

Feasibility of *Porphyra haitanensis* as protein source for ruminants

J.K. Wang¹, H.L. Mao¹, Y.W. Zhou¹, J.X. Liu^{1,3} and X.J. Yan²

¹College of Animal Sciences, Institute of Dairy Science, Ministry of Education
Key Laboratory of Molecular Animal Nutrition, Zhejiang University
Hangzhou 310029, P.R. China

²Ningbo University, Ningbo 315211, P.R. China

ABSTRACT

The present study was conducted to investigate the feasibility of *Porphyra haitanensis* (PH) as protein source for ruminants using *in vitro* gas test. The PH was inferior to soyabean meal in terms of gas production (GP) and rumen fermentation. When the soyabean meal in the mixed substrates was replaced by PH at levels of 20-100%, GP and volatile fatty acids decreased, while microbial protein yield was not reduced. Inclusion of PH at a level of 30% did not have adverse effect on rumen parameters. It is suggested that PH could be used as protein source to replace soyabean meal.

KEY WORDS: *Porphyra haitanensis*, rumen fermentation, *in vitro*

INTRODUCTION

Porphyra haitanensis (PH), a traditional edible red alga, occupies 80% of the cultivated algae in China. Growing period of PH is about eight months normally, from September to the second year. However, as food for human being, the virtual period is one to two months, and less than four months even for the best variety (Wang, 2005). A large amount of PH produced is treated as waste every year. Moreover, PH cannot survive in hot summer, dried-up PH will cumulate during this season. On the other hand, shortage of feed grain is one of the main constraints for development of livestock production in China. It has been forecasted that the deficit of protein feedstuff will be 48 million tonnes in 2020 (Yuan and Zhang, 2004). Total amino acids content of dry PH is above 30% (Chen, 1999), and utilization of the wasted PH as protein source for livestock may be beneficial to both PH production and livestock industry.

³ Corresponding author: e-mail: liujx@zju.edu.cn

The objective of the present study was to evaluate the feasibility of PH used as protein source to replaced soyabean meal for ruminants.

MATERIAL AND METHODS

Porphyra haitanensis and soyabean meal

The PH at early stage was cultivated in Zhejiang Province. The crude protein content of this PH was 33% in the DM, and lower than that of soyabean meal (47%). Ash content of PH was higher than that of soyabean meal, 12 vs 6% of the DM (Chinese Feed Database, 2003).

Experimental design

In Trial 1, soyabean meal in the mixed substrate (%: Chinese wild rye 60, maize meal 25 and soyabean meal 15, w/w) was replaced by PH at levels of 0, 20, 40, 60, 80 and 100%, respectively. Calibrated glass syringes (Häberle Labortechnik, Germany) were used as incubators. Six syringes were incubated at the same times. Three samples were used for gas test and others used to determine ammonia-N, volatile fatty acids (VFAs) and microbial protein (MCP) after 24 h incubation. The gas production (GP) was also determined for soyabean meal and PH In Trial 2, PH was included in low protein substrate (60% rice straw and 40% maize meal) and was exchanged for the mixed substrate (both rice straw and maize meal at 60:40) included at 15 and 30% to evaluate the feasibility of high level of PH in ruminant diet.

In vitro gas production studies

The GP was determined according to Menke and Steingass (1988). Rumen fluid used for determination of GP was collected from two rumen-fistulated sheep fed on a diet (60% Chinese wild rye plus 40% concentrate mixture) at 1.3 times maintenance. The GP value recorded at 2, 4, 8, 12, 24, 36, 48 and 72 h of incubation, were fitted to the equation: $GP = a + b(1 - e^{-ct})$, where: a the intercept of the gas production curve, $a + b$ - the potential gas production, c - the rate of gas production. Measurement of the *in vitro* fermentation parameters, ammonia, VFAs and MCP were also performed.

Measurement of in vitro fermentation parameters

Concentrations of the ammonia-N were determined with colorimetry. Gas chromatography was used to determine VFAs, and the details are as described by Hu et al. (2005). Concentrations of MCP were determined based on purine.

Statistical analysis

The effects of PH on gas production and fermentation parameters were analysed according to one-way analysis by GLM procedure of SAS, and the difference of means was tested by using Duncan's new multiple range test. The linear and quadratic effects of increasing level of PH were determined using polynomial contrasts (Steel and Torrie, 1980).

RESULTS

The cumulative GP curves are presented in Figure 1 for PH, soyabean meal, and all the mixed substrates. The GP of PH was much lower than that of soyabean meal, 19.2 vs 47.8 ml/200 mg DM after 72 h incubation. The GP values of the mixed substrates decreased with increasing levels of PH to soyabean meal.

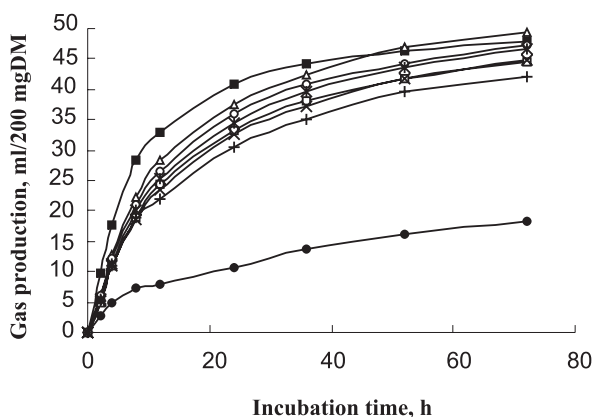


Figure 1. Gas production curves for soyabean meal (SBM, ■), *P. haitanensis* (PH, ●), and the mixed ration in which SBM was replaced by PH at levels of 0 (Δ), 20 (○), 40 (□), 60 (*), 80 (+), and 100% (×), respectively

The potential GP, total VFA, and ammonia-N of PH were lower than that of soyabean meal, but no difference in MCP was observed (Table 1). With the increasing level of PH to replace soyabean meal ($P < 0.05$), potential GP, total VFA, and ammonia-N of the mixed substrates decreased linearly ($P < 0.05$), without difference in ammonia-N concentration between the different treatments ($P > 0.05$). Moreover, there were no significant differences between MCP in the mixed substrates ($P > 0.05$).

Table 1. Gas production and fermentation parameters of soyabean meal (SBM), *P. haitanensis* (PH), and the mixed ration in which SBM was replaced by PH at different levels

Experimental substrates	PH	SBM	Mixed ration						SEM	Effect ¹	
			levels of PH replacing SBM, %							L	Q
			0	20	40	60	80	100			
<i>Gas production parameters</i> ²											
a + b, ml	19.2	46.1	48.0	45.9	43.4	45.4	41.1	43.9	0.86	*	NS
c, ml/h	0.7	5.0	3.3	3.2	2.8	2.9	2.5	2.6	0.09	*	NS
<i>Fermentation parameters after 24 h incubation</i> ³											
total VFA, mmol/l	32.7	42.7	38.0	37.4	36.8	36.7	36.1	35.5	0.35	*	NS
acetate, %	74.3	72.3	73.6	73.5	74.1	73.7	74.0	73.3	0.29	NS	NS
propionate, %	21.8	23.9	22.4	22.6	22.0	22.4	22.1	22.7	0.30	NS	NS
butyrate, %	3.9	3.8	4.0	3.9	3.9	3.9	3.8	4.0	0.09	NS	NS
A/P	3.4	3.0	3.3	3.3	3.4	3.3	3.3	3.2	0.06	NS	NS
Ammonia-N, mg/100 ml	38.5	56.8	26.5	27.8	25.9	26.2	25.3	24.9	0.64	*	NS
MCP, mg/ml	2.2	2.2	2.5	1.9	2.0	2.2	2.0	1.8	0.18	NS	NS

¹L - linear, Q - quadratic, *P<0.05, NS - not significant; ²a + b - potential gas production, c - rate of gas production; ³VFA - volatile fatty acid, A/P - ratio of acetate to propionate, MCP - microbial protein

When the level of PH in the mixed substrate was increased to 15 and 30% in a low-protein diet, the GP, total VFA, and butyrate proportion decreased, while acetate, ammonia-N and MCP yield increased (Table 2). Though, the GP and total VFA was decreased in the substrate with 30% of PH, there were no differences between 15 and 30% substrates (P>0.05).

DISCUSSION

Though gas production is not a good method for evaluation of protein source, the digestibility of the exchanged diet could be evaluated by GP, and the protein value by MCP and ammonia-N in the end-products of fermentation. The GP and fermentation parameters of PH were significant lower than that of soyabean meal (P<0.05), there was little difference in ammonia-N concentration between the 0% and other levels. There was also no significant difference in MCP between the mixed substrates with different levels of PH (P>0.05). These results revealed that replacement of soyabean meal with PH as protein source for ruminant could not affect the nitrogen utilization of soyabean meal, but the energy in the diet should be considered. Similar results could be revealed in Table 2. Ventura (1994) reported that *Ulva* (a species of algae) was not a suitable ingredient for poultry diets because of its low metabolizable energy content. With a digestible energy content of 9.1 MJ/kg DM, the inclusion of 20% *Ulva lactuca* in ruminant diets was not refused (Arieli et al., 1993).

Table 2. Gas production and fermentation parameters of mixed substrates (60% rice straw and 40% maize meal) in which *P. haitanensis* (PH) occupied different levels

Experimental substrates	Levels of PH in the substrate, %			SEM
	0	15	30	
<i>Gas production parameters</i> ¹				
GP ₂₄ , ml	39.2 ^a	35.6 ^b	35.0 ^b	0.37
<i>Fermentation parameters after 24 h incubation</i> ²				
total VFA, mmol/l	42.7 ^a	41.3 ^{ab}	39.6 ^b	0.54
acetate, %	73.7 ^b	74.2 ^{ab}	74.5 ^a	0.23
propionate, %	15.7	15.5	15.5	0.16
butyrate, %	10.7 ^a	10.3 ^{ab}	10.0 ^b	0.17
A/P	4.7	4.8	4.8	0.06
Ammonia-N, mg/100 ml	32.8 ^b	35.0 ^{ab}	37.0 ^a	1.00
MCP, mg/ml	1.4 ^b	1.6 ^{ab}	1.8 ^a	0.10

¹ GP₂₄ - gas production at 24 h incubation; ² VFA - volatile fatty acid, A/P - ratio of acetate to propionate, MCP - microbial protein; ^{a,b,c} means within the same row with different superscripts differ (P<0.05)

Total amino acids content of dry PH cultivated in Fujian Province was 35.5% in the DM, the contents of Ca, Fe, Zn and Mg were 3.63, 0.56, 0.095 and 3.29 mg/g, respectively (Chen et al., 2001), comparable to the content in soyabean meal (Chinese Feed Database, 2003). However, the effect of higher amounts of minerals from PH on ruminal fermentation could not be detected in the *in vitro* incubation system with abundant minerals. In addition to abundant minerals, bioactive matters existed in PH also. Porphyrin, one of the main components of *Porphyra*, has been shown to stimulate immune response (Yashizawa et al., 1995), scavenge active oxygen radicals *in vitro* (Kuda et al., 2005). However, the direct effects of *Porphyra* on ruminants had not been detected.

CONCLUSIONS

Porphyra haitanensis could be used in ruminant diets, but not as the only protein source to replace soyabean meal. Not only the protein supply but also energy needs to be considered when *P. haitanensis* is used as a supplement. Further research is required to evaluate the feeding value of *P. haitanensis*.

REFERENCES

- Arieli A., Sklan D., Kissil G., 1993. A note on the nutritive value of *Ulva lactuca* for ruminants. Anim. Prod. 57, 329-331
- Chen B.L., Lin Y.X., Huang J., 2001. Nutritional evaluation of *Porphyra haitanensis*. Chin. Oceanol. Medi. 80, 50-53

- Chen R., 1999. Analyses on nutritive composition of *Porphyra haitanensis*. J. Ocenogr. Taiwan Strait 18, 465-468
- Chinese Feed Database, 2003. China Feed 2003. 21, 22-27 22, 26-33
- Hu W.L., Liu J.X., Ye J.A., Wu Y.M., Guo Y.Q., 2005. Effect of tea saponin on rumen fermentation in vitro. Anim. Feed Sci. Tech. 120, 333-339
- Kuda T., Tsunekawa M., Goto H., Araki Y., 2005. Antioxidant properties of four edible algae harvested in the Noto Peninsula, Japan. J. Food Compos. Anal. 18, 625-633
- Menke K.H., Steingass H., 1988. Estimation of the energetic feed value obtained from chemical analysis and in vitro gas production using rumen fluid. Anim. Res. Dev. 28, 7-55
- Steel R.D.G., Torrie J.H., 1980. Principles and Procedures of Statistics. 2nd Edition. McGraw-Hill Book Company. New York, pp. 137-194, 363-372
- Ventura M.R., 1994. Nutritional value of seaweed (*Ulva rigida*) for poultry. Anim. Feed Sci. Tech. 49, 87-94
- Wang Q.X., 2005. Thinking on development of *Porphyra haitanensis* industry in Fujian province. J. Fujian Fisheries 6, 71-73
- Yashizawa Y., Ametani A., Tsunehiro J., Numura K., Itoh M., Fukui F., Kaminogawa S., 1995. Macrophage stimulation activity of the polysaccharide fraction from a marine alga (*Porphyra yezoensis*): structure-function relationships and improved solubility. Biosci. Biotechnol. Biochem. 59, 1933-1937
- Yuan T., Zhang W.L., 2004. The present situation of protein feedstuff in China. Jiangxi Feed 2, 25-48