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Original Research Article

Effect of High-Voltage Electrostatic Field-Assisted Curing on the Quality of Fermented Niuganba

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Abstract: This study investigated the effects of High Voltage Electric Field (HVEF)-assisted curing on the quality characteristics of fermented Niuganba. The results demonstrated that compared with conventional refrigerator curing, HVEF-assisted cured beef exhibited faster decreases in pH value, water activity, and product yield during the fermentation process. Both TBARS and TVB-N values maintained significantly lower levels ($P < 0.05$). After fermentation, the hardness and chewiness showed significant reductions ($P < 0.05$), while redness (a^* value) increased remarkably ($P < 0.05$). The comprehensive sensory evaluation score was significantly improved ($P < 0.05$), accompanied by a notable increase in free amino acid content ($P < 0.05$). E-nose analysis effectively distinguished flavor differences between curing methods, with HVEF-treated samples showing enhanced response values in sensors detecting inorganic/organic sulfides, alcohols, aldehydes, and ketones. These findings collectively indicate that HVEF-assisted curing can effectively improve color attributes, texture, and flavor characteristics of fermented Niuganba while enhancing its nutritional value. This research provides both theoretical foundation and innovative insights for the application of HVEF technology in fermented meat products processing.

Keyword: High-voltage electrostatic field, Curing, Fermentation, Niuganba, Quality.

INTRODUCTION

Niuganba is a traditional fermented meat product originating from Southwest China, produced through sequential curing and fermentation of fresh beef. Renowned for its savory flavor and high nutritional value, it has gained considerable consumer popularity (Yin F, *et al*., 2024). However, conventional production methods often suffer from prolonged curing periods, inconsistent product quality, and inadequate safety assurance, which substantially hinder industrial-scale production efficiency and product standardization. With evolving consumer demands for premium-quality meat products with shortened processing cycles, there is an urgent need to develop efficient and controllable processing aids. Such innovations could optimize production efficiency while enhancing the quality attributes of Niuganba, addressing both market expectations and industrial challenges.

The High Voltage Electrostatic Field (HVEF), a non-thermal processing technology, offers advantages such as high efficiency, energy-saving features, and operational simplicity. An HVEF system typically comprises three key components: a high-voltage generator, a DC power supply, and a treatment chamber. The DC power supply converts low voltage into high voltage through the generator, which is then applied to electrodes in the chamber to generate an electrostatic field (Wang L P, *et al*.,2024). Studies have demonstrated that HVEF-assisted meat curing accelerates salt penetration, shortens processing duration, and enhances textural and flavor properties (Sha K, *et al*., 2022, Chen C B, *et al.*, 2025).

Fermentation critically determines the final quality of Niuganba. Starter cultures have been widely adopted in industrial fermented meat production to standardize processes, reduce fermentation time, and improve product quality and safety (Shang H, *et al*.,

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2024). Notably, Zhou Y *et al*. (2025) revealed that pulsed electric field pretreatment of goose meat prior to inoculated fermentation not only accelerated fermentation but also enhanced textural attributes, flavor profiles, and microbial safety of the final product, suggesting that optimizing meat quality before fermentation positively influences subsequent processing outcomes.

Based on these insights, this study employs meat samples pretreated with moderate HVEF-assisted curing (8 kV, 15 h, 4°C) based on preliminary experiments as raw materials. These samples were inoculated with starter cultures to produce fermented Niuganba, aiming to systematically evaluate the impacts of HVEF-assisted curing on the quality characteristics of fermented Niuganba.

1. MATERIALS AND METHODS

1.1. Materials

Fresh yellow cattle hind leg meat, obtained within 12 hours post-slaughter under hygienic conditions, was purchased from Rongrong Supermarket (Yibin, China). Chemical reagents (Boric acid, Methyl Red, Trichloroacetic acid, Bromocresol Green, and 2-Thiobarbituric Acid) were procured from Macklin Biochemical Technology Co., Ltd. (Shanghai, China).

1.2. Preparation of fermented Niuganba

Niuganba was prepared following the method described by Wen *et al*. (2021) with modifications. Fresh yellow cattle hind leg meat from acid-removed yellow cattle was trimmed to remove visible connective tissue, surface moisture was absorbed using filter paper, and standardized into blocks (10 cm × 5 cm × 5 cm). Six meat blocks were selected for curing and fermentation. A curing mixture containing 3% (w/w) NaCl, 0.3% (w/w) glucose, 0.1% (w/w) sodium D-isoascorbate, and 0.015% (w/w) sodium nitrite relative to meat weight was uniformly applied to the meat surfaces. The mixture was massaged until fully integrated with meat exudate. Samples were wrapped in aluminum foil and randomly divided into two groups (3 blocks per group, with the completion of initial curing designated as cured 0d). The first group (RM group) underwent curing in a 4°C constant-temperature refrigerator. The second group (HM group) was treated using a HVEF system (8 kV, 15 h, 4°C) provided by Induce Induction Technology Co., Ltd. (Wuxi, China). After 15 h of curing, aluminum foil was removed, and samples from both groups were vacuum-sealed in labeled bags. All vacuum-packed samples were subsequently cured at 4°C for an additional 3 d (designated as fermentation day 0). Following the completion of curing, meat samples from the RM and HM groups were subjected to bacterial inoculation. A mixed lyophilized culture powder (0.02%, w/w) containing *Lactobacillus sakei, Staphylococcus xylosus, Staphylococcus carnosus, Pediococcus pentosaceus, and Debaryomyces hansenii* was pre-activated by rehydration in physiological saline at 37°C for 24 h prior to inoculation. After the inoculation process, the meat samples were individually suspended in fermentation chambers (supplied by Shanghai Yuejin Medical Equipment Co., Ltd., Shanghai, China) for controlled fermentation. The fermentation conditions were programmed as follows: temperature 20℃ and relative humidity (RH) 90% on fermentation day 0; temperature 20℃ and RH 88% on fermentation day 2; temperature 18℃ and RH 88% on fermentation day 5, with these parameters maintained until the termination of fermentation on day 9. Upon completion of the 9-day fermentation process, the final products from both RM and HM groups were collected for subsequent quality index analyses.

1.3. Determination of pH

The pH value was determined according to the method described by Hou C P *et al*. (2025) with modifications. Briefly, 2.0 g of minced meat sample was transferred into a beaker, mixed with 18 mL of physiological saline, and homogenized using a magnetic stirrer for 20 min. The pH measurement was subsequently conducted at room temperature using a calibrated pH meter. Triplicate measurements were performed for each group, and mean values were calculated as final results.

1.4. Determination of Water activity (Aw)

The water activity (Aw) was determined according to the *Chinese National Food Safety Standard: Determination of Water Activity in Foods* (GB 5009.238-2016). Briefly, 1.0 g of meat sample was transferred to a sample dish and analyzed using a calibrated water activity analyzer under ambient laboratory conditions.

1.5. Determination of Yield

The yield of Niuganba was measured according to the method described by Shi S *et al*. (2021) with modifications, and was calculated using Equation (1), where M_0 represents the weight (g) of the beef sample before curing, and M_t denotes the weight (g) of the beef sample after curing and fermentation for a duration of t days.

$$Yield\ (\%) = \frac{M_t}{M_0} \times 100\% \quad (1)$$

1.6. Determination of thiobarbituric acid (TBARS)

The determination of thiobarbituric acid (TBARS) was performed following the method described by Chen G B *et al*. (2025) with modifications. Briefly, 5.0 g of minced meat sample was weighed and homogenized with 25 mL of 7.5% trichloroacetic acid solution (containing 0.1% EDTA), followed by centrifugation at 4000 rpm for 4 min at 4℃ The supernatant was subsequently filtered, and 5 mL of the filtrate was mixed with 5 mL of 0.02 mol/L thiobarbituric acid (TBA) solution. The mixture was incubated in a boiling water bath at 100℃ for 30 min, cooled to room temperature, and the absorbance values at 532 nm and 600 nm were measured using a spectrophotometer.

$$\text{TBARS（mg/kg）} = 4.65 \times （A_{532} - A_{600}） \quad (2)$$

1.7 Determination of Total Volatile basic nitrogen (TVB-N)

The total volatile basic nitrogen (TVB-N) content in meat samples was determined in accordance with the *Chinese National Food Safety Standard: Determination of Total Volatile Basic Nitrogen in Foods* (GB 5009.228-2016).

1.8. Sensory evaluation

Sensory evaluation was conducted following the method described by Aykın-Dinçer *et al*. (2019) with modifications. Niuganba samples fermented for 9 days were subjected to sensory analysis by a panel of 10 food science graduate students (5 males and 5 females, mean age 24 years). Prior to evaluation, panelists underwent a two-day training session on meat product sensory analysis to ensure accurate identification of Niuganba's sensory attributes. For evaluation, fermented Niuganba samples were cooked at 90°C for 15 min, cut into 1 cm^3 cubes, and randomly arranged on white plastic plates. Samples from different treatment groups (three replicates per group) were labeled with three-digit random codes and presented to panelists in randomized order. Each panelist evaluated 12 samples, with 10-min breaks after every three samples to prevent sensory fatigue. Evaluations were performed using a gradient scoring system based on color, taste, aroma, chewiness, and overall acceptability.

1.9. Determination of texture

Texture analysis was performed according to the method described by Chang H J *et al*. (2014) with modifications. Fermented Niuganba samples were trimmed into uniform cubes (1.5 cm × 1.5 cm × 1.5 cm) and analyzed using a TA-XT Plus Texture Analyzer (Stable Micro Systems, UK) equipped with an HDP/USR probe. Testing parameters were set as follows: pre-test speed 1.00 mm/s, test speed 5.00 mm/s, and post-test speed 5.00 mm/s, with a trigger force of 5.0 g and 50% compression strain applied perpendicular to the muscle fiber orientation.

1.10. Determination of color

The determination of Color was conducted following the method of Liu S X *et al*. (2019) with modifications. Fermented Niuganba samples were sectioned from their surface portion and analyzed using an UltraScanVIS benchtop colorimeter (Xinlian Chuangzuo Electronics Co., Ltd., Shanghai, China). Instrument calibration was performed against a standard white plate prior to each measurement session. Triplicate measurements were performed for each sample, with mean values calculated for color parameters expressed as L^* (lightness), a^* (redness), and b^* (yellowness).

1.11. Determination of free amino acid content

The free amino acid content in meat samples was determined using an L-8900 fully automatic amino acid analyzer (Hitachi Co., Ltd., Tokyo, Japan) in accordance with the *Chinese National Food Safety Standard: Determination of Amino Acids in Foods* (GB 5009.124-2016).

1.12. Determination of E-nose

Electronic nose (E-nose) analysis was performed according to the method described by Liu M *et al*. (2021) with modifications. Briefly, 3.0 g of minced meat sample was placed in a headspace vial and equilibrated at 40°C for 40 min in a water bath prior to analysis using a PEN3 E-nose system (Shanghai Angshen Intelligent Technology Co., Ltd., Shanghai, China). Operational parameters were set as follows: injection preparation time 10 s, sampling duration 70 s, gas flow rate 0.4 L/min, and purge time 100 s, with triplicate measurements conducted for each sample group.

2. Statistical Analysis of Data

Experimental data were processed using Microsoft Excel 2021 (Microsoft Corp., USA), with statistical significance analyzed via IBM SPSS Statistics 25 (IBM Corp., USA), where differences at $P<0.05$ were considered statistically significant. Graphical representations were generated using OriginPro 2021 (OriginLab Corp, USA), and all results are presented as mean values ± standard deviation (SD) derived from triplicate measurements.

3. RESULTS AND DISCUSSION

3.1 Effect of HVEF-assisted curing on pH of Niuganba during fermentation

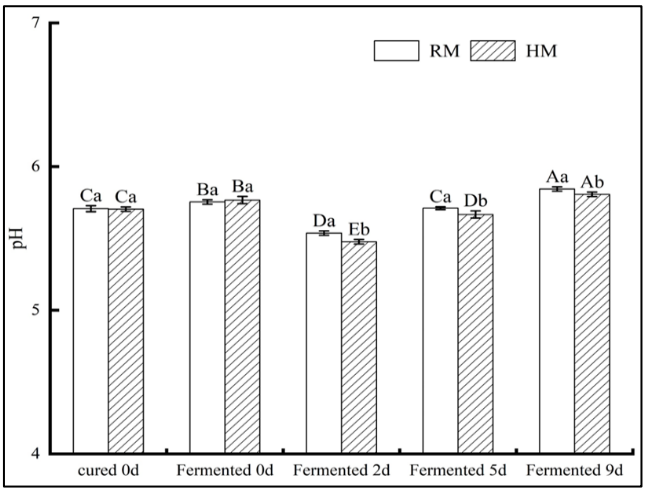

Figure 1: Effect of HVEF-assisted curing on pH of Niuganba during fermentation

Notes: Significant differences (P<0.05) between the same treatment across different time points are denoted by superscript capital letters, while differences at the same time point across different treatments are indicated by superscript lowercase letters (P<0.05).

The effect of HVEF-assisted curing on pH variations during Niuganba fermentation is illustrated in Figure 1. Both treatment groups exhibited an initial decline followed by a gradual increase in pH values with prolonged fermentation time. Throughout the curing phase, no significant differences ($P > 0.05$) were observed between the pH values of HM and RM groups. On fermentation day 2, the pH values of both groups reached their minimum, with the RM group decreasing to 5.54 and the HM group demonstrating a statistically significant reduction to 5.48 ($P < 0.05$) compared to the RM group. Previous studies have indicated that during natural meat fermentation, acid-producing bacteria proliferate exponentially as microorganisms enter the logarithmic growth phase, intensifying carbohydrate metabolism. This metabolic activity leads to organic acid accumulation (primarily lactic acid), consequently reducing meat pH (Rubio R *et al*., 2013). The rapid pH decline in both RM and HM groups can be primarily attributed to the potent acid-producing capacity of *Lactobacillus sakei* within the inoculated mixed starter culture. Simultaneously, microbial competition between non-starter populations and acidogenic bacteria during fermentation critically influenced pH dynamics. The HM group, pretreated with HVEF during curing, likely exhibited reduced initial contamination by non-starter microbial populations prior to inoculation. This preconditioning enabled dominant acid-producing strains in the starter culture to preferentially utilize nutrients for accelerated acidogenic metabolism post-inoculation (Li D P, *et al*., 2017).

3.2 Effect of HVEF-assisted curing on Aw of Niuganba during fermentation

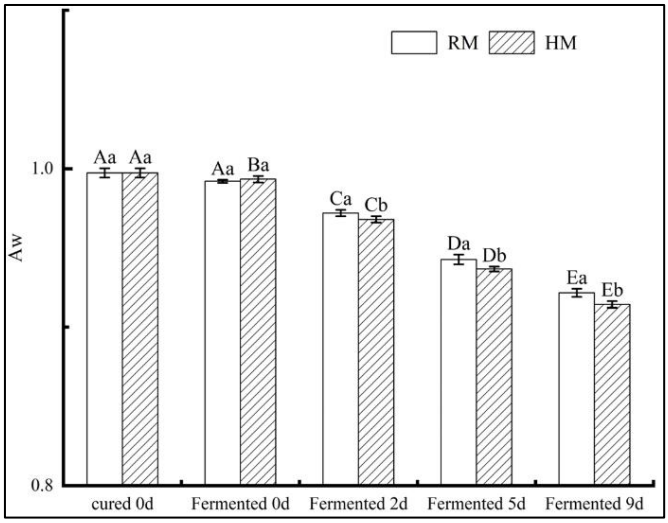

Figure 2: Effect of HVEF-assisted curing on Aw of Niuganba during fermentation

The variation in water activity (Aw) reflects microbial proliferation during meat fermentation and is intrinsically correlated with product yield. As shown in Figure 2, HVEF-assisted curing significantly influenced Aw dynamics during Niuganba fermentation. Both RM and HM groups exhibited a consistent downward trend in Aw values throughout the curing-fermentation period, attributable to microbial growth, reproduction, and metabolic water consumption – a pattern consistent with Aw variations observed in fermented mutton sausages by Sun X Y*et al*. (2020). At curing day 0, the highest Aw value (0.997) was recorded. No significant intergroup difference ($P > 0.05$) was detected during initial fermentation stages. By fermentation day 2, the HM group demonstrated a statistically significant reduction in Aw (0.968, $P < 0.05$) compared to the RM group, with this differential persisting through day 9. Previous studies suggest that as meat pH approaches protein isoelectric points during fermentation, protein gelation occurs, diminishing water retention capacity and facilitating free water diffusion from the matrix, thereby accelerating Aw reduction. Combined with pH observations, this mechanism suggests that HVEF pretreatment in HM group accelerated pH decline, subsequently driving more rapid Aw decrease through enhanced protein-water interactions.

3.3 Effect of HVEF-assisted curing on the yield of Niuganba during fermentation

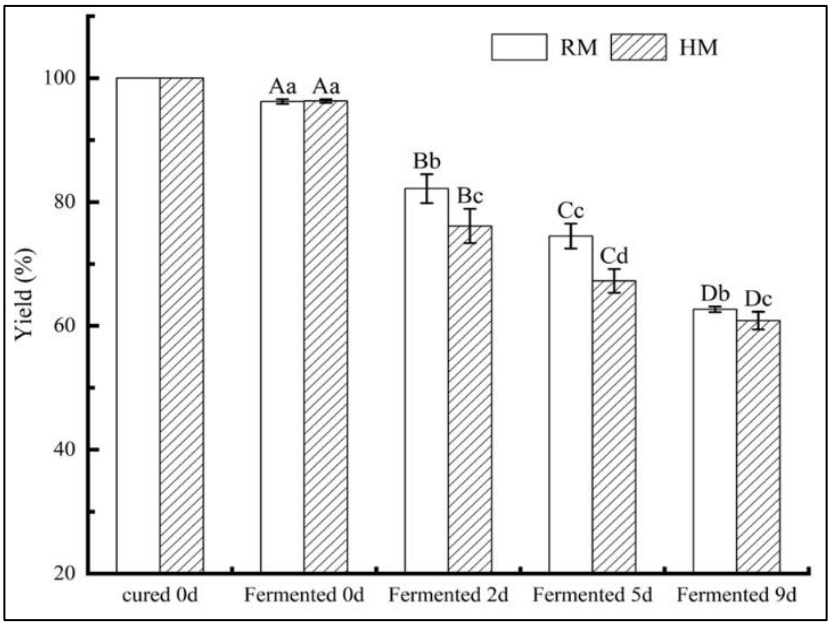

Figure 3: Effect of HVEF-assisted curing on the yield of Niuganba during fermentation

The product yield, defined as the percentage ratio of processed meat weight to raw material weight, serves as a critical indicator for evaluating raw material utilization efficiency and production cost-effectiveness in meat processing. Excessively high yields may indicate incomplete fermentation, resulting in compromised flavor and texture quality alongside reduced storage stability, while excessively low yields reflect poor material utilization and increased production costs. For fermented Niuganba, optimal yield is typically maintained within 55%–65% to balance these factors. As shown in Figure 3, HVEF-assisted curing significantly influenced yield patterns during Niuganba fermentation, with both RM and HM groups exhibiting decreasing yield trends over time. No intergroup difference ($P > 0.05$) was observed at fermentation day 0. By day 2, the HM group demonstrated significantly lower yield ($P < 0.05$) compared to RM, a pattern sustained until process termination. The HM group ultimately achieved the lowest final yield (60.84%) at day 9. Previous studies (Zhang M C *et al*., 2024) have established that meat product yields decrease proportionally with reductions in water activity and pH. This suggests that accelerated pH and Aw declines in HM samples likely drove rapid yield reduction, enabling earlier attainment of fermentation endpoints. These findings collectively indicate that HVEF-assisted beef curing effectively shortens the fermentation duration required for Niuganba production.

3.4 Effect of HVEF-assisted curing on thiobarbituric acid value of Niuganba during fermentation

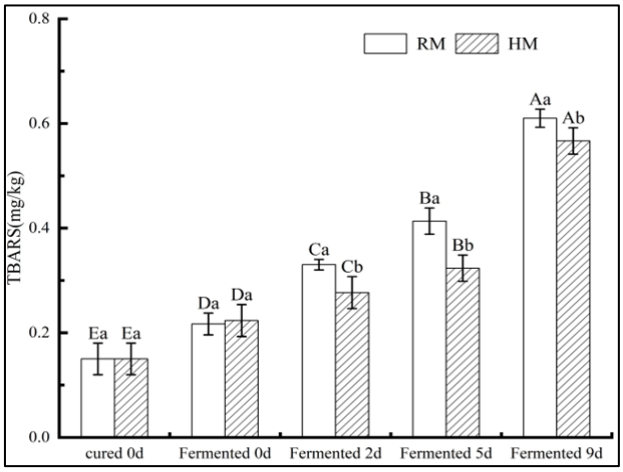

Figure 4 Effect of HVEF-assisted curing on thiobarbituric acid value of Niuganba during fermentation

The thiobarbituric acid reactive substances (TBARS) value serves as a critical indicator for evaluating the formation of peroxidation products such as malondialdehyde (MDA), indirectly reflecting the extent of lipid oxidation. Lipid peroxidation in meat products leads to quality deterioration, characterized by off-odors, color alterations, reduced nutritional value, and potential health risks (Özünlü O, *et al*., 2018). TBARS values, which primarily quantify secondary oxidation products of free fatty acids, are widely employed to assess lipid oxidation levels during meat processing. In this study, TBARS values represented the cumulative MDA content generated from unsaturated fatty acid decomposition. As shown in Figure 4, the lowest TBARS value (0.15 mg/kg) was recorded at curing day 0. Both HM and RM groups exhibited progressive increases in TBARS values during fermentation. No significant difference ($P > 0.05$) was observed between the two groups in TBARS values at fermentation day 0. However, by day 2, the HM group demonstrated significantly lower TBARS values (0.28 mg/kg, $P < 0.05$) compared to the RM group, with this differential maintained throughout subsequent fermentation stages. The observed TBARS increase can be attributed to lipid hydrolysis during fermentation, which elevates free fatty acid content. Subsequent oxidation of these fatty acids generates hydroperoxides that serve as precursors for MDA formation, thereby driving TBARS accumulation (Huang Y, *et al*., 2013). Previous studies have demonstrated that *Lactobacillus sakei*, *Pediococcus pentosaceus*, and *Staphylococcus xylosus* exhibit potent antioxidant activities capable of inhibiting lipid oxidation during fermentation processes (Wang J Y, *et al*., 2020; Zhang Y, *et al*., 2017). This suggests that compared to the RM group, HVEF pretreatment in the HM group may have created a more favorable microenvironment for the proliferation and metabolic activity of these starter cultures. Consequently, at equivalent fermentation time points, the HM group likely sustained higher populations of antioxidant-active microbial strains, thereby effectively suppressing lipid peroxidation and maintaining lower TBARS values. Collectively, these findings indicate that HVEF-assisted beef curing effectively mitigates lipid oxidation during fermentation, resulting in reduced TBARS accumulation.

3.5 Effect of HVEF-assisted curing on total volatile base nitrogen content of Niuganba during fermentation

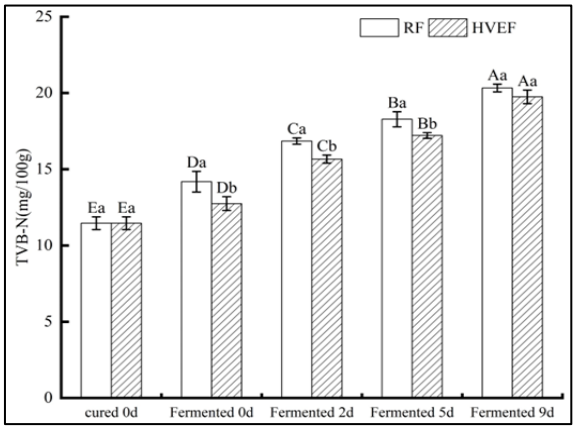

Figure 5 Effect of HVEF-assisted curing on total volatile base nitrogen content of Niuganba during fermentation

Total volatile basic nitrogen (TVB-N), comprising alkaline metabolites such as ammonia and biogenic amines derived from microbial degradation of nitrogen-containing compounds including amino acids and proteins, was analyzed for its dynamics during Niuganba fermentation under HVEF-assisted curing (Figure 5). The initial TVB-N content at curing day 0 was 11.46 mg/100 g. At fermentation day 0 (corresponding to 3 days of curing), the HM group exhibited a significant reduction in TVB-N compared to the RM group ($P < 0.05$), which was attributed to the inhibitory effect of HVEF treatment on microbial proliferation. This observation aligns with findings by Qi M Y, *et al*. (2022), who reported suppressed TVB-N accumulation in HVEF-treated salmon during storage. Throughout fermentation, both groups displayed progressive TVB-N increases, consistent with decarboxylation of free amino acids by decarboxylase-positive bacteria during proteolysis (Ikonić P, *et al*., 2019). From fermentation day 2 onward, the HM group consistently maintained significantly lower TVB-N levels than the RM group ($P < 0.05$). Suzzi G *et al*. (2003) proposed that rapid pH decline during fermentation inhibits amine-producing microorganisms, a mechanism supported by our pH data suggesting accelerated acidification in the HM group contributed to its reduced TVB-N accumulation. Notably, all TVB-N values remained below the maximum acceptable threshold (30 mg/100 g) proposed by Hu Y Y *et al*. (2021), confirming preserved product quality in both groups throughout fermentation.

3.6 Sensory evaluation

Table 1: Sensory score

Sensory quality	RM	HM
Color	7.37±0.04^{b}	7.68±0.04^{a}
Flavor	7.52±0.08^{b}	8.22±0.05^{a}
Chewiness	8.20±0.03^{b}	9.16±0.08^{a}
Organizational status	8.72±0.06^{b}	8.88±0.05^{a}
Overall acceptance	8.80±0.02^{b}	9.27±0.05^{a}

Notes: Significant differences ($P < 0.05$) between different columns within the same row are denoted by distinct lowercase letters.

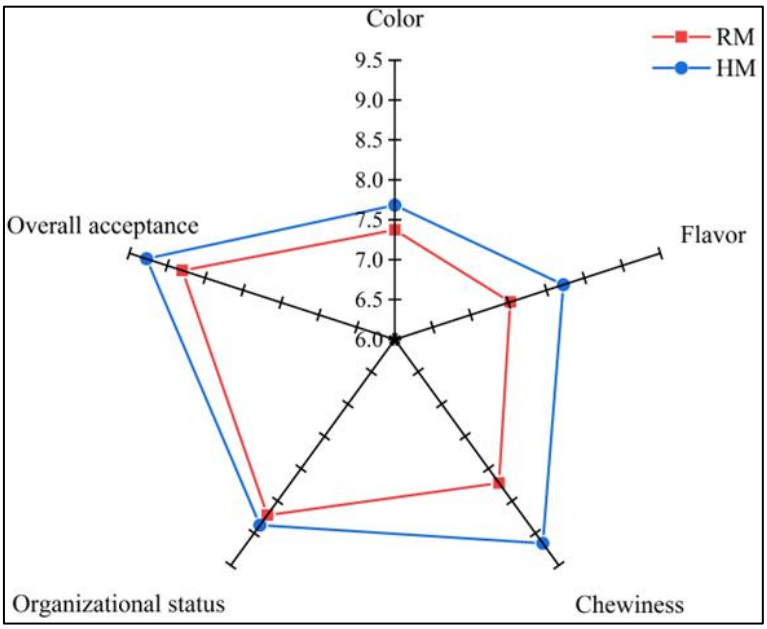

Figure 6: Sensory score radar map

Meat product quality is defined by consumer acceptability (Gao X Q, *et al*., 2014). Sensory evaluation serves as a critical methodology for assessing meat product attributes, enabling comprehensive analysis of color, flavor, chewiness, and other characteristics to identify products that better meet consumer preferences. Sensory evaluation results for fermented Niuganba produced by both treatments are presented in Table 1 and Figure 6. The analysis revealed statistically significant differences ($P < 0.05$) in sensory scores between RM and HM groups across all parameters: color, flavor, chewiness, Organizational status, and overall acceptability. For color evaluation, both groups exhibited uniform light reddish hues, with HM samples achieving significantly higher scores (7.68 vs. 7.37 for RM, $P < 0.05$). Flavor profiles showed similar differentiation, with HM group scoring significantly higher ($P < 0.05$) than RM group (7.52), indicating enhanced fermentation-derived aroma complexity and typicality in HVEF-treated samples. The most pronounced intergroup

difference occurred in chewiness assessment, where HM group achieved 9.16 versus 8.20 for RM group, demonstrating that HVEF-assisted curing facilitates superior textural development during fermentation. Significant advantages in Organizational status and overall acceptability were also observed in HM group ($P < 0.05$), confirming that HVEF pretreatment significantly improves both the organoleptic quality and structural properties of fermented Niuganba.

3.7 Effect of HVEF-assisted curing on texture properties of fermented Niuganba

Table 2: Effect of HVEF-assisted curing on texture properties of fermented Niuganba

Treatment method	Hardness/g	Springiness /mm	Chewiness /g
RM	16583.88 ± 372.94^{a}	0.54 ± 0.03^{a}	6585.96 ± 192.62^{a}
HM	15595.34 ± 297.24^{b}	0.53 ± 0.03^{a}	5608.54 ± 212.70^{b}

Notes: Significant differences ($P < 0.05$) between different rows within the same column are indicated by distinct lowercase letters.

The influence of HVEF-assisted curing on textural properties of fermented Niuganba is presented in Table 2. Compared to the RM group without HVEF treatment, the HM group exhibited significant reductions in hardness (5.96%) and chewiness (14.84%) ($P < 0.05$), while no intergroup difference in springiness was observed ($P > 0.05$). These results indicate that HVEF pretreatment combined with mixed-strain fermentation effectively improves textural softness and palatability of Niuganba. Mechanistically, HVEF treatment induces electroporation in muscle cell membranes, leading to structural weakening and tenderization (Kantono K, *et al*., 2019). Concurrently, proteolytic degradation of myofibrillar proteins by endogenous muscle enzymes and microbial-derived exogenous proteases during fermentation contributes to textural modification (Fang Y, *et al*., 2016). Previous studies suggest that endogenous proteases dominate early-stage protein hydrolysis, while microbial proteases gradually become predominant as fermentation progresses. It is hypothesized that HVEF pretreatment enhances endogenous protease activity during initial fermentation stages, accelerating myofibrillar protein degradation. This process generates bioavailable nutrients (e.g., peptides, free amino acids) that promote microbial proliferation and subsequent secretion of exogenous proteases. The synergistic proteolytic action disrupts intramuscular connective tissue integrity and reduces protein binding forces (Chen X, *et al.*, 2021), ultimately yielding a softer texture in HVEF-treated Niuganba.

3.8 Effect of HVEF-assisted curing on color of fermented Niuganba

Table 3: Effect of HVEF-assisted curing on color of fermented Niuganba

Treatment method	L^*	a^*	b^*
RM	49.40 ± 0.44^{a}	8.93 ± 0.29^{b}	5.34 ± 0.23^{a}
HM	49.81 ± 0.88^{a}	10.42 ± 0.32^{a}	5.48 ± 0.19^{a}

Notes: Significant differences ($P < 0.05$) between different rows within the same column are indicated by distinct lowercase letters.

Color, a critical quality indicator of meat products, was significantly influenced by HVEF-assisted curing in fermented Niuganba as shown in Table 3. Compared to the RM group without HVEF treatment, the HM group exhibited significantly higher a^* values (redness) ($P < 0.05$), while no intergroup differences were observed in L^* (lightness) or b^* (yellowness) values ($P > 0.05$). The characteristic bright red color of fermented meat products primarily originates from nitrosylmyoglobin formation. During fermentation, starter cultures with nitrite reductase activity reduce nitrite to nitric oxide, which subsequently binds to the central iron ion of myoglobin to form nitrosylmyoglobin (Luo Y L, *et al*., 2020). Previous studies have demonstrated that lower pH enhances both the stability and yield of nitrosylmyoglobin in fermented meats, thereby intensifying product coloration (Dasiewicz K, *et al*., 2024). This may be attributed to the combined effect of HVEF pretreatment and mixed starter cultures maintaining lower pH levels during Niuganba fermentation, potentially promoting greater nitrosylmyoglobin accumulation in the HM group compared to RM, resulting in elevated a^* values.

3.9 Effect of HVEF-assisted curing on free amino acid content of fermented Niuganba

Table 4: Effect of HVEF-assisted curing on free amino acid content of fermented Niuganba

Name of amino acid	Treatment method	
	RM	HM
Asp	86.88 ± 0.58^{b}	90.47 ± 0.85^{a}
Glu	152.65 ± 0.85^{b}	157.51 ± 1.04^{a}
Thr	40.92 ± 0.36^{b}	42.13 ± 0.47^{a}

Ser	39.82±0.41[b]	41.81±0.48[a]
Gly	40.62±0.20[b]	45.47±0.2[a]
Ala	51.5±0.56[b]	55.5±0.11[a]
Pro	37.83±0.41[b]	41.2±0.35[a]
Val	35.57±0.27[b]	36.54±0.09[a]
Met	24.24±0.43[a]	24.73±0.43[a]
Ile	30.42±0.21[a]	30.37±0.12[a]
Leu	69.17±0.72[b]	72.02±0.64[a]
Phe	42.7±0.64[a]	43.84±0.35[a]
Lys	76.87±0.41[a]	78.82±0.64[a]
Tyr	29.65±0.21[b]	30.54±0.31[a]
His	37.25±1.24[b]	37.89±0.57[a]
Arg	53.52±0.22[b]	55.38±0.21[a]
Flavor AA	239.54±1.36[b]	247.97±1.88[a]
Sweet AA	210.68±1.68[b]	226.11±0.83[a]
Bitter AA	399.38±0.66[b]	410.13±2.53[a]
Total free AA	849.60±3.35[b]	884.22±5.12[a]

Notes: Significant differences ($P < 0.05$) between different columns within the same row are denoted by distinct

The effect of HVEF-assisted curing on free amino acid (FAA) content in fermented Niuganba is presented in Table 4. Both RM and HM groups contained 16 FAAs, including 8 essential amino acids and 8 non-essential amino acids. The HM group exhibited a total FAA content of 884.22 mg/100 g, representing a significant 3.92% increase compared to the RM group (849.60 mg/100 g, $P < 0.05$). Previous studies have established that FAA accumulation during meat fermentation primarily results from proteolytic activities mediated by endogenous muscle enzymes and microbial-derived proteases (Aro J M, *et al*., 2009). These findings suggest enhanced proteolysis occurred in HM samples, a conclusion corroborated by textural property observations. This phenomenon may be attributed to HVEF-assisted curing promoting the activity of both endogenous proteases and microbial exoproteases, thereby significantly elevating FAA liberation during Niuganba fermentation.

3.10 Effects of HVEF-assisted curing on taste characteristics of free amino acids and TAV of fermented Niuganba

Table 5: Effects of HVEF-assisted curing on taste characteristics of free amino acids and TAV of fermented Niuganba

Name of amino acid	Flavor characteristics	Threshold value/(mg/100g)	RM	HM
Asp	Flavor(+)	100	0.87	0.90
Glu	Flavor (+)	30	5.09	5.25
Thr	Sweetness(+)	260	0.16	0.16
Ser	Sweetness(+)	150	0.27	0.28
Gly	Sweetness(+)	130	0.31	0.35
Ala	Sweetness(+)	60	0.86	0.93
Pro	Sweetness(+)	300	0.13	0.14
Val	Bitterness(-)	40	0.89	0.91
Met	Bitterness(-)	30	0.81	0.82
Ile	Bitterness(-)	90	0.34	0.34
Leu	Bitterness(-)	190	0.36	0.38
Phe	Bitterness(-)	90	0.47	0.49
Lys	Bitterness(-)	50	1.54	1.58
Tyr	Bitterness(-)	260	0.11	0.12
His	Bitterness(-)	20	1.86	1.89
Arg	Bitterness(-)	50	1.07	1.11

Notes: Significant differences ($P < 0.05$) between different columns within the same row are denoted by distinct

The impact of HVEF-assisted curing on taste characteristics and taste activity values (TAV) of free amino acids in fermented Niuganba is detailed in Table 5. Free amino acids contribute differentially to taste profiles, categorized as umami (Asp, Glu), sweet (Thr, Ser, Gly, Ala, Pro), and bitter (Val, Met, Ile, Leu, Phe, Lys, Tyr, His, Arg). The HM group contained 247.97 mg/100 g flavor amino acids, 226.11 mg/100 g sweet amino acids, and 410.13 mg/100 g bitter amino acids, compared to 239.54 mg/100 g (flavor), 210.68 mg/100 g

(sweet), and 399.38 mg/100 g (bitter) in the RM group, indicating enhanced taste complexity in HVEF-treated samples. Notably, taste contribution is quantified through TAV, calculated as the ratio of amino acid concentration to its taste threshold, with TAV > 1 indicating perceptible flavor impact. As shown in Table 5, In the two groups of Niuganba, Glu is the largest free amino acid of TAV, indicating that it contributed the most to the flavor taste of Niuganba. Among them, the TAV of Niuganba in the RM group was 5.09, and the meat sample in the HM group increased by 3.05% compared with it, indicating that the Niuganba in the HM group has a more delicious taste. This is consistent with the result of sensory evaluation. The TAV of the two bitter amino acids, Lys and Arg, in the Niuganba of both groups is greater than 1, indicating that these two amino acids contributed significantly to the bitterness of the meat samples. Although sweet amino acids exhibited TAV < 1, their synergistic accumulation may collectively enhance sweet perception (Duan J Y, *et al*., 2020). These findings collectively demonstrate that HVEF-assisted curing not only elevates proteolytic efficiency to improve nutritional value but also optimizes umami intensity through selective amino acid enrichment.

3.11 E-nose

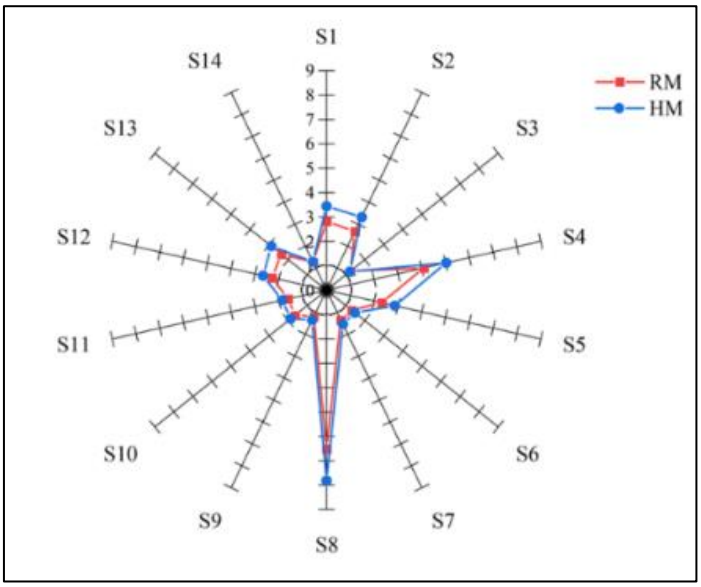

Figure 7: Electronic nose radar diagram

Electronic nose (E-nose), a highly sensitive analytical tool for detecting subtle variations in volatile compounds, has been widely employed in flavor characterization of meat products. As shown in Figure 7, E-nose profiling revealed distinct volatile patterns between HVEF-treated (HM) and control (RM) Niuganba. Both groups elicited strong responses on S4 (sensitive to nitrogen oxides) and S8 (responsive to sulfides) sensors, suggesting significant contributions of these compound classes to Niuganba's volatile profile. Previous studies have demonstrated that nitrogen oxides and sulfides serve as characteristic markers for meaty and roasted aromas in processed meats, with higher concentrations correlating with intensified flavor perception (Zhang W K, *et al*., 2022). Notably, the HM group exhibited significantly higher response values on S1, S2, S4, S5, S8, S12, and S13 sensors compared to RM group. These differential responses indicate HVEF-assisted curing facilitated increased accumulation of nitrogen oxides, organic/inorganic sulfides, alcohols, aldehydes, ketones, and long-chain alkanes during fermentation. Collectively, these findings suggest HVEF pretreatment effectively enhances both meaty aroma complexity and overall flavor perception in fermented Niuganba.

4. CONCLUSION

This study investigated the effect of HVEF-assisted curing on the quality of fermented Niuganba. The results showed that compared with the conventional refrigerator curing group (RM), the pH value, water activity, and yield of beef cured with HVEF assistance decreased rapidly during the fermentation process; the values of TBARS and the content of TVB-N remained at lower levels. For the fermented Niuganba produced by HVEF-assisted curing, the redness value increased significantly ($P < 0.05$); t the content of free amino acids increased, and the nutritional value was improved; the electronic nose differentiated the flavor differences of Niuganba in different curing groups, and the response values of the fermented Niuganba in the HVEF treatment group on the electronic nose sensors for inorganic and organic sulfides, alcohols, aldehydes, and ketones increased significantly. In conclusion, HVEF assisted curing can effectively improve the texture, color, flavor and other quality characteristics of fermented Niuganba.

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