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## Characterization of meat quality, storage stability, flavor-related compounds, and their relationship in Korean Woorimatdag No. 2 chicken breast meat during cold storage

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#### ABSTRACT

We evaluated the quality, storage stability, and flavor-related compounds of breast meat from a novel Korean native chicken breed (Woorimatdag No. 2; WRMD2) and commercial broiler (CB) during seven days of aerobic cold storage. We found that pH and drip loss increased gradually during storage and WRMD2 exhibited a significantly lower pH and higher drip loss than CB. In both groups, aerobic plate counts, volatile basic nitrogen, and lipid oxidation levels increased, whereas creatine and dipeptide levels gradually decreased during storage. WRMD2 exhibited a significantly higher anserine content and lower carnosine-to-anserine ratio than CB. Flavor nucleotide content was influenced more by the storage period, whereas fatty acid composition was affected more by genetic differences. WRMD2 exhibited significantly higher levels of polyunsaturated fatty acids, especially C20:4n6 and C22:6n3, than CB. Interestingly, multivariate analysis highlighted several volatile compounds, including methyl salicylate (day 1), dodecanal (day 3), naphthalene (day 5), and 2,4-decadienal (day 7) as potential biomarkers to distinguish between WRMD2 and CB on each storage day. Correlation analysis identified five key meat quality traits, including drip loss, aerobic plate counts, and anserine that are strongly associated with flavor substances, such as inosine monophosphate, guanosine monophosphate, 1-octen-3-ol, and hexanal. These results offer insights into the distinctive meat quality and flavor compounds in WRMD2 during storage, providing fundamental data that could improve the management and quality of native chicken meat.

### Introduction

Chicken meat is a healthy dietary option owing to its high protein content, nutritional value, bioactive compounds (e.g., creatine and dipeptide), and affordable price (Kim et al., 2020). As the global demand for high-quality meat continues to grow, consumers are increasingly prioritizing high standards for selecting their chicken meat (Lee et al., 2022). In South Korea, Korean native chicken (KNC) has been traditionally consumed on special occasions as a premium chicken meat given its superior taste and texture. Currently, several highly-bred KNCs dominate the KNC meat market. These include the Hanhyup, Sorae, and Woorimatdag breeds, which are the outcomes of decades of selective

breeding by the South Korean government or companies to achieve specific commercial goals, such as growth performance and organoleptic properties. In 2012, the South Korean government launched the 'Golden Seed Project' to discover and conserve genetic resources, while aiming to establish the sustainable production of KNC and maintain the genetic biodiversity of breeds (Jin et al., 2017). Accordingly, the breed Woorimatdag No. 1 was developed by the National Institute of Animal Science in South Korea through three-way-cross-mating of original and adapted KNCs with the aim of enhancing both taste quality and affordability (Jung et al., 2024). This breed contains high levels of taste-related and bioactive compounds; yet it has demonstrated low growth performance, thereby making it challenging to meet market demand (Barido et al.,

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2022). Therefore, Woorimatdag No. 2 (WRMD2) was developed via crossbreeding of Brown KNC/Rhode Island Red females with Black Cornish/Brown Cornish males. This resulted in a shortened rearing period while maintaining taste and nutritional value of KNC (Kim and Kang, 2014).

The quality and freshness of meat and meat products affect consumer preferences. These traits can be represented as a set of parameters, including safety, nutritional value, functionality, flavor, and organoleptic characteristics. Fresh chicken meat is highly perishable owing to its substantial protein and water activity, which limits its shelf-life regardless of cold storage (Katiyo et al., 2020). During refrigerated storage, meat may deteriorate, which leads to decreased freshness due to the loss of nutrients and moisture, proliferation of spoilage microorganisms, lipid oxidation, and accumulation of undesirable volatile compounds (Sujiwo et al., 2018). Hence, the analysis and prediction of meat quality changes during storage can help to assure sensory, taste, nutritional, and safety standards of chicken meat products. The most common methods used to evaluate the freshness of meat products are chemical, physical, and microbiological analyses, which have long been used both individually and in combination (Pereira et al., 2021). From this perspective, numerous studies have monitored changes in chicken meat quality and freshness during cold storage. These studies analyzed bioactive compounds, fatty acids (Kim et al., 2020), lipid and protein oxidation, microbial changes (Sujiwo et al., 2018), nucleotide-related products (Lee et al., 2022), and volatile organic compounds (VOCs) (Chmiel et al., 2020). Although these studies primarily focused on commercial broilers (CB), several attempts have been made to evaluate changes in the stored meat quality of various indigenous chicken breeds in South Korea (Utama et al., 2016), China (Lv et al., 2023), Ireland (Alexandrakis et al., 2012), and Bangladesh (Ali et al., 2022). However, to the best of our knowledge, detailed information on the quality characteristics that underwent alterations during cold storage, of novel KNC breeds, especially WRMD2, has not been sufficiently explored.

In this study, we aimed to fill gaps in our current understanding by investigating the changes in breast meat quality, storage stability, and flavor-related compounds of WRMD2 and CB during cold storage. We hypothesized that meat obtained from different chicken breeds would exhibit distinct quality and flavor characteristics that could serve as potential indicators to differentiate native chickens from CB during cold storage. Additionally, correlation analysis was conducted to explore the relationship between meat quality and flavor substances, enhancing our understanding of how genetic differences in chickens contribute to their meat quality and resulting flavor profiles. Our results may provide a baseline for developing strategies to enhance and optimize meat quality and flavor in the native chicken industry.

## Materials and methods

## Birds and sampling

This study followed the guidelines for the use of experimental animals of the National Institute of Animal Science (Wanju, South Korea, NIAS-2021-0519). We obtained market-aged male WRMD2 (n=72, 1,550–1,650 g, 12-wk-old) and CB (n=72, 1,150–1,250 g, 6-wk-old) chickens from the National Institute of Animal Science for the study. WRMD2 and CB are typically slaughtered at 12 weeks and 6–7 weeks of age, respectively, in South Korea.

All birds were raised on an environmentally-controlled farm (Jirisan Black Farm, Sancheong, South Korea) and were fed a commercial diet (metabolic energy, 2,500 kcal/kg; crude protein, 15.0 %; calcium, 3.60 %; phosphorus, 0.48 %), according to the Korean Feeding Standards for Poultry (2022). For slaughter, birds were transported for approximately 100 min from the farm to the commercial poultry slaughterhouse (Dasol Co., Jangheung, South Korea). Chickens were stunned with 65–80 % CO<sub>2</sub> gas and slaughtered following the Livestock Products Sanitary Control Act of Korea. After slaughter, chicken carcasses were

immediately transported for approximately 4 h (400 km) via refrigerated trucks (2  $\pm$  1 °C) to a walk-in refrigerator (4  $\pm$  2 °C) in the laboratory. The left side of the chicken breast (pectoralis major) was separated without skin and visible fat. The breast meat was then directly packaged using polystyrene trays and low-density polyethylene film (oxygen transmission rate, 35,273 cc/(m²-day-atm)), and subsequently stored at 4 °C for 7 days. Of the 72 breast meat samples, 18 were randomly selected on each sampling day (days 1, 3, 5, and 7). Of these 18 breast meat samples, 12 were randomly collected to create six replicates by randomly pooling two pieces of breast meat. The remaining six samples were subjected to shear force analysis. The proximate composition of WRMD2 and CB on the first day of storage are shown in Supplementary Table S1.

Physicochemical characteristics assessment

pI

A 3g sample with 27 mL distilled water was homogenized (Polytron PT-2500E; Kinematica, Lucerne, Switzerland) (15 s, 12,000 rpm) and analyzed using a pH meter (Orion 230A, Thermo Fisher Scientific, MA, USA).

#### Drip loss

Drip loss was calculated as the percentage weight difference between the initial weight and the final weight at each storage period (Zheng et al., 2023).

#### Water-holding capacity (WHC)

The WHC was evaluated as described by Kim et al. (2020). A 0.5 g sample was placed in a tube (Millipore Ultrafree-MC; Millipore, MA, USA), heated in a water bath (20 min, 80 °C), cooled (10 min, 23  $\pm$  1 °C), and centrifuged (10 min, 4 °C, 2,000×g) to measure water loss and the WHC (%) was calculated using following equation:

$$\textit{Water loss} = \frac{(\textit{Weight before centrifugation} - \textit{Weight after centrifugation})}{\textit{Sample Weight} \ \times \ (1 - (\textit{crude fat\%/100})} \\ \times 100$$

 $\textit{WHC} = (\textit{Moisture content\%} - \textit{Water loss}) / (\textit{Moisture content\%}) \times 100$ 

## Instrumental color

The instrumental color was measured with a Chroma Meter CR-400 instrument (Minolta, Osaka, Japan) using CIE L\*, CIE a\*, and CIE b\* after calibration (Y, 93.60; x, 0.3134; y, 0.3194).

## Warner-Bratzler shear force (WBSF)

A TA1 texture analyzer (Lloyd Instruments, Berwyn, IL, USA) combined with a Warner–Bratzler blade (load cell, 500 N; cross-head speed, 50 mm/min) was used to determine the WBSF, as described by Jung et al. (2023).

Storage stability and bioactive metabolites assessment

#### Microorganism

A 3 g sample was homogenized with 27 mL of sterile saline solution for 45 s using a Bag Mixer 400 stomacher (Interscience, Saint-Nom-la-Bretèche, France). Aerobic plate counts (APC) and coliforms were determined using 3M Petrifilms (3M Company, MN, USA) after incubation (48 h, 37  $^{\circ}\text{C}$ ) and the results are expressed as log CFU/g.

## 2-Thiobarbituric acid reactive substances (TBARS)

The TBARS content was evaluated as described by Jung et al. (2024). A 5 g sample was homogenized with 15 mL distilled water and 50  $\mu$ L 7.2 % butylated hydroxyanisole (30 s, 12,000 rpm). Subsequently, 2 mL 20 mM thiobarbituric acid in 15 % trichloroacetic acid was added to 1 mL

homogenate, heated (15 min, 90 °C), cooled (10 min, 23  $\pm$  1 °C), and centrifuged (10 min, 4 °C, 2,000×g). The absorbance of the supernatant was determined at 531 nm using a spectrophotometer (M2e, Molecular Devices, CA, USA), and the results are presented as mg malondialdehyde (MDA)/kg of sample.

#### Total volatile basic nitrogen (VBN)

The VBN content was determined using the micro-diffusion method (Kim et al., 2020). A 10 g sample was homogenized with 50 mL distilled water using a magnetic stirrer for 30 min at 4 °C. The homogenate was filtered using Whatman No. 1 filter paper, and 1 mL filtrate was added to the outer chamber of a Conway micro-diffusion cell. Then 1 mL 0.01 N  $\rm H_2SO_4$  and 1 mL saturated  $\rm K_2CO_3$  were added to inner and outer cells, respectively. The cell was covered and incubated (60 min, 25 °C). Afterwards, 10  $\mu L$  Brunswick reagent was added to the inner cell, titrated with 0.01 N NaOH, and the results are expressed as mg/100 g.

#### Bioactive metabolites

Bioactive metabolites were extracted from the samples, as described by Kim et al. (2020). An Atlantis HILIC silica column (150  $\times$  4.6 mm, 3.0  $\mu m$ ; Waters, MA, USA) equipped with an HPLC system (Agilent Infinity 1260, Agilent Technologies, CA, USA) was utilized to determine the bioactive metabolites. The creatinine content was determined at 236 nm, and creatine and dipeptide contents were assayed at 214 nm, using their respective standards (Sigma-Aldrich, MO, USA). Mobile phases consisted of solvent A (pH 5.5, 0.65 mM ammonium acetate in water: acetonitrile, 25:75, v/v) and solvent B (pH 5.5, 4.55 mM ammonium acetate in water: acetonitrile, 70:30, v/v). Solvent B was supplied at a linear gradient (0 to 100 %) at 1.4 mL/min for 13 min.

#### Flavor-related compounds assessment

#### Fatty acid composition

Lipid extraction and derivatization were performed as described by Barido et al. (2022). An Agilent 7890N gas chromatograph (Agilent Technologies) equipped with an Omegawax 250 capillary column (30 m  $\times$  0.25 mm  $\times$  0.25 µm; Supelco, PA, USA) was utilized to analyze the fatty acid composition. Carrier gas, flow rate, and split ratio were helium, 1.2 mL/min, and 1:100, respectively. The injection port and flame ionization detector temperatures were 250 °C and 260 °C, respectively. The column temperature program was as follows: start at 150 °C for 2 min, then increase gradually to 220 °C at 4 °C/min and hold for 30 min. Fatty acids were identified using their respective standards (PUFA No. 2-Animal Source, Supelco).

## Flavor nucleotides

Flavor nucleotides were extracted as described by Jung et al. (2023). The HPLC (Agilent 1260 Infinity, Agilent Technologies) equipped with a Nova-pak C18 column (150  $\times$  3.9 mm, 4- $\mu$ m particles, Waters) eluting 1 % trimethylamine · phosphoric acid (pH 6.5) at a 1.0 mL/min flow rate was employed to quantify the flavor nucleotides. Injection volume, running time, and column temperature were 10  $\mu$ L, 30 min, and 40  $^{\circ}$ C, respectively. Nucleotide content was determined at 254 nm using adenosine triphosphate, adenosine diphosphate, adenosine monophosphate (AMP), inosine monophosphate (IMP), guanosine monophosphate (GMP), hypoxanthine, and inosine standards (Sigma-Aldrich).

## Volatile organic compounds

VOCs extraction was conducted using the headspace solid-phase micro-extraction method (Jung et al., 2024). Gas-chromatography mass-spectrometry (Agilent 8890 and 5977B, Agilent Technologies) with a DB-5MS column (30 m  $\times$  0.25 mm  $\times$  0.25 µm, Agilent Technologies) was utilized to identify the VOCs. The carrier gas and flow rate were helium and 1.3 mL/min, respectively. The analysis was operated in spitless mode at 250 °C for 5 min. The oven temperature was set at 40 °C

for 5 min, programmed at 5 °C/min to reach 250 °C, and maintained for 5 min. The interface temperature was set at 280 °C. The mass spectrometer was operated in electron impact mode with an electron energy of 70 eV and a scan range of 30–300 m/z (scan rate: 4.37 scans/s, gain factor:1, resulting EM voltage:1140 V). The temperature of the mass spectrometer source and quadrupole was set at 230 and 150 °C, respectively. VOCs detected in more than four of six replicates were selected for analysis. The values are expressed as the sum of the characteristic anion abundances of each component (area unit: AU  $\times$   $10^5$ ). Flavor descriptions of the VOCs were obtained using the method described by Jung et al. (2024). Every detected VOCs (including the mass-to-charge value and linear retention index) and its flavor descriptors are listed in Supplementary Tables S2 and S3, respectively.

## Statistical analysis

Mean values and standard deviations were obtained from six replicates analyses. Statistical analyses were conducted using one-way analysis of variance with a general linear model (SAS 9.4, SAS Institute Inc., NC, USA). Significant differences between mean values were evaluated using Tukey's test at a significance level of 0.05. For VOCs, partial least squares-discriminant analysis (PLS-DA) and correlation analysis were conducted using Metaboanalyst 6.0 (www.metaboanalyst. ca) using log-transformation and auto-scaling prior to multivariate and correlation analyses.

#### Results and discussion

Physicochemical characteristics of WRMD2 and CB

pΕ

The pH value of WRMD2 (5.67–5.78) was significantly lower than that of CB (6.04–6.18) (Table 1). This outcome is consistent with previous observations (Choe et al., 2010; Jeon et al., 2010). A strong genetic correlation between chicken breast and ultimate pH has been well documented (Le Bihan-Duval et al., 2008). Meanwhile, the pH of CB and WRMD2 increased gradually from day 1 to its maximum level on day 7 (P < 0.05). The observed increase in pH during storage could be attributed to the accumulation of amines and ammonia resulting from the proteolytic decomposition of muscle proteins via microbial actions and endogenous enzymes (Lee et al., 2022). The pH of fresh chicken meat ranges from 5.69 to 6.13 (Sujiwo et al., 2018). Herein, the pH of CB and WRMD2 met the reported criteria, except for CB on day 7. As reported by Sujiwo et al. (2018), meat spoilage may begin when the pH exceeds 6.20. However, throughout all storage periods, neither CB nor WRMD2 exhibited a pH above this threshold.

## Drip loss

Drip loss is a key factor from a commercial perspective because low drip loss indicates a high WHC of meat products, which can consequently enhance tenderness and visual acceptability upon consumption (Zheng et al., 2023). The drip loss increased progressively during storage for both CB and WRMD2 (Table 1). Specifically, the drip loss of CB showed a significant increase on days 1-3 (0.17 %-0.98 %) and reached maximum levels on day 7 (1.81 %). Similar trends were observed for WRMD2, where the drip loss increased significantly on each storage day. Previous studies have reported an increase in drip loss in chicken meat during cold storage (Ab Aziz et al., 2020; Zheng et al., 2023). Multiple factors affect the moisture loss in raw meat including myofibril shrinkage, interfilamentous spacing, protein degradation, cell membrane permeability to water, and drip channel formation (Hughes et al., 2014). Interestingly, WRMD2 exhibited a significantly higher drip loss than CB, with an approximately 6-fold higher loss in WRMD2 on the first day of storage. These differences may be attributed to the different genetic backgrounds and ultimate pH of chicken meat. Le Bihan-Duval et al. (2008) highlighted a substantial negative correlation (r = 0.89)

**Table 1**Physicochemical characteristics of commercial broiler (CB) and Woorimatdag No. 2 (WRMD2) breast meat during aerobic cold storage.

Items	Treatment		Storage day						
		Day 1	Day 3	Day 5	Day 7				
pН	CB	$6.04 \pm$	6.10 $\pm$	6.12 $\pm$	$6.18 \pm$				
		0.05 <sup>Ab</sup>	0.05 <sup>Aab</sup>	0.06 <sup>Aab</sup>	$0.06^{Aa}$				
	WRMD2	5.67 $\pm$	5.71 $\pm$	5.73 $\pm$	5.78 $\pm$				
		$0.08^{Bb}$	$0.05^{\text{Bab}}$	$0.03^{\text{Bab}}$	$0.01^{Ba}$				
Drip loss	CB	0.17 $\pm$	0.98 $\pm$	1.11 $\pm$	1.81 $\pm$				
(%)		$0.03^{Bc}$	$0.12^{Bb}$	$0.22^{\mathrm{Bb}}$	$0.32^{Ba}$				
	WRMD2	$1.16~\pm$	$1.66~\pm$	2.36 $\pm$	2.85 $\pm$				
		0.13 <sup>Ad</sup>	0.14 <sup>Ac</sup>	$0.42^{Ab}$	$0.29^{Aa}$				
WHC (%)	CB	54.68 $\pm$	53.41 $\pm$	55.03 $\pm$	54.09 $\pm$				
		4.58	3.20	3.01	5.60				
	WRMD2	57.28 $\pm$	56.09 $\pm$	55.23 $\pm$	52.45 $\pm$				
		8.18	6.22	4.23	1.97				
CIE L*	CB	62.88 $\pm$	$61.36~\pm$	61.02 $\pm$	59.44 $\pm$				
		2.23	2.31	3.96	1.48				
	WRMD2	64.23 $\pm$	62.67 $\pm$	60.24 $\pm$	58.44 $\pm$				
		2.08 <sup>a</sup>	2.51 <sup>ab</sup>	4.07 <sup>ab</sup>	1.72 <sup>b</sup>				
CIE a*	CB	$1.88~\pm$	$1.20\ \pm$	1.02 $\pm$	0.96 $\pm$				
		$0.23^{a}$	$0.16^{Bb}$	$0.17^{Bb}$	$0.11^{Bb}$				
	WRMD2	1.74 $\pm$	$1.57~\pm$	$1.36~\pm$	$1.16~\pm$				
		0.21 <sup>a</sup>	0.14 <sup>Aab</sup>	$0.12^{Abc}$	0.18 <sup>Ac</sup>				
CIE b*	CB	4.28 $\pm$	$4.60 \pm$	4.91 $\pm$	5.76 $\pm$				
		0.84 <sup>b</sup>	0.61 <sup>Ab</sup>	0.53 <sup>ab</sup>	$0.59^{a}$				
	WRMD2	$3.70 \pm$	$3.83 \pm$	4.89 $\pm$	5.08 $\pm$				
		0.63 <sup>b</sup>	$0.26^{Bb}$	0.74 <sup>a</sup>	0.58 <sup>a</sup>				
WBSF (N)	CB	25.60 $\pm$	22.95 $\pm$	22.45 $\pm$	20.71 $\pm$				
		2.16 <sup>a</sup>	1.44 <sup>ab</sup>	$2.17^{b}$	1.58 <sup>b</sup>				
	WRMD2	26.14 $\pm$	$22.23 \pm$	20.81 $\pm$	19.51 $\pm$				
		1.49 <sup>a</sup>	1.55 <sup>b</sup>	1.46 <sup>b</sup>	$3.69^{b}$				

 $<sup>^{</sup>A,B}$  Different letters represent significant differences between broiler and Woorimatdag No. 2 within the same storage period (P < 0.05).

WHC, water-holding capacity; WBSF, Warner-Bratzler shear force.

between drip loss and ultimate pH of chicken breasts.

#### Water-holding capacity

Although the drip loss increased gradually during storage, the WHC for both CB (53.41 %–55.03 %) and WRMD2 (52.45 %–57.28 %) remained unchanged (Table 1). Moreover, no significant differences in the WHC were observed between CB and WRMD2 throughout the entire storage period. These results are consistent with those of Lee et al. (2022), who reported that the WHC of chicken thigh meat remained unaltered during aerobic storage. Similarly, Choe et al. (2010) found no significant difference in WHC between CB and KNC, supporting our findings.

#### Instrumental color

Generally, there was no significant difference in color between CB and WRMD2, except for the a\*- (days 3-7) and b\*-values (day 3) (Table 1). Choe et al. (2010) reported higher a\*- and b\*-values for CB than for KNC, which differed slightly from the results of the present study. Despite the reduction in the L\*-value during storage, no significant change was observed in the CB, which was also reported by Sujiwo et al. (2018). The L\*-value of WRMD2, however, decreased gradually on days 1–5 (P > 0.05), reaching its lowest level after day 7 (P < 0.05). A decrease in the L\*-value may be attributed to light scattering, which is mainly caused by protein oxidation and alterations in muscle structure (Lee et al., 2022). A negative correlation has been reported between pH and L\*-values (Le Bihan-Duval et al., 2008). Similarly, an increase in the pH of CB and WRMD2 was observed. The a\*-value was significantly reduced on days 1-3 in the CB and days 1-5 in the WRMD2. This supports earlier observations (Jaspal et al., 2021; Zheng et al., 2023). The reduction in the a\*-value may be due to the denaturation of meat globin during storage (Ab Aziz et al., 2020). Conversely, the b\*-values of CB

and WRMD2 gradually increased during storage. On day 7, CB showed a significant increase in the b\*-value compared to days 1–3, whereas days 5–7 for WRMD2 exhibited a significant increase compared to days 1–3. Numerous studies have reported an increase in the b\*-value of chicken meat (Jaspal et al., 2021; Kim et al., 2020; Sujiwo et al., 2018; Zheng et al., 2023). An increase in the b\*-value is associated with various factors, including lipid and protein oxidation, microbial spoilage, and metmyoglobin formation (Lee et al., 2022).

## Warner-Bratzler shear force

No significant difference in the WBSF was observed between the CB and WRMD2 groups (Table 1). CB and WRMD2 exhibited significant reductions in WBSF on days 1-5 and days 1-3, respectively. WBSF is an effective tool for assessing meat tenderness and is closely related to consumer satisfaction (Jung et al., 2023). The range of WBSF for CB and WRMD2 was 20.71-25.60 N and 19.51-26.14 N, respectively, which is in line with previous studies (Kim et al., 2020; Lee et al., 2022). The tenderization of meat during storage depends on the level of myofibrillar protein proteolysis by endogenous proteolytic enzyme systems (Sujiwo et al., 2018). In particular, u-calpains participated in costameres degradation and loss of cytoskeletal integrity, while m-calpains, along with increased ionic strengths and decreased pH, can considerably denature the myofibrillar proteins (Khan et al., 2016). The breast meat used in this study is generally described as tender because chicken breasts with a WBSF below 45 N are still considered tender by most consumers (Kim et al., 2020).

Storage stability and bioactive metabolites of WRMD2 and CB

#### Microorganisms

As shown in Table 2, APC for CB and WRMD2 increased gradually during storage (2.91–5.49 vs. 3.46–4.92 Log CFU/g), which is in line with previous observations (Kim et al., 2020; Lee et al., 2022; Sujiwo et al., 2018). WRMD2 showed significantly higher coliform levels than CB; however, no significant changes were observed during storage. Although the initial APC of CB was significantly lower than that of WRMD2, CB exhibited a significantly higher APC than WRMD2 from day

Table 2
Microorganisms, lipid oxidation, and volatile basic nitrogen content of commercial broiler (CB) and Woorimatdag No. 2 (WRMD2) breast meat during aerobic cold storage.

Items	Treatment	Storage day					
		Day 1	Day 3	Day 5	Day 7		
APC (Log CFU/g)	СВ	$\begin{array}{c} 2.91 \pm \\ 0.11^{\text{Bd}} \end{array}$	3.78 ± 0.35 <sup>c</sup>	$\begin{array}{l} 4.56 \pm \\ 0.19^{Ab} \end{array}$	5.49 ± 0.33 <sup>Aa</sup>		
-	WRMD2	$3.46 \pm 0.34^{Ac}$	$3.56 \pm 0.13^{c}$	$\begin{array}{l} 4.19 \pm \\ 0.27^{\text{Bb}} \end{array}$	$\begin{array}{l} \textbf{4.92} \pm \\ \textbf{0.15}^{\text{Ba}} \end{array}$		
Coliforms (Log CFU/g)*	СВ	$\begin{array}{l} 1.48 \pm \\ 0.22^{\text{B}} \end{array}$	$\begin{array}{c} 1.51 \; \pm \\ 0.18^{\mathrm{B}} \end{array}$	$1.31 \pm 0.43^{B}$	$\begin{array}{l} 1.23 \pm \\ 0.19^{\mathrm{B}} \end{array}$		
	WRMD2	$\begin{array}{c} \textbf{2.13} \pm \\ \textbf{0.24}^{\textbf{A}} \end{array}$	$\begin{array}{c} \textbf{2.56} \pm \\ \textbf{0.30}^{\textbf{A}} \end{array}$	$\begin{array}{l} 2.36 \pm \\ 0.39^{A} \end{array}$	$\begin{array}{l} \textbf{2.58} \pm \\ \textbf{0.14}^{\textbf{A}} \end{array}$		
TBARS (mg MDA/kg)	СВ	$\begin{array}{l} 0.11 \pm \\ 0.01^{\text{Bb}} \end{array}$	$\begin{array}{l} 0.12 \pm \\ 0.01^{\mathrm{Bb}} \end{array}$	$\begin{array}{l} 0.16 \pm \\ 0.01^{Ba} \end{array}$	$0.17 \pm 0.03^{\text{Ba}}$		
	WRMD2	$\begin{array}{l} 0.15 \pm \\ 0.02^{\text{Ac}} \end{array}$	$\begin{array}{l} 0.25 \pm \\ 0.01^{Ab} \end{array}$	$\begin{array}{l} 0.26 \pm \\ 0.01^{Ab} \end{array}$	$\begin{array}{l} 0.31 \; \pm \\ 0.01^{\text{Aa}} \end{array}$		
VBN (mg/100 g)	CB	$11.49 \pm \\ 0.99^{Ab}$	$11.21 \pm \\ 1.15^{\text{Bb}}$	$12.22 \pm \\ 1.62^{\rm b}$	$15.32 \pm 0.83^{a}$		
<u>.</u>	WRMD2	$\begin{array}{l} 9.37 \pm \\ 1.05^{\text{Bc}} \end{array}$	$\begin{array}{c} 12.59 \pm \\ 0.92^{Ab} \end{array}$	$12.30 \; \pm \\ 0.70^{b}$	$15.56 \pm 1.49^{a}$		

 $<sup>^{\</sup>rm A,B}$  Different letters represent significant differences between broiler and Woorimatdag No. 2 within the same storage period (P < 0.05).

 $<sup>^{\</sup>rm a-d}$  Different letters represent significant differences between storage days within the same chicken breed (P < 0.05).

 $<sup>^{\</sup>rm a\cdot d}$  Different letters represent significant differences between storage days within the same chicken breed (P < 0.05).

APC; aerobic plate counts; TBARS, 2-thiobarbituric acid reactive substances; MDA, malondialdehyde; VBN, volatile basic nitrogen.

 $<sup>^{\</sup>ast}$  E.coli was not detected in either WRMD2 or CB throughout the entire storage period.

5. It is possible that competitors of other aerobic microorganisms, including lactic acid bacteria, proliferate during storage, thereby altering the microbial status of chicken meat (Chmiel et al., 2020). Consumers typically prefer microbiologically safe fresh chicken meat because the growth of spoilage microorganisms, whether inside or outside the meat, can accelerate lipid rancidity, protein oxidation, and the formation of off-odor compounds, ultimately leading to undesirable organoleptic and nutritional properties (Abdel-Naeem et al., 2021). Nevertheless, the APC of CB and WRMD2 satisfied the levels recommended by the Ministry of Food and Drug Safety, Korea (<6.7 Log CFU/g; Kim and Jang, 2021) in addition to those of the International Commission on Microbiological Specifications for Foods (<7 Log CFU/g; Kim et al., 2020).

#### 2-Thiobarbituric acid reactive substances

Acceleration of lipid oxidation results in the deterioration of meat quality and the development of off-odor compounds. TBARS values exceeding 0.8 mg MDA/kg indicate perceptible rancidity (Jung et al., 2024). TBARS values for CB and WRMD2 increased gradually and were 0.11-0.17 and 0.15-0.31 mg MDA/kg, respectively (Table 2). The observed ranges are comparable to those reported in previous studies (Chmiel et al., 2019; Ham et al., 2020; Kim et al., 2020). The significantly higher TBARS values observed in WRMD2 than in CB could be attributed to the high proportion of polyunsaturated fatty acids (PUFA) (Table 4), which are more susceptible to oxidation than monounsaturated fatty acids (MUFA), as confirmed in Italian chickens (Dalle Zotte et al., 2020). Chaiwang et al. (2023) otherwise reported higher TBARS values in CB than in Thai chickens. It is possible, therefore, that genetic variations influence the lipid oxidation status of chicken meat, thereby potentially leading to differences in both the initial TBARS values and their subsequent increase. Multiple factors affecting lipid oxidation have been extensively documented, including the animal species and their endogenous antioxidant enzymes, free iron content (Utama et al., 2016), fat content and quality (Chmiel et al., 2019), unsaturated fatty acids, and pH status (Sujiwo et al., 2018).

#### Total volatile basic nitrogen

The decomposition of proteins and other nitrogen-containing components in meat via microbial activity and proteolytic enzymes results in the accumulation of organic amines, which are commonly collected as VBN value (Bekhit et al., 2021). A significant increase in the VBN value was observed on days 5–7 in the CB, and on days 1–3 and 5–7 in WRMD2 (Table 2). The VBN values were well below the recommended shelf-life limit of 20 mg/100 g (Sujiwo et al., 2018), thereby suggesting that all chicken meat used in this study may still be consumable. Herein, the alteration in VBN during storage exhibited a trend similar to that of APC and pH, which is in accordance with previous observations (Kim et al., 2020; Lee et al., 2022). This trend may be attributed to the fact that microbial activity is responsible for the production of VBN compounds. Sujiwo et al. (2018) reported high correlation coefficients between the total microorganisms and VBN (r = 0.92). Similarly, VBN has been demonstrated to exhibit a robust positive correlation with pH, as an increase in pH provides better growth conditions for microbes present in meat (Bekhit et al., 2021). Meanwhile, a significantly higher VBN value was observed in CB (day 1) and WRMD2 (day 3). However, no significant difference was found between the CB and WRMD2 groups from day

## Bioactive metabolites

As bioactive metabolites, creatine and creatinine play an important role in energy metabolism, muscle performance enhancement, and neuroprotection (Jung et al., 2013; Kim and Jang, 2021). CB significantly reduced creatine levels on days 1–7, whereas no significant changes were observed in WRMD2 (Table 3). Conversely, creatinine levels increased significantly in CB and WRMD2 on each storage day. These results are broadly consistent with those of Kim et al. (2020).

**Table 3**Bioactive metabolites of commercial broiler (CB) and Woorimatdag No. 2 (WRMD2) breast meat during aerobic cold storage.

Items	Treatment	Storage day					
		Day 1	Day 3	Day 5	Day 7		
Creatine	CB	446.42 $\pm$	435.28 $\pm$	429.07 $\pm$	424.00 $\pm$		
		5.41 <sup>Aa</sup>	17.21 <sup>Aab</sup>	9.43 <sup>Aab</sup>	$7.02^{Ab}$		
	WRMD2	385.26 $\pm$	381.78 $\pm$	$378.69 \pm$	$372.62 \pm$		
		8.58 <sup>B</sup>	27.92 <sup>B</sup>	10.64 <sup>B</sup>	12.48 <sup>B</sup>		
Creatinine	CB	$2.07~\pm$	3.32 $\pm$	4.44 $\pm$	5.28 $\pm$		
		$0.09^{Bd}$	0.16 <sup>Bc</sup>	0.13 <sup>b</sup>	$0.16^{Ba}$		
	WRMD2	$2.67~\pm$	$3.76 \pm$	4.33 $\pm$	4.84 $\pm$		
		0.09 <sup>Ad</sup>	$0.25^{Ac}$	$0.18^{b}$	$0.21^{Ba}$		
Carnosine	CB	175.14 $\pm$	173.22 $\pm$	173.55 $\pm$	$148.63 \pm$		
		15.49 <sup>Ba</sup>	16.45 <sup>Ba</sup>	15.15 <sup>a</sup>	12.31 <sup>b</sup>		
	WRMD2	248.05 $\pm$	217.57 $\pm$	178.28 $\pm$	$169.90 \pm$		
		23.45 <sup>Aa</sup>	35.99 <sup>Aab</sup>	26.05 <sup>bc</sup>	20.44 <sup>c</sup>		
Anserine	CB	$267.60~\pm$	$260.33~\pm$	240.67 $\pm$	232.58 $\pm$		
		20.44 <sup>Ba</sup>	23.08 <sup>Bab</sup>	17.01 <sup>Bab</sup>	$6.11^{Bb}$		
	WRMD2	515.85 $\pm$	519.95 $\pm$	479.69 $\pm$	477.29 $\pm$		
		26.11 <sup>Aab</sup>	13.23 <sup>Aa</sup>	38.89 <sup>Aab</sup>	18.60 <sup>Ab</sup>		
C:A ratio	CB	$0.66 \pm$	$0.67 \pm$	$0.73~\pm$	$0.64 \pm$		
		0.09 <sup>A</sup>	0.09 <sup>A</sup>	0.09 <sup>A</sup>	0.06 <sup>A</sup>		
	WRMD2	0.48 $\pm$	$0.42 \pm$	$0.38 \pm$	$0.36 \pm$		
		$0.05^{Ba}$	0.07 <sup>Bab</sup>	$0.08^{\mathrm{Bb}}$	0.04 <sup>Bb</sup>		

 $<sup>^{</sup>A,B}$  Different letters represent significant differences between broiler and Woorimatdag No. 2 within the same storage period (P < 0.05).

Increases in creatinine with decreases in creatine are primarily attributed to the non-enzymatic conversion of creatine to creatinine in muscle protein during cold storage (Kim and Jang, 2021). Carnosine and anserine provide valuable biological benefits, including physiological buffering of skeletal muscle, antioxidation, and neurotransmitter functions (Ali et al., 2021). A significant decrease in carnosine levels was observed in CB (days 5-7) and WRMD2 (days 1-5). Similarly, the anserine content decreased significantly in CB (days  $1\mbox{--}7$ ) and WRMD2 (days 3-7). Proteolysis can create extra space inside the myofibrils, which can subsequently elute the water-soluble dipeptides with water, or carnosinase present in muscle protein can break down carnosine (Kim and Jang, 2021). We found that creatine and anserine levels were genetically dependent in chickens. CB exhibited higher creatine levels than WRMD2 (424.00-446.42 vs. 372.62-385.26 mg/100 g), whereas WRMD2 showed approximately 2-folds significantly higher anserine levels than CB (477.29-519.95 vs. 232.58-267.60 mg/100 g). The carnosine-to-anserine ratio (C:A ratio) was significantly lower in WRMD2 (0.36-0.48) than CB (0.64-0.73). This finding aligns with previous observations, thereby indicating that various KNC strains exhibited a ratio of 0.21-0.31, whereas broilers demonstrated an approximate ratio of 0.60 (Ali et al., 2021; Jung et al., 2013; Kim et al., 2020). Native chickens have higher amounts of anserine than broilers, possibly because of differences in muscle fiber type; native and crossbred chickens typically contain higher levels of type IIB muscle than broilers (Charoensin et al., 2021).

## Flavor-related compounds of WRMD2 and CB

#### Fatty acid composition

Fatty acids play a crucial role in determining the flavor and nutritional profile of meat. C18:1n9 (31.13–39.49 %), C16:0 (22.67–23.77 %), and C18:2n6 (15.03–18.01 %) were the three predominant fatty acids in CB and WRMD2 (Table 4), as confirmed by previous observations (Chmiel et al., 2019; Kim et al., 2020; Lee et al., 2022). WRMD2 displayed significantly higher levels of C18:0 and total saturated fatty acids (SFA) than CB from day 3. Furthermore, WRMD2 contained significantly higher proportions of PUFA, including C22:4n6 and

 $<sup>^{\</sup>mathrm{a-d}}$  Different letters represent significant differences between storage days within the same chicken breed (P < 0.05).

C:A ratio, ratio of carnosine-to-anserine.

Table 4
Fatty acid composition of commercial broiler (CB) and Woorimatdag No. 2 (WRMD2) breast meat during aerobic cold storage.

Items	tems Treatment Storage day								
		Day 1	Day 3	Day 5	Day 7				
C14:0	СВ	0.89 ±	$0.88 \pm$	$0.82 \pm$	0.84 ±				
		0.06 <sup>a</sup>	0.01 <sup>a</sup>	0.02 <sup>b</sup>	0.01 <sup>ab</sup>				
	WRMD2	0.93 ±	0.91 ± 0.08	0.86 ±	0.89 ±				
C16:0	СВ	$0.09 \\ 23.59 \pm$	$0.08$ $23.26 \pm$	$0.07 \\ 23.68 \pm$	$0.16\ 22.67 \pm$				
G10.0	СБ	0.44 <sup>ab</sup>	0.31 <sup>ab</sup>	0.81 <sup>a</sup>	0.57 <sup>Bb</sup>				
	WRMD2	23.60 ±	23.59 ±	23.27 $\pm$	23.77 ±				
		0.91	1.24	0.80	0.68 <sup>A</sup>				
C16:1n7	CB	5.29 ±	$5.52 \pm$	5.44 ±	$5.11 \pm$				
		0.42 <sup>A</sup>	0.74 <sup>A</sup>	0.67 <sup>A</sup>	0.66 <sup>A</sup>				
	WRMD2	$2.35 \pm 0.60^{B}$	2.08 ± 0.51 <sup>B</sup>	$1.85 \pm 0.33^{B}$	$2.07 \pm 0.39^{B}$				
C18:0	СВ	8.24 ±	7.48 ±	$7.81~\pm$	7.84 ±				
010.0	02	$0.48^{B}$	0.58 <sup>B</sup>	0.56 <sup>B</sup>	$0.65^{B}$				
	WRMD2	$9.60 \pm$	9.85 $\pm$	10.29 $\pm$	9.33 $\pm$				
		1.27 <sup>A</sup>	0.91 <sup>A</sup>	0.87 <sup>A</sup>	0.58 <sup>A</sup>				
C18:1n9	CB	$39.01 \pm 1.22^{A}$	$39.49 \pm 1.16^{A}$	$38.19 \pm 1.57^{A}$	$38.01 \pm 1.23^{A}$				
	WRMD2	$33.13 \pm$	$31.96 \pm$	31.38 ±	$32.09 \pm$				
	WIGHDZ	1.84 <sup>B</sup>	1.81 <sup>B</sup>	1.96 <sup>B</sup>	1.35 <sup>B</sup>				
C18:1n7	CB	3.02 $\pm$	2.98 $\pm$	$3.12\ \pm$	2.95 $\pm$				
		0.21 <sup>A</sup>	0.11 <sup>A</sup>	0.21 <sup>A</sup>	0.08 <sup>A</sup>				
	WRMD2	$2.28 \pm 0.16^{Bb}$	2.25 ±	2.33 ±	2.69 ±				
C18:2n6	СВ	0.16 <sup>33</sup> 15.03 ±	0.06 <sup>Bb</sup> 15.50 ±	$0.10^{ m Bb} \ 15.37 \pm$	$0.26^{\mathrm{Ba}} \ 16.79 \pm$				
C10.2110	СБ	0.81 <sup>Bb</sup>	0.94 <sup>Bab</sup>	1.19 <sup>Bab</sup>	$0.99^{a}$				
	WRMD2	18.01 $\pm$	17.76 $\pm$	17.51 $\pm$	17.62 $\pm$				
		1.81 <sup>A</sup>	1.04 <sup>A</sup>	1.38 <sup>A</sup>	0.48				
C18:3n6	CB	0.13 ±	0.17 ±	0.15 ±	0.18 ±				
	WRMD2	$0.01^{ m Ab} \ 0.08 \pm$	0.01 <sup>Aa</sup> 0.06 ±	$\begin{array}{c} 0.01^{\text{Ab}} \\ 0.05 \ \pm \end{array}$	$0.01^{ m Aa} \ 0.14 \pm$				
	WIGNIDZ	0.03 ± 0.02 <sup>Bb</sup>	0.00 ± 0.01 <sup>Bb</sup>	0.03 ± 0.01 <sup>Bb</sup>	$0.14 \pm 0.03^{\text{Ba}}$				
C18:3n3	CB	$0.77 \pm$	$0.83 \pm$	$0.77 \pm$	$0.85 \pm$				
		$0.06^{Ab}$	$0.03^{Aab}$	0.05 <sup>Aab</sup>	$0.03^{Aa}$				
	WRMD2	0.56 ±	0.53 ±	0.48 ±	0.48 ±				
C20:1n9	СВ	0.09 <sup>B</sup> 0.64 ±	$0.06^{\mathrm{B}} \\ 0.63 \pm$	$0.08^{\mathrm{B}} \ 0.63 \pm$	$0.02^{ m B} \ 0.58 \pm$				
620.1117	CD	0.04 <sup>A</sup>	0.04 <sup>A</sup>	0.07 <sup>A</sup>	0.02 <sup>A</sup>				
	WRMD2	0.34 $\pm$	$0.34 \pm$	$0.38 \pm$	0.38 $\pm$				
		$0.06^{B}$	$0.06^{B}$	$0.08^{B}$	$0.06^{B}$				
C20:4n6	CB	2.37 ±	2.30 ±	2.74 ±	2.95 ±				
	WRMD2	$0.26^{B}$ $7.03 \pm$	0.40 <sup>B</sup> 8.36 ±	$0.39^{B}$ $9.08 \pm$	$0.57^{ m B} \ 8.13 \pm$				
	WIGNIDZ	1.84 <sup>A</sup>	2.55 <sup>A</sup>	1.88 <sup>A</sup>	1.37 <sup>A</sup>				
C20:5n3	CB	0.13 $\pm$	0.10 $\pm$	0.12 $\pm$	0.11 $\pm$				
		$0.02^{A}$	0.02 <sup>A</sup>	0.03 <sup>A</sup>	0.01				
	WRMD2	$0.04 \pm 0.01^{\text{Bb}}$	$0.06 \pm 0.02^{ m Bb}$	$0.03 \pm 0.00^{ m Bb}$	0.10 ±				
C22:4n6	СВ	0.01 <sup></sup> 0.57 ±	$0.02^{-2}$ $0.61 \pm$	$0.00^{-2}$ $0.81 \pm$	$0.02^{a} \ 0.78 \pm$				
622.4110	CD	0.09 <sup>B</sup>	0.01 ± 0.11 <sup>B</sup>	0.20 <sup>B</sup>	0.70 ± 0.22 <sup>B</sup>				
	WRMD2	1.04 $\pm$	$1.03\ \pm$	$1.33~\pm$	$1.15~\pm$				
		0.35 <sup>A</sup>	0.16 <sup>A</sup>	0.36 <sup>A</sup>	0.17 <sup>A</sup>				
C22:6n3	CB	$0.24 \pm 0.04^{Bab}$	0.19 ±	0.29 ±	$0.27 \pm 0.05^{Bab}$				
	WRMD2	0.04 0.95 ±	$0.03^{ m Bb} \ 1.14 \pm$	$0.06^{\mathrm{Ba}} \ 1.08 \pm$	$1.08 \pm$				
	WIGNIDZ	0.22 <sup>A</sup>	0.40 <sup>A</sup>	0.16 <sup>A</sup>	0.14 <sup>A</sup>				
SFA	CB	32.73 $\pm$	$31.63 \pm$	32.32 $\pm$	$31.37~\pm$				
		0.88	0.41 <sup>B</sup>	$1.18^{B}$	$0.82^{B}$				
	WRMD2	34.14 ±	34.37 ±	34.43 ±	34.00 ±				
UFA	СВ	$1.30 \\ 67.26 \pm$	$1.63^{A}$ $68.36 \pm$	$1.50^{ m A}\ 67.67~\pm$	$0.78^{A}$ $68.62 \pm$				
UFA	CD	0.88	0.41 <sup>A</sup>	1.18 <sup>A</sup>	0.82 <sup>A</sup>				
	WRMD2	65.85 ±	65.62 ±	65.56 ±	65.99 ±				
		1.30	1.63 <sup>B</sup>	1.50 <sup>B</sup>	$0.78^{B}$				
MUFA	CB	47.98 ±	48.64 ±	47.39 ±	46.67 ±				
	MIDMIDO	1.26 <sup>A</sup>	1.81 <sup>A</sup> 36.63 ±	$1.72^{A}$ 35.96 $\pm$	$1.94^{A}$ 37.25 $\pm$				
	WRMD2	$38.11 \pm 2.46^{B}$	$36.63 \pm 2.22^{B}$	35.96 ± 2.34 <sup>B</sup>	$37.25 \pm 1.85^{B}$				
PUFA	CB	19.27 $\pm$	$19.72 \pm$	20.28 $\pm$	$21.95 \pm$				
		$0.87^{Bb}$	1.47 <sup>Bab</sup>	1.43 <sup>Bab</sup>	$1.78^{Ba}$				
	WRMD2	27.74 ±	28.98 ±	29.60 ±	28.74 ±				
		2.73 <sup>A</sup>	3.18 <sup>A</sup>	1.07 <sup>A</sup>	1.73 <sup>A</sup>				

Table 4 (continued)

Items	Treatment	Storage day				
		Day 1	Day 3	Day 5	Day 7	
MUFA/ SFA	СВ	$\begin{array}{c} \textbf{1.46} \pm \\ \textbf{0.07}^{\textbf{A}} \end{array}$	$\begin{array}{c} 1.53 \pm \\ 0.07^{\text{A}} \end{array}$	$1.46 \pm 0.09^{A}$	$1.48 \pm 0.08^{A}$	
	WRMD2	$\begin{array}{c} 1.11 \; \pm \\ 0.08^{B} \end{array}$	$1.06 \pm 0.06^{B}$	$\begin{array}{c} \textbf{1.04} \pm \\ \textbf{0.11}^{\textbf{B}} \end{array}$	$1.09 \pm 0.06^{B}$	
PUFA/ SFA	СВ	$0.58 \pm 0.03^{\mathrm{Bb}}$	$\begin{array}{c} 0.62 \pm \\ 0.04^{Bab} \end{array}$	$\begin{array}{l} \textbf{0.62} \pm \\ \textbf{0.05}^{\text{Bab}} \end{array}$	$\begin{array}{l} 0.70 \; \pm \\ 0.06^{\text{Ba}} \end{array}$	
	WRMD2	$0.81 \pm 0.09^{A}$	$\begin{array}{l} \textbf{0.84} \pm \\ \textbf{0.12}^{\textbf{A}} \end{array}$	$\begin{array}{c} \textbf{0.86} \pm \\ \textbf{0.02}^{\textbf{A}} \end{array}$	$0.84 \pm 0.05^{A}$	
n6/n3	СВ	$15.75 \pm 0.67$	$16.48 \pm \\ 0.61$	$16.00 \pm 0.78^{\text{B}}$	$16.71 \pm \\ 0.60$	
	WRMD2	$16.98 \pm \\1.89$	$15.95 \pm 2.39$	$17.41 \pm 0.95^{\text{A}}$	$16.16\ \pm$ $1.77$	

 $<sup>^{</sup>A,B}$  Different letters represent significant differences between broiler and Woorimatdag No. 2 within the same storage period (P < 0.05).

SFA, sum of saturated fatty acid; UFA, sum of unsaturated fatty acid; MUFA, sum of monounsaturated fatty acid; PUFA, sum of polyunsaturated fatty acid.

C22:6n3, than CB. Conversely, CB dominated six unsaturated fatty acids, including C16:1n7, C18:1n9, C18:1n7, and total MUFA, compared with WRMD2. These findings are consistent with those of a previous study on KNC thigh meat (Barido et al., 2022). A MUFA/SFA (1.5: 1 recommended), PUFA/SFA (>0.4 recommended), and n6/n3 ratio (5-10 recommended) are the nutritional parameters of meat fat (Jung et al., 2023; Martínez-Álvaro et al., 2018). CB generally met the recommended MUFA/SFA ratio (1.46-1.53), and the PUFA/SFA ratio was well within the suggested ranges in both CB (0.58-0.70) and WRMD2 (0.81-0.86). The MUFA/SFA and PUFA/SFA ratios were significantly higher in CB and WRMD2, respectively, whereas the n6/n3 ratio was unfavorable. A significant increase in C18:1n7, C18:3n6, and C20:5n3 was observed on days 5-7 in WRMD2, whereas C18:2n6, C18:3n3, and PUFA increased significantly on days 1-7 in CB. Many factors, including breeding, feeding system, post-mortem changes, and weight, affect meat fatty acid status (Xiao et al., 2021). Previous studies demonstrated the relationship between specific fatty acids and organoleptic properties (Barido et al., 2022; Jung et al., 2023). C18:1n9 and C20:4n6 were significantly higher in CB and WRMD2, respectively. These are positively responsible for the savory flavor of meat or are further oxidized to produce numerous volatile substances such as 1-hexanol and saturated aldehydes (Jung et al., 2024). C18:2n6 and C22:6n3, which are characterized by umami/sweet and sour tastes, respectively (Barido et al., 2022), were significantly higher in WRMD2 than in CB, except for C18:2n6 on day 7. Our study suggests that the fatty acid composition of chicken breast is more commonly influenced by genetic background than by storage.

#### Flavor nucleotides

Nucleotides are critical to the perception of meat because they provide a characteristic taste and are susceptible to changes during storage (Hou et al., 2018). Both IMP and GMP are flavor-potentiating compounds that enhance meat flavor intensity on their own or synergistically with monosodium glutamate, imparting umami and meaty notes (Dashdorj et al., 2015). IMP decreased gradually on days 1–3 and 5–7 in CB, whereas in WRMD2, IMP remained at similar levels on days 1-5 and decreased significantly on day 7 (Table 5). Similarly, CB showed a significant decrease in GMP on days 1-3 and 3-5, whereas WRMD2 showed a significant reduction in GMP on days 1-7, thereby suggesting that WRMD2 is better than CB at preserving umami-nucleotides. According to Hou et al. (2018), the simultaneous reduction in IMP in chicken breast tissue may be attributed to the faster degradation of IMP than its synthesis. Moreover, IMP can be further broken down into inosine and converted to hypoxanthine by IMP phosphatase and nucleosidase during storage. Inosine and hypoxanthine are of importance, as they can indicate a bitter taste in meat (Dashdorj et al., 2015). A significant increase

 $<sup>^{\</sup>rm a,b}$  Different letters represent significant differences between storage days within the same chicken breed (P < 0.05).

**Table 5**Nucleotide-related compounds of commercial broiler (CB) and Woorimatdag No. 2 (WRMD2) breast meat during aerobic cold storage.

Items	Treatment	Storage day					
		Day 1	Day 3	Day 5	Day 7		
IMP	СВ	249.79 $\pm$	199.79 $\pm$	188.22 $\pm$	140.31 ±		
		37.66 <sup>Aa</sup>	27.78 <sup>b</sup>	$23.22^{b}$	24.16 <sup>c</sup>		
	WRMD2	190.52 $\pm$	176.17 $\pm$	184.33 $\pm$	137.09 $\pm$		
		30.71 <sup>Ba</sup>	21.02 <sup>a</sup>	21.89 <sup>a</sup>	$21.05^{b}$		
GMP	CB	5.27 $\pm$	4.17 $\pm$	2.98 $\pm$	$2.58~\pm$		
		$0.30^{Aa}$	0.43 <sup>Ab</sup>	0.47 <sup>c</sup>	0.18 <sup>c</sup>		
	WRMD2	$3.83~\pm$	3.30 $\pm$	3.37 $\pm$	$2.76~\pm$		
		$0.57^{Ba}$	$0.39^{\text{Bab}}$	$0.32^{ab}$	$0.14^{b}$		
Inosine	CB	75.15 $\pm$	87.81 $\pm$	94.02 $\pm$	$111.89\ \pm$		
		15.04 <sup>c</sup>	9.05 <sup>bc</sup>	6.48 <sup>b</sup>	11.56 <sup>Aa</sup>		
	WRMD2	73.46 $\pm$	79.98 $\pm$	85.33 $\pm$	88.10 $\pm$		
		13.50	10.32	11.52	9.79 <sup>B</sup>		
Hypoxanthine	CB	$6.30 \pm$	11.95 $\pm$	16.12 $\pm$	19.30 $\pm$		
		$0.92^{Bd}$	2.12 <sup>c</sup>	$1.15^{Ab}$	1.36 <sup>Aa</sup>		
	WRMD2	8.45 $\pm$	$11.90~\pm$	12.18 $\pm$	15.79 $\pm$		
		1.43 <sup>Ac</sup>	$0.98^{b}$	$1.32^{\mathrm{Bb}}$	$0.56^{Ba}$		
ATP	CB	8.26 $\pm$	8.26 $\pm$	6.40 $\pm$	7.27 $\pm$		
		$0.27^{Ba}$	0.40 <sup>Ba</sup>	0.94 <sup>Bb</sup>	$0.42^{Bb}$		
	WRMD2	9.04 $\pm$	9.38 $\pm$	$10.09~\pm$	9.18 $\pm$		
		0.55 <sup>Ab</sup>	0.42 <sup>Aab</sup>	1.04 <sup>Aa</sup>	$0.25^{Aab}$		
ADP	CB	6.10 $\pm$	5.86 $\pm$	5.60 $\pm$	5.45 $\pm$		
		0.24 <sup>Aa</sup>	0.41 <sup>ab</sup>	0.31 <sup>Bab</sup>	$0.22^{b}$		
	WRMD2	5.65 $\pm$	5.70 $\pm$	6.35 $\pm$	$5.39~\pm$		
		0.43 <sup>Bb</sup>	$0.21^{b}$	0.45 <sup>Aa</sup>	$0.12^{b}$		
AMP	CB	10.41 $\pm$	10.41 $\pm$	9.95 $\pm$	10.41 $\pm$		
		0.82 <sup>A</sup>	0.40 <sup>A</sup>	0.57	0.87 <sup>A</sup>		
	WRMD2	8.40 $\pm$	8.96 $\pm$	10.25 $\pm$	9.36 $\pm$		
		$0.65^{Bc}$	$0.39^{\text{Bbc}}$	0.76 <sup>a</sup>	$0.39^{\text{Bab}}$		

 $<sup>^{\</sup>mathrm{A,B}}$  Different letters represent significant differences between broiler and Woorimatdag No. 2 within the same storage period (P < 0.05).

IMP, inosine monophosphate; GMP, guanosine monophosphate; AMP, adenosine monophosphate; ADP, adenosine diphosphate; ATP, adenosine triphosphate.

in inosine was observed in the CB on days 1-5 and 5-7, whereas no alterations were identified in the WRMD2. Similarly, CB significantly increased the hypoxanthine levels during storage. WRMD2 also demonstrated a significant increase in hypoxanthine levels on days 1–3 and 5-7. An increase in inosine and hypoxanthine with prolonged storage may be due to the enzymatic degradation of IMP, as discussed above. AMP by itself can provide the sweet perception of meat at low concentrations (50-100 mg/100 g) or works synergistically with IMP to induce an umami taste (Jung et al., 2023). The only significant increase in AMP was observed in WRMD2 on days 1-5. However, minor nucleotide variations were observed between the chicken groups. CB showed a significantly higher nucleotide content than WRMD2, such as IMP (day 1) and GMP (days 1-3). WRMD2, however, exhibited higher levels of some nucleotides than CB, such as hypoxanthine (day 1), adenosine diphosphate (day 5), and adenosine triphosphate (day 1–7). This finding was confirmed by Barido et al. (2022), who reported the effects of genetic variations on the nucleotide content of chicken meat. In the present study, nucleotides were influenced more by storage time than by genetic differences, in contrast to the findings observed for fatty acids.

## Volatile organic compounds

A total of 64 individual VOCs were identified including alcohols (12), aldehydes (14), esters (10), hydrocarbons (23), and ketones (5) (Table S1). Total VOCs exhibited a significant decrease in the CB on days 1–7, whereas no significant alterations were observed in WRMD2 (Table 6). Esters and hydrocarbons are the most abundant VOCs. WRMD2 exhibited significantly higher total VOCs than CB from day 3. CB exhibited a significant increase in (S)-3-methyl-1-pentanol, 1-octen-3-ol, and total alcohols on days 1–7, whereas no significant changes in

alcohols were observed in WRMD2. Multiple factors, including microbial spoilage, proteolytic activity, lipid oxidation, and reduction of aldehydes and 2-ketones, have been reported to affect alcohol status in meat (Casaburi et al., 2015). WRMD2 showed significantly higher levels of total alcohols, including 1-heptanol, 1-octanol, and 1-octen-3-ol, than CB. Interestingly, 1-octen-3-ol was the predominant alcohol in WRMD2, accounting for approximately 82 % of total alcohol abundance. Previous studies have demonstrated that 1-octen-3-ol is a characteristic alcohol found in Korean and Chinese chicken meat (Barido et al., 2022; Wang et al., 2024). Triacylglycerol hydrolysis and fatty acid metabolism are the primary pathways responsible for aldehyde formation in meat (Pereira et al., 2021). Overall, no clear alterations in aldehyde levels were observed during storage, except for benzeneacetaldehyde, dodecanal, and hexanal. Further, 2-methylpropanal, 5-methylhexanal, hexanal, nonanal, and octanal dominated the aldehyde proportions. CB and WRMD2 contained 2-methylpropanal at approximately 82 % and 40 % of the total aldehyde abundance, respectively. WRMD2 showed significantly higher levels of eight aldehydes than CB, including 2,4-decadienal, hexanal, nonanal, and octanal, which is in accordance with the findings of Wang et al. (2024). Moreover, aldehydes may have a significant impact on meat flavor owing to their low olfactory thresholds. Several aldehydes, including benzeneacetaldehyde, 2,4-decadienal, octanal, and nonanal, have been reported as good contributors to meat flavor (Barido et al., 2022; Jung et al., 2023).

Esters are derived from the esterification of alcohols and carboxylic acids as a result of microbial esterase activity (Casaburi et al., 2015). Arsenous acid, tris(trimethylsilyl) ester represents a large group of esters in chicken meat (Barido et al., 2022), and their levels decreased significantly in the CB on days 1–3. Significantly higher levels of benzoic acid, 2-hydroxy-, ethyl ester, methyl salicylate, and n-caproic acid vinyl ester were observed in WRMD2 than in CB. Ethyl esters, with their sweet/fruity descriptors and low odor threshold, significantly contribute to meat flavor profiles (Casaburi et al., 2015). Benzoic acid, 2-hydroxy-, ethyl ester and dodecanoic acid, ethyl ester were exclusively detected in WRMD2; however, benzoic acid, 2-hydroxy-, ethyl ester decreased significantly on days 1–5. Slight changes in hydrocarbons were observed in CB (including 3,3-dimethyl-1,2-epoxybutane and sec-butylamine) and WRMD2 (including decane and dodecane) during storage. Compared with CB, WRMD2 exhibited significantly higher levels of total hydrocarbons with five individual substances, except for naphthalene. Notably, 2-pentylfuran and trimethylene oxide were exclusively present in WRMD2, whereas pentane, 2,3-dimethyl- was observed at approximately 22-fold higher levels in WRMD2 than in CB. Hydrocarbons are commonly produced by the oxidative decomposition of fatty acids and are often considered insignificant contributors to meat flavor owing to their high odor threshold (Jung et al., 2023). Numerous studies have reported that 2-ketones (or methyl ketone, the product of  $\beta$ -keto acids) are considered to contribute much to meat aroma (Bassam et al., 2022; Jung et al., 2024). Significantly higher levels of 2,3-butanedione were observed in WRMD2 on day 5 than on days 3 and 7, whereas no significant changes were observed in the CB. 2-Octanone was newly generated in WRMD2 from day 5 (P < 0.05). WRMD2 exhibited significantly higher amounts of (+)-2-bornanone and acetophenone than CB. Consequently, variations in VOCs could be affected by either storage period or breed difference, as demonstrated in this study. Barido et al. (2022) reported numerous factors, including age, breed, and storage conditions that are strongly correlated with the development of chicken meat aroma.

PLS-DA is a robust analytical tool that enables the visualization of treatment differences with multiple and complex variables, such as volatiles, in a single straightforward image, thereby enhancing data interpretation. In the CB, there was a similar VOC cluster between days 1 and 3, and between days 5 and 7 (Fig. 1). The VOC clusters on days 5 and 7 could be clearly distinguished from those on days 1 and 3, and even between each other. In WRMD2, however, the VOC cluster on day 1 was clearly differentiated from that on days 3, 5, and 7, whereas no clear

 $<sup>^{\</sup>rm a-d}$  Different letters represent significant differences between storage days within the same chicken breed (P < 0.05).

Table 6
Major volatile organic compounds of commercial broiler (CB) and Woorimatdag No. 2 (WRMD2) breast meat during aerobic cold storage.

Items (AU $\times$ 10 <sup>5</sup> )			CB			WRI	MD2		
	Day 1	Day 3	Day 5	Day 7	Day 1	Day 3	Day 5	Day 7	
Alcohols									
(S)-3-Methyl-1-pentanol	$\begin{array}{l} 0.37~\pm\\ 0.22^{Bb} \end{array}$	$\begin{array}{l} 0.72 \pm \\ 0.14^{Bab} \end{array}$	$\begin{array}{l} 0.83 \pm \\ 0.32^{Bab} \end{array}$	$1.06 \pm 0.47^{Ba}$	$3.03\pm1.23^{\text{A}}$	$\textbf{4.42} \pm \textbf{2.07}^{\textbf{A}}$	$3.57\pm1.93^{\text{A}}$	$2.93 \pm 0.70$	
1-Heptanol	$0.23\pm0.09^{\text{B}}$	$0.23\pm0.04^{\text{B}}$	$0.27\pm0.16^{\mathrm{B}}$	$0.24 \pm 0.06^{B}$	$1.64\pm0.93^{\text{A}}$	$1.99\pm1.08^{\text{A}}$	$1.73\pm1.20^{\text{A}}$	$1.22 \pm 0.54$	
1-Hexanol, 2-ethyl-	$1.21\pm0.28^{b}$	$2.23\pm0.57^{\text{Aa}}$	$2.26\pm0.75^{Aa}$	$\begin{array}{c} 2.38 \pm \\ 0.69^{\text{Aa}} \end{array}$	$\textbf{0.74} \pm \textbf{0.46}$	$1.10\pm0.67^{\text{B}}$	$1.08\pm0.65^{\text{B}}$	$1.53\pm0.62$	
l-Nonen-4-ol	$ND^{B}$	$ND^{B}$	$ND^{B}$	ND	$0.32\pm0.20^{\text{A}}$	$0.32 \pm 0.25^{\text{A}}$	$0.40\pm0.37^{\text{A}}$	$0.25 \pm 0.27$	
1-Octanol	$0.48\pm0.18^{\text{B}}$	$0.39 \pm 0.07^{\text{B}}$	$0.40\pm0.14^{\text{B}}$	$0.33\pm0.13^{\text{B}}$	$1.28\pm0.66^{\text{A}}$	$1.81\pm0.89^{\text{A}}$	$1.55\pm0.98^{\text{A}}$	$1.01 \pm 0.42$	
1-Octen-3-ol	$\begin{array}{c} 1.33 \pm \\ 0.11^{\text{Bb}} \end{array}$	$\begin{array}{l} 1.72 \pm \\ 0.46^{Bab} \end{array}$	$\begin{array}{l} 1.92 \pm \\ 0.30^{Bab} \end{array}$	$\begin{array}{l} \textbf{2.97} \pm \\ \textbf{1.58}^{\text{Ba}} \end{array}$	$44.39 \pm 28.53^{A}$	$68.26 \pm \\ 42.20^{A}$	$65.21 \pm 40.54^{A}$	$42.05 \pm 16.2^{A}$	
2-Octen-1-ol, (E)-	$ND^{B}$	$ND^{B}$	$ND^{B}$	$ND^{B}$	$0.72\pm0.45^{\text{A}}$	$2.04\pm1.60^{\text{A}}$	$1.74\pm1.55^{\text{A}}$	$0.80 \pm 0.69$	
2-Octen-1-ol, (Z)-	$ND^{B}$	$ND^{B}$	ND	$ND^{B}$	$0.75\pm1.27^{\text{A}}$	$1.34\pm0.84^{\text{A}}$	ND	$0.98 \pm 0.79$	
2,4-Dimethylcyclohexanol	$ND^{B}$	$ND^{B}$	$ND^{B}$	$ND^{B}$	$0.20 \pm 0.15^{A}$	$0.30\pm0.31^{\text{A}}$	$0.30 \pm 0.25^{A}$	$0.18\pm0.15$	
Subtotal*	$3.65 \pm 0.52^{\text{Bb}}$	$5.45 \pm 0.81^{Bab}$	$5.69 \pm 1.39^{Bab}$	$7.00 \pm 2.65^{Ba}$	$53.34 \pm 32.11^{A}$	$83.54 \pm 48.06^{A}$	77.81 $\pm$ 47.28 <sup>A</sup>	$51.79 \pm 19.12^{A}$	
Aldehyde									
2,4-Decadienal	$ND^{B}$	$ND^{B}$	ND <sup>B</sup>	ND <sup>B</sup>	$0.11\pm0.02^{\rm A}$	$0.12 \pm 0.03^{A}$	$0.14 \pm 0.08^{A}$	$0.07 \pm 0.00$	
2-Octenal, (E)-	ND	$ND^{B}$	ND <sup>B</sup>	$ND^{B}$	$0.19 \pm 0.25$	$0.41 \pm 0.16^{A}$	$0.31 \pm 0.29^{A}$	$0.25 \pm 0.13$	
5-methylhexanal	$0.37 \pm 0.15^{B}$	$0.39 \pm 0.14^{B}$	$0.41 \pm 0.17^{B}$	$0.27 \pm 0.07^{B}$	$4.16 \pm 2.68^{A}$	$4.45 \pm 2.06^{A}$	$4.21 \pm 2.68^{A}$	$2.56 \pm 1.47$	
Benzeneacetaldehyde	0.26 ± 0.14	$0.31 \pm 0.04$	$0.37 \pm 0.11$	$0.43\pm0.10$	$0.22 \pm 0.15^{D}$	$0.45 \pm 0.15^{a}$	$0.40 \pm 0.09^{ab}$	$0.41 \pm 0.07^{ab}$	
Dodecanal	$ND^{Bb}$	$\mathrm{ND}^{\mathrm{Bb}}$	$ND^{Bb}$	$\begin{array}{l} 0.02 \pm \\ 0.01^{\text{Ba}} \end{array}$	$0.09\pm0.04^{\mathrm{A}}$	$0.11\pm0.02^{\mathrm{A}}$	$0.12\pm0.03^{\mathrm{A}}$	$0.07 \pm 0.01$	
Hexanal	$0.34 \pm 0.09^{Bb}$	$\begin{array}{l} \textbf{0.71} \pm \\ \textbf{0.12}^{\text{Bab}} \end{array}$	$0.89\pm0.24^{\text{Ba}}$	$\begin{array}{l} \textbf{1.08} \pm \\ \textbf{0.44}^{\text{Ba}} \end{array}$	$2.02\pm0.68^{\text{Ab}}$	$\begin{array}{l} 3.49 \pm \\ 0.86^{Aab} \end{array}$	$\textbf{4.34} \pm \textbf{1.22}^{\text{Aa}}$	$\begin{array}{l} 3.62 \pm \\ 0.79^{\text{Aa}} \end{array}$	
Hexadecanal	ND <sup>B</sup>	ND <sup>B</sup>	$0.02\pm0.03^{\text{B}}$	$0.02 \pm 0.03^{B}$	$0.07\pm0.02^{\text{A}}$	$0.15 \pm 0.08^{A}$	$0.11\pm0.03^{\text{A}}$	$0.08 \pm 0.03$	
Nonanal	$0.86\pm0.94^{\text{B}}$	$1.30\pm1.02^{\text{B}}$	$2.03\pm1.09^{\mathrm{B}}$	$0.79\pm0.86^{\text{B}}$	$5.38\pm2.86^{\text{A}}$	$6.39 \pm 2.81^{\text{A}}$	$5.97 \pm 3.18^{\text{A}}$	$3.52\pm1.91$	
Octanal	$0.31\pm0.10^{\text{B}}$	$0.33\pm0.13^{\text{B}}$	$0.36\pm0.13^{\text{B}}$	$0.28\pm0.09^{\text{B}}$	$2.97\pm2.04^{\hbox{\scriptsize A}}$	$3.08\pm1.02^{\text{A}}$	$2.64\pm1.64^{\text{A}}$	$1.70 \pm 0.93$	
Pyran aldehyde	$ND^{B}$	$ND^{B}$	$ND^{B}$	$ND^{B}$	$0.12\pm0.05^{\text{A}}$	$0.17\pm0.16^{\text{A}}$	$0.18\pm0.12^{\text{A}}$	$0.15\pm0.06$	
2-Methylpropanal	$8.25 \pm 9.72$	$12.37\pm1.68^{\text{A}}$	$9.29 \pm 5.19$	$10.95\pm6.84$	$8.06 \pm 4.86$	$6.12\pm5.09^{\text{B}}$	$\textbf{7.08} \pm \textbf{3.83}$	$5.77\pm1.27$	
Subtotal*	$9.80\pm9.72^{\text{B}}$	$14.80\pm2.00^{B}$	$12.53\pm5.27^{\mathrm{B}}$	$12.90\pm7.68$	$18.18 \pm \\8.55^{A}$	$18.23 \pm \\ 6.16^{\text{A}}$	$18.05 \pm 5.95^{A}$	$12.93 \pm 2.80$	
Ester									
2-Butenoic acid, 2-methyl-, 2- methylpropyl ester	ND <sup>B</sup>	$ND^{B}$	$0.01\pm0.01$	$0.02\pm0.03^{\mathrm{B}}$	$0.26\pm0.17^{\mathrm{A}}$	$0.53\pm0.52^{\mathrm{A}}$	$0.35 \pm 0.39$	$0.51\pm0.06$	
Arsenous acid, tris(trimethylsilyl) ester	$\begin{array}{l} 213.65 \pm \\ 52.48^{\text{Aa}} \end{array}$	$\begin{array}{c} 102.38 \pm \\ 7.88^{\mathrm{b}} \end{array}$	$103.82 \pm 22.85^{\rm b}$	$110.66 \pm \\ 44.11^{b}$	$104.16 \pm \\ 46.08^{B}$	$99.33 \pm 3.58$	$115.06 \pm 34.15$	$131.30 \pm 4.91$	
Benzoic acid, 2-hydroxy-, ethyl ester	$ND^B$	$ND^{B}$	$ND^{B}$	$ND^{B}$	$0.27\pm0.08^{\text{Aa}}$	$\begin{array}{l} 0.22 \pm \\ 0.12^{Aab} \end{array}$	$0.12\pm0.05^{\text{Ab}}$	$\begin{array}{l} 0.10 \pm \\ 0.02^{Ab} \end{array}$	
Dodecanoic acid, ethyl ester	$ND^B$	$ND^{B}$	$ND^{B}$	$ND^B$	$0.13\pm0.04^{\text{A}}$	$0.12 \pm 0.14^{A}$	$0.20\pm0.09^{\text{A}}$	$0.02$ $0.28 \pm 0.14$	
Methyl salicylate	$0.28 \pm 0.29^{B}$	$0.30 \pm 0.05^{B}$	$0.31 \pm 0.10^{B}$	$0.25\pm0.08^{\mathrm{B}}$	$3.05\pm0.52^{\text{Aa}}$	$2.35 \pm 1.16^{Aa}$	$1.18\pm0.36^{\text{Ab}}$	1.04 ± 0.40 <sup>Ab</sup>	
n-Caproic acid vinyl ester	$0.32\pm0.33^{\text{B}}$	$0.48 \pm 0.35^{\text{B}}$	$0.58\pm0.36^{\text{B}}$	$0.71\pm0.31^{\text{B}}$	$13.41 \pm \\ 10.02^{\text{A}}$	$18.47 \pm \\7.61^{\text{A}}$	$\begin{array}{c} 20.21 \; \pm \\ 11.10^A \end{array}$	13.06 ± 4.07 <sup>A</sup>	
Subtotal*	214.40 $\pm$	103.31 $\pm$	104.90 $\pm$	112.05 $\pm$	$10.02$ $121.83 \pm$	$122.02 \pm$	$138.63 \pm$	4.07 148.11 ±	
	52.57 <sup>Aa</sup>	7.77 <sup>Bb</sup>	22.61 <sup>b</sup>	44.08 <sup>b</sup>	49.67 <sup>B</sup>	9.09 <sup>A</sup>	37.98	5.97	
Hydrocarbon 1,3-Dioxolane, 2-(1-ethylpentyl)-	162.97 $\pm$	157.94 $\pm$	$161.92~\pm$	114.25 $\pm$	138.93 $\pm$	155.89 $\pm$	155.56 $\pm$	141.80 $\pm$	
1,0 Dioxolane, 2-(1-emylpentyl)-	41.69	157.94 ± 43.48	161.92 ± 22.58	114.25 ± 57.73	138.93 ± 28.48	36.05	20.18	40.43	
2-Pentylfuran	ND <sup>B</sup>	ND <sup>B</sup>	ND <sup>B</sup>	ND <sup>B</sup>	$1.25 \pm 1.32^{\text{A}}$	$1.32 \pm 0.92^{A}$	$1.44 \pm 1.04^{A}$	$0.77 \pm 0.41$	
3,3-Dimethyl-1,2-epoxybutane	0.79 ±	1.44 ±	$1.63 \pm 0.45^{\text{Ba}}$	1.73 ±	$8.53 \pm 3.69^{A}$	11.66 ±	10.73 ±	$7.96 \pm 2.14$	
	$0.14^{Bb}$	0.33 <sup>Bab</sup>		$0.65^{Ba}$		4.03 <sup>A</sup>	5.78 <sup>A</sup>		
Decane Dodecane	$\begin{array}{c} 0.09 \pm 0.10 \\ ND^B \end{array}$	$\begin{array}{l} 0.06 \pm 0.06^{\text{A}} \\ \text{ND}^{\text{B}} \end{array}$	$\begin{array}{c} 0.14 \pm 0.03 \\ ND \end{array}$	$\begin{array}{c} 0.12 \pm 0.01^{\text{A}} \\ \text{ND} \end{array}$	$\begin{array}{c} 0.32 \pm 0.37^{a} \\ 0.36 \pm 0.21^{Aa} \end{array}$	$ ext{ND}^{ ext{Bb}} \ 0.22 \ \pm$	$\begin{array}{c} 0.13 \pm 0.14^{ab} \\ 0.18 \pm 0.20^{ab} \end{array}$	${ m ND}^{ m Bb}$ ${ m ND}^{ m b}$	
					D	0.17 <sup>Aab</sup>	D		
Naphthalene Pentane, 2,3-dimethyl-	$0.22 \pm 0.11^{A}$ $4.86 \pm 3.38^{B}$	$0.12 \pm 0.03^{A} \\ 6.09 \pm 3.84^{B}$	$0.15 \pm 0.02^{A} $ $6.12 \pm 2.09^{B}$	$\begin{array}{l} 0.18 \pm 0.09^{A} \\ 4.06 \pm 1.36^{B} \end{array}$	$ ext{ND}^{ ext{B}}$ 130.32 $\pm$	$0.04 \pm 0.05^{B}$ $139.10 \pm$	$ ext{ND}^{ ext{B}}$ 132.28 $\pm$	$0.07 \pm 0.05  81.05 \pm$	
sec-Butylamine	0.87 $\pm$	$0.89 \pm 0.67^{\text{Ba}}$	$ND^{Bb}$	$ND^{Bb}$	$70.83^{A}$ $16.38 \pm$	$55.46^{A}$ $16.59 \pm$	$76.74^{ m A} \ 12.52 \pm$	$50.46^{A}$ $7.56 \pm 5.21$	
	0.55 <sup>Ba</sup>		D	D	6.84 <sup>A</sup>	5.38 <sup>A</sup>	7.75 <sup>A</sup>		
Tetradecane	$0.21 \pm 0.23$	$0.21 \pm 0.23^{B}$	ND <sup>B</sup>	ND <sup>B</sup>	$0.29 \pm 0.23$	$0.50 \pm 0.12^{A}$	$0.45 \pm 0.07^{A}$	$0.44 \pm 0.09$	
Trimethylene oxide	ND <sup>B</sup>	ND <sup>B</sup>	ND <sup>B</sup>	ND <sup>B</sup>	$8.04 \pm 6.49^{A}$	5.13 ± 3.35 <sup>A</sup>	$3.12 \pm 2.65^{A}$	$3.39 \pm 2.14$	
Subtotal*	$172.36 \pm 44.21^{B}$	$169.59 \pm 44.58^{B}$	$189.75 \pm 36.33^{B}$	$124.37 \pm 57.09^{B}$	$310.29 \pm 105.17^{A}$	$335.20 \pm 62.90^{A}$	$324.83 \pm 94.88^{A}$	$248.55 \pm 41.34^{A}$	
Ketone	11.21	11.50	30.00	37.03	100.17	52.70	J 1.00	11.07	
(+)-2-Bornanone	$ND^B$	$ND^B$	$ND^B$	$ND^B$	$0.52\pm0.21^{\text{A}}$	$0.58 \pm 0.40^{\text{A}}$	$0.52\pm0.22^{\text{A}}$	$0.59 \pm 0.41$	
2,3-Butanedione	$0.91 \pm 0.80$	$\textbf{0.33} \pm \textbf{0.38}$	$ND^{B}$	$1.02\pm1.09^{\text{A}}$	$1.04\pm0.89^{ab}$	ND	$1.18\pm1.09^{\text{Aa}}$	$ND^{Bb}$	
2-Octanone	ND	ND	$ND^{B}$	$ND^{B}$	$\mathrm{ND^b}$	$\mathrm{ND}^\mathrm{b}$	$0.18\pm0.16^{\text{Aa}}$	$\begin{array}{l} 0.14 \pm \\ 0.12^{\text{Aa}} \end{array}$	
5-Hepten-2-one, 6-methyl-	$0.25\pm0.15^{\text{A}}$	$0.18\pm0.11^{\textcolor{red}{A}}$	$0.20\pm0.10^{\text{A}}$	$0.10\pm0.15$	$ND^{B}$	$ND^{B}$	$ND^{B}$	ND	
Acetophenone	$\mathrm{ND}^{\mathrm{Bb}}$	$0.08\pm0.07^{\text{Ba}}$	$0.16\pm0.03^{\text{Ba}}$	$\begin{array}{l} 0.13 \pm \\ 0.07^{\text{Ba}} \end{array}$	$0.17\pm0.09^{Ab}$	$0.35\pm0.05^{\text{Aa}}$	$0.41\pm0.06^{\text{Aa}}$	$0.44 \pm 0.05^{Aa}$	

(continued on next page)

Table 6 (continued)

Items (AU $\times$ 10 <sup>5</sup> )		СВ				WRMD2			
	Day 1	Day 3	Day 5	Day 7	Day 1	Day 3	Day 5	Day 7	
Subtotal*	$1.17\pm0.79$	$0.60 \pm 0.47$	$0.37\pm0.10^{\mathrm{B}}$	$1.26\pm1.04$	$1.74\pm1.02^{ab}$	$0.94\pm0.43^{b}$	$2.30\pm0.94^{\text{Aa}}$	1.19 ± 0.46 <sup>ab</sup>	
Total*	$401.41 \pm 91.03^{a}$	$\begin{array}{l} 293.77 \pm \\ 49.26^{Bab} \end{array}$	$313.26 \pm \\ 36.26^{Bab}$	$257.61 \pm \\88.69^{Bb}$	$505.39 \pm \\178.35$	$559.95 \pm 120.15^{A}$	$561.65 \pm \\ 168.12^{\text{A}}$	$462.59 \pm \\ 62.89^{A}$	

 $<sup>^{</sup>A,B}$  Different letters represent significant differences between broiler and Woorimatdag No. 2 within the same storage period (P < 0.05).

<sup>\*</sup> Subtotal and total amounts of volatile compounds are given as the sum of each detected volatile compounds listed in Supplementary Table S1; ND, not detected.

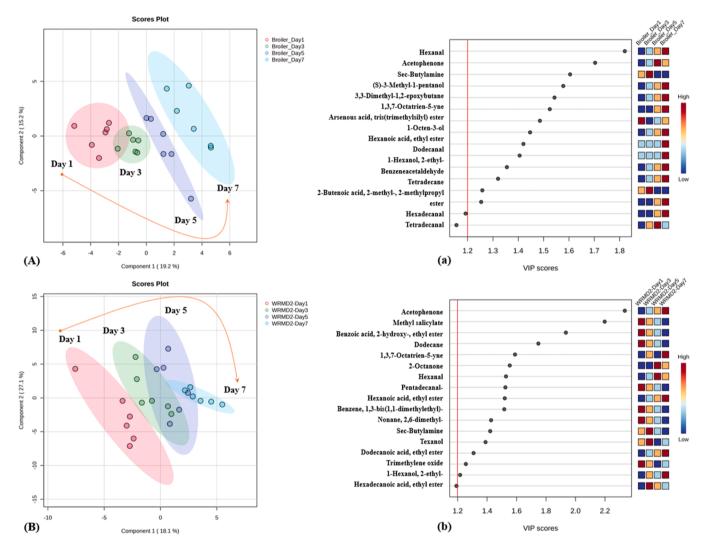


Fig. 1. Partial least squares-discriminant analysis (PLS-DA) and its variable importance in projection (VIP≥1.2) in volatile organic compounds of Woorimatdag No. 2 (WRMD2) and commercial broiler breast meat during cold storage. Capital letters (A, B) and small letters (a, b) indicate the PLS-DA and VIP score, respectively, of Woorimatdag No. 2 and commercial broiler breast meat for 7 days of cold storage.

separation of the VOC cluster was observed between days 3, 5, and 7. This result suggests that relatively small changes could occur during aerobic cold storage in WRMD2 compared with CB. A clearly distinct VOC cluster was observed between CB and WRMD2 throughout the storage period (Fig. 2). Compounds with variable importance in projection (VIP) scores of at least 1.0 may be regarded as potential markers for distinguishing between treatment groups (Jung et al., 2024). Although CB and WRMD2 exhibited obvious separation of VOCs during storage, the types and numbers of variates with high VIP score ( $\geq$ 1.2) were notably distinct: day 1 (15 types including methyl salicylate, 2, 4-decadienal, and hexanal), day 3 (12 types including dodecanal,

acetophenone, and hexanal), day 5 (9 types including naphthalene, tetradecane, and acetophenone), and day 7 (14 types including 2,4-decadienal, decane, and tridecanal).

Correlation between meat quality traits and flavor-related compounds

Drip loss, APC, coliforms, and anserine were linked to the most quality traits in CB, whereas pH, APC, TBARS, VBN, and anserine were linked to the most traits in WRMD2 (Fig. 3). Approximately 1.8 times more flavor-related compounds were correlated with quality traits in CB than WRMD2 (176 vs. 97 compounds;  $r \geq 0.50$  or  $r \leq -0.50$ ). In CB,

 $<sup>^{</sup>a,b}$  Different letters represent significant differences between storage days within the same chicken breed (P < 0.05).

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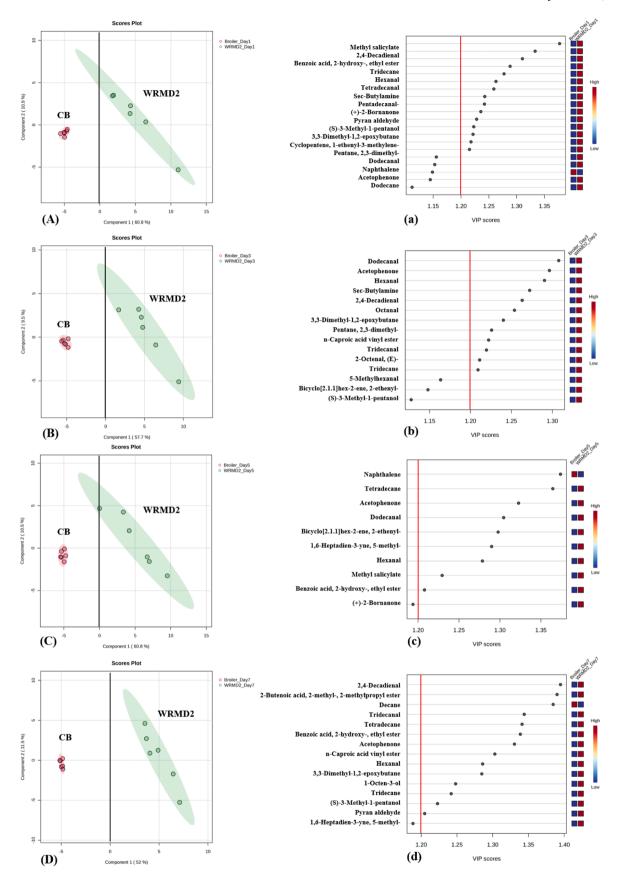


Fig. 2. Partial least squares-discriminant analysis (PLS-DA) and its variable importance in projection (VIP $\geq$ 1.2) in volatile organic compounds of Woorimatdag No. 2 (WRMD2) and commercial broiler (CB) breast meat during cold storage. Capital letters (A, B, C, D) and small letters (a, b, c, d) indicate the PLS-DA and VIP scores, respectively, of volatile organic compounds between Woorimatdag No. 2 and commercial broiler breast meat at each storage day (day 1, 3, 5, 7).

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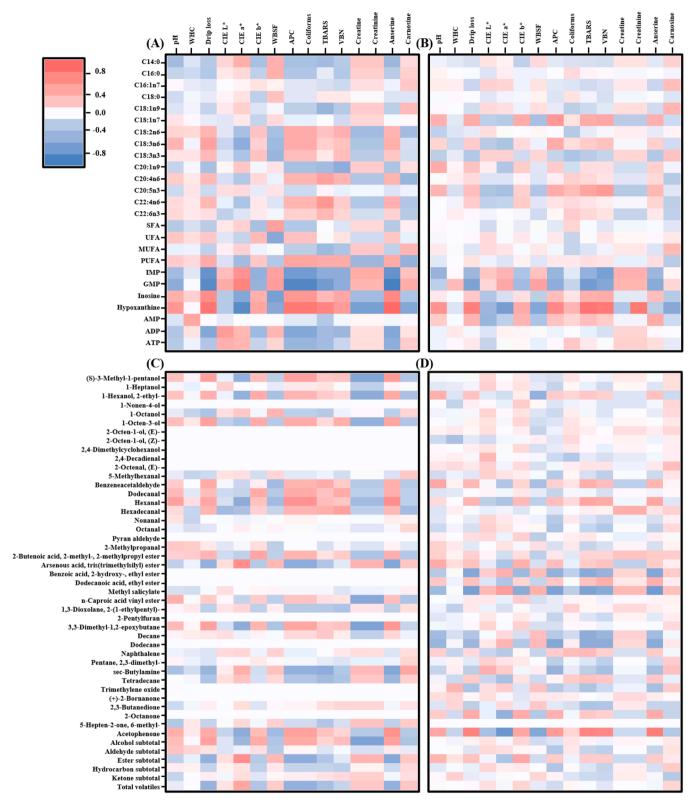


Fig. 3. Correlation analysis between meat quality traits and flavor-related compounds in Woorimatdag No. 2 and commercial broiler breast meat during cold storage. (A) and (B) represent correlation map of meat quality traits and taste-related compounds (fatty acids and flavor nucleotides) of commercial broiler and Woorimatdag No. 2, respectively. (C) and (D) represent correlation map of meat quality traits and volatile organic compounds of commercial broiler and Woorimatdag No. 2, respectively. Red and blue color indicate positive and negative correlation, respectively. The darker the color the higher the correlation. (Supplementary Table S4 provided the detailed correlation coefficient to color this figure).

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drip loss, APC, coliforms, and anserine were significantly negatively correlated with IMP and GMP but were positively associated with inosine and hypoxanthine. Conversely, in WRMD2, IMP and GMP were negatively correlated with drip loss, APC, TBARS, VBN, and anserine, whereas hypoxanthine was positively associated with creatinine and all these traits, excluding anserine. Total alcohols and 1-octen-3-ol were positively correlated with drip loss, b\*-value, APC, coliforms, and anserine in the CB. This is likely because alcohol formation depends on various factors, including microbial activity, lipid oxidation, and protein degradation (Bassam et al., 2022). Moreover, total esters and VOCs were negatively correlated with drip loss, APC, coliforms, and anserine, and positively associated with the a\*-value in the CB. Conversely, in WRMD2, hexanal was positively correlated with TBARS, thereby suggesting that these aldehydes could be derived from lipid oxidation, such as C18:2 and C20:4 (Man et al., 2023). Several esters, including benzoic acid, 2-hydroxy-, ethyl ester and methyl salicylate were negatively correlated with drip loss, APC, TBARS, VBN, and anserine content in WRMD2. Consequently, cold storage alters meat quality and leads to changes in meat-flavor substances and differential flavor profiles. Anserine was found to be highly associated with changes in chicken meat flavor substances compared to other bioactive metabolites, thereby suggesting that a reduction in anserine in meat during storage can result in significant changes in meat flavor.

## Conclusions

In this study, we confirmed that WRMD2 chickens are more susceptible to lipid oxidation during cold storage than CB, as indicated by the higher levels of TBARS and lipid oxidative volatiles, such as 1-octanol, 1-octen-3-ol, hexanal, nonanal, and octanal, whereas protein degradation levels remained comparable between the two types of chickens. Over the 7 days of cold storage, WRMD2 generally exhibited different meat quality compared to CB, including low pH, creatine content, IMP, and GMP content, but high drip loss, dipeptide content, PUFA levels, hypoxanthine, and AMP content. These differences in meat quality may influence the pathways of volatile compound formation, potentially contributing to different aroma profiles in chicken meat. Notably, we identified five key quality parameters (i.e., drip loss, APC, TBARS, VBN, and anserine) that are highly correlated with meat flavor compounds: IMP, GMP, esters, 1-octen-3-ol, and hexanal. Moreover, PLS-DA indicated that the variations in VOC profiles between CB and WRMD2 on each day could be due to the varying types and numbers of specific VOCs with high VIP scores (> 1.2), including methyl salicylate, dodecanal, naphthalene, and 2,4-decadienal. These VOCs are expected to be key aroma compounds that differentiate WRMD2 from CB at a specific storage day. Our findings on the relationships among meat quality, storage stability, and flavor profiles in WRMD2 may serve as preliminary data for further research into flavor improvement and meat quality optimization in native chicken industries globally.

#### **Conflict of Interest**

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

#### Author contribution

Conceptualization: Jung Y, Choo HJ, Jo C, Nam KC, Lee JH, Jang A., Data curation: Jung Y, Oh S, Lee S, Lee HJ, Jang A., Formal analysis: Jung Y, Oh S, Lee S, Lee HJ, Jang A., Methodology: Jung Y, Oh S, Lee S, Lee HJ, Jang A., Validation: Jung Y, Choo HJ, Jo C, Nam KC, Lee JH, Jang A., Investigation: Jung Y, Oh S, Lee S, Lee HJ, Jang A., Funding acquisition: Choo HJ, Jang A., Writing - original draft: Jung Y., Writing - review & editing: Jung Y, Jang A.

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#### Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.psj.2024.104566.

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