moved to a 22 mm², 0-size coverslip. The coverslip was mounted off-center on a 12-mm diam brass stub coated with clear fingernail polish, and the specimens were dried for scanning.

A Denton DV-502 high vacuum evaporator operating at 3 x 10⁻⁵ Torr was used to spread a standard layer of gold on the specimens. A Jeol JSM-2 SEM with an accelerated voltage of 35 kv scanned the parasitized larvae. A light compound microscope was also used to look at the specimens. Kodak Tri-X Ortho film No. 4163 was used to make the photographs.

SEM showed root-knot larvae parasitized by 7-12 D. penetrans spores attached to the nematode cuticles (Fig. 1). The spores were rounded above and flattened below. A round cap was part of the mature spore (Fig. 2, 3). The spores averaged 4.3 μm diam by 1.6 μm in depth, and the spore cap 2.8 μm diam by 0.4 μm in depth. Except for the wrinkled lines at their juncture, the spore and its cap were smooth. Pressure on the cuticle probably caused the flattened spore bottom.

Buds and tubes were seen on the spore bases (Fig. 3). The buds apparently form tubes that penetrate the host cuticle. SEM photos of buds and tubes on mature spores confirm the observations of Allen (1).

SEM photos of mature spore forms and caps generally confirm descriptions of D. penetrans provided by Thorne (5). Spore morphology is like that of a protozoan; spore size was greater than Bacillus spp. cells. Spores did not have the crescent shape of Bacillus popilliae sporangia nor of any known bacterium. Endospores or rod-shaped spores were not seen, although bacteria were noted near parasitized larvae (Fig. 1). Hyphae, thalli, and other fungal structures were not found. For these reasons, we prefer to keep this parasite in the protozoa.

LITERATURE CITED

Comparative Biology of the Wyoming and Louisiana Populations of Reesimermis nielseni, Parasitic Nematode of Mosquitoes

JAMES J. PETERSEN

The mermithid nematode, Reesimermis nielseni Tsai and Grundmann, was first described from collections made at an elevation of 2450 m near Lone Tree, Wyoming in May and June, 1965 and 1966 (5), and was reported to infect larvae, and occasionally pupae and adults, of six species of univoltine mosquitoes. Concurrently, a second population of mermithids was found in four species of multivoltine mosquitoes at an elevation of 7.5 m near Lake Charles, Louisiana (1). This latter population later proved to be the same species as the Wyoming population of R. nielseni (2). However, the lack of living material of the Wyoming population at that time prevented comparative studies on the biology of both populations.

In May 1974 (through the courtesy of Lewis Nielsen, University of Utah, and Jim Ross, University of Waterloo, Canada), I obtained about 200 female and 700 male post-parasitic juveniles from the Lone Tree site.

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The nematodes were separated into six groups of 25 females and 50 males each and placed in small glass dishes containing sterile sand. Two groups each were then held at 17, 20, and 23 C.

Observations on development, though limited, indicated that the Wyoming and Louisiana populations were different. After 2 weeks at 17 C, all of the Wyoming R. nielseni were adults and some were ovipositing. At 4 and 7 weeks, 20 and 100% of the surviving females, respectively, had completed oviposition. Development was slower at 20 C than at 17 C since all the males and 67% of the females were adults after 2 weeks, and 80% of the females had completed oviposition at 7 weeks. After 7 weeks at 23 C, all the Wyoming nematodes were mature, but none had laid eggs. After being transferred to 17 C, some females eventually oviposited, although about 20% of the females failed to oviposit after 10 months. The Wyoming preparasites hatched best in cultures held at 17 C, and peak production occurred around 18-20 weeks, with small hatches extending over 10 months.

When the Louisiana population was held in a similar environment, the rate of development increased with an increase in temperature. At 17 C, 11-12 weeks were required for 90% of the females to mature, and 23 weeks were required for the first females to complete oviposition. At 23 C, the nematodes matured in 2-3 weeks, and the females completed oviposition in 3-7 weeks. Thus at 17 C, 95% of the Wyoming females had completed oviposition in 7 weeks in comparison with the Louisiana females which required more than 1 year.

*Culex pipiens quinquefasciatus* Say and *Culiseta inornata* (Williston) have been reported as excellent mosquito hosts of the Louisiana population of *R. nielseni*, and no host resistance is known (3, 4). However, attempts to propagate the Wyoming population produced high levels of infection, a good sex ratio of postparasites, and no host resistance. Wyoming nematodes also were found to develop in pupae of *C. p. quinquefasciatus*, a condition that did not occur with the Louisiana nematodes.

Cross mating was limited by the amount of Wyoming material available. Six groups of 25 postparasitic juvenile females from the Louisiana population were placed in small glass dishes containing moist sand and treated as follows: 50 postparasitic juvenile males from the Louisiana population were added to each of two containers; 50 postparasitic juvenile males from the Wyoming population were added to each of two containers; and no males were added to two containers. The cultures were examined at 2, 3, 7, and 10 weeks. The two cultures containing both sexes of the Louisiana population averaged 33% mating and 12% egg development in the females after 2 weeks, and 42 and 85% oviposition after 3 and 7 weeks, respectively. No egg development was observed in the cultures without males or in the cultures containing the Wyoming males. No mating occurred between the Wyoming males and the Louisiana females.

The limited data suggest that the two populations of *R. nielseni* develop at different rates (especially at low temperatures), have different effects on a given host, and will not cross mate in the laboratory. The two populations appear to differ sufficiently biologically to consider them distinct species or at least subspecies. Therefore, a more detailed taxonomic study of the two populations should be made, especially since material from the Wyoming population is now available.

**LITERATURE CITED**


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