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# Sarcodontia crocea (Basidiomycota, Polyporales) is unrelated to Spongipellis

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*Sarcodontia crocea* (Schwein.) Kotl. (Kotlaba 1953: 117), is a resupinate wood-inhabiting basidiomycete with waxy, dentateaculeate (toothed-spiny) hymenophore. This is characterized by the subulate, densely crowded spines, pale to sulphurchrome yellow, turning to ochre and amber-brown with age; and a remarkable unpleasant acid-sweetish-fruity odour of young basidiomes. The preferred habitat of the fungus is decayed wood under bark or in cavities of old, unmanaged apple trees (*Malus* spp.), rarely other broadleaved hosts (Bernicchia and Gorjón 2010, Breitenbach and Kränzlin 1986).

The genus *Sarcodontia* was established for *Sarcodontia mali* Schultzer (Schultzer *et al.* 1866: 41), but later the name was synonymized with *Hydnum setosum* Pers. (Persoon 1825: 213). However Kotlaba (1953) revealed *Sistotrema croceum* Schwein. (Schweinitz 1822: 102) is the oldest validly published name of the species. The genus had been traditionally kept as monotypic, the most of the *Sarcodontia* combinations published till 2001 were later synonymized with other names and are not accepted. In 2001, Spirin combined genus *Spongipelis* Pat. (Patouillard 1887: 140) under synonymy of *Sarcodontia* due to similar spores with cyanophilous reaction (Spirin 2001). *Spongipellis* is a polyporoid genus of annual, mostly pileate basidiomes with white to cream, mostly duplex context, poroid or dentate hymenophore and globose or ellipsoid, thick-walled and cyanophilous basidiospores, typified with *Spongipellis spumeus* (Sowerby) Pat. (Patouillard 1887: 140). Nevertheless, *S. crocea* has basidiospores with cyanophilous reaction of variable intensity–e.g. Bernicchia and Gorjón (2010) treated *S. crocea* as acyanophilous. Moreover, the waxy consistency and bright yellow colour of *S. crocea* are not typical for *Spongipellis*, therefore, I decided to test phylogenetic relations between *S. crocea* and *Spongipellis*.

Macroscopic descriptions of examined specimens were based on dried basidiomes. Herbarium abbreviations followed Thiers (2016). Authors of fungal names were cited according to the recommendations of the Authors of Fungal Names (http://www.indexfungorum.org/AuthorsOfFungalNames.htm). Microscopic features were described from dried material mounted in 10% KOH, Melzer's reagent and cotton blue using an Olympus BX-50 light microscope (Tokyo, Japan) with a magnification of 1000× and Motic Images Plus 2.0 (Hong Kong, China) software.

To present the size range of the basidiospores, 5% of the measurements were excluded from each end of the range and are given in parentheses. For basidiospores, the factors Q (mean of length/ width quotients of any one spore in single specimen) and Q mean (average value of all measured specimens) are used.

DNA was extracted from dried basidiocarps using the PowerSoil DNA Isolation Kit (MoBio). DNA fragments encompassing the internal transcribed spacer (ITS) and large subunit-28S (LSU) regions of nuclear ribosomal RNA gene were amplified as previously described (Tomšovský 2012, Tomšovský *et al.* 2010). Amplified DNA was purified and sequenced by Macrogen (Korea).

The LSU and ITS sequence datasets were enriched with previously published sequences of Polyporales including examples of *Spongipellis* spp. (Floudas and Hibbett 2015, Tomšovský 2012, Westphalen *et al.* 2015), three species and the sequences were aligned in Mafft v.7 using a Q-INS-i strategy (Katoh and Standley 2013). The outgroups were selected as follows: the three sequences from Russulales–LSU dataset and *Fomitopsis pinicola* (Polyporales, Antrodia clade)–ITS dataset.

The Bayesian analyses were run in MrBayes 3.2.6 (Huelsenbeck and Ronquist 2003). Likelihood settings from the best-fit model (GTR+I+G =LSU dataset; TIM2+I+G=ITS dataset) were selected using the Bayesian information criteria in jModelTest2 (Darriba *et al.* 2012). The four chains were run for 10 million generations. The burn-in value (10 % of trees) was estimated in Tracer v.1.5 (http://beast.bio.ed.ac.uk). Sampling frequency was set to every 100th generation. The Maximum likelihood (ML) analyses were performed with RAxML-HPC v. 8 (Stamatakis 2014) with a GTRCAT model of evolution. Nodal support was determined by non-parametric bootstrapping (BS) with rapid bootstrapping option setting number of replicates automatically (the 456 replicates of the LSU dataset and 156 replicates of the ITS dataset).

The LSU dataset included a total of 1082 positions; among these positions, 736 were conserved, 332 were variable and 82 were singleton sites, as determined in MEGA 6.06. (www.megasoftware.net). The ITS dataset included 833 positions

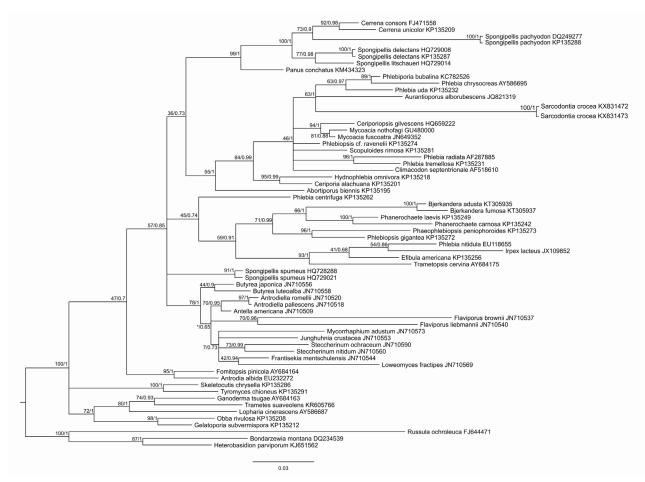
(333 conserved, 371 variable and 89 singleton). The Genbank accession numbers of the new sequences are KX831470-KX831473.

The morphological characters of the sequenced specimens are as follows and are in agreement to the description in literature (Bernicchia and Gorjón 2010, Breitenbach and Kränzlin 1986): Basidiomes are resupinate to effused, adnate, forming patches several centimeters in extent, sulfur-yellow to ferrugineous brown (turning dark red in KOH), waxy, hymenophore with teeths-spines 2-10 (-15 in literature) mm long, up to 1 mm thick.

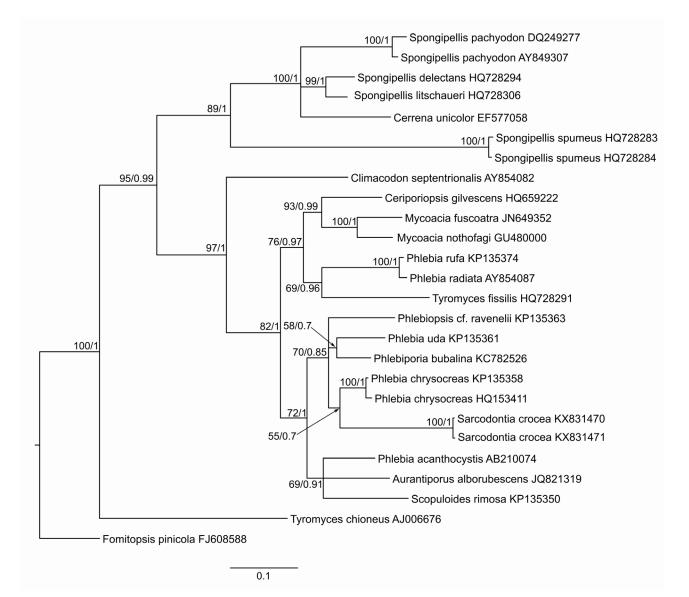
Hyphal system monomitic-pseudodimitic; generative hyphae with clamps (2.6-)3-5  $\mu$ m wide, thin-walled, often incrusted. Basidia clavate with basal clamp and four sterigmata, 20-30 × 4-6  $\mu$ m. Basidiospores subglobose-ellipsoid, often drop-shaped, acyanophilous to weakly cyanophilous, negative in Melzer's reagent, (4.5–)4.8–6.1(–6.3) × (2.9–)3–4.4  $\mu$ m; mean values=5.37×3.83, Q= 1.37–1.43, Q mean=1.40 (n= 83/4).

Materials studied:—CZECH REPUBLIC. Brno, Líšeň, *Malus domestica*, 25 August 2009, *A. Vágner* (BRNM 721609; Genbank: KX831470 and KX831472). Hodonín, Nechvalín, *Malus domestica*, 26 July 2012, *R. Maňák* (BRNM 761841; Genbank: KX831471 and KX831473). Hodonín, Veselí nad Moravou, *Pyrus* sp., 17 January 2013, *R. Maňák* (BRNM 766575). Hodonín, Velká nad Veličkou, National Nature reserve Zahrady pod Hájem, *Malus sp.* 1 May 2016, *R. Maňák* (BRNM 781256).

The results of phylogenetic analyses (Fig. 1, Fig. 2) confirmed clear separation of *S. crocea* from *Spongipellis* spp. and therefore *Spongipellis* spp. should not be kept in *Sarcodontia*. *S. crocea* belongs to the phlebioid clade but its exact position remains unresolved. The closely related species are *Phlebia chrysocreas* (Berk. & M.A. Curtis) Burds. (Lombard *et al.* 1975: 497), *Phlebia uda* (Fr.) Nakasone (Nakasone 1997: 72) and *Phlebiporia bubalina* Jia J. Chen, B.K. Cui & Y.C. Dai (Chen and Cui 2013: 569). This result supports the opinion of Jülich (1981), who placed *Sarcodontia* in family *Phlebiaceae*, currently treated as synonym of *Meruliaceae* (Cannon and Kirk 2007). Consequently, the combinations of *Spongipellis* species in *Sarcodontia*: *S. delectans* (Peck) Spirin (2001: 68), *S. pachyodon* (Pers.) Spirin (2001: 68), *S. spumea* (Sowerby) Spirin (2001: 68) and *S. unicolor* (Schw.) Zmitr. & Spirin (Zmitrovich *et al.* 2006: 25) do not respect the phylogeny of the species, although they have been occasionally used (e.g. Luchi *et al.* 2011).



**FIGURE 1.** A phylogenetic tree obtained from Bayesian analysis of the LSU alignment. Numbers at branch nodes indicate Maximum Likelihood bootstrap proportions (left; values <50 are replaced with asterisk) and Bayesian posterior probability values (right). The bar indicates number of expected substitutions per position.



**FIGURE 2.** A phylogenetic tree obtained from Bayesian analysis of the ITS alignment. Numbers at branch nodes indicate Maximum Likelihood bootstrap proportions (left) and Bayesian posterior probability values (right). The bar indicates the number of expected substitutions per position.

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