Relationship between the Lactic Acid Content and Sour Taste of Broiler Broth and the Broth of Choshu-Kurokashiwa—a Japanese Jidori Chicken

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The present study aimed to determine whether lactic acid content is associated with the intensity of the sour taste of Choshu-Kurokashiwa broth. Chicken broth was prepared using breast (pectoralis major) and leg (thigh + drumstick) meat of male and female Choshu-Kurokashiwa and broilers. These broths were assessed by a screened and trained panel and analyzed for sour taste substances (lactic, citric, pyruvic, malic, succinic, acetic, phosphoric, aspartic, and glutamic acids) and pH. The sensory sour taste was significantly higher in the Choshu-Kurokashiwa breast broth than in the broiler breast broth \((P<0.001)\), and no significant difference was observed in the leg broths among the breeds \((P>0.05)\). Choshu-Kurokashiwa breast broth had a significantly higher lactic acid content than broiler breast broth \((P<0.001)\). The leg meat broth of male Choshu-Kurokashiwa had a significantly lower lactic acid content than that of female Choshu-Kurokashiwa and broiler leg meat \((P<0.01)\). The sensory sour taste score was significantly and positively correlated with lactic acid content in the breast broth \((P<0.001)\), but not in the leg broth \((P>0.1)\). No other organic acids were detected. Phosphoric acid and glutamic acid contents were higher in broiler broth than in Choshu-Kurokashiwa broth for both breast and leg meat \((P<0.001)\). In the breast broth, the aspartic acid content was not significantly different \((P>0.1)\), and in the leg broth, it was higher in broiler and female Choshu-Kurokashiwa broth than in male Choshu-Kurokashiwa broth \((P<0.001)\). The present study suggests that high lactic acid content contributes to the sour taste of the Choshu-Kurokashiwa breast broth and demonstrated that the lactic acid content is an essential indicator for determining the sour taste of Choshu-Kurokashiwa breast meat.

Key words: broiler, Choshu-Kurokashiwa, indigenous chicken, lactic acid, sensory, sourness

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Introduction

Consumers have diverse preferences for chicken meat. In Japan, broilers account for 90% of the chicken meat market (Ministry of Agriculture, Forestry, and Fisheries, 2021). However, some consumers are willing to purchase high-quality Japanese chicken meat, known as “jidori” meat. Even though the price of jidori meat is two to five times that of broiler meat, consumers buy jidori meat because of its unique texture, odor, and taste (Koizumi et al., 1991; Hikichi et al., 2020).

Jidori breeders face the challenge of improving the growth performance of jidori while maintaining its sensory characteristics. The production of jidori is more costly than that of broilers because of the lower stocking densities, longer fattening period, and the cost associated with open non-cage floor rearing systems based on rulings of the Japanese Agricultural Standard (Ministry of Agriculture, Forestry and Fisheries of Japan, 1999). Therefore, it is necessary to improve profitability through weight gain of chicken. However, breeding without considering sensory factors may lead to a loss of palatability.

Some studies (Fujimura et al., 1996; Matsuishi et al., 2005) have focused on the umami taste of jidori meat and reported that the palatability of jidori meat was not based on the strength of the umami taste. Horinouchi et al. (2016) prepared jidori and broiler breast broth and asked the panel to choose one of the five basic tastes to describe the characteristics of the broth. They found that 38% of the panel chose umami taste, and 57% chose sour taste as the characteristic taste of jidori broth, whereas 75% of the panel chose umami taste and only 10% chose sour taste as the characteristic taste of broiler broth. Thus, although tastes other than umami, especially sourness, likely characterize the taste of jidori breast meat, only a few studies have focused on the components that

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contribute to the sour taste.

Organic acids are responsible for the sour taste of foods (Neta et al., 2007). In particular, lactic acid, which is contained in approximately 0.5 g of 100 g chicken meat (Zhang et al., 2012), intensifies the sour taste. In chicken meat, lactic acid accumulates during glycolysis after slaughter (Pösö and Puolanne, 2004). The final lactic acid content of chicken meat is negatively correlated with the chicken growth rate (Berri et al., 2001; Le Bihan-Duval et al., 2008). Given that jidori grows more slowly than broilers because of their genetic characteristics (Rikimaru and Takahashi, 2010; Oosaka et al., 2017), more lactic acid is assumed to accumulate in jidori meat. Therefore, the sour taste of jidori is thought to be caused by the high accumulation of lactic acid. However, the relationship between lactic acid content and sour taste intensity in jidori meat has not been thoroughly studied.

In the present study, I aimed to determine whether lactic acid content is associated with the intensity of the sour taste of jidori meat. Therefore, I prepared chicken broth of Choshu-Kurokashiwa, a jidori breed, and a broiler, and sensory evaluation of both broths was done by a screened and trained panel. The substances responsible for the sour taste were also analyzed.

Materials and Methods

Samples
All chicken breast and leg meat used in this study were purchased directly from the Fukawa Poultry Cooperative commercial farm (Nagato-shi, Yamaguchi, Japan). Both Choshu-Kurokashiwa and broilers were fed at the farm. Because the rearing period of Choshu-Kurokashiwa was divided into sex, both sexes were purchased. The sex of broilers was unknown. Choshu-Kurokashiwa (male and female) and broilers were slaughtered at 84–86, 98–100, and 46–49 days of age, respectively, according to the normal selling age. Eleven males and females each of Choshu-Kurokashiwa and 11 broilers were used for chicken broth preparation. At the farm, right after the slaughter, the left breast (pectoralis major) and right leg meat (thigh + drumstick) were removed from each carcass, and the leg meat was deboned. The meat was then transported to the authors’ laboratory and stored at 4°C for 24 h to complete post-mortem glycolysis. On the day following slaughter, the meat samples were weighed, and the skin and visible fats were trimmed off. The samples were then cut into 2.5×2.5×2.5-cm pieces and minced through a 5-mm plate using an electric mincer (MG-116J, Yasukichi, Gifu, Japan). Finally, 66 minced chicken meat samples (two muscle parts × three chicken groups × eleven individuals) were prepared.

Chicken Broth Preparation
Chicken broth was prepared according to the National Livestock Breeding Center (2005), with some modifications. The minced chicken meat (100 g) and purified water (200 mL) were added to a 500-mL beaker (HARIO SCIENCE, Tokyo, Japan). The beakers were then covered with aluminum foil and placed in a pot of boiling water. After the temperature of the broth reached 85°C, which was monitored using a thermometer (SN3000, Netsuken, Tokyo, Japan), the broth was stirred and heated for another 60 min. The beakers were then removed from the pot, cooled under running water for 1 h, and stored in a refrigerator at 4°C for 18 h. After that, the oil layer was removed from the broth using a 0.3-mm mesh sieve. Next, the obtained chicken broth was filtered through two layers of paper towels (NIPPN PAPER CRECIA, Tokyo, Japan) to remove the meat residues. Finally, the volume of the filtered samples was adjusted to 250 mL with purified water and stored in polypropylene bottles (250 mL; AS ONE, Osaka, Japan) at −30°C until sensory evaluation and chemical component analysis.

General Procedures for the Sensory Evaluation
The temperature of the test room was maintained at 21°C using an air conditioner. The room was illuminated with fluorescent light. The panelists sat at a long table 2 m apart. The sensory panelists were informed of the safety of the chicken meat samples according to the National Livestock Breeding Center (2005).

Screening of Candidates
Descriptive sensory evaluation was performed using a screened and trained panel. This method helps quantify the intensity of sour taste and confirms that sour taste is a characteristic of Choshu-Kurokashiwa broth. In addition, this method prevents the “damping effect” and allows the panel to evaluate the appropriate attribute intensities (Lawless and Heymann, 2013).

Sensory panel screening was performed according to the method described by Sasaki et al. (2012). Forty-five candidates were recruited from the Yamaguchi Prefectural Technology Center for Agricultural and Forestry (Yamaguchi-shi, Yamaguchi, Japan). The screening was performed in three substages: (1) recognition test of five basic tastes, (2) discrimination test of different seasoning concentrations, and (3) discrimination test of differences in food characteristics of odors. As a result, 11 members (six males and five females, aged 28–61 years) were screened. All participants took part in three training sessions and three evaluation sessions.

Generation of Descriptors and References
The panel developed sensory attributes, definitions, and references in three training sessions according to Lawless and Heymann (2013), with some modifications. All training sessions were performed for 1 h each. In the first training session, the panel generated a lexicon of sensory attributes using Kelly’s repertory grid method (Moskowitz, 1983). The sensory panel received three pairs of chicken broths randomly combined from six different broths (two muscle parts × three chicken breeds) and were asked to describe the similarities and differences between the pairs using a Japanese candidate lexicon for the descriptive sensory evaluation of meat (Sasaki et al., 2018). In the second training session, the panelists were exposed to the samples and potential reference foods and reached a consensus on the definition of attributes using the potential references (Table 1). In the final training session, six different samples (two muscle parts × three chicken breeds) and references were provided under the actual testing conditions. The panelists evaluated the samples and received
Table 1. Sensory Attributes, Definitions, and References Used in the Descriptive Analysis

| Attribute              | Definition                          | Reference |
|------------------------|-------------------------------------|-----------|
| Odor intensity         | Total intensity of the odors        | Nr        |
| Roasted-like odor      | Roasted-like odor                   |           |
| Chicken meat-like odor | Chicken meat-like odor              |           |
| Sour odor              | Sour odor                           |           |
| Baked odor             | Baked odor                          |           |
| Flavor/taste intensity | Total flavor/taste intensity        | Nr        |
| Vegetable soup like flavor | Vegetable soup like flavor       |           |
| Sour taste             | Sour taste                          | 0.03 g citric acid per 100 mL of chicken stock |
| Umami taste           | Umami taste                        | 0.13 g MSG per 100 mL of chicken stock |
| Salty taste           | Salty taste                         | 0.16 g NaCl per 100 mL of chicken stock |
| Sweet taste           | Sweet taste                         | 0.5 g sucrose per 100 mL of chicken stock |
| Koku taste            | Complexity, body, mouthfulness, and/or continuity | 0.01 g γ-Glu-Val-Gly per 100 mL of chicken stock |

Nr, no reference

The pH of the broth was measured using a pH meter (DKK-TOA, Tokyo, Japan). For the organic acid and phosphoric acid content measurements, 10 mL of broth was mixed with 14 mL of dH₂O and 6 mL of 10% (w/v) sulfoisalicylic acid. The mixture was centrifuged at 10,000 x g for 15 min. Six milliliters of the supernatant was adjusted to pH 6.8 with KOH and diluted to 20 mL with dH₂O. The solution was filtered through a 0.45-μm membrane (DISMIC 13HP045AN, Toyo Roshi Kaisha, Tokyo, Japan). The organic and phosphoric acid contents were analyzed using high-performance liquid chromatography (LC20AD; Shimadzu, Kyoto, Japan), using a Shim-pack SCR-102(H) column (Shimadzu, Kyoto, Japan). The mobile phase and reaction solution used for the experiment were 5 mM p-toluene sulfonic acid, 100 μM EDTA, and 20 mM Bis-Tris, respectively. The flow rate and oven temperature were 0.8 mL/min and 45°C, respectively.

Free amino acid levels were assessed according to Qi et al. (2017), with some modifications. After protein precipitation and centrifugation using the method used for organic acid analysis, 6 mL of the supernatant was mixed with 2 mL of n-hexane. The water phase was adjusted to pH 3.0 with KOH and diluted to 20 mL with dH₂O. The solution was filtered through a 0.45-μm membrane (DISMIC 13HP045AN, Toyo Roshi Kaisha, Tokyo, Japan) before free amino acid analysis. Free amino acids were analyzed using an automatic amino acid analyzer (L-8900; Hitachi, Tokyo, Japan).

**Statistical Analysis**

All statistical analyses were performed using R version 3.6.1 (R Core Team, 2019). The descriptive sensory score was analyzed using a linear mixed model with the package ‘lme4’. The type of chicken (male or female Choshu-Kurokashiwa or broiler), muscle parts (breast or leg), serving order, and sessions were used as fixed effects. Panelists were used as random effects.

The chemical component characteristics were analyzed by two-way analysis of variance, including variables such as the type of chicken, muscle parts, and interaction. In addition, multiple comparisons were performed using the Tukey–Kramer test using the package ‘multcomp’.

Pearson’s correlation coefficients were evaluated using the
'stats' package to determine the relationship between lactic acid and pH or sour taste score of the pooled results for breast meat (n=33) and the pooled results for leg meat (n=33).

**Results**

Table 2 shows the least-square means of the sensory scores for Choshu-Kurokashiwa broth and broiler broth attributes. The sour taste was the only attribute that was significantly higher in the Choshu-Kurokashiwa broth than in the broiler broth (P<0.001). The effect of breed on sourness was significant, with the sourness of Choshu-Kurokashiwa being significantly greater than that of broilers. The effect of the muscle part was also significant, with the breast being significantly more sour than the leg (P<0.001). Furthermore, the interaction between chicken breed and muscle part was significant (P<0.001), with the Choshu-Kurokashiwa breast being significantly more sour than the broiler breast (P<0.001), whereas no differences in the sourness of the leg meats were observed among the breeds (P>0.1). Except for sourness, the only significant difference among the chicken breeds was umami. Broilers had a stronger umami taste than Choshu-Kurokashiwa in breast meat, but there was no difference in the leg meat. Significant differences were found in odor intensity, chicken meat-like odor, baked odor, flavor/taste intensity, umami taste, and sweet taste between the muscle parts, except for the sour taste. In addition, chicken breed and muscle parts interacted with chicken meat-like odor.

Table 3 shows the weight of chicken meat, acid composition, and pH of chicken broth. A statistically significant interaction for breast meat broth, the lactic acid content was higher in Choshu-Kurokashiwa than in broilers (P<0.001). In contrast, in the leg broth, the lactic acid content of the male Choshu-Kurokashiwa was lower than that of the broilers and female Choshu-Kurokashiwa (P<0.05), but the difference among the chicken breeds was smaller than that in the breast broth. The other organic acids analyzed, namely citric acid, pyruvic acid, malic acid, succinic acid, and acetic acid, were not detected in any of the broths. Phosphoric acid and glutamic acid contents were higher in the broiler broth than in the Choshu-Kurokashiwa broth for both breast and leg meat (P<0.001). In the breast broth, the aspartic acid content was not significantly different (P>0.1), and in the leg broth, it was higher in the broiler and female Choshu-Kurokashiwa broths (P<0.001) than in the male Choshu-Kurokashiwa broth. Among the different broths, the pH was the lowest for the Choshu-Kurokashiwa breast broth, and the leg broth was almost neutral. The breast meat of male and female Choshu-Kurokashiwa was more than 100-g lighter than that of the broilers (P<0.001), whereas the leg meat did not differ significantly among the breeds (P>0.05). Fig. 1 shows the relationship between the lactic acid content and the pH of each broth. The Choshu-Kurokashiwa breast broth had a higher lactic acid content and lower pH than the broiler breast broth. The pH of the leg broth decreased as the lactic acid content increased in both the Choshu-Kurokashiwa and broilers.

Figure 2 shows the relationship between the lactic acid content and the sour taste score of each broth. The Choshu-Kurokashiwa breast broth had a higher lactic acid content and a higher sour taste score than broiler breast broth.

Table 2. Descriptive Sensory Characteristics of Choshu-Kurokashiwa Broth and Broiler Broth

| Attribute                  | Breast       | Leg        | Part | Breed |
|----------------------------|--------------|------------|------|-------|
|                            | CK² BR       | CK BR      |      |       |
|                            | Male Female  | Male Female|      |       |
| Odor                       |              |            |      |       |
| Odor intensity             |              |            |      |       |
| Roasted-like odor          |              |            |      |       |
| Chicken meat-like odor     |              |            |      |       |
| Sour odor                  |              |            |      |       |
| Baked odor                 |              |            |      |       |
| Flavor/taste intensity     |              |            |      |       |
| Vegetable soup like flavor|              |            |      |       |
| Sour taste                 |              |            |      |       |
| Umami taste               |              |            |      |       |
| Salty taste               |              |            |      |       |
| Sweet taste               |              |            |      |       |
| Koku taste                |              |            |      |       |

* Values with different superscripts within a row differ significantly (P<0.05).

1 Samples were evaluated using 15-cm lined scales from 0 cm (not perceived) to 15 cm (extremely strong). Values are expressed as least-square means.

2 CK, Choshu-Kurokashiwa; BR, broiler.

3 Standard error of the mean.

4 ns, not significant; *, P<0.05; **, P<0.01; ***, P<0.001.
As shown in Table 4, negative correlations between lactic acid and pH of the broth were observed for both breast and leg broths ($P<0.001$). Lactic acid content was significantly and positively correlated with the sour taste score for breast broth ($P<0.001$), but not with that for leg broth ($P>0.1$).

### Table 4. Pearson’s Correlation Coefficients between Lactic Acid and pH or Sour Taste Score in Chicken Broth

|                     | Breast       | Leg          |
|---------------------|--------------|--------------|
| Lactic acid - pH    | $-0.82^{***}$ | $-0.919^{***}$|
| Lactic acid - Sour taste score | $0.719^{***}$ | $0.289^{ns}$ |

$^{ns}$, not significant; $^{***}$, $P<0.001$. 

**Kurokashiwa** breast broth and broiler breast broth were plotted for different groups. On the other hand, leg meat broth showed little difference in lactic acid content and sour taste score among the chicken breeds.

As shown in Table 4, negative correlations between lactic acid and pH of the broth were observed for both breast and leg broths ($P<0.001$). Lactic acid content was significantly and positively correlated with the sour taste score for breast broth ($P<0.001$), but not with that for leg broth ($P>0.1$).
Discussion

The sour taste was found to be a crucial characteristic of Choshu-Kurokashiwa breast broth, but not of the leg broth (Table 2). This result is consistent with those of other studies (Rikimaru et al., 2011; Horinouchi et al., 2016). As described above, Horinouchi et al. (2016) reported that sour taste is an essential characteristic of the jidori breast broth; however, they did not mention why the panel perceived the sour taste to be stronger in the jidori breast broth. Rikimaru et al. (2011) fed Hinai-jidori and broilers raised to 22 weeks in the same environment and performed sensory evaluation by a trained panel. They reported no difference in the intensity of sour taste between chicken thigh meat and soup. The findings of these studies are in accordance with the results of the present study, which indicated that the breed and muscle part interacted with the intensity of the sour taste of chicken broth (Table 2).

The chicken breed and muscle part also interacted with the lactic acid content of the broth and the weight of meat (Table 3). The breast meat of Choshu-Kurokashiwa was lighter and had a higher lactic acid content than broilers. Le Bihan-Duval et al. (2008) compared chicken traits of approximately 600 chickens and found a positive correlation between breast muscle weight and ultimate pH. The pH of the breast meat of Thai and Taiwan native chickens, which grow more slowly than broilers, was lower than that of broilers (Jaturasitha et al., 2002; Chunmgoen and Tan, 2015). These chicken breasts presumably had a high lactic acid content, which was highly and negatively correlated with pH. These results are consistent with the results of the present study, where the breast meat of Choshu-Kurokashiwa was lighter, and its broth had a higher lactic acid content than broiler breasts.

The relationship between weight and lactic acid content in the leg broth was similar to that in breast broth. The leg meat of male Choshu-Kurokashiwa was heavier and had a lower lactic acid content than that of the broiler, even though the weights were not significantly different among the breeds (Table 3). Therefore, the effect of the chicken breed on the lactic acid content depends on the part of the chicken to be cooked, and the weight of each part of the chicken needs to be considered rather than the growth rate of the chicken.

The present study suggests that the high lactic acid content of the Choshu-Kurokashiwa breast broth might intensify its sour taste. Chemical components and descriptive analyses showed that the sour taste score was significantly and positively correlated with the lactic acid content in the breast broth (Fig. 2, Table 4). Because the Choshu-Kurokashiwa breast broth had 20% more lactic acid than broiler breast broth (Table 3), the trained panel could probably discriminate the difference in the sour taste intensity between the Choshu-Kurokashiwa and broiler breast broth. Furthermore, no organic acids, other than lactic acid, were detected in the broths, and phosphoric and glutamic acids were more abundant in the broiler broth than in the Choshu-Kurokashiwa broth for breast meat. The aspartic acid content of the breast broth was not significantly different among the breeds. These results suggest that lactic acid is responsible for the sour taste of the Choshu-Kurokashiwa breast broth.

Conversely, no significant correlation was observed between lactic acid content and sour taste score in leg broth (Table 4). Although the lactic acid content of the leg broths differed significantly among the chicken breeds, the differences were small (Table 3). Because the taste threshold of lactic acid is 126 mg per 100 mL (Rotzoll et al., 2006), the panel felt a slightly sour taste and could not discriminate the intensity of the sour tastes.

The pH and lactic acid content interact to affect the intensity of sour taste in foods. For solutions with the same lactic acid content, the lower the pH, the sourer the solution (Sowalsky et al., 1998; Neta et al., 2009). In the present study, the broth with a higher lactic acid content had a lower pH (Fig. 1). Therefore, in the Choshu-Kurokashiwa breast meat broth, both high lactic acid content and low pH may increase the sour taste. However, in 10 mM lactic acid solution, a change from pH 4.5 to pH 3.5, does not affect sour taste intensity (Neta et al., 2009). In addition, the closer the pH of the solution is to neutral, the lower the effect of pH on sour taste intensity (Neta et al., 2009). The present study did not demonstrate a change in sour taste when the pH was changed; however, the difference in pH of the broths in the present study was less than 1 and the pH ranged from 6 to 7. Based on a previous report (Neta et al., 2009), the effect of pH on the sour taste of chicken breast was smaller than that of lactic acid (Table 3).

The sensory evaluation results in this study showed that the umami taste was stronger in the broiler broth than in the Choshu-Kurokashiwa broth for both breast and leg meat (Table 2). This is in agreement with a previous study (Horinouchi et al., 2016). There were significant differences in odor intensity, chicken meat-like odor, baked odor, flavor/taste intensity, umami taste, and sweet taste between the parts, except for sour taste. In addition, an interaction between chicken breed and part of the chicken meat-like odor was observed. Future studies should characterize the volatile compounds and sugars that may contribute to the sensory attributes of Choshu-Kurokashiwa and broiler meat.

Lopez et al. (2011) reported the pH values of breast meat for sexes and showed that female broilers had a slightly lower ultimate pH than males (5.87 vs. 5.94). In the present study, the sex of broilers was unknown. However, because the sex differences in pH of broiler meat broth predicted from the study described by Lopez et al. (2011) was smaller than the differences between broiler and Choshu-Kurokashiwa (Table 3), the effect of sex differences on the sour taste score is probably negligible.

The effects of fatty acids were not considered in this study. According to Kiyohara et al. (2011), arachidonic acid contributes to the palatability of Hinai-jidori chickens. Oleic acid is also a preferred fatty acid in meat (Choe et al., 2010), while docosahexaenoic acid has been shown to suppress sourness and bitterness and enhance umami intensity (Koriyama et al., 2002). In the future, it will be necessary to cook meat with the fat retained and conduct the sensory evaluation.
In conclusion, in the present study, I demonstrated that lactic acid content is an essential indicator for determining the sour taste of Choshu-Kurakashiwa breast broth. On the other hand, for leg broth, the lactic acid content is not important for the Choshu-Kurakashiwa-like taste because the trained panel could not discriminate the intensity of the sour tastes between the Choshu-Kurakashiwa and broiler. Although sour taste in breast meat is one of the major characteristic tastes of jidori breast meat, preference of the consumers for the sour taste derived from lactic acid of jidori meat is unclear. Future studies should investigate the effect of the lactic acid content of jidori meat on consumer preference before using it as an indicator for breeding jidori.

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Author Contributions

The author confirms sole responsibility for the following: study conception and design, data collection, analysis and interpretation of results, and manuscript preparation.

Conflict of Interest

The author declares no conflict of interest.

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