Management of *Meloidogyne hapla* on Lettuce in Organic Soil with Sudangrass as a Cover Crop

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ABSTRACT

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Host suitability for *Meloidogyne hapla* of six cover crops was tested in the greenhouse. Sudangrass cv. Trudan 8 and rye (mixture of cultivars) were nonhosts; oat cv. Porter was a poor host; and phacelia cv. Angelia, oilseed radish cv. Renova, and yellow mustard cv. Martigena were maintenance hosts. When incorporated as a green manure before planting of lettuce cv. Montello, sudangrass was the most effective of the cover crops in reducing egg production of *M. hapla*. Soil amendment with all parts of sudangrass resulted in lower reproduction of *M. hapla* on lettuce than soil amendment with only roots of sudangrass. Soil incorporation of 2-month-old (or younger) tissues of sudangrass was more effective in reducing nematode reproduction on subsequent lettuce plants than incorporation of 3-month-old tissues. Sudangrass was grown as a cover crop after lettuce for three growing seasons in field microplots and incorporated as a green manure before the first fall frost. Weight of lettuce heads was significantly higher and reproduction of *M. hapla* was significantly lower in sudangrass-amended microplots compared with those left fallow between lettuce crops, but results varied with year and nematode infestation level.

Additional keywords: *Avena sativa*, *Brassica hirta*, *Lactuca sativa*, northern root-knot nematode, *Phacelia tanacetifolia*, *Raphanus sativus* var. *oleifera*, *Secale cereale*, *Sorghum sudanense*

The northern root-knot nematode (*Meloidogyne hapla* Chitwood) causes considerable economic losses in the commercial production of lettuce (*Lactuca sativa* L.) on organic (muck) soils in New York. Root systems that are severely infected with this nematode are unable to provide sufficient water and nutrients for normal plant growth, resulting in the production of small and often unmarketable lettuce heads. Growers rely on soil fumigation with nematicides for control of the nematode, because resistant lettuce varieties are not available and current crop rotation practiced on organic soils is of limited value. However, high costs and inconsistent results of nematicide application, together with reduced availability of nematicides as a result of increasing concern for the environment and for public health, have augmented interest in alternative methods for the management of plant-parasitic nematodes (17).

Rotation with crops that are poor or nonhosts for the northern root-knot nematode is not feasible on the high-valued muck soils in New York, because they cannot provide an income comparable to that generated by lettuce production. Most of the crops rotated with lettuce, such as onion, carrot, celery, beans, and potatoes, are also good hosts for *M. hapla* (7,8). The potential use of cover crops, grown in periods between the main crop production (21), for suppression of *M. hapla* may be a better alternative to chemical control of the nematode. It is known that the addition of organic matter to the soil improves soil drainage, reduces wind and water erosion, and decreases leaching of nutrients (12,21). The desirable properties of a cover crop grown for the management of a plant-parasitic nematode are its poor host status for the target nematode and additional suppression of the nematode population after its incorporation into the soil as a green manure.

Oilseed radish (*Raphanus sativus* L. var. *oleifera*), yellow mustard (*Brassica hirta* Moench), and phacelia (*Phacelia tanacetifolia* Benth.) have been used as rotation crops and as green manures in Germany for the control of the sugar beet-cyst nematode, *Heterodera schachtii* Schmidt (10); however, the host status of these crops to *M. hapla* has not been reported. Sudangrass (*Sorghum sudanense* (Piper) Stapf), sorghum-sudangrass hybrids (*Sorghum bicolor* (L.) Moench × *S. sudanense*)

and sudangrass hybrids (*S. sudanense* × *S. sudanense*) have been used on a limited scale as cover crops on organic soils in New York. Mojtahedi et al. (14) reported that all the cultivars of sudangrass and sorghum-sudangrass hybrids tested suppressed populations of *Meloidogyne chitwoodi* Golden et al. when the plants were used as green manures. In addition, they reported that none of the tested cultivars were found to be hosts for *M. hapla*. Oat (*Avena sativa* L.) and rye (*Secale cereale* L.) have been used traditionally on organic soils to improve soil structure. They have been reported as nonhosts (7,8) or poor hosts (8) for *M. hapla*. This study was undertaken to assess the host status of selected cover crops (phacelia, yellow mustard, oilseed radish, oat, rye, and sudangrass) to *M. hapla* and to evaluate their usefulness as soil amendments in controlling the nematode and in improving lettuce production in organic soil.

MATERIALS AND METHODS

All tests were performed with lettuce cv. Montello, a cultivar of iceberg lettuce. The cover crops included in the experiments were: phacelia cv. Angelia (Hydrophyllaceae), oilseed radish cv. Renova, yellow mustard cv. Martigena (Brassicaceae), oat cv. Porter, rye (mixture of cultivars), and sudangrass hybrid cv. Trudan 8 (Poaceae). Sudangrass was supplied by Northrup King Company and was pretreated with pirimiphos-methyl (Nugro) and captan (Captane). The other seeds were dusted with a mixture of metalaxyl (Apron, 12.5% a.i.) and captan (Captane, 48.9% a.i.) before seeding to minimize loss of seedlings by damping-off diseases caused by *Pythium* spp. and other plant-pathogenic fungi. Seeds of oilseed radish, yellow mustard, and phacelia were provided by L. Stivers (Area Extension Specialist, P.O. Box 150, Albion, NY 14411). Seeds of oat and rye were obtained from the Seed Testing Laboratory at the Agricultural Experiment Station, Cornell University, Geneva, New York.

The organic (muck) soil used in all tests was obtained from a commercial lettuce field in Oswego County, New York. Soil used in the greenhouse experiments was passed through a sieve with 1.5-cm-diameter openings to remove big pieces of undecayed plant material before it was added to the pots. Soil was autoclaved (121°C for 30 min) for the host suitability tests only, in order to eliminate plant-parasitic nematodes.

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sitic nematodes and other soilborne pathogens and pests that would affect plant growth. Untreated soil was used in the other tests because microorganisms were necessary for the decomposition of the green manures.

M. hapla was maintained on tomato (*Lycopersicon esculentum* Mill.) cv. Rutgers in the greenhouse. Eggs were extracted from infected tomato roots with sodium hypochlorite (11). The egg suspension was pipetted on top of the soil, immediately followed by light watering of the pots. All tests infested in this way received eight *M. hapla* eggs per cm³ of soil, the estimated damage threshold density of *M. hapla* to lettuce under greenhouse conditions (25). The soils in the field microplot tests and in the green manure tests were infested with chopped roots containing egg masses of *M. hapla*. The amount of roots added to the soil was based on the number of eggs extracted from five samples (5 g each) with sodium hypochlorite (11). In the green manure test, roots were thoroughly mixed with the soil by hand before the pots were filled. To infest the field microplots, roots were incorporated into the upper 15 cm of soil in each tile with a trowel. Plants were watered daily, fertilized weekly with 30 ml of a solution of complete fertilizer (N-P-K, 20-20-20), and sprayed for the control of foliar insects as needed. Additional light was provided when day length was less than 10 h. The temperature in the greenhouse varied between 24 and 27°C.

At harvest time, plants were weighed and roots were carefully washed and evaluated for root gallings caused by *M. hapla*. Root gallings were rated on a scale from 1 (no gallings) to 9 (≥80% of the root system galled). Ratings of 2 to 8 indicated that 1 to 3, 4 to 10, 11 to 25, 26 to 35, 36 to 55, 56 to 65, and 66 to 80% of the root system was galled, respectively. Eggs of *M. hapla* were extracted from the root systems with sodium hypochlorite (11), but roots were shaken with a wrist-action shaker for 15 min instead of 4 min. The volume of the egg suspension was adjusted, and the eggs were counted at ×40 magnification.

Host suitability tests. The host status of phacelia, oilseed radish, yellow mustard, oat, rye, and sudangrass to *M. hapla* was tested in the greenhouse. Lettuce was included as a control. Seeds of phacelia and lettuce were germinated on moist filter papers in petri dishes. Three-day-old seedlings were planted in pots filled with 500 cm³ of autoclaved organic soil. Seeds of oilseed radish, yellow mustard, sudangrass, oat, and rye were direct-seeded in identical pots. Ten plants were grown in each pot, except lettuce, for which only two seedlings were planted per pot. Plants were kept in a growth chamber at 22°C with a 12-h photoperiod (light intensity = 235 μE·s⁻¹·m⁻²). After 10 days, each pot was infested with 4,000 eggs of *M. hapla*

(initial population, P_i) and was removed to the greenhouse the next day. Pots were arranged in a completely randomized design with six replications, and plants were maintained for 7 weeks. The number of eggs of *M. hapla* extracted from roots in each pot was considered as the final egg population (P_f). The reproduction factor (R), calculated as $P_f \div P_i$, was used as a measure for the host suitability of the plant to *M. hapla*. The host status was described according to the four host categories defined by Ferris et al. (5) for *M. chitwoodi*, as follows: $R > 10$, excellent host; $10 > R > 1$, good host; R close to 1, maintenance host; and $R < 1$, poor host or nonhost. The host suitability of oilseed radish, sudangrass, and oat were tested in two additional greenhouse experiments (tests 2 and 3) as part of the investigation dealing with their use as green manures. The methods used in these experiments were similar to the one described above, except that the soil was not autoclaved.

Incorporation of cover crops into soil as green manures. The influence of incorporation of the six cover crops into the soil on the population of *M. hapla* and on the growth of lettuce was investigated in the greenhouse. Organic soil was mixed with chopped tomato roots containing mature egg masses of *M. hapla* (2 g of roots (16,000 eggs)/liter of soil) and placed in 1-liter clay pots. Three days later, pots were planted with 2, 30, 10, 10, 20, 20, and 20 seeds of lettuce, phacelia, oilseed radish, yellow mustard, oat, rye, and sudangrass, respectively. Phacelia was planted at the high rate of 30 seeds per pot because seed germination and seedling survival were low in previous tests. Soil in pots left unplanted was included as a control treatment. Ten pots of each treatment were arranged in a randomized complete block design in the greenhouse. Nine weeks later, stems and leaves of the cover crops were cut off at the soil line, weighed, and chopped into pieces approximately 4 cm long. Pots were emptied, roots were broken up by hand, and all the plant parts were mixed with the soil. The amended soil was then used to refill the same pots that the plants were grown in. Lettuce leaves were also cut off but were not incorporated into the soil in order to simulate normal commercial practice. Soil of the unplanted treatment was also mixed and put in the same pots. A saucer was placed under each pot to prevent loss of leachates and nematodes, and to prevent contamination between pots. Pots were left on the greenhouse bench for 19 days to allow for partial decomposition of the plant material. Two lettuce seeds were then planted in every pot, and lettuce plants were grown for 8 weeks. At harvest, lettuce plants were weighed, root gallings were evaluated, and eggs were extracted from the two root systems in each pot. The numbers of eggs of *M. hapla* extracted per root system were

transformed to $\log(x + 1)$ values before analysis of variance was performed. Root gallings ratings and weights of lettuce were also evaluated with analysis of variance (ANOVA). Means of all the treatments were compared with the mean of the unplanted control treatment with the least significant difference test (LSD).

Incorporation of the whole plant versus roots alone. Oilseed radish, oat, and sudangrass were selected for further study of their potential to reduce populations of *M. hapla* and increase yields of lettuce when used as green manures. The experiment was conducted in the greenhouse in 1993 and was repeated in 1994. Twenty seeds of either oilseed radish, oat, or sudangrass were seeded in 1-liter pots filled with organic soil. Thirty-two pots were seeded with each cover crop and 16 pots were left unplanted. Half of the pots of each treatment were infested with *M. hapla* (8,000 eggs per pot) 1 day after seeding. Soil in all pots was kept moist, and plants were maintained for 9 and 8 weeks in the 1993 and 1994 tests, respectively. Stems and leaves of the cover crops were then cut off at the soil line, weighed, and chopped up into pieces approximately 4 cm long. The soil of half of the pots of each treatment planted with cover crops was mixed with the chopped-up plant parts obtained from these pots, whereas the soil of the other half of the pots was mixed with only the roots that were already present in the pots. The total amount of plant tissues or roots of each cover crop was redistributed over all eight replicate pots before incorporation to obtain equal amounts of soil amendment in each replication. Soil of the pots with the unplanted treatments was also mixed and returned to the same pots. Pots were arranged in randomized complete blocks, and a saucer was placed under each pot. They were left on the greenhouse bench for 3.5 and 5.5 weeks in 1993 and 1994, respectively, to allow for decomposition of the cover crops. After this period, two lettuce seedlings, pregerminated (5 days) on wet filter paper in petri dishes, were planted in each of the pots. Lettuce was harvested 8 weeks after planting. Plants were weighed, and roots were evaluated for root gallings and reproduction of *M. hapla*. In the second test, eggs were not extracted from roots grown in uninfested soils. Data were analyzed as a factorial experiment with added controls, by ANOVA. The two added controls were the unplanted treatment with infested and uninfested soils. The numbers of eggs of *M. hapla* extracted per root system were transformed to $\log(x + 1)$ values to standardize the variance before statistical analysis. The treatment sum of squares from the ANOVA was partitioned into orthogonal contrasts for comparisons of groups of treatments.

Effect of age of sudangrass. The influence of the age of sudangrass tissues at the

time of their incorporation into the soil on the population of *M. hapla* and on lettuce growth was evaluated in two greenhouse tests in 1994. For the first test, sudangrass was planted in pots or trays in the greenhouse at 2-week intervals to obtain plants that were 2, 4, 6, or 8 weeks old. For the second test, sudangrass was planted in the field at monthly intervals so that 1-, 2-, and 3-month-old plants were available. Stems and leaves of sudangrass were cut in pieces approximately 4 cm long and incorporated into organic soil at a rate of 20 g/liter of soil. This amount corresponds to 40 t/ha if the sudangrass is incorporated 20 cm deep. Clay pots (15 cm diameter) were filled with soil amended with sudangrass tissues of different ages. One day after the incorporation of sudangrass, soils were infested with *M. hapla* (eight eggs per cm³ of soil). Two control treatments, consisting of unamended soil with and without eggs of *M. hapla*, were included in each test. A saucer was placed under each pot, and all pots were incubated in the greenhouse for 8 (test 1) or 6 weeks (test 2) to allow for decomposition of sudangrass and breakdown of phytotoxic components released by the green manures. Soil of each treatment was thoroughly mixed before it was put in 10-cm-diameter clay pots. There were 10 and 6 pots of each treatment in the first and second test, respectively, except for the treatment with 2-week-old sudangrass, which was replicated only five times in the first test. Pots were arranged in blocks, and two lettuce seeds were planted per pot. Plants were maintained for 8.5 and 10 weeks in the first and second test, respectively. Analysis of variance was performed on weight of lettuce, root-gall rating, and egg production per root system. Data obtained from the uninfested controls were not included for the last two parameters. The numbers of eggs of *M. hapla* per root system in the second test were transformed to log($x + 1$) values to stabilize the variance. The least significant difference (LSD) test was used to compare means.

Field microplot tests with sudangrass. Sudangrass was planted as a cover crop in field microplots at the Research Farm of the New York State Agriculture Experiment Station in Geneva, for 3 consecutive years. The microplots consisted of unglazed clay drainage tiles (25 cm diameter and 30 cm height) inserted into the field soil with the top of the tiles about 5 cm above the soil surface. The spacing of the tiles was 1 m within rows and 1.5 m across rows. Mineral soil of the test field was used to fill the lower halves of the tiles, and the upper halves were filled with organic soil that was either left uninfested or infested with a low or a high density of *M. hapla*. Chopped roots of tomato (test 1) or lettuce (tests 2 and 3) containing egg masses of *M. hapla* were used to infest the tiles. In August 1992 (test 1), microplots were infested with six eggs per cm³ of soil

(low infestation level) or 24 eggs per cm³ of soil (high infestation level). The low and high infestation rates in August 1993 (test 2) and 1994 (test 3) were two and four eggs of *M. hapla*/cm³ of soil, respectively. In addition, nematode-free lettuce roots were added to microplot soils with zero and low nematode infestation levels of tests 2 and 3, so that the total amount of added roots was the same in every tile. Half of the microplots of each infestation level were left unplanted (fallow). The other half was planted to sudangrass (30 seeds per microplot) 1 day after the nematode infestation. The experimental design was a 2 × 3 factorial experiment (with or without sudangrass, three infestation levels). The microplots were arranged in a randomized complete block design with each treatment replicated 13 times.

Sudangrass was grown for 53, 45, and 68 days in test 1, test 2, and test 3, respectively. At the end of this growing period, the number of sudangrass plants, the total weight of the aboveground plant parts, and the average height of the plants were recorded for every microplot. The leaves and stems of each microplot were then chopped

up into 5- to 10-cm-long pieces, and the total amount of plant tissues of each treatment was redistributed over the replicate microplots of the same treatment. Plant tissues and roots were incorporated into the organic soil (15 cm deep) and left to decompose during winter. Lettuce cv. Montello was planted in the microplots at a rate of eight seeds per microplot the following spring in tests 1 and 2. Seedlings were thinned gradually so that two plants per tile were left after 3 weeks. In test 3, 2-week-old lettuce seedlings grown in sterile soil in the greenhouse were transplanted into the microplots. Each microplot was fertilized with 5 g of a complete fertilizer (N-P-K, 13-13-13) 2 weeks after planting. The plots were watered and weeded as needed, and plants were sprayed twice with acephate (Orthene 75 S, 0.6 g a.i./m²) to control leafhoppers. Lettuce was harvested when heads were fully formed in the uninfested treatments (9 weeks after planting in tests 1 and 2, and 6.5 weeks after planting in test 3). Roots of each lettuce plant were dug up carefully, washed, and evaluated for root galling and production of eggs of *M. hapla*. ANOVA techniques were used to

Table 1. Host suitability to *Meloidogyne hapla* of selected cover crops grown in organic soil under greenhouse conditions

Cover crop ^w	Root gall rating ± SE ^x	Reproduction ± SE ^y	Host status ^z
Phacelia	2.2 ± 0.4	1.0 ± 0.5	Maintenance host
Oilseed radish	2.2 ± 0.2	1.4 ± 0.5	Maintenance host
Yellow mustard	3.0 ± 0.9	1.5 ± 0.5	Maintenance host
Oat	1.1 ± 0.1	0.3 ± 0.1	Poor host
Rye	1.0 ± 0	0 ± 0	Nonhost
Sudangrass	1.0 ± 0	0 ± 0	Nonhost
Lettuce	6.3 ± 1.0	25.8 ± 9.0	Excellent host

^w Values for oilseed radish, yellow mustard, and sudangrass are the means of three tests with six, eight, and eight replicate pots, respectively. Values for the other crops are the means of six replicate pots.

^x Root galling was evaluated on a scale from 1 (no galls) to 9 (>80% of roots galled).

^y Each pot (500 cm³) was infested with 4,000 eggs of *M. hapla* (Pi). After 7 weeks, eggs were extracted (Pf) and reproduction was calculated as Pf ÷ Pi.

^z Host categories: R > 10, excellent; 10 > R > 1, good; R ± 1, maintenance; R < 1, poor to nonhost.

Table 2. Effect of soil incorporation of selected cover crops on *Meloidogyne hapla* egg production and damage to lettuce in greenhouse tests

Soil amendment ^w		Parameters of <i>M. hapla</i> and lettuce ^x		
Crop	Fresh weight per pot (g) ± SE	Egg production per root ^y	Root gall rating ^z	Weight of lettuce (g)
Phacelia	8.0 ± 1.3	5,267 *	7.2 *	6.5 ns
Oilseed radish	23.2 ± 2.2	1,999 *	4.0 *	4.1 ns
Yellow mustard	16.2 ± 0.7	2,511 *	6.5 *	5.7 ns
Oat	22.8 ± 1.0	3,123 *	5.0 *	5.0 ns
Rye	11.9 ± 0.6	3,867 *	7.7 *	4.4 ns
Sudangrass	11.3 ± 0.4	710 ns	2.6 ns	3.4 *
Lettuce	—	3,217 *	6.7 *	4.2 ns
None (unplanted)	—	680	2.6	5.2 ns
MSE		0.06	1.42	2.93

^w Cover crops were grown for 9 weeks in pots with *M. hapla*-infested soil (16 eggs per cm³ of soil) and incorporated. Lettuce was planted 19 days after incorporation of the cover crops and harvested 8 weeks later.

^x Values are means of 10 replicate pots, except for phacelia and yellow mustard, which were replicated eight and nine times, respectively. Means marked with * differ significantly ($P < 0.01$) from the mean for the unplanted treatment, according to the least significant difference (LSD) test.

^y Means of egg production were transformed to log($x + 1$) values before analysis. The observed values are presented.

^z Root galling was evaluated on a scale from 1 (no galls) to 9 (>80% of roots galled).

examine the effects of soil cover and nematode infestation level on the weight of lettuce and the number of eggs of *M. hapla* produced per root system. A *t* test was used to compare the effects of the two soil covers (sudangrass and fallow) at each nematode infestation level.

RESULTS

Host suitability tests. Lettuce was an excellent host for *M. hapla*, with a reproduction factor (R) of 25.8 (Table 1). None of the cover crops tested were as good a host as lettuce, but the brassicas (oilseed radish and yellow mustard) and phacelia

were able to maintain the nematode population (R close to 1). Oat was a poor host for *M. hapla*, as indicated by the limited egg production on its roots in trials 2 and 3 ($R < 1$). Rye and sudangrass did not support any reproduction of the northern root-knot nematode ($R = 0$) and were, therefore, considered nonhosts. The degree of root galling was closely related to the host status of the plant to *M. hapla* (Table 1). Galls incited by *M. hapla* on the roots of phacelia, oilseed radish, and mustard were difficult to see, as they were much smaller than those on roots of lettuce.

Fig. 1. Egg production of *Meloidogyne hapla* on lettuce roots and weight of lettuce planted in soil amended with all tissues or roots only of oilseed radish (R), oat (O), sudangrass (S), or in soil left unplanted (U), in 1994. Soil was infested with *M. hapla* (+) prior to planting of the cover crops or was left uninfested (-).

Table 3. Orthogonal comparisons of the egg production of *Meloidogyne hapla* (Mh) on lettuce roots and weight of lettuce grown under greenhouse conditions, in uninfested or *M. hapla*-infested soil that was left unplanted or amended with either all plant tissues or only roots of oilseed radish, oat, or sudangrass^x

Orthogonal contrast	Egg production of <i>M. hapla</i> ^y (mean square)		Weight of lettuce (mean square)	
	1993	1994	1993	1994
Cover vs. unplanted	16.68 *** ^z	5.06 ***	4,092.2 ***	319.3 ns
Oilseed radish vs. (oat + sudangrass)	20.07 ***	7.45 ***	1,514.8 ***	610.3 *
Oat vs. sudangrass	3.69 *	3.15 ***	364.3 *	486.8 +
Oilseed radish +Mh vs. -Mh	74.59 ***	-	429.2 **	8,375.4 ***
all tissues vs. roots / +Mh	0.20 ns	0.30 +	473.1 **	193.2 ns
all tissues vs. roots / -Mh	1.78 ns	-	380.3 **	18.3 ns
Oat +Mh vs. -Mh	118.86 ***	-	0.6 ns	33.0 ns
all tissues vs. roots / +Mh	0.02 ns	0 ns	20.5 ns	18.1 ns
all tissues vs. roots / -Mh	2.33 +	-	5.3 ns	26.8 ns
Sudangrass +Mh vs. -Mh	107.01 ***	-	0.7 ns	1.4 ns
all tissues vs. roots / +Mh	1.21 ns	0.31 +	69.7 ns	651.5 *
all tissues vs. roots / -Mh	0 ns	-	829.4 ***	754.9 *
Unplanted: +Mh vs. -Mh	8.87 ***	-	154.4 ns	440 +

^x Analysis of variance (ANOVA) was for a $2 \times 3 \times 2$ factorial experiment with added controls. There were three treatment factors: type of soil infestation (with [+Mh] or without *M. hapla* [-Mh]), type of cover crop (oilseed radish, oat, sudangrass), and type of amendment (incorporation of all plant tissues or only roots). The added controls were unplanted treatments in either infested or uninfested soil. Treatments were arranged in a randomized complete block design with eight replications.

^y Egg production data were transformed to $\log(x + 1)$ values before analysis. Egg production of *M. hapla* in the uninfested soil was not determined in 1994.

^z Statistical significance at *** is $P \leq 0.001$, ** is $P \leq 0.01$, * is $P \leq 0.05$, + is $P \leq 0.10$. ns = no significant difference.

Incorporation of cover crops into soil.

The biomass of each cover crop added per liter of soil varied between 8.0 g (phacelia) and 23.2 g (oilseed radish) (Table 2). At the time of incorporation, oilseed radish and mustard were at the flowering stage, whereas oat was at the physiological maturity stage. Only the soil amendment with sudangrass resulted in a significant reduction in the population of *M. hapla* (measured by the number of eggs produced per root system of lettuce), which was equal to that in the unplanted treatment (Table 2). This reduction in nematode population was also reflected in the low root-gall rating given to roots of lettuce grown after the incorporation of sudangrass, and was comparable to that of the unplanted treatment. In contrast, root galling was moderate to high on roots of lettuce grown in the other soil cover treatments (Table 2). However, the weight of lettuce grown in soil amended with sudangrass was lower than the weight of lettuce grown in the soils of the other treatments, suggesting a phytotoxic effect (Table 2).

Incorporation of the whole plant versus only roots. Plants of oilseed radish, oat, and sudangrass were between 20 and 30 cm tall when they were incorporated into the soil. At this time, oilseed radish and oat were at the flowering stage, with some seed production in evidence in the 1993 test. These two cover crops were already at the flowering stage about 1 month after planting in the test of 1994. In 1993, the amount of aboveground plant tissues that was mixed with soil in each pot was 66.1 g, 27.5 g, and 43.1 g for oilseed radish, oat, and sudangrass, respectively; whereas the weight of the incorporated plant parts was 31.4 g, 15.4 g, and 37.6 g per pot for the respective cover crops in 1994.

The highest number of eggs of *M. hapla* was produced on roots of lettuce grown in the soil amended with oilseed radish, followed by oat, sudangrass, and unplanted soil (Fig. 1). Amending the soil with all plant tissues of sudangrass prior to planting of lettuce resulted in lower reproduction of *M. hapla* compared with nematode reproduction in soil amended with only the roots of sudangrass, although the difference was significant only in the 1994 test (Fig. 1, Table 3). Incorporating all plant parts of oat did not affect nematode reproduction on the subsequent lettuce crop in the nematode-infested soil, whereas the effect of soil amendments with oilseed radish tissues was opposite in both years and therefore inconclusive (Fig. 1, Table 3). In 1993, amending all plant parts of oilseed radish resulted in lower numbers of eggs per root system of lettuce compared with amending only roots of oilseed radish; whereas in 1994, amending all plant parts resulted in higher numbers of nematode eggs on the subsequent lettuce crop. Eggs of *M. hapla* were also extracted from roots

of lettuce grown in uninfested soil in the 1993 test. Few eggs were found per root system of lettuce grown after incorporation of oilseed radish (around 1,000) and oat (290), indicating that the untreated field soil was initially infested with a very low population of *M. hapla*. Root-gall rating of lettuce roots grown after the different soil amendments was correlated with the patterns of production of nematode eggs and was lowest in the sudangrass-amended soils (data not shown).

Amending the soil with all plant tissues of oilseed radish, in both nematode-infested and uninfested soils, increased the weight of lettuce significantly in 1993 but not in 1994. Incorporating all tissues of oat did not affect lettuce weight (Fig. 1, Table 3). However, the weight of lettuce grown in soils amended with the leaves and stems of sudangrass was less than the weight of lettuce grown in soils amended with only the roots of sudangrass, regardless of nematode infestation (Fig. 1, Table 3).

Effect of age of sudangrass. Infestation of unamended soil with *M. hapla* resulted in 26% reduction in the weight of lettuce (Table 4). In addition, plants infected with *M. hapla* exhibited severe root galling and a high egg production of *M. hapla* per root system (Table 4). Amending *M. hapla*-infested soil with 1- or 2-month-old tissues of field-grown sudangrass significantly increased lettuce weight, which was as high as that obtained in the uninfested soil. Root galling and reproduction of *M. hapla* were significantly reduced by the incorporation of 1- or 2-month-old tissues of sudangrass, but not by the 3-month-old tissues (Table 4). Actually, lettuce grown in soil amended with 3-month-old sudangrass tissues exhibited the highest root galling, supported the greatest egg production, and had the lowest weight. In contrast, egg production of *M. hapla* on roots of lettuce grown in soil amended with 1- and 2-month-old sudangrass tissues was reduced by 86 and 95%, respectively, compared with egg production in the unamended soil.

Likewise, egg production of *M. hapla* on roots of lettuce was significantly reduced by soil amendment with the greenhouse-grown sudangrass tissues that were 0.5, 1.0, 1.5, and 2 months old (Table 5). The incorporation of 1- and 1.5-month-old tissues of sudangrass resulted in the greatest (60 and 74%, respectively) reductions in egg production on lettuce roots. Root-gall ratings were low in all the treatments in this test, which was also evident in the limited differences in weight of lettuce.

Field microplot tests with sudangrass. The average weight of the incorporated sudangrass per microplot was higher in test 3 (179 g) than in tests 1 (64 g) and 2 (32 g). In all three tests, lettuce yield was affected by the incorporation of sudangrass and by the infestation level of *M. hapla*. There was an interaction between these two factors in the third test only (data not

shown). In test 1, lettuce yields were 31% higher ($P \leq 0.05$) in the sudangrass-amended soils than in the fallow treatment at the low infestation level (Fig. 2). In 1994, significantly higher lettuce yields were obtained in sudangrass-amended microplots compared with fallow microplots at both low and high infestation levels of *M. hapla* (20 and 34% increase in yield, respectively) (Fig. 2). In the 1995 test, lettuce yield was 24% higher ($P \leq 0.05$) in sudangrass-amended plots compared with plots left fallow at the high nematode infestation level (Fig. 2).

The numbers of eggs of *M. hapla* produced on lettuce roots was not affected by the sudangrass amendment in the first test (Fig. 2). However, in the following year, fewer eggs were produced on the roots of lettuce grown in microplots where sudangrass had been incorporated into the soil, compared with microplots that were left fallow, at all infestation levels (Fig. 2). In test 3, soil amendment with sudangrass resulted again in lower egg production on lettuce roots, but the difference was not significant for lettuce grown in soil with the low nematode infestation level (Fig. 2). Root-gall ratings of lettuce roots in the

three tests followed the trends observed for lettuce weight and reproduction of *M. hapla* (data not shown).

DISCUSSION

Results of the host suitability tests confirmed that lettuce cv. Montello is a good host for *M. hapla*, as indicated by the high reproductive factor (R) of 26. Results obtained for sudangrass, oat, and rye suggested that these crops are appropriate cover crops for potential use in a management program aimed at reducing the population of *M. hapla*. Reproduction of the nematode on these crops was nonexistent or very low, which is in agreement with other reports in the literature (8,10,14). Phacelia, oilseed radish, and yellow mustard were identified as maintenance hosts (R close to 1) for *M. hapla*. Yellow mustard was previously reported to be as good a host for *M. hapla* as lettuce (23), but it was found later that this was due to contamination with *M. incognita*.

Sudangrass was the most effective cover crop in decreasing the number of *M. hapla* eggs on the roots of a subsequent lettuce crop. Incorporation of stems and leaves of sudangrass into the soil resulted in a

Table 4. Effect of the age of field-grown sudangrass on the population of *Meloidogyne hapla* and its damage to lettuce grown in organic soil

Soil treatment		Parameters of <i>M. hapla</i> and lettuce ^w		
<i>M. hapla</i> ^x	Age of sudangrass (mo) ^y	Egg production per root	Root-gall rating ^z	Weight of lettuce (g)
–	0	0	1.0	41.4 a
+	0	13,820 b	6.3 b	30.6 b
+	1	1,913 c	2.3 c	44.9 a
+	2	731 c	2.1 c	45.1 a
+	3	75,900 a	9.0 a	31.7 b
MSE		0.08	1.26	52.60

^w Values are means of eight replicate pots. Means in each column followed by a common letter are not significantly different ($P < 0.05$) according to the least significant difference (LSD) test. The treatment without infestation of *M. hapla* was not included in the analysis of egg production and root-gall rating. Egg production data were transformed to $\log(x + 1)$ values before analysis, but original means are presented.

^x Soil was infested with eight eggs of *M. hapla* per cm³ 1 day after the addition of sudangrass.

^y Leaves and stems were incorporated into soil that was planted to lettuce 6 weeks later.

^z Root galling was evaluated on a scale from 1 (no galls) to 9 (>80% of roots galled).

Table 5. Effect of the age of greenhouse-grown sudangrass on the population of *Meloidogyne hapla* and its damage to lettuce grown in organic soil

Soil treatment		Parameters of <i>M. hapla</i> and lettuce ^w		
<i>M. hapla</i> ^x	Age of sudangrass ^y (mo)	Egg production per root	Root-gall rating ^z	Weight of lettuce (g)
–	0	0	1.0	44.0 bc
+	0	20,010 a	2.7 a	42.1 bc
+	0.5	12,220 b	2.2 b	47.4 ab
+	1	7,970 bc	2.6 a	49.8 a
+	1.5	5,120 c	2.1 b	42.7 bc
+	2	11,650 b	2.2 b	41.0 c
MSE		18,720	0.27	18.82

^a Values are means of 10 replicate pots, except for the treatment with 2-week-old sudangrass, which was replicated five times. Means in each column followed by a common letter are not significantly different ($P < 0.05$) according to the least significant difference (LSD) test. The treatment without infestation of *M. hapla* was not included in the analysis of egg production and root-gall rating.

^b Soil was infested with eight eggs of *M. hapla* per cm³ 1 day after addition of sudangrass.

^c Leaves and stems were incorporated into soil that was planted to lettuce 8 weeks later.

^d Root galling was evaluated on a scale from 1 (no galls) to 9 (>80% of roots galled).

greater reduction of the nematode population than amendment of the soil with the sudangrass roots only. This cannot be explained solely by the fact that the plant is a nonhost for *M. hapla*, as the other poor and nonhosts tested (oat and rye) were not capable of reducing the nematode population. An additional factor responsible for the decreased nematode population might be the release of toxic compounds by decomposing plant parts of sudangrass. These toxins may have killed the eggs and juveniles of *M. hapla* that survived in the soil since the initial infestation. All plant parts of sorghum and sudangrass, except for the seeds, contain a cyanoglycoside called dhurrin (3). Dhurrin yields hydrogen cyanide when hydrolyzed, a compound that has been suggested to be nematotoxic. Leaves of sudangrass were reported to be effective in reducing populations of *M. chitwoodi* when used as a green manure (15).

The greenhouse tests showed that the incorporation of 3-month-old sudangrass into organic soil was not as effective in reducing the population of *M. hapla* as was incorporation of younger plant tissues. The amount of hydrogen cyanide has been reported to decrease with maturity and with plant height. In addition, stems contain less dhurrin than leaves (9). The 3-month-old sudangrass plants were about 2 m high and had already produced seeds. Most of the 20 g of plant tissue added per liter of soil consisted of stem parts, as these were heavier than leaves. Another reason for the high reproduction of *M. hapla* on lettuce in the soil amended with the 3-month-old sudangrass could be the slow decomposition of the thick plant parts. Slow decomposing tissues possibly released lower quantities of dhurrin into the soil. Mojtahedi et al. (14,16) showed that homogenized tissues of rapeseed or sudangrass were superior in the control of *M. chitwoodi* to chopped

tissues. They hypothesized that this was due to the slower release of toxic compounds from chopped plant parts compared with homogenized plant materials.

The field microplot tests confirmed the effectiveness of sudangrass for the control of *M. hapla*, although reduction of the nematode population was only obtained in the second and third years. It is possible that incorporation of sudangrass favors the increase of microbial organisms that are antagonistic to *M. hapla*, and that abundance of these organisms in the first year was not yet adequate to suppress the population of *M. hapla*. Antagonistic fungi have been isolated from juveniles and egg masses of *M. hapla* in organic soils in New York (24). Increase in microbial organisms following the incorporation of organic compounds has been documented (2,20,22). It is also possible that the initial nematode infestation levels were too high in the first year, or that not enough toxic

Fig. 2. Effect of sudangrass as a green manure over three consecutive seasons (1993, 1994, 1995) on egg production of *Meloidogyne hapla* on lettuce roots and on lettuce yield in field microplots at different infestation levels of *M. hapla* (zero = uninfested; low = six eggs per cm³ of soil in test 1, and two eggs per cm³ of soil in tests 2 and 3; high = 24 eggs/cm³ soil in test 1, and 4 eggs/cm³ soil in tests 2 and 3). Bars represent the untransformed means. Data were transformed to log($x + 1$) values before a two-sampled *t* test (one-tailed) was performed at each infestation level of *M. hapla*. ** = significant at 1% level, * = significant at 5% level, ns = not significant.

compounds were released by the decomposing sudangrass. The amount of dhurrin in sudangrass depends on several factors, such as weather conditions, age and height of the plant, and the nitrogen level of the soil (9). It is possible that the plants in the second and third year yielded more hydrogen cyanide than the plants in the first year. This, together with a lower initial nematode density and a possible increase in antagonistic microorganisms, may have contributed to the suppression of the population of *M. hapla* and increased lettuce yields.

Leaving the soil unplanted in the greenhouse tests was as effective as the incorporation of sudangrass in reducing the population of *M. hapla*. There were more eggs of *M. hapla* on roots of lettuce planted after a fallow treatment in the microplots, however, than on roots of lettuce planted after the incorporation of sudangrass. The higher temperature and constant high moisture content of the soil in pots might have resulted in more hatching of nematode eggs soon after they were added to the soil, compared with that in soil in field microplots. As the hatched juveniles were unable to find a host during a period of 12 weeks in the greenhouse tests, it is very likely that most of them died and that few managed to infect the roots of lettuce planted after the incubation period, resulting in low final population densities.

It is not known why the incorporation of green manures of oat, rye, phacelia, oilseed radish, and yellow mustard did not reduce the population of *M. hapla*. Even though oat and rye were poor and nonhosts, respectively, it is possible that egg masses from the initial soil infestation (16 eggs of *M. hapla* per cm³ of soil) survived inside the very dense root systems of these cover crops, or that certain root exudates prevented hatching. Rye was reported to release nematotoxic compounds (18), but its incorporation did not result in reduction of the population of *M. hapla*. Even though oat was not as effective as either sudangrass or the unplanted treatment in reducing the nematode population, its use as a cover or rotation crop is preferred to a fallow treatment because it improves the soil structure and suppresses weeds (1).

It is well known that plants belonging to the Brassicaceae contain glucosinolates (4). It was shown that isothiocyanates and other compounds released upon the hydrolysis of glucosinolates in rapeseed were detrimental for *Caenorhabditis elegans* and *Xiphinema americanum* (13). The reduction of the population of *M. chitwoodi* in soil amended with leaves of rapeseed (*Brassica napus* L.) was directly related to the concentration of glucosinolates present in the plant (16). In another study by Mojtahedi et al. (14), rapeseed cv. Jupiter was effective in reducing the population of *M. chitwoodi*, although the cultivar was a suitable host (R = 5.7). It was hypothesized

that oilseed radish and yellow mustard might be able to release compounds toxic to *M. hapla* when they are incorporated into the soil. Apparently, the amount of toxins produced was insufficient to reduce the number of *M. hapla* in the soil as effectively as soil amendment with sudangrass.

Incorporation of green manures can also result in phytotoxic effects on the growth of subsequent plants. This can be caused by toxic compounds released by the decomposing plant parts or by a low C:N ratio of the added plant tissues (16,19,20,22). Phytotoxic effects may be responsible for the low weight of lettuce obtained in the experiment where all cover crops were incorporated as green manures. Other factors involved were the relatively high initial infestation level of *M. hapla* and the increased water retention of the soil due to the incorporated plant materials. Lettuce seedlings growing in soil amended with sudangrass tissues were noticeably smaller than the other seedlings, especially when stems and leaves were incorporated into the soil. This resulted in lower weights of lettuce at harvest time in soils amended with sudangrass tissues compared with lettuce in unamended soils, regardless of infestation levels of *M. hapla*. Phytotoxic effects on lettuce of a decomposing sorghum-sudangrass hybrid, as well as of decomposing rye, have been reported (6,19). It is very likely that phytotoxic substances are also released during the decomposition of sudangrass cv. Trudan 8.

The results of the greenhouse experiments indicate that oat and rye are able to reduce the population of *M. hapla* under field conditions, as they are poor or nonhosts. However, the further reduction in nematode population by the amendment of sudangrass makes this cover crop a better choice in the management of the northern root-knot nematode. Sudangrass has the ability to reduce *M. hapla* population densities more effectively than any of the other cover crops tested. There is a need to investigate the role of dhurrin in nematode suppression observed in sudangrass-amended soils, as well as other compounds released upon decomposition of the plant material that could be nematocidal. A study of the change in the microflora in the soil after addition of sudangrass could also answer some of the questions related to the mode of action of sudangrass as a nematode suppressor. The impact of growing sudangrass as a cover crop on other plant-parasitic nematodes and their damage to lettuce should also be studied. Future investigation of the effect of the incorporation of cover crops, or of their different tissues, on lettuce yield and population of *M. hapla* should consider in detail the weight and the physiological stage of the incorporated plant tissues, as well as the duration of the decomposition of the cover crop before lettuce planting.

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