

MYCOLOGY.—A *Harposporium* infecting eelworms by means of externally adhering awl-shaped conidia.<sup>1</sup> CHARLES DRECHSLER, Bureau of Plant Industry, Soils, and Agricultural Engineering.

The genus *Harposporium* was erected by Lohde (6) in 1874 on a hyphomycetous nematode parasite that produced sickle-shaped conidia on slender sterigmata arising from globose diverticulations borne laterally on filamentous hyphae extended from the body of the animal host. This parasite he designated as *H. anguillulae*, but since in its brevity the original descriptive account—apparently only a recorder's report of an oral discourse—contained little intimate detail, a really precise definition of the species was lacking until Zopf (7) 14 years later furnished very adequate particulars from study of an eelworm parasite that he had found in several localities in Germany and held identical with Lohde's. There is, indeed, much likelihood that the identification was correct, for although the characterization given by Lohde would seem to fit tolerably well one other nematode-destroying form that has appeared in my cultures, the fungus described by Zopf seems of far more frequent occurrence, at least in the Middle Atlantic States, than any hyphomycetous nematode parasite that could be regarded as congeneric with it. Conidia not typically falcate are produced by four allied species I have set forth as members of the genus (1, pp. 793–800; 4) under the binomials *H. helicoides*, *H. oxycoracum*, *H. diceraeum*, and *H. bysmatosporum*; the inclusion of these species being justified more especially by the subglobose shape of the lateral cells bearing the sterigmata on which the conidia are formed. More recently another nematode parasite was found that seems best referable to the genus, though its conidiiferous cells depart rather markedly from the subspherical type.

This parasite came to light in more than a dozen maize-meal-agar plate cultures which after being overgrown with mycelium of *Pythium ultimum* Trow, had been further planted with small quantities of decaying grass taken from an old weed pile in a garden near Beltsville, Maryland, on February 2,

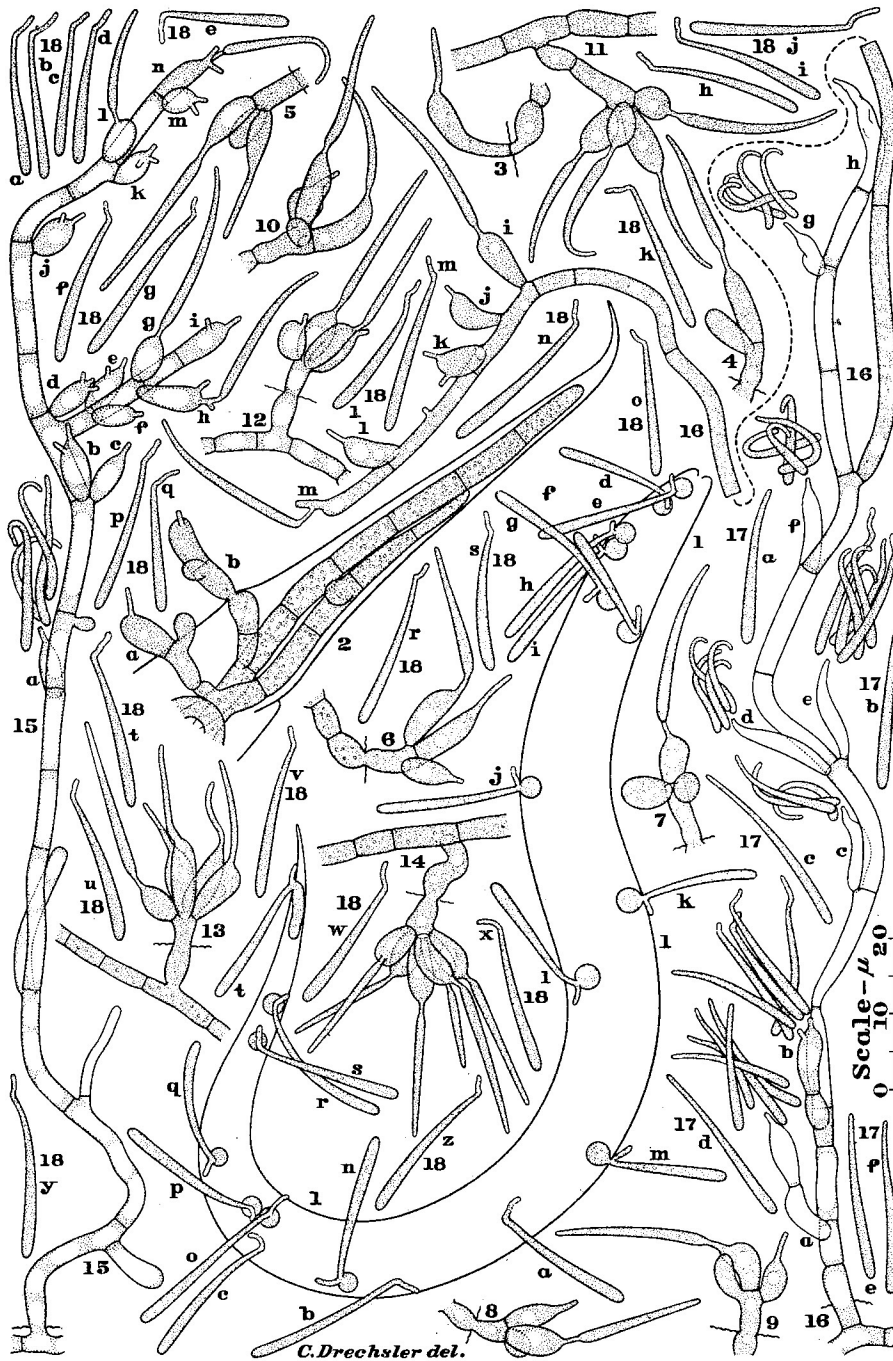
1950. Among the intermixed decaying leaves and culms, inflorescences of crabgrass (*Digitaria sanguinalis* (L.) Scop.), goosegrass (*Eleusine indica* (L.) Gaertn.), bristlegrass (*Setaria* sp.), and Bermuda-grass (*Cynodon dactylon* (L.) Pers.) remained easily recognizable. Rapid multiplication of eelworms ensued after the detritus was added, so that within ten days the cultures became liberally populated with these animals. One of the several species of eelworms that occurred in greatest number—a sharp-tailed species of *Rhabditis*<sup>2</sup> whose adult males, present in some abundance, were distinguished by a spicule of unusually large dimensions—then suffered mounting losses, and eventually the destruction of all motile individuals. Attack on a host nematode always proceeded from slender awl-shaped conidia that adhered to the integument. Early in the course of an epizootic within a plate culture, animals were most often found encumbered with one or, perhaps, two spores, but at later stages many eelworms could be seen moving about laden with a score of the tapering bodies (Fig. 1, *a-t*). Each conidium adhered by its narrower distal end, where it bore a slender process, mostly 2 to 5 $\mu$  long, that was directed usually at a wide angle with the main axis. This slender apical process was always formed before the spore adhered, and for some little time following affixture was the only modification to be seen (Fig. 1, *a-c*). Fairly soon, however, the spore would intrude a delicate germ tube through the animal's integument, which on reaching the fleshy interior widened immediately to form a globose infection bladder (Fig. 1, *d-s*). Before long the conidium showed increasing vacuolization as its contents migrated gradually into the bladder, and finally its membranous envelope was wholly emptied of protoplasm. Thereupon, or sometimes a lit-

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the earlier (Fig. 1, *t*), the infection bladder began autonomous development by elongating as an assimilative hypha. The course of the growing hypha was marked by globu-

lose degeneration of musculature and organs, with the usual unfortunate result that for a time the invading filament and all its branches were largely obscured from view.



FIGS. 1-18.—(See opposite page for legend).

Later, after the degenerating materials had mostly been absorbed, the assimilative hyphae, whether originating from single or plural infections, could be seen permeating the animal's interior somewhat uniformly from head to tail (Fig. 2). At this state they revealed cross-walls at moderate or rather short intervals, and the cells composing them were filled rather completely with protoplasm containing numerous granules. Subsequently as the animal's contents became further diminished, the continued withdrawal of substance necessary for the production of conidiophores and conidia was accompanied by increasing vacuolization, until finally the membranous envelopes of the assimilative hyphae, like the host integument surrounding them, were emptied of all protoplasmic residues.

The fungus always sporulates outside of the dead animal. Lateral branches are given off from scattered segments of the assimilative hyphae, and soon push their way out narrowly through the host integument (Fig. 2, *a*, *b*). Their external development is influenced markedly by the positional relationship of the dead eelworm in or on the infested material. If the animal host chanced to succumb on the surface, the lateral branch on erupting from the integument may at once give rise to an external cell that soon pushes up an apical protrusion (Fig. 3) which will develop into a sterigma and conidium. After one terminal conidiiferous cell, or phialide, has been delimited, usually through deposition of a cross-wall about  $5\mu$  outside the integument, a second phialide is often burgeoned forth laterally just below the first

(Fig. 4). Other phialides may be produced, likewise in lateral positions directly below the first or terminal one, thereby giving rise to clusters of three (Figs. 5-9), four (Figs. 10-12), five (Fig. 13), and even six (Fig. 14). Dead host animals found on the surface of infested materials thus come to bristle with phialides in many places, and eventually the areas surrounding them are thickly bestrewn with detached conidia.

In instances where the infected eelworms happen to succumb in positions well under the surface of an agar plate culture, much rangier conidial apparatus is developed. Many of the branches here, on erupting from the host integument, do not give rise to phialides directly, but grow upward or ascendingly through the ambient, and on reaching the surface often elongate procumbently (Figs. 15, 16). The resulting filaments, which often exceed 100 or  $200\mu$  in length, and which soon become partitioned into cells mostly 5 to  $15\mu$  long, show some meager branching of ordinary character apart from the more abundant special type of branching entailed in their production of lateral phialides (Fig. 15, *a-h*, *j-m*; Fig. 16, *a-g*, *i-l*) from the distal end of many hyphal segments other than the terminal segments. The lateral phialides are produced singly or in two's immediately below the distal cross-wall, their plural production usually being less frequent in submerged positions (Fig. 16, *d*, *e*) than in positions on or above the surface (Fig. 15, *b*, *c*; *e*, *f*; *g*, *h*; *k*, *l*). In addition to the apical sterigma with which they are regularly provided, they often show one (Fig. 15, *b*, *e*, *j*, *m*; Fig. 16, *b*, *k*) or two (Fig. 15, *d*, *k*) sterig-

FIGS. 1-18.—*Harposporium subuliforme*, sp. nov., drawn to a uniform magnification with the aid of a camera lucida,  $\times 1000$ : 1, Small specimen of *Rhabditis* sp. to which are affixed 20 conidia, *a-t*: three of them, *a-c*, show only an adhesive spur at the apex; in 16, *d-s*, an infection bladder is present as well as an adhesive spur; in one, *t*, the infection bladder has begun to elongate into an assimilative hypha. 2, Tail end of a specimen of *Rhabditis* sp., showing interior of animal occupied by assimilative hyphae from which two reproductive hyphae, *a* and *b*, have been extended. 3, Short reproductive hypha terminating in a single phialide. 4, Short reproductive hypha with a full-grown terminal phialide and an incompletely developed lateral phialide. 5-9, Short reproductive hyphae, each with three phialides, full grown or in course of development. 10-12, Short reproductive hyphae, each with four phialides, full grown or in course of development. 13, Short reproductive hypha bearing five fully developed phialides. 14, Short reproductive hypha with six fully developed phialides. 15, Long reproductive hypha from an eelworm host that succumbed under agar; the proximal portion of the hypha whereon is borne one phialide, *a*, with its four abjoined conidia, is submerged, while the distal portion with 13 phialides, *b-n*, is partly procumbent and partly ascending. 16, Long reproductive hypha extended from a submerged animal host; the proximal portion of the hypha whereon are borne eight phialides, *a-h*, is submerged, so that the conidia formed on seven of these phialides, *a-g*, have remained clustered near the sterigmata; the five phialides, *i-m*, borne on the procumbent distal portion of the branch prolonging the hypha are denuded of their fully developed conidia. 17, Detached conidia, *a-d*, without adhesive spurs. 18, Abjoined conidia, *a-z*, each with an adhesive spur at its tip.

mata arising a short distance from the tip. Occasionally, besides, a sterigma may become branched (Fig. 15, *h*). The terminal hyphal segments here, as in related fungi, serve directly as phialides, and like the lateral cells of similar function, may have simple apical sterigmata (Fig. 16, *h*), branched sterigmata (Fig. 16, *m*), or plural sterigmata (Fig. 15, *i, n*).

The lateral phialide may rather correctly be described as flask-shaped, for with its usually elongated ellipsoidal venter, often two or three times longer than wide, and its slender though short sterigma, it offers much resemblance in outward form to some flasks of glass or metal that are widely familiar in our commerce. Only now and then is its venter so nearly globose that the shape of a Florence flask is approximated. As the Florence-flask type of lateral cell is strongly characteristic of *Harposporium anguillulae*, and occurs, besides, in the four congeneric species more recently made known, the present fungus might seem, if only the shape of its phialides were considered, less aptly referable to *Harposporium* than to *Acrostalagmus* or *Cephalosporium*. However, its conidia, when newly formed (Fig. 17, *a-d*), are slender awl-shaped bodies of dimensional proportions not usual in either of the two genera last mentioned. Wherever they develop undisturbed in submerged positions they form groups of four, or five, or six, at the tips of the individual phialides (Fig. 15, *a*; Fig. 16, *a-g*). In these groups they very often show pronounced curvature (Fig. 15, *a*; Fig. 16, *c, d, f, g*), and thereby come to resemble rather strongly the conidia of *H. anguillulae*. As a rule the conidia that have developed in the air, whether on phialides surmounting eelworms which succumbed in surface positions, or on phialides arising from procumbent or ascending prolongations of hyphae extended from dead animal under the surface (Fig. 15, *c-n*; Fig. 16, *i-m*), are larger and more nearly straight than those of submerged origin. Soon after they have been cut off, the conidia very commonly will grow out obliquely at the tip to form the delicate process (Fig. 18, *a-z*) by means of which they adhere to suitable roving eelworms. In respect to this curious incipient germination, the fungus shows parallelism with the several

clamp-bearing basidiomycetous forms of similar biological habit that have been made known as members of the genus *Nematoclonus* (1, pp. 773-780; 2; 3; 5). More especially it appears comparable with my *N. tylosporus* (1) since its adhesive substance is not collected in a visible droplet. Development of the adhesive spur is here not accompanied, as in species of *Nematoclonus*, by partial evacuation of the conidium.

Presumably the fungus, like the species of *Harposporium* described earlier, is to be reckoned in the same series of interrelated clampless hyphomycetes to which belong the insectivorous as well as the nematode-destroying species of *Acrostalagmus* and *Cephalosporium*. A term having reference to its awl-shaped conidia may serve as an appropriately suggestive specific name.

**Harposporium subuliforme**, sp. nov. Mycelium nutritum ramosum, incoloratum, septatum, intra vermiculos nematoideos viventes evolutum, in cellulis plerumque 5-15 $\mu$  et 2-3.8 $\mu$  crassis constant. Hyphae fertiles extra animal emortuum evolutae, interdum in materia animal ambiente omnino immersae interdum omnino vel magnam partem procumbentes vel ascendentes, in axe simplicibus vel parum ramosae, saepius 5-300 $\mu$  longae, in cellulis plerumque 5-25 $\mu$  longis et 2-3 $\mu$  crassis constantes, ex plerisque cellulis 1-5 ramulos conidiferos sursum ferentes; cellula terminali etiam ramulis conidiferis (phialis) vulgo lageniformibus, 5-12 $\mu$  longis, 2.8-4.3 $\mu$  crassis, supra in 1-3 sterigmata abeuntibus, saepe 4-6 conidia gignentibus; sterigmatibus simplicibus vel rarius ramosis, vulgo 1-2.5 $\mu$  longis, 0.6-.8 $\mu$  crassis, conidiis incoloratis, subuliformibus, 12-26 $\mu$  longis, basi rotundatis, ibi 1-1.8 $\mu$  crassis, prope apicem 0.4-0.6 $\mu$  crassis, ex apice ramusculum glutinosum 2-5 $\mu$  longum et circa 0.6 $\mu$  crassum emittentibus.

Vermiculos nematoideos specie *Rhabditis necans* habitat in foliis caulibusque graminum (*Digitariae sanguinalis*, *Eleusinis indicae*, *Cynodontis dactyli*, *Setariae* sp.) putrescentibus prope Beltsville, Maryland.

Assimilative mycelium colorless, branched, septate, growing within living nematodes, composed of cells mostly 5 to 15 $\mu$  long and 2 to 3.8 $\mu$  wide. Conidiophorous hyphae developed outside of dead host, sometimes immersed entirely in the surrounding material, sometimes in part or wholly procumbent or ascending, only rather little given to

ordinary branching, often 5 to 300 $\mu$  long, composed of cells mostly 5 to 25 $\mu$  long and 2 to 3 $\mu$  wide, the terminal cell regularly being conidiiferous, many of the other axial segments bearing 1 to 5 conidiiferous branches (phialides) distally; conidiiferous branches commonly flask-shaped, often 5 to 12 $\mu$  long, 2.8 to 4.3 $\mu$  wide, provided distally with 1 to 3 sterigmata on which collectively are often borne 4 to 6 conidia; sterigmata usually simple, occasionally branched, commonly 1 to 2.5 $\mu$  long and 0.6 to 0.8 $\mu$  wide; conidia colorless, awl-shaped, straight or curved, 12 to 26 $\mu$  long, rounded at the base, proximally 1-1.8 $\mu$  wide, distally often 0.4 to 0.6 $\mu$  wide, soon putting forth obliquely from the tip an adhesive spur 2 to 5 $\mu$  long and about 0.6 $\mu$  wide.

Parasitic on a species of *Rhabditis*, it occurs in decaying leaves and stems of various grasses (*Digitaria sanguinalis*, *Eleusine indica*, *Cynodon dactylon*, and *Setaria* sp.) near Beltsville, Md.

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