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SUPPRESSION OF *MELOIDOGYNE INCOGNITA* BY COMBINATIONS OF OLIVE POMACE OR WHEAT STRAW WITH UREA

by

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Summary. The suppressive action of soil amendment with exhausted olive pomace and wheat straw, alone or in combination, and their possible synergism with urea were tested on *Meloidogyne incognita* on tomato in three glasshouse experiments. Olive pomace reduced the nematode reproduction at all the rates tested, but it was phytotoxic at high dosages. Its nematicidal effect was improved by the addition of urea at rates ≥ 500 mg N/kg soil. Wheat straw suppressed the population of *M. incognita* at all the dosages tested. Combining with urea improved the nematicidal effect of the straw and limited the phytotoxic effects of the highest rates of the fertilizer. Mixing olive pomace with straw improved the nematicidal effect of olive pomace but did not prevent phytotoxicity at the highest rate tested.

Soil amendment with crop residues or agro-industrial waste materials has been shown to have a suppressive action on phytoparasitic nematodes (D'Addabbo, 1995). However, their use is economic only if the materials are available at low or zero cost. Olive and wheat are two of the most economically important crops in southern Italy, and generally in the Mediterranean basin, and these generate large amounts of olive pomace and wheat straw, respectively.

Exhausted pomace is the waste product remaining after the extraction of residual oil from fresh olive pomace by chemical solvents. This material has been shown to suppress the root-knot nematode *Meloidogne incognita* on tomato without incurring much phytotoxicity (D'Addabbo and Sasanelli, 1996).

Straw from wheat or other cereal crops, used alone or in combination with practices like soil solarization, has also been found to suppress various phytoparasitic nematode species (Esmenjaud *et al.*, 1990; Greco *et al.*, 1992).

The addition of urea or other nitrogen sources to fresh olive pomace increases the nematicidal effect of the soil amendment, enabling it to be used at lower dosages and thus reducing phytotoxicity (Rodríguez-Kábana *et al.*, 1992; 1995). Urea is a nitrogen fertilizer that counteracts the depletion of nitrogen caused by the incorporation into the soil of materials with a large C:N ratio such as wheat straw; it also has a directly suppressive effect on nematodes (D'Addabbo *et al.*, 1996; Rodríguez-Kábana, 1986).

Three pot experiments were undertaken in a glasshouse to ascertain the combination of exhausted olive pomace, wheat straw and urea that could be used most effectively and with limited phytotoxicity for the control of *M. incognita*.

Materials and methods

Experiments were undertaken using pots of sterilized sand to which were added exhausted olive pomace, wheat straw and urea at the rates and combinations shown in Tables I, II, III. The 12 cm diam clay pots contained 500 cm^3 soil, six replicates of each treatment and were arranged in a randomized block design in a glasshouse at 25±2 °C.

Four weeks after the soil amendment treatments, a single tomato (*Lycopersicon esculentum* Mill.) seedling cv. Rutgers was transplanted into each pot. After a further week each pot was inoculated with 10,000 eggs and juveniles of an Italian population of *Meloidogyne incognita* (Kofoid *et* White) Chitw. host race 1 (Taylor and Sasser, 1978), corresponding to an initial population density (*Pi*) of 20 eggs and juveniles/cm^3 soil.

Tomato plants were uprooted 60 days after transplanting and their height, fresh and dry top and root weights were recorded. Final population density (*Pf*) and reproduction rate (r = *Pf/Pi*) of *M. incognita* were determined by extracting eggs and juveniles from each tomato root by the Hussey and Barker (1973) method and processing the soil in each pot by the modified Coolen (1979) method (Di Vito *et al.*, 1985). The root gall index (G.I.) was evaluated on a 0-5 scale (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).

Data were statistically analyzed by the analysis of the variance (ANOVA) followed by Fisher's L.S.D. test at P = 0.01.

TABLE I - *The effect of soil amendment with different combinations of exhausted olive pomace and urea on the reproduction of* Meloidogyne incognita *and growth of tomato.*

Amendment rate		Plant growth parameters				Nematode reproduction parameters			
Olive pomace (g/kg soil)	Urea (mg N/kg soil)	Plant height (cm)	Fresh top weight (g)	Dry top weight (g)	Fresh root weight (g)	Eggs and J2/g roots (x 1000)	*Pf* (eggs and J2/g soil)	r (*Pf/Pi*)	Root gall index (G. I.)
–	–	37.2	33.2	4.7	14.2	32.9	1,015	50.7	4.8
–	250	39.8	39.1	5.9	14.1	21.5	582	29.1	4.3
–	500	39.0	37.9	5.2	12.5	1.4	30	1.5	3.7
–	1,000	44.8	32.8	4.3	6.7	1.0	15	0.7	3.8
–	2,000	*	*	*	*	*	*	*	*
20	–	40.2	31.0	4.5	13.5	22.1	538	26.9	4.6
20	250	37.8	36.2	5.0	17.4	13.3	460	23.0	4.8
20	500	34.7	28.7	3.9	10.1	9.9	205	10.2	4.2
20	1,000	39.0	31.3	4.5	9.5	0.8	34	1.7	3.3
20	2,000	*	*	*	*	*	*	*	*
40	–	41.0	37.2	5.1	14.7	13.9	413	20.7	4.2
40	250	39.8	27.5	3.7	11.5	17.7	434	21.7	4.5
40	500	38.0	27.9	3.1	9.2	7.3	186	9.3	3.7
40	1,000	*	*	*	*	*	*	*	*
40	2,000	*	*	*	*	*	*	*	*
L.S.D. (P = 0.05)		8.5	11.6	1.7	3.8	7.0	193	9.6	0.5
(P = 0.01)		11.4	15.4	2.3	5.1	9.4	257	12.8	0.7

* Severe phytotoxicity/no data.

TABLE II - *The effect of soil amendment with different combinations of wheat straw and urea on the reproduction of* M. incognita *and growth of tomato.*

Amendment rate		Plant growth parameters				Nematode reproduction parameters			
Wheat straw (g/kg soil)	Urea (mg N/kg soil)	Plant height (cm)	Fresh top weight (g)	Dry top weight (g)	Fresh root weight (g)	Eggs and J2/g roots (x 1000)	*Pf* (eggs and J2/g soil)	*r* (*Pf/Pi*)	Root gall index (G. I.)
–	–	41.7	37.2	5.3	15.9	36.8	1,137	56.8	4.8
2.5	–	64.6	71.3	9.6	20.6	16.6	614	30.7	4.7
5.0	–	56.0	67.5	8.5	14.8	15.9	596	29.8	4.4
10.0	–	63.0	72.1	7.8	18.2	14.0	494	24.7	4.7
20.0	–	61.6	72.1	7.7	12.8	13.5	385	19.3	4.2
40.0	–	66.9	64.2	7.8	26.6	1.7	135	6.7	3.6
2.5	400	52.4	60.2	6.6	20.0	1.3	251	12.5	4.0
5.0	400	55.8	70.8	7.5	16.2	4.2	140	6.9	3.7
10.0	400	64.2	70.7	8.8	15.2	7.0	245	12.3	2.7
20.0	400	67.2	68.4	8.3	19.0	2.9	125	6.3	3.0
40.0	400	60.1	64.7	7.4	22.9	3.6	181	9.1	3.8
–	250	44.6	42.7	7.7	15.8	24.1	652	32.6	4.3
–	500	43.7	42.4	5.7	14.0	1.6	34	1.7	3.7
–	1,000	50.2	36.7	4.8	7.5	1.1	17	0.8	3.8
–	2,000	*	*	*	*	*	*	*	*
40.0	250	52.4	54.9	6.8	16.2	4.4	138	6.9	3.7
40.0	500	54.6	64.3	8.9	24.1	3.5	157	7.8	4.0
40.0	1,000	53.4	45.9	7.8	21.5	0.2	34	1.6	3.8
40.0	2,000	59.1	54.2	8.6	23.1	0.5	49	2.5	3.6
L.S.D. ($P = 0.05$)		11.4	17.5	2.8	6.9	7.1	150	8.4	0.6
($P = 0.01$)		15.6	23.2	3.7	7.8	9.4	212	10.1	0.7

* Severe phytotoxicity/no data.

Results

Olive pomace alone at rates ≥ 20 g/kg soil suppressed nematode reproduction in the tomato roots, but did not prevent the formation of galls (Tables I, III). The highest dosage caused a significant reduction of tomato growth, both alone or combined with urea or straw. The combination of 20 g pomace/kg soil with urea reduced significantly the multiplication of *M. incognita* compared with pomace alone. The mixtures of olive pomace with wheat straw were more suppressive than pomace alone at all the tested dosages.

Wheat straw always reduced the root population and the reproduction rate of *M. incognita*, but did not limit the number of galls on the tomato roots (Tables II, III). The suppressive effect was higher at 40 g/kg soil than at the lower dosages. Nematode reproduction was significantly less in the soil amended with 10 and 20 g straw/kg soil than at the corresponding rates of olive pomace, but no statistical difference was found among the two materials at the highest incorporation rate. In a second experiment the presence of the wheat straw also significantly affected the plant growth parameters com-

TABLE III - *The effect of soil amendement with different combinations of exhausted olive pomace and urea on the reproduction of* M. incognita *and growth of tomato.*

Amendment rate		Plant growth parameters				Nematode reproduction parameters			
Olive pomace (g/kg soil)	Wheat straw (g/kg soil)	Plant height (cm)	Fresh top weight (g)	Dry top weight (g)	Fresh root weight (g)	Eggs and J2/g roots (x 1000)	*Pf* (eggs and J2/g soil)	*r* (*Pf/Pi*)	Root gall index (G. I.)
–	–	60.9	86.8	9.6	30.9	59.6	3,743	187	5.0
–	10	62.8	82.4	9.8	34.2	48.2	3,041	152	4.7
–	20	59.2	81.6	10.0	36.1	35.4	2,563	128	4.5
–	40	59.3	58.9	7.3	23.6	29.9	1,440	72	4.8
10	–	72.3	85.2	11.2	41.2	21.2	1,676	84	4.2
20	–	77.6	101.0	12.4	43.3	12.8	1,201	60	3.5
40	–	75.7	102.7	10.8	45.3	8.6	854	43	3.3
10	10	72.1	83.7	10.0	36.4	22.5	1,620	81	4.0
10	20	84.0	85.1	10.1	34.2	19.2	1,251	63	3.7
10	40	75.2	61.9	8.0	27.7	11.2	669	33	2.9
20	10	78.8	92.1	11.3	39.1	15.5	1,239	62	3.0
20	20	80.9	79.5	9.9	40.2	5.7	449	22	3.8
20	40	72.6	49.0	7.3	21.1	4.7	223	11	3.9
40	10	88.5	72.2	9.1	29.5	6.1	437	22	3.6
40	20	76.0	57.7	7.5	28.7	11.8	731	36	3.5
40	40	69.7	33.4	4.0	18.1	5.4	278	14	3.7
L.S.D. (P = 0.05)		13.8	19.7	2.9	11.7	9.8	715	36	0.5
(P = 0.01)		19.4	29.2	3.9	15.9	13.0	952	48	0.7

pared with the unamended soil, but this difference was not confirmed by a third trial. The combination with 400 mg N/kg urea improved the suppressivity of the straw alone, except at the rate of 40 g/kg soil, but did not affect plant growth. The addition of 40 g/kg olive pomace to 10 and 20 g/kg rates of straw caused a significant reduction of the nematode multiplication compared with straw alone.

Urea strongly suppressed multiplication and galling of *M. incognita* on the tomato roots, but was phytotoxic at the rate of 2,000 mg N/kg soil (Tables I, II). Phytotoxicity was avoided when it was combined with 40 g straw/kg soil. The relationship between rate of urea, alone or combined with olive pomace, and final population density of *M. incognita* was tested using different interpolation formulae (linear, power, exponential and logarithmic). The best fit with the data available was obtained by the exponential equation ($y = \mathrm{b}\ e^{mx}$) (Fig. 1).

Discussion

A previous trial (D'Addabbo and Sasanelli, 1996) demonstrated the suppressive effect of exhausted olive pomace on *M. incognita* juveniles and this was confirmed in our experiments. However, phytotoxicity generally occurred only at high amendment dosages, although in the first experiment no phytotoxic effect was found at the rate of 40 g/kg soil. Lower doses of olive pomace combined with urea avoided phytotoxicity without reducing the suppressive action on *M. incognita*. It seems likely that other nitrogen sources, like ammonia, guanidine or chicken litter, could be used in combination with exhausted olive pomace, as in other glasshouse and microplots experiments they were not phytotoxic, although suppressive to *Meloidogyne* spp. on tomato when combined with fresh olive pomace (Rodríguez-Kábana *et al.*, 1995). However, a synergistic action of exhausted pomace with urea could not be hypothesised, as evident also by the interpolation lines in Fig. 1.

Soil amendment with wheat straw produced a strong suppression of *M. incognita* populations at low rates of incorporation: Esmenjaud *et al.* (1990) found that populations of *Pratylenchus* spp. were significantly suppressed in a wheat monoculture when straw was ploughed into the soil compared to when it was removed. In other field experiments, the incorporation of 10 t/ha wheat straw to a soil infested by the carrot cyst nematode, *Heterodera carotae*, followed by an eight week solarization period, resulted in a significant yield increase, whereas the addition of 5 t/ha straw followed by the same solarization period prevented the infestation of *Ditylenchus dipsaci* on onion and improved the yield of marketable bulbs (Greco *et al.*, 1992). At the same dosage straw was much more suppressive than olive pomace, and thus lower dosages could be used to achieve the same nematicidal effect. The effect of straw was improved only by the addition of the largest rate of pomace, but the phytotoxicity makes this mixture practically useless. Lower dosages were needed to obtain the same suppressive effect when the straw was combined with urea at 400 mg N/kg soil.

Our experiments confirmed that straw does not enhance the nematicidal properties of urea,

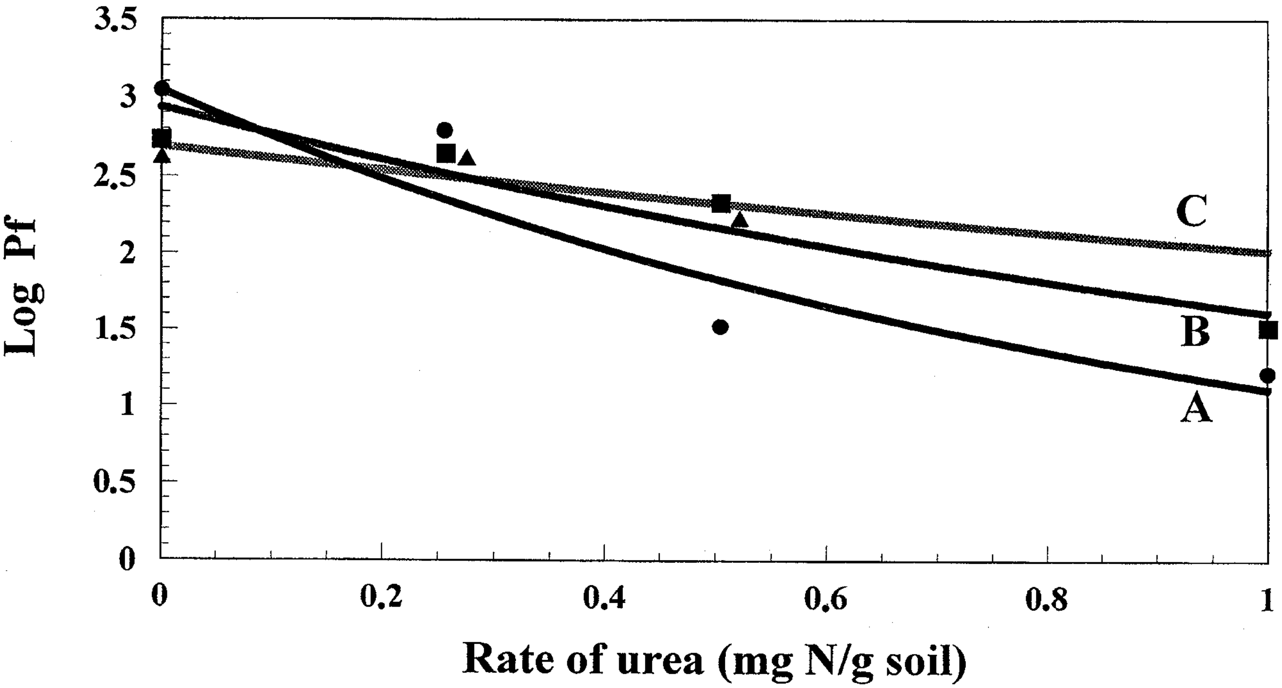

Fig. 1 - Relationship between rate of urea, alone or combined with exhausted olive pomace, and final population density of *Meloidogyne incognita* on tomato (cv. Rutgers) plants: ● A = urea alone ($y = 3.043\ e^{-1.018X}$; r = -0.939); ■ B = urea + 20 g exhausted olive pomace/kg soil ($y = 2.935\ e^{-0.605X}$; r = -0.966); ▲ C = urea + 40 g exhausted olive pomace/kg soil ($y = 2.687\ e^{-0.29X}$; r = -0.841).

but at rates > 1,000 mg N/kg soil it avoided its phytotoxic effects, by supplementing the available carbon needed for the microbial utilization of ammoniacal nitrogen deriving from the decomposition of urea.

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