

# Comparison of body size and reproductive hormones in high- and low-yielding Wulong geese

Jingjing Liu,<sup>\*,1</sup> Demin Zhang,<sup>†,1</sup> Zhenwei Zhang,<sup>\*</sup> Wenqiong Chai,<sup>\*</sup> Jiqing Zhang,<sup>\*</sup> Mengmeng Li,<sup>\*</sup> Yun Wang,<sup>‡</sup> Shuer Zhang,<sup>†</sup> and Mingxia Zhu<sup>\*,2</sup>

<sup>\*</sup>College of Agronomy of Liaocheng University, Liaocheng 252000, China; <sup>†</sup>Shandong Animal Husbandry General Station, Jinan 250010, China; and <sup>‡</sup>Liaocheng Academy of Agricultural Sciences, Liaocheng 252000, China

**ABSTRACT** Wulong geese are renowned for their egg-laying and reproductive abilities. This work investigated the potential of using body size traits in the selection and breeding of high-yielding Wulong geese. A total of forty 479-day-old female geese (high-yielding geese,  $n = 20$ ; low-yielding geese,  $n = 20$ ) were selected to evaluate the relationship between body weight, body size trait indicators, serum reproductive hormones, and biochemical indicators. The results showed that serum estradiol (**E2**), glucose (**GLU**), and triglyceride (**TG**) concentrations, together with pubic spacing and abdominal circumference were significantly higher in high-yielding geese ( $P < 0.01$ ), whereas the opposite was true

for neck circumference, neck length, and tibial circumference. In addition, the serum testosterone (**T**) concentration and body weight were higher in high-yielding geese ( $P < 0.05$ ). Neck circumference and neck length were negatively correlated with E2 and TG ( $P < 0.01$ ); while pubic spacing and abdominal circumference were positively correlated with E2, GLU, and TG ( $P < 0.01$ ), the highest correlation coefficient was 0.777 between TG and pubic spacing; T was also strongly associated with neck circumference ( $P < 0.01$ ). In conclusion, high-yielding Wulong geese can be selected through neck circumference, neck length, pubic spacing, and abdominal circumference.

**Key words:** Wulong goose, body size, reproductive hormone, biochemical indicator, correlation

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## INTRODUCTION

The Wulong goose, also known as the Huoyan goose, is one of the smaller varieties of the Chinese white goose breed. Wulong geese originate in Laiyang, Shandong Province, China, and the name derives from the Wulong River that runs through the area. The Wulong goose is a native breed that tolerates poor feed and cold temperatures, has high egg-laying rates and is extremely adaptable (Min et al., 2007). Generally, the useful breeding life of female geese is 3 to 4 yr, with less egg production in the first year, increasing to a maximum in the second and third years, and declining after the fourth year (Wang, 2020). It is regarded as a national treasure by the Chinese goose industry. Even on a global scale, Wulong geese are renowned for their outstanding egg-

laying and reproductive performance (Chen et al., 2004; Luan et al., 2013).

Two methods are commonly used to select high-laying geese: selection according to physical appearance and physiological characteristics and selection according to their production. The present study was undertaken to investigate the selection of high-yielding geese based on their physical appearance, with applications to breeding farms that provide commercial geese.

Endocrine and environmental factors such as photoperiod length and the amount of feeding can affect egg production (Lewis and Gous, 2006). Endocrine studies have identified several genes associated with egg-laying performance; these genes are mainly related to reproductive hormones (Ismail et al., 2000; Kuo et al., 2005; Syed et al., 2005; Schmahl et al., 2008; Mcderment et al., 2012; Qin et al., 2015a). Egg production in chickens is regulated by a variety of reproductive endocrine factors and hormones. These hormones are mainly involved in the hypothalamic-pituitary-gonadal axis (**HPG**), which is closely related to egg-laying in chickens (Feng et al., 1997; Onagbesan et al., 2006; Chen et al., 2007; Bhattacharya et al., 2011; Qin et al., 2015b). However, little is known about the association

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<sup>1</sup>These contributed equally to this study and share first authorship.

<sup>2</sup>Corresponding author: [zhumingxia@lcu.edu.cn](mailto:zhumingxia@lcu.edu.cn)

between body size and reproductive hormones. In this study, we selected high (high-yielding geese, **HYG**) and low (low-yielding geese, **LYG**) geese in terms of their egg production, compared the body size and reproductive hormones of the two groups in the late egg-laying period, and analyzed the correlation between body size and reproductive hormones. Our aim was to investigate the use of body size phenotypic traits for the early selection of HYG, thus contributing to the selection and breeding of the Wulong goose.

## MATERIALS AND METHODS

### Animals

Based on the individual egg-laying records, twenty Wulong geese (479 days old) from HYG and LYG groups were selected. All the geese were provided by Laiyang Wulong Goose Technology Development Co., Laiyang, China.

### Measurement of Weight and Body Size

The animal handling and sampling protocols were approved by the Animal Care and Use Committee of Liaocheng University. Seven body measurement parameters were recorded, namely, body weight, neck circumference, neck length, pubic spacing, tibial length, tibial circumference, and abdominal circumference.

- Neck length: the distance between the first cervical vertebra and the last cervical vertebra.
- Neck circumference: the circumference of the middle of the neck.
- Tibial length: the straight-line distance between the superior tibial joint and the third and fourth toes.
- Tibial circumference: the circumference of the middle of the shin.
- Abdominal circumference: the diameter at the widest point perpendicular to the abdominal midline.
- Pubic spacing: the distance between the two pubic bones.

### Measurement of Reproductive Hormones and Biochemical Parameters

Blood samples were collected from the geese using 5 mL vacuum tubes. The tubes were centrifuged at 3,500 rpm for 15 min and the serum supernatant (1 mL)

was removed and stored at  $-20^{\circ}\text{C}$ . Serum follicle-stimulating hormone (**FSH**), luteinizing hormone (**LH**), prolactin (**PRL**), progesterone (**P**), testosterone (**T**), and estradiol (**E2**) concentrations were determined by radioimmunoassay by the Beijing Northern Institute of Biotechnology (Beijing, China) using the XH6080 exonerator. The glucose (**GLU**) concentration was determined by the glucose oxidase method and the triglyceride (**TG**) concentrations by the GPO-PAP method, both using a Toshiba 120 automatic biochemistry instrument (Tokyo, Japan).

### Statistical Analysis

Data were analyzed by *t* test using SPSS Statistics software (version 26.0; IBM Corp., Armonk, NY) and expressed as means  $\pm$  standard deviation (**SD**). Differences were considered highly significant or significant at  $P \leq 0.01$  or  $P \leq 0.05$ , respectively. The relationships between body size and reproductive hormones were evaluated using Pearson correlation coefficients.

## RESULTS

### Weight and Body Size Parameters of the Geese

Table 1 shows the body weight and size parameters of the HYG and LYG. The pubic spacing and abdominal circumference of the HYG were significantly higher than those of the LYG ( $P < 0.01$ ), whereas the opposite was true for neck circumference, neck length, and tibial circumference. The weights of the HYG were higher than those of the LYG ( $P < 0.05$ ). No significant difference ( $P > 0.05$ ) was recorded between HYG and LYG for tibial length.

### Serum reproductive Hormones and Biochemical Parameters in Geese

The serum concentrations of the reproductive hormones and biochemical indicators of the HYG and LYG groups are shown in Table 2. Serum E2, GLU, and TG levels were all significantly higher in the HYG compared with the LYG ( $P < 0.01$ ). The serum T concentration was significantly higher ( $P < 0.05$ ) in the HYG while serum FSH, LH, PRL, and P concentrations were similar between the two groups.

**Table 1.** Body weights and body size indices in Wulong geese.

	Weight (kg)	Neck circumference (cm)	Neck length (cm)	Pubic spacing (mm)	Tibial length (cm)	Tibial circumference (cm)	Abdominal circumference (cm)
LYG	$3.06 \pm 0.21^a$	$9.61 \pm 0.36^B$	$26.15 \pm 2.01^B$	$40.68 \pm 8.11^A$	$10.15 \pm 0.80$	$5.00 \pm 0.29^B$	$18.01 \pm 2.45^A$
HYG	$3.27 \pm 0.29^b$	$8.94 \pm 0.43^A$	$23.39 \pm 2.59^A$	$67.35 \pm 6.00^B$	$10.17 \pm 0.61$	$4.57 \pm 0.31^A$	$22.77 \pm 1.41^B$

Abbreviations: HYG, High yielding geese; LYG, Low yielding geese.

Upper case (A, B) letters in the same column indicate highly significant differences ( $P < 0.01$ ), lower case (a, b) letters indicate significant differences ( $P < 0.05$ ), and no letters indicates non-significant differences ( $P > 0.05$ ).

**Table 2.** Reproductive hormones and biochemical indicators in Wulong geese.

	FSH (mIU/mL)	LH (mIU/mL)	PRL (uIU/mL)	P (ng/mL)	T (ng/mL)	E2 (pg/mL)	GLU (mmol/l)	TG (mmol/l)
LYG	2.40 ± 0.66	3.41 ± 1.70	45.37 ± 14.28	0.25 ± 0.12	0.17 ± 0.75 <sup>a</sup>	1.01 ± 0.28 <sup>A</sup>	13.44 ± 1.40 <sup>A</sup>	1.80 ± 0.84 <sup>A</sup>
HYG	2.41 ± 0.55	2.53 ± 1.06	49.51 ± 30.86	0.25 ± 0.10	0.28 ± 0.16 <sup>b</sup>	1.93 ± 0.50 <sup>B</sup>	16.93 ± 3.79 <sup>B</sup>	9.05 ± 3.24 <sup>B</sup>

Abbreviations: FSH, follicle stimulating hormone; E2, estradiol; GLU, Glucose; HYG, High yielding geese; LH, luteinizing hormones; LYG, Low yielding geese; PRL, prolactin; P, progesterone; T, testosterone; TG, triglyceride.

Upper case (A, B) letters in the same column indicate highly significant differences ( $P < 0.01$ ), lower case (a, b) letters indicate significant differences ( $P < 0.05$ ), and no letters indicates non-significant differences ( $P > 0.05$ ).

### Correlations Between Body Size and Reproductive Hormones in Geese

The correlations between body size and reproductive hormones in Wulong geese are shown in Table 3. Except for the tibial length, there was a strong correlation between pubic spacing and all other body measurements ( $P < 0.01$ ) with the highest correlation coefficient of 0.773 between pubic spacing and abdominal circumference. The neck circumference and neck length were both positively correlated with tibial circumference, and negatively correlated with abdominal circumference ( $P < 0.01$ ). The neck circumference was positively correlated with neck length ( $P < 0.05$ ) while the abdominal circumference was significantly associated with both the body weight and tibial circumference ( $P < 0.05$ ).

E2 was strongly associated with both T and TG ( $P < 0.01$ ). GLU was strongly correlated with LH and TG ( $P < 0.01$ ), while LH was negatively associated with E2 and TG ( $P < 0.05$ ). PRL levels were significantly related to both FSH and P ( $P < 0.05$ ), while T was positively correlated with TG ( $P < 0.05$ ).

Apart from the tibial length, there was a strong correlation between TG and all other body measurements ( $P < 0.01$ ), with the highest correlation coefficient (0.777) between TG levels and pubic spacing. E2 was strongly associated with the neck circumference, neck length, pubic spacing, and abdominal circumference ( $P < 0.01$ ); and significantly associated with the tibial circumference ( $P < 0.05$ ). There was a highly significant correlation between GLU levels and both pubic spacing, and abdominal circumference ( $P < 0.01$ ). T was strongly associated with neck circumference ( $P < 0.01$ ); and significantly associated with neck length ( $P < 0.05$ ), while LH was significantly associated with the neck circumference, neck length, and abdominal circumference ( $P < 0.05$ ).

## DISCUSSION

### Analysis of Results of Body Weight and Body Size Trait Measurements

The goose body size is an important indicator of their growth and development and is closely related to their egg production and reproductive performance. Here, we found a significant weight difference between HYG and LYG ( $P < 0.05$ ), probably because the LYG were in the feathering stage, resulting in a lower weight than the HYG. The neck circumferences and neck lengths of

the HYG were lower than those of the LYG while the pubic spacing, tibial circumference, and abdominal circumference were higher in the HYG group. A previous study has shown that low-yield ducks had thick and short necks, while high-yield ducks had wide pubic spacing and large and soft abdomens (Cheng, 2006). In the present study, we observed longer neck lengths in the LYG; this inconsistency may be due to species differences between the duck and the goose and will be investigated in more in-depth studies in the future. The neck circumference, neck length, pubic spacing, tibial circumference, and abdominal circumference were significantly different between the HYG and LYG ( $P < 0.01$ ), indicating significant differences in body size traits and suggesting that these differences could be used for selection in the breeding process.

### Analysis of the Results of Reproductive Hormone and Biochemical Parameters

Zhang et al. (2018) found that FSH, E2, and P levels were significantly higher in the high-yielding group than in the low-yielding group, while the levels of LH and T did not differ significantly between the two groups. In the present study, the serum T, E2, GLU, and TG concentrations were found to be higher in the HYG, while no significant differences in the serum FSH, LH, PRL, and P concentrations were found between the two groups. The results for the T, FSH, and P concentrations are inconsistent with the findings of Zhang et al, this may be related to breed differences and differences in the egg-laying period.

Fu (2000) reported that higher E2 levels are one of the main endocrine factors causing higher performance in Shaoxing ducks. Female goose estrogens (largely estradiol and estriol) are mainly secreted by follicular endometrial cells and follicular granulosa cells. The main physiological roles of these hormones are to promote the development of the female reproductive organs, as well as follicular maturation and the release of ovulation hormones. They also increase the plasma lipid, calcium, and protein contents, which provide the raw materials for egg formation (Wei, 2016). TG is a component of the blood fat and its level reflects the absorption and metabolism of lipids and their utilization, with lower values indicating higher utilization of fat (Chen et al., 2013). The serum GLU concentration is an indicator of the body's energy metabolism, reflecting the production and consumption of sugar. In this study, TG was positively

**Table 3.** Correlations between body size and reproductive hormones in Wulong geese.

	Weight	Neck circumference	Neck length	Pubic spacing	Tibial length	Tibial circumference	Abdominal circumference	FSH	LH	PRL	P	T	E2	GLU	TG
Weight	1														
Neck circumference	0.007	1													
Neck length	-0.178	0.379*	1												
Pubic spacing	0.442**	-0.615**	-0.495**	1											
Tibial length	0.181	0.271	0.133	-0.083	1										
Tibial circumference	0.170	0.606**	0.451**	-0.500**	0.101	1									
Abdominal circumference	0.379*	-0.432**	-0.533**	0.773**	0.115	-0.399*	1								
FSH	-0.074	-0.099	0.235	-0.012	0.223	-0.075	-0.023	1							
LH	-0.110	0.360*	0.369*	-0.240	-0.053	0.290	-0.424**	0.148	1						
PRL	0.063	0.043	0.128	-0.059	0.081	-0.054	0.082	0.315*	0.229	1					
P	-0.178	0.037	-0.059	-0.128	0.097	-0.105	0.034	0.002	0.003	0.356*	1				
T	0.027	-0.404**	-0.341*	0.205	-0.005	-0.245	0.221	0.066	-0.209	-0.261	0.031	1			
E2	0.162	-0.529**	-0.408**	0.440**	0.075	-0.377*	0.453**	0.035	-0.363*	-0.267	-0.059	0.851**	1		
GLU	0.281	-0.302	-0.270	0.573**	-0.195	-0.184	0.497**	-0.073	-0.405**	-0.018	-0.051	-0.051	0.119	1	
TG	0.459**	-0.492	-0.515**	0.777**	-0.076	-0.458**	0.713**	-0.039	-0.325*	0.176	0.046	0.319*	0.522**	0.680**	1

Abbreviations: E2, estradiol; FSH, follicle stimulating hormone; GLU, glucose; LH, luteinizing hormone; PRL, prolactin; P, progesterone; T, testosterone; TG, triglycerides.

\*Significantly different at  $P \leq 0.05$ .\*\*Highly significant difference at  $P \leq 0.01$ .

correlated with both GLU and E2 ( $P < 0.01$ ). E2 stimulates lipid metabolism, promotes adipocyte growth and division, and controls the TG stores. Han et al. (2009) found that GLU is a major inducer of hepatic lipogenesis and that it can induce TG accumulation. Therefore, E2, GLU, and TG function synergistically to improve the egg-laying performance of Wulong geese.

PRL is a protein hormone that originates from the anterior pituitary gland and acts on the ovaries; its physiological role is to inhibit the growth and development of follicles; it also promotes nesting behavior in birds. P can accelerate follicle maturation and ovulation, thus improving egg production, and the androgen T can be converted into E2 by aromatization (Li et al., 1996). Fang et al. (2005) found that PRL levels were negatively correlated with those of E2 throughout the breeding cycle of the western Anhui white goose. Reddy et al. (2002) found that PRL was negatively correlated with egg production, E2, and P. PRL acts directly on ovarian granulosa cells, inhibiting FSH-induced granulosa cell aromatase activity, reducing E2 synthesis, and inhibiting ovulation (Fortune et al., 1986). In this study, PRL was positively correlated with FSH, LH, and P, and PRL was positively correlated with P ( $P < 0.05$ ), which differs from the findings of Reddy et al. However, our findings that PRL correlated negatively with T, E2, GLU, and TG supports their observations.

FSH and LH are the main gonadotropins that regulate the development of poultry follicles. When FSH and LH act synergistically on the gonads, they regulate the secretion of various reproductive hormones by ovarian germ cells, thus affecting the proliferation, development, and maturation of follicles (Yoshimura et al., 1994). Our findings showed that FSH correlated positively with PRL ( $P < 0.05$ ) while LH was negatively correlated with GLU, E2, TG, and T, with correlation coefficients of 0.405, 0.363, 0.325, and 0.209, respectively. There is controversy concerning the action of LH on follicular membrane cells: Marrone and Hertelendy (1985) suggested that LH stimulates follicular membrane cells to synthesize E2, while Onagbesan and Peddie (1989) reported contrary results. Therefore, the mechanism of action of LH remains to be further investigated.

### Analysis of the Correlation Between Body Size and Reproductive Hormones

Zhu et al. (2014) reported that the TG concentration in the Gaoyou duck positively correlated with body weight, while Wang et al. (2002) found that TG levels in the blood of the Muscovy duck correlated negatively with the body weight at 70 days old. In the present study, there was a positive correlation between body weight and TG ( $P < 0.01$ ). The results of this study are inconsistent with the report by Wang et al. but agree with the findings of Zhu et al. Neck circumference and neck length were negatively correlated with E2 and TG

( $P < 0.01$ ); pubic spacing and abdominal circumference were negatively correlated with E2, GLU, and TG ( $P < 0.01$ ). These findings suggest that, in the process of conservation breeding, reproductive performance can be predicted by using these body size traits, both saving cost and accelerating the breeding process.

## CONCLUSIONS

There were significant differences in neck circumference, neck length, pubic spacing, tibial circumference, abdominal circumference, E2, GLU, and TG between the HYG and LYG groups. The neck circumference, neck length, and tibial circumference of the HYG were significantly lower than those of the LYG while other indices were significantly higher than those of the LYG. E2, GLU, and TG play crucial roles in egg production in Wulong geese; the pubic spacing and abdominal circumference were positively correlated with E2, GLU, and TG, while neck circumference and neck length were negatively correlated with E2 and TG. Therefore, female geese with short thin necks, wide pubic spacing, and large bellies can be selected as HYG for breeding.

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## DISCLOSURES

The authors declare no competing financial interest.

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