# Runoff nutrient losses from tall fescue pastures varying in endophyte association, fertilization, and harvest management

D.M. Endale, H.H. Schomberg, A.J. Franzluebbers, D.S. Seman, D.H. Franklin, and J.A. Stuedemann

Abstract: Tall fescue (Lolium arundinaceum [Schreb.] S.J. Darbysh.) is a widely adopted cool-season perennial forage in the southeastern United States. Historically, tall fescue contained a symbiotic fungal endophyte, Epichloë coenophiala, that produced an ergot alkaloid toxic to livestock. New fescue biotypes have been developed that are endophyte free or include endophytes that produce low-toxicity alkaloids. Ecological implications of these biotypes are not fully understood. We evaluated nutrient runoff quality for three tall fescue biotypes (common, high alkaloid endophyte [wild]; low alkaloid endophyte [novel]; and endophyte free [free]) under two fertilizer sources (broiler litter [BL] and inorganic fertilizer) in grazed systems and a haved system with inorganic fertilization of endophyte free fescue. The seven treatments were replicated twice on 1 ha paddocks near Watkinsville, Georgia. Grazed paddocks were stocked in September each year with weaned Angus (Bos taurus) heifers (2002 to 2007) and steers (2008 to 2009) at rates of ~3 to 6 head paddock<sup>-1</sup> adjusted every 28 days to maintain 1 to 3 Mg ha<sup>-1</sup> of available forage. Weather conditions varied from very dry to very wet and allowed 15 grazing periods (50 to 198 days). Tall fescue with novel-endophyte association had lower or equal runoff concentrations and loads for inorganic nitrogen (N), total N (TN), ortho-phosphorus (ortho-P), and total P (TP) than fescue with wild- or free-endophyte association (fescue treatment mean concentrations [mg L<sup>-1</sup>]: <0.8 inorganic N, <4.0 TN, <3.5 ortho-P and TP; mean loads [kg ha<sup>-1</sup>]: <0.08 inorganic N, <0.40 TN, <0.30 ortho-P and TP; all original untransformed data). This indicates increased adoption of novel-endophyte fescue, as previously recommended, presents no additional risk for nutrient losses regardless of fertilization source. Weather variability had impact on assessed variables. Mean total runoff across all paddocks during above average (27.2%) and average (20.6%) monthly rainfall periods were 15.2- and 1.9-fold greater than that from the below average (52.2%) monthly rainfall period. Nutrient loads also were greater: 3.4-fold (TP) to 14.6-fold (ammonium-N). Pollutant loads in the upper quartile were several fold greater than those from the rest of the data. The presence of cattle increased ortho-P load up to 3-fold compared with periods with no cattle.

**Key words:** broiler litter—extreme rainfall—fescue toxicosis—grazing—livestock—water quality

Tall fescue (Lolium arundinaceum [Schreb.] S.J. Darbysh.) is a cool-season perennial forage grown on more than 15 million ha in the United States, primarily in the east-central, mid-southern, and the Pacific Northwest regions (Rogers and Locke 2013; Hoveland 2009; Hannaway et al. 2009). It establishes easily, tolerates variable soil and climatic conditions, and provides good forage availability (Belesky and West 2009; Fribourg et al. 2009), which reduces

stored feed requirements compared to other cool-season grasses (Roberts et al. 2009). It is well adapted for lawn, turf, and conservation uses (Hoveland 2009).

Historically, tall fescue contained a symbiotic endophytic fungus, *Epichloë coenophialum*, which is transmitted in the seed (Leuchtmann et al. 2014; pre-2014 genus *Neotyphodium coenophialum* [Shelby and Darlymple 1987]; Bacon and Siegel 1988; USDA NRCS 2001; Roberts et al. 2009).

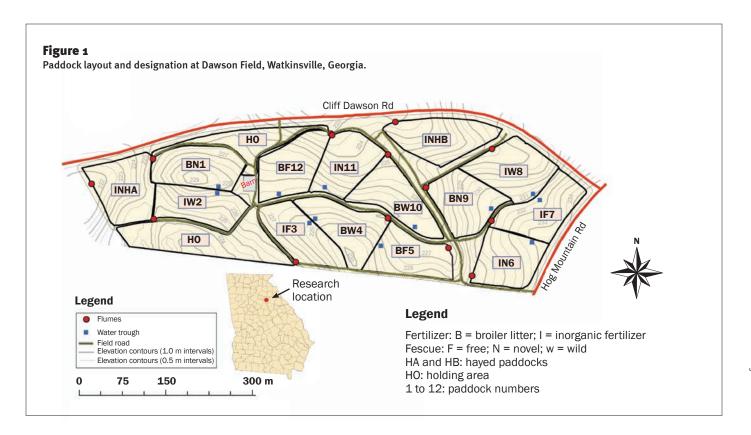
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The symbiosis enhances tall fescue tolerance to biotic and abiotic stresses, thus improving growth, reproduction, and persistence. However, the endophyte produces the ergot alkaloids peramine and loline (Rogers and Locke 2013), which reduce herbivory and adversely affect grazing animal performance and productivity, especially during spring and fall (fescue toxicosis) (Stuedemann and Hoveland 1988; Hill et al. 1994; Bouton et al. 2002; Hoveland 2009). Beef industry losses due to fescue toxicosis range from US\$0.5 to US\$1 billion annually (Rogers and Locke 2013; Gunter and Beck 2004).

Endophyte-free or novel nontoxic ergot alkaloid producing endophyte lines of tall fescue have been developed for grazing. Endophyte-free tall fescues have demonstrated low persistence (Waller 2009; Hopkins et al. 2009), while novel-endophyte tall fescues have demonstrated animal performance comparable to endophyte-free tall fescue and persistence comparable to wild-endophyte tall fescue (Bouton et al. 2002; Waller 2009; Belesky and West 2009). The first of the commercially available novel-endophyte biotypes, MaxO, was a 'Jesup' tall fescue cultivar hosting a novel endophyte AR542 (Pennington Seed Co., Madison, Georgia) (Bouton et al. 2002).

Beef cattle production in the southeastern United States is typically located on less productive crop lands. Franzluebbers (2007) reported that 53% of farms in the region had cattle and 41% had pastures with a mean area of 30 ha. Many of these farms have a mixture of poultry (*Gallus gallus domesticus*) and beef cattle production. The combination is advantageous since the broiler litter (BL; 1.0 kg bird<sup>-1</sup>) (Ritz and Merka 2013) can be applied to nearby pastures as fertilizer (more than 40% in Georgia), or sold (Starkey 2003).

Dinku M. Endale (corresponding author) is an agricultural engineer with the USDA Agricultural Research Service (ARS), Tifton, Georgia. Harry H. Schomberg is an ecologist with the USDA ARS, Beltsville, Maryland. Alan J. Franzluebbers is an ecologist with the USDA ARS, Raleigh, North Carolina. Dwight S. Seman is a retired animal scientist with the USDA ARS, Lexington, Kentucky. Dorcas H. Franklin is an associate professor with the University of Georgia, Crop and Soil Sciences Department, Athens, Georgia. John A. Stuedemann was a retired animal scientist (deceased) at the USDA ARS, J. Phil Campbell Sr. Natural Resource Conservation Center, Watkinsville, Georgia.



Repeated application of BL on pastures can lead to excessive loading of nutrients, which, along with the relatively high annual rainfall and high runoff potential across the southeastern United States, (~1,250 mm) raises concern about edge-of-pasture sediment and nutrient losses contributing to water quality impairment (Phillip et al. 2015).

Pasture renovation to improve forage availability creates an opportunity to consider new tall fescue biotypes, but the selection presents the following economic and sustainability issues: (1) wild-endophyte associated tall fescue provides excellent stand persistence, but periodic poor animal performance; (2) endophyte-free tall fescue produces excellent animal performance, but potentially has poor stand persistence; and (3) novel-endophyte fescue promises good stand persistence and excellent animal performance, but has substantially higher seed cost.

The fescue-endophyte association also has ecological impacts related to organic carbon (C) inputs affecting soil quality, nutrient cycling, surface and subsurface hydrologic ramifications, water quality, etc. In a meta-analysis on how grass-endophyte symbiosis affects subsurface soil processes and ecosystem services, Omacini et al. (2012) found significant changes in root biomass, root exudates, and mycorrhizal colonization of endophyte infected plants compared with

noninfected comparative pairs. No pattern in the response of soil fauna to endophyte infection was identified and no significant change was detected in the rate of processes like soil microbial respiration and litter decomposition. Effects on other processes like nutrient mineralization/cycling, nitrogen (N) fixation, contribution of bacteria and fungi to soil activity, etc., could not be assessed due to limited empirical data.

Fescue endophyte association's impact on cattle grazing behavior can result in alteration of surface soil properties, the distribution of soil organic C and total N, and creation of areas of soil compaction within pastures. Cattle in endophyte-infected tall fescue with high alkaloid production spend less time grazing and more time under shades and near water sources in environmentally stressful periods such as high temperatures (Stuedemann and Hoveland 1988). Given the choice, cattle show grazing preference of tall fescue free of endophyte or of low alkaloid. The true hydrologic responses of pastures under these myriad possible variabilities have not been well elucidated.

Our objective here is to present results from a long-term grazing study on the influence of three management factors: (1) fescue-endophyte associations, (2) fertilizer source, and (3) forage management on field-scale runoff nutrient quality. Extended wet

and dry periods during the study provided additional opportunity to assess impacts of variable weather on nutrient. Franzluebbers and Stuedemann (2006) and Franzluebbers et al. (2009, 2012a) previously presented results from this field-scale study on animal performance, plant stand, and soil quality aspects of managing wild-, novel-, and endophyte-free tall fescue as a function of fertilization and harvest management, while Endale et al. (2013) presented an analysis of treatment effects on runoff.

#### **Materials and Methods**

Site Description. The approximately 20 ha experimental site in the Southern Piedmont near Watkinsville, Georgia (33°53'07" N, 83°26'04" W), was transitioned for the research between 1998 and 2002 (Dawson Field; figure 1). For about 10 years before beginning the research, the field was maintained as naturalized herbaceous fallow with periodic mowing. Prior to 1988 the area was cropped to corn (Zea mays L.), cotton (Gossypium hirsutum L.), and soybeans (Glycine max) using conventional tillage for several decades by a local producer. As described in Endale et al. (2013), paddock attributes of surface area, area of slope class of 1% to 9% in increments of 1%, cumulative percentage area up to each slope class, area weighted slope (3.3% to 4.8%), length of each

of five orders of runoff flow paths (analogous to stream orders), and total flow path length (1,337 to 3,692 m) were determined based on analysis at 0.1 m cell resolution using global positioning and geographic information system. A survey in early- to mid-1980s established the soil series as predominantly (~80%) Cecil, a series that covers almost half of the Southern Piedmont, with 12% Pacolet and 8% Appling (all fine, kaolinitic, thermic Typic Kanhapludults) (unpublished data). Surface texture is sandy loam to sandy clay loam underlain by a clayey Bt horizon over a loamy saprolite C horizon. The soil is well drained, moderately permeable, and deep unless located at eroded landscape positions. Long-term (1937 to 2011; http://www. ncdc.noaa.gov; http://georgiaweather.net) mean annual rainfall was 1,240 mm with long-term monthly means varying from 79 mm in October to 136 mm in March. Despite the high mean annual rainfall, periods of drought are common (Endale et al. 2011). Mean annual temperature is 16.5°C.

**Experimental Design.** The primary study objectives focused on productivity and persistence of tall fescue-endophyte associations, animal performance, soil quality, and hydrology. Availability of land, animal resources, and long-term funding challenges led to design compromises that limited the study to two replications per treatment. Fourteen  $1.00\pm0.05$  ha grazing paddocks, large enough to allow multiple head of beef cattle to graze, were established and equipped for hydrologic monitoring and sampling. Topographic characteristics of paddocks and their effects on runoff was presented by Endale et al. (2013).

The experiment was laid out as a randomized block design with three levels of fescue-endophyte association (wild, novel, and free; producing high, low, and zero ergot alkaloids, respectively) and two sources of nutrients (BL and inorganic fertilizer) (six treatments; figure 1). Two paddocks were maintained for having (treatment no. 7) to compare with the corresponding grazed treatment with novel-endophyte fescue and inorganic fertilizer. Inorganic fertilizer and BL were applied as split applications in early fall and late winter (September/October and February/March). Broiler litter for the duration of the study came from several broiler houses managed similarly by a nearby poultry producer.

Grazing began in April of 2002 using weaned Angus heifers (2002 to 2007) and

steers (2008 to 2009) at a variable stocking rate of ~3 to 6 head ha-1. Stocking (other than the first year) was in September, and rate was adjusted every 28 days to maintain 1 to 3 Mg ha<sup>-1</sup> of available forage per paddock. Weather conditions influencing soil water and forage production dictated when cattle were stocked and removed. During periods when estimated forage biomass fell <1 Mg ha<sup>-1</sup> for most paddocks, cattle were moved to a nearby holding area of endophyte-free fescue and supplemented with hay until sufficient forage regrowth occurred (1 to 3 Mg ha<sup>-1</sup>) to allow restocking. Herd size across paddocks varied between 39 and 66 animals. Fescue was cut and baled in hayed paddocks two to three times a year whenever forage mass accumulated above 1 Mg ha<sup>-1</sup>. Paddocks were occasionally mowed in late spring to remove aboveground weed reproductive structures. The study concluded in April of 2010.

Data collected on animal grazing patterns and performance included grazing period (total of 15), start and finish dates of each grazing period, days per grazing period, and animal numbers per grazing period (tables 1 and 2). In each paddock, individual animal weights determined every 28 days were used to compute animal units. One animal unit, equivalent to 454 kg of live animal weight, provides a convenient normalized measure across different types of livestock (Kellogg et al. 2014). Animal units were used to estimate

wet manure mass and wet manure N and phosphorus (P) contents using coefficients from Kellogg et al. (2014). The given coefficients for pastured beef calves, heifers, cows, bulls, and stockers and grass-fed are 10.61 t of manure as excreted per animal unit per year as wet weight; 5.44 kg of N per wet tonne manure weight; and 2.26 kg of P per wet tonne manure weight. Nitrogen and P are considered in elemental forms and include those in urine. Number of runoff events during grazing, days with and without cattle before runoff, and animal units before runoff were compiled. Fertilizer management variables included days from N and P fertilization to a subsequent runoff event and amount of N and P fertilization immediately preceding a runoff event. (See table 3 for a complete list of variables.) The statistical methodology described below highlights how these data were used in the analyses.

Runoff, Sample Collection and Processing, and Analyses. Rainfall amounts for five-minute periods were measured near Paddock BW10 (figure 1) with a tipping bucket gauge (Model TR-525M, Texas Electronics, Dallas, Texas) and stored in a Campbell Scientific CR10X data logger (Logan, Utah). Surface runoff was measured at the lowest point (outlet) of each paddock using a combination of a 0.46 m H flume (Brakensiek et al. 1979) and a 17.24 kPa flow depth sensing transducer (Druck, New Fairfield, Connecticut) positioned in

**Table 1**Grazing period start and end dates, grazing days, and number of runoff events during each of 15 grazing periods.

Period #	Beginning date	Ending date	No. of days*	Runoff events
1	Apr. 11, 2002	May 30, 2002	50	1
2	Oct. 1, 2002	Dec. 10, 2002	71	3
3	Mar. 13, 2003	Sept. 25, 2003	197	11
4	Nov. 18, 2003	Jan. 29, 2004	73	1
5	Mar. 18, 2004	Sept. 30, 2004	197	6
6	Oct. 19, 2004	Jan. 11, 2005	85	1
7	Apr. 7, 2005	Oct. 21, 2005	198	4
8	Nov. 10, 2005	Jan. 5, 2006	57	1
9	Mar. 23, 2006	June 22, 2006	92	0
10	Oct. 5, 2006	Feb. 2, 2007	121	3
11	Apr. 3, 2007	June 14, 2007	73	0
12	Oct. 16, 2007	Dec. 11, 2007	57	0
13	Mar. 27, 2008	Aug. 14, 2008	141	0
14	Oct. 28, 2008	Jan. 7, 2009	72	2
15	Apr. 2, 2009	July 7, 2009	97	2

\*No. of days: mean  $\pm$  SE 105.4  $\pm$  13.7; median 85.0; total 1,581.

 Table 2

 Descriptive statistics for animal-related variables during 15 grazing periods from April of 2002 to July of 2009.

		Organic			Inorganic		
		Novel	Wild	Free	Novel	Wild	Free
		<b>1</b> and 9	4 and 10	5 and 12	6 and 11	2 and 8	3 and 7
Variable	Statistics*	1	2	3	4	5	6
Grazing animal numbers (GAN)	Mean	352.8	444.4	350.3	350.6	481.8	349.3
	SE	30.4	43.7	32.4	33.1	52.1	31.8
	Median	289.0	377.5	291.0	283.0	363.0	286.5
	Minimum	146.0	171.0	160.0	171.0	187.0	160.0
	Maximum	650.0	934.0	706.0	760.0	1217.0	760.0
	Total	10,585.0	13,331.0	10,510.0	10,517.0	14,454.0	10,479.0
Grazing animal unit (GAU)	Mean	226.3	267.3	225.9	224.3	281.6	224.0
	SE	24.8	31.0	27.2	25.7	34.8	25.3
	Median	165.6	189.3	155.7	160.7	194.5	157.4
	Minimum	69.2	77.4	79.4	95.6	105.6	80.5
	Maximum	496.5	658.1	531.3	524.6	787.3	509.7
	Total	6,789.1	8,018.5	6,778.1	6,728.8	8,449.0	6,720.7
Wet manure weight (Mg)	Mean	6.6	7.8	6.6	6.5	8.2	6.5
	SE	0.7	0.9	0.8	0.7	1.0	0.7
	Median	4.8	5.5	4.5	4.7	5.7	4.6
	Minimum	2.0	2.3	2.3	2.8	3.1	2.3
	Maximum	14.4	19.1	15.4	15.3	22.9	14.8
	Total	197.4	233.2	197.1	195.7	245.7	195.4
Wet manure elemental N weight (kg) (MAN)	Mean	39.5	46.6	39.4	39.1	49.1	39.1
	SE	4.3	5.4	4.7	4.5	6.1	4.4
	Median	28.9	33.0	27.2	28.0	33.9	27.5
	Minimum	12.1	13.5	13.9	16.7	18.4	14.0
	Maximum	86.6	114.8	92.7	91.5	137.4	88.9
	Total	1,184.6	1,399.0	1,182.6	1,174.0	1,474.2	1,172.6
Wet manure elemental P weight (kg) (MAP)	Mean	16.5	19.4	16.4	16.3	20.5	16.3
5, ,	SE	1.8	2.3	2.0	1.9	2.5	1.8
	Median	12.0	13.8	11.3	11.7	14.1	11.4
	Minimum	5.0	5.6	5.8	7.0	7.7	5.9
	Maximum	36.1	47.8	38.6	38.1	57.2	37.1
	Total	493.6	582.9	492.8	489.2	614.2	488.6

Note: Treatment no. 7 was the hayed only treatment with novel endophyte and inorganic fertilizer. Fertilizer = organic and inorganic. Fescue = novel, wild, and free. Paddocks = 1 through 12. Treatment no. = 1 through 6. SE = standard error.

the flume stilling well and connected to a data logger. Five-minute average flow depth and flow rate, based on the flume calibration curve (Bos 1978), were used to determine event runoff characteristic throughout the eight-year study period. Endale et al. (2013) performed Principal Component (PC) analysis using 23 variables describing rainfall, runoff, and landscape attributes (see site description above), and identified event rainfall amount, length of flow path order 1, cumulative percentage area up to slope class 2%, and area-weighted slope as the best variables to account for the effects of landscape on runoff variability.

Runoff samples were collected using a 2.5 cm diameter polyvinyl chloride (PVC) pipe securely attached below the outlet of each flume. The pipe had 1.6 mm diameter holes drilled at 2.5 cm spacing along its length. It was placed at a slight angle centered on the outflow path to continuously intercept and direct part of the runoff to a 15 cm diameter PVC pipe located to one side of the flume outlet. The 15 cm pipe was sealed at the bottom with a PVC cap, and for protection, housed inside a 20 cm diameter PVC pipe capped at the top and bottom. Runoff samples were removed from the field within 24 hours of a rainfall event. They were placed on ice in coolers and transported to the laboratory ~2.5 km away where they were stored at 4°C. Within 24 hours, a 50 mL subsample was filtered through a preweighed 0.45 micron cellulose-nitrate-acetate membrane. A second 50 mL unfiltered sample was also saved. Filtered and unfiltered subsamples were kept frozen until analyzed. Unfiltered samples were acidified with hydrogen chloride (HCl).

Filtered samples were analyzed on a Technicon Autoanalyzer II (SEAL Analytical, Mequon, Wisconsin) for dissolved ammonium-N (NH<sub>4</sub>-N) using the salicylate-hypochlorite method (Crooke and Simpson 1971) and nitrate-N (NO<sub>3</sub>-N) by the Griess-Ilosvay method (Keeney and

<sup>\*</sup>Statistics are based on 30 observations per treatment (2 paddocks and 15 grazing periods).

**Table 3**List of covariates and nutrient concentration and load abbreviations.

Covariate*	ID	Nutrient concentration and load	ID	Obs†
Amount nearest N-FERT to runoff	ANF	Ammonium-N concentration	AMC	731
Amount nearest P-FERT to runoff	APF	Ammonium-N load	AML	733
Days grazed before runoff‡	DGZ			
Days from runoff to nearest N-FERT	DNF	Nitrate-N concentration	NTC	731
Days from runoff to nearest P-FERT	DPF	Nitrate-N load	NTL	739
Days since last runoff	DSLR	Ortho-P concentration	OPC	721
Days with cattle before runoff	DWC	Ortho-P load	OPL	725
Days without cattle before runoff	DWOC	Total N concentration	TNC	266
Grazing animal unit	GAU	Total N load	TNL	266
Grazing during runoff	GCDR	Total P concentration	TPC	266
Precipitation amount (mm)	PAM	Total P load	TPL	267
Precipitation duration (h)	PDU			
Precipitation intensity	PIN			
Runoff date	ROD			
Runoff volume	ROV			

<sup>\*</sup>N-FERT and P-FERT signify nitrogen (N) and phosphorus (P) fertilization, respectively.

Nelson 1982) after reduction of NO, with a cadmium (Cd) column. Filtered samples were analyzed for soluble P (ortho-P) by the molybdate blue method (Murphy and Riley 1962). Total P (TP) and total N (TN) of unfiltered samples were determined colorimetrically as above following digestion by the alkaline persulfate oxidation technique (Franson 1998a,1998b). Data for TN and TP are available only from events in 2007 and later due to logistical issues associated with analysis of earlier samples. Nutrient loads were calculated by multiplying the nutrient concentration by the runoff volume for an event and correcting for differences in paddock areas (divided by the paddock area). Additional details about methods and materials are available in Endale et al. (2013), Franzluebbers et al. (2009, 2012a), and Franzluebbers and Stuedemann (2006).

Statistical Analysis. Over the eight-year period, 77 runoff events were recorded from one or more paddocks (Endale et al. 2013). Paddocks with zero runoff were excluded from the analyses. Due to limited runoff volume for some samples, analysis for the

complete suite of nutrients was not possible. The number of observations for each nutrient concentration and load is available in table 3. Statistical analyses of the data considered two cohorts. The first included all useable data. The second cohort consisted of only the observations in the upper quartile, representing those events with large concentrations or large loads. In selecting the second cohort for each nutrient, the upper quartile for each paddock was identified and combined with selections from the other paddocks for subsequent analysis.

Statistical analyses were conducted using SAS ver. 9.4 within SAS Enterprise Guide ver. 7.1 (SAS Inst. 2014). Like the approach taken in the analyses of treatment effects on water runoff (Endale et al. 2013), we identified a set of nontreatment variables that could affect runoff nutrient concentration and load (listed in table 3 under the covariate column heading) for a total of 15 variables. Principal component analysis of these variables was conducted using method PRIN in PROC FACTOR of SAS. Results of the varimax rotation (results not shown) identi-

fied five PCs with Eigen values >1. Variable loadings on the PCs related to grazing animal units, days since nutrient application, amount of nutrient applied immediately before runoff events, runoff volume, and precipitation characteristics. Scores estimated for the five PCs were used as covariates in the analysis of variance to evaluate treatment effects.

The SAS MIXED procedure was used for analysis of variance. Nutrient concentration and load data were transformed to natural logarithms +1 prior to analysis. To facilitate analysis of the unbalanced experimental factors, treatment combinations were coded 1 through 7 (table 2). Treatment and PC scores were considered fixed effects. Block (replicate) was considered a random effect. Runoff events over time were repeated effects with subjects identified as the individual paddocks. A spatial exponential covariance structure was used because runoff events occurred at unevenly spaced time intervals. Specific treatment effects were determined from a piori constructed contrasts using LSMESTIMATE statements. Least square treatment means and upper and lower 95% confidence limits were determined within the LSMEANS and LSMESTIMATE statements and were back transformed for presentation in data tables.

The repeated measures analysis provided an indication of the annualized loss of nutrients associated with management treatments. The long-term nature of the data set provided an indication of management influences on median losses of nutrients. Because of the inherent variability associated with rainfall events, field topography, and grazing animal behavior along with the limitation of having only two replications, we chose to consider differences significant at  $\alpha \leq 0.10$ .

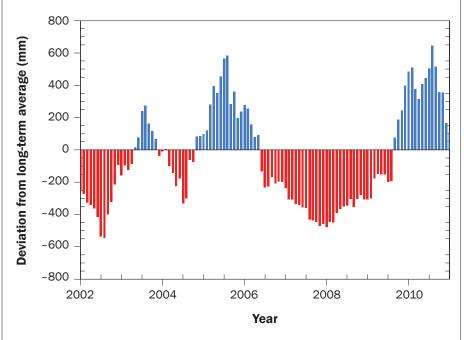
#### **Results and Discussion**

Rainfall Variability and Impact on Runoff and Runoff Nutrient Loss. Endale et al. (2011) previously presented an analysis of the impact of rainfall on runoff in this study. A summary of their results illustrating the prominent sustained drought and wet periods is presented here. The 12-month running average rainfall deviation from the long-term annual average is shown in figure 2. Comparing the 30 months of positive deviation versus the 63 months of negative deviation (April of 2002 to December of 2009), for all paddocks total runoff was 2.1 times greater (4,631 mm and 2,127 mm, respectively), total ortho-P load was ~1.7 times greater, total NH,-N load

<sup>†</sup>Obs is the number of observations in the analysis data set. Data for total concentration and load came from 2007 through the end of the project period only.

<sup>‡</sup>If cattle are present during runoff, then DGZ equals days from start of grazing to runoff. If cattle are absent during runoff, then it equals days grazed during the grazing period immediately preceding runoff.

Figure 2
Deviation of the 12 months running average rainfall during the study from the 74-year (1937 to 2010) annual average of 1,240 mm near Watkinsville, Georgia.



was about the same, and total  $NO_3^-$  load was  $\sim 2.9$  times lower (data not shown).

Individual monthly rainfall varied from below to above long-term monthly averages and are more directly related to runoff generation and nutrient losses. Supplementary tables S1 and S2 summarize runoff and runoff nutrient losses from May of 2002 (first month of runoff data) through December of 2009 for periods of monthly rainfall below average, average, and above average (52%, 21%, and 27.2% of the time, respectively). Monthly mean rainfall for above average and average periods was 3.1 and 1.6 times that of the below average period (183.5, 96.5, and 60.1 mm, respectively). Mean total runoff during above and average periods were 15.2 and 1.9 times that of the below average period (15.99, 2.02, and 1.06 mm ha<sup>-1</sup>, respectively). Across all paddocks, total nutrient loads were 3.4-fold (TP) to 14.6-fold (NH<sub>4</sub>-N) greater from the above average than the below average period. See Supplemental table S2 for more details.

Grazing Activity. Cattle grazed during 15 periods from April of 2002 to July of 2009 and grazing days per period varied from 50 to 198 (mean  $105.4 \pm 13.7$  SE; table 1). Runoff events occurred in 11 grazing periods, with 1 to 4 events per period except for period 3 in 2003 with 11 and period 5 in 2004 with 6 runoff events. Descriptive sta-

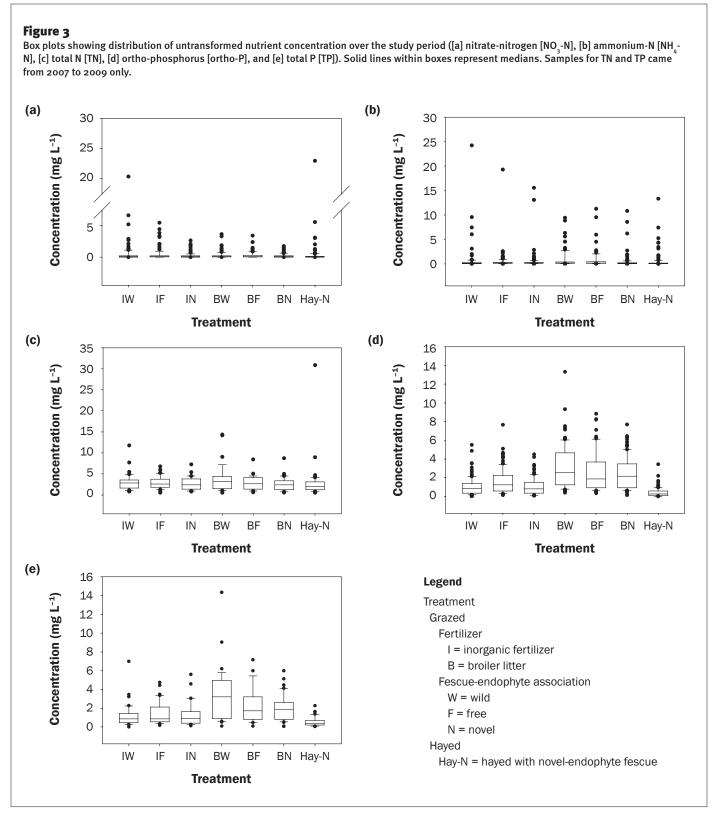
tistics for grazing animal number, grazing animal unit, estimated wet manure weight, and wet manure elemental N and P weights for the 15 grazing periods are given in table 2. Refer to figure 1 and table 2 for details.

Overall Assessment of Water Quality. In this section, we present numerical comparisons for concentrations and loads related to presence and absence of cattle and upper quartile versus rest of the data using the original untransformed data. Loads are given in units of kilogram per hectare here and in cited supplementary material, whereas in figures and for statistical comparisons and summaries (treatment comparisons, see further below) units of gram per hectare are used. Figure 3 shows the overall distribution of nutrient concentrations (untransformed data) from the seven treatments over the study period. Supplementary table S3 gives summary statistics for nutrient concentrations and loads based on untransformed data, parsed into periods with and without cattle as well as all periods. Mean concentrations (mg L<sup>-1</sup>) with cattle were 1.3- to 7.3-fold greater than those without cattle for NO3-N (0.29 to 1.03 versus 0.08 to 0.26); 1.5- to 2.3-fold greater for ortho-P (1.32 to 3.46 versus 0.72 to 2.57); 1.1- to 1.9-fold greater for TP (1.42 to 4.77 versus 1.13 to 3.06); and up to 1.8 times greater for TN (3.63 to 6.06 versus 2.48 to 3.34). For NH<sub>4</sub>-N, concentration was less from periods with cattle than without except for free fescue with inorganic fertilizer (4.2-fold greater) and wild fescue with BL fertilization (1.2-fold greater).

Total load (kg ha<sup>-1</sup>) (figure 4, shown as g ha<sup>-1</sup>) was greater with than without cattle for NO<sub>3</sub>-N (up to 5.2-fold; 0.24 to 4.43 versus 0.15 to 0.2.44) and for ortho-P (up to 2.1-fold; 5.12 to 16.36 versus 3.38 to 12.12). Total NH<sub>4</sub>-N load was up to 3.0-fold greater but only in BL treatments (~1.78 versus 0.78); in inorganic fertilizer treatments, load with cattle was ~0.27 that of without cattle. For TN and TP total load with cattle was ~0.03 that without cattle; a factor affecting this is that more of the data came from periods with no cattle (n = 22 to 36) than with cattle (n = 4 to 9). Refer to supplementary table S3 for additional details.

Comparisons of concentration and load between the upper quartile and those in the rest of the data showed the following (data not presented). In grazed treatments, upper quartile mean concentrations (mg L-1) were greater by 10- to 31-fold for NO<sub>3</sub>-N (0.60 to 2.08 versus <0.09); 13- to 28-fold for NH.-N (1.44 to 2.61 versus <0.15); 2.9- to 3.9-fold for ortho-P (2.28 to 6.25 versus <2.10); 2.2- to 3.0-fold for TN (4.70 to 7.61 versus < 2.60); and 3.0- to 4.3-fold for TP (2.65 to 6.86 versus <2.20). For haved treatments differences were 39.1-fold for NO<sub>2</sub>-N, 27.8-fold for NH<sub>4</sub>-N, 5.3-fold for ortho-P, and ~3.8-fold for TN and TP with concentrations <2.0. Total loads in grazed treatments (kg ha<sup>-1</sup>) in the upper quartile were greater by 4.7- to 10.8-fold for NO<sub>2</sub>-N for all but wild fescue with inorganic fertilization (0.43 to 2.66 versus <0.15); 3.6- to 12.1-fold for NH<sub>4</sub>-N (1.35 to 4.69 versus <0.60); 3.2- to 6.8-fold for ortho-P (8.56 to 22.30 versus <6.20); 2.0- to 3.0- fold for TN (4.56 to 9.76 versus <3.60); and 2.4- to 4.2-fold for TP (2.67 to 5.28 versus <1.50). For haved treatment, mean loads for upper quartile data were greater compared with means for the rest of the data up to 15-fold for ortho-P, TN, and TP, 56-fold for NH,-N and 177-fold for NO<sub>3</sub>-N with loads of 1.71 to 9.02 kg ha<sup>-1</sup> for the upper quartile and < 2.40 kg ha<sup>-1</sup> from the rest.

Supplementary tables S4 and S5 give breakdown of annual load estimates. Total loads (kg ha<sup>-1</sup>) across the study period were numerically greater from organic fertilizer (BL) treatments than inorganic treatments for ortho-P (2.0-fold; 69.8 versus 35.8), TP (1.3-



fold; 19.8 versus 15.3), and NH<sub>4</sub>-N (1.4-fold; 10.6 versus 7.6). In a reversal, total loads for NO<sub>3</sub>-N and TN were numerically greater from inorganic than organic fertilizer treat-

ments (3.5- and 1.4-fold, respectively; 11.2 versus 3.2 and 33.9 versus 23.6, respectively). Across all treatments and years total load (kg ha<sup>-1</sup>) was ~23 for inorganic N treatments, 68

for TN, 109 for ortho-P, and 37 for TP (TN and TP data from 2007 to 2009 only).

Correlation between ortho-P and TP concentrations per treatment was high ( $r^2 =$ 

Figure 4 Box plots showing distribution of untransformed nutrient load over the study period ([a] nitrate-nitrogen [NO\_-N], [b] ammonium-N [NH\_-N], [c] total N [TN], [d] ortho-phosphorus [ortho-P], and [e] total P [TP]). Solid lines within boxes represent medians. Samples for TN and TP came from 2007 to 2009 only. (a) (b) 7,000 2,500 6,000 2,000 5,000 Load (g ha $^{-1}$ ) Load (g ha<sup>-1</sup>) 1,500 4,000 3,000 1,000 2,000 500 1,000 0 0 IW IF IN BW BF BN Hay-N IŴ BW BF BN Hay-N IF **Treatment Treatment** (c) (d) 2,500 5,000 2,000 4.000 Load ( $g ha^{-1}$ ) 1,500 Load (g ha<sup>-1</sup>) 3,000 1,000 2,000 500 1.000 0 0 IW IF BW BN Hay-N IN BF BW BF BN Hay-N IW IF IN **Treatment Treatment** (e) 1,800 Legend 1.600 Treatment 1,400 Grazed 1.200 Load (g ha<sup>-1</sup>) Fertilizer 1,000 I = inorganic fertilizer 800 B = broiler litter 600 Fescue-endophyte association 400 W = wild200 F = free0 N = novelIW IF BN Hay-N IN BW BF Hayed Hay-N = hayed with novel-endophyte fescue **Treatment** 

0.89 to 0.98), indicating ortho-P was a major contributor to TP. Ortho-P was 98% of TP in grazed BL fertilized treatments (n=116; 97% in wild-associated, 99% in novel- and free-associated fescue), 86% of TP in grazed inorganic fertilized treatments (n=113; 84% in wild-associated, 89% in free-associated,

and 83% in novel-associated fescue), and 76% of TP in hayed management (n = 49).

Similarly, correlation between ortho-P and TP loads was also high ( $r^2 = 0.87$  to 0.97) with ortho-P load being a major constituent of TP load with some minor variation between treatments. As a percentage of TP load, ortho-P was 84% of TP in grazed pad-

docks with BL fertilization (n = 103; 73% in wild-associated, 83% in free-associated, and 99% in novel-associated fescue), 72% of TP in grazed paddocks with inorganic fertilization (n = 113; 74% in wild-associated, 69% in free-associated, and 73% in novel-associated fescue), and 69% of TP in hayed paddocks (n = 50).

Table 4

Mixed model analysis of all observations (complete data set) used for evaluating treatment and covariate (as Principal Component [PC]) effects on nutrient concentrations (mg L<sup>-1</sup>) and loads (g ha<sup>-1</sup>) in runoff from fescue paddocks at Watkinsville, Georgia. Analysis was conducted on natural log transformed values of concentration and load.

	Ammoni	um-N	Nitrate-N		Total N		Ortho-P		Total P	
Effect	F value	Prob F	F value	Prob F	F value	Prob F	F value	Prob F	F value	Prob F
Concentration										
Treatment*	37.7	<0.0001	37.09	< 0.0001	249.3	<0.0001	101.7	<0.0001	152.2	<0.0001
PC_1†	29.6	<0.0001	56.19	< 0.0001	55.9	<0.0001	121.5	<0.0001	99.2	<0.0001
PC_2	1.7	0.1985	0.44	0.5074	2.6	0.1054	83.7	<0.0001	8.6	0.0036
PC_3	16.8	<0.0001	3.12	0.0777	2.1	0.1472	12.0	0.0006	0.3	0.5714
PC_4	16.4	<0.0001	8.86	0.003	0.1	0.8286	2.5	0.1121	0.2	0.6595
PC_5	15.6	<0.0001	6.57	0.0106	1.5	0.2156	18.0	<0.0001	6.4	0.0124
Load										
Treatment*	79.3	<0.0001	56.88	<0.0001	316.3	<0.0001	89.0	<0.0001	87.5	<0.0001
PC_1†	4.0	0.0451	81.78	<0.0001	9.6	0.0022	1.5	0.2231	0.2	0.6749
PC_2	0.1	0.7688	18.16	< 0.0001	2.0	0.158	39.1	<0.0001	0.0	0.8881
PC_3	2.1	0.1482	0	0.9956	13.0	0.0004	2.1	0.1465	11.7	0.0007
PC_4	293.0	<0.0001	178.29	<0.0001	271.7	<0.0001	589.1	<0.0001	220.7	<0.0001
PC_5	35.0	<0.0001	0.06	0.8044	17.8	<0.0001	10.3	0.0014	28.4	<0.0001

Notes: N = nitrogen. P = phosphorus.

Water Quality by Treatment. Results are presented further below per cohort of data (full data set and data in the upper quartile). Here, we briefly discuss the covariates associated with the five PCs (data not shown), which collectively accounted for approximately 11% of the variance. Dominant loadings of variables were: PC\_1 DNF, DWOC, DPF, and GCDR (see table 3 for list of covariate abbreviations); PC\_2 DWC, GAU, and DGZ; PC 3 APF and ANF; PC\_4 ROV and PAM; and PC\_5 PIN and PDU. Dominant variables had loading values greater than 0.80 for the rotated factor pattern except GCDR and DGZ, which had loadings of -0.71 and 0.77, respectively. Variable loadings for the upper quartile data were essentially the same as those listed above with minor rearrangement of the order.

In the full data set, treatment differences were indicated for concentration and load for all nutrients (table 4). In addition, PC covariates differentially accounted for significant variance for all nutrients. Runoff nutrient concentration and load main effect means for fertilizer source (BL, inorganic) and fescue endophyte status (wild, novel, free) are presented in table 5 along with indications of significant differences. Concentrations of NH<sub>4</sub>-N and NO<sub>3</sub>-N were greater for inorganically fertilized paddocks, while ortho-P

and TP concentrations were greater where BL was the fertilizer source. Concentration of TN was not different due to fertilizer source. Runoff load was greater for TN, ortho-P, and TP for BL fertilized paddocks. This is not surprising since BL contains about the same amount of both N and P but forage grasses typically require three to four times more N than P (Gaskin et al. 2013). Application of BL to meet N requirements of forage grasses leads to accumulation of P and greater potential for P release in runoff, which has resulted in the requirement for use of P based recommendations in the United States to protect water quality (USDA NRCS 2012). Our observation of greater TN loss with BL most likely reflects organic N loss.

Differences in runoff concentrations of NH<sub>4</sub>-N, NO<sub>3</sub>-N, ortho-P, TN, and TP were also observed among fescue endophyte associations (table 5). In general, nutrient concentrations were lower from paddocks with novel-endophyte fescue compared with wild-endophyte fescue. Concentrations tended to be intermediate for the free-endophyte fescue. Results for runoff loads contrasted with those observed for concentration. Loads were greatest for paddocks with free-endophyte fescue compared with the novel-endophyte and wild-endophyte fescues. The result reflects the effects of

greater runoff volume from inorganic fertilization compared with BL fertilization as reported by Endale et al. (2013) being expressed to a greater degree in the inorganic × free fescue compared to remaining fertilizer × fescue interactions. Changes in soil properties were associated with cattle response to endophyte-fescue association (Franzluebbers et al. 2012b). They reported greater rates of decline of soil bulk density with novel- and wild-endophyte fescue than with endophyte-free fescue (-0.027 versus -0.18 Mg m<sup>-3</sup> y<sup>-1</sup>). Although stocking rates were frequently adjusted to promote similar forage consumption among paddocks, less forage was consumed in the wild-endophyte treatments, resulting in greater residual mass and potentially greater surface residue input during portions of the year (Franzluebbers et al. 2009). Eroded soils in the piedmont are susceptible to crusting due to limited amounts of organic matter at the soil surface (Bruce et al. 1995). Wild-endophyte treatments had greater ground cover at certain times, which would help reduce the negative impacts of precipitation on soil crusting and would promote infiltration (Daniel 2001).

Interaction level treatment means for runoff concentration and load are provided in table 6. Differences in runoff concentration between the grazed and haved inorganic fer-

<sup>\*</sup>Treatment refers to the six grazing and one having treatments. Bolded figures indicate statistical significance at ≤ 0.10.

<sup>†</sup>Greatest loadings for Principal Components were: PC\_1 = DNF, DWOC, DPF, and GCDR; PC\_2 = DWC, GAU, and DGZ; PC\_3 = APF and ANF; PC\_4 = ROV and PAM; and PC\_5 = PIN and PDU. Refer to table 3 for acronyms.

Table 5 Grazed treatment main effect means and 95% lower and upper confidence limits (LL and UL) from all observations for nutrient concentrations (mg L-1) and loads (g ha-1) in runoff from fescue paddocks at Watkinsville, Georgia. Values were back transformed from natural log least square means.

	Nutrie	ent																		
Treatment*	AM	LL	UL		NT	LL	UL		TN	LL	UL		OP	LL	UL		TP	LL	UL	
Concentration (m	g L <sup>-1</sup> )																			
BL	0.17	0.08	0.28	B†	0.14	0.07	0.22	В	2.02	1.82	3.71	Α	1.88	1.58	2.21	Α	2.03	1.50	2.66	Α
Inorganic	0.43	0.34	0.53	Α	0.29	0.22	0.35	Α	3.15	2.33	4.18	Α	1.16	0.94	1.40	В	1.08	0.82	1.37	В
Wild	0.37	0.28	0.46	а	0.24	0.18	0.30	а	2.52	1.83	3.38	а	1.55	1.30	1.84	а	1.69	1.47	1.93	а
Novel	0.22	0.15	0.30	b	0.16	0.11	0.22	b	2.39	1.73	3.22	b	1.33	1.09	1.59	b	1.35	1.13	1.59	b
Free	0.30	0.21	0.39	ab	0.24	0.17	0.30	а	3.96	2.96	5.21	ab	1.61	1.35	1.91	а	1.50	1.26	1.76	ab
Load (g ha <sup>-1</sup> )																				
BL	5.26	3.73	7.28	Α	2.65	1.82	3.71	Α	197.5	112.5	345.9	Α	67.35	41.51	108.90	Α	139.40	73.48	263.70	Α
Inorganic	6.92	5.13	9.22	Α	3.15	2.33	4.18	Α	45.4	30.6	67.2	В	24.63	14.61	41.09	В	18.68	11.14	30.90	В
Wild	5.38	3.95	7.20	b	2.52	1.83	3.38	b	74.9	57.7	97.2	b	31.54	18.69	52.76	b	41.22	26.42	64.01	b
Novel	5.43	3.98	7.27	b	2.39	1.73	3.22	b	102.8	76.9	137.3	а	38.82	23.02	65.02	b	54.01	34.48	84.29	ab
Free	7.56	5.62	10.05	а	3.96	2.96	5.21	а	112.7	84.1	151.0	а	56.24	33.98	92.67	а	62.28	39.83	97.06	а

Notes: AM = ammonium-nitrogen. NT = nitrate-N. TN = total N. OP = ortho-phosphorus. TP = total P.

Table 6 Interaction level treatment means and 95% lower and upper confidence limits (LL and UL) for all observations for nutrient concentrations (mg L-1) and loads (g ha-1) in runoff from fescue paddocks at Watkinsville, Georgia. Values were back transformed from natural log least square means.

	Nutrie	nt																	
Treatment*	AM	LL	UL	NT	LL	UL		TN	LL	UL		0P	LL	UL		TP	LL	UL	
Concentration (	mg L <sup>-1</sup> )																		
WLG	0.27	0.14	0.42	0.17	0.08	0.26		2.52	1.84	3.35		2.26	1.89	2.67		2.60	1.96	3.37	
NLG	0.07	-0.04	0.19	0.10	0.01	0.19		1.66	1.09	2.38		1.70	1.39	2.05		1.68	1.15	2.34	
FLG	0.19	0.05	0.36	0.17	0.06	0.29		1.95	1.28	2.81		1.74	1.39	2.14		1.89	1.27	2.67	
WFG	0.48	0.34	0.63	0.32	0.23	0.43		3.12	2.44	3.92		1.00	0.78	1.25		1.01	0.71	1.37	
NFG†	0.40	0.27	0.55 <b>a</b>	0.23	0.15	0.33	а	2.80	2.14	3.60	а	1.01	0.79	1.26	а	1.06	0.73	1.45	á
FFG	0.42	0.29	0.56	0.31	0.22	0.40		2.98	2.35	3.72		1.50	1.23	1.79		1.17	0.85	1.54	
NFH	0.35	0.23	0.48 <b>a</b>	0.25	0.17	0.34	а	2.45	1.92	3.08	а	0.47	0.32	0.65	b	0.40	0.21	0.63	k
Load (g ha <sup>-1</sup> )																			
WLG	4.77	3.20	6.92	2.15	1.34	3.23		148.3	83.2	263.7		55.8	34.0	91.0		110.2	57.8	209.5	
NLG	4.65	3.09	6.81	2.38	1.50	3.58		250.2	130.6	478.5		76.5	46.7	125.0		170.9	83.2	349.8	
FLG	6.56	4.23	9.93	3.57	2.22	5.51		210.7	104.9	422.3		72.4	43.0	121.7		146.0	68.0	312.0	
WFG	6.05	4.20	8.56	2.94	1.97	4.22		37.6	22.9	61.5		17.7	10.6	29.1		15.0	8.2	26.9	
NFG	6.31	4.41	8.89 <b>a</b>	2.40	1.57	3.50	а	41.9	24.7	70.5	а	19.4	11.7	31.9	а	16.6	8.9	30.3	á
FFG	8.69	6.24	11.98	4.37	3.11	6.02		60.1	37.5	95.9		43.6	26.7	70.7		26.2	14.9	45.6	
NFH	4.62	3.22	6.50 <b>b</b>	2.85	1.96	4.00	а	34.2	21.5	54.0	а	7.5	4.3	12.6	b	6.7	3.5	12.0	ı

<sup>\*</sup>Three letter treatment designations represent fescue, fertilizer, and forage management, respectively. Fescue = wild (W), novel (N), and free (F). Fertilizer = broiler litter (L) and inorganic fertilizer (F). Forage management = grazed (G) and hayed (H).

tilized novel-endophyte fescue treatments were observed for ortho-P and TP concentrations and load, and for NH<sub>4</sub>-N load. Removal of nutrients in baled hay and lack

of animal manure on the soil surface most likely contributed to reduced P losses from the hayed paddocks.

Results from the mixed models analysis of the upper quartile of data are presented in table 7. As for the full data set, treatment effects were detected for runoff concentra-

<sup>\*</sup>Treatments are organic (broiler litter [BL]) and inorganic fertilizer and three fescue-endophyte associations.

<sup>†</sup>Letters indicate differences between fertilizer source (capital letters) or among fescue type (lower case letters) means at p < 0.10 as determined from contrast statements.

<sup>†</sup>Bolded cells are for the comparison of grazed versus hayed paddocks under novel-endophyte fescue with inorganic fertilization. Differences indicated at p < 0.10.

**Table 7**Mixed model analysis on upper quartile cohort of observations evaluating treatment and covariate (as Principal Component [PC]) effects on nutrient concentrations (mg L<sup>-1</sup>) and loads (g ha<sup>-1</sup>) in runoff from fescue paddocks at Watkinsville, Georgia. Analysis was conducted on natural log transformed values of concentration and load.

	Ammoni	um-N	Nitrate-N	I	Total N		Ortho-P		Total P	
Effect	F value	Prob F	F value	Prob F	F value	Prob F	F value	Prob F	F value	Prob F
Concentration										
Treatment*	22.6	<0.0001	20.1	<0.0001	79.5	<0.0001	418.6	<0.0001	171.9	<0.0001
PC_1	4.3	0.0396	1.4	0.245	3.6	0.0613	0.0	0.9683	0.3	0.5875
PC_2	5.3	0.0224	0.2	0.6887	1.6	0.218	0.8	0.3824	11.4	0.0012
PC_3	2.8	0.0984	0.4	0.5227	4.8	0.0312	0.1	0.8222	8.5	0.0047
PC_4	7.5	0.0068	0.5	0.4655	0.0	0.8603	0.1	0.718	2.0	0.1642
PC_5	3.6	0.0604	0.1	0.7746	13.2	0.0005	1.4	0.2451	2.6	0.1133
Load										
Treatment*	111.7	<0.0001	186.6	<0.0001	334.7	<0.0001	206.8	<0.0001	134.4	<0.0001
PC_1	8.0	0.0052	0.1	0.7061	10.5	0.0019	4.6	0.0341	47.4	<0.0001
PC_2	15.0	0.0002	1.5	0.2216	4.4	0.0396	13.8	0.0003	5.9	0.018
PC_3	5.5	0.0204	1.2	0.2834	2.5	0.1174	7.0	0.0089	12.0	0.0009
PC_4	3.5	0.0618	1.6	0.2106	7.2	0.009	0.3	0.6059	0.1	0.7844
PC_5	4.5	0.0358	26.7	<0.0001	83.9	<0.0001	55.6	<0.0001	63.2	<0.0001

Notes: N = nitrogen. P = phosphorus.

**Table 8**Grazed treatment main effect means and 95% lower and upper confidence limits (LL and UL) for upper quartile cohort of observations nutrient concentrations (mg L $^{-1}$ ) and loads (g ha $^{-1}$ ) in runoff from fescue paddocks at Watkinsville, Georgia. Values were back transformed from natural log least square means.

	Nutri	Nutrient																	
Treatment*	AM	LL	UL		NT	LL	UL		TN	LL	UL	OP	LL	UL		TP	LL	UL	
Concentration (mg	g L <sup>-1</sup> )																		
BL	0.86	0.47	1.35	A†	0.62	0.34	0.96	Α	5.27	3.49	7.75 A	5.01	4.30	5.80	Α	3.92	2.93	5.16	Α
Inorganic	1.44	1.04	1.93	Α	1.07	0.79	1.39	Α	4.81	3.57	6.38 A	2.47	2.16	2.81	Α	2.96	2.39	3.63	Α
Wild	1.49	1.10	1.96	а	0.95	0.69	1.25	а	6.10	4.78	7.70 a	3.54	3.20	3.91	b	3.90	3.37	4.51	а
Novel	0.83	0.55	1.17	b	0.63	0.42	0.86	b	4.45	3.41	5.74 b	3.15	2.84	3.50	С	2.98	2.50	3.53	b
Free	1.13	0.77	1.56	ab	0.93	0.66	1.25	а	4.71	3.62	6.06 b	4.07	3.66	4.51	а	3.43	2.90	4.02	ab
Load (g ha <sup>-1</sup> )																			
BL	27.1	16.3	44.5	В†	24.9	15.9	38.7	Α	1,259	720	2,202 A	589	376	923	Α	470	192	1,145	Α
Inorganic	53.2	35.0	80.6	Α	24.1	17.2	33.5	Α	448	316	636 E	245	152	394	В	284	154	522	Α
Wild	40.4	27.0	60.3	а	22.8	16.4	31.5	ab	690	592	804 b	329	202	536	b	376	228	618	ab
Novel	28.7	19.1	42.9	b	19.6	14.2	26.8	b	731	607	882 a	b 337	205	554	b	316	194	515	b
Free	47.8	31.6	71.9	а	33.2	23.7	46.4	а	857	706	1,041 a	503	312	811	а	417	256	678	а

Notes: AM = ammonium-nitrogen. NT = nitrate-N. TN = total N. OP = ortho-phosphorus. TP = total P.

tion and load of NH<sub>4</sub>-N, NO<sub>3</sub>-N, ortho-P, TP, and TN. Results of contrasts evaluating differences in nutrient concentrations and loads due to fertilizer source or fescue

endophyte association for the upper quartile cohort are presented in table 8.

Runoff nutrient concentration response within the upper quartile data were similar for the two fertilizer sources (table 8). However, fertilizer source influences on loads were evident for  $NH_4$ -N (inorganic > BL) and for TN and ortho-P (BL > inorganic). The effect of the fescue endophyte

<sup>\*</sup>Treatment refers to the six grazing and one haying treatments. Bolded figures indicate statistical significance at ≤ 0.10. Greatest loadings for Principal Components were: PC\_1 = DNF, DWOC, DPF, and GCDR; PC\_2 = DZG, DWC, and GAU; PC\_3 = PDU and PIN; PC\_4 = APF and ANF; and PC\_5 = ROV, PAM, and DSLR. Note slight difference with those in table 3. Refer to table 3 for acronyms.

<sup>\*</sup>Treatments are organic (broiler litter [BL]) and inorganic fertilizer and three fescue-endophyte associations.

<sup>†</sup>Letters indicate differences between fertilizer source (capital letters) or among fescue type (lower case letters) means at p < 0.10 as determined from contrast statements.

**Table 9**Treatment level means and 95% lower and upper confidence limits (LL and UL) for upper quartile cohort of observations for nutrient concentrations (mg L<sup>-1</sup>) and loads (g ha<sup>-1</sup>) in runoff from fescue paddocks at Watkinsville, Georgia. Values were back transformed from log least square means.

	Nutrie	ent																		
Treatment*	AM	LL	UL		NT	LL	UL		TN	LL	UL		0P	LL	UL		TP	LL	UL	
Concentration (	mg L <sup>-1</sup> )																			
WLG	1.29	0.72	2.06		0.69	0.34	1.14		7.25	4.93	10.47		5.64	4.79	6.60		5.30	3.95	7.03	
NLG	0.46	0.09	0.95		0.45	0.14	0.84		4.14	2.52	6.50		4.36	3.62	5.21		3.09	2.10	4.41	
FLG	0.94	0.36	1.77		0.73	0.30	1.30		4.85	2.86	7.86		5.13	4.20	6.23		3.64	2.51	5.12	
WFG	1.72	1.07	2.57		1.25	0.80	1.80		5.10	3.61	7.08		2.11	1.75	2.51		2.82	2.10	3.69	
NFG†	1.31	0.77	2.02	а	0.82	0.47	1.26	а	4.79	3.33	6.75	а	2.22	1.85	2.64	а	2.88	2.14	3.80	а
FFG	1.33	0.82	2.00		1.16	0.76	1.66		4.57	3.23	6.33		3.18	2.73	3.70		3.22	2.50	4.10	
NFH	1.19	0.71	1.81	а	0.94	0.58	1.37	а	4.62	3.25	6.43	а	0.97	0.76	1.21	b	1.28	0.88	1.76	b
Load (g ha <sup>-1</sup> )																				
WLG	24.5	14.0	42.6		16.6	9.5	28.5		1,114	673	1,842		494	314	777		516	232	1,147	
NLG	20.8	11.6	36.6		23.1	13.2	40.1		1,312	711	2,422		594	379	931		404	153	1,065	
FLG	39.1	19.9	75.9		40.5	20.6	78.5		1,395	736	2,643		711	443	1,141		505	185	1,374	
WFG	66.1	39.2	111.1		31.2	18.8	51.3		427	293	623		219	140	344		273	145	516	
NFG	39.4	23.5	65.6	а	16.5	10.0	26.9	а	407	273	608	а	192	122	300	а	247	127	481	а
FFG	58.4	35.9	94.6		27.2	17.1	43.0		527	361	768		356	227	559		344	182	650	
NFH	37.5	23.0	60.7	а	23.3	14.6	36.7	a	378	258	555	а	74	47	117	b	116	61	218	b

<sup>\*</sup>Three letter treatment designations represent fescue, fertilizer, and forage management, respectively: Fescue = wild (W), novel (N), and free (F). Fertilizer = broiler litter (L) and inorganic fertilizer (F). Forage management = grazed (G) and hayed (H).

association varied with nutrient, and no clear indication of trend is apparent.

Interaction level treatment means for runoff concentration and load for the upper quartile of data are provided in table 9. Differences in runoff concentration between the grazed and hayed inorganic fertilized novel-endophyte fescue treatments were observed for ortho-P and TP concentrations and load. Removal of nutrients in baled hay and lack of animal manure on the soil surface most likely contributed to reduced P losses from the hayed paddocks. Losses of NH<sub>4</sub> and NO<sub>3</sub><sup>-</sup> although not desirable were relatively low (over the study period, ~2.8 kg ha<sup>-1</sup> y<sup>-1</sup> for both for all observations; supplemental table S5).

The results above reflect the complexities of interactions of the various factors that determine temporal and spatial nutrient loss patterns from pastures. Five PCs encompassing several cattle, hydrologic, and management variables were needed as covariates to isolate treatment effects. Endale et al. (2013) showed how one of the variables, runoff, encompassed in PC\_4, is itself affected by other factors. Through PC analysis using 23 variables, Endale et al. (2013) identified event rainfall amount, flow path order #1, cumulative percentage area up

to slope class 2, and area-weighted slope as critical variables to account for effects of landscape on runoff variability at this site.

Endale et al. (2017) presented a summary of results on ortho-P and TP concentrations and loads from four other studies alongside results from their 1999 to 2009 study at a 7.7 ha rotationally grazed pasture 1.5 km northeast of the present study. Results were similar from all five studies. For example, concentration of ortho-P (mg L<sup>-1</sup>) ranged 0.6 to 24.2 in manure applied fields and 0.6 to 3.8 in inorganically fertilized fields (Edwards et al. 1996); 4.6 to 14.2 (Nash et al. 2000); and 0.3 to 7.5 (Endale et al. 2017), which are similar to ranges observed in the current study.

Endale et al. (2017) showed that a few large storms accounted for most of the total P loss. Further, runoff events occurring within 24 hours of fertilization resulted in greatest runoff concentrations, as would be expected. Over the 10 years of data, the six largest P load events (probability of exceedance <15%) accounted for 53% of the total P loss from all 43 monitored events and coincided with cattle presence in the watershed. As shown in the current study (above), total nutrient loads from data in the upper quartile

were several fold greater compared with the rest of the data.

Loss of TP in the current study was estimated as being 4.0% of the total P applied from inorganic, BL, and manure sources. Total P lost in runoff was estimated as 147 kg ha<sup>-1</sup> with an assumption of ortho-P being 75% of total P (had ortho-P data for whole period whereas TP data came from 2007 through 2009). Total P applied from inorganic (18-9-18) and organic (BL) sources was estimated at 466 k ha<sup>-1</sup> assuming 1.0 kg P per 80.0 kg BL (unpublished data). Estimated P in cattle manure during the 15 grazing periods was 3,227 kg ha<sup>-1</sup>.

## **Summary and Conclusions**

Runoff nutrient concentrations and loads of N and P species from tall fescue pastures in the Southern Piedmont were evaluated over eight years for three management variables: (1) tall fescue-endophyte association (wild, novel, and free); (2) nutrient source (BL and inorganic fertilizer); and (3) harvest management (grazing and haying). The following points identify significant results and conclusions:

 Tall fescue with novel-endophyte association had lower or equal runoff concentrations and loads for inorganic

<sup>†</sup>Bolded cells are for the comparison of grazed versus hayed paddocks under novel-endophyte fescue with inorganic fertilization. Differences indicated at p < 0.10.

N, ortho-P, and TP compared to wild- or free-endophyte fescue associations. Based on our results and previous companion results, novel-endophyte fescue (1) poses no additional risk of runoff compared to other endophyte fescue paddocks, and (2) has improved average daily gain by cattle and stand persistence, therefore its adoption in the southeastern United States, regardless of fertilization source, is encouraged.

- Weather variability plays a significant role in nutrient losses from grazed systems in the region. Mean total runoff from all paddocks were 15.2- and 1.9-fold greater during above average (27.2%) and average (20.6%) compared to below average (52.2%) monthly rainfall periods. Across all paddocks, nutrient loads were 3.4-fold (TP) to 14.6-fold (NH<sub>4</sub>-N) greater from above average compared to below average monthly rainfall periods. Modelers, land managers, and policy makers can use these data to better assess impacts of variable weather, field-scale pasture attributes, and management on water quantity and quality.
- Use of PC that encompass temporal and spatial factors in our analysis of variance allowed us to isolate true treatment effects on nutrient loss patterns from pastures. A similar approach was used in a previous companion report on runoff variability. Covariance analysis with the PCs proved to be a valuable tool. These analyses reflect on the complexities of interactions of the various factors that determine temporal and spatial nutrient loss patterns from pastures.
- Most nutrient losses occur from few elevated levels of pollutant fluxes the result of the convergence of hydrologic events favoring pollutant transport along with forage and fertilizer management variables. Pollutant loads in the upper quartile were several fold greater than those from the rest of the data. The presence of cattle increased ortho-P load up to three-fold compared with periods with no cattle, for example.
- Maintaining forage at a threshold to avoid overgrazing is a management strategy for improved nutrient cycling as evidenced by loss of only 4% of the applied inorganic P and from cattle manure.

#### **Supplemental Material**

Supplemental material for this manuscript is provided that adds and describes five additional data tables and one fig-

ure. https://www.ars.usda.gov/southeast-area/tifton-ga/southeast-watershed-research/research/documentation/supplementary-information/.

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