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Sporormiella octomegaspora, a new hairy species with eight–celled ascospores from Spain

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Abstract — An ascolocular ascomycete with semi–immersed, hairy and pyriform pseudothecia, abundant pseudoparaphyses, fissitunicate 8-spored asci, and dark, very large 8-celled ascospores has been isolated from deer dung in Andalusia (Spain). Based on morphological features, a new species is erected and accommodated in *Sporormiella*, which the authors regard as a genus independent of *Preussia*. The new species is discussed and placed in a key, and a previous worldwide key to *Sporormiella* species with 8-celled spores is updated.

Key words — coprophily, phylogeny, *Pleosporales*, relationships, *Sporormiaceae*

Introduction

Sporormiella Ellis & Everh. (*Sporormiaceae* Munk, *Pleosporales*) is characterised by ascoloculate, aperiphysate, ostiolate pseudothecia, fissitunicate, elongated, 8-spored asci with a scarcely developed apical apparatus, dark colored, transversely septate, 4- to poly-celled ascospores with germ-slits and usually with a gelatinous envelopment, and preferable growth on dung (Ellis & Everhart 1892, Ahmed & Cain 1972, Barr 2000).

Preussia Fuckel, in the same family, has morphological features so similar to *Sporormiella* that its independence has been questioned. We refer to previous works on this subject (Doveri 2004, 2005, 2007, 2011; Doveri & Coué 2008) to explain why the senior author regards *Sporormiella* as distinct from *Preussia* (Cain 1961; Ahmed & Cain 1972; Barrasa & Checa 1991; Lumbsch & Huhndorf 2007, 2010; Kirk et al. 2008). That view has been strengthened by an extensive phylogenetic study on *Sporormiaceae* (Kruys & Wedin 2009), which does not definitely resolve the question of synonymy.

Another view (von Arx 1973, Guarro et al. 1997a, Chang & Wang 2009) that regards *Sporormiella* as a later synonym of *Preussia* has led to the recombination of several *Sporormiella* species in *Preussia* (von Arx 1973; Valldosera & Guarro

1990; Guarro et al. 1997a,b; Abdullah et al. 1999; Arenal et al. 2004, 2005, 2007; Chang & Wang 2009, Kruys & Wedin 2009) or even the description of new *Sporormiella* species in *Preussia* sensu lato (Arenal et al. 2005, 2007; Chang & Wang 2009, Asgari & Zare 2010).

Sporormiella encompasses, at the present, more than 80 species, mostly with glabrous pseudothecia and 4-celled ascospores, a minority (12) with hairs, and/or more than 4-celled ascospores, very few with ascospores consisting of a variable number of cells (2–10).

A recent collection from Andalusia (Spain) of a hairy species with consistently 8-celled ascospores has been submitted to our observation. Its morphological features are unique, thus allowing us to recognise it as a new *Sporormiella* species.

The aim of our study is to define the new species and establish its relationships with other hairy *Sporormiella* spp. and a group of *Sporormiella* with 8-celled ascospores.

Materials & methods

Dried and fresh specimens of *Sporormiella octomegaspora* were sent us from Spain, the fresh material still on small pieces of red-deer (*Cervus elaphus*) dung. Fresh material (five specimens) was utilised for macro- and microscopic studies and for attempting, unsuccessfully, isolation in axenic culture. The specimens were picked from the dung with a sterile needle, washed in distilled water, and placed in a drop of water on a microscope slide. Microscopic examinations were carried out on specimens mounted in water, Melzer's reagent, and methyl blue. Spore size was measured in water and calculated on 50 ascospores discharged from mature asci in 4 ascomata. The collection has been preserved as dried material and slides (MCVE). Herbarium abbreviation follows Holmgren & Holmgren (2012).

Sporormiella spp. that have not been recombined in that genus, are indicated by presenting the current binomials with the genus enclosed in quotation marks (e.g., "*Sporormia*" *carpinea*, "*Preussia*" *variispora*).

Taxonomy

Sporormiella octomegaspora Doveri & Sarrocco, **sp. nov.** Pl. 1–3

MycoBank 803276

Differs from all other *Sporormiella* species by its larger 8-celled ascospores.

Type: Spain, Andalusia, Huelva, Doñana National Park (approx. 37°N 6.5°W), on red deer dung, 28.10.2008, leg. A. Suárez (**Holotype**, MCVE 27409)

Etymology: From the Greek *octo* = eight, *megas* = large, *spora* = spore, referring to the large, 8-celled ascospores

Macrocharacters — pseudothecia 720–900 × 400–500 µm, ostiolate, pyriform, semi-membranous, dark brown to blackish, rough, tomentose owing to a dense net of hyphoid hairs spread all over the ascoma. Neck differentiated, 200–250 × 180–200 µm, cylindrical, somewhat darker, subcoriaceous, with the

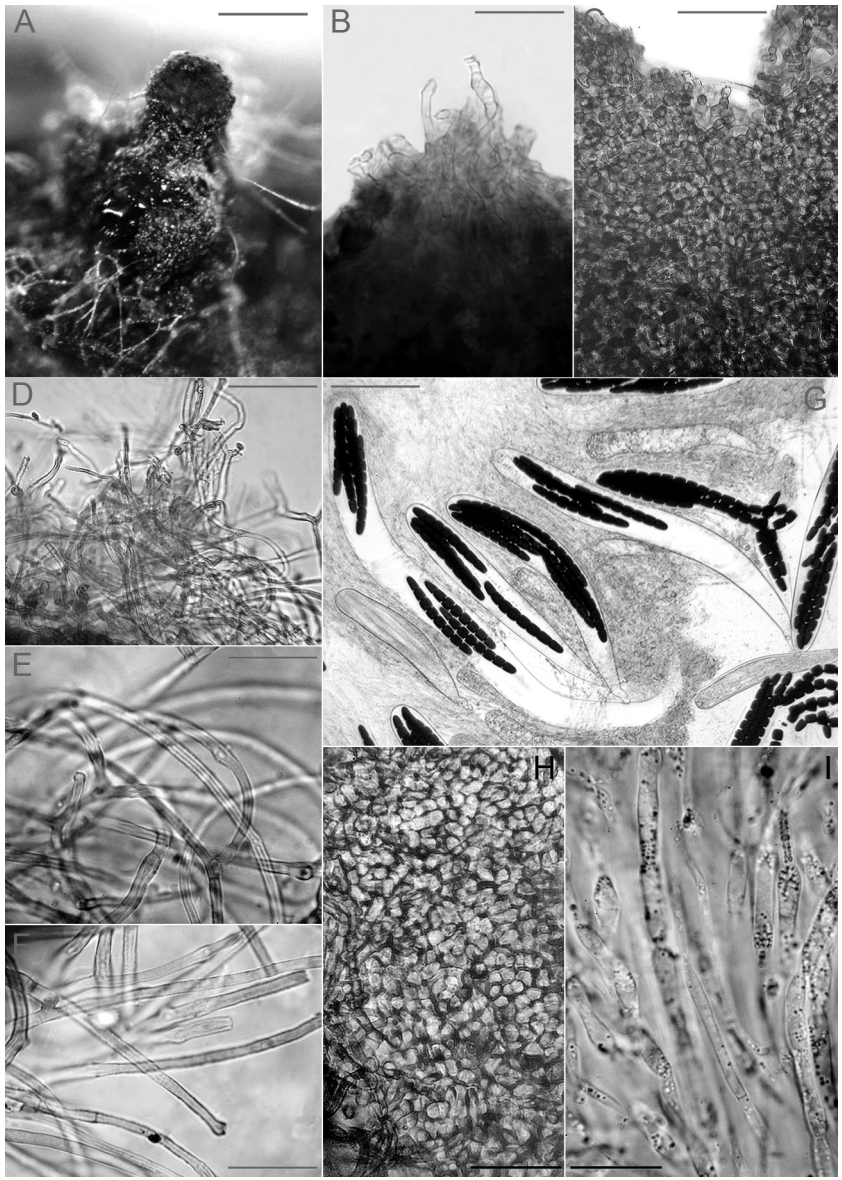

Plate 1. *Sporormiella octomegaspora* (holotype): A = ascoma on dung; B = hairs at the neck apex; C = detail of exoperidium at the neck; D–F = peridial hairs; G = centrum; H = detail of exoperidium; I = Pseudoparaphyses. Scale bars: A = 300 µm; B, E–F = 20 µm; C = 80 µm; D, H = 40 µm; G = 150 µm; H = 25 µm.

same net of hairs at its base and shorter, more isolated hairs at the apex, hardly observable at low magnification (10 ×).

Microcharacters — peridium pseudoparenchymatous, two-layered: endostratum of pale brown to almost hyaline, thin–walled, polygonal cells, 13–18 × 12–15 µm, exostratum a textura angularis of dark brown, thick–walled, polygonal cells, 6–10 × 5–8 µm, with intervals of cylindrical cells in rows (textura prismatica), a textura globulosa-angularis of blunt angular to roundish and somewhat darker cells at the neck, 5–9 µm diam., papillate around the ostiole. Some carbonaceous deposits sprinkled all over, but more abundant on the neck; peridial hairs hyphoid, very long, wavy, strongly intertwined, fairly thick–walled, quite dark brown, septate, often branched, sometimes anastomosed and somewhat encrusted, blunt at the tips, 2–3.5 µm diam.; neck hairs hyphoid at the base, similar in size and shape to peridial hairs, polymorphous, paler, stiffer and shorter at the apex, 15–30 µm long, usually with a rounded, sometimes clavate tip; pseudoparaphyses mixed with the asci and exceeding them, cylindric–filiform, sometimes submoniliform, 3–7 µm diam., often branched, septate, usually slightly tapering upwards, somewhat constricted at the septa, containing abundant hyaline vacuoles; asci 325–450 × 47–72 µm, fissitunicate, inamyloid, 8-spored, cylindric-clavate to clavate, lacking an apical apparatus, roundish or dome–shaped or even slightly flattened at the apex, usually quite abruptly narrowing below in a short stalk, 12–30 µm long; ascospores (148–)152–175(–180) × 16–18(–20) µm, usually three to five bundled in the upper portion of the ascus, the others placed at various levels below, and one usually the lowest, hyaline in the early stages, pale brown later, finally dark brown, cylindric-subfusiform, individually surrounded by a broad gelatinous envelope, smooth, thick-walled, constantly 8-celled, deeply and transversely septate. Cells easily separable at any level, each with a sigmoidal, parallel germ slit. End cells conical, with blunt tips, narrower and usually longer than the middle ones, but often not longer than the second and the seventh, third to sixth cells from the upper end sub-cubical, almost as long as wide, and the third usually the broadest, sometimes slightly wider than long, the second and seventh cell cylindrical, usually longer than wide, all cells with fairly acute angles.

Ecology & Distribution—About fifteen scattered specimens on red-deer (*Cervus elaphus*) dung in the field in October. To date only known from the type locality.

Discussion

Sporormiella octomegaspora is characterised by large and densely hairy ascomata, with hairs widespread both on the pseudothecial venter and neck, large and short-stalked asci, and very large, constantly 8-celled, subfusiform ascospores with sigmoidal germ slits and a particular arrangement inside

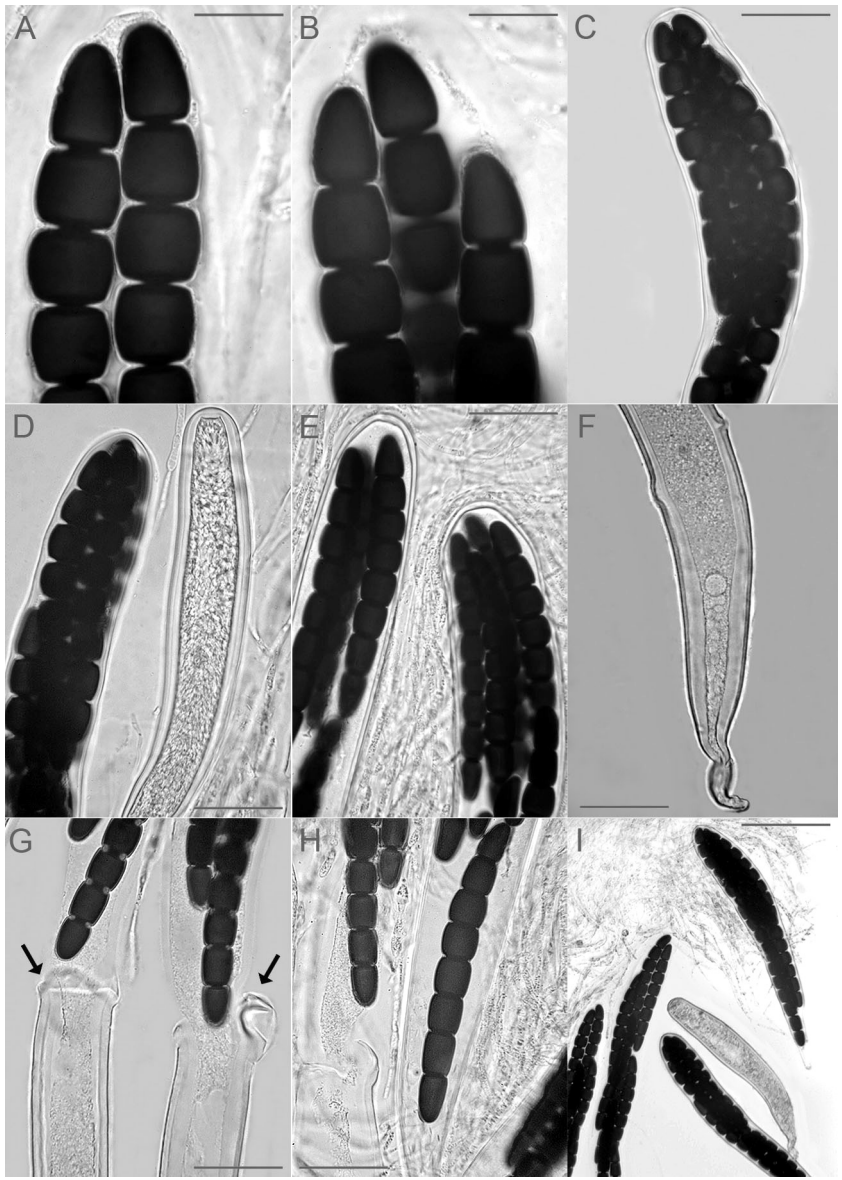

Plate 2. *Sporormiella octomegaspora* (holotype): A–B = upper cells of ascospores at the ascus apex; C-E = upper and middle parts of asci with ascospores; F = ascus base; G = detail of fissitunicate (arrows) asci; H = ascus bases with ascospores. Scale bars: A–B = 20 µm; C–H = 50 µm; I = 150 µm.

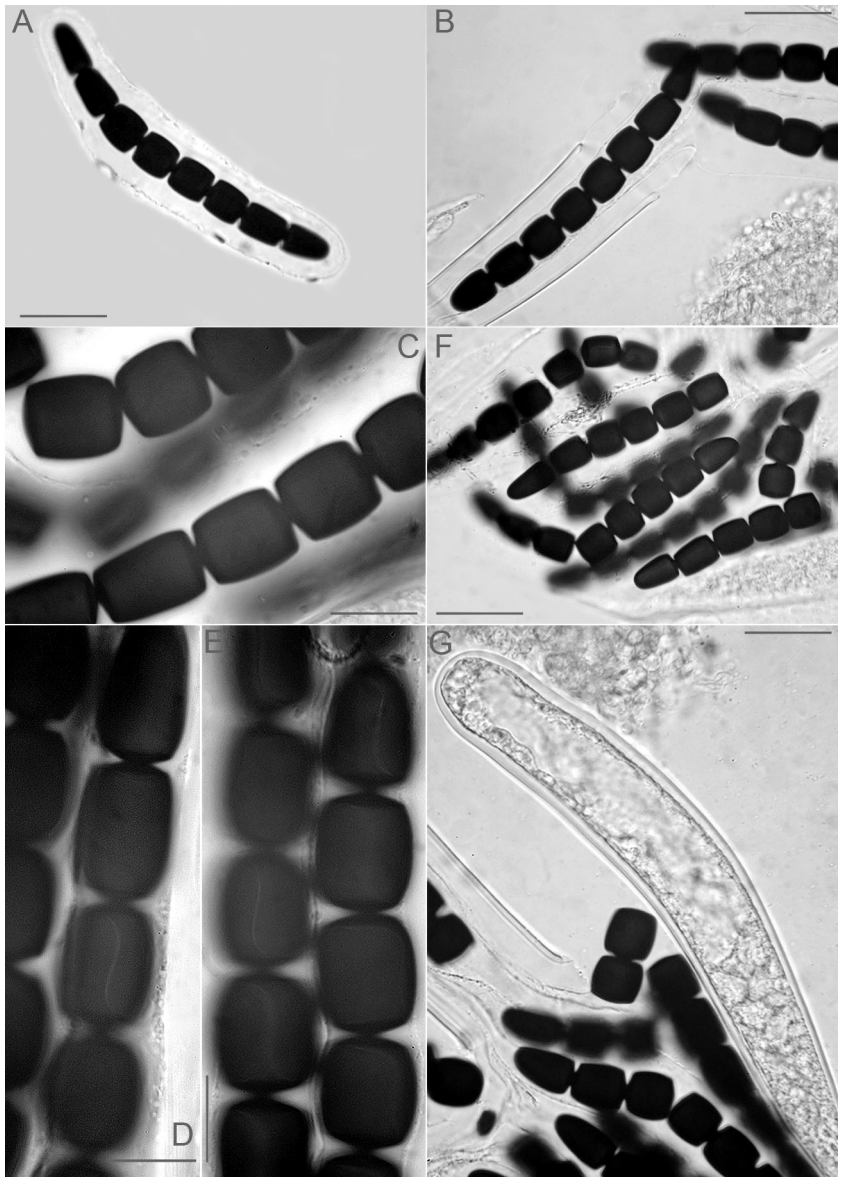

Plate 3. *Sporormiella octomegaspora* (holotype): A–B = ascospores; C–E = detail of ascospores; F–G = spore cells. Scale bars: A–B, F = 35 µm; C = 18 µm; D–E = 15 µm; G = 30 µm.

the asci. The whole of these features allows us to separate *S. octomegaspora* from species with hairy pseudothecia and/or 8-celled ascospores and easily distinguish it from all other *Sporormiella* spp.

In their basic monograph on *Sporormiella*, Ahmed & Cain (1972) reported 3 hairy species with 4-celled ascospores and 18 species with constantly 8-celled ascospores, including the coprophilous "*Sporormia*" *octoloculata* Fabre and "*Sporormia*" *pulchra* E.C. Hansen (both cited as doubtful or not examined by them) and the non-coprophilous, non-examined "*Sporormia*" *carpinea* Fautrey, "*Sporormia*" *gigaspora* Fuckel, *S. subticinensis* (Mouton) Dugan & R.G. Roberts, and *S. ticinensis* (Pirotta) Doveri (Doveri 2004). They also reported 2 species with inconsistently 8-celled ascospores, *S. commutata* (Niessl) S.I. Ahmed & Cain (7–9-celled), and "*Sporormia*" *variabilis* G. Winter (5–8-celled). All *Sporormiella* species described by Ahmed & Cain (1972) — with the exception of *S. insignis* (Niessl) S.I. Ahmed & Cain and *S. splendens* (Cain) S.I. Ahmed & Cain — whether with constantly or inconstantly 8-celled ascospores, differ from *S. octomegaspora* due to ascospores that are much shorter than 100 µm.

Sporormiella splendens has, like *S. octomegaspora*, short–stalked asci, but can be easily distinguished by its glabrous ascomata, smaller pseudothecia and asci, and somewhat shorter but notably narrower cylindrical ascospores (9–13 µm diam., Cain 1934, Ahmed & Cain 1972, Furuya & Udagawa 1972, Leenurm 1998, Treigienė 2004), with middle cells longer than broad and an oblique to diagonal germ slit.

Although it has short-stalked asci, and ascospores with cubical middle cells and an arrangement inside the asci similar to *S. octomegaspora*, *S. insignis* differs in having glabrous and much smaller pseudothecia, smaller asci (Winter 1887, Bayer 1924, Prokhorov & Armenskaya 2003), and somewhat narrower and notably shorter (100–122 × 14–15 µm; Niessl 1878, Ahmed & Cain 1972) ascospores, with middle cells equal in size to each other (so the spore shape is cylindrical rather than cylindric-fusiform) and diagonal germ slits.

Bell (2005), who collected the species in Australia on Eastern Grey kangaroo dung and which she described as *S. insignis*, straddles *S. insignis* and *S. splendens*, "having the ascospore width of the former and the ascospore length of the latter," but in our opinion drawings and measurements reported in her work match those of *S. octomegaspora*. Although the Australian material was unfortunately inadequate for a herbarium sample (Bell, pers. comm.), our examination of a slide from Australia strengthens our original opinion, as Bell's material has only slightly wider spores on average.

Doveri (2004) mentioned and briefly described or discussed (sometimes as *Preussia*) all new *Sporormiella* spp. published after Ahmed & Cain's (1972) monograph, some of which are characterised, like *S. octomegaspora*, by hairy pseudothecia and/or 8-celled ascospores. Among them, the hairy species,

S. lasiocarpa Lorenzo and "*Preussia*" *aquilirostrata* Guarro et al. (Guarro et al. 1997b), have 4-celled and much smaller ascospores [respectively 62–76 × 10–12 µm (Lorenzo 1994), 34–44 × 8.8–11.5 µm (Guarro et al. 1997b)] than *S. octomegaspora;* on the contrary, the species with 8-celled ascospores — *S. oblongiclavata* M.E. Barr & Malloch, *S. tomilinii* O.V. Korol., and "*Preussia*" *variispora* Abdullah et al. — do not have hairy pseudothecia, and "*Preussia*" *variispora* has both 7- and 8-celled ascospores, while all have much smaller ascospores (Abdullah et al. 1999, Barr 2000, Korolyova 2000) than *S. octomegaspora*.

Besides establishing new species of *Sporormiella*, Doveri (2005, 2007), Doveri & Coué (2008), and Chang & Wang (2009) updated the list of new *Sporormiella* spp. (sometimes as *Preussia*) published since 2004. None of them has consistently 8-celled ascospores.

The Welt & Heine (2007) worldwide key to *Sporormiella* spp. with 8-celled ascospores is here updated with the addition of *S. octomegaspora* and other species not previously included:

Worldwide key to *Sporormiella* species with 8-celled ascospores

[N.B.: "***Sporormia octoloculata***" and "***Sporormia***" ***carpinea*** have been omitted from the key, as the original descriptions are meagre]

1a. Ascospores 5–10-celled (never consistently 8-celled) **2**

1b. Ascospores consistently 8-celled **7**

2a. Ascospores 5–8-celled, 62–75 × 14–19 µm (Winter 1874), cylindrical, with middle cells usually broader than long ..... "***Sporormia*" *variabilis***

2b. Ascospores 7–9-celled, different in shape **3**

3a. Ascospores (7–)8–9(–10)-celled, 45–53 × 8–10 µm, fusiform with middle cells wider than long, and the third cell usually the broadest ..... ***Sporormiella* sp.** (described by Richardson 2004b)

3b. Ascospores longer, cylindric-clavate **4**

4a. Ascospores 50–60 × 8–10.5 µm (Ahmed & Cain 1972), usually 9-celled, sometimes 7–8-celled, with the middle cells broader than long, and the third cell from the upper end the broadest ..... ***S. commutata***

4b. 9-celled ascospores rare or absent **5**

5a. Ascospores 65–75 × 7–12 µm, 7–8-celled, with middle cells as long as broad, and the third cell from the upper end the largest ..... "***Preussia*" *variispora***

5b. Ascospores mostly 8-celled, sometimes 7–9-celled **6**

6a. Ascospores 70–90 × 11–13 µm, with the third or fourth cell from the upper end the largest ..... ***Sporormiella* sp.**1 (described by Richardson 2004a)

6b. Ascospores 58–72 × 8–9.5 µm, cylindrical, with isodiametric cells, to cylindric-clavate, with the upper cells shorter than wide ..... ***Sporormiella* sp.** 2 (described by Richardson 2004a)

7a (1b). Ascospores >100 µm long **8**
7b. Ascospores <100 µm long **10**

8a. Ascomata hairy, 720–900 × 400–500 µm. Ascospores 152–175 × 16–18 µm, cylindric-subfusiform with third to sixth cells from the upper end almost as long as wide, and the third usually the broadest. Germ slits sigmoid and parallel . . . . . ***S. octomegaspora***
8b. Ascomata glabrous, smaller. Ascospores narrower, cylindrical. Germ slits oblique to diagonal **9**

9a. Ascomata 550–600 × 350–400 µm. Ascospores 135–165 × 9.5–12.5 µm (Cain 1934), with middle cells longer than broad, cylindrical . . . . . ***S. splendens***
9b. Ascomata subglobose, 250–330 µm diam. Ascospores 100–122 × 14–15 µm (Ahmed & Cain 1972), with middle cells almost equal in size, cubical . . . . . ***S. insignis***

10a. (7b) Ascospores variable in shape, cylindrical to cylindric-fusoid or slightly cylindric-clavate, 50–58 × 6–8 µm, with middle cells equal in width, or the fourth and/or the fifth, or even some other cell, the broadest . . . ***Sporormiella* sp.** (described by Cribb 1994)
10b. Ascospores quite constant in shape **11**

11a. Ascospores cylindrical, with middle cells almost equal in width **12**
11b. Ascospores (sub)clavate, with one broader cell **16**

12a. Ascospores more than 60 µm long **13**
12b. Ascospores less than 60 µm long **14**

13a. Ascospores 79–95 × 14–16 µm (Ahmed & Cain 1972). Middle cells broader than long and with an obliquely transverse germ slit . . . . . ***S. platymera***
13b. Ascospores 72 × 9 µm (Fuckel 1871), with middle cells as long as wide . . . . . "***Sporormia***" ***gigaspora***
(***S. subticinensis*** with ascospores 62–75 × 9–12 µm (von Arx & Storm 1967), is hardly separable from "***Sporormia***" ***gigaspora***, although Mouton (1897) noted that it differs in having clavate rather than oblong asci and much smaller ascomata)

14a (12b). Ascospores 47–57 × 12–14 µm, with middle cells usually broader than long . . . . . "***Sporormia***" ***pulchra***
14b. Ascospores narrower, with middle cells almost as long as wide **15**

15a. Ascospores 48–58 × 6–7 µm with diagonal germ slits . . . . . ***S. bipartis***
15b. Ascospores 40–44 × 8 µm (Pirotta 1878). Germ slits not described . . ***S. ticinensis***

16a (11b). Ascospores 30–48 × 4–7 µm, with the second cell from the upper end the broadest, and with almost parallel germ slits. Non-coprophilous . . . . . ***S. oblongiclavata***
16b. Third or fourth cell the broadest. Germ slits oblique to diagonal (not described in ***S. tomilinii***). Coprophilous species **17**

17a. Third cell the broadest. Cells easily separable to non-separable **18**
17b. Fourth cell the broadest. Cells hardly separable **23**

18a. Ascospores 38–43.5 × 8–9 µm (Doveri 2008) (40 × 5–6 µm, Auerswald 1868; 40–48 × 7–8 µm, Ahmed & Cain 1972). Cells easily separable from each other, and the end ones conic-ovoid. Asci gently narrowing below in a long stalk ..... ***S. octomera***

18b. Ascospores larger **19**

19a. Asci abruptly contracted in a very short stipe **20**

19b. Asci gently narrowed in a stipe. Ascospores with (sub)conical end cells **21**

20a. Ascospores 48.5–63 × 12.5–14.5 µm (Doveri 2008) (48–58 × 12–14 µm, Ahmed & Cain 1972). Spore cells hardly separable, end cells hemispheric ..... ***S. octonalis***

20b. Ascospores 52–57 × 8–9 µm (Ahmed & Cain 1972), with cells easily separable, end cells subconical ..... ***S. schadospora***

21a (19b). Ascospores 71–90 × 12–16 µm (Bommer & Rousseau 1886) (65–80 × 12–15 µm, Ahmed & Cain 1972). Asci short-stipitate ..... ***S. affinis***

21b. Ascospores shorter **22**

22a. Ascospores 52.5–60 × 9.5–10.5 µm (Doveri 2008) (50–59 × 10–11.5 µm, Ahmed & Cain 1972). Cells not separable. Asci long-stipitate ..... ***S. corynespora***

22b. Ascospores 55–70 × 13–15 µm. Cells easily separable. Asci short-stipitate ..... ***S. tomilinii***

23a (17b). Ascospores 49–58 × 9–10 µm (Ahmed & Cain 1972). Asci abruptly short-stipitate ..... ***S. ontariensis***

23b. Ascospores smaller **24**

24a. Ascospores 40–51.5 × 7.5–8.5 µm (Doveri 2008) (40–49 × 8–9 µm, Ahmed & Cain 1972). Asci abruptly short-stipitate ..... ***S. pascua***

24b. Ascospores 34–42 × 7 µm (Doveri 2008) (32–36 × 5.5–6.5 µm, Ahmed & Cain 1972). Asci gently short-stipitate ..... ***S. minipascua***

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