Protected Methionine Supplementation with Extruded Blend of Soybeans and Soybean Meal for Dairy Cows[1]

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ABSTRACT

Methionine may be the first amino acid limiting milk production in early lactation cows. To evaluate this further, 23 high producing Holstein cows (9 multiparous and 14 primiparous) were fed an extruded blend of soybeans and soybean meal (40:60) without or with 15 g of added DL-methionine as 50 g of ruminally protected methionine product during wk 4 to 16 postpartum. Cows were fed a 15.8% crude protein total mixed ration consisting of 30% (dry basis) corn silage, 15% alfalfa hay, and 55% concentrate mix. Covariant-adjusted yields of milk (35.3 and 33.9 kg/d) and solids-corrected milk (29.3 and 28.2 kg/d) were lower for cows fed ruminally protected methionine, whereas yields of 4% fat-corrected milk (28.2 and 27.4 kg/d) were similar. Percentages of fat (2.68 and 2.69) and solids-not-fat (8.82 and 8.83) were similar, and percentages of protein (2.86 and 2.90) were higher from cows fed supplemental methionine. Dry matter intakes (20.5 and 21.6 kg/d) were higher for cows fed ruminally protected methionine. Methionine concentrations in arterial and venous serum were elevated slightly by feeding supplemental methionine. Although methionine was still the first-limiting amino acid as calculated by two different methods, supplementation of this diet with ruminally protected methionine did not increase production of early lactation cows.

INTRODUCTION

Increased milk yields occur with postruminal administration of proteins and amino acids (5, 8, 11, 13, 25, 29, 30). Also, the feeding of protein of low ruminal degradability has led to increases in milk production during early lactation (15, 18, 26, 27). These findings suggest that deficiencies of certain amino acids may be limiting peak milk production in high producing cows during early lactation. Methionine has been suggested (1, 6, 8, 10, 28, 29, 33) as the first-limiting amino acid for milk protein synthesis; however, direct evidence is limited. Fisher (11) reported an increase in the milk protein percentage when 8 g/DL-methionine was administered intravenously. Illg et al. (13) reported approximately a 7% increase in milk production and a 2% relative increase in the percentage of milk protein by supplementing 15 g/d of ruminally protected DL-methionine, but no effect was seen for other milk components. In contrast, intravenous infusion of methionine (8) or the feeding of a ruminally protected methionine source (5, 22, 23, 30, 33) did not increase the production of milk, milk fat, or milk proteins. The inconsistency of responses to supplemental protected methionine may indicate that methionine is not always the first-limiting amino acid or that other amino acids or factors may be colimiting or affecting the physiological utilization of methionine for milk production.

In early lactation, milk production is often limited by energy intake and the addition of fat is one way to raise the caloric density (20). Extruded whole soybeans contain approximately 21% ether extract and may be a more consistent and economical way to raise the caloric density of a ration. In addition, the extrusion process increases the amount of protein escaping degradation in the rumen

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TABLE 1. Ingredient content of total mixed rations.

Ingredient		Diet
		(% of dry matter)
Concentrate mix[1]		55.0
	(% of mix)	
Corn, ground shelled	63.2	
Extruded soybeans-soybean meal[2]	35.0	
Dicalcium phosphate	1.3	
Trace mineral salt	.5	
Alfalfa hay, chopped		15.0
Corn silage		30.0

[1] Plus 4840 IU of vitamin A, 968 IU of vitamin D, and .48 IU of vitamin E added per kilogram of concentrate mix.

[2] Extruded blend of whole soybeans (40%) and soybean meal (60%), (Triple F Feeds, Des Moines, IA).

without reducing quality of protein reaching gastrointestinal absorption sites (1, 15, 18, 27). Ruegsegger and Schultz (26) reported higher milk production for cows fed heat-treated whole soybeans in early lactation. Mielke and Schingoethe (17) observed no benefit when feeding extruded soybeans to cows beyond peak production. Milk production was increased by feeding heat-treated soybean meal to cows in early lactation (15, 18, 27).

Methionine is the first limiting amino acid in soybean meal and remained first limiting in heat-treated soybean meal (1, 27, 28). Supplementation of an extruded blend of whole soybeans and soybean meal with ruminally protected methionine may allow additional increases in milk production. Our objective was to measure the response of early lactating high producing dairy cows receiving a ration containing an extruded blend of whole soybeans and soybean meal without or with supplemental ruminally protected DL-methionine product.

MATERIALS AND METHODS

A 13-wk lactation trial utilized 23 Holstein cows (9 multiparous and 14 primiparous) randomly assigned at approximately 3 wk postpartum to one of two treatments to measure the lactations response of supplementing a ruminally protected methionine source to cows fed a low degradable protein supplement. Multiparous cows were yielding at least 27 kg and primiparous cows at least 23 kg milk/d before assignment to the experiment. Treatment 1 (ESB) utilized an extruded blend of 60% soybean meal and 40% soybeans as the major protein supplement. Treatment 2 (ESB + Met) was ESB plus 50 g of ruminally protected methionine product[3] blended at the time of feeding, which provided 15 g of additional DL-methionine daily. Daugaard (9) found that 80% of the methionine in Ketionin passed the rumen undegraded, 20% of this was lost in the feces and 60% was available for absorption. Composition of the total mixed ration is given in Table 1.

Cows were housed in a freestall barn and individually fed a total mixed ration once daily for ad libitum intake using Calan feeding doors (American Calan, Inc., Northwood, NH) with amounts fed and refusals recorded daily. After collection of pretreatment samples, cows were gradually switched to their respective treatment rations the last few days of the 3rd wk postpartum with the 4th wk postpartum as initiation of the 13-wk experimental period. Body weights were recorded 3 consecutive d at the start and conclusion of the experimental period as well as once every 2 wk during experimental period.

[3] Ketonin. Prills composed of 30% DL-methionine; 58% saturated and unsaturated fatty acids with 12 to 22 carbon atoms; 6% calcium carbonate; 1 to 2% glucose; 4% flavoring, antioxidant, and stabilizer. Supplied by Rumen Kjemi a/s, division of Peter Möller a/s, Oslo, Norway.

Cows were milked twice daily with milk weights recorded at each milking. Two 24-h (p.m. plus a.m.) milk samples were collected from each cow during wk 3 postpartum (pretreatment) and one 24-h sample was taken each week throughout the trial. Samples were analyzed for fat by Babcock (2), total solids by Mojonnier (3), and crude protein by Kjeldahl procedure (2). Remaining amounts of two milk samples taken approximately at wk 4 and 11 of the trial were frozen until analyzed for fatty acid distribution. Milk fat was extracted by the Roese-Gotlieb procedure (3) and butyl esters of fatty acids were prepared by methods of Jones and Davidson (14). Butyl esters were separated by gas-liquid chromatography using a 305 cm × 2 mm i.d. glass column packed with 10% SP-2330 on 100/120 Chromasorb W AW (Supelco, Inc., Bellefonte, PA). Column temperature was linearly programmed from 75 to 200°C at 6°C/min. Nitrogen was used as a carrier gas having a flow rate of 20 ml/min and injector and flame ionization detector temperatures were 230 and 240°C, respectively.

Concentrate mix, corn silage, and alfalfa hay were sampled weekly throughout the trial. Four weekly samples were combined into monthly composites for analyses of dry matter, crude protein, ether extract, and ash (2). Acid detergent fiber and permanganate lignin were determined by the procedures of Goering and Van Soest (12). Neutral detergent fiber was determined by the procedures of Robertson and Van Soest (24). Amino acid contents of feeds were determined on composites made from monthly composites of grain mix, corn silage, and alfalfa hay hydrolyzed in 6 N HCl in sealed tubes containing N_2 gas for 24 h at 145°C. Hydrolysates were evaporated to dryness, diluted with sodium citrate buffer (pH 2.2), filtered, and analyzed on a Spinco 120 automated amino acid analyzer (Beckman Instruments, Inc., Palo Alto, CA). Separation of amino acids was on ion-exchange columns with sodium citrate buffers ranging in pH from 3.49 to 6.40.

Ruminal contents were sampled from each cow three times during the experimental period approximately once every 4 wk. Samples were obtained 2 to 4 h after feeding by applying vacuum to an esophageal tube fitted with a suction strainer into a 250-ml sample bottle containing .5 ml saturated mercuric chloride. Samples were analyzed for pH and then filtered through four layers of cheesecloth. A 10-ml aliquot of the filtrate was centrifuged at 475 × *g* for 10 min. The supernatant was decanted and acidified with .5 ml of .1 N HCl and frozen until analyzed for rumen ammonia concentration (7). An additional 10-ml aliquot was acidified with 2 ml of 25% metaphosphoric acid, centrifuged at 475 × *g* for 10 min. The supernatant was decanted and frozen until analyzed for VFA by gas-liquid chromatography using a 10% SP-1200/1% phosphoric acid on 80/100 Chromasorb W AW (19).

Jugular vein blood was withdrawn into heparinized tubes at time of rumen sampling. Samples were centrifuged for 20 min at 800 × *g*. The supernatant fraction was decanted and frozen until analyzed for urea nitrogen (7) and glucose (Sigma Chemical Co., St. Louis, MO). Samples of tail vessel and mammary vein blood were obtained approximately 6 to 8 wk postpartum. Serum amino acid composition was determined in samples from 6 cows per treatment group by methods described by Drackley and Schingoethe (10). Amino acid uptake from blood transversing the mammary gland was estimated using differences in amino acid concentration of arterial and venous blood (AV) with mammary blood flow (MBF) estimated as described by Drackley and Schingoethe (10). This method of estimating blood flow was selected because it is simple, can be used without disturbing the cow, and is apparently as accurate (x ± 5%) as more direct methods of measuring blood flow (10). Averaged daily milk production and Kjeldahl nitrogen content of the milk produced during the arteriovenous blood sampling week and amino acid composition of milk (10) were used to estimate amino acid output in milk. Transfer efficiency of amino acids by mammary gland and ratios of amino acid uptake to output as described in (10) were used to identify amino acids limiting milk secretion.

Milk production and composition data were adjusted by analysis of covariance (31) using milk production and composition during the 3rd wk postpartum as covariates. Differences due to treatment, time, lactation number (multiparous vs. primiparous), and all interactions were considered. All data were subjected to least square analysis of variance (31) by the

TABLE 2. Chemical composition of concentrate mix, forages, and total ration.

Measurement	Concentrate mix	Corn silage	Alfalfa hay	Total diet[1]
Dry matter (DM), %	91.5	40.2	86.2	75.3
		(% of DM)		
Crude protein	19.5	8.1	17.5	15.8
Ether extract	4.7	2.8	1.6	3.7
Acid detergent fiber	6.1	27.5	40.2	17.6
Neutral detergent fiber	21.8	46.4	51.4	33.6
Permanganate lignin	2.0	4.8	9.5	4.0
Ash	6.5	4.9	8.4	6.3

[1]Calculated at ratio of 55:30:15 concentrate mix:corn silage:alfalfa hay.

general linear model procedure (SAS Institute, Cary, NC) with results expressed as least squares means.

RESULTS AND DISCUSSION

Lactation Study

Chemical analyses of concentrate mix, alfalfa hay, corn silage, and calculated total diet are in Table 2. Actual production of milk, 4% fat-corrected milk, and solids-corrected milk were similar for cows fed ESB and ESB + Met (Table 3). However, covariant adjusted milk and solids-corrected milk were lower for cows fed ruminally protected methionine. It is not known why cows fed supplemental methionine had lower adjusted production of milk and solids-corrected milk, but this response was likely not caused by dietary treatment. The negative response could have been due to cows fed ESB + Met having higher milk production (29.8 kg/d) than cows fed ESB (27.4 kg/d) during the pretreatment period (wk 3 postpartum). This may have indicated that higher producing cows were assigned to the ESB + Met diet, or those cows had fewer postpartum subclinical complications that allowed for a greater increase in production during the pretreatment period. Five cows assigned to ESB diet had problems such as dystocia, metritis, or parturient paresis, whereas only 2 cows assigned to ESB + Met had such complications. Except

TABLE 3. Covariant adjusted and unadjusted milk production and composition for cows fed diets containing an extruded blend of soybeans and soybean meal without (ESB) or with added protected methionine (ESB + Met).

Measurement	Unadjusted ESB	Unadjusted ESB + Met	SE	Adjusted[1] ESB	Adjusted[1] ESB + Met	SE
Milk, kg/d	35.1	35.1	.48	35.3	33.9[b]	.23
4% Fat-corrected milk, kg/d	28.2	28.2	.54	28.2	27.4	.28
Solids-corrected milk, kg/d	29.1	29.3	.50	29.3	28.2[a]	.25
Fat, %	2.70	2.69	.05	2.68	2.69	.04
Protein, %	2.85	2.91[a]	.02	2.86	2.90[a]	.01
Solids-not-fat, %	8.78	8.84	.03	8.82	8.83	.03
Total solids, %	11.48	11.54	.05	11.54	11.52	.05
Dry matter inake, kg/d	20.5	21.6[b]	.21			
Body weight, kg	598	604	5.99			

[a]Different from ESB ($P<.05$).

[b]Different from ESB ($P<.01$).

[1]Adjusted by analysis of covariance (31).

for a study by Illg et al. (13), which reported a 7% increase in milk production when cows were fed protected methionine with regular soybean meal, other researchers (5, 22, 23, 32, 33) reported similar production when cows were fed protected methionine.

Percentages of fat, solids-not-fat, and total solids were similar for cows fed ESB and ESB + Met (Table 3). However, the protein percentage was higher with methionine supplementation, indicating an increase in milk protein synthesis. Yang et al. (33) reported higher total solids content from cows fed ruminally protected methionine, although they concluded this resulted from a combined increase in fat and protein concentrations. Illg et al. (13), using the same source of protected methionine, reported an increase in the protein percent but no change in other milk components. Other studies utilizing ruminally protected methionine products reported no effect on milk composition (5, 22, 23, 32).

Fatty acid composition of milk fat (Table 4) was not affected by supplementation with protected methionine. Concentrations of short chain acids indicated no effect of supplemental methionine on de novo synthesis of short chain fatty acids in the mammary gland. Addition of whole soybeans to the concentrate mix increased the concentrations of long chain unsaturated fatty acids compared with those reported when cows were fed heat-treated soybean meal (33) or soybean meal (10, 17). Mielke and Schingoethe (17) also reported high concentrations of long chain unsaturated fatty acids when soybeans were fed than when soybean meal was fed.

Dry matter intakes (Table 3) were higher ($P<.01$) for cows fed ESB + Met. Some researchers (13, 22, 33) also reported increased dry matter intake with protected methionine, whereas others (23, 32) observed no difference in dry matter intake, and dry matter intakes were depressed in other trials when large concentrations of methionine were added to the diets (5, 21).

Molar proportions of individual VFA, total VFA concentrations, and ratio of acetate: propionate (Table 5) were similar for cows fed both diets. Ruminal pH and ammonia concen-

TABLE 4. Fatty acid composition of milk fat from cows fed diets containing an extruded blend of soybeans and soybean meal without (ESB) or with added protected methionine (ESB + Met).[1]

Fatty acid[2]	Diet		
	ESB	ESB + Met	SE
	(g/100 g)		
4:0	3.8	3.4	.27
6:0	2.4	2.4	.12
8:0	1.4	1.4	.08
10:0	3.0	3.4	.19
12:0	3.3	3.8	.19
14:0	10.4	11.1	.37
16:0	24.9	25.3	.54
16:1	2.8	2.8	.14
18:0	11.1	9.0	.67
18:1	27.6	25.9	.88
18:2	3.9	3.7	.19
18:3	.3	.4	.05
Short chain (4:0 to 14:0)	24.4	25.5	.89
Long chain (16:0 to 18:3)	70.5	67.1	1.14
Saturated	60.3	59.8	1.01
Unsaturated	34.6	32.8	1.00

[1] Means of samples taken wk 8 and 13 postpartum.

[2] Expressed as number of carbons:number of double bonds.

TABLE 5. Volatile fatty acids, ammonia, and pH in ruminal fluid, and serum urea and glucose concentrations in cows fed diets containing extruded blend of soybeans and soybean meal without (ESB) or with added protected methionine (ESB + Met).

	Diet		
Measurement	ESB	ESB + Met	SE
Volatile fatty acids			
Acetate, molar %	55.3	54.0	.80
Propionate, molar %	28.5	29.7	.92
Isobutyrate, molar %	.9	.8	.06
Butyrate, molar %	11.7	11.9	.50
Isovalerate, molar %	1.7	1.6	.12
Valerate, molar %	1.8	2.0	.14
Acetate:propionate	2.0	1.9	.10
Total, μmol/ml	97.9	101.2	4.08
Ruminal pH	6.48	6.36	.06
Ruminal ammonia, mg/dl	8.3	8.1	.90
Serum urea, mg/dl	16.9	15.5	.75
Serum glucose, mg/dl	66.1	65.0	1.76

trations as well as serum urea and glucose concentrations were unaffected by methionine supplementation.

Amino Acid Evaluation

Amino acid content of individual feeds and calculated total diet are in Table 6. Average daily methionine intake was 41 and 58 g for cows fed ESB and ESB + Met diets. Calculations prior to initiation of the experiment suggested that methionine would be the first-limiting amino acid with lysine being close to colimiting when comparing amino acid content of the feed to amino acid content of milk protein (10). Slight reductions in the amount of lysine available in heat-treated soybean meals (27, 28) could lead to potential change in order of limiting amino acid. The amino acid content of feed (Table 6) relative to amino acid content of milk protein (10) indicated that methionine was first-limiting with lysine almost colimiting. This would indicate that supplementation of methionine would immediately make lysine the first-limiting amino acid and no potential for a possible response. However, a mixture of dietary bypass and microbial protein is presented for gastrointestinal absorption and such a mixture should contain more than adequate amounts of lysine because of rumen microbial protein synthesis (4).

Concentrations of arterial and venous serum and arteriovenous differences are in Table 7. The essential amino acids (AA) isoleucine, leucine, and valine (branched chain AA) were higher in arterial and venous serum whereas arginine concentration was reduced in venous serum in cows fed additional methionine. The concentrations of all essential amino acids, except arginine, in arterial and venous serum were numerically higher in cows fed ESB + Met. Arginine is an important precursor for nonessential amino acid synthesis (8, 16). Total essential and total nonessential amino acids were slightly higher in cows fed ESB + Met. Why supplementation of methionine would raise concentrations of other amino acids is not known. However, increased concentrations of amino acids may be due to the increased dry matter and protein intake and not to the supplementation of methionine. Arteriovenous differences were similar for all amino acids for cows fed both diets except phenylalanine and valine, which had higher arteriovenous differences for cows fed ESB + Met.

Estimated uptakes [arteriovenous difference times estimated daily blood flow (10)] of essential amino acids relative to output in milk protein were adequate for milk protein synthesis as indicated by uptake to output ratios greater than 1.0 (Table 8). Uptake to output ratios for phenylalanine and valine were higher for cows fed ESB + Met. Valine may be oxidized or

TABLE 6. Amino acid content of concentrate mix, corn silage, alfalfa hay, and total diet.

Amino acid	Concentrate mix	Corn silage	Alfalfa hay	Total diet[1]
	(% of dry matter)			
Arginine	1.21	.26	.57	.83
Histidine	.52	.15	.29	.37
Isoleucine	.91	.29	.56	.67
Leucine	1.85	.74	.91	1.38
Lysine	1.01	.23	.68	.73
Methionine	.13	.32	.21	.20
Phenylalanine	1.02	.36	.70	.77
Threonine	.76	.28	.51	.58
Valine	1.07	.47	.88	.86
Total essential amino acids[2]	8.48	3.10	5.31	6.39
Alanine	1.08	.59	.71	.88
Aspartic acid	2.08	.56	2.96	1.76
Half-cystine	.09	.31	.12	.16
Glutamic acid	3.86	1.13	1.34	2.66
Glycine	.85	.33	.68	.67
Proline	1.29	.52	1.41	1.08
Serine	.89	.31	.56	.67
Tyrosine	.76	.40	.46	.61
Total nonessential amino acids	10.90	4.15	8.24	8.48
Total amino acids	19.38	7.25	13.55	14.87
Ammonia	.42	.21	.64	.39

[1] Calculated at ratio of 55:30:15 concentrate mix:corn silage:alfalfa hay.

[2] Tryptophan was not determined.

converted to nonessential amino acids (8, 16). Uptake of arginine was in excess of amounts secreted as milk protein, agreeing with previous studies (8, 16, 33).

A ranking based on the ratio of uptake to output (Table 8) suggests histidine, methionine, tryptophan, lysine, and phenylalanine as the first five limiting amino acids for cows fed ESB. However, methionine was the first-limiting amino acid for cows fed ESB + Met. Yang et al. (33) suggested that histidine was the first-limiting amino acid when cows were fed heat-treated soybean meal and ruminally protected methionine. A problem with this method of determining limiting amino acids is that it ranks amino acids from least to most used in pathways other than milk protein synthesis, which may not be an accurate prediction of amino acids limiting milk protein synthesis (8).

Transfer efficiencies of arterial amino acids incorporated into milk protein were similar for cows fed both diets (Table 8), except for branched chain amino acids isoleucine and valine, which had lower transfer efficiencies for cows fed ESB + Met. This would likely reflect the higher concentrations that were initially found in arterial serum (Table 7). Methionine, lysine, and phenylalanine would rank as first-, second-, and third-limiting amino acids with leucine and isoleucine being fourth and fifth for cows fed ESB. Threonine and leucine would be fourth and fifth for cows fed ESB + Met. If tyrosine was considered a limiting amino acid, it would rank third for cows fed both diets. The slightly lower transfer efficiency of methionine for cows fed ESB + Met suggests that methionine status was improved by methionine supplementation; however, methionine still remains the first-limiting amino acid.

In conclusion, the supplementation of extruded blend of whole soybeans and soybean meal with ruminally protected methionine did not increase milk production but increased protein percentage of milk. Blood serum amino acid analysis indicated that methionine status was improved, but the lack of increased

TABLE 7. Concentration of amino acids in arterial and venous serum, and arteriovenous (A-V) difference in cows fed diets containing extruded blend of soybeans and soybean meal without (ESB) or with added protected methionine (ESB + Met).

	Arterial serum			Venous serum			A-V difference		
Amino acid	ESB	ESB + Met	SE	ESB	ESB + Met	SE	ESB	ESB + Met	SE
	(μmol/dl)								
Arginine	16.6	16.3	.89	12.2	10.2[b]	.54	4.4	6.0	.79
Histidine	4.9	6.1	.46	3.7	4.2	.19	1.1	1.9	.36
Isoleucine	10.7	13.4[a]	1.00	5.9	7.4[a]	.54	4.9	6.0	.69
Leucine	15.9	20.4[a]	1.60	8.3	11.2[a]	1.04	7.6	9.1	.86
Lysine	7.7	7.8	.58	3.3	3.1	.28	4.4	4.7	.33
Methionine	1.7	2.0	.14	.5	.7	.14	1.2	1.3	.08
Phenylalanine	4.8	5.5	.39	2.4	2.5	.32	2.4	3.0[a]	.22
Threonine	10.2	10.3	1.11	6.8	6.2	.72	3.4	4.2	.49
Tryptophan	3.6	4.2	.58	3.0	3.1	.45	.5	1.1	.43
Valine	24.0	31.8[b]	1.97	17.8	22.9[b]	1.48	6.2	8.9[b]	.81
Total essential amino acids	100.6	118.3	7.63	62.6	70.6	4.12	38.0	47.7	4.52
Alanine	24.2	28.5	2.60	20.7	22.8	2.46	3.5	5.7	.97
Asparagine	5.1	5.9	.60	2.7	3.2	.43	2.4	2.7	.37
Aspartic acid	1.0	1.4	.23	1.5	1.3	.09	−.5	.1	.23
Half-cystine	.3	.2[b]	.01	.3	.2[a]	.02	.0	.0	.02
Glutamic acid	9.2	7.9	.61	4.4	3.7[b]	.25	4.8	4.2	.48
Glutamine	20.5	20.5	1.42	15.1	16.8	1.92	5.4	3.7	1.09
Glycine	46.0	45.9	4.96	44.8	42.6	5.27	1.2	3.3	1.26
Proline	10.0	11.9	.88	8.2	9.4	.93	1.8	2.5	.31
Serine	12.3	12.2	1.14	9.4	8.3	.91	2.9	3.9	.45
Tyrosine	4.1	4.9	.51	1.7	2.3	.34	2.4	2.7	.25
Citrulline[1]	9.0	9.8	1.07	8.6	8.7	.94	.4	1.1	.33
Ornithine[1]	5.2	5.6	.57	2.4	2.8	.33	2.7	2.8	.36
Taurine[1]	7.7	8.5	.63	7.4	7.8	.63	.2	.7	.25
Total nonessential amino acids	133.4	140.8	9.08	110.1	118.8	8.98	23.3	29.0	2.82

[a]Different from ESB ($P<.1$).

[b]Different from ESB ($P<.05$).

[1]Not included in total nonessential amino acids.

TABLE 8. Estimated ratio of uptake to output and transfer efficiencies of serum amino acids in cows fed diets containing extruded blend of soybeans and soybean meal without (ESB) or with added protected methionine (ESB + Met).

Amino acid	Uptake/output			Transfer efficiency[1]		
	ESB	ESB + Met	SE	ESB	ESB + Met	SE
Arginine	3.57 (10)[2]	4.74 (10)	.59	7.4 (10)	7.9 (10)	.36
Histidine	1.09 (1)	1.71 (5)	.32	24.2 (6)	18.3 (7)	3.36
Isoleucine	1.77 (8)	2.10 (7)	.22	26.3 (5)	21.8[a] (6)	1.57
Leucine	1.69 (7)	1.95 (6)	.17	28.9 (4)	23.8 (5)	1.99
Lysine	1.31 (4)	1.34 (2)	.09	44.2 (2)	46.2 (2)	3.07
Methionine	1.12 (2)	1.13 (1)	.06	62.7 (1)	58.4 (1)	4.51
Phenylalanine	1.33 (5)	1.61[a] (3)	.11	37.6 (3)	35.4 (3)	2.20
Threonine	1.45 (6)	1.70 (4)	.19	23.5 (7)	25.1 (4)	2.66
Tryptophan	1.22 (3)	2.55 (9)	.99	12.8 (9)	11.8 (8)	1.87
Valine	1.80 (9)	2.49[a] (8)	.22	14.6 (8)	11.3[b] (9)	.85
Tyrosine	1.41	1.49	.13	43.3	38.2	3.70

[a]Different from ESB ($P<.1$).

[b]Different from ESB ($P<.05$).

[1] $\frac{\text{Amino acid output in milk (g/d)} \times 100}{\text{Arterial serum amino acid (g/L)} \times \text{serum flow (L/d)}}$

[2] Number in parenthesis indicate apparent limiting order.

milk production would indicate that some factor or factors other than methionine status were limiting production.

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