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Bioactive Compounds in Breast Meat of Broiler Chickens Fed with Black Soldier Fly Wholemeal

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Abstract

This study investigated the effects of dietary supplementation with Black Soldier Fly (BSF) wholemeal on the content of bioactive compounds in broiler chicken breast meat. The experiment involved 45 male Ross 308 broiler chickens randomly assigned to three dietary groups: control diet, control diet supplemented with 5% (HI5), or 10% (HI10) black soldier fly (BSF) wholemeal. The diets were administered for 35 days. The study found that higher levels of BSF wholemeal meal inclusion significantly improved creatine and carnosine levels, with increases of 22% and 26%, respectively, in the HI10 group compared to the control group. In addition, HI supplementation improved the fatty acid profile, significantly increasing the levels of EPA, DHA, and conjugated linoleic acid (CLA), while reducing the total PUFA and ALA levels. Antioxidant activity, measured using the FRAP and ABTS assays, was also significantly higher in the BSF-fed groups, particularly in the HI10 group. These results suggest that BSF wholemeal flour can improve the functional and nutritional qualities of chicken meat, thereby enhancing its potential as a sustainable ingredient in poultry diets.

Keywords: antioxidant activity; bio-peptides; chicken meat; functional fatty acids; *Hermetia illucens* wholemeal flour

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1. Introduction

In recent years, the poultry industry has faced increasing challenges related to sustainability, efficiency, and animal welfare. Researchers and industry professionals have explored new food sources, among them, including black soldier fly larvae (BSFL), which contribute to nutrient reuse by converting organic materials of both animal and plant origin into biomass during the decomposition phase [1,2]. BSFL is a highly sustainable nutritional resource, both for the environment and for conventional feeds used for poultry feeding. Its nutritional value is comparable to that of conventional feeds used in feed formulations [3,4]. Whole BSFL contains up to 59% crude protein (CP) and up to 34% lipid in dry matter, which are attractive characteristics for functional poultry nutrition [5]. An important parameter is the quality of fats characterized by high concentrations of saturated fatty acids (SFA,

such as lauric, myristic, palmitic, and monounsaturated oleic acids [2]. It also has an amino acid profile composed of a high concentration of essential AA, such as histidine, lysine, leucine, and arginine, which are important in the formulation of diets that must ensure an adequate supply of all essential amino acids in each growth phase [6]. In the literature, there are few reports on the effect of the addition of whole BSFL powder to broiler chicken diets on the nutritional quality and functional capacity of meat [7]. The scientific community has contributed significantly to the understanding of the effects of larval integration in the diet from both nutritional and physiological perspectives [8]. However, there is still little scientific evidence regarding the bioactive component content in chicken meat fed these biomasses. Meat and meat products fall into the category of functional foods because they contain numerous compounds that play a functional biological role when consumed [9]. Meat is a good source of nitrogenous bioactive substances, such as creatine, carnosine, L-carnitine, glutathione, and taurine, as well as lipid nature, such as PUFAω3, ALA, C18:1cis9, and conjugated linoleic acid (CLA) [9]. Bioactive peptides are organic compounds consisting of amino acids that are inactive within the native protein and are released only after hydrolysis, cooking, or fermentation processes [10]. Many functional peptides that exhibit antioxidant activity have been detected and described in meat [9]. Carnosine is a dipeptide synthesized from β-alanine and L-histidine and is mainly present in the skeletal muscle and brain of vertebrates [11]. Its physiological functions are multiple: anti-aging, antioxidant, anti-glycation, and anti-inflammatory [12]. Gulcin and Alwasel [13] demonstrated the antioxidant action of carnosine through its ability to chelate metals and eliminate free radicals, preventing and/or limiting lipid oxidation. L-carnitine (γ-trimethylamino-βhydroxybutyric acid) is a bioactive component synthesized from lysine and methionine [14]. Its presence is fundamental in the metabolism of fats; in fact, it transports fatty acids to the mitochondria to generate energy. However, deficiency causes disturbances in the metabolism of fatty acids [15]. Furthermore, L-carnitine has been recognized as having an antioxidant action capable of increasing the antioxidant activity of enzymes, although its mechanism of action is not yet clear. However, it is known that it follows two lines of action: indirect action by enhancing the cellular energy state and direct action by maintaining mitochondrial integrity [14]. Although L-carnitine and carnosine can be synthesized in humans, their dietary intake may be beneficial for maintaining or improving health [9]. Important fatty acids like ole-ic acid, linolenic acid, and ω-3 series acids like eicosapentaenoic acid (EPA, C20:5 ω-3) and do-cosahexaenoic acid (DHA, C22:6 ω-3), as well as the class of conjugated fatty acids called conjugated linoleic acid isomers have been found to have a high bioactive potential [16]. CLAs commonly possess antitumor, antiadipogenic, anti-inflammatory, and immunomodulatory properties. However, the mechanisms underlying the bioactivity of these compounds have not yet been fully elucidated [17]. Nonetheless, current data suggest that these fatty acids may influence the expression of several oncogenes, cell cycle regulators, and energy metabolism-related genes. The type of diet influences the content of bioactive compounds in chicken meat [18]. There is extensive scientific evidence on the effects of nutrition on fatty acid and CLA content [9] and dipeptide content [19]. The importance of feed improvement in poultry feeding must consider the rationalization and optimal use of nutrients in order to benefit animal welfare and improve production performance and meat quality [20]. Therefore, the aim of the present study was to evaluate the effect of the level of dietary in order to assess the impact of the amount of black soldier fly meal (*Hermetia illucens*) supplementation on the compositional properties of meat, the current study focused on the content of specific bioactive compounds in the breast meat of broiler chickens.

2. Materials and Methods

All the birds and the experimental protocols in this study were approved by "Organismo Preposto al Benessere Animale (OpBA)" of University of Basilicata (Potenza, Italy)—Protocol code: OpBA 13_2024_UNIBAS

2.1. Birds, Diets, and Experimental Design

The study was conducted on 45 male broiler chickens (Ross 308), raised on the farm "Quadrifoglio—Aziende agricole", owned by Antony Palumbo (Picerno-Potenza, Basilicata, Italy). The birds were randomly divided into three groups: the control group (n = 15 birds; C) was fed a basal diet, the second group (n = 15 birds; HI5) was fed a basal diet plus 5% BSFL wholemeal flour; the third group (n = 15 birds; HI10) was fed a basal diet plus 10% BSFL wholemeal flour. The basal diet was designed to meet the age-specific nutritional requirements of broiler chickens. Each group of birds was divided into three replicates, with five birds per replicate, equipped with shared feeders and drinkers with teats. The litter in each pen consisted of approximately 8 cm of rice husks and wood shavings. Food and water were provided ad libitum during the experiment. A temperature of 32–34 °C was maintained for the first three days and then reduced to 22 °C at a rate of 2–3 °C per week. The light/dark cycle (14/10 h) and ventilation system of the house were automatically adjusted according to the growth stage of the chicks. The three groups of broilers (n = 45) were fed the respective diets (C, HI5, and HI10) for a period of two weeks until 35 days of age. The commercial basal diet included a grower ration (0–21 days) and a finishing ration (22–35 days), as shown in Table 1. All broiler chickens were slaughtered at 35 days of age, at a mean slaughter weight of 1.75 ± 0.24 kg. The breast (Pectoralis Major Muscle) was removed from each bird, vacuum-packed, and stored at −20 °C until analysis. Wholemeal flour BSFL was supplied by Xflies s.r.l. (Potenza, Italy). After hatching, the newly hatched larvae were reared on the standard Gainesville diet, supplied by the animal feed factory Mangimi Losasso s.r.l.—Balvano (Potenza, Italy), composed of 20% corn, 30% alfalfa meal, and 50% wheat bran (Scala et al. 2020) at 82% humidity, for 5 days, as described by Scala et al. [21]. On the sixth day, BSFL were transferred to plastic boxes containing a standard diet. The larvae were cultured at 27 ± 1 °C and 70 ± 2% relative humidity. At the end of the feeding period, the larvae were separated from the residual material (larval excreta) using an industrial sieving machine (mesh size 4 mm) (Guangzhou Flysource Biotechnology Co., Ltd., Guangzhou, China), which removed most of the adherent material. They were stored at −20 °C for 24h and then dehydrated using an industrial microwave (MAX Industrial Microwave, Yantai, China) (as suggested by IPIFF—Guide on Good Hygiene Practices). To obtain fine particles and a homogeneous powder, the dried larvae were ground using a food mill (Tom Press Italia, Mantova, Italy). The ingredients and chemical composition (%) of the broiler grower and finisher diets are presented in Table 1. The fatty acid profile of the Black Soldier Fly (BSF) meal supplemented to the standard diet is presented in Table 2. Furthermore, in a previous study by Scieuzo et al. (2022) [1], the average amino acid profile and chitin content of the same BSFL flour used in this study were reported.

Table 1. Ingredients and chemical composition (%) of the growing and finishing diets of broiler chickens.

Ingredients (%)	Grower Ration (0–21 Days)	Finisher Ration (22–35 Days)	BSFL (22–35 Days)
Corn	61.00	65.45	
Soybean meal	26.89	29.95	
Wheat bran	10.05	2.75	
Dicalcium phosphate	1.45	1.30	
Salt	0.30	0.30	
DL-methionine	0.11	0.05	
Vitamin Premix *	0.10	0.10	
Mineral Premix **	0.10	0.10	
Minerals, %			
Calcium	1.60	0.85	1.47
Phosphorus	0.60	0.48	0.95
Magnesium	0.25	0.20	0.54
Chemical composition, %DM			
Crude Protein	21.20	18.89	44.74
Crude Fat	5.60	4.5	19.80
Crude Fiber	2.85	3.06	22.70
Crude ash	5.5	5.0	12.76

* Composition of the vitamin premix (per kg of diet): vitamin A = 5500 IU; vitamin D3 = 1100 IU; vitamin E = 10 IU; riboflavin = 4.4 mg; vitamin B12 = 12 mg; nicotinic acid = 44 mg; menadione = 1.1 mg; biotin = 0.11 mg; thiamine = 2.2 mg; ethoxyquin = 125 mg. ** Composition of the mineral premix (per kg of diet): Mn = 120 mg; Zn = 100 mg; Fe = 60 mg; Cu = 10 mg; Se = 0.17 mg; I = 0.46 mg; Ca minimum = 150 mg; Ca maximum = 180 mg.

Table 2. Fatty acid profile of Black Soldier Fly (BSF) meal fed a standard diet. The values are expressed as percentages of the total lipid fraction.

Fatty Acids	Standard Diet
C12:0	37.63
C14:0	8.42
C14:1	0.29
C15:0	0.21
C16:0	16.74
C16:1	0.40
C17:0	3.64
C17:1	0.30
C18:1 cis 6	0.23
C18:0	3.02
C18:1 trans 9	-
C18:1 trans 11	0.44
C18:1 cis 9	12.31
C18:1 cis 10	-
C18:1 cis 11	0.05
C18:2 cis n6	8.38
C20:0	0.14
C20:1	0.11
C18:3 n3	1.86
C22:0	0.64
C22:1	0.15
$\sum$ SFA [1]	70.53
$\sum$ MUFA [2]	14.28
$\sum$ PUFA [3]	10.24
$\sum$ n-6	9.01
$\sum$ n-3	1.86

[1] SFA, saturated fatty acids; [2] MUFA, monounsaturated fatty acids; [3] PUFA, polyunsaturated fatty acids.

2.2. Meat Quality Measurements

The determination of total protein, total lipid, ash, dry matter, and pH on raw meat samples was in accordance with the AOAC [22]. Each determination was performed in triplicate. Meat tenderness was measured following the protocol of Khan et al. [23], using an Instron instrument (Instron 6800 Series, Norwood, MA, USA). Instead, the color indices, L* (brightness), a* (redness), and b* (yellowness), were analyzed with a MINOLTA Chromameter CR-300 (Minolta Camera Corp., Meter Division, Ramsey, NJ, USA) equipped with a D65 illuminant, the 10° O b server, and zero and blank calibration [24].

2.3. Quantification of Fatty Acids and CLA by Gas Chromatography

The lipid fraction of the chicken meat samples was extracted using the method described by Folch et al. [25]. Fatty acid methyl esters (FAME) were identified and quantified following the method described by Perna et al. [26]. A known volume of the extract was injected into a Varian 3400 gas chromatograph (Varian, Turin, Italy) equipped with a split-splitless injector and a TR-FAME capillary column (120 m × 0.25 mm i.d. × 0.25 µm film thickness (Thermo Fisher Scientific, Milan, Italy) and a flame ionization detector (FID). Galaxie™ (Varian, Inc., Walnut Creek, CA, USA) was used for chromatogram acquisition. Each peak acquired by the system was compared with the peaks identified in the pure methyl ester standard (SUPELCO, Bellefonte, PA, USA). Finally, the results were expressed as the percentage of total FAME.

2.4. Preparation of Meat for Antioxidant Activity

For the extraction of the meat sample, the method described by Grassi et al. [27] was followed. Briefly, approximately 2 g of chicken breast meat was added to 6 mL of distilled water and homogenized using a Polytron (PT-MR 2100, Kinematica AG, Littau, Lucerne, Switzerland) at 13,500 rpm for 15 s. In order to enhance extraction, the samples were placed in an ultrasonic bath (US) (Elma Transsonic 460/H, Singen, Germany) for 10 min at room temperature. Finally, the samples were centrifuged at 5000× *g* at 4 °C for 20 min, and the supernatant was filtered through a 0.45 µm cellulose acetate membrane filter (Sigma-Aldrich, Milan, Italy) and analyzed.

2.5. Ferric-Reducing Antioxidant Power (FRAP)

The ability to reduce ferric to ferrous ions was evaluated using the original method of Benzie and Strain [28], with some modifications. The FRAP reagent consisted of 300 mM acetate buffer (pH 3.6), 10 mM TPTZ in 40 mM HCl, and 20 mM $FeCl_3$ (10:1:1; *vol*/*vol*/*vol*). The reagent was maintained at 37 °C before analysis. In a cuvette, 100 µL of sample extracts and 2.9 mL of FRAP reagent were mixed, and the blank was prepared using distilled water instead of the sample. After 30 min of incubation, the absorbance was measured at 593 nm against a blank. A calibration curve was prepared using different concentrations of Trolox (2.2–0.25 µM), and the results were expressed as millimolar Trolox per gram of meat sample (mMTE/g meat). The analyses were performed in triplicate.

2.6. 2-2′-Azino-di-[3-Ethylbenzthiazoline Sulfonate] (ABTS) Radical Scavenging Activity

Radical scavenging activity was evaluated as suggested by Grassi et al. [27]. The ABTS radical cation (7.00 mM) was produced by reaction with potassium persulfate (2.45 mM) and incubated in the dark at room temperature for 12–16 h before use. An absorbance of 0.700 ± 0.020 at 734 nm was obtained using distilled water. The extract (100 µL) was added to 2.9 mL of ABTS solution and incubated at room temperature for 30 min. The percentage inhibition (I%) was evaluated using the following equation:

$$(I\% = A_b - A_s/A_b \times 100)$$

where A_b = absorbance of blank sample; A_s = absorbance of sample. The analyses were performed in triplicate.

2.7. Thiols Group

Thiols are organic compounds capable of neutralizing the action of reactive oxygen species (ROS) owing to the presence of the functional group—SH. The content of free thiols was evaluated using the method proposed by Grassi et al. [29]. The sample aliquot (250 µL) was added to 250 µL of reaction buffer and 50 µL of Ellman's reagent. The reaction buffer was composed of phosphate buffer (0.1 M with 1 mM EDTA, pH 8.0), and Ellman's reagent was composed of 3 mM DTNB in 0.1 M phosphate buffer containing 1 mM EDTA, pH 8.0. The incubation time was 15 min at 37 °C, and the change in absorbance was evaluated at 412 nm. The following equation was used:

$$C_{(\mu mol-SH/L)} = (\Delta A/\varepsilon \times b)$$

where ΔA is the change in absorbance in 15 min at 412 nm, ε is the molar extinction coefficient (14,150 M^{-1} cm^{-1}), and b is the optical path (cm). Thiols were expressed as nmol/mg of protein. The analyses were performed in triplicate.

2.8. Creatine and Carnosine Analysis

The protocol used for sample extraction was that of Simonetti et al. [30]. Samples were injected into a high-performance liquid chromatography (HPLC) system equipped with a Varian ProStar Model 210 pump, Rheodyne injector with 20 µL loop, Varian ProStar Model 325 UV-VIS detector, Varian ProStar Model 363 fluorescence detector, and Galaxie™ chromatography software (Varian, Inc., Walnut Creek, CA, USA, 2002–2005; 03-914947-00:Rev. 13). The silica column used was an Atlantis HILIC (4.6 × 150 mm, 3 µm) connected to an Atlantis HILIC Silica guard column (10 × 4.0 mm, 5 µm) (Thermo Fisher Scientific). The UV detection wavelength was set at 214 nm. Creatine and carnosine standards (Sigma-Aldrich; 0.01–0.30 and 0.01–0.50 mg/mL, respectively) were used for identification and quantification, and the results were expressed in mg/100 g meat.

2.9. Statistical Analysis

The data were statistically analyzed by one-way analysis of variance (ANOVA) with the general linear model (GLM) at a 95% confidence level ($p < 0.05$) in the SAS 1972 software (Version 7th ed):

$$y_{ij} = \mu + \alpha_i + \varepsilon_{ij}$$

where y_{ij} is the experimental observation, µ is the mean, α_i is the fixed effect of the dietary treatment (control, HI5, and HI10), and ε_{ijk} is the random error. For chemical analyses performed on chicken breast meat, each chicken was considered an experimental unit. Before setting the values, expressed in percentage terms, they were subjected to an arcsine transformation. The Tukey *post hoc* test was used to compare the means, and differences were considered significant ($p < 0.05$). The results are presented as mean ± standard deviation (µ ± SD).

3. Results and Discussion

3.1. Chemical Composition of Breast Meat

The centesimal composition of chicken breast from broilers fed the control diet and diets supplemented with 5 and 10% BLSF is presented in Table 3. Overall, the centesimal composition did not show any significant difference; in fact, the protein and fat content of the BLSF meal did not influence the chicken breast ($p > 0.05$). Our results are in line with

those of Popova et al. [31], who found differences only in chickens fed 5% wholemeal flour. Furthermore, no differences were detected in pH values and shear force ($p > 0.05$).

Table 3. pH, texture (Share Force, kg/g), color (L*, a*, b*), protein content (%), and lipid content (%) in chicken breast fed a control diet (CTRL) and supplemented with 5% and 10% BSF meal.

Parameters	C [1] μ ± SD	HI5 μ ± SD	HI10 μ ± SD
pH	5.80 ± 0.03	5.81 ± 0.04	5.81 ± 0.09
Share force$_{(kg/g)}$	2.67 ± 0.16	2.51 ± 0.14	2.59 ± 0.26
L*	52.03 ± 1.42	52.48 ± 1.76	52.85 ± 1.02
a*	2.87 ± 0.26	2.81 ± 0.14	2.82 ± 0.25
b*	3.46 ± 0.36	3.62 ± 0.36	3.75 ± 0.28
Total protein$_{(\%)}$	21.35 ± 0.61	20.99 ± 0.83	21.67 ± 0.53
Total lipid$_{(\%)}$	1.59 ± 0.10	1.49 ± 0.07	1.44 ± 0.14

[1] Values are expressed as mean ± standard deviation.

The effect of BLSF meal supplementation did not significantly affect the lightness (L*), redness (a*), and yellowness (b*) of chicken breast ($p > 0.05$). These values contrast with those found by Popova et al. [31] in the breast and thigh muscles of chickens fed whole or defatted BLSF meal. In particular, the authors observed a significant increase in the brightness index (L*) in the meat of chickens fed whole BSF meal. The authors associated these results with a low pH and low water holding capacity. In Table 4, the fatty acid profiles of chicken breast are reported separately for groups C, HI5, and HI10. Overall, diet significantly influenced the percentage content of individual fatty acids ($p < 0.05$).

The content of saturated fatty acids, such as C12:0, C14:0, and C17:0, in the breast meat of the HI5 and HI10 groups demonstrated that the supplementation of BLSF flour significantly increased the content of the studied fatty acids ($p < 0.05$), as shown in Table 4. Conversely, the change was not statistically significant for C16:0 and C18:0 content ($p > 0.05$). Among the polyunsaturated fatty acids, C18:1 cis 9, C18:2 cis n6, and C18:3 n3 decreased in proportion to increasing supplementation (HI10 > HI5; $p < 0.05$). Supplementation with BLSF flour significantly increased the total SFA content in breast meat ($p < 0.05$) and significantly decreased the total PUFA and MUFA content ($p < 0.05$). The addition of different fat sources to the diet of broiler chickens influences their fatty acid composition [32]. From our results, it is evident that BLSF flour has a high content of saturated fatty acids; in particular, the SFA class represents about 71% of the total FAME, of which about 38% is represented by C12:0 (Table 2). The latter content influences the percentage of SFA in the meat, which increases as a function of the level of dietary inclusion of the black soldier fly. Our results are supported by Skrivan et al. [33], who highlighted an increase in ileal fat digestibility following the administration of diets enriched with a fat source that increases ileal fat digestibility. Furthermore, the flour administered to the two treated groups, HI5 and HI10, was rich in lauric acid, as reported in Table 2 and in line with what was found by Popova et al. [31] and Schiavone et al. [34]. Many authors have associated the high C12:0 content with low pH values in meat; however, the results are contrasting [31,35]. In addition, a significant increase in SFA is associated with an increase in the atherogenic index (AI) and thrombogenic index (TI), indicators of lipid quality, which indicate the potential effects of lipids on the development of cardiovascular diseases. Some studies have investigated the potential effects of lauric acid on broiler diets, showing an improvement in feed conversion efficiency and an antimicrobial effect of medium-chain fatty acids in the intestine [35]. Furthermore, C12:0 significantly improves the microbiological safety of chicken breast meat by reducing pathogens such as *Campylobacter* [35] and *Salmonella* [36] and enriches the nutritional quality of meat by improving its fatty acid profile, particularly omega-3 fatty acids [37].

Table 4. Fatty acid composition (% of total lipid fraction) in chicken breast fed a control diet (C) and supplemented with 5% and 10% BSF meal.

Fatty Acids	% of Total FAME		
	C [1] μ ± SD	HI5 μ ± SD	HI10 μ ± SD
C12:0	0.23 ± 0.05 [b]	3.11 ± 0.58 [a]	4.54 ± 0.87 [a]
C13:0	0 [b]	0.02 ± 0.01 [a]	0.03 ± 0.001 [a]
C14:0	0.67 ± 0.02 [c]	1.91 ± 0.27 [b]	2.60 ± 0.20 [a]
C14:1	0.10 ± 0.02 [a]	0.05 ± 0.03 [b]	0.06 ± 0.02 [b]
C15:0	0.01 ± 0.01 [b]	0.12 ± 0.01 [a]	0.11 ± 0.02 [a]
C16:0	21.66 ± 1.32	20.84 ± 0.77	21.03 ± 1.50
C16:1	1.10 ± 0.14	1.34 ± 0.43	1.56 ± 0.67
C17:0	0.18 ± 0.01 [b]	0.23 ± 0.02 [a]	0.23 ± 0.03 [a]
C18:1 cis 6	0.03 ± 0.03 [b]	0.08 ± 0.03 [b]	0.09 ± 0.01 [a]
C18:0	11.01 ± 0.287	10.90 ± 0.48	10.84 ± 0.43
C18:1 trans 9	0.22 ± 0.08	0.14 ± 0.01	0.15 ± 0.05
C18:1 cis 9	28.15 ± 0.87 [a]	24.27 ± 1.97 [b]	21.55 ± 0.28 [c]
C18:1 cis 10	1.51 ± 0.16	1.55 ± 0.35	1.57 ± 0.27
C18:1 cis 11	0.06 ± 0.01	0.06 ± 0.01	0.07 ± 0.01
C18:2 cis n6	22.92 ± 0.79 [a]	21.82 ± 2.27 [a]	18.94 ± 1.39 [b]
C20:0	0.13 ± 0.01	0.13 ± 0.02	0.11 ± 0.01
C18:3 n6	0.07 ± 0.03 [a]	0.08 ± 0.02 [a]	0.06 ± 0.01 [b]
C20:1	0.22 ± 0.04 [a]	0.16 ± 0.08 [ab]	0.17 ± 0.02 [a]
C18:3 n3	1.24 ± 0.15 [a]	1.18 ± 0.48 [a]	0.71 ± 0.07 [b]
C18:2 cis9 trans11	0.06 ± 0.02 [b]	0.51 ± 0.05 [a]	0.56 ± 0.04 [a]
C20:2 n6	0.26 ± 0.03 [b]	0.38 ± 0.08 [a]	0.36 ± 0.01 [a]
C22:0	0.04 ± 0.01 [b]	0.08 ± 0.01 [a]	0.06 ± 0.01 [ab]
C20:3 n6	0.04 ± 0.01 [b]	0.09 ± 0.05 [ab]	0.07 ± 0.01 [a]
C22:1	0.32 ± 0.04	0.45 ± 0.17	0.37 ± 0.08
C20:4 n6	3.01 ± 0.34 [b]	4.76 ± 0.73 [a]	4.45 ± 0.12 [a]
C23:0	0.02 ± 0.001	0.02 ± 0.01	0.03 ± 0.002
C22:2 n6	0.08 ± 0.02	0.09 ± 0.03	0.08 ± 0.01
C24:0	0.62 ± 0.12 [b]	0.83 ± 0.38 [ab]	0.89 ± 0.10 [a]
C20:5 n3	0.14 ± 0.01 [b]	0.19 ± 0.10 [ab]	0.20 ± 0.02 [a]
C22:5 n3	0.33 ± 0.06 [b]	0.60 ± 0.36 [ab]	0.54 ± 0.02 [a]
C22:6 n3	0.19 ± 0.01 [b]	0.37 ± 0.22 [ab]	0.33 ± 0.06 [a]
∑ SFA [2]	34.58 ± 1.65 [c]	38.18 ± 0.75 [b]	40.43 ± 0.35 [a]
∑ MUFA [3]	31.72 ± 0.85 [a]	28.10 ± 1.86 [b]	25.59 ± 0.82 [b]
∑ PUFA [4]	28.35 ± 0.44 [b]	30.07 ± 0.80 [a]	26.30 ± 1.61 [b]
∑ n-6	26.39 ± 0.40 [a]	27.22 ± 0.66 [a]	23.96 ± 1.52 [b]
∑ n-3	1.97 ± 0.17 [a]	2.34 ± 0.21 [a]	1.78 ± 0.13 [b]
CLA	0.06 ± 0.02 [b]	0.51 ± 0.05 [a]	0.56 ± 0.04 [a]
PUFA/SFA	0.82 ± 0.05 [a]	0.79 ± 0.02 [a]	0.65 ± 0.04 [b]
n6/n3	13.94 ± 1.17 [a]	11.70 ± 0.78 [b]	13.47 ± 0.40 [a]
LA/ALA	18.67 ± 1.99 [b]	20.54 ± 6.47 [ab]	26.69 ± 1.16 [a]
AA/EPA	20.98 ± 0.70 [b]	24.51 ± 1.31 [a]	22.54 ± 1.79 [ab]

[1] Values are expressed as mean ± standard deviation. [2] SFA, saturated fatty acids; [3] MUFA, monounsaturated fatty acids; [4] PUFA, polyunsaturated fatty acids. [a,b,c] Different letters indicate statistically significant differences between groups for each parameter ($p < 0.05$).

3.2. Bioactive Peptides: Carnosine e Carnitine

The creatine and carnosine contents of the chicken breast are shown in Figure 1. Among the bioactive peptides in meat, carnosine and creatine are considered valuable antioxidant components capable of sequestering and binding to some transition metals, preventing lipid oxidation, and stabilizing the color of meat [38]. On average, the carnosine and creatine contents in group C were 278.11 mg/100 g of meat and 396.1 mg/100 g of

meat, respectively. The creatine content in the C group detected in this study was consistent with that detected by Jung et al. [39] in Korean native chicken, while the carnosine content was higher than that detected in Hubbard Flexi chickens by Kopec et al. [40], who reported significantly lower values (122 mg/100 g of meat). This result could be linked to the genetic type, which significantly influenced the content of these bioactive peptides, in agreement with the findings of Intarapichet and Maikhunthod [41]. Furthermore, Juniper and Rymer [42] conducted a study between different species (pheasant vs. free-range chicken) and observed similar contents of these peptides, suggesting that diet could be a highly influential factor with respect to the impact of the species. A diet characterized by a high carbohydrate content significantly influenced the content of bioactive peptides ($p < 0.01$). In particular, the HI10 inclusion level resulted in a significant increase in creatine content by 22% compared to the control (508.2 vs. 396.1 mg/100 g, respectively; $p < 0.05$) and carnosine content by 26% (278.11 vs. 377.20 mg/100 g, respectively; $p < 0.05$). In the present study, although the HI5 group had slightly higher mean values than the control group, this difference was not statistically significant ($p > 0.05$). Our results may be due to differences in the amino acid utilization efficiency of the different diets, which may have resulted in differences in the free L-histidine pool available for the synthesis of the studied peptides. According to Gkarane et al. [43], changes in the amino acid profile of the diet may influence the peptide content in the breast meat of male chickens (Ross 308). Bioactive peptides are biological molecules in chicken meat that are involved in numerous physiological and health-promoting functions: antioxidants, anti-glycation, and anti-aging, as well as pH buffering in muscle [38]. In the animal muscular system, carnosine is synthesized from histidine (His) and β-alanine (β-Ala) by carnosine synthetase. Creatine, on the other hand, is a nitrogenous organic acid synthesized from l-arginine, glycine, and L-methionine [11]. The positive effects of bioactive peptides on human health are still being studied. However, it is known that ingestion of low histidine-rich peptides could reduce scavenging capacity with little protection from cardiovascular disease [44]. Furthermore, low creatine content may negatively affect the defense systems against neurodegenerative disorders, as anti-oxidant, anti-apoptotic, and anti-excitotoxic properties are decreased [45].

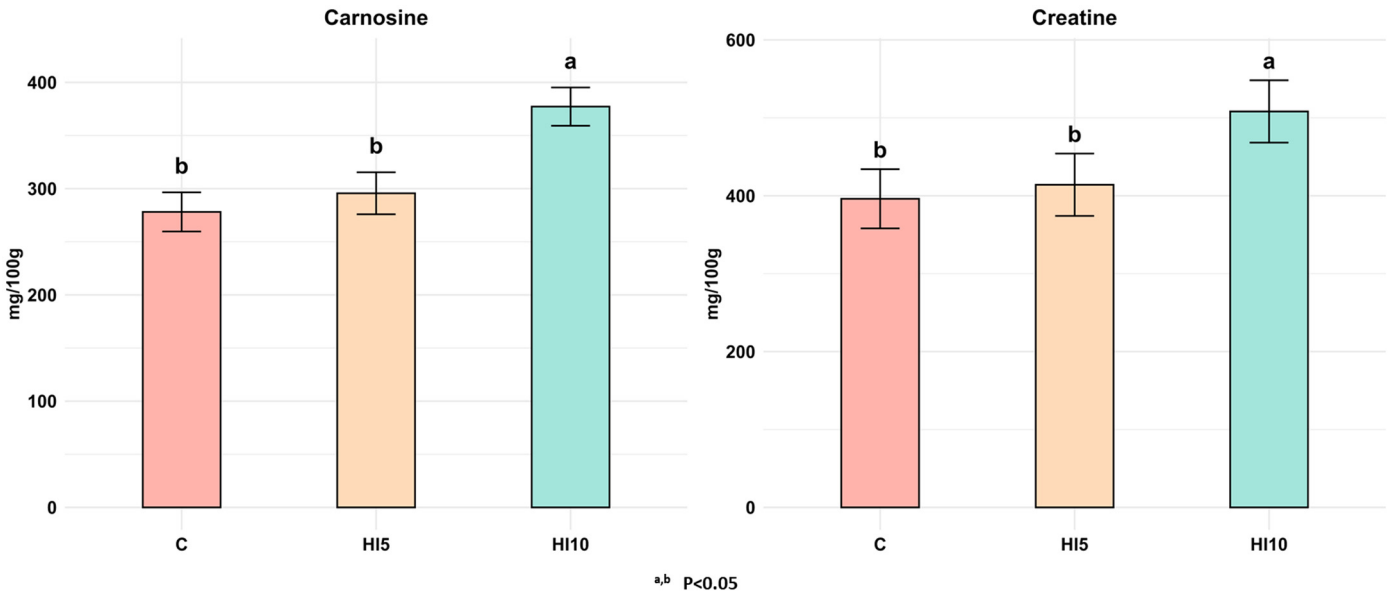

Figure 1. Creatine and carnosine content (mg/100 g of fresh meat) of chicken breast meat. [a,b] Means with different superscripts differ ($p < 0.05$). C = standard diet; HI5 = 5% of level inclusion in diet with BLSF; H10 = 10% of level inclusion in diet with BLSF.

3.3. Functional Fatty Acids Content in Breast Meat

Chicken meat contains several bioactive compounds derived from lipids that contribute to human health, including monounsaturated and polyunsaturated fatty acids (MUFA and PUFA) and conjugated linoleic acid (CLA), which can be considered functional fatty acids. The concentrations of functional fatty acids (MUFA, PUFA, and CLA) in chicken breast are presented in Table 5. Dietary HI supplementation significantly affected the functional fatty acid content of chicken breast meat ($p < 0.01$). Among the monounsaturated fatty acids (MUFA), oleic acid (C18:1 n-9) has been shown to prevent SFA-induced inflammation [46]. MUFA intake and cardiovascular heart disease risk reduction have been positively correlated by Skeaff and Miller [47]. Oleic acid has demonstrated health benefits in diseases like coronary heart disease, rheumatoid arthritis, and cancer. HI supplementation resulted in a decrease in the C18:1 content, from 28.04% in the C group to 24.27% in HI5 and 21.58% in HI10. The differences between the factor levels were statistically significant ($p < 0.05$). Similar results were reported by several authors who investigated the effect of HI inclusion on the quality of chicken meat [48]. Popova et al. [31] confirmed our results on chicken breast of broilers fed HI larvae meal, compared to the control group. As reported by Poureslami et al. [49], the increased intake of SFA in the HI diet resulted in the inhibition of C18:1 n-9 biosynthesis by suppressing elongase activity. Furthermore, Smink et al. [50] attributed the inhibition of δ-9 desaturase in the liver and subsequent reduction in the conversion of SFA to MUFA to the high amount of C18:2 n-6 in the diet. In contrast, Schiavone et al. [34] found an increased proportion of MUFA in the breast muscles of chickens, and Cullere et al. [51] found an increase in MUFA in meat quails fed defatted HI larval meal. The ∑ PUFA content in the breast meat was significantly lower in the HI group than in the control group ($p < 0.05$). Additionally, it was negatively correlated with the dietary inclusion levels of HI (Table 5). These results are consistent with those of many studies that reported an inverse correlation between supplementation with HI maggot meal and total PUFA concentration in chicken meat [52]. However, the fatty acid profile and quality of chicken meat are significantly influenced by nutritional conditions or diet characteristics. Among the ω3 PUFAs, ALA (C 18:3 n3) is an essential fatty acid that must be introduced into the diet. Many clinical studies have confirmed the different functions of human metabolism [16]. ALA has antioxidant properties; it acts as a radical scavenger of hypochlorous acids and peroxy radicals and is capable of chelating metal ions [53]. Furthermore, it plays a key role in the multienzyme complexes of pyruvate dehydrogenase, alpha-ketoglutarate dehydrogen-ase, and branched-chain alpha-keto acid dehydrogenase. [54]. Feeding BSFL resulted in a significant decrease in ALA between the C and treated groups (1.26% vs. 1.18% vs. 0.69% for C, HI5, and HI10, respectively; $p < 0.01$). In particular, the decrease amounted to 45% in the HI10 group. The decreasing trend was similar to what was found by Cullere et al. [55] but in contrast with Schiavone et al. [34], in chicken meat and Secci et al. [56] in Barbaresco partridge meat, observing an increase in ALA correlated to the increase in the level of HI in the diet. The differences could be determined by the method of conservation (freezing) and killing of HI larvae, which influence the concentration of the lipase enzyme [55]. Breast meat is characterized mainly by triacylglycerol and phospholipids, which are rich in n-3 PUFA. Eicosapentaenoic acid (EPA; C20:5 n3) and docosahexaenoic acid (DHA; C22:6 n3) are biological components with beneficial effects on human health [56]. Both are known for their anti-inflammatory [57] and protective of the brain [58] and exert protective functions on endothelial tissue [59]. EPA and DHA levels in the HI group showed a significant increase in their concentration ($p < 0.05$), compared to the control group. In particular, the increase was 26% for both HI5 and HI10 groups. No significant differences were observed between the HI5 and HI10 meat groups. Our results are in line with those of De Souza

Vilela et al. [7], who observed a decrease in the level of total PUFA, while EPA increased in the meat of broilers fed more than 20% whole dried BSFL. However, Schiavone et al. [34] in chicken meat, Cullere et al. [60] in Japanese quail meat, and Secci et al. [56] in Barbary partridge meat, observed a decrease in DHA and EPA content as the level of HI inclusion in the diet increased. However, Renna et al. [61] found a decrease in EPA and DHA contents in trout fillet muscles fed HI compared to other groups. Conjugated fatty acids have been recognized as bioactive molecules owing to their anti-carcinogenic component, conjugated linoleic acid (CLA), which has been found in minced meat [62]. Numerous researchers have associated CLA isomers with numerous biological activities, including antidiabetogenic, anti-obesogenic, anti-inflammatory, antimicrobial, and anti-atherosclerotic activities [63]. Conjugated linoleic acid (CLA) contains all the isomers of linoleic acid. Their structural characteristic is that the two C=C double bonds are conjugated and separated by a single bond. Monogastric animals, such as chickens, do not have a high quantity of CLA-producing bacteria in their digestive system; therefore, endogenous synthesis could be the only source of CLA [64]. It is known that the CLA isomers present in the animal's tissues derive from the diet and that the CLA in chicken covers a small percentage of the total fatty acids (0.1–0.2%) [65]. In the present study, the CLA content in the C group was 0.16% of the total fatty acids, which is consistent with the findings of Seyedalmoosavi et al. [64]. Furthermore, the same research group confirmed that chicken breast meat fed with increasing levels of thawed BSF larvae resulted in an increased CLA content. In our study, a significant increase in its content was observed ($p < 0.05$). In particular, the increase in CLA was 68% for the HI5 group and 71% for HI10 compared to the control. The results of this study indicate the potential use of HI flour in modulating the CLA content in chicken meat. The presence of CLA in BSFL was previously reported by Renna et al. [61], confirming that diet is the main factor that determines a change in fatty acid content, especially in monogastrics [51].

Table 5. Functional fatty acids (% of total FAME) in the breast meat of broiler chickens in the control, HI5 and HI10 groups.

% of Total FAME						
Parameter	**C**		**HI5**		**HI10**	
	μ	**SD**	**μ**	**SD**	**μ**	**SD**
PUFA	28.39 ^{a}	0.93	27.41 ^{b}	1.73	25.89 ^{c}	1.40
C18:1 n-9	28.04 ^{a}	0.87	24.27 ^{b}	1.79	21.58 ^{c}	0.22
C18:3 n-3 (ALA)	1.26 ^{a}	0.15	1.18 ^{a}	0.11	0.69 ^{b}	0.06
C20:5 n-3 (EPA)	0.14 ^{b}	0.01	0.19 ^{a}	0.01	0.19 ^{a}	0.02
C22:6 n-3 (DHA)	0.19 ^{c}	0.02	0.37 ^{a}	0.04	0.31 ^{b}	0.04
CLA	0.06 ^{c}	0.01	0.51 ^{b}	0.05	0.57 ^{a}	0.04

^{a,b,c} Means within a row with different superscripts differ ($p < 0.05$). C = standard diet; HI5 = 5% of level inclusion in diet with BLSF; H10 = 10% of level inclusion in diet with BLSF; PUFA = polyunsaturated fatty acid; ALA = α-linolenic acid; EPA = Eicosapentaenoic acid; DHA = Docosahexaenoic acid.

3.4. Antioxidant Activity

The antioxidant capacity of chicken breast, as determined using three in vitro antioxidant tests, is shown in Figure 2.

Antioxidant activity provides valuable information regarding the functional properties of meat. However, it cannot be evaluated using a single method, as meat is a complex and heterogeneous food composed of multifunctional natural antioxidant compounds. Over the decades, numerous methods have been developed to test the antioxidant activity of food matrices [66]. The percentage of inhibition of ABTS radical action, the FRAP test to estimate the ferric reducing antioxidant power, and the number of thiol groups, i.e., sulfhydryl

groups, using the DTNB reagent, which forms 5-thionitrobenzoic acid and mixed disulfide, were considered in this study.

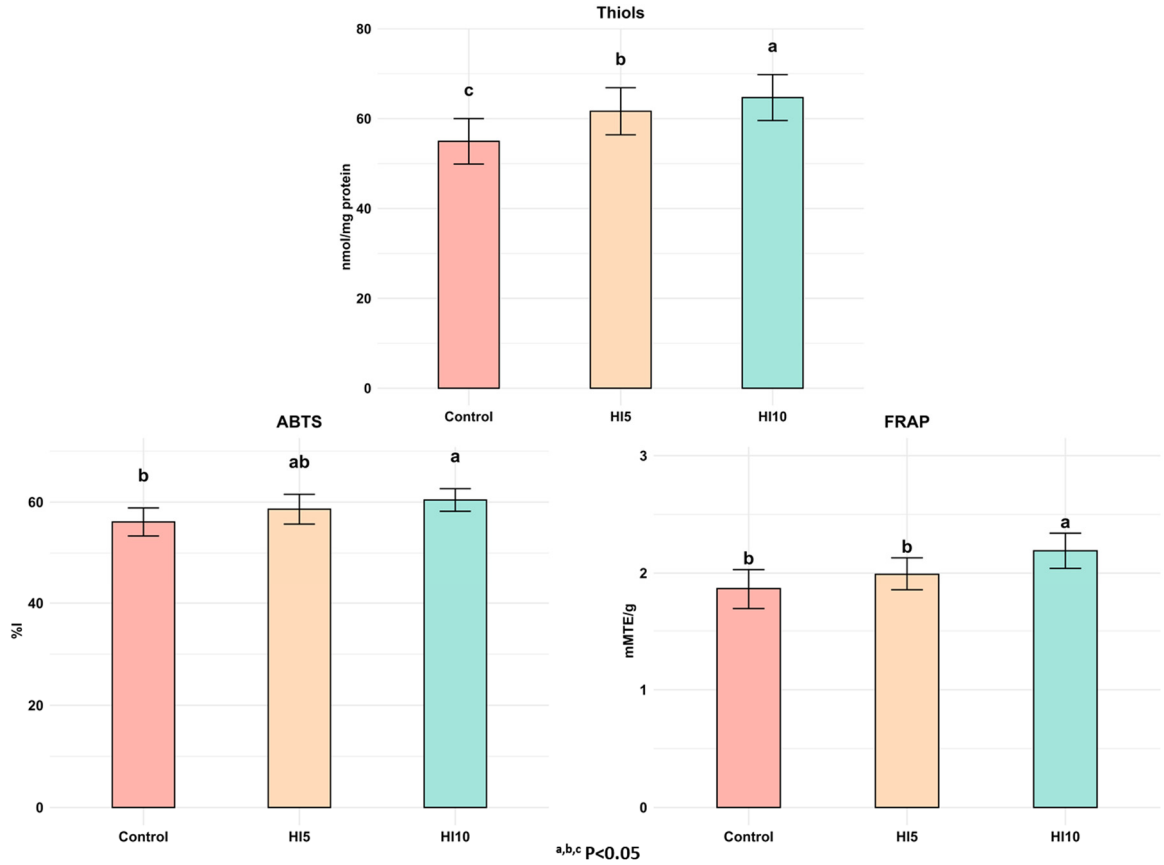

Figure 2. Antioxidant activity, thiols, ABTS, and FRAP in breast meat of broiler chickens, distinctly by group: control, HI5 (5% of inclusion level in diet with BLSF), and HI10 (10% of inclusion level in diet with BLSF). [a,b,c] Means within a row with different superscripts differ ($p < 0.05$).

Overall, the control chicken meat showed an antioxidant activity equal to 56.11% I, 1.86 mMTE/g, and 54.95 nmol -SH groups/mg of protein, which is a desirable characteristic that increases the nutritional value of meat. These results are consistent with those of Sehrawat et al. [67] and Sultana et al. [68] for broiler meat.

Effect of BSF supplementation on ABTS and FRAP antioxidant activity: The ABTS values were significantly influenced by the different levels of HI inclusion ($p < 0.05$), with percentages of 56.11% in the control group (C), 58.61% in the HI5 group, and 60.42% in the HI10 group. Overall, dietary supplementation with HI powder enhanced ABTS radical-scavenging activity, with the highest effect observed in the HI10 group, followed by HI5 and the control group. The FRAP assay values were consistent with the ABTS values. In particular, a significantly higher value was found in HI10 (2.19 ± 0.15 mMTE/g; $p < 0.05$), with an increase in antioxidant activity of 15%, while no significant difference was observed between HI5 and C. Oxidative stress occurs when there is a strong imbalance between the production of reactive oxygen species (ROS) and the volume of antioxidant systems [69]. Insects used as a supplement in animal nutrition, especially for broiler chickens, are an innovative and natural food strategy that contributes to the transfer of natural antioxidant components into muscle tissues [70]. Insects are a rich source of vitamins E and C, carotenoids, and phenolic compounds that enrich meat with beneficial compounds [71]. Many researchers have investigated the potential use of BSF in animal feed. Park et al. [72] reported that the use of BSFL and pupae could be a functional source

with antioxidant properties; Mouithys-Mickalad et al. [73] studied the antioxidant capacity of BSF derivatives and confirmed the removal of ROS. Lu et al. [5] instead demonstrated the presence of functional proteins and peptides with antioxidant properties following enzymatic digestion of BSF. Bioactive peptides are encoded in the protein sequence and are released following enzymatic hydrolysis or fermentation. BSF is rich in proteins; therefore, following gastrointestinal digestion, many bioactive peptides can be released, enter the blood circulation, and be metabolized in organs and tissues [74]. Furthermore, amino acids such as histidine, methionine, and cysteine, which are hydrophobic and aromatic in nature, increase antioxidant activity due to the transfer of a single electron and the ability to chelate metals. However, the antioxidant effects of HI could also be derived from the chitinous exoskeleton, as suggested by Abdel-Latif et al. [75]. Although the chitin content in the larvae is low (5.35% on a dry matter basis). Eggink and Dalsgaard [76] and Ngo et al. [77] clarified that chitin has excellent free radical scavenging ability to inhibit and prevent biological molecular damage in living cells.

Effect of BSF supplementation on thiol groups: A very important aspect of meat quality is the evaluation of oxidation stability, owing to the intrinsic antioxidant capacity of the meat itself. Thiols, along with other components, possess antioxidant properties due to their ability to eliminate free radicals and bind metal ions. This phenomenon is attributable to the presence of -SH groups along the amino acid chain and to the processes of muscle proteolytic degradation by endogenous enzymes, which could expose reactive thiol groups, which is why they are also useful in determining protein oxidation [78]. During intense oxidative stress, a decrease in free sulfhydryls and an increase in disulfides are noted [57]. This test verified the ability of thiols to neutralize free radicals and chelate pro-oxidative metals. In this study, the average content of free thiols in group C was 54.95 ± 5.06 nmolSH/mg protein, a value in line with that reported by Khatun et al. [79] but higher than that reported by Terevinto et al. [80]. Dietary supplementation with HI positively influenced the thiol content ($p < 0.01$). The content in the -SH groups was 61.64 and 64.69 nmolSH/mg protein in HI5 and HI10, respectively ($p < 0.01$). Compared to the control, the percentage increase was 11% for HI5 and 15% for HI10. These results could be due to the supply of amino acids, such as cysteine, present in the experimental diet, which have SH groups linked to the monoacid chain. Insect-enriched diets, rich in protein, contribute to providing highly nutritious components, such as dietary amino acids (AA), and can release bioactive substances with antimicrobial and antioxidant properties [81]. Kar et al. [82] observed that dietary inclusion of BSF enriched metabolic pathways in pig blood plasma, and that they are influenced by amino acid metabolism. These researchers analyzed and quantified the amino acid profiles consisting of essential and non-essential amino acids, as well as intermediate metabolites of amines, which play an important role in cellular protein synthesis and in the biosynthesis of precursors of other active nitrogenous compounds, such as glutathione, a cysteine-rich tripeptide present in the blood plasma of pigs.

4. Conclusions

The results of our research indicate that chicken meat from chickens fed BSFL may possess characteristics typical of a functional food, owing to its higher antioxidant content. Whole grain BSFL supplementation positively influenced antioxidant activity, presumably due to the interaction between endogenous antioxidant compounds from meat and whole grain HI flour, suggesting that the use of BSFL and pupae could represent a useful source of functional feeds. The inclusion of HI improved the content of biopeptides, which could positively contribute to human health and increase the antioxidant activity of meat. Furthermore, HI showed a strong contribution in modifying the functional FA content of

meat lipids. In this study, the inclusion of BSFL positively influenced the contents of CLA, DHA, and EPA, while the contents of total PUFA, ALA, and C18:1 n:9 decreased. These results shed light on the functionality and antioxidant components of meat from chickens fed HI. However, further investigations are needed to identify and evaluate the efficacy, dose response, and in vivo safety of bioactive compounds derived from chicken meat fed diets containing BSFL wholemeal.

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Informed Consent Statement: Not applicable.

Data Availability Statement: The original contributions presented in this study are included in the article. Further inquiries can be directed to the corresponding author.

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References

1. Scieuzo, C.; Franco, A.; Salvia, R.; Triunfo, M.; Addeo, N.F.; Vozzo, S.; Piccolo, G.; Bovera, F.; Ritieni, A.; Francia, A.D.; et al. Enhancement of Fruit Byproducts through Bioconversion by *Hermetia illucens* (Diptera: Stratiomyidae). *Insect Sci.* **2023**, *30*, 991–1010. [CrossRef] [PubMed]
2. Franco, A.; Scieuzo, C.; Salvia, R.; Pucciarelli, V.; Borrelli, L.; Addeo, N.F.; Bovera, F.; Laginestra, A.; Schmitt, E.; Falabella, P. Antimicrobial Activity of Lipids Extracted from Hermetia illucens Reared on Different Substrates. *Appl. Microbiol. Biotechnol.* **2024**, *108*, 167. [CrossRef] [PubMed]
3. English, G.; Wanger, G.; Colombo, S.M. A Review of Advancements in Black Soldier Fly (*Hermetia Illucens*) Production for Dietary Inclusion in Salmonid Feeds. *J. Agric. Food Res.* **2021**, *5*, 100164. [CrossRef]
4. Siddiqui, S.A.; Ristow, B.; Rahayu, T.; Putra, N.S.; Widya Yuwono, N.; Nisa', K.; Mategeko, B.; Smetana, S.; Saki, M.; Nawaz, A.; et al. Black Soldier Fly Larvae (BSFL) and Their Affinity for Organic Waste Processing. *Waste Manag.* **2022**, *140*, 1–13. [CrossRef]
5. Lu, S.; Taethaisong, N.; Meethip, W.; Surakhunthod, J.; Sinpru, B.; Sroichak, T.; Archa, P.; Thongpea, S.; Paengkoum, S.; Purba, R.A.P.; et al. Nutritional Composition of Black Soldier Fly Larvae (*Hermetia illucens* L.) and Its Potential Uses as Alternative Protein Sources in Animal Diets: A Review. *Insects* **2022**, *13*, 831. [CrossRef]
6. Spranghers, T.; Ottoboni, M.; Klootwijk, C.; Ovyn, A.; Deboosere, S.; De Meulenaer, B.; Michiels, J.; Eeckhout, M.; De Clercq, P.; De Smet, S. Nutritional Composition of Black Soldier Fly (*Hermetia illucens*) Prepupae Reared on Different Organic Waste Substrates. *J. Sci. Food Agric.* **2017**, *97*, 2594–2600. [CrossRef]
7. De Souza Vilela, J.; Alvarenga, T.I.R.C.; Andrew, N.R.; McPhee, M.; Kolakshyapati, M.; Hopkins, D.L.; Ruhnke, I. Technological Quality, Amino Acid and Fatty Acid Profile of Broiler Meat Enhanced by Dietary Inclusion of Black Soldier Fly Larvae. *Foods* **2021**, *10*, 297. [CrossRef]
8. Ullah, H. Feed Sustainability and Efficiency. In *Poultry Science*; IntechOpen: London, UK, 2023; ISBN 978-1-80356-155-4.
9. Kulczyński, B.; Sidor, A.; Gramza-Michałowska, A. Characteristics of Selected Antioxidative and Bioactive Compounds in Meat and Animal Origin Products. *Antioxidants* **2019**, *8*, 335. [CrossRef]
10. Cruz-Casas, D.E.; Aguilar, C.N.; Ascacio-Valdés, J.A.; Rodríguez-Herrera, R.; Chávez-González, M.L.; Flores-Gallegos, A.C. Enzymatic Hydrolysis and Microbial Fermentation: The Most Favorable Biotechnological Methods for the Release of Bioactive Peptides. *Food Chem. Mol. Sci.* **2021**, *3*, 100047. [CrossRef]

11. Jukić, I.; Kolobarić, N.; Stupin, A.; Matić, A.; Kozina, N.; Mihaljević, Z.; Mihalj, M.; Šušnjara, P.; Stupin, M.; Ćurić, Ž.B.; et al. Carnosine, Small but Mighty—Prospect of Use as Functional Ingredient for Functional Food Formulation. *Antioxidants* **2021**, *10*, 1037. [CrossRef]
12. Xing, L.; Li, G.; Toldrá, F.; Zhang, W. The Physiological Activity of Bioactive Peptides Obtained from Meat and Meat By-Products. In *Advances in Food and Nutrition Research*; Elsevier: Amsterdam, The Netherlands, 2021; Volume 97, pp. 147–185. ISBN 978-0-12-824580-4.
13. Gulcin, İ. Antioxidants and Antioxidant Methods: An Updated Overview. *Arch. Toxicol.* **2020**, *94*, 651–715. [CrossRef]
14. Thakur, M.; Gupta, N.; Sharma, H.K.; Devi, S. Physicochemical Characteristics and Mineral Status of Honey from Different Agro-Climatic Zones of Himachal Pradesh, India. *BFJ* **2021**, *123*, 3789–3804. [CrossRef]
15. Virmani, M.A.; Cirulli, M. The Role of L-Carnitine in Mitochondria, Prevention of Metabolic Inflexibility and Disease Initiation. *Int. J. Mol. Sci.* **2022**, *23*, 2717. [CrossRef]
16. Borsini, A.; Nicolaou, A.; Camacho-Muñoz, D.; Kendall, A.C.; Di Benedetto, M.G.; Giacobbe, J.; Su, K.-P.; Pariante, C.M. Omega-3 Polyunsaturated Fatty Acids Protect against Inflammation through Production of LOX and CYP450 Lipid Mediators: Relevance for Major Depression and for Human Hippocampal Neurogenesis. *Mol. Psychiatry* **2021**, *26*, 6773–6788. [CrossRef]
17. Hennessy, A.A.; Ross, P.R.; Fitzgerald, G.F.; Stanton, C. Sources and Bioactive Properties of Conjugated Dietary Fatty Acids. *Lipids* **2016**, *51*, 377–397. [CrossRef] [PubMed]
18. Dal Bosco, A.; Cavallo, M.; Menchetti, L.; Angelucci, E.; Cartoni Mancinelli, A.; Vaudo, G.; Marconi, S.; Camilli, E.; Galli, F.; Castellini, C.; et al. The Healthy Fatty Index Allows for Deeper Insights into the Lipid Composition of Foods of Animal Origin When Compared with the Atherogenic and Thrombogenicity Indexes. *Foods* **2024**, *13*, 1568. [CrossRef] [PubMed]
19. Lackner, J.; Hess, V.; Marx, A.; Hosseini-Ghaffari, M.; Sauerwein, H. Effects of Dietary Supplementation with Histidine and β-Alanine on Blood Plasma Metabolome of Broiler Chickens at Different Ages. *PLoS ONE* **2022**, *17*, e0277476. [CrossRef]
20. Zampiga, M.; Calini, F.; Sirri, F. Importance of Feed Efficiency for Sustainable Intensification of Chicken Meat Production: Implications and Role for Amino Acids, Feed Enzymes and Organic Trace Minerals. *World's Poult. Sci. J.* **2021**, *77*, 1–21. [CrossRef]
21. Scala, A.; Cammack, J.A.; Salvia, R.; Scieuzo, C.; Franco, A.; Bufo, S.A.; Tomberlin, J.K.; Falabella, P. Rearing Substrate Impacts Growth and Macronutrient Composition of *Hermetia illucens* (L.) (Diptera: Stratiomyidae) Larvae Produced at an Industrial Scale. *Sci. Rep.* **2020**, *10*, 19448. [CrossRef]
22. Official Methods of Analysis Program—AOAC International. Available online: https://www.aoac.org/scientific-solutions/standards-and-official-methods/ (accessed on 23 February 2023).
23. Khan, I.A.; Parker, N.B.; Löhr, C.V.; Cherian, G. Docosahexaenoic Acid (22:6 n-3)-Rich Microalgae along with Methionine Supplementation in Broiler Chickens: Effects on Production Performance, Breast Muscle Quality Attributes, Lipid Profile, and Incidence of White Striping and Myopathy. *Poult. Sci.* **2021**, *100*, 865–874. [CrossRef]
24. Perna, A.; Gambacorta, E.; Simonetti, A.; Grassi, G.; Scopa, A. Effect of Ozone Treatment Exposure Time on Oxidative Stability of Cream Milk. *Eur. J. Lipid Sci. Tech.* **2022**, *124*, 2100238. [CrossRef]
25. Folch, J.; Lees, M.; Sloane Stanley, G.H. A Simple Method for the Isolation and Purification of Total Lipides from Animal Tissues. *J. Biol. Chem.* **1957**, *226*, 497–509. [CrossRef]
26. Perna, A.; Simonetti, A.; Grassi, G.; Gambacorta, E. Effect of a Cauliflower (*Brassica oleraceae* Var. Botrytis) Leaf Powder-Enriched Diet on Performance, Carcass and Meat Characteristics of Growing Rabbit. *Meat Sci.* **2019**, *149*, 134–140. [CrossRef] [PubMed]
27. Grassi, G.; Capasso, G.; Rando, A.; Perna, A.M. Antioxidant Activity of Beef, Pork and Chicken Burgers before and after Cooking and after In Vitro Intestinal Digestion. *Foods* **2023**, *12*, 4100. [CrossRef]
28. Benzie, I.F.; Strain, J.J. The Ferric Reducing Ability of Plasma (FRAP) as a Measure of "Antioxidant Power": The FRAP Assay. *Anal. Biochem.* **1996**, *239*, 70–76. [CrossRef] [PubMed]
29. Grassi, G.; Di Gregorio, P.; Rando, A.; Perna, A.M. Quality and Sensorial Evaluation of Beef Burgers Added with Sicilian Sumac (*Rhus coriaria* L.). *Heliyon* **2024**, *10*, e26848. [CrossRef]
30. Simonetti, A.; Gambacorta, E.; Perna, A. Antioxidative and Antihypertensive Activities of Pig Meat before and after Cooking and in Vitro Gastrointestinal Digestion: Comparison between Italian Autochthonous Pig Suino Nero Lucano and a Modern Crossbred Pig. *Food Chem.* **2016**, *212*, 590–595. [CrossRef]
31. Popova, T.L.; Petkov, E.; Ignatova, M. Effect of Black Soldier Fly (*Hermetia illucens*) Meals on the Meat Quality in Broilers. *Agric. Food Sci.* **2020**, *29*, 177–188. [CrossRef]
32. Londok, J.; Sumiati, S.; Wiryawan, K.; Manalu, W. Antioxidant Enzyme Activity and Malondialdehyde Concentration on Broiler Fed Contain Lauric Acid and Areca Vestiaria Giseke. *Bul. Peternak.* **2018**, *42*. [CrossRef]
33. Skřivan, M.; Marounek, M.; Englmaierová, M.; Čermák, L.; Vlčková, J.; Skřivanová, E. Effect of Dietary Fat Type on Intestinal Digestibility of Fatty Acids, Fatty Acid Profiles of Breast Meat and Abdominal Fat, and mRNA Expression of Lipid-Related Genes in Broiler Chickens. *PLoS ONE* **2018**, *13*, e0196035. [CrossRef]

34. Schiavone, A.; Dabbou, S.; Petracci, M.; Zampiga, M.; Sirri, F.; Biasato, I.; Gai, F.; Gasco, L. Black Soldier Fly Defatted Meal as a Dietary Protein Source for Broiler Chickens: Effects on Carcass Traits, Breast Meat Quality and Safety. *Animal* **2019**, *13*, 2397–2405. [CrossRef] [PubMed]
35. Hankel, J.; Popp, J.; Meemken, D.; Zeiger, K.; Beyerbach, M.; Taube, V.; Klein, G.; Visscher, C. Influence of Lauric Acid on the Susceptibility of Chickens to an Experimental Campylobacter Jejuni Colonisation. *PLoS ONE* **2018**, *13*, e0204483. [CrossRef] [PubMed]
36. Wessels, K.; Rip, D.; Gouws, P. Salmonella in Chicken Meat: Consumption, Outbreaks, Characteristics, Current Control Methods and the Potential of Bacteriophage Use. *Foods* **2021**, *10*, 1742. [CrossRef]
37. Cui, X.Y.; Gou, Z.Y.; Abouelezz, K.F.M.; Li, L.; Lin, X.J.; Fan, Q.L.; Wang, Y.B.; Cheng, Z.G.; Ding, F.Y.; Jiang, S.Q. Alterations of the Fatty Acid Composition and Lipid Metabolome of Breast Muscle in Chickens Exposed to Dietary Mixed Edible Oils. *Animal* **2020**, *14*, 1322–1332. [CrossRef] [PubMed]
38. Ahmed, T.; Sun, X.; Udenigwe, C.C. Role of Structural Properties of Bioactive Peptides in Their Stability during Simulated Gastrointestinal Digestion: A Systematic Review. *Trends Food Sci. Technol.* **2022**, *120*, 265–273. [CrossRef]
39. Jung, S.; Bae, Y.S.; Kim, H.J.; Jayasena, D.D.; Lee, J.H.; Park, H.B.; Heo, K.N.; Jo, C. Carnosine, Anserine, Creatine, and Inosine 5′-Monophosphate Contents in Breast and Thigh Meats from 5 Lines of Korean Native Chicken. *Poult. Sci.* **2013**, *92*, 3275–3282. [CrossRef]
40. Kopec, W.; Jamroz, D.; Wiliczkiewicz, A.; Biazik, E.; Pudlo, A.; Korzeniowska, M.; Hikawczuk, T.; Skiba, T. Antioxidative Characteristics of Chicken Breast Meat and Blood after Diet Supplementation with Carnosine, L-Histidine, and β-Alanine. *Antioxidants* **2020**, *9*, 1093. [CrossRef]
41. Intarapichet, K.-O.; Maikhunthod, B. Genotype and Gender Differences in Carnosine Extracts and Antioxidant Activities of Chicken Breast and Thigh Meats. *Meat Sci.* **2005**, *71*, 634–642. [CrossRef]
42. Juniper, D.; Rymer, C. The Effect of Rearing System and Cooking Method on the Carnosine and Anserine Content of Poultry and Game Meat. *J. Food Nut. Agric.* **2018**, *1*, 35–39. [CrossRef]
43. Gkarane, V.; Ciulu, M.; Altmann, B.; Mörlein, D. Effect of Alternative Protein Feeds on the Content of Selected Endogenous Bioactive and Flavour-Related Compounds in Chicken Breast Meat. *Foods* **2020**, *9*, 392. [CrossRef]
44. Ohata, M.; Uchida, S.; Zhou, L.; Arihara, K. Antioxidant Activity of Fermented Meat Sauce and Isolation of an Associated Antioxidant Peptide. *Food Chem.* **2016**, *194*, 1034–1039. [CrossRef]
45. Chang, H.; Leem, Y.-H. The Potential Role of Creatine Supplementation in Neurodegenerative Diseases. *Phys. Act. Nutr.* **2023**, *27*, 48–54. [CrossRef] [PubMed]
46. Piccinin, E.; Cariello, M.; De Santis, S.; Ducheix, S.; Sabbà, C.; Ntambi, J.M.; Moschetta, A. Role of Oleic Acid in the Gut-Liver Axis: From Diet to the Regulation of Its Synthesis via Stearoyl-CoA Desaturase 1 (SCD1). *Nutrients* **2019**, *11*, 2283. [CrossRef]
47. Skeaff, C.M.; Miller, J. Dietary Fat and Coronary Heart Disease: Summary of Evidence from Prospective Cohort and Randomised Controlled Trials. *Ann. Nutr. Metab.* **2009**, *55*, 173–201. [CrossRef] [PubMed]
48. Daszkiewicz, T.; Murawska, D.; Kubiak, D.; Han, J. Chemical Composition and Fatty Acid Profile of the Pectoralis Major Muscle in Broiler Chickens Fed Diets with Full-Fat Black Soldier Fly (*Hermetia illucens*) Larvae Meal. *Animals* **2022**, *12*, 464. [CrossRef]
49. Poureslami, R.; Turchini, G.M.; Raes, K.; Huyghebaert, G.; De Smet, S. Effect of Diet, Sex and Age on Fatty Acid Metabolism in Broiler Chickens: SFA and MUFA. *Br. J. Nutr.* **2010**, *104*, 204–213. [CrossRef]
50. Smink, W.; Gerrits, W.J.J.; Hovenier, R.; Geelen, M.J.H.; Verstegen, M.W.A.; Beynen, A.C. Effect of Dietary Fat Sources on Fatty Acid Deposition and Lipid Metabolism in Broiler Chickens. *Poult. Sci.* **2010**, *89*, 2432–2440. [CrossRef] [PubMed]
51. Cullere, M.; Tasoniero, G.; Giaccone, V.; Miotti-Scapin, R.; Claeys, E.; De Smet, S.; Dalle Zotte, A. Black Soldier Fly as Dietary Protein Source for Broiler Quails: Apparent Digestibility, Excreta Microbial Load, Feed Choice, Performance, Carcass and Meat Traits. *Animal* **2016**, *10*, 1923–1930. [CrossRef]
52. Hwangbo, J.; Hong, E.C.; Jang, A.; Kang, H.K.; Oh, J.S.; Kim, B.W.; Park, B.S. Utilization of House Fly-Maggots, a Feed Supplement in the Production of Broiler Chickens. *J. Environ. Biol.* **2009**, *30*, 609–614.
53. Isenmann, E.; Trittel, L.; Diel, P. The Effects of Alpha Lipoic Acid on Muscle Strength Recovery after a Single and a Short-Term Chronic Supplementation—A Study in Healthy Well-Trained Individuals after Intensive Resistance and Endurance Training. *J. Int. Soc. Sports Nutr.* **2020**, *17*, 61. [CrossRef]
54. Salinthone, S.; Yadav, V.; Bourdette, D.N.; Carr, D.W. Lipoic Acid: A Novel Therapeutic Approach for Multiple Sclerosis and Other Chronic Inflammatory Diseases of the CNS. Available online: https://www.ingentaconnect.com/content/ben/emiddt/2008/00000008/00000002/art00008 (accessed on 2 May 2024).
55. Cullere, M.; Schiavone, A.; Dabbou, S.; Gasco, L.; Dalle Zotte, A. Meat Quality and Sensory Traits of Finisher Broiler Chickens Fed with Black Soldier Fly (*Hermetia illucens* L.) Larvae Fat as Alternative Fat Source. *Animals* **2019**, *9*, 140. [CrossRef]
56. Secci, G.; Moniello, G.; Gasco, L.; Bovera, F.; Parisi, G. Barbary Partridge Meat Quality as Affected by Hermetia Illucens and Tenebrio Molitor Larva Meals in Feeds. *Food Res. Int.* **2018**, *112*, 291–298. [CrossRef]

57. Ian Givens, D.; Gibbs, R.A. Current Intakes of EPA and DHA in European Populations and the Potential of Animal-Derived Foods to Increase Them: Symposium on 'How Can the *n*-3 Content of the Diet Be Improved? *Proc. Nutr. Soc.* **2008**, *67*, 273–280. [CrossRef] [PubMed]
58. von Schacky, C. Importance of EPA and DHA Blood Levels in Brain Structure and Function. *Nutrients* **2021**, *13*, 1074. [CrossRef] [PubMed]
59. Cunnane, S.C.; Ho, S.Y.; Dore-Duffy, P.; Ells, K.R.; Horrobin, D.F. Essential Fatty Acid and Lipid Profiles in Plasma and Erythrocytes in Patients with Multiple Sclerosis. *Am. J. Clin. Nutr.* **1989**, *50*, 801–806. [CrossRef]
60. Cullere, M.; Singh, Y.; Pellattiero, E.; Berzuini, S.; Galasso, I.; Clemente, C.; Dalle Zotte, A. Effect of the Dietary Inclusion of Camelina Sativa Cake into Quail Diet on Live Performance, Carcass Traits, and Meat Quality. *Poult. Sci.* **2023**, *102*, 102650. [CrossRef] [PubMed]
61. Renna, M.; Schiavone, A.; Gai, F.; Dabbou, S.; Lussiana, C.; Malfatto, V.; Prearo, M.; Capucchio, M.T.; Biasato, I.; Biasibetti, E.; et al. Evaluation of the Suitability of a Partially Defatted Black Soldier Fly (*Hermetia illucens* L.) Larvae Meal as Ingredient for Rainbow Trout (Oncorhynchus Mykiss Walbaum) Diets. *J. Animal Sci. Biotechnol.* **2017**, *8*, 57. [CrossRef]
62. Bhattacharya, A.; Banu, J.; Rahman, M.; Causey, J.; Fernandes, G. Biological Effects of Conjugated Linoleic Acids in Health and Disease. *J. Nutr. Biochem.* **2006**, *17*, 789–810. [CrossRef]
63. Li, S.; Xu, L.; Qing, J.; Wu, X.; Li, H.; Chen, H.; Liu, X. Multiple Biological Activities and Biosynthesis Mechanisms of Specific Conjugated Linoleic Acid Isomers and Analytical Methods for Prospective Application. *Food Chem.* **2023**, *409*, 135257. [CrossRef]
64. Seyedalmoosavi, M.M.; Mielenz, M.; Görs, S.; Wolf, P.; Daş, G.; Metges, C.C. Effects of Increasing Levels of Whole Black Soldier Fly (*Hermetia illucens*) Larvae in Broiler Rations on Acceptance, Nutrient and Energy Intakes and Utilization, and Growth Performance of Broilers. *Poult. Sci.* **2022**, *101*, 102202. [CrossRef]
65. Polidori, P.; Vincenzetti, S.; Pucciarelli, S.; Polzonetti, V. CLAs in Animal Source Foods: Healthy Benefits for Consumers. In *Bioactive Molecules in Food*; Mérillon, J.-M., Ramawat, K.G., Eds.; Reference Series in Phytochemistry; Springer International Publishing: Cham, Switzerland, 2019; pp. 667–698. ISBN 978-3-319-78029-0.
66. Liu, K.; Luo, M.; Wei, S. The Bioprotective Effects of Polyphenols on Metabolic Syndrome against Oxidative Stress: Evidences and Perspectives. *Oxidative Med. Cell. Longev.* **2019**, *2019*, e6713194. [CrossRef] [PubMed]
67. Sehrawat, R.; Sharma, R.; Ahlawat, S.; Sharma, V.; Thakur, M.S.; Kaur, M.; Tantia, M.S. First Report on Better Functional Property of Black Chicken Meat from India. *Indian J. Anim. Res.* **2021**, *55*, 727–733. [CrossRef]
68. Sultana, K.; Jayathilakan, K.; Pandey, M.C. Evaluation of Antioxidant Activity, Radical Scavenging, and Reducing Power of Clove Oil and Clove Oleoresin in Comparison with Natural and Synthetic Antioxidants in Chevon (Capra Aegagrus Hircus) and Chicken Meat. *Def. Life Sci. J.* **2017**, *3*, 51. [CrossRef]
69. Monaghan, P.; Metcalfe, N.B.; Torres, R. Oxidative Stress as a Mediator of Life History Trade-offs: Mechanisms, Measurements and Interpretation. *Ecol. Lett.* **2009**, *12*, 75–92. [CrossRef]
70. Torres-Castillo, J.A.; Olazarán-Santibáñez, F.E. Insects as Source of Phenolic and Antioxidant Entomochemicals in the Food Industry. *Front. Nutr.* **2023**, *10*, 1133342. [CrossRef] [PubMed]
71. Rumpold, B.A.; Schlüter, O.K. Potential and Challenges of Insects as an Innovative Source for Food and Feed Production. *Innov. Food Sci. Emerg. Technol.* **2013**, *17*, 1–11. [CrossRef]
72. Park, S.; Chang, B.S.; Yoe, S.M. Detection of Antimicrobial Substances from Larvae of the Black Soldier Fly, *Hermetia illucens* (Diptera: S Tratiomyidae). *Entomol. Res.* **2014**, *44*, 58–64. [CrossRef]
73. Mouithys-Mickalad, A.; Schmitt, E.; Dalim, M.; Franck, T.; Tome, N.M.; Van Spankeren, M.; Serteyn, D.; Paul, A. Black Soldier Fly (*Hermetia illucens*) Larvae Protein Derivatives: Potential to Promote Animal Health. *Animals* **2020**, *10*, 941. [CrossRef]
74. Zielińska, E.; Karaś, M.; Jakubczyk, A. Antioxidant Activity of Predigested Protein Obtained from a Range of Farmed Edible Insects. *Int. J. Food Sci. Tech.* **2017**, *52*, 306–312. [CrossRef]
75. Abdel-Latif, H.M.R.; Abdel-Tawwab, M.; Khalil, R.H.; Metwally, A.A.; Shakweer, M.S.; Ghetas, H.A.; Khallaf, M.A. Black Soldier Fly (*Hermetia illucens*) Larvae Meal in Diets of European Seabass: Effects on Antioxidative Capacity, Non-Specific Immunity, Transcriptomic Responses, and Resistance to the Challenge with Vibrio Alginolyticus. *Fish Shellfish Immunol.* **2021**, *111*, 111–118. [CrossRef]
76. Eggink, K.M.; Dalsgaard, J. Chitin Contents in Different Black Soldier Fly (*Hermetia illucens*) Life Stages. *J. Insects Food Feed* **2023**, *9*, 855–864. [CrossRef]
77. Ngo, D.-N.; Lee, S.-H.; Kim, M.-M.; Kim, S.-K. Production of Chitin Oligosaccharides with Different Molecular Weights and Their Antioxidant Effect in RAW 264.7 Cells. *J. Funct. Foods* **2009**, *1*, 188–198. [CrossRef]
78. Ulrich, K.; Jakob, U. The Role of Thiols in Antioxidant Systems. *Free Radic. Biol. Med.* **2019**, *140*, 14–27. [CrossRef] [PubMed]
79. Khatun, J.; Loh, T.C.; Foo, H.L.; Akit, H.; Mohamad, R.; Shazali, N. Effects of Vitamin E, an Oil Blend and L-Arginine on Breast Meat from Broiler Chickens. *S. Afr. J. Anim. Sci.* **2021**, *50*, 745–757. [CrossRef]
80. Terevinto, A.; Del Puerto, M.; Da Silva, A.; Cabrera, M.C.; Saadoun, A. Effect of Chia Seeds (*Salvia hispanica* L.) Inclusion in Poultry Diet on n-3 Enrichment and Oxidative Status of Meat during Retail Display. *CyTA—J. Food* **2023**, *21*, 93–100. [CrossRef]

81. Jayasena, D.D.; Jung, S.; Bae, Y.S.; Kim, S.H.; Lee, S.K.; Lee, J.H.; Jo, C. Changes in Endogenous Bioactive Compounds of Korean Native Chicken Meat at Different Ages and during Cooking. *Poult. Sci.* **2014**, *93*, 1842–1849. [CrossRef]
82. Kar, S.K.; Schokker, D.; Harms, A.C.; Kruijt, L.; Smits, M.A.; Jansman, A.J.M. Local Intestinal Microbiota Response and Systemic Effects of Feeding Black Soldier Fly Larvae to Replace Soybean Meal in Growing Pigs. *Sci. Rep.* **2021**, *11*, 15088. [CrossRef]