# Three new species of *ljuhya* (*Bionectriaceae, Hypocreales*) from metropolitan France, French Guiana and Spain, with notes on morphological characterization of *ljuhya* and allied genera



**Abstract:** Three new species of *ljuhya* are described and illustrated, based on material collected in metropolitan France, French Guiana and Spain. Based on morphological and phylogenetic comparison with the known *ljuhya* species as well as the related genera *Lasionectria* and *Lasionectriella*, we propose *l. salviicola*, *l. saulensis*, and *l. somiedoensis* as new species. The three species were cultured but only *l. salviicola* produced an acremonium-like asexual morph. Morphological characteristics distinguishing *ljuhya*, *Lasionectria* and *Lasionectriella* are discussed and illustrated. The new combination *Lasionectria boothii* is proposed and *Lasionectria lecanodes* (Ces.) Petch is re-instated.

Keywords: Ascomycota, ribosomal DNA, taxonomy.

**Résumé :** Trois nouvelles espèces d'*ljuhya* sont décrites et illustrées à partir de récoltes effectuées en Espagne, en France métropolitaine et en Guyane française. Sur la base d'une comparaison morphologique et phylogénétique avec les espèces connues d'*ljuhya* ainsi qu'avec les genres apparentés *Lasionectria* et *Lasionectriella*, nous proposons *I. salviicola*, *I. saulensis* et *I. somiedoensis* comme espèces nouvelles. Les trois espèces ont été cultivées mais seule *I. salviicola* a produit un stade asexué de type acremonium. Les caractéristiques morphologiques qui distinguent *ljuhya*, *Lasionectria* et *Lasionectriale* sont commentées et illustrées. La nouvelle combinaison *Lasionectria boothii* est proposée et *Lasionectria lecanodes* (Ces.) Petch est rétablie. **Mots-clés :** ADN ribosomal, Ascomycota, taxinomie.

## Introduction

The current concept of *ljuhya* Starbäck was established by Ross-MAN *et al.* (1999) to accommodate the bionectriaceous fungi formerly assigned to *Peristomialis* (W. Phillips) Boud. by SAMUELS (1988). After the revision of *l. vitrea* Starbäck, SAMUELS (1976) showed that this name was an earlier synonym of *Nectria peristomialis* (Berk. & Broome) Samuels [ $\equiv$  *Peristomialis berkeleyi* Boud.], the type species of *Peristomialis*, and thus reinstated *ljuhya*, with *l. peristomialis* (Berk. & Broome) Rossman & Samuels as type species.

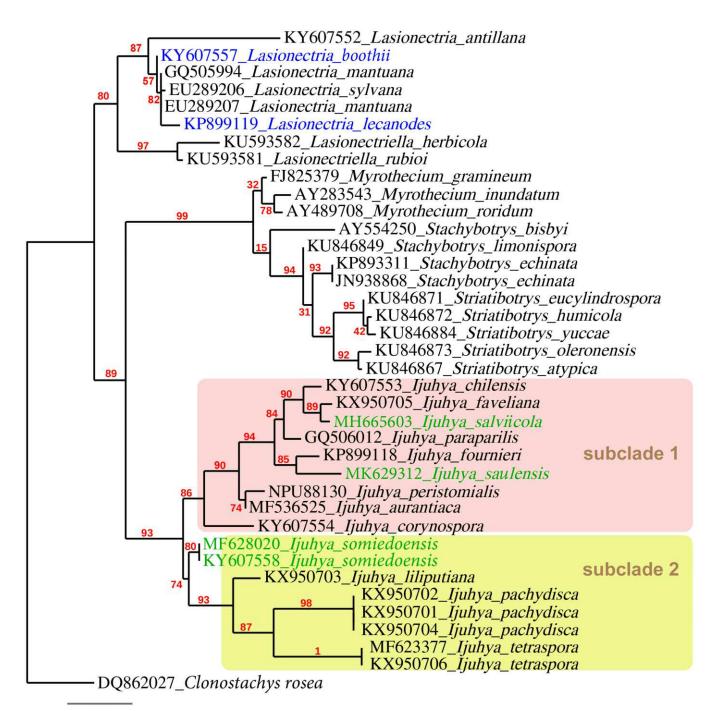
Within the Bionectriaceae, ljuhya is morphologically characterized by subglobose, usually light-coloured, non-stromatic, superficial ascomata with a discoid apex either lined by a crown of triangular fasciculate hairs or by intertwined hairs forming a prominent margin around the disc. Out of the ten species included by ROSSMAN et al. (1999), I. bambusina (Syd. & P. Syd.) Rossman & Samuels should probably be excluded, based on its minute, umber to blackish, scurfy, pyriform ascomata with acute apex (ROSSMAN et al., 1993). Since this date, twelve new species were described from both temperate and tropical regions by Fröhlich & Hyde (2000), Zhuang et al. (2007), Lechat & BARAL (2008), LECHAT & COURTECUISSE (2010), LECHAT & HAIRAUD (2012), LECHAT et al. (2015), LECHAT & FOURNIER (2017a), LECHAT et al. (2017c) and ASHRAFI et al. (2017). By widening the range of morphological variations, the increasing number of species described in ljuhya makes its delimitation from the morphologically similar genera Lasionectria (Sacc.) Cooke and Lasionectriella Lechat & J. Fourn. occasionally challenging, if not misleading. Two species formerly assigned to ljuhya based on morphological traits, I. antillana Lechat & Courtec. and I. oenanthicola Lechat & Hairaud, were shown to be phylogenetically distant from ljuhya and were recombined in Lasionectria by ASHRAFI et al. (2017). On the same grounds, I. dentifera (Samuels) Rossman & Samuels, though featuring ascomata with a flattened discoid apex lined by a crown of fasciculate hairs typical of ljuhya (SAMUELS, 1976), was likewise shown by these authors to be phylogenetically distant from Ijuhya, with closer affinities with both Ochronectria Rossman & Samuels and Protocreopsis Doi, but without formal taxonomic recombination. The latest described species is I. vitellina Schroers, Ashrafi & W. Maier, only known as an asexual morph isolated from cyst nematodes eggs in Turkey (ASHRAFI et al., 2017).

We introduce and document in this paper three new species of *ljuhya* from France, French Guiana and Spain, based on morphological, cultural and molecular data and we provide an updated dichotomous key taking into account the new species for which a sexual state is known. The removal of *I. antillana* and *I. oenanthicola* and the addition of our three new species bring the number of currently known species of *ljuhya* to twenty-two.

Lasionectria boothii (D. Hawksw.) Lechat & J. Fourn. is proposed as synonym of *L. oenanthicola* (Lechat & Hairaud) Schroers, Ashrafi & W. Maier and *Nectriopsis lecanodes* (Ces.) Diederich & Schroers is reinstated as *L. lecanodes* (Ces.) Petch based on molecular data. Finally we discuss the validity of the morphological features on which *ljuhya* can be distinguished from *Lasionectria* and *Lasionectriella*.

# **Materials and methods**

Dry specimens were rehydrated and examined using the method described by Rossman et al. (1999). Microscopical observations and measurements were made in water. The holotype specimen and paratypes were deposited in LIP herbarium (University of Lille) and living cultures in the CBS (Collection of the Westerdijk Fungal Biodiversity Institute, Utrecht, The Netherlands), and at CIRM-CF (Centre International des Ressources Microbiennes, Marseille, France). Cultures of the living specimens were made on PDA (Potato Dextrose Agar) with 5 mg/l of streptomycin in Petri dishes 9 cm diam incubated at 25 °C. DNA extraction, amplification, and sequencing were performed by ALVALAB (Santander, Spain): Total DNA was extracted from dry specimens blending a portion of them using a micropestle in 600 µL CTAB buffer (CTAB 2%, NaCl 1.4 M, EDTA pH 8.0 20 mM, Tris-HCl pH 8.0 100 mM). The resulting mixture was incubated for 15 min at 65 °C. A similar volume of chloroform:isoamylalcohol (24:1) was added and carefully mixed with the samples until their emulsion. It was then centrifuged for 10 min at 13,000 g, and the DNA in the supernatant was precipitated with a volume of isopropanol. After a new centrifugation of 15 min at the same speed, the pellet was washed in 70% cold ethanol, centrifuged again for 2 min and dried. It was finally resuspended in 200 µL ddH<sub>2</sub>O. PCR amplification was performed with the primers LROR and LR5 (VILGALYS & HESTER, 1990) to amplify the 28S nLSU region. PCR reactions were performed under a program consisting of a hot start at 95 °C for 5 min, followed by 35 cycles at 94 °C, 54 °C and 72 °C (45, 30 and 45 s respectively) and a final 72 °C step 10 min. Chromatograms were



<sup>0.08</sup> 

**Fig. 1** – Maximum likelihood phylogeny (-lnL = 2892.31521) of the three new *ljuhya* species, inferred by PhyML 3.0, model HKY85 from a 892 bp matrix of 28S rRNA sequence, rooted with *Clonostachys rosea*.

checked searching for putative reading errors, and these were corrected.

Analyses were performed online at www.phylogeny.lirmm.fr (DEREEPER *et al.*, 2008). Maximum likelihood phylogenetic analyses were performed with PhyML 3.0 aLRT (ZWICKL, 2006), using the GTR + I +  $\Gamma$  model of evolution. Branch support was assessed using the non-parametric version of the approximate likelihood-ratio test, implemented in PhyML SH-aLRT (ANISIMOVA & GASCUEL, 2006). Nomenclature follows MycoBank (Westerdijk Fungal Biodiversity Institute, Utrecht, The Netherlands).

### Taxonomy

<b>Ijuhya salviicola</b> Lechat & J. Fourn., <i>sp. nov</i> .	Fig. 2
MycoBank MB 830218	

Diagnosis: Differs from other *ljuhya* species having a crown of long fasciculate hairs around upper margin by long-fusiform, 1-septate, striate ascospores (35-)45- $60(-62) \times 4.5$ - $5.5 \mu m$ .

Holotype: FRANCE, Bretagne, Côtes-d'Armor, Queffioec, on twig of *Salvia officinalis* L. (*Lamiaceae*), 1 May 2018, *leg*. Brigitte Capoen, BC010518 (LIP BC010518); Genbank LSU sequence: MH665603.

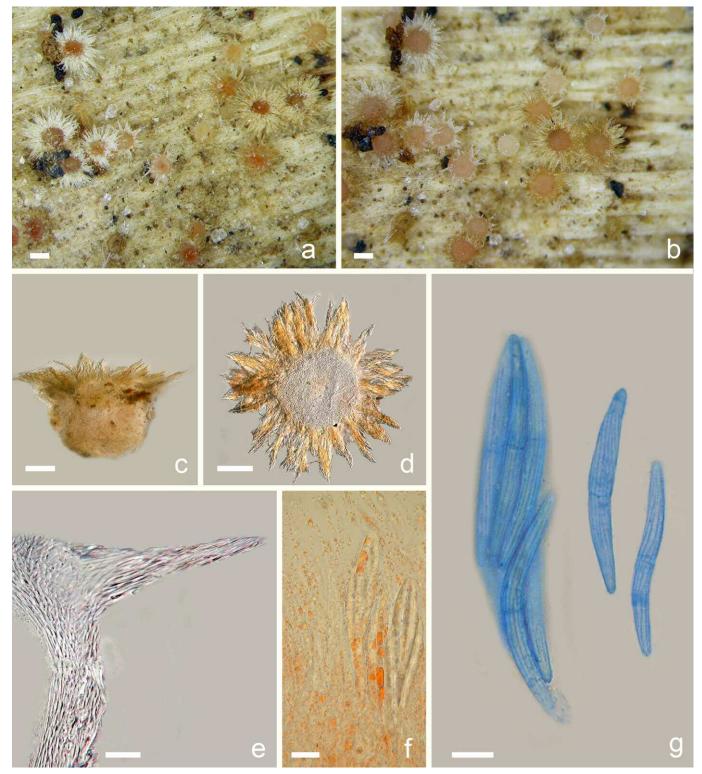
Etymology: The specific epithet "salviicola" refers to the host Salvia officinalis.

**Ascomata** superficial, solitary or in groups of 2–4, difficult to remove from substrate, non-stromatic, subglobose, (250–)270–300 (-320)  $\mu$ m high × (220–)240–260(–270)  $\mu$ m diam (Me = 280 × 250  $\mu$ m, n = 20), pale yellow to orange, not collapsing when dry, not changing colour in 3% KOH or lactic acid. **Ascomatal apex** flat, discoid, 200–230  $\mu$ m diam, slightly collapsing cupulate when dry,

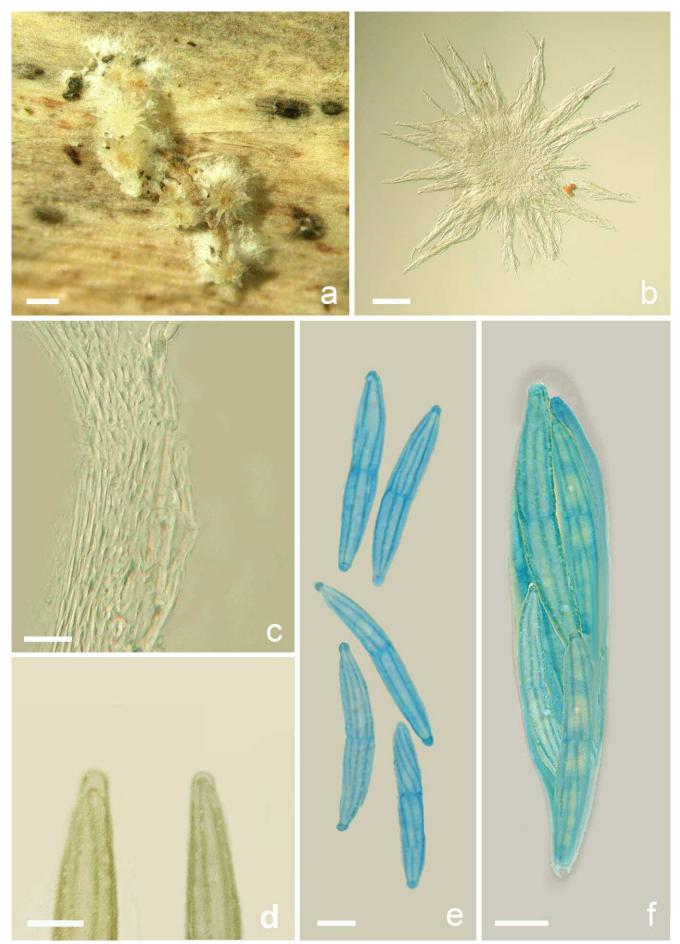
without visible papilla. **Ascomatal surface** obscured by smooth, hyaline to pale yellow, thick-walled hyphal elements 2–2.5  $\mu$ m wide, arising from base of ascomata, proliferating and agglutinating to form triangular teeth 50–180  $\mu$ m long, 20–35  $\mu$ m wide at base, slightly flexuous, with hyphal elements rounded at free ends, arranged in a stellate fringe around upper margin of ascomata. In vertical section, ascomatal wall 28–35  $\mu$ m thick, excluding loose outermost hyphal elements, of a single region composed of thick-walled, globose to ellipsoidal cells 3–11 × 2.5–5  $\mu$ m, with pale yellow wall 1.5–2.5  $\mu$ m thick, becoming hyaline, thin-walled and elongated toward interior. **Asci** evanescent, clavate to fusiform, wider in mid-

part, attenuated and rounded at apex, without ring, containing orange oily droplets and eight multiseriate to fasciculate ascospores,  $(70-)80-90(-95) \times 10-14 \ \mu m$  (Me =  $85 \times 12 \ \mu m$ , n = 20). **Interthecial elements** not seen. **Ascospores** (35-)45-60(-62)  $\times$  4.5-5.5  $\ \mu m$  (Me =  $55 \times 5 \ \mu m$ , n = 30), long fusiform, straight to slightly curved, occasionally twisted, equally 1-septate, not constricted at septum, hyaline, longitudinally striate, completely filling each ascus.

**Cultural characteristics:** Colony after three weeks 15–18 mm diam, pale yellow in center, with some rosy tinges in middle area, white at margin producing an acremonium-like asexual morph; reverse pale yellow to pale orange. Aerial mycelium composed of



**Fig. 2** – a–g: *ljuhya salviicola* (Holotype BC010518). a, b: Ascomata in natural environment. c: Close-up of ascoma in side view in water. d: Ascomatal apex in top view showing fasciculate hairs at periphery of upper disc. e: Vertical section through ascomatal wall. f: Asci and ascospores in water. g: Ascus and ascospores in lactic acid/cotton blue. Scale bars: a-b =200  $\mu$ m; c, d =100  $\mu$ m; e = 20  $\mu$ m; f, g = 10  $\mu$ m.



**Fig. 3** – a–f: *ljuhya saulensis* (Holotype CLLG18040). a: Ascomata in natural environment. b: Ascomatal apex in top view showing the fasciculate hairs at periphery of the upper disc. c: Vertical section through the lateral ascomatal wall. d: Close-up of tips of ascospores in water; e: Ascospores in lactic acid/cotton blue showing striate ornamentation and cyanophilous tips. f: Ascus and ascospores in lactic acid/cotton blue. Scale bars: a = 200  $\mu$ m; b = 100  $\mu$ m; c, e, f = 10  $\mu$ m; d: = 5  $\mu$ m.

smooth, septate, hyaline hyphae 2–2.8 µm wide bearing flexuous, hyaline conidiophores 30–55 µm long, 2–2.8 µm wide at base, subulate with a flared collarette. Conidia ellipsoidal to sub-cylindrical, attenuated at base with an abscission scar,  $5.5-7 \times 1.5-2$  µm, smooth, hyaline. Culture of this specimen no longer viable.

*ljuhya saulensis* Lechat & J. Fourn., *sp. nov.* Fig. 3 MycoBank: MB 830219

Diagnosis: Differs from other *ljuhya* species having long fasciculate hairs around upper margin by long-fusiform, 1-septate, striate ascospores  $(30-)35-45(-48) \times 5-6.5 \mu m$  with cyanophilous tips.

Holotype: FRENCH GUIANA, Saül, Gros Arbres trail, on dead leaf of *Phenakospermum guyanense* (*Strelitziaceae*), 25 Aug. 2018, *leg*. Christian Lechat, CLLG18040 (LIP); Genbank LSU sequence MK629312.

Etymology: The specific epithet "*saulensis*" refers to Saül, the locality where this species was collected.

Ascomata superficial, solitary, scattered on host surface, with a thin basal white subiculum, non-stromatic, subglobose, (200–)230– 270(–290)  $\mu$ m high  $\times$  200–240  $\mu$ m diam, white to pale yellow, laterally pinched to slightly cupulate when dry, not changing colour in 3% KOH or lactic acid. Ascomatal apex flat, discoid, 170–200 μm diam. Ascomatal surface completely obscured by smooth, hyaline hyphal elements arising from base of ascomata and subiculum, proliferating and agglutinating to form triangular teeth 170–250 µm long, up to 50 µm wide at base, composed of fasciculate hairs 2-2.5 µm wide, hyaline, flexuous, rounded at tip, arranged in a stellate crown around upper margin of ascomata. In vertical section, ascomatal wall 18–25  $\mu$ m thick, of a single region composed of thickwalled, globose to ellipsoidal cells  $3-8 \times 2-2.5 \mu m$ , with pale yellow wall 1–1.5 µm thick, becoming hyaline and elongated toward interior. Asci clavate to widely fusiform, evanescent, inconspicuous,  $(65-)75-90(-95) \times 12-18(-20) \mu m$  (Me =  $80 \times 15 \mu m$ , n = 20), apices rounded, without ring, with eight multiseriate to fasciculate ascospores. **Ascospores**  $(30-)35-45(-48) \times 5-6.5 \mu m$  (Me =  $40 \times 6 \mu m$ , n = 30), long fusiform, slightly curved, equally 1-septate, slightly constricted at septum, hyaline, coarsely striate with 2-3 finely verrucose striae per half spore, striae longitudinally oriented to slightly spiralling; ends rounded, strongly cyanophilous, 1.5–2.5 µm diam.

**Cultural characteristics:** Colony after three weeks 25–35 mm diam, entirely white; aerial mycelium composed of smooth, septate, hyaline, hyphae 2–2.5  $\mu$ m diam; not sporulating after four weeks. Culture of this specimen no longer viable.

*Ijuhya somiedoensis* Lechat & J. Fourn., *sp. nov.* Fig. 4 MycoBank MB 830220

Diagnosis: Differs from the most similar *ljuhya corynospora* in having smaller 1-septate ascospores  $18-22 \times 2.5-3 \mu m$  vs  $27-42 \times 3-4.5 \mu m$  and 5-9(-13)-septate.

Holotype: SPAIN, Asturias, Natural Park of Somiedo, La Peral, on dead stem of *Juncus* sp. (*Juncaceae*), 7 Jun. 2017, *leg*. Marja Pennanen, CLL17015 (LIP); ex-holotype culture CBS142016, Genbank LSU sequence: MF628020.

Etymology: The specific epithet "somiedoensis" refers to the natural park of Somiedo where the fungus was collected twice.

**Ascomata** superficial, solitary or in groups of 4–30, easy to remove from substrate, non-stromatic, globose to subglobose,  $(170-)180-210(-220) \mu m$  diam (Me = 200  $\mu m$ , n = 10), pale yellow to orange, not collapsing when dry, not changing colour in 3% KOH or lactic acid. **Ascomatal apex** flat, discoid, 170–190  $\mu m$  diam, with a minute, acute papilla. **Ascomatal surface** completely obscured by smooth, hyaline hyphal elements 1.5  $\mu m$  wide, hyaline, slightly flexuous, rounded at tip, arising from base of ascomata, whose free

ends form a crown around margin of disc. In vertical section, ascomatal wall 25–30 µm thick, excluding hyphal elements, of a single region composed of thick-walled, globose to ellipsoidal cells 4–10 × 2–3 µm, with pale yellow wall 1–1.5 µm thick, becoming hyaline, thin-walled and elongated toward interior. **Asci** clavate, evanescent, (50–)60–70(–80) × 8.5–12 µm (Me = 67 × 10 µm, n = 30), apices rounded, without ring, with eight irregularly biseriate to multiseriate ascospores. No **interthecial elements** observed. **Ascospores** (16–) 18–22(–25) × 2.5–3 µm (Me = 21 × 2.8 µm, n = 30), fusiform, with narrowly rounded to subacute ends, equally 1-septate, not constricted at septum, with upper cell slightly swollen and basal cell slightly curved, hyaline, smooth, completely filling each ascus.

**Cultural characteristics:** Colony after three weeks 10–15 mm diam, orange in center, white at margin; aerial mycelium composed of smooth, septate, hyaline, hyphae 2.5–3  $\mu$ m wide; not sporulating after five weeks.

Additional specimen: SPAIN, Asturias, Natural Park of Somiedo, La Malva, on dead twigs of *Genista* sp. (*Fabaceae*), 7 Jun. 2013, *leg*. Christian Lechat, CLL13022 (LIP); culture CBS 136677; GenBank sequence LSU: MH660388.

#### Discussion

As mentioned in the introduction, two species formerly assigned to *ljuhya* based on morphology proved to belong to *Lasionectria* when submitted to molecular phylogenetic analysis (ASHRAFI *et al.*, 2017). Furthermore, these authors showed that *l. dentifera*, though morphologically typical for *ljuhya*, clusters outside the *ljuhya* clade, with unexpected affinities with *Ochronectria* and *Protocreopsis*.

On the other hand, in absence of morphological data on its sexual state, *I. vitellina* was assigned to *ljuhya* based only on the similarity of its LSU sequence with that of *I. paraparilis* (Samuels) Rossman & Samuels, despite its ITS sequence showed closest affinities with *Stromatonectria* Jaklitsch & Voglmayr, a genus morphologically different from *ljuhya* (JAKLITSCH & VOGLMAYR, 2011). Moreover, its asexual state on natural substrate and in culture consists of chlamydospore-like structures that develop into microsclerotia, unlike the acremonium-like asexual state of *ljuhya* spp. if known.

These apparent discrepancies suggest that a polythetic approach based on both morphology of the holomorph and phylogenetic data is necessary to assign a collection to a taxon with reliability. For this reason, we review the morphological differences delineating *ljuhya* from its closest relatives *Lasionectria* and *Lasonectriella* already explained by LECHAT & FOURNIER (2017a), with the addition of illustrations and a re-definition of the few discriminating characters.

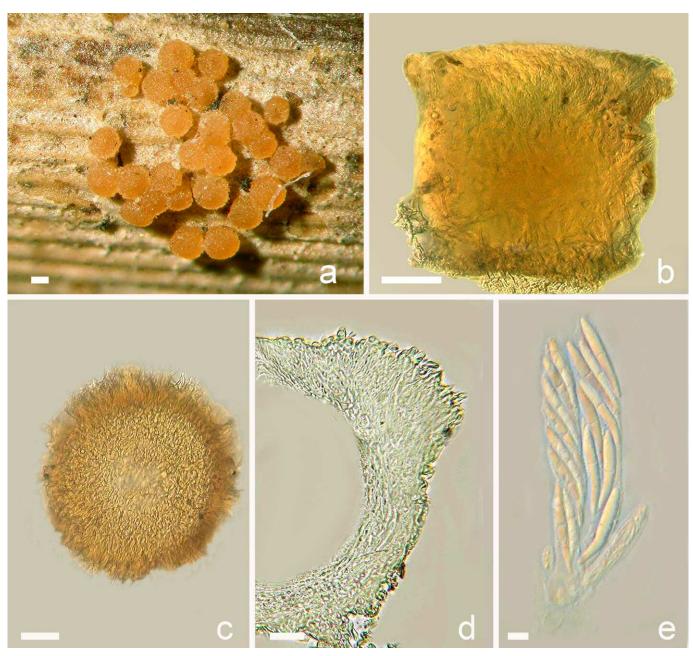
*ljuhya, Lasionectria* and *Lasionectriella* belong to the *Bionectriaceae* as defined by RossMAN *et al.* (1999). They have in common the usually light-coloured, hairy ascomata not changing colour in 3% KOH or lactic acid and occurrence on monocotyledonous and dicotyledonous woody substrates, dead leaves or herbaceous debris. Morphological characteristics of ascomatal apex, ascomatal wall structure and arrangement of superficial hairs are needed to separate these genera.

Typically, *ljuhya* is characterized by ascomata with a flat discoid apex pierced by a small, faintly prominent papilla and a one-layered ascomatal wall covered by hyphal elements arising from base, proliferating over ascomatal surface and becoming fasciculate to form teeth arranged in a stellate fringe around upper margin (Fig. 5a). Some species likewise feature hairs arising from the ascomatal base but instead of differentiating into fasciculate teeth, they are agglutinated and intertwined to form a thick apical margin, as in *l. hubeiensis* Y. Nong & W.Y. Zhuang, *l. pachydisca* Lechat & J. Fourn. (Fig. 5b), and *l. tetraspora* Lechat & J. Fourn. A slightly deviating morphology may be encountered in species such as *l. somiedoensis* (Fig. 5c) in which the disc margin is replaced by a thick crown of shorter, agglutinated hairs with free ends. *ljuhya lilliputiana* Lechat & J. Fourn. differs from all known species in having cylindrical protuberances extending upwards around the upper margin of the ascomata, resulting from proliferation of cells of the ascomatal wall (Fig. 5d); this unique configuration of the ascomatal apex makes this species distinctive.

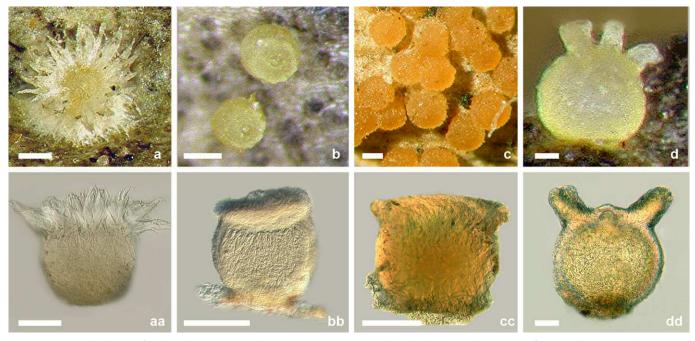
As noted by LECHAT & FOURNIER (2017a), species included in the phylogenetic analysis of the genus *ljuhya* appeared roughly distributed into two well-supported subclades, one with species having fasciculate hairs (subclade 1), the other with those lacking fasciculate hairs (subclade 2). This hypothesis is supported by the inclusion of the three new species, two of them with fasciculate hairs, *l. salviicola* and *l. saulensis*, clustering with species in subclade 1, while *l. somiedoensis* shows affinities with subclade 2 (Fig. 1). It is interesting to note that in this subclade, *l. lilliputiana* and *l. somiedoensis*, in correlation with their deviating morphology of ascomatal apex, appear distantly related to *l. pachydisca* and *l. tetraspora*, the two species with sharply delimited apical disc. However, this segregation of two morphological types in two subclades is contradicted by the placement of *l. paraparilis*, a species with a glabrous disc, in the subclade with species having fasciculate hairs. Congruent results were obtained by ASHRAFI *et al.* (2017) with a LSU sequence from a specimen of *I. paraparilis* from China and in the present study with a LSU sequence from a specimen from Japan (Fig. 1).

The most similar genera that could be confused with *ljuhya* are *Lasionectria* and *Lasionectriella*. *ljuhya* and *Lasionectriella* have ascomata covered by hyphal elements arising from base, but the latter differs in having a convex to conical apex, ascomatal wall of two regions, and thick-walled hairs merging with cells of ascomatal wall whose free ends form a short fringe or a crown around the upper margin of the perithecium (Fig. 6).

Lasionectria differs from *ljuhya* in having an ascomatal wall composed of two regions as does *Lasionectriella* and a convex ascomatal apex. The ascomatal surface in *Lasionectria* is covered by variously arranged flexuous hairs, but differs in having hairs consistently arising from cells of the ascomatal wall (Fig. 7), unlike *ljuhya* and *Lasionectriella* whose hairs arise from base and proliferate over the ascomatal wall. The fasciculate hairs may form a stellate apical crown, as in *L. antillana*, in a way which strongly recalls typical *ljuhya* (Fig 7d). Critical microscopical observation shows that, unlike typical



**Fig. 4** – a–e: *Ijuhya somiedoensis* (Holotype CLL17015). a: Ascomata in natural environment. b: Close-up of ascoma in side view in water. c: Ascomatal apex in top view; d: Vertical section through ascomatal wall. e: Asci and ascospores in water. Scale bars: a =100  $\mu$ m; b = 50  $\mu$ m; c: = 40; d = 20  $\mu$ m; e = 5  $\mu$ m.



**Fig. 5** – a-c: Morphological features characterizing the genus *ljuhya* in natural environment in top view. a: *l. fournieri* showing stellate crown of fasciculate hairs around upper margin; b: *l. pachydisca* with a flat, thick apical disk; c: *l. somiedoensis* and its crown of hyphal elements; d: *l. lilliputiana* in side view, showing protuberances extending upwards around upper margin. aa–dd: Same species as above in side view in water. All scale bars = 100  $\mu$ m except d, dd = 25  $\mu$ m.

*ljuhya*, the fasciculate hairs of *L. antillana* arise from the ascomatal wall cells, which is diagnostic (Fig 7e). In this context, the morphology of ascomatal wall and its vestiture as reported by FRÖHLICH & HYDE (2000) might prove *I. hongkongensis* J. Fröhl. & K.D. Hyde to be excluded from *ljuhya*.

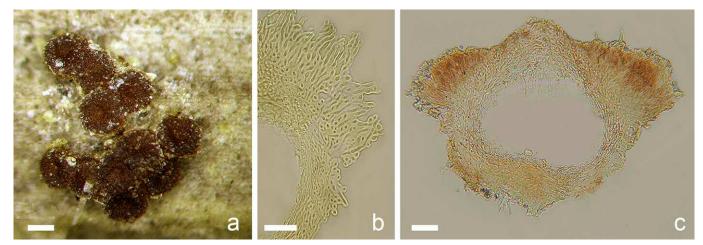
The recently introduced genus *Lasionectriopsis* Lechat & P.-A. Moreau should likewise be considered in this discussion as it morphologically resembles some species of *Lasionectria*. It differs in having ascomata embedded in a dense subiculum spreading over the ascomatal wall and phylogenetic analysis shows it is only distantly related to the genera discussed above (LECHAT *et al.*, 2019).

The three new species we propose herein are shown by phylogenetic analysis to belong to *ljuhya* (Fig. 1) and conform well to the morphological definition of *ljuhya* given above.

Morphologically, the closest species to *I. salviicola* is *I. peristomialis*, the type species, which likewise has long-fusiform, striate ascospores in the same size range, but differs in having longer fascicles of hairs up to 300  $\mu$ m long and ascospores (1–)5–7(–11)-septate. In our phylogenetic analysis, *I. salviicola* is distant from *I. peristomialis* 

and is nested on a sister branch to *I. faveliana* (Fig. 1). The latter, known from French Guiana on palm leaves, differs from *I. salviicola* in having shorter ascospores  $15-17 \times 3-3.5 \mu m$  (Lechat & FOURNIER, 2017a) and only 96% similarity of their LSU sequences.

In the key proposed below, *l. saulensis* keys out next to *l. salviicola*, with which it shares ascomata with teeth-like fasciculate hairs around the upper margin and long-fusiform, 1-septate, striate ascospores in a similar size range. It differs from *l. salviicola* by its tropical occurrence, whitish vs orange ascomata and slightly smaller, barely overlapping ascospore dimensions  $35-45 \times 5-6.5 \,\mu$ m vs  $45-58 \times 4.5-5.5 \,\mu$ m. The most distinctive feature of *l. saulensis* is that the ascospores have cyanophilous rounded tips, which sets it apart from all known species in the genus. We failed to elucidate unambiguously the nature of these structures but we assume that they result from a local loosening or thickening of the striate perispore (Fig. 3-d) more than from the presence of cellular or non-cellular appendages. In our phylogram (Fig. 1), *l. saulensis* is nested on a sister branch to *l. fournieri*, a palm-inhabiting species known from French Guiana, which resembles *l. saulensis* in ascomatal and ascospore



**Fig. 6** – a-b: *Lasionectriella herbicola*; a: Ascomata in natural environment; b: Vertical section through ascomatal wall showing hairs merging with cells of ascomatal wall; c: Vertical section through ascomata showing conical apex. Scale bars:  $a = 100 \mu m$ ; b, c: = 20  $\mu m$ .

morphology but differs in having significantly smaller ascospores 22–23.5(–25)  $\times$  4.5–5  $\mu m$  that lack cyanophilous tips. Both species display 98% similarity of LSU sequences, which supports their phylogenetic affinities but also their distinctiveness.

*ljuhya somiedoensis* is characterized by pale orange subglobose ascomata with a discoid apex lined by a prominent crown of agglutinated hyphae, and fusiform, hyaline, smooth, 1-septate ascospores  $18-22 \times 2.5-3 \mu m$  with slightly swollen upper cell and subacute ends. This combination of characters sets it apart from all known species of *ljuhya*. The most similar species is *l. corynospora* (Samuels) Rossman & Samuels, known from subtropical New Zealand (Auckland region), which likewise features a wide apical disc with prominent margins and hyaline and smooth ascospores. *ljuhya corynospora* differs from our new species by having larger clavate ascospores  $27-42 \times 3-4.5 \mu m$  that are 5-9(-13)-septate (SAMUELS, 1978; ROSSMAN *et al.*, 1999). It is interesting to note that ascospores of *l. somiedoensis* are swollen in the upper part like those of *l. corynospora* but they are subacute at both ends while those of *l. corynospora* are apically obtuse and thus typically clavate.

Recently, based on phylogenetic analyses of ITS and LSU sequences, ASHRAFI et al. (2017) showed that *l. oenanthicola* belongs to *Lasionectria* and introduced the new combination *L. oenanthicola* (Lechat & Hairaud) Schroers, Ashrafi & W. Maier. This is in agreement with our definition of this genus, but the authors were apparently unaware that in the meantime *l. oenanthicola* had been recognized as *l. boothii* (D. Hawksw.) Lechat & J. Fourn. (2017b). Accordingly, the new combination *Lasionectria* boothii (D. Hawksw.) Lechat & J. Fourn. comb. nov. is proposed to accommodate the basionym *Nectria boothii* Hawksworth (1980). Finally, based on morphological characteristics and phylogenetic analysis of its LSU sequence (Fig. 1), *Lasionectria* lecanodes is re-instated to accommodate the basionym *Sphaeria* lecanodes.

## **Taxonomic novelties**

ljuhya salviicola Lechat & J. Fourn., sp. nov. – MycoBank: MB 830218 ljuhya saulensis Lechat & J. Fourn., sp. nov. – MycoBank: MB 830219 ljuhya somiedoensis Lechat & J. Fourn., sp. nov. – MycoBank: MB 830220

*Lasionectria boothii* (D. Hawksw.) Lechat & J. Fourn., *comb. nov.* – MycoBank: MB 830224.

Basionyme: Nectria boothii D. Hawksw., Transactions of the British Mycological Society, 74 (3): 572 (1980).

≡ Ijuhya boothii (D. Hawksw.) Lechat & J. Fourn. (2017).

= ljuhya oenanthicola Lechat & Hairaud (2012).

= *Lasionectria oenanthicola* (Lechat & Hairaud) Schroers, Ashrafi & W. Maier (2017).

*Lasionectria lecanodes* (Ces.) Petch, *Transactions of the British Mycological Society*, 21 (3-4): 267 (1938) [re-instated].

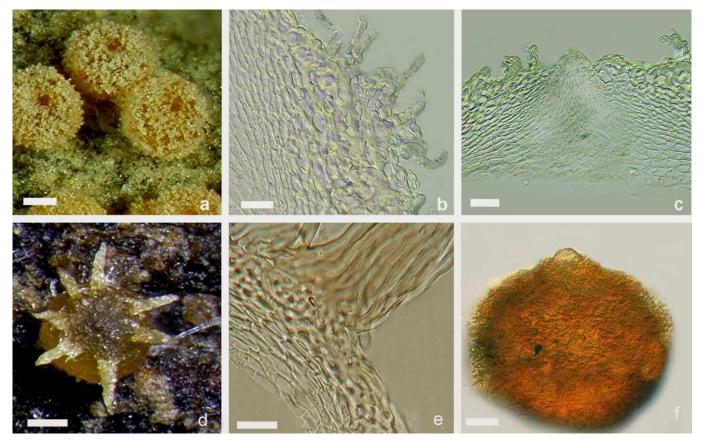
Basionym: Sphaeria lecanodes, in Rabenhorst, Klotzschii Herb. Viv. Mycol., Edn Nov, Ser. Sec., Cent. 6: no. 525 (1857) – MycoBank: MB 269144.

- = Nectria lecanodes Ces. (1857).
- ≡ Dialonectria lecanodes (Ces.) Cooke (1884).
- ≡ Cucurbitaria lecanodes (Ces.) Kuntze (1898).

≡ *Nectriopsis lecanodes* (Ces.) Diederich & Schroers (1999).

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**Fig. 7** – a-f: Morphological features characterizing *Lasionectria*; a-c: *L. lecanodes*; a: Ascomata in natural environment; b: Hairs arising from cells of ascomatal wall; c: Convex apex of perithecium in vertical section; d, e: Fasciculate hairs of *L. antillana* arising from cells of ascomatal wall: f: *L. mantuana* (type species) and its convex apex. Scale bars: a, d = 100; b,  $e = 10 \mu m$ ;  $c = 20 \mu m$ ;  $f = 50 \mu m$ .

# Key to the genera allied to *ljuhya*

1.	Ascomata with flat discoid apex; ascomatal wall of a single region; hairs arising from base, proliferating over ascomatal surface, becoming fasciculate to form teeth, arranged in a stellate fringe around upper margin or with hairs agglutinating to form a thick, apical disc, an obtuse crown or more rarely with cylindrical protuberances originating from ascomatal wall cells (Fig. 5)
2.	Hairs arising from cells of ascomatal wall, evenly covering ascomatal surface, solitary or fasciculate; ascospores smooth or striate (Fig. 7)

# Updated key to species of *ljuhya* based on morphological characteristics of ascomata and ascospores

	Ascomata with long, fasciculate hairs forming triangular teeth, arranged in a stellate fringe around upper margin	
	Ascomata with shorter, non-fasciculate hairs, whose free ends form a crown around upper margin	
С.	Ascomata with hairs agglutinating to form a thick, exceeding apical disk with rounded margin or with cylindrical protuberances	
	protruding upwards around upper margin1	3
	Ascospores finely spinulose, $6-8(-9) \times 3-4 \mu m$ ; on bark and decorticated wood (New Zealand)	
	Ascospores smooth or striate	
	Ascospores smooth	
	Ascospores striate	
	Ascospores 1-septate	
3.	Ascospores dictyosporous, $48-97 \times 10-16 \mu$ m, with 7–13 transverse septate and one longitudinal septum; on herbaceous stems	
	(Colombia and Venezuela)	
	Ascospores cylindrical, 12.5–17.6 × 2–2.5 μm; on Calamus spp. (Hong Kong)	
	Ascospores oblong to ellipsoidal 7.2–9 × 2–2.7 μm; on dead leaves of <i>llex</i> (USA)	
	Ascospores multiseptate	
	Ascospores 1-septate	7
	Ascomata hyaline to pale yellow in fresh state; ascospores (1–)5–7(–11)-septate, (24–)30–60(–110) × 4–7(–8) $\mu$ m; on wood and herbaceous debris (widespread)	is
6.	Ascomata bright orange in fresh state; ascospores (1–2–)3–5(–6)-septate, (30–)35–45(–50) $\times$ 5–7 µm; on twigs of <i>Convolvulus</i>	
	canariensis (Spain, Canary Islands) I. aurantiac	
	Ascospores over 40 μm long	
7.	Ascospores less than 30 µm long	9
8.	Ascospores with bipolar cyanophilous tips, (30–)35–45(–48) $\times$ 5–6.5 µm; on dead leaf of <i>Phenakospermum guyanense</i> (French	
_	Guiana)I. saulens	
	Ascospores lacking cyanophilous tips, $(35-)45-58(-62) \times 4.5-5.5 \mu m$ ; on twig of <i>Salvia officinalis</i> (France) <i>I. salviicol</i>	a
9.	Fasciculate hairs up to 250 $\mu$ m long; ascospores (18–)22–23.5(–25) × 4.5–5(–5.5) $\mu$ m; on dead leaves of <i>Astrocaryum</i> sp.	
_	(French Guiana)	
9.	Fasciculate hairs less than 200 μm long	0
10.	Fasciculate hairs 28–80 $\mu$ m long; ascospores (8.5–)9.5–11.5(–12.5) × 2.8–3.2(–3.5) $\mu$ m, finely striate; on <i>Equisetum hyemale</i>	
	(Germany)	
	Fasciculate hairs over 80 μm long; ascospores longer and coarsely striate; on different hosts	
	Fasciculate hairs up to 100 $\mu$ m long; ascospores (19–)21–28 × 3.5–4.5 $\mu$ m; on <i>Lobelia lupa</i> and <i>Nolina micrantha</i> (Chile). <i>I. chilens</i>	IS
11.	Fasciculate hairs up to 190 $\mu$ m long; ascospores (14–)15–17(–18) × 3–3.5(–4) $\mu$ m; on dead leaves of <i>Astrocaryum</i> sp.	
	(French Guiana)	a
12.	Ascospores ellipsoidal to oblong (7.5–)8.5–10(–11) × 2–2.7(–3) $\mu$ m; on old perithecia of <i>Thelonectria jungneri</i> (Jamaica)	_
12	I. leucocarp	
	Ascospores fusiform, (16–)18–22(–25) $\times$ 2.5–3 $\mu$ m; on <i>Juncus</i> and <i>Genista</i> (Spain) <b>1</b> somiedoens. Ascospores smooth or striate	
	Ascospores spinulose <b>1</b> Ascospores clavate, smooth, 27–42 × 3–4.5 µm, 5–9(–13)-septate; on woody and herbaceous debris (New Zealand)	0
14.	Ascospores clavate, smooth, 27–42 x 5–4.5 μm, 5–9(–15)-septate; on woody and herbaceous debris (new Zealand)	
1.4	Ascospores fusiform, striate, smaller	
	Ascompties rushoffin, strate, smaller Ascomatal apex with a flat glabrous disc; ascospores (19.5–)21.5–24.5(–25.5) × 4–5 µm; on black mycelium on bamboo culm	5
15.	(French Guiana)	ic
15	Ascomatal apex with cylindrical protuberances protruding upwards around upper margin; ascospores $(8.5-)9-10(-11) \times 2.5-$	12
13.	Ascomatal apex with cylindrical protuberances protruding upwards around upper margin; ascospores (8.5–)9–10(–11) × 2.5– 2.8 μm; on dead leaves of <i>Astrocaryum</i> sp. (French Guiana) <b>I. lilliputian</b>	
16	2.8 μm; on dead leaves of Astrocaryum sp. (French Gulana)	
	Asci 2–4-spored	
	Ascospores $(11-)12-14(-15) \times 2.8-3.2(-3.5) \mu$ m; on dead leaves of <i>Astrocaryum</i> sp. (French Guiana) <b>I. pachydisc</b>	
	Ascospores $(11-)12-14(-15) \times 2.6-5.2(-5.5)$ µm; on dead leaves of Astrocaryan sp. (Fightri Gularia)	
	Ascospores $14.5-20 \times (2.5-)5-5(-5.4)$ µm; on nerbaceous debns (riench Guiana, 0.5.A.)	
	Asci 4-spored, ascospores (14–)15–18(–19) × 3.5–4(–4.5) µm; on woody debris or dead stromata of ascomycota	
10.	$h$ solution as cospores (14-)15-10(-19) $\times$ 5.5-4(-4.5) µm, on woody depits of dedu stromated of as completed <b>I. tetraspor</b>	u

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