REPRODUCTION OF *MELOIDOGYNE* SPECIES ON YELLOW GRANEX ONION AND POTENTIAL YIELD SUPPRESSION

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ABSTRACT

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The suitability of onion (*Allium cepa* cv. Sweet Vidalia) as a host for *Meloidogyne incognita*, *M. arenaria*, and *M. javanica* was evaluated. The effect of *M. incognita* on yield and economic return of direct-seeded and transplanted onions also was evaluated. Nematode reproduction was evaluated in two greenhouse trials with six replications each. All three nematode species increased with final egg counts of 19,300 for *M. incognita*, 32,100 for *M. arenaria*, and 40,350 for *M. javanica* in the first trial and 167,200 for *M. incognita*, 71,600 for *M. arenaria*, and 101,950 for *M. javanica* in the second trial. Final egg counts were similar ($P \geq 0.05$) among the three species in the first trial, but *M. incognita* produced more eggs ($P \leq 0.05$) than *M. arenaria* in the second trial. The application of 1,3-D in direct-seeded onions increased the weight (kg/ha) of large and colossal sizes in both seasons and the weight of small and medium sizes in 2002-2003. In transplanted onions, the weight of colossal onions was increased in 2001-2002, but weights were unaffected in 2002-2003. Onion is a good host for all three *Meloidogyne* species tested, and *M. incognita* can reduce yields and economic return when onions are direct-seeded. Transplanted onions in this study did not suffer economic loss.

Key words: 1,3-dichloropropene, *Allium cepa*, economic loss, host status, *Meloidogyne arenaria*, *Meloidogyne incognita*, *Meloidogyne javanica*, root-knot nematodes.

RESUMEN

Davis, R. F., and D. B. Langston. 2003. Reproducción de especies de *Meloidogyne* y supresión potencial de cosecha de cebolla amarilla Granex. Nematropica 33:179-188.

La indoneidad de cebolla (*Allium cepa* cv. Sweet Vidalia) como huésped de *Meloidogyne incognita*, *M. arenaria*, y *M. javanica* fue evaluada. El efecto de *M. incognita* sobre la cosecha e ingresos comerciales de cebollas sembradas de semilla y cebollas trasplantadas también fue evaluado. Reproducción de nemátodos fue evaluada en dos experimentos de invernadero, con seis replicaciones cada uno. Todas las especies de nemátodos incrementaron, con números de huevos de 19,300 para *M. incognita*, 32,100 para *M. arenaria*, y 40,350 para *M. javanica* en el primer experimento y 167,200 para *M. incognita*, 71,600 para *M. arenaria*, y 101,950 para *M. javanica* en el segundo experimento. Números finales de huevos eran similares entre las tres especies en el primer experimento ($P \geq 0.05$), pero *M. incognita* produjo mas huevos ($P \leq 0.05$) que *M. arenaria* en el segundo experimento. La aplicación de 1,3-D en cebollas sembradas de semilla incrementó el peso (kg/ha) de los tamaños grande y colosal en los dos períodos, y el peso de los tamaños pequeños y medianos en el 2002-2003. En cebollas trasplantadas el peso de cebollas colosales incrementó en el 2001-2002, pero el peso no fue afectado en 2002-2003. La cebolla es un buen huésped para todos las tres especies de *Meloidogyne* ensayadas, y *M. incognita* puede reducir la cosecha y la ganancia económica cuando las cebollas se plantan de semilla. Cebollas trasplantadas no sufrieron pérdidas económicas en este estudio.

Palabras claves: 1,3-dichloropropano, *Allium cepa*, pérdida económica, estado del huésped, *Meloidogyne arenaria*, *Meloidogyne incognita*, *Meloidogyne javanica*, nemátodos agalladores.

INTRODUCTION

Several species of root-knot nematodes are reported to parasitize onion (*Allium cepa* L.). *Meloidogyne graminicola* Golden and Birchfield and *M. hapla* Chitwood, have been documented to cause yield suppression (Gergon *et al.*, 2002; MacGuidwin *et al.*, 1987; Sherf and Stone, 1956), and *M. chitwoodi* Golden *et al.* can be pathogenic to some onion cultivars, though other cultivars are non-hosts (Mojtahedi *et al.*, 1987; Westerdahl, *et al.*, 1993). Onion is documented as a host for *Meloidogyne incognita* (Kofoid and White) Chitwood, *M. javanica* (Treub) Chitwood, and *M. arenaria* (Neal) Chitwood, but their relative reproductive ability on onion has not been reported (Glazer *et al.*, 1985; Martin, 1958; Martin, 1961).

Onion is grown worldwide and is common in most countries. *Meloidogyne incognita* and *M. javanica* are common throughout the world, and *M. arenaria* is found less frequently, though it also is widely distributed (Shepherd and Barker, 1990). *Meloidogyne incognita*, *M. javanica*, and *M. arenaria* are present in some of the major onion-producing states in the U.S. such as California, Texas, and Georgia, and there is the potential that onions could be planted in fields with high population densities of these nematodes. Soil samples from onion fields in Georgia often contain juveniles of *Meloidogyne* sp., sometimes at densities of several hundred per 150 cm^3 of soil (R. F. Davis, unpublished), though it is not known which species are present or if the onion crop has been damaged. A previous study found that the yield of direct-seeded onions was negatively correlated with final population densities of *M. incognita* (Hall *et al.*, 1988), but the relationship between *M. incognita* and transplanted onions is not known. Onions produced commercially in Georgia are transplanted, and producers do not use nematicides because it is not known if the *Meloidogyne* species found in their fields can damage transplanted onions.

Our primary objective was to determine the relative abilities of *M. incognita*, *M. javanica*, and *M. arenaria* to reproduce on yellow Granex onion. A secondary objective was to document the effect of *M. incognita* on yield and economic return of direct-seeded and transplanted yellow Granex onion.

MATERIALS AND METHODS

The relative reproduction of *M. incognita* race 3, *M. javanica*, and *M. arenaria* race 1 on yellow Granex onion (cv. 'Sweet Vidalia') was measured in two greenhouse trials in 2003. Onion transplants were grown in 15-cm-diameter pots with one plant per pot. Each pot held approximately 1.5 l of pasteurized soil (Tifton loamy sand; 83% sand, 9% silt, 7% clay, and ≤1% organic matter). Six replicate pots for each nematode species were arranged in a completely randomized design. Eggs of the three nematode species were collected (Hussey and Barker, 1973) for inoculum from the roots of tomato (*Lycopersicon esculentum* Mill, 'Rutgers') and each pot was inoculated with 8,000 eggs of one species. Inoculum was placed into two 3-cm-deep holes, one on each side of the plant. Soil temperatures during the study varied between 16 and 32°C. The first trial was inoculated on 17 January 2003 and the second trial was inoculated on 30 January 2003.

Nematode eggs were extracted from the onion roots 54 days after inoculation. The entire root system of a single plant was cut into 5-cm pieces, placed in a 1-liter flask, and agitated for 4 minutes in a 1% NaOCl solution (Hussey and Barker, 1973). Eggs were collected, rinsed with tap

water on nested 150- and 25-µm-pore sieves, and a 1 ml subsample was counted. A two-way analysis of variance followed by Fisher's protected LSD ($P \leq 0.05$) was used to determine differences in reproduction among the three species. Additional data collected from the greenhouse trials included fresh root weight, fresh bulb weight, and fresh foliage weight. Prior to extraction of eggs, root galling was rated on a linear 0 to 10 scale where 0 = no galling, 1 = 1-10% of the root system galled, 2 = 11-20% galled, 3 = 21-30% galled, and so forth with 10 = 91-100% galled.

Yield suppression of yellow Granex onion (cv. 'Sweet Vidalia') was evaluated in field studies on the University of Georgia Blackshank farm in the 2001-2002 season and on the Gibbs farm in the 2002-2003 season in Tifton, GA. Onions in Georgia are either seeded in August or September or transplanted in November or December and harvested in April or May of the following year. Soil at the Blackshank site was a Fuquay loamy sand (loamy, siliceous, thermic Arenic Plinthic Paleudults; 88% sand, 9% silt, 3% clay, <1% organic matter) and soil at the Gibbs site was a Tifton loamy sand (fine, loamy, siliceous, thermic Plinthic Kandiudults; 85% sand, 11% silt, 4% clay, <1% organic matter). Both fields were naturally infested with *M. incognita*. The Blackshank site had been planted with cowpeas (*Vigna unguiculata* (L.) Walp.) and the Gibbs site had been planted with okra (*Abelmoschus esculentus* (L.) Moench) during the summer prior to planting onions and visual inspection of roots had confirmed the presence of root-knot nematodes. The experimental design was a split-plot with six replications in which whole-plots were direct-seeded onions or transplanted onions and sub-plots were rates of the soil fumigant 1,3-dichloropropene (1,3-D) (97.5% active ingredient at 0.0, 37.4, 74.8, or 112.2 l of formulated product per ha). The fumigant was applied approximately 30-cm deep on 9 October 2001 and 2 October 2002 for direct-seeded onions and on 19 November 2001 and 26 November 2002 for transplants.

All plots were on 1.8-m-wide raised beds which were formed by an implement that incorporated a rototiller followed by a bed shaper with attached subsoil chisels spaced 0.3 m apart. Plots were 9.1 m long and consisted of 4 rows of onions spaced 35.5 cm apart on a single bed. Data collection was restricted to 6.1-m sections in the center of the middle two rows. Direct-seeded onions were planted at a rate of 6.7 seed/m of row on 23 October 2001 and 22 October 2002. Transplants, spaced 15 cm apart (6.7 plants/m of row), were planted on 5 December 2001 and 12 December 2002. Applications of fertilizer, insecticides, and herbicides followed University of Georgia Extension Service recommendations and were the same for plots with the same planting method but differed between direct-seeded and transplanted plots. In both growing seasons, plots were hand weeded twice because the recommended herbicide regimes were insufficient. Overhead irrigation was applied as needed.

Onions were pulled up and allowed to air dry for three days before the foliage and roots were clipped off and the bulbs were bagged. Bulbs were harvested on 6 May 2002 and 6 May 2003, and were cured in a drying bin at 37°C for approximately 48 hours prior to grading and weighing. Air was circulated by fans through the drying bins. Onions were classified by industry standards (U.S. Department of Agriculture, 1995) as culls, small, medium, large, or colossal (also called jumbo) and then weighed. Data analysis was performed only on the weights of marketable bulbs, which included all sizes except culls.

Soil samples for nematode assay were collected on 8 October 2001 and 1 May 2002 from the first trial and on 2 October 2002 and 29 April 2003 from the second trial. Soil samples consisted of a composite of 8 to 10 cores per plot (2.5-cm diameter and approximately 20-cm deep) collected from each plot. Nematodes were extracted from 150 cm^3 of soil by centrifugal flotation (Jenkins, 1964).

Degree-days were calculated for each growing season from soil temperature data collected at a 20-cm depth in a field approximately 50 m away. Soil temperature was recorded every 50 minutes, and a daily mean temperature was calculated. A threshold temperature for development of *M. incognita* was assumed to be 10°C (Roberts and Van Gundy, 1981; Tyler, 1933), and degree-days were calculated as daily mean temperature minus the base developmental temperature. Soil temperatures were recorded during the first season of the study beginning 1 January 2002 and during the second season beginning 1 December 2002, though onions were planted or transplanted before those dates.

Greenhouse reproduction, root galling, and plant growth data were analyzed by analysis of variance with mean separation by Fisher's protected least significant difference ($P \leq 0.05$). Onion yield and economic return data was analyzed by analysis of covariance with rate of 1,3-dichloropropene as a quantitative factor and planting method (direct-seeded or transplanted) as a qualitative factor. Nematode population density data from the field trials was analyzed by analysis of variance with mean separation by Fisher's protected LSD ($P \leq 0.05$). Economic analysis was based on estimated average production costs in Georgia of $7,837/ha for transplanted onions and $7,125/ha for direct-seeded onions. The cost of 1,3-D was estimated to be $3.04/l. Estimated economic return per hectare data was analyzed by analysis of covariance with rate of 1,3-dichloropropene as a quantitative factor and planting method as a qualitative factor.

RESULTS

The three *Meloidogyne* species increased in number in both greenhouse trials, however, the increases were greater in the second trial and differed by species (trial × species interaction, $P = 0.006$), which precluded combining data. Mean reproduction factors (final egg count divided by inoculum level) ranged from 2.4 to 5.0 in the first trial and 9.0 to 20.9 in the second trial. Mean root gall indices, fresh bulb weight, fresh root weight, and fresh foliage weight did not differ among nematode species in either trial (Table 1). Final egg counts did not differ among species in the first trial, but *M. incognita* produced more eggs than *M. arenaria* in the second trial (Table 1).

Population densities of *M. incognita* were below expectations in the 2001-2002 season with a mean of 9 juveniles per 150 cm^3 of soil prior to fumigation. In the 2002-2003 season, mean density prior to fumigation was 357/150 cm^3. Nematode densities prior to fumigation did not differ among plots in either growing season. Nematode densities in the spring, as measured by juveniles in the soil, were very low: no juveniles were recovered from soil samples on 1 May 2002, and the mean density on 29 April 2003 was 2 juveniles per 150 cm^3 of soil. The accumulated degree-days above 10°C were estimated to be 1,077 in the 2001-2002 season and 1,027 in the 2002-2003 season.

Fumigation with 1,3-D had a more consistent effect on direct-seeded onions than on transplanted onions. The application of 1,3-D increased the weight (kg/ha) of

Table 1. Meloidogyne incognita, M. javanica, and M. arenaria egg counts, root gall indices, fresh bulb weight, fresh root weight, and fresh foliage weight of yellow Granex onions in two greenhouse trials eight weeks after inoculation with 8,000 eggs.

Trial	Species	Eggs		RGI[1]		Bulb weight[2]		Root weight[2]		Foliage weight[2]	
1	*M. incognita*	19,300	a	4.2	a	18.1	a	9.2	a	48.5	a
1	*M. javanica*	40,350	a	7.0	a	17.4	a	11.3	a	43.3	a
1	*M. arenaria*	32,100	a	5.0	a	11.0	a	8.8	a	38.2	a
2	*M. incognita*	167,200	a	6.8	a	28.4	a	10.8	a	42.6	a
2	*M. javanica*	101,950	ab	6.8	a	31.8	a	11.2	a	64.0	a
2	*M. arenaria*	71,600	b	5.5	a	20.5	a	9.9	a	42.6	a

[1]Root gall index on a 0-10 scale where 0 = no galling, 1 = 1-10% of the root system galled, 2 = 11-20% galled, etc.
[2]All weights are in grams and are fresh weights measured immediately after harvest and before egg extraction.
[3]Means within a trial followed by the same letter are not significantly different according to Fisher's protected least significant difference ($P \leq 0.05$).

large and colossal direct-seeded onions in the 2001-2002 season and the weight of small, medium, large and colossal direct-seeded onions in 2002-2003 (Fig. 1). The weight of colossal transplanted onions was increased in 2001-2002, whereas the weight of medium transplanted onions decreased in 2001-2002. The weights of transplanted onions were unaffected by fumigation with 1,3-D in the 2002-2003 season. Significant regressions for each method of planting that relate the weight of onions of a given size to the rate of 1,3-D applied are given in Table 2.

Economic return was not affected significantly by fumigation in 2001-2002 in either direct-seeded or transplanted onions (Fig. 2). In 2002-2003, the economic return of direct-seeded onions was increased by fumigation, though the economic return of transplanted onions was not. The regression equation that described the increase in value in 2002-2003 of direct-seeded onions with increasing rates of 1,3-D was value ($/ha) = 15140 + 151.34*(1,3-D rate) ($R^2 = 0.49$).

DISCUSSION

It is clear that all three species are capable of significant reproduction on onion and must be considered potential pathogens. This corroborates reports that onion is a host for *M. incognita*, *M. arenaria*, and *M. javanica* (Glazer *et al.*, 1985; Martin, 1958; Martin, 1961), and provides new information that these species can increase on onion. Though inoculum levels in greenhouse trials were the same for *M. incognita*, *M. javanica*, and *M. arenaria*, differences in inoculum survival among nematode species and between trials probably occurred. Unfortunately, egg hatch was not measured, so inoculum survival is not known. Differential inoculum survival could explain both the greater reproduction of all species and the differential reproduction of species in the second trial. *Meloidogyne javanica* and *M. arenaria* had levels of reproduction similar to each other in both trials, and such consistency indicates that the nematodes have similar reproductive ability on onion. The results of *M. incog-*

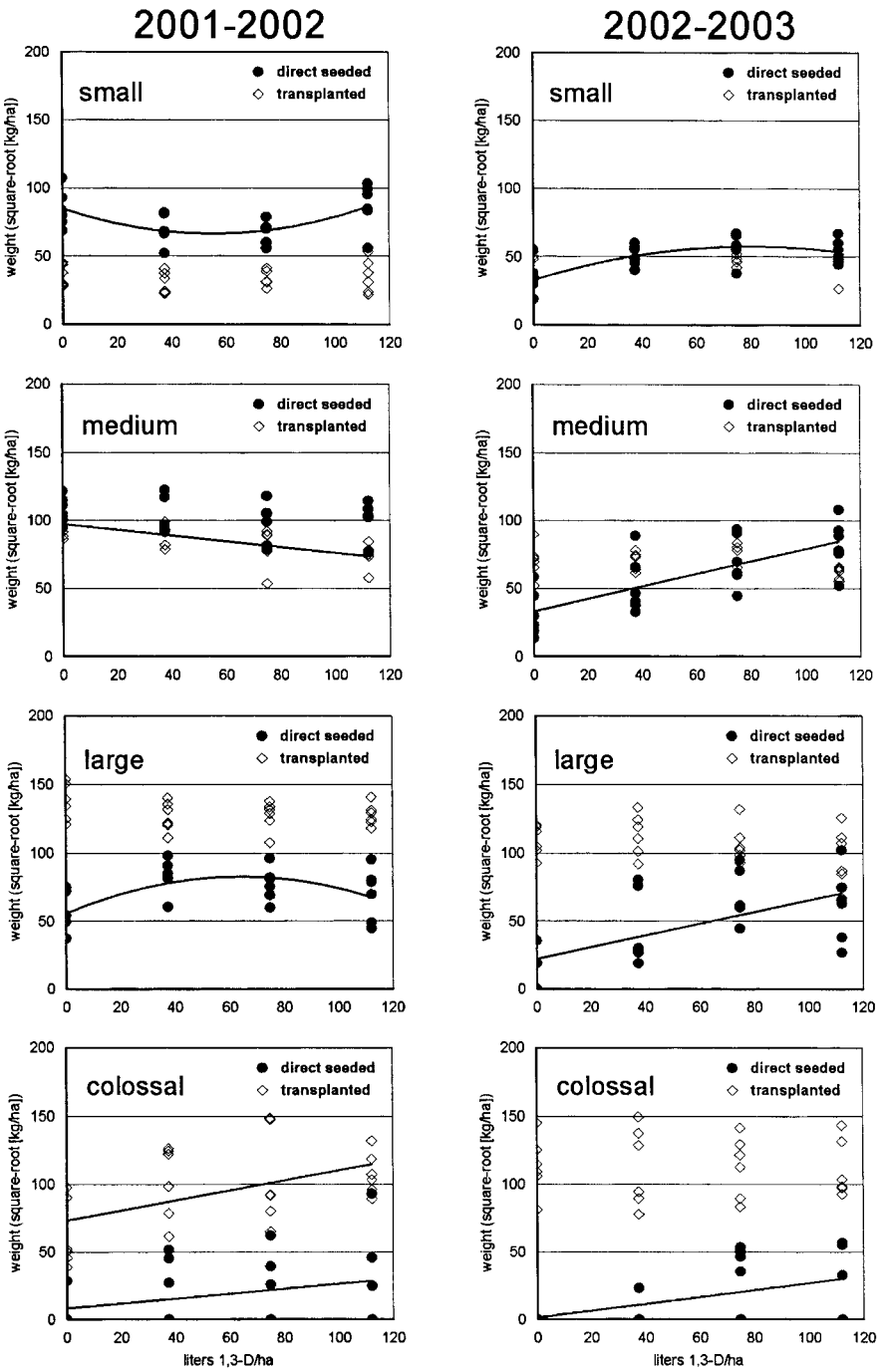

Fig. 1. The relationships between the yields of small medium, large, and colossal size onions and rate of fumigation with 1,3-dichloropropene in the 2001-2002 and 2002-2003 seasons for direct-seeded and transplanted onions. Only significant regression lines ($P \leq 0.05$) are shown.

Table 2. Equations with non-zero regression coefficients relating rate of 1,3-D applied to the weight of onions harvested.

Planting method and onion size	Regression equation[1]	R^2
2001-2002		
small direct-seeded	84.78-6.24*(1,3-D rate)+0.53*(1,3-D rate)^2	0.34
large direct-seeded	55.68+7.83*(1,3-D rate)-0.57*(1,3-D rate)^2	0.36
colossal direct-seeded	8.25+1.71*(1,3-D rate)	0.09
medium transplanted	97.54-2.00*(1,3-D rate)	0.44
colossal transplanted	73.52+3.468*(1,3-D rate)	0.26
2002-2003		
small direct-seeded	32.90+5.74*(1,3-D rate)-0.33*(1,3-D rate)^2	0.50
large direct-seeded	33.22+4.29*(1,3-D rate)	0.54
large direct-seeded	22.18+4.02*(1,3-D rate)	0.39
colossal direct-seeded	1.36+2.39*(1,3-D rate)	0.24

[1]Weight calculated by equations is the square root of kg/ha.

nita reproduction were not consistent between trials, so the reproductive ability of *M. incognita* relative to the other two species cannot be concluded with confidence.

Despite demonstration in the greenhouse of the potential for reproduction of *M. incognita* on onion, our study did not document *M. incognita* reproduction in the field during the growing season. During both onion growing seasons, soil temperatures in Tifton were high enough to accumulate more than 1,000 degree-days above 10°C, so soil temperatures were adequate to allow *M. incognita* to complete at least one generation (Tyler, 1933). Even though the 2002-2003 season began with relatively high nematode densities, very few nematodes were detected in soil samples the following spring, even in non-fumigated plots. The amount of winter attrition in our study is unknown, and winter attrition would affect the final nematode population levels. Similar results with *M. incognita* have been reported previously (Hall *et al.*, 1988). This seems to indicate that soil samples collected in late April or early May do not accurately reflect the nematode population densities in a field. Nematodes were not extracted from roots, which may have provided a better estimate of nematode population levels. It seems likely that population increases could be documented where significantly more degree-days accumulate during the onion growing season, as would be expected in locations with warmer winters or where onions are planted in the spring rather than the fall.

Nearly all of the onions currently produced in Georgia are transplanted rather than direct-seeded. However, many growers are interested in switching to direct seeding because the cost of production would be reduced by approximately $700 per hectare. Total onion yields and economic return were lower for direct-seeded onions than for transplanted onions in both growing seasons in our study regardless of 1,3-D fumigation, so switching to

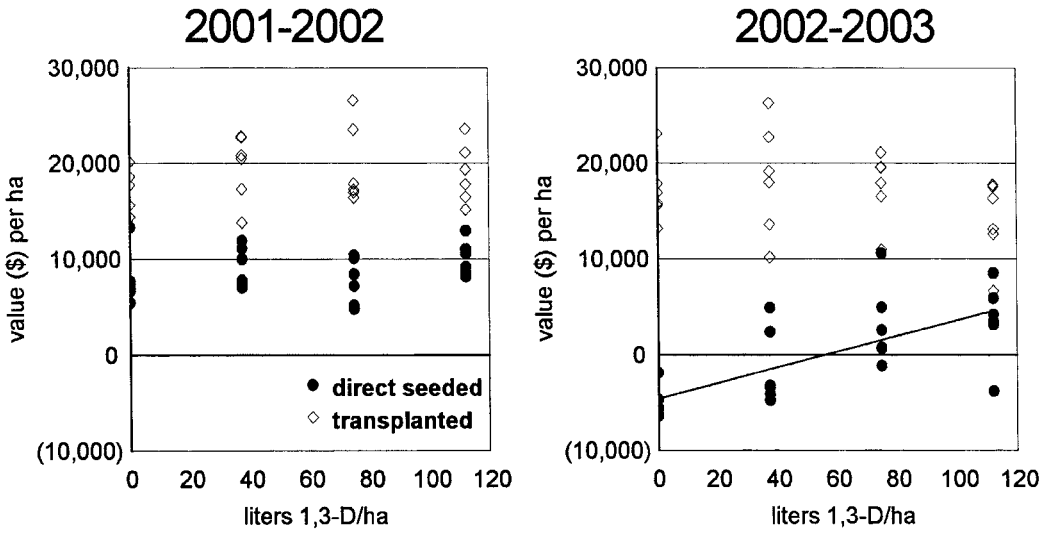

Fig. 2. The relationships between estimated economic return and rate of fumigation with 1,3-dichloropropene in the 2001-2002 and 2002-2003 seasons for direct-seeded and transplanted onions. Only significant regression lines ($P \leq 0.05$) are shown.

direct-seeded onion production should not be recommended unless yields and profitability are improved.

One reason that direct-seeded onions produced less than transplanted onions in our study was that plant spacing was less uniform which resulted in some onions being more crowded and smaller. Another reason was that weeds were more of a problem in direct-seeded onion plots. Fields are tilled much later in the year for transplanted onions, which results in fewer and smaller weeds in the spring, and herbicide regimes are different. In our study, plots were weeded by hand to supplement herbicide applications, but differential weed growth could have affected yields.

In our study, damage attributable to *M. incognita* was observed in both growing seasons, but damage was more widespread and resulted in reduced economic return only in direct-seeded onions in the second season. This is consistent with a previous report that fumigation with DD (1,3-dichloropropene, 1,2-dichloropropane) increased yield of direct-seeded onion in a field infested with *M. incognita* (Hall *et al.*, 1988). A likely reason for the difference between seasons in our study is that *M. incognita* population densities were much lower in the first season, though visual inspection of galled roots from the crop preceding onion in the first season suggested to us that nematode pressure was higher than indicated by the soil samples. Additional studies need to be conducted to determine a damage threshold level for direct-seeded onions.

In summary, *M. incognita*, *M. arenaria*, and *M. javanica* are all capable of significant reproduction on yellow Granex onion, though soil temperatures during the onion growing season in Georgia probably limit reproduction of these nematodes to one or two generations. Soil temperatures may not be so limiting in other onion production systems. Direct-seeded onions can suffer significant reductions in yield, which results in reduced economic return. Transplanted onions appear

to be more tolerant and suffer little or no reduction in yield or economic return, though it is not known if transplanted onions would be less tolerant in areas where more degree-days accumulate. Regardless of nematode damage, direct-seeded onions in Georgia have a lower economic return than transplanted onions even though production costs are significantly lower. If the economic return of direct-seeded onions can be improved to the point where growers begin to use direct seeding, then nematode management will become an important part of onion production. Transplanted onions did not suffer significant damage or economic loss in either year of our study. Initial infection by nematodes on transplanted onions occurs on larger and older plants, which likely caused them to be more tolerant of feeding by nematodes. Reports of significant damage in transplanted onions caused by *M. graminicola* show that tolerance imparted by transplanting can be overcome (Gergon *et al.*, 2001; Gergon *et al.*, 2002). Because onions in Georgia are transplanted late in the fall when soil temperatures are approaching the minimum temperature needed for development of *M. incognita*, nematode development and reproduction would be reduced compared to that on direct-seeded onions which are available for parasitism when temperatures are warmer.

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