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A NEW NEMATODE-DESTROYING HARPOSPORIUM[1]

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ABSTRACT

A mucedinaceous parasite, observed destroying eelworms in a maize-meal-agar plate culture prepared by the addition of some forest duff from western Maryland, is newly described as *Harposporium cycloides*. Its conidia differ from the crescentic or semicircular conidia of the familiar *H. anguillulae* in that their curvature extends to an angular magnitude of approximately a full circle. Its chlamydospores, though sometimes found united in pairs, are much more often produced singly than are the chlamydospores of *H. anguillulae*.

IN THE REPORT that includes the original account by Lohde (1874) of a nematode-destroying fungus which he named *Harposporium anguillulae*, the morphological treatment provides no dimensional measurements, and accordingly serves better in defining the genus, then newly presented, than in delimiting the species on which it was erected. Later Zopf (1888) described and illustrated, as *H. anguillulae*, a fungus whose endozoic assimilative hyphae and external reproductive hyphae would seem, from the magnification given for relevant figures, to measure 2.5–4 μ in width. Here and there along the external hyphae were produced globose bodies, 2.5–3.6 μ in diam, which Zopf in conformity with earlier usage designated as basidia, since they gave rise on delicate sterigmata to conidia. These conidia, of the distinctive crescentic shape signalized in the generic term invented by Lohde, were stated to measure 1–2 μ in width; and their two ends, which after disjunction of the spore became equally pointed, inclosed a chord 7–13 μ long. Although Lohde made no mention of spores other than conidia, Zopf also observed the production of distended or barrel-shaped chlamydospores, apparently about 4.5–9 μ long and 5.5–7.5 μ wide, which were formed from short segments of the assimilative hyphae. At times they were produced singly, but oftener they originated plurally in chains. Much agreement and some noteworthy disagreement with the morphological features of *Harposporium anguillulae* Lohde emend. Zopf is evident in a recently found, congeneric fungus that similarly subsists parasitically on nematodes.

MATERIALS AND METHODS—The congeneric fungus in question made its appearance in a petri plate of maize-meal agar that had been planted 30 days earlier with three small pinches of leaf mold taken from deciduous (predominantly oak) woods near Cumberland, Md., on 12 November 1967. When first seen the fungus was restricted to an area of approximately 100 square mm adjoining one of the three deposits of decaying duff. During the next 20 days, it spread slowly by attacking eelworms apparently referable to a species of *Rhabditis* (Fig. 1); earlier it had manifestly destroyed also some scattered individuals of an unusually long slender nematode species (Fig. 2). After the culture was 60 days old, the activity of the parasite gradually diminished and eventually halted. It was estimated that about 300 animals, scattered over an area of approximately 200 square mm, succumbed; some died on the surface of the agar, others in submerged positions. While the fungus remained active, portions of agar containing infected eelworms were removed at weekly intervals for microscopic examination.

RESULTS—In all examinations the fungus showed the same combination of features that distinguishes it from all hitherto known species of *Harposporium*. It is therefore described as a new number of the genus.

Harposporium **cycloides** sp. nov. [2] (Fig. 1–6)—Assimilative hyphae colorless, developing within nematodes, mostly about 2.5 μ wide. Conidiophores produced outside of moribund or dead

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[2] Hyphae assimulantes incoloratae, intra vermiculos nematoideos evolutae, vulgo circa 2.5 μ latae. Hyphae fertiles extra animal moribundum vel emortuum evolutae, procumbentes vel intra materiam animal circumdantem dispositae, saepius 20–80 μ longae et 2–3 μ latae, in cellulis 7–35 μ longis consistentes; cellula terminalis saepe conidia in apice sterigmatis gignens, aliae cellulae nonnullos (vulgo 1–4) ramulos conidiferos a latere ferentes. Ramuli conidiferi (phialae) in parte inferiore globosa plerumque 3.5–4.5 μ crassa et sterigmate cylindraceo plerumque 2–3 μ longo et 0.8–1.0 μ lato constantes. Conidia incolorata, in spiram semel ad modum caulis humuli lupuli cochleatim convoluta, vulgo 20–30 μ longa et 1.3–1.9 μ crassa, basi circa 1 μ crassa et ibi aburpte rotundata, apice rostro acuto inani 1.5–2 μ longo praedita. Chlamydosporae flavidae, in hyphis assumentibus plerumque singulatim oriundae sed

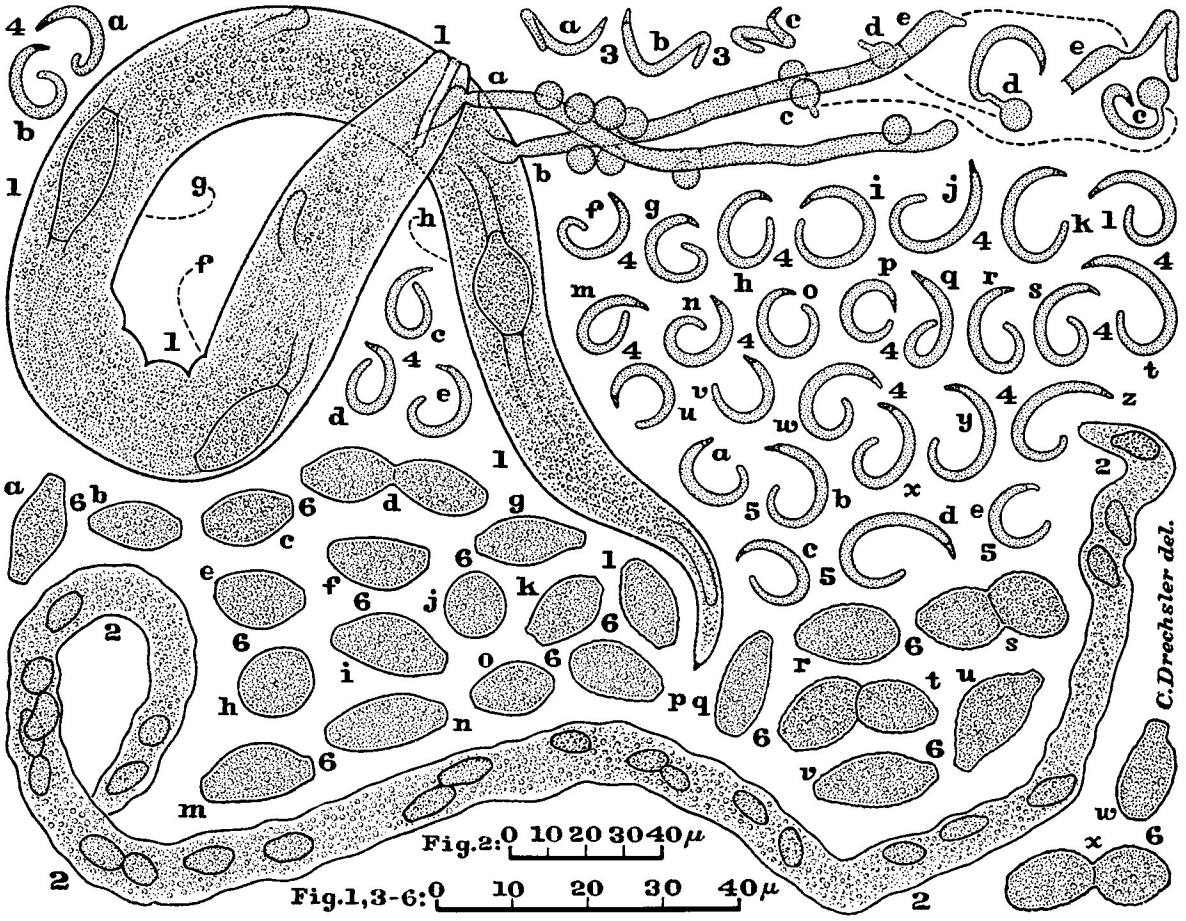

Fig. 1–6. *Harposporium cycloides*.—Fig. 1. An infected nematode (*Rhabditis* sp.) from which have been extended two conidiophores, a-b, each bearing laterally several globose sporiferous branches; two of these branches, c-d, and the terminal cell, e, are also shown separately nearby, following the production of attached conidia during the ensuing 11 hr; f-h, chlamydospores formed on endozoic assimilative hyphae, ×1,000.—Fig. 2. Remains of a long slender nematode within which have been formed 18 single and 3 binary chlamydospores, ×500.—Fig. 3. Conidia, a-c, as seen when viewed from a direction nearly perpendicular to their helical axes, ×1,000.—Fig. 4 (a-z), Fig. 5 (a-e). Conidia as usually seen somewhat flattened under a cover-glass, with their ends in many instances more widely separated than while still in undisturbed condition, ×1,000.—Fig. 6. Chlamydospores, a-x, mostly produced singly, but including several binaries, d, s, t, x; some, a-k, m-o, appear smooth; others, l, p-x, are minutely roughened, ×1,000.

host animal, either on or below surface of substratum, mostly 20–80 μ long and 2–3 μ wide, composed of segments usually 7–35 μ long, the terminal segment often producing a sterigma, the other segments commonly bearing several (often one to four) sporiferous branches laterally. Sporiferous branches (phialides) composed of a globose basal part, usually 3.5–4.5 μ in diam, together with a cylindrical sterigma mostly 2–3 μ long and 0.8–1.0 μ wide. Conidia colorless, convolved in a helicoid spiral of approximately one turn and always of left-handed rotation, mostly 20–30 μ in total length along the curved axis, usually 1.3–1.9 μ in greatest width, about 1 μ wide at the abruptly rounded base, furnished at the tip with a rather thick-walled beak mostly 1.5–2 μ long. Chlamydospores produced intercalarily in assimilative hyphae, mostly singly but sometimes in pairs; yellowish, usually elongate-ellipsoidal or globose or barrel-shaped, often smooth but sometimes showing a minutely rough outer contour, mostly 8–19 μ long and 6–9 μ in greatest width.

In a nematode infected by *Harposporium cycloides* (Fig. 1) the mycelium extending lengthwise through the fleshy interior is for the most part only indistinctly visible. Portions of assimi-

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interdum binae conjunctae ortae, vulgo elongato-ellipsoideae vel globosae vel dolioformes, saepius leves sed aliquando minute asperae, plerumque 8–19 μ longae, 6–9 μ crassae.

Vermiculos nematoideos necans habitat in foliis arborum (magnam partem quercorum) putrescentibus prope Cumberland, Maryland. Typus: Figurae 1–6.

lative hyphae are often clearly discernible only in the animal's tail and in peripheral positions where conidiophores are soon to be extended after narrow perforation of the host cuticle. As in congeneric species the young conidiophore (Fig. 1a, b) gives rise laterally to globose cells, each of which, together often with the terminal cell, develops into a phialide by putting forth a delicate sterigma. Production of a conidium on a sterigma has in several observed instances (Fig. 1c-e) been found to have been accomplished during a period of approximately 11 hr. The disposition of the conidium in a helicoid spiral comprised approximately of a single distally widening coil of left-handed rotation (Fig. 1e; 3a-e) is not well revealed in material mounted under a cover-glass, where the spore usually becomes flattened into a somewhat circular or elliptical shape (Fig. 4a-z; 5a-e). Helicoid spiral conformation of left-handed rotation is found also in the conidia of *H. anguillulae* and *H. helicoides* Drechsler (1941), but the spores of the former species comprise only one-half of a coil, while those of the latter species comprise three-quarters of a coil. By way of further contrast *H. cocleatum* Drechsler (1965), which parasitizes rotifers, gives rise to helicoid conidia that comprise individually a single coil of right-handed rotation.

Chlamydospores of *Harposporium cycloides* often develop within infected nematodes (Fig. 1f-h) at the same time when conidiophores and conidia are being produced externally. Originating singly (Fig. 6a-c, e-r, u-w), or occasionally in pairs (Fig. 6d, s, t, x), they look very unlike the hyphae on which they are borne (Fig. 1); whereas the chlamydospores of *H. anguillulae* commonly originate in chains and at all stages generally remain recognizable as indurated hyphal segments. At early maturity the chlamydospores of the new species ordinarily appear smooth (Fig. 6a-k, m-o), but after aging for several days or a few weeks some among them may show a minutely roughened surface (Fig. 6l, p-x).

The same difference in amount of conidial curvature that separates *Harposporium cycloides* from *H. anguillulae* must be held to separate it also from *H. lilliputanum* Dixon (1952), a species with arcuate conidia measuring 4.5–9 × 1–1.5 μ, as well as from *H. crassum* Shepherd (1955), a species with arcuate conidia 18–22 μ long and 2–3 μ wide. Among investigators in the United States, Norton (1963) reported the occurrence of *H. lilliputanum* in Iowa, and the same parasite has from time to time come under observation in some of the author's nematode-infested agar cultures that had been inoculated with specimens of decaying plant material from Maryland, New Hampshire, and Wisconsin. A fungus, agreeing fairly well with *H. crassum* in its main dimensions, has likewise appeared at various times in the cultures, but its identification remains uncertain, since its conidia are generally rather blunt at the ends, not markedly acuminate like many of the spores shown in Shepherd's figure.

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