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Effect of Nematicides, Telone® II and Vorlex®, on Microflora and Nitrification in Tobacco Soil

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Fumigant nematicides are widely used in southwestern Ontario to control damage from the root lesion nematode, Pratylenchus penetrans (Cobb) Filip. and Stek. 1941, which causes brown-root rot of tobacco (Nicotiana tabacum L.). An estimated 70% of the flue-cured tobacco acreage in Ontario is fumigated annually. An investigation by Elliot and Mountain (1963) indicated that fall application of organochlorine fumigants 3 weeks after nitrogen fertilization of the rye, inhibited nitrification, thus causing deleterious effects on the quality of the tobacco crop. It was demonstrated previously by Tu (1973) that fumigant nematicides had a temperature-dependent effect on nitrification. Unless fumigant application was properly timed, it adversely affected the nitrogen cycle as it related to tobacco production. Marks et al. (1972) found lower nitrate (NO_3^--N) levels in a soil up to 8 weeks after fumigation with Telone (1,3-dichloropropene and related C_3 hydrocarbons), but the effect on the soil nitrification had disappeared by 11 weeks after application. Elliot et al. (1974) found that levels of NH_4^+-N in the soil tended to be higher and NO_3^--N lower with the fumigant DD (1,3-dichloropropene, 1,2-dichloropropane, and related C_3-hydrocarbons) than the control sample. Other fumigant and non-fumigant nematicides are effective for the control of nematodes in flue-cured tobacco in Ontario (Marks and Elliot 1975), but the effect of these chemicals on the soil microflora and nitrification of added $(NH_4)_2SO_4$ has not been studied in field plots of tobacco in Ontario. The objective of this study was to determine under field conditions, the effects of certain fumigants, Telone^R II and Vorlex^R, on microbial populations and nitrification in the loamy sand before the tobacco was planted.

MATERIALS AND METHODS

Experiments were conducted on Fox loamy sand with two treatments and a control in two consecutive years. Each replicate was 2, 3 and 4 times in a randomized block design. Telone^R II (92% 1,3-dichloropropene, 8% inert

materials) and Vorlex^{R} (80% 1,3-dichloropropene and related C_3 hydrocarbons, and 20% methyl isothiocyanate) were applied at 73 and 28 L/ha, respectively on May 4, 9, 14, and 19 in both experiments. The fumigants were injected into the soil 15-20 cm deep and ridged with an additional 15 cm of soil to give an effective injection depth of 30-35 cm. Each 0.005 ha plot consisted of two 24.4-m rows. The rows of the untreated control plots were ridged in the same manner as the fumigated rows.

Microbial populations and activities in nitrification were determined from 20 soil cores, 20 cm in depth, taken with 2-cm i.d. soil tube on May 10, 15 and 20 from each experiment in each year. The treated and untreated soils were incubated at 28°C in 236-mL milk bottles, which were closed with 0.038-mm thick polyethylene film for 3 weeks. Moisture was maintained at 60% of soil moisture holding capacity. Controls, with soil only, were included. Numbers of microorganisms were counted by a soil-dilution plate technique. Sodium albuminate agar (Waksman and Fred 1922) was used for bacteria and rose bengal-streptomycin agar (Martin 1950) for fungi.

Nitrification of ammonium-N from soil organic matter and of $(NH_4)_2SO_4$-N added at 200 μg/g was measured by the phenol disulphonic acid method (Harper 1924) for nitrates and diazotization method (A.P.H.A. 1955) with sulphanilic acid, α-naphthylamine hydrochloride and sodium acetate buffer for nitrites. A source of inorganic nitrogen, $(NH_4)_2SO_4$, was mixed thoroughly into the soil to determine nitrification. Results were expressed on the oven-dry basis of triplicate samples. Statistical analysis was performed as described by Elliot et al. (1972).

RESULTS AND DISCUSSION

Numbers of microorganisms, before and after incubation at all sampling dates, were higher in the 1st than in the 2nd experiment (Table 1). Incubation had no consistent effect on fungi, but bacterial counts declined with incubation in both experiments. These declines probably resulted from the fact that over a long incubation period, aeration was inadequate, the sources of available nutrients were depleted, and waste metabolites had accumulated (Eno 1960; Stotzky et al. 1962).

In both experiments, the effect of different fumigants on fungal populations was measured on May 10, which was 6- and 1-day after treatments. Plate counts of 1st experiment samples indicated that fungal numbers were lower than the control before and after incubation. No significant inhibition of fumigants on fungal population was observed in 2nd experiment. Inhibitory effects were observed before incubation in 1st experiment in samples

Table 1. Effects of time of application of nematicides on population of microorganisms in tobacco soils before and after incubation.*

Treatment	Sampling Date	Experiment 1 Fungi ($X10^3$/g)		Experiment 2 Fungi ($X10^3$/g)		Experiment 1 Bacteria ($X10^5$/g)		Experiment 2 Bacteria ($X10^5$/g)	
		Incubation Time (wk)							
		0	3	0	3	0	3	0	3
Control	May 10	68	56	58	42	102	83	86	70
Telone II (May 4)		30*	19*	75	50	142	29*	105	54
Vorlex (May 4)		23*	32*	75	74*	201*	64	116*	100*
Telone II (May 9)		26*	16*	58	33	161*	46*	70	49
Vorlex (May 9)		27*	31*	66	50	133	99	86	57
Control	May 15	67	36	75	33	103	73	49	32
Telone II (May 4)		62	50	116*	33	100	64	84*	27
Vorlex (May 4)		44	35	83	58*	137*	101	70	30
Telone II (May 9)		59	48	66	42	138*	51	62	30
Vorlex (May 9)		20*	25	58	33	165*	50	57	32
Telone II (May14)		28*	34	66	33	141	49	59	27
Vorlex (May14)		18*	39	100	33	104	59	78*	27
Control	May 20	20	27	83	33	80	43	46	24
Telone II (May 4)		37	38	91	33	93	56	51	24
Vorlex (May 4)		16	28	116	33	127*	46	68	30
Telone II (May 9)		28	32	191	42	109	48	41	27
Vorlex (May 9)		32	57	149	25	135*	48	68	41
Telone II (May14)		31	43	91	33	47	46	62	32
Vorlex (May14)		25	28	91	75*	115	42	59	32
Telone II (May19)		24	31	75	25	67	45	59	27
Vorlex (May19)		8	12	66	50	95	50	73*	32

* Values within each column for each sampling date are significantly different from control at P = 0.05.

of May 15, 6-day after treatments with Vorlex and 1-day after treatments with Telone II and Vorlex. However, after 3 weeks' incubation, fungal populations recovered to a level equal to that found in the control.

Significantly greater fungal numbers were observed with Telone II 11-day after treatment in 2nd experiment before incubation, while fungal numbers were greater with Vorlex after 3 weeks incubation. No inhibitory effect on fungal population was observed in all treatments before and after incubation in both experiments of May 20. An inhibitory effect was observed on the bacterial populations with Telone II from samples of May 10, 6-and 1-day after treatment in 1st experiment after incubation, while bacterial numbers in 2nd experiment were greater before and after incubation in 6-day after treatment with Vorlex than that in the control samples. In soil samples of both May 15 and 20, higher levels of bacteria were found in many samples before incubation.

Table 2. Effect of time of application of nematicides on soil nitrification μg(NO_2^-+NO_3^-)-N/g soil before and after incubation*

Treatment	Sampling Date	Experiment 1 Incubation Time(wk) 0	1 3	2 0	2 3
		Addition of N-source			
		0	0 200μg $(NH_4)_2SO_4$-N/g	0	0 200μg $(NH_4)_2SO_4$-N/g
Control	May 10	2.5	5.4 16.0	1.3	9.7 17.4
Telone II	(May 4)	1.9	7.2 35.5	1.4	13.1* 14.6
Vorlex	(May 4)	1.2*	6.7 58.2	1.5	8.8 14.6
Telone II	(May 9)	2.0	6.9 55.3	1.1	11.1 16.9
Vorlex	(May 9)	2.4	5.9 31.1	1.3	9.7 20.5
Control	May 15	3.9	7.3 19.9	1.1	7.3 21.8
Telone II	(May 4)	1.7*	7.2 12.3	1.1	10.3 13.8
Vorlex	(May 4)	1.1*	6.3 20.3	1.1	10.1 14.8
Telone II	(May 9)	1.2*	5.6 9.7	0.9	6.9 17.7
Vorlex	(May 9)	1.5*	6.5 13.4	1.0	8.6 21.3
Telone II	(May14)	3.3	10.1 18.5	1.0	11.1* 16.4
Vorlex	(May14)	3.4	8.8 40.8	1.0	10.3 17.4
Control	May 20	2.4	6.1 58.3	1.5	8.4 22.1
Telone II	(May 4)	1.3	10.3 34.0	1.5	11.1* 15.9
Vorlex	(May 4)	3.4	7.2 41.7	1.6	11.2* 17.4
Telone II	(May 9)	1.9	5.3 14.7	1.1*	11.0* 20.5
Vorlex	(May 9)	1.8	8.8 11.1	1.1*	8.5 19.2
Telone II	(May14)	2.1	4.0 19.2	1.0*	8.9 13.8
Vorlex	(May14)	3.0	4.6 13.0	0.9*	10.3 14.5
Telone II	(May19)	3.6	5.1 8.1	1.1*	8.9 15.1
Vorlex	(May19)	4.9	3.6 16.7	1.0*	9.0 18.2

* Values within each colum for each sampling date are significantly differenmt from control at P = 0.05.

The soil samples were collected before planting, and the samples were taken between the fertilizer bands. The content of NH_4^+-N for nitrification believed to be negligible in the non N-source added samples.

Without incubation, fumigants depressed nitrification in samples of May 10, 6-day after treatment with Vorlex before incubation in 1st experiment (Table 2). Inhibitory effect on nitrification was also observed in samples of May 15, 11- and 6-day after treatments with both fumigants in 1st experiment and of May 20, 11-, 6- and 1-day after treatment with both fumigants in 2nd experiment. No inhibition on nitrification of native organic nitrogen occurred in the incubated soils. The decreased effect of the fumigants on nitrification in some samples after 3 weeks incubation in both experiments would indicate either that the fumigants underwent transformation and detoxication in soil during that time or that the nitrifiers had adapted to the fumigants. Addition of $(NH_4)_2SO_4$ and/or incubation increased

nitrification markedly in both experiments. By contrast, the nitrification capability was weaker before incubation in some samples of May 15 in 1st experiment and of May 20 in 2nd experiment. It probably was due to the presence of weak populations of NH_4^+-N-oxidizers which required a longer incubation time to restore the activity of nitrification. Fumigants had no marked or consistent influence on nitrification.

Partial sterilization of the soil by fumigants makes soil organic matter more available to decomposition by microorganisms (Paul and Tu 1965; Tu and Miles 1976). Readjustment and adaptation of indigenous saprophytic soil microflora and multiplication of certain groups and species follow destruction of some competitors and antagonists. As a result of this, surviving microorganisms recover and rapidly replace the sensitive species, thus maintaining the metabolic integrity of the soil.

Although the fumigant-nematicides had some significant effects on soil microbial populations and activities important to soil fertility, recovery was generally rapid. Apparently the soil microorganisms can tolerate the nematicides currently in use, neverthless the effects of new compounds on soil microorganisms and their activities should be evaluated before these compounds receive wide spread use.

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