

# Effect of feeding tamarind kernel powder extract residue on digestibility, nitrogen availability and ruminal fermentation in wethers

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Submitted Feb 22, 2016; Revised Apr 16, 2016;  
Accepted May 13, 2016

**Objective:** This study was to examine *in vivo* digestibility, nitrogen balance and ruminal fermentation of tamarind (*Tamarind indica*) kernel powder extract residue (TKPER) compared to soybean products and by-products in wethers.

**Methods:** Four wethers with initial body weight (BW) of 51.6±5.5 kg were assigned in a 4×4 Latin square design to investigate nutritional characteristics of TKPER, dry heat soybean (SB), dry soybean curd residue (SBCR) and soybean meal (SBM) feeding with ryegrass straw (R) at a ratio of 1:1 at 2% of BW in dry matter (DM) on a daily basis.

**Results:** The digestibility of DM, crude protein, and ether extract (EE) of TKPER-R diet were 57.0%, 87.0%, and 86.0%, respectively. Higher non-fiber carbohydrates digestibility was observed in TKPER-R diet (83.2%) than in SB-R diet (73.9%,  $p<0.05$ ). Wethers fed the TKPER-R diet had lower retention of nitrogen (N) and ruminal ammonia nitrogen (NH<sub>3</sub>-N) contents at 4 h after feeding than those fed the SBM-R diet ( $p<0.05$ ), which had values similar to the SB-R or SBCR-R diet. The TKPER feeding had higher propionate (C3) and lower butyrate content, as well as lower acetate to propionate ratio (C2:C3) in rumen fluid than SBM feeding at 4 h after feeding ( $p<0.05$ ).

**Conclusion:** TKPER did not bring any side effect to the wethers although it was lack of fiber, and could be used as a high protein and energy ingredient in concentrate with appropriate roughage to meet the fiber requirement for ruminants.

**Keywords:** By-product; Digestibility; Nitrogen Balance; Rumen Condition; Tamarind Kernel Powder; Wethers

## INTRODUCTION

Utilization of by-products is of great importance in rearing of livestock, both in developing and developed countries. In India, for instance, various by-products such as rubber seed cake, mango seed kernel, tea waste and tamarind seed have been recommended for use at the 10% to 30% level in the concentrate of livestock rations [1]. Taking Japan as an example of developed countries, the food industry is a major part of the economy, accounting for 10% of total industrial production [2], and a huge amount of industrial by-products are disposed each year. Livestock feeding in Japan relies heavily on imported feeds, which are easily affected by price fluctuation due to shortages. Utilization of by-products as feed ingredients has been promoted in order to improve self-sufficiency [3].

The nutritional characteristics of several food industrial by-products have been investigated in comparison to conventional feeds. In Japan, studies have examined the mixing and ensiling of these by-products with other feed ingredients. Ishida et al [4] reported that total mixed ra-

tion (TMR) silage, including soy sauce cake and noodle waste that was well preserved with high fermentation quality showed significantly higher digestibility of dry matter (DM) and organic matter (OM) compared to barley- and corn-based concentrate [4]. As for fattening of Japanese Black heifers is concern, no significant difference in DM intake or daily gain were observed between TMR silage feeding and the concentrate feeding [5]. Yani et al [6] demonstrated that intake and adequacy of total digestible nutrients (TDN) were significantly higher in dairy cows fed the TMR silage than those fed commercial concentrate-based TMR silage [6]. Later, Yani et al [7] studied the effects of utilization of local food by-products as TMR silage materials in sheep. The TMR silage including potato waste, noodle waste and soybean curd residue had significantly higher DM and neutral detergent fiber (aNDFom) digestibility and TDN content than control feeding which contained commercial concentrate in sheep [7].

Tamarind (*Tamarindus indica*), a member of the family Leguminosae, is mainly distributed throughout the tropical and subtropical regions, and is considered to be one of most important plant resources as food materials [8,9]. There are several processed products of tamarind, such as juice concentrate, jam, pickle, toffees as well as candy [10]. Tamarind seed, which is rich in polysaccharide, protein, and lipid, is a by-product of the commercial utilization of tamarind fruit. In Japan, tamarind kernel powder (TKP) has been used as the raw material in the production of polysaccharide thickener after being peeled and ground for thickening stabilizing and gelling in food. It is widely used as a food additive for improving viscosity and texture of processed foods [11]. Tamarind kernel powder extract residue (TKPER) is a by-product of the processing of polysaccharide thickener. Numerically 400 tons of TKPER are produced annually in one of the major factories in Japan and most of them are incinerated or buried in landfill in spite of its high crude protein (CP) and energy contents. It's well-known that protein is a quite important resource of nitrogen for microorganism to synthesize microbial protein in the rumen synchronizing with utilization of other nutrients in the diets, such as fermentable carbohydrate. Oh et al [12] reported that rumen degraded protein increased by higher level and degradability of dietary protein, may increase release of free amino acids, peptides and soluble proteins in the rumen of Hanwoo steers [12]. In rearing of dairy cows, protein in diet is a key factor which could insure an adequate supply of metabolizable protein and essential amino acids to allow maximal production of milk and milk protein. Milk production increased 0.75 kg/d when dietary CP was increased from 15% to 16% and 0.35 kg/d when CP was increased from 19% to 20% [13]. Moreover, TKPER has advantages not only in protein content but also in price, which will cost less than several commercial concentrate. Hence, it might be possible to use TKPER as an alternative to certain conventional ingredients of commercial concentrate, such as soybean products

and by-products.

Nevertheless, to our knowledge, there have been no studies on the use of TKPER as a by-product feed in livestock production. The objective of this study was to examine *in vivo* digestibility, nitrogen balance and ruminal fermentation of TKPER compared to soybean products and by-products in wethers.

## MATERIALS AND METHODS

### Animal care

The wethers used in the experiment were managed according to the guidelines of the Kyoto University Animal Ethics Committee.

### Preparation of feeds

The TKPER was prepared by a chemical company in the Osaka Prefecture, Japan. Briefly, TKP was processed from dehulled and ground tamarind seeds, and then treated with alkaline solution and refined to extract polysaccharide for food additives. The residue from extraction was concentrated and dried to get TKPER. The dry heat soybean (SB) and soybean meal (SBM) were purchased from a feed company located in Hyogo Prefecture, Japan. The dry soybean curd residue (SBCR) was processed and dried in a food factory in Kyoto Prefecture, Japan. The chemical composition of feeds was analyzed as shown in Table 1. TKPER had similar CP and ether extract (EE) contents to SB on a DM basis. No aNDFom content was detected in TKPER. The non-fiber carbohydrate (NFC) content of TKPER was higher than that of SB, SBCR, and SBM.

### Experimental design, treatments and sampling

Four ruminal cannulated wethers with initial body weight (BW) of  $51.6 \pm 5.5$  kg were used in a 4×4 Latin square design experiment. The wethers were housed individually in four metabolic cages at the animal shelter at Kyoto University, Kyoto, Japan. Each experimental period was 14 days, consisting of a 9-day adaptation period and a 5-day sample collection period.

The experiment consisted of four treatments which were SB,

**Table 1.** Chemical composition (%) of the test feeds and ryegrass straw

Item	SB	SBCR	SBM	TKPER	Ryegrass straw
Dry matter	93.4	92.6	89.8	88.5	89.2
Organic matter <sup>1)</sup>	94.3	96.5	93.3	91.6	95.8
Crude protein <sup>1)</sup>	39.7	27.4	51.5	42.4	5.8
Ether extract <sup>1)</sup>	20.6	9.4	1.9	15.0	2.1
aNDFom <sup>1)</sup>	16.9	28.1	10.8	ND	65.0
Crude ash <sup>1)</sup>	5.7	3.5	6.7	8.4	4.2
NFC <sub>2)</sub>	17.1	31.6	29.1	34.3	22.9

SB, dry heat soybean; SBCR, dry soybean curd residue; SBM, soybean meal; TKPER, tamarind kernel powder extract residue; aNDFom, neutral detergent fiber exclusive of residual ash; ND, not detected; NFC, non-fiber carbohydrate; CA, crude ash.

<sup>1)</sup> On a dry matter basis.

<sup>2)</sup> Calculated by  $100 - (CP + EE + aNDFom + CA)$ .

SBCR, SBM, and TKPER fed with ryegrass straw (R) at a ratio of 1:1; these are referred to as the SB-R, SBCR-R, SBM-R, and TKPER-R treatments, respectively. The wethers were fed with the experimental diets at 2% of BW on a DM basis in two equal portions daily, at 08:30 h and 17:30 h. They were allowed free access to water and mineral blocks. No refusal was observed throughout the experimental period.

All feces and urine were collected every morning before feeding during the 5-day sample collection period. The feces samples were collected manually, weighed, and then dried in an oven at 60°C for 48 h, after which the DM was weighed. The 5 days' collected feces from each wether were mixed together, and 50 g of the mixed samples were ground with a Willey mill to pass through a 1-mm screen, and then stored for further analyses. The digestibility and TDN contents of the test feeds were calculated by subtracting the digestibility and TDN contents of ryegrass straw from those of total experimental diets, according to the method described by the National Agriculture and Food Research Organization [14].

The digestibility of a nutrient in the test feed being fed in form of mixed feed is calculated as follow: Digestibility of nutrient in test feed (%) =  $(A - B \times C) \times 100 / D$ , A: Digestibility of nutrient in total diet; B: Digestibility of nutrient in basal diet (ryegrass straw); C: proportion of total nutrient in diet supplied by basal diet; D: proportion of total nutrient in diet supplied by test feed.

The TDN content of each test feed was estimated by the following equation: TDN content = digestible OM content + digestible EE content  $\times 1.25$  [14]. The urine samples were collected in plastic trays, 45 mL sulfuric acid (20%) was added, and the volume was measured. The 5 days' collected urine samples from each wether were mixed together, and 50 mL of the mixed samples were put into a plastic bottle and stored at -20°C for further analyses.

The rumen fluid samples were collected at 0 h (before feeding) and 4 h after feeding on the morning of the last day of each experimental period. The rumen fluid was filtered through four layers of gauze, and then analyzed for pH immediately using a glass electrode pH meter (Horiba Ltd., Kyoto, Japan). It was then centrifuged at 500 $\times$ g for 5 min and the supernatants were stored at -20°C for further analyses.

Jugular blood samples of approximately 10 mL were collected one time from each wether in vacuum plasma tubes at 0 h (before feeding) on the last day of each experimental period. The collected blood samples were centrifuged at 2,600 $\times$ g for 15 min, and the plasma was stored at -20°C for further analyses.

### Chemical analyses

The four test feeds, SB, SBCR, SBM, and TKPER, and ryegrass straw were sampled from 4 batches during the metabolic experiment, and analyzed for DM, CP, EE, and crude ash (CA) according to the standards of the Association of Official Analytical Chemists [15]. The content of OM was calculated as weight

loss through ashing. aNDFom was analyzed according to the procedure described in Van Soest et al [16]. The content of NFC was calculated from CP, EE, aNDFom, and CA using the following equation:  $NFC = 100 - (CP + EE + aNDFom + CA)$ .

The feces samples were analyzed for DM, CP, EE, OM, aNDFom, and NFC, as described above. Urine samples were analyzed for nitrogen (N) content using the Kjeldahl procedure. Ruminal ammonia nitrogen (NH<sub>3</sub>-N) was determined by the microdiffusion method [17]. Ruminal volatile fatty acid (VFA) concentrations were measured by gas chromatography (GC-14B; Shimadzu, Kyoto, Japan) equipped with a flame ionization detector. The chromatography was carried out with a packed glass column (Thermon 3000-2% Shimalite TPA 60/80 3.2 mm $\phi$  $\times$ 2.1; Shimadzu, Japan). The temperatures of the column, detector, and injection were 120°C, 250°C, and 250°C, respectively, using nitrogen as the carrier gas.

The blood plasma samples were analyzed for glucose (Glu), total protein (TP), albumin (Alb), blood urea nitrogen (BUN), nonesterified fatty acid (NEFA), total cholesterol (T-Chol), phospholipids (PL), calcium (Ca) and inorganic phosphorus (IP) using diagnostic kits (Glucose-HR II, NEFA-HR, Albumin-HR II, L type Wako UN, L type Wako CHO•H, L type Wako Phospholipids, CalciumE-HA and Inorganic phosphorus-HR II, Wako Pure Chemical Industries, Ltd., Tokyo, Japan). Aspartate aminotransferase (AST), alanine transaminase (ALT) and  $\gamma$ -glutamyl transpeptidase ( $\gamma$ -GTP) activities were analyzed according to the standard methods established by the Japan Society of Clinical Chemistry [18].

### Statistical analysis

Data on chemical composition of diets, intake, apparent digestibility, nitrogen balance, ruminal pH, ruminal NH<sub>3</sub>-N and blood metabolites were analyzed using general linear model procedure of the Statistical Analysis System (SAS Inst. Inc., Cary, NC, USA). The model was  $Y_{ijkl} = \mu + T_i + P_j + A_k + e_{ijkl}$ , where  $\mu$  = the overall mean,  $T_i$  = the fixed effect of treatment,  $P_j$  = the fixed effect of period,  $A_k$  = the random effect of animal and  $e_{ijkl}$  = residual error. Significance was declared at  $p < 0.05$ .

## RESULTS

### Chemical composition of the experimental diets

The DM and OM contents of the diets ranged from 88.8% to 91.3% and from 92.1% to 95.8%, respectively, and no significant differences were found in DM or OM among these four experimental diets (Table 2). The CP content of TKPER-R was significantly higher than that of SBCR-R and lower than that of SBM-R ( $p < 0.05$ ). There was no significant difference observed in EE content between TKPER-R and the other three experimental diets, respectively. The aNDFom content of TKPER-R at 32.4% was lower than that of SB-R and SBCR-R ( $p < 0.05$ ). The NFC content of TKPER-R, 28.4%, was significantly higher than that

**Table 2.** Chemical composition of the experimental diets

Item	SB-R	SBCR-R	SBM-R	TKPER-R
Dry matter (%)	90.6	91.3	89.4	88.8
Organic matter (%) <sup>1)</sup>	94.7	95.8	93.7	92.1
Crude protein (%) <sup>1)</sup>	23.4 <sup>b</sup>	17.9 <sup>a</sup>	35.4 <sup>c</sup>	24.6 <sup>b</sup>
Ether extract (%) <sup>1)</sup>	10.3 <sup>b</sup>	5.5 <sup>ab</sup>	1.4 <sup>a</sup>	6.7 <sup>ab</sup>
aNDFom (%) <sup>1)</sup>	40.3 <sup>b</sup>	50.1 <sup>c</sup>	34.3 <sup>a</sup>	32.4 <sup>a</sup>
Crude ash (%) <sup>1)</sup>	5.3 <sup>a</sup>	4.2 <sup>a</sup>	6.3 <sup>ab</sup>	7.9 <sup>b</sup>
NFC (%) <sup>2)</sup>	20.7 <sup>a</sup>	22.3 <sup>ab</sup>	22.7 <sup>ab</sup>	28.4 <sup>b</sup>

SB-R, dry heat soybean with ryegrass straw on a dry matter basis; SBCR-R, dry soybean curd residue with ryegrass straw on a dry matter basis; SBM-R, soybean meal with ryegrass straw on a dry matter basis; TKPER-R, tamarind kernel powder extract residue with ryegrass straw on a dry matter basis; aNDFom, neutral detergent fiber exclusive of residual ash; NFC, non-fiber carbohydrate; CA, crude ash. Means in the same row with different superscripts differ significantly ( $p < 0.05$ ).

<sup>1)</sup> On a dry matter basis.

<sup>2)</sup> Calculated by  $100 - (CP + EE + aNDFom + CA)$ .

of SB-R ( $p < 0.05$ ).

**Digestibility and nitrogen balance**

DM intake and digestibility of DM and OM were not different among treatments (Table 3). The CP digestibility of TKPER-R was significantly higher than SBCR-R ( $p < 0.05$ ), but not from SB-R or SBM-R. No significant difference was observed among SB-R, SBCR-R and TKPER-R in EE digestibility and the EE digestibility of SBM-R was half of those of SB-R, SBCR-R, and TKPER-R. The aNDFom digestibility of TKPER-R was lower than that of SBCR-R ( $p < 0.05$ ). Meanwhile, NFC digestibility of TKPER-R was similar to that of SBCR-R and SBM-R, and higher than that of SB-R ( $p < 0.05$ ).

**Table 3.** DM intake, apparent digestibility and nitrogen balance of the experimental diets in wethers

Item	SB-R	SBCR-R	SBM-R	TKPER-R	SEM
DM intake (g/d/BW <sup>0.75</sup> )	48.6	49.0	47.9	47.6	0.30
Digestibility (%)					
Dry matter	56.5	59.9	60.2	57.0	0.42
Organic matter <sup>1)</sup>	69.7	76.9	72.2	72.4	0.47
Crude protein <sup>1)</sup>	83.4 <sup>ab</sup>	80.3 <sup>a</sup>	90.7 <sup>c</sup>	87.0 <sup>bc</sup>	0.59
Ether extract <sup>1)</sup>	88.6 <sup>b</sup>	83.1 <sup>b</sup>	43.5 <sup>a</sup>	86.0 <sup>b</sup>	1.49
aNDFom <sup>1)</sup>	54.8 <sup>ab</sup>	64.2 <sup>b</sup>	56.9 <sup>ab</sup>	49.0 <sup>a</sup>	3.07
NFC <sup>1)</sup>	73.9 <sup>a</sup>	83.4 <sup>b</sup>	85.6 <sup>b</sup>	83.2 <sup>b</sup>	1.32
N balance (g/d/BW <sup>0.75</sup> )					
N intake	1.81 <sup>a</sup>	1.40 <sup>a</sup>	2.71 <sup>b</sup>	1.88 <sup>ab</sup>	0.01
Fecal N	0.30	0.28	0.25	0.24	0.05
Urinary N	1.20 <sup>ab</sup>	0.82 <sup>a</sup>	1.75 <sup>b</sup>	1.32 <sup>ab</sup>	0.13
Retention N	0.32 <sup>a</sup>	0.30 <sup>a</sup>	0.71 <sup>b</sup>	0.31 <sup>a</sup>	0.05
N retention (% N intake)	18.02	21.46	26.13	16.79	3.36

DM, dry matter; SB-R, dry heat soybean with ryegrass straw on a dry matter basis; SBCR-R, dry soybean curd residue with ryegrass straw on a dry matter basis; SBM-R, soybean meal with ryegrass straw on a dry matter basis; TKPER-R, tamarind kernel powder extract residue with ryegrass straw on a dry matter basis; SEM, standard error of means; BW, body weight; aNDFom, neutral detergent fiber exclusive of residual ash; NFC, non-fiber carbohydrate. Means in the same row with different superscripts differ significantly ( $p < 0.05$ ).

<sup>1)</sup> On a dry matter basis.

The estimated digestibility and TDN of TKPER were calculated by subtracting the digestibility and TDN of ryegrass straw. The DM, CP, and NFC digestibility and TDN content were 58.7%, 94.6%, 84.7%, and 93.8%, respectively.

As for nitrogen balance, the intake N, fecal N, and urinary N levels of TKPER-R did not significantly differ from those of SB-R, SBCR-R, and SBM-R. The retention N of TKPER-R was significantly lower than that of SBM-R ( $p < 0.05$ ). There was no significant difference among treatments in N retention (% N intake).

**Ruminal fermentation**

No significant differences were found in the pH of ruminal fluid before feeding among the four treatments (Table 4). At 4 h after feeding, the pH of SBCR-R was lower than those of SB-R and TKPER-R ( $p < 0.05$ ), but no significant differences were observed between SB-R, SBM-R, and TKPER-R. As for the ruminal NH<sub>3</sub>-N, SBCR-R was lowest and SBM-R was highest in concentration ( $p < 0.05$ ) at both sampling times. The NH<sub>3</sub>-N content in TKPER-R did not significantly differ from SB-R or SBM-R before feeding, or from SB-R and SBCR-R at 4 h after feeding.

The total VFA concentration (mmol/L) of each treatment before feeding was 60.6, 77.6, 78.8, and 66.9 for SB-R, SBCR-R, SBM-R, and TKPER-R, respectively. No significant differences were found in the composition of acetate (C2), propionate (C3)

**Table 4.** Ruminal fermentation in wethers fed with the experimental diets at 0 and 4 h after feeding

Item	SB-R	SBCR-R	SBM-R	TKPER-R	SEM
Ruminal pH					
0 h	6.98	6.82	6.73	7.05	0.08
4 h	6.42 <sup>b</sup>	5.92 <sup>a</sup>	6.25 <sup>ab</sup>	6.62 <sup>b</sup>	0.10
NH <sub>3</sub> -N (mg N/dL)					
0 h	43.40 <sup>ab</sup>	31.50 <sup>a</sup>	61.25 <sup>c</sup>	47.95 <sup>bc</sup>	3.71
4 h	51.10 <sup>b</sup>	28.35 <sup>a</sup>	73.33 <sup>c</sup>	43.58 <sup>ab</sup>	4.93
Total VFA (mmol/L)					
0 h	60.6	77.6	78.8	66.9	10.17
4 h	74.4	83.4	76.6	84.6	5.94
Acetate (C2) (%)					
0 h	53.6	57.2	54.5	52.9	1.12
4 h	51.1	54.0	52.1	50.2	1.29
Propionate (C3) (%)					
0 h	28.1	25.5	24.1	28.5	1.34
4 h	30.6 <sup>ab</sup>	32.4 <sup>b</sup>	24.8 <sup>a</sup>	34.6 <sup>b</sup>	1.32
Butyrate (nC4) (%)					
0 h	13.1	13.4	15.9	12.6	1.11
4 h	14.7 <sup>ab</sup>	11.8 <sup>a</sup>	18.8 <sup>b</sup>	12.2 <sup>a</sup>	0.98
C2:C3					
0 h	1.9	2.3	2.3	1.9	0.13
4 h	1.7 <sup>ab</sup>	1.7 <sup>ab</sup>	2.1 <sup>b</sup>	1.5 <sup>a</sup>	0.11

SB-R, dry heat soybean with ryegrass straw on a dry matter basis; SBCR-R, dry soybean curd residue with ryegrass straw on a dry matter basis; SBM-R, soybean meal with ryegrass straw on a dry matter basis; TKPER-R, tamarind kernel powder extract residue with ryegrass straw on a dry matter basis; SEM, standard error of means. Means in the same row with different superscripts differ significantly ( $p < 0.05$ ).

and butyrate (nC4) contents, or in the C2:C3 ratio among treatments before feeding. At 4 h after feeding, there were no significant differences in total VFA concentration and C2 content among the treatments. The TKPER-R had higher C3 but lower nC4 contents and lower C2:C3 ratio than SBM-R ( $p < 0.05$ ), whereas no significant differences in C3 content, nC4 content, or C2:C3 ratio were found between SB-R, SBCR-R, and TKPER-R.

### Blood metabolites

No significant difference was observed in Glu, TP, Alb, NEFA, AST, ALT,  $\gamma$ -GTP, Ca, and IP concentrations among four treatments (Table 5). The T-Chol and PL concentrations of TKPER-R did not differ significantly compared to the other three treatments, respectively. The BUN concentration of TKPER-R was significantly higher than that of SBCR-R.

## DISCUSSION

### Chemical composition, digestibility and nitrogen balance

The CP contents of TKPER are similar to those of soybean, and slightly less than those of SBM; the EE contents are similar to those of SB and SBCR according to the NARO [14]. TKPER had high NFC content and no aNDFom content, which was one of its most important characteristics compared to the other test feeds. The high NFC content in TKPER was attributed to polysaccharide escaping from the extraction process for food additives, and the quite low aNDFom content was due to the alkaline treatment before extraction. In the diet of dairy cows, NFC is an important nutrient, as it can increase milk protein content and prevent laminitis [19]. Increasing NFC diet content

to 34.0% to 40.0% can increase rumen bacteria production, and consequently, milk protein yield [19].

The TKPER-R diet was ingested completely by wethers fed at 2% of BW on a DM basis and the DM intake was similar among the four treatments (Table 3), which indicated that TKPER was not less palatable for wethers than SB, SBM, or SBCR. The CP digestibility of TKPER-R was higher than that of SBCR-R ( $p < 0.05$ ), and the EE digestibility of TKPER-R was higher than that of SBM-R ( $p < 0.05$ ), which might have resulted from its relatively higher CP and EE contents. The aNDFom digestibility of TKPER-R was significantly lower than that of SBCR-R. This was attributed to the non-detected aNDFom in TKPER (Table 1). The NFC digestibility of TKPER-R was higher than that of SB-R ( $p < 0.05$ ), which could be explained by the higher NFC content of TKPER-R ( $p < 0.05$ ).

Regarding nitrogen balance, results implied that the TKPER-R diet, in which TKPER was mixed with ryegrass straw at a ratio of 1:1, did not have any effects on nitrogen balance, compared to the other experimental diets. The SBM feeding in SBM-R increased CP digestibility, excretion N in urine, retention N, and ruminal  $\text{NH}_3$ -N content in the present experiment, which was consistent with previous studies [20, 21]. The high CP content in SBM-R increased CP digestibility and ruminal  $\text{NH}_3$ -N content at 4 h after feeding. The highest urinary N in SBM-R suggested that excess nitrogen from the diet was mainly excreted in urine. The retention N, however, was not aggravated but improved by SBM feeding.

### Ruminal fermentation

In general, ruminal pH decreases following ingestion due to ruminal fermentation. The value, however, is controlled between 5.5 and 7.0 [22]. The values of TKPER-R, 7.05 and 6.62, were not significantly different from the values of SB-R or SBM-R at both sampling times (Table 4). This suggested that the TKPER feeding did not have a negative effect on ruminal pH in wethers. Meanwhile, the ruminal pH in SBCR-R showed acute decrease and was lower than that in SB-R and TKPER-R ( $p < 0.05$ ) at 4 h after feeding. The higher digestibility of aNDFom and NFC in SBCR-R compared to TKPER-R (aNDFom) and SB-R (NFC), respectively, might have produced this difference.

Higher ruminal C3 content and lower C2:C3 ratio at 4 h after feeding in wethers fed the TKPER-R diet (Table 4) were observed in this study. This was likely due to the relatively higher NFC content, digestibility, and lack of aNDFom content in TKPER. Bramley et al. indicated that high NFC percentage in diet provides conditions that favor the growth of bacteria fermenting sugars and starch [23]. Combining these results with the lower ruminal  $\text{NH}_3$ -N content in wethers fed TKPER-R diet at 4 h after feeding compared to before feeding, it is likely that ruminal microorganisms could utilize protein and energy efficiently for their growth. Higher NFC diet could provide more fermentable energy in the rumen, which should reduce ruminal ammonia

**Table 5.** Blood metabolites in wethers fed with the experimental diets

Item	SB-R	SBCR-R	SBM-R	TKPER-R	SEM
Glu (mg/dL)	59.50	62.75	65.25	60.50	2.69
TP (g/dL)	7.05	6.98	6.93	6.93	0.11
Alb (g/dL)	4.05	4.03	4.00	4.03	0.11
BUN (mg/dL)	34.43 <sup>ab</sup>	25.10 <sup>a</sup>	39.20 <sup>b</sup>	36.35 <sup>b</sup>	2.16
NEFA (mEq/L)	0.24	0.18	0.10	0.23	0.03
T-Chol (mg/dL)	122.25 <sup>b</sup>	115.25 <sup>b</sup>	71.25 <sup>a</sup>	100.25 <sup>ab</sup>	9.93
PL (mg/dL)	176.00 <sup>b</sup>	168.75 <sup>b</sup>	95.75 <sup>a</sup>	148.25 <sup>ab</sup>	12.72
AST (IU/L)	97.25	102.75	61.00	104.75	14.06
ALT (IU/L)	13.75	13.00	10.75	13.50	2.07
$\gamma$ -GTP (IU/L)	58.75	62.50	65.50	68.25	5.41
Ca (mg/dL)	9.43	10.10	9.50	9.10	0.60
IP (mg/dL)	6.95	5.88	6.05	7.63	1.01

SB-R, dry heat soybean with ryegrass straw on a dry matter basis; SBCR-R, dry soybean curd residue with ryegrass straw on a dry matter basis; SBM-R, soybean meal with ryegrass straw on a dry matter basis; TKPER-R, tamarind kernel powder extract residue with ryegrass straw on a dry matter basis; SEM, standard error of means; Glu, glucose; TP, total protein; Alb, albumin; BUN, blood urea nitrogen; NEFA, nonesterified fatty acid; T-Chol, total cholesterol; PL, phospholipids; AST, aspartate aminotransferase; ALT, alanine transaminase;  $\gamma$ -GTP,  $\gamma$ -glutamyl transpeptidase; Ca, calcium; IP, inorganic phosphorus. Means in the same row with different superscripts differ significantly ( $p < 0.05$ ).

concentrations [24]. In the present experiment, microorganisms could efficiently break down protein interacting with energy from TKPER and take advantage of  $\text{NH}_3\text{-N}$  faster after ingesting the TKPER-R diet.

### Blood metabolites

The wethers exhibited no visible symptoms of metabolic disorders during the experiment. The concentration of Glu, TP, Alb, NEFA, Ca, and IP were within the normal range of values (Glu, 44 to 81 mg/dL; TP, 6.0 to 7.9 g/dL; Alb, 2.7 to 3.7 g/dL; NEFA, <0.30 mEq/L; Ca, 9.0 to 11.5 mg/dL; IP, 4.0 to 7.3 mg/dL) [25]. Solomon et al reported that feeding diets with palm oil increased serum cholesterol [26]. The higher EE contents in the experimental diets might have contributed to higher plasma T-Chol and PL concentration.

The wethers in each treatment had higher serum BUN concentrations than the normal range of values (10 to 26 mg/dL) [25], since the CP contents in the experimental diets were high. SBCR-R had the lowest BUN due to its lower CP content, because BUN is affected by dietary levels of protein [27]. The concentrations of AST, ALT, and  $\gamma$ -GTP in plasma are indicators of liver functions. The concentrations among the four treatments did not show significant differences and were within the range of normal values (AST, 307 IU/L; ALT, 30 IU/L;  $\gamma$ -GPT, 17 to 69 IU/L) [25]. Hence, we concluded that liver function in the experimental wethers was normal.

## CONCLUSION

The present results indicate that TKPER did not have any side effect on digestibility, nitrogen balance and ruminal fermentation in wethers. It could be concluded that TKPER can be used as a high-protein source feed to be substituted for an expensive ingredient for growing livestock, or as a high-NFC nutrition energy source feed for lactating and fattening livestock. Due to the absence of fiber in TKPER, appropriate roughage source should be chosen together with TKPER to meet the fiber requirement of ruminants.

## CONFLICT OF INTEREST

We certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

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