Effects of dietary fat and sire breed on puberty, weight, and reproductive traits of F_1 beef heifers[1]

M. A. Lammoglia*, R. A. Bellows†,[2], E. E. Grings†, J. W. Bergman‡, S. E. Bellows†, R. E. Short†, D. M. Hallford§, and R. D. Randel¶

*BovaGen, San Antonio, TX 78216; †USDA-ARS, Fort Keogh Livestock and Range Research Laboratory, Miles City, MT 59301; ‡Eastern Agricultural Research Center, Sidney, MT 59270; §Department of Animal and Range Sciences, New Mexico State University Las Cruces, 88003; and ¶Texas A&M University, Texas Agricultural Experiment Station, Overton 75684

ABSTRACT: Prepubertal F_1 heifers (n = 246; from crossbred dams bred to either Hereford [H], Limousin [L], or Piedmontese [P] sires) were fed 1.9% (LF) or 4.4% (HF) dietary fat from 254 ± 4 d of age until they reached puberty or the breeding season started. Safflower seeds (37% oil with 79% linoleic acid) were the added fat source. Blood samples and backfat thickness measurements were obtained from 60 randomly selected heifers representing the sire breeds and diets studied. In addition, five H-sired heifers from both diets were serially bled at 28-d intervals. Total gain, ADG, body condition score, and backfat thickness were affected by sire breed ($P < 0.001$) but not diet. Backfat thickness was affected ($P < 0.01$) by the diet × time on feed interaction. Diet did not affect pubertal age ($P > 0.10$) but tended ($P = 0.08$) to affect the percentage of heifers pubertal by the beginning of breeding (June 4). Sire breed effects on puberty age at beginning of breeding, percentage pubertal at the beginning of breeding, and puberty age during the entire study were all highly significant. The effect of the diet × sire breed interaction on percentage of heifers pubertal at beginning of breeding ($P < 0.05$) was 74.4 vs 76.3% in H-sired, 69.8 vs 60.5% in L-sired, and 76.2 vs 97.6% in P-sired heifers (LF vs HF, respectively). Number of AI services per pregnancy and final pregnancy percentage were not affected by diet or the diet × sire breed interaction. Diet affected progesterone ($P < 0.05$) and cholesterol ($P < 0.001$) concentrations, and sire breed tended to affect ($P = 0.06$) cholesterol concentrations. The effect of the diet × time on feed interaction on cholesterol concentrations was highly significant. There were no effects of diet or sample period on insulin or growth hormone concentrations in serially collected blood samples. We conclude that effects of supplemental dietary fat may be breed-dependent and hypothesize that a feeding period of approximately 60 d duration may be more appropriate than the 162 d used in this study.

Key Words: Beef Breeds, Dietary Fat, Puberty, Safflower

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Introduction

Age at puberty is an important production trait when heifers are bred to calve as 2 yr olds and in systems that impose restricted breeding seasons. Hormones, nutrition, and genotype play key roles in attainment of puberty (Patterson et al., 1992; Bellows and Hall, 1996). Heifers that have an increase in serum progesterone before first estrus (Berardinelli et al., 1979) have a higher incidence of development of a functional corpus luteum (**CL**) following estrus (Rutter and Randel, 1986). Byerley et al. (1987) found that heifers bred at their third postpubertal estrus had a pregnancy rate of 78%, compared with 57% for those bred at their pubertal estrus. Staigmiller et al. (1993) concluded this increase resulted from "maturation" of the uterine environment and pregnancy maintenance mechanisms, which extended the hypothesis of Del Vecchio et al. (1991) that attainment of puberty involves synchronous communication between the uterus and ovaries.

Feeding cows supplemental dietary fat increased serum progesterone, serum and follicular fluid cholesterol

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[2]Correspondence: Rt. 1, Box 2021 (phone: 406/232-4970; fax: 406/232-8209; E-mail: bob@larrl.ars.usda.gov).

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Table 1. Experimental design and number of F_1 heifers per subgroup

Diet	Fat content, %	Sire breed			Total number of heifers
		Hereford	Limousin	Piedmontese	
Low fat	1.9	39	43	41	123
High fat	4.4	39	42	42	123
Sire breed totals	—	78	85	83	246

concentrations, and the area occupied by lipids in the small and large luteal cells (Williams, 1989; Hightshoe et al., 1991; Ryan et al., 1992). Cows supplemented with fat had increased number and size of large follicles (Lucy et al., 1991), increased number of cows with luteal activity (Wehrman et al., 1991), prolonged lifespan of induced CL (Williams, 1989), increased serum progesterone concentrations after the first postpartum ovulation (Hightshoe et al., 1991), and increased pregnancy rates (Bellows et al., 1999). Thus, studies on the effects of feeding supplemental fat on puberty in heifers are warranted. The objectives of this study were to evaluate the effects of supplemental dietary fat and sire breed on puberty and subsequent pregnancy rates, backfat thickness, serum progesterone, insulin, GH, and $PGF_{2\alpha}$ concentrations in replacement heifers.

Materials and Methods

Two hundred forty-six spring-born, prepubertal F_1 heifers from crossbred dams of mixed breeding bred to either Hereford (**H**), Limousin (**L**), or Piedmontese (**P**) sires (Grings et al., 1999) were randomly allotted by sire and breed to receive a diet containing either 1.9% (**LF**) or 4.4% (**HF**) dietary fat (Table 1). Average initial body weights were 228.1 ± 3.1 kg, 244.8 ± 3.3 kg, and 232.5 ± 2.9 kg for H-, L-, and P-sired heifers, respectively. Safflower (*Carthamus tinctorius* L. var. Centennial) seeds containing 37% oil with 79.1% linoleic acid, 6.2% palmitic, 2.1% stearic, 10.3% oleic, and 2.3% other fatty acids were used as the supplemental fat source. Seeds were rolled sufficiently to crack the seed hulls (Lammoglia et al., 1999a) before being added to the diet mix. Diets were formulated to be approximately equal in energy and protein content (Table 2).

Heifers were brought to the feedlots 40 d before the beginning of the study for an adaptation period and fed the diet containing 1.9% dietary fat. Epididymectomized bulls equipped with marking harnesses were placed with the heifers (1 bull/25 heifers) to aid in detection of estrus, which was recorded twice daily. Heifers were fed their respective diets in 10 pens containing 24 or 25 animals each from approximately 254 d of age until either puberty was attained or the breeding season began on June 4. The time from starting the experimental diets (December 24) until beginning of the breeding season was 162 d. Diet consumption and weight gains were evaluated every 56 d and, when necessary, amounts fed were adjusted (without changing fat concentrations) to maintain an ADG of approximately 0.6 kg and meet NRC (NRC, 1984) requirements for protein.

Heifers were weighed every 28 d and palpated body condition scores (**BCS**; 1 = emaciated; 10 = obese) were given every 56 d by two experienced individuals. Sixty heifers, randomly selected to represent both diets and the three sire breeds, had backfat thickness measured at 56-d intervals using ultrasonography (LS-1000 with a 5-MHz transducer; Tokyo Keiki, Japan). Measurements were taken between the 12th and 13th rib on the left side of the animal. In addition, these heifers were bled via tail venipuncture every 28 d starting on 254 ± 4 d of age until puberty was reached or the breeding season began. Blood samples were kept at 5°C and centrifuged within 12 h following collection to yield serum. Serum samples were maintained at –20°C until cholesterol concentrations were determined using commercially available enzymatic reaction kits (Kit 2340; DMA, Arlington, TX).

Starting at 254 d of age until heifers reached puberty or the breeding season began, serial blood samples were collected via jugular vein catheter every 28 d from an additional 10 F_1 H heifers representing both diets (five per group). Catheters were inserted under local anesthesia using aseptic techniques. Due to limitations in facilities available for cannulation and for collecting serial blood samples, it was necessary to limit this phase of work to a single sire breed. Hereford-sired heifers were selected because of their temperament and

Table 2. Composition of experimental diets

Ingredient and item	Diet	
	Low fat	High fat
Corn silage	51.8[a]	51.6[a]
Ground alfalfa hay	33.3	33.2
Safflower seeds	—	7.8
Ground barley	13.8	7.4
Soybean oil meal	1.1	—
Chemical analyses[b]		
Crude protein, %	11.7	10.8
Fat, %	1.9	4.4
Dry matter, %	46.2	46.4
Estimated TDN, %[c]	64.6	65.1

[a]Percentage of diet; DM basis. Heifers had ad libitum access to trace mineralized salt.
[b]Analyses of weekly composite samples collected throughout feeding period.
[c]Calculated value based on ADF (Adams, 1980).

they represented a common beef breed. Serial blood samples were collected prior to feeding at 15-min intervals for 240 min. Approximately 20 mL of blood was drawn per sample and placed in tubes containing EDTA. Blood samples were placed in ice and centrifuged within 30 min after collection to yield plasma. Plasma samples were kept at –20°C until GH, insulin, and $PGF_{2\alpha}$ concentrations were determined using RIA procedures. Plasma $PGF_{2\alpha}$ concentrations were determined with a direct single antibody technique as described by Del Vecchio et al. (1990) and modified by Velez et al. (1991). The intra- and interassay CV were 9.8 and 9.1%, respectively. Plasma insulin and GH concentrations were determined by procedures described by Sanson and Hallford (1984) and Hoefler and Hallford (1987), respectively. Plasma insulin concentrations were determined in two assays with an intra- and interassay CV of 8.9 and 13.7%, respectively, and a recovery of added mass of 92%. Plasma GH concentrations were quantified in two assays with intra- and interassay CV of 12.5 and 15%, respectively.

A blood sample was collected via tail venipuncture from all estrual heifers between 7 and 10 d after behavioral estrus. Blood samples were maintained at 5°C and processed within 12 h to yield serum, which was maintained at –20°C until progesterone concentrations were determined using coated tubes (Kit TKPGX; DPC, Los Angeles, CA) as described by Bellows et al. (1991). Intra- and interassay CV were 8.0 and 9.7%, respectively. Serum progesterone concentrations were used to evaluate function of the CL. Also, between 7 and 10 d after estrus, the ovaries were scanned ultrasonically (LS-1000 with a 5-MHz transducer; Tokyo Keiki, Japan) for the presence of a CL. Puberty was defined as the occurrence of estrus with subsequent formation of a CL capable of maintaining a minimum serum progesterone concentration of 1 ng/mL between d 7 and 10 of the estrous cycle. Heifers attaining these three criteria were considered pubertal but were continued on their respective diets until the beginning of the breeding season. Heifers exhibiting estrus but failing to attain any of these three criteria were considered to be prepubertal and to have exhibited a nonpubertal estrus and samplings were repeated at the next observed estrus.

Heifers were moved to an irrigated mixed-grass pasture at the beginning of the breeding season and all supplemental diet feeding was terminated. Heifers that did not reach puberty during the diet-feeding period (n = 42) were considered pubertal the day they exhibited estrus and were artificially inseminated during the subsequent breeding season. The presence of heifers that did not exhibit estrus by the end of the 54-d AI breeding season was essentially random throughout the diet-sire breed subgroups. These animals (n = 23) were considered pubertal the day after the breeding season ended. The data were included in subsequent statistical analyses and mean values are presented in Table 4 as puberty age over the entire study.

Data were analyzed using SAS GLM procedures for analysis of variance and differences for diet, sire breed, day, and the interactions among groups were determined by least squares means methods (SAS, 1994). Initial body weights and condition scores were used as covariates in appropriate analyses. Significance of breed effects was tested using the mean square for sire-within-breed, and other effects were tested using the residual mean squares as error terms. Progesterone concentrations were analyzed using day of the estrous cycle on which the blood sample was obtained as a covariate. Comparisons of the percentage of pubertal heifers at the start of the breeding season and pregnancy rates in both main and interaction effects were made using CATMOD procedures (SAS, 1994).

Results and Discussion

Diet and sire-breed main effect and interaction means, pooled standard errors, and statistical significance are summarized in Tables 3 through 5. Significant interactions with days on diet treatment or time of obtaining serial blood samples that were pertinent to hypotheses under test are shown in Figures 1 to 4. All other interactions were nonsignificant.

Average body weights at the beginning of the breeding season were 348.5 ± 4.5 kg, 365.6 ± 4.8 kg, and 334.0 ± 4.1 kg for H-, L-, and P-sired heifers, respectively. Total heifer gain during the feeding period, ADG, and BCS were not affected ($P > 0.10$) by diet (Table 3). These results were expected because diets were formulated to be approximately equal in energy and protein content and diets were adjusted to obtain the desired daily gain. However, sire-breed effects on these end points were highly significant, with H- and L-sired heifers exceeding P-sired heifers in all three traits.

Diet main effects on backfat thickness approached significance ($P = 0.09$). Because BCS was not affected by diet, we interpret this result to indicate that ultrasound measurements of backfat thickness were a more critical and accurate measure of diet effects than were the palpated condition scores. Sire-breed effect on backfat thickness was highly significant, with P-sired heifers having the least backfat. The interaction of diet × days on diet treatment ($P = 0.02$) is summarized in Figure 1. Figure 1 suggests there was no diet effect on the change in backfat thickness from 0 to 56 d of feeding. At that time backfat thickness in HF heifers increased more rapidly than that in LF heifers, and the difference was maintained throughout the remainder of the feeding period.

Feeding supplemental fat has been reported not only to increase energy density, but also to improve energetic efficiency (Palmquist, 1994; Wu and Huber, 1994). Rhodes et al. (1978) found greater fat deposition in lipid-fed cattle than in controls, but there were no differences in ADG. Even though diets in the present study were formulated to be similar in energy and protein content, addition of fat to the diets influenced backfat

Table 3. Least squares mean values and statistical significance for diet and sire-breed effects on heifer gains, condition scores, and backfat thickness

Item and diet	Breed of sire			Diet means	Statistical significance (source, probability)
	Hereford	Limousin	Piedmontese		
Total gain, kg[a]					
Low fat (n = 123)	122.2 ± 4.2	116.8 ± 3.7	98.7 ± 4.5	112.5 ± 2.4	Diet (D), $P > 0.10$
High fat (n = 123)	118.1 ± 4.3	123.6 ± 3.5	101.3 ± 5.5	114.3 ± 2.6	Sire breed (B), $P < 0.001$
Sire breed means	120.1 ± 3.0	120.2 ± 2.5	100.0 ± 3.6	—	D × B, $P > 0.10$
Average daily gain, kg[a]					
Low fat (n = 123)	0.84 ± 0.03	0.80 ± 0.02	0.68 ± 0.03	0.77 ± 0.02	Diet, $P > 0.10$
High fat (n = 123)	0.81 ± 0.03	0.85 ± 0.02	0.69 ± 0.04	0.78 ± 0.02	Sire breed, $P < 0.001$
Sire breed means	0.82 ± 0.02	0.82 ± 0.02	0.68 ± 0.02	—	D × B, $P > 0.10$
Body condition score[b]					
Low fat (n = 123)	6.0 ± 0.3	6.9 ± 0.2	5.8 ± 0.2	6.2 ± 0.1	Diet, $P > 0.10$
High fat (n = 123)	7.3 ± 0.2	6.6 ± 0.2	5.2 ± 0.3	6.4 ± 0.2	Sire breed, $P < 0.001$
Sire breed means	6.7 ± 0.2	6.7 ± 0.1	5.5 ± 0.2	—	D × B, $P < 0.001$
Backfat thickness, mm[b]					
Low fat (n = 30)	4.0 ± 0.4	3.4 ± 0.2	2.6 ± 0.3	3.4 ± 0.2	Diet, $P = 0.09$
High fat (n = 30)	4.7 ± 0.3	4.3 ± 0.3	2.8 ± 0.4	3.9 ± 0.2	Sire breed, $P < 0.001$
Sire breed means	4.4 ± 0.2	3.8 ± 0.2	2.7 ± 0.2	—	D × B, $P > 0.10$

[a]During the entire feeding period.
[b]At the end of the diet feeding period (162 d).

thickness. We suggest that the differences could be a result of greater utilization efficiency of dietary energy in HF heifers.

The interaction effect of sire breed × days on diet treatment on backfat thickness was highly significant and is summarized in Figure 2. The increase in backfat thickness was greatest in H-sired heifers, least in P-sired heifers, and intermediate in L-sired heifers. These differences agree with Ferrell (1982), who reported breed differences in heifer BCS. In this study P-sired heifers had the lowest BCS and backfat thickness, which supports the findings that this breed has the genetic predisposition for muscular hypertrophy and low body fat (Arthur, 1995). Cattle with muscular hypertrophy had less total fat than cattle with no muscular hypertrophy (Karima and Berg, 1985). Hall et al. (1995) reported an increase in BCS and backfat thickness 75 d before beef heifers reached puberty. Buckley et al. (1990) demonstrated that beef heifers had a dramatic increase in fat content from 8 to 14 mo of age, which is the time when most beef heifers reach puberty, supporting results found in this study (Figure 2). Smith et al. (1987) suggested that age of animal dictates time of onset of the de novo lipogenesis (which is between d 167 and 236 in steers), whereas diet modulates the amplitude of the rate of lipogenesis. Therefore, time changes in backfat thickness in the present study could have been caused by an age and diet effect on de novo lipogenesis.

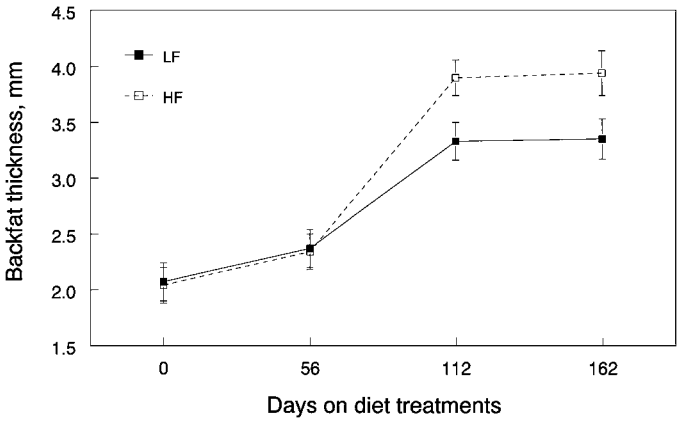

Figure 1. Least squares means plot (pooled SEM = 0.17) of backfat thickness of heifers receiving 1.9% (LF) or 4.4% (HF) dietary fat as affected by dietary treatment × days on diet treatment ($P = 0.02$; n = 60).

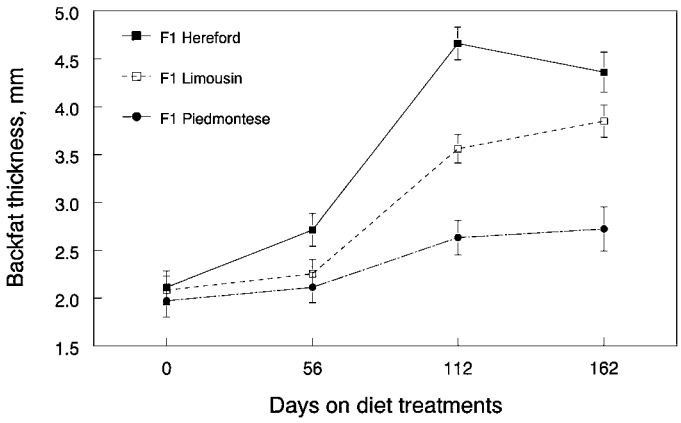

Figure 2. Least squares means plot (pooled SEM = 0.17) of backfat thickness of Hereford, Limousin, and Piedmontese-sired heifers as affected by sire breed × days on diet treatment ($P < 0.001$; n = 60).

Table 4. Least squares mean values and statistical significance for puberty and reproduction data

Item and diet	Breed of sire			Diet means	Statistical significance (source, probability)
	Hereford	Limousin	Piedmontese		
Puberty age (d) of heifers pubertal by begin breeding					
Low fat (n = 92)	357.4 ± 6.8	379.0 ± 6.7	338.1 ± 6.7	358.2 ± 3.9	Diet (D), $P > 0.10$
High fat (n = 95)	360.9 ± 6.5	380.7 ± 7.6	349.2 ± 5.9	363.6 ± 4.0	Sire breed (B), $P < 0.001$
Sire breed means	359.1 ± 4.7	379.8 ± 5.1	343.7 ± 4.5	—	D × B, $P > 0.10$
Percentage pubertal at begin breeding					
Low fat (n = 123)	74.4 ± 0.07	69.8 ± 0.07	76.2 ± 0.06	73.4 ± 0.04	Diet, $P = 0.08$
High fat (n = 123)	76.3 ± 0.07	60.5 ± 0.07	97.6 ± 0.02	78.1 ± 0.04	Sire breed, $P < 0.01$
Sire breed means	75.3 ± 0.05	65.1 ± 0.05	86.9 ± 0.03	—	D × B, $P < 0.05$
Puberty age (d) entire study[a]					
Low fat (n = 119[b])	390.3 ± 9.6	407.4 ± 9.2	368.6 ± 9.4	388.8 ± 5.4	Diet, $P > 0.10$
High fat (n = 121)	388.7 ± 9.9	416.5 ± 9.1	362.8 ± 9.3	389.3 ± 5.4	Sire breed, $P < 0.001$
Sire breed means	389.5 ± 7.5	412.0 ± 7.0	365.7 ± 7.1	—	D × B, $P > 0.10$
Breeding data					
Number AI services per pregnancy					
Low fat (n = 104)	1.25 ± 0.14	1.35 ± 0.13	1.54 ± 0.14	1.38 ± 0.08	Diet, $P > 0.10$
High fat (n = 94)	1.41 ± 0.14	1.38 ± 0.17	1.54 ± 0.14	1.44 ± 0.09	Sire breed, $P > 0.10$
Sire breed means	1.33 ± 0.10	1.36 ± 0.11	1.54 ± 0.10	—	D × B, $P > 0.10$
Pregnancy, %					
Low fat (n = 119[b])	79.5 ± 0.06	82.9 ± 0.06	65.0 ± 0.08	75.5 ± 0.04	Diet, $P > 0.10$
High fat (n = 121)	81.6 ± 0.06	68.3 ± 0.07	68.3 ± 0.07	72.7 ± 0.04	Sire breed, $P = 0.10$
Sire breed means	80.6 ± 0.04	75.6 ± 0.05	66.6 ± 0.05	—	D × B, $P > 0.10$

[a]Includes heifers pubertal during the breeding season and assigned age for heifers not showing estrus by the end of the breeding season (see text for calculation of assigned age).
[b]Four Limousin- and Piedmontese-sired heifers culled for factors not related to this study at the beginning of breeding season.

Effects of diet, sire breed, and the diet × sire breed interaction on reproduction traits are summarized in Table 4. Diet main effects on puberty age of heifers pubertal at the beginning of the breeding season and puberty age for the entire study were not significant. However, the diet main effect showed at the beginning of the breeding season (June 4) more ($P = 0.08$) heifers fed the HF diet were pubertal than heifers fed the LF diet. Sire-breed main effects on age of puberty of heifers pubertal at the beginning of the breeding season, percentage pubertal at the beginning of the breeding season, and age of puberty for the entire study were all significant or highly significant. These differences were consistently in the direction of L-sired heifers being the oldest, P-sired the youngest, and H-sired heifers intermediate. The interaction of diet × sire breed was significant for the percentage of heifers pubertal at the beginning of the breeding season. Diet effects tended to be minimal in H-sired heifers, a lower percentage were noted in L-sired heifers on the HF diet, and a higher percentage were seen in P-sired heifers on the HF diet. We interpret this interaction effect to indicate that the effects of fat supplementation are breed-dependent. Diet × sire breed interaction effects on puberty age at the beginning of the breeding season and for the entire study were nonsignificant.

Martin et al. (1992) reported breed differences in age at puberty in heifers. Ferrell (1982) suggested that breeds selected for greater milk production reached puberty at younger ages and lighter weights relative to mature weight than breeds selected for growth and beef production. Grings et al. (1999) studied age at puberty and milk production in heifers of the same breeding as those used in the present study. Piedmontese-sired heifers were younger at puberty than H- or L-sired heifers and milk production at 70 d of lactation did not differ among the three breeds. However, at 120 d of lactation P-sired heifers produced approximately the same amount of milk as H-sired heifers but less than L-sired heifers. In the present study, P-sired heifers receiving the HF ration had the highest percentage of puberty at the beginning of the breeding season and had the lowest BCS and backfat thickness. When evaluated as a breed, P-sired heifers had the lowest average BCS and backfat thickness, but feeding the HF diet increased the percentage of P-sired heifers pubertal at the beginning of the breeding season by 21.4 percentage points. We interpret these results to suggest that because P-sired heifers have a low body fat component (Arthur, 1995) and had the lowest backfat thickness in the present study (Figure 2), they may have a greater dietary fat "requirement" than did either the H- or L-sired heifers, thus resulting in their positive response to supplemental dietary fat.

Breeding data are also summarized in Table 4. Diet effects on number of AI services per pregnancy or final pregnancy percentage at the end of the 54 d breeding season were not significant. Effects of sire breed on

Table 5. Least squares mean values and statistical significance of effects of diet, sire breed, and collection date on hormone and metabolite concentrations

Compound and diet	Breed of sire: Hereford	Limousin	Piedmontese	Diet means	Statistical significance (source, probability)
Progesterone, ng/mL					
Low fat (n = 92)	2.87 ± 0.27	3.52 ± 0.26	3.13 ± 0.25	3.17 ± 0.15	Diet (D), $P < 0.05$
High fat (n = 95)	3.46 ± 0.25	3.49 ± 0.32	3.84 ± 0.23	3.60 ± 0.16	Sire breed (B), $P > 0.10$
Sire breed means	3.16 ± 0.18	3.51 ± 0.21	3.48 ± 0.17	—	D × B, $P > 0.10$
Cholesterol, mg/dL					
Low fat (n = 25)	107.2 ± 4.4	119.7 ± 4.3	102.5 ± 4.4	109.8 ± 2.6	Diet, $P < 0.001$
High fat (n = 26)	164.9 ± 4.0	177.2 ± 3.8	167.3 ± 4.2	169.8 ± 2.3	Sire breed, $P = 0.06$
Sire breed means	136.1 ± 3.0	148.4 ± 2.9	134.9 ± 3.1	—	D × B, $P > 0.10$

	Sample period (day of feeding period): 0	28	56	84	112	140		
Data from serial bleedings[a]								
Insulin, ng/mL								
Low fat (n = 5)	2.06 ± 0.41	0.99 ± 0.44	2.32 ± 0.41	1.58 ± 0.41	1.18 ± 0.42	0.80 ± 0.65	1.49 ± 0.19	Diet, $P > 0.10$
High fat (n = 5)	1.31 ± 0.42	0.83 ± 0.42	1.08 ± 0.42	2.12 ± 0.42	2.78 ± 0.42	1.96 ± 0.46	1.68 ± 0.18	Sample period, $P > 0.10$
Period means	1.68 ± 0.29	0.91 ± 0.30	1.70 ± 0.29	1.85 ± 0.29	1.98 ± 0.30	1.38 ± 0.40	—	Time of sample,[b] $P > 0.10$
								Interactions, $P > 0.10$
Growth hormone, ng/mL								
Low fat (n = 5)	3.70 ± 0.26	2.61 ± 0.26	3.25 ± 0.26	3.80 ± 0.26	4.44 ± 0.26	3.21 ± 0.43	3.50 ± 0.12	Diet, $P > 0.10$
High fat (n = 5)	2.96 ± 0.26	2.90 ± 0.26	2.02 ± 0.26	4.50 ± 0.26	2.94 ± 0.26	3.36 ± 0.30	3.11 ± 0.11	Sample period, $P > 0.10$
Period means	3.33 ± 0.18	2.76 ± 0.18	2.63 ± 0.19	4.15 ± 0.18	3.68 ± 0.18	3.29 ± 0.26	—	Time of sample,[b] $P < 0.001$
								Interactions, $P > 0.10$

[a]Hereford F_1 heifers only.
[b]Sequence of sample collected within period (1 to 17).

number of AI services per pregnancy were not significant, and the effect on final pregnancy percentage approached significance ($P = 0.10$). Hereford-sired heifers had the highest pregnancy percentage (80.6%) and P-sired the lowest (66.6%), and L-sired heifers were intermediate (75.6%). Effects of the diet × sire breed interaction on number of AI services and pregnancy rate were not significant.

The lack of diet effect on pregnancy is of interest. Byerley et al. (1987) reported that breeding heifers at the third postpubertal estrus increased pregnancy rate by 21 percentage points. Staigmiller et al. (1993) concluded this increase was a result of maturation of the pregnancy maintenance mechanisms as the heifer advanced from the first to third estrus. In the present study, more P-sired heifers were pubertal and cycling at the beginning of the breeding season, but these heifers had the lowest pregnancy rate. Studies have shown positive effects of relatively short-term feeding (approximately 8 wk) of supplemental dietary fat to dams on cold tolerance of newborn calves (Lammoglia et al., 1999a; b), ovarian follicular populations (Lucy et al., 1991; De Fries et al., 1998), and pregnancy rates (Grummer and Carroll, 1991; De Fries et al., 1998; Bellows et al., 1999). In the present study, the increase in backfat thickness changed dramatically after 56 d on the dietary treatments (Figure 1). We hypothesize that the cited studies and data from the present work suggest that the 162-d fat-feeding period in the present study may have been excessively long, leading to a dietary fat-reproduction antagonism, and that a feeding period of approximately 60 d before the beginning of the breeding season may have been more suitable. The validity of this hypothesis cannot be determined by results of the present work and awaits further study. Another possibility is that weight changes occurring on pasture following termination of the supplement phase of the study differed between LF and HF heifers. However, body weights at the end of the breeding season were not obtained.

Results of blood analyses for hormone and metabolite concentrations are summarized in Table 5. Diet had a significant effect on progesterone concentrations at d 7 to 10 of the pubertal estrous cycle, and higher progesterone concentrations were found in heifers that received the HF diet. The HF diet also increased ($P < 0.001$) cholesterol concentrations. Because cholesterol is utilized in progesterone synthesis, these differences may represent a cause-and-effect relationship. Grummer and Carroll (1991) and Thomas and Williams (1996) reported greater serum progesterone concentrations in cows supplemented with fats, agreeing with results found in this study. Hawkins et al. (1995) reported that elevated serum progesterone concentrations in cows supplemented with fats resulted from a decrease in progesterone clearance rate from the circulatory system rather than from an increase in progesterone secretion by the CL. Furthermore, Lammoglia et al. (1997) demonstrated that cultured luteal cells from fat-supplemented cows and from cows receiving the control diet responded similarly to a LH challenge. Results of the present study do not allow determination of the cause(s) of the observed increased progesterone concentration.

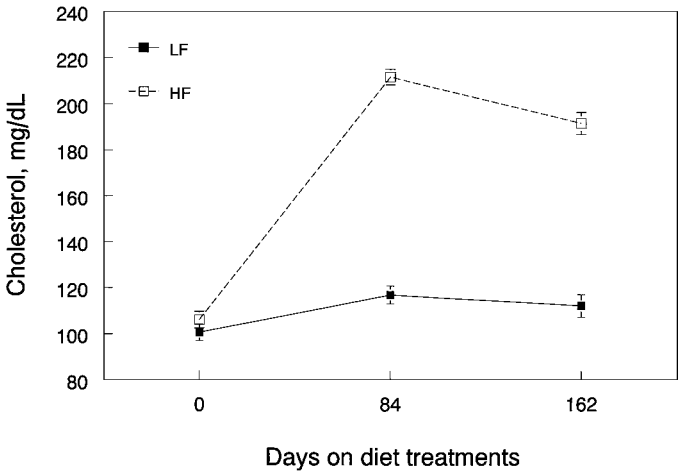

Figure 3. Least squares means plot (pooled SEM = 4.02) of serum cholesterol concentrations of heifers receiving 1.9% (LF) or 4.4% (HF) dietary fat as affected by dietary treatment × days on diet treatment ($P < 0.001$; n = 60).

Additional data on cholesterol changes are shown in Figure 3, which summarizes the diet × days on diet treatment interaction ($P < 0.001$). Heifers fed the HF diet had greater cholesterol concentrations after 84 d of feeding and maintained the higher concentrations throughout the feeding period. The sire-breed effect on progesterone concentrations was not significant, but L-sired heifers tended ($P = 0.06$) to have higher blood cholesterol concentrations than H- or P-sired heifers. Diet × sire breed interaction effects were nonsignificant for progesterone and cholesterol concentrations.

Data obtained from serial bleedings of the 10 Hereford heifers are also summarized in Table 5. Diet and sample period (day of feeding period) did not affect insulin or GH concentrations. Lammoglia et al. (1997) reported elevated blood insulin concentrations in cows receiving HF diets, which disagrees with results of the present study. Feeding cows diets containing high amounts of long-chain fatty acids increased hepatic gluconeogenesis because of increased propionate production in the rumen (Selner and Schultz, 1980; Chalupa et al., 1986; Keele et al., 1989). It may be possible that we did not increase propionate production in the rumen and subsequent gluconeogenesis of heifers receiving the HF ration. It may also be possible that level of stress during the serial bleedings was similar in both groups of heifers, resulting in cortisol release and stimulation of compensatory gluconeogenesis.

The main effect of time of sample collection within the serial bleeding series on GH concentrations was highly significant. These changes are shown in Figure 4 and suggest an episodic GH rise approximately every 4 h, which agrees with work in sheep reported by Driver

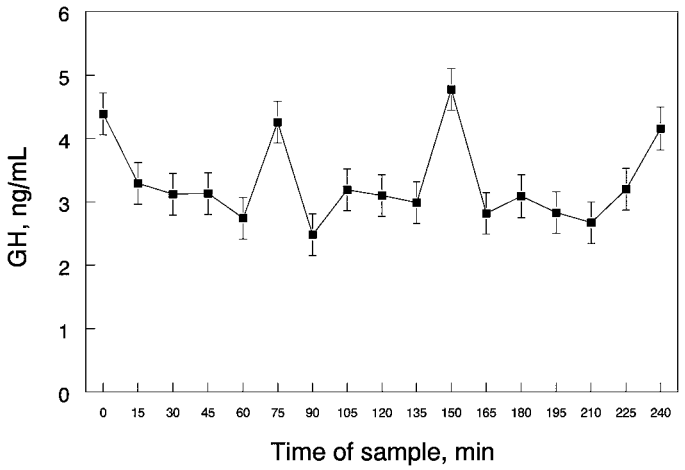

Figure 4. Least squares means plot (pooled SEM = 0.33) of effect of time of sample collection on growth hormone (GH) concentrations ($P < 0.001$; n = 10).

and Forbes (1981). Circulating growth hormone concentrations were similar among dietary groups in this study, which agrees with previous work in cycling beef cows (Lammoglia et al., 1997) and lactating dairy cows (Cummins and Sartin, 1987) but disagrees with results reported by Thomas and Williams (1996). Differences among these studies could have been caused by differences in age of the animals, breed of sire, or fatty acid composition of the diet. Plasma $PGF_{2\alpha}$ concentrations could not be compared among treatments because concentrations were not detectable with the assay used. This result indicates that $PGF_{2\alpha}$ concentrations were low in prepubertal heifers studied, because the detection limit of the assay was 14 to 25 pg/mL (Del Vecchio et al., 1990).

Implications

Feeding 4.4% dietary fat increased the percentage of heifers pubertal by the beginning of the breeding season. However, the diet effect interacted with sire breed, and we hypothesize that the response to supplemental dietary fat may be breed-dependent. Heifers with a low body fat composition may have a dietary fat requirement different from that of heifers with a greater body fat composition and may benefit from supplemental fat. The feeding period in the present study was 162 d in duration. Based on the results of other studies with supplemental fat and backfat changes in the present study, we hypothesize that a feeding period of approximately 60 d before the beginning of the breeding season may be more effective in improving reproduction in replacement heifers.

Literature Cited

Adams, R. S. 1980. Pennsylvania State forage testing service revised regression equations. Dairy Sci. Ext. Memo DSE-80-56. Penn. State Univ., University Park.

Arthur, P. F. 1995. Double muscling in cattle: A review. Aust. J. Agric. Res. 46:1493–1515.

Bellows, R. A., and J. B. Hall. 1996. Physiology and management of the replacement heifer. In: L. M. Rode (ed.) Animal Science Research and Development, Meeting Future Challenges. Proc. Canadian Soc. Anim. Sci. Lethbridge Res. Centre, Lethbridge, Alberta. pp 217–234.

Bellows, R. A., D. D. Simms, E. E. Grings, D. A. Phelps, S. E. Bellows, N. R. Bellows, R. E. Short, R. N. Funston, and T. W. Geary. 1999. Effects of feeding supplemental fat during gestation on reproduction in primiparous beef heifers. J. Anim. Sci. 77 (Suppl. 1):236 (Abstr.).

Bellows, R. A., R. B. Staigmiller, J. M. Wilson, D. A. Phelps, and A. Darling. 1991. Use of bovine FSH for superovulation and embryo production in beef heifers. Theriogenology 35:1069–1082.

Berardinelli, J. G., R. A. Daily, R. L. Butcher, and E. K. Inskeep. 1979. Source of progesterone prior to puberty in beef heifers. J. Anim. Sci. 49:1276–1280.

Buckley, B. A., J. F. Baker, G. E. Dickerson, and T. G. Jenkins. 1990. Body composition and tissue distribution from birth to 14 months for three biological types of beef heifers. J. Anim. Sci. 68:3109–3123.

Byerley, D. J., R. B. Staigmiller, J. B. Berardinelli, and R. E. Short. 1987. Pregnancy rates of beef heifers bred either on puberal or third estrus. J. Anim. Sci. 65:645–650.

Chalupa, W., B. Vecchiarelli, A. Elser, D. S. Kronfeld, D. Sklan, and D. L. Palmquist. 1986. Ruminal fermentation in vivo as influenced by long-chain fatty acids. J. Dairy Sci. 69:1293–1301.

Cummins, K. A., and J. L. Sartin. 1987. Response of insulin, glucagon, and growth hormone to intravenous glucose challenge in cows fed high fat diets. J. Dairy Sci. 70:277–283.

De Fries, C. A., D. A. Neuendorff, and R. D. Randel. 1998. Fat supplementation influences postpartum reproductive performance in Brahman cows. J. Anim. Sci. 76:864–870.

Del Vecchio, R. P., C. C. Chase, Jr., P. Bastidas, and R. D. Randel. 1990. Oxytocin-induced changes in plasma 13,14-dihydro-15-keto-prostaglandin $F_{2\alpha}$ concentrations on days 10, 20 and 30 postpartum in the bovine. J. Anim. Sci. 68:4261–4266.

Del Vecchio, R. P., D. A. Neuendorff, R. C. Stahringer, and R. D. Randel. 1991. The onset of puberty in the bovine: Uterine and ovarian hormonal interrelationships. Domest. Anim. Endocrinol. 8:521–526.

Driver, P. M., and J. M. Forbes. 1981. Episodic growth hormone secretion in sheep in relation to time of feeding, spontaneous meals and short term fasting. J. Physiol. (Lond.) 317:413–424.

Ferrell, C. L. 1982. Effects of postweaning rate of gain on onset of puberty and productive performance of heifers of different breeds. J. Anim. Sci. 55:1272–1283.

Grings, E. E., R. B. Staigmiller, R. E. Short, R. A. Bellows, and M. D. MacNeil. 1999. Effects of stair-step nutrition and trace mineral supplementation on attainment of puberty in beef heifers of three sire breeds. J. Anim. Sci. 77:810–815.

Grummer, R. R., and D. J. Carroll. 1991. Effects of dietary fat on metabolic disorders and reproductive performance of dairy cattle. J. Anim. Sci. 69:3838–3852.

Hall, J. B., R. B. Staigmiller, R. A. Bellows, R. E. Short, W. M. Moseley, and S. E. Bellows. 1995. Body composition and metabolic profiles associated with puberty in beef heifers. J. Anim. Sci. 73:3409–3420.

Hawkins, D. E., K. D. Niswender, G. M. Oss, C. L. Moeller, K. G. Odde, H. R. Sawyer, and G. D. Niswender. 1995. An increase in serum lipids increases luteal lipid content and alters the disappearance rate of progesterone in cows. J. Anim. Sci. 73:541–545.

Hightshoe, R. B., R. C. Cochran, L. R. Corah, G. H. Kiracofe, D. L. Harmon, and R. C. Perry. 1991. Effects of calcium soaps of fatty acids on postpartum reproductive function in beef cows. J. Anim. Sci. 69:4097–4103.

Hoefler, W. C., and D. M. Hallford. 1987. Influence of suckling status and type of birth on serum hormone profiles and return to estrus

in early- postpartum spring-lambing ewes. Theriogenology 27:887–895.

Karima, A. S., and R. T. Berg. 1985. Fat growth and partitioning among the depots in double muscled and normal cattle. Can. J. Anim. Sci. 65:295–306.

Keele, J. W., R. E. Roffler, and K. Z. Beyers. 1989. Ruminal metabolism in nonlactating cows fed whole cottonseed or extruded soybeans. J. Anim. Sci. 67:1612–1622.

Lammoglia, M. A., R. A. Bellows, E. E. Grings, and J. W. Bergman. 1999a. Effects of prepartum supplementary fat and muscle hypertrophy genotype on cold tolerance in newborn calves. J. Anim. Sci. 77:2227–2233.

Lammoglia, M. A., R. A. Bellows, E. E. Grings, J. W. Bergman, R. E. Short, and M. D. MacNeil. 1999b. Effects of feeding beef females supplemental fat during gestation on cold tolerance in newborn calves. J. Anim. Sci. 77:824–834.

Lammoglia, M. A., S. T. Willard, D. M. Hallford, and R. D. Randel. 1997. Effects of dietary fat on follicular development and circulating concentrations of lipids, insulin, progesterone, estradiol-17β, 13, 14-dihydro-15-keto-prostaglandin-$F_{2\alpha}$ and growth hormone in estrous cyclic Brahman cows. J. Anim. Sci. 75:1591–1600.

Lucy, M. C., C. R. Staples, F. M. Michael, and W. W. Thatcher. 1991. Effect of feeding calcium soaps to early postpartum dairy cows on plasma prostaglandins $F_{2\alpha}$, luteinizing hormone, and follicular growth. J. Dairy Sci. 74:483–489.

Martin, L. C., J. S. Brinks, R. M. Bourdon, and L. V. Cundiff. 1992. Genetic effects on beef heifer puberty and subsequent reproduction. J. Anim. Sci. 70:4006–4017.

NRC. 1984. Nutrient Requirements of Beef Cattle. 6th ed. National Academy Press, Washington, DC.

Palmquist, D. L. 1994. Metabolism of fats and their role in animal efficiency. J. Nutr. 124:1377–1381.

Patterson, D. J., R. C. Perry, G. H. Kiracofe, R. A. Bellows, R. B. Staigmiller, and L. R. Corah. 1992. Management considerations in heifer development and puberty. J. Anim. Sci. 70:4018–4035.

Rhodes, R. C., III, M. M. McCartor, and R. D. Randel. 1978. Effects of feeding protein-protected lipid upon growth and reproductive development of yearling heifers. J. Anim. Sci. 46:769–777.

Rutter, L. M., and R. D. Randel. 1986. Nonpuberal estrus in heifers. J. Anim. Sci. 63:1049–1053.

Ryan, D. P., R. A. Spoon, and G. L. Williams. 1992. Ovarian follicular characteristics, embryo recovery, and embryo viability in heifers fed high-fat diets and treated with follicle-stimulating hormone. J. Anim. Sci. 70:3505–3513.

Sanson, D. W., and D. M. Hallford. 1984. Growth response, carcass characteristics and serum glucose and insulin in lambs fed tolazamide. Nutr. Rep. Int. 29:461–466.

SAS. 1994. SAS User's Guide: Statistics (Version 6.10). SAS Inst. Inc., Cary, NC.

Selner, D. R., and L. H. Schultz. 1980. Effects of feeding oleic acid or hydrogenated vegetable oils to lactating cows. J. Dairy Sci. 63:1235–1241.

Smith, S. B., T. Jenkins, and R. L. Prior. 1987. Carcass composition and adipose tissue metabolism in growing sheep. J. Anim. Sci. 65:1525–1530.

Staigmiller, R. B., R. A. Bellows, R. E. Short, M. D. MacNeil, J. B. Hall, D. A. Phelps, and S. E. Bartlett. 1993. Conception rates in beef heifers following embryo transfer at the pubertal or third estrus. Theriogenology. 39:315 (Abstr.).

Thomas, M. G., and G. L. Williams. 1996. Metabolic hormone secretion and FSH induced superovulatory responses of beef heifers fed dietary fat supplements containing predominantly saturated or polyunsaturated fatty acids. Theriogenology 45:451–458.

Velez, J. S., R. D. Randel, and D. A. Neuendorff. 1991. Effect of uterine manipulation on postpartum fertility and plasma 13,14-dihydro-15-prostaglandin-$F_{2\alpha}$ in Brahman cows and first calf heifers. Theriogenology 36:987–998.

Wehrman, M. E., T. H. Welsh, Jr., and G. L. Williams. 1991. Diet-induced hyperlipemia in cattle modifies intrafollicular cholesterol environment, modulates ovarian follicular dynamics, and hastens the onset of postpartum luteal activity. Biol. Reprod. 45:514–523.

Williams, G. L. 1989. Modulation of luteal activity in postpartum beef cows through changes in dietary lipid. J. Anim. Sci. 67:785–793.

Wu, Z., and J. T. Huber. 1994. Relationship between dietary fat supplementation and milk protein concentration in lactating cows: A review. Livest. Prod. Sci. 39:141–149.