

CONTRIBUTION TO THE TAXONOMIC KNOWLEDGE OF THE FAMILY INOCYBACEAE IN BUDAPEST, HUNGARY

CSIZMÁR, M.^{1*} – CSEH, P.¹ – DIMA, B.² – ASSAMERE, A.^{1,3} – ORLÓCI, L.⁴ – BRATEK, Z.¹

¹*Department of Plant Physiology and Molecular Plant Biology, Eötvös Loránd University, Pázmány Péter sétány 1/C, H-1117 Budapest, Hungary*

²*Department of Plant Anatomy, Eötvös Loránd University, Pázmány Péter sétány 1/C, H-1117 Budapest, Hungary*

³*Department of Biology, Natural and Computational Sciences, Mekdela Amba University, South Wollo, P. O. Box 32, Tuluawulia, Ethiopia*

⁴*Botanical Garden, Eötvös Loránd University, Illés utca 25, H-1083 Budapest, Hungary*

**Corresponding author*

e-mail: csimiz@gmail.com; phone: +36-20-241-4475

(Received 23rd Aug 2022; accepted 11th Nov 2022)

Abstract. Members of the family Inocybaceae (Agaricales, Basidiomycota), one of the most diverse ectomycorrhiza-forming macrofungi of urban landscapes were studied. We performed phylogenetic analysis of 28 specimens (23 *Inocybe* spp., 3 *Mallocybe* spp., 2 *Pseudosperma* spp.) collected in anthropogenically influenced habitats of Budapest, Hungary. The internal transcribed spacer region of the nuclear ribosomal RNA cistron (nrDNA ITS) was sequenced for barcoding purposes of our Inocybaceae specimens. Identification to a presumptive species level was successful in the case of 24 species although one interesting *Inocybe* sp. and three *Mallocybe* spp. could not be determined. Five species, *Inocybe ghibliana*, *I. pararubens*, *I. psammobrunnea* (syn. *I. griseotarda*), *I. obscuroides* (syn. *I. cincinnata* var. *major*) and *Pseudosperma permelliolens* were collected for the first time in Hungary.

Keywords: *urban environment, molecular taxonomy, Inocybe, Mallocybe, Pseudosperma*

Introduction

Inocybaceae is an ectomycorrhizal-forming and taxonomically highly diverse family of Agaricales. In the last decade, mostly in Europe and North America, a number of phylogenetic studies on Inocybaceae have been conducted. The new, phylogeny-based classification also resulted in the discovery of an enormous amount of new species of this highly diverse family (e.g. Bandini et al., 2018; Matheny et al., 2019). Between 2020 and 2021, at least 59 species were identified as new in Europe (Bandini and Oertel, 2020; Bandini et al., 2020a, b, c, 2021a, b, c; Cervini et al., 2020; Cervini, 2021; Dovana et al., 2021; Mesic et al., 2021). Based on six-gene molecular phylogenetic studies, Inocybaceae have recently been adopted as an independent family, and simultaneously separated into seven major clades, all of them recognized in generic rank: *Auritella*, *Inocybe* sensu stricto, *Inosperma*, *Mallocybe*, *Nothocybe*, *Pseudosperma*, and *Tubariomyces* (Matheny, 2005; Matheny et al., 2019; Ryberg et al., 2010). The estimated number of the species worldwide in the largest genera of the family, *Inocybe* sensu stricto, is ca. 1050 species according to Matheny and Kudzma (2019), but this number will probably rise (Daskalopoulos et al., 2021). Only in Europe, at least 470 species are known so far (Bandini et al., 2021a). *Pseudosperma* is a smaller genus, representing around 70 species worldwide according to Matheny et al. (2019), of

which more than 40 species occur in the European countries (Bandini and Oertel, 2020). The genus *Mallocybe* contains more than 55 species (Matheny et al., 2019; Daskalopoulos et al., 2021), while *Inosperma* is represented by more than 70 known species with at least 13 species in Europe (Matheny et al., 2019; <https://www.inocybe.org>). *Auritella*, *Nothocybe* and *Tubariomyces* are rather small, mostly extra-European genera (Matheny et al., 2019).

The process of urbanization is well-known nowadays; global annual urban expansion is all over 5%, much higher than the growth of the urban world population rates (Güneralp et al., 2020). Most of the world population concentrates in cities and their urban agglomeration environments. This fact necessitates more relevant urban research and biodiversity protection (Angel et al., 2005; Oke et al., 2021). Usually, the most significant anthropogenic activities affect the cities and their agglomeration though at the same time the substantial cities, like Budapest and other capitals built typically in unique natural environments.

Members of the Inocybaceae are considered as cosmopolitan fungi and are able to form ectomycorrhizal associations with plants of at least 23 families (Matheny et al., 2019). Major part of the Inocybaceae prefer more or less calcareous or neutral rich soil and growing near the rim of paths or roads, in cemeteries, or in parks and other typical urban habitats (Kuyper, 1986; Csizmár et al., 2021). Many of the newly and also formerly described species were collected in urban territories, like gardens, parks, side of roads or in botanical gardens (Kuyper, 1986; Matheny et al., 2019; Bandini et al., 2018, 2021a). Therefore, the aim of the present study is to improve the available knowledge of European species of Inocybaceae with special focus on Budapest, the capital of Hungary. We also tried to give a taxonomic overview of the collected specimens by phylogenetic analysis. Surveys of urban biodiversity and the better understanding of urban species composition could act as a springboard for conservationists to develop relevant and effective biodiversity conservation programs.

Materials and methods

Sampling

The basidiomata samples (23 *Inocybe* spp., 3 *Mallocybe* spp., 2 *Pseudosperma* spp.) were registered in different districts (VIII., X., XI., XII., XIII., XIV., XXII.) of Budapest, Hungary between 2017 and 2021. Basidiomata were collected by our research group in Department of Plant Physiology and Molecular Plant Biology, Eötvös Loránd University (see Acknowledgements). All collections were found in anthropogenically influenced habitats, such as tree avenues, gardens, parks and botanical garden in association with different broadleaved trees and conifers. Typical soil of the collection sites is calcareous sand, deposited mainly by the Danube and occasionally mixed with clay. However, in these areas, the soil is frequently disturbed and the original soil layers could be mixed or contaminated. More collections were photographed and described in fresh state. Spores and other microscopic features were examined with a Nikon Optiphot-2 research light microscope in 5% KOH or in Aqueous Congo red dye (which was using also in microphotography) and with a Nikon SMZ-U stereo microscope. The length/width ratio (Q value) of the spores and sometimes of the cystidia were also calculated. Dried tissue samples were placed in the fungarium of the research group at the Department of Plant Physiology and Molecular Plant Biology, Eötvös Loránd University. The nomenclature and taxonomy applied here follow the Index Fungorum (<http://www.indexfungorum.org>).

DNA extraction, PCR and sequencing

Dried or fresh basidiomata were sampled and genomic DNA was isolated using the DNeasy Plant Mini Kit (Qiagen, Courtaboeuf, France) following the protocols. During the amplification process of the ITS region ITS1F/ITS4 primers were used (White et al., 1990; Vilgalys and Hester, 1990; Gardes and Bruns, 1993). Amplification products were inspected through gel electrophoresis on 1% agarose gel. For staining ethidium-bromide was used. PCR products were purified with the QIAquick® PCR Purification Kit following the protocols. Sanger sequencing was done by BIOMI Ltd. (Gödöllő, Hungary) and BaseClear (Leiden, Netherlands).

Phylogenetic analyses

We generated 28 new sequences which were submitted to GenBank (*Table 1*) with accession numbers OM228846-OM228873. To check the electropherograms, FinchTV V. 1.4.0 (Geospiza, Inc., Seattle, WA, USA; <http://www.geospiza.com>) was used. Sequences were assembled and edited using MEGA-X v. 10.0.5 (Kumar et al., 2018) while software MAFFT (Katoh and Toh, 2008) was used to align the sequences. The newly generated sequences were subjected to BLAST search tool of GenBank at NCBI (National Center for Biotechnology Information) (Zhang et al., 2000) and UNITE (Altschul et al., 1997; Nilsson et al., 2018) public databases. Thereafter, taxonomically most relevant sequences were gathered for further evaluation regarding the available ecological and distributional data. The final dataset includes our newly generated sequences and the selected reference sequences. Selection of the final matching sequences was based on the current taxonomic results. RAxML 8.2.12 (Stamatakis, 2014) as implemented on CIPRES (Miller et al., 2010) were carried out to perform the Maximum Likelihood (ML) analyses, using fast bootstrap (1000×). Likelihood of final tree was optimized under GAMMA substitution model and FigTree V. 1.4.4 (Rambaut, 2010) was used to visualize the phylogenetic tree (*Fig. 1*). As outgroup *Crepidotus applanatus* (KR673659) and *Crepidotus mollis* (JF907959) were chosen.

Results

We could successfully sequence the ITS region of 28 specimens which were all collected in Budapest, urban environment (*Table 1*). All of the specimens could be assigned to a presumptive species level. 23 registered specimens belong to the genus *Inocybe*, three specimens to the genus *Mallocybe* and two specimens to the genus *Pseudosperma*.

Totally 18 different species of the family Inocybaceae were registered. Phylogenetic tree was inferred to consider phylogenetic relationship in case of this taxonomically diverse and difficult family (*Fig. 1*).

Four *Inocybe* species, *I. ghibliana* Bandini & B. Oertel, *I. pararubens* Carteret & Reumaux, *I. psammobrunnea* Bon (syn. *I. griseotarda*) and *I. obscuroides* P.D. Orton (syn. *I. cincinnata* var. *major*) and one *Pseudosperma*, namely *Pseudosperma permelliolens* (Carteret & Reumaux) Matheny & Esteve-Rav. were not previously collected in Hungary. *Mallocybe* aff. *heimii* (M117) and *M. aff. plebeia* (M118) are tentatively labelled clades, because their phylogenetic position seems to be unclear. Based on our analysis of the ITS locus, both sequences differ from the clades of *M. heimii* s. str. and *M. plebeia* s. str (formerly identified as *M. latispora* clade) (*Fig. 1*).

Table 1. Sequenced specimens of the present study. Collections are given with their specimen voucher, location (Roman numerals indicate the proper district of Budapest), habitat type, the date of collection, ectomycorrhizal host plants within the range of 15 m (suspected mycorrhizal partners were underlined) and GenBank accession numbers

Specimen voucher	Taxon	Location	Habitat type	Date of collection	Hosts	GenBank accession (ITS)
M48	<i>Inocybe aeruginascens</i> Babos	XIII. Budapest	Park	4 June 2019	<i>Pinus nigra</i>	OM228846
M114	<i>Inocybe alluvionis</i> Stangl & J. Veselský	XIV. Budapest	Garden	27 September 2020	<u><i>Betula pendula</i></u> , <i>Salix alba</i>	OM228847
M122	<i>Inocybe alluvionis</i> Stangl & J. Veselský	XI. Budapest	Park	23 October 2020	<i>Carpinus betulus</i>	OM228848
M4	<i>Inocybe alluvionis</i> Stangl & J. Veselský	XI. Budapest	Park	31 October 2017	<i>Populus</i> sp.	OM228849
M34	<i>Inocybe furfurea</i> Kühner	XI. Budapest	Park	20 September 2019	<i>Carpinus betulus</i>	OM228850
M128	<i>Inocybe ghibliana</i> Bandini & B. Oertel	XI. Budapest	Park	26 October 2020	<i>Carpinus betulus</i>	OM228851
M5	<i>Inocybe ghibliana</i> Bandini & B. Oertel	XI. Budapest	Park	29 September 2017	<i>Carpinus betulus</i>	OM228852
M9	<i>Inocybe ghibliana</i> Bandini & B. Oertel	XIV. Budapest	Garden	17 September 2017	<i>Picea pungens</i>	OM228853
M137	<i>Inocybe psammobrunnea</i> Bon	XXII. Budapest	Garden	23 November 2020	<i>Picea abies</i>	OM228854
M123	<i>Inocybe griseovelata</i> Kühner	XI. Budapest	Park	23 October 2020	<i>Carpinus betulus</i>	OM228855
M140	<i>Inocybe</i> aff. <i>inodora</i> Velen.	XI. Budapest	Park	1 June 2021	<u><i>Tilia tomentosa</i></u> , <i>Carpinus betulus</i>	OM228856
M119	<i>Inocybe obscuroides</i> P.D. Orton	XIV. Budapest	Garden	18 October 2020	<i>Betula pendula</i>	OM228857
M135	<i>Inocybe obscuroides</i> P.D. Orton	VIII. Budapest	Botanical Garden	28 October 2020	<i>Picea pungens</i>	OM228858
M132	<i>Inocybe pararubens</i> Carteret & Reumaux	XII. Budapest	Garden	12 October 2017	<i>Quercus</i> sp.	OM228859
M2	<i>Inocybe phaeoleuca</i> Kühner	XI. Budapest	Park	29 September 2017	<i>Carpinus betulus</i>	OM228860
M125	<i>Inocybe phaeoleuca</i> Kühner	XI. Budapest	Park	23 October 2020	<i>Carpinus betulus</i>	OM228861
M127	<i>Inocybe phaeoleuca</i> Kühner	XI. Budapest	Park	22 October 2020	<i>Betula pendula</i>	OM228862
M133	<i>Inocybe pusio</i> P. Karst.	VIII. Budapest	Botanical Garden	28 October 2020	<i>Quercus robur</i> , <i>Carpinus betulus</i>	OM228863
M46	<i>Inocybe semifulva</i> Grund & D.E. Stuntz	XIII. Budapest	Park	4 June 2019	<i>Tilia</i> sp., <i>Pinus</i> sp.	OM228864
INOCBP1	<i>Inocybe semifulva</i> Grund & D.E. Stuntz	XI. Budapest	Park	8 September 2021	<i>Tilia tomentosa</i> , <i>Carpinus betulus</i>	OM228865
M14	<i>Inocybe semifulva</i> Grund & D.E. Stuntz	XIV. Budapest	Garden	23 September 2017	<i>Salix alba</i>	OM228866
M141	<i>Inocybe semifulva</i> Grund & D.E. Stuntz	XI. Budapest	Park	13 June 2021	<u><i>Tilia tomentosa</i></u> , <i>Carpinus betulus</i>	OM228867
M129	<i>Inocybe grammopodia</i> Malençon	VIII. Budapest	Botanical Garden	28 October 2020	<i>Quercus robur</i> , <i>Carpinus betulus</i>	OM228868
M117	<i>Mallocybe</i> aff. <i>heimii</i> (Bon) Matheny & Esteve-Rav.	XI. Budapest	Tree avenue; Roadside	20 October 2020	<i>Tilia tomentosa</i>	OM228869
M118	<i>Mallocybe</i> aff. <i>plebeia</i> Bandini, B. Oertel & U. Eberh.	XIV. Budapest	Garden	18 October 2020	<i>Betula pendula</i> , <i>Picea</i> sp.	OM228870
M120	<i>Mallocybe</i> aff. <i>malenconii</i> (R. Heim) Matheny & Esteve-Rav.	XI. Budapest	Tree avenue; Roadside	14 October 2020	<i>Tilia tomentosa</i>	OM228871
M134	<i>Pseudosperma obsoletum</i> (Romagn.) Matheny & Esteve-Rav.	X. Budapest	Park	24 June 2020	<u><i>Populus</i> sp.</u> , <i>Pinus</i> sp., <i>Picea</i> sp.	OM228872
M130	<i>Pseudosperma permelliolens</i> (Carteret & Reumaux) Matheny & Esteve-Rav.	VIII. Budapest	Botanical Garden	28 October 2020	probably <i>Quercus robur</i> , <i>Carpinus betulus</i>	OM228873

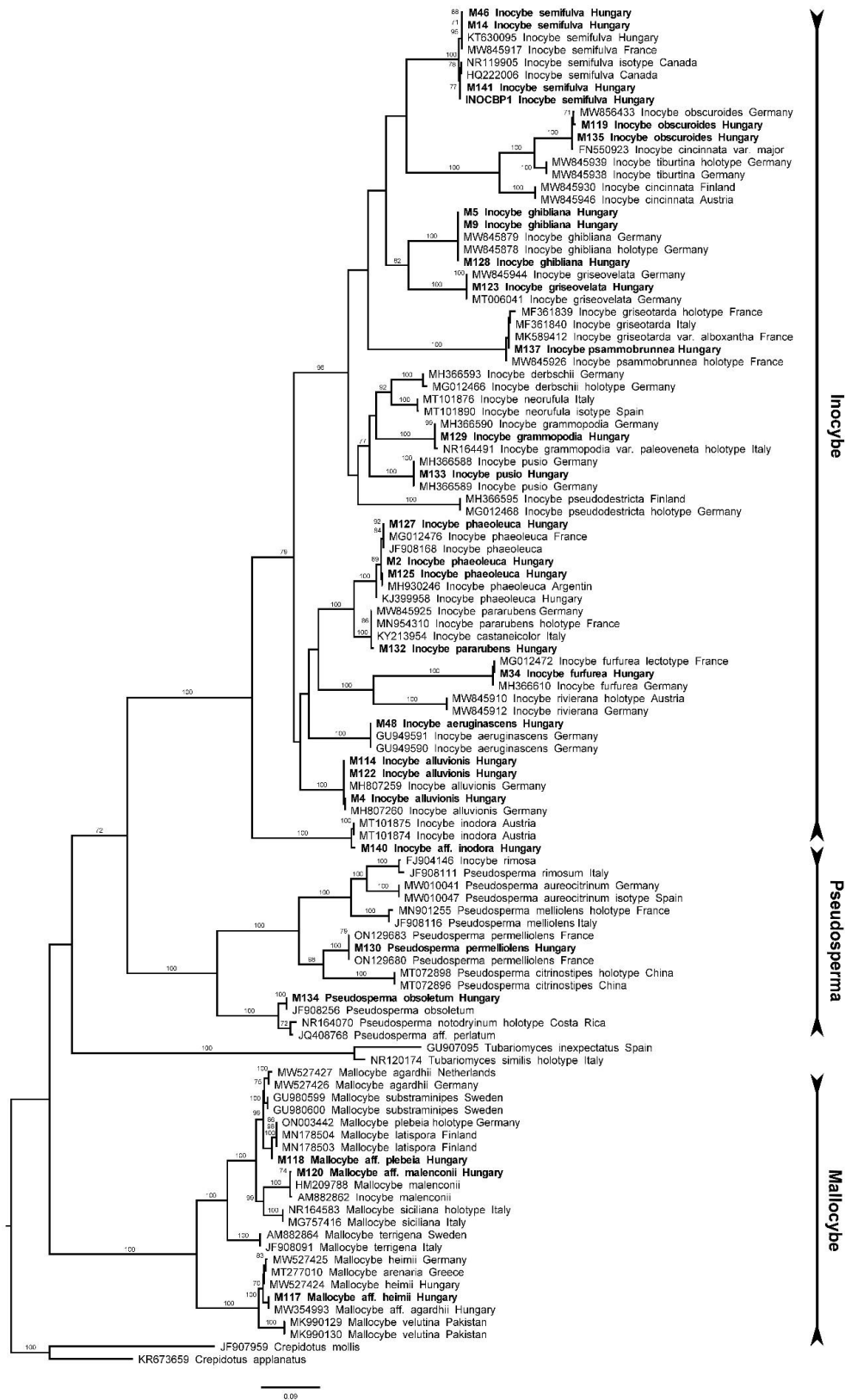


Figure 1. ML phylogenetic tree of family Inocybaceae based on analysis of the nrDNA internal transcribed spacer (ITS) sequences. *Crepidotus applanatus* (KR673659) and *Crepidotus mollis* (JF907959) were chosen as outgroup. Newly generated sequences are in bold. Maximum Likelihood (ML > 70%) bootstrap values are indicated at the branches

Photographs of these two interesting collections are shown in *Figures 2* and *3*. We also demonstrate in this paper a new collection of *Mallocybe* aff. *malenconii* (M120) with photograph (*Fig. 4*) which might be a new species as we introduced already in our previous work (Csizmár et al., 2021). Another interesting collection, unfortunately without good quality photograph, is *Inocybe* aff. *inodora* (M140) which shows a close genetic relation to *Inocybe inodora* s. str. reported from Austria.



Figure 2. Basidiomata of *Mallocybe* aff. *heimii* (M117); 20 October 2020, district XI., Budapest, Hungary. Scale bar 1 mm. Photo: M. Csizmár



Figure 3. Basidiomata of *Mallocybe* aff. *plebeia* (M118); 18 October 2020, district XIV., Budapest, Hungary. Scale bar 1 mm. Photo: M. Csizmár



Figure 4. Basidiomata of *Mallocybe* aff. *malenconii* (M120); 14 October 2020, district XI., Budapest, Hungary. Scale bar 1 mm. Photo: M. Csizmár

Discussion

Several specimens from our collections like *Inocybe aeruginascens*, *I. furfurea*, *I. griseovelata*, *I. pusio* sensu lato, *I. semifulva* reported as more common species in Europe, generally from dry, sandy soils, frequently growing on urban environments with diverse broadleaved trees but also with conifers (Nagy and Nagy, 2011; Bandini et al., 2018; Csizmár et al., 2021). *Inocybe phaeoleuca* sensu lato, a certainly common species was collected three times during our study. As a consequence of missing ITS sequence data of the type specimen, we could not label our “phaeoleuca” specimens (M2, M125, M127) based on the many available matching sequences in GenBank and UNITE public databases. Judging of our ITS data, the *I. phaeoleuca* complex contains two clades which implies that the taxonomic position of the group is probably unresolved. Other collected common species, *Inocybe alluvionis* known as growing riverine, sandy soils but prefers moister, shaded habitats (Bandini et al., 2019). Some collected species were less known probably by reasons of unclear taxonomic position and not suitable identification keys. *Inocybe pararubens*, *I. obscuroides*, *I. psammobrunnea* and *I. ghibliana* which were reported here as new to Hungary seem like relatively common species regarding to Europe. *Inocybe pararubens* were described from France by Carteret and Reumaux (2012) under *Fagus sylvatica* (holotype sequence: GenBank MN954310). Later the same species were identified as *I. castaneicolor* by La Rosa et al. (2017) from Italy on sandy soil under *Quercus suber*. Furthermore, *I. pararubens* were reported from the alpine zone of Sweden associated with *Salix* sp. on calcareous wet ground (Vauras and Larsson, 2020). *Inocybe*

obscuroides belongs to the difficult “*cincinnata*-group” with lilac stipe and mostly were identified as *I. cincinnata* var. *major* but the taxonomic studies of Bandini et al. (2021a) revealed that this taxon deserves species rank based on molecular genetic and morphological differences. *Inocybe griseotarda* Poirier, a rather frequent species in Italy, often associated with *Pinus sylvestris* (Bizio et al., 2017) was synonymized with *I. psammobrunnea* Bon by Bandini et al. (2021a). *Inocybe ghibliana* is a newly described species, reported by Bandini et al. (2021a) (our collections M5, M9 were also studied in this paper). Public databases contain around 10 EcM and basidiomata sequences only from Europe which possibly belong to this species. *Inocybe ghibliana* also prefers growing in dry habitats even in extreme locations, e.g., sun exposed habitats (Bandini et al., 2021a). Frequently collected under conifers but broadleaved tree species can be similarly adequate mycorrhizal partners. Our collections were collected in a park on sandy soil under *Carpinus betulus* and *Picea* sp.

The genus *Mallocybe* and *Pseudosperma* are less studied clades of Inocybaceae in Europe. We could find three taxa of *Mallocybe* and two of *Pseudosperma* but most of the specimens were labelled only tentatively through no available sequences of the type specimens found in the public databases. In the case of *Mallocybe* aff. *heimii* (M117) and *M. aff. plebeia* (M118) interesting collections (Figs. 2 and 3) and also the already discussed *M. aff. malenconii* (M120) (Fig. 4) (Csizmár et al., 2021), our ITS phylogenetic tree shows that all sequences represent three different species level groups (Fig. 1). Thus, these taxa need more morphological and multigene phylogenetic examinations to clarify their taxonomic positions.

Conclusions

The new era of phylogenetic research on family Inocybaceae was also helped reaching new taxonomic views and, very importantly, clarifying rather complicated identification processes based only on morphological characters. Recent studies in Europe revealed numerous new species, and the taxonomists were trying to apply molecular and morphological analyses of the old type specimens which is essential to avoid describing a previously described species as new. The high diversity of the species with complicated morphological characters resulted more misidentifications in already existing fungal literatures, moreover in scientific publications too. Most of the currently published papers with newly described species originated from Germany, France, Spain, Italy or from the northern part of Europe (Estonia, Sweden). From Hungary and the adjacent countries only, a few data on the Inocybaceae are available particularly with molecular support. This paper can contribute and extend the missing published data on Inocybaceae with studying 18 species in Hungary including phylogenetic analysis. Therefore, this study represents that urban areas could also be potentially diverse habitats with wide range of rare or already undescribed macrofungal species.

Acknowledgements. This work was financially supported by the Pannon Breeding Program (GINOP 2.2.1-15-2017-00042). The work of Bálint Dima was supported by the János Bolyai Research Scholarship of the Hungarian Academy of Sciences, the National Research, Development and Innovation Office of Hungary (OTKA FK-143061) and the ELTE Institutional Excellence Program 2020 financed by the National Research, Development and Innovation Office of Hungary (TKP2020-IKA-05). We warmly thank to Balázs Péter for providing more *Inocybe* collections. We also thank to Ádám Heller for helping in mycological observations.

Declaration of competing interests. The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

REFERENCES

- [1] Altschul, S. F., Madden, T. L., Schaffer, A. A., Zhang, J., Zhang, Z., Miller, W., Lipman, D. J. (1997): Gapped BLAST and PSI-BLAST: a new generation of protein database search programs. – *Nucleic Acids Res* 25: 3389-3402.
- [2] Angel, S., Sheppard, S. C., Civco, D. L. (2005): *The Dynamics of Global Urban Expansion*. – Transport and Urban Development Department, World Bank, Washington, DC.
- [3] Bandini, D., Oertel, B. (2020): Three new species of the genus *Pseudosperma* (Inocybaceae). – *Czech Mycol* 72(2): 221-250.
- [4] Bandini, D., Oertel, B., Ploch, S., Ali, T., Vauras, J., Schneider, A., Scholler, M., Eberhardt, U., Thines, M. (2018): Revision of some central European species of *Inocybe* (Fr.: Fr.) Fr. subgenus *Inocybe*, with the description of five new species. – *Mycol Prog* 18: 247-294.
- [5] Bandini, D., Oertel, B., Moreau, P. A., Thines, M., Ploch, S. (2019): Three new hygrophilous species of *Inocybe*, subgenus *Inocybe*. – *Mycol Prog* 18: 1101-1119.
- [6] Bandini, D., Vauras, J., Weholt, Ø., Oertel, B., Eberhardt, U. (2020a): *Inocybe woglindeana*, a new species of the genus *Inocybe*, thriving in exposed habitats with calcareous sandy soil. – *Karstenia* 58: 41-59.
- [7] Bandini, D., Oertel, B., Schüssler, C., Eberhardt, U. (2020b): Noch mehr Risspilze: fünfzehn neue und zwei wenig bekannte Arten der Gattung *Inocybe*. – *Mycologia Bavarica* 20: 13-101.
- [8] Bandini, D., Sesli, E., Oertel, B., Krisai-Greilhuber, I. (2020c): *Inocybe antoniniana*, a new species of *Inocybe* section *Marginatae* with nodulose spores. – *Sydowia* 72: 95-106.
- [9] Bandini, D., Oertel, B., Eberhardt, U. (2021a): A fresh outlook on the smooth-spored species of *Inocybe*: type studies and 18 new species. – *Mycol Prog* 20: 1019-1114.
- [10] Bandini, D., Oertel, B., Eberhardt, U. (2021b): *Inocybe blandula*, eine neue höckersporige Art der Gattung *Inocybe*, Sektion *Marginatae*. – *Zeitschrift für Mykologie* 87(2): 211-228.
- [11] Bandini, D., Oertel, B., Eberhardt, U. (2021c): Noch mehr Risspilze (2): Dreizehn neue Arten der Familie Inocybaceae. – *Mycologia Bavarica* 21: 27-98.
- [12] Bizio, E., Ferisin, G., Dovana, F. (2017): Note sul campo di variabilità di *Inocybe griseotarda*. – *Rivista di Micologia* 60(1): 59-70.
- [13] Carteret, X., Reumaux, P. („2011”, publ. 2012.): Miettes sur les *Inocybes* (6e série); étude de quelques nains de feuillus de la plaine, accompagnée d’une clé de détermination des taxons de la section *Lilacinae* R. Heim. – *Bull Trimest Soc Mycol Fr* 127(1/2): 1-53.
- [14] Cervini, M. (2021): *Inocybe messapica* (Inocybaceae, Agaricales, Basidiomycota), a new species in section *Splendentes*, from Mediterranean oak woods. – *Phytotaxa* 480(2): 174-184.
- [15] Cervini, M., Bizio, E., Alvarado, P. (2020): Quattro nuove specie italiane del Genere *Pseudosperma* (Inocybaceae) con odore di miele. – *Rivista di micologia* 63(1): 3-36.
- [16] Csizmár, M., Cseh, P., Dima, B., Orlóci, L., Bratek, Z. (2021): Macrofungi of urban *Tilia* avenues and gardens in Hungary. – *Glob Ecol Conserv* 28: e0167228.
- [17] Daskalopoulos, V., Polemis, E., Fryssouli, V., Kottis, L., Bandini, D., Dima, B., Zervakis, G. I. (2021): *Mallochybe heimii* ectomycorrhizae with *Cistus creticus* and *Pinus halepensis* in Mediterranean littoral sand dunes - assessment of phylogenetic relationships to *M. arenaria* and *M. agardhii*. – *Mycorrhiza* (4): 497-510.

- [18] Dovana, F., Bizio, E., Garbelotto, M., Ferisin, G. (2021): *Inocybe cervenianensis* (Agaricales, Inocybaceae), a new species in the *I. flavoalbida* clade from Italy. – *Phytotaxa* 484: 227-236.
- [19] Gardes, M., Bruns, T. D. (1993): ITS primers with enhanced specificity for basidiomycetes application to the identification of mycorrhizae and rusts. – *Mol Ecol* 2: 113-118.
- [20] Güneralp, B., Reba, M., Hales, B., Wentz, E., Seto, K. (2020): Trends in urban land expansion, density, and land transitions from 1970 to 2010: a global synthesis. – *Environ Res Lett* 15(4): 044015.
- [21] Katoh, K., Toh, H. (2008): Recent developments in the MAFFT multiple sequence alignment program. – *Brief Bioinformatics* 9: 286-298.
- [22] Kumar, S., Stecher, G., Li, M., Knyaz, C., Tamura, K. (2018): MEGA X: molecular evolutionary genetics analysis across computing platforms. – *Mol Biol Evol* 35(6): 1547-1549.
- [23] Kuyper, T. (1986): A revision of the genus *Inocybe* in Europe. I. Subgenus *Inosperma* and the smooth-spored species of subgenus *Inocybe*. – *Persoonia Supplement* 3(1): 1-247.
- [24] La Rosa, A., Bizio, E., Saitta, A., Tedersoo, L. (2017): *Inocybe castaneicolor* (Agaricales, Basidiomycota), a new species in section Splendentes. – *Phytotaxa* 316(1): 79-87.
- [25] Matheny, P. B. (2005): Improving phylogenetic inference of mushrooms using RPB1 and RPB2 sequences (*Inocybe*; Agaricales). – *Mol Phylogenet Evol* 35: 1-20.
- [26] Matheny, P., Kudzma, L. V. (2019): New species of *Inocybe* (Inocybaceae) from eastern North America. – *J Torrey Bot Soc* 146(3): 213.
- [27] Matheny, P. B., Hobbs, A., Esteve-Raventós, F. (2019): Genera of Inocybaceae: new skin for the old ceremony. – *Mycologia* 112(102): 1-38.
- [28] Mesic, A., Haelewaters, D., Tkalcec, Z., Liu, J., Kušan, I., Aime, M., Pošta, A. (2021): *Inocybe brijunica* sp. nov., a new ectomycorrhizal fungus from Mediterranean Croatia revealed by morphology and multilocus phylogenetic analysis. – *J Fungi* 7(3): 199.
- [29] Miller, M. A., Pfeiffer, W., Schwartz, T. (2010): Creating the CIPRES science gateway for inference of large phylogenetic trees. – *Proceedings of the Gateway Computing Environments Workshop (GCE)*, New Orleans, Louisiana.
- [30] Nagy, I., Nagy, G. L. (2011): *Inocybe aeruginascens* and *Inocybe javorkae* (Agaricales, Basidiomycota), two species described from Hungary. – *Clusiana* 50(1): 23-35.
- [31] Nilsson, R. H., Larsson, K. H., Taylor, A. F. S., Bengtsson-Palme, J., Jeppesen, T. S., Schigel, D., Kennedy, P., Picard, K., Glöckner, F. O., Tedersoo, L., Saar, I., Kõljalg, U., Abarenkov, K. (2018): The UNITE database for molecular identification of fungi: handling dark taxa and parallel taxonomic classifications. – *Nucleic Acids Res* 46(12): D259-D264. DOI: 10.1093/nar/gky1022.
- [32] Oke, C., Bekessy, S. A., Frantzeskaki, N., Bush, J., Fitzsimons, J. A., Garrard, G. E., Grenfell, M., Harrison, L., Hartigan, M., Callow, D., Cotter, B., Gawler, S. (2021): Cities should respond to the biodiversity extinction crisis. – *Urban Sustain* 1, 11.
- [33] Rambaut, A. (2010): FigTree v1.3.1. Institute of Evolutionary Biology. – University of Edinburgh, Edinburgh.
- [34] Ryberg, M., Larsson, E., Jacobsson, S. (2010): An evolutionary perspective on morphological and ecological characters in the mushroom family Inocybaceae (Agaricomycotina, Fungi). – *Mol Phylogenet Evol* 55: 431-442.
- [35] Stamatakis, A. (2014): RAxML version 8. A tool for phylogenetic analysis and post-analysis of large phylogenies. – *Bioinformatics* 30: 1312-1313.
- [36] Vauras, J., Larsson, E. (2020): First records of *Inocybe melleiconica* and *I. pararubens* for Northern Europe with a new variety from the alpine zone of the Scandinavian mountains. – *Karstenia* 58(1): 29-40.
- [37] Vilgalys, R., Hester, M. (1990): Rapid genetic identification and mapping of enzymatically amplified ribosomal DNA from several *Cryptococcus* species. – *J Bacteriol* 172: 4238-4246.

- [38] White, T. J., Bruns, T., Lee, S., Taylor, J. W. (1990): Amplification and Direct Sequencing of Fungal Ribosomal RNA Genes for Phylogenetics. – In: Innis, M. A., Gelfand, D. H., Sninsky, J. J., White, T. J. (eds.) PCR Protocols: A Guide to Methods and applications. Academic Press, New York, pp. 315-322.
- [39] Zhang, Z., Schwartz, S., Wagner, L., Miller, W. (2000): A greedy algorithm for aligning DNA sequences. – J Comput Biol 7(1-2): 203-14.

APPENDIX

Appendix 1. Exact GPS coordinates of collected fungal materials

Specimen voucher	Taxon	Location	GPS coordinate	Habitat type	Date of collection	Hosts	GenBank accession (ITS)
M48	<i>Inocybe aeruginascens</i> Babos	XIII. Budapest	47°31'57.2"N 19°03'03.4"E	Park	4 June 2019	<i>Pinus nigra</i>	OM228846
M114	<i>Inocybe alluvionis</i> Stangl & J. Veselský	XIV. Budapest	47°31'48.2"N 19°07'12.0"E	Garden	27 September 2020	<i>Betula pendula</i> , <i>Salix alba</i>	OM228847
M122	<i>Inocybe alluvionis</i> Stangl & J. Veselský	XI. Budapest	47°28'21.0"N 19°03'37.2"E	Park	23 October 2020	<i>Carpinus betulus</i>	OM228848
M4	<i>Inocybe alluvionis</i> Stangl & J. Veselský	XI. Budapest	47°28'29.7"N 19°03'45.9"E	Park	31 October 2017	<i>Populus</i> sp.	OM228849
M34	<i>Inocybe furfurea</i> Kühner	XI. Budapest	47°28'13.4"N 19°03'38.6"E	Park	20 September 2019	<i>Carpinus betulus</i>	OM228850
M128	<i>Inocybe ghibliana</i> Bandini & B. Oertel	XI. Budapest	47°28'13.7"N 19°03'28.3"E	Park	26 October 2020	<i>Carpinus betulus</i>	OM228851
M5	<i>Inocybe ghibliana</i> Bandini & B. Oertel	XI. Budapest	47°28'13.7"N 19°03'28.3"E	Park	29 September 2017	<i>Carpinus betulus</i>	OM228852
M9	<i>Inocybe ghibliana</i> Bandini & B. Oertel	XIV. Budapest	47°31'46.1"N 19°07'10.7"E	Garden	17 September 2017	<i>Picea pungens</i>	OM228853
M137	<i>Inocybe psammobrunnea</i> Bon	XXII. Budapest	47°25'43.1"N 19°01'21.9"E	Garden	23 November 2020	<i>Picea abies</i>	OM228854
M123	<i>Inocybe griseovelata</i> Kühner	XI. Budapest	47°28'21.0"N 19°03'37.2"E	Park	23 October 2020	<i>Carpinus betulus</i>	OM228855
M140	<i>Inocybe</i> aff. <i>inodora</i> Velen.	XI. Budapest	47°28'14.3"N 19°03'36.8"E	Park	1 June 2021	<i>Tilia tomentosa</i> , <i>Carpinus betulus</i>	OM228856
M119	<i>Inocybe obscuroides</i> P.D. Orton	XIV. Budapest	47°31'47.8"N 19°07'11.9"E	Garden	18 October 2020	<i>Betula pendula</i>	OM228857
M135	<i>Inocybe obscuroides</i> P.D. Orton	VIII. Budapest	47°29'01.1"N 19°05'04.4"E	Botanical Garden	28 October 2020	<i>Picea pungens</i>	OM228858
M132	<i>Inocybe pararubens</i> Carteret & Reumaux	XII. Budapest	47°31'14.9"N 18°56'36.2"E	Garden	12 October 2017	<i>Quercus</i> sp.	OM228859
M2	<i>Inocybe phaeoleuca</i> Kühner	XI. Budapest	47°28'13.7"N 19°03'28.3"E	Park	29 September 2017	<i>Carpinus betulus</i>	OM228860
M125	<i>Inocybe phaeoleuca</i> Kühner	XI. Budapest	47°28'21.0"N 19°03'37.2"E	Park	23 October 2020	<i>Carpinus betulus</i>	OM228861
M127	<i>Inocybe phaeoleuca</i> Kühner	XI. Budapest	47°28'12.9"N 19°03'26.6"E	Park	22 October 2020	<i>Betula pendula</i>	OM228862
M133	<i>Inocybe pusio</i> P. Karst.	VIII. Budapest	47°29'03.8"N 19°05'03.7"E	Botanical Garden	28 October 2020	<i>Quercus robur</i> , <i>Carpinus betulus</i>	OM228863

M46	<i>Inocybe semifulva</i> Grund & D.E. Stuntz	XIII. Budapest	47°31'30.1"N 19°02'48.7"E	Park	4 June 2019	<i>Tilia</i> sp., <i>Pinus</i> sp.	OM228864
INOCBP-1	<i>Inocybe semifulva</i> Grund & D.E. Stuntz	XI. Budapest	47°28'14.3"N 19°03'36.7"E	Park	8 September 2021	<i>Tilia tomentosa</i> , <i>Carpinus betulus</i>	OM228865
M14	<i>Inocybe semifulva</i> Grund & D.E. Stuntz	XIV. Budapest	47°31'46.2"N 19°07'09.5"E	Garden	23 September 2017	<i>Salix alba</i>	OM228866
M141	<i>Inocybe semifulva</i> Grund & D.E. Stuntz	XI. Budapest	47°28'14.3"N 19°03'36.7"E	Park	13 June 2021	<i>Tilia tomentosa</i> , <i>Carpinus betulus</i>	OM228867
M129	<i>Inocybe grammopodia</i> Malençon	VIII. Budapest	47°29'05.2"N 19°05'03.9"E	Botanical Garden	28 October 2020	<i>Quercus robur</i> , <i>Carpinus betulus</i>	OM228868
M117	<i>Mallocybe</i> aff. <i>heimii</i> (Bon) Matheny & Esteve-Rav.	XI. Budapest	47°28'16.6"N 19°03'41.9"E	Tree avenue; Roadside	20 October 2020	<i>Tilia tomentosa</i>	OM228869
M118	<i>Mallocybe</i> aff. <i>plebeia</i> Bandini, B. Oertel & U. Eberh.	XIV. Budapest	47°31'43.6"N 19°07'07.4"E	Garden	18 October 2020	<i>Betula pendula</i> , <i>Picea</i> sp.	OM228870
M120	<i>Mallocybe</i> aff. <i>malenconii</i> (R. Heim) Matheny & Esteve-Rav.	XI. Budapest	47°28'10.1"N 19°03'38.4"E	Tree avenue; Roadside	14 October 2020	<i>Tilia tomentosa</i>	OM228871
M134	<i>Pseudosperma obsoletum</i> (Romagn.) Matheny & Esteve-Rav.	X. Budapest	47°28'43.7"N 19°06'15.7"E	Park	24 June 2020	<i>Populus</i> sp., <i>Pinus</i> sp., <i>Picea</i> sp.	OM228872
M130	<i>Pseudosperma permelliolens</i> (Carteret & Reumaux) Matheny & Esteve-Rav.	VIII. Budapest	47°29'02.3"N 19°05'04.0"E	Botanical Garden	28 October 2020	probably <i>Quercus robur</i> , <i>Carpinus betulus</i>	OM228873