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Excess Rumen Undegradable Protein Alters Parameters of Reproductive Function in Beef Cows

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Summary and Implications
Evolution of the ethanol industry will likely result in future distiller’s grains feedstuffs that are more concentrated in crude protein (CP) and less concentrated in fat. However, the direct effects of excess dietary CP on reproductive function in beef cows have not been adequately addressed. In the present study, we observed that excess dietary CP derived from a rumen undegradable protein (RUP) abundant feedstuff impacted ovarian follicular growth patterns and affected follicular estradiol production and circulating concentrations of progesterone in the subsequent estrous cycle. The mechanisms by which these physiological and endocrine changes occur as a result of excessive RUP supplementation have not been fully elucidated. However, the need exists to continue research to determine how CP from these feedstuffs, when fed as a supplemental energy source in low quality forage-based diets, may impact reproductive efficiency in the beef herd.

Introduction
In the Eastern Cornbelt, corn residue is an invaluable forage resource for beef cows. However, when fed alone, baled corn residue fails to meet the energy and CP requirements of a beef cow. The accessibility of byproduct feeds such as distiller’s grains, gluten feed and condensed distiller’s syrup, are readily available, and when fed in combination with corn residue, can constitute an economical wintering ration for beef cows. However, when distiller’s dried grains with solubles (DDGS) are added to corn residue, the diet is often in gross excess of the CP requirement of that beef cow. To date, the affects of excess CP on reproductive function in beef cows has not been well elucidated.

Recent studies have reported improved pregnancy rates to timed artificial insemination (TAI) in suckled beef cows when DDGS were fed as a primary energy source during early lactation. Moreover, feeding DDGS as an energy source was also reported to increase dominant follicle diameter during the postpartum anestrous period. Given that DDGS are abundant in both fat and CP, it could not be determined which dietary constituent regulated these effects. While previous studies in dairy cattle have reported increased ovarian follicle growth and improved pregnancy rates from supplementation of unsaturated fatty acids, there is little data that definitively addresses the effects of excess CP, specifically when derived from a feedstuff that is predominantly rumen undegradable, on reproductive processes in beef cattle. Therefore, the objective of this experiment was to determine if feeding excess dietary CP, abundant in RUP, to beef cows would impact ovarian follicular growth and circulating steroid hormone concentrations. We hypothesized that excess dietary CP containing a large proportion of RUP would increase ovulatory follicle growth and increase preovulatory estradiol and post-ovulatory progesterone concentrations.

Materials and Methods
The experimental design is located in Figure 1. Non-pregnant, non-lactating beef cows (n = 20) were stratified by age, BCS, and BW, and assigned to one of 2 dietary treatments designed to deliver similar amounts of daily energy but differ in daily CP. The control diet (CON) consisted of ad libitum consumption of corn residue bales with supplementation of corn silage and corn gluten feed to meet the maintenance and CP requirements of a non-lactating, non-pregnant beef cow. The experimental diet consisted of ad libitum consumption of corn residue bales with supplemental gluten meal to meet energy requirements of the cows (PRO). Thus, in the PRO treatment daily consumption of CP that was 150% of the daily requirement. Corn gluten was used as the supplemental feed source as it is similar in protein degradability to DDGS, but lacks the fat content that may have been responsible for differences in reproductive measures discussed previously.

A dietary adaptation period of 20 days was used at the start of the trial. At the end of the adaptation period, all cows were pre-synchronized using the 5-day CO-Synch + CIDR protocol. Ten days after the conclusion of the pre-synchronization, 2.5 mg of estradiol benzoate (EB) was administered to initiate a new follicular wave. Starting at EB and daily thereafter, transrectal ultrasonography was utilized to diagram location and size of all antral follicles ≥ 3 mm in diameter on both ovaries. Ultrasound was conducted until ovulation was confirmed by the disappearance of the dominant follicle. Estrous detection was conducted twice daily throughout the ultrasound period. Blood samples were collected daily for analysis of circulating concentrations of estradiol.

Seven days after observed estrus, corpus lutea (CL) height and length were measured using the caliper function of the ultrasound. CL volume was calculated using the formula for a rotary ellipsoid (V = 4/3 π a b², where a =
longitudinal axis, and $b =$ transverse axis). Blood samples were also collected for evaluation of progesterone concentrations. It should be noted that 2 cows from each treatment became cystic and their data were thus removed.

Data were analyzed using the MIXED and GLIMMIX procedures of SAS, for continuous and binary data, respectively. REPEATED measured were used when appropriate. Cow age was used as a covariate for follicle and hormone parameters.

**Results and Discussion**

As designed, cow BW and BCS did not differ between treatments at experiment initiation or termination ($P \geq 0.76$). Moreover, as was expected, plasma urea nitrogen was greater in PRO than CON treatment at EB administration ($P < 0.001$; Table 1), indicating that PRO treated cows indeed ingested more CP than CON treated cows. Ovulatory follicle diameter was greater ($P = 0.004$; Figure 2) from day 6 after initiation of the ovulatory follicular wave to ovulation in the PRO than CON treatment. Thus, it can be concluded that increasing CP from a primarily rumen undegradable source in the diet directly affects growth rate of the eventual ovulatory follicle. Average total daily antral follicle count tended to be greater ($P = 0.06$; Table 1) in PRO than CON treatment. Moreover, peak estradiol concentrations tended to be greater ($P = 0.10$; Table 1) in the PRO than CON treatment. Numerous studies have demonstrated that greater follicular estradiol production prior to ovulation is critical for optimal fertility in cattle. Although the number of cows in this study precluded direct assessment of fertility parameters, based on previous research, the combination of increased ovulatory follicle diameter, coupled with increased antral follicle count and preovulatory estradiol concentrations would suggest that cows on the PRO treatment may be more fertile. Although ovulatory follicle diameter was greater in PRO than CON treatment, corpus lutea (CL) volume did not differ ($P = 0.34$; Table 1). This is in contrast to previous reports which have documented a strong positive relationship between ovulatory follicle and CL size. Furthermore, circulating concentrations of progesterone 7 days after estrus tended to be lesser ($P = 0.10$) in PRO than the CON treatment. This was not expected, and could be of particular concern as reduced progesterone concentrations are typically indicative of reduced capacity to maintain a pregnancy. Collectively these data illustrate that feeding increased CP abundant in RUP fraction impacted reproductive parameters prior to ovulation that would be indicative of improved fertility in cattle. However, the effect of the PRO treatment on post-ovulatory CL function is still in question. The mechanisms by which these physiological and endocrine changes occur as a result of RUP supplementation have not been fully elucidated. However, in an attempt to maintain reduced wintering feed costs in the Eastern Cornbelt, future research is warranted to better determine the impacts of CP and fat from DDGS on reproductive capacity in beef cows.
Figure 2. Effect of excess CP supplementation, abundant in RUP, on ovulatory dominant follicle growth. A treatment × day interaction ($P = 0.06$) was observed. $P$-values for treatment and day were 0.009 and $< 0.001$, respectively. Days on which ovulatory follicle diameter differed between treatments ($P < 0.05$) is indicated with *. Due to differences in wavelength, the above figure comprises a reducing data set.