

## New collections of *Flammulina rossica*

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Two specimens from Germany and one from South Dakota, USA, were identified as *Flammulina rossica* during the examination of the München herbarium (M) material of the genus *Flammulina*. Micromorphological characters of these specimens are described and illustrated. The variability and delimitation of *F. rossica* is discussed and the knowledge of its distribution, ecology, morphology and biology is summarized.

**Key words:** fungi, Basidiomycota, *Xerulaceae*, distribution, morphology.

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Počas revidovania herbárového materiálu rodu *Flammulina* z Mníchova (M) sme dva zbery z Nemecka a jeden z Južnej Dakoty v USA určili ako *Flammulina rossica*. V práci uvádzame opis a vyobrazenie mikromorfologických znakov týchto zberov. Hodnotíme variabilitu a ohraničenie druhu *F. rossica* a sumarizujeme poznatky o jeho rozšírení, ekológii, morfológii a biológii.

### INTRODUCTION

*Flammulina rossica* Redhead et R. H. Petersen was described as a new species based on material from eastern Russia and USA: Alaska (Redhead and Petersen 1999). Fruitbodies of this species were also collected in these two regions (Hughes et al. 1999), which suggested the idea of its migration between Eurasia and North America via the Bering land bridge during the last glacial maximum (Redhead and Petersen 1999, Hughes et al. 1999). However, more recently, the fungus has been detected in western Russia, Japan, USA: New York State, northern Thailand (Petersen and Hughes 2007), Canada: British Columbia (Petersen et al. on-line) and Tibet (Anonymus on-line).

During our study of taxonomy and biogeography of the genus *Flammulina* in Central Europe in the years 2004–2007, we identified three specimens at the München herbarium (M) as *F. rossica*. Two of these collections were collected in

Germany, one in USA: South Dakota. Thus, these collections also support the fact that *F. rossica* is widespread (but rare) in the Northern Hemisphere and not only trans-Beringian (Petersen and Hughes 2007).

The aim of our paper is to present the three mentioned collections of *F. rossica* from herbarium M, and to summarize current knowledge of its distribution, ecology, morphology and biology.

#### MATERIAL AND METHODS

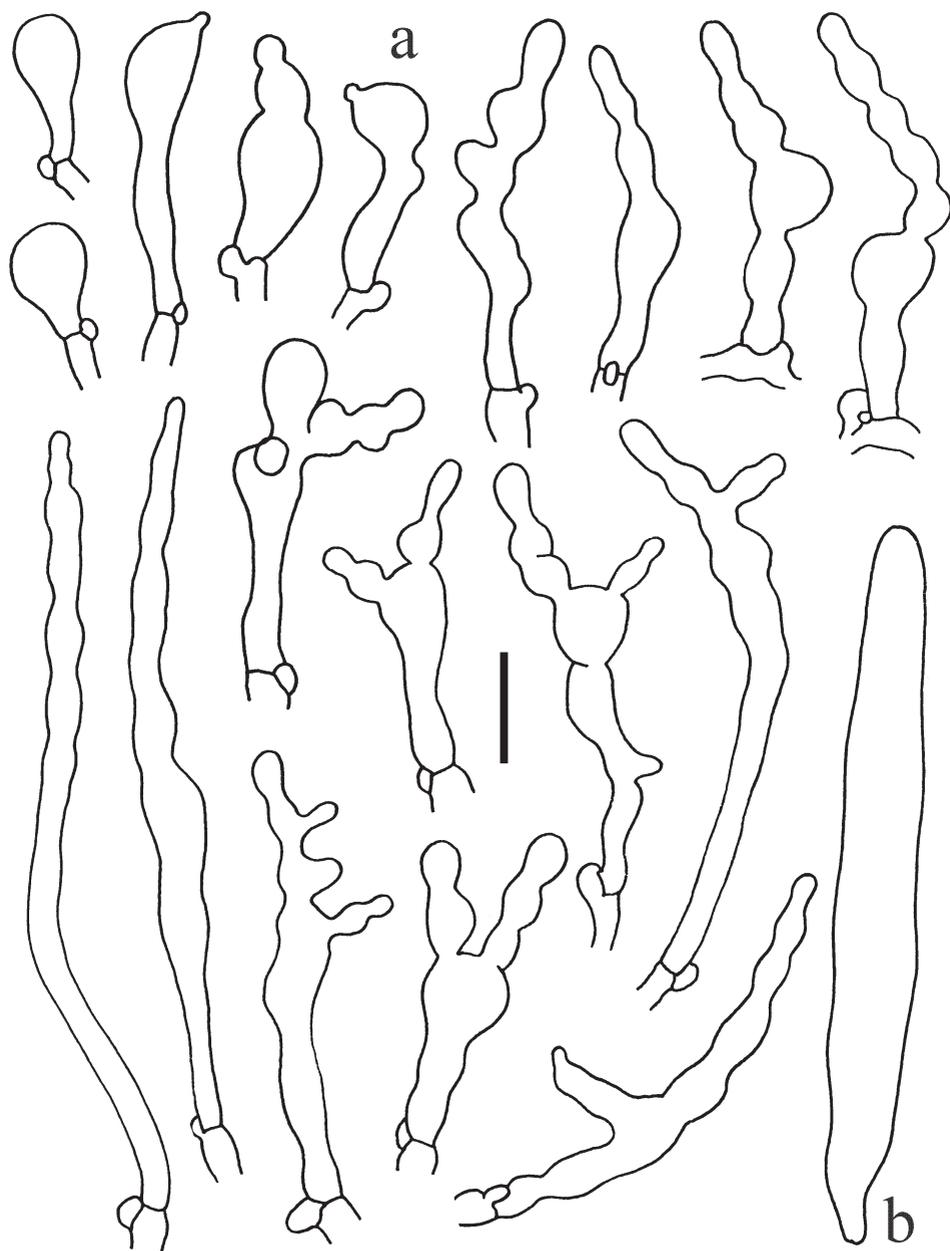
More than 40 *Flammulina* specimens deposited in herbarium M were studied in 2004. Among them 3 specimens of *F. rossica* were identified. The micromorphological characters were observed in dried material under an Olympus CX41 light microscope in oil immersion. Fragments of lamellae, stipe and pileipellis were examined in 5 % KOH, Melzer's reagent and a solution of Congo Red in ammonia (1 ml of 25 % ammonia dissolved in a filtrated solution of 1.5 g Congo Red and 50 ml distilled water). Extreme values of microcharacters were estimated as 10 and 90 percentiles of 30 measurements, the values in parentheses are the 5 and 95 percentiles. Q is the ratio of spore length and width. The abbreviations of herbaria are cited in accordance with the Index Herbariorum (Holmgren et al. 1990). Literature data on specimens are presented in their original form.

#### RESULTS AND DISCUSSION

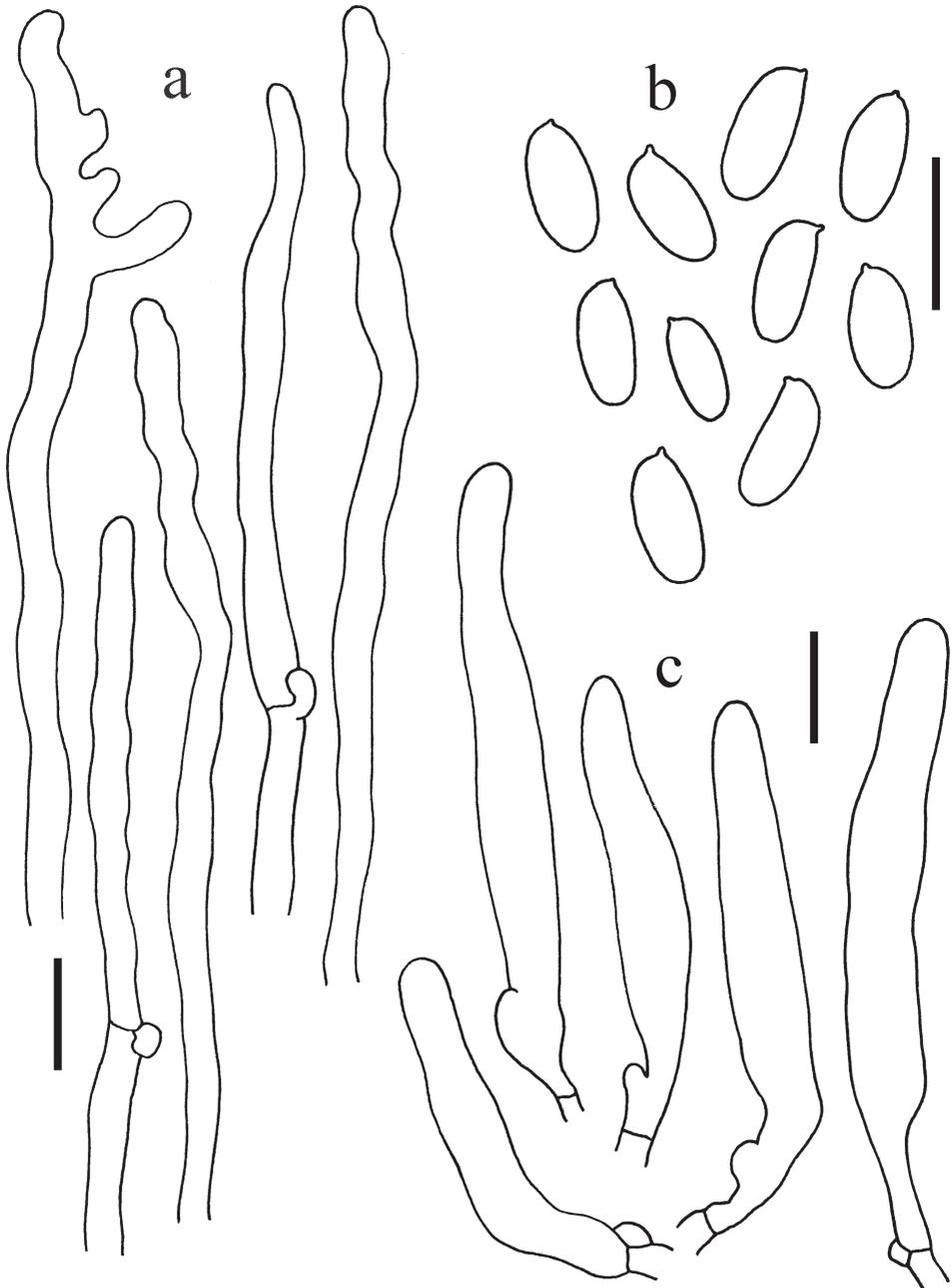
***Flammulina rossica*** Redhead et R. H. Petersen, Mycotaxon 71: 290, 1999.

**Holotype:** Russia, Primorski Terr., Dist. Ternai, Sichote Alin Biosphere Reserve, Meise, on *Populus* log, 12 Sept. 1990, R. H. Petersen 3232 (TENN 49489).

**Macromorphological characters:** Redhead and Petersen (1999), Petersen et al. (on-line). **Micromorphological characters** are based on material studied by us (see below). Basidia 4-spored. Spores (8.9–)9.2–11.9(–12.4) × (3.5–)3.6–4.9(–5) µm, av. 10.3 × 4.2 µm, Q = (1.95–)2–2.82(–2.99), av. Q = 2.45, narrowly ellipsoid to almost cylindrical, smooth, thin-walled, inamyloid, hyaline, with short and small hilar appendage. Pleurocystidia 36–64 × 6–8 µm, mostly fusiform, rarely indistinctly lageniform, at the tips narrowed and obtuse, at the base constricted and often nodulose, with walls thickened towards the base. Cheilocystidia of similar shape and size as pleurocystidia (not measured). Pileipellis a transition between a trichoderm and a hymeniderm, composed of scattered pileocystidia and numerous ixohyphidia. Terminal cells of ixohyphidia up to 80 µm long, variable in shape and dimensions, mostly of three types: 1) prevailing moniliform cells with



**Fig. 1.** *Flammulina rossica* (M 0065365): a – terminal cells of ixohyphidia, b – pileocystidium. Scale bar = 10  $\mu$ m.



**Fig. 2.** *Flammulina rossica* (M 0065365): a – hyphal terminations on stipe surface, b – spores, c – pleurocystidia. Scale bars = 10  $\mu$ m.

distinct central or terminal globose inflation (5–9 µm) and frequently with up to three lateral nodules or short branches; 2) frequent moniliform cells, which are narrower (3–6 µm), longer, attenuated and often branched; 3) occasional are sphaeropedunculate cells without lateral nodules, regular in shape. Pileocystidia of similar shape and size as pleurocystidia. Surface of stipe made up of long cylindrical hyphae, only few terminal cells shorter than 100 µm, 3–5 µm thick, with thickened walls, especially towards base, with brown pigment, mostly even, at most moniliform in terminal part; branched or nodulose terminal cells scarce. Caulocystidia scarce or absent. Hyphae in all tissues with clamp connections (Figs. 1, 2).

Delimitation of the species. *Flammulina rossica* Redhead et R. H. Petersen is one of the recently described *Flammulina* species of the Northern Hemisphere, which further include *F. elastica* (Lasch) Redhead et R. H. Petersen, *F. fennae* Bas, *F. mexicana* Redhead, Estrada-Torres et R. H. Petersen, *F. ononidis* Arnolds, *F. populicola* Redhead et R. H. Petersen, and *F. velutipes* (Curtis) Singer (Hughes et al. 1999).

Macromorphologically, basidiocarps of *F. rossica* are similar to those of *F. populicola* and *F. velutipes*, although they possess a very pale pileus – whitish to yellowish ochraceous (Redhead and Petersen 1999). The delimitation of the species is based on micromorphological characters. The species is distinctive by the combination of characters of the pileipellis (a more or less hymeniform suprapellis with typical pileocystidia) and spores (elongated, ellipsoid or cylindrical at times; for size, see Tab. 1) (Redhead and Petersen 1999).

Our observations have shown that the size and shape of the spores is more variable than that described by Redhead and Petersen (1999) in the original description: the spores might be longer (M 0065361) as well as narrower (M 0065415). In general, our measurements more or less agree with those presented by the authors (Redhead and Petersen 1999) in the Commentary (i. e. commentary text after the Latin diagnosis, p. 290–292). It is confusing that the average values of spore characters in the Latin diagnosis are smaller and out of the range of values given in the Commentary (Tab. 1).

The large variation in spore characters and the shape of the ixohyphidia makes identification difficult in some instances, as was pointed out by Redhead and Petersen (1999) and Petersen et al. (on-line). For example, the Q value of spores of the collection from South Dakota (M 0065415) is in the variability range of *F. elastica* f. *longispora* (Bas) Redhead et R. H. Petersen as estimated by Redhead and Petersen (1999).

According to our experience, the distinctive character useful for delimiting *F. rossica* from related taxa should be the presence of sphaeropedunculate or broadly clavate ixohyphidia. We have observed several collections of *F. elastica* identified according to ITS (Adamčík et al. in preparation), and if this type of ixohyphidia is present in *F. elastica* (it might also be absent), it is scarce to occasional and always has lateral nodules.

**Tab. 1.** Comparison of values measured on spores of *Flammulina rossica* in the Latin diagnosis and Commentary by Redhead and Petersen (1999) and measurements of specimens from the München herbarium (M). The boldfaced values are averages; extreme values of our observations were estimated as the 5 and 95 percentiles.

Source of measurements of <i>F. rossica</i>	spore length	spore width	Q
Latin diagnosis (Redhead and Petersen 1999)	7.4– <b>8.88</b> –11.0	3.8– <b>4.21</b> –4.5	<b>2.04</b>
Commentary (Redhead and Petersen 1999)	9.2–10.3	3.9–4.5	2.05– <b>2.3</b> –2.58
Germany (M 0065361)	9.7– <b>11.2</b> –12.6	4.1– <b>4.3</b> –5.0	2.13– <b>2.62</b> –3.06
Germany (M 0065365)	8.9– <b>9.9</b> –11.1	4.1– <b>4.6</b> –5.0	1.91– <b>2.16</b> –2.48
South Dakota (M 0065415)	8.5– <b>9.6</b> –10.4	3.4– <b>3.8</b> –4.1	2.32– <b>2.56</b> –2.88

*Flammulina rossica* and *F. elastica* are not only morphologically similar. Mating experiments gave the results that isolates of *F. rossica* and *F. elastica* were partially compatible with one another, but incompatible with those of other taxa of the genus (Petersen et al. 1999).

Based on ribosomal ITS sequences, *F. rossica* was found in the large clade with *F. mexicana*, *F. populicola* and *F. fennae* but not with *F. elastica* as might have been expected according to the results of mating experiments (Hughes et al. 1999, Petersen et al. on-line).

Using restriction enzymes (Hae III and Bst F51), isolates of *F. rossica* showed a pattern of 2–1, which was unique in the tested taxa of the genus *Flammulina* (Methven et al. 2000, Petersen et al. on-line).

Recently, Hughes and Petersen (2001) reported hybridization between *F. velutipes* and *F. rossica* which resulted in a homogenized ribosomal repeat, containing elements of both parents.

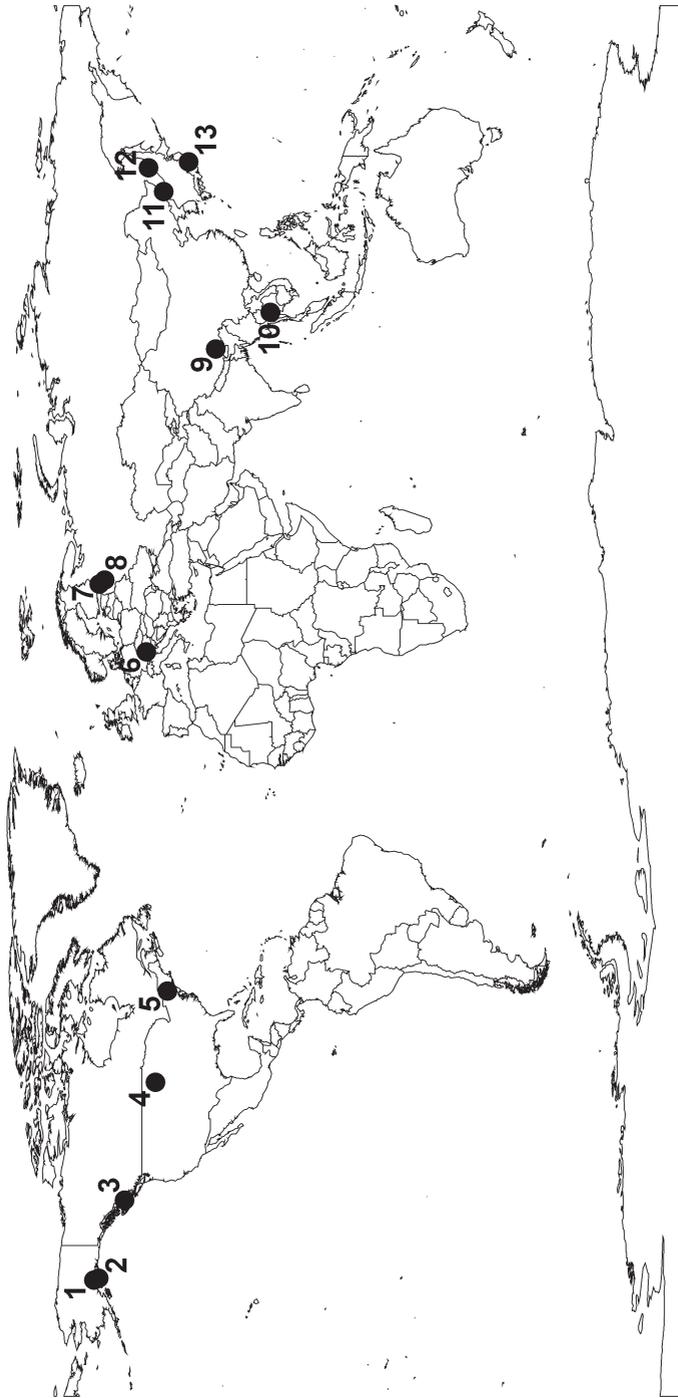
**Ecology and distribution.** The information is based on material studied and literary data (see below). *Flammulina rossica* was found on trunks of *Alnus* sp., *Betula* sp.?, *Populus* sp., *Salix amygdaloides*, *S. caprea* and *Salix* sp., from July to January.

*F. rossica* is hitherto known from USA (Alaska, South Dakota, New York State), Canada (British Columbia), Germany, Russia, Tibet, Thailand and Japan (Fig. 3).

Although we have studied *Flammulina* specimens also from other countries in Central Europe (Austria: W, Czech Republic: BRNM, Slovakia: BRA, SAV, SLO), we have not confirmed *F. rossica*.

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**Fig. 3.** The world distribution of *Flammulina rossica*. 1. Alaska (Anchorage, Valley of Moon Park), 2. Alaska (Seward), 3. British Columbia, 4. South Dakota (Northville), 5. New York State, 6. Germany (Nationalpark Berchtesgaden; 2 specimens), 7. Russia (St. Petersburg), 8. Russia (Valdai Reserve in Novgorod Region), 9. Tibet (Changdu Xian; 2 localities), 10. Thailand, 11. Russia (Vladivostok), 12. Russia (Sichote Alin Biosphere Reserve), 13. Japan.



### Material studied of *Flammulina rossica*

- Germany, Nationalpark Berchtesgaden, b. Hintersee, MTB 8343/3, alt. 800 m, auf liegenden Stamm von *Salix caprea*, 5 Oct 1981, leg. Schmid-Heckel (M 0065365 as *Flammulina velutipes*).
- Germany, Nationalpark Berchtesgaden, Hirschbichlstraße oberhalb Wildfütterungsplatz, MB 8442/2, alt. 860 m, am *Alnus*, 30 Dec 1981, leg. Schmid-Heckel (M 0065361 as *Flammulina velutipes*).
- USA, Dakota, S. D., Northville, on *Salix amygdaloides*, 3 Jan 1927, leg. J. F. Brenckle (M 0065415 as *Collybia velutipes*).

### Literary data on *Flammulina rossica*

- Russia, Primorski Terr., Dist. Ternai, Sichote Alin Biosphere Reserve, Meise, on *Populus* log, 12 Sep 1990, leg. R. H. Petersen 3232 (TENN 49489) (Redhead and Petersen 1999).
- Russia, Terr. Primorsk: vic. Vladivostok, Sep 1994, leg. I. Bullakh (CULTENN “Bullakh”) (Hughes et al. 1999).
- Russia, Valdai Reserve in Novgorod Region (Petersen and Hughes 2007).
- Russia, St. Petersburg (Petersen et al. on-line).
- Canada, coastal British Columbia (Petersen et al. on-line).
- USA, Alaska, Anchorage, Valley of the Moon Park, on log of ?*Betula*, 17 Sep 1995, leg. J. Johnson and R. H. Petersen 8195 (TENN 54170) (Redhead and Petersen 1999).
- USA, Alaska, vic. Seward, Exit Glacier Rd., 60° 11.037' N, 149° 33.580' W. On *Salix*, 14 Sep 1995, leg. K. W. Hughes and R. H. Petersen 8171 (TENN 54169; CULTENN 8171) (Hughes et al. 1999).
- USA, New York State, leg. T. J. Baroni (Petersen and Hughes 2007).
- Thailand, northern Thailand (Petersen and Hughes 2007).
- Tibet (Xizang), Changdu Xian, NE of the city of Changdu (Chamdo) along the Lancang Jiang (Mekong River – “Zachu” in Tibetan) on road (highway 317) to Jianga (Gyamda); at Qiongka, near Qiongka Diaoqiao (Qiongka suspension bridge). Scrub and heavily grazed vegetation along river and adjacent steep slopes; shrubs thorny or aromatic. 31° 18' 9" N, 97° 9' 35" E; 3290 m. On *Salix*, 27 July 2004, leg Z. W. Ge 190 (Anonymus on-line).
- Tibet (Xizang), Changdu Xian, road (highway 317) from Changdu (Chamdo) to Riwoqe (Riwoche) on E side of Zhuoga-La (pass). *Picea* forest and adjacent stream margin. 31° 4' 16" N, 96° 58' 29" E; 4215 m. On *Salix*, 7 Aug 2004, leg Z. W. Ge 296 (Anonymus on-line).
- Japan, Japanese strains (Petersen and Hughes 2007).

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### REFERENCES

- ANONYMUS (on-line): Biodiversity of the Hengduan Mountains and adjacent areas of south-central China. – [http://hengduan.huh.harvard.edu/fieldnotes/specimens/specimen\\_detail.zpt?specimen\\_id=18962](http://hengduan.huh.harvard.edu/fieldnotes/specimens/specimen_detail.zpt?specimen_id=18962) [accessed 26 Feb 2008].

- HOLMGREN P. K., HOLMGREN N. H. and BARNETT L. C., eds. (1990): Index Herbariorum 1: The Herbaria of the World. – 693 p. Bronx, New York.
- HUGHES K. W. and PETERSEN R. H. (2001): Apparent recombination or gene conversion in the ribosomal ITS region of a *Flammulina* (fungi, *Agaricales*) hybrid. – *Mol. Biol. Evol.* 18(1): 94–96.
- HUGHES K. W., MCGHEE L. L., METHVEN A. S., JOHNSON J. E. and PETERSEN R. H. (1999): Patterns of geographic speciation in the genus *Flammulina* based on sequences of the ribosomal ITS-5.8S-ITS2 area. – *Mycologia* 91(6): 978–986.
- METHVEN A., HUGHES K. W. and PETERSEN R. H. (2000): *Flammulina* RFLP patterns identify species and show biogeographical patterns within species. – *Mycologia* 92(6): 1064–1070.
- PETERSEN R. H. and HUGHES K. W. (2007): Some agaric distribution patterns involving Pacific landmasses and Pacific Rim. – *Mycoscience* 48: 1–14.
- PETERSEN R. H., HUGHES K. W. and REDHEAD S. A. (on-line): The genus *Flammulina*, a Tennessee tutorial. – <http://fp.bio.utk.edu/Mycology/Flammulina/default.html> [accessed 26 Feb 2008].
- PETERSEN R. H., HUGHES K. W., REDHEAD S. A., PSURTSEVA N. and METHVEN A. S. (1999): Mating system in the *Xerulaceae* (*Agaricales*, Basidiomycotina): *Flammulina*. – *Mycoscience* 40: 411–426.
- REDHEAD S. A. and PETERSEN R. H. (1999): New species, varieties and combinations in the genus *Flammulina*. – *Mycotaxon* 71: 285–294.