

## Effect of Growth Performance and N-Balance of Growing Raccoon Dogs Fed Reduced Crude Protein, Lysine and DL-Methionine Supplementation Diets

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**Abstract:** A production trial was carried out on 100 male growing raccoon dogs to study the effects of low protein diets with Lysine (Lys) and DL-Methionine (Met) supplementation on growth performance and N-balance. The treatment codes were P30, P27, P24, P21 and P18. The protein contents [g kg<sup>-1</sup> DM (dry matter)] of the diets were 295, 268, 237, 202 and 175, respectively. Lysine and DL-methionine were added to P27, P24, P21 and P18 to yield a total dietary content of Met and Lys corresponding to that in P30. From July to mid-September, Average Daily Feed Intake (ADFI), Average Daily Gain (ADG), Nitrogen (N) intake, Faecal Nitrogen (FN), Urinary Nitrogen (UN) and N retention were significantly reduced with dietary protein level decreased ( $p < 0.05$ ), no significant differences among P30, P27 and P24 ( $p > 0.05$ ). In conclusion, when the dietary protein decreased to 24% with Met and Lys supplementation for raccoon dogs would be beneficial in terms of reduced feed expenses and lower nitrogen emissions to the environment.

**Key words:** Raccoon dog, low protein diet, amino acid, N-balance, ADG, China

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### INTRODUCTION

The raccoon dog (*Nyctereutes procyonoides*), an omnivorous canid is a carnivorous mammal of the Canidae family. The diet of raccoon dog thus contains a relatively high proportion of animal protein. As protein is the most expensive dietary nutrient, any reduction in protein level contributes to a saving in production cost. A lot of experiments had proved that supplementing low-protein diets with amino acids may increase the metabolisable value of the diet, develop the growth performance of pig and poultry (Easter and Baker, 1980; Kerr *et al.*, 1995). The experimental data of another fur animal showed that as the Lys and Met were added in diet and protein level was reduced reasonably, it could significantly improve the performance and digestive metabolism of growing blue fox (Zhang *et al.*, 2008). Another study reported that methionine supplementation may offer possibilities to reduce the dietary levels of protein to below current requirements as shown in mink without adverse effects on late growth and fur quality. Reduced the dietary protein supplemented with amino acid should make the dietary protein level as close to the requirement as possible and a high digestibility of N is preferable and that should also minimize the N excretion from feces and urine to the environment.

The objective of the present study was to investigate the influence of low levels of dietary protein with Met and

Lys supplementation, on the performance and N-balance of growing raccoon dogs and found the optimal protein level of raccoon dog's diet.

### MATERIALS AND METHODS

The experiment was conducted at the Fur Animal Breeding Base of the Institute of Special Economic Animal and Plant Sciences, the Chinese Academy of Agricultural Sciences (44.02N, 126.15E) in the Northeast of China. The animals used in this research in the period from July to middle September in 2008. The experimental raccoon dogs were housed in roofed standard sheds with open sides.

**Animals and experimental design:** The experiment began on 27 July with a total of 100 weaned raccoon dog males. The average weight of the animals at the start was  $2.60 \pm 0.17$  kg. The animals were allotted to five treatment groups of 20 animals each. The experiment was preceded by 1 week adjustment period, during which the animals were accustomed to the experimental feed. The experimental treatments were: P30 and P27, P24, P21, P18M with Met and Lys supplementation. Animals had free access to drinking water and were fed twice a day at 08:00 a.m. and 16:00 p.m. The design of the experiment diets were shown in Table 1. All training and testing of the animals was performed by the same person. Individual body weights were recorded at began and the end of experiment.

**N-balance experiments:** The N-balance experiments were carried out with eight kits from the respective treatment groups. The faecal and urine collection period lasted 3 days after the experiment start 15 days. Faeces and urine were collected quantitatively daily and kept frozen pending analyses. To avoid ammonia evaporation from the urine, 20 mL sulphuric acid (5% solution) was added to the urine collection bottles and the urine collection trays were sprayed with citric acid (20% solution) once per day. In the N-balance calculations, retained N was determined as ingested N- (FN+UN).

**Chemical analyses:** The chemical composition of feed and faeces was analysed by standard methods. Dry Matter (DM), ash and Crude Protein (CP: Kjeldahl-N $\times$ 6.25), calcium and phosphorus contents were analyzed according to AOAC (2003) procedures. Crude Carbohydrate (CC) was calculated as the difference by

subtracting ash, CP and EE from the DM content. The calculation of ME content and the proportional composition of ME were based on the digestibility coefficients achieved and the following values of ME: protein 18.8 MJ kg<sup>-1</sup>, fat (EE) 39.8 MJ kg<sup>-1</sup> and carbohydrate 17.6 MJ kg<sup>-1</sup> (Hansen *et al.*, 1991). The concentration of amino acids were determined on Agilent 1100 High Performance Liquid Chromatographic (Agilent Technologies, Inc. Santa Clara. USA).

**Statistical analyses:** The data were subjected to analysis of variance with the General Linear Model (GLM) procedure of SAS (Statistical Analysis Systems Institute, 1989). The models that were used were a linear regression. Factors which were found to have significant impact were included in the model as a covariate. The differences between the diets were tested by means of the least significant difference method in the statistical model. Probability values  $p < 0.05$  were considered statistically significant.

## RESULTS

**Feed intake and growth performance:** Effects of low protein, Lys and Met supplemented diets on performance of growing raccoon dogs were shown in Table 2. The results showed that reducing dietary protein decreased the weight gain of raccoon dogs ( $p < 0.05$ ). Furthermore, lowering dietary protein with Met and Lys addition also decreased ADFI ( $p < 0.05$ ) and ADG ( $p < 0.05$ ). However, the final weight, ADFI and ADG had no significant differences among P30, P27 and P24 ( $p > 0.05$ ). The ADFI final weights and ADG of raccoon dogs in P21 and P18 groups were significant lower than other treatments.

**Nutrients digestibility:** The results of nutrients digestibility of items were shown in Table 3. Reducing dietary protein with Met and Lys addition significantly influenced the digestibility of DM, N and fat ( $p < 0.05$ ). The digestibility of DM, N and fat had the same variational trend with dietary protein decreased and amino acids supplemented in diets. They were all decreased with

Table 1: Composition and nutrient levels of diets (g kg<sup>-1</sup>)

Items	P30	P27	P24	P21	P18
<b>Ingredients<sup>a</sup></b>					
Extruded corn	350.0	402.0	446.0	512.0	630.0
Soybean meal	186.0	130.0	100.0	70.0	50.0
DDGS <sup>b</sup>	50.0	50.0	50.0	50.0	60.0
Corn germ meal	150.0	180.0	210.0	210.0	120.0
Fish meal	200.0	170.0	120.0	80.0	60.0
Soy oil	40.0	40.0	42.0	43.0	43.0
CaHPO <sub>4</sub>	10.0	10.0	10.0	10.0	10.0
NaCl	4.0	4.0	4.0	4.0	4.0
Lysine	-	3.0	6.0	8.5	10.0
Methionine	-	1.0	2.0	2.5	3.0
Premix <sup>c</sup>	10.0	10.0	10.0	10.0	10.0
Total	100.0	100.00	100.0	100.0	100.0
<b>Nutrient levels<sup>d</sup></b>					
ME (MJ kg <sup>-1</sup> )	12.1	12.2	12.2	12.4	12.9
Crude protein	295.0	268.0	237.0	202.0	175.0
Lys	17.4	17.7	17.7	170.5	17.1
Met	5.6	5.9	5.9	5.7	5.8
Crude fat	77.5	73.5	74.7	77.3	75.6
Ca	16.6	15.3	12.5	11.5	13.6
P	4.8	4.8	4.9	4.7	4.3

<sup>a</sup>Air-dry basis; <sup>b</sup>Distillers dried grains with soluble; <sup>c</sup>The premix provides following for per kg diet: Fe (as ferrous sulfate) 16 mg. Cu (as Copper sulfate) 4.0 mg. Zn (as Zinc sulfate) 10 mg. VA 300 IU. VB<sub>1</sub> 0.15 mg. VB<sub>2</sub> 0.40 mg. VB<sub>6</sub> 0.30 mg. folic acid 0.30 mg. nicotinic acid 1.60 mg. D-pantothenic acid 1.3 mg; <sup>d</sup>ME is calculated value, other nutrient levels are measured values. ME calculated according to Hansen *et al.* (1991), digestibility from Dahlman *et al.* (2002)

Table 2: Effects of low protein, Lys and Met supplemented diets on performance of growing raccoon dogs

Items	P30	P27	P24	P21	P18
Initial weights (g)	2.75 $\pm$ 0.220	2.79 $\pm$ 0.370	2.77 $\pm$ 0.170	2.68 $\pm$ 0.240	2.68 $\pm$ 0.230
Final weights (kg)	5.32 $\pm$ 0.540 <sup>a</sup>	5.33 $\pm$ 0.700 <sup>a</sup>	5.32 $\pm$ 0.530 <sup>a</sup>	4.80 $\pm$ 0.490 <sup>b</sup>	4.55 $\pm$ 0.580 <sup>c</sup>
ADFI (g)	199.58 $\pm$ 12.31 <sup>a</sup>	194.50 $\pm$ 11.65 <sup>a</sup>	191.46 $\pm$ 10.56 <sup>a</sup>	156.25 $\pm$ 13.14 <sup>b</sup>	117.50 $\pm$ 12.57 <sup>c</sup>
ADG (g)	65.01 $\pm$ 11.07 <sup>a</sup>	63.49 $\pm$ 9.490 <sup>a</sup>	63.51 $\pm$ 15.89 <sup>ab</sup>	54.08 $\pm$ 7.600 <sup>b</sup>	46.83 $\pm$ 14.61 <sup>b</sup>

Table 3: Effects of low protein, Lys and Met supplemented diets on fat absorptivity of growing raccoon dogs

Items	P30	P27	P24	P21	P18
Digestibility of DM (g kg <sup>-1</sup> )	64.31 $\pm$ 6.02 <sup>b</sup>	62.00 $\pm$ 5.28 <sup>b</sup>	61.33 $\pm$ 4.99 <sup>b</sup>	71.51 $\pm$ 3.67 <sup>a</sup>	73.61 $\pm$ 2.85 <sup>a</sup>
Digestibility of N (g kg <sup>-1</sup> )	69.11 $\pm$ 5.13 <sup>b</sup>	65.42 $\pm$ 6.01 <sup>b</sup>	63.54 $\pm$ 7.12 <sup>b</sup>	65.12 $\pm$ 5.47 <sup>b</sup>	78.14 $\pm$ 9.15 <sup>a</sup>
Digestibility of fat (%)	72.05 $\pm$ 9.52 <sup>a</sup>	64.52 $\pm$ 8.25 <sup>ab</sup>	59.77 $\pm$ 7.59 <sup>b</sup>	65.26 $\pm$ 4.52 <sup>ab</sup>	64.22 $\pm$ 7.37 <sup>ab</sup>

The values are the mean $\pm$ SD; Values within rows with different letters differ significantly ( $p < 0.05$ )

Table 4: Effects of low protein, Lys and Met supplemented diets on Nitrogen (N) balance of raccoon dogs (%)

Items	P30	P27	P24	P21	P18
N intake (g day <sup>-1</sup> )	9.42±0.66 <sup>a</sup>	8.38±0.58 <sup>b</sup>	7.26±0.55 <sup>c</sup>	5.05±0.60 <sup>d</sup>	3.29±0.53 <sup>e</sup>
FN (g day <sup>-1</sup> )	2.91±0.36 <sup>a</sup>	2.85±0.34 <sup>a</sup>	2.65±0.26 <sup>a</sup>	1.79±0.31 <sup>b</sup>	0.66±0.36 <sup>b</sup>
UN (g day <sup>-1</sup> )	3.94±0.83 <sup>a</sup>	2.73±0.69 <sup>b</sup>	1.75±0.67 <sup>c</sup>	1.30±0.35 <sup>cd</sup>	0.84±0.14 <sup>d</sup>
N retention (g day <sup>-1</sup> )	2.58±0.68 <sup>a</sup>	2.51±0.49 <sup>a</sup>	2.47±0.63 <sup>a</sup>	2.05±0.58 <sup>ab</sup>	1.79±0.25 <sup>b</sup>

The values are the mean±SD; Values within rows with different letters differ significantly (p<0.05)

dietary protein level declined but when the dietary protein level decreased to 24% as the dietary protein continued decreased the digestibility of DM, N and fat increased. No significant differences of the digestibility of DM, N and fat among P30, P27 and P24 (p>0.05).

**N-balance:** Effects of low protein, Lys and Met supplemented diets on N balance of raccoon dogs were shown in Table 4. The results showed that daily N intake of P30 was significantly higher than the low protein groups (p<0.05) and as dietary protein reduced daily N intake of raccoon dogs decreased significantly. The datas on FN and UN revealed that as the decreased of dietary protein while the nitrogen emissions from FN and UN reduced significantly and the datas in P30 were significantly higher than others (p<0.05). Growth of the animals was related to N retention. The N retention of P21 and P18 was significantly poorer than other groups (p<0.05) and no significant differences while the dietary protein reduced from 300-240 [g kg<sup>-1</sup> DM] with Met and Lys addition (p>0.05).

## DISCUSSION

Feed intake which affects the growth and performance of animals, it can be influenced by many factors including the number of calories, palatability of feeds, the balance of amino acids and other nutrients and feed digestibility. In the study, feed intake significantly decreased with reducing dietary protein with Met and Lys addition, it may have been related to the increasing dietary content of extruded corn with decreasing protein level. The results showed that when the dietary protein level decreased from 30-24% with Met and Lys supplementation, the diets would not affect the growth performances of raccoon dogs in these groups.

The results confirm observations made in recent and earlier studies on pigs and poultry that free amino acids are used as efficiently as protein-bound ones and that it is therefore, possible to obtain the same performance with low protein diets supplemented with amino acids as with high protein controls (Deschepper and de Groote, 1995; Gruber *et al.*, 2000; Roth *et al.*, 2001). As other trails reported supplementing low-protein diets with amino acids may develop the growth performance of animals (Quiniou *et al.*, 1994; Kerr *et al.*, 1995). A large number of factors can influence the digestibility of

protein, fat and other nutrients. As early study showed that the lower the dietary protein level, the lower was the digestibility of the DM, CP and other nutrients in fox (Dahlman *et al.*, 2002). The influence of protein level on the apparent digestibility of protein has been under intensive research in other animals, pigs in particular. A number of studies suggest that reductions in dietary protein lead to increases in relative amount of endogenous N secretion which in turn, reduce the apparent digestibility of protein (Low, 1980; Baker, 1989). In contrast in studies by Li *et al.* (1993), the apparent digestibility of protein was not affected by the dietary level of protein. In this study, the digestibility of DM, N and fat were elevatory at begaining and degressive later with the dietary protein level decreased and with Met and Lys supplementation. That might because when the dietary protein level reduced to 21%, the dietary protein was limited in order to meet the growth requirement of animals, animals accommodated the nutrients digestibility by themselves. A reduction in the dietary protein content was found to reduce pancreatic secretions of trypsin, chymotrypsin, amylase and lipase in pigs (De Lange *et al.*, 1989). Although not reported, the reason for this may have been a reduction in other amino acids content. Dahlman (2003) pointed that methionine supplementation in low-protein diets significantly improved the digestibility of EE in blue fox.

From an environmental point of view, a substantial reduction in N emissions can be achieved by decreasing the dietary protein level.

According to the present research, N excretion in urine declines significantly when the protein level in the diet is lowered from P30-24 and did not affected the performance of minks. The results are consistent with earlier findings that N excretion declined noticeably along with a reduction in dietary protein pigs (Jongbloed and Lenis, 1992; Valaja *et al.*, 1993). According to Valaja *et al.* (1993), a reduction in dietary protein from 180-120 (g kg<sup>-1</sup>) resulted in a 38.3% decrease in urinary N excretion in growing pigs. The research shows that when the dietary protein level decreased from P30-24 resulted in a 35% decrease in UN and FN excretion in growing raccoon dogs.

As early study showed that the lower the protein level in the diet the better is the utilisation of N and the smaller the proportion excreted (Dahlman, 2003).

## CONCLUSION

Sufficient dietary protein and amino acids are of great importance for the growth of young raccoon dogs but excessive N in diet will increase the production costs and moreover, to an increment in Nitrogen (N) emissions. According to the results while Lys and Met were added in diets and protein level reduced to 24% of DM, there were no significant effects on the performance of growing raccoon dogs compared with high protein level group. Low-protein diets also reduced the pollution of N and save the feed protein resource.

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