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SOME NEW SPECIES OF SACCOBOLUS

J. VAN BRUMMELEN
Rijksherbarium, Leiden

(With five Text-figures)

Five new species of *Saccobolus* are described: *Saccobolus sphaerosporus* from Germany, *S. purpureus* and *S. parvisporus* from Libya, *S. eleutherosporus* from Belgium, and *S. diaphanus* from Thailand.

Since the publication of my monograph of *Ascobolus* and *Saccobolus* (van Brummelen, 1967) many new data have come available for both genera. Especially in the genus *Saccobolus* several new species have been found, some of which have shown to be constant in cultures during long periods.

In *Saccobolus* the arrangement of the ascospores in a cluster proved to be a valuable character, not only for the distinction of species but also as a character to recognize natural sections within the genus.

Apart from the patterns of arrangement, which can be distinguished after the disposition of the ascospores in the cluster (van Brummelen, l.c.: 40) it is important to notice eventual changes in the disposition during ripening. In species with a changing arrangement of the ascospores or in species with loose ascospores the typical pattern of arrangement can usually be established just before or shortly after the violet pigment becomes visible on the surface of the epispore.

The coherence between the ascospores in different species of *Saccobolus* shows a wide range of possibilities. The spores in the spore-cluster may be completely loose to very firmly united. More or less independently from this the longitudinal contraction of the mature spore-cluster proves to be constant in most species.

Thanks are due to Mr. St. Aubyn Glynn and the late Mr. A. Vervliet for sending living material and to Dr. H. O. Sleumer and Dr. C. F. van Beusekom for providing samples of dung from tropical countries.

Saccobolus sphaerosporus Brumm., *spec. nov.*—Fig. 1

Apothecia angustata parva sessilia, 0,2–0,5 mm diam. Receptaculum initio globulare usque ovoideum et album, denique applanatum usque pulvinatum et dilute brunneum, laeve. Excipulum ad basim strato superficiali e textura globulosa praeditum. Asci initio late obovati, denique late clavati, apice tholiformes, 72–100 × 25–36 μm, 8-spori, pariete iodo caerulescente. Sporarum fasciculi saepe elongati (44–85 × 11–22 μm), interdum compactissimi (29 × 22 μm). Ascosporae saepe in series irregulariter dispositae, sphaericae, violascentes, 9,8–11,2(–12,9) μm diam., fissuris brevibus foveolisque minutis ornatae. Paraphyses sparsim ramosae, cylindrico-clavatae, 1,8–2,2 μm latae, apice leviter inflatae, cellulis terminalibus materia lutea repletis. In fimo ovino crescens.

TYPUS: *St. Aubyn Glynn*, Wirceburgum, Germania, 14.VI.1973 (L).

Apothecia solitary or closely crowded, superficial, sessile on a rather narrow base, 0.2–0.5 mm across, 0.2–0.4 mm high, watery-fleshy. Receptacle at first globular to ovoid and white, then flattened to pulvinate and pale brownish, smooth, without margin. Disk at first convex and yellow to amber-coloured, then more flattened and becoming brownish, dotted with the dark-brown protruding tips of asci. Hymenium 50–80 μm thick. Hypothecium very thin, of only a few layers of subglobular or ellipsoid cells 3–10 × 3–4.5 μm. Flesh not clearly differentiated. Excipulum consisting of a group of closely compacted subglobular cells 3–6 × 3–4 μm (textura globulosa) near the base of the fruit-body and a palisade of thin-walled hyaline hyphae up to 4 μm wide more upwards. Asci at first broadly obovate, then broadly clavate, gradually tapering downwards into a rather thick base, with broad dome-shaped apex, 72–100 × 25–36 μm, 8-spored, the wall blue in Melzer's reagent. Spore-clusters usually elongated, 44–85 × 11–22 μm, but sometimes also very compacted down to 29 × 22 μm. Ascospores irregularly disposed in a cluster, often in one row of eight

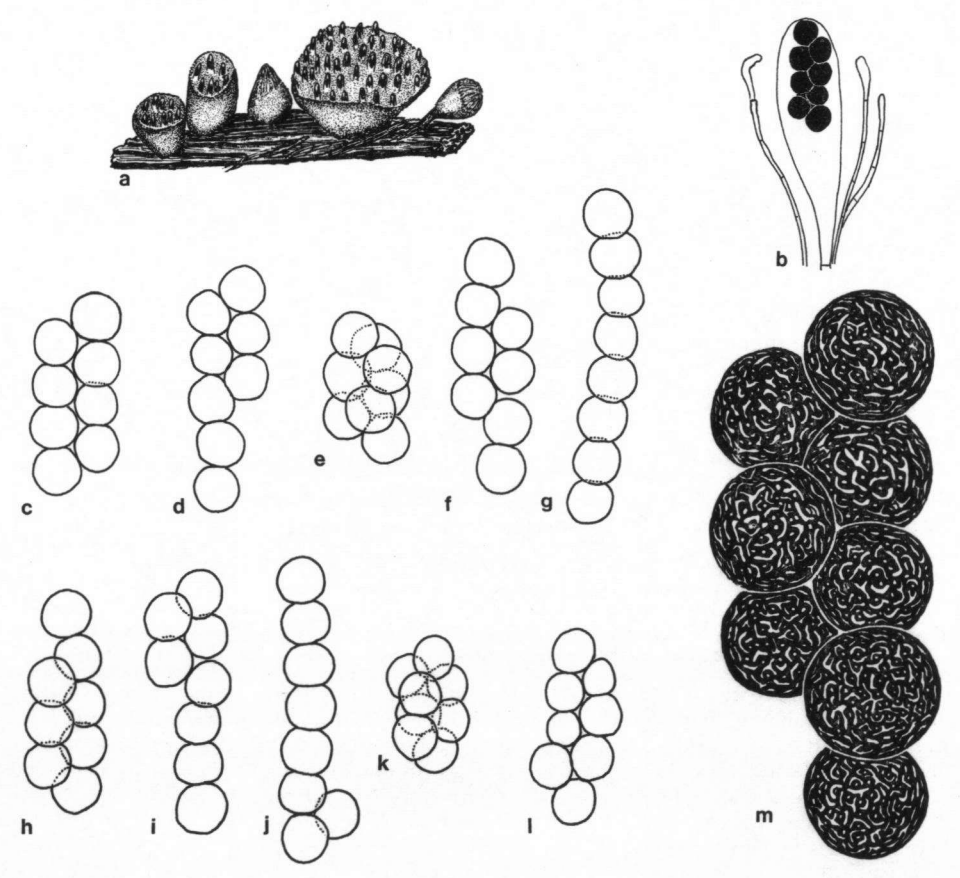

Fig. 1. *Saccobolus sphaerosporus*. — a. Habit of fruit-bodies × 50. — b. Ascus and paraphyses × 320. — c–l. Diagrams of spore-clusters × 630. — m. Spore-cluster × 1600. (From type.)

spores or two rows of one to seven spores, more rarely densely united in an ellipsoid mass, spherical, at first hyaline, then pinkish-violet, finally violet to purplish-brown, 9.8–11.2(–12.9) μm in diameter, ornamented with a very fine pattern of small pits and short more or less branching curved fissures; pigment 0.4–0.8(–1.2) μm thick. Paraphyses cylindric-clavate, sparsely branched, septate, 1.8–2.2 μm thick, enlarged up to 3.5 μm thick at the tip, especially the upper part with yellow contents.

HABITAT.—Only known from dung of sheep.

ETYMOLOGY.—From Greek, σφαιρα, a ball, a globe and σπορα, a seed: with spherical spores.

SPECIMENS EXAMINED.—GERMANY: *St. Aubyn Glynn*, on sheep dung and blotting paper, Würzburg, 14.VI.1973. A pure culture, isolated from this material, has also been examined (L, type).

Mainly on the ground of the presence of yellowish pigment in the paraphyses and the absence of any violet or brownish intercellular pigment, this species is placed in *Saccobolus* sect. *Saccobolus*.

The spherical shape of the ascospores does not lead to a constant or characteristic arrangement of the spores in a cluster.

Judging by the occasionally very compact spore-clusters, the ornamentation and the volume of the ascospores, *S. sphaerosporus* is related to *S. truncatus* Vel.

Another species which seems to be related is *Saccobolus platensis* Gam. & Ran. But here the spores are subfusoid with truncate ends, while longitudinal shortening of the spore-cluster is not reported by Gamundi & Ranalli (1969).

Saccobolus purpureus Brumm., *spec. nov.*—Fig. 2

Apothecia sessilia, 0,15–0,50 mm diam. Receptaculum initio hemisphaericum et dilute purpureo-roseum, denique obconicum et magis fusco-purpureum, postremo pulvinatum vel raro subcylindricum, laeve. Excipulum strato superficiali e textura globulosa praeditum, pigmento roseo-purpureo incrustatum. Asci clavati, apice truncati, 110–120 × 22–26 μm, 8-spori, pariete iodo caerulescente. Sporarum fasciculi elongati, arcte adpressi, 34–36 × 14–15 μm. Ascosporae secundum typum II dispositae, initio fusiformi-ellipsoideae et hyalinae, denique asymmetricae, trigonae vel ventricosae et intense purpureo-violascentes, 13,8–15,6 × 7,3–8,3 μm, laeves, saepe lineis irregularibus tenuissimis ornatae. Paraphyses ramosae, cylindricae, 2,5–3,7 μm latae, apice leviter inflatae, interdum pigmento purpureo-roseo obtectae. In fimo asinorum et animalis insectivori ignoti invenitur.

TYPUS: *van Brummelen 3290*, prope Jabo in Djebel Nefusa, Libya (L).

Apothecia solitary or in small groups, superficial, sessile on a broad base, 0.15–0.50 mm across, 0.25–0.35 mm high, watery-fleshy. Receptacle at first hemispherical and pale purple-red, then obconical and more brownish purple, finally pulvinate or rarely subcylindrical; surface smooth; margin not differentiated. Disk convex, at first colourless, then dark violet from ripe ascospores, roughened by the protruding tips of ripe asci. Hymenium 80–100 μm thick. Hypothecium very thin, of only a very few layers of isodiametric cells 2.5–7 × 2.5–6 μm. Flesh not clearly differentiated. Excipulum 6–10 μm thick, of one or two layers of isodiametric or slightly oblong cells 5–9 × 3–7 μm (textura globulosa), with maturity restricted to the base of the fruit-body, with purple-red, amorphous, intercellular, water-soluble pigment. Asci clavate, with truncate apex, 110–120 × 22–26 μm, 8-spored, the wall blue in Melzer's

reagent. Spore-clusters very compact, elongated, 34–36 × 14–15 μm, with unilateral mucilaginous substance. Ascospores arranged according to a very compact form of pattern II, at first fusiform-ellipsoid and hyaline, then asymmetrical, subtrigonal, or ventricose and dark purple-violet, finally brownish-purple, 13.8–15.6 × 7.3–8.3 μm, smooth with a few irregular secundary cracks taking the shape of delicate lines over the whole of the spore-cluster; pigment in a rather thick layer (0.5–1.3 μm thick). Paraphyses branched, septate, cylindrical, 2.5–3.7 μm thick, not or very slightly enlarged up to 5 μm at the tip, with colourless contents; the apex sometimes covered with small purple-red crystals.

HABITAT.—On dung of donkey and of unknown insectivorous animal.

ETYMOLOGY.—From Latin, *purpureus*, purple or purple-red colour.

SPECIMENS EXAMINED.—LIBYA: *van Brummelen 3290*, on dung of donkey (comm. Dr. H. O. Sleumer), near Jabo in the Djebel Nefusa, c. 170 km S. W. of Tripoli,

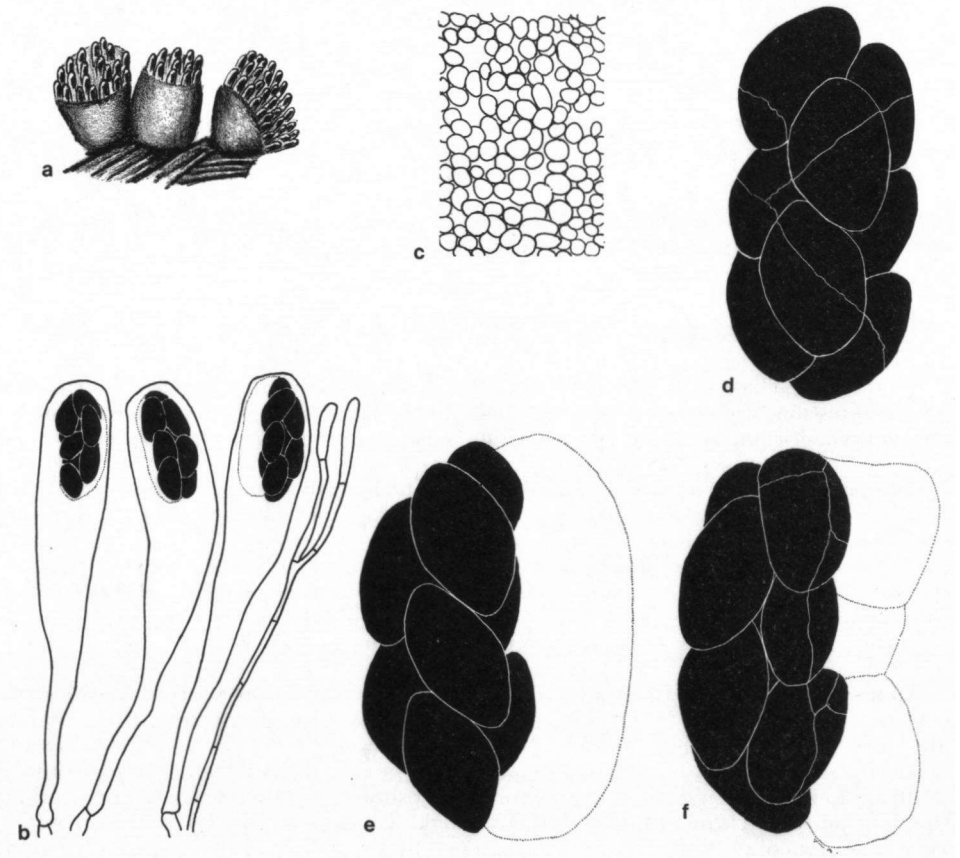

Fig. 2. *Saccobolus purpureus*. — a. Habit of fruit-bodies × 50. — b. Asci and paraphysis × 500. — c. Texture of excipulum seen from outside × 1000. — d–f. Spore-clusters × 1600. (From type.)

14.V.1971 (L, type); *van Brummelen 3295*, on dung of unknown insertivorous animal (comm. Dr. H. O. Sleumer), near Bir Ghanam, c. 70 km S. of Tripoli, 9.VIII.1971 (L).

This is a representative of *Saccobolus* sect. *Eriobolus* Sacc. Obviously *Saccobolus purpureus* is close to *S. versicolor* (P. Karst.) P. Karst., from which it can be distinguished by the conspicuous purple-red colour and the very compact spore-clusters. In *S. versicolor* the pigment of the receptacle is bluish-violet and does not tend to purple-red. The spore-clusters in *S. versicolor* may sometimes be rather compact. They measure 40–62 × 14–19(–23) μm (van Brummelen, 1967), which is considerably longer than in *S. purpureus*.

In *Saccobolus verrucisporus* Brumm. ascospores and spore-clusters of the same shape and size occur, but in this species the ascospores are ornamented with coarse, isolated warts, while the receptacle is whitish with a shade of violet.

Saccobolus parvisporus Brumm., *spec. nov.*—Fig. 3

Apothecia sessilia, solitaria, interdum confluentia, 0,3–0,6 mm diam. Receptaculum initio subglobulare et dilute roseum usque dilute violaceum, denique pulvinatum et sordide roseum usque dilute carneum, laeve. Excipulum strato superficiali textura globulosa praeditum, hyalinum. Asci clavati, apice truncati, 122–132 × 15–18 μm, 8-spori, pariete iodo intense caerulescente. Sporarum fasciculi elongati, valde laxi, raro compacti, (23–)27–29 × 10–11 μm. Ascosporae secundum typum II vel interdum secundum typum IIIa dispositae, ellipsoideae, violascentes, 9,5–10,3 × 5,1–6,3 μm. Episporium pigmenti disposito verrucis admodum grossis granulisque irregularibus obtectum, basi saepe strato tenui. Paraphyses simplices vel ramosae, cylindraceae, 1,8–2,3 μm latae, apice leviter inflatae, hyalinae. In fimo asinorum invenitur.

TYPUS: *van Brummelen 3288*, prope Jabo in Djebel Nefusa, Libya (L).

Apothecia solitary or in small coherent groups, sometimes apparently confluent, superficial, sessile on a broad base, 0.3–0.6 mm across, 0.3–0.4 mm high, watery-fleshy. Receptacle at first subglobular and pale pinkish to pale violet, then pulvinate and dingy pinkish to pale flesh-coloured, smooth, without margin. Disk convex, translucent pale pinkish, roughened by protruding asci. Hymenium 90–110 μm thick. Hypothecium thin, of closely compacted isodiametric cells 3–7.5 μm in diameter. Flesh not differentiated. Excipulum very thin, consisting of subglobular cells (textura globulosa), hyaline, scarcely pigmented, with maturity restricted to base of fruit-body. Asci clavate, with a long stalk, with truncate apex, 122–132 × 15–18 μm, 8-spored; the wall deep blue in Melzer's reagent. Spore-clusters elongated, 27–29 × 10–11 μm, sometimes becoming more compact 23–26 × 11 μm. Ascospores rather loosely united in a cluster, arranged according to pattern II, sometimes changing into pattern IIIa, ellipsoid, at first hyaline, then pale violet, becoming darker in concentrations of pigment, 9.5–10.3 × 5.1–6.3 μm; ornamented with irregular large warts and granules, often with thin uniform layer at base; mucilaginous substance very fugacious or absent. Paraphyses simple or branched, septate, cylindrical, 1.8–2.3 μm thick, slightly enlarged up to 4 μm at the tip, colourless.

HABITAT.—Known only from dung of donkey.

ETYMOLOGY.—From Latin, *parvus*, little, small and *sporus*, a seed: with small spores.

SPECIMEN EXAMINED.—LIBYA: *van Brummelen 3288*, on dung of donkey (comm. Dr. H. O. Sleumer), near Jabo in the Djebel Nefusa, c. 170 km S. W. of Tripoli, 11.V.1971 (L, type).

Judging by the arrangement of the ascospores in the cluster and the absence of yellowish pigment in the paraphyses this is a typical representative of *Saccobolus* sect. *Eriobolus*. The ascospores are longitudinally disposed with two rows of three and one row of two spores. In rare cases the cluster becomes longitudinally contracted with rather irregular arrangement of the spores.

As a result of the irregular distribution of the pigment the ascospores are only loosely united in the cluster. In squash-preparations the spores will easily break apart.

This species resembles *Saccobolus thaxteri* Brumm. in many respects. However, the

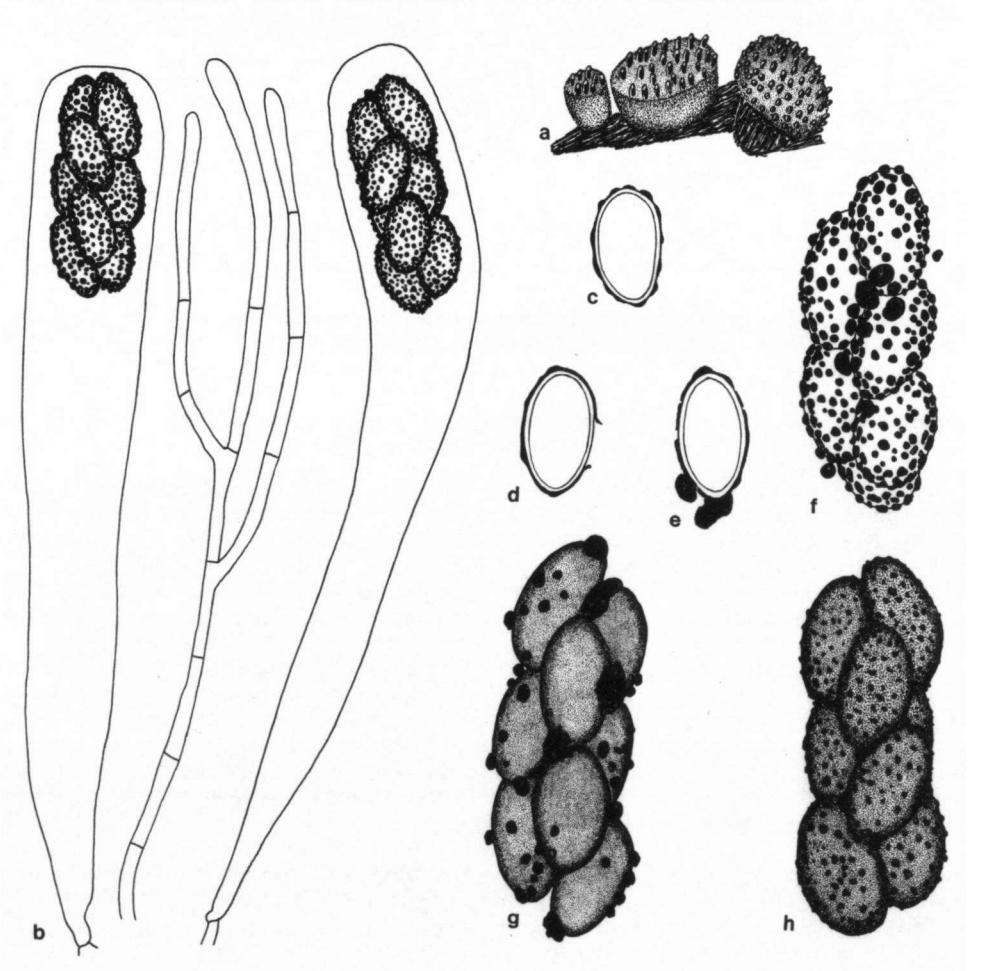

Fig. 3. *Saccobolus parvisporus*. — a. Habit of fruit-bodies × 50. — b. Asci and paraphysis × 1000. — c–e. Ascospores in optical section × 1600. — f–h. Spore-clusters × 1600. (From type.)

differences are evident. In *Saccobolus parvisporus* the ascospores are considerably smaller, the length-breadth ratio is higher, the spore-clusters are more slender, and the asci are much longer.

In *Saccobolus infestans* (Batista & Pontual) Brumm. ascospores of about the same size (9–11 × 5–6.5 μm) occur, but the arrangement of the spores in the cluster is quite different (c.f. van Brummelen, 1967).

Saccobolus eleutherosporus Brumm., *spec. nov.*—Fig. 4

Apothecia sessilia, basi angustata, 0,10–0,25 mm diam. Receptaculum initio subglobulare et dilute roseum, denique applanatum usque pulvinatum et malvinum, leviter asperum. Excipulum strato superficiali e textura globulosa praeditum. Asci cylindrico-clavati, apice truncati, 95–120 × 22–27 μm, 8-spori, pariete iodo caerulescente. Sporarum fasciculi elongati, valde laxi, facile soluti. Ascosporae non congregatae, secundum typum II dispositae, elongato-ellipsoideae, initio hyalinae, denique violascentes, 16,5–17,5 × 6,4–7,6 μm. Episporium pigmenti verrucis vel molibus irregulariter obtectum, ascosporis ipsis pro parte majore pigmento

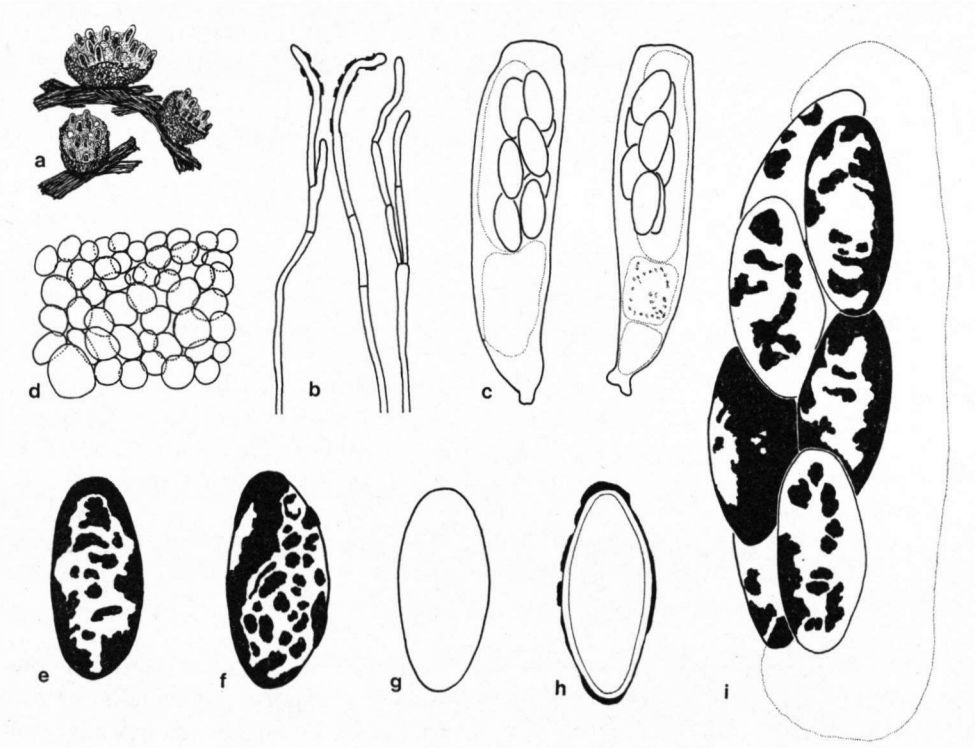

Fig. 4. *Saccobolus eleutherosporus*. — a. Habit of fruit-bodies × 50. — b. Paraphyses × 500. — c. Asci × 500. — d. Texture of excipulum seen from outside × 500. — e–g. Ascospores × 1600. — h. Ascospore in optical section × 1600. — i. Spore-cluster × 1600. (From type).

destitutis. Paraphyses valde ramosae, irregulariter filiformes, 2–3 μm latae, apice leviter inflatae, pigmento dilute roseo obtectae. In fimo Phasiani colchici invenitur.

TYPUS: *A. Vervliet*, Ekeren prope Antverpiam, Belgia, 28.II.1971 (L).

Apothecia solitary or in small groups, not confluent, superficial, sessile on a narrow base, 0.10–0.25 mm across, 0.10–0.15 mm high, soft fleshy. Receptacle at first subglobular and pale pinkish, then flattened to pulvinate and pale purplish pink, in a few cases slightly yellowish; surface finely roughened by subglobular cells; margin not differentiated. Disk convex, at first pink, then pale purplish pink, roughened by the pale violet protruding tips of ripe asci. Hymenium 90–100 μm thick. Hypothecium very thin, of subglobular cells 5–15 μm in diameter (textura globulosa). Asci cylindric-clavate, with a short stalk, truncate at the apex, 95–120 × 22–27 μm, 8-spored; the wall blue in Melzer's reagent. Spore-clusters elongated, very loose. Ascospores not cemented together by their pigment, at first free, then clinging together according to pattern II, finally often more or less free, ellipsoid or elongated-ellipsoid (length-breadth ratio 2.2–2.7), often somewhat ventricose; hyaline, then pale violet, finally pale purplish brown; 16.5–17.5 × 6.4–7.6 μm; ornamented with very irregular coarse warts and thick lumps of pigment, leaving large parts of the wall uncovered; with common unilateral mucilaginous substance. Paraphyses rather frequently branched, septate, irregularly filiform, 2–3 μm thick, not or only slightly enlarged up to 4 μm at the tip; covered near the ends with pale pinkish, amorphous pigment; pigment staining brownish red with iodine.

HABITAT.—Known only from dung of pheasant.

ETYMOLOGY.—From Greek, ελευθερος, free and σπορα, a seed: with free spores.

SPECIMEN EXAMINED.—BELGIUM: *A. Vervliet*, on dung of pheasant, Ekeren near Antwerp, 28.II.1971 (L, type).

This material was sent to me as *Saccobolus saccoboloides* (Seaver apud Dodge & Seaver) Brumm., which it resembles because of the free ascospores. In *Saccobolus saccoboloides*, however, the ascospores are arranged in a cluster according to pattern I (van Brummelen, 1967), the ornamentation of the ascospores consists of a uniform, thin layer, and the terminal elements of the paraphyses are filled with yellowish contents. This makes *S. saccoboloides* a typical representative of *Saccobolus* sect. *Saccobolus*.

Saccobolus eleutherosporus is also characterized by free ascospores, but the ascospores are arranged in a cluster according to pattern II and the pigment is irregularly distributed over the surface of the ascospores. The contents of the paraphyses are colourless, while their terminal elements are covered with amorphous pinkish pigment. These characters together with the unilateral attachment of the mucilaginous substance to the spore-cluster make the new species a typical representative of *Saccobolus* sect. *Eriobolus* Sacc.

Saccobolus eleutherosporus is closely related to *Saccobolus versicolor* (P. Karst.) P. Karst., which is a very variable species. But it can be distinguished from this by the free ascospores and the very pale, irregularly distributed sporal pigment.

At maturity the spores of *S. eleutherosporus* are loosely united in the upper part of the ascus by the mucilaginous substance. Soon after ascospore discharge this substance dissolves and the spores are set completely free.

Saccobolus diaphanus Brumm., *spec. nov.*—Fig. 5

Apothecia sessilia, 0,20–0,25 mm diam. Receptaculum initio obconicum, basi leviter constrictum, deinde applanatum, albidum, laeve. Excipulum strato superficiali e textura globulosa usque angularis praeditum. Asci late clavati, apice truncati, 73–86 × 21–23 μm, (7–)8-spori, pariete iodo caerulescente. Sporarum fasciculi satis laxi, facile soluti, 27–32 × 10–12 μm. Ascosporae secundum typum II, vel saepius irregulariter dispositae, late ellipsoideae usque ellipsoideae, 9,5–10,7 × 5,3–6,5 μm, saepe omnino laeves, tantum raro subtiliter punctatae. Paraphyses simplices, irregulariter cylindricae, 1,7–2,2 μm latae, hyalinae. In fimo equorum invenitur.

TYPUS: *van Brummelen 2800*, Chiengmai, Doi Pui, Thailandia (L).

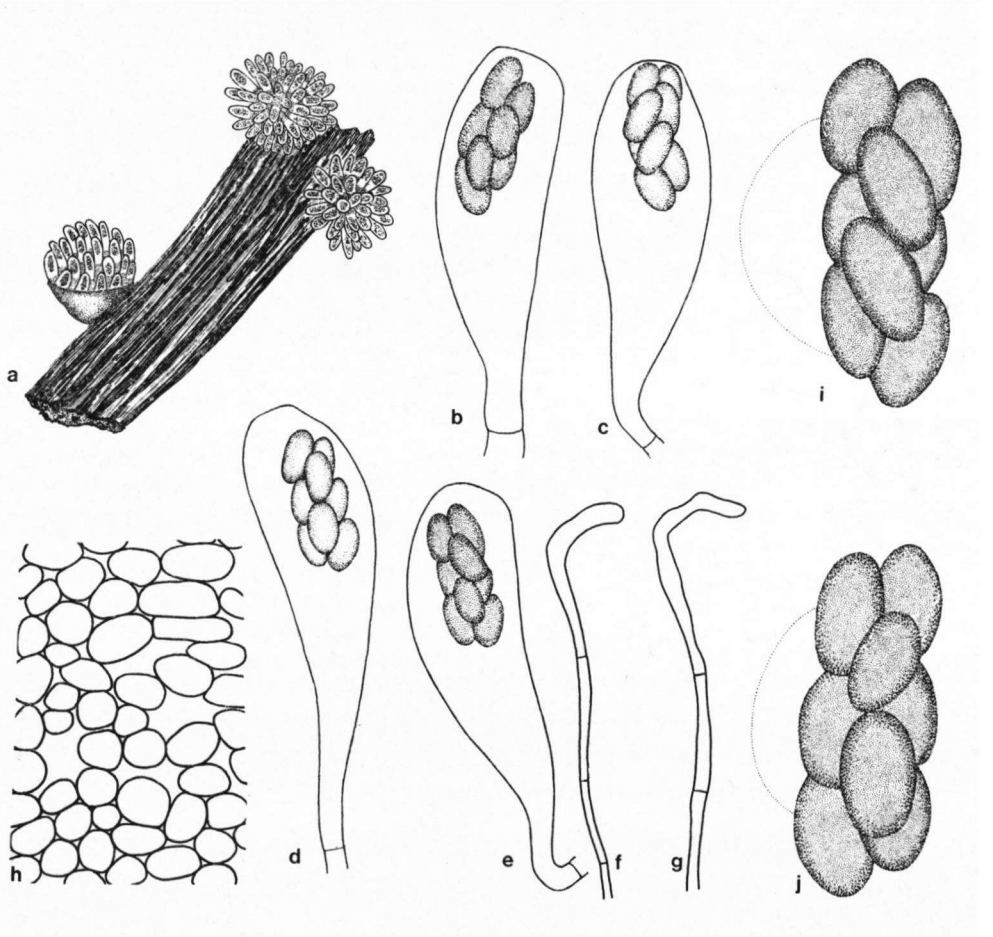

Fig. 5. *Saccobolus diaphanus*. — a. Habit of fruit-bodies × 50. — b–e. Asci × 630. — f, g. Upper parts of paraphyses × 1000. — h. Texture of excipulum seen from outside × 630. — i, j. Spore-clusters. (From type.)

Apothecia solitary, superficial, sessile, 0.20–0.25 mm across, about 0.20 mm high. Receptacle at first obconical with a narrow base, then more expanded, white, smooth; margin retracted towards the base at maturity. Disk convex, translucent white, then pale violet, dotted with the brown protruding tips of ripe asci. Hymenium up to about 65 μm thick. Hypothecium very thin, of isodiametric cells 4–8 μm wide. Flesh not or scarcely differentiated. Excipulum of only one layer of isodiametric or slightly elongated (sometimes angular) cells 8–14×8–10 μm (textura globulosa to angularis), hyaline. Asci broadly clavate, gradually tapering downwards into a rather thick base, with truncate apex, 73–86×21–23 μm, (7–)8-spored; the wall blue in Melzer's reagent. Spore-clusters rather loose, 27–32×10–12 μm. Ascospores loosely arranged according to pattern II or more often irregularly disposed, broadly ellipsoid to ellipsoid (length-breadth ratio 1.5–1.9), at first hyaline, then very pale violet, finally pale brownish violet, 9.5–10.7×5.3–6.5 μm, usually wholly smooth, more rarely finely punctate; pigment in a very thin layer 0.2–0.4 μm thick. Paraphyses simple, septate, irregularly cylindrical, 1.7–2.2 μm thick, frequently hooked at the slightly enlarged tips (up to 3 μm), hyaline.

HABITAT.—Known only from dung of horse.

ETYMOLOGY.—From Greek, διαφανης: transparent, translucent.

SPECIMEN EXAMINED.—THAILAND: *van Brummelen 2800*, on dung of horse (comm. Dr. C. F. van Beusekom), Chiengmai, Doi Pui (alt. c. 1500 m), 14.IV.1970 (L, type).

In this species the layer of pigment covering the ascospores is very thin, which gives them the pale, translucent colour. At maturity the ascospores are scarcely or not cemented together by their pigment. This leaves them often rather loose in the spore-cluster. At a certain stage of maturity a typical arrangement of the spores, according to pattern II (i.e. with two longitudinal rows of three and one of two spores), is attained. But this disposition is not sufficiently fixed by the scanty pigment, so that more or less aberrant arrangements are found.

In rare cases asci with only seven spores were found. In such asci usually one spore was found to be bigger than the others, measuring 11.9–13.4×6.5–7.0 μm.

Saccobolus diaphanus is a representative of *Saccobolus* sect. *Eriobolus* Sacc. that should be placed close to *Saccobolus depauperatus* (Berk. & Broome) E. C. Hansen. From this it can be distinguished by the pale, somewhat smaller (especially shorter) ascospores, that are only loosely united.

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