Meloidogyne fallax

Scientific Name

Meloidogyne fallax (Karssen, 1996)

Synonyms:

Meloidogyne chitwoodi B-type

Meloidogyne fallax was named for its morphological similarity to *M. chitwoodi* and was initially considered a new race of *M. chitwoodi*.

Common Name

False Columbia root-knot nematode

Type of Pest Plant-pathogenic nematode

Taxonomic Position

Class: Secernentea, **Order:** Tylenchida, **Family:** Meloidogynidae

Reason for Inclusion in Manual

Solanaceous pest; Former additional pests of concern listing; unconfirmed U.S. detection in California in 2012.

Pest Description

From Karssen (1996):



<u>Second-Stage Juveniles (J2s)</u>: J2s are 0.403 mm \pm 0.015 mm in length. Other measurements include 0.014 mm in diameter at the widest point, a stylet length of 0.010 mm, a metacorpus valve length of 0.004 mm, and a tail length of 0.049 mm \pm 0.002 mm.

The J2 body is moderately long and vermiform, with tapering at both ends, but the taper is more pronounced on the posterior end (Fig. 1). The body annules are small but distinct. The lateral field has four incisures that are not areolated. The head region is truncated and slightly set off from the shaft. The cephalic framework is weakly sclerotized with a distinct vestibule extension. The stylet is slender and moderately long



Figure 1: Photomicrograph of a J2 *M. fallax* nematode. Photo courtesy of Michael McClure.

with a straight cone. The shaft is cylindrical. The knobs are distinct, rounded, and set off from the shaft. The pharynx has a faintly outlined procorpus and an oval shaped metacorpus with a distinct valve. The oesophageal gland lobe is variable in length, and overlaps the intestine on the ventral side. The hemizonid is distinct at the level of the excretory pore. The tail is moderately sized and gradually tapers until the hyaline tail terminus with an inflated proctodeum. Phasmids are small and difficult to observe but can be found slightly posterior to the anus. A rounded hypodermis marks the anterior

position of the smooth hyaline tail terminus which ends in a broadly rounded tip. The terminus is generally marked by faint cuticular constrictions.

<u>Females:</u> Females are 0.491 mm \pm 0.074 mm in length. Other measurements include 0.361 mm \pm 0.057 mm in diameter at the widest point, a neck length of 0.149 mm \pm 0.033 mm, a neck diameter of 0.097 mm \pm 0.023 mm, a stylet length of 0.014 mm, and a vulval slit length of 0.024 mm \pm 0.01 mm.

The body is annulated, pearly white, globular to pear shaped, with slight posterior protuberance and distinct neck region projecting from the body axis at an angle of up to 90° to one side. The head is set off from the body, marked by one or two annules. The head cap is distinct but variable in shape, and the labial disc is slightly elevated. The cephalic framework is weakly sclerotized with distinct vestibule extension. The stylet cone is dorsally



Figure 2. Symptoms of *M. fallax* on potato. Photos courtesy of EPPO.

curved, and the shaft is cylindrical. The knobs are large, rounded to transversely ovoid, with slight sloping posterior from the shaft. The excretory pore is located between the head end and metacorpus levels. One or two large vesicles and several smaller ones are located along the lining of the lumen. Pharyngeal glands vary in size and shape. The perineal pattern is ovoid to oval shaped, sometimes rectangular. The dorsal arch ranges from low to moderately high, with coarse striae. The lateral lines are indistinct, appearing as weak indentations that increase toward the tail terminus, and result in a relatively large area without striae. The ventral pattern region is oval to angular shaped with moderately coarse striae.

<u>Males</u>: Males are 1.171 mm \pm 0.193 mm in length. Other measurements include 0.036 mm \pm 0.002 mm in diameter at the widest point, a head region height of 0.046 mm, a stylet length of 0.019 mm, spicules that are 0.026 mm \pm 0.002 mm long, a gubernaculum 0.007 mm in length, and testis 0.0496 mm \pm 0.144 mm in length.

The male body is vermiform with slight tapering anteriorly and blunt rounding posteriorly. The cuticle has distinct transverse striae. The lateral field has four incisures with outer bands that areolated irregularly. A fifth broken longitudinal incisure is rarely present near the mid-body. The head is slightly set-off with a single post-labial annule that is usually partly subdivided by a transverse incisures. The labial disc is rounded, elevated, and fused with medial lips. The prestoma is hexagonal in shape

with six inner cephalic sensilla adjacent to the rim. Medial lips are crescent shaped with raised with raised edges at the lateral sides. There are four cephalic sensilla that are small and marked by cuticular depressions on the medial lips. There are amphidial openings that appear as elongated slits between the labial disc and medium sized lateral lips. The cephalic framework is moderately sclerotized with the vestibule extension distinct. The stylet cone is straight, the shaft cylindrical, the knobs are large and rounded and set off from the shaft. The pharynx has a slender procorpus, and an oval metacorpus with a pronounced valve. The ventrally overlapping pharyngeal gland lobe is variable in length. There are two to four annules anterior to the excretory pore. The testes are usually long, monarchic, with reflexed or outstretched germinal zone. The tail is short and twisted, the spicules are slender, and curved ventrally. The gubernaculum is slightly crescentshaped, and the phasmids are located anterior to the cloaca.



Figure 3. "Pimples" on potatoes due to gall development of *M. fallax.* Photo courtesy of Farhat Shah.

Biology and Ecology

Nematodes are unsegmented roundworms. Most plant parasitic types are very small and feed on roots by means of a stylet, a hollow, needle-like structure used to pierce plant cells and withdraw nutrients. *Meloidogyne fallax* has a life cycle very similar to most root-knot nematodes.

Typically found in sandy to sandy-loam soils that are slightly acidic (Waeyenberge and Moens, 2001), female *M. fallax* lay eggs in gelatinous masses found on the surface or inside of gall tissue. The egg sac is initially sticky and hyaline in color, but becomes harder and darker brown



Figure 4: Cross-section of a potato infected with *M. fallax*. Photo courtesy of Farhat Shah.

with age. The eggs mature into first, then second-stage juveniles (J2s). Once the J2

molt is complete, the juveniles hatch and begin to search for another root location to bury into. Typically, the J2s begin to feed from the roots at the root tip, and then move up the root to find a permanent spot. J2 feed on the protoxylem and protophloem cells in the root, which causes their cells to differentiate into specialized nurse (feeding) cells called giant cells. Once these cells develop, the J2 remains sedentary and enlarges. After feeding, the J2s differentiate into either male or females and continue to molt up through the fourth stage juvenile (J4) and eventually into adults (Perry et al., 2009).



Figure 5. Brown speckling due to *M. fallax* in the potato. Photo courtesy of Farhat Shah.

Symptoms and Signs

In potato: Symptoms are very similar to those caused by *Meloidogyne chitwoodi*. *Meloidogyne fallax* females cause galls (Fig. 2, 3) on the below-ground regions of host plants. Each gall develops as a small raised lump above a developing female underneath the host epidermis. These galls can be white, cream, or light brown and can vary in size from 1 to 5 mm (0.039 to 0.197 in.) in diameter. On potato in particular, *M. fallax* results in raised swellings on the tuber surface above the developing nematodes (Fig. 2, 3). Gall development on potato tubers depends on cultivar and level of infestation in the soil. It can be difficult to detect an infestation in freshly harvested tubers, but as they sit in storage, the egg sacs on the cortices of the potato turn from translucent to brown, giving the potato flesh a speckled appearance (NAPPO, 1996; Shah et al., 2010). Internal tissue below the gall is often damaged, necrotic, and brownish in color when infected by *M. fallax* (Fig. 2, 4, 5).

Some potato cultivars can be infested with *M. chitwoodi* or *M. fallax* without clear external symptoms (Karssen, personal communication).

Due to the root damage caused by the gall formation and the stimulation of giant cell growth at feeding sites in parenchyma and phloem, above-ground symptoms of *M. fallax* infestation resemble nutrient or water deficiency. These above ground symptoms include: yellowing, wilting, premature senescence, and stunting (Davis and Venette, 2004). The reduction in tuber and vegetable quality results in lower yield and, subsequently, economic losses.

On turf: There are numerous root knot nematodes which cause turf damage, and symptoms are similar for each nematode (Fig. 6). Symptoms include yellowing and death of grass, root stunting, death of roots, and knots on roots. However, visual symptoms alone are similar to symptoms of nutrient deficiency, lack of moisture, and other diseases. Soil samples must be taken to verify the presence of nematodes.

Pest Importance

Meloidogyne species are among the most important plant-parasitic pests in worldwide crop production. Crop losses from nematode damage alone are estimated at 10 to 11% worldwide. but this is thought to be an underestimate. If unregulated, nematode related crop losses to potatoes grown in the Northwest region of the United States could be as much as \$40 million (Davis and Venette, 2004). The economic impact of *M. fallax* alone is difficult to measure because this species frequently occurs in mixed populations. As a result, it is possible to ascribe nematode damage within a field to *Meloidogyne* sp. in general but



Figure 6: A golf course green infested with *M. minor*. Symptoms are similar to those of *M. fallax* infestation. Photo courtesy of Colin Fleming.

not *M. fallax* alone. Due to its large host range of *M. fallax* across monocot and dicot plant species, there is also potential for this pest to disrupt the ecosystem function in the environments that support the affected host (Davis and Venette, 2004).

The presence of *M. fallax* in Europe poses a threat to the potato trade there. In 2005, countries in the European Union produced 18.5% of the global share of potatoes. EU-25 trade in potato products (not including starch and sweet potatoes) was worth about €2.76 billion (\$3.69 billion) in the period 2003/05 (EC, 2007). Total human consumption of potatoes amounted to roughly 38 million tons in the EU-25 during the period from 2001-2003.

Turfgrass is the largest irrigated crop in the United States and is a major industry. Golf is a primary consumer and producer of turf. The impact of *Meloidogyne* sp. on the golf course industry, estimated to have an annual economic impact of \$195 billion in the United States alone, is already significant and could become even greater if *M. fallax* becomes established. McClure et al. (2012) surveyed 238 golf courses in the Western United States and found root knot nematodes in 60% of the putting greens sampled.

M. fallax is listed as a harmful organism in the following countries: Albania, Austria, Belgium, Brazil, Bulgaria, Chile, Croatia, Cyprus, Czech Republic Denmark, Estonia, Finland, France, Germany, Greece, Holy See (Vatican City State), Hungary, Ireland, Italy, Japan, Korea (South), Latvia, Lithuania, Luxembourg, Malta, Monaco, Montenegro, Morocco, Netherlands, Poland, Portugal, Romania, San Marino, Serbia, Slovakia, Slovenia, Spain, Sweden, United Kingdom (USDA-PCIT, 2013).

There may be trade implications with these countries if this nematode becomes established in the United States.

Known Hosts

Major hosts: *Daucus carota* (carrot), *Scorzonera hispanica* (oyster plant), *Solanum nigrum* (black nightshade), *Solanum physalifolium* (hairy nightshade), and *Solanum tubersoum* (potato) (CABI, 2013)

Minor hosts: *Allium ampeloprasum* (leek), *Allium moly* (wild onion/garlic), *Arachis hypogeae* (peanut), *Beta* sp. (sugarbeet), *Chenopodium album* (lambquarters), *Cynara* sp. (artichoke), *Eriogonum* sp. (buckwheat), *Fragaria* sp. (strawberry), *Lactuca sativa* (lettuce), *Lolium perenne* (ryegrass), *Medicago* sp. (alfalfa), *Phacelia tenacetifolia* (lacy phacelia), *Poa annua* (annual meadowgrass), *Raphanus sativus* (radish), *Sinaphis alba* (white mustard), *Solanum esculentum* (tomato), and *Zea* sp. (corn) (SON, 2003; Davis and Venette, 2004; van der Sommen et al., 2005; Vanstone and Nobbs, 2007; Fleming, personal communication).

Note: Corn is a reported minor host of *M. fallax* by Waeyenberge and Moens (2001). The Society of Nematologists (2003) and Ferris (2011), however, suggests that corn is not a host of *M. fallax*. Karssen (personal communication) suggests that corn can be considered a very poor host.

Experimental field hosts: Acer palmatum (Japanese maple), Aconitum napellus (venus' chariot), Adiantum sp. (fern), Apium graveolens (celery), Asparagus sp. (asparagus), Betula pendula (birch), Borago officinalis (borago), Chionodoxa luciliae, Cichorium endivia (endive), Cichorium intybus (chicory), Clematis sp. (clematis), Crocus sp. (crocus), Delphinium sp. (larkspur), Dahlia sp. (dahlia), Dicentra spectabilis (bleeding hearts), Fagopyrum sp. (buckwheat), Foeniculum vulgare (fennel), Gladiolus sp. (gladiolus), Hemerocallis sp. (daylily), Hordium vulgare (barley), Hyacinthus orientalis (hyacinth), Iris sp. (iris), Laburnum anagyroides (golden chain tree), Lilium sp. (lily), Lonicera xylosteum (honeysuckle), Narcissus sp. (daffodil), Oenothera sp. (primrose), Petrosolinum crispum (parsley), Scilla siberia (squill), Secale cereale (rye), Sinapis alba (mustard), Triticum sp. (wheat), and Tulipia sp. (tulip) (den Nijs et al., 2004).

Hosts for *M. fallax* which are not shared with *M. chitwoodi* include: *Hemerocallis* sp., *Dicentra spectabilis, Oenothera erythrosepala*, and *Phacelia tenacetifolia* (Ferris, 2011). Differential hosts which are infected by *M. chitwoodi* but not by *M. fallax* are bean (*Phaseolus vulgaris*) and corn (*Zea mays*) (Ferris, 2011).

Known Vectors or Associated Organisms

As a soil-borne nematode, this pest is not vectored by another organism. DNA fingerprinting and bacterial community level physiological profiles have demonstrated *that M. fallax* egg masses supported bacterial communities distinct from the surrounding rhizosphere. However, it is still not fully understood how the communities on the egg masses interact with the surrounding rhizosphere communities, or how they affect the pathogenicity of the nematode (Papert et al., 2004).

Known Distribution

Europe: Belgium, France, Germany, the Netherlands, Switzerland, and England, United Kingdom (CABI, 2013; Fleming, personal communication).

Eradication efforts in the field and management techniques in glasshouses are underway in two locations in France (Gamon and Lenne, 2012).

Oceania: Australia and New Zealand (Marshall et al., 2001; Nobbs et al., 2001; Waeyenberge and Moens, 2001; Vanstone and Nobbs, 2007; Perry et al., 2009; Eder et al., 2010; CABI, 2013).

North America: United States.

Meloidogyne fallax was recently detected in San Francisco County, California in a sample taken from a golf course green of *Poa annua* exhibiting symptoms of yellow patch disease (Nischwitz et al., 2013).

Note: There has been no official APHIS confirmation of the detection of *M. fallax* in the United States. *Meloidogyne fallax* was detected from a single location in the United States (Nischwitz et al., 2013), and we are awaiting confirmation from an official sample (NPAG, 2013).

There is a report from South Africa, but it is not considered reliable (CABI, 2013). There was also an erroneous report of this pest in Canada (Shah et al., 2010), but it has never been found there.

Pathway

Meloidogyne fallax spreads primarily via movement of infect tubers or bulbs. Additionally, the movement of any materials contaminated with *M. fallax* infested soil can contribute to spreading the nematode. These materials could include non-host plant material like seedling transplants or nursery stock (NAPPO, 1996). There are many possible pathways due to the large number of hosts and the difficulty of detecting nematodes visually.

There are no regulations on the import of *Daucus carota* plant material into the United States (USDA, 2013). *Daucus carota* is a major host of *M. fallax*. This appears to be an open pathway.

Since 2003, there were 78 shipments of *Solanum* sp. propagative material from known host countries. While the majority of these shipments appear to be seed, several of them were measured in plant units and may have been plant material. There have also been 113 interceptions of *Solanum* sp. propagative material since 2003, including 102 from the Netherlands alone (AQAS, 2013). Effective May 20, 2013, under federal regulation 7 CFR 319.37-2a, import of all *Solanum* sp. propagules except seeds from countries other than Canada is prohibited pending a pest risk analysis of *Tuta absoluta*, the tomato leafminer (USDA, 2013).

Potential Distribution within the United States

A recent risk analysis by APHIS-PPQ-CPHST based on the presence of susceptible host plants shows that Illinois, Indiana, Illinois, Indiana, Minnesota, Nebraska, North Dakota, Ohio, South Dakota, Wisconsin, parts of Colorado, and Montana, Oklahoma, and Washington are at the highest risk from this nematode pest.

The top potato producing states in 2015 were: Idaho, Washington, Oregon, North Dakota, Colorado, and Wisconsin (USDA-NASS, 2016).

Survey

CAPS-Approved Method*:

Soil sampling: Send sample to nematology diagnostic lab where nematodes will be extracted and identified.

Visual: Collect symptomatic tubers.

Detection and inspection methods are similar to those for *M. chitwoodi*. The presence of *M. fallax* in infested soil can be determined by sampling and extraction of the second-stage juveniles, using a standard nematode extraction procedure for free-living nematodes of this size or techniques used for root-knot nematodes (Baermann trays, Baermann trays with elutriation or sieving, centrifugal flotation, flotation-sieving, semiautomatic elutriation and Cobb decanting and sieving).

External symptoms on tubers are obvious in the case of heavy infestations but, where nematode numbers are low or in the early stages of infection, such symptoms are not obvious. Clearing and staining of the tissues can show the presence of nematodes (Hooper, 1986), but this can be a laborious procedure. Storage of lightly infested tubers may lead to the development of obvious external symptoms.

*For the most up-to-date methods for survey and identification, see Approved Methods on the CAPS Resource and Collaboration Site, at <u>http://caps.ceris.purdue.edu/</u>.

Literature-Based Methods:

<u>Soil sampling:</u> Collect 20 soil core samples (1 cm diameter x 300 cm deep) at random from the plant rows. These will serve as one bulk field sample. Extract nematodes from 100 cm³ soil samples from the bulk field sample using a modified sieve and centrifuge technique described by Dropkin (Dropkin, 1989; Powers et al., 2005).

Root-knot nematodes are extracted from soil using a variety of techniques. Six methods (and subtle variations thereof) are particularly common: Baermann trays; Baermann trays with elutriation or sieving; centrifugal flotation; flotation-sieving; semiautomatic elutriation; and Cobb's decanting and sieving. These methods are described in detail by Barker (1985). The efficiency of the nematode extraction is influenced by the amount of soil that is processed at one time. Extraction efficiencies are greatest when 100 g to 450 g of soil are processed. Extraction efficiencies for

Meloidogyne sp. are frequently low and can vary between 13 and 45% (Davis and Venette, 2004).

Key Diagnostics/Identification

CAPS-Approved Method*:

Morphological: Characteristics of the males, females, and juveniles (Karssen, 1996).

*For the most up-to-date methods for survey and identification, see Approved Methods on the CAPS Resource and Collaboration Site, at <u>http://caps.ceris.purdue.edu/</u>.

Literature Based Methods:

<u>Molecular:</u> Common molecular techniques to identify *M. fallax* include RFLP, SCAR, and RAPD (Random Amplification of Polymorphic DNA)-based procedures.

Petersen and Vrain (1996) and Wishart et al. (2002) developed PCR primers to identify *M. fallax, M. chitwoodi*, and *M. hapla*.

Zijlstra (1997) used a multiplex PCR to distinguish *M. fallax, M. hapla, M. chitwoodi*, and *M. incognita*.

Zijlstra (2000) used SCAR-PCR to identify *M. fallax, M. hapla*, and *M. chitwoodi*. **Update:** This PCR was designed before the description of *M. minor*, and subsequent research by Nischwitz et al. (2013) showed that *M. minor* amplification also occurs and cannot be distinguished from *M. fallax* using this SCAR-PCR. Therefore, it is not advised to use this protocol if a mixed presence of both *M. fallax* and *M. minor* is possible in the same sample.

Adam et al. (2007) developed a molecular diagnostic key to identify juveniles of seven species, including *M. fallax, M. incognita, M. javanica, M. arenaria, M. mayaguensis, M. hapla*, and *M. chitwoodi*. Petersen et al. (1997) used a multiplex PCR to distinguish the juveniles and eggs of the same seven species. Fourie et al. (2001) distinguished the same species except *M. mayaguensis*.

A real-time PCR for the simultaneous detection of *M. fallax* and *M. chitwoodi* has been developed (Zijlstra and van Hoof, 2006).

Nischwitz et al. (2013) describe a method to distinguish *M. fallax* from *M. minor* and *M. chitwoodi* by amplification and sequencing of Hsp90 genomic DNA.

Easily Confused Pests

Meloidogyne fallax can be easily confused with *M. chitwoodi*, and was originally described as a race of the *M. chitwoodi* species. The two species differ in stylet length in females and males, by the shorter life cycle completion time for *M. fallax*, and by the longer tail and hyaline portion of the tail of the second stage juveniles. There are also biochemical and molecular techniques to separate the two species, including isozyme assays, fatty acid assay, and species specific primers in polymerase chain reaction assays (SON, 2003). Biochemically, *M. fallax* is characterized by the unique malate dehydrogenase phenotype known as N1b, and a complete lack of any major esterase activity (Perry et al., 2009).

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In addition to *M. chitwoodi*, there are numerous other *Meloidogyne* sp. which are easily confused with *M. fallax*. These include *M. hapla*, *M. incognita*, and *M. minor*. *M. minor* in particular has very similar morphology to *M. fallax* and also cross reacts with *M. fallax* using the SCAR-PCR described in Zijlstra et al. (2000) (Nischwitz et al., 2013).

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Draft log

July, 2016: Updated maps, Potential Distribution sections