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A NEW HYPHOMYCETE PARASITIC ON A SPECIES OF NEMATODE

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In a paper published in 1941 (2), I described as new 10 nematode-destroying hyphomycetes found occurring in leaf mold and other decaying vegetable materials originating from several localities in Maryland, Virginia, and Wisconsin. Though these fungi and their biological relationships are not visible by direct examination of the opaque natural materials, they could be studied quite conveniently in transparent maize-meal-agar cultures which, after being permeated with *Pythium* mycelium, had been further planted with small quantities of the partly decomposed detritus harboring the fungi. They all operate in a typically parasitic rather than predaceous manner: their conidia become affixed externally to the host eelworms by adhesion, and then individually thrust a germ tube through the integument to invade the fleshy interior with an assimilative mycelium. The 4 species presented under the binomials *Acrostalagmus bactrosporus, A. obovatus, Cephalosporium balanoides,* and *Spicaria coccospora* closely resemble the insect parasites assigned to the same genera, except that their display of reproductive parts is far less luxuriant, owing to the much smaller size of the animals serving them as hosts. There is good reason to hold that these 4 species are intimately related to the series of entomogenous forms distributed among *Acrostalagmus, Cephalosporium, Spicaria,* and such allied genera as *Verticillium, Cladobotryum,* and *Beauveria.* They would seem truly kindred taxonomically to the species of *Beauveria* and *Spicaria,* for example, which because of destructiveness to the maize borer (4), the maize earworm (1), the pine bark beetle (3), and various elm insects (5), have in recent times received attention in the pages of this journal.

A fifth nematode-destroying hyphomycete of similar kinship developed abundantly in more than a dozen maize-meal-agar plate cultures that after being overgrown with mycelium of *Pythium ultimum* Trow had been further planted with small quantities of partly decayed crabgrass (*Digitaria sanguinalis* (L.) Scop.) refuse collected from several weed piles near a community vegetable garden in Arlington, Virginia, on April 25, 1944. Although several species of nematodes present in the decaying refuse multiplied freely in the cultures, the fungus apparently restricted its attack to eelworms belonging to a single species, which Dr. G. Steiner has kindly identified as *Panagrolaimus subelongatus* (Cobb) Thorne. Growth of the parasite within infected specimens could not be followed successfully while invasion was progressing, as all fleshy host structures, except the conspicuously resistant oesophagus and oesophageal bulb, soon undergo globuliferous degeneration

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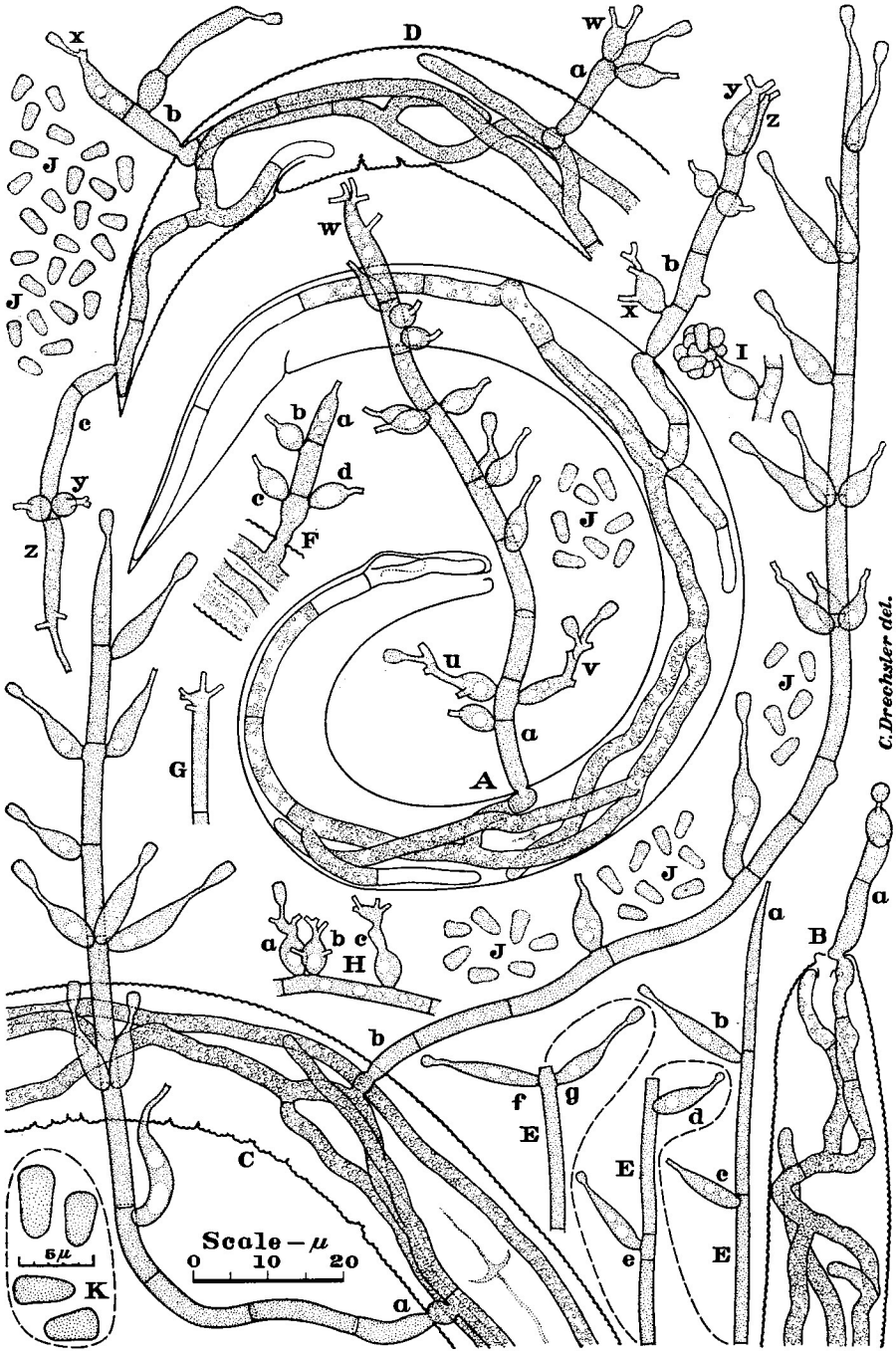

FIG. 1. *Acrostalagmus zeosporus*, drawn to a uniform scale with the aid of a camera lucida. Magnification: A–J, ×1000; K, ×2000.

and thereby offer optical difficulties that prove all the more troublesome since the contents of the young hyphae are likewise pronouncedly globuliferous. After the host contents, apart from the rather slowly evanescent oesophagus and bulb, have been largely assimilated by the fungus, and have been utilized for the production externally of conidiophorous filaments (Fig. 1, A, a, b; B, a; C, a, b; D, a–c), the assimilative mycelium is more clearly discernible. In small eelworms it may consist only of a single axial filament extending from head to tail, together with half a dozen branches of variable lengths (Fig. 1, A). Animals of greater dimensions show correspondingly more profuse hyphal development (Fig. 1, B, C), though in these also the tapering tail is usually occupied by only a single filament (Fig. 1, D). As long as the hyphae remain densely filled with globulose contents few cross-walls can be seen in them (Fig. 1, C). It is uncertain whether at this stage cross-walls are actually few or whether they are in large part obscured owing to the nature of the protoplasm wherein they are immersed. At all events, after the hyphal contents have suffered some reduction in density from the continuous transfer of substance required for formation of conidial apparatus, partitions may be distinguished at fairly close intervals (Fig. 1, A, B). Before long, further transfer of substance entails complete withdrawal of contents from terminal segments of the mycelium (Fig. 1, A, D), and thereupon evacuation of other segments proceeds until the empty host integument surrounds only the equally empty and rapidly evanescent envelopes of the assimilative filaments.

As in allied forms the character of the conidial apparatus produced by the fungus is greatly influenced by positional relationships to the substratum. In instances where the animal host has succumbed on the surface of the substratum the conidiophorous hyphae may at once grow erectly or ascendingly into the air (Fig. 1, A, a, b; D, a; F); or they may grow procumbently for some distance, and then continue their further growth ascendingly (Fig. 1, C, a, b); or, again, they may conclude their growth while still procumbent throughout their length (Fig. 1, B, a; D, b, c). In instances where the animal host has succumbed under the substratum, but not in a deeply submerged position, the conidiophorous hyphae usually make their way to the surface, often by widely divergent paths, before giving rise to conidiiferous branches, or phialides, from their prostrate or their ascending prolongations. Where, however, the animal has succumbed deep under the substratum the conidiophorous hyphae mostly fail to reach the surface, and will then give rise to phialides in submerged positions. Submerged and prostrate portions of conidiophorous hyphae show generally a rather widely spaced arrangement of phialides; many of the segments here being sterile, many others bearing only a single phialide (Fig. 1, B, a; C, b; E, a–e), and few bearing as many as 2 phialides (Fig. 1, E, f, g). In the ascending portions of conidiophorous hyphae, on the other hand, virtually all segments are fertile, with many of them bearing 2 or 3 phialides.

For the most part the phialides are of the familiar flask-shape type that

taper off distally into a single sterigma (Fig. 1, C, a, b; E, a–g). However, instances in which they support plural sterigmata are not altogether infrequent (Fig. 1, A, u–z; D, w–z; G; H, a–c). In such instances 2 (Fig. 1, D, w), 3 (Fig. 1, D, z), or 4 (Fig. 1, G) sterigmata often project separately from the same cell, and must have been put forth independently of one another; but often, again, a rachiform arrangement of sterigmatic tips gives evidence that an apical sterigma, after abstricting some conidia, grew out below its apex to form a second sporogenous tip (Fig. 1, D, x, y), which in turn gave rise to a third (Fig. 1, H, a) and possibly, on repetition of the process, to a fourth and a fifth sporogenous tip (Fig. 1, A, v). Frequently where plural sterigmata of separate origin are present, some may remain simple while one or more of the others undergo successive Beauveria-like prolongation (Fig. 1, A, u, w, x; G; H, b); so that in the end the phialide may offer a rather promiscuously appendaged aspect suggestive of the phialides figured by Petch (7) for his *Cladobotryum ovalisporum.*

It may be presumed that phialides with multiple sterigmata are generally somewhat more prolific than those of unitary make-up. The latter, as a rule, abstrict 5 to 15 conidia, which remain attached in a cohering cluster (Fig. 1, I). Clusters formed aloft in the air disintegrate when the structures supporting them finally collapse; the spores then are left strewn about on the substratum, ready to adhere apically to any specimen of the host nematode that may visit the seeded area. They recall rather strongly the conidia of *Cephalosporium balanoides,* being similarly flattened at the broad distal end and rounded at the narrower proximal end, but owing to their greater length, they show in longitudinal profile (Fig. 1, J, K) less resemblance to an acorn than to a maize (*Zea mays* L.) kernel. A term compounded in part of the generic name of maize is accordingly adopted as an appropriate epithet for the fungus. In view of its frequently liberal production of ascending conidiophorous hyphae, the species is assigned to *Acrostalagmus,* though its varied expression of reproductive habit might be more fully taken into account by resorting to the double citation "*Cephalosporium* (*Acrostalagmus*)" preferred by Petch (6) in naming related entomogenous forms.

Acrostalagmus **zeosporus** sp. nov.

Mycelium nutritum incoloratum, ramosum, septatum, intra vermiculos nematoideos viventes evolutum, in hyphis 2–3 μ crassis constans. Hyphae fertiles extra animal emortuum evolutae, subinde in materia animal ambiente omnino immersae sed saepius magna parte repentes vel ascendentes, hyalinae, vulgo simplices sed subinde ramosae, 10–300 μ longae, plerumque 1.8–3 μ crassae, in cellulis plerumque 5–25 μ longis constantes, plerisque cellulis 1–3 ramulos conidiferos sursum ferentes; ramulis conidiferis (phialis) vulgo lageniformibus, 5–20 μ longis, 2.5–4 μ crassis, plerumque sursum in unico sterigmate 0.6–0.8 μ crasso abeuntibus et 5–15 conidia gignentibus, quandoque tamen 2–4 sterigmates ex ventre proferentibus; sterigmatibus quandoque prope apicem deinceps repullulantibus ad modum *Beauveriae;* conidiis primo cohaerentibus, continuis, incoloratis, inversum elongato-nuciformibus, apice aliquid complanatis, deorsum attenuatis, basi rotundatis, plerumque 3.5–4.6 μ longis, 1.7–2.1 μ crassis.

Panagrolaimum subelongatum necans habitat in foliis caulibusque *Digitariae sanguinalis* putrescentibus in Arlington, Virginia.

Assimilative mycelium colorless, branched at moderate intervals, septate, growing within living nematodes, composed of filamentous hyphae 2 to 3 μ wide. Conidiophorous

hyphae developed outside of dead animal host, sometimes immersed entirely in the surrounding material, but more often largely prostrate or ascending, colorless, commonly unbranched but occasionally branched, 10 to 300 μ long, often 1.8 to 3 μ wide, consisting of cells mostly 5 to 25 μ long, the terminal one, often 25 μ long and as little as 1.5 μ wide, abstricting conidia at its narrow apex, the others mostly bearing 1 to 3 conidiiferous branches (phialides) at the forward end; conidiiferous branches (phialides) commonly flask-shaped, 5 to 20 μ long, 2.5 to 4 μ wide, mostly abstricting 5 to 15 conidia from a single terminal sterigma, but occasionally putting forth 2 to 4 separate sterigmata; the sterigmata whether present singly or plurally sometimes through successive subapical elongation forming one or more additional sporogenous tips in zigzag arrangement; conidia first cohering in a cluster, continuous, coloriess, inversely elongate-nuciform, somewhat flattened at the distal end, narrowing downward, rounded at the proximal end, mostly 3.5 to 4.6 μ long and 1.7 to 2.1 μ wide.

Parasitizing *Panagrolaimus subelongatus* it occurs in decaying leaves and stems of *Digitaria sanguinalis* in Arlington, Virginia.

In connection with the morphological tendencies evident in the new species it may be appropriate to mention here that more than half a century ago a mold very similar to *Botrytis* (now *Beauveria*) *bassiana* was briefly noted by Zopf (8, p. 340) among several fungi he had observed causing nematode diseases.

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