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Adam F. Summers
University of Nebraska-Lincoln

Robert A. Cushman
University of Nebraska-Lincoln, Bob.Cushman@ars.usda.gov

Stetson P. Weber
University of Nebraska-Lincoln

Karl V. Moline
University of Nebraska-Lincoln, kmoline1@unl.edu

Jeffrey W. Bergman
University of Nebraska-Lincoln, jbergman1@unl.edu

See next page for additional authors

Authors
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Andrea S. Cupp[1]

Summary

Developing heifers were offered either a modified distillers (MOD), distillers based (DDG), or corn gluten feed based (CGF) supplement while grazing pastures during development. Prior to breeding, antral follicle count (AFC), uterine horn diameter (UHD), ovarian area, and reproductive tract score (RTS) were determined via rectal ultrasonography to examine the effect of protein supplement on heifer reproductive characteristics. Heifers developed on MOD diets had greater RTS, ovarian area, and total AFC compared to DDG and CGF heifers. Small and medium follicle counts had a positive correlation with total AFC. Heifers developed on DDG and CGF had greater overall pregnancy rates compared to MOD heifers. We also conclude that there is a positive relationship between AFC and small and medium follicle counts.

Introduction

Producer profitability is related to cow longevity, with failure to become pregnant a primary reason why cows are removed from the herd (Cushman et al., *Journal of Animal Science.* 2009 87: 1971-1980). Many producers provide protein supplementation to heifers developed on dormant winter range or pasture to improve reproductive success. Previous research indicates developing heifers on dried distillers grains does not reduce reproductive success. However, reproductive tract characteristics were not measured (2007 *Nebraska Beef Cattle Report*, pp. 5-6). Measures such as antral follicle count (AFC), reproductive tract score (RTS), and uterine horn diameter (UHD) have shown to be effective prediction tools for fertility. Cushman et al. (*Journal of Animal Science,* 2009, 87: 1971 - 1980) reported increased pregnancy rates in heifers classified as high AFC compared to low. The objective of the current study was to determine if protein supplementation during development and AFCs influence heifer reproductive characteristics and success.

Procedure

Heifers from two herds at the University of Nebraska–Lincoln Agricultural Research and Development Center were used. Heifers (Angus and Angus x Simmental hybrids) from the teaching herd (n = 56) were fed 3.5 lb/day (32% CP, DM) of a modified dried distillers grain (MOD) supplement from weaning (mid September) through May. MARC III (1/4 Angus, 1/4 Hereford, 1/4 Red Poll, 1/4 Pinzgauer) x Red Angus heifers from the physiology herd (n = 173) were randomly assigned to 1 of 2 groups and fed supplements similar to that reported by Martin et al. (2007 *Nebraska Beef Cattle Report,* pp. 5-6). Heifers received either a dried distillers grain based (DDG) or corn gluten feed based supplement (CGF) offered at 0.59% (27% CP, DM) and 0.78% BW (20% CP, DM), respectively, from mid-November through May. Supplements fed to the physiology herd heifers (DDG and CGF) were formulated to be isocaloric but differed in rumen undegradable protein. All heifers were fed ad libitum meadow hay through winter while grazing dormant pasture.

Prior to breeding, heifers underwent transrectal ultrasonography. A single technician scanned each ovary using an Aloka-500 linear array transrectal probe (7.5-MHZ transducer, Aloka Ultrasound, Wallingford, Conn.) and counted small (3-5 mm), medium (6-10 mm), and large (> 10 mm) follicles to determine AFC. Uterine horn diameter, presence of CL, and ovarian length and height were also determined. Each heifer received a RTS based on the methods reported by Martin et al. (*Journal of Animal Science,* 1992, 70: 4006-40017) as described in Table 1.

Estrus was synchronized with two injections of prostaglandin $F_{2\alpha}$ administered 14 days apart. Estrous detection was performed 5 days following the second injection. Heifers observed in estrous were artificially inseminated approximately 12 hours after initial estrous detection. Approximately 10 days after estrous detection was performed, heifers were placed with fertile bulls for 45 days. Conception rates for both AI and total pregnancy rates were performed via rectal palpation approximately 45 days following AI and bull removal, respectively.

Statistical analysis was performed using the MIXED and GLIMMIX procedures of SAS (SAS Inst., Inc., Cary, N.C.) with a $P \leq 0.05$ considered significant. The model included heifer treatment as a classification effect, total AFC as a covariate, and year as a random variable. Initial analysis included breed; however, it was not significant and was removed from the model.

Results

Heifer performance data are reported in Table 2. Heifers fed MOD supplement had greater ($P < 0.05$) RTS, total AFC, larger ovaries, and a greater proportion of heifers with a CL present when compared to both CGF and DDG supplemented heifers.

Table 1. Explanation of reproductive tract scores[1].

RTS	Uterine Horns	Approximate Size of Ovaries			Ovarian Structures
		Length, mm	Height, mm	Width, mm	
1	Immature, < 20 mm in diameter, no tone	15	10	8	No palpable follicles
2	20 to 25 mm in diameter, no tone	18	12	10	8 mm follicles
3	25 to 30 mm in diameter, slight tone	22	15	10	8-10 mm follicles
4	30 mm in diameter, good tone	30	16	12	> 10 mm follicles, CL possible
5	> 30 mm in diameter, good tone, erect	> 32	20	15	> 10 mm follicles; CL present

[1]Adapted from Martin et al. (*Journal of Animal Science,* 1992, 70: 4006 – 4017).

Table 2. Effect of protein supplementation and antral follicle count on developing heifers.

Item	Treatment[1]			SEM	*P*-value	
	CGF	DDG	MOD		Treatment	Total AFC
No. of heifers	87	86	56			
Initial age, day	391	389	412	16	< 0.01	0.29
RTS	4.09	4.28	4.4	0.13	0.03	0.15
Ovarian area, mm^2	32.10	34.57	42.28	1.19	< 0.01	< 0.01
Small follicles[2]	26.46	26.23	26.40	0.20	0.53	< 0.01
Medium follicles	1.41	1.18	1.40	0.16	0.32	0.01
Large follicles	0.99	1.48	1.07	0.16	0.01	0.03
Total AFC[3]	23.09	23.29	32.52	1.94	< 0.01	—
UHD, cm^2	16.76	15.28	12.35	0.48	< 0.01	0.10
CL present, %	9.24	8.17	42.05	5	< 0.01	0.51
AI bred, %	38.73	57.09	43.72	7	0.06	0.18
Total pregnant, %	92.08	90.49	77.43	6	0.03	0.10

[1]Heifers were fed meadow hay and supplemented from November to pre-breeding with 0.78% BW corn gluten feed based supplement (CGF), 0.59% BW distillers based supplement, or 3 lb/day modified distillers grain supplement (MOD).
[2]Small follicle statistical model includes heifer treatment as a classification effect and total AFC as a covariate
[3]Total AFC statistical model does not include total AFC as a covariate.

There were no differences in small or medium follicles among treatments; however, there was a positive correlation for small follicle numbers with total AFC [AFC = 2.41 + (1.0016 x small follicles); $P < 0.01$, $r^2 = 0.97$]. Heifers supplemented with DDG had a greater ($P = 0.01$) number of large follicles compared to CGF heifers.

Uterine horn diameter was larger ($P = 0.02$) for CGF heifers compared to DDG and MOD supplemented heifers, and DDG heifers had a larger ($P < 0.01$) UHD compared to MOD.

The percent of heifers AI pregnant was greater ($P = 0.05$) for DDG heifers compared to CGF. However there was no difference in total pregnancy rate for CGF and DDG heifers. Differences in AI pregnancy rates for DDG and CGF heifers are similar to those previously reported (Martin et al., 2007 *Nebraska Beef Cattle Report*, pp. 5-6). Both CGF and DDG had increased total pregnancy rates compared to MOD heifers. Although AI pregnancy rates were greater for DDG heifers compared to CGF, reproductive tract characteristics were similar suggesting more research is needed to understand the hormonal or mechanistic actions allowing for improved AI conception rates in DDG fed heifers. These findings also suggest a correlation between small and medium follicle numbers and total AFC.

[1]Adam Summers, graduate student, University of Nebraska–Lincoln Department of Animal Science; Robert Cushman, physiologist, U.S. Meat Animal Research Center, Clay Center, Neb.; Stetson Weber, graduate student, UNL Department of Animal Science; Karl Moline, Cow/Calf unit manager; Jeff Bergman, Cow Calf unit research technician, Mead, Neb.; Matthew Spangler, assistant professor; Andrea Cupp, professor, UNL Department of Animal Science.