ENERGY AND PROTEIN REQUIREMENTS OF KHAKI CAMPBELL × THAI NATIVE GROWING DUCKS

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Summary

The experiment was carried out to estimate the energy and protein requirements of Khaki Campbell × Thai Native growing ducks. The growing stage was divided into 2: the first was a starting stage from 0 to 4 weeks of age and the second was a growing stage from 4 to 18 weeks of age. In the starting stage, the birds were given diets containing 2.70, 2.75, 2.80, 2.85 and 2.90 Mcal ME/kg and ME/protein ratio of 150/1 for the energy requirement trial, and diets containing 17.0%, 18.5%, 20.0% and 21.5% protein and 2.8 Mcal ME/kg for the protein requirement trial. It was found that there were no significant differences in body weight gain of feed efficiency among the energy or protein levels. In the growing stage, no significant differences in body weight gain or feed efficiency were found among the energy levels from 2.70 to 2.90 Mcal ME, or protein levels from 15.5 % to 20.0%. The age at first lay was 105-117 days and this is earlier than that observed in purebred Khaki Campbell ducks. The feed cost for raising ducks was lower in the low energy and low protein diets. In conclusion, Khaki Campbell × Thai native ducks can be raised with a diet low in ME as 2.7 Mcal/kg during the whole growing period. As for the protein, 17.0% in the starting stage and 15.5% in the growing stage is practical.

(Key Words: Energy/Protein Requirement, Growing Duck, Khaki Campbell × Thai Native)

Introduction

In Thailand, Khaki Campbell × Thai Native duck is most commonly used for egg production, however, little information is available on its nutrient requirements not only for egg production but also for growth. Siregar and Farrell (1980) noted that the nutrient requirements of ducks are mainly based on those of chickens, but the nutrient requirements of the two species must differ due to differences in growth rate, body composition, digestive physiology and starvation heat production. Ducks have no crop and their proventriculus is not fusiform but cylindrical (Das et al., 1965). There is an evidence that energy metabolism is different between both species (Mohamed et al., 1984).

Pan et al. (1978) found that a 15% protein diet could be used for 0 to 4-week old Tsaiya ducks without any ill-effects. Reddy et al. (1980) reported that 23% protein was required for the maximum growth of Khaki Campbell ducks up to 4 weeks of age. Prasad et al. (1988) and Gowd et al. (1983) concluded that starting and growing Khaki Campbell ducks required 16% dietary protein. There have been several studies on the protein requirements of White Pekin ducks (Dean et al., 1965; Du Preez and Wessels, 1970; Chin and Hutagalung, 1984).

Since the extent of nutrient requirements varies with age, sex, species and probably with strains (Bodwell et al., 1980; Bondi, 1987), a series of experiments have been carried out to investigate the nutrient requirements of Khaki Campbell × Thai Native ducks. In this paper energy and protein requirements of growing ducks from 0 to 18 weeks of age are reported.

Materials and Methods

The growing stage was divided into 2; the first was a starting stage from 0 to 4 weeks of age and the second a growing stage from 4 to 18 weeks of age. The experiment consisted of

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4 trials; energy requirements for the starting and growing stages, and protein requirements for both stages.

Day-old female ducks were introduced from a commercial hatchery and a certain number of birds were selected based on body weight and health condition. In the trials for growing stage a certain number of ducks were selected at 4 weeks of age from the flock which had been raised with a ration commonly used in the MIAT farm. In the trials for starting and growing stages, 15 and 12 birds, respectively, were confined together in a bamboo slat floor pen of 1.20 × 1.85 m^2 and fed diets as moist mash *ad libitum* under the practical environmental conditions. During the 18-week experimental period, body weight was weekly measured by group weighing and at the same time feed intake was determined. The age was recorded when the first egg was found in the group. In the trial for protein requirement, the hemoglobin and hematocrit values were determined at 4, 8, 12 and 16 weeks of age by the methods of Wong (1928) and Bush (1975), respectively.

Experimental diets were formulated using ingredients commonly used in Thailand. In the trials for energy requirement, 5 diets having 2.70, 2.75, 2.80, 2.85 and 2.90 Mcal ME/kg diet were used. All diets were formulated to be equal in ME/CP ratio (150/1 for the starting stage and 180/1 for the growing stage). In the trials for protein requirement, 4 diets having 17.0%, 18.5 %, 20.0% and 21.5% of protein for the starting stage and 15.5%, 17.0%, 18.5% and 20.0% of protein for the growing stage were used. ME content was fixed to be 2.8 Mcal/kg. Calcium and phosphorus contents were 0.8% and 0.6%, respectively, in all the diets. The diets were formulated using linear programming, and the least cost was estimated using retail prices. The ingredient compositions of the experimental diets are indicated in table 1.

In all trials each dietary treatment comprised 3 replicates, and the results obtained were subjected to an analysis of variance for a completely randomized design (Steel and Torrie, 1980). Duncan's new multiple range test was employed to determine which means were significantly different (Duncan, 1955). All statements of significance were based on 0.05 level of probability.

TABLE 1. INGREDIENT COMPOSITION OF THE EXPERIMENTAL DIETS

(1) For energy requirement[1]

ME level (Mcal/kg)	2.70	2.75	2.80	2.85	2.90
Starting					
Broken rice	64.20	66.00	63.50	65.30	65.50
Rice bran	10.00	10.00	10.00	6.08	3.20
Soybean meal (44%)	10.60	11.75	12.75	14.00	15.25
Fish meal (55%)	10.60	10.80	11.00	11.20	11.40
Ipil-ipil meal	3.20	–	–	–	–
Ground shell	0.27	0.40	0.45	0.03	–
Bone meal	0.44	0.40	0.30	0.74	1.00
Tallow	–	–	1.35	2.00	3.00
Price (Baht/kg)	6.33	6.46	6.62	6.79	6.97
Growing					
Broken rice	68.97	70.70	73.17	70.75	68.45
Rice bran	11.59	11.30	10.00	10.00	9.96
Rough rice bran	1.55	–		–	–
Soybean meal (44%)	4.55	4.98	6.07	7.00	7.86
Fish meal (55%)	9.06	9.22	9.39	9.56	9.73
Ipil-ipil meal	3.00	2.52	–	–	–
Ground shell	0.63	0.63	0.59	0.60	0.61
Bone meal	–	–	0.10	0.05	–
Tallow	–	–	0.03	1.39	2.74
Price (Baht/kg)	5.55	5.66	5.78	5.94	6.09

(2) For protein requirement[2]

Protein level (%)	15.5	17.0	18.5	20.0	21.5
Starting					
Ground corn	–	32.00	32.60	29.90	30.12
Broken rice	–	32.32	30.00	31.56	30.00
Rice bran	–	11.53	9.22	7.00	4.58
Soybean meal (44%)	–	7.69	11.05	15.00	18.50
Fish meal (55%)	–	10.39	11.20	12.10	13.00
Ipil-ipil meal	–	5.00	5.00	3.57	3.04
Ground shell	–	0.42	0.28	0.22	0.11
Price (Baht/kg)	–	4.14	4.38	4.66	4.91
Growing					
Ground corn	20.00	20.00	20.00	20.00	–
Broken rice	50.28	49.11	47.07	45.47	–
Rice bran	8.41	5.45	3.54	1.15	–
Rough rice bran	2.20	1.59	2.68	2.67	–
Soybean meal (44%)	7.50	10.05	14.53	17.16	–
Fish meal (55%)	9.39	10.30	11.20	12.12	–
Ipil-ipil meal	1.00	2.50	–	–	–
Ground shell	0.57	0.25	0.33	0.18	–
Bone meal	–	0.10	–	–	–
Price (Baht/kg)	4.01	4.27	4.50	4.75	–

All diets contained 0.4% common salt and 0.25% premix which supplies (per kg diet) Vitamin A 9000 IU, Vitamin D_3 1600 ICU, Vitamin E 1.4 mg, Vitamin K 1.2 mg, Vitamin B_1 0.4 mg, Vitamin B_2 4 mg, Vitamin B_6 3 mg, Vitamin B_{12} 0.015 mg, Ca pantothenate 7 mg, niacin 40 mg, folic acid 0.4 mg, biotin 0.04 mg, Co 1 mg, Cu 8 mg, I 1 mg, Mn 25 mg, Se 0.1 mg, Zn 50 mg, Fe 50 mg, ethoxyquin 4 mg.

[1] ME/CP ratio: 150/1 for starting and 180/1 for growing stages.

[2] ME content: 2.8 Mcal/kg diet for both stages.

Results and Discussion

The effects of dietary energy level on body weight gain, feed intake and feed conversion ratio in the starting and growing stages are presented in table 2. In the starting 4-week period, the highest body weight gain was observed in 2.75 Mcal ME/kg diet, being 635 g, and the lowest

TABLE 2. THE EFFECTS OF DIETARY ENERGY LEVEL ON BODY WEIGHT GAIN, FEED INTAKE AND THE FEED CONVERSION RATIO (FCR) FOR STARTING (0-4 WEEKS) AND GROWING (4-18 WEEKS) DUCKS

Energy level (Mcal/kg)	2.70	2.75	2.80	2.85	2.90	SEM
Starting (0-4 weeks)[1]						
Weight gain (g/period)	628	635	628	608	590	11
Feed intake (kg/period)	1.68	1.77	1.69	1.64	1.59	0.06
FCR (g diet/g gain)	2.68	2.79	2.70	2.69	2.70	0.04
Growing (4-18 weeks)[2]						
Weight gain (g/period)	782	778	749	786	804	25
Feed intake (kg/period)	11.9	12.0	12.1	12.0	11.9	0.1
FCR (g diet/g gain)	15.2	15.4	16.2	15.3	14.8	0.5

No significant difference ($p > 0.05$) was found among the treatments.

[1] Average initial body weight was 59 g, ME/CP ratio was 150/1.

[2] Average initial body weight was 677 g, ME/CP ratio was 180/1.

in 2.90 Mcal, being 590 g, however, the difference was only 45 g and not significant. This is in accordance with the result of Pan et al. (1978) who reported no significant difference in body weight gain in Tsaya ducks between 11.7 and 10.9 MJ (2.8 and 2.6 Mcal) ME/kg. Reddy et al.(1980) also reported the energy requirement of Khaki Campbell to be 2.80 Mcal/kg diet. Although no significant difference was found, the weight gain tended to decrease from 2.75 Mcal to 2.90 Mcal/kg diet, and this disagrees with the findings of Siregar and Farrell (1980) and Dean (1968), who reported that 3.15 Mcal ME/kg was necessary for maximum performance of starting ducks. Feed intake ranged from 1.59 to 1.77 kg, and no significant difference was found among the dietary treatments. Likely the body weight gain, feed intake tended to decrease as dietary ME level increased from 2.75 to 2.90 Mcal. Reflecting the results of body weight gain and feed intake, the feed conversion ratio was almost identical in all treatments, being around 2.70. In the growing 14-week period, the highest and the lowest feed intake were obtained in 2.90 Mcal ME/kg, and consequently the feed conversion ratio was superior in 2.90 Mcal/kg to the lower energy levels, although there was no significant difference. Dean (1968) found that Pekin ducks given 12.97 MJ (3.1 Mcal) ME/kg exhibited heavier body weight, and Adams and Stadelman (1979) reported that growing female ducks showed poorer feed conversion ratio when 2.97 Mcal/kg was given than when 3.08 3.19 Mcal/kg was given. The result of the present experiment is in disagreement with those of the above mentioned workers, but is in agreement with the result of Gowd et al. (1983), who reported that 2.8 Mcal/kg with 16% protein was optimal for growing Khaki Campbell ducks. It can be seen in the table that in the growing stage the body weight gain was slow and feed intake was very high compared to that in the starting stage. Consequently, the feed conversion ratio was very high compared with that in the starting stage. This result was similar to that obtained by Sahoo et al. (1985), who reported that the most rapid growth rate of female Khaki Campbell ducks was observed up to 4 weeks of age, the lowest from 13 to 16 weeks of age, and the feed conversion ratio increased with an increase in age.

The effects of dietary crude protein level on body weight gain, feed intake and feed conversion ratio are indicated in table 3. In this experiment the diets containing 17.0%, 18.5%, 20.0% and 21.5% crude protein were fed to the starting ducks, and the diets containing 15.5%, 17.0%, 18.5% and 20.0% curde protein were fed to the growing ducks. In the starting 4-week period, the highest body weight gain was observed in the 20.0% protein diet but the difference between the highest and the lowest body weight gains was only 39 g. Although no significant difference was found among the treatments, the body weight gain tended to decrease with a decrease in the

TABLE 3. THE EFFECTS OF DIETARY CRUDE PROTEIN LEVEL ON BODY WEIGHT GAIN, FEED INTAKE AND THE FEED CONVERSION RATIO (FCR) FOR STARTING (0-4 WEEKS) AND GROWING (4-18 WEEKS) DUCKS

CP level (%)	15.5	17.0	18.5	20.0	21.5	SEM
Starting (0-4 weeks)[1]						
Weight gain (g/period)		738	751	777	750	14
Feed intake (kg/period)	–	2.05[a]	1.96[b]	2.08[a]	2.04[a]	0.02
FCR (g diet/g weight gain)	–	2.77	2.61	2.68	2.72	0.07
Growing (4-18 weeks)[2]						
Weight gain (g/period)	650	702	734	649		33
Feed intake (kg/period)	11.1	11.9	11.9	12.5	–	0.4
FCR (g diet/g weight gain)	17.0	16.9	16.2	19.3	–	1.2

All diets were isocaloric (2.8 Mcal ME/kg diet). Means not sharing a common superscript letter are significantly different ($p < 0.05$).

[1,2] Average body weight was 58 g and 746 g, respectively.

protein level. This result is in accord with the results of several workers. Pan et al. (1978) reported that weight gain of Tsaiya from 0 to 4 weeks was not significantly different between 15% and 19% protein diets. Prasad et al. (1988) concluded that starting and growing Khaki Campbell ducks require 16% protein, and Leclercq and De Carville (1976) reported that protein requirement of Muscovy ducks from 0 to 4 weeks of age did not exceed 17%. The optimum protein levels observed in these experiments were lower than 23% which was reported by Reddy et al. (1980) in Khaki Campbell. Dean et al. (1965) found that weight gain of White Pekin from 0 to 1 week, from 1 to 2 weeks and from 2 to 3 weeks of age was maximized at levels of 22%, 20% and 18% protein, respectively. Chin and Hutagalung (1984) reported that 22-24% protein was required by White Pekin from 0 to 6 weeks of age, and Oluyemi and Fetuga (1978) reported that 24% protein was required for maximum growth of 0 to 8-week old Nigerian ducks. Du Preez and Wessels (1970) reported that optimal 14-day live weight of White Pekin was achieved with a diet containing approximately 19% crude protein. Feed intake was also not different among the treatments except for the 18.5% protein diet which showed significantly lower feed intake. The reason why such a significant difference occured was not clear. The feed conversion ratio was not significantly different among the treatments. In the growing stage, the body weight gain was highest in the 18.5% protein diet and it decreased with a decrease in the protein level, although no significant difference was found among the treatments. Feed intake and feed conversion ratio were also not significantly different among the treatments. Pan et al. (1978) reported that there was no significant difference in body weight gain and feed efficiency among growing Tsaiya ducks fed diets of different protein levels. Adams and Stadelman (1979) fed a 18% protein diet and least cost diets containing 15.4-15.9% protein to female ducks from 0 to 48 days of age, and they found no significant difference in body weight gain between the treatments. According to Bandong et al. (1977), growing Philippine ducks required 15-18% protein from 3 to 13 weeks of age and 16% protein from 13 weeks to first lay. Gowd et al. (1983) and Prasad et al. (1988) also reported the protein requirement of 16% for growing Khaki Campbell and Khajarern and Khajarern (1987) recommended 10-15% protein for growing Thai ducks from 3 to 10 weeks of age. The recommendation of Shafiuddin (1985) was 17% protein for growing Bangladesh ducks, and that of Sainsbury (1980) was 15% for growing ducks.

In order to observe the health condition, hematocrit value and hemoglobin content were determined, and the results are shown in table 4. Both values were almost identical not only for different dietary treatments but also for growing stages. Sreeraman et al. (1979) reported that the average hemoglobin content for 1-2 year old female Khaki Campbell was 12.2 g/100 ml, and the present data were a little higher than that value. Since Morgan (1962) and Hamilton and Whitney (1980) indicated that the low he-

TABLE 4. THE EFFECTS OF DIETARY PROTEIN LEVEL ON HEMATOCRIT AND BLOOD HEMOGLOBIN OF GROWING DUCKS

Protein level (%)	15.5	17.0	18.5	20.0
Hematocrit (%)				
4	38.6	40.3	40.1	39.6
8	45.3	43.0	45.6	46.3
12	45.3	44.9	44.7	45.1
16	46.4	44.4	46.2	46.6
Hemoglobin (g/100 ml)				
4	13.3	13.5	14.2	13.8
8	15.2	15.5	15.5	15.1
12	13.8	13.5	12.8	13.1
16	15.3	14.7	14.8	15.6

moglobin value would be shown in a low protein diet, the protein content used in the present experiment seemed to be adequate for growing ducks. Mortality in each trial was low and it seemed not to be affected by the diets used in this experiment. As shown in table 5, the age at first lay was 114-117 days in the trial of energy level and 105-113 days in the trial of protein level. Eswaran et al. (1984) reported that the first lay of Khaki Campbell was 163 days. The crossbred between Khaki Campbell and Thai Native seemed to begin lay earlier than Khaki Campbell pure breed.

As mentioned before, all diets were formulated using a linear programming to get a least cost, and the feed price of each deit was indicated in table 1. Ground corn was used in the trial for protein requirement but not in the trial for energy requirement. It is apparent that the feed price would be lessened by using ground corn. In both trials the feed price increased with an increase either in the energy level or in the protein level. The feed costs for raising ducks are indicated in table 6. In the trial for energy requirement, the lowest cost was shown in the 2.7 Mcal ME diet both in the starting and growing stages, and

TABLE 5. THE EFFECT OF DIETARY ENERGY AND PROTEIN LEVELS ON AGE AT FIRST LAY

Energy level (Mcal/kg)	2.70	2.75	2.80	2.85	2.90
Age at first lay (days)	117	114	115	117	114
Protein level (%)	15.5	17.0	18.5	20.0	
Age of first lay (days)	113	109	112	105	

36 birds per treatment

TABLE 6. FEED COST (BAHT / PERIOD) PER BIRD FOR RAISING DUCKS

Energy level (Mcal/kg)	2.70	2.75	2.80	2.85	2.90
Starting	10.64	11.46	11.21	11.10	11.09
Growing	66.01	67.67	70.00	71.23	72.37
Protein level (%)	15.5	17.0	18.5	20.0	21.5
Starting	–	8.47	8.60	9.69	10.02
Growing	44.40	50.70	53.40	59.50	–

the highest cost in the 2.75 Mcal ME diet in the starting stage and in the 2.90 Mcal ME diet in the growing stage. The difference between the highest and the lowest was only 0.82 Baht in the starting 4-week period and 6.36 Baht in the growing 14-week period. In the trial for protein requirement, the lowest cost in the starting and growing stages was shown in the 17.0% and 15.5% protein diets, respectively, and the highest cost was in the 21.5% and 20.0% protein diets. The difference between the lowest and the highest costs was 1.55 Baht for the starting 4-week period and 15.1 Baht for the growing 14-week period.

In conclusion, Khaki Campbell × Thai Native ducks can be raised with the low energy (2.70 Mcal ME/kg) and low protein (17.0% in the starting 4 weeks and 15.5% in the growing 14 weeks) diet without any ill-effects, and such diets are more practical from the economical point of view. In this experiment replication was not enough to make a precise comparison, and the energy and protein levels used here did not give

any difference in the weight gain and the feed efficiency. In this sense, further experiments should be done using diets much lower in energy and protein levels.

Literature Cited

Adams, R. L. and W. J. Stadelman. 1979. Energy and protein levels for growing female ducks. Poult. Sci. 58:1030.

Bandong, F. C., A. L. Alejar, C. R. Arboledga, D. M. Calvo, E. C. Coligado, S. L. Eduarde, F. M. Fronda, D. L. Evangelista, D. V. Mondonedo, P. S. Faylon and A. N. Eusebio. 1977. The Philippines Recommends for Duck Raising. The Philippines Council for Agriculture and Resources Research.

Bodwell, C. E., J. D. Saterlee and L. R. Hackler. 1980. Protein digestibility of same protein preparations by human and rat assays and by *in vitro* enzymic digestion methods. Am. J. Clin. Nutr. 33:677-686.

Bondi, A. A. 1987. Animal Nutrition. John Wiley & Sons Ltd. NY. pp. 402-406.

Bush, B. M. 1975. Veterinary Laboratory Manual. William Heinemann Medical Books Ltd. London. pp. 125-129.

Chin, D. T. F. and R. I. Hutagalung. 1984. Energy and protein requirements of Pekin broiler ducks in tropical environment. Proc. Conf. Malay. Soc. Animal Prod.

Das, I. N., D. B. Mishra and G. Biswal. 1965. Comparative anatomy of the domestic duck. Ind. Vet. J. 42:320-326.

Dean, W. F. 1968. Effect of energy-protein balance, energy intake and methionine on growth and body composition of ducklings. Poult. Sci. 47:1665-1666.

Dean, W. F., M. L. Scott and R. J. Young. 1965. Protein requirement of ducklings at different stages of growth. Poult. Sci. 44:1363.

Duncan, D. B. 1955. Multiple range and multiple F test. Biometrics. 11:1-42.

Du Preez, J. J. and J. P. H. Wessels. 1970. The effect of different dietary treatments on the two-week body weight and carcass composition of ducklings. Agroanimalia. 2:185-190.

Eswaran, K. R., A. Ramakrishnan, C. K. Venugopalan and G. R. Nair. 1984. Comparative performance of Khaki Campbell and desi ducks. Ind. J. Poult. Sci. 19:70-73.

Gowd, K. P. L., V. R. Reddy and P. V. Rao. 1983. Protein and energy requirements of Khaki Campbell grower ducks. Ind. J. Animal. Sci. 53:1271-1276.

Hamilton, E. M. and E. Whitney. 1980. Nutrition Concepts and Controversies. West Publishing Co. St. Paul. pp. 461-462.

Khajarern, J. and S. Khajarern. 1987. Poultry Feeds and Feeding. Khon Kaen Univ. p. 360.

Leclercq, B. and H. De Carville. 1976. Influence of the protein and energy contents of the diet on the growth of young Muscovy ducklings. Arch. Geflugelk. 40:117-119.

Mohamed, K., B. Leclercq, A. Anwar, H. El-Alaily and H. Soliman. 1984. A comparative study of metabolizable energy in duckLings and domestic chicks. Anim. Feed Sci. Tech. 11:199-209.

Morgan, A. F. 1962. Nutrition of the aging. Gerontologist. 2:77-84.

Oluyemi, J. A. and H. L. Fetuga. 1978. The protein and energy requirements of ducklings in the tropics. Br. Poult. Sci. 19:261-266.

Pan, C. M., C. Tai, J. C. Chen, H. H. Huang and T. F. Shen. 1978. The protein and metabolizable energy requirements of growing ducks. Taiwan Livest. Res. 11:1-10.

Prasad, S. S., V. R. Reddy and P. V. Rao. 1988. Protein and energy requirements for starter and grower Khaki Campbell ducks. Ind. J. Poult. Sci. 23:296-305.

Reddy, M. S., V. R. Reddy and P. M. Reddy. 1980. Studies of protein and energy requirements in Khaki Campbell ducklings. Ind. J. Poult. Sci. 15:137-144.

Sahoo, G., P. Panda, M. Mishra and S. C. Sahoo. 1985. Study of growth in Khaki Campbell ducks. Ind. J. Poult. Sci. 20:220-222.

Sainsbury, D. 1980. Poultry Health and Management. Grada Publishing Ltd. London. p. 365.

Shafiuddin, A. 1985. Duck production in Bangladesh. Proc. Duck Prod. Workshop, Bogor. Nov. 18-22, 1985.

Siregar, A. P. and D. J. Farrell. 1980. A comparison of the energy and nitrogen metabolism of ducklings and chicken. Br. Poult. Sc. 21:213-227.

Sreeraman, P. K., M. N. Ahmad, P. R. Rao and G. A. Sastry. 1979. Hematology of ducks. Ind. Vet. J. 56:100-104.

Steel, R. G. D. and J. H. Torrie. 1980. Principles and Procedures of Statistics: A Biometric Approach. McGraw Hill Co. NY. pp. 137-145.

Wong, S. Y. 1928. Colorimetric determination of iron and hemoglobin in blood. J. Biol. Chem. 77 : 409-415.