Ascobolus lineolatus Brumm. DPM AEB 1378 (= PDD 121670) – a good match

<u>Collection site</u>: Kāpiti Island nature reserve, map courtesy of Ian Flux (The red loop encompasses the weka dung collection area, on the ridge crest at this site.) Mixed regenerating forest of senescent manuka/kanuka.



Collection date: 10 June 2024 Substrate: weka (*Gallirallus australis*) dung Collector: Ian Flux Identifiers: Ann Bell & Dan Mahoney Voucher materials: dried herbarium material DPM AEB 1378 (= PDD 121670) accompanied by two Shear's mounting fluid (SMF) semipermanent slide mounts; several in-situ dissecting scope photos of apothecia on the dung and several photos of microscopic detail from water and SMF mounts.

Dan's brief comments: The closely matching mature violet ellipsoid ascospores $(13-14 \times 8-9 \mu m)$ ornamented with delicate, closely-spaced transverse to oblique, parallel to subparallel lines that often anastomose make this an easily identifiable species. Apothecia were numerous, highly fertile and variously sized – closely matching Brummelin's 1967 description (see the next page for his description and illustrations). In the present collection, his comments RE 'greenish', when describing the appearance of the apothecium, were more yellowish than greenish.

This collection represents the first report of *A. lineolatus* at PDD and its first record worldwide on weka dung. However, Ann, in her 1983 publication 'Bell, A. 1983. Dung fungi: an illustrated guide to coprophilous fungi in New Zealand. Victoria University Press, Wellington, New Zealand. 88 pp.' provides an earlier NZ record (see her pages 30 and 66).

Brummelen J. (van). 1967. A World-monograph of the genera Ascobolus and Saccobolus (Ascomycetes, Pezizales). Persoonia, supplement, I: 1–260. Pages 120–121.

120 A WORLD-MONOGRAPH OF THE GENERA ASCOBOLUS AND SACCOBOLUS

22. Ascobolus lineolatus Brumm., spec. nov.-Fig. 30; Pl. 9, figs. A-F

Apothecia sessilia, usque ad 1 mm diam. Receptaculum initio subglobulare, deinde hemisphaericum, denique paullo applanatum, virescens, granulatum, margine interdum crenulato. Excipulum textura globulosa. Asci cylindrico-clavati, 130–150 × 15–18 μ , 8-spori, pariete iodo haud caerulescente. Ascosporae ellipsoideae, primum hyalinae, tum violascentes, 13–14-5 × 7.5–8.5 μ , lineis delicatissimis, parum inter sese distantibus, transversis vel obliquis, subparallelis, iterum et iterum anastomosantibus ornatae. Paraphyses ramosae, cylindricae, 2–2.5 μ crassae. — Ab Ascobolo crenulato imprimis ascospororum sculptura diversus. Fimum muris ratti incolit. Typus: van Brummelen 1981 (L).

Apothecia scattered, superficial, sessile, up to 1.0 mm across, 0.4-0.5 mm high. Receptacle at first subglobular and closed, then opening and hemispherical with an obconical base, finally slightly flattened, greenish, covered with small, isolated, white granules, with a narrow, sometimes slightly crenulate margin. Disk concave, then flat, greenish, roughened by the protruding tips of ripe asci. Hymenium about 130 μ thick. Hypothecium 15–23 μ thick, of subglobular cells 4–7 μ diameter. Flesh 140-200 μ thick, of subglobular, angular or oblong cells 7-42 \times 6-23 μ , hyaline. Excipulum near the margin $28-35 \mu$ thick, near the base up to 75μ thick, of subglobular or elongated cells $8-35 \times 8-23 \mu$ (textura globulosa), at the base mainly consisting of a layer of closely compacted, intertwined, cylindrical, septate hyphae 3.5-6 μ wide, pale yellowish-brown, covered with small isolated groups of globular cells. Asci cylindric-clavate, tapering downwards, rounded above, 130-150 \times 15-18 μ , 8-spored; the wall not blue in Melzer's reagent. Ascospores biseriate; ellipsoid; at first hyaline, then violet; $13-14.5 \times 7.5-8.5 \mu$; ornamented with very delicate, closely spaced, transverse or oblique, subparallel lines that anastomose repeatedly; pigment very thin, about 0.3 μ thick. Paraphyses branched, septate, cylindrical, 2.0-2.5 µ thick, not or scarcely enlarged at the tip, embedded in greenish mucus.

Only known from dung of rat.

ETYMOLOGY.-From Latin, lineolatus, marked with fine or obscure lines.

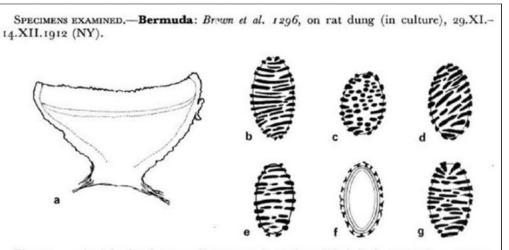


Fig. 30. — Ascobolus lineolatus: a, diagrammatic section of fruit-body \times 50; b-e, g, ascospores \times 1600; f, ascospore in optical section \times 1600 (different layers of primary spore wall, staining differently with trypan blue). (a-c, from *Brown et al.* 1296; d-g, from type.

ASCOBOLUS

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North Borneo: van Brummelen 1981, on dung of rat (comm. Dr. Ding Hou), Mesilau Camp (alt. 1500 m). Mt. Kinabalu, 4.VII.1966 (type of A. lineolatus, also subcultured on sterilized horse dung, L).

Of this species fine cultures from North Borneo could be studied. It is only known from rat dung of two very remote localities. It is related to *A. crenulatus*, from which it differs in the smaller apothecia and the ornamentation of the episporium.

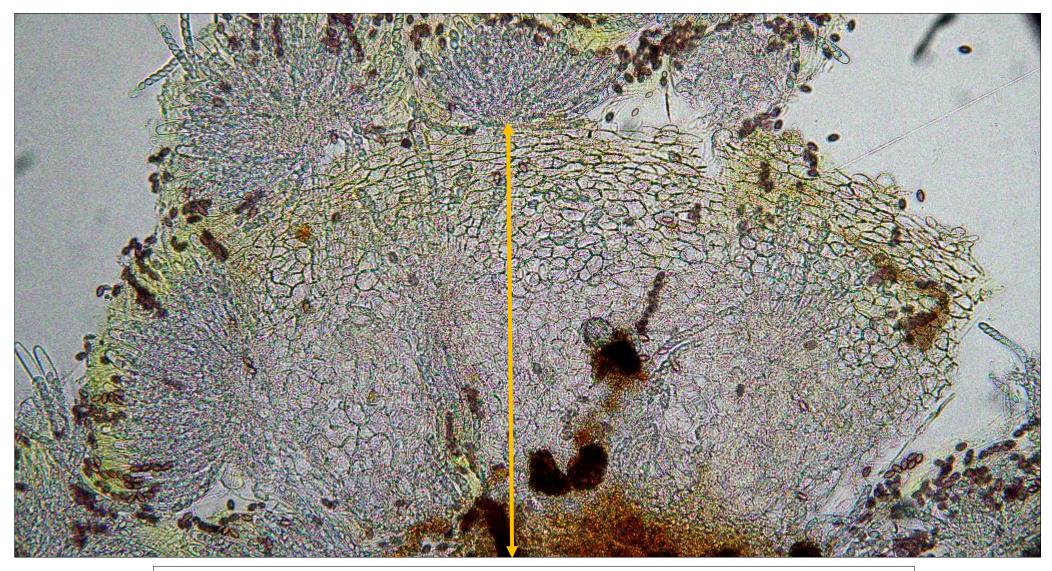
The episporium of A. lineolatus consists of a pattern of transverse or oblique, subparallel crevices that anastomose repeatedly. The crevices, however, are broad and the ribbons of pigment relatively narrow. Therefore the episporium may look like subparallel rows of short ribbons of pigment. These ribbons are T-shaped in transverse section and rest with their narrow side on the primary spore-wall (Fig. 30f).



DPM AEB 1378. In-situ fertile apothecia of various sizes on weka dung in a moisture chamber. Photo 19 June 2024.



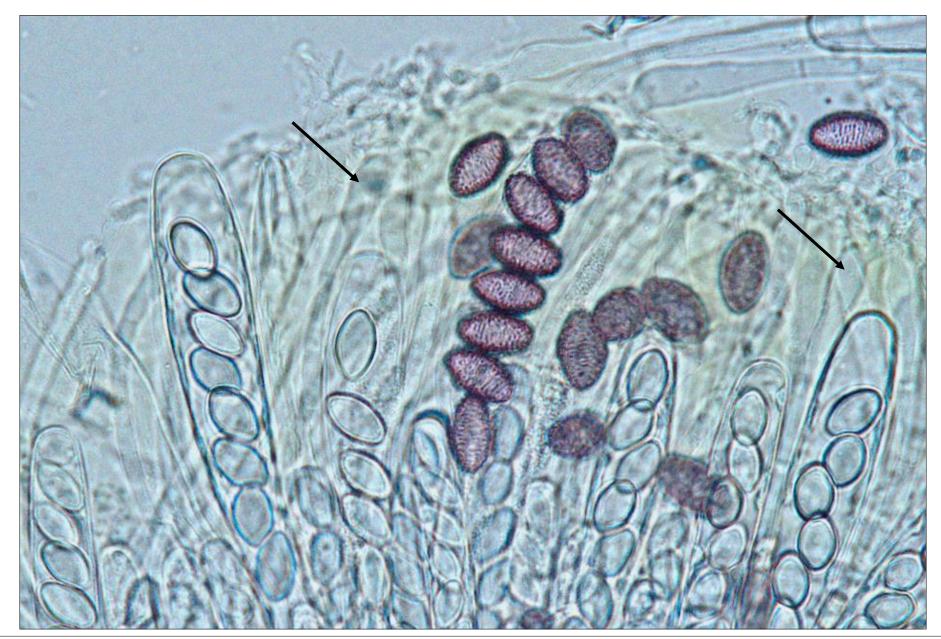
DPM AEB 1378. Closeup of in-situ fertile apothecia from the previous page.



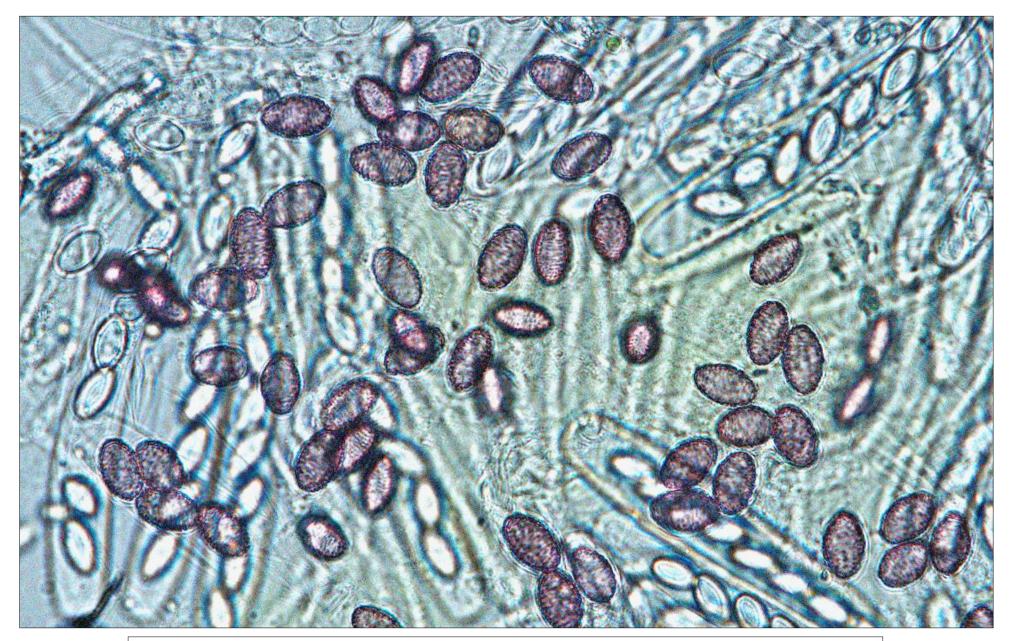
DPM AEB 1378. Apothecium ectal excipulum in a water mount squash using the X20 objective & brightfield microscopy. Note the various ectal excipulum cell shapes from near the apothecium base to its apical rim (arrowed). Photo 19 June 2024.



DPM AEB 1378. Asci and ascospores in a SMF squash mount near the yellowish apothecial hymenium surface using the X40 objective (enlarged view) and brightfield microscopy. Note the ascospore color, shape & ornamentation. Photo 21 June 2024.



DPM AEB 1378. Immature and mature asci and ascospores in a water mount squash using the X100 objective (view enlarged) & brightfield microscopy. Note the faint yellowish-green color at the apothecial hymenium surface and the septate paraphyses with their slightly enlarged tips (arrowed). Photo 19 June 2024.



DPM AEB 1378. Immature and mature asci and ascospores in a water mount squash using the X100 objective (view enlarged) & brightfield microscopy. Emphasis here is on ascospore color, shape and especially ornamentation. Photo 19 June 2024.