

Multigene phylogeny and taxonomy of the genus Melanconiella (Diaporthales)

**Hermann Voglmayr, Amy Y. Rossman,
Lisa A. Castlebury & Walter M. Jaklitsch**

Fungal Diversity

An International Journal of Mycology

ISSN 1560-2745

Volume 57

Number 1

Fungal Diversity (2012) 57:1-44

DOI 10.1007/s13225-012-0175-8



Your article is protected by copyright and all rights are held exclusively by Mushroom Research Foundation. This e-offprint is for personal use only and shall not be self-archived in electronic repositories. If you wish to self-archive your work, please use the accepted author's version for posting to your own website or your institution's repository. You may further deposit the accepted author's version on a funder's repository at a funder's request, provided it is not made publicly available until 12 months after publication.

Multigene phylogeny and taxonomy of the genus *Melanconiella* (Diaporthales)

Hermann Voglmayr · Amy Y. Rossman ·
Lisa A. Castlebury · Walter M. Jaklitsch

Received: 7 March 2012 / Accepted: 15 May 2012 / Published online: 11 July 2012
© Mushroom Research Foundation 2012

Abstract Molecular phylogenetic analyses of LSU rDNA demonstrate monophyly of the genus *Melanconiella*, and its status as a genus distinct from *Melanconis* is confirmed. Data of macro- and microscopic morphology, pure cultures and phylogenetic analyses of partial SSU-ITS-LSU rDNA, *tef1* and *rpb2* sequences revealed 13 distinct species of *Melanconiella*, six of which are described as new (*M. chryso-discosporina*, *M. chryso-melanconium*, *M. chryso-orientalis*, *M. echinata*, *M. elegans*, *M. meridionalis*). *Melanconiella hyperopta* var. *orientalis* is described as a new variety. *Diaporthe carpinicola*, *D. ellisii*, *D. flavovirens*, *D. hyperopta* and *D. ostryae* are formally combined into *Melanconiella*. The name *Melanconiella chrysostroma* is excluded from *Melanconiella*, as it is an obligate synonym of *Wuestneia xanthostroma*. The type of *Melanconiella* is confirmed as *M. spodiaea*. Several species are lecto- and/or epitypified. A key to all treated species of *Melanconiella* is provided, and the circumscriptions of the genera *Melanconis* and *Melanconiella* are emended. Most *Melanconiella* species revealed by molecular phylogenetic analyses can be well characterised by a suite of morphological traits including ascospore shape, length and width, colour, absence/presence and shape of appendages and the anamorph. Anamorph-teleomorph connections were confirmed by pure culture and DNA data, revealing

the presence of a single melanconium- or discosporina-like anamorph for each species. Colony growth was found to be characteristic of the respective species. *Melanconiella* is shown to be confined to the host family *Betulaceae*, and all species are found to be highly host-specific, mostly confined to a single host species. The biodiversity of *Melanconiella* was determined to be centred on the genus *Carpinus* with nine species, five of which have been confirmed on *C. betulus*. Europe appears to be the geographic centre of *Melanconiella* biodiversity.

Keywords High diversity · Host specificity · *Melanconidaceae* · Molecular phylogeny · New species · Pathogen · Systematics

Introduction

The *Diaporthales* are an order of ascomycetes within the *Sordariomycetidae* that are characterised by forming black perithecia with or without a well-developed stroma or pseudostroma and a diaporthalean-type centrum development, i.e. with broad deliquescent paraphyses and asci that float free at maturity, often having a refractive apical ring (Barr 1978; Castlebury et al. 2002). Many species occur on hardwood trees in temperate regions although a few species have been discovered in subtropical areas. Many species are opportunistic pathogens and endophytes (Sieber 2007), and some of those are serious threats to forestry and agriculture.

Within the *Diaporthales*, the family *Melanconidaceae* has traditionally been conceived to include many genera having perithecia immersed in a well-developed stroma with ostioles (“beaks”) that emerge through an ectostromatic disc (Barr 1978). Based on phylogenetic data, it was concluded that the *Melanconidaceae* should be restricted to a single genus *Melanconis* (Castlebury et al. 2002). This narrowly

H. Voglmayr (✉) · W. M. Jaklitsch
Department of Systematic and Evolutionary Botany,
Faculty Centre of Biodiversity, University of Vienna,
Rennweg 14,
1030, Wien, Austria
e-mail: hermann.voglmayr@univie.ac.at

A. Y. Rossman · L. A. Castlebury
Systematic Mycology & Microbiology Laboratory, USDA,
Agricultural Research Service,
B010A, 10300 Baltimore Ave,
Beltsville, MD 20705, USA

circumscribed family is closely related to the *Gnomoniaceae*. Castlebury et al. (2002) determined that species previously placed in the genus *Melanconis* are phylogenetically distributed throughout the *Diaporthales*.

The genus *Melanconis*, based on the type species *M. stilbostoma*, includes species with perithecia immersed in the substrate arranged in a circle with oblique or lateral ostioles convergent and erumpent through an ectostromatic disc and hyaline ascospores. *Melanconis stilbostoma* occurs on *Betula* and is congeneric with *M. alni* and *M. marginalis*, which also occur on members of the *Betulaceae* (Castlebury et al. 2002).

The genus *Melanconiella* Sacc. 1882 was established for species that are similar to *Melanconis* but have dark coloured ascospores, although even Saccardo placed a hyaline-spored species (*M. chrysostroma*) in this genus. Subsequently, 21 taxa were placed in *Melanconiella* primarily in the late nineteenth and early twentieth century. Following Wehmeyer (1941), recent authors including Müller and von Arx (1962) and Barr (1978) considered *Melanconiella* to be a synonym of *Melanconis*, whereas Munk (1957), Petrak (1952a) and Dennis (1968) preferred to maintain separate genera.

During many years of collecting, the last author noticed a high variability of ascospore size and morphology, especially within the hyaline-spored *Melanconis* species growing on *Carpinus betulus* and *Ostrya carpinifolia*, which all keyed out as *Melanconis* (= *Melanconiella*) *chrysostroma* based on the available taxonomic literature (Wehmeyer 1941; Ellis and Ellis 1997). Results of recent studies on ascomycetes with similar ecology (e.g. Voglmayr and Jaklitsch 2008, 2011; Mejía et al. 2011a) revealed a much higher biodiversity than previously perceived, which cast doubt on the wide species concept commonly applied to *Melanconiella chrysostroma*. The fact that no recent monographic treatment is available for the genera *Melanconis* and *Melanconiella* led us to initiate thorough morphological and molecular phylogenetic studies on *Melanconiella* involving extensive studies of numerous fresh collections and types from various hosts to evaluate morphological variability, resolve species boundaries and clarify host ranges. Cultures were obtained from both ascospores and conidia to elucidate the anamorph-teleomorph connections by DNA data. In addition, molecular phylogenetic analyses were performed to determine the phylogenetic relationships within the genus as well as the phylogenetic position of *Melanconiella* within the *Diaporthales*.

Materials and methods

Sample sources

Details of the specimens used for morphological investigations are listed in the [Taxonomy](#) section under the respective descriptions. The herbarium acronyms are given according to Thiers

(2012): B—Botanischer Garten und Botanisches Museum Berlin-Dahlem, Germany; BPI—U.S. National Fungus Collections, Beltsville, USA; DAOM—Agriculture and Agri-Food Canada, Ottawa, Canada; G—Conservatoire et Jardin botaniques de la Ville de Genève, Switzerland; GZU—Karl-Franzens-Universität Graz, Austria; K—Royal Botanic Gardens Kew, UK; M—Botanische Staatssammlung München, Germany; NY—New York Botanical Garden, USA; UPS—Uppsala University, Sweden; W—Naturhistorisches Museum Wien, Austria; WU—Universität Wien, Austria.

Morphology

Morphological observations and measurements were carried out on a Nikon SMZ 1500 stereo-microscope and after mounting in tap water or 3 % KOH on a Zeiss AxioImager A1 compound microscope using Nomarski differential interference contrast (DIC). Images were recorded with a Nikon DS-Fi1, a Nikon Coolpix 4500 (cultures) or a Zeiss AxioCam ICc3 digital camera. Measurements are reported as maxima and minima in parentheses and the mean plus and minus the standard deviation of a number of measurements given in parentheses.

Culture preparation, DNA extraction, PCR and sequencing

Single ascospore or conidium isolates were prepared and grown on 2 % malt extract agar (MEA), or on 2 % corn meal agar plus 2 % w/v dextrose (CMD) with subsequent transfer and storage on CM agar slants. To record growth characteristics, they were grown on 2 % MEA at room temperature under natural daylight.

Mycelium for DNA extraction was grown, freeze-dried and ground according to Voglmayr and Jaklitsch (2011). DNA was extracted using the modified CTAB protocol described in Riethmüller et al. (2002). In collections of *Melanconiella echinata*, *M. elegans* and *M. ellisii* for which no cultures could be obtained, DNA was isolated directly from ascomata or conidiomata according to Voglmayr and Jaklitsch (2011), but after the chloroform-isoamyl alcohol step of the CTAB extraction, the supernatant was further processed with the NucleoSpin Extract II kit (Macherey-Nagel, Düren, Germany) according to the manufacturer's instructions to obtain highly purified DNA.

The partial nuSSU-complete ITS-partial nuLSU rDNA region was amplified using the primers V9G (de Hoog and Gerrits van den Ende 1998) and LR5 (Vilgalys and Hester 1990). For amplification of DNA from herbarium specimens, ITS and LSU were sometimes amplified separately, with the primers V9G or ITS5 (White et al. 1990) and LR1 for ITS and with LR0R and LR5 (Vilgalys and Hester 1990) for LSU. For the type specimen of *M. echinata*, ITS1 and ITS2 were separately amplified with the primers ITS5/f5.8SR and f5.8SF

(Jaklitsch and Voglmayr 2011)/LR1, respectively. As the LSU sequence of *M. echinata* could not be revealed with the primers listed above, it was amplified as three separate fragments about 300 bp long, using the primers LR0R/LR2-A (5' TGCTTTTCATCTTTCGATCAC 3'), LR2R-A (5' CAGAGACCGATAGCGCAC 3')/LR3 (Vilgalys and Hester 1990) and LR3R (<http://www.biology.duke.edu/fungi/mycolab/primers.htm>)/LR5. A ca. 1.4 kb fragment of the *tefl* (translation elongation factor 1 alpha) gene was amplified with the primers EF1728F (Chaverri and Samuels 2003) and TEF1LLErev (Jaklitsch et al. 2006). The latter fragment includes the fourth and the fifth intron and a significant portion of the last large exon. A ca. 1.2 kb fragment of RNA polymerase II subunit B (*rpb2*) was amplified using the primer pair fRPB2-5f and fRPB2-7cr (Liu et al. 1999). PCR products were purified using an enzymatic PCR cleanup (Werle et al. 1994) as described in Voglmayr and Jaklitsch (2008). DNA was cycle-sequenced using the ABI PRISM Big Dye Terminator Cycle Sequencing Ready Reaction Kit v. 3.1 (Applied Biosystems, Warrington, UK) and the PCR primers; in addition, primers ITS4 (White et al. 1990) and LR3 were used for the partial nuSSU-complete ITS-partial nuLSU rDNA region. Sequencing was performed on an automated DNA sequencer (ABI 3730xl Genetic Analyzer, Applied Biosystems).

Data analysis

To reveal the phylogenetic position of *Melanconiella* within the *Diaporthales* and to investigate whether the genus is monophyletic, a phylogenetic analysis was performed with nuLSU rDNA sequences. Sequences of representative species were selected from Castlebury et al. (2002) and supplemented with sequences from GenBank; two accessions of *Gaeumannomyces* (*Magnaportheaceae*) were included as outgroup. GenBank accession numbers are given in the phylogenetic tree (Fig. 1). For detailed investigations of species relationships and delimitation within *Melanconiella*, a combined matrix of nuSSU-ITS1-5.8S-ITS2-LSU rDNA, *rpb2* and *tefl* sequences was produced for phylogenetic analyses, with *Melanconis stilbostoma* as outgroup. The GenBank accession numbers of sequences used in these analyses are given in Table 1.

Sequence alignments for phylogenetic analyses were produced with either MUSCLE version 3.6 (Edgar 2004) or MAFFT 6.847 (Katoh et al. 2002; Katoh and Toh 2008) implemented in UGENE 1.10.0 (<http://ugene.unipro.ru>), with a maximum of 100 iterative refinements and gap opening penalties of 1.03 for ITS-LSU and 0.83 for *tefl*, respectively. The resulting alignments were checked using BioEdit version 7.0.9.0 (Hall 1999). After the exclusion of excessive leading and trailing gap regions and of ambiguously aligned positions,

the LSU matrix contained 1363 characters. The combined data matrix contained 4370 characters; viz. 1689 nucleotides of SSU-ITS-LSU, 1173 nucleotides of *rpb2* and 1508 nucleotides of *tefl*.

In Maximum Likelihood (ML) and Bayesian analyses, substitution model parameters were calculated separately for the different gene regions included in the combined analyses. For ML analyses, 500 rounds of random addition of sequences as well as 500 fast bootstrap replicates were computed with RAxML (Stamatakis 2006) as implemented in raxmlGUI 0.95 (Silvestro and Michalak 2011) using the GTRGAMMA algorithm.

For Bayesian analyses (BA), the GTR + I + G model was implemented. Bayesian analyses were performed with the computer program MrBayes (version 3.1.2; Huelsenbeck and Ronquist 2001). Three parallel runs of four incrementally heated, simultaneous Markov chains were performed over 5 million generations of which every 500th and 100th tree was sampled in each run in the LSU and combined analyses, respectively. The first 1000 and 10000 trees sampled were discarded in the LSU and combined analyses, respectively, and a 90 % majority rule consensus of the remaining trees was computed to obtain posterior probabilities. To test convergence of runs, the results were analysed using AWTY (Nylander et al. 2008); no indication of lack of convergence was detected.

Maximum parsimony (MP) bootstrap analyses were performed with PAUP* version 4.0 b10 (Swofford 2002), using 1000 replicates of heuristic search with 5 rounds of random addition of sequences and subsequent TBR branch swapping (MULTREES option in effect, COLLAPSE = MAXBRLEN, steepest descent option not in effect); each bootstrap replicate was limited to 1 million rearrangements.

Results

Molecular phylogenetic analyses

Of the 1363 characters in the LSU alignment, 218 were parsimony informative. Figure 1 shows the best ML tree (lnL = -5505.723). Tree topologies of the Bayesian analyses were fully compatible with the ML analyses. The three Bayesian runs revealed highly similar posterior probabilities (PP). ML and MP bootstrap support above 60 % and Bayesian posterior probabilities above 90 % are given in Fig. 1 at the first, second and third positions, respectively, above or below the branches.

Of the 4370 characters in the combined data matrix, 1009 were parsimony informative. Figure 2 shows the best ML tree (lnL = -16890.279). Tree topologies of the Bayesian analyses were compatible with the ML tree except for minor topological differences within some species; in addition, *M.*

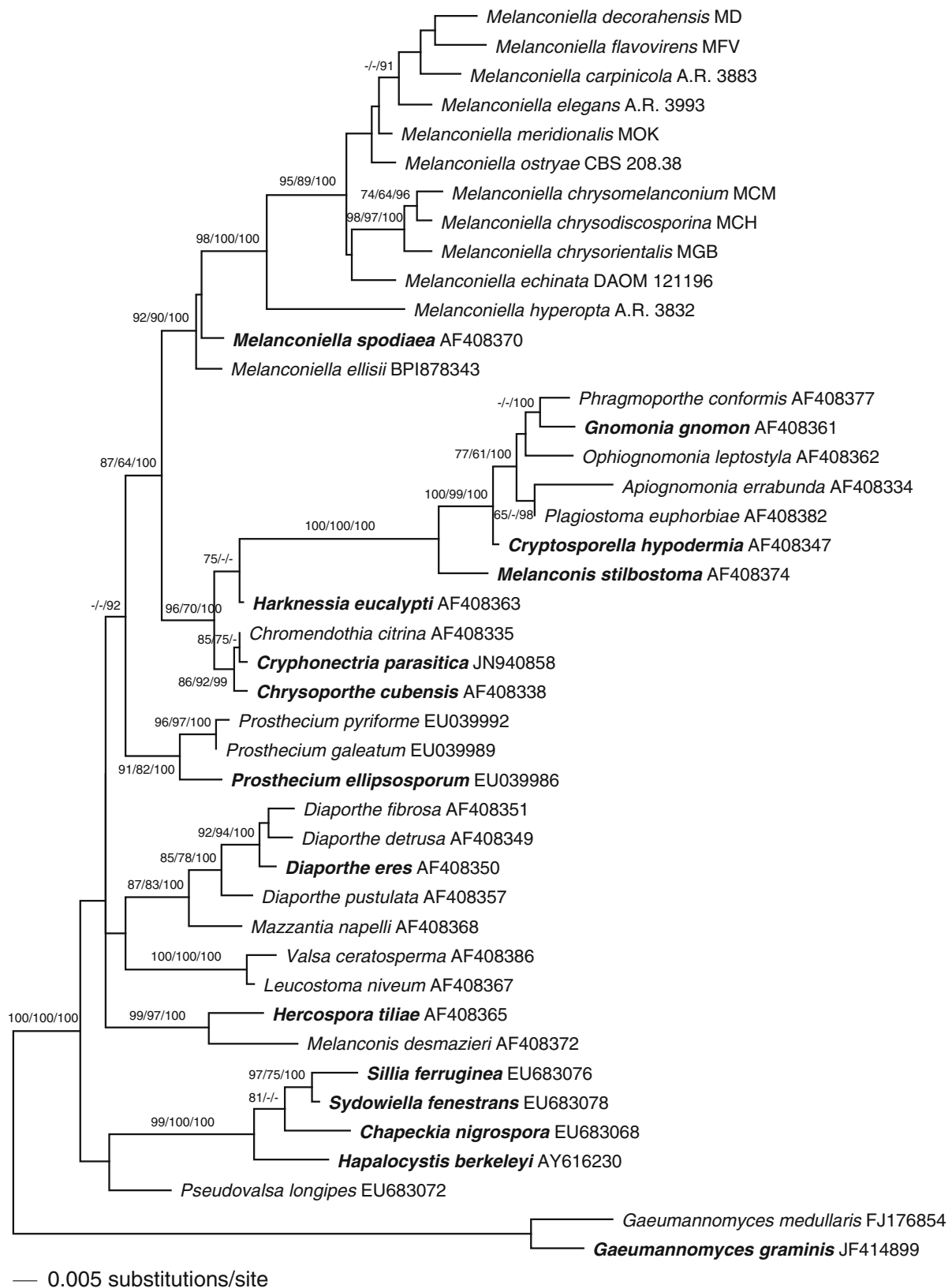


Fig. 1 Phylogram of the best ML tree (lnL=-5505.723) revealed by RAxML from an analysis of the LSU matrix of selected *Diaporthales*, showing the phylogenetic position of *Melanconiella*. ML and MP bootstrap support above 60 % and Bayesian posterior probabilities

above 90 % are given at the first, second and third positions, respectively, above or below the branches. GenBank accession numbers are given following the taxon names; generic types are formatted in bold

Table 1 Taxa, sources of DNA extracts and GenBank accession numbers of *Melanconis* and *Melanconiella* sequences used for the phylogenetic multi-gene analyses or species identification. Details on the strains are found under specimens examined in the [Taxonomy](#) section

Taxon	Strain/culture or specimen	Isolated/extracted from	ITS-LSU	<i>rpb2</i>	<i>tef1</i>
<i>Melanconiella carpinicola</i>	A.R. 3883	ascospore	JQ926230		
<i>M. carpinicola</i>	MENU	conidium	JQ926231	JQ926303	JQ926369
<i>M. carpinicola</i>	MNM	ascospore	JQ926232	JQ926304	JQ926370
<i>M. carpinicola</i>	MNP	ascospore	JQ926233	JQ926305	JQ926371
<i>M. carpinicola</i>	MNUK	ascospore	JQ926234	JQ926306	JQ926372
<i>M. carpinicola</i>	MSMI	ascospore	JQ926235	JQ926307	JQ926373
<i>M. carpinicola</i>	MSR	ascospore	JQ926236	JQ926308	JQ926374
<i>M. chrysodiscosporina</i>	MCAAn	conidium	JQ926237	JQ926309	JQ926375
<i>M. chrysodiscosporina</i>	MCH	ascospore	JQ926238	JQ926310	JQ926376
<i>M. chrysodiscosporina</i>	MCHA	conidium	JQ926239	JQ926311	JQ926377
<i>M. chrysodiscosporina</i>	MEE	conidium	JQ926240	JQ926312	JQ926378
<i>M. chrysodiscosporina</i>	MEL	ascospore	JQ926241	JQ926313	JQ926379
<i>M. chrysodiscosporina</i>	MGG	ascospore	JQ926242	JQ926314	JQ926380
<i>M. chrysodiscosporina</i>	MGJ	ascospore	JQ926243	JQ926315	JQ926381
<i>M. chrysodiscosporina</i>	MH1	ascospore	JQ926244	JQ926316	JQ926382
<i>M. chrysodiscosporina</i>	MH2	ascospore	JQ926245	JQ926317	JQ926383
<i>M. chrysodiscosporina</i>	MRi	ascospore	JQ926246	JQ926318	JQ926384
<i>M. chryso melanconium</i>	MCM	ascospore	JQ926247	JQ926319	JQ926385
<i>M. chryso melanconium</i>	MEH	conidium	JQ926248	JQ926320	JQ926386
<i>M. chryso melanconium</i>	MEUK	conidium	JQ926249	JQ926321	JQ926387
<i>M. chryso melanconium</i>	MGH	ascospore	JQ926250	JQ926322	JQ926388
<i>M. chryso melanconium</i>	MGL	ascospore	JQ926251	JQ926323	JQ926389
<i>M. chryso melanconium</i>	MGR	ascospore	JQ926252	JQ926324	JQ926390
<i>M. chryso melanconium</i>	MGS	ascospore	JQ926253	JQ926325	JQ926391
<i>M. chryso melanconium</i>	MGSt	ascospore	JQ926254	JQ926326	JQ926392
<i>M. chryso melanconium</i>	MGUK	ascospore	JQ926255	JQ926327	JQ926393
<i>M. chryso orientalis</i>	MGB	ascospore	JQ926256	JQ926328	JQ926394
<i>M. chryso orientalis</i>	MGP	ascospore	JQ926257	JQ926329	JQ926395
<i>M. chryso orientalis</i>	MGV	ascospore	JQ926258	JQ926330	JQ926396
<i>M. chryso orientalis</i>	MVH	conidium	JQ926259	JQ926331	JQ926397
<i>M. decorahensis</i>	CBS 159.26*	ascospore	JQ926260	JQ926332	JQ926398
<i>M. decorahensis</i>	MD	ascospore	JQ926261	JQ926333	JQ926399
<i>M. decorahensis</i>	MED	conidium	JQ926262	JQ926334	JQ926400
<i>M. echinata</i>	DAOM 121196**	stroma	JQ926263		
<i>M. elegans</i>	CBS 131494*	ascospore	JQ926264	JQ926335	JQ926401
<i>M. elegans</i>	CBS 131617*	ascospore	JQ926265	JQ926336	JQ926402
<i>M. elegans</i>	BPI 843574**	conidioma	JQ926266	JQ926337	JQ926403
<i>M. elegans</i>	BPI 872067**	conidioma	JQ926267	JQ926338	JQ926404
<i>M. ellisii</i>	BPI 843491**	stroma	JQ926268		JQ926405
<i>M. ellisii</i>	BPI 883227**	conidioma	JQ926269		
<i>M. ellisii</i>	BPI 883227**	stroma	JQ926270		
<i>M. ellisii</i>	BPI 878343**	stroma	JQ926271	JQ926339	JQ926406
<i>M. flavovirens</i>	MFA	conidium	JQ926272	JQ926340	JQ926407
<i>M. flavovirens</i>	MFV	ascospore	JQ926273	JQ926341	JQ926408
<i>M. flavovirens</i>	MFV1	ascospore	JQ926274	JQ926342	JQ926409
<i>M. flavovirens</i>	MFV2	ascospore	JQ926275	JQ926343	JQ926410
<i>M. flavovirens</i>	MFV3	ascospore	JQ926276	JQ926344	JQ926411
<i>M. hyperopta</i>	A.R. 3619	ascospore	JQ926277		

Table 1 (continued)

Taxon	Strain/culture or specimen	Isolated/extracted from	ITS-LSU	<i>rpb2</i>	<i>tefl</i>
<i>M. hyperopta</i>	A.R. 3832	ascospore	JQ926278		
<i>M. hyperopta</i>	CBS 114600*	ascospore	JQ926279	JQ926345	JQ926412
<i>M. hyperopta</i>	MCHBV	ascospore	JQ926280	JQ926346	JQ926413
<i>M. hyperopta</i>	MCHE	ascospore	JQ926281	JQ926347	JQ926414
<i>M. hyperopta</i>	MCL	ascospore	JQ926282	JQ926348	JQ926415
<i>M. hyperopta</i>	MCR	ascospore	JQ926283	JQ926349	JQ926416
<i>M. hyperopta</i>	MCS	ascospore	JQ926284	JQ926350	JQ926417
<i>M. hyperopta</i>	MHG	conidium	JQ926285	JQ926351	JQ926418
<i>M. hyperopta</i> var. <i>orientalis</i>	MHP	ascospore	JQ926288	JQ926352	JQ926420
<i>M. hyperopta</i> var. <i>orientalis</i>	MHVA	conidium	JQ926287	JQ926353	JQ926419
<i>M. hyperopta</i> var. <i>orientalis</i>	MSK	ascospore	JQ926286	JQ926354	JQ926421
<i>M. meridionalis</i>	MOA	conidium	JQ926289	JQ926355	JQ926422
<i>M. meridionalis</i>	MOK	ascospore	JQ926290	JQ926356	JQ926423
<i>M. meridionalis</i>	MOM	ascospore	JQ926291	JQ926357	JQ926424
<i>M. meridionalis</i>	MON	ascospore	JQ926292	JQ926358	JQ926425
<i>M. meridionalis</i>	MOS	ascospore	JQ926293	JQ926359	JQ926426
<i>M. meridionalis</i>	MOSJ	ascospore	JQ926294	JQ926360	JQ926427
<i>M. meridionalis</i>	MOSO	ascospore	JQ926295	JQ926361	JQ926428
<i>M. meridionalis</i>	MOU	ascospore	JQ926296	JQ926362	JQ926429
<i>M. ostryae</i>	CBS 208.38*	ascospore	JQ926297	JQ926363	JQ926430
<i>M. spodiaea</i>	MSH	ascospore	JQ926298	JQ926364	JQ926431
<i>M. spodiaea</i>	MVS	conidium	JQ926299	JQ926365	JQ926432
<i>M. spodiaea</i>	SPOD	ascospore	JQ926300	JQ926366	JQ926433
<i>M. spodiaea</i>	SPOD1	conidium	JQ926301	JQ926367	JQ926434
<i>Melanconis stilbostoma</i>	CBS 121894*	ascospore	JQ926229	JQ926302	JQ926368
<i>M. stilbostoma</i>	CBS 109778*	ascospore	AF408374	EU219299	DQ836910

* DNA isolated from CBS cultures, ** from herbarium specimens

chrysodiscosporina, *M. chrysmelanconium*, *M. chrysorientalis*, *M. decorahensis*, *M. flavovirens*, *M. meridionalis* and *M. ostryae* form a monophylum with moderate support (92–95 % PP) in the Bayesian trees (not shown). The three Bayesian runs revealed highly similar posterior probabilities. ML bootstrap support above 70 %, MP bootstrap support above 70 % and Bayesian posterior probabilities above 90 % are given in Fig. 2 at the first, second and third positions, respectively, above or below the branches.

In most cases, ITS sequences were identical or nearly identical within the species. However, in one accession of *M. chrysodiscosporina* (MEL), the ITS showed major differences, and another accession of the same species (MCAn) had two different ITS copies, but both accessions had *rpb2* and *tefl* sequences identical to other accessions of *M. chrysodiscosporina*. Therefore, the ITS sequences of both deviating accessions were removed from the combined matrix before analysis.

Molecular phylogenetic analyses of the nuLSU matrix (Fig. 1) confirmed that *Melanconiella* based on *M. spodiaea*

is distinct from *Melanconis*, the latter represented in our study by its generic type, *M. stilbostoma*. In addition, monophyly of *Melanconiella* is highly supported, but its closest relatives within the *Diaportheales* remain uncertain due to partly low backbone support. Internal support is mostly lacking within *Melanconiella* in Fig. 1, but the basal position of *M. ellisii*/*M. spodiaea*, the position of their neighbour *M. hyperopta* and sister group relationship of *M. hyperopta* to all other *Melanconiella* species received high support (Fig. 1). All 13 *Melanconiella* species included in the LSU matrix are revealed as phylogenetically distinct (Fig. 1).

Molecular phylogenetic analyses of the combined matrix revealed 12 distinct lineages within *Melanconiella* (Fig. 2), which are recognised at the species level, and one described at the varietal level. Of these, *Melanconiella spodiaea* and *M. decorahensis* have already been validly combined; five lineages represent already described taxa requiring formal transfer to *Melanconiella* (*Diaporthe carpinicola*, *D. ellisii*, *D. flavovirens*, *D. hyperopta* and *D. ostryae*); and five represent yet undescribed species. All species were

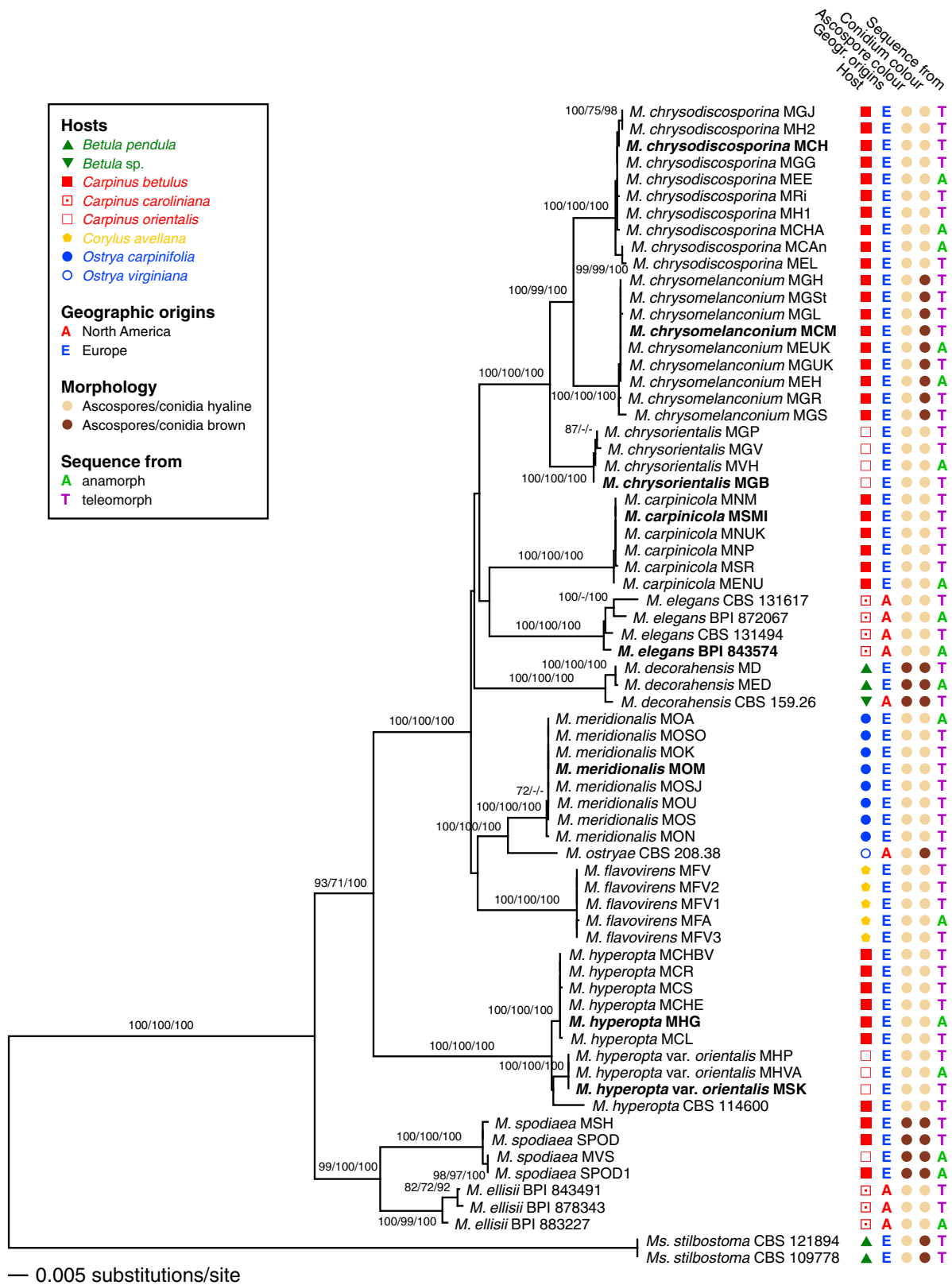


Fig. 2 Phylogram of the best ML tree (lnL=−16890.279) revealed by RAxML from an analysis of the combined ITS-LSU-*rbp2-tef1* matrix of *Melanconiella*, with *Melanconis stilbostoma* selected as outgroup. ML and MP bootstrap support above 70 % and Bayesian posterior

probabilities above 90 % are given at the first, second and third positions, respectively, above or below the branches. Strain/culture or herbarium specimen are given following the taxon names; holo- or epitype strains/specimens are formatted in bold

molecularly highly distinct and received high internal support. The clade containing *M. ellisii* and *M. spodiaea* was placed as sister to the other species with medium (MP) to high (ML, BA) support (Fig. 2), and within the latter, *M. hyperopta* was sister clade to the residual lineages with high support. Also the two species of *Melanconiella* on *Ostrya* (*M. meridionalis*, *M. ostryae*) form a highly supported clade. *Melanconiella chrysodiscosporina*, *M. chrysmelanconium* and *M. chrysorientalis*, which are highly similar in their stroma and ascospore features, consistently formed a highly supported monophylum in nuLSU and combined analyses, and relationships within that clade also received high support.

Anamorph-teleomorph relationships and cultures

For each *Melanconiella* species, a single characteristic anamorph could be identified by DNA data obtained from ascospore and conidial cultures or directly from ascomata and conidiomata, apart from *M. echinata* for which no anamorph is known. Both dark brown melanconium-like conidia as well as hyaline discosporina-like conidia were present within the genus *Melanconiella*, but both were never found within the same species; conidial morphology was constant within a species. The production of dark brown and hyaline conidia did not correlate with phylogenetic relationships, but in several cases, phylogenetic species pairs with one having hyaline, the other dark brown conidia were revealed (*M. chrysodiscosporina*/*M. chrysmelanconium*; *M. meridionalis*/*M. ostryae*).

Culture characteristics like growth rates, morphological patterns of colonies on MEA and pigment formation were observed to be highly distinct for each species (Fig. 3). Conidiation was not observed on MEA cultures, but one PDA culture of *M. elegans* produced aberrant conidia of variable shapes, the size of which was within the range observed on the natural substrate.

Discussion

Circumscription of the genus *Melanconiella*

Morphologically *Melanconiella* is similar to *Melanconis*. Species of the latter genus cause more conspicuous bumps in the bark of the hosts than those of *Melanconiella* and form light-coloured, white or yellowish ectostromatic discs. Wehmeyer (1941) used this trait to distinguish his section *Stilbostomae* from his *Chrysostromae*, which are characterized by dark coloured discs. Light coloured discs are not uncommon in *Melanconiella*, as exemplified by e.g. *M. hyperopta* or *M. meridionalis*. *Melanconiella* as circumscribed here basically matches Wehmeyer's (1941) section

Chrysostromae of his subgenus *Eumelanconis*, except for *M. carthusiana*, *M. desmazierii*, *M. juglandis*, *M. pallida* and *M. platystroma*, which do not belong here. Wehmeyer's (1941) sect. *Stilbostomae* corresponds to *Melanconis* sensu stricto, although *M. apocrypta*, *M. antarctica* and *M. fennica* may not belong there. *Melanconis everhartii* has been recognized in *Macrodiaporthe*. Other subgenera that Wehmeyer (1941) recognized in *Melanconis* have been transferred or relocated to other genera, such as *Chapeckia* (Barr 1978), *Hapalocystis* (Jaklitsch and Voglmayr 2004), *Macrodiaporthe*, *Massariovalsa* (Barr 1978), *Prostheciium* (Voglmayr and Jaklitsch 2008), *Pseudovalsa* (*M. modonia*, De Silva et al. 2009) and *Pseudovalsella* (von Höhnelt 1918).

Molecular phylogeny, species delimitation and barcoding

Extensive morphological and molecular phylogenetic data for *Melanconiella* reveal high species diversity within this genus. Most of the phylogenetically distinct species of *Melanconiella* show a suite of teleomorph characteristics enabling species identification based on morphology. In some cases, teleomorph features do not allow distinction of closely related species, but host association as well as anamorph-teleomorph relationships provide important information for reliable identification. As in many host-specific ascomycetes inhabiting recently dead corticated branches, knowledge of the host is crucial for correct species identification.

At present the genus *Melanconiella* does not appear to belong to any established family within the *Diaporthales*. Phylogenetic analyses remain inconclusive about its relationships within the order (Fig. 1). Although previously placed as a synonym of *Melanconis* in the *Melanconidaceae*, this genus and thus the family have been shown to be closely related to the *Gnomoniaceae* (Fig. 1).

Within the genus *Melanconiella*, all terminal branches are highly supported, however, the infrageneric relationships remain often unresolved due to lack of backbone support (Fig. 2). The short branch lengths in the phylograms indicate that speciation occurred within a short time frame, obscuring the evolutionary relationships.

The ITS region has been proposed as the primary barcoding locus for fungi (Schoch et al. 2012). In most accessions of *Melanconiella* sequenced in our study, the ITS enabled unequivocal identification. However, in *M. chrysodiscosporina* we found a deviating sequence in one accession and two different ITS copies in another accession, whereas both accessions had *rpb2* and *tef1* sequences identical to other accessions of *M. chrysodiscosporina*. This variation indicates that the ITS region is not always suitable for reliable species identification, and results of ITS barcoding have to be treated with caution. In addition, it also demonstrates that a polyphasic approach is necessary in species-

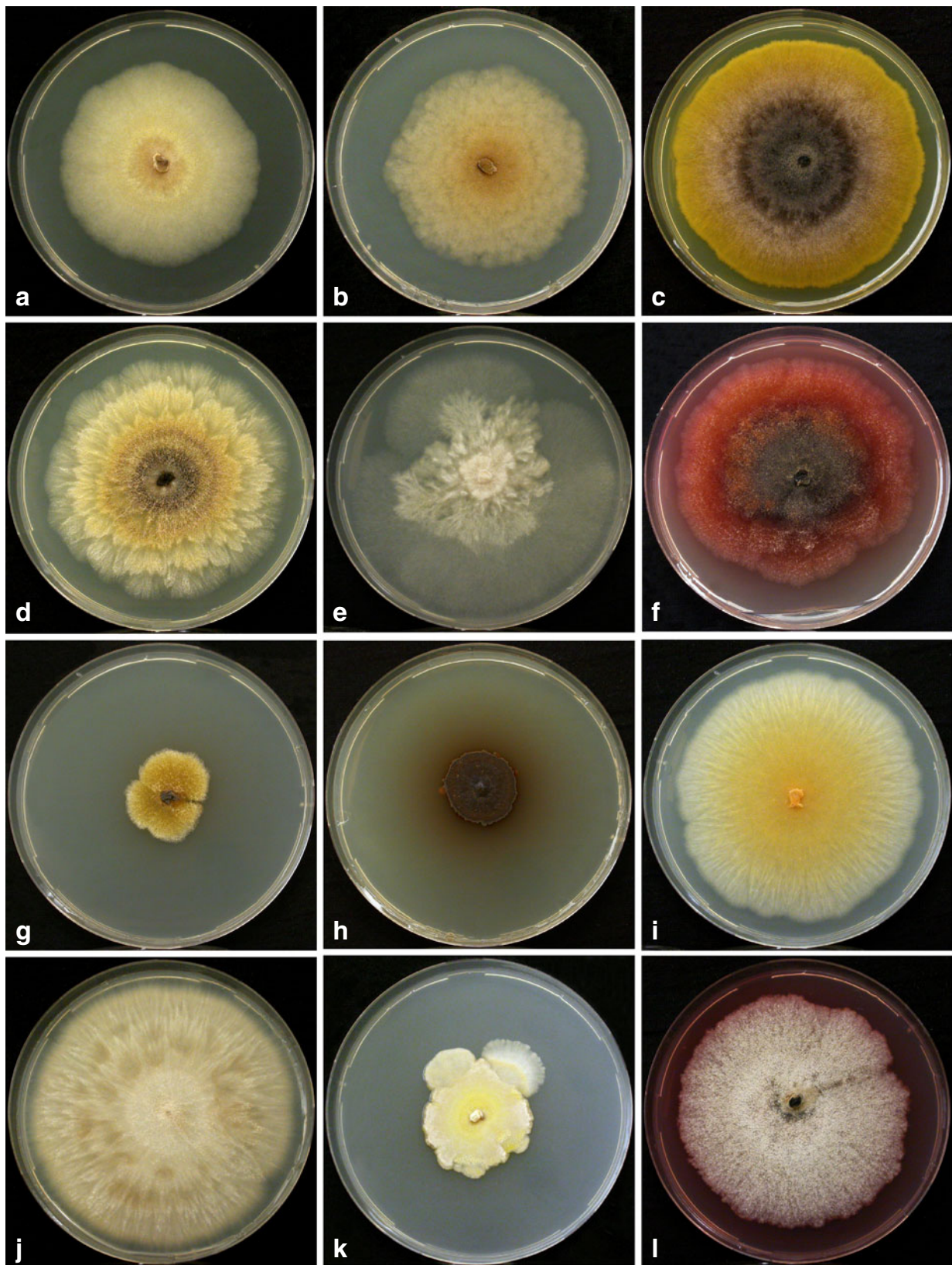


Fig. 3 *Melanconiella* cultures on MEA at room temperature, all after 20 d except where noted. **a.** *M. carpinicola* (MSMI). **b.** *M. chrysodiscosporina* (MGJ). **c.** *M. chrysmelanconium* (MEH, 28 d). **d.** *M. chrysoorientalis* (MGB). **e.** *M. decorahensis* (MED). **f.** *M. elegans*

(AR3830, 25 d). **g.** *M. flavovirens* (MFV3). **h.** *M. flavovirens* (MFA, 63 d). **i.** *M. hyperopta* (MCL). **j.** *M. hyperopta* var. *orientalis* (MHP, 31 d). **k.** *M. meridionalis* (MOM, 65 d). **l.** *M. spodiaea* (MSH, 28 d)

Table 2 List of *Melanconiella* species, their hosts and distribution. Taxa in bold represent new species, varieties or combinations

Host species	<i>Melanconiella</i> species	Distribution
<i>Betula</i> spp.	<i>M. decorahensis</i>	Europe, North America
<i>C. betulus</i>	<i>M. carpinicola</i> comb. nov.	Europe
<i>C. betulus</i>	<i>M. chrysodiscosporina</i> sp. nov.	Europe
<i>C. betulus</i>	<i>M. chryso-melanconium</i> sp. nov.	Europe
<i>C. betulus</i>	<i>M. hyperopta</i> comb. nov.	Europe
<i>C. betulus</i> , <i>C. orientalis</i>	<i>M. spodiaea</i>	Europe
<i>C. caroliniana</i>	<i>M. echinata</i> sp. nov.	North America
<i>C. caroliniana</i>	<i>M. elegans</i> sp. nov.	North America
<i>C. caroliniana</i>	<i>M. ellisii</i> comb. nov.	North America
<i>C. orientalis</i>	<i>M. chrysorientalis</i> sp. nov.	Europe
<i>C. orientalis</i>	<i>M. hyperopta</i> var. <i>orientalis</i> var. nov.	Europe
<i>Corylus avellana</i>	<i>M. flavovirens</i> comb. nov.	Europe, ?North America
<i>Ostrya carpinifolia</i>	<i>M. meridionalis</i> sp. nov.	Europe
<i>O. virginiana</i>	<i>M. ostryae</i> comb. nov.	Europe

rich groups for robust and reliable identification, and it is important to sequence also other genes (e.g. Gazis et al. 2011). ITS alone is definitely not sufficient to provide reliable characterisation of closely related species in many ascomycete lineages (e.g. Voglmayr & Jaklitsch 2008; Jaklitsch 2009, 2011; Pavlic et al. 2009; Druzhinina et al. 2010; Jaklitsch et al. 2012).

Biodiversity and nomenclature of *Melanconiella* on *Carpinus*

Until now, only two species of *Melanconiella* (usually as *Melanconis*) have been commonly recognised on *Carpinus*: the brown-spored *M. spodiaea* and the hyaline-spored *M. chrysostroma*. This was mainly influenced by Wehmeyer (1941), who was subsequently widely followed, e.g. by Ellis and Ellis (1997). Wehmeyer (1941) listed numerous synonyms for *M. chrysostroma*, however, he may not have examined all of the numerous authentic or type specimens he cited, as re-examination of several type specimens revealed that they are not even diaphragmated (see below). According to the spore size (14–23 × 3.5–7 μm) listed in Wehmeyer (1941), his concept of *M. chrysostroma* was apparently based on a mixture of *M. carpinicola* and *M. hyperopta*. Wehmeyer (1941) also established *Melanconis chrysostroma* var. *ellisii* for North American collections from *Carpinus*, which were considered to differ mainly by smaller ascospores.

Present investigations show that *M. chrysostroma* cannot be retained, because it is an obligate synonym of *Wuestneia xanthostroma* (see below). The molecular phylogenetic investigations show that specimens referred to as *M. chrysostroma* are highly heterogeneous, consisting of at least eight species. Three of these taxa (*M. carpinicola*, *M. ellisii*, *M. hyperopta*) have been described in the 19th century with accurate descriptions, but had been synonymised under *M. chrysostroma*.

Anamorphs

Our pure culture and DNA sequence data revealed the presence of a single, characteristic anamorph for each species of *Melanconiella*, which is either melanconium- or discosporina-like. This contrasts with Wehmeyer (1937, 1941), who recorded at least two different types of conidia (alpha and beta) for several species now placed in *Melanconiella*. In his pure culture studies of *M. decorahensis*, *M. ellisii* and *M. ostryae*, he reported the production of only a single anamorph, and his conclusions on alpha- and beta-conidia were mainly based on associations found in herbarium specimens. Knowing that several species of *Melanconiella* can co-occur on the same branch, it is not surprising that this observational evidence resulted in recording different conidial forms for one species.

Several of the anamorphs of *Melanconiella* have been described as species of *Melanconium*. According to Sutton (1980), more than 200 binomials have been described in *Melanconium*, and no generic revision is available. As many of the *Melanconium* binomials date back to the early 19th century, they cannot be interpreted based on the scanty original descriptions, which often indicate a mixture of discordant elements based on the hosts listed. Original material is often not extant, in bad condition, or unavailable for examination. In addition, the few morphological characters and the similarity of many *Melanconium* species are serious obstacles for appropriate interpretation. Most *Melanconium* names have not been used in the last decades. Therefore, clear preference should be given to the teleomorph epithets, which are mostly well interpretable, as well as to the respective teleomorph genera that were historically associated with *Melanconium*.

In addition to *Melanconium*, species of the anamorph genera *Discosporium* and *Discosporina* have been linked to teleomorphs now placed in *Melanconiella*. According to Sutton (1980), the teleomorph of the type species of *Discosporium*,

D. populeum (Sacc.) Sutton, is *Cryptodiaporthe populea* (Sacc.) Butin (now *Plagiostoma populinum* (Fuckel) L.C. Mejía; Mejía et al. 2011b). All *Discosporium* species attributable to *Melanconiella* have been transferred to *Discosporina* (Petra 1962); of the four species placed in *Discosporina*, three (*D. carpinicola* and *D. deplanata* from *Carpinus*; *D. sulphurea* from *Corylus*) are anamorphs of *Melanconiella*, but only *D. sulphurea* can be unequivocally attributed to a species, viz. *M. flavovirens*, the latter having priority.

Ecology and host range

Like many *Diaporthales* inhabiting woody plants, all species of *Melanconiella* are restricted to recently dead corticated branches of their hosts, often colonising extensive parts. This is indicative of weak pathogenicity or opportunistic growth, which appears to be common within the *Diaporthales*. As the *Diaporthales* have been shown to be common endophytes in broadleaf trees, often remaining dormant until host tissue senescence (Sieber 2007), species of *Melanconiella* may be widely present in healthy branches. This is supported by the rapid and extensive development of *Melanconiella* anamorphs commonly observed on recently cut branches.

All species of *Melanconiella* were observed to be highly host-specific as they were found to be confined mostly to a single, sometimes a few closely related hosts (Table 2). It is not surprising that we found a new species and variety on *Carpinus orientalis* (*M. chrysorientalis*, *M. hyperopta* var. *orientalis*) and a new species on *Ostrya carpinifolia* (*M. meridionalis*), hosts that are largely understudied. Whereas most host genera of *Melanconiella* harbour only a single or two species that do not co-occur in the same area, the situation is markedly different with *Carpinus*, which harbours the vast majority of *Melanconiella* biodiversity. On *Carpinus betulus*, several species of *Melanconiella* can co-occur in close vicinity, sometimes even on the same branches, which can pose problems for identification. However, there appears to be some ecological differentiation. Some species were only found on

branches still attached to the tree in open habitats (e.g. *M. carpinicola*, *M. chryso-melanconium*), whereas others mostly occurred on recently fallen or cut branches within forests (*M. hyperopta*, *M. chrysodiscosporina*), or were concentrated on branches still attached to the trees within forests (*M. spodiaea*). Some of the species also show different seasonality. While *M. hyperopta* has mainly been found in the winter season from late autumn to spring, *M. carpinicola*, *M. chrysodiscosporina* and *M. chryso-melanconium* were mostly found in the summer season from late spring to autumn. The reasons for this evolutionary radiation on a single host genus are unknown, but similar cases are known for other corticolous ascomycetes, e.g. *Prosthecium* (Voglmayr and Jaklitsch 2008) or *Massaria* (Voglmayr and Jaklitsch 2011), where biodiversity is concentrated on the host genus *Acer*. In those cases, co-occurrence of several related species on the same host has been observed, indicating that this may be a widespread phenomenon in weakly pathogenic corticolous ascomycetes.

Geographic distribution

Inhabiting hosts belonging to the temperate family *Betulaceae*, the genus *Melanconiella* is confined to the north temperate zone. Remarkably, the diversity of *Melanconiella* is highest in Europe with currently nine species and one variety, followed by North America with five confirmed species. The report of *M. spodiaea* on *Castanea crenata* from Japan by Kobayashi (1970) is likely based on a misidentification. The only additional Asian record referable to *Melanconiella* has been recently published by Vasilyeva and Stephenson (2010), who report *M. ellisii* (as *M. carpinigera*) from the Russian Far East. Considering that several taxa were included in *M. ellisii* until the current study, detailed morphological and DNA studies are necessary to reveal its true identity. There is little species overlap between Europe and North America; only *M. decora-hensis* has been confirmed for both continents in the present study, whereas conspecificity of North American and European accessions of *M. flavovirens* remains to be verified.

Taxonomy

Key to the genera *Melanconis* and *Melanconiella*

Ectostroma conspicuous, typically projecting up to 1 mm above the substrate, light coloured, white, cream or yellowish; conidia brown	<i>Melanconis</i>
Ectostroma inconspicuous, typically projecting up to 0.5 mm above the substrate, light or dark coloured; conidia hyaline or brown	<i>Melanconiella</i>

Melanconis Tul. & C. Tul., *Selecta fung carpol* (Paris) 2:115 (1863), emend.

Type: *Melanconis stilbostoma* (Fr.) Tul., *Ann Sci nat*, ser 4, 5:109 (1856).

Pseudostromata consisting of a well-developed, conspicuous, distinctly erumpent, light coloured, white or yellow ectostromatic disc causing a coarsely pustulate bark surface. *Ectostromatic disc* surrounded by bark or not. *Central*

column beneath the disc more or less conical, comprising hyaline hyphae mixed with a hyaline or light-coloured powdery amorphous substance; residual entostroma between perithecia of scant hyaline hyphae. *Stromatic zones* lacking. *Perithecia* often appearing as rounded bumps beneath the bark surface surrounding the ectostromatic disc, oblique or horizontal, often arranged in a circle around the central column, with long lateral ostioles that converge at the margin of the column and emerge at the margin or within the ectostromatic disc, flat discoid or slightly projecting and convex, typically not obscuring the disc. *Paraphyses* broad bands, deliquescent at maturity. *Asci* oblong or fusoid, octosporous, with an apical ring typically distinct when fresh; asci becoming detached from their base. *Ascospores* hyaline, ellipsoid, bicellular, with a central or slightly eccentric, not or slightly constricted septum, smooth, with or without blunt or pointed appendages.

Conidial state: melanconium-like. *Conidiomata* acervular; typically possessing the same type of ectostromatic disc and central column as the teleomorph, usually preceding it. *Conidiophores* branched only at the base, septate, few-celled, smooth, hyaline. *Conidiogenous cells* annellidic. *Conidia* brown, ellipsoid or subglobose.

Melanconiella Sacc., Syll Fung 1:741 (1882), emend.

Type: *Melanconiella spodiaea* (Tul.) Sacc., Syll Fung 1:741 (1882); **lectotype designated by** Müller and von Arx (1962).

Pseudostromata consisting of an inconspicuous, erumpent, light or more frequently dark coloured ectostromatic disc or pulvillus (referred to as ectostromatic disc below) causing a more or less pustulate bark surface. *Ectostromatic disc* convex, flat or concave, variously coloured, surrounded by bark or not. *Central column* beneath the disc more or less conical, comprising hyaline or pigmented hyphae mixed with a pigmented, most commonly cream, yellow, olive, brownish or gray, powdery amorphous substance; residual entostroma between perithecia mostly of scant hyphae. *Stromatic zones* lacking. *Perithecia* inconspicuous at the bark level or appearing as rounded bumps beneath the bark surface surrounding the ectostromatic disc, oblique or horizontal, scattered or often arranged in a circle around the central column, with long lateral ostioles that converge at the margin of the central column or within. *Ostioles* emerging in various positions in the ectostromatic disc, flat in the disc or slightly projecting, rarely distinctly projecting and cylindrical, often obscuring the disc. *Paraphyses* broad bands, deliquescent at maturity. *Asci* oblong or fusoid, octosporous, with a distinct apical ring, becoming detached from their base. *Ascospores* hyaline, yellowish or brown, fusoid or ellipsoid, bicellular, with a central or slightly eccentric, not or slightly constricted septum, smooth; wall

swelling in water mounts or not, with or without short, blunt appendages and sometimes with a narrow gelatinous sheath.

Conidial state: melanconium- or discosporina-like. *Conidiomata* acervular, peridermal or subperidermal; possessing the same type of ectostromatic disc and central column or lighter than that of the teleomorph, usually preceding it. *Conidiophores* branched and septate only at the base, few-celled, smooth, hyaline to light brown. *Conidiogenous cells* annellidic (in melanconium-like conidiation) or phialidic (in discosporina-like conidiation). *Conidia* brown or hyaline, ellipsoid, subglobose, ovoid or oblong, with or without distinct hyaline sheath.

Notes: In the original description of *Melanconiella*, Saccardo (1882) mentioned uniseptate ascospores becoming brown at maturity and he listed two species, *M. spodiaea* and *M. chrysostroma*, without selecting a generic type. Although Clements and Shear (1931) lectotypified the genus with *M. chrysostroma*, this species is in serious conflict with the generic protologue, as it is an obligate synonym of *Wuestneia xanthostroma* (see below), a taxon of the *Diaporthales* with one-celled, hyaline ascospores. The lectotypification of Clements and Shear (1931) is therefore superseded according to the ICBN Art. 9.17(b). We agree with Müller and von Arx (1962) who considered *M. spodiaea* to be the type species of *Melanconiella* and thus the lectotypification is attributed to them.

Anamorphs of *Melanconis* and *Melanconiella* are usually referred to as *Melanconium*. Although *Melanconium* Link, Mag. Gesell. naturf. Freunde, Berlin 3(1–2): 9 (1809) is older than the teleomorph genera, it is not used here. Its type species *M. atrum* is said to occur on *Fagus sylvatica* (Sutton 1980), but intense searches for many years has not yielded any *Melanconium* on *Fagus*. Re-examination of the lectotype of *M. atrum* (K(M) 171588) revealed conidia with a lighter brown equatorial zone and a size of 10–13×6–8 µm, which do not fit known anamorphs of *Melanconiella*. Conidial size and morphology of *M. atrum* is similar to the anamorph of *Melanconis alni*, which, however, does not occur on *Fagus*. Identity, systematic affiliation and phylogenetic position of *Melanconium atrum* are therefore obscure.

Melanconiella carpinicola (Fuckel) Voglmayr & Jaklitsch, comb. nov. (Fig. 4)

Mycobank MB 800113

Basionym: *Diaporthe carpinicola* Fuckel, Jb Nassau Ver Naturk 27–28:37 (1874) [1873–74].

Synonym: *Diaporthe kunzeana* Sacc., Nuovo Giorn Bot Ital 8:180 (1876) (type not seen).

Pseudostromata 0.8–1.7 mm diam, inconspicuous, slightly projecting or not, perithecia only rarely projecting; centre bluntly conical. *Ectostromatic disc* circular or oblong,

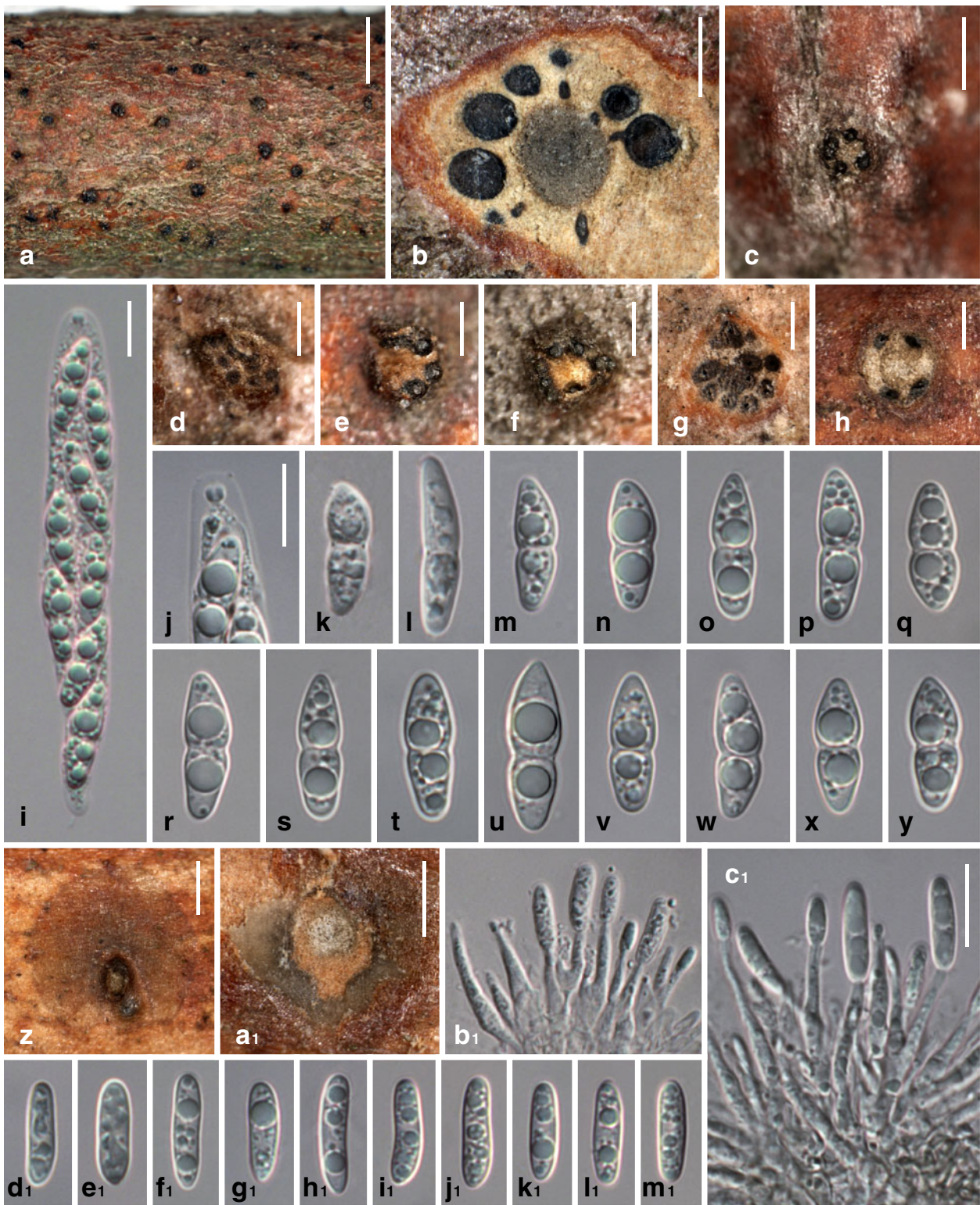


Fig. 4 *Melanconiella carpinicola*. **a**. Panoramic view of pseudostromata. **b**. Pseudostroma in transverse section showing central column and perithecia immersed in the entostroma. **c**. Pseudostroma showing perithecial bumps, ectostromatic disc and ostioles in surface view. **d–f**. Ectostromatic discs and ostioles in surface view. **g, h**. Transverse section below ectostromatic disc. **i**. Ascus. **j**. Ascus tip with apical ring. **k–y**. Ascospores. **z**. Conidioma in surface view. **a1**. Transverse

section of conidioma, showing central column. **b1, c1**. Conidiophores. **d1–m1**. Conidia. **k, l** in 3 % KOH, all other mounts in water. $a=2\text{ mm}$, $b, c, z, a1=500\text{ }\mu\text{m}$, $d-h=300\text{ }\mu\text{m}$, $i-y, b1-m1=10\text{ }\mu\text{m}$. Sources: **a, m**. WU 31806; **b, c, f, h**. WU 31802; **d, g, k, l**. G 00111395 (lectotype); **i, j, n–s**. WU 31801; **t–y, c1, f1, g1, h1**. WU 31805; **z–b1, il–m1**. WU 31807; **d1, e1**. M-0177882 (isotype)

cream, pale yellowish to dark brown or black, 0.15–0.6 mm long, sometimes concealed by densely arranged ostioles, often pulvinate; bark often darkened in a ring around the disc. *Central column* grey. *Entostroma* indistinct. *Ostioles* 1–10 per disc, 40–125 µm diam, plane or slightly papillate, black. *Perithecia* 0.3–0.5 mm diam, arranged in various configurations. *Asci* fusoid, (60–)67–82(–92)×8–11(–13) µm ($n=29$), containing 8 irregularly biseriolate ascospores, with apical ring distinct when fresh, 2.5–2.8 µm diam, 1.3–1.8 µm high, only faintly visible in older herbarium specimens. *Ascospores* hyaline, fusoid, distinctly constricted at the septum, (12–)15–18(–22)×(3.0–)3.8–5.3(–6.5), $l/w=(2.5–)3.1–4.3(–6.2)$ ($n=388$) µm; without appendages; cells dimorphic, upper cell mostly larger, with subacute end, lower cell narrowly rounded, multiguttulate containing mostly one large and numerous small guttules per cell; wall *ca.* 0.2–0.4 µm thick, not swelling.

Anamorph: discosporina-like. *Conidiomata* 1–2.5 mm diam, visible as darker spots, mostly with central or eccentric stromatic column; at maturity covered by whitish discharged conidial masses. *Conidiogenous cells* phialidic, 10–18×1.5–2.5 µm, hyaline. *Conidia* hyaline, elongate to sub-ellipsoid, (10–)12–14(–15.5)×(2.7–)3.2–3.7(–4.0) µm, $l/w=(2.8–)3.3–4.2(–5.1)$ ($n=115$), multiguttulate; wall *ca.* 0.2–0.4 µm, without gelatinous sheath.

Culture: Colony on MEA after 17 d at room temperature *ca.* 55 mm diam, without a distinct radial arrangement of surface hyphae, first white, turning dull yellow, eventually dark brown; aerial hyphae frequent only in the centre; diffusing pigment lacking.

Habitat and host range: dead corticated twigs and branches of *Carpinus betulus* attached to the tree; it has only been found in open sites like forest edges and hedges.

Distribution: Europe; uncommon but widely distributed.

Typification: GERMANY, Hessen, near Oestrich, on dry branches of *Carpinus betulus*, autumn, L. Fuckel, Fungi Rhenani 2660 (G 00111395, **lectotype of *Diaporthe carpinicola* here designated**; M-0177882, M-0177883 and WU: **isotypes**). AUSTRIA, Niederösterreich, Michelbach Markt, Mayerhöfen, map grid 7860/4, on corticated branches of *Carpinus betulus*, 18 June 2011, H. Voglmayr & W. Jaklitsch, strain MSMI (from teleomorph) (WU 31801, **epitype of *Diaporthe carpinicola* here designated**; *ex*-epitype culture CBS 131706), *ex*-epitype sequences JQ926235 (ITS-LSU), JQ926307 (*rpb2*), JQ926373 (*tef1*).

Additional specimens examined: AUSTRIA, Kärnten, St. Margareten im Rosental, Kramerstrauch, map grid 9452/4, 12 May 2010, H. Voglmayr, strain MNM (from teleomorph) (WU 31802). *ibid.*, 6 July 2002, W. Jaklitsch, strain A.R. 3883 (from teleomorph) (BPI 841338). *ibid.*, Gupf, near Brici (Writze), 6 July 2006, W. Jaklitsch, W.J. 2924 (WU 31803). *ibid.*, Triebblach, near Ledra, soc. *M. chrysomelanconium*, 7 June 1996, W. Jaklitsch, W.J. 891 (WU 31804). Oberösterreich,

Raab, map grid 7647/2, soc. *M. chrysomelanconium*, 1 Aug. 2010, H. Voglmayr, strain MSR (from teleomorph) (WU 31805). GERMANY, Sachsen-Anhalt, Eisleben, Oct. 1875, G. Winter, Thümen, Mycoth. Univ. 469 (WU). SLOVENIA, Gorenjska, Povodje, 16 May 2010, H. Voglmayr & W. Jaklitsch, strain MNP (from teleomorph) (WU 31806). SWEDEN, Skåne, Äsphult par., Forshult, 8 July 1978, K. & L. Holm (UPS 412042a, as *Melanconis chrysostroma*). UNITED KINGDOM, West Yorkshire, Wakefield, Newmillerdam N Barnsley, Newmillerdam Country Park, soc. *M. chrysomelanconium*, 17 May 2011, H. Voglmayr & W. Jaklitsch, strains MNUK (from teleomorph), MENU (from anamorph) (WU 31807, culture CBS 131700).

Notes: *Melanconiella carpinicola* is well characterised by protruding and narrowly aggregated ostioles mostly at the margin of a small inconspicuous ectostromatic disc, which, at low magnification, gives them a typical appearance of distinct, small, elevated, black bumps on the twigs. In addition, its small ascospores without swelling walls in combination with its growth on *Carpinus betulus* are distinctive. Ecologically, this species is apparently confined to open habitats, where it is commonly associated with *M. chrysomelanconium* even on the same twigs. When co-occurring, the areas of both species can be easily distinguished by their abundant conidiomata which produce hyaline conidia in *M. carpinicola* and dark brown conidia in *M. chrysomelanconium*. The description of conidia by Fuckel (1874) is in agreement with our data; the anamorph-teleomorph connection was proven in the present study by DNA data from conidial and ascospore cultures.

Considering the preservation and abundance of the collection, the part of Fuckel's original collection of Herbarium Barbey-Boissier housed in G is designated as lectotype here. To ensure nomenclatural stability, a recent collection, for which a culture and sequences are available, is designated here as epitype. Although no type specimen could be located and investigated, *Diaporthe kunzeana* is clearly a synonym of *M. carpinicola* according to the general description, particularly the ascospore size (16–19×3.5–4 µm) and host given in Saccardo (1876).

Melanconiella chryso-discosporina Voglmayr & Jaklitsch, sp. nov. (Fig. 5)

Mycobank MB 800114

Etymology: referring to the yellow stroma and the discosporina-like anamorph.

Pseudostromata typically indistinct, ill-defined, confluent, irregular, sometimes with circular outline, 1.5–3.4 mm diam, typically projecting by less than 0.5 mm; perithecial bumps sometimes distinct. *Ectostromatic disc* minute, 0.2–0.6(–0.8) mm long, circular when young, then usually



Fig. 5 *Melanconiella chrysodiscosporina*. **a**. Panoramic view of pseudostromata. **b**. Pseudostroma in transverse section. **c**. Three pseudostromata in surface view. **d–g**. Ectostromatic discs and ostioles in surface view. **h**. Transverse section below ectostromatic disc. **i**. Ascus. **j**. Ascus tip with apical ring. **k–w**. Ascospores (k, w showing

appendages). **x**. Conidioma in surface view. **y**. Transverse section of conidioma, showing central column. **z**, **a1**. Conidiophores. **b1–h1**. Conidia. All mounts in water. $a=2\text{ mm}$, $b, c, x, y=500\ \mu\text{m}$, $d-h=300\ \mu\text{m}$, $i-w, z-h1=10\ \mu\text{m}$. Sources: a, e. WU 31861; b–d, x, y, b1–f1. WU 31867; f, g. WU 31864; h, z, a1. WU 31862; g1, h1. WU 31824

narrowly fusoid-oblong-linear, often filled by ostioles, light or bright yellow or greyish to shades of brown. *Central column* yellow, greyish yellow, yellow-olive or (light

olive. *Entostroma* crumbly, of subhyaline to yellowish hyphae. *Ostioles* 1–8(–10) per disc, 50–160 μm diam, variously arranged in the disc, black, short papillate,

apically rounded or flat, sometimes acute with tips 15–30 μm wide, only slightly projecting above the disc level. *Perithecia* 0.3–0.8 mm. *Asci* cylindrical to fusoid, (90–)100–119(–127) \times (10–)11–15(–18) μm ($n=39$), containing 8 uni- or irregularly biserial ascospores, with distinct apical ring, 3.0–3.8 μm diam, 2.0–3.3 μm high. *Ascospores* hyaline, broadly ellipsoid, not constricted at the septum, (15.5–)17–20(–22) \times (6.2–)7.2–10.0(–14.5) μm , $l/w=(1.4–)1.9–2.4(–2.9)$ ($n=276$); ends broadly rounded to truncate, with hyaline cap-like appendages 1–2.5 μm long, 2.2–5.2 μm wide, fading and eventually absent in microscope mounts; cells monomorphic, multi-guttulate with one large and numerous small guttules per cell; wall distinctly swelling and sometimes distinctly bulging particularly at the septum, 0.6–1.4 μm thick, becoming *ca.* 1.2–4.7 μm thick at the septum.

Anamorph: discosporina-like. *Conidiomata* 0.7–4.7 mm diam, visible as often lobed, darker spots lined by a distinct dark brown to blackish marginal zone, with a central or eccentric stromatic column; at maturity covered by whitish discharged conidial masses. *Conidiogenous cells* phialidic, 18–32 \times 2.5–4.5 μm , hyaline. *Conidia* hyaline, ellipsoid, oblong or cylindrical, (12–)13–15.5(–19) \times (4.3–)4.8–5.5(–6.5) μm , $l/w=(2.1–)2.4–3.2(–4)$ ($n=227$), with two large and numerous small guttules; wall *ca.* 0.4 μm thick, with distinct gelatinous sheath *ca.* 0.6–0.8 μm thick.

Culture: Colony on MEA after 17 d at room temperature *ca.* 56 mm diam, not zonate, surface sometimes appearing mottled due to varying mycelial density, aerial hyphae irregularly arranged, abundant only in the centre or causing a finely floccose surface; first whitish or pale yellowish, turning brown from the centre, margin turning dull yellow-brown or black; diffusing pigment lacking; odour indistinct.

Habitat and host range: dead corticated twigs and branches of *Carpinus betulus* mostly lying on the ground.

Distribution: confined to Europe where it is common.

Type: AUSTRIA, Niederösterreich, Michelbach Markt, Mayerhöfen, map grid 7860/4, on corticated branches of *Carpinus betulus*, 20 June 2009, W. Jaklitsch & H. Voglmayr, strain MCH (from teleomorph) (WU 31859, **holotype**; *ex*-holotype culture CBS 125597), *ex*-holotype sequences JQ926238 (ITS-LSU), JQ926310 (*rpb2*), JQ926376 (*tefl*).

Additional specimens examined (all on *Carpinus betulus*): AUSTRIA, Kärnten, St. Margareten im Rosental, map grid 9452/4, 23 Sep. 2010, H. Voglmayr & W. Jaklitsch (WU 31860). Niederösterreich, Gießhübl, Seewiese, map grid 7863/3, 6 June 2010, H. Voglmayr, strain MGG (from teleomorph) (WU 31861). Gießhübl, Wassergspreng, map grid 7963/1, 15 Aug. 2010, H. Voglmayr (WU 31862). Hardegg, Merkersdorf, Semmelfeld, map grid 7161/3, 8 Aug. 2010, W. Jaklitsch & H. Voglmayr (WU 31863). Mannersdorf am Leithagebirge, Schweingraben, map grid 8065/2, soc. *M. hyperopta* (teleomorph), 17 April 2010, H.

Voglmayr & I. Greilhuber, strain MCA_n (from anamorph) (WU 31824). Michelbach Markt, Mayerhöfen, map grid 7860/4, soc. *M. chryso melanconium*, 20 June 2009, W. Jaklitsch & H. Voglmayr, strain MCH (WU 31864). *ibid.*, 3 June 2000, W. Jaklitsch, W.J. 1464 (BPI 747940, WU 31872). Mödling, Hinterbrühl, map grid 7963/2, 20 June 2010, H. Voglmayr (WU 31865). Oberösterreich, Raab, Riedlhof, map grid 7647/2, 4 April 2010, H. Voglmayr, strain MR_i (from teleomorph) (WU 31866). Steegen, Rittberg, map grid 7648/3, 28 June 2010, H. Voglmayr, strain MEE (from anamorph) (WU 31867). Wien, Döbling, Hermannskogel, map grid 7763/2, 18 April 2010, H. Voglmayr, strains MH₁, MH₂ (from teleomorph), MCHA (from anamorph) (WU 31868). Favoriten, Laaerbergstraße, map grid 7864/3, 8 April 2010, W. Jaklitsch & H. Voglmayr, strain MEL (from teleomorph) (WU 31869). Floridsdorf, Jedlersdorferstraße/Marchfeldkanalweg, map grid 7764/1, soc. *M. chryso melanconium*, 9 May 2010, W. Jaklitsch, strain MGJ (from teleomorph) (WU 31870, culture CBS 131703). Liesing, Maurerwald, map grid 7863/1, soc. *M. chryso melanconium*, 10 July 1993, W. Jaklitsch (WU 15980). CZECH REPUBLIC, Moravia, Lednice, palace grounds, 1 Oct. 2010, W. Jaklitsch & H. Voglmayr (WU 31871). SWEDEN, Skåne, Äsphult par., Forshult, 8 July 1978, K. & L. Holm (UPS 412042b, as *Melanconis chrysostroma*).

Notes: *Melanconiella chryso discosporina* is similar to the two closely related species *M. chryso melanconium* and *M. chryso orientalis*, differing mainly in ITS, *rpb2* and *tefl* sequences; in addition *M. chryso melanconium* has dark brown conidia and slightly larger ascospores than those of *M. chryso discosporina* and *M. chryso orientalis* occurs on a different host, *Carpinus orientalis*. *Melanconiella chryso discosporina* is most commonly found on recently cut branches lying on the ground, where it is often associated with *M. hyperopta*. As *M. chryso discosporina* has sometimes been found to co-occur on the same branches with the highly similar *M. chryso melanconium*, reliable species identification is only possible on anamorph traits. The anamorph-teleomorph connection was repeatedly proven by DNA data from conidial and ascospore cultures.

Melanconiella chryso melanconium Voglmayr & Jaklitsch, sp. nov. (Fig. 6)

Mycobank MB 800115

Etymology: referring to the yellow stroma and the melanconium-like anamorph.

Pseudostromata 1.5–2.6 mm diam, more or less circular, typically prominent, usually causing distinct bumps in the bark, but often densely aggregated or confluent, projecting to *ca.* 0.5 mm; individual perithecial contours causing small bumps in the bark. *Ectostromatic disc* small, 0.2–0.7(–0.9)



Fig. 6 *Melanconiella chrysolanconium*. **a.** Panoramic view of pseudostromata. **b.** Pseudostroma in transverse section. **c.** Pseudostroma showing ectostromatic disc and ostioles in surface view. **d–g.** Ectostromatic discs and ostioles in surface view. **h.** Transverse section below ectostromatic disc. **i.** Ascus. **j.** Ascus tip with apical ring. **k–u.** Ascospores (k–n, s, u showing appendages). **v.** Conidioma in surface

view. **w.** Transverse section of conidioma (left), showing central column, and of young stroma and perithecium (right). **x.** Conidiophores. **y–c1.** Conidia. All mounts in water. a=2 mm, b, c, v, w=500 μ m, d–h=300 μ m, i–u, x–c1=10 μ m. Sources: a, d, e, j. WU 31878; b, i, p–s. WU 31874; c, x–c1. WU 31879; f–h, o, w. WU 31875; k, l. WU 31873; m, n. WU 31876; t, u. WU 15180; v. WU 31877

mm, usually fusoid or oblong, sometimes circular, yellow, yellow-brown, greyish yellow or light to dark brown. *Central column* often crumbly, bright yellow, grey, brown, olive. *Entostroma* inconspicuous or crumbly and olive. *Ostioles* 1–10 (commonly up to 6) per disc, 50–160 μm diam at their base, ca. 30–65 μm at their tips, variably located in the disc, papillate or with flat apex, brown or shiny black. *Perithecia* 0.4–0.8 mm diam; connection to the central column often difficult to ascertain. *Asci* cylindrical, (93–)109–128(–133) \times (12–)14–19(–22) μm ($n=22$), containing 8 uni- or irregularly biseriolate ascospores, with distinct apical ring, 3.5–4.5 μm diam, 2.4–3 μm high. *Ascospores* hyaline, broadly ellipsoid, not constricted at the septum, (16–)19–23(–27.5) \times (7.5–)9.0–12.3(–15.5) μm , $l/w=(1.1–)1.8–2.2(–2.6)$ ($n=307$); ends broadly rounded to truncate, with hyaline cap-like appendages 1.7–3 μm long, 3–5.4 μm wide, fading and disappearing in microscope mounts; cells monomorphic, multiguttulate with one large and numerous small guttules per cell; wall 0.6–2.6 μm thick, distinctly swelling particularly at the septum, sometimes distinctly bulging out becoming 1.5–4.8 μm thick at the septum.

Anamorph: melanconium-like. *Conidiomata* 0.6–4 mm diam, visible as dark brown to blackish spots, with central or eccentric stromatic column; at maturity covered by black discharged conidial masses. *Conidiogenous cells* annellidic, 18–40 \times 3–5 μm , hyaline. *Conidia* dark brown, broadly ellipsoid to broadly pip-shaped, (12.5–)13–15(–16) \times (8.5–)8.8–9.5(–10.0) μm , $l/w=(1.3–)1.4–1.6(–1.8)$ ($n=98$), multiguttulate with 1–2 large and numerous small guttules; wall ca. 0.6–0.8 μm thick, with distinct gelatinous sheath 0.8–1.1(–1.4) μm thick.

Culture: Colony on MEA after 17 d at room temperature ca. 56 mm diam, after 4 wk plate nearly entirely covered by mycelium. Colony circular, with diffuse concentric zones; aerial hyphae short, only frequent or abundant in the centre, forming diffuse, transparent whitish zones, below these zones colony pale yellowish-greenish, turning dark brown, grey to nearly black from the centre; margin dull golden yellow to olive; odour slightly sour; diffusing pigment lacking, agar faintly yellowish.

Habitat and host range: dead corticated twigs and branches of *Carpinus betulus* attached to the tree; it has only been found in open sites like forest edges, parks and hedges.

Distribution: Europe; uncommon but widely distributed.

Type: AUSTRIA, Wien, Floridsdorf, Marchfeldkanalweg, map grid 7764/1, on corticated branches of *Carpinus betulus*, 9 Nov. 2008, W. Jaklitsch, strain MCM (from teleomorph) (WU 31873, **holotype**; ex-holotype culture CBS 124271), ex-holotype sequences JQ926247 (ITS-LSU), JQ926319 (*rpb2*), JQ926385 (*tef1*).

Additional specimens examined (all on *Carpinus betulus*): AUSTRIA, Kärnten, St. Margareten im Rosental, Triebblach, near Ledra, map grid 9452/4, soc. *M. carpinicola*, 7 June 1996, W. Jaklitsch, W.J. 891 (WU 31804). Niederösterreich,

Michelbach Markt, Mayerhöfen, map grid 7860/4, soc. *M. chrysodiscosporina*, 20 June 2009, W. Jaklitsch & H. Voglmayr (WU 31864). Oberösterreich, Raab, map grid 7647/2, soc. *M. carpinicola*, 1 Aug. 2010, H. Voglmayr, strain MGR (from teleomorph) (WU 31877). Wien, Favoriten, Laaerbergstraße/Fontanastraße, map grid 7864/3, 30 May 1993, W. Jaklitsch (WU 15180). Floridsdorf, Lazarsfeldgasse, map grid 7764/2, 23 May 2010, W. Jaklitsch, strain MGL (from teleomorph) (WU 31874). Floridsdorf, Jedlersdorferstraße, map grid 7764/1, soc. *M. chrysodiscosporina*, 9 May 2010, W. Jaklitsch (WU 31870). Landstraße, Botanical Garden, map grid 7864/1, 23 May 2010, H. Voglmayr, strains MGH (from teleomorph), MEH (from anamorph) (WU 31875, culture CBS 131699). Landstraße, Stadtpark, map grid 7764/3, 3 Sep. 2010, H. Voglmayr & W. Jaklitsch, strain MGSt (from teleomorph) (WU 31876). Liesing, Maurerwald, map grid 7863/1, soc. *M. chrysodiscosporina*, 10 July 1993, W. Jaklitsch (WU 15980). SPAIN, Asturias, Lueca, finca 'Fonte Baixa', 8 May 2010, E. Rubio Domínguez, E.R.D. 5108, strain MGS (from teleomorph) (WU 31878). UNITED KINGDOM, West Yorkshire, Wakefield, Newmillerdam N Barnsley, Newmillerdam Country Park, soc. *M. carpinicola*, 17 May 2011, H. Voglmayr & W. Jaklitsch, strains MGUK (from teleomorph), MEUK (from anamorph) (WU 31879, culture CBS 131701).

Notes: *Melanconiella chrysomelanconium* is similar to the closely related *M. chrysodiscosporina* and *M. chrysorientalis*, from which it differs mainly by dark brown conidia, slightly larger ascospores and ITS, *rpb2* and *tef1* sequences; *M. chrysorientalis* differs also by the host (*Carpinus orientalis*). Although *M. chrysodiscosporina* and *M. chrysomelanconium* share the same host, they appear to be ecologically separated: *Melanconiella chrysodiscosporina* has been mainly collected on fallen or cut branches within forests, whereas *M. chrysomelanconium* is apparently confined to open habitats, where it is commonly associated with *M. carpinicola* on twigs attached to the tree. However, *M. chrysomelanconium* has sometimes also been found to co-occur on the same branches with *M. chrysodiscosporina* at the edges of larger forests. Therefore, reliable identification is only possible with the anamorph. The anamorph-teleomorph connection was repeatedly proven by DNA data from conidial and ascospore cultures. The teleomorph of the Spanish collection is beautifully illustrated with macro- and microphotographs at http://www.ascofrance.com/search_recolte/2696 or <http://www.asturnatura.com/fotografia/setas-hongos/melanconis-chrysostroma-fr-tul-c-tul-3/9039.html>.

Melanconiella chrysorientalis Voglmayr & Jaklitsch, sp. nov. (Fig. 7)

Mycobank MB 800116

Etymology: referring to the yellow stroma and its host, *Carpinus orientalis*.



Fig. 7 *Melanconiella chrysorientalis*. **a**. Panoramic view of pseudostromata. **b**. Pseudostroma in transverse section. **c**. Three pseudostromata in surface view. **d–g**. Ectostromatic discs and ostioles in surface view. **h**. Transverse section below ectostromatic disc. **i**. Ascus. **j**. Ascus tip with apical ring. **k–y**. Ascospores (k–m showing appendages). **z**.

Conidioma in surface view. **a1**. Transverse section of conidioma, showing central column. **b1**, **c1**. Conidiophores. **d1–m1**. Conidia. All mounts in water. a=2 mm, b, c, z, a1=500 μ m, d–h=300 μ m, i–y, b1–m1=10 μ m. Sources: a, b, e–h, k–m, z–m1. WU 31858; c, d. WU 31880; i, j, n–y. WU 31881

Pseudostromata inconspicuous, 0.9–2.4 mm diam, slightly (0.1–0.3 mm) projecting, circular or contours ill-defined, merging; perithecial contours often evident. *Ectostromatic discs* 0.1–0.6 mm, often densely disposed, with oblong, elliptic or circular outline, yellowish, light yellow, dull yellow, dark yellow-brown or grey. *Central column* crumbly, dull yellow, greenish yellow or olive; *entostroma* of light or olive-green hyphae. *Ostioles* 1–10 per disc, 50–140 µm diam, shiny black or brown with black centre, convex-papillate, scarcely projecting. *Perithecia* 0.3–0.6 mm diam; connection to central column often unclear. *Asci* cylindrical, (104–)114–130(–137)×(11–)13–15(–17) µm ($n=33$), containing 8 uni-, rarely biseriata ascospores, with distinct apical ring, 3.8–4.3 µm diam, 2.5–3.2 µm high. *Ascospores* broadly ellipsoid, not constricted at the septum, (15.7–)17–19(–22)×(6.0–)7.5–9.3(–10.2) µm, $l/w=(1.7–)1.9–2.4(–3)$ ($n=111$); ends broadly rounded to sometimes truncate, with hyaline cap-like appendages 1.3–2 µm long, 2.4–4.2 µm wide, fading and eventually absent in microscope mounts; cells monomorphic, multiguttulate, each with one large and numerous small guttules; wall distinctly swelling and sometimes distinctly bulging out particularly at the septum, 0.6–1.1 µm thick, 1.3–3 µm at the septum.

Anamorph: discosporina-like. *Conidiomata* 1–2 mm diam, only scarcely visible in face view, with diffuse margins, with central or eccentric stromatic column; at maturity covered by discharged whitish to greyish conidial masses. *Conidiogenous cells* phialidic, 9–18×2.5–4 µm, hyaline. *Conidia* hyaline, ellipsoid, oblong or cylindrical, (11.7–)12.3–14.0(–15.3)×(5.5–)6.0–6.8(–7.3) µm, $l/w=(1.7–)1.9–2.2(–2.7)$ ($n=79$), with 1–2 large and numerous small guttules; wall ca. 0.5 µm thick, with distinct gelatinous sheath ca. 0.6–0.9 µm thick.

Culture: Colony on MEA after 17 d at room temperature ca. 61 mm diam, after 4 wk mycelium (nearly) covering the entire plate. Colony forming several concentric zones, each covered by a conspicuous, whitish, stellate mat of aerial hyphae; brown zones progressing from the centre, eventually dark brown below the whitish structure; agar pale yellow to brownish; odour sour.

Habitat and host range: dead corticated twigs and branches of *Carpinus orientalis*; only found on branches attached to the trees.

Distribution: South-eastern Europe, uncommon; collected in Croatia and Greece (Corfu) but likely to co-occur with its host in other countries.

Type: CROATIA, Istria, Barbariga, on corticated branches of *Carpinus orientalis*, 15 May 2010, H. Voglmayr & W. Jaklitsch, strain MGB (from teleomorph) (WU 31880, **holotype**); *ex-holotype* culture CBS 131702, *ex-holotype* sequences JQ926256 (ITS-LSU), JQ926328 (*rpb2*), JQ926394 (*tefl*).

Additional specimens examined (all on *Carpinus orientalis*): CROATIA, Istria, Poreč, Gedici, 1 Nov. 2010, H.

Voglmayr & I. Greilhuber, strain MGP (from teleomorph) (WU 31881). Vrsar, soc. *M. spodiaea* (anamorph), 14 May 2010, H. Voglmayr & W. Jaklitsch, strains MG V (from teleomorph), MVH (from anamorph) (WU 31858). GREECE, Kerkyra (Corfu), W Analipsis, 24 April 2012, H. Voglmayr & W. Jaklitsch (WU 32000). SE Ano Korakiana, 24 April 2012, H. Voglmayr & W. Jaklitsch (WU 32001). S Kouramaditika, 22 April 2012, H. Voglmayr & W. Jaklitsch (WU 32002).

Notes: *Melanconiella chrysorientalis* is similar to the closely related *M. chrysodiscosporina* and *M. chryso-melanconium*, from which it differs mainly by ITS, *rpb2* and *tefl* sequences and a different host (*C. orientalis*) and from *M. chryso-melanconium* by hyaline conidia. The anamorph-teleomorph connection was proven by DNA data from conidial and ascospore cultures.

Melanconiella decorahensis (Ellis) Sacc., Syll Fung 2: Addenda Vol I:LIV (1883). (Fig. 8)

Mycobank MB 210729

Basionym: *Melanconis decorahensis* Ellis, [as '*decoraensis*'], Am Nat 17:195 (1883).

Synonyms: *Melanconis decorahensis* var. *subviridis* Ellis & Everh., North American Pyrenomycetes 528 (1892) (type not seen).

Melanconiella subviridis (Ellis) Dearn. & House, Bull NY State Mus 266:81 (1925).

Pseudostromata 1.5–3 mm diam, circular, often inconspicuous, with their centre typically projecting to ca. 0.6, rarely 0.8 mm; perithecial contours only rarely evident. *Ectostromatic discs* variable, mostly narrow, fusoid or linear, 0.2–1.6 mm long, pale yellow, bright yellow, greenish yellow, greenish grey, olive or dull greyish brown. *Central column* grey, greenish grey or olive-brown. *Entostroma* inconspicuous. *Ostioles* variously arranged in the disc, 1–20 per disc, 40–140(–160) µm diam, shiny black, flat or papillate, tips typically flattened. *Perithecia* 0.3–0.5 mm diam. *Asci* narrowly cylindrical, (114–)125–145(–150)×(11–)12–14(–16) µm ($n=35$), containing 8 uniseriate ascospores, with an apical ring distinct when fresh, 3.7–4.2 µm diam, 3.7–4 µm high, often indistinct in older herbarium specimens. *Ascospores* dark brown, ellipsoid, distinctly constricted at the septum, (13.5–)16–20(–22)×(7.5–)8.5–10(–11) µm, $l/w=(1.6–)1.8–2.2(–2.4)$ ($n=57$); ends broadly rounded, without appendages; cells more or less monomorphic, multiguttulate; wall not swelling, ca. 0.7–0.8 µm.

Anamorph: melanconium-like. *Conidiomata* 1.5–2.5 mm diam, blackish brown, at maturity covered by black discharged conidial masses, forming conspicuous thick black pustules or spots 1–3 mm diam, sometimes confluent to 1–2 cm. *Conidiogenous cells* distinctly annellidic, 20–35×3.5–4 µm,



Fig. 8 *Melanconiella decorahensis*. **a.** Panoramic view of pseudostromata. **b.** Pseudostroma in transverse section. **c.** Pseudostroma in surface view. **d–f.** Ectostromatic discs and ostioles in surface view. **g–i.** Transverse sections below ectostromatic disc. **j.** Ascus. **k.** Ascus tip with apical ring. **l–x.** Ascospores. **y.** Conidioma in surface view. **z, a1.**

Conidiophores. **b1–i1.** Conidia. **v–x** in 3 % KOH, all other mounts in water. **a**=2 mm, **b–g, y**=500 μ m, **h, i**=200 μ m, **j–x, z–i1**=10 μ m. Sources: **a–c, f–h, j–u, y–e1.** WU 31808; **i.** Wehmeyer 3644 (NY); **v–x, f1–i1.** Ellis & Everhart, N. Amer. Fungi. Ser. II: 1562 (NY)

at first hyaline, becoming brown with age. *Conidia* dark brown, pip-shaped, (13–)15–17(–19)×(8–)9–10(–11) μm , $l/w=(1.2–)1.5–2(–2.3)$ ($n=121$), multiguttulate when fresh, with a large guttule when dead; wall *ca.* 0.6–0.7 μm thick, with hyaline gelatinous sheath *ca.* 0.5 μm thick.

Culture: Colony on MEA after 17 d at room temperature *ca.* 67 mm diam, whitish, outline irregular, covering the plate in 3–4 wk, aerial hyphae nearly lacking at the margin, but dense in central areas, forming more or less radially arranged spots; aged cultures only pale yellowish brown; diffusing pigment lacking.

Habitat and host range: dead corticated twigs and branches of *Betula* spp.

Distribution: common in North America (USA, Canada); rare in Europe (France). The French collection investigated in the present study is the first verified record from Europe. Wehmeyer (1941) mentioned a European collection deposited in PC without providing further details, but considered this record to be dubious due to the similarity of *M. decorahensis* to *M. spodiaea*, where ascospore appendages may not be seen in old material.

Typification: USA, Iowa, Decorah, on corticated branches of *Betula* sp., Aug. 1882, E. W. D. Holway (NY 922009, **lectotype of *Melanconis decorahensis* here designated**; NY 922008, **isotype**).

Additional selected specimens examined: FRANCE, Ariège, Rimont, on twigs of *Betula pendula*, 14 July 2010, J. Fournier, J.F. 10095, strains MD (from teleomorph), MED (from anamorph) (WU 31808, culture CBS 131698). USA, Alabama, Marion Co., Hamilton, Buttahatchee (“Ballahatchie”) River, on twigs of *Betula* sp., 14 July 1925, L. E. Wehmeyer 3644 (DAOM 121211, NY). Iowa, Decorah, on twigs of *Betula* sp., E. W. D. Holway, Ellis & Everhart, N. Amer. Fungi. Ser. II: 1562 (NY, 3 specimens). Massachusetts, Waverly, on twigs of *Betula* sp., 20 Sep. 1925, L. E. Wehmeyer 3806 (DAOM 121219, culture CBS 159.26). Indiana, Kankakee River & Route # 43, on twigs of *Betula lutea*, 3 Sep. 1938, L. E. Wehmeyer 3876 (DAOM 121187).

Notes: *Melanconiella decorahensis* is characterised by dark brown ascospores lacking appendages, pip-shaped dark brown conidia and growth on *Betula* spp. It is most similar to *M. spodiaea*, which mainly differs by the presence of cap-like ascospore appendages, differently shaped conidia and different hosts (*Carpinus* spp.). *Melanconis stilbostoma*, which is common on *Betula* spp., mainly differs by hyaline ascospores and whitish ectostromatic discs. The anamorph-teleomorph connection was proven by culture (Wehmeyer 1926) and DNA data.

We have not been able to study the type of *M. decorahensis* var. *subviridis* (= *M. subviridis*), but according to Wehmeyer (1941) it represents merely a well-developed specimen of *M. decorahensis*.

Melanconiella echinata Voglmayr & Jaklitsch, sp. nov. (Fig. 9)

Mycobank MB 800117

Etymology: referring to the ostioles projecting from the prominently erumpent ectostromatic discs, resulting in a spiny appearance.

Pseudostromata 1.2–2.5 mm diam, projecting 0.3–0.6(–0.8) mm, well-defined, distinct, circular, causing a ring-like elevation of the bark surrounding the ectostromatic disc; perithecial bumps indistinct. ***Ectostromatic disc*** 0.4–0.9(–1.3) mm diam, well-developed, distinctly projecting, often pulvinate, with circular, angular or ellipsoid outline, cream, yellow, honey, cinnamon, orange brown to brown. ***Central column*** bright olive-yellow when young, later olive-green to olive-brown. ***Entostroma*** well-developed, compact, of compacted hyphae, olive-green or olive-brown. ***Ostioles*** 1–25(–30) per disc, 90–190 μm diam, typically regularly arranged in the disc, cream to brown with shiny black tips, distinctly projecting, papillate or conical, tips flat or rounded. ***Perithecia*** 0.4–0.8 mm diam. ***Asci*** fusoid, (66–)72–81(–83)×(13–)14–16.5(–18) μm ($n=33$), containing 8 uni- or biseriolate ascospores, with a distinct apical ring, *ca.* 3 μm diam, 2 μm high. ***Ascospores*** hyaline, fusoid, slightly curved, slightly to distinctly constricted at the septum, (18–)20–22(–26)×(6.0–)6.5–7.5(–8.7) μm , $l/w=(2.5–)2.8–3.2(–3.7)$ ($n=121$); ends subacute to narrowly rounded, with hyaline cap-like appendages 1–1.5 μm long, 2–2.5 μm wide, soon fading in microscope mounts; cells slightly dimorphic, with four large guttules when fresh, mostly disappearing in herbarium specimens; wall *ca.* 0.4 μm thick, sometimes distinctly swelling at the septum.

Anamorph: not observed.

Habitat and host range: dead corticated twigs and branches of *Carpinus caroliniana*; probably rare.

Distribution: North America (USA).

Type: USA, New York, Ringwood, Lloyd-Cornell Preserve, on corticated branches of *Carpinus caroliniana*, 6 Sep. 1952, R. M. Horner (DAOM 121196, **holotype**).

Additional specimens examined: USA, Pennsylvania, West Chester, on *Carpinus caroliniana*, Aug. 1883, Haines & Everhart, issued as *Melanconis chrysostroma* in Ellis & Everhart, N. Amer. Fungi. Ser. II: 1563 (GZU).

Notes: As no recent collections have been available, only the ITS-LSU region could be obtained for *M. echinata*, and it could not be included in the combined phylogenetic analyses. The LSU demonstrates that the fungus belongs to *Melanconiella*, and that it is also distinct from all other species (Fig. 1). Morphologically it fits well in *Melanconiella*, except for its distinctly pustulate stromata, which have not been observed in any other species of the genus. Although otherwise being similar to *M. elegans*, *M. echinata* is distinct due to its differently shaped, larger

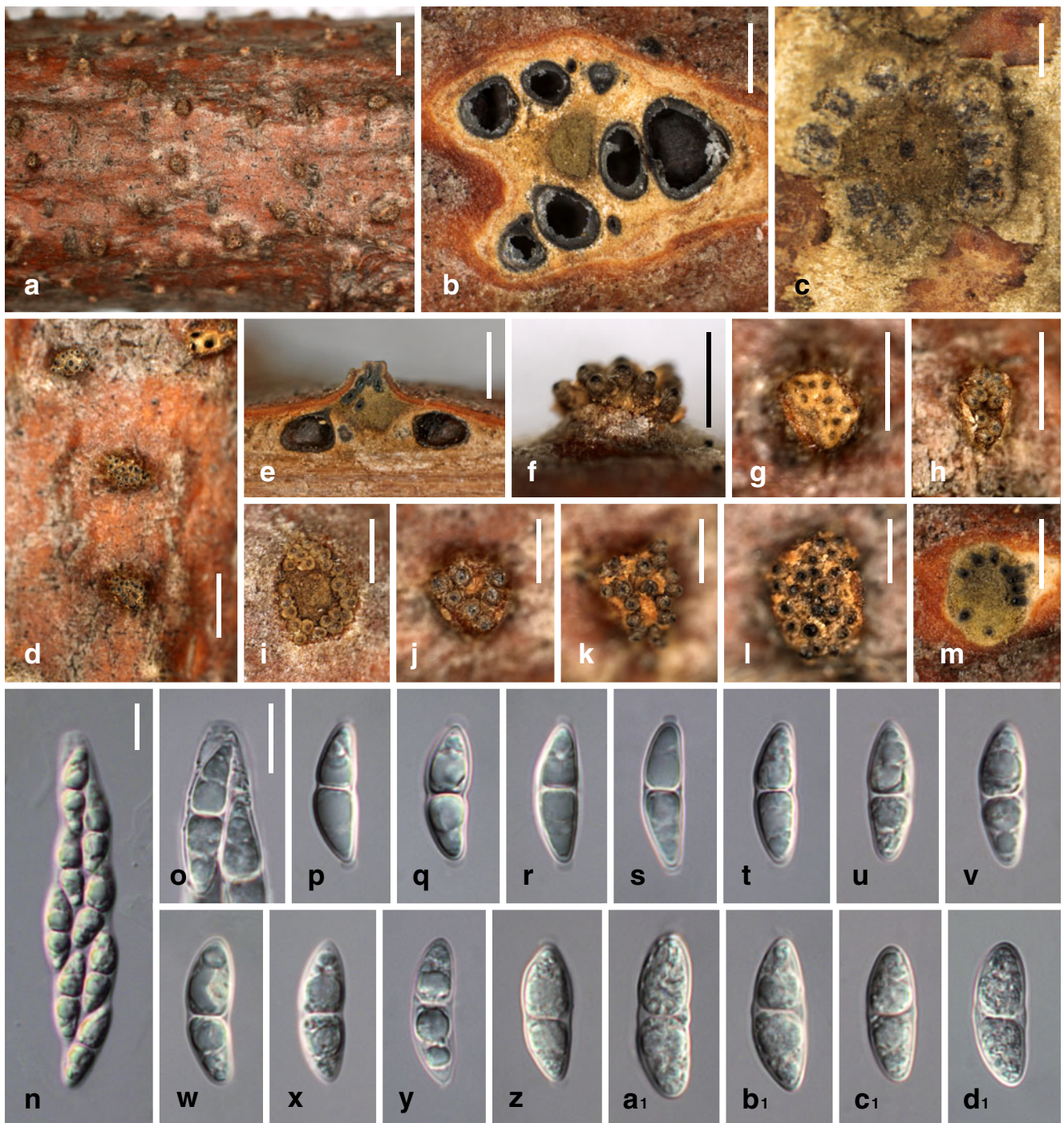


Fig. 9 *Melanconiella echinata*. **a.** Panoramic view of pseudostromata. **b.** Pseudostroma in transverse section. **c.** Groups of detached perithecia in well developed entostroma from below. **d.** Three pseudostromata in surface view. **e.** Vertical section of pseudostroma. **f.** Ectostromatic disc with projecting ostioles in side view. **g–i.** Young ectostromatic discs and ostioles in surface view. **j–l.** Fully developed ectostromatic discs

and ostioles in surface view. **m.** Transverse section below ectostromatic disc. **n.** Ascus. **o.** Ascus tip with apical ring. **p–d1.** Ascospores (p–s, z showing appendages). n–t in water, u–d1 in 3 % KOH. a=2 mm, b, c, e–m=500 μm, d=1 mm. Sources: a, b, d–y. DAOM 121196; c, z–d1. Ellis & Everhart, N. Amer. Fungi. Ser. II: 1563 (GZU)

ascospores, its prominent erumpent ectostromatic discs causing a coarsely pustulate bark surface similar to *Melanconis* and its distinctly projecting ostioles. In addition, the entostroma is well developed and of distinctly olive-

yellow to olive-green colour. Although the label of Ellis and Everhart, N. Amer. Fungi 1563 states that the specimens show no asci, fully mature perithecia were present in the specimen from GZU, having asci and ascospores fully

in agreement with the type. Interestingly, this exsiccatum was issued as *Melanconis chrysostroma*, and it was distributed in the same fascicle as the type collection of *Melanconiella ellisii* (N. Amer. Fungi 1567, issued as *Valsa (Diaporthe) ellisii*), which was collected at the same locality. This suggests that Ellis and Everhart considered these collections to represent distinct taxa. Ellis and Everhart (1892) based their description of *Melanconis chrysostroma* on the exsiccatum N. Amer. Fungi 1563, applying the name with some doubts. In the broad species concept of Wehmeyer (1941), the specimen was listed under *Melanconis chrysostroma* var. *ellisii*. We found no anamorph on the two collections examined.

Melanconiella elegans Voglmayr & Jaklitsch, sp. nov. (Fig. 10)

Mycobank MB 800118

Etymology: referring to the regular yellowish ectostromatic discs with evenly distributed ostioles, providing an elegant appearance.

Pseudostromata 1.2–3 mm diam, projecting to ca. 0.6(–0.8) mm, well-defined, distinct, circular, or confluent with densely disposed discs, causing small bumps in the bark; perithecial bumps often distinct. **Ectostromatic disc** 0.2–0.8 mm diam, rarely larger (1.6 mm), well-developed, distinctly projecting, often pulvinate, with typically circular or angular, sometimes ellipsoid, fusoid or oblong outline, pale or bright yellow to light brown, or concealed by ostioles. **Central column** yellow, olive, grey or brownish. **Entostroma** well-developed, compact, of compacted hyphae, dull yellow, olive or brownish. **Ostioles** 1–20 per disc, 30–140(–160) μm diam, typically evenly spaced in the disc, brown with shiny black tips, slightly projecting, papillate or conical, tips flat, rounded or nearly acute. **Perithecia** 0.2–0.6 mm diam. **Asci** fusoid, (49–)54–69(–77) \times (9–)10–13(–15) μm ($n=67$), containing 8 uni- or biserial ascospores, with distinct apical ring, 2.4–2.8 μm diam, 1.8–2.6 μm high. **Ascospores** hyaline, fusoid, slightly to distinctly constricted at the septum, (11.8–)14.5–17.0(–19.5) \times (3.5–)4.3–5.3(–6.7) μm , $l/w=(2.3\text{--})2.9\text{--}3.7(5.0)$ ($n=261$); ends narrowly rounded to subacute, appendages mostly indistinct; cells slightly dimorphic, each containing 2 large guttules; wall not swelling, ca. 0.4 μm thick.

Anamorph: discosporina-like. **Conidiomata** inconspicuous, typically only the central or eccentric ectostromatic disc visible at the substrate surface or their presence indicated by discharged conidial masses, 1–2.5 mm diam. **Conidiogenous cells** phialidic, 10–25 \times 2–3 μm , hyaline. **Conidia** hyaline, fusoid or suballantoid, (7.5–)8.8–12.0(–14.3) \times (2.2–)2.5–3.0(–3.5) μm , $l/w=(2.3\text{--})2.9\text{--}4.4(5.8)$ ($n=215$), multiguttulate

with numerous small guttules; wall and hyaline gelatinous sheath each ca. 0.3 μm thick.

Culture: Colony on MEA after 14 d at room temperature ca. 14–35 mm diam, indistinctly zonate, surface slightly farinose due to loosely disposed, small collapsed flocks of aerial hyphae, first whitish, turning reddish-brown or rosy mixed with yellow tones from the centre; after ca. 3 wk purple pigment diffusing into the agar, colony turning dark reddish-brown and eventually dark brown, grey to black; diffusing pigment transient, unstable, grading off into brown.

Habitat and host range: dead corticated twigs and branches of *Carpinus caroliniana*; widespread.

Distribution: North America (USA).

Type: USA, Maryland, Prince George's County, Beltsville/Greenbelt, Beaver Dam Road, on corticated branches of *Carpinus caroliniana*, 14 May 2003, W. Jaklitsch & H. Voglmayr W.J. 2118, strain A.R. 3993 (BPI 843574, **holotype**; WU 31809, **isotype**; ex-holotype culture CBS 131617), ex-holotype sequences JQ926265 (ITS-LSU), JQ926336 (*rpb2*), JQ926402 (*tef1*).

Additional selected specimens examined (all on *Carpinus caroliniana*): CANADA, Ontario, London, July 1891, J. Dearness, Ellis & Everhart, N. Amer. Fungi. Ser. II: 2742 (K(M) 171590, issued as *Diaporthe decipiens*). USA, Maryland, Beltsville, Little Paint Branch, soc. *Discosporina carpinicola*, 20 May 1950, F. Petrak; Poelt & Scheuer, Reliqu. Petrak. 2815 (GZU, issued as *Discosporina carpinicola*). New York, vic. Ithaca, Arnot Forest, 10 July 2002, L. Vasilyeva, strain A.R. 3830 (BPI 870990, culture CBS 131494). Tennessee, Gatlinburg, Great Smoky Mountains National Park, Old Sugarlands Trail, 24 May 2003, W. Jaklitsch & H. Voglmayr, W.J. 2213 (BPI 872067). Virginia, Edinburg, George Washington National Forest, Columbia Furnace/Wolf Gap, 27 May 2003, W. Jaklitsch & H. Voglmayr, W.J. 2238 (WU 31810).

Notes: Asci, ascospores and anamorph of *Melanconiella elegans* are similar to those of *M. ellisii*. *Melanconiella elegans* has beautiful cream, yellowish to bright yellow ectostromatic discs, which are uniform in shape and characterised by evenly scattered ostioles. The colour of its central stromatic column is remarkably variable, ranging from cream, light to dark grey, yellow, olive to brownish. *Melanconiella elegans* is morphologically most similar to the European *M. carpinicola*, which may be the closest relative (Fig. 2). As the anamorph-teleomorph connection could not be directly proven with pure cultures, DNA was extracted from conidiomata present on the natural substrate. Three independent DNA extractions from two specimens (BPI 843574, BPI 872067) each confirmed the connection.

Morphological descriptions of *Melanconis xanthostroma* by Wehmeyer (1937), later referred to as *Melanconis chrysostroma* var. *ellisii* by him (Wehmeyer 1941), are likely to

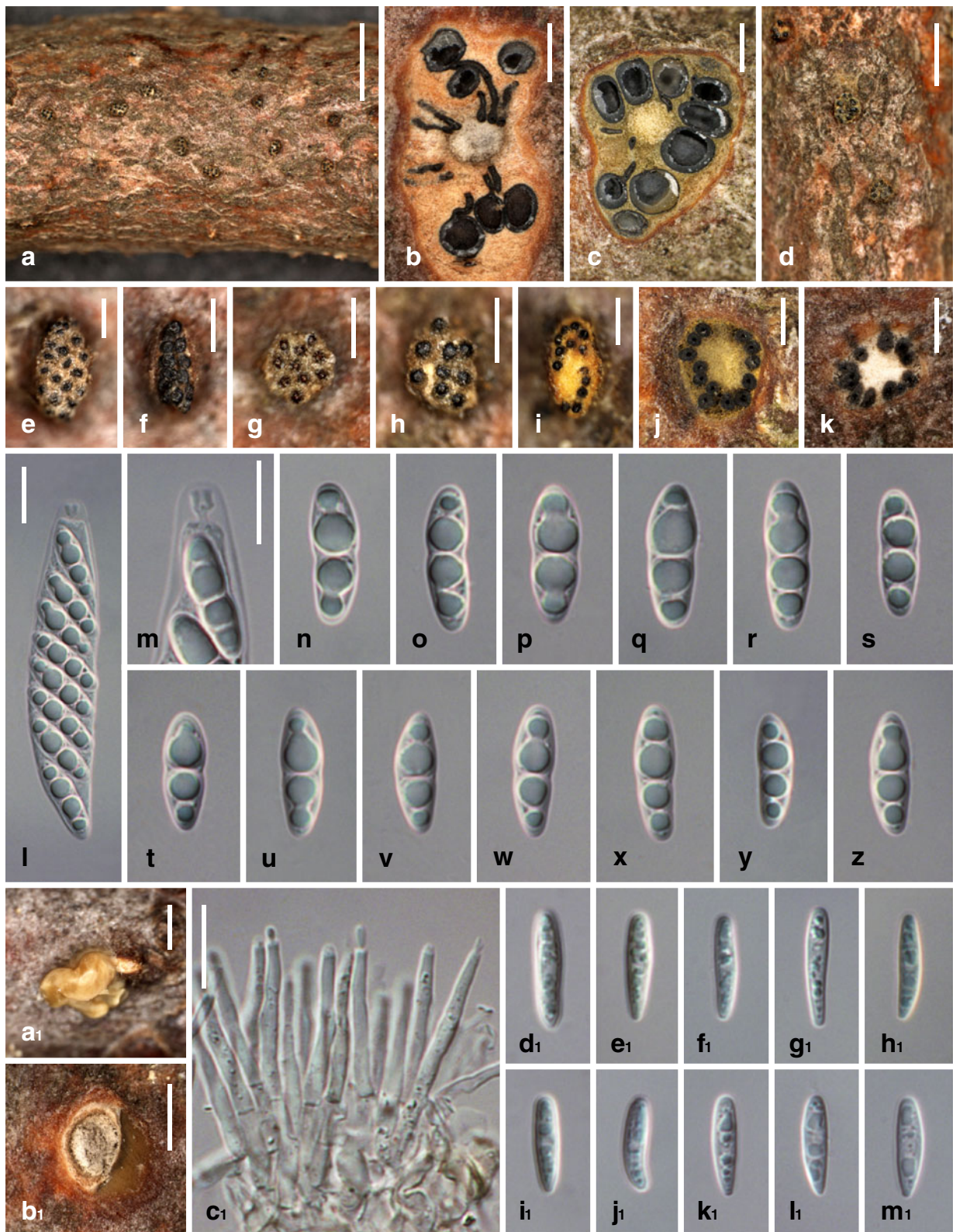


Fig. 10 *Melanconiella elegans*. **a**. Panoramic view of pseudostromata. **b, c**. Pseudostromata in transverse section. **d**. Two pseudostromata in surface view. **e–i**. Ectostromatic discs and ostioles in surface view. **j, k**. Transverse section below ectostromatic disc. **l**. Ascus. **m**. Ascus tip with apical ring. **n–z**. Ascospores. **a1**. Conidioma in surface view. **b1**.

Transverse section of conidioma, showing central column. **c1**. Conidiophores. **d1–m1**. Conidia. **d1–f1, i1, j1** in water, all other mounts in 3 % KOH. **a=2 mm, b, c, a1, b1=500 μm, d=1 mm, e–k=300 μm, l–z, c1–m1=10 μm**. Sources: **a, b, g, h, k, b1**. WU 31809; **c, h1**. BPI 872067; **e, f, j, m–s**. BPI 870990; **i, l, t–a1, c1–g1, i1–m1**. BPI 873574

be based on *M. elegans*. In his studies Wehmeyer (1937) reported the production of large alpha conidia (16–30(–37)×3.5–5 µm) in culture and similar conidia in a few herbarium specimens. However, his cultures did not produce the small beta-conidia he frequently observed on herbarium specimens. In one of our collections of *M. elegans* (BPI 870990), numerous conidiomata containing such large conidia were present. However, these could not be confirmed to represent a conidial state of *M. elegans*. A PDA culture from ascospores of BPI 870990 produced conidia similar in size (9–11×3–4 µm) to the small beta-conidia of Wehmeyer (1937, 1941), which were confirmed to represent the anamorph of *M. elegans* in our study. Sequences obtained from the large-spored anamorph on the natural substrate were either impure (from two extraction trials containing conidia, conidiophores and stromatic columns), or were revealed as an unidentified, probably diaporthean fungus not belonging to *Melanconiella* by a BLAST search (sequences from a DNA extract obtained only from conidia). This indicates that the conidia belong to a fungus that probably parasitizes stromata of *M. elegans*.

The type collection of *Discosporina carpinicola*, which was considered to be the anamorph of *M. ellisii* by Petrak (1952b), includes the teleomorph of *M. elegans*. However, morphologically the conidia present on the specimen are more similar to those of *M. ellisii*. Considering common co-occurrence and intermingled growth of teleomorphs and anamorphs of different species on the same branches in *Melanconiella* and the subtle morphological differences between the anamorphs of *M. ellisii* and *M. elegans*, it remains uncertain to which species of *Melanconiella* *Discosporina carpinicola* is connected. The species epithet is a later homonym of *Diaporthe carpinicola* and therefore not available for use in *Melanconiella*.

Melanconiella ellisii (Rehm ex Ellis & Everh.) Voglmayr & Jaklitsch, comb. nov. (Fig. 11)

Mycobank MB 800119

Basionym: *Diaporthe ellisii* Rehm ex Ellis & Everh., Bull Torr Bot Club 10:89 (1883).

Synonyms: ?*Discosporina carpinicola* Petr., Sydowia 6:357 (1952).

Melanconis chrysostroma var. *ellisii* (Rehm ex Ellis & Everh.) Wehm., Univ Michigan Stud Sci Ser 14:33 (1941).

Pseudostromata 1–2.6 mm diam, projecting up to ca. 0.7 mm, inconspicuous, circular, distinct or confluent; distinct perithecial bumps uncommon. *Ectostromatic disc* 0.2–1.2(–2) mm diam, with circular, angular or oblong outline, commonly surrounded by irregular edges or flaps of bark, drab, light to dark grey or dark brown. *Central column* grey or brownish. *Entostroma* typically indistinct, sometimes compact above perithecia, of subhyaline hyphae. *Ostioles*

1–15 per disc, 30–160(–170) µm diam at the base, typically arranged at the disc margin, shiny black, plane, papillate or conical and projecting up to 180 µm, tips flattened, rounded or acute. *Perithecia* 0.2–0.5 mm diam. *Asci* fusoid, (39–) 50–69(–80)×(7–)9–11(–13) µm (*n*=84), containing 8 uni- or biseriate ascospores, with distinct apical ring, 1.8–2.5 µm diam, 1.4–2 µm high. *Ascospores* hyaline, fusoid, slightly to distinctly constricted at the septum, (10–)12.5–16(–20)×(3.5–)4.0–5.5(–6.5) µm, l/w=(2.2–)2.7–3.3(–4) (*n*=239); ends narrowly rounded to subacute, mostly without appendages; cells mono- to slightly dimorphic, with (1–)2 large and numerous small guttules; wall not swelling, ca. 0.3 µm thick.

Anamorph: discosporina-like. *Conidiomata* inconspicuous, typically only their central or eccentric ectostromatic disc visible on the substrate surface or their presence indicated by discharged conidial masses, 0.5–2 mm diam. *Conidiogenous cells* phialidic, 10–18×2–3.5 µm, hyaline. *Conidia* hyaline, fusoid, narrowly ellipsoid, elongate to suballantoid, (8.5–)9.5–11.5(–13.0)×(2.5–)3.0–3.5(–4.0) µm, l/w=(2.2–)2.8–3.9(–4.8) (*n*=148), multiguttulate, commonly with four to six large guttules; wall ca. 0.3 µm, hyaline gelatinous sheath ca. 0.4 µm thick.

Habitat and host range: dead corticated twigs and branches of *Carpinus caroliniana*; widespread.

Distribution: North America (USA, Canada).

Typification: USA, Pennsylvania, West Chester, on *Carpinus caroliniana*, without date, Haines & Everhart, Ellis & Everhart, N. Amer. Fungi. Ser. II: 1567 (M-0177942, **lectotype of *Diaporthe ellisii* here designated**; BPI bound, K(M) 171591, **isotypes**).

Additional selected specimens examined (all on *Carpinus caroliniana*): USA, Maryland, Beltsville/Greenbelt, Beaver Dam Road, 14 May 2003, W. Jaklitsch & H. Voglmayr, W.J. 2118a (BPI 883227; WU 31811). Maryland, Prince George's Co., Beltsville Agricultural Research Station, Bottomland Research Forest, 3 June 2002, L. Vasilyeva (BPI 843491). Tennessee, Great Smoky Mountains National Park, vic. Cosby, 8 May 2006, L. Vasilyeva (BPI 878343).

Notes: *Melanconiella ellisii*, commonly referred to as *Melanconis chrysostroma* var. *ellisii*, was previously considered to be a well-defined taxon in North America (Wehmeyer 1941). However, the current study shows that it is a heterogeneous taxon, and *M. elegans* is separated as a distinct species. *Melanconiella ellisii* is characterised by its drab, grey to dark brown ectostromatic discs of irregular shape with ostioles inserted mostly at the margin of the ectostromatic disc, which is reminiscent of the closely related *M. spodiarea*. Unfortunately, no pure cultures have been available for *M. ellisii*, but DNA extraction from teleomorphs and anamorphs of three collections of *M. ellisii* revealed consistent results.



Fig. 11 *Melanconiella ellisii*. **a**. Panoramic view of pseudostromata. **b**. Pseudostroma in transverse section. **c**. Six pseudostromata in surface view. **d–i**. Ectostromatic discs and ostioles in surface view. **j**. Transverse section below ectostromatic disc. **k**. Ascus. **l**. Ascus tip with apical ring. **m–z**. Ascospores (m, r showing appendages). **a1**. Conidioma in surface view. **b1**. Transverse section of conidioma showing

central column. **c1**. Conidiophores. **d1–m1**. Conidia. All mounts in 3 % KOH. a=2 mm, b, d–i=500 μ m, c, j=1 mm, k–z, c1–m1=10 μ m, a1, b1=300 μ m. Sources: a, d, e, l, s–v. BPI 843491; b, f, j, k, w–z. BPI 878343; c–g, m, a1–b1, i1–m1. M-0177942 (lectotype); h, i, n–r. K(M) 171591 (isotype); c1–h1. BPI 883227)

Ellis and Everhart (1892) described the conidia as four-nucleate, measuring $8\text{--}11 \times 2\text{--}2.5 \mu\text{m}$. On the original collection deposited in Kew (K(M) 171591), the conidia are distinctly wider ($4\text{--}5.5 \mu\text{m}$) than those of the collection deposited in Munich (M-0177942), as well as in our collection BPI 883227 (WJ 2118a) for which the connection was confirmed by sequence data. It remains to be verified whether the conidia of *M. ellisii* show a variable width or whether these wider conidia belong to a different taxon.

For reasons of priority, Petrak (1952b) proposed that the species epithet *carpinigera* should be used for the species. However, this is a misapplied name, as the type specimen of the basionym, *Diatrype carpinigera*, is clearly not a diaporthean fungus but rather a *Valsaria* (see below). He also described *Discosporina carpinicola* as the anamorph of *M. ellisii*, but the type specimen contains the teleomorph of only *M. elegans*. As *D. carpinicola* is morphologically more similar to the anamorph of *M. ellisii*, it is here considered a synonym of the latter.

The recent record of *M. ellisii* (as *Melanconis carpinigera*) from *Carpinus cordata* near Vladivostok, Russian Far East by Vasilyeva and Stephenson (2010) cannot be evaluated without detailed study of the specimen.

Melanconiella flavovirens (G.H. Otth) Voglmayr & Jaklitsch, comb. nov. (Fig. 12)

Mycobank MB 800120

Basionym: *Diaporthe flavovirens* G.H. Otth, Mitt naturf Ges Bern 1868:47 (1869).

Synonyms: *Melanconis flavovirens* (G.H. Otth) Wehm., Mycologia 29:602 (1937).

?*Diaporthe flavovirens* var. *tetraspora* G.H. Otth, Mitt naturf Ges Bern 1868:47 (1869).

Diaporthe olivaestroma (Cooke) Berl. & Voglino [as 'elaeostroma'], in Saccardo, Syll Fung, Addit I–IV:105 (1886).

Diaporthe sulphurea Fuckel, Jb Nassau Ver Naturk 23–24:205 (1870) [1869–70] (type not seen).

Discodiaporthe sulphurea (Fuckel) Petr., Hedwigia 62:294 (1921).

Discosporina sulphurea (Sacc.) Petr., Sydowia 16:187 (1963) [1962].

Discosporium sulphureum (Sacc.) Petr., Hedwigia 62:292 (1921).

Melanconis sulphurea (Fuckel) Petr., Ann Mycol 21:321 (1923).

Myxosporium sulphureum Sacc., Syll Fung 3:725 (1884).

Valsa olivaestroma Cooke, Grevillea 14(no. 70):48 (1885).

Pseudostromata 1.5–3.4 mm diam, mostly irregularly confluent, with slightly projecting centre, sometimes well-defined, circular with circinate arranged perithecial

bumps. *Ectostromatic disc* 0.3–1.3 mm diam, circular, elliptic or oblong, often pulvinate, in this case not surrounded by bark, powdery, mostly dull yellowish green to olive, but also cream or yellow-brown, eventually black when old. *Central column* grey, olive-brown, dull green or lacking. *Entostroma* comprising olive or yellow mycelium, more or less well developed. *Ostioles* 1–15(–25) per disc, emerging in various positions in the disc, 40–160 μm diam, shiny black, slightly papillate, less commonly plane, sometimes cylindrical and projecting up to 150 μm when the disc disintegrates. *Perithecia* 0.3–0.6(–0.8) mm diam. *Asci* fusoid, (84–)95–107(–114) \times (13–)14–17(–18) μm ($n=31$), containing 8 biseriate ascospores, with distinct apical ring, 3.2–4.0 μm diam, 2.5–3.3 μm high. *Ascospores* hyaline, inequilaterally ellipsoid or broadly fusoid, not to slightly constricted at the septum, (17–)20.5–24.5(–30) \times (7.0–)7.5–8.8(–9.8) μm , l/w=(2.1–)2.5–3.0(–3.7) ($n=134$); ends narrowly rounded or subacute, with distinct persistent, knob-like hyaline appendages 0.7–1.4 μm long, 1.6–2.4 μm wide; cells mostly monomorphic, distinctly triangular-ovate in outline, with one large and numerous small guttules; wall not swelling, ca. 0.5–0.6 μm thick, at the septum 1.5–2 μm .

Anamorph: discosporina-like. *Conidiomata* 1.5–2.5 mm diam, indistinct, mostly with central or eccentric stromatic column; usually no discharged conidial masses evident. *Conidiogenous cells* phialidic, 16–27 \times 2.5–3.5 μm , hyaline. *Conidia* hyaline, variable in shape, pip-shaped, narrowly ellipsoid, elongate to suballantoid, sometimes with a distinct apiculus, (11–)12–15(–17.5) \times (4.7–)5.0–5.5(–6.5) μm , l/w=(1.9–)2.3–2.8(–3.3) ($n=65$), multiguttulate, often with one or two larger and numerous small guttules; wall and hyaline gelatinous sheath each ca. 0.5 μm thick.

Culture: Colony on MEA after 17 d at room temperature ca. 23 mm diam, after reaching a diam of 2–2.5 cm growth discontinued or at most small irregular lobes formed; colony dull greenish-yellow to yellow-brown with whitish margin, turning dark brown to black from the centre; aerial hyphae scant or abundant, causing a floccose surface; after 3–4 wk pale to medium brown pigment diffusing into the agar; odour reminiscent of fermenting yeast.

Habitat and host range: dead corticated twigs and branches of *Corylus avellana* lying on the ground.

Distribution: Europe, probably also North America (Canada, USA); common. Collections from North American *Corylus* species need to be thoroughly studied to evaluate whether they are conspecific.

Typification: SWITZERLAND, near Steffisburg, without date, G. Otth 31 (B 700014743, **lectotype of *Diaporthe flavovirens* here designated**; B 700014742, **isotype**). SWITZERLAND, 2 May 1867, P. Morthier, *Herb. Fuckel* 56 (G 00111561, **holotype of *Diaporthe sulphurea***). SWITZERLAND, Jura, 31 March 1871, P. Morthier, *Herb.*



Fig. 12 *Melanconiella flavovirens*. **a.** Panoramic view of pseudostromata. **b.** Pseudostroma in transverse section. **c.** Two pseudostromata in surface view. **d–g.** Ectostromatic discs and ostioles in surface view. **h.** Transverse section below ectostromatic disc. **i.** Ascus. **j.** Ascus tip with

apical ring. **k–w.** Ascospores. **x.** Transverse section of conidioma. **y.** Conidiophores. **z–k1.** Conidia. All mounts in water. **a–d,** **j–n,** **x–k1.** WU 31822; **e.** WU 31815; **f–i,** **o–w.** WU 31816

Fuckel 27 (G 00111404, **lectotype of *Myxosporium sulphureum* here designated**). UK, England, Middlesex,

Hendon, *M.C. Cooke 209* (K(M) 174473, **holotype of *Valsa olivaestroma***)

Additional specimens examined (all on *Corylus avellana*): AUSTRIA, Kärnten, St. Margareten im Rosental, Sabosach, map grid 9452/3, 30 Oct. 1994, W. Jaklitsch, W.J. 322 (WU 31812). *ibid.*, shrubs between the village and Stariwald, map grid 9452/4, 10 June 1993, W. Jaklitsch (WU 15584). *ibid.*, 27 Aug. 1993, W. Jaklitsch (WU 15510). *ibid.*, 19 Feb. 1994, W. Jaklitsch (WU 15376). *ibid.*, Triebloch, below Kucher, map grid 9452/2, 25 March 1995, W. Jaklitsch, W.J. 536 (WU 31813). *ibid.*, Schwarzgupf, map grid 9452/4, 10 Sep. 1998, W. Jaklitsch, W.J. 1202 (WU 31814). *ibid.*, Wograda, map grid 9452/3, 27 Oct. 2000, W. Jaklitsch, W.J. 1666 (WU 31815). *ibid.*, Zabrde, map grid 9452/4, 17 Oct. 2011, H. Voglmayr, strain MFV2 (from teleomorph) (WU 31816). Niederösterreich, Orth an der Donau, Tierboden, map grid 7866/3, 1 Nov. 2011, H. Voglmayr & W. Jaklitsch, strain MFV4 (from teleomorph) (WU 31817). Stockerau, Unterzögersdorf, map grid 7663/1, 24 April 2010, H. Voglmayr (WU 31818). Oberösterreich, Raab, Rotes Kreuz, map grid 7648/1, 6 Sep. 2009, H. Voglmayr, strain MFV (from teleomorph) (WU 31819, culture CBS 125598). St. Willibald, Aichet, map grid 7648/1, 3 June 2011, H. Voglmayr, strain MFV1 (from teleomorph) (WU 31820). Steiermark, Pöllau, Schönauklamm, map grid 8760/2, 12 Sep. 2002, W. Jaklitsch, W.J. 1952 (WU 31821). GERMANY, Reichartshausen, spring, without date, L. Fuckel, Fungi Rhen. Exs. 2539 (K(M) 174474, issued as *Diaporthe sulphurea*). ITALY, Lombardia, Vobarno, 21 Oct. 2011, H. Voglmayr & W. Jaklitsch, strains MFV3 (from teleomorph), MFA (from anamorph) (WU 31822). SWITZERLAND, Neuchâtel, 14 April 1869, P. Morthier, *Herb. Barbey-Boissier 105* (BPI 617275 as *Diaporthe sulphurea*). same place, date and collector (G 00111562 as *Diaporthe sulphurea*).

Notes: *Melanconiella flavovirens* is characterised by growth on *Corylus*, by mostly large ectostromatic discs with distinctly yellowish green tints and by broad ascospores of cells that are distinctly triangular to ovate in outline and by small persistent knob-like appendages. The anamorph is less prominent than in other *Melanconiella* species, inconspicuous and rarely seen. Despite thorough studies of several fresh collections, conidiomata were only found in a single collection from Italy. The anamorph, *Myxosporium sulphureum*, was formally described by Saccardo (1884), who referred to the description by Fuckel (1871). Apart from a few omissions, the wording of the diagnosis (including measurements) in Saccardo (1884) is identical with Fuckel's description (1871:318–319). Therefore, *Myxosporium sulphureum* should be based on a collection studied by Fuckel, and the specimen from the Fuckel herbarium housed in G is here designated as lectotype. The measurements of Fuckel (1871), repeated in Saccardo (1884), agree with ours. The anamorph-teleomorph connection was proven in the

present study by DNA data from conidial and ascospore cultures.

We examined two specimens of the type collection of *Diaporthe flavovirens* preserved in B. Considering the preservation and abundance of the collection, B 700014743 is here designated as lectotype. According to the detailed description and the host (Fuckel 1870), *Diaporthe sulphurea* is a synonym of *Melanconiella flavovirens*, which has been confirmed by examination of the type. Otth (1869) described a four-spored variety of *M. flavovirens*, but no such specimen has been seen.

Melanconiella hyperopta (Nitschke ex G.H. Otth) Voglmayr & Jaklitsch, comb. nov. (Fig. 13)

Mycobank MB 800121

Basionym: *Diaporthe hyperopta* Nitschke ex G.H. Otth, Mitt naturf Ges Bern 1868:47 (1869).

Synonyms: *Diaporthe decipiens* Sacc, Fung Ven 4:6 (1875) (type not seen).

Melanconis hyperopta (Nitschke ex G.H. Otth) Wehm., Univ Michigan Stud Sci Ser 9:254 (1933).

Pseudostromata typically distinct, well-defined, with circular outline, 1.5–3.4 mm diam, projecting up to 0.5 mm; perithecial bumps often evident; sometimes confluent and irregular. *Ectostromatic disc* concave, flat or slightly pustulate, surrounded by bark or not, little projecting above the perithecial level, 0.3–0.9 mm long, circular, fusoid to linear, whitish, cream, pale or dull yellow to brown. *Central column* pale, yellowish, pale greyish brown or brownish. *Entostroma* inconspicuous, pale, slightly lighter than bark interior, comprising subhyaline to yellowish hyphae. *Ostioles* 1–15 per disc, 60–160 μm diam, marginal or emergent within the disc, plane, discoid or papillate with rounded or flattened apices. *Perithecia* 0.3–0.8 mm diam. *Asci* fusoid, (73–)80–98(–111) \times (12–)13–17(–21) μm ($n=71$), containing 8 biseriata ascospores, with distinct apical ring, 2.6–3.1 μm diam, 2.4–3 μm high. *Ascospores* hyaline, fusoid, distinctly constricted at the septum, (17.7–)21–24(–29.5) \times (4.5–)5.5–6.8(–8.3) μm , $l/w=(2.9–)3.3–4(–5.1)$ ($n=472$); ends subacute to narrowly rounded, with hyaline cap-like appendages 1.3–2.4 μm long, 3.5–5.6 μm wide, soon fading in microscope mounts; cells dimorphic, upper cell mostly larger, multiguttulate; wall not swelling, *ca.* 0.4 μm thick.

Anamorph: discosporina-like. *Conidiomata* 1–1.5 mm diam, often lined by a distinct, narrow blackish marginal zone, with a central or eccentric stromatic column; at maturity covered by whitish discharged conidial masses. *Conidiogenous cells* phialidic, 10–30 \times 1.5–3 μm , hyaline. *Conidia* hyaline, variable in shape, narrowly ellipsoid, elongate to slightly allantoid, (10–)11.5–14(–16.5) \times (3.0–)3.8–4.5(–5.0) μm , $l/w=(2.2–)2.7–3.5(–4.6)$ ($n=172$),



Fig. 13 *Melanconiella hyperopta*. **a**. Panoramic view of pseudostromata. **b**. Pseudostroma in transverse section. **c**. Two pseudostromata in surface view. **d–g**. Ectostromatic discs and ostioles in surface view. **h**. Transverse section below ectostromatic disc. **i**. Ascus. **j**. Ascus tip with apical ring. **k–w**. Ascospores. **x**. Conidioma in surface view. **y**. Trans-

verse section of conidioma. **z, a1**. Conidiophores. **b1–l1**. Conidia. **k, l** in 3 % KOH, all other mounts in water. **a**=2 mm, **b–h, x, y**=500 μ m, **i–w, z–l1**=10 μ m. Sources: **a**. WU 31828; **b–e**. WU 31831; **f, g, k, l**. Otth 115 (B, holotype); **h, v**. WU 31827; **i, j, u, x–l1**. WU 31836; **m, n**. WU 31823; **o–p**. WU 31832; **q–t**. WU 31826; **w**. WU 31825

multiguttulate, often with one to four larger and few to numerous small guttules; wall *ca.* 0.4 μm thick; sometimes with an indistinct hyaline gelatinous sheath *ca.* 0.5 μm thick.

Culture: Colony on MEA after 17 d at room temperature *ca.* 75 mm diam, after *ca.* 20 d plate nearly entirely covered (no further growth); colony not zonate, surface with distinct radial to substellate arrangement of surface hyphae, aerial hyphae irregularly arranged, inconspicuous or causing minute floccules on the surface; colony first white, turning bright yellow to orange from the centre, eventually dull orange; diffusing pigment lacking, agar slightly yellowish.

Habitat and host range: dead corticated twigs and branches of *Carpinus betulus* mostly lying on the ground.

Distribution: Europe; common.

Typification: SWITZERLAND, near Bern, on corticated branch of *Carpinus betulus*, without date, *G. Otth 115* (B 700009156; **lectotype designated by Wehmeyer (1933)**). Niederösterreich, Gaaden, Badener Bürgerspitalswald, map grid 7963/1, on corticated branch of *Carpinus betulus*, 3 Dec. 2012, H. Voglmayr & I. Greilhuber, strain MHG (from anamorph) (WU 31836, **epitype here designated**; *ex-epitype* culture CBS 132231), *ex-epitype* sequences JQ926285 (ITS-LSU), JQ926351 (*rpb2*), JQ926418 (*tef1*).

Additional specimens examined (all on *Carpinus betulus*): AUSTRIA, Burgenland, Neusiedl/See, Edelstal, Spitzerberg, map grid 7967/2, 10 April 2010, H. Voglmayr & I. Greilhuber, strain MCS (from teleomorph) (WU 31823). Kärnten, St. Margareten im Rosental, Triebblach, near Ledra, map grid 9452/4, 1 May 2002, W. Jaklitsch, W.J. 1878, strain A.R. 3832 (BPI 843521, culture CBS 131492). Niederösterreich, Mannersdorf am Leithagebirge, Schweingraben, map grid 8065/2, soc. *M. chrysodiscosporina* (anamorph), 17 April 2010, H. Voglmayr & I. Greilhuber (WU 31824). Mauerbach, near the cemetery, map grid 7763/1, 28 Sep. 1996, W. Jaklitsch, W.J. 968 (WU 31825). *ibid.*, 12 June 2011, W. Jaklitsch (WU 31826). Oberösterreich, Raab, Riedlhof, map grid 7647/2, 4 April 2010, H. Voglmayr, strain MCR (from teleomorph) (WU 31827). Raab, Wetzlbach, map grid 7648/1, 4 April 2010, H. Voglmayr (WU 31828). St. Willibald, Loitzmayr, map grid 7648/3, 8 April 2010, H. Voglmayr, strain MCL (from teleomorph) (WU 31829, culture CBS 131697). Wien, Döbling, Cobenzl, map grid 7763/2, 11 Feb. 2001, W. Jaklitsch, W.J. 1715, strain A.R. 3619 (BPI 840939, WU 31830, culture CBS 131493). Döbling, Hermannskogel, map grid 7763/2, soc. *M. chrysodiscosporina* (anamorph), 18 April 2010, H. Voglmayr, strain MCHE (from teleomorph) (WU 31831, culture CBS 131696). Landstraße, Botanical Garden, map grid 7864/1, 31 March 2010, H. Voglmayr, strain MCHBV (from teleomorph) (WU 31832). Liesing, Maurer Wald, map grid 7863/1, 3 Oct. 1998, W. Jaklitsch, W.J. 1224 (WU 31833). *ibid.*, 22 April

2006, W. Jaklitsch, W.J. 2911 (WU 31834). Ottakring, Wilhelminenberg, Kreuzeichenwiese, map grid 7763/4, 19 March 2006, H. Voglmayr (WU 31835). GERMANY, Bayern, München, Allach, Lochholz, 16 March 1998, L. Beenken, Triebel, Microf. Exs. 273 (UPS 173895). Sachsen-Anhalt, Eisleben, Oberrissdorfer Tal, May 1875, J. Kunze, Fungi Sel. Exs. 122 (M-0177941, issued as *Diaporthe decipiens*). SWEDEN, Skåne, Helsingborg, Gyhults gård, 20 Jan. 1997, S.-Å. Hanson 25634 (UPS 200066). Skåne, Helsingborg, Frederiksdals Friluftsmuseum, 28 Feb. 2012, S.-Å. Hanson 2012-007 (WU 31615). Skåne, Södra Mellby par, Stenshuvud, 20 Oct. 1990, K. & L. Holm 5607a (UPS 412043, UPS 412044, culture CBS 114600). UKRAINE, Stryj, Podhorce, 27 Dec. 1916, F. Petrak, Fungi Polon. Exs. 637 (M-0157851, W).

Notes: Besides *Melanconiella spodiaea*, *M. hyperopta* is the most common species of the genus occurring on *Carpinus betulus* in Europe, where it is mainly found on broken or fallen branches lying on the ground within forests. It is commonly associated with *M. chrysodiscosporina*. During attempts to establish anamorph-teleomorph connections in the present study, *M. chrysodiscosporina* was isolated several times from conidiomata closely associated with stromata of *M. hyperopta*. Although the conidia of both species are hyaline and of similar size, those of *M. chrysodiscosporina* can be distinguished by their more regular, cylindrical shape and the presence of two large guttules in the cell content. The anamorph-teleomorph connection was proven by DNA data from conidial and ascospore cultures. A culture of a Swedish collection (CBS 114600) had distinct ITS-LSU, *rpb2* and *tef1* sequences (Fig. 2), but the microscopic features of the specimen (UPS 412044) agreed fully with the other collections from *Carpinus betulus*. The collections from *Carpinus orientalis* are described as a new variety based on slightly smaller ascospores and the different host (see below).

Wehmeyer (1933) proposed the combination *Melanconis hyperopta*, but he included *M. carpinicola* in his species concept. Finally, he synonymised *M. hyperopta* with *M. chrysostroma* (Wehmeyer 1941). According to the spore sizes given by Munk (1957) and Ellis and Ellis (1997), their concept of *Melanconis chrysostroma* was evidently based on collections of *M. hyperopta*. According to Gerhardt and Hein (1979), the herbarium of Nitschke, containing duplicates of Otth's material, is present in the herbarium in Berlin (B). The spore size given in Otth (1869) is somewhat smaller (18–20 \times 4–5 μm) than that revealed for the authentic collection deposited in Berlin (20–26 \times 5–7.5), which is within the typical range of *M. hyperopta*. A similar case of underestimated spore sizes given by Otth (1869) was also recorded for *Massaria pyri* (Voglmayr and Jaklitsch 2011), indicating that the measurements given by Otth may

commonly be too small. To ensure nomenclatural stability, a recent collection, for which cultures and sequences are available, is designated here as epitype.

The basionym of *M. hyperopta*, *Diaporthe hyperopta*, has been commonly ascribed to Nitschke, following Otth (1869) who attributed the taxon to Nitschke. However, there is no indication that Nitschke provided a diagnosis, and Otth himself stated in the introduction that the descriptions were intended to be provisional and based on his own observations (Otth 1869:37–38). Therefore, the author citation for *D. hyperopta* should be Nitschke ex G.H. Otth.

Although no type specimen was available for study, *Diaporthe decipiens* is clearly a synonym of *M. hyperopta*, according to spore size and host. *Diaporthe nigro-annulata* Kunze, considered a synonym of *M. hyperopta* by Wehmeyer (1933), has apparently never been validly published; the schedae of the exsiccatum listed as type (Kunze, Fungi Sel. Exs. 122) were issued as *Diaporthe decipiens* Sacc., giving “Diap. nigro-annulata Johs. Kze. in sched.” as a synonym. Re-examination of two specimens of Kunze, Fungi Sel. Exs. 122 from Munich revealed one as *M. hyperopta* (M-0177941), whereas the other contains immature *M. carpnicola* (M-0177940).

Melanconiella hyperopta* var. *orientalis Voglmayr & Jaklitsch, var. nov. (Fig. 14)

Mycobank MB 800122

Etymology: referring to the epithet of the host, *Carpinus orientalis*.

Pseudostromata 1.5–3 mm diam, typically distinct, well-defined, with circular outline, projecting up to 0.5 mm, perithecial bumps often evident; sometimes confluent and irregular. *Ectostromatic disc* typically flat, little projecting above the perithecial level, 0.2–0.6(–0.8) mm long, circular to linear, whitish, cream, light or pale yellow. *Central column* whitish, grey or brownish, entostroma inconspicuous. *Ostioles* 1–12 per disc, 50–160 µm diam, marginal or disposed within the disc, plane, discoid or papillate with rounded or flattened apices. *Perithecia* 0.3–0.6 mm diam. *Asci* fusoid, (84–)87–103(–107)×13–17(–18) µm ($n=12$), containing 8 biseriolate ascospores, with distinct apical ring, 2.5–3.6 µm diam, 1.8–2.3 µm high. *Ascospores* hyaline, fusoid, distinctly constricted at the septum, (17.5–)18.7–22(–24)×(4.7–)5.7–6.7(–7.5) µm, $l/w=(2.7–)3.0–3.5(–4.4)$ ($n=110$); ends subacute to narrowly rounded, with hyaline cap-like appendages 1–2 µm long, 2.7–4.2 µm wide, soon fading in microscope mounts; cells dimorphic, upper cell mostly larger, multiguttulate; wall not swelling, *ca.* 0.4 µm thick.

Anamorph: discosporina-like. *Conidiomata* 0.9–1.5 mm diam, visible as darker spots and lined by a darker marginal zone, mostly with a central or eccentric stromatic column; at

maturity covered by whitish conidial masses. *Conidiogenous cells* phialidic, 7–22×2–3.5 µm, hyaline. *Conidia* hyaline, variable in shape, narrowly ellipsoid, elongate to slightly allantoid, (9.3–)10.5–12.7(–14.5)×(3.0–)3.5–4.0(–4.5) µm, $l/w=(2.2–)2.8–3.6(–4.4)$ ($n=71$), multiguttulate, often with one to three larger and few to numerous small guttules; wall and hyaline gelatinous sheath each *ca.* 0.5 µm thick.

Culture: Colony on MEA after 17 d at room temperature 83 mm diam, after 4–5 wk plate nearly entirely covered by mycelium. Colony white, with pale brownish spots, or centre white, followed by a diffuse brownish, less commonly rosy zone, otherwise not zonate, with distinct radial to sub-stellate arrangement of surface hyphae; aerial hyphae scant, sometimes abundant in the centre, diffusing pigment lacking; odour slightly musty.

Habitat and host range: dead corticated twigs and branches of *Carpinus orientalis*; only found on branches attached to the trees.

Distribution: South-eastern Europe, uncommon; only known from Croatia but likely to co-occur elsewhere with its host.

Type: CROATIA, Istria, Krnica, on corticated twigs of *Carpinus orientalis*, 15 May 2010, H. Voglmayr & W. Jaklitsch, strain MSK (from teleomorph) (WU 31837, **holotype**; ex-holotype culture CBS 131705), ex-holotype sequences JQ926286 (ITS-LSU), JQ926354 (*rpb2*), JQ926421 (*tefl*).

Additional specimen examined: CROATIA, Istria, Poreč, Gedici, on corticated twigs of *Carpinus orientalis*, 1 Nov. 2010, H. Voglmayr & I. Greilhuber, strains MHP (from teleomorph), MHVA (from anamorph) (WU 31838).

Notes: *Melanconiella hyperopta* var. *orientalis* is similar to the closely related *M. hyperopta*, from which it differs mainly by host, slightly smaller ascospores and conidia and ITS, *rpb2* and *tefl* sequences. As these differences are subtle, the fungus on *Carpinus orientalis* is described as a new variety of *M. hyperopta*. The spores and conidia exhibit pronounced drought resistance, as most were still vital after storage for more than one year. The anamorph-teleomorph connection was proven by DNA data from conidial and ascospore cultures.

Melanconiella meridionalis Voglmayr & Jaklitsch, sp. nov. (Fig. 15)

Mycobank MB 800123

Etymology: referring to the southern distribution within Europe.

Pseudostromata 0.8–2.3 mm diam, projecting up to *ca.* 0.5 mm, inconspicuous, indistinct, confluent, less commonly distinct and circular, causing minute bumps in the bark; perithecial bumps distinct or not. *Ectostromatic disc*

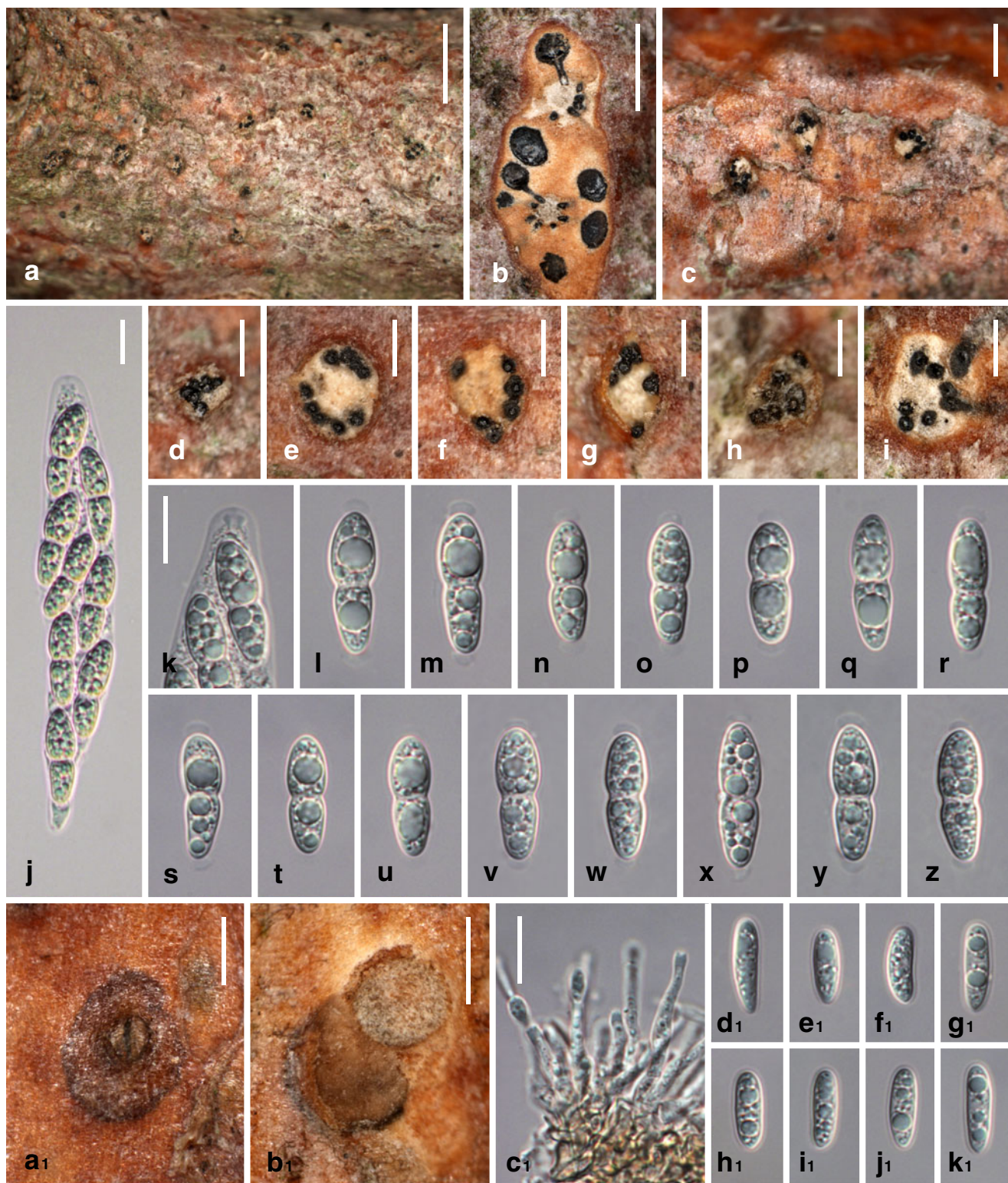


Fig. 14 *Melanconiella hyperopta* var. *orientalis*. **a**. Panoramic view of pseudostromata. **b**. Two pseudostromata in transverse section. **c**. Three pseudostromata in surface view. **d–h**. Ectostromatic discs and ostioles in surface view. **i**. Transverse section below ectostromatic disc. **j**. Ascus. **k**. Ascus tip with apical ring. **l–z**. Ascospores. **a1**. Conidioma

in surface view. **b1**. Transverse section of conidioma showing central column. **c1**. Conidiophores. **d1–k1**. Conidia. All mounts in water. a= 2 mm, b=1 mm, c=500 μm, d–i, a1, b1=300 μm, j–z, c1–k1=10 μm. Sources: a, b, h, i, v–z. WU 31837; c–g, j–u, a1–k1. WU 31838

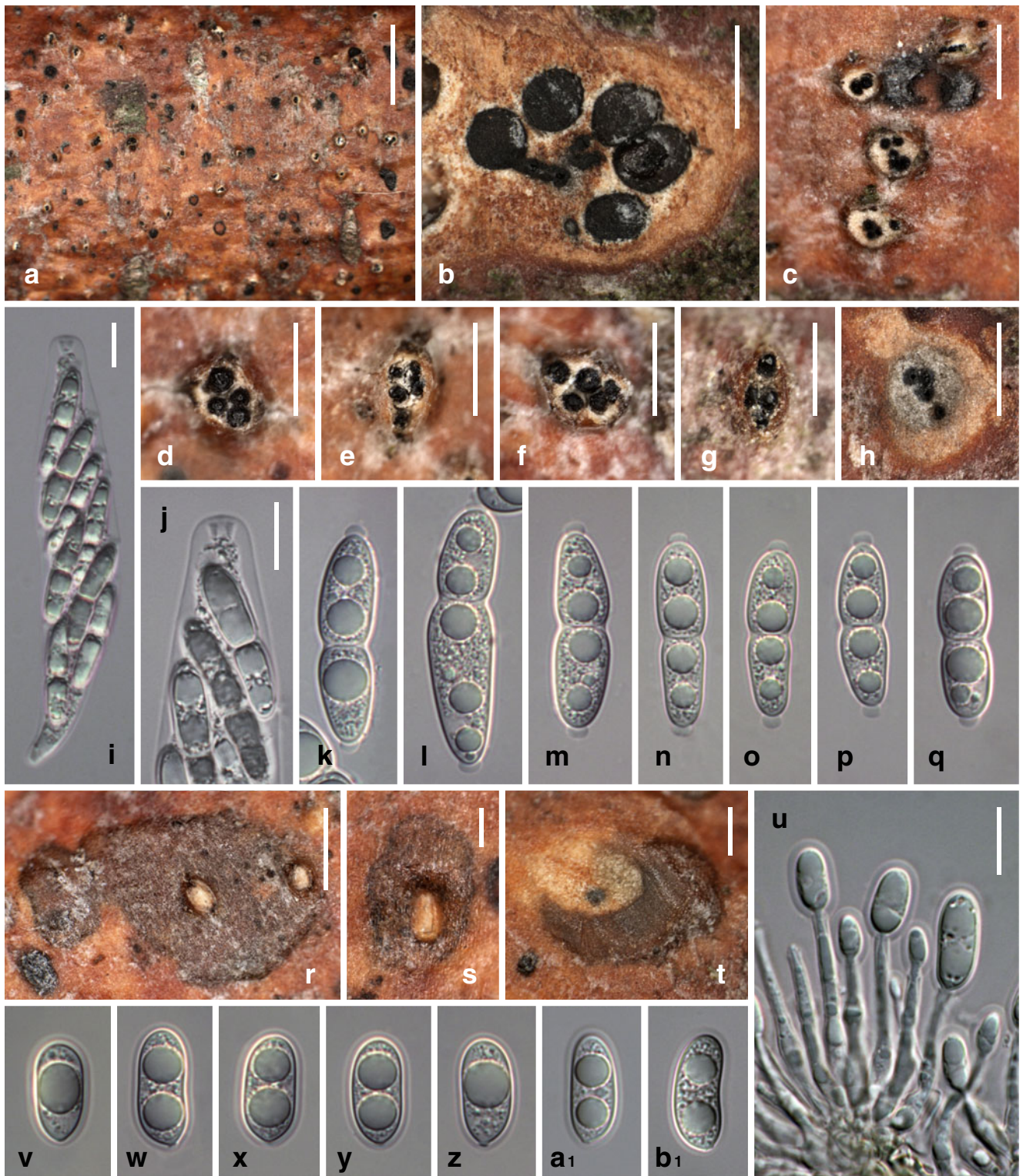


Fig. 15 *Melanconiella meridionalis*. **a.** Panoramic view of pseudostromata. **b.** Pseudostroma in transverse section. **c.** Group of pseudostromata in surface view. **d–g.** Ectostromatic discs and ostioles in surface view. **h.** Transverse section below ectostromatic disc. **i.** Ascus. **j.** Ascus tip with apical ring. **k–q.** Ascospores. **r, s.** Conidiomata in surface view. **t.**

Transverse section of conidioma. **u.** Conidiophores. **v–b1.** Conidia. All mounts in water. **a=2 mm, b–h, r=500 μm, s, t=200 μm, i–q, u–b1=10 μm.** Sources: **a, c, d–f, r, s, u.** WU 31845; **b, g–j.** WU 31844; **k–p.** WU 31849; **q.** WU 31846; **t.** WU 31839; **v–b1.** WU 31840

typically minute, 0.1–0.6 mm long, circular, oblong or linear, whitish, cream, pale yellowish to greyish or greyish brown, or concealed by ostioles. *Central column* (pale) grey or greyish brown. *Entostroma* whitish, of hyaline hyphae, scant or well-developed. *Ostioles* few, 1–5 per disc, commonly only 1–3, rarely 7, 50–125(–160) μm diam, shiny black, plane or slightly papillate with flattened or rounded tips. *Perithecia* valsoid around the column or solitary, 0.2–0.5(–0.8) mm diam. *Asci* broadly cylindrical to fusoid, (85–)91–104(–115) \times (13–)15–18(–21) μm ($n=31$), containing 8 uni- or irregularly biserial ascospores, with distinct apical ring, 3.5–4.4 μm diam, 2.4–3 μm high. *Ascospores* hyaline, fusoid, constricted at the septum, (20–)23–28(–38) \times (5.0–)6.5–8.0(–9.5) μm , $l/w=(2.8–)3.2–3.9(–4.6)$ ($n=90$); ends narrowly rounded, with hyaline persistent cap-like appendages 1–1.8 μm long, 2.8–3.4 μm wide; cells dimorphic, upper cell mostly larger, with (1–)2 large and numerous small guttules; wall not swelling, 0.4 μm thick.

Anamorph: discosporina-like. *Conidiomata* 0.4–1.5 mm diam, visible as dark brown spots lined by a darker marginal zone, with a central or eccentric stromatic column; at maturity covered by whitish discharged conidial masses. *Conidiogenous cells* phialidic, 19–37 \times 3–4 μm , hyaline. *Conidia* hyaline, cylindrical to suballantoid, (13–)15–17.5(–19.5) \times (5.8–)6.3–7.0(–8.0) μm , $l/w=(1.7–)2.2–2.7(–3.2)$ ($n=120$), with (1–)2 large and numerous small guttules; wall ca. 0.4–0.6 μm thick, with prominent gelatinous sheath 0.8–1.2 μm thick.

Culture: On MEA growth conspicuously slow, colony reaching a diam of ca. 16 mm after 17 d and ca. 22 mm after 4 wk, white with diffuse yellow centre, dense, compact, thick; aerial hyphae short, forming delicate flocks; diffusing pigment lacking; odour slightly yeast-like.

Habitat and host range: dead corticated twigs and branches of *Ostrya carpinifolia* mostly still attached to the tree.

Distribution: Southern Europe; known from Austria, Croatia, Greece, Italy and Slovenia; common in the natural distribution range of its host.

Type: AUSTRIA, Kärnten, St. Margareten im Rosental, on corticated branches of *Ostrya carpinifolia*, map grid 9452/4, 12 May 2010, H. Voglmayr, strain MOM (from teleomorph) (WU 31839, **holotype**; ex-holotype culture CBS 131704), ex-holotype sequences JQ926291 (ITS-LSU), JQ926357 (*rpb2*), JQ926424 (*tef1*).

Additional specimens examined (all on *Ostrya carpinifolia*): AUSTRIA, Kärnten, St. Margareten im Rosental, Novina, map grid 9452/4, 3 July 2007, W. Jaklitsch, W.J. 3106 (WU 31842). *ibid.*, shrubs W of Stariwald, map grid 9452/4, 17 May 2010, H. Voglmayr & W. Jaklitsch, strain MOA (from anamorph) (WU 31840). *ibid.*, map grid 9452/4, 10 Sep. 2010, W. Jaklitsch (WU 31841). *ibid.*, Wograda, map grid 9452/3, 13 Sep. 2001, W. Jaklitsch, W.J. 1810

(WU 31843). CROATIA, Primorsko-goranska (Istria), nature park Učka, Veprinac WSW Opatija, 15 May 2010, H. Voglmayr & W. Jaklitsch, strain MOU (from teleomorph) (WU 31844). Vela Učka, Poklon, 15 May 2010, H. Voglmayr & W. Jaklitsch, strain MOK (from teleomorph) (WU 31845). GREECE, Kerkyra (Corfu), Kastellani near Troumpettas, 21 April 2012, H. Voglmayr & W. Jaklitsch (WU 32003). ITALY, Toscana, Grosseto, Sovana, 29 July 2009, H. Voglmayr, W. Jaklitsch & W. Gams, strain MOSO (from teleomorph) (WU 31846). Trentino, Nembia, 19 Oct. 2011, H. Voglmayr & W. Jaklitsch (WU 31847). SLOVENIA, Vipava, Mt. Nanos massif, Rebrnice NE Lozice, 13 June 2009, H. Voglmayr & I. Greilhuber, strain MON (from teleomorph) (WU 31848). Divača, Škocjan, 12 June 2009, H. Voglmayr & I. Greilhuber, strain MOS (from teleomorph) (WU 31849). Divača, Škocjanska jama, 12 June 2009, H. Voglmayr & I. Greilhuber, strain MOSJ (from teleomorph) (WU 31850, culture CBS 125599).

Notes: *Melanconiella meridionalis* is characterised by its large, mostly biguttulate ascospores with dimorphic cells and the growth on *Ostrya carpinifolia*. In addition, it has the least developed stroma of all *Melanconiella* species. It is closely related to the North American *Melanconiella ostryae*, which differs by distinctly smaller ascospores (15–23 \times 4–8 μm), dark brown conidia with a lighter equatorial zone and the different host *O. virginiana*. The anamorph-teleomorph connection was proven by DNA data from conidial and ascospore cultures.

Melanconiella ostryae (Dearn.) Voglmayr & Jaklitsch, comb. nov. (Fig. 16)

Mycobank MB 800124

Basionym: *Diaporthe ostryae* Dearn., Mycologia 18:246 (1926).

Synonym: *Melanconis ostryae* (Dearn.) Wehm., Mycologia 29:601 (1937).

Pseudostromata 1–2.2 mm diam, typically inconspicuous, sometimes distinct, circular, slightly projecting to ca. 0.5 mm, perithecial bumps uncommon. *Ectostromatic disc* small, well-defined, with circular or elliptic outline, 0.1–0.5 mm long, whitish, cream, grey to pale greyish brown. *Central column* whitish, grey or brownish. *Entostroma* whitish or cream, of hyaline hyphae. *Ostioles* 1–5(–7) per disc, variously arranged, 40–125 μm diam, shiny black, flat or papillate. *Perithecia* 0.2–0.5 mm diam. *Asci* cylindrical to fusoid, (55–)77–100(–120) \times 10–13(–14) μm ($n=57$), containing 8 irregularly biserial ascospores, with apical ring distinct when fresh, 2.8–3 μm diam, 2.2–3 μm high, only faintly visible in older herbarium specimens. *Ascospores* hyaline, fusoid, slightly to not constricted at the septum, (14.5–)16.5–19(–23) \times (4.0–)5.5–7.0(–8.3) μm , $l/w=(2.1–)2.5–3.3(–4.2)$ ($n=209$); ends narrowly rounded to subacute,



Fig. 16 *Melanconiella ostryae*. **a**. Panoramic view of pseudostromata. **b**. Pseudostroma in transverse section. **c**. Three pseudostromata in surface view. **d–g**. Ectostromatic discs and ostioles in surface view. **h**. Transverse section below ectostromatic disc. **i**. Ascus. **j**. Ascus tip with apical ring. **k–y**. Ascospores (k, u–y showing appendages). **z**, **a1**. Conidiomata in surface view. **b1**. Transverse section of conidioma,

showing central column. **c1**, **d1**. Conidiophores. **e1–k1**. Conidia. k–v in 3 % KOH, all other mounts in water. a=2 mm, b, c, z–b1=500 μm, d–h=200 μm, i–y, c1–k1=10 μm. Sources: a, e–q, b1. Bowser 37–102 (NY); b–d, u, v. Dearness 27 July 1893, Ellis 2162 (NY); r–t, a1. M.E.B.B. 4365 (NY); w–y. Ellis 2162B (NY); z. TRTC 44844 (NY); c1–k1. BPI 615036

with hyaline cap-like appendages 0.6–2 μm long, 2.2–3.8 μm wide, soon disappearing in microscope mounts; cells slightly dimorphic, upper cell mostly larger, multiguttulate, sometimes with one large and numerous small guttules per cell; wall not swelling, *ca.* 0.4 μm thick.

Anamorph: melanconium-like. *Conidiomata* 1–1.5 mm diam, margins not visible on the surface, with central or eccentric stromatic column; at maturity covered by black discharged conidial masses. *Conidiogenous cells* annellidic, 20–32 \times 3–5 μm , hyaline. *Conidia* dark brown, with a light brown equatorial zone *ca.* 1.5–2.2 μm wide, ellipsoid, slightly constricted in the middle, (12–)13–15.5(–17) \times (6.2–)7.0–8.0(–8.8) μm , l/w=(1.6–)1.7–2.1(–2.4) ($n=76$), without guttules or granules; wall and hyaline gelatinous sheath each *ca.* 0.5–0.7 μm thick.

Habitat and host range: dead corticated twigs and branches of *Ostrya virginiana*.

Distribution: North America (USA, Canada); common.

Typification: CANADA, Ontario, Mt. Pleasant near London, on corticated branches of *Ostrya virginiana*, 6 July 1904, J. Dearness 3152 (DAOM, **lectotype of *Diaporthe ostryae* here designated**); same place, date and collector, herbarium L. E. Wehmeyer 3278 (DAOM 120563, **isotype**).

Additional specimens examined (all on *Ostrya virginiana*): CANADA, Ontario, London, 27 July 1893, J. Dearness, (NY, 2 specimens: Ellis 2162 & 2162B). *ibid.*, without date, J. Dearness, Ellis & Everhart, N. Amer. Fungi. Ser. II: 3430 (NY). Huron Distr., Bruce Co., Brinkman Corner, 31 May 1964, R. F. Cain, TRTC 44844 (NY). Bruce Co., Garrick twp., Con. XIII, Lot 31, 22 July 1960, R. L. Bowser 37-102 (NY). York Co., Nashville, 2 May 1954, R. F. Cain (NY). USA, Vermont, Lamoille Co., Stowe, Covered Bridge Road, 2 July 1964, H. E. & M. E. Bigelow M.E.B.B. 4191 (NY). *ibid.*, Pinnacle Trail, 21 July 1964, H. E. & M. E. Bigelow M.E.B.B. 4365 (NY).

Notes: *Melanconiella ostryae* is characterised by growth on *Ostrya virginiana* and dark brown conidia with a distinctly lighter brown equatorial zone, which is unique in *Melanconiella*. Due to the dark brown conidia, which are produced in large numbers, colonised twigs are blackened, providing a strong contrast to the whitish ectostromatic discs. In several collections, no black ostioles were visible in the ectostromatic discs, and sections revealed that no or only aborted perithecia were present. Due to the lack of fresh collections, no cultures and sequences could be obtained from the anamorph in the present study. However, the anamorph-teleomorph connection was experimentally proven in pure culture studies by Wehmeyer (1937). *Melanconiella ostryae* is closely related to *M. meridionalis*, which differs by distinctly larger ascospores (20–38 \times 5–10 μm), hyaline conidia, a different host (*O. carpinifolia*) and European distribution.

Melanconiella spodiaea (Tul.) Sacc., Syll Fung 1:740 (1882). (Fig. 17)

MycoBank MB210704

Basionym: *Melanconis spodiaea* Tul., Ann Sci nat, ser 4, 5:109 (1856) (type not seen).

Pseudostromata inconspicuous, 1.3–3 mm diam, little projecting, circular, perithecial contours typically not evident. *Ectostromatic disc* 0.25–1.1 mm long, mostly oblong, sometimes circular or triangular, generally laterally enclosed by bark, projecting 0.1–0.5 mm, often concealed by densely disposed ostioles, light to dark grey, greyish brown, light to dull or dark brown. *Central column* well-defined, light to dark grey, greyish brown or grey-olive, perforated by ostioles. *Entostroma* of subhyaline or yellowish hyphae. *Ostioles* often in 1 or 2 rows, 1–15 per disc, 60–190 μm diam, shiny black, stout, papillate to short cylindrical, with rounded, sometimes flattened apices, distinct or fused in the disc. *Perithecia* 0.3–0.5 mm diam, up to 20 per stroma. *Asci* cylindrical, (104–)110–120(–121) \times (14–)15–17(–18) μm ($n=16$), containing 8 uniseriate ascospores, with distinct apical ring, 3–3.8 μm diam, 1–1.5 μm high. *Ascospores* dark umber brown, broadly ellipsoid, slightly constricted at the straight to slightly oblique septum, (13.5–)15.5–18.3(–19.5) \times (6.5–)7.5–8.8(–9.8) μm , l/w=(1.6–)1.9–2.3(–2.5) ($n=82$); ends broadly rounded, with broad, persistent, hyaline cap-like appendages 1–1.7 μm long, 4–5.5 μm wide; cells monomorphic to slightly dimorphic, multiguttulate, often with one large guttule almost entirely filling the lumen and few peripheral small guttules; wall not swelling, *ca.* 0.7 μm thick.

Anamorph: melanconium-like. *Conidiomata* 1–4 mm diam, blackish, mostly with central or eccentric stromatic column; at maturity covered by black discharged conidial masses. *Conidiogenous cells* annellidic, 20–29 \times 2.5–5 μm , hyaline. *Conidia* dark umber brown, variable in shape, ovoid, obovoid, ellipsoid, oblong, sometimes shmoo-shaped or slightly constricted in the middle, (12.5–)13.3–15.2(–16.5) \times (7.0–)7.5–8.5(–9.0) μm , l/w=(1.4–)1.6–1.9(–2.1) ($n=31$), multiguttulate; wall and gelatinous sheath each *ca.* 0.7 μm thick.

Culture: Colony on MEA after 17 d at room temperature *ca.* 64 mm diam, with continuous or distinctly lobed margin, surface whitish, farinose or granulose due to aerial hyphae that form densely disposed floccules; floccules producing hyaline, rosy to violaceous brown drops; reverse and agar turning rosy, later reddish brown 9–10DE5–8 by a diffusing pigment; eventually colony reverse dark red, purple to nearly black, surface dull whitish to grey; agar decolourised to pale brownish; odour typically distinctly musty (like fermenting fruits).

Habitat and host range: dead corticated twigs and branches of *Carpinus betulus* and *C. orientalis*.



Fig. 17 *Melanconiella spodiaea*. **a**. Panoramic view of pseudostromata. **b**. Two pseudostromata in transverse section. **c**, **h**. Transverse sections below ectostromatic disc. **d–g**. Ectostromatic discs and ostioles in surface view. **i**. Ascus. **j**. Ascus tip with apical ring. **k–w**. Ascospores. **x**, **y**. Conidiomata in surface view. **z**. Transverse section of

two conidiomata, showing central columns. **a1**. Conidiophores. **b1–h1**. Conidia. All mounts in water. **a**, $x=2$ mm, **b–h**, **y**, $z=500$ μ m, **i–w**, **a1–h1** $=10$ μ m. Sources: **a–d**, **i**, **p–u**, **x–h1**. WU 31854; **e–h**, **v**, **w**. WU 31855; **j–o**. WU 31614

Distribution: Europe; common and widespread.

Specimens examined: On *Carpinus betulus*: AUSTRIA, Burgenland, Hornstein, Lebzelterberg, map grid 8064/4, 11 Sep. 2010, H. Voglmayr, strain MSH (from teleomorph) (WU 31851). Kärnten, St. Margareten im Rosental, Ledra, map grid 9452/4, 7 June 1996, W. Jaklitsch, W.J. 893 (WU 31852). Niederösterreich, Michelbach Markt, Mayerhöfen, Hegerberg, map grid 7860/4, 20 June 2009, W. Jaklitsch & H. Voglmayr, strain SPOD (WU 31853, culture CBS 125641). *ibid.*, 3 June 2000, W. Jaklitsch, W.J. 1485, strain A.R. 3462 (BPI 747944, culture CBS 109762). Mauerbach, near the cemetery, map grid 7763/1, 12 Nov. 2011, W. Jaklitsch, strain SPOD1 (from anamorph) (WU 31854). *ibid.*, 22 Aug. 1992, W. Jaklitsch (WU 16067). Hainbuch, map grid 7763/1, 21 Aug. 1993, W. Jaklitsch (WU 15993). Gaaden, Badener Bürgerspitalswald, map grid 7963/1, 3 Dec. 2012, H. Voglmayr & I. Greilhuber (WU 31614). Oberösterreich, Raab, Großbrothmayr, map grid 7647/2, 4 April 2010, H. Voglmayr (WU 31855). Wien, Döbling, Hermannskogel, map grid 7763/2, 15. Feb. 1998, W. Jaklitsch, W.J. 1135 (WU 31856). *ibid.*, 27 March 1999, W. Jaklitsch, W.J. 1295 (WU 31857). Hietzing, Lainzer Tiergarten, map grid 7863/1, 28 June 1992, W. Jaklitsch (WU 15204). Liesing, Maurerwald, map grid 7863/1, 10 July 1993, W. Jaklitsch (WU 15037). On *Carpinus orientalis*: CROATIA, Istria, Vrsar, soc. *M. chrysorientalis*, 14 May

2010, H. Voglmayr & W. Jaklitsch, strain MVS (from anamorph) (WU 31858). GREECE, Kerkyra (Corfu), ESE Ano Korakiana, W Ag. Vasilios, 23 April 2012, H. Voglmayr & W. Jaklitsch (WU 32004).

Notes: *Melanconiella spodiaea* is the most common and conspicuous species of the genus in Europe. It is often associated with *Prosthecium ellipsosporum*, which appears to be rarer. *Melanconiella spodiaea* is most similar to *M. decorahensis*, which has dark brown ascospores of similar size, but differs by the absence of ascospore appendages, by pip-shaped conidia, an ectostroma of light to dark grey or brown colour and different hosts (*Betula* spp.). Colonised twigs are often distinctly blackened by numerous dark brown conidia and ascospores. The anamorph-teleomorph connection was repeatedly proven by DNA data from conidial and ascospore cultures.

Several *Melanconium* species have been described, which could represent the conspicuous melanconium-like anamorph of *M. spodiaea*. However, as the original descriptions are very brief and incomplete and as original material is often unavailable for study, these cannot be confidently connected to the currently accepted *Melanconiella* species. In addition, they often consist of mixed elements considering the various hosts listed, and most of these binomials have not been used in recent literature. They should therefore be considered dubious names.

Key to species of *Melanconiella*

Note: Although the anamorph is often important and helpful in the distinction of similar species and is therefore essential for identification, this feature has to be evaluated carefully in species growing on *Carpinus betulus*. In the present study two different species were often found on the same twig, e.g. species with hyaline and dark brown conidia: *M. carpinicola*/*M. chryselanconium* and *M. chryselanconium*/*M. chrysodiscosporina*; or *M. hyperopta*/*M. chrysodiscosporina*, both with hyaline conidia but with different conidial shapes and contents. Ascospores and conidia should be examined in water mounts, as in KOH some features are either quickly disappearing (appendages, hyaline sheath) or less evident (swelling of ascospore walls).

- | | |
|---|------------------------------|
| 1 Ascospores dark brown; conidia dark brown | 2 |
| Ascospores hyaline; conidia hyaline or dark brown | 3 |
| 2(1) Ascospores with hyaline, broad cap-like appendages; conidia usually not distinctly pip-shaped; on <i>Carpinus</i> in Europe | <i>M. spodiaea</i> |
| Ascospores without appendages; conidia pip-shaped; on <i>Betula</i> spp. in Europe and North America | <i>M. decorahensis</i> |
| 3(1) On <i>Carpinus</i> | 4 |
| On other hosts | 12 |
| 4(3) Ascospores with distinctly swelling ascospore wall and with monomorphic, (sub)globose to bullet-shaped ascospore cells mostly wider than 7 µm; commonly with bright yellow ectostroma; in Europe | 5 |
| Ascospores without distinctly swelling ascospore wall and with slightly to distinctly dimorphic cells mostly narrower than 7 µm; ectostroma grey, brownish, cinnamon, cream or yellow | 7 |
| 5(4) Ascospores (16–)19–23(–27.5) × (7.5–)9–12.5(–15.5) µm; always associated with conspicuous conidiomata containing dark brown conidia; on <i>C. betulus</i> in open habitats (parks, hedges, forest edges) | <i>M. chryselanconium</i> |
| Ascospores (15.5–)17–20(–22) × (6–)7–10(–15) µm; with conidiomata containing hyaline conidia | 6 |
| 6(5) On <i>C. betulus</i> ; conidia (12.5–)13.5–16(–19) × 4.5–5.5(–6) µm, l/w=(2.2–)2.5–3.3(–4) | <i>M. chrysodiscosporina</i> |
| On <i>C. orientalis</i> ; conidia (11.5–)12.5–14(–15.5) × (5.5–)6–7(–7.5) µm, l/w=(1.7–)1.9–2.2(–2.7) | <i>M. chrysorientalis</i> |
| 7(4) Ascospores mostly wider than 5.5 and longer than 18.5 µm | 8 |
| Ascospores mostly narrower than 5.5 µm and shorter than 18.5 µm | 10 |

- 8(7) On *Carpinus* in Europe; ectostromatic discs concave, flat or slightly pustulate and little projecting above the perithecial level, whitish to dull yellow or brownish; central column pale yellowish to pale greyish brown or brownish, entostroma inconspicuous, pale, slightly lighter than bark interior 9
- On *C. caroliniana* in North America; ectostromatic discs well-developed, often pulvinate, commonly with distinctly projecting ostioles thus appearing spiny, cream yellow, honey, cinnamon, orange brown to brown; central column young bright olive-yellow, then olive-green to olive-brown; entostroma well-developed, of compacted hyphae, olive-green or olive-brown *M. echinata*
- 9(8) On *C. betulus* *M. hyperopta*
- On *C. orientalis* *M. hyperopta* var. *orientalis*
- 10(7) On *C. betulus* in Europe; in open habitats (parks, hedges, forest edges) *M. carpinicola*
- On *C. caroliniana* in North America 11
- 11(10) Ectostromatic disc mostly irregularly shaped, commonly surrounded by irregular edges or flaps of bark, drab, light to dark grey or dark brown; ostioles mostly marginal in the disc; central column grey or brownish *M. ellisii*
- Ectostromatic discs regularly shaped, mostly circular, angular or ellipsoid, distinctly projecting, often pulvinate, pale or bright yellow to light brown; ostioles mostly regularly arranged in the disc; central column yellow, olive grey or brownish *M. elegans*
- 12(3) On *Corylus*; ascospores broadly fusoid; ascospore ends distinctly subacute with persistent knob-like hyaline appendages *M. flavovirens*
- On *Ostrya*; ascospores fusoid; ascospore ends narrowly rounded to subacute, without or with cap-like appendages 13
- 13(12) Ascospores (20–)23–28(–37.5)×(5–)6.5–8(–9.5) μm; conidia hyaline; on *Ostrya carpinifolia* in Europe *M. meridionalis*
- Ascospores (14.5–)16.5–19(–23)×(4–)5.5–7(–8.5) μm; with dark brown conidia having a light brown equatorial zone; on *Ostrya virginiana* in North America *M. ostryae*

Species excluded from *Melanconiella*

Apart from the species of *Melanconiella* recognised in the current study, most names described or combined in the genus are either incompletely known or doubtful members of *Melanconiella*. As we have not been able to study material of some taxa, the list below contains only species for which we have some evidence. The account also includes taxa that were previously synonymised with *M. chrysostroma* (fide Wehmeyer 1941) and now found to be unrelated to *Melanconiella* after studying their types.

Diatrype carpinigera Berk. & M.A. Curtis, Grevillea 4:96 (1876).

Synonym: *Melanconis carpinigera* (Berk. & M.A. Curtis) Petr., Sydowia 6:355 (1952).

Based on a rudimentary description, this name was combined into *Diaporthe* by Saccardo (1883), subsequently synonymised with *Melanconis chrysostroma* var. *ellisii* by Wehmeyer (1941) and combined into *Melanconis* by Petrak (1952b). Re-examination of the type (K(M) 171592) showed that it is not a diaporthean fungus; stroma morphology and shape and size (18.5–22.5×8.5–11.5 μm) of its brown, two-celled ascospores rather suggest a *Valsaria* sp.

Melanconiella appendiculata (G.H. Otth) Sacc., Syll Fung 11:XXIX (1895).

This species is currently classified as *Prosthecium appendiculatum* (G.H. Otth) M.E. Barr and is phylogenetically unrelated to *Melanconiella* (unpubl. data).

Melanconiella chrysostroma (Fr.) Sacc., Syll Fung 1:741 (1882).

Basionym: *Valsa chrysostroma* Fr., Sum Veg Scand:412 (1849).

[= *Sphaeria xanthostroma* Mont. & Fr. (1834), non Schwein. (1832), non J.C. Schmidt (1817)].

Synonym: *Melanconis chrysostroma* (Fr.) Tul., Selecta fung carpol (Paris) 2:125 (1863).

Melanconiella chrysostroma is based on *Valsa chrysostroma*, which was established by Fries (1849) as a new name for the homonym *Sphaeria xanthostroma* Mont. & Fr. (Montagne 1834), non Schwein. (von Schweinitz 1832). However, *Sphaeria xanthostroma* Mont. & Fr. is the basionym of *Wuestneia xanthostroma* (Reid and Booth 1989) and represents a taxon differing from *Melanconiella* by unicellular ascospores. No morphological description was provided by Fries (1849). Tulasne and Tulasne (1863), who apparently did not know *Wuestneia xanthostroma*, considered the description and illustrations of Montagne (1834: pl. 12, Fig. 4) to be unreliable (“de peritheciorum forma et thecarum fecunditate non fida”), misapplied the name for a taxon with two-celled ascospores and combined the name into *Melanconis*, explicitly citing *Sphaeria xanthostroma* Mont. as a synonym and therefore following the nomenclatural concept of Fries (1849). Because the basionym, *Valsa chrysostroma*, is based on the type and description of *Sphaeria xanthostroma* Mont., *Melanconis chrysostroma* is an obligate synonym of *Wuestneia xanthostroma*, although the concept of Tulasne and Tulasne (1863) applies to a

different taxon belonging to *Melanconiella*. Their description does not fit a single *Melanconiella* species, but is rather based on a mixture of several taxa. Tulasne and Tulasne (1863) record both brown, ovate conidia, 13–15 × 10 µm, as well as biguttulate hyaline conidia, 10–13 × 3.5–5 µm. Their descriptions, measurements and illustrations could be based on a mixture of *M. chrysodiscosporina* (with hyaline biguttulate conidia) and *M. chryso-melanconium* (with brown conidia), which agrees with the illustration of biguttulate ascospores. However, they do not give ascospore sizes, but state that they are of similar width as the hyaline conidia, and they record a biserial ascospore arrangement, features which are not in line with the former two species, but rather imply *M. hyperopta* or *M. carpinicola*. As we did not have the opportunity to examine the Tulasne collections, their concept of *Melanconis chrysostroma* remains obscure.

Melanconiella corni (Wehm.) Petr., Sydowia 6:15 (1952).

This species is currently classified as *Hapalocystis corni* (Wehm.) M.E. Barr and is phylogenetically unrelated to *Melanconiella* (unpubl. data).

Melanconiella leucostroma (Niessl & Rehm) Sacc., Syll Fung 2: Addenda Vol I:LIV (1883).

According to Wehmeyer (1941) this name is a synonym of *Cryptodiaporthe galariculata*.

Melanconiella meschuttii (Ellis & Everh.) Berl. & Voglino, in Saccardo, Syll Fung, Addit. I–IV:129 (1886).

According to Wehmeyer (1941) this name is a synonym of *Melanconis nigrospora*, now classified as *Chapeckia nigrospora* (Peck) M.E. Barr.

Melanconiella nigrospora (Peck) Dearn. & House, Bull N Y State Mus 266:81 (1925).

This is a synonym of *Chapeckia nigrospora*, which is unrelated to *Melanconiella* (Fig. 1).

Melanconiella nyssaegena Ellis & Everh., J Mycol 8:18 (1902).

According to Wehmeyer (1941) this name is a synonym of *Massariovalsa sudans* (Berk. & M.A. Curtis) Sacc.

Melanconiella obruta (Ellis & Everh.) Sacc. & P. Syd., Syll Fung 16:509 (1902).

According to Wehmeyer (1941) this name is a *nomen dubium* which cannot be interpreted because the type collection contains only a melanconium-like anamorph.

Melanconis platystroma Wehm., Mycologia 29:613 (1937).

Melanconis platystroma was described from *Carpinus caroliniana* in Michigan and included in Wehmeyer

(1941). Although it may belong in *Melanconiella*, it is unlike any species included here and differs in having truncated, grey to yellow grey ectostromatic discs with scarcely visible or barely erumpent ostioles, large, brown ascospores (25–31 × 12.5–14.5 µm) with a distinctly greenish tint when fresh, and large (20–31 × 16–18 µm), brown conidia (Wehmeyer 1937, 1941). Wehmeyer (1937) noted that he was unable to germinate the ascospores and conidia despite numerous attempts, which is atypical for species of *Melanconiella*, conidia and ascospores of which germinate easily in pure culture. Wehmeyer (1941) noted that its stromata closely match those of *Massariovalsa sudans* which, according to our experience, is also difficult to grow. Apparently known only from the type collection, this species cannot be appropriately classified until fresh material becomes available.

Melanconiella stellata Petch, Ann Roy Bot Gard Peradeniya 7:306 (1922).

According to Wehmeyer (1941) this name may be a species of *Valsaria*.

Valsa bitorulosa Berk. & Br., Ann Mag Nat Hist Ser 3, 3:367 (1846).

Synonym: Diaporthe bitorulosa (Berk. & Broome) Sacc., Syll Fung 1: 608 (1882).

Based on a scanty description, this name was combined into *Diaporthe* by Saccardo (1882) and subsequently synonymised with *Melanconis chrysostroma* by Wehmeyer (1937, 1941). Re-examination of the type (K(M) 171593) showed that it is not a diaporthean fungus; stroma morphology and shape and size (19–20 × 5.5–7 µm) of its brown, four-celled ascospores rather suggest a *Thyridaria* sp.

Acknowledgements We thank Walter Gams for hospitality and excursion support in Italy; Jacques Fournier, Enrique Rubio Domínguez, Sven-Åke Hanson and Larissa Vasilyeva for collecting and communicating *Melanconiella* specimens; Irmgard Greilhuber and her family for organising and participating in numerous collecting trips together with HV; the fungarium curators of B, BPI, DAOM, G, GZU, K, M, NY, UPS and W for the loan of specimens; Scott Redhead (DAOM) for providing notes of L. E. Wehmeyer and for allowing DNA extraction from the type specimen of *M. echinata*; Walter Till (WU) for managing the herbarium loans; and the British Mycological Society for invitation to the BMS Spring Foray 2011 in Yorkshire.

References

- Barr ME (1978) The *Diaportheales* in North America, with emphasis on *Gnomonia* and its segregates. Mycol Mem 7:1–232
- Castlebury LA, Rossman AY, Jaklitsch WJ, Vasilyeva LN (2002) A preliminary overview of the *Diaportheales* based on large subunit nuclear ribosomal DNA sequences. Mycologia 94:1017–1031

- Chaverri P, Samuels GJ (2003) *Hypocrea/Trichoderma* (Ascomycota, Hypocreales, Hypocreaceae): species with green ascospores. *Stud Mycol* 48:1–116
- Clements FE, Shear CL (1931) Genera of fungi, 2nd edn. H.W. Wilson, New York
- de Hoog GS, Gerrits van den Ende AHG (1998) Molecular diagnostics of clinical strains of filamentous basidiomycetes. *Mycoses* 41:183–189
- De Silva H, Castlebury LA, Green S, Stone JK (2009) Characterisation and phylogenetic relationships of *Anisogramma virgultorum* and *A. anomala* in the *Diaporthales* (Ascomycota). *Mycol Res* 113:73–81
- Dennis RWG (1968) British Ascomycetes. J. Cramer, Stuttgart
- Druzhinina IS, Kubicek CP, Komoń-Zelazowska M, Mulaw TB, Bissett J (2010) The *Trichoderma harzianum* demon: complex speciation history resulting in coexistence of hypothetical biological species, recent agamospecies and numerous relict lineages. *BMC Evol Biol* 10:94
- Edgar RC (2004) MUSCLE: multiple sequence alignment with high accuracy and high throughput. *Nucleic Acids Res* 32:1792–1797
- Ellis MB, Ellis JP (1997) Microfungi on land plants. An identification handbook. Richmond Publishing, Slough
- Ellis JB, Everhart BM (1892) North American pyrenomycetes. Ellis & Everhart, Newfield
- Fries EM (1849) Summa Vegetabilium Scandinaviae. Sectio posterior. 259–572. A. Bonnier, Stockholm, Leipzig
- Fuckel KWGL (1870) Symbolae Mycologicae. Beiträge zur Kenntnis der Rheinischen Pilze. Jahrb Nassau Ver Naturkd 23–24:1–459
- Fuckel KWGL (1871) Symbolae Mycologicae. Erster Nachtrag. Jahrb Nassau Ver Naturkd 25–26:289–346
- Fuckel KWGL (1874) Symbolae Mycologicae. Zweiter Nachtrag. Jahrb Nassau Ver Naturkd 27–28:1–99
- Gazis R, Rehner S, Chaverri P (2011) Species delimitation in fungal endophyte diversity studies and its implications in ecological and biogeographic inferences. *Mol Ecol* 20:3001–3013
- Gerhardt E, Hein B (1979) Die nomenklatorischen Typen der von Th. Nitschke beschriebenen Arten im Pilzherbar des Botanischen Museums Berlin-Dahlem. *Willdenowia* 9:313–329
- Hall TA (1999) BioEdit: a user-friendly biological sequence alignment editor and analysis. program for Windows 95/98/NT. *Nucleic Acids Symp Ser* 41:95–98
- Huelsenbeck JP, Ronquist F (2001) MRBAYES: Bayesian inference of phylogenetic trees. *Bioinformatics* 17:754–755
- Jaklitsch WM (2009) European species of *Hypocrea* part I. The green-spored species. *Stud Mycol* 63:1–91
- Jaklitsch WM (2011) European species of *Hypocrea* part II. species with hyaline ascospores. *Fungal Divers* 48:1–250
- Jaklitsch WM, Voglmayr H (2004) *Hapalocystis occidentalis* - a new species of *Diaporthales* from North America and a key to the species of *Hapalocystis*. *Stud Mycol* 50:229–234
- Jaklitsch WM, Voglmayr H (2011) *Nectria eustromatica* sp. nov., an exceptional species with a hypocreaceous stroma. *Mycologia* 103:209–218
- Jaklitsch WM, Komon M, Kubicek CP, Druzhinina IS (2006) *Hypocrea voglmayrii* sp. nov. from the Austrian Alps represents a new phylogenetic clade in *Hypocrea/Trichoderma*. *Mycologia* 97:1365–1378 [‘2005’]
- Jaklitsch WM, Stadler M, Voglmayr H (2012) Blue pigment in *Hypocrea caerulea* sp. nov. and two additional new species in sect. *Trichoderma*. *Mycologia* 104(4):17
- Katoh K, Toh H (2008) Recent developments in the MAFFT multiple sequence alignment program. *Brief Bioinform* 9:286–298
- Katoh K, Misawa K, Kuma K, Miyata T (2002) MAFFT: a novel method for rapid multiple sequence alignment based on fast Fourier transform. *Nucleic Acids Res* 30:3059–3066
- Kobayashi T (1970) Taxonomic studies of Japanese *Diaporthaceae* with special reference to their life-histories. *Bull Gov Forest Exp Sta* 226:1–242
- Liu YL, Whelen S, Hall BD (1999) Phylogenetic relationships among ascomycetes: evidence from an RNA polymerase II subunit. *Mol Biol Evol* 16:1799–1808
- Mejía LC, Castlebury LA, Rossman AY, Sogonov MV, White JF Jr (2011a) A systematic account of the genus *Plagiostoma* (Gnomoniaceae, Diaporthales) based on morphology, host-associations, and a four-gene phylogeny. *Stud Mycol* 68:211–235
- Mejía LC, Rossman AY, Castlebury LA, White JF Jr (2011b) New species, phylogeny, host-associations and geographic distribution of genus *Cryptosporella* (Gnomoniaceae, Diaporthales). *Mycologia* 103:379–399
- Montagne C (1834) Notice sur les plantes cryptogames récemment découvertes en France, contenant aussi l'indication précise des localités de quelques espèces les plus rares de la flore Française. *Ann Sci Nat Bot Sér* 2(1):295–307
- Müller E, von Arx JA (1962) Die Gattungen der didymosporen Pyrenomyceten. *Beitr Kryptogamenfl Schweiz* 11(2):1–922
- Munk A (1957) Danish pyrenomycetes. A preliminary flora. *Dansk Bot Ark* 17:1–491
- Nylander JA, Wilgenbusch JC, Warren DL, Swofford DL (2008) AWTY (are we there yet?): a system for graphical exploration of MCMC convergence in Bayesian phylogenetics. *Bioinformatics* 24:581–583
- Oth G (1869) Sechster Nachtrag zu dem in Nr. 15–23 der Mittheilungen enthaltenen Verzeichnisse schweizerischer Pilze. *Mitth Natforsch Ges Bern* 1868:37–70
- Pavlic D, Slippers B, Coutinho TA, Wingfield MJ (2009) Multiple gene genealogies and phenotypic data reveal cryptic species of the *Botryosphaeriaceae*: a case study on the *Neofusicoccum parvum*/*N. ribis* complex. *Mol Phyl Evol* 51:259–268
- Petrak F (1952a) Fungi beltsvillenses. III. *Sydowia* 6:5–16
- Petrak F (1952b) Fungi beltsvillenses. IV. *Sydowia* 6:352–360
- Petrak F (1962) Mykologische Beiträge zur österreichischen Flora. *Sydowia* 16:155–198
- Reid J, Booth C (1989) On *Cryptosporella* and *Wuestneia*. *Can J Bot* 67:879–908
- Riethmüller A, Voglmayr H, Göker M, Weiß M, Oberwinkler F (2002) Phylogenetic relationships of the downy mildews (*Peronosporales*) and related groups based on nuclear large subunit ribosomal DNA sequences. *Mycologia* 94:834–849
- Saccardo PA (1876) Fungi Veneti novi vel critici. Series V. *Nuovo Giornale Botanico Italiano* 8:161–211
- Saccardo PA (1882) Sylloge Fungorum 1. Padova: published by the author
- Saccardo PA (1883) Sylloge Fungorum 2. Padova: published by the author
- Saccardo PA (1884) Sylloge Fungorum 3. Padova: published by the author
- Schoch CL, Seifert KA, Huhndorf S, Robert V, Spouge JL, Levesque CA, Chen W et al. (2012) Nuclear ribosomal internal transcribed spacer (ITS) region as a universal DNA barcode marker for *Fungi*. *Proc Natl Acad Sci USA*, www.pnas.org/cgi/doi/10.1073/pnas.1117018109
- Sieber TN (2007) Endophytic fungi in forest trees: Are they mutualists? *Fungal Biol Rev* 21:75–89
- Silvestro D, Michalak I (2011) raxmlGUI: a graphical front-end for RAXML. *Org Divers Evol*. doi:10.1007/s13127-011-0056-0
- Stamatakis E (2006) RAXML-VI-HPC: maximum likelihood-based phylogenetic analyses with thousands of taxa and mixed models. *Bioinformatics* 22:2688–2690
- Sutton BC (1980) The Coelomycetes. Commonwealth Mycological Institute, Kew

- Swofford DL (2002) PAUP*: Phylogenetic Analysis Using Parsimony (*and Other Methods), Version 4.0b10. Sinauer Associates, Sunderland, Massachusetts
- Thiers B (2012) Index Herbariorum: a global directory of public herbaria and associated staff. New York Botanical Garden's Virtual Herbarium. <http://sweetgum.nybg.org/ih/>
- Tulasne LR, Tulasne C (1863) *Selecta Fungorum Carpologia*, vol. 2. Paris
- Vasilyeva LN, Stephenson SL (2010) Biogeographical patterns in pyrenomycetous fungi and their taxonomy. 1. The Grayan disjunction. *Mycotaxon* 114:281–303
- Vilgalys R, Hester M (1990) Rapid genetic identification and mapping of enzymatically amplified ribosomal DNA from several *Cryptococcus* species. *J Bacteriol* 172:4238–4246
- Voglmayr H, Jaklitsch WM (2008) *Prosthecium* species with *Stegosporium* anamorphs on *Acer*. *Mycol Res* 112:885–905
- Voglmayr H, Jaklitsch WM (2011) Molecular data reveal high host specificity in the phylogenetically isolated genus *Massaria* (*Ascomycota*, *Massariaceae*). *Fungal Divers* 46:133–170
- von Höhnelt F (1918) *Mycologische Fragmente*. *Ann Mycol* 16:35–174
- von Schweinitz LD (1832) Synopsis fungorum in America boreali media degentium. *Trans Am Philos Soc II* 4:141–316
- Wehmeyer LE (1926) Cultural life histories of *Melanconis* and *Pseudovalsa*. *Mycologia* 18:257–273
- Wehmeyer LE (1933) The genus *Diaporthe* Nitschke and its segregates. *Univ Michigan Stud Sci Ser* 9:1–349
- Wehmeyer LE (1937) Studies of certain species of *Melanconis* on *Carpinus*, *Ostrya* and *Corylus*. *Mycologia* 29:599–617
- Wehmeyer LE (1941) A revision of *Melanconis*, *Pseudovalsa*, *Prosthecium* and *Titania*. *Univ Michigan Stud, Sci Ser* 14:1–161
- Werle E, Schneider C, Renner M, Völker M, Fiehn W (1994) Convenient single-step, one tube purification of PCR products for direct sequencing. *Nucleic Acids Res* 22:4354–4355
- White TJ, Bruns T, Lee S, Taylor J (1990) Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis MA, Gelfand DH, Sninsky JJ, White TJ (eds) *PCR protocols: a guide to methods and applications*. Academic, San Diego, pp 315–322