Incidence and Distinguishing Characteristics of Meloidogyne chitwoodi and M. hapla in Potato from the Northwestern United States[1]

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Abstract: From September 1980 to June 1981, a survey was conducted in the major potato growing regions of northern California, Idaho, Nevada, Oregon, and Washington to determine the distribution of *Meloidogyne chitwoodi* and other *Meloidogyne* spp. *Meloidogyne chitwoodi* and *M. hapla* were the only root-knot nematode species detected parasitizing potato in all the states surveyed. *Meloidogyne chitwoodi* occurred alone in 83% of the samples and *M. hapla* in 11%, with 6% of all samples containing both species. The greater incidence of *M. chitwoodi*, as compared to *M. hapla*, may be due to the cool growing season encountered in 1980 (which favored *M. chitwoodi* but not *M. hapla*) and to the increased acreage of small grains (which are good hosts for *M. chitwoodi* but not *M. hapla*) planted in rotation with potato. Differentiation between these two species can be determined by a differential host test, perineal patterns of mature females, and shape of the tail tip and of the tail hypodermal terminus of L_2 juveniles.
Key words: Meloidogyne chitwoodi, M. hapla, potato.

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Potato (*Solanum tuberosum* L.) is an important agricultural commodity of the Pacific Northwest. In 1980, four of the five leading states in potato production were from northwestern United States, with Idaho and Washington ranked first and second, respectively (1). Root-knot nematodes (*Meloidogyne* spp.) are a severe problem in potato production because they reduce the quality of the potato tuber (9).

Until recently, the northern (*Meloidogyne hapla* Chitwood), southern (*M. incognita* [Kofoid and White] Chitwood), and Thames (*M. thamesi* Chitwood) root-knot nematodes were the only species recognized as parasitizing potatoes in the Pacific Northwest (7). *Meloidogyne hapla* was reported widespread in Idaho, Oregon, and Washington; *M. incognita* in limited regions of Oregon and Washington; and *M. thamesi* in limited regions of Washington (3,7,10). In 1978, a previously unreported root-knot nematode, the Columbia root-knot nematode (*M. chitwoodi* Golden et al.) was found in potato tubers from Washington and Idaho (5,10). Examination of tubers from several production areas revealed that this nematode was present along the Columbia River Basin of Washington, the upper Snake River regions of Idaho, and isolated areas of northeastern Oregon and northwestern Washington (10). This survey indicated that *M. chitwoodi* was more widespread than initially presumed. In September 1980, a more extensive survey of the major potato growing regions of the Pacific and Northwestern United States was conducted to determine more precisely the incidence of *M. chitwoodi* and other *Meloidogyne* spp. This paper reports results of that study.

MATERIALS AND METHODS

Potato tubers and/or soil samples containing *Meloidogyne* spp. were collected from the major potato growing regions of northern California, Idaho, Nevada, Oregon, and Washington. From September 1980 to June 1981, tuber samples were obtained from processing facilities as they were being placed in or removed from storage. When regulatory inspectors detected root-knot nematode infection or symptoms on tubers, three or four of the tubers from that infected lot were removed, location to nearest town recorded, and the sample mailed to the nematology laboratory at Prosser, Washington. During the survey, a limited number of soil samples were obtained from a private consultant.

Root-knot nematode populations were increased and maintained in a greenhouse by removing 15–20 egg masses from an in-

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fected tuber and placing them on the roots of two tomato seedlings *Lycopersicon esculentum* Mill. 'Columbian'. Each tomato plant was planted in a separate 10-cm-d plastic pot containing methyl bromide fumigated sandy loam soil. Infested soil samples were placed in pots and planted directly with a tomato seedling.

Eggs were extracted from the tomato roots after 55 days using the method of Hussey and Barker (6) to prepare inocula for a differential host test. Inoculations were made by pipetting 2,000 eggs per pot into depressions made in the soil around each host's hypocotyl. Plants used in our standard differential host test included pepper (*Capsicum frutescens* L. 'California Wonder'), watermelon (*Citrullus vulgaris* Shrad 'Charleston Grey'), and wheat (*Triticum aestrum* L. 'Nugaines'). The North Carolina differential host test (11) was used whenever a population did not appear to conform to our standard differential test (Table 1). Treatments consisted of inoculating two seedlings of each cultivar, and the pots were randomly arranged on greenhouse benches. Fiberglass dividers (112 × 36 cm) separated individual populations on the same bench in order to prevent cross contamination during watering. Ambient greenhouse temperatures ranged from 24 to 26 C. Plants were watered daily and fertilized every 2 wk.

After 55 days, the plants were harvested and the roots washed in tap water and stained with phloxine B (150 mg/l for 15 min.) (2). Root galls and egg masses on each root system were counted, and plants were rated as a host or nonhost. The following rating system was used: 0 = no galls or egg masses, 1 = 1–2 galls or egg masses, 2 = 3–10 galls or egg masses, 3 = 11–30 galls or egg masses, 4 = 31–100 galls or egg masses, and 5 = > 100 galls or egg masses (11). A root system rating of 0–2 was considered a non to poor host. After the root rating was determined, eight mature females and their egg masses were removed from each root system. Morphological criteria, in conjunction with the differential host test, were used to identify the root-knot population to species. The morphological criteria were the length, tail tip shape, and the shape of the tail hypodermal terminus from 20 freshly hatched second-stage juveniles (L_2), perineal patterns, stylet length, and presence/absence of median bulb vesicles of mature females. Juveniles were obtained by incubating egg masses in water for 24 h at 25 C. The survey was terminated at the end of June 1981.

RESULTS

Meloidogyne chitwoodi and *M. hapla* were the only species of root-knot nematode detected in our survey. Both species were found in major potato producing regions of the states sampled, occurring either along a major waterway used for irrigation or in isolated areas in northern California, southern Oregon, and northwestern Washington (Fig. 1). *Meloidogyne chitwoodi* occurred alone in 83% of the samples, *M. hapla* alone in 11%, and 6% of the samples contained both species (Table 2). *Meloidogyne chitwoodi* usually occurred as a single species in the samples, while *M. hapla* was primarily present mixed with *M. chitwoodi,*

Table 1. Differential host test for *Meloidogyne* spp.*

Meloidogyne species	Differential host†						
	Wheat	Pepper	Water-melon	Tobacco	Cotton	Peanut	Tomato
M. chitwoodi	+	–	–	–	–	–	+
M. hapla	–	+	–	+	–	+	+
M. incognita‡	+	+	+	–	–	–	+
M. arenaria‡	+	+	+	+	–	+	+
M. javanica	+	–	+	+	–	–	+

*+ = host; – = nonhost. See reference # 11.

†Wheat, 'Nugaines'; pepper, 'California Wonder'; watermelon, 'Charleston Grey'; tobacco, 'NC 95'; cotton, 'Deltapine 16'; peanut, 'Florrunner'; tomato, 'Rutgers.'

‡Refers to race one of *Meloidogyne* spp. See reference # 11 for race differentiation within a species.

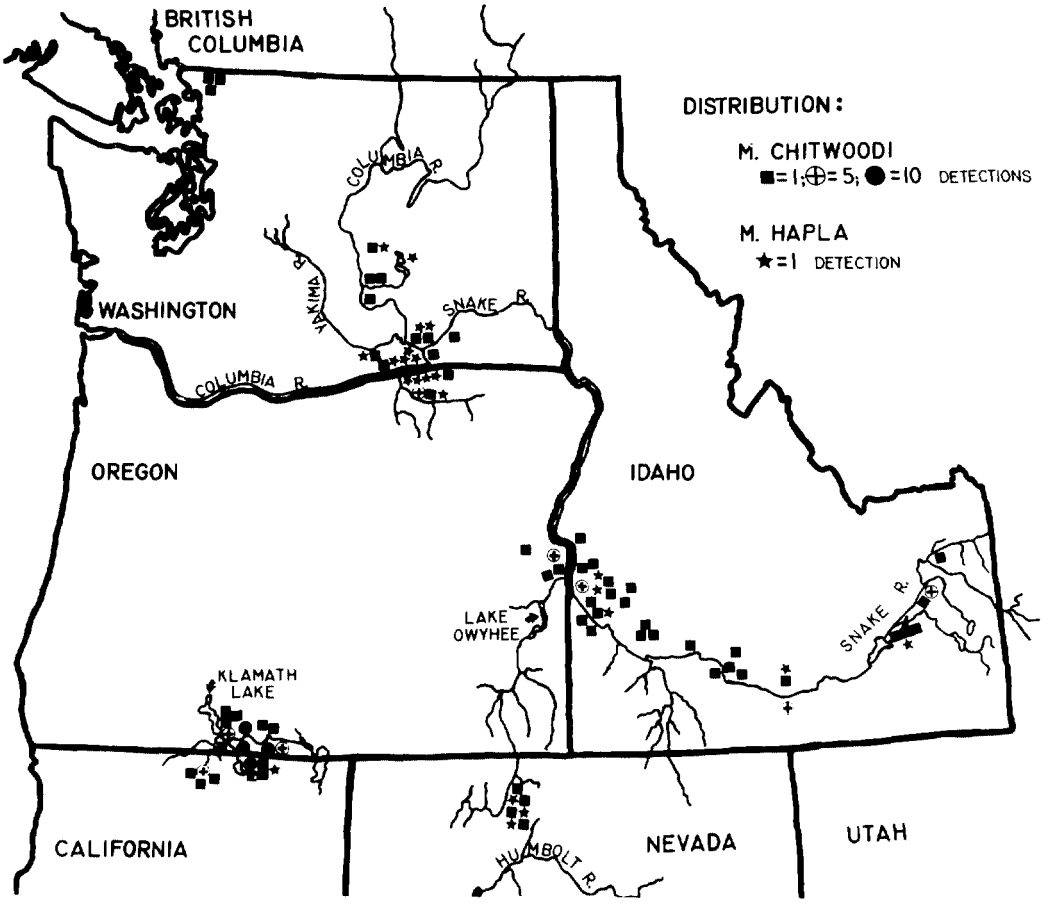


Fig. 1. Distribution of *Meloidogyne chitwoodi* and *M. hapla* on potato in five northwestern states surveyed, September 1980 to June 1981.

except in a few samples received from Washington and northeastern Oregon. *Meloidogyne chitwoodi* was the predominant root-knot nematode found in all states sampled except Nevada.

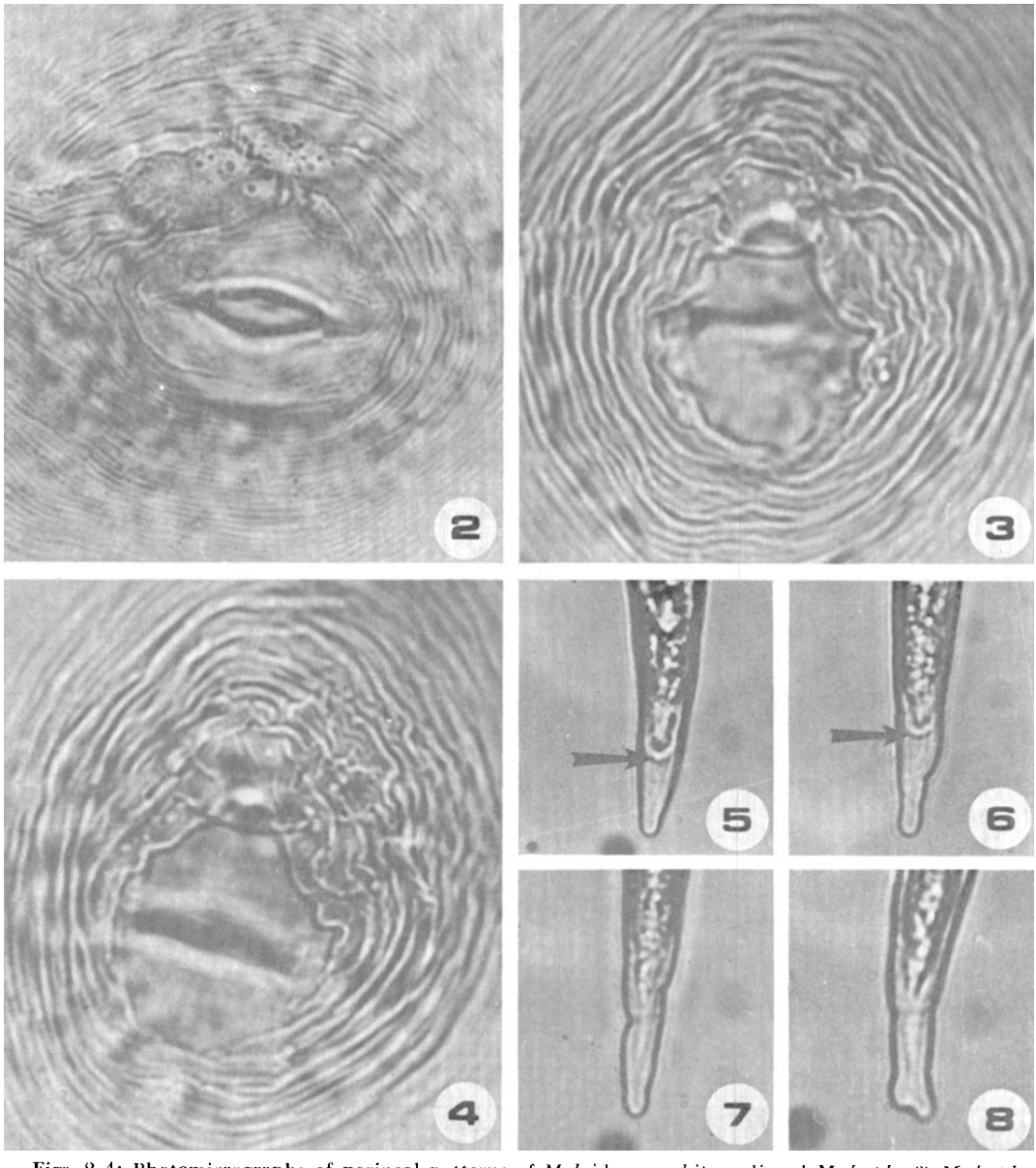
'California Wonder' pepper and 'Nugaines' wheat are excellent differential hosts for *M. chitwoodi* and *M. hapla*. Questionable results for species identification occurred in only one sample which was received from northern California. However, after repeating the original differential host test in conjunction with the North Carolina test and using morphological characteristics, it was confirmed as *M. chitwoodi*.

Morphological characteristics were used

Table 2. Occurrence of *Meloidogyne chitwoodi* (Mc) and *M. hapla* (Mh) alone or together in potato tubers and soil samples collected from the northwestern United States, September 1980 to June 1981.

State	Number and type of samples received			Number of samples infested with		
	Tuber	Soil	Total	Mc	Mh	Mc + Mh
Washington	41	3	44	25	17	2
Idaho	40	0	40	36	0	4
Oregon	72	4	76	71	3	2
California	23	0	23	22	0	1
Nevada	0	4	4	1	0	3
Total	176	11	187	155	20	12

to identify each root-knot nematode population to species. The most reliable characters differentiating *M. chitwoodi* from *M. hapla* included the adult females' perineal pattern and L_2 juvenile tail tip shape and shape of the posterior end of the hypodermis in the tail. Perineal patterns of these two species are distinctly different. Patterns of *M. hapla* (Fig. 2) were usually delicate without distinct lateral lines, the striae were smooth and parallel, and the arch was flattened to more or less rounded. Punctations were present around the tail region, whereas wing formation and number varied. Perineal patterns of *M. chitwoodi* (Fig. 3) had arches that were usually oval with thick, twisted striae in and around the anal region, and an anus and vulva set in a depression



Figs. 2–4: Photomicrographs of perineal patterns of *Meloidogyne chitwoodi* and *M. hapla*. 2) *M. hapla*. 3) *M. chitwoodi*. 4) *M. chitwoodi* pattern with higher arch (× 400).

Figs. 5–8. Photomicrographs of L_2 juvenile tails of *Meloidogyne chitwoodi* and *M. hapla*. 5) *M. chitwoodi* tail. 6) Variation of *M. chitwoodi* tail, arrow indicates bluntly rounded hypodermal terminus of the tail. 7) Tapered tail of *M. hapla*. 8) Toe-shaped tail of *M. hapla* (note tapered hypodermal terminus of *M. hapla* tails) (× 640).

(sunken). More variation occurred within patterns of *M. chitwoodi* populations than within *M. hapla*. Some patterns of *M. chitwoodi* had higher arches (Fig. 4), yet the striae were typical of *M. chitwoodi*.

Tail tip shape and the shape of the tail hypodermal terminus of freshly hatched heat-relaxed L_2 juveniles were distinctly different in the two species. The slightly tapered and bluntly rounded tail tip of *M. chitwoodi* L_2 juveniles (Fig. 5) was previously described and illustrated as being characteristic of this species (5). However, some variations between and within populations were noted. The tail tip of some L_2 juveniles were not always bluntly rounded, but sometimes deformed, exhibiting a slightly clavate tail tip (Fig. 6). However, the perineal pattern of the female from which these L_2 juveniles were obtained was typical for *M. chitwoodi*, as was the parasitism in the differential host test. One tail characteristic that was consistent among all *M. chitwoodi* L_2 juveniles was the shape of the tail hypodermal terminus, which was bluntly rounded even when variations occurred in the tail tip (Figs. 5, 6; see arrows).

Two tail tip shapes were encountered between and within populations of *M. hapla*. The tail tip was either uniformly tapered to a bluntly rounded tip (Fig. 7) or 'toe-shaped' (Fig. 8). The shape of the tail hypodermal terminus was more tapered (Figs. 7, 8) than in *M. chitwoodi*.

DISCUSSION

Meloidogyne chitwoodi and *M. hapla* were the only root-knot nematode species found in the major potato growing regions of the Northwest. Although *M. incognita* had previously been reported in Washington, it was not detected in this survey. Slides (courtesy L. R. Faulkner) containing perineal patterns of nematodes identified as *M. incognita* in 1962 have been reexamined and are now identified as *M. chitwoodi*. Failure to detect other root-knot species was not surprising since *M. chitwoodi* and *M. hapla* have a relatively high tolerance for cooler soil temperatures compared to the other three most common root-knot species—*M. arenaria, M. incognita,* and *M. javanica*—in the United States (9,11). In four of the five states surveyed, the daily annual average temperature from 1969 to 1979 taken in seven locations was 10.5 C; the daily maximum and minimum temperatures were 17.2 and 3.6 C, respectively (Table 3). This would result in relatively low soil temperatures. These relatively low soil temperatures help explain why these two species predominate in the Northwest. *Meloidogyne chitwoodi* is more of a problem than *M. hapla* on potato in years when spring temperatures are unusually cool. Santo and O'Bannon (9) have shown that *M. chitwoodi* reproduces more on potato at lower temperatures than *M. hapla*. Therefore, *M. chitwoodi* would tend to have more generations than *M. hapla* during the growing season, resulting in earlier tuber infection and a greater reduction in tuber quality. The low temperatures in the spring of 1980 could account for the higher incidence of *M. chitwoodi* found in potato tubers in this survey. Nematode analysis of soil samples from Idaho, Nevada, Oregon, and Washington (Santo, unpublished) show that *M. hapla* occurs more frequently than indicated by this survey where the majority of the samples were potato tubers.

Crop rotation plays an important role in determining which of these two root-knot nematode species predominates in a particular field. In the Pacific Northwest, alfalfa, wheat, other small grains and cereals, peppermint, and sugarbeet are the principle crops rotated with potato. Wheat,

Table 3. Daily annual average temperature at seven locations in Northwestern United States, 1969–79.

State and station	Temperature (C)		
	Maximum	Minimum	Average
Idaho			
Boise	17.1	4.2	10.7
Pocatello	13.7	0.9	7.9
Oregon			
Medford	19.2	4.8	12.0
Pendleton	17.3	5.3	11.3
Nevada			
Winnemucca	19.2	0.1	9.7
Washington			
Quincy	16.6	2.7	9.7
Walla Walla	17.4	6.9	12.2
Mean	17.2	3.6	10.5

other small grains, and cereals are good hosts for *M. chitwoodi* but poor to non hosts for *M. hapla* (10). Alfalfa was the primary crop rotated with potato in most Northwestern states (particularly Idaho and Washington), but since it is a good host for *M. hapla* it is not often used in the rotation. However, alfalfa is a poor to non host to *M. chitwoodi* (10). Peppermint is a poor host for *M. chitwoodi* but a good host for *M. hapla,* whereas sugarbeet is a good host for both nematodes (3,8). Knowing the cropping history of a field infested with *Meloidogyne* spp. could help determine which of these two species is predominant. Six percent of all samples contained both species, and whenever this occurred one species always predominated. The previous cropping sequence was probably the determining factor.

Results from this survey suggest that dissemination of *M. chitwoodi* and *M. hapla* in the potato growing regions of the Northwest occurred principally by two means; reused irrigation water (canal and river water), and infected seed potato. Irrigation water has been shown to be an excellent source for disseminating *Meloidogyne* L_2 juveniles (4). It has been estimated that the Yakima Valley and Columbia Basin of Washington receive approximately 0.144×10^6 to 15.362×10^6 plant parasitic nematodes per hectare per year in irrigation water alone. Nematode contaminated irrigation water would explain the distribution of *M. chitwoodi* and *M. hapla* along the major waterways used in irrigating potato fields in Idaho, Oregon, and Washington. Root-knot infected seed potato would account for the occurrence of both nematodes in areas removed from major waterways, such as northern California, southern Oregon, and northwestern Washington (Fig. 1). Three tuber samples infected with *M. chitwoodi* came from potato fields grown for seed, two in northwesten Washington and one in eastern Idaho.

Since only *M. chitwoodi* and *M. hapla* were detected in the survey, the perineal patterns and characteristics of L_2 juvenile tails were the easiest and most reliable criteria used to differentiate between these two species. Vesicle-like structures in the median bulb of *M. chitwoodi* females were less reliable because their number varied from 1 to 15. Other characteristics examined included total length of L_2 juveniles; position of the excretory pore in females, which is posterior to the stylet knobs for both species; the DGO, which is reported as being 4.3 μ and 5–6 μ for *M. chitwoodi* and *M. hapla* females, respectively; and female stylet lengths which are 12 μ and 13μ for *M. chitwoodi* and *M. hapla,* respectively. Though these morphometric criteria are essentially similar in *M. chitwoodi* and *M. hapla,* they would be useful in separating these two species from many other *Meloidogyne* spp.

The differential host test was a useful tool in determining which species were present. However, since races of *Meloidogyne* spp. are known to occur, the host test should not be the sole basis for species identification. Taylor and Sasser (11) emphasize that accuracy in identifying root-knot nematodes to species is increased as more criteria are used.

Meloidogyne chitwoodi and *M. hapla* were two root-knot nematode species found in major potato growing regions of northwestern United States in 1980-81. Knowledge of which species is present in a field will enable the grower to implement the proper rotation sequence in addition to other control practices, thereby suppressing root-knot nematode soil populations enough to increase potato tuber quality.

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