

Protein Protection to Increase Ruminant Feed Protein Efficiency *in Vitro*

Wahidin Teguh Sasongko^{1*}, Firsoni¹, Teguh Wahyono¹

¹National Research and Innovation Agency, Indonesia ^{*}Corresponding author. Email: wteguhs@gmail.com

ABSTRACT

This study aimed to determine the effect of protein protection to increase ruminant feed protein efficiency in vitro. The benefit of making this protected protein is that it can provide information to farmers that the provision of protein can be more efficient. The material used as protein protection is the use of tannins from jackfruit (*Arthocarpus heterophilus*) leaves. The experimental design used was a completely randomized design with 4 treatments and 6 replications. Treatment 1: control (tithonia); treatment 2: addition of 0.7% condensed tannins + tithonia; treatment 3: addition of 1.7% condensed tannins + tithonia; treatment 4: addition of 2.1% condensed tannins + tithonia. Parameters observed: degree of acidity (pH); dry matter digestibility (DMD); organic matter digestibility (OMD); Crude protein digestibility (CPD). The results showed that the highest degree of acidity (pH), DMD, OMD and CPD were 7.15; 70.42%, 78.84% and 66.26%. This study concluded that the treatment with the addition of 0.7% condensed tannins, gave a total CPD of 47.58% more efficient than the control and not significantly different from other treatments.

Keywords: Tannins, Condensed Tannins, Protein, In vitro, dry matter digestibility, organic matter digestibility, crude protein digestibility.

1. INTRODUCTION

Protein is one of the important nutritional elements in animal feed. This protein content is one source of nitrogen from feed that can be used for microbial protein synthesis in the rumen [1]. Classification of proteins in terms of structure is divided into two major groups, namely simple proteins and combined proteins. Simple proteins are proteins that consist of amino acid molecules, while combined proteins are proteins that consist of proteins and non-protein groups. Simple proteins are divided into two parts according to their molecular shape, namely fiber proteins and globular proteins.

The main source of essential amino acids for ruminants is microbial protein and protein that is not degraded in the rumen. Microbial proteins and feed proteins that are not degraded in the rumen enter the abomasum and are degraded by digestive enzymes into amino acids and short-chain peptides, which will then be absorbed by livestock for protein synthesis. Protein synthesis is used to meet the needs of life, growth, reproduction and production of both meat and milk.

Classification of proteins in terms of their structure is divided into two major groups, namely simple proteins and combined proteins. Simple proteins are proteins that consist of amino acid molecules, while combined proteins are proteins that consist of proteins and non-protein groups. Simple proteins are divided into two parts according to their molecular shape, namely fiber proteins and globular proteins [3].

Methods to protect the protein from microbial degradation in the rumen have been developed, including the addition of tannins. Several studies have shown that the addition of low concentrations of tannins in feed has the potential to increase the efficiency of fermentation in the rumen by maximizing the formation of microbial protein [4] and increasing the efficiency of protein utilization by ruminants [5]. Application of condensed tannins with low concentrations between 10 – 40 g/kg dry matter (DM), will provide the advantage of improving productivity [6]. Another study stated that

Table 1. Data analysis of the nutrition content of jackfruit leaves and *Tithonia diversifoila*

	Leaf	DM (%)	Ash (%)	CP (%)	EE (%)	CF (%)
	Jackfruit leaf	91.18	14.26	10.79	2.77	19.23
	Tithonia leaf	92.13	14.21	24.79	-	-
<u>`</u>						

Dry Matter (DM), Crude Protein (CP), Ether extract (EE), Crude Fiber (CF)

1 g of condensed tannins could bind 28.89 g of BSA protein [7].

This in vitro study is useful for formulating ruminant feed by utilizing protein bypass from jackfruit leaf tannins. It is hoped that this research will also be useful for breeders, because feeding ruminants with high protein content will be more efficient.

2. MATERIALS AND METHODS

This research was conducted in the laboratory of the Research and Technology Center for Isotope and Radiation Applications (PRT-AIR), Nuclear Power Research Organization (OR-TN), National Research and Innovation Agency (BRIN).

The research material used as protein protection was tannin from jackfruit (*Arthocarpus heterophilus*) leaves. While the protein source of feed used is paitan leaf (*Tithonia diversifolia*).

The initial research was carried out by analyzing the nutritional content of jackfruit leaves and paitan leaves [8], then continued with an analysis of jackfruit leaf tannins (measurement of total phenols, total non-tannin phenols and condensed tannins) [9].

The next stage is an in vitro test using the Tilley and Terry 2 stage method, the first stage is the stage where the feed ingredients are in the rumen. The second stage is the stage where the feed ingredients are in the abomasums [10]. The experimental design used was a complete randomized design with 4 treatments and 6 replications.

The in vitro test treatments used are as follows:

- 1. Control (tithonia)
- 2. 0.7% condensed tannins + tithonia
- 3. 1.4% condensed tannins + tithonia
- 4. 2.1% condensed tannins + tithonia

Parameters observed: degree of acidity (pH); dry matter digestibility (DMD); organic matter digestibility (OMD) and crude protein digestibility (CPD).

3. RESULTS AND DISCUSSION

The nutritional content of samples of jackfruit leaves and tithonia leaves which include dry matter (DM), organic matter (OM), crude protein (CP), ether extract (EE) and crude fiber (CF) are presented in Table 1.

The results of the analysis of tannins from jackfruit leaves consisting of measurements of total phenol, total non-tanin phenol, total tannin and condensed tannin are presented in Table 2.

Table 2. Data analysis of jackfruit leaf tannin content

Measurement	Concentration (%)
Total phenol	32.11
Total phenol non tannin	7.56
Total tannin	24.55
Condensed tannin	9.96

The measurement results above show that the condensed tannins from jackfruit leaves are 9.96%, higher than the previous study which reported that the condensed tannin content in jackfruit leaves was 5.57% [11].

The results of in vitro analysis using the Tilley and terry method obtained the results of the degree of acidity (pH) as shown in Table 3.

Table 3. Data on the degree of acidity of protein protection *in vitro*

Treatment	рН
Control	7.06
0.70%	7.11
1.40%	7.15
2.10%	7.08

The pH value in this study was still within the normal range for rumen microbes in carrying out their activities. The normal rumen pH ranged from 5.5 to 7.2 for microbial activity in degrading feed and continued for rumen microbes [12] and 6.5 - 7, with the optimum temperature for the system rumen microbial enzymes between $39 - 41^{0}$ C [13]. The best pH value for protein supplementation is approximately 7 [14].

The data above shows that with the addition of condensed tannins the digestibility decreases, so this study is the addition of tannins will reduce nutrient utilization, so that DMD and ODM with the addition of condensed tannins have lower digestibility compared to controls [15]. The decrease in DMD and ODM is a sign

Treatment	DMD (%)		OMD (%)			
	Rumen	After rumen	Amount	Rumen	After rumen	Amount
Control	70.42b	8.05	78.47b	78.84b	5.62	84.45c
0.70%	61.62a	8.02	69.64a	68.75a	5.77	74.52a
1.40%	66.27ab	6.93	73.20ab	71.25a	6.33	77.58b
2.10%	63.57ab	8.21	71.78ab	70.04a	5.17	75.22ab

Table 4. Dry matter digestibility (DMD) and organic matter digestibility (OMD) of protein protection

Different letter on the same column indicates significantly different (P<0.05)

of protein protection in high-protein feed ingredients by condensed tannins, from the degradation effect of rumen microbes.

Crude protein digestibility (CPD) is shown in Table 5.

Table 5. Crude protein digestibility (CPD) of protein protection

Treatment	DMD (%)			
	Rumen	After rumen	Amount	
Control	66.26a	1.07a	67.34	
0.70%	47.58b	16.68b	65.44	
1.40%	48.19b	22.81b	65.92	
2.10%	46.20b	15.39b	66.77	

Different letter on the same column indicates significantly different (P<0.05)

The addition of condensed tannins for the protection of high protein feed ingredients was found to decrease the digestibility in the rumen. However, post-rumen digestibility increased when compared to the control.

The higher the percentage of addition of condensed tannins (2.1%) the lower crude protein digestibility, this indicates that the protection by tannins is so strong that those degraded in the rumen cannot be released while in the post-rumen, in line with the opinion of Getachew *et al.* [16] which states that tannins can reduce protein digestibility.

4. CONCLUSION

From the results of the above discussion, it can be concluded that the addition of condensed tannins of 0.7% is the optimal number for protein protection from protein degradation in the rumen in vitro.

AUTHORS' CONTRIBUTIONS

WAHIDIN TEGUH SASONGKO performed the experiment, data analysis, wrote and revised the article draft; TEGUH WAHYONO designed and supervised the experiment; FIRSONI checked data analysis and revised the article draft.

ACKNOWLEDGMENTS

The author thanks IRAWAN SUGORO as the coordinator for agriculture, also thanks DEDI ANSORI, YUNIDA MAHARANI and UDIN SIMAN for technical assistance during the research.

REFERENCES

- [1] Sampurna, I Putu. 2013. Kebutuhan Nutrisi Ternak. Fakultas Kedokteran Hewan Universitas Udayana. Bali.
- [2] El-Wazyri, AM., Nasser, M.E.A. and Sallam, S.M.A. 2005. Processing Methodes of Soybean Meal : 1-Effect of Roasting and Tannic Acid Treated-Soybean Meal on Gas Production and Rumen Fermentation In Vitro. Journal of Applied Sciences Reseach 1(3): 313-320.
- [3] Nelson, D.L. and M.M. Cox. 2000. Lehninger Principles of Biochemistry. First Edition. W. H. Freemann and Company New York. New York. The United States.
- [4] Makkar, H.P.S., 1999. Role of Tannins and Saponin in nutrition. In Proceeding of the seventh scientific workshop in Tromso : Effects of antinutritional value of legume diets.
- Broadhurst, R.J. Johns, W.T. 1978. Analysis of Condensed Tannins using Acidified Vanillin. J. Sci. Food Agric. 29: 783-794. https://doi.org/10.1002/jsfa.2740290908.
- [6] Butter. N. L., J. M. Dawson, and P.J. Buttery. 1999. Effects of dietary tannins on ruminants. In: Caygill. J.C and I. Mueller-Harvey. Secondary Plant Products, Antinutritional and beneficial actions in animal feeding. Nottingham University Press. UK.
- [7] Sasongko, W. T., L. M. Yusiati, Z. Bachruddin dan Mugiono. 2010. Optimalisasi pengikatan tanin daun nangka dengan protein bovine serum albumin. Bul. Peternak. 34(3): 154-158.



https://doi.org/10.21059/buletinpeternak.v34i3.8 4

- [8] AOAC (2005). Official Method of Analysis. Maryland:Association of Official Analytical Chemists.
- [9] Makkar, H.P.S. 2003. Quantification of Tannins in Three and Shrub Foliage : A Laboratory Manual. Kluwer Academic Publ. The Netherland. https://doi.org/10.1007/978-94-017-0273-7
- [10] Tilley J M A & Terry R A. A two-stage technique for the in vitro digestion of forage crops. J. Brit. Grassland Soc. 18:104-11, 1963. https://doi.org/10.1111/j.1365-2494.1963.tb00335.x
- [11] Sasongko, W.T. 2010. Pemanfaatan tanin daun nangka untuk meningkatkan nilai rumen undegraded protein pada bahan pakan protein tinggi. Tesis. Fakultas Peternakan, Universitas Gadjah Mada, Yogyakarta.
- [12] Owens, F. N and A. L. Goetsch. 1988. Ruminal Fermentation. In : D.C. Church (Ed), The Ruminant Animal Digestive Physiology and Nutrition. Reston Book Prentice Hall, Englewood Cliffs, New Jersey.
- [13] Czerkawski, J. W., 1986. An Introduction to Rumen Studies. Pergamon Press. Oxford. https://doi.org/10.1016/B978-0-08-025486-9.50007-4
- [14] Owens, F. N and R. Zinn. 1988. Protein metabolism of Ruminant animals. In : D.C. Church (Ed), The Ruminant Animal Digestive Physiology and Nutrition. Reston Book Prentice Hall, Englewood Cliffs, New Jersey.
- [15] Makkar, H.P.S. 2003. Quantification of Tannins in Ttree and Shrubfoliage : A Laboratory Manual. Kluwer Academic Publ. The Netherland. https://doi.org/10.1007/978-94-017-0273-7
- [16] Getachew, G., W. Pittroff., D.H. Putnam., A. Dandekar., S. Goyal, and E.J. DePeters. 2008. The influence of addition of gallic acid, tannic acid, or quebracho tannins to alfalfa hay on in vitro rumen fermentation and microbial protein synthesis. Anim. Feed Sci. Technol. 140: 444-461.

https://doi.org/10.1016/j.anifeedsci.2007.03.011