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Effects of Fermentation on the Physicochemical Properties and Aroma of Lamb Liver Paste

Ting Liu [1], Taiwu Zhang [1], Lirong Yang [1], Yanni Zhang [1], Letian Kang [1], Le Yang [1], Yujia Zhai [2], Ye Jin [1], Lihua Zhao [1] and Yan Duan [1,*]

[1] College of Food Science and Engineering, Inner Mongolia Agricultural University, 306 Zhaowuda Road, Hohhot 010018, China
[2] Institute of Product Quality Inspection, Inner Mongolia Autonomous Region, Shihua Road, Hohhot 010070, China
* Correspondence: duanyan@imau.edu.cn

Abstract: The probiotic fermentation of lamb liver paste is a new method with which to utilize sheep by-products and address the issue of waste. In this study, a pH meter, chromaticity meter, texture analyzer, and gas chromatograph–mass spectrometer (GC–MS) were used to determine various indicators. The objective was to investigate the effect of fermentation on the physical properties and aroma of lamb liver paste. The results showed that the L* (brightness), a* (redness), and b* (yellowness) of the samples were significantly higher in the starter fermentation group than in the other two groups after storage for 0, 1, 7, 14, 21, and 28 days ($p < 0.05$). In addition, cohesiveness, adhesion, and chewiness were lower in the starter fermentation group after 7 days ($p < 0.05$). TVB-N and fat were lower in the starter fermentation group compared to the sterilization group at 28 days. pH was significantly lower in the starter fermentation group at the beginning of storage, and lactic acid bacteria numbers were significantly higher than in the sterilization groups ($p < 0.05$). Important aroma compounds, such as 2-undecenal, 1-octen-3-ol, and anethole, were significantly higher in the starter fermentation group than in the sterilization group ($p < 0.05$). Fermented lamb liver paste is a new by-product that exhibits a high degree of freshness and a low degree of fat oxidation during storage. This study provides a theoretical basis for future industrial production.

Keywords: lamb liver; by-product; GC–MS; texture; volatile compounds

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1. Introduction

Lamb meat is tender, tasty, unique in flavor, rich in nutrients, and loved by consumers. It also contributes to sustainable development in animal husbandry and the meat industry. The high demand for lamb has led to an increase in the production of sheep by-products. However, these by-products have not been efficiently utilized, resulting in great economic losses in animal husbandry and negative impacts on the ecological environment. A low comprehensive utilization rate of sheep by-products, a lack of product variety, and resource waste are problems associated with the process of animal slaughter. To address these issues, animal liver is worthy of attention in terms of converting animal by-products into valuable animal products. The vitamin (vitamin B2, vitamin C, and vitamin A), mineral (trace element selenium) [1,2], and other nutrient contents in animal liver are higher than those in milk, eggs, meat, fish, and other foods. The nutrients in lamb liver are components of many enzymes and coenzymes in human biochemical metabolism, which accelerate the body's metabolism, relieve visual fatigue, and enhance the body's immunity [3–5]. Consequently, fermented products can both alleviate these problems and provide benefits to the body.

Fermentation technology refers to the artificial application of methods (conditions such as temperature and pH) to control microorganisms (bacteria, yeast, etc.) to use organic matter as a medium for fermentation. The addition of probiotics induces host digestive protease and peptidase activity and also improves the absorption of small peptides and amino

acids in the organism by improving epithelial absorption and enhancing translocation [6]. Moreover, endogenous meat enzyme activity, microbial growth, and lipid oxidation reactions are partly responsible for the production of many aroma and carrion compounds [7]. Studies have found that, when lactic acid bacteria are added to fermented sausages, their metabolites (lactic acid, formic acid, acetic acid, etc.) give the product a special sour flavor [8,9]. Casaburi used two strains with differing abilities to decompose protein and fat as starter cultures to make fermented sausage. The experiment proved that both strains improved the nutritional quality of fermented sausage [10]. Antara found that after mixing *Lactobacillus plantarum* and *Pediococcus lactis* into sausages, the control of enterotoxin production by metabolizing lactic acid extended the shelf life of fermented sausages [11]. The Xing TK study showed that the addition of lactic acid bacteria and Staphylococcus significantly promoted the accumulation of free amino acids during the fermentation of beef jerky sausage [12]. In summary, the fermentation technology used for meat products is sophisticated, but there are few reports on the fermentation of by-products.

Microorganisms decompose sugars to produce acids, reducing the pH value of meat products. The rapid production of lactic acid, resulting in a decrease in pH, inhibits the growth of pathogenic and spoilage microorganisms and prolongs the shelf life of fermented foods. The pH value is often used to measure the maturity of fermented meat products and indicates the fermentation stage according to the evaluation index. This is used to establish the best fermentation time for the fermented product. In addition, other metabolites, such as lactic acid, acetic acid, propionic acid, benzoic acid, hydrogen peroxide, and bacteriocins, improve food safety [13]. Therefore, with the continuous improvement of comprehensive animal liver processing technology, animal liver will eventually be utilized to its full potential. Therefore, this experiment investigated the effects of fermentation on the probiotic count, texture, and aroma of lamb liver paste. The fermented lamb liver paste in this study had a pleasant aroma and represents a high-value sheep by-product. Moreover, it increases the variety of sheep by-products available, which will act to reduce waste in this industry.

2. Materials and Methods

2.1. Preparation of Fermented Lamb Liver Paste (Basic Recipe and Process Flow)

The preparation of goat liver paste refers to the "Liver Paste" product in the GOST 12319-77 standard report [14]. (1) Trimming: Raw lamb livers were trimmed for membranes, bile ducts, and other inclusions, and rinsed of blood. (2) Maceration: Trimmed lamb livers were macerated in 1% salt water for 3 h. (3) Pickling: According to the recipe (salt 2.5%, sugar 2%, Chinese rice cooking wine 3%, sodium nitrite 50 mg/kg, and vitamin C 0.05%), the livers were pickled at 0–4 °C for 12 h. (4) Cooking: The pickled lamb livers were cooked for 8–10 min (star anise 0.05%, pepper 0.05%, chili 0.1%, cinnamon 0.1%, dahurica 0.1%, garlic 0.5%, and fresh ginger 0.5%), and then placed on a rack to cool at room temperature. (5) Mixing: The cooled lamb livers were cut into pieces, and the small pieces were put into a ZB-40 chopper (Ruiheng Food Machinery Factory) for grinding. Spices (five-spice powder 0.3%, black pepper powder 0.2%, ginger powder 0.2%, salt 1.5%, sugar 1.5%, and corn starch 4%), emulsifier 3% (glycerol monostearate: sodium caseinate = 1:1), thickening agent 5% (sodium carboxymethyl cellulose: β-cyclodextrin = 4:1), ice water 40%, corn germ oil 9%, and starter culture 0.02% were continually added during mixing. (6) Fermentation and storage: The mixed filling was fermented in a humidity chamber at a constant temperature (39 °C, 7 h) to obtain the finished product, which was vacuum packed and stored at 4 °C in a refrigerator.

2.2. Experimental Design Grouping of Mutton Liver Paste during Storage

Starter fermentation group (SF): 0.02% starter F-1 Bactoferm® (*Staphylococcus xylosus* DD-34 and *Pediococcus pentosaceus* PCFF-1, Chr. Hansen Holding A/S, Hørsholm, Denmark) was added to basic formula lamb liver paste before fermenting in a humidity box at a constant temperature (39 °C, 7 h). Natural fermentation group (NF): basic formula lamb

liver paste with no starter culture added was fermented in a humidity box at a constant temperature (39 °C, 7 h). Sterilization treatment group (ST): the basic formula was sterilized at a high temperature (121 °C, 10 min) without adding the starter culture.

The changes in microbes and physicochemical indexes of the three lamb liver paste groups were compared on day 0, the 1st day, 7th day, 14th day, 21st day, and 28th day of storage. In order to explore the quality changes in the lamb liver paste during storage, the most suitable storage period was selected to provide a theoretical basis.

2.3. Indicator Measurement

2.3.1. Microbial Analysis

Under sterile conditions on an ultraclean bench (Haier Co., Ltd., Qindao, China), 25 g of the sample was weighed from the middle of the saucer, added to 225 mL of sterile physiological saline, vigorously shaken for 30 min, and then diluted in gradient after mixing. The appropriate dilution was selected for pouring into agar medium plates, and after culturing at 37 °C for 48 h, a count was performed. PCA medium (plate counting agar medium) was used to measure the total number of colonies. PCA medium ingredients: tryptone 5.0 g, yeast extract 2.5 g, glucose 1.0 g, agar 15.0 g, distilled water 1000 mL, and pH 7.0 ± 0.2. MRS medium (de Man, Rogosa, and Sharpe medium) was used to measure the number of lactic acid bacteria. MRS medium: ingredients: peptone 10.0 g, beef paste 10.0 g, yeast paste 5.0 g, diammonium hydrogen citrate [$(NH_4)_2HC_6H_5O_7$] 2.0 g, glucose ($C_6H_{12}O_6$-H_2O) 20.0 g, Tween 80 1.0 mL, sodium acetate (CH_3COONa-$3H_2O$) 5.0 g, dipotassium hydrogen phosphate (K_2HPO_4-$3H_2O$) 2.0 g, magnesium sulfate ($MgSO_4$-$7H_2O$) 0.58 g, manganese sulfate ($MnSO_4$-H_2O) 0.25 g, agar 18.0 g, distilled water 1000 mL, and pH 6.4 ± 0.2.

2.3.2. Determination of Physical and Chemical Indicators

pH value: The pH value was measured following the method described by Perea-Sanz et al. [15]. Using a portable pH meter (PB-10, Zhicheng Analytical Instrument Manufacturing Co., Ltd., Shanghai, China), which was calibrated in buffers with pH 4.60 and 7.00, each sample was measured three times. Triplicates were taken. Each 25 g sample was mixed with 225 mL of normal saline and shaken for 30 min, and the pH value was measured with a pH meter.

Color: Meat color was measured using a CR-410 chromometer (Konica Minolta, Tokyo, Japan). Standard white plate parameters were used for calibration (D65 light source; Y = 92.6, x = 0.3162, y = 0.3324). Lightness (L*), redness (a*), and yellowness (b*) values of each sample were measured, and the mean values were regarded as the product color [16].

Water activity (Aw): The Aw value was determined with an HD-3A intelligent water activity meter (Huake Instrument Co., Ltd., Wuxi, China).

Textural properties: The lamb liver paste was placed in a 4 cm cylindrical model and arranged into a cylindrical shape. We referred to the texture profile analysis method of Omana et al. [17] and used the TA.XT Express Stable Micro Systems Texture Analyser (Stable Micro systems Ltd., Godalming, UK). The setting parameters of the texture analyzer were as follows: "TPA" setting mode, the strain area was 50%, and the aluminum cylinder probe (diameter 36 mm) was used for double compression cycle test. The compression cycle of each sample lasts for 5 s, the test speed was 1.5 mm/s, and the lamb liver paste was measured in three cycles. The hardness, elasticity, cohesion, adhesiveness, and chewiness of each group of samples were assessed in parallel in the three groups. The parameters were quantified when the product regained its original position [16].

A simple summary of the procedure for TVB-N should be included [18]. Three parallel groups were formed for each group of samples. First, 10 g of goat liver paste was transferred into a distillation tube. Then, 75 mL distilled water was added and homogenized for 30 min. Next, 1 g of magnesium oxide was added to the distillation tube containing the treated sample. Finally, TVB-N content was determined by automatic Kjeldahl nitrogen analyzer (Kjeltec 8200, FOSS, Hilleroed, Denmark).

Thiobarbituric acid-reactive substances (TBARS): According to the method described by Gong et al. [19], lipid oxidation was determined by TBARS. Then, 10 g liver samples were collected and ground, 50 mL of 7.5% trichloroacetic acid was added (containing 0.1% EDTA), the mixture was shaken for 30 min, and the resulting product was filtered twice with double-layer filter paper. Thereafter, 5 mL of supernatant was aspirated with a pipette and added to 5 mL of 0.02% MTBA solution. This was then heated in a water bath at 90 °C for 40 min. The heated sample was cooled, centrifuged for 5 min (16,000 r/min), and then, after 1 h, 5 mL of chloroform was added to the supernatant, and it was well shaken. After standing for stratification, the supernatant was assessed by colorimetry at 532 and 600 nm. TBARS content was expressed as mg malonaldehyde (MDA) per kg liver paste.

$$\text{TBARS (mg/kg)} = (A_{532} - A_{600})/155 \times (1/10) \times 72.6 \times 1000$$

Nutritional indicators: The moisture, protein, and fat content of the samples were determined according to the method described by Perea-Sanz et al. [15]. Three groups of parallel tests were carried out for each group of samples.

2.4. Analysis of Volatile Compounds

The vacuum-preserved lamb liver paste was removed from the 4 °C refrigerator, 5 g of the sample was placed into a 20 mL sample bottle, and 5 mL of saturated sodium chloride solution and 1 μL of 2-methyl-3-heptanone solution (0.168 μg/mL) were added. The mixed sample was placed in the rotor, shaken, and placed on a magnetic stirrer. The extraction head was inserted into the sample bottle at a distance of 1 cm from the sample and removed after adsorption at 60 °C for 45 min. Then, the GC injection port was inserted, and the sample underwent desorption at 250 °C for 4 min.

Referring to the method by Luo et al. [20], analyses were performed on a Trace 1300 Series GC gas chromatograph fitted with an ISQ mass spectrometer and a Xcalibur ChemStationa (Thermo Fisher Scientific, Waltham, MA, USA). A DB-5 chromatographic column (TR-5MS 30 m × 0.25 mm, film thickness 0.25 μm) with Helium as the carrier gas was used for the gas chromatograph–mass spectrometer (GC–MS) determination of volatile aroma components. The carrier gas flow rate was 1.0 mL/min, and the temperature of the inlet and the interface was 250 °C. Heating program: The initial temperature was 40 °C. This was maintained for 3 min, increased to 150 °C at 4 °C /min, and then held for 1 min before increasing to 200 °C at 5 °C/min, again increasing to 230 °C at 20 °C/min, and holding for 5 min, with no split injection. The ion source temperature was 250 °C, the transmission line temperature was 250 °C, the mass scanning range was m/z 30–400, and the solvent delay was 1 min. The mass spectrum was qualitatively analyzed with MEANLIB (MathWorks, Inc., R2006a), library database (NIST MS Search Program 2.0), and Wiley Library, and the matching degree was greater than 98% as the identification basis. Additionally, 2-Methyl-3-heptanone was selected to be the internal standard for the determination of volatile flavor compounds. Quantitative analyses were carried out according to the peak area of 2-methyl-3-heptanone of known mass concentration.

2.5. Data Analysis

An analysis of variance (ANOVA) test and Pearson correlation analysis were performed using the Statistical Package for the Social Science (SPSS Inc., version 26.0). Values are expressed as mean ± standard deviation (RSD). p-values less than 0.05 were considered statistically significant. The origin version from 2021 developed by OriginLab was used for mapping. PCA diagrams were drawn using the R Programming Language "factoextra" (R version 4.1.1).

3. Results

3.1. The Total Number of Colonies and the Number of Lactic Acid Bacteria

Changes in the total number of colonies during storage of lamb liver paste are shown in Figure 1A and Table S1. On day 0 of storage, the total number of colonies in the SF group

was significantly higher than in the other two groups ($p < 0.05$), while the total number of colonies in the ST group in each storage period was significantly lower ($p < 0.05$). On the 28th day of storage, the total number of colonies in the three groups significantly increased ($p < 0.05$), and the total number of colonies in the ST group was significantly lower than in the other two groups ($p < 0.05$).

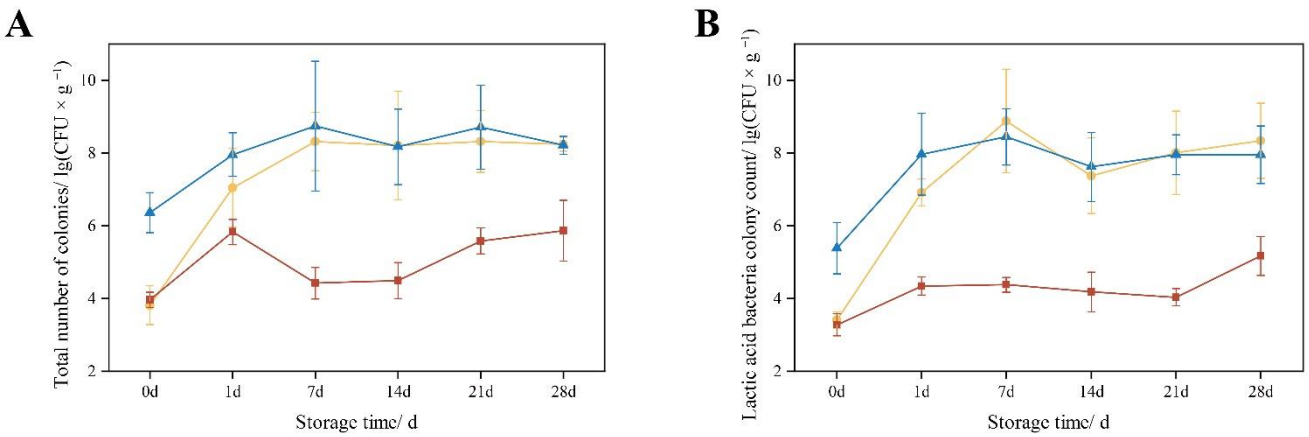

Figure 1. The changing trend of the total number of colonies and the number of lactic acid bacteria in lamb liver paste during the storage period. Note: yellow represents the natural fermentation group (NF); blue represents the starter fermentation group (SF); red represents the sterilization group (ST). (**A**) represents the change of the total colony number of the lamb liver paste with the storage time, and (**B**) represents the change of the lactic acid bacteria colony count of the lamb liver paste with the storage time.

The number of lactic acid bacteria during the storage of lamb liver paste is shown in Figure 1B and Table S2. During the whole storage period, the number of lactic acid bacteria in the ST group was significantly lower than in the other two groups ($p < 0.05$). The high-temperature treatment inhibited the activity of lactic acid bacteria, and the number of lactic acid bacteria in the SF group was significantly higher than in the NF group on the 1st day. The SF group and NF group had more lactic acid bacteria than the ST group.

3.2. pH Value

Acidic metabolites reduce the pH of the fermented product and increase the acidity of the product. As can be seen from Figure 2 and Table S3, after the 1st day of fermentation, the pH value of the SF group was 4.32, which was significantly lower than that of the other two groups ($p < 0.05$). The pH value of the SF group changed less during the storage period. In the NF group, the pH value decreased rapidly from 5.89 to 4.43 on the 7th day and then leveled off. There was no significant change in pH in the ST group ($p > 0.05$).

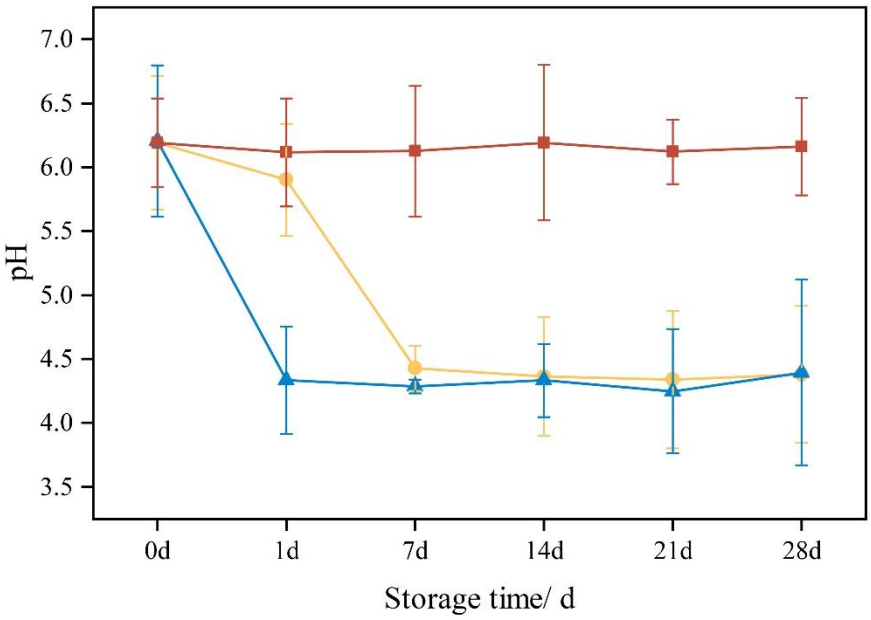

Figure 2. Change trend of pH value of lamb liver paste during storage period. Note: yellow represents the natural fermentation group (NF); blue represents the starter fermentation group (SF); red represents the sterilization group (ST).

3.3. Aw Value

Changes in the Aw value of lamb liver paste during storage are shown in Figure 3 and Table S4. The changing trend of the Aw value of the three groups of lamb liver paste is the same. During the whole storage process, there was no significant change in the Aw value of lamb liver paste in the three groups, and the Aw value was around 0.86.

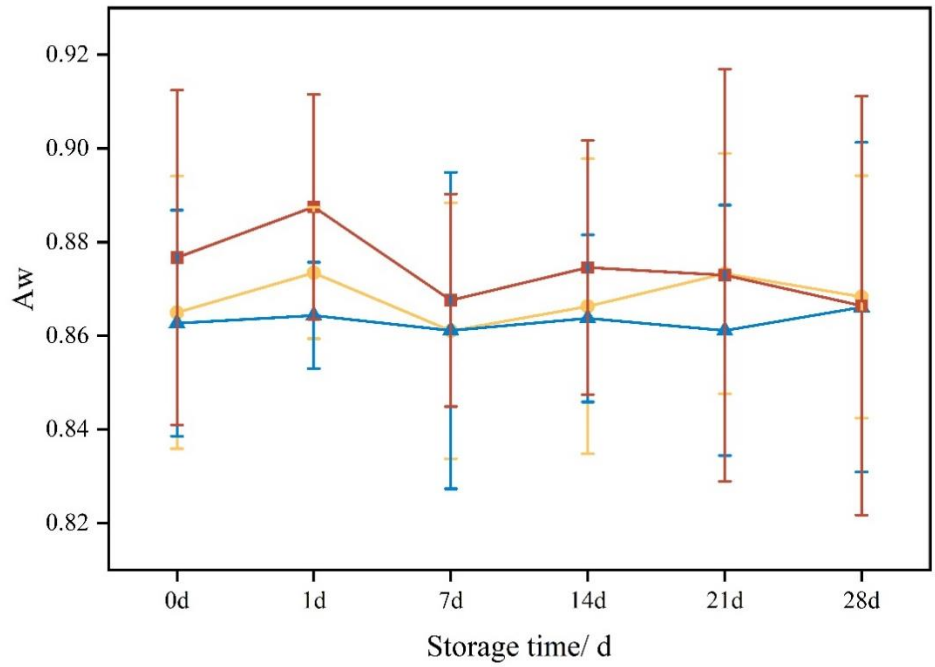

Figure 3. Change trend of Aw value of lamb liver paste during storage period. Note: yellow represents the natural fermentation group (NF); blue represents the starter fermentation group (SF); red represents the sterilization group (ST).

3.4. Chromatic Properties

The changes in L*, a*, and b* during the storage process of lamb liver paste are shown in Figure 4. Figure 4A and Table S5 show the increase in L* in the NF group and the SF group with the prolongation of storage time. The ST group had a significantly lower L* than other groups on the 28th day ($p < 0.05$). Figure 4B and Table S6 show that the a* values of lamb liver paste in the three groups did not change on day 0. The a* of the SF group was significantly higher than those of the other two groups, and the a* of the NF group was significantly higher than that of the ST group after 1 day of storage ($p < 0.05$). With the increase in storage time (Figure 4C and Table S7), the b* of lamb liver paste in the three groups also increased. The b* of the SF group was significantly higher than in the other two groups ($p < 0.05$).

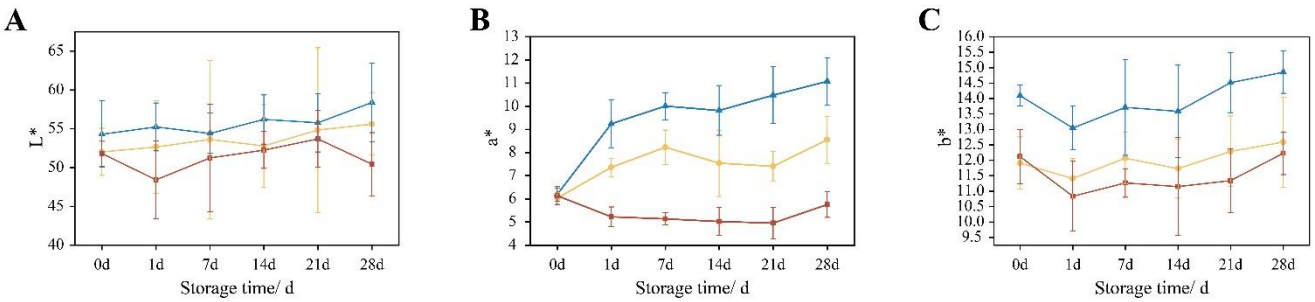

Figure 4. Change trend of color difference (a*, b*, L*) of lamb liver paste during storage period. Note: yellow represents the natural fermentation group (NF); blue represents the starter fermentation group (SF); red represents the sterilization group (ST). (**A**) represents the change of the Lightness value (L*) of the lamb liver paste with the storage time, (**B**) represents the change of the Redness value (a*) of the lamb liver paste with the storage time, and (**C**) represents the change of the Yellowness value (b*) of the lamb liver paste with the storage time.

3.5. Texture

The textural properties reflect the tissue structure and physical state of lamb liver paste. The textural properties of lamb liver paste throughout the storage period are shown in Table 1. The hardness, elasticity, cohesion, adhesiveness, and chewiness of the SF group were significantly higher than the other two groups on day 0 ($p < 0.05$). The adhesion of the SF group was significantly higher than the other two groups on the 1st day ($p < 0.05$), and there was no significant difference in the other indicators ($p > 0.05$). The elasticity, cohesiveness, adhesiveness, and chewiness of the sterilization group were significantly higher than the other two groups on the 7th day and 14th day of storage ($p < 0.05$). With the prolongation of the storage period, there was no significant difference in elasticity among the three groups on the 21st day and 28th day ($p > 0.05$). Cohesion, adhesion, and chewiness were significantly higher in the ST group ($p < 0.05$), but there was no significant difference between the NF group and the SF group.

Table 1. Differences in the texture of lamb liver paste during storage.

	Groups	0 d	1 d	7 d	14 d	21 d	28 d
Hardness/g	NF-group	1770.55 ± 67.60 ^{Bb}	1963.21 ± 26.12 ^{Aa}	1621.91 ± 119.71 ^{Bb}	1592.06 ± 45.13 ^{Bc}	1367.97 ± 47.51 ^{Cc}	1353.04 ± 52.84 ^{Cb}
	SF-group	1963.45 ± 37.48 ^{ABa}	2054.15 ± 55.98 ^{Aa}	2017.42 ± 47.64 ^{Aa}	1870.76 ± 25.70 ^{Bb}	1472.28 ± 12.69 ^{Cb}	1409.58 ± 26.61 ^{Cb}
	ST-group	1844.84 ± 41.44 ^{Cab}	1964.04 ± 13.45 ^{BCa}	2097.47 ± 55.96 ^{Ba}	2056.36 ± 92.52 ^{Ba}	2262.87 ± 27.78 ^{Aa}	2348.97 ± 73.99 ^{Aa}
Elasticity	NF-group	0.18 ± 0.01 ^{Ab}	0.18 ± 0.01 ^{Aa}	0.15 ± 0.01 ^{Ab}	0.16 ± 0.03 ^{Aab}	0.17 ± 0.05 ^{Aa}	0.13 ± 0.02 ^{Aa}
	SF-group	0.30 ± 0.05 ^{Aa}	0.19 ± 0.02 ^{Ba}	0.14 ± 0.01 ^{Bb}	0.12 ± 0.02 ^{Bb}	0.14 ± 0.04 ^{Ba}	0.18 ± 0.04 ^{Ba}
	ST-group	0.18 ± 0.01 ^{Ab}	0.18 ± 0.02 ^{Aa}	0.18 ± 0.01 ^{Aa}	0.17 ± 0.01 ^{Aa}	0.18 ± 0.01 ^{Aa}	0.19 ± 0.02 ^{Aa}
Cohesiveness	NF-group	0.14 ± 0.02 ^{Ab}	0.12 ± 0.01 ^{ABa}	0.09 ± 0.01 ^{BCb}	0.08 ± 0.01 ^{Cb}	0.08 ± 0.00 ^{Cb}	0.08 ± 0.01 ^{Cb}
	SF-group	0.19 ± 0.02 ^{Aa}	0.12 ± 0.01 ^{Ba}	0.10 ± 0.01 ^{Cab}	0.09 ± 0.00 ^{Cb}	0.09 ± 0.01 ^{Cb}	0.08 ± 0.00 ^{Cb}
	ST-group	0.15 ± 0.01 ^{Ab}	0.12 ± 0.00 ^{Ba}	0.12 ± 0.00 ^{Ba}	0.13 ± 0.01 ^{ABa}	0.12 ± 0.00 ^{Ba}	0.13 ± 0.01 ^{ABa}
Adhesion	NF-group	261.05 ± 8.48 ^{Ab}	227.93 ± 3.04 ^{Bc}	152.57 ± 10.05 ^{CDc}	160.30 ± 11.51 ^{Cb}	127.43 ± 14.35 ^{DEb}	107.15 ± 8.17 ^{Eb}
	SF-group	374.55 ± 8.42 ^{Aa}	268.12 ± 5.58 ^{Ba}	199.36 ± 4.94 ^{Cb}	167.51 ± 2.11 ^{Db}	121.36 ± 1.52 ^{Eb}	116.24 ± 1.62 ^{Eb}
	ST-group	256.90 ± 1.39 ^{CDb}	242.28 ± 5.13 ^{Db}	251.53 ± 2.21 ^{Da}	283.36 ± 8.57 ^{ABa}	274.28 ± 1.60 ^{BCa}	296.84 ± 14.48 ^{Aa}
Chewiness	NF-group	49.89 ± 1.76 ^{Aab}	41.09 ± 1.41 ^{Aa}	23.47 ± 3.33 ^{Bb}	23.35 ± 5.25 ^{Bb}	22.53 ± 8.58 ^{Bb}	14.85 ± 1.19 ^{Bb}
	SF-group	58.71 ± 7.13 ^{Aa}	49.99 ± 7.10 ^{Aa}	28.51 ± 0.28 ^{Bab}	21.34 ± 2.02 ^{BCb}	15.79 ± 3.52 ^{Cb}	15.68 ± 0.81 ^{Cb}
	ST-group	44.91 ± 0.92 ^{Ab}	41.87 ± 1.98 ^{Aa}	41.24 ± 8.24 ^{Aa}	51.39 ± 8.47 ^{Aa}	42.42 ± 2.02 ^{Aa}	41.85 ± 7.55 ^{Aa}

Uppercase letters denote significant differences in the same row; lowercase letters denote significant differences in the same column and the same metric.

3.6. TVB-N Value

The TVB-N value during the storage of lamb liver paste is shown in Figure 5 and Table S8. TVB-N values represent the freshness of the food, and a higher TVB-N content indicates higher amino acid destruction. The TVB-N value of the NF group was significantly higher than those of the other two groups on the 1st day of storage ($p < 0.05$). The TVB-N value of the SF group and the ST group were significantly higher than that of the NF group after 7 days ($p < 0.05$). After the 28th day, the TVB-N value of the SF group was lower than those of the other two groups.

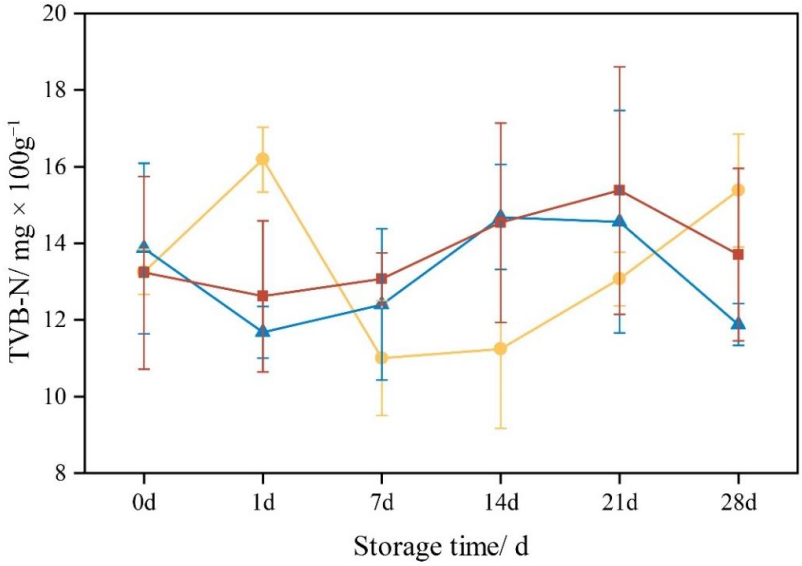

Figure 5. Variation trend of TVB-N value during storage of lamb liver paste. Note: yellow represents the natural fermentation group (NF); blue represents the starter fermentation group (SF); red represents the sterilization group (ST).

3.7. TBARS Value

The TBARS value indicates the degree of fat oxidation according to the amount of malondialdehyde—a secondary product—that is formed by fat oxidation. Fat oxidation

causes spoilage and deterioration of meat quality and reduces the storability of meat. The higher the TBARS value, the higher the degree of fat oxidation and the greater the spoilage of the meat. The changes in TBARS values during the storage of lamb liver paste are shown in Figure 6 and Table S9. The TBARS values of the three groups decreased during processing and storage with increasing time. The TBARS values were significantly higher in the NF group than in the other two groups on the 1st day and significantly lower on the 14th day ($p < 0.05$).

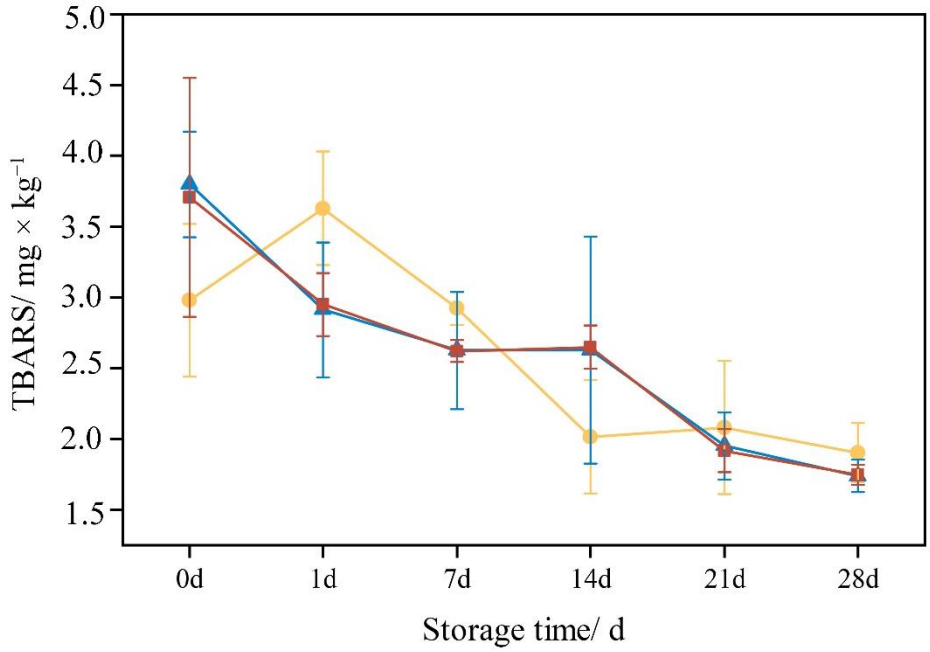

Figure 6. Variation trend of TBARS value during storage of lamb liver paste. Note: yellow represents the natural fermentation group (NF); blue represents the starter fermentation group (SF); red represents the sterilization group (ST).

3.8. Basic Nutritional Indicators

3.8.1. Moisture Content

The moisture content of lamb liver paste during storage is shown in Figure 7A and Table S10. The moisture content of the three groups was similar for all storage days ($p > 0.05$).

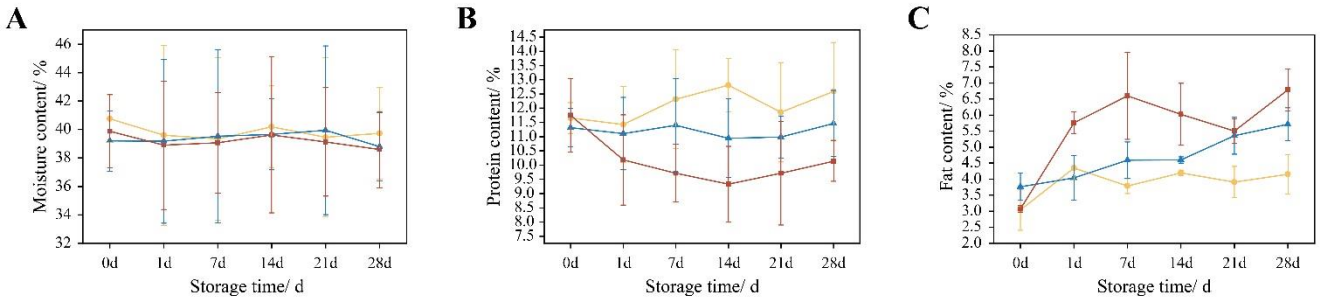

Figure 7. Changes in the basic nutritional quality of lamb liver paste during storage. Note: yellow represents the natural fermentation group (NF); blue represents the starter fermentation group (SF); red represents the sterilization group (ST). (**A**) represents the change of the moisture content of the lamb liver paste with the storage time, (**B**) represents the change of the protein content of the lamb liver paste with the storage time, and (**C**) represents the change of the fat content of the lamb liver paste with the storage time.

3.8.2. Protein Content

The protein content of lamb liver paste during storage is shown in Figure 7B and Table S11. The protein content of the ST group was significantly lower than that of the NF group and the fermentation group throughout the storage period ($p < 0.05$). The protein content of the NF group was significantly higher than that of the SF group and ST group ($p < 0.05$).

3.8.3. Fat Content

The fat content of lamb liver paste during storage is shown in Figure 7C and Table S12. In the ST group, the fat content increased significantly from day 0 to the 1st day, and the fat content was higher than in the other groups ($p < 0.05$). Except for the 1st day, the fat content in the SF group was significantly higher than in the NF group for all the storage times ($p > 0.05$).

3.9. Volatile Profile

The differences between groups were analyzed using the dimensionality reduction method and were ranked by principal component analysis (PCA). It can be seen from the PCA graph that the aroma of the three groups of samples were well-differentiated throughout the storage period (Figure 8). The scattered areas of the sample points are relatively dense, and the differences within the groups are small. The values of PC1 and PC2 accounted for 97.8%, representing the whole sample, indicating that the lamb liver pastes with different treatments were different.

The aroma analysis of lamb liver paste is shown in Table 2. Benzaldehyde in the NF group and SF group was significantly higher than in the ST group ($p < 0.05$). Octanal, decanal, 2,4-decadienal, and tetradecanal were significantly higher in the SF group and NF group as compared to the ST group ($p < 0.05$). The proportions of 2-undecenal and 2-octenal in the SF group and NF group were significantly higher than in the ST group ($p < 0.01$). The content of 1-hexanol in the NF group was significantly higher than in the other two groups ($p < 0.05$). Additionally, 1-Heptanol in the SF group was significantly higher than in the other two groups ($p < 0.05$). The 1-octen-3-ol in the SF group and the NF group was significantly higher than in the ST group ($p < 0.05$). Benzaldehyde, 4-(1-methyl ethyl)- and anethole in the SF group were significantly higher than in the other two groups ($p < 0.05$). Furthermore, 2-Nonanone was significantly higher in the NF group ($p < 0.05$).

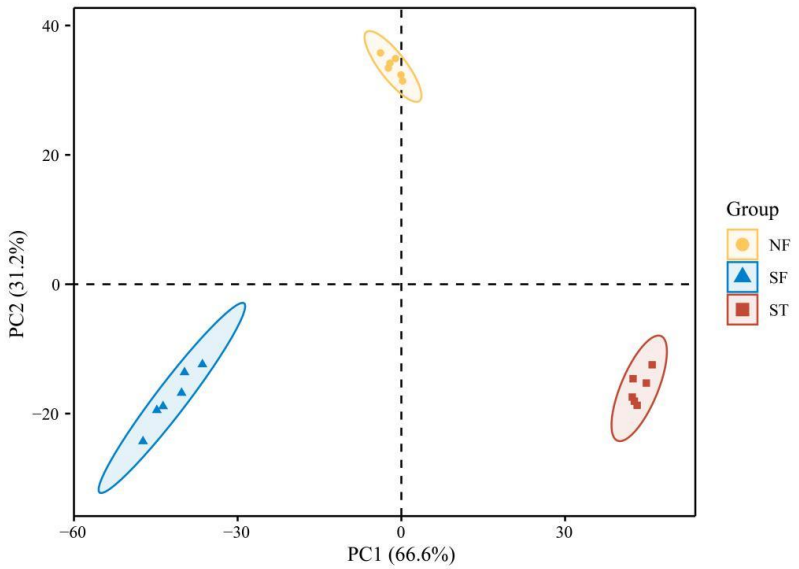

Figure 8. Principal coordinate analysis (PCoA) scatter plot analysis of two groups of samples. Note: yellow represents the natural fermentation group (NF); blue represents the starter fermentation group (SF); red represents the sterilization group (ST).

Table 2. Effects of three processing methods on volatile flavor compounds in lamb liver paste (μg/kg).

Volatile Flavor Compounds	Groups	0 d	1 d	7 d	14 d	21 d	28 d
Pentanal	NF	7.42 ± 0.85^{Db}	24.62 ± 0.82^{Cb}	32.24 ± 3.18^{Ba}	21.44 ± 2.67^{Cb}	31.19 ± 1.55^{Ba}	40.94 ± 3.08^{Aa}
	SF	7.29 ± 0.41^{Cb}	27.01 ± 1.80^{Ba}	34.88 ± 3.03^{Aa}	26.00 ± 3.28^{Ba}	2.36 ± 0.24^{Db}	3.25 ± 0.38^{Db}
	ST	20.08 ± 2.68^{Aa}	4.44 ± 0.57^{Cc}	15.93 ± 1.05^{Bb}	16.51 ± 1.50^{Bc}	1.77 ± 0.10^{Db}	2.16 ± 0.32^{Db}
Hexanal	NF	146.18 ± 15.29^{Cb}	492.83 ± 20.58^{Ab}	84.92 ± 7.92^{Da}	380.33 ± 38.01^{Bb}	18.46 ± 1.22^{Ea}	16.23 ± 1.97^{Eb}
	SF	112.24 ± 8.49^{Dc}	540.75 ± 21.68^{Ba}	662.09 ± 63.30^{Aa}	451.44 ± 39.54^{Ca}	15.76 ± 1.45^{Eb}	18.13 ± 1.78^{Eab}
	ST	413.27 ± 18.15^{Aa}	64.47 ± 2.09^{Cc}	272.52 ± 28.61^{Bb}	274.21 ± 15.69^{Ba}	17.15 ± 2.36^{Dab}	19.36 ± 2.01^{Da}
Heptanal	NF	25.54 ± 1.63^{Cb}	8.44 ± 1.09^{Db}	41.82 ± 5.19^{Aa}	31.50 ± 3.09^{Bb}	5.36 ± 0.58^{Db}	4.18 ± 0.36^{Db}
	SF	21.81 ± 1.48^{Bc}	40.55 ± 2.82^{Aa}	42.22 ± 3.03^{Aa}	39.10 ± 6.23^{Aa}	4.74 ± 0.65^{Cb}	4.78 ± 0.77^{Cb}
	ST	38.99 ± 2.97^{Aa}	9.67 ± 1.10^{Cb}	21.46 ± 1.45^{Bb}	22.01 ± 1.29^{Bc}	7.70 ± 0.52^{Ca}	7.57 ± 1.13^{Ca}
Octanal	NF	43.54 ± 3.30^{Cb}	81.70 ± 4.87^{Bb}	106.83 ± 16.95^{Aa}	91.79 ± 7.27^{Bb}	14.16 ± 2.07^{Da}	18.82 ± 2.26^{Da}
	SF	40.62 ± 5.38^{Bb}	137.87 ± 20.03^{Aa}	125.30 ± 15.41^{Aa}	130.46 ± 10.86^{Aa}	6.44 ± 0.53^{Cb}	8.42 ± 0.87^{Cb}
	ST	72.43 ± 12.41^{Aa}	7.11 ± 1.08^{Cc}	51.21 ± 3.38^{Bb}	49.14 ± 3.46^{Bc}	12.92 ± 1.13^{Ca}	9.94 ± 0.79^{Cb}
Nonanal	NF	126.47 ± 15.31^{Cb}	168.31 ± 17.91^{Aa}	158.63 ± 12.55^{Ba}	145.30 ± 12.91^{Cb}	34.67 ± 1.82^{Db}	30.51 ± 3.23^{Db}
	SF	127.41 ± 10.25^{Cb}	145.92 ± 15.58^{Cb}	165.23 ± 6.47^{Ba}	184.53 ± 18.41^{Aa}	27.10 ± 3.70^{Dc}	30.52 ± 3.48^{Db}
	ST	167.78 ± 13.44^{Aa}	55.53 ± 6.50^{Cc}	91.29 ± 10.52^{Bb}	94.32 ± 7.34^{Bc}	42.44 ± 4.37^{Ca}	43.65 ± 3.68^{Ca}
Benzaldehyde	NF	60.94 ± 4.13^{Cb}	112.34 ± 14.23^{Ba}	130.81 ± 15.94^{Aa}	99.75 ± 5.07^{Bb}	38.25 ± 3.93^{Db}	32.26 ± 2.68^{Db}
	SF	36.13 ± 3.74^{Bc}	118.83 ± 14.76^{Aa}	113.52 ± 5.31^{Ab}	115.86 ± 7.56^{Aa}	34.12 ± 2.09^{Bb}	34.69 ± 4.06^{Bb}
	ST	97.58 ± 9.94^{Aa}	59.12 ± 6.16^{Cb}	83.73 ± 5.10^{Bc}	85.84 ± 5.34^{Bc}	51.45 ± 4.55^{Ca}	48.79 ± 6.37^{Ca}
Tetradecanal	NF	1.77 ± 0.18^{Ec}	4.72 ± 0.40^{Db}	7.30 ± 0.62^{Ba}	5.72 ± 0.25^{Cb}	12.41 ± 0.87^{Aa}	6.25 ± 0.55^{Cb}
	SF	6.88 ± 0.48^{Ba}	7.42 ± 0.67^{Aa}	4.57 ± 0.57^{Cb}	7.61 ± 0.52^{Aa}	4.98 ± 0.52^{Cb}	6.30 ± 0.60^{Bb}
	ST	1.77 ± 0.18^{Db}	5.10 ± 0.66^{Cb}	4.56 ± 0.66^{Cb}	4.35 ± 0.39^{Cc}	12.14 ± 1.54^{Aa}	8.81 ± 1.23^{Ba}
2-Undecenal	NF	10.16 ± 1.00^{Ca}	55.30 ± 5.80^{Bb}	89.24 ± 11.83^{Aa}	65.28 ± 6.48^{Bb}	2.90 ± 0.27^{Ca}	2.36 ± 0.30^{Ca}
	SF	13.65 ± 1.03^{Ca}	92.88 ± 8.53^{Aa}	77.12 ± 4.86^{Bb}	85.07 ± 13.86^{Ba}	2.32 ± 0.13^{Cb}	2.39 ± 0.33^{Ca}
	ST	10.01 ± 1.42^{Ba}	3.88 ± 0.23^{Cc}	18.31 ± 1.68^{Ac}	19.21 ± 2.47^{Ac}	1.86 ± 0.19^{Cc}	1.91 ± 0.20^{Cb}
2-Octenal	NF	45.60 ± 3.01^{Db}	168.06 ± 15.66^{Bb}	212.57 ± 10.47^{Aa}	146.82 ± 10.09^{Cb}	12.18 ± 0.70^{Ea}	14.16 ± 1.19^{Ea}
	SF	37.79 ± 2.31^{Cb}	216.19 ± 22.17^{Aa}	222.56 ± 15.11^{Aa}	168.40 ± 14.09^{Ba}	9.86 ± 0.58^{Db}	11.00 ± 1.84^{Db}
	ST	102.38 ± 9.95^{Aa}	18.30 ± 1.21^{Cc}	93.61 ± 4.96^{Bb}	90.52 ± 8.31^{Bc}	8.77 ± 0.67^{Cc}	11.58 ± 0.34^{Cb}
2-Nonenal	NF	2.85 ± 0.21^{Db}	5.95 ± 0.54^{Bb}	7.05 ± 1.08^{Ab}	6.01 ± 0.49^{Bb}	7.13 ± 0.31^{Aa}	4.04 ± 0.17^{Ca}
	SF	2.60 ± 0.29^{Db}	10.97 ± 0.88^{Ba}	23.27 ± 3.46^{Aa}	6.68 ± 0.52^{Ca}	1.84 ± 0.28^{Db}	3.57 ± 0.26^{Db}
	ST	6.62 ± 0.68^{Ba}	1.49 ± 0.14^{Dc}	2.89 ± 0.24^{Cc}	2.97 ± 0.28^{Cc}	7.44 ± 0.76^{Aa}	1.71 ± 0.21^{Dc}
2-Decenal	NF	14.38 ± 0.84^{Db}	62.14 ± 3.81^{Ca}	117.49 ± 6.83^{Aa}	82.98 ± 8.66^{Bb}	7.39 ± 0.73^{Da}	6.49 ± 0.75^{Da}
	SF	15.83 ± 2.52^{Db}	63.46 ± 2.84^{Ba}	46.77 ± 4.38^{Cb}	99.53 ± 8.95^{Aa}	5.14 ± 0.34^{Eb}	5.70 ± 0.33^{Eb}
	ST	38.45 ± 4.53^{Aa}	8.19 ± 1.00^{Cb}	28.32 ± 1.55^{Bc}	29.78 ± 3.33^{Bc}	5.79 ± 0.49^{Cb}	4.99 ± 0.24^{Cb}
2,4-Decadienal	NF	3.86 ± 0.39^{Dc}	15.30 ± 1.79^{Cb}	26.54 ± 2.53^{Ba}	35.84 ± 4.29^{Aa}	28.55 ± 1.54^{Bb}	25.66 ± 4.09^{Ba}
	SF	4.73 ± 0.57^{Db}	30.18 ± 2.57^{Ca}	25.08 ± 2.05^{Ca}	31.94 ± 2.91^{Ba}	36.25 ± 5.58^{Aa}	29.43 ± 2.05^{Ca}
	ST	9.32 ± 0.39^{Aa}	4.52 ± 0.63^{Cc}	8.22 ± 0.68^{Bb}	7.51 ± 0.43^{Bb}	4.56 ± 0.77^{Cc}	4.04 ± 0.31^{Cb}
2,4-Dodecadienal	NF	18.98 ± 0.98^{Bb}	89.33 ± 7.75^{Aa}	53.79 ± 4.28^{Ba}	82.05 ± 6.75^{Ab}	5.47 ± 0.61^{Da}	3.36 ± 0.21^{Db}
	SF	19.27 ± 1.99^{Bb}	12.24 ± 1.76^{Cb}	11.75 ± 1.50^{Cc}	118.49 ± 8.87^{Aa}	3.27 ± 0.25^{Db}	3.32 ± 0.25^{Db}
	ST	62.12 ± 2.93^{Aa}	9.22 ± 0.84^{Cb}	38.86 ± 5.48^{Bb}	38.38 ± 4.03^{Bc}	5.62 ± 0.75^{Ca}	5.11 ± 0.35^{Ca}

Table 2. *Cont.*

Volatile Flavor Compounds	Groups	0 d	1 d	7 d	14 d	21 d	28 d
1-Hexanol	NF	9.59 ± 0.76 ^{Ba}	3.78 ± 0.38 ^{Db}	11.34 ± 1.07 ^{Aa}	7.47 ± 1.03 ^{Ca}	2.89 ± 0.20 ^{Ea}	2.14 ± 0.19 ^{Ea}
	SF	2.96 ± 0.10 ^{Cb}	4.40 ± 0.30 ^{Ba}	3.82 ± 0.36 ^{Bb}	5.17 ± 0.44 ^{Ab}	3.08 ± 0.23 ^{Ca}	0.98 ± 0.46 ^{Db}
	ST	1.38 ± 0.22 ^{Bc}	2.06 ± 0.34 ^{Ac}	2.12 ± 0.22 ^{Ac}	2.15 ± 0.28 ^{Ac}	2.34 ± 0.31 ^{Ab}	1.39 ± 0.20 ^{Bb}
1-Heptanol	NF	3.75 ± 0.29 ^{Cc}	19.05 ± 2.20 ^{Bb}	27.14 ± 2.97 ^{Aa}	16.08 ± 1.87 ^{Bb}	3.83 ± 0.43 ^{Ca}	1.76 ± 0.18 ^{Cc}
	SF	12.06 ± 1.25 ^{Ca}	25.44 ± 2.56 ^{Ba}	23.68 ± 3.67 ^{Ba}	46.03 ± 3.55 ^{Aa}	2.88 ± 0.19 ^{Db}	3.00 ± 0.19 ^{Db}
	ST	5.27 ± 1.04 ^{Bb}	17.58 ± 1.56 ^{Ab}	3.30 ± 0.55 ^{Cb}	3.26 ± 0.24 ^{Cc}	3.63 ± 0.37 ^{Ca}	3.99 ± 0.63 ^{Ca}
1-Octanol	NF	6.35 ± 0.37 ^{Db}	15.24 ± 1.63 ^{Ba}	18.52 ± 2.35 ^{Aa}	12.58 ± 1.73 ^{Cb}	7.13 ± 0.21 ^{Da}	4.78 ± 0.41 ^{Da}
	SF	6.06 ± 0.66 ^{Cb}	15.99 ± 1.33 ^{Ba}	19.37 ± 1.25 ^{Aa}	15.96 ± 1.71 ^{Ba}	3.28 ± 0.28 ^{Db}	2.44 ± 0.22 ^{Db}
	ST	9.88 ± 0.62 ^{Aa}	3.41 ± 0.24 ^{Cb}	7.92 ± 0.93 ^{Bb}	7.83 ± 0.91 ^{Bc}	1.85 ± 0.11 ^{Dc}	1.86 ± 0.19 ^{Dc}
1-Octen-3-ol	NF	78.13 ± 2.42 ^{Cab}	109.64 ± 6.39 ^{Aa}	89.92 ± 5.07 ^{Ba}	60.79 ± 4.45 ^{Db}	7.96 ± 0.77 ^{Eb}	6.77 ± 0.71 ^{Eb}
	SF	72.55 ± 4.8 ^{Cb}	91.86 ± 10.26 ^{Ab}	84.32 ± 8.81 ^{Ba}	76.48 ± 4.86 ^{Ca}	10.45 ± 0.67 ^{Da}	10.66 ± 1.05 ^{Da}
	ST	85.34 ± 7.21 ^{Aa}	19.50 ± 1.85 ^{Cc}	51.85 ± 3.94 ^{Bb}	47.36 ± 3.87 ^{Bc}	7.31 ± 0.91 ^{Db}	9.31 ± 1.31 ^{Da}
Benzaldehyde, 4-(1-methyl lethyl)-	NF	24.14 ± 2.46 ^{Ab}	15.39 ± 1.17 ^{Bb}	11.68 ± 0.73 ^{Cb}	8.17 ± 1.17 ^{Dc}	7.56 ± 0.35 ^{Db}	6.54 ± 0.58 ^{Dc}
	SF	31.69 ± 2.50 ^{Ca}	101.55 ± 10.90 ^{Aa}	83.26 ± 11.65 ^{Ba}	16.57 ± 2.12 ^{Da}	15.98 ± 2.35 ^{Da}	15.48 ± 1.19 ^{Da}
	ST	15.34 ± 0.66 ^{Bc}	19.08 ± 0.90 ^{Ab}	14.19 ± 0.80 ^{Bb}	13.75 ± 1.24 ^{Bb}	18.45 ± 2.00 ^{Aa}	15.89 ± 1.82 ^{Ba}
2-Heptanone	NF	2.82 ± 0.16 ^{Db}	6.97 ± 0.65 ^{Aa}	4.99 ± 0.44 ^{Ba}	3.19 ± 0.44 ^{Db}	2.56 ± 0.24 ^{Db}	3.50 ± 0.24 ^{Cb}
	SF	2.86 ± 0.20 ^{Cb}	6.10 ± 0.86 ^{Aa}	5.45 ± 0.71 ^{Aa}	4.43 ± 0.48 ^{Ba}	2.86 ± 0.19 ^{Cab}	2.62 ± 0.34 ^{Cc}
	ST	4.42 ± 0.53 ^{Ba}	3.86 ± 0.55 ^{Cb}	4.16 ± 0.34 ^{Bb}	3.79 ± 0.38 ^{Cab}	3.33 ± 0.45 ^{Ca}	5.51 ± 0.32 ^{Aa}
2-Nonanone	NF	4.21 ± 0.35 ^{Cb}	10.79 ± 0.40 ^{Ba}	4.37 ± 0.38 ^{Cb}	37.34 ± 3.70 ^{Aa}	3.01 ± 0.31 ^{Ca}	3.64 ± 0.38 ^{Ca}
	SF	4.73 ± 0.68 ^{Bb}	5.78 ± 0.33 ^{Ab}	5.44 ± 0.32 ^{Aa}	3.86 ± 0.34 ^{Cb}	1.63 ± 0.19 ^{Eb}	2.36 ± 0.27 ^{Db}
	ST	13.77 ± 1.44 ^{Aa}	3.70 ± 0.33 ^{Bc}	2.41 ± 0.21 ^{Ca}	2.50 ± 0.15 ^{Cb}	2.46 ± 0.25 ^{Cb}	2.47 ± 0.17 ^{Cb}
2-Tridecanone	NF	16.31 ± 2.13 ^{Aa}	10.73 ± 1.03 ^{Ca}	11.46 ± 0.89 ^{Ca}	12.15 ± 1.02 ^{Bb}	9.58 ± 0.58 ^{Ca}	9.40 ± 0.94 ^{Ca}
	SF	17.94 ± 3.11 ^{Aa}	8.72 ± 0.32 ^{Bb}	6.64 ± 0.57 ^{Bb}	17.60 ± 0.69 ^{Aa}	1.76 ± 0.16 ^{Cc}	2.39 ± 0.25 ^{Cb}
	ST	3.92 ± 0.28 ^{Ab}	2.60 ± 0.13 ^{Bc}	2.09 ± 0.21 ^{Dc}	2.06 ± 0.18 ^{Dc}	2.65 ± 0.13 ^{Bb}	2.44 ± 0.24 ^{Cb}
Ethyl octanoate	NF	6.89 ± 0.56 ^{Db}	10.40 ± 1.07 ^{Ab}	8.71 ± 0.88 ^{Cb}	7.66 ± 0.76 ^{Db}	10.02 ± 1.00 ^{Ba}	10.05 ± 1.19 ^{Bb}
	SF	11.31 ± 1.00 ^{Ba}	116.18 ± 9.85 ^{Aa}	106.08 ± 14.01 ^{Aa}	12.17 ± 0.85 ^{Ba}	10.29 ± 0.76 ^{Ba}	12.54 ± 1.65 ^{Ba}
	ST	12.36 ± 1.30 ^{Aa}	5.14 ± 0.48 ^{Cb}	3.09 ± 0.29 ^{Bb}	3.13 ± 0.17 ^{Bc}	3.97 ± 0.39 ^{Bb}	6.41 ± 0.62 ^{Bc}
Ethyl nonanoate	NF	5.73 ± 0.51 ^{Bc}	3.77 ± 0.31 ^{Cc}	11.17 ± 0.70 ^{Aa}	5.38 ± 0.36 ^{Bc}	2.25 ± 0.13 ^{Dc}	2.13 ± 0.21 ^{Da}
	SF	6.73 ± 0.52 ^{Db}	11.74 ± 1.08 ^{Ba}	7.13 ± 1.04 ^{Db}	14.10 ± 1.33 ^{Aa}	7.64 ± 0.60 ^{Db}	9.93 ± 0.81 ^{Ca}
	ST	9.42 ± 0.57 ^{Ba}	8.42 ± 0.68 ^{Db}	7.26 ± 0.61 ^{Db}	7.05 ± 0.56 ^{Db}	10.47 ± 1.00 ^{Aa}	8.50 ± 1.18 ^{Cb}
Ethyl tridecanoate	NF	17.15 ± 1.52 ^{Cb}	18.58 ± 2.26 ^{Cb}	23.86 ± 1.31 ^{Ba}	20.60 ± 1.95 ^{Ca}	26.21 ± 2.73 ^{Aa}	22.42 ± 2.48 ^{Ba}
	SF	30.55 ± 3.33 ^{Aa}	23.34 ± 1.86 ^{Ca}	18.94 ± 0.75 ^{Cb}	22.21 ± 2.14 ^{Ca}	22.84 ± 3.34 ^{Cab}	25.11 ± 3.45 ^{Ba}
	ST	14.43 ± 1.02 ^{Cb}	18.26 ± 1.68 ^{Bb}	12.05 ± 1.39 ^{Dc}	12.18 ± 0.85 ^{Db}	20.44 ± 1.18 ^{Bb}	21.20 ± 1.61 ^{Aa}
Phenol	NF	10.92 ± 0.82 ^{Cb}	3.60 ± 0.11 ^{Dc}	67.85 ± 7.38 ^{Aa}	16.75 ± 0.90 ^{Bb}	7.51 ± 0.84 ^{Db}	11.42 ± 1.12 ^{Ca}
	SF	27.03 ± 2.96 ^{Ba}	15.13 ± 0.90 ^{Cb}	17.51 ± 0.97 ^{Cb}	20.69 ± 2.92 ^{Ca}	69.03 ± 6.81 ^{Aa}	7.31 ± 0.67 ^{Db}
	ST	8.49 ± 0.89 ^{Bb}	19.91 ± 2.43 ^{Aa}	5.49 ± 0.75 ^{Cc}	5.61 ± 0.55 ^{Cc}	7.73 ± 0.38 ^{Bb}	5.33 ± 0.88 ^{Cc}
D-Limonene	NF	20.07 ± 2.83 ^{Ab}	16.36 ± 1.69 ^{Cb}	13.27 ± 1.24 ^{Cb}	13.52 ± 1.12 ^{Cb}	18.83 ± 2.10 ^{Bb}	20.61 ± 1.55 ^{Ab}
	SF	30.01 ± 3.60 ^{Aa}	14.45 ± 0.57 ^{Db}	11.73 ± 0.42 ^{Db}	15.89 ± 0.63 ^{Ca}	23.65 ± 1.49 ^{Ba}	23.32 ± 1.25 ^{Bb}
	ST	15.90 ± 1.58 ^{Cb}	28.29 ± 2.23 ^{Aa}	16.29 ± 1.70 ^{Ca}	16.50 ± 2.23 ^{Ca}	22.94 ± 2.51 ^{Ba}	29.50 ± 3.64 ^{Aa}

Table 2. *Cont.*

Volatile Flavor Compounds	Groups	0 d	1 d	7 d	14 d	21 d	28 d
Caryophyllene	NF	37.72 ± 5.8^{Cb}	34.58 ± 3.23^{Db}	35.88 ± 1.94^{Da}	29.96 ± 2.68^{Db}	48.19 ± 5.27^{Aa}	44.27 ± 3.31^{Bb}
	SF	69.13 ± 5.06^{Aa}	33.70 ± 2.69^{Db}	19.17 ± 2.87^{Eb}	43.10 ± 4.25^{Ca}	32.26 ± 3.37^{Db}	51.46 ± 6.40^{Ba}
	ST	33.36 ± 2.74^{Bb}	48.43 ± 5.83^{Aa}	21.31 ± 3.01^{Cb}	22.20 ± 1.49^{Cc}	44.45 ± 4.27^{Aa}	49.58 ± 3.63^{Aab}
3-Ethyl-2-methyl-1,3-hexadiene	NF	7.15 ± 0.73^{Cb}	21.53 ± 1.69^{Aa}	23.63 ± 2.41^{Aa}	17.29 ± 1.82^{Bb}	16.85 ± 1.64^{Bb}	1.99 ± 0.12^{Db}
	SF	7.77 ± 0.35^{Cb}	20.54 ± 3.02^{Ba}	24.07 ± 3.43^{Ba}	20.59 ± 1.98^{Ba}	27.28 ± 1.44^{Aa}	2.56 ± 0.26^{Da}
	ST	13.71 ± 1.55^{Ba}	12.19 ± 0.60^{Bb}	11.67 ± 1.69^{Bb}	12.87 ± 1.29^{Bc}	14.85 ± 1.06^{Ab}	2.54 ± 0.33^{Ca}
Oxime-, methoxy-phenyl-	NF	5.15 ± 0.25^{Da}	20.62 ± 10.00^{Bb}	33.58 ± 5.09^{Ab}	19.75 ± 1.10^{Bb}	12.80 ± 1.27^{Cc}	2.02 ± 0.16^{Da}
	SF	5.48 ± 0.51^{Ea}	31.65 ± 3.70^{Aa}	20.60 ± 2.20^{Bc}	11.00 ± 1.24^{Dc}	16.16 ± 0.47^{Cb}	2.04 ± 0.30^{Fa}
	ST	2.47 ± 0.14^{Db}	14.82 ± 0.86^{Ca}	51.84 ± 10.70^{Aa}	48.70 ± 7.11^{Aa}	25.12 ± 3.61^{Ba}	1.54 ± 0.18^{Db}
Anethole	NF	25.04 ± 3.31^{Db}	27.23 ± 3.86^{Da}	32.26 ± 2.46^{Ca}	33.09 ± 2.01^{Bb}	31.64 ± 2.35^{Cab}	39.78 ± 3.98^{Aa}
	SF	50.17 ± 7.23^{Aa}	10.29 ± 1.20^{Cb}	6.65 ± 0.65^{Cc}	39.67 ± 4.36^{Ba}	35.05 ± 2.42^{Ba}	37.36 ± 2.72^{Ba}
	ST	26.89 ± 2.45^{Bb}	26.06 ± 1.91^{Ba}	17.86 ± 2.10^{Cb}	18.89 ± 1.60^{Cc}	30.51 ± 2.28^{Ab}	27.59 ± 1.38^{Bb}
Pentadecane	NF	6.52 ± 0.78^{Aa}	4.25 ± 0.39^{Ca}	4.17 ± 0.48^{Ca}	3.89 ± 0.19^{Cb}	5.78 ± 0.85^{Bb}	5.33 ± 0.61^{Ba}
	SF	7.30 ± 0.68^{Aa}	4.94 ± 0.47^{Ca}	4.57 ± 0.24^{Ca}	5.13 ± 0.50^{Ca}	5.48 ± 0.60^{Bb}	5.51 ± 0.43^{Ba}
	ST	4.50 ± 0.62^{Bb}	4.65 ± 0.60^{Ba}	3.30 ± 0.33^{Cb}	3.54 ± 0.25^{Cb}	7.03 ± 0.83^{Aa}	5.25 ± 0.31^{Ba}

Different capital letters in the same row indicate significant differences in the same volatile flavor compound under different storage times within the same group; lowercase letters in the same column indicate significant differences among the three groups for the same volatile flavor at the same storage time.

4. Discussion

Fermentation is a key process in the production of fermented lamb liver paste and can give the product a special fermentation aroma and play a certain role in removing the muttony taste. There are few reports on the application of fermentation technology to liver products, and systematic research on the formulation and process is scarce. Mokhtar [21] compounded *Lactobacillus plantarum*, *Bifidobacterium lactis*, and *Bifidobacterium bifidum* into a starter culture, applied it to the production of sausages, and found that the contents of tyramine, putrescine, cadaverine, and tryptamine were significantly reduced. Zang [22] studied the mixed starter culture to help improve the flavor quality of traditional Chinese fermented fish. The Cenci-Goga [23] study found that the addition of dairy starter cultures and commercial probiotics inhibited the growth of undesirable microorganisms in salami and improved its sensory properties. Wang [24] used high-throughput sequencing technology to determine the bacterial community in sausages, dry-cured sausages, and smoked sausages and found that the main microorganisms in fermented sausages were Staphylococcus and Lactobacillus. Cano Garxia [25] found that yeast decomposes proteins and fats in dry fermented sausages to produce phenols and alcohols. Alcohols can react with lactic acid produced by lactic acid bacteria to give sausages a certain ester flavor. Antara [11] found that, after mixing *Lactobacillus plantarum* and *Pediococcus lactis* into sausages, the shelf life of fermented sausages was prolonged by metabolizing lactic acid to control enterotoxins. The main fermenting bacteria in fermented lamb liver paste are lactic acid bacteria, which participate in the metabolism of various substances during the fermentation process.

In the storage of lamb liver paste, the total number of colonies and lactic acid bacteria were higher in the NF and SF groups than in the ST group. On the one hand, this may have been due to the higher protein content of the NF group and SF group providing sufficient nitrogen sources for microbial growth and reproduction, and on the other, it may have been due to the addition of the starter culture increasing the total number of initial microorganisms [26]. With the increase in storage time, the total number of colonies in each group exhibited an upward trend. The NF group and the SF group exhibited the same trend.

The ST group underwent high-temperature treatment, which inactivated most of the heat-labile microorganisms. The microorganisms in the NF group and the SF group both grew and reproduced well under suitable fermentation conditions, increasing the number of microorganisms. After the 7th day of storage, the total number of colonies in the NF and SF groups was similar. However, after 14 days of storage, the TVB-N of the NF group continued to increase, and the highest degrees of amino acid destruction and lowest nutritional value were found in the lamb liver paste of the NF group at 28 days of storage. In conclusion, starter culture is of great significance to the quality of fermented meat products. In this research, the fermented lamb liver paste was made with lamb liver as the main raw material, and the paste products were made by adding an emulsifier, thickener, seasoning, other auxiliary materials, and starter culture. Probiotic products promote nutrient absorption in the small intestine, affect immune homeostatic cell signaling pathways in the intestinal mucosa, inhibit pathogenic bacteria, and improve intestinal health [27]. Therefore, a new type of lamb liver fermented product was developed in this experiment, which provides a theoretical basis for the high-value utilization of lamb liver.

Microorganisms decompose sugars to produce acid, which reduces the pH of meat products and prevents the growth of spoilage bacteria [13]. The pH value of the ST group was stable between 6.1 and 6.2. After the product is sterilized, a large number of microorganisms are inactivated, which does not affect the pH value of the product. The product was vacuum packed and stored at 4 °C. The environmental humidity of lamb liver paste storage was unchanged, and the Aw value of the three groups of lamb liver paste was not significantly different at any point. The L*, a*, and b* in the SF group were higher than in the ST group, which may be related to the effect of microorganisms in the fermentation process. With the increase in storage time, an upward trend was observed, with the SF group exhibiting significantly higher values than the other two groups. This situation shows that the use of leavening agents promotes the formation of product color. The microbes in the starter culture break down the proteins in the liver paste. The decreased adhesiveness of liver paste during storage is the result of microbial action. The ST group was treated at high temperature and pressure, and most of the microorganisms were inactivated, which also inactivated and denatured most of the proteins. This ultimately led to increased interstructural forces in the liver paste and the increased adhesion of the paste in the ST group.

The TVB-N and TBARS values in the SF group were lower than in the NF groups on the 1st day and 28th day, indicating that the fermentation technology ensures the quality of the product within 1 month of storage. The change in the TVB-N value in the NF group may have been caused by the mutual inhibition of bacteria in the early stage and the decrease in bacterial activity in the later stage. The analysis of the results on the 28th day showed that the addition of starter culture reduced the TVB-N value and improved the safety of lamb liver paste. There was no significant change in the ST group, indicating that the oxidation of fat was inhibited by autoclaving. In addition, lactic acid bacteria in the starter culture may also inhibit the production of malondialdehyde from peroxides and reduce fat oxidation. The addition of starter culture improved the freshness of lamb liver paste.

The protein content of the three groups of lamb liver paste was generally stable, i.e., remaining between 10 and 12%. This indicated that, as compared with fresh lamb liver (23.26%), the effect of 1-month storage time on protein content was relatively small. It is possible that the protein content of the lamb liver paste was reduced after the addition of water, emulsifiers, and thickeners. Studies have shown that under the dual decomposition of endogenous enzymes and microbial enzymes, protein degradation produces free amino acids as a taste substance [28]. The protein content of animal liver is lower as compared to the muscle [29]. Therefore, fewer proteins were available for microbial decomposition during the production of fermented products, resulting in insignificant changes in the protein content of the fermented liver paste. The low protein content of the ST group may have been caused by the destruction of the protein structure by autoclaving. The higher content of naturally fermented histones may have been due to the catabolism of proteins

by miscellaneous bacteria. The fat content of the NF group, the SF group, and the ST group changed from 3% to 4%, from 3.7% to 5.5%, and from 3% to 7%, respectively. Except for the 21st day, the fat content of the NF group and SF group was significantly lower than that of the ST group, probably due to the consumption of the carbon source by microorganisms during fermentation.

Liver-endogenous enzymes degrade fat and protein to produce free fatty acids, amino acids, and volatile flavor substances, which not only improve the nutritional value but also give fermented meat products a unique flavor [2,30]. Lactic acid bacteria are the focus of microbial starter culture screening for fermented meat products. They use carbohydrate fermentation to produce by-products, such as acetic acid, formic acid, and succinic acid, which have a certain positive effect on the flavor of fermented meat products. The results of the flavor analysis revealed several flavor compounds that we were concerned about during the storage period [8,31]. Aldehydes give lamb liver paste an almond and sweet aroma. Benzaldehyde is one of the representatives of the aromatic aldehydes that has cherry and nut aromas. Octanal, decanal, 2,4-decadienal, and tetradecanal have a gentle oily, slightly citrus, and iris-like aroma, with an aroma strength value of 2 but with a short duration [32]. The key aroma compounds in pork soup—2-undecenal and 2-octenaland hexanal—were characterized in the directional aroma analysis, which showed that the meat aroma was stronger in the SF group. Bai Shuang [33] found in an experiment on the formation mechanism of volatile compounds in fried mutton that aldehydes, alcohols, and esters produced by fat oxidation during the frying process were the main sources of volatile compounds. Alcohols are one of the main aroma compounds of fermented lamb liver paste. In addition, 1-Hexanol has an herbaceous aroma and also affects actin interactions [34]. Consistent with changes in hydrophobic interactions at the interface of myosin and actin in transition from a weakly to strongly bound state, 1-hexanol accelerated Pi release from myosin [35]. In addition, 1-heptanol also has a relatively strong fruit aroma and exhibits a binding ability to the myofibrillar protein, which can make the meat have a stronger flavor [36]. Furthermore, 1-Octen-3-ol has an important contribution to meat aroma. Mevalonate, a key substance in cholesterol synthesis, leads to a decrease in corticosterone content, affects ZNF414 and KLF15 gene expression, and positively regulates 1-octen-3-ol production in chicken [37]. The platycodon grandiflorum extracts anethole (40.27%) and 4-methoxy benzaldehyde (4.25%) can activate the acquired immune response and are beneficial to the human body [38]. Ketones are also part of the aroma of lamb liver paste. The fermented aroma compound 2-nonanone has a pleasing aroma and acts as a pheromone component to improve olfactory learning through persistent modulation of appetite motivation [39]. Limonene is the most common terpene in nature and a major constituent of several citrus oils (orange, lemon, mandarin, lime, and grapefruit). As a solvent for cholesterol, limonene has been clinically used to dissolve cholesterol-containing gallstones [40]. The limonene in the ST group was significantly higher than in the other two groups ($p < 0.05$). Caryophyllene is a class of bicyclic sesquiterpenes with cloves and turpentine aroma notes as functional food factors [41]. It is naturally found in lemon, nutmeg, and cinnamon leaf oils [42]. Studies have shown that exposure to volatile BCP in mice is detectable in the lung, olfactory bulb, brain, serum, heart, liver, kidney, epididymal adipose, and brown adipose tissue. Furthermore, inhaled volatile BCP is widely distributed in mouse tissues and affects the kinetics of metabolites in the liver [43]. Caryophyllene in the SF group and NF group was significantly higher than in the ST group ($p < 0.05$). As a new type of liver paste product, fermented lamb liver paste has a long storage period. In future research, the effect of functional substances on fermented lamb liver paste can be explored to improve its quality and nutritional value.

5. Conclusions

In conclusion, the probiotic fermented lamb liver paste in this study represents a high-value sheep by-product. The freshness, the number of lactic acid bacteria, and the L*, a*, and b* values were higher in the starter fermentation group, and the fat content was

lower. The lower pH in the early storage period of the starter fermentation group acted to inhibit the growth of microorganisms to a certain extent and prolong the storage period of fermented lamb liver paste. Volatile aroma compounds, such as aldehydes, 1-octen-3-ol, anethole, and 2-nonanone, as detected by GC–MS, contributed to the aroma composition of the fermented lamb liver paste.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/fermentation8120676/s1, Table S1: Changes of the total colony number in lamb liver paste with three different treatments during storage period; Table S2: Changes of the lactic acid bacteria colony count in lamb liver paste with three different treatments during storage; Table S3: Changes of pH value of lamb liver paste with three different treatments during storage period; Table S4: Changes of Aw value of lamb liver paste with three different treatments during storage period; Table S5: Changes of L* value of lamb liver paste with three different treatments during storage period; Table S6: Changes of a* value of lamb liver paste with three different treatments during storage period; Table S7: Changes of b* value of lamb liver paste with three different treatments during storage period; Table S8: Changes of TVB-N value in lamb liver paste with three different treatments during storage period; Table S9: Changes of TBARS value in lamb liver paste with three different treatments during storage period; Table S10: Changes of moisture content in lamb liver paste with three different treatments during storage period; Table S11: Changes of protein content in lamb liver paste with three different treatments during storage period; Table S12: Changes of fat content in lamb liver paste with three different treatments during storage period.

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