

RESEARCH ARTICLE

Productive Traits, Biochemical Parameters, Meat Quality, and the Gene Expression in Different Duck Breeds

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Abstract

This research was performed to evaluate “productive traits, biochemical parameters, meat quality and the expression of related genes in different duck breeds”. This experiment was conducted on 80 ducks (20 Pekin, 20 Star 53, 20 Muscovy, and 20 Mulard). Each breed was reared under the same ecological, managerial, and sanitary conditions from one day old until the end of the experiment (12th week of the age). The growth performance, biochemical characteristics, physicochemical parameters of meat, and gene expression of Growth hormone (GH), Insulin like growth factor 1 (IGF1), and calpain genes were analyzed and estimated. The findings showed that Mulard ducks had significantly greater ($p < 0.05$) body weight (4234 g) followed by Muscovy (4029 g), Star 53 (3659 g), and Pekin (2961 g) although the feed conversion ratio (FCR) of Pekin ducks was higher ($p < 0.05$) than others. Mulard had the longest shank, body length, keel length, and body circumference (8.28, 78.50, 20.21, and 38.60 cm, respectively) at the end of the experiment. Pekin ducks recorded the highest values of total cholesterol (TC), superoxide dismutase (SOD), and glutathione peroxidase (GPX). While, Mulard recorded the highest values of Glucose, high-density lipoprotein (HDL), low-density lipoprotein (LDL), and malondialdehyde (MDA). The lowest shear force value for breast and thigh muscles was observed in Muscovy ducks. GH, IGF-1 expression was higher in Mulard than other breeds. While, Calpain gene expression was higher ($p < 0.05$) in Muscovy than in Mulard, Star 53, and Pekin. From the obtained results, it could be concluded that the observed highly productive traits in Mulard ducks might be attributed to the difference in gene expression related to growth. However, the good meat quality in Muscovy ducks was owing to the high expression of the Calpain gene.

Keywords:

duck breeds; biochemical blood parameters; body measurements; Meat quality; Gene expression.

Introduction

The progress of duck production had expanded over the past 20 years, where about 1.15 billion ducks were kept around the world in 2017 [1]. Egypt has recently focused on increasing meat production, particularly ducks, which are believed to be the easiest domestic bird to raise

[2,3]. Pekin, Aylesbury, Rouen, and Muscovy ducks are the most popular breeds used for meat production [4,5]. Pekin ducks have white plumage and are distinguished by their tasty meat [6]. Due to their quick growth and disease resistance, white Pekin ducks are utilized to improve strains [7]. Muscovy ducks are very popular because of their various

environmental adaptations, distinctive taste, high breast meat, and low-calorie level [8]. The Mulard (a Muscovy and Pekin duck hybrid) is raised for its meat and fattened liver[9].

The weighing of animals is the simplest method for evaluating an animal's mass. But in certain circumstances, a scale could not be accessible. A substitute is to measure a body portion and correlate the measurement to body weight (BW). Shank length is the body portion that is usually evaluated in poultry to correlate with BW. There has been research showing a direct correlation between BW and shank length [10]. Body size and body measurement have been used as parameters for selection by local sellers and research [11]. Comparing phenotypes based on morphological characteristics is a useful way to illustrate the genetic diversity among populations. The selection of appropriate breeding objectives and plans depends on the genetic diversity of the duck populations[12].

The term "biochemical" describes the study of chemical processes that occur within and correlate with living organisms. Hematological and biochemical tests have not frequently been effective for the diagnosis of avian species, but these tests may be a suitable diagnosis mechanism for assessing health status, observing sick ducks' responses to treatment plans, and providing a prognosis for some duck diseases [13].

One of the most significant economic characteristics in the domestic animal industry is body growth which is controlled by many factors including endocrine factors. Consequently, many endocrinological investigations have been carried out to enhance body growth in numerous species[14,15]. More

specifically, growth factors, such as *IGF-I* have been shown to stimulate body growth in mammals [16]. Many of the anabolic activities of *GH* are regulated by *IGF-1* [17]. The growth of body tissues is stimulated by *IGF-I* levels, which are influenced by *GH* production [18]. *GH* controls the growth of various tissues after birth, including skeletal muscles [19].

The maintenance and improvement of human health and well-being depend on food quality. Previously, it had been stated that duck meat is abundant in nutrients and low in cholesterol. Researchers have become more interested in duck meat quality and related food quality concerns[20,21]. Some of the parameters that influence meat quality are tenderness, water-holding capacity, color, nutritional content, and safety. The value of these traits' changes depending on the product type and the consumer characteristic [22].

One of the most essential factors influencing overall meat quality is tenderness. Meat tenderness varies according to breed, nutrition, husbandry, and slaughter methods. The calpain system (Ca^{2+} + activated neutral proteinases) is probably one of the major components related to meat tenderness, and its influences on postmortem tenderization have been well reported. Ca^{2+} can activate calpain after slaughtering the animal and causes the breakdown of postmortem myofibrillar protein as well as tenderness improvement [23,24]. There were few studies on the *Calpain* gene in ducks, so the first aim of this research is to evaluate the level of the *Calpain* gene in breast muscle among different duck breeds and its association with meat tenderness. The second aim is to assess the level of *GH* and *IGF1* expression and their

relation with productive traits.

Materials and Methods

Birds and Management

This research was approved by the Animal Care and Welfare Committee of Zagazig University, Egypt (ZU-IACUC/2/F/149/2022). A total of 80 ducks (20 Pekin, 20 Star 53, 20 Muscovy, and 20 Mulard ducks) were obtained from EL-Wafaa Farm Company in Giza and reared under the same ecological, managerial, and sanitary conditions from one day old until the end of the experiment (12th week of age). They were kept in pens with similar floor area (5 ducks/ m²) covered with 5 cm thickness of wood shaving. The environmental temperature was provided at 34°C for the first three days, then declined by about 2°C per week till it reached 25°C, and the light was continuous. The feed was supplied ad libitum with the starter ration 22% (cp) was offered from 1st to 4th week of age. While, from 5th to 12th week of age, the grower/finisher ration 18% (cp) offered by NRC [25].

Measurements and observations

Productive traits

Growth performance traits included initial BW, final BW, average feed intake (AFI), and feed conversion ratio (FCR). The initial BW of all ducks was recorded on the first day of age. At 12th week of age, we evaluated the final BW of all ducks after 12h fast. AFI and FCR were estimated on replicate basis. Feed consumption for the total period divided by total body weight gain to estimate the feed conversion ratio (FCR). Body measurements were measured bi-weekly to determine shank length, keel length, breast circumference, and body

length according to Makram *et al.* [26] as the following:

Shank Length: was evaluated on live duck by determining the length of the tibiotarsus from the top of the hock joint to the foot pad with a digital caliper (cm).

Body Length: Starting from the beak to the end of the bird's foot (cm).

Keel Length: was determined for each duck individually by a digital caliper (cm).

Breast circumference: was measured under the wings at the edge of the sternum.

Serum biochemical parameters

Blood samples were taken during slaughtering at the 12th week of age from the jugular vein under aseptic circumstances. Samples were collected in test tubes without EDTA and then centrifuged at 2500 rpm for 12 min for serum collection. Serum samples were kept at -20°C until used for serum biochemical characterization analysis. Serum glucose, albumin, uric acid, total protein, triacylglycerol, total cholesterol, high-density lipoprotein and low-density lipoprotein were determined in a digital spectrophotometer (Biomate 5, Thermo Electron Corporation, Rochester, NY, USA) by using commercial kits (Nanjing Jiancheng Institute of Bioengineering, Nanjing, China). Serum malondialdehyde, glutathione peroxidase and superoxide dismutase activities were estimated using ELISA Kit of QuantiChrom™ (Hayward, CA, USA), BioAssay Systems (Hayward, CA, USA) and Cayman Chemical Company (Hayward, CA, USA); respectively.

Physico-chemical properties of meat

Breast and thigh muscles were stored at 4°C for 24h post-mortem to assess pH, color, water holding capacity, drip loss, cooking loss, shear force, and thio-barbituric acid reducing substances. Ultimate pH (pHu) was measured after 24 h of chilling and evaluated by operating an electrical pH meter (Bye model 6020, USA) that was adjusted daily with standard pH buffers of 4.0 and 7.0 at 25°C [27]. The surface color as CIE L* (lightness), a* (redness), and b* (yellowness) values were calculated using a Hunter Lab Color Meter (D25-INC4750- Hunter Associate Lab, Reston, Virginia, USA) [28].

Water holding capacity was estimated as the ratio (%) of the volume of released water to the tested intact duck meat [29]. Drip loss (%) was estimated according to the difference between the first weight of the samples measured at the facility and the second weight of them calculated after 1-day post-mortem during chilling storage at 4°C [30]. Cooking loss was determined as the proportion of weight loss to a constant weight after cooking (W2) and the initial weight (W1) [31].

$$\text{Cooking loss\%} = \frac{W1 - W2}{W1} \times 100$$

The shear force was measured with a blade (68mm wide × 72 mm long × 3mm thick) by using instron device 1195 (England). The blade speed 10 mm/min was used to perform shear force analysis and the pickup force of the calculating head was 50 Kg with the muscle fibers parallel to the direction of the blade. The

findings were given in kilograms of shear force [32]. The quantities of thio-barbituric acid reducing substances (TBARS) were represented as milligrams of malondialdehyde per kilogram of meat [33].

Determination of the relative expression of growth-related genes (Growth hormone (GH), Insulin like growth factor 1 (IGF-1)), and Calpain gene.

For the expression of *IGF-1*, *GH*, and *Calpain* genes, samples of breast muscle were taken and put in sterile tubes (RNase-free), which were then directly submerged in liquid nitrogen prior to storage at -80°C until use. RNA extraction from tissue samples was utilized using the QIAamp RNeasy Mini kit (Qiagen, Germany, GmbH). Preparation of tissue samples and extraction of RNA was prepared following kit instructions. 25- μl reaction containing 12.5 μl of the TransScript Green One-Step qRT-PCR SuperMix®, 1 μl of RT Enzyme Mix (20X), 0.5 μl of each primer (forward and reverse) of 20 pmol concentration, 0.5 μl reference dye, 5 μl of water, and 5 μl of RNA template. The reaction was performed in a step one real time PCR machine. Primers [34, 35] used were supplied from Metabion (Germany) and are listed in Table (1). Amplification curves and CT values were established by step one software. To evaluate the variation of gene expression on the RNA of the various samples, the CT of each sample was compared with that of the control group (Pekin duck) according to the "ΔΔCT" method declared by Yuan *et al.* [36] utilizing the following ratio: ($2^{-\Delta\Delta CT}$).

Table 1. Primer sequences and cycling conditions for SYBR green real-time PCR.

Target gene	Primers sequences (5'→3')	Reverse transcription	Primary denaturation	Amplification (40 cycles)			Reference
				denaturation	Annealing	Extension	
<i>B-actin</i>	F:5'-GTGGATCAGCAAGCAGGAGT-3'						[34]
	R:5'-TTTGT CACAAGGGTGTGGGT-3'						
<i>Calpain</i>	F:5'-AAGCGACATTGGCGAACT-3'						[35]
	R:5'-CCAGCCCACAAGACATCC-3'	50°C	94°C	94°C	55°C	72°C	
¹ <i>IGF-1</i>	F:5'-GCCATCTGCAGGATACTTTGC-3'	30 min.	15 min.	15 sec.	30 sec.	30 sec.	[34]
	R:5'-CTGGGAGAATGCCCATTTGGT-3'						
² <i>GH</i>	F:5'-TTT GCC AAC GCT GTGC-3'						[8]
	R:5'-CTG GGC ATC ATC CTT CC-3'						

The ¹*IGF-1* primer (insulin growth factor-1), ²*GH* primer(growth hormone).

Statistical analysis

Results were described as mean \pm SEM (standard error of mean). In order to evaluate the effect of the 4 breeds, one-way analysis of variance (ANOVA) was applied. Duncan multiple tests as post hoc test were utilized to determine the difference between means. The value of $P < 0.05$ was employed to demonstrate statistical significance. All analyses and charts were drawn with Statistical Package for Social Sciences version 24.0 (SPSS, IBM Corp., Armonk, NY) and Graph Pad Prism 8.0.2 (GraphPad Software, Inc).

Results

There was a significant difference in growth performance among different duck breeds. The Mulard breed exhibited the highest final BW (4234 g) followed by the Muscovy (4029 g), Star 53 (3659 g) and Pekin (2961 g). Furthermore, AFI was significantly reduced in Muscovy breed (14316 kg) compared to Star 53 (16786 kg). The highest feed conversion ratio was recorded for Pekin ducks followed by Star 53. While Muscovy and Mulard exposed no significant change in feed conversion ratio as shown in (Figure 1).

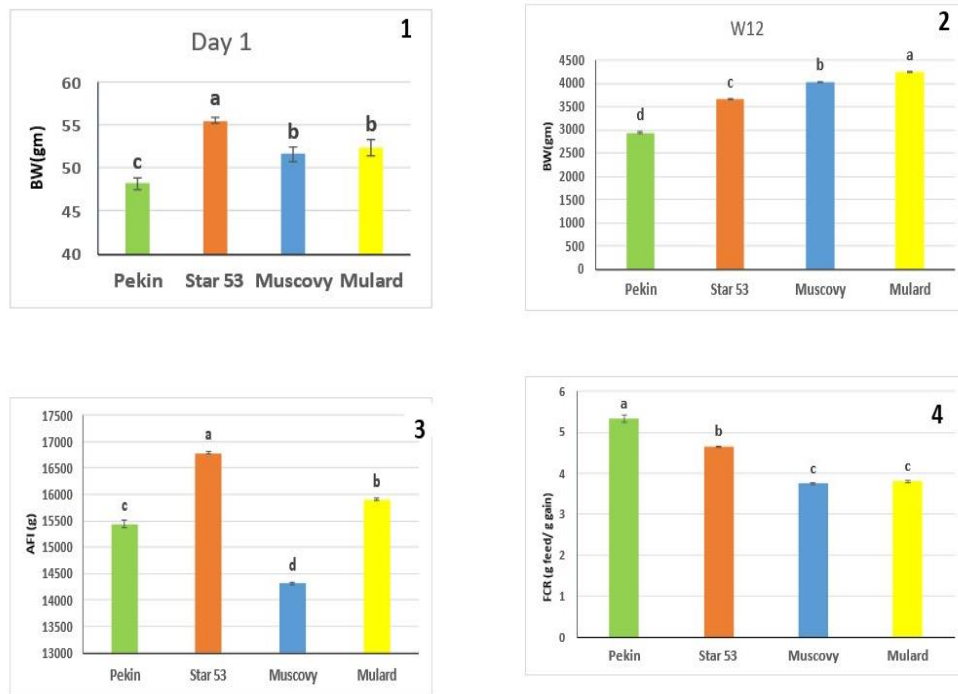


Figure 1. Growth performance of Pekin, Star 53, Muscovy, and Mulard represented as (mean \pm SEM). Groups with different letters are significantly different ($P < 0.05$). (1) Initial body weight, (2) final Body weight, (3) Average feed intake (AFI) and (4) feed conversion ratio (FCR).

The shank length, body length, keel length, and body circumference at all studied ages were affected by breed as presented in Table (2). There is no difference between Star 53, and Mulard at 2, 4, and 8 weeks of age for shank length. A similar trend was noticed at 2, 4, 6, and 8 weeks of age for body length and at 4 weeks for keel length. Star 53 had a significantly higher shank length (7.33 cm) compared to Mulard (7.05 cm), Muscovy (6.80 cm), and Pekin (6.68 cm) at 6 weeks of age. A similar trend was noticed for body length at 4 weeks of age.

Concerning body circumference, and keel length, it could be noticed that the Star 53 duck had considerably greater body measurements at 2, 6, and 8 weeks of age compared to Mulard, Muscovy, and Pekin. At 12th week of age, Mulard had longer shank, body, and keel lengths (8.28, 78.50, and 20.21 cm., respectively) and also had a longer body circumference (38.60 cm). While Pekin ducks recorded shorter body, keel lengths and had a smaller body circumference (72, 15.5, and 34.5cm., respectively).

Table 2. Body measurements (Shank length, Body length, Keel length and Body circumference) for four duck breeds under investigation

Strains	Pekin	Star 53	Muscovy	Mulard	P-value
Shank length (cm)					
2 nd week	4.60±0.11 ^b	5.20±0.14 ^a	4.73±0.03 ^b	4.92±0.04 ^{ab}	<0.05
4 th week	5.20±0.11 ^c	6.93±0.13 ^a	5.55±0.01 ^b	6.94±0.01 ^a	<0.05
6 th week	6.68±0.09 ^c	7.33±0.02 ^a	6.80±0.04 ^c	7.05±0.03 ^b	<0.05
8 th week	6.68±0.09 ^c	7.46±0.03 ^a	7.21±0.04 ^b	7.43±0.04 ^{ab}	<0.05
10 th week	7.42±0.06 ^c	7.58±0.05 ^b	7.61±0.05 ^b	7.85±0.07 ^a	<0.05
12 th week	7.58±0.08 ^b	7.61±0.08 ^b	7.81±0.07 ^b	8.28±0.04 ^a	<0.05
Body length (cm)					
2 nd week	35.01±0.07 ^c	38.60±0.29 ^a	36.20±0.09 ^b	38.10±0.07 ^a	<0.05
4 th week	53.00±0.59 ^d	64.95±1.08 ^a	55.60±0.11 ^c	61.50±0.11 ^b	<0.05
6 th week	64.95±1.08 ^b	67.45±0.40 ^a	65.30±0.36 ^{ab}	65.50±0.11 ^{ab}	<0.05
8 th week	67.45±0.40 ^c	72.00±0.23 ^a	70.35±0.24 ^b	71.60±0.22 ^a	<0.05
10 th week	68.70±0.11 ^d	74.55±0.11 ^c	75.75±0.099 ^b	76.47±0.05 ^a	<0.05
12 th week	72.00±0.23 ^d	75.75±0.09 ^c	76.50±0.12 ^b	78.50±0.12 ^a	<0.05
Keel length (cm)					
2 nd week	4.04±0.04 ^d	5.30±0.11 ^a	4.40±0.07 ^c	5.02±0.01 ^b	<0.05
4 th week	4.71±0.01 ^c	9.05±0.31 ^a	8.32±0.03 ^b	8.82±0.06 ^{ab}	<0.05
6 th week	8.43±0.28 ^d	14.5±0.19 ^a	10.8±0.05 ^c	12.5±0.11 ^b	<0.05
8 th week	9.70±0.036 ^d	16.25±0.057 ^a	14.40±0.06 ^c	15.23±0.04 ^b	<0.05
10 th week	13.93±0.18 ^d	17.6±0.11 ^c	18.08±0.03 ^b	19.05±0.03 ^a	<0.05
12 th week	15.5±0.47 ^c	18.30±0.03 ^b	19.11±0.04 ^b	20.21±0.04 ^a	<0.05
Body circumference (cm)					
2 nd week	14.13±0.15 ^d	22.25±0.09 ^a	19.15±0.04 ^c	20.16±0.04 ^b	<0.05
4 th week	22.25±0.099 ^d	32.24±0.047 ^a	27.25±0.34 ^c	29.70±0.11 ^b	<0.05
6 th week	28.30±0.03 ^d	32.80±0.09 ^a	29.07±0.11 ^c	31.02±0.09 ^b	<0.05
8 th week	32.80±0.092 ^c	33.80±0.15 ^a	32.60±0.11 ^c	33.30±0.11 ^b	<0.05
10 th week	33.80±0.15 ^d	34.50±0.11 ^c	36.50±0.11 ^b	37.60±0.11 ^a	<0.05
12 th week	34.50±0.11 ^d	36.50±0.11 ^c	37.60±0.11 ^b	38.60±0.11 ^a	<0.05

There was significant diversity in cholesterol, Triglyceride, alanine biochemical parameters between the four duck breeds. The results revealed that the highest values of Glucose, HDL, LDL, Globulin, and MDA were observed in Mulard ducks. While Pekin ducks recorded the highest value of Total

cholesterol, Triglyceride, alanine aminotransferase (ALT), SOD, and GPX. The lowest values of total protein, Globulin and GPX were recorded in Star 53. Muscovy ducks showed the highest level of uric acid, Total protein and albumin Table (3).

Table 3. The effect of Duck breeds reared under the same environmental conditions on their biochemical parameters at 12th week of age

Parameters	Pekin	Star 53	Muscovy	Mulard	P-value
Glucose(mg/dL)	142.33±0.21 ^d	152±0.86 ^c	163.5±0.43 ^b	172±0.86 ^a	< 0.001
uric acid(mg/dL)	3.20±0.04 ^d	9.82±0.31 ^c	24.5±0.34 ^a	12.17±0.17 ^b	< 0.001
Total cholesterol(mg/dL)	202.67±0.88 ^a	150.5±0.22 ^d	167.83±1.9 ^c	195.67±0.80 ^b	< 0.001
HDL (mg/dL)	50.00±0.37 ^b	50.00±0.00 ^b	38.17±0.17 ^c	62.83±0.70 ^a	< 0.001
LDL (mg/dL)	150.17±0.60 ^b	100.50±0.22 ^c	67.67±0.61 ^d	260.00±1.37 ^a	< 0.001
Triglyceride(mg/dL)	153.17±0.17 ^a	142.33±0.21 ^b	131.17±0.4 ^c	61.67±0.42 ^d	< 0.001
Total protein (g/dl)	3.77±0.02 ^b	1.13±0.11 ^c	5.43±0.17 ^a	5.7±0.04 ^a	< 0.001
Albumin(g/dl)	1.80±0.04 ^c	0.67±0.07 ^d	3.25±0.18 ^a	2.62±0.06 ^b	< 0.001
Globulin(g/dl)	1.85±0.01 ^c	0.40±0.02 ^d	2.25±0.06 ^b	3.22±0.05 ^a	< 0.001
A/G ratio	0.97±0.02 ^b	1.67±0.18 ^a	1.45±0.09 ^a	0.81±0.02 ^b	< 0.001
ALT(g/dl)	38.50±0.01 ^a	24.62±0.15 ^b	15.17±0.17 ^c	24.17±0.17 ^b	< 0.001
AST(g/dl)	57.50±0.02 ^b	82.17±0.31 ^a	21.5±0.34 ^c	19.17±0.17 ^d	< 0.001
ALP(g/dl)	167.17±0.6	154.00±0.63	170.67±0.6	169.33±18.27	>0.05 ^{NS}
SOD(U/mL)	132.00±0.26 ^a	63±0.58 ^b	6.23±0.24 ^c	5.35±0.19 ^c	< 0.001
GPX(U/mL)	267.00±0.37 ^a	24.17±0.17 ^d	134.67±1.2 ^c	142.33±0.67 ^b	< 0.001
MDA (nmol/L)	4.18±0.05 ^d	10.17±0.17 ^c	17.50±0.34 ^b	26.17±0.31 ^a	< 0.001

^{abcd} Means with different superscript within same row are statistically significant; statistical significance p <0.001., NS: not significant. HDL: high density lipoprotein; LDL: low density lipoprotein; ptn: protein; A/G: albumin to globulin; ALT: alanine aminotransferase; AST: aspartate aminotransferase; ALP: alkaline Phosphatase; SOD: superoxide dismutase; GPX: glutathione peroxidase; MDA: malondialdehyde.

Breast meat color and quality characteristics of four duck breeds was represented in Table (4). Duck breeds had an important influence on color values for Lightness (L*), Redness (a*), and Yellowness (b*). Muscovy duck had higher (a*) and (b*) values (13.12 and 11.23) than other breeds and the lowest (L*) value (45.33). The lowest a* value was recorded for the Pekin duck (7.82).

Mulard duck had the lowest WHC (45.33%) with the highest drip loss (3.8%) and cooking loss (24.4%). The highest WHC (72.67%), PH (6.84), and TBARS (0.32 mg/kg) were denoted for Star 53 ducks. While the highest shear force value (20.77 kg) was shown in Pekin ducks. Muscovy ducks had the lowest cooking loss, shear force, and TBARS.

Table 4.comparative breast meat color and quality characteristics of four duck breeds

Parameters	Pekin	Star 53	Muscovy	Mulard	P-value
pH	6.15±0.01 ^b	6.84±0.03 ^a	5.74±0.01 ^c	5.73±0.03 ^c	< 0.001
L*	47.50±0.22 ^c	55.77±0.05 ^a	45.33±0.33 ^d	51.77±0.05 ^b	< 0.001
a*	7.82±0.09 ^d	10.72±0.05 ^b	13.12±0.02 ^a	10.30±0.04 ^c	< 0.001
b*	2.23±0.12 ^c	2.35±0.1 ^c	11.23±0.02 ^a	4.35±0.1 ^b	< 0.001
WHC (%)	61.00±0.37 ^b	72.67±0.21 ^a	47.17±0.31 ^c	45.33±0.21 ^d	< 0.001
Drip loss (%)	1.47±0.02 ^c	1.23±0.05 ^d	1.76±0.00 ^b	3.80±0.04 ^a	< 0.001
Cooking loss (%)	21.97±0.11 ^b	19.43±0.15 ^c	10.43±0.06 ^d	24.40±0.07 ^a	< 0.001
shear force (kg/cm ²)	20.77±0.22 ^a	18.47±0.14 ^b	10.87±0.3 ^d	14.17±0.13 ^c	< 0.001
TBARS (mg/kg)	0.21±0.01 ^b	0.32±0.01 ^a	0.12±0.01 ^c	0.25±0.01 ^b	< 0.001

^{abcd} Means with different superscript within same row are statistically significant; statistical significance < 0.001; b*: Yellowness; L*: Lightness; a*: redness; WHC: water holding capacity; PH: Potential of hydrogen; TBARS: Thiobarbituric Acid Reducing Substances.

Thigh meat color and quality characteristics of four duck breeds were presented in Table (5). Duck breeds had a significant effect on color values for Lightness (L*), Redness (a*), and Yellowness (b*). Muscovy duck had higher (a*) and (b*) values (11.4 and 10.6) than other breeds and the lowest (L*) value (44.4). The lowest a* value was recorded for the Pekin duck (5.82). Mulard duck had the highest drip loss (2.12%) and cooking loss (31.17%). The highest WHC (73.7%) and PH (7.84) were denoted for Star 53 ducks. While the highest shear force value (20.8 kg) was shown in Pekin ducks. Muscovy ducks had the lowest shear force and TBARS.

Table 5. Thigh meat color and quality characteristics among different investigated duck breeds.

Parameters	Pekin	Star 53	Muscovy	Mulard	P-value
pH	7.15±0.009 ^b	7.84±0.028 ^a	5.89±0.026 ^c	5.92±0.018 ^c	< 0.001
L*	48.60±0.24 ^c	50.60±0.13 ^b	44.40±0.04 ^d	51.8±0.05 ^a	< 0.001
a*	5.82±0.09 ^d	8.72±0.05 ^c	11.4±0.08 ^a	10.30±0.045 ^b	< 0.001
b*	2.50±0.12 ^c	2.65±0.09 ^c	10.60±0.06 ^a	4.35±0.096 ^b	< 0.001
WHC (%)	62.00±0.37 ^b	73.70±0.21 ^a	47.50±0.23 ^c	48.17±0.17 ^c	< 0.001
Drip loss (%)	1.47±0.02 ^{bc}	1.23±0.05 ^c	1.72±0.004 ^b	2.12±0.19 ^a	< 0.001
Cooking loss (%)	19.40±0.15 ^b	15.45±0.07 ^c	15.45±0.07 ^c	31.17±0.20 ^a	< 0.001
shear force (kg/cm ²)	20.8±0.22 ^a	18.47±0.14 ^b	11.43±0.20 ^d	14.67±0.15 ^c	< 0.001
TBARS (mg/kg)	0.21±0.013 ^b	0.317±0.015 ^a	0.14±0.011 ^c	0.28±0.013 ^a	< 0.001

^{abcd} Means with different superscript within same row are statistically significant; statistical significance < 0.001; b*: Yellowness; L*: Lightness; a*: redness; WHC: water holding capacity; PH: Potential of hydrogen; TBARS: Thio-barbituric Acid Reducing Substances.

GH, *IGF1* and Calpain genes expression in four duck breeds was revealed (Figure 2). The Mulard had considerably ($p < 0.05$) increased expression of *GH*, *IGF1* than other breeds. In contrast, Muscovy recorded noticeably ($p < 0.05$) increased expression of the Calpain gene than Mulard, Star 53, and Pekin duck.

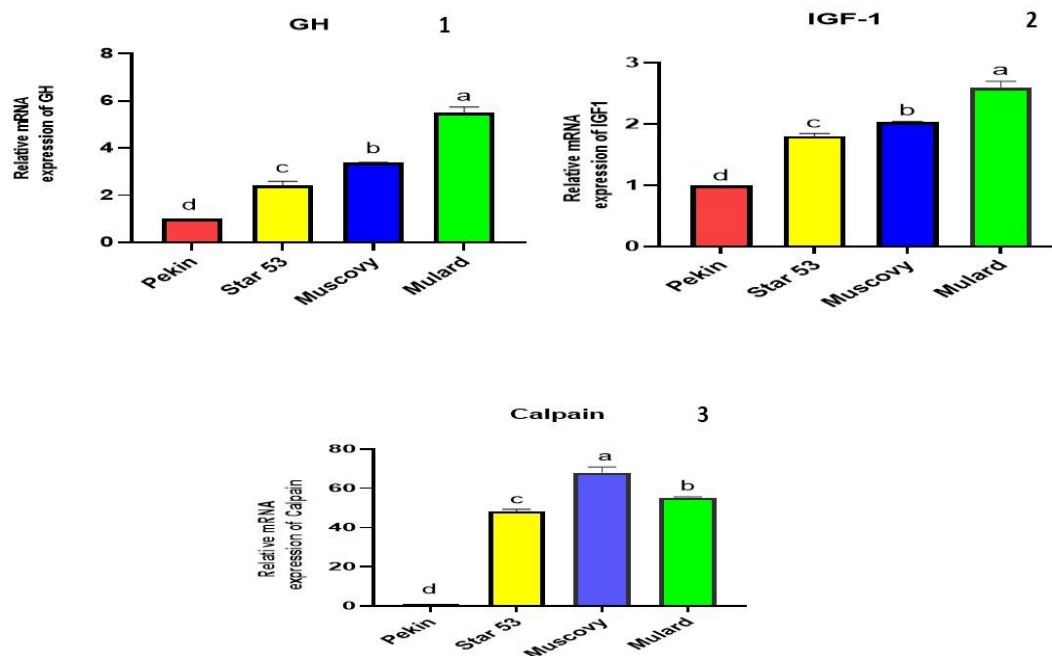


Figure 2. Graphical presentation of real-time quantitative PCR analysis of the expression of (1) *GH* (Growth Hormone), (2) *IGF-1* (Insulin growth factor-1) and (3) *Calpain* genes in breast Muscles of different duck breeds (mean \pm SEM) at 12th week. Groups with different letters are significantly different ($P < 0.05$, one-way ANOVA).

Discussion

The present experiment studied the breed effects on productive traits, biochemical parameters, meat quality, and gene expression of Pekin, Star 53, Muscovy, and Mulard ducks to investigate the greatest breed with the best qualities. This study demonstrated that the Mulard has the highest BW than other breeds. These results were in agreement with those reported by Omar *et al.* [37], and Nasr *et al.* [38] who stated that Mulard was 4,021 g at the 10th week of age. On the contrary, Galal *et al.* [3], and Hassan *et al.* [39] reported that the Muscovy showed the highest BW. However, Star 53 breed was higher than Mulard ducks which were slaughtered at the 8th week of age [40].

The result of feed intake was contrary to Galal *et al.* [3] who reported that Muscovy consumed larger feed than Pekin ducks. Also, Abdel-Rahman and Mosaad [41] showed that feed consumption for Muscovy ducks was 25.419 kg/duck. However, Hassan *et al.* [39] stated that Muscovy consumes the lowest amount of feed which was consistent with our study. The significant variations between different breeds in productive traits could be attributed to differences in genetic makeup [42].

Body measurements were obviously influenced by breeding and rise with age. The shank length of Star 53, Mulard, Muscovy, and Pekin at 6 weeks measured 7.33 cm, 7.05 cm, 6.80 cm, and 6.68 cm,

respectively. Therefore, Star 53 had a longer shank length than other strains. These results were in disagreement with Makram *et al.* [26] who recorded that the Pekin duck had a longer shank length (6.65 cm) than Muscovy (6.09 cm). While Makram *et al.* [43] found that the Muscovy duck had higher sexual dimorphism for shank length compared to Pekin ducks. The difference between different breeds in shank length was related to a positive correlation between BW and shank length [10]. Mulard duck had significantly higher BW, body circumference and body length, keel length and shank at 12th week of age compared with Star 53, Muscovy, and Pekin ducks. There was a positive relationship between chest circumference and BW [44]. Książkiewicz and Mazanowski [45] reported breast circumference in Mulard was 40.15 cm at 12th week of age which is in accordance with our study. While Muscovy ducklings had larger breast circumference, keel length, and shank length at 4 to 12th weeks of age than Sudani ducklings [46]. The difference in body measurement between breeds suggests distinct and unique in terms of body biometric characteristics [47].

Biochemical parameters were critical tools for predicting metabolic disorders [48]. The blood changed when the bird was under stress due to thermoregulatory mechanisms [49]. ALT and triglyceride values of Pekin duck were in contradiction with those of Arak *et al.* [50] who mentioned that ALT and triglyceride values were 155.16 mg/dl and 46.64 IU/L, respectively. Total cholesterol in Pekin was higher than in other breeds which may be attributable to the increased production of cholesterol by the liver and decreased mobilization by tissues [51]. Lower concentrations of total proteins in Star 53 may be a consequence of higher protein and amino acid requirements for somatic

development. Similar findings have been found in chickens and guinea fowl [52,53]. The values of biochemical parameters for Muscovy and Mulard agreed with Nasret *al.* [38]. Also, the results of total proteins, albumin, and globulin for Mulard were comparable with those reported by Omaret *al.* [37] but Muscovy values were consistent with that reported by previous researchers [49]. On the contrary, El-Fiky *et al.* [54] found that values of HDL in Muscovy (83.40 mg/dl) were higher than in Mulard (77.47 mg/dl). Free radicals and other reactive oxygen species cause cell destruction, and antioxidant capacity contributes to decreasing oxidative stress “the imbalance between reactive oxygen species and antioxidants” [55].

In the antioxidant system, superoxide dismutase operated as the initial line of protection against damage by converting oxygen free radicals into hydrogen peroxide (H₂O₂). GPX was regarded as a critical peroxide breakdown enzyme and prevented additional oxidative damage which eliminated H₂O₂ and lipid peroxides [56]. The values of serum SOD, and GPX in Pekin were inline with those reported by Ao and Kim [57]. Pekin ducks had the highest blood serum levels of SOD and GPX enzymes than other breeds and were thought to be more oxidative stress tolerant and enhance immune function [58,59]. MDA was considered a radical oxidative marker and one of the byproducts of lipid peroxidation in cell membranes and its content could be used to assess the threshold of oxidative stress in an organism [60]. Values of antioxidant enzymes for Muscovy and Mulard agreed with Nasret *al.* [38]. Variation in antioxidant breeds may be possible due to genetic regulations suggesting these selective breeding

of ducks for new strains with higher oxidative stress tolerance.

Muscle lactic acid was a pH indicator, and high accumulations of it resulted in lower meat quality [61]. The pH of Muscovy meat (breast and thigh) was between 5.74 and 5.89 [62-64] and was lower than those reported by Nasr *et al.* [38]. There was no considerable alteration in the pH of Muscovy and Mulard and these results were maintained by others [65]. The value obtained for the pH of breast Pekin was similar to those reported previously [66] and higher than those found by Huda *et al.* [67], and Michalczuk *et al.* [68]. While the pH for Star 53 breast meat was in accordance with Kokoszyński *et al.* [69] and lower than those obtained by [70]. Additionally, we noticed that the highest pH value in Star 53 breast and thigh was due to the lowest glycogen leading to improved meat shelf life [71]. In general, variations in pH values can be attributed to variations in the levels of glycogen at slaughter, reactions to stress before slaughter, or slaughter weight.

Meat color was used as a guide for meat quality and freshness [72], and directly associated with the ultimate pH [73]. The color of meat was primarily influenced by the myoglobin content and nature, the composition, and the physical state of muscle [74,75]. L*, a*, and b* colors of the breast and thigh muscle of star 53 duck agreed with those reported previously [70]. While the values of Pekin were similar to those reported by Kokoszyński *et al.* [76] and were lower than those reported by Zhenget *al.* [65]. Muscovy values were similar to those reported by Zhu *et al.* [64] and disagree with others [65]. Mulard breast values disagreed with Zhenget *al.* [65] recorded L*, a*, and b* values (37.87, 19.91, and 7.5, respectively) at 70 days of

age. In this study, the Muscovy duck had the highest redness color and lowest lightness than other breeds. These findings did not support those reported previously [3,77] in which the Muscovy ducks had lighter breast muscles (L*) than Pekin ducks. The lowest L* value in Muscovy duck breast and thigh explained the association between iron concentration and meat color, which was described in an earlier study of Kokoszyński *et al.* [47]. Significantly superior redness values were documented for the breast meat of Muscovy compared to Mulard ducks, and this was confirmed in an earlier study [65]. In contrast, the meat of Muscovy ducks was recorded to be graded lower for color, compared with the breast meat of Rouen ducks [78]. This difference could be related to the age at which ducks are slaughtered.

Water holding capacity means the ability of meat to retain water. Water will flow out gradually from the myofibril enhancing the length of postmortem time [79]. WHC% breast and thigh in Pekin and Muscovy were similar to those found by Huda *et al.* [67]. While Mulard duck breast was lower than Janiszewski *et al.* [80]. The difference in WHC within the breed was supported by the findings of other researchers [62,67,68]. The present investigation revealed that Star 53 duck had the highest value, and this may be attributable to differences in genotype and muscle fiber sizes [81]. A greater WHC was very essential for the food industry, as it could contribute positively to lowering the final product weight loss that happened during storage [67].

Drip loss referred to the percentage of water lost from meat and was considered an indirect marker of WHC subsequently, drip loss was adversely correlated with WHC [72]. Drip loss values for Muscovy, Pekin, and Mulard were lower than

reported by Eratalaret *al.* [70]. While Star 53 drip loss disagreed with Kokoszyński *et al.* [82]. Drip loss increased in Mulard ducks, which was supported by Nasret *al.* [38], and Zhenget *al.* [65]. The highest drip loss in Mulard may be due to low pH which promotes muscle fiber contraction, producing high drip loss [83]. The meat from Mulards had considerable quantities of free water, which suggested it was not very suitable for processing [62].

Cooking loss declined when the WHC was higher. The cooking loss of Pekin and Muscovy was contrary to the previous [67]. While Mulard disagreed with Zhenget *al.* [65]. The meat of Muscovy had the lowest cooking loss value followed by Star 53, Pekin, and Mulard. These findings were supported by Pingel [84], and Baeza *et al.* [85] who reported that the Muscovy ducks were characterized as meat of better quality than Pekin ducks. On the contrary, Wawro *et al.* [62], and Omojola [78] showed that Pekin ducks revealed superior meat quality. The amount of water reserved in meat after cooking has been related to the juiciness, palatability, and marketable weight of the products [86].

Another meat trait that was significant to consumers and processors was tenderness, which was evaluated using shear force values [67]. There was a significant difference in Shear force values among breeds. These results were supported by Omojola [78]. However, another researcher didn't observe any difference due to strain (Pekin, Muscovy, or their crossbred offspring) [57,77]. In this study, Shear force values for Pekin and Muscovy were higher than those reported by Omojola [78]. Muscovy recorded the lowest shear force. However, Pekin noted the highest value. These results disagreed with Kokoszyński *et al.* [76], and Omojola [78] reported that the

shear force in Pekin was lower than in Muscovy. These authors recorded shear force in Pekin ducks at 7 weeks and 12 weeks of age in Muscovy. A greater shear force in Pekin may be associated with the growth of the muscle tissue because the cross-sectional area of muscle fiber was enhanced with age [87].

The development of off flavors (rancidity) was due to lipid oxidation, which could be determined by measurement of the degradation products such as thio-barbituric acid reactive substance [88]. Undesirable changes during storage at both refrigeration and freezing temperatures for long storage periods lead to lipid deterioration and liberation of free fatty acids [89]. The Value of TBARS in Pekin was similar to that reported by Jeong [66]. While in Muscovy was contradictory with Zhu *et al.* [64] who stated that the TBARS from breast meat of Muscovy ducks was 2.82 mg/kg and 1.36 mg/kg for thigh meat.

GH and *IGF-1* were candidate somatotrophic axis genes that improved growth performance and carcass trait characteristics in chickens [90,91]. *GH* gene was a great genetic marker for improving the genetic potential of ducks [92]. *IGF-1* was a potential gene for chickens' growth, body composition, metabolism, skeletal features, development of adipose tissue, and fat deposition [93]. The current results demonstrated considerable variations in *GH* and *IGF-1* expression amongst the different breeds. These findings were supported by others [94,95]. The Mulard exhibited the highest expression, which may explain why it had greater muscular growth. *IGF-1* gene expression findings differ from those of Hassan *et al.* [39], who reported that Muscovy ducks had the most significant *IGF-1* gene expression, followed by

Mulard and Pekin ducks. This difference was related to a positive correlation between *IGF-1* concentration and body weight [96]. Transgenic over-expression of *IGF-1* in mice had shown that *IGF-1* mRNA expression was correlated with enlarged body weight. Body weight increased by 30% when *IGF-1* was overexpressed at 1.5 times the normal levels [97]. Furthermore, Wang *et al.* [98] showed that the *IGF-1* gene is tightly linked to both the body size and weight of chickens.

The Calpain gene was especially connected to connective muscle in areas where proteolysis had been related to post-mortem meat tenderness [99]. Also, Gandolfi *et al.* [100] reported that elevated calpain activity stimulated the cleavage of certain myofibrillar proteins such as titin, desmin, and vinculin, which enhanced tenderness. Piórkowska *et al.* [101] referred to the primary determinant of meat tenderness as a shear force. A low shear force revealed tender or soft meat. High shear force indicates that the meat has low proteolytic activity which is correlated with its hardness. Previous studies have shown a close connection between the calpain system and the qualities of pig meat [100], sheep [102], and chicken [103]. In this study, it was observed that the level of the Calpain gene was higher in Muscovy ducks. This breed was characterized by lower shear force. These results were consistent with previous [101] which reported that the Calpain gene was expressed at a lower level in birds with high shear force (low tenderness) than in broilers with low shear force. Contradicting our findings by Liao and Chou [104] that the difference in meat tenderness between Muscovy and Pekin duck muscles was caused by quicker calpain-1 activation in postmortem Pekin breast muscles, this might indicate that

calpain is involved in the tenderization process of duck breast muscle. These findings were confirmed by the studies of other researchers regarding the role of calpain in postmortem tenderization [24, 105-107], and goose [108].

Conclusions

This study revealed that Mulard ducks had the highest productive traits and high expression of GH, and IGF1. Muscovy ducks had better meat quality and high expression of the Calpain gene. This illuminates the crucial role played by conventional breeding programs in influencing molecular genetic systems that result in improved genotypes with potential productive qualities and meat quality.

Conflict of interest

There are no conflicts of interest among the authors to reveal.

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الملخص العربي

الصفات الانتاجية والقياسات البيوكيميائية وجودة اللحوم والتعبير الجينيلهذه الصفات فى سلالات البط المختلفة

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تهدف هذه الدراسة إلى تقييم الصفات الإنتاجية والقياسات الكيميائية الحيوية وجودة اللحوم والتعبير الجيني لهرمون النمو (GH) ووجين (IGF-1) ووجين (Calpain) فى اربعة سلالات من البط [البكينيوالفرنساوي البيوروالمسكوفى والبغالى]. أجريت هذه التجربة على 80 بطة (20 من كل سلالة) ولقد تم وضعهم تحت نفس الظروف البيئية والرعاية من عمر يوم واحد حتى نهاية التجربة (الأسبوع الثاني عشر من العمر). حيث تم تقييم أداء النمو وقياس تحاليل الدم البيوكيميائية وجودة اللحوم والتعبير الجيني لجينات النمو ووجين (Calpain). أظهرت النتائج أن البط البغالى أعلى نمو للجسم من السلالات الأخرى بينما البط البكينى كان أعلاهم فى معامل التحويل الغذائى. بالإضافة إلى ذلك أظهر البط البغالى أعلى قياسات للجسم فى نهاية التجربة. عند قياس تحاليل الدم البيوكيميائية كانت معدلات (TC), (SOD), (GPX) عالية فى البط البكينى. بينما قياسات (HDL), (LDL), (MDA) زادت فى البط البغالى. وعند تقييم جودة اللحوم أظهرت قوة ألياف لحم الصدر والفخذ أقل قيمة فى البط المسكوفى. وجد أن جين النمو (GH) ووجين (IGF-1) زاد فى البط البغالى عن السلالات الأخرى ولكن تعبير (Calpain) جين كان أعلى فى البط المسكوفى. نستنتج أن زيادة الصفات الإنتاجية فى البط البغالى ترجع إلى الاختلاف فى التعبير الجيني للنمو ولكن جودة اللحوم الجيدة فى المسكوفى قد تعزى إلى اختلاف تعبير (Calpain).