

1 **Rare and undersampled dimorphic basidiomycetes**

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42 **Abstract**

43 The diversity of yeasts has grown rapidly as the discovery of new species has benefited from
44 intensified sampling and largely improved identification techniques. An environmental study
45 typically reports the isolation of yeast species, some of which are new to science. Rare species
46 represented by a few isolates often do not result in a taxonomic description. Nucleic acid
47 sequences from these undescribed yeasts remain in public sequence databases, often without a
48 proper taxonomic placement. This study presents a constrained phylogenetic analysis for
49 many rare yeasts from unpublished but publicly available DNA sequences and from studies
50 previously conducted by the authors of this work. We demonstrate that single isolates are an
51 important source of taxonomic findings such as including new genera and species.
52 Independent surveys performed during the last 20 years on a large geographic scale yielded a
53 number of single strains, which were proved to be conspecific in the phylogenetic analyses
54 presented here. The following new species were resolved and described: *Vustinia terrea*
55 Kachalkin, Turchetti & Yurkov gen. nov. et sp. nov., *Udeniomyces caspiensis* Kachalkin sp.
56 nov., *Udeniomyces orazovii* Kachalkin sp. nov., *Tausonia rosea* Kachalkin sp. nov.,
57 *Itersonilia diksonensis* Kachalkin sp. nov., *Krasilnikovozyma fibulata* Glushakova &
58 Kachalkin, *Kwoniella fici* Turchetti sp. nov., *Heterocephalacria fruticeti f.a.* Carvalho, Roehl,
59 Yurkov & Sampaio sp. nov., *Heterocephalacria gelida f.a.* Turchetti & Kachalkin sp. nov.,
60 *Heterocephalacria hypogea f.a.* Carvalho, Roehl, Yurkov & Sampaio sp. nov.,
61 *Heterocephalacria lusitanica f.a.* Inacio, Carvalho, Roehl, Yurkov & Sampaio sp. nov.,
62 *Piskurozyma arborea* Yurkov, Kachalkin, Mašínová & Baldrian sp. nov., *Piskurozyma*
63 *silvicultrix* Turchetti, Mašínová, Baldrian & Yurkov sp. nov., *Piskurozyma stramentorum*
64 Yurkov, Mašínová & Baldrian sp. nov., *Naganishia nivalis* Turchetti sp. nov., *Yurkovia*
65 *nerthusi* Yurkov & Begerow, sp. nov. In addition, two new combinations were proposed
66 *Krasilnikovozyma curviuscula* (Babeva, Lisichkina, Reshetova & Danilevich) Yurkov,
67 Kachalkin & Sampaio comb. nov., *Hannaella taiwanensis* (F.L. Lee & C.H. Huang) Yurkov
68 comb. nov. The order Cyphobasidiales T. Spribille & H. Mayrhofer is rejected in favour of
69 the older name Erythrobasidiales R. Bauer, Begerow, J.P. Sampaio, M. Weiss & Oberwinkler.
70 Other potential novel species identified in this paper await future description. Phylogenetic
71 placement of yet unpublished sequences is believed to facilitate species descriptions and
72 improve classification of yeasts from environmental sequence libraries.

73 **Keywords:** 1 new genus, 17 new species, 2 combinations, yeasts, taxonomy,
74 Tremellomycetes, Pucciniomycotina, Microbotryomycetes, Cystobasidiomycetes

75 Introduction

76 Our knowledge of a yeast stage of members of the phylum Basidiomycota dates back to the
77 19th century and [the](#) pioneering work of Brefeld, who observed the germination of teliospores
78 of the corn smut *Mycosarcoma maydis* with budding yeast-like cells (reviewed by
79 Oberwinkler 2017). Subsequent mating experiments confirmed the presence of the
80 filamentous-yeast switch in several smuts (reviewed by Oberwinkler 2017). Later, Kluyver
81 and van Niel (1927) pointed to the similarity between forcibly ejected buds of a red yeast
82 belonging to the genus *Sporobolomyces* and basidiospores of basidiomycetes. The most
83 conclusive evidence of a basidiomycete connection was the discovery of mating and a sexual
84 state in strains of *Rhodotorula glutinis* by Banno (1963, 1967), followed by the discoveries of
85 teleomorphs of several yeasts and description of the genera *Filobasidiella*, *Cuniculitrema* and
86 *Bulleromyces*. A number of morphological, biochemical, ultrastructural, and physiological
87 criteria indicated the basidiomycetous affinity of many asexual yeasts. These characters
88 include a positive diazonium blue B reaction, urease activity, enteroblastic mode of budding,
89 presence of ballistoconidia, red carotenoid pigments, a lamellate cell wall ultrastructure,
90 presence of a dolipore septum (in hyphae), the biochemical composition of the cell wall, and a
91 high GC content of genomic DNA (Boekhout et al. 2011; Kurtzman and Boekhout 2017).
92 However, evolutionary relationships between yeasts, whether asexual or sexual, and
93 teleomorphic filamentous taxa remained unknown. Early molecular evolutionary studies of 5S
94 rRNA distinguished two phylogenetic lineages, Agaricomycotina and Pucciniomycotina, and
95 these results correlated well with septal ultrastructure (reviewed by Kurtzman and Boekhout
96 2017; Oberwinkler 2017). Later, studies of the SSU rRNA gene revealed the third lineage of
97 basidiomycetes, the Ustilaginomycotina.

98 Studies of dimorphic heterobasidiomycetes followed two different directions (reviewed by
99 Begerow et al. 2017). One approach taken by traditional mycologists, who sampled fungi in
100 the field and investigated them in the laboratory. From these studies we know that sexual
101 structures of some mycoparasites (e.g. *Tremella*, *Rhynchogastrema*, *Trimorphomyces*) and
102 plant parasites (e.g. *Microbotryum*, *Mycosarcoma*) germinate with yeast states. Another
103 approach was undertaken by yeast researchers and included mating experiments to obtain
104 teleomorphic states on laboratory media (e.g. *Curvibasidium*, *Leucosporidium*, *Papiliotrema*,
105 and *Rhodosporeidium*) and the subsequent description of the relevant morphological
106 characters, including basidial and hyphal morphology. It [transpired](#) that species commonly
107 considered as yeasts form a sexual cycle *ex situ* and display features previously described as
108 an adaptation to a parasitic lifestyle, for example appressoria, colacosomes and haustoria.

109 Yeasts were among the first fungi for which nucleic acids were sequenced and
110 phylogenetically analysed, and their presence in all three subphyla in Basidiomycota was
111 demonstrated by many molecular phylogenetic studies (reviewed by Kurtzman and Boekhout
112 2017). Pioneering studies from the 1980s showed that sexual and asexual taxa are intermixed
113 in many clades suggesting that morphological dimorphism is a common feature among
114 basidiomycetous yeasts. Yeasts were also the first group of fungi subjected to DNA-
115 barcoding using ribosomal gene fragments such as LSU rRNA gene (Fell et al. 1995, 2000;
116 Begerow et al. 1997; Fonseca et al. 2000; Sampaio 2004) and ITS (Scorzetti et al. 2002).

117 These studies provided a solid background for fast and reliable identification of yeasts and
118 facilitated the discovery of new species. Although the number of known yeast species in the
119 Agaricomycotina and Pucciniomycotina was growing rapidly, the number of available
120 sequences of teleomorphic taxa remained low (Fell et al. 2000; Millanes et al. 2011; Liu et al.
121 2015a; Wang et al. 2015b). Recent changes in fungal taxonomic rules [have resulted](#) in a
122 unified classification system for naming sexual and asexual taxa, including lineages
123 containing yeasts. Consequently, large polymorphic and polyphyletic anamorphic genera like
124 *Bullera*, *Cryptococcus*, *Rhodotorula*, *Sporobolomyces* and *Trichosporon* were reclassified,
125 which resulted in 35 new genera and 296 new combinations (Liu et al. 2015a; Wang et al.
126 2015b). Similarly, several teleomorphic species of *Cystobasidium*, *Tremella* and *Syzygospora*
127 distantly related to clades that contain type species were reclassified (Liu et al. 2015a; Wang
128 et al. 2015b; Millanes et al. 2016; Spirin et al. 2018). These studies demonstrated that
129 undersampling and the lack of reference sequences hamper both taxonomical works and
130 biodiversity assessments. It has been also shown that the lack of reference sequences and
131 inaccessibility of material (culture or specimen) can result in taxonomic redundancy, as
132 exemplified by the yeast genus *Bandoniozyma* (Tremellomycetes), which coincided with
133 *Rhynchogastrema*, an earlier discovered, but not sequenced, filamentous genus (Liu et al.
134 2015a).

135 To date, most of our knowledge of diversity in Cystobasidiomycetes, Microbotryomycetes
136 and Tremellomycetes is derived from studies reporting the discovery of new yeast species
137 (Liu et al. 2015; Wang et al. 2015; Kurtzman and Boekhout 2017). Yeasts are isolated
138 worldwide from a multitude of habitats and substrates (Peter et al. 2017). Many of them
139 remain, however, undescribed because of an unclear phylogenetic placement or because they
140 are known from a limited number of isolates (e.g., Seifert and Rossman 2010). Constrained
141 phylogenetic analyses of LSU rRNA proved their usefulness and identified potential new
142 species clustering in already recognized (sometimes monotypic) lineages (Liu et al. 2015a;
143 Wang et al. 2015b). A few from these potential new species have been described recently
144 (e.g., Yurkov et al. 2016a), and others still await a description in spite of having been
145 discovered for a long time. Moreover, every new biodiversity study adds a few potential novel
146 yeasts to the already existing pool of unnamed and unplaced species. Sometimes the number
147 of potential novel yeasts reported in a study can be as large as 25-30% of the total diversity
148 (e.g., Yurkov et al. 2012a, 2016b; Mašínová et al. 2017a, 2017b). Thus, it becomes
149 challenging for researchers to [maintain](#) an overview [of the](#) ever-growing diversity of
150 undescribed yeasts.

151 The aim of this paper is to provide updated phylogenies of yeasts in Agaricomycotina and
152 Pucciniomycotina, and identify new clades in these groups. The work includes previously
153 isolated but yet undescribed species and more recent isolates obtained from a number of
154 studies performed by us. Many of these species belong to the so-called heterobasidiomycetes,
155 a group of fungi extensively studied by Franz Oberwinkler and his collaborators, including
156 Robert Bauer and Robert Bandoni. Also, several strains from the former collection of the
157 University of Tübingen served a reference for teleomorphic taxa for which no sequence was
158 available. Several new species and one new genus identified in this study are described.

159 **Material and Methods**

160 Sampling and isolation of yeasts have been independently performed in previous studies.
161 Isolations from soils in Czech Republic, Germany and Portugal followed protocols described
162 by Mašíňová et al. (2017b) and Yurkov et al. (2012a, 2016b), respectively. Strains from
163 Russia were obtained using sampling and isolation protocols described by Glushakova et al.
164 (2015). Yeasts on plant material in Portugal were studied by Inácio et al. (2002) and Inácio
165 (2003). Isolations from glacial sampling and plant materials in Italy followed protocols
166 described in Turchetti et al. (2013) and Franca et al. (2016).

167 Sequences of D1/D2 domains of the 26S rRNA gene (LSU) were aligned into [multiple](#)
168 [sequence alignments](#), previously used by Liu et al. (2015a) and Wang et al. (2015b), by
169 utilising MAFFT online service (version 7) and `--add` and `--keeplength` functions (Katoh et al.
170 2017). New sequences were either obtained from public databases or produced in previous
171 studies. Resulting alignments contained a total of [860](#) sequences, 301 for Pucciniomycotina
172 and [559](#) for Agaricomycotina. Trees were constructed with raxmlGUI 1.5b software using the
173 maximum-likelihood algorithm and GTRGAMMA (GTR substitution model with gamma-
174 distributed rate heterogeneity) model with 100 bootstrap replicates; topological constraints
175 were enforced as was described previously by Liu et al. (2015a) and Wang et al. (2015b). A
176 parsimony network was constructed from aligned LSU and ITS sequences with the program
177 TCS 1.21 (Clement et al. 2000) using a 95% connection limit and gaps treated as missing
178 data. A fragment of the gene encoding translation elongation factor 1 alpha (*TEF1*) was
179 amplified and sequenced following the previously described protocols (Yurkov et al. 2015a,
180 2015b; Spirin et al. 2018). Multiple sequence alignments were performed with the genomic
181 sequences using online version of MAFFT algorithm (Katoh et al. 2017). Phylogenetic
182 relationships were inferred from a concatenated dataset of the ITS region, LSU rRNA and
183 *TEF1* genes as described above.

184 Assimilation tests were performed on solid and in liquid media following the procedures
185 described by Kurtzman et al. (2011), with API 50CH test strips (bioMérieux) and YT and FF
186 MicroPlates (Biolog) as described previously (Yurkov et al. 2017; Mašíňová et al. 2018). For
187 observing micro-morphology, cultures were grown at 10-22°C on YM, PD, GPY and 0.5%
188 glucose-YNB agars and studied with phase-contrast optics.

189

190 **Results and discussion**

191 Phylogenetic analyses included [860](#) sequences of which [185](#) were not studied by Liu et al.
192 (2015a) [or](#) Wang et al. (2015b). The overall topology was consistent with the two previous
193 constrained LSU analyses (Liu et al. 2015a; Wang et al. 2015b), although several lineages
194 were enlarged with potential novel species (Figs. 1, 2). We discuss some of these clades
195 below.

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198

199 **Tremellomycetes**

200 **Cystofilobasidiales**

201 The order Cystofilobasidiales comprises two families, Mrakiaceae and Cystofilobasidiaceae
202 (Liu et al. 2015a). The study of Liu et al. (2015a) indicated a number of sequences
203 corresponding to potential new species in the genera *Itersonia*, *Krasilnikovozyma*, *Tausonia*
204 and *Udeniomyces*. Also, two novel *Cystofilobasidium* species were recently described from
205 Mediterranean soils (Pontes et al. 2017). Three yeasts isolated from cold soils in Russia
206 (Altay region), Italy (Alps) and Kyrgyzstan (Byshkek) showed highly similar sequences and
207 were placed close to the genus *Krasilnikovozyma* (Fig. 3). Our phylogenetic analyses based
208 on LSU, ITS and *TEF1* suggest these yeasts are likely to represent a new genus in Mrakiaceae
209 (see Taxonomy below).

210 The genus *Krasilnikovozyma* was proposed by Liu et al. (2015a) to accommodate members of
211 the so-called *Cryptococcus huempii* clade, which also included the species *Mrakia*
212 *curviuscula* and *Cryptococcus tahquamenonensis*. *C. huempii* and *M. curviuscula* were
213 sequenced from the material available at the CBS culture collection (strains CBS 8168 and
214 CBS 9136, respectively) and showed identical D1/D2 sequences (GenBank AF189844 and
215 EF118826, respectively) suggesting these yeasts belong to the same species. ITS sequence of
216 *Mrakia curviuscula* strain CBS 9136 was first published by Liu et al. (2015b). Because both
217 LSU (see above) and ITS (GenBank KY103891 and KY103892, respectively) sequences of
218 the two species were identical, *Mrakia curviuscula* was considered to be a taxonomic
219 synonym of *Krasilnikovozyma huempii* by Liu et al. (2015a, 2015b).

220 *Cryptococcus tahquamenonensis* (presently *Krasilnikovozyma tahquamenonensis*) was
221 described based on a single strain from soil; its LSU sequence (KM408125) shows 98%
222 similarity to the type strain of *Cryptococcus huempii* (AF189844) and differs in 9
223 substitutions (including two N base calls in and 1 gap in KM408125 using GenBank Blastn).
224 The similarity of the ITS sequences of the two species is 92% (GenBank KM384610 and
225 AF444322), thus allowing a proper differentiation of these yeasts with either LSU or ITS
226 sequencing.

227 The type material of *Mrakia curviuscula* strain Oz-358 was preserved in the yeast collection
228 of the Lomonosov Moscow State University as KBP Y-3618 (holotype) and deposited (ex-
229 type) in the VKM collection as VKM Y-2953 (Babjeva et al. 2002). The original culture Oz-
230 358, from which the holotype Oz-358 was derived, was revived in 2003 and sub-cultured,
231 following a request from the Portuguese Yeast Culture Collection. The progeny of Oz-358
232 was preserved in and later transferred to PYCC as PYCC 5836. Sequencing, performed
233 independently, of cultures derived from the type material, i.e. VKM Y-2953 (GenBank
234 MK244628) and PYCC 5836 (GenBank MF372143, MF372124), demonstrated that the two
235 strains share identical LSU and ITS sequences, which showed high similarity to sequences of
236 the type strain of *Krasilnikovozyma (Cryptococcus) tahquamenonensis* (ITS: KM384610,
237 LSU: KM408125). Specifically, comparison of ITS sequences showed 3 mismatches in ITS2,
238 corresponding to 1 base pair difference and two gaps. Our results suggest that the presumptive
239 ex-type CBS 9136 is not identical to two other ex-types VKM Y-2953 and PYCC 5836. In

240 our opinion, VKM Y-2953 and PYCC 5836 are the correct ex-type cultures, whereas CBS
241 9136 is a strain of *K. huempfi*. Sequencing of ex-types VKM Y-2953 and PYCC 5836
242 suggests that *M. curviuscula* is conspecific with *K. tahquamenonensis*. However, *Mrakia*
243 *curviuscula* Babeva, Lisichkina, Reshetova & Danilevich (2002) is older than *Cryptococcus*
244 *tahquamenonensis* Q.M. Wang, A.B. Hulfactor, K. Sylvester & C.T. Hittinger (2015) and,
245 thus, has a taxonomic priority. Therefore, we resurrect and recombine *Mrakia curviuscula*
246 Babeva, Lisichkina, Reshetova & Danilevich (2002) as *Krasilnikovozyma curviuscula* comb.
247 nov. and put *Krasilnikovozyma tahquamenonensis* (Q.M. Wang, A.B. Hulfactor, K. Sylvester
248 & C.T. Hittinger) A.M. Yurkov (2015) in taxonomic synonymy with the former (see
249 Taxonomy).

250 A GenBank search resulted in a number of sequences labelled as *Mrakia frigida* and *Mrakia*
251 *gelida*. Many of these strains have ITS sequences that differ from those of type strains of *M.*
252 *frigida* and *M. gelida*. Although nucleotide sequences of the ITS region were successfully
253 used to delimit species in the genus *Mrakia*, these results suggest that additional genes should
254 be used in the future to reassess the most useful criteria for species delimitations in this genus.

255

256 **Tremellales**

257 Family Bulleraceae. [A single-species lineage represented by *Cryptococcus mujuensis* in a 7-](#)
258 [gene phylogeny \(Liu et al. 2015b\) was enlarged with two more species in a LSU-phylogeny](#)
259 [\(Liu et al. 2015a\). These tree species were transferred to the newly erected genus](#)
260 [Fonsecazyma \(Liu et al. 2015a\).](#) This study also indicated that this clade contains four
261 potential new species. Yeasts of this genus have been isolated from diverse regions mostly
262 from plant material (Herzberg et al. 2002; Inácio 2003; Mittelbach et al. 2016; Sylvester et al.
263 2015). The *Tremella* clade I (sensu Millanes et al. 2011) comprises not yet re-classified
264 species of the genera *Tremella* and *Sirobasidium*, some of which are also known from living
265 cultures. We identified two more cultures in public collections, namely a yet undescribed
266 *Tremella* species represented by two cultures, DSM 104578 (Germany, FO 24396) and NBRC
267 32520 (Japan). Another species in this clade, closely related to *Cryptococcus cuniculi pro.*
268 *temp.*, is represented by strain KBP Y-5716 (MH697756). The genus *Pseudotremella* was
269 proposed to accommodate the well-supported clade including *T. moriformis* by Liu et al.
270 (2015a). These authors also reported some inconsistency between sequences of *T. indecorata*,
271 which clustered in *Pseudotremella* and *Tremella* clade I. In addition to the aforementioned
272 inconsistency, we found some heterogeneity between sequences of specimens identified as *P.*
273 *moriformis* (Fig. 1A). Specifically, *Tremella moriformis* specimen UBC F13838 (= RB284b,
274 sometimes wrongly cited as RB2846 and RJB2846) collected by Wells from UC Davis
275 (USA) was sequenced by Chen in Tübingen, Germany (GenBank AF042426, AF042244).
276 Sequences of this specimen were highly similar to another recently collected *Tremella*
277 specimen CWU (MYC) 136 from Kharkiv (Ukraine) and a yeast isolate from Italy ([DBVPG](#)
278 [10729, GenBank MK634540](#)) (Fig. 1A). Phylogenetic analyses demonstrated that they are
279 placed in the genus *Pseudotremella* but very distant from the reference specimen of *P.*
280 *moriformis* UBC F13868 (= CBS 7810, GenBank AF075493, AF444331) used by Liu et al.

281 (2015a). Another sequence representing a potentially conspecific isolate was found in
282 GenBank (MG190052); this yeast was isolated from an insect frass on an olive tree in South
283 Africa.

284 Family Carcinomycetaceae. The mycoparasitic genus *Carcinomyces* (Oberwinkler & Bandoni
285 1982) presently comprises two sexual and one asexual species. Our analysis identified
286 another, yet undescribed, species in this genus isolated from an insect gallery in the USA (Fig.
287 1A).

288 Family Cryptococcaceae. Our analyses revealed several potential new species in the genera
289 *Kwoniella* and *Cryptococcus* from soil (Yurkov et al. 2016b, Mašínová et al. 2018), plant
290 (Glushakova and Kachalkin 2017) and insect sources (Fig. 1).

291 Family Phaeotremellaceae. *Gelidatrema*, a monotypic genus proposed by Liu et al. (2015a),
292 was expanded with an additional in our phylogenetic analysis (Fig. 1A). The recently
293 described from a microbial mat in the Canadian High Arctic *Gelidatrema psychrophila* was
294 previously observed by Turchetti et al. (2013). This culture, though the sequence was
295 available for in Genbank (KC433781), was not considered in the description by Tsuji et al.
296 (2018).

297 Family Sirobasidiaceae. The genus *Fibulobasidium* is distinguished from closely related
298 *Sirobasidium magnum*, based on its unusual basidium development (Bandoni 1979). The
299 genus received strong support in recent phylogenetic analyses (Liu et al. 2015a). *Sirobasidium*
300 *magnum* (Liu et al. 2015a) and *Sirobasidium japonicum* (Liu et al. 2015b) were placed sister
301 to the genus *Fibulobasidium* (Fig. 1B). In the present study, we identified a new closely
302 related sub-clade represented by seven yeasts (Fig. 1B). These strains originate from diverse
303 habitats and regions, all of which are characterized by a substantial water limitation, i.e. from
304 Mediterranean soils, Greenland sandstone, moss-dominated desert soil crusts, grape berries
305 and floral nectar (Cadez et al. 2010; Selbman et al. 2014; Mittelbach et al. 2015; Yurkov et al.
306 2016b). A sequence of an unspecified *Sirobasidium* sp. (GenBank LC203429) formed a well-
307 supported cluster with the genus *Fibulobasidium* (Fig. 1B), while *S. intermedium* is placed in
308 *Tremella* clade I (Fig. 1A). Delimitation of the two genera will be difficult in the future
309 because the sequence of the type species of the genus *Sirobasidium*, *S. rubrofuscum* (syn. *S.*
310 *sanguineum*, Dämon and Hausknecht 2002), is not yet available.

311 Family Tremellaceae. The genus *Tremella* was re-defined to include only members of the *T.*
312 *mesenterica* clade. A new lineage consisting of three isolates from soils and insect-related
313 sources was identified close to the *Tremella* clade, though statistical support for this
314 placement was low (Fig. 1A).

315 Family Trimorphomycetaceae. The monotypic genus *Sugitazyma* was proposed to
316 accommodate *Bullera miyagiana* (Liu et al. 2015a). Our results show that several yeasts were
317 placed close to *Sugitazyma miyagiana* and *Tremella parmeliarum*, though statistical support
318 for this clustering was low (Fig. 1B). These yeasts were isolated from soils (Czech Republic,
319 Portugal), an insect (Bulgaria) and a plant (Taiwan). Two strains independently isolated in
320 Serra da Arrábida, Portugal (Inácio 2003; Yurkov et al. 2016b) represent potentially new

321 species, which were placed in the genus *Saitozyma*, though statistical support for this
322 placement was low (Fig. 1B).

324 **Filobasidiales**

325 After recent re-classification of *Cryptococcus* species in Filobasidiales, yeasts in this lineage
326 are accommodated in two existing (i.e. *Filobasidium*, *Syzygospora*), three new (i.e.
327 *Goffeauzyma*, *Piskurozyma*, *Solicoccozyma*) and two resurrected (i.e. *Heterocephalacria*,
328 *Naganishia*) genera (Liu et al. 2015a).

329 Family Filobasidiaceae. The genus *Filobasidium* has been restricted to the clades Magnus and
330 Floriforme (sensu Fonseca et al. 2000, 2011; Scorzetti et al. 2002), which contain the type
331 species *F. floriforme* (Liu et al. 2015a). Similarly, Liu et al. (2015a) resurrected the genus
332 *Naganishia*, with the type species *N. globosa*, to accommodate members of the Albidus clade
333 of the order Filobasidiales (sensu Fonseca et al. 2000, 2011; Scorzetti et al. 2002). Sixteen
334 species are presently accepted in the genus considering the latest member, *Naganishia*
335 *qatarensis*, isolated from a hypersaline marine environment (Fotedar et al. 2018). Our
336 analysis revealed a new member of the genus isolated from cold environments. The study by
337 Liu et al. (2015a) revealed four potential new species in the genus *Heterocephalacria*. Our
338 analyses expand the number of yet undescribed species in this genus to eight (Figs. 1, 4).
339 According to the available literature and GenBank records, these species were isolated from
340 Mediterranean soils and plants (Inácio 2003; Yurkov et al. 2016b), supraglacial sediments
341 (Turchetti et al. 2013), boreal swamp plants, tundra plants, and New Zealand soil.

342 Family Piskurozymaceae. The genus *Piskurozyma* was proposed to accommodate the well-
343 supported Cylindricus clade of the order Filobasidiales (Scorzetti et al. 2002; Fonseca et al.
344 2011), a single-species lineage *F. capsuligenum*, and the mycoparasite *Syzygospora sorana*.
345 The analysis performed by Liu et al. (2015a) showed two well-supported sub-clades in the
346 genus, one comprising members of the Cylindricus clade and another one represented by *P.*
347 *fildesensis* and two undescribed species, later described as *P. yama* and *P. tuonelana* (Yurkov
348 et al. 2016b). Our study substantially expanded the size of the genus with seven potentially
349 new species as suggested by phylogenetic analyses (Figs. 1, 5).

351 **Pucciniomycotina**

352 Similarly to studies in Tremellomycetes, recent taxonomic changes in yeasts belonging to
353 Cystobasidiomycetes and Microbotryomycetes were directed towards the re-classification of
354 large polyphyletic genera such as *Bensingtonia*, *Rhodotorula*, and *Sporobolomyces* (Yurkov
355 et al. 2015; Wang et al. 2015b).

356 In the Cystobasidiales, sequencing and subsequent phylogenetic analyses showed that the
357 parasitic lichen-inhabiting teleomorphic genus *Cystobasidium* is polyphyletic (Yurkov et al.
358 2015b; Millanes et al. 2016) and distributed between two clades. The first clade, containing
359 the type species *C. fimentarium*, also includes numerous yeast species from the so-called
360 *Rhodotorula minuta* clade (Yurkov et al. 2015b). The second clade, with *C. hypogymniicola*

β61 and *C. usneicola* is placed outside Cystobasidiales and to *Cyrenella elegans* in the
362 Erythrobasidiales (Fig. 2). It is important to document that *Cyrenella elegans* was not
363 included in the phylogenetic analyses that addressed the taxonomic position of the genus
β64 *Cyphobasidium* (Millanes et al. 2016; Spribille et al. 2016). Below we discuss taxonomy of
365 this genus in more detail. Recent studies expanded *Cystobasidium* and the closely genus
366 *Occultifur* with three and four new yeast species, respectively (Wang et al. 2015b; Šibanc et
367 al. 2018; Turchetti et al. 2018).

368 In the Microbotryomycetes, a number of new genera and species were described since the re-
369 classification of *Bensingtonia*, *Rhodotorula*, and *Sporobolomyces* by Wang et al. (2015b).
β70 Two genera, *Libkindia* and *Yurkovia*, were proposed to accommodate new yeasts from forest
371 soils in the Czech Republic (Mašínová et al. 2017b). The genus *Heitmania* was recently
372 described to include three novel yeasts from plants in China (Liu et al. 2017). Despite being
β73 based on multi-gene analyses, the taxonomic position of these genera remained unclear and
374 they were placed in the Microbotryomycetes as ‘*incertae sedis*’. Wang et al. (2015b) noted
375 that using the signal of the LSU rRNA gene alone is not sufficient to resolve the high-level
376 phylogenetic relationships in Microbotryomycetes. Our analyses indicated two loosely placed
377 clusters in Microbotryomycetes close to genera *Curvibasidium*, *Pseudoleucosporidium* and
378 *Sampaiozyma* (Fig. 2). These weakly supported clades contained yeasts isolated from
379 Germany (forest soil, Yurkov et al. 2016a), Portugal (plant, Inacio 2003) and Russia. A robust
380 multi-gene analysis is required to resolve the phylogenetic relationships and taxonomic
381 position of these yeasts. The constrained LSU rRNA gene phylogenetic analysis performed by
382 Wang et al. (2015 b) failed to resolve placement of several psychrophilic yeasts, including
383 *Rhodotorula svalbardensis* and a few undescribed species. In agreement with this
384 observation, our results showed the unclear placement of CRUB 1733 (GenBank FJ841888)
385 and DBVPG 10048 (GenBank KC433880) close to the filamentous fungus *Camptobasidium*
386 (Fig. 2). The analysis also suggested a potential new species in this yet monotypic genus
387 represented by an antarctic yeast, CBS 8941 (Fig. 2). Two species of *Hamamotoa* and one
388 species of *Colacogloea*, *Leucosporidium* and *Slooffia* were described recently from soils in
389 Europe (Yurkov et al. 2016a; Mašínová et al. 2017b). A few more novelties in genera
390 *Bannozyma* (KY558342, Mašínová et al. 2017b, 2018; this study), *Chrysozyma* (AB552933,
391 KX067789, Endoh et al. 2011; Wang et al. 2015b), *Colacogloea* (EU002850, FN428953,
392 Kachalkin et al. 2008; Wang et al. 2015b), *Fellozyma* (FN868158, Wang et al. 2015b),
393 *Hamamotoa* (AM039679, KU609479, AB462346, EF585181, Wang et al. 2015b; this study),
394 *Oberwinklerozyma* (FN401525, Wang et al. 2015b; Yurkov et al. 2016b), *Slooffia*
395 (AF444728, DQ531946, EF450537, Sampaio et al. 2011; Wang et al. 2015b) and *Yurkovia*
396 (FN428970, Mašínová et al. 2017b) were reported and analysed in previous studies and await
β97 a formal description. [For the first time our](#) analysis also revealed [the](#) diversity of the genus
398 *Yunzhangia* (previously known from two species only) suggesting three novel species close to
399 *Yunzhangia sonckii* (Fig. 2).

400

401

402 **Taxonomy**

403 **Tremellomycetes, Cystofilobasidiales**

404 **Description of *Vustinia* Kachalkin, Turchetti & Yurkov, gen. nov. (MB 829115)**

405 Etymology: the genus is named in honor of the Russian zymologist Dr. Michael Vustin
406 (VKPM culture collection, State Research Institute of Genetics and Selection of Industrial
407 Microorganisms, Russia) for his contributions to the study of soil yeasts and yeasts producing
408 carotenoid pigments.

409 This monotypic genus is proposed to accommodate a new species represented by two isolates,
410 which are phylogenetically close to the genus *Krasilnikovozyma* in a well-supported clade
411 (Fig. 3).

412 Phylogenetic position: Fungi, Dikarya, Basidiomycota, Agaricomycotina, Tremellomycetes,
413 Cystofilobasidiales, Mrakiaceae.

414 Basidiocarps absent. True hyphae and pseudohyphae not observed. Sexual reproduction not
415 observed. Budding cells present. Budding is on the wide basis, polar with sympodial
416 proliferation. Ballistoconidia absent. Arthroconidia absent. Urea hydrolysis and Diazonium
417 Blue B reaction are positive. Nitrate is utilized.

418 Type species: *Vustinia terrae* Kachalkin, Turchetti & Yurkov, sp. nov. (MB 829116)

419 Notes: New species of the genus *Vustinia* can be distinguished from known *Krasilnikovozyma*
420 species by orange-colored pigmentation, which is an important character in Tremellomycetes.
421 This is the third pigmented genus in Cystofilobasidiales after *Cystofilobasidium* and *Phaffia*.
422 The genus *Vustinia* was distant from known *Krasilnikovozyma* species in a concatenated ITS-
423 LSU and ITS-LSU-*TEF1* phylogenies (Figure 3).

424

425 **Description of *Vustinia terrae* Kachalkin, Turchetti & Yurkov, sp. nov. (MB 829116)**

426 Etymology: The species epithet *terrae* is derived from terra (L. gen. sing. f., n., of the earth)
427 and refers to the original substrate of isolation, namely soil.

428 After 1 week at 20 °C on PD, GPY and 2% Glucose YNB agars, streak is orange, butyrous
429 with a smooth glistening surface. Margins are smooth and entire. Cells are ellipsoidal to
430 cylindrical 6–10 × 2–3 µm in size, occurring singly or in pairs, and proliferating by polar
431 budding (Figure 7 a). Lipid-like bodies can be present in cells. Pseudohyphae and true hyphae
432 were not observed. Ballistoconidia were not observed. Teleomorph was not observed.

433 Glucose is not fermented. Positive growth on D-glucose, D-galactose, L-sorbose, D-ribose
434 (weak), D-xylose, L-arabinose, D-arabinose (weak and delayed), L-rhamnose, sucrose,
435 maltose (weak), trehalose, cellobiose, salicin (weak), arbutin (delay for some strains),
436 melibiose (variable, delayed), lactose, raffinose (variable), melezitose (variable), soluble
437 starch (variable, delayed), glycerol, erythritol, ribitol, xylitol (delay), D-mannitol, D-glucitol,
438 2-keto-D-gluconate, 5-keto-D-gluconate, D-gluconate, D-glucuronate (variable), succinate
439 (delayed) and citrate (variable). No growth on D-glucosamine, methyl-alpha-D-glucoside,

440 inulin, inositol, methanol, ethanol, galactitol, DL-lactate, L-malate and ethyl acetate.
441 Utilization of nitrogen sources: positive growth on potassium nitrate, sodium nitrite, lysine
442 and cadaverine. No growth in the presence of 10% sodium chloride, 50% D-glucose and
443 0.01% cycloheximide. Growth on vitamin-free medium is weak. Urea hydrolysis and
444 Diazonium Blue B reaction are positive. Starch-like compounds are produced. Maximum
445 growth temperature: 25 °C

446 Molecular characteristics (holotype): nucleotide sequences of ITS and LSU (D1/D2 domains)
447 rRNA, and partial sequence of *TEF1* deposited in NCBI/EMBL (GenBank) under the
448 accession numbers: MH685196, MH697747 and LS992156, respectively.

449 Deposits: holotype, strain K833b isolated from mountain soil collected in June 2015 near
450 Kosh-Agach village, Republic of Altay, Russia, preserved in a metabolically inactive state in
451 the KBP collection of Department of Soil Biology, Faculty of Soil Sciences, Lomonosov
452 Moscow State University (WDCM 1173) as KBP Y-5245^T. Ex-type cultures are deposited in
453 the All-Russian Collection of Microorganisms (VKM), Pushchino, Russia (VKM Y-3018),
454 the Russian National Collection of Industrial Microorganisms (VKPM), Moscow, Russia
455 (VKPM Y-4321) and the German Collection of Microorganisms and Cell Cultures (DSMZ),
456 Braunschweig, Germany (DSM 105056).

457 Strains studied: K833b (= KBP Y-5245^T); paratypes: DBVPG 10597 from forest soil
458 collected in Kleiner Priol (Montigg), South Tyrol, Alps, Italy (GenBank KU745368,
459 KU745306, MH986600) and KBP Y-5336 (= VKM Y-3019) from soil collected in a park in
460 Bishkek, Kyrgyzstan (GenBank MH697748, LS992158).

461 Notes: New species of the genus *Vustinia* can be distinguished from known *Krasilnikovozyma*
462 species by orange-colored pigmentation, positive growth on glycerol and erythritol and
463 inability to grow on D-glucosamine.

464

465 **Description of *Udeniomyces caspiensis* Kachalkin, sp. nov. (MB 829119)**

466 Etymology: The species epithet *caspiensis* is derived from Latin Caspius (L. adj. m., Caspian)
467 and refers to the region, where the species was isolated.

468 After 1 week at 20 °C on PD, GPY and 2% Glucose YNB agars, streak is pinkish-white, soft
469 with a dull to semi-shiny surface. Margins are smooth and entire. Cells are ovoid to
470 ellipsoidal, 6–11 × 4–5 µm in size, occurring singly or in pairs, and proliferating by polar
471 budding on a broad base ([Figure 7 b](#)). Budding scars may be present. Pseudohyphae and true
472 hyphae were not observed. Ballistoconidia were not observed. Teleomorph was not observed.

473 Glucose is not fermented. Positive growth on D-glucose, D-ribose (weak), L-arabinose, D-
474 arabinose (weak), sucrose, maltose, trehalose, cellobiose, salicin (weak), arbutin (delay for
475 some strains), raffinose, melezitose, glycerol, D-mannitol (weak), D-glucitol (weak), D-
476 glucuronate (weak), D-gluconate (weak), 2-keto-D-gluconate (weak) and 5-keto-D-gluconate.
477 No growth on D-galactose, L-sorbose, D-glucosamine, D-xylose, L-rhamnose, methyl-alpha-
478 D-glucoside, melibiose, lactose, inulin, soluble starch, ethanol, erythritol, ribitol, galactitol,
479 inositol, DL-lactate, succinate and citrate. Utilization of nitrogen sources: positive growth on

480 potassium nitrate and lysine. No growth in the presence of 10% sodium chloride and 50% D-
481 glucose. Growth on vitamin-free medium is negative. Urea hydrolysis and Diazonium Blue B
482 reaction are positive. Starch-like compounds are produced. Maximum growth temperature: 23
483 °C.

484 Molecular characteristics (holotype): nucleotide sequences of ITS and LSU (D1/D2 domains)
485 rRNA deposited in NCBI/EMBL (GenBank) under the accession number MH697745.

486 Deposits: holotype, strain K686-1b isolated from leaves of *Camphorosma sp.*
487 (Amaranthaceae) collected in May 2014 at the Djanybek Research Station of Institute of
488 Forest Science RAS, Volgograd Oblast, Russia, preserved in a metabolically inactive state in
489 the KBP collection of Department of Soil Biology, Faculty of Soil Sciences, Lomonosov
490 Moscow State University (WDCM 1173) as KBP Y-5036^T. Ex-type cultures are deposited in
491 the All-Russian Collection of Microorganisms (VKM), Pushchino, Russia (VKM Y-3016),
492 the CBS yeast collection of the Westerdijk Fungal Biodiversity Institute, Utrecht, the
493 Netherlands (CBS 15357) and the German Collection of Microorganisms and Cell Cultures
494 (DSMZ), Braunschweig, Germany (DSM 106747).

495

496 **Description of *Udeniomyces orazovii* Kachalkin, sp. nov. (MB 829120)**

497 Etymology: The species epithet *orazovii* (NL. gen. sing. masc. n.) is in honor of the
498 mycologist Prof. Hodjanazar Orazov (Institute of Botany of the Academy of Sciences of
499 Turkmenistan) for his contributions to the study of fungi in arid soils. Prof. Orazov also
500 provided the sample from which this species was isolated.

501 After 1 week at 20 °C on PD, GPY and 2% Glucose YNB agars, streak is white, butyrous
502 with a smooth glistening surface. Margins are undulating with some outgrowth in a medium
503 underneath the streak. Cells are sub-globose, ellipsoidal, elongate to cylindrical, 6–15 × 3–6
504 µm in size, occurring singly, in pairs or chains, and proliferating by polar budding on a broad
505 base (Figure 7 d). Ballistoconidia were not observed. Pseudohyphae and short true hyphae
506 occur. Terminal, lateral and intercalary spherical chlamydospores, 18–20 µm in size, with
507 refractile granules from a single cell on true hyphae formed on PDA and 2% Glucose YNB
508 agar after 8-10 d at 20 °C (Figure 7 e, f). Teleomorph was not observed.

509 Glucose is not fermented. Positive growth on D-glucose, L-sorbose (variable), D-glucosamine
510 (variable), D-ribose (weak), D-xylose (variable), L-arabinose, D-arabinose (weak), sucrose,
511 maltose, trehalose, cellobiose, salicin, arbutin, lactose (variable), raffinose, melezitose,
512 soluble starch (weak), glycerol (weak), ribitol (variable), D-glucitol, D-mannitol, inositol
513 (weak), ethanol, 2-keto-D-gluconate, 5-keto-D-gluconate, D-glucuronate (weak), succinate
514 (weak) and citrate (weak). No growth on D-galactose, L-rhamnose, methyl-alpha-D-
515 glucoside, melibiose, inulin, erythritol, galactitol, D-gluconate and DL-lactate. Utilization of
516 nitrogen sources: positive growth on potassium nitrate and lysine. No growth in the presence
517 of 50% D-glucose. Growth on vitamin-free medium is positive. Urea hydrolysis and
518 Diazonium Blue B reaction are positive. Starch-like compounds are produced. Maximum
519 growth temperature: 24 °C.

520 Molecular characteristics (holotype): nucleotide sequences of ITS and LSU (D1/D2 domains)
521 rRNA, and partial sequence of *TEF1* deposited in NCBI/EMBL (GenBank) under the
522 accession numbers: MH734791, MH697744 and LS998026, respectively.

523 Deposits: holotype, strain K515b isolated from leaves of *Halocharis hispida* (Amaranthaceae)
524 collected in May 2013 near Babadurmaz village, Turkmenistan (approx. coordinates 37.65 N,
525 59.15 E), preserved in a metabolically inactive state in the KBP collection of Department of
526 Soil Biology, Faculty of Soil Sciences, Lomonosov Moscow State University (WDCM 1173)
527 as KBP Y-4766^T. Ex-type cultures are deposited in the All-Russian Collection of
528 Microorganisms (VKM), Pushchino, Russia (VKM Y-3014), the CBS yeast collection of the
529 Westerdijk Fungal Biodiversity Institute, Utrecht, the Netherlands (CBS xxxxx) and the
530 German Collection of Microorganisms and Cell Cultures (DSMZ), Braunschweig, Germany
531 (DSM 100168).

532 Strains studied: K515b (= KBP Y-4766^T); paratype KBP Y-4704 (GenBank MH697743) from
533 soil near Babadurmaz village, Turkmenistan.

534

535 **Description of *Tausonia rosea* Kachalkin, sp. nov. (MB 829122)**

536 Etymology: The species epithet *rosea* is derived from roseus (L. adj. f., pink) and refers to the
537 culture color.

538 After 2 weeks at 20 °C on PD, GPY and 2% Glucose YNB agars, streak is light pink,
539 butyrous, with a dull and smooth surface. Margins are smooth and entire. Cells are globose,
540 sub-globose and ovoid, 7–8 × 4.5–7 µm in size, occurring singly or in pairs, and proliferating
541 by polar budding on a broad base (Figure 7 c). Budding scars may be present. Spherical
542 chlamydospore-like cells, 8–10 µm in size, may be present in older culture. Pseudohyphae
543 and true hyphae were not observed. Ballistoconidia were not observed. Teleomorph was not
544 observed.

545 Glucose is not fermented. Positive growth on D-glucose, L-arabinose, D-arabinose, sucrose,
546 maltose, trehalose, cellobiose (weak), salicin, arbutin, raffinose, melezitose, soluble starch
547 (weak), glycerol (weak), ribitol (weak), D-glucitol, D-mannitol, 2-keto-D-gluconate, 5-keto-
548 D-gluconate (weak), D-glucuronate (weak) and ethanol (weak). No growth on D-galactose, L-
549 sorbose, D-glucosamine, D-ribose, D-xylose, L-rhamnose, methyl-alpha-D-glucoside,
550 melibiose, lactose, inulin, erythritol, galactitol, inositol, D-gluconate, DL-lactate, succinate
551 and citrate. Utilization of nitrogen sources: positive growth on potassium nitrate and lysine.
552 No growth in the presence of 10% sodium chloride and 50% D-glucose. Growth on vitamin-
553 free medium is positive. Urea hydrolysis and Diazonium Blue B reaction are positive. Starch-
554 like compounds are produced. Maximum growth temperature: 23 °C.

555 Molecular characteristics (holotype): nucleotide sequences of ITS and LSU (D1/D2 domains)
556 rRNA, and partial sequence of *TEF1* deposited in NCBI/EMBL (GenBank) under the
557 accession numbers: LN871177 and LS998027, respectively.

558 Deposits: holotype, strain K744-1b isolated from leaves of *Salicornia* sp. (Amaranthaceae)
559 collected in August 2014 on the shore of lake Elton, Volgograd Oblast, Russia, (approx.

560 coordinates 49.15 N, 46.68 E), preserved in a metabolically inactive state in the KBP
561 collection of Department of Soil Biology, Faculty of Soil Sciences, Lomonosov Moscow
562 State University (WDCM 1173) as KBP Y-4584^T. Ex-type cultures are deposited in the All-
563 Russian Collection of Microorganisms (VKM), Pushchino, Russia (VKM Y-3007), the CBS
564 yeast collection of the Westerdijk Fungal Biodiversity Institute, Utrecht, the Netherlands
565 (CBS xxxxx) and the German Collection of Microorganisms and Cell Cultures (DSMZ),
566 Braunschweig, Germany (DSM 100302).

567

568 **Description of *Itersonilia diksonensis* Kachalkin, sp. nov. (MB 829123)**

569 Etymology: The species epithet *diksonensis* (NL. gen. sing. masc. n.) is derived from Dikson,
570 the urban-type settlement on Russia's Arctic Ocean coast and refers to the region where the
571 species was isolated.

572 After 2 weeks at 20 °C on GPY and 2% Glucose YNB agars, streak is salmon-pink, butyrous,
573 with a glossy wrinkled surface. Margins are crenulate and fringed with tiny colonies resulting
574 from discharged ballistoconidia. Cells are ellipsoidal, sub-globose to ovoid, 6–9 × 4.5–6 µm
575 in size, occurring singly or in pairs, and proliferating by polar and lateral budding ([Figure 7](#)
576 [g](#)). Ballistoconidia are lunate, 5.5–6 × 3.5–4 µm size, formed abundantly on cells with 1–2
577 occasionally sympodially branching sterigmata ([Figure 7 h](#)). After 2 weeks at 20 °C on PDA,
578 streak is light salmon-pink, mucoid, raised with a smooth and glossy surface. Margin is entire.
579 Cells are ovoid, ellipsoidal or elongate, 6–12 × 5–6 µm in size, occur singly, in pairs or
580 chains, and proliferating by polar budding. Rare ballistoconidia formed at the end of 10–30
581 µm long sterigmata. Pseudohyphae and true hyphae were not observed on any media.
582 Teleomorph was not observed.

583 Glucose is not fermented. Positive growth on D-glucose, L-sorbose, D-ribose, D-xylose, L-
584 arabinose, D-arabinose, L-rhamnose, sucrose, trehalose, cellobiose, salicin, arbutin, raffinose,
585 melezitose (weak), soluble starch (weak), glycerol, ribitol, D-glucitol, D-mannitol, 5-keto-D-
586 gluconate (weak), D-gluconate, D-gluconate (weak), DL-lactate (weak), succinate, citrate
587 and ethanol. No growth on D-galactose, D-glucosamine, maltose, methyl-alpha-D-glucoside,
588 melibiose, lactose, inulin, erythritol, galactitol and inositol. Utilization of nitrogen sources:
589 positive growth on potassium nitrate and lysine. No growth in the presence of 50% D-glucose.
590 Growth on vitamin-free medium is positive. Urea hydrolysis and Diazonium Blue B reaction
591 are positive. Starch-like compounds are not produced. Maximum growth temperature: 36 °C.

592 Molecular characteristics (holotype): nucleotide sequences of ITS and LSU (D1/D2 domains)
593 rRNA, and partial sequence of *TEF1* deposited in NCBI/EMBL (GenBank) under the
594 accession numbers: MH697741, MH734790 and LS998025, respectively.

595 Deposits: holotype, strain K343b isolated from a mixed sample of leaves of flowering arctic
596 plants collected in July 2012 near the settlement Dikson, Taymyr peninsular, Krasnoyarsk
597 Krai, Russia, preserved in a metabolically inactive state in the KBP collection of Department
598 of Soil Biology, Faculty of Soil Sciences, Lomonosov Moscow State University (WDCM
599 1173) as KBP Y-4765^T. Ex-type cultures are deposited in the All-Russian Collection of

600 Microorganisms (VKM), Pushchino, Russia (VKM Y-3013), the Russian National Collection
601 of Industrial Microorganisms (VKPM), Moscow, Russia (VKPM Y-4017), the CBS yeast
602 collection of the Westerdijk Fungal Biodiversity Institute, Utrecht, the Netherlands (CBS
603 xxxxx) and the German Collection of Microorganisms and Cell Cultures (DSMZ),
604 Braunschweig, Germany (DSM 100169).

605

606 **Description of *Krasilnikovozyma fibulata* Glushakova & Kachalkin sp. nov. (MB 829124)**

607 Etymology: The species epithet *fibulata* is derived from fibula (L. adj. f., with clamp) and
608 refers to the presence of hyphae with clamp connections.

609 After 2 weeks at 20 °C on PD, GPY and 2% Glucose YNB agars, streak is cream colored,
610 butyrous with a smooth glistening surface and hyphae produced at the margin. Cells are
611 fusiform, 6–10 × 2–3 µm in size, and occurring singly or in pairs, and proliferating by polar
612 budding (Figure 7 i). Ballistoconidia were not observed. Pseudohyphae and true hyphae with
613 clamp connections occur. Spherical teliospores, 15–16 µm in size, are produced laterally or
614 terminally (Figure 7 j, k). No teliospore germination was observed and basidia morphology
615 remains unknown.

616 Glucose is not fermented. Positive growth on D-glucose, D-galactose, L-sorbose, D-
617 glucosamine, D-xylose, L-rhamnose, sucrose, trehalose, cellobiose, salicin (weak), arbutin,
618 lactose, ethanol (weak), ribitol (weak), D-glucitol, 2-keto-D-gluconate, 5-keto-D-gluconate,
619 citrate (weak) succinate (weak) and ethanol. No growth on L-sorbose, D-ribose, L-arabinose,
620 D-arabinose, maltose, methyl-alpha-D-glucoside, melibiose, raffinose, melezitose, inulin,
621 soluble starch, glycerol, erythritol, D-mannitol, galactitol, inositol, D-gluconate, D-
622 glucuronate and DL-lactate. Utilization of nitrogen sources: positive growth on potassium
623 nitrate and lysine. No growth in the presence of 50% D-glucose. Growth on vitamin-free
624 medium is positive. Urea hydrolysis and Diazonium Blue B reaction are positive. Starch-like
625 compounds are produced. Maximum growth temperature: 24 °C.

626 Molecular characteristics (holotype): nucleotide sequences of ITS and LSU (D1/D2 domains)
627 rRNA, and partial sequence of *TEF1* deposited in NCBI/EMBL (GenBank) under the
628 accession numbers: MH685197, MH697754 and LS992159, respectively.

629 Deposits: holotype, strain A528 isolated from roots of *Impatiens parviflora* (Balsaminaceae),
630 in September 2010 in the Losiny Ostrov (Rus. Elk Island) National Park, Moscow, Russia,
631 preserved in a metabolically inactive state in the KBP collection of Department of Soil
632 Biology, Faculty of Soil Sciences, Lomonosov Moscow State University (WDCM 1173) as
633 KBP Y-5098^T. Ex-type cultures are deposited in the All-Russian Collection of
634 Microorganisms (VKM), Pushchino, Russia (VKM Y-3017), the Russian National Collection
635 of Industrial Microorganisms (VKPM), Moscow, Russia (VKPM Y-3827), the CBS yeast
636 collection of the Westerdijk Fungal Biodiversity Institute, Utrecht, the Netherlands (CBS
637 xxxxx) and the German Collection of Microorganisms and Cell Cultures (DSMZ),
638 Braunschweig, Germany (DSM 105054).

639

640 **Description of *Krasilnikovozyma curviuscula* (Babeva, Lisichkina, Reshetova &**
641 **Danilevich) Yurkov, Kachalkin & Sampaio comb. nov. (MB 829125)**

642 Basionym: *Mrakia curviuscula* Babeva, Lisichkina, Reshetova & Danilevich MB 529873

643 Holotype: KBP Y-3618 isolated from moss *Bryum* sp., dry white moss-pine forest, Oka
644 Nature Reserve, Ryazan Oblast, Russia, preserved in [a](#) metabolically inactive state (dried) in
645 the KBP collection of Department of Soil Biology, Faculty of Soil Sciences, Lomonosov
646 Moscow State University (WDCM 1173).

647 Ex-type cultures are deposited in the All-Russian Collection of Microorganisms (VKM),
648 Pushchino, Russia (VKM Y-2953) and the Portuguese Yeast Culture Collection (PYCC),
649 Caparica, Portugal (PYCC 5836).

650 Molecular characteristics (holotype): nucleotide sequences of ITS and LSU (D1/D2 domains)
651 rRNA deposited in NCBI/EMBL (GenBank) under the accession numbers: MK244628 (VKM
652 Y-2953), and MF372124 and MF372143 (PYCC 5836), respectively.

653 Ecology: The species is widespread. It was found in soil (Japan, Canada, USA, Denmark;
654 GenBank AB462342, KM384610, MH655679, MG914803), in mushroom (Taiwan; GenBank
655 FJ873516), pine forest litter (Russia; GenBank MK244629, MK244630), rotten wood
656 (Argentina), Sphagnum moss (Russia), [and](#) orchid flower (North America; GenBank
657 EU218880) (e.g. Taylor and McCormick 2008, Glushakova et al. 2015, Sylvester et al. 2015).

658

659 **Tremellomycetes, Tremellales**

660 **Description of *Hannaella taiwanensis* (F.L. Lee & C.H. Huang) Yurkov comb. nov. (MB** 661 **829114)**

662 Basionym: *Cryptococcus taiwanensis* F.L. Lee & C.H. Huang, Fungal Science 26 (1): 61
663 (2011); MB 560182

664 Note: The species was overlooked in the analysis by Liu et al. (2015). Sequences D1/D2
665 domains of LSU (HQ591443) of the type strain BCRC 23252 show 97% similarity to
666 *Hannaella zae*, *H. kunmingensis*, *H. siamensis*, and 96% similarity to the type species of the
667 genus *H. sinensis* (Fig. S1).

668

669 **Validation of *Tremella basidiomaticola* X.Z. Liu & F.Y. Bai, MycoKeys 47:80 (2018); MB** 670 **827184**

671 Zhao et al. (2018) described four new *Tremella* species from China. Type material of the
672 three sexual species (*Tremella cheejenii*, *T. erythrina*, and *T. salmonea*) was deposited in the
673 Herbarium of the Chinese Academy of Sciences (acronym HMAS). The species *Tremella*
674 *basidiomaticola* is known from its asexual (yeast) state, and the holotype (a strain) was
675 deposited in the China General Microbiological Culture Collection Center. The description of
676 *Tremella basidiomaticola* does not conform with the ICN Shenzhen Code. According to the
677 Article 40.8 of the ICN Shenzhen Code, for the name of a new species or infraspecific taxon

678 published on or after 1 January 2019 of which the type is a culture, the protologue must
679 include a statement that the culture is preserved in a metabolically inactive state. This
680 requirement was not fulfilled in the original description by Zhao et al. (2018). Therefore, the
681 description is validated here in accordance with the Article 9.2 of the ICN Shenzhen Code.

682 Holotype, strain CGMCC 2.5724^T isolated from a basidioma of *Tremella fuciformis* collected
683 in July 2017 by X.Z. Liu in Gutian county, Ningde city, Fujian Province, China, preserved in
684 a metabolically inactive state at the China General Microbiological Culture Collection Center,
685 Beijing, China. Ex-holotype culture is deposited in the CBS yeast collection of the Westerdijk
686 Fungal Biodiversity Institute, Utrecht, the Netherlands (CBS 15261).

688 Comments to the genus *Gelidatrema*

689 The description of the second species of the genus *Gelidatrema*, *Gelidatrema psychrophila*,
690 was published simultaneously with the preparation of the present manuscript (Tsuji et al.
691 2018). This species was described based on two isolates from a microbial mat in the Canadian
692 High Arctic. Another culture of *Gelidatrema psychrophila* (DBVPG 5459, GenBank
693 KC433781) was isolated from a snow sample in Italian Alps (Turchetti et al. 2013). This
694 culture has not been considered in the description by Tsuji et al. (2018). We compared
695 physiological profiles of *Gelidatrema psychrophila* from the original description (Tsuji et al.
696 2017) with those of the strain DBVPG 5459 (this study). In contrast to results published by
697 (Tsuji et al. 2018), the strain *G. psychrophila* DBVPG 5459 grew on D-gluconate but not on
698 D-glucuronate, N-acetyl-D-glucosamine, and succinate. Assimilation of glycerol and ethanol
699 was weak or negative (Tsuji et al. 2018; this study). Similarly to results obtained for the type
700 species of the genus, *G. spencermartinsiae*, and in contrast to results published by Tsuji et al.
701 (2018), the strain DBVPG 5459 did not assimilate potassium nitrate and sodium nitrite.

702 The two species of the genus *Gelidatrema* (*G. spencermartinsiae* and *G. psychrophila*) are
703 polytrophic utilizing a total of 22 carbon sources, namely arbutin, cellobiose, D-arabinose, D-
704 galactose, D-xylose, D-glucose, galactitol, L-arabinose, L-rhamnose, D-ribose, D-mannitol,
705 lactose, maltose, melezitose, melibiose, myo-inositol, raffinose, ribitol, salicin, sorbitol,
706 trehalose, and xylitol. Both species do not grow on citrate, erythritol, ethanol, ethyl-acetate,
707 glycerol, hexadecane, inulin, lactate, L-sorbose and methanol. Amino acids lysine and
708 cadaverine are utilised as a source of nitrogen, but no growth occurs on potassium nitrate and
709 sodium nitrite. Taking together, the results of the present study and descriptions of *G.*
710 *spencermartinsiae* and *G. psychrophila* (de Garcia et al. 2010; Tsuji et al. 2018), the two
711 species can be only distinguished on the basis of utilisation of D-ribose, sucrose, D-
712 glucosamine and ethylamine.

714 **Description of *Kwoniella fici* Turchetti & Buzzini sp. nov. (MB 829127)**

715 Etymology: The species epithet *fici* is derived from ficus (L. gen. sing. n. n., of fig tree) and
716 refers to the substrate of isolation.

717 After 1 week at 25 °C on MEA, PDA and GPYA, streak culture is greyish-white (MEA,
718 PDA) to cream-colored (GPYA), viscous to butyrous with a dull smooth surface. Margins are
719 smooth and entire and the profile is flat. Cells are spherical to globose, 5–5.5 µm, occurring
720 singly or in pairs, and proliferating by multilateral budding. Pseudohyphae and true hyphae
721 were not observed. Ballistoconidia were not observed. Teleomorph was not observed.

722 Glucose is not fermented. Positive growth on D-glucose, D-galactose (weak, delayed), D-
723 ribose, D-xylose, L-arabinose, D-arabinose, L-rhamnose, sucrose, maltose, trehalose, methyl-
724 alpha-D-glucoside (weak), cellobiose, salicin (weak, delayed), lactose, raffinose, melezitose,
725 glycerol, erythritol, ribitol, xylitol, D-glucitol, D-mannitol, galactitol, myo-inositol, glucono-
726 delta-lactone, D-gluconate, succinate and ethanol. No growth on L-sorbose, arbutin,
727 melibiose, D-glucuronate, D-galacturonate, DL-lactate, citrate, methanol, L-malate,
728 hexadecane, N-acetyl-D-glucosamine, and ethyl acetate. Utilisation of nitrogen sources:
729 positive growth on potassium nitrate, sodium nitrite, ethylamine, lysine and cadaverine. No
730 growth in the presence of 0.01% cycloheximide. Growth in the presence of 10 % NaCl and 50
731 % glucose is negative. Growth on vitamin-free medium is weak. Urea hydrolysis and
732 Diazonium Blue B reaction are positive. Starch-like compounds are abundantly produced.
733 Maximum growth temperature: 30 °C.

734 Molecular characteristics (holotype): nucleotide sequences of ITS and LSU (D1/D2 domains)
735 rRNA deposited in NCBI/EMBL (GenBank) under the accession numbers: MK070336 and
736 MK070318, respectively.

737 Deposits: holotype, strain AH-10 isolated from a fruit of the common fig (*Ficus carica*)
738 collected in September 2011 in Umbria, Italy, preserved in a metabolically inactive state in
739 the Industrial Yeasts Collection DBVPG, Department of Agricultural, Food and
740 Environmental Sciences, University of Perugia, Perugia, Italy as DBVPG 10122^T.

741

742 **Tremellomycetes, Filobasidiales**

743 **Description of *Heterocephalacria fruticeti* f.a. Carvalho, Roehl, Yurkov & Sampaio sp.** 744 **nov. (MB 829128)**

745 Etymology: The species epithet *fruticeti* is derived from fruticetum (L. gen. sing. n. n., of a
746 thicket) and refers to the substrate of isolation, which was a dense *maqui* scrubland.

747 After 1 week at 25 °C on GPY agar and PDA, streak culture is whitish to cream-coloured,
748 mucoid with a glistening smooth surface. Margins are smooth and entire and the profile is flat.
749 Cells are oval to ellipsoidal 3–5 × 5–7 µm in size, occurring singly or in pairs, and
750 proliferating by polar and multilateral budding. Pseudohyphae and true hyphae were not
751 observed. Ballistoconidia were not observed. Teleomorph was not observed.

752 Glucose is not fermented. Positive growth on D-glucose, D-galactose, L-sorbose, L-arabinose,
753 D-arabinose, L-rhamnose, sucrose, maltose, trehalose, methyl-alpha-D-glucoside, cellobiose,
754 salicin, lactose, melezitose, inulin (weak), soluble starch (delayed), ribitol, xylitol, D-glucitol,
755 D-mannitol, inositol, D-glucuronate, succinate, citrate, L-malate, L-tartrate, methanol (weak)
756 and ethanol. No growth on D-glucosamine, D-ribose, D-xylose, melibiose, raffinose, glycerol,

757 erythritol, galactitol, glucono-delta-lactone, D-gluconate and DL-lactate. Utilisation of
758 nitrogen sources: positive growth on potassium nitrate, sodium nitrite, lysine and cadaverine.
759 Growth in the presence of 0.01% and 0.1% cycloheximide is positive. Growth in the presence
760 of 10 % NaCl is negative. Growth on vitamin-free medium is negative. Urea hydrolysis and
761 Diazonium Blue B reaction are positive. Starch-like compounds are produced. Maximum
762 growth temperature: 25 °C.

763 Molecular characteristics (holotype): nucleotide sequences of ITS and LSU (D1/D2 domains)
764 rRNA deposited in NCBI/EMBL (GenBank) under the accession numbers: MK307720 and
765 KT314192, respectively.

766 Deposits: holotype, strain OR 402 isolated from dry scrubland soil collected in the Arrábida
767 Natural Park, Serra da Arrábida, Portugal, preserved in a metabolically inactive state in the
768 Portuguese Yeast Culture Collection, Caparica, Portugal as PYCC 8314^T.

769

770 **Description of *Heterocephalacria gelida* f.a. Turchetti & Kachalkin sp. nov. (MB 829129)**

771 Etymology: The species epithet *gelida* is derived from *gelidus* (L. f. adj., frosty) and refers to
772 the climatic conditions of localities, from which the species was isolated.

773 After 1 week at 25 °C on GPY agar and PDA, streak culture is whitish to cream-colored,
774 mucoid and viscous with a glistering smooth surface. Margins are smooth and entire, and the
775 profile is flat. Cells are sub-globose, oval to ellipsoidal 3–5 × 7–9 µm in size, occurring
776 singly, in pairs or short chains, and proliferating by polar and multilateral budding.

777 Pseudohyphae and true hyphae were not observed. Ballistoconidia were not observed.

778 Sediment is produced when the strain grows in ME and GPY broth at 25°C after 7 days. After
779 14 days, a superficial ring is also present. Teleomorph not observed.

780 Glucose is not fermented. Positive growth on glucose, D-galactose, L-sorbose, D-
781 glucosamine, D-ribose (weak), D-xylose, L-arabinose, D-arabinose (weak), L-rhamnose,
782 sucrose, maltose, trehalose, methyl alpha-methyl-D-glucoside, cellobiose, salicin, arbutin,
783 melibiose (weak), lactose, raffinose, melezitose, ribitol, xylitol, D-glucitol, D-mannitol,
784 galactitol, myo-inositol, glucono-delta-lactone, D-gluconate, and D-galacturonate (weak). No
785 growth on glycerol, erythritol, D-glucuronate, DL-lactate, succinate, citrate, ethanol,
786 methanol, L-malic acid, hexadecane, N-acetyl-D-glucosamine and ethyl acetate. Utilisation of
787 nitrogen sources: positive growth on potassium nitrate, sodium nitrite, ethylamine and lysine.
788 Growth in the presence of 0.01% is weak; no growth in the presence of 0.1% cycloheximide.
789 Growth in the presence of 10 % NaCl and 50 % glucose is negative. Growth on vitamin-free
790 medium is negative. Urea hydrolysis and Diazonium Blue B reaction are positive. Starch-like
791 compounds are produced. Maximum growth temperature: 25 °C.

792 Molecular characteristics (holotype): nucleotide sequences of ITS and LSU (D1/D2 domains)
793 rRNA deposited in NCBI/EMBL (GenBank) under the accession numbers: KC455903 and
794 KC433839, respectively.

795 Deposits: holotype, strain 20.9 LB.6 isolated from supraglacial sediments collected in July
796 2009 in Miage glacier, Mont Blanc massif, Alps, Italy (45.783333N, 6.866667E), preserved

797 in a metabolically inactive state in the Industrial Yeasts Collection DBVPG, Department of
798 Agricultural, Food and Environmental Sciences, University of Perugia, Perugia, Italy as
799 DBVPG 5868^T.

800 Strains studied: DBVPG 5868^T, paratype KBP Y-5466 from leaves of *Dryas punctata*
801 (Rosaceae) collected near the settlement Dikson, Taymyr peninsular, Krasnoyarsk Krai,
802 Russia.

803

804 **Description of *Heterocephalacria hypogea* f.a. Carvalho, Roehl, Yurkov & Sampaio sp.**
805 **nov. (MB 829130)**

806 Etymology: The species epithet *hypogea* is derived from hypogeus (L. f. adj., underground)
807 and refers to the substrate of isolation.

808 After 1 week at 25 °C on GPY agar and PDA, streak culture is whitish to cream-coloured,
809 mucoid with a glistening smooth surface. Margins are smooth and entire and the profile is flat.
810 Cells are oval to ellipsoidal 3–5 × 5–7 µm in size, occurring singly or in pairs, and
811 proliferating by polar and multilateral budding. Pseudohyphae and true hyphae were not
812 observed. Ballistoconidia were not observed. Teleomorph was not observed.

813 Glucose is not fermented. Positive growth on D-glucose, D-galactose, L-sorbose, D-
814 glucosamine, L-arabinose, D-arabinose, L-rhamnose, sucrose, maltose, trehalose, methyl-
815 alpha-D-glucoside, cellobiose, salicin, melibiose (weak), lactose, raffinose, melezitose, inulin
816 (weak), soluble starch (weak), ribitol, xylitol, D-glucitol, D-mannitol, galactitol, inositol, D-
817 glucuronate, succinate, citrate, L-malate, L-tartrate, methanol (weak) and ethanol (weak). No
818 growth on D-ribose, D-xylose, glycerol, erythritol, glucono-delta-lactone, D-gluconate and
819 DL-lactate. Utilisation of nitrogen sources: positive growth on potassium nitrate, sodium
820 nitrite, lysine and cadaverine. Growth in the presence of 0.01% and 0.1% cycloheximide is
821 positive. Growth in the presence of 10 % NaCl is negative. Growth on vitamin-free medium is
822 negative. Urea hydrolysis and Diazonium Blue B reaction are positive. Starch-like
823 compounds are produced. Maximum growth temperature: 25 °C.

824 Molecular characteristics (holotype): nucleotide sequences of ITS and LSU (D1/D2 domains)
825 rRNA deposited in NCBI/EMBL (GenBank) under the accession numbers: MK307721 and
826 KT253539, respectively.

827 Deposits: holotype, strain OR 262 isolated from semihumid chaparral forest soil collected in
828 the Arrábida Natural Park, Serra da Arrábida, Portugal, preserved in a metabolically inactive
829 state in the Portuguese Yeast Culture Collection, Caparica, Portugal as PYCC 6805^T.

830

831 **Description of *Heterocephalacria lusitanica* f.a. Inacio, Carvalho, Roehl, Yurkov &**
832 **Sampaio sp. nov. (MB 829131)**

833 Etymology: The species epithet *lusitanica* (L. f. adj.) is derived from the name of the ancient
834 Roman province Lusitania, which included the territory of modern Portugal. The epithet
835 refers to the locality, where the species was isolated.

836 After 1 week at 25 °C on GPY agar and PDA, streak culture is whitish to cream-coloured,
837 mucoid with a glistering smooth surface. Margins are smooth and entire and the profile is flat.
838 Cells are oval to ellipsoidal 3–5 × 5–7 µm in size, occurring singly or in pairs, and
839 proliferating by polar and multilateral budding. Pseudohyphae and true hyphae were not
840 observed. Ballistoconidia were not observed. Teleomorph was not observed.

841 Glucose is not fermented. Positive growth on D-glucose, D-galactose, L-sorbose, D-
842 glucosamine, D-ribose (variable), D-xylose (variable), L-arabinose, D-arabinose, L-rhamnose,
843 sucrose, maltose, trehalose, methyl-alpha-D-glucoside, cellobiose, salicin, melibiose, lactose,
844 raffinose (delayed), melezitose, inulin (variable), soluble starch (variable, delayed), ribitol,
845 xylitol, D-glucitol, D-mannitol, inositol, glucono-delta-lactone, D-gluconate, D-glucuronate,
846 succinate (variable), citrate, L-malate, L-tartrate, saccharate and galactarate. No growth on
847 glycerol, erythritol, DL-lactate, methanol, ethanol, D-tartrate and m-tartrate. Growth on low-
848 weight aromatic compounds is negative. Utilisation of nitrogen sources: positive growth on
849 potassium nitrate, sodium nitrite, ethylamine (variable), lysine and cadaverine. Growth in the
850 presence of 0.01% and 0.1% cycloheximide is positive. Growth in the presence of 10 % NaCl
851 is negative. Growth on vitamin-free medium is negative. Urea hydrolysis and Diazonium Blue
852 B reaction are positive. Starch-like compounds are produced. Maximum growth temperature:
853 25 °C.

854 Molecular characteristics (holotype): nucleotide sequences of ITS and LSU (D1/D2 domains)
855 rRNA deposited in NCBI/EMBL (GenBank) under the accession numbers: MK307716 and
856 EU002809, respectively.

857 Deposits: holotype, strain 2MV5 isolated from fruits of the strawberry tree (*Arbutus unedo*)
858 collected in the Arrábida Natural Park, Serra da Arrábida, Portugal, preserved in a
859 metabolically inactive state in the Portuguese Yeast Culture Collection, Caparica, Portugal as
860 PYCC 6104^T. Ex-type culture is deposited in the CBS yeast collection of the Westerdijk
861 Fungal Biodiversity Institute, Utrecht, the Netherlands (CBS 10185).

862 Strains studied: 2MV5 (= CBS 10185), paratypes 4ExS4 (= CBS 10186) from an oak exudate,
863 OR 78 (= PYCC 8313) from soil under humid sclerophyll forest. All known strains were
864 isolated from Nature Park of Arrábida on northern and southern slopes of the Serra da
865 Arrábida mountain range from plants and soil (Inacio 2003; Yurkov et al. 2016).

866

867 **Comments to the genus *Heterocephalacria***

868 The genus *Heterocephalacria* proposed by Liu et al. (2015) comprises two teleomorphic
869 mycoparasites *H. bachmannii* and *H. physciacearum* and asexual *H. arrabidensis*, which is
870 known from yeast state only. Although no teleomorph was discovered for newly described *H.*
871 *gelida* and *H. lusitanica*, a mycoparasitic lifestyle cannot be excluded. No culture is available
872 for *H. bachmannii* and *H. physciacearum*. Physiological profiles are only available for
873 asexual species, which can be distinguished based on assimilation of ethanol, glycerol, DL-
874 lactate, succinate, citrate, D-glucuronate, L-malate, nitrate and lysine. However, closely
875 related *H. gelida* and *H. lusitanica* only differ in growth on succinate, citrate, D-glucuronate

876 and L-malate. *H. hypogea* can be distinguished from the closely related *H. arrabidensis* in
877 assimilation of D-xylose, D-ribose, methanol, glycerol, DL-lactate, D-glucuronate, nitrate,
878 ethylamine, and lysine.

879

880 **Description of *Piskurozyma arborea* Yurkov, Kachalkin, Mašínová & Baldrian sp. nov.**
881 **(MB 829132)**

882 Etymology: The species epithet *arborea* refers to the habitat of isolation.

883 After 1 week at 25 °C on GPY agar and PDA, streak culture is white to cream-coloured,
884 butyrous with a glistening smooth surface. Margins are smooth and entire. Cells are ovoid,
885 cylindrical or somewhat sausage-shaped, 6-7 × 2-4 µm. Budding is polar or occurs, in the case
886 of lateral budding, near the poles of the cells. Pseudohyphae and true hyphae were not
887 observed. Ballistoconidia were not observed. Teleomorph was not observed.

888 Glucose is not fermented. Positive growth on D-glucose, D-galactose, L-sorbose, D-ribose
889 (variable), D-xylose, L-Arabinose, D-Arabinose, L-rhamnose, sucrose, maltose, trehalose,
890 cellobiose, salicin (variable), arbutin, melibiose, lactose, raffinose, melezitose, inulin, soluble
891 starch, glycerol (variable), ribitol, xylitol, arabitol, D-glucitol, D-mannitol, galactitol, inositol,
892 2-Keto-D-Gluconate, 5-Keto-D-Gluconate, D-gluconate, D-glucuronate, DL-lactate,
893 succinate, citrate and L-malate. No growth on D-glucosamine and erythritol. Utilization of
894 nitrogen sources: positive growth on potassium nitrate and lysine. Growth in the presence of
895 10 % NaCl and 50 % glucose is negative. Growth on vitamin-free medium is positive. Urea
896 hydrolysis and Diazonium Blue B reaction are positive. Starch-like compounds are not
897 produced. Maximum growth temperature: 25 °C.

898 Molecular characteristics (holotype): nucleotide sequences of ITS and LSU (D1/D2 domains)
899 rRNA deposited in NCBI/EMBL (GenBank) under the accession number KY558349.

900 Deposits: holotype, strain KT168 isolated from spruce litter collected in Kladeruby nad
901 Oslavou, Třebíč District, Vysočina Region, Czech Republic, preserved in a metabolically
902 inactive state in the German Collection of Microorganisms and Cell Cultures (DSMZ),
903 Braunschweig, Germany as DSM 103202^T.

904 Strains studied: DSM 103202^T, paratype KBP Y-4682 (= VKM Y-3011) from Sphagnum
905 moss collected in Moscow Region, Russia.

906

907 **Description of *Piskurozyma silvicultrix* Turchetti, Mašínová, Baldrian & Yurkov sp. nov.**
908 **(MB 829133)**

909 Etymology: The species epithet *silvicultrix* (L. f. adj., inhabiting forest) refers to the habitat of
910 isolation.

911 After 1 week at 25 °C on GPY agar and PDA, streak culture is white to cream-colored,
912 mucoid with a glistening smooth surface. Margins are smooth and entire and the profile is flat.
913 Cells are globose 7-7.5 µm in diameter or ovoid 6-7 × 2-4 µm, occurring singly, in pairs or

914 short chains, and proliferating by polar budding. Pseudohyphae and true hyphae were not
915 observed. Ballistoconidia were not observed. Teleomorph was not observed.

916 Glucose is not fermented. Positive growth on D-glucose, D-galactose, L-sorbose, D-xylose,
917 L-arabinose, sucrose, maltose, trehalose, methyl-alpha-D-glucoside, arbutin, melezitose,
918 soluble starch, erythritol, D-glucitol, D-mannitol, myo-inositol, glucono-delta-lactone
919 (delayed), D-gluconate (delayed), D-gluconate (delayed), D-galacturonate (delayed), DL-
920 lactate (delayed) and succinate. No growth on D-ribose, D-arabinose, L-rhamnose, cellobiose,
921 salicin, melibiose, lactose, raffinose, glycerol, ribitol, xylitol, galactitol, citrate, ethanol,
922 methanol, L-malate, hexadecane, N-acetyl-D-glucosamine and ethyl acetate. Utilisation of
923 nitrogen sources: positive growth on ethylamine; no growth on potassium nitrate, sodium
924 nitrite, L-lysine, and cadaverine. Growth in the presence of 0.01% and 0.1% cycloheximide is
925 positive. Growth in the presence of 10 % NaCl and 50 % glucose is negative. Growth on
926 vitamin-free medium is negative. Urea hydrolysis and Diazonium Blue B reaction are
927 positive. Starch-like compounds are produced. Maximum growth temperature: 25 °C.

928 Molecular characteristics (type strain): nucleotide sequences of ITS and LSU (D1/D2
929 domains) rRNA deposited in NCBI/EMBL (GenBank) under the accession numbers:
930 KU745333 and KU745299, respectively.

931 Deposits: holotype, strain 20.14 A S4 2R isolated from forest soil collected in June 2014 in
932 Kleiner Priol (Montigg), South Tyrol, Alps, Italy, preserved in a metabolically inactive state
933 in the Industrial Yeasts Collection DBVPG, Department of Agricultural, Food and
934 Environmental Sciences, University of Perugia, Perugia, Italy as DBVPG 10557^T.

935 Strains studied: DBVPG 10557^T, paratypes DSM 103194 and DSM 103201 (GenBank
936 KY558341 and KY558348, respectively) from beech litter collected in Czech Republic
937 (Mašíňová et al. 2017b).

938

939 **Description of *Piskurozyma stramentorum* Yurkov, Mašíňová & Baldrian sp. nov. (MB**
940 **829134)**

941 Etymology: The species epithet *stramentorum* is derived from stramentum (L. gen. plu. n. n.,
942 litter) and refers to the substrate of isolation.

943 After 1 week at 25 °C on GPY agar and PDA, streak culture is white to cream-coloured,
944 butyrous with a glistening smooth surface. Margins are smooth and entire. Cells are ovoid,
945 cylindrical or somewhat sausage-shaped, 6-7 × 2-4 µm. Budding is polar or occurs, in the case
946 of lateral budding, near the poles of the cells. Pseudohyphae and true hyphae were not
947 observed. Ballistoconidia were not observed. Teleomorph was not observed.

948 Glucose is not fermented. Positive growth on D-glucose, D-galactose, L-sorbose, D-
949 glucosamine, D-ribose, D-xylose, L-arabinose, D-arabinose, L-rhamnose, sucrose, maltose,
950 D-trehalose, methyl-alpha-D-glucoside (weak), cellobiose, melibiose, lactose, raffinose,
951 melezitose, ribitol, D-glucitol, D-mannitol (weak), D-sorbitol (weak), galactitol, inositol, D-
952 Gluconate, L-malate (weak) and arbutin. No growth on salicin, inulin, glycerol, erythritol,
953 xylitol, citrate, DL-lactate and succinate. Utilization of nitrogen sources: positive growth on

954 potassium nitrate, ethylamine and L-lysine. Growth on vitamin-free medium is negative. Urea
955 hydrolysis and Diazonium Blue B reaction are positive. Starch-like compounds are produced.
956 Maximum growth temperature: 25 °C.

957 Molecular characteristics (type strain): nucleotide sequences of ITS and LSU (D1/D2
958 domains) rRNA deposited in NCBI/EMBL (GenBank) under the accession number
959 KY558344.

960 Deposits: holotype, strain KT146 isolated from spruce litter collected in Kladeruby nad
961 Oslavou, Třebíč District, Vysočina Region, Czech Republic, preserved in a metabolically
962 inactive state in the German Collection of Microorganisms and Cell Cultures (DSMZ),
963 Braunschweig, Germany as DSM 103197^T.

964

965 **Description of *Naganishia nivalis* Turchetti & Buzzini sp. nov. (MB 829135)**

966 Etymology: The species epithet *nivalis* (L. f. adj., snowy) refers to the substrate of isolation.

967 After 1 week at 25 °C on MEA, PDA and GPYA, streak culture is white to cream-colored
968 (becoming brownish upon aging), viscous to butyrous with a dull wrinkled surface. Margins
969 are smooth and entire and the profile is raised. Cells are spherical to ovoid 2.5–3 × 2–2.5 µm
970 in size, occurring singly or in pairs, and proliferating by polar budding. Pseudohyphae and
971 true hyphae were not observed. Ballistoconidia were not observed. Sediment is produced
972 when the strain grows in ME and GPY broth at 25°C after 7 days. After 14 days, a superficial
973 ring is also present. Teleomorph not observed.

974 Glucose is not fermented. Positive growth on D-glucose, D-galactose, D-ribose, D-xylose, L-
975 arabinose (weak), D-arabinose (delayed), sucrose (delayed), maltose, trehalose, methyl-alpha-
976 D-glucoside, cellobiose, salicin, melibiose, lactose, raffinose, melezitose, soluble starch,
977 glycerol, erythritol, ribitol, xylitol, D-glucitol, D-mannitol, galactitol, inositol, methanol,
978 ethanol, hexadecane, N-acetyl-D-glucosamine and ethyl acetate. No growth on L-sorbose, L-
979 rhamnose, arbutin, glucono-delta-lactone, D-gluconate, D-gluconate, D-galacturonate, DL-
980 lactate, succinate, citrate and L-malate. Utilisation of nitrogen sources: positive growth on
981 potassium nitrate, sodium nitrite, ethylamine, lysine and cadaverine (weak). Growth in the
982 presence of 0.01% is negative. Growth in the presence of 10 % NaCl and 50 % glucose is
983 negative. Growth on vitamin-free medium is positive. Urea hydrolysis and Diazonium Blue B
984 reaction are positive. Starch-like compounds are produced. Maximum growth temperature: 25
985 °C.

986 Molecular characteristics (holotype): nucleotide sequences of ITS and LSU (D1/D2 domains)
987 rRNA deposited in NCBI/EMBL (GenBank) under the accession numbers: MK070337 and
988 KC433768, respectively.

989 Deposits: holotype, strain MB 10.12 isolated from snow collected in July 2010 on Glacier du
990 Geant, Mont Blanc massif, Alps, Italy (45.833333N, 6.916667E), preserved in a metabolically
991 inactive state in the Industrial Yeasts Collection DBVPG, Department of Agricultural, Food
992 and Environmental Sciences, University of Perugia, Perugia, Italy as DBVPG 5693^T.

993 Strains studied: DBVPG 5693^T, paratype DBVPG 5706 from the same locality.

994

995 **Microbotryomycetes**

996 **Description of *Yurkovia nerthusi* Yurkov & Begerow, sp. nov. (MB 828552)**

997 Etymology: The specific epithet *nerthusi* is derived from a Latinized form (Nerthus) (NL.
998 gen. sing. m. n., snowy) of Germanic goddess of earth and fertility Nerþuz.

999 After growth on YM agar plates for 1 mo at 16 °C and 22 °C, the streak culture is off-white to
1000 tan, butyrous to mucoid with a smooth and glistening surface. The margin is entire or rarely
1001 wrinkled. After growth on YM agar plates for 7 d at 16 °C and 22 °C, cells are ellipsoidal or
1002 cylindrical (2–4 × 8–10 µm), occurring singly or in pairs, and proliferating by polar budding.
1003 Pseudohyphae and true hyphae were not observed after 1 mo in Dalmau plate culture on
1004 CMA at 16–22 °C. Fermentation is absent.

1005 Assimilation of carbon compounds: Growth on D-glucose, D-galactose, L-sorbose, L-
1006 arabinose (weak), D-arabinose (weak), sucrose, maltose, trehalose, cellobiose, melezitose,
1007 inulin, starch, glycerol, ribitol, DL-lactate, succinate, citrate, ethanol (weak), L-tartaric acid,
1008 D-saccharic acid, 3,4-dihydroxybenzoic acid and 4-hydroxybenzoic acid. No growth on
1009 raffinose, D-ribose, L-rhamnose, salicin, melibiose, lactose, erythritol, xylitol, galactitol and
1010 myo-inositol. Assimilation of nitrogen compounds: growth on potassium nitrate, sodium
1011 nitrite, ethylamine and lysine. Growth in the presence of 0.01% cycloheximide is positive.
1012 Growth in the presence of 10 % NaCl and in medium with 50% and 60% glucose is
1013 negative. Urea hydrolysis and Diazonium Blue B reaction are positive. Maximum growth
1014 temperature: 25 °C.

1015 Molecular characteristics (holotype): nucleotide sequences of LSU (D1/D2 domains) rRNA
1016 gene and ITS deposited in NCBI/EMBL (GenBank) under the accession number FN428970
1017 and **KY083054**, respectively.

1018 Deposits: holotype, strain HEW-2-3 isolated from spruce forest soil collected in Stadtwald
1019 Mühlhausen, Thuringia, Germany (51.21N, 10.37E), preserved in a metabolically inactive
1020 state in the German Collection of Microorganisms and Cell Cultures (DSMZ), Braunschweig,
1021 Germany as DSM 26788^T. Ex-type cultures are deposited in the CBS yeast collection of the
1022 Westerdijk Fungal Biodiversity Institute, Utrecht, the Netherlands (CBS 11560) and the
1023 Mycothèque de l'Université Catholique de Louvain (BCCM/MUCL), Louvain-la-Neuve,
1024 Belgium (MUCL 53962).

1025 Strains studied: DSM DSM 26788^T; paratype AEG-2-20 (= CBS 11560) isolated from
1026 grassland soil in Swabian Alb, in the proximity of Sternberg Wanderheim, Gomadingen,
1027 Baden-Wuerttemberg, Germany (48.391389N, 9.376889E); GenBank FN428969, KY083054.

1028 Notes: Strain AEG-2-20 was recovered on modified Browns' nitrogen deficient media
1029 (Yurkov et al. 2011), whereas strain HEW-2-3 was isolated on nutrient-rich acidified with
1030 lactic acid YPD agar (Yurkov et al. 2012a).

1031

1032 **Validation of *Leucosporidium drummii* Yurkov, A.M. Schäfer & Begerow, International**
1033 **Journal of Systematic and Evolutionary Microbiology 62: 730 (2012); MB 563455**

1034 *Leucosporidium drummii* was described from soil by Yurkov et al. (2012b). However, the
1035 description does not conform to the ICN Melbourne Code Article 40.7, which requires [that](#)
1036 the single herbarium or collection or institution in which the type is conserved must be
1037 specified. We here fulfil the requirements for valid publication of *Leucosporidium drummii*.

1038 Holotype: strain DSM 106046^T, isolated from grassland soil collected near Günterberg,
1039 Angermünde, Brandenburg, Germany, preserved in a metabolically inactive state in the
1040 German Collection of Microorganisms and Cell Cultures (DSMZ), Braunschweig, Germany.
1041 Ex-type cultures are deposited in the CBS yeast collection of the Westerdijk Fungal
1042 Biodiversity Institute, Utrecht, the Netherlands (CBS 11562) and the Mycothèque de
1043 l'Université Catholique de Louvain (BCCM/MUCL), Louvain-la-Neuve, Belgium (MUCL
1044 52878).

1045
1046 **Validation of *Leucosporidium krtinense* Mašínová, A. Pontes, C. Carvalho, J.P. Samp. &**
1047 **Baldrian, International Journal of Systematic and Evolutionary Microbiology 67: 904**
1048 **(2017); MB 815370**

1049 *Leucosporidium krtinense* was described from soil by Mašínová et al. (2017b). However, the
1050 description does not conform to the ICN Melbourne Code Article 40.7, which requires that
1051 the single herbarium or collection or institution in which the type is conserved must be
1052 specified. We here fulfil the requirements for valid publication of *Leucosporidium krtinense*.

1053 Holotype: strain PYCC 6879^T isolated from beech litter collected in October 2013 in the
1054 Křtiny forest area (49.303278N, 16.747389E), Czech Republic, preserved in a metabolically
1055 inactive state in the Portuguese Yeast Culture Collection, Caparica, Portugal. Ex-type cultures
1056 are deposited in the CBS yeast collection of the Westerdijk Fungal Biodiversity Institute,
1057 Utrecht, the Netherlands (CBS 14304) and the in the German Collection of Microorganisms
1058 and Cell Cultures (DSMZ), Braunschweig, Germany (DSM 101892).

1059
1060 **Validation of *Libkindia masarykiana* Mašínová, A. Pontes, C. Carvalho, J.P. Samp. &**
1061 **Baldrian, International Journal of Systematic and Evolutionary Microbiology 67: 906**
1062 **(2017); MB 815373**

1063 *Libkindia masarykiana* was described from soil by Mašínová et al. (2017b). However, the
1064 description does not conform to the ICN Melbourne Code Article 40.7, which requires that
1065 the single herbarium or collection or institution in which the type is conserved must be
1066 specified. We here fulfil the requirements for valid publication of *Libkindia masarykiana*.

1067 Holotype: strain PYCC 6886^T isolated from oak litter collected in April 2014 in the Křtiny
1068 forest area (49.266944N, 16.721167E), Czech Republic, preserved in a metabolically inactive
1069 state in the Portuguese Yeast Culture Collection, Caparica, Portugal. Ex-type cultures are
1070 deposited in the CBS yeast collection of the Westerdijk Fungal Biodiversity Institute, Utrecht,

1071 the Netherlands (CBS 14275) and the in the German Collection of Microorganisms and Cell
1072 Cultures (DSMZ), Braunschweig, Germany (DSM 101891).

1073

1074 **Validation of *Yurkovia mendeliana* Mašínová, A. Pontes, C. Carvalho, J.P. Samp. &**
1075 **Baldrian, *International Journal of Systematic and Evolutionary Microbiology* 67: 907**
1076 **(2017); MB 815372**

1077 *Yurkovia mendeliana* was described from soil by Mašínová et al. (2017b). However, the
1078 description does not conform to the ICN Melbourne Code Article 40.7, which requires that
1079 the single herbarium or collection or institution in which the type is conserved must be
1080 specified. We here fulfil the requirements for valid publication of *Yurkovia mendeliana*.

1081 Holotype: strain PYCC 6884^T isolated from beech litter collected in October 2013 in the
1082 Křtiny forest area (49.324944N, 16.652750E), Czech Republic, preserved in a metabolically
1083 inactive state in the Portuguese Yeast Culture Collection, Caparica, Portugal. Ex-type cultures
1084 are deposited in the CBS yeast collection of the Westerdijk Fungal Biodiversity Institute,
1085 Utrecht, the Netherlands (CBS 14273) and the in the German Collection of Microorganisms
1086 and Cell Cultures (DSMZ), Braunschweig, Germany (DSM 101889).

1087

1088 **Cystobasidiomycetes**

1089 Cyphobasidiales T. Sprib. & H. Mayrhofer (2016) Index Fungorum 309: 1 (MB 552589)
1090 nomen dubium

1091 The order Cyphobasidiales T. Sprib. & H. Mayrhofer (2016) and family Cyphobasidiaceae T.
1092 Sprib. & H. Mayrhofer (2016) were proposed to accommodate lichenicolous parasite
1093 *Cyphobasidium* Millanes, Diederich & Wedin (2016) and lichen-inhabiting fungi represented
1094 by environmental sequences (Spribille et al. 2016). The monophyly of *Cyphobasidium* was
1095 not supported in the ML analysis of rRNA genes by Millanes et al. (2016). The analysis was
1096 also lacking *Cyrenella elegans*, which is the closest relative of *Cyphobasidium* according to
1097 sequence similarities from NCBI Blast and our phylogenetic analysis (Fig. 2). According to
1098 Bauer et al. (2006) *Cyrenella elegans* is placed in order Erythrobasidiales R. Bauer, Begerow,
1099 J.P. Samp., M. Weiss & Oberw. (2006). The order Cyphobasidiales coincides with
1100 Erythrobasidiales and the latter is older name for the order. Although the principle of
1101 taxonomic priority does not apply above the rank of family (ICN Shenzhen Code, Art. 11.10),
1102 the name Cyphobasidiales T. Sprib. & H. Mayrhofer (2016) is a duplicate and nomen dubium.
1103 Its delimitation from the order Erythrobasidiales is unclear and was not supported in our
1104 analyses. The order Erythrobasidiales includes genera *Bannoa*, *Buckleyzyma*, *Cyrenella*,
1105 *Hasegawazyma*, *Erythrobasidium* and *Symmetrospora* (Aime et al. 2006; Bauer et al. 2006;
1106 Wang et al. 2015a, 2015b). Erythrobasidium is an older generic name (ICN Shenzhen Code,
1107 Recommendation 16A.1) and the order Erythrobasidiales has been widely used by yeast
1108 taxonomists, mycologists and botanists (e.g. Aime et al. 2006; Bauer et al. 2006; Ruggiero et
1109 al. 2015; Oberwinkler 2017; Zhao et al. 2017). In our opinion, the name Cyphobasidiales
1110 should not be used in favour of the name Erythrobasidiales.

1111 Similarly, the proposal of the family Cyphobasidiaceae T. Sprib. & H. Mayrhofer (2016) is
1112 premature since its delimitation from Buckleyzymaceae Q.M. Wang, F.Y. Bai, M. Groenew.
1113 & Boekhout (2015), Erythrobasidiaceae Denchev (2009) and Symmetrosporaceae Q.M.
1114 Wang, F.Y. Bai, M. Groenew. & Boekhout (2015) was not supported by a robust
1115 phylogenetic analysis.

1116

1117 **Discussion**

1118 Two important evolutionary characters, such as the ability of basidiospores to germinate by
1119 secondary spore formation and active spore discharge, connected yeasts with some
1120 Basidiomycetes (reviewed by Oberwinkler 1987). The evolution of filamentous habit was
1121 essential for penetration and growth in the substrate and production of fruiting bodies as well
1122 as for the emergence specialised organs like haustoria and sporangia. Although
1123 basidiomycetous yeast species with a sexual state do not form macroscopic fruiting bodies
1124 (basidiocarps) in culture, filaments and haustoria have been observed in several yeasts.
1125 Filamentous heterobasidiomycetes display diverse germination patterns, including budding,
1126 secondary spores, microconidia and hyphae. A close connection between filamentous
1127 parasites like *Carcinomyces*, *Syzygospora* and *Tremella*, and teleomorphic yeasts like
1128 *Filobasidium* and *Filobasidiella* (presently *Cryptococcus*) has been suggested based on the
1129 basidial morphology, presence of haustoria and on ultrastructural markers (Bandoni 1987;
1130 Oberwinkler 1987). The first molecular studies already confirmed close relationships between
1131 some filamentous Heterobasidiomycetes and yeasts, and subsequent works demonstrated that
1132 many basidiomycetous yeasts represent cultivable morphs of Heterobasidiomycetes presently
1133 classified in the classes Cystobasidiomycetes, Microbotryomycetes and Tremellomycetes
1134 (Boekhout et al. 1993; Fell et al. 2000; Scorzetti et al. 2002; Sampaio 2004; Bauer et al. 2006;
1135 Millanes et al. 2011; Liu et al. 2015a; Wang et al. 2015b). Also, large filamentous genera
1136 *Tremella*, *Sirobasidium*, *Syzygospora* and *Cystobasidium* were shown to be polyphyletic. The
1137 implementation of rRNA and multi-gene phylogenies has provided valuable information for
1138 the re-evaluation of the classification of these fungi (Liu et al. 2015a; Wang et al. 2015b).
1139 However, despite substantial progress in species sequencing, taxon sampling for teleomorphic
1140 taxa is meagre. Very few sequences were obtained for species of genera like *Carcinomyces*,
1141 *Holtermannia*, *Sirobasidium*, *Sirotrema*, *Syzygospora*, *Tetragoniomyces* and several lineages
1142 of *Tremella*. Sequences of type species of genera like *Mycogloea*, *Occultifur*, *Sirobasidium*
1143 and *Spiculogloea* are lacking. No sequences are available for *Phragmoxenidium*, *Phyllogloea*,
1144 *Sigmogloea*, *Sigmatrema*, *Tremellina*, *Xenolachne* and *Zygogloea*. Unfortunately, few culture
1145 experiments have been carried out to isolate and preserve these fungi. In addition, many
1146 species are rarely collected. New collections studied molecularly and preserved *ex-situ* would
1147 improve our understanding of the diversity and systematics of this group. The recent
1148 reassessment of the genus *Phaeotremella* provides a good example of the relevance of such
1149 taxonomic studies. Along with the re-collection the availability of cultures, isolates from
1150 several *Tremella* species collected by Franz Oberwinkler and his co-workers were
1151 authenticated and re-sequenced (Spirin et al. 2018). Another recent study by Pontes et al.
1152 (2017) studied the culture of *Tetragoniomyces uliginosus*, which was earlier obtained by

1153 Franz Oberwinkler (PYCC database). Nucleotide sequences from this culture suggest that the
1154 phylogenetic placement of this species in the Trichosporonales (Liu et al. 2015a) is likely to
1155 be an artefact of a short sequence, which was originally obtained from a herbarium material
1156 (Millanes et al. 2011).

1157 Our phylogenetic analyses included sequences from single strains from different studies.
1158 Interestingly, some of them represent conspecific isolates, e.g. in genera *Cryptococcus*,
1159 *Heterocephalacria*, *Piskurozyma* and *Pseudotremella* (Figs. 1, 4, 5). A number of potential
1160 new species were shown to enlarge small or previously monotypic genera, e.g. *Carcinomyces*,
1161 *Fibulobasidium*, *Gelidatrema*, *Heterocephalacria*, *Sugitazyma* and *Yurkovia* (Figs. 1, 2). This
1162 observation legitimises to some extent the re-classification of large polymorphic genera into
1163 smaller, sometimes monotypic entities. A considerable number of species awaiting
1164 description would be otherwise assigned to genera *Bensingtonia*, *Bullera*, *Cryptococcus*,
1165 *Rhodotorula*, and *Sporobolomyces*, increasing taxonomic complexity of many groups. Newly
1166 erected genera of Microbotryomycetes accommodated species previously classified as
1167 *Bensingtonia*, *Rhodotorula*, and *Sporobolomyces* and, in some cases, closely related species
1168 were assigned to different genera (Wang et al. 2015a, 2015b). Similarly, GenBank sequences
1169 of yeasts belonging to such problematic groups were mislabelled or had a doubtful taxonomic
1170 assignment up to the rank of a family. This situation is often confusing; it does not facilitate
1171 description of new species nor does it help to identify them in the environment. Sequences
1172 obtained from Genbank and a few other repositories were placed into the two LSU rRNA
1173 datasets used by Liu et al. (2015a) and Wang et al. (2015b) in order to provide an update on
1174 phylogenetic relationships in Tremellomycetes, Cystobasidiomycetes and
1175 Microbotryomycetes. The presented phylogenetic analyses (Figs. 1, 2) show the diversity of
1176 basidiomycetous yeasts and provide authors of potential new species with a robust
1177 phylogenetic analysis to ease future species descriptions. Discovery of a single strain
1178 representing a new species seldom warrants a publication. However, public sequences
1179 representing yet undescribed species become an extremely important source of rare fungi and
1180 urge researchers to release unpublished data of potential new species to facilitate their formal
1181 description. In the present study, sharing sequence data helped us to identify many closely
1182 related and conspecific strains originating from a number of independent studies.

1183

1184 **Rare and rarely sampled**

1185 Yeasts thrive in the environment not as pure cultures but as a part of a microbial community
1186 which varies in time and space. Physico-chemical characteristics of the environment,
1187 substrate colonisation, dissemination of propagules, competition with other species, and
1188 interactions with potential vectors and predator-prey relationships determine composition of
1189 the community (reviewed in Yurkov 2017; Yurkov and Pozo 2017). As a result, most of
1190 known yeast communities consist of a few dominating species and a large number of species
1191 which are found in low numbers. While isolation of dominant species is easy to achieve,
1192 cultivation of less numerous species requires a larger sampling and cultivation effort.
1193 Consequently, it is well documented and understandable that some species are recovered from

1194 the environment as single strains. These yeasts represent rare species, which are also rarely
1195 sampled.

1196 Yeasts used in this study were isolated from different substrates and regions, suggesting that
1197 basidiomycetous yeasts are rather widespread and not restricted to a specific location or
1198 substrate. Recent surveys of soils and cold habitats reported a substantial proportion of
1199 potential new yeasts (Buzzini et al. 2017; Sannino et al. 2017). These habitats are often
1200 characterized by very uneven structure of yeast communities with a few dominating species
1201 and a large number of rare yeasts. Most of [the](#) new soil-borne basidiomycetes recently
1202 described from soils represent rare taxa (e.g. Yurkov et al. 2016a, 2016b; Mašínová et al.
1203 [2017a, 2017b](#); Pontes et al. 2017). Mediterranean plants sampled in Serra da Arrábida
1204 (Portugal) during years 1997-1999 yielded diverse yeast communities, including many new
1205 species known from very few isolates (Inacio 2003). A number of these yeasts remained
1206 undescribed and were not found again until the recent resampling of the same biotopes
1207 (Yurkov et al. 2016b).

1208 The number of known basidiomycetous species has increased dramatically in recent years. It
1209 is important to highlight that [a number of species and genera](#) in some groups were described
1210 from a very few strains [and that this trend has increased in the last few years](#). [For example,](#)
1211 [80% of species descriptions published in IJSEM since 2018 have been based on 1-3 strains. It](#)
1212 [is also important to mention that taxonomic publications often do not provide essential](#)
1213 [information on the number of samples, isolated strains, species abundance or sampling depth.](#)
1214 [Therefore, it is not possible to conclude whether the sampling effort was sufficient in the](#)
1215 [aforementioned studies and these novel yeasts are rare species. Descriptions of either](#)
1216 [prokaryotic or eukaryotic species are expected to include a careful examination of species](#)
1217 [properties and ecology, which is difficult \(if possible at all\) to achieve on a limited number of](#)
1218 [strains. In our opinion, publication policies need to be modified to ensure that descriptions of](#)
1219 [novel taxa, especially genera, include \(considering availability of both the material and data\)](#)
1220 [all related strains and sequences.](#)

1221 Non-pigmented Microbotryomycetes, previously classified in the genera *Bensingtonia*,
1222 *Rhodotorula* and *Sporobolomyces*, are slow-growing species that are rarely isolated from the
1223 environment. Some species and genera are psychrophilic which makes their cultivation more
1224 complicated. During our past studies, we have also observed yeasts which grow slowly after
1225 isolation but lose their viability after a few passages. Therefore, fast and efficient preservation
1226 of yeast cultures is essential to keep the material for further investigations. Public databases,
1227 GenBank and culture collections contain nucleotide sequences of potential novel species
1228 which were isolated long time ago but await a formal description. Despite ongoing studies of
1229 European forest soils, no additional strain of species recently described by Yurkov et al.
1230 (2016a) such as *Colacogloea*, *Hamamotoa* and *Slooffia* were recovered even though they
1231 were isolated in 2008. Likewise, *Yurkovia nerthusi* described here is known from only two
1232 strains, and no additional culture of this species was found since the first isolation. However,
1233 several yeasts isolated by Fonseca et al. (2000) and Inacio (2003) from the Nature Park of
1234 Arrábida, were not found elsewhere. *Heterocephalacria* (formerly *Cryptococcus*)
1235 *arrabidensis* was isolated from plants and more species of this genus were identified and

1236 described in this study (Figs. 1, 4). One of them is represented by two strains isolated from the
1237 same locality with a gap of 20 years. Similarly, *Cystofilobasidium alribaticum* was described
1238 after a recent resampling in Serra da Arrábida (Pontes et al. 2016). These examples show that
1239 rare species can persist in the environment for a long period. However, since these yeasts are
1240 rare and cultured as single isolates, a substantial sampling effort is required to resample them
1241 or to isolate them from a new source. We do not call for vast species descriptions based on a
1242 single culture but it is important to note that an effort to obtain additional isolates of some
1243 yeasts can be far too difficult.

1244

1245 **Acknowledgements**

1246 We owe much of our present knowledge of diversity and systematics of so-called dimorphic
1247 basidiomycetes heavily relies on specimens, sequence data and publications to Professor
1248 Franz Oberwinkler and his collaborators, including students and trainees of the Tübingen
1249 School.

1250 Álvaro Fonseca is much acknowledged for assistance in sampling in Arrábida Natural Park
1251 and analysis of yeasts from this locality. Angela Schäfer (RUB), Julian Federici (RUB) and
1252 Gabrielle Gresenz (DSMZ) are acknowledged for assistance in the lab. Two anonymous
1253 reviewers are acknowledged for their valuable comments on the manuscript. We thank
1254 MycoBank curator Dr. Konstanze Bensch for her advice on Latin names and Dr. Lara Barnes
1255 (School of Pharmacy and Biomolecular Sciences, University of Brighton) for correcting the
1256 English text.

1257 Ana Pontes, Claudia Carvalho and José Paulo Sampaio acknowledge funding from FCT grant
1258 UID/Multi/04378/2013 (Portugal).

1259

1260 **References**

1261 Babjeva IP, Lisichkina GA, Reshetova IS, Danilevich VN (2002) *Mrakia curviuscula* sp.
1262 nov.: a new psychrophilic yeast from forest substrates. Microbiology (Moscow) 71:449-454

1263 Bandoni RJ (1979). *Fibulobasidium*: a new genus in the Sirobasidiaceae. Can J Bot 57:264-
1264 268

1265 Bandoni RJ (1987) Taxonomic overview of the Tremellales. In: de Hoog GS, Smith MTh,
1266 Weijman ACM (eds), The Expanding Realm of Yeast-like Fungi. Elsevier, Amsterdam, pp
1267 87-110

1268 Banno I (1963) Preliminary report on cell conjugation and mycelial stage in *Rhodotorula*
1269 yeasts. J Gen Appl Microbiol 9:249-251

1270 Banno I (1967) Studies on the sexuality of *Rhodotorula*. J Gen Appl Microbiol 13:167-196

1271 Bauer R, Begerow D, Sampaio JP, Weiß M, Oberwinkler F (2006) The simple-septate
1272 basidiomycetes: a synopsis. Mycol Prog 5:41-66

- 1273 Begerow D, Bauer R, Oberwinkler F (1997) Phylogenetic studies on nuclear large subunit
1274 ribosomal DNA sequences of smut fungi and related taxa. *Can J Bot* 75:2045-2056
- 1275 Begerow D, Kemler M, Feige A, Yurkov A (2017) Parasitism in yeasts. In: Buzzini P,
1276 Lachance MA, Yurkov A (eds) *Yeasts in Natural Ecosystems: Ecology*. Springer, Cham, pp
1277 179-210
- 1278 Boekhout T, Bandoni RJ, Fell JW, Kwon-Chung KJ, Sampaio JP, Fonseca Á (2011)
1279 Discussion of teleomorphic and anamorphic genera of heterobasidiomycetous yeasts. In:
1280 Kurtzman CP, Fell JW, Boekhout T (eds) *The yeasts, a taxonomic study*, 5th edn. Elsevier,
1281 Amsterdam, pp 1339-1374
- 1282 Boekhout T, Fonseca Á, Sampaio JP, Golubev WI (1993) Classification of
1283 heterobasidiomycetous yeasts: characteristics and affiliation of genera to higher taxa of
1284 Heterobasidiomycetes. *Can J Microbiol* 39:276-290
- 1285 Buzzini P., Turk M, Perini L, Turchetti B, Gunde-Cimerman N (2017) Yeasts in Polar and
1286 Subpolar Habitats. In: Buzzini P, Lachance MA, Yurkov A (eds) *Yeasts in Natural*
1287 *Ecosystems: Diversity*. Springer, Cham, pp 331-365
- 1288 Cadez N, Zupan J, Raspor P (2010) The effect of fungicides on yeast communities associated
1289 with grape berries. *FEMS Yeast Res* 10:619-630
- 1290 Clement M, Posada D, Crandall K (2000) TCS: a computer program to estimate gene
1291 genealogies. *Mol Ecol* 9:1657-1660
- 1292 [Dämon W, Hausknecht A \(2002\) First report of a *Sirobasidium* species in Austria, and a](#)
1293 [survey of the Sirobasidiaceae. *Österr Z Pilzk* 11:133-151](#)
- 1294 [de García V, Brizzio S, Russo G, Rosa CA, Boekhout T, Theelen B, Libkind D, van Broock](#)
1295 [M \(2010\) *Cryptococcus spencermartinsiae* sp. nov., a basidiomycetous yeast isolated from](#)
1296 [glacial waters and apple fruits. *Int J Syst Evol Microbiol* 60:707-711](#)
- 1297 Endoh R, Suzuki M, Okada G, Takeuchi Y, Futai K (2011) Fungus symbionts colonizing the
1298 galleries of the ambrosia beetle *Platypus quercivorus*. *Microb Ecol* 62:106-120
- 1299 Fell JW, Boekhout T, Fonseca Á, Scorzetti G, Stätzell-Tallman A (2000) Biodiversity and
1300 systematics of basidiomycetous yeasts as determined by large-subunit rDNA D1/D2 domain
1301 sequence analysis. *Int J Syst Evol Microbiol* 50:1351-1372
- 1302 Fell JW, Boekhout T, Freshwater DW (1995) The role of nucleotide sequence analysis in the
1303 systematics of the yeast genera *Cryptococcus* and *Rhodotorula*. *Stud Mycol* 38:129-146
- 1304 Fonseca Á, Boekhout T, Fell JW (2011). *Cryptococcus* Vuillemin (1901). In: *The yeasts: a*
1305 *taxonomic study* (Kurtzman CP, Fell JW, Boekhout T, eds). Elsevier, Amsterdam, pp 1661-
1306 1737
- 1307 Fonseca Á, Scorzetti G, Fell JW (2000) Diversity in the yeast *Cryptococcus albidus* and
1308 related species as revealed by ribosomal DNA sequence analysis. *Can J Microbiol* 46:7-27

- 1309 Fotedar R, Kolecka A, Boekhout T, Fell JW, Anand A, Al Malaki A, Zeyara A, Al Marri M
 1310 (2018) *Naganishia qatarensis* sp. nov., a novel basidiomycetous yeast species from a
 1311 hypersaline marine environment in Qatar. *Int J Syst Evol Microbiol* 68:2924-2929
- 1312 França L, Sannino C, Turchetti B, Buzzini P, Margesin R (2016) Seasonal and altitudinal
 1313 changes of culturable bacterial and yeast diversity in Alpine forest soils. *Extremophiles*
 1314 20:855-873
- 1315 Glushakova AM, Kachalkin AV (2017) Endophytic yeasts in *Malus domestica* and *Pyrus*
 1316 *communis* fruits under anthropogenic impact. *Microbiology (Moscow)* 86:128-135
- 1317 Glushakova AM, Kachalkin AV, Chernov IY (2015) Effect of invasive herb species on the
 1318 structure of soil yeast complexes in mixed forests exemplified by *Impatiens parviflora* DC.
 1319 *Microbiology (Moscow)* 84:717-721
- 1320 Herzberg M, Fischer R, Titze A (2002) Conflicting results obtained by RAPD-PCR and large-
 1321 subunit rDNA sequences in determining and comparing yeast strains isolated from flowers: a
 1322 comparison of two methods. *Int J Syst Evol Microbiol.* 52:1423-1433
- 1323 Inácio J (2003) Yeast occurrence and diversity on the phylloplane of selected plants from the
 1324 Arrábida Natural Park. PhD thesis (in Portuguese). Universidade Nova de Lisboa, Portugal
- 1325 Inácio J, Pereira P, de Carvalho M, Fonseca Á, Amaral-Collaço MT, Spencer-Martins I
 1326 (2002) Estimation and diversity of phylloplane mycobiota on selected plants in a
 1327 Mediterranean-type ecosystem in Portugal. *Microb Ecol* 44:344-353
- 1328 Kachalkin AV, Glushakova AM, Yurkov AM, Chernov IYu (2008) Characterization of yeast
 1329 groupings in the phyllosphere of sphagnum mosses. *Microbiology (Moscow)* 77:474-481
- 1330 Katoh K, Rozewicki J, Yamada KD (2017) MAFFT online service: multiple sequence
 1331 alignment, interactive sequence choice and visualization. *Brief Bioinform* bbx108
- 1332 Kluyver AJ, van Niel CB (1927). *Sporobolomyces*: ein Basidiomyzet? *Ann Mycol* 25:389-
 1333 394
- 1334 Kurtzman CP, Boekhout T (2017) Yeasts as distinct life forms of Fungi. In: Buzzini P,
 1335 Lachance MA, Yurkov A (eds) *Yeasts in Natural Ecosystems: Ecology*. Springer, Cham, pp
 1336 1-37
- 1337 Kurtzman CP, Fell JW, Boekhout T, Robert V (2011) Methods for isolation, phenotypic
 1338 characterization and maintenance of yeasts. In: Kurtzman CP, Fell JW, Boekhout T (eds) *The*
 1339 *yeasts, a taxonomic study*, 5th edn. Elsevier, Amsterdam, pp 97-107
- 1340 Liu XZ, Wang QM, Göker M, Groenewald M, Kachalkin AV, Lumbsch HT, Millanes AM,
 1341 Wedin M, Yurkov AM, Boekhout T, Bai FY (2015a) Towards an integrated phylogenetic
 1342 classification of the Tremellomycetes. *Stud Mycol* 81:85-147
- 1343 Liu XZ, Wang QM, Theelen B, Groenewald M, Bai FY, Boekhout T (2015b) Phylogeny of
 1344 tremellomycetous yeasts and related dimorphic and filamentous basidiomycetes reconstructed
 1345 from multiple gene sequence analyses. *Stud Mycol* 81:1-26.

- 1346 Mašínová T, Bahnmann BD, Větrovský T, Tomšovský M, Merunková K, Baldrian P (2017a)
1347 Drivers of yeast community composition in the litter and soil of a temperate forest. FEMS
1348 Microbiol Ecol 93:fiw223
- 1349 [Mašínová T, Pontes A, Carvalho C, Sampaio JP, Baldrian P \(2017b\) *Libkindia masarykiana*](#)
1350 [gen. et sp. nov., *Yurkovia mendeliana* gen. et sp. nov. and *Leucosporidium krtinense* fa sp.](#)
1351 [nov., isolated from temperate forest soils. Int J Syst Evol Microbiol 67:902-908.](#)
- 1352 Mašínová T, Yurkov A, Baldrian P (2018) Forest soil yeasts: decomposition potential and the
1353 utilization of carbon sources. Fungal Ecol 34:10-19
- 1354 Millanes AM, Diederich P, Ekman S, Wedin M (2011) Phylogeny and character evolution in
1355 the jelly fungi (Tremellomycetes, Basidiomycota, Fungi). Mol Phylogenet Evol 61:12-28.
- 1356 Millanes AM, Diederich P, Wedin M (2016) *Cyphobasidium* gen. nov., a new lichen-
1357 inhabiting lineage in the Cystobasidiomycetes (Pucciniomycotina, Basidiomycota, Fungi).
1358 Fungal Biol 120:1468-1477
- 1359 Mittelbach M, Yurkov AM, Stoll R, Begerow D (2016) Inoculation order of nectar-borne
1360 yeasts opens a door for transient species and changes nectar rewarded to pollinators. Fungal
1361 Ecol 22:90-97
- 1362 Oberwinkler F (1987) Heterobasidiomycetes with ontogenetic yeast-stages – systematic and
1363 phylogenetic aspects. In: de Hoog GS, Smith MTh, Weijman ACM (eds), The Expanding
1364 Realm of Yeast-like Fungi. Elsevier, Amsterdam, pp 61-74
- 1365 Oberwinkler F (2017) Yeasts in Pucciniomycotina. Mycol Progress 16:831-856
- 1366 Péter G, Takashima M, Čadež N. (2017) Yeast habitats: different but global. In: Buzzini P,
1367 Lachance MA, Yurkov A (eds) Yeasts in Natural Ecosystems: Ecology. Springer, Cham, pp
1368 39-71
- 1369 Pontes A, Röhl O, Maldonado C, Yurkov AM, Sampaio JP (2017) *Cryptotrichosporon argae*
1370 sp. nov., *Cryptotrichosporon brontae* sp. nov. and *Cryptotrichosporon steropae* sp. nov.,
1371 isolated from forest soils. Int J Syst Evol Microbiol 67:3610-3614
- 1372 [Ruggiero MA, Gordon DP, Orrell TM, et al. \(2015\) A higher level classification of all living](#)
1373 [organisms. PloS One 10:e0119248](#)
- 1374 Sampaio JP (2004) Diversity, phylogeny and classification of basidiomycetous yeasts. In:
1375 Agerer R, Piepenbring M, Blanz P (eds), Frontiers in Basidiomycete Mycology. IHW Verlag,
1376 Eching, pp 49-80
- 1377 Sampaio JP (2011) *Rhodotorula* Harrison (1928). In: Kurtzman CP, Fell JW, Boekhout T
1378 (eds) The yeasts: a taxonomic study, 5th edn. Elsevier, Amsterdam, pp 1873-1927
- 1379 Sannino C, Tasselli G, Filippucci S, Turchetti B, Buzzini P (2017) Yeasts in Nonpolar Cold
1380 Habitats. In: Buzzini P, Lachance MA, Yurkov A (eds) Yeasts in Natural Ecosystems:
1381 Diversity. Springer, Cham, pp 367-396

- 1382 Scorzetti G, Fell JW, Fonseca Á, Stazzell-Tallman A (2002) Systematics of basidiomycetous
1383 yeasts: a comparison of large subunit D1D2 and internal transcribed spacer rDNA regions.
1384 FEMS Yeast Res 2: 495-517
- 1385 Seifert KA, Rossman AY (2010) How to describe a new fungal species. IMA Fungus. 1:109-
1386 116
- 1387 Selbmann L, Zucconi L, Onofri S, Cecchini C, Isola D, Turchetti B, Buzzini P (2014)
1388 Taxonomic and phenotypic characterization of yeasts isolated from worldwide cold rock-
1389 associated habitats. Fungal Biol 118:61-71
- 1390 Šibanc N, Zalar P, Schroers HJ, Zajc J, Pontes A, Sampaio JP, Maček I (2018) *Occultifur*
1391 *mephitis f.a.*, sp. nov. and other yeast species from hypoxic and elevated CO₂ mofette
1392 environments. Int J Syst Evol Microbiol. 68:2285-2298
- 1393 Spribille T, Tuovinen V, Resl P et al (2016) Basidiomycete yeasts in the cortex of ascomycete
1394 macrolichens. Science 353:488-492
- 1395 Spirin V, Malysheva V, Yurkov A et al (2018) Studies in the *Phaeotremella foliacea* group
1396 (Tremellomycetes, Basidiomycota). Mycol Prog 17:451-466
- 1397 Sylvester K, Wang QM, James B, Mendez R, Hulfachor AB, Hittinger CT (2015)
1398 Temperature and host preferences drive the diversification of *Saccharomyces* and other
1399 yeasts: a survey and the discovery of eight new yeast species. FEMS Yeast Res 15:fov002
- 1400 Taylor DL, McCormick MK (2008) Internal transcribed spacer primers and sequences for
1401 improved characterization of basidiomycetous orchid mycorrhizas. New Phytol 177:1020-
1402 1033
- 1403 Turchetti B, Goretti M, Branda E, Diolaiuti G, D'Agata C, Smiraglia C, Onofri A, Buzzini P
1404 (2013) Influence of abiotic variables on culturable yeast diversity in two distinct Alpine
1405 glaciers. FEMS Microbiol Ecol 86:327-340
- 1406 Turchetti B, Selbmann L, Gunde-Cimerman N, Buzzini P, Sampaio JP, Zalar P (2018)
1407 *Cystobasidium alpinum* sp. nov. and *Rhodospordiobolus oreadorum* sp. nov. from European
1408 Cold Environments and Arctic Region. Life 8:9
- 1409 Wang QM, Groenewald M, Takashima M, Theelen B, Han PJ, Liu XZ, Boekhout T, Bai FY
1410 (2015a) Phylogeny of yeasts and related filamentous fungi within Pucciniomycotina
1411 determined from multigene sequence analyses. Stud. Mycol 81:27–53
- 1412 Wang QM, Yurkov AM, Göker M, Lumbsch HT, Leavitt SD, Groenewald M, Theelen B, Liu
1413 XZ, Boekhout T, Bai FY (2015b) Phylogenetic classification of yeasts and related taxa within
1414 Pucciniomycotina. Stud Mycol 81:149-189
- 1415 Yurkov AM (2017) Temporal and Geographic Patterns in Yeast Distribution. In: Buzzini P,
1416 Lachance MA, Yurkov A (eds) Yeasts in Natural Ecosystems: Ecology. Springer, Cham, pp
1417 101-130

- 1418 Yurkov AM, Pozo MI (2017) Yeast Community Composition and Structure. In: Buzzini P,
1419 Lachance MA, Yurkov A (eds) Yeasts in Natural Ecosystems: Ecology. Springer, Cham, pp
1420 73-100
- 1421 Yurkov AM, Kemler M, Begerow D (2012a) Assessment of yeast diversity in soils under
1422 different management regimes. Fungal Ecol 5:24-35
- 1423 Yurkov AM, Schäfer AM, Begerow D (2012b) *Leucosporidium drummii* sp. nov., a member
1424 of the Microbotryomycetes isolated from soil. Int J Syst Evol Microbiol 62:728-734.
- 1425 Yurkov A, Guerreiro MA, Sharma L, Carvalho C, Fonseca Á (2015a) Multigene Assessment
1426 of the Species Boundaries and Sexual Status of the Basidiomycetous Yeasts *Cryptococcus*
1427 *flavescens* and *C. terrestris* (Tremellales). PLoS One 10:e0120400
- 1428 Yurkov AM, Kachalkin AV, Daniel HM, Groenewald M, Libkind D, de Garcia V, Zalar P,
1429 Gouliamova DE, Boekhout T, Begerow D (2015b) Two yeast species *Cystobasidium*
1430 *psychroaquaticum* f.a. sp. nov. and *Cystobasidium rietchieii* f.a. sp. nov. isolated from natural
1431 environments, and the transfer of *Rhodotorula minuta* clade members to the genus
1432 *Cystobasidium*. Antonie van Leeuwenhoek 107:173-185
- 1433 Yurkov AM, Wehde T, Federici J et al (2016a) Yeast diversity and species recovery rates
1434 from beech forest soils. Mycol Prog 15:845-859
- 1435 Yurkov AM, Röhl O, Pontes A et al. (2016b) Local climatic conditions constrain soil yeast
1436 diversity patterns in Mediterranean forests, woodlands and scrub biome FEMS Yeast Res 16:
1437 fov103
- 1438 Yurkov AM, Dlačhy D, Péter G (2017) *Meyerozyma amylolytica* sp. nov. from temperate
1439 deciduous trees and the transfer of five *Candida* species to the genus *Meyerozyma*. Int J Syst
1440 Evol Microbiol 67:3977-3981
- 1441 [Zhao RL, Li GJ, Sánchez-Ramírez S, et al. \(2017\). A six-gene phylogenetic overview of](#)
1442 [Basidiomycota and allied phyla with estimated divergence times of higher taxa and a](#)
1443 [phyloproteomics perspective. Fungal Divers 84:43-74.](#)
- 1444 [Zhao Y, Liu XZ, Bai FY \(2019\) Four new species of *Tremella* \(Tremellales, Basidiomycota\)](#)
1445 [based on morphology and DNA sequence data. MycoKeys 47:75-95](#)

1446

1447 **Figure captions**

1448 Figure 1 – Phylogenetic relationships of yeasts and related taxa in Agaricomycotina obtained
1449 by maximum-likelihood analysis of LSU (D1/D2 domains) rRNA gene. Tree topology was
1450 constrained according to the topology of the seven genes-based tree (for details see Liu et al.
1451 2015a, 2015b) with nodes showing bootstrap values >85 % inforced to be monophyletic. Taxa
1452 not included in the previous analysis by Liu et al. (2015) are in red. The numbers provided on
1453 branches are frequencies (> 50 %) with which a given branch appeared in 100 bootstrap
1454 replications. The scale bars indicate the numbers of expected substitutions accumulated per
1455 site.

1456 Figure 2 – Phylogenetic relationships of yeasts and related taxa from Pucciniomycotina
1457 lineages obtained by Maximum-Likelihood analysis of the LSU (D1/D2 domains) rRNA
1458 gene. Tree topology was constrained according to the topology of the seven genes-based tree
1459 (for details see Wang et al. 2015a, 2015b) with nodes showing bootstrap values >85 %
1460 enforced to be monophyletic. Taxa not included in the phylogenetic analysis of the seven
1461 genes (Wang et al. 2015a) are indicated in red. The numbers provided on branches are
1462 frequencies (> 50 %) with which a given branch appeared in 100 bootstrap replications. The
1463 scale bars indicate the numbers of expected substitutions accumulated per site.

1464 Figure 3 – Phylogenetic relationships of yeasts and related taxa from the order
1465 Cystofilobasidiales in Tremellomycetes obtained by maximum-likelihood analysis of a
1466 concatenated alignment of the (A) ITS region and LSU (D1/D2 domains) rRNA gene and (B)
1467 ITS region, LSU rRNA gene and *TEF1*. The numbers provided on branches are frequencies
1468 (> 50 %) with which a given branch appeared in 100 bootstrap replications. The scale bars
1469 indicate the numbers of expected substitutions accumulated per site.

1470 Figure 4 – Maximum likelihood analysis of a concatenated alignment of the ITS region and
1471 LSU (D1/D2 domains) rRNA gene for the genus *Heterocephalacria*. The numbers provided
1472 on branches are frequencies (> 50 %) with which a given branch appeared in 100 bootstrap
1473 replications. The scale bars indicate the numbers of expected substitutions accumulated per
1474 site.

1475 Figure 5 – Maximum likelihood analysis of a concatenated alignment of the ITS region and
1476 LSU (D1/D2 domains) rRNA gene for the genus *Piskurozyma*. The numbers provided on
1477 branches are frequencies (> 50 %) with which a given branch appeared in 100 bootstrap
1478 replications. The scale bars indicate the numbers of expected substitutions accumulated per
1479 site.

1480 Figure 6 – Maximum likelihood analysis of a concatenated alignment of the ITS region and
1481 LSU (D1/D2 domains) rRNA gene for the genus *Naganishia*. The numbers provided on
1482 branches are frequencies (> 50 %) with which a given branch appeared in 100 bootstrap
1483 replications. The scale bars indicate the numbers of expected substitutions accumulated per
1484 site.

1485 Figure 7 – Micrographs showing morphology of new species: *Vustinia terrae* (a), vegetative
1486 cells on PDA after 7 d at 20 °C; *Udeniomyces caspiensis* (b), vegetative cells on 2% Glucose
1487 YNB agar, after 7 d at 20 °C; *Tausonia rosea* (c), vegetative cells on PDA, after 7 d at 20 °C;
1488 *Udeniomyces orazovii* (d-f), vegetative cells (d), microcolony (e), and chlamydospores (f) on
1489 PDA after 10 d at 20 °C; *Itersonilia diksonensis* (g, h), vegetative cells (g) and sympodially
1490 branched sterigmata with ballistoconidia (h) on 2% Glucose YNB agar, after 14 d at 20 °C;
1491 *Krasilnikovozyma fibulata* (i-k), vegetative cells (i), hyphae with clamp connections and
1492 teliospores (j, k) on PDA after 14 d at 20 °C. Scale bars: 10 µm (a, b, c, d, g, h, i, k), 20 µm
1493 (f), 60 µm (e) and 70 µm (j).