# Characterization of Protein Degradation in Tropical Dairy Feedstuff Using the *In Sacco* Method

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#### ABSTRAK

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Penelitian ini bertujuan untuk menentukan karakteristik degradasi protein pada 27 bahan pakan tropis untuk ransum ternak perah dengan dua puluh tujuh jenis bahan pakan. Dua puluh dua bahan pakan tropis dikelompokkan menjadi A1 (pakan lokal rendah serat rendah protein: jagung, dedak padi, gaplek, onggok, gandum, dan pollard), A2 (pakan lokal rendah serat tinggi protein: bungkil inti sawit, ampas tahu, dan ampas tempe), dan A3 (pakan lokal tinggi serat: akasia, alfalfa, narra, gliricidia, indigofera, kaliandra, bauhinia, leucaena, albizia, agati, piper, kelor, dan daun jack), dibandingkan dengan A4 (pakan impor rendah serat tinggi protein: kedelai, kedelai sangrai, DDGS, CGM, dan CGF) menggunakan metode in sacco. Hasil penelitian menunjukkan bahwa A1, A2, dan A3 memiliki kandungan protein lebih rendah namun tinggi serat dibandingkan A4. Fraksi kelarutan protein (a) lebih tinggi pada A1 dan A2, sedangkan fraksi yang berpotensi terdegradasi (b) lebih tinggi pada pada A2 dan A3. A1 dan A2 memiliki fraksi RDP lebih tinggi daripada A3 dan A4. Pakan tinggi RDP meliputi pollard, gandum, kedelai, CGF, ampas tempe, alfalfa, amtoro, sengon, daun asam jawa, sirih hutan, daun nangka, kedelai sangrai, bungkil kedelai, CGM. Pakan tropis menunjukkan karakteristik degradasi protein yang beragam sehingga bermanfaat dalam memformulasikan ransum yang tepat pada sapi perah.

Kata Kunci: Hijauan, Konsentrat, Rumen Degradable Protein, Rumen Undegradable Protein, Sapi Perah

#### ABSTRACT

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A study was conducted to determine the protein degradation characteristics of 27 tropical feedstuffs for dairy rations. Twentytwo tropical feedstuffs were grouped into A1 (local low fiber and low protein sources: corn, rice bran, cassava, cassava waste, wheat, pollard), A2 (local low fiber and low protein sources: palm kernel meal, tofu waste, tempe waste), and A3 (local high fiber sources: acacia, alfalfa, narra, gliricidia, indigofera, calliandra, bauhinia, leucaena, albizia, agati, piper, moringa, jack leaves), and compared to A4 (imported low fiber high protein sources: soybean, roasted soybean, DDGS, CGM, CGF) using the *in sacco* method. The study revealed that A1, A2, and A3 had lower protein content but higher crude fiber than A4. Protein solubility (*a*) was higher in A1 and A2, while the potentially degraded fraction (*b*) was higher in A2 and A3. A1 and A2 had higher RDP fractions than A3 and A4. High RDP feedstuffs include pollard, wheat, soybean, CGF, tempe waste, alfalfa, gliricidia, indigofera, agati, and moringa. In contrast, high RUP feedstuffs include corn, palm kernel meal, narra, calliandra, leucaena, albizia, tamarind, piper, jack leaves, roasted soybean, soybean meal, and CGM. Tropical feedstuffs exhibit diverse protein degradation characteristics, making them valuable for strategic ration formulation in dairy cattle.

Key Words: Concentrate, Dairy Cattle, Forages, Rumen Degradable Protein, Rumen Undegradable Protein

### **INTRODUCTION**

The tropical regions have a high biodiversity potential and natural resources. The characteristics of tropical ecosystems are species richness in various taxa and complex biotic interactions among component species (Orians 2000). High species diversity allows for exploring plants, including grains and forages, that have the potential as livestock feed. Lee (2018) indicated that tropical forage plants have a lower nutritional value compared to those in temperate regions, with a 2% reduction in crude protein (CP), a 12% decrease in dry matter digestibility (DMD), and 49% organic matter digestibility (OMD). In contrast, tropical forages have higher fiber content, including 19% more neutral detergent fiber (NDF), 11% more acid detergent fiber (ADF), and 3% more acid detergent lignin (ADL). Hence, tropical feedstuffs have various qualities that fluctuate between the dry and rainy seasons (Despal et al., 2014). For instance, Napier grass (*Pennisetum purpureum*) has lower CP and DMD in the dry season (12.4% CP and 59.1% DMD) compared to the rainy season (13.7% CP and 61.8% DMD) (Evitayani et al. 2004). Conversely, *Leucaena leucocephala* shows

higher CP content during the dry season (24.19%) compared to the rainy season (21.75%) (Tahuk et al. 2018). Moyo and Nsahlai (2021) reported that feed degradability was highest in the cold (715 g/kg) and temperate regions (745 g/kg), compared to lower values in tropical (664 g/kg) and arid regions (621 g/kg).

Dairy cattle feed should provide mainly energy, protein, minerals, and vitamins. In the ruminant, the protein quality of feed is expressed by amino acid content, degradability, and digestibility (Patton et al. 2014). Rumen degradable protein (RDP) and rumen undegradable protein (RUP) are also used to determine the quality of protein for dairy cattle (NRC 2001). RDP expresses the amount of protein degraded by rumen microbes into ammonia (NH<sub>3</sub>), which then is used to synthesize microbial protein (Hristov et al. 2019) and is essential to maintain the balance of the microbial population (Uddin et al., 2015). RUP is feed protein that by-pass from the rumen. The RDP content of the feed is correlated with crude protein content but uncorrelated with crude fiber (Rosmalia et al. 2021). The Dairy National Research Council (NRC) standard (2001) recommends that the minimum RDP and RUP requirements be 60% and 40%, respectively.

The common feed used for dairy ration in Indonesia, namely concentrates and forages (Rosmalia et al. 2022; Sahroni et al. 2021), still adopts the dairy NRC standard for the formulation without considering RDP and RUP due to the lack of information on local feed. Concentrate feed supplies protein and energy (Woods et al. 2003), including agro-industrial by-products such as soybean meal, corn gluten meal, tofu waste, and palm kernel meal. By-product feeds have undergone mechanical or physical processing, especially heating, which can reduce soluble protein and RDP (Doiron et al. 2009). Cereal grains provide energy in the rumen and are a source of carbon skeletons for microbial protein synthesis when RDP is sufficient (Ferraretto et al., 2013; Rastgoo et al., 2020). Cereal grains provide protein and support the formulation of cost-effective dairy rations due to their relatively low prices, such as wheat pollard at 210 USD per metric ton compared to soybean meal at 500 USD per metric ton. Legume has a high protein content and is an alternative feedstuff in the tropical dairy ration to substitute protein content in feed concentrate (Castro-Montoya et al. 2019). A previous study reported that feeding dairy cattle a legume-based ration led to higher dry matter intake (DMI) by 1.3 kg/day and increased milk production by 1.6 kg/day compared to a grass-based ration (Johansen et al. 2018). The use of legumes in dairy cattle rations is limited by the low degradation in the rumen caused by crude fiber content and the presence of antinutrients (Jouan et al., 2020; Piluzza et al., 2014).

It is important to know the degraded and nondegraded fractions in the rumen related to the utilization of protein for rumen microbes and the host. Efforts to

increase the productivity of dairy cows through improved feed with a fulfillment approach based on protein adequacy and a balance of RDP and RUP will provide an accurate measurement of the estimated dairy production so that feed protein efficiency increases and costs incurred for reduced feed. Dairy ration based on RDP and RUP balance at the optimum level can also reduce nitrogen emission to the environment by improving nitrogen efficiency (Martins et al. 2019). The objective of this study was to compare degradation characteristics between tropical and imported (temperate origin) feed and to identify the RDP and RUP content in tropical feedstuff, including concentrate and forage protein, as an inventory of information on the nutritional content of the feed to further serve as a reference in preparing feed formulations and meeting nutrient needs for dairy cattle in Indonesia.

## MATERIALS AND METHODS

### **Tropical feedstuff preparation**

A total of 29 kinds of feedstuff, including concentrate and forage protein, were used in this study. Feedstuffs were grouped into A1 (local low fiber low protein sources), A2 (local low fiber high protein sources), A3 (local high fiber sources), and A4 (imported low fiber high protein sources). Local energy sources comprised corn, rice bran, cassava, cassava waste, wheat, and pollard. Protein sources include palm kernel meal, tofu, and tempe waste). Forage sources consisted of acacia (Acacia mangium), alfalfa (Medicago sativa), narra (Pterocarpus indicus), gliricidia (Gliricidia sepium). indigofera (Indigofera zollingeriana). calliandra (Calliandra calothyrsus), bauhinia (Bauhinia purpurea), leucaena (Leucaena leucocephala), albizia (Albizia chinensis), agati (Sesbania grandiflora), piper (Piper aduncum L.), moringa (Moringa oleifera), and jack leaves (Artocarpus heterophyllus). In contrast, imported sources consisted of soybean, roasted soybean, DDGS, CGM, and CGF.

The concentrate samples were ground through a 2mm screen for concentrate feedstuff and dried in an oven at 60 °C for 48 h. The leaves and stems commonly eaten by dairy cattle were taken as samples for forage sources. The forage samples were chopped and dried in the room for three days, then dried in an oven at 60 °C for 48 h before ground to a 2-mm size. All feedstuff was analyzed for nutrient composition, such as dry matter (DM), ash, crude protein (CP), ether extract (EE), crude fiber (CF), and nitrogen-free extract (NFE) using AOAC (2005) method, and gross energy (GE) was estimated according to Weiss and Tebbe (2019).

### In sacco degradability measurement

This research was conducted using the *in sacco* method according to NRC (2001). The samples were

weighed 5 g and put in a nylon bag (ANKOM, porosity  $\pm$ 50 µm), 5 x 10 cm for concentrate feed and 10 x 20 cm for forage protein feed (Despal et al. 2022; NRC 2001; Van Emon et al. 2015). Each sample consists of 3 bags. The nylon bag is tied with a rope and inserted into the fistulated dairy cattle rumen. The nylon bags were inserted before morning feeding. This study used two fistulated Friesian Holstein bulls (BW±510 kg). Dairy cattle were fed 2% of BW twice daily at 7.00 am and 3.00 pm. Diets contained 60% Napier grass (*Pennisetum purpureum*) and 40% concentrate mixture (%DM basis). The diets contained 10.25% CP, 20.70% CF, and 60% TDN.

The nylon bags were incubated in the rumen for 0, 3, 6, 9, 12, 15, 24, and 48 hours for concentrate feed, then until 72 h incubation for forage protein feed. For 0 h incubation, the nylon bags were only rinsed under tap water. The nylon bag contains a sample of feed at a predetermined time. The nylon bag was washed and dried at  $60^{\circ}$ C for 48 hours in an oven. The nylon bag was weighed, and the residual sample was separated for protein analysis using the Kjeldahl method (AOAC 2005). Crude protein disappearance (CPD) (%) was estimated as follows:

$$CPD = ((BW + S1) - (BW + RW))x \ 100 \ (S1xCP)$$
  
where BW is bag weight, RW is residue weight, S1 is  
sample weight, and CP is the crude protein of the original  
sample.

The degradation of protein or kinetic parameters was calculated based on an exponential equation according to the (Ørskov & Mcdonald 1979):

$$y = a + b(1 - e^{(-ct)})$$
(1)

Where y is protein disappearance in the rumen (%), a is a soluble fraction (%), b is an insoluble but potentially degradable fraction (%), c is degradation rate constant of the b fraction (%/h), t is degradation time (0, 3, 6, 9, 12, 15, 24, 48 and 72 h), e is base for natural logarithm. Potential degradation was calculated with the formula:

$$PD = a + b \tag{2}$$

The RDP and RUP were calculated, which refer to NRC (2001) with the following equation:

$$RDP = a + b \left[ \frac{c}{c+k} \right]$$
(3)

 $RUP = 100 - RDP \tag{4}$ 

where a, b, and c are the same as in equation (1), k is the rumen outflow rate, assumed to be 6%/h.

#### Statistical analysis

The nutrient composition and ruminal degradability of protein were analyzed descriptively. A One-Way Analysis of Variance (ANOVA) was performed using the SAS program (SAS Institute Inc., Cary, NC, USA) to evaluate the kinetic parameters and estimate RDP and RUP. The animals were treated as a block. Duncan's Multiple Range Test further tested the differences (P<0.05).

### **RESULTS AND DISCUSSION**

#### Nutrient composition

In this study, there were four groups of tropical dairy feedstuff consisting of local low-fiber and low-protein sources (A1), local low-fiber and high-protein sources (A2), local high-fiber sources (A3), and imported lowfiber and high protein sources (A4). The nutrient composition of the tropical dairy feedstuff varied depending on the feed sources presented in Table 1. The crude protein content (CP) of A4 was highest compared to other group feedstuff, with an average of 38.30%±14.26. The data showed that imported feed sources had higher protein content than local feed sources in both concentrate (low fiber) and forage (high fiber) categories. Moyo and Nsahlai (2021) reported that the CP content of feed from cold and temperate climates (imported protein sources) was higher than in tropical climates (local protein sources) due to rapid lignification in tropical climates.

In contrast, the A1 group as an energy source had the lowest protein and the highest nitrogen free-extract (NFE), averaging  $8.76\%\pm4.74$  and  $70.00\%\pm19.05$ , respectively. Storage carbohydrates act as energy sources, so the A1 group is supposed to have low protein content and is easily degradable (Klevenhusen and Zebeli 2021). The gross energy predicted using the Weiss and Tebbe (2019) equation showed that the A4 group had the highest energy density, driven by its high CP and ether extract (EE) content. In contrast, although A1 was rich in carbohydrates (NFE), its overall energy density was moderated by lower protein and fat levels, resulting in the lowest GE among the groups.

The A1 group is a low-fiber and low-protein feed source group primarily used as an energy source in formulating dairy cow rations. Its low protein content also influences the extent of protein degradation in the rumen due to the different carbohydrate components. Pollard has the highest protein content compared to other A1 feed ingredients. Besides being used as an energy source, pollard is also a protein source for dairy cows (Chuzaemi et al. 2020).

The CP content is closely related to effective rumen degradation (Fulkerson et al., 2007). Tofu waste, a feedstuff commonly used by smallholder dairy farmers, had the highest CP content with low crude fiber (CF) and high NFE compared to other A2 group feedstuff. For the A3 group, indigofera had a high CP, followed by gliricidia and leucaena. Meanwhile, tamarind had the lowest CP. However, the crude fiber (CF) content of A3 was lower than A2, 16.30%±5.32 and 25.98%±10.56, respectively. It is due to protein sources from by-product agroindustry and non-conventional feed containing high

		Nutrient composition							
Group	Feedstuff	DM (%)	Ash (%DM)	CP (%DM)	EE (%DM)	CF (%DM)	NFE (%DM)	GE (Mcal/kg) <sup>1</sup>	
A1	Corn	90.59	1.31	7.88	3.74	1.24	85.83	4.45	
	Rice bran	92.61	16.21	6.91	3.22	25.60	48.06	3.78	
	Cassava	91.17	1.78	5.18	0.43	1.60	91.02	4.22	
	Cassava waste	93.35	37.04	3.72	0.52	11.69	47.03	2.72	
	Wheat	89.52	1.50	12.78	1.73	3.63	80.36	4.41	
	Pollard	91.58	6.80	16.10	1.80	7.59	67.71	4.23	
Averag	;e±SD	91.47±1.38	10.77±14.08	8.76±4.74	1.91±1.36	8.56±9.25	70.00±19.05	3.97±0.65	
A2	Palm kernel meal	94.02	4.77	15.87	11.24	28.46	39.66	4.81	
	Tofu waste	92.68	3.47	18.80	4.50	14.40	58.83	4.55	
	Tempe waste	91.72	2.62	12.65	2.51	35.07	47.15	4.40	
Averag	e±SD	92.81±1.16	3.62±1.08	15.77±3.08	6.08±4.58	25.98±10.56	48.55±9.66	4.59±0.21	
A3	Acacia	24.89	4.68	16.63	1.93	19.04	50.04	4.34	
	Alfalfa	26.22	9.53	14.47	2.79	28.40	38.18	4.15	
	Narra	29.39	6.00	21.63	1.98	21.51	42.34	4.35	
	Gliricidia	19.86	9.46	25.18	2.25	14.15	41.34	4.27	
	Indigofera	24.16	9.32	26.18	2.37	12.52	40.23	4.30	
	Calliandra	31.41	5.78	21.39	1.55	15.84	48.41	4.34	
	Bauhinia	37.04	8.94	21.71	3.02	23.07	35.76	4.29	
	Leucaena	28.56	7.92	22.68	2.46	16.33	43.26	4.31	
	Albizia	34.55	4.61	17.56	2.31	14.02	54.64	4.37	
	Tamarind	39.21	7.12	11.60	1.99	18.51	55.30	4.17	
	Agati	23.40	8.39	19.69	2.54	10.50	50.82	4.26	
	Piper	20.98	18.53	21.69	1.47	10.44	39.54	3.80	
	Moringa	18.55	11.41	20.59	3.44	10.38	46.08	4.19	
	Jack leaves	29.91	11.12	15.53	0.86	13.42	51.89	4.00	
Average±SD		27.72±6.33	8.77±3.54	19.75±4.12	2.21±0.66	16.30±5.32	45.56±6.34	4.22±0.16	
A4	Soybean	92.67	6.13	34.78	13.59	11.99	33.50	5.14	
	Roasted soybean	94.32	6.23	38.92	15.31	9.55	30.00	5.28	
	Soybean meal	92.10	8.47	49.30	1.39	2.26	38.58	4.61	
	DDGS	88.77	5.77	29.81	6.15	6.73	51.55	4.69	
	CGM	92.94	1.74	58.63	2.51	0.72	36.40	5.08	
	CGF	90.42	4.90	18.37	2.49	10.13	64.11	4.38	
Averag	e±SD	91.87±1.98	5.54±2.21	38.30±14.26	6.91±6.08	6.90±4.54	42.36±12.95	5 4.86±0.35	

Table 1. Nutrient composition of tropical dairy feedstuff

DM= dry matter, CP= crude protein, EE= ether extract, CF= crude fiber, NFE= nitrogen-free extract, GE= gross energy, A1= local low fiber low protein sources, A2= local low fiber high protein sources, A3= local high fiber sources, A4= imported low fiber high protein sources. <sup>1</sup>GE was calculated using the equation proposed by Weiss and Tebbe (2019) with GE = (%CP x 0.056) + (%EE x 0.094) + ((100 - %CP - %EE - %ash) x 0.042)

fiber. Abdeltawab and Khattab (2018) reported that the CF content of palm kernel meal reached 24.90%, with ADF 43.70% and NDF 66.70%. Most feedstuffs in the A4 group had a high protein content except CGF due to the gluten (protein) cut off during the wet-milling process (Li et al. 2011).

# *The In Sacco* protein degradability of tropical dairy feedstuff

The ruminal CP degradation (CPD) of tropical dairy feedstuff at each incubation time is shown in Figure 1. The rise of CPD was followed by increasing incubation time. At 0 h incubation time, protein degradation showed the amount of soluble protein without incubation in the rumen. Protein solubility is the main factor determining proteolytic microbes' activity to access feed protein and degrade it (Bach et al. 2005). The data showed a wide variety of protein degradation at 0 h incubation. The range of protein degradation at 0 h incubation time for concentrate source (A1, A2, and A4) and fiber source (A3) were 0%-78% and 0%-32%, respectively. The highest protein degradation at 0 h incubation time for concentrate and forage sources was found in cassava meal and agati; this indicates that cassava meal and agati contain more soluble protein than other feeds.

In the A1 group, over 50% of CPD rice bran, cassava, cassava waste, wheat, and pollard had been degraded for three hours of incubation. Corn had to be degraded by over 50% CPD for 12 hours. Tempe waste had lower time incubation (3 h) to reach more than 50% CPD compared to tofu waste (9 h) and palm kernel meal (12 h). In the A4 group, CGF had more than 50% CPD with lower time incubation (3 h) rather than soybean and DDGS (9 h), roasted soybean and soybean meal (15 h), and CGM (48 h). According to Figure 1, the level of CPD between soybean and roasted soybean was different due to the heating process obtained by roasted soybean. The heating process in roasted soybeans causes low protein degradation (Petit et al. 2002). Heating treatment of feed reduced RDP by decreasing the soluble fraction (a) and the potential degradation (a+b) (Rosmalia et al. 2024). The low CPD in CGM and corn was due to CGM and corn coming from maize protected by a complex endosperm texture, starch structure, and starch granule shape associated with protein and fat, inhibiting rumen microbes from accessing and degrading the protein (Rastgoo et al. 2020). The characteristics of high-starch feed that interact with protein or fat take longer to be degraded (Menezes et al. 2019).

In the forage group, alfalfa, indigofera, agati, and moringa had reached 50% of CPD at 3 h incubation. Meanwhile, gliricidia and bauhinia got more than 50% of CPD at 9 h, piper at 12 h; jack leaves at 15 h, then narra and Leucaena at 48 h. In contrast, acacia and albizia have not reached 50% degradation after 72 h incubation. Protein degradation of forage protein depends on the part of the plant, fertilization rates, maturities, and antinutrients, which have different characteristics among forages (Elizalde et al. 1999). Acacia and albizia contained high tannin compared to agati and gliricidia (Alam et al. 2007; Yusiati et al. 2018).

# Kinetic of protein degradation on tropical dairy feedstuff

The kinetic degradation of protein in tropical dairy feedstuff is presented in Table 2. The values of a, b, and PD were significantly different among types of feed (p < 0.05), while the coefficient c was not significant (p > 0.05).The average soluble fraction (a) on A1 (32.06%) and A2 (30.96%) was higher than A3 (16.24%) and A4 (12.40%). In the A1 group, cassava was the highest value, followed by cassava waste, pollard, corn, wheat, and rice bran. The high soluble fraction of cassava is due to high nonstructural carbohydrate concentration (Daza et al., 2019). The most significant value in the A2 group was tempe waste compared to tofu waste and palm kernel meal. The high percentage of total carbohydrates in tempe waste can affect soluble fractions. Lee et al. (2017) revealed that the soluble fraction positively correlates with carbohydrates, especially non-fiber carbohydrate (NFC) content. The highest and lowest values in the A3 group were agati and acacia, respectively. The low value can be caused by the inability of rumen microbes and their enzymes to degrade the substrate at a certain level. The presence of associated nutrients (cross-linking bonds) that are difficult to degrade is also a limiting factor (Dijkstra et al. 2005). In the A4 group, CGF had the highest value, while soybean meal and roasted soybean had a lower value than other A4 feedstuff. The negative value in soybean meal and roasted soybean can be related to the loss of finer particles from the bags in this treatment instead of a higher solubility (Belachew et al. 2013).

The value of b was highest in A4 (90.03%) compared to A2 65.00%, A1 55.64%, and A3 59.10%; this is in line with Moyo and Nsahlai (2021) that the bvalue was higher for concentrate feed compared to roughages. Wheat and corn had a high b value, followed by pollard, rice bran, cassava waste, and cassava. Tofu waste had the highest b value compared with palm kernel meal and tempe waste. In the forage group, the b value was highest for piper, followed by moringa, jack leaves, indigofera, gliricidia, agati, leucaena, bauhinia, alfalfa, narra, tamarind, acacia, calliandra, and albizia. In the A4 group, soybean meal, roasted soybean, and soybean had a high b value. In contrast, DDGS, CGM, and CGF had the same level of *b* coefficient ranging from 35%-60%. The b values of soybean meal and roasted soybean were overestimated by more than 100%. Referring to Figure 1, this might be due to the low and slow degradation rate

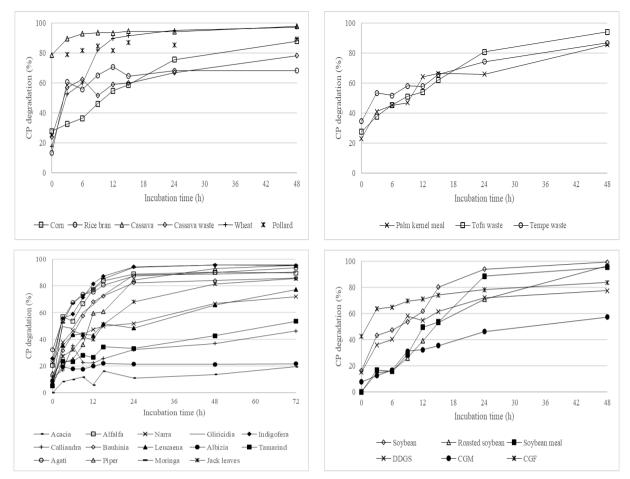


Figure 1. Ruminal crude protein degradation (CPD) of tropical feedstuff in the rumen

at the beginning of incubation for up to 15 hours of incubation time. After 24 hours of incubation, more than 70%–80% of the protein had been degraded in the rumen.

The PD value describes potential degradation. Imported protein sources had the highest PD value, followed by local protein, energy, and forage protein sources. Corn, wheat, and cassava had a higher potential degradation, followed by pollard, cassava waste, and rice bran. There is no difference in PD values among feedstuff in A4 (P>0.05). The potential degradation for tofu waste was highest compared to other A4 feeds. Piper, moringa, and agati had a high PD value, while albizia had the lowest.

The rate of degradation (c value) was not significantly different among types of feed, including the feedstuff in the group (P>0.05). The c value ranged from 0.04–0.63 h<sup>-1</sup>. However, corn's degradation rate tends to be low compared to other A1 feedstuff. According to Herrera-Saldana et al. (1990), corn contains high starch and has a low rate of starch degradation. Table 2 shows that the c value decreased for soybeans heated by roasting. The heating process is one way to protect feed protein from rumen degradation (Micek et al. 2020). Petit et al. (2002) reported that the rate of protein degradation decreased, and the concentration of RUP increased with heating temperature due to the Maillard reaction.

# Estimation of rumen degradable protein and rumen undegradable protein

Table 3 shows the estimation of RDP and RUP on tropical dairy feedstuff, including concentrate and forage protein. The data indicate that RDP and RUP values differed for all types of feed (P<0.05). A1 had the highest RDP value, followed by A2, A4, and A3. In contrast, A3 had the highest RUP values in comparison to other types. Almost 70% of the protein of A1 had degraded in the rumen used to serve the energy and the carbon skeleton for microbial protein synthesis (Rastgoo Cassava had the highest RDP value et al. 2020). compared to other A1 feedstuff. The high RDP is related to the degradability of dry matter in feedstuff. Wanapat and Kang (2015) reported that dry matter degradability for cassava, cassava waste, rice bran, and corn were 92.5%, 63.6%, 63.0%, and 59.3%, respectively. A1 feeds contain carbohydrates, including sugar, starch, fructan, and pectin (Villalba et al., 2021). It was reported that cassava had a high pectin substance

Group	Feedstuff	Coefficient of kinetic parameters <sup>1</sup>					
Group	Feedstuff	a (%)	<i>b</i> (%)	PD (%)	<i>c</i> (h <sup>-1</sup> )		
	Corn	24.59 <sup>bc</sup>	75.94ª	100.54 <sup>a</sup>	0.04		
	Rice bran	13.84°	52.74 <sup>bc</sup>	66.58°	0.63		
. 1	Cassava	78.83 <sup>a</sup>	16.29 <sup>d</sup>	95.12ª	0.40		
A1	Cassava waste	31.40 <sup>b</sup>	42.54°	73.95°	0.11		
	Wheat	17.96 <sup>bc</sup>	79.89ª	97.85ª	0.16		
	Pollard	25.42 <sup>bc</sup>	59.86 <sup>b</sup>	85.29 <sup>b</sup>	0.84		
Average±SD		32.06±24.15ª	55.64±23.37 <sup>b</sup>	87.70±13.38 <sup>ab</sup>	0.39±0.36		
	Palm kernel meal	26.00 <sup>b</sup>	60.44 <sup>b</sup>	86.44 <sup>b</sup>	0.06		
A2	Tofu waste	27.53 <sup>b</sup>	79.69ª	107.22 <sup>a</sup>	0.04		
	Tempe waste	39.36 <sup>a</sup>	54.87 <sup>b</sup>	94.23 <sup>ab</sup>	0.04		
Average±SD		30.96±6.66ª	65.00±11.79 <sup>ab</sup>	95.96±9.72 <sup>ab</sup>	0.05±0.02		
	Acacia	6.25 <sup>e</sup>	$30.88^{\mathrm{f}}$	37.13 <sup>gh</sup>	0.29		
	Alfalfa	17.23 <sup>bcd</sup>	50.37°	67.60 <sup>d</sup>	0.12		
	Narra	15.75 <sup>bcd</sup>	43.94 <sup>e</sup>	59.68 <sup>ef</sup>	0.06		
	Gliricidia	22.26 <sup>ab</sup>	54.37 <sup>cde</sup>	76.63 <sup>bc</sup>	0.11		
	Indigofera	21.04 <sup>bc</sup>	61.99 <sup>bcd</sup>	83.03 <sup>ab</sup>	0.13		
	Calliandra	15.37 <sup>bcd</sup>	27.91 <sup>f</sup>	43.28 <sup>g</sup>	0.05		
	Bauhinia	14.26 <sup>bcde</sup>	50.99 <sup>de</sup>	65.25 <sup>de</sup>	0.11		
A3	Leucaena	17.15 <sup>bcd</sup>	51.27 <sup>de</sup>	68.42 <sup>d</sup>	0.07		
	Albizia	12.44 <sup>cde</sup>	22.22 <sup>f</sup>	34.65 <sup>h</sup>	0.39		
	Tamarind	21.95 <sup>ab</sup>	$32.94^{\mathrm{f}}$	54.89 <sup>f</sup>	0.10		
	Agati	29.69 <sup>a</sup>	55.00 <sup>cde</sup>	84.69ª	0.12		
	Piper	9.18 <sup>de</sup>	79.90ª	89.09ª	0.06		
	Moringa	20.60 <sup>bc</sup>	66.24 <sup>b</sup>	86.84ª	0.16		
	Jack leaves	12.21 <sup>cde</sup>	63.51 <sup>bc</sup>	75.72°	0.05		
Average±SD		16.24±9.91 <sup>b</sup>	59.10±24.64 <sup>b</sup>	75.35±27.60 <sup>b</sup>	0.28±0.97		
	Soybean	18.78 <sup>b</sup>	86.01 <sup>bc</sup>	104.80	0.07		
	Roasted soybean	-5.52 <sup>d</sup>	143.81 <sup>ab</sup>	138.30	0.04		
	Soybean meal	-6.57 <sup>d</sup>	157.61 <sup>a</sup>	151.05	0.03		
A4	DDGS	16.62 <sup>b</sup>	60.51°	77.13	0.10		
	CGM	5.81°	56.94°	62.75	0.05		
	CGF	45.30 <sup>a</sup>	35.28°	80.58	0.15		
Average±SD		12.40±18.82 <sup>b</sup>	90.03±50.53ª	102.43±38.44ª	$0.07 \pm 0.05$		

Table 2. Kinetic degradation of protein

<sup>1</sup>Means in the same row with different superscripts differ significantly (P<0.05). a= soluble fraction, b= insoluble but potentially degradable fraction, PD= potential degradation, c= degradation rate constant of the b fraction, A1= local low fiber low protein sources, A2= local low fiber high protein sources.

Group	Feedstuff	CP (%DM)	RDP (%CP)	RUP (%CP)
	Corn	7.88	55.00°	45.00 <sup>a</sup>
	Rice bran	6.91	61.42°	38.58 <sup>a</sup>
A1	Cassava	5.18	92.61ª	7.39 <sup>c</sup>
AI	Cassava waste	3.72	59.19°	40.81 <sup>a</sup>
	Wheat	12.78	76.33 <sup>b</sup>	23.67 <sup>b</sup>
	Pollard	16.10	$80.77^{b}$	19.23 <sup>b</sup>
Average±SD		8.76±4.74	71.95±14.32 <sup>a</sup>	28.05±14.32 <sup>b</sup>
	Palm kernel meal	15.87	57.04 <sup>b</sup>	42.96 <sup>a</sup>
A2	Tofu waste	18.80	59.19 <sup>ab</sup>	40.81 <sup>ab</sup>
	Tempe waste	12.65	62.08 <sup>a</sup>	37.92 <sup>b</sup>
Average±SD		15.77±3.08	59.43±3.05 <sup>ab</sup>	40.57±3.05 <sup>ab</sup>
	Acacia	16.63	10.46 <sup>g</sup>	89.54ª
	Alfalfa	14.47	69.79 <sup>a</sup>	30.21 <sup>g</sup>
	Narra	21.63	$47.49^{d}$	52.51 <sup>d</sup>
	Gliricidia	25.18	64.08 <sup>ab</sup>	$35.92^{\mathrm{fg}}$
	Indigofera	26.18	73.88 <sup>a</sup>	26.12 <sup>g</sup>
	Calliandra	21.39	26.79 <sup>ef</sup>	73.21 <sup>bc</sup>
A 2	Bauhinia	21.71	58.89 <sup>bc</sup>	41.11 <sup>ef</sup>
A3	Leucaena	22.68	46.15 <sup>d</sup>	53.85 <sup>d</sup>
	Albizia	17.56	19.38 <sup>fg</sup>	80.62 <sup>ab</sup>
	Tamarind	11.60	28.90 <sup>e</sup>	71.10°
	Agati	19.69	72.64 <sup>a</sup>	27.36 <sup>g</sup>
	Piper	21.69	52.59 <sup>cd</sup>	47.41 <sup>de</sup>
	Moringa	20.59	72.41ª	27.59 <sup>g</sup>
	Jack leaves	15.53	46.45 <sup>d</sup>	53.55 <sup>d</sup>
Average±SD		19.75±4.12	49.28±21.07 <sup>b</sup>	50.72±21.07ª
	Soybean	34.78	65.31 <sup>ab</sup>	34.69 <sup>cd</sup>
	Roasted soybean	38.92	42.92 <sup>cd</sup>	57.08 <sup>ab</sup>
A 4	Soybean meal	49.30	50.72 <sup>bc</sup>	49.28 <sup>bc</sup>
A4	DDGS	29.81	54.27 <sup>bc</sup>	45.73 <sup>bc</sup>
	CGM	58.63	31.95 <sup>d</sup>	68.05ª
	CGF	18.37	69.85 <sup>a</sup>	30.15 <sup>d</sup>
Average±SD		38.30±14.26	52.50±15.08 <sup>b</sup>	47.50±15.08ª

Table 3. Estimation of RDP and RUP on tropical dairy feedstuff

 $\overline{CP}$ = crude protein, RDP= rumen degradable protein, RUP= rumen undegradable protein, A1= local low fiber low protein sources, A2= local low fiber high protein sources, A3= local high fiber sources, A4= imported low fiber high protein sources

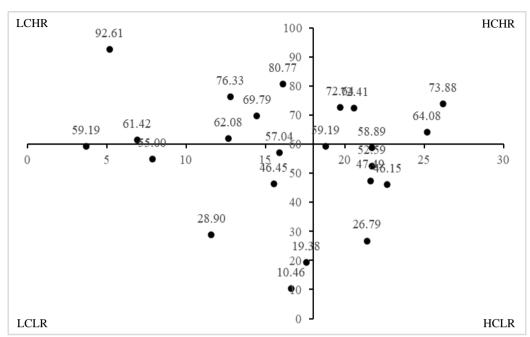


Figure 2. Tropical dairy feedstuff groups based on CP and RDP content

(Staack et al. 2019), and corn was rich in starch, approximately 70% (Hall et al. 2010). Zhao et al. (2015) revealed that the degradation rate for feedstuffs rich in pectin was higher than for feedstuffs rich in starch. This is in line with this study's finding that the RDP value of cassava was higher than corn.

Indigofera, agati, moringa, alfalfa, and gliricidia had high RDP values, followed by bauhinia, piper, narra, jack leaves, leucaena, tamarind, calliandra, albizia, and acacia. Putri et al. (2019) also showed that the RDP value of indigofera (74.72%) was high compared to gliricidia and leucaena. It indicates that indigofera can be a forage protein source with a high RDP content. The RDP value of alfalfa in this study (69.79%) was lower than that reported by Elizalde et al. (1999), ranging from 72.9%-81.1% at different stages of maturity (midvegetative, early bud, early flowering, and late flowering). The high RUP of acacia was due to its high fiber content. Abdulrazak et al. (2000) reported that acacia had a high proportion of ADF fraction, which means high cellulose and lignin content. Also, the CP is negatively correlated with ADF and the polyphenolic compound.

For A4, CGF had the highest RDP value compared to other imported feedstuff. Maskal'ová et al. (2014) reported that the RDP content of soybean meal, CGM, and CGF were 71.0%, 20.3%, and 74.8%, respectively. The data shows that the roasted soybean's RUP value was higher than the soybean's due to the heating process. A previous study revealed that soybeans under heat treatment had a higher RUP value than unheated soybeans (Petit et al. 2002). Tempe waste had the highest RDP value in A2, while palm kernel meal had the lowest. The low RDP value of palm kernel meal is due to its high crude fiber content (Abdeltawab and Khattab 2018). Crude fiber is negatively correlated to the RDP value of feed (Rosmalia et al. 2021).

The quality of feed protein sources is distinguished by their CP content and a balanced RDP and RUP. Feed protein sources contained a minimum protein of 18%-20%, and the minimum RDP and RUP contents were 60% and 40% (NRC 2001). Based on CP and RDP content, tropical dairy feedstuff can be divided into 4 groups, which are shown in Figure 2. The four groups are LPLR (low CP and low RDP), HCLR (high CP and low RDP), LCHR (low CP and high RDP), and HCHR (high CP and high RDP). For concentrate feed, the LCPLR group is corn, cassava waste, and palm kernel meal; LCHR includes cassava, rice bran, wheat, pollard, and tempe waste; HCLR is a soybean meal, roasted soybean, DDGS, CGM, and tofu waste; HCHR is soybean and CGF. For forage feed, LCLR includes acacia, albizia, tamarind, and jack leaves; LCHR is alfalfa; HCLR is narra, calliandra, bauhinia, leucaena, and piper; HCPHR includes gliricidia, indigofera, moringa, and agati. Some factors that influence RDP level are the types of protein, the proportion of NPN, the physical and chemical properties of the protein, the presence of disulfide bonds and cross-linking, antinutrients, retention time, rumen pH, and proteolytic microbial activity (Bach et al. 2005; Broderick et al. 1991; Doiron et al. 2009).

#### CONCLUSION

The degradability and the RDP and RUP content of tropical dairy feedstuff vary and differ among types of feed. Most energy feed sources are highly degraded, so they should be combined with high RDP from the protein feed sources. Forage protein could be added to dairy ration as a source of RUP. Furthermore, this information can be used in dairy formulation by considering the protein degradation of feedstuff.

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#### REFERENCES

- Abdeltawab AM, Khattab MSA. 2018. Utilization of palm kernel cake as a ruminant feed for animal: A review. Asian J Biol Sci. 11:157–164. DOI:10.3923/ajbs.2018.157.164.
- Abdulrazak SA, Fujihara T, Ondiek JK, Ørskov ER. 2000. Nutritive evaluation of some Acacia tree leaves from Kenya. Anim Feed Sci Technol. 85:89–98. DOI:10.1016/S0377-8401(00)00133-4.
- Alam MR, Amin MR, Kabir AKMA, Moniruzzaman M, McNeill DM. 2007. Effect of tannins in Acacia nilotica, Alblzia procera, and Sesbanla aculeate foliage determined *in vitro*, *in sacco*, and *in vivo*. Asian Austral J Anim Sci. 20:220–228. DOI:10.5713/ajas.2007.220
- AOAC. 2005. Official Methods of Analysis. 18<sup>th</sup> ed. Horwitz W, Latimer GW, editors. Gaithersburg, Maryland.
- Bach A, Calsamiglia S, Stern MD. 2005. Nitrogen metabolism in the rumen. J Dairy Sci. 88:E9–E21. DOI:10.3168/jds.S0022-0302(05)73133-7.
- Belachew Z, Yisehak K, Taye T, Janssens GPJ. 2013. Chemical composition and *in sacco* ruminal degradation of tropical trees rich in condensed tannins. Czech J Anim Sci. 58:176–192. DOI:10.17221/6712-CJAS.
- Broderick GA, Wallace RJ, Ørskov ER. 1991. Control of rate and extent of protein degradation. In: Physiol Asp Dig Metab Ruminants. pp. 541–592. DOI:10.1016/b978-0-12-702290-1.50030-8.
- Castro-Montoya J, Gownipuram R, Mendoza M, Solano N, López F, Dickhöfer U, Corea EE. 2019. Effects of feeding tropical forage legumes on nutrients digestibility, nitrogen partitioning and performance of crossbred milking cows. Anim Feed Sci Technol. 247:32–40. DOI:10.1016/j.anifeedsci.2018.10.017.
- Chuzaemi S, Mashudi, Eryantristan H, Huda AN. 2020. Effect of pollard and soybean meal protected with condensed tannin in concentrate on *in vitro* gas production. In: IOP Conf Ser: Earth Environ Sci. 478(012053). DOI:10.1088/1755-1315/478/1/012053.
- Daza J, Benavides D, Pulido R, Balocchi O, Bertrand A, Keim J. 2019. Rumen *in vitro* fermentation and *in situ*

degradation kinetics of winter forage brassicas crops. Animals 9:1–14. DOI:10.3390/ani9110904.

- Despal D, Alifianty OF, Pratama AP, Febrianti F, Evvyernie D, Wijayanti I, Nuraina N, Agustiyani I, Rosmalia A. 2022. *In situ* degradation of dairy cattle feedstuffs using reusable local nylon fabric bags. Vet World. 15:2234– 2243. DOI: 10.14202/vetworld.2022.2234-2243.
- Despal, Malyadi J, Destianingsih Y, Lestari A, Hartono H, Abdullah L. 2014. Seasonal feeding practice impact on lactating cow performances kept in Bogor lowland small enterprise dairy farming. In: Proc 16th AAAP Anim Sci Congr. Vol. II. Yogyakarta, Indonesia.
- Dijkstra J, Forbes JM, France J. 2005. Quantitative Aspects of Ruminant Digestion and Metabolism. CABI.
- Doiron K, Yu P, McKinnon JJ, Christensen DA. 2009. Heatinduced protein structure and subfractions in relation to protein degradation kinetics and intestinal availability in dairy cattle. J Dairy Sci. 92:3319–3330. DOI:10.3168/jds.2008-1946.
- Elizalde JC, Merchen NR, Faulkner DB. 1999. In situ dry matter and crude protein degradation of fresh forages during the spring growth. J Dairy Sci. 82:1978–1990. DOI:10.3168/jds.S0022-0302(99)75434-2.
- Evitayani, Warly L, Fariani A, Ichinohe T, Fujihara T. 2004. Seasonal changes in nutritive value of some grass species in West Sumatra, Indonesia. Asian Austral J Anim Sci. 17:1663–1668. DOI:10.5713/ajas.2004.1663.
- Ferraretto LF, Crump PM, Shaver RD. 2013. Effect of cereal grain type and corn grain harvesting and processing methods on intake, digestion, and milk production by dairy cows through a meta-analysis. J Dairy Sci. 96:533–550. DOI:10.3168/jds.2012-5932.
- Fulkerson WJ, Neal JS, Clark CF, Horadagoda A, Nandra KS, Barchia I. 2007. Nutritive value of forage species grown in the warm temperate climate of Australia for dairy cows: Grasses and legumes. Livest Sci. 107:253–264. DOI:10.1016/j.livsci.2006.09.029.
- Hall MB, Larson CC, Wilcox CJ. 2010. Carbohydrate source and protein degradability alter lactation, ruminal, and blood measures. J Dairy Sci. 93:311–322. DOI:10.3168/jds.2009-2552.
- Herrera-Saldana RE, Huber JT, Poore MH. 1990. Dry matter, crude protein, and starch degradability of five cereal grains. J Dairy Sci. 73:2386–2393. DOI:10.3168/jds.S0022-0302(90)78922-9.
- Hristov AN, Bannink A, Crompton LA, Huhtanen P, Kreuzer M, McGee M, Nozière P, Reynolds CK, Bayat AR, Yáñez-Ruiz DR, Dijkstra J, Kebreab E, Schwarm A, Shingfield KJ, Yu Z. 2019. Invited review: Nitrogen in ruminant nutrition: A review of measurement techniques. J Dairy Sci. 102:5811–5852. DOI:10.3168/jds.2018-15829.
- Johansen M, Lund P, Weisbjerg MR. 2018. Feed intake and milk production in dairy cows fed different grass and legume species: A meta-analysis. Animal 12:66–75. DOI:10.1017/S1751731117001215.

- Jouan J, Ridier A, Carof M. 2020. Legume production and use in feed: Analysis of levers to improve protein selfsufficiency from foresight scenarios. J Cleaner Prod. 274:123085.DOI:10.1016/j.jclepro.2020.123085.
- Klevenhusen F, Zebeli Q. 2021. A review on the potentials of using feeds rich in water-soluble carbohydrates to enhance rumen health and sustainability of dairy cattle production. J Sci Food Agric. 101:5737–5746. DOI:10.1002/jsfa.11358.
- Lee MA. 2018. A global comparison of the nutritive values of forage plants grown in contrasting environments. J Plant Res. 131:641–654. DOI:10.1007/s10265-018-1024-y.
- Lee YH, Kim YI, Oh YK, Ahmadi F, Kwak WS. 2017. Yield survey and nutritional evaluation of garlic stalk for ruminant feed. J Anim Sci Technol. 59:1–7. DOI:10.1186/s40781-017-0147-3.
- Li Q, Gao Y, Cao Y, Feng Z, Li J. 2011. Effects of rumendegradable protein balance on rumen fermentation in continuous culture fermenters. Front Agric China. 5:598–604. DOI:10.1007/s11703-011-1138-7.
- Martins CMMR, Fonseca DCM, Alves BG, Arcari MA, Ferreira GC, Welter KC, Oliveira CAF, Rennó FP, Santos MV. 2019. Effect of dietary crude protein degradability and corn processing on lactation performance and milk protein composition and stability. J Dairy Sci. 102:4165– 4178. DOI:10.3168/jds.2018-15553.
- Maskaľová I, Vajda V, Krempaský M, Bujňák L. 2014. Rumen degradability and ileal digestibility of proteins and amino acids of feedstuffs for cows. Acta Vet Brno 83:225–231. DOI:10.2754/avb201483030225.
- Menezes ACB, Valadares Filho SC, Carneiro Pacheco MV, Pucetti P, Pereira JMV, Rotta PP, Zanetti D, Silva BC, Costa E Silva LF, Detmann E, Neville TL, Caton JS. 2019. Single point ruminal incubation times necessary to estimate rumen degradable protein content in concentrate feeds. Transl Anim Sci. 3:1686–1690. DOI:10.1093/tas/txz058.
- Micek P, Słota K, Górka P. 2020. Effect of heat treatment and heat treatment in combination with lignosulfonate on *in situ* rumen degradability of canola cake crude protein, lysine, and methionine. Can J Anim Sci. 100:165–174. DOI:10.1139/cjas-2018-0216.
- Moyo M, Nsahlai I. 2021. Consequences of increases in ambient temperature and effect of climate type on digestibility of forages by ruminants: A meta-analysis in relation to global warming. Animals 11:1–17. DOI:10.3390/ANI11010172.
- NRC. 2001. Nutrient Requirements of Dairy Cattle. 7<sup>th</sup> Ed. Washington DC: National Academy Press.
- Orians GH. 2000. Biodiversity and ecosystem processes in tropical ecosystems. Rev Biol Trop. 48:297–303.
- Ørskov ER, Mcdonald I. 1979. The estimation of protein degradability in the rumen from incubation measurements weighted according to rate of passage. J Agric Sci. 92:499–503. DOI:10.1017/S0021859600063048.

- Patton RA, Hristov AN, Lapierre H. 2014. Protein feeding and balancing for amino acids in lactating dairy cattle. Vet Clin Food Anim. 30:599–621. DOI:10.1016/j.cvfa.2014.07.005.
- Petit HV, Tremblay GF., Tremblay E, Nadeau P. 2002. Ruminal biohydrogenation of fatty acids, protein degradability, and dry matter digestibility of flaxseed treated with different sugar and heat combinations. Can J Anim Sci. 82:241–250. DOI:10.4141/A01-083.
- Piluzza G, Sulas L, Bullitta S. 2014. Tannins in forage plants and their role in animal husbandry and environmental sustainability: A review. Grass Forage Sci. 69:32–48. DOI:10.1111/gfs.12053.
- Putri EM, Zain M, Warly L, Hermon H. 2019. In vitro evaluation of ruminant feed from West Sumatera based on chemical composition and content of rumen degradable and rumen undegradable proteins. Vet World. 12:1478–1483. DOI:10.14202/vetworld.2019.1478-1483.
- Rastgoo M, Kazemi-Bonchenari M, HosseinYazdi M, Mirzaei M. 2020. Effects of corn grain processing method (ground versus steam-flaked) with rumen undegradable to degradable protein ratio on growth performance, ruminal fermentation, and microbial protein yield in Holstein dairy calves. Anim Feed Sci Technol. 269:114646.

https://doi.org/10.1016/j.anifeedsci.2020.114646

- Rosmalia A, Dewi NA, Permana IG, Despal. 2022. Reformulation of dairy cattle concentrate based on rumen degradable protein to undegradable protein ratio at different energy levels: *in vitro* study. In: IOP Conf Ser: Earth Environ Sci. 1020(1). DOI:10.1088/1755-1315/1020/1/012008.
- Rosmalia, A, Permana, IG, Despal, Zahera R. 2021. Estimation rumen degradable protein of local feeds in dairy cattle using in sacco method. In: IOP Conf Ser: Earth Environ Sci. 883:012010. DOI:10.1088/1755-1315/883/1/012010.
- Rosmalia A, Permana IG, Despal, Toharmat T. 2024. In sacco and in vitro evaluation of heating and formaldehyde treated protein feed. Am J Anim Vet Sci. 19(1): 74–85. DOI:10.3844/ajavsp.2024.74.85
- Sahroni WP, Permana IG, Despal. 2021. Reformulation of dairy cow diets based on rumen degradable protein and total digestible nutrient with varying levels on *in vitro* fermentability and digestibility. In: IOP Conf Ser: Earth Environ. Sci. 888 012075. DOI:10.1088/1755-1315/888/1/012075.
- Staack L, Della Pia EA, Jørgensen B, Pettersson D, Rangel Pedersen N. 2019. Cassava cell wall characterization and degradation by a multicomponent NSP-targeting enzyme (NSPase). Sci Rep. 9:1–11. DOI:10.1038/s41598-019-46341-2.
- Tahuk PK, Baliarti E, Budhi, SPS, Panjono P. 2018. The effect of season on the feed quantity and quality and growth performance of male Bali cattle fattened in smallholder

farms. Bul Peternak. 42:203–209. DOI:10.21059/buletinpeternak.v42i3.33058

- Uddin MJ, Khandaker ZH, Khan MJ, Khan MMH. 2015. Dynamics of microbial protein synthesis in the rumen - A Review. Ann Vet Anim Sci. 2:116–131.
- Van Emon ML, Loy DD, Hansen SL. 2015. Determining the preference, *in vitro* digestibility, *in situ* disappearance, and grower period performance of steers fed a novel algae meal derived from heterotrophic microalgae. J Anim Sci. 93:3121–3129. DOI:10.2527/jas.2014-8654.
- Villalba JJ, Ates S, MacAdam JW. 2021. Non-fiber carbohydrates in forages and their influence on beef production systems. Front Sustain Food Syst. 5:1–12. DOI:10.3389/fsufs.2021.566338.
- Wanapat M, Kang S. 2015. Cassava chip (*Manihot esculenta* Crantz) as an energy source for ruminant feeding. Anim Nutr 1:266–270. DOI:10.1016/j.aninu.2015.12.001.

- Weiss W, Tebbe A. 2019. Estimating digestible energy values of feeds and diets and integrating those values into net energy systems. Transl Anim Sci. 3:953–961. DOI:tas/txy119.
- Woods VB, Moloney AP, O'Mara FP. 2003. The nutritive value of concentrate feedstuffs for ruminant animals Part II: *In situ* ruminal degradability of crude protein. Anim Feed Sci Technol. 110:131–143. DOI:10.1016/S0377-8401(03)00222-0.
- Yusiati LM, Kurniawati A, Hanim C, Anas MA. 2018. Protein binding capacity of different forages tannin. IOP Conf Ser: Earth Environ Sci. 119(1). DOI:10.1088/1755-1315/119/1/012007.
- Zhao XH, Gong JM, Zhou S, Fu CB, Liu CJ, Xu LJ, Pan K, Qu MR. 2015. Effects of degradable protein and non-fibre carbohydrates on microbial growth and fermentation in the rumen simulating fermenter (Rusitec). Ital J Anim Sci. 14:220–225. DOI:10.4081/ijas.2015.3771.