

Article



# Performance, Carcass Composition, and Meat Quality during Frozen Storage in Male Layer-Type Chickens

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Abstract: An experiment was carried out in the Institute of Animal Science-Kostinbrod, Bulgaria, to investigate the growth performance of male layer-type chickens (Lohmann Brown Classic), raised to 6 and 9 weeks of age, to evaluate the economic aspects of this rearing, as well as to present changes in the quality characteristics of the meat during frozen storage. The chickens were reared in a controlled microclimate with an initial stocking density of 9 birds/ $m^2$ . After 6 weeks of age, fragmentation of the stocking density was applied, and then it diminished to 3 birds/m<sup>2</sup>. The chickens were slaughtered at 6 and 9 weeks of age. Ten 9-week-old chickens were subjected to carcass analysis. Meat quality parameters (pH, color), degree of proteolysis (free amino groups), and lipid oxidation (content of peroxides and TBARS) were assessed in fresh breast and thigh meat (0 d) and in samples stored for 60 and 120 days at -18 °C in chickens slaughtered at 6 and 9 weeks old. The mean live weight of the male layer-type chickens at 6 weeks was 608.81 g, while the 9-week-old chickens reached 1115.93 g. The feed conversion ratio (FCR) for the whole period of rearing was 2.75. There were no considerable deviations in the meat traits, indicating quality deterioration over the course of the frozen storage. There was a significant increase in the pH of the breast and thighs, reaching maximum values for 60 days of storage in the 6-week-old chicks, while in the 9-week-old birds, pH peaked in the samples stored for 120 days. The changes in the dynamics of pH corresponded to those of proteolysis. There was an increase in lightness (L\*), allowing for higher values in the samples stored for 60 days to be reached regardless of the type of meat and age of the chickens. The content of the peroxides increased considerably for 60 days of frozen storage and decreased afterwards. During storage, there was a constant increase in the secondary products of lipid oxidation. Our results indicated that the application of practices such as the fragmentation of stocking density and finding the suitable age for slaughter have significant importance for the profitability of producing meat product from male layer-type chickens. We found that rearing this type of bird until 9 weeks of age resulted in lower costs and higher economic efficiency.

Keywords: male layer-type chickens; growth performance; economics; meat; frozen storage

# 1. Introduction

Meat is an important component in the human diet. It provides high value protein, iron, vitamin B12, zinc, selenium, and phosphorus. Removing it from the diet poses a risk of severe nutritional deficiencies, imbalanced diet, and impaired human health [1]. Poultry meat makes no exception, and it is even considered healthier compared to red meats, having a lower content of saturated fatty acids but still being a very good source of protein. The inclusion of poultry meat in the diet, especially when substituting red meat, has been



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**Copyright:** © 2024 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). associated with a reduced risk of developing cardiovascular diseases [2], obesity, and type 2 diabetes mellitus [3,4]. Furthermore, poultry consumption might decrease the risk of certain cancers [5,6]. Chicken meat is the most consumed among all the poultry meats [7]. Fastgrowing broilers have been the main source of chicken meat; however, in recent decades, consumers have become more and more aware of the benefits of eating meat from slowgrowing chickens. In comparison to fast-growing broilers, the latter have a more favorable nutritional profile in terms of protein and lipid content [8] and fatty acid composition [9]. An alternative source of this meat could be male layer-type chickens. In fact, their rearing for meat has been considered as an option in addition to in ovo sex determination and the development of dual-purpose breeds to avoid the common practice of culling them immediately after hatching [10]. Based on previous studies, showing that the meat of male layer-type chickens is not inferior to fast- and slow-growing chickens, we focused on their age at slaughter as a factor to produce high-quality meat and meat products, as well as achieve more economically efficient production. Since the raising of male layer-type chickens is a longer and less profitable process, to avoid accumulating higher costs, we applied fragmentation of the stocking density at a certain age to the birds, that being the age of first slaughter. This method has been modified in the Institute of Animal Science-Kostinbrod, and its advantages are described in more detail elsewhere [11]. Our previous studies have shown that the most appropriate age for applying density fragmentation is 5 weeks. However, since the carcasses of the slaughtered birds at this age are quite small (approx. 200 g) [11], for the purposes of this study, we decided to decrease the stocking density of the birds at 6 instead of 5 weeks of age and to assess the economic efficiency of the process.

After harvesting the meat, it is of crucial importance to store it properly so that its quality and nutritional value are maximally preserved. Freezing is one of the most important preservation methods that allows for a longer shelf life of meat and meat products to be generated, including poultry. However, during frozen storage, meat components can be involved in many reactions, such as oxidation. Oxidative reactions in meat can significantly contribute towards quality deterioration, negatively affecting color, pigments, flavor, texture, and nutritional value. Poultry meat is particularly susceptible to oxidation due to its high contents of polyunsaturated fatty acids [12]. Although extensive research has been carried out on the changes in the quality of poultry meat during lowtemperature long-term storage, such studies on male layer-type chickens remain rather scarce. So far, studies on the meat of male layer-type chickens have focused on comparisons with fast-growing genotypes and local or dual-purpose breeds [13–16], as well as on the effect of production systems [17]. This study was conducted to present the growth performance of male layer-type chickens, raised to 6 and 9 weeks of age, economical evaluation of this rearing, as well as the changes in the quality characteristics of the meat during frozen storage.

# 2. Materials and Methods

# 2.1. Experimental Birds and Housing

This trial was carried out in the experimental poultry farm of the Institute of Animal Science—Kostinbrod, Bulgaria, involving a total of 800 one-day-old Lohmann Brown Classic male chickens, supplied by Bulagro AD. The birds were distributed into 5 pens, each containing 160 chickens, and were reared conventionally until 9 weeks of age at an initial stocking density of 9 chickens/m<sup>2</sup>. When the chickens reached 6 weeks of age, fragmentation of the stocking density was applied to reach 3 birds/m<sup>2</sup>. The decrease in the stocking density of the chickens was achieved through the preliminary weighing of each bird and differentiation by live weight. By applying the fragmentation of the density, approximately <sup>1</sup>/<sub>4</sub> of the chickens set for trial remained and were reared until 9 weeks of age. For each of the groups, the limit weight was individually determined, and all the birds of a lower weight ( $\leq$ 540 g) were slaughtered. Male layer-type chickens were reared in deep litter and a controlled microclimate. The lighting regime was 3 h light and 3 h dark,

repeated during the 24 h cycle. Feed and water for the birds were provided ad libitum. Their diet consisted of standard broiler feed containing maize, wheat, sunflower meal, and soybean meal (crude protein 20%, crude fat 5.99%, crude fiber 4.40%, Ca 0.94%, P 0.80%, Lys 1.30%, Met 0.53%, metabolizable energy 3000 kcal/kg) and was purchased from a certified feed supplier. Water was provided through gravity drinkers. During the trial period, the live weight (LW) and feed intake (FI) of the birds were controlled weekly. These indicators were further used to calculate the body weight gain for the controlled periods (BWG) and the feed conversion ratio (FCR) of the male layer-type chickens.

#### 2.2. Slaughtering, Carcass Analysis, Sampling, and Storage

The chickens were slaughtered at the age of 6 and 9 weeks in a certified poultry abattoir in Pazadrzik, Southern Bulgaria. After stunning, decapitating, and bleeding, the carcasses were plucked and eviscerated. Their feet and edible viscera (heart, liver, gizzard) were removed in order to obtain the ready-to-cook carcass. The carcasses were then placed in a refrigerator at 4 °C for 24 h. On the next day, ten 9-week-old chickens were subjected to carcass analysis. Each of the carcasses were weighed and cut to neck, breast, thighs, wings, and back. Sampling for meat quality analysis and storage was performed in the breast and thighs of chickens at both ages (n = 10). Briefly, the breast and thighs of each carcass were collected, and their skin and bones were removed. A part of the samples was used for the analysis of the meat quality and oxidative stability on the same day before freezing (day 0), while the others were frozen and stored at -18 °C for subsequent analysis after 60 and 120 days.

### 2.3. Economic Evaluation

Methodology of Calculation of the Costs:

The total costs for obtaining kg live weight and kg product by the chickens were calculated according to the following equations:

$$LWC = \frac{(\sum_{i=1}^{n} FC_i + \sum_{j=1}^{m} VC1_j)}{TLW}$$
(1)

$$TC = \frac{\left(\sum_{i=1}^{n} FC_{i} + \sum_{j=1}^{m} VC1_{j} + \sum_{k=1}^{l} VC2_{k}\right)}{TPW},$$
(2)

where *TLW* is the total live weight of all the chickens (kg), *TPW* is the total weight of the ready product obtained (ready-to-cook carcass) (kg); LWC denotes the costs for rearing the chickens before slaughter (EUR/kg)—as all the costs are further presented in the same units—and TC denotes the total costs to obtain kg of ready product.

 $FC_i$  are fixed costs, comprising the rent (depreciation) of the building used for rearing the chickens and preparation of the premises, including covering and preliminary heating the one-day-old chickens.

 $VC1_j$  are a group of variable costs that are used to calculate the cost price of the live weight of the chickens and comprise labor costs; price of a one-day-old chicken; cost for vaccines and decontamination of the premises; energy costs; and feed costs.

 $VC2_k$  are a second group of variable costs used to calculate the price cost of the ready product: costs for slaughter; transportation; and packaging and freezing.

## 2.4. Analysis of Meat Quality

The analysis of the meat quality parameters was carried out in the Department of Technology of Meat and Fish Products at the University of Food Technology, Plovdiv, Bulgaria.

## 2.4.1. Measurement of Meat pH and Color

pH and color were measured on the fresh meat (day 0) and on day 60 and day 120 of the frozen storage. The pH measurements were performed using a portable pH meter (HI99163, Hanna Instruments, Inc., Smithfield, RI, USA) equipped with a specialized probe

(FCO99) with a conical tip and stainless-steel blade. Calibration prior to use at pH 4.0 and 7.0 was performed. The color of the breast and thigh meat surface was described by the CIELab color space. The lightness (L\*), redness (a\*), and yellowness (b\*) were measured using a chroma meter CR-410 (Konica Minolta Holding, Inc., Ewing, NJ, USA) at the following settings: aperture = 8 mm, standard observer  $2^{\circ}$ , and illuminant D65. The instrument was calibrated using a standard white plate (Y = 94.3, x = 0.3134 and y = 0.3197). Measurements of the pH and color were performed at three locations in the meat samples, and the results were averaged.

#### 2.4.2. Free Amino Groups

The content of the free amino groups was determined in muscle proteins after extraction, as described Khan et al. [18], with slight modifications. Meat samples (2.5 g) were homogenized with 48.5 mL PBS buffer (49 mM Na<sub>2</sub>HPO<sub>4</sub>.7H<sub>2</sub>O, 4.5 mM NaH<sub>2</sub>PO<sub>4</sub>.H<sub>2</sub>O, KCl to obtain I = 0.55). The homogenate was kept in a refrigerator for 12 h and then centrifuged at  $1000 \times g$  for 15 min. The free amino groups were measured according to Vassilev et al. [19] as follows: 2 mL of supernatant was transferred in a test tube and mixed with 1 mL solution of ninhydrin reagent (0.5% ninhydrin; 10%Na<sub>2</sub>HPO<sub>2</sub>·12H<sub>2</sub>O; 6%KH<sub>2</sub>PO<sub>4</sub>; 0.3% fructose). The mixture was heated in boiling water for 16 min and then cooled at room temperature for 20 min and diluted with 5 mL water–ethanol (3:2) solution of KIO<sub>3</sub> (2%). The absorbance was read at 570 nm against the blank sample using a Camspec, M 550 two-beam UV-VIS spectrophotometer (Spectronic Camspec Ltd., Leeds, UK). The concentration of free amino groups was calculated using a standard curve made by leucine and presented as mg leucine/g meat.

# 2.4.3. Lipid Oxidation

The lipids from the breast and thigh meat were extracted according to the method of Bligh and Dyer [20].

The peroxide value was determined as described by Shantha and Decker [21]. The extracted lipid (0.1 g) was mixed in a glass tube with 50  $\mu$ L iron (II) solution and 50  $\mu$ L NH<sub>4</sub> SCN (300 mg/mL) and CHCl<sub>3</sub>:CH<sub>3</sub>OH (3:5, v/v) to form a final volume of 10 mL. The samples were incubated for 5 min at room temperature, and the absorbance was determined by a spectrophotometer at 507 nm against a blank containing all the reagents except the sample. The results were presented as meqO<sub>2</sub>/kg lipid.

Thiobarbituric acid reactive substances (TBARSs) were determined according to the method described by Botsoglou et al. [22] with slight modifications. A total of 10 g of meat was homogenized with 50 mL NaCl (0.9%) and left for 5 min. Further, 50 mL of trichloroacetic acid (10%) was added, and the samples were filtered through grade 391 filter paper. The filtered samples (4 mL) were then mixed with 1mL 2-thiobabituric acid (1%) and were incubated at 70 °C for 30 min. After cooling to room temperature, the absorbance of the samples was determined at 532 nm against a blank containing distilled water instead of the sample. The TBARS concentrations were calculated using 1, 1, 3, 3 tetraethoxypropane as standard. The results were expressed as mg MDA/kg meat or TBARS units.

#### 2.5. Statistical Evaluation

Statistical evaluation of the data was performed using JMP v.7 software package [23]. A one-way ANOVA was applied to assess the effect of the storage duration on the examined quality traits and oxidation in the breast and thigh meat of the chickens slaughtered at 6 and 9 weeks of age. In the case of significance, the difference between means was further assessed through post hoc comparisons (Tukey HSD, p < 0.05). Data were presented as means and standard deviations.

# 3. Results

# 3.1. Growth Performance and Carcass Composition

The initial live weight of the chickens was 36.43 g. At 6 weeks of age, when density fragmentation was applied, the average live weight of the male layer-type chickens was 601.81 g, and until the end of the experiment (9 weeks of age), their weight increased to 1115.93 g (Figure 1).



Figure 1. Live weight of the male layer-type chickens during the experiment.

As seen from Table 1, the weight gain of the male layer-type chickens increased until the 6th week.

**Table 1.** Dynamics of growth performance traits of the male layer-type chickens during the trial period.

Trait	BWG, g	FI, g/Bird	FI Cumulative g/Bird	FCR Feed/Gain
1 week	30.52 (3.16)	65.00 (1.20)	65.00 (1.20)	2.15 (0.25)
2 week	55.40 (1.73)	74.32 (2.62)	139.32 (3.06)	1.34 (0.06)
3 week	76.79 (1.12)	183.07(2.47)	322.38 (4.47)	2.38 (0.04)
4 week	111.32 (3.98)	250.04 (4.21)	572.43 (8.14)	2.25 (0.10)
5 week	117.25 (7.74)	349.40 (2.68)	921.83 (8.46)	2.99 (0.18)
6 week	181.10 (11.04)	369.00 (0.99)	1290.83 (7.53)	2.04 (0.13)
7 week	151.79 (12.67)	464.79 (16.55)	1755.61 (15.71)	3.07 (0.15)
8 week	166.73 (20.75)	597.96 (21.28)	2353.57 (35.63)	3.63 (0.42)
9 week	188.60 (22.11)	618.92 (30.37)	2972.49 (63.31)	3.31 (0.31)

On the 7th and 8th week of the trial, the values of this trait decreased; however, body weight constantly increased. As presented by the data, despite the lower weight gain measured at the age of 7 and 8 weeks, the feed intake remained higher and was associated with higher values of FCR. Maximum weight gain was reached at the end of the experimental period (9 weeks). The chickens gained 572.38 g to reach the maximum weight for the first slaughter at 6 weeks with FCR 2.25, while the FCR for the whole period was 2.75.

The weight at slaughter for 6-week-old chickens was 527.42 g, and for the birds at 9 weeks it was 1110.13 g. The carcass yield for the younger chicks was 57.58%, while for the 9-week-old birds the value of this parameter was 65.18%. The carcass analysis of the male layer-type chickens showed that thighs had the highest proportion of all parts, followed by breast and back (Figure 2).



Figure 2. Proportion of the carcass parts in male layer-type chickens (9 weeks old).

3.2. Economic Evaluation of the Rearing of Male Layer-Type Chickens

Comparisons of the costs to produce 1 kg of live weight between the chickens raised to 6 and 9 weeks of age are shown in Table 2. The differences between both ages are small (0.07 EUR/kg) in favor of the chickens raised until 9 weeks old.

Table 2. Costs for producing 1 kg live weight of ma	ale layer-type chickens.
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Crown of Costs	Parameters —	Costs (EUR/kg Live Weight)		
Group of Costs		6 Weeks Old	9 Weeks Old	Difference
EC (Eived costs)	Preparing the premises	0.43	0.22	0.21
re (rixed costs)	Rent	0.76	1.07	-0.31
	Price of one day chicken	0.29	0.14	0.16
	Energy	0.76	0.70	0.06
VC1 (Variable costs 1)	Labor	0.96	1.26	-0.30
	Feed	1.49	1.41	0.07
	Medication	0.35	0.16	0.18
TC (Total costs)		5.03	4.96	0.07
Live weight (kg)		304.698	237.440	

Source: own calculation.

Table 3 presents the comparison of the costs made to produce 1 kg ready product between the two age groups. The differences are again in favor of the older birds; however, here they are considerable (1.84 EUR/kg).

Crown of Costs	Parameters -	Cost (EUR/kg Product Weight)		
Group of Costs		6 Weeks Old	9 Weeks Old	Difference
EC (Eined seats)	Preparation of premises	0.74	0.33	0.41
FC (Fixed costs)	Rent	1.32	1.64	-0.32
	Price of one-day-old chick	0.51	0.21	0.30
	Energy	1.32	1.07	0.25
VC1 (Variable costs 1)	Labor	1.66	1.93	-0.27
	Feed	2.58	2.16	0.42
	Medication	0.60	0.25	0.35
	Transport	0.73	0.83	-0.10
VC2 (Variable costs 2)	Slaughter	1.34	0.56	0.78
	Package	0.17	0.14	0.03
TC (Total costs)		10.97	9.13	1.84
Product weight (kg)		175.439	154.770	

Table 3. Costs for producing 1 kg ready product of male layer-type chickens.

Source: own calculation.

# 3.3. Meat Quality

## 3.3.1. pH and Color

The values of the pH in breast and thigh (Table 4) meat were significantly affected (p < 0.0001) by the storage period in the chickens slaughtered at 6 and 9 weeks of age. The pH of the breast and thigh meat samples of the 6-week-old birds stored for 60 days increased, but the values decreased in the meat stored for 120 days. The meat of the 9-week-old birds, however, had a significant increase in pH at the end of storage (120 days).

Table 4. pH and color of meat in male layer-type chickens.

Trait	Age (Weeks)	Storage (Days)			Significance (p)
Breast		0	60	120	
pН	6	6.06 (0.004) <sup>b</sup>	6.13 (0.02) <sup>a</sup>	5.87 (0.06) <sup>c</sup>	< 0.0001
-	9	5.87 (0.04) <sup>b</sup>	5.94 (0.07) <sup>b</sup>	6.24 (0.03) <sup>a</sup>	< 0.0001
L*	6	58.51 (1.25) <sup>b</sup>	61.86(1.81) <sup>a</sup>	58.53 (1.99) <sup>b</sup>	0.0001
	9	56.51 (0.28) <sup>b</sup>	59.76 (0.65) <sup>a</sup>	54.96 (1.20) <sup>c</sup>	< 0.0001
a*	6	13.60 (0.75) <sup>a</sup>	13.19 (0.99) <sup>a</sup>	9.54 (1.29) <sup>b</sup>	< 0.0001
	9	14.44 (0.17) <sup>a</sup>	7.66 (0.89) <sup>b</sup>	13.70 (1.36) <sup>a</sup>	< 0.0001
b*	6	8.46 (0.41) <sup>b</sup>	10.90 (0.68) <sup>a</sup>	6.19 (0.65) <sup>c</sup>	< 0.0001
	9	7.62 (0.19) <sup>a</sup>	6.76 (0.43) <sup>b</sup>	6.49 (0.85) <sup>b</sup>	0.0003
Thigh					
pН	6	6.55 (0.007) <sup>b</sup>	6.67 (0.03) <sup>a</sup>	6.54 (0.04) <sup>b</sup>	< 0.0001
	9	6.42 (0.03) <sup>b</sup>	6.41 (0.02) <sup>b</sup>	6.59 (0.008) <sup>a</sup>	< 0.0001
L*	6	52.06 (0.90) <sup>a</sup>	52.97 (1.60) <sup>a</sup>	47.91 (2.32) <sup>b</sup>	< 0.0001
	9	47.95 (1.42) <sup>b</sup>	53.10 (2.88) <sup>a</sup>	49.06 (1.52) <sup>b</sup>	< 0.0001
a*	6	16.52 (0.23)	16.01 (1.26)	16.26 (1.25)	0.5501
	9	18.49 (0.81) <sup>a</sup>	13.55 (1.49) <sup>c</sup>	16.96 (1.36) <sup>b</sup>	< 0.0001
b*	6	5.23 (0.67) <sup>c</sup>	8.65 (0.84) <sup>a</sup>	6.89 (0.88) <sup>b</sup>	< 0.0001
	9	4.86 (0.75) <sup>b</sup>	4.22 (0.95) <sup>b</sup>	7.76 (0.83) <sup>a</sup>	< 0.0001

Means connected with different letters within an age group differ significantly (p < 0.05).

The storage time had a significant effect on most of the color parameters of the meat (p < 0.0001); however, different trends for the change in their values were observed both according to the age of the birds and the type of meat. Lightness (L\*) significantly increased in the breast meat of 6- and 9-week-old chickens stored for 60 days and then decreased until 120 days of storage. Such a trend was registered in the thigh meat of 9-week-old chickens

as well. Similar to breast, the lightness of the thigh meat showed the lowest values at the end of the storage period. The dynamics of the changes in the redness (a\*) and yellowness (b\*) of the meat showed patterns in the breast and thighs of the chickens that differed in the different age groups. The redness of the breast of the 6-week-old chickens underwent negligible changes for 60 days of storage; however, this then significantly decreased for 120 days. Likewise, the yellowness of the breast was lowest in the samples stored for 120 days; however, the values of this parameter peaked in the middle of the storage period compared to those that were measured initially and at the end of storage. In the birds slaughtered at 9 weeks old, both redness and yellowness decreased over the course of storage as the redness showed substantially decreased values in the samples stored for 60 days.

The redness of the thigh meat was only affected by the storage period in 9-week-old birds, and its values decreased gradually in the samples stored for 120, with minimal values on day 60. On the other hand, b\* was affected by the storage of the thigh meat of both 6- and 9-week-old birds. The former displayed more yellow meat when stored for 60 days and decreased values of this trait for 120 days of storage. The thigh meat of the 9-week-old birds had higher b\* values at the end of storage.

### 3.3.2. Proteolysis in Meat

The content of the free amino groups showed similar trends of change during storage in breast and thigh meat depending on the age at slaughter of the chickens and was affected by frozen storage (Figure 3). There was a significant increase in the proteolysis in the breast and thighs of the younger birds stored for 60 days and a gradual decrease afterwards until day 120. In the meat samples stored for 60 days, the content of the free amino groups showed maximum values. In the meat of the older birds, we observed a constant increase in this parameter over the course of the frozen storage as the highest content was registered in the meat samples stored for 120 days.

### 3.3.3. Lipid Oxidation

Lipid oxidation in terms of PV showed that the content of peroxides was affected significantly by the storage period and showed similar trends of changes in the birds slaughtered at 6 and 9 weeks. The highest content of lipid peroxides was measured in the breast and thigh meat stored for 60 days. In the samples stored for 120 days, the levels of the peroxides significantly decreased (Figure 4).



Figure 3. Cont.



**Figure 3.** Content of free amino groups in the breast (**A**) and thigh (**B**) meat of male layer-type chickens during frozen storage. Means connected with different letters within an age group differ significantly (p < 0.05).



**Figure 4.** Content of peroxides in the breast (**A**) and thigh (**B**) meat of male layer-type chickens during frozen storage. Means connected with different letters within an age group differ significantly (p < 0.05).

The TBARS values showed a constant significant increase (p < 0.0001) over the course of storage in the breast and thigh (Figure 5) regardless of the age of the birds. The maximum TBARS contents were measured in the meat stored for 120 days and varied within 1.99–2.46 for the breast and 2.31–2.33 for the thigh meat.



**Figure 5.** Content of TBARSs in the breast (**A**) and thigh (**B**) meat of male layer-type chickens during frozen storage. Means connected with different letters within an age group differ significantly (p < 0.05).

# 4. Discussion

Raising male layer-type chickens for meat is one option to avoid their culling right after hatching. Yet, despite studies reporting the positive characteristics of their meat [13,24], this option is still considered challenging by many, mainly due to economic reasons. There are certain factors, however, that can affect the rearing of this type of bird to achieve lower costs. Stocking density and the suitable age for slaughter are important for economically efficient production. In a previous study [11], we focused on the growth performance of male layer-type chickens reared at an initial stocking density of 22 birds/m<sup>2</sup> until 5 weeks of age, when a part of the birds was slaughtered. For the present trial, we opted for a much lower initial density and higher age of first slaughter to obtain a higher weight for the ready-to-cook carcass, particularly for small chickens. After applying fragmentation of the

stocking density after 6 weeks of age to reduce it to 3 birds/ $m^2$ , the average final live weight of the birds was 1115.93 g. Though the difference in the final body weight is negligible, the body weight of the birds reared at a lower density before and after fragmentation remained higher than that previously reported for the birds reared at a higher stocking density [11]. The live weight of the male layer-type chickens that we measured in this study was considerably lower than that of Habig et al. [25] in Lohmann Brown males for the whole trial period. Similarly, other studies with male layer-type chickens reported live weights higher than ours until the 4th [26] and 5th week [27]. Additionally, Putra et al. [27] reported a lower growth rate of the birds after the 6th week, which we can explain with the fragmentation of the stocking density that we applied to select birds with a higher live weight for further rearing. The final live weight of the male layer-type chickens at 9 weeks of age was comparable to that of 3-week-old fast-growing broilers [14]. For the whole period until 9 weeks of age, the FCR was 2.75. This value was lower than that reported by Mueller et al. [15] for Lohmann Brown chickens reared for 67 and 84 days, but it is closer to that determined for dual-purpose breeds and slow-growing broilers. It can be considered acceptable when compared to the FCR for rearing male layer-type chickens reported in previous studies [28,29].

The carcass composition of the male layer-type chickens in this study showed that thighs had the highest proportion of all parts, followed by the breast and back. The content of the thighs is approximately 10% higher than that of the breast. This is in agreement with our previous research on layer cockerels [11], slow-growing chickens [30], and dualpurpose crosses [31]. A higher percentage of thighs in male layer-type chickens has also been reported in other studies regardless of age [16,32]. In contrast, in fast-growing broilers, both conventionally and organically reared, the yield of breast was reported to be 10% higher than that of thighs (37% vs. 27%) [33]. The body composition of the layer strains makes them less attractive to consumers compared to fast-growing broilers at the same slaughter weight. On the other hand, when slaughtered at an early age, male layer-type chickens might be used as "coquelets", as shown by Koenig et al. [26] in a study with two different layer genotypes (medium–heavy and light). It was found that this was economically feasible; however, the carcass composition differed between the genotypes in favor of the heavier strain.

The economic analysis of the production of male layer-type chickens revealed that the total costs for producing 1 kg live weight were lower for birds reared until 9 weeks of age. A detailed breakdown of the individual parameters revealed that rearing the chickens to older age was associated with higher labor and rental costs. The latter was a direct consequence of the reduced stocking density, whereas the increase in labor costs was due to the negligible difference in the activities for rearing to 6 weeks (for total weight of 304.698 kg) and 9 weeks of age (for total weight 237.44 kg). Higher costs were compensated by the rest of the parameters, the preparation of the premises, the price of a one-day-old chick, and the costs for medication, having the highest share. The first two costs were made once at the beginning of the experiment, and as the weight of the chickens increased, it was obvious that their proportion decreased. On the other hand, medication costs included decontamination of the premises (constant parameter) and vaccine costs, which were used mainly until the chickens were 6 weeks old.

Again, when evaluating the efficiency of the production of 1 kg ready product, we found that the total costs were lower for the 9-week-old birds. This was mainly due to two factors. First was the lower cost for slaughter. At the same price for slaughtering a chicken (as it is in our case), it is clear that it is economically more efficient to slaughter chickens of higher weight. The second factor is the higher dressing percentage of 9-week-old chickens (65.18%) compared to that of 6-week-old chickens (57.58%). The dressing percentage obtained by us is slightly higher than the one presented in the study of Mueller et al. [15], where the value is 57.28% of Lohmann Brown Classic chickens slaughtered at the age of 67 days. It could be suggested that lower costs can also be obtained by slaughtering

at an older age, but not more than 12 weeks, because it has been found that after this age, the growth rates for this breed decrease significantly, and the economic costs are unjustified.

The pH of meat is an important factor in its quality. It reflects the rate of the postmortem glycolysis, and variations in the pH values affect the ability of meat to retain water, as well as its color. The highest pH values in chickens slaughtered at 6 weeks were observed in meat stored for 60 days, followed by a decrease, while in 9-week-old birds, the values of pH peaked in the meat stored for 120 days. The trends of the changes in the pH values of the meat in this study were consistent with the changes in the free amino groups over the course of the frozen storage. The increase in the pH values during storage corresponded with the increased content of the free amino groups, and point towards the association of pH, with proteolysis occurring in meat [19]. So far, the results reporting the effect of the frozen storage on meat pH remain contradictory. Our results, showing an increase in pH, coincide with those for goose meat when subjected to frozen storage [34]. Other studies, however, report no change [35,36] or decrease [37,38] in pH during frozen storage. Moreover, the different patterns of the change in pH in the age groups regardless of the type of meat suggest an interaction between storage and age. Since we refer to 6and 9-week-old chickens as a source of two different products, we have not examined the effect of age, and in this study, both age groups are separated in regard to meat quality characteristics. However, in our previous study, when comparing the meat quality traits of layer cockerels slaughtered at 5 and 9 weeks of age, we found significant differences in the pH of breast and thigh meat between the two ages [24].

During the course of frozen storage, we observed a significant increase in L\* values in the breast of 6- and 9-week-old birds stored for 60 days, as well as in the thigh of 9-week-old male chickens. The thighs of the chickens slaughtered at 6 weeks old also displayed this trend. The increased values of the L\* in the breast and thighs of the 9-week-old chickens measured after 60 days of frozen storage corresponded to the lower degree of redness. On the other hand, the dynamics of the changes in b\* showed similar patterns in birds at 6 weeks of age in the breast and thigh, with increased values in the samples stored for 60 days. The lower values of a\* and increased values of b\* suggest denaturation of the myoglobin, particularly during 60 days of frozen storage. The effect of the frozen storage on the color parameters has been reported to be different among studies. Constant increased values of L\* and b\* during 18 months of frozen storage were reported for pork [39]; however, for a\*, the authors observed an increase from 3 to 12 months and then a decrease until 18 months of frozen storage. In chicken meat, Lee at al. [40] observed a gradual decrease in L\* until 6 months of storage and an increase afterwards until the 8th month. The authors reported a similar trend for a\* and a constant decrease in b\*. Our results coincide with those of Agustynska-Prejsnar et al. [41], that demonstrated a decrease in L<sup>\*</sup> in chicken meat during long periods of frozen storage compared to shorter ones. On the other hand, Ali et al. [42] reported an increase in L\* in chicken meat for 6 months of frozen storage, which is in line with our findings. The authors also observed increased b\* values for this period, that we only observed in the meat of 6-week-old chickens, again suggesting an interaction between storage with age.

The changes in the content of the free amino groups reflect the accumulation of the end products of proteolysis. The initial and final content of free amino groups in this study were higher in the breast than in the thigh meat. Vassilev et al. [19] also reported a higher content of free amino groups in breast meat compared to leg when refrigerated for 7 days. These observations indicate that the trends of proteolysis are dependent on the different muscle fiber characteristics [43]. It could be seen that the concentration of the free amino groups showed different trends during the frozen storage process in the meat of the chickens slaughtered at 6 and 9 weeks of age. The highest levels of amino groups were measured after 60 days of storage in the breasts and thighs of younger chicks, while in the meat of 9-week-old chickens, the value of this trait reached maximum at day 120 of storage. It could be suggested that proteolytic processes have different intensities depending on the age of the birds due to the different activity of the enzyme systems [44].

In this study, the peroxide value and TBARSs were selected to represent the oxidative processes that occur in the chicken meat during storage. The peroxide value is a measure of the formation of peroxides that are primary products of lipid oxidative processes and determine their extent at the initial stages. We observed considerable augmentation of the content of the peroxides for 60 days of frozen storage, regardless of the type of the meat or the age of the birds, followed by a reduction in the peroxide values. The decrease in peroxide formation during frozen storage is not unusual. In line with our results, a reduction in PV was reported after frozen storage in breast and leg meat, respectively, after 2 and 3 months [45]. Another study on chicken meat also reported a dramatic decrease in peroxide formation at 6 months of frozen storage regardless of the presence of antioxidants [46]. As stated by Yi et al. [47], the ability to form peroxides is relevant to meat quality since it precedes the formation of an off-flavor as well as the crosslinking of proteins. Furthermore, it was showed [48] that even in small concentrations, lipid peroxides can exert toxic effects on the cell. Lipid peroxides are unstable and susceptible to decomposition [49] and the formation of other products, such as alcohols, ketones, and aldehydes. This could explain the decrease in their content during frozen storage that we observed in this study, corresponding to the dramatic increase in the TBARSs that we measured in the meat stored for 120 days. We observed a constant increase in the TBARS values over the course of storage. This is in line with the results obtained in goose meat stored for 365 days [50], poultry meat frozen for 3 months [51], as well as in the meat of other species—lamb [52], horse [53], and pork [39]. The major TBA reactive substance is malondialdehyde (MDA)—an important secondary product of the oxidation of polyunsaturated fatty acids. It is associated with rancid odors in meat even at low concentrations. According to some studies [54,55], MDA, in an amount of 2.5 mg/kg meat, has been set as a limit below which no rancidity was detected. The TBARSs content presented in this study did not exceed the mentioned threshold; hence, frozen storage was not associated with negative changes in the flavor of the meat of male layer-type chickens.

#### 5. Conclusions

This study aimed to demonstrate the potential of male layer-type chickens to produce meat as an option to avoid their culling after hatching and to further investigate the influence of frozen storage on the quality of their meat derived at two ages of slaughter. Over the course of frozen storage, no considerable deviations in the quality parameters indicating deterioration of the meat were observed. Yet, it had significant effect on pH, color, proteolysis, and lipid oxidative processes in the breast and thigh meat, and in some of these parameters the changes were different according to the age of slaughter. Further, our results showed that the application of some rearing practices such as fragmentation of stocking density, as well as finding a suitable age for slaughter, are very important for obtaining good economic results. We found that rearing male layer-type chickens until an older age (9 weeks) had lower costs and higher economic efficiency.

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**Institutional Review Board Statement:** The experimental protocol used in this study was designed in compliance with the guidelines of the European and Bulgarian legislation regarding the protection of animals used for experimental and other scientific purposes (Directive 2010/63; EC, 2010–put into law in Bulgaria with Regulation 20/2012). The protocol was based on the permit for use of animals

in experiments No. 277 of the Bulgarian Food Safety Agency (Statement No. 193 of the Bulgarian Animal Ethics Committee, prot.No. 18/02.07.2020).

**Data Availability Statement:** The data presented in this study are available upon request from the corresponding author.

Conflicts of Interest: The authors declare no conflicts of interest.

# References

- 1. Pereira, P.M.C.C.; Vicente, A.F.R.B. Meat nutritional composition and nutritive role in the human diet. *Meat Sci.* 2013, 93, 586–592. [CrossRef]
- Papp, R.E.; Hasenegger, V.; Ekmekcioglu, C.; Schwingshackl, L. Association of poultry consumption with cardiovascular diseases and all-cause mortality: A systematic review and dose response meta-analysis of prospective cohort studies. *Crit. Rev. Food Sci. Nutr.* 2023, 63, 2366–2387. [CrossRef]
- Bonpoor, J.; Petermann-Rocha, F.; Parra-Soto, S.; Pell, J.P.; Gray, S.R.; Celis-Morales, C.; Ho, F.K. Types of diet, obesity, and incident type 2 diabetes: Findings from the UK Biobank prospective cohort study. *Diabetes Obes. Metab.* 2022, 24, 1351–1359. [CrossRef] [PubMed]
- 4. Ibsen, D.B.; Warberg, C.K.; Würtz, A.M.L.; Overvad, K.; Dahm, C.C. Substitution of red meat with poultry or fish and risk of type 2 diabetes: A Danish cohort study. *Eur. J. Nutr.* **2019**, *58*, 2705–2712. [CrossRef]
- 5. Daniel, C.R.; Cross, A.J.; Graubard, B.I.; Hollenbeck, A.R.; Park, Y.; Sinha, R. Prospective investigation of poultry and fish intake in relation to cancer risk. *Cancer Prev. Res.* 2011, *4*, 1903–1911. [CrossRef]
- 6. Lo, J.J.; Park, Y.-M.M.; Sinha, R.; Sandler, D.P. Association between meat consumption and risk of breast cancer: Findings from the Sister Study. *Int. J. Cancer* 2020, 146, 2156–2165. [CrossRef] [PubMed]
- 7. FAOSTAT. Available online: https://www.fao.org/faostat/en/#data/QCL (accessed on 20 November 2023).
- 8. Valenta, J.; Chodová, D.; Tůmová, E.; Ketta, M. Carcass characteristics and breast meat quality in fast-, medium- and slow-growing chickens. *Czech J. Anim. Sci.* 2022, 67, 286–294. [CrossRef]
- 9. Sirri, F.; Castellini, C.; Bianchi, M.; Petracci, M.; Meluzzi, A.; Franchini, A. Effect of fast-, medium- and slow-growing strains on meat quality of chickens reared under the organic farming method. *Animal* **2011**, *5*, 312–319. [CrossRef]
- 10. Popova, T.; Petkov, E.; Ignatova, M.; Vlahova-Vangelova, D.; Balev, D.; Dragoev, S.; Kolev, N. Male layer-type chickens—An alternative source for high quality poultry meat: A review on the carcass composition, sensory characteristics and nutritional profile. *Braz. J. Poult. Sci.* **2022**, *24*, 1–10. [CrossRef]
- 11. Popova, T.; Petkov, E.; Ignatova, M.; Dragoev, S.; Vlahova-Vangelova, D.; Balev, D.; Kolev, N. Growth Performance, carcass composition and tenderness of meat in male layer-type chickens slaughtered at different age. *C. R. Acad. Bulg. Sci.* **2023**, *76*, 156–164.
- 12. Angelovičová, M.; Angelovič, M.; Čapla, J.; Zajác, P.; Folvarčíková, P.; Čurlej, J. The effect of oregano essential oil on chicken meat lipid oxidation and peroxidation. *Potravin. S. J. Food Sci.* **2021**, *15*, 1056–1068. [CrossRef]
- 13. Lichovníková, M.; Jandásek, J.; Jùzl, M.; Draèková, E. The meat quality of layer males from free range in comparison with fast growing chickens. *Czech J. Anim. Sci.* **2009**, *54*, 490–497. [CrossRef]
- 14. Mueller, S.; Kreuzer, M.; Siegrist, M.; Mannale, K.; Messikommer, R.E.; Gangnat, I.D.M. Carcass and meat quality of dual-purpose chickens (Lohmann Dual, Belgian Malines, Schweizerhuhn) in comparison to broiler and layer chicken types. *Poult. Sci.* **2018**, *97*, 3325–3336. [CrossRef]
- 15. Mueller, S.; Taddei, L.; Albiker, D.; Kreuzer, M.; Siegrist, M.; Messikommer, R.E.; Gangnat, I.D.M. Growth, carcass, and meat quality of 2 dual-purpose chickens and a layer hybrid grown for 67 or 84 D compared with slow-growing broilers. *J. Appl. Poult. Res.* **2020**, *29*, 185–196. [CrossRef]
- Choo, Y.K.; Oh, S.T.; Lee, K.W.; Kang, C.W.; Kim, H.W.; Kim, C.J.; Kim, E.J.; Kim, H.S.; An, B.K. The growth performance, carcass characteristics, and meat quality of egg-type male growing chicken and White-Mini Broiler in comparison with commercial broiler (Ross 308). *Korean J. Food Sci. Anim. Resour.* 2014, 34, 622–629. [CrossRef]
- 17. Evaris, E.F.; Sarmiento-Franco, L.; Sandoval-Castro, C.A. Meat and bone quality of slow-growing male chickens raised with outdoor access in tropical climate. *J. Food Compos. Anal.* **2021**, *98*, 103802. [CrossRef]
- 18. Khan, A. Extraction and fractionation of proteins in fresh chicken muscle. J. Food Sci. 1962, 27, 430–434. [CrossRef]
- 19. Vassilev, K.; Ivanov, G.; Balev, D.; Dobrev, G. Protein changes of chicken light and dark muscles during chilled storage. *J. EcoAgriTourism* 2012, *8*, 263–268.
- 20. Bligh, E.G.; Dyer, W.J. A rapid method of total lipid extraction and purification. *Can. J. Biochem. Physiol.* **1959**, 37, 911–917. [CrossRef]
- 21. Shantha, N.C.; Decker, E.A. Rapid, sensitive, iron-based spectrophotometric methods for determination of peroxide values of food lipids. *J. AOAC Int.* **1994**, *7*, 421–424. [CrossRef]
- 22. Botsoglou, N.A.; Fletouris, D.J.; Papageorgiou, G.E.; Vassilopoulos, V.N.; Mantis, A.J.; Trakatellis, A.G. Rapid, sensitive, and specific thiobarbituric acid method for measuring lipid peroxidation in animal tissue, food, and feedstuff samples. *J. Agric. Food Chem.* **1994**, *42*, 1931–1937. [CrossRef]
- 23. *JMP*, version 7; SAS Institute Inc.: Cary, NC, USA, 2007.

- 24. Popova, T.; Petkov, E.; Ignatova, M.; Vlahova-Vangelova, D.; Balev, D.; Dragoev, S.; Kolev, N.; Dimov, K. Meat quality of male layer-type chickens slaughtered at different ages. *Agriculture* **2023**, *13*, 624. [CrossRef]
- Habig, C.; Bayerbach, M.; Kember, N. Comparative analyses of layer males, dual purpose males and mixed sex broilers kept for fattening purposes regarding their floor space covering, weight-gain and several animal health traits. *Arc. Geflügelkd.* 2016, *80*, 1–10.
- Koenig, M.; Hahn, G.; Damme, K.; Schmutz, M. Utilization of laying-type cockerels as coquelets: Influence of genotype and diet characteristics on growth performance and carcass composition. *Arc. Geflügelkd.* 2012, 76, 197–202.
- 27. Putra, W.P.B.; Riaz, R.; Gunawan, A.A.; Orman, A. Comparison of growth curve in male layer chickens. *J. Res. Vet. Med.* 2021, 40, 49–53. [CrossRef]
- Evaris, F.E.; Sarmiento, F.L.; Sandoval, C.C.; Segura, C.J.; Caamal, M.J.A. Male Layer chicken's response to dietary *Moringa oleifera* meal in a tropical climate. *Animals* 2022, 12, 1843. [CrossRef] [PubMed]
- De Silva, P.; Wickramasinghe, Y.; Kalubowila, D. Growth performance and carcass quality of layer type cockerels and broiler chicken. *Iran. J. Appl. Anim. Sci.* 2016, 6, 429–433.
- Petkov, E.; Popova, T.; Ignatova, M. Carcass and meat composition in f1 crosses of two lines of slow-growing chickens reared in conventional or alternative system with access to pasture. *Int. J. Innov. Approaches Agric. Res.* 2018, 2, 359–374. [CrossRef]
- Petkov, E.; Popova, T.; Ignatova, M.; Sharkova, V.; Dimov, K. Development of dual-purpose cross for meat and egg production I. Growth performance and carcass composition of the crossbred chickens in comparison to the parent lines. *Arch. Zootech.* 2022, 25, 119–129. [CrossRef]
- 32. Murawska, D.; Bochno, R. Comparison of the slaughter quality of Layer-type cockerels and broiler chicken. *J. Poult. Sci.* 2007, 44, 105–110. [CrossRef]
- 33. Sarica, M.; Yamak, U.S.; Boz, M.A.; Erensoy, K.; Cilavdaroglu, E.; Noubandiguim, M. Performance of fast, medium and slow growing broilers in indoor and free-range production systems. *S. Afr. J. Anim. Sci.* **2019**, *49*, 1127–1138. [CrossRef]
- Wereńska, M.; Okruszek, A. Impact of frozen storage on some functional properties and sensory evaluation of goose meat. *Poult. Sci.* 2023, 102, 102894. [CrossRef] [PubMed]
- Śmiecińska, K.; Hnatyk, N.; Daszkiewicz, T.; Kubiak, D.; Matusevičius, P. The effect of frozen storage on the quality of vacuumpackaged Turkey meat. Vet. Zootech. 2015, 71, 61–66.
- Kluth, I.K.; Teuteberg, V.; Ploetz, M.; Krischek, C. Effects of freezing temperatures and storage times on the quality and safety of raw turkey meat and sausage products. *Poult. Sci.* 2021, 100, 101305. [CrossRef]
- Wei, R.; Wang, P.; Han, M.; Chen, T.; Xu, X.; Zhou, G. Effect of freezing on electrical properties and quality of thawed chicken breast meat. *Asian-Australas J. Anim. Sci.* 2017, 30, 569–575. [CrossRef]
- 38. Saewa, S.; Khidhir, Z.; Al Bayati, M. The impact of storage duration and conditions on the formation of biogenic amines and microbial content in poultry meat. *Iraqi J. Vet. Sci.* 2021, *35*, 183–188. [CrossRef]
- Medić, H.; Kušec, I.D.; Pleadin, J.; Kozačinski, L.; Njari, B.; Hengl, B.; Kušec, G. The impact of frozen storage duration on physical, chemical and microbiological properties of pork. *Meat Sci.* 2018, 140, 119–127. [CrossRef] [PubMed]
- 40. Lee, Y.S.; Saha, A.; Xiong, R.; Owens, C.M.; Meullenet, J.F. Changes in broiler breast fillet tenderness water-holding capacity and colour attributes during long-term frozen storage. *J. Food Sci.* 2008, 73, 162–168. [CrossRef]
- Augustyńska-Prejsnar, A.; Hanus, P.; Sokołowicz, Z.; Kačániová, M. Assessment of technological characteristics and microbiological quality of marinated turkey meat with the use of dairy products and lemon juice. *Anim. Biosci.* 2021, 34, 2003–2011. [CrossRef]
- 42. Ali, S.; Rajput, N.; Li, C.; Zhang, W.; Zhou, G. Effect of Freeze-thaw cycles on lipid oxidation and myowater in broiler chickens. *Braz. J. Poult. Sci.* 2016, *18*, 35–40. [CrossRef]
- 43. Cheng, H.; Song, S.; Park, T.S.; Kim, G.D. Proteolysis and changes in meat quality of chicken pectoralis major and iliotibialis muscles in relation to muscle fiber type distribution. *Poult. Sci.* 2022, 108, 102185. [CrossRef]
- 44. Teye, G.A.; Okutu, I. Effect of ageing under tropical conditions on the eating qualities of beef. *Afr. J. Food Agric. Nutr. Dev.* **2010**, *9*, 1901–1913. [CrossRef]
- 45. Soyer, A.; Özalp, B.; Dalmış, Ü.; Bilgin, V. Effects of freezing temperature and duration of frozen storage on lipid and protein oxidation in chicken meat. *Food Chem.* **2010**, *120*, 1025–1030. [CrossRef]
- 46. Lai, M.M.C.; Zhang, H.A.; Kitts, D.D. Ginseng prong added to broiler diets reduces lipid peroxidation in refrigerated and frozen stored poultry meats. *Molecules* **2021**, *26*, 4033. [CrossRef] [PubMed]
- Yi, G.; Haug, A.; Nyquist, N.F.; Egelandsdal, B. Hydroperoxide formation in different lean meats. *Food Chem.* 2013, 141, 2656–2665. [CrossRef] [PubMed]
- 48. Angeli, J.P.F.; Garcia, C.C.M.; Sena, F.; Freitas, F.P.; Miyamoto, S.; Medeiros, M.H.G.; Di Mascio, P. Lipid hydroperoxide-induced and hemoglobin-enhanced oxidative damage to colon cancer cells. *Free Radic. Biol. Med.* **2011**, *51*, 503–515. [CrossRef] [PubMed]
- Talbot, G. The stability and shelf life of fats and oils. In *The Stability and Shelf Life of Food*; Subramaniam, P., Ed.; Elsevier: Amsterdam, The Netherlands, 2016; pp. 461–503.
- 50. Wereńska, M.; Okruszek, A.; Haraf, G.; Wołoszyn, J.; Goluch, Z. Impact of frozen storage on oxidation changes of some components in goose meat. *Poult Sci.* 2022, 101, 101517. [CrossRef] [PubMed]

- 51. Konieczka, P.; Czauderna, M.; Rozbicka-Wieczorek, A.; Smulikowska, S. The effect of dietary fat, vitamin E and selenium concentrations on the fatty acid profile and oxidative stability of frozen stored broiler meat. *J. Anim. Feed Sci.* 2015, 24, 244–251. [CrossRef]
- 52. Pinheiro, R.S.B.; Francisco, C.L.; Lino, D.M.; Borba, H. Meat quality of Santa Inês lamb chilled-then-frozen storage up to 12 months. *Meat Sci.* 2019, 148, 72–78. [CrossRef]
- 53. Seong, P.N.; Seo, H.W.; Kim, J.H.; Kang, G.H.; Cho, S.H.; Chae, H.S.; Park, B.Y.; Van Ba, H. Assessment of frozen storage duration effect on quality characteristics of various horse muscles. *Asian-Australas J. Anim. Sci.* 2017, 30, 1756–1763. [CrossRef]
- 54. Campo, M.M.; Nute, G.R.; Hughes, S.I.; Enser, M.; Wood, J.D.; Richardson, R.I. Flavour perception of oxidation in beef. *Meat Sci.* **2006**, 72, 303–311. [CrossRef] [PubMed]
- Zhang, Y.; Holman, B.W.B.; Ponnampalam, E.N.; Kerr, M.G.; Bailes, K.L.; Kilgannon, A.K.; Collins, D.; Hopkins, D.L. Understanding beef flavour and overall liking traits using two different methods for determination of thiobarbituric acid reactive substance (TBARS). *Meat Sci.* 2019, 149, 114–119. [CrossRef] [PubMed]

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