

NUTRIENT AND BACTERIAL TRANSPORT FROM AGRICULTURAL LANDS FERTILIZED WITH DIFFERENT ANIMAL MANURES

Anurag Mishra

Thesis submitted to the faculty of Virginia Polytechnic Institute and State University in partial fulfillment of the requirements of the degree of

Master of Science
in
Biological Systems Engineering

Saied Mostaghimi, Chair
Greg Mullins
Theo Dillaha

Dec 4, 2003
Blacksburg, Virginia

Keywords: nutrients, phosphorus, nitrogen, indicator bacteria, fecal coliform, escherichia coli, enterococcus, rainfall simulator

© Copyright 2003, Anurag Mishra

Nutrient and Bacterial Transport from Agricultural Lands Fertilized with Different Animal Manures

Anurag Mishra

Abstract

The increase of animal agriculture coupled with excess manure production, and the reduced availability of land has led to the over application of animal manure on agricultural fields. The excessive application of manure is responsible for nutrient and bacterial pollution of downstream waterbodies. Manure application based on the crop phosphorus (P) requirements has been recommended as a viable method to reduce nutrient pollution. A plot scale study was conducted to measure the loss of nutrients and bacterial transport in runoff from cropland treated with poultry litter, dairy manure and inorganic fertilizer according to the P requirements of the crop.

Three simulated rainfall events were conducted 1, 2 and 35 days after planting of corn. Highest P and N concentrations were observed in the runoff from plots treated with poultry litter, followed by dairy manure and inorganic fertilizer. The poultry litter treated plots exhibited highest concentrations of bioavailable P in the runoff, compared to all other treatments. The P from poultry litter treated plots was also mostly in the soluble form, which underscores the need to control the runoff from cropland in order to decrease the P losses from the poultry litter treated fields. The edge of the field nutrient concentrations observed in this study were high enough to cause severe to moderate eutrophication problems in downstream waterbodies unless they are diluted. In general, nutrient concentrations were lower during the second simulated event, compared with those from the first event. A significant reduction in the nutrient concentrations of runoff was observed from the second to the third simulated event for all the treatments. This reduction was attributed to the loss of nutrients by natural rainfall-runoff events during the time period between the second and the third simulated rainfall event, plant uptake of nutrients, sorption and leaching processes.

The indicator bacteria analyzed in the present study were fecal Coliform (FC), Escherichia Coli (E.Coli) and Enterococcus (ENT). The bacterial concentrations reported in the runoff for the first and second simulated events were 10^4 to 10^5 times higher than the federal and state limits for primary contact recreation waters. No significant effect of treatments was observed on the bacterial concentrations in runoff. The highest concentrations were observed for FC, followed by ENT and EC in the runoff. The ratio of bacteria removed in runoff to the bacteria applied also

followed the above trend. The concentrations of bacteria generally increased from the first to second simulated event; unlike the nutrients. However, the bacterial concentrations dropped significantly from second to the third simulated rainfall event to the levels lower than those designated for primary contact recreation water limits. This reduction was attributed to the washing away of bacteria by the heavy rainfall-runoff events in the period between second and third simulated rainfall events and the die-off of bacteria.

The results reported from this study suggest that the manure application based on crop P requirements can also be a significant source of nutrient pollution and should be coupled with other best management practices (BMPs) also to reduce nutrient pollution. The results also suggest that the manure treated cropland can be a source for significant indicator bacterial pollution and appropriate BMPs are required to mitigate their effect.

Acknowledgement

I would like to thank my advisor Dr. Saied Mostaghimi, for his encouragement and support throughout this research work. Dr. Mostaghimi helped me to complete the work in time by reviewing my work even on weekends. I would also like to extend my acknowledgements to Dr. Theo Dillaha and Dr. Greg Mullins for their continuous support and resources during the research.

Imagining field work without Jan Carr and Jeff Wynn is impossible. Their hard working and humorous nature kept everything going even in unsupportive weather. Support of Michelle Soupir from plot set-up, until the completion of thesis greatly helped me complete the work on time. Long days of field work are the characteristics of this kind of plot scale studies. Support from Michael Johnston and Laura Teany, in the field was gratifying. Jae Pil Cho, Megan Laird, Christine Nelson, Adam Faulkner, Leigh Ann Henry, Marco Caiado, Matthew Habersack, Rachel Wagner, Tess Wynn, Robert Burgholzer worked during the long and hot summer days in field. Their extensive support made the daunting field work possible. I also appreciate the help from NSF students (summer, 2003) during the field works.

I would like to thank all the faculty and staff members for their help during the process of rainfall simulations. I am thankful to Julie Jordan and Katie for the sample analysis especially in the wee hours of the day. Dr. Christine Anderson Cock helped me a lot regarding the statistical design and result analysis. I want to thank Sudhanshu Mishra for helping me during my initial period in United States and supporting me throughout my research work. Ebrahim, Rakesh and Hitesh helped me a lot, especially in my difficult times and during my research work.

My parents deserve kudos for their patience and letting me study in United States, thousand of miles away from them. I would like to thank my fiancée Vineeta, who supported and encouraged me from miles away for my work.

Table of Contents

Abstract	ii
Acknowledgement	iv
Acknowledgement	iv
Table of Contents	v
List of Tables	vii
List of Figures	ix
List of Figures	ix
Chapter 1 Introduction	1
1.1 Goals and Objectives	3
Chapter 2 Literature Review	4
2.1 Water quality in U.S.....	4
2.2 Eutrophication.....	4
2.3 Manure.....	6
2.3.1 Properties of Manure.....	7
2.4 Nitrogen (N).....	7
2.4.1 N in Manure.....	9
2.4.2 Surface transport of N.....	10
2.5 Phosphorus (P).....	11
2.5.1 P and Its Forms.....	12
2.5.2 P in manure.....	13
2.5.3 P loading in surface runoff.....	13
2.6 Pathogen.....	15
2.7 Bacteria in animal waste.....	17
2.8 Fate and survival of bacteria.....	17
2.9 Die off of bacteria.....	19
2.10 Bacteria loading in surface runoff.....	20
2.11 Summary.....	21
Chapter 3 Materials and Methods	23
3.1 Site Selection.....	23
3.2 Site preparation.....	24
3.3 Plot preparation.....	25
3.4 Treatments.....	26
3.5 Rainfall simulation.....	31
3.6 Sampling Procedure.....	33
3.7 Additional Monitoring.....	34
3.8 Flow weighting the samples.....	34
3.9 Laboratory analysis.....	34
3.10 Statistical Analysis.....	36
Chapter 4 Results and Discussion	37
4.1 Hydrology.....	37
4.1.1 Canopy Cover.....	38
4.1.2 Rainfall.....	39
4.1.3 Runoff.....	39
4.1.4 Total Suspended Solids (TSS).....	46
4.1.5 Summary of Hydrology Results.....	49
4.2 Nutrients.....	50
4.2.1 Nutrient composition of manure.....	50
4.2.2 Dissolved Reactive Phosphorus (DRP).....	51

4.2.3	Bioavailable Phosphorus (BAP)	55
4.2.4	Total Phosphorus (TP)	59
4.2.5	Nitrate-Nitrogen (NO ₃ ⁻ -N).....	63
4.2.6	Ammonia Nitrogen (NH ₄ ⁺ -N).....	67
4.2.7	Total Nitrogen (TN).....	70
4.2.8	Summary of nutrient transport study	74
4.3	Bacteria	76
4.3.1	Fecal Coliform (FC)	76
4.3.2	E. Coli (EC)	80
4.3.3	Enterococcus (ENT)	84
4.3.4	Summary of bacteria transport study	87
Chapter 5 Summary and Conclusions		89
5.1	Nutrient Transport	89
5.2	Bacterial Transport	91
5.3	Conclusions.....	92
5.4	Implications and Recommendations	93
5.5	Recommendations for future study	94
References		95
Appendix A Manure Analysis		101
Appendix B Soil Properties		104
Appendix C Rainfall Data		106
Appendix D Runoff Data		108
Appendix E Nutrient Analysis.....		114
Appendix F Bacterial Analysis Results.....		118

List of Tables

Table 2.1 Federal standards for common indicator bacterial species and their individual uses.	17
Table 3.1 Average textural and chemical properties of soil at the experimental plots.....	27
Table 3.2 Nutrient analysis results of manure used in the experiment. The dates in parenthesis show the date, the samples were procured. Table also shows nutrient composition of inorganic fertilizer.....	29
Table 3.3 Amount of manure and fertilizer applied per unit area and per plot for each treatment	30
Table 3.4 Water quality parameters and test number conducted on runoff water samples.....	35
Table 4.1 Average canopy cover development from various treatments, 42 days after planting of corn.....	38
Table 4.2 Average time in minutes to start of the runoff from each treatment for all the simulated events	40
Table 4.3 Mean peak runoff rate from the plots for all the simulated events in mm/hour	42
Table 4.4 Mean runoff volume (mm) from the treatments for all the simulated events.....	44
Table 4.5 Mean TSS concentrations and yield in runoff from different treatments for all the simulated events.....	46
Table 4.6 Descriptive statistics of the TSS (kg/ha) yield in runoff from different treatments for each simulated event.....	48
Table 4.7 Average nutrient composition for dairy manure samples, poultry litter samples and inorganic fertilizer.....	51
Table 4.8 Descriptive Statistics for the DRP concentrations of runoff from different treatments for each simulated event.....	52
Table 4.9 Descriptive Statistics for the DRP yield in runoff from different treatments for each simulated event.....	54
Table 4.10 Descriptive statistics fro the BAP concentrations in runoff from different treatments for each simulated event.....	56
Table 4.11 Descriptive statistics for the BAP yield (g/ha) in runoff from different simulated events for each simulated event	58
Table 4.12 Descriptive statistics for the TP concentrations (mg/l) in runoff from different treatments for all the simulated events	60
Table 4.13 Descriptive statistics for the TP yield (g/ha) in runoff from different treatments for each simulated event.....	62
Table 4.14 Descriptive statistics for the NO ₃ ⁻ -N concentration (mg/l) in runoff from different treatments for each simulated event.....	64
Table 4.15 Descriptive statistics for the NO ₃ ⁻ -N yield in runoff from different treatments for each simulated event.....	66
Table 4.16 Mean NH ₄ ⁺ -N concentrations and yield in runoff from different treatments for all the simulate events.....	68
Table 4.17 Descriptive statistics for the NH ₄ ⁺ -N yield (g/ha) in runoff from different treatments for each simulated event.....	70
Table 4.18 Mean TN concentrations and yield in runoff from different treatments for all the simulated events.....	71
Table 4.19 Descriptive statistics for the TN yield (g/ha) in runoff from different treatments for each simulated event.....	73
Table 4.20 Descriptive statistics for the FC concentrations of runoff from different treatments for all the simulated rainfall events.....	77
Table 4.21 Descriptive statistics for the FC yield in runoff from different treatments for each simulated event.....	80

Table 4.22 Descriptive statistics for the E. Coli concentration in runoff from different treatments for all the simulated events	81
Table 4.21 Descriptive statistics for EC yield in runoff from different treatments for each simulated rainfall event	83
Table 4.22 Descriptive statistics for ENT concentrations in runoff (cfu/100ml) from different treatments for all the simulated events	85
Table 4.23 Enterococcus yield in runoff (cfu/ha) from different treatments for each simulated rainfall event	86
Table A1 Dairy manure analysis results	102
Table A2 Chicken Litter analysis results	103
Table B1 Particle size analysis of soil samples	105
Table B2 Chemical Properties of the Soil	105
Table C1 Rainfall reported from different raingages for all the simulated rainfall events	107
Table C2 Raingage readings recorded between simulated rainfall events in mm	107
Table D1 Time to initiate runoff from each treatment plot, in minutes. The symbol in bracket shows the treatment	109
Table D2 Peak runoff rate (mm/h) from each plot	109
Table D3 Total runoff volume (mm) from each plot	110
Table D4 Rate of runoff (mm/h) recorded at the time of sampling from each plot for the first simulated event	111
Table D5 Rate of runoff (mm/h) recorded at the time of sampling from each plot for the second simulated event	111
Table D6 Rate of runoff (mm/h) recorded at the time of sampling from each plot for the third simulated event	112
Table D7 Volume to prepare flow weighted samples for each plot for first simulated rainfall event	112
Table D8 Volume to prepare flow weighted samples for the second simulated rainfall event ..	113
Table D9 Volume to prepare the flow weighted samples for the third simulated event	113
Table E1 Nutrient analysis results from the flow weighted samples from each plot for the first simulated rainfall event	115
Table E2 Nutrient analysis results from the flow weighted samples from each plot for the second simulated rainfall event	116
Table E3 Nutrient analysis results from the flow weighted samples from each plot for the third simulated rainfall event	117
Table E4 Nutrient analysis results of the raw water used in each simulated event	117
Table F1 Bacterial analysis results from each plot for the first simulated event	119
Table F2 Bacterial analysis results from each plot for the second simulated event	119
Table F3 Bacterial analysis results from each plot for the third simulated event	120

List of Figures

Figure 2.1 Aerial view of lake fertilized with P	5
Figure 3.1 Contour map of the site and the experiment area marked for final plot construction.	24
Figure 3.2 Schematic diagram of the layout of experimental plots on the field	25
Figure 3.3 Rectangular flume attached to a stage recorder.....	26
Figure 3.4 Schematic diagram of the layout of rainfall simulator over the experimental plots	31
Figure 3.5 Rainfall simulator in operation.....	32
Figure 3.6 A graduate student (Tess Wynn) collecting the runoff samples flowing through the flume	33
Figure 4.1 Average times to start of runoff for each treatment during the three simulated rainfall events.....	40
Figure 4.2 Mean peak runoff rate from each treatment for all the three simulated rainfall events	43
Figure 4.3 Mean runoff volume resulted from different treatments for all the three simulated rainfall events	45
Figure 4.4 Mean TSS concentration in runoff from different treatments for all the simulated events	47
Figure 4.5 Amount of TSS lost in surface runoff per unit area per from different treatments for all the simulated events	49
Figure 4.6 Mean SP concentration in runoff from different treatments in all simulated rainfall events.....	52
Figure 4.7 Soluble Phosphorus yield in runoff from different treatments for all the simulated events.....	55
Figure 4.8 Concentration of Bioavailable Phosphorus in runoff from different treatments for the simulated events.....	56
Figure 4.9 Amount of Bioavailable Phosphorus in runoff per unit area from different treatments for all the simulated events	59
Figure 4.10 Mean concentration of TP in runoff from different treatments for all the simulated events.....	60
Figure 4.11 Mean amount of Total Phosphorus in runoff per unit area from different treatments for all the simulated events	63
Figure 4.12 Mean NO_3^- -N concentration of runoff from the treatments for different simulated events.....	65
Figure 4.13 Nitrate-Nitrogen losses in surface runoff per unit area for different treatments for all the simulated events	67
Figure 4.14 Mean NH_4^+ -N concentrations in runoff from the different treatments for all the simulated events.....	68
Figure 4.15 Mean amount of ammonia-Nitrogen lost in runoff per unit area from different treatments for all the simulated events	70
Figure 4.16 Mean TN concentrations (mg/l) of surface runoff from different treatments for each simulated event.....	72
Figure 4.17 Total Nitrogen yield from different treatments for each the simulated rainfall event.....	74
Figure 4.18 Mean FC concentration in runoff from different treatments for all the simulated events	77
Figure 4.19 Mean amount of Fecal Coliform per unit area in runoff from different treatments for all the simulated events	80

Figure 4.20 Mean E. Coli concentrations in runoff from different treatments for all the simulated events.....	82
Figure 4.21 Mean counts of E. Coli in runoff from different treatments for all the simulated events	84
Figure 4.22 Mean concentration of Enterococcus in runoff from different treatments for all the simulated events.....	85
Figure 4.23 Mean Enterococcus count in runoff from different treatments for all the simulated events.....	87

Chapter 1 Introduction

Animal agriculture is a growing industry in the nation, which produces \$98.8 billion per year in farm revenue (GAO, 1999). A large amount of waste is produced by the animal industry and the application of these animal wastes on land is an economically viable method to utilizing animal waste and recycling nutrients back into the system. However, land application of animal wastes could impair water quality by introducing pollutants such as nutrients, organic matter, sediment, pathogens, heavy metals, hormones, antibiotics and ammonia (GAO, 1999). Despite tremendous progress achieved in reducing water pollution, 40% of the nation's water bodies assessed by the states do not meet water quality standards (NRAES, 2000). According to the U.S.EPA, agricultural activities are the leading sources of impairment to the nation's rivers and lakes in the U.S., and are a significant source of impairment to its coastal waters and ground water (GAO, 1995). The U.S. EPA also reported that eight percent or nearly 19,811 river miles in U.S. are impaired by pathogens (NRAES, 2000). The EPA's water quality Inventory also reported that agriculture is the leading source of impairment in estuaries.

In Virginia, more than 1,300 poultry operations produce over 4 million metric tons of manure each year (CBF, 1998). It is estimated that poultry farmers generate 159,000 tons more poultry waste than can be properly applied to the land in Rockingham County alone (Pease et al., 1988). In Virginia, 46 river miles, 1500 acres of lakes and 1539 acres of estuaries have been reported impaired due to excessive nutrients (VDEQ, 2002). Pathogen indicators are responsible for impairment of 3111 river miles, 1061 acre lakes and 115 square miles of estuaries (VDEQ, 2000). On the Virginia eastern shore, close to one-third of the N and two-fifth of the P entering the Chesapeake Bay from that region are attributed to animal waste (CBF, 1998). Excess nutrients produced by the animal operations needs to be managed in order to minimize the pollution threat to receiving water sources. These management operations, as directed by Unified National Strategy (USDA, 1998) should include modification of animal diets, improved manure handling, use of manure in energy production and application to cropland in such a manner that it would not introduce an excess of nutrients and minimizes runoff.

Phosphorus (P) and nitrogen (N) are two of the sixteen elements that are essential for plant growth and their economic benefits are well documented (AWMFH, 2002). Manure is very effective in improving the soil properties and crop production as it contains nutrients and organic matter. Plants require nutrients for their development, and organic matter improves the structure of the soil and consequently facilitates good vegetation growth (Risse and Gilley, 2001).

However, high concentration of these nutrients in surface runoff is a potential pollution threat to receiving waters. The U.S. EPA reported that N and P from agriculture accelerate biological production in receiving waters and results in a variety of problems including clogged pipelines, fish kills and reduced recreational opportunities (NRAES, 2000). Agricultural loss of P in runoff is the primary cause of eutrophication in many fresh water systems (Birr and Mulla, 2001).

These nutrients are applied in several forms on agricultural land including inorganic fertilizer, poultry manure, dairy manure, beef cattle manure, and sewage sludge; depending upon their availability and cost. However, different types of manure have different compositions and they also vary according to the ration of animals, method of collection and type of storage. Although studies have been carried out comparing the effects of poultry litter with inorganic fertilizer on runoff water quality (Wood et.al.,1999; Heathman et al., 1995; Vories et al., 2001), few investigations have been conducted to compare water quality impacts of inorganic fertilizer with beef cattle manure (Eghball and Gilley, 1999) and dairy manure (Paul et al., 2001). There is a need for a comprehensive study to compare the effects of different types of animal manure and inorganic fertilizer on surface runoff quality.

Fecal materials from warm blooded animals contain several microorganisms, some of which may be pathogenic. Some diseases carried by animals are transmittable to human beings (AWMFH, 2002). Since, it is impractical to test water for individual pathogens, impairment by pathogens is often quantified based on indicator species such as total coliform (TC), fecal coliform (FC) and fecal streptococcus (FS), which are found in the intestines of warm-blooded animals (Walker et al., 1990). The sources of fecal contamination are land applied manure, manure deposited by grazing animals, wildlife feces, combined sewer outflows, and septic systems. The state of Virginia currently uses E. Coli, FC and enterococcus (ENT) as indicator organisms for fecal contamination. The present study investigated the fate and transport of these bacterial indicators..

Even though much research on N and P has been done in recent years, many aspects regarding agricultural production and environmental quality are not completely understood (Sharpley and Tunney, 2000) and there is a need to create a balance between these two important issues. With the increase in animal production and large amounts of waste produced each year, a strategy needs to be developed so that animal waste is appropriately applied to agricultural land in an effort to reduce downstream water quality problems. This study provides

an insight into the behavior of nutrients and indicator bacteria in runoff from cropland treated with different types of animal manures.

1.1 Goals and Objectives

The goal of this study was to quantify nutrient and bacterial losses from croplands treated with animal manure and inorganic fertilizer. The specific objectives of this research are as follows

1. Evaluate the impact of dairy manure, poultry litter and inorganic fertilizer, applied according to crop P requirements on P and N transport in runoff from cropland.
2. Evaluate the impact of dairy manure and poultry litter applied according to crop P requirements on the transport of indicator bacteria in runoff from cropland.

Chapter 2 Literature Review

Water quality and its impairment by several factors has always been an issue of discussion and research. Various agriculture activities have been responsible for impairment of the nation's water quality (GAO, 1995). Animal waste or manure application is done to increase soil fertility and productivity and also to recycle animal waste. With the increase in animal agriculture in the US, the land application of manure has turned into a problem, as the chemicals and pathogens from these wastes may flow into streams or creeks and impair water quality. This section discusses the relevant work conducted in this field.

2.1 Water quality in U.S.

Over 40% of the nation's water bodies do not meet water quality standards, despite the tremendous progress made in reducing water pollution (USEPA, 2003). Nitrogen and P from agriculture accelerate biological production in receiving waters resulting in a variety of problems including clogged pipelines, fish kills and reduced recreational opportunities (NRAES, 2000). Agricultural activities are the leading sources of impairment to the nation's rivers and lakes and a significant source of impairment to its coastal waters and ground water (GAO, 1995). The U.S. EPA also reported that about 19,811 river miles in the nation are polluted by pathogens.

In Virginia, 46 river miles, 1500 acres of lakes and 1539 acres of estuaries have been reported impaired due to excessive nutrients (VDEQ, 2002). Pathogen indicators are responsible for impairment of 3111 river miles, 1061 acre lakes and 115 square miles of estuaries (VDEQ, 2000). Animal wastes are responsible for one third of N and two fifth of P lost to the Chesapeake Bay from Virginia's Eastern Shore (CBF, 1998).

2.2 Eutrophication

Eutrophication is a natural process by which lakes or a waterbody gradually age and become more productive and it takes many years for a lake to become eutrophic (ELA, 2003). However, the addition of excess nutrients into the water bodies through various pathways actually accelerates this process. These nutrients can overstimulate the growth of algae, interfering with designated use of the waterbody. Algal blooms blocks the sunlight and causes underwater grasses to die disrupting the natural ecosystem around the waterbody. As the algae start dying and decompose, the organic processes

increase and oxygen is used up. This depletion of oxygen can cause the aquatic animals to die (USEPA, 2003).

There is some research which suggests that the factors which limit algal growth vary by season (Conley, 2000), however, phytoplankton growth in estuarine and coastal systems is limited by Nitrogen (N) availability (Nixon, 1995 and Howarth et al., 2000). Phosphorus is generally the limiting nutrient in the development of algae in most of the fresh water sources. When P enters fresh water sources it causes the nuisance growth of algae and accelerates the aging process in lakes (AWMFH, 2000). The same phenomena can be observed in estuaries where N is the limiting nutrient. Due to the excessive growth of algae, the available oxygen in the water sources begin to decrease and could cause fish kills. Eutrophication also results in clogged pipelines and reduced recreational opportunities (NRAES, 2000). Agriculture is considered to be the leading cause of these impairments in estuaries according to the EPA's water quality inventory report. Figure 2.1 by Karen Scott (1994) shows the visual effects of adding P to a freshwater lake for the 26th consecutive year. The lake in the background is unfertilized.



Figure 2.1 Aerial view of lake fertilized with P. Lake in background is unfertilized.
Source: ELA, 2003. Photo by Karen Scott, 1994

With the increase in eutrophication as the single largest water quality problem in the world (ELA, 2003), there is a need to understand the movement of nutrients from different types of agricultural lands, treated by different kinds of fertilizers, and under various tillage practices. This research helps to understand the movement of nutrients from cultivated lands treated with animal manure according to the P requirement of the field.

2.3 Manure

Manure is an organic material that is used to fertilize land and is comprised mainly of animal feces and urine of livestock with or without accompanying material such as bedding straw or hay. Farm animal defecate nutrients such as, Nitrogen (N), Phosphorus (P), and Potassium (K) which are supplied to them in their ration. Manure or organic agricultural wastes improves the tilth of soil, decreases crusting, adds organic matter and increases infiltration (AWMFH, 2002). Risse and Gilley (2001) reported that manures improve the physical condition of soil and enhance crop production because they contain nutrients and organic matter. Animal manure is primarily land applied as it serves as the dual purpose of soil enrichment and waste removal. Animal manures are also utilized in energy production (Wudi et al., 2002), fertilizer production (drying, pelletization and granulation), and home and farm land application after composting (Pease, 2002). The other methods of manure utilization are not very popular and land application is primarily used to dispose animal manure. Manures as a by- product of animal agriculture are very cheap alternative to inorganic fertilizer and land application provides an efficient way to get rid of the animal waste.

Animal agriculture is a growing industry in the nation and produces \$98.8 billion per year in farm revenue (GAO, 1999). About 1.37 billion tons of animal manure is produced in the US per year (US Senate, 1997). The large amounts of waste produced by these industries are a potential threat to surface and ground water resources. In Virginia, there are more than 1300 unregulated poultry operations that produce more than one billion pounds of manure each year (CBF, 1998).

Animal manure, if not applied properly on land, can impair water quality by introducing pollutants such as nutrients, organic matter, sediment, pathogens, heavy metals, hormones, antibiotics and ammonia (GAO, 1999). Several plot and field scale studies have been conducted to assess the effects of excessive manure application on surface water quality. Researchers have shown that continued application of manure in some areas has lead to increased N and P levels in soils and surface runoff (Heathman et al., 1995; Westerman et al., 1983; McLeod and Hegg, 1984; Edwards and Daniel, 1993; Burwell et al., 1977). These nutrients upon reaching surface water systems, may lead to their eutrophication (Birr & Mulla, 2001; EPA, 1994).

Manures can also be responsible for introducing microorganisms to surface water systems. These microorganisms can contaminate food and water supplies of both animals and human beings (Patni et al., 1985; Jack and Hepper, 1969; Miner et al., 1967).

2.3.1 Properties of Manure

Animal manures are categorized by their physical and chemical properties. The physical properties include consistency, moisture content, solid content, etc. These properties help in deciding the storage and management techniques for the manure. Chemical properties include the amount and forms of different nutrients and their forms present in the manure. It also includes the Chemical Oxygen Demand (COD) and Biological Oxygen Demand (BOD). These properties depends upon the type of animal, species of animal, kind of facility, waste management techniques, type of ration, weather etc. (AWMFH, 2002).

The waste leaving from an animal facility not only includes feces and urine, but other materials such as bedding, soil, wasted feed, wasted water, dust, hair, etc. In general, these extra things get added and cannot be avoided, and are considered part of animal waste. The waste from a typical dairy facility is mixed with water and is in the form of slurry (solid content less than 5%), while the waste from a chicken or turkey facility includes the feces of these animals, wasted feed, bedding materials, feathers and is collectively referred to as Litter. Average N content of dairy liquid manure in Virginia is 22.61 lbs/1000 gal and the average P content is 12.07 lbs/1000 gal, while the average N content of litter in Virginia is 62.58 lbs/ ton and the average P content is 62.12 lbs/ ton (VA DCR, 1995). Nitrogen and P being the key nutrients which help in the growth and development of plants are the main nutrients that are used in determining the application rate of animal manure. These nutrients are discussed in detail in the following sections.

2.4 Nitrogen (N)

Nitrogen is one of the macro-nutrients, required for complete plant development. It is a key component of dioxynucleic acid (DNA), ribonucleic acid (RNA), amino acids and proteins required for plant growth. Plants extract N from agricultural systems through

different processes. Farmers supply N to field crops in the form of animal manures and fertilizers.

Nitrogen changes many forms from the time of its application until the time it is taken up by plants, or transported by surface runoff to other water bodies. This movement of N in the whole system, between the atmosphere, soil and living beings is termed as the "Nitrogen Cycle". A fairly simplistic approach to the N cycle is explained in subsequent paragraphs to describe the major forms of N available in soil. Nitrogen exists mainly in the form of atmospheric molecular dinitrogen (N_2) and hence not usable by most plants, except for some leguminous plants. The other forms of N such as ammonia (NH_3), ammonium (NH_4^+ -N) and nitrate (NO_3^- -N) are available to plants through fertilizer application, N fixation by bacteria, manure application, decomposition of plant residues and precipitation.

Nitrogen in soils is generally in the form of organic compounds (org-N), NH_4^+ -N, and NO_3^- -N ions. Org-N is not readily available to plants, but can be converted into available forms by soil microorganisms. Nitrogen in fresh animal manure is organic by 60 to 80%, but the manure in lagoons has about 20-30% of org-N and 40-90% of the org-N in manure is "mineralized" into ammonia within 4-5 months of application (AWMFH, 2002). Mineralization is the process of conversion of org-N to NH_4^+ -N.

Ammonium Nitrogen or ammoniacal nitrogen or NH_4^+ -N is readily available to plants as it can attach itself to the negatively charged clay particles in soil. Inorganic fertilizers are provided in the form of Urea which rapidly hydrolyses into NH_4^+ -N in soils. Animal manure provides N in the form of NH_4^+ -N also. NH_4^+ -N is relatively immobile in soils as it attaches itself to soil particles. However, it can move with eroded soil particles. A significant amount of NH_4^+ -N volatilizes as NH_3 during the time of manure application. If applied as irrigation from sprinkler head, as much as 25% of NH_4^+ -N is lost as NH_3 before it reaches the surface (AWMFH, 2002).

Unionized ammonia is poisonous and its limit in surface water has been proposed as 0.02 mg/l to protect aquatic life. The criteria for NH_4^+ -N in fresh water and saltwater have been specified by Virginia DCR, and it varies according to the pH and temperature of the

water (DEQ, 2003). At pH of more than 8, the criteria for NH_4^+ -N gets as low as less than 1.00 mg/l and at this pH, criteria further reduces with temperature.

In addition to being taken up by plants, NH_4^+ -N can undergo the process of “Nitrification” by the chemotrophic bacteria in the soil. In this process, NH_4^+ -N is converted to Nitrite (NO_2^-), which is very short lived and then to NO_3^- (Novotny and Olem, 1994). Nitrate is relatively soluble in water and is readily used by plants. This process of utilizing N by plants and converting them to proteins is termed as “Nitrogen Accumulation” or “Biological Uptake”. The NO_3^- being soluble can leach through the soil and reach subsurface water. Under anoxic conditions, which prevail when the soil is saturated, rhizomes of some plants create aerobic packets near the roots. Nitrate can serve as an electron acceptor and then reduce to its gaseous forms (Novotny and Olem, 1994). This process of conversion of NO_3^- to its gaseous forms is termed as “denitrification”.

Nitrates are not toxic to aquatic animals, but they are a source of enrichment for aquatic plants. NO_3^- can help promote algal blooms and production of other aquatic vegetation, provided other nutrients are available. The EPA has not set any limits for NO_3^- concentration in surface waters. Although a limit of 10 mg/l of NO_3^- -N is set for drinking water (AWMFH, 2002).

2.4.1 N in Manure

About 60-80% of the total N in fresh animal manure is in organic forms. An anaerobic lagoon has 20-30% of the total N in organic forms. The N in the solid fraction of most animals is in the form of complex organic compounds, while N in liquid is in the form of Urea, or NH_4^+ -N. If the stored manure is well aerated, the waste may have NO_3^- -N. The nitrogen content of manure is represented as Total N, which includes all the N compounds discussed above. Total Kjeldahl Nitrogen or TKN represents the amount of organic N and ammoniacal N. Nitrogen content in manure can also be represented in the form of individual components and the amount of available N depending upon the method of application.

DCR of Virginia (1995) and AWMFH (2000) report on nutrient content of different types of animal manure and it reported that N content of manure depends upon the type of animal, type of animal facility, type of manure storage facility, and diet of animals. The N

content of fresh excreted waste from dairy animals, beef, swine and veal vary from 0.30 to 0.60 lb/d/AU, while the N content of poultry can vary from 0.5 to 1.10 lb/d/AU. The N content of waste from milking facilities can vary from 0.7 to 1.6 lb/1000 gal, while the N content of dairy manure from lagoon can vary from 1.6 lb/1000 gal (supernatant) to about 20 lb/1000 gal (sludge). Similarly poultry waste in form of litter can have TKN content of about 60 lbs/ton and liquid poultry waste can have TKN content of about 50 lbs/1000 gal. Forms of N available in manure also vary according to the type of animal, type of manure storage. On an average in Virginia, N in dry poultry has 77% of org-N and rest as NH_4^+ -N, while liquid poultry has 36% of N in the form of org-N and rest as NH_4^+ -N. Liquid dairy has 58% of N in org-N form and 42% in NH_4^+ -N form.

2.4.2 Surface transport of N

Nitrogen to rivers or lakes is transported from air borne, surface or underground sources. However, much of the N in lakes or rivers comes through the eroded sediments and soil organic matter or dissolved in surface runoff (Follett and Delgado, 2002). The kinetic forces of rainfall or irrigation detach the soil particles, increase their suspension and transport important components, soluble nutrients, and soil organic matter out of the fields (Delgado, 2002). Many water quality studies have been conducted to measure the amount of N lost through surface runoff in different agricultural systems treated with different fertilizers or different animal manures.

The concentration of total N in surface runoff from cropland and grassed areas has been reported to increase following a fertilizer or manure application (Heathman et al., 1995; Edwards and Daniel, 1994 and 1993). Follett and Delgado, (2001) gave a nationwide estimate of inorganic N lost, as attached to sediments as about 6.6 to 10 million t/yr and this amount does not account for the loss of dissolved N in the form of NO_3^- -N to surface waters and ground water. Nitrogen is necessary for aquatic plant growth, but as explained earlier, it can lead to eutrophication of surface water sources, if it is the limiting nutrient.

Loss of N to surface water sources, however, depends on several factors such as type of manure, timing of application, method of application, intensity of rainfall, timing and frequency of rainfall or irrigation, soil conditions, type of tillage, and slope. Vories et al. (2001) studied the quality of runoff from a cotton field fertilized with poultry litter for three

years and reported a reduction in the amount of $\text{NO}_3\text{-N}$ from fields receiving poultry litter compared to the fields receiving inorganic fertilizers. However, the concentration of nutrients associated with the solid fraction of chicken litter increased and the yield was lower for field having poultry litter as the only fertilizer source. Wood et al., (1999) studied the effect of broiler litter and inorganic fertilizer on the field with a corn and winter rye cropping system and reported that the concentration of all the nutrients (except Ca) in runoff was enough to support eutrophication in surface water.

Eghball and Gilley (1999) studied the concentration of P and N in runoff following an application of beef cattle manure and compost. The N based manure application resulted in excess $\text{NH}_4^+\text{-N}$ concentration in runoff when the soil was not tilled, compared to disked soil. They recommended P based manure application to be agronomically and environmentally sound. However, the authors did not report about the yield with the P based management system. Heathman et al. (1995) studied the water quality following application of poultry litter on farm land and reported an increase in $\text{NO}_3\text{-N}$ concentrations in runoff, in tilled and no-till plots compared to the control treatment. They also reported an increase in soil N following application of poultry litter.

Most of the water quality studies have reported an increase in the N concentration of runoff following application of manure or fertilizer. This loss of N depends on the time of rainfall event in the cropping season (Burwell et al., 1977) and could be controlled by more restrictive N and P management (Sharpley, 1997). This loss can also be controlled by reducing the sediment movement from the field.

This study simulates the worst case scenario in which the simulated rainfall occurs just after planting corn in conventionally tilled plots fertilized with P based management system. The study will help in assessing the effects of P based management system on N loss in surface runoff and will also describe if different kinds of manures respond equally to P based manure management.

2.5 Phosphorus (P)

Phosphorus is also one of the macro nutrients required for complete development of plants. Phosphorus is a part of several key plant structure compounds and acts as a catalyst in several key biochemical reactions. It is also an important part of DNA, RNA

(genetic units) and ATP (Energy releasing compounds). Phosphorus is generally supplied to the plants in the form of animal manures or phosphate fertilizers.

2.5.1 P and Its Forms

Phosphorus found in soils is broadly classified as organic-P and inorganic-P. Organic-P is found in the organic matter present in soil while the inorganic-P consists of apatite, complexes of iron and aluminum phosphates and P adsorbed to clay particles of soil. The inorganic forms of P are subdivided into soluble and sediment bound P. The solubility of both of these forms is very low, and very low amounts of organic P are available as solution in soil (EFU Manual, 2002).

Hansen et al. (2002) discussed the presence of P in soil as if they are distributed in three hypothetical pools namely; stable P, reactive P and soluble P. Stable and reactive pools contain both organic and inorganic P. Soluble P mostly consists of H_2PO_4 and HPO_4 and is most readily available to plants (Hansen et al., 2002; AWMFH, 2002), but the soluble P comprises of less than 1% of the total P in soil (Brady and Weil, 1999). Phosphorus in the reactive pool is in equilibrium with soluble P, and whenever plant uptake the soluble P, P from the reactive pool replaces it by the processes such as desorption, dissolution, and mineralization. Stable P is not biologically available and is made up of occluded, insoluble or tightly sorbed P forms.

Organic-P is a part of all living beings and in soils; it is attached to the microbial tissue or stays in the dead roots, plant residues, unmineralized wastes etc. This form of organic P is present in both reactive and stable P forms. About 73% of P in fresh waste of various livestock is in the form of organic-P (AWMFH, 2002) and it is present in the reactive P pool. The organic P also competes with the inorganic-P for P adsorption sites in soil.

Inorganic P is present in all three P pools in soils. In the solution P pool, inorganic P is present in the form of H_2PO_4 and HPO_4 . In the reactive P pool, P is loosely bound to Al, Fe and Ca. When plants uptake P from the soil solution, P from these loosely exchangeable sites replaces them and maintains the equilibrium with solution P. As the inorganic P from these sites is depleted, organic-P replaces the exchange sites and maintains the equilibrium in the reactive P pool. The reverse of this process is also true. When the organic-P is applied to the soil, it replaces P from adsorption sites and makes

more P available in solution form, thereby making more P available for plant uptake (AWMFH, 2002).

In principle, stable P also maintains equilibrium with reactive P, but the reactions of stable P forms are too slow to be significant for agricultural activities. In the stable pool, P is present in the crystalline Al, and Fe or Ca compounds (Brandy and Weil, 1999).

2.5.2 P in manure

Phosphorus in fresh manure is mostly in organic forms. However, for aged manure or composts, Sharpley and Moyer (2000) investigated the forms of P and reported that aged manure and composts consists of more inorganic P (63-92%) than organic-P (5-25%). They also reported that the P and N content of poultry litter is more than that of dairy manure and poultry litter also contains more water soluble P than dairy manure. Phosphorus content in fresh waste varies from 0.07 lb/day/AU in dairy animals to 0.14 lb/day/AU in beef and 0.22 lb/day/AU in swine. The P content in poultry wastes is about 0.31 lb/day/AU in fresh poultry waste (AWMFH, 2000). The average P content in dairy manure in Virginia is about 12.07 lbs/1000 gal, however, in liquid poultry; the P content is 41.01 lbs/ 1000 gal. The average P content of poultry litter is 62.12 lbs/ ton (VDCR, 1995).

Farmers use to apply manure to agricultural fields according to the N requirement of crops as a general practice, which increases the amount of available P which cannot be utilized by plants completely. This excess P is subject to loss in runoff and to nearby surface water sources or it can build up the soil P level and this excess P may be available for surface runoff in subsequent rainfalls.

2.5.3 P loading in surface runoff

Phosphorus loading in surface runoff is a complex process. Phosphorus can be lost in surface runoff as DP (Dissolved P) or with eroded soil particles as PP (Particulate P). PP forms a major part of the P transported from cultivated land (Schuman et al., 1973; Sharpley et al., 1987). Sharpley (1993) discussed the movement of P in the landscape in detail and classified different factors responsible for P loading as transport factors and source factors. Transport factors include erosion and runoff.

Phosphorus is sorbed by soil materials and hence erosion is responsible for PP movement (Burwell et al., 1977; Garbrecht and Sharpley, 1992; Schuman et al., 1973). During detachment and movement of sediment in runoff, finer size particles are preferentially eroded. The finer particles have higher sorption capacity and hence they sorb more P than the surface soil. As these finer fractions move through the soil, they keep on sorbing the DP present in the runoff or streamflow. As the rainfall or irrigation water interacts with the soil surface various processes such as desorption, dissolution and extraction of P from soil and plant material takes place. These processes lead to enrichment of surface runoff with DP.

The source factors responsible for P loss in runoff include soil test P (STP) levels, amount of P applied and the method and timing of P application on soils (Sharpley, 1995). Addition of more P, than required by plants, to soils increases STP levels of soils overtime and increases the potential of P to be lost in surface runoff. Sharpley (1995) also suggested that no further economic advantage can be obtained by applying more P, than a certain threshold value. The potential for loss of P is also increased, if fertilizer or manure is surface applied compared to incorporation. Another source factor controlling P loss in runoff is the timing of application. The loss of P in surface runoff can be reduced by avoiding application of fertilizer or manure when a storm or rainfall is imminent (Sharpley, 1995). An investigation by Westerman and Overcash (1980) suggested that the TP loss in runoff can be reduced by 90% if the first runoff event is delayed to 3 days from 1 day.

Phosphorus loss in runoff is measured in the forms of dissolved P (DP), bioavailable P (BAP) and total P (TP). DP includes the orthophosphate (OP), which is dissolved in runoff and is readily available for algal growth (Peters, 1981; Vollenweider, 1968; Walton and Lee, 1972). Bioavailable P (BAP) includes not only all of DP, but some OP bound to sediments (Sharpley et al., 1992). The OP in sediment bound P can vary from 10-90% depending upon the watershed (Sharpley, 1997). Since, some of the OP in sediments is also bioavailable, BAP provides a better estimate than DP, about the amount of P which is available for algal growth in waterbodies. Total P (TP) includes P in all the forms in runoff.

The loss of P in runoff can be controlled by better management of manure and fertilizer, and by controlling erosion and runoff. Management includes application of manure and fertilizer according to the P requirement of soils, incorporation of manure, and applying manure and fertilizer when the weather is clear, so that ample time can be provided for plants to take up the applied P. However, manure or fertilizer applications cannot always be scheduled according to weather, as it involves using farm machineries, which are generally rented on a daily basis. Moreover, during the season of field preparation, the laborers and machineries get extremely busy and a prior appointment is generally taken for these field works. P loss is also controlled by installing one or more BMP and thereby controlling runoff and erosion.

The study presented henceforth evaluates the effects of P based fertilizer and manure management on cropland on P loadings. Application of manure was based on the P requirements of the crops in contrast to N requirement of the crop as is the general practice. Since the amount of N cannot be supplied by manure alone, the N requirement was satisfied by application of additional inorganic fertilizer.

2.6 Pathogen

Excreta from warm blooded animals contain several micro-organisms, some of which are pathogenic and could transmit diseases to human beings (AWMFH, 2002). When manure is applied on agricultural land, the pathogens are exposed to environment. Their survival depends upon many environmental and ambient factors. Pathogens transported to surface waters from these sources lead to impaired water quality. Pathogens are one of the leading causes of water quality impairments in rivers nationwide (USEPA, 2000). About 35% of river miles assessed in 1998 were reported to be impaired by pathogen indicators, primarily Fecal Coliform (USEPA, 2000).

Pathogens present in animal manure could cause illness problems in human beings. They can be transferred to human beings by wind, insects, rodents, flowing waters etc. The potential communicable diseases from infected cattle to human beings are Salmonellosis, Leptospirosis, Anthrax, Tuberculosis, Johne's disease, Brucellosis, Listeriosis, Tetanus, Tularemia, Erysipelas, and Colibacillosis (Azedevo and Stout, 1974). However, only three major disease outbreaks have been associated with agriculture (USDA, 2000).

Morrison and Martin (1977) studied the human health aspects of animal waste application on land and suggested that crops which are eaten raw should not receive manure application unless sufficient time for pathogen die off is available. They also suggested avoiding application near high population density areas and avoiding frequent application on the same land.

It is impractical to test a water sample for all known pathogens; most information concerning water quality addresses the concentrations of the indicator groups (Walker et al., 1990). Indicator organisms are good indications of the potential pollution from the waste of warm blooded animals. The indicator organisms are used, rather than the actual pathogens, since they are easier to isolate, are safer to work with and are usually present in larger numbers than actual pathogens (Wang and Mankin, 2001). The indicator organism used widely to test for the presence of pathogens is Fecal Coliform (FC) (AWMFH, 2002). A positive test for FC clearly indicates the pollution from warm-blooded animals and a high count indicates a greater probability of the presence of pathogenic organisms.

Fecal Coliform is a subgroup of Total Coliform (TC) and originates from the intestinal tract of warm blooded animals. Some other indicators used are E.Coli, and Enterococcus. E. Coli is a member of FC group. This subgroup is used as an indicator because of its correlation with illness from swimming and gastrointestinal problems. Enterococcus correlates well with human illness in recreational waterbodies (USDA, 2000). The federal standards of indicator bacteria for primary contact with human beings are listed in Table 2.1 Individual state standards may be more stringent than these, but not lower. The table also lists the common characteristics of these indicators (Landry and Wolfe, 1999).

Table 2.1 Federal standards for common indicator bacterial species and their individual uses

Indicator Bacteria	Federal Standard	Comments
Total Coliform (TC)	1000 cfu/100 ml	Very generic and widely distributed in nature
Fecal Coliform (FC)	200 cfu/100 ml*	Originates specifically from intestinal tracts of warm blooded animals.
Escherichia Coli (E. Coli)	126 cfu/100ml*	Correlates well with illness from swimming.
Fecal Streptococcus (FS)	FC/FS > 4.0 (human source) 0.7 < FC/FS < 4.0 (multiple sources) FC/FS < 0.7 (animal source)	Used primarily to find the source of fecal contamination.
Enterococcus	33 cfu/100 ml*	Also correlate well with illness from swimming.

*30 day geometric mean

EPA has targeted 2003 for all states to start using E. Coli and Enterococcus as indicators instead of FC and TC (USEPA 1999d). E. Coli and Enterococcus provide better explanation of health risks to human beings, those who are in primary contact with the polluted water (USEPA, 1996).

2.7 Bacteria in animal waste

Fecal Coliform is found in excreta of all warm blooded animals. This includes wild animals like deer, birds, beavers etc. Therefore, a background level of fecal coliform is always present in all natural waters without the influence of human beings or domestic animals (AWMFH, 2002).

Moore et al. (1988) summarized the indicator bacterial concentrations in different types of animal manures. They reported 1.3×10^6 FC organisms per gram in chicken manure, and 3.4×10^6 FS per gram in fresh chicken waste. 1.3×10^6 FS per gram was measured for fresh cow deposits, 7.3×10^6 FC per dry weight gram in fresh dairy waste and 1.014×10^7 FS per gram dry weight. These bacterial concentrations are average values and can vary according to animal age, ration, season, housing and manure management system.

2.8 Fate and survival of bacteria

Bacteria present in animal manure are faced with different ambient and environmental conditions after they are land applied. The various variables which affect the survival of these enteric organisms are physical and chemical properties of the soil; pH; porosity; organic matter content; texture and particle size distribution; elemental composition; temperature; moisture content; absorption and filtration properties; availability of nutrients; atmospheric conditions including sunlight; humidity, precipitation, and temperature; biological interactions of organisms including competition from indigenous microflora, antibiotics and toxic substances; and density of the organisms in the waste material (Crane et al., 1983). The ambient conditions in both, storage and field affect the fate of bacteria.

A study by Smallbeck and Bromel (1975) showed that the concentrations of FC in lagoon storage decreased from 10,000 cfu/100 ml to 1000 organisms / 100 ml in five months. However the concentration again increased to about 9000 cfu/100 ml after agitation. The concentrations again started declining after agitation and reached to 800 cfu/100 ml, and similar results were obtained by Panhorst (2002).

Survival of bacteria and its fate after field application of animal manure has also been studied by researchers. Gerba et al. (1975) determined that almost 92-97% of the bacteria applied to soils remain in the top 1 cm of soil. Fraust (1982) reported maximum concentrations of FC in the top 7 cm of soil and a rapid decrease after this depth. Soils also act as filter and trap the bacteria. Organic and clay particles in soil effectively trap viruses and smaller bacteria and protozoa (Mawdsley et al., 1995). This occurs because the microbes are adsorbed to the negatively charged surfaces on clay. Larger bacteria, protozoa and helminthes are filtered by narrow pore size and bridging but, in coarse textured soil, there are more chances of viruses to migrate through the soil pores. Howell et al. (1996) also compared the FC survival in different soils and found that the survival of bacteria is greater in fine textured soils compared with coarse textured ones.

A study by Van Donsel et al. (1967) showed that the survival of bacteria is greatly reduced in summer compared to autumn and winter. The bacterial survival decreases with increased temperature (Reddy et al., 1980), and reduced soil moisture content (Boyd et al., 1969). Bacterial survival also decreases if the pH range is beyond 5.8-8.4 (Lambert, 1974) and if manure, which is the bacteria source, is incorporated into the soil

(Giddens et al., 1973). However, rainfall can extend the life of micro-organisms by providing the needed moisture and washing the organisms from the plant into the soil environment (USDA, 2000).

An “after growth” or “re-growth” has also been reported by some studies. Van Donsel et al. (1967) noticed the after growth of both tracer FC and nonfecal coliforms. After growth can take place if the bacteria are exposed to favorable environmental factors outside the host (USDA, 2000). Although a single factor cannot be attributed to re-growth of bacteria, but the soil moisture and favorable temperature helps the bacteria to grow again. Sometimes the nutrients supplied through manure help the bacteria to survive.

2.9 Die off of bacteria

Bacterial die off after they leave their host has been estimated and modeled by several investigators. The die off rate helps to determine whether application of manure is safe. If enough time is available for bacterial die off then there would be lesser chance of bacteria polluting water sources. The simplest and most commonly used equation to represent bacterial die off rate is the first order decay equation or Chick’s Law (Moore et al., 1988). The equation can be written as

$$\frac{N_t}{N_0} = 10^{-kt}, \quad (1)$$

where, N_t = Number of bacteria at time t

N_0 = Number of bacteria at time t_0

k = first order decay rate constant (1/day)

t = time elapsed since t_0 (days)

This model has been used by many researchers due to its simplistic nature. Moore et al (1988) reported die off rate constants for different kinds of animal manure in different kinds of ambient environments. Crane and Moore (1985) also conducted a study of storage experiments and reported the die off rates for FC for different animal manure storage structures.

Modifications by Mancini et al. (1978), Polprasert et al. (1983), and Reddy et al. (1981), adjusted the die off rate constant to account for the different environmental factors. MWASTE model use this equation for calculating die off in both agricultural lands and manure storages (Moore et al., 1988). A study by Kriss and Gifford (1984) reported that

the concentration of bacteria in runoff decreased from 10^7 FC/100 ml in manure samples to 4.0×10^4 FC/100 ml 30 days after application. The concentrations further reduced to 10^4 FC/100 ml after 100 days. Peak FC concentration decreased after each rainfall event.

2.10 Bacteria loading in surface runoff

Rainfall washes micro-organisms available in the feces material applied to the soil surface and directs them into soil or through surface runoff to surface water sources. Water contaminated by pathogens is a potential health risk if used for swimming or, recreational uses. Agriculture is recognized as the major cause of water quality impairment based on indicator bacteria standards. Moore et al. (1988) summarized the bacterial count in runoff from different agricultural systems. They reported a FC count of 3.0×10^4 per 100 ml of runoff from solid manure application and 5.0×10^4 per 100 ml of runoff from liquid manure application. They also reported a FC count of 9.6×10^3 per 100 ml in runoff from a poultry waste disposal area. FC counts of 9.2×10^3 to 2.9×10^4 per 100 ml of runoff were reported from cropland treated with dairy slurry.

Bacterial loading to surface runoff depends upon the duration and intensity of rainfall, fecal deposit or manure application age, and erosive properties of soil. Dudley and Karr (1979) suggested a higher amount of bacterial contamination from peak runoff because of increased transport of surface materials, despite of the increased dilution effect. There has been an attempt to quantify the number of bacteria leaving the site versus the number of bacteria applied through manure. Robin et al. (1971) studied different kinds of livestock operations and determined that 3 to 23 percent of FC deposited on the field by manure application or directly by animals is lost in runoff when averaged over the whole year. The big range accounts for the animal type, type of livestock operation and manure management strategy. McCaskey et al. (1971) reported small bacteria counts from dairy application sites where dairy manure was applied in the form of liquid, semi-liquid, or solid form at application rates varying from 20 to 300 metric tons of dry matter per hectares per year. The maximum removal rates for TC, FC and FS observed were 0.06 %, 0.007% and 0.008% of those applied for one year.

Kunkle et al. (1979) conducted a plot study and reported a decline in the population of FC in runoff with time. The population of TC, FS and enterococci stayed constant, which

were attributed to high background levels of these bacteria. They also observed that the majority of bacteria were lost in the first irrigation event after manure application. Similar studies by Crane et al. (1978) yielded similar results and they concluded that the residence time of manure on soil is an important factor in determining the amount of bacteria lost in surface runoff. It appears that some time dependant process such as adsorption, fixation of bacteria on soil, or die-off takes place which reduces the amount of bacteria lost in runoff if sufficient time is allowed between the manure application and rainfall or irrigation event.

A study by Khaleel et al. (1979) modeling various parameters, such as particle size, density, precipitation intensity, and area covered by manure and relating them to manure and sediment transport revealed that several variables are important in controlling bacterial loading to runoff. The first is the residence time as discussed above. The other variable is surface soil. Surface soil can have immediate effects in immobilizing bacteria. The other factors include the method of application and the type of waste. A modeling study by Walker et al. (1990) found incorporation to be the best method of manure application to reduce bacterial loading.

2.11 Summary

A study of relevant literature in the field of nutrient transport and bacterial loading to surface runoff indicates that eutrophication and excess bacterial loading in surface water sources are long existing problems and need to be addressed systematically. The imbalance between manure production, land availability, and crop uptake needs to be engineered so that surface water sources can be saved from deterioration. The present study will give an insight into the effects of P based manure application, and will also suggest if this method is equally good for different manures and fertilizer. It has also been reported by several studies that apart from manure and its method of application, many factors control the movement of nutrients to surface waters, such as soil type, climate, local weather and some field specific factors such as tillage type, topography, and crop type. The plot scale studies bridge the data gap by giving us more information on the behavior of N and P in different conditions.

Bacterial standards in the state river are not achieved and render water unsafe for use unless treated by some costly methods. Our understanding of bacterial transport is still

not as developed as for nutrient behavior in the field. Unlike nutrients, loading of bacteria in surface water also includes the survival, die-off and after-growth between the time of its production until they reach the surface water source. There have been several attempts to quantify and model bacterial loss from manure applied areas and evaluate the factors responsible for it. A large data gap exists in the bacteria research studies because the survival and fate of bacteria depends upon a larger number of factors compared with those affecting nutrient behavior. Plot and field scale experimental studies are required to identify the behavior of bacteria at small scale. These studies are required to model the bacteria loss and to help in controlling the bacteria loading to surface water sources.

Chapter 3 Materials and Methods

The study was conducted at the Prices Fork Research and Experiment Station, Virginia Tech from May through June, 2003. The selection, survey and preparation of the experimental site were started in March 2003. Site preparation included tilling the land, applying the manure, preparing the plots, installing the rainfall simulator and planting the corn. The first set of rainfall simulation experiments were conducted on May 14 and 15 and the next set of simulation occurred on June 17. The runoff samples collected during the experiment were analyzed in the Water Quality Laboratory of the BSE Department for TSS, nutrient concentrations and bacteria counts. Soil and manure parameters were tested before the experiments.

3.1 Site Selection

To conduct the experiment, an area of about a half acre in size and with a gentle slope in one direction was selected. The history of the site was verified with the farm manager for any previous experiments involving manure applications. The site was used as an experiment location about 4 years ago to study the movement of pesticides in runoff and it had been left fallow since. The area also had minor influences of wild life (mainly deer and rabbit) as it could be observed with a few footprints. The effect of pesticides applied four years ago was expected to have diminished to negligible levels in the soil.

The site was cleared of bushes and grass. A detailed contour map of the study site was prepared using Total Survey Stations (Topcon Inc.) and a GIS Software ArcGIS 8.2 (ESRI Inc., 1999). The contour map of the area is shown in Figure 3.1. The average slope of the area was approximately 2%. An area of 50m x 20m with good uniform slope was selected to construct the experimental plots. The selected area is shown in Figure 3.1 with black borders.

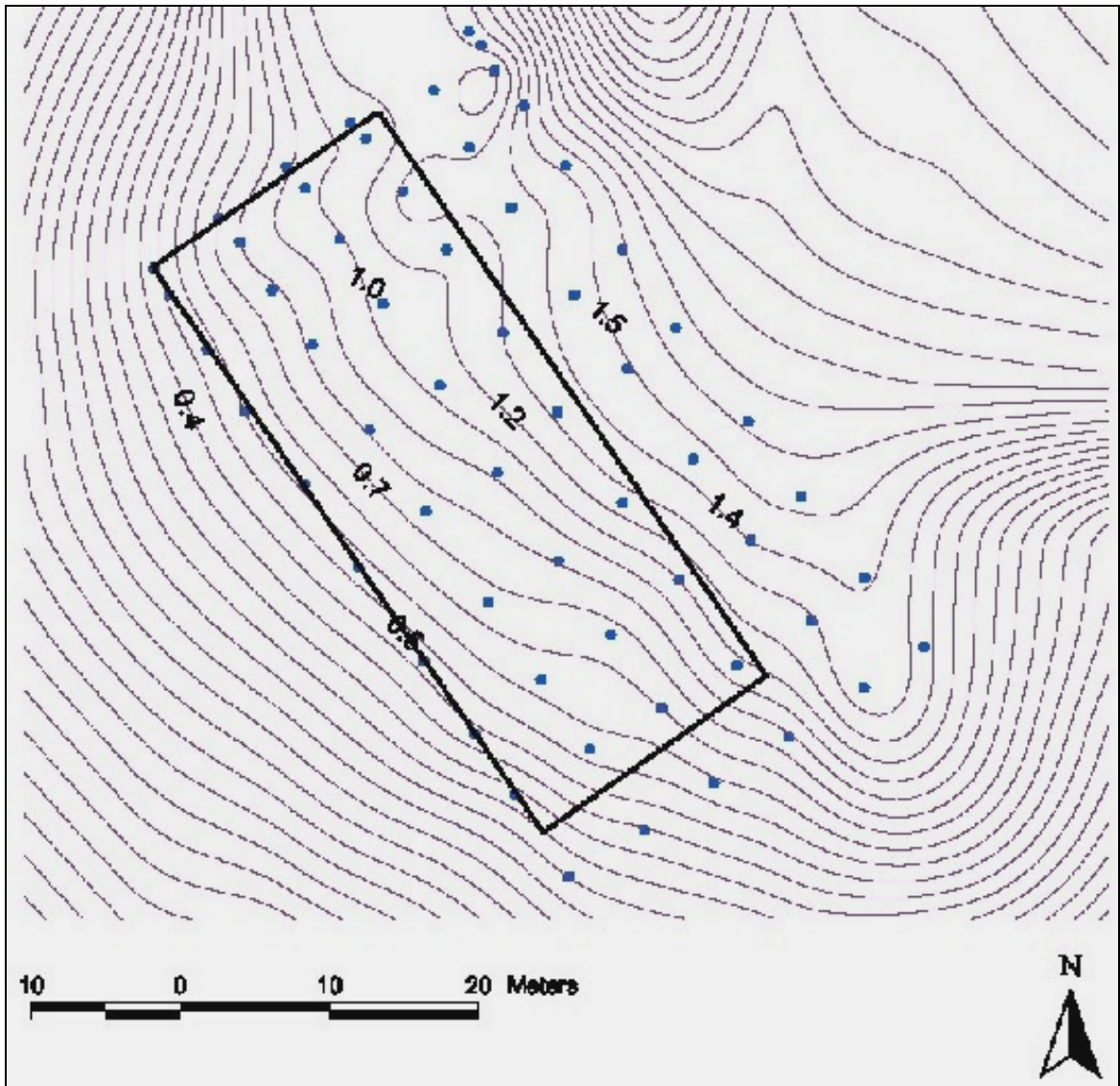


Figure 3.1 Contour map of the site and the experiment area marked for final plot construction

3.2 Site preparation

The selected area was plowed 150 mm deep with a tractor operated tiller to uproot the vegetation and stubbles in the field during late April. Due to excess vegetation, the soil could not be turned properly and mixed. The soil was again plowed two weeks later to till the soil properly. Soil samples from the field were collected after the first tillage operation and analyzed for chemical and textural properties. The field was treated with 2-4 D herbicide, about a week after the first tillage, to kill emergent weeds. After the second plowing, 15 plots, each 18.29m x 3.05m (60' x 10') in size were marked with wooden stakes and nylon strings on the selected area shown in Figure 3.1.

3.3 Plot preparation

The plots were established by inserting plywood boards about 10 - 15cm deep into the ground, so that water would not flow from one plot to another. The plywood borders were attached with screws and they were held upright with wooden stakes. The borders were also caulked at ends to prevent any water movement between the adjacent plots. The borders at the downslope end of plots were made to converge in the shape of a 'V' leaving 0.3m (1') space at the constricted end of V. The schematic diagram of experimental plots is shown in Figure 3.2. The borders at the upslope end were not fixed until the manure application, as two treatments (Incorporated Dairy Manure and Inorganic Fertilizer) needed tilling to incorporate the manure and fertilizer after their application. As soon as the incorporation was completed, metal borders were fixed at the upslope end to prevent flow of water outside the plot. In each plot, two raingages were also installed to measure the rainfall amount. The raingages were kept in place after the simulation events to measure the natural rainfall amounts between the second and the third simulations.

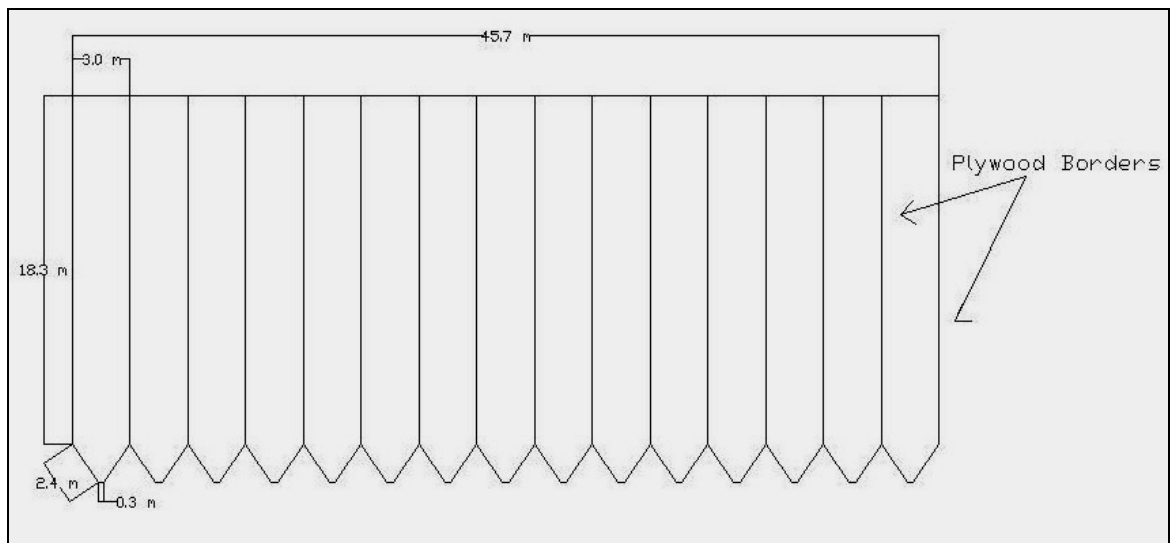


Figure 3.2 Schematic diagram of the layout of experimental plots on the field

At the end of each plot, a 6" metal rectangular 'H' flume, constructed according to USDA specifications (USDA, 1962) was installed. The metal flumes are used to measure the flow rate. The flumes were attached to a stage recorder to record the runoff hydrograph, as shown in Figure 3.3.



Figure 3.3 Rectangular flume attached to a stage recorder

3.4 Treatments

The number of treatments was five with three replications each. The treatments were; Surface applied dairy manure (DS), Incorporated dairy manure (DI), Poultry litter (PL), Inorganic fertilizer (IF) and Control (CO) with no manure or fertilizer. For the surface applied dairy manure (DS) treatment, the dairy manure was filled in buckets and uniformly spread over the plot. The dairy manure was also spread likewise in the incorporated dairy manure (DI) treatment, but it was plowed after the application. Inorganic fertilizer was manually broadcasted over the plots in the IF treatments, followed by plowing. Poultry litter was surface applied by manually broadcasting it over the plot in the PL treatments.

The treatments were assigned to each plot using a Randomized Complete Block Design (RCBD) (Ott and Longnecker, 2001). The detail of this statistical design is included in section 3.8. Plots were grouped in three blocks of five adjacent plots each. All the five treatments were assigned to each block and the treatments were assigned randomly to each plot in each block. Before application, each plot was flagged with a treatment name and an alphabetically assigned plot number.

The amount of manure and fertilizer applied to each plot was determined according to the estimated P removal rates of corn crop. The state of Virginia requires application of poultry waste according to crop P requirements. To determine the P requirement, soil samples were collected for analysis after the first plowing. A one inch diameter soil auger was used to collect 150 mm deep soil samples. The samples were collected from 5 different locations around the field and were thoroughly mixed to make a composite sample for each plot. The mixed and dried sample was sent to the Soil Testing Laboratory, Virginia Tech for chemical analysis, and a sub-sample were also sent to Soil Physics laboratory, Virginia Tech for particle size analysis. The relevant properties of the soil are shown in Table 3.1. The soil properties were used to determine the amount of nutrients required for good crop yield. Recommendations from Virginia DCR guide (VA DCR, 1995) were used to calculate the nutrient requirements. The nutrient requirement for a corn crop is also shown in Table 3.1.

Table 3.1 Average textural and chemical properties of soil at the experimental plots

Textural Properties	% Sand	% Silt	% Clay	Textural Class
	30.01	57.94	12.05	Silt
Chemical Properties	pH	Phosphorus (ppm)	Potassium (ppm)	Calcium (ppm)
	5.99	7	85	611
Crop Requirements	Nitrogen (kg/ha)	Phosphorus (kg/ha)	Nitrogen (kg/plot)	Phosphorus (kg/plot)
	170	90	1.00	0.53

Dairy manure was obtained from the Virginia Tech Dairy farm where it was stored in liquid form in a lagoon. When the dairy manure was collected, it was near the end of the storage period of about 6 months. It is a normal practice to apply dairy manure after six month period of storage in a lagoon (AWMFH, 2000). While the dairy manure was being collected, it was constantly stirred and mixed with a pump, which helped to provide a

good representative sample of dairy manure. About 1000 gallons of dairy manure was collected, and during the collection process three manure samples were also collected. These samples were shipped to the Agriculture Laboratory, Clemson University, for nutrient analysis within a day of their collection. The manure was stored in three 350 gallon capacity, plastic tanks near the experimental plots and covered with a tarp to reduce the effect of sun on the properties of manure, as the dairy manure was collected about two weeks before application. Dairy manure samples were also collected and sent for nutrient analysis at the time of application. Poultry litter was obtained from Valley Pike farm, Harrisonburg, directly from the chicken houses, just one day before the application. The poultry litter samples were collected during the time of application to the plots and were sent to the Clemson Agriculture Laboratory, Clemson for nutrient analysis. The dairy manure and poultry litter samples collected just before their application were also sent to the Water Quality Laboratory, Department of Biological Systems Engineering, Virginia Tech to conduct the bacterial analysis. The test was conducted to measure the concentration of indicator bacteria in source manure and thereby help in assessing the amount of bacteria applied to each treatment.

Since the dairy manure was collected two weeks before the experiment, there was ample time available to obtain and use the results from Clemson Agriculture Laboratory to calculate the dairy manure required for each plot. However, for poultry litter, the old nutrient results were used as a guide to calculate the litter required for each plot. Table 3.2 shows the nutrient analysis results of manure. The table also shows the date, when the samples were shipped for nutrient analysis. The results from the previous samples were used to calculate the manure required per unit area and the amount of manure applied on each plot for each treatment. The inorganic fertilizers used were Ammonium Nitrate and Triple Super Phosphate. The table also shows the nutrient composition of inorganic fertilizers.

Table 3.2 Nutrient analysis results of manure used in the experiment. The dates in parenthesis show the date, the samples were procured. Table also shows nutrient composition of inorganic fertilizer

Manure	Available Nitrogen	Phosphorus as P₂O₅
Dairy Manure (4/26/03)	8.13 (lbs/1000 gal) (Surface Applied)	6.65 (lbs/1000gal)
	10.02 (lbs/1000 gal) (Incorporated)	
Dairy Manure (5/13/03)	8.26 (lbs/1000 gal) (Surface Applied)	5.59 (lbs/1000gal)
	9.85 (lbs/1000 gal) (Incorporated)	
Chicken Litter (Previous year record)	45.00 (lbs/ton)	49.27 (lbs/ton)
Chicken Litter (5/13/03)	40.00 (lbs/ton)	62.44 (lbs/ton)
Ammonium Nitrate	34% by weight	
Triple Super Phosphate		45% by weight

The N requirement of the corn crop was estimated according to the recommendations from the Virginia DCR Guide (VDCR, 1995). When applying the manure according to the P requirement, Nitrogen (N) is generally not adequate for proper development of plant. Thus, the N requirement of the crop was fulfilled with inorganic N fertilizer. The amount of manure and fertilizer applied for each treatment are shown in Table 3.3.

Table 3.3 Amount of manure and fertilizer applied per unit area and per plot for each treatment

Treatment	Manure/fertilizer applied for P	N deficiency fulfilled by Urea	Urea applied
Dairy manure, surface applied (DS)	27909 gal/ha	148 kg/ha	435 kg/ha
	165 gal/plot	0.87 kg/plot	2.57 kg/plot
Dairy manure, incorporate (DI)	27909 gal/ha	95 kg/ha	280.4 kg/ha
	165 gal/plot	0.56 kg/plot	1.65 kg/plot
Poultry litter (PL)	3780 kg/ha	114.5 kg/ha	336.7 kg/ha
	22.35 kg/plot	0.76 kg/ha	2 kg/plot
Inorganic Fertilizer (IF)	200 kg/ha (Triple Super Phosphate)	170 kg/ha	500 kg/ha
	1.2 kg/plot	1 kg/plot	3 kg/plot
Control (C)	N/A	N/A	N/A

The plots were subdivided into 5 feet sections to ensure uniform application of manure and fertilizer in each plot. The amount of manure and fertilizer to be applied on each section was also calculated. The measured amount of manure and fertilizer were collected in small buckets and spread in the sections of respective blocks. The spreading was done manually. A tractor mounted tiller was used to incorporate the dairy manure in DI (incorporated dairy manure) and IF (inorganic fertilizer) after the application of manure and fertilizer in the respective plots. After the tilling operation, metal borders were fixed on the upslope end of all the plots.

The plots were planted with corn (*Zea Mays*). The corn variety used was “Early Sunglow Sweet”, which is a popular variety in South-West Virginia. The planting was done in rows across the contours. The planting was done by hand according to the recommendations by the seed company. A 5 cm (2”) deep furrow was constructed, in plots, 0.9 m (36”) apart. About 300 g of seed were planted in each 100’ row.

3.5 Rainfall simulation

A rainfall simulator (Dillaha et al., 1987) was installed after the plots were prepared and treated. The schematic of the layout of the rainfall simulator over the plots is illustrated in Figure 3.4. The rainfall simulations were conducted on May 14 and 15, 2003, and on June 17, 2003. Water was obtained from a local pond. Water from a main line goes to risers through laterals and then moves into sprinkler heads which revolve and spreads water in the form of rain, as shown in Figure 3.5.

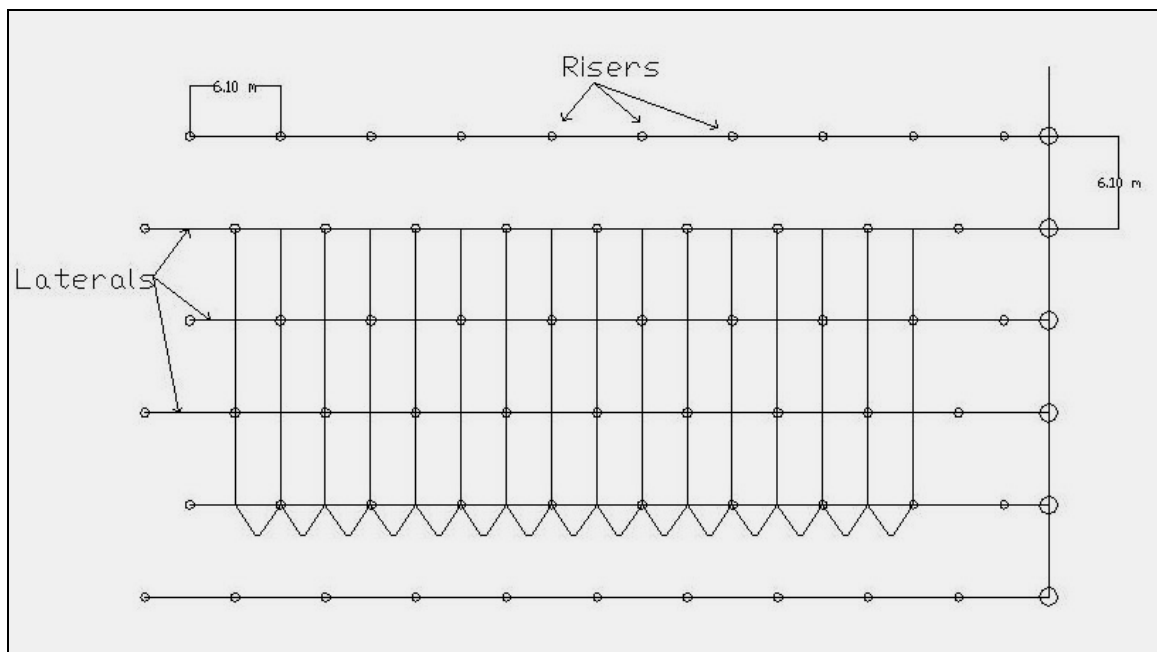


Figure 3.4 Schematic diagram of the layout of rainfall simulator over the experimental plots

A standard set of three simulation events on dry, wet and very wet soil conditions is conducted to simulate various antecedent soil moisture conditions for erosion research studies (e.g. Gilley et al., 1986). However, this being a nutrient and bacterial transport study, only two runs were conducted. The first simulation event was conducted on dry soil for an hour on May 14, 2003 and the second event was conducted on May 15, 2003 until the runoff from all the plots reached a steady state and stabilized. The total time for the second simulation event was 45 minutes. The first event was a dry run as it was conducted on dry soil, and the second event was a wet run as it was conducted on soil wetted from the simulation event on the previous day. The same sequence of simulation

events however could not be repeated after one month, due to intermittent heavy natural rainfall events. Due to wet field conditions, dry rainfall simulation event could not be conducted. The simulation event conducted on June 17, 03 represented the wet rainfall simulation event as the soil was wet due to a rainfall event occurring during the previous 24 hours. The third event was also conducted until the runoff hydrographs from all the plots stabilized and the total time of rainfall simulation was 51 minutes.

The rainfall intensity was maintained around 50 mm/hour by maintaining a pressure of 35 psi in the water supply line. The final readings as discussed in chapter 4 section 4.1.2, shows that the average intensity of rainfall was about 48 mm/hour in the first and second simulation event and 50 mm/hour for the third simulation event.



Figure 3.5 Rainfall simulator in operation

Before the start of rainfall simulation events, each plot was assigned to a student who helped to sample the runoff water. They were provided with 15, 200 ml sampling bottles for collecting samples. Each student was also provided with a measuring bucket and a stop watch to measure the flow rate after sampling.

3.6 Sampling Procedure

Samples were collected after the initiation of visible runoff from the plots. Runoff start time from each plot was recorded, and the samples were collected at each three minute intervals. Initially, the rise in the hydrograph from individual plots was steep and hence the time interval was kept as 3 minutes. As soon as the rise in hydrograph started decreasing, the sampling interval was increased to 6 minutes. About 200 ml of runoff flowing through the flume was collected at each sampling interval by the students. Flow rate was also measured just after each sampling with a measuring bucket and a stop watch. This flow rate was measured to verify the flow rates obtained from each stage recorder hydrograph.



Figure 3.6 A graduate student (Tess Wynn) collecting samples of runoff flowing through the flume

Two minutes after the completion of each simulation event, one additional runoff sample was collected and the time at which runoff stopped. The first simulation event was conducted for an hour and 9-12 samples were collected from each plot. The number of samples collected depended upon the time for runoff to start from individual plots. The second simulation was conducted for 45 minutes and 8-9 samples were collected from

each plot. The third simulation was 51 minutes long and 10-12 samples were collected from each plot.

3.7 Additional Monitoring

The raingage readings were noted after each simulation event to measure a rainfall amount. Raingages were also used to measure the natural rainfall between simulation events. Soil was tested for antecedent soil moisture conditions before the simulations by the gravimetric method (Black, 1965). Soil moisture was not measured before the first two simulation events and antecedent moisture conditions prior to the third simulation only was recorded. Before the experiment, soil samples were collected to analyze the nutrient content of soil and conduct particle size analysis, as described in Section 3.4 and shown in Table 3.1.

Canopy cover was also calculated 6 weeks after planting using digital pictures of each plot. These pictures were analyzed using ArcGIS 8.2 (ESRI Inc., 1999) and the green pixels were calculated. The ratio of green pixels to total number of pixels provided an estimate of canopy cover. To account for variation, three pictures per plot were used to calculate canopy cover.

3.8 Flow weighting the samples

Water samples were transported from the experiment site to the Water Quality Laboratory at the Department of Biological Systems Engineering, Virginia Tech after each simulation event. Stage recorder hydrographs for each plot were digitized with a digitizer and HAS software (Carr et al., 1988). The results provided information on the amount of each sample required to make 1 liter of flow weighted composite sample for each plot. Individual samples were mixed before preparing the flow weighted composite samples. Samples were tested for indicator bacteria within 5 hours of the simulation runs. The rest of the samples were stored in a cooler to preserve and tested later for nutrient content.

3.9 Laboratory analysis

The flow weighted composite samples were tested for nutrient content and indicator bacteria counts. The name and number of all the tests performed on the runoff water samples are listed in Table 3.4.

Table 3.4 Water quality parameters and test number conducted on runoff water samples

Sample	Test
Escherichia Coli (EC)	EPA 1603 9215 C (for manure sample)
Fecal Coliform (FC)	SM 9222 D 9215 C (for manure sample)
Enterococcus (ENT)	SM 9230 C 9215 C (for manure sample)
Dissolved Reactive Phosphorus (DRP)	EPA 365.2
Total Phosphorus (TP)	EPA 365.3
Bioavailable Phosphorus (BAP)	EPA 365.2
Ammonium Nitrogen (NH₄⁺-N)	EPA 350.2
Nitrate Nitrogen (NO₃⁻-N)	EPA 353.3
Total Suspended Solids (TSS)	EPA 160.2
Total Nitrogen	HACH 10071/10072

The tests for FC, EC, and enterococcus in runoff were performed within 5 hours of sample collection. The appropriate sample volume was passed through a membrane filter (0.45 µm) to retain the bacteria present. The filter was placed on an agar medium (in a Petri dish) that is selective for coliform or Enterococci growth. The Petri dish containing the agar medium was incubated, upside down for 24-48 hours, at a temperature range of 35-44.5° C. After the incubation period, the colonies developed were counted using a low power microscope.

To calculate the TSS, well mixed sample was filtered through a pre-weighted standard glass-fiber filter and the residue retained on the filter was dried to a constant weight in an oven at 103 to 105° C. The analysis procedure for the DRP and the BAP was the same. The analysis for DRP was conducted on a filtered sample and the BAP analysis was conducted on the unfiltered sample. The orthophosphate in the sample was reacted with ammonium molybdate and antimony potassium tartrate in an acid medium resulting in the formation of a heteropoly acid-phosphomolybdic acid. This heteropoly acid-phosphomolybdic acid reduced to intensely colored molybdenum blue by ascorbic acid. The intensity of the color is proportional to the amount of orthophosphorus in the sample.

To calculate the total P, the runoff sample was heated in the presence of sulfuric acid and potassium persulfate and the rest if the reaction is same as that for DRP.

The NH_4^+ -N in the filtered sample was reacted with chlorine to form monochloramine. Monochloramine was reacted with salicylate to form 5-aminosalicylate. The 5-aminosalicylate was oxidized in the presence of a sodium nitroprusside catalyst to form a blue colored compound. This blue color was masked by yellow color from the excess reagent resulting in a green color. This green color was proportional to the amount of NH_4^+ -N in the sample. To analyze the nitrate, the filtered solution was reduced to form nitrite from nitrate with the help of cadmium metal. The nitrite ion was reacted in an acid medium with sulfanilic acid to form a diazonium salt. The salt was coupled with gentisic acid to form amber colored solution which is proportional to the NO_3^- -N concentration of the sample. To calculate TN, the sample was heated in the presence of alkaline persulfate, converting all forms of nitrogen to nitrate. Sodium metabisulfate was added after heating. Nitrate then reacted with chromotropic acid under strongly acidic conditions to form a yellow complex which is proportional to the amount of TN in the sample.

3.10 Statistical Analysis

The experiment design was a Randomized Complete Block Design (RCBD) (Ott and Longnecker, 2001). The experiment consisted of 5 treatments with 3 replications each, making 15 experimental units or plots. Blocks were introduced to avoid the error due to the location of plots. The plots were divided in three blocks with five adjacent plots. All 5 treatments were randomly assigned to the plots in each block.

The analysis was conducted with the help of a statistical software SAS (SAS Inc.). The “proc glm” method was used in SAS to conduct the analysis of variance. The significance level of 5% was used to calculate the significant difference between the treatments and the simulated events. Adjusted Tukey method was used for comparison of treatment means. The null hypotheses assumed for the analysis were no difference between treatments, no difference between simulation events and no interaction between simulation events and treatments.

Chapter 4 Results and Discussion

The laboratory analyses of the water samples were completed within one month of each simulated rainfall event, except for the bacterial analysis which was performed within five hours of sample collection. The results were plotted for visual comparison among the treatments and simulated events. The analysis was also carried out statistically with SAS (SAS Institute, Inc.) statistical software to evaluate the effect of different treatments and simulated events on the water quality parameters. As described in Chapter 3, section 3.5, the simulated rainfall events were carried out at three different times; on May 14 and 15 and finally on June 17, 2003. The comparison of the first two simulated events indicates the effect of antecedent soil moisture conditions on the runoff and the reduction in pollutant concentration and load due to the significant loss of pollutants during first simulated event. However, the comparison between the second and third simulated events evaluates the effect of time elapsed between the two simulated events and the amount of natural rainfall and runoff which occurred between the two events. As recorded by the raingages installed in the field, about 270 mm of rainfall occurred between the second and third simulated rainfall events.

The results and discussion is divided into three sections; hydrology, nutrients and bacteria. Each section describes the concentration of respective components in runoff and explains the underlying reasons behind the results. The amount of each pollutant lost in runoff is also calculated to integrate the effects of the pollutant's concentration and runoff amount. In the section on bacteria the number of indicator bacteria applied and indicator bacteria lost in runoff are also calculated. As shown in Section 3.4, Table 3.1, the five treatments are labeled by two letter symbols and the same symbols are used throughout this chapter.

4.1 Hydrology

This section discusses the hydrologic response of the treatments to the simulated rainfall events. The canopy cover is included as a subsection as it affects the hydrologic response of various treatments. The subsection on rainfall provides information about the amount of rainfall applied and the uniformity of rainfall application. In the runoff subsection, time to the start of runoff, the peak runoff rate and the volume of runoff from each treatment are discussed. The subsection on TSS discusses the concentration and yield of Total Suspended Solids (TSS) in runoff from the treatments.

4.1.1 Canopy Cover

The corn plants started emerging in about 10 days after the second simulated event. Plants varying from 10-25 cm in height could be observed before the third simulated event. Thinning was performed five days before the third simulated event to assist in development of healthy plants. Herbicide 2, 4-D was also applied just after thinning to inhibit the development of weeds. Canopy cover was recorded one week after the third simulated event. Although canopy cover was recorded one week after the third simulated event, it gives us a good idea of the differences in canopy development among the treatments. To calculate canopy cover, digital pictures of treatments were taken and the relative area covered by green pixels was calculated using ArcGIS 8.2 (ESRI Inc., 1999). The average canopy cover for each treatment is shown in Table 4.1.

Table 4.1 Average canopy cover development from various treatments, 42 days after planting of corn

Treatments	Average Canopy Cover
Incorporated Dairy Manure (DI)	27.03%
Poultry Litter (PL)	46.34%
Surface applied Dairy Manure (DS)	45.61%
Inorganic Fertilizer (IF)	48.69%
Control (CO)	29.13%

Although the maximum canopy development was observed on the Inorganic Fertilizer (IF) treatment, Poultry Litter (PL) and Surface applied dairy manure (DS) treatments had canopy cover similar to the IF treatment. These results indicate that inorganic fertilizer is a more immediate supplier of plant available nutrients than the other treatments, followed closely by poultry litter and dairy manure. However, when the dairy manure was incorporated, it exhibited different results. The canopy cover from DI treatments is almost equal to the canopy cover from CO treatment, which may be due to the immobilization of some plant nutrients. Since the manure was thoroughly mixed into the soil and the soil was nutrient deficient, as reported in Chapter 3, Section 3.4, most of these nutrients might have been adsorbed on the soil particles. The canopy cover in the Control (CO) treatments was higher than that on the Incorporated Dairy Manure (DI) treatment but the difference was not statistically significant.

4.1.2 Rainfall

Rainfall was applied using a rainfall simulator. The first simulated rainfall event was conducted on May 14, 2003. Forty eight mm of rainfall was applied in a period of one hour, with a uniformity coefficient of 95.3%. Runoff samples were collected for TSS, nutrients, and bacterial analysis. The second simulated rainfall event was conducted on May 15, 2003. An overnight rain of 0.87 mm was also recorded on May 15, 2003 before the second simulated event. This additional rainfall amount was not significant enough to cause a runoff event from the plots. During the second simulated event, 36 mm of rainfall was applied over a period of 45 minutes with a uniformity coefficient of 93.0%. The reduction in uniformity coefficient was due to the excessive wind speed observed during the second simulated event.

The third and last simulated rainfall event was conducted on June 17, 2003. A total of 270 mm of rainfall was recorded during the period between the second and third rainfall simulated events. During the third simulated event, a total rainfall amount of 42.8 mm was applied in 51 minutes with a uniformity coefficient of 91.4%. The reduction in uniformity coefficient was due to the failure of a few nozzles during the simulation.

4.1.3 Runoff

Runoff rate and volume from the plots were recorded using a stage recorder and manual measurements using a stop watch and a 5-liter bucket. The runoff starting time varied for different treatments. The time to start runoff was greatest for DS treatments and was shortest for DI treatments, compared to rest of the treatments investigated. The descriptive statistics for the time to initiate runoff are shown in Table 4.2. The mean runoff initiation time for each treatment in all the simulated rainfall events is illustrated in the Figure 4.1.

Table 4.2 Descriptive statistics for the time to initiate the runoff from each treatment for all the simulated events*

Treatments	Simulation 1 ^a			Simulation 2 ^b			Simulation 3 ^b		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	22:25 ^A	23:35	20:40	03:22 ^A	05:25	01:40	05:57 ^A	10:50	03:30
PL	19:28 ^A	23:00	14:55	03:04 ^A	04:40	01:41	04:47 ^A	07:45	03:06
DS	22:40 ^A	25:30	20:00	05:46 ^A	10:40	01:39	07:42 ^A	11:30	03:06
IF	21:09 ^A	24:30	17:06	04:00 ^A	07:10	01:39	06:20 ^A	10:20	03:10
CO	18:17 ^A	20:20	15:30	03:10 ^A	04:38	01:42	04:05 ^A	07:20	01:40

*The values with different uppercase letter within a column are significantly different. The simulated events with different lowercase letters are significantly different.

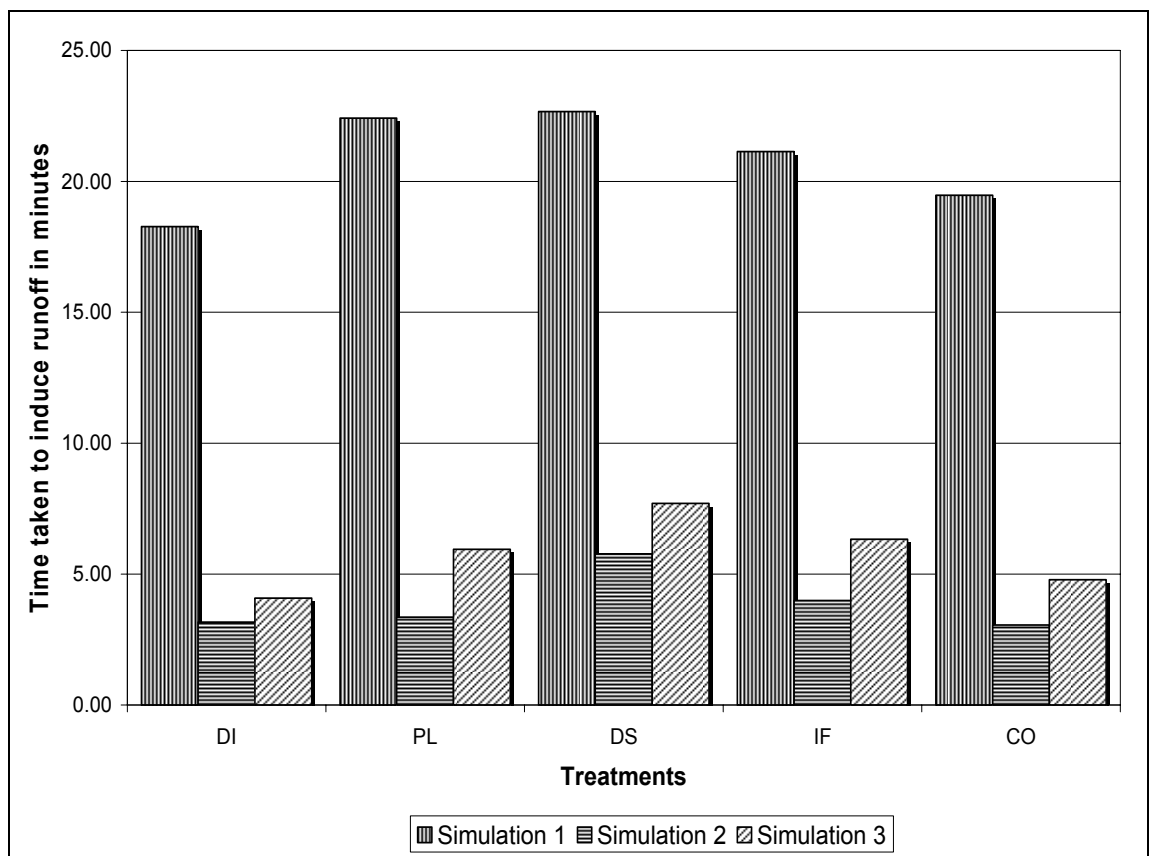


Figure 4.1 Mean time to initiate the runoff for each treatment during the three simulated rainfall events

The simulated events had a significant effect on the time to start of runoff from the treatments. Statistical results suggest that the time to start runoff for the first simulated event was significantly different from the other two, while those for the second and third simulated events were not different from each other. The average time taken for runoff to start was about 21 minutes in the first simulated event. In the second simulated event,

the time to initiate the runoff was reduced by about 17 minutes to 4 minutes. Before the first simulated event, the soil was dry and although soil samples were not collected before the first and second simulated events for moisture analysis, it was clearly visible that before the second simulated event, the soil was saturated due to the first simulated rainfall event. The infiltration capacity of the soil was satisfied, which resulted in runoff to starting as soon as rainfall was applied. The average runoff start time was about 6 minutes for the third simulated event. Before the third simulated rainfall event, the soil was also wet due to the natural rainfall event which occurred the day before the simulated event. The average soil moisture content before the third simulated event was 26.47%. The roughness of soil surface was also reduced due to the heavy rainfall events which occurred between the second and third simulated rainfall events. This decrease in roughness helped increase runoff volume from the soil surface. However, in the one month time period between the second and third simulated rainfall events, the canopy had developed and covered 30 to 45% of the plot area for various treatments. The canopy cover provided resistance to the rain drop impact on the soil and hence the time to initiate the runoff was greater during the third simulated event, compared with that recorded for the second simulated event.

Statistical analysis suggested no significant effect of treatments on the time to start of runoff within each simulated event. Since conventional tillage was performed on all the plots, which were adjacent to each other, with very little hydrologic differences, the hydrologic response time was not significantly different for various treatments. The minor variation observed could be attributed to the physical properties of manure and fertilizer applied on the plots. Some variations could also be attributed to the different nutrients application methods discussed previously in this section.

No significant interaction was observed among different treatments and the simulated rainfall events. Thus, the relative behavior of treatments with each other remained the same for all three simulated events. There were also no significant interaction between simulated events and treatments with regard to peak runoff rate and runoff volume.

The peak runoff rates from all the treatments were calculated in mm/hour. Variation in peak runoff rates among different treatments and simulated events is shown in Table 4.3, and is graphically illustrated in Figure 4.2. The DS treatments exhibited the lowest

peak runoff rate for the first rainfall simulated event. As seen earlier, the DS treatments also had the longest time to initiate the runoff. It can be inferred that the dairy manure, when surface applied, has an impeding effect on runoff. When the dairy manure was surface applied, the soil was dry, and the liquid from manure infiltrated into the soil. The solid portion of manure formed a layer over the soil surface. Since the manure was applied earlier on the day of May 13 and simulated rainfall event was conducted later in the day on May 14, ample time was available for the upper portion of manure layer to dry. The dry and windy weather conditions also helped this manure layer to dry quickly. When the simulated rainfall event was conducted on the following day, the layer of organic material over soil absorbed the water and acted as a sponge to reduce runoff volume. This organic layer worked as mulch over the soil surface. This thin layer of organic material was observed even 10 days after the third simulated rainfall event. The highest peak runoff rate was observed from IF treatment compared to all other treatments.

Table 4.3 Descriptive statistics for the peak runoff rate from the treatments during each simulated event (mm/hour)*

Treatments	Simulation 1 ^a			Simulation 2 ^b			Simulation 3 ^a		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	28.89 ^A	39.15	19.57	37.26 ^A	48.41	30.34	26.85 ^A	29.48	22.08
PL	26.57 ^A	30.45	24.36	34.75 ^A	35.37	34.37	19.04 ^A	23.88	11.82
DS	20.48 ^A	24.91	18.27	34.65 ^A	38.80	30.66	26.52 ^A	35.11	18.26
IF	34.90 ^A	39.15	31.32	38.40 ^A	40.31	36.09	24.86 ^A	26.99	21.41
CO	25.81 ^A	30.45	19.57	29.67 ^A	34.49	26.31	23.91 ^A	24.83	23.15

*The values with different uppercase letter within a column are significantly different. The simulated events with different lowercase letters are significantly different.

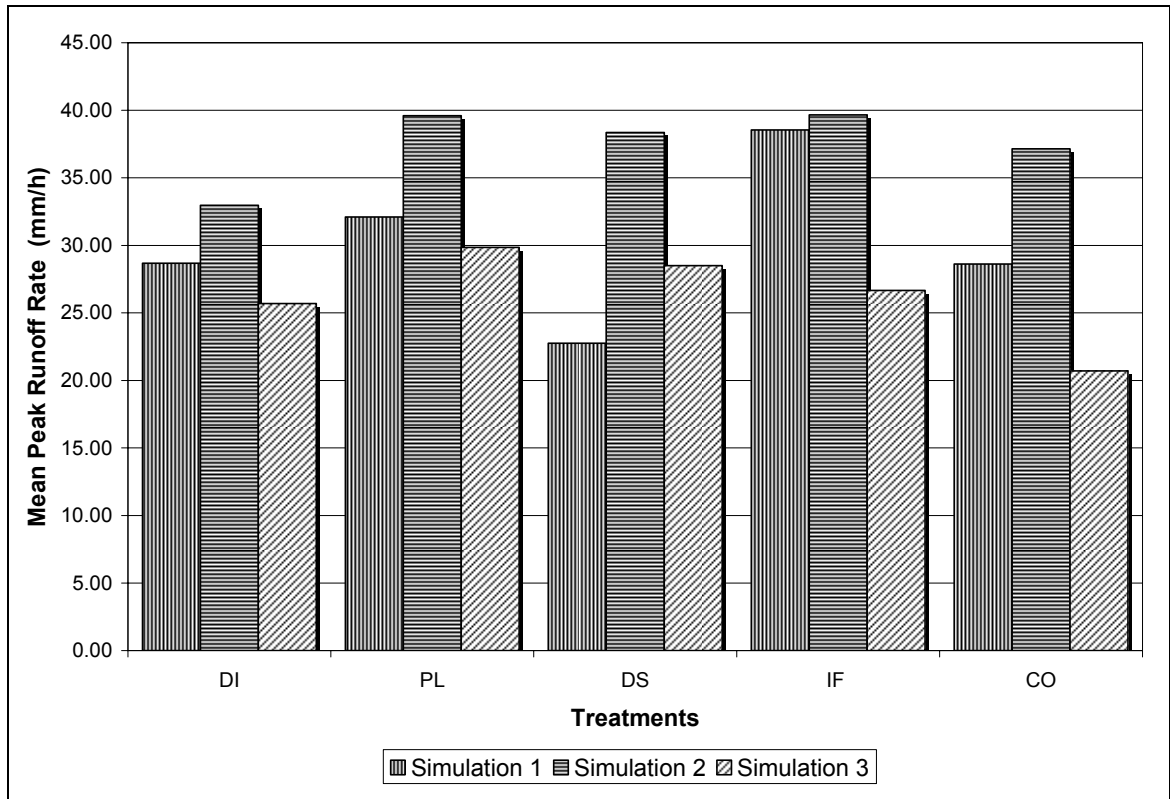


Figure 4.2 Mean peak runoff rate from each treatment for all the three simulated rainfall events

The change in peak runoff from the first to second simulated event could be attributed to the change in hydrologic properties of the plots due to excessive rainfall. The soil was dry and freshly tilled before the first simulated event, which resulted in a rough soil surface. The rough surface allowed soil to retain water and reduce runoff. The dry soil also allowed the water to infiltrate throughout the first simulated event and consequently reduced the runoff. During the second simulated event, the soil roughness was reduced due to the rainfall event on the preceding day and the soil was also saturated. These conditions helped the runoff volume and peak runoff to increase significantly from the first simulated event to the second simulated event. The soil conditions before the second and third simulated events were approximately the same due to the occurrence of rainfall events during the preceding day. During the third simulated event, significant canopy development was observed, which intercepted the rain drops and hence reduced runoff. Statistically significant effect of simulated events on the peak runoff was observed. The peak runoff rate recorded in second simulated event was significantly higher than the peak runoff rate recorded from the first and third simulated rainfall events.

The runoff volumes from each treatment were calculated and are reported in millimeters. The mean runoff volumes recorded for each treatment for all three simulated events are shown in Table 4.4, and are graphically illustrated in Figure 4.3. Statistical analysis suggests significant treatment effects on runoff volume. The minimum runoff volume was observed from DS treatment for the first simulated event. During the first two simulated events, runoff volume from IF treatment was significantly different than those measured for the DS treatments. The reason for the low runoff volume measured for DS treatment is the mulching effect of dairy manure over the soil surface as explained earlier in the section. The effect of treatment was not observed for the third simulated rainfall event.

Table 4.4 Descriptive statistics for the runoff volume from the treatments during each of the simulated rainfall events*

Treatments	Simulation 1 ^a			Simulation 2 ^b			Simulation 3 ^a		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	7.72 ^A	10.56	6.25	14.13 ^A	16.46	12.86	10.63 ^A	14.72	3.66
PL	9.37 ^A	14.23	4.71	14.18 ^A	14.74	13.41	6.41 ^A	8.59	2.08
DS	4.16 ^{AB}	5.92	3.18	10.63 ^{AB}	12.72	8.70	7.78 ^A	11.37	5.62
IF	14.22 ^{AC}	16.11	12.66	16.89 ^{AC}	17.43	15.88	9.25 ^A	11.35	6.52
CO	7.42 ^A	9.40	5.01	11.26 ^A	14.14	8.93	9.24 ^A	11.59	7.98

*The values with different uppercase letters within a column are significantly different from each other. The simulated events with different lowercase letters are significantly different from each other.

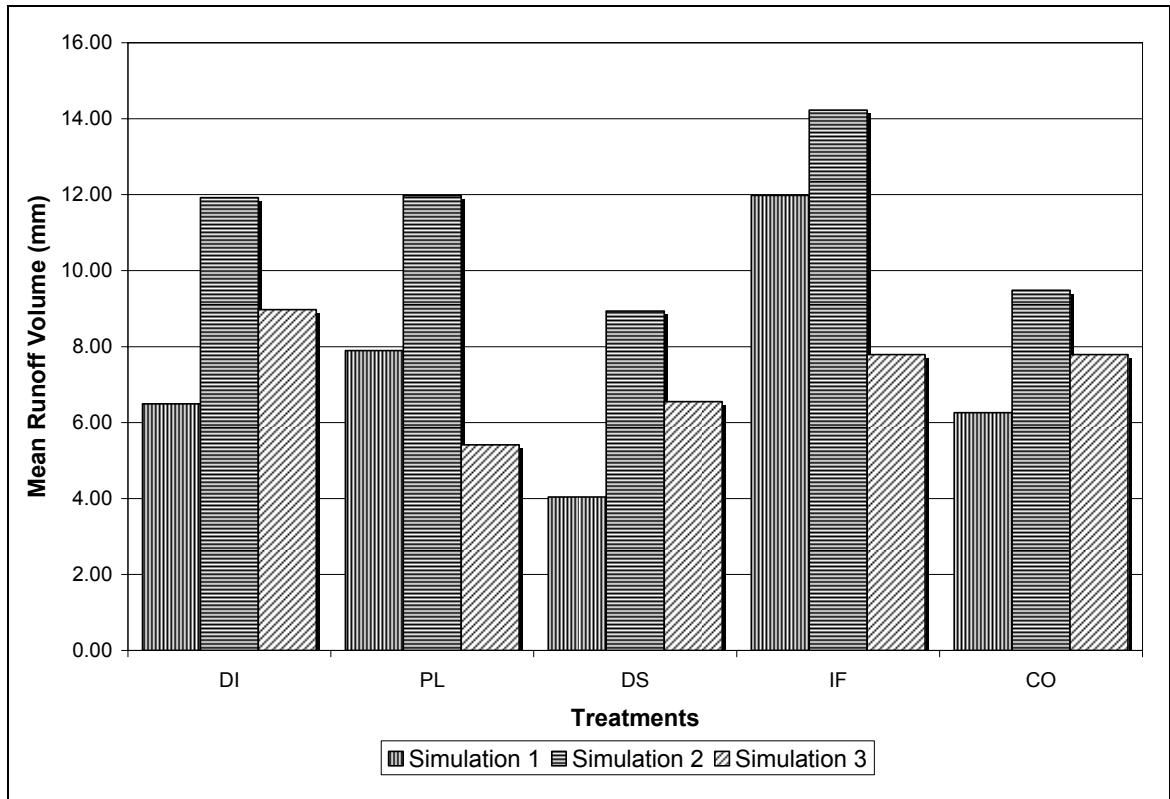


Figure 4.3 Mean runoff volume resulted from different treatments during each simulated rainfall events

Statistical analysis suggested that the first simulated event produced significantly lower runoff volume than the second event and the second event resulted in more runoff volume than the third simulated event. In the first simulated event, of the 48 mm of rainfall applied to plots, about 15% was lost as runoff, while in the second simulated event, 32% of the applied rainfall water was lost as runoff. Before the second simulated event, the plots were already wet and the soil was saturated, because of the simulated rainfall event on the previous day. During the third simulated event, about 17% of the 42.8 mm rainfall applied on plots was lost as runoff. The ground was saturated before the third simulated event also, because of 270 mm of total natural rainfall during the entire month and the significant rainfall event on the day preceding the third simulated event. The soil was wet, but the lower runoff volume during the third simulated event, compared with the volume during the second simulated event, can be attributed to increased canopy cover at the time of third simulated event.

The results obtained in this study are similar to those from previous investigations where no significant effects of different manure and fertilizer treatments have been observed on runoff from the cropland (Wood et al., 1999; Edwards and Daniel, 1993, 1994).

4.1.4 Total Suspended Solids (TSS)

The mean TSS concentration of runoff for different treatments and simulated events is illustrated in Figure 4.4. The descriptive statistics for the TSS concentrations of runoff are shown in Table 4.5. The highest TSS concentration was observed from PL treatment (4.13 g/l) in the first simulated event and it was significantly higher than that from the DS treatment. The dry and loose poultry litter made both soil and organic material susceptible to suspend in and move with runoff as soon as the rainfall event occurred, Concentrations of TSS in runoff from plots treated with dairy manure, (DI (2.7 g/l) and DS (2.9 g/l)) were lowest among all the treatments for the first simulated event. Dairy manure was applied in liquid form, and in the 24-28 hour period between manure application and the first simulated event, the moisture available in the manure bonded the loose soil particles together and reduced the TSS loss from dairy manure treated plots.. Similar values of TSS concentrations in runoff were reported by Vories at al. (2001) from cotton fields in a long term field study.

Table 4.5 Descriptive statistics of the TSS concentrations in g/ml in runoff from different treatments for each simulated rainfall event*

Treatments	Simulation 1 ^a			Simulation 2 ^b			Simulation 3 ^c		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	3.82 ^A	4.38	3.28	3.32 ^A	3.44	3.09	0.28 ^A	0.43	0.10
PL	4.13 ^{AB}	4.55	3.43	2.84 ^A	3.32	2.25	0.27 ^A	0.43	0.06
DS	2.99 ^{AC}	3.84	2.47	2.84 ^A	3.46	2.47	0.19 ^A	0.37	0.04
IF	3.64 ^A	4.21	2.85	3.11 ^A	3.38	2.95	0.14 ^A	0.22	0.07
CO	3.41 ^A	3.82	3.19	2.66 ^A	3.17	2.10	0.29 ^A	0.50	0.08

*The values with different uppercase letters within a column are significantly different from each other. The simulated events with different lowercase letters are significantly different from each other.

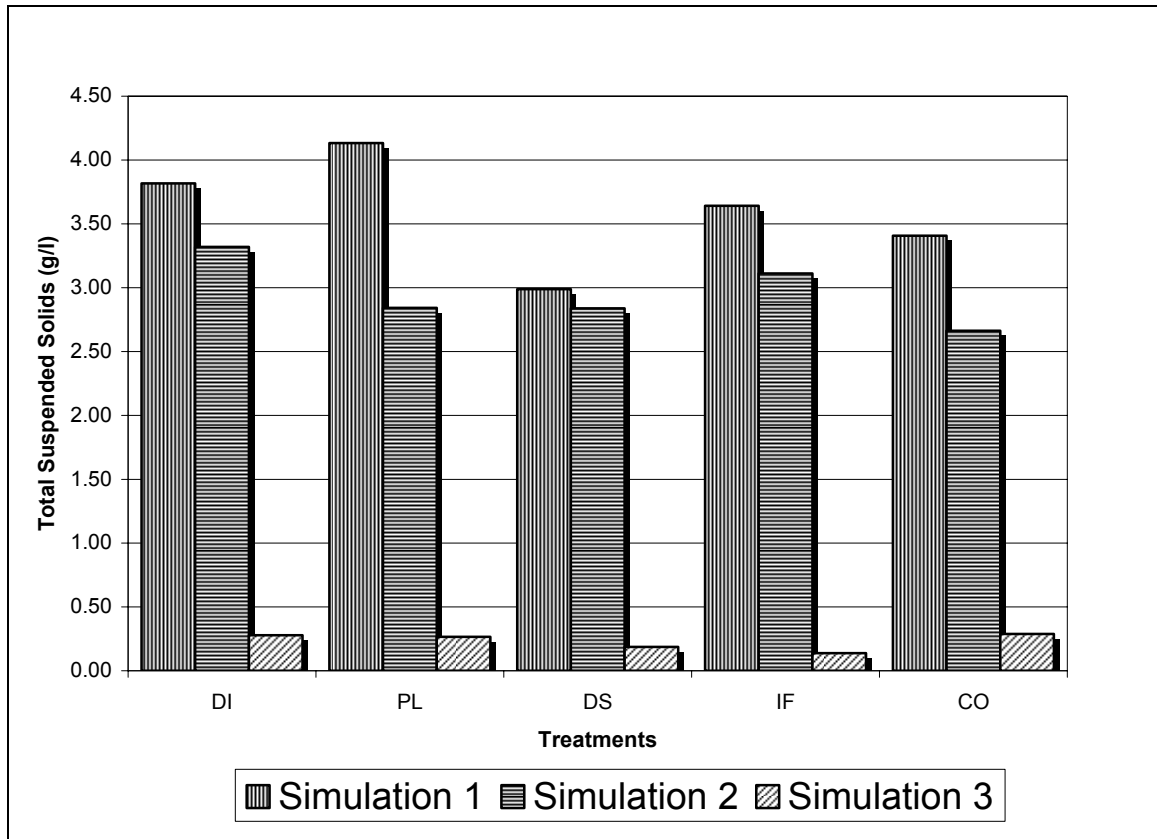


Figure 4.4 Mean TSS concentration in runoff from different treatments for all the simulated events

No significant treatment effect was observed for the TSS concentrations in runoff from second and third simulated rainfall events. Overall, statistical comparison showed that all the TSS concentrations in runoff from the first simulated event were significantly higher than the concentrations from the second and third simulated event. Furthermore, the TSS concentrations in runoff from PL from second simulated event were significantly higher than the third simulated event. The TSS concentrations for the third simulated event were reduced by more than 90%, compared with TSS concentrations in the second simulated event for all treatments. The reduction in TSS concentrations for all the treatments from first to second simulated rainfall event could be attributed to significant losses of loose soil particles and organic material on the soil surface during the first rainfall event. The reduction in TSS concentrations from second to third simulated event was significantly higher than the reduction from first to second simulated event because several natural rainfall events occurred between the second and third simulated rainfall events. Statistical analysis showed no significant interaction between simulated rainfall events

and treatments. This shows that the relative behavior of treatments with each other was the same for all the simulated events.

The descriptive statistics for TSS yield in runoff from each treatment are shown in Table 4.6 and graphically illustrated in Figure 4.5. The TSS yield from IF treatment was higher than those from the other treatments for first and the second simulated event. The inorganic fertilizer was incorporated with a tiller, which loosened the soil, thus making it more susceptible to move with runoff; this effect, coupled with the highest runoff volume from the IF treatment, compared to other treatments, resulted in the highest TSS loadings from IF treatment. Although dairy manure was also incorporated after its application in the DI treatment, the liquid in dairy manure helped the loose particles and organic material to bind and prevented them from suspending in runoff. The lower volume of runoff from the DI treatment compared to other treatments, except DS, also helped reduce the TSS loadings from the DI treatment. Total Suspended Solid yield in runoff from DS treatment were lowest among all the simulated events; this can be attributed to the smallest runoff volume recorded for DS treatment and smallest TSS concentrations, compared to all treatments, except for the DI treatment.

Table 4.6 Descriptive statistics of the TSS (kg/ha) yield in runoff from different treatments for each simulated event

Treatments	Simulation 1			Simulation 2			Simulation 3		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	294.53	400.71	204.83	470.39	565.17	404.18	35.38	58.03	3.81
PL	404.74	647.85	161.70	405.06	478.53	302.11	21.51	36.94	1.33
DS	131.82	227.16	83.74	301.91	362.63	215.33	12.16	23.33	4.21
IF	522.44	622.26	360.74	526.34	586.07	467.87	13.94	24.85	4.70
CO	256.82	359.03	159.62	290.33	297.33	282.77	26.38	40.24	6.91

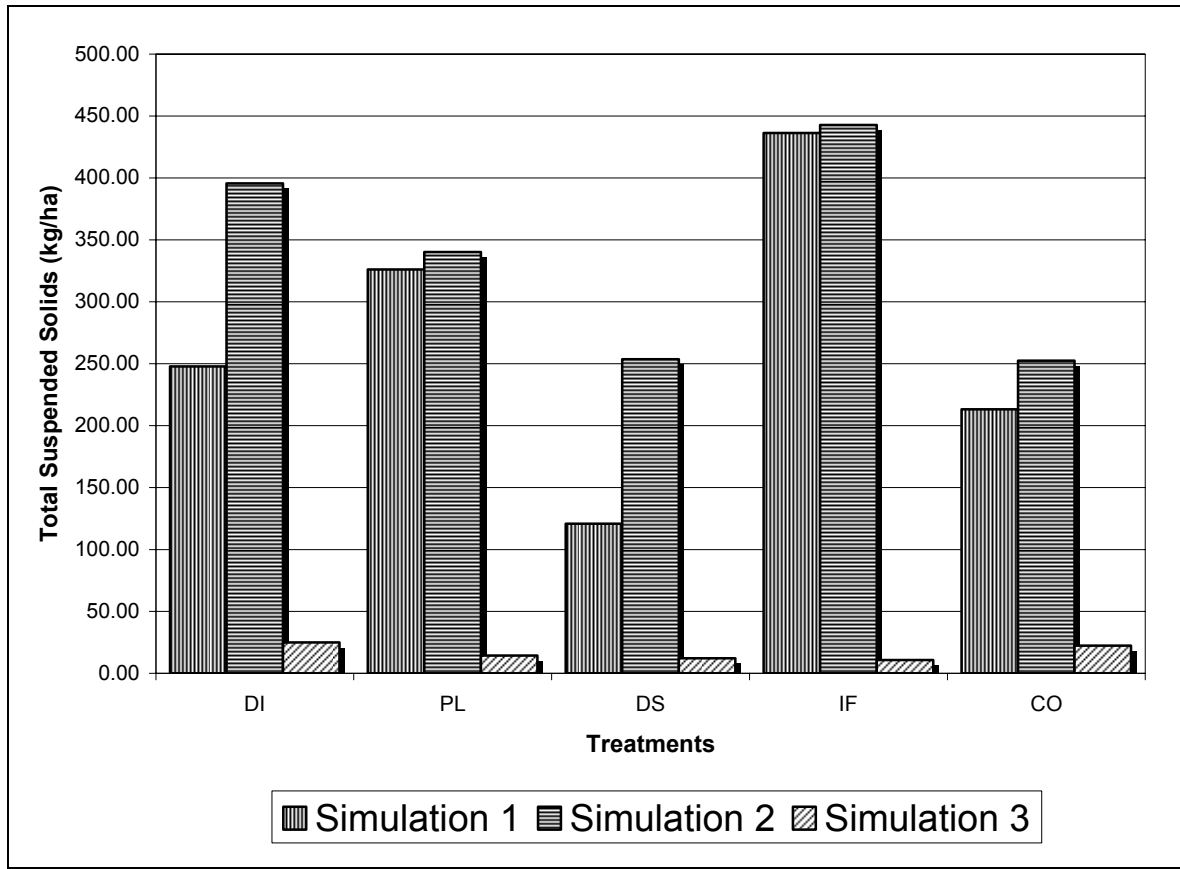


Figure 4.5 TSS yield in surface runoff, per unit area from different treatments for all the simulated events

4.1.5 Summary of Hydrology Results

The hydrology results showed that the liquid nature of dairy manure, whether surface applied or incorporated, helped reduce runoff and erosion from cropland, compared to the other treatments. However, surface application was more effective than incorporation with regard to plant development. Time to initiate runoff was greatest for the DS treatments, which also exhibited minimum runoff loss and minimum TSS yield in runoff from the first two simulated events. The PL treated plots exhibited highest TSS concentrations in runoff. Heavy rainfall events such as the events simulated in the present study, shortly after the application of poultry litter can lead to the loss of a significant amount of organic material present on the surface. Highest TSS yields were observed from the IF treatment compared to other treatments due to incorporation of fertilizer and disturbance of the soil surface shortly before the first simulated rainfall event. It can be concluded that the TSS loadings can be reduced by avoiding application of dry manure (such as poultry litter) and inorganic fertilizer, if inclement weather is

imminent. The heavy rainfall events in the period between the second and third simulated events helped in smoothening of the soil surface and washed of loose soil and organic particles on the surface. The hydrologic properties of all the treatments were very similar during the third simulated rainfall event.

4.2 Nutrients

All Nutrient analyses were conducted within one month of sample collection. The samples were stored in a cooler until analysis. The results of nutrient analysis are discussed in detail in the following sections.

4.2.1 Nutrient composition of manure

Dairy manure samples were sent for nutrient analysis to Clemson University, Clemson as soon as it was procured. The purpose of this analysis was to determine the amount of dairy manure to be applied to treatments to satisfy the Phosphorus (P) requirement of the corn crop. Samples were also collected just before the land application of manure and sent for analysis to assess if any changes occurred in manure composition during the 15 day storage period.

The chicken litter was procured one day before its application. The nutrient analysis records of chicken litter which were about six months old were used to calculate the amount of chicken litter to be applied to cropland to satisfy the P requirement of the crop. Samples of chicken litter were also collected before its application and sent to Clemson University, Clemson to measure the nutrient content. The manure did not have enough nitrogen (N) to fulfill the N requirements of crop, since they were applied based on crop phosphorus (P) requirements. Therefore, the N requirements of the crop were fulfilled by applying urea, as discussed in Chapter 3, section 3.4.

The nutrient analysis of dairy manure and poultry litter samples are summarized in Table 4.6. Table 4.6 also shows the nutrient composition of inorganic fertilizer. The P content of the dairy manure was reduced by about 16% during the storage and no significant differences were observed in available N content during the storage period. The P content of chicken litter was 26% higher and the available N was 11% lower than the same recorded for previous year analysis. Since, the earlier analysis results of dairy manure and previous year record of chicken litter were used to calculate the nutrient

requirement of the crop; we ended up applying about 26% more P and 11% less N to the PL treatment, while we applied 16% less P for both DI and DS treatments.

Table 4.7 Average nutrient composition for dairy manure samples, poultry litter samples and inorganic fertilizer

Manure	Available Nitrogen	Phosphorus as P₂O₅
Dairy Manure (before storage)	8.13 lbs/1000 gal (Surface Applied)	6.65 lbs/1000gal
	10.02 lbs/1000 gal (Incorporated)	
Dairy Manure (after storage)	8.26 lbs/1000 gal (Surface Applied)	5.59 lbs/1000gal
	9.85 lbs/1000 gal (Incorporated)	
Chicken Litter (Previous year record)	45.00 lbs/ton	49.27 lbs/ton
Chicken Litter	40.00 lbs/ton	62.44 lbs/ton
Ammonium Nitrate	34% by weight	
Triple Super Phosphate		45% by weight

4.2.2 Dissolved Reactive Phosphorus (DRP)

The DRP analysis result indicated the amount of dissolved reactive P in the runoff. This form of P is readily available to aquatic plants for their biological development. However, DRP does not include all the readily available P for biological growth. Some reactive P is also bound to sediments (Sharpley, 1997). The analysis of bioavailable P, which includes reactive P in both the dissolved and sediment forms, is discussed in section 4.2.3.

Table 4.8 shows the descriptive statistics for the concentration of DRP in runoff from different treatments for all the simulated events. Mean DRP in runoff from all the treatments for each simulation are illustrated graphically in Figure 4.6. The DRP concentrations in runoff from the PL treatment for the first and second simulated events (4.76 and 3.49 mg/l, respectively) were 5 to 10 times higher than those measured for other treatments. Since the amount of P applied on all the treatments was approximately the same, high concentrations of DRP from the PL treatment suggests high availability of dissolved P in poultry litter compared to dairy manure. This result is in accordance with the results from Sharpley and Moyer (2000), who showed that the poultry litter contains more water soluble P than dairy manure.

Table 4.8 Descriptive statistics for the DRP concentrations of runoff from different treatments for each simulated event*

Treatments	Simulation 1 ^a			Simulation 2 ^b			Simulation 3 ^c		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	0.37 ^A	0.58	0.26	0.45 ^A	0.65	0.27	0.24 ^A	0.40	0.10
PL	4.77 ^B	7.60	3.20	3.49 ^B	5.80	2.12	0.20 ^A	0.40	0.00
DS	0.96 ^A	1.52	0.42	1.21 ^C	1.59	0.92	0.21 ^A	0.23	0.20
IF	0.84 ^A	1.41	0.40	0.45 ^A	0.54	0.39	0.17 ^A	0.21	0.10
CO	0.37 ^A	0.47	0.20	0.44 ^A	0.62	0.22	0.25 ^A	0.40	0.00

*The values with different uppercase letters within a column are significantly different from each other. The simulated events with different lowercase letters are significantly different from each other.

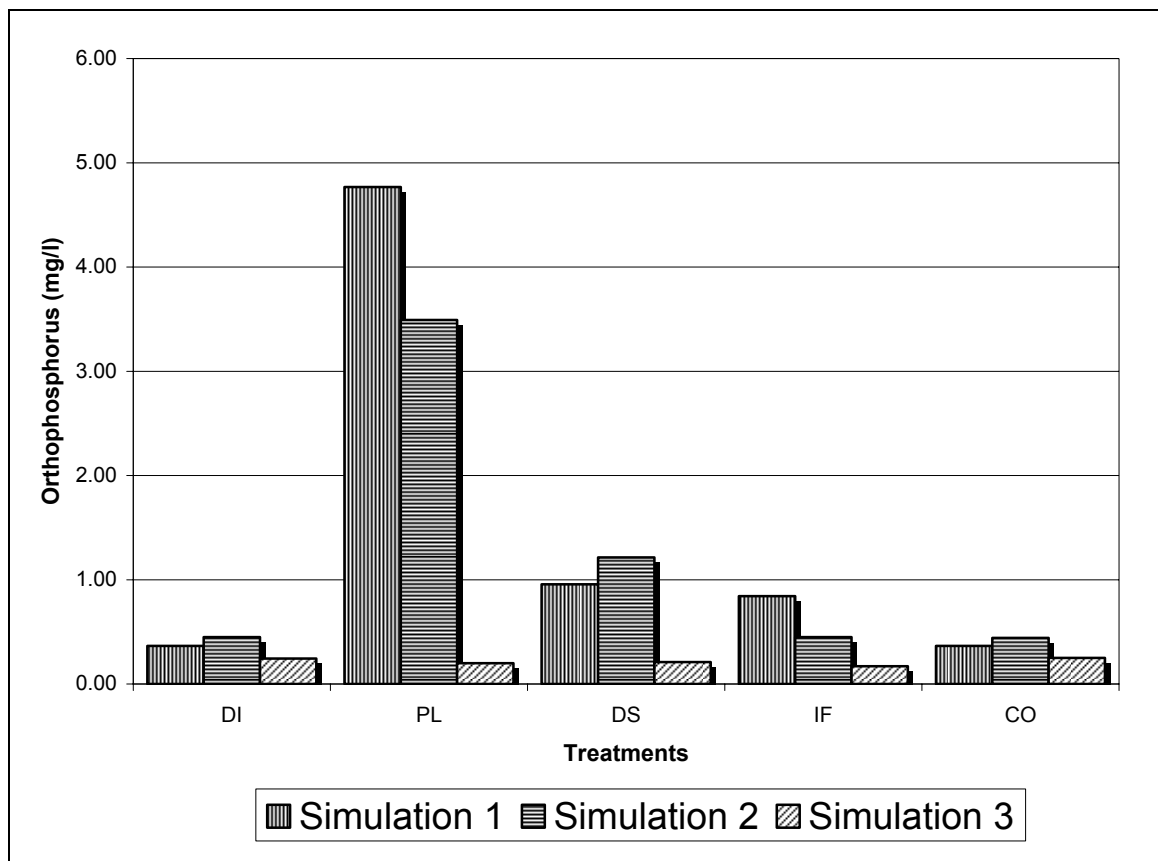


Figure 4.6 Mean DRP concentration in runoff from different treatments for each simulated rainfall event

The DRP concentrations of runoff reported in this study for the DI and CO treatments were very close to the concentration of DRP in raw water (0.24 mg/l and 0.12 mg/l for the first and second simulated events, respectively). The low concentration of DRP from the DI treatment compared to DS treatment (0.95 and 1.21 mg/l for first and second

simulated events, respectively) suggests that a significant amount of ortho-P present in dairy manure attached to soil particles shortly after it was incorporated. The liquid nature of dairy manure might have been helpful in this interaction. The dairy manure was surface applied in DS treatment and hence not enough interaction between soil particles and manure was possible compared to the DI treatment in which the manure was incorporated. The DRP concentration in runoff from DI treatments was about 2.5 times less than that from IF treatment, which suggests higher solubility of P in the inorganic fertilizer, as the dairy manure in DI treatment and the inorganic fertilizer in the IF treatment were incorporated. Dissolved Reactive P concentrations in the runoff from DI and IF plot were similar during the second simulated event, which suggests slow release of ortho-P from dairy manure. A 50% decrease in DRP concentrations in runoff from the IF treatment during the second simulated event not only suggests significant losses of nutrients during the first simulated rainfall event, but is also indicative of the highly soluble form of P present in the inorganic fertilizer (TSP).

The DRP concentrations in runoff from the PL treatment decreased by about 25% in the second simulated event compared to first simulated event. The primary reason for this reduction was the washing away of significant amount of organic material and nutrients by runoff during the first simulated rainfall event. A marginal increase in DRP concentrations in runoff from the first to second simulated event for DI and DS treatments, in spite of increase in runoff volume, suggests that the release of P from dairy manure is slow and can extend over a period of time. The concentration of DRP in runoff from the CO treatment was not significantly different from those measured for the pond water used for the experiments.

The concentration of DRP dropped significantly from the second to third simulated event for all the treatments. Mean comparison suggests that the DRP concentrations in runoff from the third simulated event were significantly lower than the DRP concentrations in runoff from the second simulated event. This decrease in concentration can be attributed to several factors including washing away of nutrients by the excessive amount of natural rainfall which occurred between the second and third simulated rainfall events, plant uptake of nutrients and the sorption of orthophosphorus to soil particles. A laboratory study by Sharpley (1997) also indicated the decrease in DRP concentrations in runoff from soils treated with poultry litter from first to 10th rainfall runoff event.

However, the concentrations reported by Sharpley (1997) in the first simulated event were 10-20 times lower than those measured in the present study. The primary reason may be that the first simulated event in Sharpley's (1997) study occurred 7 days after litter application which allowed enough time for interaction between soil and P, in contrast to the present study, where the first simulated event was conducted within 24 hours of poultry litter application. Significant interaction between treatments and simulated events was also observed suggesting that the rate of DRP release and consequently, the mechanism responsible for DRP release by dairy manure, poultry litter and inorganic fertilizer are different.

The descriptive statistics for the DRP yields in runoff from all the treatments for each simulation are shown in Table 4.9 and illustrated graphically in Figure 4.7. Dissolved Reactive Phosphorus yield followed almost the same pattern as the DRP concentrations, shown in Figure 4.6. For the first two simulated events, the DRP yield was highest from the PL treatment (376.2 and 418.3 g/ha) which were about 15 times greater than that from the DI treatment (23.8 g/ha) and about 4 times greater than the yield measured for the IF treatment (100.95 g/ha) for the first simulated event. Similar trends were also observed for the second simulated event.

Table 4.9 Descriptive statistics for the DRP yield in runoff from different treatments for each simulated event

Treatments	Simulation 1			Simulation 2			Simulation 3		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	31.34	61.27	16.26	65.87	107.00	35.31	26.93	58.87	8.43
PL	517.83	1081.76	150.95	501.15	854.77	305.45	8.48	17.10	0.00
DS	35.31	51.49	24.87	132.32	202.25	96.35	16.18	22.73	11.24
IF	116.56	178.47	55.22	76.31	94.11	61.92	15.43	22.70	9.89
CO	26.54	40.42	15.68	51.77	69.27	19.65	24.94	46.38	0.00

The DRP yield increased from the first to second simulated event for all treatments, except for the IF treatment, due to increased runoff volume. In the DI and DS treatments, the increase was magnified due to the increased runoff volume and DRP concentrations. However, the increase in DRP yield from the PL treatment was only due to increased runoff volume. The inorganic fertilizer is extremely soluble and hence in the IF treatment, the maximum DRP yield was observed in the first simulated event, as the amount of P available for dissolution with water reduced for the second simulated event. The reduction in DRP yield from second to third simulated event was significant and it was a

direct consequence of substantial decrease in DRP concentrations as shown in Figure 4.7.

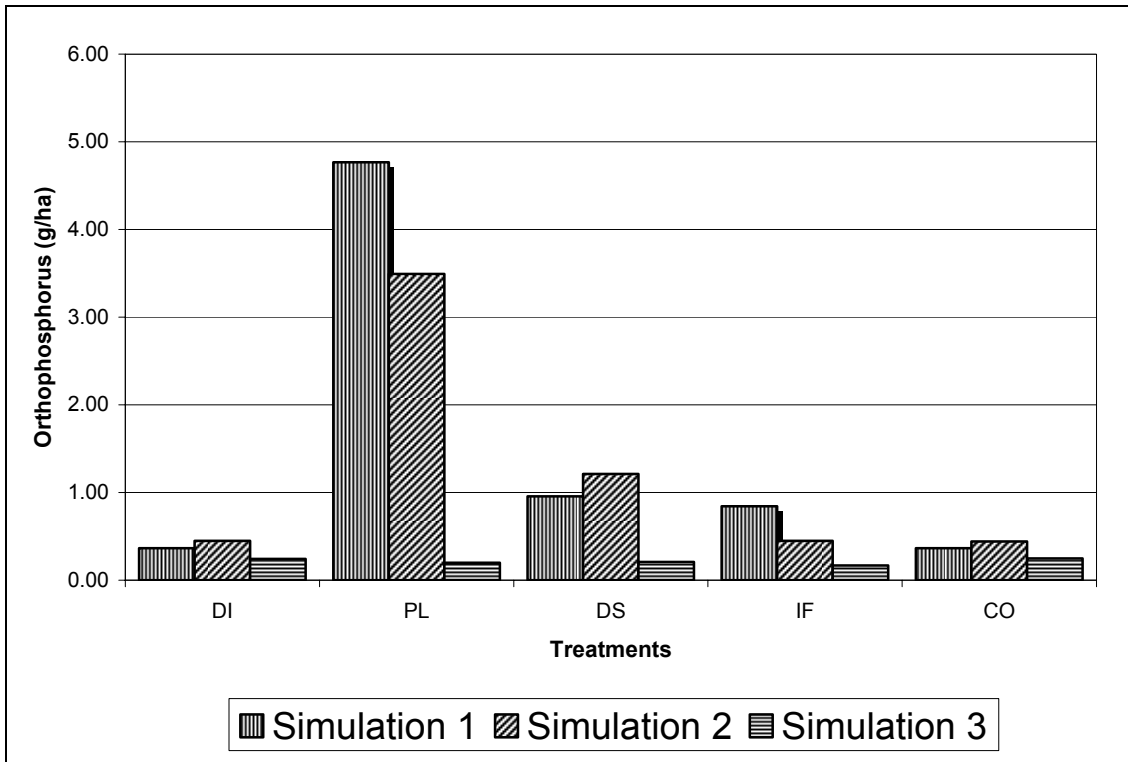


Figure 4.7 Dissolved Reactive Phosphorus yield in runoff from different treatments for all the simulated events

4.2.3 Bioavailable Phosphorus (BAP)

Bioavailable P consists of all the orthophosphorus which is dissolved and sediment bound. Bioavailable P is readily available for algal uptake in water bodies. Descriptive statistics for the BAP concentrations in runoff from different treatments for each simulated event are presented in Table 4.10 and mean BAP concentrations are graphically illustrated in Figure 4.8. A significant treatment effect was observed by the analysis of variance, and the mean comparison indicated that the concentrations of BAP in runoff from PL treatment were significantly higher than those measured for all other treatments, for the first and second simulated events; a trend similar to DRP concentrations in runoff. Clearly, the bioavailability of P in poultry litter is greater than the dairy manure.

Table 4.10 Descriptive statistics for the BAP concentrations in runoff from different treatments for each simulated event

Treatments	Simulation 1 ^a			Simulation 2 ^b			Simulation 3 ^c		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	2.74 ^A	4.02	1.46	0.85 ^A	0.99	0.65	0.35 ^A	0.48	0.27
PL	4.80 ^B	5.17	4.26	3.02 ^B	3.88	1.97	0.54 ^A	0.60	0.47
DS	2.24 ^A	2.39	2.10	1.39 ^A	1.85	1.06	0.71 ^A	0.99	0.43
IF	2.20 ^A	2.59	1.73	0.91 ^A	1.33	0.40	0.41 ^A	0.48	0.37
CO	1.92 ^A	2.46	1.59	0.65 ^A	0.74	0.52	0.46 ^A	0.62	0.36

*The values with different uppercase letters within a column are significantly different from each other. The simulated events with different lowercase letters are significantly different from each other.

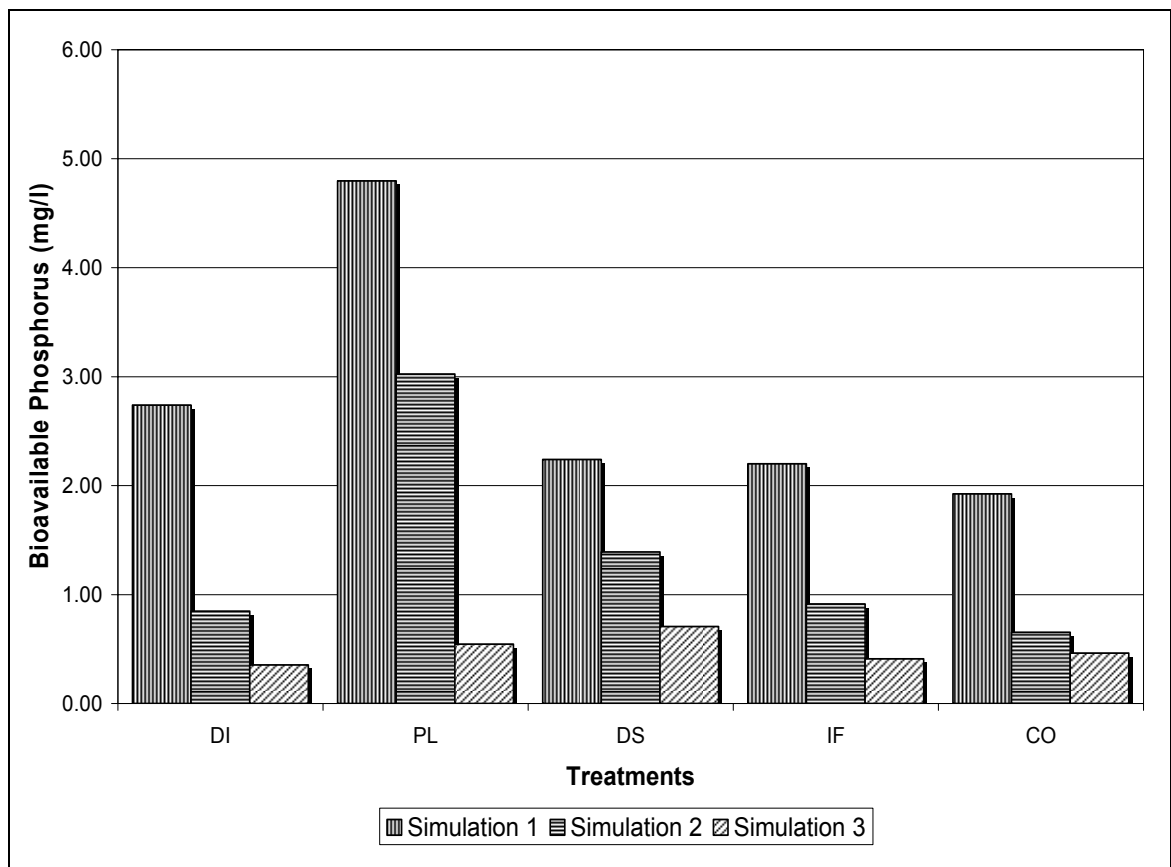


Figure 4.8 Concentration of Bioavailable Phosphorus in runoff from different treatments for the simulated events

The BAP concentrations in runoff from PL treatment (4.80 mg/l) were 1.5 to 2.5 times higher than those measured for other treatments for the first simulated event. As reported in section 4.2.2, DRP concentrations in runoff from PL treatment were 5-10 times greater than those measured for other treatments in the first simulated event. This reduced difference in BAP concentrations, compared to DRP concentrations, could

indicate that the P in dairy manure and inorganic fertilizer attaches itself to soil particles quicker than the P available in poultry litter. This phenomenon is further explained by the BAP concentrations resulting from the DS treatments (2.24 mg/l). Results indicate lower BAP concentrations in runoff from the DS, than the DI (2.74 mg/l) for the first simulated event, although DS had high DRP concentration in runoff than DI. These results show that the greater portion of ortho-P is attached to soil particles when dairy manure is incorporated. As shown in Figure 4.4, the concentration of TSS in runoff from DI treatment was marginally less than that for the DS treatment and significantly less than that for PL treatment; a further evidence that the binding of ortho-P to soil particles is quicker in dairy manure than in poultry litter treatments. Based on the results that the poultry litter has more water soluble form of P, reducing runoff volume should be the major emphasis in preventing the loss of BAP to downstream water bodies.

A significant decrease in the BAP concentration was observed from the first to the second simulated event for all treatments (Figure 4.8). The concentration of BAP was reduced by about 35 to 70% from the first to second simulated event for various treatments. These concentrations were further reduced by about 30 to 80% during the third simulated event, compared to the second simulated event. The decrease in concentration can be attributed to a decrease in DRP concentrations, decrease in TSS concentrations and increase in runoff volume from the first to second simulated events. The reduction between the second and the third simulated rainfall events reflects the effects of washing away of nutrients in the runoff resulting from heavy rainfall events, the uptake of nutrients by plants and the sorption of ortho-P to soil particles. The interaction between treatments and simulated events was also observed on the BAP concentrations in runoff suggesting that the relative behavior of treatments with each other changed with the simulated events. The primary reason behind the interaction between treatments and simulated event is that mechanism controlling the P release differed for different manure and inorganic fertilizer.

Table 4.11 Descriptive statistics for the BAP yield (g/ha) in runoff from different simulated events for each simulated event

Treatments	Simulation 1			Simulation 2			Simulation 3		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	229.54	424.66	92.59	121.24	162.97	85.01	39.48	70.65	11.36
PL	434.88	606.35	243.70	433.29	571.81	264.13	34.45	51.31	11.65
DS	94.52	141.51	70.90	144.10	161.00	111.01	50.32	62.75	39.35
IF	315.31	417.25	240.09	156.87	231.78	63.51	38.61	54.47	24.79
CO	144.71	192.83	79.62	73.57	98.95	55.68	42.10	49.51	29.27

Descriptive statistics for the BAP yield in runoff from different treatments for all the simulated events are shown in Table 4.11 and mean BAP concentrations are graphically illustrated in Figure 4.9. The maximum amount of BAP yield was observed from PL treatment among all the simulated events; a direct result of high BAP concentration in runoff. The increase in BAP yield from first to second simulated event for PL and DS treatments was primarily due to an increase in runoff volume. The BAP yield from all treatments decreased by about 60 to 80% from the second to the third simulated event, because of significant loss of manure during the heavy rainfall events between the second and the third simulated events, plant uptake of nutrients and sorption of P to the soil particles.

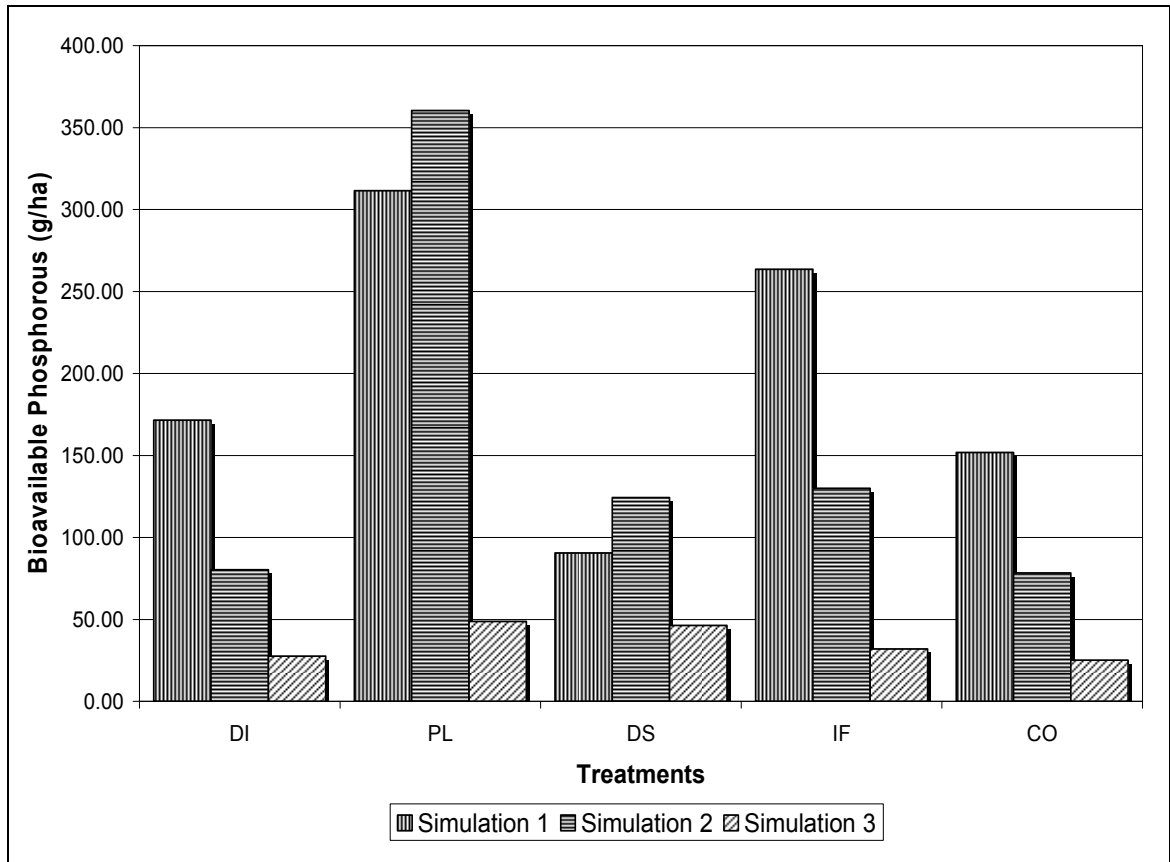


Figure 4.9 Amount of Bioavailable Phosphorus in runoff per unit area from different treatments for all the simulated events

4.2.4 Total Phosphorus (TP)

Total Phosphorus (TP) in runoff includes all forms of P in soluble and sediment bound phases. All sediment bound P is not available to aquatic plants for biological development, but it can be released into ambient water slowly in the dissolved form and become bioavailable in due time. Descriptive statistics for the TP concentrations in runoff from different treatments for each simulated event are presented in Table 4.8 and mean TP concentrations are graphically illustrated in Figure 4.10. Analysis of variance did not suggest significant treatment effects on the TP concentration in runoff for the simulated events.

Table 4.12 Descriptive statistics for the TP concentrations (mg/l) in runoff from different treatments for all the simulated events

Treatments	Simulation 1 ^a			Simulation 2 ^b			Simulation 3 ^b		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	8.10 ^A	11.00	5.60	2.00 ^A	2.80	1.10	1.95 ^A	3.05	1.38
PL	9.03 ^A	13.70	3.90	3.23 ^A	5.70	0.10	2.10 ^A	2.93	1.44
DS	5.90 ^A	8.60	3.20	3.73 ^A	5.40	1.80	1.55 ^A	2.29	0.85
IF	3.38 ^A	4.00	2.45	3.03 ^A	5.40	0.60	1.24 ^A	1.54	0.93
CO	4.93 ^A	9.80	2.20	0.60 ^A	1.70	0.00	1.57 ^A	2.44	0.85

*The values with different uppercase letters within a column are significantly different from each other. The simulated events with different lowercase letters are significantly different from each other.

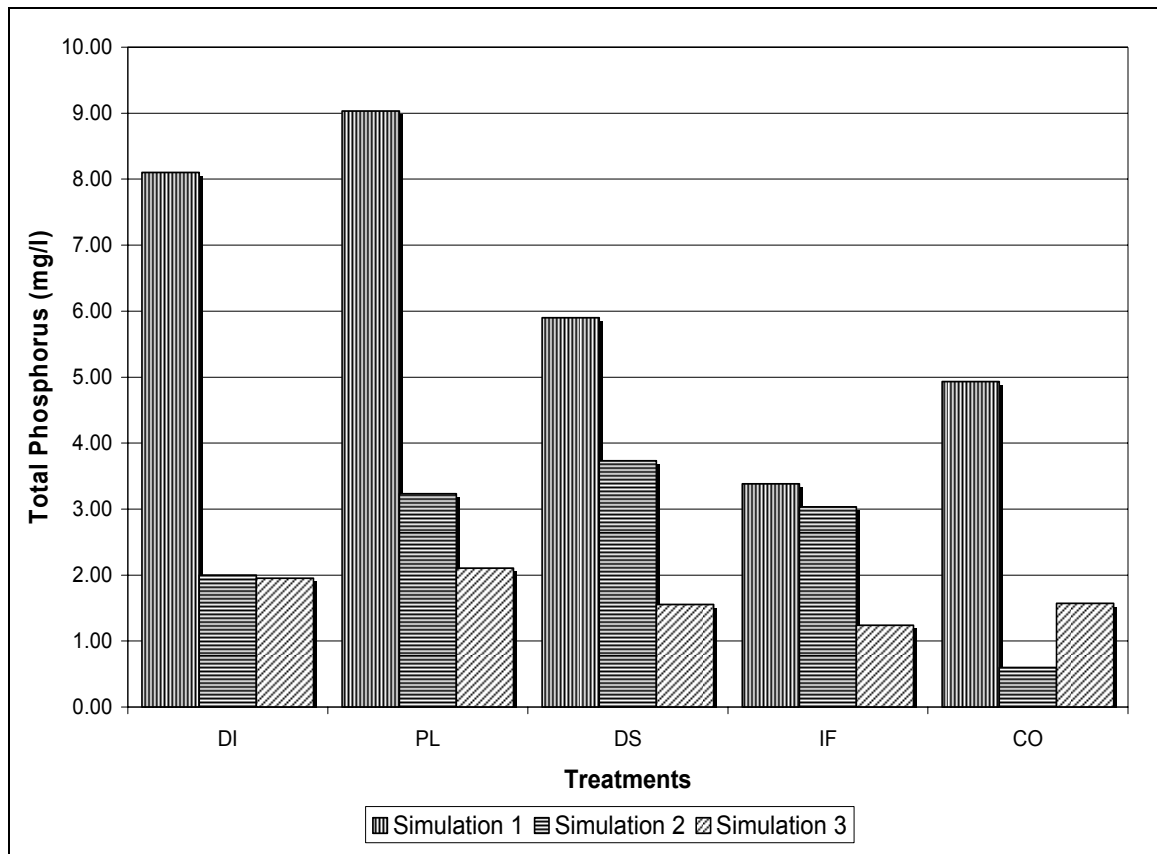


Figure 4.10 Mean concentration of TP in runoff from different treatments for all the simulated events

As reported in section 4.2.2, the concentrations of DRP in runoff from the DI treatment were lowest compared to other treatments, and the TP content of runoff from the DI treatment (8.1 mg/l) was the second highest during first simulated event. Thus, it can be deduced that a significant amount of P in the DI treatment is adsorbed on soil particles.

These conclusions support our inference derived in section 4.2.3 regarding sorption of P from dairy manure to soil in the DI treated plots. Among all the treatments investigated, the maximum and minimum concentrations of TP in runoff were observed in the PL (9.03 mg/l) and IF treatments (3.38 mg/l), respectively for the first simulated event. The high TP concentration in the PL treatment suggests higher P availability in poultry litter, moreover, a higher amount of P was also applied in the PL treatment compared to other treatments, as discussed in section 4.2.1..The concentrations of TP from IF treatment were less than those from all other treatments for the first simulated event The concentrations of TP from IF treatment were even lower than those measures from the CO treatment, which is due to a large value (9.8 mg/l) of TP concentration measured from one control plot.

Analysis of variance suggested a significant simulation effect on the TP concentrations in runoff. Mean comparison suggested that TP concentrations from the first simulated event were significantly higher than those from the second and third simulated events. However, the second and third simulated events were not significantly different from each other. This is particularly interesting because in the one month period between the second and third simulated events, significant amount of natural rainfall occurred and plant uptake of P also took place. These observations suggest that the majority of P was lost during the first simulated event, although contradictory results were obtained for DRP analysis. The TP concentration in runoff from DI and PL reduced significantly from first to second simulation events by 75 and 64%, respectively. The reduction in concentration was due to the significant loss of nutrients in runoff during the first simulated event and dilution by higher runoff volume during the second simulated event. The concentration of TP was reduced by 60% in the IF treatment to 2% in the DI treatment from second to third simulated event. A laboratory study conducted by Sharpley (1997) on the effect of poultry litter application on various types of soil, also reported reductions in TP concentration from the first simulated rainfall event to the 10th simulated event. However, the reduced TP concentration values, even after the 10th event, were greater than TP concentrations from control treatment for all soils. The 7 day incubation period between litter application and first simulated event might have provided enough time for soil and P interaction and thus more P sorption was possible in the study by Sharpley (1997). The mean TP concentrations observed in the study by Sharpley (1997) were 4 to 5 times less than the TP concentration observed from poultry

litter treated plots in the present study. No significant interactions between the simulated rainfall events and treatments were observed, which suggested similar behavior of treatments with respect to each other regarding TP concentrations in runoff for all the simulated events.

The TP concentrations of runoff reported in the present study are 100 to 1000 times higher than the water quality criteria recommended by EPA for this ecoregion (XI). The water quality criteria for lakes and reservoir is 8 µg/l and water quality criteria for rivers and streams is 10 µg/l to prevent nutrient overenrichment of waterbodies. Bases on these criteria, the raw pond water used for the study was itself overenriched. However, these criteria are just guidelines and the actual standards may be different. Nevertheless, the TP concentrations reported in the study can overenrich the downstream waterbodies with nutrients and cause eutrophication. The results indicate that cropland applied with poultry litter has higher potential than dairy manure treated cropland to release P in runoff.

Table 4.13 Descriptive statistics for the TP yield (g/ha) in runoff from different treatments for each simulated event

Treatments	Simulation 1			Simulation 2			Simulation 3		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	590.25	697.58	481.60	291.62	460.91	143.87	224.22	411.61	50.57
PL	690.44	870.66	555.11	469.81	821.26	13.41	149.19	251.68	29.96
DS	270.27	509.18	101.75	372.61	469.95	228.97	109.07	145.16	85.44
IF	480.89	596.08	340.28	524.80	941.07	95.27	118.26	152.27	60.66
CO	305.68	490.75	206.81	83.08	240.32	0.00	142.86	194.83	69.11

The descriptive statistics for TP yield in runoff are shown in Table 4.13 and mean TP yields are graphically illustrated in Figure 4.11. The loss of TP was highest in the PL treatment during the first simulated event and these results were similar to the trend observed for DRP and BAP amounts in runoff. The TP yield in the DI treatment was higher than that recorded from the DS treatment. The higher amount of TP in DI treatment is a direct consequence of higher concentration of TP and higher volume of runoff in DI compared with the DS treatment for first simulated event. The TP yield reduced from first simulated event to second simulated event for DI, PL and CO treatments, but, the TP yield increased for IF and DS treatments. The primary reason could be the fact that the decrease in concentration of TP in runoff from the DS

treatment was less than the corresponding increase in runoff volume, while in the IF treatment, both concentration and runoff increased from the first to second simulated event. A decrease in the TP yield was observed for all treatments from the second to the third simulated event, primarily because of heavy rainfall and runoff events occurring prior to the third simulated event, plant uptake and the various processes taking place in soil to stabilize soil P.

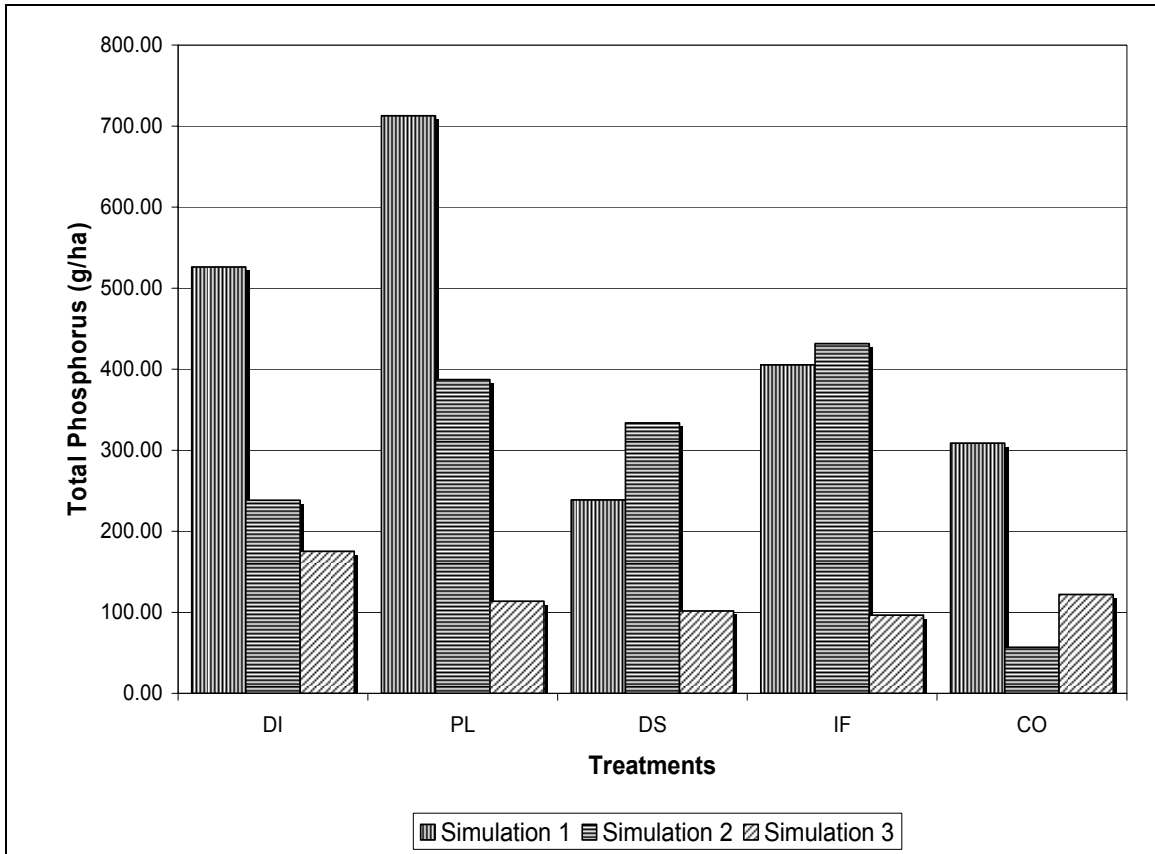


Figure 4.11 Mean amount of Total Phosphorus in runoff per unit area from different treatments for all the simulated events

4.2.5 Nitrate-Nitrogen (NO_3^- -N)

The descriptive statistics for the concentrations of NO_3^- -N in runoff from different treatments for each simulated event are shown in Table 4.10 and mean NO_3^- -N concentrations in runoff are graphically illustrated in Figure 4.12. Analysis of variance suggested significant treatment effects for the first and second simulated events. The maximum NO_3^- -N concentration in runoff was observed for the first two simulated events from the DS treatment (1.7 and 1.96 mg/l, respectively). Reduced volume of surface

runoff from the DS treatment, could partially explain the higher concentrations of NO_3^- -N in runoff. Mean comparison of treatments suggested that the concentration of NO_3^- -N in runoff from DS treatment was significantly higher than those measured for DI (1.33 mg/l) and IF treatments (1.17 mg/l) for the first simulated event and significantly higher from all other treatments for the second simulated event. The lowest concentration of NO_3^- -N in runoff from CO treatment (1.06 mg/l) indicates that the application of manure or fertilizer to cropland contributes to enriched NO_3^- -N levels in runoff. However, the maximum concentration of NO_3^- -N in runoff measured from any treatment was less than 2.00 mg/l which is much less than the 1000 mg/l level, a safe limit for freshwater fish (AWMFH, 2000). Although NO_3^- -N is an important nutrient for aquatic plant development which can lead to eutrophication, the USEPA has not set any specific limit for NO_3^- -N concentration in surface waters.

Table 4.14 Descriptive statistics for the NO_3^- -N concentration (mg/l) in runoff from different treatments for each simulated event

Treatments	Simulation 1 ^a			Simulation 2 ^a			Simulation 3 ^b		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	1.33 ^A	1.60	1.10	1.40 ^A	1.50	1.30	0.80 ^A	1.00	0.50
PL	1.57 ^A	1.70	1.40	1.23 ^A	1.40	1.00	0.93 ^A	1.10	0.70
DS	1.70 ^{AB}	2.30	1.30	1.97 ^B	2.00	1.90	0.93 ^A	1.10	0.80
IF	1.17 ^A	1.31	1.10	1.50 ^A	1.60	1.40	1.37 ^A	1.80	0.90
CO	1.07 ^{AC}	1.10	1.00	1.23 ^A	1.30	1.20	0.83 ^A	1.10	0.50

*The values with different uppercase letters within a column are significantly different from each other. The simulated events with different lowercase letters are significantly different from each other.

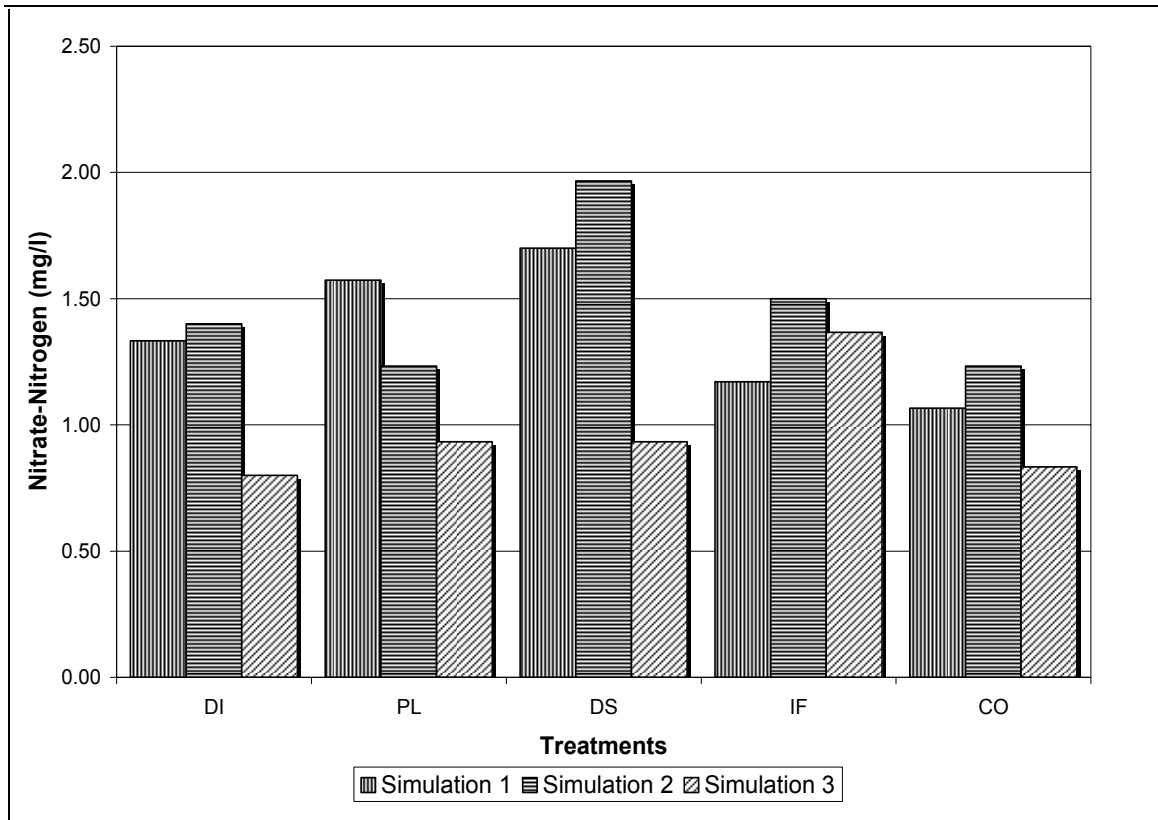


Figure 4.12 Mean NO_3^- -N concentration of runoff from the treatments for different simulated events

Although the volume of runoff increased from the first simulated event to the second simulated event, the concentration of NO_3^- -N was increased for all treatments in the second simulated event, except for the PL treatment. It appears that when the dairy manure and fertilizer are applied to well aerated soils, NH_4^+ -N is rapidly converted to NO_3^- -N and hence higher concentrations of NO_3^- -N was observed during the second simulated event. However, the same phenomenon was not observed for the PL treatment. It can be concluded that NO_3^- -N in poultry litter does not hydrolyze as quickly as it does in dairy manure and inorganic fertilizer. The higher concentration of TSS in runoff compared to the DS and DI treatments (Figure 4.4) also supports this notion. However, previous studies by Sharpley (1985) and Sharpley et al. (1997) have reported a lack of any significant relationship between NO_3^- -N in runoff and the soil. No significant treatment effect was observed for the NO_3^- -N concentrations in runoff during the third simulated event. The concentrations of NO_3^- -N in the third simulated event were less than those observed for first and second simulated events. Plant uptake might have reduced NO_3^- -N in the soil since the NO_3^- -N concentration in runoff was almost equal to

the concentration of NO_3^- -N in the pond water used for the three simulated rainfall events.

The overall comparison between the simulated events suggested that the third simulated event was significantly different from the first two simulated events with regard to NO_3^- -N concentrations in runoff. A significant interaction between treatments and simulated rainfall events was also observed. The interactions can be the result of PL and IF treatments behaving differently in different simulated events. PL treatment resulted in lower concentrations of NO_3^- -N in runoff from the second runoff, in contrast to all other treatments, while the IF treatment showed higher concentrations of NO_3^- -N in runoff from the third simulated event compared to the first simulated event, in contrast to all other treatments.

The NO_3^- -N yields in runoff are shown in Table 4.15 and mean NO_3^- -N yields are graphically illustrated in Figure 4.13. The maximum NO_3^- -N yield occurred in runoff from the IF treatment for all three simulated events. The lowest NO_3^- -N yield occurred for the CO treatment during the first two simulated events, except DS in first simulated event. Although the highest concentration of NO_3^- -N in runoff was observed from the DS treatment for the first simulated event (Figure 4.12), the minimum amount of NO_3^- -N yield observed in the DS treatment were probably due to very low runoff volume from the DS treatment. The increase in NO_3^- -N yield from first to second simulated event was due to the increase in NO_3^- -N concentration as well as to increased runoff volume.

Table 4.15 Descriptive statistics for the NO_3^- -N yield in runoff from different treatments for each simulated event

Treatments	Simulation 1			Simulation 2			Simulation 3		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	102.53	137.33	68.80	199.00	246.92	170.03	95.66	147.19	18.32
PL	143.83	199.27	76.42	175.90	206.32	134.07	64.85	94.08	14.56
DS	76.11	136.18	41.33	209.74	254.41	165.35	72.33	102.29	44.97
IF	166.18	182.08	139.23	252.82	261.41	242.99	128.19	204.27	88.99
CO	79.90	103.40	50.08	138.66	169.64	107.16	72.99	87.83	57.97

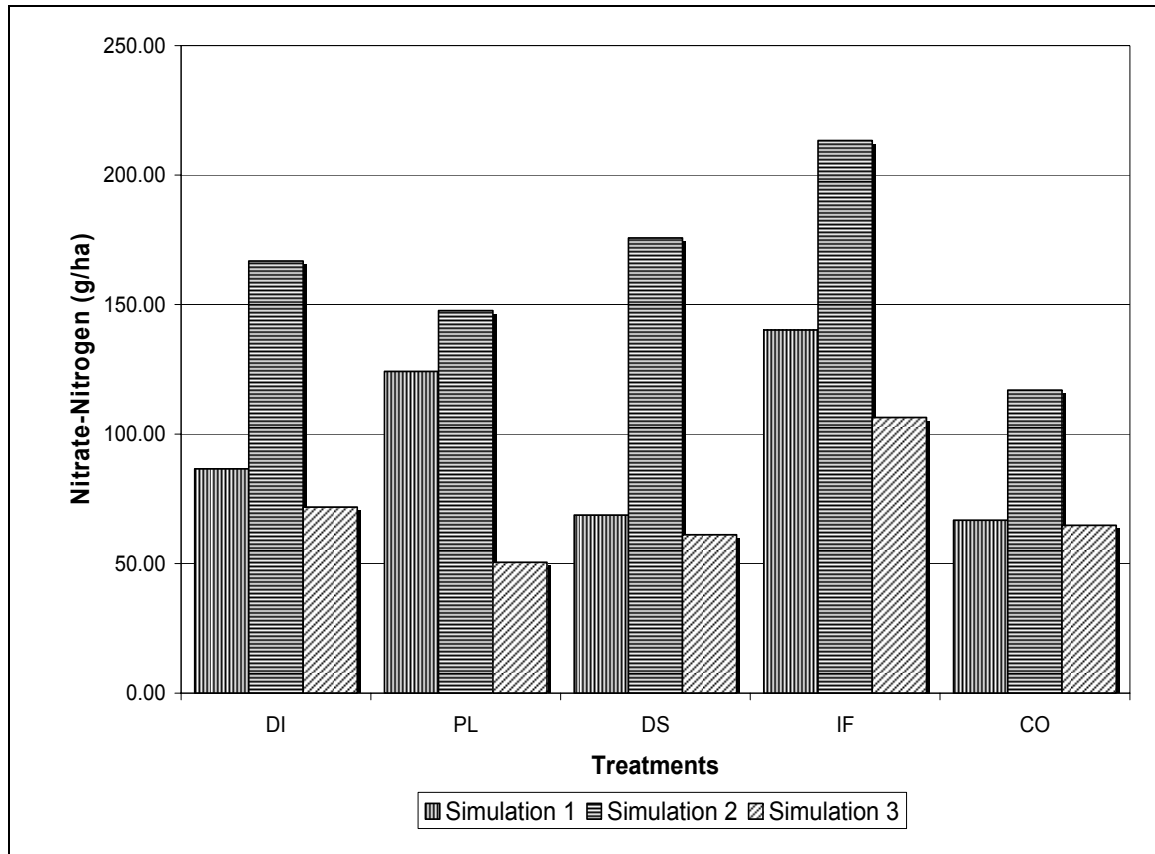


Figure 4.13 Nitrate-Nitrogen yield in surface runoff, per unit area for different treatments for all the simulated events

4.2.6 Ammonia Nitrogen ($\text{NH}_4^+\text{-N}$)

Ammonia-Nitrogen concentrations of runoff from different treatments for each simulated rainfall event are shown in Table 4.16 and mean $\text{NH}_4^+\text{-N}$ concentrations of runoff are graphically illustrated in Figure 4.14i. No significant differences in $\text{NH}_4^+\text{-N}$ concentrations were observed among the treatments for the simulated events. The maximum $\text{NH}_4^+\text{-N}$ concentration was observed from the PL treatment (1.42 mg/l and 1.6 mg/l, respectively) during the first two simulated events. The corresponding lowest $\text{NH}_4^+\text{-N}$ concentrations were observed for the CO treatment (0.56 and 0.09 mg/l). Safe concentrations of $\text{NH}_4^+\text{-N}$ varies with pH and temperature for different uses in Virginia (SWCB, Va, 2003) and these runoff concentrations would violate the standard in situations with high pH (> 8) and low temperature (< 15°C).

Table 4.16 Descriptive statistics for the NH_4^+ -N concentrations (mg/l) in runoff from different treatments for each simulated rainfall event

Treatments	Simulation 1 ^a			Simulation 2 ^a			Simulation 3 ^b		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	1.23 ^A	2.50	0.40	0.73 ^A	1.40	0.20	0.05 ^A	0.10	0.02
PL	1.42 ^A	1.50	1.27	1.60 ^A	2.90	0.60	0.15 ^A	0.30	0.05
DS	1.17 ^A	1.40	0.70	0.97 ^A	1.80	0.50	0.07 ^A	0.20	0.00
IF	0.68 ^A	1.10	0.19	0.41 ^A	0.70	0.20	0.23 ^A	0.40	0.10
CO	0.57 ^A	1.00	0.20	0.09 ^A	0.20	0.00	0.17 ^A	0.40	0.00

*The values with different uppercase letters within a column are significantly different from each other. The simulated events with different lowercase letters are significantly different from each other.

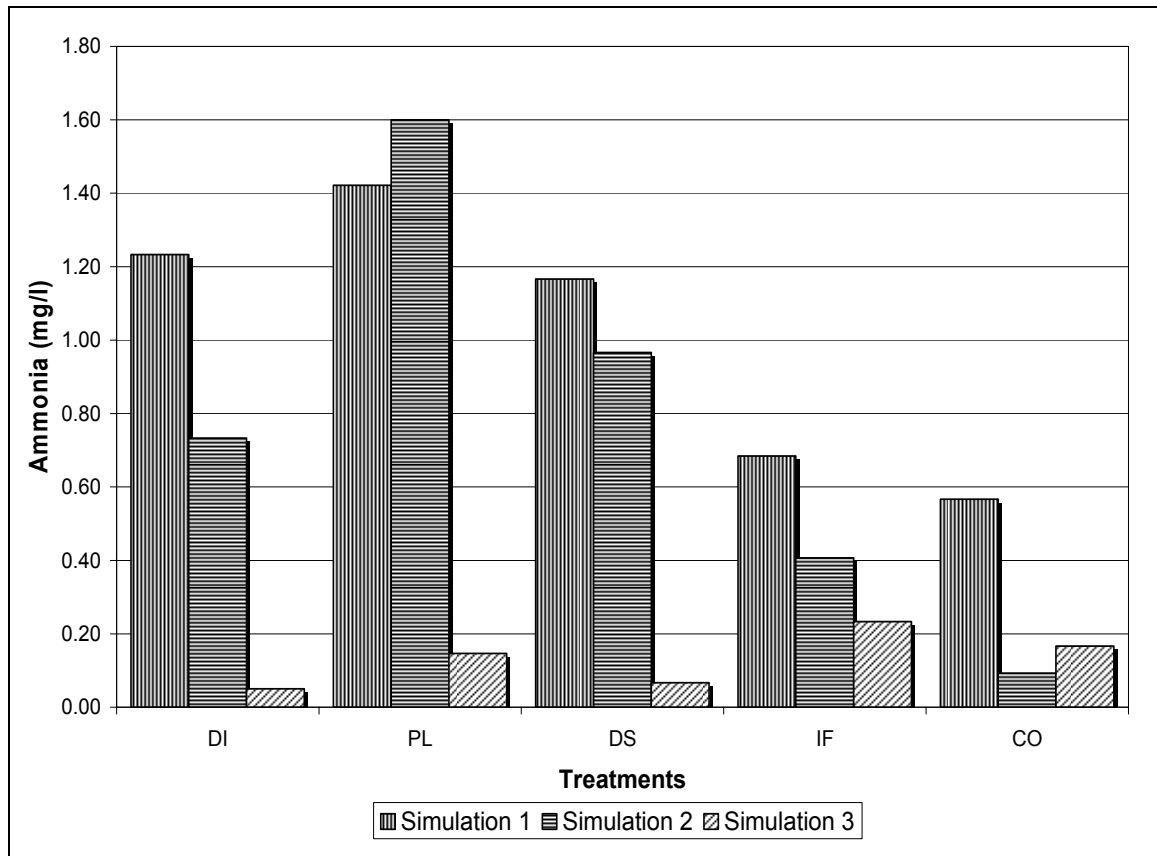


Figure 4.14 Mean NH_4^+ -N concentrations in runoff from the different treatments for all simulated events

In contrast to NO_3^- -N, maximum NH_4^+ -N concentrations observed in surface runoff were from the PL treatment for the first two simulated events. The NH_4^+ -N concentrations in runoff increased from the first to the second simulated event for the PL treatment, in

contrast to other treatments. Results would indicate that the conversion of $\text{NH}_4^+\text{-N}$ to $\text{NO}_3^-\text{-N}$, when poultry litter is exposed to aerobic condition is not as spontaneous when compared with dairy manure. The liquid nature of dairy manure helped in better spreading of manure over the surface in DS treatment and hence dairy manure was better exposed to aerobic conditions than poultry litter in PL treatment. A field study by Vories et al. (2001), when comparing the inorganic fertilizer and poultry litter, reported slightly higher concentrations of $\text{NH}_4^+\text{-N}$ in runoff from plots treated with poultry litter, but their study consisted of natural rainfall events spread over a longer time period. Edwards and Daniel (1994) conducted a field study on fescue grass and reported highest $\text{NH}_4^+\text{-N}$ concentrations from plots treated with inorganic fertilizer rather than with poultry litter, as indicated in this study. However, with the subsequent rainfall events, no difference between the treatments was observed in their study.

The $\text{NH}_4^+\text{-N}$ concentrations in runoff decreased for all treatments, except for the PL treatment, from the first to second simulated rainfall event, but not significantly. The decrease in $\text{NH}_4^+\text{-N}$ concentration can be attributed to the loss of $\text{NH}_4^+\text{-N}$ in runoff during the first event or the conversion of $\text{NH}_4^+\text{-N}$ to $\text{NO}_3^-\text{-N}$ prior to subsequent simulations. The concentrations were reduced significantly from second to third simulated event (by about 90%) for all treatments, except for CO. The same pattern of $\text{NH}_4^+\text{-N}$ loss was observed by Sharpley (1997) in a laboratory study, where the $\text{NH}_4^+\text{-N}$ concentration reached the background level by the 10th simulated rainfall event. The increase in $\text{NH}_4^+\text{-N}$ concentration in the CO treatment during the third simulated event can be considered a minor fluctuation in natural levels of $\text{NH}_4^+\text{-N}$ in soil.

The descriptive statistics for the $\text{NH}_4^+\text{-N}$ yields in runoff from different treatments for each simulated event are shown in Table 4.17 and mean $\text{NH}_4^+\text{-N}$ yield are graphically illustrated in Figure 4.15. The maximum amount of $\text{NH}_4^+\text{-N}$ in runoff was measured in the PL treatment compared to other treatments for the first two simulated events, followed by those for the DI treatment. The high amount of $\text{NH}_4^+\text{-N}$ lost from the PL treatment can be attributed to high $\text{NH}_4^+\text{-N}$ concentrations in runoff and high runoff volume. Minimum amounts of $\text{NH}_4^+\text{-N}$ loss was measured from the CO treatment.

Table 4.17 Descriptive statistics for the $\text{NH}_4^+\text{-N}$ yield (g/ha) in runoff from different treatments for each simulated event

Treatments	Simulation 1			Simulation 2			Simulation 3		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	89.36	158.54	25.02	97.15	180.04	32.92	6.17	14.72	1.10
PL	136.89	213.50	59.68	221.51	388.81	88.42	6.08	7.73	4.28
DS	50.86	82.89	22.26	94.36	156.65	62.84	4.23	12.68	0.00
IF	102.37	177.21	24.05	69.67	121.50	31.76	19.07	26.09	11.35
CO	40.74	78.39	18.80	9.52	21.42	0.00	18.17	46.38	0.00

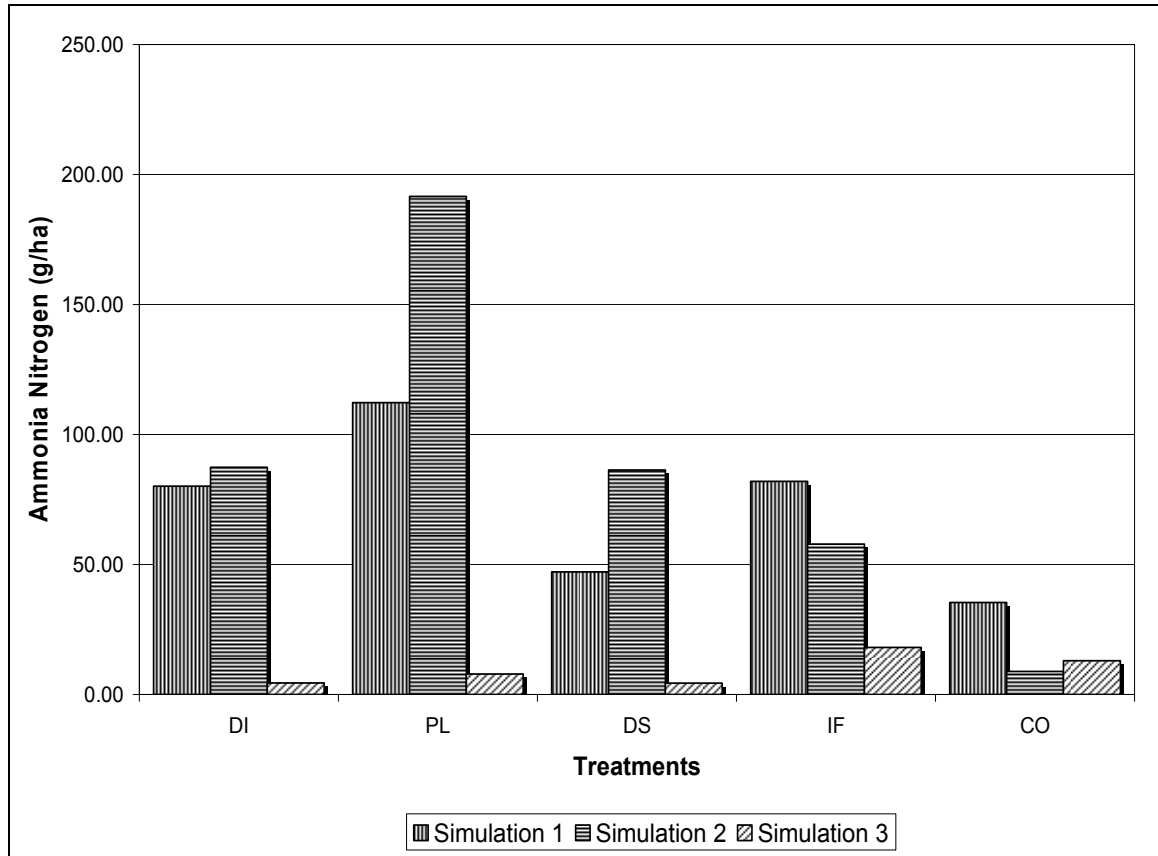


Figure 4.15 Mean $\text{NH}_4^+\text{-N}$ yield in runoff per unit area from different treatments for all the simulated events

4.2.7 Total Nitrogen (TN)

Total Nitrogen (TN) concentrations in the runoff from different treatments for each simulated event are shown in Table 4.18 and mean TN concentrations are graphically illustrated in Figure 4.16. Significant treatment effect on the TN concentration in runoff was observed during the first simulated event. The TN concentration in runoff from the PL treatment (21.08 mg/l) was significantly higher than that from DS (13.47 mg/l) and CO (12.67 mg/l) treatment for the first simulated event. The maximum concentration of

TN was observed for the PL treatment in the first simulated event, followed by DI, IF and DS treatments, respectively. The TN concentration dropped slightly in the runoffs from second simulated event for all treatments except for the DS treatment for which it increased by about 40%. This increase can be attributed to increased NO₃⁻-N concentrations as discussed in section 4.2.2. In the third simulated event, however, the concentrations were reduced by about 85 to 90% for all treatments.

Table 4.18 Descriptive statistics for the TN concentrations (mg/l) in runoff from different treatments for all the simulated events*

Treatments	Simulation 1 ^a			Simulation 2 ^a			Simulation 3 ^b		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	19.27 ^A	21.50	16.30	19.17 ^A	21.20	17.80	2.73 ^A	3.10	2.10
PL	21.08 ^{AB}	24.90	17.25	18.30 ^A	22.70	13.80	2.10 ^A	2.80	1.60
DS	13.47 ^{AC}	16.40	10.50	19.03 ^A	24.10	15.30	1.37 ^A	1.90	1.10
IF	15.64 ^A	17.93	14.10	14.90 ^A	16.40	13.70	2.10 ^A	2.80	1.60
CO	12.67 ^{AC}	14.20	11.60	12.23 ^A	13.60	10.40	1.87 ^A	2.50	1.50

*The values with different uppercase letters within a column are significantly different from each other. The simulated events with different lowercase letters are significantly different from each other.

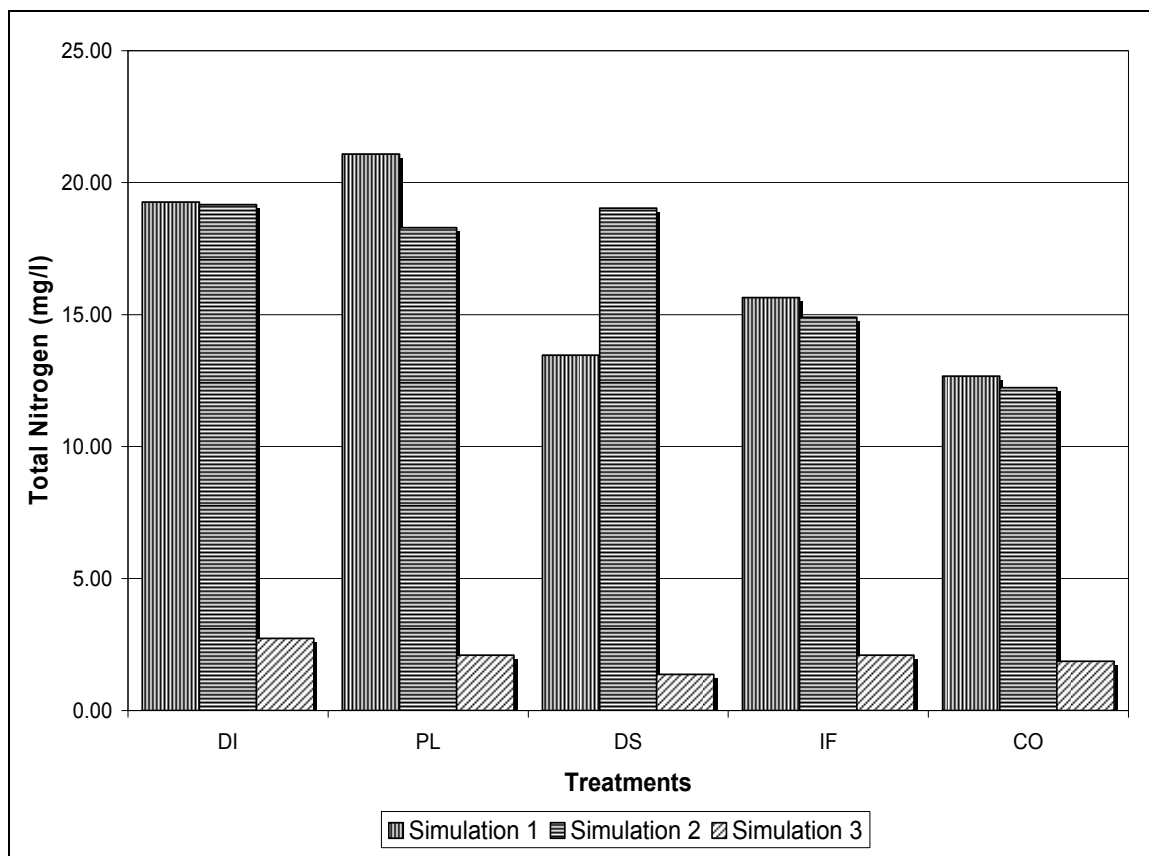


Figure 4.16 Mean TN concentrations (mg/l) of surface runoff from different treatments for each simulated event

The concentrations of TN in runoff from all treatments were significantly higher than the sum of $\text{NH}_4^+\text{-N}$ and $\text{NO}_3^-\text{-N}$ concentrations in the runoff from all the treatments. It can be inferred that a large amount of organic nitrogen was present in the soil. The experimental plots, when tilled, had a fair amount of vegetation on the surface. The uprooted vegetation were left to dry under the sun after the tillage operation. There were few rainfall events between the first tillage operation and the setup of experiments. These conditions might have stimulated the organic matter and mineralization of organic-N. The soil was not tested for N content as it is not covered in the Virginia Tech routine soil testing services. Determination of soil N may have provided additional insight into the reason for high TN levels in runoff.

The TN concentrations of runoff reported in this study are well above the nutrient criteria of 0.34 mg/l recommended by USEPA to cause overenrichment in the lakes, reservoirs,

rivers and streams for this ecoregion (ecoregion XI) (USEPA, 2002). However, these values are just recommendations and different concentrations may be set as standards by the respective states for different designated uses. However, compared to pristine water sources these concentrations have potential to cause eutrophic problem in waterbodies.

A significant effect of simulated rainfall events was observed on the TN concentrations in runoff from different treatments. Overall, comparisons suggested that the concentration of TN in runoff from third simulated event was significantly lower than that from the first and second simulated events. This reduction in TN concentration from the second and third simulated rainfall events can be attributed to several processes taking place in soil during the time between second and third simulated event. Apart from washing away of nutrients with the surface runoff following the heavy rainfall event, plant uptake of N, leaching and mineralization processes might have reduced the availability of TN in the soil surface. Significant interaction between the simulated rainfall events and treatments was also observed with regard to TN concentrations in runoff. The interaction was observed primarily due to contrasting response regarding TN concentration in runoff of DS treatment from first to second simulated event, compared to other treatments.

Total Nitrogen yield from the different treatments for each simulated rainfall event are shown in Table 4.19 and mean TN yield are graphically illustrated in Figure 4.17. The amount of TN lost in runoff was maximum for the IF treatment (1874 g/ha) during the first simulated event. The TN loss was minimum in runoff from DS treatment for first and third simulated events, compared to the other treatments, and TN loss in the DS treatment during the second simulated event was higher than that from the CO treatment only. In the third simulated event, TN loss in runoff decreased by about 90% for all treatments.

Table 4.19 Descriptive statistics for the TN yield (g/ha) in runoff from different treatments for each simulated event

Treatments	Simulation 1			Simulation 2			Simulation 3		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	1498.55	2112.72	1019.48	2694.00	2930.10	2379.15	279.52	441.56	113.61
PL	2097.02	3544.18	813.09	2610.85	3270.65	1850.22	145.32	239.47	33.29
DS	535.50	621.68	429.24	2035.67	2523.98	1331.53	102.43	125.02	61.83
IF	2215.27	2488.29	1885.98	2522.05	2846.48	2175.29	185.48	215.62	158.20
CO	919.28	1090.43	711.08	1353.49	1470.18	1134.06	179.90	289.85	119.77

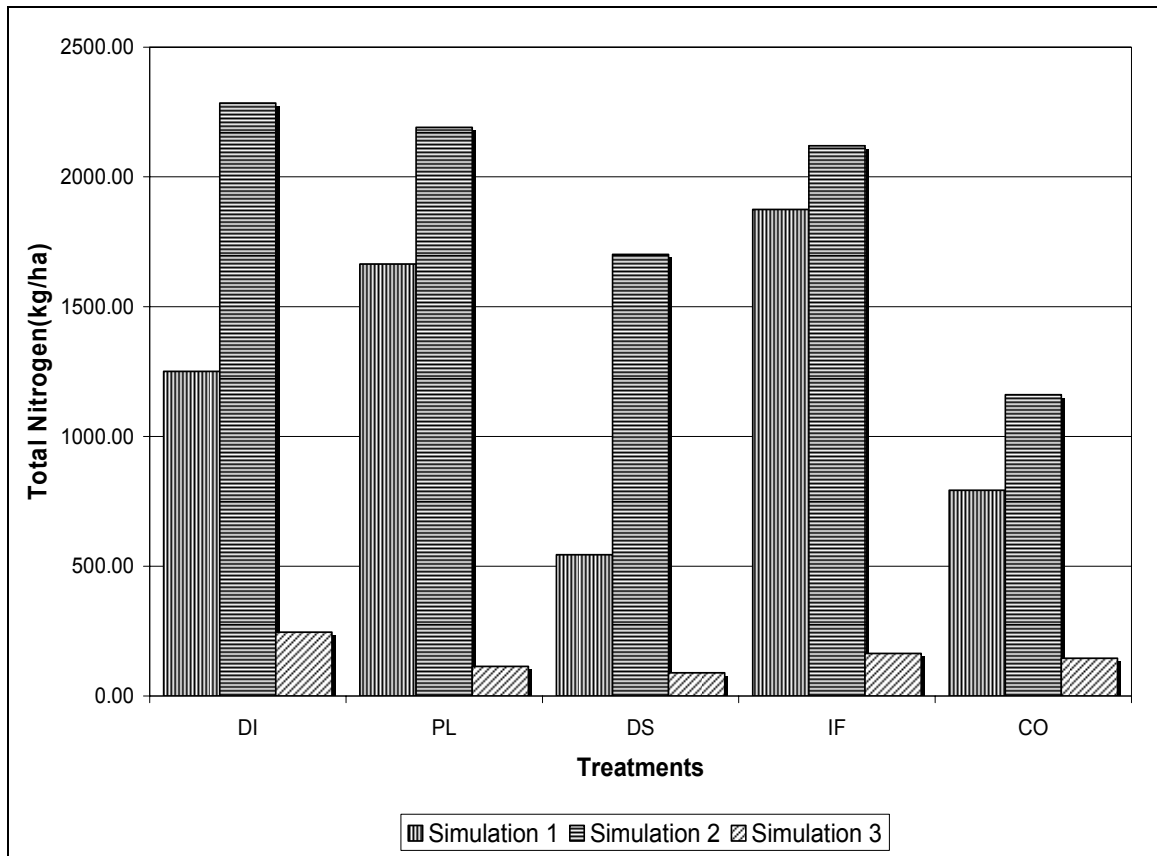


Figure 4.17 Total Nitrogen yield from different treatments for each the simulated rainfall event

4.2.8 Summary of nutrient transport study

The nutrient transport study illustrates the effect of P-based manure application on nutrient concentrations of runoff. The results show that the P loss is significantly high from the poultry litter compared to dairy manure, in spite of P-based management. The DRP and BAP concentrations from the PL treatment were 5-10 times higher than those measured for other treatments. The TP concentrations were 2-3 times higher in runoff from PL treatments than those reported for other treatments, except DS. These results also imply that the P from PL treatments is mostly in the soluble form (as opposed to sediment bound), and hence the P loss to downstream water bodies can be controlled by implementing BMPs that reduce runoff from agricultural fields treated with poultry litter.

The TP concentrations of runoff from all the treatments are well above the nutrient criteria of 8 µg/l for lakes and reservoirs and 10 µg/l for rivers and streams in this ecoregion (ecoregion XI) (USEPA, 2002). The P levels measured in runoff in this study were also higher than those reported by similar studies in the literature. The results show that manure application according to the P requirement of crops would not necessarily reduce eutrophication of downstream water bodies. However, P-based management helps reduce the P loadings compared to N based application (Sharpley, 1997).

The NO₃⁻-N concentrations of runoff from all treatments and simulated events were two orders of magnitude lower than the lethal concentrations specified for aquatic organisms (AWMFH, 2000). The NO₃⁻-N concentrations of runoff generally increased from the first to the second simulated event, which is in contrast with trends observed for all other nutrients, and reached a stable value equal to the nitrate levels measured in the raw water during the third simulated event. The increase in NO₃⁻-N concentration could be attributed to the rapid conversion of NH₄⁺-N to NO₃⁻-N available near soil surface. The NH₄⁺-N concentrations in runoff were well within the federal limits for primary contact recreation water. The highest NH₄⁺-N concentrations were observed in runoff from PL treatments, which is mainly due to the high NH₄⁺-N content of poultry litter compared to dairy manure as shown in the Table A1 and A2 in appendix A. There is no federal limit for TN concentrations in surface waters. However, the TN concentrations reported in this study were above the threshold of 0.46 mg/l and 0.36 mg/l which can cause nutrient overenrichment in lakes and reservoirs and rivers and streams, respectively (USEPA, 2002). These concentrations are edge of field concentrations and some dilution is expected as runoff reaches a water body. The nutrient yields decreased significantly in the third simulated event, compared to the first and the second simulated events. The reasons for nutrient yield reduction were uptake of nutrients by plants, washing away of nutrient in runoff by the large rainfall amounts occurring between the second and third simulated events, transformation and mineralization processes taking place between soil particles and nutrients, and leaching. However, the TN concentrations in runoff from the third simulated event were still above the threshold of 0.46 mg/l, which may cause nutrient overenrichment in water bodies (USEPA, 2003).

4.3 Bacteria

Runoff samples were analyzed for three indicator bacteria species; Fecal Coliform (FC), Escherichia Coli (EC) and Enterococcus (ENT). The runoff samples from the IF treatment were not analyzed for bacteria since no manure was applied on this treatment and it was not expected to contribute any bacteria to the runoff. The analysis was conducted within five hours of sample collection in the water quality laboratory of Department of Biological Systems Engineering, Virginia Tech. The original manure samples were also analyzed for their bacterial content. The following sections discuss the concentration of these bacterial species in runoff from each treatment. The effect of different treatments and the simulated event timings on bacterial concentrations of runoff are also discussed.

The statistical analysis of all bacterial concentrations was conducted on the log values of concentrations at base 10. The log values are also used to display the data graphically, so that the low values from CO treatment can be displayed as well. It is a common practice to perform this logarithmic transformation before conducting an analysis of variance as the desired characteristics of additivity, constant variance and normality are achieved for pollutant data. This transformation is also conducted before mean comparison tests to look for significant differences among the means (Gilbert, 1987).

4.3.1 Fecal Coliform (FC)

Fecal Coliform bacteria concentrations in runoff varied are shown in Table 4.20 and mean FC concentrations are graphically illustrated in Figure 4.18. The FC concentrations of runoff are presented on logarithmic scale on vertical scale to show the small values observed during the last simulated event. The maximum concentration of FC in the first simulated event was measured in the runoff from PL treatment (1.043×10^6 cfu/100ml), while the minimum concentrations were recorded from the CO treatment (2.64×10^4 cfu/100ml) for the first simulated event. However, the concentrations of FC remained in the same order of magnitude for DI, DS and PL treatments for first two simulated events. No significant treatment effects were observed for the any simulated rainfall event. The high concentration of FC in the CO treatment, even though 6 to 40 times less than the concentrations measured for other treatments, was surprising, since no manure was applied to the CO treatment plots. The reasons for the high FC in runoff

from CO treatment could be the cross-contamination from other treatments or the influence of wildlife or random experimental errors. The sources of cross contamination could be the cross-walking of graduate students through the plots while planting the corn, or setting up the rainfall simulated event, or the blowing wind may have spread some poultry manure to other treatments during its application to the PL treatment.

Table 4.20 Descriptive statistics for the FC concentrations of runoff from different treatments for all the simulated rainfall events

Treatments	Simulation 1 ^a			Simulation 2 ^a			Simulation 3 ^b		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	3.70E+05 ^A	1.00E+06	1.60E+04	3.74E+05 ^A	6.45E+05	1.07E+05	4.95E+01 ^A	9.05E+01	2.30E+01
PL	1.04E+06 ^A	1.74E+06	3.50E+05	1.38E+05 ^A	2.10E+05	6.80E+04	8.63E+01 ^A	1.40E+02	2.60E+01
DS	1.70E+05 ^A	4.20E+05	4.00E+04	2.85E+05 ^A	3.15E+05	2.30E+05	5.72E+01 ^A	6.50E+01	5.15E+01
CO	2.64E+04 ^A	4.00E+04	1.28E+04	8.00E+02 ^B	9.00E+02	7.00E+02	7.68E+01 ^A	8.65E+01	6.65E+01

*The values with different uppercase letters within a column are significantly different from each other. The simulated events with different lowercase letters are significantly different from each other.

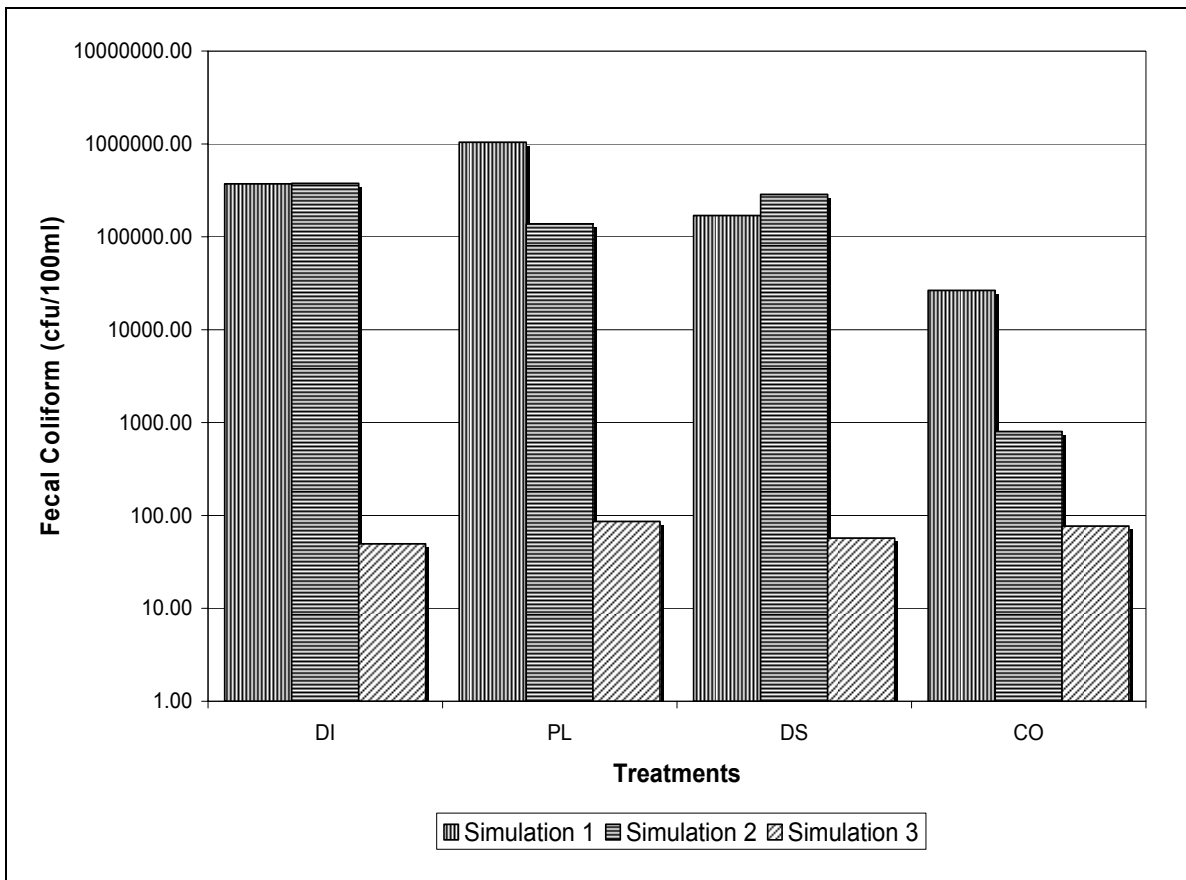


Figure 4.18 Mean FC concentration in runoff from different treatments for all the simulated rainfall events

The concentrations of FC in runoff from all treatments observed in the present study are much higher than the FC standards set by Virginia Department of Environmental Quality (VDEQ, 2003) for any designated water use. In fact, these concentrations violate the water quality standards of any state by orders of magnitude. The FC concentrations reported in the present study are higher than the values reported by other studies by 1 to 3 orders of magnitude (Barker and Sewell, 1973; Janzen et al., 1974 and Robbins et al., 1971). Although these are edge of field concentrations, and some dilution is expected when the runoff mixes into the streams, and some bacterial die off is also expected, these values can at least temporarily raise the FC concentrations of small streams at the confluence points. These results show that manure applications on agricultural lands can negatively impact water quality of downstream waters. The runoff from agricultural lands treated with animal manure needs to be controlled through the implementation of appropriate Best Management Practices (BMP) to reduce downstream pollution of water bodies.

The method of application does not seem to have significant effect on the FC concentration in runoff as no significant difference was observed between the FC concentration in runoff from DI and DS treatments, although incorporation is recommended as a method to reduce pathogen concentrations in runoff (USDA, 2000; Walker et al., 1990). Even one day lapse between the application of manure and the rainfall application could substantially reduce bacterial concentrations in runoff (Crane et al., 1978). The first simulated rainfall event took place within 20-24 hours of manure application, and the soil was wet enough to reduce significant bacteria die-off during the one day period between the first and the second simulated event. No significant change in the FC concentrations of runoff was observed from the first to second simulated event. The concentration of FC from dairy manure treatments (DI and DS) increased slightly during the second simulated event., although runoff volume and peak runoff rate increased from the first to second simulated event and a dilution effect was expected. The increase in bacterial concentrations with increase in runoff volume and peak runoff rate has also been observed in some previous studies (Dudley and Karr, 1979; Kunkle, 1970; Robbins et al., 1971). These results are particularly significant as they show that the release of bacteria from animal manure is gradual and that the bacterial contamination from animal manure treatments can extend over a period of time, unlike

nutrient contamination where significant losses of nutrients were observed during the first simulated rainfall event.

Fecal Coliform was also detected in runoff from the third simulated event. Although the FC concentrations in the third simulated event was significantly lower than those from the first and second simulated events, presence of FC shows that that FC can survive in soils even a month after application. The soil was wet throughout the month because of intermittent heavy rainfall events and probably the high soil moisture content helped the FC to survive through this period. However, the FC concentrations in runoff from the third simulated event were well below the federal standards limit for primary contact recreation waters and this reduction in FC concentrations in runoff from the third simulation event could be attributed to the die off of bacterial population in the time period between second and third simulated rainfall events.

Significant interactions between treatments and simulated events were also observed. The behavior of treatments with respect to other treatments was significantly different for different simulated events.

The FC count in runoff from different treatments for each simulated event are shown in Table 4.21 and mean FC count are graphically illustrated in Figure 4.19. The FC counts per unit area are drawn on a logarithmic scale to show all the values. A study by Kunkle et al. (1979) noted that majority of FC loss occurred during the first rainfall-runoff event and loss of FC in the subsequent events was negligible. Dunigan and Dick (1980) also reported the same trend but the period between the first and second rainfall-runoff event in their study was a dry period. In the present study, the second rainfall-runoff event was responsible for greater FC loss in runoff than the first event, but it was conducted within 24 hours of first simulated event. The greater FC counts in the runoff from second simulated event can be attributed to the increased transport of surface materials with higher volume of runoff. Similar results were reported by Dudley and Karr (1979). However, during the third simulated event, the FC loss in runoff was reduced to less than 0.01% of FC loss in first and second simulated events. The heavy rainfall events and exposure to sun during the one month period between the second and the third rainfall were mainly responsible for the reduction in FC count in runoff from the third

simulate event. These conditions were responsible for the loss of FC in runoff resulting from respective rainfall events and die off of bacteria populations.

Table 4.21 Descriptive statistics for the FC yield in runoff from different treatments for each simulated event

Treatments	Simulation 1			Simulation 2			Simulation 3		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	3.76E+12	1.06E+13	1.00E+11	5.30E+12	8.44E+12	1.38E+12	5.90E+08	1.33E+09	1.28E+08
PL	1.40E+13	2.47E+13	3.21E+12	1.93E+12	2.82E+12	1.00E+12	4.39E+08	8.03E+08	2.22E+08
DS	9.27E+11	2.49E+12	1.36E+11	3.04E+12	4.01E+12	2.41E+12	4.36E+08	5.85E+08	3.09E+08
CO	2.20E+11	3.76E+11	6.41E+10	9.77E+09	9.90E+09	9.64E+09	7.21E+08	1.00E+09	5.41E+08

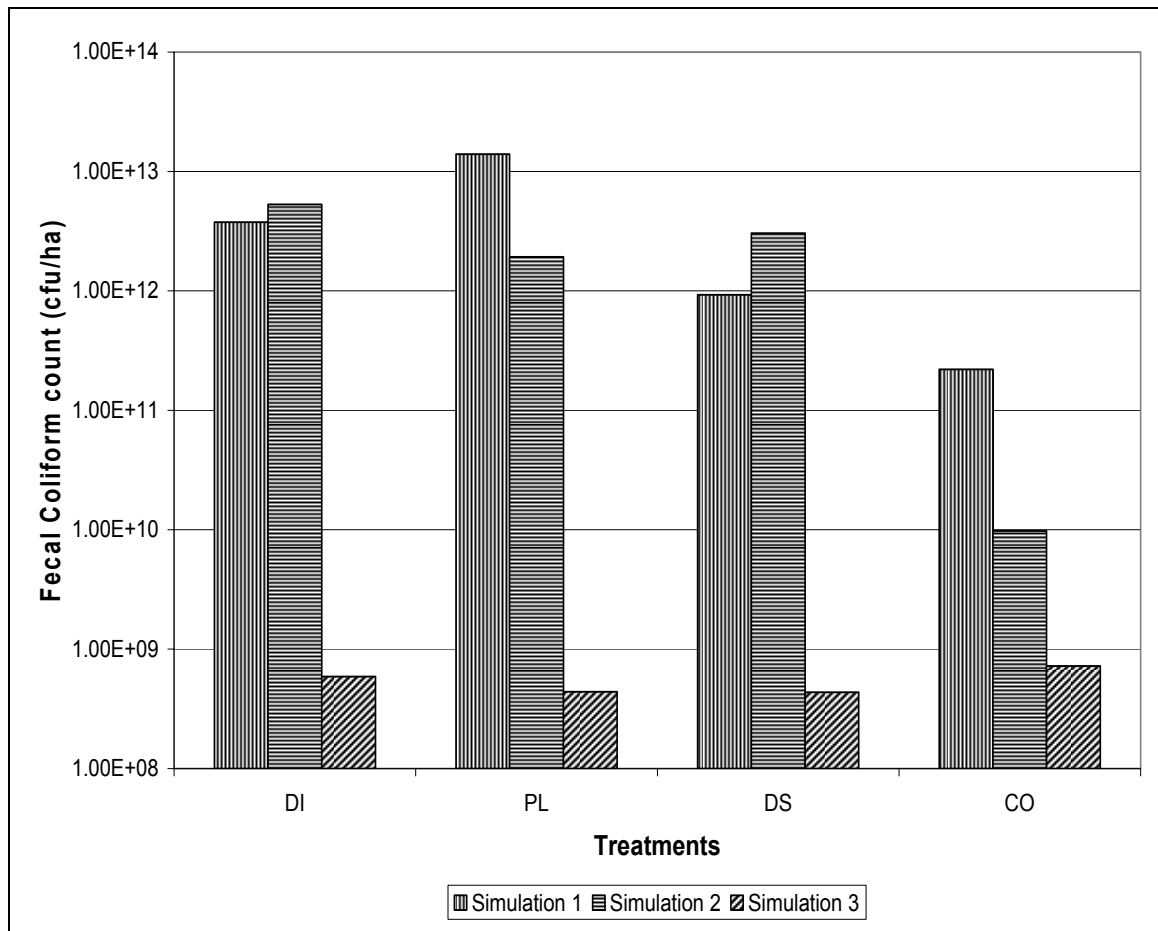


Figure 4.19 Mean amount of Fecal Coliform per unit area in runoff from different treatments for all the simulated events

4.3.2 E. Coli (EC)

Escherichia Coli or E. Coli (EC) concentrations in runoff from different treatments for all the simulated events are shown in Table 4.22 and mean EC concentration are

graphically illustrated in Figure 4.20 Statistical analysis indicated no significant treatment effect on EC concentration for any of the simulated rainfall events. The concentrations of EC for all the treatments were in the same order of magnitude for the first simulated event. For the second simulated event, an increase in EC concentrations compared to first simulation event was observed in DI and PL treatments. Although not significant, this increase in EC concentration is consistent with the observation made by previous studies (Dudley and Karr, 1979; Kunkle, 1970; Robbins et al., 1971). A decrease in concentration of EC from DS treatment was observed from the first to second simulated event; however this decrease was not significant. The presence of bacteria in the runoff from CO treatment can be attributed to the cross-contamination from other treatments, as explained in the section 4.3.2 earlier.

Table 4.22 Descriptive statistics for the E. Coli concentration in runoff from different treatments for all the simulated events*

Treatments	Simulation 1 ^a			Simulation 2 ^a			Simulation 3 ^b		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	6.50E+03 ^A	1.30E+04	2.15E+03	1.05E+04 ^A	1.85E+04	2.40E+03	1.00E+01 ^A	2.90E+01	0.00
PL	1.93E+03 ^A	3.35E+03	5.00E+02	2.76E+04 ^A	6.90E+04	3.20E+03	2.33E+00 ^A	7.00E+00	0.00
DS	8.58E+03 ^A	1.13E+04	7.00E+03	7.75E+03 ^A	1.14E+04	4.15E+03	7.27E+01 ^A	1.95E+02	0.00
CO	1.93E+03 ^A	3.50E+03	3.50E+02	3.63E+03 ^A	9.70E+03	1.00E+02	2.30E+01 ^A	4.00E+01	13.00

*The values with different uppercase letters within a column are significantly different from each other. The simulated events with different lowercase letters are significantly different from each other.

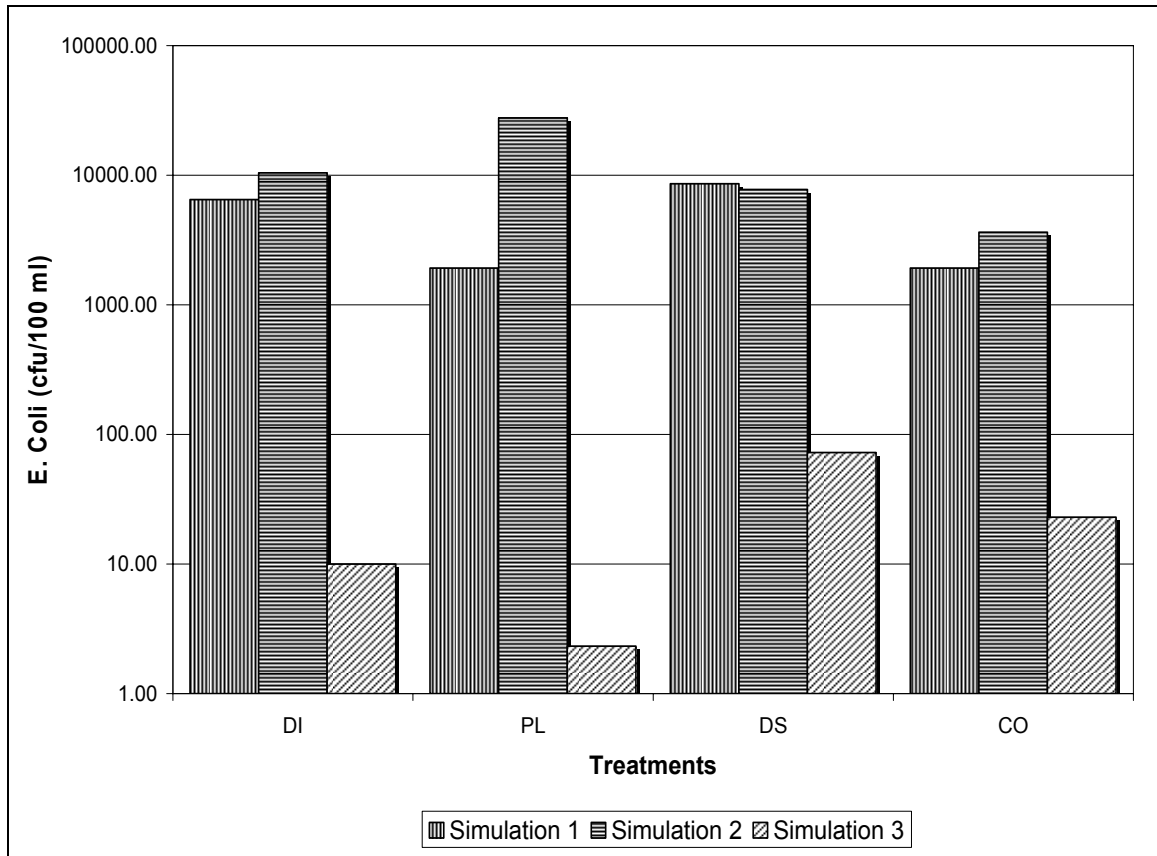


Figure 4.20 Mean E. Coli concentrations in runoff from different treatments for all the simulated events

The concentration of EC from all treatments was 10 to 100 times higher than the water quality standard 126 cfu/100ml set by state and federal agencies. Although, a dilution effect is expected as water moves to downstream water bodies, the concentration of FC in streams can increase temporarily at the confluence point. In small watersheds streams could be severely affected by such high bacterial concentration in runoff.

Statistical comparison suggests significant effects of simulated rainfall events. Mean comparisons suggested that the EC concentrations in runoff from the third simulated event were significantly lower than those measured from the first and second simulated events. The concentration of EC in runoff from the third simulated event was negligible compared to other two events and was below the federal standards for primary contact recreation water.

The mean EC count in runoff per unit area is shown in Figure 4.21 for different treatments and simulated rainfall events. The EC count is presented on a log scale to

show the data for all the simulated events. The EC count lost in runoff is less than the FC count lost in runoff for all treatments and simulated events. The EC lost in runoff increased in second simulated event for all treatments due to increase in concentration and increase in runoff volume. Although the EC concentration in runoff from DS treatment decreased slightly, the increase in runoff volume masked that decrease. The EC count decreased significantly by the third simulated event and the reasons could be die-off of bacteria in the period between the second and the third simulated events as well as the filtering of EC into the soil.

Table 4.21 Descriptive statistics for EC yield in runoff from different treatments for each simulated rainfall event

Treatments	Simulation 1			Simulation 2			Simulation 3		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	4.70E+10	8.13E+10	1.36E+10	8.98E+10	2.38E+11	0.00E+00	1.44E+08	4.27E+08	0.00E+00
PL	2.61E+10	4.77E+10	4.58E+09	3.75E+11	3.75E+11	4.72E+10	2.00E+07	5.99E+07	0.00E+00
DS	3.49E+10	4.44E+10	2.23E+10	5.05E+10	5.05E+10	0.00E+00	4.55E+08	1.24E+09	0.00E+00
CO	1.04E+10	1.75E+10	3.29E+09	3.33E+10	3.33E+10	1.41E+09	2.04E+08	3.19E+08	1.06E+08

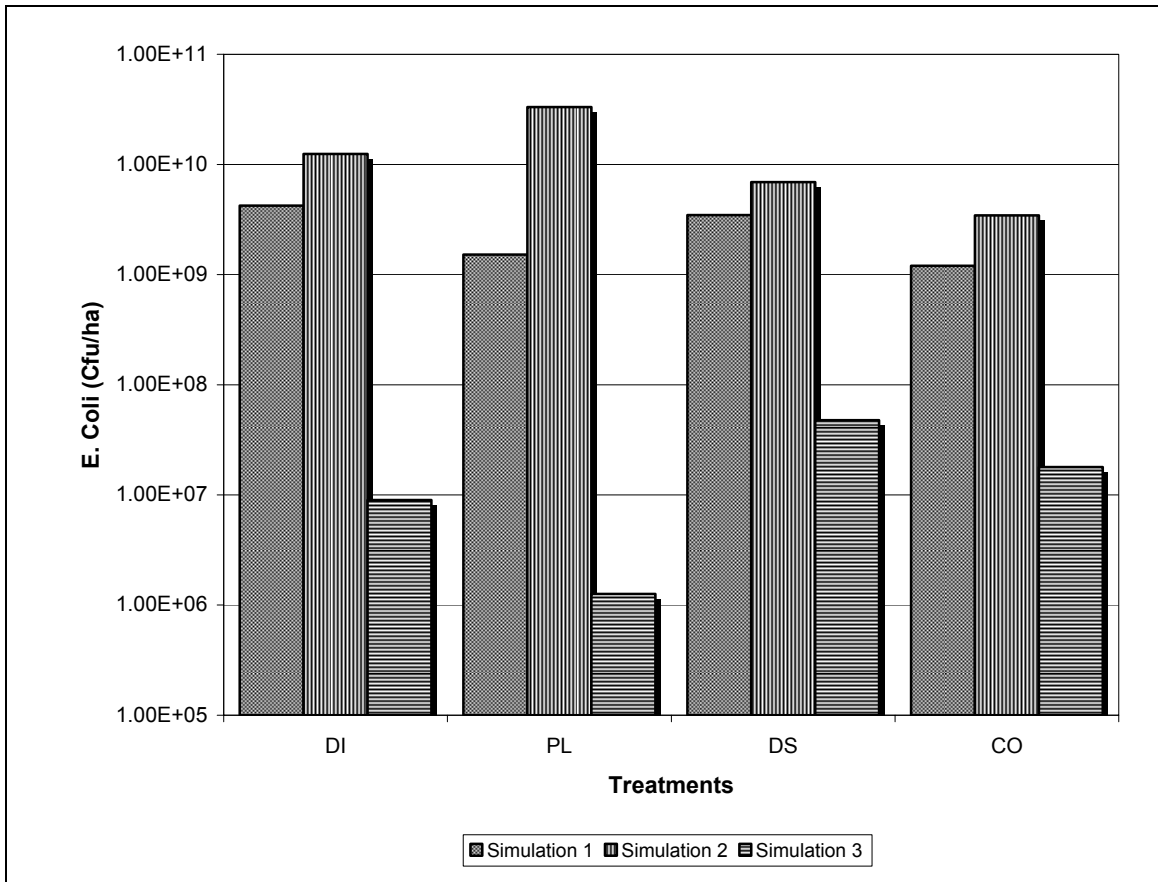


Figure 4.21 Mean counts of E. Coli per unit area in runoff from different treatments for all the simulated events

4.3.3 Enterococcus (ENT)

Enterococcus (ENT) is a subgroup of Fecal Streptococcus. ENT presence in water bodies correlates well with human illness due to fishing and swimming (USDA, 2000). Along with E. Coli, ENT is now being used by all states as pathogen indicators. The concentration of ENT in runoff from different treatments for each simulated event is shown in Table 4.24. The mean EC concentrations in runoff are graphically displayed in Figure 4.22. The concentrations are represented on a log scale to show the small values measured for the third simulated event. Although, no statistically significant treatment effect was observed, the concentration of ENT from the DI treatment (4.14×10^4 cfu/100ml) was an order of magnitude lower than the values in runoff from the PL (2.768×10^5 cfu/100ml) and the DS treatments (7.882×10^5 cfu/100ml) for the first simulated event. The concentrations of ENT from the DS treatment were higher than the concentrations of ENT from the DI treatment (4.15×10^4 cfu/100ml), which indicates an

effect of manure incorporation. However, no such trend could be observed with other bacterial indicators. ENT was also measured in runoff from CO treatment which indicated cross-contamination of CO plots from other treatments and possibility of some experimental errors.

Table 4.22 Descriptive statistics for ENT concentrations in runoff (cfu/100ml) from different treatments for all the simulated events

Treatments	Simulation 1 ^a			Simulation 2 ^a			Simulation 3 ^b		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	4.15E+04 ^A	5.65E+04	1.50E+04	4.30E+04 ^A	7.25E+04	2.45E+04	3.28E+01 ^A	6.80E+01	3.50E+00
PL	2.77E+05 ^A	2.85E+05	2.69E+05	8.58E+04 ^A	9.70E+04	7.50E+04	3.75E+01 ^A	6.45E+01	6.50E+00
DS	7.88E+05 ^A	2.36E+06	3.50E+03	8.28E+04 ^A	1.60E+05	1.75E+04	1.80E+02 ^A	3.11E+02	4.70E+01
CO	3.30E+03 ^A	7.50E+03	9.00E+02	8.55E+03 ^B	1.27E+04	4.20E+03	4.18E+01 ^A	6.15E+01	2.55E+01

*The values with different uppercase letters within a column are significantly different from each other. The simulated events with different lowercase letters are significantly different from each other.

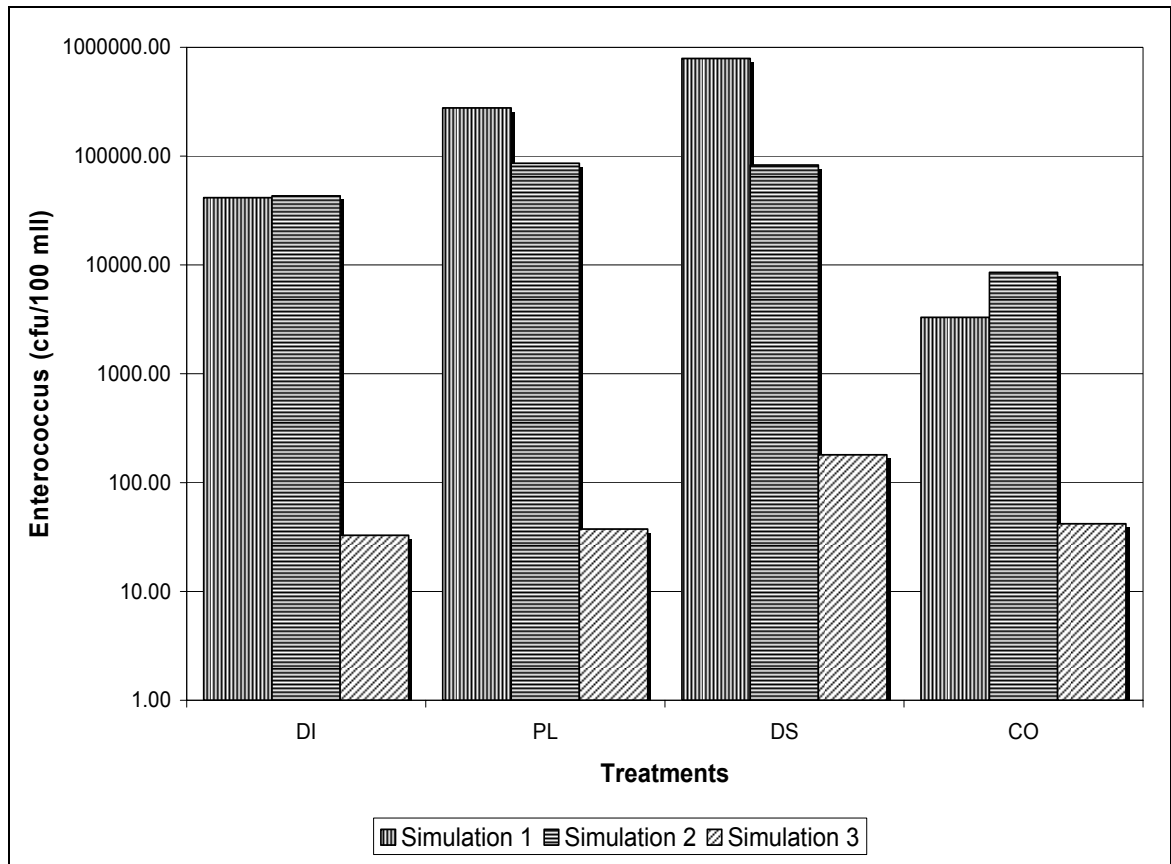


Figure 4.22 Mean concentration of Enterococcus in runoff from different treatments for all the simulated rainfall events

A decrease in concentration of ENT in runoff from the first to second simulated event was observed in DS and PL treatments, while the ENT concentrations from DI treatments remained the same. These results were opposite of the trend observed for other bacterial indicators. However, ENT concentration from CO treatments increased from the first to second simulated event. The ENT concentration in runoff observed from the first and second simulated event was higher by 3 to 4 orders of magnitude than the federal standard for primary contact (33 cfu/100ml). Again, these are runoff concentrations and are expected to be diluted as soon as they reach some surface water source and some die off is also expected, but these concentrations also emphasize the requirement of appropriate BMPs to treat runoff water from cropland before they reach surface water sources.

Statistical analysis suggested that the ENT concentrations in runoff from the third simulated event were significantly lower than those recorded from the first and second simulated events. The concentration of ENT from the third simulation was close to the federal standard limit for primary contact recreation water for DI, PL and CO treatments. However, the concentration of ENT from the DS treatment (180 cfu/100ml) was about six times higher than the federal standard for primary contact recreation water.

Enterococcus count in runoff per unit area is shown in Table 4.23 and the mean ENT count is graphically illustrated in Figure 4.23. The amount is shown on a log scale to show the low values observed in the third simulated event. In general, the ENT yields in runoff are less than the FC and more than the EC yields. The ENT yield increased for DI and CO and decreased for PL and DS treatments.

Table 4.23 Enterococcus yield in runoff (cfu/ha) from different treatments for each simulated rainfall event

Treatments	Simulation 1			Simulation 2			Simulation 3		
	Mean	Max	Min	Mean	Max	Min	Mean	Max	Min
DI	3.41E+11	5.97E+11	9.51E+10	6.42E+11	1.19E+12	3.20E+11	4.59E+08	1.00E+09	1.28E+07
PL	2.14E+12	3.82E+12	0.00E+00	1.22E+12	1.22E+12	1.11E+12	2.32E+08	5.54E+08	5.56E+07
DS	4.66E+12	1.39E+13	1.11E+10	8.39E+11	8.39E+11	2.23E+11	1.43E+09	2.06E+09	2.64E+08
CO	1.96E+10	3.76E+10	7.05E+09	9.41E+10	9.41E+10	4.50E+10	4.09E+08	7.13E+08	2.07E+08

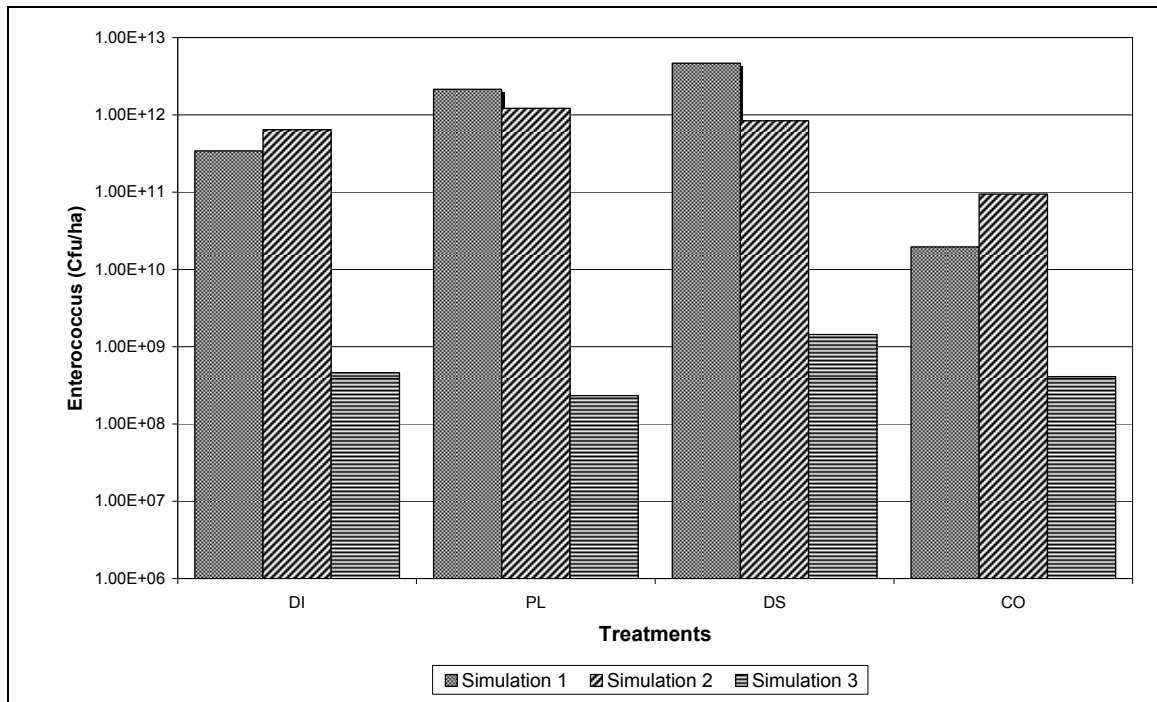


Figure 4.23 Mean Enterococcus count in runoff from different treatments for all the simulated events

4.3.4 Summary of bacteria transport study

The bacteria transport study shows that the edge of field concentrations of selected bacteria in runoff from all treatments violate the federal standards by orders of magnitude. Although, die off and dilution effects are expected before runoff reaches water bodies, the high runoff concentrations observed in this study, could temporarily raise the concentrations at the confluence point in the water bodies. If the watershed is small, high concentrations of such magnitude can impair the downstream water body. The simulated rainfall events in the present study, represent the worst case scenario of rainfall occurring within 24 hours of manure application and as such very little bacteria die off is expected before the start of the rainfall simulation. Manure applications, if avoided when the weather is inclement, can decrease the amount of bacteria in runoff. The runoff from all treatments had the greatest concentration of FC than other bacterial indicators, followed by ENT. Fecal Coliform can include many different bacterial species and hence the FC concentration is highest in runoff. On the other hand, ENT is selective for 20 species of bacteria and E. Coli is selective for only one species for bacteria which may be the reason for higher ENT removed in surface runoff compared to E.Coli.

The FC may have been removed by the downward movement of bacteria, through the infiltrating water, beyond the root zone as observed by Stoddard et al. (1998). Krone et al. (1958) found surface soil to be very effective in reducing the concentrations of bacteria in infiltrating liquids by the process of filtration and adsorption. The large population of bacteria from manure may have been adsorbed on soil particles, filtered by the soil or died-off to result in such a small fraction being removed by runoff. Evans and Owen (1972) observed highest concentrations of E. Coli in drainage water several hours after application of large volume of liquid manure on a sandy clay loam soil. These results show that the movement of E. Coli is quickest through the soil profile and hence the lowest E. Coli was removed by runoff.

Significantly lower concentrations of bacteria were observed in runoff from all treatments during the third simulation. The primary reason attributed to this reduction is the die off of bacteria in the hostile environmental conditions. The bacterial concentrations in runoff were also lower than the federal limits for primary contact recreation during the third simulated event. The presence of bacteria, although at low concentrations, can be explained by the observation made by Mulcock et al. (1973) who reported that once in the soil profile, microorganisms can persist for as long as two months, and that is why we observed bacterial concentrations in runoff during the third simulated event.

Chapter 5 Summary and Conclusions

A plot-scale study was conducted at the Prices Fork Research Farm, Virginia Tech, to evaluate the transport of nutrients and bacteria from conventionally tilled cropland, treated with different animal manures and inorganic fertilizer. The treatments included the surface applied dairy manure (DS), incorporated dairy manure (DI), surface applied poultry litter (PL) and incorporated inorganic fertilizer (IF). Simulated rainfall events were conducted on the plots and discrete runoff samples were collected manually throughout the experiment. The runoff samples were flow-weighted for the nutrient and bacterial analysis. The experiment consisted of three simulated rainfall events. The first simulated event lasted for one hour and was conducted within 24 hours of manure and inorganic fertilizer application. The second simulated event was conducted within 24 hours of the first simulated event until runoff from the field plots reached steady state. The third simulated event was conducted one month after the second simulated event. The primary nutrients of interest were Phosphorus (P) and Nitrogen (N). The samples were also tested for three indicator bacteria: Fecal Coliform (FC), Escherichia Coli (EC) and Enterococcus (ENT).

5.1 Nutrient Transport

Runoff samples were collected and analyzed in the Water Quality Laboratory, Department of Biological Systems Engineering, Virginia Tech. The water quality analysis results show that the runoff concentrations of various P species were significantly higher from poultry litter treatment (PL) than all the other treatments for the first two simulated events. The dissolved reactive phosphorus (DRP) concentrations of runoff from PL treatment were 5-10 times higher than those measured for other treatments. Bioavailable Phosphorus (BAP) concentrations of runoff were 2-5 times higher in runoff from PL treatments than from other treatments. Similar trends were observed for Total Phosphorus (TP) concentrations. Since the amount of P applied to all the treatments was approximately the same, these results indicate that the availability of P in poultry litter is higher than dairy manure. These results also indicate that the P in poultry litter is in highly soluble and bioavailable form. The P concentrations of runoff reported in this study were higher than the EPA nutrient criteria of 10 µg/ml to cause severe overenrichment of nutrients in waterbodies. The P concentrations decreased significantly during the third simulated event for all the treatments, due to plant uptake of nutrient, washing away of nutrients by heavy rainfall events which occurring between the second

and third simulated rainfall events, and the various processes taking place in soil, responsible for sorption and fixation of P.

The Nitrate (NO_3^- -N) form of Nitrogen measured in runoff was not significantly different from the source water concentrations for most of the treatments. The DS treatment consistently resulted in higher NO_3^- -N concentrations in runoff, but it was less than 0.002 times the concentration considered lethal for aquatic organisms. The Ammonia-Nitrogen (NH_4^+ -N) concentration was highest from PL treatments and the primary reason could be the presence of higher NH_4^+ -N in poultry litter compared to dairy manure and the slower hydrolysis of NH_4^+ -N in poultry litter. However, the NH_4^+ -N concentrations in runoff from all the treatments were below the acute limits of NH_4^+ -N in fresh waters, imposed by Virginia Department of Environmental Quality (VDEQ, 2001). The Total Nitrogen (TN) concentrations of runoff from all treatments were higher than the EPA recommended criteria of 0.46 mg/l, to cause overenrichment of waterbodies. The TN concentrations were significantly higher than the sum of NO_3^- -N and NH_4^+ -N, mainly due to the presence of excess organic nitrogen in the soil. Significantly higher TN concentration was observed from PL treatments compared to other treatments. Although the TN concentrations decreased significantly from the third simulated event compared with those measured from the third simulated event, the TN concentrations were significantly higher than the 0.46 mg/l criteria.

The nutrient analysis results show that the manure application to cropland based on P requirements of crop does not necessarily prevent nutrient enrichment of downstream waterbodies. Heavy rainfall events following manure application could increase the nutrient input to downstream waterbodies. Based on the P results, it can be inferred that the runoff should be controlled from agricultural fields through implementation of appropriate BMPs, to reduce soluble P input to downstream waterbodies, especially from poultry litter-treated fields. Reducing runoff also would reduce erosion and other associated parameters such as Total Suspended Solids (TSS) and sediment bound pollutants.

5.2 Bacterial Transport

The runoff samples were also analyzed for three indicator bacterial species within five hours of sample collection. Bacterial concentrations reported from all treatments, except from the CO (Control) plots were orders of magnitude higher than the federal and state limits established for primary contact recreation waters. Although these are edge of field concentrations and some dilution and die-off is expected as the runoff reaches waterbodies, the concentrations of this magnitude can temporarily raise the levels of these indicator bacteria at the confluence point of a waterbody. The bacterial concentrations reported in this study were generally higher than the results reported in the literature for similar studies. One possible reason for higher bacteria concentrations in this study is that the rainfall was applied within 24 hours of manure application and no sufficient time was available for the bacterial die-off. No significant difference was observed among treatments regarding bacteria concentrations in runoff. Although the volume and peak runoff rate increased significantly from the first to second simulated event, the concentration and yield of the indicator bacteria in runoff during the second simulated event generally increased, compared with the first simulated event. Apparently, the increased runoff during the second event caused more organic materials to float and move with the runoff. A significant decrease in the bacterial concentration was observed from the third simulated event, compared with the second event. The one month time lapse between the second and third simulated period, along with heavy rainfall events during this period were the main cause of bacterial die-off and bacterial removal in the runoff.

The highest concentrations and yield in runoff was observed for Fecal Coliform (FC), followed by Enterococcus (ENT) and Escherichia Coli (EC). A similar trend was observed for the amount of bacteria in runoff. The primary reason for differences in bacteria concentrations was that the FC includes many bacterial species, while ENT includes 20 different bacterial species and EC includes only one. Other reasons could be retention of bacteria in the soil particles and the infiltration of bacteria to the subsurface. Although the bacterial concentrations of top soil and the subsurface were not measured, the available literature supports these assumptions. It can be hypothesized that the EC is more preferentially retained and infiltrated by soil followed by ENT and FC. However, a detailed investigation is required for more information on soil retention and infiltration of bacteria.

Application of manure to cropland is generally based on the nutrient requirement of the crop and no consideration is given to the bacterial contamination resulting from it. This study shows that manure application can result in bacterial contamination of runoff, and consequently the downstream waterbodies. The runoff from manure-treated agricultural lands should be controlled using appropriate BMPs to alleviate bacterial contamination of downstream waterbodies.

5.3 Conclusions

The following conclusions can be drawn from the study.

Objective 1

- The concentrations of dissolved reactive phosphorus (DRP) and bioavailable phosphorus (BAP) were higher in runoff from croplands treated with the surface applied poultry manure than with dairy manure in the rainfall-runoff events occurring one and two days after manure application.
- The total phosphorus (TP) concentrations of runoff from the treatments were not significantly different for the individual rainfall-runoff events, but the TP concentrations were 100 to 1000 times the EPA nutrient criteria for nutrient overenrichment of waterbodies.
- Total Nitrogen (TN) concentrations of runoff from poultry litter treated plots were higher than all other treatments for rainfall-runoff events occurring within one and two days of manure application and the TN concentrations from all the treatments were higher than EPA nutrient criteria for nutrient overenrichment of waterbodies.
- The nutrient concentration from all the treatments decreased significantly in the runoff event occurring one month after the manure application, and the probable reasons for these reductions are washing away of nutrients during the natural rainfall-runoff events occurring in one month period, plant uptake of nutrients, leaching and processes leading to immobilization and transformation of nutrients.

Objective 2

- No significant effect of type of manure was observed on indicator bacterial concentration of runoff from the plots. No significant effect of method of application was observed for the dairy manure on indicator bacterial concentrations of runoff.

- The bacterial concentrations in runoff from all treatments were 10^4 to 10^5 times higher than federal and state limits for the primary contact recreation waters, during rainfall-runoff events occurring within one and two days of manure application.
- The highest bacterial yield was observed for the Fecal Coliform followed by Enterococcus and E. Coli which might be because of higher retention and infiltration of E. Coli in the soil.
- The Fecal Coliform and E. Coli concentration generally increased in the second rainfall-runoff event occurring two days after manure application, due to high runoff volume and high peak runoff rate during the second event.
- The bacterial concentrations in runoff from the the rainfall-runoff event occurring one month after the manure application declined significantly due to die-off of bacteria and washing away in the runoff resulting from natural rainfall events in the one month period between the second and third simulated rainfall events.

5.4 Implications and Recommendations

With the increase in animal agriculture and resulting manure production, and reduced availability of land, excessive amounts of manure are applied to agricultural fields. Manure, if not properly applied could result in nutrient and bacterial pollution of downstream waterbodies. Manure application based on P requirement of crop has been suggested as a practical way to reduce nutrient pollution of downstream waterbodies. The present study suggests that this practice can also pollute downstream waterbodies. The high nutrient concentrations in runoff reported in the present study could cause eutrophication problems and nutrient overenrichment of downstream waterbodies. The study results indicate that poultry litter can cause significant phosphorus loss to downstream waterbodies, compared with the other treatments investigated. The P from poultry litter is mostly water soluble and hence to control the nutrient pollution, efforts should be concentrated on controlling runoff from agricultural fields treated with poultry litter. Dairy manure was also a significant source of runoff nutrient enrichment but requires less attention than poultry litter. The focus of BMP implementation on the agricultural fields should be to control runoff, and manure applications should be avoided when rainfall is imminent.

Manures could be a potential significant source of bacterial contamination of waterbodies. The study showed that the bacterial concentrations in runoff from various

manures applied to cropland are several orders of magnitude higher than the federal and state limits established for primary contact recreation waters. The type of manure and the application method did not significantly affect bacterial concentration in runoff. The results from this study suggest that manure application should be avoided on agricultural fields at times when rainfall is imminent and sufficient BMPs need to be installed to reduce loss of bacteria in runoff.

5.5 Recommendations for future study

Several recommendations can be made for future studies based on the results reported from the present study. This study investigated nutrient losses from cropland. The nutrient loss from other agricultural systems and soils needs to be studied as well. As reported in this study, significant nutrient loss takes place in the dissolved phase; therefore proper BMPs need to be developed to control the runoff from the agricultural lands. A reliable method for bacterial analysis needs to be developed so that the confidence in results and consequently, models can be increased. There is a need for an in-depth investigation of the fate of bacteria applied on the soils. Apart from die off, some retention and infiltration also takes place which needs to be quantified and modeled. Appropriate BMPs need to be evolved and tested for their effectiveness for trapping bacteria and nutrients.

References

- Azedevo, J. and, P.R. Stout. 1974. Farm animal manure: An overview of their role in the agriculture environment. *California Agriculture Experiment Station Extension Service, Manual 44, 109pp.*
- Baker, J.L., J.M. Lafen, A. Mallarino, and B. Stewart. 2001. Developing a phosphorus index using WEPP. In Proc. Int. Symp. (3-5 January 2001, Honolulu, HI, USA)Eds. J.C. Ascough II and D.C. Flanagan. St. Joseph, MI: ASAE.701P0007
- Birr, A.S., and D.J. Mulla. 2001. Evaluation of the phosphorus index in watersheds at the regional scale. *J. Environ. Qual.* 30: 2018-2025.
- Brady, N.C., and R.R. Weil. 1999. Soil phosphorus, potassium, and micronutrients. *Elements of the Nature and Properties of Soil. Upper Saddle River, NJ:Prebtice Hall.* 391-434.
- Burwell, R.E., G.E. Schuman, H.G. Heinemann, and R.G. Spomer. 1977. Nitrogen and phosphorus movement from agricultural watersheds. *J. Soil and Water Cons.* 226-231.
- Conley, D. J. 2000. Biogeochemical Nutrient Cycles and Nutrient Management Strategies. *Hydrobiologia.* 410: 87-96.
- Crane S.R. and J.A. Moore. 1985. Modeling enteric bacterial die off: A review. Journal paper No. 6699. *Oregon Agricultural Experiment Station, Corvallis.*
- Crane, S.R., J.A. Moore, M.E. Grismer and J.R. Miner. 1983. Bacterial pollution from agricultural sources: A review. *Trans. ASAE.*:858-866.
- Crane, S.R., M.R. Overcash, and P.W. Westermann. 1978. Swine manure microbial die off and runoff transport under controlled boundary conditions. *Unpublished paper*, 15 pp.
- CBF. 1998. The need to regulate poultry: What Science and Experts Say. Fact Sheet. Chesapeake Bay Foundation.
- Chinkuyu, A.J., and R.S. Kanwar. 2001. Predicting soil nitrate-nitrogen losses from incorporated poultry manure using the GLEAMS model. *Trans ASAE.* 44(6): 1643-1650.
- Delgado, J.A. 2002. Quantifying the Loss Mechanisms of Nitrogen. *J. of Soil and Water Conservation.* 57(6): 389-398.
- Doran, J.W., and D.M. Linn. 1979. Bacteriological quality of runoff water from pastureland. *Applied Environmental Microbiology.* 37(5): 985-991.
- Dudley, D.R., and J.R. Karr. 1979. Concentration and sources of fecal and organic pollution in an agricultural watershed. *Water Resource Bull.* 15(4): 911:923.
- Edwards, D.R., and T.C. Daniel. 1993. Effects of poultry litter application rate and rainfall intensity on quality of runoff from fescuegrass plots. *J. Environ. Qual.* 22: 361-365.

- EFU Manual. 2002. Efficient Fertilizer Use Manual. IMC Global. Available at http://www.imcglobal.com/general/efu_manual.htm Aug 23, 2003.
- Eghball, B., and J. E. Gilley. 1999. Phosphorus and nitrogen in runoff following beef cattle manure or compost application. *J. Environ. Qual.* 28(4): 1201-1210.
- ELA. 2003. Eutrophication (Nutrient Pollution). Experimental Lakes Area. Available at <http://www.umanitoba.ca/institutes/fisheries/eutro.html> Jan 20, 2003.
- ESRI, Incorporated. 1999. ArcGIS 8.3. © Copyright 1999-2002.
- Follett, R.F., and J.A.Delgado. 2002. Nitrogen fate and transport in agricultural systems. *J. Soil and Water Cons.* 57(6):402-408.
- Garbrech, J., and A.N. Sharpley. 1992. Sediment-phosphorus relationships in watersheds. pp. 601-610. In P. Larson (ed.) Sediment management. Proc. 5th Intl Symp. On river sedimentation, Karlsruhe, Germany, 5-9 Apr. Unive of Karlsruhe Press.
- Geldrich, E.E., R.H. Bordner, C.B. Huff, H.F. Clark, and P.W. Kabler. 1962. Type distribution of coliform bacteria in the feces of warm blooded animals. *J. Water Poll. Control Fed.* 34(3): 295-301.
- Gilbert, Richard O. 1987. Statistical Methods for Environmental Pollution Monitoring, 148-149. John Wiley and Sons Inc.
- Hanson, N.C., T.C. Daniel, A.N. Sharpley, and, J.L. Lemunyon. 2002. The fate and transport of phosphorus in agricultural systems. *J. Soil and Water Conservation.* 57(6): 408-417.
- Heathman, G.C., A.N. Sharpley, S.J. Smith, and J.S. Robinson. 1995. Land application of poultry litter and water quality in Oklahoma, U.S.A. *Fertilizer Research.* 40: 165-173.
- Howarth, R., D. Anderson, J. Cloern, C. Elfring, C. Hopkinson, B. Lapointe, T. Malone, N. Marcus, K. McGlarchy, A. N. Sharpley, and D. Walker. 2000. Nutrient pollution of coastal rivers, bays and seas. *In Issues in Ecology. Ecological Society of America, Washington, D.C.*
- Howell, J.M., M.S. Coyne, and P.L. Cornelius. 1996. Effect of sediment particle size and temperature on fecal bacteria mortality rates and the fecal Coliform/fecal streptococci ratio. *J. Environ. Qual.* 25: 1216-1220.
- Jack, E. and P. Hepper. 1969. An outbreak of Salmonella Typhmuriium infection in cattle associated with the spreading of slurry. *Vet. Rec.* 84:198.
- Kenner, B.A., H.F. Clark, and P.W. Kabler. 1960. Fecal Streptococci II. Quantification of streptococci in feces. *Amer. J. Pub. Health* 50:1553-1559.
- Khaleel, R.G., G.R. Foster, K.R. Reddy, M.R. Overcash, and P.W. Westerman. 1979. A nonpoint source model for land areas receiving animal wastes: III. A conceptual model for sediment and manure transport. *Trans ASAE.* 11(6): 1353-1361.

- Kunkle, S.H. 1979. Using bacteria to monitor the influences of cattle waste on water quality. USDA-SEA-ARR. ARR-NE-3, September.
- Kunkle, S.H. 1970. Concentration and cycles of bacterial indicators in farm surface runoff. In: Relationship of agriculture to soil and water pollution. Cornell Univ. Conf. on Agricultural Waste Management. Jan 19-21. Cornell Univeristy, Ithaca, NY. pp 49-60.
- Krogstad, T., and O. Lovstad. 1989. Erosion, phosphorus and phytoplankton response in rivers of Southeastern Norway. *Hydrobiologia*. 183:33-41.
- Landry, M.S., and M.L. Wolfe. 1999. Fecal bacteria contamination of surface waters associated with land application of animal waste. Presented at the July 18-21, 1999 ASAE/CSAE-SCGR Annual International Meeting, Paper No. 994024. St. Joseph, MI 49085-9659.
- Levine, S.L., and D.W. Schindler. 1989. Phosphorus nitrogen and carbon dynamics of experimental lake 303 during recovery from eutrophication. *Can. J. Fish Aquatic Sci.* 46:2-10.
- Miner, J.R., L.R. Fina and C. Platt. 1967. *Salmonella infantis* in cattle feedlot runoff. *Appl. Microbiol.* 15:627:628.
- Moore, J.A., J Smyth, S. Baker, and J.R. Miner. 1988. Evaluating coliform concentrations in runoff from various animal waste management systems. *Special report 817, Agriculture Experiment Station, Oregon State University, Corvallis.*
- Mulcock, A.P., M.J. Noonan, and C.A. Reay. 1973. Soil contamination by septic tank effluent. Lincoln College, Canterbury, New Zealand.
- National Resource, Agriculture and Engineering service. 2000. Managing nutrients and pathogens from animal agriculture. *NRAES-130*
- Nixon, S. W. 1995. Coastal Marine eutrophication: a definition, social causes and future concerns. *Ophelia* 41: 199-219.
- Patni, N.K., R. Toxopeus, A.D. Tenant, and F.R.Hore. 1985. Bacterial quality of runoff from manured and non-manured cropland. *Trans ASAE*. 28: 1871-1877.
- Paul, J.A. Withers, Stephen D. Clay, and Victor G. Breeze.2001. Phosphorus transfer in runoff following application of fertilizer, manure and sewage sludge. *J. Environ. Qual.* 30: 180-188.
- Pease, J., M. Alley, and S. Mostaghimi. 1998. *Presentation to Senate Subcommittee Studying. HB1207.*
- Pease, James. 2002. Organic waste generation and disposal in Virginia. *Presentation to AAEC.*

- Peters, R.H. 1981. Phosphorus availability in Lake Memphremagog and its tributaries. *Limnol. Oceanogr.* 26: 1150-1161.
- Reddy, G.B., C.W. Raczkowski, M.R. Reyes, and G.A. Gayle. 2001. Surface losses of N, P, and herbicides from a long term tillage study at North Carolina A & T state university. *ASAE Pub #701P0007*.
- Risse, L.M., and J.E. Gilley. 2001. Modeling the impacts of manure on soil and water losses. Proc. Int. Symp. (3-5 January 2001, Honolulu, HI, USA). Eds. J.C. Ascough II and D.C. Flanagan. St. Joseph, MI: ASAE.701P0007
- Rosen, Barry H. 2000. Waterborne Pathogens in Agricultural Watersheds. *Watershed Science Institute*. USDA.
- Schuman, G.E., R.G. Spomer, and R.F. Piest. 1973. Phosphorus losses from four agricultural watersheds on Missouri valley loess. *Soil Sci. Soc. Am. Proc.* 37:424-427.
- Senate Agriculture, Nutrition and Forestry Committee. 1997. Animal Waste Pollution: An Emerging National Problem. *United States Senate*.
- Sharpley, A.N. 1995. Dependence of runoff phosphorus on extractable soil phosphorus. *J. Environ. Qual.* 24: 920-926.
- Sharpley, A.N. 1995. Identifying sites vulnerable to phosphorus loss in agricultural runoff. *J. Environ. Qual.* 24: 947-951.
- Sharpley, A.N. 1997. Rainfall frequency and nitrogen and phosphorus loss amended with poultry litter. *J. Environ. Qual.* 26(4): 1127-1132.
- Sharpley, A.N., and B. Moyer. 2000. Phosphorus forms in manure and composts and their release during simulated rainfall. *J. Environ. Qual.* 29: 1462-1469.
- Sharpley, A.N., and Hubert Tunney. 2000. Phosphorus research strategies to meet agricultural and environmental challenges of the 21st century. *J. Environ. Qual.* 29: 176-181.
- Sharpley, A.N., R.W. McDowell, J.L. Weld, and P.J.A. Kleinman. 2001. Assessing site vulnerability to phosphorus loss in agricultural watershed. *J. Environ. Qual.* 30: 2026-2036.
- Sharpley, A.N., T.C. Daniel, and D.R. Edwards. 1993. Phosphorus movement in the landscape. *J. Prod. Agric.* 6:492-500.
- Sharpley, A.N., S.J. Smith, O.R. Jones, W.A. Berg, and G.A. Coleman. 1992. The transport of Bioavailable Phosphorus in Agricultural Runoff. *J. Environ Qual.* 21:30-35.
- Sharpley, A.N., S.J. Smith, and J.W. Naney. 1987. The environmental impact of agricultural nitrogen and phosphorus use. *J. Agric. Food Chem.* 36:812-817.

- Smallbeck, D.R. and M.C. Bromel. 1975. Bacterial analysis and land disposal of farm based lagoon waters. In Proc. 3rd International Symposium on Livestock Wastes, 318-321. Urbana Champaign, IL, April.
- Stoddard, C.S., M.S. Coyne and J.H. Grove. 1998. Fecal Bacteria Survival and Infiltration through a Shallow Agricultural Soil: Timing and Tillage effects. *J. Environ Qual.* 27:1516-1523.
- Soupir, M. 2003. Release and Transport of Bacteria from Nutrients from Livestock Manure Applied to Pastureland. Virginia Tech, Blacksburg, Virginia, United States.
- Topcon. Total Survey Stations. © Copyright 2002 Topcon Positioning Systems.
- U.S.EPA. 1999. Animal feeding operation unified strategy. Available at http://cfpub.epa.gov/npdes/afo/ustrategy.cfm?program_id=7 July 31, 2002.
- USEPA. 2002. Water Quality Inventory Report. U.S. Environmental Protection Agency. Available at <http://www.epa.gov/305b/2000report> May 18, 2003.
- USEPA. 2003. Overview of Current Total Maximum Daily Load -TMDL- Program and Regulations.: U.S. Environmental Protection Agency. Available at <http://www.epa.gov/owow/tmdl/overviewfs.html> April 10, 2003.
- USEPA. 2003. Eutrophication. U.S. Environmental Protection Agency. Available at <http://www.epa.gov/maia/html/eutroph.html> Mar 11, 2003.
- USEPA. 1994. National Water Quality Inventory: 1994 Report to Congress. United States Environmental Protection Agency. Available at <http://www.epa.gov/305b/94report/index.html> Dec 16, 2002.
- USDA. 2000. Waterborne Pathogens in Agricultural Watersheds. Barry H. Rosen. University of Vermont, Burlington.: NRCS, Watershed Science Institute.
- VaDCR. 1995. Virginia Nutrient Management Standards and Criteria. Richmond, Virginia.
- VaSWCB. 2003. Water Quality Standards. Virginia State Water Control Board. Statutory Authority: § 62.1-44.15(3a) of the Code of Virginia.
- Van Donsel, Dale J., Edwin E. Geldrich and Norman A. Clarke. 1967. Seasonal variations in survival of bacteria in soil and their contribution to storm-water pollution. *American Soc. of Microbiology.* 15. 1362-1370.
- Vietor, D.M., R.H. White, C.L. Munster, and T.L. Provin. 2002. Reduced nonpoint source pollution through manure use and export in turfgrass sod. In Proceedings of the March 11-13, 2002 Conference, (Fort Worth, Texas, USA) Publication Date March 11, 2002. ASAE Publication Number 701P0102, ed. Ali Saleh.
- Vollenweider, R.A. 1968. Scientific fundamentals of the eutrophication of lakes and flowing waters with particular reference to nitrogen and phosphorus as factors in eutrophication. Tech. Rep. DA 5/SCI/58.27. OECD. Paris.

- Vories, E.D., T.A. Costello, and R.E. Glover. 2001. Runoff from cotton fields fertilized with poultry litter. *Trans. ASAE*. 44(6): 1495-1502
- Walker, S.E., S. Mostaghimi, T.A. Dillaha, and F.E. Woeste. 1990. Modelling animal waste management practices: Impacts on bacteria levels in runoff from agricultural lands. *Trans. ASAE*. 33(3)(5/6): 807-811.
- Wang, L., and K.R. Mankin. 2001. Dieoff and release of fecal pathogens from animal manure. ASAE Meeting Paper No. 01-2237. St. Joseph, Mich.: ASAE.
- Westerman, P.W., T.L. Donnelly, and M.R. Overcash. 1983. Erosion of soil and poultry manure- A laboratory study. *Trans. ASAE* 26:1070-1078, 1084.
- Whalen, Joann K., and Chi Chang. 2001. Phosphorus accumulation in cultivated soils from long term annual applications of cattle feedlot manure. *J. Environ. Qual.* 30: 229-237.
- Wood, B.H., C.W. Wood, K.H. Yoo, K.S. Yoon, and D.P. Delany. 1999. Seasonal surface runoff losses of nutrients and metals from soils fertilized with broiler litter and commercial fertilizer. *J. Environ. Qual.* 28(4): 1210-1218.
- Walker, S.E., S. Mostaghimi, T.A. Dillaha, and F.E. Woeste. 1990. Modeling animal waste management practices: Impacts on bacteria levels in runoff from agricultural lands. *Trans. ASAE*. 33(3) (5/6): 807-811.
- Wudi, Zhang, Song Hongchuan, Li Jianchang, Wei Xiaokui. 2002. Comprehensive utilization of human and animal wastes. Provincial key lab of Rural Energy Engineering, Yunnan Normal University, Kunming. P. R. China.

Appendix A Manure Analysis

Table A1 Dairy manure analysis results

Parameter	Dairy Manure					
	4/26/03 (Before Storage)			5/13/03 (After Storage)		
Date	1	2	3	1	2	3
Replication	1	2	3	1	2	3
Ammonium Nitrogen (lbs/1000gal)	6.18	6.18	6.51	6.59	5.17	6.59
Organic Nitrogen (lbs/1000gal)	7.76	7.68	7.09	7.43	8.85	7.34
Nitrate Nitrogen (lbs/1000gal)	0.58	0.47	0.42	0.42	0.06	0.20
Available Nitrogen (incorporated) (lbs/1000gal)	10.17	10.01	9.89	10.15	9.51	9.88
Available Nitrogen (Surface Applied) (lbs/1000gal)	8.32	8.16	7.93	8.17	7.95	7.90
Phosphorus as P₂O₅ (lbs/1000gal)	6.55	6.42	6.99	5.81	4.57	6.33
Potassium as K₂O (lbs/1000gal)	16.46	15.97	17.10	14.89	15.29	16.62
Calcium (lbs/1000gal)	5.34	5.53	5.97	5.30	5.42	5.81
Magnesium (lbs/1000gal)	2.75	2.73	2.89	2.61	2.35	2.88
Sulfur (lbs/1000gal)	1.35	1.44	1.56	1.35	1.33	1.44
Zinc (lbs/1000gal)	0.04	0.05	0.05	0.04	0.04	0.05
Copper (lbs/1000gal)	0.11	0.12	0.13	0.11	0.12	0.12
Manganese (lbs/1000gal)	7.04	0.06	0.07	0.06	0.05	0.06
Sodium (lbs/1000gal)	2.73	2.74	2.74	2.23	2.29	2.50
pH	6.5	6.5	6.5	6.6	7.3	6.5
Moisture	96.89%	96.55%	96.55%	96.08%	96.68%	96.32%
Calcium Carbonate Equivalency (lbs/1000gal)	2457.6	3848.71	3646.76	326.29	326.29	353.83
Soluble Phosphorus (lbs/1000gal)	2.34	2.24	2.79	2.29	0.94	2.33

Table A2 Chicken Litter analysis results

Parameter	Chicken Litter			
	Previous Analysis Results	5/13/03 (Fresh Litter)		
Replication	1	1	2	3
Ammonium Nitrogen (lbs/ton)	15.00	14.60	11.60	13.40
Organic Nitrogen (lbs/ton)		51.73	56.99	49.08
Nitrate Nitrogen (lbs/ton)		1.19	1.11	0.93
Available Nitrogen (incorporated) (lbs/ton)	51.19	44.63	45.25	41.65
Available Nitrogen (Surface Applied) (lbs/ton)	45.19	40.25	41.77	37.63
Phosphorus as P ₂ O ₅ (lbs/ton)	49.27	67.18	59.54	60.62
Potassium as K ₂ O (lbs/ton)	45.77	60.31	56.89	56.20
Calcium (lbs/ton)	34.00	48.03	38.64	40.48
Magnesium (lbs/ton)	7.31	9.64	9.59	8.82
Sulfur (lbs/ton)	7.84	11.51	9.88	10.08
Zinc (lbs/1000gal)	0.58	0.77	0.82	0.71
Copper (lbs/ton)	0.54	0.74	0.64	0.69
Manganese (lbs/ton)	0.52	0.66	0.62	0.64
Sodium (lbs/ton)	8.26	12.39	11.21	11.62
pH		8.6	8.6	8.7
Moisture	33.59%	28.09%	27.37%	29.97%
Calcium Carbonate Equivalency (lbs/ton)		42.14	28.33	0.00
Soluble Phosphorus (lbs/ton)		10.35	9.59	9.80

Appendix B Soil Properties

Table B1 Particle size analysis of soil samples

Sample ID	Block 1			Block 2			Block 3		
Replication Number	1	2	3	1	2	3	1	2	3
% VCS	4.7	5.7	5.3	5.3	5.3	4.8	6.5	5.1	5.1
%CS	3.0	3.6	3.6	3.7	3.3	3.0	4.4	3.4	2.7
%MS	5.1	4.1	4.6	4.3	5.1	5.1	5.9	5.7	5.3
%FS	0.2	9.8	8.9	8.9	9.0	9.0	7.7	11.2	9.7
%VFS	15.7	0.6	9.0	10.2	7.7	6.6	7.7	8.4	6.2
Total % Sand	28.6	23.9	31.4	32.5	30.3	28.4	32.2	33.7	29.1
%CSI	14.7	21.0	10.6	11.5	12.4	10.9	8.1	9.5	12.5
%MSI	34.5	28.3	31.0	29.2	34.3	32.9	36.1	32.9	34.0
%FSI	9.6	12.1	15.7	14.0	12.0	15.0	12.6	12.1	13.8
Total % Silt	58.9	61.4	57.3	54.8	58.6	58.8	56.7	54.5	60.3
Total % Clay	12.5	14.7	11.2	12.8	11.0	12.8	11.1	11.8	10.6
Textural Class	SIL	SIL	SIL	SIL	SIL	SIL	SIL	SIL	SIL

Table B2 Chemical Properties of the Soil

Sample ID	1(03/12/03)	Block 1 (05/13/03)			Block 2 (05/13/03)			Block 3 (05/13/03)		
Replication	1	1	2	3	1	2	3	1	2	3
pH	5.99	5.57	5.72	5.62	5.59	5.59	5.69	5.85	5.82	5.85
P	7	4	4	4	4	5	5	7	7	7
K	85	116	103	115	110	105	105	114	106	102
Ca	611	521	489	521	501	501	478	548	543	527
Mg	116	97	92	97	96	96	92	119	119	114
Zn	1.5	0.9	0.9	0.9	0.9	0.9	0.8	1.0	0.9	0.9
Mn	17.7	10.0	9.7	10.5	10.6	10.5	10.2	10.7	10.0	10.1
Cu	1.1	0.2	0.2	0.2	0.3	0.3	0.3	0.2	0.2	0.2
Fe	8.5	7.1	6.8	7.0	8.1	8.3	8.5	6.4	6.0	6.3
B	0.3	0.4	0.4	0.3	0.3	0.3	0.3	0.4	0.4	0.4

Appendix C Rainfall Data

Table C1 Rainfall reported from different raingages for all the simulated rainfall events

Raingage	Simulated Event 1	Simulated Event 2	Simulated Event 3
1	54.0	39.0	38
2	51.0	37.0	40
3	51.0	32.0	38
4	46.0	35.0	44
5	47.0	34.0	40
6	40.0	34.0	48
7	48.0	29.0	40
8	47.0	35.0	45
9	46.0	39.0	*NR
10	45.0	38.0	46
11	47.0	38.0	44
12	49.0	33.0	47
13	52.0	39.0	45
14	52.0	36.0	45
15	50.0	34.0	45
16	45.7	35.0	36
17	50.8	35.0	35
18	*NR	34.0	38
19	46.0	29.0	46
20	47.0	35.0	40
21	48.5	39.0	42
22	48.0	38.0	40
23	46.0	38.0	46
24	47.0	39.0	50
25	49.0	41.0	49
Average	48.0	35.7	43.0
Uniformity Coefficient	95.3	93.0	91.4

*Not Reported

Table C2 Raingage readings* recorded between simulated rainfall events in mm

Date	Rainfall
5/14/2003	0.9
5/19/2003	75.0
5/23/2003	15.0
5/28/2003	20.0
6/2/2003	25.0
6/10/2003	25.0
6/17/03	100.0

*The readings were taken after major rainfall events, but may include the rainfall from some small events in between spread over few days. The values do not correspond to any single rainfall event in between.

Appendix D Runoff Data

Table D1 Time to initiate runoff from each treatment plot

Plot	Treatment	Simulated Event 1	Simulated Event 2	Simulated Event 3
A	DI	0:20:40	0:01:40	0:03:30
B	PL	0:20:30	0:01:41	0:03:30
C	CO	0:20:20	0:01:42	0:03:15
D	IF	0:21:50	0:01:39	0:03:10
E	DS	0:20:00	0:01:39	0:03:06
F	PL	0:14:55	0:02:50	0:03:06
G	DI	0:23:35	0:05:25	0:03:30
H	IF	0:17:06	0:03:10	0:05:30
I	DS	0:25:30	0:10:40	0:11:30
J	CO	0:15:30	0:03:10	0:01:40
K	DS	0:22:30	0:05:00	0:08:30
L	IF	0:24:30	0:07:10	0:10:20
M	CO	0:19:00	0:04:38	0:07:20
N	DI	0:23:00	0:03:00	0:10:50
O	PL	0:23:00	0:04:40	0:07:45

Table D2 Peak runoff rate (mm/h) from each plot

Plot	Treatment	Simulated Event 1	Simulated Event 2	Simulated Event 3
A	DI	21.75	36.69	32.23
B	PL	33.83	38.32	23.79
C	CO	33.83	38.32	25.72
D	IF	34.06	38.32	29.99
E	DS	27.68	43.11	39.02
F	PL	27.68	38.19	26.53
G	DI	31.07	33.71	32.75
H	IF	38.06	40.58	23.79
I	DS	20.30	33.65	20.29
J	CO	21.75	29.23	27.59
K	DS	20.30	38.32	26.18
L	IF	43.50	40.10	26.19
M	CO	30.45	31.35	23.76
N	DI	43.50	48.41	24.53
O	PL	24.36	34.93	11.82

Table D3 Total runoff volume (mm) from each plot

Plot	Treatment	Simulated Event 1	Simulated Event 2	Simulated Event 3
A	DI	5.35	13.87	12.41
B	PL	7.71	12.16	7.25
C	CO	7.94	11.94	9.73
D	IF	11.70	14.68	9.52
E	DS	6.61	8.78	9.57
F	PL	12.00	12.44	7.22
G	DI	8.89	11.04	11.40
H	IF	13.57	14.62	5.51
I	DS	2.85	7.29	4.74
J	CO	4.22	7.50	6.75
K	DS	2.68	10.74	5.33
L	IF	10.67	13.39	8.32
M	CO	6.61	9.01	6.85
N	DI	5.25	10.85	3.11
O	PL	3.97	11.32	1.76

Table D4 Rate of runoff (mm/h) recorded at the time of sampling from each plot for the first simulated event

Time	A (DI)	B (PL)	C (CO)	D (IF)	E (DS)	F (PL)	G (DI)	H (IF)	I (DS)	J (CO)	K (DS)	L (IF)	M (CO)	N (DI)	O (PL)
13:40:00															
13:43:00															
13:46:00															
13:49:00															
13:52:00															
13:55:00															
13:58:00						0.81		1.69		1.34					
14:01:00	0.57		1.31		0.98	1.50		3.65		2.65			0.61		
14:04:00	0.55		9.49		1.41	2.90		10.87		2.80	1.19			1.47	1.25
14:07:00	0.76	2.54	15.17		1.69	5.71	0.88	21.00		3.34	1.39	9.61	1.35	1.65	
14:10:00	1.07	7.43	15.64	7.05	2.11	7.61	1.28	25.37	1.46	4.61	1.55	14.05		1.88	1.47
14:13:00	1.18	19.03	12.18	20.85	2.60	11.28	3.54	33.83	2.00	6.34	2.31	19.03	1.52	2.21	2.12
14:16:00	1.18	23.42	15.22	24.01	3.54	16.02	6.48	33.83	2.14	7.25	2.54	26.48	3.58	12.18	2.57
14:22:00	10.15	26.48	21.75	28.53	6.48	21.75	16.28	33.83	8.01	9.82	2.74	30.45	15.22	20.30	9.37
14:28:00	16.02	29.56	33.83	34.80	20.30	25.37	21.75	38.06	16.91	17.91	8.70	38.06	20.30	27.68	16.24
14:34:00	17.91	30.45	31.23	31.81	25.37	27.68	30.45	33.83	20.30	17.91	16.02	38.06	23.42	33.83	16.24
14:40:00	21.75	33.83	33.83	34.06	27.68	27.68	31.07	38.06	20.30	21.75	20.30	43.50	30.45	43.50	24.36
14:42:00	6.09	11.89	9.51	10.43	11.71	10.57	13.24	15.53	8.46	6.34	7.43	17.91	8.46	13.84	8.12

Table D5 Rate of runoff (mm/h) recorded at the time of sampling from each plot for the second simulated event

Time	A (DI)	B (PL)	C (CO)	D (IF)	E (DS)	F (PL)	G (DI)	H (IF)	I (DS)	J (CO)	K (DS)	L (IF)	M (CO)	N (DI)	O (PL)
13:36:00															
13:40:00	1.09	1.19	1.06	5.75	1.72	3.45								1.79	
13:44:00	1.99	8.21	8.62	19.71	6.21	10.35	2.21	9.32		3.45	2.01	3.94	0.94	2.16	8.90
13:48:00	10.36	24.64	19.16	27.59	22.99	22.62	7.99	24.64	5.31	8.62	7.66	26.53	15.00	16.23	18.39
13:54:00	28.74	38.32	38.32	38.32	34.49	35.56	23.02	38.32	20.69	21.56	31.35	33.16	24.64	36.07	29.99
14:00:00	31.35	38.32	34.49	38.32	38.32	33.16	27.77	36.30	27.59	24.99	31.35	38.32	26.53	37.80	32.85
14:06:00	34.49	38.32	34.49	43.11	43.11	34.77	32.45	44.79	30.66	27.37	38.32	38.32	26.53	42.45	34.93
14:12:00	36.69	38.32	38.32	38.32	43.11	35.41	31.73	40.58	29.35	27.59	38.32	40.10	31.35	48.41	35.37
14:21:00	36.69	38.32	38.32	38.32	43.11	38.19	33.71	40.58	33.65	29.23	38.32	39.64	28.74	41.18	34.93
14:23:00	18.85	15.00	18.15	18.15	20.29	29.48	17.55	16.19	12.54	8.62	13.27	15.33	9.20	12.54	13.33

Table D6 Rate of runoff (mm/h) recorded at the time of sampling from each plot for the third simulated event

Time	A (DI)	B (PL)	C (CO)	D (IF)	E (DS)	F (PL)	G (DI)	H (IF)	I (DS)	J (CO)	K (DS)	L (IF)	M (CO)	N (DI)	O (PL)
10:20:00															
10:23:00															
10:26:00	18.30	7.34	10.30	7.84	10.67	4.31	11.50	4.06		4.76					
10:29:00	22.32	11.13	17.92	14.08	20.25	8.57	13.01	10.61		10.14	3.72		1.21		1.06
10:32:00	24.81	15.33	19.49	21.11	24.00	12.32	15.80	10.89	5.75	13.80	8.57	3.28	2.76	3.59	0.99
10:35:00	26.74	15.33	14.92	17.17	24.25	13.80	22.06	13.27	10.35	18.15	11.71	6.44	8.26	5.75	0.96
10:38:00	25.72	16.99	20.42	22.00	30.63	16.99	24.81	14.37	12.26	17.24	18.01	8.62	8.41	7.64	1.47
10:44:00	31.02	21.03	21.15	22.53	33.58	20.29	26.11	18.15	16.42	21.56	24.46	14.80	15.22	13.78	3.58
10:50:00	32.02	18.95	24.65	26.03	35.27	21.56	29.13	20.29	16.74	23.79	22.34	17.17	17.69	15.58	5.17
10:56:00	30.25	20.29	23.74	28.02	36.65	22.99	29.99	20.90	20.05	25.17	23.58	19.45	20.09	18.87	6.57
11:02:00	32.23	21.16	24.03	26.23	35.93	23.46	31.07	22.25	20.29	25.55	25.41	23.92	22.99	20.86	8.28
11:08:00	30.99	22.84	25.72	27.59	39.02	26.53	32.75	21.56	20.29	27.59	26.18	23.10	22.31	24.53	9.85
11:11:00	32.23	23.79	23.79	29.99	29.99	26.53	27.61	23.79	20.29	27.37	25.48	26.19	23.76	23.38	11.82
11:13:00	12.76	0.00	13.14	9.38	14.86	8.62	13.30	10.00	10.30	9.58	11.04	11.65	21.56	7.21	1.89

Table D7 Volume to prepare flow weighted samples for each plot for first simulated rainfall event

Sample Number	A (DI)	B (PL)	C (CO)	D (IF)	E (DS)	F (PL)	G (DI)	H (IF)	I (DS)	J (CO)	K (DS)	L (IF)	M (CO)	N (DI)	O (PL)
1	6.98	6.93	1.45	9.29	20.82	10.12	7.02	0.22	9.26	2.80	10.23	27.25	23.22	0.67	10.62
2	0.53	28.82	2.34	19.34	9.90	6.38	6.13	2.64	3.97	6.46	7.05	33.43	7.50	0.48	8.48
3	0.65	55.34	4.33	36.18	9.13	14.04	12.53	13.52	7.54	10.61	10.72	47.44	9.13	0.38	10.11
4	1.54	70.97	13.19	56.43	15.24	24.16	25.19	30.47	29.34	13.11	12.24	103.49	15.24	2.49	11.77
5	3.26	80.87	29.48	106.04	34.61	33.64	83.69	47.02	102.96	17.32	27.43	166.40	34.61	50.86	38.39
6	24.07	132.58	83.28	157.81	102.55	47.19	164.79	60.84	206.42	25.80	145.20	196.03	102.55	143.34	106.61
7	130.20	194.01	165.81	185.41	182.86	99.67	228.60	109.78	275.86	57.17	355.79	216.63	182.86	223.21	194.98
8	250.55	211.93	224.04	206.41	224.75	164.25	268.05	161.33	197.22	112.70	342.01	142.59	224.75	272.14	268.32
9	305.72	162.47	248.74	138.34	213.32	201.10	156.50	176.72	167.41	189.27	98.90	66.75	213.32	185.30	201.09
10	201.62	54.58	159.81	84.75	100.93	217.04	47.50	186.72	-	260.90	8.41	-	100.93	121.13	149.61
11	74.87	1.47	67.54	-	85.89	127.31	-	122.77	-	189.26	-	-	85.89	-	-
12	-	-	-	-	-	55.12	-	87.95	-	114.60	-	-	-	-	-

Table D8 Volume to prepare flow weighted samples for the second simulated rainfall event

Sample Number	A (DI)	B (PL)	C (CO)	D (IF)	E (DS)	F (PL)	G (DI)	H (IF)	I (DS)	J (CO)	K (DS)	L (IF)	M (CO)	N (DI)	O (PL)
1	0.88	17.15	4.51	7.02	1.88	8.26	13.91	7.82	38.86	1.93	28.40	22.52	14.26	2.21	11.47
2	5.99	49.02	22.38	29.81	15.86	30.13	56.41	14.8	96.56	8.79	93.24	84.63	70.52	10.79	58.52
3	52.68	109.60	80.17	83.92	68.60	84.25	119.56	73.84	148.55	47.76	157.07	138.89	142.37	67.73	124.16
4	117.19	153.01	140.94	131.31	118.40	140.05	164.12	131.47	179.67	111.69	179.18	159.95	169.68	131.80	153.52
5	158.72	165.91	162.11	148.60	148.21	159.02	187.41	149.39	196.69	155.04	196.69	170.84	176.98	156.98	165.78
6	183.50	174.92	171.68	161.06	168.98	164.47	200.18	163.82	138.57	176.80	203.91	177.01	184.34	174.22	176.45
7	194.36	177.31	181.70	167.30	168.85	165.08	136.84	174.10	201.10	190.76	117.16	120.80	126.13	191.74	120.51
8	133.16	109.92	124.93	112.93	110.90	109.08	121.55	116.23	-	131.06	24.35	125.36	11.70	132.89	189.59
9	153.50	43.07	111.60	158.10	198.30	139.66	-	176.26	-	176.16	-	-	-	131.63	-

Table D9 Volume to prepare the flow weighted samples for the third simulated event

Sample Number	A (DI)	B (PL)	C (CO)	D (IF)	E (DS)	F (PL)	G (DI)	H (IF)	I (DS)	J (CO)	K (DS)	L (IF)	M (CO)	N (DI)	O (PL)
1	97.58	39.29	16.74	21.27	5.30	10.57	27.46	7.21	14.37	14.62	31.57	130.67	24.68	7.19	2.45
2	55.34	40.01	28.61	35.09	14.89	17.51	29.75	12.81	32.14	24.36	33.26	63.29	27.32	12.04	1.87
3	56.42	49.02	42.71	52.24	30.58	31.32	38.58	23.45	75.40	36.54	42.21	114.08	39.49	44.45	4.03
4	57.76	52.64	49.89	54.72	41.10	43.12	46.85	37.69	125.89	46.63	91.30	168.92	82.91	142.73	23.49
5	93.32	115.66	83.64	84.53	74.86	80.84	87.78	75.33	148.96	85.48	147.68	185.31	130.62	97.49	74.50
6	132.41	161.58	122.30	129.64	112.63	126.04	133.87	117.65	166.92	131.38	170.22	194.09	148.37	142.73	130.33
7	141.41	143.52	132.92	141.99	119.59	142.92	146.95	133.22	184.97	148.07	184.63	120.70	168.64	178.41	190.13
8	145.42	140.84	140.15	146.39	131.04	153.35	155.68	149.85	144.31	164.85	186.67	22.69	185.03	216.95	234.08
9	138.19	142.91	142.30	151.19	155.39	160.19	162.70	171.61	71.55	173.83	101.73	0.00	133.52	180.94	192.87
10	72.36	90.33	108.15	115.32	125.57	123.10	117.84	133.56	35.47	121.36	10.71	0.38	47.41	85.57	103.53
11	7.89	20.36	61.39	49.45	67.92	65.52	42.72	70.00	-	42.54	-	-	12.01	34.21	42.72
12	1.89	3.75	71.21	18.17	121.14	45.51	9.82	67.63	-	10.33	-	-	-	-	-

Appendix E Nutrient Analysis

Table E1 Nutrient analysis results from the flow weighted samples from each plot for the first simulated rainfall event

Plot	Treatment	TSS (g/l)	Nitrate (mg/l)	Ammonia (mg/l)	Total Nitrogen (mg/l)	Orthophosphorus (mg/l)	Bioavailable Phosphorus (mg/l)	Total Phosphorus (mg/l)
A	DI	4.38	1.60	2.50	21.50	0.26	1.46	11.00
B	PL	4.42	1.70	1.50	21.10	3.50	4.96	9.50
C	C	3.82	1.10	0.20	11.60	0.43	1.72	2.20
D	IF	4.21	1.31	0.76	17.93	0.40	1.73	2.45
E	DS	3.84	2.30	1.40	10.50	0.42	2.39	8.60
F	PL	4.55	1.40	1.50	24.90	4.20	4.26	8.30
G	DI	3.79	1.30	0.80	20.00	0.58	4.02	5.60
H	IF	3.86	1.10	1.10	14.10	0.72	2.59	3.70
I	DS	2.47	1.50	1.40	16.40	1.52	2.10	5.90
J	C	3.19	1.00	0.50	14.20	0.47	1.59	9.80
K	DS	2.66	1.30	0.70	13.50	0.93	2.23	3.20
L	IF	2.85	1.10	0.19	14.90	1.41	2.28	4.00
M	C	3.21	1.10	1.00	12.20	0.20	2.46	2.80
N	DI	3.28	1.10	0.40	16.30	0.26	2.74	7.70
O	PL	3.43	1.62	1.27	17.25	3.20	5.17	13.70

Table E2 Nutrient analysis results from the flow weighted samples from each plot for the second simulated rainfall event

Plot	Treatment	TSS (g/l)	Nitrate (mg/l)	Ammonia (mg/l)	Total Nitrogen (mg/l)	Orthophosphorus (mg/l)	Bioavailable Phosphorus (mg/l)	Total Phosphorus (mg/l)
A	DI	3.43	1.50	0.20	17.80	0.65	0.99	2.80
B	PL	3.32	1.30	1.30	22.70	2.12	3.22	5.70
C	C	2.10	1.20	0.00	10.40	0.49	0.70	1.70
D	IF	3.01	1.50	0.32	14.60	0.54	1.33	5.40
E	DS	3.46	2.00	0.60	24.10	0.92	1.06	4.00
F	PL	2.95	1.40	0.60	18.40	2.24	3.88	7.60
G	DI	3.09	1.30	0.60	21.20	0.27	0.65	1.10
H	IF	3.38	1.40	0.70	16.40	0.42	1.01	3.10
I	DS	2.47	1.90	1.80	15.30	1.13	1.85	5.40
J	C	3.17	1.20	0.08	12.70	0.20	0.74	2.30
K	DS	2.58	2.00	0.50	17.70	1.59	1.26	1.80
L	IF	2.95	1.60	0.20	13.70	0.39	0.40	0.60
M	C	2.72	1.30	0.20	13.60	0.05	0.52	1.18
N	DI	3.44	1.40	1.40	18.50	0.43	0.90	2.10
O	PL	2.25	1.00	2.90	13.80	1.85	1.97	2.70

Table E3 Nutrient analysis results from the flow weighted samples from each plot for the third simulated rainfall event

Plot	Treatment	TSS (g/l)	Nitrate (mg/l)	Ammonia (mg/l)	Total Nitrogen (mg/l)	Orthophosphorus (mg/l)	Bioavailable Phosphorus (mg/l)	Total Phosphorus (mg/l)
A	DI	0.30	1.00	0.10	3.00	0.40	0.48	1.43
B	PL	0.43	1.00	0.09	1.50	0.00	0.47	2.93
C	C	0.28	0.50	0.40	2.50	0.40	0.41	1.42
D	IF	0.22	1.80	0.10	1.90	0.20	0.48	1.25
E	DS	0.04	0.90	0.00	1.10	0.20	0.43	0.85
F	PL	0.31	1.10	0.05	2.50	0.20	0.60	1.94
G	DI	0.43	0.90	0.02	2.10	0.10	0.27	3.05
H	IF	0.07	1.40	0.40	2.80	0.21	0.38	0.93
I	DS	0.16	0.80	0.00	1.10	0.20	0.70	1.52
J	C	0.50	1.10	0.00	1.50	0.00	0.62	2.44
K	DS	0.37	1.10	0.20	1.90	0.23	0.99	2.29
L	IF	0.12	0.90	0.20	1.60	0.10	0.37	1.54
M	C	0.08	0.90	0.10	1.60	0.35	0.36	0.85
N	DI	0.10	0.50	0.03	3.10	0.23	0.31	1.38
O	PL	0.06	0.70	0.30	1.00	0.40	0.56	1.44

Table E4 Nutrient analysis results of the raw water used in each simulated event

Simulated Event	TSS (g/l)	Nitrate (mg/l)	Ammonia (mg/l)	Total Nitrogen (mg/l)	Orthophosphorus (mg/l)	Bioavailable Phosphorus (mg/l)	Total Phosphorus (mg/l)
First	0.00	N/A	0.07	N/A	0.15	0.22	0.43
Second	0.00	0.9	0.04	1.3	0.12	0.17	0.31
Third	0.00	0.9	0.40	1.3	0.00	0.06	0.63

Appendix F Bacterial Analysis Results

Table F1 Bacterial analysis results from each plot for the first simulated event

Plot	Treatment	E.Coli (cfu /100ml)	Enterococcus (cfu /100ml)	Fecal Coliform (cfu /100ml)
A	DI	2150.00	15000.00	95000.00
B	PL	500.00	285000.00	350000.00
C	CO	350.00	1500.00	40000.00
D	IF	-	-	
E	DS	7500.00	2355000.00	420000.00
F	PL	3350.00	268500.00	1735000.00
G	DI	4350.00	56500.00	1000000.00
H	IF	-		
I	DS	11250.00	6000.00	40000.00
J	CO	3500.00	7500.00	12800.00
K	DS	7000.00	3500.00	50000.00
L	IF	-		
M	CO	-	900.00	
N	DI	13000.00	53000.00	16000.00
O	PL	-	-	-

Table F2 Bacterial analysis results from each plot for the second simulated event

Plot	Treatment	E.Coli (cfu /100ml)	Enterococcus (cfu /100ml)	Fecal Coliform (cfu /100ml)
A	DI	-	72500.00	370000.00
B	PL	10700.00	97000.00	136500.00
C	CO	100.00	8800.00	700.00
D	IF	0.00		
E	DS	-	160000.00	230000.00
F	PL	3200.00	75000.00	68000.00
G	DI	2400.00	24500.00	645000.00
H	IF			
I	DS	11350.00	71000.00	310000.00
J	CO	9700.00	12650.00	Missing
K	DS	4150.00	17500.00	315000.00
L	IF			
M	CO	1100.00	4200.00	900.00
N	DI	18500.00	32000.00	107000.00

Table F3 Bacterial analysis results from each plot for the third simulated event

Plot	Treatment	E.Coli (cfu /100ml)	Enterococcus (cfu /100ml)	Fecal Coliform (cfu /100ml)
A	DI	29.00	68.00	90.50
B	PL	0.00	64.50	93.50
C	CO	16.00	61.50	86.50
D	IF			
E	DS	0.00	181.50	51.50
F	PL	7.00	6.50	26.00
G	DI	0.00	27.00	23.00
H	IF			
I	DS	23.00	47.00	55.00
J	CO	40.00	38.50	77.50
K	DS	195.00	311.00	65.00
L	IF			
M	CO	13.00	25.50	66.50
N	DI	1.00	3.50	35.00
O	PL	0.00	41.50	139.50