

CHARACTERIZATION OF MANURE EXCRETION AND ENVIRONMENTAL  
IMPACTS OF NUTRIENT MANAGEMENT IN  
DAIRY PRODUCTION SYSTEMS

By

TAMILEE DAWN NENNICH

A dissertation submitted in partial fulfillment of  
the requirements for the degree of

DOCTOR OF PHILOSOPHY

WASHINGTON STATE UNIVERSITY  
Department of Animal Sciences

DECEMBER 2004

To the Faculty of Washington State University:

The members of the Committee appointed to examine the dissertation of  
TAMILEE DAWN NENNICH find it satisfactory and recommend that it be accepted.

---

Chair

---

---

---

---

## ACKNOWLEDGMENTS

My first and ultimate thanks go to my Heavenly Father for all of the wonderful blessings he has sent my way. I know that God has led me to this point in my life and it is only his grace that has brought me to where I am today.

My advisor, Dr. Joe Harrison has been one of the best mentors I could have hoped for. These past few years have been filled with educational and enjoyable experiences. Even some of my bad habits weren't too big of a challenge for you to change, even if you met a little resistance along the way. I am a better person today than when I arrived in Washington and I know that you are directly responsible for many of the positive changes. I only hope that my future endeavors will make you proud.

A special thanks goes to my graduate committee for their patience and assistance along the way. Dr. Ron Kincaid did a wonderful job as my substitute advisor during my time in Pullman. Your thoughts and ideas have contributed to my educational experiences. Dr. Shulin Chen brought me into the world of agriculture engineering. Though engineering may never be my specialty, you made it more enjoyable. Dr. Normand St-Pierre gets extra thanks for the many hours and extreme patience that was shown to me as I tried to gain an understanding of statistics. Your support and understanding from across the miles was much appreciated.

Deb, though your job title may be a research technician, you have filled the roles of mentor, assistant, student, and friend. You have patiently helped me along the way and my research projects would not have been possible if you had not been there.

My research would not have been possible without the donations of land and time made by the Mallonee family. The entire family freely provided a place to conduct research and the assistance and effort to make everything come together. I hope that your contributions will ultimately prove to be helpful to you in the end.

Parents are a blessing from God, and I can only say I am truly blessed to have had parents who care so much. Your unconditional love and support has been humbling. I am one of those lucky people that can say my parents are also my friends. I know that it has been difficult for both of you to have so many miles between us, but you have been extremely supportive and let me go where I needed to.

Leaving my “little” brother in Minnesota was hard, but I didn’t need to worry because God’s plans were to have you join us here in Washington. If my presence here were only to assist you along your path to greatness, than it would have been worth it all. You have brought a little bit of home to Washington as well as expanding my knowledge and challenging me to think about areas outside of my chosen field. I also wanted to thank you for your contributions to my research, not everyone would volunteer to assist their big sister with the field sampling portion of their graduate projects.

I have also appreciated the thoughts and prayers of my other family members from the Talento’s (Honesto, Myrna, Myronie, Sam, and Kai), Nennich’s (Terry, Carrie, T. J., and Alexis), and Krause’s (Erma Jean). Thanks go to A. J. and Terry and Carrie for opening their home and providing transportation to make my visits home much more enjoyable and less stressful.

To my many friends, both new and old, your presence has made life more enjoyable. Fellow graduate students have brought both inspiration and enjoyment to my graduate experiences. I have to thank my best friend, Charity, for both your friendship and contributions to my research projects. I appreciated the patient helpfulness you provided, even though it was your vacation time. And to Sue, my new friend that I can only describe as a gift from God.

Last, but definitely not least, my thanks go to Mark, my husband and best friend. I know that our time in Washington has been a time for you to put your dreams aside and to let me have mine, and I will forever be grateful. I hope that the experiences we have had over the past few years will just be the start of our life together and the many adventures we have left to come.

CHARACTERIZATION OF MANURE EXCRETION AND ENVIRONMENTAL  
IMPACTS OF NUTRIENT MANAGEMENT IN  
DAIRY PRODUCTION SYSTEMS

Abstract

By Tamilee Dawn Nennich, Ph.D.  
Washington State University  
December 2004

Chair: Joseph H. Harrison

Determining amounts of nutrients excreted in manure and the best management practices for handling that manure in an environmentally friendly manner is important knowledge for dairy producers. Actual amounts of manure and nutrients excreted are difficult to determine outside of labor-intensive research studies and is extremely difficult for commercial dairy operations. New operations need a method of estimating manure and nutrients that will be excreted from dairy cows. Prediction equations are a method of estimating amounts of manure excretion and nutrients present in manure. Datasets from several universities were combined to develop prediction equations for estimating total manure, total solids, nitrogen, phosphorus, and potassium excretion of lactating dairy cows, dry cows, and heifers. Previous excretion estimates and prediction equations were also evaluated using this dataset. Equations for prediction of urine, urinary N, and urinary mineral excretion were developed to determine specific factors affecting urinary excretion. Two studies were conducted to evaluate environmental affects of manure application in a grazing-based dairy production system. Dairy slurry

was surface applied to pastureland during winter months. The effect of two rates of slurry application on N utilization and dry matter yields of grass were evaluated. A second study evaluated the risk of transport of nutrients and fecal coliform bacteria to surface waters. Application of dairy slurry during winter months was found to have little environmental impact. Knowledge of amounts of manure and nutrient excretion from dairy cattle and best management practices for preventing environmental impacts are important aspects of whole-farm nutrient management.

## TABLE OF CONTENTS

	Page
ACKNOWLEDGMENTS.....	iii
ABSTRACT.....	vi
LIST OF TABLES.....	xii
LIST OF FIGURES.....	xiii
CHARACTERIZATION OF MANURE EXCRETION AND ENVIRONMENTAL IMPACTS OF NUTRIENT MANAGEMENT IN DAIRY PRODUCTION SYSTEMS.....	1
INTRODUCTION.....	1
REFERENCES.....	6
PREDICTION OF MANURE AND NUTRIENT EXCRETION FROM DAIRY CATTLE.....	8
ABSTRACT.....	9
INTRODUCTION.....	9
MATERIALS AND METHODS.....	11
Sample collection and analysis.....	14
Statistical analysis.....	15
RESULTS AND DISCUSSION.....	16
Lactating cows.....	16
Total manure excretion.....	16
Dry matter excretion.....	20
Nitrogen excretion.....	21
Phosphorus excretion.....	23



Potassium excretion.....	25
Dry cows.....	27
Heifers.....	29
Calves.....	31
CONCLUSIONS.....	34
ACKNOWLEDGMENTS.....	35
REFERENCES.....	35
PREDICTION AND EVALUATION OF URINE AND URINARY NITROGEN AND MINERAL EXCRETION FROM DAIRY CATTLE.....	48
ABSTRACT.....	49
INTRODUCTION.....	50
MATERIALS AND METHODS.....	52
Sample collection and analysis.....	54
Statistical analysis.....	55
RESULTS AND DISCUSSION.....	56
Urine excretion.....	56
Effect of minerals on urine excretion.....	58
Urinary nitrogen excretion.....	61
Evaluation of urinary N excretion equations.....	63
Urinary mineral excretion.....	65
CONCLUSIONS.....	67
REFERENCES.....	68

WINTER MANURE APPLICATION ON A GRAZING BASED DAIRY: 1.) EVALUATION OF NITROGEN USE IN A NATIVE PASTURE.....	78
ABSTRACT.....	79
INTRODUCTION.....	79
MATERIALS AND METHODS.....	82
Statistical analysis.....	86
RESULTS AND DISCUSSION.....	86
Forage yields.....	88
Efficiency of nitrogen uptake.....	92
Soil nitrate and ammonium.....	94
CONCLUSIONS.....	97
REFERENCES.....	98
WINTER MANURE APPLICATION ON A GRAZING BASED DAIRY: 2.) EVALUATION OF FECAL BACTERIA AND NUTRIENT TRANSPORT TO SURFACE WATER.....	109
ABSTRACT.....	110
INTRODUCTION.....	111
MATERIALS AND METHODS.....	114
Site location and sampling.....	114
Sample collection.....	115
December application.....	115
January application.....	117
Laboratory analysis.....	117
Nutrient analysis.....	118

Bacterial source tracking.....	118
RESULTS AND DISCUSSION.....	119
Slurry application and sampling.....	119
Rainfall.....	120
Fecal bacteria on soil.....	121
Bacteria in surface water.....	124
Bacterial source tracking.....	127
Nitrogen in surface water.....	128
Phosphorus in surface water.....	130
CONCLUSIONS.....	131
ACKNOWLEDGMENTS.....	132
REFERENCES.....	132
CHARACTERIZATION OF MANURE EXCRETION AND ENVIRONMENTAL IMPACTS OF NUTRIENT MANAGEMENT IN DAIRY PRODUCTION SYSTEMS.....	145
SUMMARY AND CONCLUSIONS.....	145
APPENDIX	
1. PREDICTION OF MANURE AND NUTRIENT EXCRETION FROM DAIRY CATTLE.....	151
2. PREDICTION AND EVALUATION OF URINARY NITROGEN AND MINERAL EXCRETION FROM DAIRY CATTLE...	172
3. WINTER MANURE APPLICATION ON A GRAZING BASED DAIRY: 1.) EVALUATION OF NITROGEN USE IN A NATIVE PASTURE.....	188
4. WINTER MANURE APPLICATION ON A GRAZING BASED DAIRY: 2.) EVALUATION OF FECAL BACTERIA AND NUTRIENT TRANSPORT TO SURFACE WATER.....	197

## LIST OF TABLES

PREDICTION OF MANURE AND NUTRIENT EXCRETION FROM DAIRY CATTLE.....	38
1. Experiments included in the datasets used for development of excretion equations.	
2. Animal and production characteristics for the datasets used for development of prediction equations.	
3. Definitions of variables used in the prediction equations.	
4. Estimated typical manure characteristics as excreted by dairy animals.	
5. Average excretion values for animals in the datasets.	
PREDICTION AND EVALUATION OF URINE AND URINARY NITROGEN AND MINERAL EXCRETION FROM DAIRY CATTLE.....	71
1. Experiments included in the datasets used for development of urine and urinary mineral excretion equations.	
2. Animal and production characteristics for the datasets used for development of prediction equations.	
3. Average excretion values for animals in the datasets.	
4. Previously published prediction equations evaluated.	
WINTER MANURE APPLICATION ON A GRAZING BASED DAIRY: 1.) EVALUATION OF NITROGEN USE IN A NATIVE PASTURE.....	101
1. Application rates and composition of dairy slurry applied to plot during 2002 and 2003.	
2. Total cumulative yields of dry matter and nitrogen for plots.	
3. Soil nitrate N and ammonia N for plots.	
WINTER MANURE APPLICATION ON A GRAZING BASED DAIRY: 2.) EVALUATION OF FECAL BACTERIA AND NUTRIENT TRANSPORT TO SURFACE WATER.....	135
1. Characteristics of slurry applied to grassland.	
2. Classification of Escherichia coli isolates taken from water or soil samples.	

## LIST OF FIGURES

### PREDICTION OF MANURE AND NUTRIENT EXCRETION FROM DAIRY CATTLE.....43

1. Relationship between dry matter intake (DMI) and trial adjusted manure excretion for lactating cows (554 cow-periods). The solid line is equal to manure excretion,  $\text{kg/d} = (\text{DMI, kg/d} \times 2.63) + 9.4$ , Residual SE = 7.1, Inter-study SE = 9.5.
2. Relationship between P intake and trial adjusted P excretion for lactating cows (66 cow-periods). The solid line is equal to P excretion,  $\text{g/d} = (\text{P intake, kg/d} \times 560.7) + 21.1$ , Residual SE = 9.7, Inter-study SE = 9.2.
3. Relationship between crude protein (CP) intake and trial adjusted N excretion for heifers (60 animal-periods). The solid line is equal to N excretion,  $\text{g/d} = (\text{CP intake, kg/d} \times 78.39) + 51.4$ , Residual SE = 10.8, Inter-study SE = 24.5.
4. Relationship between crude protein (CP) intake and trial adjusted N excretion for calves (46 animal-periods). The solid line is equal to N excretion,  $\text{g/d} = (\text{CP intake, k/d} \times 112.55)$ , Residual SE = 8.2.
5. Relationship between P intake and trial adjusted P excretion for calves (32 animal-periods). The solid line is equal to P excretion,  $\text{g/d} = (\text{P intake, kg/d} \times 622.03)$ , Residual SE = 1.4, Inter-study SE = 0.6.

### PREDICTION AND EVALUATION OF URINE AND URINARY NITROGEN AND MINERAL EXCRETION FROM DAIRY CATTLE.....75

1. Relationship between the dietary cation-anion difference  $[(\text{Na} + \text{K}) - (\text{S} + \text{Cl})]$  and trial adjusted urine excretion (kg/d) for lactating cows (MINERAL dataset,  $n = 115$ ). The solid line is equal to Urine excretion (kg/d) =  $(\text{DCAD, meq/kg} \times 0.337) + 13.2$ , Residual SE = 6.1, Inter-study SE = 3.8.
2. Relationship between the rumen degradable protein supply (g/d) as calculated by the 2001 NRC model and trial adjusted urinary N excretion (g/d) for lactating cows (LACT dataset,  $n = 372$ ). The solid line is equal to Urinary N excretion (g/d) =  $(\text{RDP supply, g/d} \times 0.0628) + 55.6$ , Residual SE = 42.8, Inter-study SE = 34.4.
3. Plot of residuals (observed – predicted) versus predicted values of urinary N excretion (g/d) from evaluation of the equation [Urinary N,  $\text{g/d} = (0.0259 \times \text{BW, kg} \times \text{MUN, mg/dl})]$  by Kauffman and St-Pierre, 2001. The equation was evaluated using a combination of the LACT and VALIDATE datasets. The solid line on the graph represents the equation  $y = 7.98(\pm 10.3) -$

0.531( $\pm$ 0.047)(X – 214). Evaluation of the equation did not result in a mean bias. The linear bias was significant (-0.532; P < 0.01).

WINTER MANURE APPLICATION ON A GRAZING BASED DAIRY: 1.)  
EVALUATION OF NITROGEN USE IN A NATIVE PASTURE.....104

1. Crude protein concentration of harvested foraged from each clipping taken throughout the growing season. One cutting of grass was taken from each plot during the growing season (30 May 2002 and 23 May 2003).
2. Cumulative forage nitrogen yields for 2002 for plots receiving no slurry (control), 1x, or 2x slurry application rates. Slurry was applied to 1x and 2x plots on 15 Jan. 2002 and on 7 June 2002. The plots were harvested for hay on 30 May 2002.
3. Cumulative forage nitrogen yields for 2003 for plots receiving no slurry (control), 1x, or 2x slurry application rates. Slurry was applied to the 1x and 2x plots on 10 Jan. 2003 and on 11 June 2003. The plots were harvested for hay on 23 May 2003.
4. Cumulative removal of nitrogen in forage compared to the amount of nitrogen in dairy slurry applied to the plots in 2002 and 2003. The solid lines indicate the removal of N compared to the total amount of nitrogen in the applied slurry. The dashed lines indicate removal of N compared to nitrogen applications assuming 20% loss of NH<sub>4</sub>-N after the winter slurry applications and 80% loss of NH<sub>4</sub>-N after the summer slurry applications. Applications of dairy slurry, indicated on the graph with a \*, occurred on 15 Jan. 2002, 7 June 2002, 10 Jan. 2003, and 11 June 2003.
5. Average soil NH<sub>4</sub>-N concentrations during 2002 and 2003 for the control, 1x, and 2x plots. Average soil temperatures during the year are indicated on the graph with the (●).

WINTER MANURE APPLICATION ON A GRAZING BASED DAIRY: 2.)  
EVALUATION OF FECAL BACTERIA AND NUTRIENT TRANSPORT TO  
SURFACE WATER.....137

1. Location of manure application area, water sampling sites, and soil plots. The slurry applications took place on 12 Dec. 2003 and 27 Jan. 2004. Site A was located 10.6 m from the point of slurry application and site B was located 216 m further downstream. Sites C and D (287 m downstream from site B) were located at the property boundary. Site C included a waterway draining the western half of the property and site D included drainage water from the entire property and wooded areas to the east of the farm property. Sampling sites A, C, and D were sampled throughout the January sample collection period. In addition, sites E and F were sampled to monitor runoff from the January

application area. Site F was located upstream from the slurry application area and was 152 m from site E. Site E was located 19.8 m from the point of slurry application and was 111 m from site A.

2. Cumulative rainfall recorded with a weather station located at the study location. The December slurry was applied on December 12, 2003 and the January slurry application took place on January 27, 2004.
3. Fecal coliform bacteria concentrations in the soil from plots X, Y, and Z before and after slurry application on December 12, 2003. Plots X and Y were located in the slurry application area and plot Z was located in the grass setback zone between the application area and the waterway.
4. Fecal coliform bacteria concentrations in the soil from plots M, N, and O before and after slurry application on January 27, 2004. Plots M and N were located in the slurry application area and plot O was located in the grass setback zone between the application area and the waterway.
5. Fecal coliform bacteria concentrations at sites A and B before and after slurry application on December 12, 2003. The concentration of fecal coliform bacteria was greater than 5000 CFU 100 ml<sup>-1</sup> at site A on d 2 after slurry application.
6. Fecal coliform bacteria concentrations at sites E, F, and A before and after slurry application on January 27, 2004.
7. Fecal coliform bacteria concentrations at sites C and D before and after slurry applications on December 12, 2003 and January 27, 2004.
8. Concentrations of ortho-P and total P in water samples collected at sites A and B before and after manure application. Manure was surface applied to pastureland on December 12, 2003.

# **Characterization of Manure Excretion and Environmental Impacts of Nutrient Management in Dairy Production Systems**

**T. D. Nennich**

## **INTRODUCTION**

Nutrient management on dairy operations is emphasized more as environmental laws and regulations are passed at both the National and State levels to protect the quality of groundwater and surface water in the United States. In April 2003, new federal legislation from the U. S. Environmental Protection Agency (EPA) for Concentrated Animal Feeding Operations went into effect (USEPA, 2004). The EPA is a federal agency with a mission to protect the waters of the United States. As part of this mission, the EPA passes national legislation, such as the Concentrated Animal Feeding Operation Rule (USEPA, 2004), to help protect water quality. Although federal legislation is in place, individual states have the responsibility to pass additional legislation to protect water quality on a local basis. Government agencies, such as the Natural Resources Conservation Service (NRCS), assist with the mission of the EPA by establishing standards, such as the Nutrient Management 590 standard (NRCS, 2001). The goal of the 590 standard is to establish best management practices (BMP) to protect groundwater and surface water.



In the state of Washington, all dairy operations were required to have a nutrient management plan (NMP) by July 1, 2002, and the NMP was to be certified as fully implemented by December 31, 2003 (Washington State Legislature, 1998). The NMPs are supposed to outline the best management practices (BMP) for dairy operations to follow to minimize movement of nutrients and bacteria into groundwater and surface water. Many of the BMP are established by the NRCS as general standards that apply across states or regions of the United States. Many management practices have not been sufficiently evaluated on their efficacy to prevent transport of nutrients and bacteria into the surrounding environment. In addition, manure handling facility design and development of NMPs have been hampered in recent years due to lack of data on manure and nutrient excretion from cows fed contemporary diets and producing at current levels of high milk production.

Nutrient management planning needs to start with accurate estimates of manure and nutrient excretion from dairy animals. Without accurate estimates for manure excretion, planning of storage facilities and determination of BMP for nutrient handling and use becomes very difficult. The American Society of Agriculture Engineers (ASAE) has published standards for estimation of manure excretion from various livestock species (ASAE, 2001). Although the ASAE standards were revised every few years, the values in the tables were usually not updated. The 2001 ASAE manure excretion values listed for dairy animals were based on cow data from the 1970s. In addition, tabular values in the 2001 ASAE standards included one column for all dairy animals and did not make any distinctions for dry cows, heifers, or calves. In 1996,

Tomlinson et al. reported that the ASAE standards underestimated excretion from high producing cows. The ASAE standards have not been changed since Tomlinson's report.

The past several decades have brought many changes to the dairy industry, including a large increase in milk production per cow along with greater manure and nutrient excretion. According to the 2001 ASAE standards, a 650 kg cow excretes 56 kg manure, 292 g N, and 61 g P per d. Wilkerson et al. (1997) predicted that cow of a similar weight producing 40 kg of milk per day would excrete approximately 25% more manure and 35% more N than estimated by the 2001 ASAE standards. As the previous example demonstrated, basing manure and nutrient excretion on animal body weight was another challenge of the 2001 ASAE standards. Methods for estimating manure and nutrient excretion that use variables with greater physiological importance will provide better estimates of excretion, which is becoming more feasible as dairy operations improve measurement abilities and record keeping.

Excretion of total manure, N, and P are usually of the greatest concern in NMPs because these excretion factors directly affect storage and land application requirements. However, development of prediction equations for estimating excretion of urinary N and minerals provides additional information and opportunities for nutrient management. Recently, researchers (Jonker et al., 1998; Kauffman and St-Pierre, 2001; Kohn et al., 2002) have focused more specifically on urinary N excretion. The largest

dataset used to develop prediction equations for urinary N included 70 observations (Jonker et al., 1998).

There is also a paucity of data available on estimating excretion of other minerals, such as K, Na, and Ca, from dairy cows. Development of prediction equations for other minerals provides a resource for nutrient management planners when additional mineral excretion information is desired.

After a better estimation of manure and nutrient excretion has been determined, there are various methods by which the manure and nutrients can be handled at the farm level. Many of the best management practices that are either recommended or discouraged have not been evaluated under controlled conditions and in a systems approach to determine whether or not the management practices increase the risk of nutrient and bacteria transport to the environment.

A management practice that is often discouraged is application of dairy slurry during winter months. In many areas, spreading slurry during winter months is discouraged because the application occurs on frozen soils. However, in locations where soils do not freeze, the effect of winter slurry application on bacteria and nutrient transport to the environment is unknown. Due to the mild winters in the Pacific Northwest, Sullivan et al. (2000) suggested that up to one-third of manure could be applied during the fall and winter months. Therefore, two studies were designed to

evaluate if application of dairy slurry on a grazing based dairy during winter months would lead to transport of nutrients to either groundwater or surface water.

A study was designed to evaluate N usage of winter applied dairy slurry in a pasture based dairy system. Nutrient leaching from grasslands is less likely because of the large quantity of N utilized by grass (Bittman et al., 1999). Nevertheless, applied nutrients must be available to the crop during a time when the crop nutrient uptake is taking place. Due to the growing conditions in the Pacific Northwest, grasses have a longer growing season and there are more opportunities for manure application throughout the year (Sullivan et al., 2000). However, N can be leached through the soil profile and out of the root zone during the high rainfall periods of the winter months. On-farm studies provide opportunities to evaluate the recovery of N when manure is applied to fields during the winter months.

Very few on-farm studies have been conducted to look at risks of nutrient and bacterial transport to surface water. A study by Frantz et al. (2003) was one of the few on-farm studies conducted to evaluate the potential movement of fecal bacteria into surface waters after application of dairy slurry. The Frantz et al. (2003) study evaluated movement of bacteria when dairy slurry was applied almost on a daily basis for over a year. Quantification of the movement of fecal coliform bacteria into surface waters is important because of state standards for bacteria levels in surface water. In the 2003 revised water quality standards, the Washington State Department of Ecology (WSDOE, 2003) established that fecal coliform was to be used as the criteria for fecal

contamination of surface waters. The numerical criteria for secondary use recreational waters in Washington State are 200 colony forming units/100 mL of water (WSDOE, 2003).

Improved methods for predicting manure excretion will provide dairy operations and nutrient management planners with better estimates of manure and nutrients that need to be managed at the farm level. In addition, controlled evaluation of various nutrient management practices provide a sound basis from which to determine the degree of environmental risks associated with these particular management practices.

## REFERENCES

- American Society of Agriculture Engineers. 2001. Manure production and characteristics. ASAE Standards D384.1. ASAE, St. Joseph, MI.
- Bittman, S., C. G. Kowalenko, D. E. Hunt, and O. Schmidt. 1999. Surface-banded and broadcast dairy manure effects on tall fescue yield and nitrogen uptake. *Agron. J.* 91:826-833.
- Frantz, E., K. Griswold, G. Apgar, B. Jacobson, and J. Haddock. 2003. Effectiveness of vegetative filter strips (VFS) for controlling pathogen loads and antibiotic resistance in dairy wastewater. Pages 67-73 in *Proc. 9<sup>th</sup> Int. Symp. of Anim., Agric., and Food Processing Wastes*, Research Triangle Park, NC.
- Jonker, J. S., R. A. Kohn, and R. A. Erdman. 1998. Using milk urea nitrogen to predict nitrogen excretion and utilization efficiency in lactating dairy cows. *J. Dairy Sci.* 81:2681-2692.
- Kauffman, A. J., and N. R. St-Pierre. 2001. The relationship of milk urea nitrogen to urine nitrogen excretion in Holstein and Jersey cows. *J. Dairy Sci.* 84:2284-2294.
- Kohn, R. A., K. F. Kalscheur, and E. Russek-Cohen. 2002. Evaluation of models to estimate urinary nitrogen and expected milk urea nitrogen. *J. Dairy Sci.* 85:227-233.

Natural Resources Conservation Service. 2001. Nutrient management. Natural Resources Conservation Service Conservation Practice Standard Code 590-1. USDA-NRCS. Washington State.

Sullivan, D. M., C. G. Cogger, A. I. Barry, and S. C. Fransen. 2000. Timing of dairy manure applications to perennial grass on well drained and poorly drained soils. *J. Soil and Water Conservation* 55:147:152.

Tomlinson, A. P., W. J. Powers, H. H. Van Horn, R. A. Nordstedt, and C. J. Wilcox. 1996. Dietary protein effects on nitrogen excretion and manure characteristics of lactating cows. *Trans. ASAE* 39:1441-1448.

United States Environmental Protection Agency (USEPA). 2004. Concentrated animal feeding operations – Final rule. Online. Available: <http://cfpub.epa.gov/npdes/afo/cafofinalrule.cfm>.

Washington State Department of Ecology (WSDOE). 2003. Water quality standards for surface waters of the state of Washington. Chapter 173-201A WAC. Online. Available: <http://www.ecy.wa.gov/pubs/wac173201a.pdf>.

Washington State Legislature. 1998. Dairy Nutrient Management. Chapter 90.64 RCW. Online. Available: <http://www.leg.wa.gov/RCW/index.cfm?fuseaction=chapterdigest&chapter=90.64>.

Wilkerson, V. A., D. R. Mertens, and D. P. Casper. 1997. Prediction of excretion of manure and nitrogen by Holstein dairy cattle. *J. Dairy Sci.* 80:3193-3204.

## **Prediction of Manure and Nutrient Excretion from Dairy Cattle**

**T. D. Nennich<sup>\*</sup>, J. H. Harrison<sup>\*</sup>, L. M. VanWieringen<sup>\*</sup>,  
D. Meyer<sup>†</sup>, A. J. Heinrichs<sup>‡</sup>, W. P. Weiss<sup>§</sup>,  
N. R. St-Pierre<sup>||</sup>, R. L. Kincaid<sup>#</sup>, D. L. Davidson<sup>\*</sup>, and E. Block<sup>£</sup>**

<sup>\*</sup>Puyallup Research and Extension Center,  
Washington State University, Puyallup 98371

<sup>†</sup>Department of Animal Sciences,  
University of California, Davis 95616

<sup>‡</sup>Department of Dairy and Animal Science,  
The Pennsylvania State University, University Park 16802

<sup>§</sup>Department of Animal Sciences,  
Ohio Agriculture Research and Development Center,  
The Ohio State University, Wooster 44691

<sup>||</sup>Department of Animal Sciences,  
The Ohio State University, Columbus 43210

<sup>#</sup>Department of Animal Sciences,  
Washington State University, Pullman 99163

<sup>£</sup>Church & Dwight Co. Inc.,  
Princeton, NJ 08543

Corresponding author:

Joseph Harrison  
7612 Pioneer Way E.  
Puyallup, WA 98371  
253/445-4638  
jhharrison@wsu.edu

## ABSTRACT

Accurate estimates of manure excretion are needed for planning of manure storage facilities and for nutrient management. Datasets from metabolism studies conducted at several universities were compiled and evaluated for excretion of total manure, N, P, and K. Animal groups included calves weighing up to 204 kg, heifers weighing between 274 to 613 kg, non-lactating cows, and lactating cows. Regression equations were developed to predict excretion of total manure, total dry matter, N, P, and K. Predictors used in the regression equations for lactating cows included milk yield, percentages of protein and fat in milk, dietary concentrations of crude protein and neutral detergent fiber, and intakes of nutrients. The regression equations provide improved predictions of excretion and enable more accurate planning of manure storage and nutrients to be managed at the farm level.

**Key words:** manure, nutrient excretion, nitrogen, phosphorus, potassium

**Abbreviation key:** **ASAE** = American Society of Agricultural Engineers, **MILK** = milk yield, **MF** = milk fat percent, **MTP** = milk protein percent, **MilkP** = P in milk, **DMD** = DM digestibility, **C<sub>CP</sub>** = dietary CP concentration, **C<sub>NDF</sub>** = dietary neutral detergent fiber concentration, **C<sub>P</sub>** = dietary P concentration, **C<sub>K</sub>** = dietary K concentration, **M<sub>E</sub>** = manure excretion, **DM<sub>E</sub>** = manure DM excretion, **N<sub>E</sub>** = N excretion, **P<sub>E</sub>** = P excretion, **K<sub>E</sub>** = K excretion.

## INTRODUCTION



Manure excretion estimates are important for designing storage facilities and developing nutrient management plans. As production and management of dairy cattle have changed in recent decades, changes also have occurred in manure and nutrient excretion. Increased feed intake and milk production of dairy cows have led to greater excretion of nutrients, and must be considered during the planning of modern dairy operations. According to the Natural Resources Conservation Service standard for nutrient management planning (NRCS, 2001), nutrient budgets should be established for N, P, and K. Recent modifications to the Clean Water Act have resulted in new regulations related to nutrient management on concentrated animal feeding operations (Environmental Protection Agency, 2003). Estimates of manure nutrients excreted are now being incorporated into nutrient management plans, necessitating greater accuracy in the estimates used. In addition, estimates of manure DM excretion are useful for designing anaerobic digesters and providing information for implementation of current, or development of future, manure treatment technologies.

Estimates for manure and nutrient excretion by dairy cattle are found in ASAE Standard D384.1 (ASAE, 2001). These estimates, based on data from the late 1960s and early 1970s, were taken from a maximum number of 85 data points. The ASAE standard was revised in 1988 to merge a dairy heifer column with a dairy cow column for one single column for all dairy cattle categories.

Prediction of manure excretion from dairy cattle was reviewed by Tomlinson et al. (1996) and Wilkerson et al. (1997). Each of these authors compiled excretion data

from lactating Holstein dairy cows producing an average of 20.3 and 29 kg of milk/d (Tomlinson et al., 1996; Wilkerson et al., 1997). However, many dairy cattle are currently producing milk at twice those levels. New manure and nutrient excretion estimates need to be developed to predict excretion from higher producing cows. Updated information is critical to owners of animals residing in environmentally sensitive areas. In addition, excretion estimates for calves and heifers have not been published in recent years and estimates of manure excretion from these animals are limited and essential to replacement heifer operations. Technical assistance providers, dairy operators, and staff from regulatory agencies are seeking site-specific information on manure volume and nutrient content to more precisely develop nutrient management plans.

Recently, a committee was developed by ASAE Structures and Environment Committee 412 and members from the Federation of Animal Science Societies to revise the ASAE manure excretion values using data from contemporary diets and levels of productivity. Therefore, the objective of our study was to develop regression equations to predict the manure, DM, and nutrient excretion of calves, heifers, non-lactating, and lactating Holstein dairy cows in an effort to revise the ASAE dairy manure excretion estimates.

## **MATERIALS AND METHODS**

Datasets from Washington State University, The Ohio State University, The Pennsylvania State University, and the University of California, Davis were combined and used for estimations of excretion from dairy cattle (Table 1). The overall dataset included records from a wide variety of animal ages, ranging from calves to multiparous lactating cows. Data were categorized into four groups: lactating cows (**LACT**), dry cows (**DRY**), heifers (**HEIFER**), and calves (**CALF**). The LACT dataset (554 cows or cow-periods from Latin square experiments) included multiparous lactating Holstein cows. DRY dataset (18 cow-periods) animals were defined as multiparous, non-lactating cows that were pregnant. The HEIFER dataset (60 animal-periods) included female, non-lactating animals that had not calved and included animals of various ages weighing > 250 kg. The CALF dataset (46 animal-periods) included animals weighing < 250 kg (Table 2).

The LACT dataset included observations from 26 individual feeding studies. Feeding studies were originally intended to evaluate nutritional hypothesis and were not designed specifically for the development of excretion prediction equations. Variation in animal location and dietary ingredients increased the errors associated with development of regression equations, but provided a broader base to account for differences in the commercial industry. An effort was made to include several equations for each dependent variable during the development of prediction equations to provide users flexibility depending on the accuracy and availability of input variables for a given dairy operation.

The independent variables included in the LACT dataset were BW, DIM, DMI, DM digestibility (**DMD**), milk yield (**MILK**), percent milk fat (**MF**), and percent milk protein (Table 3). Milk CP values in the dataset were converted to milk true protein (**MTP**) values using a conversion factor of 0.940. Dietary ingredients and characteristics were used as additional prediction factors for equations, including dietary concentrations of CP (**C<sub>CP</sub>**), NDF (**C<sub>NDF</sub>**), P (**C<sub>P</sub>**), and K (**C<sub>K</sub>**). Dependent variables included in the LACT dataset were (Table 3) manure excretion (**M<sub>E</sub>**), DM excretion (**DM<sub>E</sub>**), N excretion (**N<sub>E</sub>**), P excretion (**P<sub>E</sub>**), and K excretion (**K<sub>E</sub>**). Dry matter excretion included both fecal and urinary DM and was determined by adding actual fecal DM and 4.5% of urinary excretion.

Quadratic models were evaluated for excretion variables using the LACT dataset. Variables evaluated in equations included squared and two-way interactions of DMI, MILK, DIM, BW, and C<sub>NDF</sub> and C<sub>CP</sub>.

A subset of the LACT dataset included information on P and K intake and excretion for lactating animals. The MINERAL dataset (85 cow-periods) included cows for which excretion of feces and urine were known. Intakes of minerals were determined through analyses of both feed and orts. One study of early lactation cows (Johnson et al., 1998, Experiment 2; 15 cow-periods ranging from 16 to 61 DIM) was not included in the MINERAL dataset due to negative P and K balances for the early lactation animals.

Diets fed during the metabolism trials included a wide variety of protein supplements and forage types. Forages included corn silage, grass silage, alfalfa silage, and grass hay. The remainder of the diets included various grains, by-product feeds, and mineral supplements. Cows in these trials were fed ad libitum.

Equations given for each parameter include residual standard error (SE) and inter-study SE. Equations with lower SE are expected to provide a more precise estimation of excretion and should be used when values for the input variables are available.

### **Sample Collection and Analysis**

Total collection metabolism studies (16 studies) conducted at Washington State University included both lactating cows (399 cow-periods) and dry cows (7 cow-periods). Feeding, sample collection, and sample analyses were conducted by methods outlined by Johnson et al. (1998) and Timmermans et al. (2000). Metabolism studies from the University of California, Davis included 3 calf studies, 3 heifer studies, 2 dry cow studies, and 4 lactating cow studies. Feeding and sample collections were described by James et al. (1999) and Meyer et al. (2000). Methods used for collection during the metabolism studies (6 studies; 139 cows or cow-periods) with lactating multiparous Holstein cows at The Ohio State University were reviewed by Weiss and Wyatt (2004). Studies from The Pennsylvania State University that included 32

observations from weaned calves and 32 observations from heifers are summarized by Gabler and Heinrichs (2003a and 2003b).

Mineral analyses were not conducted on milk samples. Therefore, milk mineral contents were assumed to be equivalent to values outlined in the 2001 Dairy NRC. Milk P and K were estimated at 0.9 g/kg and 1.5 g/kg of milk, respectively (Dairy NRC, 2001). Minerals in feces were analyzed by the University of Nebraska Soil and Plant Analytical Lab, Lincoln, NE and urine minerals were analyzed by Dairyland Laboratories, Arcadia, WI.

### **Statistical Analysis**

Regression analyses were performed using PROC MIXED of SAS (SAS, 1999) with the discrete effect of study included as a random variable (St-Pierre, 2001). Equations were developed by running multiple iterations in MIXED and removing the least significant main effect each time based on Type III sums of squares. For datasets equal to or greater than 200 observations, variables were kept if  $P < 0.10$ . For datasets with less than 200 observations, variables were kept if  $P < 0.25$ . Adjusted observations were calculated for graphing purposes by adding the residual from each individual observation to the predicted value of the study regression (St-Pierre, 2001).

Equation evaluation was done by regressing residuals (predicted values subtracted from observed values) on the predicted values (St-Pierre, 2003). Predicted

values were centered by subtracting the mean of all predicted values from each prediction. This makes the slope and intercept estimates in the regression orthogonal and, thus, independent. Mean biases were assessed using the intercepts of the regression equations and the slopes of the regression equations were used to determine the presence of linear biases.

## **RESULTS AND DISCUSSION**

### **Lactating Cows**

*Total manure excretion.* Equations for  $M_E$  were developed using the LACT dataset (554 cow-periods). Manure excretion for cows in this dataset averaged 66.3 kg/d for cows weighing an average of 630 kg with a DMI of 21.7 kg/d. Wilkerson et al. (1997) reported cows with an average milk yield of 29 kg/d excreted 89 kg of manure/d per 1000 kg of BW, which is equivalent to 56.1 kg of  $M_E$ /d from a 630 kg cow averaging 17.9 kg of DMI/d. Average  $M_E$  was 8.2 kg greater for cows in the LACT dataset than for cows in the study by Wilkerson et al. (1997). Frank et al. (2002) reported even less  $M_E$ , ranging from 44.9 to 49.1 kg/d for cows consuming between 18.3 and 20.2 kg DM/d. Other researchers have indicated that the 2001 ASAE manure excretion estimates underestimate excretion from high producing cows (Tomlinson et al., 1996) and that using only BW is not a method of accurately predicting excretion (James et al., 1999).

The ASAE (2001) tables listed excretion values on a basis of 1000 kg of BW. The units of 1000 kg of BW were removed to reduce the confusion often associated with placing animals on an equal BW basis. Newly proposed table values (Table 4) provide excretion estimates on a per animal basis and include descriptions of animals used to develop the value. Table values, common in previous versions of ASAE standards (2001), were retained to provide an average value for predicting manure excretion. In addition, prediction equations were developed for each of the excretion parameters if enough data were present. The goal of including the prediction equations was to provide excretion estimates that are adaptable to particular operations in lieu of a general table value that is used by all dairy operations, regardless of the production level or size of the animals.

A simple equation with MILK as the only input variable was developed to provide a practical equation for use when other animal factors are not known. An equation using MILK as the only variable was included to provide nutrient management planners with a more precise alternative to the use of a the general table value.

$$M_E = (\text{MILK} \times 0.616 (\pm 0.057)) + 46.2 (\pm 2.3) \quad [1]$$

$$\text{Residual SE} = 10.0, \text{ Inter-study SE} = 7.1$$

Equation [1] has the greatest residual SE and is less precise than equations with different variables and lower residual SE. The simple equation with MILK provides flexibility not previously given in the ASAE standards. The ASAE (2001) value for  $M_E$



was listed as 86 kg/d per 1000 kg of BW (54.2 kg/d for a 630 kg cow). Using [1],  $M_E$  would average 58.5 and 70.8 kg/d for cows producing 15 and 40 kg/d, respectively. The lower  $M_E$  estimates given in the 2001 ASAE tables are a result of lower milk production and DMI of cows used to generate these values.

Inclusion of DMI in [2] provided a more precise estimation of  $M_E$  than equations that did not include DMI. The best single independent variable for predicting  $M_E$  in the LACT dataset was DMI [2].

$$M_E = (\text{DMI} \times 2.63 (\pm 0.10)) + 9.4 (\pm 2.8) \quad [2]$$

$$\text{Residual SE} = 7.1, \text{ Inter-study SE} = 9.5$$

Figure 1 shows the relationship between DMI and  $M_E$ . Including DMI as an independent variable improved the precision of the estimation by reducing the residual SE by 28.9% as compared to using MILK. Inclusion of DMI in the equations has become a more realistic option as producers have improved record-keeping skills and increased the use of scales.

Body weight was a significant predictor of  $M_E$ , and similar findings were reported in equations developed by Wilkerson et al. (1997). When comparisons were made between equations including either DMI or BW, equations with DMI improved the prediction across studies by 20% when predicting  $M_E$ . The less accurate relationship of BW to  $M_E$  in the prediction equations indicated that basing  $M_E$  on BW,

as was done in 2001 ASAE, is not the most accurate method of predicting  $M_E$  from lactating cows.

Dietary NDF concentration was evaluated in the LACT dataset to determine if it was a significant factor for predicting  $M_E$  as indicated by Wilkerson et al. (1997).

When  $C_{NDF}$  was evaluated in the LACT dataset, it was positively related to  $M_E$  regardless of the inclusion of DMI in the regression models. Crude protein intake was also positively related to  $M_E$ , indicating that the total amount of CP consumed affected  $M_E$ . Frank et al. (2002) also reported greater  $M_E$  as greater amounts of CP were fed.

Most of the non-linear models evaluated resulted in prediction equations that were less accurate predictors of  $M_E$  than linear equations. The best non-linear equation for describing the LACT dataset included several of the same independent variables given by Wilkerson et al. (1997) (BW, DMI, DIM,  $C_{CP}$ , and  $C_{NDF}$ ) and included the interaction of DMI and  $C_{CP}$  as well as squared terms of DIM and  $C_{NDF}$ . The non-linear equation improved the residual SE of [2] by only 1%. Because the non-linear equations only provided a very slight improvement over the linear equations, we suggest the use of the linear equations for predicting  $M_E$ .

In addition to the development of new prediction equations using the LACT dataset, previously published equations for predicting  $M_E$  were evaluated. Predictions using the equation of Wilkerson et al. (1997) [ $M_E = (0.0286 \times BW) + (0.0378 \times DIM) + (1.0689 \times MILK) + (9.67 \times C_{CP}) + (61.4 \times C_{NDF}) - 21.94$ ] resulted in significant mean

and linear biases ( $P < 0.01$ ) of 5.6 kg/d and  $-0.25$ , respectively. Although  $M_E$  in the LACT dataset was greater than was predicted using the Wilkerson et al. (1997) equation, the difference was less than the standard deviation of the residuals.

**Dry matter excretion.** The average fecal DM excretion was 7.3 kg/d and  $DM_E$  was 8.5 kg/d (Table 5). Tomlinson et al. (1996) reported fecal DM excretion ranging from 6.2 to 7.4 kg/d, values similar to those in the LACT dataset.

The best predictor of  $DM_E$  was DMI [3].

$$DM_E = (DMI \times 0.356 (\pm 0.011)) + 0.80 (\pm 0.34) \quad [3]$$

$$\text{Residual SE} = 0.78, \text{ Inter-study SE} = 1.11$$

Equation [3] indicated a direct relationship between DMI and  $DM_E$ . In 1994, Van Horn et al. reported that total solids excretion could be determined by multiplying DMI by 0.33 and adding the urine DM.

Other variables evaluated for predicting  $DM_E$  were BW, DIM, MILK, MF, and MTP, but these variables were not significant when included in the equation with DMI. Conversely,  $C_{NDF}$  was a significant variable when included in an equation with DMI, but resulted in a less precise prediction equation across studies than when DMI was the only independent variable for predicting  $DM_E$ .

Two equations were developed for predicting  $DM_E$  in the absence of DMI:

$$DM_E = (\text{MILK} \times 0.0874 (\pm 0.0070)) + 5.6 (\pm 0.30) \quad [4]$$

Residual SE = 1.21, Inter-study SE = 0.87

$$DM_E = (\text{MILK} \times 0.112 (\pm 0.008)) + (\text{BW} \times 0.0062 (\pm 0.00089)) + \\ (\text{MTP} \times 106.0 (\pm 18.8)) - 2.2 (\pm 0.95) \quad [5]$$

Residual SE = 1.15, Inter-study SE = 0.78

Equation [4] provides a prediction of  $DM_E$  based solely on MILK and [5] includes BW, MTP, and MILK. Predictions of  $DM_E$  using [4] and [5] are expected to be less accurate than predictions using [3], but provide estimates in instances where DMI is not known.

***Nitrogen excretion.*** The simple linear equation, using MILK as the only independent variable [6], indicated a significant relationship between  $N_E$  and MILK. When MILK was used as the only prediction variable, it resulted in a less precise prediction than subsequent equations evaluated, but using MILK as the only variable resulted in a 2.6% improvement in accuracy compared to use of BW to predict  $N_E$ .

$$N_E = (\text{MILK} \times 2.82 (\pm 0.42)) + 346 (\pm 18.1) \quad [6]$$

Residual SE = 70.9, Inter-study SE = 57.9

Equations were developed for situations where intake of CP is known. When evaluated in a simple equation, CP intake was significantly associated with  $N_E$  [7].

$$N_E = (\text{DMI} \times C_{CP} \times 84.1 (\pm 3.7)) + (\text{BW} \times 0.196 (\pm 0.026)) \quad [7]$$

$$\text{Residual SE} = 51.4, \text{ Inter-study SE} = 56.1$$

As expected, an increase in CP consumption resulted in greater  $N_E$ . The most precise equation developed for predicting  $N_E$  included CP intake as an independent variable [7]. Equation [7] improved the residual SE by 27.5% compared to equation [6]. The direct relationship between N intake and  $N_E$  indicates that future improvements in balancing diets to better meet the specific amino acid needs of the animal while decreasing  $C_{CP}$  may be an important step in decreasing  $N_E$  (Harrison et al., 2002).

Nitrogen intake was directly related to  $N_E$  in previous experiments (Tomlinson et al., 1996; Frank et al., 2002; James et al., 1999; Krober et al., 2000). Excess intake N is mostly excreted via urinary excretion. Tomlinson et al. (1996) indicated that  $N_E$  was closely related to N intake and DMI and somewhat related to BW, whereas Van Horn et al. (1994) stated that  $N_E$  could be estimated by subtracting the N in milk from N intake.

Quadratic models were evaluated to determine if the predictions were improved with the addition of squared terms and interactions in the model. When quadratic models were evaluated, the resulting equations did not reduce the residual SE or inter-study SE compared to the linear models. Conversely, Wilkerson et al. (1997) reported

that development of quadratic models led to a statistical improvement over the linear models developed for predicting  $N_E$ . These authors, however, did not account for the imbalance of the predictor variables across studies (the random study effect in the model) and thus may have induced the apparent nonlinearity of the prediction.

Previously published equations for predicting  $N_E$  (Wilkerson et al., 1997) were evaluated using the LACT dataset. Evaluation of the linear equation published by Wilkerson et al. in 1997 [ $N_E = (0.000232 \times BW) + (0.000342 \times DIM) + (0.00649 \times MILK) + (1.83 \times C_{CP}) + (0.280 \times C_{NDF}) - 0.440$ ] resulted in a significant mean bias of 37.4 g/d ( $P < 0.01$ ) of excreted N and a significant linear bias of  $-0.264$  ( $P < 0.01$ ). The standard deviation of the residuals of the LACT dataset was 78.4 g/d, indicating that the mean bias, although significant, was less than the variation expected between studies.

***Phosphorus excretion.*** Dietary P concentrations in the MINERAL dataset averaged 0.0044 g/g DM. Many of the studies in the MINERAL dataset were conducted before reduced P feeding was emphasized in dairy diets, which resulted in  $C_P$  greater than needed to meet animal requirements. Because cows were fed diets with  $C_P$  greater than their requirements, equations developed with the MINERAL dataset may not accurately account for diets with P supply at or below animal requirements.

The 2001 ASAE estimate of  $P_E$  for a 630 kg cow was 59.2 g/d, 14.7 g/d less than the average  $P_E$  for cows in the MINERAL dataset (Table 5). In contrast, an average  $P_E$  of 57 g/d was reported by Weiss and Wyatt (2004), which was similar to the

2001 ASAE value. However, the P intake in the MINERAL dataset averaged 0.013 kg/d greater than the cows in the Weiss and Wyatt (2004) and would account for most of the difference in  $P_E$ .

The simple equation [8] developed using MILK as the only predictor of  $P_E$  indicated a positive relationship between MILK and  $P_E$ .

$$P_E = (\text{MILK} \times 0.781 (\pm 0.230)) + 50.4 (\pm 8.6) \quad [8]$$

$$\text{Residual SE} = 13.4, \text{ Inter-study SE} = 11.3$$

The positive relationship between MILK and  $P_E$  is most likely a result of greater intakes of high producing cows. Although MILK may be used for predicting  $P_E$ , accuracy of predictions increased when P intake was included in the equations. Development of a simple equation with P intake as the only predictor reduced the residual SE by 16% [9].

$$P_E = (\text{DMI} \times C_P \times 560.7 (\pm 71.1)) + 21.1 (\pm 7.7) \quad [9]$$

$$\text{Residual SE} = 9.7, \text{ Inter-study SE} = 9.2$$

In this dataset, P intake was the best single independent variable for predicting  $P_E$  (Figure 2). Similarly, Beede and Davidson (1999) and Weiss and Wyatt (2004) found that P intake was the most important single factor in determining  $P_E$ . When an additional equation that included MILK and MTP was evaluated for predicting  $P_E$ , there was a reduction in the residual SE from 9.7 to 9.3. However, the precision of future

predictions across studies was not improved with the addition of MILK and MTP to the equation.

Other researchers have proposed equations for the prediction of  $P_E$  (Weiss and Wyatt, 2004; Beede and Davidson, 1999; Van Horn et al., 1994). Beede and Davidson (1999) evaluated several  $P_E$  equations and determined that prediction of  $P_E$  was most accurately estimated using [10].

$$P_E = (\text{DMI} \times C_P \times 1000) - (\text{MilkP} \times 1000) \quad [10]$$

Subtracting MilkP from P intake to estimate  $P_E$  assumes there is not any tissue mobilization or retention. In the MINERAL dataset, evaluation of [10] resulted in a significant linear bias (-0.409,  $P < 0.01$ ), but no mean bias ( $P > 0.22$ ). In 2004, Weiss and Wyatt proposed using [11] to estimate  $P_E$ .

$$P_E = 7.5 + ((\text{DMI} \times C_P \times 1000) \times 0.78) - (\text{MILK} \times 0.702) \quad [11]$$

Evaluation of [11] indicated a mean bias (15.5 g/d,  $P < 0.02$ ) but no linear bias ( $P > 0.26$ ) when evaluated using the MINERAL dataset. The standard deviation of the residuals for the MINERAL dataset was 13.2 g/d.

***Potassium excretion.*** Potassium excretion occurs mainly in urine, with some unabsorbed K excreted in feces (NRC, 2001). Total  $K_E$  in the MINERAL dataset



averaged 200 g/d with urinary K excretion accounting for approximately 75% of  $K_E$ . The 2001 ASAE standards estimated  $K_E$  to be 182.7 g/d.

Potassium excretion was directly related to both MILK [12] and  $C_K$  [13]. When MILK was evaluated as the only significant factor to predict  $K_E$ , a positive relationship between MILK and  $K_E$  was found.

$$K_E = (\text{MILK} \times 1.476 (\pm 0.7207)) + 154.1 (\pm 24.5) \quad [12]$$

$$\text{Residual SE} = 43.2, \text{ Inter-study SE} = 23.6$$

Milk production has been reported to have a curvilinear relationship to K intake, with the peak milk yield occurring at a  $C_K$  of 0.015 g/g (NRC, 2001). Conversely, inclusion of squared terms and interactions did not improve the models for predicting  $K_E$  in the MINERAL dataset. The lack of a curvilinear relationship in our dataset was most likely due to the low  $C_K$  in the MINERAL dataset (0.0129 g/g).

Future prediction of  $K_E$  is expected to be more accurate if  $C_K$  or K intakes are used to predict excretion. The best equation for prediction of  $K_E$  included DMI and  $C_K$  [13].

$$K_E = (\text{DMI} \times 7.21 (\pm 1.150)) + (C_K \times 15944 (\pm 2691.2)) - 164.5 (\pm 50.6) \quad [13]$$

$$\text{Residual SE} = 36.9, \text{ Inter-study SE} = 2.7$$

When DMI and  $C_K$  were included in the equation, the SE between the studies in the dataset was very small compared to equations that included other variables for predicting  $K_E$ .

*Phosphorus and potassium excretion in early lactation cows.* Cows (15 cow-periods) from an early lactation study (average of 38 DIM) were evaluated separately from the MINERAL dataset because of the greater excretion of P and K for the early lactation animals compared to cows in later lactation. For early lactation cows, P intake was not a significant factor to predict  $P_E$ , and  $P_E$  was not significantly related to MILK, DMI, or Ca intake. Phosphorus excretion of these cows averaged over 23 g/d more than would be expected based on P intake and MILK. The greater  $P_E$  for these early lactation animals is most likely a result of greater endogenous fecal P losses, possibly related to bone mobilization.

Potassium excretion of cows in the early lactation dataset was greater than  $K_E$  of cows in the MINERAL dataset. On average,  $K_E$  for early lactation cows was 143 g/d greater than cows in the MINERAL dataset, even though K intakes were only 0.008 kg/d greater (Table 2). Due to the greater  $K_E$  and the greater secretion of K in milk, early lactation cows were in a negative K balance. Silanikove et al. (1997) found that cows in early lactation are often in a negative K balance and suggested that increased amounts of K in the diet may be beneficial to milk production.

## **Dry Cows**

The DRY dataset was a small dataset and consisted of 18 cows. Of these dry cows, 15 cows were fed diets specifically formulated for dry cows and 3 cows were fed diets formulated for lactating cows. Due to the limited dataset, prediction equations were not developed for dry cows and only average intake and excretion values are reported.

Manure excretion from dry cows averaged 38.6 kg/d (Table 5), which was 12.3 kg/d more than reported by Wilkerson et al. (1997), but similar to the 36.3 kg/d estimate by Van Horn et al. (1994). The lower  $M_E$  values from Wilkerson et al. (1997) are most likely a result of the restricted intakes for dry cows in their studies. The  $DM_E$  estimate of 4.5 kg/d for dry cows was reported by Van Horn et al. in 1994 was the same as the average value in the DRY dataset.

Manure excretion for the DRY dataset was 59% of the 2001 ASAE value for  $M_E$  for a cow weighing 755 kg. However, the 2001 ASAE values do not differentiate between lactating and non-lactating animals.

Mean  $N_E$  from dry cows was 228 g/d, though the range in  $N_E$  was large (Table 5), and was 49 g/d greater than reported by Wilkerson et al. (1997). Estimates of  $N_E$  in the 2001 ASAE standards were 340 g/d for dairy cattle and 257 g/d for beef cattle. Clearly, estimates for dry cows are closer to the 2001 ASAE estimates for beef cattle than dairy cattle. Addition of dry cows to the updated standards will be an

improvement over ASAE 2001 values and will improve the flexibility and accuracy of the standards.

The DRY dataset was used to evaluate linear equations for  $M_E$  and  $N_E$  published by Wilkerson et al. (1997). Dry cows were assumed to be 230 days pregnant because day of pregnancy was not available in the dry cow dataset. When the Wilkerson et al. (1997) equation [ $M_E = (0.00711 \times BW) + (32.4 \times C_{CP}) + (25.9 \times C_{NDF}) + 8.05$ ] was evaluated, there were no significant mean or linear biases ( $P > 0.13$ ) and the equation accurately described the cows in this small dataset. However, evaluation of the  $N_E$  equation [ $N_E = (0.000107 \times BW) + (1.11 \times C_{CP}) + (0.170 \times C_{NDF}) - 0.135$ ] (Wilkerson et al., 1997) resulted in significant mean (279.5 g/d;  $P < 0.01$ ) and linear biases (1.19;  $P < 0.01$ ). In the future, more research is needed on dry cows fed diets typical in the industry to be able to develop regression equations.

## **Heifers**

The dataset for heifers included 60 observations that ranged in BW from 274 to 613 kg (Table 2). The 2001 ASAE manure excretion estimates for dairy cattle were not categorized by animal age. Excretion estimates for heifers were not specifically available and would have to be approximated using either dairy cattle or beef cattle data. Determination of new prediction equations for growing dairy heifers was difficult due to a shortage of total collection metabolism trials recently conducted on this class of animals.

Manure excretion in the HEIFER dataset was overestimated by ~54% for the average heifer (437 kg) using the 2001 ASAE dairy excretion estimates, but by only ~3% using the beef estimates. The most accurate equation for predicting  $M_E$  for heifers included BW and DMI [14]. Manure excretion was dependent on BW of the animal alone [15], although the addition of DMI to the equation provided a more precise predictor of  $M_E$ .

$$M_E = (\text{DMI} \times 4.158 (\pm 0.536)) - (\text{BW} \times 0.0246 (\pm 0.0103)) \quad [14]$$

$$\text{Residual SE} = 2.6, \text{ Inter-study SE} = 5.6$$

$$M_E = (\text{BW} \times 0.0181 (\pm 0.0104)) + 17.8 (\pm 4.8) \quad [15]$$

$$\text{Residual SE} = 3.6, \text{ Inter-study SE} = 4.0$$

Nitrogen excretion from dairy heifers in the HEIFER dataset averaged 117.3 g/d, which was 80.4 and 31.3 g/d less than predicted by the 2001 ASAE dairy and beef values, respectively. An equation [16] developed for  $N_E$  using the HEIFER dataset resulted increased in  $N_E$  when CP intake increased (Figure 3).

$$N_E = (\text{DMI} \times C_{CP} \times 78.39 (\pm 13.01)) + 51.4 (\pm 17.0) \quad [16]$$

$$\text{Residual SE} = 10.8, \text{ Inter-study SE} = 24.5$$

Hoffman et al. (2001), James et al. (1999), and Wilkerson et al. (1997) also reported increased  $N_E$  for heifers fed greater levels of N. Hoffman et al. (2001) found that growth of Holstein heifers was optimized when the  $C_{CP}$  was 0.13 g/g DM. The average  $C_{CP}$  was 0.112 g/g DM in the HEIFER dataset (Table 2). Feeding heifers diets with a  $C_{CP}$  of 0.13 g/g DM would be expected to increase  $N_E$  compared to the average in the HEIFER dataset.

Development of prediction equations for  $P_E$  in the HEIFER dataset was not possible due to the limited data available and the variation within the dataset.

Previously developed prediction equations for  $M_E$  and  $N_E$  in heifers were evaluated. When the equation for  $M_E$  [ $M_E = (0.0499 \times BW) + (44.2 \times C_{CP}) + (5.86 \times C_{NDF}) - 5.918$ ] (Wilkerson et al., 1997) was evaluated, there were no significant mean or linear biases, indicating this previously published equation adequately described the heifers in this dataset.

Evaluation of a previously published (Wilkerson et al., 1997)  $N_E$  equations [ $N_E = (0.000471 \times BW) + (0.867 \times C_{CP}) - (0.0109 \times C_{NDF}) - 0.159$ ] from heifers resulted in no linear bias, but there was a trend ( $P < 0.07$ ) towards a mean bias. The mean bias for the equation was 121.2 g/d, indicating an under-prediction of  $N_E$  from animals in the HEIFER dataset.

## **Calves**

The dataset for calves included 46 observations ranging in BW from 86 to 205 kg (Table 2). Development of equations for excretion estimates from calves used a small dataset because of a shortage of calf data from total collection metabolism studies. As with heifers, estimates of excretion for calves were not available in the 2001 ASAE manure excretion estimates for dairy cattle. Use of the 2001 ASAE manure excretion estimates for calves would not be expected to be accurate for estimating  $M_E$  or nutrient excretion.

Average  $M_E$  from the CALF dataset was approximately half the  $M_E$  of the HEIFER dataset (12.1 kg/d less), even though the average calf BW was about one-third as much as the heifers (Tables 2 and 5). The most accurate equation for predicting  $M_E$  of calves included DMI [17].

$$M_E = (\text{DMI} \times 3.45 (\pm 0.41)) \quad [17]$$

$$\text{Residual SE} = 5.6, \text{ Inter-study SE} = 2.5$$

Although DMI was the best predictor of  $M_E$  for the CALF dataset, BW was also a significant predictor of  $M_E$  [18].

$$M_E = (\text{BW} \times 0.0811 (\pm 0.0086)) \quad [18]$$

$$\text{Residual SE} = 5.6, \text{ Inter-study SE} = 2.1$$

Prediction of  $DM_E$  was similar to  $M_E$  for calves. Equation [19] shows the relationship between  $DM_E$  and DMI for the CALF dataset, with greater  $DM_E$  occurring as DMI increases.

$$DM_E = (DMI \times 0.393 (\pm 0.032)) \quad [19]$$

$$\text{Residual SE} = 0.31, \text{ Inter-study SE} = 0.24$$

Nitrogen excretion was directly related to CP intake in the CALF dataset [20] as it was for the other classes of dairy animals. In the CALF dataset, the coefficient for CP intake was greater than was seen in the HEIFER dataset (112.6 and 78.4, respectively) for the simple linear equation to predict  $N_E$  (Figures 3 and 4).

Evaluation of [20] with the HEIFER dataset resulted in a significant linear bias of  $-0.873$  ( $P < 0.01$ ) and a significant mean bias of  $-79.7$  g/d ( $P < 0.01$ ) of excreted N. The significant mean bias indicated that  $N_E$  for heifers would be overestimated if [20] was used to predict  $N_E$ .

$$N_E = (DMI \times C_{CP} \times 112.55 (\pm 2.13)) \quad [20]$$

$$\text{Residual SE} = 8.2$$

A relationship between P intake and  $P_E$  was seen in the CALF dataset (Figure 5) even though there was not a significant relationship in the HEIFER dataset. Phosphorus excretion increased with greater P intakes in the CALF dataset [21].



$$P_E = (\text{DMI} \times C_P \times 622.03 (\pm 37.58)) \quad [21]$$

Residual SE = 1.44, Inter-study SE = 0.57

Although there was not a direct relationship between  $P_E$  and P intake in the HEIFER dataset, when equation [21] was evaluated in the HEIFER dataset, there were no significant mean or linear biases. Because [21] accurately predicted the  $P_E$  from HEIFERS, this equation could be used for non-lactating heifers regardless of BW.

## CONCLUSIONS

The proposed revisions of the ASAE manure excretion estimates for dairy cattle provide updated values that more closely reflect the excretion values of contemporary animals as well as providing excretion estimates for various classes of animals, including heifers and dry cows. The 2001 ASAE standards underestimate manure and N excretion from high producing dairy cows while overestimating manure and nutrient excretion from dry cows, calves, and heifers. The most significant change to the 2001 ASAE standards included the addition of prediction equations, which provide a method by which excretion estimates can be determined according to production levels and dietary factors for a given operation. The improved ability to predict nutrient excretion will be essential information for technical service providers and producers to consider when developing nutrient management plans.

## ACKNOWLEDGMENTS

The authors would like to thank the National Center for Manure and Animal Waste Management, North Carolina State University and Church & Dwight, Co. for the financial support provided for sample analyses.

## REFERENCES

- American Society of Agriculture Engineers. 2001. Manure production and characteristics. ASAE Standards D384.1. ASAE, St. Joseph, MI.
- Beede, D. K., and J. A. Davidson. 1999. Phosphorus: Nutritional management for Y2K and beyond. Pages 51-97 *in* Tri-State Dairy Nutr. Conf., Ft. Wayne, IN.
- Environmental Protection Agency. 2003. National pollutant discharge elimination system permit regulation and effluent limitation guidelines and standards for concentrated animal feeding operations (CAFOs). Final Rule. Federal Register 68:7176-7274.
- Frank, B., M. Person, and G. Gustafsson. 2002. Feeding dairy cows for decreased ammonia emission. *Livest. Prod.* 76:171-179.
- Gabler, M. T., and A. J. Heinrichs. 2003a. Altering soluble and potentially degradable protein for prepubertal Holstein heifers. *J. Dairy Sci.* 86:2122-2130.
- Gabler, M. T., and A. J. Heinrichs. 2003b. Effects of increasing dietary protein on nutrient utilization in heifers. *J. Dairy Sci.* 86:2170-2177.
- Harrison, J. H., L. Johnson, J. Werkhoven, A. Werkhoven, S. Werkhoven, M. Vazquez-Anon, G. Winter, N. Barney, and W. Chalupa. 2002. Effectiveness of strategic ration balancing on efficiency of milk protein production and environmental impact. *J. Dairy Sci.* 85(Suppl. 1):205. (Abstr.)
- Hoffman, P. C., N. M. Esser, L. M. Bauman, S. L. Denzine, M. Engstrom, and H. Chester-Jones. 2001. Short communication: Effect of dietary protein on growth and nitrogen balance of Holstein heifers. *J. Dairy Sci.* 84:843-847.
- James, T., D. Meyer, E. Esparza, E. J. DePeters, and H. Perez-Monti. 1999. Effects of dietary nitrogen manipulation on ammonia volatilization from manure from Holstein heifers. *J. Dairy Sci.* 82:2430-2439.

- Johnson, L. M., J. H. Harrison, and R. E. Riley. 1998. Estimation of the flow of microbial nitrogen to the duodenum using urinary uric acid or allantoin. *J. Dairy Sci.* 81:2406-2420.
- Krober, T. F., D. R. Kulling, H. Menzi, F. Sutter, and M. Kreuzer. 2000. Quantitative effects of feed protein reduction and methionine on nitrogen use by cows and nitrogen emission from slurry. *J. Dairy Sci.* 83:2941-2951.
- Meyer, D., V. Erikson, E. Tooman, and T. James. 2000. Manure production characteristics from lactating Holsteins. Pages 157-159 *in Proc. Western Section ASAS, Davis, CA.*
- National Research Council. 2001. *Nutrient Requirements of Dairy Cattle.* 7<sup>th</sup> rev. ed. Natl. Acad. Sci., Washington, D. C.
- Natural Resources Conservation Service. 2001. Nutrient management. Natural Resources Conservation Service Conservation Practice Standard Code 590-1. USDA-NRCS. Washington State.
- SAS User's Guide: Statistics, Version 8.2. 1999. SAS Inst., Inc., Cary, NC.
- Silanikove, N., E. Maltz, A. Halevi, and D. Shinder. 1997. Metabolism of water, sodium, potassium, and chlorine by high yielding dairy cows at the onset of lactation. *J. Dairy Sci.* 80:949-956.
- St-Pierre, N. R. 2001. Integrating quantitative findings from multiple studies using mixed model methodology. *J. Dairy Sci.* 84:741-755.
- St-Pierre, N. R. 2003. Reassessment of biases in predicted nitrogen flows to the duodenum by NRC 2001. *J. Dairy Sci.* 86:344-350.
- Timmermans Jr., S. J., L. M. Johnson, J. H. Harrison, and D. Davidson. 2000. Estimation of the flow of microbial nitrogen to the duodenum using milk uric acid or allantoin. *J. Dairy Sci.* 83:1286-1299.
- Tomlinson, A. P., W. J. Powers, H. H. Van Horn, R. A. Nordstedt, and C. J. Wilcox. 1996. Dietary protein effects on nitrogen excretion and manure characteristics of lactating cows. *Trans. ASAE* 39:1441-1448.
- Van Horn, H. H., A. C. Wilkie, W. J. Powers, and R. A. Nordstedt. 1994. Components of dairy manure management systems. *J. Dairy Sci.* 77:2008-2030.
- Weiss, W. P., and D. J. Wyatt. 2004. Macromineral digestion by lactating dairy cows: estimating phosphorus excretion via manure. *J. Dairy Sci.* 87:2158-2166.

Wilkerson, V. A., D. R. Mertens, and D. P. Casper. 1997. Prediction of excretion of manure and nitrogen by Holstein dairy cattle. *J. Dairy Sci.* 80:3193-3204.

Zanton, G. I., and A. J. Heinrichs. 2003. Altering protein degradability and solubility on rumen fermentation, blood urea nitrogen, and nitrogen balance in 16-18 month-old heifers. *J. Dairy Sci.* 86(Suppl. 1):22.

Table 1. Experiments included in the datasets used for development of excretion equations.

Data source	Experiment source or description
Washington State University	<p><i>Published experiments</i>            Johnson et al., 1998            Timmermans et al., 2000</p> <p><i>Description of unpublished experiments</i></p> <ol style="list-style-type: none"> <li>1. Treatments included fishmeal or distillers grain plus an additional protein supplement.</li> <li>2. Dry rolled corn or high moisture corn or 2 combinations of dry rolled and high moisture corn.</li> <li>3. Dietary treatments contained either control corn silage or corn silage ensiled with inoculant.</li> </ol>
Pennsylvania State University	<p><i>Published experiments</i>            Gabler and Heinrichs, 2003a            Gabler and Heinrichs, 2003b            Zanton et al., 2003</p>
The Ohio State University	<p><i>Published experiments</i>            Weiss and Wyatt, 2004</p>
University of California, Davis	<p><i>Published experiments</i>            James et al., 1999            Meyer et al., 2000</p>

Table 2. Animal and production characteristics for the datasets used for development of prediction equations. Characteristics are included for datasets with all lactating cows (LACT), lactating cows for which mineral data was available (MINERAL), dry cows (DRY), heifers (HEIFERS), and calves (CALF).

Item	n	Mean	Minimum	Maximum	SD
<i>LACT</i>					
BW, kg	554	630	437	830	66.1
DIM	552	172	13	563	91.9
Milk, kg/d	553	31.4	1.4	86.1	11.0
DMI, kg/d	553	21.7	6.8	32.9	3.9
Milk fat, g/g milk	554	0.0362	0.0225	0.0673	0.0067
Milk protein, g/g milk	529	0.0299	0.0161	0.0447	0.0034
Dietary CP, g/g DM	529	0.175	0.124	0.244	0.0185
Dietary NDF, g/g DM	554	0.364	0.265	0.454	0.0507
<i>MINERAL</i>					
BW, kg	85	617	437	745	69.4
DIM	85	165	27	356	80.2
Milk, kg/d	85	31.5	6.0	44.3	7.8
DMI, kg/d	85	21.9	10.5	30.1	4.3
Milk fat, g/g DM	85	0.0355	0.0230	0.0550	0.0066
Milk protein, g/g DM	85	0.0292	0.0218	0.0381	0.0030
Dietary P, g/g DM	66	0.0044	0.0025	0.0060	0.0007
Dietary K, g/g DM	66	0.0129	0.0096	0.0176	0.0019
<i>DRY</i>					
BW, kg	18	755	413	934	155.3
DMI, kg/d	18	10.4	5.1	16.8	3.2
Dietary CP, g/g DM	18	0.133	0.080	0.230	0.061
Dietary NDF, g/g DM	11	0.466	0.350	0.579	0.094
<i>HEIFER</i>					
BW, kg	60	437	274	613	65.4
DMI, kg/d	60	8.34	6.23	10.66	1.12
Dietary CP, g/g DM	60	0.112	0.073	0.216	0.031
Dietary NDF, g/g DM	36	0.498	0.379	0.598	0.059
Dietary P, g/g DM	32	0.0029	0.0026	0.0035	0.0002
Dietary K, g/g DM	32	0.0147	0.0121	0.0178	0.0019
<i>CALF</i>					
BW, kg	46	152.8	86.0	203.7	31.0
DMI, kg/d	46	3.37	2.38	5.15	0.50
Dietary CP, g/g DM	46	0.166	0.092	0.227	0.024
Dietary NDF, g/g DM	44	0.395	0.299	0.512	0.055
Dietary P, g/g DM	32	0.0037	0.00032	0.0029	0.0042

Table 3. Definitions of variables used in the prediction equations developed for the proposed revisions of the ASAE manure characteristics. Units are on a per animal basis.

Variable	Description	Units
<i>Animal performance characteristics</i>		
MILK	Milk yield	kg milk/d
MF	Milk fat	g fat/g milk
MTP	Milk true protein	g protein/g milk
MilkP	Milk P	g P/g milk
DIM	Days in milk	d
BW	Body weight	kg
<i>Dietary characteristics</i>		
DMI	Dry matter intake	kg DM/d
DMD	Apparent dry matter digestibility of total ration	g/g DM
C <sub>cp</sub>	Concentration of crude protein of total ration	g crude protein/g DM
C <sub>NDF</sub>	Concentration of neutral detergent fiber of total ration	g neutral detergent fiber/g DM
C <sub>P</sub>	Concentration of P of total ration	g P/g DM
C <sub>K</sub>	Concentration of K of total ration	g K/g DM
<i>Excretion</i>		
M <sub>E</sub>	Total manure excretion	kg/d
N <sub>E</sub>	Total N excretion	g/d
P <sub>E</sub>	Total P excretion	g/d
K <sub>E</sub>	Total K excretion	g/d
DM <sub>E</sub>	Dry matter excretion	kg/d

Table 4. Estimated typical manure (urine and feces combined) characteristics as excreted<sup>1</sup> by dairy animals.

Animal Type and Production Grouping	Total manure kg/d	Total dry matter kg/d	N g/d	P g/d	K g/d
Lactating cow <sup>2</sup>	75.2	9.7	491	74	223
Dry cow <sup>3</sup>	38.6	4.5	228	NA <sup>6</sup>	NA
Heifer <sup>4</sup>	24.5	3.74	117	20	NA
Calf <sup>5</sup>	12.4	1.37	63	8	NA
2001 ASAE <sup>7</sup>	53.8	NA	281	59	181

<sup>1</sup>Prior to any changes due to dilution water addition, drying, volatilization or other physical, chemical or biological processes.

<sup>2</sup>Lactating cow excretion estimates are based on a 625 kg cow producing 40 kg milk per day with intakes of 25 kg DM, 4.38 kg CP, 0.095 kg P, and 0.325 kg K per day.

<sup>3</sup>Dry cow excretion estimates based on a 755 kg cow with intakes of 10.4 kg DM and 1.38 kg N per day.

<sup>4</sup>Heifer excretion estimates are based on a 437 kg heifer with intakes of 8.3 kg DM, 0.93 kg CP, and 0.024 kg P per day.

<sup>5</sup>Calf excretion estimates are based on a 153 kg calf with intakes of 3.4 kg DM, 0.56 kg CP and 0.013 kg P per day.

<sup>6</sup>Data not available

<sup>7</sup>ASAE values are based on a 625 kg animal.



Table 5. Average excretion values for animals in the datasets of lactating cows (LACT), lactating cows for which mineral data was available (MINERAL), dry cows (DRY), heifers (HEIFERS), and calves (CALF).

Item	n	Mean	Minimum	Maximum	SD
<i>LACT</i>					
Manure, kg/d	554	66.3	27.7	114.4	14.4
Total DM, kg/d	538	8.52	3.22	14.78	1.80
Fecal DM, kg/d	538	7.25	1.74	12.93	1.63
Urine, kg/d	554	23.1	8.4	58.7	7.19
Nitrogen, g/d	529	438.7	180.0	741.0	94.3
Fecal N, g/d	530	222.3	72.3	441.9	59.1
Urinary N, g/d	529	216.5	63.0	498.6	64.3
<i>MINERAL</i>					
Manure, kg/d	85	67.4	34.5	100.8	13.4
Total P, g/d	85	73.9	27.4	114.5	16.2
Total K, g/d	85	200.3	92.0	348.8	48.7
Urinary K, g/d	85	150.5	53.8	305.6	45.0
<i>DRY</i>					
Manure, kg/d	18	38.6	22.2	54.8	8.9
Total DM, kg/d	18	4.54	2.49	5.84	0.92
Urine, kg/d	18	15.4	8.4	32.1	6.3
Nitrogen, g/d	18	228.4	80.0	503.3	136.6
Fecal N, g/d	18	90.1	30.0	203.3	53.8
Urinary N, g/d	18	138.3	40.0	330.0	89.7
<i>HEIFERS</i>					
Manure, kg/d	60	24.5	18.1	40.3	4.8
Total DM, kg/d	32	3.74	3.18	4.47	0.32
Urine, kg/d	60	9.0	6.3	18.7	3.0
Nitrogen, g/d	60	117.3	70.0	220.0	31.7
Fecal N, g/d	60	57.7	30.0	80.0	1.2
Urinary N, g/d	60	59.7	30.0	160.0	30.5
P, g/d	32	20.4	18.73	22.14	1.13
<i>CALF</i>					
Manure, kg/d	46	12.4	5.0	28.8	6.4
Total DM, kg/d	46	1.37	0.66	2.54	0.47
Nitrogen, g/d	46	62.8	33.5	110.0	14.6
Fecal N, g/d	46	23.6	10.0	40.0	5.8
Urinary N, g/d	46	39.3	18.5	73.8	12.3
P, g/d	32	7.8	5.0	12.6	1.7

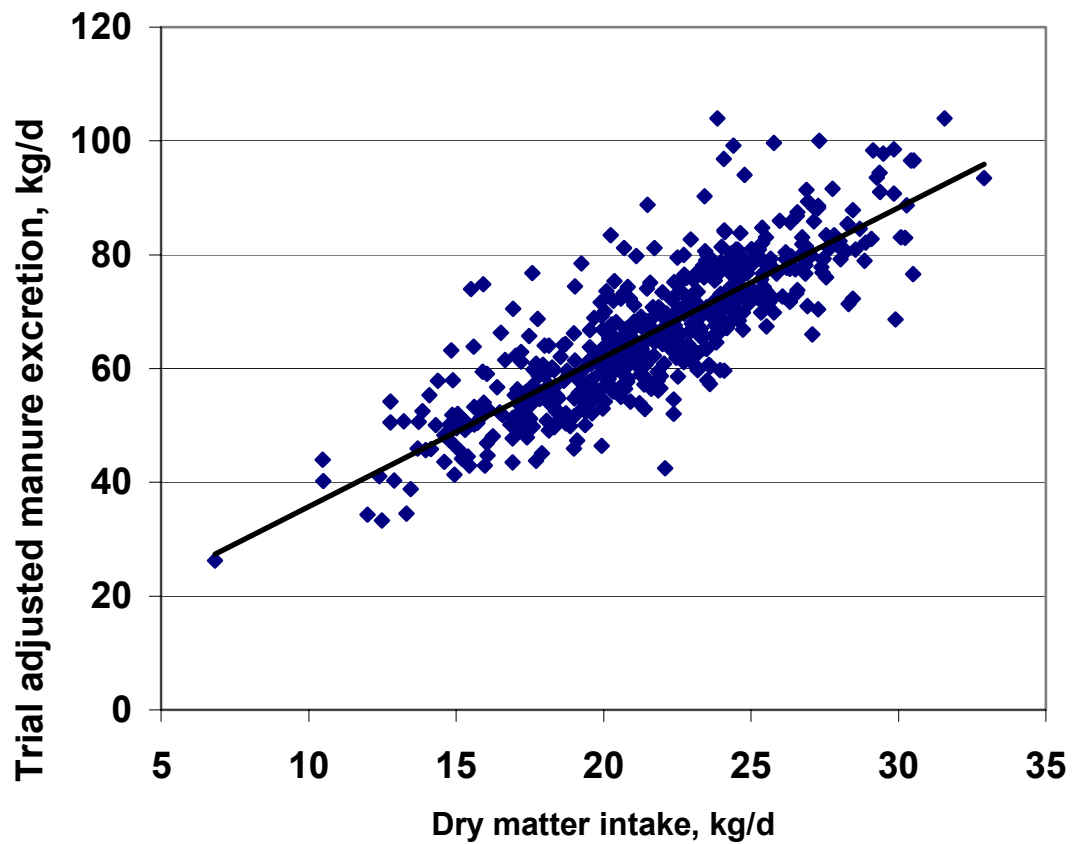


Figure 1. Relationship between dry matter intake (DMI) and trial adjusted manure excretion for lactating cows (554 cow-periods). The solid line is equal to manure excretion, kg/d = (DMI, kg/d x 2.63) + 9.4, Residual SE = 7.1, Inter-study SE = 9.5.

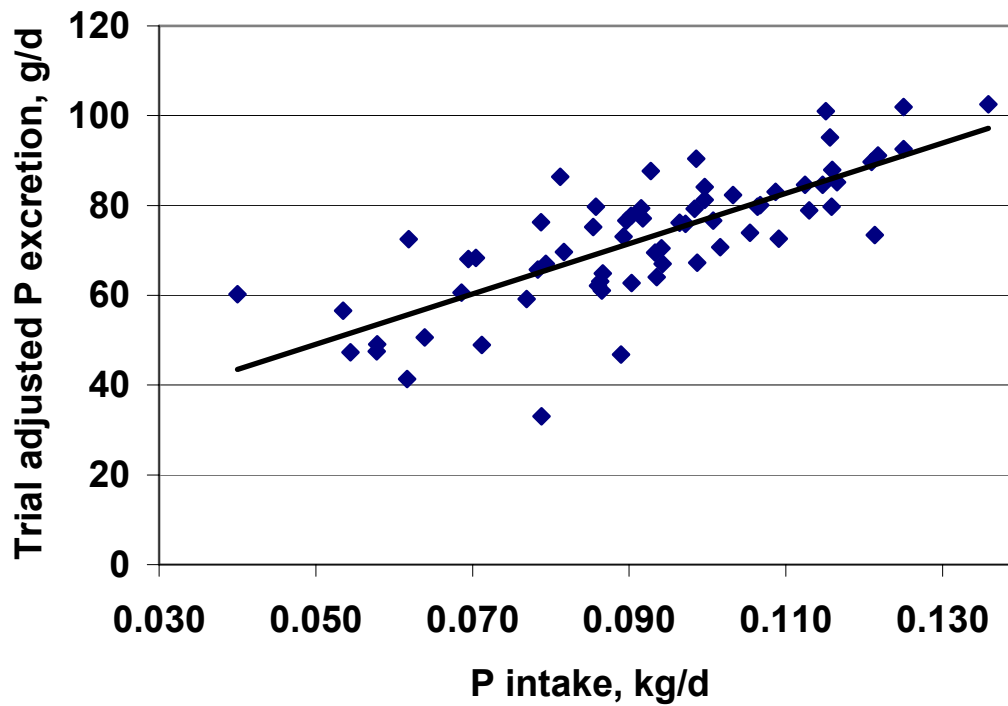


Figure 2. Relationship between P intake and trial adjusted P excretion for lactating cows (66 cow-periods). The solid line is equal to  $P \text{ excretion, g/d} = (P \text{ intake, kg/d} \times 560.7) + 21.1$ , Residual SE = 9.7, Inter-study SE = 9.2.

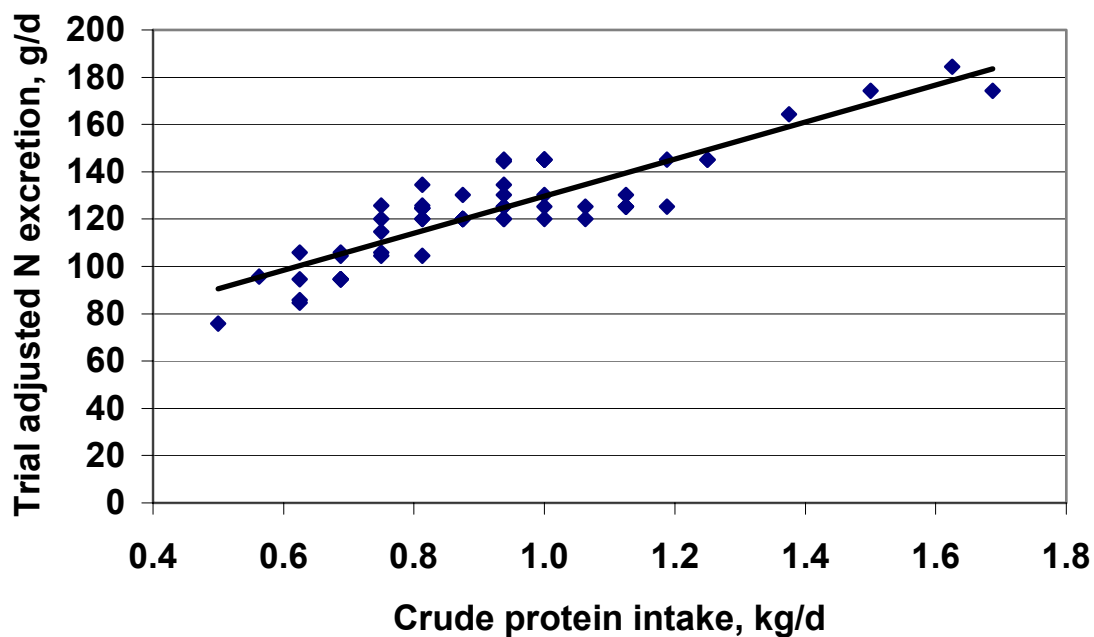


Figure 3. Relationship between crude protein (CP) intake and trial adjusted N excretion for heifers (60 animal-periods). The solid line is equal to  $N \text{ excretion, g/d} = (\text{CP intake, kg/d} \times 78.39) + 51.4$ , Residual SE = 10.8, Inter-study SE = 24.5.

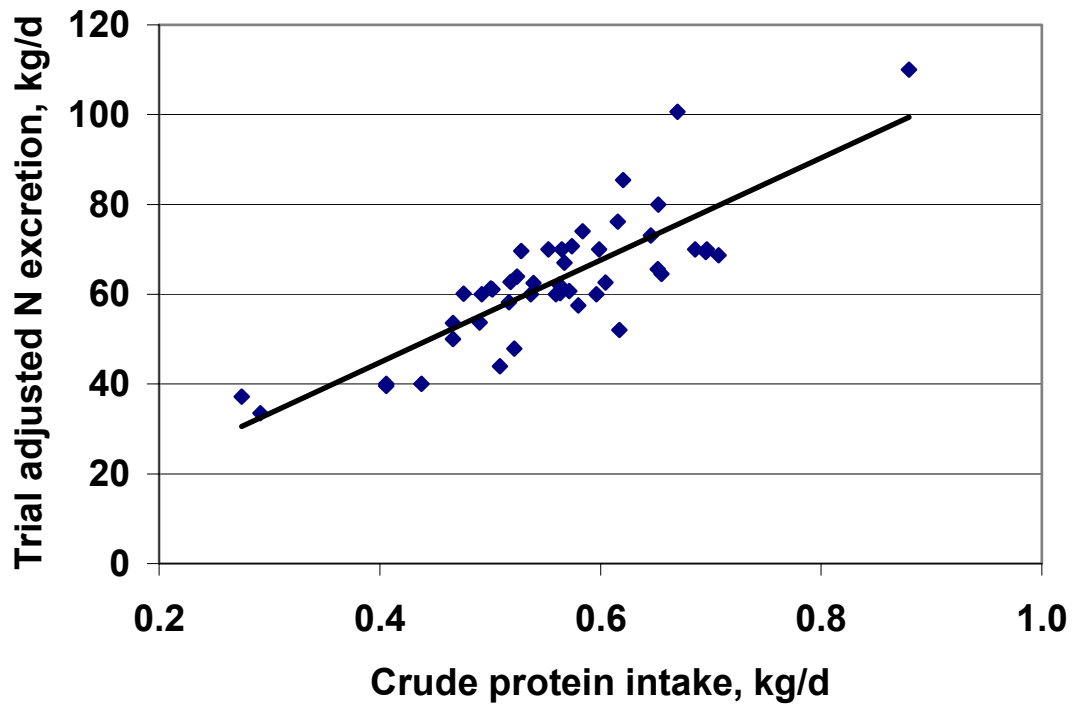


Figure 4. Relationship between crude protein (CP) intake and trial adjusted N excretion for calves (46 animal-periods). The solid line is equal to  $N \text{ excretion, g/d} = (\text{CP intake, k/d} \times 112.55)$ , Residual SE = 8.2.

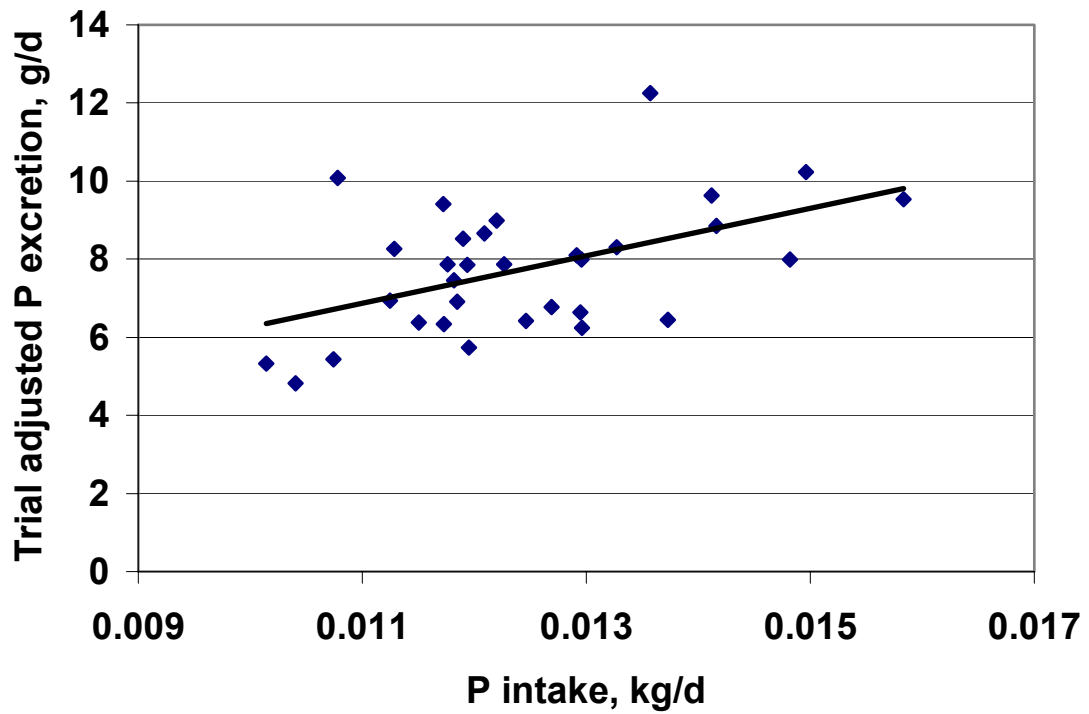


Figure 5. Relationship between P intake and trial adjusted P excretion for calves (32 animal-periods). The solid line is equal to P excretion, g/d = (P intake, kg/d x 622.03), Residual SE = 1.4, Inter-study SE = 0.6.

# **Prediction and Evaluation of Urine and Urinary Nitrogen and Mineral Excretion from Dairy Cattle**

**T. D. Nennich<sup>\*</sup>, J. H. Harrison<sup>\*</sup>, L. M. VanWieringen<sup>\*</sup>,  
N. R. St-Pierre<sup>†</sup>, R. L. Kincaid<sup>‡</sup>, M. A. Wattiaux<sup>§</sup>,  
D. L. Davidson<sup>\*</sup>, and E. Block<sup>||</sup>**

<sup>\*</sup>Puyallup Research and Extension Center,  
Washington State University, Puyallup 98371

<sup>†</sup>Department of Animal Sciences,  
The Ohio State University, Columbus 43210

<sup>‡</sup>Department of Animal Sciences,  
Washington State University, Pullman 99163

<sup>§</sup>Department of Animal Science,  
University of Wisconsin, Madison 53706

<sup>||</sup>Church & Dwight Co. Inc.,  
Princeton, NJ 08543

Corresponding author:

Joseph Harrison  
7612 Pioneer Way E.  
Puyallup, WA 98371  
253/445-4638  
jhharrison@wsu.edu

## ABSTRACT

Accurate estimates of manure excretion are needed for planning of manure storage facilities and for nutrient management. Previously developed prediction equations need to be validated to verify their accuracy across various diets and environments. The objective of this study was to develop equations for prediction of urine excretion and excretion of urinary N, Na, and K, and to evaluate previously published prediction equations for estimation of urine and urinary nutrient excretion from lactating dairy cows. Datasets from metabolism studies conducted at Washington State University were compiled and evaluated for excretion of minerals. Urine excretion averaged 24.1 kg/d and urinary nitrogen (UN) ranged from 63 to 499 g/d in the calibration dataset. Regression equations were developed to predict urine excretion, urinary N excretion, and urinary Na and K excretion. Predictors used in the regression equations included milk yield, body weight, dietary crude protein percentage, milk urea nitrogen, and nutrient intakes. Previously published prediction equations were evaluated using a combination of datasets from Washington State University and the University of Wisconsin. Mean and linear biases were evaluated by determining the regression of residuals on predicted values. Evaluation and validation of prediction equations are important to develop equations that will more accurately estimate urine and urinary nitrogen excretion from lactating dairy cows.

**(Key words:** urine, urinary nitrogen, sodium, potassium)

**Abbreviation key:** **ASAE** = American Society of Agricultural Engineers, **MILK** = milk yield, **MUN** = milk urea N, **MF** = milk fat percent, **MTP** = milk protein percent,



**NI** = N intake, **NaI** = Na intake, **KI** = K intake, **RDP** = rumen degradable protein supply, **UNaPct** = urinary Na percent, **UKPct** = urinary K percent, **U<sub>E</sub>** = urine excretion, **U<sub>N</sub>** = urinary N excretion, **U<sub>Na</sub>** = urinary Na excretion, **U<sub>K</sub>** = urinary K excretion.

## INTRODUCTION

Estimates of urine and fecal excretion from dairy cattle are becoming more important as state and federal legislation are requiring dairy operations to have nutrient management plans. Excretion estimates are important for designing storage facilities and developing nutrient management plans. Increased feed intake and milk production of dairy cows have led to greater excretion of nutrients that is often not accounted for when planning modern dairy operations.

The amount of urine excreted greatly affects total manure excretion from lactating dairy cows as urine excretion accounts for approximately one-third of total manure excretion on a weight basis (NRC, 2001). Urine excretion has been directly linked to intake of N, Na, and K (Bannink et al., 1999). Excretion of urine can potentially be reduced by supplying nutrients in levels that meet, but do not exceed, animal requirements. Sodium and K intakes have been shown to affect total urinary excretion (Bannink et al., 1999; Fisher et al., 1994). Increased urine excretion results in requirements for larger manure storage facilities and may even require additional nutrient export or land for application.

Nitrogen excretion is an environmental concern because it can lead to ammonia volatilization or movement to surface or groundwater. The main route of excretion for excess dietary N is via urine. Excretion of urea in urine results in greater ammonia volatilization due to the breakdown of the urea when it comes into contact with urease enzymes present in feces. In addition, excretion of urinary N has been directly linked with milk urea N (**MUN**) concentrations (Jonker et al, 1998; Kauffman and St-Pierre, 2001; Kohn et al., 2002).

Urinary excretion is the main route of excretion for Na and K (Miller, 1975). Because urinary excretion is the main method of homeostatic regulation mechanisms for Na and K, intake of these minerals will directly affect excretion (Miller, 1975). Potassium is a mineral of concern for livestock producers when they are developing nutrient management plans. The Natural Resources Conservation Service standard for nutrient management planning (NRCS, 2001) states that nutrient budgets should be established for N, P, and K. Although K has not been directly linked to environmental problems, excess field application of K can result in a buildup of soil K. Greater soil concentrations of K can lead to higher concentrations of K in forages as grasses are luxury consumers of K (Harrison et al., 2003). Forage K levels affect intake of K, which is a concern for periparturient dairy cows (NRC, 2001).

Innovations in management techniques are continually being explored to decrease risks of nutrients leaving livestock operations through air or water. An

example of a management strategy designed to decrease ammonia volatilization is the separation of urine and feces in animal housing facilities. In Sweden, 36% of ammonia volatilization is a result of manure storage (Frank et al., 2002). Ammonia volatilization from stored slurry decreased when urinary N excretion was reduced (Krober et al., 2000). Development of new management strategies that focus specifically on urine management requires information to better estimate urine excretion on dairy operations and to gain a better understanding of factors affecting urine and urinary nutrient excretion. The main objectives of this study were 1.) the development of new equations to predict excretion of urine, urinary N, urinary Na, and urinary K and 2.) evaluation of previously published equations developed to predict excretion of urine, urinary N, and urinary minerals. During the development of prediction equations, an effort was made to include equations for use on commercial dairy operations and to gain a better understanding of dietary and metabolic factors affecting excretion.

## **MATERIALS AND METHODS**

Datasets from Washington State University and were combined and used for estimations of urinary excretion from dairy cattle (Table 1). The overall dataset only included records from multiparous lactating cows. Data were divided into two groups including lactating cows and a subset of lactating cows for which mineral intake and excretion data were available. The LACT dataset (372 cow-periods from Latin square experiments) included lactating Holstein cows. The MINERAL dataset (115 cow-

periods) included cows for which intakes and excretion of minerals were known. Intakes of minerals were determined through analyses of both feed and orts.

The LACT dataset included observations from 16 individual feeding studies. Feeding studies were originally intended to evaluate nutritional hypothesis and were not designed specifically for development of excretion prediction equations. Variations in dietary ingredients, milk production, and days in milk increased the errors associated with development of regression equations.

The independent variables included in the LACT dataset were BW, kg/animal, DIM, DMI, kg/d, milk yield, kg/d (**MILK**), milk urea N, mg/dl (**MUN**) which was determined by colorimetry, percent milk fat (**MF**), and percent milk protein (Table 2). Milk CP values in the dataset were converted to percent milk true protein (**MTP**) values using a conversion factor of 0.940. Dietary ingredients and characteristics were used as additional prediction factors for equations, including dietary concentrations of CP, % of DM (**C<sub>CP</sub>**), NDF, % of DM (**C<sub>NDF</sub>**), and N intake, g/d (**NI**). The supply of rumen degradable protein, g/d (**RDP**) was determined for each individual cow using the 2001 Dairy NRC model. Dependent variables included in the LACT included urine excretion, kg/d (**U<sub>E</sub>**) and urinary N excretion, g/d (**U<sub>N</sub>**). The MINERAL dataset included the same variables as the LACT dataset and also included the following independent variables: Na intake, g/d (**NaI**), K intake, g/d (**KI**), urinary Na concentration, % (**UNaPct**), and urinary K concentration, % (**UKPct**). The dietary cation-anion difference (**DCAD**) was determined using the following equation: (Na +

$K) - (S + CI)$ . The MINERAL dataset also included the dependent variables of urinary Na excretion, g/d ( $U_{Na}$ ) and urinary K excretion, g/d ( $U_K$ ).

Diets fed during the metabolism trials included a wide variety of protein supplements and forage types. Forages included corn silage, grass silage, alfalfa silage, and grass hay. The remainder of the diets included various grains, by-product feeds, and mineral supplements. Cows in these trials were fed ad libitum.

Equations given for each parameter include residual standard error (SE) and inter-study SE. Equations with lower SE are expected to provide a more precise estimation of excretion and should be used when values for the input variables are available.

Evaluations of previously published prediction equations for estimating  $U_E$  and  $U_N$  were done using a combination of the LACT dataset and the VALIDATE dataset. The VALIDATE dataset included 48 observations of multiparous lactating Holstein cows conducted by Wattiaux and Karg (2004). Animals characteristics for the VALIDATE dataset are given in Table 2.

### **Sample Collection and Analysis**

Total collection metabolism studies (16 studies) conducted at Washington State University included lactating cows (372 cow-periods) and cows with mineral data (115 cow-periods). Feeding, sample collection, and sample analyses were conducted by

methods outlined by Johnson et al. (1998) and Timmermans et al. (2000). Studies from the University of Wisconsin that were included in the validation dataset included 48 observations from multiparous lactating cows. Descriptions of feeding, sampling, and collections were summarized by Wattiaux and Karg (2004).

Mineral analyses were not conducted on milk samples. Therefore, milk mineral contents were assumed to be equivalent to values outlined in the 2001 Dairy NRC. Milk Na and K were estimated at 0.63 g/kg and 1.5 g/kg of milk, respectively (Dairy NRC, 2001). Minerals in feces were analyzed by the University of Nebraska Soil and Plant Analytical Lab, Lincoln, NE and urine minerals were analyzed by Dairyland Laboratories, Arcadia, WI.

### **Statistical Analysis**

Regression analyses were performed using PROC MIXED of SAS (SAS, 1999) with the discrete effect of study included as a random variable (St-Pierre, 2001). Equations were developed by running multiple iterations in MIXED and removing the least significant main effect each time based on Type III sums of squares. For the LACT dataset, variables were kept if  $P < 0.10$ . Variables were kept if  $P < 0.25$  for the MINERAL dataset due to the fewer number of observations available in the dataset. Adjusted observations were calculated for graphing purposes by adding the residual from each individual observation to the predicted value of the study regression (St-Pierre, 2001).

Equation evaluation was done by regressing residuals (predicted values subtracted from observed values) on the predicted values (St-Pierre, 2003). Predicted values were centered by subtracting the mean of all predicted values from each prediction. This makes the slope and intercept estimates in the regression orthogonal and, thus, independent. Mean biases were assessed using the intercepts of the regression equations and the slopes of the regression equations were used to determine the presence of linear biases.

## **RESULTS AND DISCUSSION**

### **Urine Excretion**

Urine excretion in the LACT dataset averaged 24.1 kg/d and accounted for 35.4% of total manure excretion on a weight basis (Table 3). According to the 2001 ASAE manure excretion estimates of dairy cattle, urine excretion from a 625 kg cow would be 16.3 kg/d, which was much lower than the average  $U_E$  in the LACT dataset. Urinary excretion estimates from the 2001 ASAE standards were also less than values reported by Broderick et al. (2003), Sannes et al. (2002), and Valadares et al. (1999). Valadares et al. (1999) reported greater daily  $U_E$ , ranging from 31.5 to 50.7 kg/d across the various dietary treatments. Urine excretion ranged from 21.8 to 25.7 L/d and 20.8 to 27.3 L/d in studies by Sannes et al. (2002) and Broderick et al. (2003), respectively.

Animal and dietary factors were used as independent variables to determine significant factors for predicting  $U_E$  in the LACT dataset. Significant variables

included, DMI, NI, BW, and MUN. Days in milk, MILK, MTP, and MF were evaluated in the regression equations, but were not significant predictors of  $U_E$ . In the LACT dataset, MUN was the best individual factor for predicting  $U_E$  [1], which was similar to conclusions drawn by Jonker et al. (1998) and Kohn et al. (2002).

$$U_E = \text{MUN} \times 0.563 (\pm 0.115) + 17.1 (\pm 2.0) \quad [1]$$

$$\text{Residual SE} = 5.8; \text{Inter-study SE} = 4.7$$

Body weight and DMI were added to the MUN prediction equation to see if the additional variables improved the prediction of  $U_E$ . When BW, DMI, and MUN were evaluated, there was a slight reduction in the residual SE, from 5.8 for [1] to 5.5, but there was not any improvement in prediction across studies.

Dietary CP concentrations have been shown to affect  $U_E$ . An increase in  $C_{CP}$  from 15.1 to 18.4% of DM was associated with increased  $U_E$  of 6.5 L/d (Broderick, 2003). Similarly, Sannes et al. (2002) and Wattiaux and Karg (2004) reported greater  $U_E$  for high protein diets as compared to low protein diets. Conversely, Tomlinson et al. (1996) did not find increases in  $U_E$  with increasing  $C_{CP}$ .

Presently, few prediction equations for predicting  $U_E$  for lactating dairy cows are available in the literature. An equation [E1] was proposed by Fox et al. (2004) to predict  $U_E$  (Table 4). Evaluation of this equation resulted in significant mean and linear biases. In the LACT and VALIDATE datasets, mean bias for  $U_E$  was 3.74 (1.65) kg/d



greater than predicted by [E1]. Although the mean bias was significant, the errors between measurements were greater than the mean bias, indicating [E1] adequately predicted  $U_E$  in the LACT dataset. The differences between the observed  $U_E$  and  $U_E$  predicted using [E1] may have been caused by the lack of inclusion of terms accounting for N metabolism or Na and K intakes.

***Effect of minerals on urine excretion.*** The MINERAL dataset was used to predict the affect of mineral intakes and urinary mineral concentrations on  $U_E$ . Intakes of Na, K, and Cl were directly related to  $U_E$ . Similarly, Fisher et al. (1994) found that the level of K in the diet significantly increased urine output. In the MINERAL dataset, BW, NI, MUN, and DMI were also significant factors for predicting  $U_E$  as they were for the LACT dataset. The best equation for prediction of  $U_E$  using minerals intakes included the intake of NaI along with MUN [2].

$$U_E = (\text{NaI} \times 0.062 (\pm 0.016)) + (\text{MUN} \times 0.43 (\pm 0.21)) + 11.4 (\pm 3.8) \quad [2]$$

$$\text{Residual SE} = 5.8; \text{Inter-study SE} = 5.2$$

Accounting for the intake of Na and the N utilization efficiency, as measured by MUN, were the factors that accounted for the greatest variation within studies.

In the MINERAL dataset, there was a significant relationship between the DCAD and  $U_E$  (Figure 1). The DCAD was the best single predictor of  $U_E$  between studies in the MINERAL dataset [3].

$$U_E = (\text{DCAD} \times 0.337 (\pm 0.090)) + 13.2 (\pm 3.6) \quad [3]$$

$$\text{Residual SE} = 6.1; \text{Inter-study SE} = 3.8$$

The effect of DCAD on  $U_E$  is not surprising because urinary excretion is the main homeostatic regulation for Na, K, and Cl (Miller, 1975). Changes in DCAD have resulted in alterations in urine pH (Tucker et al., 1988). When NI, KI, and NaI were all used as predictors of  $U_E$ , only NaI was significant, indicating the NaI had a greater effect on  $U_E$  in the MINERAL dataset than KI or NI. When KI was evaluated individually in the MINERAL dataset, KI was directly related to  $U_E$  [4].

$$U_E = (\text{KI} \times 0.018 (\pm 0.011)) + 20.5 (\pm 3.8) \quad [4]$$

$$\text{Residual SE} = 6.2; \text{Inter-study SE} = 5.2$$

Fisher et al. (1994) also reported that KI had a significant affect on  $U_E$ . Similarly, Bannink et al. (1999) found that KI was significant when included with NaI.

Concentrations of minerals in urine and urinary mineral excretion were evaluated as predictors of  $U_E$ . Urine excretion in the MINERAL dataset was significantly related to concentrations of Na, K, and S in the urine as well as urinary excretion of N, K, and Na. An equation for predicting  $U_E$  included urinary excretion of N and Na, and the concentration of K in urine [5].

$$U_E = (UN \times 50.94 (\pm 13.58)) + (UNa \times 0.119 (\pm 0.024)) - (UKPct \times 22.8 (\pm 3.2)) + 19.8 (\pm 3.4) \quad [5]$$

Residual SE = 4.1; Inter-study SE = 5.2

In 1999, Bannink et al. developed an equation [E9] for predicting  $U_E$  using NaI and KI (Table 4). When [E9] was evaluated using the MINERAL dataset, there was a significant linear bias of  $-0.69$  and mean bias of  $-7.3$  kg/d. The standard deviation of the residuals from the evaluation of [E9] was 9.08, thus the mean bias was less than the expected variation between studies. The linear bias resulted in a value at the maximum (54.6 kg/d) predicted  $U_E$  of less than 23 kg/d, which is greater than the standard error of 7.8 kg/d.

Daily urinary excretions of N, Na, and K were used to predict  $U_E$  (Bannink et al., 1999). In 1999, Bannink et al. proposed [E8] (Table 4) to predict  $U_E$ . When [E8] was evaluated using the MINERAL dataset, there were significant mean and linear biases. When the mean bias was evaluated, the standard error of the residuals was greater than the mean bias. Therefore, the mean bias was less than the variation expected between studies even though it was significant. The value of the linear bias for the minimum (7.2 kg/d) predicted  $U_E$  was 8.1 kg/d, which was slightly greater than the standard error of 6.5 kg/d.

The final equation evaluated for prediction of  $U_E$  was developed by Bannink et al. (1999) and combined several factors including mineral intakes, MILK, and MTP

[E10] (Table 4). Evaluation of [E10] with the MINERAL dataset resulted in significant mean (-7.06) and linear (-0.693) biases. Similar to the previous equations evaluated, the mean bias, although significant, was less than the variation expected between studies. The linear bias resulted in a value of less than 23 kg/d at the maximum (55.3 kg/d) predicted  $U_E$ , which was outside the standard error of 8.3 kg/d.

### **Urinary Nitrogen Excretion**

Urinary N excretion averaged 49% of total manure N excretion for lactating dairy cows in the LACT dataset. Excess dietary N is excreted via urine in the form of urea. Urea is a soluble compound that will diffuse to various body fluids, including both blood and milk, in the lactating cow (Kauffman and St-Pierre, 2001; Nousiainen et al., 2004). Because excess dietary N is excreted mostly in the form of urea, MUN would be expected to have a direct affect on  $U_N$ . In addition, the concentration of MUN is directly affected by  $C_{CP}$  (Nousiainen et al., 2004).

Equations for estimating  $U_N$  were developed using the LACT dataset. Independent variables that were significant to predict  $U_N$  included BW, NI, DMI, DIM, and MUN. The best individual predictor of  $U_N$  within studies in the LACT dataset was NI. An increase in urinary N excretion has been associated with higher  $C_{CP}$  in several studies (Broderick, 2003; Sannes et al., 2002; Wattiaux and Karg, 2004). In 1998, Jonker et al. proposed an equation for predicting  $U_N$  that included NI and Nousiainen et al. (2004) found that  $C_{CP}$  was a better predictor of  $U_N$  than was MUN. Milk urea N was

also a significant predictor of  $U_N$  in the LACT dataset and would be expected to be 2.8% more accurate across studies than using NI.

The best simple linear equation for predicting  $U_N$  both within and between studies in the LACT dataset was the supply of RDP [6] as determined by the 2001 NRC model (Figure 2).

$$U_N = (\text{RDP} \times 0.0628 (\pm 0.00064)) + 55.6 (\pm 19.1) \quad [6]$$

$$\text{Residual SE} = 42.8; \text{Inter-study SE} = 35.4$$

Multiple regression equations were also evaluated for the prediction of  $U_N$  [7].

$$U_N = (\text{BW} \times 0.254 (\pm 0.039)) - (\text{MILK} \times 1.03 (\pm 0.40)) + \\ (\text{NI} \times 210.1 (\pm 27.8)) + (\text{MUN} \times 5.09 (\pm 0.787)) + \\ (\text{MTP} \times 21.8 (\pm 8.3)) - (\text{MF} \times 6.5 (\pm 3.5)) - 138.8 (\pm 44.0) \quad [7]$$

$$\text{Residual SE} = 38.6; \text{Inter-study SE} = 46.4$$

Although the multiple regression equation [7] decreased the variation within studies, the multiple linear regression equation did not significantly improve the prediction of  $U_N$  across studies.

*Evaluation of urinary nitrogen excretion equations.* Numerous equations have been proposed to predict  $U_N$  from lactating dairy cows (Table 4). Many of these previously published prediction equations have included MUN in the equation because equilibration of urea in the body results in a direct relationship to  $U_N$  (Kauffman and St-Pierre, 2001; Nousiainen et al., 2004). An additional advantage of the use of MUN as an independent variable for prediction of  $U_N$  is the ease and low cost of determining MUN. The challenge of using MUN as an independent variable is a result of the differences in the calibration and accuracy of the laboratory equipment used to determine the MUN values for many commercial dairy operations (Kohn et al., 2004).

Previously published equations for predicting UN have been evaluated by several researchers (Kauffman and St-Pierre, 2001; Kohn et al., 2002; Nousiainen et al., 2004). Besides evaluation of equations, many of these researchers proposed additional equations for prediction of  $U_N$ . Evaluation of previously published equations provides information as to the usefulness of the prediction equations in various dietary and management situations.

In 1998, Jonker et al. proposed the use of [E2] to predict  $U_N$ . In 2001, Kauffman and St-Pierre reported that the coefficient (12.54) from [E2] should be increased to 17.6 to account for an inaccuracy in the determination of the equipment used for calculation of MUN values. The adjustment in MUN values was confirmed by Kohn et al. (2002). When [E2] was evaluated using the LACT and VALIDATE datasets, there was a significant mean bias (57.3 g/d), which further confirmed the need

for an adjustment to the coefficient of the MUN term in [E2] (Jonker et al., 1998). Another equation for predicting  $U_N$  using adjusted MUN values was published by Kauffman and St-Pierre (2001). When [E4] was evaluated with the LACT dataset, there was not a significant mean bias, but there was a significant linear bias (-0.679). The linear bias resulted in a maximum bias of 176 g/d over the entire range of the predicted values.

Prediction equations for  $U_N$  were developed for both Holsteins and Jerseys by Kauffman and St-Pierre (2001). The development of prediction equations across breeds led to an adjustment for BW in the  $U_N$  prediction equation. Evaluation of [E5] did not result in a mean bias, but there was a significant linear bias of -0.532 (Figure 3). The maximum bias for [E5] was less than 121 g/d over the range of the predicted values and resulted in the least bias of the equations evaluated for prediction of  $U_N$ .

An additional equation that included NI was proposed by Jonker et al. (1998). When [E3] was evaluated using the LACT dataset, there was not a significant mean bias, but there was a significant linear bias (-0.652) that resulted in a maximum bias of 200 g/d over the range of the predicted values. Of the equations evaluated, [E3] resulted in the least amount of individual cow variation in the LACT and VALIDATE datasets. The greatest difficulty with [E3] occurred when early lactation cows were evaluated using the equation. The use of [E3] resulted in negative prediction values for early lactation animals. The negative predictions were most likely a result of the

negative N balance of these early lactation cows, but indicates that [E3] should not be used for cows in early lactating.

In 2002, Kohn et al. evaluated equations published by Jonker et al. (1998) and Kauffman and St-Pierre (2001), and proposed an additional equation for prediction of  $U_N$  [E6]. Evaluation of [E6] resulted in a significant linear bias of  $-0.626$ . Equation [E6] was comparable to [E5] for prediction of  $U_N$  in the LACT and VALIDATE datasets, with a maximum bias of 136 g/d over the full range of the data.

## Urinary Mineral Excretion

**Sodium.** Average  $U_{Na}$  in the MINERAL dataset was 84.8 g/d (Table 3), and accounted for 71.4% of total Na excretion. In the MINERAL dataset, the  $C_{Na}$  was above animal requirements due to the addition of sodium bicarbonate to the diets, thus resulting in greater  $UNaPct$  in the MINERAL dataset than values reported by Bannink et al., (1999), Shalit et al., (1991), and Tucker et al. (1988). Silanikove et al. (1997) reported that 65% of total Na excretion was via urine and Bannink et al. (1999) found  $U_{Na}$  accounted for 76% of total Na excretion.

Urinary Na excretion was directly related to  $NaI$  in the MINERAL dataset and in the study reported by Bannink et al. (1999). Equation [8] was developed to estimate  $U_{Na}$  based on  $NaI$ .



$$U_{\text{Na}} = (\text{NaI} \times 0.456 (\pm 0.072)) + 26.6 (\pm 12.1) \quad [8]$$

Residual SE = 20.0; Inter-study SE = 18.5

Urinary Na excretion has also been estimated by the difference in apparently digested Na and Na secreted in milk [E11] (Bannink et al., 1999). However, assumption of apparently digested Na to determine excretion of Na may not always be accurate because of Na recycling that occurs for ruminant animals, especially during periods of low NaI (NRC, 2001).

**Potassium.** Dietary concentrations of K in the MINERAL dataset averaged 1.34% of DM, which is lower than the  $C_K$  in many modern dairy diets because of high K levels in forages. The lower  $C_K$  in the MINERAL dataset resulted in KI that were 74% of KI reported by Bannink et al. (1999). Fisher et al. (1994) reported  $U_K$  of 209 g/d for diets containing 1.6%  $C_K$ . The lesser KI in our study resulted in ~50% of the  $U_K$  reported by Bannink et al. (1999) and lower K concentrations in urine than reported by Shalit et al. (1991). In contrast, Tucker et al. (1988) reported UKPct that were similar to concentrations in our dataset when the dietary concentration of (Na + K) – Cl equaled 10 meq/100 g of diet DM in their study.

Increased KI resulted in greater  $U_K$  in the MINERAL dataset [9].

$$U_K = (\text{KI} \times 0.451 (\pm 0.092)) + 40.2 (\pm 29.9) \quad [9]$$

Residual SE = 38.8; Inter-study SE = 41.6

In the MINERAL dataset, 75% of K excretion occurred via urinary routes. Conversely, Silanikove et al. (1997) reported that ~64% of K excreted in manure was through urinary excretion and Bannink et al. (1999) found that urinary excretion accounted for over 87% of total K excretion.

Bannink et al. (1999) proposed an equation to estimate  $U_K$  [E12] (Table 4). Evaluation of [12] using the MINERAL dataset resulted in a linear bias. The amount of apparently digested K is not an easily obtainable value in practical situations and would not be practical for use on an on-farm basis.

## **CONCLUSIONS**

Prediction of urine and urinary nutrient excretion is related closely to protein metabolism of lactating cows. Crude protein intake and MUN were directly related to both urine and urinary N excretion in our dataset. Inclusion of mineral intake parameters for the prediction of urine excretion resulted in direct relationships between intakes of Na and K and excretion of urine. In the MINERAL dataset, Na intake affected urine excretion to a greater extent than the intake of N or K. Intake of N, Na, and K directly affected urinary excretion of N, Na, and K, respectively. Evaluation and validation of prediction equations is important to develop equations that will more accurately estimate urine, urinary N, and urinary mineral excretion from lactating dairy cows.

## REFERENCES

- Bannink, A., H. Valk, and A. M. Van Vuuren. 1999. Intake and excretion of sodium, potassium, and nitrogen and the effects on urine production by lactating dairy cows. *J. Dairy Sci.* 82:1008–1018.
- Broderick, G. A. 2003. Effects of varying dietary protein and energy levels on the production of lactating dairy cows. *J. Dairy Sci.* 86:1370–1381.
- Fisher, L. J., N. Dinn, R. M. Tait, and J. A. Shelford. 1994. Effect of level of dietary potassium on the absorption and excretion of calcium and magnesium by lactating cows. *Can. J. of Anim. Sci.* 74:503–509.
- Fox, D. G., L. O. Tedeschi, T. P. Tylutki, J. B. Russell, M. E. Van Amburgh, L. E. Chase, A. N. Pell, and T. R. Overton. The Cornell Net Carbohydrate and Protein System model for evaluating herd nutrition and nutrient excretion. *Anim. Feed Sci. Technol.* 112:29-78.
- Frank, B., M. Person, and G. Gustafsson. 2002. Feeding dairy cows for decreased ammonia emission. *Livest. Prod.* 76:171–179.
- Harrison, J., P. Huhtanen, and M. Collins. 2003. Perennial grasses. Pages 665–745 in *Silage Science and Technology*. D. R. Buxton, R. E. Muck, and J. H. Harrison, ed. ASA-CSSA-SSSA, Madison, WI.
- Johnson, L. M., J. H. Harrison, and R. E. Riley. 1998. Estimation of the flow of microbial nitrogen to the duodenum using urinary uric acid or allantoin. *J. Dairy Sci.* 81:2406–2420.
- Jonker, J. S., R. A. Kohn, and R. A. Erdman. 1998. Using milk urea nitrogen to predict nitrogen excretion and utilization efficiency in lactating dairy cows. *J. Dairy Sci.* 81:2681–2692.
- Kauffman, A. J., and N. R. St-Pierre. 2001. The relationship of milk urea nitrogen to urine nitrogen excretion in Holstein and Jersey cows. *J. Dairy Sci.* 84:2284–2294.
- Kohn, R. A., K. R. French, and E. Russek-Cohen. 2004. A comparison of instruments and laboratories used to measure milk urea nitrogen in bulk-tank milk samples. *J. Dairy Sci.* 87:1848–1853.
- Kohn, R. A., K. F. Kalscheur, and E. Russek-Cohen. 2002. Evaluation of models to estimate urinary nitrogen and expected milk urea nitrogen. *J. Dairy Sci.* 85:227–233.

- Krober, T. F., D. R. Kulling, H. Menzi, F. Sutter, and M. Kreuzer. 2000. Quantitative effects of feed protein reduction and methionine on nitrogen use by cows and nitrogen emission from slurry. *J. Dairy Sci.* 83:2941–2951.
- Miller, W. J. 1975. New concepts and developments in metabolism and homeostasis of inorganic elements in dairy cattle. A review. *J. Dairy Sci.* 58:1549–1560.
- National Research Council. 2001. *Nutrient Requirements of Dairy Cattle*. 7<sup>th</sup> rev. ed. Natl. Acad. Sci., Washington, D. C.
- Natural Resources Conservation Service. 2001. *Nutrient management*. Natural Resources Conservation Service Conservation Practice Standard Code 590-1. USDA-NRCS. Washington State.
- Nousiainen, J., K. J. Shingfield, and P. Huhtanen. 2004. Evaluation of milk urea nitrogen as a diagnostic of protein feeding. *J. Dairy Sci.* 87:386-398.
- Sannes, R. A., M. A. Massman, and D. B. Vagnon. 2002. Form of rumen-degradable carbohydrate and nitrogen on microbial protein synthesis and protein efficiency of dairy cows. *J. Dairy Sci.* 85:900–908.
- SAS User's Guide: Statistics, Version 8.2. 1999. SAS Inst., Inc., Cary, NC.
- Shalit, U., E. Maltz, and N. Silanikove. 1991. Water, sodium, potassium, and chlorine metabolism of dairy cows at the onset of lactation in hot weather. *J. Dairy Sci.* 74:1874–1883.
- Silanikove, N., E. Maltz, A. Halevi, and D. Shinder. 1997. Metabolism of water, sodium, potassium, and chlorine by high yielding dairy cows at the onset of lactation. *J. Dairy Sci.* 80:949–956.
- St-Pierre, N. R. 2001. Integrating quantitative findings from multiple studies using mixed model methodology. *J. Dairy Sci.* 84:741–755.
- St-Pierre, N. R. 2003. Reassessment of biases in predicted nitrogen flows to the duodenum by NRC 2001. *J. Dairy Sci.* 86:344–350.
- Timmermans Jr., S. J., L. M. Johnson, J. H. Harrison, and D. Davidson. 2000. Estimation of the flow of microbial nitrogen to the duodenum using milk uric acid or allantoin. *J. Dairy Sci.* 83:1286–1299.
- Tomlinson, A. P., W. J. Powers, H. H. Van Horn, R. A. Nordstedt, and C. J. Wilcox. 1996. Dietary protein effects on nitrogen excretion and manure characteristics of lactating cows. *Trans. ASAE* 39:1441–1448.

Tucker, W. B., G. A. Harrison, and R. W. Hemken. 1998. Influence of dietary cation-anion balance on milk, blood, urine, and rumen fluid in lactating dairy cows. *J. Dairy Sci.* 71:346–354.

Valadares, R. F. D., G. A. Broderick, S. C. Valadares Filho, and M. K. Clayton. 1999. Effect of replacing alfalfa silage with high moisture corn on ruminal protein synthesis estimated from excretion of total purine derivatives. *J. Dairy Sci.* 82:2686–2696.

Wattiaux, M. A., and K. L. Karg. 2004. Protein level for alfalfa and corn silage-based diets: II. Nitrogen balance and manure characteristics. *J. Dairy Sci.* 87:3492–3502.

Table 1. Experiments included in the datasets used for development of urine and urinary mineral excretion equations.

Experiment	Experiment description
1	Treatments included either high or low RDP concentrations with nonstructural carbohydrates (Experiment 1; Johnson et al., 1998)
2	Treatments were designed to look at various levels of lysine and methionine available for absorption in the small intestine and included negative and positive control diets with 2 levels of supplement (Experiment 2, Johnson et al., 1998; Experiment 6, Timmermans et al., 2000)
3	Treatments were designed to look at various levels of lysine and methionine available for absorption in the small intestine and included negative and positive control diets with 1 level of supplement (Experiment 3, Johnson et al., 1998)
4	Grass silages contained either no enzymes or were treated with different levels of cellulase and xylanase enzymes (Experiment 4, Johnson et al., 1998; Experiment 5, Timmermans et al., 2000)
5	Corn silage with either no bacterial inoculant added or corn silages treated with 1 of 2 different bacterial inoculants (Experiment 5, Johnson et al., 1998; Experiment 4, Timmermans et al., 2000)
6	Corn silage harvested at either blackline or one-half milkline (Experiment 6, Johnson et al., 1998)
7	Corn silage harvested at 1/3 milkline, 2/3 milkline, or blackline both with and without mechanical processing (Experiment 1, Timmermans et al., 2000)
8	Corn silage harvested at dough stage, 1/3 or 2/3 milkline both with and without mechanical processing (Experiment 2, Timmermans et al., 2000)
9	Control diet of corn with 4 treatments of various barley hybrids (Experiment 3, Timmermans et al., 2000).
10	Corn silage contained no bacteria or enzyme and 3 treatments containing enzymes and/or bacteria (Experiment 7, Timmermans et al., 2000).
11	Corn silage harvested at 1/3 or 2/3 milkline or blackline both with and without mechanical processing (Experiment 8, Timmermans et al., 2000)
12	Corn silage harvested at 2/3 milkline at 10, 30, or 40 mm theoretical length of cut with and without mechanical processing (Experiment 9, Timmermans et al., 2000)
13	Corn silage harvested at 2/3 milkline at 30 or 40 mm theoretical length of cut with and without mechanical processing (Experiment 10, Timmermans et al., 2000)
14	Treatments included fishmeal or distillers grain plus an additional protein supplement (unpublished).
15	Dry rolled corn or high moisture corn or 2 combinations of dry rolled and high moisture corn (unpublished).
16	WSU PWP16

Table 2. Animal and production characteristics for the datasets used for development of prediction equations. Characteristics are included for datasets with all lactating cows (LACT), lactating cows for which mineral data was available (MINERAL), and cows in the validation dataset (VALIDATE).

Item	n	Mean	Minimum	Maximum	SD
<i>LACT</i>					
BW, kg	372	629	437	800	64.6
DIM	371	154	13	488	93.2
Milk, kg/d	372	32.7	1.4	86.1	11.3
DMI, kg/d	372	22.2	6.8	32.9	3.99
Milk fat, %	372	3.62	2.25	6.42	0.68
Milk protein, %	372	2.94	1.61	4.47	0.32
Dietary CP, %	372	175.8	128.9	243.8	18.7
MUN, mg/dl	372	13.3	6.0	27.0	3.48
<i>MINERAL</i>					
BW, kg	115	615	437	758	68
DIM	115	145	16	356	73.0
Milk, kg/d	115	34.1	3.8	86.1	13.8
DMI, kg/d	115	21.5	10.5	31.6	4.28
Milk fat, %	115	3.69	2.47	6.42	0.67
Milk protein, %	115	2.93	2.15	4.47	0.32
Dietary Na, %	115	0.66	0.28	1.15	0.20
Dietary K, %	115	1.36	0.58	1.86	0.23
Dietary Cl, %	115	0.42	0.25	0.60	0.09
Dietary S, %	115	0.26	0.14	0.42	0.05
DCAD, meq/kg	115	36.0	20.7	55.0	7.9
<i>VALIDATE</i>					
BW, kg	48	655	542	764	57.9
DIM	48	83	---	---	---
Milk, kg/d	48	41.2	22.9	53.3	6.0
DMI, kg/d	48	23.5	14.5	31.9	3.9
Milk fat, %	48	2.83	1.45	4.84	1.45
Milk protein, %	48	2.78	2.26	3.32	0.25
Dietary CP, %	48	17.2	16.3	18.1	0.6
MUN, mg/dl	48	11.9	7.7	15.5	2.08

Table 3. Average excretion values for animals in the datasets of lactating cows (LACT), lactating cows for which mineral data was available (MINERAL), and for the validation dataset (VALIDATE).

Item	n	Mean	Minimum	Maximum	SD
<i>LACT</i>					
Urine, kg/d	372	24.2	11.6	58.7	7.1
Urinary N, g/d	372	221.7	63.0	498.6	65.7
<i>MINERAL</i>					
Urine, kg/d	115	24.8	11.7	57.4	7.7
Urinary N, g/d	115	210.5	93.4	418.5	55.1
Urinary K, g/d	76	160.8	53.8	366.7	59.2
Urinary Na, g/d	76	84.8	22.0	161.6	33.9
<i>VALIDATE</i>					
Urine, kg/d	48	29.0	17.1	68.2	8.3
Urinary N, g/d	48	225.4	150.0	320.0	39.2



Table 4. Previously published prediction equations evaluated using either the MINERAL dataset or a combination of the LACT and the VALIDATE datasets.

No.	Equation	Source	Mean bias		Linear bias	
			Value	<i>P</i>	Value	<i>P</i>
E1	$U_E = 3.55 + 0.16 \times \text{DMI} + 6.73 \times \text{CPI} - 0.35 \times \text{Milk}$	Fox et al., 2004	3.74 (1.65)	<0.04	-0.528(0.075)	<0.001
E2	$U_N = 12.54 \times \text{MUN}$	Jonker et al., 1998	57.3 (10.8)	<0.001	-0.549 (0.070)	<0.001
E3	$U_N = 0.83 \times \text{NI} - \text{Milk N} - 97$	Jonker et al., 1998	NS	0.17	-0.652 (0.031)	<0.001
E4	$U_N = 17.64 \times \text{MUN}$	Kauffman and St-Pierre, 2001	NS	0.39	-0.679 (0.050)	<0.001
E5	$U_N = 0.0259 \times \text{BW} \times \text{MUN}$	Kauffman and St-Pierre, 2001	NS	0.45	-0.532 (0.047)	<0.001
E6	$U_N = 15.1 \times \text{MUN} + 27.8$	Kohn et al., 2002	NS	0.71	-0.626 (0.058)	<0.001
E7	$U_N = 75.18 + 0.719 \times ((\text{NI} - \text{Fecal N}) - \text{Milk N})$	Bannink et al., 1999	NS	0.38	-0.631 (0.052)	<0.001
E8	$U_E = 0.1343 \times U_{\text{Na}} + 0.0612 \times U_{\text{K}} + 0.0239 \times U_{\text{N}}$	Bannink et al., 1999	4.19 (1.36)	0.02	-0.279 (0.116)	<0.02
E9	$U_E = 0.1153 \times \text{NaI} + 0.0577 \times \text{KI}$	Bannink et al., 1999	-7.35 (1.86)	<0.01	-0.695 (0.092)	<0.001
E10	$U_E = 1.3442 + \text{DMI} \times (1.079 \times C_{\text{Na}} + 0.5380 \times C_{\text{K}} + 0.0203 \times C_{\text{CP}} - \text{MILK} \times (0.1216 + 0.0275 \times \text{MTP}))$	Bannink et al., 1999	-7.06 (2.10)	0.01	-0.693 (0.091)	<0.001
E11	$U_{\text{Na}} = 3.29 + 0.925 \times ((\text{NaI} - \text{Fecal Na}) - \text{Milk Na})$	Bannink et al., 1999	NS	0.58	-0.602 (0.087)	<0.001
E12	$U_{\text{K}} = 25.25 + 0.935 \times ((\text{KI} - \text{Fecal K}) - \text{Milk K})$	Bannink et al., 1999	NS	0.34	-0.44 (0.13)	<0.001

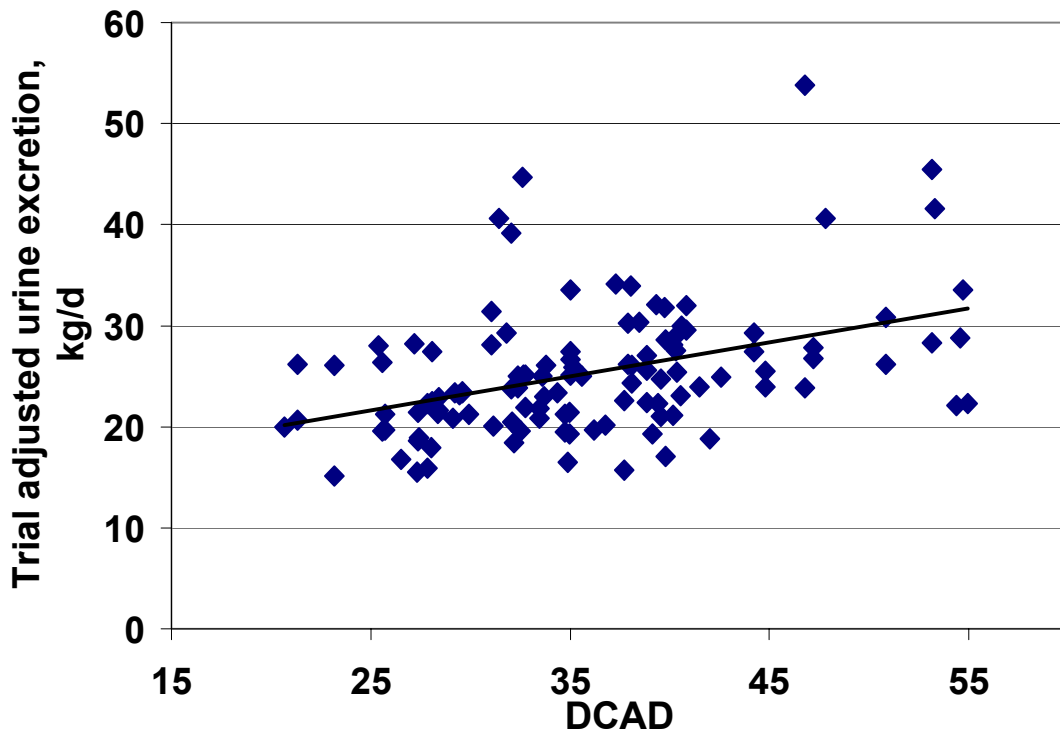


Figure 1. Relationship between the dietary cation-anion difference  $[(\text{Na} + \text{K}) - (\text{S} + \text{Cl})]$  and trial adjusted urine excretion (kg/d) for lactating cows (MINERAL dataset,  $n = 115$ ). The solid line is equal to Urine excretion (kg/d) = (DCAD, meq/kg  $\times$  0.337) + 13.2, Residual SE = 6.1, Inter-study SE = 3.8.

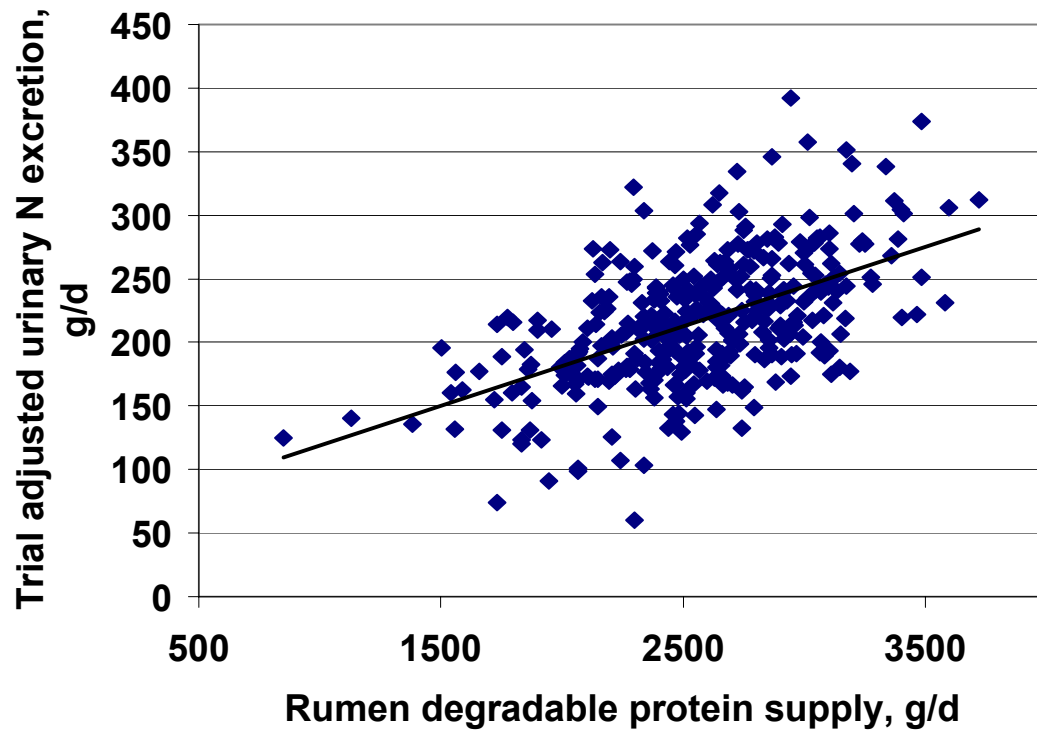


Figure 2. Relationship between the rumen degradable protein supply (g/d) as calculated by the 2001 NRC model and trial adjusted urinary N excretion (g/d) for lactating cows (LACT dataset, n = 372). The solid line is equal to Urinary N excretion (g/d) = (RDP supply, g/d x 0.0628) + 55.6, Residual SE = 42.8, Inter-study SE = 34.4.

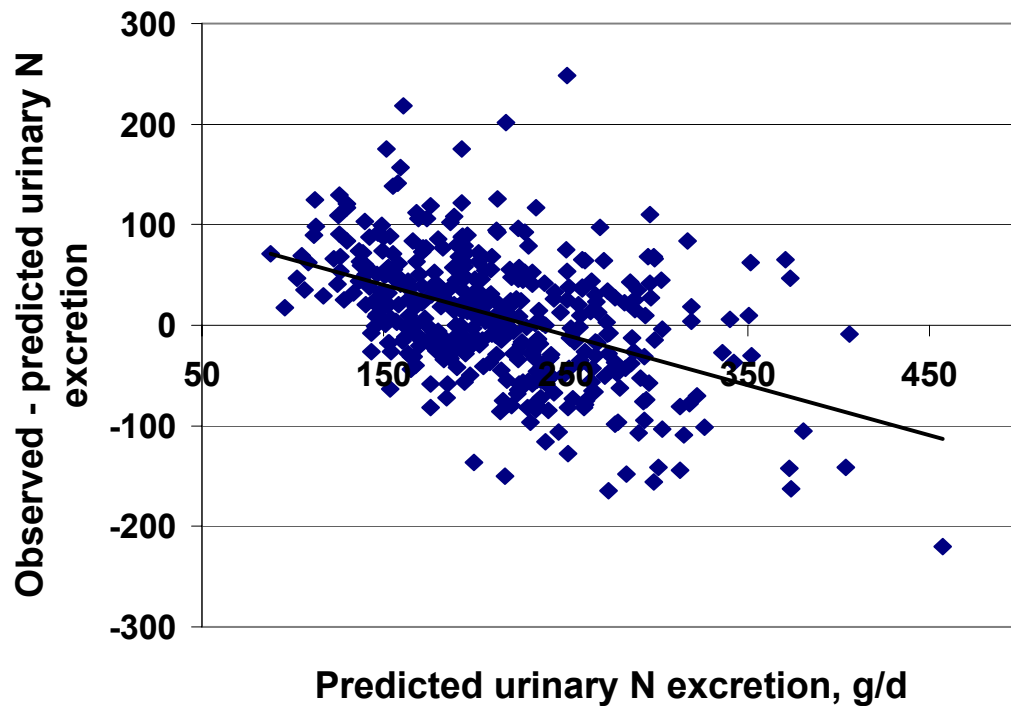


Figure 3. Plot of residuals (observed – predicted) versus predicted values of urinary N excretion (g/d) from evaluation of the equation [Urinary N, g/d = (0.0259 x BW, kg x MUN, mg/dl)] by Kauffman and St-Pierre, 2001. The equation was evaluated using a combination of the LACT and VALIDATE datasets. The solid line on the graph represents the equation  $y = 7.98(\pm 10.3) - 0.531(\pm 0.047)(X - 214)$ . Evaluation of the equation did not result in a mean bias. The linear bias was significant (-0.532;  $P < 0.01$ ).

**Winter Application of Dairy Slurry on a Grazing Based Dairy: 1.)  
Evaluation of Nitrogen Use in a Native Pasture**

**T. D. Nennich\*, J. H. Harrison\*,  
and D. L. Davidson\***

\*Puyallup Research and Extension Center,  
Washington State University, Puyallup 98371

Corresponding author:

Joseph Harrison  
7612 Pioneer Way E.  
Puyallup, WA 98371  
253/445-4638  
jhharrison@wsu.edu

## ABSTRACT

Application of manure during winter months is often discouraged due to a perceived lack of nutrient uptake by plants and increased risk of nitrogen leaching to groundwater. The objective of this study was to evaluate the effects of two rates of dairy manure application on soil nitrate levels and forage dry matter and N yields. Six plots (48.8 m by 14.6 m) in a native pasture were assigned in duplicate to one of three treatments (control, 1x, and 2x manure application rates) for a 2-yr period. Dairy slurry was surface applied to the 1x and 2x plots in January and June using a splash-plate manure applicator. Soil samples were taken on a monthly basis and grass clippings were taken once a month from April through September. No significant differences were found in soil nitrate levels among treatments or between years. In year 1, forage N yields at first cutting were greater for 2x plots (92.0 kg ha<sup>-1</sup>) than for 1x or control plots (63.1 and 55.9 kg ha<sup>-1</sup>, respectively). Cumulative N yields were 104.4, 76.3, and 64.7 kg ha<sup>-1</sup> for control, 1x, and 2x plots in year 1. In year 2, first cutting and cumulative N yields for 1x plots were greater than control plots, and 2x plot N yields were greater than both 1x and control plot yields. The results indicate winter manure application on soils with low nitrate levels increase grass N yields without increasing soil nitrate levels.

## INTRODUCTION

The importance of evaluating various management practices for field application of manure increases as environmental regulations are passed at both the state and

federal levels. Excess application of N increases leaching of nitrates through the soil profile into groundwater. Application of nutrients at rates equal to plant nutrient uptake decreases the risk of nutrient transport to groundwater or surface waters.

Applications of manure slurry during different seasons of the year has been previously evaluated (Beckwith et al., 2002; Gupta et al., 2004; Sullivan et al., 2000). Spring slurry application resulted in greater N uptake in grass than fall applications, but the effect on yield of grass DM has not always been significant (Beckwith et al., 2002). Grasses in the Pacific Northwest utilize manure N applied in the fall of the year as efficiently as spring and summer applications (Sullivan et al., 2000).

Spreading slurry during winter months is often discouraged due to increased risk of nutrient transport to surface waters or leaching through the soil profile. In many regions, the ground is frozen through the winter months, reducing incorporation of manure into the soil and further increasing the risk of runoff. However, in areas of the Pacific Northwest, the soil does not freeze during winter. Accordingly, Sullivan et al. (2000) recommended that up to one-third of manure N may be applied in the fall or late winter.

Nitrogen leaching through the soil profile can also be affected by timing of manure application. Gupta et al. (2004) found greater N leaching through soil when manure was applied in fall as compared to winter application on frozen soils. When N is applied at lower application rates, there is less likelihood of N movement into the

environment. Therefore, split applications of N fertilizer can be beneficial for environmental considerations (Bittman and Kowalenko, 2000).

Recovery of N application from dairy slurry was 12% in contrast to 63% for N applied from commercial fertilizers (Motavalli et al., 1989). The much smaller uptake of N from manure compared to commercial fertilizer sources is a result of several factors. First, approximately half of manure N is in the form of ammonia N and half is in the form of organic N. Ammonia N present in manure is susceptible to volatilization to the atmosphere. Volatilization of the ammonia N is reduced with lower temperatures and incorporation of manure into the soil. The organic N in manure must be mineralized for plant uptake to occur. Eghball et al. (2002) reported an average of 21% of the organic N in dairy manure was available during the first year of application.

Dairy producers are seeking ways to improve the management of nutrients on their operation to provide protection for the environment while maintaining low-cost handling options. Producers are encouraged to have enough storage for their manure during the winter months. However, construction of manure storage facilities can be costly and impractical for some dairy producers. The reason for the storage requirement is to prevent field application of nutrients during times of the year in which there is a perceived greater risk of nutrients moving into surface waters or leaching through the soil profile.



The Washington State Dairy Nutrient Management Act (Washington State Legislature, 1998) required nutrient management plans for every dairy operation in the state of Washington. Approval of a nutrient management plan for a low-intensity, grazing-based dairy was not granted due to a perceived lack of adequate storage. Nitrogen usage from winter slurry applications on the dairy was a primary focus of the reason for the non-approval of the nutrient management plan. It was believed that late fall and winter manure application would occur when forage could not use the N and nitrate would leach to groundwater. There was also a concern about increased transport of fecal bacteria to surface water, which was evaluated in a second study (Nennich et al., unpublished).

The objective of this study was to evaluate the utilization of N from dairy slurry during winter application on a native pasture used for hay production and grazing. Replicated plots at the dairy of concern were studied over a 2-yr period to represent typical manure management conditions of the dairy.

## **MATERIALS AND METHODS**

The study location was in Lewis County in southwestern Washington. Native pasture of predominately tall fescue (*Lolium arundinacea* Schreb.) that had not been renovated or inter-seeded for over 30 yr was used for this study. Typically, one cutting of hay is taken each spring at the beginning of the growing season, and cattle are grazed for a short period of time (<1 mo) at the end of the main forage season in August or

September. The pasture is not irrigated and rainfall during the summer is usually low. The pasture used for this study was approximately 4 ha. On the farm, there were approximately 70 ha used for management intensive grazing of dairy cattle. Unlike the pasture area used for this study, the majority of pastures used for intensive grazing were irrigated during the summer months. Average soil characteristics and nutrient concentrations of the plots at the start of the trial were: pH of 5.7, organic matter (OM) of 7.2%, 5.2 mg kg<sup>-1</sup> NO<sub>3</sub>-N, 3 mg kg<sup>-1</sup> P (Morgan), and 177 mg kg<sup>-1</sup> K.

Six plots (14.6 by 48.8 m) were assigned in duplicate to one of three treatments for a 2 yr period. The treatments included a control plot that received no slurry application, and two treatments of various application rates (Table 1). The treatments included either a 1x (0.13 acre-inches) or 2x (0.25 acre-inches) manure application rate that was achieved by adjusting tractor speed during manure application. The 1x rate was equivalent to the normal application rate for the dairy. For the purposes of the study, the 1x rate was doubled (2x rate) to determine the effects of additional slurry application.

Dairy slurry, including feces, urine, and bedding, was stored in an earthen lagoon. The lagoon was agitated with a mechanical mixer before it was pumped into a manure wagon. Dairy slurry was surface applied to plots with a splash-plate applicator two times yr<sup>-1</sup>, once in January and again in June after first the first cutting. Slurry application rates were determined by weighing each load of manure before and after application and dividing by the slurry application area.

Slurry samples were taken by obtaining a representative sample from each load of slurry applied. Nitrogen content of the slurry was estimated at the time of application using both hydrometer and Agros meters. Slurry samples were analyzed for total N and ammonia-N by Dairyland Laboratories, Inc., Arcadia, WI.

Soil samples were taken approximately every 4 wk with a 2.5 cm diameter probe at a 30-cm depth. Approximately 20 cores were taken from each plot and thoroughly mixed. Samples were analyzed for  $\text{NO}_3\text{-N}$  and  $\text{NH}_4\text{-N}$  by SoilTest Farm Consultants, Inc., Moses Lake, WA. Soil temperatures were determined at a depth of approximately 15 cm.

Glass clippings were taken once every 4 wk from April through August whenever grass growth was sufficient for sample collection. Grass clippings were not taken after cattle were allowed to graze the plot area. An additional grass clipping was taken each year approximately 2 to 3 d immediately before the plots were cut for hay to estimate yields at each cutting. Grass samples were taken using a 0.6 by 0.6 m square made of polyvinyl chloride. Grass inside the square was harvested using hand shears. Individual samples were weighed and dried at 60°C to determine dry matter (DM) harvest weights. Grass samples from each plot were composited and ground in a Wiley mill (Swedesboro, NJ) to pass through a 1-mm screen. Samples were analyzed for crude protein and  $\text{NO}_3\text{-N}$  using near infrared spectroscopy (NIR) at Cumberland Valley Analytical Services, Inc., Maugansville, MD.

Apparent N recovery (ANR) percentages were calculated using the following equation (Munoz et al. 2004; Sullivan et al., 2002):

$$\text{ANR, \%} = \frac{\text{N uptake from treatments} - \text{N uptake from control}}{\text{N applied to plots}} \times 100$$

Apparent N recovery values were calculated for total N (ANR<sub>TN</sub>). The ANR values were used to compare the efficiency of total N uptake for the 1x and 2x manure application treatments. Evaluations of ANR were also made between the N recoveries before the first cutting and N recovery through the last clipping period to determine the efficiency of N uptake between the winter and summer applications of dairy slurry.

Cumulative uptake of N was calculated by dividing the N yield of the grass by the total manure N applied to the plots. The calculations do not account for mineralization of organic N in the soil that was available for plant uptake. The cumulative N uptake was determined at each clipping of grass throughout the growing season in 2002 and 2003. The N applied at each slurry application was added to determine the uptake of N applied during the entire study period. A second calculation for cumulative N uptake was determined by accounting for expected losses of NH<sub>4</sub>-N to volatilization. Cumulative N uptake with volatilization losses were calculated with assumed losses of 20 and 80% of the applied NH<sub>4</sub>-N after winter and summer slurry applications, respectively (NRCS, 2004).

## **Statistical Analysis**

Data were analyzed using PROC MIXED of SAS (SAS, 1999). Differences in first cutting and cumulative DM and N yields were determined by using repeated measures with a compound symmetric covariance structure. Least square means were evaluated if the model was significant ( $P < 0.05$ ). There were no interactions of treatment by year ( $P > 0.10$ ) for cumulative grass DM yields, but interactions of treatment by year indicated a trend ( $P > 0.07$ ) for grass N yields.

Soil  $\text{NO}_3\text{-N}$  and  $\text{NH}_4\text{-N}$  concentrations were analyzed with repeated measures using PROC MIXED of SAS (SAS, 1999). The covariance structure used was spatial power to account for differences in timing between sampling periods. Least square means were evaluated if the model was significant ( $P < 0.05$ ). There were no interactions of treatment by year ( $P > 0.10$ ) for either soil  $\text{NO}_3\text{-N}$  or soil  $\text{NH}_4\text{-N}$  concentrations.

## **RESULTS AND DISCUSSION**

Application rates of dairy slurry applied to the plots are shown in Table 1. The application rates for the 1x plots averaged 45% of the rates for the 2x plots for the winter applications and 68% for the summer applications. The 2002 summer

application rate for the 2x plots was slightly less than the application rate for the 1x plots, which was unplanned.

Total N application rates over the 2-yr period were 270 kg ha<sup>-1</sup> for the 1x plots and 528 kg ha<sup>-1</sup> for the 2x plots. Ammonium N application rates for the 1x plots over the 2-yr period averaged 51% of the NH<sub>4</sub>-N application rates for the 2x plots. In this study, NH<sub>4</sub>-N was, on average, over 62% of the total N in manure. Conversely, Sullivan et al. (1997) found that 88% of the manure applied was in the organic N form.

In addition to NIR laboratory analyses of manure, quick tests for total N and NH<sub>4</sub>-N in manure were done using a hydrometer and Agros meter, respectively. The hydrometer determines an estimate of N concentration in manure by estimating the N concentration based on the amount of total solids in the slurry (Van Kessel et al., 1999). The hydrometer readings for the slurry in this study resulted in greater estimates of N than were found in the NIR N laboratory analysis. The large differences between the hydrometer readings and the laboratory analysis was most likely caused by a total solids concentration in the slurry that did not allow the hydrometer to float freely (Van Kessel et al., 1999). Use of a hydrometer for a quick estimate of N in slurry would be expected to be more accurate if the slurry contained less total solids or was diluted with water.

An Agros meter was used to acquire a field estimate of NH<sub>4</sub>-N present in the slurry. Over the 4 slurry applications, the Agros meter underestimated the NH<sub>4</sub>-N in manure by 13.3%. On an average individual load basis, the Agros meter estimate was

6.6 kg NH<sub>4</sub>-N ha<sup>-1</sup> less than NIR laboratory analysis, though the individual variation between loads was as great as 27.8 kg NH<sub>4</sub>-N ha<sup>-1</sup>. Most studies that have been conducted on the Agros meter to compare lab NH<sub>4</sub>-N to Agros meter NH<sub>4</sub>-N for dairy slurry have resulted in R<sup>2</sup> of ≥ 0.89, with the exception of one study with a small sample size that had an R<sup>2</sup> of 0.63 (Van Kessel et al., 1999).

## **Forage Yields**

Grass DM yields for the 2x plots did not differ from the control plots during 2002, but yields for the 2x plots were numerically 2.5 and 1.5 Mg ha<sup>-1</sup> greater than the control and 1x plots, respectively (Table 2). In 2003, significant differences in DM yields occurred between 2x and control plots. Although not statistically significant, the cumulative DM yield for 2x plots was 2.5 Mg ha<sup>-1</sup> greater than 1x plots. There was also a numerical increase of 2 Mg DM ha<sup>-1</sup> for the 1x rate as compared to the control. Increased grass DM yields with increasing slurry application rates were reported by Schmitt et al. (1999) and Sullivan et al. (2000). Yields of tall fescue were also reported to increase when application rates of biosolids were increased from 283 to 848 kg N ha<sup>-1</sup> yr<sup>-1</sup> (Cogger et al., 2001). However, Beckwith et al. (2002) did not find a significant yield difference between low and high manure application rates when N was applied at rates similar to those used in our study.

The majority of the forage growth occurred before the first cutting (30 May 2002). The results of the DM yields at the first cutting were similar to the cumulative

DM yields for the plots (Table 2) due to the lack of irrigation or rainfall during the summer months. In 2002, there were no significant differences between DM yields, though DM yields from 1x and 2x plots were numerically greater than control plots. In 2003, first cutting DM yields for 2x plots were greater ( $P < 0.05$ ) than 1x and control plots. Although they were not statistically different, the average 1x plot DM yields were  $1.9 \text{ Mg ha}^{-1}$  greater than control plots. Sullivan et al. (2000) also reported that the greatest yields were seen at the first cutting for orchardgrass plots and reed canarygrass plots (Schmitt et al., 1999).

Dry matter yields of forage declined during the summer months during each year of the study. The decline in DM yields was a result of the hot and dry weather conditions. Because the plots were not irrigated, the grass did not receive enough rainfall to maintain growth throughout the summer, resulting in declining DM and N yields as the summer progressed.

The effect of timing of slurry application on DM yields appeared to be greater for the January application as compared to the June application. In contrast, Beckwith et al. (2002) did not find any significant yield differences when slurry was applied in February as compared to October. The differences in DM yields between the different application times was most likely due to a lack of moisture for plant growth after the summer application. In addition, the hot and dry conditions that occurred after the summer applications most likely resulted in a loss of N to the atmosphere through ammonia volatilization.



All of the plots received additional nutrient application during the short grazing period; however, available nutrients limited forage DM yields for the control plots. The DM yields of grass at first cutting declined 0.3 Mg ha<sup>-1</sup> from 2002 to 2003, even though DM yields increased for the 1x and 2x plots. Declining yields for control plots were most likely a result of nutrients limiting forage growth.

The plots receiving slurry applications generally had greater concentrations of crude protein (CP) in the forage (Figure 1). The concentration of CP in the forage declined for each of the treatments as the forage matured, although the CP increase in the re-growth after the first cutting was harvested. The CP concentration of the forage declined during the dry summer period in both 2002 and 2003. In contrast, Bittman et al. (1999) found that forage N concentrations were greater in the fall than in the spring. Forage harvested from 2x plots generally had greater CP concentrations than control or 1x plots. Crude protein concentrations in the forage from 1x plots were numerically greater than control plots with the exception of the clippings that were taken immediately before first cutting.

Forage NO<sub>3</sub>-N concentrations were relatively low for all of the forage samples collected from the plots. The greatest forage NO<sub>3</sub>-N concentrations from individual plots were 0.30, 0.04, and 0.14% for the control, 1x, and 2x plots, respectively, with averages of 0.04, 0.03, and 0.04% NO<sub>3</sub>-N over both years of growth. In 2002, the concentrations of NO<sub>3</sub>-N in forages did not show any trends according to treatment.

However, in 2003 the forage  $\text{NO}_3\text{-N}$  concentrations were up to 0.05 percentage units greater at each clipping for 2x plots than control or 1x plots. Bittman et al. (1999) reported large coefficients of variation between forage  $\text{NO}_3\text{-N}$  concentrations, but generally saw an increase in  $\text{NO}_3\text{-N}$  concentrations in herbage from plots receiving fertilizer N as compared to dairy manure.

Cumulative forage N yields in the control plots numerically declined  $5.3 \text{ kg ha}^{-1}$  from 2002 to 2003 (Table 2; Figures 2 and 3). Applications of dairy slurry prior to the start of the study would have resulted in residual organic N that continued to be mineralized after the onset of the study (Eghball et al., 2002). The amount of available organic N for mineralization would decrease each year if additional slurry was not applied. Owens and Bonta (2004) reported a steady decline, from  $85$  to  $41 \text{ kg ha}^{-1}$ , in N yields from hay crops when no additional N fertilizer was applied for 4-yr.

In the 2x plots, forage N yields were significantly greater than control plots in 2002 (Table 2; Figure 2), and greater than 1x and control plots in 2003 (Table 2; Figure 3). In 2003, there was also a significant increase in forage N yields for the 1x plot as compared to the control. Cherney et al. (2002) also reported greater N removal for tall fescue with a 2x application rate of manure N as compared to a 1x rate. Similarly, N uptake of tall fescue was increased when more N was applied in the form of biosolids (Cogger et al., 2001). Nitrogen yields reported for orchardgrass were much greater than N yields of tall fescue in our study and averaged  $121$ ,  $255$ , and  $362 \text{ kg ha}^{-1}$ , respectively,

for applications of liquid dairy slurry that provided 0, 150, or 300 kg N ha<sup>-1</sup> (Kanneganti and Klausner, 1994).

### **Efficiency of Nitrogen Uptake**

The ANR<sub>TN</sub> across 1x and 2x treatments was greater ( $P < 0.08$ ) in 2003 than in 2002 (22.9 and 13.0%, respectively). In 2002, the ANR<sub>TN</sub> for 2x plots was 17.4% and 8.7% for 1x plots. In 2003, the ANR<sub>TN</sub> only varied by 2.3 percentage points between the 1x and 2x plots. In comparison, Munoz et al. (2004) reported an ANR of 16% (weighted average for 3 years) for corn when an average of 36 Mg ha<sup>-1</sup> yr<sup>-1</sup> of manure was applied. Apparent N recovery in tall fescue ranged from 28 to 40% for heat-dried and dewatered biosolids (Cogger et al., 1999).

The average ANR<sub>TN</sub> for 2x plots was significantly greater at the first cutting, with an average ANR<sub>TN</sub> over the 2 yr period of 53.4% compared to 5.8% at the end of the year. The 1x plots had average ANR<sub>TN</sub> of 16.0 and 6.1% at the first cutting and final harvest, respectively, over the 2 yr period. Similar to our study, summer application of manure with a splash-plate applicator led to lower total ANR values than spring or fall applications (Bittman et al., 1999). In contrast, Unwin et al. (1986) found greater availability of N with spring or summer applications as compared to winter applications. Mid-season N application can result in varied responses (Beckwith et al., 2002). Recovery of N was 25% in slurry applied after the first cut (Unwin et al., 1986). Beckwith et al. (2002) reported greater N uptake for February than for October manure

applications. Similarly, N recovery in forage was greater (25.5%) for the April application than for the December application, partly due to the large denitrification losses that occurred after the December application (Thompson et al., 1987).

Cumulative N removal in the forage as a proportion of the total N applied in dairy slurry was estimated for 1x and 2x plots during 2002 and 2003 (Figure 4). In our study, recovered N in forage ranged from 33.5 to 84.1% of manure N applied to plots, which was greater than the range of 28 to 70% reported by Cherney et al. (2002) for tall fescue plots receiving 1x and 2x rates of manure application. The percentage of N removal declined after the summer slurry applications for both 1x and 2x plots.

Cumulative uptake of N in forage was also determined by accounting for volatilization of  $\text{NH}_4\text{-N}$  after both the winter and summer slurry applications (Figure 4). The range of cumulative N uptake when accounting for volatilization losses ranged from 55 to 135% of applied N. Cumulative N uptakes greater than 100% indicate the contributions of N from OM present in the soil. The average OM content of the soil at the beginning of the trial was 7.2%. Summer slurry applications caused a reduction in cumulative N uptake in this study for both the 1x and 2x plots. The decreases in cumulative N uptake after the summer slurry applications were exacerbated by lack of moisture for forage growth and greater ammonia volatilization.

Volatilization of ammonia is expected to be greater during summer months. In this study, the weather after summer slurry applications were hot and dry; therefore, it

was likely that a large portion of  $\text{NH}_4\text{-N}$  volatilized after the summer application. Conversely, Thompson et al. (1987) reported that 48% of the surface applied  $\text{NH}_4\text{-N}$  was volatilized after the April application as compared to 74% after the December application.

Eghball et al. (2002) estimated that approximately 21% of the organic N would be available in the first year. A small portion of N will continue to be mineralized each consecutive year. In the second year after slurry application, up to 10% of the N was available for plant uptake (Unwin et al., 1986). Eghball et al. (2002) estimated that 14% of total N applied should be available the second year after application. Because manure had been applied to these plots on a yearly basis before the start of the trial, some residual organic N would be expected to mineralize and supply N for forage uptake. Declining levels of organic N available for mineralization are most likely a cause for the declining grass yields in the control plots during the 2 yr of the study. However, grazing cattle would be expected to provide an additional source of N for grass growth.

### **Soil Nitrate and Ammonium**

Soil  $\text{NH}_4\text{-N}$  concentrations were not different among the control, 1x, and 2x application rates (Table 3). However, there were significant differences between years, with average  $\text{NH}_4\text{-N}$  concentrations of 9.4 and 13.1  $\text{mg kg}^{-1}$  for 2002 and 2003, respectively, across treatments. The increase in soil  $\text{NH}_4\text{-N}$  appeared to result from

mineralization of soil organic matter, which was evident from the increase in  $\text{NH}_4\text{-N}$  that occurred in control plots. Although the organic matter in the manure would also be expected to mineralize, increases in  $\text{NH}_4\text{-N}$  could not have completely resulted from mineralization of manure organic matter because of the numerical increase that occurred in control plots.

Residual soil  $\text{NO}_3\text{-N}$  concentrations at the start of the trial were between 5 and  $5.5 \text{ mg kg}^{-1}$  for all plots. In 2002, there was a slight numerical increase in average soil  $\text{NO}_3\text{-N}$  levels for the 2x plots ( $1.3 \text{ mg kg}^{-1}$ ) as compared to the 1x and control plots, though the increase was not significant. The average soil  $\text{NO}_3\text{-N}$  concentration in 2003 showed a slight numerical decline of 0.3 and  $0.5 \text{ mg kg}^{-1}$  for control and 2x plots, with 1x plots showing a slight numerical increase. Soil  $\text{NO}_3\text{-N}$  did not change significantly for plots during any month of the sampling period, indicating that manure applications were not increasing residual soil  $\text{NO}_3\text{-N}$ . Similarly, Cogger et al. (1999) did not find increases in soil  $\text{NO}_3\text{-N}$  after application of biosolids. Also, Sullivan et al. (2000) did not see an increase in soil nitrates levels at the lower application rates, which were equal to N application rates in this study, but they did find an increase in soil nitrate with a greater application rate. Beckwith et al. (2002) reported that October applications of dairy slurry at the high rate ( $300 \text{ kg ha}^{-1}$  target rate) increased the soil mineral N.

Residual soil  $\text{NO}_3\text{-N}$  contents of these soils were extremely low and indicated a need for additional N fertilization for grass yields to reach their potential during the

growing season. Soil N levels in grass systems are often low due to N uptake by grasses (Sullivan et al., 2000).

Soil transformations of  $\text{NH}_4\text{-N}$  to  $\text{NO}_3\text{-N}$  will occur naturally when the temperature, moisture, and oxygen conditions are adequate. Microbial transformations of  $\text{NH}_4\text{-N}$  to  $\text{NO}_3\text{-N}$  are most efficient when the water-filled pore space of soil is between 50 and 70% and soil temperatures are between 25 to 35°C (Havlin et al., 1999). Figure 5 shows the soil  $\text{NO}_3\text{-N}$  concentrations during 2002 and 2003 and the soil temperatures during those time periods. The conversion of  $\text{NH}_4\text{-N}$  to  $\text{NO}_3\text{-N}$  in our study would be expected to be very low during the winter months due to the cold soil temperatures. Less conversion of  $\text{NH}_4\text{-N}$  to  $\text{NO}_3\text{-N}$  would reduce the risk of  $\text{NO}_3\text{-N}$  leaching because soil concentrations of  $\text{NO}_3\text{-N}$  would remain low until soil temperatures increased during the summer months (Figure 5). The increase in soil temperature also promotes forage growth, thus  $\text{NH}_4\text{-N}$  and  $\text{NO}_3\text{-N}$  are more likely to be utilized by the grass.

Excess N fertilization can result in leaching of  $\text{NO}_3\text{-N}$  into groundwater. Timing and rates of application affect leaching losses. Gupta et al. (2004) reported greatest N leaching losses occurred when manure was applied in fall. However, fall application of dairy manure at a rate around 110 kg N ha<sup>-1</sup> did not appear to increase leaching rates (Sullivan et al., 2000). In addition, leaching losses of N were negligible when cattle slurry was applied in December at a rate of 248 kg N ha<sup>-1</sup> (Thompson et al., 1987). In contrast, Decau et al. (2004) reported greater leaching losses (19.3 kg N ha<sup>-1</sup>

yr<sup>-1</sup>) in the fall than in the spring when cattle urine was applied, with applications occurring in May, July, and October, with greater amounts of <sup>15</sup>N recovered in leached water during the winter application period due to less plant uptake. Nitrogen leaching occurs naturally, as Gupta et al. (2004) found that N leaching occurred even for the control plots that did not receive manure application.

Another advantage of the greater N application rates is the reduced leaching due to increased plant water requirements (Decau et al., 2004). Greater application rates of manure have been shown to result in greater DM yields of forage (Cogger et al., 2001; Schmitt et al., 1999; Sullivan et al., 2000). In order to produce the greater yields, plants require additional water to support the increased plant growth. The increased water requirements reduce the amount of water available for leaching, thus reducing the potential of NO<sub>3</sub>-N to leach to groundwater.

## **CONCLUSIONS**

Nitrogen applied at the 1x and 2x application rates appeared to be utilized through forage uptake and indicated that N application rates were within the ability of the forage to utilize the N. After 2-yr of data collection, application of dairy slurry did not increase soil NO<sub>3</sub>-N concentrations. In addition, apparent N recovery was better for the winter manure application than for the summer application due to volatilization of NH<sub>4</sub>-N after the summer slurry application. Winter application of dairy slurry at the



applications rates used in this study did not appear to result in increased risks of N transport to the environment under the application strategy used on this dairy operation.

## REFERENCES

- Beckwith, C. P., P. J. Lewis, A. G. Chalmers, M. A. Froment, and K. A. Smith. 2002. Successive annual applications of organic manures for cut grass: short-term observations on utilization of manure nitrogen. *Grass Forage Sci.* 57:191–202.
- Bittman, S., and C. G. Kowalenko. 2000. Within-season grass herbage crude-protein- and nitrate-N concentrations as affected by rates and seasonal distribution of fertilizer nitrogen in a high yearly rainfall climate. *Can. J. Plant Sci.* 80:277–285.
- Bittman, S., C. G. Kowalenko, D. E. Hunt, and O. Schmidt. 1999. Surface-banded and broadcast dairy manure effects on tall fescue yield and nitrogen uptake. *Agron. J.* 91:826–833.
- Cherney, D. J. R., J. H. Cherney, and E. A. Mikhailova. 2002. Orchardgrass and tall fescue utilization of nitrogen from dairy manure and commercial fertilizer. *Agron. J.* 94:405–412.
- Cogger, C. G., A. I. Bary, S. C. Fransen, and D. M. Sullivan. 2001. Seven years of biosolids versus inorganic nitrogen applications to tall fescue. *J. Environ. Qual.* 30:2188–2194.
- Cogger, C. G., D. M. Sullivan, A. I. Bary, and S. C. Fransen. 1999. Nitrogen recovery from heat-dried and dewatered biosolids applied to forage grasses. *J. Environ. Qual.* 28:754–759.
- Decau, M. J., J. C. Simon, and A. Jacquet. 2004. Nitrate leaching under grassland as affected by mineral nitrogen fertilization and cattle urine. *J. Environ. Qual.* 33:637–644.
- Eghball, B., B. J. Wienhold, J. E. Gilley, and R. A. Eigenberg. 2002. Mineralization of manure nutrients. *J. Soil Water Conserv.* 57:470–473.
- Gupta, S., E. Munyankusi, J. Moncrief, F. Zvomuya, and M. Hanewall. 2004. Tillage and manure application effects on mineral nitrogen leaching from seasonally frozen soils. *J. Environ. Qual.* 33:1238–1246.

- Havlin, J. L., J. D. Beaton, S. L. Tisdale, and W. L. Nelson. 1999. Soil fertility and fertilizers: an introduction to nutrient management. 6<sup>th</sup> ed. Prentice Hall, Inc. Upper Saddle River, NJ.
- Kanneganti, V. R., and S. D. Klausner. 1994. Nitrogen recovery by orchardgrass from dairy manure applied with or without fertilizer nitrogen. *Commun. Soil Sci. Plant Anal.* 25:2771–2783.
- Motavalli, P. P., K. A. Kelling, and J. C. Converse. 1989. First-year nutrient availability from injected dairy manure. *J. Environ. Qual.* 18:180–185.
- Munoz, G. R., K. A. Kelling, J. M. Powell, and P. E. Speth. 2004. Comparison of estimates of first-year dairy manure nitrogen availability or recovery using nitrogen-15 and other techniques. *J. Environ. Qual.* 33:719–727.
- Natural Resources Conservation Service. 2004. National Engineering Handbook Part 651. Agriculture waste management field handbook. Online. Available: <http://www.wcc.nrcs.usda.gov/awm/awmfh.html>.
- Nennich, T. D., J. H. Harrison, D. L. Davidson. 2004. Winter application of dairy slurry on a grazing based dairy: 2.) Evaluation of fecal bacteria and nutrient transport to surface water. (Unpublished).
- Owens, L. B., and J. V. Bonta. 2004. Reduction of nitrate leaching with haying or grazing and omission of nitrogen fertilizer. *J. Environ. Qual.* 33:1230–1237.
- SAS User's Guide: Statistics, Version 8.2. 1999. SAS Inst., Inc., Cary, NC.
- Schmitt, M. A., M. P. Russelle, G. W. Randall, C. C. Scheaffer, L. J. Greub, and P. D. Clayton. 1999. Effect of rate, timing, and placement of liquid dairy manure on reed canarygrass yield. *J. Prod. Agric.* 12:239–243.
- Sullivan, D. M., C. G. Cogger, A. I. Barry, and S. C. Fransen. 2000. Timing of dairy manure applications to perennial grass on well drained and poorly drained soils. *J. Soil Water Conserv.* 55:147–152.
- Sullivan, D. M., S. C. Fransen, C. G. Cogger, and A. I. Bary. 1997. Biosolids and dairy manure as nitrogen sources for prairiegrass on a poorly drained soil. *J. Prod. Agric.* 10:589–596.
- Thompson, R. B., J. C. Ryden, and D. R. Lockyer. 1987. Fate of nitrogen in cattle slurry following surface application or injection to grassland. *J. Soil Sci.* 38:689–700.
- Unwin, R. J., B. F. Pain, and W. N. Whinham. 1986. The effect of rate and time of application of nitrogen in cow slurry on grass cut for silage. *Agric. Wastes* 15:253–268.

Van Kessel, J. S., R. B. Thompson, and J. B. Reeves III. 1999. Rapid on-farm analysis of manure nutrients using quick tests. *J. Prod. Agric.* 12:215–224.

Washington State Legislature. 1998. Dairy Nutrient Management. Chapter 90.64 RCW. Online. Available: <http://www.leg.wa.gov/RCW/index.cfm?fuseaction=chapterdigest&chapter=90.64>.

Table 1. Application rates and composition of dairy slurry applied to plots during 2002 and 2003.

Item	1x application rate	2x application rate
2002 winter application		
Application rate, Mg ha <sup>-1</sup>	33.8	73.4
Total N, kg ha <sup>-1</sup>	76.2	167.5
Ammonium N, kg ha <sup>-1</sup>	36.9	80.8
Hydrometer total N, kg ha <sup>-1</sup>	419.3	301.6
Agros meter ammonium N, kg ha <sup>-1</sup>	49.7	54.4
2002 summer application		
Application rate, Mg ha <sup>-1</sup>	28.9	23.2
Total N, kg ha <sup>-1</sup>	64.4	58.0
Ammonium N, kg ha <sup>-1</sup>	35.3	30.8
Hydrometer total N, kg ha <sup>-1</sup>	138.6	97.8
Agros meter ammonium N, kg ha <sup>-1</sup>	39.6	36.1
2003 winter application		
Application rate, Mg ha <sup>-1</sup>	28.0	63.5
Total N, kg ha <sup>-1</sup>	54.7	123.9
Ammonium N, kg ha <sup>-1</sup>	39.4	89.2
Hydrometer total N, kg ha <sup>-1</sup>	103.1	175.0
Agros meter ammonium N, kg ha <sup>-1</sup>	28.8	50.1
2003 summer application		
Application rate, Mg ha <sup>-1</sup>	24.9	55.8
Total N, kg ha <sup>-1</sup>	74.2	178.5
Ammonium N, kg ha <sup>-1</sup>	56.4	130.3
Hydrometer total N, kg ha <sup>-1</sup>	142.4	260.7
Agros meter ammonium N, kg ha <sup>-1</sup>	54.8	119.4

Table 2. Total cumulative yields of dry matter and nitrogen for plots receiving no slurry (control), 1x, or 2x application rates.

Item	control	1x rate	2x rate	SE
First cutting				
Dry matter yield, Mg ha <sup>-1</sup>				
2002	3.57	4.32	5.83	0.55
2003	3.24 <sup>b</sup>	5.14 <sup>b</sup>	7.55 <sup>a</sup>	0.55
Average	3.40 <sup>b</sup>	4.73 <sup>ab</sup>	6.69 <sup>a</sup>	0.50
Nitrogen yield, kg ha <sup>-1</sup>				
2002	55.9 <sup>b</sup>	63.1 <sup>b</sup>	92.0 <sup>a</sup>	5.11
2003	50.7 <sup>c</sup>	78.4 <sup>b</sup>	120.2 <sup>a</sup>	5.11
Average	53.3 <sup>c</sup>	70.8 <sup>bc</sup>	106.1 <sup>a</sup>	6.09
Apparent nitrogen recovery, %				
2002	---	10.5	21.5	6.79
2003	---	51.1	55.7	4.62
Cumulative over season				
Dry matter yield, Mg ha <sup>-1</sup>				
2002	4.53	5.51	7.00	0.67
2003	4.13 <sup>b</sup>	6.19 <sup>ab</sup>	8.66 <sup>a</sup>	0.67
Average	4.33 <sup>b</sup>	5.85 <sup>ab</sup>	7.83 <sup>a</sup>	0.60
Nitrogen yield, kg ha <sup>-1</sup>				
2002	64.7 <sup>b</sup>	76.3 <sup>ab</sup>	104.4 <sup>a</sup>	7.41
2003	59.4 <sup>c</sup>	92.1 <sup>b</sup>	136.1 <sup>a</sup>	7.41
Average	62.0 <sup>c</sup>	84.2 <sup>bc</sup>	120.3 <sup>a</sup>	6.52
Apparent nitrogen recovery, %				
2002	---	8.7	17.4	5.23
2003	---	21.8	24.1	4.15

<sup>abc</sup>Superscripts within rows differ (P < 0.05).

Table 3. Soil nitrate N and ammonia N for plots receiving no slurry (control), 1x, or 2x application rates.

Item	control	1x rate	2x rate	Yearly Average
Nitrate N, mg kg <sup>-1</sup>				
2002	5.3	5.3	6.5	5.7
2003	5.0	5.6	6.0	5.5
Average	5.2	5.5	6.3	---
Ammonium N, mg kg <sup>-1</sup>				
2002	8.4	9.2	10.5	9.4 <sup>b</sup>
2003	11.4	13.0	14.8	13.1 <sup>a</sup>
Average	9.9	11.1	12.7	---

<sup>abc</sup>Superscripts within columns differ (P < 0.06).

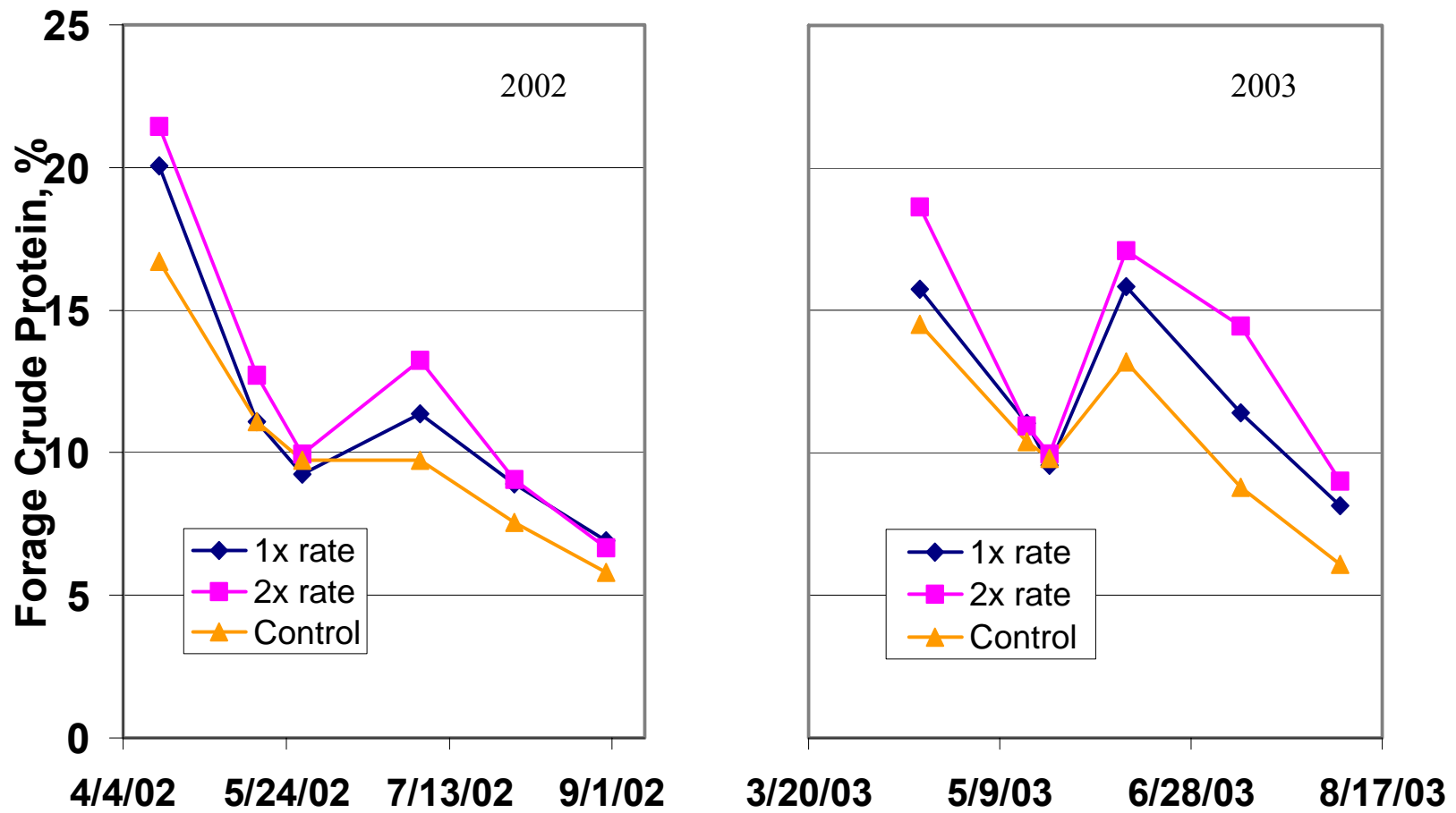


Figure 1. Crude protein concentration of harvested foraged from each clipping taken throughout the growing season. One cutting of grass was taken from each plot during the growing season (30 May 2002 and 23 May 2003).

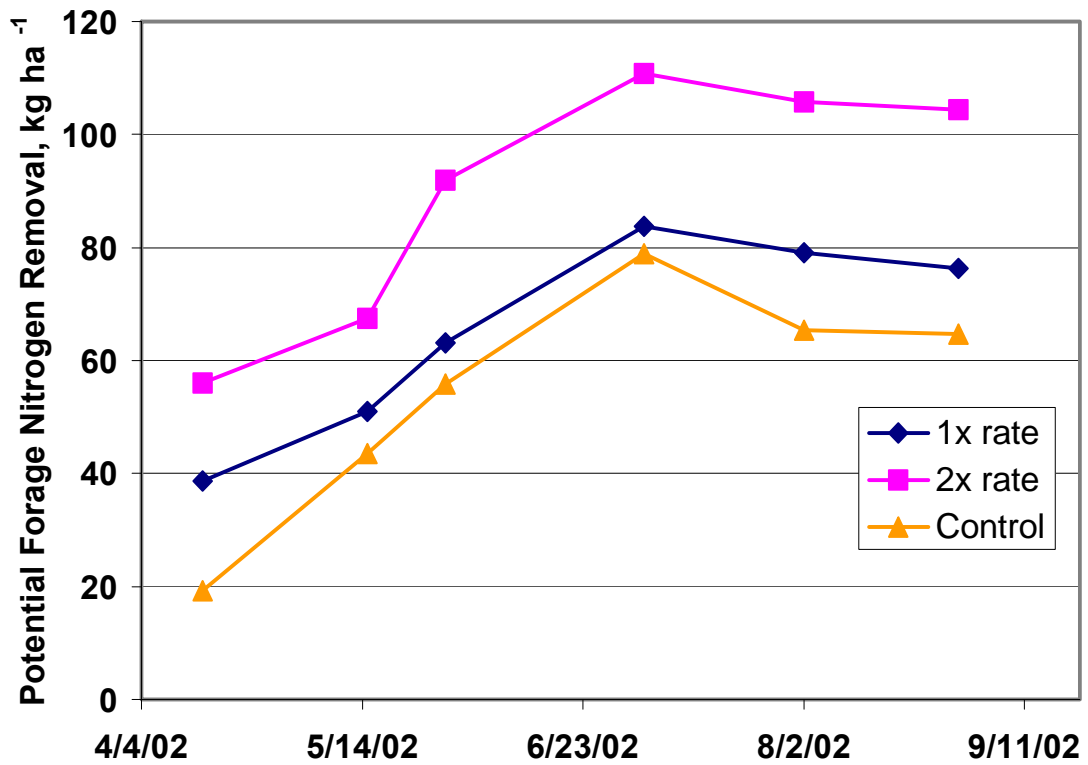


Figure 2. Potential forage N removal at each grass clipping for plots receiving no slurry (control), 1x, or 2x slurry application rates in 2002. Slurry was applied to 1x and 2x plots on 15 Jan. 2002 and on 7 June 2002. The plots were harvested for hay on 30 May 2002.



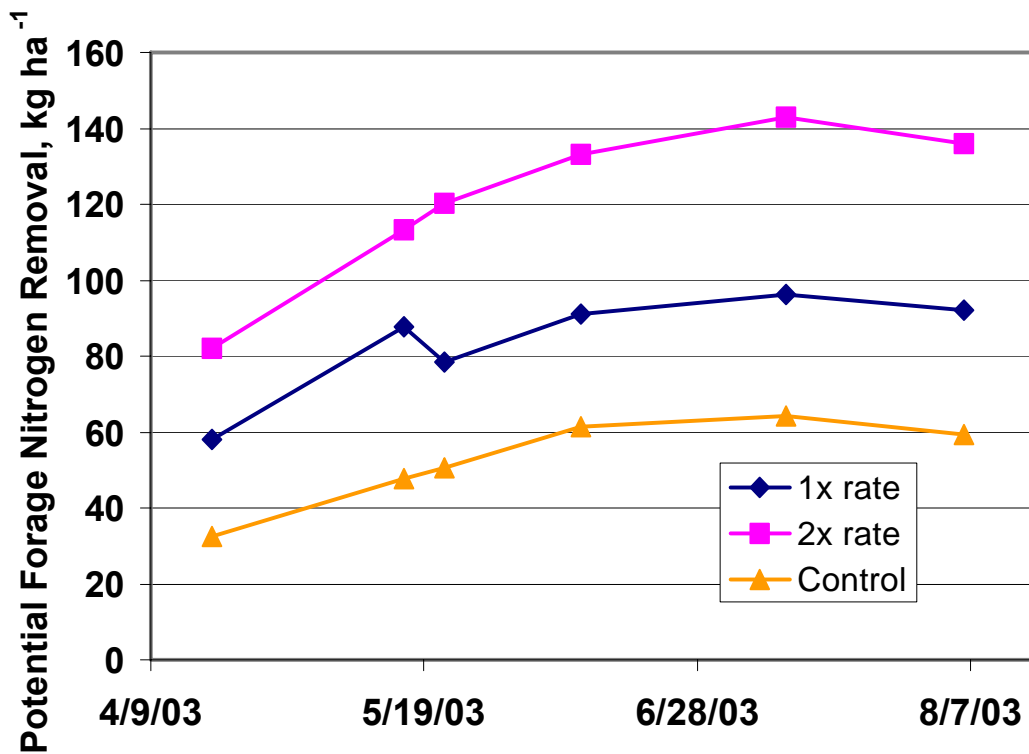


Figure 3. Potential forage N removal at each grass clipping for plots receiving no slurry (control), 1x, or 2x slurry application rates in 2003. Slurry was applied to the 1x and 2x plots on 10 Jan. 2003 and on 11 June 2003. The plots were harvested for hay on 23 May 2003.

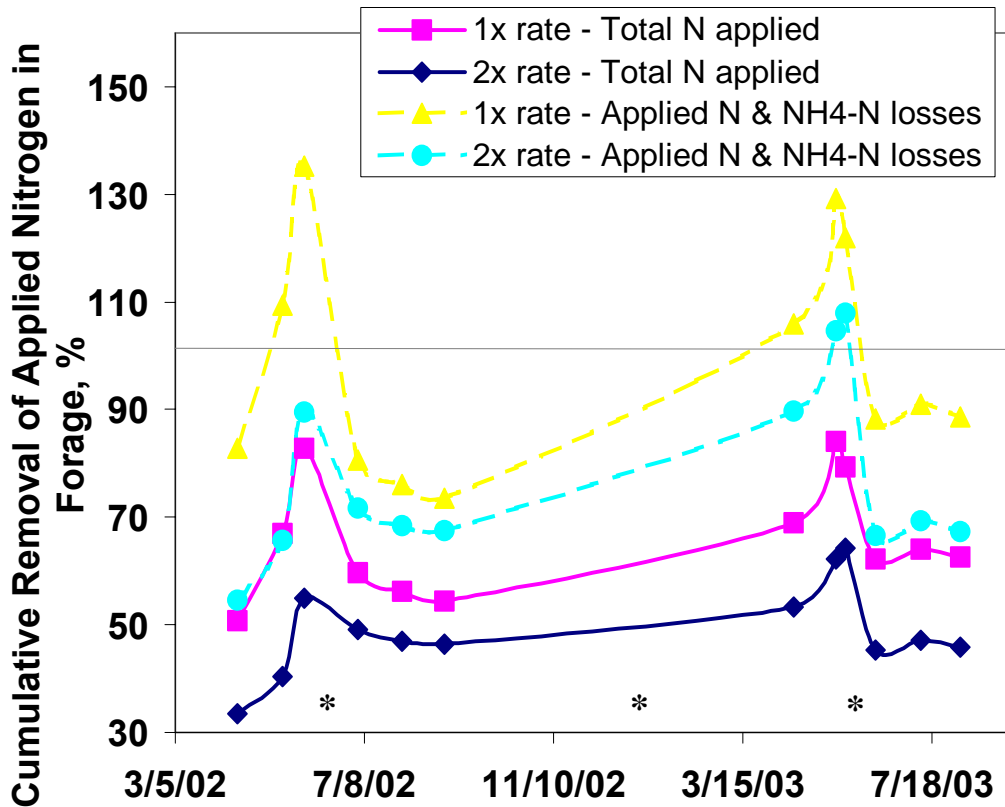


Figure 4. Cumulative removal of nitrogen in forage compared to the amount of nitrogen in dairy slurry applied to the plots in 2002 and 2003. The solid lines indicate the removal of N compared to the total amount of nitrogen in the applied slurry. The dashed lines indicate removal of N compared to nitrogen applications assuming 20% loss of NH<sub>4</sub>-N after the winter slurry applications and 80% loss of NH<sub>4</sub>-N after the summer slurry applications. Applications of dairy slurry, indicated on the graph with a \*, occurred on 15 Jan. 2002, 7 June 2002, 10 Jan. 2003, and 11 June 2003.

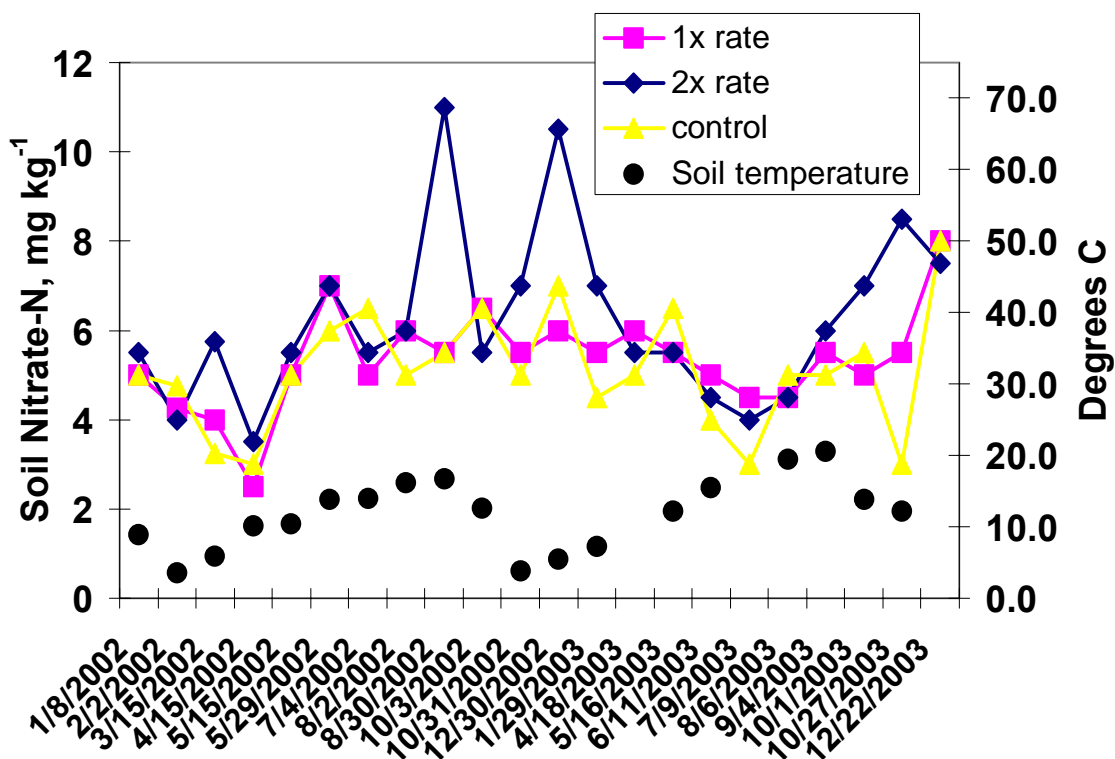


Figure 5. Average soil NO<sub>3</sub>-N concentrations during 2002 and 2003 for the control, 1x, and 2x plots. Average soil temperatures during the year are indicated on the graph with the (●).

**Winter Application of Dairy Slurry on a Grazing Based Dairy: 2.)  
Evaluation of Fecal Bacteria and Nutrient Transport to Surface  
Water**

**T. D. Nennich\*, J. H. Harrison\*,  
and D. L. Davidson\***

\*Puyallup Research and Extension Center,  
Washington State University, Puyallup 98371

Corresponding author:

Joseph Harrison  
7612 Pioneer Way E.  
Puyallup, WA 98371  
253/445-4638  
jhharrison@wsu.edu

## ABSTRACT

Application of dairy slurry during winter months has been assumed to increase the risk of nutrient and bacterial movement to the environment. The environmental risk of spreading dairy slurry during winter months, as measured by fecal coliform and nutrient levels in runoff water, was evaluated on a grazing based dairy in southwestern Washington. Dairy slurry was applied to pastureland in December 2003 and January 2004 by broadcast manure applicator to pasture areas approximately 3 to 4 times greater than routine farm practice. Sites along a grass waterway draining the farm property and surrounding areas were selected to collect water samples. Background levels of fecal coliform and *Escherichia coli* were monitored in soil and runoff water prior to slurry application. Soil samples were taken from a plot in the setback area or from two plots in the slurry application area. Water and soil samples were taken on a daily basis after slurry application and on a weekly basis for several weeks following application. Rainfall events of 2.5 cm or greater occurred within the first 48 h after each slurry application. Bacteria, N, and P concentrations increased 2 or 3 d after application at the water sampling sites closest to the slurry application area, but concentrations did not increase at downstream sites in the grass waterway. The utilization of best management practices, including limited nutrient application and use of setback zones, decreased the risk of N, P, fecal coliform, and *E. coli* movement to the environment when dairy slurry was applied during winter months.

**(Key words:** slurry, fecal coliform, nitrogen)

**Abbreviation key:** BST = Bacterial source tracking.

## INTRODUCTION

Fecal coliforms, which include *Escherichia coli*, enter the environment through animal and human wastes. Animal feces have been shown to have high concentrations of *E. coli*, with up to  $1 \times 10^9$  CFU of *E. coli* gram<sup>-1</sup> of wet feces (Mawdsley et al., 1995). Because these wastes are usually deposited on land, there is potential for fecal coliforms and *E. coli* to be transported to surface waters via runoff. *Escherichia coli* 0157:H7 has been shown to result in severe diarrhea and kidney damage in humans. Therefore, the presence of *E. coli* in water is a potential health risk to humans.

The survival of *E. coli* in the environment is controversial. The EPA document, Pathogens in Agricultural Watersheds (Rosen et al., 2000), states that *E. coli* can live in slurry for over 300 d, in soils for over 200 d, and in water for 35 d. Temperatures also play a role in bacteria survival, as longer survival times occur with colder temperatures. However, Mawdsley et al. (1995) cites studies that give survival rates of *E. coli* up to 11 weeks in slurry and survival of 7 to 8 d to a few weeks in soil.

The containment of *E. coli* in the environment through the use of buffer strips has not always been effective. Nunez-Delgado et al. (2002) reported that buffer strip length did not consistently remove fecal coliform bacteria. In the same study, soils beneath plots that were or were not fertilized with manures showed the presence of fecal coliform, indicating that these bacteria can migrate through soil (Nunez-Delgado et al., 2002).

The presence of fecal bacteria in surface water often results from numerous warm-blooded animal sources. Several methods have been explored to identify the sources of *E. coli* including DNA ribotyping and microbial resistance (Carson et al, 2001; Dombek, et al., 2000; Frantz et al. 2003). Due to the possibility of incorrect classification of the source of *E. coli* isolates, the validity of these methods for bacterial source tracking (BST) has been questioned. Frantz et al. (2003) found 77 and 97% of fecal streptococci from pigeon and cows, respectively, were correctly identified using antimicrobial resistance methods. When DNA ribotyping is used as a method for source identification of *E. coli*, the method of ribotyping used can affect the accuracy of the identification of the source of the isolate (Dombek et al., 2000). Dombek et al. (2000) found that accuracy of source identification ranged from 65 to 100% for cattle isolates depending on the fingerprints used. Similarly, when isolates from three or fewer sources were identified, greater than 89% of the sources were correctly identified and 97.1% of the ribotypes were correctly identified when human and non-human sources were differentiated (Carson et al., 2001).

Movement of N and P into surface waters is a concern because these nutrients promote eutrophication in surface waters. In particular, P is an environmental concern because it accelerates surface water eutrophication (Sharpley et al., 2003). Eutrophication causes algal blooms that decay and consume dissolved oxygen in the water, resulting in restricted water use for recreation, fisheries, and drinking water (Sharpley et al., 2003). Nitrogen is a major environmental concern because of its

potential effects on groundwater quality. Field application of N in excess of plant uptake can lead to leaching and contamination of groundwater.

The Washington State Dairy Nutrient Management Act (Washington State Legislature, 1998) required nutrient management plans for each dairy operation in the state of Washington. Approval of a nutrient management plan for a low-intensity, grazing-based dairy was not granted due to a lack of adequate storage and the perceived risks associated with late fall and winter manure applications. The non-approval of the nutrient management plan was due to the perceived risks of movement of fecal bacteria to surface waters and nitrate leaching to groundwater. In Washington State, there is a fecal coliform bacteria standard of 200 CFU 100 ml<sup>-1</sup> for secondary use recreational waters (WSDOE, 2003).

The dairy selected for this study was already using best management practices (BMP) to prevent movement of nutrients and/or bacteria into the environment. The BMP in use on the dairy included setback zones between application areas and waterways, low application rates, and applying during periods when the weather prediction for rainfall was low. The objective of this study was to confirm the value of the BMP already in use on the dairy and to determine the environmental risk of fecal coliform, *E. coli*, N, and P transport to surface waters resulting from spreading dairy slurry during winter months on a transitional-organic, grazing based dairy in southwestern Washington. This study is a companion to a previous study focused on



nitrogen usage from dairy slurry applied during winter months (Nennich et al., unpublished).

## MATERIALS AND METHODS

### Site Location and Sampling

The sampling site was located on a transitional-organic, grazing based dairy in Lewis County in Southwest Washington (Figure 1). The fields utilized for this study were native pastures of predominately Tall Fescue (*Lolium arundinacea* Schreb.) used for a combination of harvested forage and cattle grazing. A grass waterway drains the fields during the winter months, from approximately October through March. The grass waterway borders the west and east sides of a pasture that contained about 30 head of grazing beef cattle throughout the experimental period. The waterway drains hayfields, both continuously and intensively grazed pastures, and forested areas. Average rainfall for this location is approximately 190 cm yr<sup>-1</sup>, with nearly 80% of the rainfall occurring from October through April.

Daily air temperature, rainfall, and soil temperatures were monitored with a GroWeather<sup>®</sup> weather station (Davis Instruments Co., Hayward, CA) located on the farmstead. The weather station recorded air temperature, soil temperature, and rainfall data in 30-minute intervals. Rainfall during the sampling period is listed in Figure 2.

## **Sample Collection**

Water samples for fecal coliform and *E. coli* analysis were collected into sterile 250 ml plastic bottles with lids. Duplicate samples for nutrient analyses were taken in 250 ml plastic bottles and frozen for later analyses. Replicate samples at each site, along with duplicates for nutrient analyses, were taken in 5-minute intervals. Samples taken at sites C and D (Figure 1) were collected into sterile 250 ml plastic bottles by securing bottles to an extendable sampling pole.

Soil samples were taken using a 6-cm diameter soil probe at a depth of 3.8 cm. Three soil cores were taken from each plot and included grass and surface material. Each core was divided into 2 parts and placed into separate sterile sample bags. One set of cores was used for microbiological analyses and the other set was frozen. The soil probe was cleaned and sterilized between plots with a 90% isopropyl alcohol solution.

## **December Application**

Three soil plots (148.6 m<sup>2</sup>), located 30.5 m from each other, were designated for soil sampling. Two of the soil plots (X and Y) were located in the slurry application area and one plot (Z) was located in the setback zone between the application area and the grass waterway (Figure 1). The soil plots were located in a native pasture used for management intensive rotational grazing of dairy cattle. The pasture was irrigated after grazing periods during the summer months to promote forage growth. Cattle had been

removed from the pasture for over a month before any water or soil background samples were taken.

Background water samples were taken on 27 Oct. and 25 Nov. 2003.

Background soil and water samples were taken on 2 Dec, and 8 Dec. 2003. Dairy slurry was surface applied to a 1.16 ha area of pasture on 12 Dec. 2003 using a splash-plate manure applicator. Slurry was applied at a rate of  $0.036 \text{ kg (m}^2\text{)}^{-1}$ , covering an area four to five times greater than normal daily slurry applications for this dairy. A 10.6 m grass setback zone was left between the slurry application area and the grass waterway.

Water and soil samples were taken on a daily basis from 13 to 19 Dec. and on a weekly basis from 21 Dec. 2003 to 26 Jan. 2004. Three water samples were taken at 5-minute intervals for two consecutive hours, resulting in a total of six samples per site each day.

Water samples were taken from four sampling sites in the grass waterway. Site A was located 10.6 m from the point of slurry application. Site B was located 216 m further downstream and drained a larger portion of the property. Sites C and D (287 m downstream from site B) were located at the property boundary. Site C included a waterway draining the western half of the property and site D included drainage water from the entire property as well as a forested areas to the east of the farm property (Figure 1). Samples at sites A and B were taken simultaneously.

## **January Application**

The January slurry application was located on an area of pasture upstream from the first slurry application area. Sampling sites A, C, and D were sampled throughout the January sample collection period. In addition, sites E and F (Figure 1) were sampled to monitor runoff from the second application area. Site F was located upstream from the slurry application area and was 152 m from site E (Figure 1). Site E was located 19.8 m from the point of slurry application and was 111 m from site A. Background samples from the sites E and F were taken on 26 Jan. 2004.

Dairy slurry was surface applied on 27 Jan. 2004 using a splash-plate manure applicator. Slurry was applied to a 0.49 ha area at a rate of  $0.039 \text{ kg (m}^2\text{)}^{-1}$ , which covered an area two to three times greater than a normal daily slurry application for this dairy. Water and soil samples were taken on a daily basis from 28 Jan. to 31 Jan. 2004 and on a weekly basis from 2 Feb. to 9 Mar. 2004.

Soil samples were collected from three  $37.2 \text{ m}^2$  plots located 12.2 m apart (Figure 1). Plots M and N were in the slurry application area, whereas Plot O was in the grass setback zone, which was 19.7 m wide, between the slurry application area and the grass waterway.

## **Laboratory Analysis**

Water, soil, and slurry samples were analyzed for fecal coliforms within 6 h of sample collection. Slurry and water samples were diluted, if necessary, to achieve 20 to 60 CFU on each petri dish. Diluted samples were membrane filtered according to Clesceri et al., 1998. After filtration, samples were incubated at 44.5°C for 24±2 h. Bluish colonies were counted as fecal coliform CFU. After counting individual samples, plates containing 20 to 60 fecal coliform CFU were transferred to petri dishes containing Nutrient Agar with MUG and incubated at 35°C for 4 h. Colonies that fluoresced when held under a 366 nm light were counted as *E. coli*.

Soil samples were diluted in a 1:1 ratio of milliliters sterile buffer water to grams of wet soil. The soil and buffer solution mixture were placed into a stomacher for 1 minute at 200 rpm. Samples were allowed to settle for approximately 15 min, after which 5 ml of solution was pipetted from the top of the sample. Samples containing soil particulates were pre-filtered using a 2.5 µm filter. After pre-filtering, soil samples were analyzed in the same manner as outlined above. Soil samples from background sampling were retained for DNA fingerprinting.

***Nutrient analyses.*** Duplicate water samples taken during the first hour of sampling were analyzed for total nitrogen (TN), nitrate-N (NO<sub>3</sub>-N), ammonium-N (NH<sub>4</sub>-N), inorganic P, and total P (TP) at the Soil and Plant Analytical Laboratory, University of Nebraska, Lincoln, NE.

## **Bacterial Source Tracking**

Slurry samples from the 12 Dec. 2003 slurry application were retained for DNA fingerprinting (Institute for Environmental Health, Seattle, WA). Bacteria source tracking (BST) was conducted on 2 to 4 CFU from soil samples taken on 8 Dec. 2003 and on individual water samples taken from sites A, B, C, and/or D on 8, 14, 15, 21, and 30 Dec. 2003 and 8 and 11 Jan. 2004.

Plates incubated and counted for fecal coliform and *E. coli* were selected for DNA ribotyping. For isolation of the colonies, colonies appearing to be *E. coli* were selected and streaked on MacConkey agar and confirmed as *E. coli* bacteria. Fragments of DNA were isolated and restriction endonuclease digestions were done. Samples were run on a 0.8% agarose gel and underwent southern blot hybridization.

Isolates of *E. coli* were cross-referenced with a library of DNA sequences at the Institute of Environmental Health, Seattle, WA and were classified as being from cattle or unknown sources. Isolates classified as being from cattle were not specific to the slurry application and included both beef cattle and dairy cattle sources that could have been from applied slurry or from beef cattle grazing in nearby pastures.

## **RESULTS AND DISCUSSION**

### **Slurry Application and Sampling**

The January slurry application was applied at a slightly greater rate (0.039 kg (m<sup>2</sup>)<sup>-1</sup>) to 0.49 ha as compared to the December application that covered 1.16 ha at a

rate of  $0.036 \text{ kg (m}^2\text{)}^{-1}$ . The fecal bacteria concentrations in the December slurry averaged 10.4 million CFU fecal coliform  $100 \text{ ml}^{-1}$  and 9.74 million CFU *E. coli*  $100 \text{ ml}^{-1}$  (Table 1). Fecal bacteria levels in the slurry were lower for the January application, with only 3.27 CFU of both fecal coliform and *E. coli*  $100 \text{ wet grams}^{-1}$  of slurry. Frantz et al. (2003) also reported greater concentrations of *E. coli* in dairy wastewater in the fall of the year (7.08 million CFU  $100 \text{ ml}^{-1}$ ) and lower concentrations (25,000 CFU *E. coli*  $100 \text{ ml}^{-1}$ ) during the winter months. Concentrations of bacteria in the slurry were  $10^4$  to  $10^6 \text{ g}^{-1}$  less than values reported from manure directly excreted from cattle (Mawdsley et al., 1995). *Escherichia coli* O157:H7 concentrations have been reported to decline in stored cattle slurry (Jones, 1999; Kudva et al., 1998).

## **Rainfall**

The 25-yr, 24 h rainfall event for this location is approximately 15 cm. Cumulative rainfall during the study period is shown in Figure 2. Approximately 0.10 cm of rainfall occurred within the first 7 h after the application. A steady rainfall, averaging  $0.06 \text{ cm of rain hr}^{-1}$ , started approximately 14 h after the December slurry application and continued for approximately 42 h. Rainfall totaled 1.2 cm in the first 24 h and 2.8 cm in the first 48 h after slurry application.

Rainfall events after the January slurry application followed a similar pattern as the December application with a steady rainfall event, averaging  $0.10 \text{ cm of rain hr}^{-1}$ , starting approximately 12 h after application. Post-application rainfall events in January

exceeded those of December, with 0.7, 2.7, and 5.6 cm, respectively, falling within the first 24, 48, and 72 h after slurry application.

Rainfall events post-application were greater than expected. The high rainfall led to unusually large runoff events from surfaces applied with dairy slurry. On Day 2 after the December application and on Day 3 after application in January, the high rainfall events exceeded the filtering capacity of the setback zone and led to movement of slurry-laden runoff water directly into surface water, which resulted in concentrations of fecal coliform approximately 25 and 5 times greater than the Washington State fecal coliform standard for secondary recreational waters (WSDOE, 2003) at sites A and E, respectively. The fecal bacteria and nutrients that entered the grass waterway with the high rainfall events appeared to be contained by the vegetative growth in the waterway and did not move to downstream sampling sites.

### **Fecal Bacteria on Soil**

Background levels of fecal coliform and *E. coli* were very low in the soils before slurry application, with less than 10 CFU g wet soil<sup>-1</sup> in the soils before the December slurry application (Figure 3) and less than 1 CFU g wet soil<sup>-1</sup> before the January application (Figure 4). These background concentrations of fecal coliform were less than reported by Avery et al. (2004), but were similar to values reported by Stoddard et al. (1998) for soils during the month of November.



As expected, fecal coliform levels increased in plots located in the application area immediately after slurry application in both December and January. Daily soil samples taken after slurry applications indicated an increase in fecal bacteria concentrations occurred 4 to 6 d following the December slurry application and 3 to 4 d after the January application (Figures 3 and 4). Similarly, Himathongkham et al. (1999) reported that an increase in *E. coli* O157:H7 in cattle manure were seen in the first three days after the initiation of the experiment, followed by a decline in bacterial numbers. The reason for the increase in fecal coliform and *E. coli* levels after these applications is not completely known, though it is likely that the environmental conditions allowed the bacteria to come out of dormancy for a short period of time before substantial die-off occurred.

Fecal bacteria concentrations declined more than 70% in the first ten days after both slurry applications (Figures 3 and 4). Avery et al. (2004) reported 1 log declines in *E. coli* in the soil of cattle pens every 38 d. In our study, fecal coliform concentrations returned to background levels 52 and 42 d, respectively, after the December and January slurry applications. The background levels were probably reached more quickly during the January application due to the lower concentration of bacteria in the slurry applied to the pasture.

Survivability of *E. coli* in soil has been a point of debate among researchers. Avery et al. (2004) found that *E. coli* survived up to 190 d in the soil of a pen where cattle were held for 14 d. Lenehan et al. (2004) reported that *E. coli* levels returned to

background concentrations 3 mo after cattle were removed from the feeding area in which they were housed for 3 mo. In contrast, die-off time for fecal coliform bacteria on soil surfaces was less than 60 d (Stoddard et al., 1998), which more closely reflected the die-off times of bacteria seen after the slurry applications in our study. Similarly, in 2002 Jiang et al. found that *E. coli* O157:H7 survived less than 63 d in soils at 5°C. Other researchers (Himathongkham et al., 1999; Kudva et al., 1998) have noted that dehydration processes lead to decreased bacterial numbers, although Jiang et al. (2002) reported that bacteria survived in soil with 1% moisture. Conversely, the roots of rye and other grasses were found to contribute to longer survivability of *E. coli* O157:H7 (Gagliardi and Karns, 2000), which could have contributed to increased survivability of *E. coli* in our study.

Temperature plays a role in the survivability of *E. coli* on soils. Jiang et al. (2002) reported shorter survival times for *E. coli* O157:H7 at 5°C than at 15 or 21°C, though these results were in contrast to previous studies (Himathongkham et al., 1999; Jones, 1999) where lower temperatures increased *E. coli* survival time. Stoddard et al. (1998) saw the most rapid decline in fecal coliform concentrations after freezing conditions. However, freezing does not always kill *E. coli* bacteria, as Gagliardi and Karns (2002) found viable *E. coli* O157:H7 in soil samples that had been frozen. The freezing temperatures that occurred during after the December application did not appear to affect the die-off rate of the *E. coli* on the soils. Freezing air temperatures may have been a factor in the relatively short livability of the fecal bacteria in the soil of

this study. However, other contributing factors, such as competition with other soil organisms, have been reported to hasten die-off rates (Jiang et al., 2002).

The plots in the December application appeared to be located too far apart to detect any movement of bacteria from plots X or Y to the plot Z in the setback zone. However, during the January application there was some movement of bacteria across the slope of the land into the setback zone (plot O). During the January application, plot O, which was located in the setback zone, showed an increase in fecal coliform concentrations from less than 20 CFU 100 g wet soil<sup>-1</sup> to over 1500 CFU 100 g wet soil<sup>-1</sup> on Day 3 after the slurry was applied (Figure 4). The increase in soil bacteria levels on Day 3 coincided with the increased fecal bacteria levels found on Day 3 in the surface water at site E, indicating that bacteria was moving with the runoff water. The movement of runoff water was directly related to the 3.71 cm of rainfall that was received in the 72 h after slurry application.

### **Bacteria in Surface Water**

Fecal coliform concentrations in the first background samples taken from sites A and B were greater than subsequent background samples. The background samples taken on 27 Oct. 2003 exceeded the Washington State bacteria criteria for secondary recreational waters of 200 CFU of fecal coliform 100 ml<sup>-1</sup> (WSDOE, 2003). At site B, fecal coliform levels were 300 CFU 100 ml<sup>-1</sup> greater at the first background sampling

than on any other sampling day during the study. After the initial sampling day, fecal coliform levels in surface water declined to less than 50 CFU 100 ml<sup>-1</sup> (Figure 5).

On Day 2 following slurry application, an increase in fecal coliform and *E. coli* levels were seen at site A. This increase was a result of the direct movement of slurry into the surface water caused by the large rainfall event (2.8 cm), which occurred in the 48 h post-application. Even though bacteria levels at site A were increased, no increases in the concentration of bacteria were seen at the site immediately downstream (site B). The lack of bacteria detection at site B may have been due to adhesion of the bacteria to the streambed or vegetative matter in the waterway. Fajardo et al. (2001) reported that tall fescue vegetation decreased the concentration of fecal coliform in runoff water. Similarly, Lim et al. (1998) found that a 6.1 m vegetative filter strip completely reduced fecal coliform concentrations that were as high as 20.0 million CFU 100 ml<sup>-1</sup>.

Background levels of fecal coliform in surface water at sites E and F were even lower than background levels at sites A and B. Before slurry application, fecal coliform concentrations were 41 and 11 CFU 100 ml<sup>-1</sup> for sites E and F, respectively.

An increase in fecal coliform concentration was seen at site E, the surface water site closest to the slurry application area, on Day 3 following the January slurry application. There was also an explained increase at site A on Day 2 after application (Figure 6). Site A was located downstream from site E and runoff water from the

January application area appeared to flow through site E. The increase in fecal coliform and *E. coli* concentrations at site A on Day 2 appeared to be from other sources besides the slurry application areas, especially since concentrations of fecal coliform in the soil had reached background levels at the December application area before this sampling date. On Day 3 after the January application, there was an increase in *E. coli* of 200 CFU 100 ml<sup>-1</sup> that could be attributed to the increase in bacterial levels seen at site E.

The increase in fecal coliform concentration seen at site E after the January slurry application was less than 20% of the increase observed in site A following the December application. The lower bacteria levels seen in surface water during the January application may have been due to the lower concentration of bacteria in the slurry and the decreased application area. Because the January slurry application was still two to three times greater than the normal application area, transport of fecal bacteria to surface water under normal farm conditions would be expected to be much lower than levels seen during the January application. In addition, normal slurry application on the dairy is adjusted depending on predicted weather to avoid application prior to large rainfalls, as occurred during both the December and January applications. The risk of fecal bacterial runoff from this site is also decreased by the slow growing grass cover (Trevisan et al., 2002).

Increases in bacterial concentrations in the waterway leaving the farm boundary (sites C and D) occurred during several periods during the months following slurry application (Figure 7). The increases in bacteria at sites C and D did not appear to be

the result of the slurry application because fecal bacteria levels in the soils during several of these high concentrations were already at or below background levels. Large rainfall events appeared to increase the amount of bacteria entering the waterway from sources other than the slurry application. In the rainfall events on Day 2 and 3 after slurry application, bacteria concentrations leaving the farm boundary at sites C and D were increased. However, the increases in bacteria concentrations were not from the slurry application area because there were not any increases at site B, the site downstream from site A and upstream from sites C and D. The increases in fecal coliform concentrations at sites C and D could have been from numerous sources, including pastures used for grazing beef cattle or from wildlife in the forested land upstream of site D.

### **Bacterial Source Tracking**

A limited number of isolates were collected from water and soil samples immediately before and after the December slurry application. Soil samples used for BST were from background samples taken prior to slurry application (8 Dec. 2003) to establish the source of *E. coli* on the soil before slurry application. Eleven of the 12 isolates taken from the soil samples were identified as being from cattle (Table 2). The presence of an isolate from an unknown source indicates that other animals contributed to fecal bacteria present on the soil, even though cattle were expected to be the main source of *E. coli*. Contributions of fecal bacteria from wildlife was very likely as a herd of elk frequently entered the pasture and elk feces could be seen in the slurry application area. In addition, the pasture was accessible to birds and other wild animals.

All of the *E. coli* isolates taken from the waterway at site A were identified as originating from cattle sources (Table 2). The high percentage of isolates identified as cattle was expected as site A was located nearest to the slurry application area. At the farm boundary (site D), just over 60% of the bacteria isolates evaluated were from cattle. In 2003, Franz et al. found that over 87% of fecal streptococci bacteria in stream sites were classified as being from cattle. In our study, the isolates identified as being from cattle were not specific to the slurry application and could also be from beef cattle grazing in the pastures. The results of the BST indicated that source tracking may be useful in determining the contribution of bacteria from specific background sources. A challenge of BST is the expense of the procedure and the limited number of bacteria evaluated to determine the sources.

### **Nitrogen in Surface Water**

Total N, NH<sub>4</sub>-N, and NO<sub>3</sub>-N concentrations were determined in the water samples taken from the waterway. Nutrient composition of the slurry applied during the December and January applications is given in Table 1.

The concentration of TN increased at site A on Day 2 and Day 3 after the December slurry application. There was a slight increase in the concentration of TN (0.005 percentage units) at site B on Day 2 after application and a subsequent increase of 0.028 percentage units from Day 2 to Day 3. In contrast, the TN concentration in water was not affected by the January slurry application. After the January slurry

application, TN levels in the water declined on a daily basis even though there did appear to be some movement of  $\text{NH}_4\text{-N}$  into the surface water at the site closest to the slurry application area. Concentrations of TN in surface water were similar at site F, the background site, as well as sites E and A, indicating the presence of factors besides the slurry application was playing a larger role in the levels of TN in the water.

Concentrations of  $\text{NH}_4\text{-N}$  in surface water appeared to be directly related to movement of slurry into water. After the December slurry application, there was an increase in  $\text{NH}_4\text{-N}$  concentrations ( $0.65 \text{ mg kg}^{-1}$ ) seen in the water at site A. There was a very slight increase ( $0.01 \text{ mg kg}^{-1}$ ) in  $\text{NH}_4\text{-N}$  concentration at site B on Day 3 after slurry application. A similar pattern was observed in the concentration of  $\text{NH}_4\text{-N}$  after the January slurry application, with an increase in  $\text{NH}_4\text{-N}$  concentration ( $0.10 \text{ mg kg}^{-1}$ ) occurring on Day 3 after application at site E and an increase ( $0.06 \text{ mg kg}^{-1}$ ) at site A on Day 4. No  $\text{NH}_4\text{-N}$  was detected in water samples taken at site F on the days following the January slurry application. The concentrations of  $\text{NH}_4\text{-N}$  in runoff in our study were lower than the  $\text{NH}_4\text{-N}$  concentrations of  $2.34$  and  $0.93 \text{ mg L}^{-1}$ , respectively, found in runoff from manure applied plots with or without grass hedges as reported by Eghball et al. (2000).

Nitrate-N concentrations in water were lower than expected and were less than  $0.12 \text{ mg kg}^{-1}$   $\text{NO}_3\text{-N}$  at all of sampling points except for one. Similarly,  $\text{NO}_3\text{-N}$  concentrations of  $0.12 \text{ mg L}^{-1}$  or less were reported in runoff from tall fescue plots applied with manure (Fajardo et al, 2001). In contrast, Eghball et al. (2000) reported



NO<sub>3</sub>-N concentrations greater than 26.0 mg L<sup>-1</sup> for runoff water from plots receiving manure application. During the December slurry application, there was an increase in water NO<sub>3</sub>-N concentrations at site A. However, this was not expected due to the low amount of NO<sub>3</sub>-N present in slurry. After the January slurry application, there were increased concentrations of NO<sub>3</sub>-N at the background site (site F) and sites E and A. The increased concentration of NO<sub>3</sub>-N at site F indicate that the increase of NO<sub>3</sub>-N in the surface water was from a source, such as the forested land upstream, other than the slurry application.

Srivastava et al. (1996) found that TN, NO<sub>3</sub>-N, and NH<sub>4</sub>-N were reduced with longer lengths of vegetative filter strips. The vegetative setback zone between the slurry application area and the waterway appeared to adequately contain any N movement with the exception of the large runoff events that exceeded the filtering capacity of the setback zone. In addition, the plant growth in the waterway appeared to play a role in filtering N from the water, thus only small increases in N concentrations were found at the downstream sites.

### **Phosphorus in Surface Water**

In this study, there appeared to be some movement of TP and inorganic P into surface water as a result of the December slurry application. The inorganic P concentrations increased 0.82 mg kg<sup>-1</sup> on Day 2 after the December slurry application; however, the inorganic P concentration at site B only increased 0.15 mg kg<sup>-1</sup> on Day 2 (Figure 8). On Day 2 after the December slurry application, the TP concentration in the

water at site A was 3.2 times greater than the concentrations in water on the previous day. In 2000, Eghball et al. reported greater concentrations of dissolved available P ( $2.3 \text{ mg L}^{-1}$ ) in runoff from plots applied with manure and even greater concentrations ( $6.0 \text{ mg L}^{-1}$ ) of total P.

Total P and inorganic P concentrations indicated movement of nutrients into the water after the January slurry application. The inorganic P concentration was increased on Day 3 after slurry application at sites E and A, which were downstream from the point of application. However, inorganic P concentrations at site F also increased on Day 3 after the January application, indicating that there was some movement of inorganic P into the water from sources other than the slurry application. Therefore, use of inorganic P as an indicator of movement of manure nutrients into surface waters may be erroneous due to the possibility of background sources contributing to the increase in water P concentrations.

## **CONCLUSIONS**

An increase in the concentration of bacteria and nutrients in surface water was only detected when slurry was directly transported into surface water. When there was direct movement of slurry into the surface water, bacteria and nutrients appeared to move concurrently in the runoff. Besides the periods of large rainfall that resulted in direct runoff from the slurry application area, there were no increases in nutrient or bacteria concentrations in the waterway, indicating that the setback zones were

adequately containing runoff from the slurry application area. When the fecal bacteria and nutrient concentrations were increased at the sampling sites closest to the application areas as a result of the excessive runoff, the bacteria and nutrients showed little movement along the waterway during the sampling periods. The vegetative growth in the grass waterway appeared to act similar to a vegetative buffer and reduced the movement of fecal bacteria and nutrients. Results indicated that strategic winter slurry application proposes little risk of movement of bacteria and nutrients to surface water when best management practices of limited application and setback areas are utilized.

## **ACKNOWLEDGMENTS**

The authors would like to thank the Institute of Environmental Health in Seattle, WA for their laboratory assistance and interpretation for the bacterial source tracking. The authors would also like to thank the Washington State Dairy Industry for funding this project.

## **REFERENCES**

- Avery, S. M., A. Moore, and M. L. Hutchison. 2004. Fate of *Escherichia coli* originating from livestock faeces deposited directly onto pasture. *Lett. Appl. Microbiol.* 38:355–359.
- Carson, C. A., B. L. Shear, M. R. Ellersieck, and A. Asfaw. 2001. Identification of fecal *Escherichia coli* from humans and animals by ribotyping. *Appl. Environ. Microbiol.* 67:1503–1507.

- Clesceri, L. S., A. E. Greenberg, and A. D. Eaton, eds. 1998. Standard methods for the examinations of water and wastewater, 20<sup>th</sup> ed. Am. Publ. Health Assoc., Inc., Am. Water Works Assoc., and Water Environ Federation, Washington, D. C.
- Dombek, P. E., L. K. Johnson, S. T. Zimmerley, and M. J. Sadowsky. 2000. Use of repetitive DNA sequences and the PCR to differentiate *Escherichia coli* isolates from human and animal sources. *Appl. Environ. Microbiol.* 66:2572–2577.
- Eghball, B., J. E. Gilley, L. A. Kramer, and T. B. Moorman. 2000. Narrow grass hedge effects of phosphorus and nitrogen in runoff following manure and fertilizer application. *J. Soil Water Conserv.* 55:172–176.
- Fajardo, J. J., J. W. Bauder, and S. D. Cash. 2001. Managing nitrate and bacteria in runoff from livestock confinement areas with vegetative filter strips. *J. Soil Water Conserv.* 56:185–191.
- Frantz, E., K. Griswold, G. Apgar, B. Jacobson, and J. Haddock. 2003. Effectiveness of vegetative filter strips (VFS) for controlling pathogen loads and antibiotic resistance in dairy wastewater. p. 67–73. *In Anim., Agric. and Food Processing Wastes. Proc. 9<sup>th</sup> Int. Symp., Research Triangle Park, NC. 12–15 Oct. 2003. ASAE Publ., St. Joseph, MI.*
- Gagliardi, J. V., and J. S. Karns. 2002. Persistence of *Escherichia coli* O157:H7 in soil and on plant roots. *Environ. Microbiol.* 4:89–96.
- Himathongkham, S., S. Bahari, H. Riemann, and D. Cliver. 1999. Survival of *Escherichia coli* O157:H7 and *Salmonella typhimurium* in cow manure and cow manure slurry. *FEMS Microbiol. Lett.* 178:251–257.
- Jiang, X., J. Morgan, and M. P. Doyle. 2002. Fate of *Escherichia coli* O157:H7 in manure-amended soil. *Appl. Environ. Microbiol.* 68:2605–2609.
- Jones, D. L. 1999. Potential health risks associated with the persistence of *Escherichia coli* O157 in agricultural environments. *Soil Use Manage.* 15:76–83.
- Kudva, I. T., K. Blanch, and C. J. Hovde. 1998. Analysis of *Escherichia coli* O157:H7 survival in ovine or bovine manure and manure slurry. *Appl. Environ. Microbiol.* 64:3166–3174.
- Kuntz, R. L., P. G. Hartel, D. G. Godfrey, J. L. McDonald, K. W. Gates, and W. I. Segars. 2003. Targeted sampling protocol as prelude to bacterial source tracking with *Enterococcus faecalis*. *J. Environ. Qual.* 32:2311–2318.
- Lenahan, N. A., J. M. DeRouchey, T. T. Marston, M. L. Christian, and G. L. Marchin. 2004. Evaluation of round bale feeding sites on soil fecal bacteria and nutrient concentrations. *J. Dairy Sci.* 87(Suppl. 1):349.

- Lim, T. T., D. R. Edwards, S. R. Workman, B. T. Larson, and L. Dunn. 1998. Vegetative filter strip removal of cattle manure constituents in runoff. *Trans. ASAE* 41:1375–1381.
- Mawdsley, J. L., R. D. Bardgett, R. J. Merry, B. F. Pain, and M. K. Theodorou. 1995. Pathogens in livestock waste, their potential for movement through soil and environmental pollution. *Appl. Soil Ecol.* 2:1–15.
- Nennich, T. D., J. H. Harrison, D. L. Davidson. 2004. Winter application of dairy slurry on a grazing based dairy: 1.) Evaluation of nitrogen use in a native pasture. (Unpublished).
- Nunez-Delgado, A., E. Lopez-Periago, and F. Diaz-Fierros Viqueira. 2002. Chloride, sodium, potassium and faecal bacteria levels in surface runoff and subsurface percolates from grassland plots amended with cattle slurry. *Biores. Technol.* 82:261–271.
- Rosen, B. H., R. Croft, S. Stehman, E. R. Atwill, and S. E. Wade. 2000. Waterborne pathogens in agricultural watersheds. *Natural Resource, Agriculture, and Engineering Service*–147. Ithaca, NY.
- Sharpley, A. N., T. Daniel, T. Sims, J. Lemunyon, R. Stevens, and R. Parry. 2003. *Agriculture phosphorus and eutrophication*. 2<sup>nd</sup> ed. USDA ARS–149.
- Srivastava, P., D. R. Edwards, T. C. Daniel, P. A. Moore Jr., and T. A. Costello. Performance of vegetative filter strips with varying pollutant source and filter strip lengths. *Trans. ASAE* 39:2231–2239.
- 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.
- Trevisan, D., J. Y. Vansteelant, and J. M. Dorioz. 2002. Survival and leaching of fecal bacteria after slurry spreading on mountain hay meadows: consequences for the management of water contamination risk. *Water Res.* 36:275–283.
- Washington State Department of Ecology (WSDOE). 2003. Water quality standards for surface waters of the state of Washington. Chapter 173-201A WAC. Online. Available: <http://www.ecy.wa.gov/pubs/wac173201a.pdf>.
- Washington State Legislature. 1998. Dairy Nutrient Management. Chapter 90.64 RCW. [Online]. Available: <http://www.leg.wa.gov/RCW/index.cfm?fuseaction=chapterdigest&chapter=90.64>.

Table 1. Characteristics of slurry applied to grassland on December 12, 2003 and on January 27, 2004.

Item	December application	January application
Total N, %	0.26	0.25
Nitrate N, ppm	1.1	0.92
Ammonia N, ppm	1309	1373
Total P, ppm	766.7	361.5
Ortho-P, ppm	3.67	2.06
Total K, %	0.35	0.22
Fecal coliform, log <sub>10</sub> CFU/g	5.02	4.51
<i>Escherichia coli</i> , log <sub>10</sub> CFU/g	4.99	4.52
pH	7.45	7.9

Table 2. Classification of *Escherichia coli* isolates taken from water or soil samples. Isolates were DNA fingerprinted and identified as originating from cattle or unknown sources. Bacteria source tracking was conducted on 2 to 4 colonies from individual water samples taken from sites A, B, C, and/or D on December 8, 14, 15, 21, 30 and January 8 and 11 and from soil samples taken from plots X or Y on December 8, 2003.

Site	Classification of isolates				Total isolates Number
	Cattle		Unknown		
	Number	%	Number	%	
A	9	100	0	0	9
B	18	82	4	18	22
C	35	88	5	12	40
D	8	62	5	38	13
Total from water	70	83	14	17	84
Plot					
X	5	83	1	17	6
Y	6	100	0	0	6
Total from soils	11	92	1	8	12

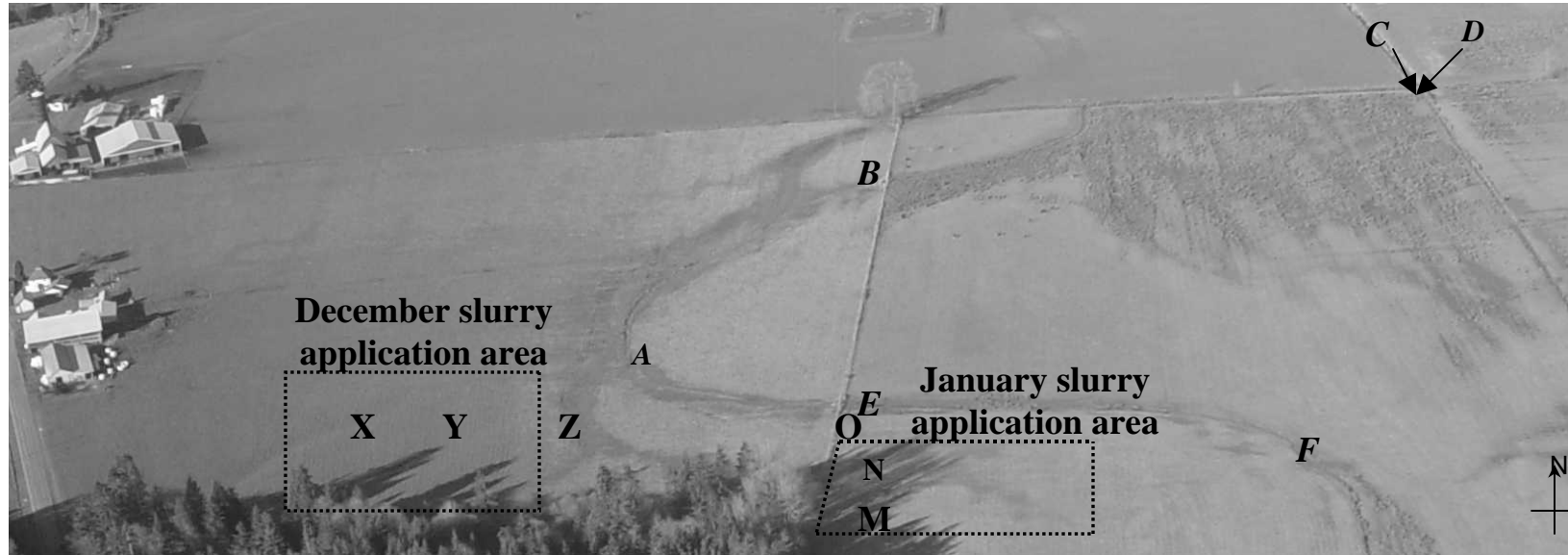


Figure 1. Location of manure application area, water sampling sites, and soil plots. The slurry applications took place on 12 Dec. 2003 and 27 Jan. 2004. Site A was located 10.6 m from the point of slurry application and site B was located 216 m further downstream. Sites C and D (287 m downstream from site B) were located at the property boundary. Site C included a waterway draining the western half of the property and site D included drainage water from the entire property and wooded areas to the east of the farm property. Sampling sites A, C, and D were sampled throughout the January sample collection period. In addition, sites E and F were sampled to monitor runoff from the January application area. Site F was located upstream from the slurry application area and was 152 m from site E. Site E was located 19.8 m from the point of slurry application and was 111 m from site A.



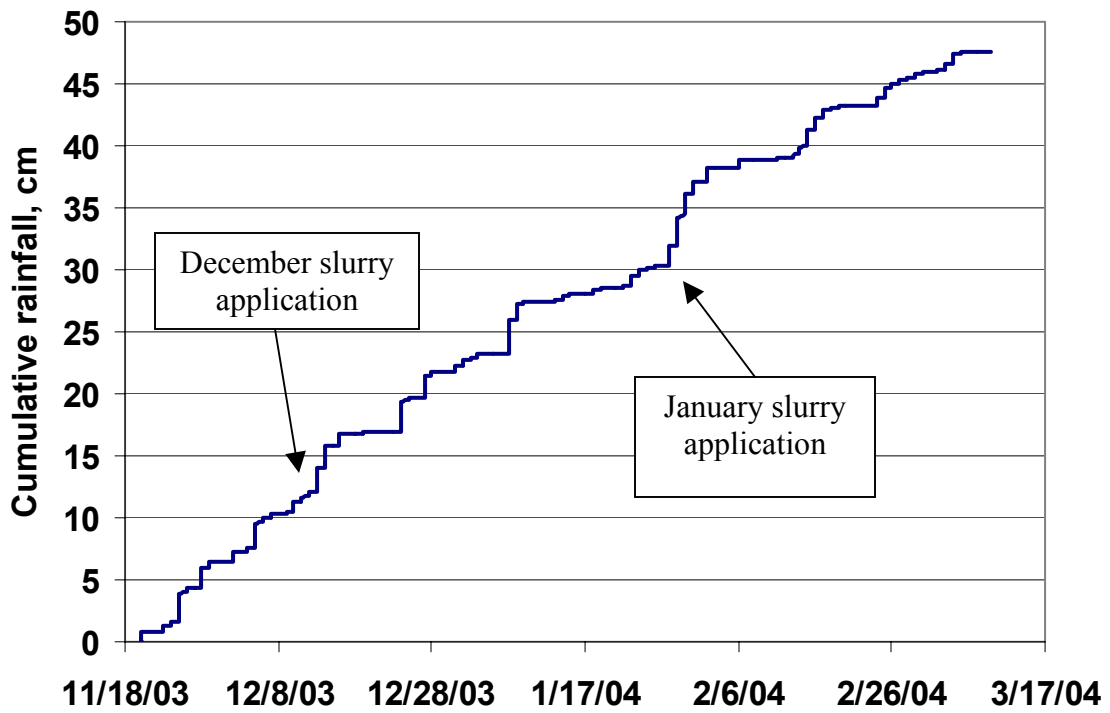


Figure 2. Cumulative rainfall recorded with a weather station located at the study location. The December slurry was applied on December 12, 2003 and the January slurry application took place on January 27, 2004.

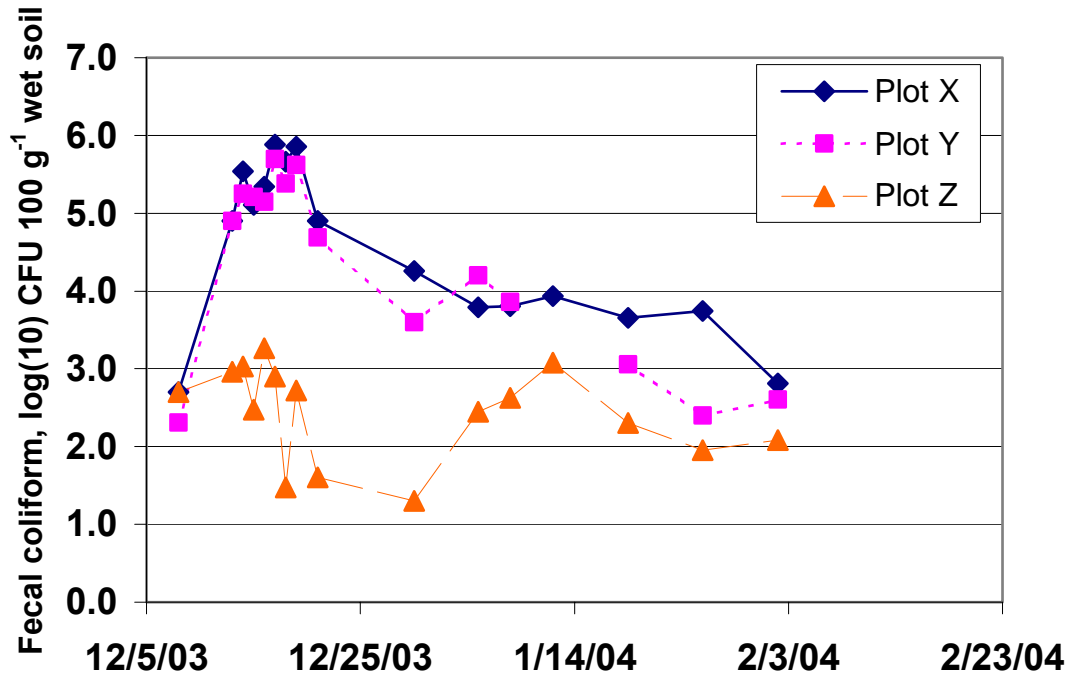


Figure 3. Fecal coliform bacteria concentrations in the soil from plots X, Y, and Z before and after slurry application on December 12, 2003. Plots X and Y were located in the slurry application area and plot Z was located in the grass setback zone between the application area and the waterway.

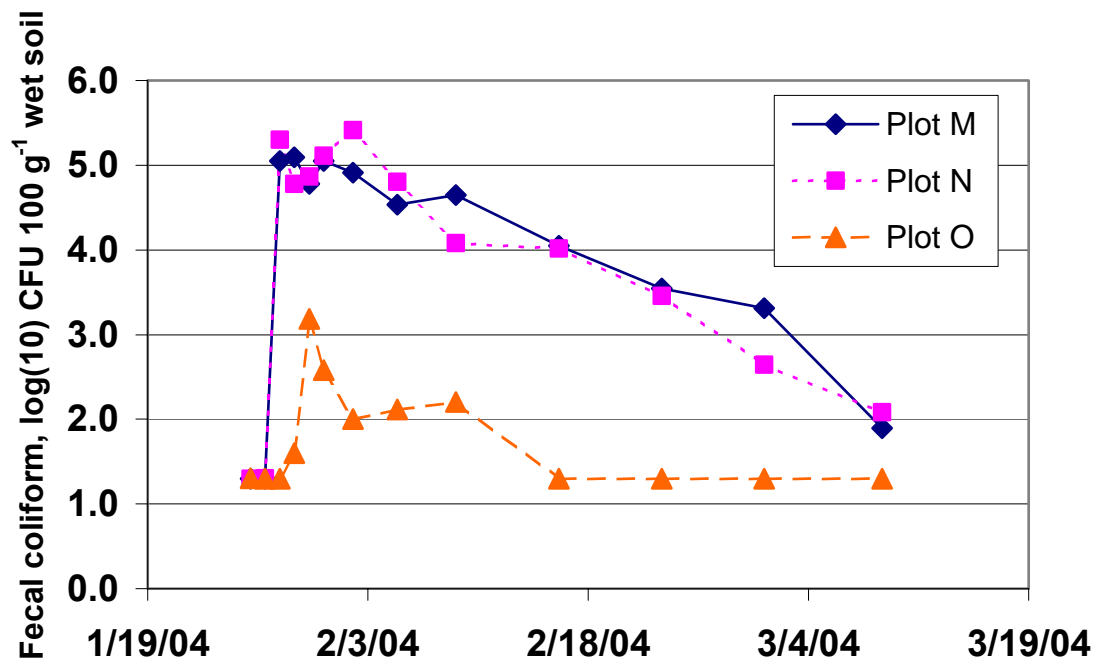


Figure 4. Fecal coliform bacteria concentrations in the soil from plots M, N, and O before and after slurry application on January 27, 2004. Plots M and N were located in the slurry application area and plot O was located in the grass setback zone between the application area and the waterway.

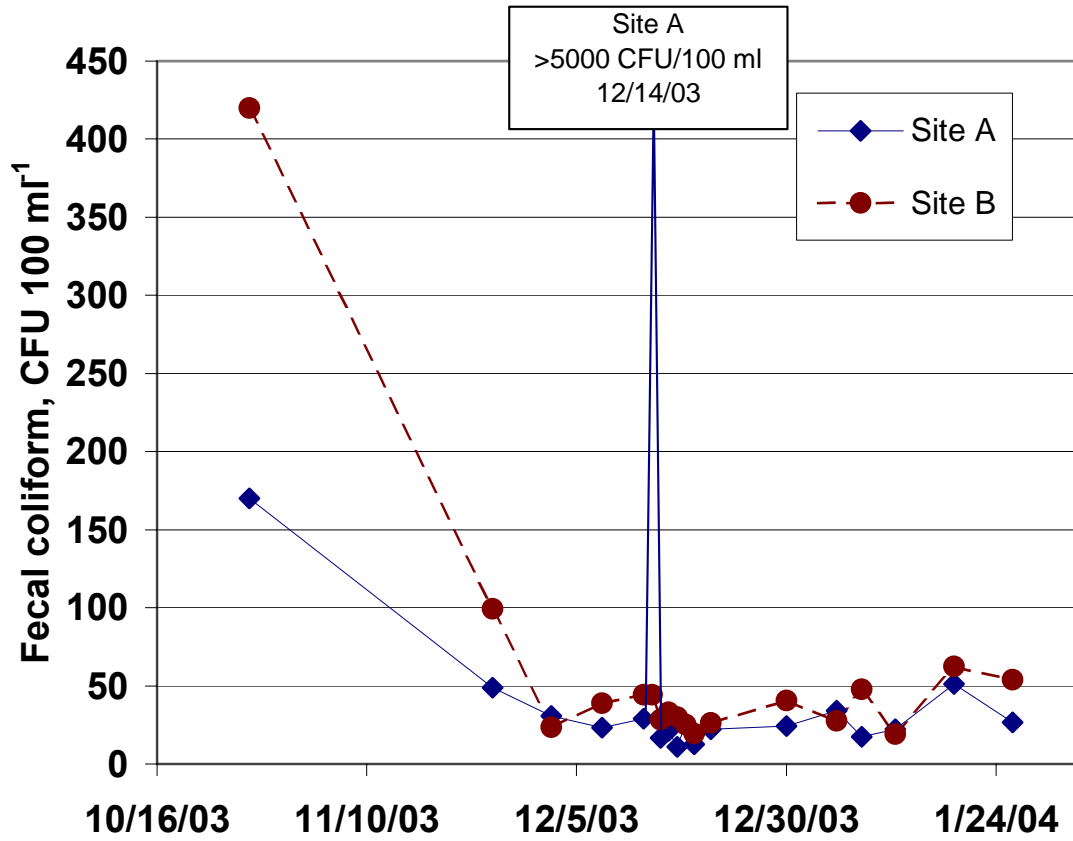


Figure 5. Fecal coliform bacteria concentrations at sites A and B before and after slurry application on December 12, 2003. The concentration of fecal coliform bacteria was greater than 5000 CFU 100 ml<sup>-1</sup> at site A on d 2 after slurry application.

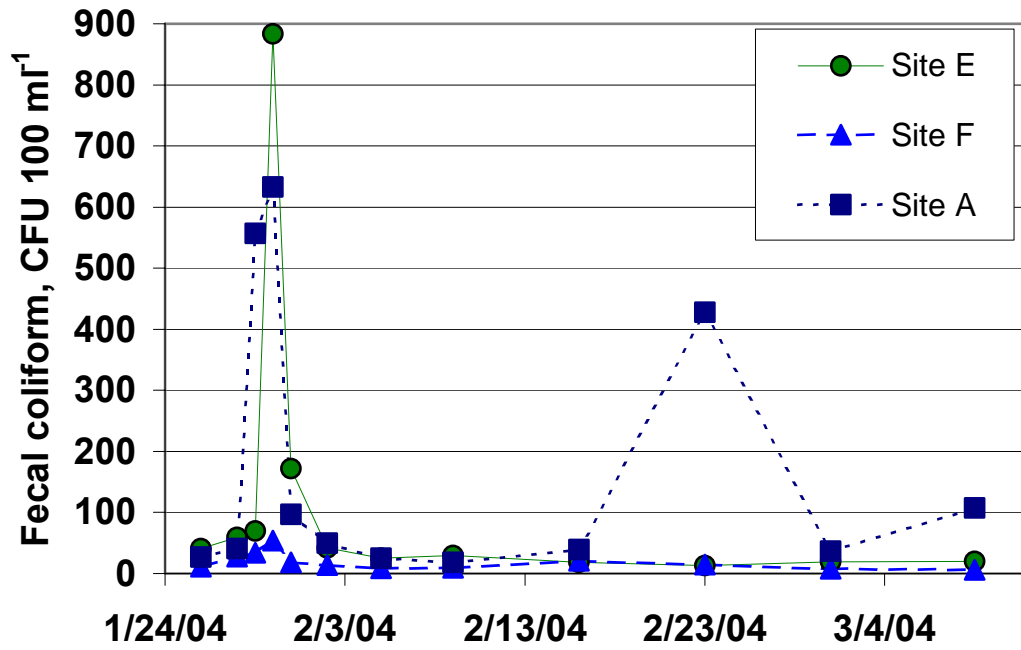


Figure 6. Fecal coliform bacteria concentrations at sites E, F, and A before and after slurry application on January 27, 2004.

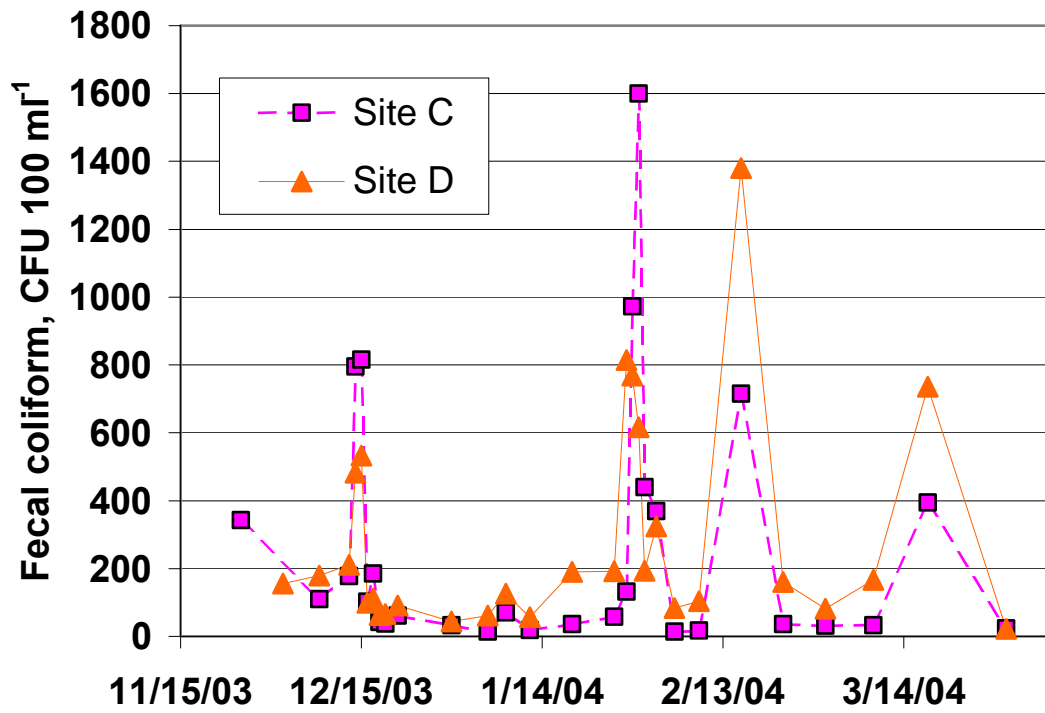


Figure 7. Fecal coliform bacteria concentrations at sites C and D before and after slurry applications on December 12, 2003 and January 27, 2004.

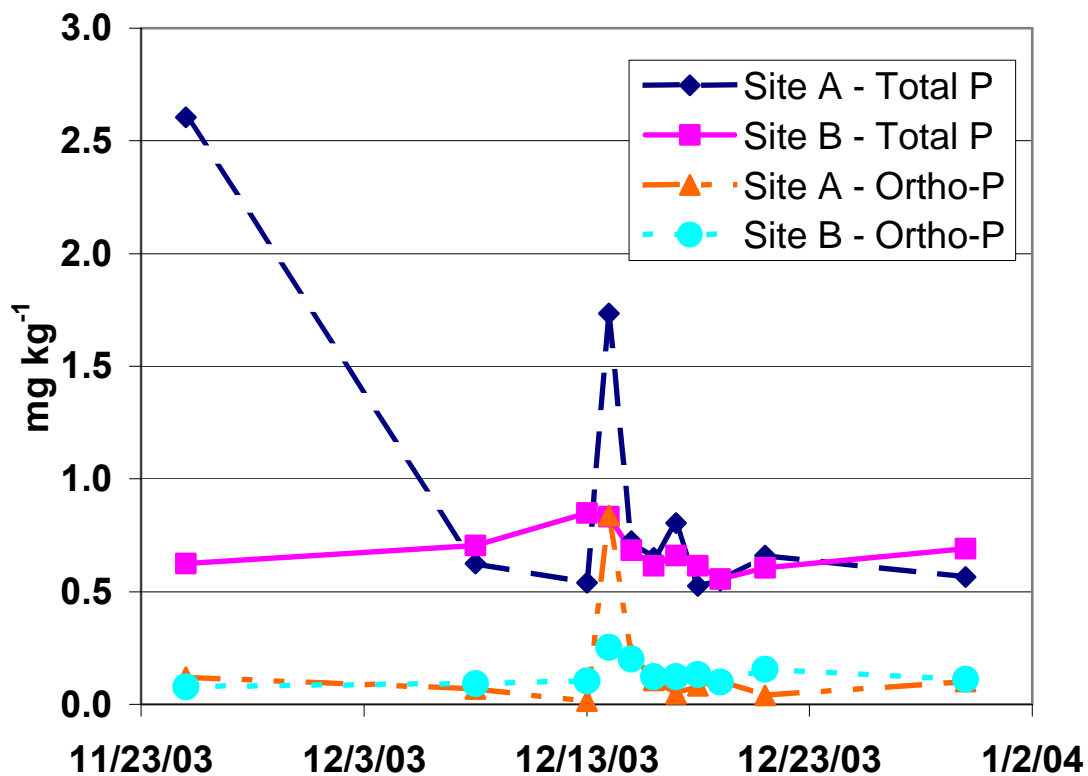


Figure 8. Concentrations of ortho-P and total P in water samples collected at sites A and B before and after manure application. Manure was surface applied to pastureland on December 12, 2003.

# **Characterization of Manure Excretion and Environmental Impacts of Nutrient Management in Dairy Production Systems**

**T. D. Nennich**

## **SUMMARY AND CONCLUSIONS**

Estimations of manure excretion from dairy cattle and best management practices for handling manure are important aspects of nutrient management. Although on-farm measurements are the best method of measuring actual manure production on a dairy operation, these measurements are not easy to obtain for many existing operations and are not available for new operations. Equations provide estimations of excretion in situations where actual measurements of excretion are not available. In 2001, an effort was initiated to update the 2001 ASAE manure excretion standards for each of the livestock species. The approach taken to revise the dairy cattle standards was to compile datasets from total collection metabolism studies from different classes of dairy animals from numerous universities. The data were compiled and evaluated for excretion of total manure, total solids, N, P, and K. Equations that include a variety of feed and animal variables were included for the prediction of manure and nutrient excretion from dairy animals. The prediction equations developed can be valuable tools for estimating manure and nutrient excretion from dairy cattle.



Updating estimation of manure and nutrient excretion from dairy cattle is important as intake and production levels of lactating dairy cows increase. The 2001 ASAE manure excretion standards underestimated total manure excretion for a 625 kg cow by ~28%. For a 100-cow dairy, manure excretion would have been underestimated by over 770,000 kg per year. The underestimation of manure excretion would have been increased to over 7.7 million kg for a 1000-cow dairy. In order to store the manure produced on a 1000-cow dairy, and an additional 7780 m<sup>3</sup> of storage space would be required to contain the manure alone.

Nitrogen balance on dairy operations is an area of concern due to the risk of nitrates leaching through soil into groundwater. The 2001 ASAE manure excretion estimates under-predicted N excretion from lactating dairy cows by approximately 210 g/d. The underestimation of N excretion would translate to over 75,000 kg of additional N excretion on a 1000-cow dairy per year. This large difference in N excretion will result in greater ammonia losses to the atmosphere as well as requiring additional acres of land to utilize the N at an agronomic rate.

Changes in the dairy industry have also brought about changes in the way dairy operations are managed. Currently, calves and heifers are often housed at separate locations or facilities than lactating cows. Because of the different facilities, manure from calves and heifers is often handled separately than manure from lactating cows. The current ASAE (2001) manure excretion standards do not provide separate estimates of manure excretion for non-lactating animals. Addition of prediction equations for

non-lactating dairy animals provide information for planning facilities and manure handling equipment for these classes of animals. The 2001 ASAE manure excretion estimates for dairy cattle overestimated manure and N excretion by 29.3 and 0.164 kg/d as compared to the average 437 kg heifer in our dataset.

Prediction equations developed for manure and urine excretion can be used as a source of information for future technologies. For an example, estimations of urine excretion provide information for management strategies, such as separation of feces and urine in animal housing facilities. An understanding of urine excretion, along with urinary N and mineral excretion, provides information to contribute to development of future technologies. Knowledge of factors affecting urine and urinary nutrient excretion, such as intake of N, Na, and K, provide an understanding of options to reduce excretion and nutrients on the farm level. The dataset developed for prediction of excretion, which included over 300 cows, allowed for evaluation of previously published equations as well as development of new prediction equations for urinary and urinary N excretion.

An understanding of manure and nutrient excretion is only the first step in managing manure and nutrients on a dairy operation. Environmentally friend handling and land-application prevents environmental contamination and risks to human health. Establishment of best management practices (BMP) are essential to provide livestock operations with guidelines as to the best ways to manage manure and nutrients. Although BMP should protect the environment, the establishment of BMP must also

provide some flexibility to dairy operations. Scientific studies provide a method of determining the risks associated with specific management practices.

Winter manure application is a practice that has been discouraged due to its association with increased risk of nutrient and bacteria transport to the environment. Although winter manure application may not be a viable management practice for many dairy operations, inhibiting the practice from all dairy operations may not be a reasonable solution.

Application of dairy slurry during winter months did not increase the risk of nutrient transport on the dairy used in these studies due to the BMP that were implemented on the dairy. The BMP utilized on this dairy to prevent movement of nutrients into waterways included low slurry application rates, setback areas between application areas and waterways, and applying during periods with a low probability of rainfall.

In the study to determine N usage from winter application of dairy slurry, dairy slurry was applied once in winter, with a second application after the first cutting of grass. Winter application of dairy slurry on a native pasture resulted in increased forage dry matter and N yields in our study. The dairy slurry applied during the winter months resulted in greater apparent N recovery than did the summer applications. Soil temperatures that were ~5°C above freezing reduced microbial activity that would have resulted in mineralization of organic N and nitrification of NH<sub>4</sub>-N in the dairy slurry.

The losses of N that occurred after the winter slurry application were most likely caused by denitrification, a process that releases  $N_2O$  and  $N_2$  to the atmosphere, and not a result of nitrate leaching. Denitrification was promoted due to anaerobic conditions resulting from soil saturation during the high rainfall of the winter months.

The summer slurry applications on the plots resulted in a large percentage of  $NH_4-N$  loss through volatilization. Ammonia volatilization is greater when the slurry is not incorporated into the soil and weather conditions are hot and dry, which were factors present in our study. The slurry applied in summer was not incorporated into the soil and there was very little rainfall after the slurry was applied to the field. Therefore, the slurry was accessible to the effects of the warm weather conditions. The very dry soil conditions during the summer months decreased mineralization of organic N to a plant available form of N.

An additional study was designed to evaluate the potential transport of fecal bacteria (fecal coliform and *E. coli*), N, and P to surface water. The use of BMP during winter slurry applications appeared to minimize bacteria and nutrient transport to surface waters. Fecal coliform and *E. coli* concentrations in the top 3.8 cm of the soil declined more than 70% within the first 10 d in our study. Fecal bacteria levels in the soils returned to background levels in 52 d or less after the slurry applications. In our study, the only influx of nutrients or bacteria into surface water occurred during large rainfall events. The risk of nutrient and bacteria transport to the waterway in our study

was much greater than typical farm practice due to the greater application area utilized in our study.

The nutrients in the dairy slurry showed similar movement to that of the fecal bacteria in the soil. Monitoring nutrient levels in waterways was complicated by the background influxes of nutrients into the waterway. In this study, concentrations of  $\text{NO}_3\text{-N}$  and P in the water were increased at the background sites, indicating the increases in nutrient concentrations in the water were not always directly related to the winter slurry application.

In summary, adequate estimations of manure and nutrient excretion from dairy cattle, as well as urine and urinary nutrient excretion from lactating dairy cows, provides estimates of the amount of manure and nutrients that need to be managed on an operation. Understanding factors affecting manure and nutrient excretion provide dairy producers with the knowledge to alter manure and nutrient excretion and provides a source of information from which development of new technologies may be based. Best management practices should be continually evaluated to determine which practices adequately protect the environment while providing dairy operations alternative ways of managing manure and nutrients.

## **APPENDIX 1**

## **Additional introduction**

Various management factors affect the amount of manure excreted from an animal. These factors include dry matter intake, body weight, and stage of lactation (Smith and Frost, 2000; Wilkerson et al., 1997). Likewise, total manure excretion increases proportionally to milk production and subsequent increases in feed intake.

Nitrogen is a major environmental concern because of its potential effects on groundwater quality. Field application of N in excess of plant uptake can lead to leaching and contamination of groundwater. Volatilization of N in the form of ammonia results in a loss of valuable nutrients to the atmosphere and has a very pungent smell that may lead to social concerns. Nutrient excretion by dairy cattle has become an even greater concern as nutrients from cattle operations have created environmental challenges in some areas of the country. Nutrient management plans that optimize feeding and fertilizing practices need to be implemented to increase efficiency of N and P use at the farm level (Kuipers et al., 1999). Nitrogen is the nutrient of concern for many dairy operations and is the basis of most nutrient management plans. Nitrogen excretion is dependent on N intake of the animal and increases proportionally as the crude protein content of the diet increases (Frank et al., 2002). Numerous studies have focused on decreasing nitrogen output from heifers (James et al., 1999) or lactating cows (Frank and Swensson, 2002; Frank et al., 2002; Harrison et al., 2002; Karg and Wattiaux, 2002; Krober et al., 2000) by reducing their crude protein intake. Reduced intake of crude protein particularly reduces the amount of N excreted via urine. Heifers fed a diet containing 9.6% CP excreted 9.4 g/d less urinary urea nitrogen

per day than heifers fed an 11% CP diet (James et al., 1999). Jonker et al. (2002) considered various management factors affecting N utilization efficiency and found increased milk production per cow had the greatest impact for increasing N efficiency.

Another nutrient of major environmental concern is P because it accelerates surface water eutrophication (Sharpley et al., 2003). Eutrophication causes algal blooms that decay and consume dissolved oxygen in the water, resulting in restricted water use for recreation, fisheries, and drinking water (Sharpley et al., 2003). Land application of manure according to N content has led to a major buildup of P in many soils in the United States. In 2000, soil tests showed as many as 24 of the 50 states have soil P levels in the high or very high categories and do not require P fertilization (Sharpley et al., 2003). Phosphorus excretion is reduced when less dietary P is fed. Wu et al. (2000) reported a 23% decrease in fecal P excretion when dietary P was reduced from 0.49 to 0.40% of the diet.

### Results and Discussion

Equations were developed that included multiple independent variables that increased the precision of the  $M_E$  estimates. An equation [1-1] was developed for operations with animal information beyond MILK, but without accurate DMI values available to be used in calculations.

$$M_E = (\text{Milk} \times 0.738 (\pm 0.062)) + (\text{BW} \times 0.050 (\pm 0.007)) + (\text{DIM} \times 0.013 (\pm 0.006)) + (\text{C}_{\text{NDF}} \times 0.46 (\pm 0.19)) - 8.2 (\pm 8.8) \quad [1-1]$$



Residual SE = 9.5, Inter-study SE = 6.7

Body weight and  $C_{\text{NDF}}$  were significant factors and were included in [1-1] because addition of these factors improved the precision of the equations. Prediction equations reported by Wilkerson et al. (1997) included BW, MILK, DIM,  $C_{\text{CP}}$ , and  $C_{\text{NDF}}$  to predict  $M_{\text{E}}$  for lactating cows. In the LACT dataset,  $C_{\text{CP}}$  was not a significant factor and  $M_{\text{E}}$  was more precisely predicted using other variables.

The most precise method of predicting  $M_{\text{E}}$  in the LACT dataset was when DMI, MTP, DIM, and DMD were included as independent variables [1-2].

$$M_{\text{E}} = (\text{DMI} \times 2.65 (\pm 0.089)) - (\text{DMD} \times 111.0 (\pm 8.0)) - (\text{DIM} \times 0.0090 (\pm 0.0042)) + (\text{MTP} \times 227.8 (\pm 106.3)) + 77.9 (\pm 6.4) \quad [1-2]$$

Residual SE = 6.3, Inter-study SE = 5.5

Apparent DMD averaged 0.666 g/g DM for cows in the LACT dataset. Although [1-2] has the lowest residual SE of all equations, DMD was not included in the proposed ASAE equations due to the impracticality of measuring DMD on commercial dairy farms.

Non-linear models were evaluated for  $M_{\text{E}}$  using the LACT dataset. Variables evaluated in equations included squared and two-way interactions of DMI, MILK, DIM, BW,  $C_{\text{NDF}}$ , and  $C_{\text{CP}}$ . Most of the models evaluated resulted in prediction equations that

would be expected to be less accurate predictors of  $M_E$  than linear equations. The best non-linear equation [1-3] for describing the LACT dataset included several of the same independent variables given by Wilkerson et al. (1997).

$$\begin{aligned}
 M_E = & (BW \times 0.017 (\pm 0.006)) + (DMI \times 0.94 (\pm 0.92)) + (DIM \times 0.030 \\
 & (\pm 0.013)) - (C_{NDF} \times 2.7 (\pm 1.7)) - (C_{CP} \times 1.5 (\pm 1.2)) - \\
 & (DIM \times DIM \times 0.0001 (\pm 0.00002)) + (DMI \times C_{CP} \times 0.091 (\pm 0.052) + \\
 & (C_{NDF} \times C_{NDF} \times 0.042 (\pm 0.023)) + 68.7 (\pm 39.3) \quad [1-3] \\
 \text{Residual SE} = & 6.9, \text{ Inter-study SE} = 10.3
 \end{aligned}$$

Equation [1-3] only improved the residual SE over other equations by 1%. Since use of the non-linear  $M_E$  equation only provided a very slight improvement over the linear equations, we suggest the use of the linear equations for predicting  $M_E$ .

***Nitrogen excretion.*** Nitrogen prediction equations were developed using a smaller dataset developed to reflect  $N_E$  estimates from cows fed N at levels close to their MP requirements. The original LACT dataset was divided into two datasets, LOWMP and HIGHMP, according to MP requirements as determined by the 2001 NRC model. Cows that were fed less than or equal to 112% of their MP requirements were in the LOWMP dataset (369 cow-periods) and cows fed greater than 112% of their MP requirements were in the HIGHMP dataset. Cows fed greater than 112% of their requirements were separated into the HIGHMP dataset to prevent the inflation of excreted N table values and equations due to less than ideal feeding practices. The

HIGHMP dataset was used as an evaluation dataset for the equations developed with LOWMP.

The simple linear equation, using MILK as the only independent variable [1-4], indicated a significant relationship between  $N_E$  and MILK. When MILK was used as the only prediction variable, it resulted in the most imprecise prediction of the equations evaluated, but still provided a more accurate estimate for planning than a singular table value.

$$N_E = (\text{Milk} \times 3.68 (\pm 0.53)) + 297.3 (\pm 21.8) \quad [1-4]$$

$$\text{Residual SE} = 63.9, \text{ Inter-study SE} = 52.7$$

Equations were developed for situations where additional information besides MILK was known. Equation [1-5] included MILK, CP intake, and MF and improved the residual SE by 29.6% over [1-4]. Equation [1-6] included MILK, DIM, BW, MTP, and  $C_{CP}$  and improved the residual SE by 11.6% over [1-4].

$$N_E = (\text{DIM} \times 0.139 (\pm 0.032)) + (\text{DMI} \times C_{CP} \times 93.1 (\pm 4.8)) + \\ (\text{MF} \times 830.8 (\pm 466.3)) + 24.8 (\pm 29.3) \quad [1-5]$$

$$\text{Residual SE} = 44.9, \text{ Inter-study SE} = 61.9$$

$$N_E = (\text{Milk} \times 5.81 (\pm 0.56)) + (\text{DIM} \times 0.229 (\pm 0.050)) + \\ (\text{BW} \times 0.221 (\pm 0.059)) + (\text{MTP} \times 5795.9 (\pm 1313.5)) +$$

$$(C_{CP} \times 1187.3 (\pm 392.5)) - 330.2 (\pm 89.6) \quad [1-6]$$

Residual SE = 56.4, Inter-study SE = 43.8

$$N_E = (NI \times 0.578 (\pm 0.031)) + 80.8 (\pm 22.2) \quad [1-7]$$

Residual SE = 46.4, Inter-study SE = 60.3

Equations developed using LOWMP were evaluated with the HIGHMP dataset to determine if the equations were applicable to cows fed protein at levels in excess of their requirements. In [1-5], which included CP intake as an independent variable, the equation developed did not have any significant mean or linear biases. However, when [1-4] and [1-6] were evaluated using the HIGHMP dataset, there were significant mean biases ( $P < 0.01$ ). Equations [1-4] and [1-6] underestimated  $N_E$  by 58.8 ( $\pm 13.92$ ) and 50.6 ( $\pm 13.24$ ) g/d, respectively, for cows in the HIGHMP dataset.

Evaluation of equations developed with LOWMP indicated that including cows in the HIGHMP dataset inflated the prediction of  $N_E$  for equations in which CP intake was not included. Therefore,  $N_E$  equations were developed using only cows in the LOWMP dataset.

Nitrogen intake was a significant independent variable for  $N_E$  when evaluated in a simple equation [1-7]. As expected, an increase in N consumption resulted in greater  $N_E$ .

The most precise equation developed for predicting  $N_E$  included CP intake as an independent variable [1-5]. The addition of the independent variables DIM and MF to [1-7] resulted in a reduction in the residual SE from 46.4 to 44.9. When [1-5] was evaluated using the HIGHMP dataset, there were not any significant mean or linear biases, indicating that the equation was applicable across various levels of CP intake.

Quadratic models were evaluated to determine if the models were improved through the addition of squared terms and interactions. When quadratic models were evaluated, the resulting equations did not reduce the residual SE or inter-study SE compared to the linear models evaluated. Conversely, Wilkerson et al. (1997) reported that development of quadratic models led to a statistical improvement over the linear models developed for predicting  $N_E$ .

## **Dry Cows**

As with the lactating cows, prediction equations were developed for dry cows. The DRY dataset only allowed for prediction of  $M_E$ ,  $DM_E$ , and  $N_E$  as the dataset did not include intake or excretion values for P, K, or other minerals. Equations listed for P [1-8] and K [1-9] excretion assume that intake is equal to excretion.

$$P_E = (((DMI \times 1000) \times C_P \times DP) - 264.386)/DP \quad [1-8]$$

$$K_E = ((DMI \times 1000) \times C_K) \quad [1-9]$$

In the case of  $P_E$  [1-8], a subtraction was made to account for P requirements of the fetus in pregnant animals. The adjustment factor for the gestation P requirement was derived from the 2001 Dairy NRC and is dependent on days pregnant.

Manure excretion and  $DM_E$  for dry cows were dependent on BW (Equations [1-10] and [1-11], respectively). Although it was expected that DMI would affect the amount of  $M_E$ , DMI was not a significant factor for  $M_E$  in the dry cow dataset, most likely a result of the extremely small dataset available for dry cows.

$$M_E = (BW \times 0.022 (\pm 0.015)) + 21.8 (\pm 11.4) \quad [1-10]$$

$$\text{Residual SE} = 5.7, \text{ Inter-study SE} = 5.9$$

$$DM_E = (DMI \times 0.18 (\pm 0.63)) + 2.73 (\pm 7.34) \quad [1-11]$$

$$\text{Residual SE} = 0.45, \text{ Inter-study SE} = 0.74$$

$$DM_E = (BW \times 0.004 (\pm 0.001)) + 1.86 (\pm 1.01) \quad [1-12]$$

$$\text{Residual SE} = 0.59, \text{ Inter-study SE} = 0.42$$

Conversely, when equations to predict  $DM_E$  were developed, DMI was the best predictor of  $DM_E$  [1-11]. A separate equation for  $DM_E$  is listed for prediction purposes if DMI data are not available [1-12].

Nitrogen excretion from dry cows in the DRY dataset was dependent on DMI and the  $C_{CP}$  [1-13].

$$N_E = (DMI \times 12.7 (\pm 4.7)) + (C_{CP} \times 1606.3 (\pm 244.8)) - 117.5 (\pm 37.1) \quad [1-13]$$

Residual SE = 45.5

In 1997, Wilkerson et al. reported that BW, days of pregnancy, and  $C_{NDF}$  were significant predictors of  $N_E$ , but these factors were not significant in the DRY dataset.

Table 1-1. Additional prediction equations developed for estimating manure, dry matter, and nitrogen excretion for lactating cows.

Equation	Residual SE	Inter-study SE
<b><i>Manure excretion</i></b>		
$M_E = (\text{MILK} \times 0.616 (\pm 0.057)) + 46.2 (\pm 2.3)$	10.0	7.1
$M_E = (\text{MILK} \times 0.792 (\pm 0.054)) + (\text{C}_{\text{NDF}} \times 43.5 (\pm 12.4)) + (\text{MTP} \times 845.2 (\pm 123.2))$	9.8	6.2
$M_E = (\text{DIM} \times -0.0074 (\pm 0.0042)) + (\text{DMI} \times 2.573 (\pm 0.096)) + (\text{BW} \times 0.018 (\pm 0.0040))$	7.1	9.4
$M_E = (\text{MILK} \times 0.846 (\pm 0.62)) + (\text{BW} \times 0.0551 (\pm 0.0071)) + (\text{C}_{\text{NDF}} \times 0.46 (\pm 0.18)) + (\text{MTP} \times 874.1 (\pm 150.3)) - 38.3 (\pm 10.3)$	9.3	6.0
$M_E = (\text{DMI} \times 2.20 (\pm 0.23)) + (\text{BW} \times 0.0140 (\pm 0.0056)) + (\text{C}_{\text{NDF}} \times 33.7 (\pm 17.04)) + (\text{C}_{\text{CP}} \times \text{DMI} \times 2.3 (\pm 1.2)) - 10.8 (\pm 7.5)$	7.0	10.3
$M_E = (\text{DMI} \times 1.54 (\pm 0.65)) + (\text{DMI} \times \text{DMI} \times 0.025 (\pm 0.015)) + 20.8 (\pm 7.3)$	7.1	9.5
<b><i>Dry matter excretion</i></b>		
$\text{DM}_E = (\text{MILK} \times 0.0874 (\pm 0.0070)) + 5.63 (\pm 0.29)$	1.21	0.87
$\text{DM}_E = (\text{DMI} \times 0.355 (\pm 0.011)) + (\text{C}_{\text{NDF}} \times 5.4 (\pm 1.9)) - 1.13 (\pm 0.77)$	0.78	1.17
$\text{DM}_E = (\text{MILK} \times 0.1124 (\pm 0.0077)) + (\text{BW} \times 0.00619 (\pm 0.00089)) + (\text{MTP} \times 106.0 (\pm 18.8)) - 2.19 (\pm 0.95)$	1.15	0.78
<b><i>N excretion</i></b>		
$N_E = (\text{MILK} \times 2.82 (\pm 0.42)) + 346.2 (\pm 18.1)$	70.9	57.9
$N_E = (\text{DMI} \times 9.4 (\pm 1.7)) - (\text{MILK} \times 1.28 (\pm 0.36)) + (\text{BW} \times 0.144 (\pm 0.027)) + (\text{DMI} \times (\text{C}_{\text{CP}}/6.25) \times 0.308 (\pm 0.054))$	49.3	66.7
$N_E = (\text{DIM} \times 0.128 (\pm 0.047)) + (\text{MILK} \times 4.59 (\pm 0.46)) + (\text{BW} \times 0.399 (\pm 0.052)) + (\text{C}_{\text{CP}} \times 632.9 (\pm 237.6)) + (\text{MTP} \times 4072.8 (\pm 1160.3)) - 219.3 (\pm 71.6)$	65.1	50.1
$N_E = (\text{DIM} \times 0.191 (\pm 0.044)) + (\text{MILK} \times 4.16 (\pm 0.44)) + (\text{BW} \times 0.377 (\pm 0.052)) + (\text{C}_{\text{CP}} \times 613.8 (\pm 239.4)) + (\text{C}_{\text{NDF}} \times 267.3 (\pm 136.1)) - 173.4 (\pm 75.9)$	65.8	48.4



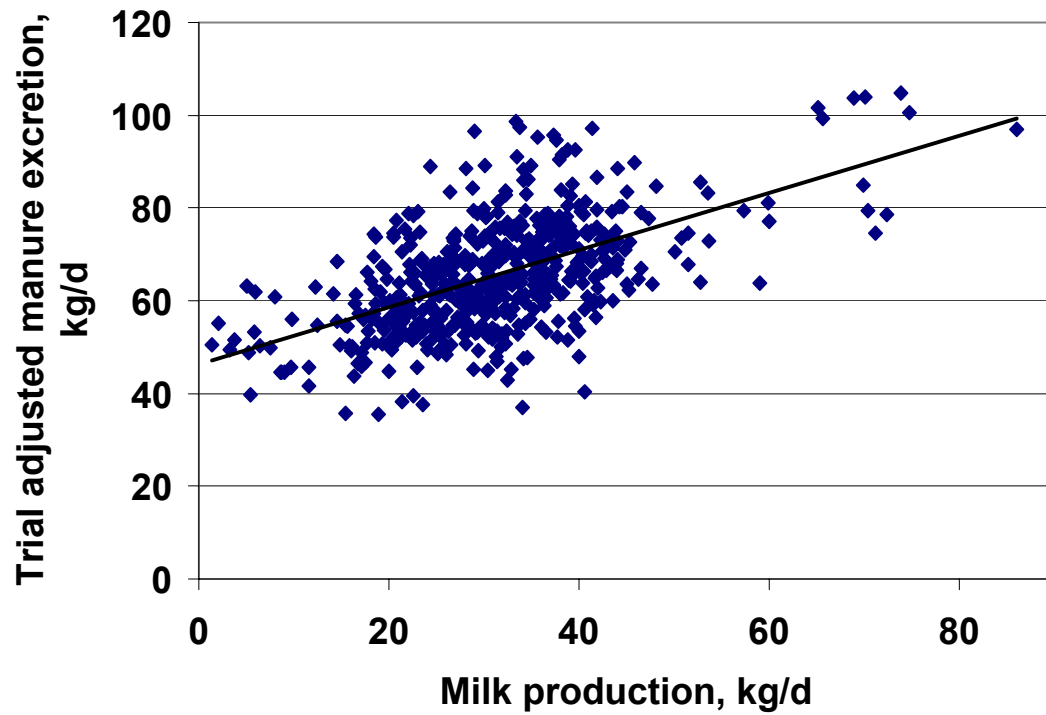


Figure 1-1. Relationship between milk production and trial adjusted manure excretion (kg/d) for lactating cows (LACT dataset, n = 553). The solid line is equal to Manure excretion (kg/d) = (Milk, kg/d x 0.647) + 43.212, Residual SE = 9.19, Inter-study SE = 6.94.

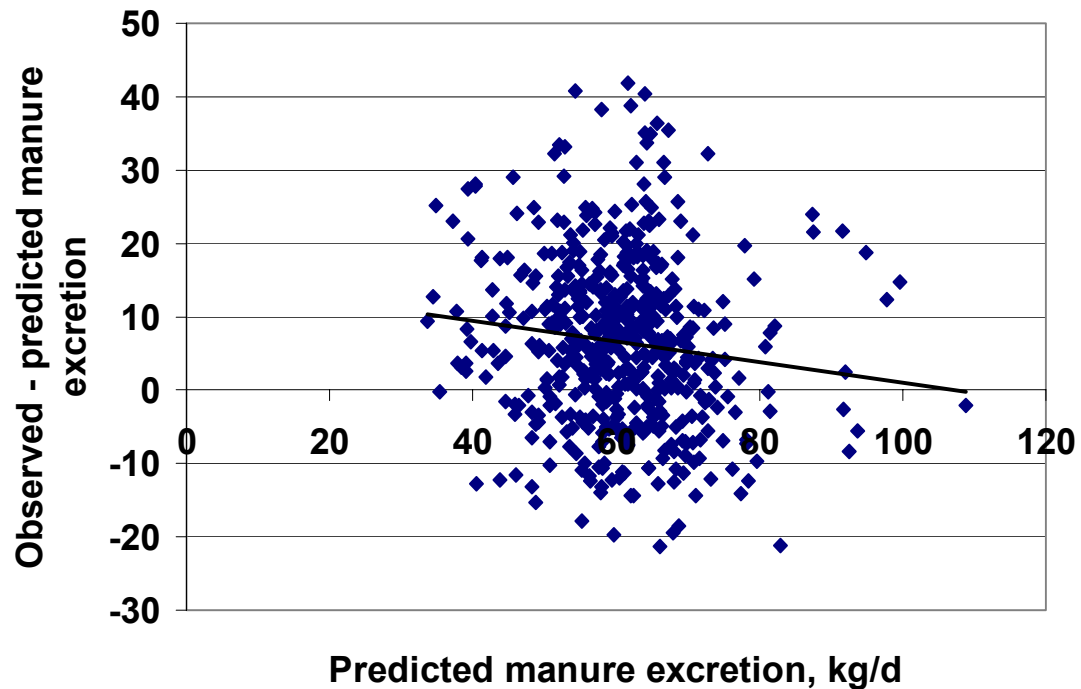


Figure 1-2. Plots of residuals versus predicted values from evaluation of the equation [total manure excretion, kg/d = (0.0286 x BW, kg) + (0.0378 x DIM) + (1.0689 x Milk, kg/d) + (0.0967 x CP, % of DM) + (0.614 x NDF, % of DM) – 21.94] by Wilkerson et al. 1997. The solid line is  $y = 5.60 - 0.25(X - 60.59)$ . There were significant mean ( $5.60 \pm 1.37$ ;  $P < 0.001$ ) and linear ( $-0.251 \pm 0.058$ ;  $P < 0.001$ ) biases.

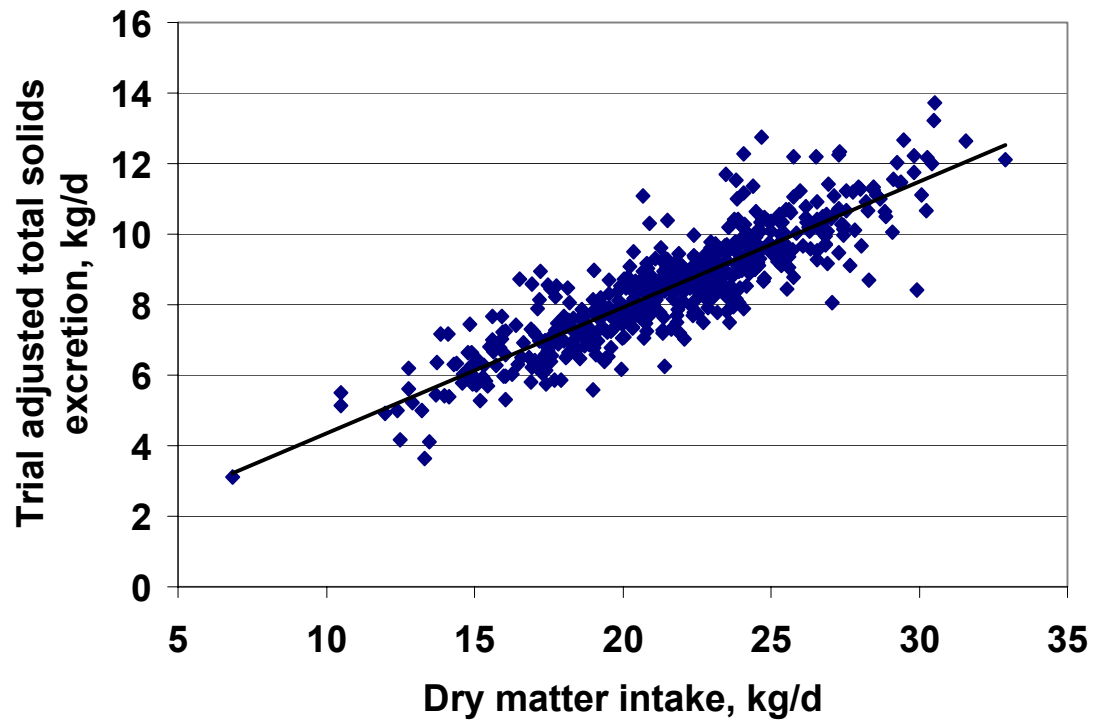


Figure 1-3. Relationship between dry matter intake and trial adjusted total solids excretion (kg/d) for lactating cows (LACT dataset, n = 553). The solid line is equal to total solids excretion, kg/d = (DMI, kg/d x 0.356) + 0.804, Residual SE = 0.78, Inter-study SE = 1.11.

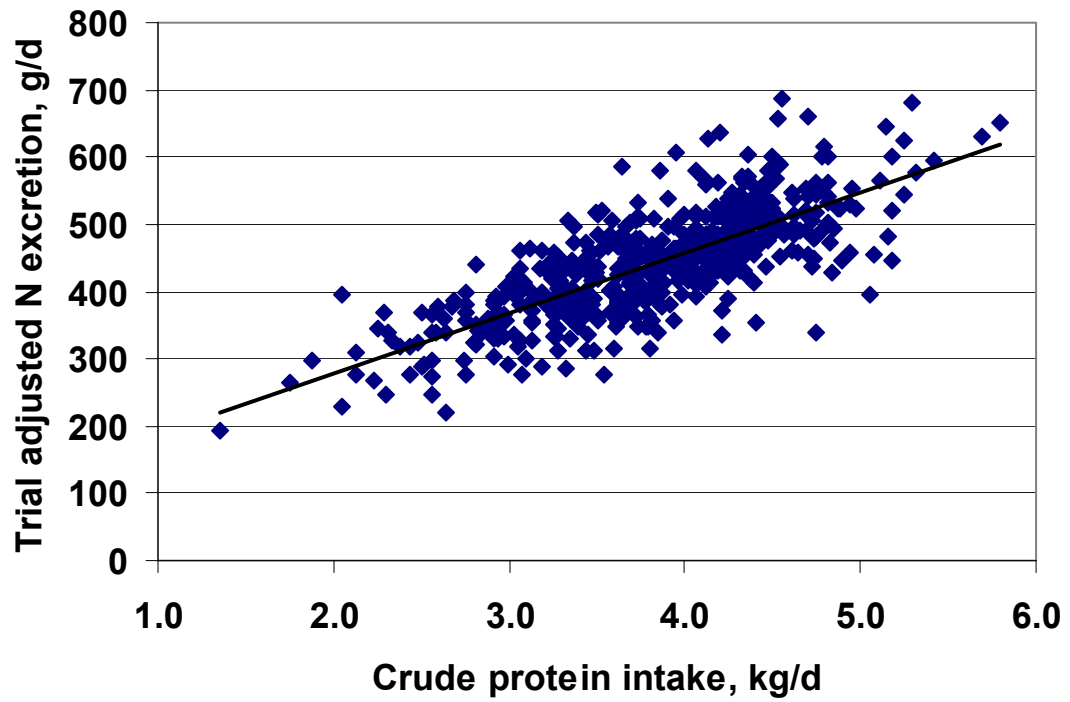


Figure 1-4. Relationship between crude protein (CP) intake and trial adjusted N excretion for lactating cows (554 cow-periods). The solid line is equal to nitrogen excretion, g/d = (CP intake, kg/d x 89.24) + 100.6, Residual SE = 52.9, Inter-study SE = 55.9.

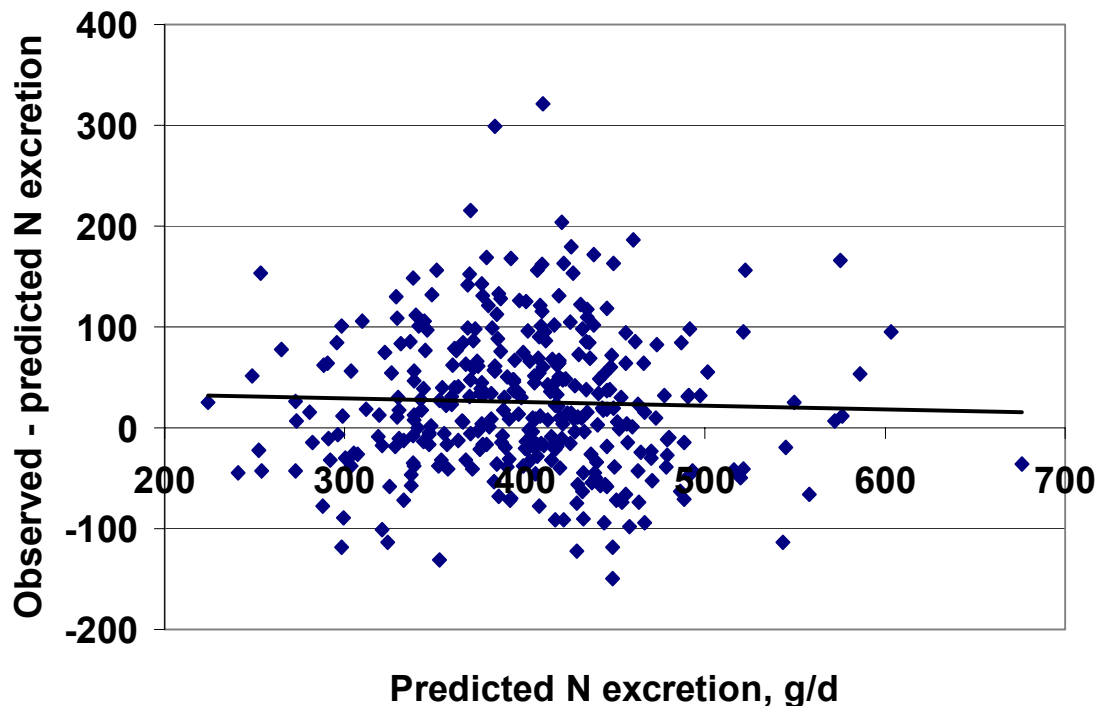


Figure 1-5. Plot of residuals versus predicted values from evaluation of the equation [N excretion, g/d = ((0.000232 x BW, kg) + (0.000342 x DIM) + (0.0064 x Milk, kg/d) + (0.0183 x CP, % of DM) + (0.00280 x NDF, % of DM) - 0.440) x 1000] by Wilkerson et al. 1997. The equation was evaluated using cows that consumed less than or equal to 112% of metabolizable protein requirements according to the 2001 Dairy NRC computer model. The solid line is  $y = 21.92 - 0.176(X - 399.6)$ . There were significant mean ( $21.92 \pm 9.18$ ,  $P < 0.03$ ) and linear ( $-0.176 \pm 0.068$ ,  $P < 0.01$ ) biases.

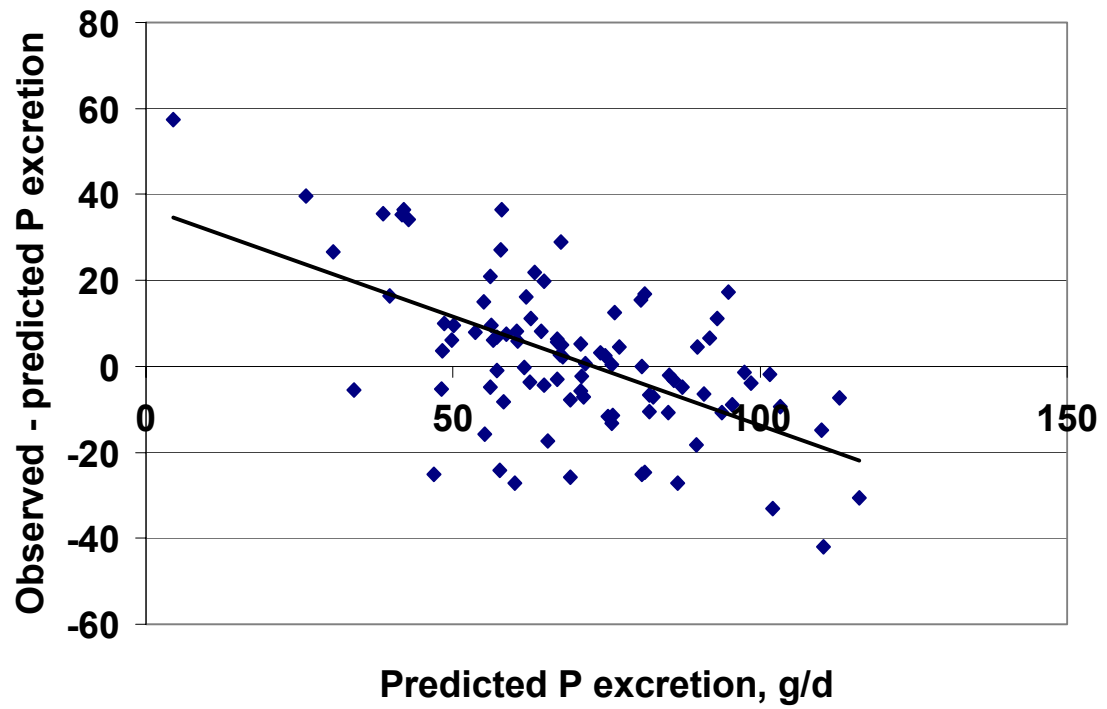


Figure 1-6. Plots of residuals versus predicted values from evaluation of the equation P excretion, g/d = PI, g/d – MilkP, g/d. The solid line is  $y = 1.168 - 0.439(X - 68.2)$ .

The mean bias was not significant, but there was a significant linear ( $-0.439 \pm 0.083$ ,  $P < 0.01$ ) bias.

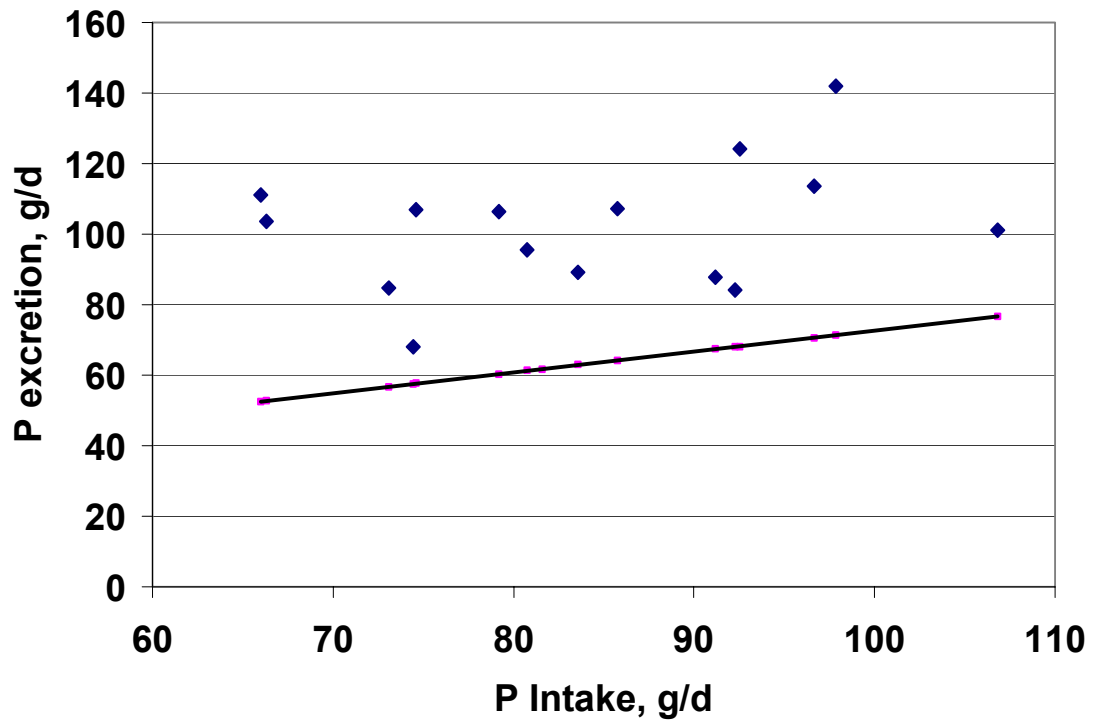


Figure 1-7. Relationship between phosphorus excretion and phosphorus intake in early lactation cows (n = 15, DIM = 36). The solid line is the predicted phosphorus excretion for cows consuming the same levels of phosphorus. The equation of the solid line is P excretion, g/d = (DMI, kg/d x C<sub>p</sub> x 586.3 (± 0.069)) + 13.931 (± 7.677), Residual SE = 11.61, Inter-study SE = 8.83. Data from cows in later lactation showed a significant relationship between phosphorus intake and excretion, but there was not a significant relationship for early lactation cows.

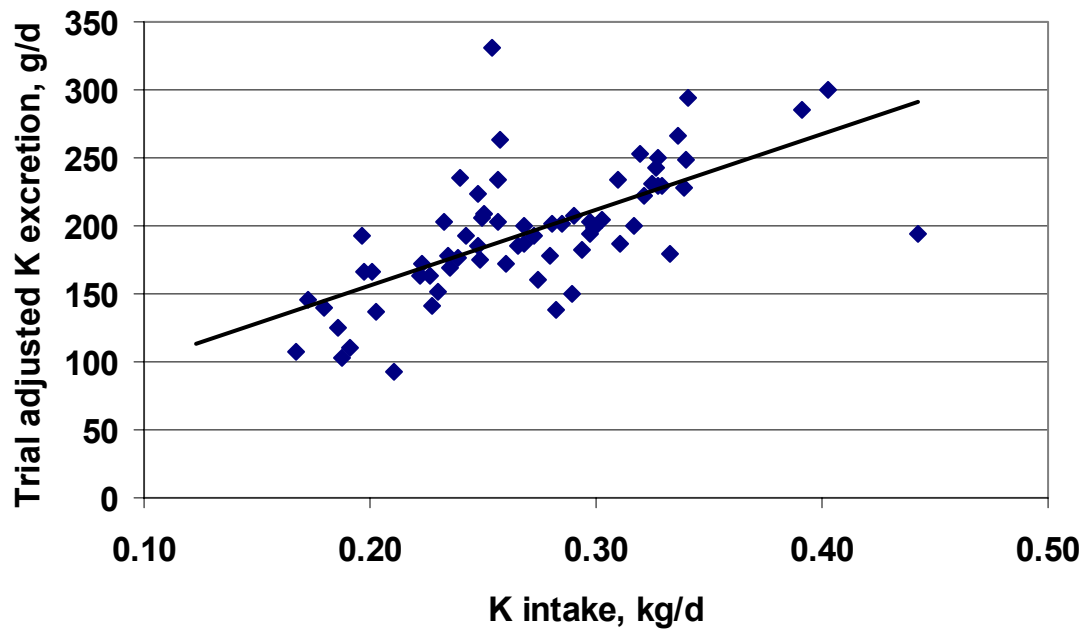


Figure 1-8. Relationship between potassium intake and trial adjusted potassium excretion (g/d) for lactating cows (MINERAL dataset, 66 cow-periods). The solid line is equal to  $K_E = (DMI \times C_K \times 0.5556) + 44.9$ , Residual SE = 36.6, Inter-study SE = 12.3.



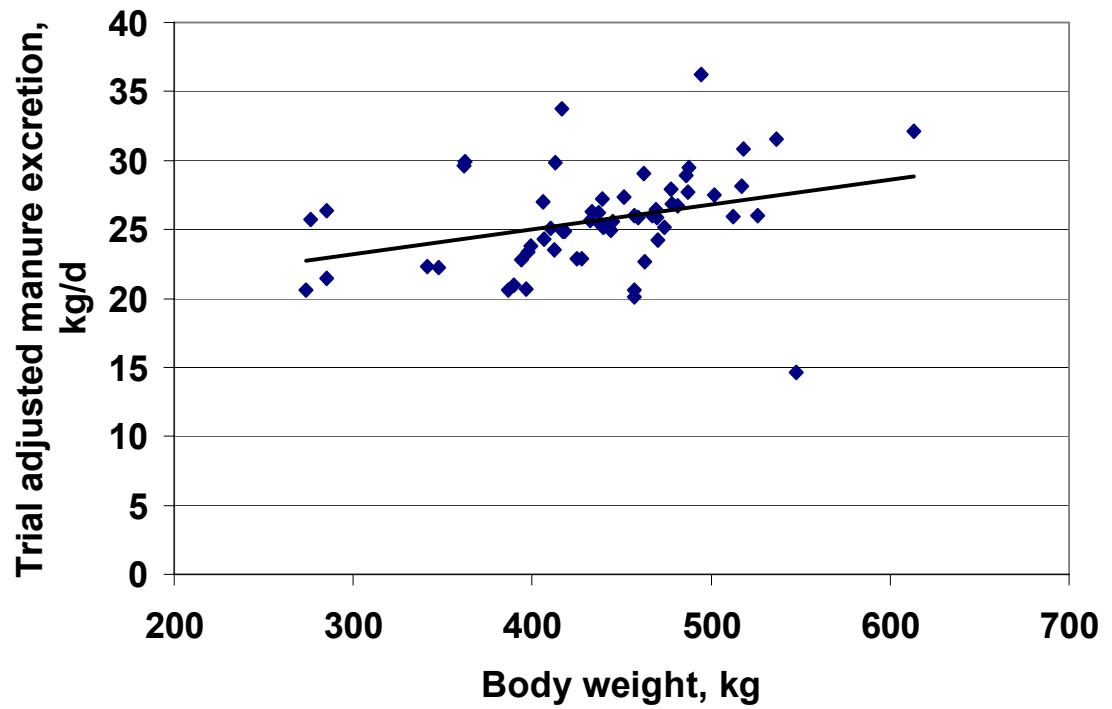


Figure 1-9. Relationship between body weight and trial adjusted manure excretion (g/d) for heifers (HEIFER dataset, n = 60). The solid line is equal to total manure excretion,  $\text{kg/d} = (\text{BW, kg} \times 0.018 (\pm 0.010)) + 17.817 (\pm 4.772)$ , Residual SE = 3.55, Inter-study SE = 4.02.

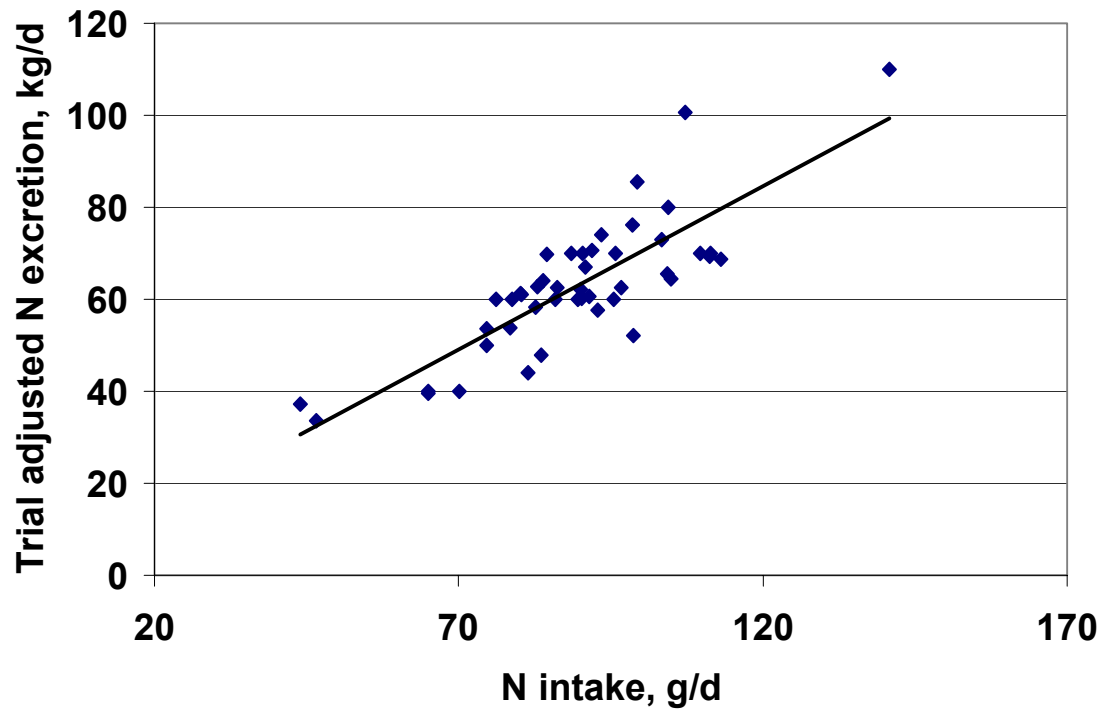


Figure 1-10. Relationship between nitrogen intake and trial adjusted nitrogen excretion (g/d) for calves (CALF dataset, n = 46). The solid line is equal to  $N \text{ excretion, g/d} = (NI, \text{ g/d} \times 0.711 (\pm 0.073)) - 0.730 (\pm 6.612)$ , Residual SE = 8.30.

## **APPENDIX 2**

Table 2-1. Additional prediction equations developed for estimating urine excretion and urinary N excretion from lactating cows.

Equation	Residual SE	Inter-study SE
<b><i>Urine excretion</i></b>		
$U_E = (\text{NaI} \times 0.062 (\pm 0.016)) + (\text{MUN} \times 0.43 (\pm 0.21)) + 11.4 (\pm 3.8)$	5.8	5.2
$U_E = (\text{NaI} \times 0.061 (\pm 0.016)) + 16.8 (\pm 2.9)$	5.9	5.0
$U_E = (\text{DCAD} \times 0.337 (\pm 0.090)) + 13.2 (\pm 3.6)$	6.1	3.8
$U_E = (\text{NaI} \times 0.018 (\pm 0.012)) + 20.5 (\pm 3.8)$	6.2	5.2
$U_E = (U_{\text{Na}} \times 0.162 (\pm 0.025)) + 11.6 (\pm 2.9)$	5.5	4.6
$U_E = (U_{\text{NaPct}} \times -18.9 (\pm 10.7)) + 32.2 (\pm 4.4)$	2.7	5.9
$U_E = (\text{UKPct} \times -17.2 (\pm 4.3)) + 36.9 (\pm 3.7)$	6.1	6.0
$U_E = (U_{\text{N}} \times 50.9 (\pm 13.6)) + (U_{\text{Na}} \times 0.119 (\pm 0.024)) - (\text{UKPct} \times 22.8 (\pm 3.2)) + 19.8 (\pm 3.4)$	4.1	5.2
$U_E = (\text{MUN} \times 0.57 (\pm 0.11)) + (\text{BW} \times 0.0188 (\pm 0.0054)) + (\text{DMI} \times 0.348 (\pm 0.093)) - (\text{C}_{\text{NDF}} \times 0.43 (\pm 0.17)) + 13.6 (\pm 7.5)$	5.5	4.4
$U_E = (\text{MUN} \times 0.58 (\pm 0.11)) + (\text{BW} \times 0.0189 (\pm 0.0054)) + (\text{DMI} \times 0.359 (\pm 0.093)) - 2.7 (\pm 4.0)$	5.5	5.3
<b><i>Urinary N excretion</i></b>		
$U_{\text{N}} = (\text{MUN} \times 6.05 (\pm 0.86)) + (\text{BW} \times 0.337 (\pm 0.041)) - 69.3 (\pm 30.7)$	43.2	41.7
$U_{\text{N}} = (\text{MP} \times 0.056 (\pm 0.0064)) + 85.4 (\pm 18.9)$	43.6	42.1
$U_{\text{N}} = (\text{NI} \times 0.223 (\pm 0.025)) + 85.9 (\pm 19.7)$	44.5	46.6
$U_{\text{N}} = (\text{MUN} \times 5.78 (\pm 0.94)) + 145.1 (\pm 17.1)$	47.0	42.4

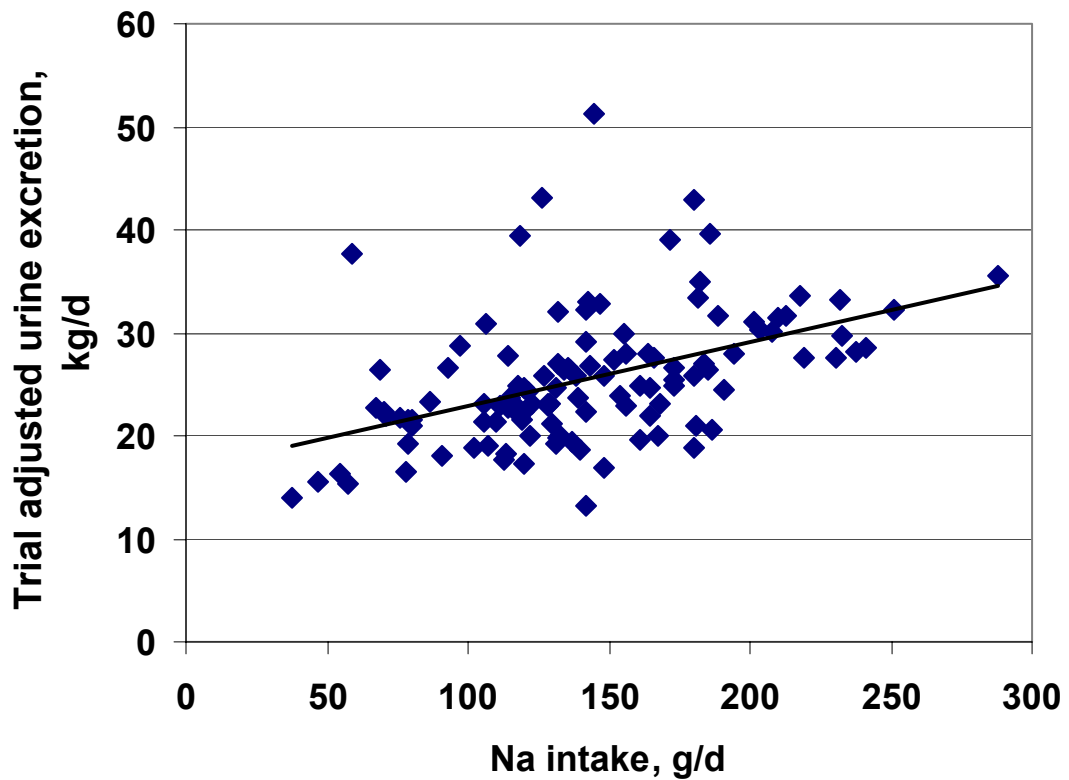


Figure 2-1. Relationship between the Na intake (g/d) and trial adjusted urine excretion (kg/d) for lactating cows (MINERAL dataset, n = 115). The solid line is equal to Urine excretion (kg/d) = (0.061 x Na intake, g/d) + 16.8, Residual SE = 5.9, Inter-study SE = 5.0.

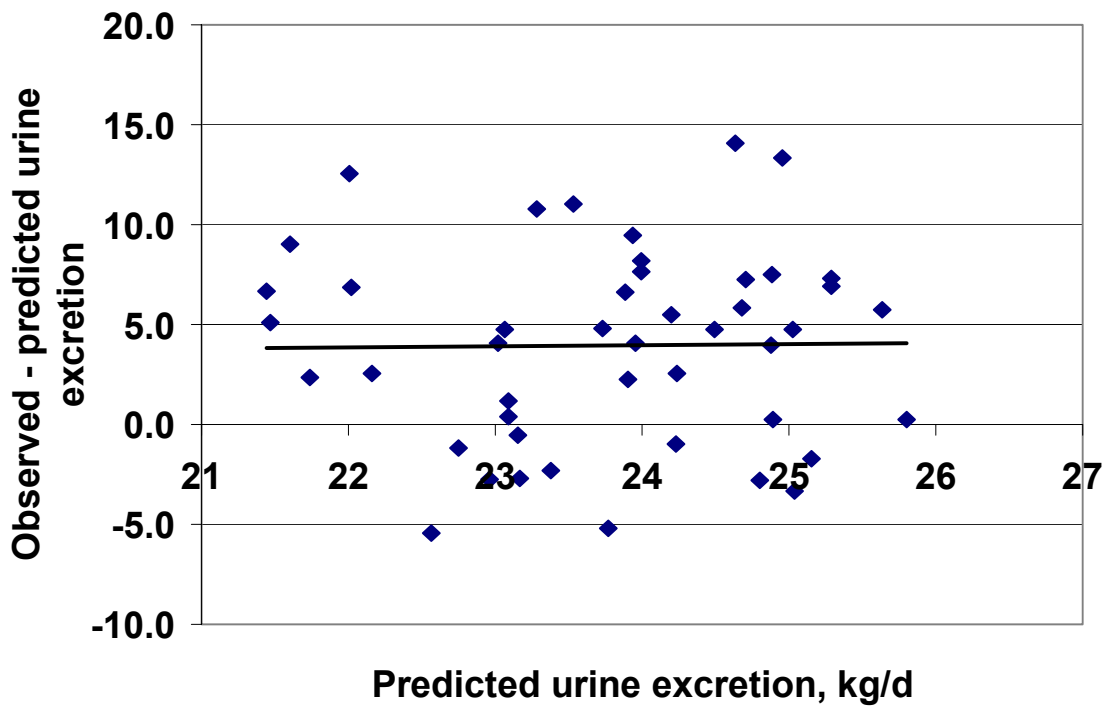


Figure 2-2. Plot of residuals (observed – predicted) versus predicted values of urine excretion (kg/d) from evaluation of the equation [Urine excretion, kg/d = 0.563 x MUN, mg/dl + 17.1] developed using the LACT dataset. The equation was evaluated using the VALIDATE dataset. The solid line on the graph represents the equation  $y = 0.049x + 2.79$ .

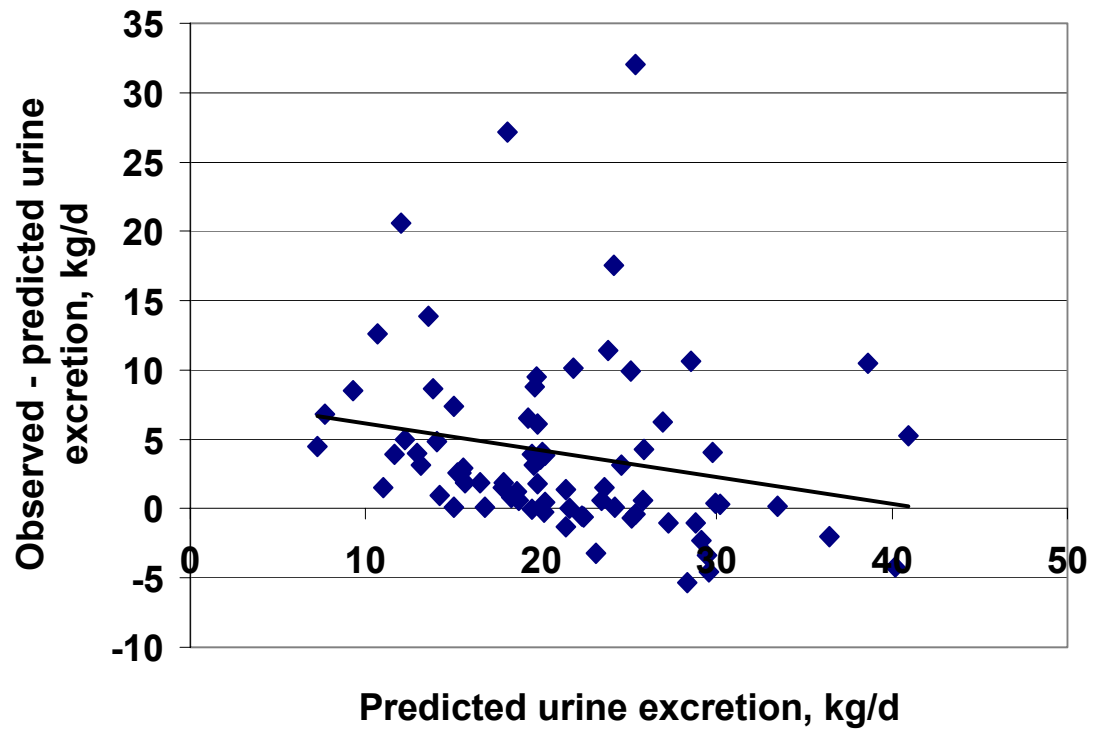


Figure 2-3. Plot of residuals (observed – predicted) versus predicted values of urine excretion (kg/d) from evaluation of the equation [Urine excretion, kg/d = 0.1343 x urinary Na, g/d + 0.0612 x urinary K, g/d + 0.0239 x urinary N, g/d] by Bannink et al., 1999. The equation was evaluated using the MINERAL dataset. The solid line on the graph represents the equation  $y = 4.19(\pm 1.36) - 0.279(\pm 0.116)(X - 21.2)$ . Evaluation of the equation resulted in significant mean (4.19) and linear biases (-0.279).

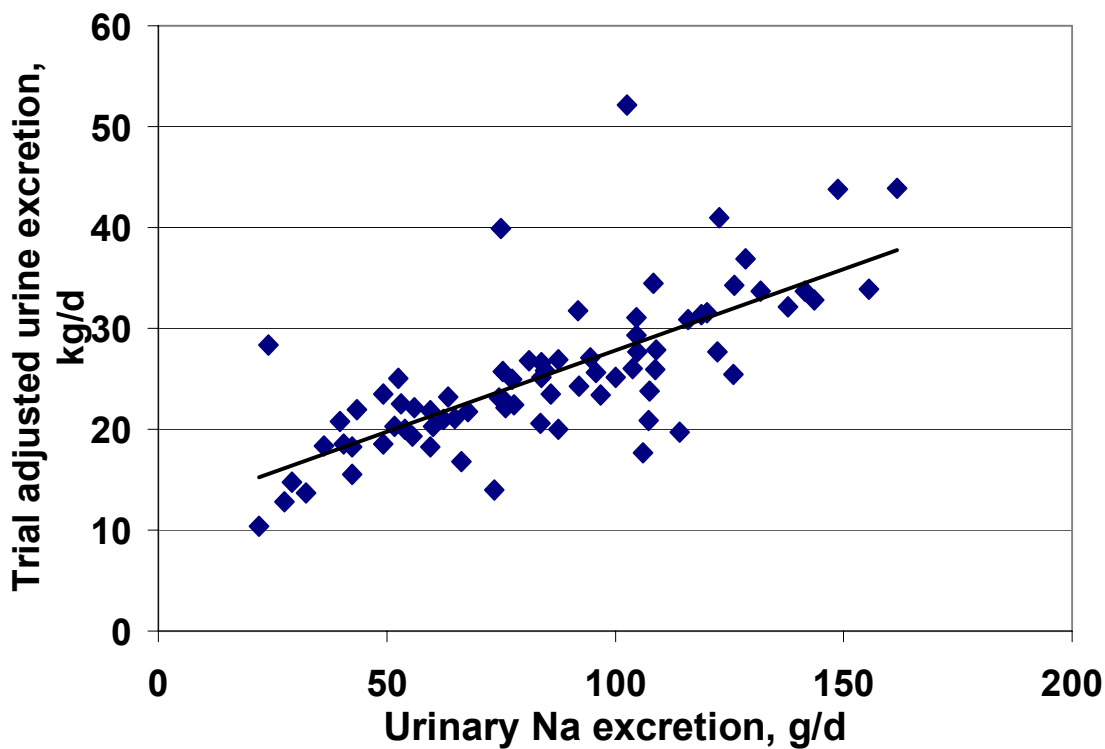


Figure 2-4. Relationship between the urinary Na excretion (g/d) and trial adjusted urine excretion (kg/d) for lactating cows (MINERAL dataset, n = 76). The solid line is equal to  $\text{Urine excretion (kg/d)} = (0.162 \times \text{Urinary Na, g/d}) + 11.6$ , Residual SE = 5.5, Inter-study SE = 4.6.



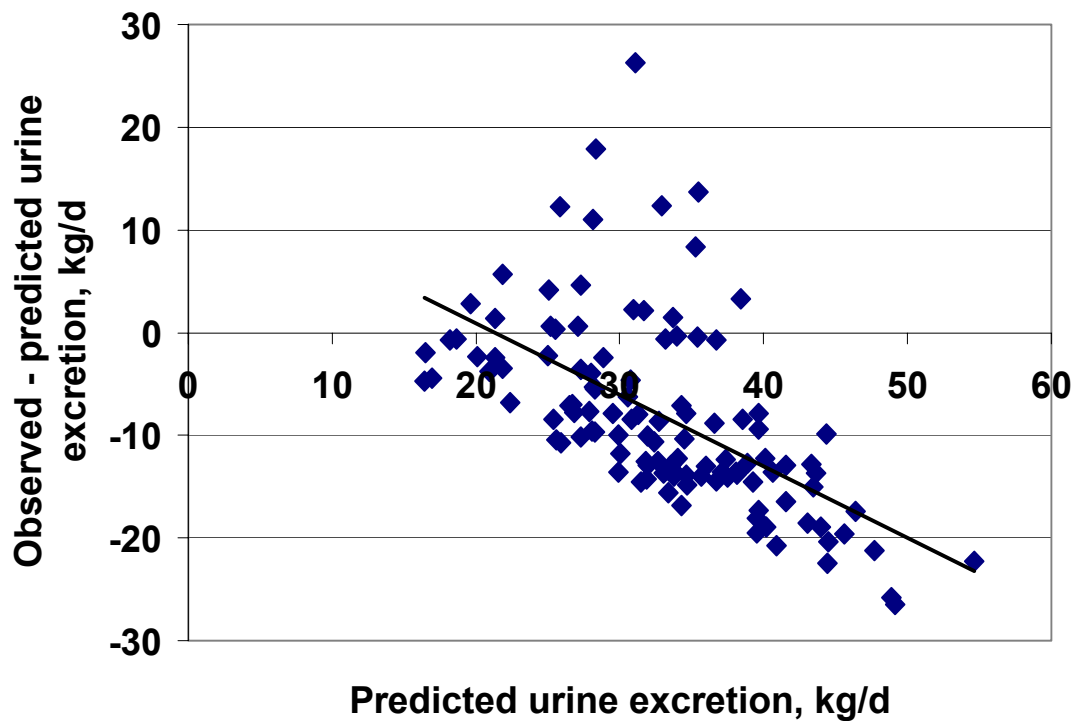


Figure 2-5. Plot of residuals (observed – predicted) versus predicted values of urine excretion (kg/d) from evaluation of the equation [Urine excretion, kg/d = 0.1153 x Na intake, g/d + 0.0577 x K intake, g/d] by Bannink et al., 1999. The equation was evaluated using the MINERAL dataset. The solid line on the graph represents the equation  $y = -7.35(\pm 1.9) - 0.695(\pm 0.092)(X - 33)$ . Evaluation of the equation resulted in significant mean (-7.35) and linear biases (-0.695).

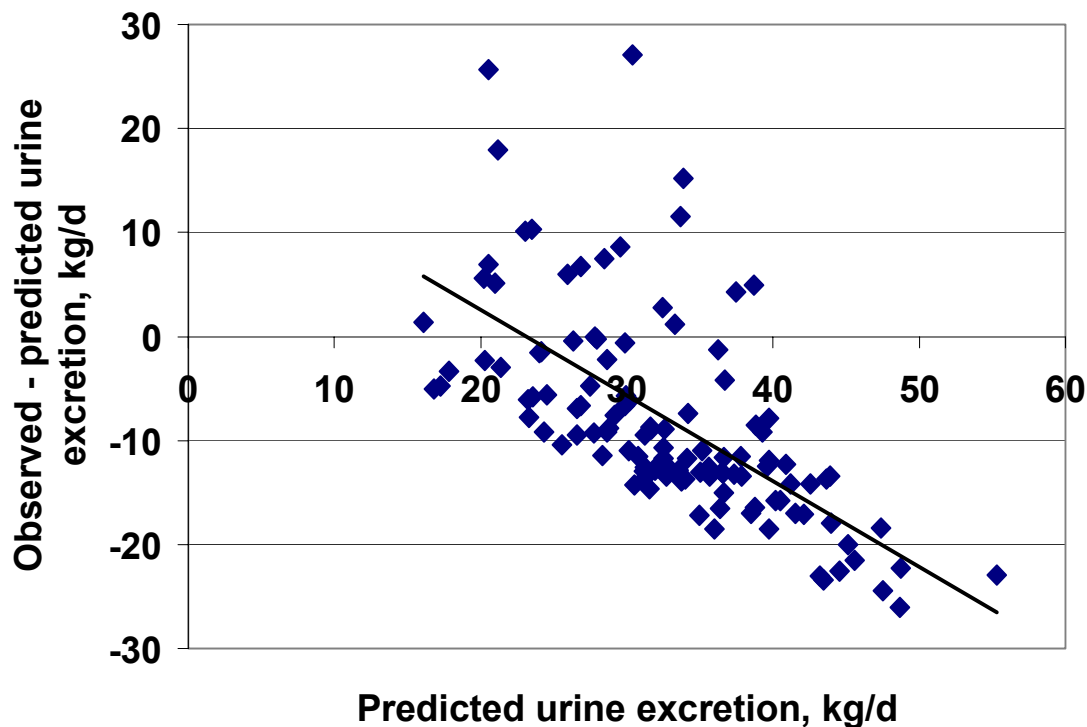


Figure 2-6. Plot of residuals (observed – predicted) versus predicted values of urine excretion (kg/d) from evaluation of the equation [Urine excretion, kg/d = 1.3442 + Dry matter intake, kg/d x (1.079 x Dietary Na, % + 0.5380 x Dietary K, % + 0.0203 x Dietary CP, % - Milk, kg/d x (0.1216 + 0.0275 x Milk protein, %)] by Bannink et al., 1999. The equation was evaluated using the MINERAL dataset. The solid line on the graph represents the equation  $y = -7.06(\pm 2.1) - 0.693(\pm 0.091)(X - 32.8)$ . Evaluation of the equation resulted in significant mean (-7.06) and linear biases (-0.693).

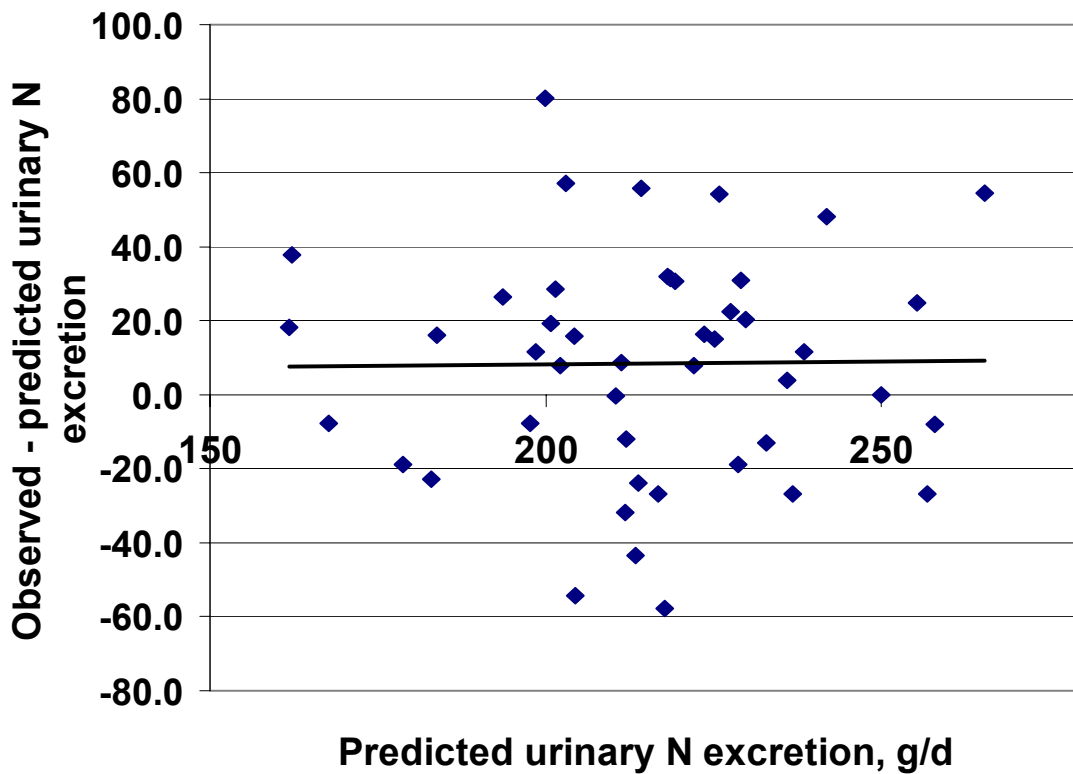


Figure 2-7. Plot of residuals (observed – predicted) versus predicted values of urinary N excretion (g/d) from evaluation of the equation [Urinary N excretion, g/d = (0.063 x RDP supplied, g/d) + 55.6] developed using the LACT dataset. The equation was evaluated using the VALIDATE dataset. The solid line on the graph represents the equation  $y = 0.0166x + 4.83$ .

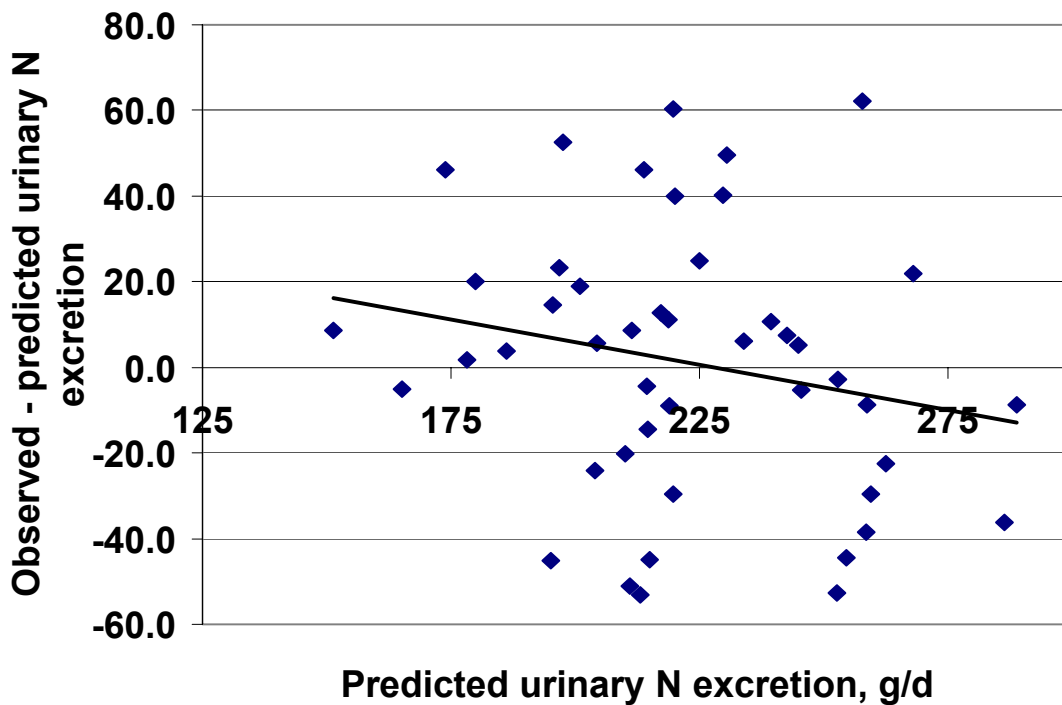


Figure 2-8. Plot of residuals (observed – predicted) versus predicted values of urinary N excretion (g/d) from evaluation of the equation [Urinary N excretion, g/d = (0.254 x BW, kg) – (1.03 x MILK, kg/d) + (210.1 x NI, g/d) + (5.09 x MUN, mg/dl) + (21.8 x MTP, %) – (6.5 x MF, %) – 138.8] developed using the LACT dataset. The equation was evaluated using the VALIDATE dataset. The solid line on the graph represents the equation  $y = -0.0212x + 48.34$ .

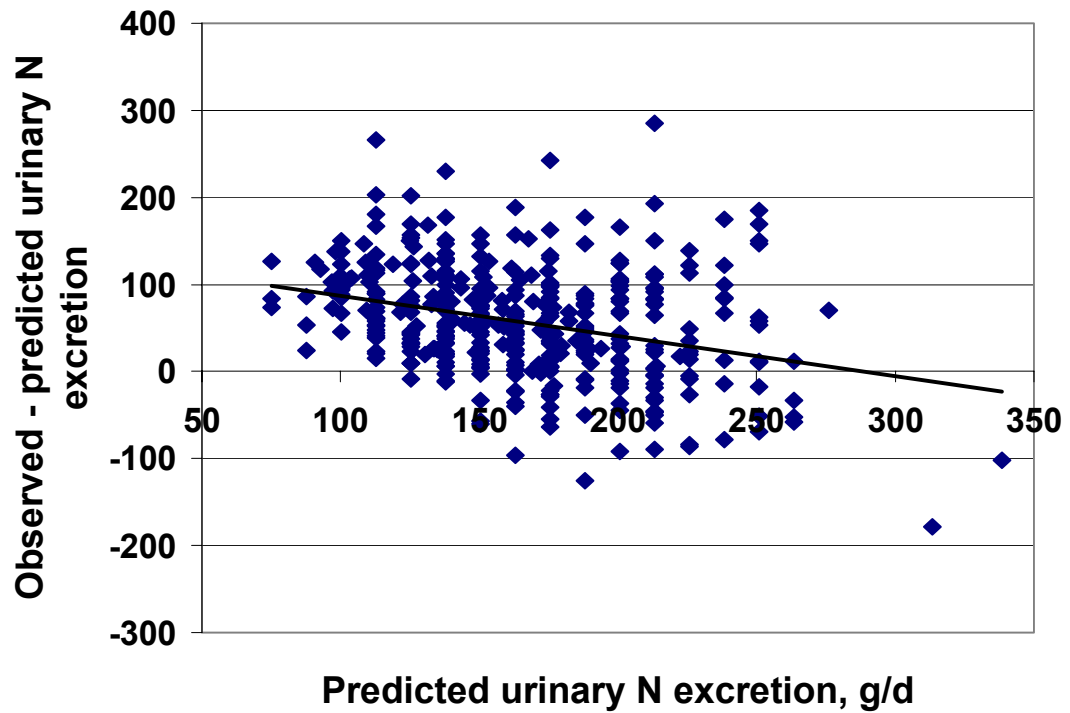


Figure 2-9. Plot of residuals (observed – predicted) versus predicted values of urinary N excretion (g/d) from evaluation of the equation [Urinary N, g/d = (12.54 x MUN, mg/dl)] by Jonker et al., 1998. The equation was evaluated using a combination of the LACT and VALIDATE datasets. The solid line on the graph represents the equation  $y = 57.3(\pm 10.8) - 0.549(\pm 0.070)(X - 165)$ . Evaluation of the equation resulted in significant mean (57.3) and linear biases (-0.549).

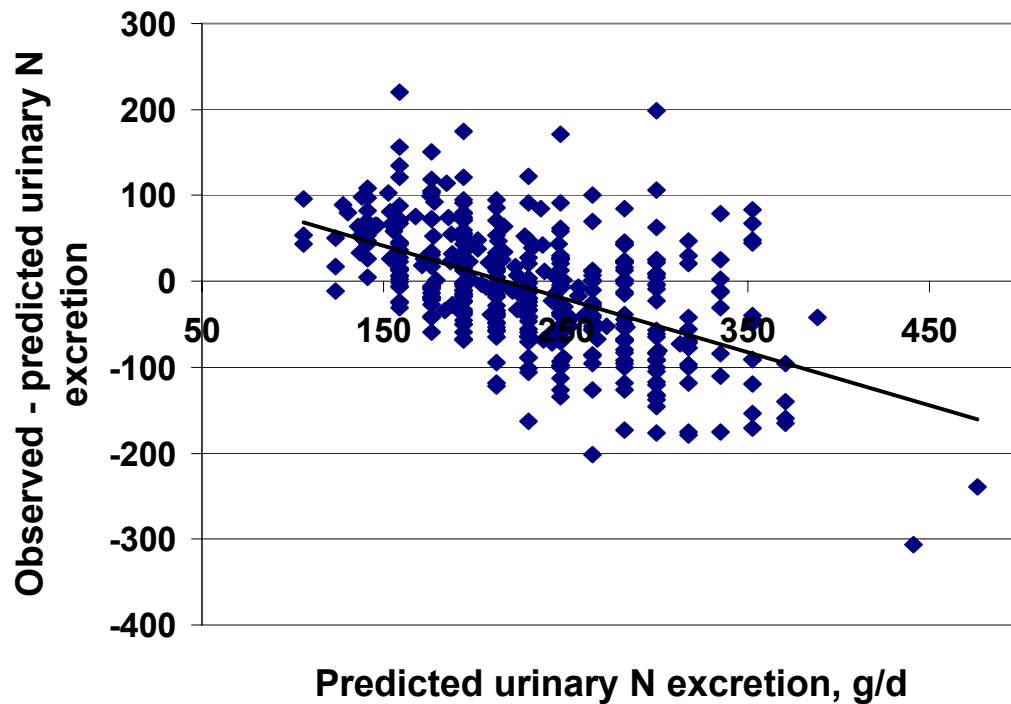


Figure 2-10. Plot of residuals (observed – predicted) versus predicted values of urinary N excretion (g/d) from evaluation of the equation [Urinary N, g/d = (17.64 x MUN, mg/dl)] by Kauffman and St-Pierre, 2001. The equation was evaluated using a combination of the LACT and VALIDATE datasets. The solid line on the graph represents the equation  $y = -9.7(\pm 10.8) - 0.679(\pm 0.050)(X - 231)$ . Evaluation of the equation did not result in a mean bias. The linear bias was significant ( $-0.679$ ;  $P < 0.01$ ).

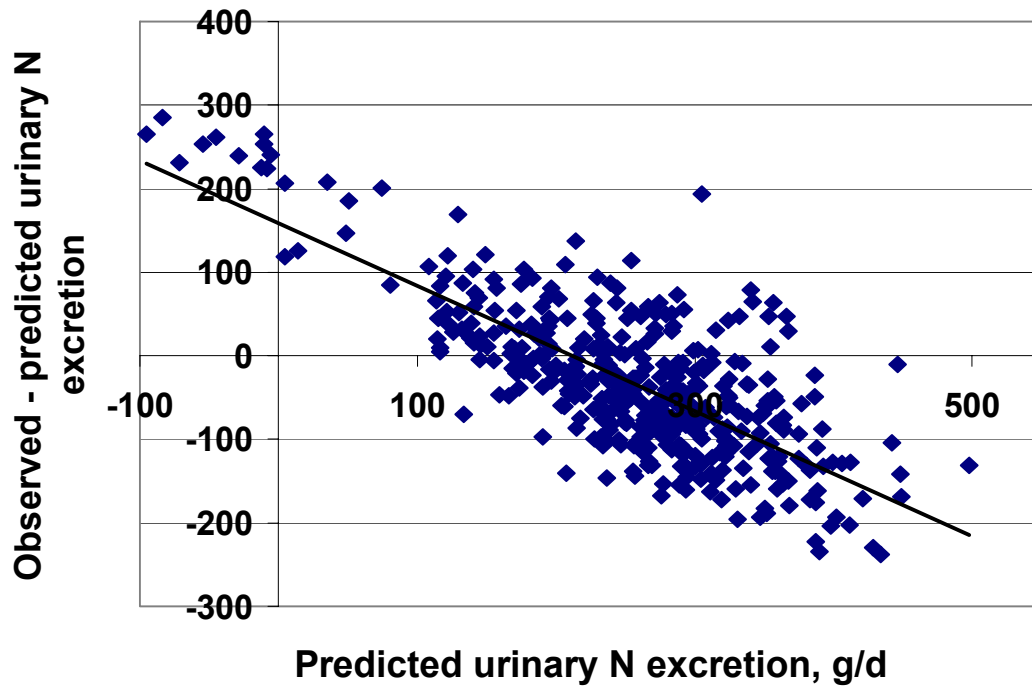


Figure 2-11. Plot of residuals (observed – predicted) versus predicted values of urinary N excretion (g/d) from evaluation of the equation [Urinary N, g/d = (0.83 x N intake – Milk N – 97)] by Jonker et al., 1998. The equation was evaluated using a combination of the LACT and VALIDATE datasets. The solid line on the graph represents the equation  $y = -18.0(\pm 12.5) - 0.652(\pm 0.031)(X - 255)$ . Evaluation of the equation did not result in significant mean and linear biases. The linear bias was significant ( $-0.652$ ;  $P < 0.01$ ).

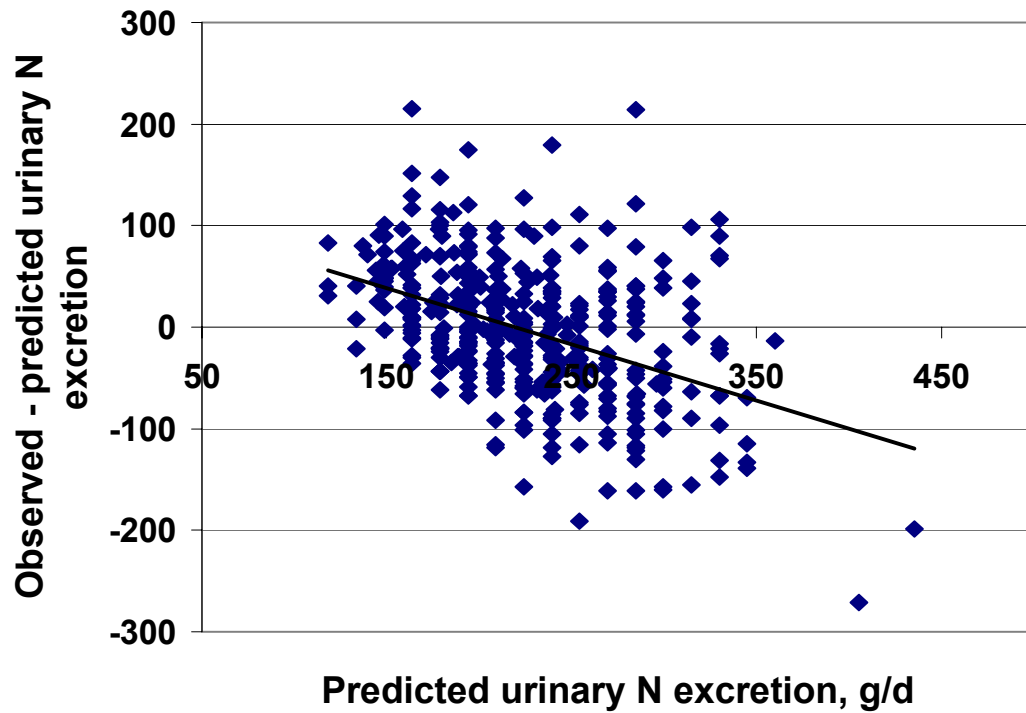


Figure 2-12. Plot of residuals (observed – predicted) versus predicted values of urinary N excretion (g/d) from evaluation of the equation [Urinary N, g/d = (15.1 x MUN, mg/dl + 27.8)] by Kohn et al., 2002. The equation was evaluated using a combination of the LACT and VALIDATE datasets. The solid line on the graph represents the equation  $y = -4.1(\pm 10.8) - 0.626(\pm 0.058)(X - 226)$ . Evaluation of the equation did not result in significant mean and linear biases. The linear bias was significant ( $-0.626$ ;  $P < 0.01$ ).



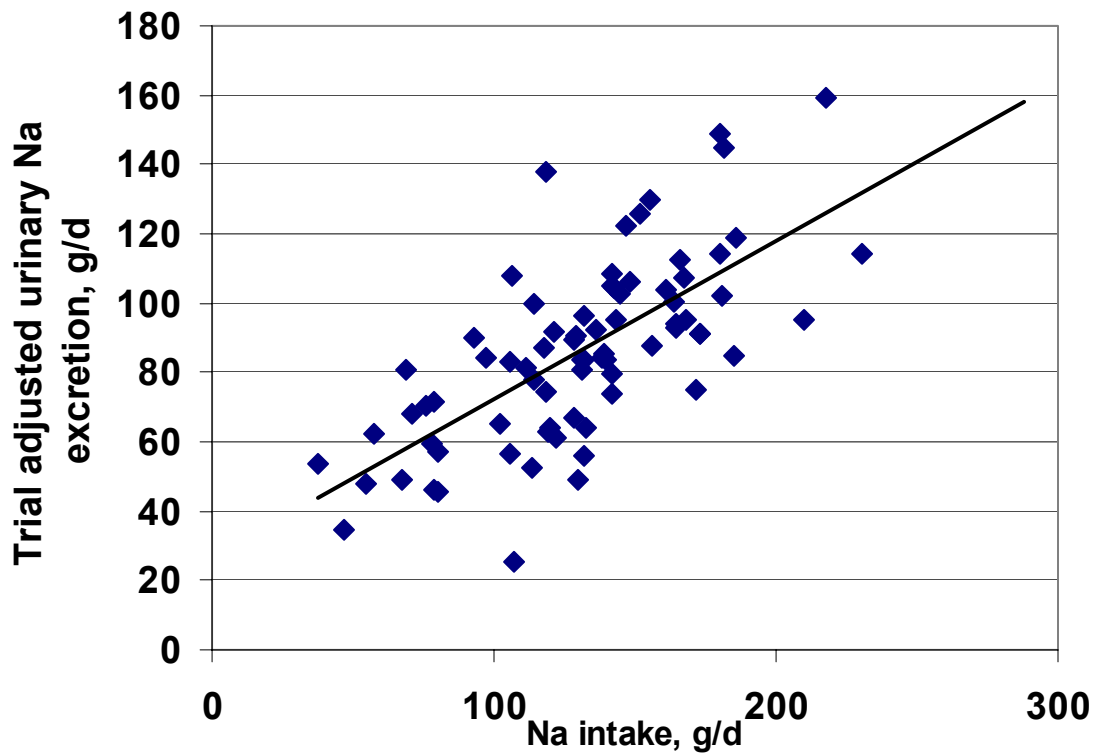


Figure 2-13. Relationship between the Na intake (g/d) and trial adjusted urinary Na excretion (g/d) for lactating cows (MINERAL dataset, n = 76). The solid line is equal to Urinary Na excretion (kg/d) = (0.456 x Na intake, g/d) + 26.6, Residual SE = 20.0, Inter-study SE = 18.5.

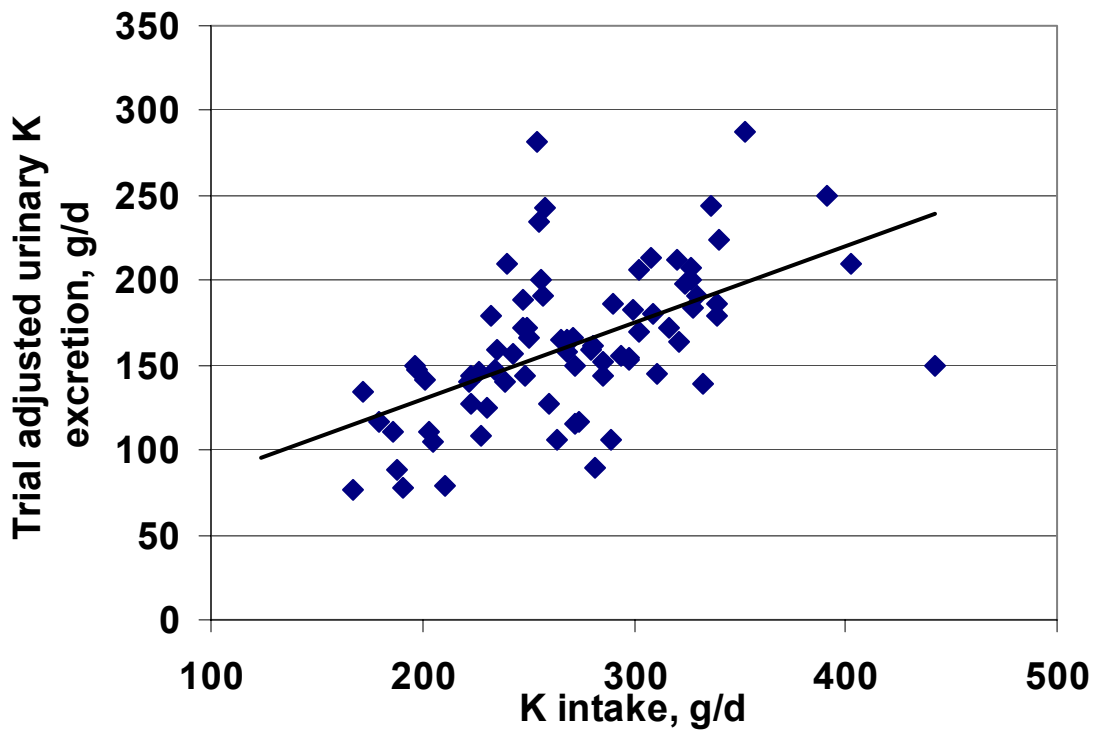


Figure 2-14. Relationship between the K intake (g/d) and trial adjusted urinary K excretion (g/d) for lactating cows (MINERAL dataset, n = 76). The solid line is equal to Urinary K excretion (kg/d) = (0.451 x K intake, g/d) + 40.2, Residual SE = 38.8, Inter-study SE = 41.6.

## **APPENDIX 3**

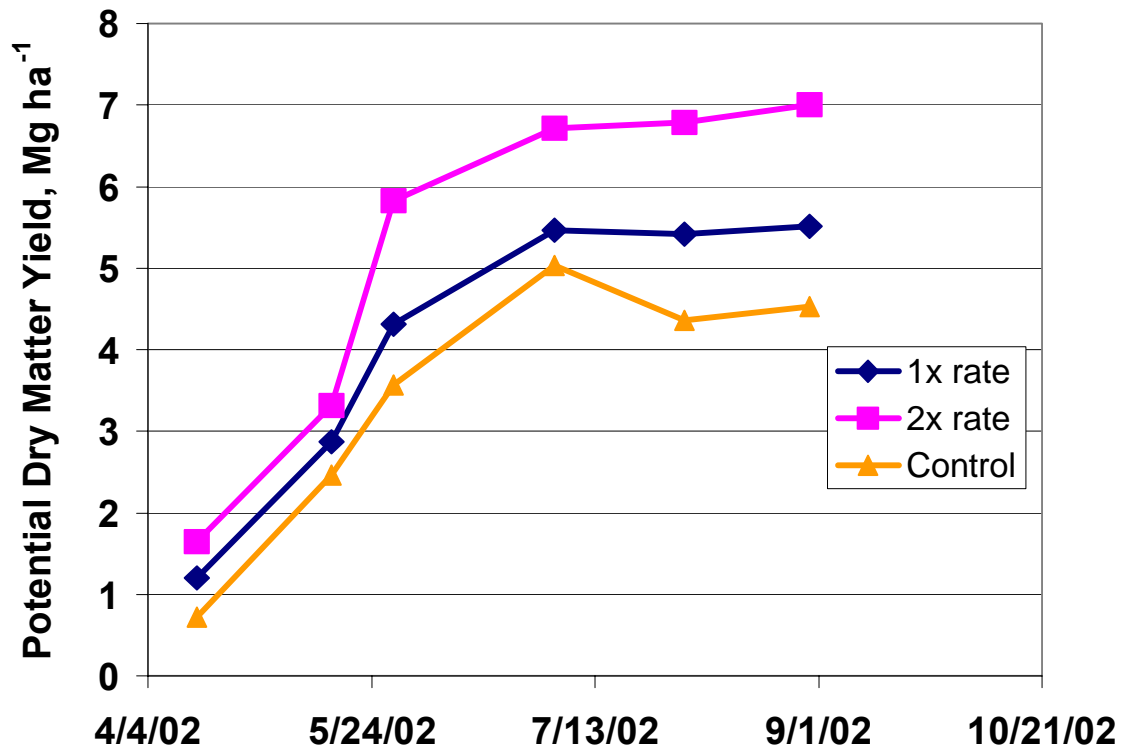


Figure 3-1. Potential forage dry matter yields at each grass clipping for plots receiving no slurry (control), 1x, or 2x slurry application rates in 2002. Slurry was applied to 1x and 2x plots on 15 Jan. 2002 and on 7 June 2002. The plots were harvested for hay on 30 May 2002.

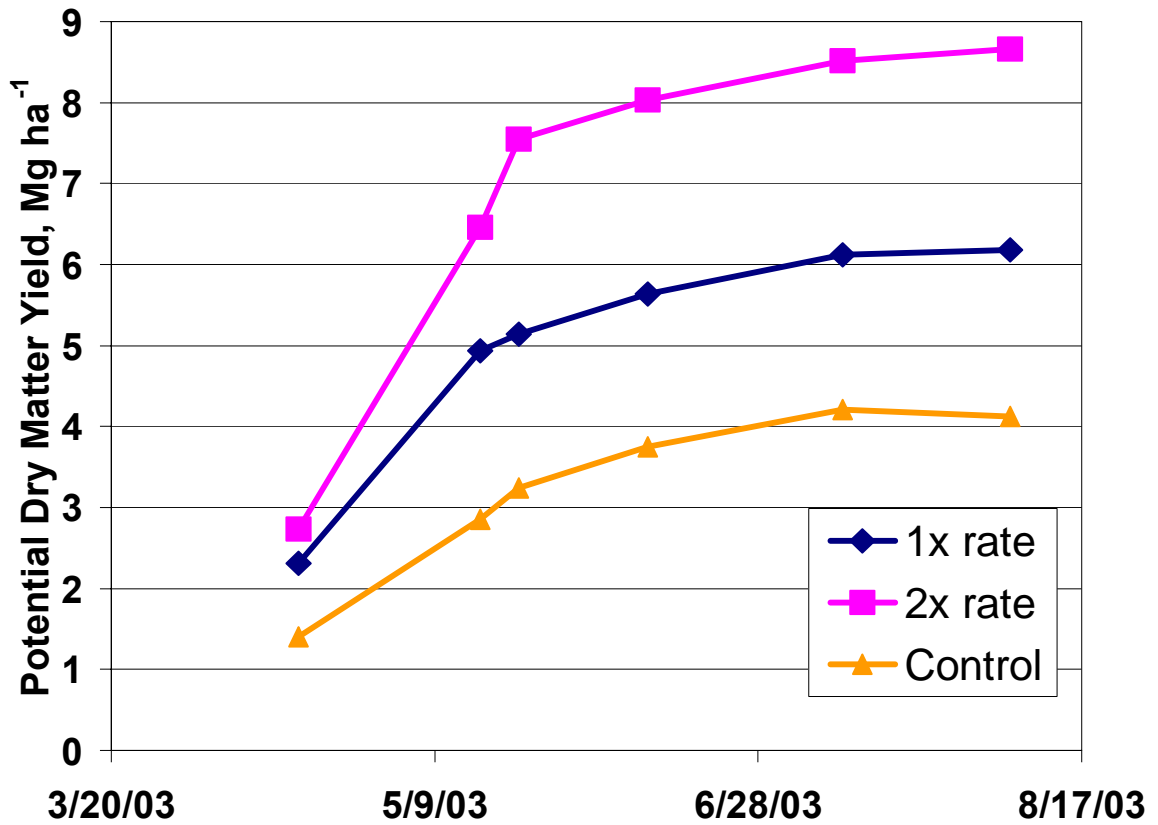


Figure 3-2. Potential forage dry matter yields at each grass clipping for plots receiving no slurry (control), 1x, or 2x slurry application rates in 2003. Slurry was applied to the 1x and 2x plots on 10 Jan. 2003 and on 11 June 2003. The plots were harvested for hay on 23 May 2003.

# **Winter Application of Dairy Slurry on a Grazing Based Dairy: 1.) Evaluation of Nitrogen Use in a Native Pasture in Year 3.**

## **MATERIALS AND METHODS**

### **Year 3**

Dairy slurry was applied to each of the plots during the January application. For the spring manure application, 1x and 2x treatment plots were divided into two sections. Dairy slurry was only applied to one-half of each of the 1x and 2x plots. The second half of the plot did not receive any slurry during the spring application period. The control plots remain unchanged with no slurry application. Application rates of dairy slurry are given in Table 3-1.

## **RESULTS AND DISCUSSION**

In 2004, the grass began to grow earlier in the season and resulted in the first clipping of grass being taken approximately 1 mo earlier in the spring than in 2002 or 2003. In addition, grass growth was sufficient for 2 cuttings of hay to be harvested from the plots as compared to one cutting taken in each of the previous years. After the second cutting of grass from the plots, the plots did not receive enough rainfall for further grass growth and no further grass clippings were taken from the plots.

In the 2x plots, forage DM yields were greater in 2004 than in 2003 or 2002. In 2004, the spring slurry application appeared to positively affect the grass DM yields (Figure 3-3).

Concentrations of soil  $\text{NO}_3\text{-N}$  declined slightly for the control, 1x, and 2x treatments. In contrast, soil  $\text{NH}_4\text{-N}$  concentrations increased in 2004 as compared to the two previous years. However, soil  $\text{NH}_4\text{-N}$  concentrations were also increased in control plots, thus the increase did not appear to be directly related to the applications of dairy slurry.

Table 3-1. Application rates and composition of dairy slurry applied to plots during 2004. Winter 1x and 2x plots only received the winter application of dairy slurry.

Item	1x application rate	2x application rate
2004 winter application		
Application rate, Mg ha <sup>-1</sup>	32.9	60.5
Total N, kg ha <sup>-1</sup>	77.5	139.8
Ammonium N, kg ha <sup>-1</sup>	52.0	90.9
Hydrometer total N, kg ha <sup>-1</sup>	91.4	273.8
Agros meter ammonium N, kg ha <sup>-1</sup>	37.7	73.8
2004 summer application		
Application rate, kg m <sup>-1</sup>	30.5	51.2
Total N, kg ha <sup>-1</sup>	68.7	122.9
Ammonium N, kg ha <sup>-1</sup>	50.6	82.4
Hydrometer total N, kg ha <sup>-1</sup>	67.1	98.2
Agros meter ammonium N, kg ha <sup>-1</sup>	43.1	64.5



Table 3-2. Total cumulative yields of dry matter for plots receiving no slurry (control), 1x or 2x rates only during the January application, or 1x or 2x application rates received during both January and May.

Item	control	Winter 1x rate	Winter 2x rate	1x rate	2x rate
First Cutting					
Dry matter, Mg ha <sup>-1</sup> 2004	2.22	NA	NA	4.87	6.66
Cumulative over season					
Dry matter, Mg ha <sup>-1</sup> 2004	4.76 <sup>b</sup>	8.29 <sup>ab</sup>	11.33 <sup>a</sup>	9.03 <sup>a</sup>	12.08 <sup>a</sup>

Table 3-3. Soil nitrate N and ammonia N for plots receiving no slurry (control), 1x, or 2x application rates in 2004.

Item	control	1x rate	2x rate	Yearly Average
Nitrate-N, mg kg <sup>-1</sup>				
2004	4.6	4.9	5.2	4.9
Ammonium-N, mg kg <sup>-1</sup>				
2004	12.8	16.5	19.6	16.3

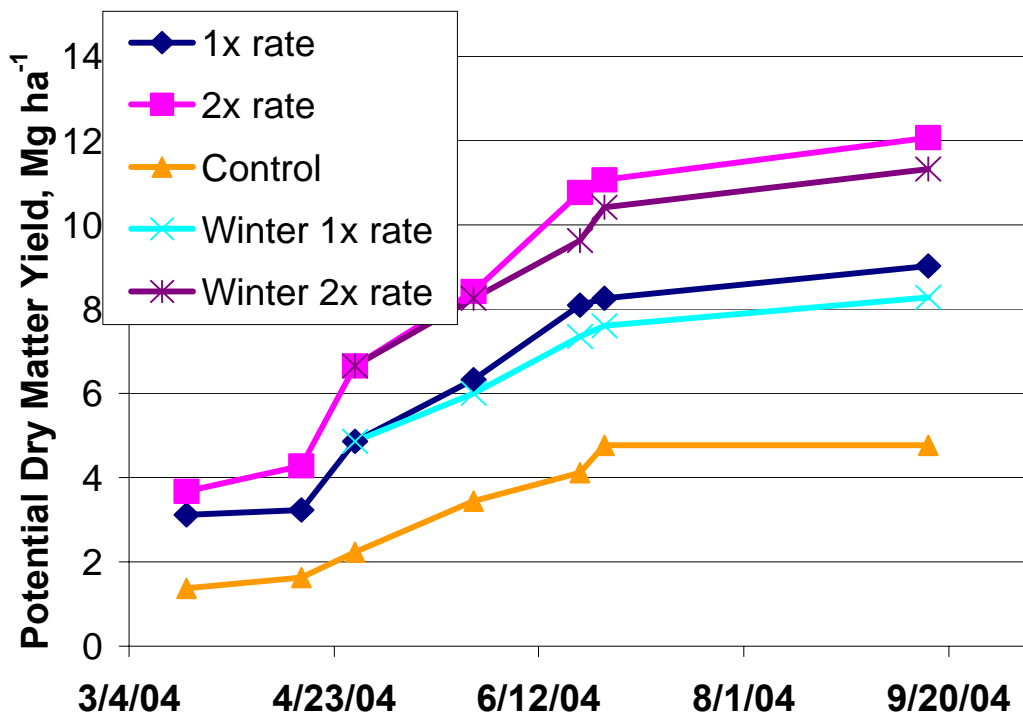


Figure 3-3. Potential forage dry matter yields at each grass clipping for plots receiving no slurry (control), 1x, or 2x slurry application rates in 2004. Slurry was applied to the 1x and 2x plots on 21 Jan. 2004 and on 7 May 2004. The plots were harvested for hay on 1 May 2004 and 28 June 2004.

## **APPENDIX 4**

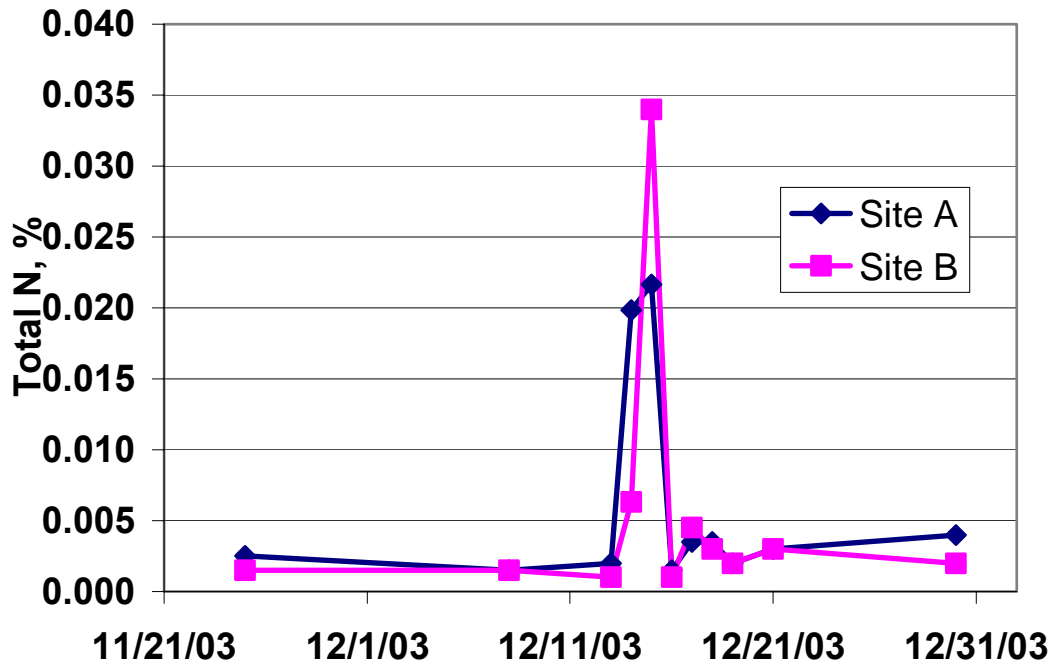


Figure 4-1. Total N concentration in the water samples taken at sites A and B after the December 12, 2003 slurry application.

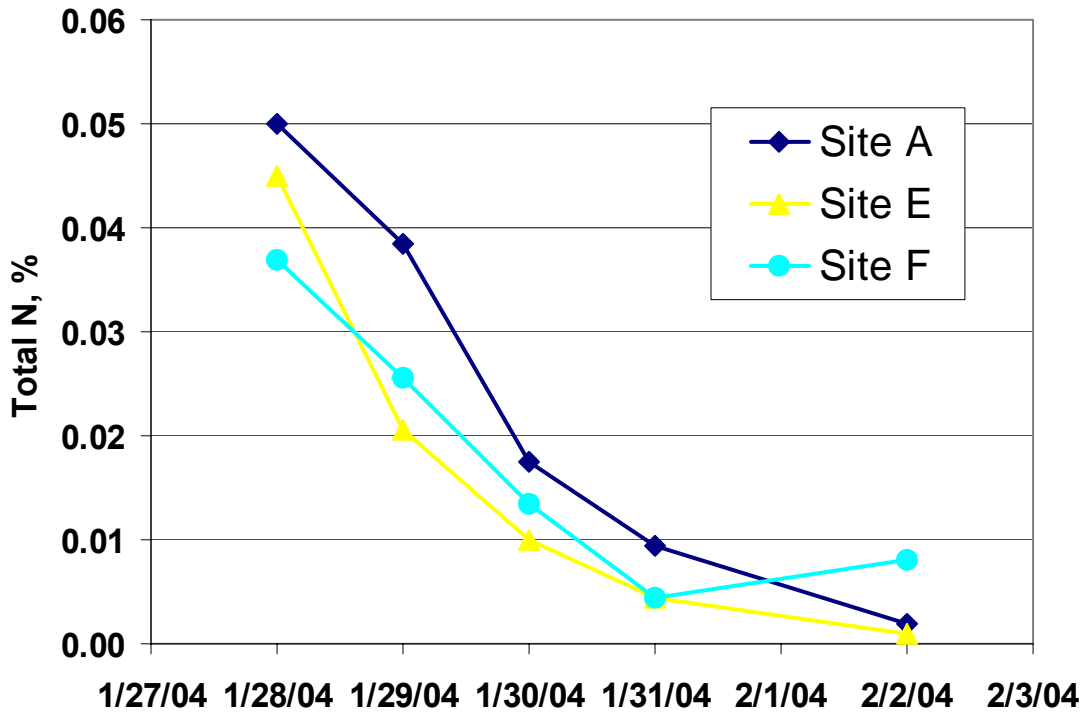


Figure 4-2. Total N concentration in the water samples taken at sites A, E, and F after the January 27, 2004 slurry application.

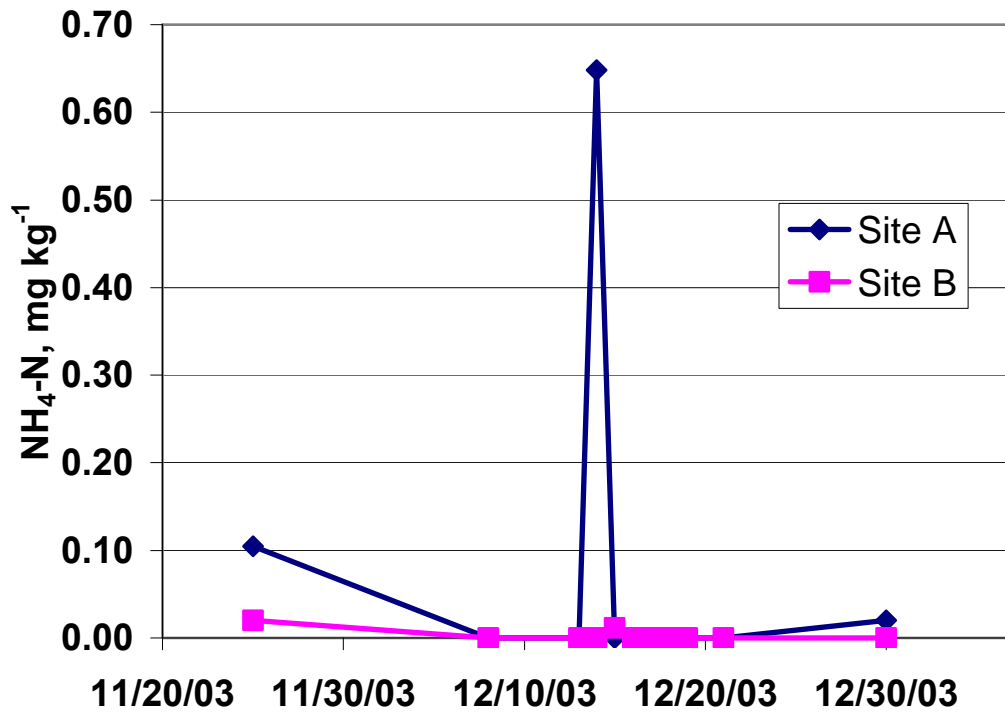


Figure 4-3. Ammonia-N concentration in the water samples taken at sites A and B after the December 12, 2003 slurry application.

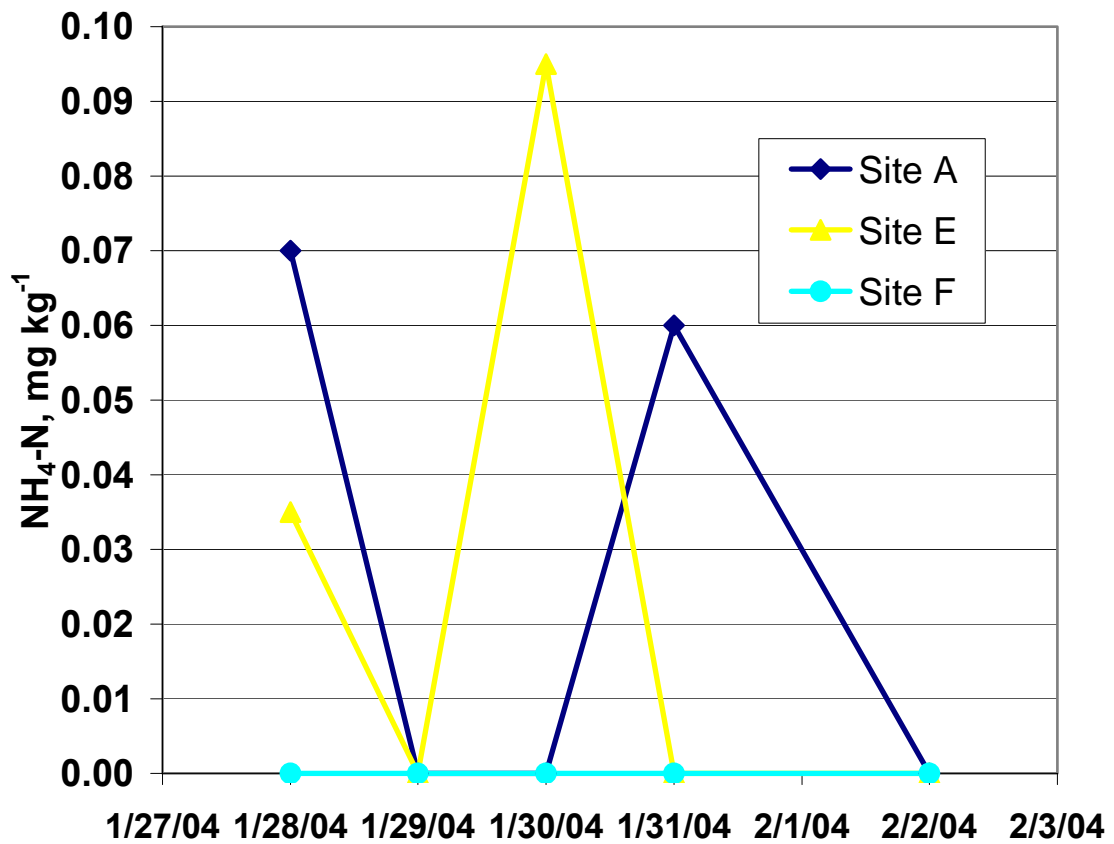


Figure 4-4. Ammonia N concentration in the water samples taken at sites A, E, and F after the January 27, 2004 slurry application.



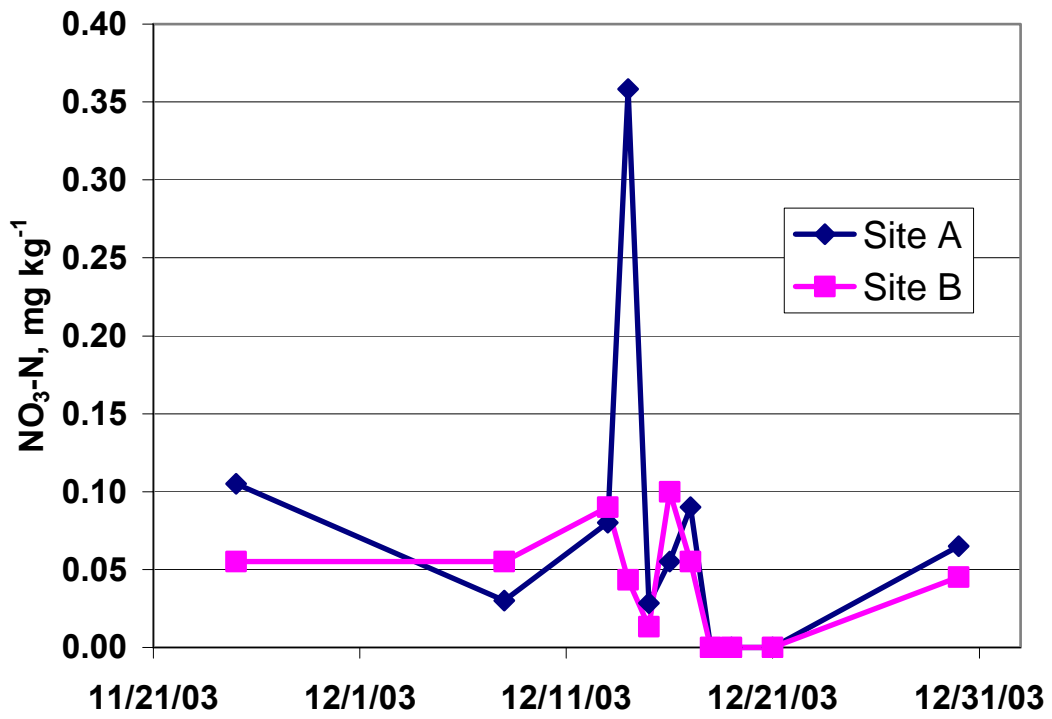


Figure 4-5. Nitrate-N concentration in the water samples taken at sites A and B after the December 12, 2003 slurry application.

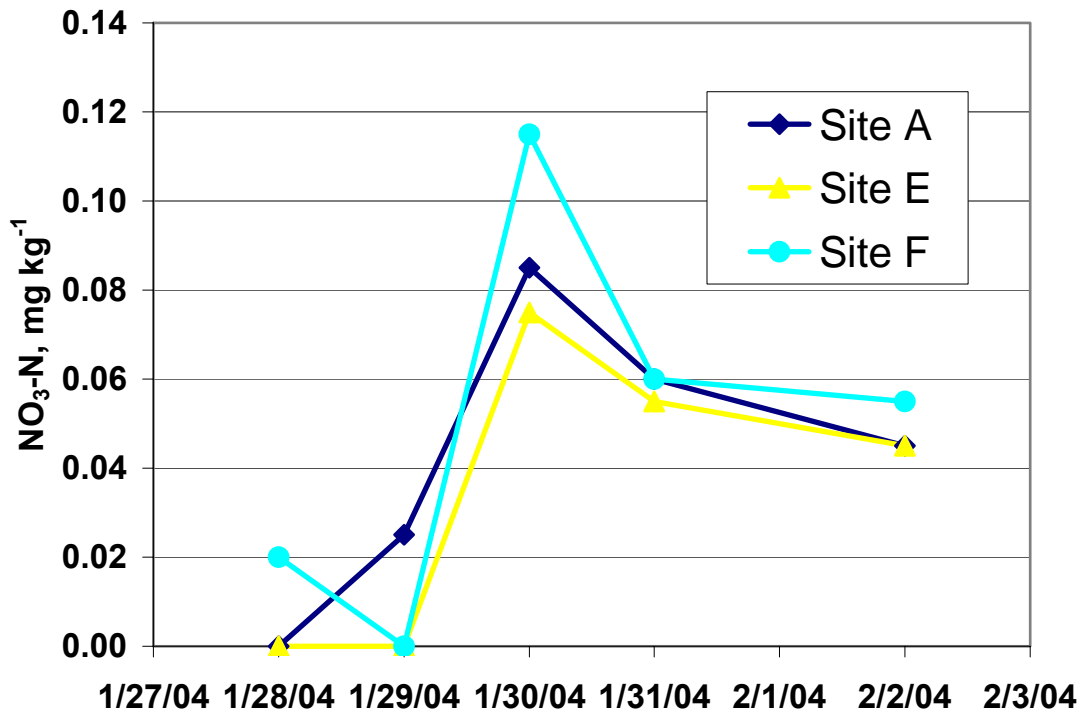


Figure 4-6. Nitrate-N concentration in the water samples taken at sites A, E, and F after the January 27, 2004 slurry application.

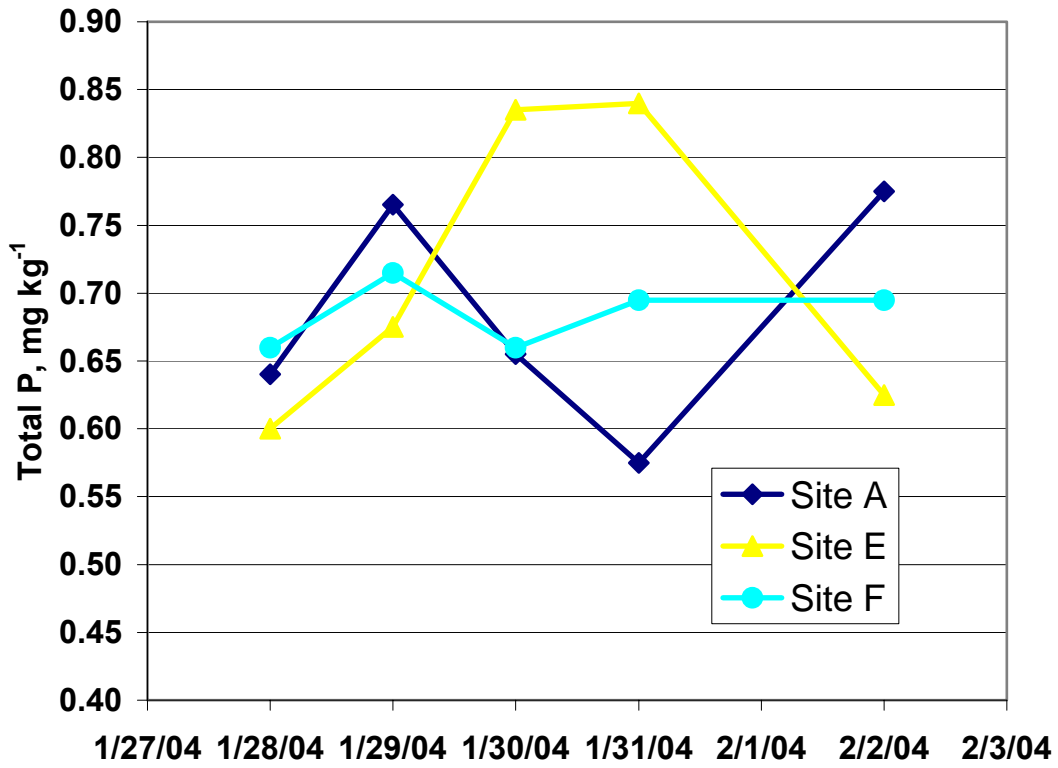


Figure 4-7. Total P concentration in the water samples taken at sites A, E, and F after the January 27, 2004 slurry application.

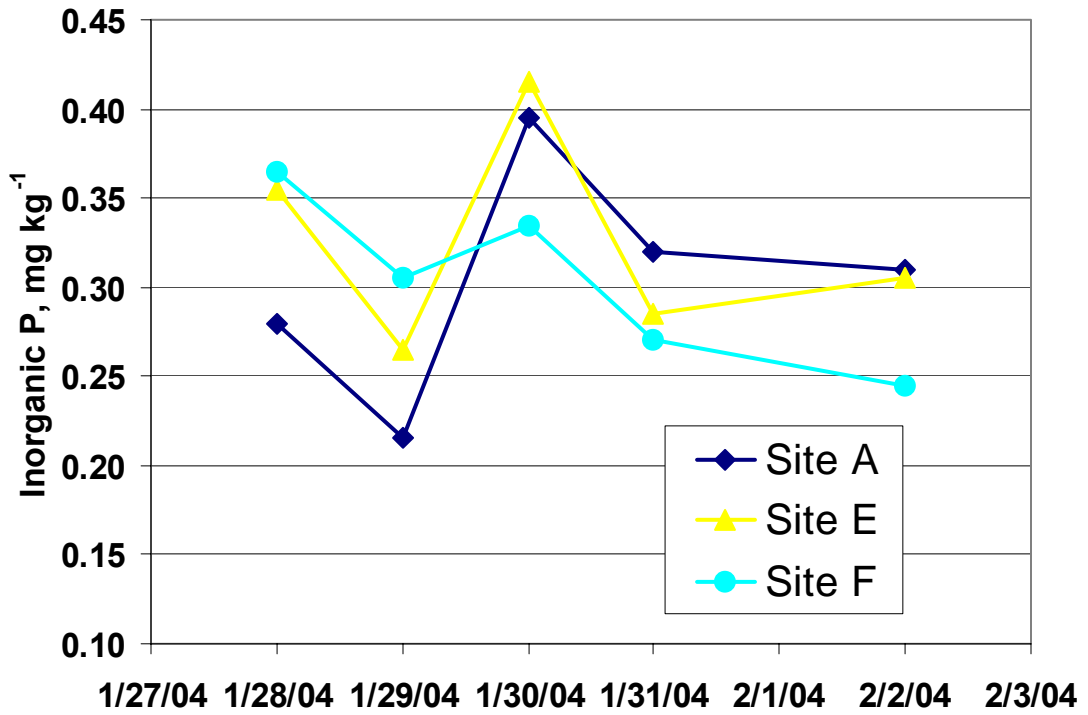


Figure 4-8. Inorganic P concentration in the water samples taken at sites A, E, and F after the January 27, 2004 slurry application.

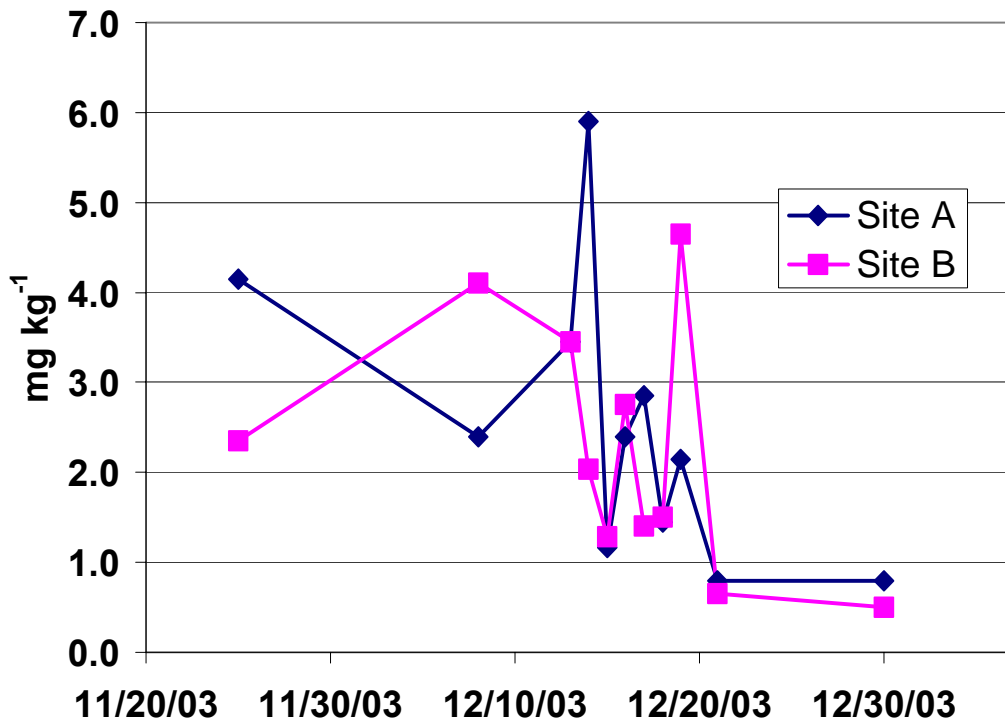


Figure 4-9. Potassium concentrations in the water samples taken at sites A and B after the December 12, 2003 slurry application.

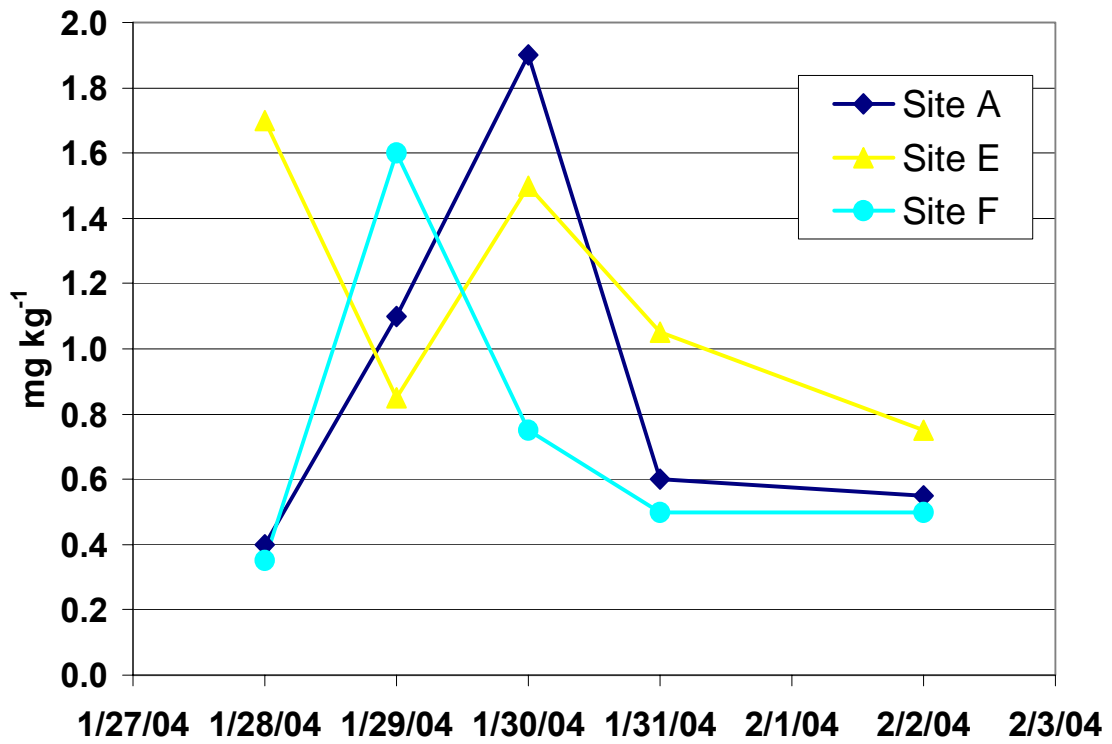


Figure 4-10. Potassium concentrations in the water samples taken at sites A, E, and F after the January 27, 2004 slurry application.

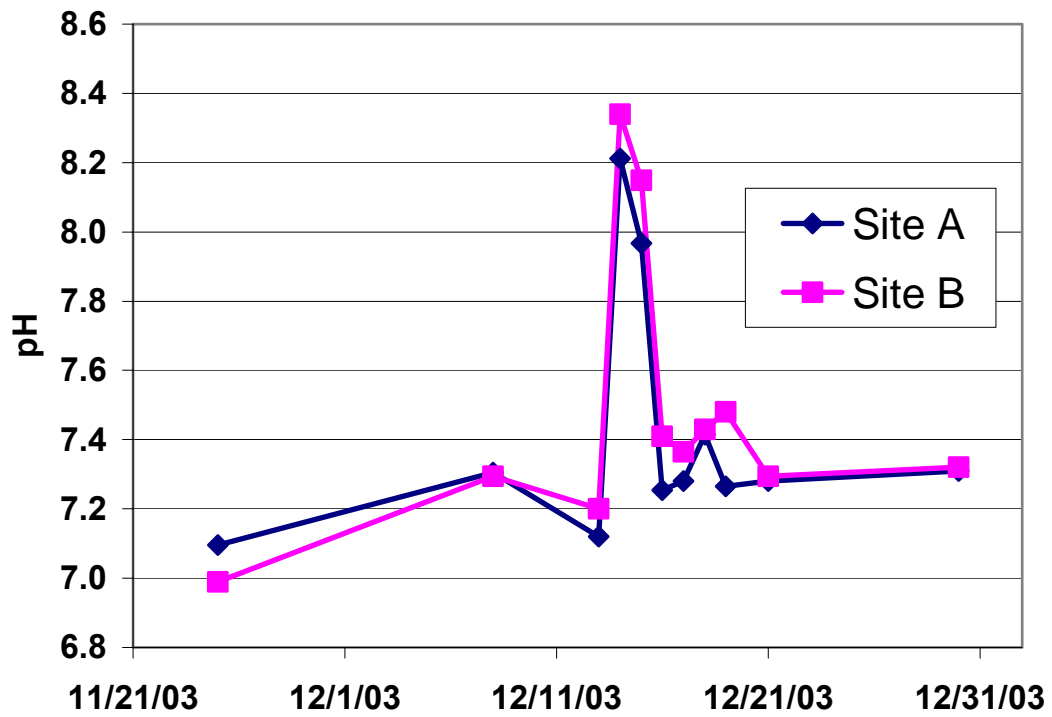


Figure 4-11. Water sample pH at sites A and B after the December 12, 2003 slurry application.

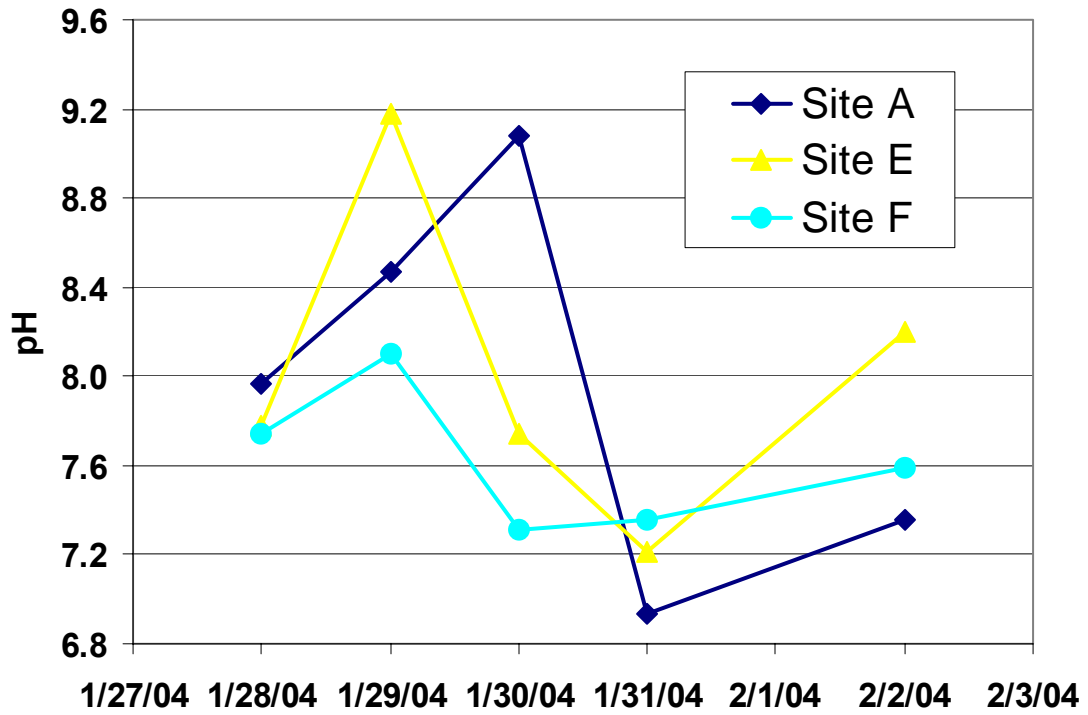


Figure 4-12. Water sample pH at sites A, E, and F after the January 27, 2004 slurry application.