

Istituto di Nematologia Agraria - C.N.R., 70126 Bari, Italy

EFFECT OF CALCIUM CYANAMIDE AND OTHER AMMONIA FERTILIZERS ON *MELOYDOGINE INCOGNITA*

by

T. D'ADDABBO, A. FILOTICO and N. SASANELLI

Summary. The nematicidal effect of calcium cyanamide at different dosages was tested on tomato in comparison with urea and ammonium sulphate against *Meloidogyne incognita*. Calcium cyanamide caused phytotoxicity when applied after transplanting, whereas suppressed nematode population only at rates over 1,000 mg N/kg soil, when distributed before transplanting. Urea and ammonium sulphate were suppressive at all the application rates, although phytotoxic at the highest dosages. Fertilizer rates and final nematode population data were related significantly by the linear equation $y = a - bx$.

The effectiveness of inorganic fertilizers in controlling phytoparasitic nematodes in infested soils is well known (Rodríguez-Kábana, 1986). Urea and other fertilizers containing ammoniacal nitrogen or formulations releasing ammonia in the soil are the most suppressive, although their nematicidal action is consistent only at application rates much higher than those required for crop fertilization (Rodríguez-Kábana and King, 1980; Huebner *et al.*, 1983). The effect of ammonium sulphate on root-knot nematodes (*Meloidogyne* spp.) also produced only erratic or inconsistent results (Dos Santos *et al.*, 1981; Prot *et al.*, 1994; Khan and Khan, 1995).

Calcium cyanamide is an amide fertilizer releasing ammonium nitrogen by a slow process of hydrolysis in the soil, but also has a pesticidal, herbicidal and nematicidal activity (Cornforth, 1971). However, application at the rates used for crop fertilization did not give sufficient control of cyst (Šedivý, 1969; Müller, 1985) or root-knot nematodes (Lamberti *et al.*, 1975). Therefore an experiment was undertaken in the glasshouse to compare the effect of calcium cy-

namide, urea and ammonium sulphate at high application rates on the root-knot nematode *Meloidogyne incognita* (Kofoid *et* White) Chitw.

Materials and methods

Clay pots (12 cm diam, 700 g soil) were filled with sterilized sandy soil. Urea, ammonium sulphate and calcium cyanamide were applied on the surface of the soil at the rates of 250, 500, 750, 1,000 and 2,000 mg N/kg. Urea and ammonium sulphate were applied in two equal parts, at transplanting and 28 days after transplanting. Calcium cyanamide was applied either in total at 28 days before transplanting or in two equal parts, 28 days before transplanting and 28 days after transplanting. The pots were arranged on benches in a glasshouse at 25 ± 2 °C in a randomized block design with six replicates of each treatment; non fertilized pots were used as control. Blocks and pots within blocks were randomized every week to avoid position effects.

One tomato (*Lycopersicon esculentum* L.) cv. Rutgers seedling was transplanted in each pot and after one week inoculated with 8,000 eggs and juveniles (*Pi*) of an Italian population of *M. incognita*, host race 1, from sugarbeet (*Beta vulgaris* L.) at Castellaneta, province of Taranto, Apulia. Eggs were extracted from the sugarbeet roots by shaking for 3 minutes in a 1% sodium hypochlorite aqueous solution (Hussey and Barker, 1973).

Sixty days after transplanting, plants were uprooted and their height, fresh and dry top and fresh root weights were recorded. Root gall index was evaluated according to a 0-5 scale, where 0 = no galls, 1 = 1-2 galls, 2 = 3-10 galls, 3 = 11-30 galls, 4 = 31-100 galls and 5 = >100 galls (Taylor and Sasser, 1978). The final nematode population density (*Pf*) in each pot was determined by processing each tomato root in 1% sodium hypochlorite (Hussey and Barker, 1973) and extracting nematodes from 500 cm³ soil by a modification of Coolen's method (Coolen, 1979; Di Vito *et al.*, 1985). The reproductive rate of the nematode was calculated ($r = Pf/Pi$).

Data were statistically analyzed by analysis of variance followed by Fisher's least significant difference.

Results

Calcium cyanamide was phytotoxic at all rates and times of application. When applied after transplanting 50% of the tomato plants survived at the lowest dosage but there was 100% mortality at rates ≥ 500 mg N/kg soil. At the same rates ammonium sulphate also caused phytotoxicity, although replacement plants then grew normally. Urea was phytotoxic only at the highest application rate.

There was no significant effect of any of the treatment on plant height (Table I). Fresh and dry weights of plants in soils fertilized with calcium cyanamide were not significantly different

from the control, but always lower than the other treatments at $\leq 1,000$ mg N/kg soil. Urea at 500 and 750 mg N/kg soil and ammonium sulphate at $\leq 1,000$ mg N/kg soil significantly increased plant weight compared with the control and with 2,000 mg N/kg soil dosage. At the latter application rate there was a significant reduction of fresh root weight with all three fertilizers.

The number of eggs and juveniles on roots was significantly decreased by calcium cyanamide only at the highest rate applied before transplanting, and at 250 mg N/kg soil when applied in two parts (Table II). Ammonium sulphate and urea at rates > 250 mg N/kg soil caused a significant reduction of egg numbers in comparison with both the control and calcium cyanamide $< 2,000$ mg N/kg soil. Calcium cyanamide, at rates $\geq 1,000$ mg N/kg soil in one application or at 250 mg N/kg soil in two applications, significantly decreased the reproduction rate of *M. incognita* compared to the untreated control. Values of *r* were significantly lower than control with urea and ammonium sulphate at rates $\geq 1,000$ mg N/kg soil and ≥ 750 mg N/kg soil, respectively. The root gall index was significantly lower than the control with calcium cyanamide at the two highest rates of application, with 2,000 mg N/kg soil urea and with ammonium sulphate at rates ≥ 750 mg N/kg soil. Reduction in root galling caused by these treatments was also statistically significant compared with the lower rates of each fertilizer.

The relationship between the final population of *M. incognita* and the application rate of the three fertilizers is described by the linear equation $y = a - bx$ (1) (Fig. 1), in which *y* is the log₁₀ final nematode population/dm³ soil and *x* the fertilizer rate (mg N/kg soil). Data from the experiment fitted this equation well, as shown by the high correlation index and level of significance.

Based on eq. (1), any infinitesimal variation of nematode population caused by a corresponding infinitesimal variation of fertilizer rate

TABLE I - Effect of different dosages of calcium cyanamide, urea and ammonium sulphate on growth of tomato (*cv. Rutgers*) in soil infested by *Meloidogyne incognita*.

Fertilizer	Dosage (mg N/kg soil)	Distribution time	Plant height (cm)	Plant top weight (g)		Root fresh weight (g)
				fresh	dry	
Control			58.7	33.8	2.7	5.3
Calcium cyanamide	250	4 wks before transplanting	56.1	43.4	3.5	6.8
"	500	"	60.3	46.0	4.2	6.6
"	750	"	64.5	47.0	4.3	5.1
"	1,000	"	62.1	40.3	3.7	4.4
"	2,000	"	60.3	27.2	2.6	2.3
Calcium cyanamide	250	1/2 4 wks before + 1/2 4 wks after transplanting	49.5	22.4	2.3	4.7
"	500	"	-	-	-	-
"	750	"	-	-	-	-
"	1,000	"	-	-	-	-
"	2,000	"	-	-	-	-
Urea	250	1/2 at transplanting + 1/2 4 wks after transplanting	71.3	53.9	4.9	6.5
"	500	"	65.0	68.5	6.4	9.0
"	750	"	74.2	68.5	6.6	7.6
"	1,000	"	59.9	48.3	4.1	4.5
"	2,000	"	60.1	31.2	2.9	3.1
Ammonium sulphate	250	1/2 at transplanting + 1/2 4 wks after transplanting	67.8	55.8	5.5	9.3
"	500	"	72.9	71.3	6.5	8.5
"	750	"	68.3	56.8	5.4	5.2
"	1,000	"	69.3	57.6	5.3	4.7
"	2,000	"	51.2	17.8	1.5	1.1
L.S.D. 0.01			15.8	20.4	2.1	3.0

is proportional to the final population of *M. incognita*, i.e.:

$$dP/df = -kP \quad (2),$$

where P = final nematode population density, f = fertilizer rate and k = a proportionality coefficient. If finite variations are considered, it will be:

$$\int_{P_u}^{P_f} 1/P \times dP = \int_{P_u}^{P_f} -k \times df \quad (3),$$

in which P_u = final nematode population in unfertilized soil and P_f = final nematode population in fertilized soil. Then:

$$\log_n P_f - \log_n P_u = -fk \quad (4), \text{ or:}$$

$$\log_{10} P_f - \log_{10} P_u = -fk/2.30258 \quad (5).$$

If $2.30258/k = a$,

$$\log_{10} P_f - \log_{10} P_u = -f/a \quad (6) \text{ or,}$$

$$\log_{10} (P_f/P_u) = -f/a \quad (7).$$

Considering a 10-fold reduction of nematode population in fertilized soil, i.e.

$P_f \times 10 = P_u$, it will be:

$\log_{10} (P_f/P_u) = \log_{10} (1/10) = -f/a = -1$ (8) and therefore,

$$a = -f/\log_{10} (P_f/P_u) = -f/(\log_{10} P_f - \log_{10} P_u) \quad (9),$$

and graphically, from Fig. 1:

$$a = \cos \varphi / \sin \varphi \quad (10),$$

where φ is the angle between abscissa and regression line. Then:

$$a = -\cotg \varphi = -1/\tg \varphi \quad (11).$$

Therefore a , i.e. the fertilizer dosage at which the nematode reproductive capacity is re-

duced by 10-fold, corresponds to the inverse of the angular coefficient of the regression lines in Fig. 1. It could be defined as "nematode reproduction decimal reduction rate".

Discussion

The high phytotoxicity of calcium cyanamide was confirmed when applied at high dosages (Dekker and Duke, 1995). A single dose applied before transplanting avoided this problem, but was suppressive to *M. incognita* only at a very high rate.

TABLE II - Effect of different dosages of calcium cyanamide, urea and ammonium sulphate on reproduction of *M. incognita* on tomato (cv. Rutgers).

Fertilizer	Dosage (mg N/kg soil)	Distribution time	No. eggs/g roots (x 1,000)	Pf (eggs and juveniles/cm ³ soil)	r (Pf/Pi)	G.I.
Control			43.0	322	26.9	5.0
Calcium cyanamide	250	4 wks before transplanting	35.9	356	29.6	5.0
"	500	"	32.2	314	26.2	4.7
"	750	"	38.1	286	23.8	4.0
"	1,000	"	37.4	168	14.0	3.7
"	2,000	"	14.8	57	4.8	3.0
Calcium cyanamide	250	1/2 4 wks before + 1/2 4 wks after transplanting	15.2	115	9.6	5.0
"	500	"	—	—	—	—
"	750	"	—	—	—	—
"	1,000	"	—	—	—	—
"	2,000	"	—	—	—	—
Urea	250	1/2 at transplanting + 1/2 4 wks after transplanting	35.9	320	26.7	4.7
"	500	"	21.8	279	23.2	4.3
"	750	"	18.2	211	17.6	4.2
"	1,000	"	21.8	141	11.7	4.0
"	2,000	"	10.8	49	4.1	2.2
Ammonium sulphate	250	1/2 at transplanting + 1/2 4 wks after transplanting	18.2	237	19.7	4.8
"	500	"	11.9	156	13.0	4.2
"	750	"	17.4	136	11.4	3.7
"	1,000	"	16.3	100	8.3	3.3
"	2,000	"	9.3	12	1.0	2.2
L.S.D. 0.01			16.8	129	10.8	1.0

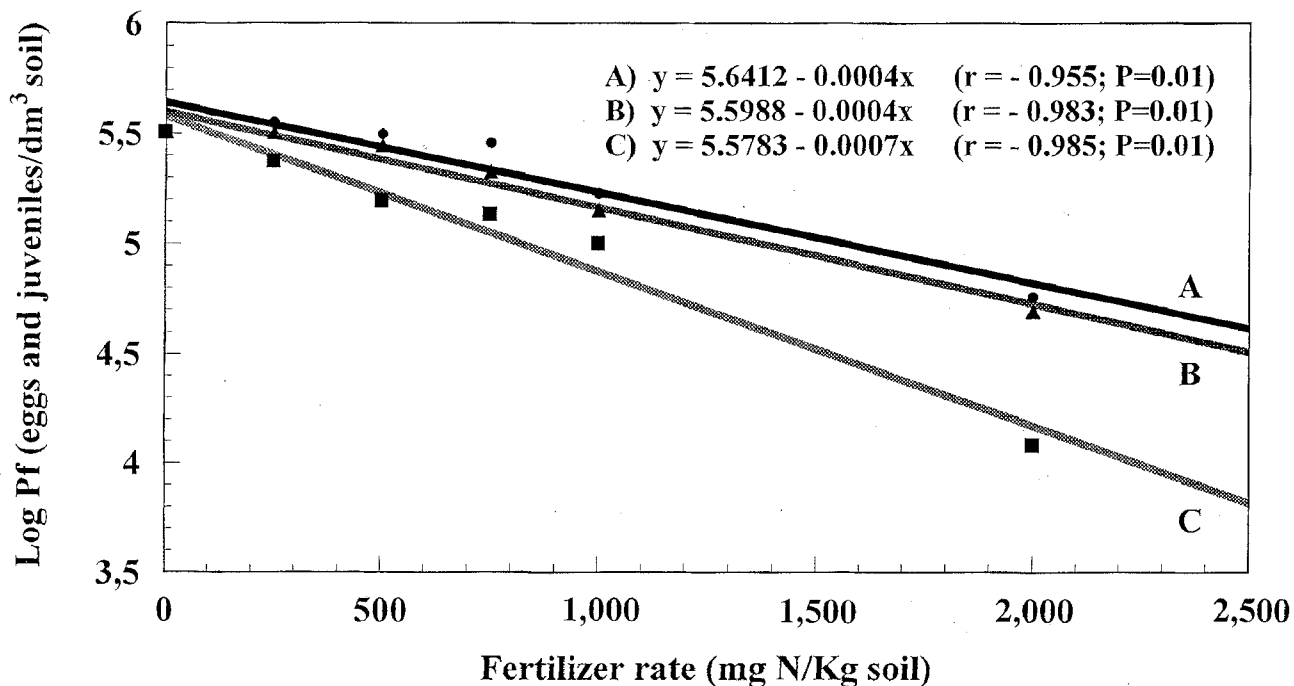


Fig. 1 - Relationship between \log_{10} final population of *Meloidogyne incognita* on tomato cv. Rutgers and application rates of different ammonia fertilizers: A) calcium cyanamide; B) urea; C) ammonium sulphate.

The suppressive action of urea was evident at rates ≥ 500 mg N/kg soil: in a previous glass-house experiment urea applied at rates of 200-1,000 mg N/kg soil reduced population of *M. incognita* but was phytotoxic to soybean (Huebner *et al.*, 1983), whereas at rates ≥ 400 mg N/kg soil it reduced the number of juveniles and root galling of *M. arenaria* on squash but was phytotoxic (Rodríguez-Kábana, 1980).

Populations of *M. incognita* and galling on tomato roots were reduced by ammonium sulphate at the lowest rate of application. This was probably due to its quick degradation in the soil and then to a high concentration of ammonia which could also explain its phytotoxicity to tomato seedlings at all the doses. In previous experiments ammonium sulphate, at increasing dosages, decreased reproduction rates and number of galls of *M. incognita* on papaya (Khan and Khan, 1995) and of *M. exigua* on

coffee (Dos Santos *et al.*, 1981), whereas at application rates much lower than those used in this experiment there was no suppressive effect on *M. graminicola* on rice (Prot *et al.*, 1994).

Fertilizer rate and final nematode population were related by a linear regression equation which gave a good fit of data from the experiment: a 10-fold reduction of final nematode population in the soil could theoretically be obtained, based on values of parameter *a*, applying calciumcyanamide and urea at 2,500 mg N/kg soil and ammonium sulphate at 1,428 mg N/kg soil.

The accumulation of nitrates and ammonia in the soil and, for ammonium sulphate, the high salt index per unit of nitrogen (Rader *et al.*, 1943), provides an explanation of the phytotoxic effect caused by high dosages of ammonia fertilizers. Addition of amendments with a

large availability of carbon can stimulate the microbial activity and consequently the utilization of nitrogen excess.

Literature cited

- COOLEN W. A., 1979. Methods for the extraction of *Meloidogyne* spp. and other nematodes from roots and soil. In: Root-knot nematodes (*Meloidogyne* spp.). Systematics, Biology and Control, pp. 317-329. (Eds. F. Lamberti and C. E. Taylor). London, UK: Academic Press.
- CORNFORTH I. S., 1971. Calcium cyanamide in agriculture. *Soils Fertil.*, 34: 463-470.
- DEKKER J. and DUKE S. O., 1995. Herbicide - resistant field crops. *Adv. Agron.*, 54: 96.
- DI VITO M., GRECO N. and CARELLA A., 1985. Population densities of *Meloidogyne incognita* and yield of *Capiscum annuum*. *J. Nematol.*, 17: 45-49.
- DOS SANTOS J. M., FERRAZ S. and DE OLIVEIRA L. M., 1981. Effect of nitrogen fertilizers on gall formation in the roots of coffee infested with *Meloidogyne exigua* and on the hatching of larvae. *Fitopatol. Brasil.*, 6: 457-463.
- HUEBNER R. A., RODRÍGUEZ-KÁBANA R. and PATTERSON R. M., 1983. Hemicellulosic waste and urea for control of plant parasitic nematodes: Effect on soil enzyme activities. *Nematropica*, 13: 37-54.
- HUSSEY R. S. and BARKER K. R., 1973. A comparison of methods of collecting inocula of *Meloidogyne* spp. including a new technique. *Plant Dis. Repr.*, 57: 1025-1028.
- KHAN A. T. and KHAN S. T., 1995. Effect of NPK on disease complex of papaya caused by *Meloidogyne incognita* and *Fusarium solani*. *Pak. J. Nematol.*, 13: 29-34.
- LAMBERTI F., COIRO M. I. and CALLIERIS F., 1975. Esperienze con la calciocianamide nella lotta contro i nematodi galligeni su tabacco levantino in provincia di Lecce. Proc. Giornate Fitopatologiche, Torino, Italy, 12-14 Nov. 1975: 259-263.
- MÜLLER J., 1985. The potential of calcium cyanamide for the control of sugarbeet nematode, *Heterodera schachtii*. *Mitteil. der Biologis. Bundesan. Land Forstwir.*, 226: 141-150.
- PROT J. C., VILLANUEVA L. M. and GERGON E. B., 1994. The potential of increased nitrogen supply to mitigate growth and yield reductions of upland rice cultivar UPL Ri-5 caused by *Meloidogyne graminicola*. *Fundam. Appl. Nematol.*, 17: 445-454.
- RADER L. F., WHITE L. M. and WHITTAKER C. W., 1943. The salt index - a measure of the effect of fertilizers on the concentration in the soil solution. *Soil Science*, 55: 201-218.
- RODRÍGUEZ-KÁBANA R., 1986. Organic and inorganic nitrogen amendments to soil as nematode suppressants. *J. Nematol.*, 18: 129-135.
- RODRÍGUEZ-KÁBANA R. and KING P. S., 1980. Use of mixtures of urea and blackstrap molasses for control of root-knot nematodes in soil. *Nematropica*, 10: 38-44.
- ŠEDIVÝ J., 1969. Control of *Heterodera rostochiensis* Woll. by using resistant varieties and nematicides. *Ochrana Rostlin*, 5 (4): 237-244.
- TAYLOR A. L. and SASSER J. N., 1978. Biology, Identification and Control of Root-knot Nematodes (*Meloidogyne* spp.). North Carolina State University Graphic, Raleigh, N.C. (USA), pp. 111.