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Influence of Poultry Litter Applications on Nematode Communities in Cotton Agroecosystems

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Abstract: The effects of the application of poultry litter at 0.0, 6.7, 13.4, and 20.1 tons/ha on population changes during the growing season on nematode communities were evaluated in two cotton production fields in North Carolina. Numbers of bactivorous nematodes increased at midseason in response to the rate at which litter was applied but decreased with increasing litter application rates at cotton harvest. Numbers of fungivores at cotton harvest were related positively to the rate of litter applied, and this affected a positive increase in the fungivore-to-bacterivore ratio at this sampling date. The rate at which poultry litter was applied resulted in an increase in the bacterivore to plant-parasite ratio, and this corresponded with increased cotton lint yield. Trophic diversity was increased by litter application rate at cotton harvest at one location but not at another. The plant-parasite maturity index was greater consistently at one site than at a second site where the *Hoplolaimus columbus* population density was above the damage threshold for cotton. The population density of *H. columbus* was suppressed with increasing rates of poultry litter application, but other plant-parasitic nematodes were affected marginally.

Key words: Columbia lance nematode, community structure, cotton, ecology, *Gossypium hirsutum, Helicotylenchus dihystera, Hoplolaimus columbus,* management, nematode, *Paratrichodorus minor,* poultry litter, population changes, *Pratylenchus brachyurus,* soil health, trophic groups.

The sustainability of agricultural ecosystems and production of food and fiber are increasingly the focus of public policy (Barker and Koenning, 1998; McSorley and Porazinska, 2001). Also, the demand for agricultural products grown with reduced or no synthetic inputs has increased dramatically in recent years (Thompson, 1998). Soil-inhabiting nematodes are important in nutrient cycling and serve to regulate many soil chemical and biological processes (Bongers and Ferris, 1999; Bulluck et al., 2002; Ferris et al., 1996; Freckman, 1988; Freckman and Ettema, 1993; Ingham et al., 1985; McSorley and Frederick, 1999; Porazinska and Coleman, 1995; Tu et al., 2003). The impacts of various cultural practices, including cropping systems, crops, tillage, inputs of synthetic or organic pesticides and fertilizer on nematode communities have been studied, but knowledge about these communities in cotton is generally lacking.

North Carolina is a leading state in poultry production. This industry generates enormous quantities of wastes that require environmentally acceptable means of disposal. These materials (manure/litter) generally are applied to agricultural land. An estimated 1.27 million tons of poultry waste with 37, 44, and 25 thousand tons of nitrogen, phosphate, and potash, respectively, are produced annually in North Carolina (Barker and Zublena, 1995). By increasing the efficient use of alternative nutrient sources, the nutritional needs of a crop can be met with fewer off-farm inputs, resulting in enhanced profitability (Edwards and Daniel, 1992). Because mineralization is required before plant-available N is released, the use of these waste products can provide a source of slow-release N in soils, which may limit groundwater contamination in soils most subject to leaching (Edwards and Daniel, 1992; Ndegwa et al., 1991).

The Columbia lance nematode, *Hoplolaimus columbus,* is limited in distribution to Georgia, North Carolina, South Carolina, and Alabama in the United States (Koenning et al., 2003a). In areas where this pathogen occurs, it can parasitize and damage cotton (*Gossypium hirsutum*), corn (*Zea mays*), and soybean (*Glycine max*), especially in sandy soils (Koenning et al., 2004). Rotation is not generally an option in fields infested with *H. columbus* because of its wide host range. A number of tactics for management of *H. columbus* in cotton have been investigated, including cultural practices, tolerant cultivars, and the application of poultry litter (Koenning et al., 2003a,b). Rates of poultry litter needed to suppress this nematode, however, may be greater than that required for plant nutrition and thus restrictive. Nematode management in cotton depends on nematicides, and nematicides are effective in preventing yield suppression by *H. columbus* and other plant-parasitic nematodes (Koenning et al., 2004). The use of nematicides, however, is increasingly under scrutiny by public and government agencies. Alternative tactics for management of the Columbia lance nematode are limited.

The addition of nitrogenous soil amendments results in the formation of ammonia, which has nematicidal properties (Rodríguez-Kábana, 1986). More recently, a range of organic amendments, including chicken litter or other animal wastes, have provided considerable protection for plants against plant-parasitic nematodes

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(Riegel et al., 1996; Rodríguez-Kábana et al., 1987). Animal manure impacts communities of soil microorganisms and may stimulate organisms antagonistic to nematodes (Opperman et al., 1993; Riegel and Noe, 2000; Rodríguez-Kábana et al., 1987; Sikora, 1992).

Several measures of nematode communities have been developed and related to ecosystem structure and function (Bongers, 1990; Bongers and Ferris, 1999; Bulluck et al., 2002; Ferris et al., 1996, 2001; Freckman and Ettema, 1993; Neher et al., 1995). The maturity index (MI) has been found useful in characterizing disturbance but is difficult to quantify because nematodes must be identified to the specific or at least the family level (Bongers, 1990; Bongers and Ferris, 1999). In contrast, the plant-parasite maturity index (PPI) is readily calculable by phytonematologists because plant-parasitic nematodes are often identified to the generic or specific level. Bongers and Ferris (1999) indicated that the PPI is generally inversely proportional to the MI. Similarly, greater diversity values are regarded as a measure of a less disturbed and potentially more stable environment. A trophic diversity index (TDI) can be calculated from data that quantify nematodes to the trophic level, which though more difficult to obtain than the PPI is certainly within the abilities of most phytonematologists. Simple ratios such as the bacterivore-to-fungivore ratio, or the bacterivore-to-plant parasite ratio, also have shown promise in measuring the activity of decomposers and nutrient cycling (Neher et al., 1995). Additions of poultry litter, and or other organic waste products, should be reflected in changes in these indices and (or) ratios. These measures may provide insight into mechanisms that impact soil health and sustainability.

The use of biologically active waste products for management of plant-parasitic nematodes is an attractive alternative to chemical nematicides. Application of poultry litter and other biological wastes to agricultural land has the potential to decrease certain pest problems and minimize air and water pollution by these materials (Koenning et al., 2003b; Sims and Wolf, 1994). The objectives of this research were to quantify the impact of the rate of application of poultry litter on the population changes of nematode communities and subsequent cotton yield.

Materials and Methods

Field experiments were conducted in 1994 in Hoke and Scotland counties, North Carolina, in fields that had been planted previously with cotton for several years. The soil type at the Hoke County location (site 1) was a Goldsboro sandy loam (67% sand, 27% silt, 6% clay, <1% organic matter), and the soil type at the Scotland County location (site 2) was a Marlborough sandy loam (67% sand, 20% silt, 13% clay, < 1% organic matter). Species of plant-parasitic nematodes typically identified at these sites included *Helicotylenchus dihystera, H. columbus, Paratrichodorus minor,* and *Pratylenchus brachyurus.* Of these plant-parasitic nematodes, only *H. columbus* is considered to be pathogenic to cotton. Although the *P. minor* and *P. brachyurus* are certainly parasitic on cotton, their pathogenicity to this crop is not well documented (Koenning et al., 2003b). The preplant population density of *H. columbus* at site 1 (preplant density 86/500 cm^3 soil) was below the damage threshold of 500/500 cm^3 soil for a fall sample, but the population density of this nematode at site 2 was 519/500 cm^3, which was above the damage threshold for this nematode in North Carolina (Anonymous 2004; Noe, 1993). The experimental design was a split-plot with main plots as rates of poultry litter applied at 0.0, 6.7, 13.4, and 20.1 tons ha, and sub-plots were treated or not treated with the growth regulator mepiquat chloride (Pix, BASF AG, Research Triangle Park, NC). Treatments with mepiquat chloride were included to evaluate the need for growth regulators to suppress excessive vegetative growth at high fertility levels. There were three options for treatment with mepiquat chloride—no treatment, treatment applied at early bloom at 0.025 l a.i./ha, or treatment at early bloom only if early-season growth were excessive according to recommendations of the North Carolina Cooperative Extension Service (Anonymous, 2004). There were six replications for each treatment combination. Plots were four rows, 6.1 m long, with 1.01-m row spacing and 3.0-m alleys.

Poultry litter was obtained from local sources, applied to selected plots, and incorporated with a disk 2 weeks prior to planting. The nutrient content of the litter was analyzed by the North Carolina Department of Agriculture and Consumer Services (Table 1). Plots

Table 1. Nutrient composition of poultry litter applied to plots at two locations in North Carolina in 1994.

	Nutrient composition[a]												
Location	DM (%)	N (%)	P (%)	K (%)	Ca (%)	Mg (%)	S (%)	Fe (ppm)	Mn (ppm)	Zn (ppm)	Cu (ppm)	B (ppm)	Na (ppm)
Scotland Co.	68.6	3.68	1.52	2.74	2.73	0.60	0.70	692	554	631	335	42.7	0.87
Hoke Co.	53.3	1.99	2.99	1.51	4.03	1.21	0.39	331	736	617	529	15.8	0.38

[a]Analysis provided by the Plant, Waste Solution Section Agronomic Division of the North Carolina Department of Agriculture and Consumer Services. DM = Percent dry matter.

that did not receive poultry litter were fertilized according to soil test recommendations for North Carolina and were approximately equivalent to the low litter rate. Plots receiving litter did not receive supplemental fertilizer. Cultivar Deltapine 5690 was planted in mid-May by the growers, and all plots received an in-furrow application of aldicarb (Temik 15G, Bayer Crop Science Inc., Research Triangle Park, NC) at 0.5 kg a.i./ha for early-season insect control. This rate of aldicarb is not generally considered to be nematicidal. Alleys between plots were cut by hand 2 weeks prior to harvest. Plots were harvested with a commercial cotton picker. Lint yield was determined by ginning sub-samples of seed cotton taken from the first two replications.

Samples for nematode assay of each plot were collected prior to litter application and at mid-season and cotton harvest. Quantification of all nematodes to trophic group and to species for plant-parasitic nematodes was done for pre-plant, mid-season, and harvest samplings (Pi, Pm, and Pf). Each soil sample consisted of 8 to 10 soil cores (2.5-cm-diam.) taken to a depth of 15 cm from the center two rows of each plot and composited. Soil samples were stored at 15 °C and processed within 3 weeks after the sample date. A 500-cm^3 sub-sample was processed by elutriation and centrifugation to extract nematodes from soil. Roots were collected from a sieve on the elutriator and placed in a mist extractor for 5 days to extract nematodes from roots and organic debris (Barker et al., 1986).

Data analysis consisted of analysis of variance (ANOVA) for a split-plot design. Orthogonal polynomial contrasts were used to quantify increasing amounts of litter applied, and contrasts were also used to compare typical fertility regimens (0.0 and 6.7 tons/ha litter rate) with a high fertility rate (13.4 and 20.1 tons/ha litter rate). The general linear models procedure (PROC GLM) was also used to evaluate the impact of rate of litter on nematode data. Repeated measures ANOVA (PROC GLM) was used to evaluate changes between sampling dates and to evaluate interactions of changes over time associated with litter application. PC/SAS software (SAS Institute, Cary, NC) was used to conduct all analyses. Because application of the growth regulator mepiquat chloride did not have an impact on nematode data or cotton lint yield, the means presented represent 18 observations. Nematode data were transformed using $\ln(x+1)$. Several ratios and indices of nematode communities were computed for analysis after transformation. The ratio of fungivores to bacterivores (FB ratio) was computed as fungivores ÷ (bacterivores + fungivores), and the ratio of bacterivores to plant parasites (BP ratio) was calculated as bactivores ÷ (bacterivores + plant parasites). The maturity index for plant parasites (PPI) was calculated as $PPI = \Sigma\ (v_i \times f_i)/n$, where v_i = the colonizer-persister (c-p) value assigned to family I, f_i = the frequency of family i in a sample, and n = total number of individuals in the sample (Bongers 1990; Neher et al., 1995). The TDI was estimated as Simpsons diversity index (Hill's N2) with $N2 = 1 \div (\Sigma[n_i \div N]^2)$, where n_i = number of individuals in trophic group (or family) i and N is the known total number of all individuals in the community.

Results

Poultry litter effects on numbers of nematodes and population changes over time: Plant-parasitic nematodes identified included *H. dihystera, H. columbus, P. minor,* and *P. brachyurus* at site 1 and *H. dihystera, H. columbus,* and *P. minor* at site 2. Poultry litter application rate had no effects on *H. columbus* numbers at site 1 at any sampling date (Fig. 1A,B). Midseason population densities of *H. columbus* at site 2 were related negatively ($r^2 = 0.056$, $P = 0.046$) to the application rate of litter. Contrasts of the 0.0 and 6.7 rates with the 13.4, and 20.1 tons/ha rates of poultry litter on Pm numbers indicated ($P = 0.088$) suppression of *H. columbus* numbers associated with high rates of litter application. Numbers of *H. columbus* were different between the two sites and also varied through time of sampling ($P \leq 0.0001$), with greatest numbers at harvest for site 2. A time × litter interaction was observed ($P = 0.01$) for site 2, but not for site 1.

There was a trend toward lower Pm densities of *P. minor* at both locations and for *P. brachyurus* at site 1 in response to the rate of litter applied, but this effect was not significant ($P \leq 0.10$). Poultry litter had no detectable effect on *H. dihystera* numbers at any time during these experiments (data not included). Numbers of total plant parasites did not differ between sites for Pi, but numbers of plant-parasitic nematodes were greater ($P = 0.0024$, $P \leq 0.0001$) at site 2 than at site 1 for Pm and Pf samplings (Fig. 1C,D). Poultry litter application rate had no effects on the density of total plant-parasitic nematode numbers at site 1 for any sampling date. Midseason population densities of plant parasites at site 2 were related negatively ($r^2 = 0.058$, $P = 0.43$) to the application rate of litter, and contrasts of the 0.0 and 6.7 rates with the 13.4, and 20.1 tons/ha rates of poultry litter on Pm numbers of this nematode showed reduced ($P = 0.095$) population densities associated with high rates of litter application. Total numbers of plant-parasitic nematodes were greatest at site 1 for the Pm samples, but greatest numbers of plant-parasitic nematodes occurred at the Pf sampling for site 2; these changes through time were significant ($P \leq 0.001$) for both sites.

The levels of bactivorous nematodes differed considerably between site 2 and site 1, with greater numbers of bacterivores at site 2 ($P \leq 0.001$) for both Pi and Pf sampling dates (Fig. 2A,B). Midseason numbers of bacterivores at site 1 increased linearly with increasing rates of litter application ($P = 0.001$, $r^2 = 0.33$), but

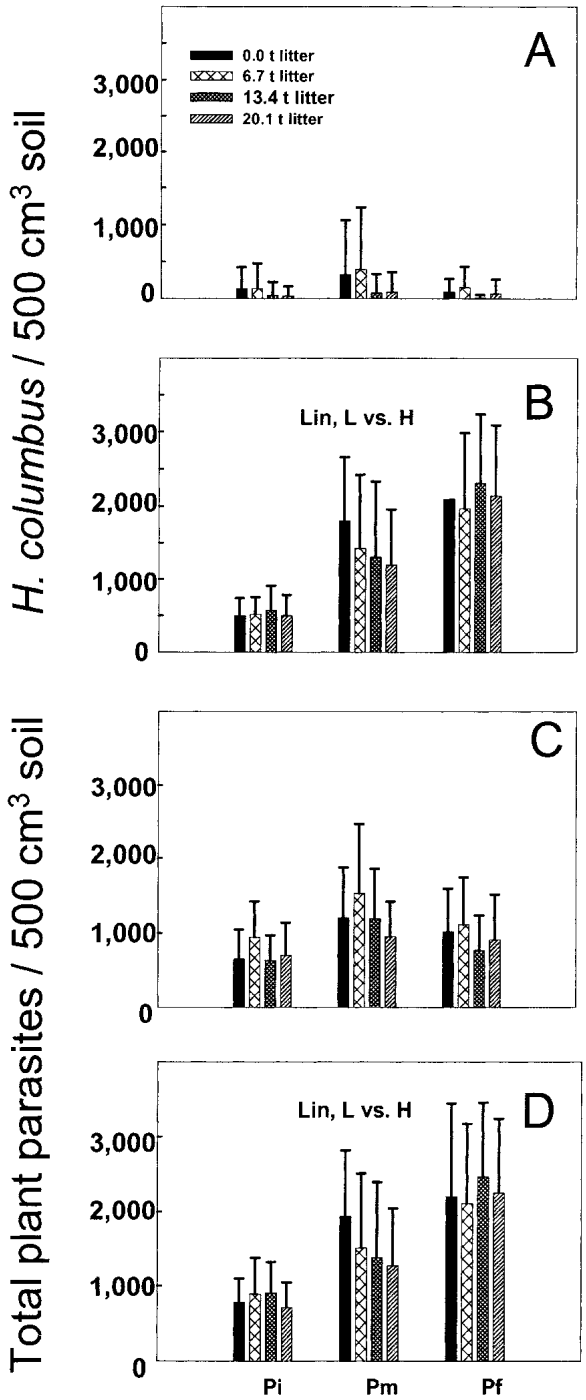

FIG. 1. Impact of pre-plant poultry litter incorporation at 0.0, 6.7, 13.4, and 20.1 tons/ha on numbers of *Hoplolaimus columbus* (A,B) and total numbers of plant parasites (C,D) per 500 cm^3 soil at two cotton production fields prior to planting (Pi), midseason (Pm), and at harvest (Pf) 1994. Site 1 is illustrated in A) and C), and Site 2 is illustrated in B) and D). Vertical lines are the standard deviation of the mean. Lin indicates a linear relationship ($P \leq 0.10$) to litter application rate, and L vs. H indicates a difference according to orthogonal contrasts ($P \leq 0.10$) of the 0.0 and 6.7 rates vs. 13.4, and 20.1 tons/ha rates of poultry litter.

bacterivore numbers at cotton harvest (Pf) were related negatively to the amount of litter applied ($r^2 = 0.045$, $P = 0.073$) at site 1. There was no impact of litter application on bacterivore population densities at any sampling date at site 2. Bacterivore numbers varied greatly through the season, with greatest densities at mid-season for site 1 compared with greatest densities at harvest for site 2.

Numbers of fungivores were greater ($P \leq 0.001$) at site 1 than 2 for Pm and Pf samples (Fig. 2C,D). Contrasts of the 0.0 and 6.7 rate with the 13.4 and 20.1 tons ha rate of poultry litter on Pm numbers of fungivores at both locations showed greater population densities at high rates of litter application ($P = 0.092$, $P = 0.098$ respectively). Final densities of fungivores were related positively ($r^2 = 0.14$, $P = 0.001$; $r^2 = 0.14$, $P = 0.002$) to the rate of litter application at both sites. Numbers of nematode fungivores differed between the two sites, and nematode numbers also varied through time of sampling ($P \leq 0.001$), with a linear increase in nematode numbers over time at site 1 but not at site 2.

Numbers of omnivores at three sampling dates were unaffected by the amount of litter applied at either site (Fig. 2E,F). The levels of omnivorous nematodes were different ($P = 0.01$) between sites, and nematode numbers also varied through time ($P = 0.001$), with greatest numbers at site 1. The greatest numbers of omnivores occurred at mid-season at site 1 compared to a density maximum at Pf for site 2. Predator numbers were greatest at site 1 for midseason samples (Fig. 2E). Litter had no measurable impact on numbers of predators at this site. Predators were not detected at site 2 (Fig. 3).

The influence of poultry litter application rate had no effects on total nematode numbers at site 2, but nematode densities at site 1 were related positively ($r^2 = 0.11$, $P = 0.005$) to the rate of litter applied (Fig. 2G,H). The final density of the total nematode community was related positively to the amount of litter applied for both locations ($r^2 = 0.14$, $P = 0.061$). The levels of all nematodes differed between locations for all sample times ($P \leq 0.001$), with greatest densities at site 1 for Pi and Pm samples but greatest abundance at site 2 for Pf. There was no litter × location interaction ($P > 0.10$), nor was there a time × litter interaction for either site. Total nematode numbers varied through time for both sites ($P \leq 0.001$).

Poultry litter effects and changes in ratios and indices over time: The BP ratio increased linearly ($r^2 = 0.28$, $P \leq 0.001$; $r^2 = 0.14$, $P = 0.059$) with increasing levels of poultry litter according to orthogonal polynomial contrasts for both Pm and Pf sample dates at site 1 (Fig. 4A). The rate at which poultry litter was applied effected an increase ($r^2 = 0.09$, $P = 0.008$) in the BP ratio for the Pm sampling only at site 2 (Fig. 4B). BP ratio values were greater ($P = 0.01$) at site 1 than at site 2 for all sampling dates. The BP ratios for each site changed ($P \leq 0.001$) through time, and the litter × time interaction was significant ($P = 0.05$) for both locations with lower BP ratios for Pf samples.

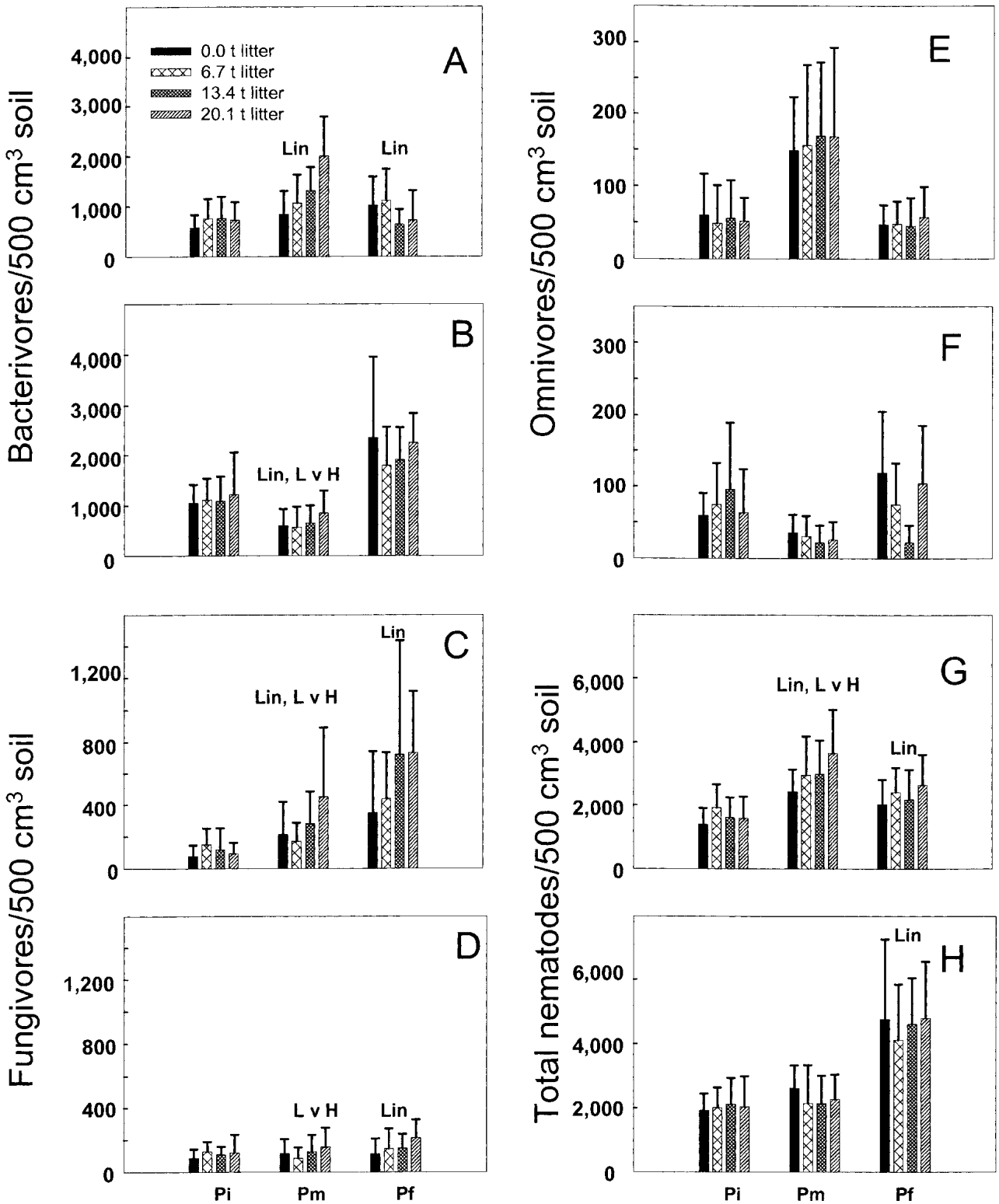

FIG. 2. Influence of the pre-plant application and incorporation of poultry litter at 0.0 (solid), 6.7 (coarse cross-hatch), 13.4 (fine cross-hatch), and 20.1 (diagonal) tons/ha on numbers of bacterivores (AB), fungivores (C,D), omnivorous (E,F), and total numbers of nematodes (G,H) per 500 cm^3 soil in a cotton production field quantified prior to planting (Pi), midseason (Pm), and at harvest (Pf) 1994. Site 1 (A,C,E,G) and Site 2 (B,D,F,H) are illustrated separately. Vertical lines represent the standard deviation of the mean. Lin indicates a linear relationship ($P \leq 0.10$) to litter application rate, and L vs. H indicates a difference according to orthogonal contrasts ($P \leq 0.10$) of the 0.0 and 6.7 rates vs. 13.4 and 20.1 tons/ha rates of poultry litter.

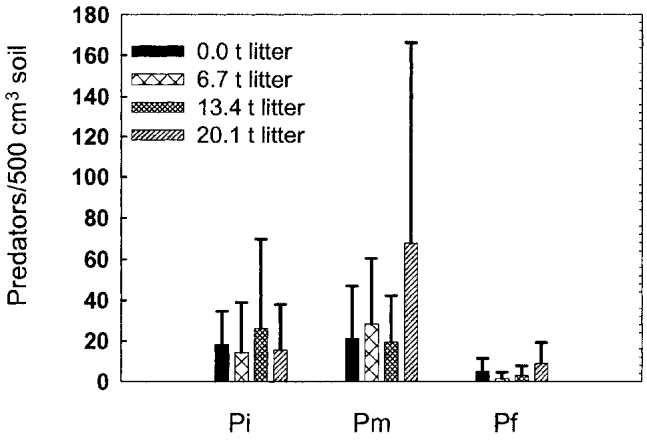

FIG. 3. Influence of the pre-plant application and incorporation of poultry litter at 0.0, 6.7, 13.4, and 20.1 tons/ha on numbers of predatory nematodes/500 cm^3 soil in a cotton production field quantified prior to planting (Pi), midseason (Pm), and at harvest (Pf) 1994. Vertical lines are the standard deviation of the mean.

The FB ratios were unaffected by litter application for Pm samples, but the Pf FB ratio increased linearly ($r^2 = 0.08$, $P = 0.02$; $r^2 = 0.15$, $P = 0.001$) with respect to the amount of litter applied at both locations (Fig. 4C,D). FB ratio values were similar between sites for Pi and Pm samples but were greater ($P = 0.001$) at site 1 than at site 2 for Pf samples. FB ratio values for each site varied ($P \leq 0.001$) through time, and a litter × time interaction was observed for both locations ($P = 0.05$) with greater FB ratio values at the Pf sample.

The TDI for the Pf sample increased linearly ($r^2 = 0.10$, $P = 0.008$) at site 1 with the amount of litter applied but was unaffected by litter application at either location for other sample times (Fig. 5A,B). The TDI was greater ($P \leq 0.03$) for all sample dates at site 1 than at site 2. The TDI for each site varied ($P \leq 0.0001$) through time with greater Pm indices at site 1 than at site 2. There was no location × litter interaction for any sampling, nor was the litter × time interaction significant for either location ($P > 0.10$).

Litter application decreased ($r^2 = 0.041$, $P = 0.059$) the PPI proportionally to the amount applied for the Pm sampling for site 2 but not at site 1 according to polynomial contrasts (Fig. 5C,D). There was no impact of litter application on either location for Pf samples on the PPI ($P \leq 0.10$). The PPI was greater ($P = 0.001$) at site 1 than at site 2 for the Pi sample date but not at other times. The PPI varied through time at site 2 with greatest PPI for Pm and Pf samples ($P \leq 0.001$). The PPI at site 1 did not differ among sampling dates. A litter × time interaction was observed ($P = 0.04$) for site 2 but not for site 1.

Influence of poultry litter application rates on cotton lint yield: Poultry litter applied at the greater rates increased ($P = 0.05$) cotton lint yield at both locations (Fig. 6). The optimum rate for litter application appears to be approximately 14.0 tons/ha. Cotton lint yields were

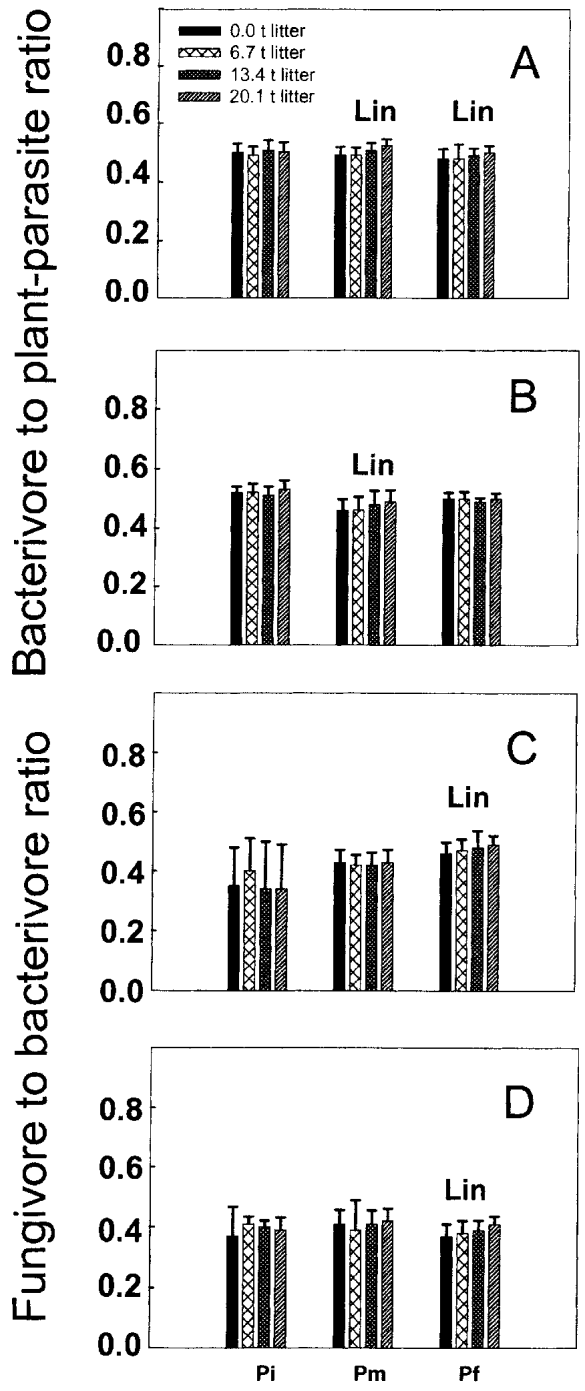

FIG. 4. Effects of pre-plant poultry litter incorporation at 0.0, 6.7, 13.4, and 20.1 tons/ha on the bacterivore-to-plant-parasite ratio (A,B) and the fungivore-to-bacterivore ratio (C,D) in cotton production fields quantified prior to planting (Pi), midseason (Pm), and at harvest (Pf) 1994. Site 1 (A,C) and Site 2 (B,D) are illustrated. Vertical lines are the standard deviation of the mean. Lin indicates a linear relationship ($P \leq 0.10$) to litter application rate.

lower at site 2 than at site 1 ($P = 0.0345$). Correlations of the various indices and ratios with cotton lint yield revealed only one consistent relationship across these two sites. The BP ratio at midseason was positively related to cotton lint yield in combined analysis ($r = 0.40$, $P \leq 0.001$).

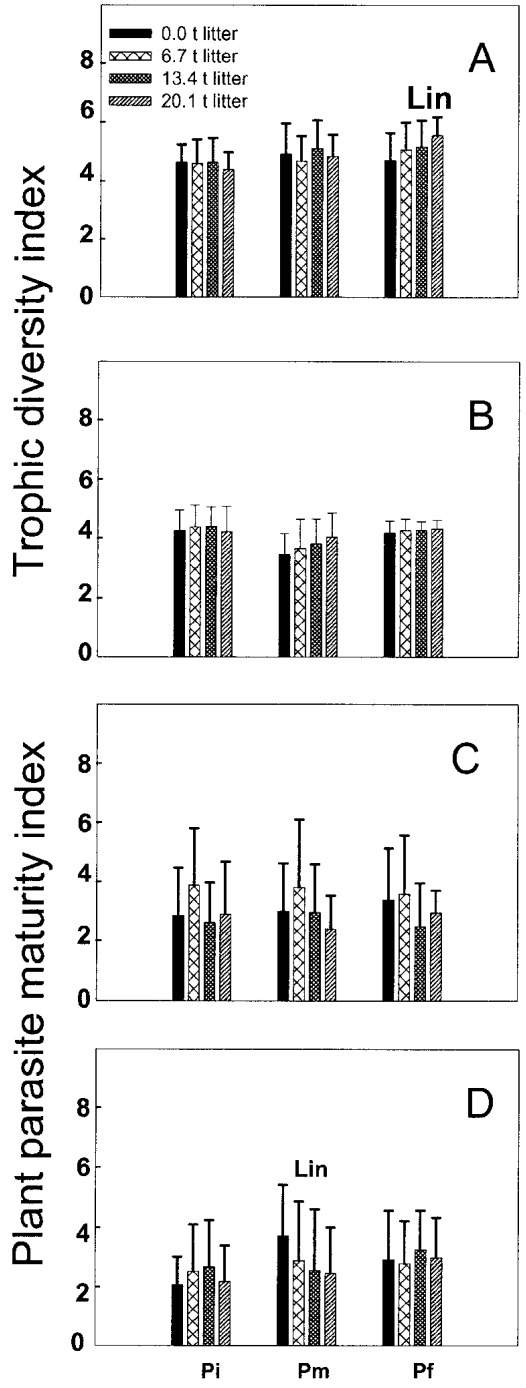

FIG. 5. Influence of the pre-plant application and incorporation of poultry litter at 0.0, 6.7, 13.4, and 20.1 tons/ha on the trophic diversity index (A,B) and plant-parasite maturity index (C,D) in cotton production fields quantified prior to planting (Pi), midseason (Pm), and at harvest (Pf) 1994. Site 1 (A,C) and Site 2 (B,D) are illustrated. Vertical lines are the standard deviation of the mean. Lin indicates a linear relationship ($P \leq 0.10$) to litter application rate.

DISCUSSION

The influence of poultry litter on primary consumers was reflected in the changes in population densities of *H. columbus* at midseason at site 2. Of the plant parasites detected in this study, only *H. columbus* is well documented for its pathogenicity to cotton (Koenning et al., 2004). The suppression of *H. columbus* numbers at midseason that occurred at site 1 associated with high rates of litter application has been observed previously (Koenning et al., 2003b). Suppression of *H. columbus* population densities at this site, however, did not persist to cotton harvest. The lack of impact on *H. columbus* at site 1 compared to site 2 can be attributed to the low population densities of this nematode at site 1 and because this nematode was absent or below detectable levels in many plots at this location.

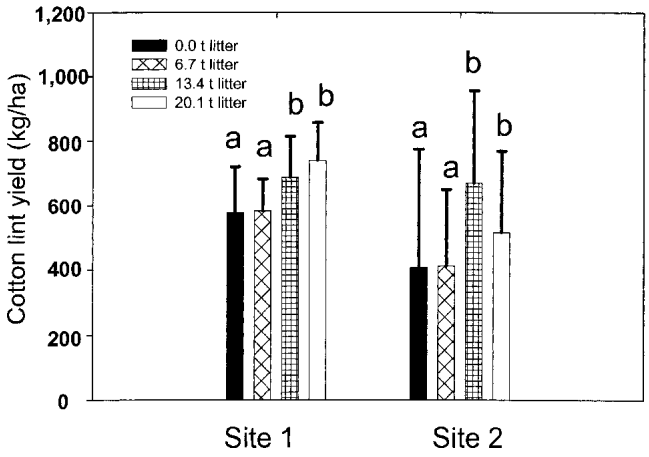

FIG. 6. Influence of pre-plant poultry litter incorporation at 0.0, 6.7, 13.4, and 20.1 tons/ha on cotton lint yield (kg/ha) at two locations in North Carolina during 1994. Means followed by the same letter do not differ according to orthogonal contrasts ($P \leq 0.10$). Vertical lines are the standard deviation of the mean.

Bongers et al. (1997) reported that the PPI was related positively to the level of nutrient enrichment that typically occurs in agroecosystems, and Freckman and Ettema (1993) reported the PPI useful in discriminating among systems representing a continuum of human intervention. The PPI was greater in organically managed soils than in conventionally managed soils in other research (Neher, 1999), but various soil amendments that increased the PPI one year did not influence PPI during the second year (Bulluck et al., 2002). Although root-knot nematode gall-index values were smaller in plots where organic amendments were applied to soil, numbers of this nematode were not influenced by these amendments. In yet another study, numbers of plant-parasitic nematodes tended to be greater in conventionally managed soils compared to organically managed soils (Ferris et al., 1996). In the current study, application of poultry litter at the high rates resulted in a lower PPI at midseason. The lower PPI at high rates of litter application at site 2 during midseason was likely due to the toxic effects of poultry litter on the dominant plant parasite (*H. columbus*) at this location. This result also suggests that the effects of ammonia and other compounds released by degradation of the poultry litter and (or) changes in biological activity in response to poultry litter may affect species of plant-parasitic nematodes differently. The lower PPI

values at site 2 compared to site 1 probably reflect the dominance of a plant parasite *H. columbus* on the community of plant-parasitic nematodes at site 2. Because *H. columbus* was above the damage threshold at site 2, but not at site 1, this nematode (a migratory endo- and ecto-parasite) probably limited root growth and thus substrate for other plant parasites. Also, *H. columbus* was in direct competition with *P. minor* (migratory ecto-parasite) and *H. dihystera* (a migratory endo- and ecto-parasite) at this location. The litter × time interaction for site 2 that did not occur at site 1 supports this conclusion.

Both primary consumers, bacterivores and fungivores, responded to application of poultry litter application, but the timing and type of response varied by group. The early increase of bacterivores at midseason in relation to poultry litter application and then decrease at cotton harvest is likely in response to enrichment of the soil environment with litter. The low C:N ratio of poultry litter promoted rapid bacterial degradation of this substrate and subsequent depletion of this organic resource for bacteria and their nematode consumers. Although some increase was noted in numbers of fungivores at the Pm sampling in response to litter application, the strong positive influence of poultry litter application on fungivore numbers at the harvest sampling for both locations is probably the result of poultry litter residue, associated fungi, and increased plant biomass from cotton roots. Poultry litter is a complex and heterogeneous substrate, as it consists not only of poultry excrement but also various cellulose-rich substances including wood shavings, fibers, and feathers that may require fungal decomposition (Ndegwa et al., 1991). Secondly, fungi—pathogenic, saprophytic, and symbiotic—readily colonize cotton roots and would increase through the growing season as root biomass increases. Thus the presence of large numbers of fungivores at cotton harvest is probably indicative of the presence of complex substrates with high C:N ratios that remain at the end of the growing system, requiring fungal activity for their decomposition (Ferris et al., 2001). The increase in FB ratio values over the course of the growing season and the interaction with litter application over time reflects the changes noted in the relative population densities of fungivores and bacterivores. Enrichment of the cotton-production environment by poultry litter, thus, had differing effects on the ratios depending on when samples are collected. The current research, however, demonstrates that the time of the year when samples are collected may affect the FB ratio. Porazinska and Coleman (1995) found no or only moderate influence of type of fertilizer (chicken litter vs. chemical fertilizer) on fungivores and bacterivores, in contrast to our research. The use of a gradient treatment design with increasing levels of chicken litter in the current study may allow for more precise quantification of this variable, or perhaps low levels of poultry litter may be insufficient to increase decomposition pathways.

Greater primary plant productivity and increased numbers of primary and secondary consumers that occurred as a result of poultry litter application at the higher application levels used in this research would be expected to increase numbers of nematode omnivores and predators in soil samples. Because the low numbers of these nematodes may have masked any effects, a longer study period may be needed, or increased resolution of any changes in taxa that may have occurred may be required to detect differences. Other researchers have noted that the low population densities of nematodes in these trophic groups make detection of treatment effects difficult (Freckman and Ettema, 1993; Neher et al., 1995). Also, there may be a time lag in the increase in numbers of predators and omnivores in response to increased productivity that was not encompassed by the time for this study. Neher et al. (1995) indicated that the preferred method for extraction of nematodes was the Cobb sieving method followed by centrifugation because this method was more efficient at recovering large nematodes, in particular, omnivores and predators. Since elutriation and centrifugation were used in combination with a modified Seinhorst mist chamber in the current research, detection of these nematodes may have been limited. However, the use of the mist extractor, to some extent, reduces the lack of detection for larger nematodes that may become caught in sieves used to remove organic debris from the sample because this portion is placed in the mist chamber.

The greater BP ratio at site 1 compared to site 2 may be related to the damaging levels of *H. columbus* that limited cotton development at this site. Supporting evidence for this hypothesis is the relationship between increasing amounts of poultry litter with increasing BP ratio that occurred at site 2 for Pm samples. This ratio is considered indicative of decomposition activity relative to reduced primary productivity (Neher et al., 1995).

The TDI presented in this study are comparable with those presented in other research (Freckman and Ettema, 1993; McSorley and Frederick, 1999; Neher and Campbell, 1994; Poranzinska and Coleman, 1995). Greater trophic diversity values indicate greater evenness and greater numbers of the trophic groups. The higher trophic diversity index values for site 1 compared to site 2 were expected because predators were absent at site 2. Predator numbers, however, were low at site 1 and would contribute relatively little to this index. The relatively low trophic diversity at site 2 was also due to the dominance of plant-parasitic nematodes at this location for Pi and Pm samples. The increase of the trophic diversity index associated with the addition of poultry litter for Pf samples at site 1 is related to the

reduction in bacterivorous nematodes population densities and a corresponding increase in fungivores.

The optimum rate of litter application appears to be approximately 14.0 tons/ha for cotton lint yield and suppression of Columbia lance nematode. Rates of litter application in excess of 20 tons/ha may actually suppress cotton lint yield, and these estimates agree with other research (Koenning et al., 2003b). The positive correlation between cotton lint yield and the BP ratio at midseason is the result of the suppression of numbers of plant-parasitic nematodes related to the rate at which poultry litter was applied and the increase in bacterial-feeding nematodes associated with litter application. Unfortunately, the value of this index as a predictive tool would be limited because it is a midseason measure. Other indices or ratios tended to have no statistical effect on cotton lint yield or were contradictory when comparing sites. The use of these indices, however, might provide useful information if applied to a larger number of fields and situations.

The current research benefited from the fact that there were a large number of replications for each treatment effect because application of mepiquat chloride had no impact on the nematode communities or cotton yield. Detection of significant treatment differences as affected by poultry litter applications was possible only because of the large number of replications and the use of a gradient treatment experimental design. The use of more sensitive measures of nematode community diversity in this research may have improved resolution of interactions with crop productivity, soil health, and nematode community structure. For example, identification of nematodes in other trophic groups at the specific or family level would permit the use of an enrichment/structural trajectory plot that may provide additional insight into changes in food web structure (Ferris et al., 2001). However, identification of fungivores, bacterivores, omnivores, and predators to the generic or specific level is time consuming. Systems that rely on automation for identification and (or) molecular tools to aid in such research are needed (van der Knapp et al., 1993)

Consideration of the entire nematode community in agricultural and other soils has been considered as part of a national program to monitor soil health (Neher and Campbell, 1994; Neher et al., 1995), and numerous other studies have been completed evaluating various measures of soil health and ecosystem function. A second important facet of this research is its application in the development and delivery of biological control of soilborne plant pathogens. A better understanding of the soil ecosystem is essential to the development of intensive biologically based systems to better manage bacterial, fungal, and nematode pathogens (Barker and Koenning, 1998).

Studies of nematode communities in cotton ecosystems are important because this intensively managed crop is grown annually on approximately 5 million hectares in the United States and annual cotton yield suppression due to nematodes is estimated at 4% (Koenning et al., 2004). Changes in agricultural practices will impact soil health and the sustainability of agroecosystems, especially in view of current government initiatives to reduce the use of nonrenewable resources. The adoption of practices that rely on nutrient cycling in the soil environment requires a better understanding of soil ecosystem structure, including the nematode community. Also, the potential for global climate change may accelerate the implementation of programs that encourage the sequestration of carbon in soil through the adoption of practices that increase soil organic matter, such as the use of manure, cover crops, and no-till farming practices. Because of the ubiquitous distribution of nematodes in soil ecosystems and their role in nutrient and carbon cycling, additional research on the various agroecosystems is warranted.

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