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Correlation between Biogenic Amines and Their Precursors in Stored Chicken Meat

Wojciech Wójcik [1,*], Krzysztof Damaziak [1], Monika Łukasiewicz-Mierzejewska [1], Olga Świder [2], Jan Niemiec [1], Michał Wójcicki [3], Marek Roszko [2], Dariusz Gozdowski [4], Julia Riedel [1] and Agata Marzec [5]

[1] Department of Animal Breeding, Institute of Animal Science, Warsaw University of Life Sciences—SGGW, Ciszewskiego 8, 02-786 Warsaw, Poland; krzysztof_damaziak@sggw.edu.pl (K.D.); monika_lukasiewicz@sggw.edu.pl (M.Ł.-M.); jan_niemiec@sggw.edu.pl (J.N.); julia_riedel@sggw.edu.pl (J.R.)
[2] Department of Food Safety and Chemical Analysis, Prof. Waclaw Dabrowski Institute of Agricultural and Food Biotechnology—State Research Institute, Rakowiecka 36, 02-532 Warsaw, Poland; olga.swider@ibprs.pl (O.Ś.); marek.roszko@ibprs.pl (M.R.)
[3] Department of Microbiology, Prof. Waclaw Dabrowski Institute of Agricultural and Food Biotechnology—State Research Institute, Rakowiecka 36, 02-532 Warsaw, Poland
[4] Department of Biometry, Faculty of Agriculture and Biology, Institute of Agriculture, Warsaw University of Life Sciences, Nowoursynowska 159, 02-786 Warsaw, Poland; dariusz_gozdowski@sggw.edu.pl
[5] Department of Food Engineering and Process Management, Institute of Food Sciences, Warsaw University of Life Sciences, Nowoursynowska 159c, 02-776 Warsaw, Poland
* Correspondence: wojciech_wojcik@sggw.edu.pl

Abstract: Biogenic amines (BAs) are biologically active substances found in the cells of microorganisms, plants, and animals. These BAs serve many vital functions in the body. However, an excessive amount can be toxic, especially for individuals taking monoamine oxidase (MAO) and diamine oxidase (DAO) inhibitors. They primarily form in products rich in amino acids, the primary substrates for BA formation. The aim of this study was to determine the formation of BAs and their precursor amino acids in chicken breast and leg muscles stored under chilling conditions. Analyses of BA and AA determinations were conducted on days 1, 3, 5, 7, and 10 of muscle storage. There was a noted increase in BAs with the storage of both muscle types ($p < 0.05$). Distinct levels of BAs were detected ($p < 0.05$) in the muscles, except for putrescine ($p > 0.05$). Interactions emerged between the two factors for various Bas, including histamine ($p = 0.001$), tyramine ($p < 0.001$), BAI index ($p < 0.001$), tryptamine ($p < 0.001$), agmatine ($p = 0.001$), spermidine ($p < 0.001$), TOTAL BA-1 ($p < 0.001$), and TOTAL BA-2 ($p = 0.016$). There was no evident interaction between the type of meat and storage time concerning amino acid content ($p > 0.05$). Correlations in breast muscles were observed for biogenic amine–amino acid pairs such as putrescine–ornithine ($r = -0.57$) ($p < 0.05$), spermidine–ornithine ($r = -0.73$) ($p < 0.05$), and phenylethylamine–phenylethylalanine ($r = -0.50$) ($p < 0.05$). In leg muscles, significant correlations were found for histamine–histidine ($r = -0.87$) ($p < 0.05$), putrescine–ornithine ($r = -0.96$) ($p < 0.05$), and phenylethylamine–phenylethylalanine ($r = -0.65$) ($p < 0.05$). The results obtained can be used in the future to estimate the levels of BAs with knowledge of the levels of individual amino acids and inversely.

Keywords: chicken meat; biogenic amines; amino acids; correlation

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1. Introduction

Biogenic amines (BAs) are small-molecule biologically active substances found in the cells of microorganisms, plants, and animals. They have many important functions in the body. Biogenic amines are compounds that are crucial for the proper course of the organism's metabolic processes, such as protein synthesis, hormone synthesis, and DNA replication, as well as maintaining cell viability [1,2]. However, due to their toxic effects (excessive consumption of BAs causes diarrhea, food poisoning, vomiting, sweating, or tachycardia), it is important to limit their levels in the human diet, especially among people

taking antidepressants and drugs from the monoamine oxidase (MAO) and diamine oxidase (DAO) groups. Furthermore, excessive amounts of BAs can act as false neurotransmitters, leading to numerous allergic reactions [1,2].

Biogenic amines are produced through three processes: the decarboxylation of amino acids, the reductive transamination of aldehydes and ketones, and the formation within tissues. The availability of a substrate (precursor) is a determinant for the creation of BAs, as it is from this substrate that the amine originates. Biogenic amines can be categorized based on the structure of the precursor amino acid—aliphatic, aromatic, or heterocyclic—and by the number of bonds between the amino acid residue and the nitrogen atom, such as monoamines, diamines, and polyamines. Putrescine arises from ornithine, tyramine from tyrosine, histamine from histidine, tryptamine from tryptophan, cadaverine from lysine, phenylethylamine from phenylethylalanine, and agmatine from arginine. Spermine and spermidine are derived from arginine and ornithine. Elevated levels of amino acids (AA) can result in significant amounts of BAs, contingent on their content [2–5].

Poultry meat is one of the most popular products among consumers. Its popularity has been influenced mainly by the lack of religious restrictions and the reduction in the rearing period of chickens, coupled with shifts in rearing methodologies. Presently, chickens are mainly kept in intensive systems in poultry production. Genetic improvement to increase the proportion of breast and leg muscles has led to a change in the structure of the meat, specifically the muscle fibers. Consequently, poultry meat tends to deteriorate quite rapidly. The ongoing proteolysis results in increased levels of free amino acids (FAAs), which serve as precursors of BAs [6,7].

The aim of this study was to determine the levels of BAs and their precursor AAs and to explore the relationship between them in the breast and leg muscles of refrigerated-stored chickens.

2. Materials and Methods

2.1. Experimental Scheme

Breast and leg muscles were obtained from chickens nourished on a diet based on wheat, maize, and soya, structured in a three-stage system: 0–16 d, starter: 11.5 MJ energy, 261 g crude protein (CP)/kg; 17–35 d, grower: 12.1 MJ energy, 221 g CP/kg; and 36–42 d, finisher: 13.4 MJ energy, 187 g CP/kg. After slaughtering and carcass cooling, 10 samples of each breast and leg muscle were obtained. Each muscle sample was individually homogenized using a meat grinder featuring 3 mm holes and was rigorously mixed to ensure homogeneity. The protein and fat contents within the procured muscles were assessed using a Food Scan™ analyzer (Foss Electric, Hillerød, Denmark). The protein level in the breast muscles was registered at 22.23 $\pm$ 0.39%, whereas it stood at 20.04 $\pm$ 0.42% for the leg muscles. Concurrently, the fat level was 2.73 $\pm$ 0.09% in the breast muscles and 7.81 $\pm$ 0.14% in the leg muscles. The obtained homogenate was subsequently divided into five samples (each sample weighed 20 g), each encased in polyethylene (PE) film string bags of dimensions 100 $\times$ 150 mm. Each pouch was tightly closed and stored under refrigeration at a temperature of 2.2 $\pm$ 0.3 °C. Assessments of BAs and amino acids were performed on days 1, 3, 5, 7, and 10 of storage.

2.2. Reagents

Liquid chromatography–mass spectrometry (LC-MS)-grade acetonitrile, hexane, and LC-MS water were supplied by Witko (Łódź, Poland). Disodium tetraborate (borax) $\geq$99% was supplied by Chempur (Piekary Śląskie, Poland). Ammonium formate $\geq$ 97% and formic acid assay 98–100% were purchased from Chem-Lab (Zedelgem, Belgium). Dansyl chloride 97% was acquired from abcr GmbH (Karlsruhe, Germany). Pure trichloroacetic acid was provided by POCH (Gliwice, Poland). Certified analytical standards (Merck, Darmstadt, Germany) included putrescine, histamine, cadaverine, tryptamine, phenethylamine, tyrosine, spermidine $\geq$ 99%, spermine $\geq$99%, agmatine $\geq$ 97%, 1,7-diaminoheptane

assay 98%, and ammonium hydroxide solute arginine, ornithine ≥ 99%, histidine, lysine, tryptophan ≥ 98%, phenylethylalanine, and tyrosine.

2.3. Preparation of Samples for Biogenic Amines and Free Amino Acid Content Analysis

Sample preparation and determination of BAs and AAs were conducted as described by Świder et al. [4]. Two grams of the homogenized meat sample was weighed into a 50 mL centrifuge tube. This was then spiked with 50 µL of the 1,7-diaminoheptane internal standard solution (1 mg × L^{-1}) and with 40 mL of 5% trichloroacetic acid. Subsequently, the sample was shaken and centrifuged at 10,000× *g* for 10 min. The supernatant was then filtered using filter paper. For the derivatization process, 1 mL of distilled water, 1 mL of 5% borax solution, and 100 µL of the sample supernatant were combined in a 15 mL polypropylene tube. Then, 2.5 mL of dansyl chloride (20 mM) dissolved in acetonitrile was added. The tube was thoroughly mixed and then placed in a shaking water bath, maintained at 30 °C for 1 h, ensuring it was kept away from light exposure. After this period, 125 µL of a 400 mM ammonia solution was added to the tube, which was then allowed to sit in a dark environment for an additional 15 min. The mixture was finally passed through a 0.45 µm syringe filter, and the filtrate was transferred to a chromatographic vial, ready for LC-MS analysis.

BA levels were analyzed for putrescine, cadaverine, tyramine, tryptamine, histamine, agmatine, phenethylamine, spermine, and spermidine, as well as their precursors, which included arginine, lysine, histidine, tyrosine, Phenylethylalanine, Tryptophan, and ornithine. The following were also analyzed: BAI—biogenic amines index (sum of putrescine, cadaverine, tyramine, and histamine); TOTAL BA-1—sum of histamine, putrescine, cadaverine, tyramine, phenylethylamine, tryptamine, and agmatine; and TOTAL BA-2—sum of Total BA-1, spermine, and spermidine. For reasons of stability, spermine and spermidine were not included in TOTAL BA-2, but the TOTAL BA-2 index was distinguished. For amino acids, the index sum of precursors of BAI =sum of lysine, histidine, tyrosine, and ornithine was distinguished. All raw results have been included in the database available in Supplementary Materials link.

2.4. Liquid Chromatography–Mass Spectrometry

An ultra-high-performance liquid chromatograph (UPLC) coupled with a high-resolution mass spectrometer Q Exactive Orbitrap Focus MS (Thermo Fisher Scientific, Waltham, MA, USA) were used for analysis. Methods used were according to Świder et al. [4]. The scan was set at Full MS, followed by All Ion Fragmentation mode with scan ranges of 200–1200 m/z and 80–1000 m/z, respectively. The analyses were conducted at a resolution of 70,000 in simultaneous scan and 35,000 in All Ion Fragmentation mode. The Cortecs UPLC C18 2.1 × 100 mm, 1.6 µm column purchased in Waters (Milford, MA, USA) was used. Ions were produced using a heated electrospray ionization (HESI) technique with spray voltage 3 kV. Liquid phases consisted of water/ACN (90:10)/0.1% FA/5 mM ammonium formate (phase A) and ACN/water (90:10)/0.1% FA/5 mM ammonium formate (phase B), which flowed by in a gradient according to the following settings: A:B (%) gradient 0–2 min—90:10—waste, 2–22min—0:100, 22–25min—0:100, 25–26min—90:10, 26–28min—90:10, at the rate of 0.3 mL/min. LC-MS-grade water and acetonitrile were purchased from Witko (Łodź, Poland). Formic acid (98–100%) and ammonium formate (≥97%) for LC-MS were supplied by Chem-Lab (Zedelgem, Belgium). Polarization was set in positive mode, and the volume of the injection was 2.5 µL. The remaining parameters were set as follows: capillary temperature: 256 °C, sheath gas flow rate: 48, auxiliary gas flow rate: 11, sweep gas flow rate: 2, probe heater temperature: 413 °C, S-lens RF level: 50. Xcalibure 4.2.47 software (Thermo Fisher Scientific, Waltham, MA, USA) was used to acquire and analyze data. The applied analytical method was validated to evaluate its statistical parameters in analyses of BAs and AAs, according to Świder et al. [4]. Recovery rates (RRs) of this method were calculated on the basis of results obtained using some spiked samples. In addition, limits of detection (LOD) and limits of quantification (LOQ)

were determined. The values of RRs ranged from 80 to 120%, the LOD was less than 0.1 mg/kg, and the LOQ was less than 0.30 mg/kg. Linearity of the calibration curves was better than 0.99.

2.5. Statistical Analysis

Evaluation of the effects of studied factors (type of meat and storage time) and their interactions was conducted using two-way analysis of variance (ANOVA) and their interactions according to the following linear models:

For one-way ANOVA:

$$Y_{ij} = \mu + A_j + e_{ij} + \text{or } Y_{ik} = \mu + B_k + e_{ik}$$

For two-way ANOVA:

$$Y_{ijk} = \mu + A_j + B_k + (AB)_{jk} + e_{ijk}$$

where Y is a dependent variable, μ is the general mean, A_j is the effect of the meat type, and B_k is the effect of the storage time.

Statistical evaluations were conducted using ANOVA, where p-values were presented along with the pooled standard errors of the means (SEM) for each studied variable. In order to facilitate multiple comparisons of means, one-way ANOVA was utilized in tandem with Duncan's multiple range test. Pearson's correlation coefficients were calculated to evaluate the relationships between selected variables. Beyond these tests, a principal component analysis (PCA) was also carried out to further understand the data. All of these statistical analyses were performed in Statistica 13.3 software. It is important to note that for all the tests and analyses performed, the significance level was set at 0.05 [8].

3. Results

3.1. Effect of Meat Type

Breast and leg muscles differed in histamine ($p < 0.001$), cadaverine ($p < 0.001$), tyramine ($p < 0.001$), BAI index ($p < 0.001$), phenylethylamine ($p = 0.032$), tryptamine ($p < 0.001$), agmatine ($p < 0.001$), spermine ($p < 0.001$), spermidine ($p < 0.001$), TOTAL BA-1 ($p < 0.001$), and TOTAL BA-2 ($p < 0.001$). There was no difference between the analyzed meat types for putrescine content ($p = 0.763$) (Table 1). Table 2 summarizes the precursor amino acids for BA. The effect of meat type was evident for the level of these amino acids: arginine ($p < 0.001$), lysine ($p = 0.008$), histidine ($p < 0.001$), tyrosine ($p < 0.001$), phenylethylalanine ($p < 0.001$), tryptophan ($p < 0.001$), ornithine ($p < 0.001$), and the sum of precursors from BAI ($p < 0.001$).

Table 1. Biogenic amines (mg/100 g of meat) in broiler breast and leg meat stored in air conditions.

	Storage Time (Day)	Item											
		Histamine	**Putrescine**	**Cadaverine**	**Tyramine**	**Index BAI**	**Phenylethylamine**	**Tryptamine**	**Agmatine**	**Spermine**	**Spermidine**	**TOTAL BA-1**	**TOTAL BA-2**
Breast meat	1	1.23 ^{aA}	1.62 ^{A}	0.77 ^{aA}	0.00 ^{A}	3.62 ^{aA}	0.00 ^{aA}	0.00 ^{A}	0.00 ^{A}	77.43 ^{bC}	14.43 ^{aAB}	3.62 ^{aA}	95.47 ^{b}
	3	2.04 ^{B}	2.43 ^{aA}	1.42 ^{aA}	0.00 ^{aA}	5.89 ^{aB}	0.55 ^{D}	0.00 ^{aA}	0.32 ^{B}	73.04 ^{bC}	13.35 ^{bA}	6.76 ^{aB}	93.16 ^{b}
	5	3.09 ^{C}	7.31 ^{B}	1.68 ^{aA}	0.18 ^{aB}	12.27 ^{aC}	0.38 ^{C}	0.85 ^{C}	0.50 ^{C}	62.68 ^{bB}	15.23 ^{bAB}	13.99 ^{aC}	91.91 ^{b}
	7	5.28 ^{D}	11.09 ^{C}	2.72 ^{aB}	0.28 ^{aB}	19.37 ^{aD}	0.19 ^{B}	0.44 ^{B}	0.60 ^{aC}	50.74 ^{bA}	16.79 ^{bB}	20.60 ^{aD}	88.13 ^{b}
	10	8.77 ^{aE}	14.42 ^{D}	4.91 ^{aC}	0.73 ^{aC}	28.83 ^{aE}	0.35 ^{C}	0.43 ^{aB}	0.93 ^{aD}	47.51 ^{bA}	16.64 ^{bAB}	30.55 ^{aE}	94.69 ^{b}
Leg meat	1	1.62 ^{bA}	1.94 ^{A}	2.01 ^{bA}	0.00 ^{A}	5.57 ^{bA}	0.22 ^{bA}	0.00 ^{A}	0.00 ^{A}	45.05 ^{aC}	17.19 ^{bC}	5.79 ^{bA}	68.03 ^{aC}
	3	2.10 ^{A}	3.15 ^{bB}	2.54 ^{bAB}	0.28 ^{bB}	8.07 ^{bB}	0.51 ^{B}	0.25 ^{bB}	0.35 ^{B}	43.81 ^{aC}	11.17 ^{aB}	9.19 ^{bB}	64.16 ^{aC}
	5	3.32 ^{B}	6.78 ^{C}	3.42 ^{bB}	0.40 ^{bB}	13.93 ^{bC}	0.49 ^{B}	0.71 ^{C}	0.63 ^{C}	27.62 ^{aB}	4.55 ^{aA}	15.75 ^{bC}	47.92 ^{aA}
	7	5.94 ^{C}	11.95 ^{D}	5.43 ^{bC}	0.96 ^{bC}	24.27 ^{bD}	0.31 ^{AB}	0.55 ^{C}	0.85 ^{bD}	25.01 ^{aB}	4.28 ^{aA}	25.99 ^{bD}	55.28 ^{aB}
	10	11.10 ^{bD}	13.46 ^{E}	12.81 ^{bD}	1.62 ^{bD}	38.98 ^{bE}	0.30 ^{AB}	1.27 ^{bD}	1.38 ^{bE}	18.03 ^{aA}	4.73 ^{aA}	41.94 ^{bE}	64.70 ^{aC}
Pooled SEM		0.78	1.80	1.33	0.02	4.72	0.03	0.05	0.03	48.58	6.62	4.88	68.13
Main effects		*p*-value											
Effect of meat type		<0.001	0.763	<0.001	<0.001	<0.001	0.032	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Effect of storage time		<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Effect of meat type x storage time		0.001	0.144	<0.001	<0.001	<0.001	0.064	<0.001	0.001	0.284	<0.001	<0.001	0.016

^{a,b}—small letters indicate significant differences between meat types within the same storage time, $p \leq 0.05$; ^{A,B,C,D,E}—capital letters indicate significant differences between storage times within the same meat type, $p \leq 0.05$ (one-way ANOVA, Duncan test); index BAI—sum of histamine, putrescine, cadaverine, and tyramine; TOTAL BA-1—sum of histamine, putrescine, cadaverine, tyramine, phenylethylamine, tryptamine, and agmatine; TOTAL BA-2—sum of Total BA-1, spermine, and spermidine.

Table 2. Precursors of biogenic amines—amino acids (g/100 g of meat) in broiler breast and leg meat stored in air conditions.

	Storage Time (Day)	Item							
		Arginine	**Lysine**	**Histidine**	**Tyrosine**	**Phenylethylalanine**	**Tryptophan**	**Ornithine**	**Sum of Precursors BAI**
Breast meat	1	1.473 ^{b}	1.569	0.809 ^{b}	0.644	0.782 ^{b}	0.204 ^{b}	0.031 ^{b}	3.053 ^{b}
	3	1.442 ^{b}	1.448	0.769 ^{b}	0.655 ^{b}	0.729 ^{b}	0.190	0.032 ^{b}	2.904 ^{b}
	5	1.471 ^{b}	1.557	0.769 ^{ba}	0.634 ^{b}	0.762 ^{b}	0.192	0.031 ^{b}	2.991 ^{b}
	7	1.487 ^{b}	1.518	0.817 ^{b}	0.623 ^{b}	0.736 ^{b}	0.201	0.031 ^{b}	2.989 ^{b}
	10	1.485 ^{b}	1.519	0.793 ^{b}	0.645 ^{b}	0.786 ^{b}	0.200	0.030 ^{b}	2.987 ^{b}
Leg meat	1	1.039 ^{a}	1.445	0.583 ^{a}	0.570 ^{B}	0.663 ^{a}	0.175 ^{a}	0.023 ^{a}	2.622 ^{a}
	3	1.044 ^{a}	1.412	0.584 ^{a}	0.530 ^{aAB}	0.643 ^{a}	0.170	0.024 ^{a}	2.550 ^{a}
	5	1.023 ^{a}	1.421	0.592 ^{a}	0.510 ^{A}	0.646 ^{a}	0.175	0.025 ^{a}	2.548 ^{a}
	7	1.022 ^{a}	1.390	0.568 ^{a}	0.508 ^{aA}	0.641 ^{a}	0.168	0.026 ^{a}	2.492 ^{a}
	10	1.039 ^{a}	1.458	0.561 ^{a}	0.532 ^{aAB}	0.667 ^{a}	0.175	0.026 ^{a}	2.578 ^{a}
Pooled SEM		0.014	0.032	0.011	0.005	0.008	0.001	0.0001	0.046
Main effects						*p*-value			
Effect of meat type		<0.001	0.008	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Effect of storage time		0.987	0.662	0.957	0.308	0.480	0.866	0.704	0.523
Effect of meat type x storage time		0.920	0.861	0.768	0.753	0.963	0.941	0.464	0.882

^{a,b}—small letters indicate significant differences between meat typeswithin the same storage time, $p \leq 0.05$; ^{A,B}—capital letters indicate significant differences between storage times within the same meat type, $p \leq 0.05$ (one-way ANOVA, Duncan test); sum of precursors of BAI = sum of lysine, histidine, tyrosine, and ornithine.

3.2. Effect of Storage Time

Table 1 shows the variations in biogenic amine content in breast and leg muscles over time. The effect of storage time was evident in the contents of histamine ($p < 0.001$), putrescine ($p < 0.001$), cadaverine ($p < 0.001$), tyramine ($p < 0.001$), BAI index ($p < 0.001$), phenylethylamine ($p < 0.001$), tryptamine ($p < 0.001$), agmatine ($p < 0.001$), spermine ($p < 0.001$), spermidine ($p < 0.001$), TOTAL BA-1 ($p < 0.001$), and TOTAL BA-2 ($p < 0.001$). However, storage time did not influence the amino acid content in either breast or leg muscles ($p > 0.05$) (Table 2).

3.3. Interaction Meat Type × Storage Time

An interaction between both factors (meat type and storage time) was observed for levels of histamine ($p = 0.001$), tyramine ($p < 0.001$), BAI index ($p < 0.001$), tryptamine ($p < 0.001$), agmatine ($p = 0.001$), spermidine ($p < 0.001$), TOTAL BA-1 ($p < 0.001$), and TOTAL BAs ($p = 0.016$) (Table 1). As storage time progressed, there was an increase in BA levels, while AA levels remained relatively consistent. For AA content in breast and leg muscles, no interaction between the meat type and storage time was detected ($p > 0.05$) (Table 2).

3.4. Correlations and Principal Component Analysis (PCA)

PCA was carried out to determine the changes in BA levels and their precursor amino acids over meat storage time. Within the PCA framework, the data from each amino acid and the BA content at every analysis interval were transformed into two new orthogonal variables, denoted as "principal components" (PC1 and PC2). The relationships between the parameters and the PCs were interpreted according to the correlations between them. Figure 1 shows the PCA for breast muscles (A) and leg muscles (B). For the breast muscles (Figure 1A), PC1 and PC2 account for 79.73% of the variability in BA levels and their precursors during the evaluated periods, leading to a 20.27% data loss. The primary component (PC1) was positively correlated with spermine, TOTAL BA-2, tyrosine, ornithine, and phenylethylamine levels while showing an inverse relationship with the rest of the BAs and AAs present in the breast muscle. Conversely, the secondary component (PC2) was negatively correlated with TOTAL BA-2, spermine, spermidine, arginine, phenylethylamine, histidine, tryptophan, and the sum of precursors of BAI and positively correlated with the remaining amines and amino acids. In the context of leg muscles (Figure 1B), PC1 and PC2 capture 86.23% of the variance observed in the levels of BAs and their precursors at the measured intervals, resulting in a 13.77% information deficit. Concentrations of spermine, spermidine, TOTAL BA-2, arginine, tyrosine, histidine, phenylethylamine, and the sum of precursors of BAI were positively correlated with PC1. Meanwhile, PC2 is positively correlated with levels of ornithine, putrescine, histidine, and phenylethylamine.

Table 3 summarizes the correlations between BAs and precursor AAs. In the breast muscle, a negative correlation was observed between ornithine and putrescine ($r = -0.57$) ($p < 0.05$) and between ornithine and spermidine ($r = -0.73$) ($p < 0.05$). A negative correlation was also found for phenylethylamine and its precursor ($r = -0.50$) ($p < 0.05$). The content of phenylethylamine in the breast muscles negatively correlated with the level of tryptophan ($r = -0.90$) and with the sum of the precursors of BAI ($r = -0.92$) ($p < 0.05$). In leg muscles, correlations were noted for histamine and its precursor histidine ($r = -0.87$) ($p < 0.05$) and between putrescine and ornithine ($r = -0.96$) ($p < 0.05$). Ornithine in leg muscles also correlated with spermine ($r = -0.95$), spermidine ($r = -0.95$), and TOTAL BA-2 ($r = -0.90$) ($p < 0.05$). A negative correlation was again found for phenylethylamine and its precursor ($r = -0.65$) ($p < 0.05$). Furthermore, the analysis revealed positive correlations for ornithine with tryptamine ($r = 0.89$) and agmatine ($r = 0.95$), and for arginine with TOTAL BA-2 levels ($r = 0.89$) ($p < 0.05$).

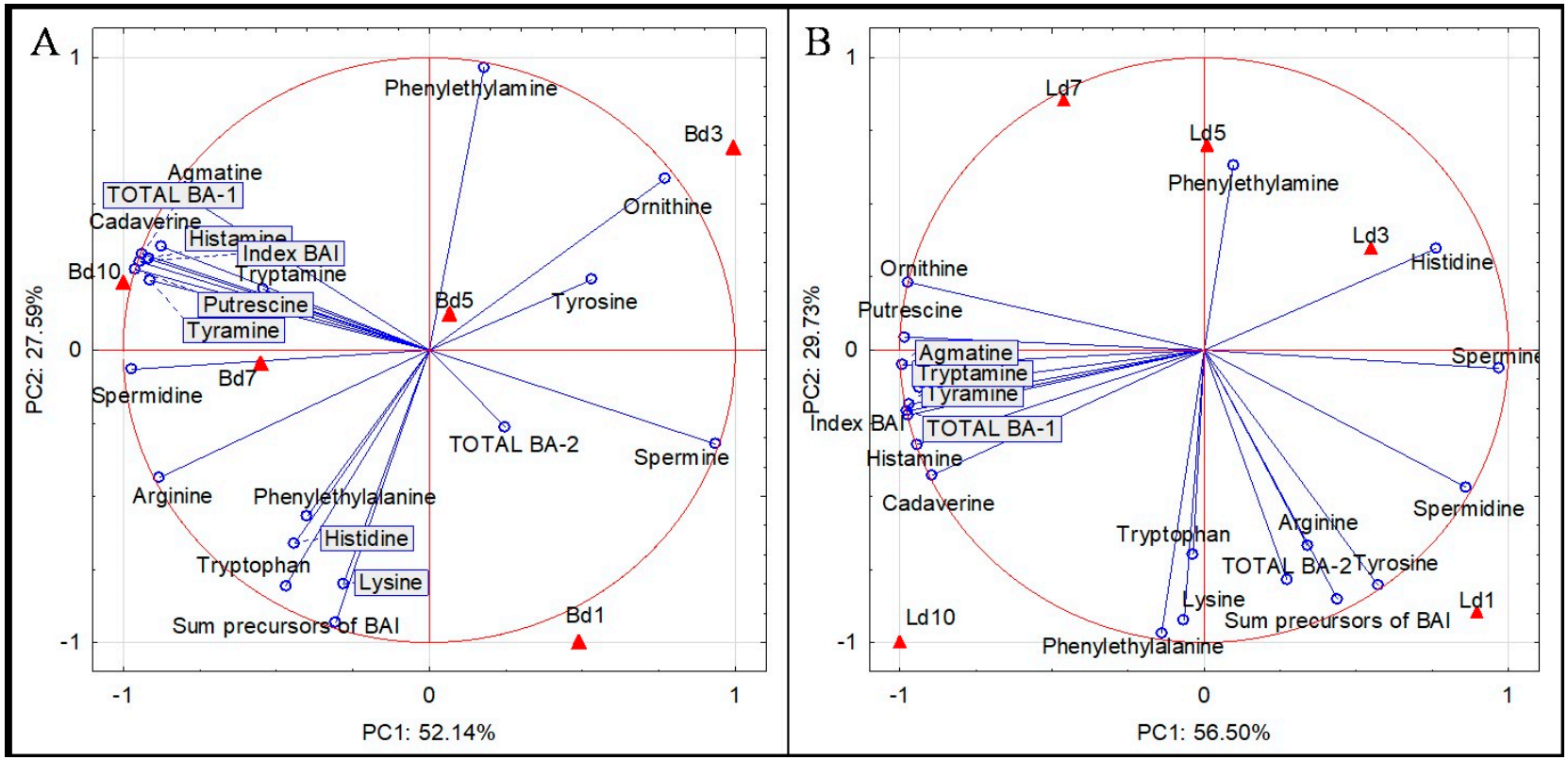


Figure 1. First two components of PCA for biogenic amines and their precursors in breast (**A**) and leg (**B**) chicken meat during storage.

Table 3. Correlation between biogenic amines and their precursors in broiler breast and leg meat.

		Histidine	Ornithine	Lysine	Tyrosine	Phenylethylalanine	Tryptophan	Arginine	Sum Precursors of BAI
Breast meat	Histamine	−0.21	−0.58	−0.07	−0.22	0.29	0.24	0.63	−0.03
	Putrescine	0.24	−0.57 *	0.05	−0.46	0.21	0.23	0.73	0.04
	Cadaverine	0.16	−0.55	−0.13	−0.12	0.30	0.20	0.56	−0.08
	Tyramine	0.17	−0.64	0.04	−0.16	0.44	0.26	0.64	0.07
	Index BAI	0.22	−0.58	−0.01	−0.33	0.26	0.23	0.68	0.01
	Phenylethylamine	−0.82	0.68	−0.76	0.41	−0.50 *	−0.90 *	−0.60	−0.92 *
	Tryptamine	−0.22	−0.18	0.39	−0.62	0.11	−0.20	0.47	0.11
	Agmatine	−0.05	−0.34	−0.18	−0.25	0.09	−0.06	0.48	−0.25
	Spermine	−0.26	0.50	0.01	0.53	−0.08	−0.18	−0.71	0.03
	Spermidine	0.54	−0.73	0.32	−0.71	0.27	0.50	0.92 *	0.35
	TOTAL BA-1	0.19	−0.56	−0.02	−0.34	0.24	0.19	0.67	−0.02
	TOTAL BA-2	−0.22	−0.18	0.15	0.79	0.68	0.13	−0.26	0.22
Leg meat	Histamine	−0.87 *	0.84	0.34	−0.28	0.43	0.21	−0.05	−0.16
	Putrescine	−0.80	0.96 *	−0.03	−0.57	0.09	−0.05	−0.41	−0.50
	Cadaverine	−0.84	0.77	0.46	−0.18	0.52	0.29	0.06	−0.03
	Tyramine	−0.87	0.90 *	0.19	−0.40	0.28	0.06	−0.11	−0.30
	Index BAI	−0.86	0.89 *	0.24	−0.37	0.34	0.14	−0.15	−0.26
	Phenylethylamine	0.52	0.07	−0.40	−0.57	−0.65 *	−0.31	−0.07	−0.39
	Tryptamine	−0.60	0.89 *	0.30	−0.48	0.28	0.29	−0.21	−0.22
	Agmatine	−0.75	0.95 *	0.14	−0.54	0.18	0.07	−0.23	−0.38
	Spermine	0.62	−0.95 *	−0.06	0.61	−0.12	−0.15	0.51	0.42
	Spermidine	0.38	−0.95 *	0.30	0.89 *	0.30	0.16	0.64	0.72
	TOTAL BA-1	−0.85	0.90 *	0.24	−0.39	0.33	0.14	−0.16	−0.27
	TOTAL BA-2	−0.33	−0.44	0.54	0.82	0.61	0.15	0.89 *	0.64

* The correlation is significant at $p \leq 0.05$; sum of precursors of BAI = sum of lysine, histidine, tyrosine, andornithine; index BAI—sum of histamine, putrescine, cadaverine, and tyramine; TOTAL BA-1—sum of histamine, putrescine, cadaverine, tyramine, phenylethylamine, tryptamine, and agmatine; TOTAL BA-2—sum of Total BA-1, spermine, and spermidine.

4. Discussion

The main objective of this study was to determine the development of changes in BA levels and their precursor amino acids in chicken breast and leg muscles during cold storage. Additionally, the study sought to understand the correlation between BAs and AAs. With the exception of spermine and spermidine, BAs increased as the storage time lengthened. Similar results were also obtained by other authors [9–12]. This is due to progressive proteolysis and increasing levels of free amino acids, which act as precursors for individual BA formation. While correlations between individual precursor amino acids and BAs have not been studied extensively, Triki et al. identified a trend in chicken meat, pointing to a correlation between BA and AA levels [11]. In their research on pickled vegetables, Świder et al. demonstrated that elevated AA levels influenced the formation of BAs in a BA-specific manner [4]. The results of this study suggest a relationship between BAs and their precursor amino acids. It is possible that the mechanisms dictating the formation of specific BAs, in the context of changes in individual amino acid levels, might vary. Leg muscles, which have a higher fat content, a lower protein level, and a slightly varied amino acid composition compared to breast muscles, might influence the synthesis of individual BAs. The authors showed that BA levels in leg muscles were markedly higher than in breast muscles. The different chemical and amino acid compositions, as well as different rates of muscle processes, lead to different correlation values between the AA precursor–BA pairs [13,14]. What is also possibly related to the different activity of the breast and leg muscles during the life of the birds, as well as the different structure of the muscle fibers, are the proteolytic processes taking place in them. In the future, it will be important to find out what factors influence the different rate of BA formation, which not only lowers the meat product itself but also the biological value of the meat protein.

5. Conclusions

The results obtained can be used to confirm correlations in the future and can be used to estimate BA levels with knowledge of the individual amino acid levels and vice versa. This is important not only for cognitive reasons but also for dietary and pro-health reasons. The estimation of BA levels may be important for people taking agents that are MAO and DAO inhibitors. The present study is only concerned with changes occurring in ROSS 308 meat chickens. In the future, it would be worthwhile to carry out similar analyses on other genotypes and species of poultry used for meat production.

Supplementary Materials: https://figshare.com/articles/dataset/Raw_datebase/24409453/1 accessed on 26 October 2023.

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References

1. Feddern, V.; Mazzuco, H.; Fonseca, F.N.; De Lima, G.J.M.M. A review on biogenic amines in food and feed: Toxicological aspects, impact on health and control measures. *Anim. Prod. Sci.* **2019**, *59*, 608–618. [CrossRef]
2. Nuñez, M.; del Olmo, A.; Calzada, J. Biogenic Amines. *Encycl. Food Health* **2016**, 416–423. [CrossRef]
3. Park, Y.K.; Lee, J.H.; Mah, J.H. Occurrence and reduction of biogenic amines in traditional Asian fermented soybean foods: A review. *Food Chem.* **2019**, *278*, 1–9. [CrossRef]
4. Świder, O.; Roszko, M.Ł.; Wójcicki, M.; Szymczyk, K. Biogenic Amines and Free Amino Acids in Traditional Fermented Vegetables-Dietary Risk Evaluation. *J. Agric. Food Chem.* **2020**, *68*, 856–868. [CrossRef] [PubMed]
5. Gallas, L.; Standarová, E.; Steinhauserová, I.; Steinhauser, L.; Vorlová, L. Formation of Biogenic Amines in Chicken Meat Stored under Modified Atmosphere. *Acta Vet. Brno* **2010**, *79*, 107–116. [CrossRef]
6. Dalle Zotte, A.; Ricci, R.; Cullere, M.; Serva, L.; Tenti, S.; Marchesini, G. Research Note: Effect of chicken genotype and white striping–wooden breast condition on breast meat proximate composition and amino acid profile. *Poult. Sci.* **2020**, *99*, 1797–1803. [CrossRef] [PubMed]
7. Salinas, Y.; Ros-Lis, J.V.; Vivancos, J.L.; Martínez-Máñez, R.; Marcos, M.D.; Aucejo, S.; Herranz, N.; Lorente, I. Monitoring of chicken meat freshness by means of a colorimetric sensor array. *Analyst* **2012**, *137*, 3635–3643. [CrossRef] [PubMed]
8. TIBCO Software Inc. *Statistica (Data Analysis Software System)*, version 13; TIBCO Software Inc.: Sydney, NSW, Australia, 2017. Available online: http://statistica.io(accessed on 15 September 2023).
9. Saewan, S.A.; Khidhir, Z.K. 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]
10. Chmiel, M.; Roszko, M.; Hać-Szymańczuk, E.; Cegiełka, A.; Adamczak, L.; Florowski, T.; Pietrzak, D.; Bryła, M.; Świder, O. Changes in the microbiological quality and content of biogenic amines in chicken fillets packed using various techniques and stored under different conditions. *Food Microbiol.* **2022**, *102*, 103920. [CrossRef] [PubMed]
11. Triki, M.; Herrero, A.M.; Jiménez-Colmenero, F.; Ruiz-Capillas, C. Quality assessment of fresh meat from several species BAed on free amino acid and biogenic amine contents during chilled storage. *Foods* **2018**, *7*, 132. [CrossRef] [PubMed]
12. Min, J.S.; Lee, S.O.; Jang, A.; Jo, C.; Lee, M. Control of microorganisms and reduction of biogenic amines in chicken breast and thigh by irradiation and organic acids. *Poult. Sci.* **2007**, *86*, 2034–2041. [CrossRef] [PubMed]
13. Pleva, D.; Lányi, K.; Monori, K.D.; Laczay, P. Heterocyclic amine formation in grilled chicken depending on body parts and treatment conditions. *Molecules* **2020**, *25*, 1547. [CrossRef] [PubMed]
14. Esposito, L.; Mastrocola, D.; Martuscelli, M. Approaching to biogenic amines as quality markers in packaged chicken meat. *Front. Nutr.* **2022**, *9*, 966790. [CrossRef] [PubMed]

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