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Potential of an Alkaline-Stabilized Biosolid to Manage Nematodes: Case Studies on Soybean Cyst and Root-Knot Nematodes

The use of treated biosolids for pest management and soil nutrient augmentation is not a new practice, but it has increased in the last two decades, primarily in the United States (22). In the late 1970s, the first land application regulations were formulated by the U.S. Environmental Protection Agency (USEPA) in response to the Clean Water Act (44). Land application of sewage sludge for soil amendment and land reclamation has increased over time as a result of the ban on ocean dumping of wastewater residuals (Ocean Disposal Ban Act of 1988). The Act also minimizes other disposal options, such as land-filling or incineration. In 1993, the Standards for the Use or Disposal of Sewage Sludge (Code of Federal Regulations Title 40, Part 503) was created (45,46). Part 503 (as it is commonly called) set pollutant limits, operational standards for human/animal pathogen and vector-attraction reduction, management practices, and other provisions intended to protect public health and the environment from any reasonably anticipated adverse effects from chemical pollutants and pathogenic organisms. In 1995, the EPA promoted the terminology "biosolids" rather than "sewage sludge" and defined biosolids as "the primarily organic solid product yielded by municipal wastewater treatment processes that can be beneficially recycled as soil amendments and meets the standards of Part 503". Although the term is sometimes controversial (33), we will use biosolid in reference to the product tested in this research.

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In 1998, approximately 6.9 million dry tons of biosolids were disposed of in the United States, of which 41% was used for land application (Table 1) (47) on less than 1% of the nation's agricultural land. A state-by-state summary of biosolid production and use for 2000 is available (11). Biosolid production is expected to increase to 8.2 million dry tons in 2010—an approximately 20% increase in just over 10 years (47). An increase in the proportion that is land-applied is also expected (Table 1).

Plant-parasitic nematodes cause nearly $10 billion in crop losses per year in the United States and about $100 billion globally (17,37). Nematodes are responsible for reductions in yield and quality over a broad range of hosts. Strategies used to control plant-parasitic nematodes include the use of nematicides, resistant varieties, nonhost rotations, cover crops, cultural practices, and biological control. Because approximately 48% of biosolids will be land-applied in 2010 (Table 1), the effects of such application, alone or in combination with other management practices, required study. In 2001, the U.S. Congress mandated that research be conducted on an alkaline-stabilized biosolid product (N-Viro Soil [NVS]) and the potential for this product to control plant-parasitic nematodes. The ensuing collaborative research effort included federal (U.S. Department of Agriculture–Agricultural Research Service), university (Iowa State University, Ohio State University, Michigan State University, North Carolina State University, and University of Florida), and private (N-Viro International, Toledo, OH) organizations. Numerous laboratory, greenhouse, and field experiments were conducted. In this paper, we summarize these studies and discuss the opportunities and challenges presented by using alkaline-stabilized biosolids for plant-parasitic nematode management.

Past Experiences with Biosolids for Nematode Management

Decreases in population densities of plant-parasitic nematodes by raw and composted sewage sludge were demonstrated in the 1970s (12). Raw sewage sludge reduced *Meloidogyne incognita* (the southern root-knot nematode) juvenile penetration, galling, and egg production on tomato roots in greenhouse pots (7). Although soil amended with heat-treated sewage sludge, alone and combined with yard waste, had few consistent effects on plant-parasitic nematodes, it reduced final *M. incognita* population densities on squash (23). Amendment of field soil with dried, pelletized biosolids increased *M. incognita* population densities and root galling on tomato compared with the nontreated control (24). More recently (2), composted sewage sludge suppressed *Meloidogyne javanica* (the Javanese root-knot nematode) on tomato, when applied at very high rates (greater than 75% wt/wt). Although results were inconsistent among studies, they indicated a potential for biosolids as a plant-parasitic nematode management tactic.

Prior to initiation of the research effort discussed herein, an alkaline-stabilized biosolid (NVS) applied for nutrient management purposes was observed to reduce population densities of *Heterodera glycines* (the soybean cyst nematode) in Ohio (T. Logan, *unpublished data*). The reduction was sustained for 3 years in some fields and enabled the farmer to insert soybeans into a crop rotation cycle 2 to 3 years earlier than in a nonhost crop rotation cycle. Similar results were reported from Canada (49), where 2 to 20 dry t/ha NVS suppressed *H. glycines*.

The N-Viro Biosolids Alkaline Stabilization Process

Since the advent of advanced wastewater treatment in the late 1960s in the United States, much attention has been given to treatment and disposal or utilization of the residual organics. Prior to the 1980s, most biosolids were biologically digested to stabilize the residual organics and to kill some pathogens. In the 1980s, more advanced technologies for biosolids treatment emerged to produce a pathogen-free product and stabilize residual organics. The two most widely used approaches are biological composting and alkaline stabilization (47). Whereas composting relies on biological degradation, heat, and drying to kill pathogens and stabilize organic matter, alkaline stabilization involves a combination of high pH, heat, and drying to achieve the same purpose. Two classes of biosolids can be produced, classes A and B. Class A biosolids contain no detectable levels of pathogens, meet strict vector attraction reduction requirements, and have low levels of metals. Class B biosolids are treated but still contain detectable levels of pathogens. Whereas class A biosolids can be applied without restrictions, class B biosolids incur land buffer requirements and crop harvesting restrictions (45).

One alkaline stabilization approach that yields a class A product is the N-Viro process (Fig. 1). The basis of the N-Viro process (6,21) is to destroy pathogens through a combination of the following stresses: (i) alkaline pH; (ii) accelerated drying; (iii) high temperature; (iv) high ammonia (NH_3); (v) salts; and (vi) indigenous microflora. These stresses are produced in the biosolid when alkaline admixtures (AA) with varying unique properties are added. A number of industrial waste alkaline materials are used alone or together as the AA in the N-Viro process. These include cement kiln dust (CKD), lime kiln dust (LKD), wood ash, various coal combustion ashes that include flue gas desulfurization byproduct (FGD), or alkaline fly ash. If the AA contains enough free lime (calcium oxide [CaO], calcium hydroxide [$Ca(OH)_2$], or other strong alkali) to give a pH rise to greater than 12.0 and an exothermic reaction necessary to achieve desired temperatures (52 to 62°C), no other additive is needed. Calcium oxide is added to supplement the free lime content of the AA if it is not "hot" enough.

Raw primary biosolids, either waste-activated or digested, with solids content of 18 to 40% can be treated (Fig. 1). Biosolids and AA are mixed with a pug mill or screw blender. The ratio of AA to biosolids varies with the solids content, with a higher ratio used for those biosolids with lower solids content. In general, the AA dose ranges from 25 to 50% of the dewatered biosolids wet weight. With proper mixing speeds, the resultant product is a granular, easy-to-handle soil-like material that is further processed by air drying or heating.

A similar N-Viro process, which differs in that it does not use heat, can be employed to treat animal manures. Alkaline

Table 1. Past and projected biosolid use and disposal in the United States (47)

		Beneficial use				Disposal			
Year	Biosolid generation (dry tons)	Land application	Advanced treatment	Other beneficial use	Total	Surface disposal/ landfill	Incineration	Other	Total
1998	6.9	41%	12%	7%	60%	17%	22%	1%	40%
2000	7.1	43%	12.5%	7.5%	63%	14%	22%	1%	37%
2005	7.6	45%	13%	8%	66%	13%	20%	1%	34%
2010	8.2	48%	13.5%	8.5%	70%	10%	19%	1%	30%

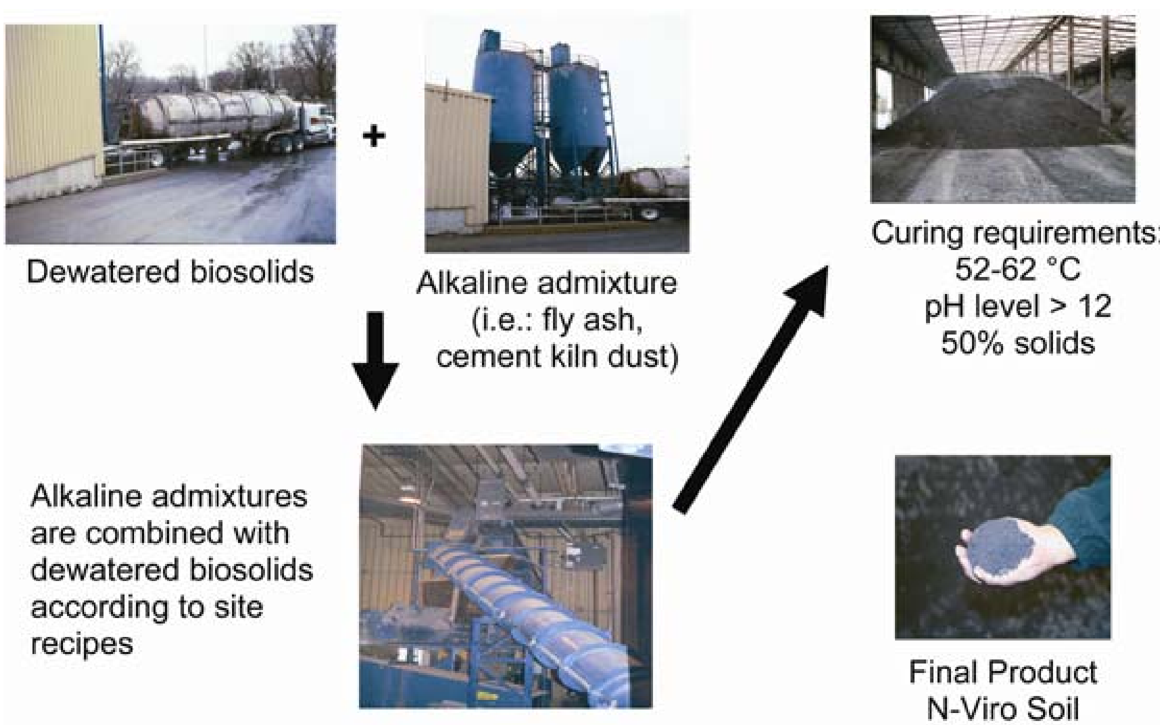

Fig. 1. The N-Viro alkaline stabilization process (photos courtesy of David Chitwood, USDA-ARS, with permission from N-Viro International, Toledo, OH).

Table 2. Typical chemical characteristics of N-Viro Soil produced in Toledo, OH (N-Viro International) expressed on a percent dry weight or mg/kg basis[a]

Chemical properties	
pH	12.2
Nitrogen (%)	1
Phosphorus (%)	0.2 – 1.1
Potassium (%)	1
Sulfur (%)	5
Calcium (%)	10 – 40
Magnesium (%)	1
Sodium (%)	<0.2
Arsenic (mg/kg)	27.4
Cadmium (mg/kg)	<1.4
Chromium (mg/kg)	65.4
Copper (mg/kg)	74.0
Lead (mg/kg)	28.4
Mercury (mg/kg)	<0.7
Molybdenum (mg/kg)	9.2
Nickel (mg/kg)	61.1
Selenium (mg/kg)	8.5
Zinc (mg/kg)	188

[a] Elemental composition and characteristics vary depending upon manufacturing location.

materials are added to raise pH above 10.0. Disinfection of the manure is achieved primarily by the high levels of gaseous NH_3 contributed by the manure at this pH.

Properties of N-Viro Soil

The strong alkalinity in NVS is responsible for its high pH (Table 2), but the pH of NVS when added to soil is very different than that of the material itself. When NVS is added to soil, an initial neutralization reaction (Fig. 2A) occurs quickly due to the high reactivity of the $Ca(OH)_2$ component compared with the $CaCO_3$ component. This reaction is very rapid and is followed by acid neutralization which occurs by calcite dissolution (Fig. 2B); this is the reaction that occurs in soil when limestone is added. It is important to understand the balance between these two reactions when applying NVS for plant-parasitic nematode suppression.

The typical chemical characteristics of NVS are given in Table 2. The chemistry of NVS is dominated by calcium (Ca); about 20% of the Ca in NVS is water-soluble. Soluble salts are high in NVS as they are in biosolid composts or heat-dried biosolids, because these processes involve the evaporation of water and concentration of salts from the biosolids. The total carbon content of NVS is about half that of digested biosolids, the difference due to the added mineral AA. The nitrogen (N) in NVS exists almost entirely as organic N because the high pH drives off free ammonia (NH_3) from the biosolids. All of the phosphorus is contributed by the biosolids and probably exists in NVS as a combination of organic phosphorus (P) and calcium phosphates. Potassium content of NVS varies widely, and is contributed by CKD, which can contain as much as 5 to 7% K. Most of the sulfur in NVS is in the form of gypsum from the fly ash in the AA; a lesser amount is contributed by sulfur-containing proteins in biosolids. Trace element contents of NVS are low compared with that of U.S. biosolids (Table 2).

$$\text{A.}\quad Ca(OH)_2 + 2H^+ = Ca^{2+} + 2H_2O$$

$$\text{B.}\quad CaCO_3 + H^+ = Ca^{2+} + HCO_3^-$$

$$\text{C.}\quad NH_4^+ + OH^- \xleftrightarrow{pK_a = 9.25} NH_3 + H_2O$$

Fig. 2. Important chemical reactions to understand when utilizing alkaline stabilized biosolids to control plant-parasitic nematodes. A, initial acid neutralization, B, calcite dissolution, and C, Henderson-Hasselbalch equation.

Traditional Uses of N-Viro Soil

N-Viro Soils are currently produced in more than 30 locations in the United States, United Kingdom, Israel, and Brazil (www.nviro.com). Existing markets or markets undergoing developmental research include those for agricultural limestone substitutes/low analysis fertilizers, land reclamation, soil amendments/urban soils, soil blend ingredients, and landfill cover materials. Agricultural limestone substitute/low analysis fertilizer is the most developed NVS market, particularly in Ohio, New York, Florida, Indiana, and North Carolina. As a limestone substitute, NVS is typically applied at a rate of 10 t/ha dry weight every 2 to 4 years, depending on location. The objective of the research presented here was to explore another possible market for the utilization of NVS, thereby increasing the value-added benefit of the product. A summary of our research efforts in Florida, Iowa, Maryland, Michigan, and North Carolina is compiled in Table 3.

Effects of N-Viro Soil on Soybean Cyst Nematode

H. glycines, the soybean cyst nematode (Fig. 3A and B), causes greater yield reduction in soybean (*Glycine max*) than any other pathogen or pest in the United States (51). It is widely spread throughout the soybean growing regions of the United States and elsewhere, causing yield losses of up to 50%. Current management practices include crop rotation with nonhosts and resistant soybean cultivars. However, the sources of resistance to *H. glycines* are limited, and overuse of resistant soybean cultivars may impose a strong selection pressure on the nematode population, resulting in the resistant cultivars' becoming ineffective (15,29). Alternative management options have been suggested to supplement the use of resistant soybean cultivars, one of which is the application of soil amendments such as NVS. NVS was evaluated in field, microplot, and greenhouse experiments in Iowa, Michigan, and North Carolina for *H. glycines* management.

Research at Iowa State University (G. L. Tylka and F. Avendano, *unpublished data*) examined the effect of NVS on *H. glycines* populations and on soybean yield under field conditions (Table 3). Experiments were conducted at different locations in central Iowa from 2002 to 2006; the soil types of experimental plots were either a Hanlon fine sandy loam or a Harps loam. Soybean cultivars, resistant or susceptible to *H. glycines*, were planted in replicated plots. Treatments were fallow, NVS at 6 and 17 dry t/ha, and a nontreated control. In 2005 and 2006, two application methods, broadcast and banded (Fig. 4A and B), were also tested. NVS was incorporated into the soil immediately after application in all cases. Data collected each year consisted of *H. glycines* egg population densities at planting and at harvest, soil pH at planting and at harvest, and soybean yield. In addition, soil pH and *H. glycines* population density were measured 30 days after application of NVS in 2005, and soil pH was measured in soil samples collected 2 days after application in 2006. Results were fairly consistent from year to year; there were no statistically significant differences in *H. glycines* population density, soybean yield, or changes in soil pH across treatments. Soil pH after NVS amendment varied from 6.5 to 7.5.

In North Carolina (16), field experiments were conducted during 2003 and 2004 on a Woodington sand (87% sand, 11% silt, 2% clay, <1% organic matter) naturally infested with *H. glycines* (Table 3). NVS at rates of 0, 7, 13, and 20 dry t/ha was broadcast-applied, and soybean was planted immediately after application. The experiment was a split-plot design with main plots of either *H. glycines*–resistant or –susceptible cultivars and split plots of the four rates of NVS. Data collected consisted of preplant soil samples to determine population densities of juveniles, midseason plant samples for cyst population

Table 3. Summary of results of experiments evaluating N-Viro Soil (NVS) for plant-parasitic nematode management conducted in multiple states from 2002 to 2006

Location	N-Viro Soil (dry t/ha)	Nematode tested	Experiment type	Application method	Soil	Description of response
IA	6 and 17	*Heterodera glycines*	Field	Incorporated	Sandy loam to loam	No reduction in *H. glycines* population densities at either rate.
NC	7, 13, 20	*H. glycines*	Field	Broadcast	Sand (87%)	Nonsignificant increase in *H. glycines* population densities and yield increase in *H. glycines*–resistant cultivars with increasing rates of NVS.
NC	7, 13, 20	*H. glycines*	Microplot	Broadcast	Sand (91%)	Increase in *H. glycines* population densities with increasing rates of NVS.
NC	13, 27, 40	*H. glycines*	Field	Incorporated	Sand (91%)	Nonsignificant decrease in *H. glycines* population densities with increasing rates of NVS. Increase in *H. glycines*–resistant soybean with increasing rates of NVS
MI	10 and 40	*H. glycines*	Greenhouse	Incorporated	Sandy loam	Numbers of preadult stages and cysts generally decreased with increasing rates of NVS.
NC	7, 13, 20	*Meloidogyne incognita*	Microplot	Broadcast	Sand (91%)	Reduction in midseason *M. incognita* juvenile population densities with increasing rates of NVS; this reduction was not sustained to the end of the growing season. Higher rates did result in an increase in cotton yields.
MD	25, 50, 75, 100	*M. incognita*	Microplot	Incorporated	Loamy sand	A 1-year decrease in *M. incognita* population densities at NVS rates >75 t/ha. This reduction was not sustained during subsequent years.
MI	10 and 40	*M. hapla*	Greenhouse	Incorporated	Sandy loam	Both rates reduced population densities of *M. hapla*, with the higher NVS rate being more effective.
FL	5, 10, 15	*M. incognita*	Microplot	Incorporated	Loamy sand	Population densities were suppressed by NVS at 36 days; however, this effect disappeared by 67 days.

densities, soil and plant samples at harvest to determine juvenile and egg population densities, and soybean yield. The number of cysts per root system was lower ($P < 0.5$) on the *H. glycines*–resistant cultivar than the *H. glycines*–susceptible cultivar, and NVS had no impact on these data. Harvest *H. glycines* juvenile and egg population densities were numerically higher following NVS treatment. Yield of the *H. glycines*–resistant cultivar increased linearly with regard to NVS application rate ($P < 0.05$). Similar trends were observed during both years. A microplot experiment to evaluate NVS for *H. glycines* suppression was also conducted in North Carolina (*H. glycines*–resistant and –susceptible cultivars combined with the same NVS rates broadcast-applied) (16) (Table 3). The soil type was a Fuquay sand (91% sand, 6% silt, 3% clay, <1% organic matter). Preplant, midseason, and harvest soil samples were collected to determine *H. glycines* juvenile and cyst population densities. NVS had no apparent impact on *H. glycines* midseason population densities, and in fact there was an increase in harvest *H. glycines* population densities related to NVS application.

In 2005, higher rates of NVS (13, 27, or 40 dry t/ha) were incorporated into a Norfolk sandy loam naturally infested with *H. glycines* in North Carolina (S. R. Koenning, *unpublished data*) (Table 3). In a split-plot design, *H. glycines*–resistant and –susceptible soybean cultivars were planted as main plots and NVS rates were subplots. Additional treatments included: aldicarb (Temik 15G at 1.18 kg a.i./ha) applied in the furrow and a no NVS control. *H. glycines* population densities were monitored during the season, and soybean yield was determined. Whereas there were no significant differences ($P > 0.05$) in *H. glycines* population densities among treatments, there was a consistent nonsignificant decrease in *H. glycines* harvest egg population densities with increasing rates of NVS regardless of cultivar. The highest

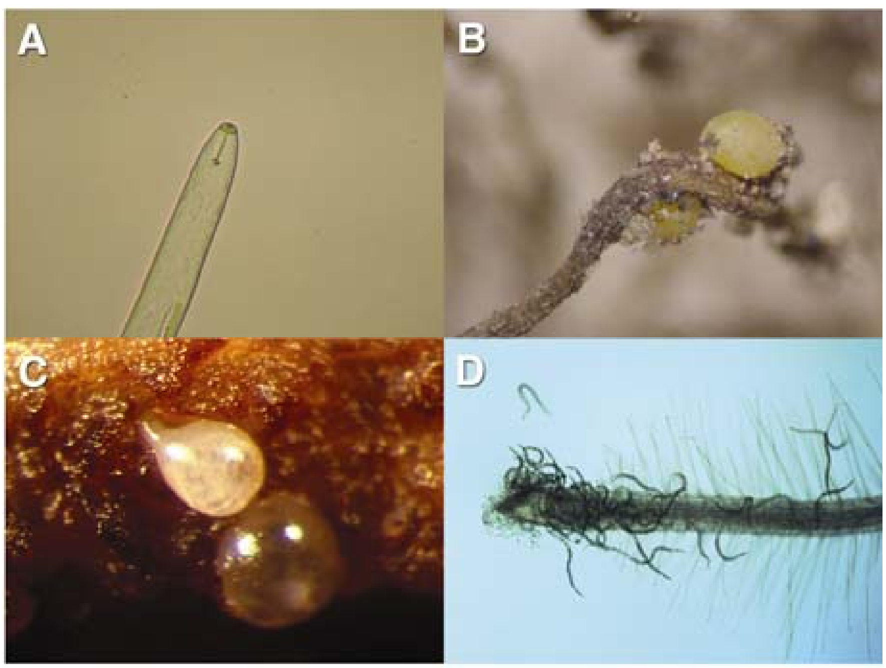

Fig. 3. Plant-parasitic nematodes cause nearly $10 billion in crop losses per year in the United States (37). A, *Heterodera glycines* juvenile; B, *H. glycines* cysts containing hundreds of nematode eggs; C, *Meloidogyne* sp. female; and D, *Meloidogyne* sp. juveniles surrounding a root tip (photos of *Meloidogyne* sp. with permission from MacTode, Blacksburg, VA).

rate of NVS tested (40 dry t/ha) resulted in an increase in *H. glycines*–susceptible soybean yield compared with those measured at the lower NVS rates, and was not different from yield in the aldicarb-treated plots.

In Michigan, the effects of 0, 0.5, or 2 dry g of NVS per 100 cm^3 (equivalent to 10 and 40 dry t/ha) on three *H. glycines* populations, GN 1, GN 2, and GN 3, classified as HG type 2, HG type 1.2, and HG type 0, respectively (30), were examined in three greenhouse experiments with glyphosate-tolerant soybean (DSR-221) (26,27) (Table 3). Whereas the responses of the three *H. glycines* populations to NVS treatment varied by experiment, the overall numbers of preadult stages and cysts generally decreased with increasing levels of NVS in all experiments.

Effects of N-Viro Soil on Root-Knot Nematodes

Root-knot nematodes, *Meloidogyne* spp. (Fig. 3C and D), were reported as the most destructive nematodes in a global survey (37). *Meloidogyne* spp. are known parasites of more than 3,000 host plants, and individual species often have wide host ranges. Unlike the situation with *H. glycines*, plant resistance to *M. incognita* is not widespread. The situation is similar for *M. hapla* (the northern root-knot nematode), a major pest of vegetables in temperate climates. In addition to a lack of usable resistance, nematicides traditionally used to control this group of nematodes are either being phased out or have already been banned (43).

Microplot experiments were conducted with *M. incognita* on cotton during 2003 and 2004 in North Carolina (S. R. Koenning, *unpublished data*) (Table 3). Microplots contained a Fuquay sand previously infested with *M. incognita*. Preplant juvenile population densities were determined prior to NVS application. Four rates of NVS (0, 7, 13, and 20 dry t/ha) were broadcast on the soil surface. Midseason and harvest *M. incognita* juvenile population densities were determined, as was cotton yield. NVS effected a linear reduction ($P < 0.05$) in midseason *M. incognita* juvenile population densities, but this reduction was not sustained; harvest population densities were unaffected. There was an increase ($P < 0.05$) in seed cotton yields with increasing rates of NVS during both years.

Microplot experiments in Maryland evaluated the effect of increasing rates of NVS (0, 25, 50, 75, and 100 dry t/ha) on *M. incognita* in soybean (53) (Table 3). The soil type was a Norfolk A loamy sand (87% sand, 8% silt, 5% clay, <1% organic matter). NVS was incorporated to a depth of 15 cm, and *M. incognita* population densities (juveniles and eggs) and soil pH were monitored over a 3-year period. During year 1, increasing rates of NVS resulted in greater soil solution pH and degrees of *M. incognita* J2 and egg suppression (Fig. 5). While soil solution

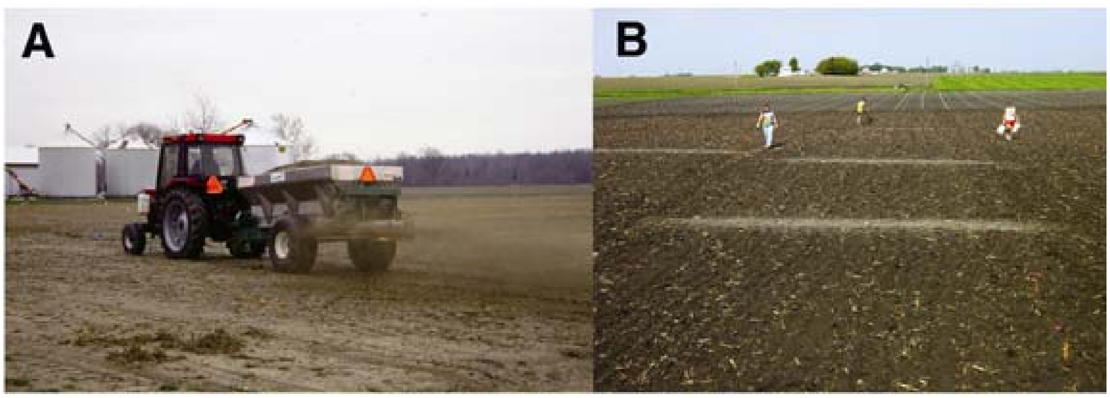

Fig. 4. A, Mechanical application of N-Viro Soil (NVS) in Ontario, Canada (photo courtesy of Tom Welacky, Agriculture and Agri-Food Canada), and B, banded application of 5 dry t/ha NVS to field plots in Iowa.

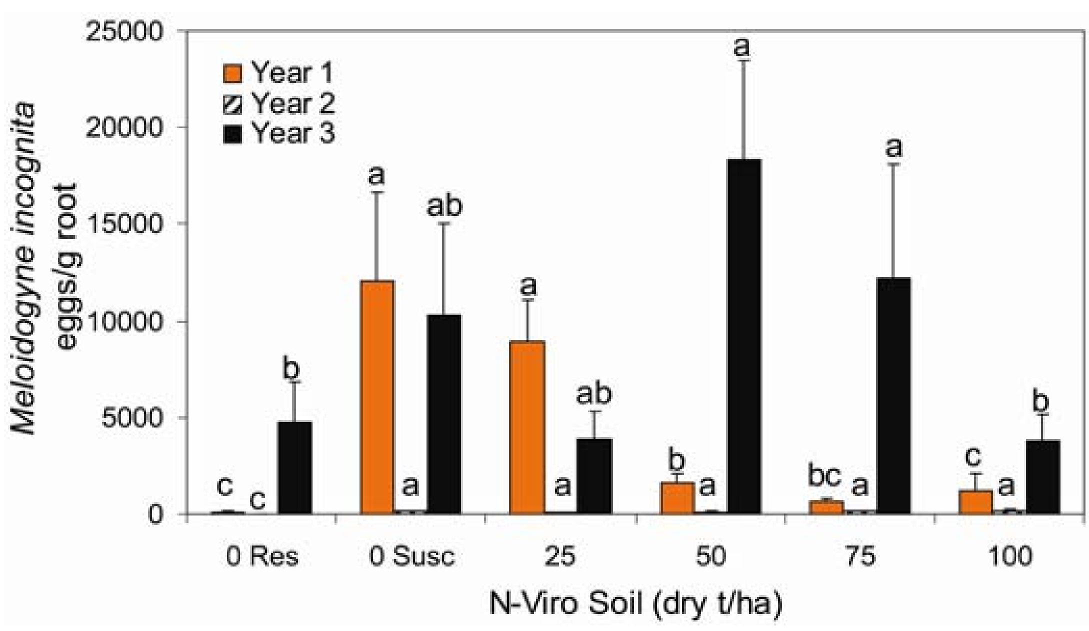

Fig. 5. *Meloidogyne incognita* egg population densities after incorporation of N-Viro Soil at different rates over time (with permission from Brill Academic Publishers, Boston, MA) (53). Nonamended resistant (res) and susceptible (susc) cultivar controls were included. Bars within years with the same letter are not significantly different according to Tukey's HSD test ($P \leq 0.001$).

pH remained higher in NVS-amended plots during years 2 and 3, *M. incognita* juvenile and egg population densities were not consistently lower at the higher NVS rates.

Microplot experiments in Florida evaluated the effect of various rates of NVS (0, 5, 10, and 15 dry t/ha) on *M. incognita* in tomato (1) (Table 3). The soil type was a Krome very gravelly loam with a soil pH of 7.6, texture of about 33% soil (<2 mm), 67% pebbles (>2 mm), and <2% organic matter. NVS was incorporated to a depth of 15 cm, and *M. incognita* population densities (juveniles and eggs) were monitored. The study showed that nematode population densities were reduced by NVS at 36 days, but this effect disappeared by 69 days, when nematode population densities increased in treated plots. However, tomatoes treated with NVS produced more biomass.

Greenhouse studies in Michigan were conducted with a steam-sterilized sandy loam (87% sand, 8% silt, 5% clay) to test the response to NVS of different populations of *M. hapla* grown in tomato (*Lycopersicon esculentum*) cv. Rutgers (25) (Table 3). Populations of *M. hapla* from loamy sand, sandy, silt loam, and muck soils with pHs of 7.2, 6.6, 7.4, and 6.3, respectively, at 0 or 600 eggs per 100 cm^3 soil, were tested. These populations came from selected Michigan nursery and vegetable production systems within a radius of about 20 km and had shown differences in their reproductive potential. When treated with 0, 0.5, or 2 g of dry NVS per 100 cm^3 of soil (equivalent to approximately 10 and 40 dry t/ha), both doses of NVS had significant effects on population densities of all *M. hapla* populations compared with the controls; the high dose was more suppressive than the low dose (Fig. 6). This suggested that NVS may be effective against these field populations.

Considerations When Using NVS to Control Nematodes

During 5 years of research evaluating NVS for plant-parasitic nematode management, the only common thread was inconsistency (Table 3). This is not surprising considering the diverse environments in which NVS was tested. Our experiences have led to the conclusion that, for consistent, reliable, and environmentally friendly plant-parasitic nematode management, NVS application will have to be implemented with a deeper understanding of the mechanism(s) responsible for nematode suppression.

Proposed mechanism of nematode suppression by NVS. To determine the mode of action of NVS against plant-parasitic nematodes, laboratory experiments were conducted (52,54). NVS was applied to sand at 0.5, 1, 2, and 3% dry wt/wt (equivalent to 10 to 60 dry t/ha) with a nonamended control. *M. incognita* and *H. glycines* mortality and changes in sand-assay chemical properties were determined after 24 h. The most important chemical property related to nematode mortality was the high pH level generated in soil solution after amendment (54). To a lesser extent, the production of NH_3 played a role in nematode suppression. In subsequent laboratory experiments, the components of NVS (biosolids and AA) were applied to sand to determine the effects on nematode survival, soil solution pH, and NH_3 concentrations (52). Alkaline-stabilization of biosolids was necessary to achieve nematode suppression; nontreated biosolids did not suppress *H. glycines* to the same level as equivalent amounts of NVS. Suppression was attributed to soil solution pH levels after biosolid amendment, which were never greater than 8.5, whereas NVS amendment resulted in pH levels greater than 10.0 at rates of 1% dry wt/wt or higher. In the same experiment, there was a weak relationship between the amount of NH_3 generated after NVS amendment and *H. glycines* mortality. These laboratory experiments provide evidence to suggest that the pH-mediated transformation of NH_4^+ to NH_3 (Fig. 2C) is at least partially responsible for the plant-parasitic nematode suppression achieved after amendment of soil with NVS. The challenge is to understand this chemical reaction and to utilize it in an environmentally responsible manner to achieve nematode suppression.

The toxicity of gaseous NH_3 to plant-parasitic nematodes and other soilborne pests is well documented (8,31,34,36, 40,48). Suppression of plant-parasitic nematodes by NH_3 depends on its concentration in soil air and solution, which is determined by several factors including NH_3/NH_4^+ equilibrium. The equilibrium between NH_3 and NH_4^+ is dependent on the pH of soil solution (and temperature), and follows the Henderson–Hasselbalch equation (Fig. 2C). For example, at pH 9.5, approximately half of the NH_3 plus NH_4^+ exists as NH_3 at a temperature of 20°C (Fig. 7). Despite NH_3 toxicity, the use of NH_3 for controlling plant-parasitic nematodes has not been accepted commercially (31). Several reasons have been cited, including the dependence of NH_3 toxicity on environmental conditions, the large amounts of NH_3/NH_4^+ needed to achieve control, and the frequent phytotoxic effects of high rates on plants. We and others (32) believe that excessive rates of nitrogenous materials have been ineffective in previous research because specific soil pH values were not present to generate toxic concentrations of NH_3 in soil solution. We believe NVS can be used to transiently raise soil pH (≥10.0), which converts most soil NH_4^+ to the active gaseous NH_3 form (Figs. 2C and 7). This is a delicate balance,

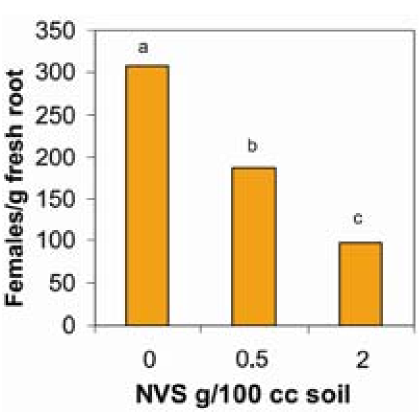

Fig. 6. Effect of 0, 0.5, and 2 g of dry N-Viro Soil (NVS) per 100 cm^3 of sandy loam soil on the densities of *Meloidogyne hapla* populations (with permission from Elsevier, Orlando, FL) (25).

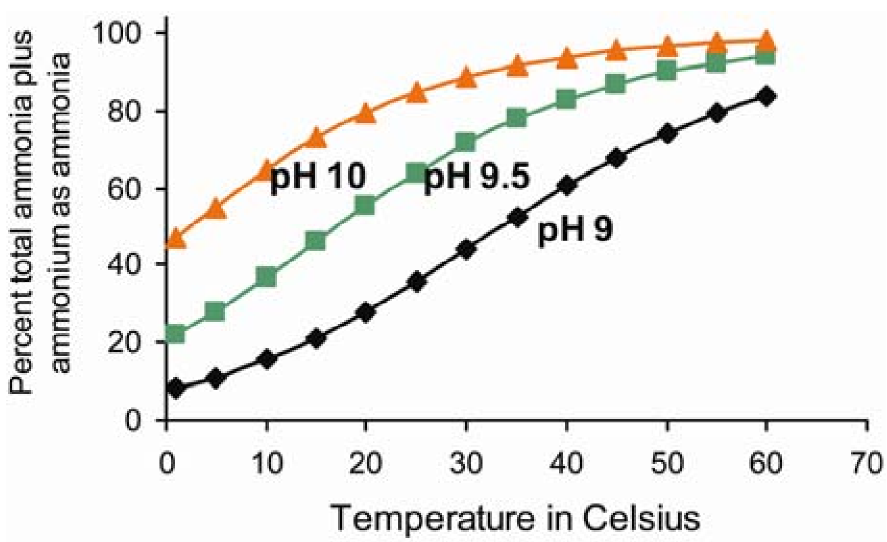

Fig. 7. The equilibrium between ammonium (NH_4^+) and ammonia (NH_3) as influenced by temperature and pH (with permission from M. Tenuta, University of Manitoba, Winnipeg, MT).

and care must be taken to minimize negative effects on soil structure and chemistry. We therefore believe that management of plant-parasitic nematodes using NVS should be advocated only in those environments where this chemical reaction (pH and NH_3) can be manipulated effectively.

Soil properties. Our field studies were conducted on a range of soil textures, from sand in North Carolina to loam in Iowa and calcareous in Florida, and results were variable across the sites (Table 3). In general, we did not observe plant-parasitic nematode suppression in the sandy loam to loam soils of Iowa, and had some nematode suppression, especially when higher rates were applied, when soil textures ranged from sand to loamy sand in North Carolina, Michigan, and Maryland. Previous research provides evidence of why NVS was effective in some soil textures while not in others. Tenuta and Lazarovits (41) identified soil properties associated with the accumulation of NH_3. Ammonia toxicity occurred in soils with low rates of nitrification, low levels of cation exchange capacity, and organic carbon contents, but high bulk densities and sand contents. In another study (42), cation exchange reactions involving the adsorption of NH_3 and NH_4^+ to soil particles was not a primary determinant of NH_3 accumulation, and bulk density only slightly affected NH_3 accumulation. The authors concluded that nitrogenous amendments should be applied to soils having low organic carbon and high sand content.

Soils also differ in their pH buffering capacities, which would influence the ability of NVS to raise soil pH. The initial pH of NVS is above 12.0, and care should be taken as pH can adversely affect the behavior of soils and nutrient availability (10,13,18). This means that NVS should be applied only to those soils where pH can be rapidly returned to normal after promoting the accumulation of NH_3 in soil. Candidate soils would include sandy, low organic matter, alkaline soils. If properly managed, pH recovery in NVS-amended soils can be rapid (20). For example, in Florida, 20 dry t/ha of NVS was applied to a Krome gravelly loam (bulk density 1.42 g/cm^3, 51% coarse material, 18% sand, 65% inorganic carbon, and 1.5% organic carbon), initial pH 7.9, and the change in soil solution pH over time was measured (Y. Li and I. A. Zasada, *unpublished data*). Immediately after NVS amendment, soil solution pH was greater than 11.0, but after 10 days the pH had returned to 8.5, slightly above the initial pH. While we do not advocate increasing soil pH to above 11.0, this does provide an example of a soil where nitrogenous amendments in combination with elevated pH could be implemented for plant-parasitic nematode management.

Environmental conditions. As discussed in a previous section, the generation of concentrations of NH_3 toxic to plant-parasitic nematodes depends on its concentration in soil air and solution, which is determined by NH_4^+ concentration, NH_3/NH_4^+ equilibrium, soil pH, and the environmental conditions present when nitrogenous amendments are added to soil. It has been shown that external factors need to be taken into consideration when predicting NH_3 volatilization following manure application (14). Two environmental conditions to consider are soil temperature and moisture.

Soil temperature was found to explain the largest variability in soil NH_3 volatilization (35) (Fig. 7). Ammonia volatilization after urea application was highest after 2 days and corresponded with daily maximum temperatures (9). The volatilization of NH_3 increased 18% as the temperature of incubation was increased 16°C, from 4 to 20°C (50). Also, whereas a low temperature favors a high equilibrium yield of NH_3, it dictates that a long time will be required to obtain the yield. Because higher soil temperatures promote the rapid conversion of NH_4^+ to NH_3, to obtain plant-parasitic nematode control and to minimize excess additions of N to soil, NVS application should occur when soil temperatures are warm. This timing may or may not be realistic depending upon the production system in which this technology would be employed.

Moisture also influences the retention and loss of NH_3 from soil. Volatilization has been shown to increase with decreasing soil water content (5,12,19,35). Adding water to soil after the application of urea decreased NH_3 losses by 15% (6). Similarly, Liu et al. (19) reported that NH_3 volatilization rate at 20% field moisture capacity (FC) was two- to three-fold greater than that at 80% FC for various forms of N fertilizers applied to four Washington and Florida soils. Heavy rainfall was a major determinant in a model predicting the transfer of NH_3 across the soil/air interface (35). It is more difficult to understand the role of moisture and how it relates to NH_3 exposure to plant-parasitic nematodes. When soils were amended with bone meal, NH_3 toxicity occurred in those soils with low moisture levels (42). In another study (41), soil moisture only slightly affected the accumulation of NH_3 in an alkaline loam soil. While low soil moisture increases NH_3 volatility, it stands to reason that the concentration of NH_3 in solution would be less dilute, therefore increasing the concentration to which nematodes are exposed. Conversely, although high soil moisture minimizes volatility, it also makes the concentration of NH_3 in solution more dilute. The exact soil moisture necessary to optimize the use of NVS, alone or in combination with another N source, is not clear but will vary from soil to soil.

Application rates. The rates of NVS evaluated in our experiments ranged from 6 to 100 dry t/ha (Table 3). As a limestone substitute, NVS would typically be applied at a rate of 10 t/ha dry weight every 2 to 4 years, depending on location. The lower rates used in our field studies did not consistently control plant-parasitic nematodes, so clearly a liming rate of NVS would not be effective. Higher rates were needed to achieve plant-parasitic nematode control, but these higher rates also resulted in dramatic increases in soil pH, a practice that is not encouraged because of negative effects on soil nutrient availability and structure. The goal is to apply a rate of NVS that facilitates a transient increase in pH and the production of toxic concentrations of NH_3 in soil solution. Because of the way NVS is produced, N exists almost entirely as organic N. To achieve toxic concentrations of NH_3, it may be necessary to add additional nitrogen in the form of fertilizers or nitrogenous amendments. Achieving nematode management through the manipulation of the nitrogen cycle is a fine balancing act and is advocated only in environments where this can be achieved in an environmentally responsible manner. It is important to point out that the source of ammoniacal-N (e.g., chicken litter, urea, bone meal) needs to be labile in order to take advantage of the rapid increase in pH that occurs after the addition of NVS to soil.

Product variability. To determine the ability to implement NVS for plant-parasitic nematode management across geographic locations, NVS from different processing facilities were tested against *H. glycines* (52). Of the five NVS sources tested (at rates equivalent to approximately 60 dry t/ha), four of them reduced *H. glycines* juvenile abundance more than 95%; a single source resulted in 56% reduction. NVS manufactured from different source materials and formulations have markedly different properties. Differences occur in the AA used, treatment of biosolids prior to stabilization (primary [3 to 7% solids] versus secondary [0.5 to 2% solids]), and the materials present in the wastewaters (28).

One of the most important considerations, regardless of NVS source, is the age and pH of the product. For NVS to be used as a vehicle to transiently increase soil pH to promote the production of NH_3, the initial pH of the NVS must be greater than 12.0. This will require that a relatively young batch (1 to 2 months) of NVS be used for plant-parasitic nematode management. Despite the differences in NVS composition from different locations, our research demonstrated that NVS produced from a diversity of by-products has the potential to be implemented on a wide geographic scale for plant-parasitic nematode management.

Application methods. In these studies, NVS was applied as a surface broadcast application, broadcast then incorporated application, and banded application (Table 3). We do not have a comparison of how higher rates of NVS, where we observed nematode mortality, performed when applied to the surface versus incorporated, but previous research indicates that narrow-banded application of manure significantly reduced NH_3 volatilization compared with broadcast application (14). The mean cumulative volatilization for broadcast application was 77% of the total ammoniacal-N applied, whereas it was 20% for narrow-banded application. These results indicate that NH_3 losses can be minimized, which would also increase exposure concentrations of NH_3 to plant-parasitic nematodes in soil.

Another application method that would minimize loss of NH_3 to the environment and promote the production of NH_3 is tarping of the soil after application. Because temperature influences the equilibrium between NH_4^+ and NH_3 (Fig. 7) and because tarping increases soil temperature in some environments (39), tarping should increase the amount of NH_3. Sealing treated soil in plastic bags reduced the amount of NH_4OH required to kill *M. javanica* (31). The use of tarps after the application of NVS to achieve nematode suppression would be most applicable to smaller acreage or higher value crops.

Future of Biosolid Use for Nematode Management

Many of the trends associated with land application of biosolids are likely to continue (3). Land application will increase, and increasing costs will discourage landfilling and incineration of biosolids. Coupled with this will be the increased production of class A, exceptional quality biosolids requiring fewer reporting requirements because they contain lower amounts of heavy metals and undesirable chemical and biological constituents. NVS is one of these products.

Public perception is the key to the acceptance of biosolids for new uses (28). Some food processors currently restrict the use of biosolids on agricultural products. Extensive public education and awareness programs need to be implemented for biosolids to be viewed as a resource rather than a disposal problem (4,28). Regardless of where an individual, company, university, or federal agency stands regarding the controversial use of biosolids on agricultural lands (38), the inescapable question still exists: how will we as a society manage our waste? With a growing population and increasing amounts of biosolids, it is essential to implement recovery options for biosolids and ultimately tactics for their disposal that are safe for humans and the environment, and are economically viable.

From a plant-parasitic nematode management perspective, the two largest obstacles for the implementation of this technology are its inconsistent performance and the acceptance of using NH_3 in combination with elevated soil pH as a soil disinfectant. Our results indicate that if NVS is to be applied alone for plant-parasitic nematode management, then the rate will have to be greater than 50 dry t/ha. This is not economically or environmentally acceptable. The amount of NVS, when applied alone, required for nematode suppression results in undesirable soil pH levels, which could lead to nutrient management problems. From an economic perspective, it may not be feasible to apply more than about 10 dry t/ha/year. The primary cost in utilizing NVS is the cost of transportation and spreading of this amount of material.

However, there is solid evidence from the research presented here and by others (31,32,40) that the mechanism responsible for nematode suppression by NVS, the production of NH_3 in combination with elevated pH, may be possible to achieve in some environments. In this case, the rate of NVS would be reduced and used to achieve a transient increase in soil pH to facilitate the production of NH_3. The environments where this technology could be employed will include only those soils where this chemical reaction can be maximized (sandy, low organic matter, or alkaline soils) and high value or small acreage crops where soil moisture and temperature can be manipulated by tarping or other tactics.

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