To: California Pepper Commission

RE: Research Report for 2018-19

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Project Title: Efficacy of novel nematicides and plant resistance against root-knot nematodes on bell peppers.

Statement of the problem and background.

The Southern root-knot nematode (*Meloidogyne incognita*) has been reported to cause serious damage to peppers. The second-stage nematodes (J2) are worm-shaped, move through the soil, and enter the plant roots. In roots of a host crop, the second-stage juveniles nematodes develop into females, while the root system responds to infection with the formation of galls. The fully developed females can produce up to 400 eggs, that are contained in clusters in a gelatinous material and "glued' to the outside of the roots. From these eggs second-stage juveniles can emerge to repeat the cycle, or eggs can remain in the soil during fallow period to serve as inoculum for the next crop cycle. The duration of the nematode life cycle depends primarily on the species of root-knot nematode and on soil temperature. *Meloidogyne incognita*, the most important species infecting bell-pepper, can complete its' life cycle in less than 4 weeks under an optimum soil temperature of 32C (90F), and become inactive when the soil temperature drops below 17C (62F). In most host crops, root-knot nematode infestation can easily be diagnosed because of obvious galling on the affected roots. Above-ground symptoms are however not specific, and can include chlorosis, wilting under sufficient soil moisture, stunting, and increased susceptibility of plants to fungal or bacterial root pathogens.

In the Coachella Valley of Southern California, approximately 5,000 acres are cropped with bell peppers, representing an estimated gross crop value of $90,000,000. Root-knot nematodes are widespread throughout the Coachella Valley and growers report serious damage. To control nematodes, pepper growers in the Coachella Valley commonly apply fumigant nematicides such as metam-sodium (Vapam) or 1,3-dichloropropene (Telone) as a post-harvest and/or pre-plant soil treatment through the drip tubing.

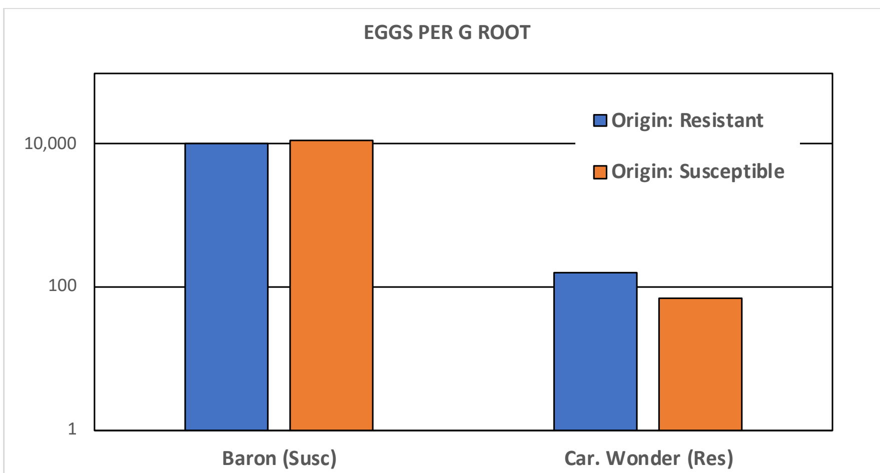
In our previous studies, also funded by the CPC, we initially found that root-knot nematodes (*M. incognita*) were relatively common in pepper fields in the Coachella Valley. Soil and root infestation levels were sometimes very high. In greenhouse pot experiments, using nematode populations isolated from Coachella pepper fields, we found that of two nematode-resistant pepper varieties tested (Charleston Belle and Carolina Wonder), the Carolina Wonder pepper was highly resistant even under high nematode inoculum levels.

In 2017 trials, these two pepper varieties were grown at two locations (CVARS-Coachella and SCREC-Irvine). The trial set-up at both locations was similar, with various novel nematicides applied pre-plant. In summary, our results indicated that the nematicides had little effect on pepper yields. At CVARS, the development nematicide Salibro resulted in a slight reduction in root galling and nematode root infestation, but this was not the case at SCREC. A surprising result was obtained at CVARS, where the resistant cultivar Carolina Wonder still exhibited substantial root galling and nematode root infestation. At SCREC, this resistant cultivar remained virtually free of galls and nematodes. Our initial speculation was that this might be related to high(er) soil temperatures at CVARS compared to SCREC.

2018 Greenhouse Trials.

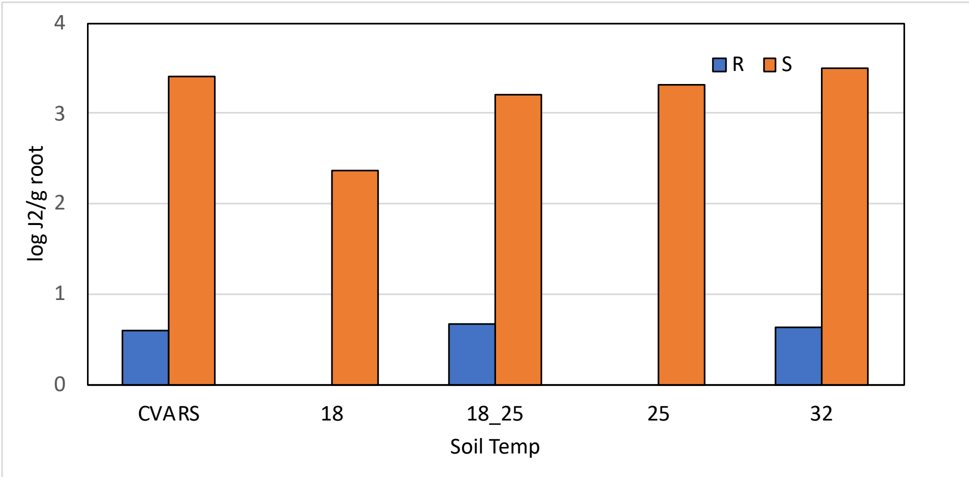
Goal of initial greenhouse trials was to determine if the breaking of resistance observed at CVARS was due to the occurrence or development of a resistance-breaking nematode population at this site. Roots from the susceptible ‘Baron’ and the resistant ‘Carolina Wonder’ from the 2017 CVARS field trial were collected and taken to the UCR Nematology lab. Second-stage juveniles of the nematodes (J2) were extracted from the roots of both cultivars, and inoculated onto greenhouse grown ‘Baron’ and ‘Carolina Wonder’ pepper plants in pots (10,000 j2/pot) with steam-sterilized sandy soil (n=5). After 8 weeks, nematode eggs were extracted from the roots and counted. The results showed that there was no difference between the nematodes that had come from the field-grown resistant or susceptible roots. They were able to reproduce on the susceptible ‘Baron’ but not on the resistant ‘Carolina Wonder’. Therefor, it was concluded that the nematode population at CVARS had not developed the ability to break resistance in ‘Carolina Wonder’.

Figure 1. Greenhouse pot trial: Egg numbers on susceptible Baron and resistant Carolina Wonder when inoculated with nematodes originating from Baron or Carolina Wonder roots from 2017 CVARS field trial.



To test if soil temperatures affect the resistance level of ‘Carolina Wonder’ a set of experiments was done with closed pots placed in waterbaths running at different temperature regimes. ‘Baron’ and ‘Carolina Wonder’ peppers were grown in closed pots, inoculated with nematodes (*M. incognita*) and pots were then placed in the waterbaths. Pot soil temperatures were a constant 18C (cool), constant 25C (medium), constant 32 (warm), a cool to medium change (18-25), and a CVARS simulation temperature sequence. At the end of the trial, root-galling and nematode root levels were determined. The results showed that at none of the temperatures resistance in ‘Carolina Wonder’ was compromised.

Figure 2. Nematode infestation (J2/g root) on roots of R (Carolina Wonder) and S (Baron) pepper grown at different soil temperatures.



Material and Methods 2018

Field Trials.

Identical field trials were done on root-knot nematode (*M. incognita*) infested field sites at two locations: CVARS (Coachella Valley Agricultural Research Station) and SCREC (SouthCoast Research and Experiment Station, Irvine).

Each site had 70 plots. Plots consisted of 20 ft long sections of 60-inch-wide (CVARS) or 40-inch-wide (SCREC) sections of beds. There were seven treatments:

1) Untreated control
2) Metam-sodium @ 75 gallon/acre, drench incorporated, 3 wk pre-plant
3) Salibro @ 30.7 oz/acre drench incorporated, 1 wk pre-plant
4) Salibro @ 46.2 oz/acre drench incorporated, 1 wk pre-plant
5) Salibro @ 61.4.7 oz/acre drench incorporated, 1 wk pre-plant
6) Nimitz @ 5 pt/acre drench incorporated, 1 wk pre-plant
7) Velum One @ 13 oz/acre drench incorporated, 1 wk pre-plant
8)

Field was designed according to a completely randomized block split-plot design with 5 replicates. Main treatments were nematicide treatments, sub-treatments were pepper variety: susceptible pepper 'Baron' and resistant 'Carolina Wonder'. Transplants of were grown in a UCR greenhouse and planted in the plots on 3/16/2018 (CVARS) and 6/19/2018 (SCREC). Plants were planted in one line per bed per bed , at 16 inch spacing, with resistant and susceptible varieties in adjacent beds for each nematicide treatment. Watering and fertigation was through buried drip.

Data collected were pre-treatment soil root-knot nematode levels, galling of pepper root systems at harvest, fruit yield at harvest, and nematode infestation levels of the roots.

Results

CVARS:

Pre-treatment root-knot nematode levels (J2/100 g soil) were low 8(J2/100 g) and not significantly different between the treatments. At-harvest soil nematode levels were not different between the nematicide treatments, but were very much affected by the pepper variety. Under the susceptible ‘Baron’, soil nematode numbers were 3,010 J2/100 g soil, whereas after the resistant ‘Carolina Wonder’ only 1 J2/100g soil was detected. The pepper variety had similar dramatic effects on the number of nematode eggs per root system, and on root galling. Of the nematicides, all three Salibro treatments reduced galling compared to the untreated control in the susceptible peppers.

Table 1. Effect of Nematicide Treatments and Resistant (R) and Susceptible (S) Bell Pepper: nematode effects.

	Pi		**Pf**		**eggs/g root**		**galling**	
Nematicide	**R**	**S**	**R**	**S**	**R**	**S**	**R**	**S**
UTC	8	5	1	5,720	6	14,845	0.0	5.7A
VPAM	3	9	3	3,560	0	19,765	0.0	6.0A
SALIBRO LOW	8	5	0	1,330	0	10,771	0.0	3.2C
SALIBRO MED	7	8	1	1,537	8	5,765	0.2	3.0C
SALIBRO HIGH	13	10	1	1,350	0	8,094	0.0	3.4BC
NIMITZ	9	11	0	3,820	0	15,440	0.0	5.7A
VELUM	8	10	0	3,750	0	11,048	0.0	5.2AB
average	**8**	**8**	**1 b**	**3,010 a**	**2 b**	**12,247 a**	**0.0b**	**4.6a**

There were no significant treatment effects on yields.

Table 2. Effect of Nematicide Treatments and Resistant (R) and Susceptible (S) Bell Pepper: yield.

	kg fruit per plant		
Nematicide	**R**	**S**	**average**
UTC	0.4	0.3	0.4
VPAM	0.4	0.4	0.4
SALIBRO LOW	0.3	0.4	0.4
SALIBRO MED	0.3	0.3	0.3
SALIBRO HIGH	0.3	0.3	0.3
NIMITZ	0.4	0.3	0.3
VELUM	0.3	0.3	0.3
average	0.3	0.3	0.3

SCREC: At the SCREC station, pre-treatment root-knot nematode levels were moderate (40 J2/100 g) and not significantly different between the treatments. At-harvest soil nematode levels were not different between the nematicide treatments, but like at CVARS, were affected by the pepper variety. Under the susceptible ‘Baron’, soil nematode numbers were 869 J2/100 g soil, whereas after the resistant ‘Carolina Wonder’ this was 106 J2/100g soil was detected. At SCREC, the pepper variety also affected the number of nematode eggs per root system, and root galling. There were no significant effects of the nematicides.

Table 3. Effect of Nematicide Treatments and Resistant (R) and Susceptible (S) Bell Pepper: nematode effects.

	Pi		Pf		eggs/g root		galling	
Nematicide	**R**	**S**	**R**	**S**	**R**	**S**	**R**	**S**
UTC	20	22	115	555	254	6,304	0.3	7.1
VPAM	21	49	125	825	175	6,697	0.3	6.7
SALIBRO LOW	29	89	125	1245	50	7,713	0.7	6.5
SALIBRO MED	68	18	100	590	197	8,536	0.6	6.1
SALIBRO HIGH	24	13	150	860	101	7,227	0.2	6.0
NIMITZ	39	38	75	1295	107	8,583	0.2	6.9
VELUM	80	54	50	705	78	9,397	0.4	5.7
average	40	41	106 b	869 a	137 b	7,811 a	0.4 b	6.4 a

At SCREC, where initial nematode levels were higher than at CVARS, the resistant peppers yielded significantly more than the susceptible peppers. In the susceptible peppers, the untreated control treatment had the lowest yield, but differences were not significant at the 95% confidence level.

Table 4. Effect of Nematicide Treatments and Resistant (R) and Susceptible (S) Bell Pepper: yield.

	kg fruit per plant		
Nematicide	**R**	**S**	**average**
UTC	1.1	0.5	0.8
VPAM	1.1	0.7	0.9
SALIBRO LOW	1.0	0.7	0.9
SALIBRO MED	1.0	0.6	0.8
SALIBRO HIGH	0.9	0.8	0.9
NIMITZ	1.0	0.7	0.9
VELUM	0.9	0.6	0.8
average	1.0 a	0.7 b	0.8 ns

Discussion & Conclusion

The initial hypothesis that the infestation of resistant peppers at CVARS in 2017 could have been due to high soil temperatures, or the presence of a resistance-breaking nematode population could not be confirmed in greenhouse pot trials. Also, in our 2018 trials, the resistant peppers remained virtually free of nematode (symptoms) at CVARS, or had very low nematode infestation levels (SCREC). There were some indications that the nematicide Salibro may have some benefit as it significantly reduced root-galling in the susceptible variety, and also resulted in the lowest at-harvest nematode levels at CVARS. At SCREC, where nematode pressure was moderately high, the resistant variety 'Carolina Wonder' yielded significantly more than the susceptible 'Baron'. We will continue to explore the potential of nematode resistance in pepper. Basic knowledge about the relationship between initial nematode levels and crop yield is important for deciding on the use of nematode control strategies. Such data are not available for root-knot nematode and pepper in California. For 2019 we propose to study this relationship for a number of different California pepper varieties both under controlled greenhouse conditions, and in field trials.