

IDENTIFICATION OF MAJOR *MELOIDOGYNE* SPECIES EMPLOYING ENZYME PHENOTYPES AS DIFFERENTIATING CHARACTERS

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The use of enzyme phenotypes for species identification and for the elucidation of the taxonomic and phyletic relationships of organisms has become quite popular during the last 20 years (16, 17, 21). Enzyme studies of root-knot nematodes were initiated in 1971 (8). This early work indicated clearly that phenotypes of some enzymes are characteristic of certain species or certain populations within a species. Since then, about 30 enzymes have been detected in various *Meloidogyne* populations (Table 1), but only a few of them have been investigated extensively and assessed for their usefulness as taxonomic characters (4, 10, 14). Dalmasso and Bergé (4) conducted an extensive survey involving 83 populations of 5 *Meloidogyne* species. These investigators used a miniaturized extraction and electrophoretic procedure that enabled them to determine the enzyme phenotype of individual females of root-knot nematodes, thus increasing their chances of detecting enzymatic variation within populations. In more recent work, the thin-slab electrophoretic procedure provided even more accurate evaluations of enzyme phenotypes of these nematodes (14). Comparisons of 20 or more nematode-protein extracts in the same slab gel, under exactly the same electrophoretic conditions, can reveal even slight isozymic variations that may exist within and between populations of a species or between species (10).

It is believed that, with standardization of the electrophoretic procedures (protein extraction, electrophoresis, and staining), enzyme phenotypes can provide a number of advantageous and objective characters for the identification of certain species or biological forms of root-knot nematodes and for taxonomic characterization of new forms.

Because the early data about certain enzymes of root-knot nematodes proved to be so promising (1, 4, 8, 12, 14), an extensive investigation was undertaken in collaboration with the International *Meloidogyne* Project in an effort to determine the practicality and reliability of the use of enzyme phenotypes in the identification of root-knot nematodes on a worldwide basis (10). About 300 populations were selected from a collection of more than 600 which had been studied cytologically (see Chapter 9) and with regard to host range and perineal patterns of adult females. Tentative identifications to species had been made on the basis of such information and, for some populations, on supplemental studies of male stylets and other morphological features of females, males and juveniles. The 300 populations selected for the biochemical study had originated from 65 countries and 20 states in the U.S.A., and represented 16 *Meloidogyne* species including the four major species (*M. arenaria*, *M. hapla*, *M. incognita*, and *M. javanica*) and most of the known cytogenetic and physiological (host range) races of these species. Studies have been completed on four enzymes (non-specific esterases, malate dehydrogenase, superoxide dismutase, and glutamate-oxaloacetate transaminase) that gave promising results in the early stages of this work. Of those, esterases proved to be the most useful enzyme for the differentiation of the major *Meloidogyne* species. This conclusion confirmed previous findings by other investigators who had studied smaller numbers of populations, not representing all of the geographic, cytogenetic, and physiological variation known to exist within each species (1, 4, 8, 12, 14). Superoxide dismutase was also useful in the characterization of certain major

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