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BOTANY.—*Two new basidiomycetous fungi parasitic on nematodes.*[1] CHARLES DRECHSLER, Bureau of Plant Industry.

Among several fungi set forth in an earlier paper (4) as attacking nematodes after the usual manner of parasites, by intrusion of hyphal elements arising through germination of adhering conidia, were included two species which from their production of clamp-connections were obviously to be reckoned among the Basidiomycetes. The two species, it was clear, were intimately akin to one another; yet owing to somewhat incidental differences in the make-up of their sporulating apparatus they could not both be assigned satisfactorily to any one mucedinaceous genus then available. Accordingly a new genus, *Nematoctonus,* was erected in which they were described under the names *N. tylosporus* and *N. leiosporus.* Subsequently two other forms, similarly parasitic on free-living nematodes, and similarly provided with clamp-connections, have been observed in transparent Petri-plate cultures. These two forms, which like those presented earlier have more than ordinary interest, since they represent basidiomycetes habitually subsisting on animals that normally remain in a motile state from the time of hatching until the approach of death, are described herein as additional species of *Nematoctonus.*

One of the two species came to light on September 1, 1942, in a maize-meal-agar plate culture that on August 24, 1942, had been planted with the softened stem of a newly damped-off tomato seedling from a greenhouse at the Bureau of Plant Industry Station near Beltsville, Md. The fungus, when first observed, was barely visible to the naked eye as a very delicate arachnoid weft festooned over a portion of the decaying tomato material. It failed to spread to other areas of the original culture, apparently for the reason that in its initial development all individuals of the susceptible species of nematode had been exterminated. However, when a small quantity of the delicate weft was transferred to another Petri-plate culture, which likewise had been started on August 24, from a damped-off tomato seedling, and which likewise had afforded ample development of free-living eelworms soon after the agar substratum became permeated with mycelium of *Pythium irregulare* Meurs, the arachnoid fungus resumed its destructive activity on a larger scale. Everywhere in the second culture it parasitized a single nematode species that manifestly was identical with the species it had exterminated in the original culture. The eelworm in question was determined by Dr. G. Steiner to belong to a group of forms that have been cited in the literature rather indiscriminately under the binomial *Rhabditis monhystera* Bütschli.

During the earlier stages of invasion the assimilative mycelium within an infected nematode is usually obscured very badly by the globulose materials resulting directly from degeneration of the host tissues. Later, when these globulose materials have in large part been appropriated by the fungus and have been utilized for the production externally of conidiophorous filaments (Fig. 1, *A, a, b*), the assimilative hyphae are better discernible. In some instances the empty membrane of the conidium (Fig. 1, *A, c*) that initiated the attack may then still be seen attached to the outside of the host integument, its prolongation in the empty germ hypha visibly communicating with the mycelium inside. Occasionally the empty envelopes of several conidia operative in accomplishing infection may be seen attached to the dead animal. The quantity of assimilative mycelium, however, would seem little influenced by the number of adhering spore envelopes, for multiple hyphae are readily produced by branching. Frequently branches arise directly from, or in close proximity to, clamp-connections, though some clamps having no special positional relationship to branches are usually present. Occurrence of clamps without relationship to branches or to any other lateral outgrowths could be noted also in external hyphae that happened to lie submerged for considerable distances under the surface of

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[1] Received March 24, 1943.

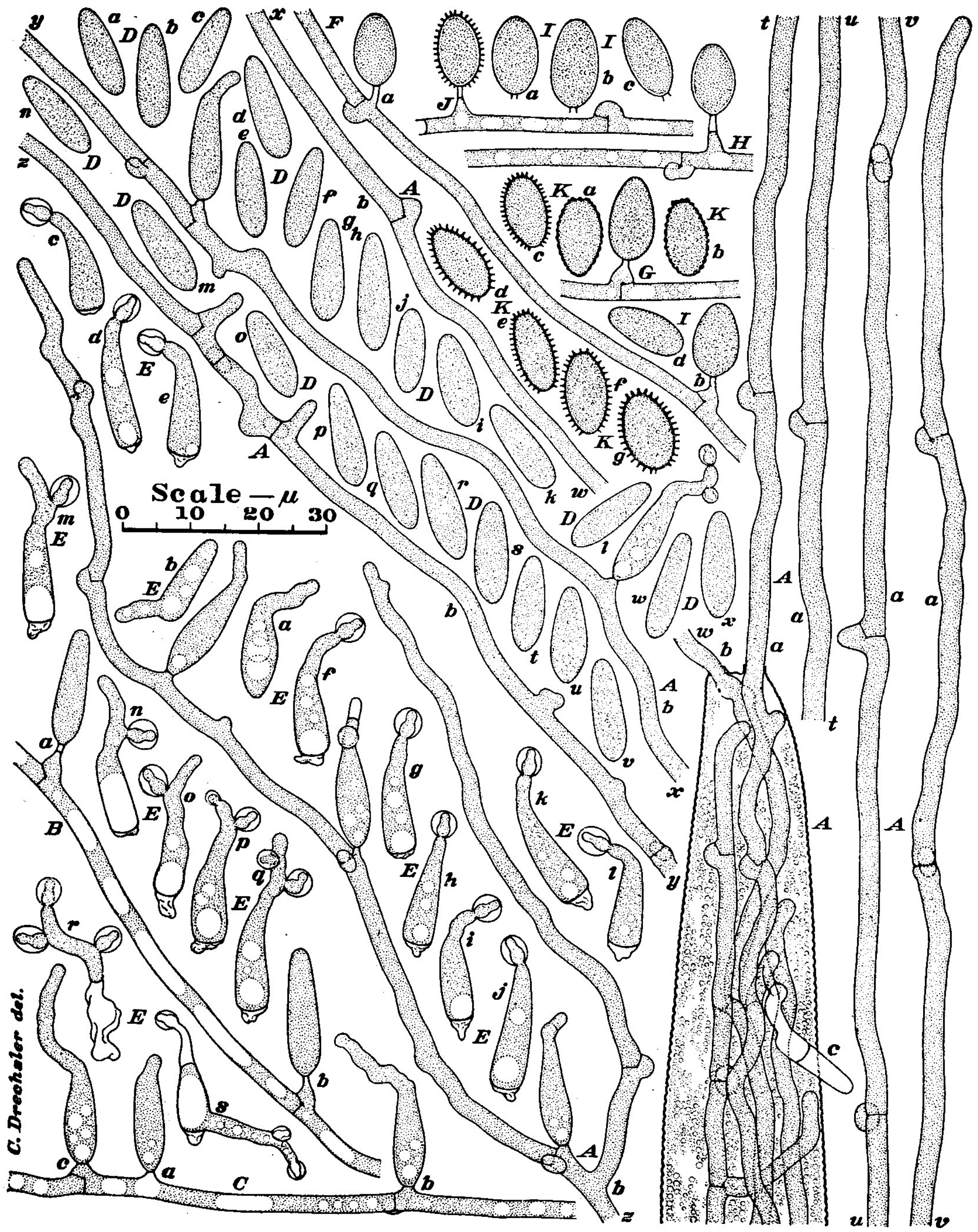

Fig. 1.—*Nematoctonus pachysporus*, drawn to a uniform magnification with the aid of a camera lucida; ×1,000 throughout. *A*, Anterior portion of nematode host permeated with assimilative mycelium from which one hypha, *a*, has been extended into the surrounding agar culture medium, while another hypha, *b*, has been extended into the air; the two external hyphae, from want of space, being shown in sections whose proper continuity is indicated by the two sequences of paired letters, *t–v* and *w–z*, respectively; *c*, empty envelope of conidium, attached externally, from which the assimilative mycelium had its origin. *B*, Portion of conidiophorous hypha, bearing conidia on two longish sterigmata, *a* and *b*. *C*, Portion of conidiophorous hypha bearing solitary conidia on three short sterigmata, *a-c*. *D*, Conidia, *a-x*, showing variations in size and shape previous to germinative development. *E*, Conidia, *a-s*, showing variations in germinative development. *F*, Portion of conidiophorous hypha with two sterigmata, *a* and *b*, whereon are borne solitary ovoid spores destined for conversion into resting spores. *G*, *H*, Portions of conidiophorous hypha, each with an ovoid spore soon to be converted into a resting spore. *I*, Three ovoid spores before conversion into resting spores. *J*, Portion of conidiophorous hypha showing an echinulate resting spore borne on a sterigma. *K*, Resting spores, *a-g*, showing variations in size, shape, and echinulation.

the agar culture medium (Fig. 1, *A*, *a*).

A submerged position, of course, is not a usual one for the external, conidiophorous filaments. Most often they grow out somewhat ascendingly into the air to attain lengths ranging from 1 to 1.5 mm. As their development continues they sooner or later decline to the substratum, so that eventually they come to lie prostrate in areas where the surface is smooth, or are draped loosely over prominences in more rugged areas. Occasionally a conidiophorous filament may grow out in a procumbent posture.

Whatever their posture may be, the aerial filaments become studded at intervals with clamp-connections, which often give rise, on short narrow sterigmata, to erect strobiliform conidia (Fig. 1, *A*, *b; B*, *a*, *b*). Conidia may, however, arise without any close positional relationship to clamp-connections (Fig. 1, *C*, *a*). Sometimes a conidium is borne almost sessile on the parent filament in close proximity to a clamp-connection (Fig. 1, *C*, *b*)*;* or, again, it is attached, without any noticeable sterigma, directly to the dorsal side of a clamp (Fig. 1, *C*, *c*). Ordinarily its original strobiliform shape (Fig. 1, *D*, *a–x*) is soon modified as the result of germinative development. A short broad process is extended usually from the distal end (Fig. 1, *E*, *a*) or, in rare instances, from the basal end (Fig. 1, *E*, *b*). This process gives rise at its tip to a globose adhesive body, measuring usually 3 or 4μ in diameter, and consisting apparently of a narrowed hyphal termination together with a layer of glutinous secretion (Fig. 1, *E*, *c–l*). Thereupon the outgrowth may resume elongation (Fig. 1, *E*, *m–o*) to produce terminally a second adhesive body (Fig. 1, *E*, *p*, *q*). Elongation may then be resumed again, with eventual development of a third adhesive body (Fig. 1, *E*, *r*). In some instances where a germ outgrowth is put forth from the basal end as well as from the distal end, one of the outgrowths may form a single adhesive body while the other may produce two such bodies (Fig. 1, *E*, *s*). The transfer of protoplasmic materials required for such incipient germinative development is accompanied by vacuolization and evacuation usually of the basal portion of the conidium, and by collapse of the emptied portion of conidial envelope. Occasionally the entire protosplasmic contents may migrate into the stout germ outgrowth (Fig. 1, *E*, *r*).

In addition to the colorless thin-walled conidia discussed so far, the fungus produces resting spores. These likewise are mostly borne on short sterigmata arising from clamp-connections or in close proximity to clamp-connections (Fig. 1, *F*, *a*, *b; G; H*). During their earlier stages of development they resemble conidia, though usually they may be distinguished even then by their broader ovoid shape (Fig. 1, *I*, *a–d*). In their ripe condition (Fig. 1, *J*) they have a perceptibly yellowish coloration, and individually are surrounded by a thicker wall, which sometimes is modified externally with bullate sculpturing (Fig. 1, *K*, *a*, *b*), but oftener is closely beset with slender spiny protuberances (Fig. 1, *K*, *c–g*). As these resting spores have never been seen to germinate, it may be presumed that like the resting spores of *Nematoctonus tylosporus* they are adapted for tiding over unfavorable periods.

The greater thickness of its conidia relative to the conidia of the three known congeneric species suggests the epithet proposed for the fungus.

Nematoctonus pachysporus, sp. nov.

Hyphae assumentes incoloratae, irregulariter ramosae, plerumque 2–3.5μ crassae, in modum Hymenomycetum septato-nodosae, intra vermiculum nematoideum viventem crescentes, post mortem animalis aliquot hyphas fertiles extra emittentes; hyphis fertilibus incoloratis, simplicibus vel parce ramosis, primo plerumque ascendentibus postea procumbentibus, mediocriter septato-nodosis, vulgo 0.5–1.5 mm longis, 2.2–3.2μ crassis, conidia vel sporas perdurantes quandoque protinus ex nodis quandoque ex sterigmatis singulatim gerentibus; sterigmatis erectis, 0.5–5μ longis, basi 1–2.5μ crassis, apice 0.6–1μ crassis; conidiis incoloratis, primo continuis et erectis, levibus, elongato-ellipsoideis vel strobiliformibus, plerumque 12–19μ longis, 4–5.5μ crassis, ex apice vel rarius ex basi hypham germinationis brevem erectam emittentibus; hac hypha 1–3 corpora glutinosa 2.5–5.5μ crassa deinceps proferente. Sporae perdurantes continuae, ovoideae, flavidae, verrucosae

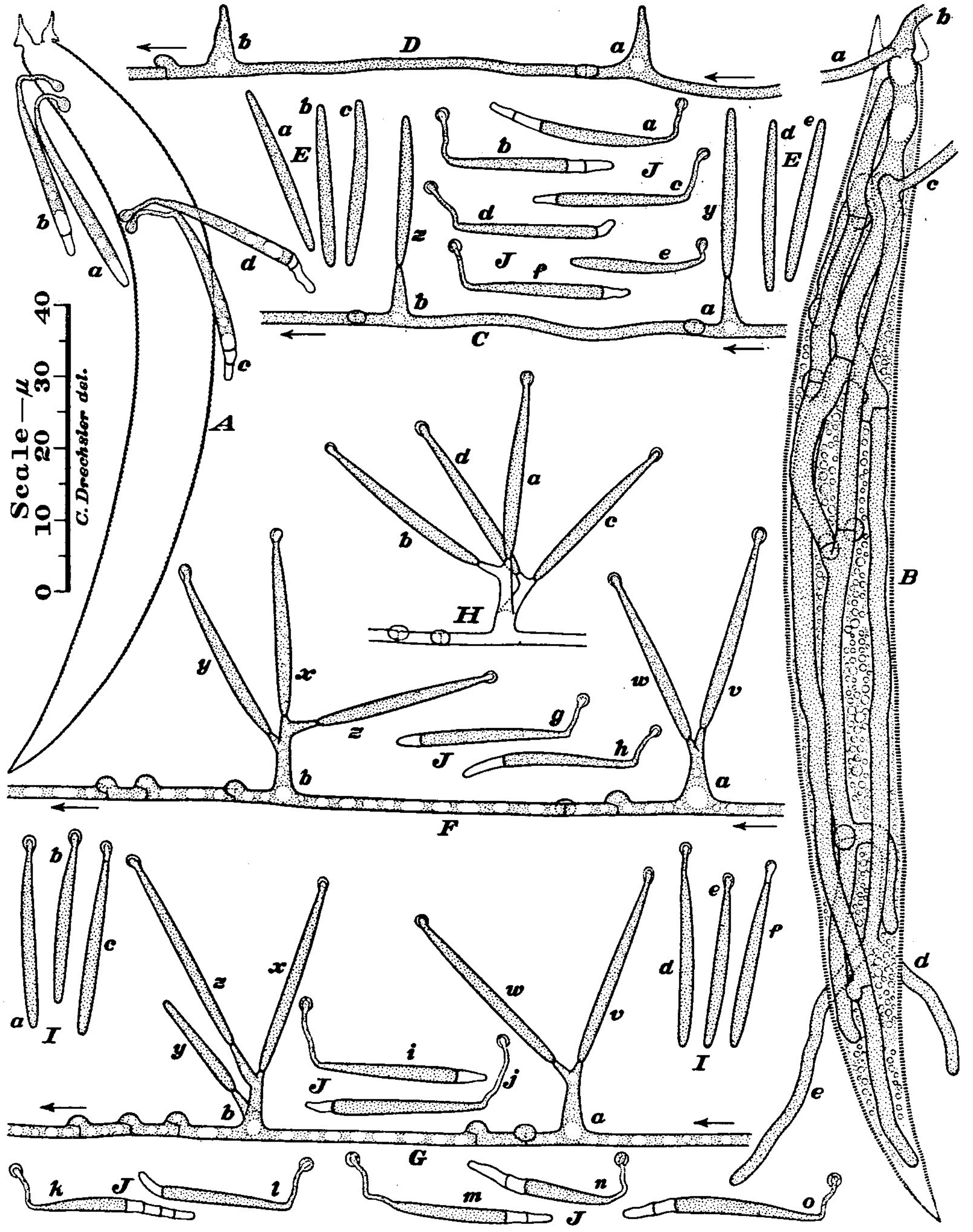

Fig. 2.—*Nematoctonus leptosporus,* drawn to a uniform magnification with the aid of a camera lucida; ×1,000 throughout. *A,* Young specimen of *Bunonema* sp. to which are attached four germinating conidia whose vacuolate condition indicates that each may be extending an infective hypha into the animal. *B,* Nematode host permeated with a mycelium from which five hyphae, *a-e,* have begun to grow externally. *C,* Portion of conidiophorous hypha with two sterigmata, *a* and *b,* each bearing a single conidium that shows no apical adhesive modification. *D,* Portion of conidiophorous hypha with two denuded sterigmata, *a* and *b.* *E,* Detached conidia, *a-e,* showing variations in size and shape before undergoing apical modification. *F, G,* Portions of conidiophorous hypha, each with two branching sterigmata, one of them, *a,* bearing two spores, *v* and *w,* while the other, *b,* supports three spores, *y-z.* *H,* Portion of conidiophorous hypha with a branched sterigma bearing four conidia, *a-d,* all modified at the apex. *I,* Conidia, *a-f,* each of which formed an adhesive knob at its apex before becoming detached from its sterigma. *J,* Conidia, *a-o,* that after falling on moist agar culture medium have each sent up a delicate apical process terminating in a small adhesive knob.

vel crebre echinulatae, 10–13μ longae, 5.5–7.5μ crassae.

Vermiculum nematoideum Rhabditis monhysterae adfinem enecans habitat in radicibus Lycopersici esculenti putrescentibus prope Beltsville, Maryland.

Assimilative hyphae colorless, irregularly branched, mostly 2 to 3.5μ wide, provided with clamp-connections, developing within living nematodes, after death of host animal giving rise externally to several conidiophorous hyphae. Conidiophorous hyphae colorless, simple or somewhat branched, at first usually ascending, later prostrate or festooned on the substratum, commonly 0.5 to 1.5 mm. long, 2.2 to 3.2μ wide, at moderate intervals (mostly 10 to 75μ) forming clamp-connections, producing solitary conidia or solitary resting spores sometimes directly on clamp-connections and at other times on sterigmata 0.5 to 5μ long, 1 to 2.5μ wide at the base, 0.6 to 1μ wide at the apex. Conidia erect, colorless, smooth, at first continuous, elongate-ellipsoid or strobiliform, mostly 12 to 19μ long, 4 to 5.5μ wide, before or after disjunction usually becoming partly evacuated of contents in giving rise at the apex or more rarely at the base to a short erect process whereon are borne successively 1 to 3 globose adhesive bodies 2.5 to 5.5μ in diameter. Resting spores aseptate, yellowish, ovoid, at maturity warty or strongly echinulate, measuring mostly 10 to 13μ in length and 5.5 to 7.5 μ in width.

Destroying a species of nematode belonging to the *Rhabditis monhystera* group, it occurs in decaying roots of *Lycopersicon esculentum* near Beltsville, Md.

The other species of *Nematoctonus* to be presented herein was found developing abundantly in Petri plates of maizemeal agar, which after being permeated with *Pythium* mycelium had been further planted with pinches of friable leaf mold taken from deciduous woods near Fairfax, Va., on November 10, 1942. In these cultures it subsisted exclusively on a species of *Bunonema* introduced with the forest refuse. Invasion of the small eelworm was manifestly initiated by continued germinative development of adhering conidia (Fig. 2, *A*, *a–d*), though owing to optical difficulties arising from globulose degeneration of the host tissues, not to mention further difficulties attributable to pronounced sculpturing of the host integument, the progress of mycelial advance could not be followed. However, after the granular materials had been largely appropriated the assimilative mycelium was revealed, though often only rather indistinctly, as a branching system of hyphae studded here and there with clamp-connections (Fig. 2, *B*). Usually before this somewhat transparent condition came about, conidiophorous filaments were being extended over the surface of the adjacent substratum (Fig. 2, *B*, *a–e*).

These filaments, while still elongating, give rise at moderate intervals to erect tapering sterigmata (Fig. 2, *C*, *a*, *b*; *D*, *a*, *b*), each bearing at its tip a slender, slightly tapering, rod-shaped conidium (Fig. 2, *C*, *y*, *z*) whose apex in the beginning shows no special modification (Fig. 2, *E*, *a–e*). Sometimes a sterigma grows directly from a clamp-connection but more often it arises some little distance backward from a clamp, that is, some little distance nearer the origin of the filament (Fig. 2, *C*, *a*, *b*; *D*, *a*, *b*). After the individual sterigma (Fig. 2, *F*, *a*; *G*, *a*) has produced its first conidium (Fig. 2, *F*, *v*; *G*, *v*), it ordinarily continues in its reproductive function by putting forth a short lateral spur on which a second conidium is formed (Fig. 2, *F*, *w*; *G*, *w*). Many sterigmata thereupon will put forth a second lateral spur, and thus will come to support three conidia (Fig. 2, *F*, *x–z*; *G*, *x–z*). In mature portions of conidiophorous hyphae some sterigmata can be found bearing as many as four conidia, each, of course, borne on a separate sterigmatic tip (Fig. 2, *H*, *a–d*).

Branched sterigmata bearing two or three conidia are nearly always to be found in proximity to a corresponding number of clamp-connections (Fig. 2, *F*, *G*). Apparently the clamps of later origin are formed in successively more distal positions.

Soon after they have been cut off by a basal septum, the conidia undergo noticeable germinative development. Those that remain supported on their sterigmata produce at the tip a globose knob consisting of a glandular part thinly surrounded with adhesive secretion (Fig. 2, *F–H*; *I*, *a–f*). Those that become detached and fall on a moist surface produce a similar adhesive knob terminally on a delicate, erect or ascending process extended from the tip (Fig. 2, *J*, *a–o*). The materials required for this germinative development are supplied through evacuation of protoplasm from the basal portion of the conidium. To separate the living portion of the spore from the emptied portion at least one retaining wall is laid down. In instances where presumably the movement of protoplasm takes place rather slowly, two (Fig. 2, *J*, *a*, *b*, *m*, *n*, *o*) or even four (Fig. 2, *J*, *k*) retaining walls may be laid down successively.

Comparable development of adhesive knobs on delicate processes arising from fallen conidia has not been noted in *Nematoctonus tylosporus*. The fungus differs further from *N. tylosporus* in its markedly stronger tendency toward production of conidia plurally on branching sterigmata. Its conidia, moreover, are appreciably narrower and longer than those of *N. tylosporus*, and, of course, pronouncedly narrower and longer than the conidia of *N. leiosporus* and *N. pachysporus*. A term having reference to its slender spores may therefore serve as an epithet sufficiently descriptive to set the fungus apart from the three known congeneric forms.

Nematoctonus leptosporus, sp. nov.

Hyphae assumentes incoloratae, plus minusve ramosae, plerumque 2–3.5μ crassae, in modum Hymenomycetum septato-nodosae, intra vermiculum nematoideum viventem crescentes, post mortem animalis aliquot fertiles hyphas extra emittentes; hyphis fertilibus incoloratis, saepius procumbentibus, modice septato-nodosis, vulgo 250–750μ longis, 1.6–2μ crassis, conidia ex erectis sterigmatis gerentibus; his sterigmatis inter se saepius 35–65μ distantibus, 5–10μ altis, basi 2.5–4μ crassis, sursum attenuatis, apice .5–1μ crassis, primo simplicibus, postea 1–3 ramusculos emittentibus, itaque vulgo 2 vel 3 etiam quandoque 4 conidia proferentibus; conidiis incoloratis, bacillaribus, sursum leviter attenuatis, utrinque obtusulis vel rotundatis, 21–28μ longis, 1.7–2.2μ crassis, primo continuis et protoplasmatis omnino repletis, mox in parte infera vacuis et apice tuberculo glutinoso circa 2μ crasso praeditis, postea tuberculum ejusmodi in apice hyphae erectae vel ascendentis 3–10μ longae .6μ crassae ferentibus.

Vermiculum nematoideum speciei Bunonematis necans habitat in humo silvestri prope Fairfax, Virginia.

Assimilative hyphae colorless, provided with clamp-connections, somewhat branched, mostly 2 to 3.5μ wide, developing within living nematodes, after death of host animal producing several conidiophorous hyphae externally; conidiophorous hyphae colorless, usually prostrate, commonly 250 to 750μ long, 1.6 to 2μ wide, forming clamp-connections at moderate intervals, giving rise to conidia on erect sterigmata; the sterigmata spaced mostly at intervals of 35 to 65μ, at first simple, commonly 5 to 10μ high, 2.5 to 4μ wide at the base, tapering upward, mostly 0.5 to 1μ wide at the tip, later usually putting forth 1 to 3 lateral spurs and by producing a conidium on each spur eventually coming to support 2 or 3 or sometimes even 4 conidia; the conidia colorless, staff-shaped, tapering slightly toward apex, somewhat obtuse or bluntly rounded at both ends, mostly 21 to 28μ long and 1.7 to 2.2μ wide, at first continuous, later often empty at the base and provided at the tip with a globose adhesive knob about 2μ wide, or after falling off producing such a knob terminally on an erect or ascending process, 3 to 10μ long and 0.6μ wide, that is extended obliquely or perpendicularly from the tip.

Parasitic on a species of *Bunonema* in leaf mold near Fairfax, Va.

With respect to outward shape the knob-bearing outgrowth commonly produced by the conidium of *Nematoctonus leptosporus* after falling on a moist surface offers curious similarity to the empty basal appendage on the conidium of *Euryancale sacciospora* Drechsl. (3) as well as to the proximal portion of the conidium of *Harposporium oxycoracum* Drechsl. (4). This similarity would seem in large measure illustrative of convergence, since the three fungi, remote from one another taxonomically, all subsist as obligate parasites on nematodes of the genus *Bunonema*. The adaptive modifications here concerned may well have been developed to facilitate attachment of the conidia to the strongly sculptured integument so characteristic of the host animals in their adult condition.

In *Nematoctonus pachysporus* the development of adhesive bodies plurally, together with the frequently pronounced exhaustion of the spore, makes for an appearance not wholly unlike that offered in the development of basidiospores on basidia. The homologies thus suggested can not readily be dismissed until adverse cytological evidence has been brought foreward, or until structures have been discovered more closely corresponding to basidia than any I have observed hitherto. The plural adhesive bodies, it is true, are almost certainly of the same character as the single adhesive knobs formed in the three congeneric species; but the possibility remains that these single knobs, however commonplace their appearance, might yet represent abortive basidiospores modified for adhesion. Nevertheless, the thin-walled aerial spores still seem best interpretable as conidia, especially since in their manner of formation they offer strong parallelism with the binucleate conidia described by Nobles (5) as being produced on clamp-bearing mycelia of *Corticium incrustans* Höhn. & Litsch. If the four parasitic species so far described all produce typically straight conidia, the Hawaiian nematode-capturing fungus to which reference was made earlier (4, p. 780) and which almost certainly is intimately related to the parasitic species, produces conidia that resemble those of *C. incrustans* in being of curved allantoid shape.

Their constant production of clamps rather definitely removes all five of the fungi habitually subsisting on eelworms from close kinship with *Septobasidium* Pat., a large genus of basidiomycetes whose parasitism on scale insects, affirmed by Reinking (6) in 1919, has more recently been set forth in detail by Couch (2). On similar grounds they must be considered taxonomically remote from *Uredinella* Couch, likewise a genus of basidiomycetes parasitic on scale insects, since at least in *U. coccidiophaga* Couch (1), just as in all species of *Septobasidium*, clamp-connections are absent.

LITERATURE CITED

(1) COUCH, J. N. *A new fungus intermediate between the rusts and* Septobasidium. Mycologia **29**: 665–673. 1937.

(2) ———. *The genus* Septobasidium. 480 pp. Chapel Hill, N. C., 1938.

(3) DRECHSLER, C. *Five new Zoopagaceae destructive to rhizopods and nematodes.* Mycologia **31**: 388–415. 1939.

(4) ———. *Some hyphomycetes parasitic on free-living terricolous nematodes.* Phytopathology **31**: 773–802. 1941.

(5) NOBLES, MILDRED K. *Production of conidia by* Corticium incrustans. Mycologia **29**: 557–566. 1937.

(6) REINKING, O. A. *Diseases of economic plants in southern China.* Philippine Agr. **8**: 109–135. 1919.