ZOOSPORE DEVELOPMENT FROM OOSPORES OF PYTHIUM ULTIMUM AND PYTHIUM DEBARYANUM AND ITS RELATION TO ROOTLET-TIP DISCOLOR ATION

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Oospores of Pythium ultimum Trow taken from maizemeal-agar plate cultures 3 months old produced zoospores in moderate abundance within 18 hours after they were transferred to a thin layer of water in Petri dishes kept at a temperature of 10°C. During the 3 months a large proportion of the cospores had lost the internal organization distinctive of their resting condition; the large central reserve globule together with the conspicuous refringent body having disappeared, and the oospore wall having become reduced, through resorption of an inner layer, to somewhat less than half its original thickness. In the densely granular texture and obscurely vacuolate character of their protoplasmic contents, as also in the appearance of their diminished envelopes, the after-ripened oospores showed general resemblance to globose asexual spores (conidia) formed abundantly on the mycelial hyphae of the species. At 25° C they germinated promptly by production of a germ hypha, even without addition of fresh water, while they were undergoing microscopical examination after their removal to a glass slide. On shallow irrigation at a temperature of 10° C an evacuation tube mostly 3 to 6 µ wide above its frequently broadened base was pushed through the loosely surrounding oogonial envelope. After it had attained a total length of 10 to 40 µ the tube formed a hyaline cap of dehiscence, which soon yielded to permit the protoplasmic contents to migrate into a vesicle for conversion usually into 8 to 10 motile zoospores. After escaping from the vesicle and swimming about for a time the zoospores came to rest and rounded up into cysts mostly 8 to 10 µ -- rarely up to 13 \mu - in diameter. These commonly germinated by putting forth 1 or 2 germ hyphae mostly 2 to 3 µ wide, but rather often, again, they gave rise individually to a secondary motile zoospore by extending an evacuation tube 2.5 to 4 µ long and 2 to 2.5 µ wide.

Zoospore formation has not hitherto been recorded for Pythium ultimum. The operation of this familiar species and of P. debaryanum Hesse as damping-off parasites in seedling beds has little obvious relation to development of swarmers, since here spread of infection evidently is accomplished for the most part by rangy extension of extramatrical mycelium from one plant host to another. However, the frequent occurrence of P. ultimum, especially during the cooler wet portions of the growing season, as the cause of blackening of rootlet tips in various crop plants as well as in many uncultivated phanerogams, appears rather strongly suggestive of infection by swarmers. It seems likely that under natural conditions swarm-spore development in P. ultimum may take place more abundantly through germination of after-ripened cospores than through reproductive germination of sporangia (conidia) borne directly on mycelial hyphae; the behavior of the fungus inviting comparison

with such congeneric forms as P. anandrum and P. ostracodes. Addition of partly decayed leaf mold to maizemeal-agar plate cultures after the substratum has been well permeated by the fungus often results in very pronounced increase in number of cospores formed. It appears possible that leaf mold supplies substances that besides permitting more cospores to develop make them more capable of giving rise to swarmers.

The same treatment that evoked zoospore production in cospores of Pythium ultimum was successful, likewise, in evoking such production in cospores of P. debaryanum taken from maizemeal-agar plate cultures 2 months old. The evacuation tube extended through the oogonial wall is here noticeably shorter, usually measuring only 7 to 15 μ in total length. After discharge of the protoplasmic contents into a vesicle the persistent outer layer of the cospore wall was found surrounding a structurally separate sporangial membrane more often than in P. ultimum. Oospores of average size, here as in the case of P. ultimum, gave rise commonly to 8 or 9 motile swarmers which after coming to rest and encysting measured usually 8 to 9 µ in diameter; the encysted bodies here, too, sometimes giving rise individually to a motile secondary swarmer. Zoospore development in P. debaryanum, of course, does not require a temperature as low as 10°C, nor any substances not usually present in maizemeal-agar containing in suspension a moderate quantity of finely divided maizemeal; for often when microscopic examinations are carried out on sparingly moistened slabs excised from maizemeal-agar cultures 15 days old many of the subspherical asexual sporangia in favorable positions will promptly put forth an evacuation tube to give rise to motile swarmers, though the laboratory may have a temperature near 18° C. During prolonged wet periods P. debaryanum, like P. ultimum, can often be isolated in quantity from blackened rootlet tips of various crop plants. The dispersed distribution of rootlet tips at varying depths in the soil would seem to offer more scope for infection by zoospores than for infection by extramatrical mycelium.

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