
***Portalia* gen. nov. (Ustilaginomycotina)**

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González, V., Vánky, K., Platas, G. and Lutz, M. (2007). *Portalia* gen. nov. (Ustilaginomycotina). *Fungal Diversity* 27: 45-59.

A new, monotypic genus, *Portalia* is described for *Cintractia uljanishcheviana*, a smut fungus collected for the first time in Europe (Spain), in addition to its type locality in Kazakhstan. The phylogenetic relationship of the new genus to other smut genera, such as *Cintractia*, *Dermatosorus*, *Gymnocintractia*, *Leucocintractia*, *Parvulago*, *Pilocintractia* and *Stegocintractia* is discussed, based on ribosomal DNA LSU sequence analyses. A detailed description, including spore germination, obtained for the first time, as well as illustrations are given for *Portalia uljanishcheviana*.

Key words: new combination, new genus, LSU rDNA, molecular phylogeny, *Portalia*, *P. uljanishcheviana*, *Scirpus*, smut fungi, taxonomy, Ustilaginomycotina

Introduction

Schwarzman (1960: 162) described a new smut fungus from Kazakhstan, *Cintractia uljanishcheviana*, in the 'ovaries' of *Holoschoenus vulgaris* Link (= *Scirpus holoschoenus* L., *Cyperaceae*). It is characterised by black spore masses and single, brown, verrucose, rather agglutinated spores. At that time, the genus *Cintractia* Cornu was heterogeneous and all smut fungi with black, agglutinated spore masses with single spores were included. Based on the 2-celled basidia, Brefeld (1895: 144) separated the genus *Anthracoidea*, but it was neglected until reinstated by Kukkonen (1963). Similar smut fungi, which were not on host plants in the *Cyperaceae*, were also removed from *Cintractia* (comp. Vánky, 1987). Based on morphological and molecular data Piepenbring *et al.* (1999), and Piepenbring (2000) split the large, heterogeneous *Cintractia* into four genera, recognising also *Anthracoidea*. Piepenbring (2000: 362)

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considered *C. uljanishcheviana* to be a species of *Anthracoidea*, but did not make a combination into *Anthracoidea*.

In 2004 a smut fungus was collected in Spain by V. González *et al.* on *Scirpus holoschoenus*. In molecular analyses it clustered with *Dermatosorus cyperi* and *Parvulago marina*. Morphological studies showed that it is identical with "*Cintractia*" *uljanishcheviana*, for which a new genus is proposed.

Materials and methods

Specimens examined

Sorus and spore characteristics were studied using either dried herbarium specimens (preserved partly in Herbarium *Ustilaginales* Vánky, HUV 12116), or freshly collected material from Spain, [Cuenca Prov., Huete, road CM 2025 Sacedón-Huete, Arroyo Valquemado, 40°09'58.44" N, 2°40'49.5" W, alt. 720 m, 4.V.2004, leg. V. González, G. García and A. Quintanar, on dried, past years inflorescences of *Scirpus holoschoenus* (HUV 21361); *ibidem*, 8.VIII.2006, leg. V. González (HUV 21372)].

For light microscopy (LM) and spore measurements, dried spores were rehydrated in lactophenol by gently heating to boiling point. For scanning electron microscopy (SEM), dried spores were dusted on double-sided adhesive tape, mounted on a specimen stub, sputter-coated with gold-palladium, *ca.* 20 nm, and examined in a SEM at 10 kV.

Spore germination was studied on water agar (WA), at room temperature. The spores were either collected from plants that were stored outdoor over the winter, or freshly collected spores were suspended in distilled water, kept at –20°C for 12 hours and then dispersed on WA.

DNA extraction, PCR amplification, and DNA sequencing

For DNA extraction, two thirds of the cone shaped bottom of an Eppendorf tube was filled with spores of *Portalia uljanishcheviana*, and processed as previously described (Peláez *et al.*, 1996). Amplification of the ITS2-D1-D2 region of the 28S rDNA was carried out using the primers ANT-1 5' CAACTTTTGGCAACGGATCTC 3'a that matches a zone of the 5.8S gene of *Ustilaginales* and NL4 (O'Donnell, 1993).

PCR reactions were performed following standard procedures recommended by the manufacturer (5 minutes at 93°C followed by 40 cycles of 30 s at 93°C, 30 s at 53°C and 2 min at 72°C) with Taq DNA polymerase (QBiogene, Inc.). The amplification products (0.10 µg/ml) were sequenced

using the Big Dye Terminator Cycle Sequencing Kit (Applied Biosystems) following the manufacturer's recommendations. Amplification products were sequenced with the same primers used for the initial amplification plus NL1 (O'Donnell, 1993). Partial sequences obtained in sequencing reactions were assembled using Bioedit 7.0.5.3 (Hall, 1999).

The sequence obtained was compared to GenBank database using FastA applications (GCG Wisconsin Package, Version 10.3-UNIX, Accelrys Inc.). To get insight into the phylogenetic relationships of *Portalia* and other members of the Ustilaginomycetes, other D1-D2 28S rDNA sequences of taxa from both the *Ustilaginales* and the *Urocystales* were included in the analyses. In addition, three sequences representing *Eballistra* species (Exobasidiomycetes) were included to root the phylogenetic reconstructions. GenBank accession numbers are given in Fig. 5. Alignments of the homologous regions of the different strains were performed using the multiple alignment program ClustalW (Thompson *et al.*, 1994). The final size of the alignment was 533 bases.

Phylogenetic analysis

Phylogenetic analyses were performed using sequences obtained by the authors and mostly retrieved from GenBank public databases, representing genera and species of several families of the *Ustilaginales*. All the LSU sequences employed are listed in Table 1.

Bayesian analysis based on Markov Monte Carlo chain approach was run as implemented in the computer program MrBayes 3.01 (Huelsenbeck *et al.*, 2002). To improve mixing of the chain, four incrementally heated simultaneous Monte Carlo Markov chains were run over 2,000,000 generations. MrModeltest 2.2 (Nylander, 2004) was used to perform hierarchical likelihood ratio tests to calculate the akaike information criterion (AIC) values of the nucleotide substitution models. The model selected by AIC for the present alignment was the GTR+I+G model of DNA substitution considering a proportion of invariable sites for the substitution rates, number of substitution rates = 6. Trees were sampled every 100 generations, resulting in an overall sampling of 20,001 trees. The initial 1001 trees were not used for the posterior analysis. From those trees that were sampled after the process had reached stationarity, a 50% majority-rule consensus tree was computed to get estimates for a posteriori probabilities.

For the Maximum Likelihood analyses, MODELTEST (Posada and Crandall, 1998) was used to identify the model of DNA substitution that best fits. MODELTEST analyses selected the GTR+I+G model with the estimation of nucleotide frequencies (A = 0.2601, C = 0.1954, G = 0.21926, T = 0.3043)

Table 1. Sequences used in this study.

Species	Genbank accession number	Strain no.	Original Substrate	Reference
<i>Cintractia amazonica</i> Syd. & P. Syd.	AJ236142	MP 2008	<i>Rhynchospora barbata</i> (Vahl) Kunth	Piepenbring <i>et al.</i> , 1999
<i>Cintractia axicola</i> (Berk.) Cornu	AF009847	HUV 17460	<i>Fimbristylis tetragona</i> R. Br.	Begerow <i>et al.</i> , 1997
<i>Cintractia limitata</i> G.P. Clinton	AJ236147	MP 1310	<i>Cyperus rotundus</i> L.	Piepenbring <i>et al.</i> , 1999
<i>Cintractia limitata</i> G.P. Clinton	AJ236146	MP 1594	<i>Cyperus ligularis</i> L.	Piepenbring <i>et al.</i> , 1999
<i>Cintractia michellii</i> Vánky	AJ236149	HUV 17666	<i>Fimbristylis schultzei</i> Boeck.	Piepenbring <i>et al.</i> , 1999
<i>Dermatosorus cyperi</i> Vánky	AJ236157	HUV 15991	<i>Cyperus celluloso-reticulatus</i> Boeckeler	Piepenbring <i>et al.</i> , 1999
<i>Eballistra brachiariae</i> (Viégas) R. Bauer, Begerow, A. Nagler & Oberw.	AF009864	FO 17510	<i>Brachiaria distycha</i> (L.) Stapf	Begerow <i>et al.</i> , 1997
<i>Eballistra lineata</i> (Cooke) R. Bauer, Begerow, A. Nagler & Oberw.	AY525372	HUV 741	<i>Zizania aquatica</i> var. <i>angustifolia</i> (Hitchc.) Tsvelev	Bauer <i>et al.</i> , 2005
<i>Eballistra oryzae</i> (Syd. & P. Syd.) R. Bauer, Begerow, A. Nagler & Oberw.	AF229353	Isolate 127114	<i>Oryza sativa</i> L.	Bauer <i>et al.</i> , 2005
<i>Eriomoeszia eriocauli</i> (G.P. Clinton) Vánky	AY740094	MS246	<i>Eriocaulon cinereum</i> R. Br.	Stoll <i>et al.</i> , 2005
<i>Farysia chardoniana</i> Zundel	AF009859	MP 2062	<i>Carex polystachya</i> Swartz ex Wahlenb.	Begerow <i>et al.</i> , 1997
<i>Heterotolyposporium piluliforme</i> (Berk.) Vánky	AF009871	HUV 15732	<i>Juncus planifolius</i> R. Br.	Begerow <i>et al.</i> , 1997
<i>Leucocontractia leucodermoides</i> (Berk.) M. Piepenbr., Begerow & Oberw.	AJ236145	MP 2026	<i>Rhynchospora holoschoenoides</i> (L.C. Rich.) Herter	Piepenbring <i>et al.</i> , 1999
<i>Leucocontractia scleriae</i> (DC.) M. Piepenbr., Begerow & Oberw.	AJ236154	MP 2074	<i>Rhynchospora triflora</i> Vahl	Piepenbring <i>et al.</i> , 1999
<i>Macalpinomyces eragrostiellae</i> Vánky & C. Vánky	AY740089	MS80	<i>Eragrostiella bifaria</i> (Vahl) Bor	Stoll <i>et al.</i> , 2005

Table 1 continued. Sequences used in this study.

Species	Genbank accession number	Strain no.	Original Substrate	Reference
<i>Macalpinomyces eriachnes</i> (Thüm.) Langdon & Full.	AY740090	MS293	<i>Eriachne aristidea</i> F. Mueller	Stoll <i>et al.</i> , 2005
<i>Macalpinomyces eriachnes</i> (Thüm.) Langdon & Full.	AY740091	MS294	<i>Eriachne helmsii</i> (Domin) Harley	Stoll <i>et al.</i> , 2005
<i>Melanotaenium cingens</i> Bref.	DQ875364	Isolate 1943	<i>Linaria vulgaris</i> Mill.	Unpublished
<i>Melanotaenium euphorbiae</i> (L.W. Lenz) M.D. Whitehead & Thirum.	AJ236135	HUV 17733	<i>Euphorbia heterophylla</i> L.	Piepenbring <i>et al.</i> , 1999
<i>Melanotaenium endogenum</i> (Unger) de Bary	DQ789979	CBS 481.91	<i>Galium mollugo</i> L.	Unpublished
<i>Moreaua rhynchosporae-cephalotis</i> (Vánky & T. Vánky) Vánky	AJ236160	HUV 15199	<i>Rhynchospora cephalotes</i> (L.) Vahl	Piepenbring <i>et al.</i> , 1999
<i>Parvulago marina</i> (Durieu) R. Bauer, M. Lutz, M. Piatek & Oberw.	DQ185437	M, TUB 012166	<i>Eleocharis parvula</i> (Roem. & Schult.) Link ex Bluff, Nees & Schauer	Bauer <i>et al.</i> (unpublished)
<i>Pilocintractia fimbristylidicola</i> (Pavgi & Mundk.) Vánky	AJ236143	MP 2213	<i>Fimbristylis spadicea</i> (L.) Vahl	Piepenbring <i>et al.</i> , 1999
<i>Portalia uljanishcheviana</i> (Schwarzman) V. González, Vánky & G. Platas	EF118824	HUV 21361	<i>Scirpus holoschoenus</i> L.	This work
<i>Schizonella cocconii</i> (Morini) Liro	AJ236158	DB 614	<i>Carex flacca</i> Schreber	Piepenbring <i>et al.</i> , 1999
<i>Schizonella melanogramma</i> (DC.) J. Schröt.	AF009870	FO 37174	<i>Carex pilulifera</i> L.	Begerow <i>et al.</i> , 1997
<i>Sporisorium apludae-aristatae</i> (B.V. Patil & Thirum.) Vánky	AY740098	MS287	<i>Apluda mutica</i> L.	Stoll <i>et al.</i> , 2005
<i>Sporisorium cruentum</i> (J.G. Kühn) Vánky	AY74156	MS14	<i>Sorghum bicolor</i> (L.) Moench	Stoll <i>et al.</i> , 2005
<i>Sporisorium destruens</i> (Schltdl.) Vánky	AY747077	MS133	<i>Panicum miliaceum</i> L.	Stoll <i>et al.</i> , 2005
<i>Sporisorium fastigiatum</i> Vánky	AY740133	MS45	<i>Andropogon angustatus</i> (J. Presl) Steud.	Stoll <i>et al.</i> , 2005

Table 1 continued. Sequences used in this study.

Species	Genbank accession number	Strain no.	Original Substrate	Reference
<i>Sporisorium foveolati</i> (Maire) Vánky	AY740103	MS21	<i>Eremopogon foveolatus</i> Stapf	Stoll <i>et al.</i> , 2005
<i>Sporisorium loudetiae-pedicellatae</i> Vánky & C. Vánky	AY740106	MS252	<i>Loudetia pedicellata</i> (Stent.) Chippind.	Stoll <i>et al.</i> , 2005
<i>Sporisorium moniliferum</i> (Ellis & Everh.) L. Guo	AF453940	MS 98	<i>Heteropogon contortus</i> (L.) P. Beauv. ex Roem. & Schult.	Piepenbring <i>et al.</i> , 2002
<i>Sporisorium ophiuri</i> (Henn.) Vánky	AJ236136	HB 20	<i>Rottboellia cochinchinensis</i> (Lour.) W. Clayton	Piepenbring <i>et al.</i> , 1999
<i>Sporisorium penniseti</i> (Rabenh.) Ershad	AY740130	MS5	<i>Pennisetum setaceum</i> (Forsk.) Chiov.	Stoll <i>et al.</i> , 2005
<i>Sporisorium sorghi</i> Ehrenb. ex Link.	AY745726	CBS104.17	unknown	Unpublished
<i>Sporisorium sorghi</i> Ehrenb. ex Link.	AF009872	Isolate 1056	<i>Sorghum bicolor</i> (L.) Moench	Begerow <i>et al.</i> , 1997
<i>Sporisorium veracruzianum</i> (Zundel & Dunlap) M. Piepenbr.	AY740114	Isolate 1335	<i>Panicum viscidellum</i> Scribn.	Stoll <i>et al.</i> , 2005
<i>Stegocintractia luzulae</i> (Sacc.) M. Piepenbr., Begerow & Oberw.	AJ236148	MP 2340	<i>Luzula pilosa</i> (L.) Willd.	Piepenbring <i>et al.</i> , 1999
<i>Stegocintractia spadicea</i> (Liro) M. Piepenbr. & Begerow	AJ236155	MP 2350	<i>Luzula alpinopilosa</i> (Chaix) Breistr.	Piepenbring <i>et al.</i> , 1999
<i>Tolyposporium junci</i> (J. Schröt.) Woronin	AF009876	HUV 17168	<i>Juncus bufonius</i> L.	Begerow <i>et al.</i> , 1997
<i>Trichocintractia utriculicula</i> (Henn.) M. Piepenbr.	AF009877	MP 2075	<i>Rhynchospora corymbosa</i> (L.) Britton	Begerow <i>et al.</i> , 1997
<i>Ustanciosporium cubense</i> (M. Piepenbr.) M. Piepenbr. & Begerow	AJ236153	MP 2252	<i>Rhynchospora microcephala</i> Britton	Piepenbring <i>et al.</i> , 1999
<i>Ustanciosporium montagnei</i> (Tul. & C. Tul.) M. Piepenbr., Begerow & Oberw.	AJ236150	MP 2344	<i>Rhynchospora alba</i> (L.) Vahl	Piepenbring <i>et al.</i> , 1999

Table 1 continued. Sequences used in this study.

Species	Genbank accession number	Strain no.	Original Substrate	Reference
<i>Ustanciosporium neomontagnei</i> M. Piepenbr. & Begerow	AJ236151	MP 925	<i>Rhynchospora globosa</i> (Kunth) Roem & Schult.	Piepenbring <i>et al.</i> , 1999
<i>Ustanciosporium rhynchosporae</i> Vánky	AJ236144	MP 2335	<i>Rhynchospora alba</i> (L.) Vahl	Piepenbring <i>et al.</i> , 1999
<i>Ustanciosporium samanense</i> (Cif.) M. Piepenbr.	AJ236152	MP 2106	<i>Rhynchospora fascicularis</i> (Michx.) Vahl ssp. <i>fascicularis</i>	Piepenbring <i>et al.</i> , 1999
<i>Ustanciosporium standleyanum</i> (Zundel) M. Piepenbr.	DQ846888	JAG 73	unknown	Unpublished
<i>Ustanciosporium taubertianum</i> (Henn.) M. Piepenbr. & Begerow	DQ839599	H AJB10528	unknown	Unpublished
<i>Ustanciosporium taubertianum</i> (Henn.) M. Piepenbr. & Begerow	AJ236156	MP 1801	<i>Rhynchospora tenuis</i> Link	Piepenbring <i>et al.</i> , 1999
<i>Websdanea lygniae</i> (Websdane, Sivasith., K.W. Dixon & Pate) Vánky	AJ236159	HUV 17900	<i>Lyginia barbata</i> (Labill.) R.Br.	Piepenbring <i>et al.</i> , 1999

with the shape parameter of the gamma distribution ($\alpha = 0.6237$) to accommodate rate variations among sites. Maximum Likelihood analysis was performed with PAUP 4.0. The starting tree was obtained via neighbour-joining; the branch-swapping algorithm used was TBR.

The multiple alignment and phylogenetic tree performed with Bayesian analysis were deposited on TREEBASE under the number SN3137.

Results

Two phylogenetic reconstructions based on the comparison of D1-D2 28S rDNA sequences were made employing Bayesian (BY) and Maximum Likelihood (ML) analyses. This was done to obtain an accurate view on the systematic relationships between *Portalia*, the new smut genus here proposed, and several related smut fungi such as *Cintractia s. l.*, including genera recently segregated from this genus on the basis of molecular and morphological evidence (Piepenbring *et al.*, 1999), and some other smut genera belonging to the *Ustilaginales* and the *Urocystales*. Thus, molecular analyses covered several genera in the *Ustilaginales* (*Sporisorium*, *Moreaua*, *Dermatosorus*, *Farysia*, *Eriomoeszia*, *Macalpinomyces* and *Websdanea*), plus one genus, *Melanotaenium*, presently considered within the order *Urocystales*.

Topologies resolved by means of Maximum Likelihood analysis were identical (data not shown) to those obtained with Bayesian methods. Thus, results described and further discussion is based on the phylogram obtained with Bayesian analysis.

Phylogenetic analysis of the D1 and D2 domains of the LSU rDNA (Fig. 5) provided evidence to consider "*Cintractia*" *uljanishcheviana* to be outside the genus *Cintractia*. The LSU sequence representing "*Cintractia*" *uljanishcheviana* clustered together with sequences representing two other members of the *Ustilaginales* with an uncertain systematic placement: *Ustilago marina* Durieu (= *Parvulago marina*) and *Dermatosorus cyperi* Vánky. The latter, a peculiar smut fungus recently accommodated (Vánky, 2001) in the family *Dermatosoraceae*, was revealed (with a support of 95%) as sister taxon to "*Cintractia*" *uljanishcheviana*. *Parvulago* was recently erected (M. Lutz, pers. comm., in prep.) to accommodate *Ustilago marina* (on *Eleocharis parvula*), a taxon that exhibits morphological and molecular features different to those accepted for the modern concept of the large and complex genus *Ustilago*.

Furthermore, phylogenetic reconstruction suggests a polyphyletic origin for both the *Ustilaginaceae* and the *Cintractiaceae*. Thus, topology resolved in

the Bayesian analysis (Fig. 5), showed several main clusters, where genera from these two families grouped and were distributed and partly also mixed.

All the species belonging to *Cintractia s. str.* clustered in a single clade on a basal position with the rest of the genera analyzed. Such topology validates the prevailing view that considered this family as a polyphyletic and heterogeneous group. Some allied, recently segregated genera, such as *Stegocintractia*, *Ustanciosporium* and *Leucocintractia* (Piepenbring *et al.*, 1999) appeared not closely related and even distant (in the case of *Stegocintractia*) with the rest of the taxa included in the *Cintractia s. str.* “core” clade resolved in Fig. 5.

Phylogenetic reconstruction also provided evidence to consider *Pilocintractia fimbristylidicola*, a taxon recently removed (Vánky, 2004) from the concept of *Cintractia s. str.* Topology obtained here arranged *P. fimbristylidicola* on a single branch, external and not related with *Cintractia* or any of the recently segregated *Cintractia*-like genera. Furthermore, one sequence representing *Eriomoeszia eriocauli* (= *Moesziomyces eriocauli*) (Vánky, 2005) was found to group together with *P. fimbristylidicola* but clustering on a separate branch.

Discussion

On account of the molecular and morphometrical evidence (including morphology, cytology and teliospore germination) provided here, the new genus *Portalia* could be considered as a natural taxon of the *Ustilaginales*, being removed out of the concept of genus *Cintractia s. l.* The relative position of the isolate of *P. uljanishcheviana* in the phylogenetic reconstruction (Fig. 5) suggested that *Portalia* is not closely related to *Cintractia*, nor with any of the several *Cintractia*-like genera recently erected.

Considering *Dermatosorus cyperi* and *Parvulago marina*, the species which were revealed as closest relatives of *Portalia* in the molecular analyses, the morphology of the types of these three genera is so markedly different (for *Ustilago marina* comp. Vánky, 1994:366 and 417, for *Dermatosorus* comp. Vánky, 2002:44-45) that we consider the erection of a new genus for *Cintractia uljanishcheviana* is justified.

Concerning the family concept for the *Ustilaginaceae*, current systematic schemes based on morphological features and host relationships do not fit well with molecular-based grouping provided here and in previous phylogenetic studies (e.g. Begerow *et al.*, 1997; Piepenbring *et al.*, 1999; Stoll *et al.*, 2005; Vánky *et al.*, 2006). There is no molecular support to consider the *Cintractiaceae* as a monophyletic group. Thus, we prefer to retain all those

genera (representing the *Ustilaginaceae*, *Cintractiaceae*, *Dermatosoraceae* and *Farysiaceae*) under a big and extensive concept of *Ustilaginaceae* until more indepth combined analyses suggest more stable taxonomical conclusions within the group.

In summary, molecular analyses performed have: 1) provided evidence for considering the new genus *Portalia* as a natural taxon within the *Ustilaginales*; 2) supported the previously assumed heterogeneity of the *Ustilaginaceae*; 3) suggested a possible polyphyletic origin for the family *Cintractiaceae*; and 4) not fully resolve the systematic placement of *P. uljanishcheviana* within the *Cintractiaceae*.

As a result of our morphological and molecular biology investigations of *Cintractia uljanishcheviana* we propose the erection of a new genus:

***Portalia* V. González, Vánky & G. Platas, gen. nov.**

Mycobank: 510390.

Etymology: *Portalia* derived from the name María Ángeles Portal, the wife of Dr. Vicente González.

Sori in floribus familiae *Cyperaceae* (*Scirpus*), sine peridio et stromate fungali, e massa sporarum nigra, mature pulverea, e sporis tantum compositi, sine cellulis sterilibus. Infectio systemica. *Sporae* singulares, pigmentatae (brunneae), ornamentatae. *Germinatio sporarum* basidiis septatis (phragmobasidiis) cellulis eorum compatilibus fuis hyphae dicaryoticae producentibus. Typus generis:

Sori in the flowers of *Cyperaceae* (*Scirpus*), peridium and fungal stroma lacking, spore mass black, when mature powdery, composed of spores only, no sterile cells. Infection systemic. *Spores* single, pigmented (brown), ornamented. *Spore germination* results in septate basidia (phragmobasidia) of which compatible cells fuse producing dikaryotic hyphae.

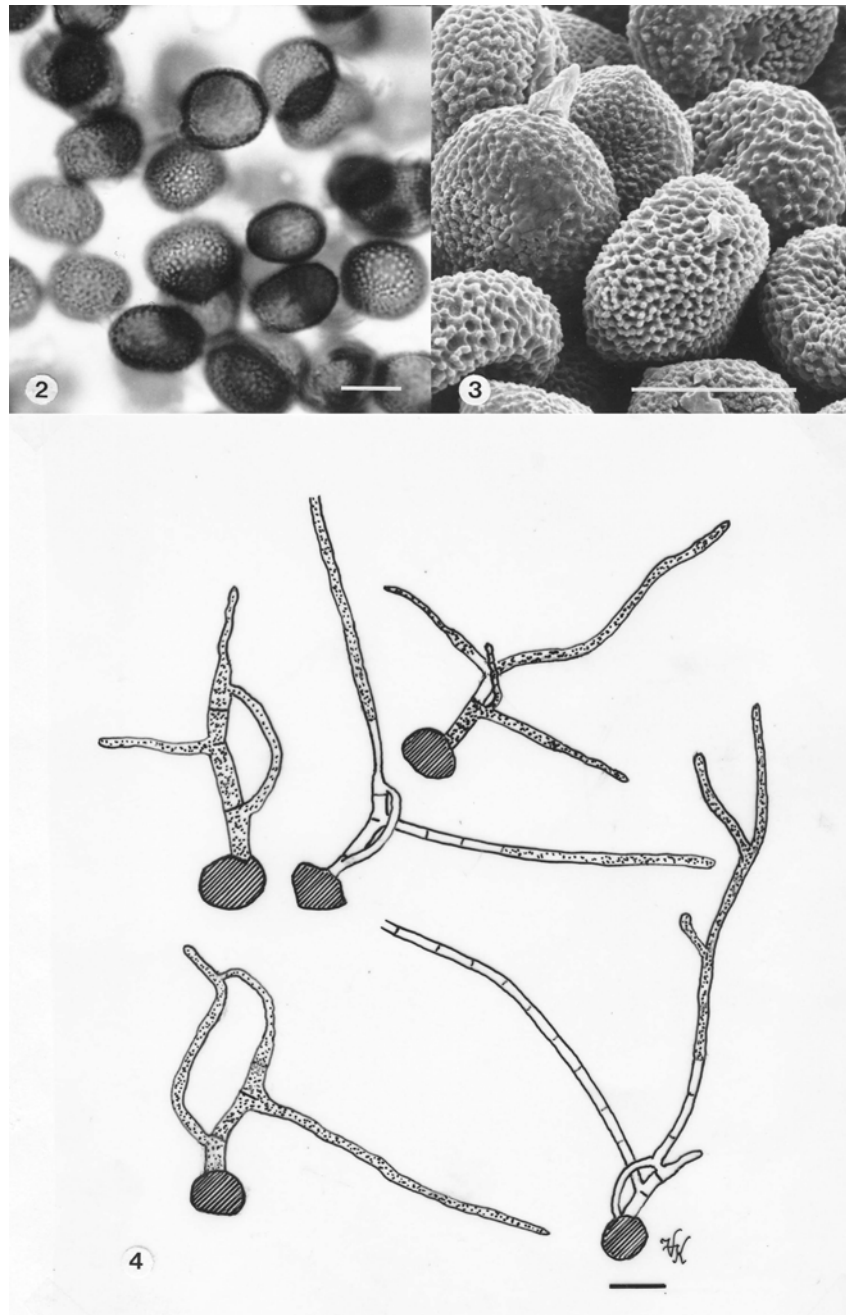
***Portalia uljanishcheviana* (Schwarzman) V. González, Vánky & G. Platas, comb. nov.** (Figs 1-4)

Basionym (≡): *Cintractia uljanishcheviana* Schwarzman, *Fl. spor. rast. Kazakhstana* 2: 162, 1960 (as "*uljanishchevianum*"). — Type on *Holoschoenus vulgaris* (= *Scirpus holoschoenus*), Kazakhstan, Dzhambul'skaya Obl., Ul'kun-Burul, Lake Bijlikul, 12.VII.1949, S.R. Schwarzman. **Holotype** in AA, isotype in HUV 12116.

Sori in the flowers, usually affecting all in a spikelet, destroying the innermost floral organs replacing them by a black, semiagglutinated to powdery mass of spores completely hidden by the outermost floral envelopes. Peridium and stroma lacking. Infected spikelets swollen, globoid or ovoid, 1-2 mm in diameter. Infection systemic; all spikelets (or nearly all) (Fig. 1b) of a head, and all heads of an inflorescence are affected (Fig. 1a). *Spores* (Figs 2, 3) single,



Fig. 1. Sori of *Portalia uljanishcheviana* in the flowers of *Scirpus holoschoenus*. **a:** Habit; **b:** Aspect of healthy inflorescences (from Spain, HUV 21372).



Figs. 2, 3. Spores of *Portalia uljanishcheviana* on *Scirpus holoschoenus* (from type). **1.** LM. **2.** SEM. **Fig. 4.** Germinating spores of *Portalia uljanishcheviana* on *Scirpus holoschoenus*, on WA, at room temperature, in 2-3 days (from Spain, HUV 21372). Bars = 10 μ m.

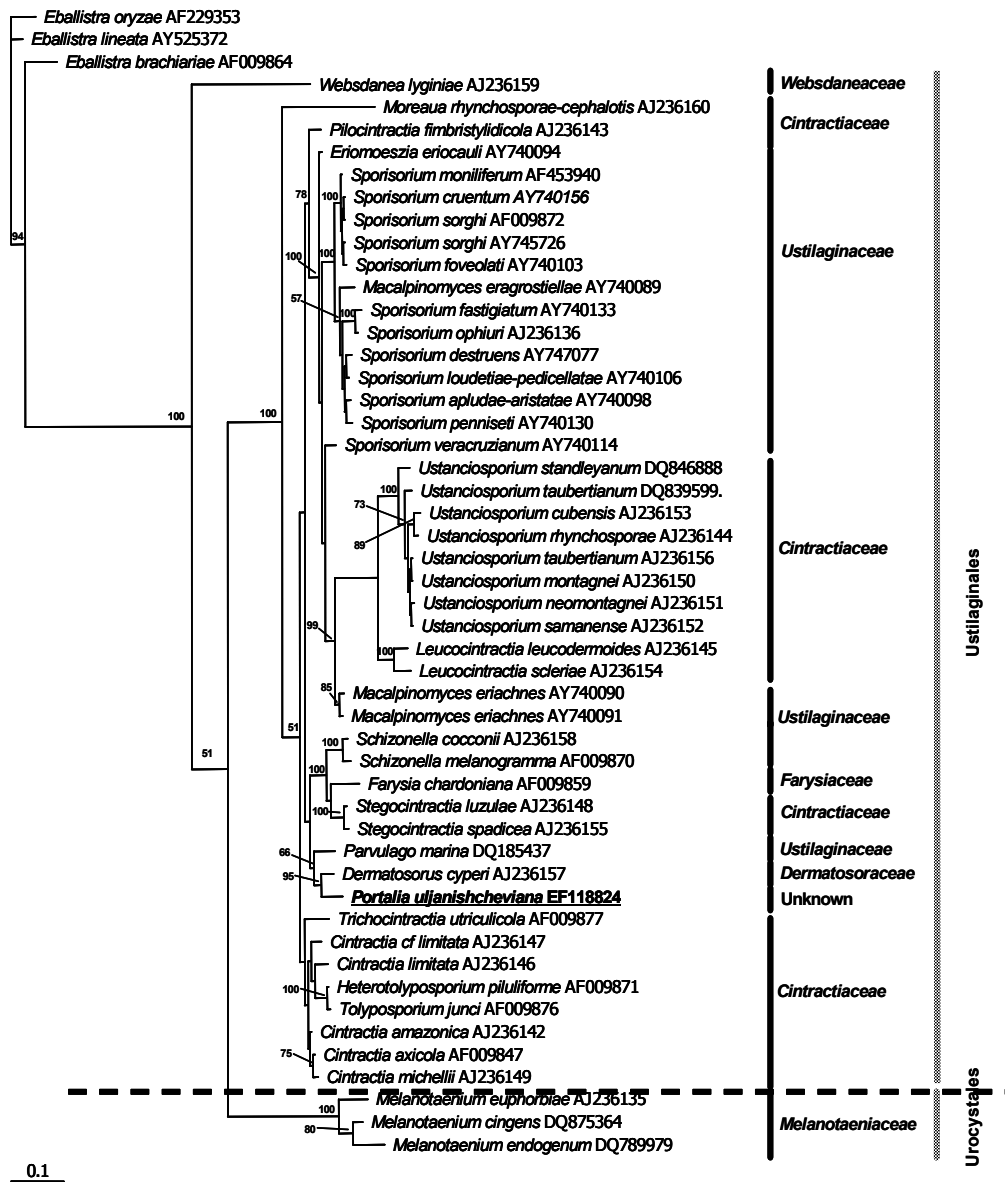


Fig. 5. Phylogenetic tree based on the comparison of LSU rDNA sequences generated by Bayesian analysis, representing several genera and families of the *Ustilaginales* and *Urocystales*, including *Portalia uljanischeviana*.

subglobose, ovoid, ellipsoidal, elongated or irregular, sometimes with a short subacute tip and also with a short, hyaline papilla, 10.5-15 × 12-18.5 µm, reddish- or olivaceous-brown; wall even to slightly uneven, 0.5-1(-1.5) µm thick, finely to coarsely verrucose, warts isolated or often connected by low ridges forming a fine, irregular and incomplete reticulum, as seen in SEM. Spore profile in LM wavy to finely, bluntly serrulate. *Spore germination* (Fig. 4; on WA, at room temp., after 2-3 days) were obtained with both methods employed and results in a 4-celled basidium measuring 2.5-3 × 15-37 µm. After conjugation of compatible basidial cells, c. 2 µm wide, long, dikariotic, ramifying hyphae are produced with retraction septa.

On *Cyperaceae*: *Scirpus holoschoenus* L. (*Holoschoenus vulgaris* Link); Europe (Spain), Asia (Kazakhstan). Probably more common but overlooked.

Acknowledgements

The authors are grateful to Dr. S. Tóth (Gödöllő, Hungary) for providing the Latin description, to Dr. F. Peláez (MSD-CIBE, Spain), for reading the manuscript and checking its English, to the Director and Curator of the Herbarium AA (Alma Ata) for the isotype of *Cintractia uljanishcheviana*, and to Dr. E. McKenzie (New Zealand Fungal Herbarium-PDD) for critically reading and checking the manuscript.

References

- Bauer, R., Lutz, M. and Oberwinkler, F. (2005). *Gjaerumia*, a new genus in the *Georgefischeriales* (Ustilaginomycetes). *Mycological Research* 109: 1250-1258.
- Begerow, D., Bauer, R. and Oberwinkler, F. (1997/1998). Phylogenetic studies on nuclear large subunit ribosomal DNA sequences of smut fungi and related taxa. *Canadian Journal of Botany* 75: 2045-2056.
- Brefeld, O. (1895). *Untersuchungen aus dem Gesamtgebiete der Mykologie*. XII. Hemibasidii. Brandpilze III. Münster i. W., Commissions-Verlag v. H. Schöningh. IV + 99-236 pp. + Pls. VI-XII.
- Hall, T.A. (1999). BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acids Symposium Serie* 41: 95-98.
- Huelsenbeck J.P., Larget, B., Miller, R.E. and Ronquist, F. (2002). Potential applications and pitfalls of Bayesian inference of phylogeny. *Systematic Biology* 51: 673-688.
- Kukkonen, I. (1963). Taxonomic studies on the genus *Anthracoidea* (Ustilaginales). *Annales Botanici Societatis Zoologicae Botanicae Fennicae "Vanamo"* 34: 1-122.
- Nylander, J.A.A. (2004). *MrModeltest v2*. Program distributed by the author. Evolutionary Biology Centre, Uppsala University.
- O'Donnell, K. (1993). *Fusarium* and its near relatives. In: *The fungal holomorph: mitotic, meiotic and pleomorphic speciation in fungal systematics* (eds. D.R. Reynolds, and J.W. Taylor). CAB international, Wallingford, U.K : 225-233.
- Peláez, F., Platas, G., Collado, J. and Díez, M.T. (1996). Intraspecific variation in two species of aquatic hyphomycetes, assessed by RAPD analysis. *Mycological Research* 100: 831-837.

- Piepenbring, M. (2000). The species of *Cintractia* s. l. (*Ustilaginales*, Basidiomycota). *Nova Hedwigia* 70: 289-372.
- Piepenbring, M., Begerow, D. and Oberwinkler, F. (1999). Molecular sequence data assess the value of morphological characteristics for a phylogenetic classification of species of *Cintractia*. *Mycologia* 91: 485-498.
- Piepenbring, M., Stoll, M. and Oberwinkler, F. (2002). The generic position of *Ustilago maydis*, *Ustilago scitaminea*, and *Ustilago esculenta* (*Ustilaginales*). *Mycological Progress* 1: 71-80.
- Posada D. and Crandall, K.A. (1998). Modeltest: testing the model of DNA substitution. *Bioinformatics* 14: 817-818.
- Schwarzman, S.R. (1960). Golovnevye griby. (Smut fungi; in Russian). Flora sporovykh rastenij Kazachstana 2. Alma Ata.
- Stoll, M., Begerow, D. and Oberwinkler, F. (2005). Molecular phylogeny of *Ustilago* and *Sporisorium*, and related taxa based on combined analysis of rDNA sequences. *Mycological Research* 109: 342-356.
- Thompson, J.D., Higgins, D.J. and Gibson, T.J. (1994). CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. *Nucleic Acids Research* 22: 4673-4680.
- Vánky, K. (1987). Illustrated genera of smut fungi. *Cryptogamic Studies* 1: 1-159 (ed. W. Jülich). Gustav Fischer Verlag, Stuttgart/New York.
- Vánky, K. (1994). *European smut fungi*. Gustav Fischer Verlag, Stuttgart, Germany.
- Vánky, K. (2001). The emended *Ustilaginaceae* of the modern classificatory system for smut fungi. *Fungal Diversity* 6: 131-147.
- Vánky, K. (2002). *Illustrated Genera of Smut Fungi*. 2nd Ed. APS Press, St. Paul, Minnesota, USA.
- Vánky, K. (2004). *Pilocintractia* gen. nov. (Ustilaginomycetes). *Mycologia Balcanica* 1: 169-174.
- Vánky, K., Lutz, M. and Shivas, R. (2006). *Anomalomyces panici*, new genus and species of Ustilaginomycetes from Australia. *Mycologia Balcanica* 3: 119-126.
- Vánky, K. (2005). The smut fungi (Ustilaginomycetes) of *Eriocaulaceae*. I. *Eriomoeszia* gen. nov. *Mycologia Balcanica* 2: 105-112.

(Received 31 January 2007; accepted 11 July 2007)