aJava

Asian Journal of Animal and Veterinary Advances

Academic
Journals Inc.

www.academicjournals.com

OPEN ACCESS Asian Journal of Animal and Veterinary Advances

ISSN 1683-9919
DOI: 10.3923/ajava.2018.295.300

Research Article
Influence of Lighting Systems on Some Muscle Development Related Genes and Production Traits in Japanese Quail

[1]Walaa Slouma Hamouda Abd El Naby and [2]Heba Abdo Basha

[1]Genetics and Genetic Engineering in Department of Animal Husbandry and Animal Wealth Development, Faculty of Veterinary Medicine, Alexandria University, Alexandria, Egypt
[2]Poultry Breeding and Production, Department of Animal Husbandry and Animal Wealth Development, Faculty of Veterinary Medicine, Alexandria University, Alexandria, Egypt

Abstract

Background and Objective: Light is an important environmental aspect of growth and reproduction. This study was conducted to evaluate the effect of natural light (13 h L:11 h D) (group I) versus artificial continuous light (23 h L:1 h D) (group II) on the gene expression of myogenic regulatory factors (*MRFs*) genes (*MyoG* and *MyoD*), Myostatin (*MSTN)* and signal transducer and activator of transcription 5b (*STAT5b*) in the pectoralis major muscle of male Japanese quail (*Coturnix coturnix japonica*) at 6 weeks of age and its association with the production performance. **Materials and Methods:** Newly hatched Japanese quail chicks (200) were used in this study. Allocated into 2 groups at the 5th day of age and each group exposed to different lighting systems (13 h L:11 h D) and (23 h L:1 h D). The average body weights of Japanese quail at 7 and 14 days of age was calculated, then weekly individual weights were recorded until 42 days. Daily feed intake, overall daily weight and feed conversion ratio were calculated. The expression analysis of myogenic regulatory factors (*MRFs*) genes, *MSTN* and *STAT5b* in the pectoralis major muscle of male birds at 6 weeks of age was done using quantitative real-time PCR. **Results:** The results showed that *MyoG* and *STAT5b* mRNA levels showed an increase in group II relative to group I this was in reverse to *MyoD* and *MSTN* transcript levels. Expression changes of these genes were associated with alteration of productive traits. Since group II had heavier final mature body weight and weight gain than group I. **Conclusion:** The lighting system influenced the expression profile of myogenic regulatory factors (*MRFs*) genes (*MyoG* and *MyoD*), *MSTN* and *STAT5b* which consequently reflected on the productive traits of Japanese quail.

Key words: Japanese quail, lighting system, body weight, MRFs, STAT5b expression

Received: October 23, 2017 Accepted: December 28, 2017 Published: April 15, 2018

Citation: Walaa Slouma Hamouda Abd El Naby and Heba Abdo Basha, 2018. Influence of lighting systems on some muscle development related genes and production traits in Japanese quail. Asian J. Anim. Vet. Adv., 13: 295-300.

Corresponding Author: Walaa Slouma Hamouda Abd El Naby, Genetics and Genetic Engineering in Department of Animal Husbandry and Animal Wealth Development, Faculty of Veterinary Medicine, Alexandria University, Egypt Tel: +201020285003

Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

The lighting system is a critical component that affects bird's body weight. Higher body weight is the main goal of the poultry breeder. It is a quantitative trait which affected by genes and environmental conditions. The poultry house system is one of the main factors that should be efficient maintained to achieve the productivity success. Light hour's length is one of the environmental factors, which affects poultry production and causing physiological changes in the growth parameters[1,2] carcass traits[3,4] and sexual maturity[5,6].

The modulation of the light and dark period that the bird receives affects its body weight, via the change in energy intake and loss[1,7]. Also, Charles *et al.*[8] illustrated the different body weight in various photoperiodism due to the change of the balance between feed intake and feed digestion. Moreover, the external stimuli of light proved to affect the proliferation and differentiation of the muscle cells through its primary effect on the gene expression of myogenic regulatory factors (*MRFs*) which include myogenic determination factor (*MyoD)*, Myogenin (*MyoG*), Myogenic factor 5 (*Myf 5*) and *MRF4* (*Myf 6* or herculin)[9].

Skeletal muscle development in birds was controlled by several *MRFs*[10]. *MyoD* and *Myf5* are factors involved in the determination of myogenic cells, whereas *MyoG* is mainly engaged in the muscle terminal differentiation process and triggers the expression of myotube-specific genes[11,12]. Moreover, Myostatin (*MSTN*) is an essential factor for growth and development of muscle mass in vertebrate species[13]. Signal transducer and activator of transcription 5b gene (*STAT5b*) participates in regulation of several components of the GH-insulin-like growth factor 1 (*IGF-1*) axis, long recognized as fundamental for growth-promoting actions of GH[14,15].

Japanese quail (*Coturnix coturnix japonica*) is derived from wild Japanese quail. Its production considered an alternative source of meat due to its fast growth rate, early sexual maturity, easy handling, high immunity, inexpensive rearing costs and great laying ability[16,17].

The aim of the present study was to investigate the impact of two lighting systems, natural light (13 h L: 11 h D) at fall season and artificial continuous lighting (23 h L: 1 h D) on the expression of some *MRFs* genes (*MyoG* and *MyoD*), *MSTN* and Signal transducer and activator of transcription 5b (*STAT5b*) in Japanese quails. Moreover, their effect on some productive traits as body weight, feed intake, daily weight gain and feed conversion ratio.

MATERIALS AND METHODS

This experiment was conducted at fall season (End of September and first of October, 2016), at the Department of Animal husbandry and animal wealth development, Faculty of Veterinary Medicine, Alexandria University.

Birds sample and lighting system: Newly hatched Japanese quail chicks (n = 200) were used in this study. For the first 5 days of age the chicks were subjected to 24 h L: 0 h D (13 h natural and 11 h artificial). The birds were randomly allocated into two separate groups (100 birds/each) at the 5th day of hatch. The group I exposed to natural light depended on sunlight (average 13 h L: 11 h D). Group II was transported to light tight room where the birds were exposed to artificial continuous lighting system 23 h L: 1 h D. Both groups were reared on a deep litter system. The temperature was started at 36°C then reduced 3°C daily until reaching 21°C.

Housing and feeding management: All birds had access to feed and water *ad libitum* during the experimental period. The bird ration contains all needed vitamins and amino acids according to the NRC recommendation[18]. The ingredients and chemical composition of the basal diets were presented in (Table 1). Where, vegetable oil composed of soybean oil, cotton seed oil and sun-flower oil. Dicalcium phosphate 18% phosphorus, 23% calcium. Minerals and vitamins premix (Pharma Mix) purchased by Egypt Pharma for pharmaceuticals and feed additives industries (Bach No. 02100033). Each 3 kg contains: vitamin A 12.000.000 I.U., vitamin D_3 2.500.000 I.U., vitamin E:10.000 mg, vitamin K_3: 2.00 mg, vitamin B_1 1.000 mg, vitamin B_2: 5.000 mg, vitamin B_6:1.500 mg, vitamin B_{12}10 mg, niacin: 30.000 mg, folic acid: 1.000 mg, biotin: 50 mg, pantothenic acid: 10.000 mg, copper 10.000 mg, iodine: 10.000 mg, selenium: 100 mg, iron: 30.000 mg, manganese 60.000 mg, zinc: 50.000 mg, cobalt: 100 mg, $CaCo_3$ add to 3000 g mL-Lysine 78% produced by Archer Daniels Medl and

Table 1: Ingredient composition (% DM) of the basal diets

Ingredients	Starter-grower
Ground yellow corn	54.90
Soya bean meal (44% CP)	35.50
Corn gluten meal (60% CP)	6.00
Vegetable oil	0.61
Di-calcium phosphate	0.82
Ground limestone	1.30
Common salt	0.40
Mineral and vitamin premix	0.25
Lysine	0.20
Methionine	0.03

Company De Caur I.L. made in USA (ADM). DL-Methionine 99% Canadian registration number 990137 guaranteed analysis L-Methionine 99.5%, DL-Methionine 99%.

Productive parameters: The average body weights of Japanese quail at 7 and 14 days of age were calculated by division of the sum of each group weights on its individual number. On the 21st day, the birds were sexed by the color pattern of the breast feathers and wing banded for identification of individual birds (46 males and 54 females in group I and 53 males and 47 females in group II) also, weekly individual weights were recorded until 42 days. Daily feed intake was measured by subtracting the weight of left over feed from the introduced feed at the previous day.

Tissue collection: At 6 weeks of age, 3 male birds from both groups were slaughtered and the pectoralis major muscle was rapidly collected. Tissue samples were immediately stored at -80°C until further use. Tissue was collected only from male birds as mature female quail weights were not completely actual due to the development of the heavier female reproductive organs than those of males[19].

Total RNA extraction and cDNA synthesis: Total RNA of pectoralis major muscle was extracted using the Biozol (Bioflux, Japan) according to manufacturer's instructions. Subsequently, RNA was reverse-transcribed into cDNA using the SensiFAST™cDNA Synthesis Kit (Bioline, United Kingdom) according to manufacturer's instructions. Briefly, 4 µL of total RNA was mixed with 4 µL 5X buffer, 1 µL reverse transcriptase and 11 µL RNase\ DNase free H_2O added. The thermal cycler program carried out by 25°C for 10 min, 42°C for 15 min (reverse transcription) and 4°C hold. The getting cDNA was checked by *GAPDH* gene, then stored at -20°C until further use.

Table 2: Primers used for quantitative real-time PCR

Gene	Sequence (5`-3`)	Tm (°C)	Bp
MSTN	F: GCAAAAGCTAGCAGTCTATG	59	119
(NM_001001461)	R: TCCGTCTTTTTCAGCGTTCT		
MyoD	F: GATTTCCACAGACAACTCCACAT	60	116
(NM_204214)	R: GAATCTGGGCTCCACTGTCACT		
MyoG	F: GTGGGATGGTGATGCTGGAA	60	109
(NM_204184)	R: TTGGAGAGGAGTGGGAAAGGA		
STAT5b	F: CTGCTGTGTGATGGAGTACC	60	154
(NM_204779)	R: GACACTGAACTGCGACTCAA		
GAPDH	F: AACCCATTTTTAGAGGTCAGAG	60	147
(NM_204305)	R: TCATGAGCACCCGCAACGAT		

MSTN: Myostatin, *MyoD*: Myogenic factor 1, *MyoG*: Myogenin, *STAT5b*: Signal transducer and activator of transcription 5b, *GAPDH*: Glyceraldehyde 3-Phosphate dehydrogenase, Tm: Annealing temperature and BP: Base pair

Quantitative real-time PCR analysis and data analysis: Quantitative Real-Time PCR (RT-PCR) was carried out to reveal the expression profiles of *MyoG*, *MyoD*, *MSTN* and *STAT5b* genes using SensiFAST™ Syber green with low Rox (Bioline, United Kingdom) according to manufacturer's instruction. The thermal program was set as 95°C for 10 min as an initial denaturation followed by 40 cycles of 15 sec (95°C) and annealing temperature (Table 2) for 15 sec. A dissociation curve was performed at the end of the last cycle. The *GAPDH* gene was selected as a reference gene for expression analysis. The experimental data were analyzed by comparative threshold cycle method $2^{-\Delta\Delta ct}$ and the results was reported as fold change[20].

Statistical analysis: All collected data were compared using t-test in groups. Statistical significance was based on $p<0.05$.

RESULTS

The different lighting systems did not affect on Japanese quail growth performance during the first 2 weeks of age. The average body weight of Japanese quail subjected to natural light (group I) had no significant difference ($p<0.05$) compared to that reared in the artificial lighting system (group II) at 7 days of age (23.92±0.78 and 25.25±0.52) and at 14 days of age (68.29±1.23 and 68.29±1.23). While the recorded weights from 28-42 days of age showed that the lighting system affected significantly on the body weight ($p<0.05$), also the female quails of artificial lighting system were heavier after 28 days of the age than males in the same group (Table 3). The weights of males and females in both

Table 3: Effect of different light systems on the average body weights of different sex of Japanese quail at 3-6 weeks of age

	Male		Female	
Age (days)	Group I	Group II	Group I	Group II
21	98.70±2.97^a	101.05±1.82^a	97.70±2.43^a	97.57±2.56^a
28	151.52±2.32^b	160.00±1.62^a	160.36±1.99^b	170.00±1.10^a
35	193.95±1.56^b	198.42±1.21^a	201.67±2.21^b	211.14±2.03^a
42	213.40±1.56^b	225.83±1.83^a	230.50±2.49^b	252.71±2.98^a

Each value is expressed as Mean±SE, Mean values carrying different superscript letters within the same row are significant different ($p\leq0.05$). Group I (Natural light) and Group II (Artificial light)

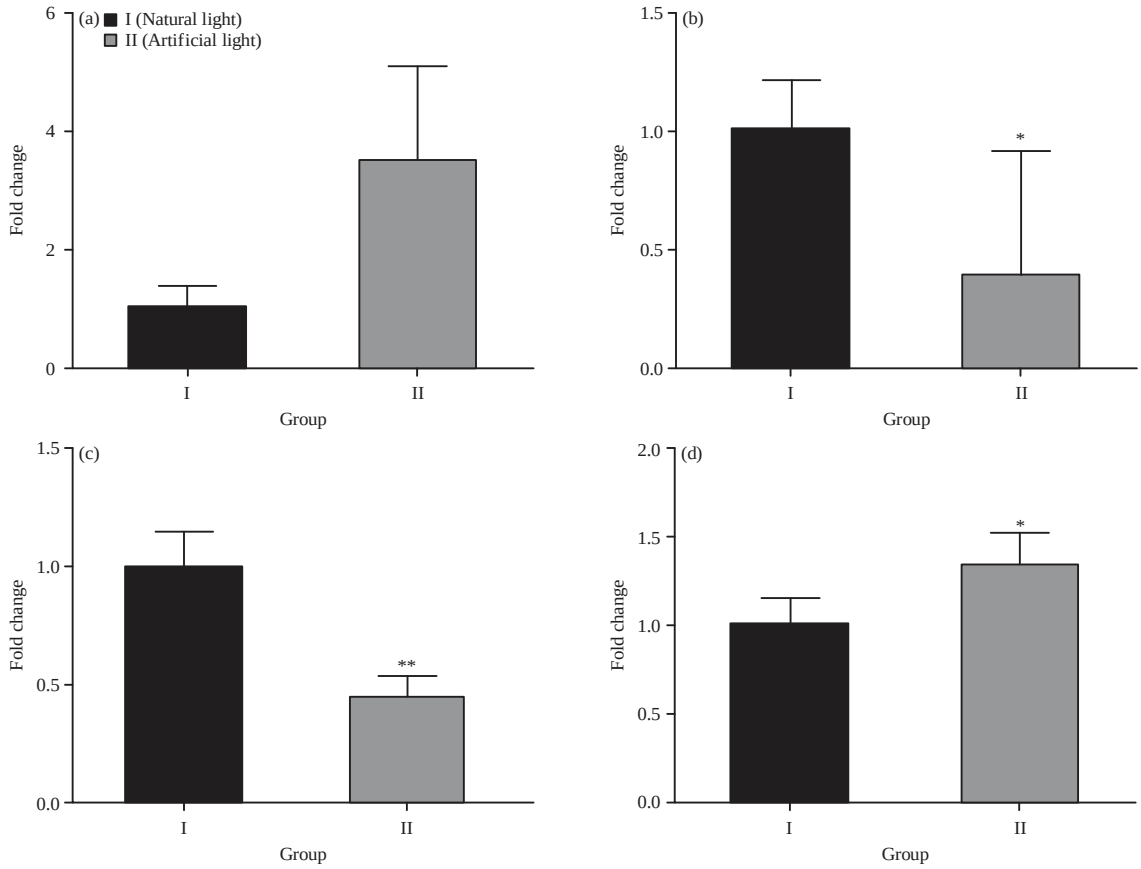

Fig. 1(a-d): Relative expression of selected genes in pectoralis major muscles of male Japanese quail exposed to group I (Natural light) and group II (Artificial light) at 6 weeks of age (a) *MyoG*, (c) *MyoD*, (c) *MSTN* and (d) *STAT5b*
*Indicate significant differences between the group I and group II, when *p<0.05, **p<0.01

Table 4: Effect of different light systems on daily feed intake, overall daily weight and feed conversion ratio of Japanese quail

Parameters	Age (days)	Group I	Group II
Feed intake	7	10.68±0.87[a]	10.45±0.60[a]
	14	18.50±0.67[a]	17.00±1.09[a]
	21	23.20±0.35[a]	21.40±1.25[a]
	28	27.74±0.93[a]	24.43±0.43[b]
	35	30.07±0.93[b]	32.62±0.40[a]
	42	36.66±0.59[a]	38.80±0.96[a]
Feed conversion ratio	1-42	4.63±1.33[a]	4.23±0.58[a]
Overall daily weight	1-42	5.05±0.38[b]	5.46±0.62[a]

Each value is expressed as Mean±SE, Mean values carrying different superscript letters within the same row are significant different ($p \leq 0.05$). Group I (Natural light) and Group II (Artificial light)

groups increased in the same sequence from 21-42 days (Table 3), but at the 4th week males and females of group II, had higher body weight than those in the group I (Table 3).

Comparison of the average of feed intake of Japanese quail in the studied groups showed that the differences between the two groups at all recorded ages were non- significant except at 28 days of age, where the group I excelled on group II (Table 4). During the experiment period, feed conversion ratio and daily weight gain in group I was 4.63 and 5.05. Whereas, in the group II were calculated as 4.23 and 5.46 (Table 4).

The expression analysis of *MyoG*, *MyoD*, *MSTN* and *STAT5b* genes in Pectoralis major muscle of male birds in both groups were shown in (Fig. 1). The *MyoG* mRNA level showed an increase in group II, which exposed to artificial light relative to group I (Fig. 1a). The relative expression of *MyoD* mRNA revealed a significant decrease (0.39±0.52) in group II compared to the group I which exposed to natural light (Fig. 1b). Also, the *MSTN* expression level was decreased significantly ($p<0.05$) in group II relative to group I (Fig. 1c). While the expression level of *STAT5b* gene revealed a significant increase (1.34±0.18) in an artificial light group relative to group exposed to natural light (Fig. 1d).

DISCUSSION

In this study the continuous lighting led to an increase of body weights and overall daily weight gain compared to birds

subjected to natural light. This may be due to permanent food availability in front of the birds. Also, the decrease of the photoperiod leads to decline of the live body weight and weight gain of the Japanese quail[7,21]. These previous studies attributed the change of body weights and weight gain in different lighting systems due to change of feeding behavior and balance between energy loss and gain by the bird's body.

The present results showed differences in expression level of *MSTN*, *MyoD* and *MyoG* in the studied groups. Other studies reported that expression of *MyoG* is necessary for the formation and differentiation of multinucleated myotubes[22,23]. The *MyoG* expression was increased in quail which have a significant increase in body weight due to exposed to continuous light. This may be the birds with continuous light have more exercise which enhances expression of *MyoG* and increases muscle fiber diameter. Moreover, Fergany *et al.*[24] showed increased expression of *MyoG* in high body weight Cobb broiler chicken at 37 days of age relative to the low body weight group. In additions, Yin *et al.*[25] indicated that the expression level of *MyoG* and *MyoD* in pectoralis muscles were greater in high weight selection relative to the low weight selection at 28 and 56 days of the adult broiler. While in the present study, *MyoD* expression level was decreased significantly (0.39 ± 0.52) in group II relative to the group I which exposed to natural light and have low body weight. So this decreased in *MyoD* expression, may be return to its involvement in the determination not in differentiation of myogenic cells, as in the case of *MyoG*.

Myostatin gene in vertebrate regulates growth negatively by limiting muscular growth during the pre-hatch and post-hatch period[26]. The relative expression level of *MSTN* gene decreased significantly in group II, which increased in body weight relative to the group I that exposed to natural lights. Also, both chicken lines (broiler and layer) revealed the lowest expression of *MSTN* gene at 6 weeks of age, which allow muscle growth and get better body weight[27]. Moreover, Liu *et al.*[28] reported that after 30 days of age, breast muscle *MSTN* expression was higher ($p<0.05$) in Wuding chicken (not selected for fast growth) than in broilers.

Several studies have indicated that *STAT5b* is the important modulators of the growth hormone, growth hormone receptor, *IGF* and insulin signaling pathways. It involved in growth, reproduction and metabolism[15,29,30,31]. The result in present study showed a significant ($p<0.05$) increase in expression of *STAT5b* in quail reared under the artificial lighting system (group II) relative to group I. Moreover, Zhao *et al.*[32] suggested that *STAT5b* gene served as a potential genetic marker for growth trait evaluation. So, the expression pattern of these studied genes is compatible with recorded body weights of the studied groups and need further study to clarify the molecular mechanism.

CONCLUSION

The present study showed that rearing Japanese quail in continuous lighting system for 6 weeks of age affected significantly on the body weight and expression profile of some muscle development and growth modulator genes. Thus it is recommended that the poultry breeder to use the continuous lighting system to promote growth performance of Japanese quail.

SIGNIFICANCE STATEMENT

This study connected for the first time between the expression profile of some muscle growth and development promoting genes and the response of body weight under different lighting systems for Japanese quails. So, this study will aid in the progress of Japanese quail production as an alternative source of meat and obtain high economic profit by application of continuous lighting system.

ACKNOWLEDGMENT

We would like to thank all staff and workers in Animal husbandry and animal wealth development, Faculty of Veterinary Medicine, Alexandria University who helped us to achieve this work.

REFERENCES

1. Bayram, A. and S. Ozkan, 2010. Effects of a 16-hour light, 8-hour dark lighting schedule on behavioral traits and performance in male broiler chickens. J. Applied Poult. Res., 19: 263-273.
2. Molino, A.B., E.A. Garcia, G.C. Santos, J.A.V. Filho, G.A.A. Baldo and I.C.L. Almeida Paz, 2015. Photostimulation of Japanese quail. Poult Sci., 94: 156-161.
3. Coban, O., E. Lacin, N. Sabuncuoglu and Z. Ozudogru, 2009. Effect of self-photoperiod on live weight, carcass and growth traits in quails (*Coturnix coturnix* Japonica). Asian-Aust. J. Anim. Sci., 22: 410-415.
4. Li, W.B., Y.L. Guo, J.L. Chen, R. Wang, Y. He and D.G. Su, 2010. Influence of lighting schedule and nutrient density in broiler chickens: Effect on growth performance, carcass traits and meat quality. Asian-Australasian J. Anim. Sci., 23: 1510-1518.

5. Jatoi, A.S., M.K. Khan, A.W. Sahota, M. Akram, K. Javed, M.H. Jaspal and S.H. Khan, 2013. Post-peak egg production in local and imported strains of Japanese quails (*Coturnix coturnix* Japonica) as influenced by continuous and intermittent light regimens during early growing period. J. Anim. Plant Sci., 23: 727-730.
6. Abd El Naby, W.S.H. and H.A. Basha, 2016. Expression of melatonin receptor subtype genes and its impact on reproductive traits in Japanese quail in different lighting systems. Avian Biol. Res., 9: 250-256.
7. Wikelski, M., L.B. Martin, A. Scheuerlein, M.T. Robinson and N.D. Robinson *et al*., 2008. Avian circannual clocks: Adaptive significance and possible involvement of energy turnover in their proximate control. Philos. Trans. R. Soc. London B: Biol. Sci., 363: 411-423.
8. Charles, R.G., F.E. Robinson, R.T. Hardin, M.W. Yu, J. Feddes and H.L. Classen, 1992. Growth, body composition and plasma androgen concentration of male broiler chickens subjected to different regimens of photoperiod and light intensity. Poult. Sci., 71: 1595-1605.
9. Sławinska, A., J. Brzezinska, M. Siwek and G. Elminowska-Wenda, 2014. Expression of myogenic genes in chickens stimulated *in ovo* with light and temperature. Reprod. Biol., 13: 161-165.
10. Berkes, C.A. and S.J. Tapscott, 2005. MyoD and the transcriptional control of myogenesis. Semin. Cell Dev. Biol., 16: 585-595.
11. Bentzinger, C.F., Y.X. Wang and M.A. Rudnicki, 2012. Building muscle: Molecular regulation of myogenesis. Cold Spring Harb. Perspect. Biol., Vol. 4. 10.1101/cshperspect.a008342.
12. Singh, K. and F.J. Dilworth, 2013. Differential modulation of cell cycle progression distinguishes members of the myogenic regulatory factor family of transcription factors. FEBS J., 280: 3991-4003.
13. Joulia-Ekaza, D. and G. Cabello, 2006. Myostatin regulation of muscle development: Molecular basis, natural mutations, physiopathological aspects. Exp. Cell Res., 312: 2401-2414.
14. Rosenfeld, R.G., A. Belgorosky, C. Camacho-Hubner, M.O. Savage, J.M. Wit and V. Hwa, 2007. Defects in growth hormone receptor signaling. Trends Endocrinol. Metab., 18: 134-141.
15. Rotwein, P., 2012. Mapping the growth hormone-Stat5b-IGF-I transcriptional circuit. Trends Endocrinol. Metab., 23: 186-193.
16. Hyánková, L. and F. Starosta, 2012. Divergent selection for shape of growth curve in Japanese quail. 6. Hatching time, hatchability and embryo mortality. Br. Poult. Sci., 53: 592-598.
17. Olanrewaju, H.A., J.L. Purswell, S.D. Collier and S.L. Branton, 2013. Interactive effects of photoperiod and light intensity on blood physiological and biochemical reactions of broilers grown to heavy weights. Poult. Sci., 92: 1029-1039.
18. NRC., 1994. Nutrient Requirements of Poultry. 9th Rev. Edn., National Academy of Science Press, Washington, DC., USA.
19. Wilson, W.O., U.K. Abbott and H. Abplanalp, 1961. Evaluation of coturnix (Japanese quail) as pilot animal for poultry. Poult. Sci., 40: 651-657.
20. Rao, X., X. Huang, Z. Zhou and X. Lin, 2013. An improvement of the 2ˆ (-delta delta CT) method for quantitative real-time polymerase chain reaction data analysis. Biostatistics Bioinf. Biomath., 3: 71-85.
21. Wagan, S.A., W. Ali, V. Nasir, R. Syed and K. Fareed *et al*., 2017. Effect of light duration on productivity of Japanese quail. Int. J. Curr. Res., 9: 45594-45596.
22. Gutierrez, J. and E. Brandan, 2010. A novel mechanism of sequestering fibroblast growth factor 2 by glypican in lipid rafts, allowing skeletal muscle differentiation. Mol. Cell. Biol., 30: 1634-1649.
23. Meadows, E., J.H. Cho, J.M. Flynn and W.H. Klein, 2008. Myogenin regulates a distinct genetic program in adult muscle stem cells. Dev. Biol., 322: 406-414.
24. Fergany, A.A.M., S.A. Hemeda, A.F. El-Nahas and W.S.H. Abd El Naby, 2017. Polymorphism and expression of some myogenic genes at embryonic stages and 37 days age of cobb broiler chickens and their impact on the marketing weights. Int. J. Recent Scient. Res., 8: 19435-19440.
25. Yin, H., S. Zhang, E.R. Gilbert, P.B. Siegel and Q. Zhu *et al*., 2014. Expression profiles of muscle genes in postnatal skeletal muscle in lines of chickens divergently selected for high and low body weight. Poultr. Sci., 93: 147-154.
26. Sato, F. and M. Kurokawa, 2006. Gene silencing of myostatin in differentiation of chicken embryonic myoblasts by small interfering RNA. Am. J. Physiol. Cell Physiol., 291: C538-C545.
27. Bhattacharya, T.K., R.N. Chatterjee, K. Dushyanth and R. Shukla, 2015. Cloning, characterization and expression of myostatin (growth differentiating factor-8) gene in broiler and layer chicken (*Gallus gallus*). Mol. Biol. Rep., 42: 319-327.
28. Liu, L.X., T.F. Dou, Q.H. Li, H. Rong and H.Q. Tong *et al*., 2016. Myostatin mRNA expression and its association with body weight and carcass traits in Yunnan Wuding chicken. Genet. Mol. Res., Vol. 15, No. 4. 10.4238/gmr15048967.
29. Barclay, J.L., C.N. Nelson, M. Ishikawa, L.A. Murray and L.M. Kerr *et al*., 2011. GH-dependent STAT5 signaling plays an important role in hepatic lipid metabolism. Endocrinology, 152: 181-192.
30. Hennighausen, L. and G.W. Robinson, 2008. Interpretation of cytokine signaling through the transcription factors STAT5A and STAT5B. Genes Dev., 22: 711-721.
31. Pilecka, I., A. Whatmore, R.H. van huijsduijnen, B. Destenayes and P. Clayton, 2007. Growth hormone signalling: Sprouting links between pathways, human genetics and therapeutic options. Trends Endocrinol Metab., 18: 12-18.
32. Zhao, X.H., J.Y. Wang, G.X. Zhang, Y. Wei, Y.P. Gu and Y.B. Yu, 2012. Single nucleotide polymorphism in the *STAT5b* gene is associated with body weight and reproductive traits of the Jinghai Yellow chicken. Mol. Biol. Rep., 39: 4177-4183.