Project Number: 87-W1
Primary Researcher: Ramming
Secondary Researchers: Culver and McKenry
Title of Project: Rootstock Resistance to Lesion and Ring Nematodes

Summary of research on the development of *Prunus* rootstocks resistant/tolerant to the root lesion nematode, *Pratylenchus vulnus*, and the ring nematode, *Criconemella xenoplax* conducted from January 1 to December 1, 1987

Prepared by

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and

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I. Primary Greenhouse Screening Genotypes for Root Lesion Nematode Resistance.

Using the primary screening procedures developed in 1986, the following genotypes were screened in 1987:

Screen #1	Screen #2
1) Dura	1) P114-37
2) P70-101	2) P114-42
3) Fla 9-4	3) P114-47-84L
4) Fla 14-25	4) P114-47-85S
5) Fla 14-11	5) P114-52
6) Nemaguard	6) P114-55
	7) P114-62
	8) P114-66
	9) P114-84
	10) Nemaguard

Screen #1 was inoculated on April 28, 1987 and evaluated on June 12, 1987 (45 DAT); P70-101 and Nemaguard were also evaluated on July 9, 1987 (90 DAT). Screen #2 was inoculated on June 5, 1987 and evaluated July 22, 1987 (47 DAT). The results of those screens are shown in Table 1. P70-101 had somewhat reduced counts at 45 DAT, but the evaluation at 90 DAT showed no differences from the standard, Nemaguard. In Screen #2, all of the Israeli almond genotypes were highly susceptible to attack and establishment by root lesion nematode.

Table 1. Results of root lesion nematode primary screen of genotypes conducted in 1987.

Screen #1[1/]

	Total nematodes		Nematodes/g. of root	
Genotype	45 DAT	90 DAT	45 DAT	90 DAT
Dura	300.8	-	107.8	-
P70-101	107.8	413.2	34.1	74.8
Fla 14-25	276.4	-	93.7	-
Fla 14-11	242.4	-	98.7	-
Fla 9-4	283.6	-	107.8	-
Nemaguard	195.0	419.6	47.3	60.0

Screen #2[2/]

	Total nematodes	Nematodes/g. of root
Genotype	47 DAT	47 DAT
P114-37	507.2	214.9
P114-42	639.6	256.9
P114-47-84L	355.4	133.1
P114-47-85S	228.6	150.4
P114-52	407.6	107.5
P114-55	564.4	244.3
P114-62	433.0	253.2
P114-66	276.6	78.4
P114-84	414.4	114.2
Nemaguard	54.4	6.1

1/ Inoculated with 150 vermiform larvae/plant, 5 plants/genotype, on April 28, 1987.

2/ Inoculated with 150 vermiform/plant, 5 plant/genotypes, on June 5, 1987.

The following genotypes have been rooted and should be run through the root lesion screen in August:

1) P70-95

2) P70-100

3) P70-107

4) P70-135

5) P70-141

6) P70-149

7) Marianna 2624 (tolerant)

8) Nemaguard (standard)

The P70-107 is being run through the screen a second time to test the reproducibility of screen results. P70-107 showed good resistance in the 1986 fall primary screen, but showed no resistance in the secondary screen in 1987 (see Section II). It should be determined whether the primary screen results were not accurate for P70-107 or whether the resistance of the genotype was, in fact, lost over time.

II. Secondary Greenhouse Screening Genotypes for Root Lesion Nematode Resistance/Tolerance.

Based on the results from the primary greenhouse screen of genotypes in 1986, the following inoculated genotypes were moved up from 2 3/8" x 2 3/8" x 5" bands to 6" pots in February of 1987:

1) P70-107

2) P. tomentosa "Orient"

3) P. japonica

4) Citation

5) P70-133

6) P70-139

7) P70-150

8) Nemaguard

9) Myrobalan 29C

Genotypes 1 through 4 were selected because they demonstrated possible resistance. Genotypes 5 through 7 were selected because they demonstrated possible tolerance. Genotypes 8 and 9 were the susceptible standard and the tolerant standard, respectively. An evaluation was made on July 1 and October 30, 1987 and the results of that evaluation are shown in Table 2. Total root weights may provide an indication of dwarfing effects.

Table 2. Results of root lesion nematode secondary screen of genotypes conducted in 1987[1/].

	Total root weight	Nematodes/g. of root	
Genotype	October 30	July 1	October 30
P70-107	88	157.8	243
P. tomentosa "Orient"	26.6	1.9	0.24
P. japonica	20.9	4.5	3.0
Citation	52.1	168.8	149
P70-133	92.8	241.4	249
P70-139	48.3	298.3	221
P70-150	53.8	249.5	180
Nemaguard	83.5	379.7	152
Myrobalan 29C	98.5	85.0	345

1/ Genotypes were initially inoculated with 150 vermiform larvae/plant on 9/10/86 on a primary screen. Plants were moved up to 6" pots in February, 1987.

The primary screen for these genotypes indicated that the most resistant genotype was P70-107. However, this secondary screen indicated that, over time, P70-107 had become as susceptible as the standard, Nemaguard. P. tomentosa "Orient" and P. japonica were clearly resistant to root lesion nematode. The growth of Citation appeared to be significantly affected by root lesion nematode, although no measurements of infected and non-infected plants were taken. These results were further verified with nematode counts and growth measurements on October 30, 1987.

III. Initial Results of Tests Designed to Identify the Procedures for Screening for Ring Nematode Resistance.

In these trials, genotypes were selected that had been grown in 1986 in the above-noted bands in two different media. These media were: 1) Mix (2:1:1, peat:vermiculite:sand), and 2) soil (3:1, Kearney Agricultural Center sandy loam soil:sand). Test plants were moved up to 6" pots with all additional soil being sand. The genotypes selected and the band media they were in were as follows:

1) "Nemaguard" peach in soil
2) "Nemaguard" peach in mix
3) P. tomentosa "Orient" in mix
4) Pistacia atlantica in soil
5) Pistacia atlantica in mix

There were ten plants of each combination set up on April 10, 1987. The plants were moved into the 6" pots and immediately inoculated with 35 cc of sandy soil which contained approximately 500 ring nematode motile forms. A separate group of five "Nemaguard" peach in soil was inoculated with 2,500 ring

nematode motile forms. The infected soil was placed in a trough around the plant after it had been planted into the 6" pot and water applied in the trough. At 90 DAT, a 250 cc soil sample was extracted from each plant for each treatment. The results are shown in Table 3.

Table 3. Ring nematode motile counts taken 90 and 180 DAT.

Treatment	Ring nematode motile 250 cc[1/] 7/9/87 (90 DAT)	10/7/87 (180 DAT)
"Nemaguard" in soil	58.7	132
"Nemaguard" in mix	29.6	19.4
P. tomentosa "Orient" in mix	14.4	34.3
Pistacia atlantica in soil	44.0	42.6
Pistacia atlantica in mix	13.9	45.8
"Nemaguard" in soil (high inoculum level)[2/]	167.2	425

1/ Numbers take into account a 75% extraction efficiency. 90 DAT = July 9, 1987, 180 DAT = October 7, 1987. Original inoculum was approximately 500 motile forms of ring nematode/plant.

2/ Original inoculum was approximately 2,500 motile forms of ring nematode/plant.

As can be seen, the numbers of ring nematode were disappointingly low. However, in referring to the work of Nyczepir et al. (1987) with inoculation levels of ring nematode and population increases over time, several procedural differences came to light. To summarize, Nyczepir et al. planted "Nemaguard"

seed into loamy sand (86% sand) and inoculated 12 days after emergence. Even at inoculation rates of 14,000+ ring nematodes per 6" pot, no growth effects were seen at 90 DAT. However, at an inoculation rate similar to ours (448 nematodes/pot), there was a seven-fold population increase seen at 90 DAT which did not occur in our test. We will evaluate our test once more at 180 DAT to see if populations have increased. The procedures of Nyczepir, et al. should be incorporated in future testing for developing a ring nematode screen.

References

1) Nyczepir, A.P, C.C. Reilly, and W.R. Okie. 1987. Effect of Initial Population Density of Criconemella xenoplax on Reducing Sugars, Free Amino Acids, and Survival of Peach Seedlings over Time. J. Nemol. 19(3):296-303.

Proposed Trials for 1988

I. Ring Nematode Screen

A.R. Nyczepir, C.C. Reilly, and W.R. Okie recently reported results of the effect of various populations of ring nematode, Criconemella xenoplax, on "Nemaguard" peach seedlings (4). It is recommended that the basic methodology employed in these trials be employed in the screen of Prunus genotypes for resistance to ring nematode. Specifically, it appears that the following criteria should be used in the screen:

1) Test soil should be 85-90% sand.
2) Plants should be healthy, but with a minimum amount of roots.
3) Initial population density of C. xenoplax per 1500 cm^3 soil should be 8,000 to 10,000.

Based on the results of Nyczepir et al. with Nemaguard, significant effects on percent survival, dry root weight, and height increase of susceptible Prunus genotypes will occur using the above noted test conditions 180 days after inoculation.

The initial genotypes to test should be "Nemaguard" peach (check), S60 selection(s) (as determined by M.V. McKenry), and Lovell selection(s) (as determined by M.V. McKenry). Genotypes should be propagated by rooting cuttings and tested in 6" pots. Additional tests should be conducted on P. tomentosa "Orient" and P. japonica.

II. Field Testing of P. tomentosa "Orient" and P. japonica

Using the field screening methodology developed in 1985, a trial should be conducted to evaluate the resistance of P. tomentosa "Orient" and P. japonica to lesion nematode, P. vulnus, using "Nemaguard" peach as the susceptible genotype.

III. Status of Selections to Screen for Lesion and Ring Nematodes

Budwood of the following genotypes was supplied by Dr. W.R. Okie, USDA-ARS, Byron, Georgia, in July 1987 and given to Burchell Nursery for propagation on Nemaguard seedlings:

Genotype	Description
1) TNR 2	Tennessee Natural selection
2) 14 DR 51	Read leaf selection originally isolated by Fogle
3) PI 442380	Evergreen peach seedling from Mexico
4) FL 4-1-11	o-p selection from Fla 14-9 peach
5) FL 2-1-11	o-p selection from Fla 14-9 peach
6) FL 4-3-16	o-p selection from Fla 14-18 peach
7) FL 2-3-16	o-p selection from Fla 14-18 peach
8) SL 2760	o-p Blue Goose (plum)
9) SL 1410	o-p Blue Goose (plum)
10) SL 1665	o-p Satsuma
11) SL 1089	o-p 520-9 peach (from o-p Nemaguard)
12) SL 1090	o-p 520-9 peach (from o-p Nemaguard)
13) SL 1990	o-p Edible Sloe (plum)
14) SL 1171	o-p 7446-14 (plum)

All of these genotypes appear to have varying degrees of resistance/tolerance to ring nematode in the field based primarily on percent survival in soil having high populations of ring nematode and a high incidence of peach tree short-life (PTSL). There are two other genotypes that need to be obtained from Dr. Okie which have been outstanding in his field trials. These are PI82413 (Baladi #1) and PI134151 (Transvaal Yellows).

Another group of genotypes worth evaluating were originally tested by Day and Serr in 1953 (1). These were field evaluations of genotypes for resistance to root lesion nematode. The more outstanding genotypes were:

Genotype	Description
1) Bokhara	Peach seedling
2) Yunnan 55885	Peach seedling
3) Macedonian Wild	*P. cerasifera*?
4) Etter's Best	*P. subcordata* x *P. domestica*?
5) Bruce	*P. salicina* x *P. angustifolia*

Lastly, there are 4 genotypes identified in the literature which appear to have good resistance or tolerance to root lesion nematode. These are the following:

Genotype	Description
1) G_2S2544-1-2	South Korean peach
2) P2037	*P. besseyi* x Myrobalan?
3) P322 x P871	Myrobalan x (Myrobalan x peach)
4) S.B./II	Italian peach seedling?

The first three genotypes were reported by La Massese in 1975 (2) and the S.B./II was reported by Manzo in 1978 (3).

IV. Screening the S60 and Lovell Selections of Dr. M.V. McKenry for Resistance/Tolerance to Root Lesion Nematode

Dr. McKenry will be generating selections of S60 and Lovell peaches which will have performed well in his root knot nematode field screening trials. These genotypes should be run through the greenhouse root lesion nematode screen at the USDA-ARS, Fresno facility.

References

1) Day, L.H. and E.F. Serr. 1953. Comparative resistance of rootstocks of fruit and nut trees to attack by a root-lesion or meadow nematode. Proc. Amer. Soc. Hort. Sci. 57:150-154.

2) La Massese, C.S. 1975. Tests d'hotes de quelaues porte-greffe et varietes fruitieres a l'egard de Pratylenchus vulnus Allen et Jensen. Comptes Rendus des Seances de l'Academie d'Agriculture de France 61(17):1088-1095.

3) Manzo, P. 1978. Comparison among different peach rootstocks in fumigated and non-fumigated replanted soil. Note II. Annali dell' Istituto Sperimentale per la Frutticoltura 9:49-56.

4) Nyczepir, A.P., C.C. Reilly, and W.R. Okie. 1987. Effect of initial population density of Criconemella xenoplax on reducing sugars, free amino acids, and survival of peach seedlings over time. J. of Nematology 19(3):296-303.

AES/CE MAR 84 Workgroup/Department _____

University of California
Division of Agricultural Sciences

Project Plan/Research Grant Proposal

Project Year 1988 Anticipated Duration of Project _____

Project Leader Dave Ramming Location USDA, Fresno; Kearney Agric. Center

Cooperating Personnel M.V. McKenry

Project Title Development of Prunus Rootstocks Resistant/Tolerant to Root Lesion and Ring Nematode

Keywords _____

Commodity(s) California Almond, Cling Peach, Prune, Fresh Tree Fruit Agreement Relevant AES/CE Project No. _____

Problem and Its Significance:

The root lesion nematode (Pratylenchus vulnus) and ring nematode (Criconemella xenoplax) are prevalent in many regions of stone fruit and almond production due to the predominant use of rootstocks which are susceptible to these nematodes. Nemaguard, Lovell, Marianna 2624 are susceptible to root lesion nematode while Marianna 2624 is a tolerant host. A recent survey in almonds indicated that sandy soils, including more than half the acreage in San Joaquin, Stanislaus and Merced Counties, are infested with relatively high population levels of ring nematode. More than 25 percent of the almond orchards statewide also have root lesion nematode. These two nematodes can occur together. The association of ring nematode with the bacterial canker complex provides good symptomology for identification of the fields where ring nematode occurs. Where there are high infestation levels of root lesion nematode, Dr. McKenry has found yield reductions of 23 percent and 14 percent in plums on Nemaguard and Lovell, respectively. Significant reductions in the levels of calcium in plum leaves of trees infected with root lesion nematode were also noted. CONTINUED ON OTHER SIDE

Objectives:

(1) Develop protocols for testing rootstock material for resistance/tolerance to lesion and ring nematodes. (2) Identify candidate rootstock material that is resistant/tolerant to lesion and ring nematode.

Plans and Procedures:

Candidate rootstock materials are presently being tested in the greenhouse for lesion nematode resistance/tolerance using nematode inoculation and evaluation techniques validated in research conducted in 1986. This screening program is for root lesion nematode only. However, the technology should be applicable to the identification of resistance/tolerance to ring nematode also. Rootstock materials that appear promising in the greenhouse in terms of root lesion resistance/tolerance will be taken to the field and tested. Again, the technology should be applicable to field testing for resistance/tolerance to ring nematode.

CONTINUED ON OTHER SIDE

BUDGET REQUEST Budget Year 1988

Funding Source California Almond Board, California Tree Fruit Agreement, Prune Board, Cling Peach Board

Salaries and Benefits		
Postdocs/RA's		
SRA's		
Lab/Field Assistance		
Subtotal		Sub 2
Employee benefits		Sub 6
		TOTAL
Supplies and Expenses		Sub 3
Equipment		Sub 4
Operating Expenses and Equipment Travel (Davis campus only)		Sub 5
Travel		Sub 7
		TOTAL $38,182

Department account number

(continued from front)

PROBLEM AND ITS SIGNIFICANCE (continued): With the recent detection of a number of soil applied pesticides in groundwater in California and other states, there is now a genuine concern expressed by EPA, CDFA and the populace in general (as evidenced by the overwhelming passage of Proposition 65) about the contamination of ground water with pesticides. The most efficient and environmentally sound solution to these problems is to develop nematode resistant rootstocks. It should be noted that there is already excellent control for root-knot nematode in the industry through the use of the resistant rootstocks Nemaguard and Marianna 2624.

PLANS AND PROCEDURES (continued): Nematode inoculation rates for screening in the greenhouse and field have been established for root lesion nematode but not for ring nematode.

The primary criteria used in the root lesion nematode screen are nematodes/gram of root, root weight and the changes in the levels of calcium and magnesium in plant tissues, particularly in the roots. Root weight and nematode counts in the soil will be taken in the initial ring nematode screening trials to identify the most reliable criteria to use in the ring nematode screening program.

The identification of the inoculum rates and evaluation criteria to use for the ring nematode screen will involve collecting biological measurements from three rootstock selections with known or suspected relative susceptibilities. Since ring nematode is an ectoparasite, nematode counts from the soil will be necessary as will measurements of fibrous root differences. See attached our specific plans for 1988.

Michael V. McKenry for Dave Ramming Date 12/10/87
Originator's Signature

funding to be submitted to USDA, Fresno as in 1987

COOPERATIVE EXTENSION
County Director Date
Program Director Date

AGRICULTURAL EXPERIMENT STATION
Department Chair Date

LIAISON OFFICER Date

D2454-2 (1/84)