

On the identity of *Lachnum winteri* (Ascomycota, *Helotiales*)

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Syntypes of *Lachnum winteri* (specimens of the exsiccatae collection Rehm, Ascomyceten, No. 113) deposited in the herbaria S, K and M were examined. The syntypes contain the same species with short-stalked apothecia possessing whitish, pale yellow to yellow, sparsely warted and apically attenuated hairs corresponding to the genus *Albotricha*. The species differs from *A. albotestacea* not only in the colour of the apothecia and frequent presence of whitish subicular hyphae at the base of the stalks, but also in the structure of the ectal excipulum. The thinner ectal excipulum cell walls are considered to be the main character distinguishing the species from *A. albotestacea*. The species is regarded to be a good member of the genus *Albotricha*, hence the new combination *A. winteri* is proposed. A lectotype is designated and a description, line drawing and photographs are presented. The concept of *L. winteri* introduced by J. Velenovský is also discussed.

Key words: lachnoid fungi, *Albotricha*, type study, lectotypification, taxonomy.

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V rámci předložené studie byly prozkoumány syntypy *Lachnum winteri* (položky z Rehmovy exsikátové sbírky Ascomyceten, No. 113) uložené v herbářích S, K a M. Syntypy představují ten samý druh s krátce stopkatými apothecii s bělavými, bledě žlutými až žlutými, řídké bradavčitými a nahoře ztenčenými chlupy, odpovídajícími rodu *Albotricha*. Tento druh se liší od *A. albotestacea* nejen barvou apothecia a častou přítomností bělavých subikulárních hyf na bázi stopek, ale též strukturou vnějšího excipula. Tenčí stěna buněk vnějšího excipula je považována za hlavní znak odlišující tento taxon od *A. albotestacea*. Jelikož je považován za dobrý druh rodu *Albotricha*, je navržena nová kombinace *A. winteri*. V práci je vystaven lektotyp a prezentován popis, kresby mikroznaků a fotografie. Diskutován je též koncept *L. winteri* v pojetí J. Velenovského.

INTRODUCTION

The *Lachnaceae* include primarily ascomycetes with relatively tiny, scattered or gregarious apothecia with warted or in some parts smooth hairs of pale, less frequently reddish, olivaceous or brown colours (Rehm 1893, Suková 2005, Baral in

Jaklitsch et al. 2016). The hairs or parts of hairs are densely or irregularly covered with warts which are sometimes enlarged and coloured, the colour and partly also the mass of warts being often soluble in potassium hydroxide (e.g. Chlebická 2013).

Rehm (1893) distinguished *Lachnum winteri* from *L. albotestaceum*. The main differences according to his descriptions of these species are the gregarious and sulphur-coloured apothecia in *L. winteri*. His descriptions were based on the type material of *L. winteri* from the Botanical Garden in Leipzig, on *Phragmites*, and two specimens of *L. albotestaceum* from Germany, on *Calamagrostis*. *Lachnum albotestaceum* is generally known from grasses and has pink apothecia and white or whitish to rusty hairs (Raitviir 1970, Sacconi 1986, Vesterholt 2000). Ellis & Ellis (1997) reported specimens with flesh-pink apothecia and pale yellow hairs from various grasses, including *Phragmites*. Höhnelt (1918) examined the type specimen of *L. albotestaceum* by making a cross-section of an apothecium. The characters given by him (structure of the apothecium and excipulum tissues) are important for the taxonomy of this group of species, although they are hitherto known in just a few of them (e.g. Ono & Hosoya 2001) because of a lack of modern descriptions and also because of their tiny apothecia and the fact that the species are not fairly common.

According to Ellis & Ellis (1997), the most common inoperculate discomycetes on *Phragmites australis* are *Lachnum acutipilum*, *L. controversum*, *Lophodermium arundinaceum* and *Trichobelonium kneiffii*. *Lachnum acutipilum* differs from *L. albotestaceum* and *L. winteri* by its longer ascospores (Rehm 1893, Raitviir 1970, Vesterholt 2000, Baral on-line). *Lachnum controversum* differs from *L. albotestaceum* and *L. winteri* by its shorter and obtuse hairs (Rehm 1893, Dennis 1949) and also by the reddening of the apothecia due to numerous refractive vacuolar guttules in fresh state (Baral in Baral & Krieglsteiner 1985). Raitviir (1970) established the genus *Albotricha*, to which he transferred *A. acutipila* and *A. albotestacea*. The genus is characterised by tapering hairs with scattered warts in their non-apical part (Raitviir 1970, Huhtinen 1985, Ono & Hosoya 2001).

Lachnum winteri in its original sense had not been reported in the literature after it was last mentioned by Rehm (1893). However, it was misinterpreted by some authors (Velenovský 1934, Svrček 1979, Baral in Baral & Krieglsteiner 1985). The aim of the present study is to examine the original concept of *L. winteri* and to define the differences between *L. winteri*, *A. albotestacea* and *L. winteri* ss. auct.

MATERIAL AND METHODS

Herbarium specimens were prepared under a stereomicroscope using a small drop of tap water and studied in 3% KOH at a magnification of 1000× under an Olympus BX-51 light microscope using an oil immersion lens. Besides 3% KOH, the microscopic characters of the hairs were also examined in tap water. The reactions of the asco-apical ring were tested using Melzer's reagent (further referred to as MLZ; Huhtinen 1987) and Lugol's solution (further referred to as IKI; Baral 1987) without KOH pretreatment. The colours of the apothecia and hairs were examined in dried state under a stereomicroscope illuminated with an Olympus KL 1500 LCD light source and described according to Kornerup & Wanscher (1981). The lipid content in the ascospores was characterised using a linear scale from 0 to 5 (0 = devoid of lipid; 5 = maximum lipid content) following Baral (1992).

Examined specimens are deposited in the herbaria S (Swedish Museum of Natural History, Stockholm, Sweden), K (Royal Botanic Gardens, Kew, UK), M (Botanische Staatssammlung München, Munich, Germany) and PRM (National Museum, Prague, Czech Republic).

RESULTS

Albotricha winteri* (Cooke) Šandová, **comb. nov.*

Figs. 1, 2, 3

(MycoBank MB 826983)

Basionym: *Peziza winteri* Cooke, Grevillea 4(30): 67, 1875

≡ *Dasyscyphus winteri* (Cooke) Rehm, Ber. Naturhist. Ver. Augsburg 26: 30, 1881 [*Dasyscypha*"]

≡ *Trichopeziza winteri* (Cooke) Sacc., Discomyceteae et Phymatosphaeriaceae, p. 420, 1889

≡ *Lachnum winteri* (Cooke) Rehm, in Rabenh. Krypt.-Fl., Edn 2 (Leipzig) 1.3 (Lief. 41), p. 904, 1893

≡ *Lachnella winteri* (Cooke) Boud., Hist. Class. Discom. Eur., p. 124, 1907

Lectotype (designated here, MycoBank MBT 383721). Germany, Leipzig, Botanical Garden, on decaying shoots of *Phragmites australis*, July 1872, leg. G. Winter, S-F6139 (deposited in Swedish Museum of Natural History, Stockholm).

Etymology: *winteri* – dedicated to Heinrich Georg Winter, who collected the type material and is said to have prepared most of Rehm's exsiccatae of Fascicles I and II, i.e. Nos. 1 to 50 and 51 to 100, respectively, for distribution (Cooke 1875, see also Stevenson 1971).

Description (specimen S-F6139 unless otherwise stated, observations in 3% KOH unless otherwise stated). Dry apothecia 0.4–1.0 mm in diam., 0.3–0.7 mm high, with golden yellow [5-B7], orange [6-B8] to brown orange [6-C8] disc, outer surface covered by pale yellow [3-A5] to yellow [3-A6] hairs which are colourless to white at margin. Stalks of apothecia up to 0.3 mm wide and up to 0.2 mm long. Fragment of apothecium in K(M)157912 showing a ochre-yellow colour in KOH.

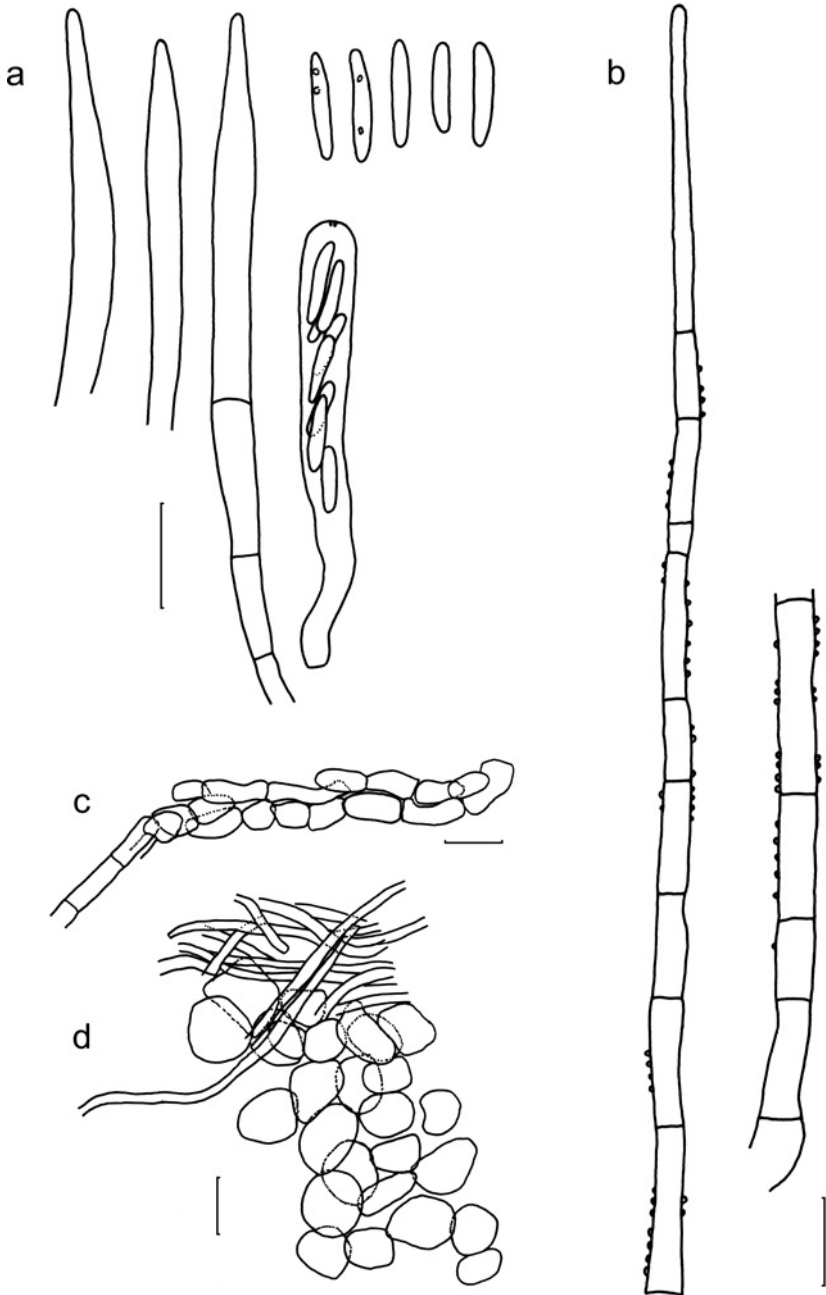


Fig. 1. *Albotricha winteri* (S-F6139, lectotype): **a** – paraphyses, ascus and ascospores; **b** – hair; **c** – cells of excipulum near margin and basal parts of hairs (left); **d** – medullary hyphae and cells of excipulum in lower flank area. Medium: 3% KOH. Scale bars = 10 μ m. Line drawings by M. Šandová.

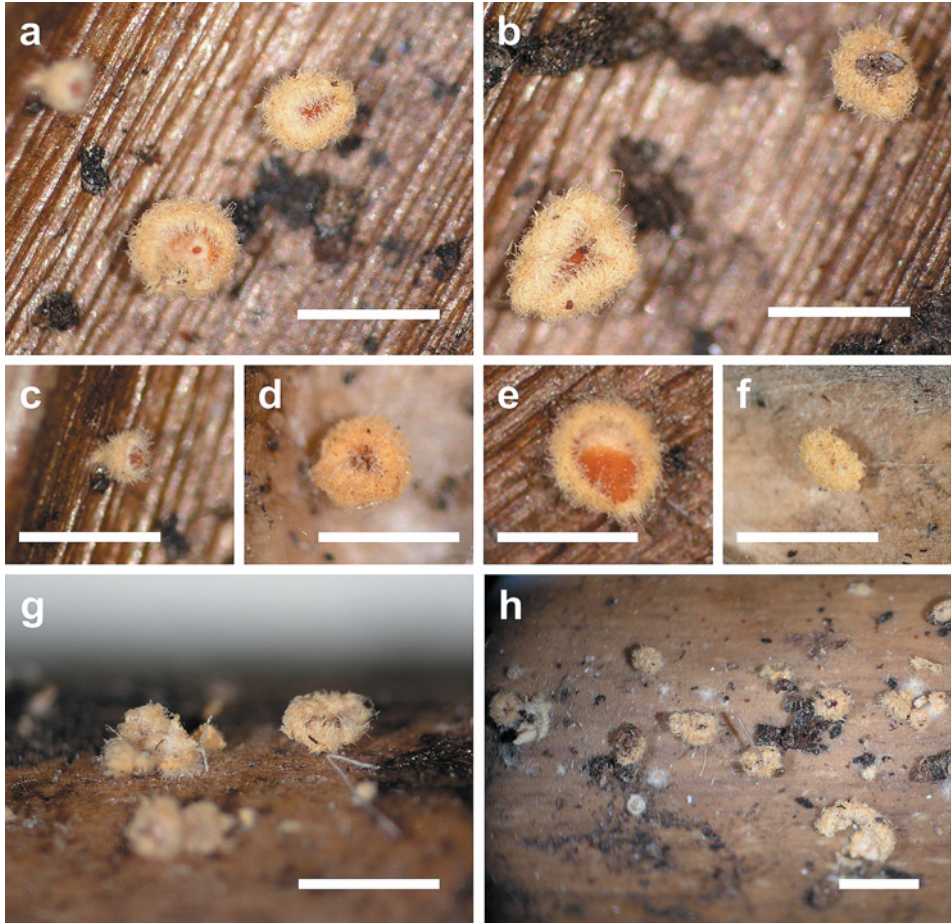


Fig. 2. Apothecia of *Albotricha winteri* in dried state: **a–e** – specimen S-F6139 (lectotype); **f–g** – specimen K(M)157912 (**g** – apothecia overgrown with a hyphomycete); **h** – specimen M-0229590. Scale bars = 1 mm. Photos by M. Šandová.

Hairs subhyaline, 125–200 μm long, septate in intervals of 8–20 μm (105–160 μm long and 6–9(10)-septate in M-0229590), 3.3–3.9 μm wide in lower part, 2.3–2.6 μm wide in apical part, with scattered to relatively dense warts in central part, subhyaline in water (M-0229590), with scattered, up to 1 μm broad warts and scattered larger, up to 4 μm broad warts or wall deposits.

Ectal excipulum hyaline to subhyaline, composed of isodiametric, up to 10–13 μm large cells with 0.6–0.9 μm thick cell wall. Medullary excipulum hyaline, composed of up to 2–3 μm wide hyphae.

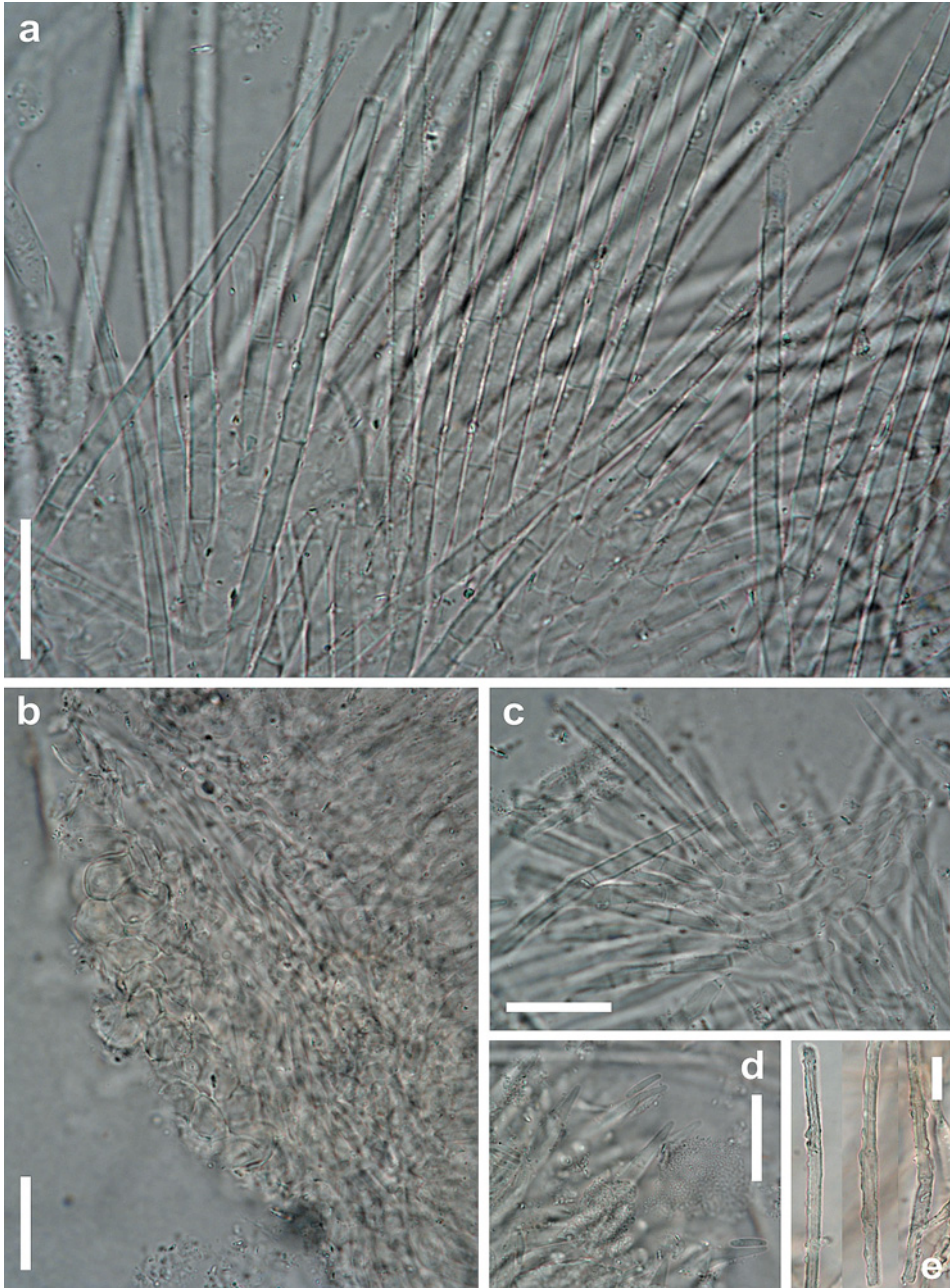


Fig. 3. *Albotricha winteri* (a–d: lectotype specimen S-F6139; e: specimen M-0229590): **a** – hairs; **b** – excipulum in cross-section; **c** – excipulum in marginal part of apothecium in cross-section; **d** – ascospores and apical parts of paraphyses; **e** – hairs. Medium: 3% KOH (a–d), tap water (e). Scale bars = 20 μm (a–d), 10 μm (e). Photos by M. Šandová.

Asci cylindrical, narrower in basal part, with rounded apex, $40\text{--}44 \times 4\text{--}5 \mu\text{m}$, apical ring (M-0229590) blue in IKI and MLZ without KOH pretreatment. Ascospores fusiform, hyaline, smooth, $7.8\text{--}9.8(10.8) \times 1.5\text{--}1.8 \mu\text{m}$, non-septate, with 0–7 small lipid bodies per ascospore, lipid content 0–1, less frequently up to 1–2. Paraphyses lanceolate, hyaline, smooth, $3.5\text{--}5 \mu\text{m}$ wide, exceeding the asci for $7.5\text{--}14 \mu\text{m}$.

Studied specimens (specimens under the name *Dasyscypha kneiffii* Wallr. from the exsiccatae collection Ascomyceten by Rehm, Fasc. 3, No. 113)

Germany. Leipzig, Botanical Garden, on decaying shoots of *Phragmites australis*, July 1872, leg. G. Winter [S-F6139 – lectotype; K(M)157912; M-0229590].

Part of specimen K(M)157912 contains old apothecia overgrown with a white hyphomycete, but the microcharacters observed in part of one of the apothecia are identical to *L. winteri* (non-thickened wall of cells of ectal excipulum, up to $125 \mu\text{m}$ long, $2.8\text{--}3.8 \mu\text{m}$ wide, 6–9-septate hairs, narrowed in their apical part; asci and paraphyses not observed probably because of the old apothecium; ascospores non-septate, $6.7\text{--}11.7 \times 1.3\text{--}1.8 \mu\text{m}$ in size).

DISCUSSION

Taxonomy

Lachnum winteri is here considered to be different from *Trichopeziza* and to belong to *Albotricha* because of its stalked apothecia and attenuate hairs (Fig. 5). It differs from *A. albotestacea* by the structure of its ectal excipulum (Figs. 4c–e, 6c–e), the abundant yellow larger warts or wall deposits on the hairs, the apothecium colour in dead state tending towards yellowish [3-A6 to 3-A7] in *A. winteri* vs. light orangish [5-4A to 5-4B] in *A. albotestacea* (Figs. 4a–b, 6a–b), and the frequent presence of whitish subicular hyphae at the base of stalks (Fig. 4b). The ectal excipulum of *A. albotestacea* is characterised by its thickened cell walls ($0.8\text{--}2.7 \mu\text{m}$ thick vs. $0.6\text{--}0.9 \mu\text{m}$ in *A. winteri*). The type material of *A. albotestacea* was not examined during this study, but information on the taxon was obtained from the protologue (Desmazières 1843) and from Höhnel's revision (Höhnel 1918). The specimen of *A. albotestacea* examined in the present study (Figs. 6–7) is identified on the basis of the thickened cells of its ectal excipulum (compare Höhnel 1918) and the colour of the apothecia (compare Desmazières 1843). The colours used by Desmazières (*testaceus* and *carneus*, i.e. *incarnatus*) were interpreted using Stearn (2004) and Saccardo (1894).

An ITS sequence was only obtained for *A. albotestacea* during this study (see below), although specimens of *A. winteri* deposited in the PRM herbarium were also examined. The sequence does not concur with available sequences of *A. albotestacea* in GenBank (AB481235, AB481236), which probably represent a different species.

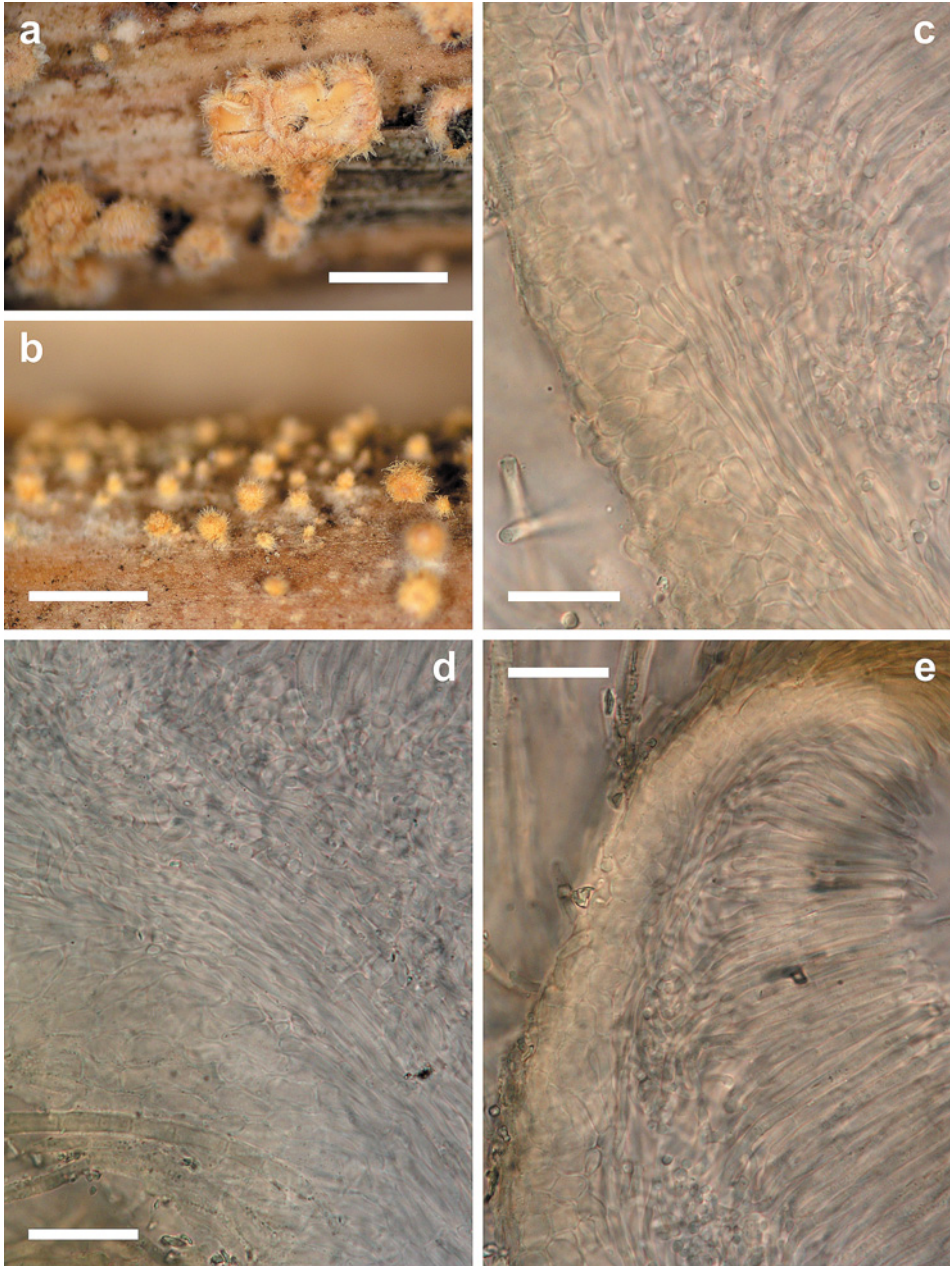


Fig. 4. *Albotricha winteri* (PRM 816192): **a–b** – dried apothecia; **c–e** – excipulum in cross-section. Medium: 3% KOH (c–e). Scale bars = 1 mm (a–b), 20 µm (c–e). Photos by M. Šandová.

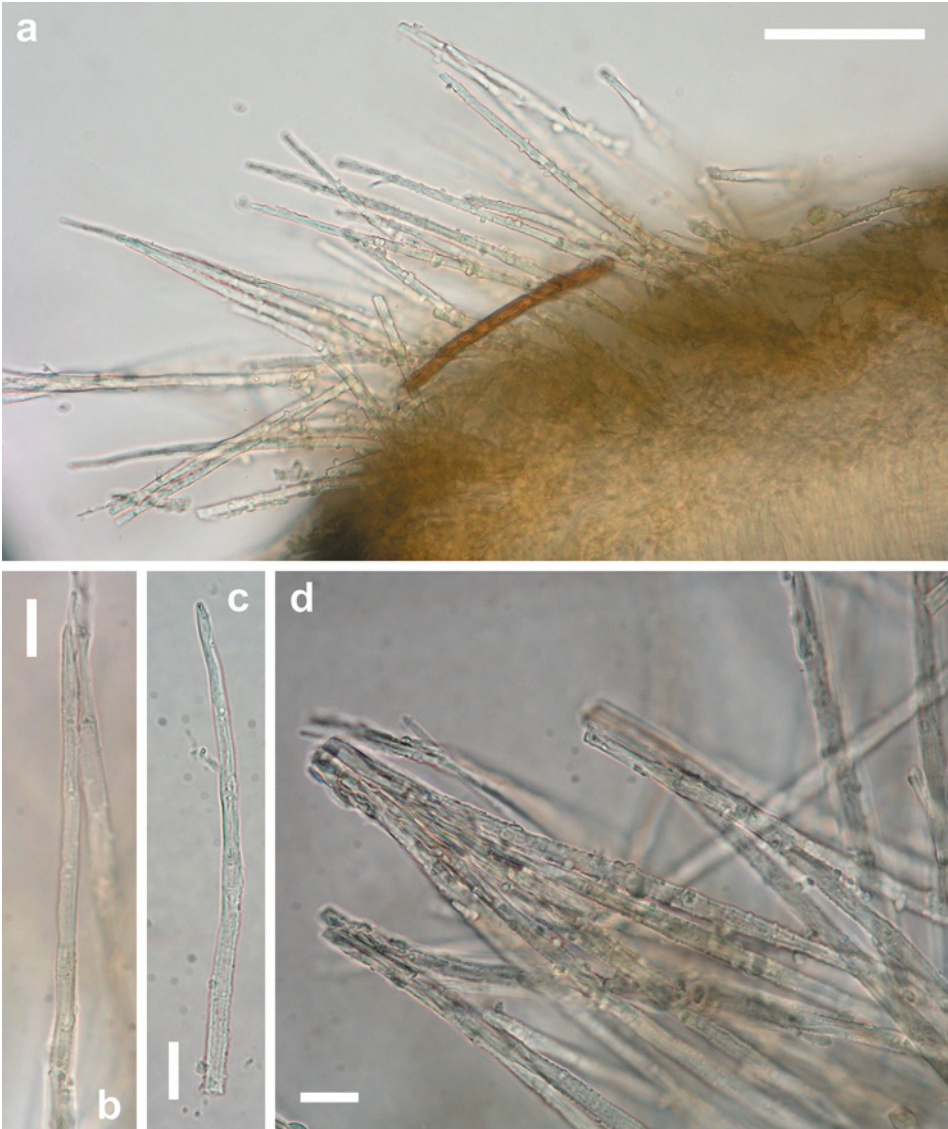


Fig. 5. *Albotricha winteri* (a: specimen PRM 819557; b–d: specimen PRM 816192): a–d – hairs. Medium: tap water. Scale bars = 50 μm (a), 10 μm (b–d). Photos by M. Šandová.

The character of the ascus bases was not determined with certainty in the type material of *A. winteri*, but according to the specimens deposited in the PRM herbarium, the asci arise from simple septa.

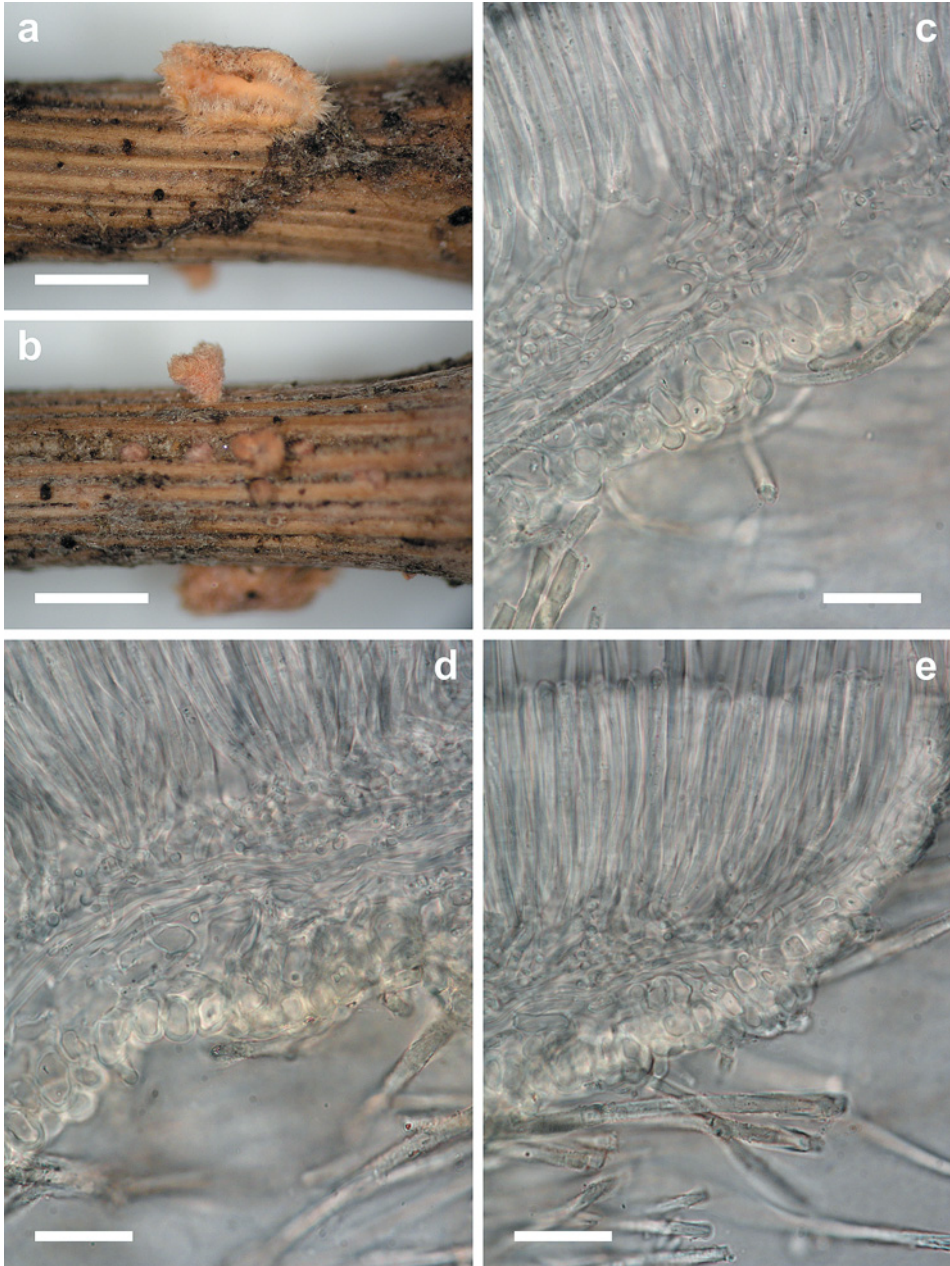


Fig. 6. *Albotricha albotestacea* (PRM 908618): **a–b** – dried apothecia; **c–e** – excipulum in cross-section. Medium: 3% KOH (c–e). Scale bars = 1 mm (a–b), 20 μ m (c–e). Photos by M. Šandová.

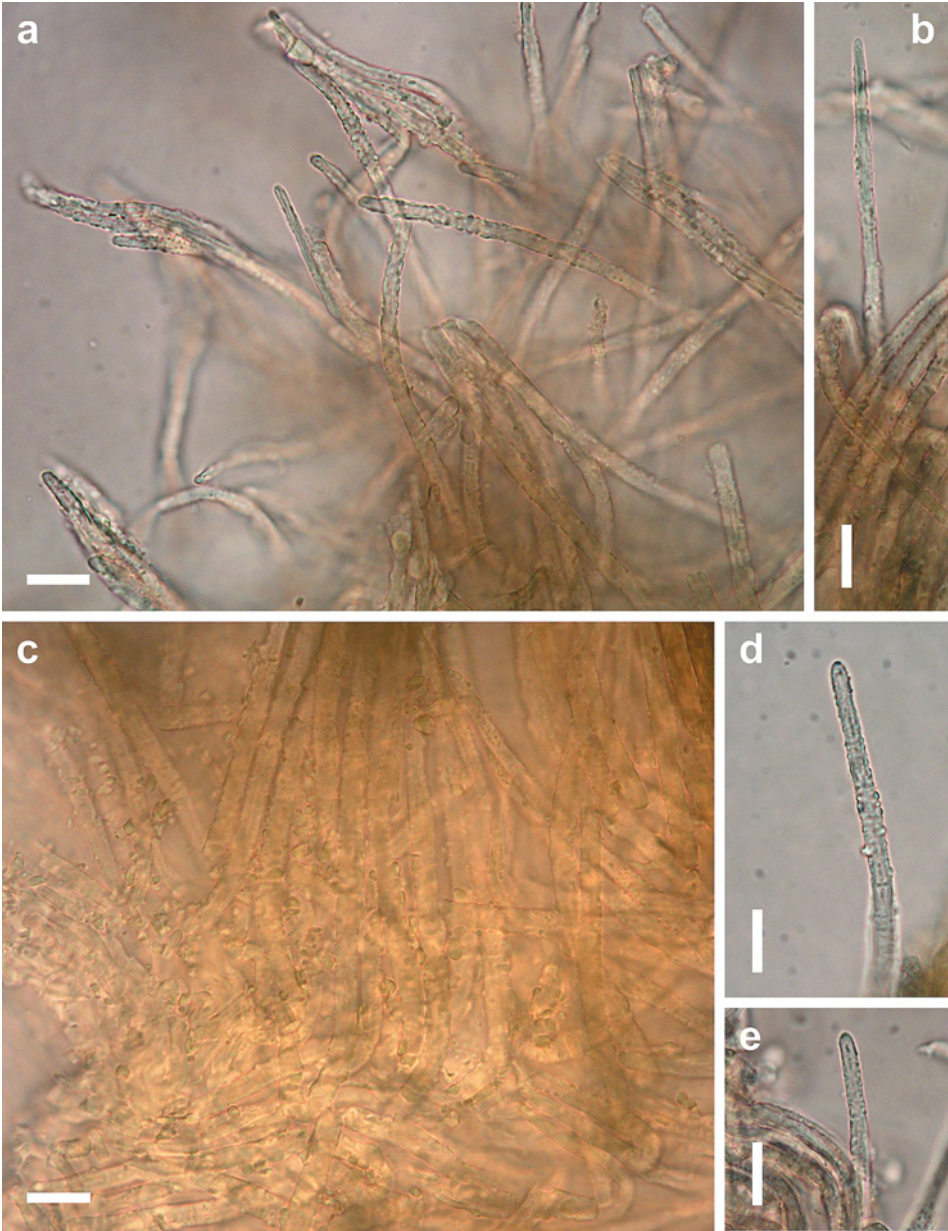


Fig. 7. *Albotricha albotestacea* (PRM 908618): **a–e** – hairs. Medium: tap water. Scale bars = 10 μm . Photos by M. Šandová.

Studied specimens illustrated in Figs. 4–5 (*A. winteri*)

Czech Republic. South Bohemia, Třeboň, *Alnetum* “U Jindřů”, on *Phragmites australis*, 13 April 1957, leg. J. Kubička, det. M. Svrček (as *Dasyscyphus albotestaceus*; PRM 816192). – South Bohemia, Vodňany, in verge of Podvinice pond, on *Phragmites australis*, 10 June 1977, leg. M. Svrček & J. Kubička, det. M. Svrček (as *D. albotestaceus*; PRM 819557).

Studied specimen illustrated in Figs. 6–7 (*A. albotestacea*)

Poland. SE Poland, Bieszczady National Park, ESE of Wołosate, S ridge of Mt. Rozsypaniec, alt. 1155 m, on grass, 10 August 2006, leg. & det. M. Chlebická (PRM 908618; GenBank no. LS997507).

Since a sequence was obtained from the specimen, a description is added. Fresh apothecia subsessile, broad, pale orange [5-A3 to 6-A3], relatively long-haired, dried apothecia 0.35–1.5 mm in diam., 0.3–0.65 mm high, with orange yellow [4-A5 to 4-A6] disc and outer surface covered by yellow-orange [4-A6 with 5-A4 to 5-A5 tint] to light orange [5-A4 to 5-B4] hairs, which are concolorous to white at margin. Stalks of apothecia in dried state up to 0.2 mm wide and 0.25 mm long. Hairs in 3% KOH subhyaline to hyaline, 8–10(13)-septate, 105–215 µm long, 3.0–3.9 µm wide, septate in intervals of 6–23(31) µm, with scattered (but frequently relatively dense) warts in central part; in water hyaline, yellowish or pale brownish yellow, relatively densely warted, apices warted or smooth, more or less attenuated, in some parts with scattered, larger, up to 3.8 µm wide warts or wall deposits. Ectal excipulum hyaline to yellowish, composed of isodiametric to ellipsoidal, angular cells with 3.4–11.3 × 2.8–6.8 µm large lumen, l/w ratio 1.05–2.3(2.6), cell wall 0.8–2.7 µm thick. Medullary excipulum hyaline, composed of up to 1.8–2.3 µm wide hyphae. Asci arising from simple septa, cylindrical, narrower in basal part, with rounded apex, 35–51 × 4–5 µm, apical ring blue in IKI and MLZ without KOH pretreatment. Ascospores narrowly fusiform, hyaline, smooth, 7.8–11.6 × 1.4–1.8 µm, non-septate, with 0–8 small lipid bodies per ascospore, lipid content 0–2. Paraphyses lanceolate, hyaline, smooth, 2.6–3.2 µm wide, exceeding the asci for 13.5–21 µm.

Nomenclature

The name *Peziza winteri* was mentioned for the first time by Cooke (1875). He introduced it for a species distributed by Rehm in 1872 as *Dasyscypha kneiffii*, Ascomyceten, No. 113. Cooke (1875) did not provide a description, but specimens of the Ascomyceten exsiccatae collection were distributed without descriptions on labels too. The labels of specimen No. 113 were checked in several herbaria (see under Specimens studied) and detailed information on the labels of this exsiccatae collection is also given by Lizoň (2016). Rehm’s Ascomyceten exsiccatae collection was not accompanied by printed descriptions or diagnoses in the time of its distribution and bears only names on the labels. Neither a description nor a diagnosis of distributed *Dasyscypha kneiffii* specimens is given in the list of exsiccatae published in the Flora journal (Rehm 1873). The text accompanying Fascicle III, which includes exsiccate No. 113, was published in *Berichte des Naturhistorischen Vereins in Augsburg* 26, 1881 (Stevenson 1971, Stafleu & Cowan 1983, Lizoň 2016). This is the place where a description for *Dasyscyphus winteri* is given (Rehm 1881, as *Dasyscypha*). The too general and unintended characterisation of the fungus given by Cooke (1875) may or may not be considered a diagnosis under Art. 38.1 (Turland et al. 2018), since Cooke (1875) only delineated a difference between *Peziza kneiffii* Wallr. ss. Rehm, for

which he proposed the name *Peziza winteri*, and *Eustegia arundinacea* (DC.) Fr. (syn. *Peziza kneiffii* Wallr.), with which it was confused by Rehm. *Peziza winteri* Cooke is used in the present study as a basionym, following the recent survey by Lizoň (2016). However, a request for a binding decision will be submitted to Taxon under Art. 38.4. (Turland et al. 2018) in order to obtain an official statement on whether the first valid publication of the species is in Cooke (1875) or not. In the latter case, the nearest valid publication would be in Rehm (1881).

Velenovský's concept

Velenovský (1934), followed by Svrček (1979) and Baral (in Baral & Krieglsteiner 1985), presented a different concept of *Lachnum winteri*. These authors used this name for a *Lachnum* similar to *L. controversum*, but containing yellow guttules in fresh paraphyses and hairs (Baral, op. cit.). Svrček (1979) did not provide a detailed description, but specimens identified by him as *L. winteri* deposited in the PRM herbarium contain a *Lachnum* which fully agrees with *Lachnum controversum* in its characters: dried apothecia up to 0.45 mm high, up to 0.7 mm in diam., stalked; hairs warted, cylindrical, with rounded apices, 40–65 × 3.8–4.2 µm; asci arising from simple septa, 39–44 × 4.5–5 µm, with euamyloid apical ring; ascospores 8–10.8 × 1.8–2.1 µm, containing a few small lipid bodies; paraphyses lanceolate, 2.9–3.6 µm wide, exceeding the asci for 11.5–20 µm.

Since Rehm's (1881) description of the type of *L. winteri* does not provide information on the character of the apothecia and the hair shape and length, the concept introduced by Velenovský and followed by the mentioned authors may be in agreement with this incomplete description. The revision of the exsiccatae specimens of *L. winteri* published in the present study, however, shows that the specimens agree with Rehm's later, more detailed description (Rehm 1893) and that the epithet *winteri* in its original meaning is not related to the above-mentioned yellow form of *Lachnum controversum*.

Studied specimen (*L. controversum*)

Czech Republic. Central Bohemia, Kosoř, "Kosořský rybníček", on *Phragmites australis*, 7 June 1953, leg. & det. M. Svrček (as *Dasyscyphus winteri*; PRM).

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