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Edited by
Gary A. Laursen
School of Agriculture and Land Resources Management
University of Alaska
Fairbanks, Alaska

Joseph F. Ammirati
Department of Botany
University of Washington
Seattle, Washington

and
Scott A. Redhead
Biosystematics Research Centre
Ottawa, Ontario, Canada

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THREE 4-SPORED *SACCOBOLUS* SPECIES FROM NORTH EAST GREENLAND

Henry Dissing

University of Copenhagen, Institut for Sporeplanter
ϕFarimagsgade 2 D, DK-1353 Copenhagen K, Denmark

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ABSTRACT

Three 4-spored species of the coprophilous genus *Saccobolus* (Order Pezizales) from North East Greenland are treated, viz. *Saccobolus quadrisporus*, *S. groenlandicus* sp. nov., and a third taxon for which only a preliminary description is provided. *Saccobolus groenlandicus* is also reported from Arctic Canada.

INTRODUCTION

During July and August in 1982 and 1983 the author collected Discomycetes on Ella Island (72°30'N 25°W) and around Mestersvig (72°15'N 24°W) in the southern part of the National Park in North East Greenland. About 80 species of Operculate Discomycetes (order Pezizales) were encountered. Many of these are undescribed taxa.

Three 4-spored species of the coprophilous genus *Saccobolus* are treated below. These are *Saccobolus quadrisporus* Mass. & Salm., *S. groenlandicus* sp. nov., and a third species for which a formal description will be postponed until more material is available.

Material of *Saccobolus groenlandicus* also has been obtained from Danmarks Havn (76°46'N 18°48'W), Jameson Land (71°45'N 23°W), in North East Greenland, and from Ellesmere Island (78°53'N 75°35'W), Canada.

Material and methods.

The excellent laboratory facilities in Nyhavn made it possible to prepare detailed notes on microscopic and macroscopic characters of fresh material for all species. Also, fresh goose dung was dried carefully and taken to Copenhagen, where samples were incubated on filter paper in moist chambers at room temperature in daylight.

For sectioning, *Saccobolus groenlandicus* ascocarps were isolated from dung in moist chambers and fixed according to methods described by Jensen (1982: 725). Specimens were then dehydrated and embedded in Spurr's resin and polymerized at 70°C. Sections 2 µm thick were cut on a LKB pyramitone with a glass knife. Sections were stretched in 10% acetone

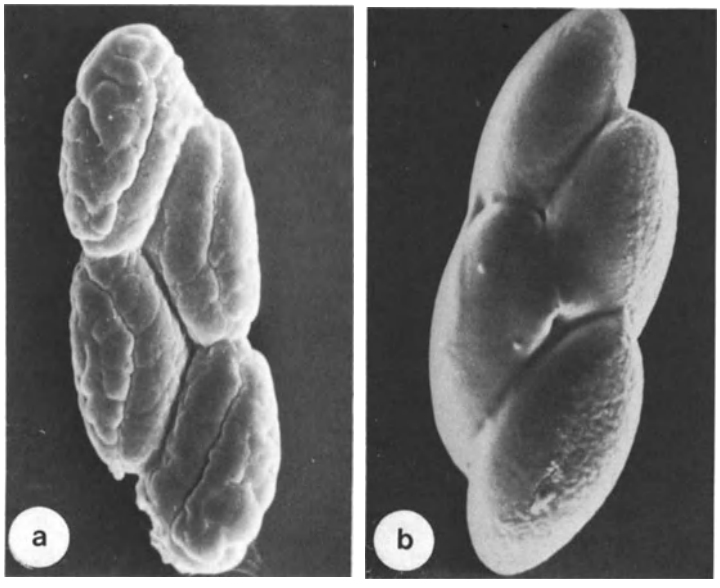

Fig. 1. SEM micrographs of spore clusters. a. *Saccobolus quadrisporus*, Gr. 83.154, b. *S. groenlandicus*, Gr. 83.149. a-b, x 1732.

on a hot plate, dried, then stained in 0.1% methylene blue, followed by 0.3% safranin in 30% ethanol. Next they were rinsed in tap water, dried and mounted in Spurr's reagent. Unless otherwise stated, the material from Greenland is deposited in the Botanical Museum, Copenhagen (C) while material from Ellesmere Island will be deposited at the University of Toronto (TRTC), with duplicates in Copenhagen (C).

Saccobolus quadrisporus Mass. & Salmon Figs. 1a, 2a, and 6a.

Ascocarps turbinate to disc-shaped, finally pulvinate, 0.3-0.6 mm broad, pale purplish to purplish brown. Asci 4-spored, 168-188 x 26-30 µm, staining pale bluish overall in Melzer's reagents. Paraphyses straight, above slightly enlarged to 6-7 µm broad, septate, content hyaline to pale brownish. Ascospores 21-23 x 10-12 µm, ellipsoid to inequilateral, at first hyaline, smooth, then pale greyish purple; when mature very dark purplish, almost opaque in the light microscop; when mature the pigment deposited in a very characteristic way, i.e., in two zones, separated by a longitudinal whitish line: half of the spore towards the center of a cluster nearly smooth, while the side towards the periphery is heavily and irregularly pigmented with nearly confluent warts. Ascospore clusters 50-55 x 17-20 µm (when the spores are displaced in two rows) or 56-58 x 14-16 µm (when displaced in one row), with two separated, clearly visible gelatinous coverings.

Material examined: East Greenland, Jameson Land, Ørsteddalen, fresh dung from Barnacle Goose, collected 17 July 1982, leg. David Boertmann. Incubated in moist chambers 11 January 1983, isolated 27 January 1983 where it was fruiting together with *Saccobolus* sp. nov. (Gr. 82.326) and an undescribed *Ascobolus* species, Gr. 82.327; - Ella Island, near Langesø, on dung from Barnacle Goose, 30 July 1983, Gr. 83.61; - ibid., fresh dung from Barnacle Goose, collected 30 July 1983. Incubated 6 January 1984, isolated 4, 6 and 13 February 1984, Gr. 83.153, 83.154 &

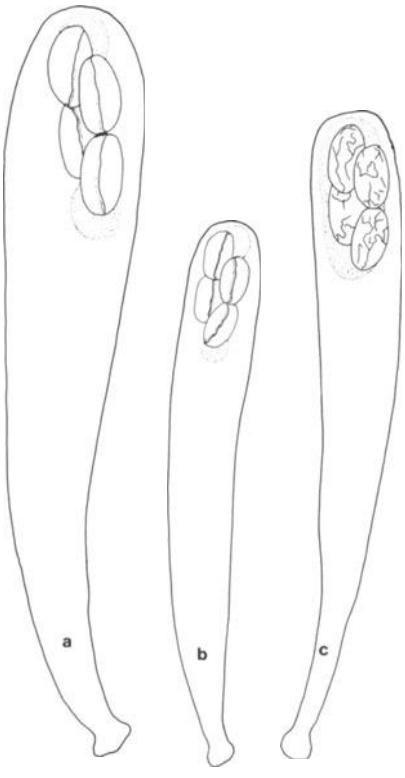

Fig. 2. Asci with spore clusters, schematic. a. *Saccobolus quadrisporus*, Gr. 82.327, b. *Saccobolus* sp. nov., Gr. 82.326, c. *S. groenlandicus*, EI. 84.116, a-c, x 495.

83.158; - Mestersvig, moist area 1 km SW of Nyhavn, on dung from Barnacle Goose, 3 August 1983, Gr. 83.92 & 83.94; ibid., 9 August 1983, Gr. 83.122.

DISCUSSION

The above description is based on notes made in Greenland on fresh material and on moist chamber material in Copenhagen. In all collections examined, the gelatinous coverings of the spore clusters were clearly visible as two separate areas. In nature a brownish amorphous substance was often deposited between cells in the excipulum and between the paraphyses in the hymenium in old apothecia.

Until now *Saccobolus quadrisporus* had been known from only two localities, viz. the type locality in Kew Garden where it was found on goose dung in November 1900 (Massee & Salmon 1901; Brummelen 1967), and from the arctic island of Spitsbergen, also on goose dung (Eckblad 1968).

By chance *S. quadrisporus* was described before *S. groenlandicus* even though the latter seems to be much more common. An example may illustrate this: one day in 1983 while working in the field in Greenland, a great number of goose dung samples (probably all from Barnacle Goose) were inspected with a hand lens. Apothecia of a *Saccobolus* could be seen on about 20 samples. A later inspection of the fresh material in the laboratory revealed that of the 20 samples only 2 were *S. quadrisporus* while 18 were identified as *S. groenlandicus*.

Fig. 3. *Saccobolus groenlandicus*, spore cluster with unilateral gelatinous covering, EI. 84.116, x 1072.

Saccobolus groenlandicus sp. nov. Figs. 1b, 2c, 3, 4a,b, and 5.

Carposoma primum turbinatum vel disciforme, deinde pulvinatum, 0.2-0.6 mm latum, hymenio aqueo-albido vel pallide purpureo ascis prominentibus punctato, extra glabrum infra hyphas fixatorias emittens, pallide purpureum vel - imprimis ad marginem rotundatum inconspicuum versus - vinaceum.

Excipulum ad 600 µm crassum, parietibus cellularum omnino tenuibus; pars medullaris e cellulis brevibus, hyalinis, globularibus vel hyphaceis et hyphis ramificatis, septatis ascogenibus intermixtis composita; cellulae in parte exteriore infra globulares, 6-18 µm diam., supra series plus minus manifestas, sub angulo subrecto ad superficiem directas formantes, extrorsum sensim elongatae, claviformes, 20-30 x 3-5 µm magnae, superficiales contento vinaceo insignes. Subhymenium paulum manifestum.

Hymenium 100-200 µm altum. Asci plerumque 4 spori, maturi longe prominentes, ad 140 µm longi, 20-25 µm lati, infra in bases angustas sensim attenuati, supra truncati, liquore Melzeri omnino pallide coerulescentes. Operculum 6-9 µm latum.

Paraphyses rectae, supra paulum dilatatae, 6-8 µm latae, ramificatae, septatae, brevicellules, interdum anastomosantes, hyalinae vel in summis cellulis contentum pallide vinaceum praebentes. Sporae valde cohaerentes, apicibus sese paulum tegentes, unam seriem vel duas parallelas dislocatas formantes, 16.5-18.5 x 8.5-9.5 µm magnae, ellipsoides vel inaequilaterales, primum hyalinae, deinde purpurascentes, demum colore floris Hellebori nigri, strato pigmentoso tenui, ut in Saccobolo glabro fissuras irregulares in lateribus convexis liberis praebente.

Glomus sporarum duas series formantium 32-40 x 14-16.5 µm, sporarum uniseriatarum 36-42 x 12-14 µm magnum, in altero latere strato gelatineo continuo crasso tectum.

Holotypus die 3 Augusti anni 1983 in fimo Brantae leucopsis in loco aqua transmissa uvido a Mestersvig sinus Kong Oscars Fjord 2 km in occidentem sito (lat. bor. 72°12', long. occ. 24°10') a H. Dissing et S. Sivertsen sub numero 83.95 lectus, siccus in Museo Botanico Hauniensi (C) depositus.

Ascocarps at first turbinate to disc-shaped, then pulvinate, 0.2-0.6 mm broad, hymenium watery whitish to pale purplish, dotted with protruding

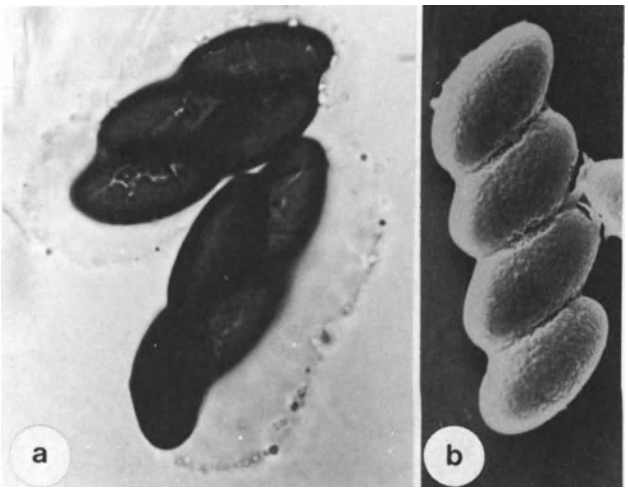

Fig. 4. Spore clusters, *Saccobolus groenlandicus*. a. LM photograph of cluster in water, EI. 84.116, b. SEM micrograph of spores displaced in one row, Gr. 83.149, a. x 1074, b. x 1247.

asci, outside glabrous, below with anchoring hyphae, pale purplish to vinaceous, especially towards the inconspicuous, rounded margin.

Excipulum up to 600 µm thick, throughout of thin walled cells; medullary excipulum of small globose to hyphae-like, short, hyaline cells, intermixed with branching, septate, ascogenous hyphae; outer excipulum below of globose cells, 6-18 µm broad, above tending to form perpendicular rows of cells which gradually elongate and become club shaped, 20-30 x 3-5 µm; outermost cells with vinaceous content. Subhymenium indistinct.

Hymenium 100-120 µm high. Asci normally 4-spored, strongly protruding when mature, then up to 140 µm long, 20-25 µm broad, below gradually tapering into a slender base, above truncate, all over staining pale bluish in Melzer's reagent. Operculum 6-9 µm broad.

Paraphyses straight, above slightly enlarged to 6-8 µm broad, branching, septate, of short cells, sometimes anastomozing, hyaline or with very pale vinaceous content in uppermost cells.

Spores strongly adhering in clusters, slightly overlapping, in one row, or in two displaced parallel rows. Individual spores 16.5-18.5 x 8.5-9.5 µm, ellipsoid to inequilateral, at first hyaline, then purplish, finally purplish brown, pigmented layer thin, with irregular crackings on the free convex side like *Saccobolus glaber*.

Spore clusters 32-40 x 14-16.5 µm (when displaced in two rows) or 36-42 x 12-14 µm (when displaced in one row), with one continuous, prominent, unilateral gelatinous covering.

Material examined: Type - Greenland, 2 km W of Mestersvig, near Nyhavn at Kong Oscars Fjord, 72°12'N 24°10'W, on dung of *Branta leucopsis* (Barnacle Goose) in moist area with seeping water, 3 August 1983, H. Dissing & S. Sivertsen, Gr. 83.95 (C, Holotype); East Greenland, Mestersvig; Gåsesøen near the airport, on dung of *Branta leucopsis*, 9

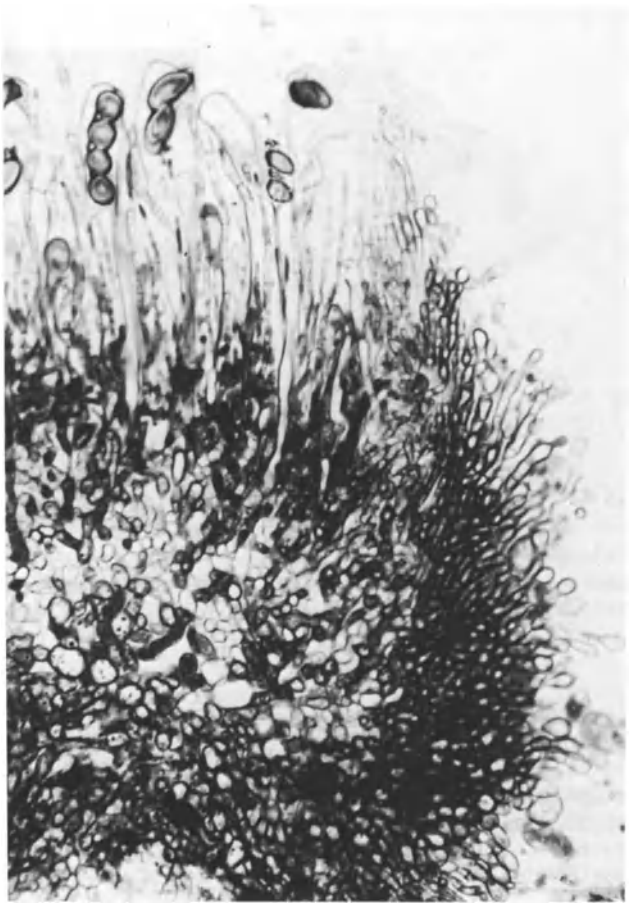

Fig. 5. *Saccobolus groenlandicus*, section of fruitbody, EI. 84.116, x 450.

August 1982, Gr. 82.137; - Store Blydal, 10 km SW of Mestersvig, on dung of goose (undetermined), 27 July 1983. Incubated 6 January 1984, isolated 24 January 1984, Gr. 83.142.

In addition more than 30 collections from Nyhavn, Gåsesøen and Store Blydal around Mestersvig have been studied. Most of these are deposited in the Botanical Museum, Copenhagen (C), but supplementary collections are deposited in Herbaria at the Universities of Trondheim (TRH) and Leiden (L).

East Greenland, Danmarks Havn; near Hvalrosodden, fresh dung from Barnacle Goose, collected 10 August 1984, leg. Børge Lauritsen. Incubated 19 January 1985, isolated 19 February 1985, Gr. 85.10. Canada, Ellesmere Island; Alexandra Fjord, 17 July 1984, EI.84.12, 84.13; - ibid., 21 July 1984, EI.84.19, 84.20; - ibid., 1 August 1984, EI.84.79, 84.85; - fresh dung from goose, collected 27 July 1984. Incubated 15 April 1985, isolated 1 May 1985, EI.84.114 and incubated 24 May 1985, isolated 20 June 1985, EI.84.116.

DISCUSSION

The above description is based on notes from fresh material of the

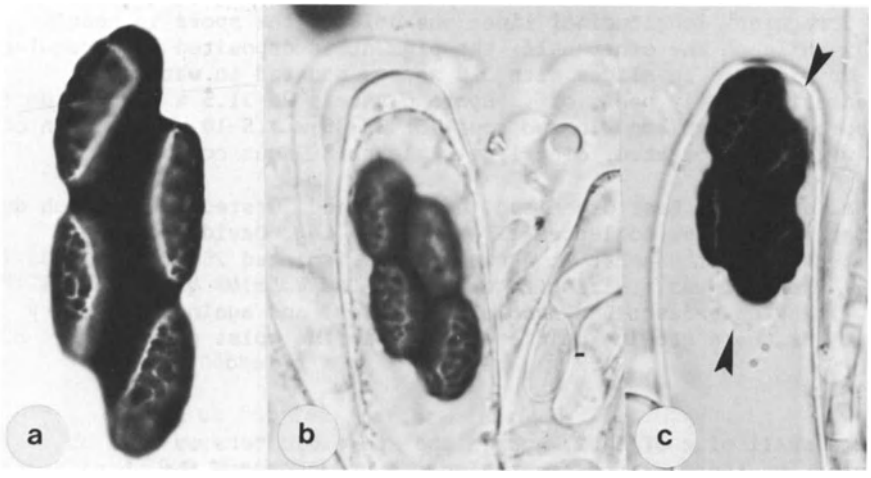

Fig. 6. LM photographs of spore clusters, a. *Saccobolus quadrisporus*, Gr. 82.327, b. *Saccobolus* sp. nov., note the curved paraphyses and bean-shaped immature spores, Gr. 82.326, c. *Saccobolus* sp. nov., ascus with spores with two separated gelatinous coverings, arrows, Gr. 82.326. a-c. x 895.

type as well as notes from material developed in moist chambers in the laboratory in Copenhagen. In nature, old apothecia will often be dark brownish, with amorphous substances deposited between cells in the excipulum and among paraphyses in the hymenium.

In *Saccobolus groenlandicus*, the asci are normally 4-spored, with the spores displaced in two rows. However, a few asci with spores in one row will be seen in almost all ascocarps. Asci with 3 or 5 spores were rarely found, and on two occasions only, asci with 8 spores in a cluster were seen (Hare dung, Gr. 83.127 and Goose dung, EI. 84.85).

Saccobolus groenlandicus can easily be distinguished from *S. quadrisporus* on characters of the size of asci, single spores and spore clusters. Besides colors of the ascocarps, characters of the spore ornamentation and gelatinous covering separate the two species.

In East Greenland, *S. groenlandicus* has been found on dung of Barnacle Goose (*Branta leucopsis*), Arctic Hare (*Lepus arcticus*) and Lemming (*Lemmus lemmus*). No collection has yet been made on dung from Pink-footed Goose (*Anser brachyrrhynchus*), which also nests there. In Ellesmere Island the goose dung most probably stems from Snow Goose (*Anser coerulescens*) or Brent Goose (*Branta bernicla*). It would certainly be interesting to study winter dung from the above mentioned geese in their respective winter quarters.

Saccobolus sp. nov. Figs. 2b and 6b.

Ascocarps 0.2-0.5 mm broad, disc-shaped to pulvinate, pale purplish to vinaceous or lilac. Asci 4-spored, 115-122 x 13-16 µm, staining pale bluish all over in Melzer's reagent. Paraphyses septate, branching, above curved, slightly enlarged to 5-6.5 µm broad. Spores 12-13.5 x 7-9.5 µm, ellipsoid to bean-shaped, at first hyaline, smooth, when mature reddish brown; in mature spores the pigment is deposited in two zones, separated

by an irregular, longitudinal line: one half of the spore is nearly smooth, while on the other half, the pigment is deposited on irregular warts or ridges. In slides with old spores mounted in water the ornamentation easily peels off. Spore clusters 26-31.5 x 12.5-14 µm (when the spores are displaced in two rows) or 33-35 x 7.5-10 µm (when in one row), with two separated, clearly visible gelatinous coverings.

Material examined: East Greenland, Jameson Land, Ørsteddalen, fresh dung from Barnacle Goose, collected 17 July 1982, leg. David Boertmann. Incubated in moist chamber 11 January 1983, isolated 25 January 1983 (Gr. 82.326) where it was fruiting together with *Saccobolus quadrisporus* (82.327) and an undescribed *Ascobolus* species, and again 16 February 1983 it was where growing on filter paper in the moist chamber, Gr. 82.341.

DISCUSSION

The small size of asci, spores and spore clusters as well as characters of the spore ornamentation seem to separate the above described taxon very well from *Saccobolus quadrisporus* and *S. groenlandicus*. However, a formal description of a new species is postponed until material from nature is available.

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Joop van Brummelen made pertinent suggestions on the correct delimitation of *Saccobolus quadrisporus*. Tyge Christensen prepared the Latin diagnosis. Jørgen Fuglsang Nielsen operated the Cambridge electron microscope. Lene Christiansen and Lisbeth Haukrogh gave valuable technical assistance. Doris Thye-Petersen typed the manuscript.

Sigmund Sivertsen (1982), Henrik Gøtzsche (1983), and Linda M. Kohn (1984) were brilliant co-workers during the field work.

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