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# Suppression of *Pratylenchus penetrans* Populations in Potato and Tomato using African Marigolds

Samuel A. Alexander[1] and Christine M. Waldenmaier[1]

*Abstract:* Current strategies for management of *Pratylenchus penetrans* in both white potato and tomato consist of the use of fumigant or non-fumigant nematicides or crop rotation. The objective of this study was to determine if double-cropping African marigolds (*Tagetes erecta*) with potatoes or tomatoes could reduce *P. penetrans* populations. Plots were 10 m × 3 m arranged in a randomized complete block design with four replications. Treatments included marigolds, potatoes or tomatoes, and natural weedy fallow followed by either potatoes or tomatoes. Nematode populations were sampled before spring planting, between crops in August and after harvest in November. During the 3 years of the study, *P. penetrans* soil population density declined by an average of 93% from the pre-plant level when marigold was grown in rotation with potato and by 98% when marigold was grown.in rotation with tomato. Weedy fallow preceding potato resulted in an average decline in *P. penetrans* soil population density of 38%, and a similar decrease (37%) was seen when fallow preceded tomato. There was a significant reduction in the number of *P. penetrans* found in both potato and tomato roots when the crops followed marigolds. These results suggest that *P. penetrans* population density may be significantly reduced when marigolds are double-cropped with potatoes or tomatoes.

*Key words:* biological control, double-cropped, lesion nematode, marigolds, potato early dying, *Pratylenchus penetrans,* thiophenes, and tomato.

*Pratylenchus penetrans* (Cobb) Filipjev & Schuurmans Stekhoven can be a chronic recurring problem for white potato (*Solanum tuberosum* L.) producers (Oostenbrink, 1958). Damage to potato occurs not only from nematodes feeding but also from interactions with other disease-causing organisms that take advantage of wounds created by nematodes (Abawi and Chen, 1998). This interaction can lead to a syndrome referred to as ''potato early dying,'' where the plants yellow and senesce early and yields are reduced (Burpee and Bloom, 1978; Rowe et al., 1985). Crop rotation is often not effective because many alternative crops are also hosts to *P. penetrans* (Barker, 1998; Chen et al., 1995), making fumigant or contact nematicides necessary to reduce nematode population densities below the economic threshold (Easton et al., 1991; Kimpinski, 1982; Olthof, 1989).

*Pratylenchus penetrans* may also cause significant damage to tomatoes (*Lycopersicon esculentum* Mill.) (Jensen, 1972). Potter and Olthof (1977) reported a decrease in tomato fruit weight and size, as well as delay in fruit ripening when nematode populations reached 230–590 nematodes per g of root. Also, fungal and bacterial root pathogens in tomatoes can be enhanced by the presence of *P. penetrans* (Shurtleff and Averre, 2000).

African marigolds (*Tagetes erecta* L.: tribe tageteae) have been shown to inhibit the reproduction of nematodes (Davide, 1979; Huang, 1984; Ijani and Mmbaga, 1988; Kimpinski et. al., 2000). The active nematicidal compounds in marigolds are thiophenes, which are found in the foliage and roots (Chitwood, 1992; Tang et al., 1986). Studies conducted on the Eastern Shore of Virginia from 1986 to 1993 demonstrated an average reduction in *P. penetrans* of 89%, and an average increase in potato tuber yield of 33% when potatoes were cropped the following year after a full season of marigolds (Baldwin, pers, comm). Experiments conducted in Prince Edward Island, Canada, showed an average increase in potato tuber yield of 10% in potatoes following marigolds over tuber yields of potatoes following other cover crops (Kimpinski et al., 2000).

Planting tomatoes the year following marigolds also reduced the number of *P. penetrans* in tomato roots compared to tomatoes following soybeans (Baldwin, pers. comm). Unfortunately, cropping a field to marigolds for a full growing season keeps the field out of vegetable production for that year. The objective of this study was to determine the effects of double-cropping African marigolds with potatoes or tomatoes on *P. penetrans* soil and root populations, and subsequent yields in potatoes and tomatoes.

## Materials and Methods

The study was conducted at the Eastern Shore Agricultural Research Extension Center (ESAREC) at Painter, Virginia, in different fields in 1996 (Year 1), 1997 (Year 2), and 1999 (Year 3). An experiment in 1998 was lost due to severe drought conditions. Experimental plots were planted in winter rye, which was incorporated in early spring prior to initiation of each study. Two experiments conducted each year were potatoes following a crop of either potatoes or marigolds or following weedy fallow (PMF) and tomatoes following either tomatoes or marigolds or following weedy fallow (TMF). The general timeline for the PMF and TMF experiments was as follows: in late spring, potatoes (cv. Belrus) or tomatoes (cv. Sunbeam), marigolds (cv. Crackerjack), and fallow were established; at midsummer, whole potato plants with tubers or whole tomato plants with fruit were removed, and the marigolds and weedy fallow were mowed and incorporated into

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[1] Associate Professor and Research Specialist, Eastern Shore Agricultural Research and Extension Center, Virgnia Polytechnic Institute & State University, Painter, VA 23420.
E-mail: salex@vt.edu
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the soil; 2 weeks later, either potatoes (PMF) or tomatoes (TMF) were planted in the plots; in early November, potatoes in the PMF and tomatoes in the TMF were harvested from each of the plots.

Potato plots consisted of three 10-m-long rows spaced 1 m apart with a 25-cm plant spacing within the rows. Marigold plots consisted of eight 10-m-long rows spaced 30 cm apart with a 10-cm plant spacing within the row. Tomato plots consisted of three 10-m-long rows spaced 1.5 m apart with a 30-cm plant spacing within the row. Weedy fallow plots were 10 m × 3 m and were left undisturbed to grow in naturally occurring vegetation until planted with potatoes or tomatoes.

The initial potato crop (PMF) was planted using potato seed pieces on 21 May 1996, 15 April 1997, and 12 May 1999. The initial tomato crop (TMF) was transplanted on 23 May 1996, 23 May 1997, and 26 May 1999. Marigolds were direct-seeded on 6 June 1996, 11 June 1997, and 1 June 1999. In all 3 years during the first week of August, potato plants and tubers and tomato plants and fruit were removed, and marigold and weedy fallow plots were mowed and the residue incorporated. The plots were left unplanted between crops for 14 days in Year 1, 11 days in Year 2, and 15 days in Year 3 and then all plots were replanted with potatoes or tomatoes.

The second planting of potato or tomato was made on 19 August 1996, 15 August 1997, and 19 August 1999. Potato tubers were harvested on 11 November 1996, 20 November 1997, and 12 November 1999. Tomato fruits were harvested on 18 November 1996 and 5 November 1997. Yield data were taken from the center row in each plot. Total tomato fruit weight per plot was measured in Year 1. In Year 2, tomato fruit also were graded into large (>6.1-cm-diam.), medium (3.9 to 6.1-cm-diam.), and small (<3.9-cm-diam.) categories. A killing frost occurred before marketable fruit were produced in Year 3, preventing the collection of tomato yield data in Year 3.

The PMF and TMF experiments were established adjacent to each other on a Bojac sandy loam soil with organic matter <1% and pH 6.0. Marigolds were fertilized with 560 kg/ha of 10-10-10 (N,P,K) broadcast incorporated before each planting. Potatoes were fertilized with 1,120 kg/ha of 10-10-10, banded next to the seed prior to each planting. Tomatoes were fertilized with 1,120 kg/ha of 10-10-10 that was broadcast and incorporated prior to each planting. For weed control in potatoes, S-metolachlor 8E at 1.67 kg a.i./ha + metribuzin 4F at 0.56 kg a.i./ha was sprayed over the plots just after the initial planting in the spring and S-metolachlor 8E at 0.84 kg a.i./ha + linuron 50DF at 0.42 kg a.i./ha for the second planting. Weeds were controlled in tomato plots before transplant by incorporating trifluralin 4EC at 0.56 kg a.i./ha + metribuzin 75W at 0.28 kg a.i./ha. Weeds were controlled in the marigold plots with pronamide 50W at 1.12 kg a.i./ha + linuron 50DF at 0.l 1 kg a.i./ha. Imidacloprid 1.6F at 0.06 kg a.i./ha was used for control of the Colorado potato beetle (*Leptinotarsa decemlineata* Say) in potato and tomato, and acephate 75S at 0.84 kg a.i./ha was applied for control of thrips, *Frankliniella tritici* (Fitch), on marigold. Disease control was unnecessary.

*Nematode sampling and extraction:*

*Potato and tomato root extraction.* Twelve plants were systematically sampled in each plot at fall harvest. Plants were removed from the soil, taking care not to damage fine roots. The root systems were removed from each of the 12 plants, placed in a plastic bag, and stored at 10 °C until processed. Roots were washed, sliced into 3-cm lengths, and mixed thoroughly. A 10-g random sample of fine root tissue was removed and placed in a 250-ml Erlenmeyer flask, washed with distilled water, and shaken for 3 days on an Eberbach platform shaker (Eberbach Corp., Ann Arbor, MI). The distilled water surrounding each root sample was decanted onto a sieve with 45-μm pore openings and then back-washed onto a wet-strength tissue in a modified Baermann funnel. After 48 hours of incubation, nematodes were drawn off in 20 ml water and the water volume standardized to 25 ml. The sample was stirred for 5 seconds, and a 5-ml aliquot was pipeted into a clear polystyrene 60 × 15-mm tissue culture dish for nematode counting and identification. Nematodes were counted in the 5-ml subsample and multiplied by 5 to give the number of nematodes in 10 g of roots.

*Soil extraction.* Soil samples were taken prior to planting in the spring, prior to the second planting in the summer, and at harvest in the fall. Fifteen soil cores, 15 to 20 cm deep, were randomly collected from each plot with a standard 2.0-cm i.d. sampling tube, approximately 10 cm from the plant stems. The soil cores from each plot were combined and mixed thoroughly. A 250-$cm^3$ subsample was diluted in 3 liters of water, stirred for 30 seconds, allowed to settle for 60 seconds, and decanted through a 595-μm-pore sieve placed over a 45-μm-pore sieve. Contents of the 45-μm-pore sieve were backwashed onto a wet-strength tissue in a modified Baermann funnel. After 48 hours at room temperature, nematodes were drawn off in 20 ml water and the water volume was standardized to 25 ml. The sample was stirred for 5 seconds before pipeting a 5-ml aliquot into a clear polystyrene 60 × 15-mm tissue culture dish for nematode counting and identification. Counts from the subsample were multiplied by 5 to calculate the number of nematodes per 250-$cm^3$ soil sample.

Data were subjected to log transformation before analysis. Mean separation was calculated by Duncan's new multiple-range test using Agricultural Research Manager software (Gyllings Data Management, Brookings, SD). The nematode data are presented as back-transformed means in the tables.

TABLE 1. Effects of double-cropping marigolds, weedy fallow, and potatoes with potatoes on soil population densities of *Pratylenchus penetrans.*

| | Number of *P. penetrans* per 250 $cm^3$ soil | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | Year 1 | | | Year 2 | | | Year 3 | | |
| First crop[a] | I[b] | II | III | I | II | III | I | II | III |
| Marigold | 29 a | 2 b | 2 b | 98 a | 12 c | 10 b | 10 a | 2 b | 0 b |
| Fallow | 41 a | 5 b | 21 a | 131 a | 65 b | 69 a | 13 a | 10 b | 5 a |
| Potato | 40 a | 42 a | 27 a | 106 a | 364 a | 92 a | 9 a | 44 a | 11 a |

Data are means of four replicates. Means within a column followed by a common letter are not different according to Duncan's new multiple-range test ($P \leq 0.05$). Mean separation was based on log transformation.
[a] Marigolds and fallow were incorporated and potatoes removed in August, and all plots were replanted with potatoes.
[b] Sampling periods: I—preplant, first crop; II—between crops; III—harvest, second crop.

## RESULTS AND DISCUSSION

The PMF and TMF experiments were planted immediately adjacent to one another in different locations in Years 1, 2, and 3. Initial numbers of *P. penetrans* in the soil varied considerably depending on year, with 1997 having the largest initial nematode number in the soil for both potatoes and tomatoes (Tables 1, 4).

In Year 1, the marigold treatment lowered the soil and root nematode populations at harvest (III) in both the PMF and TMF experiments ($P \leq 0.05$) (Tables 1, 2, 4, 5). There were no differences ($P \leq 0.05$) in the number of *P. penetrans* in the soil when potato followed either potato or weedy fallow, or when tomato followed either tomato or weedy fallow in Year 1 (Tables 1, 4). The weedy fallow treatment lowered ($P \leq 0.05$) *P. penetrans* numbers prior to the second potato planting (sampling period II) in the PMF experiment, but this difference was not seen at harvest (sampling period III). The number of lesion nematodes per 10 g of roots was lower ($P \leq 0.05$) in the potatoes following marigolds than in potatoes following either potatoes or fallow (Table 2). Similar results were found in tomatoes when the number of lesion nematodes per 10 g roots was lower in tomatoes following marigold than when tomatoes followed either tomatoes or fallow (Table 5).

In Year 2, nematode numbers in the soil in the PMF experiment were lower ($P \leq 0.05$) after the first crop and at harvest in the potatoes following marigolds or fallow (sampling period II) compared to the potatoes following potatoes, but by harvest (sampling period III) only the potatoes following marigolds was significantly lower ($P \leq 0.05$) (Table 1). In the TMF experiment, nematode numbers in the soil were lower ($P \leq 0.05$) in the tomatoes following marigolds or fallow than in the tomatoes following tomatoes between crops (sampling period II), but by harvest (sampling period III) only the tomatoes following marigolds resulted in lower *P. penetrans* numbers (Table 4). The number of root-inhabiting nematodes was lower ($P \leq 0.05$) in the potatoes or tomatoes following marigolds than in the potatoes or tomatoes following fallow, potatoes, or tomatoes (Tables 2, 5).

In Year 3, soil population densities of *P. penetrans* were low at the beginning of the experiment in all plots in both the PMF and TMF experiments (Tables 1, 4). In the PMF experiment, nematode numbers in the potatoes following marigold and fallow were lower ($P \leq 0.05$) than in the potatoes following potatoes (sampling period II). By harvest the numbers were so low in all plots that there was no separation between treatment means ($P > 0.05$). The soil nematode population density in the tomatoes following marigolds in the TMF was lower ($P \leq 0.05$) at harvest than in the tomatoes following fallow or tomatoes (Table 4). The number of nematodes found in the potato roots in the PMF and in tomato roots in the TMF experiments were lower ($P \leq 0.05$) where marigolds were grown than in the fallow or tomato treatments (Tables 2, 5).

Overall, in the PMF experiments, the marigold treat-

TABLE 2. Effects of marigolds, weedy fallow, and potatoes double-cropped in a single season with potatoes on the number of *Pratylenchus penetrans* in potato roots at harvest.

| | Number of *P. penetrans* per 10 g roots | | |
|---|---|---|---|
| First crop[a] | Year 1 | Year 2 | Year 3 |
| Marigold | 10 b | 31 b | 6 b |
| Fallow | 501 a | 141 a | 47 a |
| Potato | 549 a | 161 a | 51 a |

Data are means of four replicates. Means within a column followed by a common letter are not different according to Duncan's new multiple-range test ($P \leq 0.05$). Mean separation was based on log transformation.
[a] Marigolds and fallow were incorporated and potatoes removed in August, and all plots were replanted with potatoes.

TABLE 3. Effects of marigolds, weedy fallow, and potatoes double-cropped in a single season with potatoes on tuber yield.

| | Tuber yield (kg/ha) | | |
|---|---|---|---|
| First crop[a] | Year 1 | Year 2 | Year 3 |
| Marigold | 5,831 a | 3,071 a | 9,251 a |
| Fallow | 6,478 a | 3,158 a | 9,711 a |
| Potato | 5,546 a | 2,997 a | 5,496 b |

Data are means of four replicates. Means within a row followed by a common letter are not different according to Duncan's new multiple-range test ($P \leq 0.05$).
[a] Marigolds and fallow were incorporated and potatoes removed in August, and all plots were replanted with potatoes.

TABLE 4. Effects of double-cropping marigolds, weedy fallow, and tomatoes with tomatoes on soil population densities of *Pratylenchus penetrans*.

| | Number of *P. penetrans* per 250 cm$^3$ soil | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | Year 1 | | | Year 2 | | | Year 3 | | |
| First crop[a] | I[b] | II | III | I | II | III | I | II | III |
| Marigold | 58 a | 2 a | 2 b | 137 b | 13 c | 2 b | 26 a | 2 a | 0 b |
| Fallow | 53 a | 19 a | 31 a | 213 a | 121 a | 83 a | 12 a | 14 a | 7 a |
| Tomato | 85 a | 29 a | 43 a | 154 ab | 37 b | 76 a | 23 a | 13 a | 10 a |

Data are means of four replicates. Means within a column followed by a common letter are not different according to Duncan's new multiple-range test ($P \leq 0.05$). Mean separation was based on log transformation.

[a] Marigolds and fallow were incorporated and tomatoes removed in August, and all plots were replanted with tomatoes.

[b] Sampling periods: I—preplant, first crop; II—between crops; III—harvest, second crop.

ments were more effective in reducing the *P. penetrans* populations in the soil than either the fallow or potato treatments. In general, soil *P. penetrans* populations were reduced by an average of 55% by the marigold treatments over the fallow treatments, and greater than 87% over the potato treatments. Soil lesion nematode numbers for the PMF experiment were reduced by an average of 93%, 38%, and 14%, respectively, for the marigold, weedy fallow, and potato treatments over all 3 years. The marigold treatment was consistently more effective than either the fallow or potato treatments in reducing the number of *P. penetrans* in the roots. In the TMF experiments the number of nematodes in the soil was numerically lower in the marigold treatment at each successive sampling period for all 3 years. Soil lesion nematode numbers were reduced in the TMF experiment by an average percent of 98%, 37%, and 30% for the tomatoes following marigolds, weedy fallow, and tomato, respectively, for all 3 years. Nematode numbers in the tomato root tissue also were lower in each of the 3 years in the marigold treatments than in the other treatments.

Nematode numbers in the soil tended to increase more where potatoes followed potatoes than where tomatoes followed tomatoes, but nematode numbers in the roots were generally higher in the TMF experiment than in the PMF experiment. This may indicate that tomato was a more suitable host for *P. penetrans* than potato. Nematode soil counts for potatoes and tomatoes following weedy fallow decreased by the end of the experiments in late fall (as would normally occur at that time of the year).

Potato yields in the PMF experiment were higher ($P \leq 0.05$) in the potatoes following marigolds and fallow than in the potatoes following potatoes only in Year 3 (Table 3). The statistical differences in yields in Year 3 were probably not associated with the number of nematodes because they were considerably lower than the previous 2 years. Most likely, soft rot bacteria initially developing in the first potato crop increased during a very wet September (>7.6 cm over the 59-year average rainfall) and reduced the final yields in the potatoes following potatoes. The highest number of nematodes in the soil occurred in Year 2, where the average was 423 nematodes/250 cm$^3$ soil. This is equivalent to approximately 1,692 nematodes per kg soil. Olthof and Potter (1973) showed tuber yield reduction in potatoes when nematode numbers in the soil were greater than 2,000 per kg soil. Similarly, Kimpinski and McRae (1988) found large tuber yield increases in Superior potato when nematode populations at planting were at least 3,900 per kg soil. Initial nematode numbers in the soil were highest in Year 2, however, and averaged approximately 447 nematodes per kg. Low initial lesion nematode populations may explain the lack of yield response in all 3 years in the PMF experiment.

Fruit yields in the TMF experiment were highest ($P \leq 0.05$) in the tomatoes following marigold and fallow in Year 1 (Table 6). In Year 2, percent large-size tomato fruit (fruit >6.8 cm diam.) was higher ($P \leq 0.05$) in the tomatoes following marigolds than in both the tomatoes following fallow or tomatoes following tomatoes (Table 6). Percent large fruit was also higher ($P \leq 0.05$) in the tomatoes following fallow than in the tomatoes following tomato (Table 6).

In summary, *P. penetrans* can be a serious problem in vegetable production (Jensen, 1972; Olthof and Potter,

TABLE 5. Effects of marigolds, weedy fallow, and tomatoes double-cropped in a single season with tomatoes on the number of *Pratylenchus penetrans* in tomato roots at harvest.

| | Number of *P. penetrans* per 10 g roots | | |
|---|---|---|---|
| First crop[a] | Year 1 | Year 2 | Year 3 |
| Marigold | 4 b | 43 b | 3 b |
| Fallow | 557 a | 574 a | 52 a |
| Tomato | 671 a | 472 a | 73 a |

Data are means of four replicates. Means within a column followed by a common letter are not different according to Duncan's new multiple-range test ($P \leq 0.05$). Mean separation was based on log transformation.

[a] Marigolds and fallow were incorporated and tomatoes removed in August, and all plots were replanted with tomatoes.

TABLE 6. Effects of marigold, weedy fallow, and and tomatoes double-cropped in a single season with tomatoes on fruit yield.

| | Fruit yield in kg/ha | | | |
|---|---|---|---|---|
| | | Year 2 | | |
| First crop[a] | Year 1 | Total | % Large[c] | Year 3[b] |
| Marigold | 2,311 a | 6,128 a | 31 a | --- |
| Fallow | 2,281 a | 6,769 a | 26 b | --- |
| Tomato | 1,484 b | 6,322 a | 21 c | --- |

Data are means of four replicates. Means within a column followed by a common letter are not different according to Duncan's new multiple-range test ($P \leq 0.05$).

[a] Marigolds and fallow were incorporated and tomatoes removed in August, and all plots were replanted with tomatoes.

[b] An early frost prevented the collection of yield data in Year 3.

[c] Fruit >6.8 cm diam.

1973; Oostenbrink, 1958). Losses may occur either directly due to*P. penetrans* or indirectly through interactions with other pathogens (Rowe et al., 1985; Shurtleff and Averre, 2000). *Tagetes* sp. have been shown to reduce nematode populations in many different crops (Davide, 1979; Huang, 1984; Ijani and Mmbaga, 1988; Kimpinski et al., 2000). The future loss of methyl bromide, environmental concerns about the use of many pesticides, and increased demands for organically grown vegetables make alternative methods for nematode management, such as crop rotation with marigolds, attractive. In developing countries the use of marigolds may be particularly applicable due to either lack of availability of nematicides or expense of applying them.

In this study, marigolds double-cropped with potato or tomato in the same growing season lowered lesion nematode populations. Double-cropping within the same year allows the field to be kept in production each year instead of every other year, as reported in other studies (Davide, 1979; Huang, 1984; Kimpinski et al., 2000). African marigolds double-cropped with potatoes or tomatoes in the same year significantly reduced ($P \leq 0.05$) *P. penetrans* populations in the soil and roots. This study suggests that planting marigolds prior to potatoes or tomatoes in the same growing season can reduce *P. penetrans* populations both in the soil and in the roots.

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