PERSOONIA
Published by the Rijksherbarium, Leiden
Volume 5, Part 2, pp. 189-199 (1968)

NEW SPECIES OF THYSANOPHORA AND CUSTINGOPHORA GEN. NOV.

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(With Plate 9 and four Text-figures)

Two monoverticillate species of *Thysanophora* are described and figured, *T. canadensis* spec. nov. and *T. taxi* (Schneider) comb. nov. A new hyphomycetous genus, *Custingophora*, related to *Thysanophora* and *Phialocephala*, is proposed with *C. olivacea* spec. nov. as type species.

The genus *Thysanophora* was erected by Kendrick (1961a), who included two species, *T. penicillioides* (Roum.) Kendrick, the type species, and *T. longispora* Kendrick. Both species are characterized by brown-coloured sympodially branched conidiophores, bearing biverticillate penicilli resembling those of *Penicillium*. In this paper two species with monoverticillate penicilli are added to the genus.

The third fungus dealt with in this paper resembles superficially an *Aspergillus* but shows some significant differences from this genus. Ontogenetically it rather resembles a monoverticillate *Penicillium* but has slimy spores, just like *Phialocephala* Kendrick (1961b) resembles a polyverticillate *Penicillium*. Since the fungus cannot be satisfactorily placed in any existing genus of the Moniliales either, a new genus is proposed for it, *Custingophora*.

Thysanophora canadensis Stolk & Hennebert,

spec. nov.—Text-fig. 1, Pl. 9, figs. 1–4

Fungus imperfectus. Coloniae in natura inconspicuae, e conidiophoris atro-brunneis, conidiis griseo-viridibus, hyphis repentibus et sclerotiis constistentes, in vitro variabiliter crescentes, centro zonatae, velutinae, griseo-virides vel olivaceae, fertiles, margine sparsae et pallidiores, sclerotia formantes, facie reversa atro-brunnea. Mycelium e hyphis immersis hyalinis vel subhyalinis 1.5–3 μ latis, hyphisque aeriis in natura atro-brunneis prostratis reticulatis in vitro subhyalinis lanosis formatum. Conidiophora mononemata, solitaria vel adjuncta, in hyphis immersis vel aeriis enata, erecta, simplicia et successive proliferantia. Stipites cylindracei, 150–1000 μ longi, 4.5–6.5 μ lati, septati, atro-brunnei, basi parce vel haud inflati, cellulis duabus distalibus hyalinis vel subhyalinis usque ad 2–2.5 μ attenuatis, apice abrupte usque ad 4.5–6 μ incrassatis, pariete laevi. Proliferationes singulae laterales e cellula subdistali formatae, sympodiales, parce vel haud septatae, 15–30 μ longae, 2.5–3 μ latae, fertiles. Penicilli verticillo unico phialidum 3–8 instructi. Phialides hyalinae, parallelae

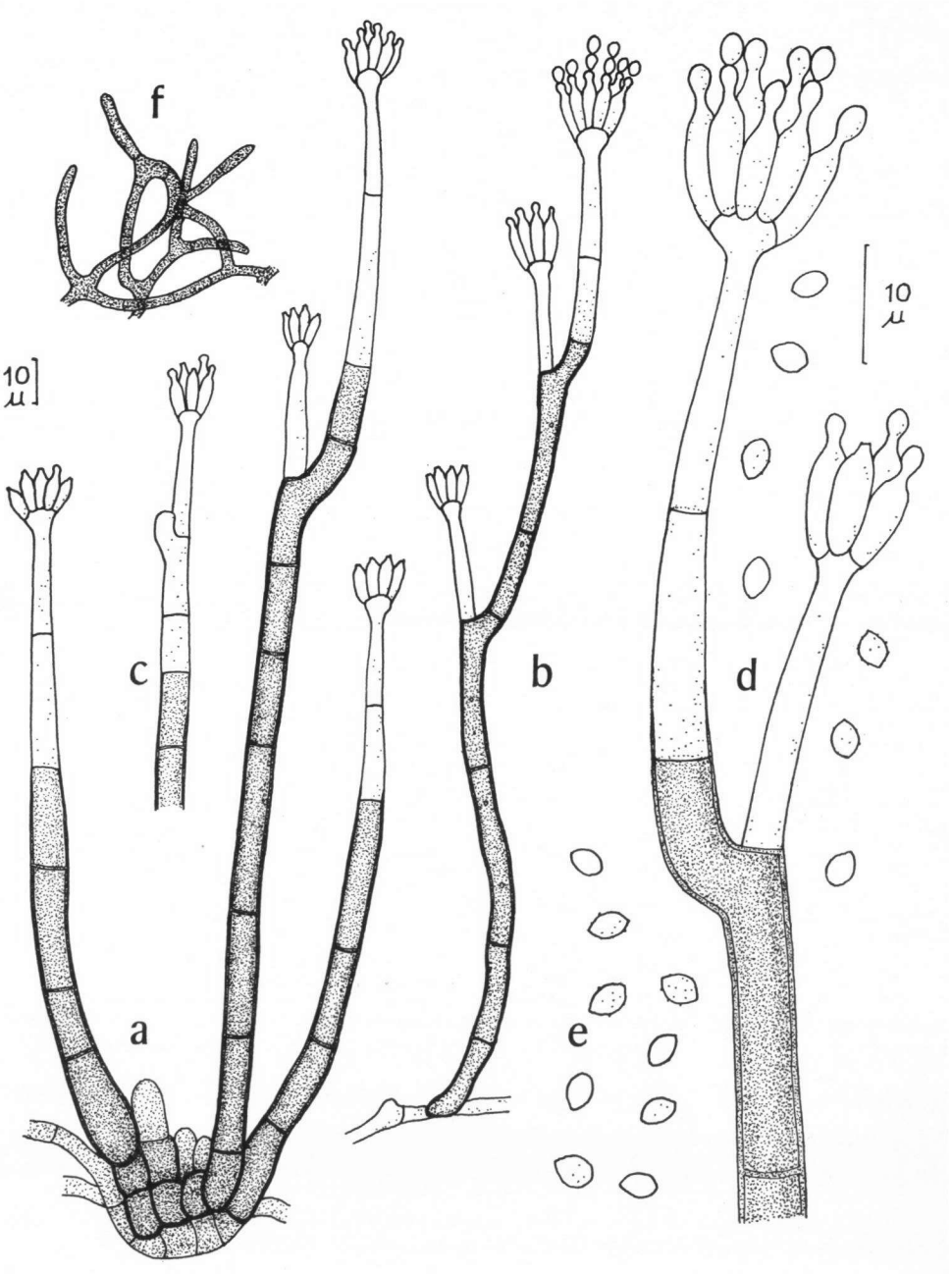


Fig. 1

parum divaricatae, cylindraceae, tubulares, apice in collum curtum 1 μ diam. attenuatae, collario nullo, 7.5–12 × 2.5–4 μ. Phialosporae continuae, siccae, catenas basipetales formantes, postea ovoideae, apiculatae, verruculosae, 2–3.5 × 1.7–2.5 μ. Sclerotia 250–400 μ diam.; medulla e cellulis hyalinis crasso-tunicatis formata; cortex cellulis brunneis instructus, hyphisque brunneis anastomosantibus intertextis 2–4 μ latis setisque radiantibus sterilibus tectus.

Typus in aciebus *Tsugae canadensis* repertus, Bell's Corners, Ontario, Canada, Sept. 1961, *G. L. Hennebert*, et in culturis desiccatis et vivis conservatus (G.L.H. 2497-B, CBS 334.68).

The first subcultures made from the fresh material, preserved as dried cultures (Plate 9, fig. 1), grew rapidly on both malt agar and potato dextrose agar. They were zonate, producing abundant conidial structures in the centre of the colonies, but more scarcely in the external zone, while this zone was drying out. Sclerotia were produced in a more or less radiate arrangement, at first appearing whitish, but later becoming surrounded by brown shining hyphae and then appearing dark brown.

After 7 years of preservation under mineral oil the cultural appearance has changed considerably (Pl. 9, fig. 2). Colonies on malt agar grow slowly, attaining a diameter of 4 cm within two weeks at 25° C. They consist of a well-developed, largely submerged mycelium, upon which abundant conidial structures are produced, forming a thin, velutinous, slightly zonate layer, pale greyish green coloured near Tea Green (Ridgway, Pl. 47) when young, but soon turning more greyish approximating Deep Olive-Gray (Ridgway, Pl. 51). Colony margins consisting of submerged hyphae often show dark green colours. Sclerotia are not produced any more. Reverses of colonies are zonate, showing greenish black shades near Ivy Green and Olivaceous Black (Ridgway, Pls. 31, 46).

Colonies on oatmeal agar grow more slowly, about 2 cm within two weeks at 25° C. They are very thin, greyish brown, approximating Grayish Olive (Ridgway, Pl. 46). Sclerotia are not produced. Reverses show grey to brownish shades.

Colonies on Czapek agar grow very restrictedly, about 1 cm within 14 days at 25° C, and are sterile.

In culture the mycelium is hyaline to subhyaline, septate, 1.5–3 μ in diameter. Production of conidial structures is most pronounced on malt agar. Conidiophores occur singly, erect or ascending, arising mostly as branches from submerged hyphae but occasionally, especially in the centre of the colonies, also from aerial hyphae. Stipes are septate, olivaceous when young, brown-coloured in age, but paler toward the apex, with the upper two cells hyaline or subhyaline; they are somewhat sinuous, unbranched or sometimes sympodially branched by successive single subapical proliferations of the penultimate cell of the stipe, and consequently geniculate. They are variable in length, ranging from 150–1000 μ, with a diameter of 4.5–6.5 μ, uniform over almost their entire length, but tapering very gradually in the upper two cells to a diameter of 2–2.5 μ, enlarging again abruptly to about 4.5–6 μ diameter at the apex. Their walls are smooth, about 0.5 μ thick, becoming thinner toward the apex. Distal cells may become lateral, following the proliferation of the penultimate cell of the stipe, and then appear like branches (metulae), 15–30 ×

EXPLANATION OF FIGURE 1

Text-fig. 1. *Thysanophora canadensis*, CBS 334.68 — a. Group of conidiophores emerging from the host. — b. Conidiophore in culture. — c. Development of a proliferation of the penultimate cell of the stipe (culture). — d. Upper part of the conidiophore with monoverticillate penicilli (culture). — e. Phialospores (culture). — f. Reticulum of hyphae surrounding the sclerotium (host).

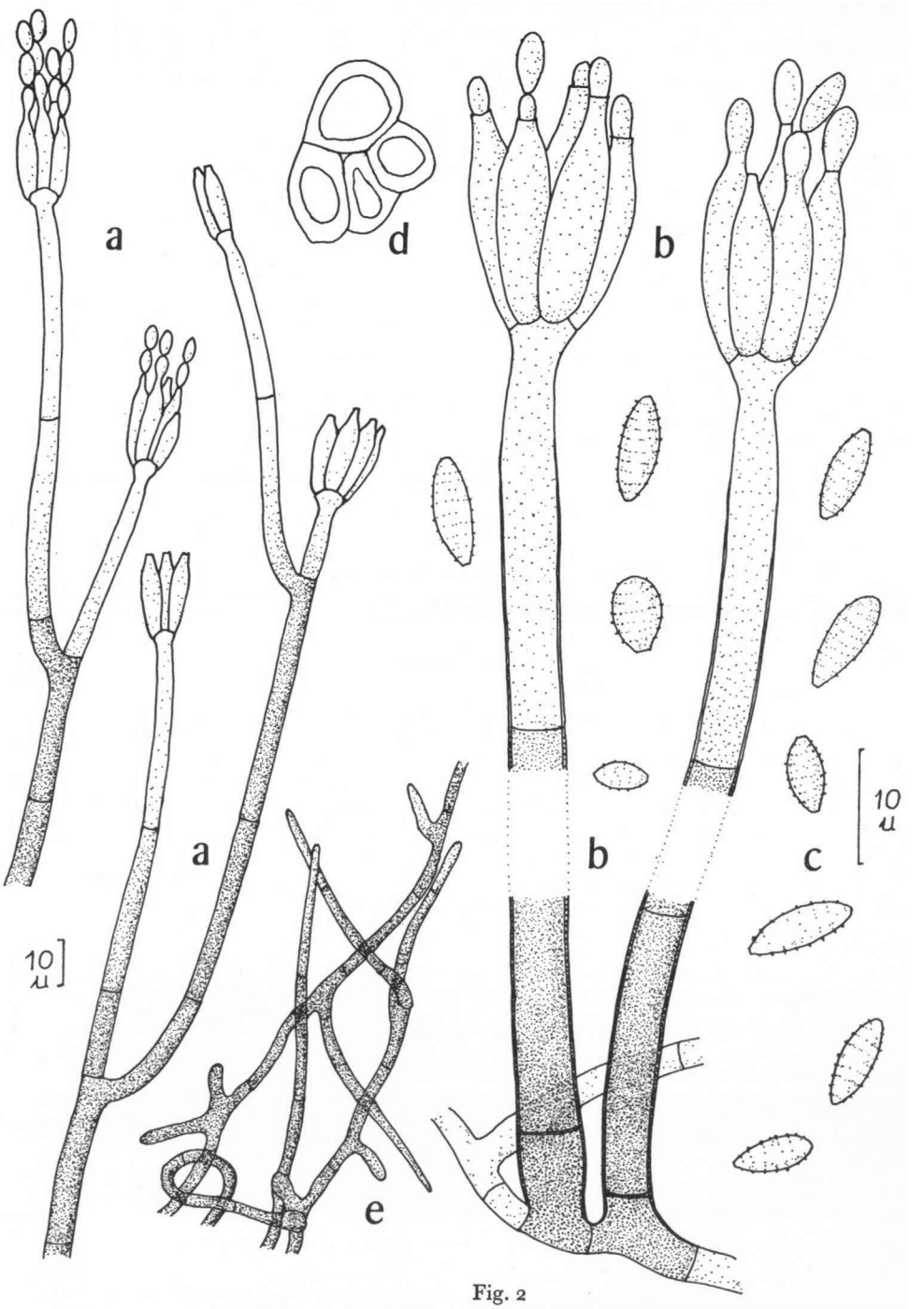


Fig. 2

2.5–3 μ. Proliferations are very variable in length, reaching 100 μ, 1–4-septate, concolorous with the stipe; up to 3 or 4 proliferations may develop successively on one stipe. Penicilli consist of a verticil of 3–8 phialides, borne in a parallel or slightly divergent cluster at the apex of the stipe. Phialides, measuring 7.5–12 × 2.5–4 μ, are concolorous with the distal cell of the stipe, flask-shaped, cylindrical at the base, tapering abruptly to a small narrow neck of about 1 μ diameter; a collarette is lacking. Phialospores are borne in basipetal chains, they are dry, continuous, subhyaline, globose to subglobose at first, soon becoming ovoid to broadly ellipsoid, slightly apiculate at both ends, with walls smooth or very finely roughened, 2–3.5 × 1.7–2.5 μ. Sclerotia are white when young, becoming dark brown in age, occurring singly or in small clusters, about 250–400 μ in diameter, consisting of a medulla of thick-walled hyaline cells and a cortex of one layer of similar brownish cells; they are surrounded by a dense reticulum of brown, septate, abundantly branching and anastomosing hyphae 2–4 μ in diameter emerging from the cortex. Toward the outside these hyphae become more loosely woven, producing sterile seta-like projections at the periphery.

On the host the mycelium is partly immersed in the tissues and hyaline, partly creeping on the surface, thick-walled, dark brown, sinuous, branched, and often anastomosed, forming a reticulum. Conidiophores are erect, either emerging in groups of 6–16 from small thick-walled, dark brown, roundish cells filling the stomata, or ascending solitarily from creeping hyphae. Conidiophores are 120–350 μ in length by 6–9 μ in diameter, being longer and slightly thinner in culture. The conidial heads and conidia in culture look like those developing on the host.

The holotype is represented by natural specimen, dried and living cultures from needles of *Tsuga canadensis*, Bell's Corners, Ontario, Canada, 10 Sept. 1961, *G. L. Hennebert*, (G. L. H. 2497-B, CBS 334.68).

Conidial structures of this new species were detected on the host amongst the much larger structures of *T. penicillioides*. The two species are closely related, *T. canadensis* differing from *T. penicillioides* in producing monoverticillate conidial heads, whereas those of *T. penicillioides* are biverticillate. Moreover there is a slight difference in size and shape of the phialospores, those of *T. canadensis* being less ellipsoid than those of *T. penicillioides* (1.8–6.7 × 1.4–4 μ). *Thysanophora canadensis* differs from the monoverticillate species *T. taxi* by the much smaller phialides and phialospores.

Thysanophora taxi (Schneider) Stolk & Hennebert,

comb. nov.—Text-fig. 2, Pl. 9, figs. 5, 6

Penicillium taxi Schneider *in* Zentbl. Bakt. ParasitKde, II Abt., **110**: 47. 1956 (basionym).

Colonies on malt agar grow restrictedly, attaining a diameter of about 2.5 cm within two weeks at 25° C. They consist of a well-developed mycelium, but largely submerged, upon which abundant conidial structures are borne. Colonies are

EXPLANATION OF FIGURE 2

Text-fig. 2. *Thysanophora taxi*, CBS 206.57. — a. Branched conidiophores. — b. Conidiophores showing monoverticillate penicilli and foot cells. — c. Phialospores. — d. Thick-walled cells of the sclerotium. — e. Reticulum of hyphae surrounding the sclerotium.

velutinous, plane, showing dark olivaceous green colours near Lincoln Green (Ridgway, Pl. 41), becoming more grey-green near Grayish Olive (Ridgway, Pl. 46) in age. Sclerotia, produced in limited number, are white when young. Reverses of colonies show dark green to almost black shades, approximating Olivaceous Black (Ridgway, Pl. 51).

Colonies on oatmeal agar grow about 1.2 cm within two weeks at 25° C, they are thinner than those on malt agar, producing somewhat more vegetative mycelium and sporulating less abundantly, and show olivaceous green shades near Grayish Olive, becoming brownish grey near Drab and Hair Brown in age (Ridgway, Pl. 46). Sclerotia are produced in limited numbers, and become black soon. Reverses of colonies show brown colours.

Colonies on Czapek agar grow very restrictedly, about 1 cm within 14 days at 25° C, producing only limited conidial structures.

The mycelium is hyaline to subhyaline or brownish, with the hyphae 2–4 μ in diameter. Conidiophores arise singly as branches from submerged or aerial hyphae. Stipes are erect or ascending, septate, olivaceous when young, brown-coloured in age, mostly paler toward the apex. They are somewhat sinuous, unbranched or sometimes branched by successive, single, subapical proliferations of the penultimate cells of the stipes, occasionally of one of the lower cells, and consequently becoming geniculate. They are variable in length, ranging from 100–1000 μ in length, and 4–6 μ in diameter, tapering in the upper cell to a constriction beneath the apex of 3–4 μ diameter, abruptly enlarging again at the apex to a diameter of 6–7 μ. Their walls are smooth, up to 0.5 μ thick, becoming thinner toward the apex. Distal cells may become lateral following proliferation of cells of the stipe, then appearing like branches. These branch-like structures are subhyaline or brownish, of variable length, the aseptate ones mostly about 40 μ long, but up to 100 μ when 1- or 2-septate. Proliferations are usually concolorous with the stipe or slightly paler, 1–4-septate, very variable in length, up to 200 μ. Penicilli consist of one verticil of phialides, which are mostly parallel-ranged. Phialides are subhyaline to pale olivaceous when young, mostly darkening in age, usually concolorous with the distal cell of the stipe. They are smooth-walled, 17–24 × 5–7 μ, cylindrical at the base, tapering abruptly to a small narrowed neck of about 1.5 μ diameter; a collarette is lacking. Phialospores are continuous, dry, subhyaline to slightly olivaceous, ovoid to ellipsoid, showing scars at each end, slightly roughened, with the roughenings often arranged in parallel bands, 6.5–10 × 2–3.5 μ, forming very long, parallel to slightly divergent chains, appearing silky when viewed under low magnifications. Sclerotia are white when young, becoming dark brown to almost black in age, occurring singly or in clusters, 300–600 μ in diameter, very hard, consisting of a medulla of thick-walled hyaline cells and a cortex of similar brownish cells. They are surrounded by a reticulum of brown, septate, abundantly branching and anastomosing hyphae 2–2.5 μ in diameter, producing sterile seta-like projections at the periphery of up to 40 μ long.

CBS 206.57, representing the holotype, was isolated by Dr. R. Schneider from needles of *Taxus baccata* in March 1953 and sent to the CBS as strain 74.80 in 1957.

Dr. Schneider reported the production of abundant sclerotia on malt agar, developing in concentric zones. After having been cultivated for 15 years, the production of sclerotia is very much reduced.

The penicillate structures producing long conidial chains as well as the colour of the colonies of *T. taxi* suggest a *Penicillium*. However, this fungus clearly belongs in *Thysanophora* because of its brown-coloured sympodially branched conidiophores. In

addition the structure of the sclerotia is identical with that occurring in *Thysanophora*. The species is closely related to *T. longispora*, since both produce large phialides and large spores, but differs from it in producing monoverticillate penicilli. The shape and size of the phialospores (*T. longispora*: 8–16.2 × 1.8–3 μ) and the cultural appearance of the two species are also different. The branching of the conidiophores is more irregular in *T. taxi* than in other species of the genus in that proliferations develop from lower cells of the stipe.

Custingophora Stolk, Hennebert & Klopotek, *gen. nov.*[1]

Deuteromycetes, Moniliales.

Hyphae hyalinae vel fuliginosae, ramosae, septatae. Conidiophora mononemata, simplicia vel sympodialiter proliferantia, erecta, fuliginea, septata, basi saepe radicibus munita, apice in vesiculam inflata. Phialides uniseriatae, conidiophori apice successive enatae, brunneae. Phialosporae mucosae, continuae.

Species typica: *C. olivacea* Stolk, Hennebert & Klopotek.

Hyphae hyaline or coloured, branched, septate. Conidiophores mononematous, simple or sympodially branched by subapical proliferation, erect or ascending, brown, septate, arising from foot cells which may produce rhizoids, enlarging apically to form a more or less distinct vesicle. Phialides produced successively on the apex of the conidiophore or vesicle, brown. Phialospores continuous, collecting at the apex of the conidial head in drops of slime.

Type species: *C. olivacea* Stolk, Hennebert & Klopotek.

The new genus shows similarities with the genera *Aspergillus* Micheli ex Fr., *Thysanophora* Kendrick, and *Phialocephala* Kendrick (1961b).

The conidial heads of *Custingophora* resemble superficially those of *Aspergillus*, yet the two genera differ in some important characteristics. In *Aspergillus* the development of the conidial head starts with the production of an apical swelling of the conidiophore to form the vesicle, then phialides develop simultaneously on this vesicle. In *Custingophora*, on the other hand, phialides develop successively on the apex of the conidiophores, which enlarges gradually to accommodate the increasing number of phialides until finally a vesicle is formed (Text-fig. 4). In *Aspergillus* conidiophores are always simple, in *Custingophora* sympodially branched conidiophores occur. Moreover, the conidia in the new genus do not adhere in dry conidial chains, as is usual for most *Aspergillus* species, but they collect in conspicuous slime balls.

Before the apex of the stipe swells to produce the characteristic vesicle, the developing conidial heads of *Custingophora* resemble those of the genus *Thysanophora*. Moreover in both genera sympodially branched brown conidiophores occur, which develop by subapical proliferation of the stipe. However, the genera *Custingophora* and *Thysanophora* can easily be separated on the characters of the phialospores. In *Thysanophora* phialospores are dry amerospores forming conspicuous long chains, whereas in

[1] Etymology: κύστιγξ, small vesicle, φόρειν, to bear.

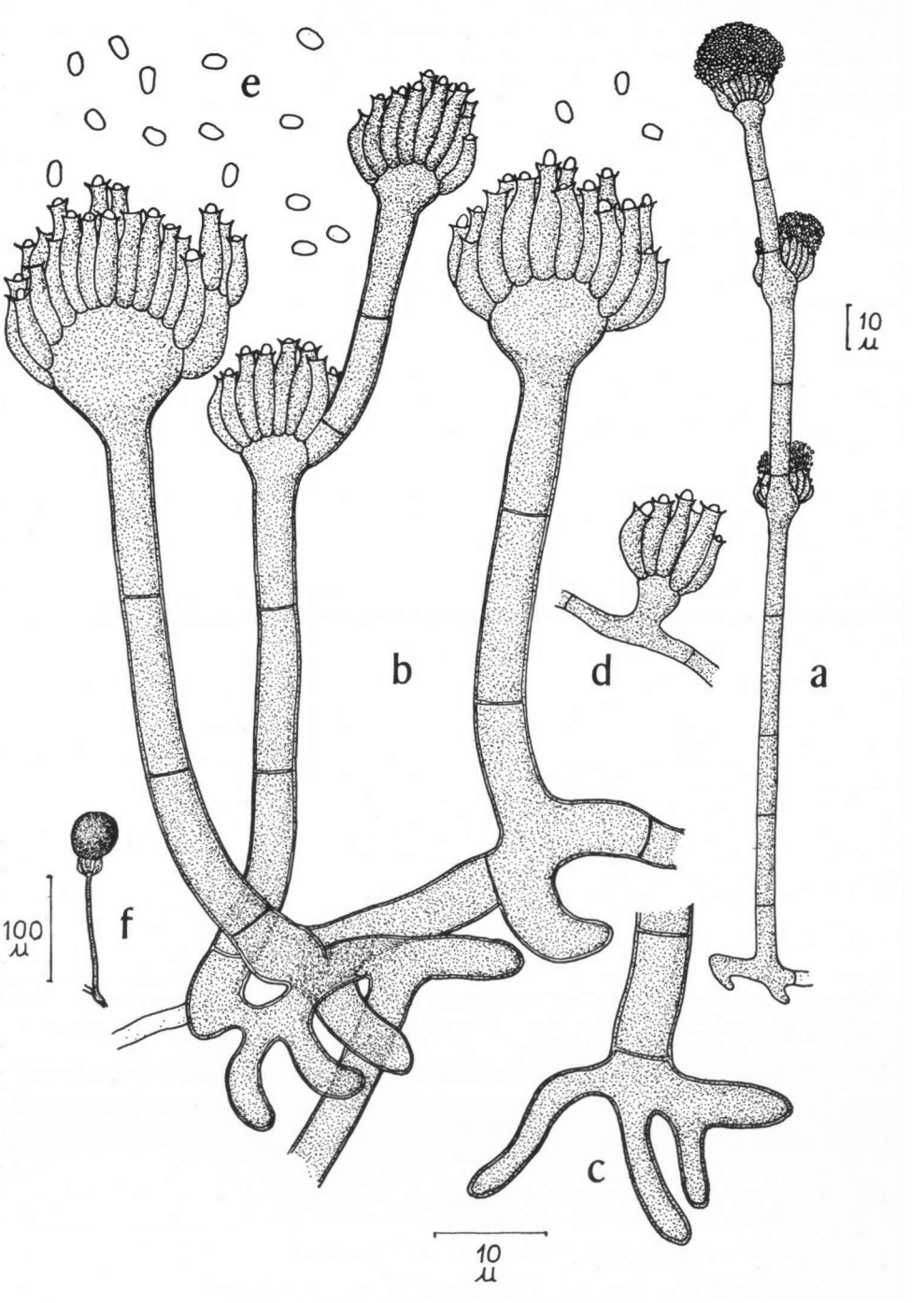


Fig. 3

Custingophora phialospores collect in slime balls. In addition the shape and the colour of the phialides are different in the two genera, those of *Thysanophora* resembling the phialides of *Penicillium*, whereas in *Custingophora* the phialides are similar to those of *Phialophora*.

The new genus has in common with *Phialocephala* the brown colour of the stipe, the phialides with well-marked collarettes and the phialospores collecting in drops of slime. However, they differ in the structure of the sporogenous head and in the branching of the conidiophore. In *Custingophora* the conidial head consists of one series of phialides covering the conspicuous swollen vesicle-like apex of the stipe, in *Phialocephala*, on the other hand, the sporogenous heads are complex structures, consisting of one to several series of metulae with the ultimate series bearing the phialides. Neither the apex of the stipe nor the apices of the phialide-bearing metulae form vesicle-like swellings. The stipe of *Custingophora* may become geniculate by successive proliferation, in *Phialocephala* no geniculate stipes occur.

Custingophora olivacea Stolk, Hennebert & Klopotek, *spec. nov.*—Text-figs. 3, 4, Pl. 9, figs. 7, 8

Fungus imperfectus. In vitro coloniae satis rapide crescentes, velutinae, olivaceo-brunneae, azonatae, margine alba, facie reversa olivacea vel atro-brunnea, saepe sectores pallidiores formantes. Mycelium e hyphis hyalinis vel subhyalinis 1.5–2.5 μ latis formatum. Conidiophora numerosa, erecta vel ascendentia, plerumque e hyphis submersis rarius aeriis formata, plerumque singula interdum pauca aggregata, in omnibus partibus uniformiter brunnea. Stipites septati, simplices vel sympodialiter proliferationibus succesivis simplicibus et subapicalibus ramosi, ideoque geniculati, e cellulis basalibus rhizoidibus conspicuis aseptatis digitaliformibus notatis enascentes, 20–200 μ longi, 2.5–5.5 μ diam., basi usque ad 7 μ diam., apicem versus attenuati, interdum constricti, vesicula distali, brunnea, subglobosa, 6–12 μ diam. Phialides successive enatae, uniseriatae, parallelae, externae incurvatae, internae rectae, laeves, cylindraceae vel botuliformes, brunneae, insuper constrictae collarioque conspicuo praeditae 6.5–10 × 2–2.5 μ. Phialosporae continuae, mucosae, ellipsoideae vel ovoideae, subhyalinae, laeves, 1.8–3 × 1–1.5 μ.

Typus in cultura desiccata et viva, e materiis vegetalibus putrescentibus, Germania, *A. Klopotek*, 1965 (CBS 335.68, G.L.H. 9398).

Colonies on malt agar grow fairly rapidly, attaining a diameter of about 9 cm within two weeks at 30° C. They are composed of a largely submerged vegetative mycelium bearing a very thin layer of conidial structures, which lend the colonies an olivaceous to brownish colour near Dark Grayish Olive (Ridgway, Pl. 46); they are azonate, showing a tendency to produce almost colourless sectors. The margin is broad and colourless. Reverses of colonies are olivaceous or dark brown.

Colonies on oatmeal agar in general agree with those on malt agar. Sporulation

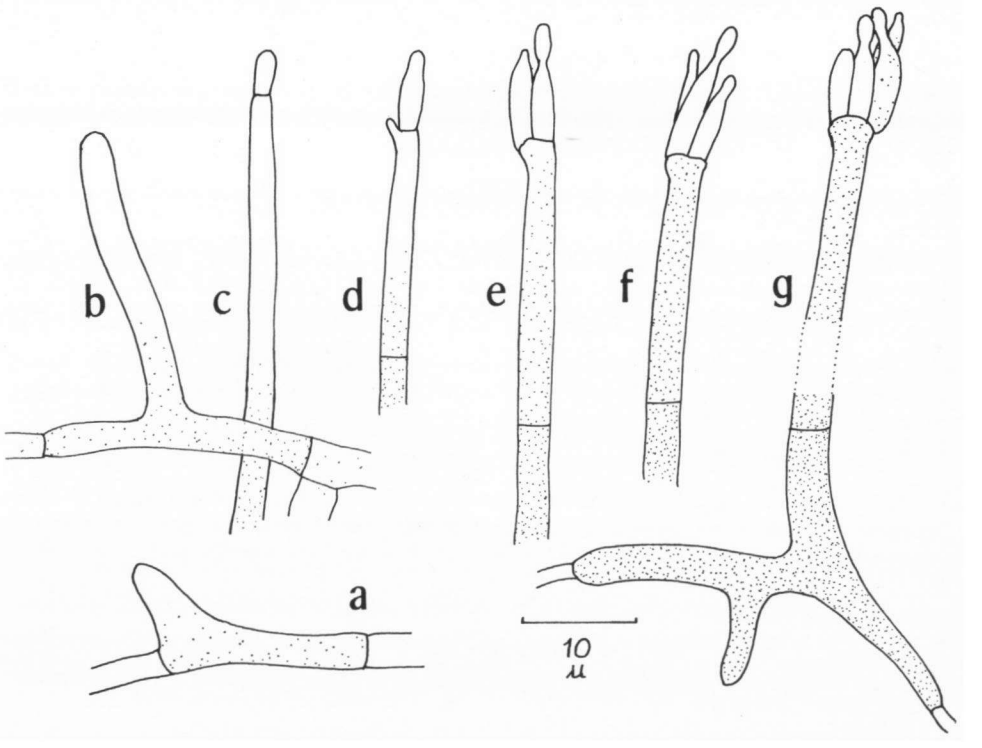
EXPLANATION OF FIGURE 3

Text-fig. 3. *Custingophora olivacea*, CBS 335.68. — a. Sympodially branched conidiophores. — b. Different types of conidiophores. — c. Foot cell showing finger-shaped rhizoids. — d. Reduced conidial head. — e. Phialospores. — f. Habit sketch of a conidiophore with a slime ball consisting of phialospores.

is more pronounced. Colonies range from Dark Grayish Olive to Olivaceous Black (Ridgway, Pl. 46).

Colonies on Czapek agar develop only poorly, they are almost sterile.

The mycelium is hyaline to subhyaline, with the hyphae 1.5–2.5 μ in diameter. Conidiophores arise abundantly as erect or ascending branches mostly from submerged but also from aerial hyphae, usually singly, occasionally in small groups, all parts of them being evenly brown-coloured. Stipes are septate, simple or sympodially branched by successive, single, subapical proliferation, and consequently geniculate; they develop from foot cells which are characterized by conspicuous, aseptate, finger-shaped rhizoids, and range from 20–200 μ in length, 2.5–5.5 μ in diameter, the base being somewhat thicker, about 3.5–7 μ; at the apex they develop into a vesicle beneath which they are sometimes slightly constricted; they have smooth walls, up to 0.2 μ thick. Proliferations arise from the vesicle, developing successively up to 4 per conidiophore, and measuring usually 20 to 40 μ in length. Vesicles are mostly subglobose or slightly ellipsoid, 6–12 μ in diameter, with a dense mass of phialides covering one half to two thirds of its surface. Phialides develop successively on the apex of the conidiophore, which gradually enlarges to form the vesicle (Text-fig. 4); they are parallel and the outer ones definitely incurved, cylindrical to flask-shaped, showing an inconspicuous slightly narrowed neck,



Text-fig. 4. *Custingophora olivacea*, CBS 335.68. — a–g. Development of the conidial head and the foot cell.

provided with a marked collarette, and measure 6.5–10 × 2–2.5 μ. Phialospores are hyaline to subhyaline, continuous, smooth-walled, almost cylindrical when young but soon becoming ovoid or ellipsoid, 1.8–3 × 1–1.5 μ, collecting in slime balls, which are colourless or creamish when young but brownish in age, about 40 μ in diameter.

Reduced conidial heads are often found to develop on aerial hyphae with very short stipes of about 5 μ in length and with very small vesicles.

Sclerotia are not known to occur, a perfect state has not been observed.

The holotype, CBS 335.68, G.L.H. 9398, was isolated from compost by Dr. A. von Klopotek, Giessen, Germany, in 1965.

The species is thermotolerant, having its optimal development at 30° C. At 40° C the colonies reach a diameter of 5 cm within two weeks, but do not sporulate. At 10° C and 45° C growth ceases.

The authors wish to thank Miss J. B. Pannebakker for making the photographs reproduced in Pl. 9.

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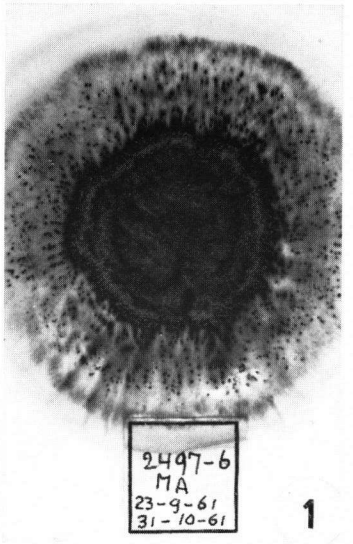
EXPLANATION OF PLATE 9

Figs. 1–4. *Thysanophora canadensis*, CBS 334.68. — 1. Dried culture made soon after isolation. — 2. Living subculture on malt agar, made 7 years later, originating from same isolate. — 3. Conidiophore, 480 ×. — 4. Section through a sclerotium, 400 ×.

Figs. 5, 6. *Thysanophora taxi*, CBS 206.57. — 5. Phialospores, 1200 ×. — 6. Conidiophore, 1200 ×.

Figs. 7, 8. *Custingophora olivacea*, CBS 335.68. — 7. Conidiophore, 1200 ×. — 8. Conidiophore, 480 ×.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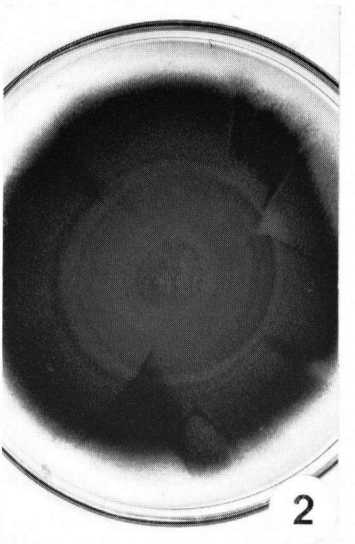
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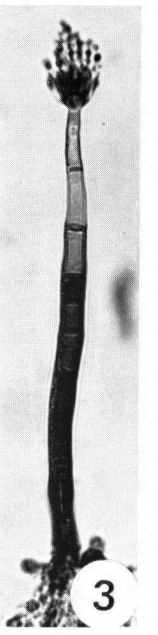
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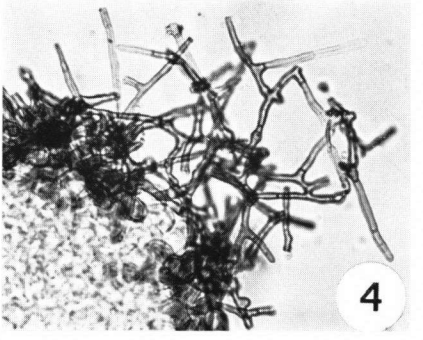
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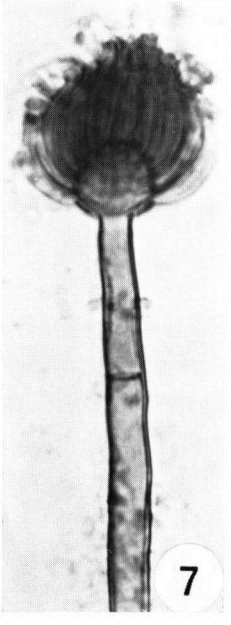
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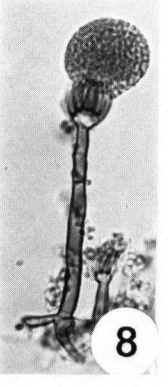
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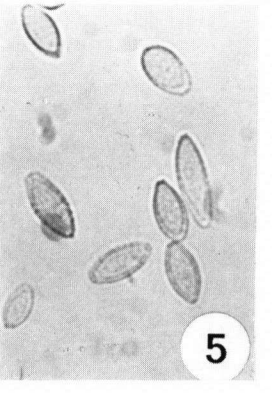
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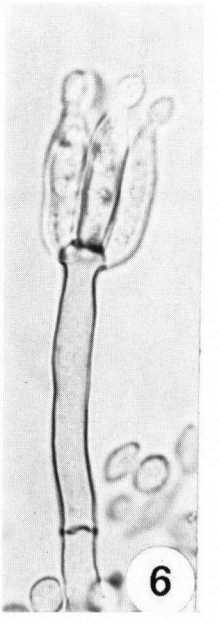
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