EFFECTS OF NITROGEN FERTILIZATION ON PHOSPHORUS UPTAKE AND MOVEMENT AMONG FORAGE, SOIL AND WATER IN A YEAR-ROUND PASTURE SYSTEM

by

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Abstract

A 3-year study was conducted to investigate manipulation of nitrogen (N) fertilization regime and incorporation of seasonally adapted annual forages for re-establishing phosphorus (P) equilibrium and sustainability in a year-round grazed pasture system characterized by high background soil-test P. In the fall of 2009, 2010 and 2011, six 0.28-ha plots were overseeded with triticale (*Triticum secale*) and crimson clover (*Trifolium incarnatum*) into a tall fescue (*Lolium arundinacea*)/bermudagrass (*Cynodon dactylon*) sod and randomly assigned to 1 of 3 N fertilizer treatments (n = 2): 100% of N recommendation in a split application, 50% in a single application, and 0% of N recommendation for triticale. Cattle were placed into plots the following spring for grazing until May. In the summer, plots were overseeded with cowpea (*Vigna unguiculata*), fertilized at the same rates by reference to N recommendations for bermudagrass, and grazed by cattle until September. There were no effects of N fertilization rate on acid or alkaline soil phosphatase activity, soil pH, soil electrical conductivity, or soil concentrations of water-soluble P or N. Concentration of extractable soil-P was decreased in plots receiving 50% of the recommendation for N. However, increasing N fertilization to 100% of recommendation in a split application resulted in no further reduction in soil-test P. Forage mass, foliar P and N concentrations and forage P mass were not affected by N fertilization rates at the plant-community level;, but responses were observed within individual forage species. Forage characteristics were

affected more by growing season and forage management than by fertilization, similar to effects observed for soils, suggesting that foliar P mass could be increased with minimal N inputs, use of seasonally adapted annual forages and incorporation of grazing cattle. Runoff water from plots contained greater concentrations of extractable P, PO_4-P and NH_4-N in unfertilized than fertilized paddocks, possibly due to lower pasture productivity. Neither N fertilization regime nor grazing season affected intake or fecal excretion of water-soluble P by grazing cattle, but total P excretion was greater with greater N fertilization in the spring grazing period. Cattle P requirements for growth were met by grazed forage, and sufficient P was returned to pasture to meet forage requirements, with no N fertilization. There was no effect of N fertilization on rates of decomposition or P disappearance from fecal pats, or on P concentration in soil beneath decomposing fecal pats; however, N fertilization increased N removal from and increased soil N concentrations beneath fecal pats. Results are interpreted to mean that, in order to effectively decrease nutrient concentrations in water runoff and to increase forage P mass and soil quality in a year-round grazed grass-legume pasture, N fertilization at no more than 50% of the recommended rate for the seasonally-adapted grass species within the stand was sufficient. Also, intake and fecal returns of P and foliar uptake of P were causally associated with N status of the P-enriched pasture system, but rate and extent of assimilation of P returns into the soil profile from degradation of fecal material were not.

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0N	0% of nitrogen recommendation
50N	50% of nitrogen recommendation in a single application
100N	100% of nitrogen recommendation in a split application
ADF	Acid detergent fiber
ADG	Average daily gain
ADL	Acid detergent lignin
BW	Body weight
C	Celsius
Ca	Calcium
Cl	Chlorine
cm	Centimeter
CP	Crude protein
Cr	Chromium
CS	Cool season
Cu	Copper
d	Day
DAA	Days after application
df	Degrees of freedom
DM	Dry matter

DNA	Deoxyribonucleic acid
EC	Electrical conductivity
g	Gram
h	hour
ha	hectare
hd	Head
ICAP	Inductively couple argon plasma
IVDMD	*In vitro* dry matter digestibility
K	Potassium
kg	Kilogram
L	liter
m	Meter
Mg	Megagram
Mg	Magnesium
mg	milligram
mM	millimoles
mm	millimeter
mol	mole
Mt	Megaton
N	Nitrogen
NDF	Neutral detergent fiber
O	Oxygen
P	Phosphorus

P_i	Inorganic phosphorus
pNP	para-Nitrophenol
ppm	Parts per million
RNA	Ribonucleic acid
S	Sulfur
US	United States
wk	Week
WS	Warm season
yr	Year
Zn	Zinc
µg	Microgram
µmol	Micromoles
µohms	Micro ohms

I. LITERATIURE REVIEW

Ruminant nutrient utilization

Nitrogen. Ruminant animals metabolize N in a unique manner that enables them to recycle N, primarily in the form of urea. Like most animals, the major source of N in the ruminant diet is protein. Due to the function of rumen microorganisms, ruminants can subsist without a source of pre-formed dietary protein and, because of recycling of N through saliva, ruminants can survive for extended periods of time with very little dietary N (Owens and Zinn, 1988).

Rumen microbes are able to utilize dietary nonprotein N and recycled urea N for amino acid and protein synthesis. Cattle can therefore grow, reproduce and lactate when the diet contains nonprotein N as the sole source of N (Owens and Zinn, 1988). Plants provide 60 to 80% of their total plant N as true protein; the remainder is mostly soluble nonprotein N, with a small fraction of insoluble nitrogenous compounds associated with plant cell walls (Van Soest, 1994). Rumen microbial metabolism is the sole means of converting nonprotein N into high-quality protein for use by the host animal; therefore, it is important to consider their metabolic needs when feeding ruminant animals. Optimization of microbial yield requires optimal use of N, which is modulated via synchrony with carbohydrate fermentation and rate of rumen outflow. Also, high rates of feed intake result in more rapid microbial growth and ruminal turnover rates (Van Soest,

1994). Animal protein requirements typically exceed those of the microbes. However, even in a growing animal, microbial output typically exceeds animal requirements and, therefore, there is often no growth response to supplementation with greater than 10 to 12% CP (Van Soest, 1994).

Nitrogen is recycled in the form of salivary urea that can diffuse across the rumen wall. Owens and Zinn (1988) state that 23 to 92% of plasma urea is recycled to the digestive tract, with higher values associated with decreased N intakes. With forage diets, typically 15 to 50% of urea is recycled via the saliva, with the remainder being diffused across the rumen wall directly from the plasma. Urea is utilized by urease-producing bacteria that make up approximately 10 to 15% of the bacteria that adhere to the rumen wall surface. Owens and Zinn (1988) also state that cattle can recycle up to 60 g N/d. After conversion by urease in the rumen, recycled N either is absorbed across the rumen wall as ammonia or becomes incorporated into microbial CP that can then be utilized by the animal. Once ammonia is in the blood, it is quickly converted back to urea by the liver to prevent any toxic side effects (Van Soest, 1994). Nearly all microbial protein is digested and absorbed in the small intestine of the animal, much like in monogastric animals (Owens and Zinn, 1988).

When dietary N intake exceeds the needs of both the rumen microbes and the animal tissues, the extra N is filtered by the kidneys and is excreted via the urine in the form of urea. However, a small amount of N is excreted through the feces. This fecal N is typically endogenous N, N from sloughed animal cells or from indigestible fractions of microbial cells (Van Soest, 1994). The fecal N fraction is not discretionary like urinary output, so it establishes the threshold for minimal obligatory dietary N intake because the

animal loses approximately 0.6% of dietary DM intake as fecal N losses (Van Soest, 1994).

Phosphorus. Metabolism of P is critical to extent of P excretion by ruminant animals. In grazing ruminants, P is one of the most common deficiencies because of the oftentimes low P concentrations in pasture forages, especially in arid and tropical regions (Kincaid, 1988). Concentration of P in forages ranges from 0.3% in early growth to 0.15% in mature growth, and may be deficient for cattle subsisting on mature, dry forages. Additionally, rumen microorganisms contain a high proportion of DNA and RNA that are both P-rich, causing their P requirements to be elevated (Van Soest, 1994) in relation to host-animal requirements. NRC (1996) states that the maintenance P requirement is 16 mg P/kg BW for beef cattle, which is based on fecal endogenous P levels that dictate the minimum P requirement. NRC (1996) also states that true absorption of P is only 68%, and it is even less from forage-based diets.

Similar to N, P can be recycled through the saliva in the form of phosphates. Phosphorus is absorbed in the ortho-phosphate form; however, pyro- and meta-phosphates can be absorbed, but are less biologically available. Phytate P, which is largely unavailable to monogastrics and makes up much of P in plants, can be hydrolyzed by rumen microorganisms (Kincaid, 1988). Phosphorus from the small intestine is recycled via the saliva and returned for use in microbial metabolism (Van Soest, 1994).

Phosphorus is typically excreted via the feces; however, a small portion of endogenous losses occur through the urine, typically 0.27 g P/d for cattle grazing a grass and legume pasture (Van Soest, 1994). Additionally, Van Soest (1994) states that fecal losses consist of endogenous losses from incompletely degraded microbial cell walls, as

well as indigestible forage components. When dietary intake of P exceeds animal and microbe requirements, extraneous P is excreted via the feces (Van Soest, 1994).

Agronomic characteristics and nutrient-uptake capacity of experimental forages

Tall Fescue. Tall fescue (*Lolium arundinacea*) originated in Europe and was first brought to North and South America in the late 1700s; however, not much attention was given to tall fescue until the release of two cultivars from Oregon State University and University of Kentucky. In 1940, Oregon State University released the tall fescue cultivar 'Alta,' and in 1943 the University of Kentucky released the cultivar 'Kentucky 31'(KY 31). William C. Johnston, Extension Specialist with the University of Kentucky, recommended KY 31 based on its dependability, adaptability to a wide range of soils, potential for year-round grazing and palatability for livestock. In 1940, only 16,000 ha in the US were planted in tall fescue; however, by 1973, 12 to14 million ha throughout the US were planted in tall fescue (Buckner et al., 1979).

There are currently over 14.2 million ha of tall fescue throughout the US, including over 121,000 ha within the State of Alabama (Ball et al., 2007). Tall fescue is a cool-season bunch grass that grows to a height of approximately 1 m, and it has been reported to have a rooting depth of up to 1.22 m with a total root mass of 10,000 kg/ha.(Ball et al., 2007). Tall fescue exhibits seasonal production from September through December and from March through June or July, and it can tolerate low-fertility and acidic soils; also, tall fescue responds exceptionally well to N fertilization. It is best adapted to clay or loam soils, tolerant of poor drainage and relatively tolerant to drought (Ball et al., 2007). Establishment guidelines include a seed drill rate of 17 to 22 kg/ha in

August, September or October; furthermore, because it is a perennial plant, it does not require reseeding once it is successfully established. Standard fertilization recommendation is approximately 112 kg N/ha in a split-application (Stricker et al., 1979). Other studies (Cogger et al., 2001; Archer and Decker, 1976) have shown that increasing N fertilization rate up to 403 kg N/ha can further increase forage DM yield.

Over the last 60 yr, tall fescue has become one of the most widely utilized cool-season pasture forages, due in large part to its high yield potential and nutritive quality for livestock. Balasko (1977) reported that, with a N application rate of 240 kg N/ha, tall fescue produced 10,290 kg DM·ha^{-1}·yr^{-1} cumulatively over 3 harvests, with an average IVDMD of 64.6% and 9.3% CP over an entire season. Similarly, Pedersen et al. (1990) reported that endophyte-infected KY 31 yielded 9,415 kg DM·ha^{-1}·yr^{-1}, and contained 17% CP and 57% IVDMD.

While tall fescue offers significant potential as a livestock feed, its relationship with the fungal endophyte *Neotyphodium coenophialum* contributes to ergot alkaloid toxicity in cattle and other livestock (Schmidt and Osborne, 1993). Fescue foot is caused by vasoconstriction in body part extremities, which can lead to gangrene and eventual loss of hooves and/or portions of ears or tails. Fescue toxicity is characterized by poor animal gains, reduced conception rates, intolerance to heat, failure to shed winter hair coat and elevated body temperatures. Due to the expansiveness of the fescue geographic range of adaptation, fescue toxicosis is considered the most common and economically important syndrome in cattle grazing tall fescue pastures (Ball et al., 2007).

Tall fescue is also important in its use as a phytoremediator of nutrient-enriched soils in areas of concentrated dairy, poultry and swine production where environmentally

benign disposal of manures can be problematic. Chenery et al. (2002) reported that a 3-yr old stand of tall fescue produced 9.87 Mg DM/ha when 286 kg N/ha was applied in the form of fresh dairy manure, which resulted in removal of 156.6 kg N/ha from the stand over the entire season. Additionally, in the following season with only 140 kg N/ha applied in the form of ammonium nitrate, the same plots produced 12.86 Mg DM/ha and removed 201.4 kg N/ha. The authors reported that manure residual effects were evident for at least the next 3 yr of growth due to the large organic N content of animal manures that was not immediately available for plant uptake. Sistani et al. (2006) reported that tall fescue fertilized with poultry litter at a rate of 4,480 kg litter/ha, equivalent to approximately 55 kg N/ha, produced 3,611 kg DM$\cdot$ha$^{-1}\cdot$yr^{-1} when harvested twice during a season. The fescue plots removed 23.87 kg N/ha; however, increasing tall fescue DM yield would have allowed for greater extent of N removal.

Animal manures are oftentimes land-applied to satisfy crop N requirements. However, N:P ratio of animal manures is typically narrower than that of the crops onto which they are applied, potentially causing P accumulation in soils; therefore, P phytoremediation is becoming increasingly important. Sistani et al. (2006) reported that tall fescue would remove 4.34 kg P$\cdot$ha$^{-1}\cdot$yr^{-1} when fertilized with poultry litter at a rate that delivered approximately 55 kg N/ha. Fescue fertilized with 150 kg N/ha and 150 kg P/ha in the form of ammonium nitrate and triple superphosphate removed 14.1 kg P/ha (Sikora and Enkiri, 2005). These studies indicate that, with adequate N and excessive P, tall fescue has the ability to remove some amount of P from the soil.

Triticale. Triticale (×*Triticosecale rimpaui* Wittmack) is a hybrid of wheat (*Triticum*) and rye (*Secale*) (Briggle, 1969). Triticale first appeared in the scientific

literature in 1876 and was described by Wilson (1876) as consisting of Durum wheat and rye crosses to produce an amphiploid hybrid (McCloy et al., 1971). In the 1910s, naturally occurring wheat × rye hybrids were discovered in experimental wheat plots at the USDA Farm in Arlington, VA. Triticale was first developed as a grain crop that would possess the grain quality, productivity and disease resistance of wheat, and the vigor and hardiness of rye (Briggle, 1969).

Within the southeastern US, triticale was first promoted as a grain crop, and several cultivars were developed and released in the 1980s. However, due to abnormally cold winters in the 1980s, considerable winter kill caused a decrease in production and interest in triticale as a grain crop (Myer et al., 2009); additionally, initial triticale cultivars being developed for grain production did not bode well for use in cool-season annual forage systems. Recently, forage cultivars have been released recently, causing a surge in acreage of triticale planted across the country and within the Southeast. Myer et al. (2009) estimated that, in 2008, there were approximately 4,000 to 8,100 ha of triticale in the Southeast, and the authors expected the acreage to increase rapidly in the near future. For comparison, other annual cool-season forages range in acreage from 202,343 ha of oat and rye to 485,623 ha of annual ryegrass in the Southeast (Myer et al., 2009).

Triticale is an annual, cool-season bunch grass that is optimally planted in September or October and grazed from December until April or May, depending on temperature and rainfall (Myer et al., 2009). Seeding rate when planted alone should be 101 to 135 kg/ha; however, if it is used as a mixture, only 67 to 101 kg/ha of seed are required (Ball et al., 2007). Triticale will grow to approximately 0.6 to 1.2 m tall, and then it will form a spiked seed head. Ball et al. (2007) suggest that stocking rates should

be adequate to utilize forage and allow new leaf growth, and prevent maturation of vegetation. Generally, triticale is more tolerant of heavy, wet soils than rye, and is more cold tolerant than wheat (Ball et al., 2007).

When used for forage, triticale produces DM yields and has nutritive quality similar to those of other cool-season small grains such as rye, wheat and oat. Brown and Almodares (1976) reported that triticale planted in October in northern Georgia produced a seasonal total of 5,983 kg DM/ha from 4 harvests between December 7 and April 10, which was similar to that of rye (7,440 kg DM/ha) and oat (6,723 kg DM/ha) planted and harvested at the same time as the triticale. The authors reported that the triticale contained 29.63% CP at the first cutting, and the final cutting in April contained only 11.9% CP; similar values were observed for rye and oat. Glass and van Santen (2008) reported that 3 different triticale varieties had a seasonal DM yield of 5,822 to 6,101 kg/ha over 3 yr when fertilized with a total of 179 kg N/ha in a split application. Garcia del Moral et al. (1995) reported very low average yield values ranging from 295 to 1,759 kg DM/ha, but CP concentration averaged 22.1% and ranged from 18.5 to 26.1% CP.

Triticale and other small grains have high DM yield and high CP concentration, suggesting efficient N utilization. Gibson et al. (2007) reported that winter triticale grown in Iowa had the least DM yield, N concentration and N uptake values when 0 kg N/ha was applied; however, DM yield, N concentration and N uptake increased with increasing N fertilization rates and were greatest at 99 kg N/ha, the highest level tested. Foliar N concentration declined as the season progressed, while forage DM increased at all fertilization rates (0, 33, 66 and 99 kg N/ha), resulting in N uptake throughout the season ranging from 45 (0 kg N/ha fertilization rate) to 105 kg N/ha (99 kg N/ha

fertilization rate). Schwarte et al. (2005) reported similar results for winter triticale planted in Iowa and fertilized at a rate of 56 kg N/ha. Recently, there has been interest in use of triticale for removal of residual soil nitrate to prevent leaching of nitrate. Nance et al. (2007) reported a residual available NO_3-N of 48 to 80 kg at a soil depth of 120 cm, depending upon the previously planted crop. When additional N fertilizer was used to stimulate triticale growth, the authors reported up to 41.6 kg N/ha was captured from the residual soil NO_3-N, reducing nitrate leachate by up to 40% compared with no cover crop at all (Nance et al., 2007).

Winter forages also provide an opportunity to mitigate pollution potential of animal manures that can contain relatively high concentrations of P. Brown and Almordares (1976) conducted a study to determine the capacity of fall-planted triticale to remove P from Idaho soils enriched with P due to excessive dairy manure application. When 112 kg N/ha fertilizer was utilized, triticale produced an average 6.61 Mg DM/ha with a P concentration of 3.26 g/kg, which equates to removal of 21.9 kg P/ha from the soil. This rate of removal was greater than that from other winter annuals, including winter wheat that removed only 18.0 kg P/ha during the growing season. The authors also reported that soil-test P was reduced within the top 30 cm of soil from 28.1 mg P/kg in 1999 to 11.8 mg P/kg in 2001, resulting in a P removal of 18.3 kg P/ha.

Crimson Clover. Crimson clover (*Trifolium incarnatum*) is an annual cool-season legume that is indigenous to Eurasia, mainly the British Isles, Spain, France, Italy, the Balkan Peninsula and Turkey, where it has been grown as a winter annual forage (Frame, 2005). It was introduced into the US in the early 1800s and has been grown extensively in the Southeast and Pacific-coastal zones for cool-season grazing. Crimson clover is

also grown in higher latitudes as a warm-season annual, but in the Southeast is has been mainly used as a green manure cover crop (Frame, 2005).

Crimson clover grows to approximately 0.3 to 1.0 m in height and produces a brilliant crimson flower on each stem (Ball et al., 2007). While it is an annual species, it does re-seed itself quite well, and a productive multi-year stand can be established with proper management. It is seeded in late August to October, typically via aerial broadcasting at a rate of 22 to 34 kg/ha. While fairly tolerant of soil acidity, crimson clover is not especially tolerant of calcareous or poorly drained soils. For grazing, forage production occurs during November and, due to cold weather, does not become productive again until March through April in the northern part of Alabama, but in the southern counties it is productive through November and December, as well as in February through April (Ball et al., 2007). Crimson clover is also useful in grass/legume mixtures and has been successfully cultivated in Italian ryegrass (*Lolium multiflorum*), bahiagrass (*Paspalum notatum*) and bermudagrass (*Cynodon dactylon*) stands (Frame, 2005).

Crimson clover has long been used as both forage, singly and in mixtures, as well as a green manure largely because of its ability to fix atmospheric N_2 into NO_3, which is readily available to plants. Holderbaum et al. (1990) reported that a ryegrass/crimson clover cover crop that was managed with forage harvesting and return of nutrients to simulate cattle grazing produced the greatest DM yield of corn compared with no cover, cover with no harvest, or hay removal of cover crop (15.2, 13.4, 12.9 and 12.8 Mg /ha, respectively) when fertilized with 90 kg N/ha. The authors also reported that N uptake was greater (180 kg N/ha) when the ryegrass/crimson clover cover crop was managed for

nutrient removal compared with the other treatments. Karpenstein-Machan and Stuelpnagel (2000) conducted a study to determine how legume cover crops affected a subsequent maize crop. The authors employed 8 treatments: rye and crimson clover at 25%, 50% and 75% clover, a pure crimson clover stand, rye and winter pea at 25% pea, 50% pea and 75% pea, and a pure winter pea stand. Soil NO_3-N concentrations (0 to 90 cm depth) were measured in July, August and October, and stands of both the crimson clover and winter pea had the greatest soil NO_3-N concentrations in July at 75 kg/ha each when no N fertilizer was used on the subsequent maize crop. When a N fertilizer rate of 225 kg N/ha was used, soil NO_3-N concentrations were still greatest (300 kg NO_3-N/ha) in the pure legume stands compared with the other treatments. In August and October, the same trends were observed among treatments and fertilizer regimes; however, soil NO_3-N was decreased to 50 kg/ha in August and 43 kg/ha in October under pure crimson clover. However, the authors did not report any differences in total maize DM yield among treatments within each fertilizer regime, even though soil NO_3-N concentrations were different.

Legumes have the ability to fix atmospheric N, which allows them to achieve high biomass production with little to no exogenous N input. Frame (2005) reported that crimson clover interseeded with Italian ryegrass produced a total forage yield of 5.3 to 6.4 $t \cdot ha^{-1} \cdot yr^{-1}$; however, crimson clover represented only approximately 20% of the sward biomass. Knight (1970) reported that crimson clover grown in a 'Coastal' bermudagrass sod had a 4-yr mean yield of 2.9 $t \cdot ha^{-1} \cdot yr^{-1}$. Similarly, Evers and Newman (2008) reported that greatest DM yield of crimson clover was 11,000 kg/ha, which occurred in late April. Number of cuttings (used to simulate defoliation by grazing) had no effect on

total-season yield of the crimson clover, which the authors attributed to the superior capacity of crimson clover to withstand periodic defoliation compared with other clover species. Because of greater foliar N concentrations, concentration of CP in clover is normally greater than that of grass species. Dunavin (1982) reported that 'Dixie' crimson clover grown in a bahiagrass sod and fertilized with 139 kg N/ha, 26 kg P/ha and 67 kg K/ha during the summer season produced forage with a CP value of 25.4%. Lloveras and Iglesias (2001) reported whole-plant CP concentrations of 12.4 to 10.7%; however, leaf concentrations were as high as 27.9% CP. These values decreased with increasing maturity of the forage.

Even though they are able to fix atmospheric N, legumes will utilize soil N if there is an adequate amount that is readily plant-available. Pederson et al. (2002) reported that swards of crimson clover had a greater whole-plant N content (17.6 g N/m^2) than ryegrass (10.0 g N/m^2); however, the values for crimson clover do not represent only N uptake from soil, but also that from N fixation. Edmeades et al. (1986) investigated foliar N concentration of crimson clover as affected by both N fixation and soil N uptake. The authors reported that N uptake by crimson clover was 95 kg N/ha, and that symbiotic N fixation was 170 kg N/ha. The authors also noted that, when seeded with perennial ryegrass, the contribution of soil N uptake by clover was 18%.

Phosphorus is an essential element for plant growth, and legume uptake of P is typically greater than in grasses per kg of DM production due to the large complement of P-enriched N-fixing enzymes. However, because grasses typically produce substantially greater DM yield than legumes, P uptake is greater for grasses than legumes (Caradus, 1980). Brink et al. (2001) reported that crimson clover fertilized with 9 Mg/ha broiler

litter (34, 20 and 32 g/kg N, P and K, respectively) in a sandy loam soil with low soil-test P had a foliar concentration of 2.9 g P/kg DM. The forage produced 9,720 kg/ha, which equates to 28 kg P/ha uptake. However, due to greater DM yield of ryegrass than crimson clover, this value was not significantly different from the annual P uptake of ryegrass. McKell et al. (1962) compared P utilization by eight legumes including crimson clover. The authors reported that crimson clover had the greatest P uptake among all legume species tested at 15.5°C (22 kg P/ha), and at 21°C had the next greatest P uptake (20 kg P/ha). However, P uptake was significantly decreased to 9 kg P/ha at 10°C, which the authors attributed to decreased aerial plant and root growth at colder temperatures.

Other studies (Jackson and McDermid, 1964) have reported that application of 0, 60 or 1,350 kg of $P_2O_5 \cdot ha^{-1} \cdot yr^{-1}$ did not affect foliar P concentrations of 'Dixie' crimson first-cut clover (0.27 – 0.30% P). This lack of response to rate of P_2O_5 application was also observed in other clover species tested (0.25, 0.26, 0.30 and 0.24 for 'Tallarook' subterranean clover, 'Ladino' white clover, 'New Zealand' white clover and 'Kenland' red clover, respectively). Adams et al. (1966) reported that crimson clover grown at various N, P and K fertilization rates with bermudagrass had a linear increase in foliar P concentration with increasing P fertilization rates (0.29, 0.32, 0.38 and 0.40% P at 0, 25, 48 and 98 kg P/ha, respectively). However, the authors did not report a difference in foliar P concentrations among different N fertilization treatments (0, 112, 224 and 448 kg N/ha). Additionally, increasing rates of K fertilization (0, 46, 93 and 185 kg K/ha) tended to increase foliar P concentrations (0.34, 0.33, 0.34 and 0.39% P, respectively); however, this trend was not as strong as with P fertilization. The authors also reported that P uptake by crimson clover was lowest with the lowest P fertilization rate, 7.1 kg

P/ha, and increased linearly with the highest P fertilization rate producing the highest P uptake, 16.8 kg P/ha.

Phosphorus has been shown to affect nodulation and, therefore, N fixation by legumes. Ae et al. (1990) reported that legume growth is typically P-limited; not only does P limit growth, but Smith (1992) reported that nodulation of roots was reduced by inadequate P levels. Therefore, not only does low P concentration limit the quantity of N incorporation due to biomass restriction, but it also can reduce the amount of *Rhizobia* found in association with a legume species. Also, the N-fixing enzyme nitrogenase is limited by how much available inorganic N is already available in soil, as nitrogenase activity decreases at elevated N levels. Therefore, the supply of N and P in terrestrial ecosystems can regulate rates of symbiotic N-fixation, both altering the success of legumes and by influencing the activity of nitrogenase (Smith, 1992). Lynd et al. (1984) reported that the addition of P solution (100 mg P/kg soil) to soil of greenhouse-grown arrowleaf clover (*Trifolium vesiculosum* Sav., an annual cool-season clover) more than doubled nitrogenase activity (21.6 and 48.2 μmol $C_2H_4 \cdot g^{-1}$nodule$\cdot h^{-1}$ without and with P, respectively). The authors also reported that inclusion of P resulted in nearly 100% increase in aboveground plant P content (0.86 and 1.74 g/plant for without and with P, respectively); similar trends were also observed in nodule weight. Additionally, P concentration of root nodules was significantly different between P treatments, 47.8 and 116.2 μg P/g nodule, without and with P, respectively.

Bermudagrass. Bermudagrass (*Cynodon dactylon*) is a warm-season perennial that is well suited for grazing and hay production (Hancock et al., 2012). Common bermudagrass was introduced into the US from southeastern Africa in the early 1750s by

Governor Henry Ellis of Georgia. Through natural dispersion, it then spread across the Southern states and covered most of the Southeast by the late 19th century. It was used as both a pasture and hay forage; however, many producers considered it a noxious weed, a reputation it retains in some regions today (Hanna et al., 2011a). Bermudagrass cultivation and breeding began in 1928 at the University of Georgia Coastal Plain Experiment Station in Tifton, GA. The same group of breeders later released the still popular cultivars 'Coastal', 'Tifton 44' and 'Tifton 85' (Hanna et al., 2011b). The latter variety, 'Tifton 85', is noted for its increased DM yield, long grazing season and increased ADG of grazing cattle (Hanna et al., 2011c). Today, over 6 million ha of the Southeast are planted in various varieties of bermudagrass (Hanna et al., 2011a).

Bermudagrass is a sod-forming grass that spreads by rhizomes, stolons and seeds (in some cultivars), and grows to approximately 38 to 61 cm (Ball et al., 2007). It is best adapted to sandy soils and is extremely drought tolerant, making it a popular forage in both the Coastal Plain and Piedmont regions of the Southeast. Common bermudagrass can be planted using seed at a seeding rate of 5.6 to 11.2 kg/ha in spring; however, some hybrid varieties require planting of sprigs in March or April at 0.9 m^3/ha in rows or 2.2 to 3.5 m^3/ha broadcast. Bermudagrass is highly responsive to N fertilization; however, requirement for K is high compared with other warm-season forages, and K limitation is the most common reason for stand declines. Bermudagrass is productive from late May through September in Alabama; in warmer climates it can be productive from early May through October (Ball et al., 2007).

Bermudagrass is noted for both its high yield potential and medium nutritive quality. Overman et al. (1992) reported that 'Coastal' bermudagrass interseeded with

crimson clover had a maximum annual DM yield of 19.28 Mg·ha^{-1}·yr^{-1}, which was greater than 'Coastal' bermudagrass alone that produced up to 18.90 Mg DM·ha^{-1}·yr^{-1}; these values were derived from yield responses to 5 different N fertilization rates that were analyzed using a statistical model for predicting maximum forage yield potential. For both stands, the maximum forage production was achieved at approximately 500 kg N·ha^{-1}·yr^{-1}. These values were also significantly greater than those for common bermudagrass with and without clover modeled in the same manner (12.12 Mg/ha and 11.92 Mg/ha, respectively). Franzluebbers et al. (2004) reported 'Coastal' bermudagrass yields as affected by inorganic (NH_4NO_3, 225 kg N·ha^{-1}·yr^{-1}), clover + inorganic (135 kg N·ha^{-1}·yr^{-1}) and broiler litter (194 kg N·ha^{-1}·yr^{-1}) fertilization regimes. Stands had an initial DM mass of 3.28, 2.70 and 2.71 Mg/ha for inorganic, clover + inorganic and broiler litter, respectively. After a 140-d growing season, stand total production was 6.15 Mg/ha (inorganic fertilizer), 5.97 (clover + inorganic) and 5.71 Mg/ha (broiler litter); however, these values were not different. Similarly, Silveria et al. (2007) reported 'Coastal' fertilized with 135 kg N·ha^{-1}yr^{-1}of urea in a single application produced 14.3 Mg DM/ha total forage when harvested for hay every 4 to 5 wk during the growing season. Hill et al. (1993) reported that percentage CP in whole-masticate samples from steers grazing 'Tifton 85'averaged 14% over the season, but varied during the season from 11.9 (May sampling) to 15.6% (September sampling). Due to selective grazing, these values represent the best quality forage, and mean percentage CP in the stand was most likely less (Hill et al., 1993). Hoveland et al. (1978) reported that 'Coastal' sod supported 187 grazing d/ha for beef cow-calf pairs, a cow ADG of 0.22 kg and a calf ADG of 0.71 kg. The authors also reported that the maximum cow-calf stocking rate was

2.5 pairs/ha, which occurred from June through October. Mandebvu et al. (1999) reported a season average 16.2% CP in 'Tifton 85' fertilized with 112 kg $N \cdot ha^{-1} \cdot yr^{-1}$. The greatest percentage CP was recorded during the second wk of the experiment (20.8%), and it declined linearly until it reached 11.1% in Wk 7. The authors also reported overall seasonal values of 69.2% NDF, 30.7% ADF and 63.2% IVDMD.

Bermudagrass has significant capacity for biomass production and therefore has been successfully used to phytoextract nutrients that have accumulated in soil, including N and P. Silveria et al. (2007) reported that 'Coastal' bermudagrass fertilized with 45 kg N/ha as NH_4NO_3 had a N uptake of 202 kg N/ha. When fertilized with 90 kg N/ha, the N uptake rate increased to 288 kg N/ha; and when fertilized with 135 kg N/ha, the N uptake rate increased to 310 kg N/ha. The authors calculated relative N uptake efficiency [N efficiency (%) = $\frac{N_{uptake}\,(fertilized\ plot) - N_{uptake}\,(control\ plot)}{N\ rate\ applied} \times 100$] as 67%, averaged across 3 yr of data. Production efficiency (kg DM/kg N) calculated as $\frac{\text{yield fertilized plot - yield unfertilized control plot}}{\text{N application rate} \times \text{number of applications}}$, was reported as 55.7 kg DM/kg N at the 45 kg N/ha fertilization rate. When plots were fertilized with 90 kg N/ha, production efficiency was 35.3 kg DM/kg N and, when fertilized with 135 kg N/ha, production efficiency was 28.8 kg DM/kg N. However, even though production efficiency decreased with increasing N fertilization rate, overall N uptake still increased due to increased biomass production. McLaughlin et al. (2004) investigated the effect of swine effluent on N uptake by 'Coastal' and common bermudagrass when applied at a rate to deliver 371 kg $N \cdot ha^{-1} \cdot yr^{-1}$. Over three yr, common bermudagrass had an average N uptake of 153.4 kg $N \cdot ha^{-1} \cdot yr^{-1}$, and 'Coastal' bermudagrass had an average N uptake of

148.5 kg N·ha^{-1}·yr^{-1}. These values were greater than those of all other warm-season perennials tested (eastern gamagrass, indiangrass, johnsongrass and switchgrass).

Bermudagrass has also been extensively studied for its capacity to extract P from high-P soils. Recent studies (Read, 2012) reported P uptake by bermudagrass and ryegrass grown in rotation and fertilized with broiler litter and inorganic N fertilizer. After 4 yr of broiler litter application at 9 Mg·ha^{-1}yr^{-1} (246 ±18 kg N/ha and 116 ± 11 kg P/ha), surface soil-test P (0 to 15 cm) was 103.143 mg P/kg, which is considered to be high. After 2 yr of broiler litter application (9 Mg/ha) supplemented with 112 kg N·ha^{-1}·yr^{-1} inorganic N fertilizer, surface soil test P was 53.08 mg P/kg. However, this experiment was conducted as a small field-plot experiment, and actual soil P values at greater spatial scales would most likely decline at a slower rate due to uneven distribution of both forages and broiler litter application. Coblentz et al. (2004) reported that P uptake by common bermudagrass during the first of 3 seasonal harvests responded linearly to N fertilization rate and was greatest (8.5 kg P/ha) with the highest N fertilization rate (168 kg N/ha). However, foliar P concentration did not differ among the 4 fertilization regimes, averaging 0.32% P. The same trend was observed for the second harvest of the season, with the greatest P uptake also being 8.5 kg P/ha; foliar P concentrations were greater (0.33%), but this difference was not biologically significant. The greatest foliar P concentrations were observed during the last harvest (0.41% P), which also resulted in greater P uptake during the third harvest (up to 14.5 kg P/ha), for which no differences were observed among fertilization rates. The authors reported that maximum P uptake in common bermudagrass occurred at approximately the 280 kg N·ha^{-1}·yr^{-1} fertilization rate; when the N fertilization rate was increased above this value, P uptake declined (30.5 and

28.5 kg P/ha, respectively). Similar results were observed when the same plots were used during a second season; P uptake was maximized at 224 kg $N \cdot ha^{-1}yr^{-1}$ (50.8 kg P/ha) and did not change with increasing N fertilization rates of 280 and 336 kg $N \cdot ha^{-1}yr^{-1}$ (49.9 and 50.0 kg P/ha, respectively). Read et al. (2007) reported that after a "build-up" phase using broiler litter to increase soil P levels, total surface soil P was 562 mg/kg at 0 to 5 cm depth. In 2 subsequent yr in which 'Coastal' bermudagrass was fertilized with 268 kg $N \cdot ha^{-1} \cdot yr^{-1}$ with inorganic fertilizer, total soil P was reduced to 389 mg/kg using bermudagrass alone and to 367 mg/kg using ryegrass in rotation with bermudagrass. The subsurface soil (5 to 15 cm) had a maximum soil total P of 212 mg/kg initially; however, after two years of bermudagrass management, the levels were reduced to 157 mg/kg using ryegrass-bermudagrass rotation.

Cowpea. Cowpea (*Vigna unguiculata*) is an annual, warm-season legume that is commonly called southern pea, blackeye pea, crowder pea, lubia neibe, coupe or frijole. Cowpea originated in Africa and is still widely grown in Africa, Latin America, southeast Asia and the southern US (Davis et al., 1991). Cultivation of cowpea dates back 5 to 6 thousand yr ago in Ethiopia and, while it is commonly used as a grain crop for human consumption, it is also utilized as forage for livestock (Ball et al., 2007).

Cowpea is a rapidly growing summer legume that produces a viney, weak steam with three large leaflets and single white flowers (Mullen, 1999; Ball et al., 2007). Maximum canopy height is 50 to 60 cm, depending on the variety; however, cowpea is not tolerant of overgrazing, and defoliation below 15 cm can lead to plant death (Davis et al., 1991). Cowpea is useful both as pasture forage and hay due to its good forage quality, and it is adapted to a variety of soils ranging from light, sandy soils to well-drained,

loamy soils. Cowpea may be drilled into a prepared seed-bed or no-till drilled at a rate of 33 to 44 kg/ha; however, if broadcast aerially, the seeding rate must be increased to 112 to 134 kg/ha. Cowpea should be planted when soil temperatures are above 18° C, from approximately May to early June in the southern region of the US (Ball et al., 2007). It is tolerant of drought and heat, making it a useful summer forage crop in the Southeast, and it is productive from June until September, or until the first frost (Mullen, 1999; Ball et al., 2007; Davis et al., 1991). Because it is a legume, cowpea requires no N fertilization; however, to increase nodulation success and rates, cowpea seeds should be inoculated with the appropriate *Rhizobium* species (Mullen, 1999).

When managed properly, cowpea can be very productive and produce significant DM yield. Boe et al. (1991) reported that cowpea grown at two different sites in South Dakota produced 5,800 to 8,000 kg DM/ha. Muir (2002) reported that cowpea grown with or without dairy manure and harvested every 6 weeks during the growing season produced 3,194 kg DM·ha^{-1}·yr^{-1} during the first yr; however, forage DM yield in the second yr was decreased to 511 kg DM·ha^{-1}·yr^{-1} due to unusually low precipitation during the summer, and dairy manure did not affect forage yield. The author also reported that forage harvested only once in the fall produced 807 and 300 kg DM·ha^{-1}·yr^{-1} during yr 1 and yr 2, respectively, which suggests that cowpea is not especially well adapted to stockpiling, but can produce large quantities of forage DM when harvested or grazed regularly. Muir (2002) also reported that cowpea grown with or without dairy cattle manure contained 13.5 to 22.3% CP, and that there was no difference between fertilizer treatments or harvest treatments for CP concentration. Use of dairy manure or timing of harvest did not affect ADF or ADL concentrations in cowpea; ADF values ranged from

17.8 to 21.5% and ADL from 39.2 to 40.4%, in agreement with Thro and Shock (1987) who reported that whole-planted harvested cowpea contained 22.0% ADF.

Holzknecht et al. (2000) reported that highly utilized cow pea (defined as removal of all leaves and some plant stems) supported ADG of 1.17 kg/d in Charolias × Brahaman cattle grown in Queensland, Australia. Cowpea leaves contained 22.4% CP and stems contained only 7.8% CP, and IVDMD was 64% in leaves and 58% in stems. The authors also reported a herbage mass of 4,890 kg DM/ha with a residual mass of 3,473 kg DM/ha after weekly grazing rotations, and that the optimal stocking rate of 2.8 hd/ha was slightly greater than the low-utilization treatment of 2.4 hd/ha.

As with most forage legumes, N fixation and residual N are major factors that should be considered in selecting the best plant species for a given production system. Stamford et al. (2003) reported that cowpea grown in tropical soils of Brazil had the highest nodulation rate (a presumptive measure of N fixation potential) with application of 600 kg S/ha, but the highest shoot total N values were found in inoculated plants fertilized with the lowest level of S (600 kg S/ha) and/or with gypsum; these values ranged from 160 to 190 mg N/plant. Sanginga et al. (2000) studied the effect of P fertilization rate on different breeding lines of cowpea and reported that nodulation rate on P-responding varieties increased with increasing P application rate (0 to 60 kg P/ha applied). The percentage N derived from the atmosphere averaged 70% and was affected by P application in the P-responder group. The greatest P-responder total shoot N was 28.9 kg N/ha, averaged across all P application rates; total N_2 fixed ranged from 13.1 (0 kg P/ha applied) to 31.9 kg/ha (60 kg P/ha applied). The non-P-responder group had a total shoot N of 25.35 kg N/ha, and total N_2 fixed ranged from 17.2 to 30.4 kg/ha.

Phosphorus is often the most limiting nutrient for legumes species, and they are therefore relatively efficient at using residual soil P for growth. Cassman et al. (1981) reported that when cowpea is grown without any N fertilizer inputs, the maximum DM yield of approximately 6,000 kg DM/ha was achieved at a P fertilizer application rate of 220 to 230 kg P/ha. These values were generated by growing cowpea at various P fertilization rates and creating a model to ascertain where P use efficiency was greatest. Muir (2002) reported that cowpea fertilized with and without dairy cattle manure and harvested in autumn only or periodically throughout the growing season had greater foliar P concentration when 20 Mg dairy manure/yr were applied than 0 Mg dairy manure/yr (2.56 and 2.22 g P/kg DM, respectively). When averaged across manure treatments, foliar P concentrations were not different between the different harvesting regimes, ranging from 1.60 to 2.75 g P/kg DM. The use of dairy manure over 2 yr increased plant-available soil P, 40 mg/kg soil without manure and 19.1 mg/kg soil with manure; however, neither forage type nor harvest regime produced specific patterns of change in soil P. Sanginga et al. (2000) reported P use efficiency of 0.16 to 0.30 g shoot DM/ mg P in shoots in P-responding cowpea varieties, and was greatest at the lowest P application rate (0 kg P/ha). Efficiency of P uptake ranged from 55 to 227 mg P in shoot/g dry roots; the greatest P-uptake efficiency was reported at the median P application rate of 40 kg P/ha compared with 0, 20 and 60 kg P/ha application rates. Fernandez and Ascencio (1994) reported that acid phosphatase activity (a plant enzyme that catalyzes conversion of organic P into plant-available inorganic P) was not affected by plant age (2 or 4 wk) or P concentration of nutrient solution (1 mM P and 0.02 mM P); however, root tissues had greater acid phosphatase activity than either mature leaves or young leaves.

Environmentally important plant nutrients

Phosphorus. Phosphorus is an essential element for all life forms; it is a component of bones, teeth, phospholipids, nucleotide phosphates and many other compounds (Correll, 1998). Phosphorus is an important plant macronutrient that constitutes approximately 0.2% of plant DM and 0.68% of animal weight (Schachtman et al., 1998; Ball et al., 2007). After N, P is considered the most commonly limiting plant macronutrient; however, much of soil P is in the organic form and is not readily plant-available (Schachtman et al., 1999).

The P cycle in the biosphere includes both terrestrial and aquatic systems; however, it does not typically include an atmospheric component with the exception of transport via volcanic activity and/or dust/aerosol particles. Atmospheric flux rates are slow compared with those in surface waters and soils (Correll, 1998). Significant sources of P include synthetic fertilizer, animal excreta and weathering of P-rich rock. However, P in rock is present in poorly soluble forms (i.e., calcium phosphate) and not readily available to plants (Smil, 2000). Fertilizer forms of P include inorganic phosphate (PO_4^{-3}) that is readily available to plants, and organic forms that must first be converted to phosphate before being utilized by plants (Filippelli, 2002).

Adequate amounts of P increase the response of plants to applications of both N and K; adequate P also promotes growth of young tissues including roots, flowers and seeds (Smil, 2000). Globally, all major field crops including forages grown on arable land assimilate approximately 12 Mt P annually; however, weathering and atmospheric deposition supply no more than 4 Mt P/yr (Smil, 2000). Plant uptake of P is limited by

the amount of inorganic P (P_i) in soil solution, which rarely exceeds 10μM (Schachtman et al., 1998). However, plants have developed specialized transporters at the root-soil interface for extraction of P_i from soil solution and transporting it across membranes between intracellular compartments against a concentration gradient. Therefore, under normal physiological conditions, energy is required for transport of P_i across the plasma membrane (Schachtman et al., 1998).

Regulation of P_i uptake into plants is controlled to maintain a cytoplasmic P_i maintenance concentration of approximately 5 to 10 mM. This is independent of the external P_i concentration, except under severe P depletion. In contrast, vacuolar P_i concentrations vary widely and are largely dependent on external concentrations of P_i. If P_i is limited, plants will grow more roots to increase the amount of uptake from the soil, translocate P_i from older leaves and deplete vacuolar stores of P_i, and the roots may become more extensively colonized by mycorrhizal fungi (Schachtman et al., 1998). However, if plants have adequate amounts of P_i and are absorbing it at rates that exceed need, P_i is converted into organic storage forms, uptake rate is reduced, and P_i is removed via efflux in order to prevent toxic levels of P_i from accumulating in plant tissues (Schachtman et al., 1998).

In more than 90% of terrestrial plant species, P uptake is not solely by root cells but also by symbiotically root-associated mycorrhizal fungi. In these plants, fungal hyphae play a vital role in uptake of P by the plant; plants transfer C from plant tissues in exchange for P and other mineral nutrients from the fungus. Smith and Reed (1997) reported that influx of P in roots colonized by mycorrhizal fungi can be 3 to 5 times greater than in non-mycorrhizal roots. There are several factors that may contribute to

increased rate of P_i uptake, including an extensive network of hyphae extending from the roots that increases the surface area for acquisition of P from organic sources that are not directly available to plants (Schachtman et al., 1998).

Phosphorus in plants is transferred to animal tissues as a result of digestive processing by herbivores, notably ruminants. Phosphorus is the second most plentiful mineral in the body of cattle (Ternouth, 1990). Morse et al. (1992) reported that efficiency of absorption of P from mixed diets fed to lactating dairy cows was only approximately 50%. Dietary intake of P is affected by both feed P concentration and feed intake and can range from 2 to 24 g P/d for a 300-kg steer (Ternouth, 1990). According to Playne (1976), approximately 50% of P present in forages is P_i, and up to 30% of the remainder is phyate-P; however, microorganisms located in the rumen allow for the breakdown of esterified P including phyate-P. Tamminga (1996) reported that true availability of P in forages is 65 to 100%. Additionally, 30% of the P outflow from the rumen is endogenous microbial P from rumen microorganisms (Playne, 1976). Phosphorus secretion in saliva is the most important source of endogenous P and accounts of 30 to 60 g of P/d, which is one to six times the daily P intake (Tamminga, 1996).

Small quantities of P can be absorbed throughout the digestive tract of the ruminant animal. The duodenum and jejunum are the main sites of P absorption. Phosphorus absorption occurs through both active transport and passive diffusion, and the amount of absorbed P equals that secreted in the saliva and that ingested from the diet, dependent on the solubility of the P and the P status of the animal (Ternouth, 1990). Excretion of P through urine is normally of little significance in the ruminant, and daily

loss is less than 1 g/d for mature cattle (Ternouth, 1990). The kidneys act to retain certain nutrients within the blood, including P that is almost completely reabsorbed from the kidney fluids (Ternouth, 1990). However, studies with sheep have shown that, when fed high levels of P, sheep appear to have higher urinary P excretion (Scott et al., 1984).

The majority of P excretion in cattle occurs through fecal material and, of the total P ingested by domestic farm animals, about 70% is excreted (Barnett, 1994a). Tamminga (1992) reported that a typical dairy cow excretes 13.7 kg P/yr in feces; additionally, Tamminga (1996) states that this P is largely organic P, mainly nucleic acids and phospholipids. Phosphorus excretion is determined largely by P intake, and amounts and chemical forms of P excreted vary considerably (Morse et al., 1992). Other studies (Barnett, 1994b) report that cattle excreted 4.32 to 7.27 mg P/g fecal DM. Toor et al. (2005) reported that P concentrations in feces were related to P forms in the diet in all but 1 of 6 dairy farms. Also, as diet P quantity increased, the amount of P_i excreted increased as both a quantity but also as a percentage of total P, 51% of total P excreted at the lowest P intake and 70% at the greatest P intake. Barnett (1994a) reported that 47.1% of total P excreted was P_i, 37.0% was residual P (nucleic acid-type material), 13.9% was contained in acid-soluble organic P (phytic acid) and 2.1% as phospholipids. However, Barnett is quick to state that there are few references for these values, and most research is over 50 yr old.

Phosphorus is returned to the soil via crop residue decomposition and deposition of feces from grazing animals. Soil P is tightly bound in highly weathered, acid soils that contain high concentrations of Fe and Al, leaving it virtually unavailable to plants (Bellows, 2001). However, Fe-P and Al-P complexes give these soils a large capacity to

store P from manure and residue inputs (Friesen et al., 1997). Eghball et al. (1996) reported that movement of P in soils from land- applied fertilizer was only 4 cm in 94 d, and that most P movement occurred within the first few wk after application of P fertilizer. However, Sharpley et al. (1984a) reported that application of cattle feedlot manure resulted in increased soil P test to a 0.3 m soil depth. Eghball et al. (1996) also reported that maximum soil P adsorption occurred at a lower depth in manure-amended soil than when inorganic-P fertilizer was applied (0.7 and 0.4 m, respectively); additionally, adsorption was greater in manure-amended soil than soil to which inorganic fertilizer had been applied (205 and 175 mg/kg, respectively). Vadas et al. (2007) reported an increase of 121 mg P/kg soil after bare soil (0 to 2 cm depth) was amended with poultry litter at a rate of 13 Mg/ha; however, after 50 d the soil P concentration was reduced to within 31 mg P/kg of the initial soil P value. Deeper soil (2 to 5 cm) P only increased by 21 mg P/kg soil with application of poultry litter, and little reduction in soil-test P was observed after 50 d. Crop residues also offer opportunities to recycle nutrients, including P in pasture systems. Franzleubbers et al. (2002) reported that total soil P in an unharvested (biomass cut but left in field) bermudagrass pasture fertilized with broiler litter was 537 mg/kg at a soil depth of 0 to 6 cm and 5 yr of management. However, when hayed (less crop residue available), total soil P was 460 mg/kg, and when light grazing intensity was used, soil P was 575 mg P/kg.

Most P returned to soil from organic sources is organic P and unavailable for plant use; however, through bacterial transformations, organic P can become mineralized and available for plant growth in limited amounts. Despite the widespread deficiency of plant-available P, most soils contain large quantities of total P (Richardson, 2001). The

rhizosphere supports large populations of soil microorganisms and contains a wide range of plant and microbial exudates and metabolites (including the phosphatase enzyme); therefore, unavailable soil P is released by a variety of mechanisms within this zone (Richardson, 2001). Compared with bulk soil, the rhizosphere has increased phosphatase activity, due in part to depletion of P stores and increased populations of soil microorganisms. Organic acids excreted by plant roots create a reduced environment within the rhizosphere that results in increased solubility of various forms of precipitated P (e.g., Ca-phosphates). Other root processes (e.g., uptake of NH_4^+ ions) also result in net acidification of the rhizosphere, thereby also increasing P solubility (Richardson, 2001). Recent evidence also indicates that organic acids increase the accessibility of soil organic-P substrates to enzyme hydrolysis (Hayes et al., 2000).

Microorganisms directly affect P solubilization, mineralization and immobilization. Various forms of precipitated P are solubilized by soil bacteria and fungi; predominantly *Bacillus*, *Pseudomonas*, *Penicillium* and *Aspergillus* spp. (Richardson, 2001). Laboratory screening studies indicate that up to 40% of the culturable soil microorganism population is able to solubilize P; however, it is important to note that laboratory screening methods for identifying and quantifying soil microbiota may misrepresent actual microorganism populations (Kucey, 1983). Richardson (2001) suggests that solubilization of P by microorganisms is a major mechanism for plant growth promotion. Soil microorganisms also play an important role in mineralization of soil organic P and represent a significant component of total soil phosphatase activity. Richardson (2001) stated that organic P is rapidly degraded when added to soil and that microorganisms obtain P from various sources of organic P, evidence of which is the

rapid turnover rate of organic matter in soil. However, quantification of amounts of P mineralization and its relative contribution to plant P nutrition remains poorly documented (Richardson, 2001). Studies have shown that phytates in soil are resistant to mineralization and accumulate as a result of adsorption and precipitation. Numbers of studies of microorganisms that possess phytase activity and studies conducted on the ability to inoculate plants with these microorganisms have increased recently; however, in these studies the response to inoculation are limited (Richardson, 2001).

Plants and soil microorganisms produce phosphatases that convert organic P into inorganic P. Phosphatases are a broad group of enzymes that catalyze hydrolysis of both esters and anhydrides of phosphoric acid. These include phosphoric monoester hydrolases, enzymes acting on phosphoryl-containing anhydrides and enzymes acting on P-N bonds (phosphoamidases) (Eivazi and Tabatabai, 1977). Acid phosphatase (phosphomonoesterase) is responsible for the conversion of orthophosphoric monoester into phosphate, and it is also the most extensively studied of phosphatase enzymes produced by both plants and soil microorganisms. (Eivazi and Tabatabai, 1976). Alkaline soil phosphatase also converts orthophosphoric monoester into phosphate; however, its optimum pH is higher (7 to 9) than that of acid phosphatase (approximately 4 to 6), and it is produced by soil microorganisms but not plants (Eivazi and Tabatabai, 1977).

Due to the nature of these enzymes, variability of soil conditions affects enzyme activity. Dick et al. (2000) conducted a study to determine the effect of pH on both acid and alkaline soil phosphatase activity. Five different soils were incubated with 100% of the lime requirement for the specific soil, and soils were analyzed for 67 d following lime

addition. Soil 1 had an original pH of 3.3, and after 67 d the pH was 7.1. The alkaline phosphatase to acid phosphatase activity ratio increased from 1.1 to 1.8 between d 0 and d 67. A similar trend was observed in soils 2, 3 and 4 (original soil pH of 4.7, 5.0 and 5.4, respectively); however, the pH of soil 5 (initially 6.6) decreased from 1.0 to 0.6 over the 67-d period. Hýsek and Šarapatka (1998) reported that the number of alkaline phosphatase-active colonies in the soil was positively correlated with organic C and the number of ammonification bacteria in the H horizon. However, the A horizon was almost biologically inactive except for some acid phosphatase activity that was positively correlated with organic matter content. More importantly, the authors did not report any correlation between number of acid or alkaline phosphatase-active bacteria colonies and acid and alkaline phosphatase activity, which the authors attributed to production of phosphatases by organisms other than bacteria. Additionally, the authors found that the presence of plants positively affects phosphatase activities.

Olander and Vtousek (2000) reported that potential phosphatase activity in the O and A horizons ranged from 7.8 to 22.7 $\mu mol \cdot g \cdot^{-1} h^{-1}$, with greater activity in the O horizon. The authors also reported that P fertilization decreased phosphatase activity, from 10.30 to 3.82 $\mu mol \cdot g \cdot^{-1} h^{-1}$ in the O horizon and from 22.71 to 11.38 $\mu mol \cdot g \cdot^{-1} h^{-1}$ in the A horizon. However, the addition of both N and P fertilizer did not affect phosphatase activity in either horizon. Rojo et al. (1990) reported phosphatase activity in a temperate pasture with calciferous alkaline soil was 4.4 $\mu mol\ pNP \cdot g^{-1} soil \cdot h^{-1}$, and was 11.6 $\mu mol\ pNP \cdot g^{-1} soil \cdot h^{-1}$ in an acidic soil.

The microbial biomass in soil contains a significant amount of P that generally accounts for 1 to 10% of total soil P, and may represent as much as 100 kg P/ha

(Richardson, 2001). Microbial P is a dynamic portion of the soil-P pool and is responsive to soil-P status, season-specific factors and management practices. Brookes et al. (1984) reported that soil biomass P ranged from 3.6 to 4.7% of total soil P at a depth of 0 to 15 cm in a permanent grassland. Additionally, soil biomass P was 13.7 ± 3.5% of total soil organic P. Incubation studies in bulk soil have shown that microorganisms are important for maintaining levels of both inorganic and organic P; however, Richardson (2001) stated that whether microorganisms act as a net source of P within the rhizosphere or as a temporary sink for plant-available P is not fully understood.

A high percentage of soil P is adsorbed to soil particles; thus, little to no P is leached from soils. However, through soil erosion, aquatic systems can become overloaded with P, causing eutrophication of water bodies. According to the USEPA (1996), eutrophication is the main cause of impaired surface water quality in the US, and eutrophication of most water bodies is accelerated by P inputs. Lake water concentrations above 0.02 ppm P generally accelerate eutrophication; however, the soil solution P concentration necessary for plant growth is 0.2 to 0.3 ppm (Sharpley, 2003).

Surface and subsurface runoff from agricultural fields is often the source of P that enters water bodies; additionally, P in runoff can exist as either dissolved or in the predominant form which is sediment-bound (Sharpley, 2003). Sediment-bound P includes both P associated with soil particles and organic material eroded during flow events, and constitutes about 80% of P transported in surface runoff from cultivated land. Due to the filtering capacity of pasture and grasslands, surface runoff from non-cultivated land usually contains little sediment-bound P; however, dissolved P can still be transported from pasture runoff. For eutrophication remediation purposes, it is important

to note that most dissolved P is readily plant available, whereas sediment-bound P must be transformed by chemical and/or enzymatic reactions prior to being plant-available (Sharpley, 2003). Eghball and Gilley (1999) reported that sorghum fields fertilized with feedlot cattle manure at a rate of 49.4 Mg/ha had 7.7 mg/L total P, 6.9 mg/L particulate P and 0.73 mg/L dissolved P in runoff. In the same study, commercial fertilizer applied at a rate of 151 kg N/ha and 25.5 kg P/ha produced 13.8 mg/L total P, 8.6 mg/L particulate P and 5.20 mg/L dissolved P in runoff. The authors reported similar results when the field was planted with wheat and fertilized with feedlot manure; total P was 5.4 mg/L, particulate P was 4.0 mg/L and dissolved P was 1.30 mg/L in runoff.

Nitrogen. Nitrogen is an essential nutrient for all living organisms, and makes up essential cellular components. Unlike the P cycle that includes only terrestrial and aquatic forms, the N cycle includes terrestrial, aquatic and atmospheric forms; these include NH_4, NO_3, N_2O, N_2 and organic N. Nitrogen makes up approximately 70% of the earth's atmosphere; inorganic N (NH_x, NO_x) makes up 70% of this total and organic N the rest (Schlesinger, 2008). Throughout its cycle, N is transformed by various microorganisms, as well as by climatic events such as wind and temperature.

During the 20th century, the development of synthetic N fixation gave producers a faster and cheaper method of N fertilization application via ammonium nitrate and other commercial N fertilizers. Prior to the "Green Revolution," the only ecologically-derived N inputs were from organic matter decomposition and biological N-fixation that transforms $N_{2\,(g)} \rightarrow NH_{4\,(soil)}$. Nitrogen fixation is accomplished by bacteria, either free-living (e.g., cyanobacteria) or living in a symbiotic relationship (e.g., *Rhizobia* or *Frankia*) with plant species, typically legumes. These bacterial species are obligate

anaerobes, facultative anaerobes or obligate aerobes (Reed et al., 2011), and reside in nodules along the roots of the plants.

The nitrogenase enzyme is responsible for the fixation of N_2 into NH_4. There are 4 known nitrogenases, and each one requires Fe and typically Mo or V for full functionality. The most abundant nitrogenase is the Mo-dependent enzyme. The chemical equation for the reaction catalyzed by the Mo-dependent nitrogenase is represented by:

$$N_2 + 8H^+ + 16\ Mg{\cdot}ATP + 8e^- \rightarrow 2NH_3 + H_2 + 16\ Mg{\cdot}ADP + 16\ P_i.$$

(Seefeldt et al., 2009)

Nitrogen fixation is one of the most costly, in terms of ATP, metabolic processes on Earth; *Azotobacter vinelandii* was found to use > 16 mol of ATP per mol of N_2 fixed, and > 100 g of glucose to fix one gram of N_2 (Gutschick 1981; Hill 1992). Nitrogenase contains two components, the Fe protein (component II) and the Mo·Fe protein (component I); the Fe protein is responsible for delivering the electrons one by one to the Mo·Fe protein, which is responsible for the actual conversion of N_2 into NH_4 (Seefeldt et al., 2009).

Non-leguminous plants take up N from the soil solution but are limited to only NO_3 and NH_4; legumes in association with N-fixing *Rhizobia* transfer NH_4 from the root nodules directly into the plant tissues in exchange for carbohydrates from the plant. Additionally, legumes can transfer up to 40% of their fixed N to surrounding grasses during the growing season (Bellows, 2001); a pasture containing 20 to 45% legumes can meet the N requirement for the rest of the grass species in the pasture for the entire

growing season (Thomas, 1992). This occurs via the development of mycorhizzal associations between grass and legume roots in the soil matrix (Brophy et al., 1987).

The actual uptake of N occurs across the cell membrane of plant roots. Plants transfer one NO_3 via a symport with two H^+ ions, and NH_4 is transferred via a uniport (Engles and Marschner, 1995). Nitrate-N has been shown to saturate the transport system at 0.2 to 0.5 mM concentration; additionally, plants exhibit a preference for NH_4, which is represented by evidence showing that the presence of NH_4 inhibits plant cell uptake of NO_3 (Engles and Marschner, 1995).

Ruminants consume N through ingestion of plant materials containing largely organic N. Typically, digestible proteins are located within the cytoplasm of plant cells, and relatively indigestible proteins are found in within cell walls of plant tissues (Ball et al., 2007). However, due to the unique characteristics of the ruminant digestive system, ruminants can exist without a source of dietary protein (Owens and Zinn, 1988). When protein is ingested, rumen microorganisms that have proteolytic activity utilize this protein for amino acid and microbial protein synthesis (Ball et al., 2007). Rumen microbes then pass to the abomasum and small intestine, and the host animal is able to gain necessary protein from the microbes themselves. This source of protein is very concentrated, with rumen microbes typically being 20 to 60% CP by DM weight; bacteria alone have approximately 50% CP by DM weight (Owens and Zinn, 1988). Once the microbial protein reaches the intestines, it is absorbed and utilized within the small intestines in the same method as monogastric animals.

In addition to protein digestion, ammonia can be absorbed across the rumen wall and into the blood stream. Ammonia in the rumen is from recycled N in the saliva, or

from nonprotein N compounds (e.g., urea) that are converted in the rumen to ammonia and absorbed across the wall (Van Soest, 1994). Ammonia in the blood is quickly converted into urea by the liver, making it nontoxic to the animal. This method of recycling N allows ruminants to survive for extended periods of time on limited N intake. Amino acids that are absorbed in the small intestine from microbial digestion are transported to animal tissues or the liver. In the liver, these amino acids are converted to urea and enter the blood urea pool. The animal then uses this pool to produce saliva, which allows rumen microorganisms to survive if there is a N shortage, or any overflow is filtered through the kidneys and into the urine (Van Soest, 1994).

Fecal loss of N is only approximately 0.6% of DM intake; this amount is much less variable than urinary loss and is equivalent to a dietary protein intake of 3 to 4% (Van Soest, 1994). Nitrogen in feces is from metabolic losses and includes microbial debris and endogenous substances (e.g., Ca and Mg salts, sloughed-off animal cells and mucus); microbial debris typically includes cell walls that are highly indigestible and include nitrogenous compounds. Van Soest (1994) also states that in normal feces there should be no evidence of potentially digestible feed protein.

In the absence of N_2-fixation, the predominant N inputs are from the degradation of organic N from crop residues and animal excretions, mainly urea but also a small amount of fecal N. In the soil, organic N compounds are converted into inorganic forms (NH_4 and NO_3) via mineralization. Mineralization of organic N occurs through two separate but linked processes, ammonification and nitrification (Stevenson and Cole, 1999). Ammonification is an enzymatic process in which N is liberated as NH_4 from organic nitrogenous compounds (e.g., proteins and peptides). Enzymes responsible for

ammonification include proteinases, peptidases, amino acid dehydrogenases, amino sugar kinases, ureases and nucleases, to name a few (Ladd and Jackson, 1982). The process of nitrification then converts NH_4 into NO_3, which is also plant-available. Nitrification is a highly spontaneous process that rapidly converts NH_4 to NO_3 in a two-step process; the first conversion ($NH_4 \rightarrow NO_2$) is achieved by *Nitrosomonas*, and the second ($NO_2 \rightarrow NO_3$) occurs via *Nitrobacter*. Both *Nitrosomonas* and *Nitrobacter* are Gram-negative, chemautotrophic, archebacteria (Stevenson and Cole, 1999). Both NH_4 and NO_3 make up the pool of inorganic plant-available N in the soil. This pool is in equilibrium with immobile, organic N. Immobilization is simply the inclusion of soil inorganic N in soil microorganisms, which renders it inaccessible. However, as soil microorganisms die and decay, this N is returned to the inorganic N pool (Stevenson and Cole, 1999).

While the majority of N returns to the soil, a portion of plant/animal N is returned to the atmosphere via NH_3 volatilization. Stevenson and Cole (1999) estimated that 26 to 53×10^9 kg N/year return to the atmosphere via volatilization. In a typical agricultural setting, N losses are predominantly via commercial fertilizer or manure application, and can range from 3% to greater than 50% depending on the type of fertilizer and prevailing climatic conditions (Stevenson and Cole, 1999). In cattle urine, the volatilized ammonia is derived mainly from the urea component that is generally 65 to 90% of total N (Lockyer and Whitehead, 1990). Lockyer and Whitehead (1990) reported that the proportion of N volatilized from cattle urine applied to a grass sward ranged from 3.7 to 26.9% of the total N. The experiment was conducted during the cooler months, but showed that the majority of volatilization occurs within the first 4 d after application. Vallis et al. (1982) reported that volatilization rates in a subtropical

pasture were highest 24 h after application and had relatively low rates of volatilization within 3 to 4 d after application. Additionally, they reported that average volatilization rates were higher in February than June, possibly due to differences in soil moisture during those months. Vallis et al. (1982) also reported that annual losses of NH_3 were calculated to be only 17 to 34 kg $N \cdot ha^{-1} \cdot yr^{-1}$.

The last process of the N cycle is dentrification; it is the major process for which N is reduced and returned to the atmosphere as N_2 and to a less extent N_xO (Firestone, 1982). Firestone (1982) estimated that N loss in agricultural soils via dentrification are as high as 70% of applied fertilizer N. Dentrification can only occur when soils are waterlogged or contain anaerobic microsites within them; both *Azospirillum* and *Thiobacillus*, among others, are capable of dentrification. The rate of dentrification is enhanced with poor soil drainage, increased soil temperature, neutral soil pH and an abundant supply of readily degraded organic matter (Stevenson and Cole, 1999).

II. EFFECTS OF NITROGEN FERTILIZATION ON SOIL NUTRIENT CONCENTRATION AND PHOSPHATASE ACTIVITY, FORAGE NUTRIENT UPTAKE, AND NUTRIENT LOAD IN RUNOFF FROM A YEAR-ROUND PASTURE SYSTEM

INTRODUCTION

Animal manures have been used for millennia as a source of nutrients for crops of all types. With the advent of modern animal agriculture, manure-derived nutrients may become concentrated in locales that cannot fully utilize them. Consequently, land can become enriched in P due to repeated application over many years. Phosphorus does not leach from soil and can accumulate over time, possibly creating environmentally harmful situations such as eutrophication (Eghball et al, 1996).

In the past, production of high-yielding forage crops such as tall fescue (*Lolium arundinacea*) and bermudagrass (*Cynodon dactylon*) for hay was regarded as the easiest and quickest method by which to phytoextract P from P-enriched soils (Whalen and Chang, 2001). This approach requires a commitment from the producer to keep affected areas in hay production for many years. In the southeastern US where land is typically the most limiting resource for livestock production, many producers are unable to make this commitment. Ball et al. (2007) have stated that N is the single nutrient that normally produces the most dramatic growth response in forage grasses; therefore, N fertilization can increase the amount of P that can be phytoextracted from the soil by simply increasing forage DM production. Grazing cattle year-round offers a viable method for generating animal product while mitigating P accumulation in the soil (Sharpley et al., 1994).

The objective of this research was to determine the impact of different N-application regimes on P transformation and movement through a P-enriched, grazed-pasture system. We hypothesized that manipulation of N application regime, in conjunction with overseeding of a N-fixing legume into a permanent pasture sod, would affect patterns of plant uptake and soil concentrations of P, as well as P loss via water runoff such that phytoextraction and efficiency of P cycling are enhanced, pollution potential is reduced, and a more sustainable P equilibrium is re-established.

MATERIALS AND METHODS

Research Site

The experimental site contained six instrumented runoff plots (Figure 1) constructed in 2007 at the Stanley P. Wilson Beef Teaching Center of the Auburn University Department of Animal Sciences, Auburn, AL (32° 53' 34.43" N latitude, 85° 30' 3.32" W longitude, 187 m above MSL). Plots (91.4 × 30.5 m, 0.28 h each) ranged from 1 to 10% slope, and consisted of a permanent common bermudagrass (*Cynodon dactylon*) and tall fescue (*Lolium arundinaceum*) sod on a Marvyn loamy sand (Fine-loamy, Kadinitic, thermic Typic Kanhapludult) and a Pacolet sandy loam (Fine, Kaolinitic, thermic Typic Kanhapludult) soil. Each plot was bordered with flexible plastic conveyor belt material to define drainage boundaries and confine overland and subsurface flow within the plot. For measurement and sampling of runoff from each plot, a Model 3700 portable ISCO automatic runoff sampler (Teledyne Isco, Inc., Lincoln, NE), and a 30-cm H-flume (Plasti-Fab, Inc., Tualatin, Oregon) were installed at the lower end of each plot with a V-shape approach zone. Two tipping-bucket rain gauges and a National Weather Service Class A evaporation pan were also installed at the lower end of the pasture.

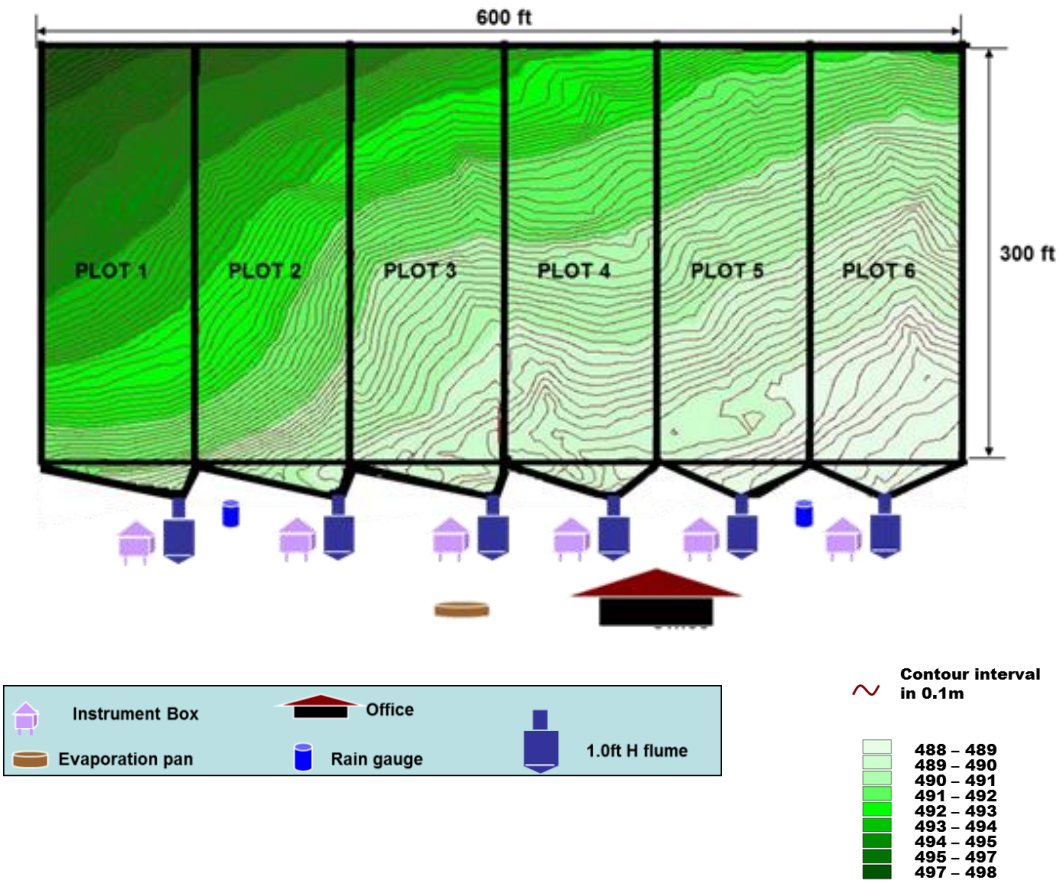

Figure 1. Experimental watershed site

Forage Establishment and Fertilization

In October, 2009, plots were no-till drilled (Great Plains model 3P606NT with small seed box) with triticale (× *Triticosecale rimpaui* Wittm var. 'Trical 2700') at a seeding rate of 125.5 kg/ha and pre-inoculated crimson clover (*Trifolium incarnatum* var. 'Dixie') at a seeding rate of 33.6 kg/ha, and randomly assigned to 1of 3 treatments: 0% of N (0N) recommendation for triticale, 50% of N (50N) recommendation for triticale (50.4 kg N/ha) in a single application at planting, and 100% of N (100N) recommendation for triticale (100.8 kg N/ha) in a split-application; the first at planting, and the second directly prior to turning out cattle into plots. On October 15, the 50N and 100N treatments received 50.4 kg N/ha in the form of ammonium sulfate-urea. On February 18, 2010, 100N plots received an additional 50.4 kg of N/ha. On May 23, 2010, plots were mob-grazed with 12 mature cows (mean BW, 635 kg) in order to achieve a forage stubble height of 10 cm; all cattle were allowed to graze each plot for 72 h, and were then moved to the next plot until all plots were grazed.

On June 16, 2010, plots were no-till drilled with cowpea (*Vigna unguculata* var. 'Iron and Clay') at a seeding rate of 57 kg/ha. Cowpea seeds were inoculated prior to planting with EL Type inoculant at a rate of 170 g inoculant per 23 kg seed. Plot assignments to fertilization treatments were the same as in the previous winter-spring (cool) growing season (CS); for the summer (warm) growing season (WS), N fertilization rates were based on recommendations for bermudagrass. The 0N treatment thus received 0 kg N, the 50 N treatment received 56.0 kg N/ha at time of planting, and the 100N treatment received 112.0 kg N/ha in a split-application at planting and prior to turning out cattle into plots on July 14, 2010.

The experiment was repeated without re-randomization of N-fertilization treatments among plots for 2 additional yr (2011 and 2012). On October 19, 2010, plots were seeded with triticale and crimson clover using the same seeding rates as in 2009. The 50N and 100N treatments received 50.4 kg N/ha as ammonium sulfate-urea at the time of planting, and the 100N treatment received an additional 50.4 kg N/ha on February 16, 2011. On November 1, 2011, plots were no-till drilled with triticale and crimson clover using the same seeding rates as in the 2 previous yr. The 50N and 100N treatments received 50.4 kg N/ha as ammonium sulfate-urea at the time of planting, and the 100N treatment received an additional 50.4 kg on January 12, 2012. In May 2011 and 2012, after completion of CS grazing, plots were mowed to a 10-cm height using a mechanical mower.

The WS portion of the experiment was also repeated for 2 additional yr, with plot assignments to treatments remaining the same as in 2010. On July 5, 2011 plots were no-till drilled with inoculated cowpea, and the 50N and 100N treatments were fertilized with 56.0 kg N/ha. On August 2, 2011, the 100N treatment received an additional 50.6 kg N/ha. On June 14, 2012, plots were seeded with cowpea, and 50N and 100N treatments were fertilized with 56.0 kg N/ha. On August 1, 2012, the 100N treatment received an additional 56.0 kg N/ha.

Cattle Grazing Management

On April 7, 2010, 6 Angus heifers and 6 Angus steers (216 ± 56 kg BW) were randomly assigned to graze in the plots (2 cattle/plot) with the proviso that 1 steer and 1 heifer were assigned to each plot. Cattle were turned out when forage availability had

achieved 1,000 kg DM/ha. Cattle were allowed free access to water, salt and shade for the duration of the grazing season. Once forage availability had decreased below 500 kg DM/ha or forage had matured to seed head stage (May 18, 2010), the grazing season was terminated and cattle were removed from the plots. The WS grazing period commenced on July 20, 2010 with 12 Angus heifers (279 ± 33 kg BW) assigned randomly to plots (2 heifers/plot). As in the CS grazing period, cattle were given free access to water, salt and shade. Once forage DM availability had fallen below 500 kg DM/ha (on August 31), cattle were removed from the plots.

For the CS grazing period in 2011, 6 Red Angus × Beefmaster cattle (4 steers and 2 heifers, 328 ± 60 kg BW) were randomly assigned to plots (1 animal/plot) and turned in to graze on February 23. Cattle were allowed to graze until May 5, 2011 when forage availability had decreased below 500 kg DM/ha. On August 9, 2011, 6 Red Angus × Beefmaster steers (361 ± 23 kg) were randomly assigned to plots and allowed to graze. Grazing continued until September 21, 2011 when forage DM availability had decreased below the 500 kg DM/ha threshold.

Four Angus steers and two Angus heifers (345 ± 60 kg BW) were randomly assigned to plots (1 animal/plot) and turned out to graze on January 18, 2012. Cattle were allowed to graze until May 10, 2012 when the grazing season was terminated because forage DM availability had decreased below 500 kg DM/ha. During the WS grazing period in 2012, six Angus steers (312 ± 31 kg BW) were randomly assigned to plots (1 steer/plot) and allowed to graze until September 26, 2012 when forage DM availability had decreased below 500 kg DM/ha.

Soil Core Sampling

Prior to planting on October 10, 2009, 6 random 1-m soil cores were taken in each plot using a 1-m hydraulic, soil-probe (Giddings Machine Company, Windsor, CO). Soil cores were separated into depth strata of 0 to 10 cm, 10 to 20 cm, 20 to 40 cm, 40 to 60 cm, 60 to 80 cm and 80 to 100 cm. All 6 samples of the same depth from each plot were mixed thoroughly, and a subsample was taken and dried at 60°C for 72 h, sieved to pass a 2-mm screen, and stored for laboratory analysis. An additional set of 1-m soil cores were taken after cattle removal from plots 3 yr later on October 12, 2012. The same procedure was followed to collect, dry, sieve, and store samples for laboratory analysis.

Concentrations of soil total N and C were determined via dry combustion using a LECO TruSpec CN Analyzer (LECO Corp, St Joseph, MI). Soil samples were extracted using dilute nitric/hydrochloric acid solution (Mehlich I) and analyzed by inductively coupled argon plasma (ICAP) spectroscopy (SPECTRO CIROS CCD, Germany) to determine concentrations of P, K, Ca, Mg, Zn and Cu (Hue and Evans, 1986).

Forage Sampling

Three forage samples were taken randomly from each plot prior to cattle turn out to estimate forage DM availability and nutrient concentration. Samples were randomly taken using a 0.25-m^2 quadrat and hand clippers to cut forage at a 2-cm height. After cattle had been turned out for grazing, 3 forage samples were taken randomly from each plot biweekly until cattle were removed from plots. Prior to each season, three 2-m^2 exclusion cages were randomly placed within each of the 6 pastures to prevent cattle grazing. After cattle turn out, 1 sample was taken from each of the 3 exclusion cages every 2 wk. Samples of previously unharvested forage were collected so that seasonal

accumulations of DM and nutrients could be determined for ungrazed, primary-growth forage. Forages were dried at 60° C for 72 h after collection. Once dry and air-equilibrated, forages samples were weighed and separated by species into tall fescue, triticale, crimson clover or 'other' (any other plant species) in the CS; and bermudagrass, cowpea or 'other' in the WS. Within each species, sub-samples were mixed thoroughly for uniformity, and then ground to pass a 1-mm screen in a Wiley Mill (Thomas Scientific, Philadelphia, PA).

Concentrations of forage total N and C were determined via dry combustion using a LECO TruSpec CN Analyzer (LECO Corp, St Joseph, MI). Concentrations of P, K, Ca, Mg, Zn and Cu in forage samples were analyzed by dry-ashing followed by ICAP spectroscopy (Hue and Evans, 1986).

Soil Phosphatase Sampling for Analysis

Prior to cattle turnout in the CS grazing periods of 2011 and 2012, 3 15-cm soil samples were taken randomly from each plot. An additional 3 soil samples were taken from each plot after termination of the WS grazing periods in 2011 and 2012. Samples from the same plot were mixed, and a subsample was taken and air-dried for 24 h. After air drying, soil samples were sieved to pass a 2-mm screen. A 0.2-g sample from each plot was analyzed for acid and alkaline phosphomonoesterase activity using the Tabatabai (1982) procedure.

Runoff Water Collection and Sampling

The ISCO sampler in each plot enabled automated recording of flow rate, total rainfall and ambient air temperature data that were electronically recorded by a CR5000

datalogger (Campbell Scientific, Inc., Logan, Utah). Sample collection commenced on October 1, 2009 and was continued until September 26, 2012. The sampler was programmed to collect a 10-ml sample every 2 min during a runoff event, and was triggered for collection by the liquid-level actuator that detected the presence of runoff in the flume; measurable runoff typically occurred when rainfall totaled 4 cm in a 24-h period. Water samples were kept at 4°C until they could be collected and transported to the laboratory, where they were then frozen at 0°C and stored for laboratory analysis.

All water samples were analyzed for concentrations of water-soluble P (Murphy and Riley, 1962), Total Kjeldhal N (Bremner, 1965), NO_3- and NH_4-N (Sims et al., 1995) and total P, K, Ca, MG, Zn and Cu by ICAP spectroscopy (Hue and Evans, 1986; SPECTRO CIROS, CCD).

Total nutrient mass in runoff was calculated by the following equation: Total nutrient mass = $[N_x] \times (FR_x \times D_x)$, where $[N_x]$ is the nutrient concentration, FR_x is the flow rate for a specific runoff event, and D_x is the duration of a specific runoff event.

Statistical analysis

Forage, soil core, water and soil phosphatase data were analyzed as a completely randomized design using PROC MIXED of Statistical Analysis Software (SAS, Cary, NC). The experimental unit was considered to be paddock (n = 2). For acid and alkaline soil phosphatase activity, the statistical model: included fixed effects of N-fertilization treatment, season and their interaction; year was considered a random effect. Components of the soil-core statistical model included fixed effects of N-fertilization treatment,

season, depth, year and their interactions. Forage data were analyzed using a statistical model that included main effects of N-fertilization treatment, season and their interactions, with year considered a random effect and successive samples within a single season treated as repeated measures. Water data were analyzed using a statistical model that included main effects of N-fertilization treatment and timing of samples (background or trial period). Treatment df were partitioned into single-df orthogonal contrasts that were used to compare the 0N treatment with 50N and 100N treatments (0N vs. [50N + 100N]), and compare the 50N with the 100N treatment (50N vs. 100N). In recognition of the low statistical power characteristic of field studies that employ limited numbers of replicates, treatment differences were considered significant when $P < 0.10$ (Peterman, 1990).

RESULTS

Acid and Alkaline Soil Phosphatase Activity

Acid soil phosphatase activity (μg p-nitrophenol [NP]·g^{-1}·h^{-1}) was not different (P = 0.944) between seasons (Table 1), but was greater for 0N than [50N + 100N] (P = 0.068) in CS. Alkaline phosphatase activity was greater (P = 0.071) in WS than CS, and did not differ between 0N and [50N + 100N] or between 50N and 100N (P = 0.872 and 0.251, respectively).

Table 1. Acid and alkaline soil phosphatase activity in pastures receiving different N-fertilization treatments

		N Treatment [a]				
Item	Season[b]	0N	50N	100N	Mean	SE[c]
Acid Phosphatase (μg pNP·g^{-1}·h^{-1})						
	CS[d]	30.9	8.8	8.5	16.1	
	WS	14.6	20.3	14.9	16.6	
	Mean	22.7	14.5	11.7	16.4	5.4
Alkaline Phosphatase (μg pNP·g^{-1}·h^{-1})						
	CS	9.9	12.3	7.0	8.7[e]	
	WS	15.6	20.5	14.6	17.0[f]	
	Mean	12.9	16.4	10.8	12.9	2.7

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]n = 4.
[d]0N vs. [50N + 100N] differ (P < 0.10).
[e,f]Within a column, means without a common superscript differ (P < 0.10).

Soil Characteristics

There were no differences in soil pH (Table 2) between 0N and [50N + 100N] treatments at soil depth increments of 0 to 10 cm, 10 to 20 cm, 20 to 40 cm, or 40 to 60 cm, or between the 50N and 100N treatments at the same soil depth increments ($P >$ 0.151). Soil pH in 2010 was less ($P < 0.0001$) than in 2012 over the entire 0 to 60 cm soil-depth range. However, pH was not different ($P > 0.174$) between 0 and [50N + 100N] treatments or between 50 and 100N treatments over the entire 0 to 60 cm soil-depth range.

Electrically conductivity (EC; Table 3) of the soil cores was not different between 0N and [50N + 100N] for 0 to 10, 20 to 40 or 40 to 60 cm depth increments, but was greater ($P = 0.003$) for 0N (0.019 siemens/m) than [50N + 100N] (0.007 siemens/m) at the 10 to 20 cm depth in 2012. There was no difference between the 50N and 100N treatments at any depth interval or over the entire 0 to 60 cm depth range ($P > 0.298$). However, EC in 2010 was lower ($P < 0.0001$) than in 2012 over the 0 to 60 cm depth range.

There were no differences in soil concentration of extractable P (Table 4) between 50N and 100N treatments in 0 to 10, 10 to 20, 20 to 40 or 40 to 60 cm soil depth increments, or between 0N and [50N + 100N] at the 10 to 20, 20 to 40 or 40 to 60 cm soil depths. However, 0N had a greater ($P = 0.01$) extractable P concentration than [50N + 100N] in the 0 to 10 cm depth interval. Over the 0 to 60 cm soil depth range, concentration of extractable P was not different ($P = 0.422$) between the 0N and [50N + 100N] treatments, but was less ($P < 0.023$) for 100N than 50N. Extractable P concentration in soil was greater ($P = 0.019$) in 2010 than in 2012.

Over the entire 0 to 60 cm soil-depth range, concentration of water-soluble P (Table 5) was not different ($P = 0.504$) between 0N and [50N + 100N] or between 50N and 100N treatments ($P = 0.208$). Water-soluble P was less ($P < 0.0001$) in 2010 than in 2012 over the entire 0 to 60 cm soil-depth range. In 2012 at the 0 to 10 cm depth interval, 0N had a greater ($P = 0.071$) water soluble P concentration (11.0 mg/kg) than [50N + 100N] (8.2 mg/kg). In the same year at the 10 to 20 cm depth interval, [50N + 100N] (7.2 mg/kg) had a greater ($P = 0.071$) water-soluble P concentration than 0N (4.5 mg/kg) and, 50N (8.8 mg/kg) had a greater ($P = 0.069$) water–soluble P concentration than 100N (5.5 mg/kg).

There was no difference in soil concentration of total N (Table 6) between 50N and 100N treatments at the 0 to 10, 10 to 20, 20 to 40 or 40 to 60 cm soil depth increments; however, 0N (2.4 mg/kg) was greater ($P = 0.027$) than [50N + 100N] (1.8 mg/kg) in 2012 at the 0 to 10 cm depth interval. Total N concentration in soil was not different ($P > 0.193$) between 0N and [50N + 100N] or between 50N and 100N treatments over the 0 to 60 cm soil-depth range. Also, there was no difference ($P = 0.310$) in total N concentration between 2010 and 2012 over the 0 to 60 cm soil-depth range.

Table 2. pH of 1-m soil cores in pastures receiving different N-fertilization treatments

	N Treatment[a]				
Item	0N	50N	100N	Mean	SE[b]
Depth, cm					
0 to 10	6.31	6.32	6.16	6.26	
10 to 20	6.39	6.21	6.20	6.27	
20 to 40	6.11	6.01	5.92	6.01	
40 to 60	5.95	5.87	5.96	5.93	
0 to 60	6.19	6.10	6.06	6.12	0.07
Year					
2010	5.63	5.43	5.61	5.56[c]	
2012	6.75	6.78	6.51	6.68[d]	
Mean	6.19	6.10	6.06	6.12	0.04

[a]0N = 0% of N recommendation, 50N = 50% of N recommendation, 100N = 100% of N recommendation based on N requirement of grass species.
[b]n = 4.
[c,d]Within a column, means without a common superscript differ ($P < 0.001$).

Table 3. Electrical conductivity (siemens/m) of 1-m soil cores in pastures receiving different N-fertilization treatments

	N Treatment[a]				
Item	0N	50N	100N	Mean	SE[b]
Depth, cm					
0 to 10	0.010	0.006	0.006	0.007	
10 to 20[c]	0.011	0.004	0.004	0.006	
20 to 40	0.004	0.005	0.003	0.004	
40 to 60	0.003	0.004	0.003	0.003	
0 to 60	0.007	0.005	0.004	0.005	0.001
Year					
2010	0.002	0.002	0.002	0.002[d]	
2012	0.012	0.008	0.007	0.009[e]	
Mean	0.007	0.005	0.004	0.005	0.001

[a]0N = 0% of N recommendation, 50N = 50% of N recommendation, 100N = 100% of N recommendation based on N requirement of grass species.
[b]n = 4.
[c]0N vs. [50N + 100N] differ ($P < 0.01$).
[d,e]Within a column, means without a common superscript differ ($P < 0.001$).

Table 4. Concentration of extractable P (mg/kg) in 1-m soil cores from pastures receiving different N-fertilization treatments

	N Treatment[a]				
Item	0N	50N	100N	Mean	SE[b]
Depth, cm					
0 to 10[c]	86.0	75.5	71.2	77.5	
10 to 20	71.4	72.6	60.9	68.3	
20 to 40	32.0	49.9	19.5	33.8	
40 to 60	10.7	10.8	2.9	8.1	
0 to 60[d]	50.0	52.2	38.6	47.0	4.7
Year					
2010	53.6	64.2	40.2	52.7[e]	
2012	46.4	40.4	37.0	41.3[f]	
Mean	50.0	52.2	38.6	47.0	2.7

[a]0N = 0% of N recommendation, 50N = 50% of N recommendation, 100N = 100% of N recommendation based on N requirement of grass species.
[b]n = 4.
[c]0N vs. [50N + 100N] differ ($P < 0.01$).
[d]50N vs. 100N differ ($P < 0.05$).
[e,f]Within a column, means without a common superscript differ ($P < 0.05$).

Table 5. Concentration of water-soluble P (mg/kg) in 1-m soil cores from pastures receiving different N-fertilization treatments

	N Treatment[a]				
Item	0N	50N	100N	Mean	SE[b]
Depth, cm					
0 to 10	6.2	4.5	5.1	5.2	
10 to 20	2.3	4.9	3.1	3.4	
20 to 40	1.1	3.0	1.5	1.9	
40 to 60	1.7	1.7	1.1	1.4	
0 to 60	2.8	3.5	2.7	3.0	0.5
Year					
2010	0.4	1.0	0.7	0.7[c]	
2012	5.1	6.1	4.8	5.3[d]	
Mean	2.8	3.5	2.7	3.0	0.4

[a]0N = 0% of N recommendation, 50N = 50% of N recommendation, 100N = 100% of N recommendation based on N requirement of grass species.
[b]n = 4.
[c,d]Within a column, means without a common superscript differ ($P < 0.001$).

Table 6. Concentration of total N (mg/kg) in 1-m soil cores from pastures receiving different N-fertilization treatments

	N Treatment[a]				
Item	0N	50N	100N	Mean	SE[b]
Depth, cm					
0 to 10	1.9	1.6	1.5	1.7	
10 to 20	0.6	0.6	0.7	0.6	
20 to 40	0.4	0.3	0.4	0.4	
40 to 60	0.3	0.3	0.4	0.4	
0 to 60	0.8	0.7	0.8	0.8	0.01
Year					
2010	0.7	0.6	0.8	0.7	
2012	0.9	0.8	0.8	0.8	
Mean	0.8	0.7	0.8	0.8	0.03

[a]0N = 0% of N recommendation, 50N = 50% of N recommendation, 100N = 100% of N recommendation based on N requirement of grass species.
[b]n = 4.

Forage

Grazed crimson clover was greater (P = 0.019) proportion of CS (Table 7) than ungrazed clover and grazed bermudagrass was greater (P = 0.053) proportion of WS forage than ungrazed bermudagrass. However, tall fescue and WS 'other' were greater (P = 0.076 and 0.032, respectively) proportions of CS forage when ungrazed. Ungrazed triticale was greater proportion in 100N than 50N, but 0N and [50N + 100N] were not different. Grazed tall fescue was greater proportion of CS forage in 100N than 50N, and grazed 'other' was greater proportion of WS forage in 50N than 100N, but neither species were different between 0N and [50N + 100N].

Within the CS, crimson clover had greater (P = 0.092) DM mass (Table 8) in grazed than ungrazed forage and, bermudagrass had greater (P = 0.016) DM mass in grazed than WS forage. Ungrazed triticale had greater forage mass in 100N than 50N, but 0N and [50N + 100N] were not different. Additionally, ungrazed bermudagrass had greater mass in 0N than [50N + 100N] forage. Both triticale and crimson clover in the CS had greater forage in 0N than [50N + 100N] forage but 50N and 100N were not different. Tall fescue had greater DM mass in [50N + 100N] than 0N forage, but 50N and 100N were not different. Grazed bermudagrass had a greater DM mass in the 50N than 100N treatment.

Foliar N concentration (Table 9) was not different between grazed and ungrazed forages or among treatments for CS forages. Bermudagrass had a greater (P = 0.098) foliar N concentration in ungrazed than grazed forages, and ungrazed bermudagrass had a greater foliar N concentration in 100N than 50N, but 0N was not different from [50N + 100N].

Foliar P concentration (Table 10) of forages was not different between ungrazed and grazed forages for tall fescue, crimson clover, CS 'other' and cowpea. Both bermudagrass and WS 'other' had greater ($P < 0.070$) foliar P concentrations in ungrazed than grazed forage; however, triticale had a greater ($P = 0.029$) foliar P concentration in grazed plots than ungrazed forage. Ungrazed bermudagrass had a greater foliar P in 0N than [50N + 100N] treatments, but 50N and 100N were not different. Warm-season 'other' species had a greater foliar P in [50N + 100N] than 0N, but 50N was not different from 100N.

Grazed tall fescue had less forage P mass (Table 11) in 0N than [50N + 100N] treatments. Grazed crimson clover had greater forage P mass in 50N than 100N. Ungrazed triticale had greater forage P mass in 0N than [50N + 100N], but grazed triticale had greater forage P mass in 0N than [50N + 100N]. Ungrazed bermudagrass had greater ($P < 0.001$) P mass than grazed bermudagrass. Ungrazed bermudagrass had a greater forage P mass in 0N than [50N + 100N], but 50N and 100N were not different. However, grazed bermudagrass had a greater forage P mass in 100N forage than 50N plots, but 0N was not different from [50N + 100N].

Table 7. Proportion (%) of ungrazed and grazed forage species in pastures receiving different N-fertilization treatments

Forage Management	Season[b]	Forage Species	N Treatment[a]				Contrasts			
							0N vs. [50N + 100N]		50N vs. 100N	
			0N	50N	100N	Mean	*P* value	SE[c]	*P* value	SE[c]
Ungrazed										
	CS	Tall fescue	55.7	55.8	52.2	54.6[d]	0.623	3.5	0.374	4.0
		Triticale	10.6	8.2	18.7	12.5	0.414	3.5	0.009	4.0
		Clover	18.9	15.2	13.8	16.0[f]	0.200	3.5	0.724	4.0
		Other	14.7	20.8	15.3	16.9	0.339	3.5	0.168	4.0
	WS	Bermudagrass	81.4	81.7	85.6	82.9[h]	0.601	4.4	0.439	5.0
		Cowpea	4.0	5.6	4.0	4.5	0.856	4.4	0.748	5.0
		Other	14.6	12.7	10.4	12.6[j]	0.481	4.4	0.727	5.0
Grazed										
	CS	Tall fescue	41.0	53.8	56.1	50.3[e]	< 0.001	3.8	0.598	4.4
		Triticale	18.0	9.3	10.0	12.4	0.422	3.8	0.257	4.4
		Clover	26.7	18.9	19.4	21.6[g]	0.046	3.8	0.901	4.4
		Other	13.0	18.5	13.5	15.0	0.027	3.8	0.881	4.4
	WS	Bermudagrass	91.8	92.1	83.0	89.0[i]	0.610	5.0	0.115	5.8
		Cowpea	3.5	2.7	9.4	5.2	0.617	5.0	0.248	5.8
		Other	4.7	5.2	7.6	5.8[k]	0.727	5.0	0.675	5.8

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]n = 6.
[d,e]Within a column, means without a common superscript differ ($P < 0.10$).
[f,g]Withina column, means without a common superscript differ ($P < 0.05$).
[h,i]Within a column, means without a common superscript differ ($P < 0.10$).
[j,k]Within a column, means without a common superscript differ ($P < 0.05$).

Table 8. Forage mass (kg DM/ha) of ungrazed and grazed forage species in pastures receiving different N-fertilization treatments

Forage Management	Season[b]	Forage Species	N Treatment[a]				Contrasts			
							0N vs. [50N + 100N]		50N vs. 100N	
			0N	50N	100N	Mean	*P* value	SE[c]	*P* value	SE[c]
Ungrazed										
	CS	Tall fescue	1,118	1,120	1,302	1,180	0.485	133	0.237	154
		Triticale	223	168	511	301	0.383	133	0.026	154
		Clover	445	370	402	406[d]	0.661	133	0.836	154
		Other	207	324	311	281	0.409	133	0.935	154
	WS	Bermudagrass	2,439	2,149	2,081	2,223[f]	0.054	168	0.722	194
		Cowpea	114	171	102	129	0.897	168	0.722	194
		Other	398	307	235	313	0.450	168	0.709	194
Grazed										
	CS	Tall fescue	1,018	1,271	1,509	1,266	0.011	145	0.156	168
		Triticale	577	210	264	350	0.020	145	0.749	168
		Clover	826	485	619	643[e]	0.059	145	0.426	168
		Other	215	383	305	301	0.373	145	0.642	168
	WS	Bermudagrass	2,634	2,527	2,154	2,439[g]	0.130	194	0.095	224
		Cowpea	108	61	175	115	0.960	194	0.610	224
		Other	153	151	232	179	0.841	194	0.718	224

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]n = 6.
[d,e]Within a column, means without a common superscript differ ($P < 0.05$).
[f,g]Within a column, means without a common superscript differ ($P < 0.10$).

Table 9. Foliar N concentration (%) of ungrazed and grazed forage species in pastures receiving different N-fertilization treatments

Forage Management	Season[b]	Forage Species	N Treatment[a]				Contrasts: 0N vs. [50N + 100N]		Contrasts: 50N vs. 100N	
			0N	50N	100N	Mean	*P* value	SE[c]	*P* value	SE[c]
Ungrazed										
	CS	Tall fescue	2.57	2.64	2.85	2.69	0.399	0.21	0.389	0.25
		Triticale	2.46	2.68	2.55	2.56	0.509	0.23	0.641	0.28
		Clover	3.22	3.31	3.40	3.31	0.605	0.24	0.736	0.27
		Other	2.63	3.03	2.63	2.73	0.181	0.23	0.109	0.25
	WS	Bermudagrass	2.61	2.28	3.01	2.63[d]	0.894	0.26	0.017	0.30
		Cowpea	3.00	3.03	3.17	3.07	0.788	0.36	0.709	0.37
		Other	2.22	2.59	2.29	2.64	0.438	0.29	0.369	0.32
Grazed										
	CS	Tall fescue	2.47	2.57	2.89	2.64	0.249	0.23	0.207	0.26
		Triticale	2.94	2.46	2.57	2.66	0.114	0.27	0.722	0.31
		Clover	3.05	3.18	3.32	3.18	0.417	0.25	0.657	0.31
		Other	2.69	2.49	2.63	2.60	0.638	0.27	0.618	0.29
	WS	Bermudagrass	2.07	2.19	2.70	2.32[e]	0.208	0.30	0.151	0.35
		Cowpea	2.46	3.00	2.93	2.80	0.325	0.52	0.882	0.27
		Other	1.94	2.20	2.32	2.16	0.448	0.42	0.792	0.47

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]n = 6.
[d,e]Within a column, means without a common superscript differ ($P < 0.10$).

Table 10. Foliar P concentration (%) of ungrazed and grazed forage species in pastures receiving different N-fertilization treatments

							Contrasts			
Forage		Forage	N Treatment[a]				0N vs. [50N + 100N]		50N vs. 100N	
Management	Season[b]	Species	0N	50N	100N	Mean	*P* value	SE[c]	*P* value	SE[c]
Ungrazed										
	CS	Tall fescue	0.29	0.29	0.29	0.29	0.862	0.02	0.875	0.02
		Triticale	0.31	0.31	0.30	0.31[d]	0.629	0.02	0.615	0.02
		Clover	0.28	0.30	0.29	0.29	0.478	0.02	0.736	0.02
		Other	0.29	0.28	0.30	0.29	0.976	0.02	0.288	0.02
	WS	Bermudagrass	0.28	0.24	0.28	0.26[f]	0.518	0.02	0.119	0.03
		Cowpea	0.38	0.39	0.36	0.37	0.907	0.03	0.443	0.03
		Other	0.28	0.27	0.27	0.27[h]	0.673	0.03	0.850	0.03
Grazed										
	CS	Tall fescue	0.31	0.30	0.31	0.31	0.990	0.02	0.660	0.02
		Triticale	0.34	0.33	0.34	0.34[e]	0.974	0.02	0.794	0.03
		Clover	0.28	0.27	0.29	0.28	0.937	0.02	0.317	0.02
		Other	0.30	0.29	0.29	0.29	0.626	0.02	0.977	0.03
	WS	Bermudagrass	0.19	0.16	0.19	0.18[g]	0.608	0.03	0.272	0.03
		Cowpea	0.34	0.35	0.35	0.35	0.762	0.05	0.964	0.04
		Other	0.18	0.29	0.23	0.24[i]	0.021	0.04	0.165	0.04

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]n = 6.
[d,e]Within a column, means without a common superscript differ ($P < 0.05$).
[f,g]Within a column, means without a common superscript differ ($P < 0.001$).
[h,i]Within a column, means without a common superscript differ ($P < 0.10$).

Table 11. Phosphorus mass (kg/ha) of ungrazed and grazed forage species in pastures receiving different N-fertilization treatments

Forage Management	Season[b]	Forage Species	N Treatment[a]				Contrasts			
							0N vs. [50N + 100N]		50N vs. 100N	
			0N	50N	100N	Mean	*P* value	SE[c]	*P* value	SE[c]
Ungrazed										
	CS	Tall fescue	3.01	3.21	3.60	3.28	0.390	0.46	0.456	0.53
		Triticale	0.78	0.50	1.71	1.00	0.532	0.52	0.042	0.59
		Clover	1.57	1.62	1.52	1.57	0.999	0.53	0.871	0.60
		Other	0.53	0.83	1.05	0.80	0.392	0.47	0.679	0.53
	WS	Bermudagrass	6.63	4.94	5.29	5.62[d]	0.011	0.59	0.615	0.69
		Cowpea	0.42	0.71	0.40	0.58	0.922	0.81	0.715	0.84
		Other	1.43	1.01	0.81	1.08	0.408	0.64	0.786	0.73
Grazed										
	CS	Tall fescue	2.90	3.70	4.53	3.71	0.015	0.50	0.137	0.56
		Triticale	2.16	1.01	1.09	1.42	0.047	0.56	0.905	0.63
		Clover	2.55	1.34	1.85	1.92	0.062	0.51	0.390	0.59
		Other	0.20	1.10	0.88	0.73	0.544	0.56	0.724	0.62
	WS	Bermudagrass	4.68	3.15	4.72	4.18[e]	0.277	0.69	0.057	0.82
		Cowpea	0.48	0.42	0.79	0.57	0.921	1.22	0.734	1.07
		Other	0.20	0.29	0.90	0.47	0.643	0.86	0.549	1.01

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]n = 6.
[d,e]Within a column, means without a common superscript differ ($P < 0.01$).

Neither forage management nor N-fertilization treatment affected ($P > 0.135$) total DM mass (Table 12) across grazing seasons. Cool-season forages (2,364 kg DM/ha) had less ($P = 0.030$) DM mass than WS forages (2,698 kg DM/ha). Ungrazed forage had greater ($P = 0.015$) DM mass in WS than CS and, within CS forage, ungrazed forage had less ($P = 0.037$) DM mass than grazed forage.

Foliar N concentration (Table 13) was greater ($P = 0.002$) in grazed forages than ungrazed forages. Warm-season grazed forages had a greater foliar N concentration than WS ungrazed forages ($P = 0.001$); however, there was no difference between CS grazed and ungrazed forages ($P = 0.485$). Within WS, 50N (2.39%) had less ($P = 0.040$) foliar N than 100N (2.72%).

Foliar P concentration (Table 14) was not affected by N-fertilization treatment ($P > 0.260$) across forage management practices and grazing seasons. However, ungrazed forage tended to have ($P = 0.177$) greater foliar P concentrations than grazed forage, and CS (0.32%) had greater ($P = 0.100$) foliar P than WS (0.24%). Grazed CS forages had greater forage P mass (Table 15) than CS ungrazed forages ($P = 0.024$), whereas WS ungrazed forages had greater forage P mass than WS grazed forages ($P = 0.008$).

Table 12. DM mass (kg DM/ha) of ungrazed and grazed forage in pastures receiving different N-fertilization treatments

						Contrasts			
Forage		N Treatment [a]				0N vs. [50N + 100N]		50N vs. 100N	
Management	Season[b]	0N	50N	100N	Mean	*P* value	SE[c]	*P* value	SE[c]
Ungrazed									
	CS	1,993	1,982	2,526	2,167[d,f]	0.700	293	0.306	338
	WS	2,951	2,627	2,417	2,665[e]	0.531	391	0.081	311
	Mean	2,472	2,305	2,472	2,416				
Grazed									
	CS	2,636	2,349	2,696	2,560[g]	0.332	269	0.693	311
	WS	2,895	2,739	2,561	2,732	0.206	338	0.591	391
	Mean	2,766	2,544	2,629	2,646				

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]n = 6.
[d,e]Within a column, means without a common superscript differ ($P < 0.05$).
[f,g]Within a column, means without a common superscript differ ($P < 0.05$).

Table 13. Foliar N concentration (%) of ungrazed and grazed forage in pastures receiving different N-fertilization treatments

Forage Management	Season[b]	N Treatment[a] 0N	50N	100N	Mean	SE[c]
Ungrazed						
	CS	2.25	2.20	2.26	2.23	
	WS	2.04	2.10	2.41	2.20[d]	
	Mean	2.15	2.15	2.34	2.22[f]	0.07
Grazed						
	CS	2.39	2.11	2.46	2.32	
	WS	2.61	2.62	3.04	2.76[e]	
	Mean	2.50	2.37	2.75	2.54[g]	0.08

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]n = 6.
[d,e]Within a column, means without a common superscript differ ($P < 0.001$).
[f,g]Within a column, means without a common superscript differ ($P < 0.05$).

Table 14. Foliar P concentration (%) of ungrazed and grazed forage in pastures receiving different N-fertilization treatments

Forage Management	Season[b]	N Treatment[a] 0N	50N	100N	Mean	SE[c]
Ungrazed						
	CS	0.27	0.28	0.28	0.28	
	WS	0.27	0.27	0.30	0.28	
	Mean	0.27	0.28	0.29	0.28	0.03
Grazed						
	CS	0.29	0.29	0.30	0.29	
	WS	0.19	0.20	0.23	0.21	
	Mean	0.24	0.25	0.27	0.25	0.03

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]n = 6.

Table 15. Forage P mass (kg/ha) of ungrazed and grazed of forage in pastures receiving different N-fertilization treatments

Forage Management	Season[b]	N Treatment[a] 0N	50N	100N	Mean	SE[c]
Ungrazed						
	CS	5.33	5.77	7.29	6.09[d]	
	WS	7.61	6.94	6.69	7.08[f]	
	Mean	6.47	6.35	6.94	6.59	0.33
Grazed						
	CS	7.78	6.67	7.97	7.47[e]	
	WS	5.23	4.07	5.62	4.97[g]	
	Mean	6.50	5.37	6.80	6.22	0.37

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]n = 6.
[d,e]Within a column, means without a common superscript differ ($P < 0.05$).
[f,g]Within a column, means without a common superscript differ ($P < 0.01$).

Water Runoff

Water runoff pH (Table 16) prior to first N treatment application was greater ($P < 0.001$) than during the experiment. There was no difference in water runoff pH among N-fertilization treatments during the experiment. Electrical conductivity (Table 17) of water runoff was not different ($P = 0.345$) between background and study-period samples or among treatments during the experiment.

Ammonium-N (NH_4-N) concentration in water runoff (Table 18) was not different among treatments during the experiment, and there was no difference ($P = 0.390$) between background water runoff and runoff during the experiment. Nitrate-N (NO_3-N; Table 19) concentration in water runoff was also not different among study-period treatments. However, background water runoff had a greater ($P < 0.001$) concentration of NO_3-N than water sampled during the experiment. Kjeldahl-N (Table 20) concentration was not different among study-period treatments, and there was no difference ($P = 0.3891$) between background water runoff and water runoff sampled during the experiment.

Concentration of phosphate-P (PO_4-P; Table 20) was greater ($P = 0.076$) in samples collected during the experiment than in pre-trial water runoff, but there was no difference among experimental treatments. Concentration of total P (Table 21) was greater in 0N than in [50N+100N] treatments, but was not different between 50N and 100N treatments. Also, samples collected during the experimental period had greater ($P < 0.001$) total P concentration than pre-trial samples.

Table 16. pH of water runoff from grazed pastures receiving different N-fertilization treatments

	N Treatment [a]				
Item	0N	50N	100N	Mean	SE[b]
Study-period					
	6.4	6.6	6.7	6.6[c]	0.2
Background					
				7.0[d]	0.1

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]n = 6.
[c,d]Within a column, means without a common superscript differ ($P < 0.001$).

Table 17. Electrical conductivity (siemens/m) of water runoff from grazed pastures receiving different N-fertilization treatments

	N Treatment [a]				
Item	0N	50N	100N	Mean	SE[b]
Study-period					
	26.7	181.5	68.6	92.3	100.1
Background					
				45.7	75.5

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]n = 6.

Table 18. Concentration of NH_4-N (mg/L) of water runoff from grazed pastures receiving different N-fertilization treatments

	N Treatment [a]				
Item	0N	50N	100N	Mean	SE[b]
Study-period					
	0.29	0.28	0.15	0.24	0.43
Background					
				0.11	0.25

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]n = 6.

Table 19. Concentration of NO_3-N (mg/L) of water runoff from grazed pastures receiving different N-fertilization treatments

	N Treatment [a]				
Item	0N	50N	100N	Mean	SE[b]
Study-period					
	0.07	0.08	0.07	0.07[c]	0.02
Background					
				0.11[d]	0.01

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]n = 6.
[c,d]Within a column, means without a common superscript differ ($P < 0.01$).

Table 20. Concentration of Kjeldahl N (mg/L) of water runoff from grazed pastures receiving different N-fertilization treatments

	N Treatment [a]				
Item	0N	50N	100N	Mean	SE[b]
Study-period					
	1.4	0.7	0.3	0.8	1.2
Background					
				0.4	0.7

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]n = 6.

Table 21. Concentration of PO_4-P (mg/L) of water runoff from grazed pastures receiving different N-fertilization treatments

	N Treatment [a]				
Item	0N	50N	100N	Mean	SE[b]
Study-period					
	0.06	0.05	0.06	0.06[c]	0.02
Background					
				0.03[d]	0.01

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]n = 6.
[c,d]Within a column, means without a common superscript differ ($P < 0.10$).

Table 22. Concentration of total P (mg/L) of water runoff from grazed pastures receiving different N-fertilization treatments

	N Treatment [a]							
Item	0N	50N	100N	Mean	SE[b]	Contrasts	*P* value	SE[b]
Study-period								
	3.0	1.8	1.9	2.2[c]		0N vs. [50N + 100N]	0.009	0.3
						50N vs. 100N	0.774	0.3
Background								
				1.2[d]	0.2			

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]n = 6.
[c,d]Within a column, means without a common superscript differ ($P < 0.01$).

The total volume of water runoff per paddock (Table 23) was not different (P = 0.347) between pre-experimental samples and samples collected during the experiment. During the experiment, 0N treatment had greater mean runoff volume than [50N + 100N] treatments, but mean runoff volume was not different between 50N and 100N.

There was no difference (P > 0.696) in content of NH_4-N (Table 24) in runoff between pre-trial and experimental periods. However, during the study period, 0N had greater NH_4-N content per runoff event than [50N + 100N], but no differences were observed between 50N and 100N. Content of NO_3-N (Table 25) was greater (P = 0.036) in background samples than study period samples. During the experiment, 0N had a greater NO_3-N than [50N + 100N], but 50N and 100N were not different. Total Kjeldahl N content (Table 26) was not different between pre-trial and experimental runoff samples, or among N-fertilization treatments during the study period.

Total PO_4-P (Table 27) contrasts was not different (P = 0.666) between pre-trial water runoff and water runoff collected during the experiment. However, PO_4-P content during the study period was greater in 0N than [50N + 100N] treatments, but not different between 50N and 100N. Total P content (Table 28) was not different (P = 0.964) between pre-trial water runoff and water runoff collected during the experiment. However, total P content was greater in 0N than [50N + 100N], but not different between 50N and 100N.

Table 23. Volume (m^3) of water runoff from grazed pastures receiving different N-fertilization treatments

	N Treatment [a]							
Item	0N	50N	100N	Mean	SE[b]	Contrasts	*P*-value	SE[b]
Study-period								
	4.1	1.0	0.1	1.7		0N vs. [50 + 100N]	0.007	1.2
						50N vs. 100N	0.522	1.3
Background								
				2.2	0.8			

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]n = 6.

Table 24. NH_4-N (g) in water runoff from grazed pastures receiving different N-fertilization treatments

	N Treatment [a]							
Item	0N	50N	100N	Mean	SE[b]	Contrasts	*P*-value	SE[b]
Study-period								
	0.57	0.11	0.02	0.23		0N vs. [50 + 100N]	0.003	0.16
						50N vs. 100N	0.598	0.17
Background								
				0.26	0.10			

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]n = 6.

Table 25. NO_3-N (g) in water runoff from grazed pastures receiving different N-fertilization treatments

	N Treatment [a]							
Item	0N	50N	100N	Mean	SE[b]	Contrasts	*P*-value	SE[b]
Study-period								
	0.20	0.08	0.01	0.10[c]		0N vs. [50 + 100N]	0.047	0.08
						50N vs. 100N	0.374	0.09
Background								
				0.16[d]	0.03			

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]n = 6.
[c,d]Within a column, means without a common superscript differ ($P < 0.05$).

Table 26. Kjeldahl N (g) in water runoff from grazed pastures receiving different N-fertilization treatments

	N Treatment [a]				
Item	0N	50N	100N	Mean	SE[b]
Study-period					
	6.1	2.0	0.3	2.8	2.9
Background					
				2.9	1.5

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]n = 6.

Table 27. Phosphate ($P0_4$-P, g) in water runoff from grazed pastures receiving different N-fertilization treatments

	N Treatment [a]							
Item	0N	50N	100N	Mean	SE[b]	Contrasts	*P*-value	SE[b]
Study-period								
	0.17	0.03	0.01	0.07		0N vs. [50 + 100N]	0.005	0.05
						50N vs. 100N	0.627	0.05
Background								
				0.08	0.03			

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]n = 6.

Table 28. Total P (g) in water runoff from grazed pastures receiving different N-fertilization treatments

	N Treatment [a]							
Item	0N	50N	100N	Mean	SE[b]	Contrasts	*P*-value	SE[b]
Study-period								
	14.7	1.7	0.4	5.6		0N vs. [50 + 100N]	0.008	4.9
						50N vs. 100N	0.805	5.1
Background								
				5.7	3.0			

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]n = 6.

DISCUSSION

Acid and Alkaline Soil Phosphatase Activity

Acid phosphatase activity was not different between seasons or among N-fertilization treatments, in agreement with Ajwa et al. (1999) who observed that acid phosphatase activity was not different between surface soils (0 to 5 cm) in plots that had been amended with 100 kg N/ha N fertilization and a control plot. Also, the authors reported that there was no difference in acid phosphatase activity of either fertilized or unfertilized soil among April, June, August, or October sampling dates.

Alkaline phosphatase activity was not different among treatments in the current study, in agreement with Ajwa et al. (1999) who reported a sustained difference in alkaline phosphatase activity between control and N-fertilized plots. However, the authors reported a transient increase in both acid and alkaline soil phosphatase activity directly following N fertilizer application, which Olander and Vitousek (2000) suggest is mediated through direct use of N as a primary component of these N-rich enzymes, indirectly through increased productivity and P demand in response to alleviation of N deficiency, or a combination of both. Olander and Vitousek (2000) reported an increase in phosphatase activity in surface soils (0 to 10 cm) of plots receiving long-term application of 100 kg $N \cdot ha^{-1} \cdot yr^{-1}$ compared with low-N surface soils. However, the authors did not observe an effect of N fertilization on phosphatase activity at a soil depth of 10 to 18 cm. In contrast, Johnson et al. (2005) reported that addition of 120 kg $N \cdot ha^{-}$

$^{1}\cdot yr^{-1}$ did not have an effect on phosphatase activity at 0 to 5 cm and 5 to 10 cm soil depths.

Soil Characteristics

Soil core pH values were not different between 0N and [50N + 100N] or between 50 and 100N treatments. However, soil pH increased by 1.1 units from pre- to post-experiment. Franzluebbers et al. (2004) reported that bermudagrass plots fertilized with either 200 kg $N\cdot ha^{-1}\cdot yr^{-1}$ or 100 kg $N\cdot ha^{-1}\cdot yr^{-1}$ with addition of legumes had an average soil pH of 6.3 after 5 yr of low grazing pressure (forage maintained at 3.0 Mg/ha availability), which is consistent with a soil pH of 6.47 after 3 yr of application of the 100N treatment in the current study. Eghball (2002) reported that application of 151 kg $N\cdot ha^{-1}\cdot yr^{-1}$ over 4 yr decreased soil pH by 0.3 units; however, application of cattle manure on the basis of crop P requirement and supplemented with ammonium nitrate did not affect soil pH to a soil depth of 15 cm. This is critical because, for maximum nutrient availability, it is necessary to maintain a soil pH between 6 and 7 (Ball et al., 2007).

Electrical conductivity increased from pre- to post-experiment by 0.007 siemens/m, in agreement with Eghball (2002) who reported an increase in EC of 0.01 siemens/m over 4 yr in surface soil (0 to 15 cm) amended with 151 kg $N\cdot ha^{-1}\cdot yr^{-1}$. Increased EC indicates an increase in yield potential of the field as a result of increased OM, increased water holding capacity, increased cation-exchange capacity, or a combination of these. Increased EC from pre- to post-experiment in the current study could conceivably have resulted from increased soil OM due to forage decomposition and fecal deposition; however, soil OM was not measured in the current experiment. Plots

receiving P-based manure application were unchanged in EC of soil (Eghball, 2002). Kingery et al. (1994) reported EC values similar to those in the current study (0.009 and 0.01 siemens/m, respectively) in northern Alabama soils amended with various N fertilization rates.

Total P concentration averaged 81.5 mg/kg in the 0 to 10 cm soil depth, which is less than 167.1 mg/kg reported by Capece et al. (2007) for grazed-pasture soils in Florida. Also, mean soil P was less in 2012 than 2010, which differs from findings by Eghball and Power (1999) who reported that soils with repeated N fertilizer application (151 kg $N \cdot ha^{-1} \cdot yr^{-1}$) over 4 yr were unchanged in concentration of soil P (75 mg/kg). Silveira et al. (2013) reported that long-term grazing of common bermudagrass with N fertilization (279 to 461 kg $N \cdot ha^{-1} \cdot yr^{-1}$) and P fertilization (< 51 kg $P \cdot ha^{-1} \cdot yr^{-1}$) increased extractable P from 29.4 to 45.0 mg/kg in 10 yr. Phosphorus concentration decreased as soil depth increased in the present study, in agreement with Kingery et al. (1994).

Franzleubbers et al. (2002) reported that inorganic P in surface soil (0 to 6 cm) increased in plots with greater commercial N fertilization (65 and 110 kg N/ha); in contrast to the current study in which the 0N treatment had greater water-soluble P concentration than [50N + 100N] in surface soil (0 to 10 cm). Water-soluble P increased over the 2-yr study, suggesting that the presence of grazing cattle contributed to increased organic P. Sharpley et al. (1984b) reported an increase in the proportion of inorganic P in soils amended with feedlot waste, but did not observe an increase in inorganic P when plots were amended with mineral-P fertilizer. For this reason, the authors suggested that P from feedlot waste is more crop available and transferable within the soil than mineral-P fertilizer. Ternouth (1990) states that the majority of P excreted from typical ruminant

diets is endogenous P, mainly from salivary excretions, and is therefore largely inorganic P, which explains the increase in inorganic P in excreta and thus in soils amended with ruminant excreta. Stroia et al. (2011) reported that organic P values were greater in plots not fertilized with P than plots fertilized with P or with N and P, which indicates that limiting P application increased the fraction of P in the organic form by approximately 59 and 45%, respectively.

In the current study, concentration of total soil N was not different among treatments or between yr, in agreement with Burns et al. (2009) who reported no difference in total N of soil fertilized with 101, 202 or 303 kg $N \cdot ha^{-1} \cdot yr^{-1}$ planted to coastal or 'Tift 44' bermudagrass and grazed to maintain a herbage mass of 2.5 to 4.0 Mg DM/ha. In the current study, soil N was greater in the surface soil than deeper soil strata, in in agreement with Burns et al. (2009) who reported a decrease in soil total N concentration from 0 to 15 cm to 15 to 30 cm.

Forage

Lorenz and Rogler (1972) reported a decrease (34 to 17%) in the proportion of grass in a mixed prairie with increasing N fertilization (0 to 180 kg N/ha application rates). Each forage species responded differently to increasing N-fertilization rates in the present study. Both tall fescue and 'other' species represented increasing proportions of grazed CS forage at the expense of crimson clover with increasing N-fertilization rate; the proportion of triticale in ungrazed forage increased with increasing N-fertilization rate. Nitrogen fertilization typically increases the competitive edge for sod-grasses and 'other' species at the expense of legumes and drilled annual grasses. Bermudagrass and

crimson clover represented a greater proportion of WS and CS stands, respectively, in grazed than ungrazed plots, in agreement with Jones and Evans (1960) who reported grass species and clovers in a northern California grassland constituted a greater proportion of the stand when plots were grazed (41 and 4%, respectively) than when the plots were ungrazed (33 and 3%, respectively). In the same study, increasing N fertilization decreased the proportion of clover and grasses, but increased the proportion of 'other' species (forbs) within the stand, in agreement with the current study. In N-limiting systems (0N), legume species have a competitive advantage; however, in a system with at least adequate N, grass and 'other' species gain this competitive edge (Jones and Evans, 1960).

Tall fescue had greater mass of grazed forage in [50N + 100N] than 0N. Lorenz and Rogler (1972) also reported that mass of the grass component of a mixed prairie increased with increasing N-fertilization rate. In the current study, grazed crimson clover mass decreased with increasing N fertilization, in agreement with George et al. (1995) who reported that increasing N fertilization of a switchgrass-red clover mixture decreased mass of clover within the stand. As with N fertilization effects on botanical composition of pasture (i.e., proportions of component species), this effect is most likely related to differences in plant physiology and responsiveness to N fertilization that drive competition between grasses and legumes. Across all N-fertilization treatments, the presence of grazing cattle increased the DM mass of crimson clover and bermudagrass compared with ungrazed plots in the present study.

Ungrazed bermudagrass had greater foliar N concentration than grazed bermudagrass, in contrast to Chaneton et al. (1996) who reported that grazed grasses in

an Argentinian grassland contained greater foliar N concentration (2.23%) than ungrazed grasses (1.43%). Also, ungrazed bermudagrass receiving [50N + 100N] treatments had greater foliar N concentration than bermudagrass receiving 0N treatment. Brink et al. (2002) reported that increasing N fertilization of bermudagrass not only increased the foliar N concentration, but also foliar N mass.

Ungrazed bermudagrass and 'other' WS forages had greater foliar P concentration than grazed forages, but grazed triticale had greater foliar P concentration than ungrazed triticale. Chaneton et al. (1996) reported that, among forages in a grassland in Argentina, grazed forages contained greater foliar P concentration than ungrazed forages at the beginning of the season, but ungrazed forages contained greater foliar P concentration at the end of the season. The authors suggested that this is an adaptive response to defoliation; i.e., in order to be able to maintain growth under grazing pressure, plants reduce P transfer from roots to leaf and steam tissues. There was a greater foliar P concentration in grazed triticale than ungrazed; however, bermudagrass did not follow this pattern**.** Osborne and Rengel (2002) reported a foliar P mass efficiency in triticale of up to 44 mg foliar P/g P applied when a sufficient supply of P was available. By comparison, Brink et al. (2002) reported a foliar P mass efficiency of only 22 mg foliar P/g P applied to 'Alicia' bermudagrass.

Forage P mass was not affected by forage management across N-fertilization treatments. McLaughlin et al. (2004) reported foliar P mass values of 10.1 to 43.8 kg P/ha in common bermudagrass. These values are greater than those from the current study (4.7 to 6.6 kg P/ha); however, McLaughlin et al. (2004) applied N fertilizer at a greater rate (371 kg $N \cdot ha^{-1} \cdot yr^{-1}$) and applied P fertilizer as well (61 kg $P \cdot ha^{-1} \cdot yr^{-1}$), and

plots were harvested for hay production every 30 d. Grazed tall fescue had greater forage P mass in the [50N + 100N] treatments than the 0N treatment, in agreement with Vervoort et al. (1998) who reported increased foliar P and N mass in tall fescue with increasing N-fertilization rates. Also, P mass was greater in grazed bermudagrass for the 100N than 50N treatment. The opposite pattern was observed in ungrazed bermudagrass, with 0N having greater forage P mass than [50N + 100N]. Robinson (1996) reported that a positive correlation exist between forage mass and forage foliar P mass in hay harvested systems and, because the same trend was observed for mass of bermudagrass in the current study, these findings are not surprising. Forage P mass was greater in ungrazed triticale for the 100N than 50N treatment, but grazed triticale receiving 0N had greater forage P mass than the [50N + 100N] treatments.

Numerous studies (Lorenz and Rogler, 1972; Johnson et al., 2001) have reported increased yields in both cool- and warm-season forages with increasing N fertilization. Interestingly, mass of ungrazed bermudagrass was greater for 0N than [50N + 100N] in the present study. Mass of ungrazed forage DM was greater for WS than CS forage, and grazed CS forage had greater DM mass than ungrazed CS forage. Weeda and During (1987) reported that the presence of grazing cattle decreased standing herbage mass of cool-season pastures, and Burns and Sollenberger (2002) reported less standing herbage mass in grazed samples; however, the authors point out that overall seasonal production was greater in grazed than ungrazed pastures, even though standing herbage mass was typically less. These patterns were not observed in the current study, possibly due to the relatively low stocking densities employed.

Foliar N concentrations were greater in grazed than ungrazed forages, and the response was attributable primarily to differences in foliar N concentration between grazed and ungrazed WS forage. Chaneton et al. (1996) also reported that total-sward foliar N concentration was greater in grazed than ungrazed pastures, and this pattern was present in both early season samples and late season samples. Eriksen and Whitney (1981) observed increasing foliar N concentrations with increasing N-fertilization rate; however, their highest fertilization rate was 365 kg $N \cdot ha^{-1} \cdot yr^{-1}$, which is much greater than that in the current study. Also, plots were harvested every 8 wk, and forage was removed from the site. Because forage was removed, no foliar N was returned to the soil via grazing-animal excreta, and little was returned from crop residue decomposition. In the current study, the majority of foliar N was returned to the soil via animal excreta and crop residue decomposition because no forage was removed from the plots during the 3-yr study.

Foliar P concentrations were not affected by N fertilization rate, season or forage management. Chaneton et al. (1996) found similar results in total-sward foliar P concentrations, and percentage P was not different between grazed and ungrazed pastures in either early- or late-season samples. Balasko (1977) reported that foliar P concentrations in tall fescue in control and N-fertilized (240 kg $N \cdot ha^{-1} \cdot yr^{-1}$) plots did not differ (0.21%), and that foliar P concentrations decreased in both plots from early (0.21%) to late season (0.17%). Other studies (Sharpley et al., 1984b and Olander and Vitousek, 2000) suggest that forage in pasture with excessive soil N, and therefore foliar N mass, would have greater foliar mass of P as well. In the current study, plots received minimal to adequate amounts of N fertilization.

Mass of foliar P was greater for grazed than ungrazed CS forage, but greater for ungrazed than grazed WS forage, reflecting integration of patterns observed for DM mass and foliar P concentration. Evers (2002) observed that foliar P mass above high-P soils receiving broiler litter increased with increasing N-fertilization rate in cool-season grasses up to an application rate of 168 kg $N \cdot ha^{-1} \cdot yr^{-1}$. Nitrogen-fertilization rate did not have as much of an impact in the WS, but foliar P mass increased from 14.2 kg/ha to 18.6 kg/ha. Brink et al. (2001) reported crimson clover with a foliar P mass of 16 to 28 kg/ha in fields amended with 306 kg/ha N and 180 kg P/ha in the form of poultry litter. These values are considerably greater than those in the current study; however, Brink et al. (2001) applied both P and N, whereas there were no additional P inputs from outside the pasture system in the current study. Brink et al. (2001) concluded that the greater factor influencing nutrient foliar mass is not foliar concentration, but total DM mass of forage. These authors observed that P mass in warm-season grasses receiving both N and P in the form of poultry litter (9 $Mg \cdot ha^{-1} \cdot yr$) ranged from 43 to 50 kg P/ha. Foliar P mass of WS forages in the current study was 5.21 to 7.78 kg P/ha from plots that did not receive any fertilization with P. Brink et al. (2002) have stated that there is a positive correlation between DM mass and forage P mass; however, results of the current study indicate that available soil P is also a factor affecting both forage P concentrations and forage P mass.

Water Runoff

Pre-experiment water runoff had greater pH than samples collected during the experimental period. Long (1979) reported that water runoff from plots receiving 45 $mt \cdot ha^{-1} \cdot yr^{-1}$of dairy manure had greater pH than control plots receiving 370 kg $N \cdot ha^{-1} \cdot yr^{-}$

[1]. Sauer et al. (1999) reported that pH of water runoff from control plots (7.2) was less than from plots receiving urine and feces from a dairy (7.4). In contrast to the current study, these two studies utilized dairy manure (a mixture of feces, urine and bedding material), and ammonia in urine could conceivably explain the increase in pH of water runoff. In the current study, most ammonia from urine would either have volatilized or be adsorbed onto soil, resulting in little, if any, effect on pH; however, other non-volatile compounds such as nitrate could reduce the pH. Additionally, fertilization with ammonium sulfate-urea will decrease soil pH by releasing H^+ during the conversion of NH_4^+ to NO_3^-, which could also lead to decreased pH of water runoff. Electrical conductivity was not different between background and experimental samples nor among N-fertilization treatments. Long (1979) reported that the EC of control plots and manured plots was not different. Sauer et al. (1999) reported that the EC of plots receiving urine and feces from a dairy was greater (201 μohms/cm) than from control plots (162 μohms/cm). Sauer et al. (1999) utilized rainfall simulation 1 d after application of poultry litter and reported an increase in water runoff EC that was most likely due to dissolved nutrients from the poultry litter itself. However, in the current study and in Long (1979) in which manure was applied, natural rainfall produced runoff. In these latter experiments, nutrients from animal excreta could at least be partially returned to the soil prior to a significant rainfall event, thus producing no difference in EC values between manured or grazed plots and control plots.

In the current study, NH_4-N concentration in runoff was not affected by N-fertilization treatment, and was not different between pre-test and experimental samples. Schepers and Francis (1982) reported that pastures grazed with 1.2 cow-calf pairs/ha in

rotation and fertilized with 67 kg $N \cdot ha^{-1} \cdot yr^{-1}$ had the same amount of NH_4^+ in runoff as ungrazed pastures. Similarly, Long (1979) reported that NH_4^+ concentrations in runoff from control plots (370 kg $N \cdot ha^{-1} \cdot yr^{-1}$) was not different from plots receiving dairy cattle manure applied at a rate of 45 $mt \cdot ha^{-1} \cdot yr^{-1}$ so that plots were isonitrogenous. Sauer et al. (1999) observed greater NH_4^+ concentration in water runoff from plots receiving dairy feces and urine (5.07 mg/L) than control plots (0.13 mg/L).

Nitrate concentrations in water runoff were not different between treatments; however, background samples had greater concentration of NO_3-N than samples collected during the experiment. Other studies (Long, 1979; Schepers and Francis, 1982; Sauer et al., 1999) have reported an increase in NO_3^- concentrations when either grazing animals were present or plots received cattle manure. Values recorded in the current study were much lower than those reported in previous literature (Long, 1979; Schepers and Francis, 1982; Sauer et al., 1999).

In the current study, concentration of Kjeldahl-N was not different among seasons or between background and samples collected during the experiment. Both Long (1979) and Schepers and Francis (1982) reported greater Kjeldahl-N values in grazed plots than control plots. Sauer et al. (1999) reported slightly greater Kjeldahl-N concentrations in control plots (0.76 mg/L) than plots receiving dairy feces and urine (0.74 mg/L); however, these values are most likely not biologically significant, and are similar to concentration values recorded in the current study.

Phosphate concentrations in water runoff were greater in the experimental period than the background samples; however, they were not different among N-fertilization treatments. Schepers and Francis (1982) observed that runoff from grazed pastures (5.24

μg/L) had greater soluble-P concentration than from ungrazed pastures (4.67 μg/L). In contrast, Sauer et al. (1999) reported that pastures fertilized with dairy-cattle feces and manure had a phosphate concentration that was not different from control plots. Total P concentration was greater in runoff from 0N plots than [50N + 100N] plots, and experimental samples had a greater total P concentration than background samples. Schepers and Francis (1982) reported that total P in runoff was greater from grazed plots (2.14 μg/L) than ungrazed plots (1.28 μg/L).

Water runoff volume following significant precipitation events was not different between background (2009) and samples collected during the experimental period (2010 to 2012). However, N-fertilization treatment affected water runoff volume. The effect of treatment is possibly confounded with design of the watershed. Paddocks 1 and 2 were randomly selected as the 100N treatments and typically generated the least amount of runoff, whereas paddocks 4 and 6 assigned to the 0N treatment had a slightly greater slope and therefore generated not only more volume but also more runoff events than the other paddocks.

Ammonium concentration in water runoff did not differ between background and study-period samples, but 0N had greater total NH_4^+ concentration than [50N + 100N]. Steinman et al. (2003) reported similar findings, and also observed that NH4-N concentration decreased as grazing-animal density increased in improved pastures. Eghball and Gilley (1999) observed no difference in total amounts of NH_4-N from plots receiving manure, commercial fertilizer or a non-fertilized control. Total NO_3-N was not different among N-fertilization treatments in the current study, but was greater in the background samples than study-period samples. Eghball and Gilley (1999) observed

similar results, with a non-fertilized control having a numerically greater total NO_3-N concentration than both commercial fertilized and manured plots; however, the values were not different. Total N was not different between seasons, among treatments or between study-period and background samples. Steinman et al. (2003) observed similar results with improved pastures, with no difference in total Kjeldahl-N in water runoff regardless of whether grazing cattle were present. Additionally, Eghball and Gilley (1999) observed no difference in total N of plots receiving commercial fertilizer, cattle manure or no N fertilizer.

Total PO_4-P in water runoff was not different between background and study-period samples, but was greater in 0N than [50N + 100N] treatments. Steinman et al. (2003) reported that the presence of grazing cattle did not have an effect on total P concentration in water runoff. However, Eghball and Gilley (1999) reported that total dissolved P was greater in plots receiving commercial fertilizer than manured or control plots; manured and control plots were not different.

Total P was not different between background and study-period samples, but was greater in 0N than [50N + 100N] treatments. Eghball and Gilley (1999) observed no difference in total P of water runoff from commercial-fertilized, manured and control plots. Also, Steinman et al. (2003) reported no difference in total P in water runoff from pastures with and without cattle present. The results from the current study suggest that, with limited N availability, excess P can be lost in water runoff; however, when adequate soil-N is available, both PO_4-P and total P can be incorporated into soil to greater extent such that runoff has lower P concentration.

CONCLUSIONS

While different N-fertilization rates did not affect acid and alkaline soil phosphatase activity, soil pH, soil EC, or soil concentrations of water-soluble P or N, total soil P was reduced in plots receiving at least 50% of the agronomic recommendation for N for the grass component of a mixed species pasture system. However, increasing N fertilization to 100% in a split application resulted in no further reduction in soil-test P. Factors causing the greatest effect on soil P were animal and pasture management and time, suggesting that soil P can be reduced with proper N fertilization, forage, animal and soil management. Overall, forage DM mass, foliar P and N concentrations and forage P mass did not respond to differing N-fertilization rates; however, there was a response within individual forage species. Growing season and forage management had greater effects on forage characteristics than did N fertilization, similar to effects on soils, suggesting that foliar P mass can be increased with minimal N inputs, use of seasonally adapted annual forages, and incorporation of grazing animals. Nitrogen fertilization affected chemical properties of water runoff. Runoff contained greater concentrations of total P, PO_4-P and NH_4^+ in unfertilized than fertilized paddocks, possibly due to lower pasture productivity. Therefore, increasing N-fertilization to at least 50% of the agronomic recommendation for the grass component could reduce concentrations of the nutrients and, in order to decrease nutrient concentrations in water runoff and to increase forage P mass and soil quality in a year-round grazed grass-legume pasture, producers might apply N fertilizer at a rate of 50% of the recommendation for the grass species within the stand.

III. NITROGEN FERTILIZATION IMPACTS ON INTAKE AND FECAL EXCRETION OF PHOSPHORUS BY GRAZING BEEF CATTLE

INTRODUCTION

Nutrients such as P are potentially a major concern when considering quality of water runoff from beef cattle operations. According to Williams and Haynes (1990), 70% of P that is ingested by grazing cattle is excreted and returned to soil in feces. When using manure from an animal feeding operation, manure is evenly spread across a pasture using a mechanical spreader or irrigation. However, in grazed pastures, urine and feces are not uniformly distributed throughout the pasture. A single beef-cow dung patch covers approximately 0.06 m^2 (Fame, 1971). When calculated on an annual basis, the total surface area of a pasture that receives dung may only be 27 to 40% of the total area (Saunders, 1984). Jawson et al. (1982) reported that total P losses were about 8 to 12 times greater from grazed than un-grazed watersheds; however, grazing did not present much of a nutrient pollution hazard.

The objective of the current study was to determine the effect of N fertilization on intake and fecal excretion of P by grazing cattle, and to characterize temporal patterns of P return to the soil ecosystem from fecal pats. We hypothesized that N fertilization would alter available forage DM mass and foliar P concentration such that P intake and thus fecal excretion of P by cattle would be modified. Secondly, we hypothesized that

pasture response to N fertilization would affect temporal patterns of disappearance of fecal pats such that feces from cattle grazing during the summer would disappear more rapidly and result in greater soil N and P concentrations than feces from cattle grazing during winter/spring.

MATERIALS AND METHODS

Research Site, Forage Establishment and Cattle Grazing management

The experiment was conducted from October, 2010 through January, 2013 at the Auburn University Stanley P. Wilson Beef Teaching Center in Auburn, Alabama (32° 53' 34.43" N latitude, 85° 30' 3.32" W, 187 m above MSL). A complete description of the research site, forage establishment, and cattle grazing management is provided in Materials and Methods of Chapter II.

Chromium Pellet Fabrication

In August 2010, chromic oxide (Cr_2O_3)/corn pellets were fabricated for administration to grazing cattle in order to determine indirectly their daily fecal DM output by Cr dilution technique. A mixture of Cr_2O_3 and ground corn was blended in a ratio of 20.4 kg corn:2.3 kg Cr_2O_3 and fabricated into pellets containing 6.8% Cr by weight. Pellets were formed using a laboratory-scale pellet mill (model CL5, California Pellet Mill Co., San Francisco, CA) using a 4.76-mm dye. Pellets were dried for 24 h at room temperature and stored in an air-tight container until use.

Phosphorus Intake and Fecal Excretion

Daily forage DM intake was estimated by first determining fecal excretion of Cr from consumption of Cr_2O_3/corn pellets. Cattle were individually fed 50 g ground corn twice daily for 10 d to familiarize them with hand-feeding by personnel. Next, cattle were individually fed 10 g/d of Cr_2O_3/corn pellets for 7 d at 0800 h. On d 7, each animal was moved at 0800 h to a holding facility at the Stanley P. Wilson Beef Teaching Unit where feces were collected from a concrete floor immediately following excretion and placed into a bucket. All feces from 0800 to 1500 h from each animal were mixed, and a 500-g subsample (wet wt) was retained for Cr analysis. This experiment was conducted twice, midway and late in the grazing season, for each of CS (March 31, 2011, March 1, 2012 and April 21, 2011, April 20, 2012, respectively) and WS (August 31, 2011, September 5, 2012 and September 14, 2011, September 19, 2012, respectively) seasons in 2011 and 2012. Prior to analyses, fecal samples were dried at 60° C for 72 h and ground with a Wiley Mill (Thomas Scientific, Philadelphia, PA) to pass a 1-mm screen.

In order to enable calculation of forage DM intake, 3 forage samples were clipped in the vicinity of where cattle had been observed to recently graze in each plot within 7 d of fecal collection. Forage samples were mixed, dried at 60° C for 72 h, and ground with a Wiley Mill (Thomas Scientific, Philadelphia, PA) to pass a 1-mm screen. Forage *in vitro* dry matter digestibility (IVDMD) was determined according to the Van Soest et al. (1991) modification of the Tilley and Terry procedure (1963) using the Daisy^{II} incubator system (Ankom Technology^{TM}). Ruminal fluid was collected from a fistulated, dry Holstein cow at the Auburn University College of Veterinary Medicine. The cow was fed a corn silage-based diet containing cottonseed meal and Megalac^{TM} supplement, and

given free access to bermudagrass pasture and alfalfa (*Medicago sativa*) hay. Fluid was stored in pre-warmed thermos containers to maintain a temperature supportive of the microbial population and transported to the Auburn University Ruminant Nutrition laboratory where it was then prepared for the batch-culture IVDMD procedure.

Fecal samples were analyzed for Cr using dry-ashing followed by ICAP spectroscopy (Hue and Evans, 1986). Total fecal Cr in feces was used to determine total fecal output using the following equation: daily fecal output = $\frac{Cr\ intake}{Cr\ concentration\ in\ feces}$. Forage DM intake was calculated using the equation: daily forage DM intake = fecal DM output / % indigestibility (Streeter, 1969). Intake and fecal excretion of P were then calculated by multiplying forage and fecal concentrations of P by forage DM intake and fecal DM output, respectively.

Fecal Pat Degradation

Twenty-four 1-m^2 plots were demarcated outside of the larger pasture plots at the research site. These plots were mowed to a 4-cm height and sprayed with glyphosate prior to fecal-pat degradation experiments conducted over a 2-yr period. The study was arranged as a completely randomized design with two replications. Prior to feces collection in each of cool and warm grazing seasons, three 20-cm soil samples were taken from each 1-m^2 plot and separated into 0 to 5, 5 to 10 and 10 to 20 cm depth strata. In the CS and WS of 2011 (April 21 and September 14, 2011, respectively) and 2012 (April 11 and September 12, 2012, respectively), feces were collected directly from each animal in each pasture. Each animal was brought to the Stanley P. Wilson Beef Teaching Unit holding facility at 0800 h where feces were collected from a concrete floor immediately

following excretion and placed into a bucket. All feces from 0800 to 1500 h from each individual were composited and allocated to 0-, 28-, 56-, 84- and 112-d after application (DAA) treatments, and a 0.5-kg (wet basis) fecal sample was prepared for each DAA treatment. Zero-DAA fecal samples were taken directly to the laboratory and dried at 60° C for 72 h. All remaining treatment aliquots were transported to the experimental plots and randomly placed in the center of the 1-m^2 plot. Feces were applied in a circular fashion until a 20-cm fecal pat was formed that simulated an animal defection.

On the assigned treatment DAA, remaining feces that had not decomposed was recovered and weighed. Three additional soil samples were taken from directly beneath fecal pats and separated into 0 to 5, 5 to 10 and 10 to 20 cm depth strata. Feces and soil were dried at 60° C for 72 h. Fecal samples were ground with a Wiley Mill (Thomas Scientific, Philadelphia, PA) to pass a 1-mm screen, and all soil samples were sieved to pass a 2-mm screen.

Soil and fecal concentrations of N and C were determined via dry combustion using a LECO TruSpec CN Analyzer (LECO Corp, St Joseph, MI). Soil samples were extracted using dilute HCl and HNO_3 (Mehlich I), and analyzed by ICAP spectroscopy to determine P, K, Ca, Mg, Zn and Cu (Hue and Evans, 1986; SPECTRO CIROS CCD). Concentrations of P, K, Ca, Mg, Zn and Cu in fecal samples were analyzed by dry-ashing followed by ICAP spectroscopy (Hue and Evans, 1986). Fecal samples were also analyzed for water-extractable P using the Murphy and Riley (1962) method.

Total remaining nutrients in feces were calculated by the following equation: Nutrient remaining = $([N_x] * D_x)/1000$ where $[N_x]$ is the concentration of the nutrient on a given DAA and D_x is the dry weight of the feces on a given DAA.

Statistical Analysis

Data were analyzed as a completely randomized design using PROC MIXED of Statistical Analysis Software (SAS, Carey, NC). Fecal nutrients and P utilization data were analyzed using N-fertilization treatment, season and their interaction as main effects. The soil nutrient model included N-fertilization treatment, season, soil depth, DAA and their interactions as main effects. Orthogonal contrasts were used to compare the 0N treatment with 50N and 100 N treatments, and 50N and 100N treatments. In recognition of the low statistical power characteristic of field studies that employ limited numbers of replicates α was set to equal 0.10 (Peterman, 1990).

RESULTS

Phosphorus intake and fecal excretion

Phosphorus intake by cattle (Table 29) was not different between grazing seasons or among N-fertilization treatments ($P > 0.10$). A treatment × season interaction (P 0.087) existed such that fecal P output in CS increased with N fertilization but was not affected in WS. Fecal output of water-soluble P was not different among treatments or between seasons ($P > 0.10$)

Phosphorus concentration in forage available to cattle during the experimental period (Table 30) was not different between seasons or among treatments ($P > 0.10$). However, forage DM mass was greater ($P = 0.008$) in CS forages than WS forages, and P mass was greater ($P = 0.023$) in CS forage than WS forage. Neither DM mass nor forage P mass were different among N-fertilization treatments ($P > 0.10$).

Table 29. Phosphorus intake, fecal P output and fecal water-soluble P output of cattle grazing pastures receiving different N-fertilization treatments

						Contrasts			
		N Treatment [a]				0N vs. [50N + 100N]		50N vs. 100N	
Item	Season[b]	0N	50N	100N	Mean	*P* value	SE[c]	*P* value	SE[c]
P intake (g/d)									
	CS	11.2	25.1	33.4	23.2	0.229	14.7	0.578	14.7
	WS	14.2	14.6	13.7	14.2	0.998	12.3	0.951	14.2
	Mean	12.7	19.8	23.5	18.7				
P output (g/d)									
	CS	7.1	12.1	29.5	16.2	0.154	9.4	0.093	10.1
	WS	16.9	17.0	15.8	16.6	0.954	8.5	0.905	10.1
	Mean	12.0	14.6	22.7	16.4				
Water-soluble P output (g/d)									
	CS	1.8	0.6	1.5	1.3	0.717	2.1	0.718	2.4
	WS	2.6	4.4	1.4	2.8	0.876	2.1	0.205	2.4
	Mean	2.2	2.5	1.4	2.1				

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]n = 6.

Table 30. Forage DM mass, foliar P concentration and forage P of pastures receiving different N-fertilization treatments

		N Treatment [a]				
Item	Season[b]	0N	50N	100N	Mean	SE[c]
DM mass (kg/ha)						
	CS	3,360	3,580	3,945	3,628[d]	
	WS	2,722	2,492	2,622	2,612[e]	
	Mean	3,401	3,036	3,284	3,120	258
P concentration (%)						
	CS	0.28	0.24	0.22	0.25	
	WS	0.23	0.28	0.22	0.24	
	Mean	0.25	0.26	0.22	0.25	0.01
Forage P mass (kg/ha)						
	CS	3.4	3.6	3.9	3.6[f]	
	WS	2.7	2.5	2.6	2.6[g]	
	Mean	3.1	3.1	3.3	3.1	0.3

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]n = 6.
[d,e]Within a column, means without a common superscript differ ($P < 0.10$).
[f,g]Within a column, means without a common superscript differ ($P < 0.05$).

Fecal Degradation

The P concentration of soil (Table 31) beneath fecal pats was not affected by N-fertilization treatment; however, soil in the WS contained a greater P concentration than in CS. Also, the 0 to 5 cm depth interval (115.1 mg/kg) had greater ($P < 0.001$) P concentration than both 5 to 10 and 10 to 20 cm (36.4 and 43.3 mg/kg, respectively); however, 5 to 10 and 10 to 20 cm were not different ($P > 0.10$). Concentration of soil P on 0 DAA (31.8 mg/kg) was less ($P < 0.060$) than on both 56 DAA (67.0 mg/kg) and 112 DAA (52.9 mg/kg), but was not different from 28 DAA (98.6 mg/kg) and 84 DAA (74.6 mg/kg). Also, soil on 28 DAA had greater ($P = 0.033$) soil P concentration than on 56 DAA.

The N concentration of soil (Table 32) beneath fecal pats was not affected by DAA or season. The 0N (1.3 mg/kg) treatment was not different from [50N + 100N] ($P > 0.10$); however, 100N (1.5 mg/kg) had a greater ($P = 0.018$) N concentration than 50N (0.12%). Also, N concentration decreased as soil depth increased ($P < 0.061$), with 0 to 5 cm depth interval (2.3 mg/kg) having the greatest N concentration, 5 to 10 cm having the median value (0.10%), and 10 to 20 cm having the least value (0.8 mg/kg). There was a season × N-fertilization treatment interaction such that 0N (1.2 mg/kg) had lesser ($P = 0.001$) N concentration than [50N + 100N] (1.4 mg/kg), and 50N (1.2 mg/kg) had lesser ($P = 0.050$) N concentration than 100N (1.7 mg/kg) in the CS.

Percent of fecal-pat DM remaining (Table 33) was not different among N-fertilization treatments or between seasons. The percent remaining at 28 DAA (52.2%) was greater ($P < 0.037$) than at 112 DAA (31.2%), but was not different ($P > 0.10$) from

56 and 84 DAA (55.4 and 45.7%, respectively). Percent remaining at 112 DAA was less ($P < 0.037$) than at 0, 28 and 56 DAA.

Percent remaining P in fecal pats (Table 34) was not different among treatments or between seasons ($P > 0.10$). 0 DAA had a greater ($P < 0.002$) percent remaining (100.0%) than all other DAAs. Additionally, 28 DAA (64.5%) had a greater ($P < 0.0001$) remaining P than 84 (41.8%) and 112 DAA (25.9%). However, 56 (45.4%), 84 and 112 DAA were not different ($P > 0.10$). Within the WS, percent remaining of P at 112 DAA was greater in 100N than 50N, and greater in [50N + 100N] than 0N].

Percent remaining of water-soluble P (Table 35) was not different among treatments or between seasons ($P > 0.10$). Additionally, there was no difference among DAA ($P > 0.10$). Within the WS, the percent reaming of water-soluble P at 112 DAA was greater in 100N than 50N, and 100N was greater than 50N in the CS at 28 DAA.

Percent remaining of N (Table 36) was not different among treatments or between seasons ($P > 0.10$). 0 DAA (100.0%) had a greater ($P < 0.001$) than all other DAAs and 28 DAA (65.9%) had a greater percent remaining of N than 84 (48.8%) and 112 DAA (34.5%). However, 56 (51.1%), 84 and 112 DAA were not different ($P > 0.10$).

Table 31. Extractable P concentration (mg/kg) in soil from beneath feces from cattle grazing pastures receiving different N-fertilization treatments

		Cool Season					Warm Season				
		N Treatment [a]					N Treatment				
DAA[b]	Depth, cm	0N	50N	100N	Mean	SE[c]	0N	50N	100N	Mean	SE[c]
0	0 to 5	31.4	42.3	41.9	38.5	47.0	30.8	34.1	32.8	32.6	35.2
	5 to 10	32.7	30.0	37.7	33.4	35.2	24.6	24.5	23.8	24.3	35.2
	10 to 20	33.7	32.9	40.6	35.7	47.0	23.3	25.5	29.4	26.1	35.2
28	0 to 5	64.6	51.0	60.2	58.6	42.3	249.3	484.5	341.2	358.3	35.2
	5 to 10	38.9	27.0	37.8	34.6	35.2	59.7	78.1	55.5	47.8	35.2
	10 to 20	37.3	29.4	53.4	40.0	35.2	34.4	35.5	37.8	35.9	35.2
56	0 to 5	54.4	39.8	13.6	35.9	49.8	185.2	358.4	88.6	210.8	37.1
	5 to 10	42.7	16.5	13.8	24.3	45.5	56.3	63.5	89.1	64.4	37.1
	10 to 20	33.5	30.5	21.5	28.5	49.8	32.3	48.7	17.4	32.8	40.7
84	0 to 5	20.6	40.0	74.6	45.1	35.2	205.2	244.8	183.9	211.3	37.1
	5 to 10	24.5	27.5	35.2	29.1	49.8	86.5	55.0	62.9	69.6	37.1
	10 to 20	34.5	37.2	76.6	49.4	39.0	51.2	39.9	42.2	44.4	37.1
112	0 to 5	36.5	32.4	43.3	37.4	35.2	96.8	187.5	85.5	123.3	35.2
	5 to 10	28.3	20.3	63.8	37.4	43.9	37.2	66.0	40.2	68.1	35.2
	10 to 20	32.4	19.7	48.2	33.4	37.1	35.5	49.0	29.1	37.8	35.2
			Season Mean		37.4[d]	11.0		Season Mean		92.5[e]	9.4

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]DAA = days after application
[c]n = 6.
[d,e]Within a row, means without a common superscript differ ($P < 0.001$)

Table 32. Nitrogen concentration (mg/kg) in soil beneath feces from cattle grazing pastures receiving different N-fertilization treatments

		Cool Season					Warm Season				
		N Treatment [a]					N Treatment				
DAA[b]	Depth, cm	0N	50N	100N	Mean	SE[c]	0N	50N	100N	Mean	SE[c]
0	0 to 5	0.7	0.8	2.3	1.3	0.3	2.1	1.7	1.6	1.8	0.2
	5 to 10	0.9	0.6	1.9	1.1	0.3	1.2	1.3	1.0	1.2	0.2
	10 to 20	0.7	0.7	2.1	1.2	0.3	0.4	0.9	0.7	0.7	0.2
28	0 to 5	2.4	1.7	2.8	2.3	0.2	3.4	2.3	3.2	3.0	0.2
	5 to 10	1.4	0.7	0.9	1.0	0.2	1.0	1.0	0.8	0.9	0.2
	10 to 20	1.1	1.1	0.6	0.9	0.2	0.1	0.4	0.3	0.3	0.2
56	0 to 5	2.7	2.7	1.7	2.4	0.3	2.6	2.4	2.7	2.6	0.3
	5 to 10	0.7	0.9	0.6	0.8	0.3	0.8	0.8	0.8	0.8	0.2
	10 to 20	0.4	0.3	0.5	0.4	0.3	0.3	0.4	0.4	0.4	0.2
84	0 to 5	1.8	1.8	2.0	1.9	0.2	3.1	3.1	2.4	2.9	0.3
	5 to 10	0.6	0.7	1.9	1.2	0.2	1.1	1.0	1.2	1.1	0.2
	10 to 20	0.8	0.8	2.3	1.3	0.3	0.8	0.5	0.4	0.6	0.2
112	0 to 5	2.1	1.7	3.1	2.3	0.2	2.6	2.8	2.8	2.7	0.2
	5 to 10	0.7	0.9	1.9	1.2	0.2	0.3	0.7	0.7	0.6	0.2
	10 to 20	0.5	1.9	2.1	1.2	0.2	1.2	0.4	0.4	0.7	0.2
			Season Mean		1.3	0.1		Season Mean		1.3	0.1

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]DAA = days after application
[c]n = 6.

Table 33. DM mass (% of initial) remaining in fecal pats from cattle grazing pastures receiving different N-fertilization treatments

		N Treatment [a]				
Season[b]	DAA[c]	0N	50N	100N	Mean	SE[d]
CS						
	0	100.0	100.0	100.0	100.0	9.3
	28	51.8	49.0	47.5	49.4	12.2
	56	50.3	53.7	45.4	49.8	9.3
	84	58.0	49.4	53.3	53.6	9.3
	112	52.9	48.4	19.2	40.2	12.3
WS						
	0	100.0	100.0	100.0	100.0	9.4
	28	47.8	61.8	55.5	55.0	9.5
	56	54.2	77.7	50.8	60.9	12.3
	84	24.0	46.4	42.8	37.7	12.2
	112	19.8	6.5	40.4	22.2	9.5

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]DAA = days after application
[d]n = 6.

Table 34. Phosphorus remaining in fecal pats (% of initial) from cattle grazing pastures receiving different N-fertilization treatments

						Contrasts			
		N Treatment [a]				0N vs. [50N + 100N]		50N vs. 100N	
Season[b]	DAA[c]	0N	50N	100N	Mean	*P* value	SE[d]	*P* value	SE[d]
CS									
	0	100.0	100.0	100.0	100.0	1.000	20.4	1.000	20.4
	28	36.0	65.6	44.6	48.9	0.499	21.0	0.344	21.0
	56	33.7	47.3	40.9	40.6	0.658	22.3	0.782	22.3
	84	32.5	27.2	43.9	34.5	0.688	20.4	0.417	20.4
	112	13.6	31.4	33.0	26.0	0.403	22.0	0.607	22.0
WS									
	0	100.0	100.0	100.0	100.0	1.000	20.4	1.000	20.4
	28	86.9	92.0	61.0	80.0	0.714	31.0	0.451	31.0
	56	70.7	45.7	33.8	50.1	0.301	36.9	0.527	36.9
	84	49.6	50.2	47.4	49.1	0.620	35.3	0.800	35.3
	112	0.0	0.0	77.4	25.8	0.018	28.8	0.010	28.8

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]DAA = days after application
[d]n = 6.

Table 35. Water-soluble P remaining in fecal pats (% of initial) from cattle grazing pastures receiving different N-fertilization treatments

						Contrasts			
		N Treatment [a]				0N vs. [50N + 100N]		50N vs. 100N	
Season[b]	DAA[c]	0N	50N	100N	Mean	*P* value	SE[d]	*P* value	SE[d]
CS									
	0	100.0	100.0	100.0	100.0	1.000	56.4	1.000	56.4
	28	107.3	46.9	158.5	104.2	0.329	56.4	0.054	56.4
	56	67.2	85.2	73.4	75.3	0.879	55.2	0.763	57.9
	84	45.5	103.4	21.2	56.7	0.536	56.4	0.240	69.1
	112	23.5	128.4	90.5	80.8	0.211	65.1	0.564	37.9
WS									
	0	100.0	100.0	100.0	100.0	1.000	60.4	1.000	60.4
	28	42.1	85.4	146.0	91.2	0.293	97.7	0.435	60.4
	56	59.9	85.4	110.1	85.1	0.494	72.8	0.332	65.7
	84	89.9	105.8	94.2	96.6	0.911	65.1	0.912	97.7
	112	0.0	0.0	85.7	28.6	0.501	65.1	0.084	72.8

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]DAA = days after application
[d]n = 6.

Table 36. Nitrogen remaining in fecal pats (% of initial) from cattle grazing pastures receiving different N-fertilization treatments

		N Treatment [a]				
Season[b]	DAA[c]	0N	50N	100N	Mean	SE[d]
CS						
	0	100.0	100.0	100.0	100.0	12.9
	28	59.2	46.6	42.5	49.4	14.9
	56	54.1	38.2	41.3	44.5	13.2
	84	49.8	37.6	15.8	34.4	12.9
	112	32.3	34.8	22.0	29.7	32.3
WS						
	0	100.0	100.0	100.0	100.0	12.9
	28	79.3	92.1	75.7	82.4	18.3
	56	70.7	62.3	40.0	57.6	25.9
	84	33.4	105.3	49.9	62.9	18.3
	112	67.5	0.0	50.2	39.2	12.9

[a]0N = 0% N fertilization, 50N = 50% N fertilization, 100N = 100% N fertilization based on N requirement of grass species.
[b]CS = cool season and WS = warm season.
[c]DAA = days after application
[d]n = 6.

DISCUSSION

Phosphorus intake fecal excretion

Daily intake and fecal excretion of P by grazing cattle were not different between seasons or among N-fertilization treatments. According to NRC (1996), medium-framed growing beef cattle require approximately 10 to 12 g P/d. In the current study, cattle were consuming at least 11.2 g P/d, which is within the recommended intake. Karn (2001) has stated that cattle grazing forage typically only become P deficient when soil is deficient in P because foliar P concentration and animal performance are both positively correlated with soil P concentration. Several studies (Karn, 1997, Winter, 1990 and Winks et al, 1977) have reported that fecal P excretion is directly linked to P intake, and that a positive correlation exists between increased P in the diet and increased fecal P excretion. Winks et al. (1977) have suggested that dietary P intake could be estimated from fecal P excretion. Evers (2002) observed that P uptake from high-P soils increased with increasing N-fertilization rate in CS grasses up to an application rate of 168 kg $N \cdot ha^{-1} \cdot yr^{-1}$; in WS; N-fertilization rate did not have as much of an impact, but P uptake increased from 14.2 kg/ha to 18.6 kg/ha. Increased foliar P uptake could explain increased P intake and fecal P excretion observed in the CS of the current study. Foliar concentration of water-soluble P was not different between seasons or among treatments, but the mean concentration (0.2 g/kg) is similar to values (~ 0.7g/kg) reported by Kleinman et al. (2005) for fresh beef cattle feces.

Concentration of P in forages consumed by cattle within 7 d of intake measurements was not different between seasons or among treatments, but P mass tended to be greater in CS than WS because forage DM mass was greater in the CS than WS. Foliar P concentrations and, therefore, forage P mass are affected by soil P status, P fertilization, stage of forage maturity, forage management practices, P apportionment among individual plant species, and meteorological conditions (Karn, 2001).

Fecal Degradation

Concentration of P in soil beneath fecal pats was not affected by treatment, but was greater in WS than CS; surface soils had a greater P concentration than deeper soil layers. Chardon et al. (2007) reported that P concentration in soil beneath cattle manure using a lysimeter had the greatest P concentration from 0 to 2 cm (648 mg/kg), and P concentration decreased until 8 to 10 cm (236 mg/kg). Similarly, Lithourgidis et al. (2007) reported that soil P increased initially after manure application, but returned to approximately initial soil P values, in agreement with the finding in the current study that soil P concentration in the CS returned to approximately initial soil P concentration by 112 DAA.

The N concentration of soil beneath fecal pats was not affected by DAA or season; however, soil N concentration was greatest at the greatest N-fertilization rate. Lithourgidis et al. (2007) reported similar findings; i.e., that N concentration in soils beneath applied cattle manure did not differ between the beginning and end of the experiment (0- to 30- cm soil depth). Also, soil N concentration decreased with increasing soil depth in the current study.

Percent of DM remaining in fecal pats was not different between seasons or among treatments. Lupwayi and Haque (1999) reported that cattle manure pats had only been reduced by 15% after 15 weeks. Brown (2010) reported that cattle manure applied in the winter had decayed by 25% in 140 d, and in the summer by 46% in 140 d. Mundus et al. (2008) reported that cattle manure decomposed to approximately 50% of initial mass in 80 d. In the current study, fecal pats were reduced by an average 68.8% in 112 d which is greater than values reported by Lupwayi and Haque (1999), but similar to both Brown (2010) and Mundus et al. (2008).

Water-soluble P remaining in fecal pats were affected by season, N-fertilization treatment and DAA, and total P was not affected by season or N-fertilization. Whereas mass of total P and water-soluble P remaining in fecal pats were not different among DAA, values decreased over time. Esse et al. (2001) reported that P disappearance from fecal pats mirrored disappearance of organic matter of soil-applied manure, consistent with the current study in which values for percent of DM remaining and percent of P remaining are similar at every DAA. Lupwayi and Haque (1999) reported that, after 15 wk, 80% of P remained within manure pats; however, the experiment only used a 10 g (DM-basis) sample, and disappearance was much less than in the current study for which approximately 74% disappearance of P was observed. Chardon et al. (2007) stated that cow manure, due its large proportion of water-soluble P, can be a long-term source of P that can be transported either by surface water runoff or be leached into the soil. Results of the current study support this concept because almost half of the initial water-soluble P in fecal pats was still remaining in the pats 112 days later. Additionally, the water-soluble P in the fecal pats increased from 0 DAA to 28 DAA in the CS. Kleinman et al.

(2005) state that manure P has a long-term effect on soil due to the slow decomposition by macro- and microorganisms within the soil and on the surface. Therefore, the increase in water-soluble P could possibly be due to conversion of organic P in the manure into water-soluble P.

Remaining N in fecal pats was not different between seasons. Esse et al. (2001) observed that N liberation from fecal pats, much like P, mirrored that of fecal pat DM disappearance; this pattern was observed in the current study, with the greatest N content recorded at 0 DAA and all other subsequent DAA having decreased total N. Mundus et al. (2008) reported that the percent N remaining in surface-applied cattle manure was less than 20% in only 80 d; however, this experiment was conducted on a small scale (10 g DM) and, therefore, due to increased fecal pat surface area to volume ratio, decomposition and nutrient transfer would be assumed to occur at a faster rate.

CONCLUSIONS

Neither N fertilization regime nor grazing season affected intake or fecal excretion of water-soluble P by grazing cattle, but total P excretion was greater with greater N-fertilization application in the CS. These findings indicate that cattle P requirements were met and that sufficient P was returned to pasture to meet forage requirements in the absence of fertilization with N. There was no effect of N fertilization on the decomposition of or P removal from fecal pats, or on P concentration in soil beneath fecal pats. However, N fertilization increased N removal from and increased soil N concentrations beneath fecal pats. These observations indicate that in grazed pastures with high soil-test P, N-fertilization did not affect intake and fecal returns of P, foliar uptake of P and rate or extent of assimilation of P returns into the soil profile from degradation of fecal material.

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