Supplemental Enzymes, Yeast Culture and Effective Micro-organism Culture to Enhance the Performance of Rabbits Fed Diets Containing High Levels of Rice Bran

T. Shanmuganathan, K. Samarasinghe* and C. Wenk[1]

Dept. of Animal Science, Faculty of Agriculture, University of Peradeniya, Peradeniya, Sri Lanka

ABSTRACT : An experiment was carried out to study the effects of exogenous enzymes (cellulases and proteases), yeast culture and effective micro-organism (EM) culture on feed digestibility and the performance of rabbits fed rice bran rich diets over a period of ten weeks. Twenty four, 8 to 9 weeks old male and female New Zealand White rabbits were allotted to 4 dietary treatments; a basal (control) feed containing 43% rice bran, basal feed supplemented with either enzymes, yeast culture or EM. Individual feed intake, body weight gain, nutrient digestibility, carcass characteristics and feed cost were studied. Sex of the rabbits had no significant ($p<0.05$) influence on the parameters studied. The control group showed the lowest daily feed intake (104.8 g), body weight gain (12.8 g) and the highest feed/gain ratio (8.20 g/g). The highest daily feed intake (114.3 g), body weight gain (20.42 g) and the lowest feed/gain ratio (5.60) were observed with enzymes. Compared to the control, yeast significantly ($p<0.05$) improved the feed intake, body weight gain and feed/gain ratio by 4.9, 34.4 and 22.0%, respectively, while EM improved ($p<0.05$) them by 4.0, 32.6 and 21.6%, respectively. All the additives improved ($p<0.05$) the digestibility of dry matter, crude protein, crude fiber and energy by 4.9-8.7, 3.6-10.7, 5.9-8.3 and 4.3-6.4%, respectively. Higher weights of pancreas (by 38.5-56.4%) and caecum (by 13.1-26.8%, compared to the control) were recorded with all additives but liver weight was increased only by yeast (24.5%) and enzymes (26.7%). Significantly ($p<0.05$) higher carcass recovery percentages were observed with enzymes (60.55), yeast (60.47) and EM (56.60) as compared to the control (48.52). Enzymes, yeast and EM reduced ($p<0.05$) the feed cost per kg live weight by 23.8, 15.9 and 15.5%, respectively. Results revealed that enzymes, yeast culture and EM can be used to improve the feeding value of agro-industrial by-products for rabbits in Sri Lanka and thereby to reduce the feed cost. Under the present feeding system, enzyme supplement was the best. ***(Asian-Aust. J. Anim. Sci. 2004. Vol 17, No. 5 : 678-683)***

Key Words : Rabbits, Enzymes, Yeast, Effective Micro-organisms, Feed, Digestibility

INTRODUCTION

Rabbits possess various attributes that are advantageous in comparison to other livestock. Rao et al. (1979) and Taylor et al. (1989) noted that rabbit meat is of excellent protein quality, low in total as well as saturated fat, cholesterol and sodium. Therefore, rabbit production is considered as a good source of meat in tropical developing countries where there is an abundance of by-product feedstuffs. Currently commercial rabbits in Sri Lanka are reared mostly on expensive poultry feeds, as commercial feeds are not available for rabbits. This is one of the important factors limiting the expansion of rabbit farming in Sri Lanka. Agro-industrial by-products such as rice bran and coconut poonac, which are relatively inexpensive and readily available in Sri Lanka have great potential for feeding rabbits. However both rice bran and coconut poonac are nutritionally poor in quality due to their high fiber contents. Coconut poonac marketed in Sri Lanka contains 21.8% crude protein, 21.5% crude fiber, 9.4% ether extract and 6.5% ash (Ravindran et al., 1982). Metabolizable energy content of coconut poonac is low due to its high fiber.

Several studies carried out on poultry and pigs have shown that the nutritive value of feed stuffs, especially of poor quality, can be enhanced by supplemental enzymes (Choct and Annison, 1992; Ward, 1995; Cowan et al., 1996). On the other hand, probiotics such as yeast culture can be used to manipulate rumen fermentation in ruminants (Williams and Newbold, 1990). Rabbits may also benefit from probiotic feed additives as they are highly dependent on hind gut fermentation to utilize their feed efficiently. However the use of such feed additives in the rabbit has not been extensively studied. If the feeding value of tropical feeds such as rice bran and coconut poonac for rabbits can be improved with enzymes and probiotics, it will help the rabbit industry remarkably. Therefore, the present study was carried out to investigate the possibility of using exogenous enzymes, yeast culture and effective micro-organism culture (EM; a live microbial culture of Japanese origin) to improve the performance of rabbits fed diets rich in rice bran and coconut poonac.

MATERIALS AND METHODS

A feeding trial was conducted using eight to nine weeks old, 12 male (1,984 g body weight) and 12 female (1,987g

* Corresponding Author: K. Samarasinghe. Tel: +94-08-38-71-79, Fax: +94-08-38-64-92, E-mail: ksamarasinghe2002@yahoo.com

[1] Institute of Animal Science, ETH-Z, CH 8092 Zurich, Switzerland.

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Table 1. Ingredient composition (as fed basis) and the analysed nutrient composition (on DM basis) of the basal feed

Ingredients	Amount (%)	Ingredients	Amount (%)
Rice bran	43	Molasses	4
Coconut poonac	15	Dicalciumphosphate	1.2
Grass meal	15	Salt	0.25
Maize meal	12	Vitamin-mineral premix[1]	0.25
Soybean meal	5	Lysine·HCl	0.25
Fish meal	4	DL-methionine	0.05
Analyzed nutrient composition			
Dry matter (%)	92.08	Crude fiber (%)	13.01
Ash (%)	6.25	Ether extract (%)	5.52
Crude protein (%)	19.75	Gross energy (kcal/kg)	4,900

[1] Contained 150,000 IU, vit. A; 300,000 IU, vit. D_3; 0.3 g, vit. B_1; 0.6 g, vit. B_2; 0.5 g, vit. B_6; 1.5 g, vit. B_{12}; 2.5 g, vit. E; 1.5 g, vit. K; 0.3 g, calcium pantothenate; 0.15 g, folic acid; 3 g, nicotinic acid; 15 g, choline chloride; 0.1 g, cobolt; 5 g, selenium; 4 g, iron; 8 g, manganese; 1 g, copper; 5 g, zinc; 0.1 g, iodine; 5 g, antioxidants and 50 g DL-methionine per kg.

body weight) New Zealand White rabbits over a period of 10 weeks to study the effect of enzymes and probiotics on their performance when fed on rice bran and coconut poonac based diets. Rabbits were housed in individual wire net cages having a floor space of 2,160 sq.cm (72 cm× 30 cm), equipped with nipple drinkers and a nylon net fixed under each cage for faeces collection.

A basal ration (Table 1) containing 43% rice bran (un-defatted) and 15% coconut poonac (expeller extract) was formulated to contain all the nutrients as required by growing rabbits (NRC, 1977). Guinea grass (*Panicum maximum*) was cut at pre-bloom stage, dried under shade for few days, ground to a meal and used as the main source of fiber in the basal ration. The total amount of basal feed required for the trial was mixed as one batch using a horizontal feed mixer. Then the basal feed was divided in to four equal portions and one portion was taken as the control feed. The remaining three portions were supplemented either with enzymes (a mixture of cellulases and proteases at 400 ppm), yeast culture (Yea-Sacc[1026] at 200 ppm) or effective micro-organisms (1 %) to get three test diets.

Effective micro-organisms (EM) culture is a cocktail of live beneficial micro-oragnisms introduced by Japanese scientists (Higa and Parr, 1994). According to Higa and Parr (1994), EM contains selected species of microbes such as photosynthetic bacteria (*Rhodopseudomonas palustris* and *Rhodobactor spaeroides*), lactic acid bacteria (*Lactobacillus plantarum, Lactobacilluscasei* and *Streptococcus lactis*), yeast (*Saccharomyces cereviciae* and *Candida utilis*), ray fungi (*Streptomyces albus* and *Streptomyces griseus*) and fungi (*Aspergillus oryzae* and *Mucor hiemalis*).

All the four rations were separately pelleted using a 3.5 mm die in a pelleting machine (CPM LP 4001-Aerphilly Mid-Glam-Eriez magnetics). In the case of EM treatment, EM culture solution was sprayed on pellets immediately before feeding every day. EM could not be added to the feed before pelleting because the viability of bacteria gets reduced when exposed to sunlight. As recommended by producers, EM solution was also added to drinking water every day at the rate of 1:2,000 (v/v). Rabbits in other treatments received fresh water without EM. Each dietary treatment was then assigned to six rabbits (sex ratio 1:1) in a Randomized Complete Block Design. Feed and water was given *ad libitum*.

Individual feed intake and body weight were recorded weekly. Samples of faeces from individual rabbits were collected separately during three consecutive days for three collection periods starting from 31st, 38th and 45th day after commencement of the experiment. Soon after collection, the samples were stored in a deep freezer until they were taken for analyses. Feed samples were also collected during each collection period and stored for subsequent analyses.

At the termination of the experiment, all the animals were subjected to a slaughter study. They were weighed, stunned by dislocating the neck and decapitated. After bleeding for few minutes, the head, skin and feet were removed. The abdomen was opened by making a cut across and animals were eviscerated manually. The weights of liver, pancreas and empty caecum were recorded. The weight of the carcass without head, feet and giblets was taken to estimate the carcass recovery (dressing percentage).

Faeces samples were dried at 60°C for 48 h and ground using a 1 mm sieve. Feed samples were also ground using a similar sieve size. Dry matter (DM), ash, crude protein (CP), crude fiber (CF) and ether extract (EE) of feeds and DM, CP and CF of faeces were analyzed (AOAC, 1992). Gross Energy of feed and faeces were determined using a bomb calorimeter (Gallenkamp Ballistic bomb calorimeter). Acid insoluble ash (AIA) contents of feed and faeces were determined (AOAC, 1992) and the digestibility values were then calculated according to an indicator method using AIA as the indicator (Prabucki et al., 1975; Scott and Boldaji, 1997).

Data were subjected to Analysis of Variance (ANOVA) using statistical analysis system (SAS, 1990). Feed intake, weight gain, feed conversion ratio (feed intake per unit live weight gain) and nutrient digestibility were statistically

Table 2. Feed intake, weight gain, feed efficiency and carcass characteristics of rabbits as affected by dietary treatments (Mean±SD)

Parameter	Dietary treatments			
	Control	EM	Yeast	Enzymes
Mean daily feed intake (g)	$104.77^{a}\pm0.85$	$108.97^{b}\pm0.89$	$109.89^{b}\pm0.69$	$114.26^{c}\pm0.91$
Male	$105.2^{a}\pm0.85$	$109.1^{b}\pm0.89$	$110.0^{b}\pm0.69$	$115.3^{c}\pm0.81$
Female	$104.3^{a}\pm0.90$	$108.8^{b}\pm0.81$	$109.8^{b}\pm0.71$	$114.5^{c}\pm0.97$
Mean daily weight gain (g)	$12.78^{a}\pm0.25$	$16.95^{b}\pm0.13$	$17.18^{b}\pm0.14$	$20.42^{c}\pm0.21$
Male	$12.91^{a}\pm0.25$	$16.92^{b}\pm0.13$	$17.16^{b}\pm0.14$	$20.52^{c}\pm0.21$
Female	$12.65^{a}\pm0.32$	$16.98^{b}\pm0.32$	$17.20^{b}\pm0.24$	$20.32^{c}\pm0.23$
Mean feed/gain ratio (g/g)	$8.20^{c}\pm0.19$	$6.43^{b}\pm0.20$	$6.40^{b}\pm0.19$	$5.60^{a}\pm0.15$
Male	$8.15^{c}\pm0.09$	$6.50^{b}\pm0.12$	$6.40^{b}\pm0.08$	$5.61^{a}\pm0.16$
Female	$8.24^{c}\pm0.10$	$6.40^{b}\pm0.10$	$6.38^{b}\pm0.09$	$5.63^{a}\pm0.11$
Carcass characteristics (% live weight)				
Mean carcass weight	$48.52^{a}\pm1.61$	$56.60^{b}\pm0.73$	$60.47^{c}\pm1.00$	$60.55^{c}\pm1.11$
Male	$48.23^{a}\pm1.21$	$56.32^{b}\pm0.62$	$60.28^{c}\pm0.69$	$60.48^{c}\pm0.54$
Female	$48.81^{a}\pm0.98$	$56.88^{b}\pm0.70$	$60.66^{c}\pm0.74$	$60.62^{c}\pm0.61$
Mean liver weight	$1.770^{a}\pm0.05$	$1.770^{a}\pm0.08$	$2.203^{b}\pm0.10$	$2.243^{b}\pm0.07$
Mean pancreas weight	$0.039^{a}\pm0.002$	$0.054^{b}\pm0.001$	$0.061^{c}\pm0.002$	$0.061^{c}\pm0.001$
Mean caecum weight	$1.584^{a}\pm0.04$	$1.791^{b}\pm0.03$	$1.993^{c}\pm0.04$	$2.008^{c}\pm0.05$

$^{a, b, c}$ Means with different superscripts in a row are significantly different ($p<0.05$).

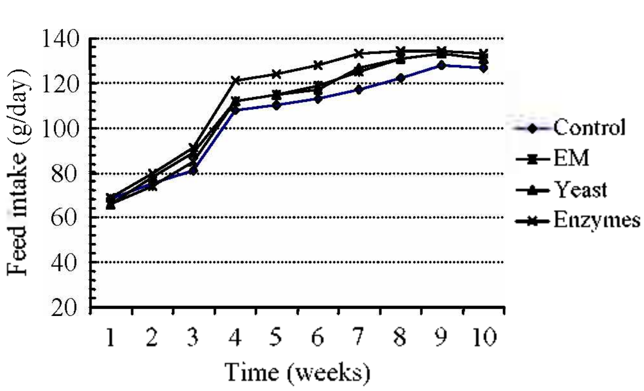

Figure 1. Mean feed intake of rabbits during the experiment.

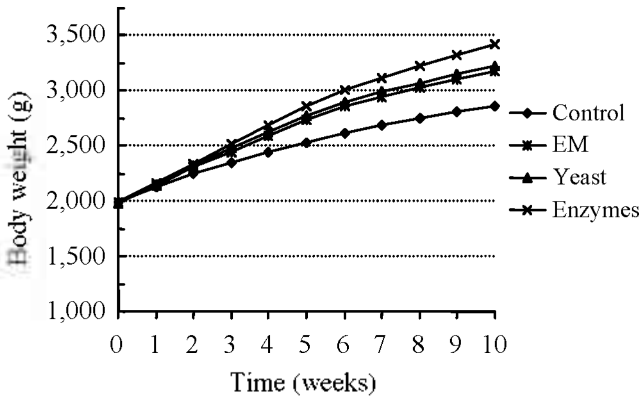

Figure 2. Mean body weight of rabbits during the experiment.

analyzed by considering treatment, sex and time as three factors. Weight of the internal organs and dressing percentages were analyzed by considering treatment and sex as two factors. Means were compared using Duncan's New Multiple Range Test.

RESULTS AND DISCUSSION

The nutrient composition of the basal feed offered to rabbits is presented in Table 1. Though the feed was formulated to contain 16% CP, analytical results show a higher level (19.8%) probably due to higher actual CP levels, especially in rice bran and grass meal, as compared to their table values that were used in formulating the feed. Feed contained somewhat lower crude fiber value than expected (14%) which is also attributed to the variation in ingredient quality. Except for these two, all the other nutrients were within expected levels.

The gender of rabbits did not have any influence on the parameters studies. However, significant influences ($p<0.05$) were observed when the basal diet was supplemented with additives. As shown in Table 2, all the additives have significantly ($p<0.05$) increased the feed intake of rabbits, while enzymes resulted in highest intake (114 g/day). Yeast and EM have increased the feed intake by 4.4% as compared to the control (104.8 g). Increased feed intake has been reported in most of the studies on animals fed yeast culture supplemented diets (Phillips and Tungeln, 1985; Malcom and Kiesling, 1986; Fallon and Harte, 1987; Gunther, 1989; Williams and Newbold, 1990; McLeod et al., 1990; Williams et al., 1991 and Erasmus et al., 1992). Rabbits have consumed more from supplemented diets soon after introducing them and maintained the higher intake throughout the experimental period (Figure 1).

Parallel to the increased feed intake, all the feed additives remarkably increased the growth of rabbits from the beginning of the experiment (Figure 2). Maximum weight gain was observed with enzymes (20.4 g/day)

Table 3. Energy and nutrient digestibility of rabbits and the cost of experimental diets (Mean±SD)

Parameter	Dietary treatments			
	Control	EM	Yeast	Enzymes
Dry matter	$0.610^{a}\pm0.006$	$0.640^{b}\pm0.003$	$0.647^{b}\pm0.004$	$0.663^{b}\pm0.011$
Crude protein	$0.698^{a}\pm0.004$	$0.723^{b}\pm0.005$	$0.762^{c}\pm0.006$	$0.773^{c}\pm0.002$
Crude fiber	$0.169^{a}\pm0.0011$	$0.179^{b}\pm0.0003$	$0.183^{c}\pm0.0002$	$0.183^{c}\pm0.0005$
Energy	$0.654^{a}\pm0.005$	$0.682^{b}\pm0.004$	$0.686^{b}\pm0.003$	$0.696^{b}\pm0.005$
Feed cost/kg live weight (Rs[1])	$102.86^{c}\pm4.22$	$86.91^{b}\pm2.00$	$86.50^{b}\pm2.10$	$78.33^{a}\pm1.80$
Feed cost/kg dressed weight (Rs)	$211.99^{d}\pm7.45$	$153.55^{c}\pm2.52$	$143.05^{b}\pm1.71$	$129.26^{a}\pm2.86$

[a, b, c, d] Means with different superscripts in a row are significantly different ($p<0.05$).

[1] One US$ equals to Sri Lankan Rupee (Rs) 96.57 (Exchange rate in December 2003).

followed by yeast (17.2 g/day) and EM (17.0 g/day) (Table 2). The increased weight gain observed in test groups must have contributed to the increased feed intake.

Though the feed intake and weight gain showed a similar trend among treatments, the enzyme supplementation resulted in significantly ($p<0.05$) lower feed conversion ratio (FCR) (5.6) compared to other treatments. The FCR observed with yeast (6.40) and EM (6.43) were also lower ($p<0.05$) than that of the control (8.20). This indicates that the improvement in growth rate was higher than the increased rate of feed intake thus showing a better nutrient utilisation with additives. This is well established with regard to enzymes and yeast (Rexen, 1981; Hollister et al., 1990; Schutte et al., 1993; Singh et al., 1995). According to Singh et al. (1995), feed required per unit gain was reduced by 3% in NZW rabbits given *Saccharomyces cerevisiae* (strain N0. 2094, ITCCF). In the same experiment, about 5% improvement in body weight gain had been observed. Hollister et al. (1990) also reported an improved ($p<0.05$) feed:gain ratio in rabbits fed on diets with Lacto-Sacc. The results of the present study are in line with earlier reports. Though such information on EM is scantly, Li and Umemura (1995) and Inciong (1996) have reported higher feed efficiency of chicken fed diets supplemented with EM.

However, the FCR values obtained in the present study were less comparable to the values reported by most of the other workers (Raharjo et al., 1988; Maertens and Villamide, 1998). Raharjo et al. (1988) reported a FCR of 3.12 when 40% rice bran diet supplemented with essential nutrients was fed to rabbits during 5 to 10 weeks of age. Abd El-Baki et al. (1993) reported an average FCR of 5.65 in New Zealand White rabbits fed a commercial feed from their 5th week to the 20th week of age. In their experiment, the initial body weight of rabbits was 735 g and the average body weight gain was 20.2 g/day. In the present study, the rabbits had an initial body weight of 1,986 g and an average weight gain of 12.65 g/day (Control group) mainly due to their higher age. Therefore the age difference must be the main reason for higher FCR values observed in the present study. Further, it should be noted that the FCR value and the daily weight gain reported by Abd El-Baki et al. (1993) are well comparable to the values obtained for the enzyme group in the present study.

As expected, apparent nutrient digestibility values showed no differences between males and females. It is reported that no sex differences could be found with respect to digestion either in the growing or adult rabbits (Fekete and Lebas cited by Fekete, 1988; Fekete and Bokori, cited by Fekete, 1988). These findings are in agreement with the present results. However, all additives tested had a significant effect ($p<0.05$) on energy and nutrient digestibility (Table 3). Enzyme supplementation resulted in maximum digestibility values of dry matter (0.663), crude protein (0.773), crude fiber (0.183) and energy (0.696). EM and yeast increased the above values by 5.5, 6.4, 7.1 and 4.6%, respectively, whereas yeast increased the crude protein digestibility by 9.2% while EM increased it by only 3.6% ($p<0.05$) as compared to the control. The highest nutrient digestibility with enzyme supplemented feed was also reflected by the lowest FCR. It is a well known fact that rabbits can get a part of their energy requirement through fiber degradation in the hind gut. High crude fiber digestion with test diets may have contributed to an elevated energy digestibility. Therefore it is clear that the faster growth rate observed with test diets was primarily due to increased digestibility of nutrients and energy, which must have resulted in better utilization of nutrients for growth.

Maertens and de Groote (1992) have reported that yeast can colonize in the gut and contribute to the maintenance of flora equilibrium in rabbits. Improved colonization and self-replication of beneficial microbes with yeast and EM might have helped in gut fermentation in the present study, which in turn has helped in improved feed digestion. The improvement in digestion of feeds in monogastrics and ruminants supplemented with probiotics was reported by several workers (Glade, 1991; Williams et al., 1991; Erasmus et al., 1992). According to the producers, EM consists of *lactobacillus spp*. Therefore, it may show a similar mode of action of other probiotics in colonization of intestine to improve nutrient digestion.

Enzymes and yeast remarkably increased the carcass recovery by 24.7% while EM increased it by 16.7% in spite of heavier giblets (Table 2). This improvement in carcass yield could be attributed to better nutrient utilization as observed by Gowda et al. (1996). As shown in Table 2, enzyme and yeast supplements have increased the weights of liver, pancreas and caecum of rabbits. Addition of EM has increased the weights of pancreas and caecum but not the liver. However, the reasons for heavier organs observed with additives are not clear.

Feed cost analysis

In this study, labor and transportation costs involved in feeding were not included in the calculation, but the cost of additives were considered. Since labor and transportation costs were similar in all test diets, the present comparisons are justified. Among the treatments tested, enzyme supplementation had the lowest feed cost per live weight as well as dressed weight compared to other treatments (Table 3). Yeast and EM also reduced the feed cost markedly. Present data revealed that enzyme supplementation is the most economical feeding strategy, which would give more profit to the farmer. When live weight of rabbit is considered, the cost of feed can be reduced by 24% with enzyme supplements. When the same was calculated on dressed weight basis, the feed cost could be reduced by 39% with enzyme supplements.

CONCLUSIONS

Appropriate enzymes, yeast and effective micro organism (EM) cultures significantly increase the body weight gain, feed intake, feed efficiency, nutrient digestibility and carcass recovery in rabbits fed on rice bran rich diets. The feed cost can be reduced remarkably by using such additives. Among the additives tested, enzymes resulted in the maximum beneficial effects under the present feeding system.

Present findings clearly demonstrate that rabbits can be produced more economically by feeding locally available, inexpensive agro-industrial by-products supplemented with appropriate enzymes, yeast or EM culture.

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