

Phosphorus Supplementation of Ammoniated Rice Straw on Rumen Fermentability, Synthesised Microbial Protein and Degradability *in Vitro*

Mardiati Zain, N. Jamarun and A. S. Tjakradidjaja

Abstract—The effect of phosphorus supplementation of ammoniated rice straw was studied. The *in vitro* experiment was carried out following the first stage of Tilley and Terry method. The treatments consisting of four diets were A = 50% ammoniated rice straw + 50% concentrate (control), B = A + 0.2% Phosphor (P) supplement, C = A + 0.4% Phosphor (P) supplement, and D = A + 0.6% Phosphor (P) supplement of dry matter. Completely randomized design was used as the experimental design with differences among treatment means were examined using Duncan multiple range test. Variables measured were total bacterial and cellulolytic bacterial population, cellulolytic enzyme activity, ammonia (NH₃) and volatile fatty acid (VFA) concentrations, as fermentability indicators and synthesized microbial protein, as well as degradability indicators including dry matter (DM), organic matter (OM), neutral detergent fibre (NDF), acid detergent fibre (ADF) and cellulose. The results indicated that fermentability and degradability of diets consisting ammoniated rice straw with P supplementation were significantly higher than the control diet (P < 0.05). It is concluded that P supplementation is important to improve fermentability and degradability of rations containing ammoniated RS and concentrate. In terms of the most effective level of P supplementation occurred at a supplementation rate of 0.4% of dry matter.

Keywords—ammoniated rice straw, phosphorus, fermentability, degradability and synthesized microbial protein.

I. INTRODUCTION

IMPROVE fiber use following treatment with alkalis [1] suggest that scope should exist to derive more nutrients from fiber by microbial fermentation in the rumen. However potentially degradable fibre may be transported from the rumen before fermentation is complete. The extent of fermentation in the available time depends on the biomass of cellulolytic bacteria. Nutritive value of some straws and other by-product feeds can be improved simply by adding urea and

minerals as such as P. These bacteria are needed for pregastric digestion of fibre in cattle. Replication and growth of the bacteria are dependent upon a P supply [2]. P supplementation is important for rumen fermentation and growth of rumen microbia, therefore, a study was conducted to examine the effects of P addition on *in vitro* fermentability, synthesised microbial protein and degradability of ammoniated rice straw.

II. MATERIAL AND METHODS

The experimental diet composed of 50% ammoniated rice and 50% concentrate, and this diet was used as a control diet (A). The rice straw was previously treated with 4% urea. The crude protein of the diet was 10.16%. P₂O₅ was used as a P source and added to the diet 0.2, 0.4 and 0.6% on dry matter, respectively. *In vitro* fermentability and degradability of nutrients were determined following the first stage of the Tilley and Terry procedure [3]. Ruminal fluid was obtained from a cannulated steer. Fermentation tubes contained of 10 ml of ruminal fluid and 40 ml of McDougall buffer solution. Samples were incubated in duplicate in 100 ml polyethylene tubes in 39 °C in a shaken water bath for 48 h. Two fermentation tubes that did not contain diets were also incubated and used as blanks. Sample was taken from each fermentor for bacterial counting. Fermentation was terminated at 48 h by injecting the tubes with 1 ml of HgCl₂. Tubes were then centrifuged at 2000 x g for 15 min and the supernatant was removed. Tubes with residue were dried at 60 °C for 48 h and weighed and the data were used for degradability determination. These residues were also analyzed for its DM, OM and N by using standard procedures (4), the NDF, ADF, and cellulose of residues were determined by [5] procedures. Supernatants were used in order to determine NH₃ concentration (microdiffusion Conway method), total VFA concentration (Gas chromatography) and rumen fluid pH. Total and cellulolytic bacterial population was determined by methods described in [6], cellulase enzyme activity and the amount of synthesized microbial protein was, determined by methods described in [7, 8] respectively. A completely randomized design was used as experimental design consisting of four treatments. Data were analyzed by ANOVA using the GLM procedure. Differences between the control treatment and P supplementation treatment were analyzed by Duncan multiple range test (DMRT) [9].

Mardiati Zain, . Author is with the *Department of Animal Nutrition, Faculty of Animal Science, Andalas University, Kampus Limau Manis, Padang - 25163, Indonesia, Telp/HP: 0751-72400/08126703875 E-mail mardiat@faterna.unand.ac.id*

N. Jamarun. Author, Author is with the *Department of Animal Nutrition, Faculty of Animal Science, Andalas University, Kampus Limau Manis, Padang - 25163, Indonesia, Telp/HP: 0751-72400/08126608179 E-mail novirmanj@yahoo.co.id*

A.S. Tjakradidjaja Author is with the *Department of Animal Nutrition and Feed Technology, Faculty of Animal Science, Bogor Agricultural University, Kampus Dramaga, Bogor - 16680, Indonesia, Email : atj.yanuar@gmail.co.id*

III. RESULT AND DISCUSSION

Table shows results of P supplementation effects on bacterial population and other variables of rumen fermentability. Effects of treatments were significant ($P < 0.05$) for ammonia and total VFA concentrations. Data on *in vitro* degradability of ammoniated RS are presented in the table and show that the addition of P at different level affected all degradability variables ($P < 0.05$).

TABLE I

EFFECT OF PHOSPHORUS SUPPLEMENTATION ON TOTAL AND CELLULOLYTIC BACTERIAL POPULATION AND FERMENTATION IN THE RUMEN AND *IN VITRO* DEGRADABILITY

Variabel	P Supplementation (%DM)			
	0	0.2	0.4	0.6
Synthesised microbial protein (%/g)	0.19	0.18	0.21	0.13
N-NH ₃ (mM)	11.09 ^a	10.02 ^b	9.25 ^c	8.80 ^d
Total VFA (mM)	88.75 ^c	98.12 ^b	106.87 ^a	111.87 ^a
Dry matter degradability (%)	52.91 ^c	54.85 ^{bc}	57.66 ^a	60.79 ^a
Organic matter degradability (%)	54.69 ^c	58.43 ^b	60.18 ^a	62.69 ^a
NDF degradability (%)	39.31 ^b	41.58 ^b	43.94 ^a	50.91 ^a
ADF degradability (%)	27.99 ^c	32.78 ^{bc}	37.59 ^a	40.30 ^a
Cellulose degradability (%)	29.47 ^b	33.04 ^b	38.74 ^a	41.61 ^a

Means within rows with the same superscript letter are significantly different at $P < 0.05$

Ammonia concentrations decreased from 11.09 mM in control diet to 10.02, 9.25 and 8.80, respectively in rations B, C and D which consisted of S supplements at 0.2, 0.4 and 0.6% on dry matter. Differences in ammonia concentration among treatment diets were significant ($P < 0.05$). Reverse results from ammonia concentrations were obtained in total VFA concentrations. The addition of P supplement increased total VFA concentrations from 88.75 mM in diet A to 98.12 in diet B, 106.87 mM in diet C and 111.87 mM in diet D. Total VFA concentrations differed significantly between all rations ($P < 0.05$), except that between rations C and D.

The present results demonstrate that addition of P supplement at diet had reduced ammonia concentration, and increased total bacterial population. Although the treatments did not produce significant effect on cellulolytic bacterial population, S supplementations had increased cellulolytic enzyme activity which subsequently increased total VFA concentration. This means that cellulolytic rumen bacteria were present in small population in liquid phase used as samples for counting cellulolytic bacterial numbers; most of cellulolytic bacteria may be attached to fibrous components in solid phase samples. The results also indicate that P plays important role in rumen microbial growth and cellulolytic enzyme activity which then increased fibre degradation and total VFA concentration. These results had agreed with the works reported by [10].

Comparisons among diets supplemented with P, the best results in fermentability study had been obtained at 0.4% P supplement. This ratio was the same that was obtained by [2] who suggested that the best P supplement to beef cattle was 0.4-0.5% On dry matter on diet low quality roughage.

in vitro degradability

Data on *in vitro* degradability of ammoniated RS are presented in the Table 2. The addition of P at different level on dry matter affected all degradability variables ($P < 0.05$). Control diet (A) had the lowest DM, OM, NDF, ADF and cellulose degradabilities ($P < 0.05$). An increase in P supplementation from 0.2% (B) 0.4% (C) and 0.6% (D) increased the degradabilities of DM, OM, NDF, ADF and cellulose, and the increase in degradabilities of DM, OM, and fibrous fractions followed linier patterns with the levels of S supplementation.

The present results in degradabilities of DM, OM and fibrous fraction of diets consisting of ammoniated RS and concentrate were in association with the increase in total VFA concentration as affected by P supplementation. These indicate that P supplementation had promoted rumen bacterial growth and cellulolytic enzyme activities which increased fermentability and degradability of diets consisting ammoniated RS and concentrate. This study has also shown that diets consisting of ammoniated RS were deficient in P, and P supplementation is important to improve fibre degradation of fibrous feedstuffs. The present results were in agreements with the results of (11:12). The last researchers had indicated that improvement in fibre degradation by P supplementation occurred through its specific stimulation on growth of rumen cellulolytic bacteria and anaerobic rumen fungi. The present study has suggested that the best result was obtained by supplementing P at 0.4% on dry matter.

IV. CONCLUSION

Fermentability and degradability of rations containing ammoniated RS and concentrate. The effects occurred through reduction in ammonia concentration, increase in total bacterial population, cellulolytic enzyme activity, total VFA concentration, and degradabilities of DM, OM, and fibrous fraction. The best level of P supplementation is obtained at a 0.4% on dry matter.

ACKNOWLEDGMENT

This work was supported by an Insentif Riset Dasar Programme at Ministry of Research and Technology, Republic of Indonesia in 2007.

REFERENCES

- [1] Van Soest, P.J. 2006. Rice straw the role of silica and treatment to improve quality. *J.Anim.Feed. Sci. and Technology* Volume 130, pp. 137-171.
- [2] Patterson, J. 2002.. *Proc. 37th Pac NW Anim Nutr Conf.* pp. 11-19.
- [3] Tilley, J. M., and R. A. Terry. 1969. A two stage technique for *in vitro* digestion of forage crops. *Journal of British Grassland Society* 18 (2), pp 104 – 111
- [4] AOAC. 1990. Official methods of analysis. 15th edn. Association of Official Analytical Chemists, Arlington, Virginia.
- [5] Goering, H.K. and Van Soest, P.J. 1970. Forage fiber analysis(apparatus, reagents, procedure and some applications. In: United States Department of Agricultural (Ed), *Agricultural Handbook*, 379. US government printing Office, Washington D.C., pp 1-20
- [6] Suryahadi. 1990. *Penuntun praktikum Ilmu Nutrisi Ruminansia*. Pusat Antar Universitas Ilmu Hayat. IPB. Bogor

- [7] Widyastuti, A.T. 2005. Isolasi dan uji kemampuan bakteri simbiosis rayap pendegradasi serat. Skripsi. Fakultas Peternakan. Institut Pertanian Bogor.
- [8] Gopar, A.H., 1981. Pengaruh perubahan laju penggantian isi rumen terhadap kegiatan metabolisme mikrobanya dalam kondisi *in vitro*. Skripsi. Fakultas Peternakan. Institut Pertanian Bogor.
- [9] Steel, R.G.D. and J.H. Torrie. 1980. Principles and Procedure of Statistics. McGraw-Hill Book Co. Inc. New York.
- [10] Komizarczuk, S., and Durand M. 1991. Effect of mineral on microbial metabolism. In: Rumen Microbial Metabolism and Ruminant Digestion. J.P. Jouany (Ed) INRA publ. Versailles, France
- [11] Bravo, D., D. Sanvant, C. Bogaert and F. Meschy. 2003. Quantitative aspect of phosphorus absorption in ruminant. *Reprod. Nutr. Dev.* 43: 271-284. INRA. EDP. Sciences.
- [12] Durand, M. and R. Kawashima. 1980. Influence of minerals in rumen microbial digestion. In: Ruckebush and Thivend (eds): *Digestive Physiology and Metabolism in Ruminants*. MTP Press, Ltd., Lancaster, England. Pp 375-408.
- [13] Rodehutsord, M. Heuvers, H. Pfeffer, 2000. Effect of organic matter digestibility on obligatory faecal phosphorus loss in lactating goats, determined from balance data. *Anim. Sci.* 70: 561-568.