*Journal of Animal and Feed Sciences, 16, Suppl. 2, 2007, 604–608*

# Effect of *Picrasma quassioides* plant extract, yeast culture and monensin on *in vitro* mixed ruminal microorganism fermentation of wheat starch*

**J.A. Wei, H.S. Xin, Q.X. Meng, L.P. Ren and Z.M. Zhou[1]**

*College of Animal Science and Technology, Beef Cattle Research Center,*
*China Agricultural University*
*Beijing 100094, P.R. China*

ABSTRACT

The objective of this study was to compare the plant extract from *Picrasma quassioides* (EPQ), yeast culture and monensin on *in vitro* rumen fermentation characteristics of wheat starch. Compared with the control (no additive), EPQ decreased the dry matter digestibility and the total VFA production ($P<0.05$), and increased the ratio of acetate and propionate. Both EPQ and yeast culture increased the final rumen pH ($P<0.05$), but monensin had a negative effect on decline of rumen pH. In conclusion, the addition of EPQ can efficiently retard rumen fermentation rate of wheat starch *in vitro*, and increase the final pH of ruminal fluid.

KEY WORDS: *Picrasma quassioides* extract, yeast culture, monensin, fermentation *in vitro*, rumen acidosis

## INTRODUCTION

Diets rich in readily fermented carbohydrate will induce acute and chronic acidosis of ruminant. Since the Food and Drug Administration approved the feeding of ionophore-antibiotics to animals in the mid-1970s, these compounds have been used to prevent rumen acidosis and improve production performance of beef cattle. Monensin is the most widely used ionophore (Russell and Strobel, 1989). It can decrease $CH_4$ production (Thornton and Owens, 1981) and increase ruminal pH by decreasing volatile fatty acid and lactate production (Bergen and Bates, 1984). Yeast culture has been supplemented in ruminant diet for many

* Supported by National Natural Science Foundation of China, Grant No. 30125033 and 30270944, and 948 Grant of Ministry of Agriculture of China, Grant No. 2003 - Z77
[1] Corresponding author: e-mail: zhouzhenming2001@sohu.com

years. Researchers have reported that, in some cases, yeast culture could influence the ruminal fermentation *in vitro* and then stimulated lactate uptake (Harris and Webb, 1990). Unfortunately, there were not consistent results about the effect of yeast culture in *in vivo* and *in vitro* fermentation (Martin and Nisbet, 1992). EPQ is used as herbal antimicrobial to prevent inflammation in human medicine, which shows that the EPQ may effect on the bacteria. So the objective of this study is to compare the effects of the EPQ, yeast culture and monensin on ruminal fermentation *in vitro* of readily fermented carbohydrate (wheat starch).

## MATERIAL AND METHODS

Wheat starch was used as a substrate for the rumen fermentation *in vitro*. EPQ purchased from Shengzhitang Company (Guangdong, China) was added to form a final concentration of 100 mg/l mixed culture fluid. Yeast culture product, purchased from Piotech Company (USA), was added at the concentration of 200 mg/l. Monensin was purchased from Haizheng Company (China) and added at the concentration of 3 mg/l mixed culture fluid.

*In vitro* rumen fermentation was carried out with rumen fluid which was obtained from four rumen cannulated steers fed concentrate supplement twice a day. Ruminal contents were obtained in the morning, mixed and squeezed through four layers of cheesecloth into a 1.000 ml flask with an $O_2$-free $CO_2$ headspace and maintained in a 39°C water bath. Particle-free fluid from the flask was anaerobically mixed (1:4) with a buffer (pH 6.5) containing the following compounds, mg/l: $K_2HPO_4$ 292, $KH_2PO_4$ 240, $(NH_4)_2SO_4$ 480, NaCl 480, $MgSO_4 \cdot 7H_2O$ 100, $CaCl_2 \cdot 2H_2O$ 64, $Na_2CO_3$ 4000, and cysteine hydrochloride 600 (Lynch and Martin, 2002). After mixing, 30 ml buffered rumen fluid was dispensed in to 100 ml syringe containing 300 mg of wheat starch and different additives described previously. The glass syringes (HFT000025, Häberle Maschinenfabrik GmbH, Germany) were sealed by clips and incubated at 39°C for either 24 h (for 24 h fermentation traits) or 72 h (for 72 h gas production). Each treatment had three syringes as replicates. The next three syringes were served as blank incubation (rumen fluid + buffer). After 24 (or 72) h, fermentation was stopped and the syringes were put into an ice-cooled bath.

After cooling, all the contents of the syringes were centrifuged (5400 *g*, 4°C, 15 min) for dry matter digestibility (DMD) determination. The concentrations of VFA were determined by the gas chromatography procedure (Beauchemin et al., 2003) with the Agilent G 6890 Gas Chromatography. $NH_3$-N concentration was measured according to the method of Broderick and Kang (1980), lactate concentration by ion chromatography procedure (Dionex ICS 2500).

The data of 72 h dynamic gas production was estimated using the non linear regression procedure of SAS (1996) the model is GP=B*[1-exp (-c*(t-lag))].

The differences in pH, total and individual volatile fatty acids, lactate, $NH_3$-N production as well as dry matter digestibility (DMD) and other parameters were analysed by analysis of variance using GLM procedure of SAS (1996).

## RESULTS AND DISCUSSION

The dynamic change of cumulative gas production of different additives was shown in Figure 1. EPQ significantly decreased ($P<0.05$) the rate of the gas production and increased ($P<0.05$) the lag time (Table 1). This might be due to the EPQ could depress the activity of ruminal microorganisms and influenced the fermentation, which resulting in the decreased rate of gas production. Monensin significantly decreased ($P<0.05$) the amount of gas production and the yeast culture decreased ($P<0.05$) the lag time of the fermentation.

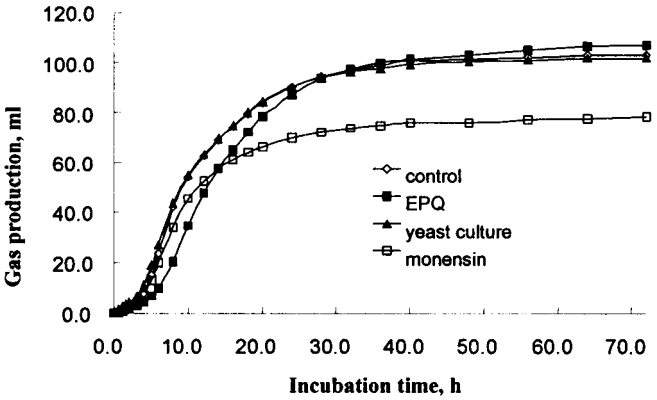


Figure 1. The dynamic change of cumulative gas production of wheat starch added different additives

Table 1. The effect of different additives on dynamic traits of *in vitro* gas production of wheat starch

| Parameter | Treatment | | | | SEM | P |
|---|---|---|---|---|---|---|
| | control | EPQ | yeast culture | monensin | | |
| 24 h gas production, ml | 90.4[a] | 87.1[a] | 90.3[a] | 69.7[b] | 1.45 | <0.001 |
| 72 h gas production, ml | 102.9[a] | 107.0[a] | 101.8[a] | 78.2[b] | 1.32 | <0.001 |
| Maximum gas production, ml | 106.5[b] | 114.7[a] | 104.7[b] | 79.6[c] | 1.43 | <0.001 |
| Rate of gas production, ml/h | 0.075[c] | 0.054[d] | 0.078[b] | 0.086[a] | 0.001 | <0.001 |
| Lag time, h | 1.34[b] | 1.80[a] | 1.09[c] | 1.27[b] | 0.04 | <0.001 |

means with different superscript letter within the same line differ significantly ($P<0.05$

The effect of different additives on ruminal fermentation characteristics *in vitro* was shown in Table 2. Compared with the control, EPQ and yeast culture increased

the final pH, and EPQ had the highest value. Some studies showed that yeast culture increased the pH during the ruminal fermentation *in vitro*, but there were not consistent results between *in vivo* and *in vitro* (Martin and Nisbet, 1992). EPQ decreased ($P<0.05$) the DMD after 24 h fermentation. This result showed that the plant extract of *Picrasma quassioides* decreased the activity of the ruminal microbes and then decreased the feed digestibility. The concentration of $NH_3$-N was increased ($P<0.05$) by addition of EPQ and monensin, the greater value corresponding to the EPQ.

Table 2. The effect of different additives on rumen fermentation characteristics *in vitro*

| Parameter | Treatment | | | | SEM | P |
|---|---|---|---|---|---|---|
| | control | EPQ | yeast culture | monensin | | |
| pH | 5.61$^{c}$ | 5.94$^{a}$ | 5.74$^{b}$ | 5.54$^{d}$ | 0.02 | <0.001 |
| DMD, % | 57.87$^{a}$ | 49.47$^{b}$ | 54.67$^{a}$ | 56.70$^{a}$ | 1.09 | 0.003 |
| $NH_3$-N, mg/l | 22.37$^{c}$ | 36.89$^{a}$ | 25.83$^{bc}$ | 30.62$^{b}$ | 1.84 | 0.003 |
| Lactate, mM | 0.24$^{b}$ | 0.09$^{b}$ | 0.10$^{b}$ | 0.88$^{a}$ | 0.06 | <0.001 |
| Total VFA, mM, % | 68.77$^{a}$ | 56.33$^{b}$ | 65.38$^{a}$ | 64.62$^{a}$ | 1.83 | 0.008 |
| acetate | 51.34$^{b}$ | 54.09$^{a}$ | 51.17$^{b}$ | 48.44$^{c}$ | 0.27 | <0.001 |
| propionate | 35.38$^{b}$ | 27.05$^{c}$ | 35.46$^{b}$ | 47.54$^{a}$ | 0.21 | <0.001 |
| isobutyrate | 0.42$^{b}$ | 0.65$^{a}$ | 0.40$^{b}$ | 0.29$^{b}$ | 0.04 | 0.003 |
| butyrate | 12.15$^{b}$ | 17.22$^{a}$ | 12.21$^{b}$ | 3.28$^{c}$ | 0.27 | <0.001 |
| isovalerate | 0.46$^{b}$ | 0.68$^{a}$ | 0.45$^{b}$ | 0.30$^{c}$ | 0.01 | <0.001 |
| valerate | 0.26$^{b}$ | 0.31$^{a}$ | 0.31$^{a}$ | 0.14$^{c}$ | 0.01 | <0.001 |
| A : P value | 1.45$^{b}$ | 2.00$^{a}$ | 1.44$^{b}$ | 1.02$^{c}$ | 0.02 | <0.001 |

means with different superscript letter within the same line differ significantly ($P<0.05$)

Monensin decreased the molar ratios of acetate to propionate ($P<0.05$). It was in agreement with the result described by Russell and Strobel (1989). Yeast culture almost had no effect on the production of VFA. However, EPQ could significantly decrease the total VFA production ($P<0.05$), and increased the molar ratio of acetate and propionate. Because little lactate (<1 mmol/l) was detected in the present study, the increased final pH of the EPQ treatment might be due to the decreased total VFA production.

## CONCLUSIONS

Addition of extract from *Picrasma quassioides* (EPQ) decreased ruminal fermentation rate and 24 h DMD *in vitro*. This plant extract showed a function in prevention of the rumen pH decline, suggesting a potential use as a rumen fermentation modulator. A deceased 24 h *in vitro* DM digestibility and less production of total VFA due to addition of the plant extract from EPQ indicated an alteration of rumen digestion site or extent resulting from such EPQ addition. EPQ

also changed the molar proportion of individual VFA, suggesting an alteration occurring in the ruminal fermentation pattern. Further studies are needed to investigate the response of animal performance to the addition of this plant extract and its responding mechanisms.

## REFERENCES

Beauchemin K.A., Yang W.Z., Morgavi D.P., Ghorbani G.R., Kautz W., Leedle J.A.Z., 2003. Effects of bacterial direct-fed microbials and yeast on site and extent of digestion, blood chemistry, and subclinical ruminal acidosis in feedlot cattle. J. Anim. Sci. 81, 1628-1640

Bergen W.G., Bates D.B., 1984. Ionophores: Their effect on production efficiency and mode of action. J. Anim. Sci. 58, 1465-1483

Broderick G.A., Kang J.H., 1980. Automated simultaneous determination of ammonia and total amino acids in ruminal fluid and in vitro media. J. Dairy Sci. 63, 64-75

Harris B., Webb D.W., 1990. The effect of feeding a concentrate yeast culture to lactating dairy cows. J. Dairy Sci. 73, Suppl.1, 266 (Abstr.)

Lynch H.A., Martin S.A., 2002. Effects of *Saccharomyces cerevisiae* culture and *Saccharomyces cerevisiae* live cells on in vitro mixed ruminal microorganism fermentation. J. Dairy Sci. 85, 2603-2608

Martin S.A., Nisbet D.J., 1992. Effect of direct-fed microbials on rumen microbial fermentation. J. Dairy Sci. 75, 1736-1744

Russell J.B., Strobel H.J., 1989. Effects of ionophores on ruminal fermentation. Appl. Environ. Microbiol. 55, 1-6

SAS, 1996. SAS System for Windows, Release 6.12, SAS Institute Inc. Cary, NC

Thornton J.H., Owens F.N., 1981. Monensin supplementation and *in vivo* methane production by steers. J. Anim. Sci. 52, 628-634