

# Chapter 20

## NEMATODES

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### Introduction

Numerous plant-parasitic nematodes (eelworms) are associated with roots and soils of beans and other plants throughout the world (Table 1). Many of these nematodes have been reported to cause considerable damage to many crops, including beans (Abawi and Jacobsen, 1984; Costa, 1972; Keplinger and Abawi, 1976; Mai et al., 1977; Manzano et al., 1972; McSorley, 1980; McSorley et al., 1981; Melton et al., 1985; Navarro-A. and Barriga-O., 1974; Freire, 1976; Freire and Ferraz, 1977a; Renaud and Thomason, 1973; Rhoades, 1983; Riedel, 1978; Sen and Jensen, 1969; Taylor, 1965; Taylor and Sasser, 1978; Taylor et al., 1970; Zaumeyer and Thomas, 1957). However, only the species of the *Meloidogyne* and *Pratylenchus* genera are frequently and consistently found on beans in relatively high densities in Latin and North America.

Nematode infestations at high initial population densities cause significant yield losses. For example, yield losses may reach 10% to 80% with lesion nematodes (Elliott and Bird, 1985; Robbins et al., 1972), and 50% to 90% with root-knot nematodes (Freire and Ferraz, 1977a; Varón de Agudelo and Gálvez 1974; Varón de Agudelo and Riedel, 1982; Zaumeyer and Thomas, 1957). In addition, plant-parasitic nematodes, particularly the root-knot nematodes, are known to predispose many crop plants to various soil-borne microorganisms that induce root rot and wilt diseases (Elliott et al., 1984b; Powell, 1979; Ribeiro and Ferraz, 1983; Schuster, 1959; Singh et al., 1981b; Walker and Wallace, 1975).

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Table 1. Nematodes frequently found in association with roots of common beans and other plants.

Scientific name	Common name
<i>Aphelenchoïdes</i> spp.	Bud-and-leaf nematode
<i>Belonolaimus gracilis</i> Steiner	Sting nematode
<i>Belonolaimus longicaudatus</i> Rau	Sting nematode
<i>Criconemella</i> spp.	Ring nematode
<i>Ditylenchus destructor</i> Thorne	Potato-rot nematode
<i>Ditylenchus dipsaci</i> (Kühn) Filipjev	Stem-and-bulb nematode
<i>Helicotylenchus</i> spp.	Spiral nematode
<i>Heterodera glycines</i> Ichinohe	Soybean-cyst nematode
<i>Heterodera humuli</i> Filipjev	Hop-cyst nematode
<i>Heterodera schachtii</i> Schmidt	Sugarbeet nematode
<i>Heterodera trifolii</i> Goffart	Clover-cyst nematode
<i>Meloidogyne arenaria</i> (Neal) Chitwood	Root-knot nematode
<i>Meloidogyne hapla</i> Chitwood	Root-knot nematode
<i>Meloidogyne incognita</i> (Kofoid et White) Chitwood	Root-knot nematode
<i>Meloidogyne javanica</i> (Treub) Chitwood	Root-knot nematode
<i>Pratylenchus brachyurus</i> (Godfrey) Filipjev et Schuurmans Stekhoven	Root-lesion nematode
<i>Pratylenchus penetrans</i> (Cobb) Filipjev et Schuurmans Stekhoven	Root-lesion nematode
<i>Pratylenchus scribneri</i> Steiner	Root-lesion nematode
<i>Rotylenchulus reniformis</i> Linford et Oliveira	Reniform nematode
<i>Trichodorus</i> spp.	Stubby-root nematode
<i>Tylenchorhynchus</i> spp.	Stunt nematode
<i>Xiphinema elongatum</i> Schuurmans Stekhoven et Teunissen	Dagger nematode
<i>Xiphinema krugi</i> Lordello	Dagger nematode
<i>Xiphinema setariae</i> Luc	Dagger nematode

This chapter will only summarize available information on root-knot nematodes (*Meloidogyne* spp.) and root-lesion nematodes (*Pratylenchus* spp.) found on beans. For general information on plant-parasitic nematodes, see Mai and Lyon (1975) for taxonomic treatments with an easy-to-use pictorial key for the identification of

plant-parasitic nematodes; Zuckerman et al. (1971) for the principles of plant nematology and the ecology, biology, and management of nematodes as plant pathogens; Varón de Agudelo and Riedel (1982) for the main nematodes found on beans and their control (an auditorial prepared at the Centro Internacional de Agricultura Tropical (CIAT) for training programs); and Sasser and Kirby (1979), Taylor and Sasser (1978), and Taylor et al. (1970) for detailed information dealing with the worldwide distribution, ecology, epidemiology, and management of root-knot nematodes (International Meloidogyne Project publications).

Common names frequently used for *Meloidogyne* species in Latin America include "nematodos de las nudosidades radicales" and "galhas das raízes." Names commonly used for *Pratylenchus* species include "nematodos de las lesiones radicales," "lesiones por nematodos," and "definhamento de nematoide."

## Epidemiology and Life Cycle

### Root-knot nematodes

Although there are about 50 reported species of root-knot nematodes, four major species (*M. arenaria*, *M. hapla*, *M. incognita*, and *M. javanica*) have accounted for about 99% of all populations collected from cultivated crop species, including beans. Differential host tests and cytogenetical analysis have identified four races of *M. incognita*, two races of *M. arenaria* populations, and one race each of *M. javanica* and *M. hapla*. Populations of *M. hapla* occur in relatively cold areas since they tolerate temperatures as low as -15 °C. The other three species are adapted to and occur in high-temperature areas. *Meloidogyne incognita* and *M. javanica* are the most prevalent root-knot species in tropical and subtropical regions.

Root-knot nematodes are obligate, endoparasites with a wide host range, including agronomic crops and weeds that belong to many plant families. These nematodes are most abundant and cause serious damage in coarse-textured soil with good drainage (Crispín-Medina et al., 1976; Taylor et al., 1982) such as the coastal soils of Peru. Very few populations of *Meloidogyne* spp. have been found in

soils with more than 40% clay or 50% silt fractions (Taylor et al., 1982). Root-knot nematodes survive in soil as eggs and larvae. Length of survival in soil varies with the nematode species, stage of development, soil texture, soil moisture, and soil aeration (Taylor and Sasser, 1978). Dissemination of nematodes among fields and growing regions can be by irrigation water, vegetative plant parts, and soil infested with eggs or larvae which adhere to farm implements, animals, or man (Crispín-Medina et al., 1976; Vieira, 1967).

The life cycle of *Meloidogyne* spp., as is the case with other plant-parasitic nematodes, involves five developmental stages. Eggs are deposited by mature females in an egg sac consisting of a gelatinous matrix (glycoprotein-type substance) secreted by the female. This sac protects the eggs from dehydration (Figure 152) (Bird and Soeffky, 1972) and may contain as many as 1000 eggs. Eggs are oval to ellipsoidal and slightly concave (Figure 153). They are 30-52  $\mu\text{m}$  by 67-128  $\mu\text{m}$  in size (Thorne, 1961). The vermiform first-stage larvae and, later after the first molt, the second-stage larvae develop in the egg. The second-stage juvenile hatches by breaking the egg shell with repeated thrusting of its well-developed stylet (about 10  $\mu\text{m}$  long). These juveniles (Figure 154) are 375-500  $\mu\text{m}$  long and 15  $\mu\text{m}$  in width.

Second-stage, infective juveniles of *Meloidogyne* spp. move through the soil in search of host roots. Usually, they penetrate roots just behind the root cap and migrate inter- and intracellularly upwards through cortical tissue toward the stele (Ngundo and Taylor, 1975c). The juvenile head is inserted into the vascular system near the region of elongation to obtain plant nutrients. Plant cells in the vicinity of the juvenile increase in number (hyperplasia) and size (hypertrophy) as a result of nematode secretions. Giant cells form near the juvenile head by the fusion and enlargement of plant cells in response to nematode feeding. These giant cells (syncytia) produce root swellings called galls or knots.

Sedentary juveniles continue to enlarge during the formation of giant cells and galls, completing the second and third molts after which the sexes can be differentiated. Males and females are mature after the fourth molt. Adult males are vermiform, measure 0.03-0.36 by 1.20-1.50 mm, lack a bursa, and have a well-developed stylet.

Males are not essential for reproduction. Adult females are pyriform (Figure 155), pearly white, visible on roots without magnification, have a soft cuticle, and measure 0.27-0.75 by 0.40-1.30 mm (Southey, 1965).

Depending upon the host and soil temperature (Tyler, 1933), the entire life cycle (Figure A) may be completed in 17-57 days (Ngundo and Taylor, 1975a). Slight plant injury is apparent 10 days after penetration, but within 40 days epidermal cells often collapse, particularly if females had deposited eggs near the outer root surface (Ngundo and Taylor 1975b). Penetration by and pathogenicity of *Meloidogyne* spp. are affected by plant age, susceptibility, size of nematode populations, and the environment (Gilvonio-Vera and Ravines, 1971; McClure et al., 1974; Ngundo and Taylor, 1975c; Sosa-Moss and Torres, 1973).

Infection of beans by root-knot nematodes results in the reduction and malformation of the root system. There are accompanying physiological changes and a decreased efficiency in the absorption of water and nutrients (Melakeberhan et al., 1985; Wilcox and Loria, 1986). In addition, root-knot nematodes interact with other plant pathogens, resulting in increased plant damage caused by other diseases such as fusarium wilt (Ribeiro and Ferraz, 1983; Singh and Reddy, 1981b), rhizoctonia root rot (Reddy et al., 1979), bean rust (Bookbinder and Bloom, 1980), bacterial wilt (Schuster, 1959), and tobacco ring spot virus (Walker and Wallace, 1975). Infection by nonhost nematodes also reduces rhizobium nodulation (Singh and Reddy, 1981a).

## Root-lesion nematodes

Species of *Pratylenchus* are migratory endoparasites and are vermiform during all five developmental stages (Thorne, 1961). Although there are about 40 reported species of *Pratylenchus*, only *P. brachyurus*, *P. penetrans*, and *P. scribneri* are frequently found on beans. These three species are widely distributed and have numerous host crops in many plant families. Eggs, juveniles, and adults survive in infected roots or free in soil. Juveniles and adults can penetrate unsterilized plant roots and move through and between root cells causing cell breakdown and necrosis. Breakdown

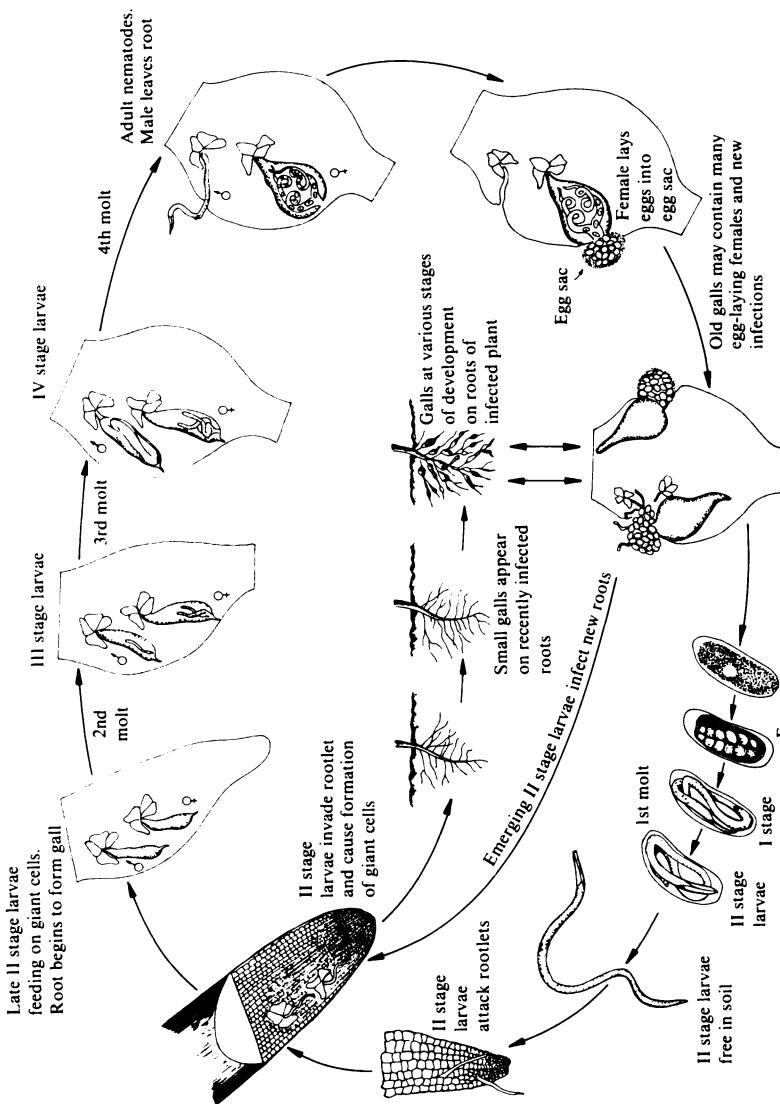


Figure A. Life cycle of *Meloidogyne* sp. (from Agrios, 1978).

of cell walls results, in part, from the mechanical action of nematode spears (stylets), pressure of their body movements in roots, and from enzymes and other substances secreted by the nematodes. Root-lesion nematodes are restricted to the root cortex (Thomason et al., 1976; Thorne, 1961).

Females lay eggs in clusters in root tissues. First-stage larvae and, after the first molt, second-stage juveniles form within the egg. After hatching, second-stage juveniles begin to feed in or migrate outside root tissues in search of other roots to parasitize (Figure 156). Except for the sexual organs, males and females of *Pratylenchus* spp. are similar. They are about 20-25  $\mu\text{m}$  long and 0.4-0.7  $\mu\text{m}$  wide. In some species males are numerous and are required for the reproduction of the species (Mai et al., 1977). Length of the life cycle (Figure B) is variable, depending on nematode species, host crop, and environmental conditions. It ranges from 25-50 days.

Damage to crops, including beans, depends on initial nematode density in soil. A recent greenhouse study (Elliott and Bird, 1985) showed that the growth of susceptible beans was reduced by an initial soil population density of 50 or more *P. penetrans* per 100  $\text{cm}^3$  soil. Yield of susceptible bean cultivars was reduced 43%-47% at densities of 150 *P. penetrans* per 100  $\text{cm}^3$  soil. Species of *Pratylenchus* interact with other soil-borne organisms infecting bean roots. For example, infection by *P. penetrans* increases the incidence and severity of fusarium root rot (Hutton et al., 1973) and of the mycorrhizal fungus *Glomus fasciculatus* (Thaxter *sensu* Gerdemann) Gerdemann et Trappe (Elliott et al., 1984b).

## Symptomatology

Plants infected with species of *Meloidogyne* or *Pratylenchus* do not necessarily exhibit characteristic foliar symptoms. Severely infected plants may show chlorosis, stunting, necrosis of leaf margins, and wilting during periods of moisture stress (Figure 157). Distribution of infected plants within a field depends on the history of nematode infestation and the cropping system practiced. In a newly infested field, infected plants showing foliar symptoms may be restricted to one or a few small areas. If a susceptible crop is grown repeatedly in an infested field, the small areas in which growth is poor will

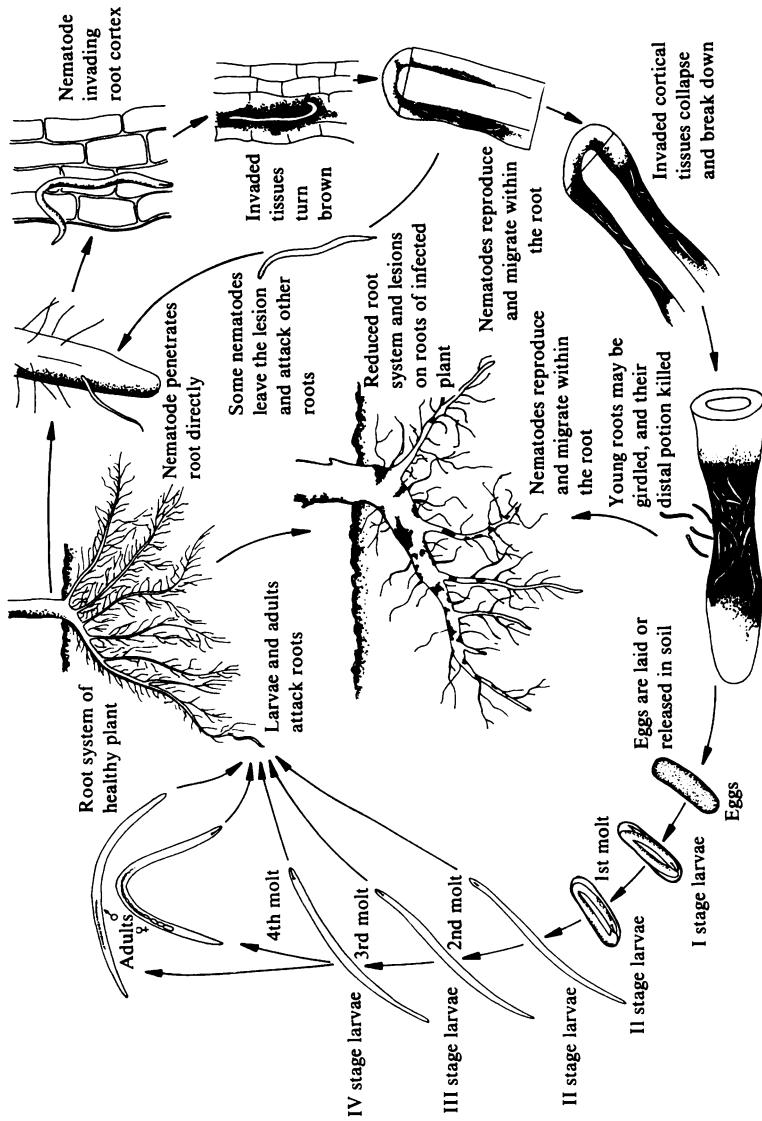


Figure B. Life cycle of *Pratylenchus* sp. (from Agrios, 1978).

gradually enlarge and affected areas with stunted and chlorotic plants will appear throughout the field.

Diagnostic symptoms for nematode infection can, however, be found more clearly on the root system. For proper examination of bean roots, plants must be dug up carefully and the soil removed with as little disturbance to fibrous roots as possible. Roots of bean plants infected by *Meloidogyne* spp. exhibit galls or root knots, usually on the primary and secondary roots (Figure 158). Depending on the species involved, galls may range in size from as small as a pinhead to 12 mm or more in diameter. In addition, the root system becomes malformed with shortened and thickened individual roots which may appear as a mass of galls. Intensive galling interferes seriously with normal root functions, often causing premature defoliation and plant stunting, but rarely death. Stem and hypocotyl tissues may become infected and also exhibit galls, especially when bean seeds are planted too deep (Fassuliotis and Deakin, 1973). Galls induced by root-knot nematodes cannot be detached from the root system without breaking the root. In comparison, nodules formed by nitrogen-fixing *Rhizobium* bacteria are loosely attached to the sides of roots (Renaud and Thomason, 1973).

Bean plants heavily infected by root-lesion nematodes have a reduced root system and, depending on the cultivar, may exhibit brown or black small lesions on the roots (Figure 159). These lesions result from penetration and feeding activities of nematodes in epidermal and cortical tissues (Ngundo and Taylor, 1975b; Thomason et al., 1976). However, diagnostic proof of damage by these nematodes requires extraction of larval and adult stages from roots and adjacent soil. Parasitic nematodes can also be observed directly inside roots by using a compound microscope. However, they can be confused with bacterial feeders unless staining techniques are used by trained observers.

Under natural field conditions infections of bean roots by species of *Meloidogyne* and *Pratylenchus* occur in the presence of many pathogenic and saprophytic soil microorganisms in the rhizosphere. Thus, these nematodes play an important role as a component of the microbial complexes that cause discoloration, necrosis, and eventually decay of plant roots. Decay results from various interactions that can occur among nematodes and soil microorganisms, as well

as from the ability of the nematodes to affect the physiology of plant roots and so predispose them to the detrimental activities of rhizosphere fauna and flora (Elliott et al., 1984b; Powell, 1979; Ribeiro and Ferraz, 1983; Schuster, 1959; Singh et al., 1981b; Walker and Wallace, 1975).

## Control by Cultural Practices and Biological Agents

Crop rotation can reduce population levels of root-knot nematodes when beans are planted once every two or three years in rotation with nonhosts such as maize. Growing crops antagonistic to nematodes such as *Tagetes minuta* L. (marigolds), *Crotalaria spectabilis* Roth. (rattlebox) (Hackney and Dickerson, 1975; Navarro-A. and Barriga-O., 1970; Zaumeyer and Thomas, 1957), or *Indigofera hirsuta* L. (hairy indigo) can reduce populations of both root-knot and root-lesion nematodes (Rhoades, 1976). However, many plant-parasitic nematodes such as *Meloidogyne* and *Pratylenchus* species have a wide host range which make crop rotation at times hard to formulate or impractical.

Other cultural practices which reduce nematode populations include long fallow periods, deep plowing, weed control, and, where practical, flooding for one or two weeks (Crispín-Medina et al., 1976; Taylor and Sasser, 1978; Vieira, 1967). Several parasitic and antagonistic microorganisms of eggs and adult stages of plant-parasitic nematodes have been described (Barron, 1977; Kerry, 1980; Mankau, 1980; Sayre, 1980). However, the field effectiveness of these organisms and their economic commercial use are not encouraging.

## Control by Chemicals

Chemical control of plant-parasitic nematodes with nematicides is very effective and used widely on annual agronomic crops. However, use of nematicides is expensive for a crop like beans and requires care in handling and often the use of special equipment for application. Fumigant nematicides such as D-D soil fumigant (1,3-dichlorpropene and related hydrocarbons), methyl bromide, chloropicrin, and Vorlex, have been used successfully on beans and other

crops (Hartmann, 1968a; Jiménez, 1976; Johnson et al., 1979; McSorley and Parrado, 1983; Powell, 1974; Reddy, 1984; Rhoades, 1976 and 1983; Robbins et al., 1972).

In addition, control of nematodes and increase of bean yield have been obtained with the use of nonfumigant nematicides such as aldicarb, phenamiphos, carbofuran, and oxamyl, applied as a broadcast or band and incorporated into the soil (Abawi and Crosier, 1985; Elliott et al., 1984a; Jiménez, 1976; Rhoades, 1983; Singh and Reddy, 1981b). The application of the nematicide oxamyl to beans as a foliar spray has been effective against many nematodes (Abawi and Mai, 1975; McSorley, 1980; Smittle and Johnson, 1982). However, its activity against the root-knot nematode is limited and a combination of a soil treatment with foliar sprays of oxamyl is recommended (Starr et al., 1978). There have been some encouraging results from the application of nematicides such as oxamyl, as seed treatments to beans (Carvalho et al., 1981; Ngundo and Taylor, 1974; Parisi et al., 1972; Sosa-Moss and Camacho-Guerrero, 1973; Truelove et al., 1977).

## Control by Plant Resistance

The use of bean cultivars highly tolerant to plant-parasitic nematodes is the most efficient control strategy, especially for small farmers with limited production inputs. Numerous reports are available that describe the evaluation and identification of bean germplasm with tolerance to plant-parasitic nematodes, especially the *Meloidogyne* spp. (Arias and Ranaud, 1982; Blazey et al., 1964; Cabanillas, 1982; Dickerson and Franz, 1974; Elliott and Bird, 1985; Fassuliotis et al., 1970; Ginoux et al., 1979; Hadisoeganda and Sasser, 1982; Hartmann, 1968a, 1968b, and 1971; López, 1980; Ngundo, 1977; Reddy et al., 1979; Sasser and Kirby, 1979; Singh et al., 1981a; Taha et al., 1977; Varón de Agudelo and Gálvez, 1974; Vieira, 1967; Wilcox and Loria, 1986; Wyatt and Fassuliotis, 1979; Wyatt et al., 1980a, 1980b, and 1983; Zaumeyer and Meiners, 1975; Zaumeyer and Thomas, 1957). The cultivars and breeding lines that are reported as tolerant to root-knot nematodes are Alabama 1, 2, 8, and 19, Spartan, State, P.I. 165426, Rico 23, Manteigão Fosco 11, Porto-Alegre-Vagem-Roxa, Coffee Wonder, Manão Wonder,

Spring Water Half Runner, Wingard Wonder, P.I. 165435, P.I. 313709, Nyakahuti, Red Haricot, Rono, Saginaw, Kibu, Bountiful, Tender Pod, Brittle Wax, My Finca, E.E.U.U. 1-263, Contender, Tender Green, Nema Snap, B 4175, and Strike.

Saginaw, Seafarer, Tuscola, and others are reported as tolerant to the root-lesion nematode (*P. penetrans*). Resistant lima bean cultivars include Hopi, L 5980, Nema Green, Westan, and White Ventura (Allard, 1954; Wester et al., 1958).

Root-knot resistant germplasm is stable (Taylor and Sasser, 1978), but resistance to one race or species of root-knot nematodes is often independent of other races or species. For example, the bean cultivar Contender was highly resistant to races 2, 3, and 4 of *M. incognita*, but only moderately resistant to race 1 (Hadisoeganda and Sasser, 1982). P.I. 165426 is resistant to *M. incognita* (Fassuliotis et al., 1970), but is susceptible to simultaneous infection by *M. incognita* and *M. javanica* (Ngundo, 1977).

Resistance to gall formation and resistance to the buildup of nematode populations in root systems are characters independent of tolerance to yield reduction. They are probably governed by separate genetic mechanisms (Hadisoeganda and Sasser, 1982; Wyatt, 1976). Selection of tolerant bean germplasm is often based upon root galling, egg-mass formation, and number of eggs produced per gram of root tissue. However, the galling index does not always correlate with yield (Ngundo, 1977). Galling, female development, and egg-mass production increase as temperature is raised from 16 to 28 °C (Fassuliotis et al., 1970; Freire and Ferraz, 1977b). A hypersensitive necrotic (resistant) response may appear about four days after inoculation (Fassuliotis et al., 1970). A recent report has suggested that cultivar tolerance in beans to root-knot nematodes is related to the effects of nematodes on plant-water relations (Wilcox and Loria, 1986).

Only limited information is available on the inheritance of resistance to plant-parasitic nematodes in beans. Resistance to *M. incognita* is governed by two or three dominant (Hartmann, 1971) and two recessive genes (Ginoux et al., 1979).

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