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Effect of *Pediococcus pentosaceus* Inoculation on Physicochemical Quality and Volatile Flavor Profiles of *Suanyu* During Fermentation

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Abstract

The effect of inoculating *Pediococcus pentosaceus* (*P. pentosaceus*) on the physicochemical quality and volatile flavor profiles of fermented *Suanyu* was explored. Compared to natural fermentation, inoculation with *P. pentosaceus* significantly increased whiteness, accelerated the decrease in water activity and pH, altered the temporal TBARS pattern, enhanced gel strength and textural properties (hardness, gumminess and chewiness), and improved sensory properties. A total of 31 volatile compounds were detected in *Suanyu*, including 10 alcohols, 7 aldehydes, 6 ketones, 4 esters, 1 acid, and 3 hydrocarbons. The total signal intensity of volatile compounds in the *P. pentosaceus*-inoculated group was significantly higher than in the natural fermentation group, especially for aldehydes and ketones. Overall, the inoculation of *P. pentosaceus* effectively enhanced the physicochemical quality and volatile flavor profiles of *Suanyu*, offering a theoretical basis for its industrial-scale fermentation.

Keywords: quality; volatile flavor; *Suanyu*; *Pediococcus pentosaceus*

1. Introduction

As the most extensively cultivated freshwater fish species in China, grass carp (*Ctenopharyngodon idellus*) plays a pivotal role in national aquaculture. In 2023, its production accounted for approximately 21.44% of the country's total freshwater fish output, reaching 5.94 million tons [1]. Valued for its cost-effectiveness and nutritional richness, particularly its high protein and polyunsaturated fatty acid content, grass carp contributes positively to human dietary balance and health [2]. However, its broader processing and utilization are limited by several inherent drawbacks, including a distinct earthy odor, numerous intermuscular spines, high moisture content, elevated endogenous enzyme activity, and rapid spoilage [3].

Fermentation is a well-established food preservation technique that has evolved alongside human civilization and remains relevant in modern food systems. Traditional fermented fish products such as *Suanyu*, a regional specialty from Southwest China, are produced through biochemical transformations driven by environmental microorganisms and endogenous fish enzymes [4]. Under controlled temperature and humidity, ingredients including fish, salt, spices, and cooked grains (e.g., corn flour) undergo microbial

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succession, typically dominated by lactic acid bacteria (LAB). These bacteria metabolize carbohydrates into organic acids, primarily lactic acid, lowering pH and suppressing spoilage organisms. Concurrently, enzymatic proteolysis and lipolysis modify texture, enhance nutritional profiles, and generate characteristic flavors [5–7].

Traditional spontaneous fermentation is often associated with inconsistent product quality, extended processing time, and nutrient loss [8]. To address these limitations, controlled fermentation using selected starter cultures has been adopted to standardize production and improve quality. Studies have demonstrated the effectiveness of LAB such as *Lactiplantibacillus plantarum* and *Saccharomyces cerevisiae* in enhancing fermented fish products [9,10]. Among LAB, *Pediococcus pentosaceus* has been identified as a dominant autochthonous bacterium in fermented fish and is frequently applied as an exogenous culture in sausage fermentation. For instance, Li et al. reported that *P. pentosaceus* addition accelerated volatile flavor formation and reduced spoilage microorganism abundance in tilapia sausage [11]. Similarly, Zhao et al. linked *P. pentosaceus* metabolism to flavor development in Sichuan sausages via pathways such as arginine biosynthesis and glycerophospholipid metabolism [12]. Despite these promising applications, the specific effects of *P. pentosaceus* monoculture inoculation on *Suanyu* quality remain underexplored. A systematic comparison of physicochemical and flavor profiles between spontaneous and *P. pentosaceus*-inoculated fermentation is needed to elucidate its potential in optimizing traditional fermented fish production.

Therefore, the main goal of this study was to investigate the effects of *P. pentosaceus* inoculation on physicochemical quality and volatile flavor profiles during the fermentation of *Suanyu*, and the related indexes, including colorimetry, pH, water activity (Aw), thiobarbituric acid reactive substances (TBARS), gel strength, texture profile analysis (TPA), sensory evaluation, and volatile compounds, were analyzed. The naturally fermented *Suanyu* sample was used as the control. The study may provide some valuable insights into the industrial production of high-quality *Suanyu* products.

2. Materials and Methods

2.1. Experimental Materials

Thirty fresh grass carp weighing 3.0 ± 0.2 kg were purchased from a supplier at the Huguang Vegetable Market in Mazhang District, Zhanjiang City, Guangdong Province. *P. pentosaceus* (strain no. 192624) was purchased from Beijing Beina Chuanglian Biotechnology Research Institute. Salt, white sugar, and corn flour were purchased from the Fu you Wanjia Supermarket in Mazhang District, Zhanjiang City, Guangdong Province. All other chemicals were analytically pure and supplied by Sinopharm Chemical Reagent Co. Ltd. (Shanghai, China).

2.2. Preparation of P. pentosaceus

P. pentosaceus powder was activated in MRS liquid medium and preserved on slant culture at 4 °C. A single colony was picked and cultured in MRS at 37 °C for 24 h. Then, 0.1 mL of the bacterial suspension was inoculated into 10 mL of MRS and incubated in a shaker at 37 °C for 16 h. After cultivation, the bacterial cells were collected by centrifugation at 4 °C and 9700× *g* for 10 min, then washed twice with an equal volume of physiological saline and resuspended in physiological saline. The final concentration of the bacteria was adjusted to 10^8–10^9 CFU/mL.

2.3. Preparation of Suanyu Samples

The preparation of *Suanyu* samples was carried out according to the previous study with slight modifications [13]. Thirty fish were randomly divided into the naturally fermented

(NF) and the fermented with *P. pentosaceus* (PF) groups (15 fish per group). Each group was further divided into three independent fermentation batches (5 fish per batch). For each batch, dorsal muscle samples were pooled, rinsed, cut into cubes (3 × 2 × 2 cm), and processed in separate fermentation containers. The fish cubes were then thoroughly mixed with white sugar (2% *w*/*w*) and salt (3% *w*/*w*), followed by marination at 4 °C for 24 h. After marination, 20% (*w*/*w*) roasted corn flour and 1% (*v*/*w*) *P. pentosaceus* culture solution were incorporated into the fish pieces and mixed uniformly. The mixtures were transferred into fermentation containers, which were covered with lids and sealed with water. Fermentation was carried out at 25 °C for 15 days. Samples fermented with *P. pentosaceus* (PF) were collected on days 0, 5, 10, and 15, with a naturally fermented (NF) group sample serving as the control. The NF control was mixed with an equal volume (1%, *v*/*w*) of sterile physiological saline instead of the *P. pentosaceus* culture. For each treatment and each sampling time point, samples were obtained from three independently prepared fermentation containers (n = 3 biological replicates).

2.4. Determination of Color

The color properties of *Suanyu* samples were analyzed using a CR9 Chroma meter (Shenzhen Sanenji Technology Co., Ltd., Shenzhen, China). The color parameters were presented as L* (lightness), a* (positive value indicating red; negative value indicating green), and b* (positive value indicating yellow; negative value indicating blue), and the whiteness index was subsequently calculated according to the following Equation (1):

$$\text{Whiteness} = 100 - \sqrt{(100 - L^*)^2 + a^{*2} + b^{*2}} \quad (1)$$

2.5. Determination of pH and Water Activity (Aw)

The pH values of *Suanyu* samples were determined using the PHSJ-6L digital pH-meter (Shanghai Yile Scientific Instruments Co., Ltd., Shanghai, China). The Aw of the sample was measured with a water activity meter at 25 °C [14].

2.6. Determination of Texture Profile and Gel Strength

The texture profile and gel strength of *Suanyu* samples were determined with a texture analyzer (TA. XT plus C, Stable Micro System, Godalming, UK), following the method described by Liu et al. [15]. For texture profile analysis, the instrument was set to a crosshead speed of 1 mm/s, a target distance of 15 mm, two compressions per sample with a 1 s interval, and a 5 kg load cell. Gel strength was assessed under the following conditions: a displacement of 1.5 cm, a trigger load of 5.0 g, and a test speed of 1.0 mm/s. Gel strength value was calculated as the product of breaking strength and depression depth.

2.7. Determination of Thiobarbituric Acid Reactive Substance (TBARS)

A 1 g sample was homogenized in 10 mL of ice-cold 7.5% (*w*/*v*) trichloroacetic acid (TCA) for 1 min, then incubated at 4 °C for 10 min. Subsequently, 3 mL of the filtrate was combined with 5 mL of 0.02 mol/L 2-thiobarbituric acid and heated in a boiling water bath for 40 min. The absorbance of the resultant solution was measured at 532 nm. The TBARS value, expressed as mg malondialdehyde (MDA)/kg, was quantified against a standard curve prepared with 1,1,3,3-tetraethoxypropane (TEP) [16].

2.8. Sensory Evaluation Test

Twelve sensory panelists were invited for the sensory analysis, which included evaluating odor, color, texture, taste and overall acceptance. All panelists passed screening tests based on ISO 8586 standards (2023) [17]. Panelists underwent three preliminary training

sessions (3 h each) to establish consensus on sensory attributes. A 5-point intensity scale (1 = lowest, 5 = highest) was used for the analytical attributes (odor, color, texture, and taste), whereas overall acceptance was evaluated separately as a hedonic attribute using a 5-point acceptability scale [18].

2.9. Gas Chromatography–Ion Mobility Spectrometry (GC-IMS) Analysis

GC-IMS was used to determine the volatile flavor components of fermented *Suanyu* samples according to the method of Xiao et al. [19]. First, 2.0 g of sample was placed in a 20 mL headspace glass sampling bottle and incubated at 60 °C with a rotation speed of 500 r/min for 15 min. The sample headspace gas (500 μL) was injected into an MXT-5 capillary column (15 m × 0.53 mm i.d. × 10 μm film thickness) using splitless injection, and the injection rate and temperature were 30 mL/min and 85 °C, respectively. The incubation speed was 500 rpm. Chromatographic separation was performed using gas chromatography (GC) coupled to ion mobility spectrometry (IMS) maintained at 45 °C. The GC run time was 30 min. GC was performed with nitrogen (≥99.999%) as the carrier gas at an initial flow of 2.0 mL min^{-1} (held 2 min), raised to 10 mL min^{-1} over 8 min, then to 100 mL min^{-1} over the next 10 min and maintained for 10 min. Each sample was tested three times, and the retention index (RI) was calculated using normal paraffin (C4-C9) chromatographic reagents as external references. Volatile compounds were qualitatively determined by comparing the RI and drift time of standards in the GC-IMS library search.

2.10. Statistical Analysis

All analytical measurements were performed in triplicate on each biological replicate. To account for the repeated measures structure (the same three independent fermentation vessels were sampled over time), the longitudinal fermentation data were analyzed using a linear mixed-effects model, with treatment, fermentation time, and their interaction as fixed effects, and fermentation batch as a random intercept. Post hoc multiple comparisons were conducted using Tukey's test. The significance level was set at $p < 0.05$. The results are visualized using Origin2019b software (OriginLab Corporation, Northampton, MA, USA). Fingerprint analysis of volatile compounds was carried out using the Gallery Plot plugin.

3. Results and Discussion

3.1. Physicochemical Quality Analysis

3.1.1. Color Analysis

Color serves as a key quality attribute in the assessment of fermented fish products and significantly influences consumer acceptance [20]. As shown in Figure 1A–C, both NF- and PF-group *Suanyu* samples demonstrated a continuous decline in L* values during the whole fermentation process, accompanied by a steady rise in a* and b* values. Meanwhile, as the fermentation time increased, the whiteness of both the NF- and PF-group *Suanyu* samples decreased gradually (Figure 1D), indicating that fermentation could reduce the color quality of fish meat. However, it is notable that in the later stage of fermentation (10–15 d), compared with the samples in the NF group, significantly higher whiteness was observed in the samples of the PF group, suggesting that inoculation of *P. pentosaceus* can effectively improve the color and quality of *Suanyu*.

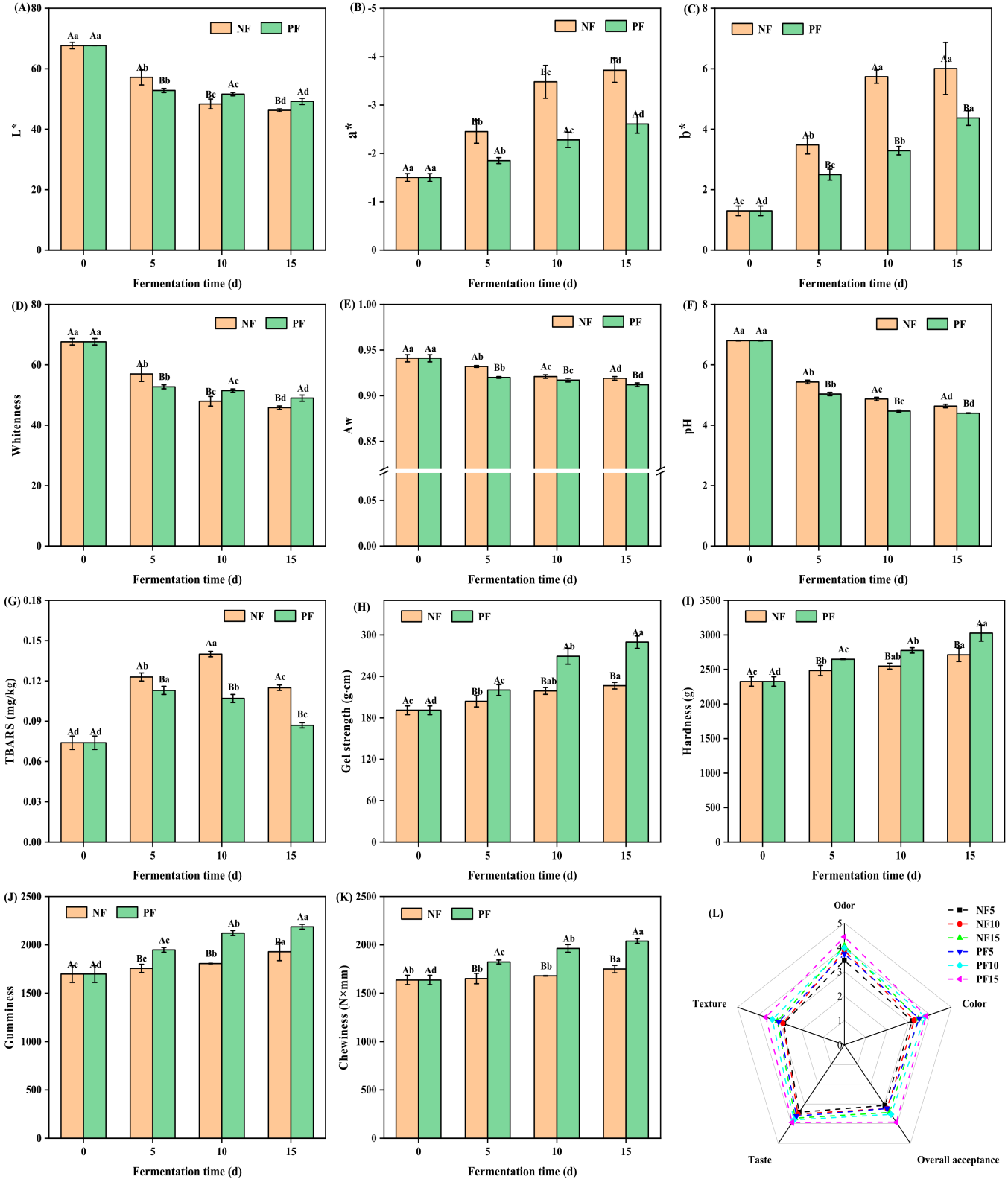


Figure 1. Physicochemical quality analysis of *Suanyu* samples during natural fermentation and fermentation inoculated with *P. pentosaceus*. (**A**) *L* value*; (**B**) *a* value*; (**C**) *b* value*; (**D**) *Whiteness*; (**E**) *Aw (water activity)*; (**F**) *pH*; (**G**) *TBARS value*; (**H**) *Gel strength*; (**I**) *Hardness*; (**J**) *Gumminess*; (**K**) *Chewiness*; (**L**) *Sensory radar plot*. Different lowercase letters indicate significant differences among fermentation time points within the same treatment ($p < 0.05$), and different uppercase letters indicate significant differences between treatments at the same time point ($p < 0.05$).

3.1.2. Aw Analysis

Low Aw is widely recognized to inhibit microbial growth and extend the shelf-life of fermented fish products [13]. As shown in Figure 1E, the Aw of Suanyu samples decreased under both natural fermentation and fermentation inoculated with P. pentosaceus. The initial Aw of the sample was 0.941±0.004, significantly lower than that of fresh fish flesh, likely due to the salt-curing pretreatment. Salt-curing has been shown to markedly reduce Aw in meat products, thereby delaying spoilage [21]. Throughout fermentation, Aw continued to decline in both treatments, which may be linked to the reduction in pH. Related studies indicated that the water-holding capacity of muscle products decreases significantly as pH drops or approaches the protein isoelectric point [22]. After 10 days of natural fermentation, Aw reached 0.921, whereas a similar value was attained in the *P. pentosaceus*-inoculated group after only 5 days. This accelerated decline in Aw may be related to enhanced protein hydrolysis promoted by *P. pentosaceus*, which reduces water retention capacity.

3.1.3. pH Analysis

pH is recognized as a critical factor influencing the safety of fermented products [23]. The pH values of *Suanyu* samples subjected to natural fermentation and to fermentation inoculated with *P. pentosaceus* are presented in Figure 1F. The initial pH of the *Suanyu* sample was 6.80. Both groups showed a decreasing trend throughout the fermentation period. Notably, the pH of the NF-group *Suanyu* sample decreased to 4.63 in 15 days, whereas the pH of the PF-group sample decreased to 4.47 within 10 days. These results demonstrated that inoculation with *P. pentosaceus* could accelerate acidification in *Suanyu*, thus contributing to product stability.

3.1.4. TBARS Analysis

The TBARS assay is widely used to evaluate lipid oxidation in meat and meat products [24]. Higher TBARS levels indicate the accumulation of secondary oxidation products, and acceptable fish products should not exceed 5 mg MDA/kg [10]. As shown in Figure 1G, as fermentation time increased, the TBARS levels of *Suanyu* samples first increased and then decreased under both natural fermentation and inoculated fermentation with *P. pentosaceus*. The sample exhibited an initial TBARS value of 0.074 mg/kg. For the NF group sample, the TBARS value peaked (0.140 MDA/kg) after 10 days, whereas the PF group reached the maximum level (0.113 MDA/kg) after 5 days of fermentation. Similar trends were found in the study of Gao et al. [16]. Notably, the TBARS value decreased in the two fermentation conditions at the end of fermentation, which might be attributed to the degradation of aldehydes or additional interactions with other compounds, such as Maillard reaction intermediates and amino acids, resulting in the formation of various flavor-related compounds [24]. In addition, at the end of fermentation (day 15), the TBARS value of the PF group (0.115 mg MDA/kg) was significantly higher than that of the NF group (0.087 mg MDA/kg) ($p < 0.05$), indicating a higher TBARS level in the inoculated group at the final stage.

3.1.5. Gel Strength and TPA Analysis

Gel strength and textural properties are major physical characteristics of fermented fish products. As shown in Figure 1H, the gel strengths of *Suanyu* samples increased under both natural fermentation and inoculated fermentation with *P. pentosaceus*. After 15 days of fermentation, the gel strength of the NF-group *Suanyu* sample was 226.5 g·cm, while that of the PF-group *Suanyu* sample reached 289.5 g·cm, indicating that the inoculation of *P. pentosaceus* could improve the gel quality of the *Suanyu* samples compared to natural fermentation. The further enhanced gel strength might be related to the lower pH value

produced by the fermentation of fish meat due to the inoculation of *P. pentosaceus*. The low pH conditions may accelerate the aggregation of proteins, triggering the gelation phenomenon induced by acidic substances [25]. In addition, as the fermentation time increased, the hardness, gumminess and chewiness of the two groups of *Suanyu* all showed a gradually increasing trend (Figure 1I–K). Meanwhile, significantly higher hardness, gumminess and chewiness existed in the PF group sample, indicating that the addition of *P. pentosaceus* could enhance the rigidity of the *Suanyu* samples. The accelerated loss of water in fish meat and the acid-induced protein gelation of fish meat might enhance the hardness, gumminess and chewiness of fermented fish meat [10].

3.1.6. Sensory Evaluation Analysis

The results of the sensory evaluation of *Suanyu* samples subjected to natural fermentation and fermentation inoculated with *P. pentosaceus* are shown in Figure 1L. The five sensory attribute scores of the two groups of *Suanyu* samples increased with the extension of fermentation time. After 15 days of fermentation, the odor, color, texture, taste and overall acceptability scores of the PF samples were significantly higher than those of the NF group, indicating that inoculation fermentation with *P. pentosaceus* could significantly improve the sensory quality of *Suanyu*.

3.2. Volatile Flavor Analysis

3.2.1. GC-IMS Fingerprint Chromatogram Analysis of Volatile Compounds in *Suanyu* Samples

Volatile compounds in *Suanyu* samples with natural fermentation and fermentation inoculated with *P. pentosaceus* were analyzed by HS-GC-IMS. The three-dimensional top view, two-dimensional vertical plane view, and fingerprint spectra of volatile compounds of the *Suanyu* samples are displayed in Figure 2A–C. As shown in Figure 2A, the X-, Y-, and Z-axes corresponded to drift time, retention time, and peak signal intensity, respectively. A color gradient from blue to red encoded relative abundance, with intensifying red hues indicating progressively higher signal intensities [26]. To comprehensively compare the changes in their flavor components, dimensionality reduction techniques were employed to obtain the two-dimensional (2D) spectrum (Figure 2B). The vertical axis of the two-dimensional spectrum represents the retention time of gas chromatography, while the horizontal axis represents the ion migration time [27]. On the blue background, the red vertical line at the horizontal coordinate of 1.0 indicates the normalized reaction ion peak (RIP). Each individual data point located on both sides of the RIP represents a volatile compound, and the color intensity reflects the concentration level of the corresponding molecule. Signals are abundant in the retention time range of 200 s to 400 s and the ion migration time range of 1.0 ms to 1.5 ms, indicating a high content of valid volatile compounds in *Suanyu* samples within this region. As presented in Figure 2A–C, although the volatile compounds in the two groups of *Suanyu* samples at different fermentation times showed similarities in terms of types, the signal intensity of each volatile compound exhibited significant differences between natural fermentation and fermentation with *P. pentosaceus*. As presented in Figure 2D, the total signal intensity of volatile compounds in the *Suanyu* samples gradually increased with the extension of fermentation time. Among them, the total signal intensity of volatile compounds of the PF group sample was significantly higher than that of the NF sample, indicating that *P. pentosaceus* inoculation could effectively enhance the volatile flavor of *Suanyu* samples.

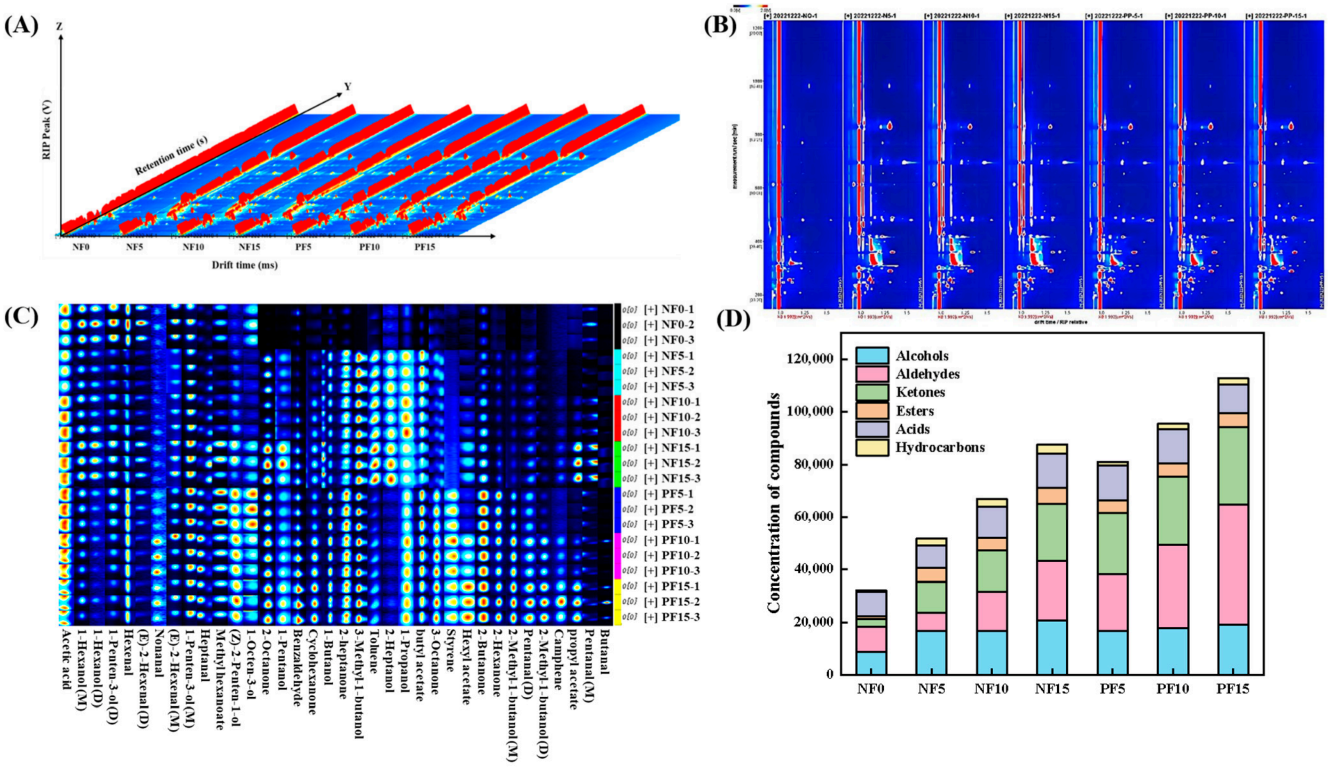


Figure 2. GC-IMS fingerprint chromatogram analysis of volatile components in *Suanyu* samples during natural fermentation and fermentation inoculated with *P. pentosaceus*. (**A**) Three-dimensional top view of the GC-IMS spectra, with the X-, Y-, and Z-axes representing drift time, retention time, and peak intensity, respectively. (**B**) Two-dimensional vertical plane view of the GC-IMS spectra, with the vertical axis representing GC retention time and the horizontal axis representing ion mobility drift time; the red vertical line at a drift time of 1.0 ms indicates the normalized reaction ion peak (RIP), and each data point on either side of the RIP represents a volatile compound. (**C**) Fingerprint spectra of volatile compounds in *Suanyu* samples from the NF and PF groups at different fermentation time points. (**D**) Total signal intensity of volatile compounds in *Suanyu* samples during fermentation.

3.2.2. Differences in Volatile Compounds of *Suanyu Samples*

A total of 36 volatile compounds were identified in the *Suanyu* samples (Table 1). Among these volatile compounds, 5 compounds (e.g., 1-penten-3-ol, 2-methyl-1-butanol, 1-hexanol, pentanal, and (E)-2-hexenal) were detected in monomeric and dimeric forms. As a result, 31 unique volatile compounds were identified in *Suanyu* samples (Table 1 and Figure 2), of which 10 were alcohols, 7 were aldehydes, 6 were ketones, 4 were esters, 1 was an acid, and 3 were hydrocarbons. It is worth noting that compared to the NF group, significantly higher total signal intensities of alcohols, aldehydes, and ketones were observed in the PF group during the fermentation process.

During fermentation of *Suanyu*, the accumulation of alcohol primarily originates from the oxidative degradation of polyunsaturated fatty acids [28]. After 15 days of fermentation, the total alcohol signal intensity in the NF and PF groups was comparable. Notably, the signal intensity of 2-methyl-1-butanol exhibited an increasing trend throughout the fermentation process in both groups, with significantly higher values observed in the PF group than in the NF group. 2-methyl-1-butanol has a wine, grassy and fatty odor [29]. In this study, the signal intensity of 1-penten-3-ol with a fruity, grassy, tomato, and mint odor in the NF group first decreased and then increased, whereas in the PF group it initially increased before declining. At 5 and 10 days of fermentation, the signal intensity of 1-penten-3-ol in the PF group was significantly higher than in the NF group, indicating that the inoculation fermentation with *P. pentosaceus* can accelerate the formation of 1-penten-3-ol.

Table 1. Volatile compounds analysis of *Suanyu* samples during natural fermentation and fermentation inoculated with *P. pentosaceus*.

Volatiles	CAS#	Formula	Retention Time [s]	Drift Time [s]	Retention Index	Peak Area (a.u.)						
						NF0	NF5	NF10	NF15	PF5	PF10	PF15
Alcohols (13)												
1-Propanol	C71238	C_3H_8O	390.184	1.08527	1027.2	261.97 ± 80.52	627.86 ± 38.15	804.92 ± 23.98	558.45 ± 28.33	718.93 ± 14.23	752.88 ± 49.96	741.22 ± 37.45
1-Butanol	C71363	$C_4H_{10}O$	490.786	1.17003	1105.8	652.52 ± 30.26	3276.21 ± 64.05	3219.08 ± 163.81	3997.88 ± 100.31	2799.36 ± 82.43	3218.41 ± 192.49	3400.85 ± 60.09
1-Penten-3-ol (M)	C616251	$C_5H_{10}O$	613.456	0.93996	1104.8	1609.00 ± 154.28	1406.84 ± 15.83	1761.31 ± 69.52	2209.50 ± 67.05	2472.70 ± 30.06	2226.04 ± 56.57	2109.38 ± 18.33
1-Penten-3-ol (D)	C616251	$C_5H_{10}O$	613.456	1.35878	1174.6	528.59 ± 71.15	192.26 ± 28.90	211.78 ± 29.11	444.88 ± 47.72	815.58 ± 6.81	551.61 ± 54.93	497.77 ± 14.86
(Z)-2-Penten-1-ol	C1576950	$C_5H_{10}O$	770.762	1.61	1267.3	63.07 ± 7.48	93.06 ± 5.58	104.27 ± 10.03	150.87 ± 49.67	220.77 ± 11.88	247.52 ± 3.11	251.41 ± 29.49
1-Pentanol	C71410	$C_5H_{12}O$	696.427	1.80257	1222.9	96.78 ± 12.45	1077.70 ± 58.36	1334.08 ± 54.32	2223.00 ± 254.23	1284.98 ± 23.54	1578.53 ± 1.40	1944.04 ± 19.12
3-Methyl-1-butanol	C123513	$C_5H_{12}O$	698.054	1.49641	1223.9	388.62 ± 32.93	6500.31 ± 336.35	5228.63 ± 68.93	5801.23 ± 53.41	4159.35 ± 30.24	4580.08 ± 244.71	5117.73 ± 240.36
2-Methyl-1-butanol (M)	C137326	$C_5H_{12}O$	534.993	1.20975	1132.4	88.07 ± 2.63	302.21 ± 20.62	361.66 ± 18.65	430.07 ± 14.43	784.72 ± 3.16	1476.61 ± 61.52	1799.21 ± 27.71
2-Methyl-1-butanol (D)	C137326	$C_5H_{12}O$	534.292	1.44	1132	102.38 ± 5.78	117.91 ± 6.09	144.86 ± 6.92	218.90 ± 8.09	194.78 ± 3.79	571.78 ± 48.74	917.28 ± 33.92
1-Hexanol (M)	C111273	$C_6H_{14}O$	983.844	1.32867	1369.4	2795.34 ± 229.69	1702.02 ± 39.87	2006.08 ± 230.67	2577.88 ± 70.95	1805.47 ± 123.60	1481.29 ± 246.38	1343.75 ± 326.63
1-Hexanol (D)	C111273	$C_6H_{14}O$	982.429	1.64239	1368.8	484.85 ± 63.99	327.74 ± 11.90	409.17 ± 71.18	515.21 ± 24.72	272.31 ± 45.87	227.70 ± 49.43	228.63 ± 38.68
2-Heptanol	C543497	$C_7H_{16}O$	982.429	1.3933	1368.8	150.42 ± 26.44	372.11 ± 24.73	367.61 ± 18.74	479.23 ± 23.26	106.97 ± 6.37	123.64 ± 17.28	134.75 ± 21.78
1-Octen-3-ol	C3391864	$C_8H_{16}O$	1070.098	1.1624	1469.4	1503.47 ± 140.61	640.29 ± 21.64	790.46 ± 52.39	1140.25 ± 49.84	1066.51 ± 85.16	793.49 ± 34.79	690.47 ± 69.88
Aldehydes (9)												
Butanal	C123728	C_4H_8O	250.684	1.28382	832.2	84.36 ± 8.20	292.16 ± 27.73	155.03 ± 11.98	382.37 ± 28.98	95.95 ± 6.84	166.45 ± 96.28	402.93 ± 25.96
Pentanal (M)	C110623	$C_5H_{10}O$	400.332	1.39134	1036.1	485.44 ± 203.55	255.17 ± 24.36	683.31 ± 53.37	1633.00 ± 29.54	465.20 ± 42.24	482.79 ± 23.80	475.41 ± 67.78
Pentanal (D)	C110623	$C_5H_{10}O$	402.535	1.4204	1009.7	75.46 ± 6.52	144.39 ± 15.48	145.22 ± 3.69	211.35 ± 20.15	217.72 ± 7.46	308.28 ± 28.75	437.07 ± 20.00
Hexanal	C66251	$C_6H_{12}O$	480.007	1.25873	1098.9	2448.79 ± 410.31	1196.22 ± 158.88	1812.56 ± 316.98	2077.49 ± 93.42	3191.03 ± 42.16	2942.37 ± 56.49	2684.63 ± 98.64
(E)-2-Hexenal (M)	C6728263	$C_6H_{10}O$	581.307	1.18395	1158	453.83 ± 31.24	350.73 ± 8.23	392.49 ± 17.34	500.67 ± 31.91	370.33 ± 39.40	451.76 ± 163.62	383.72 ± 76.48
(E)-2-Hexenal (D)	C6728263	$C_6H_{10}O$	770.61	1.50782	1267.2	435.50 ± 103.15	177.04 ± 9.59	219.52 ± 26.59	349.67 ± 19.80	260.22 ± 37.27	200.37 ± 16.30	184.27 ± 14.25
Heptanal	C111717	$C_7H_{14}O$	661.502	1.33157	1200.3	263.94 ± 29.21	395.78 ± 29.00	541.82 ± 60.00	752.80 ± 40.27	622.46 ± 72.66	862.18 ± 57.36	882.16 ± 42.49

Table 1. *Cont.*

Volatiles	CAS#	Formula	Retention Time [s]	Drift Time [s]	Retention Index	Peak Area (a.u.)						
						NF0	NF5	NF10	NF15	PF5	PF10	PF15
Nonanal	C124196	$C_9H_{18}O$	1069.549	1.47403	1403.8	345.18 ± 25.41	338.38 ± 6.28	342.27 ± 37.57	377.39 ± 20.66	387.34 ± 7.74	552.35 ± 28.42	484.23 ± 51.47
Benzaldehyde	C100527	C_7H_6O	1331.365	1.15475	1493.9	5075.61 ± 2165.04	3891.85 ± 557.81	10552.77 ± 2217.59	16178.69 ± 1051.10	16002.33 ± 1040.75	25665.87 ± 2145.09	39665.17 ± 932.80
Ketones (6)												
2-Butanone	C78933	C_4H_8O	300.919	1.25185	917.1	1091.54 ± 81.84	1728.72 ± 80.00	2287.92 ± 333.79	2734.23 ± 315.25	6265.39 ± 111.80	6068.32 ± 136.89	6400.65 ± 212.81
2-Hexanone	C591786	$C_6H_{12}O$	399.771	1.4813	1035.7	162.70 ± 4.05	205.52 ± 11.14	180.30 ± 12.49	280.70 ± 15.57	700.18 ± 41.97	643.33 ± 56.78	788.02 ± 26.40
2-Heptanone	C110430	$C_7H_{14}O$	360.043	1.37359	999.3	412.69 ± 129.79	3563.38 ± 83.18	3361.81 ± 32.19	3082.49 ± 37.11	3983.55 ± 19.89	3963.79 ± 39.93	3974.05 ± 25.68
Cyclohexanone	C108941	$C_6H_{10}O$	766.22	1.16719	1264.7	328.63 ± 5.56	536.38 ± 27.09	870.21 ± 51.30	1146.1833.32	787.05 ± 17.91	1345.22 ± 34.42	1619.78 ± 55.96
2-Octanone	C111137	$C_8H_{16}O$	833.591	1.33555	1301.2	441.99 ± 73.60	4850.25 ± 719.93	8192.84 ± 351.01	15062.35 ± 940.65	10429.35 ± 229.59	12692.01 ± 80.71	15615.90 ± 77.84
3-Octanone	C106683	$C_8H_{16}O$	773.425	1.7174	1268.8	326.04 ± 14.32	800.73 ± 87.27	653.46 ± 33.77	671.61 ± 36.32	979.56 ± 30.45	1079.71 ± 13.39	1095.85 ± 15.24
Esters (4)												
Propyl acetate	C109604	$C_5H_{10}O_2$	328.969	1.47717	958.5	227.97 ± 2.66	64.68 ± 7.41	201.55 ± 21.96	805.29 ± 45.63	166.18 ± 13.26	290.03 ± 17.29	754.64 ± 38.59
Butyl acetate	C123864	$C_6H_{12}O_2$	417.504	1.25093	1050.7	454.51 ± 63.78	5084.15 ± 95.83	4276.91 ± 44.75	4567.44 ± 113.81	3792.26 ± 8.20	4066.58 ± 157.64	3698.64 ± 131.95
Methyl hexanoate	C106707	$C_7H_{14}O_2$	655.18	1.25942	1196.1	421.25 ± 27.57	159.92 ± 12.21	248.04 ± 24.86	590.45 ± 131.39	847.16 ± 9.11	572.79 ± 15.88	537.82 ± 18.27
Hexyl acetate	C142927	$C_8H_{16}O_2$	766.443	1.44023	1264.8	30.49 ± 2.66	51.37 ± 7.83	89.19 ± 12.93	150.08 ± 3.43	72.72 ± 2.36	184.66 ± 12.22	287.11 ± 2.27
Acids (1)												
Acetic acid	C64197	$C_2H_4O_2$	1338.518	1.05254	1496.1	9191.26 ± 1774.59	8370.07 ± 612.56	11993.24 ± 682.44	12908.74 ± 101.62	13209.29 ± 115.40	13116.17 ± 207.98	10968.51 ± 250.26
Hydrocarbons (3)												
Styrene	C100425	C_8H_8	789.7	1.08514	1277.9	65.15 ± 13.60	71.08 ± 7.20	80.87 ± 4.00	76.63 ± 4.92	383.35 ± 20.94	432.40 ± 24.29	384.65 ± 3.52
Toluene	C108883	C_7H_8	425.44	1.04751	1057.3	544.98 ± 98.02	2553.35 ± 69.20	2793.52 ± 141.89	3249.79 ± 8.04	981.07 ± 9.36	1463.93 ± 32.19	1694.80 ± 19.87
Camphene	C79925	$C_{10}H_{16}$	543.191	1.74737	1137.1	47.59 ± 3.82	63.98 ± 5.77	84.45 ± 18.28	136.30 ± 15.92	65.25 ± 3.96	148.34 ± 15.48	310.53 ± 88.25

Note: M and D represent the monomeric and dimeric forms of volatile compounds, respectively.

Aldehydes, which contribute pleasant aromas, are predominantly formed by the oxidation of monounsaturated fatty acids and ω-6 polyunsaturated fatty acids [30]. During fermentation, the PF group exhibited a significantly higher total aldehyde signal intensity than the NF group. Benzaldehyde, responsible for an almond and cherry odor, was the most abundant aldehyde in both groups. Over the fermentation period, benzaldehyde signal intensity levels increased in both treatment groups, with the PF group demonstrating markedly higher signal intensity than the NF group. In the NF group, hexanal first declined and then increased, whereas in the PF group it initially increased before decreasing. In addition, after 15 days of fermentation, the butanal, hexanal, heptanal, and nonanal, which showed a fatty, fruity and green grass odor [30–33], remained higher in the PF group than in the NF group, indicating that inoculation fermentation with *P. pentosaceus* can significantly increase the production of straight-chain aldehydes.

Ketone compounds endow fermented foods with unique fruity and aromatic odors, and these substances primarily originate from the autoxidation of UFAs or the amino acid degradation [34]. After 15 days of fermentation, the total signal intensity of ketones in the PF group was significantly higher than that in the NF group, and the signal intensity levels of six ketones (2-butanone, 2-hexanone, 2-heptanone, cyclohexanone, 2-octanone, and 3-octanone) were all higher in the PF group than in the NF group, suggesting that inoculation fermentation with *P. pentosaceus* can promote the formation of fruity and aromatic odors.

4. Conclusions

The inoculation of *P. pentosaceus* during *Suanyu* fermentation markedly influenced physicochemical and flavor characteristics. It enhanced whiteness, accelerated acidification and water activity reduction, and improved gel strength and textural properties (i.e., hardness, gumminess and chewiness). Sensory results showed that *Suanyu* fermented with *P. pentosaceus* achieved higher sensory scores for odors, texture, and overall acceptance. Volatile flavor analysis identified 31 compounds (10 alcohols, 7 aldehydes, 6 ketones, 4 esters, 1 acid, and 3 hydrocarbons). Compared with the natural fermentation group, the *P. pentosaceus*-inoculated group exhibited a higher total signal intensity of volatile compounds, with a notable increase in aldehydes and ketones. The results of the study could provide guidance for the use of *P. pentosaceus* as a starter culture to improve the quality of industrialized fermented fish products.

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Institutional Review Board Statement: This sensory study was exempt from formal ethical review in accordance with Article 32 of the Measures for Ethical Review of Life Science and Medical Research Involving Human Beings (issued by the National Health Commission of the People's Republic of China in 2023, NHC Science and Education Development [2023] No. 4). The study involved only normal food consumption by healthy adult volunteers, utilized anonymized data, posed no harm to human subjects, and did not involve sensitive personal information or commercial interests. This exemption is also consistent with the institutional policy of Guangdong Ocean University.

Informed Consent Statement: Written informed consent was obtained from all subjects involved in the sensory evaluation after they were fully informed of the study's purpose, procedures, and potential risks. All data were anonymized to protect participant privacy.

Data Availability Statement: The original contributions presented in this study are included in the article. Further inquiries can be directed to the corresponding author.

Conflicts of Interest: The authors declare no conflicts of interest.

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