



Original Scientific Paper

A contribution to the knowledge of *Amanita coryli* (Amanitaceae, Agaricales)

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ABSTRACT:

The authors present descriptions and illustrations of *Amanita coryli*, rarely featured in the mycological literature, based on molecularly characterised by nrITS sequences specimens from the Balkan Peninsula (Bulgaria and Turkey). The collections studied here suggest that the species is probably not restricted to the presumed host-trees of the genus *Corylus* and may also occur with some Fagaceae. Further, the analysis of previously released sequences in public databases show that it is a species with a wide distribution in Eurasia, probably much more common than currently known, but likely confused with other members of the section *Vaginatae*.

Keywords:

Amanita Sect. *Vaginatae*, *Amanitopsis*, biogeography, mycota of Southeastern Europe, taxonomy

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INTRODUCTION

The genus *Amanita* Pers. is the core genus of the family Amanitaceae E.-J. Gilbert and is one of the best known genera to mycophiles around the globe, encompassing both deadly poisonous and highly appreciated edible species (TULLOSS & RODRÍGUEZ-CAYCEDO 2011; CUI *et al.* 2018; LOIZIDES *et al.* 2018; ULLAH *et al.* 2019). In Europe the genus has been the focus of mycologists for a long time and its representatives have regularly been featured in iconographic treatises since the 18th century (SCHÄFFER 1774; BULLIARD 1791, 1793; PAULET 1793). Consequently, it has been extensively studied by means of the morphological approach and a vast number of dedicated works have been published, particularly since the beginning of the 20th century (see e.g. QUÉLET & BATAILLE 1902; GILBERT 1918, 1941; SEYOT 1930; VESELÝ 1933; PARROT 1960; BAS 1962a, b, 1969; MERLO & TRAVERSO 1983; GARCIN 1984; PERSSON 1992; WASSER 1992; FRAITURE 1993; BREITENBACH & KRÄNZLIN 1995; TRAVERSO 1998; GALLI 2001; CONTU 2003; KRIEGLSTEINER 2003; NEVILLE & POUMARAT 2004, 2009; RUNE 2006; VESTERHOLT 2008; KIBBY 2012). Among its sub-

divisions, the section containing ringless amanitas, i.e. the section *Vaginatae* Quél., is known as the most troublesome, containing numerous taxonomic and nomenclatural problems (FRAITURE 1993; MALYSHEVA & KOVALENKO 2015; HANSS & MOREAU 2020). Notwithstanding, in Europe the molecular approach to the study of its diversity started very recently, in the second half of the last decade (VIZZINI *et al.* 2016; LOIZIDES *et al.* 2018; HANSS & MOREAU 2020; CROUS *et al.* 2021; ŠEVČÍKOVÁ *et al.* 2021). Among such studies, the recent publication of HANSS & MOREAU (2020) provides a cornerstone in research into the section, having generated sequences of a number of type materials of European taxa. Despite being still relatively limited in number, published studies regarding the European *Vaginatae* have revealed cases of staggering variability, unexpected synonymies, and the existence of shrouded species. Furthermore, in some biodiversity hotspots, such as the Balkan Peninsula and Turkey, the genus *Amanita* has so far been tackled from the morphological approach alone and the knowledge of its diversity seems surprisingly limited judging from the existing dedicated works and checklists (ZERVAKIS *et al.* 1998; MEŠIĆ & TKALČEC 2002; SESLI & DENCHEV 2008;

DENCHEV & ASSYOV 2010; LAZAREVIĆ *et al.* 2011; LUKIĆ 2013; KARADELEV *et al.* 2018).

Among the species of the section *Vaginatae*, *Amanita coryli* Neville & Poumarat was described recently on materials from Western Europe (NEVILLE & POUMARAT 2009) and remains one of the species of the section on this continent which may be difficult to identify in the field and consequently has a largely unknown variability and distribution. The authors of this paper recently started a taxonomic study of *Amanita* in the Balkans and Turkey through a combined molecular and morphological approach (ASSYOV *et al.* 2021). Their studies revealed several specimens of *A. coryli*, providing the opportunity to make a contribution to the knowledge of the morphology and distribution of this uncommonly featured species in the mycological literature, so far unknown in the east of Europe.

MATERIALS AND METHODS

Fungal specimens. The specimens were collected during field trips and documented with colour photographs in situ and field notes as appropriate. The basidiomata were then dried in a dehydrator at 40°C for 12 h. The voucher specimens of the Bulgarian collections are deposited in the Mycological Collection of the Institute of Biodiversity and Ecosystem Research (SOMF). The specimens from Turkey are noted in the text below by the abbreviation “FBozok” and are conserved in the fungarium of Osmaniye Korkut Ata University.

Morphological study. The macroscopic characters were observed in their fresh state. The colours were matched as far as possible to the Flora of British Fungi Colour Identification Chart (ANONYMOUS 1969) and noted in parenthesis in the description with their colour code with the prefix “BFF”. The primary colour is referred to and its intensity may be additionally described as “pale” or “very pale” as appropriate. The colours for which no code number is mentioned refer to vernacular colour terms rather than chart entries. The study of the microscopic characters was performed using dried samples. The required material was extracted by freehand sections with a razor blade. The sections were then mounted to re-inflate in 5% KOH, pressed as needed and observed. Congo red in ammonia was used to improve the visibility of the microscopic structures, except for the spores, which were always observed and measured in KOH. Melzer’s reagent was used to test the possible amyloid reactions of different tissues, including the “K-K reaction” (thus termed by NEVILLE & POUMARAT 2004), i.e. the occurrence of amyloid substance in the cytoplasm of hyphae, first discovered by KOTILOVÁ-KUBIČKOVÁ (1982). An AmScope T360B microscope with an AmScope MU900 imaging device attached was used for the observations. The measurements were read from digital images with the aid of

Piximètre software v. 5.10 (©Alain Henriot & Jean-Louis Cheyep; <http://www.piximetre.fr/>). From each studied basidioma 30 randomly selected mature and normally developed spores were measured and the spore quotient (Q) was calculated for each spore. In the description, the abbreviations L_{av} , W_{av} and Q_{av} refer to the average values of the spore length, width and quotient, respectively. The designations “n =”, “m =” and “p =” stand for the number of spores, number of collections and number of basidiomata assessed. The spore data from unsequenced basidiomata are reported separately. For the remaining microscopic structures mentioned in the description, the minimum and maximum values are reported, based on 15 measurements per collection. Illustrations of the microscopic features were prepared by tracing objects from digital images onto semi-transparent paper.

DNA extraction, amplification and sequencing. The methods used for the total genomic DNA isolation, amplification and sequencing of the ITS rDNA region were those described in BOZOK *et al.* (2020). The sequences obtained in this study (forward and reverse reads) were assembled and edited as required with Sequencher version 5.4.5 (Gene Codes, Ann Arbor, MI, USA).

Phylogenetic analysis. The dataset included the sequences generated in this study, as well as 40 derived from previously published works (MALYSHEVA & KOVALENKO 2015; VIZZINI *et al.* 2016; ALONSO & RIGUEIRO 2020; HANSS & MOREAU 2020) or unpublished accessions available on the GenBank and UNITE databases. It was composed, aligned and visually checked in BioEdit Sequence Alignment Editor software (HALL 1999). A sequence of *Amanita submembranacea* (Bon) Gröger was selected as an outgroup, taking into consideration the phylogenetic inference in HANSS & MOREAU (2020). The phylogenetic analysis was performed online using the the Phylogeny.fr platform (DEREEPER *et al.* 2008; <http://www.phylogeny.fr/>). Multiple sequence alignment was carried out using MUSCLE 3.8.31 (EDGAR 2004). The phylogenetic analysis employed PhyML 3.1/3.0 (GUINDON & GASCUEL 2003; GUINDON *et al.* 2010), using the HKY85 substitution model. The Shimodaira-Hasegawa version of the approximate likelihood-ratio test (SH-aLRT) was applied for the estimation of the branch support (ANISIMOVA *et al.* 2011). The phylogenetic tree was constructed by TreeDyn 198.3 (CHEVENET *et al.* 2006) and the final technical formatting of the resulting image was applied in InkScape ver. 1.0.2-2 (<https://inkscape.org>).

RESULTS

Amanita coryli Neville & Poumarat, Fungi Non Delin-eati, 51–52: 34 (2009); Figs. 1–2.

Pileus up to 9 cm across, initially campanulate, then expanding to convex or applanate or somewhat depressed,

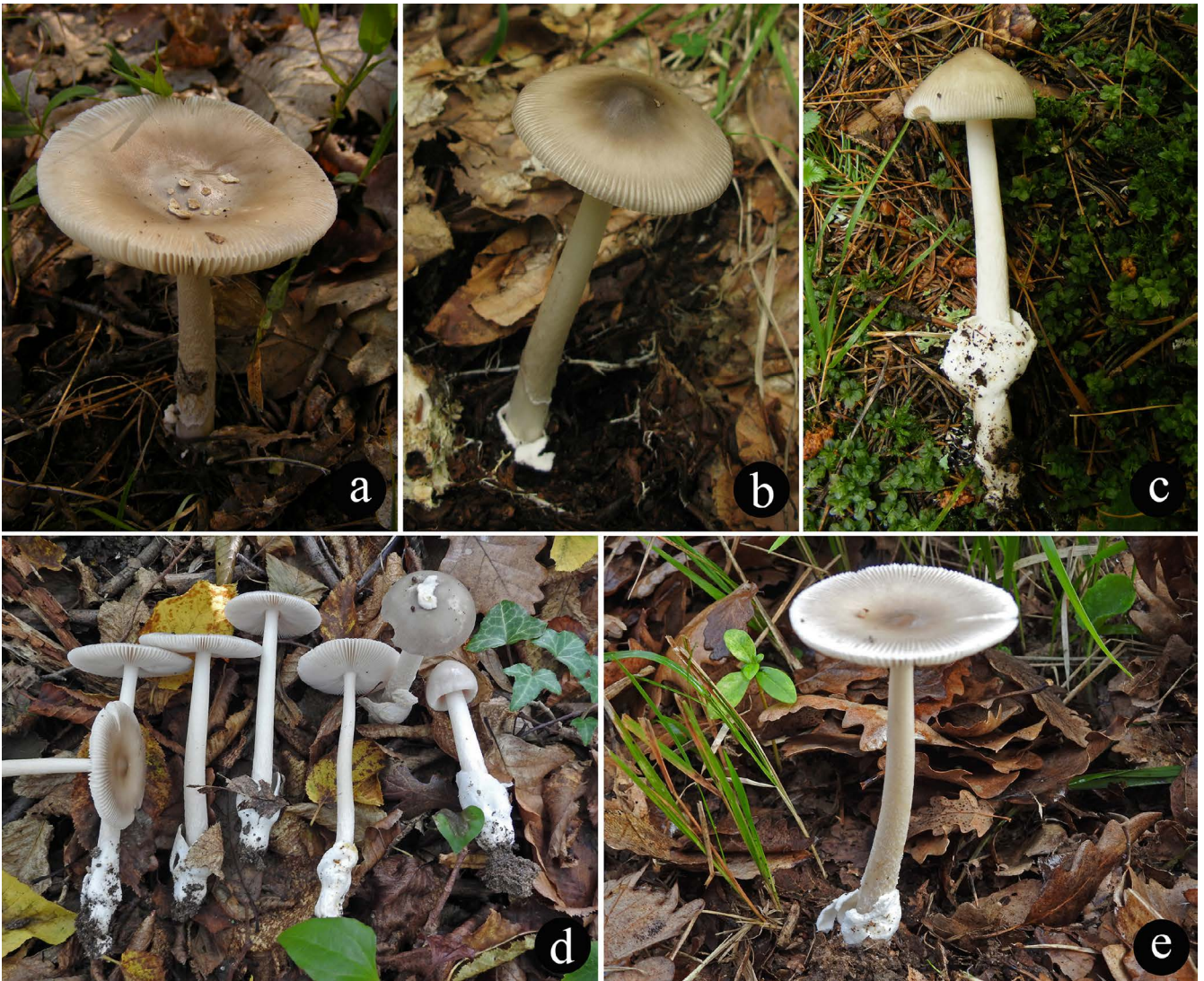


Fig. 1. Basidiomata of *Amanita coryli* from sequenced collections: a – SOMF 29444, b – SOMF 30420, c – SOMF 30416; d – FBozok 001131; e – FBozok 001160. The figures are not to scale.

usually more or less umbonate, striate to 1/5–1/3 of the pileal radius; pileal surface dry to slightly viscid in humid weather, pale vinaceous buff (BFF 31), pale clay buff (BFF 32), darker – drab (BFF 33), hazel (BFF 27) to sepia (BFF 26) in the centre, paler outwards and slightly darker at the striate marginal areas, or sometimes almost entirely off-white with a very pale cream centre or smoke grey (BFF34); velar remnants on the pileal surface sometimes present, scarce to numerous, initially whitish, then tinted yellowish buff or buff (BFF 52). Lamellae white to whitish cream, becoming salmon (BFF 45) to peach coloured (BFF 46) in exsiccata and retaining such tints for several months; lamellar edge almost smooth (not dentate), concolorous with the lamellar faces and remaining so with age; lamellulae common, truncate. Spore print not seen. Stipe up to 11 × 2 cm, up to 3.5 times longer than the pileal diameter, subcylindrical to

narrowly clavate with a non-bulbous base, finely whitish to very pale buff (BFF 52) or very pale vinaceous buff flocculose (BFF 31) or sometimes almost smooth; surface white, off-white or very pale buff (BFF 52), unchanging on handling; volva membranose, vaginate (type III), extending up to 3.5 cm from the stipe base, whitish on the outside, whitish or pale cream on the inside, yellowish buff spots sometimes present on the outer surface, unchanging with age and on handling; limbus internus not seen. Context white; odour not distinctive; taste not recorded. Macrochemical reactions: KOH 5% on the pileal context, stipe and lamellae – nil; phenol 10% on the stipe, pileal context and lamellae – purple; guayacol – on the pileal context, stipe and lamellae – nil. Basidiospores 8.8–11.9 × 8.2–11.4 μm, $Q = 1-1.31$, $L_{av} = 10.2-10.7$ μm, $W_{av} = 9.3-9.8$ μm, $Q_{av} = 1.06-1.14$ ($n = 180$, $m = 6$, $p = 6$; sequenced basidiomata); 9.8–13.3 × 9.0–12.7 μm,

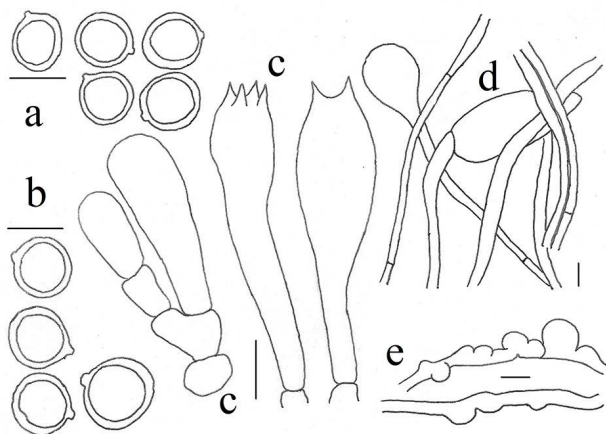


Fig. 2. Microscopic features of *Amanita coryli*: a – basidiospores from sequenced basidiomata, b – basidiospores from unsequenced basidiomata, c – basidia and basidioles with elements of subhymenium, d – elements of the external volval layer, e – thromboplerous hyphae from the stipe context. Scale bars = 10 µm.

$Q=1.00\text{--}1.20$, $L_{av} = 10.9\text{--}11.0$ µm, $W_{av} = 10.2\text{--}10.6$ µm, $Q_{av} = 1.05\text{--}1.07$ ($n = 90$, $m = 1$, $p = 3$; unsequenced basidiomata), subglobose (predominantly), globose (common) or broadly ellipsoid (rarely), thin-walled and with a large central guttule in KOH, inamyloid. Basidia generally 4-spored, but 2-spored also present and common in some basidiomata, clavate or rarely ventricose, $38.8\text{--}61.2 \times 10.8\text{--}17.2$ µm; sterigmata 2–4 µm long. Lamellar edge sterile; marginal cells abundant, mostly spherical, or more rarely ovoid, pyriform or spheropedunculate, $11\text{--}43.7 \times 10.1\text{--}37.1$ µm, thin-walled. Hymenial trama collapsed and structure not resolved; subhymenium cellular. Pileipellis of filamentose hyphae up to 5 µm broad. Universal veil on the lower stipe (volva) on the outer side composed predominantly of filamentous undifferentiated hyphae, 2–5 µm wide, scattered inflated hyphae, 10–14 µm wide, and very rare spherical or ovoid elements, 12–18 µm across; on the inner side similar, inflated elements slightly more common, spherical elements rare; thromboplerous hyphae scarce in both layers, not branched, flexuous, without swollen excrescences, occasionally septate, not constricted at the septum, with honey yellow content in KOH. Universal veil remnants on the pileus of similar architecture as the volva. Stipe context longitudinally acrophysalidic; undifferentiated hyphae 1.4–4.7 µm wide; inflated cells ovoid or narrowly clavate, $16.9\text{--}62.7 \times 6\text{--}15$ µm; thromboplerous hyphae common, 3.9–10.4 µm wide, sinuous, mostly with abundant subspherical or kidney-shaped swellings, in places septate and constricted at the septum, in KOH with uniform, honey-coloured content. Clamp connections not seen in any tissue. Microchemical reactions: K-K reaction negative.

Specimens examined: – BULGARIA, Sofia region, Sofia distr., Kostinbrod municipality, Drenovo village, N $42^{\circ}56'53.9''$, E $23^{\circ}10'56.7''$, elev. ca. 927 m, xerophytic oak forests, under *Quercus* spp., 14 June 2013, leg. B. Assyov & I. Assyova (SOMF 29444; GenBank ON115211); idem, 14 June 2013, leg. B. Assyov & I. Assyova (SOMF 30421); idem, 4 June 2022, leg. B. Assyov (SOMF 30426); Western Stara Planina Mts., Sofia distr., Gintsi municipality, Petrohan Pass, N $43^{\circ}06'57.2''$, E $23^{\circ}07'33.4''$, elev. ca. 1447 m, beech forest, under *Fagus sylvatica* L., 4 August 2018, leg. B. Assyov & I. Assyova (SOMF 30420; GenBank ON115210); Rila Mts., Sofia distr., Samokov municipality, between the locality of Gyulechitsa and the Malyovitsa ski centre, boreal coniferous forest, dominated by *Picea abies* (L.) H. Karst., under *Corylus avellana* L., 18 August 2020, leg. B. Assyov (SOMF 30416; GenBank ON115212); TURKEY, Tekirdağ province, Saray distr., Çamlıköy Nature Park, N $41^{\circ}34'24.7''$, E $28^{\circ}06'03.8''$, elev. ca. 66 m, under *Carpinus* spp. and *Quercus* spp., 5 November 2021, leg. H. Taşkın, F. Bozok & M. Yazar (FBozok 001131; GenBank ON115214); Istanbul Province, Istanbul, Belgrad Forest, N $41^{\circ}04'01.8''$, E $28^{\circ}42'30.0''$, elev. ca. 176 m, forest of *Quercus* spp. and *Fagus* spp., 7 November 2021, leg. H. Taşkın, F. Bozok & M. Yazar (FBozok 001160; GenBank ON115219); Istanbul, Atatürk Arboretum, N $41^{\circ}10'32.1''$, E $28^{\circ}58'52.2''$, elev. ca. 58 m, forest of *Quercus* spp. and *Fagus* spp., 7 November 2021, leg. H. Taşkın, F. Bozok & M. Yazar (FBozok 001171; GenBank ON115235).

DISCUSSION

Sequences were successfully obtained from six specimens from distant collecting sites in Bulgaria and Turkey. The initial BLAST search suggested they are closely related to the sequences of *A. coryli*, generated by HANSS & MOREAU (2020). When matched against the sequence MN490667 from the holotype of *A. coryli*, prior to trimming the new sequences showed between 98.93% and 99.82% similarity. The three Bulgarian sequences are not completely identical and a pairwise comparison of the ITS1-5.8S-ITS2 region shows that ON115211 differs from the other two by one substitution and two indels, resulting in 99.61% similarity. Their comparison to the sequence from the holotype results in 98.82% or 99.01% identity. The three Turkish sequences are also not totally identical either with sequence ON115235 differing from the remaining two by five indels throughout the alignment of the ITS1-5.8S-ITS2 region, resulting in 99.03% identity. Compared to the sequence from the holotype they show identity of 98.82% and 99.80% respectively. The phylogenetic analysis (Fig. 3) strongly supports the resolved six major lineages previously known in Clade 5 of section *Vaginatae* as defined by HANSS & MOREAU (2020), and treated informally as stirpes *Albogrisescens* (incl. *A. albogrisescens* and *A. alseides*), *Betulae* (incl.

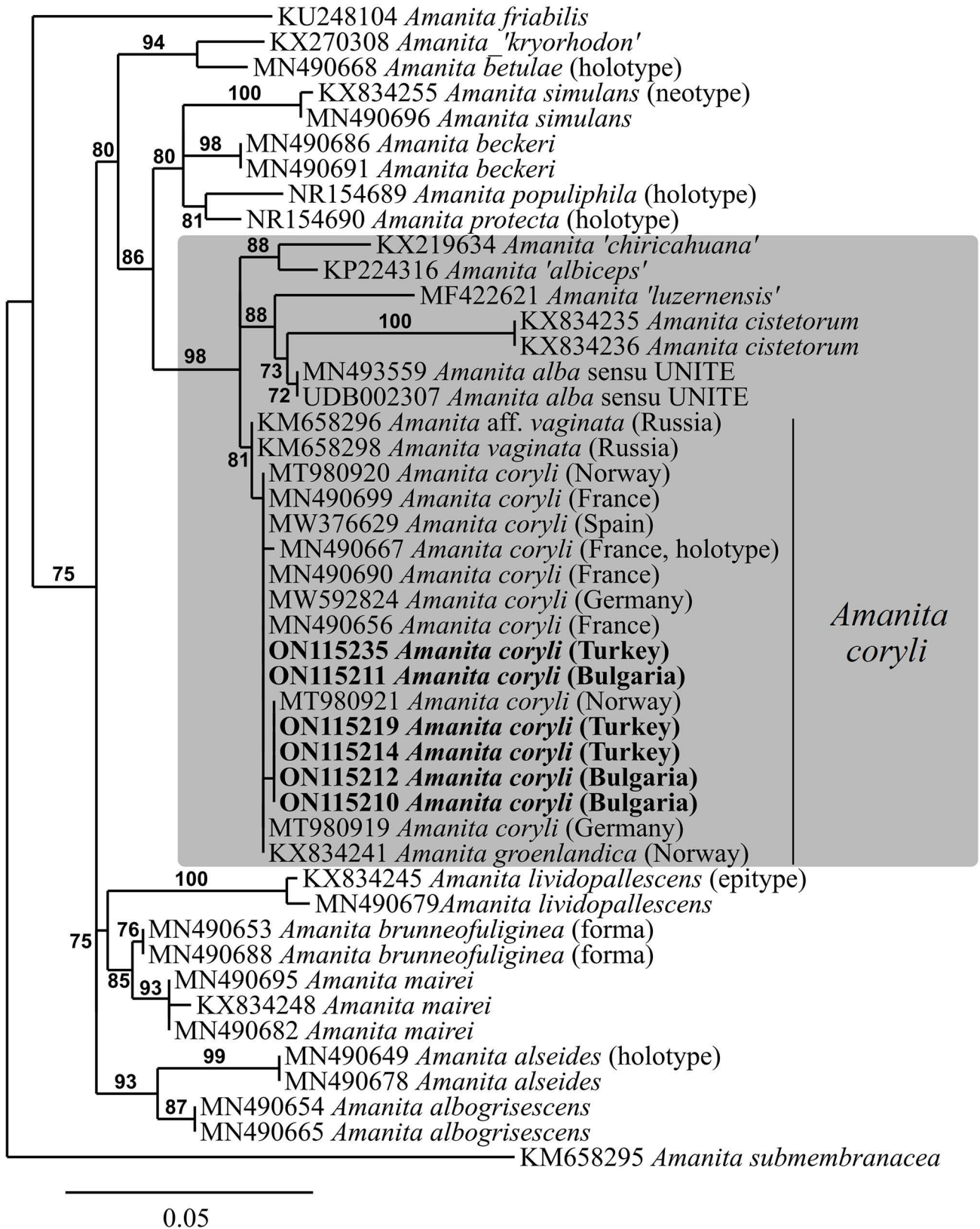


Fig. 3. Phylogenetic tree of ITS sequences of Clade 5 of *Amanita* section *Vaginatae*, obtained by analysis in PhyML. The branch support values obtained by the SH-aLRT-test are shown next to the branches as percentages; only those values above 50% are annotated. The sequences generated in this work are shown in boldface. The clade corresponding to stirps *Coryli* is highlighted in grey.

A. betulae and *A. 'kryorhodon'*), *Coryli* (see below), *Lividopallescens* (incl. *A. lividopallescens*), *Mairei* (incl. *A. brunneofulginea* f. *ochraceopallida* and *A. mairei*) and *Populiphila* (incl. *A. beckeri*, *A. populiphila*, *A. protecta* and *A. simulans*). *Amanita friabilis* (P. Karst.) Bas appears as a separate clade and has not yet been assigned to particular stirps (cf. HANSS & MOREAU 2020). Stirps *Coryli* in particular receives high branch support (98%) and encompasses sequences of *A. alba* sensu UNITE, *A. 'albiceps'* Lamoureux (nom. illeg., nom. prov.), *A. 'chiricahuana'* Tulloss (nom. illeg., nom. prov.), *A. cistetorum* Contu & Pacioni, *A. coryli* and *A. 'luzernensis'* Tulloss & coll. (nom. illeg., nom. prov.), which is also in line with the findings of HANSS & MOREAU (2020). Our six sequences nest in a clade, containing the sequence from the holotype of *A. coryli*, along with 12 other sequences, accessioned as *A. coryli*, *A. groenlandica*, *A. aff. vaginata* and *A. vaginata*. This clade is well supported (81%). The sequences KM658296 and KM658298 from the Asian part of Russia are somewhat divergent from the main clade, differing from the holotype sequence by two or three base pairs, while the remaining terminal sister branches in the stirps *Coryli* differ from each other by 11 to 59 base pairs. The difference observed in the Russian sequences is thus likely not significant and receives no support in the analysis, although it may be interesting to examine more molecularly characterised collections of *A. coryli* from Asia in future. The sequences from two of the Turkish and two of the Bulgarian specimens (ON115210, ON115212, ON115214, and ON115219), together with MT980921 from Norway, are also slightly divergent and form a short branch of their own, which is nested deeply in the *coryli*-clade and receives no statistical support. The assessment of the available sequences of *A. coryli* reveals the existence of a certain degree of polymorphism in the ITS region. Since no consistent pattern could be found suggesting the existence of discrete groups, and no geographic or host correlation seems to exist, we feel it is justified to consider *A. coryli* as a single entity for the time being.

Amanita coryli is characterised by a more or less umbonate pileus, which is generally buff-coloured, usually somewhat zonate with a darker centre and paler outer parts, but slightly darker in the zone of the marginal striations. The volva and the volval remnants on the pileus in particular tend to become yellowish ochraceous spotted with time. The spores are subglobose to globose and the volval tissues are mainly composed of filamentous hyphae with relatively few inflated elements in the different volval layers. In terms of the structure of the volva it should be taken into account that in our specimens its plectenchyma appeared rather collapsed and further observations on fresh specimens seem worthy. The same stands for the peculiarities of the hymenial trama.

NEVILLE & POUMARAT (2009) reported the spore quotient of *A. coryli* as $Q = 0.94\text{--}1.26(1.29)$, $Q_{av} = 1.00\text{--}1.12$

and for f. *albida* Neville, Poumarat & M. Rovira – $Q = (0.95)\text{--}1.00$, $Q_{av} = 1.00$; in the data for both taxa there seem to be some inaccuracies in the calculations of the quotient minimum values. Spore measurements derived from molecularly supported specimens are reported here for the first time. In our specimens the minimum value for the average Q appears to be slightly higher than the values obtained by NEVILLE & POUMARAT (2009), but this is not unexpected as in their calculation they included values below 1. Overall, the remaining spore parameters in the specimens studied here are within the limits given in the protologue, although some variability exists and the spore dimensions vary even between basidiomata from the same locality. As in other recently described and little-known species, more data from barcoded specimens would be welcome in order to gain a better understanding of the spore variability. In terms of the remaining macroscopic and microscopic characters in the collections presented here, they also correspond well to the original description and the accompanying illustrations. Notably, one of the Turkish collections shows that albeit rarely, *A. coryli* may sometimes produce basidiomata with uniformly grey-coloured pilei at least when young (Fig. 1d, the two basidiomata on the right). Similar striking chromatic variability has already been documented in some other members of the section *Vaginatae* (HANSS & MOREAU 2020; ASSYOV *et al.* 2021). NEVILLE & POUMARAT (2009) distinguished two forms – f. *coryli* and f. *albida*, the latter being set apart by the entirely whitish pileus. Such a white form is also illustrated by Tulloss (<http://www.amanitaceae.org/?Amanita+coryli>). Curiously, the collection from the Rila Mts. studied here is somewhat intermediate between the two above, featuring a whitish pileus with a darker, pale cream umbo. We are hesitant to assign this specimen to f. *albida* and feel that a consensus on the taxonomic treatment of the distinctive colour forms in *Amanita* is needed.

NEVILLE & POUMARAT (2009) reported a positive K-K reaction for *A. coryli*. In our specimens we could not confirm its presence. To what extent this is a stable and taxonomically valuable character at species level in section *Vaginatae* is at present unknown and the matter is certainly worthy of further study on molecularly barcoded specimens.

On drying the pilei in *A. coryli* turn pale cinnamon (BFF 10), pale sienna (BFF 11) or pale fulvous (BFF 12) and the stipe becomes mottled with the same colours or somewhat buff (BFF 51). The lamellae develop a striking salmon (BFF 45) to peach tint (BFF 46), which is retained for several months, a trend which we have observed in several other *Vaginatae* (Assyov *et al.* 2021). For the moment it is unknown whether the colours in the exsiccata could be of some use for species identification, but they are mentioned here for the sake of completeness.

Amanita betulae Neville & Poumarat, a species as far as currently known associated only with *Betula*

spp., seems to be morphologically close. While its microscopic features considerably overlap with those in *A. coryli*, it seems to be distinguished by the more brightly coloured pilei (orange ochraceous to tawny and becoming more greyish in age), the stipe often almost concolorous with the pileus zebroid bands, and gill edges, which quickly become dark-coloured; when present the velar remnants on the pileus are said to remain whitish (NEVILLE & POUMARAT 2009; KIBBY 2011, 2012; FERNÁNDEZ-VICENTE 2021). When discussing the difference between the two species, NEVILLE & POUMARAT (2009) mentioned the more slender stipe in *A. betulae* as a distinguishing characteristic, but as evident from the illustrations they present, this is a feature which varies considerably even among different basidiomata from the same collection. Nonetheless, *A. betulae* and *A. coryli* were already shown unquestionably to belong to distinct lineages by HANSS & MOREAU (2020) as they also appear in the analysis included here.

TRAVERSO (1998) described *A. vaginata* f. *avellanea* M. Traverso, associated with *Castanea sativa* Mill., which judging from the original description and the illustration resembles *A. coryli*. This taxon caused confusion for a long time and it was suggested by M. Contu in a personal communication to G. Kibby, that it may be identical to *A. coryli* (KIBBY 2012). NEVILLE & POUMARAT (2009) studied the holotype and an additional collection from France. They believed it to be distinct and elevated it to species rank, although they did not discuss its delimitation from *A. coryli* to which Traverso's taxon is apparently close in terms of its gross morphology. An analysis of the reported microscopic characters hardly provides further clues as most of the important features overlap in the two taxa. Besides, similarly to *A. coryli*, they reported the Q of Traverso's specimen to be 0.92–1.05 and Qm = 0.98, values which are apparently partly incorrect, but nevertheless with the maximum value resting within the known range of the spore variability of *A. coryli*. Eventually, HANSS & MOREAU (2020) obtained the ITS sequence from the holotype of *A. vaginata* f. *avellanea* and showed that this name should be considered a synonym of *A. fulvoides* Neville & Poumarat. The case of Traverso's collection suggests that in the field some colour forms of *A. fulvoides* may turn out to be difficult to distinguish from *A. coryli*. In addition to the rusty spotted volva in *A. fulvoides*, the dark edged lamellae in this species could also aid the preliminary identification should such forms be further encountered.

In our phylogenetic inference the only publicly available sequence of *A. groenlandica* Bas ex Knudsen & Borgen clusters in the clade of *A. coryli*, similarly to the analysis in HANSS & MOREAU (2020). This species was described in association with dwarf members of the genera *Salix* L. and *Betula* L. from southern Greenland (KNUDSEN & BORGEN 1987). Judging from the placement of this non-type sequence, *A. groenlandica* may be phy-

logenetically close to *A. coryli*, but as inferred from its original and some later descriptions it could be separated by several morphological characters, including a greyish stipe surface and girdles, an at least partly greyish volva, dark-coloured lamellar edges and numerous spherical elements in the outer volval layer – characters suggesting phenetic similarity to the group around *A. submembranacea* (KNUDSEN & BORGEN 1987; RUNE 2006). Further attention to this poorly known species is necessary to provide additional support for either its separation or inclusion in *A. coryli*, preferably also including attempts to obtain sequences from type materials.

Among the remaining European species of the section, *Amanita cistetorum* was resolved as phylogenetically close to *A. coryli* by HANSS & MOREAU (2020) and appears similarly placed in our ITS inference. It is currently known as a Mediterranean species, described as a *Cistus*-associate on materials from Sardinia (CONTU & PACIONI 1998). It shows a certain similarity to *A. coryli*, particularly the tendency of the velar remnants on the pileus and volva to develop an ochraceous tint, as well as the scarce inflated elements in this tissue (CONTU & PACIONI 1998). It should, however, be distinguished easily from the species discussed here due to its squat stature, grey-coloured pilei and broader, subglobose to broadly ellipsoid basidiospores, as well as its peculiar habitat (CONTU & PACIONI 1998).

Amanita coryli was originally described and recorded as occurring exclusively with *Corylus* L. (NEVILLE & POUMARAT 2009; KIBBY 2012). Our findings, however, suggest that it may not be strictly related to this host since three of our Bulgarian collections were found in habitats where this shrub was not present, while only the third one appeared to be associated with *Corylus*. As this fact seemed puzzling, one of our team members (BA) re-visited the respective localities in 2020 and the lack of hazel or other species of *Betulaceae* Gray in the vicinity of the *A. coryli* collections was confirmed, although *C. avellana* and *Carpinus* spp. are generally present in both areas (but far from the collecting sites). Among the Turkish findings two collections occurred in forests of *Quercus* spp. and *Fagus* spp. and one in a mixed forest of *Carpinus* spp. and *Quercus* spp. It is hence safe to assume that species of *Quercus* L. and *Fagus* L. may possibly serve as the hosts of this particular species at least in some cases. A certain degree of host versatility has been previously documented in other species of the section *Vaginatae* (VIZZINI *et al.* 2016). The species apparently has a wide altitudinal range, occurring from lower elevations in the belts of thermophilous oak woodlands up to boreal coniferous forest belts, provided suitable mycorrhizal hosts are present.

Amanita coryli was based on numerous materials from France and Spain (NEVILLE & POUMARAT 2009). Later on, a few additional records were mentioned from Italy, Spain and the United Kingdom, including a bar-

coded Spanish collection (KIBBY 2012; RIBES *et al.* 2017; ANONYMOUS 2019; ALONSO & RIGUEIRO 2020). Some unpublished sequences from Germany and Norway are publicly available in GenBank, released by R.E. Tulloss and collaborators (MT980919, MT980920, MT980921, MW592824; see <http://www.amanitaceae.org/?Amanita+coryli>). In this paper we present the first molecularly supported collections from the Balkan Peninsula, considerably extending the geographic range of the species in the south-eastern direction. In addition, as mentioned above, two sequences designated as *A. aff. vaginata* or *A. vaginata* in MALYSHEVA & KOVALENKO (2015) also appear to belong to *A. coryli*, differing from the sequence from the holotype in terms of a small number of base pairs, a difference which we deem not to be significant, taking into consideration the overall polymorphism observed in the ITS region in the *A. coryli* clade. They originate from the Baikal Region and Primorskiy Krai in Russia and greatly extend the range of the species throughout Asia, although in fact some species of the section *Vaginatae* have already been shown to have a wide Eurasian distribution (MALYSHEVA & KOVALENKO 2015; HANSS & MOREAU 2020). It is our opinion that those findings, coupled with the wide range of the potential hosts of *A. coryli*, unequivocally evince that it must be widespread, if not common, in Eurasia, but has most probably been overlooked or unrecognised so far. The small number of published findings thus merely reflect the difficulties of identifying the ringless amanitas in Europe.

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REZIME

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Prilog poznavanju *Amanita coryli* (Amanitaceae, Agaricales)

Fuat BOZOK, Boris ASSYOV, Mahmut YARAR i Hatıra TAŞKIN

Autori predstavljaju opis i ilustracije vrste *Amanita coryli*, retko zastupljene u mikološkoj literaturi, zasnovane na molekularno okarakterisanim nrITS sekvencama primeraka sa Balkanskog poluostrva (Bugarska i Turska). Istraživane kolekcije sugerišu da vrsta verovatno nije ograničena samo na domaćine iz roda *Corylus* i da se očigledno može pojaviti i kod nekih predstavnika familije Fagaceae. Dalje, analiza prethodno objavljenih sekvenci u javno dostupnim bazama podataka pokazuje da je to vrsta sa širokim rasprostranjenjem u Evroaziji. Vrsta je verovatno mnogo češća nego što je trenutno poznato, ali je verovatno zamenjivana sa drugim članovima sekcije *Vaginatae*.

Ključne reči: *Amanita* Sect. *Vaginatae*, *Amanitopsis*, biogeografija, mikota jugoistočne Evrope, taksonomija