Effects of Dietary Crude Protein on Growth Performance, Nutrient Utilization, Immunity Index and Protease Activity in Weaner to 2 Month-old New Zealand Rabbits*

Q. X. Lei, F. C. Li[1,] and H. C. Jiao[1]**

Poultry Institute, Shandong Academy of Agricultural Science, Jinan, Shandong Province, 250100, P. R. China

ABSTRACT : An experiment was conducted to determine the effects of different dietary crude protein (CP) levels on growth performance, nutrient utilization, small intestine protease activity and immunity index of weaner to 2 month-old New Zealand rabbits. Eighty weaner rabbits were allocated in individual cages to five treatments in which they were fed diets with CP at 14%, 16%, 18%, 20% and 22%, respectively. The growth performance and nutrient digestibility of rabbits increased firstly when dietary CP increased, then decreased. The average daily gain was the highest and feed conversion rate was the lowest when dietary CP reached 20%, namely 34.9 g/d and 2.74:1, respectively. Maximum CP digestibility was 72.1% in the 18% CP group, maximum crude fiber digestibility of 28.4% occurred in the 16% CP group and was significantly different from other treatments ($p<0.01$), apparent digestibility of Lys and Val followed the same trend as CP digestibility, and reached their maximum when dietary CP was 18%. Apparent digestibility of Cys, Tyr, Leu and Thr also had a similar trend to CP digestibility. Nitrogen retention (RN) increased with CP level ($p>0.05$), and was highest for 20% CP treatment (1.5 g/d). The effect of CP level on the rate of digestible nitrogen (DN) converted RN was small. The spleen index, thymus index, chymotrypsin and trypsin activities in small intestine were highest when dietary CP was 16%, which were 1.0, 2.8, 15.7 U/g and 125.7 U/g, respectively. There was no significant difference among treatments ($p>0.05$). According to the above results, the appropriate dietary CP level from weaner to 2 month-old meat rabbits was 18-20%. *(Asian-Aust. J. Anim. Sci. 2004. Vol 17, No. 10 : 1447-1451)*

Key Words : New Zealand Rabbit, Crude Protein, Growth, Immunity Index, Protease Activities

INTRODUCTION

Protein is an important component for life processes, and is a material for renewing and repairing tissue. Studies on dietary protein were popular in the in the 1970s and 1980s, but seldom dealt with meat rabbits. It is believed that the dietary crude protein requirement of growing rabbits is 16%, which comes from NRC (1977). Omole (1982) reported the appropriate crude protein level for meat rabbits was 18%-22%. Abdella (1988) indicated that body gain, protein digestibility, slaughter performance and economy benefits were the greatest when the dietary crude protein level was 16%. Wang (1991) pointed out that the average daily gain, feed conversion rate and slaughter rate were the best when the dietary crude protein level was 15.3%-17.9%. Wang (1999) reported that the body gain, feed consumption and slaughter performance were ideal when the dietary crude protein level was only 16.5%.

The data obtained by Moughan et al. (1988) allow estimation of the amino acid requirements of growing rabbits according to their respective proportions of body protein. Several authors have studied the total amino acid requirements for rabbits on a dose-response basis (de Blas, 1998). Recent studies (Taboada et al., 1994,1996; de Blas et al., 1996) have determined the lysine, sulphur and threonine requirements. Results for growth were consistent with those obtained by Moughan et al. (1988) based on amino acid composition of the whole body. But, information regarding the requirement of amino acids is scarce.

The objectives of the research were to examine the effects of different dietary CP levels on growth performance, nutrient utilization, small intestine protease activity and immunity index of weaner rabbits, and to determine the appropriate dietary CP level in weaner to 2 month-old meat rabbits.

MATERIALS AND METHODS

Animals and diets

Eighty weaned New Zealand rabbits (mean body weight 1.13±0.47 kg, equal numbers of male and female) were randomly allocated to five groups according to average body weight, with sixteen rabbits per group. Rabbits were individually housed in metabolism cages which can separate urine from faeces. Each cage contained a feeder to provide free access to feed and a nipple waterer to provide free access to water.

Table 1 gives the composition of diets. The diets were designed according to NRC (1977) and were pelleted.

* Supported by Department of Science and Technology of Shandong Province.

** Corresponding Author: F. C. Li. Tel: +86-0538-8242593-8319, Fax: +86-0538-8241419, E-mail: chlf@sdau.edu.cn

[1] College of Animal Science and Technology, Shandong Agricultural University, Tai,an 271018, P. R. China.

Received December 19, 2003; Accepted May 31, 2004

Table 1. The composition of experimental diets

Composition (%)	Treatment 1	Treatment 2	Treatment 3	Treatment 4	Treatment 5
Peanut vine	33	32	31	30.5	30.5
Corn	29	24	18.5	13	8
Bran	24.5	24.5	25	25	24
Soya bean meal	6	12.5	18	24	30
Limestone	4.5	4	4.5	4.5	4.5
Salt	0.5	0.5	0.5	0.5	0.5
$CaHPO_4$	1.5	1.5	1.5	1.5	1.5
Premix	1	1	1	1	1
Designed DE (MJ/kg)	11.22	11.22	11.22	11.22	11.22
Designed CP	14	16	18	20	22
Determined CP	13.50	15.55	17.56	20.20	21.93
CF	12.21	12.17	12.11	12.18	12.35
Phe	0.50	0.62	0.72	0.83	0.94
His	0.27	0.33	0.38	0.43	0.48
Met	0.12	0.16	0.18	0.21	0.24
Cys	0.24	0.28	0.30	0.33	0.37
Arg	0.63	0.82	0.97	1.14	1.31
Lys	0.51	0.67	0.80	0.94	1.09
Tyr	0.33	0.42	0.48	0.56	0.64
Leu	0.82	0.98	1.11	1.25	1.39
Thr	0.41	0.52	0.61	0.71	0.80
Val	0.50	0.61	0.70	0.80	0.89
Ile	0.40	0.50	0.59	0.68	0.77

Table 2. Effects of dietary CP levels on growth performance of experimental rabbits (mean±SD)

Treatment	Initial body weight (g)	2 month body weight (g)	ADG (g/d)	Average daily intake (g/d)	F/G ratio
14% group	1,114.6±76.4	1,448.4±50.5	32.3±1.9	98±6	3.10^{a}
16% group	1,080.8±18.7	1,403.9±38.0	33.4±2.1	100±8	2.93^{b}
18% group	1,146.9±30.3	1,495.5±42.8	34.9±1.4	99±4	2.83^{b}
20% group	1,160.0±59.3	1,509.3±69.1	34.9±1.5	96±3	2.74^{b}
22% group	1,133.9±48.1	1,434.1±30.5	32.1±2.1	90±7	2.82^{b}

The data with different letters in the same column (row) differ significantly ($p<0.05$ or $p<0.01$).

Eighty weaned rabbits were fed each of five experimental diets. The CP levels of the five experimental diets were 14%, 16%, 18%, 20% and 22%, respectively.

Experimental procedures

All rabbits were divided into five groups according to average body weight. Feed was offered in equal portions at 8:30 and 17:30 daily. Total experiment consisted of a 7-day adjustment period followed by an 18 day experimental period (including a 7 day collection of faeces, urine and residual feed daily). Faeces, urine and residual feed were collected from rabbits in the metabolism cages. After the feeding trial, 10 ml blood samples were collected from each rabbit and 6 rabbits from each experimental group were slaughtered. The serum was separated by centifugation for chemical analysis.

Chemical analysis and statistics

All experimental rabbits were weighed from weaning to 2 months old, and the average daily gain was calculated. The average daily feed consumption was recorded and feed/gain ratio was calculated. Feed and faeces samples were analyzed for dry matter (DM), crude fiber (CF), nitrogen (N), crude fat and energy according to the procedure of Yang (1993). Urine samples were analyzed for nitrogen (N) according to the procedure of Yang (1993).

Spleen and thymus weight was measured after slaughter and spleen index and thymus index were calculated according to the following formulae: spleen index=spleen weight/body weight, thumus index=thymus weight/body weight.

The small intestine chyme was collected after slaughter, chymotrypsin and trypsin activities were analyzed according to the method of Xu (2002).

Data collected from the experiments were subjected to analysis of variance using the General Linear Model Procedure of SAS (1985). Treatment means comparison was done using Duncan's Multiple Range Test.

RESULTS AND DISCUSSION

Effects of different dietary CP on growth performance

Effect of different dietary CP on growth performance of experimental rabbits is shown in Table 2. The average daily gain (ADG) of 16% CP group, 18% CP group and 20% CP

Table 3. Effects of CP levels on nutrient apparent digestion of experimental rabbits (%) (mean±SD)

Item	14% CP group	16% CP group	18% CP group	20% CP group	22% CP group
Energy	61.5±4.0^{AB}	63.2±0.8^{B}	64.7±1.1^{B}	64.7±1.6^{B}	59.3±1.4^{A}
CP	64.9±7.0^{a}	68.1±3.8^{ab}	72.1±6.0^{b}	69.5±3.6^{ab}	69.4±6.1^{ab}
CF	17.0±7.8^{A}	28.4±1.8^{B}	20.9±4.9^{A}	20.9±2.9^{A}	18.2±7.0^{A}
DM	58.5±4.5	57.5±4.5	60.8±3.0	60.4±1.3	57.8±4.7
OM	64.2±4.3^{ab}	63.5±4.4^{ab}	66.8±2.5^{b}	66.5±1.2^{b}	61.0±4.2^{a}
EE	66.1±15.3^{ab}	72.6±8.1^{b}	73.6±7.1^{b}	73.1±8.2^{b}	60.1±9.8^{a}

Table 4. Effects of different CP levels on amino acid apparent digestibility (%) (mean±SD)

Traits	14% CP group	16% CP group	18% CP group	20% CP group	22% CP group
Phe	75.6±4.6	73.6±2.0	80.7±4.3	80.2±6.3	74.0±3.8
Ala	81.7±4.2^{C}	71.5±4.5^{AB}	68.0±2.4^{AB}	67.5±4.1^{A}	76.3±4.0^{BC}
Met	65.3±4.3^{AB}	63.4±2.7^{A}	71.1±2.7^{B}	71.8±4.1^{B}	72.0±2.7^{B}
Cys	58.9±3.0^{A}	67.3±4.5^{AB}	74.1±4.2^{BC}	77.6±3.8^{C}	65.4±4.7^{AB}
Glu	74.8±4.4^{a}	82.8±3.6^{ab}	84.0±5.1^{b}	76.1±7.1^{ab}	81.7±4.2^{ab}
Gly	64.9±3.9	68.8±5.1	65.8±6.2	66.5±3.1	71.6±2.8
Arg	74.9±4.8^{a}	69.8±3.9^{a}	87.5±2.5^{b}	85.6±3.8^{b}	82.8±2.8^{b}
Lys	62.3±3.9^{a}	75.9±3.7^{b}	82.7±3.0^{c}	75.5±5.1^{b}	67.1±3.7^{a}
Tyr	70.1±6.6^{a}	72.7±6.1^{ab}	80.1±5.1^{b}	80.2±6.3^{b}	76.4±4.1^{ab}
Leu	78.0±3.8	79.1±5.0	79.9±6.2	81.6±3.1	79.4±5.6
Asp	70.8±4.5^{a}	77.9±3.8^{b}	75.6±5.1^{ab}	79.7±3.5^{b}	69.1±5.1^{a}
Ser	72.3±3.3^{ab}	80.2±3.0^{b}	77.2±4.1^{bc}	69.8±4.1^{a}	70.1±3.4^{a}
Thr	61.6±5.5^{AB}	68.7±4.4^{B}	70.7±3.3^{B}	70.8±3.2^{B}	56.5±4.0^{A}
Val	63.4±4.1^{a}	68.9±4.0^{ab}	71.3±3.5^{b}	70.9±4.3^{b}	67.0±3.4^{ab}
Ile	75.9±3.9	73.5±4.7	80.5±4.7	79.6±4.8	76.0±4.1
His	74.8±6.1^{A}	80.6±4.6^{AB}	87.3±5.3^{B}	82.8±4.2^{AB}	85.6±3.8^{B}

group were higher than that of the 14% CP group. Among them, the ADG of the 20% CP group was the highest but there was no significant difference in ADG ($p>0.05$). The feed/gain (F/G) ratio of 14% CP group, 16% CP group, 18% CP group and 22% CP group were higher than that of 20% CP group and there was a significant difference between 14% CP group and other groups ($p<0.05$). The regression formula of ADG (y, g/d) and dietary CP levels (x, %) was $y=-13.24+5.37x-0.15x^2$, $R^2=0.8997$.

Many researchers have studied the effect of dietary protein levels on production performance of growing rabbits. Although protein levels were different, all researchers thought that low dietary protein levels can reduce production performance and high dietary protein levels can increase economic profit, gain, slaughter performance and nutrient digestibility. In this study, ADG rose from 14% CP group to 20% CP group, then fell for the 22% CP group F/G ratio descended then ascended when CP increased. Tang (1987) reported that ADG and F/G ratio were 19.4 g/d and 3.8 when dietary CP was 19.5%, which were higher and lower than those of 16.7% and 12% CP groups. Wang (1999) reported that ADG was 29 g/d when dietary CP was 16.5%, which was higher than those of 13.5%, 15%, 18%, 19.5% and 21% CP groups. F/G ratio was 3.5 when dietary CP was 18%, which was lower than those of 13.5%, 15%, 16.5%, 19.5% and 21%CP groups. Xu (1982) reported that many traits were the best for 6 to 14 week-old rabbits when dietary protein was 17%. ADG and microorganism concentration in intestine decreased in 22% protein group. Similar finding were reported by Wang (1991).

Effects of different dietary CP on nutrient apparent digestibility (Table 3)

Nutrient apparent digestibility was the highest with 18% CP group apart from crude fiber (CF) digestibility, and was significantly different from other groups ($p<0.05$). Lang (1981) reported that the feed conversion ratios for dietary 17%-20% protein were higher than that for 16%. Tang (1987) also reported the same results. The apparent digestibility of CF was the highest when dietary CP was 16%, and the 16% CP group was significantly different from other groups ($p<0.05$). The regression formula of CP digestion (y, %) and dietary CP levels (x, %) was $y=-0.24x^2+9.01x-15.05$, $R^2=0.8477$. The regression formula of CF digestion (y, %) and dietary CP levels (x, %) was $y=-0.37x^2+13.05x-91.14$, $R^2=0.4226$.

Similar findings were reporteds by Cheeke (1981), de Blas (1985) and Xie (1990). Xie (1990) pointed out that CP and CF digestion rose by 2% and 3.9% when dietary CP increased from 13% to 16%.

The effect of different dietary CP on apparent digestibility of amino acids of experimental rabbits is shown in Table 4. Apparent digestibility of Lys and Val followed the same trend as CP digestibility, and reached

Table 5. Effect of different protein levels on nitrogen balance (mean±SD)

Traits	14% CP group	16% CP group	18% CP group	20% CP group	22% CP group
N intake (g/d)	2.4 ± 0.3^{a}	2.9 ± 0.3^{ab}	3.1 ± 0.6^{b}	3.8 ± 0.2^{c}	3.5 ± 0.8^{c}
Faeces N (g/d)	0.8 ± 0.2^{a}	0.9 ± 0.1^{ab}	0.8 ± 0.2^{a}	1.2 ± 0.2^{c}	1.1 ± 0.2^{cb}
Urine N (g/d)	0.6 ± 0.2^{a}	0.7 ± 0.2^{a}	1.0 ± 0.2^{b}	1.1 ± 0.3^{b}	1.0 ± 0.2^{b}
Digestible N (DN, g/d)	1.6 ± 0.3^{a}	2.0 ± 0.3^{ab}	2.1 ± 0.5^{bc}	2.6 ± 0.2^{cd}	2.4 ± 0.7^{d}
Retention N (RN, g/d)	1.0±0.2	1.3±0.3	1.2±0.4	1.5±0.3	1.4±0.8
RN/N intake (%)	41.7 ± 6.7^{a}	44.8 ± 10.0^{b}	39.7 ± 6.7^{a}	39.5 ± 12.7^{a}	40.0 ± 10.0^{a}
RN/DN (%)	65.0 ± 7.8^{b}	66.8 ± 8.3^{b}	57.8 ± 10.6^{a}	56.8 ± 12.0^{a}	52.6 ± 25.8^{a}

Table 6. Effect of different protein levels on immunity index of experimental rabbits (mean±SD)

Traits	14% CP group	16% CP group	18% CP group	20% CP group	22% CP group
Spleen weight (g)	0.7±0.1	1.1±0.6	1.2±0.5	1.0±0.2	1.2±1.0
Thymus weight (g)	2.9±0.4	3.6±1.0	2.8±0.9	3.6±0.8	2.9±1.2
Spleen index	0.5±0.2	1.0±0.9	0.9±0.4	0.7±0.2	0.9±0.6
Thymus index	2.1±0.2	2.8±0.8	2.0±0.5	2.5±0.5	2.2±0.8

Table 7. Effect of different protein level on enzyme activities in small intestine of rabbits (U/g) (mean±SD)

Traits	14% CP group	16% CP group	18% CP group	20% CP group	22% CP group
Chymotrypsin	14.4±0.7	15.7±3.2	13.8±0.5	13.8±0.6	13.8±0.5
Trypsin	125.1±5.8	125.7±3.6	115.0±10.1	122.6±9.1	119.8±6.3

their maximum when dietary CP was 18%. Apparent digestibility of Cys, Tyr, Leu and Thr also had a similar trend to CP digestibility. This is consistent with the results of De Blas (1998), who pointed out that the digestibility of total protein had a positive correlation with the digestibility of single amino acids. The regression formula of digestion of Cys (y 1, %), Lys (y 2, %), Tyr (y 3, %), Leu (y 4, %), Thr (y 5, %) and Val (y 6, %) and dietary CP levels (x, %) were y 1=-0.79x^2+29.73x-203.07, R^2=0.9015; y 2=-1.04x^2+37.77 x-263.18, R^2=0.9648; y 3=-0.36x^2+13.89x-55.27, R^2= 0.8623; y 4=-0.10x^2+3.95x-42.39, R^2=0.7633; y 5=-0.80x^2+ 28.26x-178.62, R^2=0.9238; y 1=-0.39x^2+14.50x -63.20, R^2= 0.9991.

When amino acids were provided by conventional feeds the values obtained for lysine (Taboada et al., 1994), methionine (Taboada et al., 1996) and threonine (de Blas et al., 1996) were 0.74, 0.71 and 0.63, respectively, similar to results obtained in the present experiment.

Effects of different dietary CP on nitrogen balance (Table 5)

Effects of dietary CP on N balance of experimental rabbits are shown in Table 6. N intake, faces N, urine N, digestible N (DN) and retained N (RN) of different treatments increased with increasing dietary CP levels. RN/N intake and the efficiency of DN converted into RN were the highest when dietary CP was 16%, and were 44.8% and 66.8%, and had significantly different among different treatments. The regression formula of RN (y, g/d) and dietary CP levels (x, %) was y=-0.76+0.19x-0.004x^2, R^2=0.8312. In this study, Urinary N losses increased from 14% CP group to 20% CP group, and fell for the 22% CP group. Urinary N losses of 18% CP group, 20% CP group and 22% CP group were higher than those of 14% and 16% CP group ($p<0.05$). There were no significant differences among 18%, 20% and 22% CP group and between 14% and 16% CP group ($p>0.05$). The regression formula of urinary N losses (y, g/d) and dietary CP levels (x, %) was y=-0.01 x^2+0.45x-3.59, R^2=0.9027.

This research illustrated that RN increased when dietary CP increased, that is to say that the synthesis of body protein and product protein may increase with increasing dietary CP, consistent with results of Singh et al. (1988).

Effects of different dietary CP on immunity indexes

Effects of dietary CP on immunity indexes of experimental rabbits are shown in Table 6. Spleen weight, thymus weight, spleen index and thymus index were not significantly different among different groups, but spleen index and thymus index were the highest when dietary CP was 16%.

Protein and amino acids are the basic structural units of the body's immune system, and have a close relationship with the growth and development organs of the immune system. Immunity index reflects immunity function of animals in certain context. For a healthy animal, immunity function is better when immunity index is bigger. This research illustrated that spleen and thymus index of 16% CP group were the greatest and higher than that of 14% group, which indicated immunity function of weaner to 2 month-old rabbits will drop if dietary CP level is lower than 16% and may affect rabbits' immune responses.

Effects of different dietary CP on small intestine protease

Effects of different dietary CP on the activity of small intestine protease are shown in Table 7. The activity of small intestine chymotrypsin and trypsin were the highest when dietary CP was 16%, but there were no significant differences among different treatments.

Chymotrypsin and trypsin are two important enzymes in digesting and breaking down protein. This research indicated that their activities do not vary with the change of dietary protein and CP digestibility.

CONCLUSIONS

Growth performance of weaner to 2 month-old meat rabbits rose then fell when dietary CP increased from 14% to 22%. ADG was the highest and feed/gain ratio was the lowest when dietary CP was 20%, so growth performance of weaner to 2-old month meat rabbits was the best when dietary CP was 18-20%.

The digestion of CF was highest with 16% CP group. The digestibility of CP, Lys and Val were highest with 18% CP group. Apparent digestibility of Cys, Tyr, Leu, Thr and RN was greatest in the 20% CP group.

Spleen index, thymus index, chymotrypsin activity and trypsin activity of weaner to 2 month meat rabbits were highest when dietary CP was 16%.

REFERENCES

Abdella, H. M. 1988. Effect on growing rabbits of feeding different levels of crude protein. The Journal of Applied Rabbit Research 11:252-256.

Cheeke, P. R. 1981. Effect of alfalfa and dietary fiber on the growth performance of weaning rabbits. Lab. Anim. Sci. 28:167-172.

de Blas, J. C. 1985. Units for feed evaluation and requirements for commercially grown rabbits. J. Anim. Sci. 60:1021-1028.

de Blas, C., E. Taboada, N. Nicodemus, R. Campos and J. Mendez. 1996. The response of highly productive rabbits to dietary threonine content for reproduction and growth. In: (Ed. F. Labas) Proceedings of the 6th World Rabbit Congress, Toulouse. Association Francaise de Cuniculture, Lempdes, pp. 139-144.

de Blas. 1998. The Nutrition of the Rabbit.CABI Publishing. p. 42, p. 246.

Lang, J. 1981. The nutrition of the commercial rabbit. 1. physiology, digestibility and nutrient requirements. Nutr. Abstr. Rev. 51:197.

Moughan, P. J., W. H. Schultze and W. C. Smith. 1988 Amino acid requirements of the growing meat rabbit. 1. The amino acid composition of rabbit whole-body tissue-a theoretical estimate of ideal amino acid balance. Animal Production 47:297-301.

NRC. 1977. Nutient requirements of domestic animals, No. 9. Nutrient requirements of rabbits. Second Revised Ed. National academy of sciences-National Research Council, Washington, DC.

Omole, T. A. 1982. The effect of dietary protein on growth and reproduction performance in rabbits. The Journal of Applied Rabbit Research 5:83.

Singh, B. 1988. Utilization by growing rabbits of a low protein diet with or without urea and groundnut cake supplementation. The Journal of Applied Rabbits Research 11:25-28.

Taboada, E., J. Mendez, G. G. Mateos and C. de Blas. 1994. The response of highly productive rabbits to dietary lysine content. Livestock Production Science 40:329-337.

Taboada, E., J. Mendez and C. de Blas. 1996. The response of highly productive rabbits to dietary sulphur amino acid content for reproduction and growth. Reproduction, Nutrition and Development 36:191-203.

Tang, L. M. 1987. Effects of dietary energy and protein on production performances of growing rabbits. Chinese Journal of Rabbit Farming 6:21-23.

Wang, S. C. 1999. The effect of different nutrient levels on growth performance of meat rabbits. Chinese Journal of Rabbit Farming 6:20-22.

Wang, Y. X. 1991. The study of protein requirement in growing meat rabbits. Journal of Shandong Agricultural University 6:137-145.

Xie, X. H. 1990. Effects of dietary crude protein and fiber on digestibility of meat rabbit. Chinese Journal of Rabbit Farming 3:16-18.

Xu, L. D. 1982. The nutrition and diets. Feed and Animal 3:22-25.

Xu, Z. R., X. T. Zou and C. H. Hu. 2002. Effects of dietary fructooligosaccharide on digestive enzyme activities, intestinal microflora and morphology of growing pigs. Asian-Aust. J. Anim. Sci. 15:1784-1789.

Yang, S. 1993. Feed analysis and feed quality assay. Beijing Agricultural University Press, 1993.