The effect of increasing amount of glucogenic precursors on reproductive performance in young postpartum range cows¹

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ABSTRACT: Supplementing CP and propionate salts (PS) may improve economic returns in young range beef cows by increasing the dietary supply of glucogenic precursors. A 3-yr study conducted at Corona Range and Livestock Research Center (Corona, NM) from February to mid-July in 2005 (n = 80), 2006 (n= 81), and 2007 (n = 80) evaluated days to first estrus, calf weaning weight, BW change, and metabolic responses in 2- and 3-yr-old postpartum cows grazing native range. Cows were individually fed one of three 36% CP supplement treatments after parturition, with increasing glucogenic potential (GP) supplied by RUP and PS. Supplements were isoenergetic and fed at a rate of 908 g/cow per day twice weekly. Supplementation was initiated 7 d after calving and continued for an average of 95 d. Supplement treatments provided 1) 328 g of CP, 110 g of RUP and 0 g of PS (PS0); 2) 328 g of CP, 157 g of RUP, and 40 g of PS (PS40); or 3) 329 g of CP, 158 g of RUP, and 80 g of PS (PS80). Ultimately, PS0, PS40, and PS80 provided 44, 93, and 124 g of GP, respectively. Body weight was recorded weekly and serum was collected twice weekly for progesterone analysis to estimate days to first estrus. Cows were exposed to bulls for 60 d or less beginning in mid-May. Days to first estrus exhibited a quadratic (P =0.06) response to GP resulting from the fewest days to first estrus with the consumption of PS40. Pregnancy rates were 88, 96, and 94% for cows fed PS0, PS40, and PS80, respectively (P = 0.11). Total kilograms of calf weaned per cow exposed to bulls for the supplementation and following year increased quadratically (P = 0.09). However, supplement did not affect milk composition or yield $(P \ge 0.53)$. Serum acetate half-life decreased linearly (P = 0.08) with increasing GP in 2007. Predicted margins were the greatest (quadratic; P = 0.03) for cows fed PS40. Even though supplement costs were greater for PS40 and PS80, cows fed PS40 had increased profits (\$33.47/cow) compared with cows fed PS0 and PS80. This study implies that young postpartum cows fed additional glucogenic precursors may have improved reproductive efficiency and may wean more calf weight per cow exposed to breeding.

Key words: beef cow, glucogenic precursor, protein supplementation, reproduction

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INTRODUCTION

Young cows grazing primarily dormant range in the semiarid southwest experience negative energy balance

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during early lactation. Protein content of low-quality dormant forages tends to be more limiting to grazing animal performance than energy (Wallace, 1987). Therefore, nutritional needs of a beef cow may not be met by forage alone; thus, supplementation is necessary to minimize the protein deficiency. Protein supplementation has been found to enhance the intake and digestibility of dormant grass and improve cow performance (McCollum and Horn, 1990). After satisfying the RDP requirement, an MP deficiency may still exist. In that case, supplemental RUP can serve to meet MP needs. Feeding additional RUP has been shown to reduce days to first estrus and BW loss (Wiley et al., 1991) and to increase first-service conception rates in first-calf heifers (Triplett et al., 1995; Vasquez and Bastidas, 2005).

Supplementing RUP may also alter nutrient partitioning away from lactation (Hunter and Magner, 1988) and promote synthesis of maternal tissues for maintenance, growth, and reproduction by improved energy utilization (Miner et al., 1990; Waterman et al., 2006). Waterman et al. (2006) and Endecott (2006) found that 2- and 3-yr-old cows grazing dormant rangeland and provided high-RUP supplements plus propionate salt (PS) had decreased days to first estrus compared with cows fed cottonseed meal-based supplements. These findings indicate that feeding high-RUP supplements plus PS will decrease days to first estrus and improve pregnancy rates in 2- and 3-yr-old cows. Therefore, the objectives of this study were to determine the effect of increasing consumption of glucogenic precursors supplied as protein or PS in range supplements on days to first estrus, pregnancy rate, BW change, and calf weaning weight.

MATERIALS AND METHODS

All animal handling and experimental procedures were in accordance with guidelines set by the New Mexico State University Institutional Animal Care and Use Committee.

The study was conducted during the spring and summer for 3 consecutive years (2005 to 2007) at the New Mexico State University Corona Range and Livestock Research Center (Corona, NM). The average elevation at the Corona Range and Livestock Research Center is 2,000 m above sea level, with an average precipita-

tion of 400 mm. Rainfall during this study was 105% (2005), 76% (2006), and 117% (2007) of a 14-yr average (161 mm) for those months (Figure 1). The majority of precipitation occurs from July through September from convectional thunderstorms. The primary grass species found at the study site were blue grama (Bouteloua gracilis) and common wolftail (Lycurus phleoides; Knox, 1998; Forbes, 1999). Pasture was 762 ha and contained approximately 355 kg/ha of standing forage (A. Cibils, New Mexico State University, personal communication). All pastures were stocked at a rate that was 50% less than the Natural Resources Conservation Service recommended rate so that forage availability was assumed not to limit cow productivity in all 3 yr, even with a drought in 2006 (USDA-NRCS, 2002). Three ruminally cannulated cows were used to collect diet extrusa samples for analysis of CP (AOAC, 2000) and NDF (Van Soest et al., 1991) in 2006 and 2007. Extrusa samples were not collected in 2005 because of labor limitations. Extrusa samples were collected in April before breeding via the ruminal evacuation techniques described by Lesperance et al. (1960). Extrusa samples from the study pasture averaged (OM basis) 5.1 and 8.1% CP, and 78.6 and 85.9% NDF for 2006 and 2007, respectively.

Cows were 2 (n = 144) and 3 (n = 97) yr of age and were primarily Angus, with some Hereford influence (Table 1). Management before calving was similar in all 3 yr and between age groups. At least 60 d before calving, cows were fed 1.6 kg/cow of a 36% CP cube once per week. Within age, cows were stratified by calving

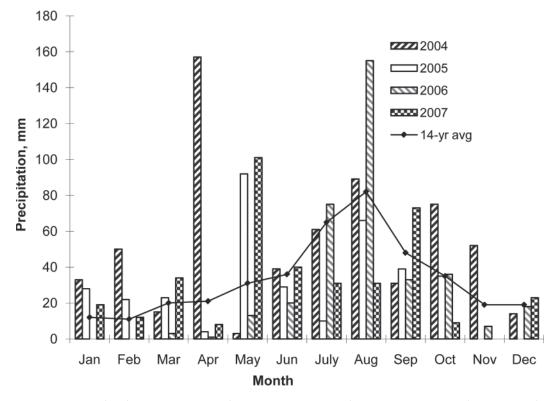


Figure 1. Annual precipitation (bars) by month for 2004 (year preceding the study), 2005, 2006, and 2007 (years of study). Line shows 14-yr average precipitation.

Table 1. Distribution (No. of cows) of 2- and 3-yr-old cows fed supplements with increasing glucogenic precursors in 2005, 2006, and 2007

Year					
	Cow age, yr	PS0	PS40	PS80	Total
2005	2	19	21	21	61
	3	5	8	6	19
2006	2	10	9	12	31
	3	17	19	14	50
2007	2	18	17	17	52
	3	8	10	10	28
Total		77	84	80	241

¹PS0 = 0 g of calcium propionate (Kemin Industries Inc., Des Moines, IA) added; PS40 = 40 g of calcium propionate added; PS80 = 80 g of calcium propionate added.

date to each supplement treatment so that age and days after calving were distributed evenly across treatments. The 2-yr-old cows were used again the next year and were reassigned randomly to treatments based on the blocking protocol. A carryover effect was not found in the second year. The breeding season began in mid-May in all 3 yr and was for a period of 60 d or less, with a bull-to-cow ratio of 1:26.

Cows were assigned randomly to 1 of 3 supplements formulated to be 36% CP on an as-fed basis, and supplements provided 1) 328 g of CP, 110 g of RUP, and 0 g of PS (**PS0**); 2) 328 g of CP, 157 g of RUP, and 40 g of PS (**PS40**); or 3) 329 g of CP, 158 g of RUP, and 80 g of PS (PS80). Ultimately, PS0, PS40, and PS80 provided 44, 93, and 124 g of glucogenic potential (**GP**), respectively. The additional GP contributed by supplemental RUP was calculated by using the equation of Preston and Leng (1987), in which 40% of the RUP is considered to be glucogenic (Overton et al., 1999). The GP of NutroCal (PS; Kemin Industries Inc., Des Moines, IA), which contains 80% propionate, is 95% glucogenic (Steinhour and Bauman, 1988). These values were used to calculate the added GP of supplements formulated with RUP and NutroCal. Cows were individually fed at a rate of 908 g/cow per day twice weekly. Supplements were formulated to be isoenergetic and were commercially cubed and milled from 2005 to 2006 at Hi-Pro Feeds (Friona, TX; Table 2) and in 2007 at Alderman Cave (Roswell, NM; 2007; Table 3). Supplementation was initiated 7 d after calving and lasted for 74 (2005), 120 (2006), and 80 (2007) d postpartum. The total number of days of supplementation was determined strategically by monitoring the average cow BW change of all the cows within each year. Supplementation was discontinued when the total cow herd BW change was no longer negative. According to these criteria, supplementation ended 14 d into breeding in 2005 and 2007 and 7 d before the end of breeding in 2006. Cows had ad libitum access to water and a loose self-fed macro- and micromineral mix yearlong.

Cows were weighed weekly from calving until the end of breeding and again at weaning (Figure 2). Days to BW nadir were determined from the lightest BW after calving. Body weight change was evaluated between

key intervals that included the beginning of supplementation to the BW nadir, the beginning of supplementation to the beginning of breeding, the BW nadir to the beginning of breeding, the end of supplementation to the end of breeding, and the initial BW to the weaning weight. Body condition scores (1 = emaciated, 9 = obese; Wagner et al., 1988) were assigned by 2 trained technicians to each cow by visual observation and palpation at the initiation of the study, branding, and weaning. Calf BW was recorded within 3 d after birth and again at branding and weaning. Calf branding and weaning weights were adjusted for a 55-d branding and 205-d weaning weight, and no adjustments were used for sex of calf or age of dam.

Blood samples (9 mL) were collected twice weekly on the days of supplementation (Monday and Friday) via coccygeal venipuncture (Corvac, Sherwood Medical, St. Louis, MO) beginning approximately 35 d postpartum (by cow) for analysis of progesterone to determine days to first estrus (2 or more consecutive progesterone concentrations >1.0 ng/mL). Blood samples were collected while cows received and consumed supplement. Blood samples were cooled and subsequently centrifuged at 4° C at $2,000 \times q$ for 20 min. Serum was collected and stored at -20° C in plastic vials for later analysis. Serum was analyzed for progesterone concentration by solid-phase RIA (Coat-A-Count, Diagnostic Products Corp., Los Angeles, CA) as described by Schneider and Hallford (1996). Inter- and intraassay CV were less than 10%, and sensitivity of the assay was 0.05 ng/mL. Cows were diagnosed as pregnant by rectal palpation at weaning or a few weeks later. Open cows at weaning were then exposed to a bull for another 60 d and palpated again the next spring. Two cows fed the PS80 did not cycle during the course of the study, and after weaning, they were placed with a bull for another 60 d. In April, these 2 cows were palpated for pregnancy and were found not to be pregnant. These cows were then considered reproductively incompetent and were removed from the study.

Serum samples were also analyzed for insulin, glucose, NEFA, urea N (\mathbf{SUN}) , and IGF-I to evaluate nutrient status. Serum samples were analyzed using commercial kits for NEFA (Wako Chemicals, Richmond, VA) and

Table 2. Composition (as-fed basis) of protein supplements containing increasing amounts of glucogenic precursors in 2005 and 2006

Item	PS0	PS40	PS80			
Ingredient, %						
Cottonseed meal	56.94	18.15	21.30			
Urea	1.20	1.20	1.20			
Wheat middlings	21.45	40.10	32.50			
Fish meal		13.00	13.00			
Hydrolyzed feather meal	0.00	12.00	12.00			
Soybean meal	10.00					
NutroCal ²	_	4.40	8.80			
Molasses	9.00	9.00	9.00			
Potassium chloride	0.95	2.00	2.05			
Monocalcium phosphate	0.30					
Vitamin A premix	0.08	0.08	0.08			
Manganese sulfate	0.06	0.05	0.05			
Trace mineral premix	0.02	0.02	0.02			
Copper sulfate	0.01	0.01				
Nutrient composition						
DM, %	87.67	88.46	88.88			
Calcium, %	0.24	1.58	2.42			
Phosphorus, %	1.00	1.09	1.06			
Magnesium, %	0.47	0.33	0.32			
Potassium, %	2.01	2.01	2.01			
Sulfur, %	0.36	0.37	0.37			
Sodium, %	0.09	0.38	0.37			
Manganese, mg/kg	210.49	210.57	210.71			
Zinc, mg/kg	109.19	199.13	284.11			
Iron, mg/kg	176.43	233.46	233.14			
Copper, mg/kg	49.82	50.45	77.84			
Selenium, mg/kg	0.24	0.53	0.53			
Cobalt, mg/kg	0.44	0.38	0.38			
Iodine, mg/kg	1.23	1.25	1.24			
Vitamin A, 1,000 IU/kg	33	33	33			
TDN, g/d	596	590	591			
CP, g/d	327	327	327			
RDP, g/d	229	167	165			
RUP, g/d	109	160	162			
Estimated glucogenic potential, g/d	44	94	126			
As fed, ³ g/d per animal	908	908	908			

 $^{^{1}\}mathrm{PS0}=0$ g of calcium propionate added; $\mathrm{PS40}=40$ g of calcium propionate added; $\mathrm{PS80}=80$ g of calcium propionate added.

SUN (Thermo Electron Corp., Waltham, MA). Glucose was analyzed with a commercial kit (enzymatic endpoint, Thermo Electron Corp., Waltham, MA). Insulin was analyzed by solid-phase RIA (Count-A-Coat, Siemens Medical Solutions Diagnostics, Los Angeles, CA) as reported by Reimers et al. (1982). Insulin assay sensitivity was 0.1 ng/mL. Serum IGF-I samples were quantified by double antibody RIA (Berrie et al., 1995) with a sensitivity of 0.1 ng/mL. Inter- and intraassay CV were less than 10%. As a chute-side measure of nutrient status and glucose sufficiency, whole-blood β-hydroxybutyrate concentrations were measured [MediSense/Abbott Laboratories, Abingdon, UK, validated by Byrne et al. (2000)] in early-May during 2006 and 2007. In 2006, the same subsample (n = 29; \sim 64 d postpartum) of cows in the glucose tolerance test (GTT) were used and whole blood was taken on subsequent

milking days. However, in 2007, the entire cow herd (n = 80; \sim 63 d postpartum) was used for the whole-blood β -hydroxybutyrate measurements on May 4.

A GTT was conducted in 2006 at approximately 64 d postpartum on a subsample of 2-yr-old (n = 13) and 3-yr-old (n = 16) cows to evaluate glucose half-life and sensitivity to endogenous insulin. Cows were brought in from pasture the day of the GTT and remained unrestrained in individual pens during the course of the challenge. A 12-gauge hypodermic needle (Ideal Instruments, Schiller Park, IL) was used to puncture the jugular vein. Approximately 0.45 m of Tygon tubing (0.10 cm i.d., 0.18 cm o.d., Cole-Parmer Instrument Company, Vernon Hills, IL) was threaded through the needle and into the jugular vein. The remaining portion (2.05 m) was secured with adhesive tape to the neck of the cow and down the middle of the back. A blunt

²Source of calcium propionate (Kemin Industries Inc., Des Moines, IA).

 $^{^3}$ Total supplement individually fed at a rate of 908 g/cow per day twice weekly.

Table 3. Composition of protein supplements (all units as fed) containing increasing amounts of glucogenic precursors in 2007

		Supplement ¹	
Item	PS0	PS40	PS80
Ingredient, %			
Cottonseed meal	57.82	59.46	61.31
Corn gluten feed	32.60	5.00	5.00
Distillers dried grain	_	17.79	10.71
Fish meal	_	7.07	9.41
$\mathrm{NutroCal}^2$	_	4.41	8.81
Urea	1.39		
Molasses	3.00	3.00	3.00
Calcium carbonate	2.94	1.56	
Monocalcium phosphate	0.63		
Potassium chloride	1.25	1.34	1.38
Copper sulfate	0.02	0.02	0.02
Manganous oxide	0.03	0.03	0.03
Selenium	0.10	0.10	0.10
Vitamin A premix	0.17	0.17	0.17
Trace mineral premix	0.07	0.07	0.07
Nutrient composition			
DM, %	90.01	90.26	90.45
Calcium, %	1.50	2.00	2.36
Phosphorus, %	1.00	1.00	1.04
Magnesium, %	0.47	0.46	0.45
Potassium, %	2.01	2.00	2.00
Sulfur, %	0.53	0.43	0.42
Sodium, %	0.43	0.42	0.38
Manganese, mg/kg	245.87	240.94	239.68
Zinc, mg/kg	135.19	138.58	138.52
Iron, mg/kg	224.05	165.01	152.51
Copper, mg/kg	51.41	51.41	51.41
Selenium, mg/kg	0.20	0.42	0.44
Cobalt, mg/kg	0.07	0.07	0.07
Iodine, mg/kg	7.78	7.78	7.78
Vitamin A, 1,000 IU/kg	33.03	33.03	33.03
TDN, g/d	613	617	587
CP, g/d	328	328	331
RDP, g/d	217	174	177
RUP, g/d	110	154	154
Estimated glucogenic potential, g/d	44	92	122
As fed, ³ g/d per animal	908	908	908

 $^{^{1}}$ PS0 = 0 g of calcium propionate added; PS40 = 40 g of calcium propionate added; PS80 = 80 g of calcium propionate added.

18-gauge needle (Salvin Dental Specialties, Charlotte, NC) was inserted into the end of the catheter, and a 10mL syringe was used as the tubing end cap. Catheters were inserted on the morning of the GTT. A 50% dextrose solution was infused at 0.5 mL/kg of BW via the indwelling jugular catheter. Blood was collected at -1, 0, 3, 6, 9, 12, 15, 20, 40, 60, 80, 100, 120, 140, 160, and 180 min relative to the infusion time. Catheters were flushed with 10 mL of a 0.9% sterile saline immediately before and after each collection time and after the infusion of glucose. At collection time -1 min, a sample was collected before infusion of glucose and 0 min immediately after infusion. Ten-milliliter blood samples were collected at each collection time and placed in Corvac serum separator tubes. Blood samples were cooled and subsequently centrifuged at 4°C at $2,000 \times g$ for 20 min. Serum was collected and stored at -20° C. Serum was stored in plastic vials at -20° C for later analysis of glucose and insulin. Insulin and glucose concentrations were analyzed as described previously. Intra- and interassay CV for both glucose and insulin were <10%.

In 2007, an acetate tolerance test (ATT) was conducted at approximately 64 d postpartum on a subsample of 2-yr-old (n = 12) and 3-yr-old (n = 12) cows to assess acetate clearance as affected by the GP of the experimental supplements. Catheter procedures were the same as reported for the GTT. A 20% acetic acid solution was infused at 1.25 mL/kg of BW via the indwelling jugular catheter. Blood collection times were -1, 0, 1, 3, 5, 7, 10, 15, 30, 60, and 90 min relative to infusion. Infusion of acetate occurred after -1 min and before 0 min. Blood samples were collected (10 mL)

²Source of calcium propionate (Kemin Industries Inc., Des Moines, IA).

³Total supplement individually fed at a rate of 908 g/cow per day twice weekly.

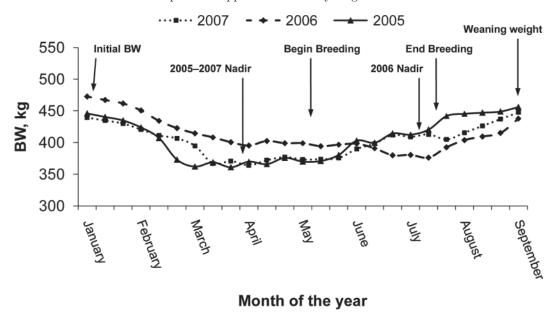


Figure 2. Timeline of specific events and average BW change that occurred during the 3-yr study.

at each collection time and were placed in Corvac serum separator tubes. Blood samples were centrifuged at $2,000 \times g$ at 4°C for 20 min, and serum was harvested. After centrifugation, samples were stored in plastic vials at -20°C for later analysis of acetate, insulin, and glucose concentrations.

Serum glucose and insulin concentrations were analyzed as described previously. Serum was filtered with a centrifugal filter device for 60 min at 4°C at $5{,}000 \times g$ for deproteinization (Millipore Amicon Ultra-4 centrifugal device, Millipore Corp., Burlington, MA). Filtered serum was mixed at a 5:1 ratio with 25\% metaphosphoric acid containing 2 g/L of 2-ethyl butyric acid as an internal standard. Samples, 1 μL in volume, were analyzed for acetate concentration by gas chromatography, as adapted from the method of Goetsch and Galyean (1983) [Varian 3400 instrument, Varian, Walnut Creek, CA; and a Nukol capillary column (30 m \times 0.25 mm; Supelco, Bellefonte, PA); temperature increase of 8°C/ min from 90°C to 200°C]. An internal standard was used to calculate final acetate concentrations, and acetate half-life was calculated as the time required for a 50% decrease in peak serum acetate concentration (Kaneko, 1989). Serum acetate, insulin, and glucose areas under the curve (AUC) were calculated using the trapezoidal summation method.

The same subsample of cows used in the GTT and ATT in 2006 (n=29) and in 2007 (n=24) were randomly selected to be equal representations of age and treatment and were milked by a portable machine (Porta-Milker, Coburn Company Inc., Whitewater, WI) approximately 57 d postpartum in 2006 and approximately 69 d postpartum in 2007. Milking procedures were a modified weigh-suckle-weigh technique described by Waterman et al. (2006). Milk weights were recorded to calculate 24-h milk production. Milk samples were analyzed for lactose, butterfat, solids-not-fat, and pro-

tein by Pioneer Dairy Labs, Dairy Herd Improvement Association (Artesia, NM).

An economic comparison was conducted to show predicted financial margins from each supplement from kilograms of calf weaned per cow exposed to breeding bulls and using treatment PS0 as the baseline. The actual postpartum feed cost was calculated for each cow with the additional yearly cost of the free-choice mineral (\$3.98/yr; Sawyer et al., 2005). All calves were valued at \$2.20/kg at weaning. The postpartum feed cost was deducted from the weaning calf value, resulting in a predicted postpartum margin (\$/cow).

In the statistical analysis, the normality of data distribution and the equality of variances of measurements were evaluated using PROC UNIVARIATE, the Levene test, and PROC GPLOT, respectively (SAS Inst. Inc., Cary, NC). Data were analyzed as a completely randomized design with cow as the experimental unit using the Kenward-Roger degrees of freedom method. The MIXED procedure (SAS Inst. Inc.) was used to test all main effects and all possible interactions. The model included the fixed effects of supplement, cow age, year, and their interactions. Covariates were calving date and days supplemented. All interactions remained in the model regardless of significance. In addition, carryover effects were tested as covariates as described by Milliken and Johnson (1984) and were not significant (P = 0.54). Preplanned contrasts were used to test for linear and quadratic effects of increasing amounts of glucogenic precursors. Serum metabolite concentrations for 2006 and 2007 were analyzed with period as the repeated factor and cow as the subject with compound symmetry as the covariance structure. The model included supplement, cow age, year, period of measurement, and associated interactions. Glucose and acetate half-lives were estimated for each animal by regressing the logarithmically transformed glucose

and acetate concentrations over time (Kaneko, 1989). Area under the curve was determined for insulin, glucose, and acetate concentrations using the trapezoidal summation method. The MIXED procedure of SAS was used to test all main effects of the ATT and GTT. The model included treatment, age, and their interaction with calving date as a covariate. Economic data were analyzed with the MIXED procedure with treatment, age, year, and their interactions in the model. Pregnancy rates were analyzed using logistic regression (PROC GENMOD of SAS) using a model that included the fixed effects of treatment, cow age, year, and their interactions. Means for statistically significant categorical data were evaluated by generating a frequency table using PROC FREQ of SAS. Significance was determined at $P \leq 0.10$.

RESULTS AND DISCUSSION

A fundamental management element leading to greater pregnancy rates in young range cows is minimizing the length of the postpartum interval (Wiltbank et al., 1961), which allows a young cow more opportuni-

ties to conceive in a defined breeding season. Days to first estrus exhibited a quadratic (P = 0.06; Table 4) response resulting from the fewest days to first estrus with consumption of PS40. Waterman et al. (2006) and Endecott (2006) found similar results when young cows were fed greater quantities of glucogenic precursors and had reduced days to first estrus. An earlier return to estrus has been shown to increase the probability that conception will occur (Randel, 1990). Furthermore, the earlier a cow conceives in the breeding season, the older, heavier, and more profitable the calf will be in the following year (Wiltbank, 1970). Pregnancy rates were 88, 96, and 94% for cows fed PS0, PS40, and PS80, respectively (P = 0.11). Therefore, supplemental GP favorably influenced days to first estrus and tended to increase pregnancy rates even though the control supplemented cows achieved a relatively high conception rate. Fewer days to first estrus did not result in a shorter calving interval the next year (P = 0.35). This response was because cows resumed estrus approximately 1 wk before the scheduled turn out of bulls.

Cow BW was similar among supplement groups at all measurement times ($P \ge 0.28$; Table 4). The cow BW change was similar for most measurement intervals

Table 4. Supplement effects on reproduction, calf and cow BW, and cow BCS for 2- and 3-yr-old postpartum cows grazing native range and fed supplements with increasing glucogenic precursors in 2005, 2006, and 2007

		$Supplement^1$			Contrast	P-value ²
Response	PS0	PS40	PS80 SEM Lin 74 2 0.4 94 — 0.1 73/78 — — — 373 3 0.3 378 2 0.9 359 2 0.7 399 2 0.4 387 2 0.6 409 3 0.5 448 3 0.6 -33 2 0.6 -3 3 0.9 38 2 0.7 13 3 0.9 38 2 0.7 11 2 0.1 -9 3 0.6 61 3 0.7 4.7 0.05 0.7 4.2 0.06 0.7 4.2 0.06 0.7 4.7 0.06 0.1 198 9 0.5	Lin	Q	
Days to first estrus	77	71	74	2	0.46	0.06
Pregnancy rate, %	88	96	94		0.13	0.11
Ratio^3	67/76	81/84	73/78	_	_	
Calving interval, d	377	376		3	0.32	0.71
Cow BW, kg						
Begin supplementation	378	379	378	2	0.90	0.77
BW nadir	359	359	359	2	0.78	0.95
End supplementation	396	396	399		0.49	0.67
Begin breeding	385	383	387		0.65	0.28
End breeding	411	409	409		0.54	0.74
Weaning	446	444	448	3	0.65	0.35
Cow BW change, kg						
Begin supplementation – BW nadir	-32	-32	-33	2	0.63	0.90
Begin supplementation — begin breeding	-3	-8	-3	3	0.98	0.07
BW nadir – end supplementation	37	35	38	2	0.71	0.32
BW nadir – begin breeding	15	9	13		0.71	0.13
BW nadir – end breeding	53	50	50	2	0.32	0.67
End supplementation — end breeding	16	14	11	2	0.11	0.88
Initial BW – weaning wt	-7	-9	-9		0.69	0.83
Days to BW nadir	59	63	61		0.76	0.35
BCS						
Initial	4.7	4.9	4.7	0.05	0.74	0.04
Branding	4.2	4.2	4.2	0.06	0.72	0.42
Weaning	4.6	4.7	4.7	0.06	0.17	0.36
Calf BW						
Kilograms weaned per cow exposed	190	207	198	9	0.52	0.18
2-year total weaned calf, kg	378	406	396	10	0.19	0.09
Predicted margins, \$/cow	0.00	33.47	-2.83	13.37	0.88	0.03

¹PS0 = 0 g of calcium propionate (NutroCal, Kemin Industries Inc., Des Moines, IA) added; PS40 = 40 g of calcium propionate added; PS80 = 80 g of calcium propionate added.

²Lin = linear contrast; Q = quadratic contrast.

³Ratio = number of cows pregnant/total number of cows in treatment.

Table 5. Supplement \times year interaction for BW change interval for 2- and 3-yr-old postpartum cows grazing native range and fed supplements with increasing glucogenic precursors in 2005, 2006, and 2007

DIV. 1		Supplement ²			Contrast	P-value ³
BW change interval ¹	PS0	PS40	PS80	SEM	Lin	Q
2005	29	16	26	6	0.64	0.07
2006	-16	-12	-24	6	0.22	0.18
2007	2	4	13	5	0.09	0.59

¹Beginning of supplementation – end of supplementation (kg).

 $(P \ge 0.11)$. However, a quadratic (P = 0.07) response to the supplements was found from the beginning of supplementation to the beginning of breeding, with PS40 cows losing the most BW. A supplement \times year interaction (P = 0.05; Table 5) also occurred from the beginning of supplementation to the end of supplementation, in which the cows in 2006 lost more BW than cows in 2005 and 2007. In 2005, cows responded quadratically (P = 0.07) to increased GP, with cows fed PS40 having the least BW gain. In 2007, BW gain during the supplementation period increased linearly (P = 0.09) with increasing GP in the diet.

Initial BCS (before calving and initiation of the experiments) exhibited a quadratic response (P = 0.04), resulting in cows in the PS40 group with a slightly greater BCS than cows fed the other 2 supplements. After calving, BCS remained similar (P = 0.17) for all cows throughout the study.

The BW nadir represents the magnitude of postpartum cow BW loss attributable to negative energy balance. In dairy cattle, the BW nadir represents the transition from negative to positive energy balance and is considered a key management indicator for the resumption of reproductive competence (Beam and Butler, 1997). The number of days to BW nadir was similar among supplement groups (P = 0.35) and did not interact with year (P = 0.16). The number of days to BW nadir in this study was longer than that reported by Endecott (2006) in a study using the same pastures with cows of the same age, but with greater precipitation in the years 2003 and 2004.

In 2006, all cows used in the GTT were considered to be insulin resistant. Supplementation did not affect $(P \ge 0.19)$ glucose AUC, insulin AUC, or glucose half-life (Table 6) as a result of a GTT. The glucose half-life of all cows was almost 3 times the normal halflife of 35 min as described by Kaneko (1989). Such a metabolic state would be consistent with the effects of the drought in 2006. The lack of supplement effects on glucose half-life, glucose AUC, and insulin AUC offers insight into the severity of undernutrition experienced by the cows in 2006, with all cows having greater than normal glucose half-lives. Thus, all cows were considered insulin resistant, and the increasing concentration of glucose supplied by supplements was likely used for milk production and was not readily taken up by insulin-sensitive tissues for BW gain and oxidative metabolism (Figure 2). Hunter and Magner (1988), Endecott (2006), and Waterman et al. (2006) have suggested that improvements in serum insulin concentrations or tissue insulin sensitivity may decrease milk and milk fat yield and potentially increase BW gain, which were not found in this current study.

Serum acetate clearance can be used as an indication of the GP of a diet and reveals the efficiency of oxidative metabolism (Cronjé et al., 1991). Additional glucogenic precursors are necessary for the efficient utilization of acetate when diets are low in protein (Cronjé

Table 6. Supplement effects on the glucose tolerance test in 2006 for 2- and 3-yr-old postpartum cows grazing native range and fed supplements with increasing glucogenic precursors

		$Supplement^1$		Contrast P-value ²		
Glucose tolerance test response	PS0	PS40	PS80	SEM	Lin	Q
Glucose half-life, min	88	97	97	15	0.66	0.83
Glucose AUC ³	10,295	10,890	13,232	1,553	0.19	0.66
Insulin AUC	183	169	188	21	0.88	0.54

 $^{^{1}}$ PS0 = 0 g of calcium propionate (NutroCal, Kemin Industries Inc., Des Moines, IA) added; PS40 = 40 g of calcium propionate added; PS80 = 80 g of calcium propionate added.

 $^{^2}$ PS0 = 0 g of calcium propionate (NutroCal, Kemin Industries Inc., Des Moines, IA) added; PS40 = 40 g of calcium propionate added; PS80 = 80 g of calcium propionate added.

 $^{^{3}}$ Lin = linear contrast; Q = quadratic contrast.

 $^{^{2}}$ Lin = linear contrast; Q = quadratic contrast.

 $^{^{3}}$ AUC = area under the curve.

Table 7. Supplement effects on acetate tolerance test in 2007 for 2- and 3-yr-old postpartum cows grazing native range and fed supplements with increasing glucogenic precursors

A		_	Contrast P-value ²			
Acetate tolerance test response	PS0	PS40	PS80	SEM	Lin	Q
Acetate half-life, min	35	29	27	3	0.08	0.08
Acetate AUC ³	247	277	247	28	0.99	0.41
Glucose AUC	9,105	8,902	7,654	515	0.06	0.42
Insulin AUC	36	34	41	7	0.63	0.57

 $^{^{1}}$ PS0 = 0 g of calcium propionate (NutroCal, Kemin Industries Inc., Des Moines, IA) added; PS40 = 40 g of calcium propionate added; PS80 = 80 g of calcium propionate added.

et al., 1991). In 2007, acetate half-life decreased linearly (P=0.08; Table 7) with increasing GP. However, acetate half-life has been reported to be as rapid as 10 min (Preston and Leng, 1987), which is approximately 3 times quicker than found in this study, suggesting that opportunities exist to further enhance oxidative metabolism. Acetate and insulin AUC were similar $(P \ge 0.41)$ among our treatment groups in 2007. However, glucose AUC decreased (P=0.06) linearly with increasing amounts of GP in the diet. These data suggest that the smaller glucose AUC indicates a faster disposal rate for cows in 2007 supplemented for greater GP, which was facilitated by a faster acetate clearance.

Whole-blood β -hydroxybutyrate concentrations can accumulate in whole blood when the rate of acetate oxidation is inhibited by an inadequate supply of cellular oxaloacetate derived from serum glucose (Kaneko, 1989). β -Hydroxybutyrate concentrations decreased linearly (P=0.01; Table 7) with increasing amounts of GP. These data concur with other findings indicating

that increasing amounts of dietary glucogenic precursors decreased ketone concentrations (Endecott, 2006). The increasing amounts of glucogenic precursors in the PS40 and PS80 groups appear to have improved utilization of metabolizable acetate, subsequently decreasing ketone concentration, which would be expected as an outcome of increased acetate clearance rate as found in the ATT in 2007.

Twenty-four-hour milk production did not differ (P = 0.26; Table 8) among supplement groups. Concentrations of milk butterfat, protein, lactose, and solids-not-fat also were not influenced $(P \geq 0.14)$ by increasing GP. In contrast, Waterman et al. (2006) found a 9% decrease in milk production and a 25% reduction in butterfat secretion for cows (~57 d postpartum) fed RUP plus 100 g of PS/d, with a 54-min glucose half-life, which was 40 min faster than in the current study. Rigout et al. (2003) also found results similar to those of Waterman et al. (2006) with a decrease in milk fat; however, they contradicted the results of Waterman et

Table 8. Supplement effects on blood ketones, milk production, and serum metabolites for 2- and 3-yr-old post-partum cows grazing native range and fed supplements with increasing glucogenic precursors in 2006 and 2007

		$Supplement^1$			Contrast P -value ²	
Response	PS0	PS40	PS80	SEM	Lin	Q
Blood ketones, mmol/L						
Whole-blood β-hydroxybutyrate	0.38	0.29	0.30	0.02	0.01	0.08
Milk, g/d						
24-h milk production	5,736	6,402	5,797	463	0.93	0.26
Butterfat	179	204	169	20	0.72	0.22
Protein	145	172	155	12	0.57	0.14
Lactose	279	315	281	22	0.96	0.19
Solids-not-fat	475	546	489	38	0.79	0.18
Serum metabolite						
SUN , $^3 mg/100 mL$	8.5	8.2	8.4	0.4	0.91	0.59
Glucose, mg/dL	54.3	55.8	57.7	1.09	0.02	0.82
Insulin, ng/mL	0.42	0.43	0.43	0.02	0.44	0.97
NEFA, mmol/L	450	481	482	15	0.13	0.42

¹PS0 = 0 g of calcium propionate (NutroCal, Kemin Industries Inc., Des Moines, IA) added; PS40 = 40 g of calcium propionate added; PS80 = 80 g of calcium propionate added.

²Lin = linear contrast; Q = quadratic contrast.

 $^{^{3}}$ AUC = area under the curve.

²Lin = linear contrast; Q = quadratic contrast.

 $^{^{3}}SUN = serum urea N.$

Table 9. Supplement \times year interaction for serum IGF-I of 2- and 3-yr-old cows grazing native range and fed supplements with increasing glucogenic precursors in 2006 and 2007

- 0		$Supplement^1\\$			Contrast	P -value 2
IGF-I, ng/mL	PS0	PS40	PS80	SEM	Lin	Q
2006 2007	37.7 51.7	48.9 51.7	42.5 59.1	5 4	0.38 0.14	0.05 0.36

 $^{^{1}}$ PS0 = 0 g of calcium propionate (NutroCal, Kemin Industries Inc., Des Moines, IA) added; PS40 = 40 g of calcium propionate added; PS80 = 80 g of calcium propionate added.

al. (2006) in that milk production increased when glucogenic precursors were infused into either the rumen or duodenum of dairy cows.

Serum urea N concentrations were similar (P=0.59; Table 8) among the 3 supplement groups. Serum urea N concentrations of 10 to 12 mg/100 mL are usually considered optimal (Hammond et al., 1993; Stateler et al., 1995). All supplement means were below that optimal concentration. Therefore, forage protein was not in abundance and the SUN concentrations were not increased by any of the supplements, which suggests that cows used supplemental protein efficiently even though the RDP:RUP ratios varied between formulations.

Serum glucose concentrations increased linearly (P=0.02) with increasing consumption of glucogenic precursors. In contrast, multiple studies have found a decrease or no increase in serum glucose with feeding increasing amounts of glucogenic precursors (Cronjé et al., 1991; Vanhatalo et al., 2003; Waterman et al., 2006). Despite cows having different serum glucose concentrations, their serum insulin concentrations were similar (P=0.44) between treatments. Serum NEFA concentrations were also similar (P=0.13) with increasing GP.

Insulin-like growth factor-I has been suggested to be a better indicator of the rebreeding performance of first-calf heifers than BCS or BW change (Roberts, 2008). Furthermore, circulating IGF-I concentration is associated with nutrient intake (McGuire et al., 1992) and is an indicator of nutrient status in dairy (Spicer et al., 1990) and beef (Roberts et al., 1997) cattle. A treatment \times year interaction was observed for IGF-I concentrations (P = 0.02; Table 9). In 2006, cows fed PS40 had a greater concentration of IGF-I than

did cows fed PS0 (quadratic; P=0.05). However, in 2007, concentration of IGF-I was not different (P=0.14) among treatments. This interaction in IGF-I values between years may have been caused by the effects of low rainfall in 2006 compared with 2007 (Figure 1). Therefore, feeding RUP supplements with PS may exhibit a more consistent response (as shown by IGF-I) even in drought conditions and may help alleviate the negative association between drought conditions and nutrient intake.

A supplement × age interaction occurred ($P \le 0.01$; Table 10) for calf BW at branding (adjusted 55 d of age). Increasing the consumption of GP did not influence (P = 0.24) the branding weight of calves from 2-yr-old cows. However, the 3-yr-old cows fed PS80 had the lightest calves at branding compared with the 3-yr-old cows fed PS0 and PS40 (quadratic; P = 0.02). A supplement × year (P = 0.10; Table 11) interaction was observed for 205-d weaning weight. Differences in GP among supplements did not affect calf weaning weights in 2005 and 2006; however, in 2007 calf weaning weights responded quadratically (P = 0.06), with calves from cows fed PS40 having the heaviest weaning weights.

The total number of kilograms of calf weaned per cow exposed to breeding bulls has been suggested to be a primary production evaluation criterion that takes into account reproductive success and calf growth potential. It is the sum of the influences of the conditions of the current year, milk production, and the response of the previous year to conception timing, reproductive rate, and culling rate (Ramsey et al., 2005). The number of calves weaned in relation to the number of breeding-age cows in the herd is a key indicator of efficiency. Increas-

Table 10. Supplement × cow age interaction for calf branding weight (55 d) for 2- and 3-yr-old postpartum cows grazing native range and fed supplements with increasing glucogenic precursors in 2005, 2006, and 2007

		Supplement ¹		Contrast P -value ²		
Cow branding wt, kg	PS0	PS40	PS80	SEM	Lin	Q
2 yr old	63	60	64	3	0.67	0.24
3 yr old	64	67	57	3	0.06	0.02

 $^{^{1}}$ PS0 = 0 g of calcium propionate (NutroCal, Kemin Industries Inc., Des Moines, IA) added; PS40 = 40 g of calcium propionate added; PS80 = 80 g of calcium propionate added.

²Lin = linear contrast; Q = quadratic contrast.

²Lin = linear contrast; Q = quadratic contrast.

Table 11. Supplement \times year interaction for calf weaning weight for 2- and 3-yr-old postpartum cows grazing native range and fed supplements with increasing glucogenic precursors in 2005, 2006, and 2007

207.1		Supplement ¹		Contrast P -value ²		
205-d weaning wt, kg	PS0	PS40	PS80	SEM	Lin	Q
2005	218	222	216	6	0.79	0.42
2006	158	155	165	6	0.37	0.30
2007	217	225	210	7	0.42	0.06

 $^{^{1}}$ PS0 = 0 g of calcium propionate (NutroCal, Kemin Industries Inc., Des Moines, IA) added; PS40 = 40 g of calcium propionate added; PS80 = 80 g of calcium propionate added.

ing amounts of GP, in this study, decreased days to first estrus, providing the opportunity to wean heavier or older calves the next year. Ramsey et al. (2005) defined ranch productivity as pounds weaned per exposed female, which integrates 3 main production variables: calving percentage, calf death loss, and breeding-season length. The importance of reproduction in young breeding females to profitability has also been demonstrated previously (Meek et al., 1999; Patterson et al., 2003). The total number of kilograms of calf weaned per cow exposed to breeding bulls was similar among treatment groups (P = 0.33; Table 4). However, the total number of kilograms weaned for the supplemental year and the subsequent year increased (quadratic; P = 0.09) with increasing consumption of GP of the diet. Over the 3 yr, predicted margins were the greatest (quadratic; P= 0.03) for cows fed PS40. Even though supplement costs were greater for PS40 and PS80, cows fed PS40 had increased profits (\$33.47/cow) compared with feeding cows PS0 and PS80. Endecott (2006) found similar results of a \$19.42/cow increase in income when feeding RUP plus 80 g of calcium propionate compared with a traditional cottonseed meal-based supplement. Cows fed PS40 were just as reproductively efficient as cows fed PS80 but were more cost effective. Within this study, feeding 40 g/d of calcium propionate (PS40) was the most likely cost-effective postpartum supplementation strategy.

Results of the measurement days to first estrus agree with the findings of Waterman et al. (2006) and Endecott (2006). Cows fed increasing amounts of glucogenic precursors returned to estrus sooner than cows not supplemented with PS. However, supplementation with glucogenic precursors did not alter nutrient partitioning, as proposed by Waterman et al. (2006) and Endecott (2006). One explanation for this discrepancy is that all cows in 2006 were insulin resistant based on GTT. Conversely, energy metabolism was improved in 2007 when cows were fed increasing amounts of glucogenic precursors. Acetate half-life, ketone concentration, and glucose AUC in 2007 decreased linearly with increasing amount of glucogenic precursors. Therefore, cows might still have been insulin insensitive, yet they were more efficient at utilizing other energy substrates such as acetate, thus allowing for improved nutrient utilization. However, the conditions of 2006 and 2007 were different because of a difference in the amount and timing of precipitation. The GTT in 2006, the ATT in 2007, and differences in the duration of BW loss suggest that cows in 2007 may have been more efficient than those in 2006. The finding of no interactions between any reproductive measures and the main factors of supplement, age, and year suggests that potentially all supplements used had consistent effects on cow metabolism during the 3 yr, independent of a BW change or change in BCS. Cows fed increasing amounts of glucogenic precursors required fewer days to first estrus, with numerically greater pregnancy rates, and they weaned heavier calves in the subsequent year, resulting in an increased predicted income.

In conclusion, supplementing young cows with 40 g of calcium propionate provided the greatest response with decreased days to first estrus in 2- and 3-yr-old range cows. Cows fed additional glucogenic precursors appeared to wean heavier calves the following year, which increased returns beyond the expense of the greater cost of supplement ingredients. Diets that supply additional glucogenic precursors may decrease serum ketone concentrations and increase acetate disappearance rate, indicating a more efficient energy metabolism and better use of forage energy. These results indicate a more efficient overall energy metabolism and reproduction by feeding additional glucogenic precursors.

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²Lin = linear contrast; Q = quadratic contrast.

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