

#### **Chapter 4: The Damage Potential of the Tobacco Cyst Nematode on Several Economically Important Hosts.**

*Abstract*-The tobacco cyst nematode (*Globodera tabacum solanacearum*) continues to spread south in the United States. Pepper, tomato, and eggplant have been reported to be hosts of the tobacco cyst nematode, but field research has not been conducted to investigate the effect of TCN on these three crops. In this study, pepper, tomato, eggplant, and TCN-susceptible (K326) and TCN-resistant (NC567) tobacco cultivars were planted in TCN infested soil. Half of the field was fumigated with 1,3-dichloropropene to compare yield and nematode reproduction between treated and non-treated areas. All three vegetable crops were hosts of TCN, with greater reproduction occurring on tomato and eggplant than on pepper. Eggplant was very sensitive to TCN infection, experiencing a 64% yield reduction in non-treated plots compared to fumigated ones. Pepper and tomato were more tolerant of TCN infection, as yields from non-treated plots were not significantly different from treated plots. This experiment needs to be repeated to verify these conclusions.

*Key terms*-Tobacco cyst nematode, pepper, eggplant, tomato, reproduction, fumigation, 1,3-dichloropropene

Tobacco cyst nematodes are divided into three subspecies. *Globodera tabacum tabacum* (Lownesbery and Lownesbery, 1954), Behrens, 1975 is a parasite of shade tobacco (*Nicotiana tabacum* L.) in Connecticut. *Globodera tabacum virginiae* (Miller and Gray, 1968), Behrens, 1975, is a parasite on wild solanaceous weeds in southeastern Virginia. *Globodera tabacum solanacearum* (Miller and Gray, 1972), Behrens, 1975, is a pathogen of flue-cured and dark-fired tobacco in Virginia, North Carolina, and one Maryland-type tobacco farm in Maryland (Johnson, 1998).

*Globodera tabacum solanacearum* (TCN) causes extensive damage on flue-cured tobacco in Virginia. Estimates place average annual losses at 15% in infested fields (Komm *et al.*, 1983); however, complete crop losses have been reported (Komm *et al.*, 1983). *Globodera tabacum solanacearum* also parasitizes many species of the *Solanum* and *Nicotiana* genera (Baldwin and Mundo-Ocampo, 1991). Most alternate hosts of TCN are weeds and other species of little economic importance, but TCN can parasitize other types of tobacco, eggplant (*Solanum melongena* L.), pepper (*Capsicum* spp. L.), and tomato (*Lycopersicon* spp. L.) (Miller and Gray, 1972). Tobacco cyst nematodes were reported to be parasitic on flue-cured, burley, dark-fired, shade, and sun-cured tobacco (Miller and Gray, 1972). Harrison and Miller (1969) examined the host range of *G. t. tabacum* and concluded that eggplant and tomato were excellent hosts and pepper was a moderate host. Due to the close relation between these two nematode subspecies, TCN could produce similar effects on these hosts.

Originally described in the southern piedmont area of Virginia (Osborne, 1961), TCN continues to be found in new locations. The first report of TCN outside of Virginia was in Warren County, North Carolina (Melton and Phillips, 1991). The nematode has since been found in seven counties in North Carolina and threatens vegetable producing areas of North Carolina. However, the potential damage to eggplant, tomato, and pepper has not been verified. Limited research involving TCN and these crops has been conducted in the greenhouse, but no damage potential data has been collected from the field. The purpose of this research was to investigate the effect of TCN on the yield of pepper, tomato, and eggplant and to examine overall reproduction of TCN on these crops in the field.

### **Materials and Methods**

*Seedling Preparation*-The following cultivars were used: X<sub>3</sub>R Camelot hybrid bell pepper, Mountain Spring tomato, F<sub>1</sub> Classic eggplant, and K326 and NC567 flue-cured tobacco cultivars. All seedlings, except for tomato plants which were purchased commercially, were reared in 200-cell greenhouse styrofoam trays. At transplanting, vegetable and tobacco seedlings were six and seven weeks old, respectively. Transplants were set on 2 June 1998 in 12.2 m long rows, 1.2 m apart, with 0.5 m spacing between seedlings.

*Experimental design and nematicide treatments*-The experiment was conducted at the Virginia Polytechnic Institute and State University, Southern Piedmont Agricultural Research and Extension Center in Blackstone, Virginia. Soil in the experimental field was classified as an Appling sandy loam. A split-plot design was implemented with main

plots consisting of nematicide-treated or non-treated plots. Each of the five crops used in this experiment were randomized within the main plots. Four replications of each cultivar-nematicide treatment combination were used. Single plots consisted of two rows, one row for destructive sampling and the other for soil sampling and yield data.

1,3-dichloropropene (Telone II) (Dow Elanco Inc.) was broadcast applied on 29 April 1998 to appropriate plots using a tractor mounted broadcast fumigator at a rate of 140 L/ha. Fumigant was lightly incorporated immediately after nematicide application using a field cultivator. The entire field received 6.2 L/ha of pebulate incorporated into the soil using a field cultivator for weed control on 1 June 1998. Additionally, 0.047 Mg/ha of nitrogen was applied to the field on 1 June 1998. Additional nitrogen was applied on 12 June 1998 to bring total rates of nitrogen to 0.078 Mg/ha for tobacco and pepper and 0.14 Mg/ha for eggplant and tomato. All crops were cultivated on 25 June 1998 and all except pepper received a layby cultivation on 13 July 1998. The study was irrigated four times (7, 14, and 28 July and 5 August 1998). Crops were also sprayed with thiodan (23 June and 21 July 1998), carbaryl (31 July and 25 August), and acephate (applied on tobacco only on 11 June, 3 August, and 8 September 1998) for insect control during the season.

*Soil and root sampling*-Soil samples were taken from each plot prior to nematicide treatment (28 April), 25 June, 31 July, and after harvesting was complete (15 October). Approximately 24 soil cores (18 cm long X 2 cm diameter) were taken from each plot. Cysts were extracted from soil using a modified Fenwick Can and crushed

with a blender (Caswell *et al.*, 1985). Nematode eggs were stained with acid fuchsin (Daykin and Hussey, 1985).

One randomly-selected plant was sampled from each plot on 6 July and 9 October to examine the number of nematodes per gram of feeder root. Plants were removed using a shovel and taken back to the lab, where roots were weighed and processed according to Byrd *et al.* (1983). Nematodes in the roots were categorized into the following four cyst nematode life stages: vermiform, swollen, pyriform, and adult.

*Yield data*-Vegetables were harvested six times during the experiment: on 7, 17, and 25 August, and 2, 11, and 28 September, 1998. All ripe vegetables were hand harvested on each harvest date. No peppers were harvested on the September 11 harvest date due to lack of harvestable fruit. Harvested vegetables were counted and weighed for each plot. Tobacco was harvested as it ripened four times during the season. After curing, individual harvests were weighed for each plot and graded by United States Department of Agriculture (USDA) inspectors. Grades were assigned a numerical grade index for analytical purposes (Bowman *et al.*, 1988).

*Statistical Analysis*-Nematode data were log-transformed [ $\log_{10}(x+1)$ ] prior to statistical analysis. Analysis of variance was conducted to evaluate treatment effects (SAS Institute, 1989). The Waller-Duncan test was used to compare individual treatment effects (K-ratio = 100). Paired t-tests (significant at  $P \leq 0.05$ ) were conducted within each crop/cultivar to determine if nematicide treatment had effectively reduced nematode populations. Harvest data (number of fruits, total fruit weight, and average weight per fruit) was subjected to t-tests for each harvest date as well as over cumulative yield.

## Results

*TCN Egg counts*-No significant differences in nematode infestation level were present in either treatment or plant blocks prior to fumigation (28 April) (Table 4.1). Additionally, no differences were found in nematode numbers across crops, cultivars, or between nematicidal treatments at the second sampling date (25 June). However, more TCN eggs were recovered from plots planted in pepper than the other crops and cultivars on 31 July 1998. More nematode eggs were produced on the tobacco cultivars (K326 and NC567) than on tomato, but only K326 produced significantly more TCN eggs than eggplant. Significant differences were found on the final soil sampling date (15 October) in number of TCN eggs found in the soil according to plant. More TCN eggs were observed on 15 October in plots planted with tomato and the tobacco cultivar K326 than in those of the other crops. Additionally, more eggs were found to be in plots planted in eggplant than the tobacco cultivar NC567. No difference was found in the number of eggs in non-treated and nematicide-treated soil.

Reproductive ratios were calculated by dividing the later sampling dates over the initial population level (Table 4.1). No significant differences were found in  $P_2/P_i$  among crop or nematicidal treatments. However, a significantly higher  $P_3/P_i$  ratio was observed for pepper than for eggplant and tomato. Additionally, both tobacco cultivars produced a higher  $P_3/P_i$  ratio than tomato. Significant differences in  $P_i/P_i$  ratios were also detected among crops and cultivars, but not between fumigation treatments. The final reproductive ratio was higher for the tobacco cultivar K326 than for any of the other

crops and cultivars. Reproductive ratio at the end of the growing season was also higher for tomato than for pepper or the tobacco cultivar NC567.

*Nematode root counts*-No differences in nematodes per gram of feeder root were found among crops or cultivars at the 7 July sampling date (Table 4.2). Fumigation also had no effect on the number of vermiform, pyriform, adult, or total nematodes in roots. However, there were significantly more swollen nematodes in the roots of all crops and cultivars for non-treated versus fumigated plots. At the end of the growing season (9 October), more vermiform nematodes were found in roots of eggplant and tobacco cultivar K326 than in tobacco cultivar NC567 or pepper. More vermiform nematodes were found in the roots of all other plants compared to pepper. There were no significant differences between eggplant, K326, or tomato in the number of swollen and total nematodes. However, more swollen and total nematodes were found in roots of eggplant, K326, and tomato, than in pepper or NC567. More swollen and total nematodes were found in the roots of NC567 than in pepper. Similar trends were found in numbers of pyriform and adult nematodes, where significantly more nematodes were found in roots of eggplant, tomato, and K326, than in pepper or NC567. Significantly more vermiform and total nematodes were found in plots not treated with a nematicide than in fumigated plots.

*Vegetable harvest data*-Fumigation increased ( $P \leq 0.05$ ) the number of eggplant fruit harvested at the 7 August, 2 September, and cumulative harvests (Table 4.3). Fruit weight for 7 and 17 August, 2 and 28 September, and cumulative harvests was also

increased by nematicide treatment. Fumigation increased the average weight per fruit for the 25 August, 28 September, and cumulative harvests.

Nematicide treatment did not increase the yield of pepper or tomato (Table 4.4 and 4.5). In fact, pepper weights were higher ( $P \leq 0.05$ ) from untreated than treated plots at the 2 September harvest date. A similar trend was noted in average weight per fruit on the 7 August harvest date.

*Tobacco yield data*-No significant cultivar-treatment interactions were observed in tobacco yield data. Fumigation did not increase yield or crop value (Table 4.6). However, the TCN-resistant cultivar NC567 produced a higher ( $P \leq 0.05$ ) yield and value than the TCN-susceptible cultivar K326. No significant differences were observed for price or grade index between cultivars or nematicidal treatments.

### **Discussion**

The results of this study confirm eggplant, pepper, and tomato as hosts of *Globodera tabacum solanacearum* under field conditions. Harrison and Miller (1969) reported eggplant and tomato to be more efficient hosts for *G. t. tabacum* than pepper. We noted a similar trend with *G. t. solanacearum*. Nematode numbers in the roots of eggplant and tomato were similar at the 9 October sampling date to those found on the TCN-susceptible flue-cured tobacco cultivar K326. Nematode numbers in roots of pepper were less than those in the TCN-resistant flue-cured tobacco cultivar NC567. Tomato and eggplant would be unacceptable as rotational crops for tobacco due to the significant TCN reproduction in the roots of these crops. However, due to low reproduction rates, pepper may be useful in tobacco rotations.



Eggplant was most severely injured by TCN, exhibiting a 64% yield reduction in nontreated plots compared to fumigated plots. In addition, TCN reproduction on eggplant was similar to that on the TCN-susceptible tobacco cultivar K326. Profitable production of eggplant in TCN-infested fields cannot be achieved without the use of a nematicide. Pepper were both tolerant to damage and TCN reproduction, exhibiting no yield loss and decreasing TCN populations in the soil. Apparently, pepper can be produced without the application of a nematicide due to low TCN reproduction rates and yield tolerance. Although, there were no significant differences in tomato yield between fumigated and non-treated plots, TCN reproduction on this crop was comparable to that found on eggplant and K326. Miller and Gray (1972) reported variable pathogenicity by TCN across varieties of tomato and pepper. The cultivars used in this study were selected due to their popularity in TCN threatened areas. Other cultivars of tomato and pepper should be tested for their host status to TCN.

This study was only conducted over a single year. A repeat test is necessary and is planned. Environmental conditions can influence the effects of TCN on tobacco (Johnson, 1998). Weather conditions during this study were wet at the beginning and very dry over the last two months. Apparent trends in our results may not be representative of nematode effects under different environmental conditions.

Fumigation seemed to have little effect on TCN population densities in the soil and within roots. It is possible that the method used to apply 1,3-dichloropropene (140 L/ha broadcast) was not adequate to achieve proper nematode control. Johnson *et al.* (1998) found that better TCN control is achieved if the fumigant is applied banded in-row

as opposed to broadcast over the entire field. Due to less than optimal TCN control, treatment differences in yield and nematode reproduction could have been harder to detect. Future repeat trials need to use in-row versus broadcast fumigation in order to more accurately investigate the effect of TCN on tested plants.

In conclusion, eggplant is highly susceptible to TCN infection, but tomato and pepper show some degree of tolerance to the nematode. However, other cultivars of these crops may respond differently to TCN infection. Reproduction of TCN on tomato and eggplant is similar to that of TCN-susceptible flue-cured tobacco, while reproduction on pepper seems to be less than or equal to that of a TCN-resistant flue-cured tobacco.

Table 4.1. Population densities of tobacco cyst nematodes in 1998 in soil either treated or untreated with a nematicide and planted to various solanaceous plant species.

Eggs/500 cc soil							
Crop	28-Apr (P <sub>i</sub> )	25-Jun (P <sub>2</sub> )	31-Jul (P <sub>3</sub> )	15-Oct (P <sub>f</sub> )	P <sub>2</sub> /P <sub>i</sub> Ratio	P <sub>3</sub> /P <sub>i</sub> Ratio	P <sub>f</sub> /P <sub>i</sub> Ratio
Eggplant	16,838 a	10,657 a	6,242 cd	11,541 b	0.67 a	0.40 bc	0.67 bc
K326	17,767 a	11,052 a	9,069 b	26,180 a	0.70 a	0.67 ab	1.73 a
NC567	14,290 a	8,807 a	6,755 bc	5,559 c	0.85 a	0.67 ab	0.58 c
Pepper	15,493 a	7,864 a	13,201 a	7,114 bc	0.51 a	0.85 a	0.47 c
Tomato	19,973 a	11,033 a	5,221 d	21,033 a	0.98 a	0.35 c	1.00 b

  

Eggs/500 cc soil							
Treatment	28-Apr (P <sub>i</sub> )	25-Jun (P <sub>2</sub> )	31-Jul (P <sub>3</sub> )	15-Oct (P <sub>f</sub> )	P <sub>2</sub> /P <sub>i</sub> Ratio	P <sub>3</sub> /P <sub>i</sub> Ratio	P <sub>f</sub> /P <sub>i</sub> Ratio
Fumigated	16,744 a	9,310 a	7,820 a	17,022 a	0.74 a	0.53 a	0.95 a
Untreated	17,000 a	10,455 a	8,374 a	11,548 a	0.74 a	0.64 a	0.83 a

Means shown in the table are from four replications. Mean separations were performed on log transformed data [ $\log_{10}(x+1)$ ] using the Waller-Duncan test (k-ratio=100) for the plant data and Tukey's test (alpha=0.05) for detection between treatments.

Table 4.2. Tobacco cyst nematodes within one gram of feeder root of various solanaceous hosts, with or without fumigation at Southern Piedmont Agricultural Research and Extension Center in Nottoway County, Virginia, in 1998.

Crop	Nematodes per gram of feeder root									
	7-Jul-98					9-Oct-98				
	Vermiform	Swollen	Pyriform	Adult	Total	Vermiform	Swollen	Pyriform	Adult	Total
Eggplant	2.8 a	4.1 a	6.7 a	15.3 a	28.8 a	201.8 a	32.1 a	59.0 a	89.0 a	381.9 a
K326	2.8 a	1.7 a	2.3 a	18.9 a	25.7 a	181.8 a	32.6 a	47.8 a	63.3 a	325.4 a
NC567	1.0 a	1.1 a	8.2 a	7.4 a	17.8 a	129.3 b	15.5 b	11.8 b	8.8 b	165.3 b
Pepper	1.8 a	1.0 a	7.1 a	5.9 a	15.7 a	51.8 c	7.5 c	9.9 b	8.6 b	77.8 c
Tomato	3.4 a	2.6 a	2.8 a	10.3 a	19.0 a	150.0 ab	30.1 a	54.9 a	67.5 a	302.5 a

Treatment	Nematodes per gram of feeder root									
	7-Jul-98					9-Oct-98				
	Vermiform	Swollen	Pyriform	Adult	Total	Vermiform	Swollen	Pyriform	Adult	Total
Fumigated	1.6 a	1.1 b	4.9 a	16.8 a	24.4 a	135.1 b	23.0 a	39.3 a	50.7 a	248.0 b
Untreated	3.1 a	3.1 a	5.9 a	6.3 a	18.3 a	150.7 a	24.2 a	34.1 a	44.2 a	253.2 a

Means shown in the table are from four replications. Mean separations were performed on log-transformed data [ $\log_{10}(x+1)$ ] using the Waller-Duncan test (k-ratio=100) for the plant data and Tukey's test (alpha=0.05) for detection between treatments.

Table 4.3. Eggplant yield data, from 1998, parasitized by tobacco cyst nematodes, with or without the presence of a nematicide, at Southern Piedmont Agricultural Research and Extension Center, in Nottoway County, Virginia.

Number of fruits per harvest per plot							
Treatment	7-Aug	17-Aug	25-Aug	2-Sep	11-Sep	28-Sep	Total
Fumigated	3.5 a	7.5 a	9.3 a	14.5 a	13.8 a	34.0 a	82.5 a
Untreated	0.0 b	3.0 a	6.0 a	4.0 b	6.5 a	17.0 a	37.0 b
p-value	0.0152*	0.0633	0.1789	0.0175*	0.0827	0.1285	0.0272*

Weight (kg) per harvest per plot							
Treatment	7-Aug	17-Aug	25-Aug	2-Sep	11-Sep	28-Sep	Total
Fumigated	1.3 a	4.4 a	4.6 a	6.0 a	5.3 a	13.2 a	34.8 a
Untreated	0.0 b	1.4 b	2.4 a	1.7 b	2.4 a	4.7 b	12.6 b
p-value	0.0144*	0.0350*	0.0744	0.0151*	0.0766	0.0072*	0.0053*

Average weight (g) per fruit for each harvest							
Treatment	7-Aug	17-Aug	25-Aug	2-Sep	11-Sep	28-Sep	Total
Fumigated	382.6	587.2 a	500.1 a	417.2 a	385.3 a	391.5 a	422.8 a
Untreated	N/A	360.5 a	387.1 b	391.9 a	265.5 a	298.6 b	355.6 b
p-value	N/A	0.1599	0.0492*	0.5715	0.2753	0.0428*	0.0247*

Values shown in tables are means of four replications. Comparisons were conducted within harvest day using t-testing ( $P \leq 0.05$ ) on log-transformed data [ $\log_{10}(x+1)$ ].

Table 4.4. Pepper yield data, from 1998, parasitized by tobacco cyst nematodes, with or without the presence of a nematicide, at Southern Piedmont Agricultural Research and Extension Center, in Nottoway County, Virginia.

Treatment	Number of fruits per harvest per plot					
	7-Aug	17-Aug	25-Aug	2-Sep	28-Sep	Total
Fumigated	26.5	86.8	34.0	5.0	71.3	223.5
Untreated	35.5	90.5	36.0	7.8	77.8	247.5
p-value	0.3673	0.6431	0.6973	0.0815	0.8104	0.5000

Treatment	Weight (kg) per harvest per plot					
	7-Aug	17-Aug	25-Aug	2-Sep	28-Sep	Total
Fumigated	3.6	12.7	4.3	0.5	5.3	26.4
Untreated	5.1	14.0	4.6	0.9	5.7	30.3
p-value	0.2960	0.3705	0.7147	0.0465*	0.8711	0.3948

Treatment	Average weight (g) per fruit for each harvest					
	7-Aug	17-Aug	25-Aug	2-Sep	28-Sep	Total
Fumigated	129.8	146.5	123.8	102.4	71.4	117.4
Untreated	145.2	155.1	126.1	113.4	72.8	122.8
p-value	0.0413*	0.2593	0.7972	0.4989	0.8605	0.3155

Values shown in tables are means of four replications. Comparisons were Conducted within harvest day using t-testing ( $P \leq 0.05$ ) on log-transformed data [ $\log_{10}(x+1)$ ].

Table 4.5. Tomato yield data, from 1998, parasitized by tobacco cyst nematodes, with or without the presence of a nematicide, at Southern Piedmont Agricultural Research and Extension Center, in Nottoway County, Virginia.

Treatment	Number of fruits per harvest per plot						Total
	7-Aug	17-Aug	25-Aug	2-Sep	11-Sep	28-Sep	
Fumigated	9.5	47.3	87.3	103.5	305.3	48.8	601.5
Untreated	4.0	29.5	48.5	112.0	404.0	45.5	643.5
p-value	0.2473	0.0982	0.2185	0.7256	0.3919	0.7012	0.7231

Treatment	Weight (kg) per harvest per plot						Total
	7-Aug	17-Aug	25-Aug	2-Sep	11-Sep	28-Sep	
Fumigated	1.2	7.4	14.5	13.9	35.6	4.7	77.3
Untreated	0.5	4.5	7.3	17.3	44.4	3.8	77.8
p-value	0.3064	0.0915	0.2371	0.1601	0.4198	0.3102	0.9622

Treatment	Average weight (g) per fruit for each harvest						Total
	7-Aug	17-Aug	25-Aug	2-Sep	11-Sep	28-Sep	
Fumigated	90.7	157.7	161.9	139.9	123.7	97.7	131.4
Untreated	129.4	151.0	148.7	163.7	110.7	81.9	122.5
p-value	0.3180	0.4422	0.3098	0.4740	0.4003	0.2327	0.5729

Values shown in tables are means of four replications. Comparisons were conducted within harvest day using t-testing ( $P \leq 0.05$ ) on log-transformed data [ $\log_{10}(x+1)$ ].

Table 4.6. Tobacco yield and quality data of two flue-cured tobacco cultivars, from 1998, parasitized by tobacco cyst nematodes, with or without the presence of a nematicide at Southern Piedmont Agricultural Research and Extension Center in Nottoway County, Virginia.

Cultivar	Price \$/lb.	Grade Index	Yield lbs/Acre	Value \$/Acre
NC567	1.68 a	51.1 a	2,810 a	4,732 a
K326	1.68 a	48.0 a	2,314 b	3,897 b

Treatment	Price \$/lb.	Grade Index	Yield lbs/Acre	Value \$/Acre
Fumigated	1.70 a	52.6 a	2,701 a	4,598 a
Untreated	1.66 a	46.5 a	2,424 a	4,031 a

Means within columns followed by the same letter are not significantly different according to the Tukey's test ( $P = 0.05$ ). Data are means of eight replications.



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