The Comparison of Muscle Protein Turnover Rate among the Egg Type, Meat Type and Japanese Native Chicken Stocks

Yoshizane MAEDA, Shin OKAMOTO, Kaoru OKANO*

Takeshi TOMITA** and Tsutomu HASHIGUCHI

Department of Animal Science, Faculty of Agriculture,
Kagoshima University, Kagoshima-shi 890

* University Farm of Faculty of Agriculture, Kyushu
University, Kasuya-machi, Fukuoka-ken 811-24

** Department of Animal Science, School of Agriculture,
Nagoya University, Chikusa-ku, Nagoya-shi 464-01

(Received November 16, 1989)

Abstract The fractional rates (%/day) of synthesis (Ks) and degradation (Kd) of muscle protein, at 4 weeks of age, of the egg type stock (two strains of White Leghorn), meat type stock (two kinds of commercial broiler) and Japanese native stock (four breeds of Japanese native breeds) of chicken were determined by measuring the output of N³-methylhistidine (N³-MH) in their excreta. The fractional rates of synthesis of muscle protein for the egg type stock, meat type stock and Japanese native stocks were 6.2-6.8%/day, 7.3-8.9%/day and 8.8-12.6%/day, respectively. The fractional rates of degradation of these stocks were 4.6-5.1%/day, 2.8-3.0%/day and 6.1-7.7%/day, respectively. The Japanese native stock showed higher rates of both synthesis and degradation of muscle protein. The Kd values for Japanese native stock were about 1.2-1.7 times higher than those for the egg type, and 2.0-2.8 times higher than those for meat type stock. The Ks/Kd values in egg type, meat type and Japanese native stocks were calculated as 1.22-1.48, 2.61-2.97 and 1.40-1.68, respectively. The accumulation efficiency of muscle protein, (Ks-Kd)/Ks, at 4 weeks of age was calculated as 17.7-32.4% in the egg type stock, 61.6-66.3% in the meat type stock and 28.8-40.5% in the Japanese native stock.

These results suggest that 1) the muscle protein turnover rate differs between egg type, meat type and native stocks, and 2) the improvements for body size, growth rate and/or feed efficiency caused changes in muscle protein turnover rate, in the relative value (Ks/Kd) of protein synthesis to degradation and in the accumulation efficiency (Ks-Kd)/Ks of muscle protein.

Jpn. J. Zootch. Sci., 61 (8) : 701-706, 1990

Key words : muscle, protein turnover, chicken, differentiation, improvement

The accumulation of muscle protein is determined by the fractional rates of synthesis and degradation of muscle protein¹⁾. Rates of synthesis and degradation of muscle protein are usually measured using specific radioactivity or urinary N³-methylhistidine (N³-MH). N³-MH is a component of actin in all muscle

and of myosin in white muscle, and approximately 90% of N³-MH is located in skeletal muscle^{2,3)}. N³-MH from muscle protein cannot be reutilized for protein synthesis and is rapidly and quantitatively excreted in the urine after its release by degradation⁴⁾. ASATOOR and ARMSTRONG⁵⁾ proposed that the rate of

degradation of muscle protein could be estimated by measuring the urinary excretion of N^{τ} -MH. By measuring the urinary excretion of N^{τ} -MH, the muscle protein turnover rate has been studied in rats^{3,6,7}, humans^{8,9}, cattle¹⁰⁻¹², chicken¹³⁻¹⁵ and Japanese quail¹⁶⁻¹⁹.

The muscle protein turnover rate is associated with the growth rate. The chicken is a typical animal having a variety of body sizes. By selective breeding for body size, egg production and/or feed utilization, many breeds and strains with various characteristics have been developed. Chickens are thus able to be classified into egg type stock, meat type stock and native stocks. These stocks have different growth rates and body sizes.

In the present study, the authors examined the fractional rates of synthesis (Ks) and degradation (Kd) of muscle protein and the characteristics of muscle protein turnover rate in egg type, meat type and native chicken stocks.

Materials and Methods

Egg type stocks (two Strains of White Leghorn), meat type stocks (two kinds of commercial broiler) and Japanese native stocks

Table 1. Standard values of body weight(gr.) of male at 4 weeks of age and adult in each stock

Stock	4 Weeks of Age	Adult
Egg type		
White Leghorn (E1) ¹⁾	250	2200
White Leghorn (E2)	250	2300
Meat type		
Commercial broiler (M1)	880	4500< ²⁾
Commercial broiler (M2)	850	4500< ²⁾
Japanese native chicken		
Gifujidori	110	1300
Tokarajidori	190	2200
Tokuchijidori	80	900
Katsura Chabo	90	750
Goishi Chabo	100	750

¹⁾ E1 : Shaver
E2 : Dekalb
M1 : Hubbard
M2 : Cobb

²⁾ Parent stock

(Gifujidori, Tokarajidori, Tokuchijidori, and 2 kinds of Chabo ; Japanes Bantum) were used in this study. The standard values of body weight for adult native chickens are shown in Table 1. All chickens were reared in an electric brooder from hatching to 2 weeks of age, and were then housed in wire-floored individual cages that were artificially lighted for 14 hrs per day and maintained at 22°C. The feed consisted of a Kagoshima University formula which contained 19% crude protein and 13.1 MJ/kg metabolizable energy. Both water and feed were available *ad libitum*. The feed intake was individually measured by the use of feeder cups²⁰.

Complete 24-hr excreta collections were individually obtained for 4 days at 4 weeks of age. Each sample was stored at -40°C until analyzed for N^{τ} -methylhistidine (N^{τ} -MH). The quantitative analysis of N^{τ} -MH was conducted via the methods described by NISHIZAWA *et al.*²¹. Each excreta sample was hydrolyzed with 80 ml of 6 N-HCl at 110°C for 24 hrs to convert acetyl N^{τ} -MH to N^{τ} -MH. After filtration of the hydrolyzed sample, the filtrates were reduced to dryness and redissolved in distilled water. This procedure of drying and redissolving was repeated eight times. The dried hydrolysate was dissolved in 10 ml of 0.2 M pyridine and 4 ml of the dissolved hydrolyzate applied to a Dowex 50-X 4 column (pyridine form 200-400 mesh). After the acidic and neutral amino acids were eluted with 200 ml 0.2 M pyridine, N^{τ} -MH was eluted with 100 ml 1 M pyridine²¹. The N^{τ} -MH assay was conducted via the methods described by WARD²².

The fractional rates of degradation and synthesis of muscle protein were calculated based on the daily amount of N^{τ} -MH excretion¹³. In order to calculate accurately the fractional rates of synthesis and degradation for myofibrillar protein, it is necessary to estimate the pool size (P) of N^{τ} -MH in the whole body skeletal muscle and the amount of

Muscle Protein Turnover Rate of Various Chicken Stocks

N^T -MH originating from the degradation of skeletal muscle. The pool size (P) of N^T -MH in the whole body skeletal muscle was calculated by the equation in the previous report¹³⁾.

Fractional rates of degradation (Kd) and synthesis (Ks) of the muscle protein in each bird were calculated by FUNABIKI *et al.*²³⁾. The accumulation efficiency of muscle protein was calculated via the equation of (Ks-Kd)/Ks that was proposed by FUNABIKI²⁴⁾. Data were analyzed by a one-way analysis of variance. The statistical significance of differences among means was evaluated via Duncan's multiple-range test.

Results

The fractional rates of synthesis and degradation of muscle protein in the egg type stock, meat type stock and native chicken stocks at 4 weeks of age are given in Table 2. The means of the fractional rate of synthesis (Ks) of muscle protein at 4 weeks of age of the egg type stock, meat type stock and native stocks were 6.2-6.8%/day, 7.3-8.9%/day and 8.8-14.2%/day, respectively. The Ks values for the egg type and meat type stocks, which are improved breeds, were statistically lower ($P < 0.05$) than those of native stocks except Chabo

(Japanese bantam). The Ks values for muscle protein in the meat type stocks were slightly higher than those in the egg types. The difference in Ks value between the egg type and meat type was significant in M_1 for commercial broilers, but not significant in M_2 . In the native stock, the Ks values for Chabo were smaller than those for other native breeds except Tokuchijidori.

The fractional rates of degradation (Kd) at 4 weeks of age for the egg type stock, meat type stock and native stocks were 4.6-5.1%/day, 2.8-3.0%/day and 6.1-9.6%/day, respectively. The Kd values for native stock were about 1.2-1.7 times higher than those for the egg type stock and 2.0-2.8 times higher than those for the meat type stock. The Kd values for meat type stocks were almost half those for the egg type stock. In the native stock, the Kd values for the Chabo (6.1-6.4%/day) were smaller than those for other breeds (7.3-9.6%/day). Both Ks and Kd values in native stock were higher than those in egg type and meat type stocks. The relative values of Ks to Kd, (Ks/Kd), in the egg type, meat type and native stocks were calculated as 1.22-1.47, 2.61-2.96 and 1.40-1.68, respectively. The values for the commercial broiler were rela-

Table 2. Fractional rate of synthesis and degradation of muscle protein at 4 weeks of age in the various chicken stocks

Stocks	No. of birds	Ks (%/day)	Kd (%/day)	Ks/Kd	(Ks-Kd)/Ks (%)
		Mean±SD	Mean±SD		
Egg type					
White Leghorn (E1) ¹⁾	5	6.9±1.0 ^{ab}	4.6±0.7 ^{bc}	1.47	32.4
White Leghorn (E2)	8	6.2±0.9 ^a	5.1±0.6 ^{cd}	1.22	17.7
Meat type					
Commercial broiler (M1)	8	8.9±1.1 ^b	3.0±0.8 ^a	2.96	66.3
Commercial broiler (M2)	5	7.3±0.9 ^{ba}	2.8±0.5 ^a	2.61	61.6
Japanese native chicken					
Gifujidori	8	12.6±1.6 ^d	7.5±0.7 ^{gh}	1.68	40.5
Tokarajidori	6	11.1±1.8 ^d	7.3±1.0 ^{fgh}	1.52	34.2
Tokuchijidori	4	10.9±1.9 ^{dc}	7.7±1.0 ^h	1.42	29.3
Chabo (N1)	6	8.8±1.2 ^b	6.1±0.9 ^{de}	1.44	30.6
Chabo (N2)	7	9.0±1.5 ^{bc}	6.4±1.0 ^{ef}	1.40	28.8

¹⁾ E1: Shaver E2: Dekalb M1: Hubbard M2: Cobb N1: Katsura Chabo N2: Goishi Chabo

*Column values with the same superscript are not statistically significant ($P < 0.05$)

tively higher (2.61-2.96) than those for egg type and Japanese native stocks. The values for the Japanese native stock were slightly higher than for the egg type stock.

The accumulation efficiency of muscle protein can be evaluated from the equation of $(K_s - K_d)/K_s$ suggested by FUNABIKI²⁴⁾. The values for the egg type, meat type and Japanese native stocks were calculated as 17.7-32.4%, 61.6-66.3% and 28.8-40.5%, respectively. The high values $(K_s - K_d)/K_s$ for the commercial broiler reflect the high growth rate in broilers.

Discussion

The muscle protein turnover rate of chicken has been estimated from the excretion of the N^{τ} -MH, in normal and dystrophic strains^{25, 26)}, via variation among chicken stock^{12, 13)}, and via the effect of the *dw* gene²⁷⁾. In order to use N^{τ} -MH excretion in urine as a measurement of muscle protein breakdown, it is necessary to demonstrate that (1) the N^{τ} -MH in muscle protein cannot be reutilized or protein synthesised and is rapidly and quantitatively excreted in the urine after it is released by degradation of myofibrillar protein and that (2) skeletal muscle is the major source of urinary N^{τ} -MH. SAUNDERSON and LESLIE²⁸⁾ demonstrated that urinary excretion is valid for estimating the muscle protein degradation rate in chicken because of quantitative recovery of N^{τ} -MH within 1 week except for turkey. However, HARRIS *et al.*²⁹⁾ reported that excretion of N^{τ} -MH by 4 to 18-week-old cockerels cannot be used as a reliable index of muscle protein breakdown *in vivo*, because recovery of radioactivity in excreta were incomplete and progressively decreased with increasing age. In his data, the recovery of radioactivity during the first 4 days showed almost 85% at 4 weeks of age. Although recovery at 4 weeks of age is not 100%, we considered that excretion of N^{τ} -MH at 4 weeks of age is not entirely invalid for a relative comparison of muscle protein degradation rate among various chicken

stocks.

It is well known that muscle protein turnover rate is associated with growth rate^{16, 30)}. MAEDA *et al.*¹⁶⁾ demonstrated that the growth rate correlates highly with the muscle protein turnover rate. In this study, the muscle protein turnover rates at 4 weeks of age differed between the chicken stocks. Both K_s and K_d values for Japanese native stock were higher than those for meat type and egg type stocks. The meat type stock showed a higher K_s and a lower K_d than the egg type stock. It was recognized that the improvement in economical traits caused the changes in both K_s and K_d to adapt to economic performance.

In previous reports, HAYASHI *et al.*¹⁴⁾ and MAEDA *et al.*^{13, 25)} measured the muscle protein turnover rates of White Leghorn, commercial broilers, White Plymouth Rock, Satsuma native fowl (one kind of Japanese native fowl) and Fayoumi. Although the muscle protein turnover rates for egg type stock and Japanese native stock in the present study are similar to the results of the previous report¹³⁾, the K_s for commercial broilers (7.3-8.9%/day) in the present study is lower than that in the previous report (11.1-12.6%/day). HAYASHI *et al.*¹⁴⁾ measured the muscle protein turnover rate of layers and broilers at 3, 6 and 9 weeks of age. From his data, if we calculate the muscle protein turnover rate at 4 weeks of age in proportion to age, we can expect that K_s and K_d values in layers are 10.4%/day and 5.6%/day, respectively, while those values in broilers are 8.6%/day and 4.3%/day, respectively. The K_s value in layer and K_d value in broilers were higher than the corresponding rates in the present study.

MAEDA *et al.*^{13, 25)} demonstrated the K_s and K_d values for muscle protein in Satsuma native fowl (Satsumadori) were 13.9%/day and 8.6%/day, respectively, and that those in Fayoumi were 13.0%/day and 10.1%/day, respectively. The muscle protein turnover rates for Satsuma native fowl and Fayoumi are similar to the

turnover rates for the Japanese native chicken stock breeds listed in Table 2.

Both K_s and K_d values in the Japanese native chicken stock were high compared to the egg type and meat type stocks. Most Japanese native fowls were not selected for any economic characteristics such as body weight, feed efficiency or egg production. Therefore native chickens are slow in growth rate and small in body weight compared to improved chickens. Skeletal muscle in the body is the major protein deposit of an animals body and has the function of controlling movement as well as preserving protein and providing amino acids in protein metabolism. Therefore high muscle protein turnover rates in the native chicken may be associated with their growth rate and body weight. If there is an association between muscle protein metabolic rate and fitness, a high metabolic rate at an early age is probably advantageous for survival. It is likely that jungle fowl in habitats in the jungle of Southeast Asia have a high metabolic rate of muscle protein.

The differences between stocks in muscle protein turnover rate were shown also in the genetically diverse quail lines selected for body size¹⁶⁾. The K_s and K_d values were significantly different among three lines of quail selected for body size at 6 weeks of age, the fractionnal rate of synthesis of the line selected for large body size (LL line : 13.2%/day) was higher than that of the non-selected line (RR line : 11.5%/day), whereas the line selected for small body size (SS line : 8.1%/day) was lower than that of the RR line. The fractional rates of degradation, at 3 weeks of age, for both the LL line (4.1%/day) and the SS line (5.6%/day) were lowr than that of RR line (7.0%/day). The muscle protein turnover rate tendencies of the three quail lines selected for body size were similar to those of the egg type, meat type and Japanese native chicken stocks.

Both K_s/K_d values and $(K_s-K_d)/K_s$ values were low in the egg type stock and high in the

meat type stock. Those values for native stock were slightly higher than those for egg type stock.

In conclusion, both K_s and K_d values in native stock were higher than those of the egg and meat type stocks (improved breeds). The meat type stock with high growth rate demonstrated relatively high K_s value and low K_d value compared to the egg type stock. These results suggest that genetic improvements in body size, growth rate and/or feed utilization caused a change in muscle protein turnover rate.

Acknowledgment

Author wish to express their thanks to Dr. K. HAYASHI, Department of Animal Science, Faculty of Agriculture, Kagoshima University, for his technical advice, and to Ms. Mary Fitzgerald, Board of Education of Ijuin-cho, Kagoshima Prefecture, for her reading of the manuscript.

References

- 1) MILLWAED, D.J., P.J. GARLICK, D.O. NNANYELGO and J.C. WATERLOW, *Biochem. J.*, **156** : 185-188. 1976.
- 2) HAVERBERG, L.N., P.T. OMSTED, H.N. MUNRO and V.R. YOUNG, *Biochim. Biophys. Acta.*, **405** : 67-71. 1975.
- 3) NISHIZAWA, N., M. SHIMBO, S. HAREYAMA and R. FUNABIKI, *Br. J. Nutr.*, **37** : 345-353. 1977.
- 4) COWGILL, R.W. and B. FREEBURG, *Arch. Biophys.*, **71** : 466-472. 1957.
- 5) ASATOOR, A.M. and M.D. ARMSTRONG, *Biochem. Biophys. Res. Commun.*, **26** : 168-174. 1967.
- 6) NISHIZAWA, N., R. FUNABIKI and S. HAREYAMA, *J. Nutr. Sci. Vitaminol.*, **21** : 383-385. 1975.
- 7) YOUNG, V.R., S.D. ALEXIS, B.S., BALIGA, H.N. MUNRO and W. MUECKE, *J. Biol. Chem.*, **247** : 3592-3600. 1972.
- 8) LONG, C.L., L.N., HAVERBERG, V.R., YOUNG, J.M. KINNY, H.N. MUNRO and J.W. GEIGER, *Metabolism*, **24** : 929-935. 1975.
- 9) MURRY, A.J., BALLERD, F.J. and F.M. TOMAS, *Anal. Biochem.*, **116** : 537-544. 1981.
- 10) HARRIS, C.I. and G. MILNE, *Br. J. Nutr.*, **45** : 411-422. 1981.
- 11) MCCARTHY, F.D., W.G., BERGEN and D.R. HAWKINS, *J. Nurt.*, **113** : 2455-2463. 1983.
- 12) NISHIZAWA, N., Y. TOYODA, T. NOGUCHI, S. HAREYAMA, H. ITABASHI and R. FUNABIKI, *Br. J.*

- Nutr., 42: 247-252. 1979.
- 13) MAEDA, Y., K. HAYASHI, S. TOYOHARA and T. HASHIGUCHI, Biochem. Genet., 22: 687-700. 1984.
- 14) HAYASHI, K., Y. TOMITA, Y. MAEDA, Y. SHINAGAWA, K. INOUE and T. HASHIZUME, Br. J. Nutr., 54: 157-163. 1985.
- 15) JONES, S.J., E.D. ABERLE and M.D. JUDGE, Poultry Sci., 65: 2142-2147. 1986.
- 16) MAEDA, Y., K. HAYASHI, T. HASHIGUCHI and S. OKAMOTO, Biochem. Genet., 24: 207-216. 1986.
- 17) MAEDA, Y., K. SATO, S. OKAMOTO and T. HASHIGUCHI, Biochem. Genet., 26: 395-399. 1988.
- 18) MAEDA, Y., N. YAMANOUCHI, S. OKAMOTO and T. HASHIGUCHI, Jpn. J. Zootech. Sci., 60: 484-490. 1989.
- 19) MAEDA, Y., M. FUJII, S. OKAMOTO and T. HASHIGUCHI, Biochem. Genet., 27: 603-611. 1989.
- 20) MAEDA, Y., H. KAWASAKI and T. HASHIGUCHI, Jpn. J. Zootech. Sci., 53: 388-394. 1982.
- 21) NISHIZAWA, N., T. NOGUCHI, S. HAREYAMA and R. FUNABIKI, Br. J. Nutr., 38: 149-151. 1977.
- 22) WARD, L.C., Anal. Biochem., 88: 598-604. 1978.
- 23) FUNABIKI, R., Y. WATANABE, N. NISHIZAWA and S. HAREYAMA, Biochim. Biophys. Act., 451: 143-150. 1976.
- 24) FUNABIKI, R., SEIKAGAKU., 58: 1150-1156. 1986.
- 25) MAEDA, Y., K. HAYASHI, M. MIZUTANI and T. HASHIGUCHI, Poultry Sci., 66: 757-759. 1987.
- 26) HILLGARTNOR, F.B., A.S. WILLIAMS, A. FLANDERS, D. MORIN and R.J. HANSEN, Biochem. J., 196: 591-601. 1981.
- 27) MAEDA, Y., S. MATSUOKA, N. FURUICHI, K. HAYASHI and T. HASHIGUCHI, Biochem. Genet., 25: 253-258. 1987.
- 28) SAUNDERSON, C.L. and S. LESLIE, Br. J. Nutr., 50: 691-700. 1983.
- 29) HARRIS, C.I., G. MINE and R. MCDIARMID, Br. J. Nutr., 57: 467-478. 1987.
- 30) MARTIN, R.J., Animal Growth Regulation (Campion, D.R., G.J. Hausman and R.J. Martin, eds.) 183-210. Plenum Publishing Corporation. New York., 1989

卵用鶏，肉用鶏および日本在来鶏における筋肉蛋白質 代謝回転速度の比較

前田芳實・岡本 新・岡野 香*・富田 武**・橋口 勉

鹿児島大学農学部，鹿児島市 890

* 九州大学農学部付属農場，福岡県粕屋町 811-24

** 名古屋大学農学部，名古屋市千種区 464

本研究では卵用種，肉用種および日本在来種のニワトリについて，4週齢時における筋肉蛋白質代謝回転速度の分析を行なった。筋肉蛋白質代謝回転速度は糞尿中の N^2 -MH の排泄量より推定した。卵用種，肉用種および在来種の筋肉蛋白質の合成速度 (K_s) はそれぞれ 6.2-6.8%/day, 7.3-8.9%/day および 8.8-12.6%/day; また分解速度 (K_d) はそれぞれ 4.6-5.1%/day, 2.8-3.0%/day および 6.1-7.7%/day と評価された。また，在来鶏は一般に合成速度および分解速度共に高い値を示す傾向にあり， K_d は卵用種および肉用種の約2倍の値を示した。 K_s/K_d 値は卵用種，肉用種および在来種において，それぞれ 1.22-1.48, 2.61-2.97 および 1.40-1.68 と評価された。筋肉蛋白質の蓄積効率と考えられる $(K_s-K_d)/K_s$ は卵用種，肉用種および在来種でそれぞれ 17.7-32.4%, 61.6-66.3% および 28.8-40.5% と計算され，ブロイラーは他の鶏種に比較して蓄積効率が高いことが示された。筋肉蛋白質代謝回転速度のこれらの鶏種間のちがいは，それぞれの鶏種の産肉性や飼料利用性の実態をよく反映している。

日畜会報, 61 (8) : 701-706, 1990