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The Effect of Manure Application Method on Nutrient and Microbial Runoff Transport and Soil Biological Health Indicators

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by

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THE EFFECT OF MANURE APPLICATION METHOD ON NUTRIENT AND MICROBIAL RUNOFF TRANSPORT AND SOIL BIOLOGICAL HEALTH INDICATORS

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Two projects were completed to provide significant new information to the agricultural industry regarding the environmental implications and soil health impacts related to land application of swine manure. The first study reports on the runoff transport of nutrients and microbials as affected by manure application method and time following application. The second study provides information about the effect of application method and time following application on soil health indicators using arthropod abundance and diversity as a biological indicator. The information gained through these studies will provide beneficial information to the pork industry on the impact of manure application method and the timing of application on limiting the movement of manure constituents with runoff while improving soil biological health.

The first field study was conducted to measure the effects of manure application method and time following application on runoff transport of nutrients and microbials. Swine slurry was applied to the soil by a commercial applicator in June 2014 using broadcast and injection methods. Simulated rainfall events were applied to the study plots following manure application in June, July, and August 2014. The broadcast treatment resulted in significantly greater dissolved and total phosphorous runoff loads than the injection treatment. Soil erosion was greater for the injection plots than for the broadcast treatment sites. Overland flow rate variation had a significant impact on all measured

water quality parameters. Significant reductions in nutrient transport loads were observed on plots where slurry was injected rather than broadcast. Nutrient and microbial transport loads decreased significantly during the 45 days following slurry application for both treatments.

The second study was conducted to determine the impact of manure application method and time following application on soil health by examining the chemical and biological responses to the treatments over time. Swine slurry for this study was also applied to field plots in June 2014 by a commercial applicator using broadcast and injection methods. The broadcast treatment resulted in a significant increase in hypogastruridae and Isotomidae populations over time. For the broadcast treatment, significant initial increases in nutrient content were observed, while the injection treatment showed very little initial response with much greater increases later in the study. Time following slurry application had a significant impact on all measured soil characteristics and all but one of the arthropod Orders that were quantified. Application method had a significant impact on all measured soil chemical characteristics, but was only significant for the Hypogastruridae, Isotomidae, and Pseudoscorpion populations.

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INTRODUCTION

United States pork producers marketed over 148 million hogs in 2014, returning $26.5 billion in gross receipts (USDA-NASS, 2015) and providing approximately 547,800 people with jobs that are associated with the pork industry (National Pork Producers Council, 2015). Daily manure production for the estimated 68.4 million pigs in the U.S. swine inventory (as of September 1st, 2015; USDA-NASS, 2015) can be estimated at approximately 342,000 tons (USDA-NRCS, 2008). The majority of livestock manure is spread, injected, sprayed, or otherwise applied to agricultural land as a nutrient resource for growing plants (USDA, 2008).

The nutrients and organic matter in swine manure are valuable resources that can be used for soil improvement and crop production. Manure application can positively influence soil physical properties by increasing water infiltration and retention, improving soil aggregate stability, and decreasing erosion and soil loss. Land application of manure can also improve soil chemical properties by increasing the concentrations of nitrogen, phosphorous, micronutrients, and organic matter, which are necessary for optimal plant growth. Unfortunately, manure application to soil carries potential environmental risks when amount, timing, and method of application are not optimized. Application of manure to agricultural land has the potential to contribute to elevated concentrations of phosphorous and nitrogen in surface waters (Bergström and Goulding, 2005) through runoff losses. Excessive phosphorous and nitrogen concentrations in fresh surface water contribute to algal blooms, reduced dissolved oxygen concentrations, and a decreased ability of aquatic ecosystems to support animal and plant life. In addition to the transport of nutrients in agricultural runoff, microbials can also be transported in runoff from

manured fields. Microbial transport has the potential to distribute antibiotic resistant bacteria and antimicrobial resistant genes in the environment along with pathogenic organisms.

The environmental risks associated with improper management of manure are well established and continue to be addressed through regulations established by federal and state regulatory agencies, including the United States Environmental Protection Agency (USEPA) and state departments of natural resources and environmental quality. These regulatory entities are responsible for enforcing the agricultural provisions of the Clean Water Act (CWA) and Effluent Limit Guidelines (ELGs), requiring all concentrated animal feeding operations (CAFOs)[1] with the potential to discharge pollutants to waters of the United States to obtain a National Pollution Discharge Elimination System (NPDES) permit and implement a nutrient management plan (NMP) (Kaplan et al., 2004; Feinerman et al., 2004). The purpose of the NMP is to ensure adequate land is available to utilize manure nutrients produced on the operation and that appropriate rates, methods, and timing of nutrient applications are planned. Though manure application to agricultural fields may be performed on a nitrogen or phosphorus basis, either application strategy must be performed at a rate that does not exceed the uptake and utilization capabilities of the soil and crop for the target nutrient. This practice minimizes the risk of nutrient loss to surface and ground water due to leaching and runoff from applied fields, while maintaining the ability to use the beneficial nutrients and organic matter in manure to improve soil fertility.

SOIL HEALTH

[1] A large swine CAFO is considered a facility housing at least 2,500 swine, weighing 55 pounds or more each, or 10,000 swine, weighing less than 55 pounds each (EPA, 2012).

Soil health, often used interchangeably with soil quality, is a complex soil characteristic that is not easily quantified, directly measured, or even distinctly defined. Part of the complexity in quantifying soil health arises from the number of factors that impact it. According to Kibblewhite et al. (2008), soil type, organisms and their functions, carbon and energy, and nutrients all play an important role in defining the "health" of soil. Given this wide range of factors impacting the quality of soil, it is difficult to quantify soil health by any single method or indicator. However, a combination of biological, chemical, and physical information about a soil can lend greatly to the understanding of its condition, especially when compared with other locations and fields within a relative area. By using relevant information about the biological, chemical, and physical properties from a population of fields, an improved understanding of soil health for a single location can be achieved.

The working definition employed in this application is derived from the similar and critical aspects of a number of other authors' proposed definitions, as well as from the NRCS definition. Kibblewhite et al. (2008) explains that "a healthy agricultural soil is one that is capable of supporting the production of food and fiber, to a level and with a quality sufficient to meet human requirements, together with continued delivery of other ecosystem services that are essential for maintenance of the quality of life for humans and the conservation of biodiversity". Rüdisser et al. (2015) defines soil health based on its stability, biological diversity, internal nutrient cycling, and resilience to stressors or disturbances. Finally, Doran and Safley (1997) consider soils healthy based on their ability to produce or function according to their potential with the understanding that this potential changes over time as a result of human use and management and natural events. Therefore, in this application, a healthy soil will be considered

one that functions as a living system and supports human, plant, and animal productivity to its maximum potential without endangering the health of these organisms.

An important clarification in the discussion of soil health is the difference between soil health and soil quality. Soil quality describes the measurable physical and chemical properties of the soil, which pertain most directly to human plant growth activities (Curell, Gross, and Stenkie, 2012). As mentioned previously, however, soil health includes a much wider environmental and biological aspect, along with soil quality measurements, in its consideration. The additional biological factors accounted for in soil health are what make it so much more complex to define and measure.

The most substantial anthropogenic influences on soil quality and health are land use and management practices (Rüdisser et al., 2015; Kibblewhite et al., 2008). The main contributing land management factors include tillage, application of animal manures and inorganic fertilizers, pesticide use, crop residue management, crop rotations, and cover crops (Kladivko and Clapperton, 2011; Sapkota et al., 2012). Tillage contributes to erosion, nitrogen losses, disruption of the soil structure, disturbance of the natural soil microbial and invertebrate environment, and decreased water infiltration capacity (Thayer et al., 2012; Gilley et al., 2007; Reicosky et al., 2011). Application of animal manures or inorganic fertilizers improves soil nutrient content and organic matter content (in the case of manure), but does have the potential to increase the risk of nutrient and microbial transport to surface waters (Eghball and Gilley, 1999; Gilley and Risse, 2000). The process of land applying livestock manure can also impact the biological component of the soil environment by altering the soil chemical balance and structure. Pesticide use improves crop resiliency but can have a detrimental effect on the biodiversity of the ecosystem at the soil microbial and invertebrate level. These factors can significantly impact nearly every

aspect of the soil system, explaining the critical role that land use and management have in maintenance of good soil health.

Soil Arthropods as Biological Indicators of Soil Health

Nature has a fascinating ability to adapt to change that is nearly impossible for humans to recreate in such detail and complexity. This adaptability holds true for every categorization and level of nature. Such adaptability and uniqueness means that some of the most quality information at our disposal can only be obtained by observing the natural reactions to and mechanisms affected by a change or disturbance imposed on an ecosystem. Therefore, the best indicators of changes in soil health may actually be the organisms living within the soil environment.

The key organisms associated with the upper soil layers that are both easily and inexpensively measured are soil arthropods (Parisi et al., 2005). These organisms exhibit a high level of sensitivity to changes in the soil environment and to land management practices (Sapkota et al., 2012). The simple, inexpensive methods of measurement and high level of sensitivity demonstrated by soil arthropods make them ideal indicators of soil health for this application.

Soil arthropod species richness is dependent upon the diversity of their feeding habits and energy sources as well as the differentiation of niches displayed in the community (van Straalen, 1997). Biodiversity within the soil biota community is essential to the long-term integrity, function, and sustainability of the terrestrial ecosystem (Pankhurst, 1997). Biological roles within the terrestrial environment that create niches and diversity in the soil arthropod communities include nutrient cycling and mobilization (i.e., carbon, nitrogen, phosphorous), degradation and

stabilization of organic matter, and soil structure maintenance (Sparling, 1997; Torstensson, Pell and Stenberg, 1998; Elia et al., 2010; Kladivko and Clapperton, 2011). Arthropods play a critical role in the balance of the soil environment by functioning as both litter transformers and soil structural engineers. Litter transformers contribute to the soil environment through the ingestion and breakdown of organic matter to forms that are usable by other soil organisms that are then responsible for the mineralization of those nutrients to plant-available inorganic forms (Muturi et al., 2011). Arthropod feces help to stabilize the soil and increase its nutrient storage capacity through the formation of soil aggregates and humus (Culliney, 2013). Therefore, the greater the diversity and population size within a soil arthropod community, the greater the level of activity within each of the biological soil roles. The quantification of soil arthropods can be reported based on individual abundance and the QBS index (Qualità Biological del Suolo) as suggested by Parisi et al. (2005). The QBS index is predicated on the concept that soil of greater quality will support a greater diversity of microarthropod groups that are well adapted to the soil habitat (Parisi et al., 2005). A taxonomic diagram of common soil arthropods is shown in figure 12. Studies investigating the effects on soil arthropod populations from organic versus inorganic fertilizers, long-term fertilization, changes in pH, and organic matter content have been conducted. However, little information can be found describing the effects of manure application method and the timing of manure application on soil arthropod abundance and diversity.

As livestock production intensifies in response to a rising world population and associated demand for protein sources, careful utilization of manure to obtain its full nutrient value while protecting environmental quality becomes increasingly important. While there are many components of livestock production that contribute to an operation or industry's overall sustainability, the management of manure as a soil amendment is certainly an important aspect of

environmental, social, and economic sustainability. A broad goal for managing manure in a sustainable manner is to manage the manure such that its value to crop production is fully recognized while environmental risk is mitigated by employing relevant management practices. The connection between livestock and crop production is an important aspect of sustainability for both industries and is a concept being promoted in Nebraska and nationwide. Demonstrating the mutual benefits to both industries of manure utilization in cropping systems as a soil amendment and promoting management practices for mitigating environmental risk associated with manure application form the basis of the thesis research presented here.

OBJECTIVES

A review of the literature relating to manure land application and the use of arthropods as biological indicators of soil health revealed the need for targeted investigations to quantify runoff transport of nutrients and microbials and to define the impact of manure application method and time since application on soil arthropod abundance and diversity. Therefore, the objectives of the research presented were:

Manuscript I. Runoff Nutrient and Microbial Transport Following Swine Slurry Application

1. Determine the effects of slurry application method and time since slurry application on runoff nutrient and microbial loads (mass or colony forming units (CFU) per unit area).
2. Compare the effects of slurry application method, time following slurry application, and runoff rate on nutrient and microbial transport rates (mass or CFU per unit area per unit time).

Manuscript II. Soil Invertebrates as Bioindicators of Agricultural Soil Quality Under Swine Slurry Treatments

1. Determine the effects of slurry application method and time following application on soil arthropod diversity and abundance.

THESIS PRESENTATION

This thesis is written in manuscript form as drafts for publication. Chapter II is written as a manuscript titled, "Runoff Nutrient and Microbial Transport Following Swine Slurry Application" and formatted for publication in *Transactions of the ASABE*. This chapter presents research on the effects of swine slurry application method and time following application on runoff transport of nutrients and microbials. The effect of varying inflow rates on nutrient and microbial transport is also reported. This information is intended to help determine how runoff nutrient and microbial loads fluctuate based on whether manure is applied to soil via broadcast or injection and whether time following manure application has an effect on runoff transport loads of these potential contaminants. The information also provides a basis for understanding how nutrient and microbial transport are impacted by varying overland flow rates given the method of application and time following application.

Chapter III is written as a manuscript titled, "Soil Invertebrates as Bioindicators of Agricultural Soil Quality Under Swine Slurry Treatments," formatted for submission to *Applied Soil Ecology*. This chapter addresses the impact of swine slurry application method and time following manure application on soil health using arthropods as biological indicators based on population diversity and abundance. This information is intended to help define how soil biological health is impacted over time following swine slurry application to soil by broadcast and injection methods.

A Manuscript Prepared for Submission to

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ABSTRACT

This study was conducted to measure the effects of swine slurry application method and time following slurry application on runoff transport of nutrients and microbials. Swine slurry from a commercial wean-to-finish swine production was applied to field plots using broadcast or injection methods at a rate required to meet annual nitrogen requirements for corn. Three simulated rainfall events were applied to the experimental plots for a 30-minute duration, separated by 24 h intervals, at an intensity of 70 mm h^{-1}. Following the third rainfall simulation event, inflow was applied at the top of each plot in four successive increments to simulate greater plot lengths. The dissolved phosphorous (DP) and total phosphorus (TP) loads of 0.35 and 0.46 kg ha^{-1} measured for the broadcast treatment were significantly greater than the 0.13 and 0.19 kg ha^{-1} obtained from the injected treatment. As time following slurry application increased from 2 to 44 days, DP and TP transport decreased from 0.35 to 0.14 and 0.52 to 0.18 kg ha^{-1}. Overland flow rate was a critical variable significantly affecting each of the measured water quality parameters. Runoff loads of DP, total phosphorous, NO3-N, and total nitrogen increased from 10.1 to 29.8, 12.9 to 35.5, 314 to 1341, and 346 to 1460 g ha^{-1} min^{-1}, respectively, as overland flow rate increased from 2.3 to 12.6 L min^{-1}. Significant reductions in nutrient transport were found on sites where slurry was injected rather than broadcast. Nutrient and microbial transport values were found to significantly decrease during the 45 days following slurry application.

Keywords.

Land application, Manure management, Manure runoff, Microbes, Nitrogen, Nutrients, Phosphorous, Runoff, Swine manure, Water quality

INTRODUCTION

Swine manure offers substantial benefits to soil when land-applied as it contains valuable organic matter and nutrients (Eghball et al., 2004) that can increase soil productivity, improve water infiltration, and reduce soil erosion potential. Phosphorous introduction to surface water is a strong contributor to reduction of water quality through eutrophication in fresh water lakes (Sims and Kleinman, 2005). Nitrogen entering surface water has the ability to produce hypoxic zones if transported to salt water systems like the Gulf of Mexico (Turner et al., 1997). Additionally, animal manure application in excess of crop requirements can result in excess salt and nutrient accumulation, which may increase the potential for groundwater contamination (Mathers et al., 1975).

Bacteria have the opportunity to enter the environment via land-applied manure (Warnemuende and Kanwar, 2000) and present a risk for pollution of soil, vegetation and surface and ground water. In North America, most of the waterborne disease outbreaks have been associated with drinking water systems in rural areas (Oun et al., 2014) and could be attributed to animal manure or domestic waste contamination. Runoff from sites with land-applied manure has been found to be an important source of water body microbial contamination (Daniel et al., 1998; Thurston-Enriquez et al., 2005). Results from previous studies suggest that large precipitation events have the potential to result in significant transfer of microbials in runoff that could significantly impact water bodies within the watershed (Thurston-Enriquez et al., 2005). Bacteria transported during a rainfall event can be quantified through total aerobic microbial

counts (TAMC) and qualified through MacConkey agar bacteria counts (MAC). TAMC quantifies the number of aerobic microbes present in a sample in colony forming units (CFU) and is a common test utilized in assessing drinking water quality. MAC isolates the Gram-negative and enteric bacilli in a sample, which are often the bacteria of greatest concern for human infection (*E. coli*, *Salmonella*, *Shigella*, etc.), and is commonly used as a test of fecal contamination present in water.

A study by Gilley and Risse (2000) found that application of manure resulted in reduced runoff and soil loss. The factors that influence runoff amount include cropping and soil management practices and timing, rate, and method of application (Khaleel et al., 1980). Application of liquid swine manure results in decreased runoff and erosion rates by providing a stabilizing effect on soil surfaces (Mitchell and Gunther, 1976). The implementation of either surface application or incorporation into the soil also significantly impacts the transport of manure constituents in runoff (Durso et al., 2011).

The rate of overland flow significantly impacts the transport of nutrients from fields receiving animal manure (Gilley et al., 2008; Gilley et al., 2013). Information collected on test-plots provides insight into the important transport mechanisms occurring on the field scale. Many factors may influence the fate and transport of nutrients and microbes from land-applied manures, including manure management (application method and timing) and source management (applied versus soil nutrients). Currently, little information is available concerning the effects of swine slurry application method and time since manure application on the concentrations of nutrients and bacteria in runoff. The objectives of this study were to: (1) determine the effects of slurry application method and time following slurry application on runoff nutrient and microbial loads (mass or colony forming units (CFU) per unit area) and (2)

compare the effects of slurry application method, time following slurry application, and runoff rate on nutrient and microbial transport rates (mass or CFU per unit area per unit time).

MATERIALS AND METHODS

STUDY SITE CHARACTERISTICS

This field study was conducted from June through August 2014 at the University of Nebraska-Lincoln Rogers Memorial Farm, located 18 km east of Lincoln, Nebraska in Lancaster County (figure 13). The Aksarben silty clay loam soil at the site (fine, smectitic, mesic Typic Argiudoll) contained 15% sand, 48% silt, 37% clay, 3.5% organic matter, 1.5% total carbon, and had a mean slope of 10% (Kettler et al., 2001). This soil developed in loess deposits under prairie vegetation and is considered a benchmark soil of the Corn Belt. The study site has been cropped using a no-till management system under a corn (*Zea Mays L.*), grain sorghum (*Sorghum bicolor* (L.) Moench), soybean (*Glycine max* (L.) Merr.), and winter wheat (*Triticum aestivum* L. cv. Pastiche) rotation. Corn was planted during the 2013 season and the site was left undisturbed following harvest in October 2013. Herbicide (glyphosate) was applied to the area as needed to control weed growth. The study site has not had a manure application since at least 1966.

PLOT PREPARATION

Thirty-six 0.75 m × 2 m plots were established, with the 2 m plot dimension parallel to the slope in the direction of overland flow (figure 14). Experimental treatments included two

manure application methods (broadcast and injected; figure1) and control plots (figure 2). Each of the treatments was replicated three times.

Swine slurry was collected from a deep pit on a commercial 8000-head wean-to-finish swine operation in north central Nebraska just prior to field application. Samples of the swine slurry were collected before application for solids and nutrient analyses, which were performed at a commercial laboratory. Mean measured values of NO_3-N, NH_4-N, TN, TP, water content, EC, and pH for the slurry were 3.91 g kg^{-1}, 0.0024 g kg^{-1}, 5.49 g kg^{-1}, 0.58 g kg^{-1}, 96.57%, 42.35 dS m^{-1}, and 8.0, respectively. A commercial operator was hired to inject and broadcast slurry at the experimental site. For injection, a v-shaped chisel (horizontal sweep) implement was used on a 6-row applicator for manure placement. For broadcasting, the applicator was lifted above the soil while maintaining a steady speed and flow rate. The slurry was applied at a rate of approximately 46,800 liters per hectare.

RAINFALL SIMULATION PROCEDURES

The rainfall simulation procedures used in the study were adopted from the National Phosphorous Research Project (Sharpley and Kleinman, 2003). Rainfall was applied to paired plots at an intensity of approximately 70 mm h^{-1} for 30 minutes by a portable rainfall simulator based on the design by Humphry et al. (2002). Two additional rainfall simulation tests were conducted on the same plots at approximately 24-hour intervals. Two rain gauges were placed along the outer edge of the plots and one was placed in the center between the plots (figure 15).

Field rainfall simulation tests were conducted 1 day, 1 week, 2 weeks, 3 weeks, and 6 weeks following slurry application (figure 1). Simulation tests were also conducted on control plots 1 week before and 5 weeks after slurry was applied to the adjoining plot areas. Eight plots

were examined during each of the test periods. Each plot was examined only once throughout the course of the study. No significant precipitation events occurred during the study period.

Water used in the study was obtained from an on-site irrigation well. Reported nutrient contents represent the difference between nutrient measurements in the runoff and those in the irrigation water. Measured mean concentrations of DP, TP, NO_3-N, NH_4-N, and total nitrogen (TN) in the irrigation water were 0.19, 0.19, 15.3, 0.04, and 15.3 mg/L, respectively. The irrigation water had a mean EC of 0.75 dS m^{-1} and a pH of 7.35.

Plot borders channeled runoff into a sheet metal lip that emptied into a collection trough that extended along the bottom of the plots. The troughs diverted the runoff into plastic buckets where it was transferred by a sump pump into large plastic containers. Following each rainfall simulation event, the containers were weighed to determine the mass of the runoff. The accumulated runoff was agitated immediately before sample collection to maintain suspension of solids. Runoff samples were collected for water quality analysis and sediment analysis within a few minutes following completion of the rainfall simulation tests.

Runoff samples collected from each rainfall event were stored in 1-L plastic bottles for nutrient and sediment analyses and kept on ice until delivery at the laboratory. The plastic bottles used for sediment analyses were transported to the laboratory and total mass was measured. Tare weights of the bottles had been previously obtained. The plastic bottles were then dried in an oven at 105°C and weighed again to determine the mass of sediment (total solids) remaining. The sediment content of the runoff samples was determined by calculating the mass of material remaining in the bottles after drying divided by the mass of water contained in the bottles before

drying (the total measured mass of liquid minus the mass of total solids). The mass of dissolved chemical constituents contained in the runoff was assumed to be negligible.

Extracts from centrifuged and filtered runoff samples were used to determine nutrient concentrations. A Lachat system (Zellweger Analytics, Milwaukee, WI) was employed to analyze all samples for dissolved phosphorous (DP), NO_3-N, and NH_4-N. Non-filtered samples were stored in a cooler at 2°C and then analyzed at a commercial laboratory for total phosphorous and total nitrogen.

ADDITION OF INFLOW

Increased runoff rates resulting from upslope contributing areas were simulated using well-established procedures (Laflen et al., 1991; Misra et al., 1996; Monke et al., 1977). Simulated overland flow was applied at the up gradient end of each plot after the first 30 min of the third rainfall simulation event to examine the influence of varying runoff rates on the transport of nutrients. Rainfall continued during the overland flow tests. Inflow was added in four successive increments to produce average runoff rates of approximately 3.2, 9.6, 14.2, and 21.2 L min^{-1}.

A mat made of a green synthetic material and generally used as an outdoor carpet was placed beneath the inflow device at the up gradient end of the plot to prevent scouring and create more uniform runoff distribution over the plot. Runoff generated during the inflow tests was transferred into a flume where a stage recorder was mounted to measure flow rate. Overland flow rate increments were only increased once the previous rate had achieved a steady runoff value (determined with the flume and stage recorder) and samples had been collected for both nutrient and sediment analysis. Each inflow increment had a duration of approximately 8 min, which was

the period of time generally required for steady-state flow conditions to become established and for nutrient and sediment analyses samples to be collected.

Information on soil nutrient values was collected to help explain differences among runoff nutrient measurements. Before the rainfall simulation tests were initiated for each test interval, three soil cores were collected by hand from each plot using a 5 cm wide x 30 cm long soil probe. The three cores from each plot were segmented into 0 – 10, 10 – 20, and 20 – 30 cm depth increments and composited. The soil samples were then air-dried, ground, and analyzed for water-soluble P (WSP), Bray and Kurtz No.1 P (BKP), NO_3-N, NH_4-N, EC and pH.

The Murphy and Riley (1962) method, which involved shaking 2 g of soil for 5 min with 20 mL of deionized water, was used to measure WSP. As an index of P availability, the BKP procedure (Bray and Kurtz, 1945) provides a relative estimate of P concentration in the soil solution that limits the growth of plants. Soil NO_3-N and NH_4-N concentrations (extracted using a 2 molar KCl solution) were measured using spectrophotometry (Lachat system from Zellweger Analytics, Milwaukee. WI).

STATISTICAL ANALYSES

The effects of manure application method (broadcast vs. injection) and time following slurry application (1 day, 1 week, 2 weeks, 3 weeks and 6 weeks) on selected runoff water quality parameters were determined by performing analysis of variance (ANOVA) (SAS, 2011). For a given plot, water quality measurements from each of the three rainfall simulation runs were treated as repeated measures. If a significant difference was identified, the least significant difference test (LSD) was used to identify differences among experimental treatments. A probability level of <0.05 was considered significant. For the inflow test runs, ANOVA was performed to determine the effects of application method, time following slurry application, and

flow rate on the measured water quality parameters. ANOVA was also used to determine if changes in water quality characteristics occurred between the two rainfall simulation periods occurring on the control plots.

The effects of timing of soil collection and soil depth on chemical and physical characteristics of the control plots were determined using ANOVA. ANOVA was also used to determine the effects of application method, timing of soil collection, and depth on the measured soil characteristics. The LSD test was used to identify differences among experimental treatments, if a significant difference was identified.

RESULTS AND DISCUSSION

SOIL CHARACTERISTICS

Mean measured soil concentrations of WSP, BKP NO_3-N, and NH_4-N for the control plots where no slurry was applied were 2.0, 20.9, 8.5, and 3.55 mg kg^{-1}, respectively (table 1). Mean EC and pH values were 0.29 ds m^{-1} and 6.52, respectively. No significant differences in measurements of water WSP, BKP, NH_4-N, or pH were found between test periods. However, timing of soil collection had a significant effect on measurements of NO_3-N and EC, with values increasing from 5.6 to 11.4 mg kg^{-1} and 0.26 to 0.31 dS m^{-1}, respectively, during the 6 week period between test intervals (table 1). The change in NO_3-N concentration on the control plots can be attributed to the process of nitrogen mineralization occurring over time.

Application method x soil depth interactions on the plots where slurry was applied were found for WSP, BKP, NO_3-N, and EC (table 2). Application method x time x soil depth interactions were found for NH_4-N and pH. Soil measurements of WSP, BKP, NO_3-N, and NH_4-N were significantly greater on the plots where manure was broadcast, with values of 2.1, 28.3,

17.1, and 7.3 mg kg^{-1}, respectively, for the broadcasted plots and 1.8, 22.1, 10.5, and 3.6 mg kg^{-1}, respectively, for the injected plots (table 2). Manure application method did not cause significant changes in measured EC or pH values. On the plots where slurry was injected, soil samples were collected between the injection slots. Since slurry was not applied at the sample collection points, soil measurements would be expected to be less at these locations.

Time since slurry application significantly affected values of WSP, BKP, NH_4-N, and EC in soil. However, time did not have a significant impact on NO_3-N or pH measurements (table 2). With the exception of NO_3-N, the largest soil parameter values were observed during the first 15 days following slurry application.

Soil depth had a significant impact on each of the measured soil characteristics. There were significantly higher concentrations of all of the measured parameters in the top ten centimeters of the soil compared to the two deeper soil layers (10-20 and 20-30 cm). Significant differences in BKP, EC, and pH measurements were found among each of the three soil depths with BKP, EC, and pH values decreasing with depth.

BKP soil concentrations were significantly greater for the 0-10 cm depth than the 10-20 or 20-30 cm depths for both the broadcast and injected plots (figure 3). The 0-10 cm depth also had the greatest NO_3-N concentrations (figure 4). The soil concentrations of NH_4-N for the broadcasted plots at the 0-10 cm depth were greatest initially then decreased steadily throughout the study. However, the NH_4-N concentration was significantly greater at the 0-10 cm depth on the broadcast plots than at the 10-20 and 20-30 cm depths (figure 5). The NH_4-N soil concentrations for the injected plots did not follow a specific trend (figure 5).

RUNOFF CHARACTERISTICS

The nutrient values reported in this study represent the difference between runoff measurements and concentrations in the irrigation water. Application method x time interactions were found for DP, PP, TP, NH_4-N, and EC (table 3). Application method significantly affected measurements of DP, PP, TP, EC, and erosion (table 3). Significant variations in measurements of DP, PP, TP, NH_4-N, pH, erosion, MAC, and TAMC were found based on the length of time that had expired following slurry application.

Phosphorous Load in Runoff

The plots that were broadcasted with slurry had significantly greater mean runoff loads of DP than those that were injected with slurry, with measured values of 0.35 and 0.13 kg ha^{-1}, respectively (table 3). The broadcast plots also had greater mean DP runoff loads for each interval following slurry application than the injected plots (figure 6). Similar results were found in a study by Gilley et al. (2013) with mean runoff DP loads being significantly greater for broadcasted application than for disk or injection application. In addition, the largest DP load occurred during the first rainfall simulation period (figure 3). The average DP load decreased from 0.35 to 0.14 kg ha^{-1} as the time since slurry application increased from 2 to 44 days (table 3).

The plots where slurry was broadcast had significantly greater mean runoff loads of TP than the plots where slurry was injected, with values of 0.46 and 0.19 kg ha^{-1}, respectively (table 3). The greatest difference in runoff TP loads between the broadcasted and injected plots were observed during the first rainfall simulation period, two days following slurry application (figure 7). The broadcasted plots had larger measured runoff loads of TP for each of the intervals following slurry application. The values for TP in runoff consistently decreased from 0.52 to 0.18 kg ha^{-1} as the time since slurry application increased from 2 to 44 days (table 3).

Nitrogen Load in Runoff

The N-related water quality parameters, NO_3-N, NH_4-N, and TN, were not significantly affected by application method (table 3). Additionally, time since slurry application did not significantly influence runoff loads of NO_3-N or TN. However, time since slurry application had a significant effect on measured NH_4-N loads, with values decreasing from 2.17 to 0.14 kg ha^{-1} as the time following slurry application increased from 2 to 44 days (table 3).

A significant application method x time interaction was found for NH_4-N runoff loads. Significantly greater NH_4-N runoff loads were observed for the broadcasted plots than the injected plots for the first set of rainfall simulation data collected on day 2 following slurry applications (figure 8). However, as simulations continued on later dates, the injected plots showed greater runoff loads for days 9 and 16. The final two simulations on days 23 and 44 had very similar but relatively low runoff loads for both application methods.

EC, pH, Runoff, and Erosion Measurements

Measurements of EC were significantly greater for the broadcast than the injected treatments, with values of 0.82 and 0.79 dS m^{-1}, respectively (table 3). However, time since slurry application did not have a significant impact on measured EC values.

Erosion measurements for the injected plots were significantly greater than for the broadcast plots, with mean erosion values of 0.42 and 0.32 Mg ha^{-1}, respectively (table 3). Similar results were found in a study by Gilley et al. (2013) with soil loss from the broadcasted plots being significantly lower than for the disked and injected plot, with values of 87, 160, and 127 kg ha^{-1} min^{-1}, respectively. Soil disturbance resulting during the injection process was likely the cause of the increased erosion values. Significant differences in erosion values resulted from

the length of time that elapsed following slurry application. Erosion measurements obtained over time followed a non-uniform pattern.

Microbial Transport and Interactions

Slurry application method did not significantly affect runoff measurements of MAC or TAMC. However, time since slurry application significantly affected the transport of microbials (table 3), with MAC values decreasing from 13.54 to 12.45 log CFU ha^{-1} and TAMC values decreasing from 11.37 to 10.65 log CFU ha^{-1}, as time since slurry application increased from 2 to 44 days (figure 9). The ratio of TAMC to MAC bacteria counts was not significantly affected by either application method or time since slurry application (table 3).

Correlation coefficients of runoff water quality characteristics with microbial measurements are reported in table 4. MacConkey agar bacteria counts were significantly correlated to several water quality characteristics including PP, TP, NH_4, TN and EC. Total aerobic microbial counts were correlated to DP, PP, TP, NH_4-N, EC, and pH. The TAMC/MAC microbial constituent ratio was not significantly impacted by any of the measured water quality parameters.

RUNOFF CHARACTERISTICS AS AFFECTED BY OVERLAND FLOW

Since the upslope contributing area under field conditions is much larger than that provided by the 2 m long experimental plots, additional inflow was introduced at the top of the plots to simulate greater slope lengths. The experimental results are applicable to a much larger range of rainfall and runoff conditions when nutrient transport rate is related to flow rate.

When simulated overland flow was introduced during the experimental tests, it was not practical to capture and store all of the runoff that occurred. Therefore, nutrient and sediment

samples were collected under steady-state runoff conditions and nutrient load values per unit time are reported for this portion of the study.

Application x runoff rate interactions were found for DP and TP (table 5). Manure application method significantly affected DP, TP, and soil loss values (table 5). Time since slurry application significantly affected NH_4-N, EC, and pH measurements. Runoff rate significantly impacted each of the measured water quality parameters.

Phosphorous Measurements

Mean transport rates for DP and TP were significantly greater for the broadcasted plots than they were for the injected plots. Transport rates of 24.6 and 16.6 kg ha^{-1} min^{-1} were obtained for DP and 29.7 and 20.5 kg ha^{-1} min^{-1} for TP for the broadcast and injected treatments, respectively. The mean transport rates for DP and TP consistently increased from 10.1 to 29.8 g ha^{-1} min^{-1} and 12.9 to 35.5 g ha^{-1} min^{-1} as runoff rate increased from 2.3 to 12.6 L min^{-1}. Regression equations were derived relating the rate of transport of DP in runoff (y) in g ha^{-1} min^{-1} to runoff rate (x) in L min^{-1} (figure 10):

For plots with broadcast manure application:

$$y = 2.24x + 7.60 \ (R^2 = 0.99) \qquad (1)$$

For plots with injected manure application:

$$y = 1.52x + 5.09 \ (R^2 = 0.98) \qquad (2)$$

Regression equations were also derived relating the rate of transport of TP in runoff (y) in g ha^{-1} min^{-1} to runoff rate (x) in L min^{-1} (figure 11):

For plots with broadcast manure application:

$$y = 2.61x + 9.90\ (R^2 = 0.99) \qquad (3)$$

For plots with injected manure application:

$$y = 2.46x + 4.36\ (R^2 = 0.90) \qquad (4)$$

Gilley et al. (2013) examined the effects of overland flow rate on runoff nutrient transport as affected by land application method, swine growth stage, and runoff rate. Inflow was added to the top of the plots to produce runoff rates of 3.2, 9.6, 14.2, and 21.2 L min^{-1}. The study reported that variations in runoff rate have significant effects on the transport of DP, PP, and TP. Similar results were found in the present investigation, supporting the fact that increasing runoff rate has a significant impact on the rate of transport of DP, PP, and TP.

Nitrogen Measurements

No significant differences in mean NO_3-N transport rates were found based on slurry application method. Additionally, time since manure application did not have a significant effect on transport rates of NO_3-N. However, overland flow rate had a significant impact on the transport rate of NO_3-N with values consistently increasing from 314 to 1341 g ha^{-1} min^{-1}, as runoff rate increased from 2.3 to 12.6 L min^{-1} (table 5).

No significant differences in transport rates of NH_4-N were found between the broadcast and injected experimental treatments. Mean NH_4-N transport rates were significantly affected by time since slurry application, with values decreasing from 39.1 to 6.6 g ha^{-1} min^{-1} as time after slurry application increased from 2 to 44 days. Additionally, runoff rate had a significant impact on NH_4-N transport rates, which increased from 13.9 to 25.1 g ha^{-1} min^{-1} as runoff rate increased from 2.3 to 12.6 L min^{-1} (table 5). The runoff transport rates for NH_4-N were much smaller than those measured for NO_3-N.

Runoff transport rates for TN were not significantly affected by manure application method or the time that had elapsed since slurry application (table 5). However, overland flow rate significantly impacted TN transport rates with values consistently increasing from 346 to 1460 g ha^{-1} min^{-1} as runoff rates increased from 2.3 to 12.6 L min^{-1}.

Gilley et al. (2013) measured the effects of overland flow rate on runoff nutrient transport as affected by land application method, swine growth stage, and runoff rate. The study applied inflow to the top of the plots, resulting in runoff rates of 3.2, 9.6, 14.2, and 21.2 L min^{-1}. The study reported significant effects on the transport rates of NO_3-N, NH_4-N, and TN with the increasing overland flow rates. The present study had similar findings, as runoff rate was found to significantly affect the rate of transport of NO_3-N, NH_4-N, and TN.

EC, pH, and Soil Loss Measurements

Measurements of EC were not significantly affected by slurry application method, with a mean measured EC value of 0.71 dS m^{-1} obtained for both the broadcasted and injected treatments (table 5). However, time since slurry application and runoff rate each significantly affected runoff EC measurements. As runoff rate increased from 2.3 to 12.6 L min^{-1}, measured EC values decreased from 0.73 to 0.70 ds m^{-1}.

Measured pH values were also not significantly affected by slurry application method. However, time since slurry application and runoff rate significantly impacted measured pH values (table 5). The largest pH value (7.97) occurred during the initial test period, but no significant differences occurred among the remaining test intervals. A significant decrease in mean pH measurements of 8.01 to 7.79 was observed as runoff rate increased from 2.3 to 12.6 L min^{-1}.

The mean soil loss measurement of 40.3 kg ha^{-1} obtained on the broadcast treatment was significantly less than the mean soil loss measurement of 56.8 kg ha^{-1} min^{-1} obtained on the injected treatment (table 5). Additionally, as runoff rate increased from 2.3 to 12.6 L min^{-1}, soil loss rate increased significantly from 16.4 to 75.4 kg ha^{-1} min^{-1}. Time since slurry application did not have a significant impact on soil loss rate.

CONTROL PLOT RESULTS

The control plots were established in this study to determine if background soil nutrient values changed over a period of several weeks. The most significant observed changes in background soil nutrient concentrations were for NO_3-N, which changed significantly with both soil depth and time. NO_3-N concentrations decreased with soil depth from 13.2 to 7.1 to 5.3 mg kg^{-1} for the 0-10 cm, 10-20 cm, and 20-30 cm depth increments, respectively, and increased from 5.6 to 11.4 mg kg^{-1} from the beginning to the end of the six-week study period (table 1). The only other characteristic affected by time was EC, which increased from 0.26 to 0.31 dS m^{-1} (table 1). Soil depth significantly affected WSP, BKP, EC, and pH with values decreasing with depth from 3.8 to 0.9 mg kg^{-1}, 36.3 to 10.3 mg kg^{-1}, 0.37 to 0.21 dS m^{-1}, and 6.83 to 6.12, respectively (table 1).

Timing of the simulation tests had a significant effect on several of the control plot water quality parameters including DP, PP, TP, NH_4-N, and erosion with values changing from 0.06 to 0.04 kg ha^{-1}, 0.02 to 0.01 kg ha^{-1}, 0.07 to 0.05 kg ha^{-1}, 0.02 to 0.01 kg ha^{-1}, and 0.27 to 0.46 Mg ha^{-1}, respectively (table 6). Time since application only had a significant effect on NH_4-N, with transport rates decreasing from 3.4 to 1.4 g ha^{-1} min^{-1} (table 7).

Overland flow rate had a significant effect on all of the measured water quality parameters for the control plots except for NO_3-N. As runoff rate increased from 2.3 to 12.6 L

min^{-1}, nutrient transport rates increased from 3.3 to 17.2, 4.0 to 19.6, 190 to 1242, 0.6 to 3.7, and 210 to 1372 g ha^{-1} min $^{-1}$ for DP, TP, NO_3-N, NH_4-N, and TN, respectively (table 7). Additionally, the measured values for EC, pH, and soil loss changed from 0.71 to 0.70 dS m^{-1}, 8.09 to 7.89, and 10.7 to 62.7 kg ha^{-1} min^{-1}, respectively.

The phosphorous transport response to increasing runoff rate can be seen in figure 12. As runoff rate increased, both DP and TP transport increased linearly. Regression equations were derived relating the rate of transport of TP in runoff (y) in g ha^{-1} min^{-1} to runoff rate (x) in L min^{-1} (figure 11):

For TP runoff transport:

$$y = 1.49x + 1.58\ (R^2 = 0.97) \qquad (3)$$

For DP runoff transport:

$$y = 1.37x + 0.54\ (R^2 = 0.99) \qquad (4)$$

Therefore, variations in background soil characteristics could have had some impact on the observed transport loads from the plots on which slurry was applied.

CONCLUSIONS

Swine slurry is applied to agricultural fields as a fertilizer source to enhance crop growth, improve soil infiltration, improve soil moisture retention, and increase the availability of phosphorous, nitrogen, and organic matter in the soil. Two common methods of slurry application, broadcast and injection, were investigated in this study to determine their effects, in

combination with time following application and overland flow rate, on nutrient and microbial transport in runoff. The mean runoff loads from plots where slurry was applied had significantly greater DP, PP, TP, and EC values. However, slurry injection resulted in greater rates of soil loss than broadcast application, likely due to the tillage required in the injection application process.

Analysis of runoff water quality parameters revealed that time since slurry application had a significant impact on the mean measured runoff values of DP, PP, TP, NH_4-N, pH, soil loss rate, MAC, and TAMC. As time following manure application increases, the nutrients present in organic forms in the manure begin to mineralize to plant-accessible forms. Therefore, the concentrations of nutrients, once applied to soil, are subject to change over time.

The effect of overland flow rate on nutrient transport in runoff was also investigated. It was determined that increasing runoff rates had a significant impact on each of the measured water quality parameters, including DP, PP, TP, NO_3-N, NH_4-N, TN, EC, pH, and soil loss. Values for each of the measured water quality parameters increased significantly with increasing overland flow rate with the exception of EC and pH. Manure application method, time since slurry application, and runoff rate should each be considered when estimating nutrient and microbial transport loads from areas on which swine slurry is applied.

ACKNOWLEDGMENTS

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APPENDIX

Figure 1. Schematic showing the plot layout and rainfall simulation period for the broadcast and injected plots

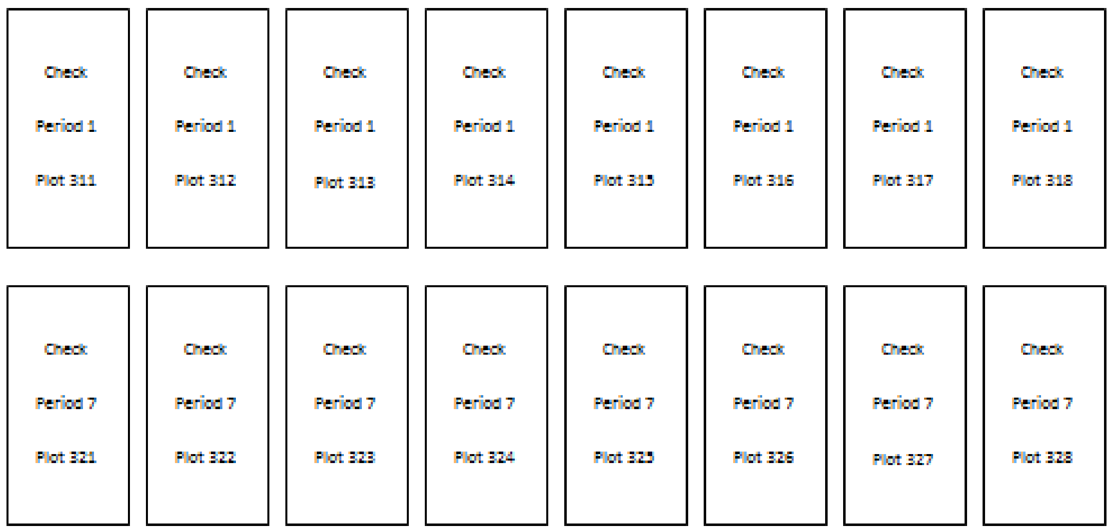

Figure 2. Schematic showing the plot layout and rainfall simulation period for the control plots

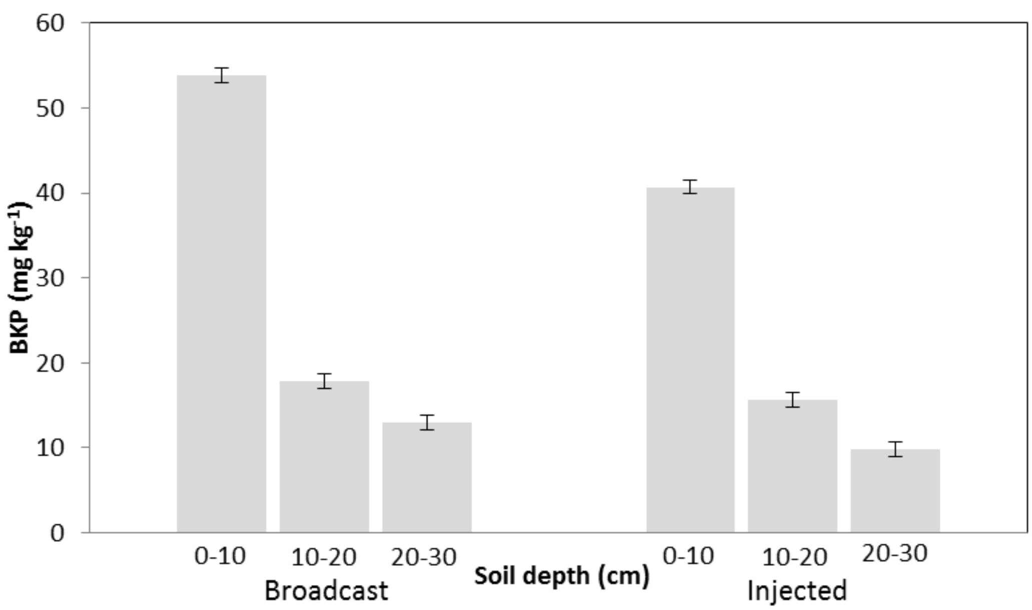

Figure 3. Bray and Kurtz No.1 P (BKP) as affected by soil depth for the broadcast and injected experimental treatments. Vertical lines represent the standard error of the mean value.

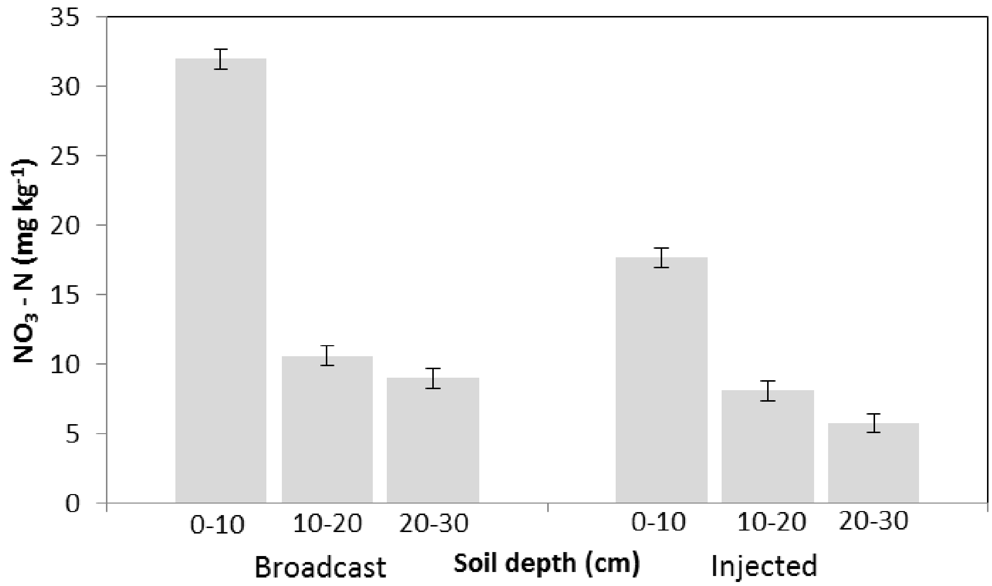

Figure 4. NO_3 - N as affected by soil depth for the broadcast and injected experimental treatments. Vertical bars represent the standard error of the mean value.

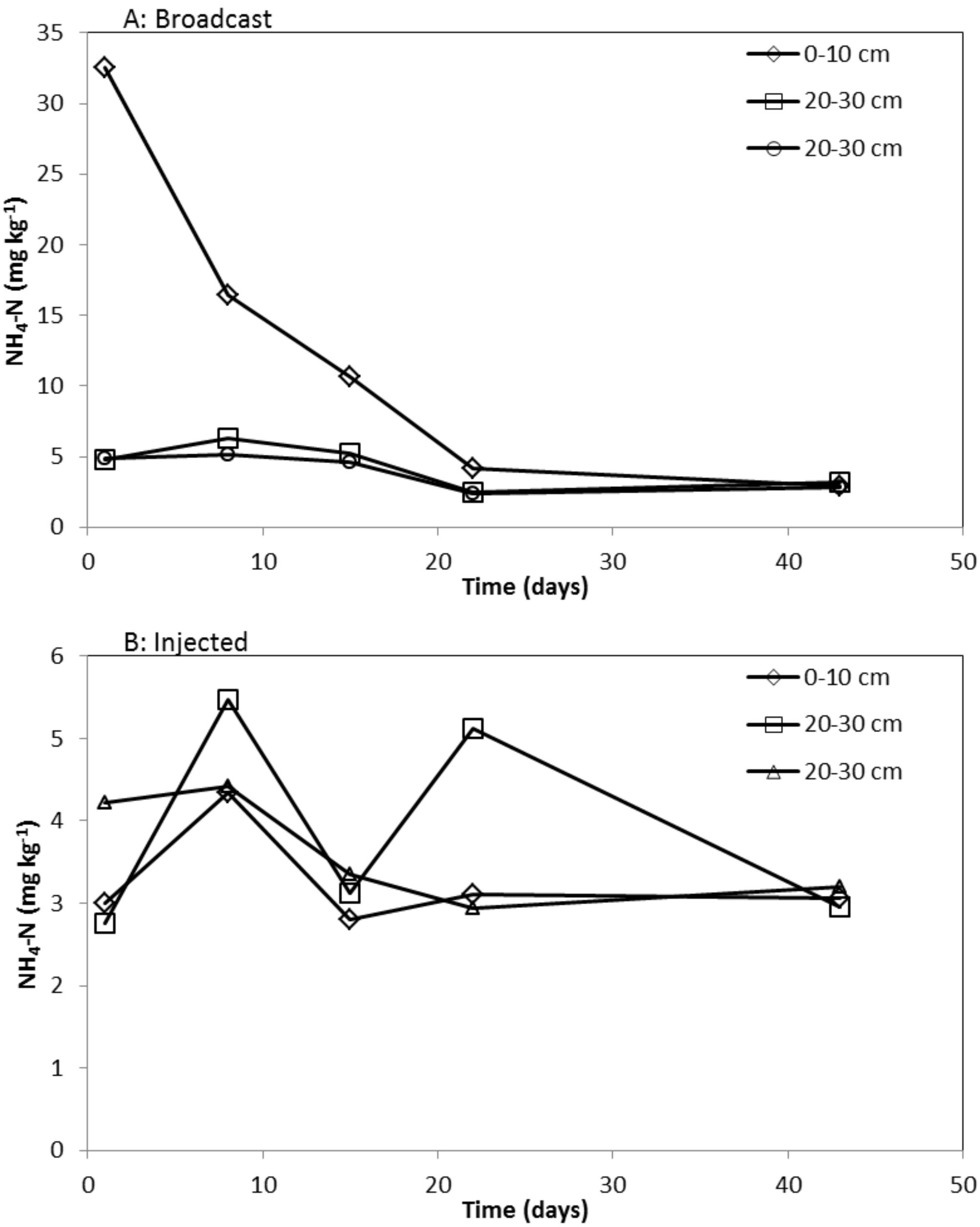

Figure 5. NH_4-N as affected by soil depth and time following slurry application for the broadcast and injected experimental treatments

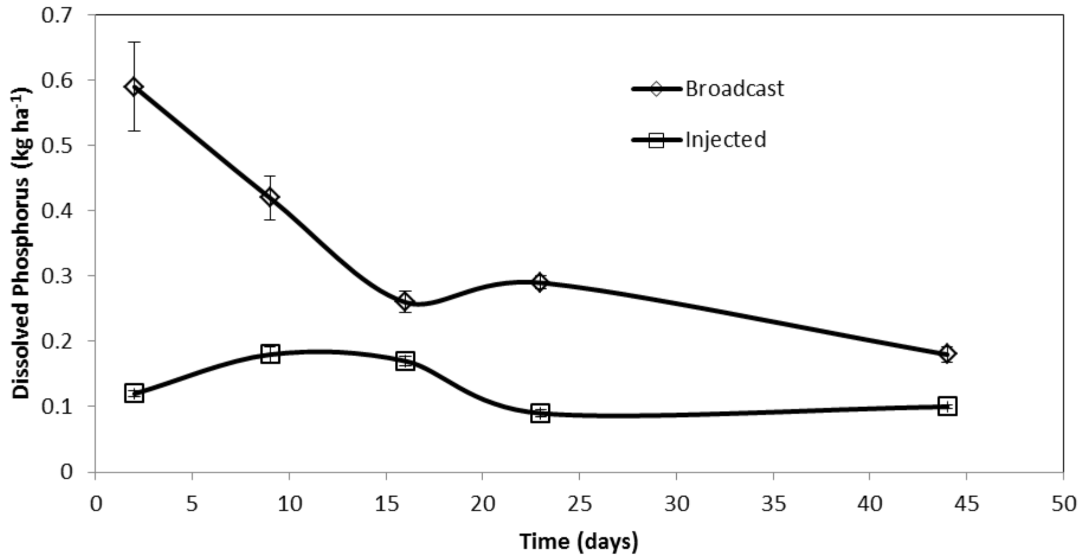

Figure 6. Dissolved phosphorus transport versus time since manure application for the broadcast and injected experimental treatments. Vertical bars represent the standard error of the mean value.

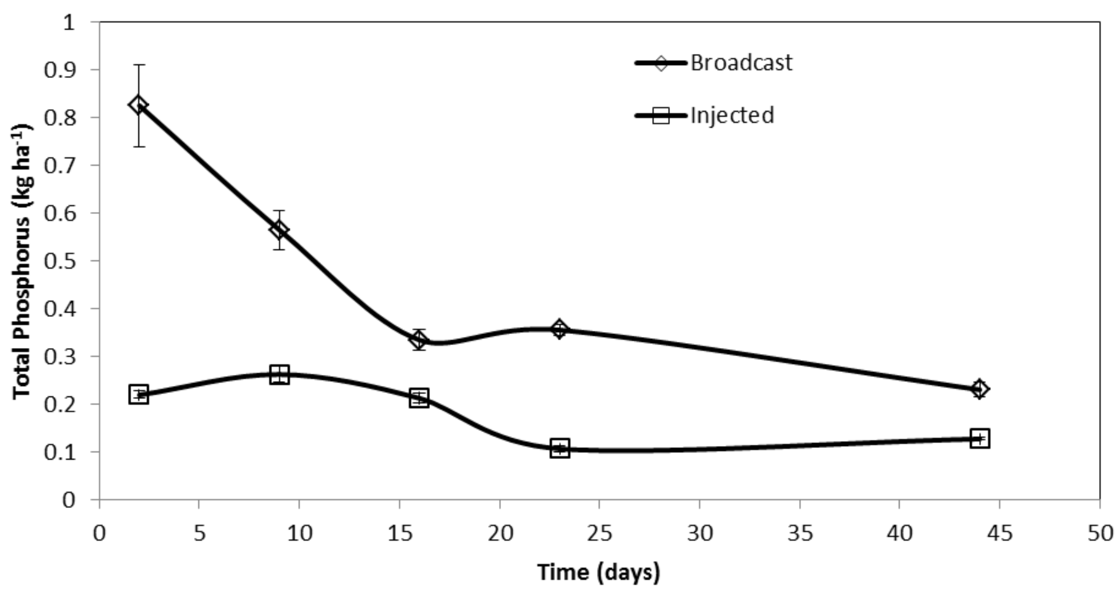

Figure 7. Total phosphorus transport versus time since manure application for the broadcast and injected experimental treatments. Vertical bars represent the standard error of the mean value.

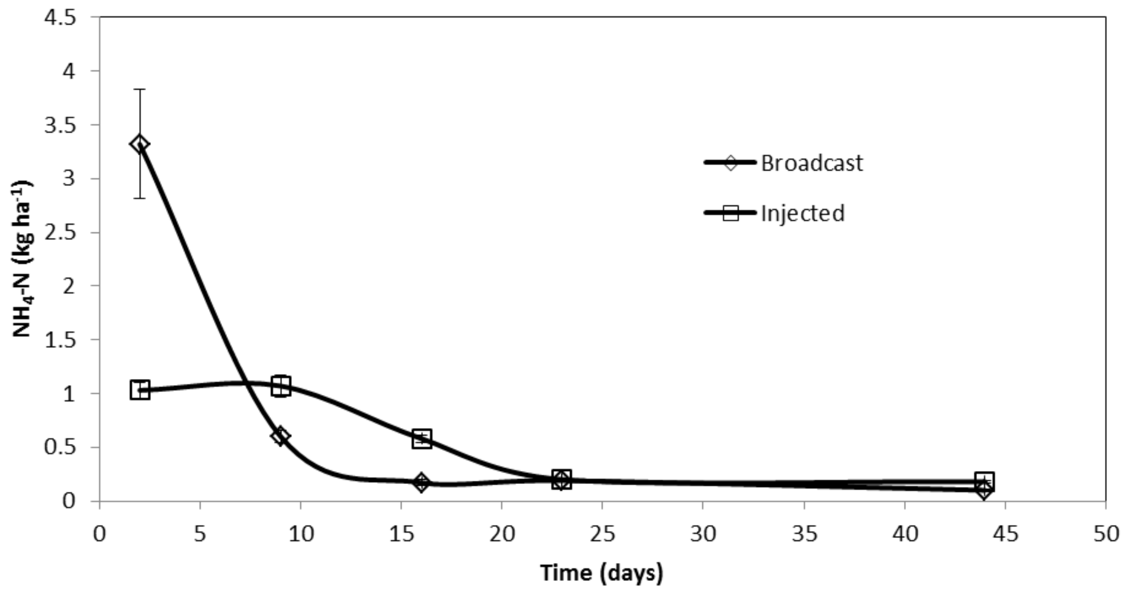

Figure 8. NH_4-N transport versus time since manure application for the broadcast and injected experimental treatments. Vertical bars represent the standard error of the mean value.

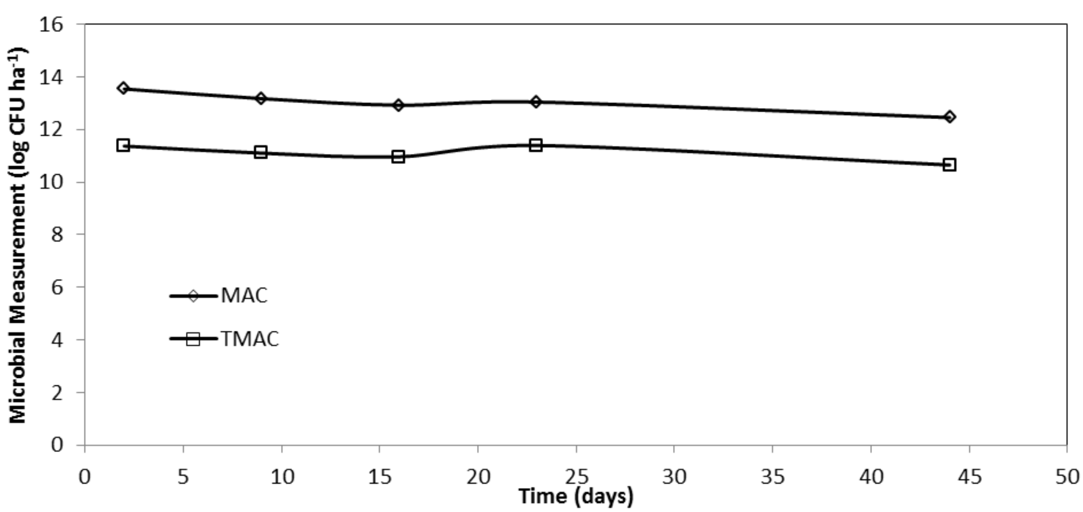

Figure 9. MacConkey agar bacteria count (MAC) and total aerobic microbial count (TAMC) versus time since manure application.

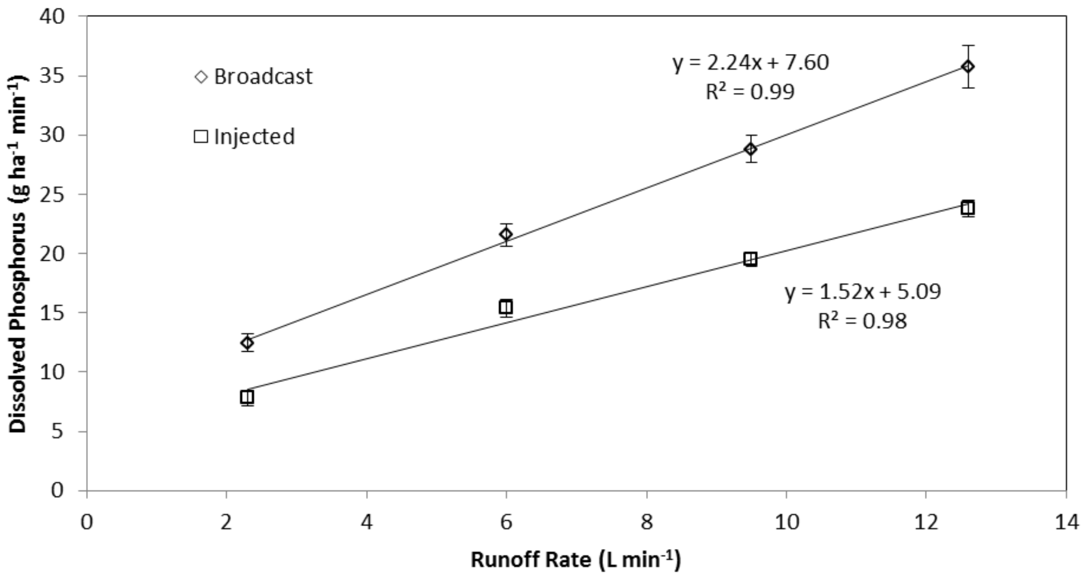

Figure 10. Dissolved phosphorus transport rate as affected by runoff rate for the broadcast and injected experimental treatments. Vertical bars represent the standard error of the mean value.

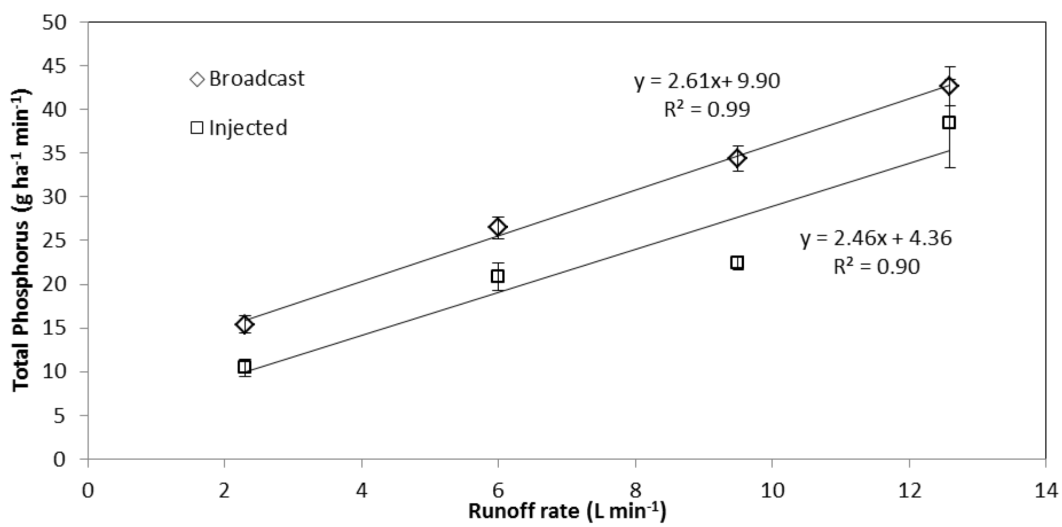

Figure 11. Total phosphorus transport rate as affected by runoff rate for the broadcast and injected experimental treatments. Vertical bars represent the standard error of the mean value.

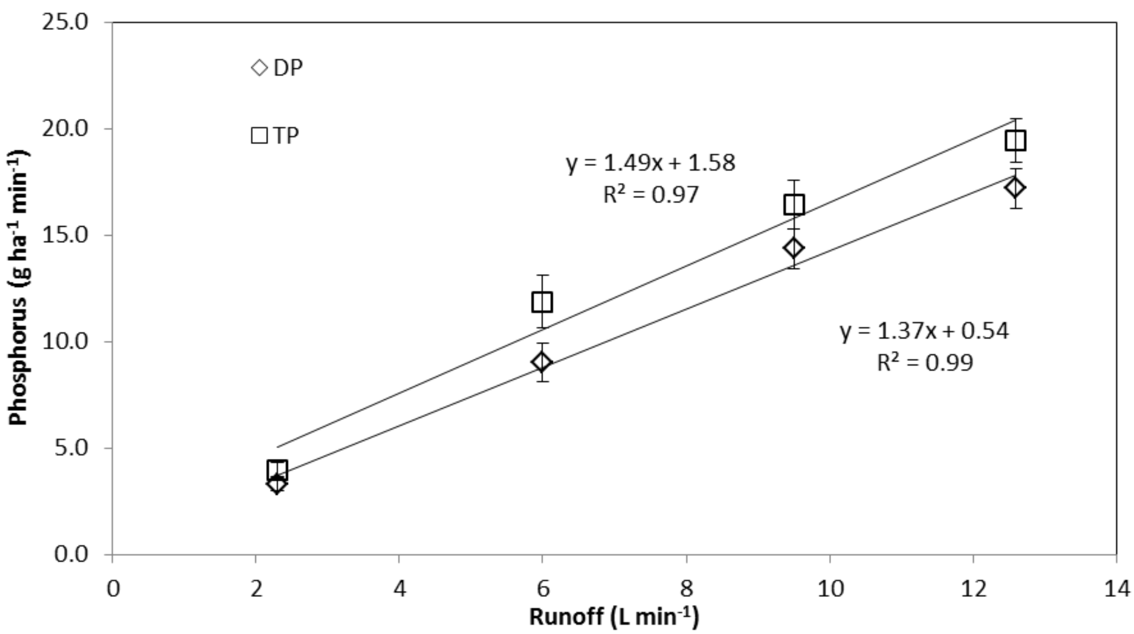

Figure 12. Dissolved phosphorus (DP) and total phosphorus (TP) transport rates as affected by runoff rate for the control plots.

Figure 13. Rogers Memorial Farm with field study site highlighted in red; North arrow located in bottom right corner

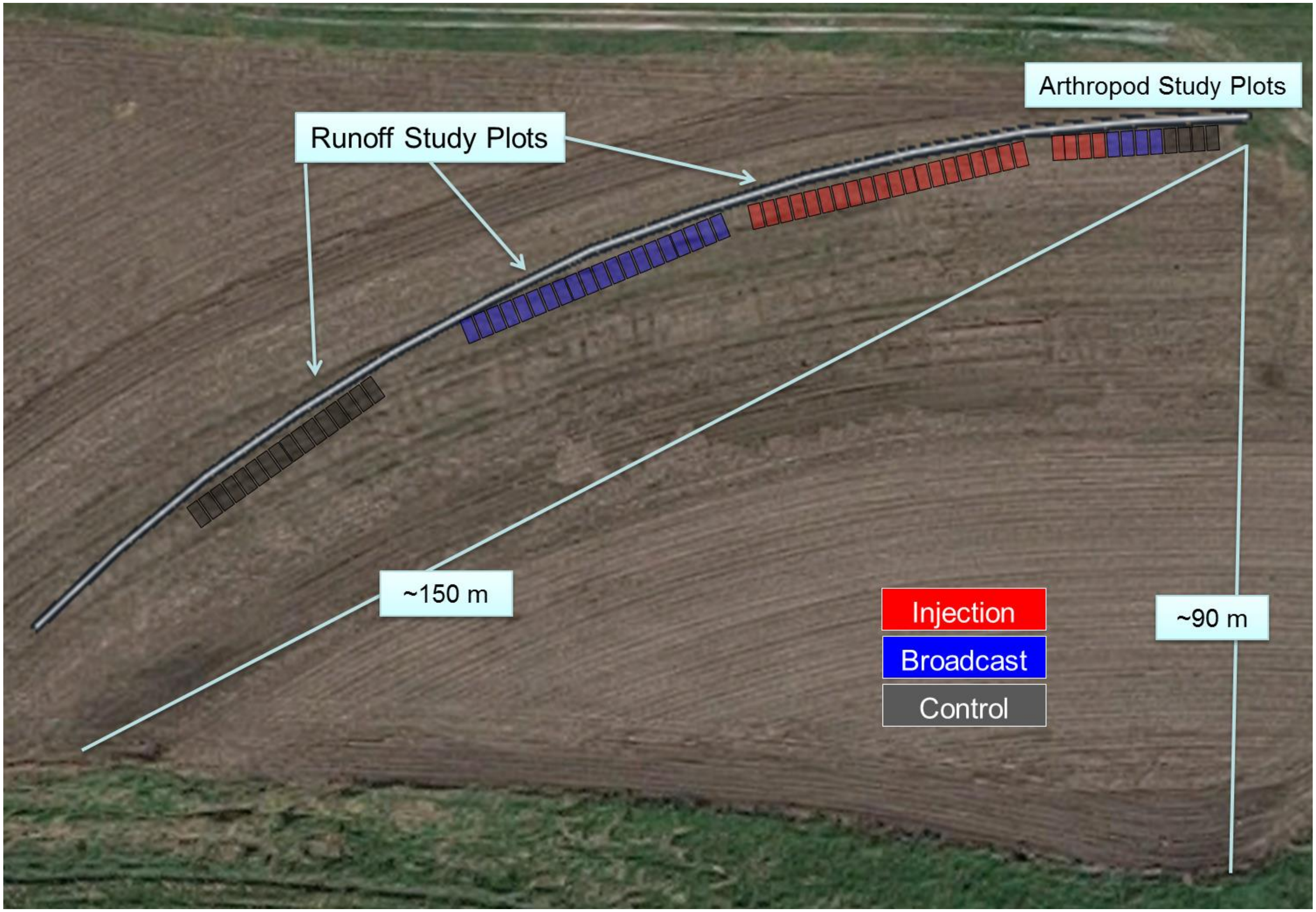

Figure 14. Aerial view of plot setup at Rogers Memorial Farm study site

Figure 15. Overhead view of plot setup looking in the direction of runoff.;Two rain gauges located on the right and left side of the plots and one located in the center between the plots

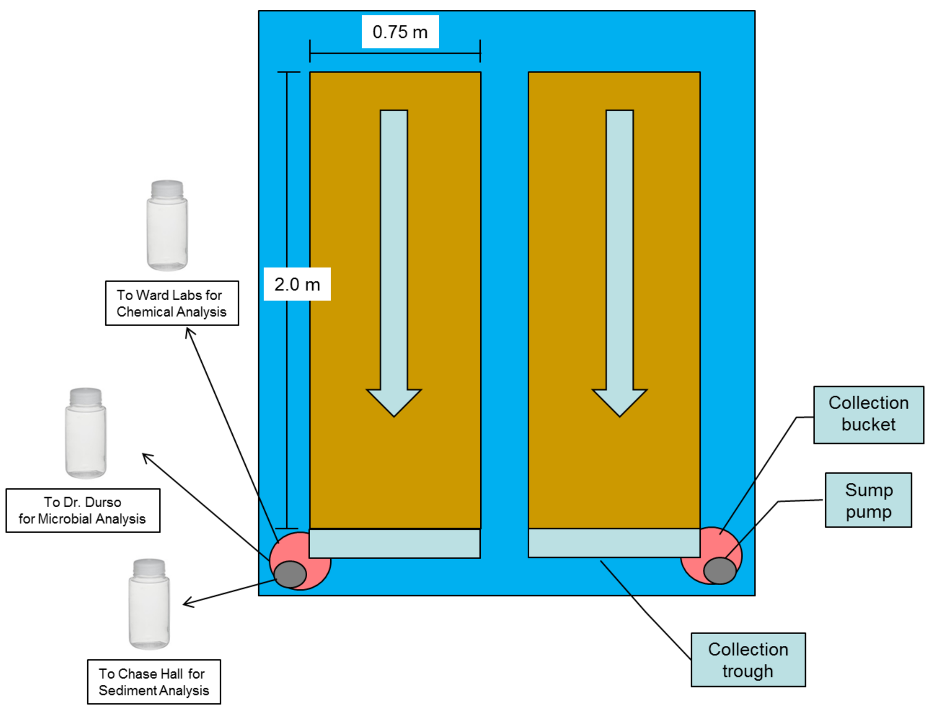

Figure 16. Rainfall simulation sample collection diagram

Table 1. Soil characteristics as affected by time and soil depth for the control plots. [a]

		WSP [b] (mg kg^{-1})	BKP [c] (mg kg^{-1})	NO_3-N (mg kg^{-1})	NH_4-N (mg kg^{-1})	EC [d] (dS m^{-1})	pH
Time							
	1	2.3	22.9	5.6^{b}	3.8	0.26^{b}	6.48
	2	1.7	18.9	11.4^{a}	3.3	0.31^{a}	6.55
Soil Depth (cm)							
	0-10	3.8^{a}	36.3^{a}	13.2^{a}	3.4	0.37^{a}	6.83^{a}
	10-20	1.4^{b}	16.1^{b}	7.1^{b}	3.9	0.27^{b}	6.59^{b}
	20-30	0.9^{c}	10.3^{c}	5.3^{c}	3.3	0.21^{c}	6.12^{c}
ANOVA		Pr > F					
	Time	0.10	0.17	0.01	0.05	0.01	0.55
	Depth	0.01	0.01	0.01	0.17	0.01	0.01
	Time x depth	0.48	0.47	0.01	0.54	0.02	0.15

[a] Values in the same column with different superscripts are significantly different at the 0.05 probability level based on the LSD test.

[b] WSP = water soluble P

[c] BKP = Bray and Kurtz No. 1]

[d] EC = electrical conductivity; EC and pH were determined in 1:1 soil: water ratio.

Table 2. Soil characteristics as affected by application method, time since slurry application, and soil depth. [a]

		WSP [b] (mg kg^{-1})	BKP [c] (mg kg^{-1})	NO_3-N (mg kg^{-1})	NH_4-N (mg kg^{-1})	EC [d] (dS m^{-1})	pH
Application Method							
	Broadcast	2.1^a	28.3^a	17.1^a	7.3^a	0.41	6.57
	Injected	1.8^b	22.1^b	10.5^b	3.6^b	0.41	6.66
Time (days)							
	1	1.9^b	23.6^b	9.7	8.7^a	0.35^b	6.61
	8	2.6^a	33.4^a	14.2	7.0^a	0.43^a	6.69
	15	1.8^b	23.3^b	15.1	4.9^b	0.45^a	6.66
	22	1.7^b	22.2^b	13.4	3.3^{bc}	0.40^{ab}	6.60
	43	1.7^b	23.4^b	16.8	3.0^c	0.42^a	6.52
Soil depth (cm)							
	0-10	3.8^a	47.3^a	24.8^a	8.3^a	0.59^a	6.91^a
	10-20	1.1^b	16.8^b	9.3^b	4.1^b	0.35^b	6.69^b
	20-30	0.8^b	11.5^c	7.3^b	3.8^b	0.29^c	6.24^c
ANOVA		Pr > F					
	Application	0.03	0.01	0.01	0.01	0.88	0.08
	Time	0.01	0.01	0.07	0.01	0.02	0.26
	Depth	0.01	0.01	0.01	0.01	0.01	0.01
	Application x time	0.28	0.26	0.93	0.01	0.01	0.01
	Application x depth	0.02	0.01	0.01	0.01	0.01	0.02
	Time x depth	0.12	0.73	0.17	0.01	0.08	0.13
	Application x time x depth	0.92	0.84	0.96	0.01	0.82	0.01

[a] Values in the same column followed by different letters are significantly different at the 0.05 probability level based on the LSD test.

[b] WSP = water soluble P.

[c] BKP = Bray and Kurtz No.1 P

[d] EC = electrical conductivity; EC and pH were determined in 1:1 soil: water ratio.

Table 3. Effects of slurry application method and time since application on selected water quality parameters.

	DP (kg ha^{-1})	PP (kg ha^{-1})	TP (kg ha^{-1})	NO_3-N (kg ha^{-1})	NH_4-N (kg ha^{-1})	TN (k g ha^{-1})	EC (ds m^{-1})	pH	Runoff (mm)	Erosion (Mg ha^{-1})	MAC (log CFU ha^{-1})	TMAC (log CFU ha^{-1})	TMAC/ MAC %
Application method [a]													
Broadcast	0.35^{a}	0.11^{a}	0.46^{a}	5.11	0.886	6.22	0.82^{a}	8.13	22	0.32^{b}	12.99	11.06	1.18
Injection	0.13^{b}	0.06^{b}	0.19^{b}	5.95	0.61	6.77	0.79^{b}	8.11	23	0.42^{a}	13.04	11.11	1.18
Time (days)													
2	0.35^{a}	0.17^{a}	0.52^{a}	4.83	2.17^{a}	7.28	0.83	8.18^{a}	26	0.41^{a}	13.54^{a}	11.37^{ab}	0.67
9	0.30^{ab}	0.11^{b}	0.41^{a}	5.85	0.84^{b}	7.16	0.79	8.11^{b}	24	0.24^{b}	13.17^{b}	11.10^{bc}	0.85
16	0.22^{bc}	0.06^{c}	0.27^{b}	6.14	0.38^{bc}	6.59	0.81	8.09^{b}	23	0.41^{a}	12.92^{c}	10.96^{cd}	1.09
23	0.19^{c}	0.05^{c}	0.23^{b}	5.60	0.20^{c}	5.81	0.80	8.10^{b}	21	0.46^{a}	13.04^{bc}	11.39^{a}	2.24
44	0.14^{c}	0.04^{c}	0.18^{b}	5.25	0.14^{c}	5.63	0.80	8.10^{b}	21	0.34^{a}	12.45^{d}	10.65^{d}	1.60
ANOVA							Pr > F						
Application	0.01	0.01	0.01	0.09	0.14	0.35	0.01	0.23	0.54	0.03	0.58	0.80	0.73
Time	0.01	0.01	0.01	0.46	0.01	0.27	0.09	0.01	0.38	0.03	0.01	0.01	0.11
Application x time	0.01	0.01	0.01	0.30	0.01	0.13	0.01	0.05	0.55	0.34	0.89	0.31	0.53

[a] Values in the same column followed by different letters are significantly different at the 0.05 probability level based on the LSD test.

Table 4. Correlation coefficients of runoff water quality characteristics with microbial characteristics[a]

Microbial Constituent	DP	PP	TP	NH_4	TN	NO_3	EC	pH
MAC [a]	-0.23 (0.06)	**-0.36 (0.01)**	**-0.26 (0.03)**	**-0.38 (0.01)**	**-0.19 (0.11)**	-0.01 (0.96)	**-0.32 (0.01)**	0.08 (0.48)
TAMC	**-0.33 (0.01)**	**-0.44 (0.01)**	**-0.36 (0.01)**	**-0.40 (0.01)**	-0.13 (0.28)	0.08 (0.52)	**-0.34 (0.01)**	**0.032 (0.01)**
TAMC/MAC	-0.11 (0.37)	-0.13 (0.28)	-0.12 (0.34)	-0.10 (0.43)	-0.07 (0.59)	-0.02 (0.85)	-0.10 (0.40)	0.19 (0.12)

[a] A correlation coefficient is significant at the 95% level if |correlation **shown in bold**| > 0.23 for n = 70.

[b] The value in parenthesis represents the Pr > |r|.

Table 5. Selected water quality parameters as affected by slurry application method, time since application, and runoff rate. [a]

		DP (g ha^{-1} min^{-1})	PP (g ha^{-1} min^{-1})	TP (g ha^{-1} min^{-1})	NO3-N (g ha^{-1} min^{-1})	NH4-N (g ha^{-1} min^{-1})	TN (g ha^{-1} min^{-1})	EC (ds m^{-1})	pH	Soil Loss (kg ha^{-1} min^{-1})
Application method [a]										
	Broadcast	24.6^{a}	5.1	29.7^{a}	818	19.6	902	0.71	7.88	40.3^{b}
	Injected	16.6^{b}	3.9	20.5^{b}	869	22.3	949	0.71	7.88	56.8^{a}
Time (days)										
	2	21.3	4.7	25.9	811	39.1^{a}	938	0.72^{a}	7.97^{a}	51.2
	9	25.2	7.2	32.3	979	31.3^{a}	1140	0.71^{b}	7.87^{b}	51.6
	16	19.9	4.7	24.5	834	14.9^{b}	849	0.70^{c}	7.83^{b}	45.7
	23	20.9	3.7	24.6	870	12.8^{b}	884	0.71^{b}	7.85^{b}	47.0
	44	16.0	2.4	18.3	722	6.6^{b}	819	0.71^{b}	7.88^{b}	47.5
Runoff rate (L min-1)										
	2.3	10.1^{d}	2.8^{b}	12.9^{d}	314^{d}	13.9^{c}	346^{d}	0.73^{a}	8.01^{a}	16.4^{d}
	6	18.5^{c}	5.2^{a}	23.7^{c}	699^{c}	21.3^{b}	770^{c}	0.71^{b}	7.90^{b}	43.1^{c}
	9.5	24.2^{b}	4.2^{ab}	28.4^{b}	1020^{b}	23.4^{a}	1130^{b}	0.70^{c}	7.82^{c}	59.4^{b}
	12.6	29.8^{a}	5.8^{a}	35.5^{a}	1341^{a}	25.1^{a}	1460^{a}	0.70^{c}	7.79^{d}	75.4^{a}
ANOVA		Pr>F								
	Application	0.01	0.27	0.01	0.53	0.57	0.61	0.46	0.58	0.01
	Time	0.24	0.08	0.11	0.39	0.01	0.21	0.01	0.01	0.93
	Runoff rate	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01
	Application x time	0.55	0.22	0.42	0.88	0.01	0.89	0.03	0.40	0.60
	Application x runoff rate	0.01	0.21	0.01	0.60	0.13	0.48	0.90	0.45	0.15
	Time x runoff rate	0.01	0.23	0.01	0.12	0.01	0.02	0.01	0.10	0.07
	Application x time x runoff rate	0.24	0.60	0.38	0.71	0.01	0.71	0.47	0.05	0.99

[a] Values in the same column followed by different letters are significantly different at the 0.05 probability level based on the LSD test.

Table 6. Effect of timing of the rainfall simulation tests on selected water quality parameters for the control plots.[a]

	DP (kg ha^{-1})	PP (kg ha^{-1})	TP (kg ha^{-1})	NO_3-N (kg ha^{-1})	NH_4-N (kg ha^{-1})	TN (k g ha^{-1})	EC (ds m^{-1})	pH	Runoff (mm)	Erosion (Mg ha^{-1})
Time										
1	0.06^{a}	0.02^{a}	0.07^{a}	2.60	0.02^{a}	2.97	0.71	8.21	15	0.27^{b}
2	0.04^{b}	0.01^{b}	0.05^{b}	2.51	0.01^{b}	2.77	0.72	8.21	14	0.46^{a}
ANOVA	Pr > F									
Time	0.03	0.04	0.03	0.72	0.01	0.52	0.06	0.88	0.53	0.01

[a] Values in the same column followed by different letters are significantly different at the 0.05 probability level based on the LSD test.

Table 7. Selected water quality parameters as affected by timing of the rainfall simulation test and runoff rate for the control plots. [a]

	DP (g ha^{-1} min^{-1})	PP (g ha^{-1} min^{-1})	TP (g ha^{-1} min^{-1})	NO_3-N (g ha^{-1} min^{-1})	NH_4-N (g ha^{-1} min^{-1})	TN (g ha^{-1} min^{-1})	EC (ds m^{-1})	pH	Soil Loss (kg ha^{-1} min^{-1})
Time									
1	9.9	2.7	12.6	704	3.4^{a}	795	0.70	8.02	30.6
2	12.1	1.3	13.4	754	1.4^{b}	811	0.70	7.91	45.1
Runoff rate (L min^{-1})									
2.3	3.3^{c}	0.7	4.0^{c}	190^{d}	0.6^{b}	210^{d}	0.71^{a}	8.09^{a}	10.7^{d}
6	9.0^{b}	2.9	11.9^{b}	533^{c}	1.5^{b}	58.4^{c}	0.70^{b}	7.98^{ab}	29.8^{c}
9.5	14.3^{a}	2.1	16.4^{ab}	951^{b}	3.8^{a}	1046^{b}	0.70^{b}	7.91^{b}	48.1^{b}
12.6	17.2^{a}	2.3	19.6^{a}	1242^{a}	3.7^{a}	1372^{a}	0.70^{b}	7.89^{b}	62.7^{a}
ANOVA									
Time	0.45	0.15	0.81	0.73	0.04	0.92	0.59	0.24	0.06
Runoff rate	0.01	0.39	0.01	0.01	0.01	0.01	0.01	0.01	0.01
Time x runoff rate	0.80	0.60	0.44	0.63	0.25	0.44	0.41	0.83	0.83

[a] Values in the same column followed by different letters are significantly different at the 0.05 probability level based on the LSD test.

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ABSTRACT

Soil arthropod abundance and diversity provide an indication of the biological quality of soil, which can impact soil fertility. Arthropods include insects, crustaceans, arachnids, myriapods, and scorpions and nearly every soil is inhabited by many different arthropod species. Row-crop soils may contain several hundred species (USDA-NRCS, 1999). Arthropods in the soil environment play a significant role in nutrient cycling and soil structure maintenance. One particular arthropod sub-Class, Acari (mites), can have a significant impact on nutrient release in soil, while Collembola are one of many arthropods that serve an important role as a link in the middle of the food web, acting as both predator and prey. For this study, the impact of swine manure slurry applied via broadcast and injection at a rate designed to meet the agronomic nitrogen needs of corn was investigated to determine the impact of manure application method

on soil arthropod abundance and diversity. Treatments included broadcast swine slurry, injected swine slurry, and non-manured control plots with four replications per treatment. Plots were monitored following manure application in June 2014 for a period of one year. Soil samples measuring 20 cm in diameter and 20 cm in depth were removed 4 d prior to manure application and at 1, 2, and 4 weeks and monthly thereafter. Arthropods were extracted by use of Berlese funnels and collected species were sorted and characterized. Application method had a significant effect on the collembolan populations, with much greater increases in the broadcast plots than the injection plots, particularly for Hypogastruridae and Isotomidae. Mite populations were not significantly impacted by manure application, regardless of method. However, Pseudoscorpions were significantly affected by application method. Time since application had a significant impact on nearly all arthropod populations throughout the yearlong study; however, this variability was likely a result of seasonality rather than time since manure application.

Keywords.

Acari, arthropods, Collembola, land application, manure management, nutrients, organic matter, soil health, soil properties, swine manure

INTRODUCTION

Agricultural soil health is a complex topic that lacks a simple, direct method of measurement, making it difficult to quantify or categorize. Instead of reporting only the physical and chemical soil characteristics as is typical in applications concerning the similar term 'soil quality,' soil health includes information about the biological health of the terrestrial environment, as well (Curell, Gross, and Steinke, 2012). However, quantification of the soil system's biological health is where the assessment of soil health becomes so complex.

Fortunately, nature has a unique ability to adapt to change that is nearly impossible for humans to recreate in such detail and complexity. Ecosystems have the inherent capability to detect and react to environmental changes including land-use patterns, anthropogenic climate change, and the management of industrial and agricultural wastes (Seebacher and Franklin, 2012). The soil food web, comprised of all organisms living part or all of their life in the soil, is dependent upon the sources of energy and carbon available for consumption by the organisms (USDA-NRCS, 1999). In agricultural soils where organic and inorganic nutrient applications are commonly applied to promote plant growth, organic matter and nutrients provided by these fertilizers serve as energy sources for organisms in the soil food web. The dependency of individual soil organisms upon interactions with each other for survival further contributes to the complexity of the soil ecosystem. Likewise, activities like tillage and fertilizer application that disrupt the soil environment trigger activity among soil organisms. Therefore, the natural reactions of an ecosystem to human-imposed environmental perturbations may reveal more about the condition of a system than any developed measurement technique. Namely, addition of organic matter to soil raises the proportion of active organic matter – the portion available to soil organisms – providing a useful food source that can support an increase in the population of soil organisms. For this reason, monitoring changes in abundance and diversity of soil organisms such as arthropods may reveal more information about the ecological health of a soil in response to external stimuli than common chemical and physical analyses that exhibit a much slower response.

According to Doran and Zeiss (2000), ideal biological indicators of soil health should fulfill several criteria: (1) sensitivity to variations in land management practices; (2) strong correlation to beneficial soil functions; (3) ability to provide information helpful for

understanding ecosystem processes; (4) proven comprehensibility and usefulness for land managers; and (5) simple and inexpensive measurement. In the terrestrial ecosystem, collection, analysis, and categorization of soil arthropods has proven to be an inexpensive and easily quantified method of gathering information about the biological response to anthropogenic changes to the environment (Parisi et al., 2005). Arthropods also show a strong degree of sensitivity to land management practices (Sapkota et al., 2012) and specific taxa within the arthropod phylum are considered to have a positive correlation to soil health (Parisi et al., 2005). These characteristics make soil arthropods exceptional biological indicators of soil health.

According to Kibblewhite et al. (2008), soil type, organisms and functions, carbon and energy, and nutrients all play an important role in soil health. Application of livestock manure to agricultural fields is a common method of maximizing crop production by improving soil fertility. Livestock manure contains valuable macro- and micro-nutrients and organic matter that offer substantial benefits to soil condition by improving water infiltration, reducing soil erosion potential, and increasing soil productivity (Eghball et al., 2004).

A system of classification called the QBS method ("Qualità Biologica del Suolo," or biological quality of soil) utilizes an eco-morphological index (EMI) score to determine the degree of soil adaptability of different arthropods in the soil (Parisi et al., 2005). The assigned EMI score reflects the particular arthropod taxon's relative soil adaptability based on a number of other factors including level of pigmentation, appendage and visual apparatus development, and size. Arthropod population abundance and diversity are an indication of the diversification of the community into niches. These niches are determined by the roles that each arthropod taxon plays in the terrestrial ecosystem including nutrient cycling, organic matter decomposition and

stabilization, and soil structure maintenance (Sparling, 1997; Torstensson, Pell and Stenberg, 1998; Elia et al., 2010; Kladivko and Clapperton, 2011).

The Acari and Collembola populations are the most abundant and diverse of the commonly represented soil arthropod Orders (Culliney, 2013). These and other arthropods in the soil environment serve as the links in the middle of the food chain, as they act as both predator and prey (Culliney, 2013; Booher et al., 2012). Arthropods, depending on species and ecosystem, are responsible for the breakdown of litter to more available forms for the soil microflora and microfauna. They also play an important role both directly and indirectly in the cycling of nutrients and improvement of soil structure (Kibblewhite et al., 2008).

As agricultural crop producers consider utilizing livestock manure as a soil amendment, the impact of swine manure on soil biological health may be a factor worth consideration. Therefore, this study focused on assessing the biological component of soil health, described in terms of soil arthropod population abundance and diversity, as impacted by swine slurry application method and time following application.

MATERIALS AND METHODS

STUDY SITE CHARACTERISTICS

This field study was conducted at Rogers Memorial Farm, located 18 km east of Lincoln, Nebraska (40°50’38.7”N, 96°28’07.1”W), from June 2014 through June 2015. The soil at the site is classified as an Aksarben silty clay loam (fine, smectitic, mesic Typic Argiudoll), containing 15% sand, 48% silt, 37% clay, 3.5% organic matter, 1.5% total carbon, and had a mean slope of 10% (Kettler et al., 2001). The site has been operated under a no-till management

system using a crop rotation of corn (*Zea mays* L.), grain sorghum (*Sorghum bicolor* (L.) Moench), soybean (*Glycine max* (L.) Merr.), and winter wheat (*Triticum aestivum* L. cv. Pastiche). The soil developed under prairie vegetation from loess deposits. The site was left undisturbed since October 2013 following a corn crop planted during the 2013 growing season. Manure has not been applied to the site since at least 1966. Herbicide (glyphosate) was applied as necessary to manage weed growth.

PLOT PREPARATION

Experimental treatments included two manure application methods (broadcast and injected) and control plots. Four 0.75 m x 2 m plots were assigned to each of the experimental treatments, resulting in a total of twelve plots established parallel to the slope in the direction of overland flow.

Swine slurry was collected just prior to field application from the deep pits of an 8000-head commercial wean-to-finish swine facility in north central Nebraska. Solids and nutrient analyses were conducted on the swine slurry samples at a commercial laboratory prior to land application. Mean measured values of NO_3-N, NH_4-N, TN, TP, water content, EC, and pH for the slurry were 3.91 g kg^{-1}, 0.0024 g kg^{-1}, 5.49 g kg^{-1}, 0.58 g kg^{-1}, 96.57%, 42.35 dS m^{-1}, and 8.0, respectively. The slurry was injected and broadcasted by a commercial operator on June 30, 2014. For injection, a v-shaped chisel (horizontal sweep) implement was used on a 6-row applicator for manure placement. For broadcast application, the operator lifted the injection apparatus above the soil while maintaining a constant speed and flow rate. The slurry for both treatments was applied at a rate of approximately 46,800 liters per hectare. Control plots did not receive any application of manure.

SOIL SAMPLE COLLECTION

Soil samples were collected two days prior to treatment applications, one and two weeks post-application of manure, and once per month, thereafter, throughout the study period. Samples were not collected during winter months when soil was frozen. The months excluded from the study included December 2014 and January, February, and March 2015.

Soil sample collection was comprised of two types of samples. Samples obtained with a 3.8-cm diameter soil probe were divided into 0-10 and 10-20 cm sections for each of the plots for nutrient analysis at a commercial laboratory. Samples measuring 20 cm in diameter and 20 cm in depth, resulting in a soil volume of 6,280 cm^3, were stored in plastic buckets with air holes in the lids, placed in coolers with ice packs, and transported to the University of Nebraska West Central Research & Extension Center in North Platte, Nebraska within 12 h of collection. These samples were then transferred to Berlese funnels for extraction of arthropods. Berlese funnels provide an arthropod extraction technique based on the principle that arthropods and other organisms that typically dwell in the dark soil environment will respond negatively to light and heat (MSU, 2006). The apparatus consists of a light situated above a soil-holding container whose base is comprised of a sieve and funnel. The sieve retains the soil particles, but is large enough to allow the soil arthropods to pass through and drop into the funnel, which then diverts the organisms into a container of ethanol solution. A 70% ethanol solution was used in this study to preserve the organisms for later analysis.

ARTHROPOD SAMPLE ANALYSES

The QBS method of classification was employed to assign an eco-morphological index (EMI) score on the basis of soil adaptability level of each arthropod Order or Family (Parisi et al., 2005). Preserved arthropods from each soil sample were manually identified and quantified using a Leica EZ4 stereo microscope (Leica Biosystems, Inc., Buffalo Grove, IL). Arthropods

were classified by Order with the exclusion of the collembolans, which were categorized by Family due to their substantial variation in soil adaptability characteristics (table 1). Members of the Order Coleoptera were individually assigned EMI scores based on specific attributes (table 2). Collembolan and coleopteran EMI scores were assigned according to suggestions by Parisi et al. (2005), as shown in tables 1 and 2, respectively.

STATISTICAL ANALYSES

The impacts of swine slurry application method (broadcast vs. injection) and time following manure application on soil arthropod populations and soil chemical characteristics was determined by performing tests of hypotheses for mixed model analysis of variance using the general linear model (GLM) procedure (SAS, 2012). The samples were tested for significant differences resulting from time and treatment, as well as for variations within the treatment samples. Following identification of any significant differences, the least significant differences (LSD) test was employed to identify specific differences among treatments. A probability level of <0.05 was considered significant.

The soil characteristics data were also tested with mixed model analysis of variance using the GLM procedure. The soil samples were tested for significant differences associated with timing of soil collection, application method, samples within treatments, soil depth, and for interactions among these factors. In the case of significant differences, the LSD test was used to identify those differences. A probability level of <0.05 was considered significant.

RESULTS AND DISCUSSION

SOIL CHARACTERISTICS

Soil characteristic values and interactions are shown in table 2. Mean measured soil concentrations of nitrate, phosphorus, potassium, calcium, magnesium, sodium, and sulfate for

the control plots where no slurry was applied were 10.2, 20.1, 3425.5, 527.6, 9.13, and 11.3 mg kg^{-1}, respectively. Mean electrical conductivity (EC), organic matter, cation exchange capacity, and pH values were 0.35 mmho cm^{-1}, 3.4%, 24.1 meq $100g^{-1}$, and 6.48, respectively for the control plots. For the injection plots, mean measured soil concentrations of nitrate, phosphorus, potassium, calcium, magnesium, sodium, and sulfate were 16.8, 18.4, 286.6, 3087.3, 670.6, 17.9, 9.9 mg kg^{-1}, respectively. The mean measured values for EC, organic matter, cation exchange capacity, and pH were 0.39 mmho cm^{-1}, 2.9%, 23.7 meq $100g^{-1}$, and 6.21, respectively for the injection plots. The mean measured soil concentrations of nitrate, phosphorus, potassium, calcium, magnesium, sodium, and sulfate for the broadcast plots were 15.2, 44.4, 411.5, 3181.1, 425.9, 11.3, and 11.8 mg kg^{-1}, respectively. Mean EC, organic matter, cation exchange capacity, and pH for the broadcast plots were 0.40 mmho cm^{-1}, 3.3%, 21.8 meq $100g^{-1}$, and 6.63, respectively.

Time since swine slurry application had a significant impact on all measured soil characteristics. Soil depth also had a significant effect on all measured soil characteristics. The measured values of magnesium, sodium, and cation exchange capacity increased with soil depth with values of 626.5 mg kg^{-1}, 13.33 mg kg^{-1}, and 24.26 meq $100g^{-1}$ at 4-8" compared to measurements of 456.2 mg kg^{-1}, 12.22 mg kg^{-1}, and 22.21 meq $100g^{-1}$ at 0-4" soil depths (table 3). However, measurements of pH, EC, organic matter, NO_3-N, P, K, Ca, and SO_4-S decreased with soil depth with values of 6.82, 0.44 mmho cm^{-1}, 3.5%, 18.7, 42.1, 408.5, 3348.0, and 12.79 kg mg^{-1}, respectively for 0-4" soil depths compared to 6.05, 0.32 mmho cm^{-1}, 2.9%, 9.5, 13.1, 280.0, 3114.6, and 9.21 mg kg^{-1}, respectively for the 4-8" soil depth range (table 3).

Manure application method had a significant impact on all measured soil characteristics except EC. Mean measured values of pH, organic matter, P, K, Ca, and SO_4-S were greater for

the broadcast plots with values of 6.63, 3.32%, 44.4, 411.5, 3181.1, and 11.79 mg kg^{-1}, respectively compared to 6.21, 2.89%, 18.4, 286.6, 3087.3, and 9.9 mg kg^{-1}, respectively for the injection plots. However, the mean measured soil values for the injection plots were significantly greater than those for the broadcast plots for NO3N, Mg, Na, and cation exchange capacity with values of 16.8, 670.6, 17.9 mg kg^{-1}, and 23.7 meq $100g^{-1}$, respectively for the injection plots compared to values of 15.2, 425.9, 11.3 mg kg^{-1}, and 21.8 meq $100g^{-1}$, respectively for the broadcast plots.

Application method x time interactions on the plots where swine slurry was applied were found for pH, EC, NO_3-N, phosphorous, K, Ca, Mg, Na, SO_4-S, and cation exchange capacity (table 3). Time x soil depth interactions were found for pH, EC, NO_3-N, phosphorous, Na, and SO_4-S. Application method x depth interactions were identified for pH, EC, organic matter, phosphorous, K, Ca, Mg, Na, SO_4-S, and cation exchange capacity. Application method x time x depth interactions were only found for pH. For the injection experimental treatment plots, soil samples were taken between injection slots. Therefore, the measured values for some soil characteristics would be expected to be less at those locations.

The measured pH values for both the broadcast and injection plots were significantly greater at the 0-4" soil depth than the 4-8" depth (figure 1). However, the mean pH for the broadcasted plots was significantly greater than the pH for the injection treatments at both the 0-4" and 4-8" soil depths (figure1). Measured soil phosphorous concentrations showed a similar trend with significantly greater concentrations at the 0-4" depth compared to the 4-8" depth for both the broadcast and injection application methods (figure 2). The broadcast plots also had a significantly greater phosphorous concentration at both the 0-4" and 4-8" soil depths than the injection plots (figure 2). The concentration of NO_3-N was significantly greater at the 0-4" depth

for the injection, broadcast, and control plots, but the injection and broadcast plots had significantly greater overall NO_3-N concentrations than the control plots (figure 3). The broadcast and injection plots did not have significantly different NO_3-N concentrations.

Mean measured soil pH values were significantly higher for the broadcast plots than the injection plots (figure 4). Soil EC values were significantly different based on application method, time since application, and through application method x time interactions (table 3). All values were similar before applications and the control plots showed only small variation throughout the experimental period (figure 5). However, the broadcast plots showed a significant initial increase in EC and then gradually decreased over time (figure 5). The injection plots showed a trend similar to the control plots until approximately four months into the study, at which time the EC value increased significantly and then gradually decreased over time, as well (figure 5).

Concentrations of NO_3-N followed a similar trend to the measured EC values, with the broadcast values increasing quickly and then gradually decreasing over time, while the values for the injection plots did not increase significantly until approximately three months into the study after which time they gradually decreased for the remainder of the experiment (figure 6). Measured concentrations of phosphorous and organic matter were significantly greater for the broadcast plots than the injection plots throughout the study (figures 7 and 8).

Arthropod Population Analyses

Select arthropod taxa were analyzed for their response to application method and time since application on population density. The arthropod groups used for analysis were selected based on relative abundance in samples throughout the study compared to other taxa identified during the categorization process. The mean population counts for Hypogastruridae, Isotomidae,

Acari (mites), coleopteran larvae, Diplura, dipteran larvae, and Pseudoscorpions for the control plots were 10.7, 15.2, 42.2, 1.9, 1.3, 1.3, and 0.4 individual organisms per 6,280 cm^3 sample, respectively.

Application method had a significant impact on the Hypogastruridae, Isotomidae, and Pseudoscorpion populations. The Hypogastruridae and Isotomidae populations were significantly larger for the broadcast treatment with counts of 20.88 and 52.18, respectively, compared to 2.93 and 21.7 for the injection plots (table 4). The Pseudoscorpion population was significantly greater in the injection plots with a count of 1.43 compared to the 0.18 count for the broadcast plots (table 4). Time following slurry application had a significant impact on all of the measured arthropod populations with the exception of Pseudoscorpions. Application method x time following application interactions were identified for the Hypogastruridae and Isotomidae populations. QBS score was impacted only by time following slurry application.

The Hypogastruridae populations were significantly impacted by both application method and time. The injection, broadcast, and control plots' Hypogastruridae populations remained at a similar low population value until approximately 60 days into the study, at which time the broadcast Hypogastruridae populations increased significantly (figure 9). After the initial increase, the Hypogastruridae population reached its maximum at 150 days, and then returned to levels similar to those at the beginning of the study (figure 9). The Hypogastruridae populations in the injection plots remained at a much steadier, low level than the broadcast plots throughout the study (figure 9). The Isotomidae populations in the broadcast plots followed a similar trend to the Hypogastruridae, increasing rapidly after the first several months and then dropping off again after the winter (figure 10). The Isotomidae populations from the injection plots did increase somewhat after the first two months, but not as drastically as was observed in the broadcasted

plots (figure 10). Pseudoscorpion populations in the injection plots were significantly greater than those in the broadcast plots throughout the study (figure 11).

Daily temperature data for June 15, 2014 to July 15, 2015 for the nearest weather station to Roger's Memorial Farm is shown in figure 12. The significant impact of time on all but the Pseudoscorpion populations was most likely due to these normal seasonal temperature changes. Similar fluctuations with time were observed for the control plots (figures 9, 10, and 11). Arthropods are also sensitive to soil moisture, which fluctuates seasonally, as well. The daily precipitation data for June 15, 2014 to July 15, 2015 for the nearest weather station to Roger's Memorial Farm is shown in figure 13.

The rapid increase in the Hypogastruridae and Isotomidae populations in the broadcast plots about three months into the study may be a result of the increase in organic matter following manure application. The initial lag was most likely caused by the disturbance of the soil environment during slurry application. The injection plots may not have experienced the same increases due to the difference in method of application. Slurry injection requires the soil to be opened up by a chisel, so that the manure can be placed below the surface of the soil. The necessary soil disturbance in the injection process may cause a significant enough change in the habitat of these arthropods to deter their proliferation, even with the addition of nutrients and organic matter to the soil environment.

Overall arthropod community adaptation to soil dwelling was quantified using the QBS method. QBS score results are shown in table 5. Similar to results found in a study by Santorufo et al. (2012), the individual abundance of different arthropod groups, as opposed to the overall taxa richness, seemed to serve as a better indicator of changes in soil characteristics. Though it

has proven effective in a number of previous studies, the QBS score did not appear to follow a specific trend for either the treatments or the control plots for this application.

Collembolans are strongly sensitive to changes in pH (Ke et al., 2004). The broadcast plots offered a significantly higher pH environment of 6.63 than the injection plots at 6.21. The significant difference in pH between the treatment plots likely had a significant impact on the other analyzed arthropod populations, as well. Similar results were found in a study by Ke et al. (2004) in which collembolans showed preference to higher pH levels. In the Ke et al. (2004) study, collembolans were placed in environments containing a range of pH values and consistently moved from the lower to higher pH locations (typically preferring a pH of approximately 8).

The increase in collembolan populations is beneficial to soil health because of their positive impact on the soil environment. Collembola are one the most important building blocks in the soil, as they play a critical role in the middle of the soil food web. From the standpoint of the lower end of the food web, collembolans perform the step necessary for the breakdown of organic matter and litter to components that are available to smaller microorganisms who then convert the collembolan by-product into plant-available nutrients. In terms of the top of the soil food web looking down, collembolans are a large portion of the diet of many larger arthropods in the soil environment. Collembolans can play a critical role in plant production, as well. In addition to the ingestion of litter and organic matter, Collembola feed on mycorrhizae fungi and smaller microorganisms that are often crop pests. Mycorrhizae fungi dwell in plant root systems. The feeding of collembolans on mycorrhizae stimulates its growth, causing it to uptake organic P and N. The mycorrhizae then convert the organic P and N to plant-available inorganic forms in

exchange for C from the plant's roots (Jonas et al., 2007). Therefore, collembolans are considered highly beneficial to soil health due to their critical role in the soil food web.

The changes in soil characteristics resulting from the differences in manure application method likely played an important role in the changes in other arthropod populations, as well. All measured soil parameters were significantly affected by application method. The concentrations of NO_3-N, SO_4-S, and organic matter are critical to soil biology, as they provide food sources and energy to soil organisms. The availability of these sources appeared to significantly impact several of the populations of interest in this study.

CONCLUSIONS

Swine slurry application to agricultural fields serves as a beneficial fertilizer source that improves soil biological, chemical, and physical properties. Different methods of manure application and the timing of application result in varying changes to soil characteristics. This study investigated the effect of application method and timing on soil biological and chemical properties.

More significant changes in the chemical properties of the plots receiving broadcasted manure where observed in the initial portion of the study, while changes in the injection plots became apparent over time. Both time and treatment had a significant impact on all measured soil chemical parameters including pH, EC, organic matter, nitrogen, and phosphorous content.

The effect of application method and timing of application on the biological component of soil health was also investigated. The most significant responses to application method were found for the collembolan populations, specifically for Hypogastruridae and Isotomidae. However, Pseudoscorpions were also significantly affected by application method. Time since application had a significant impact on nearly all analyzed populations including

Hypogastruridae, Isotomidae, mites, coleopteran larvae, diplurans, and dipteran larvae. However, changes in population size with time were likely also heavily influenced by normal seasonal fluctuations in temperature and soil moisture content.

The utilization of swine slurry as a fertilizer source is beneficial to soil health, but requires consideration of application method, timing of application, and the combination of those two factors that will select for the most beneficial soil micro-organisms to the soil environment.

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APPENDIX

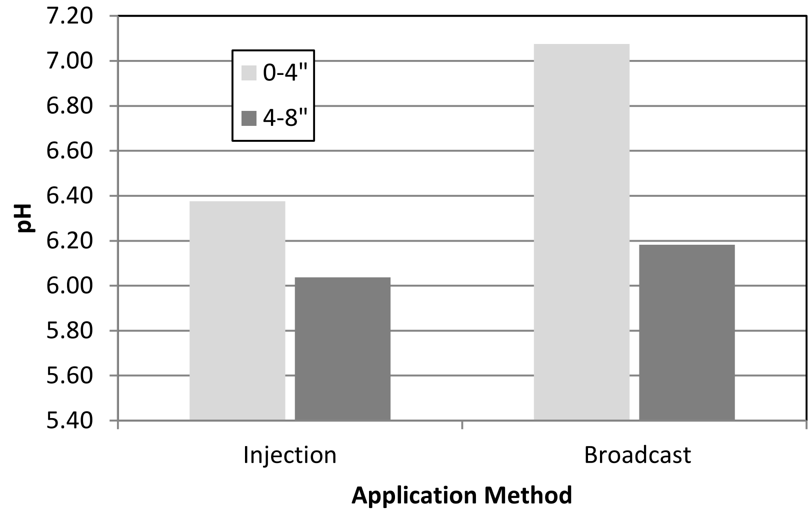

Figure 1. Mean soil pH as affected by soil depth for the injection and broadcast treatments

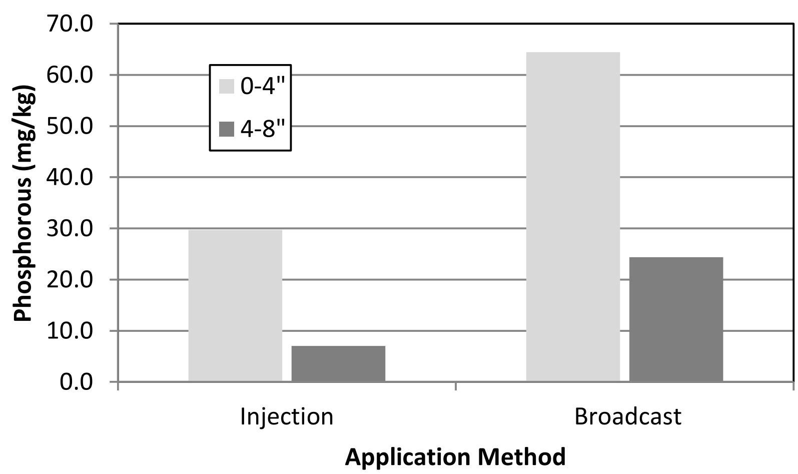

Figure 2. Mean phosphorus concentration as affected by soil depth for the injection and broadcast treatments

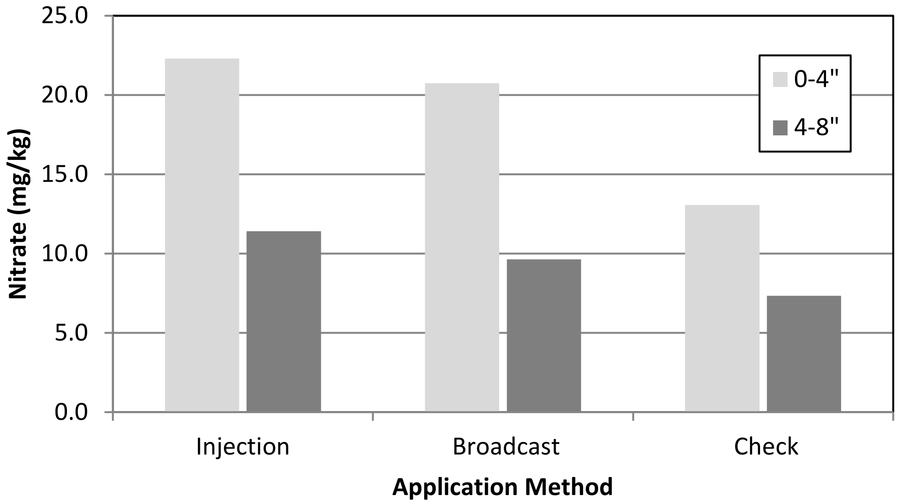

Figure 3. Mean NO_3-N concentration as affected by soil depth for the injection, broadcast, and control plots

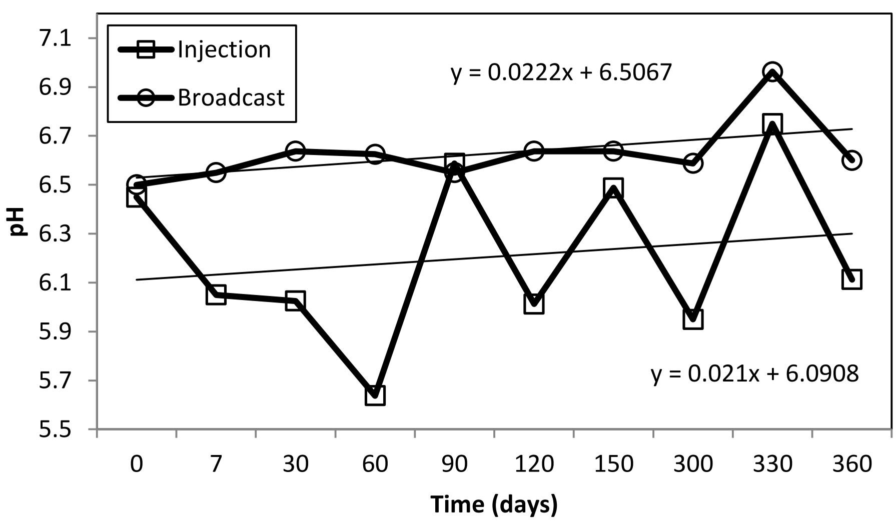

Figure 4. Soil pH as affected by time for the injection and broadcast experimental treatments

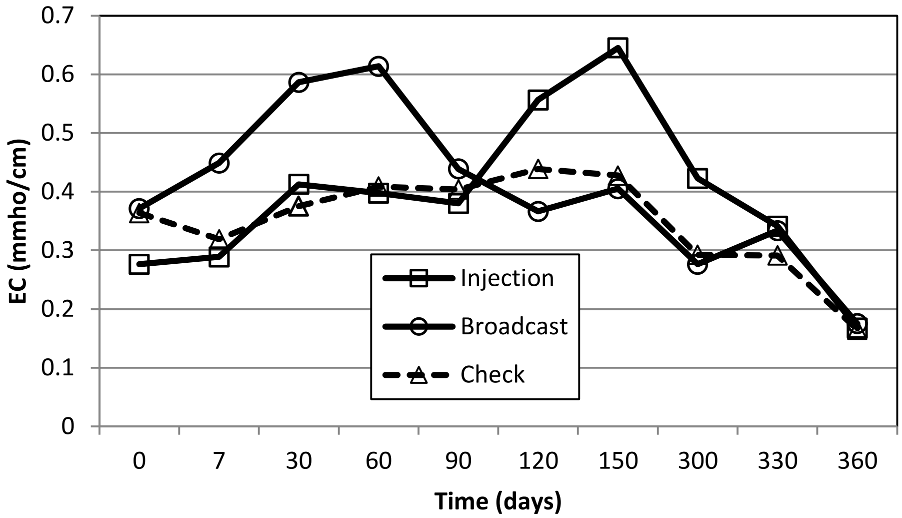

Figure 5. Electrical conductivity (EC) as affected by time for the injection, broadcast, and control plots

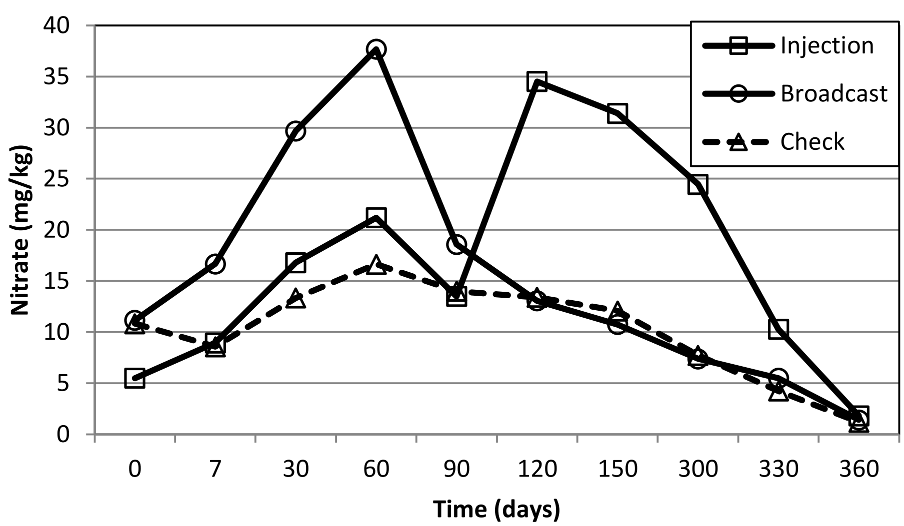

Figure 6. Nitrate-nitrogen (NO_3-N) as affected by time for the injection, broadcast, and control plots

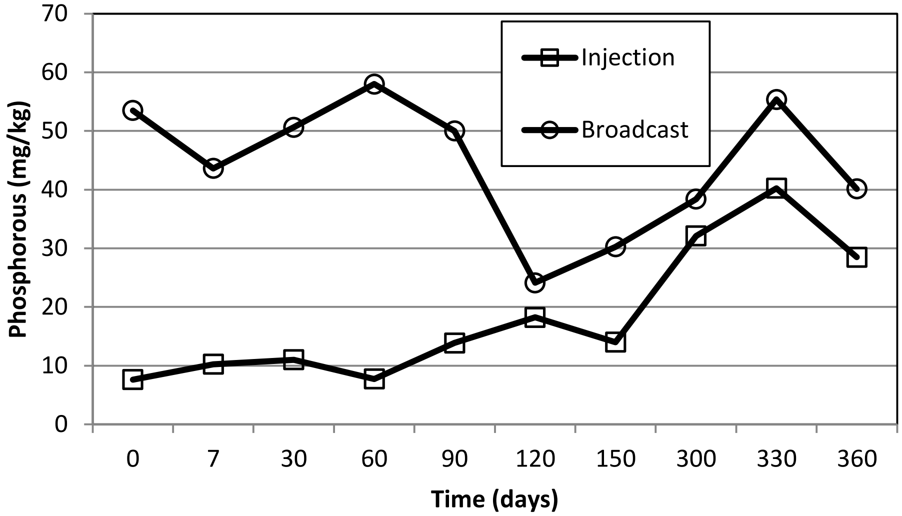

Figure 7. Phosphorous (ppm P) as affected by time for the injection, broadcast, and control plots

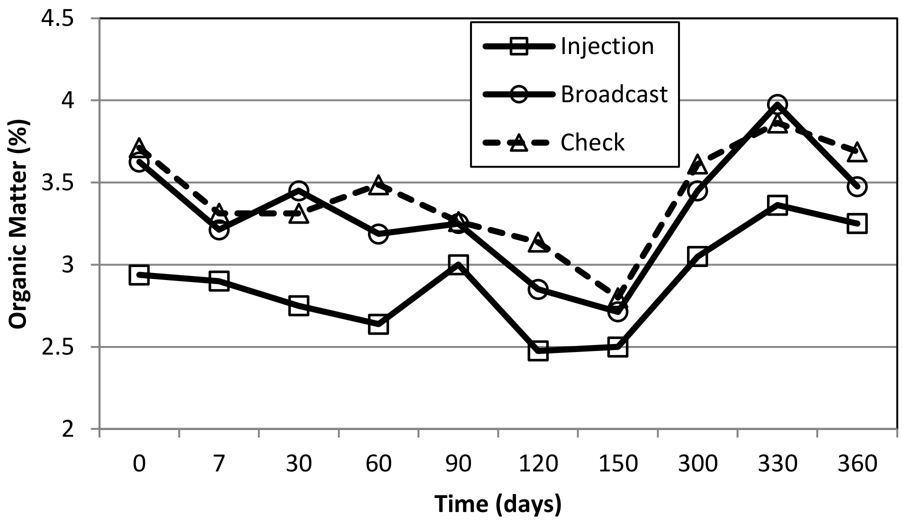

Figure 8. Organic matter content as affected by time for the injection[c], broadcast[b], and control[a] plots.

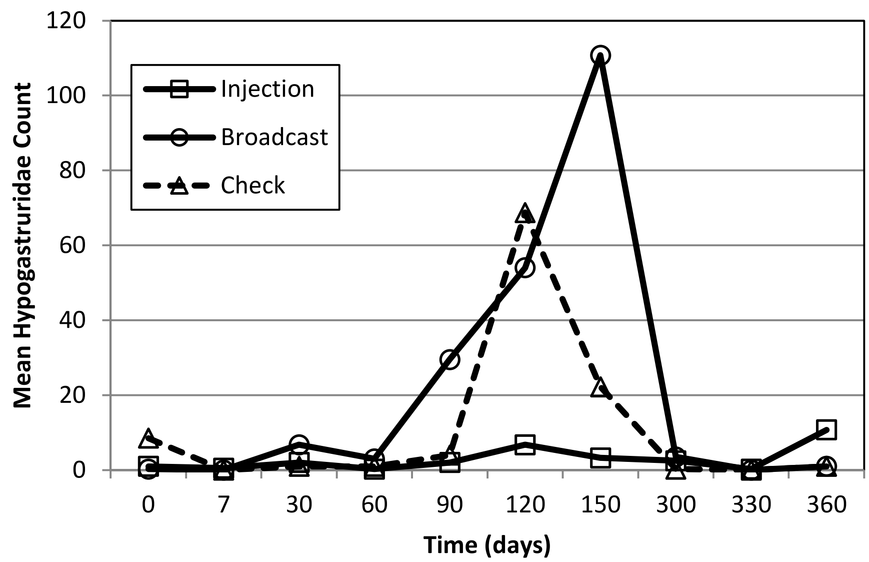

Figure 9. Mean Hypogastruridae count as affected by time since manure application for the injection[b], broadcast[a], and control[b] plots

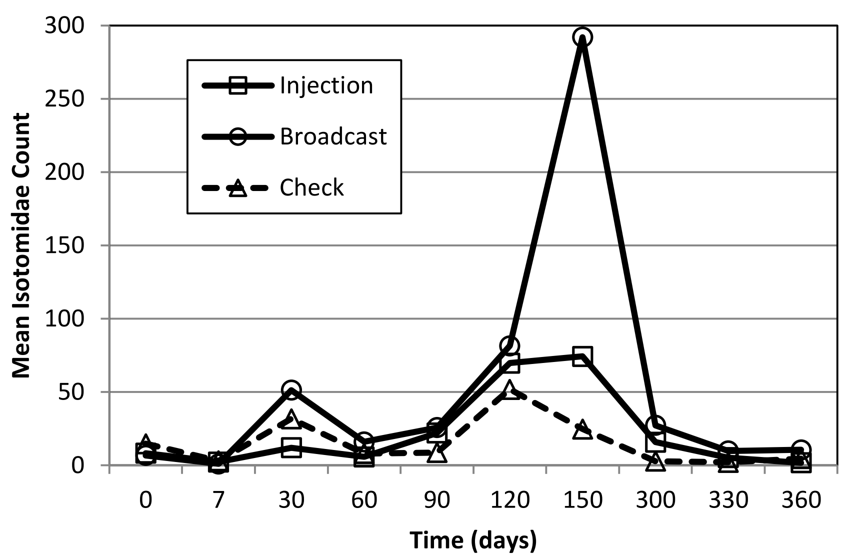

Figure 10. Mean Isotomidae count as affected by time since slurry application for the injection[b], broadcast[a], and control[b] plots

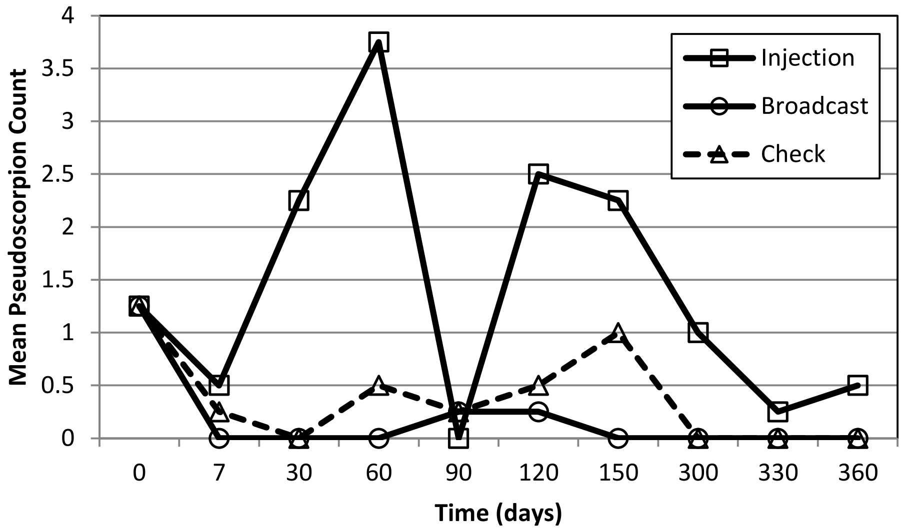

Figure 11. Mean Pseudoscorpiones count as affected by time since slurry application for the injection[a], broadcast[b], and control[b] plots

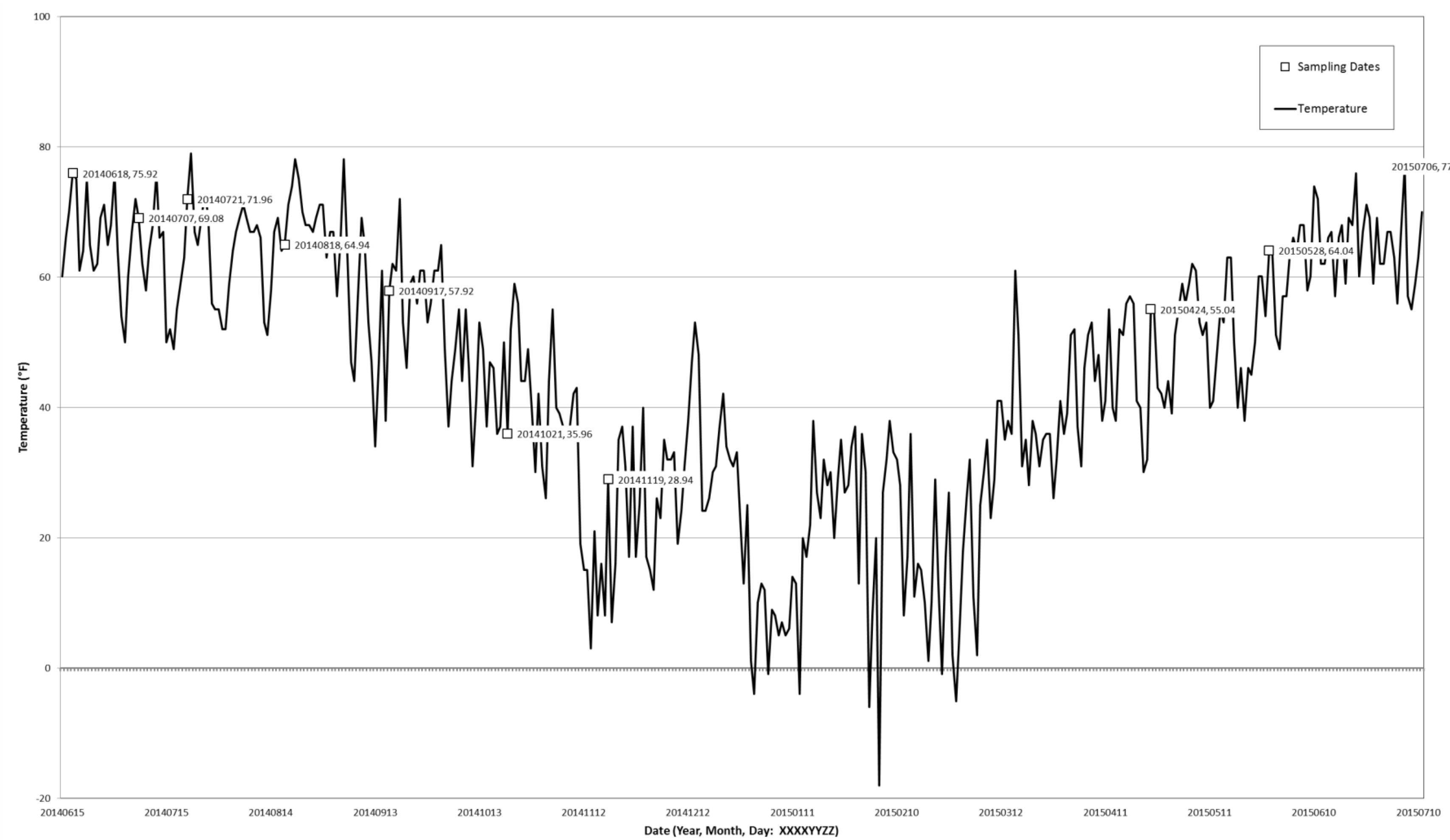

Figure 12. Daily dry bulb temperature data for June 15, 2014 to July 15, 2015 from the nearest weather station to Roger's Memorial Farm located at 40.841°, -96.514°.

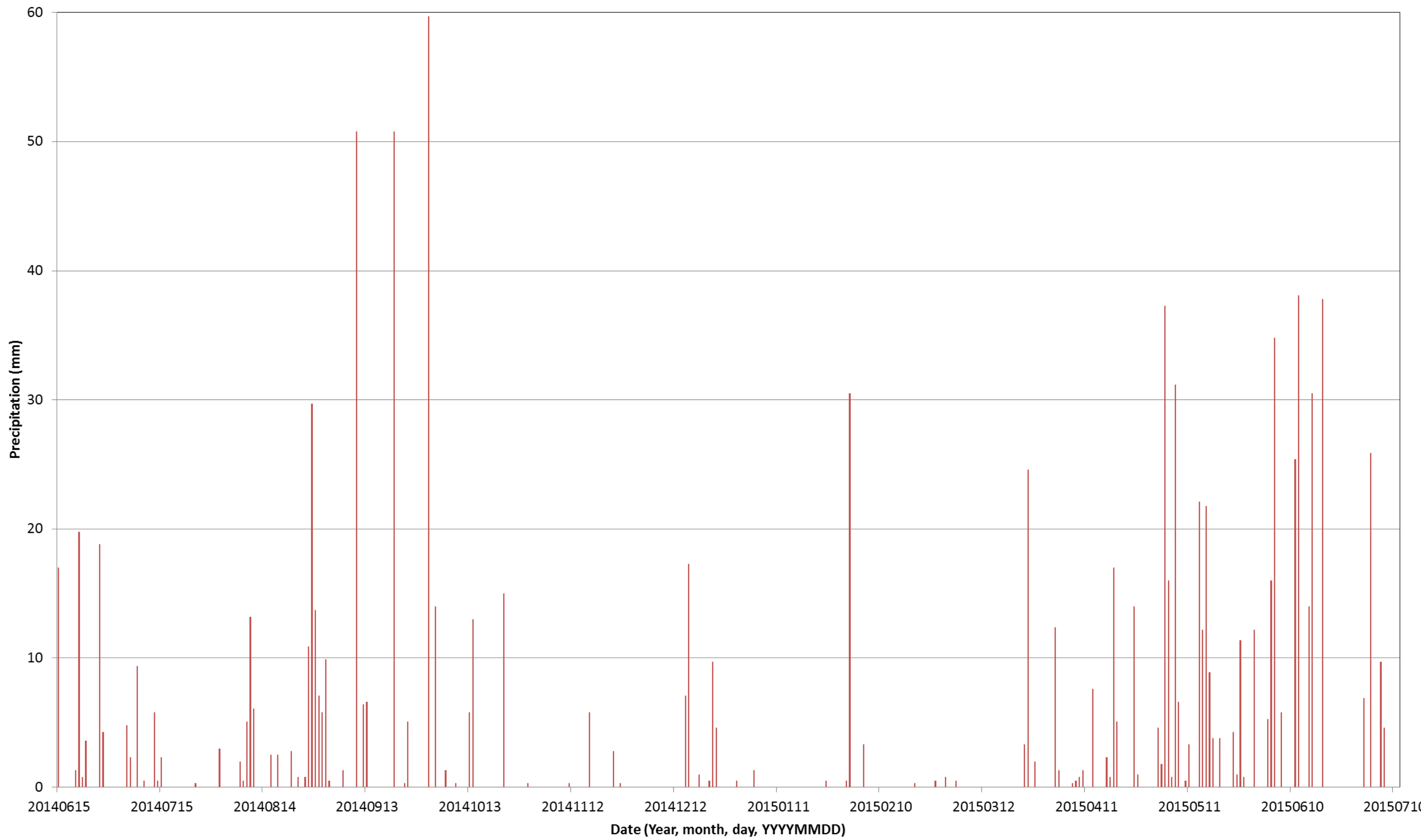

Figure 13. Daily precipitation data for June 15, 2014 to July 15, 2015 from the nearest weather station to Roger's Memorial Farm located at 40.841°, -96.514°.

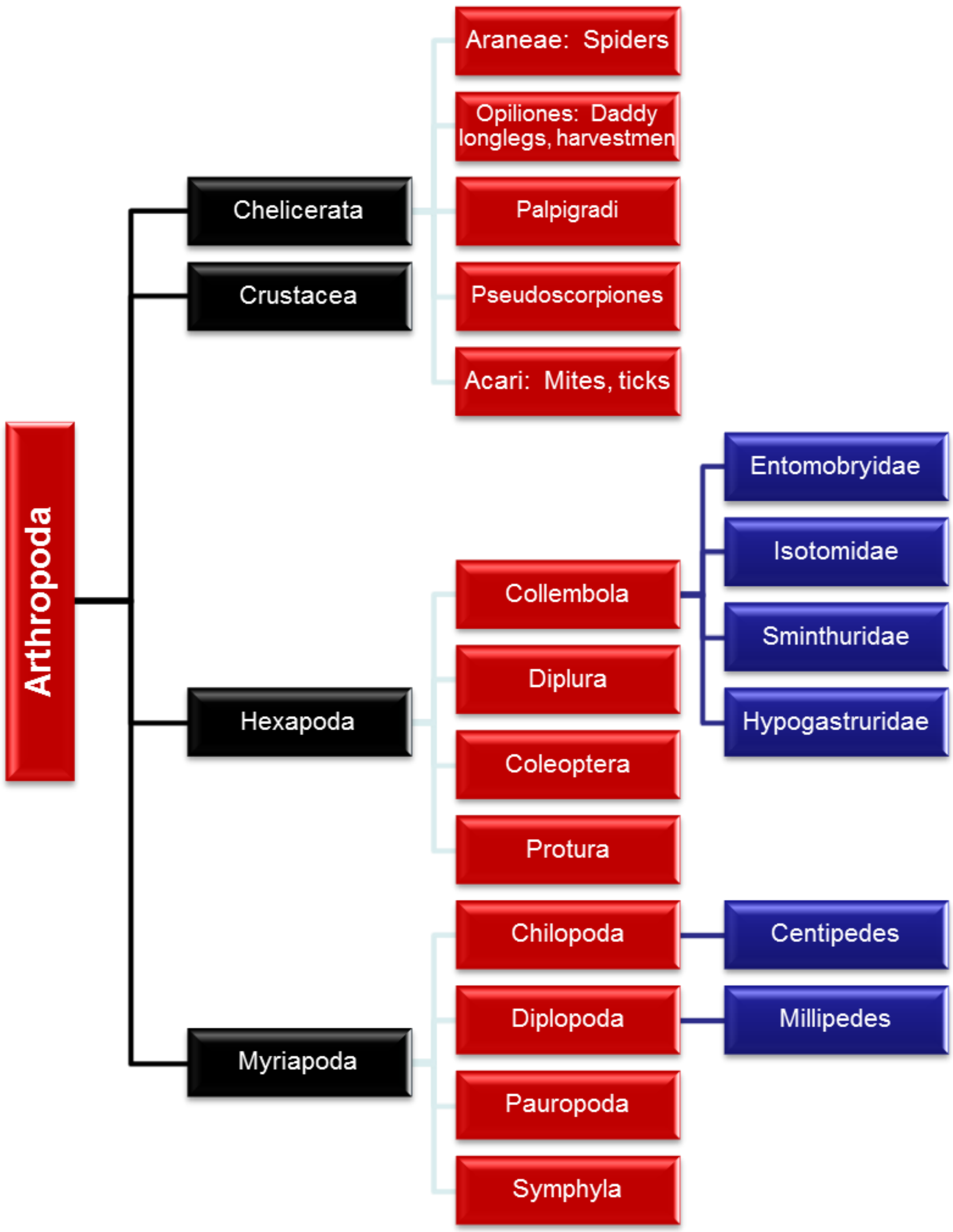

Figure 22. Arthropod taxonomy diagram.

Table 1. EMI calculation table for Collembola (from Parisi, 2001)

CHARACTERISTICS	EMI SCORE
Size	
big >3 mm	0
intermediate 2-3 mm	2
small < 2 mm	4
Pigmentation	
complex (e.g. Orchesella, Seira)	0
simple (e.g. Isotomurus, Tomocerus)	1
uniform (or limited to appendages, distally)	3
absent	6
Integument and associated structures	
great development of macro-chaetes &/or scales, presence of trichobothria	0
modest cover of integument	1
topographic specialization and reduced number of chaetes, particular sensilla on antennas, Post Antennal Organ present, AD present (not all these characters may be present)	3
scarce chaetes, sensors and particular structures present in various body parts	6
Anophtalmy	
8+8 ommatidia	0
6+6 ommatidia	2
from 5+5 to 1+1	3
no ommatidia	6
Antennas	
antennas much longer than head diagonal	0
ca. same length	2
shorter antennas	3
much shorter (often with particular sensilla)	6
Legs	
well-developed	0
intermediate	2
short	3
reduced or with lacking/reduced empodium, nail often without denticulation	6
Furca	
well-developed	0
intermediate	2
short with reduced number of setae	3
lacking mucron &/or alterations in manubria and teeth forms	5
Loss of furca or its reduction to a rudiment	6

Table 2. EMI calculation table for coleoptera (Parisi et al., 2005).

Characteristics	**EMI**
Clearly epigeous forms	1
Dimensions smaller than 2 mm	+4
Thin integument, often testaceous (tan-brown) color	+5
Hind wings highly reduced or absent	+5
Microphtalmy or anophtalmy	+5

Table 3. Soil characteristics as affected by swine slurry application method, timing of application, and soil depth.

	pH	EC (mmho/ cm)	Organic Matter (%)	NO_3-N (mg kg^{-1})	Phosphorus (mg kg^{-1})	K (mg kg^{-1})	Ca (mg kg^{-1})	Mg (mg kg^{-1})	Na (mg kg^{-1})	SO_4-S (mg kg^{-1})	CEC (me/100g)
<u>Application Method</u>											
Injection	6.21^{c}	0.39^{a}	2.9^{c}	16.8^{a}	18.4^{b}	286.6^{c}	3087.3^{c}	670.6^{a}	17.9^{a}	9.9^{b}	23.7^{a}
Broadcast	6.63^{a}	0.40^{a}	3.3^{b}	15.2^{a}	44.4^{a}	411.5^{a}	3181.1^{b}	425.9^{c}	11.3^{b}	11.8^{a}	21.8^{b}
Control	6.48^{b}	0.35^{b}	3.4^{a}	10.2^{b}	20.1^{b}	334.6^{b}	3425.5^{a}	527.6^{b}	9.1^{c}	11.3^{a}	24.1^{a}
<u>Depth</u>											
0-4"	6.82^{a}	0.44^{a}	3.5^{a}	18.7^{a}	42.1^{a}	408.5^{a}	3348.0^{a}	456.2^{b}	12.2^{b}	12.8^{a}	22.2^{b}
4-8"	6.05^{b}	0.32^{b}	2.9^{b}	9.5^{b}	13.1^{b}	280.0^{b}	3114.6^{b}	626.5^{a}	13.3^{a}	9.2^{b}	24.3^{a}
<u>Time</u>											
1 day	6.42	0.34	3.4	9.2	29.4	332.9	3,222.50	520	10.2	13.9	22.8
1 week	6.23	0.35	3.1	11.4	24.3	307.8	3,169.00	518.2	10.6	9.7	23.6
July	6.33	0.46	3.2	19.9	26.1	317.9	3,109.80	500.6	11.8	9.5	21.9
August	6.17	0.47	3.1	25.2	27	318.8	3,027.90	508.9	10.9	8.4	22.1
September	6.63	0.41	3.2	15.4	29.3	309.7	3,178.00	541	12.8	8.3	22.1
October	6.39	0.45	2.8	20.3	19.3	376.7	3,110.70	575.7	17.4	9.3	22.6
November	6.52	0.49	2.7	18.1	18.8	310.7	3,150.80	565.3	13.7	13.3	23.5
April	6.42	0.33	3.4	13.2	31.3	418.3	3,671.60	620.7	14.5	13.0	26.4
May	6.88	0.32	3.7	6.7	39.8	430.1	3,669.70	567.7	17.0	14.1	25.3
June	6.40	0.17	3.5	1.5	30.8	319.5	3,003.10	495.5	8.8	10.4	22.1
<u>GLM</u>	Pr > F										
trt	0.0027	0.0549	0.0001	0.0108	0.0003	0.0001	0.0113	0.0001	0.0001	0.0195	0.0252
sample(trt)	0.1599	0.0925	0.1404	0.1683	0.1935	0.0079	0.0011	0.0039	0.0479	0.0026	0.0074
time	0.0001	0.0001	0.0001	0.0001	0.0027	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001
trt*time	0.0028	0.0001	0.1231	0.0001	0.006	0.0067	0.0001	0.046	0.0001	0.0001	0.0175
sample*time(trt)	0.0001	0.2562	0.2641	0.2601	0.2136	0.0923	0.0517	0.0369	0.0672	0.0513	0.1231
sample*depth(trt)	0.0310	0.7996	0.4686	0.7281	0.1673	0.6406	0.9623	0.0355	0.8178	0.9644	0.2211
time*depth	0.0100	0.0085	0.4618	0.0001	0.0151	0.1346	0.4314	0.0964	0.0191	0.0002	0.1199
trt*time*depth	0.0416	0.6717	0.2197	0.5173	0.3889	0.7754	0.803	0.5877	0.2779	0.0715	0.3928
depth	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001	0.0089	0.0001	0.0001
trt*depth	0.0001	0.0200	0.0033	0.069	0.0317	0.0001	0.0001	0.0003	0.0001	0.0015	0.0013

Table 4. Selected arthropod orders and species as affected by application method and time since application.

	QBS Score	Hypogastruridae	Isotomidae	Mites	Coleoptera Larvae	Diplura	Diptera Larvae	Pseudoscorpions
Application Method								
Injection	59.88	2.93[b]	21.70[b]	45.55	1.03	1.30	0.73	1.43[a]
Broadcast	59.63	20.88[a]	52.18[a]	40.88	2.25	1.25	0.88	0.18[b]
Control	57.23	10.68[b]	15.20[b]	42.20	1.93	1.30	1.25	0.38[b]
Time (days)								
0	79.83	3.25	9.92	39.33	0.67	3.00	0.58	1.25
7	43.25	0.17	2.08	2.92	1.17	0.75	0.75	0.25
30	79.83	3.25	31.67	52.83	2.33	4.58	4.67	0.75
60	71.92	1.42	9.92	62.92	5.42	1.25	0.92	1.42
90	54.83	11.83	18.83	19.00	1.08	0.50	0.42	0.17
120	64.58	43.17	67.67	107.17	3.08	0.25	0.92	1.08
150	52.92	45.42	130.33	80.50	2.42	0.08	0.75	1.08
300	50.33	2.08	15.25	34.50	0.67	0.58	0.33	0.33
330	47.17	0.08	5.67	15.42	0.42	1.08	0.08	0.08
360	44.42	4.25	5.58	14.17	0.08	0.75	0.08	0.17
	Pr > F							
Application Method	0.8609	0.0016	0.0001	0.8828	0.3530	0.9800	0.7380	0.0030
Time	0.0001	0.0001	0.0001	0.0001	0.0290	0.0001	0.0190	0.1960
Application Method x Time	0.2687	0.0001	0.0001	0.1514	0.9140	0.0540	0.9460	0.5590

Table 5. QBS score over time by swine slurry application method

	QBS Score									
Time (days)	0	7	30	60	90	120	150	300	330	360
Treatment										
Injection	71.0	47.0	90.0	76.0	42.0	52.0	59.5	61.3	49.3	50.8
Broadcast	78.3	43.0	87.3	63.3	58.3	66.5	50.8	61.0	49.0	39.0
Control	90.3	39.8	62.3	76.5	64.3	75.3	48.5	28.8	43.3	43.5

The application of swine manure to agricultural fields serves as a beneficial amendment that improves the biological, chemical, and physical properties of the soil. When making recommendations to producers about how and when to apply manure to meet the agronomic needs of a crop, it is important to consider the impacts of swine slurry application method and the timing of application on the potential risk of runoff losses of nutrients and microbials, both of which can have detrimental impacts on the receiving surface water. It is also important to consider the impact of the slurry application method on soil fertility and biological health. Two field studies were conducted to determine the effect of manure application method and time following application on 1) the runoff concentrations of nutrients and microbials and 2) soil health as a function of soil arthropod abundance and diversity.

Injection application of swine slurry resulted in lower runoff concentrations of nutrients and microbials compared to broadcast application and non-manured control plots. However, the injection application method resulted in decreased soil pH and increased soil loss and erosion. The broadcasted slurry treatments yielded a lower rate of soil erosion and a significantly greater abundance of Collembolan populations in comparison to the injected and non-manured treatments. However, broadcast application produced greater runoff concentrations of nutrients and microbials. It was also determined that increasing overland flow rates significantly impacted runoff nutrient concentrations, water pH and EC, and soil loss. The impacts of overland flow were significant for both the injection and broadcast treatments.

The effect of application method and time since application on arthropod populations was also investigated. Application method had a significant effect on the collembolan populations, with a significantly greater abundance observed in the broadcast plots compared to the injection and control plots. The increase in collembolan population size may have been correlated to the increase in soil pH on the broadcast plots. Collembolans are particularly sensitive to changes in pH, preferring slightly higher pH levels (Ke et al., 2004). Pseudoscorpions were also significantly affected by application method. The injection plots were found to have a significantly greater Pseudoscorpion population than the broadcast or control plots. The smaller collembolan population and larger Pseudoscorpion population in the injection plots compared to the broadcast plots may have also had a correlation to Pseudoscorpion predation on collembolans.

Time following swine slurry application had a significant effect on all analyzed arthropod orders other than Pseudoscorpions, including Hypogastruridae, Isotomidae, Acari (mites), coleopteran larvae, Diplura, and dipteran larvae in both the treatment and control plots. Arthropod populations also varied significantly with time following manure application in the control plots, which would indicate that the differences in abundance were likely a function of seasonal soil temperature and moisture variations as opposed to the time since application of manure. Longer-term studies will provide an opportunity to substantiate or refute this assumption.

Swine slurry is a beneficial soil amendment that positively impacts soil arthropod abundance. Arthropods play an important role in soil structure maintenance, nutrient cycling and mobilization, and organic matter degradation and stabilization in the soil

environment. These processes contribute to improved water infiltration and soil moisture holding capacity as well as an increase in plant-available nutrients. While the application of swine slurry, regardless of application method, positively impacts soil health, it is important to consider the timing of application relative to precipitation occurrence and intensity to minimize the risk of nutrient and microbial transport to surface waters in runoff. Ideally, application of manure slurry at agronomic rates to meet the nitrogen needs of corn is most beneficial to the soil and least detrimental to surface receiving waters when performed at least 10 days prior to an anticipated precipitation event, particularly if the predicted rainfall intensity is greater than 70 mm/hr. From a practical standpoint, however, agricultural crop producers should be encouraged to utilize available manure application windows to manage and maintain capacity in manure storages while utilizing available weather prediction data to maximize time between manure application and impending precipitation.

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