

# Effect of Fish Meal and Expeller-Processed Soybean Meal Fed to Dairy Cows Receiving Bovine Somatotropin (Sometribove)<sup>1</sup>

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## ABSTRACT

Forty-eight multiparous cows were blocked by calving date and milk production and assigned randomly to a TMR formulated to contain 68 or 55% of dietary CP as ruminally degradable CP. Diets contained corn silage, alfalfa haylage, and ground corn. Supplemental CP was soybean meal for the control diet or a combination of soybean meal, expeller-processed soybean meal, and fish meal for the low degradable protein diet. Two 10-wk phases began on d  $31 \pm 3$  (phase 1) and  $110 \pm 7$  postpartum [phase 2, all cows received subcutaneous implantations of pelleted (400 mg) bST (sometribove) every 14 d]. Dietary energy, CP, ruminally degradable CP, NDF, and ADF were similar between dietary treatments. Production of FCM increased in response to bST but was not affected by dietary treatment. Cows fed the expeller-processed soybean meal and fish meal TMR produced milk that contained less milk fat in phase 1 and less milk protein content in both phases. The DMI, BW,

and body condition scores were not affected by diet. Hematocrit, plasma urea N, albumin, total protein, creatinine, glucose, and serum insulin were similar between dietary treatments. Replacing soybean meal with expeller processed soybean meal and fish meal did not affect ruminal degradation of protein or milk production but decreased milk fat and protein contents.

(Key words: fish meal, expeller soybean meal, lactation, somatotropin)

Abbreviation key: ES = expeller-processed soybean meal, FM = fish meal, SBM = soybean meal.

## INTRODUCTION

High producing dairy cows in early lactation may be in negative nutrient balance. Maximum milk production can be achieved through mobilization of body tissue stores. During early lactation, 15 to 60 kg of lipid and 0 to 15 kg of protein can be mobilized to support the metabolic demands of milk production (8, 19). Although cows can rely to some extent on body fat reserves as an energy source during early lactation, most of their protein requirement must be provided through the diet. During short-term administration of bST, lactating dairy cows experience a metabolic situation similar to that of high production in early lactation. Therefore, feeding proteins that are resistant to ruminal degradation should be beneficial both to lactating cows in early lactation and to those treated with bST (20).

Heat-treated soybean meal (SBM) has been used frequently as a source of ruminally undegradable protein, but improvements in cow

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performance have been inconsistent (13, 26). To maximize production, the proportion of dietary protein that escapes ruminal degradation and its AA profile should be considered (11). Feeding combinations of low degradable proteins with complementary AA profiles may provide a practical way to manipulate the quantity and quality of proteins reaching the small intestine (9, 10, 27). The proteins contained in fish meal (FM) are of high quality and are resistant to ruminal degradation (10, 11). Because the AA profile of FM is rich in Lys and sulfur AA, inclusion of FM in diets for dairy cows may improve the quality of the protein delivered to the small intestine and thereby enhance production.

The objective of the present study was to examine effects of partially replacing SBM with expeller-processed soybean meal (ES) and FM on lactational performance and plasma component profiles of dairy cows in early lactation and during bST administration.

**MATERIALS AND METHODS**

**Diets**

Two experimental diets were formulated to contain different amounts of ruminally undegradable protein (7) and to meet or exceed NRC (23) recommendations for cows producing 41 kg of 3.5% FCM (Table 1). Diets contained alfalfa haylage, corn silage, corn grain, animal fat, and a vitamin and mineral mix. Source of supplemental protein in the control diet was SBM. Menhaden FM (Zapata Haynie Co., Hammond, LA) and ES (SoyPlus®, West Central, Ralston, IA) partially replaced SBM as the source of supplemental protein in the ES-FM diet.

**Cows and Management**

Multiparous Holstein cows (n = 48) were housed in a loose housing, deep-pack barn at the University of Minnesota Rosemount Experiment Station. Feed was distributed individually to each cow (Calan® doors, American Calan, Inc., Northwood, NH) as a TMR once daily between 0800 and 1000 h. Cows had access to feed and water throughout the day except during milking at 1030 and 2230 h. Amounts offered and refused were recorded

TABLE 1. Ingredient and chemical composition of diets.

Composition and analysis	Diet <sup>1</sup>	
	Control	ES-FM
	— (% of DM) —	
Ingredient composition		
Alfalfa haylage	22.6	22.6
Corn silage	28.4	28.5
Ground corn	28.9	30.4
Soybean meal	15.9	2.3
Expeller soybean meal	. . .	9.0
Fish meal	. . .	3.1
Animal fat	2.0	2.0
Minerals and vitamins <sup>2</sup>	2.2	2.1
Chemical analyses		
NE <sub>L</sub> , <sup>3</sup> Mcal/kg	1.74	1.74
CP, %	19.7	19.7
ADF, %	19.9	19.4
Ether extract, %	5.4	5.9
Ca, %	.92	1.01
P, %	.51	.57

<sup>1</sup>Diets contained soybean meal (control) or a combination of soybean meal, expeller-processed soybean meal, and fish meal (ES-FM).

<sup>2</sup>Provided 8040, 2480, and 31 IU/kg of supplemental vitamins A, D, and E, respectively.

<sup>3</sup>Estimated from NRC (23).

daily. During the first 4 wk of lactation, all cows were fed the control diet. At 31 ± 3 DIM, cows were blocked by calving date and average daily milk production during d 20 to 26 postpartum and assigned randomly within blocks to either control or ES-FM diets (phase 1; 31 to 101 ± 3 DIM). In phase 2 of the study (110 to 180 ± 7 DIM), all cows continued on their respective dietary treatment and received subcutaneous implantations (400 mg every 14 d) of pelleted, recombinantly derived methionyl bST (sometribove, Monsanto Co., St. Louis, MO). Site of implantation was posterior to the shoulder and was rotated from right to left side and from upper to lower section of the shoulder. Implantation sites were inspected weekly. Cows were observed daily for general health characteristics, and all incidences were recorded.

**Sampling and Measurements**

Weekly feed samples were composited during 4-wk intervals and analyzed by AOAC methods (2) for DM, OM, CP, and ether extract. Fiber components (ADF and NDF) were

analyzed by the detergent system (34). All feed analyses were conducted by the Northeast DHI Cooperative Forage Testing Laboratory (Ithaca, NY). Milk samples were obtained weekly at two consecutive a.m. and p.m. milkings and analyzed individually by infrared spectrophotometry for fat, protein, and lactose content and by fluorescent detection of ethidium bromide incorporation in white blood cell DNA for SCC (Minnesota State DHIA, Zumbrota, MN). Body weights and body condition scores (36) were determined weekly.

Jugular blood samples were obtained by venipuncture using 18-gauge  $\times$  2.5-cm needles (Becton Dickinson and Co., Rutherford, NJ) and either monojet tubes containing K-EDTA (Sherwood Medical, St. Louis, MO) for plasma or vacutainer serum separation tubes (Becton Dickinson and Co., Rutherford, NJ) for serum. Plasma and serum samples were collected 7 and 4 d prior to phase 1 ( $24$  and  $27 \pm 3$  DIM) and every second Friday during phase 1 (41, 55, 69, 83, 94, and  $97 \pm 3$  DIM). During phase 2 (110 to  $180 \pm 7$  DIM), plasma and serum samples were obtained on d 1, 7, and 14 of the second and fifth bST implantation cycles (124, 131, 138, 166, 173, and  $180 \pm 7$  DIM). Hematocrit of blood samples containing the anticoagulant was determined within 30 min after sampling. Plasma and serum were obtained from their respective samples by centrifugation ( $4000 \times g$  for 15 min), frozen in dry ice, and stored at  $-20^{\circ}\text{C}$  until analyzed. Plasma samples were analyzed for total protein, albumin, urea N, creatinine, and glucose concentrations on an automated clinical analyzer (Dimension Analyzer, DuPont, Wilmington, DE). Serum insulin concentration was determined with a solid support radioimmunoassay system (Diagnostic Products Co., Los Angeles, CA).

#### Statistical Analyses

Cows were blocked (eight blocks) by calving date (56-d maximum interval) and average milk production during d 20 to 26 of lactation (maximum range of 9.1 kg/d) and were assigned randomly within blocks to dietary treatments. Distribution of actual calving dates and milk production of available cows and udder injury caused the removal of 4 cows (final  $n = 21$  and 23 cows for control and ES-FM diets, respectively). Effects of diet on production-

related data in phases 1 and 2 were assessed separately by analysis of covariance (covariate data collected during d 15 to 28 postpartum) in a randomized, incomplete block design. Within each phase, effects of diet on blood component concentrations were assessed by repeated measures analyses. All statistical evaluations were conducted through the general linear models procedure of SAS (28). Significance was detected at  $P < .05$  unless otherwise noted. All reported means are least squares means adjusted for pretreatment differences (covariate) unless otherwise noted.

## RESULTS

Composition of diets was similar for all analyzed components (Table 1) and met or exceeded NRC (23) recommendations for lactating dairy cows in the present experiment. Fish meal, SBM, and ES contained 9.9, .9, and 5.4% ether extract, respectively. Diets were formulated to contain 18.5% CP, but, because of the dry conditions of the year, protein content of alfalfa was higher than expected (18). Diets were formulated to contain 68 and 55% degradable protein, but estimates from a continuous culture system indicate ruminal degradabilities of 60.8 and 59.8% for control and ES-FM diets, respectively (7).

Milk and 3.5% FCM production and fat, protein, and lactose yields were not affected by dietary treatments in either phase 1 or 2 (Table

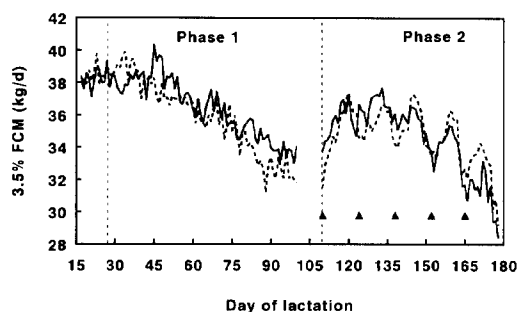


Figure 1. Least squares means for 3.5% FCM of cows consuming diets containing soybean meal (---) or a combination of soybean meal, expeller-processed soybean meal, and fish meal (—) as protein supplements during early lactation (phase 1) and during administration of a sustained-release form of bST (phase 2). Cows received 400 mg of bST/14 d, as indicated (▲).

TABLE 2. Effects of diets containing soybean meal (control) or a combination of soybean meal, expeller-processed soybean meal, and fish meal (ES-FM) on least squares means for milk production and composition in early lactation (phase 1) and during bST administration (phase 2).

Item	Phase 1 <sup>1</sup>			Phase 2 <sup>2</sup>		
	Control	ES-FM	SE	Control	ES-FM	SE
Milk production, kg/d	35.6	36.7	.8	33.0	33.3	1.1
3.5% FCM, kg/d	35.9	36.4	.9	34.7	34.5	1.2
Fat						
%	3.65 <sup>a</sup>	3.43 <sup>b</sup>	.07	3.9	3.75	.08
kg/d	1.27	1.27	.04	1.26	1.24	.05
Protein						
%	3.22 <sup>a</sup>	3.13 <sup>b</sup>	.02	3.36 <sup>a</sup>	3.26 <sup>b</sup>	.04
kg/d	1.12	1.15	.03	1.09	1.09	.04
Lactose						
%	4.97	4.97	.03	4.91	4.90	.05
kg/d	1.76	1.84	.05	1.62	1.65	.07
SCC, log <sub>10</sub>	5.27	5.26	.11	5.31	5.45	.11

<sup>a,b</sup>Means with different superscripts within a phase in the same row differ ( $P < .05$ ).

<sup>1</sup>Phase 1: 31 to 100 ± 3 DIM.

<sup>2</sup>Phase 2: 110 to 180 ± 7 DIM. All cows received 400 mg of bST/14 d.

2, Figure 1). In phase 1 (31 to 100 ± 3 DIM), average milk production of cows consuming ES-FM diets was 1.1 kg/d more than for cows consuming control diets, but differences were not significant. Production of 3.5% FCM was similar between dietary treatments in both phases, because fat content of milk was .22 percentage units less in cows fed ES-FM in phase 1 and not significantly decreased in phase 2 (Table 2). Milk protein content decreased in phases 1 and 2 (Table 2) when cows consumed ES-FM diets. Lactose concentration and SCC were not affected by dietary treatment.

Persistencies of the mean lactation curves of cows fed control or ES-FM diets from peak production to 101 ± 3 DIM (phase 1) were similar (-.70 and -.66 kg of 3.5% FCM/wk, respectively). The milk production anticipated if cows had not been treated with bST during phase 2 was calculated assuming that the persistencies of cows on control and ES-FM diets during phase 1 continued during phase 2. Actual milk production of cows during phase 2 (bST treatment phase) was 17.6% greater (an increase of 5.9 and 4.4 kg/d of 3.5% FCM for cows on control and ES-FM diets, respectively) than the milk production calculated from persistency estimates (28.8 and 30.1 kg/d of 3.5% FCM for cows on control and ES-FM diets, respectively).

Milk production response to bST administration was not affected by dietary treatment. Milk production response to the sustained-release form of bST within the five 14-d implantation cycles followed a cyclic pattern (Figure 2), peaking at about d 8 of the cycle. In contrast, milk production was relatively constant within similar 14-d intervals during phase 1 (35.9 and 36.4 ± .8 kg/d of FCM, for the control and ES-FM dietary treatments, respectively).

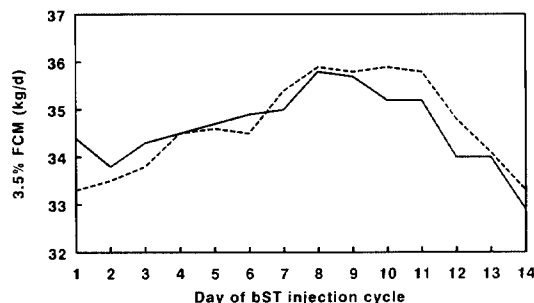


Figure 2. Least squares means for daily milk production within bST implantation cycle (n = 5 cycles) by cows receiving 400 mg of bST/14 d and consuming diets containing soybean meal (---) or a combination of soybean meal, expeller-processed soybean meal, and fish meal (—) as protein supplements.

Feed intake, BW, and body condition scores (Table 3) were not affected by dietary treatments in either phase 1 or 2 and tended to increase as lactation proceeded. Calculated feed efficiencies also were similar between dietary treatments.

All analyzed blood components were within normal physiological ranges (15) and were not affected by dietary treatment (Figures 3 and 4). Overall mean concentrations during phases 1 and 2 were 30.2 and 31.4% for hematocrit, 7.9 and 7.9 g/dl for plasma total protein, 3.45 and 3.51 g/dl for albumin, 20.5 and 19.3 mg/dl for urea N, .914 and .946 mg/dl for creatinine, 67.6 and 71.9 mg/dl for glucose, and 15.6 and 21.8  $\mu$ IU/ml for serum insulin, respectively.

Within bST implantation cycles, plasma urea N and glucose concentrations and serum insulin concentrations varied cyclically. Means for d 1, 7, and 14 of the second and fifth bST implantation cycles were 20.0, 18.4, and 19.4 mg/ml for plasma urea N and 20.1, 25.2, and 20.2  $\mu$ IU/ml for serum insulin. Plasma urea N decreased and serum insulin increased ( $P < .06$ ) on d 7 compared with values on either d 1 or 14 of the cycle.

No abnormal health characteristics were noted during either phase for cows on control or ES-FM diets (data not presented). Inspection of implantation site indicated occasional local and transient swelling.

#### DISCUSSION

Based on pre- and posttrial estimates on the degradable protein content of SBM, ES, and FM (7) and on NRC (23) estimates of other dietary components, degradable protein content of control and ES-FM diets should have differed (68.3 vs. 57.3% of dietary protein for control and ES-FM diets, respectively) by 11 percentage units (7). However, a continuous culture study conducted after completion of the lactation study indicated that the degradable protein contents of control and ES-FM diets were 60.8 and 59.8%, respectively (7). Crude protein degradability of dietary components and TMR were evaluated further by the in situ, ficin, and ammonia release techniques (7). Results from those evaluations (7) demonstrated that ES and FM proteins were, indeed, less degradable than SBM and that degradable protein content of the concentrate portion of

control and ES-FM diets agreed with values calculated from the degradable protein content of the respective ingredients. However, analyses of the TMR by those additional techniques confirmed only small differences between dietary CP degradability of control and ES-FM. If control and ES-FM diets had been prepared improperly at different points during the study,

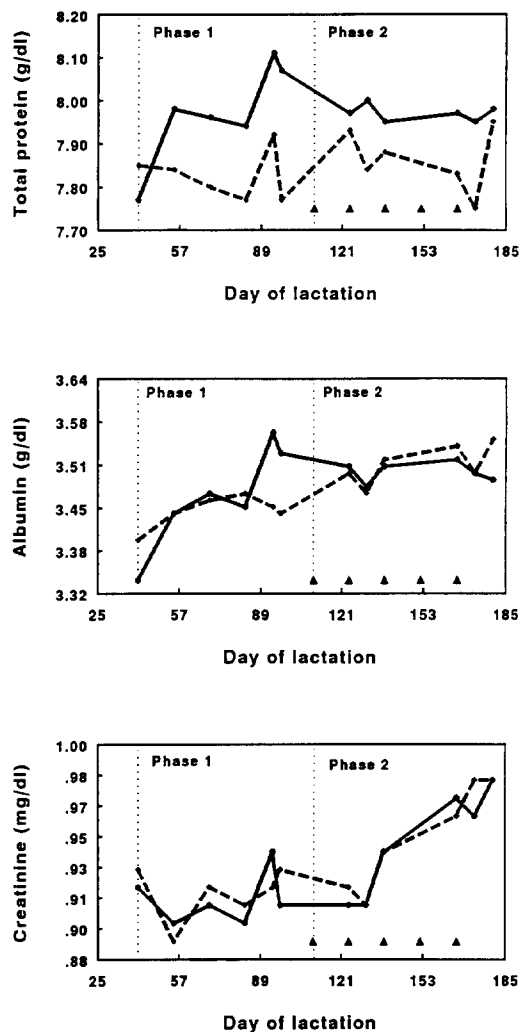


Figure 3. Effects of diets containing soybean meal (- - ♦ - -) or a combination of soybean meal, expeller-processed soybean meal, and fish meal (—●—) as protein supplements on least squares means for plasma total protein, albumin, and creatinine concentrations in early lactation (phase 1) and during bST administration (phase 2). Cows received 400 mg of bST/14 d, as indicated (▲).

TABLE 3. Least squares means for BW, body condition score, and feed efficiency of dairy cows fed diets containing soybean meal (control) or a combination of soybean meal, expeller-processed soybean meal, and fish meal (ES-FM) in early lactation (phase 1) and during bST administration (phase 2).

Item	Phase 1 <sup>1</sup>			Phase 2 <sup>2</sup>		
	Control	ES-FM	SE	Control	ES-FM	SE
DMI, kg/d	22.9	22.2	.4	23.4	22.3	.6
BW, kg	650	649	4	697	706	7
Body condition score	3.1	3.1	<.1	3.3	3.3	<.1
Apparent feed efficiency, FCM/DMI	1.58	1.62	.03	1.50	1.53	.04
Gross feed efficiency, FCM/NEI <sup>3</sup>						
Observed	1.01	.98	.01	.94	.95	.02
Corrected <sup>4</sup>	1.06	1.07	.02	1.00	1.02	.02

<sup>1</sup>Phase 1: 31 to 100 ± 3 DIM.

<sup>2</sup>Phase 2: 110 to 180 ± 7 DIM. All cows received 400 mg of bST/14 d.

<sup>3</sup>FCM per total net energy intake.

<sup>4</sup>Corrected for BW changes by the formula: FCM/[NE<sub>L</sub> - (5.12 Mcal/kg × BW gain)]; FCM = 3.5% FCM (kilograms per day), NEI = net energy intake (megacalories per day), and BW gain (kilograms per day).

the consistently lower protein and fat content of milk from cows fed ES-FM (Table 2) would not be expected. Therefore, errors in mixing the TMR appear unlikely.

The lack of difference between ruminal degradabilities of protein in control and ES-FM diets has no clear explanation. Although most reports indicate that dietary degradability can be predicted accurately from individual ingredient analyses (22, 32), others, as summarized by Calsamiglia et al. (7), have reported discrepancies between estimated ruminal degradability based on analyses of individual ingredients and actual measurements of TMR. Those results indicate that estimates of ruminal degradability obtained from TMR probably are better indicators of protein degradability than estimates obtained from individual ingredients.

It is not clear from our results whether the discrepancy between measured ruminally degradable protein content of the diet and that calculated from analyses of individual ingredients is due to altered degradation of forage, concentrate, or both (7). Because both TMR had high protein content and similar CP degradability, amounts of degradable and undegradable protein in control and ES-FM diets were equal to or above NRC (23) recommendations for cows in the present study (mean production of 35.4 kg/d of FCM) and likely contributed to the lack of a dietary effect on milk production.

Although total milk fat yield was not affected by dietary treatment in either phase 1 or 2, fat content of milk from cows fed ES-FM decreased in phase 1 and tended to decrease in phase 2 compared with controls (Table 2). Total dietary fat content was not excessive (17) in either TMR, and the small difference in dietary ether extract content between control and ES-FM diets (5.4 vs. 5.9%, respectively) most likely was insufficient to cause the observed changes in milk fat content.

Opstvedt (25) summarized effects of fish oils on milk production and composition and concluded that effects on milk fat content were dependent on the net intake of fish oils. Decreased milk fat content occurred when intakes of fish oil exceeded 38 g/d, and reduced total fat yield occurred only when intake of fish oil exceeded 100 g/d (25). In the present study, intake of fish oil by the cows consuming the ES-FM diet averaged 68.2 g/d. Production responses (decreased fat percentage, but no change in fat yield) in the present study agree with those anticipated from the regression analysis of Opstvedt (25). In addition, others (14, 31) have indicated that feeding small amounts of the long-chain polyunsaturated fatty acids in FM to cows may decrease milk fat content. We calculated that about 75% of fat from FM and ES was in the form of unsaturated fatty acids.

In phases 1 and 2, milk protein content, but not milk protein production, was reduced when

the ES-FM diet was fed (Table 2). Nonesterified fatty acids and unsaturated fatty acids have been reported to decrease milk protein content (29). Broderick (5) reported decreased milk protein content when ES replaced SBM (12.4% of dietary DM). However, the SBM control diet used by Broderick (5) also contained added soybean oil to compensate for differences in oil content. Therefore, depressed milk protein was not dependent solely on the amount of soybean oil but probably involved a more complex interaction. In contrast, no change in milk protein content was reported when ES provided less than 6% of dietary DM (5, 6). Dairy cows fed FM have responded with both increased (21) and decreased (31) milk protein content.

Feed intake, feed efficiency, BW, and body condition scores were not affected by dietary treatment (Table 3). Broderick (5) and Broderick et al. (6) reported that, compared with cows consuming diets containing SBM, those consuming diets containing ES consumed less feed but produced similar amounts of milk. They (5, 6) attributed the improved feed efficiency of the cows fed ES to an increased amount of dietary protein escaping ruminal degradation and an improved AA profile of digesta entering the small intestine. The lack of response in the present study suggests either that dietary protein was not adequately protected from ruminal degradation or that the quantity and quality of AA reaching the small intestine of control cows were not limiting production.

The estimated increase (17.5%) in milk production as a result of bST administration (Figure 1) agrees with previous reports [e.g., (3)]. Milk production response within bST implantation interval followed a cyclic pattern, reaching a maximum between d 7 through 11 (Figure 2). Milk production on d 1, 9, and 14 of each bST implantation cycle averaged 33.8, 35.8, and 33.1 kg/d of FCM. This cyclic response agrees with the observation of Bauman et al. (3) despite the difference (pelleted bST vs. an oil-based formulation) between the prolonged-release products. Although the specific dynamics of bST release from the pelleted and oil-based products may differ, the results suggest that neither formulation provides a constant delivery throughout the intended administration interval. During the last

one-third of the administration interval, delivery of bST was insufficient to maintain the response in milk production.

Concentrations of blood components in the present study were within normal physiologi-

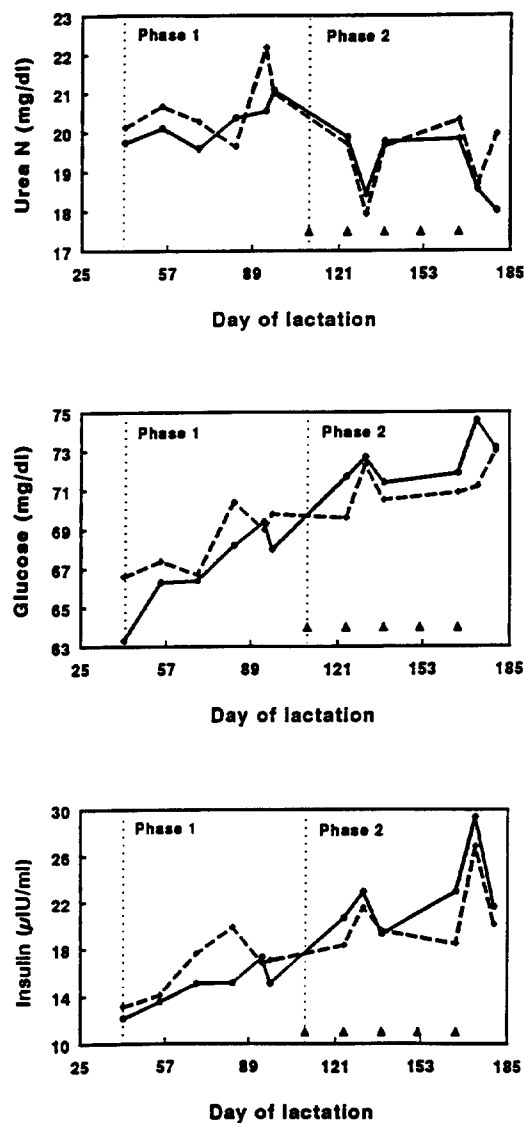


Figure 4. Effects of diets containing soybean meal (---●---) or a combination of soybean meal, expeller-processed soybean meal, and fish meal (—●—) as protein supplements on least square means for plasma urea N and glucose concentrations and serum insulin concentrations in early lactation (phase 1) and during bST administration (phase 2). Cows received 400 mg of bST/14 d, as indicated (▲).

cal ranges (15) and were not affected by dietary treatments. Hematocrit in both control and ES-FM cows tended to increase with lactation, which agrees with the inverse relationship between hematocrit and milk production and between hematocrit and stage of lactation (33). Although bST administration decreased hematocrit (30, 35), this effect probably was due to increased milk production. In agreement with other reports (16), total plasma protein and albumin concentrations were not affected by stage of lactation. The lower albumin concentration in early lactation probably resulted from increased demands for protein by the mammary gland and a dilution effect resulting from increased blood volume (33). Although Larson (16) indicated that creatinine concentrations were unaffected by stage of lactation or bST administration, creatinine in the present study increased in cows fed control and cows fed ES-FM diets during phase 2. The design of the experiment precludes discerning whether increased creatinine during phase 2 was an effect of bST administration, advancing stage of lactation, or both. However, values were within normal ranges, and changes probably were of no biological significance.

Plasma urea N responded in a cyclic pattern to bST implantation; apparent nadir occurred midcycle. Vicini et al. (35) suggested that reduction of plasma urea N during bST administration may reflect reduced hepatic oxidation of AA and increased mammary gland uptake to support synthesis of milk proteins. Consistent with observations by others (1, 4), serum insulin and plasma glucose concentrations increased as lactation progressed. Effects of bST administration on serum insulin concentration are variable (12, 24). Disagreement among reports is probably the result of differences among factors that affect insulin concentration, including stage of lactation, energy balance, and time of sampling in relation to feeding.

Within bST implantation cycles, insulin concentration varied cyclically, peaking ( $P < .06$ ) midcycle, and returning to basal concentration at the end of the 14-d period. Glucose concentration within bST implantation cycles followed the same cyclic pattern as insulin, although differences were not significant ( $P = .18$ ). Oldenbrok et al. (24) also examined blood component profiles in cows administered a 2-wk bST product, but their monthly

sampling did not allow examination of within-cycle responses. Shifts in glucose metabolism (increased glucose and insulin concentrations in blood) within implantation cycle likely reflect coordinated attempts to increase glucose availability to the mammary gland. This occurs, in part, through decreased sensitivity of peripheral tissues to insulin, which provides an advantage to the relatively insulin-insensitive mammary gland (35).

### CONCLUSIONS

Inclusion of protein supplements resistant to ruminal degradation in the diet of lactating dairy cows did not reduce protein degradability of TMR. Although no clear explanation is available, results suggest that associative effects may occur when individual ingredients are combined in TMR. Partial substitution of SBM by ES and FM did not alter milk FCM production or fat, protein, or lactose yields, but fat and protein contents of milk decreased. Blood component profiles were unaffected by dietary treatment. Administration of bST increased milk production cyclically but did not alter response to dietary treatments. Within bST implantation cycles, plasma glucose and urea N concentrations and serum insulin concentrations cycled in a pattern consistent with milk production response.

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