

¹Parasitology Research Laboratory, Biology Department, Xiamen University, China

²Department of Plant Pathology, Yunan Agricultural University, China

TEMPORAL FLUCTUATIONS IN *MELOIDOGYNE FUJIANENSIS* PARASITIZING *CITRUS RETICULATA* IN NANJING, CHINA

by

C. PAN¹, X. HU² and J. LIN¹

Summary. A study was made of the temporal fluctuation in the population density of *Meloidogyne fujianensis* parasitizing the roots of *Citrus reticulata* growing in Nanjing County, China. The lowest infection rate occurred in August (30%), and the highest in April of the following year (90%). The lowest average infectivity was recorded in August (1.8 females/2 g roots) and the highest in February of the following year (24.2 females/plant). Infection occurred throughout the year with peaks of infection in September and October and again in March to April of the following year. The peak of oviposition occurred during December and January. The population density increased with increasing monthly average temperatures. The most active growth of *C. reticulata* roots coincided with the peaks of infection of nematode juveniles (September-October, March-April), which was associated with two main periods of disease symptomatology. The ratio of males decreased with increasing monthly rainfall.

In the southern part of Fujian Province *Tylenchulus semipenetrans* causes serious reductions in the yields of citrus crops. However, *Meloidogyne fujianensis* is more widespread in this region, and appears to cause more serious damage to citrus than does *T. semipenetrans* (Pan, 1985; Pan *et al.*, 1988).

Temporal fluctuations in the population structure of *T. semipenetrans* under citrus have been reported (Al-Sayeed *et al.*, 1993) but similar studies have not been made with *M. fujianensis*. Therefore, a small area of an orange grove in Nanjing County was used to study temporal fluctuations in the population structure of *M. fujianensis* under orange.

Materials and methods

The study was made in a small area in a citrus grove (*Citrus reticulata* Blanco) on Fengtian Farm, Nanjing County, Fujian Province, China, at 24°6'N, 117°5'E. The site has a sandy loam soil, is 40 m asl., with a subtropical monsoon, frost-free climate, an annual average air temperature of 20 °C, and 1000 mm annual rainfall.

To examine temporal fluctuations in the *M. fujianensis* Pan population root samples from ten citrus trees were collected during the middle of each month from July 1983 to July 1984. The roots were thoroughly washed free from adhering soil and two sub-samples of 2 g of

rootlets were taken from each plant. The roots from one sub-sample were carefully dissected and the nematodes of all stages and egg-sacs from the galls were removed and counted. The second sub-sample of roots was stained with lactophenol to identify the presence of nematodes in the root tissue. For the months in which the population density was high, parts of the root and root-galls were selected, dissected separately, and all development stages of *M. fujianensis* were counted.

Representative samples of all weed species growing in the orange grove were collected each month. The *Meloidogyne* spp. present on each weed species were identified by directly dissecting egg-producing females and their corresponding egg-sacs from the roots. Second-stage juveniles and males were obtained by hatching individual egg-sacs under water. Half of the recovered specimens were preserved in 4% Formalin solution. The remaining specimens were used for taxonomic identification in which female body shape, cuticular pattern, distance of the excretory pore from the anterior end, female stylet length, and juvenile body length were recorded.

Results and discussion

Dissection of 250 g of roots, collected monthly, from July to the following June, from ten *C. reticulata* trees produced a total of 21576 *M. fujianensis* nematodes (11096 mature females, 7835 males, 1667 J2s, and 2168 J3s and J4s) and 6776 egg-sacs.

The proportion of *C. reticulata* root samples containing *M. fujianensis* varied between 30% and 90%, with the smallest number being recorded in August and the largest the following April. The smallest average number of *M. fujianensis* females per root sample (1.8) was recorded in August and the largest (24.2) in the following February. Nematodes were present in the root samples throughout the year, but the peak periods of invasion occurred during Sep-

tember/October, and during March/April of the following year. During these periods more second (J2), third (J3) and fourth (J4) stage juveniles were present than at other sampling dates. The peak oviposition period was December and January with 87.2 egg-sacs/2 g roots and 150.2 egg-sacs/2 g roots, respectively.

During September many lateral roots were produced by the *C. reticulata* trees, which was associated with a large increase in the *M. fujianensis* population. On average, a 0.6 cm length of root contained five laterals (fibrous roots) on each side. Many of these root segments contained root galls, frequently occurring as six small galls side by side, each approximately 0.35 cm in length and 0.2 cm in diameter. Four laterals were randomly selected and dissected. A sex differentiation phenomenon was observed as the nematodes present in the galls from a single lateral were either mainly (>80%) male, or were mainly (>80%) female. This phenomenon was frequently observed at the other sampling dates.

In September, seven galls chosen at random from *C. reticulata* tree 5 contained a mean of 15.9 ± 5.6 (5-23) *M. fujianensis* nematodes per gall, comprising 0.4 ± 0.5 (0-1) females, 1.0 ± 1.5 (0-4) males, 5.9 ± 6.4 (0-19) J2s, 3.7 ± 2.4 (2-8) pre-female J3s and 4.9 ± 3.8 (0-10) pre-male J3s. In October, ten galls from *C. reticulata* tree 8 contained a mean of 3.9 ± 9.0 (0-29) females, 54.2 ± 60.8 (0-199) males, 3.5 ± 3.6 (0-10) J2s, 1.2 ± 2.4 (0-7) pre-female J3s, 8.9 ± 11.3 (0-33) pre-male J3s, and 0.5 ± 1.6 (0-5) pre-female J4s.

The volume of the egg-sac matrix produced by *M. fujianensis* is 3 to 5 times that of the female and appears to be characteristic of the species. A consequence of the large size of the egg-sac is that the female is completely submerged within the matrix; however, there are only a few eggs in the egg sac matrix and they are situated near the vulva of the female. Ten egg-sacs from the roots of *C. reticulata* were examined and only half were found to contain eggs (108.2 ± 139.1 (5-353) eggs).

Seven weed species growing in the citrus grove were examined for the presence of *Meloidogyne* nematodes: *Alternanthera sessilis* (L.) D.C., *Hurdannia nudiflora* (L.) Brenan, *Imperata cylindrica* (L.) Breauv., *Siegesbeckia orientalis* L., *Solanum nigrum* L. var. *pauciflorum* Liou, *Solanum verbascifolium* L., and *Urena lobata* L. *Meloidogyne* nematodes were present on all weed species examined but morphological examination of these nematodes revealed that *M. fujianensis* was exclusively present on *I. cylindrica*, whereas *M. incognita* and *M. javanica*, but not *M. fujianensis*, were present on the other six weed species.

Our study revealed that the population of *M. fujianensis* parasitizing *C. reticulata* increased as a direct consequence of increasing average monthly air temperature, but that two months elapsed between the increase in temperature and the consequent increase in the nematode population density. From July to December, the air temperature was continuously high (averaging 800 day °C, above 0 °C, per month) and in December, two months after the highest monthly figure of 891.3 day °C, the population density reached its peak of 94.3 females/2 g roots. From January to June, the air temperature was continuously low (average <500 day °C) and in April, two months after the lowest monthly figure of 308.4 day °C, the population was of 117.6 females/2 g roots. The ratio of males to females in the *M. fujianensis* population was observed to decrease with increasing rainfall.

Citrus reticulata trees produced flushes of new root growth in May/June and again during September/October, coincident with the highest population densities of *M. fujianensis* and the appearance of disease symptoms. During March/April diseased trees had a slow budburst, the new-season growth turned green much more slowly than occurred in healthy trees and young trees exhibited early flower drop. During September/October diseased trees showed symptoms similar to those asso-

ciated with water and fertiliser stress e.g., leaf yellowing, early ripening and falling of fruit, and small misshapen, poor quality fruits. The larger nematode population densities were associated with greater severity of disease symptoms.

In separate studies the life-cycle of *M. fujianensis* on *C. reticulata* was found to take 55-60 days at 25 °C, with the period from root invasion by the J2 nematodes to mature female and egg production requiring 30 to 35 days (unpublished results). Similarly, in our study two months elapsed between the peak of root invasion by *M. fujianensis* and the peak in oviposition, expressed as eggs/female. Peak root invasion occurred during September (65.1±92.2 (0-257) juveniles/2 g roots) and two months later, in November, peak oviposition occurred, 1.2 eggs/female. Our study also revealed that new root production was as important, if not more so, than temperature as similar maximum numbers of nematodes were recorded in September and March. These coincided with new root production, but the monthly temperatures provided 891.3 and 409 day °C, respectively, a difference between the two months of 481.3 day °C.

Sex determination in *Meloidogyne* nematodes is influenced by environmental factors (Taylor and Sasser, 1978). During periods of low rainfall in our study the *C. reticulata* trees suffered from water stress which would cause a resulting diminution of food quality available to the nematodes invading and developing in the roots of the trees. Thus, at these times more of the nematodes would develop to males, whereas when rainfall was abundant, and the trees were not stressed, more females developed.

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