



Vasoconstrictive responses of the testicular and caudal arteries in bulls exposed to ergot alkaloids from tall fescue¹

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ABSTRACT

Color Doppler ultrasonography was used to evaluate vasoconstrictive responses of the testicular artery in yearling bulls to ergot alkaloids (*Neotyphodium coenophialum*) produced by a fungal endophyte that infects tall fescue [*Lolium arundinaceum* (Schreb.) Darbysh.]. Alkaloid-induced constriction of the testicular artery could disrupt thermoregulation. Luminal areas of the testicular artery were monitored and analyzed for 2 experiments that were designed to evaluate fertility responses of bulls to toxic endophyte-infected tall fescue. Experiment 1 (pen experiment) compared diets containing toxic endophyte-infected or endophyte-free tall fescue seed, and Exp. 2 (grazing experiment) compared grazing diets consisting of toxic endophyte-

infected and nontoxic endophyte-infected tall-fescue pasture. In Exp. 1 ultrasound images were acquired on 3 dates during the 126-d of feeding and in Exp. 2 on 4 dates during the 155-d of grazing. Luminal area of the caudal artery also was monitored as an indicator of alkaloid-induced vasoconstriction. Caudal and testicular arteries for the toxic treatment responded similarly between experiments. Across imaging dates for Exp. 1, caudal artery lumens for the toxic diet averaged 42% less ($P < 0.01$) area than the nontoxic diet, and testicular arteries for the toxic diet averaged 40% less ($P < 0.05$) area than the nontoxic diet. For Exp. 2, there were interactions ($P < 0.05$) between treatment and imaging date on luminal areas of both arteries. Differences between treatments were not detected until the last 2 image dates, with luminal areas of caudal and testicular arteries on the last date for the toxic tall fescue being 46 and 41%, respectively, less than the nontoxic counterpart. Results indicated that ergot alkaloids can induce constriction of blood flow to the testes that may adversely affect bull fertility.

Key words: bull fertility, ergot alkaloids, fescue toxicosis, tall fescue, vasoconstriction

INTRODUCTION

Tall fescue [*Lolium arundinaceum* (Schreb.) Darbysh.] is a persistent and productive cool-season perennial grass that is the most predominant grass used for pasture in the eastern United States. Unfortunately, ergot alkaloids produced by an endophyte (*Neotyphodium coenophialum*) that infects most tall-fescue plants can cause a toxicosis in cattle that reduces growth and thriftiness (Schmidt and Osborn, 1993) and reproductive performance (Porter and Thompson, 1992). From a review, Paterson et al. (1995) reported pregnancy rates and weaning weights on endophyte-infected tall fescue were reduced an average of 32 and 14%, respectively, as compared with animals on endophyte-free tall-fescue pastures.

Limited research has shown that bulls exposed to endophyte-infected tall fescue have elevated scrotal temperature (Jones et al., 2004), smaller scrotal circumference (Jones et al., 2004; Stowe et al., 2013), and lower percentages of motile and progressive sperm with high ambient temperatures (Looper et al., 2009). Although reports have been inconsistent in detection of which bull fertility char-

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acteristics were affected by ergot alkaloids, the extent of an adverse effect on these characteristics likely depends on genetics, environment, and amount of bioaccumulation of ergot alkaloids in the vasculature (Pratt et al., 2015).

Ergot alkaloids bind biogenic amine receptors in the vasculature (Oliver, 2005; Strickland et al., 2011) to constrict blood flow to peripheral tissues (Rhodes et al., 1991; Aiken et al., 2007; Klotz et al., 2007) and reduce the ability of the animal to thermoregulate body temperature. Thermoregulation of the testes is critical to maintaining fertility in bulls (Kastelic et al., 1997; Brito et al., 2004) and constriction of blood flow through the testicular artery could affect production of morphologically sound and viable sperm.

Ergot alkaloid-induced vasoconstriction of the testicular artery in bulls has not been documented. Therefore, an experiment was conducted using color Doppler ultrasonography to compare cross-sectional areas of the testicular artery between bulls fed diets with or without toxic endophyte-infected fescue seed in a feeding experiment (Exp. 1), and also between bulls grazing either toxic or nontoxic fescue pastures (Exp. 2). The caudal artery also was measured as a marker of alkaloid-induced vasoconstriction because it has been documented to be sensitive to ergot alkaloids (Aiken et al., 2007, 2009b).

MATERIALS AND METHODS

Animals and Treatments

Vasoconstrictive responses were measured for 2 groups of bulls between 13 and 16 mo of age that were used in separate experiments with objectives to compare fertility traits and semen characteristics between bulls consuming diets with or without ergot alkaloids. A feeding experiment fed either toxic or nontoxic seed as the treatment (Exp. 2), and a grazing experiment was conducted to make comparisons between bulls grazing either toxic or nontoxic endophyte-

infected tall fescue pastures (Exp. 2). Both experiments were conducted at the Simpson Research and Education Center in Pendleton, South Carolina. All animal research followed procedures approved by the Clemson University Institutional Animal Care and Use Committee.

Feeding Experiment (Exp. 1)

Seven Angus and 6 Hereford Bulls were stratified by breed, BW, and BCS for random assignment to feeding treatments that consisted of concentrate containing either toxic (E+, $n = 6$) or nontoxic (E-, $n = 7$) seed. Quantity of seed in the diet was similar between the 2 treatments and set by the quantity of toxic seed needed to provide a diet concentration of 0.8 μg of ergovaline and ergovalanine per gram of DM. Seed was assayed for ergovaline and ergovalanine using HPLC with fluorescence detection following procedures of Yates and Powell (1988) that were modified as described by Carter et al. (2010). A 2-wk diet adjustment period was done by feeding the concentrate with E- seed to all bulls. A 126-d test period (April 6 to August 10, 2011) with ad libitum feeding followed the adjustment period. Details of the experimental design and procedures are described by Stowe et al. (2013). Following the 126-d test period, 3 bulls on the toxic treatment were switched to the nontoxic diet, and 3 bulls remained on the nontoxic diet until October 19, 2011, to determine whether there was recovery from ergot alkaloid-induced vasoconstriction.

Grazing Experiment (Exp. 2)

Angus bulls were stratified by BW and visual BCS for allocation to single pastures of Kentucky 31 toxic endophyte-infected ($n = 11$) and MaxQ II [Texoma (Pennington Seed Inc., Madison, GA) infected with the nontoxic endophyte, AR584; $n = 10$] tall-fescue pastures. Although ergot alkaloid concentrations in grazed forage were not quantified, serum

prolactin concentrations were used as indicators of fescue toxicosis (Strickland et al., 1993). Blood samples were collected from the caudal arteries on d 35, 84, 114, and 140 for assaying serum prolactin following procedures of Bernard et al. (1993). Pastures were grazed for 155 d from April to August 2012. Experimental design and procedures are described by Pratt et al. (2015).

Doppler Ultrasound Imaging

Color Doppler ultrasound images of the cross-sections of the testicular and carotid arteries were collected on 3 dates for Exp. 1 (May 26, July 28, October 19) and 4 dates for Exp. 2 (May 10, June 28, August 16, and October 10). Baseline measures were not collected because logistical limitations of the sonographers caused wide time periods between imaging dates, and baselines would have been taken during cooler ambient temperatures that could have confounded comparisons between baselines and those when the bulls were on treatment diets. Images were collected using a Classic Medical TeraVet 3000 Ultrasound Unit (Classic Universal Ultrasound, Tequesta, FL) with a 12L5-VET (12 MHz) linear array transducer. Cross-sections of the medial caudal artery were imaged with a ventral, transverse orientation of the transducer on the tail at the fourth coccygeal (Cd4) vertebrae (Aiken et al., 2007, 2009b), and cross-sections of the testicular artery were imaged with a caudal, transverse orientation on the scrotum approximately 2 cm above the dorsal testes. Five images were collected for each artery using a frequency of 5.0 MHz and a pulse repetitive frequency that ranged between 3.0 to 4.5 kHz. Scan depth was set at 3 cm for both arteries. Following freezing of an individual scan, frames stored in the cine memory of the unit were searched to store the image exhibiting the maximum flow signal and assumed to be at peak systolic phase. The flow signal was traced to estimate luminal area (Aiken et al., 2009a).

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