J Anim Reprod Biotechnol 2021;36:129-136
pISSN: 2671-4639 • eISSN: 2671-4663
https://doi.org/10.12750/JARB.36.3.129

JARB
Journal of Animal Reproduction and Biotechnology
Check for updates

Original Article

Investigation of single nucleotide polymorphism in *TSH-β* and *CaSR* associated with body weight in Korean native chickens (Gray Brown)

Dongyep Oh[1], Jae Jung Ha[1], Jun Koo Yi[1], Dae Hyun Kim[1], Seung Min Oh[1], Songmi Kim[2,3], Kyudong Han[2,3] and Yong-Soo Park[4,*]

[1]Gyeongsangbuk-Do Livestock Research institute, Yeongju 36052, Korea
[2]Center for Bio-Medical Engineering Core Facility, Dankook University, Cheonan 31116, Korea
[3]Department of Microbiology, Dankook University, Cheonan 31116, Korea
[4]Korea National College of Agriculture and Fisheries, Jeonju 54874, Korea

Received July 21, 2021
Revised August 24, 2021
Accepted August 30, 2021

***Correspondence**
Yong-Soo Park
E-mail: dvmpys@korea.kr

ORCID
https://orcid.org/0000-0002-1948-0919

ABSTRACT This study identified single nucleotide polymorphisms (SNPs) that affect the body weight of chickens. Analysis of body weight showed that the Cornish breed had the highest body weight, and the Korean native chicken (Gray Brown) had the lowest body weight. *TSH* is composed of an α-subunit and a β-subunit, and the *TSH-β* gene encoding the β-subunit has been reported to be associated with obesity. In chickens, it is located on chromosome 26 and is reported to be associated with growth. The calcium-sensing receptor gene (*CaSR*) plays a role in the regulation of extracellular calcium homeostasis and is responsible for calcium absorption in the urinary tract, which affects the eggshell quality in poultry. It was shown that *TSH-β* was strongly correlated with weight in Cornish and Korean native (Gray Brown) chickens, particularly in those with the CC trait. However, *CaSR* showed no association with body weight in poultry; it was associated with calcium and the eggshell. Thus, selection for *TSH-β* can be used to produce individuals with more favorable traits in terms of body weight.

Keywords: *CaSR,* Cornish, Korean native chicken (Gray Brown), SNP, *TSH-β*

INTRODUCTION

The Korean native chicken (KNC) has chewy meat due to its high collagen content. The meat contains abundant sulphur-containing amino acids, such as methionine and cystine, which impart flavor; it is also favored for its juiciness and tenderness.

Since the CBD Convention in 2004, Korea has registered 15 unique animal genetic resources, 120 breeds, or endogenous breeds in the Domestic Animal Diversity Information System (DAD-IS), an international breeding system. Forty species, including KNC, are registered (Food and Agriculture Organization of the United Nations, 2019). Since the start of the KNC restoration project, several studies have been conducted on this breed, and many meaningful results have been reported. Among these, representative studies on the genetic characteristics of KNCs include studies on the genetic composition and genetic diversity of the population using SNP markers, MS markers, or mtDNA D-loops (Hoque et al., 2009; Lee

et al., 2010; Cho et al., 2011; Lee et al., 2011; Cho et al., 2014; Seo et al., 2015a,b; Choi et al., 2017); hair color-related genetic mutations (Heo et al., 2011; Park et al., 2013; Choi et al., 2014); the frequency and developmental patterns of plumage fading (Sohn et al., 2012; Sohn et al., 2013; Bang et al., 2018); immune and stress responses (Jung et al., 2009; Hoque et al., 2011; Sohn et al., 2014; Sohn et al., 2015); and genetic parameters for productivity and economically favorable traits and practical production using breeding combinations (Sang et al., 2005; Park et al., 2011; Kang et al., 2012a,b; Kim et al., 2012a,b; Lee et al., 2013; Lee et al., 2014; Yoo et al., 2015; Jin et al., 2017).

Although there have been various studies on KNCs (Gray Brown), there has been no concrete report on their productivity or comparison of productivity between species. However, they are not economically viable because of their slow low growth rate, long breeding period, and low weight (Kwon et al., 1995; Park et al., 2010; Kang et al., 2012a,b). Therefore, improvement of growth-related traits would enhance the market value of KNCs.

Recently, extensive studies have been conducted for the development of molecular breeding techniques to improve growth-related traits in chickens. Genes such as growth hormone (*GH*) (Seo et al., 2001), growth hormone receptor (*GHR*), growth hormone secretagogue receptor (*GHSR*) (Zhang et al., 2009), insulin-like growth factor-I (*IGF-I*) (Li et al., 2009), insulin-like growth factor binding protein-2 (*IGFBP-2*) (Leng et al., 2009), insulin (*INS*) (Lei et al., 2007), leptin receptor (*LEPR*) (Choi et al., 2003), and thyroid-stimulating hormone beta subunit (*TSH-β*) (Lei et al., 2007) are associated with growth. The growth-related traits are manifested by the various actions of multiple organs.

The thyroid plays a very important role in growth by regulating hormone metabolism in the body through the secretion of thyroid hormone (*TH*) (Ga et al., 1999). A recent report showed that thyroidectomy reduces the growth rate significantly and results in excessive accumulation of fat in the neck, back, chest, and abdominal cavity (Ga et al., 1999). The manifestation and secretion of *TH* is induced by thyrotrophin, a thyroid stimulating hormone (*TSH*), which is composed of the α-subunit and β-subunit. The *TSH-β* gene, which encodes the β-subunit, is located on chromosome 1 and is related to obesity (Nie et al., 2005; Lei et al., 2007); it is also found on chromosome 26 and is related to growth (Lei et al., 2007).

The growth of male chickens involves various factors and hormones, including testosterone and TH (Boertje et al., 2019). These two hormones support growth. During the growth of *Emberiza bruniceps* feathers, thyroid activity is strongly influenced by gonadal hormones (Pati et al., 1986). Breeders often administer testosterone to poultry for faster growth, longer crowing, and better feather color. Further, the g.1031 G>C marker in the intron region is related to the weight of Cornish.

The calcium-sensing receptor gene (*CaSR*) plays a role in regulating extracellular homeostasis of calcium and secretion of hormones and absorption of calcium in the urinary system (Hough et al., 2004; Cifuentes et al., 2005; Pidasheva et al., 2005; Yun et al., 2007). Decreased calcium concentration in the body leads to the secretion of parathyroid hormones by the parathyroid gland, decreasing the secretion of calcium ions from the kidneys and promoting the absorption of calcium ions in the small intestine. It also stimulates the secretion of growth hormone by the pituitary gland (Hall et al., 1985) and enhances progesterone secretion (Veldhuis and Klase, 1982). These results suggest that the calcium concentration in the body has major effects on hormones related to growth and reproduction.

The *CaSR* gene has been extensively studied in recent years. The polymorphism in the A986S variation site in the *CaSR* gene regulates calcium concentration and is related to diseases such as osteoporosis, hypertension, and obesity (Felderbauer et al., 2005; Yun et al., 2007). *CaSR* is a homodimeric complex located in the cell membrane and belongs to the class C G-protein-coupled receptor (*GPCR*) (Jensen and Bräuner-Osborne, 2007). It can sense subtle changes in extracellular calcium concentration and thus mediate PTH secretion to maintain calcium homeostasis by regulating intestinal absorption, bone storage and exchange, and renal reabsorption (Conigrave, 2016).

In addition, *CaSR* mediates various physiological and pathophysiological processes, such as ion channel activity, gene expression, inflammation, proliferation, differentiation, and apoptosis by inducing downstream signaling cascades. Chicken *CaSR* is 79 and 84% homologous with human *CaSR* at the nucleotide and amino acid levels, respectively. In situ hybridization has revealed that *CaSR* is present in the parathyroid, kidney, brain, and small intestine (Diaz et al., 1997). Yarden et al. (2000) showed

that *CaSR* expression in the parathyroid gland is inversely associated with changes in plasma calcium concentration. Chickens fed vitamin D-deficient diets with low *CaSR* expression were characterized by the highest concentration of PTH, whereas high *CaSR* gene expression levels in vitamin-D-depleted chickens were associated with low PTH content in the parathyroid gland (Yarden et al., 2000). The results indicate that the functional *CaSR* in chickens possesses characteristics similar to those of mammalian *CaSR* and may play a significant role in avian calcium homeostasis.

The regulatory capacity of this gene on calcium concentration makes it a promising candidate for the improvement of economic traits.

The KNC is recognized for its value as a genetic resource; however, trait improvement is required to increase its value as an industrial resource. Specific genetic characteristics should be identified for such trait improvement. Thus, through the investigation and analysis of genetic variations in *TSH-β* and *CaSR* genes, this study aims to provide basic molecular breeding data for the production of KNC with improved growth traits.

MATERIALS AND METHODS

Animals and phenotypes

Genomic DNA was extracted from Cornish (n = 192), KNC (Gray Brown) (n = 192), and Rhode Island Red (n = 192) with different body weights (Table 1). In the breeding environment, the temperature inside the windowless poultry house was maintained at 20-25℃, and the humidity was approximately 70-85%. An incandescent light bulb (60 W) is installed for the lighting time, and an automatic lighting switchgear to automatically turn off the lights for an average of 8 h per day. Feed for poultry was fed freely. For the negative control, the poultry were provided with a free supply of water passed through an ultraviolet sterilizer (Dynamics, M600, USA).

SNP genotyping

TSH-β and *CaSR* genes were retrieved from the reference sequence data (accession no. AY341265, XM_416491). The c.319 G>T SNP is a non-synonymous SNP that is substituted from serine to alanine in the 963rd amino acid position. Primer sequences were designed using the Primer3 program, and their information is shown in Table 2. For genotyping of g.1031 G>C and c.319 G>T SNPs within the *TSH-β* and *CaSR* genes, the single base extension (SBE) method was performed according to the protocol of the SNaPshot ddNTP Primer Extension Kit (Applied Biosystems, Foster City, CA).

Primer extension reactions were performed using the SNaPshot dNTP Primer Extension Kit (Applied Biosystems, Foster City, CA, USA). For primer extension, exonuclase 1 and shrimp alkaline phosphatase (SAP) were added to the reaction mixtures. Samples were cultured at 37℃ for 1 h and then inactivated at 72℃ for 15 min. PCR products were analyzed using the Genescan 120 LIZ standard and

Table 1. Mean and standard deviations of body weight at different ages in chicken populations

Trait	Cornish	Korea Native Chicken (Gray Brown)	Rhode Island Red
WT8	1800 ± 124.4	618 ± 74.4	805 ± 100.8
WT12	2264 ± 187.9	918 ± 107.2	1179 ± 95.5
WT16	2857 ± 191.1	1122 ± 156.5	1506 ± 135.2

WT8, body weight at 56 days of age; WT12, body weight at 84 days of age; WT16, body weight at 112 days of age.

Table 2. Information on SNP markers associated with *TSH-β* and *CaSR*

Gene	SNP	Chromosome	Position		Sequence	Tm	Product size
TSH-β	g.1031 G>C	26	Intron	F[1]	CGTTGTTGCGGTAGTAGGTG	60	521
				R[2]	GCGTCAGGAAGAGCTCATTG		
				E[3]	TGTAGTCAGTGTAGGTGCTT		
CaSR	c.319 G>T	1	Exon[1]	F	CAGTGCGCTACCATTGAGTC	59	468
				R	TGACCCACAGTTGTAACCAGA		
				E	TAAGTGAGCTGTCTAAATTG		

[1]Forward primer sequence, [2]Reverse primer sequence, [3]Extension primer sequence.

HiDi formamide (Applied Biosystems, Foster City, CA), followed by denaturation at 95℃ for 5 min. Electrophoresis was performed using an ABI PRISM 3500XL Genetic Analyzer and analyzed using GeneMapper v.4.0 software (Applied Biosystems, Foster City, CA).

Statistical analyses

Associations between body weight and individual SNPs for the 576 samples were analyzed using the following mixed analysis of covariance (ANCOVA) model using SPSS v19.0 (SPSS Inc., Chicago, IL, USA).

$$Y_{ijk} = \mu + B_i + P_j + G_k + \beta age + e_{ijk}$$

where Y_{ijk} is a phenotype, μ is the overall mean, B_i is a breed effect (i = Cornish, Korea Native Chicken [Gray Brown]), P_j is the fixed effect of calving place j, G_k is a fixed effect of genotype k, β is a regression coefficient, age is a covariate for age in days at slaughter, and e_{ijk} is a random residual assumed to have independent and identical normal distribution.

RESULTS

Genotype and allele frequency

The primer sequences of *TSH-β* and *CaSR* genes were designed using the Primer3 program from the reference data (Accession no. AY341265, XM_416491) from NCBI, and their information is shown in Table 2. For genotyping g.1031 G>C and c.319 G>T within the *TSH-β* and *CaSR* genes, the SBE method was performed according to the protocol of the SNaPshot ddNTP Primer Extension Kit (Applied Biosystems, Foster City, CA).

As shown in Table 1, Cornish chickens showed overall higher body weight than the KNC (Gray Brown) and Rhode Island Red species. At 112 days of age, the Cornish chickens were 2,857 g, which is approximately twice as large as 1,122 g for KNC (Gray Brown). The genotype and allele frequencies of the two selected SNPs were calculated for each of the three chickens (Table 3). Genotype distribution for the g.1031 G>C SNP in each breed indicated that the CC genotype was present in the Cornish (23.9%) and KNC (Gray Brown) (11.9%), and the CC genotype was present in Rhode Island Red (0%). For the c.319 G>T SNP, the GT, and TT genotypes were identified in Cornish and KNCs (Gray Brown), except in Rhode Island Red.

Table 3. SNP frequency in each breed and candidate gene

Gene	SNP	Genotype and allele	Genotype and allele frequency (%)		
			Cornish	KR	RIR
TSH-β	g.1031 G>C	CC	0.239	0.119	-
		CG	0.518	0.584	-
		GG	0.243	0.297	1.000
CaSR	c.319 G>T	GG	0.345	0.120	1.000
		GT	0.217	0.650	-
		TT	0.438	0.230	-

Table 4. Association of g.1031 G>C and c.319 G>T SNPs with body weight in Cornish chickens

Gene	SNP	Traits (g)	Genotype LSMEAN ± SE			*p*-value
TSH-β	g.1031 G>C		CC	CG	GG	
		WT8	1915.75 ± 108.76[b]	1895.69 ± 118.47[ab]	1795.75 ± 108.76[a]	0.019
		WT12	2374.35 ± 187.27[b]	2169.79 ± 177.97[a]	2097.99 ± 167.27[a]	0.024
		WT16	3017.72 ± 210.17[b]	2987.91 ± 195.41[ab]	2796.12 ± 195.39[a]	0.005
CaSR	c.319 G>T		GG	GT	TT	
		WT8	1795.57 ± 112.15	1816.93 ± 126.59	1812.43 ± 109.55	0.849
		WT12	2247.35 ± 187.17	2279.65 ± 167.67	2198.65 ± 157.77	0.788
		WT16	2807.69 ± 187.68	2887.49 ± 197.78	2903.59 ± 177.68	0.874

WT8, body weight at 56 days of age; WT12, body weight at 84 days of age; WT16, body weight at 112 days of age.
[a,b]Means with different superscripts within the same column are significantly different ($p < 0.05$).

Table 5. Association of g.1031 G>C and c.319 G>T SNPs with body weight in Korean native chickens (Gray Brown)

Gene	SNP	Traits (g)	Genotype LSMEAN ± SE			*p*-value
TSH-β	g.1031 G>C		CC	CG	GG	
		WT8	661.84 ± 62.82[b]	651.94 ± 71.52[b]	588.74 ± 65.72[a]	0.001
		WT12	954.11 ± 94.27[b]	939.48 ± 101.17[b]	891.21 ± 89.37[a]	0.004
		WT16	1297.79 ± 142.38[b]	1127.24 ± 151.48[ab]	1091.57 ± 149.79[a]	0.019
CaSR	c.319 G>T		GG	GT	TT	
		WT8	618.74 ± 62.12	629.94 ± 92.58	601.14 ± 78.59	0.785
		WT12	932.49 ± 99.87	912.59 ± 104.87	929.19 ± 90.87	0.984
		WT16	1219.64 ± 141.98	1127.91 ± 121.58	1297.47 ± 101.71	0.721

WT8, body weight at 56 days of age; WT12, body weight at 84 days of age; WT16, body weight at 112 days of age.
[a,b]Means with different superscripts within the same column are significantly different ($p < 0.05$).

Effect of SNP genotype in body weight

We analyzed body weight depending on the g.1031 G>C and c.319 G>T SNPs in three chicken breeds. The g.1031 G>C SNP was associated with a variety of body weights in Cornish and KNCs (Gray Brown) ($p < 0.05$, Tables 3 and 4). In particular, the g.1031 G>C SNP was associated with all the phenotypes at 56 days of age, 84 days of age, and 112 days of age. For Rhode Island red, only one genotype (GG) was identified and excluded from the analysis.

As shown in Table 4, body weight in the g.1031 G>C SNP was observed to be higher at 1,915 g, 2,374 g, and 3,017 g in the genotypes of CC ($p < 0.05$). A similar trend was found in the KNC (Gray Brown), showing a significant difference of 661 g, 954 g, and 1,297 g compared to the CC genotype (Table 5). In the *CaSR* gene, it was confirmed that there was no significant difference in body weight because it is a gene that affects calcium or eggshell.

DISCUSSION

The thyroid is an organ that plays an important role in growth by regulating hormones in the body. Thyroid-stimulating hormone (*TSH*), a hormone that stimulates the thyroid, is secreted by the anterior pituitary. This consists of the α-subunit and β-subunit; the α-subunit has amino acid sequences, which correspond to gonadotropins such as *LH, FSH,* and *hCG*.

Various characteristics of the β-subunit have been reported to influence the specific activity of *TSH* (Cho, 2007). The *TSH-β* gene in chickens is located in the 3.9 Mbp area of chromosome 26, and QTL D/B information displays various weight-related QTLs (http://www.animalgenome.org). According to a report by Nadaf et al. (2009), a QTL related to body weight at 84 days of age is located in the 2 to 4 Mbp area of chromosome 26, and this QTL area includes the *TSH-β* gene (3.9 Mbp). Nearby, there is also a QTL (2.4 to 3.4 Mbp) related to body weight at 56 days of age.

Therefore, it is considered that the *TSH-β* gene has a very close connection with growth traits. There are many different genes and genetic variations in specific traits, and these genetic structures have diverse effects on specific traits. Moreover, genetic structures of genetic variations can be identified through linkage disequilibrium (LD) analysis, and the characteristics of this LD block structure can be classified depending on the variety (Gautier et al., 2007; Amaral et al., 2008; Megens et al., 2009).

When analyzing the variation region of the *TSH-β* gene in this study, genetic characteristics depending on the varieties were revealed. It was identified that all the analyzed individuals of Rhode Island Red had a GG genotype and that Cornish and KNC had a significant correlation ($p < 0.05$) between genotype and weight.

According to a report by Seo et al. (2013), g.1031 G>C applied in this study was measured by weight. Further, comparative analysis showed a correlation with body weight only at the age of 150 days in Cornish chickens, and there was no correlation in conventional chickens. However, based on the results, Cornish and native chickens showed a strong correlation between the g.1031 G>C marker and body weight. Although the correlation analysis results for the Cornish chickens were the same, different results were obtained for conventional chickens in this study as these were KNCs (Gray Brown) restored in Gyeongsangbuk-do. Although it can be broadly classified as the same breed as conventional chickens, the phenotype and genetic characteristics of the native chicken breed used in this study are different from those of the

native chicken provided by the National Institute of Livestock Science. Further, in the previous studies analyses were carried out at 150 days and 270 days. In this study, analyses were carried out at 56 days, 84 days, and 112 days; this may have contributed to the difference in the results obtained.

It is believed that the *TSH-β* gene, particularly the g.1031 G>C variation region, is closely correlated with growth traits. Furthermore, it is anticipated that G, an allele, can be used as a biological marker for the selection of growth-related and other economically favorable traits. Although *TSH-β* is a gene closely linked with growth traits, there have not been many studies on its economic use. Therefore, varied approaches and studies on the utilization of *TSH-β* are expected to play a crucial role in the improvement of economic traits in KNCs.

CONFLICTS OF INTEREST

No potential conflict of interest relevant to this article was reported.

ACKNOWLEDGEMENTS

This study was performed with the support of the Cooperative Research Program for Agriculture Science & Technology Development (Project No. PJ015996012021), Rural Development Administration, Republic of Korea.

AUTHOR CONTRIBUTIONS

Author contribution: DO, JJH, JKY, DHK, SMO, SK, KH, YSP
Conceptualization: DO, YSP
Data curation: DO, SK
Formal analysis: DO, KH
Funding acquisition: DO
Investigation: DO, JJH, JKY, DHK, SMO
Methodology: DO, KH
Project administration: KH, YSP
Resources: DO, KH
Software: SK, KH
Supervision: KH, YSP
Validation: SK
Visualization: SK
Writing - original draft: DO, KH, YSP
Writing - review & editing: DO, KH, YSP

AUTHOR'S POSITION AND ORCID NO.

Oh D, Researcher,
https://orcid.org/0000-0003-4412-7719
Ha JJ, Researcher,
https://orcid.org/0000-0001-6785-6346
Yi JK, Researcher,
https://orcid.org/0000-0003-2593-6529
Kim DH, Researcher,
https://orcid.org/0000-0002-4820-4438
Oh SM, Researcher,
https://orcid.org/0000-0001-8848-8028
Kim S, Researcher,
https://orcid.org/0000-0002-5497-1174
Han K, Professor,
https://orcid.org/0000-0001-6791-2408
Park YS, Professor,
https://orcid.org/0000-0002-1948-0919

REFERENCES

Amaral AJ, Megens HJ, Crooijmans RP, Heuven HC, Groenen MA. 2008. Linkage disequilibrium decay and haplotype block structure in the pig. Genetics 179:569-579.

Bang MH, Cho EJ, Cho CY, Sohn SH. 2018. Study on the characteristics of feather developing pattern and morphology in early- and late-feathering Korean native chickens. Korean J. Poult. Sci. 45:155-165.

Boertje ET, Snyder NM, Reed WL, Kittilson JD, Clark ME. 2019. Testosterone and triiodothyronine in Franklin's Gull (*Leucophaeus pipixcan*) eggs. Waterbirds 42:251-260.

Cho CY, Lee PY, Ko YG, Kim HK, Park MN, Yeon SH. 2011. Multiple maternal origins of Korean native chicken based on the mtDNA D-loop variation. Korean J. Poult. Sci. 38:5-12.

Cho ES, Chung WH, Choi JW, Jang HJ, Park MN, Kim N, Kim TH, Lee KT. 2014. Genome-wide copy number variation in a Korean native chicken breed. Korean J. Poult. Sci. 41:305-311.

Cho YW. 2007. Clinical implication of serum TSH concentration. J. Korean Endocr. Soc. 22:87-94.

Choi BH, Kim TH, Cho YM, Lee HY, Jeon JT, Cheong IC. 2003. Association study between porcine LEPR-derived microsatellite polymorphisms and economic traits. J. Anim. Sci. Technol. 45:679-688.

Choi ES, Bang MH, Kim KG, Kwon JH, Chung OY, Sohn SH. 2017. Production performances and heterosis effects of Korean native chicken breed combinations by diallel crossing test. Korean J. Poult. Sci. 44:123-134.

Choi JA, Lee JH, Jang HJ, Lee KT, Kim TH, Lee HJ, Heo KN, Kim CD, Han JY, Park MN. 2014. Genetic variations of chicken TYR gene and associations with feather color of Korean native chicken (KNC). Korean J. Poult. Sci. 41:7-14.

Cifuentes M, Albala C, Rojas C. 2005. Calcium-sensing receptor expression in human adipocytes. Endocrinology 146:2176-2179.

Conigrave AD. 2016. The calcium-sensing receptor and the parathyroid: past, present, future. Front. Physiol. 7:563.

Diaz R, Hurwitz S, Chattopadhyay N, Pines M, Yang Y, Kifor O, Einat MS, Butters R, Hebert SC, Brown EM. 1997. Cloning, expression, and tissue localization of the calcium-sensing receptor in chicken (Gallus domesticus). Am. J. Physiol. 273(3 Pt 2):R1008-R1016.

Felderbauer P, Hoffmann P, Klein W, Bulut K, Ansorge N, Epplen JT, Schmitz F, Schmidt WE. 2005. Identification of a novel calcium-sensing receptor gene mutation causing familial hypocalciuric hypercalcemia by single-strand conformation polymorphism analysis. Exp. Clin. Endocrinol. Diabetes 113:31-34.

Food and Agriculture Organization of the United Nations. 2019. Domestic Animal Diversity Information System. http://www. fao.org/dad-is/browse-by-country-and-species/en/. Accessed 20 December 2019.

Ga CH, Kim CH, Chae BJ, Rhee YC. 1999. Effect of dietary thyroid hormone on growth performance, body composition, serum thyroid hormone concentration and energy metabolism of broiler chicks. Korean J. Poult. Sci. 26:237-245.

Gautier M, Faraut T, Moazami-Goudarzi K, Navratil V, Foglio M, Grohs C, Boland A, Garnier JG, Boichard D, Lathrop GM, Gut IG, Eggen A. 2007. Genetic and haplotypic structure in 14 European and African cattle breeds. Genetics 177:1059-1070.

Hall TR, Lam SK, Harvey S. 1985. Calcium control of growth hormone release from chicken pituitary glands in vitro. Gen. Comp. Endocrinol. 60:70-74.

Heo KN, Choo HJ, Seo BY, Park MN, Jung KC, Hwangbo J, Kim HK, Hong EC, Seo OS, Kang BS. 2011. Investigation of TYR and MC1R polymorphisms in Korean native chickens and the commercial chickens. CNU J. Agric. Sci. 38:465-471.

Hoque MR, Jung KC, Park BK, Choi KD, Lee JH. 2009. Genetic variability of mtDNA D-loop region in Korean native chickens. Korean J. Poult. Sci. 36:323-328.

Hoque MR, Lee SH, Jung KC, Kang BS, Park MN, Lim HK, Choi KD, Lee JH. 2011. Discrimination of Korean native chicken populations using SNPs from mtDNA and MHC polymorphisms. Asian Australas. J. Anim. Sci. 24:1637-1643.

Hough TA, Bogani D, Cheeseman MT, Favor J, Nesbit MA, Thakker RV, Lyon MF. 2004. Activating calcium-sensing receptor mutation in the mouse is associated with cataracts and ectopic calcification. Proc. Natl. Acad. Sci. U. S. A. 101:13566-13571.

Jensen AA and Bräuner-Osborne H. 2007. Allosteric modulation of the calcium-sensing receptor. Curr. Neuropharmacol. 5:180-186.

Jin S, Jayasena DD, Jo C, Lee JH. 2017. The breeding history and commercial development of the Korean native chicken. Worlds Poult. Sci. J. 73:163-174.

Jung KC, Hoque MR, Seo DW, Park BK, Choi KD, Lee JH. 2009. Genotype analysis of the major histocompatibility complex region in Korean native chicken. Korean J. Poult. Sci. 36:317-322.

Kang BS, Choo HJ, Kim HK, Kim CD, Heo KN, Hwangbo J, Suh OS, Choi HC, Hong EC. 2012a. Performance of laying period of two-way crossbreed parent stock Korean native chickens for producing of Korean native commercial chickens. Korean J. Poult. Sci. 39:133-141.

Kang BS, Kim HK, Kim CD, Heo KN, Choo HJ, Hwangbo J, Suh OS, Hong EC. 2012b. Performance of growing period of two-crossbreed parent stock Korean native chickens for producing of Korean native commercial chickens. Korean J. Poult. Sci. 39:71-76.

Kim CD, Choo HJ, Kang BS, Kim HK, Heo KN, Lee MJ, Son BR, Suh OS, Choi HC, Hong EC. 2012a. Performance of laying period of two-way crossbreed parent stock to produce laying-type Korean native commercial chickens. Korean J. Poult. Sci. 39:245-252.

Kim YS, Kim JH, Suh SW, Kim H, Byun MJ, Kim MJ, Kim JS, Lee JW, Choi SB. 2012b. Comparison of growth performance between Korean native layer chickens and imported layer chickens at early rearing stage. Korean J. Poult. Sci. 39:283-290.

Kweon YJ, Yeo JS, Sung SK. 1995. Quality characteristics of Korean native chicken meat. Korean J. Poult. Sci. 22:223-231.

Lee HK, Oh JD, Park CH, Lee KW, Lee JH, Jeon GJ, Kong HS. 2010. Comparison for genetic diversity between Korean native commercial chicken brand groups using microsatellite markers. Korean J. Poult. Sci. 37:355-360.

Lee MJ, Heo KN, Choi HC, Hong EC, Kim CD. 2014. The performance test in crossbreds of Korean native chickens for the establishment of new lines. Korean J. Poult. Sci. 41:39-44.

Lee MJ, Kim SH, Heo KN, Kim HK, Choi HC, Hong EC, Choo HJ, Kim CD. 2013. The study on productivity of commercial Korea chickens for crossbred Korean native chickens. Korean J. Poult. Sci. 40:291-297.

Lee P, Yeon SH, Kim JH, Ko YG, Son JK, Lee HH, Cho CY. 2011. Genetic composition of Korean native chicken populations - national scale molecular genetic evaluation based on microsatellite markers. Korean J. Poult. Sci. 38:81-87.

Lei M, Luo C, Peng X, Fang M, Nie Q, Zhang D, Yang G, Zhang X. 2007. Polymorphism of growth-correlated genes associated with fatness and muscle fiber traits in chickens. Poult. Sci. 86:835-842.

Leng L, Wang S, Li Z, Wang Q, Li H. 2009. A polymorphism in the 3'-flanking region of insulin-like growth factor binding protein 2 gene associated with abdominal fat in chickens. Poult. Sci. 88:938-942.

Li W, Li F, Li D. 2009. IGF-1 gene polymorphism and weight-related analysis. Int. J. Biol. 1:113-118.

Megens HJ, Crooijmans RP, Bastiaansen JW, Kerstens HH, Coster A, Jalving R, Vereijken A, Silva P, Muir WM, Cheng HH, Hanotte O, Groenen MA. 2009. Comparison of linkage disequilibrium and haplotype diversity on macro- and microchromosomes in chicken. BMC Genet. 10:86.

Nadaf J, Pitel F, Gilbert H, Duclos MJ, Vignoles F, Beaumont C, Vignal A, Porter TE, Cogburn LA, Aggrey SE, Simon J, Le Bihan-Duval E. 2009. QTL for several metabolic traits map to

loci controlling growth and body composition in an F2 intercross between high- and low-growth chicken lines. Physiol. Genomics 38:241-249.

Nie Q, Lei M, Ouyang J, Zeng H, Yang G, Zhang X. 2005. Identification and characterization of single nucleotide polymorphisms in 12 chicken growth-correlated genes by denaturing high performance liquid chromatography. Genet. Sel. Evol. 37:339-360.

Park MN, Hong EC, Kang BS, Hwangbo J, Kim HK. 2011. Performance and meat quality of three-crossbreed Korean native chickens (KNC). Korean J. Poult. Sci. 38:293-304.

Park MN, Hong EC, Kang BS, Kim HK, Seo BY, Choo HJ, Na SH, Seo OS, Han JY, Hwangbo J. 2010. The study on production and performance of crossbred Korean native chickens (KNC). Korean J. Poult. Sci. 37:347-354.

Park MN, Kim TH, Lee HJ, Choi JA, Heo KN, Kim CD, Choo HJ, Han JY, Lee T, Lee JH, Lee KT. 2013. Genetic variations of chicken MC1R gene and associations with feather color of Korean native chicken (KNC) 'Woorimatdag'. Korean J. Poult. Sci. 40:139-145.

Pati AK and Pathak VK. 1986. Thyroid and gonadal hormones in feather regeneration of the redheaded bunting, *Emberiza bruniceps.* J. Exp. Zool. 238:175-181.

Pidasheva S, Canaff L, Simonds WF, Marx SJ, Hendy GN. 2005. Impaired cotranslational processing of the calcium-sensing receptor due to signal peptide missense mutations in familial hypocalciuric hypercalcemia. Hum. Mol. Genet. 14:1679-1690.

Raisz LG. 1981. Calcium regulation. Clin. Biochem. 14:209-212.

Sang BD, Choi CH, Kim HK, Kim SD, Jang BG, Na JC, Yu DJ, Lee SJ, Sang BC, Lee JH. 2005. Estimation of breeding values for economic traits in Korean native chicken. Korean J. Poult. Sci. 32:231-237.

Seo DS, Kang WJ, Yun JS, Lee JH, Hong KC, Ko Y. 2001. Association of serum insulin-like growth factor-1 and egg productivity with growth hormone gene polymorphism in Korean Native Ogol Chicken. J. Anim. Sci. Technol. 43:303-314.

Seo DW, Park HB, Choi NR, Jin S, Yoo CK, Sultana H, Heo KN, Jo C, Lee JH. 2015a. Construction of genetic linkage map using microsatellite and SNP markers in Korean native chicken. Korean J. Poult. Sci. 42:77-86.

Seo J, Oh JD, Choi EJ, Lim HK, Seong J, Song KD, Lee JH, Lee HK, Kong HS, Jeon GJ, Shon YG, Choi KD. 2013. Effects of SNP in TSH-β gene of chicken on economic traits. Korean J. Poult. Sci. 40:115-120.

Seo JH, Oh JD, Lee JH, Seo D, Kong HS. 2015b. Studies on genetic diversity and phylogenetic relationships of Korean native chicken using the microsatellite marker. Korean J. Poult. Sci. 42:15-26.

Sohn SH, Cho EJ, Park DB, Jang IS, Moon YS. 2014. Comparison of stress response between Korean Native Chickens and Single Comb White Leghorns subjected to a high stocking density. Korean J. Poult. Sci. 41:115-125.

Sohn SH, Cho EJ, Park JA, Hong YH, Kim CD. 2015. Analysis of stress response of domestic chicken breeds for the development of a new synthetic parent stock. Korean J. Poult. Sci. 42:157-167.

Sohn SH, Kim NY, Park DB, Song HR, Cho EJ, Choi SB, Heo KN, Choi HC. 2013. Influence of early- and late-feathering phenotype on productive performance in the feather-sexing strains of Korean native chicken. Korean J. Poult. Sci. 40:263-270.

Sohn SH, Park DB, Song HR, Cho EJ, Kang BS, Suh OS. 2012. Genotype frequencies of the sex-linked feathering and their phenotypes in domestic chicken breeds for the establishment of auto-sexing strains J. Anim. Sci. Technol. 54:267-274.

Veldhuis JD and Klase PA. 1982. Calcium ions modulate hormonally stimulated progesterone production in isolated ovarian cells. Biochem. J. 202:381-386.

Yarden N, Lavelin I, Genina O, Hurwitz S, Diaz R, Brown EM, Pines M. 2000. Expression of calcium-sensing receptor gene by avian parathyroid gland in vivo: relationship to plasma calcium. Gen. Comp. Endocrinol. 117:173-181.

Yoo J, Koo B, Kim E, Heo JM. 2015. Comparison of growth performance between crossbred Korean native chickens for hatch to 28 days. CNU J. Agric. Sci. 42:23-27.

Yun FH, Wong BY, Chase M, Shuen AY, Canaff L, Thongthai K, Siminovitch K, Hendy GN, Cole DE. 2007. Genetic variation at the calcium-sensing receptor (CASR) locus: implications for clinical molecular diagnostics. Clin. Biochem. 40:551-561.

Zhang B, Chen H, Guo Y, Zhang L, Zhao M, Lan X, Zhang C, Pan C, Hu S, Wang J, Lei C. 2009. Associations of polymorphism within the GHSR gene with growth traits in Nanyang cattle. Mol. Biol. Rep. 36:2259-2263.