KSÜ Tarım ve Doğa Derg 29 (6), 1583-1604, 2026
KSU J. Agric Nat 29 (6), 1583-1604, 2026
DOI:10.18016/ksutarimdoga.vi.1866882

Assessment of the Effects of Genotype and Sex on Welfare and Fluctuating Asymmetry in Anadolu-T and Cobb500 Broilers

Hasan ELEROĞLU[1], Beyhan YETER[2]✍
[1]Department of Plant and Animal Production, Sivas Technical Sciences Vocational School, Sivas Cumhuriyet University, Sivas, Türkiye,
[2]Department of Animal Science, Agriculture Faculty, Kahramanmaraş Sütçü İmam University, Kahramanmaraş, Türkiye
✉: byeter@gmail.com

ABSTRACT

This study was conducted to compare the welfare parameters of the local broiler genotype, Anadolu-T, with the commercial genotype, Cobb 500. The research was carried out at the fully controlled R&D poultry house of Kahramanmaraş Sütçü İmam University, using a total of 240 chicks (120 from each genotype). During the experiment, body weights (BW), rectal temperatures (RT), tonic immobility (TI) durations, and bilateral body measurements were measured to determine developmental stability. Rectal temperature (RT) at 3 weeks of age was significantly affected by genotype, with Anadolu–T chickens exhibiting higher RT values than Cobb 500. At 6 weeks, RT was significantly influenced by both genotype and sex. Anadolu–T chickens continued to maintain higher RT values than Cobb500, while females showed higher RT than males. Tonic immobility durations revealed no statistically significant differences between the genotypes, suggesting that both groups experienced similar fear levels under the experimental conditions. Furthermore, the fluctuating asymmetry values calculated from bilateral measurements showed that both genotypes exhibited similar performance regarding environmental factors and developmental stability. In conclusion, while the local Anadolu-T genotype lags behind its commercial counterpart in terms of growth rate, it was determined to be comparable to Cobb 500 regarding welfare parameters, demonstrating a healthy developmental pattern.

Animal Science

Research Article

Article History
Received : 19.01.2026
Accepted : 09.04.2026

Keywords
Anadolu-T
Cobb500
Broiler
Welfare
Asymmetry

Anadolu-T ve Cobb500 Etlik Piliçlerinde Genotip ve Cinsiyetin Refah ve Dalgalı Asimetri Üzerine Etkileri

ÖZET

Bu çalışma, yerli broiler genotipi Anadolu-T ile ticari genotip Cobb 500'ün refah parametrelerini karşılaştırmak amacıyla yürütülmüştür. Araştırma, Kahramanmaraş Sütçü İmam Üniversitesi'nin tam kontrollü Ar-Ge kümesinde, toplam 240 civciv (her genotipten 120) kullanılarak gerçekleştirilmiştir. Deneme süresince, gelişimsel stabiliteyi belirlemek amacıyla vücut ağırlıkları (BW), rektal sıcaklıklar (RT), tonik immobilite (TI) süreleri ve bilateral vücut ölçümleri ölçülmüştür. 3 haftalık yaşta rektal sıcaklık (RT) genotipten önemli ölçüde etkilenmiş olup, Anadolu-T tavukları Cobb 500'e göre daha yüksek RT değerleri göstermiştir. 6 haftalık yaşta RT, hem genotip hem de cinsiyetten önemli ölçüde etkilenmiştir. Anadolu-T tavukları Cobb 500'e kıyasla yüksek RT değerlerini korurken, dişiler erkeklere göre daha yüksek RT göstermiştir. Tonik immobilite süreleri genotipler arasında istatistiksel olarak anlamlı farklılık göstermemiştir; bu durum her iki grubun da deneysel koşullar altında benzer korku düzeyleri yaşadığını düşündürmektedir. Ayrıca, bilateral ölçümlerden hesaplanan dalgalı asimetri değerleri, her iki genotipin çevre faktörleri ve gelişimsel stabilite açısından benzer performans sergilediğini göstermiştir. Sonuç olarak, yerli Anadolu-T genotipi büyüme hızı açısından ticari

Hayvan Yetiştirme

Araştırma Makalesi

Makale Tarihçesi
Geliş Tarihi : 19.01.2026
Kabul Tarihi : 09.04.2026

Anahtar Kelimeler
Anadolu-T
Cobb500
Etlik piliç
Refah
Asimetri

eşine göre geride kalmasına rağmen, refah parametreleri açısından Cobb 500 ile karşılaştırılabilir bulunmuş ve sağlıklı bir gelişimsel profil sergilediği belirlenmiştir.

Atıf İçin : Eleroğlu, H & Yeter, B (2026). Anadolu-T ve Cobb500 Etlik Piliçlerinde Genotip ve Cinsiyetin Refah ve Dalgalı Asimetri Üzerine Etkileri. *KSÜ Tarım ve Doğa Derg 29* (6), 1583-1604. DOI: 10.18016/ksutarimdoga.vi.1866882.
To Cite: Eleroğlu, H & Yeter, B (2025). Assessment of the Effects of Genotype and Sex on Welfare and Fluctuating Asymmetry in Anadolu-T and Cobb500 Broilers. *KSU J. Agric Nat 29* (6), 1583-1604. DOI: 10.18016/ ksutarimdoga.vi.1866882.

INTRODUCTION

Because the right and left sides of the body in animals are governed by the same genetic mechanisms, bilateral traits are expected to develop symmetrically when animals are exposed to low stress levels, experience high welfare conditions, and are raised under suitable management practices (Yang et al., 1997). Consequently, minimal differences between corresponding bilateral traits are generally anticipated (Yalçın et al., 2001). Although both sides typically develop under comparable environmental conditions, any deviation between the right and left sides is considered a manifestation of developmental instability (Eleroğlu et al., 2015; Eleroğlu and Yıldırım, 2019; Eleroğlu et al., 2020).

Modern broiler chickens possess a high growth potential, allowing them to attain market weight in a relatively short time (Karaarslan, 2016). Nevertheless, when rapid growth occurs under suboptimal housing, nutrition, or welfare conditions, it may predispose broilers to deteriorations in meat quality, metabolic disorders such as ascites, disrupted growth patterns, developmental instability, and a range of health-related problems (Shahin & El Azeem, 2005, 2006; Mendes, 2008). Unfavorable environmental conditions within poultry houses particularly high stocking density, inappropriate lighting regimes, and unsuitable feeding strategies can further exacerbate irregular growth and impaired physiological development (Yalçın et al., 2001). Consequently, various asymmetry-based indices are widely employed to quantify developmental disturbances in bilaterally symmetrical traits (Mendes, 2008).

Deviations from perfect right–left symmetry in animals are referred to as fluctuating asymmetry (Van Nuffel et al., 2007). This form of asymmetry has been widely used as an indicator for evaluating the adequacy of rearing conditions and management practices (Knierim et al., 2007; Van Nuffel et al., 2007). Asymmetry is considered a potential welfare-related indicator, as it reflects an animal's capacity to cope with individual challenges that may disrupt normal development at certain stages of life (Broom, 2006; Knierim et al., 2007). Asymmetry can be described as random and direct departures from symmetrical growth, which commonly arise during morphological development as a consequence of interactions between genetic background and environmental influences, and may manifest in specific body regions (Nääs et al., 2009).

In recent years, fluctuating asymmetry (FA) has gained considerable attention as an indicator of animal welfare (Moller and Pomiankowski, 1993; Van Poucke et al., 2007), and it has been reported to be influenced by both environmental conditions and genetic background (Parsons, 1992; Gomendio et al., 2000; Lens et al., 2002; Campo et al., 2002; Knierim et al., 2007; Van Nuffel et al., 2007). Previous studies have demonstrated positive associations between FA and several stress-related factors, including heat stress (Yalçın et al., 2001; Yalçın and Siegel, 2003), tonic immobility responses (Moller et al., 1995; Campo et al., 2000), locomotion disorders (Moller et al., 1999), inappropriate lighting programs (Moller et al., 1999), corticosterone responses to stress (Satterlee et al., 2000), and high stocking density (Moller et al., 1995).

This study evaluated the effects of genotype and sex on the morphological characteristics of hybrids derived from Anadolu–T and Cobb 500 genotypes reared under a closed housing system. In addition, asymmetry in bilaterally symmetrical traits was examined to assess the potential influence of genotype and sex on animal welfare.

MATERIAL and METHOD

Research Location and Animal Material: The study was conducted at the fully controlled broiler Research and Development (R&D) poultry house of the Animal Production Application and Research Center at Kahramanmaraş Sütçü İmam University. Broiler chicks belonging to the Anadolu–T and Cobb500 genotypes, obtained from parent flocks of the same age (32 weeks), were used as animal material in the experiment.

Experimental Design and Housing Conditions: The experiment consisted of two main treatment groups representing two different genotypes. Each group included four replicates, with 30 chicks allocated to each replicate, resulting in a total of 240 chicks (2 genotypes × 4 replicates × 30 chicks). The experiment was conducted using a completely randomized design, in which replicates were randomly assigned to the experimental treatments.

The chicks were housed in a fully controlled closed poultry house measuring 7 m × 19 m and were placed in pens measuring 2 × 1.5 m (3 m²), separated by wire mesh. Stocking density was maintained at 10 chicks/m² throughout the experimental period. The floor of the house was covered with pine wood shavings at a depth of 7–8 cm as litter material.

Environmental Conditions and House Management: The indoor climate and ventilation were optimized using the following equipment:

Heating: Thermostat-controlled, forced-air electric heaters were used.

Ventilation: One 60-cm fan and seven air inlets were used for minimum ventilation.

Cooling: From day 10 onward during daytime, and from day 25 onward during both daytime and night-time when cooling was required, cooling was provided via a total of 6 m² (2 × 3 m²) evaporative cooling pads located on both sidewalls of the house, in combination with three 90-cm tunnel fans.

Lighting: A photoperiod of 23 h light and 1 h darkness was applied throughout the experiment. Lighting was provided using 15-W bulbs with a daylight spectrum, and light intensity was measured at feeder level as 20 lux using a lux meter (Lutron LX-101).

Feeding and Management Program: The chicks were fed three different commercial diets obtained from a commercial feed company (broiler starter, grower, and finisher diets). The nutrient composition of the diets is presented in Table 1.

Feeding: One hanging feeder was placed in each pen, and feed was offered ad libitum after weighing.

Watering: Water was provided ad libitum through nipple drinkers (five nipples per pen). As supportive care, a multivitamin supplement (20 g/200 L) was added to the drinking water once a week.

Health Management: As part of biosecurity measures, live Newcastle disease vaccine was administered via drinking water on day 6, and live Gumboro vaccine was administered on day 13.

Sex determination: In broiler chickens, sex determination from 21 days of age was performed subjectively based on body weight and morphometric traits, including body length, breast width, thigh and shank lengths, comb and wattle development, and wing length

Table 1. Feed types provided to chicks at different ages
Çizelge 1. Farklı yaşlardaki civcivlere verilen yem çeşitleri

Ingredients	0-10 days	11-25 days	26-42 days
Dry matter %	89.63	89.42	89.26
Metabolizable enerji MJ/kg	12.20	12.56	13.08
Crude Protein %	22.00	20.50	19.50

Data Collection and Measurements

The following parameters were recorded during the experimental period:

Morphological Measurements: In the study, a total of 240 broiler chickens from both genotypes were used. Morphological measurements were analysed for 64 chicks, comprising 15 males and 15 females randomly selected from each genotype, and were taken at days 21 and 42 using a digital caliper (Van Nuffel et al., 2007). To minimize measurement error, all measurements were performed by the same researcher, and each measurement was taken three times on the same chick, with the average value used for analysis. In the study, each genotype consisted of four replicates, and four male and four female chicks were selected from each replicate. Measurements were taken at the 3rd and 6th weeks. In total, 64 chicks were evaluated (2 genotypes × 4 replicates × 8 chicks, comprising 4 males and 4 females per replicate). To evaluate the effects of genotype and sex on selected morphological traits and asymmetry, bilateral measurements were obtained from the head region of roosters on both the right and left sides. The traits measured included Comb length (CL), Comb width (CW), Eye width (EW), Beak length (BL), Facial width (FW), Wattle length (WL), Wattle width (WW) and nose Length (NL).

For the feet, morphological measurements were taken on both the right and left sides, including Joint width (JW), Tarsometatarsus length (TML Tarsometatarsus width at 1 cm above the spur (TMW-1), Tarsometatarsus width at the spur level (TMW-SL), Hind toe length (HTL), Outer toe claw length (OTCL), Fourth phalanx length of the outer toe (OP4L), Third phalanx length of the outer toe (OP3L), Middle toe claw length (MTCL), Fourth phalanx length of the middle toe (MP4L), and Third phalanx length of the middle toe (MP3L). The morphological characteristics evaluated in this study are illustrated in Figures 1 and 2 (Eleroğlu et al., 2020).

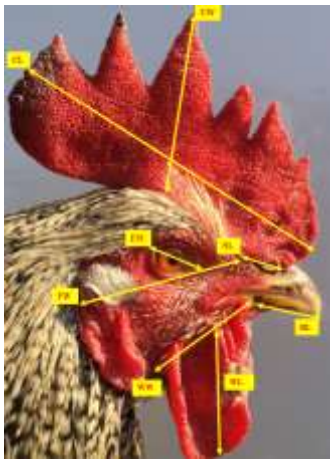


Figure 1. Head measurements
Şekil 1. Baş bölgesine ait ölçümler
CL: İbik uzunluğu, CW: İbik genişliği, EW: Göz genişliği, BL: Gaga uzunluğu, FW: Yüz genişliği, WL: Sakal uzunluğu, WW: Sakal genişliği, NL: Burun uzunluğu
Comb length (CL), Comb width (CW), Eye width (EW), Beak length (BL), Facial width (FW), Wattle length (WL), Wattle width (WW) and nose Length (NL). (Eleroğlu et al., 2020).

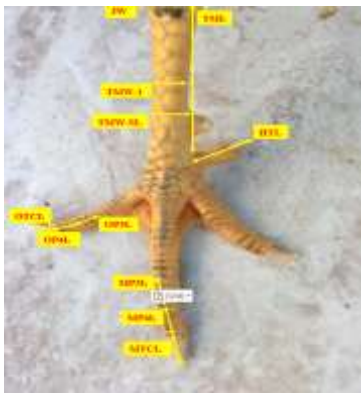


Figure 2. Foot measurements
Şekil 2. Ayak ölçümleri
JW: Eklem genişliği, TML: Tarsometatarsus uzunluğu, TMW-1: Mahmuzun 1 cm üstünde tarsometatarsus genişliği, TMW-SL: Mahmuz seviyesindeki tarsometatarsus genişliği, HTL: Arka parmak uzunluğu, OTCL: Dış parmak tırnak uzunluğu, OP4L: Dış parmağın dördüncü falanks uzunluğu, OP3L: Dış parmağın üçüncü falanks uzunluğu, MTCL: Orta parmak tırnak uzunluğu, MP4L: Orta parmağın dördüncü falanks uzunluğu, MP3L: Orta parmağın üçüncü falanks uzunluğu
Joint width (JW), Tarsometatarsus length (TML Tarsometatarsus width at 1 cm above the spur (TMW-1), Tarsometatarsus width at the spur level (TMW-SL), Hind toe length (HTL), Outer toe claw length (OTCL), Fourth phalanx length of the outer toe (OP4L), Third phalanx length of the outer toe (OP3L), Middle toe claw length (MTCL), Fourth phalanx length of the middle toe (MP4L), and Third phalanx length of the middle toe (MP3L). (Eleroğlu et al., 2020).

Fluctuating asymmetry (FA) was defined as the absolute difference between the right and left measurements, and was calculated as follows;

$FA = |R - L|$ (Campo et al., 2002).

Relative asymmetry (RA) was expressed as the ratio of the absolute asymmetry to the mean of the right and left trait values and was calculated as follows:

$RA = (|R - L| / [(R + L)/2]) \times 100$ (Yang, 1998).

Relative asymmetry (RA) of the left and right bones (%) was calculated as $RA = |R - L| / ((R + L)/2) \times 100$, where R and L are the lengths of the right and left bones (mm), $|R - L|$ is their absolute difference, and $(R + L)/2$ is the average used to standardize the difference relative to bone size.

Physiological Measurements

Rectal Temperature (RT): Cloacal temperature was measured by inserting the probe of a digital thermometer 2 cm into the cloaca.

Tonic Immobility (TI): Tonic immobility was used to evaluate fear responses in the broiler chicks. Chicks were gently placed on their backs on a padded surface, and light pressure was applied to the chest to induce immobility. The stopwatch was started after the pressure was released, and the duration until the bird attempted to stand up was recorded. Chicks that did not show a righting response within 10 minutes were excluded from the evaluation (Marx et al., 2008; Prieto & Campo, 2010).

Statistical Analysis: Data were analyzed to determine the effects of genotype and sex on the measured traits. A two-way analysis of variance (ANOVA) was performed, including genotype and sex as fixed effects. The interaction between genotype and sex was also included in the model. Statistical significance was declared at $P < 0.05$ (Düzgüneş et al., 1987; Bek et al., 2000).

Results are presented as means ± standard error of the mean (SEM).

Statistical model:

$$Y_{ij} = \mu + G_i + S_j + (G \times S)_{ij} + e_{ij}$$

Where

Y_{ij}= observed value,

μ= overall mean,

G_i= effect of genotype,

S_j= effect of sex,

$(G \times S)_{ij}$= interaction effect,

e_{ij}= random error.

RESULTS and DISCUSSION

The effects of genotype, sex, and their interaction on tonic immobility (TI) duration and rectal temperature (RT) at 3 and 6 weeks of age are presented in Table 2.

Table 2. Effects of genotype and sex on tonic immobility reaction and rectal temperature (°C) of broilers
Çizelge 2. Genotip ve cinsiyetin etlik piliçlerin tonik hareketsizlik reaksiyonu ve rektal sıcaklığı (°C) üstüne etkileri

Items		3. week		6. week	
Genotype	Sex	TI±SEM	RT±SEM	TI±SEM	RT±SEM
Anadolu - T	M	213,68±36,23	41.6±0.06	204.0±35.44	41.17±0.15
	F	279±49,85	41.48±0.07	108.9±14.42	41.66±0.09
	T	246,34±30,87	41.54±0.05	156.45±20.66	41.42±0.10
Cobb 500	M	247,08±49,53	41.34±0.08	191.14±43.83	41.07±0.09
	F	269,81±55,79	41.17±0.12	187.73±48.82	41.09±0.12
	T	258,44±36,75	41.26±0.07	189.43±30.55	41.08±0.07
Source of variation		Sig.	Sig.	Sig.	Sig.
Genotype (G)		0.389	***0.001***	0.369	***0.003***
Sex (S)		0.831	0.064	0.182	***0.023***
G × S		0.333	0.754	0.214	***0.034***

F = female; M = male; T = Total; TI = Tonic immobility; RT = Rectal temperatures

At 3 weeks of age, TI duration was not significantly affected by genotype or sex ($p > 0.05$). Although Cobb500 chickens exhibited numerically longer TI durations compared with Anadolu–T, the difference was not statistically significant. Similarly, no significant genotype × sex interaction was detected for TI at this age ($p > 0.05$). In contrast, RT at 3 weeks was significantly influenced by genotype ($p < 0.01$), with Anadolu–T chickens showing higher RT values than Cobb500. The effect of sex on RT tended to be marginal at 3 weeks ($p = 0.064$), whereas no significant genotype × sex interaction was observed.

At 6 weeks of age, tonic immobility duration remained unaffected by genotype, sex, or their interaction ($p > 0.05$). However, rectal temperature was significantly influenced by genotype ($p < 0.01$) and sex ($p < 0.05$). Anadolu–T chickens maintained higher rectal temperatures than Cobb500, while females exhibited higher RT values compared with males. Moreover, a significant genotype × sex interaction was detected for rectal temperature at 6 weeks ($p < 0.05$), indicating that the magnitude of sex-related differences in RT varied between genotypes.

Overall, these results demonstrate that while fear-related behavior measured by tonic immobility was not markedly affected by genotype or sex, physiological responses assessed via rectal temperature were significantly influenced by both factors, particularly at later ages.

Tonic immobility (TI) is widely recognized as a reliable behavioural indicator of fearfulness and stress responsiveness in poultry, with prolonged TI duration generally associated with heightened fear reactions and anti-predator responses in chicks (Forkman et al., 2007; Ghareeb, 2010; Tiemann et al., 2023). Fear-related behaviour is typically expressed as a rapid and transient response to immediate challenges; however, the magnitude and persistence of this response are strongly influenced by genetic background. Differences among genetic strains in fearfulness and temperament may therefore contribute to variability stress-related behaviour responses observed under similar environmental conditions (Campbell et al., 2022; Nelson et al., 2020; Rentsch et al., 2023). In the present study, TI duration was not significantly affected by genotype or sex at either 3 or 6 weeks of age. This finding suggests that Anadolu–T and Cobb500 chickens exhibited comparable fear-related behavioural responses under the experimental conditions. Similar results have been reported in previous studies indicating that genetic background does not always result in pronounced differences in TI duration, particularly when chicks are raised under uniform management and environmental conditions (Forkman et al., 2007).

In contrast to the behavioural findings, RT, a well-established physiological indicator of stress and thermoregulation in poultry, was significantly influenced by genotype at both ages, consistent with previous observations that genetic strain can affect core body temperature and heat stress responses in chickens (Anderson & Jones, 2012; Adu-Asiamah et al., 2021). Anadolu–T chickens consistently exhibited higher RT values than Cobb500 birds, indicating genotype-dependent differences in physiological responses.

In chickens, genetic background has been shown to influence physiological responses to heat stress, including core body temperature regulation, with certain strains exhibiting greater increases in RT under thermal challenge than others, indicating genotype-specific differences in thermoregulatory capacity (Felver-Gant et al., 2012). The higher RT observed in Anadolu–T chickens may therefore be associated with differences in metabolic rate, growth pattern, or adaptive capacity to environmental stressors compared with fast-growing commercial broilers.

The effect of sex on RT became significant at 6 weeks of age, with females exhibiting higher RT values than males. This age-related sex effect may be explained by differences in hormonal regulation, body composition, and metabolic activity that become more pronounced as chicks approach market age. Previous research has shown that physiological responses such as rectal temperature and core body temperature regulation under heat conditions can differ between male and female chickens, with males sometimes exhibiting higher RT than females under the same thermal conditions (Yehia et al., 2025).

A significant genotype × sex interaction for rectal temperature at 6 weeks indicates that thermophysiological responses differ between males and females depending on genetic background, supporting previous reports that thermoregulation and stress responses in chickens are jointly influenced by genotype and sex (Felver-Gant et al., 2012; Yehia et al., 2025)

A key outcome of the present study is the divergence observed between behavioural and physiological indicators of stress. Although tonic immobility duration showed limited sensitivity to genotype and sex, rectal temperature displayed marked differences, indicating that these responses do not necessarily vary in parallel. This supports the view that behavioural fear reactions may reflect distinct regulatory processes. Accordingly, reliance on a single welfare indicator may fail to capture the full complexity of stress responsiveness. Overall, the results suggest that genotype and sex have a more pronounced effect on physiological regulation than on fear-related behaviour, particularly at later ages, underscoring the necessity of combining behavioural and physiological measures in comprehensive welfare evaluations of broiler genotypes [L48.1] (De Jong, I. C., & Guémené, 2011; Yehia et al., 2025).

The effects of genotype and sex on non-bilateral morphological characteristics, including comb length (CL), comb width (CW), and beak length (BL), at 3 and 6 weeks of age are presented in Table 3. The maximum, median, and minimum values of Comb length, Comb width, and Beak length by genotype and sex at week 6 are presented in Figures 3, 4, and 5, respectively. At 3 weeks of age, both CL and CW were significantly influenced by genotype, with Cobb500 chickens exhibiting greater comb dimensions than Anadolu–T chicks, likely reflecting differences in early growth rate and selection intensity between commercial and locally adapted genotypes. Sex exerted a highly significant effect on both traits at this age, as males displayed markedly longer and wider combs than females across genotypes, indicating an early onset of sexual dimorphism. In contrast, BL was not affected by genotype or sex at 3 weeks, suggesting relatively uniform early beak development. No significant genotype × sex interaction was observed for CL or CW, although a tendency toward interaction was detected for BL.

By 6 weeks of age, sex remained the dominant factor influencing CL and CW, with males continuing to show substantially larger comb dimensions than females. While the main effect of genotype on comb traits was not statistically significant, a significant genotype × sex interaction was detected for CL, indicating that sex-related differences in comb length varied between genotypes and reflected genotype-specific growth trajectories of secondary sexual characteristics. At this age, BL exhibited a different pattern: genotype had a significant effect, with Anadolu–T chickens showing slightly longer beaks than Cobb500 chicks, whereas sex had no influence. This suggests that beak morphology is more strongly associated with genetic background than with sexual

differentiation (Struthers et al., 2023).

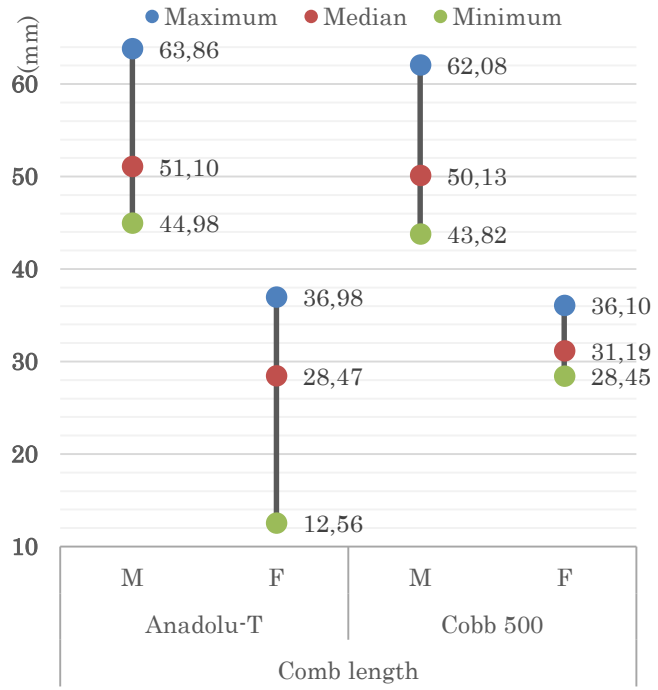


Figure 3. Maximum, Median, and Minimum Values of Comb Length by Genotype and Sex at Week 6

Şekil 3. 6. haftada genotip ve cinsiyete göre ibik uzunluğunun maksimum, medyan ve minimum değerleri

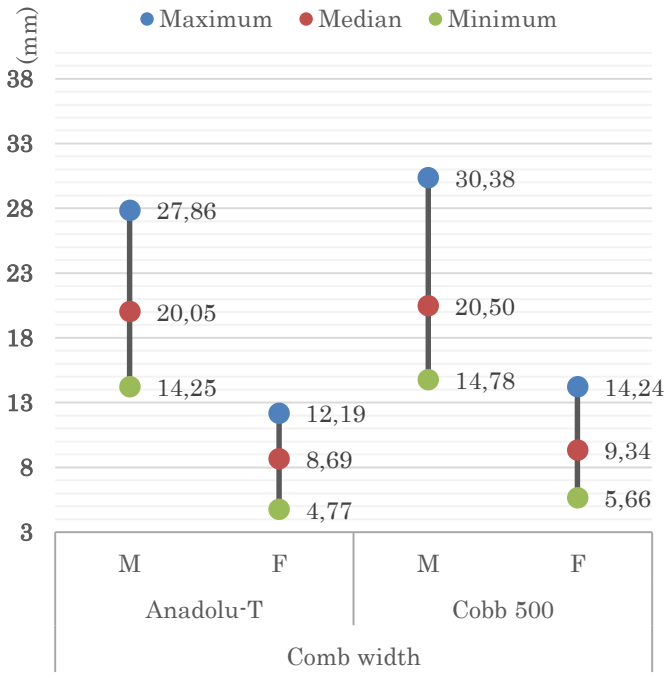


Figure 4. Maximum, Median, and Minimum Values of Comb width by Genotype and Sex at Week 6

Şekil 4. 6. haftada genotip ve cinsiyete göre ibik genişliğinin maksimum, medyan ve minimum değerleri

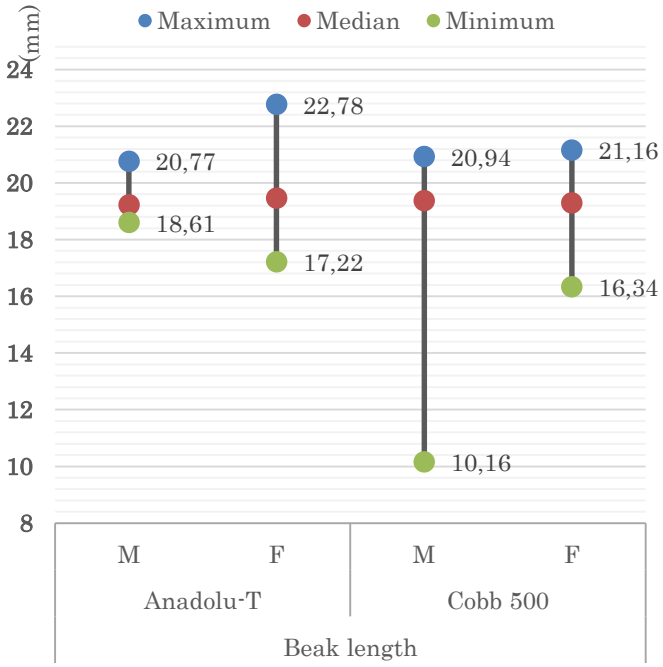


Figure 5. Maximum, Median, and Minimum Values of Beak length by Genotype and Sex at Week 6

Şekil 5. 6. haftada genotip ve cinsiyete göre gaga uzunluğunun maksimum, medyan ve minimum değerleri

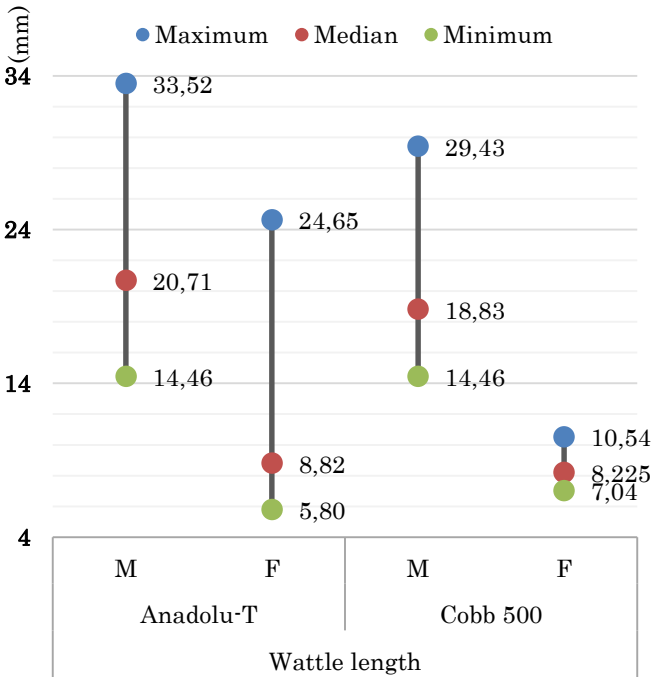


Figure 6. Maximum, Median, and Minimum Values of Wattle length by Genotype and Sex at Week 6

Şekil 6. 6. haftada genotip ve cinsiyete göre sakal uzunluğunun maksimum, medyan ve minimum değerleri

Overall, the results demonstrate that comb-related traits are strongly sex-dependent and closely linked to hormonal regulation, consistent with their role as secondary sexual characteristics. Previous studies have shown that comb development is associated with endocrine activity, particularly growth hormone and testosterone, and reflects physiological status, immune regulation, and growth performance in chickens (Qi et al., 2025). Moreover,

androgen receptor signalling has been identified as a key molecular mechanism underlying avian sexual development and the expression of secondary sexual traits such as the comb (Lengyel et al., 2024). In contrast, the age-dependent genotype effect observed for BL supports the view that beak morphology reflects divergent selection pressures related to feeding behaviour, adaptability, and genetic background rather than sexual dimorphism (Struthers et al., 2023; Knief et al., 2012).

Table 3. Influence of genotype and sex on non-bilateral morphological characteristics
Çizelge 3. Genotip ve cinsiyetin iki taraflı olmayan morfolojik özellikler üstüne etkisi

Items		3. weeks			6. weeks		
Genotype	Sex	CL±SEM	CW±SEM	BL±SEM	CL±SEM	CW±SEM	BL±SEM
Anadolu - T	M	34.62±1.09	12.87±0.63	16.16±0.25	51.96±1.40	20.34±0.96	19.55±0.19
	F	25.53±0.83	6.49±0.48	16.89±0.70	28.13±1.30	8.29±0.60	19.62±0.39
	T	30.08±1.06	9.68±0.69	16.53±0.37	40.04±2.34	14.32±1.22	19.58±0.21
Cobb 500	M	37.71±0.84	14.3±0.63	17.05±0.34	50.81±1.11	21.19±0.86	18.34±0.71
	F	26.61±0.79	8.19±0.41	16.23±0.37	31.57±0.55	9.50±0.57	18.92±0.44
	T	32.16±1.13	11.24±0.65	16.64±0.24	41.19±1.83	15.35±1.16	18.63±0.4
Source of variation		Sig.	Sig.	Sig.	Sig.	Sig.	Sig.
Genotype (G)		***0.021***	***0.005***	0.799	0.316	0.178	***0.042***
Sex (S)		***0.000***	***0.000***	0.918	***0.000***	***0.000***	0.484
G × S		0.256	0.799	0.084	***0.047***	0.811	0.583

F = female; M = male, T=Total; CL = Comb length; CW = Comb width; BL= Beak length

Taken together, these findings highlight that different non-bilateral morphological traits respond differently to genotype- and sex-related factors. While comb traits are useful indicators of sex and developmental stage, beak length appears to reflect genotype-specific growth patterns. This underscores the importance of evaluating multiple morphological traits when assessing growth dynamics, sexual dimorphism, and phenotypic differentiation among chicken genotypes (McGary et al., 2003; Struthers et al., 2023).

Fluctuating and Relative Asymmetry of Bilateral Head at 3 Weeks of Age

The fluctuating asymmetry (FA) and relative asymmetry (RA) indices calculated from bilateral head measurements at 3 weeks of age are presented in Table 4. Indices of asymmetry, particularly FA, have been widely applied as indicators of developmental stability, reflecting an individual's capacity to buffer environmental and physiological stressors during growth. Increased asymmetry is generally interpreted as reduced developmental stability under adverse environmental factors (Palmer & Strobeck, 1986; Zakharov et al., 2020).

Fluctuating Asymmetry (FA)

At 3 weeks of age, genotype significantly affected FA only for wattle width (WW) (P = 0.049), with Cobb500 broilers exhibiting higher FA values than Anadolu–T chickens. This finding suggests that wattle width may represent a particularly sensitive trait for detecting early genotype-related differences in developmental stability. In contrast, no significant genotype effects were observed for wattle length (WL), eye width (EW), nose length (NL), or facial width (FW), indicating comparable bilateral stability for most cranial traits between genotypes at this early growth stage. Sex exerted a significant influence on FA values for WW (P = 0.039) and EW (P = 0.017), with females generally displaying higher asymmetry than males. This pattern suggests slightly reduced developmental stability in females during early post-hatch growth. Such sex-related differences in asymmetry may be associated with differential growth allocation and early endocrine variation between males and females, as sex-specific hormonal environments have been shown to influence growth trajectories and morphological development from early life stages (Wang et al., 2019; Lengyel et al., 2024). Genotype × sex interactions were not significant for most FA traits; however, a marginal interaction for EW (P = 0.053) suggests that sex-related asymmetry patterns in eye width may differ subtly between genotypes.

Relative Asymmetry (RA)

For RA indices at 3 weeks of age, sex had a significant effect on EW (P = 0.007), with females exhibiting higher relative asymmetry than males across genotypes. This result is consistent with the FA findings and further indicates that eye width is particularly sensitive to sex-related variation in early developmental stability. Genotype effects on RA traits were not statistically significant, although numerical differences for EW (P = 0.085) and WL (P = 0.465) may point to emerging genotype-specific patterns that are not yet fully expressed at this age.

The lack of pronounced genotype effects on most RA traits suggests that relative asymmetry at 3 weeks is largely independent of genetic background and may instead reflect short-term physiological processes and sex-related developmental dynamics. Similarly, genotype × sex interactions for RA traits were not significant, although

marginal values for WL (P = 0.081) and EW (P = 0.052) indicate that interaction effects may become more apparent at later stages of development.

Table 4. Fluctuating (FA) and relative (RA) asymmetry indices derived from bilateral head measurements at week3
Çizelge 4. 3. haftada iki taraftan yapılan baş ölçümlerinden elde edilen dalgalı (FA) ve göreceli (RA) asimetri indeksleri

Items		3. Week Fluctuating Asymmetry (FA)				
Genotype	Sex	WL±SEM	WW±SEM	EW±SEM	NL±SEM	FW±SEM
Anadolu - T	M	1.94±0.35	0.92±0.18	0.43±0.09	0.34±0.07	1.60±0.25
	F	2.68±0.25	0.60±0.12	1.18±0.30	0.35±0.06	1.19±0.18
	T	2.31±0.22	0.76±0.11	0.81±0.17	0.35±0.04	1.39±0.16
Cobb 500	M	2.43±0.36	1.43±0.29	0.52±0.09	0.33±0.07	1.37±0.30
	F	2.10±0.39	0.90±0.20	0.60±0.10	0.52±0.08	1.74±0.36
	T	2.26±0.25	1.17±0.18	0.56±0.06	0.42±0.05	1.56±0.22
Source of variation		Sig.	Sig.	Sig.	Sig.	Sig.
Genotype (G)		0.896	***0.049***	0.147	0.280	0.546
Sex (S)		0.534	***0.039***	***0.017***	0.167	0.931
G × S		0.112	0.604	0.053	0.184	0.153
		3. Week Relative Asymmetry (RA)				
Genotype	Sex	WL±SEM	WW±SEM	EW±SEM	NL±SEM	FW±SEM
Anadolu - T	M	2.47±0.42	2.07±0.37	0.91±0.19	1.37±0.26	1.03±0.17
	F	3.89±0.37	2.43±0.46	2.47±0.57	1.56±0.28	0.75±0.12
	T	3.18±0.30	2.25±0.29	1.69±0.33	1.47±0.19	0.89±0.11
Cobb 500	M	2.90±0.42	3.23±0.65	0.99±0.17	1.15±0.26	0.83±0.18
	F	2.85±0.50	3.06±0.71	1.25±0.22	2.01±0.34	1.09±0.24
	T	2.87±0.30	3.15±0.45	1.12±0.13	1.58±0.21	0.96±0.14
Source of variation		Sig.	Sig.	Sig.	Sig.	Sig.
Genotype (G)		0.465	0.104	0.085	0.694	0.708
Sex (S)		0.101	0.860	***0.007***	0.064	0.973
G × S		0.081	0.621	0.052	0.231	0.124

F = female; M = male, T=Total; WL = Wattle length; WW = Wattle width; EW = Eye width; NL = Nose length; FW = Facial width

General Interpretation

Overall, the findings indicate that developmental instability at 3 weeks of age is trait-specific rather than generalized across head morphology. Among the evaluated traits, wattle width and eye width emerged as the most responsive indicators of asymmetry. The higher FA and RA values observed in females for certain traits suggest a sex-related sensitivity in early cranial development, potentially linked to differences in growth rate, energy allocation, and early hormonal regulation. The limited genotype effects observed at this age imply that buffering capacity against developmental perturbations is largely maintained during early growth, with clearer genotype-dependent asymmetry patterns likely to manifest at later developmental stages.

Collectively, these results support the use of FA and RA indices derived from head traits as sensitive, non-invasive indicators of early developmental stability and provide a useful framework for interpreting age-dependent changes in asymmetry in relation to genotype and sex.

Fluctuating and Relative Asymmetry of Bilateral Head at 6 Weeks of Age

The fluctuating asymmetry (FA) and relative asymmetry (RA) indices derived from bilateral head measurements at 6 weeks of age are presented in Table 5. The maximum, median, and minimum values of Wattle length, Wattle width, Eye width, Nose length, and Facial width according to genotype and sex at week 6 are illustrated in Figures 6, 7, 8, 9, and 10, respectively. At this developmental stage, asymmetry patterns were more clearly differentiated by genotype and sex, likely reflecting increased physiological demands and accelerated growth associated with increasing age. Fluctuating asymmetry has been widely used as an indicator of developmental stability, with elevated asymmetry values reflecting reduced capacity to buffer genetic, environmental, and physiological stress during growth (Palmer & Strobeck, 1986; Møller & Swaddle, 1997).

At week 6, genotype had a significant effect on facial width (FW) FA (P = 0.024), with Cobb500 broilers exhibiting higher asymmetry values than Anadolu–T chicks. This finding suggests reduced developmental stability in facial width among fast-growing commercial broilers and is consistent with previous studies reporting higher FA levels in intensively selected, fast-growing chicken genotypes (Møller et al., 1995; Van Nuffel et al., 2007). Such patterns

are commonly attributed to trade-offs between rapid somatic growth and the maintenance of developmental stability.

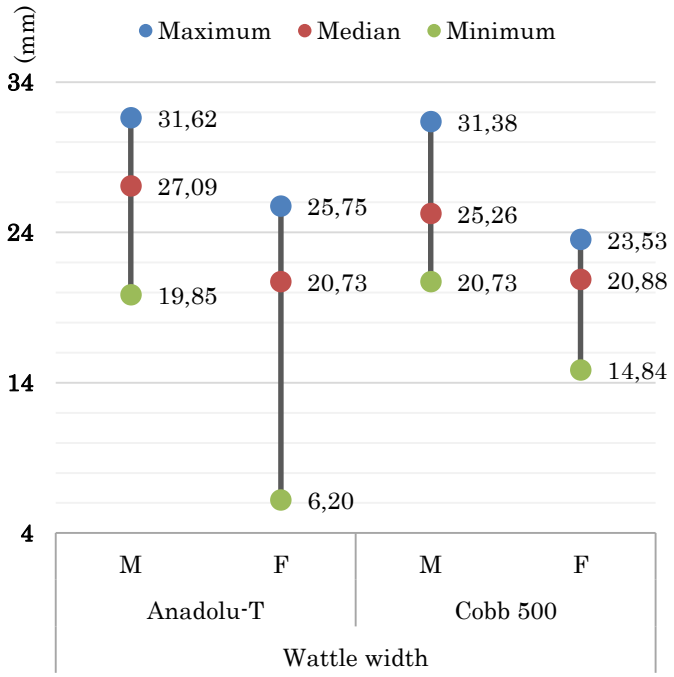


Figure 7. Maximum, Median, and Minimum Values of Wattle width by Genotype and Sex at Week 6

Şekil 7. 6. haftada genotip ve cinsiyete göre sakal genişliğinin maksimum, medyan ve minimum değerleri

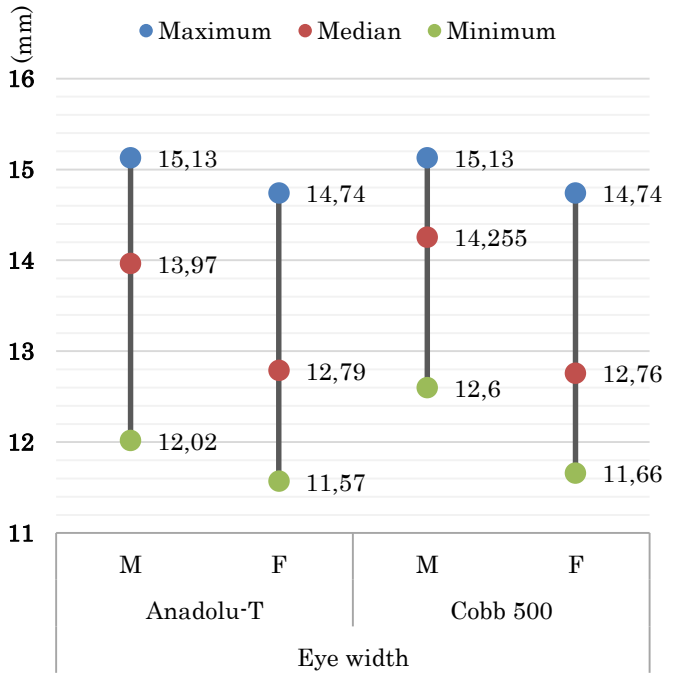


Figure 8. Maximum, Median, and Minimum Values of Eye width by Genotype and Sex at Week 6

Şekil 8. 6. haftada genotip ve cinsiyete göre göz genişliğinin maksimum, medyan ve minimum değerleri

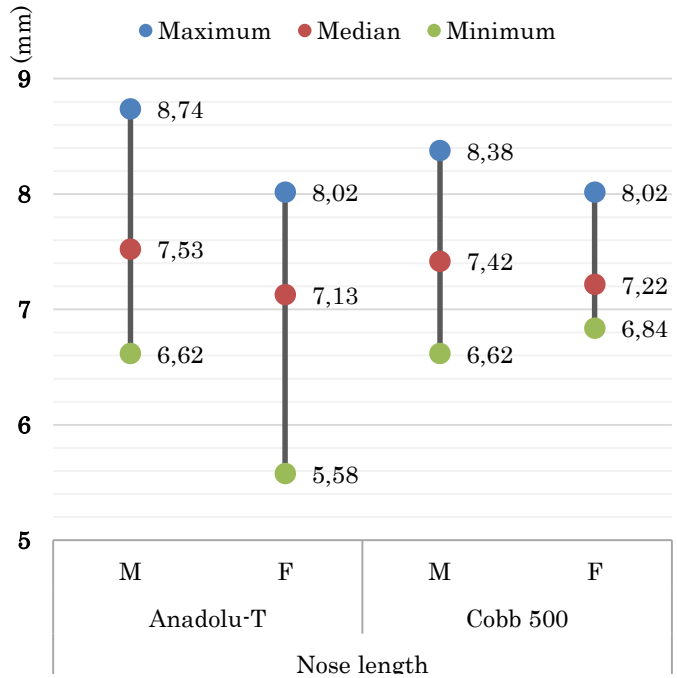


Figure 9. Maximum, Median, and Minimum Values of Nose length by Genotype and Sex at Week 6

Şekil 9. 6. haftada genotip ve cinsiyete göre burun uzunluğunun maksimum, medyan ve minimum değerleri

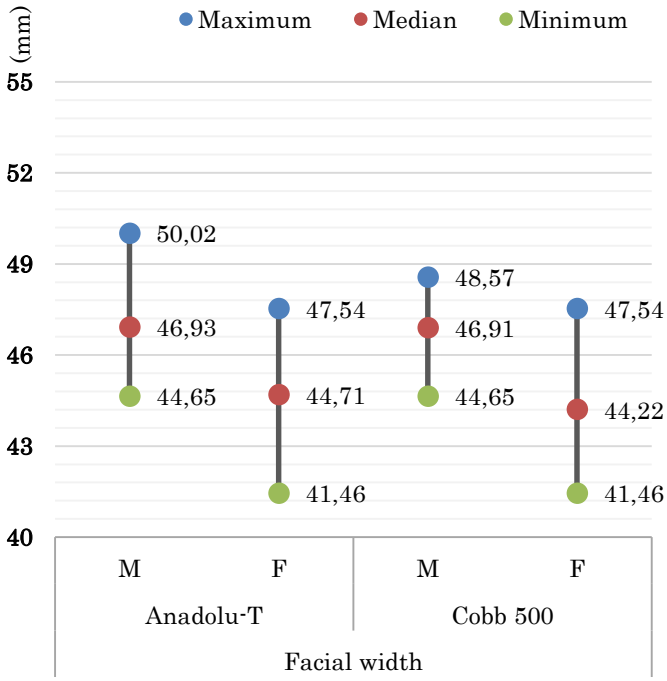


Figure 10. Maximum, Median, and Minimum Values of Facial width by Genotype and Sex at Week 6

Şekil 10. 6. haftada genotip ve cinsiyete göre yüz genişliğinin maksimum, medyan ve minimum değerleri

Table 5. Fluctuating (FA) and relative (RA) asymmetry indices derived from bilateral head measurements at week6
Çizelge 5. Altıncı haftada iki taraflı baş ölçümlerinden elde edilen dalgalı (FA) ve göreceli (RA) asimetri indeksleri

Items		6. Week Fluctuating Asymmetry (FA)				
Genotype	Sex	WL±SEM	WW±SEM	EW±SEM	NL±SEM	FW±SEM
Anadolu - T	M	3.01±0.41	2.69±0.56	0.49±0.09	0.30±0.07	1.62±0.29
	F	1.49±0.28	1.70±0.36	0.36±0.06	0.40±0.11	1.59±0.32
	T	2.25±0.28	2.19±0.34	0.42±0.05	0.35±0.06	1.61±0.21
Cobb 500	M	1.72±0.42	3.00±0.55	0.48±0.07	0.40±0.07	2.52±0.36
	F	1.42±0.40	2.28±0.71	0.33±0.10	0.40±0.08	2.20±0.38
	T	1.57±0.27	2.64±0.42	0.41±0.06	0.40±0.05	2.36±0.24
Source of variation		Sig.	Sig.	Sig.	Sig.	Sig.
Genotype (G)		0.073	0.411	0.810	0.532	***0.024***
Sex (S)		***0.017***	0.118	0.082	0.492	0.602
G × S		0.106	0.799	0.841	0.522	0.650
		6. Week Relative Asymmetry (RA)				
Genotype	Sex	WL±SEM	WW±SEM	EW±SEM	NL±SEM	FW±SEM
Anadolu - T	M	3.27±0.52	2.59±0.58	0.89±0.16	0.97±0.22	0.86±0.15
	F	3.13±0.71	2.99±0.58	0.70±0.12	1.52±0.40	0.88±0.17
	T	3.20±0.43	2.79±0.41	0.80±0.10	1.24±0.23	0.87±0.11
Cobb 500	M	2.29±0.59	2.94±0.60	0.85±0.13	1.36±0.26	1.34±0.19
	F	4.26±1.19	2.63±0.83	0.63±0.19	1.39±0.26	1.23±0.21
	T	3.28±0.63	2.79±0.47	0.74±0.11	1.37±0.17	1.29±0.13
Source of variation		Sig.	Sig.	Sig.	Sig.	Sig.
Genotype (G)		0.917	0.998	0.721	0.652	***0.020***
Sex (S)		0.227	0.939	0.172	0.310	0.817
G × S		0.165	0.573	0.911	0.367	0.697

F = female; M = male, T=Total; WL = Wattle length; WW = Wattle width; EW = Eye width; NL = Nose length; FW = Facial width

Genotype effects on other FA traits were not statistically significant; however, a marginal genotype effect was observed for wattle length (WL) (P = 0.073), indicating a trend toward increasing asymmetry differences that may become more pronounced with increasing age. Similar age-dependent increases in trait-specific asymmetry have been reported in broiler chickens as growth intensity increases (Mendes, 2008).

Sex significantly influenced WL FA at week 6 (P = 0.017), with males displaying higher asymmetry than females. This contrasts with observations at earlier ages and suggests a shift in developmental stability patterns as chicks approach sexual maturation. Increased androgen activity and sex-specific energy allocation in males during later growth stages may contribute to this pattern, as endocrine regulation plays a central role in shaping secondary sexual traits and growth trajectories (Møller & Pomiankowski 1993; Møller & Swaddle, 1997; Lengyel et al., 2024). No significant genotype × sex interactions were detected for FA traits, indicating that genotype-related differences in asymmetry were expressed similarly in both sexes at this age.

For RA indices at week 6, genotype had a significant effect on FW RA (P = 0.020), with Cobb500 broilers again exhibiting higher relative asymmetry than Anadolu–T chicks. This result corroborates the FA findings and reinforces facial width as a particularly sensitive indicator of developmental instability at later growth ages. Previous studies have shown that certain cranial and head-related traits exhibit heightened sensitivity to developmental perturbations, especially in fast-growing genotypes (Van Nuffel et al., 2007; Mendes, 2008).

In contrast, genotype effects on RA values for wattle length (WL), wattle width (WW), eye width (EW), and nose length (NL) were not significant, suggesting relatively stable bilateral development for these traits across genotypes at this age. Neither sex nor genotype × sex interactions significantly affected RA traits, indicating that relative asymmetry at week 6 was primarily genotype-dependent rather than sex-driven. This contrasts with earlier developmental stages, where sex-related effects on asymmetry were more pronounced, and highlights the dynamic nature of asymmetry expression across growth phases.

Overall, developmental instability at 6 weeks manifests in a trait-specific way, with facial width as the most responsive asymmetry indicator. The consistently higher FA and RA values observed in Cobb500 broilers suggest that rapid growth and intensive genetic selection may compromise developmental stability, particularly in cranial traits. Similar associations between fast growth, selection pressure, and elevated asymmetry have been widely reported in poultry (Møller et al., 1995; Van Nuffel et al., 2007; Mendes, 2008).

The reduced influence of sex on RA indices at this age suggests that genetic background outweighs hormonal effects in determining bilateral symmetry during later growth stages. Collectively, these findings support the use of FA

and RA indices derived from head morphology as sensitive, non-invasive tools for assessing developmental stability, genotype-related differences, and potential welfare implications in broiler production systems.

Fluctuating and Relative Asymmetry of Bilateral Leg at 3 Weeks of Age

The fluctuating asymmetry (FA) and relative asymmetry (RA) indices derived from bilateral leg (tarsometatarsus) measurements at 3 weeks of age are presented in Table 6. These indices were used to assess early developmental stability of skeletal traits, which are critical for locomotion and overall welfare in broiler chickens.

At week 3, no significant effects of genotype, sex, or genotype × sex interaction were detected for FA indices of tarsometatarsus length (TML), joint width (JW), tarsometatarsus width at 1 cm above the spur (TMW-1), or tarsometatarsus width at the spur level (TMW-SL) ($P > 0.05$ for all traits). These results indicate a high degree of bilateral stability in leg morphology during early growth, regardless of genetic background or sex.

Although numerical differences were observed such as higher TML FA values in Anadolu-T males and higher TML FA in Cobb500 females these variations did not reach statistical significance. This suggests that early leg development is relatively well buffered against genetic and sex-related variation, likely due to the fundamental functional importance of limb symmetry during the initial growth phase.

Similarly, RA indices at week 3 were not significantly affected by genotype, sex, or their interaction for any of the evaluated leg traits. Both genotypes exhibited comparable RA values for TML, JW, TMW-1, and TMW-SL, indicating consistent proportional bilateral development across genetic lines. The absence of sex effects further suggests that sexual dimorphism in leg asymmetry had not yet emerged at this early age.

Slightly higher RA values for tarsometatarsus width traits in Anadolu-T chicks compared with Cobb500 were observed numerically; however, these differences remained statistically non-significant. This pattern may reflect subtle genotype-specific growth trajectories that become more evident at later ages rather than true early developmental instability.

Table 6. fluctuating (FA) and relative (RA) asymmetry analyses derived from bilateral tarsometatarsus measurements at week 3

Çizelge 6. 3. haftada yapılan iki taraflı tarsometatarsus ölçümlerinden elde edilen dalgalı (FA) ve göreceli (RA) asimetri analizleri

Items		3. Week Fluctuating Asymmetry (FA)			
Genotype	Sex	TML±SEM	JW±SEM	TMW-1±SEM	TMW-SL±SEM
Anadolu - T	M	0.63±0.15	0.46±0.07	0.3±0.06	0.28±0.04
	F	0.56±0.12	0.41±0.08	0.31±0.07	0.40±0.10
	T	0.59±0.10	0.43±0.05	0.3±0.05	0.34±0.06
Cobb 500	M	0.35±0.09	0.41±0.10	0.27±0.07	0.26±0.03
	F	0.69±0.15	0.29±0.06	0.19±0.04	0.29±0.06
	T	0.52±0.08	0.35±0.06	0.23±0.04	0.27±0.03
Source of variation		Sig.	Sig.	Sig.	Sig.
Genotype (G)		0.548	0.297	0.230	0.289
Sex (S)		0.304	0.268	0.567	0.249
G × S		0.106	0.669	0.427	0.473
Items		3. Week Relative Asymmetry (RA)			
Genotype	Sex	TML±SEM	JW±SEM	TMW-1±SEM	TMW-SL±SEM
Anadolu - T	M	0.22±0.05	0.53±0.08	0.66±0.13	0.62±0.09
	F	0.20±0.04	0.52±0.10	0.71±0.16	0.94±0.24
	T	0.21±0.03	0.52±0.06	0.69±0.10	0.78±0.13
Cobb 500	M	0.11±0.03	0.44±0.11	0.54±0.15	0.51±0.07
	F	0.23±0.05	0.34±0.07	0.39±0.09	0.61±0.13
	T	0.17±0.03	0.39±0.06	0.46±0.09	0.56±0.07
Source of variation		Sig.	Sig.	Sig.	Sig.
Genotype (G)		0.361	0.142	0.104	0.142
Sex (S)		0.220	0.527	0.711	0.157
G × S		0.133	0.603	0.480	0.468

F = female; M = male; T = Total; TML = Tarsometatarsus length; JW = Joint width; TMW-1 = Tarsometatarsus width at 1 cm above the spur; TMW-SL = Tarsometatarsus width at the spur level

Overall, the results demonstrate that leg-related asymmetry at 3 weeks of age is minimal and not significantly influenced by genotype or sex. This finding suggests a strong developmental canalization of skeletal traits during early growth, likely driven by the mechanical and physiological demands of supporting rapid body mass increase.

Compared with head-related traits, which showed greater sensitivity to sex and genotype at similar ages, leg morphology appears more developmentally stable in early life.

These findings support the view that FA and RA indices of tarsometatarsus traits are more informative at later growth ages, when increased body weight and differential growth rates may challenge skeletal stability. Consequently, early leg asymmetry may have limited value as an indicator of developmental stress, whereas age-dependent changes may provide more meaningful insights into genotype- and welfare-related differences.

Table 7 presents the effects of genotype, sex, and their interaction on fluctuating (FA) and relative (RA) asymmetry indices of bilateral tarsometatarsus measurements at week 6. The maximum, median, and minimum values of tarsometatarsus length, joint width, tarsometatarsus width at 1 cm above the spur, and tarsometatarsus width at the spur level according to genotype and sex at week 6 are illustrated in Figures 11, 12, 13, and 14, respectively. The evaluated traits included tarsometatarsus length (TML), joint width (JW), tarsometatarsus width at 1 cm above the spur (TMW-1), and tarsometatarsus width at the spur level (TMW-SL).

Table 7. Fluctuating (FA) and relative (RA) asymmetry analyses derived from bilateral tarsometatarsus measurements at week 6

Çizelge 7. 6. haftada iki taraflı tarsometatarsus ölçümlerinden elde edilen dalgalı (FA) ve göreceli (RA) asimetri analizleri

Items		6. Week Fluctuating Asymmetry (FA)			
Genotype	Sex	TML±SEM	JW±SEM	TMW-1±SEM	TMW-SL±SEM
Anadolu - T	M	0.41±0.12	0.95±0.39	0.23±0.04	0.36±0.06
	F	0.30±0.09	0.40±0.08	0.18±0.05	0.25±0.05
	T	0.35±0.07	0.68±0.2	0.21±0.03	0.31±0.04
Cobb 500	M	0.29±0.09	0.56±0.09	0.33±0.06	0.38±0.07
	F	0.58±0.14	0.32±0.06	0.32±0.06	0.42±0.09
	T	0.43±0.08	0.44±0.05	0.33±0.04	0.4±0.05
Source of variation		Sig.	Sig.	Sig.	Sig.
Genotype (G)		0.464	0.252	***0.025***	0.148
Sex (S)		0.400	0.055	0.612	0.555
G × S		0.075	0.447	0.680	0.234
Items		6. Week Relative Asymmetry (RA)			
Genotype	Sex	TML±SEM	JW±SEM	TMW-1±SEM	TMW-SL±SEM
Anadolu - T	M	0.11±0.03	0.95±0.4	0.43±0.08	0.68±0.12
	F	0.09±0.03	0.45±0.09	0.38±0.09	0.52±0.11
	T	0.10±0.02	0.7±0.21	0.4±0.06	0.6±0.08
Cobb 500	M	0.08±0.03	0.51±0.08	0.56±0.1	0.66±0.11
	F	0.16±0.04	0.33±0.06	0.62±0.12	0.83±0.18
	T	0.12±0.02	0.42±0.05	0.59±0.07	0.75±0.1
Source of variation		Sig.	Sig.	Sig.	Sig.
Genotype (G)		0.444	0.191	0.056	0.246
Sex (S)		0.259	0.111	0.940	0.967
G × S		0.081	0.445	0.551	0.189

F = female; M = male; T = Total; TML = Tarsometatarsus length; JW = Joint width; TMW-1 = Tarsometatarsus width at 1 cm above the spur; TMW-SL = Tarsometatarsus width at the spur level

For FA indices, genotype had a significant effect only on TMW-1 ($P = 0.025$), indicating that differences in growth patterns between Anadolu-T and Cobb500 genotypes influenced bilateral symmetry of bone width above the spur. Cobb500 chickens exhibited numerically higher FA values for TMW-1 compared with Anadolu-T, suggesting greater developmental instability in this trait, likely associated with the rapid growth rate and higher metabolic demands characteristic of fast-growing commercial broilers.

No significant genotype effect was observed for TML, JW, or TMW-SL ($P > 0.05$). Similarly, sex did not significantly affect FA indices for any measured trait, although a tendency toward higher FA in males for joint width was observed ($P = 0.055$). The genotype × sex interaction was also not significant, indicating that genotypic differences in asymmetry were consistent across sexes.

Regarding RA indices, no significant effects of genotype, sex, or genotype × sex interaction were detected for any of the evaluated traits ($P > 0.05$). Although Cobb500 chicks showed numerically higher RA values for TMW-1 and TMW-SL, these differences were not statistically significant. This suggests that when asymmetry is expressed relative to trait size, compensatory growth mechanisms may reduce apparent genotypic differences in bilateral development.

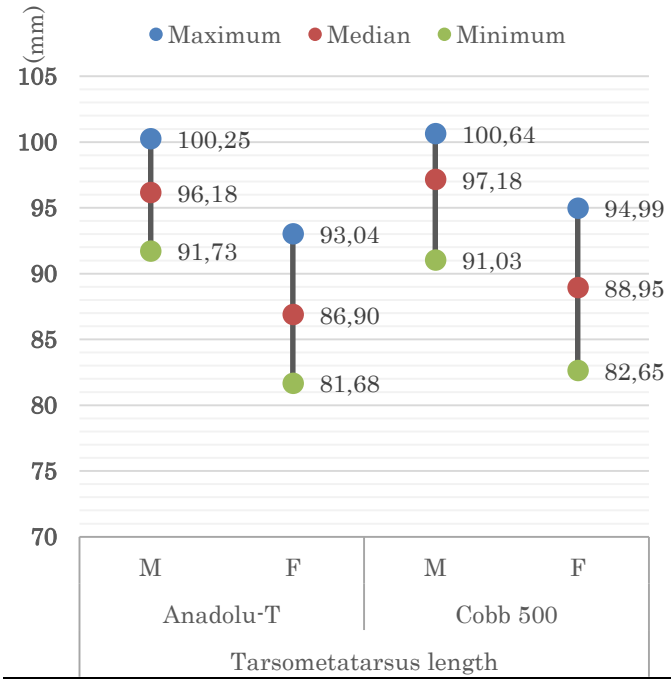


Figure 11. Maximum, Median, and Minimum Values of Tarsometatarsus length by Genotype and Sex at Week 6

Şekil 11. 6. haftada genotip ve cinsiyete göre tarsometatarsus uzunluğunun maksimum, medyan ve minimum değerleri.

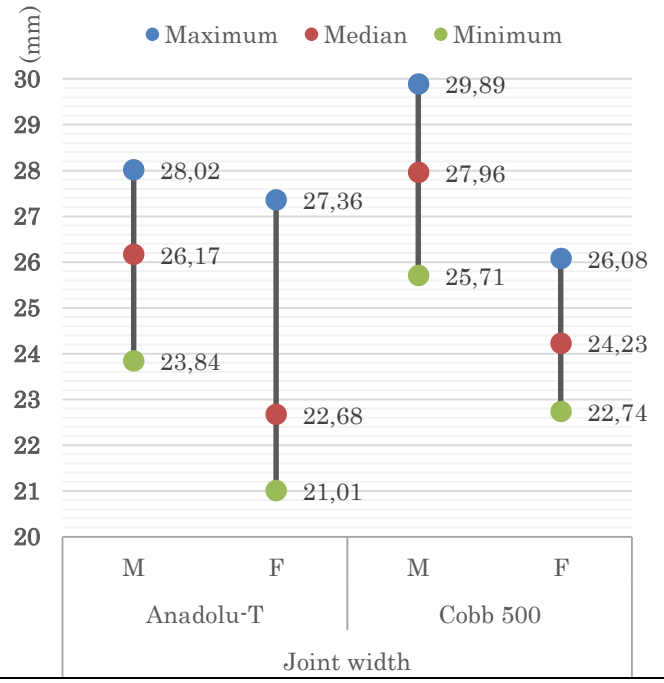


Figure 12. Maximum, Median, and Minimum Values of Joint width by Genotype and Sex at Week 6

Şekil 12. 6. haftada genotip ve cinsiyete göre eklem genişliğinin maksimum, medyan ve minimum değerleri

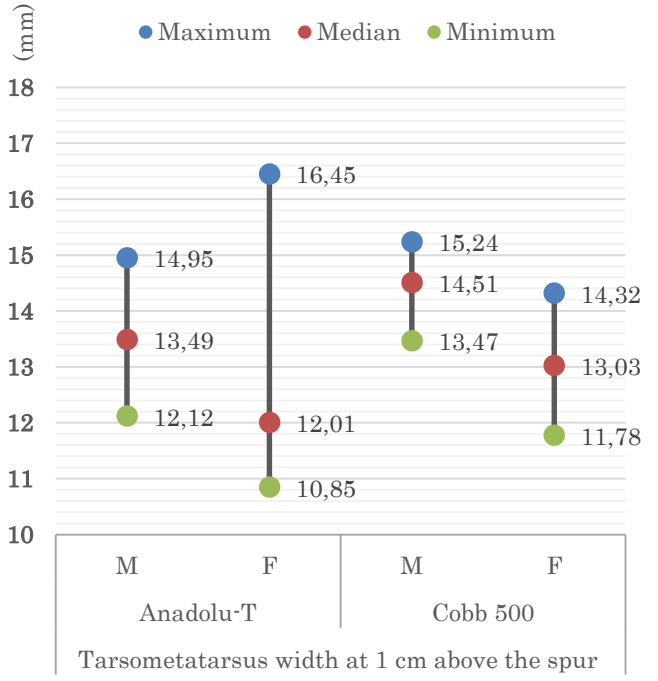


Figure 13. Maximum, Median, and Minimum Values of Tarsometatarsus width at 1 cm above the spur by Genotype and Sex at Week 6

Şekil 13. 6. haftada, mahmuzun 1 cm yukarısından alınan tarsometatarsus genişliğinin genotip ve cinsiyete göre maksimum, medyan ve minimum değerleri.

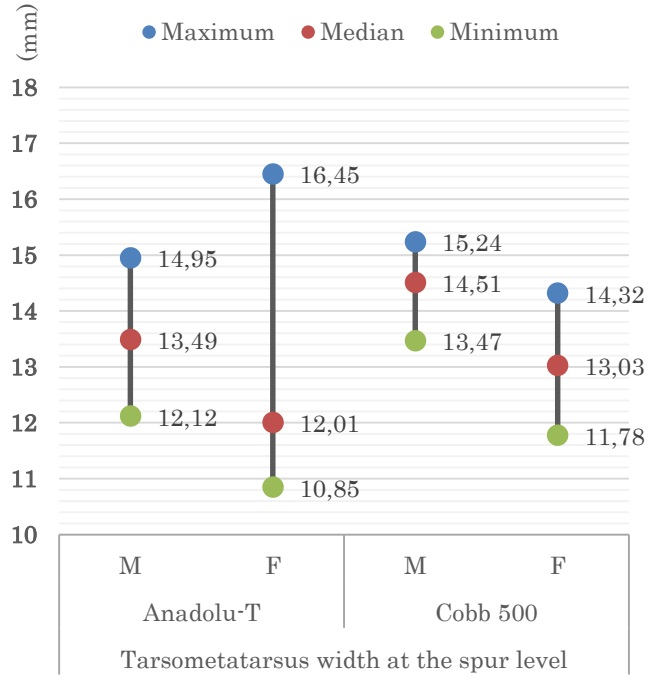


Figure 14. Maximum, Median, and Minimum Values of Tarsometatarsus width at the spur level by Genotype and Sex at Week 6

Şekil 14. 6. haftada, mahmuz seviyesi tarsometatarsus genişliğinin genotip ve cinsiyete göre maksimum, medyan ve minimum değerleri.

The significant genotype effect detected for FA in TMW-1 highlights this trait as a sensitive indicator of developmental instability under intensive growth conditions. The absence of consistent effects across other traits and in RA indices suggests that skeletal asymmetry in broilers is trait-specific and more pronounced in bone width measurements than in longitudinal growth parameters. These findings support previous reports indicating that fast-growing genotypes may exhibit localized asymmetry in skeletal traits due to increased mechanical loading and accelerated ossification processes.

Overall, the results indicate that genotype exerts a limited but trait-specific influence on bilateral tarsometatarsus asymmetry at week 6, while sex plays a negligible role. The relatively low asymmetry levels observed in Anadolu-T chickens may reflect greater developmental stability and adaptability compared with the commercial Cobb500 genotype.

At Week 3 (Table 8), FA analysis revealed that genotype had a significant effect on several foot traits. Significant genotype-related differences were observed for hind toe length (HTL) (P = 0.034), fourth phalanx length of the outer toe (OP4L) (P = 0.027), and third phalanx length of the middle toe (MP3L) (P = 0.050). In general, Cobb500 broilers exhibited higher FA values for these traits compared with the Anadolu-T genotype, indicating greater developmental instability in distal foot structures of the fast-growing genotype at early age.

Sex effects at Week 3 were mostly non-significant; however, MP3L showed a significant sex effect (P = 0.005), with females tending to exhibit higher FA values than males. In addition, a significant genotype × sex interaction was detected for middle toe claw length (MTCL) (P = 0.005), suggesting that the expression of asymmetry in this trait differed between males and females depending on genotype.

Table 8. Fluctuating (FA) and relative (RA) asymmetry analyses derived from foot measurements according to genotype and sex at week 3

Çizelge 8. 3. haftada genotip ve cinsiyete göre ayak ölçümlerinden elde edilen dalgalı (FA) ve göreceli (RA) Asimetri analizleri

Items		3. Week Fluctuating Asymmetry (FA)						
Genotype	Sex	HTL±SEM	OTCL±SEM	OP4L±SEM	OP3L±SEM	MTCL±SEM	MP4L±SEM	MP3L±SEM
	M	0.63±0.15	0.7±0.13	0.43±0.07	0.51±0.14	0.48±0.09	0.44±0.1	0.44±0.08
Anadolu - T	F	0.88±0.16	1.87±1.24	0.33±0.06	0.36±0.08	0.89±0.12	0.53±0.08	0.73±0.15
	T	0.75±0.11	1.29±0.62	0.38±0.04	0.43±0.08	0.69±0.08	0.49±0.06	0.59±0.09
	M	1.14±0.23	0.75±0.2	0.61±0.13	0.5±0.09	0.82±0.12	0.55±0.08	0.55±0.09
Cobb 500	F	1.65±0.55	0.57±0.15	0.56±0.1	1.33±0.64	0.6±0.11	0.93±0.3	1.38±0.36
	T	1.4±0.27	0.66±0.12	0.59±0.08	0.92±0.3	0.71±0.08	0.74±0.14	0.97±0.18
		Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.
Genotype (G)		***0.034***	0.330	***0.027***	0.118	0.841	0.106	***0.050***
Sex (S)		0.205	0.438	0.414	0.266	0.376	0.140	***0.005***
G × S		0.661	0.297	0.769	0.115	0.005	0.357	0.166
Items		3. Week Relative Asymmetry (RA)						
Genotype	Sex	HTL±SEM	OTCL±SEM	OP4L±SEM	OP3L±SEM	MTCL±SEM	MP4L±SEM	MP3L±SEM
	M	0.77±0.18	2.09±0.36	1.92±0.31	1.07±0.28	1.17±0.2	1.67±0.39	0.63±0.12
Anadolu - T	F	1.11±0.2	3.42±1.62	1.56±0.27	0.82±0.18	2.13±0.29	2.09±0.3	1.09±0.23
	T	0.94±0.14	2.76±0.82	1.74±0.21	0.94±0.17	1.65±0.19	1.88±0.25	0.86±0.13
	M	1.27±0.25	2.16±0.63	2.03±0.4	3.78±3.01	1.9±0.3	1.63±0.25	0.69±0.11
Cobb 500	F	2.18±0.89	1.7±0.46	2.02±0.32	3.37±2.17	1.42±0.25	2.77±0.75	2±0.62
	T	1.72±0.42	1.93±0.38	2.03±0.24	3.58±1.77	1.66±0.19	2.2±0.37	1.34±0.3
		Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.
Genotype		0.080	0.371	0.376	0.150	0.958	0.463	0.127
Sex		0.162	0.633	0.573	0.857	0.348	***0.079***	***0.006***
[10]G × S		0.521	0.329	0.580	0.964	0.007	0.418	0.176

F = female; M = male; T = Total; HTL = Hind toe length; OTCL = Outer toe claw length; OP4L = Fourth phalanx length of the outer toe; OP3L = Third phalanx length of the outer toe; MTCL= Middle toe claw length; MP4L=Fourth phalanx length of the middle toe; MP3L= Third phalanx length of the middle toe

By Week 6 (Table 9), the magnitude and statistical significance of FA differences were markedly reduced. No significant effects of genotype, sex, or their interaction were detected for most foot traits (P > 0.05). Only near-significant tendencies were observed for genotype effects on HTL (P = 0.066) and MTCL (P = 0.073), indicating that asymmetry differences evident at Week 3 tended to diminish with age. This pattern suggests a degree of developmental stabilization or compensatory growth in foot morphology as chicks matured.

Relative asymmetry analysis at Week 3 showed no significant genotype effects for any foot trait (P > 0.05), although

Cobb500 chicks generally displayed numerically higher RA values, particularly for OP3L and MP3L, reflecting size-adjusted asymmetry in toe phalanges. A significant sex effect was detected for MP3L (P = 0.006), consistent with FA findings and indicating greater proportional asymmetry in females for this trait. Moreover, a significant genotype × sex interaction was observed for MTCL (P = 0.007), reinforcing the idea that sex-related growth patterns modulate asymmetry expression differently across genotypes.

At Week 6, RA values again showed no significant genotype effects for any trait. However, a significant sex effect emerged for MP3L (P = 0.028), with females exhibiting higher RA values than males. Similar to FA, the overall reduction in significant effects at Week 6 suggests that relative asymmetry in foot traits becomes less pronounced as skeletal growth progresses.

When Tables 8 and 9 are evaluated together, a clear age-related pattern emerges. Asymmetry in foot traits was more pronounced and biologically informative at Week 3, particularly in phalangeal and toe length measurements. These early differences were mainly associated with genotype and, to a lesser extent, sex.

By Week 6, both FA and RA indices indicated a general attenuation of asymmetry, implying improved developmental stability of foot morphology with advancing age. The reduction in genotype-related differences may reflect adaptive skeletal remodelling or the completion of rapid early growth phases, especially in Cobb500 broilers. Foot and toe structures are critical for locomotion, weight distribution, and overall welfare in broiler chickens. The higher FA and RA values observed in Cobb500 chicks at Week 3 suggest that rapid early growth may impose transient developmental stress on distal limb structures. However, the subsequent decline in asymmetry by Week 6 indicates that these effects may be partially compensated over time, reducing long-term structural imbalance.

Overall, these findings support the use of foot-related asymmetry indices, particularly at early ages, as sensitive indicators of developmental instability and growth-related stress in different broiler genotypes.

Table 9. Fluctuating (FA) and relative (RA) asymmetry analyses derived from foot measurements according to genotype and sex at week 6

Çizelge 9. 6. haftada genotip ve cinsiyete göre ayak ölçümlerinden elde edilen dalgalı (FA) ve göreceli (RA) Asimetri analizleri

Items		6. Week Fluctuating Asymmetry (FA)						
Genotype	Sex	HTL±SEM	OTCL±SEM	OP4L±SEM	OP3L±SEM	MTCL±SEM	MP4L±SEM	MP3L±SEM
Anadolu - T	M	1.2±0.21	0.68±0.16	0.55±0.06	0.54±0.09	0.75±0.17	0.46±0.07	0.54±0.09
	F	0.75±0.13	0.45±0.1	0.44±0.09	0.51±0.11	1.1±0.43	0.58±0.12	0.8±0.15
	T	0.97±0.13	0.57±0.09	0.49±0.05	0.53±0.07	0.93±0.23	0.52±0.07	0.67±0.09
Cobb 500	M	1.52±0.26	0.55±0.1	0.47±0.09	0.43±0.11	0.5±0.1	0.44±0.08	0.48±0.09
	F	1.63±0.59	0.56±0.09	0.54±0.09	0.61±0.13	0.47±0.1	0.36±0.07	0.65±0.13
	T	1.57±0.29	0.55±0.06	0.5±0.06	0.52±0.08	0.49±0.07	0.4±0.05	0.57±0.08
		Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.
Genotype (G)		0.066	0.895	0.880	0.970	0.073	0.172	0.392
Sex (S)		0.600	0.342	0.768	0.469	0.510	0.841	***0.064***
G × S		0.396	0.293	0.294	0.325	0.442	0.285	0.694
Items		6. Week Relative Asymmetry (RA)						
Genotype	Sex	HTL±SEM	OTCL±SEM	OP4L±SEM	OP3L±SEM	MTCL±SEM	MP4L±SEM	MP3L±SEM
Anadolu - T	M	1.15±0.2	1.47±0.34	1.76±0.18	0.89±0.15	1.41±0.33	1.31±0.22	0.61±0.1
	F	0.8±0.14	1±0.22	1.58±0.35	0.94±0.2	1.93±0.72	1.69±0.37	1±0.19
	T	0.97±0.12	1.24±0.2	1.67±0.19	0.91±0.12	1.67±0.39	1.5±0.21	0.8±0.11
Cobb 500	M	1.49±0.25	1.25±0.25	1.48±0.29	0.67±0.17	0.94±0.19	1.24±0.25	0.53±0.11
	F	2.04±0.92	1.27±0.2	1.79±0.31	1.04±0.22	0.89±0.19	1.08±0.22	0.77±0.16
	T	1.77±0.43	1.26±0.15	1.64±0.2	0.85±0.13	0.91±0.13	1.16±0.16	0.65±0.09
		Sig.	Sig.	Sig.	Sig.	Sig.	Sig.	Sig.
Genotype		0.082	0.919	0.901	0.726	0.074	0.207	0.284
Sex		0.825	0.371	0.816	0.254	0.581	0.680	***0.028***
[10]G × S		0.322	0.332	0.377	0.367	0.492	0.308	0.606

F = female; M = male; T = Total; HTL = Hind toe length; OTCL = Outer toe claw length; OP4L = Fourth phalanx length of the outer toe; OP3L = Third phalanx length of the outer toe; MTCL= Middle toe claw length; MP4L=Fourth phalanx length of the middle toe; MP3L= Third phalanx length of the middle toe

Fluctuating asymmetry (FA), defined as small, random deviations from perfect bilateral symmetry, is widely used as an indicator of developmental instability in animals, including poultry, and can vary with age. Campo et al. (2005) highlighted that fluctuating asymmetry (FA) in chickens is influenced by both genetic and environmental factors. In the second experiment of their study, FA values were observed to vary across different ages, with certain morphological traits showing statistically significant changes, particularly around sexual maturity (8, 12, 16, … 40 weeks). Similarly, classical studies on FA, such as those by Møller et al. (1997), have demonstrated that rapid growth and environmental stress can increase FA, with faster-growing chickens generally exhibiting higher

asymmetry. This suggests that age-related changes in metabolic and developmental demands may affect FA. Moreover, Klingenberg (2003) emphasized that FA not only reflects developmental instability resulting from random variation during growth but can also provide insights into the developmental origins of morphological integration and the mechanisms that buffer developmental noise in bilateral traits.

Sex-related differences in morphological asymmetry observed in this study, particularly the higher levels of asymmetry in traits such as eye width and beard size in females, may be explained by underlying physiological and developmental mechanisms (Helle et al., 2010). One plausible explanation involves sex-specific growth rates and differences in resource allocation strategies, which can lead to increased developmental instability (Watson & Thornhill, 1994; Polak & Starmer, 2005). These findings are consistent with previous research on avian species (Helle et al., 2010; Cuervo & Møller, 1999; Aparicio & Bonal, 2002). The relationship between sex and asymmetry is complex and species-dependent, and is often influenced by ecological pressures, life-history strategies, and sexual selection (Watson & Thornhill, 1994; Liker et al., 2013; Polak & Starmer, 2005). In some cases, females may exhibit higher asymmetry due to greater energetic constraints, whereas in others, males show increased asymmetry associated with the costs of developing secondary sexual traits (Møller, 1992; Cuervo & Møller, 1999).

Descriptive statistics of foot measurements

Table 10 presents the descriptive statistics of foot measurements according to genotype and sex at Week 6. Overall, male chicks tended to exhibit higher mean values than females for most foot traits, particularly for hind toe length (HTL) and third phalanx length of the outer toe (OP3L), reflecting sex-related differences in skeletal size at this age.

Table 10. Descriptive statistics of foot measurements according to genotype and sex at week 6
Çizelge 10. 6. haftada genotip ve cinsiyete göre ayak ölçümlerinin tanımlayıcı istatistikleri

Items		6. week					
Genotype	Sex	Maximum	Median	Minimum	Mean	Std. E.M.	Range
				Hind toe length (HTL)			
Anadolu - T	M	27.83	26.32	24.20	25.99	0.306	3.63
	F	27.87	23.35	21.54	23.49	0.362	6.33
Cobb 500	M	29.28	24.90	22.02	25.21	0.495	7.26
	F	25.03	22.73	16.26	22.44	0.526	8.77
				Outer toe claw length (OTCL)			
Anadolu - T	M	12.90	11.45	10.02	11.55	0.189	2.88
	F	16.85	11.04	9.10	11.29	0.408	7.75
Cobb 500	M	12.15	11.21	9.57	10.99	0.205	2.58
	F	12.13	10.88	9.89	10.90	0.155	2.24
				Fourth phalanx length of the outer toe (OP4L)			
Anadolu - T	M	8.84	7.73	6.54	7.80	0.161	2.30
	F	8.13	7.28	5.25	7.17	0.173	2.88
Cobb 500	M	8.84	8.01	7.10	7.99	0.110	1.74
	F	8.14	7.59	6.23	7.48	0.113	1.91
				Third phalanx length of the outer toe (OP3L)			
Anadolu - T	M	16.27	14.90	13.65	15.07	0.187	2.62
	F	18.81	13.95	12.15	14.06	0.380	6.66
Cobb 500	M	17.81	16.55	14.31	16.48	0.211	3.50
	F	16.11	14.86	12.78	14.82	0.210	3.33
				Middle toe claw length (MTCL)			
Anadolu - T	M	14.82	13.62	12.37	13.53	0.175	2.45
	F	19.18	13.65	11.78	13.90	0.393	7.40
Cobb 500	M	14.62	13.34	11.90	13.30	0.181	2.72
	F	14.78	13.21	11.87	13.39	0.222	2.91
				Fourth phalanx length of the middle toe (MP4L)			
Anadolu - T	M	1.03	0.46	0.01	0.46	0.072	1.02
	F	1.75	0.52	0.05	0.57	0.122	1.70
Cobb 500	M	1.36	0.36	0.04	0.43	0.083	1.32
	F	0.85	0.31	0.01	0.35	0.063	0.84
				Third phalanx length of the middle toe (MP3L)			
Anadolu - T	M	1.16	0.47	0.10	0.53	0.089	1.16
	F	2.18	0.60	0.04	0.79	0.147	2.14
Cobb 500	M	1.39	0.38	0.15	0.48	0.094	1.24
	F	1.38	0.56	0.07	0.65	0.119	1.31

F = female; M = male

Between genotypes, Cobb500 males showed the highest maximum and mean HTL values, whereas Anadolu-T females generally displayed the lowest mean HTL and the widest range, indicating greater individual variability. Similar patterns of variability were observed for outer toe claw length (OTCL) and middle toe claw length (MTCL), where Anadolu-T females exhibited notably larger ranges compared with other groups.

Phalangeal measurements (OP4L, OP3L, MP4L, and MP3L) showed less pronounced genotype-related differences, with relatively close mean values between Anadolu-T and Cobb500 chicks. However, female chicks, especially in the Anadolu-T genotype, tended to show higher variability, as indicated by larger ranges and standard errors in MP4L and MP3L.

In general, the results suggest that while absolute foot dimensions at Week 6 were influenced by sex and genotype, within-group variability particularly among females was substantial for several traits, which may be associated with differences in growth patterns and developmental stability at later growth stages.

CONCLUSION

The present study clearly indicates that genotype- and sex-related differences in morphological traits influence developmental stability in broiler chickens in an age-dependent manner. Comparisons between the local Anadolu-T genotype and the commercial Cobb500 genotype revealed that these effects were most evident during the early growth period.

At Week 3, analyses based on fluctuating (FA) and relative asymmetry (RA) indices demonstrated that Cobb500 broilers generally exhibited higher asymmetry values in several head, foot, and tarsometatarsus traits. These findings suggest a temporary reduction in developmental stability during early ontogeny, particularly in distal skeletal structures such as toe phalanges and claws, which are known to be highly sensitive to accelerated growth and metabolic demands (Van Valen, 1962; Møller & Swaddle, 1997; Yang, 1998). In contrast, the Anadolu-T genotype showed lower or more consistent asymmetry levels, reflecting a greater capacity for developmental buffering under identical rearing conditions.

By Week 6, the majority of genotype-related differences in FA and RA indices were no longer statistically significant. This reduction in asymmetry with age indicates a process of developmental stabilization and compensatory skeletal growth, suggesting that early asymmetries do not necessarily persist into later stages when chicks are managed under controlled and uniform conditions (Klingenberg, 2003; Møller & Manning, 2003). Such patterns support the view that developmental instability in fast-growing genotypes may be transient rather than permanent.

Sex-related effects were generally trait-specific, with females often displaying greater variability and higher asymmetry values, particularly in selected foot and phalangeal measurements. These findings underline the influence of sex-dependent growth trajectories and skeletal maturation rates on morphological variability (Mendes et al., 2007; Eleroğlu & Yıldırım, 2019).

Descriptive statistics of foot measurements at Week 6 further complemented the asymmetry analyses, showing that males tended to have larger absolute dimensions, whereas females especially within the Anadolu-T genotype exhibited wider ranges and higher within-group variability. This variability likely reflects differences in growth uniformity and developmental timing rather than pathological impairment.

From a welfare perspective, the absence of marked differences between genotypes in tonic immobility duration and cloacal temperature indicates that both Anadolu-T and Cobb500 broilers were able to adapt similarly to the experimental housing conditions, without evidence of excessive stress or fear responses (Campo et al., 2000; Shahin & El-Azeem, 2005). Moreover, the maintenance of bilateral symmetry across most traits suggests that appropriate management conditions can mitigate potential welfare risks, even in high-performance genotypes (Yalçın & Siegel, 2003; Møller et al., 1999).

In conclusion, the Anadolu-T genotype emerges as a robust local alternative, exhibiting high developmental stability and favorable welfare characteristics comparable to those of modern commercial hybrids. While Cobb500 broilers displayed superior growth performance, the transient increase in asymmetry observed at early ages highlights the biological cost associated with rapid growth. These findings emphasize the importance of incorporating developmental stability indicators, such as FA and RA, alongside conventional performance traits when evaluating broiler genotypes.

Future studies investigating different environmental stressors, including lighting programs and stocking density, may further clarify the resilience and long-term welfare potential of local genotypes such as Anadolu-T. For practical poultry management, it is recommended that environmental conditions (e.g., feeding strategies, stocking density) be optimized in fast-growing genotypes such as Cobb 500 to support developmental stability during the early growth period. Regarding the conservation of local genotypes, the high developmental stability and favorable welfare parameters observed in the Anadolu-T genotype highlight its potential as an advantage for preserving

native genetic resources and for integration into sustainable production systems. For future research, examining the effects of different environmental stressors (e.g., heat stress, high stocking density) on asymmetry will help determine the resilience thresholds of different genotypes.

Ethics Approval

This study was approved by the Local Ethics Committee for Animal Experiments of Kahramanmaraş Sütçü İmam University (Approval date: 28 February 2025; Approval number: 2025/02). Morphological measurements were performed by the same researcher and the average value was used for analysis; however, no reliability test was conducted.

Limitations

Several limitations of the present study should be acknowledged. First, the experiment was conducted at a single location and during a single production cycle, which may limit the generalizability of the findings. Second, sex determination of the animals used in the experiment was performed subjectively at 21 days of age based on morphological characteristics. Third, the sample size (240 chicks in total, with 64 animals included in asymmetry measurements) may restrict the statistical power and the extent to which the results can be generalized. Fourth, measurement error in fluctuating asymmetry could not be completely controlled. Finally, welfare assessment was limited to tonic immobility (TI) and righting time (RT), while a more comprehensive evaluation including behavioral, physiological, and health indicators was not conducted.

Acknowledgments

In this study, we would like to thank TAGEM for providing the animal material we used from project number TAGEM/HAYSÜT/G/22/A4/P4/6230.

Contribution Rate Statement Summary of Researchers

Yazarlar makaleye eşit oranda katkı sağlamış olduklarını beyan eder.

The authors declare that they have contributed equally to the article.

Conflict of Interest

Makale yazarları aralarında herhangi bir çıkar çatışması olmadığını beyan ederler.

The authors of this article declare that they have no conflict of interest.

REFERENCES

Adu-Asiamah, P., Zhang, Y., Amoah, K., Leng, Q. Y., Zheng, J. H., Yang, H., Zhang, W. L., & Zhang, L. (2021). Evaluation of physiological and molecular responses to acute heat stress in two chicken breeds. *Animal, 15*(2), Article 100106. https://doi.org/10.1016/j.animal.2020.100106

Anderson, K. E., & Jones, D. R. (2012). Effect of genetic selection on growth parameters and tonic immobility in Leghorn pullets. *Poultry Science, 91*(3), 765–770. https://doi.org/10.3382/ps.2011-01799

Aparicio, J. M., & Bonal, R. (2002). Why do some traits show higher fluctuating asymmetry than others? A test with tail feathers of birds. *Heredity, 89*(2), 139–144. https://doi.org/10.1038/sj.hdy.6800118

Broom, D. M. (2006). Behaviour and welfare in relation to pathology. *Applied Animal Behaviour Science, 97*(1), 73–83. https://doi.org/10.1016/j.applanim.2005.11.019

Campbell, A. M., Johnson, A. M., Persia, M. E., & Jacobs, L. (2022). Effects of housing system on anxiety, chronic stress, fear, and immune function in Bovan Brown laying hens. *Animals, 12*(14), Article 1803. https://doi.org/10.3390/ani12141803

Campo, J. L., Gil, M. G., Muñoz, I., & Alonso, M. (2000). Relationships between bilateral asymmetry and tonic immobility reaction or heterophil to lymphocyte ratio in five breeds of chickens. *Poultry Science, 79*(4), 453–459. https://doi.org/10.1093/ps/79.4.453

Campo, J. L., Gil, M. G., Torres, O., & Dávila, S. G. (2002). Fluctuating asymmetry in male secondary sexual traits: Association with indicators of stress in chickens. *Archiv für Geflügelkunde, 66*(2), 85–89.

Campo, J. L., Gil, M. G., Dávila, S. G., & Muñoz, I. (2005). Estimation of heritability for fluctuating asymmetry in chickens by restricted maximum likelihood: Effects of age and sex. *Poultry Science, 84*(11), 1689–1697. https://doi.org/10.1093/ps/84.11.1689

Cuervo, J. J., & Møller, A. P. (1999). Phenotypic variation and fluctuating asymmetry in sexually dimorphic feather ornaments in relation to sex and mating system. *Biological Journal of the Linnean Society, 68*(4), 505–529.

https://doi.org/10.1111/j.1095-8312.1999.tb01186.x

De Jong, I. C., & Guémené, D. (2011). Major welfare issues in broiler breeders. *World's Poultry Science Journal, 67*(1), 73–82. https://doi.org/10.1017/S0043933911000067

Düzgüneş, O., Kesici, T., Kavuncu, O., & Gürbüz, F. (1987). *Research and experimental methods (Statistical methods II)* (Publication No. 1021) [in Turkish]. Ankara University, Faculty of Agriculture.

Efe, E., Bek, Y., & Şahin, M. (2000). *Statistical methods II with solutions in SPSS* (Publication No. 10) [in Turkish]. Kahramanmaraş Sütçü İmam University, Computer Research and Application Center.

Eleroğlu, H., Yıldırım, A., Duman, M., & Şekeroğlu, A. (2015). The welfare of slow growing broiler genotypes reared in organic system. *Emirates Journal of Food and Agriculture, 27*(5), 454–459. https://doi.org/10.9755/ejfa.2015.04.075

Eleroğlu, H., & Yıldırım, A. (2019). Organik sistemde kuru kekik (*Origanum vulgare* L.) yaprağı ilave edilmiş karma yemle beslenen beç tavuklarının (*Numida meleagris*) refah parametreleri. *Journal of Poultry Research, 16*(2), 74–79. https://doi.org/10.34233/jpr.650630

Eleroğlu, H., Yeter, B., & Akyar, Ö. (2020). The effect of different production systems on some morphological characteristics and asymmetry in Atak-S cocks. *Turkish Journal of Agriculture - Food Science and Technology, 8*(12), 2770–2779. https://doi.org/10.24925/turjaf.v8i12.2770-2779.4168

Felver-Gant, J. N., Mack, L. A., Dennis, R. L., Eicher, S. D., & Cheng, H. W. (2012). Genetic variations alter physiological responses following heat stress in two strains of laying hens. *Poultry Science, 91*(7), 1542–1551. https://doi.org/10.3382/ps.2011-01988

Forkman, B., Boissy, A., Meunier-Salaün, M. C., Canali, E., & Jones, R. B. (2007). A critical review of fear tests used on cattle, pigs, sheep, poultry and horses. *Physiology & Behavior, 92*(3), 340–374. https://doi.org/10.1016/j.physbeh.2007.03.016

Ghareeb, K. (2010). Presence of males within laying hens affects tonic immobility response and sociality. *International Journal of Poultry Science, 9*(12), 1087–1091. https://doi.org/10.3923/ijps.2010.1087.1091

Gomendio, M., Cassinello, J., & Roldan, E. R. S. (2000). A comparative study of ejaculate traits in three endangered ungulates with different levels of inbreeding: Fluctuating asymmetry as an indicator of reproductive and genetic stress. *Proceedings of the Royal Society B: Biological Sciences, 267*(1446), 875–882. https://doi.org/10.1098/rspb.2000.1084

Helle, S., Suorsa, P., Huhta, E., & Hakkarainen, H. (2010). Fluctuating feather asymmetry in relation to corticosterone levels is sex-dependent in Eurasian treecreeper (*Certhia familiaris*) nestlings. *Biology Letters, 6*(4), 521–524. https://doi.org/10.1098/rsbl.2009.1068

Karaarslan, S., & Nazlıgül, A. (2016). Etlik piliçlerde refah kriteri olarak bacak sağlığı, korku ve stres parametreleri üzerine aydınlatma, yerleşim sıklığı ve tünek kullanımının etkileri I: Korku ve stres parametreleri. *Animal Health Production and Hygiene, 5*(1), 426–431.

Klingenberg, C. P. (2003). A developmental perspective on developmental instability: Theory, models and mechanisms. In M. Polak (Ed.), *Developmental instability: Causes and consequences* (pp. 14–34). Oxford University Press.

Knief, U., Schielzeth, H., Kempenaers, B., Ellegren, H., & Forstmeier, W. (2012). QTL and quantitative genetic analysis of beak morphology reveals patterns of standing genetic variation in an estrildid finch. *Molecular Ecology, 21*(15), 3704–3717. https://doi.org/10.1111/j.1365-294X.2012.05661.x

Knierim, U., Van Dongen, S., Forkman, B., Tuyttens, F. A. M., Špinka, M., Campo, J. L., & Weissengruber, G. E. (2007). Fluctuating asymmetry as an animal welfare indicator: A review of methodology and validity. *Physiology & Behavior, 92*(3), 398–421. https://doi.org/10.1016/j.physbeh.2007.02.014

Lengyel, K., Rudra, M., Berghof, T. V. L., Leitão, A., Frankl-Vilches, C., Dittrich, F., Duda, D., Klinger, R., Schleibinger, S., Sid, H., Trost, L., Vikkula, H., Schusser, B., & Gahr, M. (2024). Unveiling the critical role of androgen receptor signaling in avian sexual development. *Nature Communications, 15*(1), Article 8970. https://doi.org/10.1038/s41467-024-52989-w

Lens, L., Van Dongen, S., Kark, S., & Matthysen, E. (2002). Fluctuating asymmetry as an indicator of fitness: Can we bridge the gap between studies? *Biological Reviews, 77*(1), 27–38. https://doi.org/10.1017/S1464793101005796

Liker, A., Freckleton, R. P., & Székely, T. (2013). The evolution of sex roles in birds is related to adult sex ratio. *Nature Communications, 4*, 1587. https://doi.org/10.1038/ncomms2600

Marx, B. P., Forsyth, J. P., Gallup, G. G., & Fusé, T. (2008). Tonic immobility as an evolved predator defense: Implications for sexual assault survivors. *Clinical Psychology: Science and Practice, 15*(1), 74–90. https://doi.org/10.1111/j.1468-2850.2008.00112.x

McGary, S., Estevez, I., & Bakst, M. R. (2003). Potential relationships between physical traits and male broiler breeder fertility. *Poultry Science, 82*(2), 328–337. https://doi.org/10.1093/ps/82.2.328

Mendes, M. (2008). Asymmetry measures and allometric growth parameter estimates to investigate the effect of early feed restriction on deviation from bilateral symmetry in broiler chickens. *Archiv für Tierzucht, 51*(6), 611–619. https://doi.org/10.5194/aab-51-611-2008

Møller, A. P. (1992). Female swallow preference for symmetrical male sexual ornaments. *Nature, 357*(6375), 238–240. https://doi.org/10.1038/357238a0

Møller, A. P., & Pomiankowski, A. (1993). Fluctuating asymmetry and sexual selection. *Genetica, 89*(1–3), 267–279. https://doi.org/10.1007/BF02424520

Møller, A. P., Sanotra, G. S., & Vestergaard, K. S. (1995). Developmental stability in relation to population density and breed of chickens (*Gallus gallus*). *Poultry Science, 74*(11), 1761–1771. https://doi.org/10.3382/ps.0741761

Møller, A. P., & Swaddle, J. P. (1997). *Asymmetry, developmental stability, and evolution*. Oxford University Press.

Møller, A. P., Sanotra, G. S., & Vestergaard, K. S. (1999). Developmental instability and light regime in chickens (*Gallus gallus*). *Applied Anim. Behaviour Science, 62*(1), 57–71. https://doi.org/10.1016/S0168-1591(98)00210-7

Møller, A. P., & Manning, J. T. (2003). Growth and developmental instability. *The Veterinary Journal, 166*(1), 19–27. https://doi.org/10.1016/S1090-0233(02)00262-9

Nääs, I. A., Baracho, M. S., Salgado, D. D., Sonoda, L., Carvalho, V. C., Moura, D. J., & Paz, I. C. L. A. (2009). Broilers' toes asymmetry and walking ability assessment. *Engenharia Agrícola, 29*(4), 538–546. https://doi.org/10.1590/S0100-69162009000400004

Nelson, J. R., Settar, P., Berger, E., Wolc, A., O'Sullivan, N., & Archer, G. S. (2020). Brown and white egg-layer strain differences in fearfulness and stress measures. *Applied Animal Behaviour Science, 231*, Article 105087. https://doi.org/10.1016/j.applanim.2020.105087

Palmer, A. R., & Strobeck, C. (1986). Fluctuating asymmetry: Measurement, analysis, patterns. *Annual Review of Ecology and Systematics, 17*, 391–421. https://doi.org/10.1146/annurev.es.17.110186.002135

Parsons, P. A. (1992). Fluctuating asymmetry: A biological monitor of environmental and genomic stress. *Heredity, 68*(4), 361–364. https://doi.org/10.1038/hdy.1992.52

Polak, M., & Starmer, W. T. (2005). Environmental origins of sexually selected variation and a critique of the fluctuating asymmetry–sexual selection hypothesis. *Evolution, 59*(3), 577–585.

Prieto, M. T., & Campo, J. L. (2010). Effect of heat and several additives related to stress levels on fluctuating asymmetry, heterophil:lymphocyte ratio, and tonic immobility duration in White Leghorn chicks. *Poultry Science, 89*(10), 2071–2077. https://doi.org/10.3382/ps.2010-00716

Qi, K., Amevor, F. K., Liu, Z., He, J., Xu, D., Zhai, C., Wang, Y., Wu, L., Wang, Y., Shu, G., & Zhao, X. (2025). Relationship between comb development, immune regulation, growth hormone, testosterone, and growth traits in Tianfu broilers. *Poultry Science, 104*(8), Article 105367. https://doi.org/10.1016/j.psj.2025.105367

Rentsch, A. K., Ellis, J. L., & Widowski, T. M. (2023). Fearfulness in commercial laying hens: A meta-analysis comparing brown and white egg layers. *Poultry Science, 102*(6), Article 102664. https://doi.org/10.1016/j.psj.2023.102664

Satterlee, D. G., Cadd, G. G., & Jones, R. B. (2000). Developmental instability in Japanese quail genetically selected for contrasting adrenocortical responsiveness. *Poultry Science, 79*(12), 1710–1714. https://doi.org/10.1093/ps/79.12.1710

Shahin, K. A., & El Azeem, F. A. (2005). Effect of breed, sex and diet and their interactions on carcass composition and tissue weight distribution of broiler chickens. *Archiv für Tierzucht, 48*(6), 612–626. https://doi.org/10.5194/aab-48-612-2005

Shahin, K. A., & El Azeem, F. A. (2006). Effect of breed, sex and diet and their interactions on fat deposition and partitioning among depots of broiler chickens. *Archiv für Tierzucht, 49*(2), 181–193. https://doi.org/10.5194/aab-49-181-2006

Struthers, S., Andersson, B., Schmutz, M., Matika, O., McCormack, H. A., Wilson, P. W., Dunn, I. C., Sandilands, V., & Schoenebeck, J. J. (2023). An analysis of the maxillary beak shape variation between two pure layer lines and its relationship to the underlying premaxillary bone, feather cover, and mortality. *Poultry Science, 102*(8), Article 102854. https://doi.org/10.1016/j.psj.2023.102854

Tiemann, I., Becker, S., Fournier, J., Damiran, D., Büscher, W., & Hillemacher, S. (2023). Differences among domestic chicken breeds in tonic immobility responses as a measure of fearfulness. *PeerJ, 11*, Article e14703. https://doi.org/10.7717/peerj.14703

Van Nuffel, A., Tuyttens, F. A. M., Van Dongen, S., Talloen, W., Van Poucke, E., Sonck, B., & Lens, L. (2007). Fluctuating asymmetry in broiler chickens: A decision protocol for trait selection in seven measuring methods. *Poultry Science, 86*(12), 2555–2568. https://doi.org/10.3382/ps.2007-00173

Van Poucke, E., Van Nuffel, A., Van Dongen, S., Sonck, B., Lens, L., & Tuyttens, F. A. M. (2007). Experimental stress does not increase fluctuating asymmetry of broiler chickens at slaughter age. *Poultry Science, 86*(10), 2110–2116. https://doi.org/10.1093/ps/86.10.2110

Wang, Y., Jin, G., Ma, M., & Xiang, X. (2019). Sex differences in serum steroid hormone levels during embryonic development in hen eggs. *Poultry Science, 98*(11), 6053–6062. https://doi.org/10.3382/ps/pez270

Watson, P. J., & Thornhill, R. (1994). Fluctuating asymmetry and sexual selection. *Trends in Ecology & Evolution, 9*(1), 21–25. https://doi.org/10.1016/0169-5347(94)90227-5

Yalçın, S., & Siegel, P. B. (2003). Exposure to cold or heat during incubation on developmental stability of broiler embryos. *Poultry Science, 82*(9), 1388–1392. https://doi.org/10.1093/ps/82.9.1388

Yalçın, S., Özkan, S., Türkmut, L., & Siegel, P. B. (2001). Responses to heat stress in commercial and local broiler stocks: II. Developmental stability of bilateral traits. *British Poultry Science, 42*(2), 153–160. https://doi.org/10.1080/00071660120048401

Yang, A. (1998). *Bilateral asymmetry in chickens of different genetic backgrounds* (Publication No. 304462777) [Doctoral dissertation, Virginia Polytechnic Institute and State University]. ProQuest Dissertations & Theses Global.

Yang, A., Dunnington, E. A., & Siegel, P. B. (1997). Developmental stability in stocks of White Leghorn chickens. *Poultry Science, 76*(12), 1632–1636. https://doi.org/10.1093/ps/76.12.1632

Yehia, M., Askri, A., Achour, A., Prus, J. A., Ouellet, V., & Alnahhas, N. (2025). Rectal temperature and heat transfer dynamics in the eye, face, and breast of broiler chickens exposed to moderate heat stress. *Poultry Science, 104*(2), Article 104748. https://doi.org/10.1016/j.psj.2024.104748

Zakharov, V. M., Shadrina, E. G., & Trofimov, I. E. (2020). Fluctuating asymmetry, developmental noise and developmental stability: Future prospects for the population developmental biology approach. *Symmetry, 12*(8), Article 1376. https://doi.org/10.3390/sym12081376