

Growth Performance and Caecal Fermentation in Growing Rabbits Fed on Diets Containing Graded Levels of Mulberry (*Morus alba*) Leaves

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ABSTRACT : Growth performance, nutrient digestibility and changes in caecal fermentation pattern was studied on four groups of 8 grower rabbits (soviet chinchilla) each, fed on diets containing 17 to 18% CP and 10.4-11.00 MJ DE kg⁻¹ feed. The complete diets contained mulberry leaves along with other feed ingredients at 0 (LH), 15 (ML15), 30 (ML30) and 45% (ML45) levels. In LH diet ground lucerne hay was added at 15% of the diet as roughage source. The diets were uniform in other nutritional parameters except the higher amount of hemicellulose (18.7 and 16.5%) in LH and ML15 diets. The performance of grower rabbits was better ($p < 0.05$) on LH and ML15 diets in terms of twelve weeks weight (kg), total gain (g), average daily gain (ADG) and feed efficiency (%). Different levels of mulberry leaves in the diet did not influence the dry matter intake. Digestibility of nutrients for DM, OM, CP, NDF and energy was uniform in all the diets but the digestibility of CF, ADF and cellulose was higher ($p < 0.01$) on ML15, ML30 and ML 45 diets. The hemicellulose digestibility was significantly higher ($p < 0.01$) on LH (44.52%) and ML15 (48.00%) compared to ML30 (33.54%) and ML 45 (39.17%) diets. The nitrogen retention (% of intake) was higher ($p < 0.05$) in LH and ML15 diets than ML30 and ML45 diets. The caecum weight as percent of intestine weight consistently increased ($p < 0.05$) with increasing content of mulberry leaves in diets. Total nitrogen and NH₃-N was higher on LH and ML15 diets. It is concluded that incorporation of mulberry leaves can replace Lucerne hay in complete rabbit feed (15% in diet). Even at higher level i.e. 30 and 45% of the diet mulberry leaves based complete feed gave promising results. (*Asian-Aust. J. Anim. Sci. 2003. Vol 16, No. 9 : 1309-1314*)

Key Words : Rabbit, Growth, Tree Leaves, Mulberry Leaves, Caecal Fermentation, Nutrient Utilization

INTRODUCTION

Better growth performance is desired in grower rabbits after weaning. Rabbits are generally fed on high energy diets containing starch exhibiting frequent digestive troubles (primarily gastroenteritis) linked with changes in caecal fermentation pattern of sick animals (Franck and Coulmin, 1978; Peeters and Charlier, 1984; De Blas et al., 1986; Peeters and Maerten, 1988; Bellier, 1994). The favourable effect of fibres with respect to resistance to pathogenic agents was clearly shown only recently (Licois and Gidenne, 1999). However increasing crude fibre (CF) contents of the diet to reduce gastric problems leads to reduce digestive energy and protein content (Perez et al., 1994), which become insufficient to meet the requirement of growing rabbits. The botanic origin of fibre influence the digestion and caecal microbial activity, irrespective of the quantity of the fibre (Gidenne, 2000). Though CF is still taken in to account while formulating rabbit diets but favourable effect of ADF supply on the frequency of the digestive disorder and mortality in fattening rabbits was shown by Maitre et al. (1990). Further the importance of digestible fibre fraction constituted mainly of hemicellulose can not be ignored in rabbit, as on one side it reduce gastric problems on the other side it can substitute starch with out

any adverse effect on growth performance (i.e. feed conversion, growth rate) (Jehl and Gidenne, 1996; Gidenne et al., 1998) suggesting that digestible fibres are efficiently utilized by the growing rabbits. Economisation of feed cost using cheaper feed resources (Bhatt and Sharma, 2001; Muriu et al., 2002; Vasanthakumar et al., 1999) is another important aspect for commercial rabbit production and formulation of economic diets with use of good quality fodders is not always possible at many places in tropical countries where animal population is large so alternative fodders are to be tried.

Thus the aim of this study was to examine the possibility of using dried mulberry leaves (ML) in the diet of grower rabbits replacing full quantity of Lucerne hay (LH) and to test the feasibility of using mulberry leaves at higher level as a source of energy and protein on performance and caecal fermentation pattern in growing rabbits.

MATERIALS AND METHODS

Diets

Green mulberry (*Morus alba*) leaves were collected from the Institute farm, dried under shed and grounded in hammer mill to pass through 4 mm sieve. Complete pelleted diets were prepared as per the ingredient composition presented in Table 1. Feed pellets were prepared after mixing ground ingredients with water containing 8 kg molasses, equivalent to 25% of moisture.

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Table 1. Ingredients and chemical composition (% on DM basis) of diets

Parameters	LH	ML15	ML30	ML45	Mulberry leaves
Ingredient composition (%)					
Groundnut cake	25	20	18	15	
Barley	20	30	25	15	
Wheat bran	15	10	7	5	
Deoiled rice bran	10	10	5	5	
Lucerne hay	15	-	-	-	
Mulberry leaves	-	15	30	45	
Fish meal	5	5	5	5	
Mineral mixture	1.5	1.5	1.5	1.5	
Salt	0.5	0.5	0.5	0.5	
Molasses	8	8	8	8	
Chemical composition (%)					
Organic matter	95.10	95.30	94.10	94.10	84.88
Crude protein	18.00	17.44	17.41	17.38	15.30
Crude fibre	14.85	12.00	12.60	12.65	11.63
NDF	39.40	36.40	30.90	29.90	50.45
ADF	20.65	20.25	20.55	19.75	36.65
Hemicellulose*	18.75	16.15	10.35	10.15	13.80
Cellulose	12.80	10.50	10.10	9.80	25.50
Lignin	5.30	4.20	4.50	4.20	3.74
GE (MJ/kg DM)	18.07	17.28	17.36	17.74	-
DE (MJ/kg DM)	10.72	11.00	10.41	10.41	-
Lignin/cellulose ratio	0.41	0.40	0.44	0.42	0.14

* NDF-ADF.

Prepared feed mix was passed through 4 mm die cast in a laboratory model pelleting machine and the extruded pellets were sun dried for removing moisture left in pellets and stored in gunny bags for use in the feeding experiment.

Season and temperature

The study was conducted at Central Sheep and Wool Research Institute, Avikanagar. Experimental animals were housed in asbestos roofed open sided shed in individual pens. The experiment was started in second week of January and terminated by the end of March. During experimentation the ambient temperature and RH of the animal shed was $7.6 \pm 0.3^\circ\text{C}$ and 41% minimum and $29.0 \pm 0.5^\circ\text{C}$ and 75 maximum respectively.

Animals, feeding and measurement of growth performance

Thirty two weaners (4 week) old rabbits of Soviet Chinchilla breeds were randomly assigned to four groups of eight animals each as and when they were weaned from their mothers. The rabbits were maintained in individual cages (45 cm \times 50 cm \times 37 cm) made up of wire mesh supported with welded angle iron frame having provision of feeder and watering bowls. The animals were offered *ad libitum* pelleted feed once daily and refusal of previous day was weighed and discarded before offering fresh feed. Animals had free access to clean drinking water in

individual bowls fixed in the cages. Feed intake was determined daily and the animals were weighed weekly through out the experimental period for observing growth performance. The feeding of animals was terminated when they completed 12 weeks (84 days) of age.

Digestibility trial and collection of feed, faeces and urine samples

The metabolism trial of five days duration was conducted on all animals in the middle of feeding experiment by attaching metabolism funnels to the individual cages. The metabolism funnel had the provision of automatic and quantitative collection of faeces and urine separately. During metabolism trial most of the animals had attained the age of 9 weeks with average body weight of 1.2 ± 0.17 to 1.71 ± 0.17 kg in different groups. An aliquot equivalent to 1/2 of faeces was collected daily and pooled together for nitrogen estimation while the dry matter of fixed quantity was estimated daily. An aliquot equivalent to 1/5 of urine voided was added in a Kjeldahl flask containing 25 ml of concentrated sulphuric acid, daily for nitrogen estimation. A representative 100 g sample of feed was collected from the bulk for further analysis.

Caecal digesta sampling procedure

Collection of caecal digesta was performed by slaughtering five animals from each group at 14 h after cervical dislocation and deskinning the animal. The total digestive tract was removed by severing the middle line of the stomach. The total digestive tract was taken immediately, after ward caecum was separated and weighed. The samples of caecum digestive content were collected in wide mouth 200 ml plastic bottles by gentle squeezing. The empty caecum was then cleared by water flushing and weighed after draining extra water. The caecum contents were than sampled for various parameters. Ten grams of caecal contents were added in the Kjeldahl flask containing 25 ml of concentrated sulphuric acid for total nitrogen estimation. The pH of the contents was taken by diluting 10 g of contents with 15 ml of distilled water. The samples for $\text{NH}_3\text{-N}$ and total volatile fatty acids (TVFA) estimation was done as per the procedure of Garcia et al. (1996). In which 10 g sample was acidified with 10 ml 0.2 M HCl centrifuged at 5,000 rpm for 15 minutes and supernatant was used for $\text{NH}_3\text{-N}$ estimation. A separate 20 g caecal content was preserved with 2 ml of solution containing 5% orthophosphoric acid (v/v)+1% HgCl_2 (w/v) in equal quantity. The preserved samples were after ward diluted with 20 ml of distilled water and centrifuged at 5,000 rpm for 15 minutes for sedimentation of the feed particles. The supernatant solution was used for estimation of total-N, TCA perceptible N (TCA-N) and TVFA estimation.

Table 2. Performance of broiler rabbits maintained on lucerne hay (LH) and mulberry leaves (ML15, ML30, ML45%) containing diets

Parameters	LH	ML15	ML30	ML45	SEM
Four week weight (g)	417.50	427.50	385.00	418.70	25.872
Twelve week weight (g)	2,170.00 ^b	2,111.00 ^{ab}	1,865.00 ^a	1,842.5 ^a	95.380
Total gain (g)	1,752.00 ^c	1,683.7 ^{bc}	1,480.00 ^{ab}	1,423.70 ^a	81.209
ADG (g)	31.29 ^c	30.06 ^{bc}	26.42 ^{ab}	25.41 ^a	1.450
Total feed intake (g)	6,532.70	6,391.70	6,099.50	6,379.80	395.748
Feed intake (g/d)	116.62	114.34	108.88	113.86	7.076
Feed efficiency (%)	26.83 ^b	26.76 ^b	24.77 ^{ab}	22.48 ^a	0.988

Values bearing unlike superscripts in a row differ significantly. ^{a,b,c} (p<0.05).

Table 3. Feed intake, digestibility and balance of nutrients in broiler rabbits maintained on lucerne hay (LH) and mulberry leaves (15, 30, 45%) containing diets

Parameters	LH	ML15	ML30	ML45	SEM
DMI (g d ⁻¹)	126.38	123.02	96.44	114.74	12.809
DMI (g kg ⁻¹ w ^{0.75})	91.64	83.85	84.65	89.31	7.932
Digestibility coefficients (%)					
Dry matter	59.08	64.05	63.78	63.90	1.814
Organic matter	60.13	65.67	65.61	65.59	1.991
Crude protein	70.80	72.80	68.34	67.98	2.089
Crude fibre	13.60 ^A	23.31 ^B	30.86 ^C	38.35 ^D	0.896
NDF	28.77	37.32	29.80	31.14	3.172
ADF	14.50 ^A	28.74 ^B	27.90 ^B	25.44 ^B	3.685
Hemicellulose	44.52 ^b	48.00 ^b	33.54 ^a	39.17 ^{ab}	3.126
Cellulose	20.63 ^A	34.91 ^{AB}	40.23 ^B	45.23 ^B	3.923
Energy	59.35	63.72	59.96	58.70	2.272
Intake					
DCP (g d ⁻¹)	16.07	15.56	11.29	13.53	1.442
DCP (g kg ⁻¹ w ^{0.75})	11.72	10.52	10.01	10.54	0.976
DE (MJ d ⁻¹)	1.35	1.35	0.98	1.18	0.125
DE (MJ kg ⁻¹ w ^{0.75})	0.99	0.92	0.87	0.93	0.086
DCP (g MJ ⁻¹ DE)	11.83	11.42	11.50	11.33	0.126
Nitrogen					
Intake (g d ⁻¹)	3.70	3.42	2.68	3.18	0.360
Voided in faeces (g d ⁻¹)	1.08	1.00	0.87	1.02	0.148
Voided in urine (g d ⁻¹)	0.47	0.34	0.42	0.45	0.043
N-balance (g d ⁻¹)	2.14	2.07	1.38	1.70	0.214
N retention (% of intake)	57.53 ^{bc}	60.79 ^c	51.68 ^a	53.14 ^{ab}	1.950

Values bearing unlike superscripts in a row differ significantly. ^{a,b,c} (p<0.05), ^{A,B,C,D} (p<0.01).

Analytical procedure

The analysis of feed, faeces and urine was done in duplicate. Dry matter (DM) content was determined by drying samples at 80°C for 48 h. Nitrogen (N) was measured by Kjeldahl procedure and converted to crude protein (CP) using factor 6.25. NDF, ADF, cellulose and ADL were determined according to procedure of Van Soest (1991). TVFA were measured by distillation method (Barnett and Reid, 1957) and NH₃-N by Conway's diffusion cell method (Conway, 1957). Gross energy (GE) was estimated by ballistic bomb calorimeter (Gallenkamp, UK).

Statistical procedure

Data on growth performance, digestibility of nutrient and different biochemical parameters was statistically analysed by one way analysis of variance using SPSS base 10 (SPSS, Inc. Chicago USA).

RESULTS

The diets were prepared with increasing level of mulberry leaves at 15 (ML15), 30 (ML30) and 45% (ML45) of the total diet. In control (LH) diet in place of mulberry leaves ground Lucerne hay was used at 15% (Table 2). C F, NDF and Hemicellulose contents were low in ML supplemented diets compared to LH diet. ADF contents were not affected by increasing the mulberry leaves in diets. CP, GE and digestible energy (DE) values remained uniform as per the desired formulation of diets.

Performance and feed intake during growth period

The twelve week weights of the rabbits was significantly (p<0.01) higher in LH and ML15 diet compared to ML30 and ML45 diets. On average weight were higher by 316.25 and 257.2 g in LH and ML15 diets than ML30 and ML40 groups. The total gain during the growth period of 56 days and average daily gains followed the trend of twelve week weights, though the total feed intake during growth phase and feed intake day⁻¹ did not differ significantly among the groups and ranged between 6,099.5 to 6,532.7 g and 108.88 to 116.62 g respectively. Presentation of dry matter intake (DMI) during metabolism trial on (g/d) and (g/kg W^{0.75}) basis was similar. Percent feed efficiency was higher in LH and ML15 diets suggesting better utilization of these diets by grower rabbits.

Digestibility and balance of nutrient during metabolism trial

Digestibility coefficients of nutrient for DM, OM and CP were similar in different groups. Digestibility of CF consistently increased with increasing amount of mulberry leaves from ML15 to ML45 diet. Incorporation of mulberry leaves at a similar rate to that of ground Lucerne hay i.e. 15% of diet also resulted in better crude fibre digestibility in ML15 diet (23.31%) compared to LH diet (13.60%). The

digestibility of ADF and cellulose was similar but higher in ML15, ML30 and ML45 diets as compared to LH diet.

There was no significant difference in apparent digestibility of energy. Protein was also equally digested in different groups and thus the supply of digestible crude protein and energy did not differ significantly in various groups, leading to a similar energy protein (DCP/DE) ratio. Though there is slight variation in intake of DCP and DE when presented on g day⁻¹ or MJ day⁻¹ basis (Table 3). Nitrogen intake and balance was higher in LH and ML15 diets though it did not achieve statistical significance and was 3.70 and 3.42; 2.14 and 2.07 g/d respectively. The excretion of ingested N through faeces and urine was uniform in all the diets. The N retention was better on LH (57.53) and ML15 (60.79%) fed diets compared to ML30 (51.68%) and ML45 (53.14%) diets.

Gut weight, caecum weight and changes in caecal fermentation pattern

Intestine weight did not vary between groups due to addition of mulberry leaves at different level in diets (Table 4). Caecum weight (inclusive of caecal content) consistently increased with increasing level of mulberry leaves in the diets. Caecum weight (inclusive of caecal content) was higher even when mulberry leaves were added at 15% of the diet compared to LH diet where ground Lucerne hay was added 15% of the diet. Caecum content weight followed the trend of caecum weight. Caecum weight as per cent of Intestine weight was significantly ($p < 0.05$) higher in mulberry leaves incorporated diets. Empty caecum weight was uniform in different groups. The mean caecal DM level

was 269 g kg⁻¹ across the treatments.

Increasing the level of mulberry leaves in diet resulted in slight decrease in pH of caecal content from ML15 to ML45. A significant decrease in caecal liquor total N content was registered with increase in mulberry leaves in diet from ML15 to ML45. The decrease was to the tune of 50 mg from ML15 to ML45. Similarly NH₃-N decreased from 17.54 in LH to mulberry leaves incorporated diets say 14.89, 7.03 and 7.18 m mol litre⁻¹ liquor in ML15, ML30 and ML45 diets, respectively. The decrease in NH₃-N of mulberry leaves incorporated diet was observed only up to 30% level of mulberry leaves incorporation. An increase ($p < 0.05$) in TVFA concentration was observed on all three mulberry leaves incorporated diets averaging to 62.9 as compared to 48.75 m mol litre⁻¹ in LH diet.

DISCUSSION

The crude protein content in the diet was uniform as per the desired formulation. The crude fibre of ML15, ML30 and ML45 diets was about 2.5 units less than LH diets even after incorporation of mulberry leaves at 30 and 45% of diet due to higher cell contents of mulberry leaves. The hemicellulose portion was higher in LH and ML15 diets due to presence of higher amount of wheat bran and de-oiled rice bran in these diets and is in agreement to earlier reports (Jehl and Gidenne, 1996). The digestible energy content of the diet was in the range of 10.41-11.00 MJ kg⁻¹ DM and is in accordance with the recent requirements for the grower rabbits (Gidenne and Jehl, 1996). The higher ($p < 0.05$) 12 week weight of grower rabbits (2,170 and 2,111

Table 4. Caecal characteristics of broiler rabbits maintained on lucerne hay (LH) and mulberry leaves (ML15, ML30, ML45%) containing diets

Parameters	LH	ML15	ML30	ML45	SEM
Intestine weight (g)	483.75	477.50	481.25	490.00	38.554
Caecum weight (g)	138.75	165.00	182.50	190.00	14.892
Empty caecum weight (g)	33.00	33.75	35.25	33.25	3.062
Caecum content weight (g)	105.75	131.25	147.25	156.75	13.745
Intestine weight (% of body wt.)	23.21	24.21	24.95	26.30	1.998
Caecum weight (% of intestine wt.)	28.77 ^a	35.09 ^{ab}	37.77 ^b	38.85 ^b	6.750
Empty caecum weight (% of intestine wt.)	6.84	7.08	7.47	7.16	0.636
Caecum content DM (%)	27.01	27.71	25.95	26.94	1.045
Total N of caecum content (mg/g of content)	10.92	11.02	10.15	10.60	0.870
pH of caecal content	6.19	6.08	5.99	5.93	0.061
Caecal liquor					
Total N (mg dl ⁻¹)**	378.00 ^b	329.50 ^{ab}	297.50 ^a	280.0 ^a	19.77
TCA N (mg dl ⁻¹)**	251.25	217.50	192.50	195.50	21.960
NH ₃ -N (m mole l ⁻¹)*	17.54 ^B	14.89 ^B	7.03 ^A	7.18 ^A	0.078
Non protein N (mg dl ⁻¹)	126.75	112.00	105.00	85.00	11.400
TVFA (m mol l ⁻¹)**	48.75 ^a	61.50 ^b	67.00 ^c	60.25 ^c	5.125

* 10 g caecal content was diluted with 10 ml of 0.2 M HCl and centrifuged at 5,000 rpm for 15 minutes and the supernatant solution was used for NH₃-N estimation.

** 20 g caecal contents was preserved with 2 ml of solution containing 5% orthophosphoric acid (w/v)+1% HgCl₂ (w/v) in equal quantity. The contents were then diluted with 20 ml of water and centrifuged at 5,000 rpm for 15 minutes and the supernatant solution was used for estimation of these parameters. Values bearing unlike superscripts in a row differ significantly. ^{a, b, c} ($p < 0.05$). ^{A, B} ($p < 0.01$).

g) on LH and ML15 diets was due to better feed efficiency, which is related to production of higher microbial protein as supported by higher amount of TCA precipitable-N in caecal liquor. These diets had higher amount of wheat bran and de-oiled rice bran which contained higher amount of hemicellulose favouring better microbial protein production (Jehl and Gidenne, 1996; Candau et al., 1978; Bellier, 1994; Yangxi et al., 2002). Though the 12-week weight, total gain and average daily gains were lower on ML30 and ML45 diets compared to LH and ML15 diets but were satisfactory from production point of view.

The intake of cellulose and lignin content in all the diets were within the latest normal prescribed limits of 11-12 g d⁻¹ and 5-7 g d⁻¹ (Gidenne, 2000) so these constituents did not result in any adverse effect on the digestibility of DM, OM, CP and GE (Perez et al., 1994). The presence of fibre viz. crude fibre, ADF and cellulose in unrefined form favoured the better digestibility (Chiou et al., 1994 and Gidenne and Perez, 1994) of these constituents on ML15, ML30 and ML45 mulberry leaves diets. The digestible fibre i. e. hemicellulose was present in more quantity in LH and ML15 diets and so was the digestibility of hemicellulose was higher in these diets (Merino and Carabano, 1972; Candau et al., 1978; Motta, 1990). The intake of DCP and DE was in accordance with the latest recommendations (Gidenne, 2000) for growing rabbits and ranged between 10.01 to 11.72 g d⁻¹ and 0.87 to 0.99 MJ kg⁻¹ W^{0.75}. A ratio between DCP/MJ DE of 11.33 to 11.83 was observed on different diets. The better nitrogen balance and N retention in LH and ML15 diets seems to be the effect of higher nitrogen intake and production of more amount of microbial mass on these diets.

Increasing level of mulberry leaves in diets resulted in consistent increase in caecum weight (% of intestine weight), however the empty caecum weight (wall weight) remain the same. The pH of caecal content decreased slightly with increasing levels of mulberry leaves in diet due to higher amount of TVFA on ML15, ML30 and ML45 diets, though the value did not achieve statistical significance. These results are related to the studies reported by Garcia et al. (1992) and Carabano et al. (1997) using sugar beet pulp for barley grain or Lucerne hay with increasing acidity of caecal content. The TVFA production is within the normal range of reported values (Fraga et al., 1984; Garcia et al., 1995a). Our values of ammonia nitrogen are higher than reported values in literature (Merino and Carabano, 1992; Motta et al., 1996) but these values are closed to the values reported by Morisse et al. (1985). Higher (p<0.01) total N and ammonia N concentration in caecal content on LH and ML15 diets associated with higher digestible fibre content and its nature of binding. However, these results are not in confirmity of negative influence of dietary fibre on CP of caecal content (Carabano et al., 1988; Garcia et al., 1995b).

CONCLUSIONS

Incorporation of mulberry leaves in grower rabbit diets showed the comparable performance to that of LH up to 15 per cent of the diet. At 30 and 45 per cent though the 12-week body weights were lower but viable from production point of view. Utilization of nutrients for crude fibre, ADF and cellulose was better on mulberry leaves incorporated diets. Diets containing higher amount of digestible fibre resulted in better performance of the rabbit. Increase in caecum weight was observed with increasing level of mulberry leaves in the diet. The use of mulberry leaves is quite feasible in grower rabbit diets up to 15 per cent level and even at higher level is practicable to use. This may help in reducing the feeding cost in the areas where good quality fodders are not available.

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