

Feeding heat-treated soybean to mid-lactation Holstein cows: Production performance, predicted efficiency of nitrogen utilization, and blood metabolites

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Abstract The objective of this study was to compare the effects of soybean (SB) meal (SBM) with increased rumen undegradable protein as roasted SB (RSB), extruded SB (ESB) or their equal blend (RSB + ESB) on production performance, nutrient digestibility, N-utilization efficiency, and blood metabolites in mid-lactation dairy cows. Eight lactating Holstein cows (BW = 534 ± 52 and DIM = 104 ± 5; mean ± SD) were used in a replicated 4 × 4 Latin square design with four 28-d periods receiving 4 diets: (1) 13.88% of diet dry matter (DM) as SBM; (2) 15.22% of diet DM as RSB, (3) 15.55% of diet DM as ESB, and (4) 7.69% RSB plus 7.69% ESB (RSB + ESB). Each experimental period consisted of a 14-d diet adaptation followed by 14-d data collection. Dry matter intake, actual milk and 3.5% fat-corrected milk yield were not affected by the diet. Average milk fat and protein percentages (3.42 and 3.11%) and yields (1.28 and 1.17 kg/d), respectively, were not different among the diets. Plasma urea N concentration was similar across SB products but decreased compared with SBM. Feeding processed SB reduced NH₃-N concentration in the rumen (14.0 vs. 17.2 mg/dL; processed SB vs. SBM), indicating lower ruminal degradation of processed SB protein, and thereby improved N-utilization. Based on our results, RSB and ESB and their equal blend had a similar effect on productivity and N-utilization efficiency in mid-lactation Holstein cows.

Keywords: dairy cow, soybean meal, roasted – and extruded soybean, nitrogen efficiency

Received: 13 Apr. 2019, accepted: 04 Jun. 2019, published online: 14 Oct. 2019

Introduction

Soybean (SB) is a common source of protein with a good amino acid profile in the ration of high-producing dairy cows; however, most of its protein content can be degraded by rumen microbes, leading to a surplus ammonia production in the rumen (Mielke and Schingoethe, 1981; Akbarian et al., 2014). The aim of SB processing is to reduce ruminal degradability for high quality protein supplements with no negative effects on its intestinal digestibility. Extruding (Bailoni et al., 2004; Amanlou et al., 2012; Sadr-Arhami et al.,

2019) and roasting (Amanlou et al., 2012; Júnior et al., 2017; Sadr-Arhami et al., 2019) are common methods used to process SB for lactating dairy cows. Heating SB potentially decreases the degradable fraction of dietary protein in the rumen which increases the flow of the undegradable fraction of dietary protein to the small intestine. Akbarian et al. (2014) indicated that roasting of SB reduced the ruminal degradation of SB protein and enhanced its availability in the small intestine of dairy cows. Bailoni et al. (2004) reported that extruded SB (ESB) and roasted SB (RSB) may replace SB meal (SBM) at 9% of dietary DM in dairy cows producing 37 kg/d of milk without adverse effects on feed intake, rumen fermentation characteristics, and production performance. In another study, Amanlou et al. (2012) fed ESB and RSB at 10% of dietary DM as a replacement

Abbreviations:

DM: dry matter, CP: crude protein, NFC: non-fibrous carbohydrate, NDF: neutral detergent fiber, EE: ether-extract, DMI: dry matter intake, TMR: total mixed ration, FCM: fat-corrected milk, SB: soybean, SBM: soybean meal, RSB: roasted SB, ESB: extruded SB.

for SBM to high-producing (45 kg/d of milk) and observed no change in feed intake but increased fat-corrected milk yield for cows fed SB products as compared with cows fed SBM. More recently, Giallongo et al. (2015) showed that ESB, compared with solvent-extracted SBM, increased DM intake (DMI) and milk yield without affecting milk composition and feed efficiency in dairy cows. However, feeding RSB and ESB resulted in varied milk production responses in different studies. Heat processed SB, as compared with SBM, contains a greater amount of oil (NRC, 2001), which may provide additional energy for high-producing dairy cows, assuming it does not negatively affect ruminal fermentation and fiber digestibility and thereby feed intake. Therefore, there is a need to evaluate the use of different heat-treated SB products at higher inclusion rates when dietary energy is balanced in SBM supplemented diets using an inert source of fat in the rumen.

Although the impact of roasted (Bailoni et al., 2004; Amanlou et al., 2012; Akbarian et al., 2014) or extruded SB (Guillaume et al., 1991; Amanlou et al., 2012; Giallongo et al., 2015) on performance of dairy cows have been determined in several studies, to the best of our knowledge, there is little published information on the effects of mixture of processed SB products (RSB and ESB) on production performance and N-utilization efficiency in mid-lactation Holstein cows. We hypothesised that feeding ESB or RSB would have similar effects on the feed intake and milk production compared to SBM. We also hypothesised that feeding blended SB products would perform equally to those of ESB or RSB if not affect feed intake; therefore, RSB and ESB or their blend can be used interchangeably in dairy cow diets, allowing dairy producers more choices of feed ingredients. The objectives of the present study were to investigate the effects of RSB and ESB alone, and/or in equal blend on production performance, N-utilization efficiency, and blood metabolites in mid-lactation Holstein cows.

Materials and methods

Cows, experimental design and diets

The experiment was conducted at the Dairy Research Facilities of the Lavark Research Station from Isfahan University of Technology, Isfahan (Iran). Guidelines for the care and use of animals were approved by the Animal Care Committee of the University. Eight Holstein dairy cows (BW = 534 ± 52 and DIM = 104 ± 5; mean ± SD) were used in a replicated 4 × 4 Latin square design with four 28-d periods. Each experimental period

consisted of a 14-d diet adaptation and a 14-d collection duration. Cows were assigned to 1 of the 4 experimental diets: (1) 13.88% of diet DM as SBM, (2) 15.22% of diet DM as RSB, (3) 15.55% of diet DM as ESB, and (4) equal blending of RSB and ESB at 7.69% of diet DM (RSB + ESB). Cows were housed individually in box stalls (4 m × 4 m) located in a roofed area with open sides. Each box stall was equipped with a concrete feed bunk and automatic water troughs. Clean wood shavings and sand were used for bedding and refreshed twice daily. Cows were allowed to exercise in an outdoor lot daily from 1700 to 1800 h. All diets were formulated according to the Cornell Net Carbohydrate and Protein System (version 5.0) nutrient allowance for a lactating dairy cow weighing 534 kg and producing 37 kg/d of milk with 3.11 % milk true protein and 3.40 % fat and consuming 22 kg of DM. Diets were formulated to be iso-energetic and iso-nitrogenous, however, the CP content of the diet supplemented with SBM was slightly higher than the expected level (Table 2). Dietary ingredients were mixed for approximately 8 min in a total mixed ration (TMR) mixer wagon (Fan Avaran Keshavarzi Arya Co., Isfahan, Iran). The TMR were analyzed for DM, CP, ether extract, and NDF concentrations with 2 replicates in each period. Diets were provided twice daily at 0700 and 1400 h *ad libitum* and adjusted daily to allow at least 10% refusals. In this study, whole SB (Williams-82) was roasted at 145°C for 30 min as the optimum to maximize lactation response to roasting of SBs (Faldet and Satter, 1991) in a hot air commercial roaster (Roaster Jet., Asia-Roasting Danesh Co., Isfahan, Iran). Soybeans were extruded at 150 °C for 15-20 seconds with an initial and final pressure of 2.0265 and 6.0795 bars, respectively, using a commercial extruder (Extruder Jet, Koohpayeh Co., Isfahan, Iran). Cows were weighed immediately before the morning feeding at the beginning and end of each period. Predicted urine N output, fecal N, and apparent N efficiency were calculated using the following equations: predicted urine N output = 0.0283 × MUN (mg/dL) × BW (kg) (Wattiaux and Karg, 2004); predicted fecal N = N intake – predicted urine N – milk N (Cyriac et al., 2008); and apparent N efficiency = 100 × milk N (g/d) / N intake (g/d) (Cyriac et al., 2008).

Sampling and analysis

The amounts of diets offered and refused were measured daily for each cow with DMI determined daily. Refusals from individual cows were used for calculation of DMI. To determine DM and nutrient composition, representative samples of forages (pooled within the period),

Table 1. Chemical composition and protein fractions¹ in soybean meal (SBM), roasted soybean (RSB), and extruded soybean (ESB)

Item	SBM	RSB	ESB
Dry matter	82.39	83.13	83.57
Organic matter, % DM	93.15	94.44	94.02
Crude protein, % DM	53.13	38.90	39.90
A, % CP	4.90	0.00	0.52
B ₁ , % CP	19.82	7.20	11.10
B ₂ , % CP	25.48	24.01	22.62
B ₃ , % CP	1.98	3.84	2.83
C, % CP	0.95	3.84	2.83
Rumen degradable protein (RDP), % CP	63.18	36.06	48.46
Rumen undegradable protein (RUP), % CP	36.82	63.94	51.54
RDP/RUP	1.72	0.56	0.94
Non-fibrous carbohydrate, % DM	20.56	18.99	19.24
Neutral detergent fiber, % DM	18.31	17.3	13.42
Acid detergent fiber, % DM	8.80	14.69	10.60
Ether extract, % DM	1.15	19.25	21.46
Ash, % DM	6.85	5.56	5.98

¹Fraction A is soluble in buffer and tungstic acid, fraction B₁ is soluble in buffer and precipitated by tungstic acid, fraction B₂ is insoluble in buffer but soluble in neutral detergent, fraction B₃ is soluble in acid detergent but insoluble in neutral detergent, and fraction C is insoluble in acid detergent.

riod), treatment TMR (pooled by diet within the period), and individual refusals (pooled by cow within the period) were collected immediately before the morning feeding during the final 7-d collection period. One fecal grab sample per cow was taken from the rectum daily at 4 h after the morning feeding during the last 5 days of each period and frozen at -20°C until analyzed. Frozen samples were thawed at room temperature, dried at 60°C for 72 h, ground, and pooled (by cow within the period). Dry matter, crude protein, neutral detergent fiber (without sodium sulfite), ether extract, and ash content were analyzed as described by Kargar et al. (2012). The RUP and RDP of feed ingredients were calculated according to Sniffen et al. (1992) and Licitra et al. (1996). Acid detergent insoluble ash was used as an internal marker to determine the apparent total-tract nutrient digestibility (Van Keulen and Young, 1977; Kargar et al., 2012). The non-fibrous carbohydrate component was calculated (NRC, 2001) as $100 - (\text{CP} + \text{NDF} + \text{EE} + \text{Ash})$.

Rumen fluid sampling and ammonia N analyses

Rumen contents were sampled by rumenocentesis at 4 h after the morning feeding (Kargar et al., 2012). A 10 mL sample was preserved with 2 mL of 5% sulfuric acid and frozen at -20°C until analyses for determination of $\text{NH}_3\text{-N}$. For $\text{NH}_3\text{-N}$ determination, frozen rumen fluid samples were thawed at room temperature, and 5 mL of rumen fluid was vortexed and centrifuged (Eppendorf

AG, 5810R, Hamburg, Germany) at $3000 \times g$ for 20 min at 4°C to separate supernatant. The $\text{NH}_3\text{-N}$ concentration was determined by the colorimetric phenol-hypochlorite method using a spectrophotometer (Infinity M200, Tecan Austria GmbH, Grödig, Austria) according to Broderick and Kang (1980).

Blood sampling and analysis

Blood samples were collected in tubes (Vacutainer[®], Becton Dickinson, Rutherford, NJ, USA) containing lithium heparin from the coccygeal vein at 4 h after the morning feeding on d 27 of each period and placed immediately on ice. Samples were centrifuged at $3,000 \times g$ for 20 min at 4°C within 2 h of sampling. Blood plasma was separated and frozen at -20°C for subsequent analysis. Concentrations of plasma metabolites were spectrophotometrically (Model UNICCO-2100; Zistchemi, Tehran, Iran) determined using commercially available kits [Pars Azmoon Co., Tehran, Iran; Catalogue numbers: glucose (1-500-017), cholesterol (1-500-010), triglyceride (1-500-03), total protein (1-500-028), albumin (1-500-001), globulins (1-500-011), and BUN (1-400-029)] according to the manufacturer's instructions. Plasma concentration of βHBA (Randox Laboratories Ltd., Crumlin, UK) was determined automatically by using the standard test kit on an ALCYON 300i automatic analyzer (Abbott Laboratories Ltd., USA). The analyzer was calibrated, and control samples assayed daily according to the manufacturer's instructions to ensure acceptable assay performance.

Table 2. Ingredients and chemical composition of experimental diets¹ and predicted values for metabolizable protein, lysine, methionine, and lysine: methionine ratio

Item	Diets ¹			
	SBM	RSB	ESB	RSB + ESB
Ingredient, % DM				
Alfalfa hay	20.04	20.04	20.04	20.04
Corn silage	20.04	20.04	20.04	20.04
Barley grain, ground	17.55	17.55	17.55	17.55
Corn grain, ground	21.10	21.10	21.10	21.10
Soybean meal	13.88	-	-	-
Soybean, roasted	-	15.22	-	7.69
Soybean, extruded	-	-	15.55	7.69
Corn gluten meal	2.29	2.29	2.29	2.29
Fat powder	2.04	0.70	0.37	0.54
Vitamin-mineral mix ²	1.02	1.02	1.02	1.02
Sodium bicarbonate	1.02	1.02	1.02	1.02
Calcium carbonate	0.41	0.41	0.41	0.41
Di-calcium phosphate	0.41	0.41	0.41	0.41
White salt	0.20	0.20	0.20	0.20
Chemical composition, % DM				
Dry matter, % of as-fed	55.09	55.72	55.56	54.16
Crude protein	16.06	15.47	15.07	15.04
Metabolizable protein (MP), ³ kg/d	2.72	2.82	2.72	2.77
MP from bacteria, ³ kg/d	1.39	1.34	1.32	1.33
MP from RUP, ³ kg/d	1.33	1.48	1.41	1.44
Lysine, ³ % of MP	6.06	6.00	5.99	6.00
Methionine, ³ % MP	1.89	1.86	1.87	1.87
Lysine: methionine	3.21	3.23	3.20	3.21
NFC, ⁴	37.97	38.06	38.61	35.86
Neutral detergent fiber	33.63	33.08	32.94	35.13
Ether extract	4.48	5.70	5.71	5.87
NE _L , ³ Mcal/kg DM	1.65	1.67	1.67	1.67

¹SBM: soybean meal at 13.9% of dietary DM, RSB: roasted soybean at 15.2% of dietary DM, ESB: extruded soybean at 15.5% of dietary DM, and RSB + ESB: roasted- and extruded soybean at equal blend of 7.7% of dietary DM.

²Vitamin-mineral mix contained (DM basis): 1,300,000 IU/kg of vitamin A; 360,000 IU/kg of vitamin D₃; 12,000 g/kg of vitamin E; 10 g/kg of Mn; 16 g/kg of Zn; 4 g/kg of Cu; 0.15 g/kg of I; 0.12 g/kg of Co; 0.8 g/kg of Fe; and 0.08 g/kg of Se.

³Estimated using the Cornell Net Carbohydrate and Protein System method.

⁴Non-fibrous carbohydrate was calculated (NRC, 2001) as: $100 - (\text{CP} + \text{NDF} + \text{ether extract} + \text{ash})$.

Milk yield and components

Cows were milked three times daily at 0200, 1000, and 1800 h in a herringbone milking parlor. During the 14-d collection period, milk yield was recorded at each milking and sampled into tubes containing potassium dichromate as the preservative. Milk samples were analyzed for fat and protein using an infrared analyzer (Milk-o-Scan 134 BN; Foss Electric, Hillerød, Denmark). Fat-corrected milk (FCM) was calculated as $3.5\% \text{ FCM (kg/d)} = 0.35 \text{ (kg milk/d)} + 15.0 \text{ (kg fat/d)}$. The MUN content was determined enzymatically according to Wilson et al. (1998).

Statistical analysis

Data were composited within period and subjected to

MIXED MODEL procedure (SAS Institute, 2003) to account for the effects of square, period within square, cow within square, and diet. The model included the fixed effects of the square, period within the square, and diet. Cow within square was the random effect. Differences between diets were determined using the procedure of least squares means. The REML method was used to estimate the least squares means, and the Kenward-Roger method was used to calculate the denominator degrees of freedom. Normality of distribution and homogeneity of variance for residuals were tested using PROC UNIVARIATE (SAS Institute, 2003). In case of non-normality, parameters were normalized by log transformation prior to analysis to generate a normal distribution. Effects of the factors were considered significant at $P \leq 0.05$, and as a trend at $P \leq 0.10$.

Results and discussion

Diet characteristics

Chemical composition and protein fractions of SBM, RSB, and ESB are presented in Table 1. The ingredient composition, chemical analysis of experimental TMR and predicted values of metabolizable protein (MP), lysine (Lys), methionine (Met), and Lys:Met ratios are presented in Table 2. According to Table 1, CP content for the SBM was higher than that of the RSB and ESB, although diets were formulated to be both isonitrogenous and isoenergetic. As anticipated, heat processing extensively decreased the proportions of A, B₁, and B₂ fractions, but increased proportions of B₃ and C fractions compared to SBM. This resulted in an increase in RUP content and a decrease in RDP to RUP ratio of heat-processed SB products in comparison with SBM (Table 1). Furthermore, RSB provided more available protein post-ruminally than ESB (Bailoni et al., 2004). Based on the values in Table 1 and as reported by others (Bailoni et al., 2004; Sadr-Arhami et al., 2019), RSB and, in second order ESB, are potentially able to supply greater amount of RUP relative to SBM. The ingredient composition and chemical analysis of experimental TMR are presented in Table 2. Across diets, the DM, CP, NFC, NDF, and EE concentrations (mean ± SE) were 55.1 ± 0.4, 15.4 ± 0.2, 37.6 ± 0.6, 33.7 ± 0.5, and 5.4 ± 0.3% (DM basis), respectively.

Feed intake, and milk yield and milk composition

Dry matter intake was not different across experimental diets (Table 3). Feeding heat-processed SB products (RSB or ESB) vs. SBM resulted in varied DMI in different research studies. Several studies reported increase (Anderson et al., 1984; Giallongo et al. 2015) and/or no changes (Amanlou et al., 2012; Júnior et al., 2017; Sadr-Arhami et al., 2019) in DMI for cows fed heat-processed SB products vs. SBM. However, our results are in line with Mohamed et al. (1988) and Sadr-Arhami et al. (2019) who reported no differences in DMI between heat-processed SB products and SBM. The lack of effect of the treatment on DMI was attributed to no changes in meal size or inter-meal interval among diets as substantiated by Sadr-Arhami et al. (2019). In several studies (Guillaume et al., 1991; Bailoni et al., 2004; Amanlou et al., 2012) there was no difference in DMI between RSB and ESB. Overall, in the present experiment, the mean DMI was 22.0 kg/d, or approximately 4.1% of mean BW, being within the anticipated range for dairy cows producing > 35 kg/d of milk (NRC, 2001). Actual milk (37.5 kg/d) and 3.5% fat-corrected

milk (36.6 kg/d) yields were not affected by the diets as the diets were isoenergetic in the current study and DMI did not change across diets (Guillaume et al., 1991; Bailoni et al., 2004; Amanlou et al., 2012). Conversely, Faldet and Satter (1991) and Júnior et al. (2017) suggested that cows fed heat-processed SB yielded more milk than cows fed SBM or raw SB. Zimmerman et al. (1992) showed that feeding extruded products resulted in increased release rate of oil in the rumen, and a greater rumen escape of dietary CP, which may improve performance of lactating dairy cows. In their study, yield of FCM was greater (on average 4.0 kg/d) for heat-processed SB than for SBM or raw SB. Giallongo et al. (2015) observed greater feed intake (4.6%) for cows fed heat-processed SB vs. SBM and thereby increased milk yield (8.3%). Composition and yields of milk fat and protein were not different between diets.

No differences were observed in feed efficiency, milk fat, protein, and lactose yield and percentages among diets (Table 3) as also reported by others (Mohamed et al., 1988; Amanlou et al., 2012; Júnior et al., 2017). The lack of treatment effect on milk composition may be related to no significant changes in ruminal fluid pH and fermentation characteristics as explained by Sadr-Arhami et al. (2019).

Nitrogen efficiency and apparent total tract nutrient digestibility

Ruminal fluid NH₃-N concentration decreased ($P = 0.04$) in cows fed heat-processed SB (except for ESB) compared with those fed SBM (Table 4). This may be due to rapid and extensive degradation by the ruminal microbes (NRC, 2001). This is consistent with the reports that rumen concentration of NH₃-N was reduced when cows were fed heat-processed SB compared with SBM (Mielke and Schingoethe, 1981).

Milk urea N concentration was greater ($P = 0.03$) in cows feeding on the SBM diet compared with the other diets (Table 4). Kalscheur et al. (2006) reported that MUN concentration increased linearly from 9.5 mg/dL for cows fed the lowest RDP diet (6.8% of total dietary CP) to 16.4 mg/dL for cows fed the highest RDP diet (11% of total dietary CP). Predicted urinary N decreased ($P = 0.02$) in heat-processed SB diets compared with the SBM diet. Giallongo et al. (2015) observed numerically increased rumen NH₃-N, and increased concentration of MUN and urinary urea N excretion for ESB vs. SBM in lactating dairy cows despite similar dietary CP concentration. They attributed this effect to the increased CP intake for ESB vs. SBM diets, probably due to inclusion of NPN as urea. In the current study, no significant dif-

Table 3. Feed intake, milk yield and milk composition, and feed efficiency (least squares mean) as influenced by feeding processed soybean to lactating dairy cows

Item	Diet ¹				SEM	P-value
	SBM	RSB	ESB	RSB + ESB		
Intake, kg/d						
Dry matter	22.7	21.5	22.1	21.6	0.43	0.22
Milk yield and composition, kg/d						
Actual milk	37.5	37.8	37.6	37.2	0.56	0.91
3.5% FCM ²	37.4	37.3	35.7	36.1	0.90	0.47
Fat	1.33	1.31	1.22	1.25	0.05	0.32
Protein	1.17	1.16	1.17	1.17	0.02	0.92
Milk composition, %						
Fat	3.54	3.46	3.26	3.40	0.10	0.48
Protein	3.13	3.08	3.11	3.13	0.03	0.65
Feed efficiency						
Milk yield/DMI	1.67	1.74	1.70	1.74	0.04	0.55
FCM/DMI	1.64	1.73	1.62	1.67	0.05	0.62

¹SBM: soybean meal at 13.9% of dietary DM, RSB: roasted soybean at 15.2% of dietary DM, ESB: extruded soybean at 15.5% of dietary DM, and RSB + ESB: roasted- and extruded soybean at equal blend of 7.7% of dietary DM.

²3.5% fat-corrected milk (kg/d) = (0.4255 × milk yield [kg/d]) + (16.425 × fat yield [kg/d]).

Table 4. Rumen ammonia N, N retention, N efficiency, and apparent total tract nutrient digestibility (least squares mean) as influenced by feeding processed soybean to lactating dairy cows

Item	Diet ¹				SEM	P-value
	SBM	RSB	ESB	RSB + ESB		
Rumen NH ₃ -N, mg/dL	17.23 ^a	13.26 ^b	15.29 ^{ab}	13.45 ^b	0.77	0.04
MUN, mg/dl	13.10 ^a	10.36 ^b	10.91 ^b	10.47 ^b	0.64	0.03
Milk N, g/d	183.8	181.4	183.4	183.3	2.89	0.94
Predicted urine N, ² g/d	166.5 ^a	137.8 ^b	145.2 ^b	143.5 ^b	6.00	0.02
Predicted fecal N, ³ g/d	224.6	207.5	208.4	198.1	10.27	0.36
Apparent N efficiency, ⁴ %	31.98	34.67	34.02	34.85	0.89	0.10
Apparent total tract nutrient digestibility, %						
Dry matter	73.02 ^a	69.21 ^a	70.52 ^a	60.55 ^b	1.68	<0.01
Crude protein	71.70 ^a	67.98 ^a	68.51 ^a	57.04 ^b	1.72	<0.01
Ether extract	81.70	81.50	83.11	78.05	1.88	0.31

¹SBM: soybean meal at 13.9% of dietary DM, RSB: roasted soybean at 15.2% of dietary DM, ESB: extruded soybean at 15.5% of dietary DM, and RSB + ESB: roasted- and extruded soybean at equal blend of 7.7% of dietary DM.

²Predicted urine N output = 0.0283 × MUN (mg/dL) × BW (kg) (Wattiaux and Karg, 2004).

³Predicted fecal N = N intake – predicted urine N – milk N (Cyriac et al., 2008).

⁴Apparent N efficiency = 100 × milk N (g/d) / N intake (g/d) (Cyriac et al., 2008).

^{a-b}Means within a row with common superscript(s) are not significantly different ($P > 0.05$).

ferences were found in milk N and predicted fecal N. The lack of effect of heat-processed SB on fecal N excretion is in agreement with the similar apparent total-tract CP digestibility among diets except for RSB + ESB diet that had the lowest CP digestibility ($P < 0.01$; Table 4). Greater N intake in cows fed SBM diet resulted in a decrease ($P = 0.10$; Table 4) in apparent N efficiency as compared with the heat-processed SB. Vande-Haar and St-Pierre (2006) suggested that feeding high CP diets to dairy cows to support maximal milk production could result in low N efficiency. Our result is consistent with the report of Hristov et al. (2004) and Law et al. (2009) that suggested high dietary CP levels are positively associated with protein degradation rate in the rumen (in-

creased NH₃-N concentration), and decrease the efficiency of N utilization for milk production. These results are indicative of a beneficial effect of heat processing of SB on MUN concentration and apparent N efficiency. A major factor related to the lower MUN and increased apparent N efficiency with feeding RSB and ESB in this trial is that those diets contained about 0.85% unit, on average, less CP. It appears that CP intake, and the resulting metabolizable protein and essential amino acid supplies, exceeded animal requirements on all diets, and feeding diets with about 15% CP was sufficient to support the level of production observed in this trial. Taken together, the current data show a financial cost-benefit to the dairy producer and also an envir-

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onmental benefit from feeding heat-processed SB products (low CP and low RDP diets) vs. SBM (high CP and high RDP diet) to dairy cows.

Unexpectedly, the apparent total tract digestibility of DM and CP decreased ($P = 0.01$) in cows fed RSB + ESB diet compared with cows fed other diets, but no difference was found in EE digestibility among the diets (Table 4). It is unclear to us why RSB + ESB diet decreased nutrient digestibility as compared with others. A possible explanation could be the negative effect of unsaturated long chain fatty acids released from heat-treated SB products on rumen microbial population and consequently on ruminal DM and fiber degradation. The effects of unsaturated long chain fatty acids on rumen fermentation characteristics are well known (Jenkins, 1993; Harfoot and Hazlewood, 1997). The inhibitory effect of unsaturated long chain fatty acids is related to methanogenesis (decrease in reducing equivalents and also a reduction in methanogens count) when cellulolytic bacteria and ciliate protozoa counts are decreased which provide hydrogen as a substrate for methanogens (Harfoot and Hazlewood, 1997). Although we did not measure fiber digestibility in the current experiment, rumen fermentation characteristics (Sadr-Arhami et al., 2019) and milk fat percentages were not affected by the same experimental diets. It seems that in the current experiment, forage NDF concentration and intake across diets was sufficient to promote chewing, salivation, and high rumen pH, which may have masked the detrimental effects of unsaturated long chain fatty acids in the rumen (Sadr-Arhami et al., 2019). Consistent with the results reported by Mielke and Schingoethe (1981) and Guillaume et al. (1991), the apparent total tract DM and CP digestibilities were not affected by feeding heat-processed SB.

Blood metabolites

Blood plasma glucose (57.4 mg/dL), triglyceride (13.0

mg/dL), cholesterol (245.2 mg/dL), β HBA (0.75 mmol/L), total protein (8.2 mg/dL), albumin (4.0 g/dL), and globulin (4.2 g/dL) concentrations were not affected by the experimental diets ($P > 0.05$; Table 5). Inconsistent with the results reported by Bailoni et al. (2004), plasma urea concentrations were greater ($P < 0.01$) in cows consuming SBM diet as compared with others (14.90 vs. 11.2 mg/dL). Despite lower rumen $\text{NH}_3\text{-N}$ concentration in the study by Mielke and Schingoethe (1981) when fed RSB vs. SBM to lactating dairy cows, plasma urea N was not affected but was numerically lower in RSB diet. Milk urea N and BUN concentrations become elevated whenever diet CP is not utilized efficiently, whether due to excessive degradation in the rumen, poor utilization of metabolizable protein, or both. The absorbed amino acids not incorporated into milk protein are catabolized for energy with N being converted to urea and excreted. In fact, higher BUN concentrations express more rumen $\text{NH}_3\text{-N}$ which is being absorbed into the blood. The higher levels of BUN for SBM show that more $\text{NH}_3\text{-N}$ has been formed (Table 4) and wasted through increasing BUN, MUN, urea N which led to less N utilization efficiency. Consistent with our results in the current study, Zimmerman et al. (1992) showed that SB processing, increased RUP for digestion and absorption in the digestive tract and decreased BUN concentration. Law et al. (2009) reported that an increase in dietary protein concentration significantly increased plasma urea, total protein and albumin concentrations, and decreased plasma β HBA concentrations in early and mid-lactation periods. They also mentioned that increased plasma urea concentrations indicated increased ammonia detoxification in the liver, whereas an increase in total blood protein concentrations indicated intestinal absorption of protein, which will be evident in greater dietary protein contents.

Table 5. Blood biochemical metabolites (least squares mean) as influenced by feeding processed soybean to lactating dairy cows

Item	Diet ¹				SEM	P-value
	SBM	RSB	ESB	RSB + ESB		
Glucose, mg/dL	56.75	57.16	57.11	58.36	1.09	0.73
Triglyceride, mg/dL	11.88	13.75	12.47	13.88	0.65	0.16
Cholesterol, mg/dL	232.8	241.1	251.4	255.4	6.74	0.12
β HBA, mmol/L	0.78	0.71	0.82	0.70	0.05	0.57
Total protein, mg/dL	8.53	8.15	8.19	8.08	0.15	0.20
Albumin, g/dL	4.21	3.90	3.91	3.96	0.10	0.15
Globulin, g/dL	4.31	4.25	4.28	4.11	0.10	0.50
Plasma urea N, mg/dL	14.90 ^a	11.03 ^b	11.09 ^b	11.44 ^b	0.64	<0.01

¹SBM: soybean meal at 13.9% of dietary DM, RSB: roasted soybean at 15.2% of dietary DM, ESB = extruded soybean at 15.5% of dietary DM; and RSB + ESB = roasted- and extruded soybean at equal blend of 7.7% of dietary DM.

^{a-b}Means within a row with common superscript are not significantly different ($P > 0.05$).

Conclusions

It is concluded that feeding processed SB products vs. SBM had minimal effect on production performance of Holstein dairy cows but improved N-utilization efficiency and could be considered as alternative feedstuffs when the price is competitive and N excretion in the environment is limiting factor in balancing dairy rations. Furthermore, the findings suggested that there was no difference between RSB and ESB in terms of feed intake, productivity, and N-utilization efficiency in mid-lactation cows.

Acknowledgment

The authors gratefully acknowledge Isfahan University of Technology (Isfahan, Iran) for financial support and research facilities. The authors also express their appreciation to Mohammad Javad Zamiri (Emeritus Professor; Shiraz University) for editing the final English version of this manuscript.

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Communicating editor: Omid Dayani

تغذیه دانه سویای حرارت دیده به گاوهای هلستاین در اواسط دوره شیردهی: عملکرد تولید، تخمین بازدهی استفاده از نیتروژن و فراسنجه‌های خونی

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چکیده هدف از انجام این پژوهش مقایسه اثر تغذیه کنجاله سویا (SBM) با دانه سویای برشته شده (RSB)، دانه سویای اکستروود شده (ESB) و آمیزه یکسانی از آن‌ها (RSB + ESB) بر عملکرد تولید، گوارش پذیری مواد مغذی، بازده استفاده از نیتروژن و فراسنجه‌های خونی گاوها در اواسط دوره شیردهی بود. برای این منظور از هشت رأس گاو هلستاین (با وزن 52 ± 534 و روزهای شیردهی 5 ± 104 ؛ انحراف معیار \pm میانگین) در قالب طرح مربع لاتین تکرار شده 4×4 و دوره‌های ۲۸ روزه که ۴ جیره پی‌آیند را دریافت می‌کردند استفاده شد: (۱) جیره دارای ۱۳/۸۸ درصد (بر اساس ماده خشک) SBM، (۲) جیره دارای ۱۵/۲۲ درصد (بر اساس ماده خشک) RSB، (۳) جیره دارای ۱۵/۵۵ درصد (بر اساس ماده خشک) ESB و (۴) جیره دارای آمیزه یکسانی از RSB (۷/۶۹ درصد بر اساس ماده خشک) و ESB (۷/۶۹ درصد بر اساس ماده خشک) (RSB ± ESB). هر دوره آزمایشی شامل ۱۴ روز سازگاری به جیره و ۱۴ روز جمع‌آوری داده بود. ماده خشک مصرفی و تولید شیر خام و شیر تصحیح شده برای ۳/۵ درصد چربی تحت تأثیر جیره آزمایشی قرار نگرفت. میانگین درصد (۳/۴۲ و ۳/۱۱ درصد) و تولید (۱/۲۸ و ۱/۱۷ کیلوگرم در روز) چربی و پروتئین شیر بین جیره‌ها متفاوت نبود. غلظت نیتروژن اوره‌ای پلاسما بین محصولات فرآوری شده دانه سویا یکسان بود اما در مقایسه با SBM کاهش یافت. تغذیه محصولات فرآوری شده دانه سویا در مقایسه با SBM غلظت نیتروژن آمونیاکی را در شکمبه کاهش داد (۱۴/۰ در مقابل ۱۷/۲ میلی‌گرم بر دسی لیتر) که دال بر تجزیه‌پذیری شکمبه‌ای کم‌تر آن‌ها و در نتیجه بهبود در بازده استفاده از نیتروژن است. بر اساس این نتایج، RSB و ESB و آمیزه آن‌ها اثر یکسانی بر عملکرد تولید و بازده استفاده از نیتروژن در گاوهای هلستاین اواسط دوره شیردهی داشتند.