

## ASSOCIATION OF GROWTH RATE AND GENE EXPRESSION OF GROWTH-RELATED GENES IN TWO COMMERCIAL BROILER STRAINS

**Habiba, H. Rezk and M.M. Hamed**

*Poultry Production Department, Faculty of Agriculture, Ain Shams University, P.O. Box 68 Hadayek Shoubra, 11241 Cairo, Egypt*

Email : [habiba\\_hassan@agr.asu.edu.eg](mailto:habiba_hassan@agr.asu.edu.eg); [mohamed.hamed93420@gmail.com](mailto:mohamed.hamed93420@gmail.com)

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### SUMMARY

To investigate whether a variation in the body weight gain is associated with gene expression of growth-related genes, (insulin like growth factor (IGF-I) and growth hormone (GH) profile in broilers, to genetic evaluation of body weight gain of two broiler commercial Ross and Arbo acres broiler strain by comparing gene expression of Growth Hormones and Insulin-like Growth Factor were conducted with 1000 newly hatched Ross and Arbo acres in a Poultry Breeding Farm, Poultry Production Department, Faculty of Agriculture, Ain Shams University, Shalakan under standard management. Body weight and body weight gain were recorded for all broiler birds that were reared. At 28 days of age, six samples from each replication were chosen at random from each strain for the gene expression analysis. Results showed that both body weight and total weight gain were highly significant ( $P < 0.01$ ) in Ross broilers compared to Arbo Acres broiler strains. Additionally, an increased gene expression of insulin-like growth factor (IGF-I) was observed in heavy and higher-weight gain strains (Ross strains) ( $P < 0.01$ ), relative to lighter and slower-weight gain strains (Arbo Acres strains). The heavy weight is linked to the gene expression (IGF-I) in broiler chickens. These parameters could be employed to select breeding programs for enhanced growth based on the gene expression level of the genes responsible for weight body.

**Keywords:** *Body weight gain; Ross broiler; Arbo Acres broiler; the GH and IGF-I genes; gene expression.*

### INTRODUCTION

The Ross broiler, particularly the Ross 308 strain, has become a focal point in poultry production due to its rapid growth, efficiency, and adaptability in commercial settings. The Ross 308 is the world's most popular broiler, providing integrated operations with the perfect balance of breeder, broiler, and processing performance. Through a progressive program of breeding and selection, Arbor Acres products have been developed to serve the multiple demands of markets around the world. The Arbor Acres product line is steadily improved to ensure all products consistently add value to customer operations through established breed selection processes that use both traditional scientific techniques and the latest in technology. Research has highlighted significant genetic variations among broiler strains, particularly when comparing Ross 308 to other broiler strains. Studies indicate that Ross 308 exhibits distinctive growth performance metrics, including body weight gain and feed conversion ratios, which are critical for assessing economic efficiency in poultry farming. Genetic selection plays a vital role in these outcomes, suggesting that understanding the genetic basis for performance can enhance production efficiency (Biasato *et al.*, 2018). Furthermore, the evolution of Ross broilers, such as the Ross 308 strain, reflects significant human intervention in breeding practices, resulting in increased body size and altered skeletal morphology. These changes underscore the impact of anthropogenic factors on genetic traits and raise questions about the sustainability of such practices in the long term (Bennett *et al.*, 2018).

In addition to growth performance, the quality of meat produced by Arbor Acres broilers has also garnered attention. Research comparing Arbor Acres broilers with other strains indicated that Arbor Acres exhibited higher dressing percentages and superior meat quality traits (Wang *et al.*, 2020). This is particularly beneficial for producers focusing on meat quality, as these traits can serve as significant selling points in competitive markets. Moreover, nutritional strategies that enhance muscle characteristics

and overall meat quality have been investigated. Arbor Acres broilers have been shown to possess genetic predispositions that contribute to their superior meat quality, reinforcing the need for strategic dietary manipulation to maximize these traits (Qiu *et al.*, 2021).

When growth hormones (GHs) are released from the pituitary gland, they bind to specific receptors on target cells, triggering a cascade of signaling pathways that ultimately lead to the expression of genes related to growth and metabolism. These genes encode proteins that mediate the effects of (GHs), such as stimulating cell division, protein synthesis, and tissue growth.

Similarly, insulin like growth factors (IGFs) are produced in various tissues and circulate in the bloodstream, where they can also bind to receptors on target cells. Upon binding, IGFs can activate gene expression programs that promote cell growth, differentiation, and survival. The interaction between GHs and IGFs is complex, as they often work together to regulate growth and metabolism.

The GH is involved in numerous physiological processes, such as body composition, development, aging, reproduction, and egg production (Su *et al.*, 2014), and it is essential for both growth and metabolic rates (Anh *et al.*, 2015). A hormone with several metabolic and anabolic functions, IGF-I shares structural similarities with insulin (McMurtry *et al.*, 1997). For chickens to grow normally and generate bone and fat tissue, this hormone is essential (Boschiero *et al.*, 2013). However, the liver's intermediary pathway between the pituitary gland and one of its target tissues, muscle, is where GH functions (Soendergaard *et al.*, 2017). GH in the liver is probably responsible for controlling the synthesis of many proteins by controlling the expression of their genes (Rastegar *et al.*, 2000). The GH controls the paracrine synthesis of IGF-I in numerous different tissues and promotes IGF-I synthesis in the liver (Laron, 2001). The main objective of the research is to investigate whether a variation in body weight gain is associated with gene expressions of growth-related genes (insulin-like growth factor (IGF-I) and growth hormone (GH) profile in broilers by comparing two broiler strains (Ross and Arbor Acres).

## **MATERIALS AND METHODS**

### ***Ethics statement:***

You have to determine that the experimental protocol was reviewed and approved by the Animal Ethics Committee of (Faculty of Agriculture, Ain Shams University) under approval number (Approval No.).

### ***Measurement of growth:***

This experiment was carried out at Poultry Breeding Farm, Poultry Production Department, Faculty of Agriculture, Ain Shams University, Shalakan at the beginning month of October 2024. All chicks were brooded in brooding Letter (land) from hatching to the end of the experiment at 28 days of age. All birds were reared under similar environmental, managerial, and hygienic conditions. Body weight (BW) of 500 chicks was individually recorded at hatching and at one, two, three, and four weeks of age (BW0, BW1, BW2, BW3, and BW4). The body weight gain (BWG) was calculated at one-week intervals: 0 to 1 weeks of age (BWG 0–1), 1 to 2 weeks of age (BWG 1–2), 2 to 3 weeks of age (BWG 2–3), 3 to 4 weeks of age (BWG 3–4). To extract genomic DNA and the IGF-I and GH genes, blood samples from two broiler strains (Ross and Arbor Acres), peripheral blood samples were obtained by vein puncture at 28 days of age. Six samples from each replication were chosen at random for the gene expression analysis.

### ***Material used for cDNA synthesis:***

Maxima First Strand cDNA Synthesis Kit for RT-qPCR #K1641 from Thermo Scientific One practical method designed for cDNA synthesis in two-step real-time quantitative RT-PCR (RT-qPCR) applications is the Maxima® First Strand cDNA Synthesis Kit. The complex enzyme Maxima Reverse Transcriptase, which is produced by the in vitro synthesis of M-MuLV RT, is used in the kit. Compared to wild-type M-MuLV RT, the enzyme is substantially more thermostable and robust has a greater rate of cDNA synthesis. Reproducible cDNA synthesis from a variety of starting total RNA levels (1 pg–5 µg) at high temperatures (50–65°C) is possible using the Maxima First Strand cDNA Synthesis Kit. The synthesis reaction can be completed in 15 to 30 minutes.

### ***Material used for RT-PCR:***

Following reverse transcription procedures (RT-qPCR) with varying initial RNA transcript dosages (5 ng to 0.5 fg), amplification was carried out using the Maxima SYBR Green/ROX qPCR Master Mix. The

slope varies from -3.09 to -3.58, the correlation coefficient is more than 0.99, and the reaction efficiency falls between 90 and 110 percent. The genotypes of GH and IGF-I were determined by the polymerase chain reaction (PCR) utilizing restriction fragment length polymorphism (RFLP). Table 1 lists the forward and reverse primers, enzymes, and annealing temperatures for the GH and IGF-I gene PCR-RFLP.

**Table (1): Primer sequences and their applications.**

Gene	Primer (forward/reverse)	AT (°C)	PCRproduct (bp)	Enzyme
<i>cGH</i>	5'-TCCCAGGCTGCGTTTTGTTACTC-3' 5'-ACGGGGGTGAGCCAGGACTG-3'	65	429	<i>EcoRV</i>
<i>IGF-I</i>	5'-TCAAGAGAAGCCCTTCAAGC-3' 5'-CATTGCGCAGGCTCTATCTG-3'	60	813	<i>HinfI</i>

PCR, polymerase chain reaction; GH, chicken growth hormone gene; IGF-I, insulin-like growth factor gene. Nguyen et al., 2015

#### Statistical analysis:

Data were subjected to one way analysis and their interaction using the General Linear Models (GLM) procedure of SAS User's Guide, Ver.8.2, 2001. Duncan's multiple range tests were used to separate means when separation was relevant. To estimate mRNA by using real time PCR. Then, a spread sheet program (Microsoft Excel) was used to arrange the data for each breed regarding each locus. Data were analyzed employing the Arlequin 3.5 software package after data conversion using CONVERT program.

$$Y_{ik} = \mu + S_i + e_{ik}$$

Where:

$\mu$ = Overall mean,

$S_i$ =Strain effect,

$e_{ik}$ = Experimental error.

## RESULTS AND DISCUSSION

#### Body weight and body weight gain:

Initial Weight: The initial weight between the two strains was fairly consistent (Table 2). Chick hatch was 44 g and 44.47 for Ross and Arbor acres, respectively with mean initial weight of 44.24g as presented in Table 1.

**Table 2: Effects of body weight (g) and body weight gain of broiler chicks.**

Items	initial body weight	BW one week	2 wee ks	3we ek	4 we eks	WG (0-1) 0-1	WG (1-2) 1-2	WG (2-3) 2-3	WG (3-4) 3-4	Total weight gain WG0-4
<b>Arbo Acres</b>	44.47	157.85	422. 88	100 4.14	171 9.2	113.3 8	265.0 3	581.2 6	715.0 6	1674.73
<b>Ross</b>	44	246.5	688	1370	1969	202.5	441.5	682	599	1925
<b>SEM</b>	0.41	2.04	6.43	18.6	19.9	3.44	7.80	20.01	22.30	27.80
<b>Sig.</b>	NS	*	*	*	*	*	*	*	*	*

a,b Means within column with different are significantly different, SEM=standard error of the means; NS = not significant. \*Significant at  $P \leq 0.01$ . BW= Body weight, WG= weight gain

The distribution of initial weight at hatching was fairly uniform among two strains (Arbor Acres and Ross) with initial weight means not significantly different. This lack of difference in the initial weight of

the two strains may be due to the fact that the genetic base of most commercial strains is the same, and therefore the performance traits seldom differ among commercial broilers (Moro *et al.*, 2005). Ross had significantly ( $P<0.01$ ) heavier final body weight at four weeks of 1969 g vs. 1819.2 g for and Arbor acres, as shown in Table 2. The effect of strain on the final weight at 4 weeks was highly significant ( $P<0.01$ ) for Ross broiler. Also, Total Weight Gain: Ross Strain had the recorded value of 1925 g total weight gain in the 4 weeks in (Table 2), which was significantly ( $P<0.01$ ) higher than what was recorded for the other strain. The Ross strain showed a higher weight compared to the Arbo Acres. This is evident from previous research that indicated that the Ross is one of the heaviest commercial broilers (Khalid *et al.*, 2021).

#### Gene expression of GH gene and IGF-I gene:

The present results revealed that GH and IGF-I influence growth in figures (1) and (2). Where it appeared that Arbor acres had significantly ( $P<0.01$ ) higher GH than Ross broiler. But The Ross strain showed high significance ( $P<0.01$ ) in the IGF-I gene compared to the Arbo Acres broiler. The reason for this is that studies indicate that the Ross strain is heavier and slower in growth due to structural formation and structural and metabolic functions, so it has a high significance for the IGF-I gene compared to the Arbo Acres broiler. And Nam *et al.* (1997) showed that the levels of serum GH and IGFBP-1 were suppressed in all the obese subjects ( $P<0.05$ ). Xiao *et al.* (2017) showed that growth rate has linear positive correlations with gene expression IGF-1 of broilers.

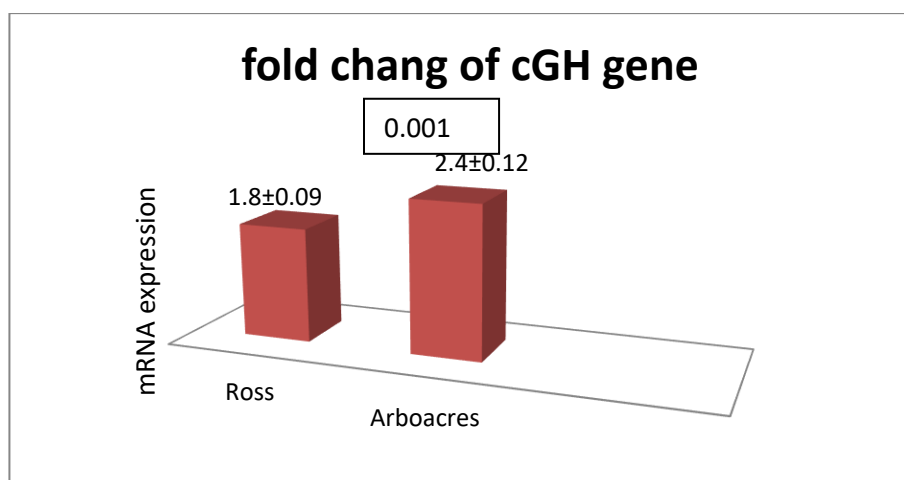


Fig 1 . The mRNA abundance of the GH gene of Ross broiler and Arbo Acres broiler at 28 days of age. significantly \*  $p \leq 0.01$ .

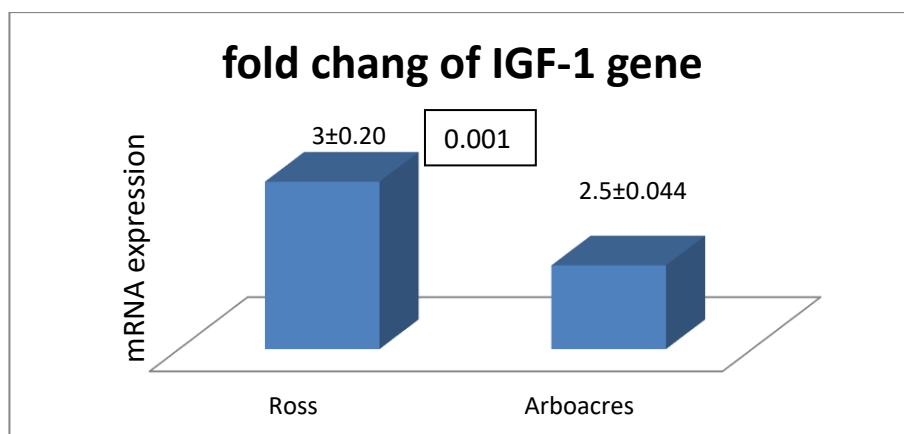


Fig 2 . The mRNA abundance of the IGF-I gene of Ross broiler and Arbo Acres broiler at 28 days of age. significantly  $p \leq 0.01$ .

Several genes are involved in growth traits; however, (GH) and IGF-I are the major hormones required to support normal growth (Kita et al., 2005). GH is involved in a wide variety of physiological functions, such as growth, body composition, egg production, aging, and reproduction (Su *et al.*, 2014), and it plays a critical role in both growth and metabolism rates (Anh et al., 2015). The IGF-I is a hormone that is structurally related to insulin and has multifunctional metabolic and anabolic properties. It is a crucial hormone for normal growth and for bone and fat tissue development in chickens (Boschiero et al., 2013).

## CONCLUSION

This study showed the superiority of the Ross broiler over Arbo Acres regarding body weight and body weight gain. Additionally, an increase of gene expression of insulin-like growth factor (IGF-I) was observed in Ross broilers. The IGF-I genes may play an important role in body weight gain. Hence, the gene can be used as a promising marker for selecting broiler strains with a higher body weight gain.

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## ارتباط معدل النمو والتعبير الجيني للجينات المرتبطة بالنمو في سلالتين تجاريتين من دجاج التسمين

حبيبة حسن رزق و محمد مصطفى حامد

قسم إنتاج الدواجن، كلية الزراعة، جامعة عين شمس، القاهرة، مصر

غالبًا ما تختلف الزيادة الوزنية للجسم ومعدلات النمو بين سلالات الدجاج اللحم من نفس السلالة أو سلالات متباينة تحت نفس الظروف البيئية. للتحقق مما إذا كان التباين في زيادة وزن الجسم مرتبطًا باختلاف التعبير الجيني للجينات المرتبطة بالنمو (عامل النمو الشبيه بالأنسولين IGF-I) وهرمون النمو (GH) في دجاج التسمين، لتقييم وراثي لزيادة وزن الجسم لسلالتين تجاريتين من دجاج التسمين من نوع روس وأربواكرز عن طريق مقارنة التعبير الجيني لهرمونات النمو وعامل النمو الشبيه بالأنسولين تم إجراء ذلك على 1000 دجاجة حديثة الفقس من نوع روس وأربواكرز في مزرعة تربية الدواجن بقسم إنتاج الدواجن بكلية الزراعة بجامعة عين شمس تحت نفس الظروف البيئية. تم تسجيل وزن الجسم وزيادة وزن الجسم لجميع طيور دجاج التسمين التي تم تربيتها. في عمر 28 يومًا، أخذت ست عينات من كل تكرار عشوائيًا لكل سلالة لتحليل التعبير الجيني. بناءً على نتائج هذه الدراسة، تم تحديد تأثير سلالة دجاج التسمين على وزن الجسم وزيادة الوزن الكلي. أظهرت النتائج أن كل من وزن الجسم وزيادة الوزن كانت ذات دلالة إحصائية عالية ( $P < 0.01$ ) في دجاج التسمين من نوع روس مقارنة بسلالات دجاج التسمين من سلالة أربواكرز. بالإضافة إلى ذلك، لوحظ ارتفاع في التعبير الجيني لعامل النمو الشبيه بالأنسولين (IGF-I) في السلالات ذات الوزن الثقيل وزيادة الوزن العالية (سلالة روس) (قيمة الاحتمال  $> 0.01$ )، مقارنة بالسلالات ذات الوزن الخفيف وبطء زيادة الوزن (سلالة أربواكرز). وتشير هذه النتائج مجتمعة إلى أن الوزن الثقيل مرتبط بالتعبير الجيني (IGF-I) في دجاج التسمين. ويمكن استخدام هذه المعايير لاختيار برامج تربية مُحسنة للنمو.

**الكلمات المفتاحية:** زيادة وزن الجسم، سلالة الروس، سلالة أربواكرز، وجينات GH و IGF-I، التعبير الجيني.