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Research Article

Effect of High Hydrostatic Pressure and Thermal Processing on the Shelf Life and Quality Attributes of Apple-Kiwi-Carrot Puree Blend

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The effects of high hydrostatic pressure (HHP) (400 MPa/2 min and 500 MPa/2 min) and thermal processing (TP) (90°C/2 min) on the microorganisms, nutrition, color, endogenous enzyme activities, antioxidant capacity, and rheological properties of blended apple-kiwi-carrot puree were comprehensively evaluated after processing and during storage at 4°C. Results showed the microbiological shelf life of the HHP or TP products was at least 24 days. TP inactivated polyphenol oxidase activity more effectively, retaining more polyphenols in samples during storage, whereas HHP treatments were more beneficial for the preservation of ascorbic acid, total carotenoids, β-carotene, and antioxidant capacity. During storage, there was no significant difference in PME activity among all treated samples, but particle size distribution and viscoelasticity tests demonstrated that the 400 MPa treated samples were more stable in their textures. Moreover, the principal component analysis (PCA) intuitively revealed that the overall sensory and nutritional attributes of the HHP-treated samples were closer to the fresh product. These results demonstrated that HHP could be a better choice than TP for use in the production of high-quality puree blends.

1. Introduction

Nowadays, fruit and vegetable purees have been popular as ready-to-eat products on the market [1], especially for infants and children, which are considered healthy, nutritious, and convenient [2]. Moreover, the properly intake of fruits and vegetables could provide dietary fiber and phytochemicals and thus produce positive effects on preventing obesity and chronic disease [1, 3]. However, the fact is that most consumptive purees are made of concentrated juice, essences, pigments, and other additives, while few are fresh fruit or vegetable purees sterilized by classic thermal processing (TP), such as strawberry puree [4], apple puree [5], avocado puree [6], carrot puree [7], and tomato puree [8]. These mixed products do not conform to the trend of "clean label," which focuses on organic and natural raw materials without artificial additives/ingredients [9]. In addition, traditional TP treatments usually lead to deterioration of the color, texture, flavor, and nutritional values of food products [10–12].

High hydrostatic pressure (HHP) has been evaluated as a burgeoning nonthermal processing technique, in which the typical applied pressure operates from 200 to 800 MPa at a low temperature of 5 to 35°C [13]. Lots of studies have found that HHP does not break covalent bonds and has a limited effect on low molecular mass food compounds, such as polyphenols, vitamins, and flavor components [14, 15]. Thus, compared with classic TP, HHP could delay the deterioration of nutrition and sensory properties like color, flavor, taste, and texture of food products, especially for heat-sensitive materials like fresh fruit and vegetables [14]. The better retention of antioxidant activity, total phenol, anthocyanin, and ascorbic acid in strawberry and blackberry purees treated by HHP has also been confirmed

[16]. Also, HHP technology has been applied in many other products like strawberry puree, blueberry puree, tomato puree, and purple sweet potato nectar to prolong the shelf life and maintain the original sensory and nutritional properties [12, 17–19]. However, compared with a single component puree, a puree blend with different combinations of supplements can clearly provide more balanced nutrients and a more diversified flavor and taste [20]. Therefore, the application of HHP in puree blend products based on the concept of "clean label" displays a great market prospect.

The main objective of this study was to compare the impacts of HHP and classical TP treatments on the microorganisms, color parameters, phytochemicals, endogenous enzyme activities, antioxidant capacity, and rheological properties of the puree blends after processing and during refrigerated storage for 24 days.

2. Materials and Methods

2.1. Preparation of Samples. In this study, the apple (*Malus domestica* Borkh.) variety "Fuji," kiwifruit (*Actinidia chinensis* Planch.) variety "Chinensis," and carrot (*Daucus carota* L.) variety "Zhunong Qicun" were all purchased from local market in Beijing (China). According to the results of preliminary experiments, pieces of apples, kiwifruits, and carrots were mixed in a mass ratio of 6 : 2 : 1 at room temperature (25 ± 2°C). The prepared puree was quickly packed into PET bags (12 ∗ 10 cm) and vacuum sealed and then stored at 4°C before use.

2.2. Chemicals. 2,2-Diphenyl-1-picrylhydrazyl (DPPH), 2,4,6-tri-2-pyridyl-1,3,5-triazine (TPTZ), 6-hydroxy-2,5,7,8-tetramethyl-chroman-2-carboxylic acid (Trolox), and Folin–Ciocalteu reagent were purchased from Sigma-Aldrich (St. Louis, USA), nutrient agar and rose bengal agar were obtained from Beijing Solarbio Co. Ltd., (Beijing, China), and other chemicals were all provided by Beijing Chemicals Co. (Beijing, China).

2.3. Sample Treatments. According to Zhang et al. [21], the HHP-treated samples were placed in a HHP (CAU-30L, Baotou Kefa Co., Ltd., Inner Mondolia, China) treatment kettle, and the distilled water was used as pressure transfer media (25 ± 2°C). The pressurization rate was about 120 MPa/min and the depressurization was immediate (<3 s).

The TP treated samples were placed in a water bath and heated until the core temperature reached 90°C and kept for 2 min. All treated samples were quickly cooled in an ice bath, and the untreated samples were used as control. All prepared samples were stored at 4°C. During storage, samples were tested at 0, 3, 6, 9, 14, 19, and 24 days.

2.4. Microbial Analysis. To count viable microorganisms in samples, the total plate count method was followed according to the method described by Xu et al. with some modifications [22]. Decimal dilutions of 10 g samples were prepared in sterile 0.1% (w/w) peptone solution and then homogenized for 2 min. The colonies were counted after incubation for 48 ± 2 h at 37°C for viable total aerobic bacteria (TAB) and 72 ± 2 h at 27°C for viable yeasts and molds (Y&M).

2.5. Analysis of pH, Total Soluble Solids (TSSs), and Total Dietary Fiber (TDF). The pH and TSS values of the samples were measured with a Thermo Orion 868 pH meter (Thermo Fisher Scientific, Inc., Waltham, MA, USA) and a digital Abbe refractometer (DR-A1, ATGO. Co., Ltd., Tokyo, Japan) at 25°C, respectively.

The content of TDF was determined according to AOAC method of Prosky et al. [23] with some modifications. Dried samples were sequential enzymatic digested by heat stable α-amylase, amyloglycosidase, and protease to remove starch and protein. Then, they were precipitated by ethanol and filtered, and the residues were washed with ethanol and acetone. Thereafter, the samples were dried, which were the final total dietary fiber residues. Results were expressed as g/100 g of dry weight.

2.6. Ascorbic Acid (AA) Analysis. Ascorbic acid was measured by the method of West et al. [24] with some modifications. Weighed 10 g sample and made up to 100 mL with metaphosphoric acid solution, and then 4 g of kaolin was added to decolorize. After centrifuged at 11000 ×g for 15 min at 4°C, 10 mL of the supernatant was taken into a conical flask. The titration was carried out with a calibrated 2,6-dichloroindophenol solution until the solution turned pink and did not fade within 15 s, the blank test was also performed.

2.7. Instrumental Color Assessment. Color assessment was conducted according to Xu et al. [22] using a color measurement spectrophotometer (Hunter Lab Color Quest XE, Hunter Associates Laboratory, Inc., VA, USA) in the reflectance mode. Color was expressed in L^*, a^*, and b^* values. In addition, the total color difference (ΔE) was calculated using the following equation, where L_0^*, a_0^*, and b_0^* are the values for the control samples:

$$\Delta E = \left[\left(L^* - L_0^*\right)^2 + \left(a^* - a_0^*\right)^2 + \left(b^* - b_0^*\right)^2 \right]^{1/2}. \quad (1)$$

2.8. Determination of Carotenoid Content

2.8.1. Total Carotenoids. The determination of total carotenoids was followed according to the method described by Bunea et al. [25] with little modifications. Weighed 3 g sample and mixed with 100 mL of acetone (containing 0.1% BHT) and then extracted for 15 min using an ultrasonic equipment (SB-800DTD, NingBo Xinzhi Biological Technology Co., Ltd., Ningbo, China) under the condition of 60 W/30°C. Then, 100 mL of 10% (w/v) KOH-methanol solution was added into the extract solution and

saponified in a shake table at 25°C for 3 h. Thereafter, another 100 mL of petroleum ether was added, and the aqueous phase was repeatedly extracted as above description. Finally, the organic phases were combined, washed with water until neutral, and then filtered, and the filtrate was placed in a 250 mL round bottom flask and rotary evaporated to near dryness at 40 ± 2°C. The residue was dissolved in *n*-hexane and transferred into a 25 mL volumetric flask. The extract was measured for absorbance at a wavelength of 450 nm using *n*-hexane as a control.

2.8.2. β-Carotenoid. The β-carotenoid was determined using the HPLC analysis described by Sánchez–Moreno et al. [26]. The preparation of the β-carotenoid extract was basically the same as total carotenoid. For HPLC analysis, the C18 column was selected, in which the column length was 250 mm, the inner diameter was 4.6 mm, and the particle size was 5 μm. The mobile phase was prepared by chloroform, acetonitrile, and methanol in a ratio of 3 : 12 : 85, containing 0.4 g/L ascorbic acid, and filtered through a 0.45 μm membrane. The flow rate was 2.0 mL/min, the detection wavelength was 450 nm, the column temperature was 35 ± 1°C, and the injection volume was 20 μL.

2.9. Determination of Total Phenol. The total phenol content of the sample was determined by the Folin–Ciocalteu reagent method with slight modifications [21]. 10 g sample was weighed and mixed with 20 mL of anhydrous methanol, then extracted at 4°C for 30 min and centrifuged at 15,000 ×g/4°C for 10 min. Then, 0.1 mL of the supernatant was diluted to 0.4 mL and mixed with 2 mL of Forinol reagent diluted 10 times with ultrapure water. After kept at room temperature for 1 h in the dark environment, 1.8 mL of 7.5% Na_2CO_3 solution was added and reacted for 15 min. Finally, the absorbance at 765 nm was measured with a spectrophotometer (UV1800, Shimadzu Co., Ltd., Kyoto, Japan), and according to the calibration curve ($Y = 0.0111X + 0.0376$, $R^2 = 0.9939$), the results were expressed in terms of mg equivalent of gallic acid (GA) per 100 g of sample.

2.10. Enzyme Activity Assays: Polyphenol Oxidase (PPO) and Pectin Methylesterase (PME). For PPO enzyme, the spectrophotometric method of Tan and Harris [27] was referred and made some modifications. To obtain a crude extract solution of polyphenol oxidase (PPO), 5 g sample was mixed with 30 mL of phosphate buffer extract (0.2 mol/L, pH = 6.5) and then centrifuged at 10,000 ×g for 10 min at 4°C after kept at 4°C for 1 h. The abovementioned clear liquid was taken as crude PPO solution and mixed with equal volume of 0.07 mol/L catechol solution (prepared with 0.2 mol/L phosphate buffer with pH = 6.5). The absorbance value was immediately monitored at 420 nm for 5 min, and the slope of the straight-line part of the curve was used as the activity of PPO and the results were expressed as relative residual activity.

The activity assay of pectin methylesterase (PME) was performed as described by Kimball [28] with slight modifications. A total volume of 60 mL of pectin solution (0.1 mol/L NaCl) was taken into a beaker of a circulating water bath at 30 ± 2°C, then 0.03 mol/L NaOH was added with an 842 Titrando automatic potentiometric titrator (Metrohm Ltd, Herisau, Switzerland) until the pH reached 7.0. Then, 5.0 mL of crude PME solution was added. During the hydrolysis of the crude PME solution, the automatic potentiometric titrator was used to add 0.03 mol/L NaOH to maintain the pH at 7.5. Finally, the amount volume of 0.03 mol/L NaOH added within 30 min was recorded. The activity of PME was defined as the number of moles of acid produced per minute by PME at pH 7.5, and the results were expressed as relative residual activity.

2.11. Determination of Antioxidant Capacity

2.11.1. DPPH Radical Scavenging Activity. The method of Zhang et al. [21] was referred with minor modifications. Sample extract (100 μL) was pipetted and added into 4 mL of DPPH solution (0.14 mM). After incubation for 45 min at room temperature in the dark, the absorbance was measured at 517 nm. Meanwhile, 100 μL of methanol was used as blank control. The concentration of Trolox equivalent to the clearance obtained was calculated in mmol TE/kg according to the calibration curve ($Y = 0.7967X + 0.07$, $R^2 = 0.9923$).

2.11.2. Ferric-Reducing Antioxidant Power (FRAP). Sample extract (100 μL) was mixed with 4 mL of TPTZ working solution (prepared from 0.3 mol/L acetate buffer, which pH was 3.6, 10 mmol/L TPTZ solution and 20 mmol/L $FeCl_3$ mixed in a volume ratio of 10 : 1 : 1). After incubation at 37°C for 10 min, the absorbance at 593 nm was measured. The results were calculated according to the calibration curve ($Y = 1.159X - 0.0062$, $R^2 = 0.9944$) and expressed in Trolox equivalents in mmol TE/kg.

2.12. Particle Size Distribution. The particle size distribution of the sample was measured using a laser particle size analyzer (LS230, Beckman coulter, Inc., Florida, USA). A large particle size measurement mode was selected, and the scattered light intensities of the sample particles were measured under laser diffraction at a wavelength of 750 nm. The volume-weighted average particle size (D [4, 3]) in the result report was used for further analysis, which is widely used to characterize the shape and size of particles.

2.13. Rheological Properties. The sample was equilibrated at 25°C for 30 min and measured using a rheometer (AR1000, TA Instrument Co., Ltd., New Castle, DE, USA). All measurements were performed on a 40 mm diameter flat plate at 25°C. For the determination of dynamic rheological properties, the stress amplitude was fixed at 1.5 Pa, and the oscillation angle frequency was varied from 0.1 to 10 rad/s in the linear viscoelastic interval.

2.14. Statistical Analysis. All test results were expressed as mean ± S.D. To ensure the reliability of the experimental results, all experiments were operated with 3 biological and 2

technical replicates, except for a special explanation. The principal component analysis (PCA) was conducted using an online tool (http://www.metaboanalyst.ca) according to Yang et al. [29]. Data were analyzed with one-way analysis of variance and Tukey multiple comparison test, in which $p < 0.05$ showed significant difference.

3. Results and Discussion

3.1. Microbiological Analysis. The counts of TAB and Y&M in the HHP and TP treated samples during storage are shown in Table 1. The initial counts of TAB and Y&M in untreated samples were 4.45 and 3.16 $\log_{10}$ CFU/g, respectively. After treatment by HHP or TP, the counts of TAB and Y&M in all samples were kept lower than the detection limit (<1 CFU/g). During storage, there were no Y&M was detected in all samples. Furthermore, the counts of TAB in 90°C/2 min, 400 MPa/2 min, and 500 MPa/2 min treated samples were gradually increased until the 9th day and increased to 1.66 $\log_{10}$ CFU/g, 1.65 $\log_{10}$ CFU/g, and 1.58 $\log_{10}$ CFU/g, respectively. Researchers also found that HHP and TP treatments could availably inhibit the microorganism growth in a mixed juice blend during the refrigerated storage of 12 days [20]. As the storage time prolonged, the damaged (sublethal state) or viable but nonculturable bacteria might restored, which caused the increase of TAB in the samples [30]. Compared with TP treatment, HHP treatment led to obvious decreases in the pH, which was beneficial to the preservation of samples. Similar results also occurred in HHP treated Valencia orange juice [31]. The pH of all samples increased slightly during storage, which might be related to the degradation of organic acids such as ascorbic acid. However, since the pH was kept lower than 4.1, the microbial growth in the samples was effectively inhibited. At the end of storage, the count of TAB did not exceed 2.0 $\log_{10}$ CFU/g and no Y&M was detected, which suggested that HHP treatment could ensure the microbial safety of the samples at least for 24 days at 4°C.

3.2. TSS, TDF, Ascorbic Acid, Total Phenol, and Carotenoids. The effects of HHP and TP treatments on the phytochemicals of the samples during storage are shown in Figure 1. After treatment and during the whole storage, there were no obvious changes in TSS were found in all treated samples, which indicated that the HHP treatment could well maintain the content of TSS in the samples (Figure 1(a)). Similar results in TSS content of cantaloupe puree treated by 300–500 MPa/5 min during refrigerated storage were reported by Mukhopadhyay et al. [32].

At day 0, the TDF of TP treated samples showed a significant increase, but there were no significant changes in HHP-treated samples (Figure 1(b)). TP treatment could produce some Maillard products, which were also analyzed as lignin and thus increased the apparent fiber content and color deterioration of the samples. Considering that the PME activity was not efficiently inactivated, the changes in TDF content could be related to the solubilization and fragmentation of carbohydrate polymers associated to plant cell walls causing the transformation of the different forms of dietary fibers present in the materials [33].

According to Figure 1(c), at day 0, the AA content of samples decreased greatly from 0.82 to 0.55 mg/100 g after TP treatment but was well maintained to 0.83 mg/100 g and 0.75 mg/100 g after 400 MPa/2 min and 500 MPa/2 min treatments, respectively. During storage, the content of AA in all treated samples significantly decreased with the prolong of storage time. At the end of storage, the AA content decreased by 75.6%, 69.5%, and 71.9% in the TP, 400 MPa/2 min, and 500 MPa/2 min treated samples, respectively. In comparison, the HHP treatment seemed to preserve more and show better stability of AA than TP treatment. Moreover, HHP treatments (400~600 MPa/5~10 min) were better for AA retention at room or lower temperature compared with TP, and the AA content of food products after HHP treated generally showed a higher stability during cold storage [34, 35].

The changes in the total phenol content of the sample during storage were shown in Figure 1(d). At day 0, the total phenolic contents of treated samples were all significantly decreased than the untreated samples. Another study reported similar decreases after 450~550 MPa/5 min for tomato puree and 70°C/30 s treatment for tomato juice [18]. Furthermore, the total phenolic contents of the HHP treated samples were significantly lower than that of the TP treated ones, which was in accordance with the results of PPO for it is supposed to be the main enzyme responsible for phenol oxidation and degradation [36]. During storage, the total phenolic content of treated samples increased significantly, which was in agreeing with the result of HHP-treated strawberry syrup during 30 days [30]. Also, this result might be attributed to the enhanced permeability of the cells after HHP or TP treatment, which caused the slowly elute of phenolic substances from the cells and resulting in a measured increase in the total phenol content [16].

Figures 1(e) and 1(f) show the changes of total carotenoids and β-carotene content in the samples during the 24 days of storage. Both HHP and TP treatments resulted in no significant change in the content of total carotenoids and β-carotene in the samples. During storage, the content of total carotenoids and β-carotene in all treated samples displayed continuously decrease, but the HHP-treated samples decreased more slowly than the TP treated samples. At the end of storage, the total carotenoids and β-carotene content decreased by 28.7% and 39.7% in the TP-treated samples, by 25.9% and 36.6% in the 400 MPa/2 min treated samples, and by 24.6% and 35.2% in the 500 MPa/2 min treated samples, respectively. Clearly, compared with the TP, HHP performed better in the preservation of carotenoids, which was in accordance with the results of Andrés et al. [37]. They found similar less decreases in the content of β-carotene and total carotenoids in the HHP-treated mixed orange smoothies (450 MPa/3 min) during storage of 45 days, compared with the TP-treated samples (80°C/3 min).

3.3. Color Measurements. As shown in Table 2, at day 0, there were no significant differences of L^*, a^*, and b^* values between the treated and untreated samples. However, the ΔE

Table 1: Variations of total aerobic bacteria (TAB) and yeasts and molds (Y&M), and pH values in puree blends after processing and during storage.

	Storage time (d)	Control	TP	400 MPa/2 min	500 MPa/2 min
TAB ($\log_{10}$ CFU/g)	0	4.45 ± 0.45	ND	ND	ND
	3	—	ND	ND	ND
	6	—	ND	ND	ND
	9	—	ND	ND	ND
	14	—	1.22 ± 0.07^{a}	1.23 ± 0.21^{a}	1.19 ± 0.20^{a}
	19	—	1.45 ± 0.05^{a}	1.46 ± 0.15^{a}	1.44 ± 0.13^{a}
	24	—	1.66 ± 0.10^{a}	1.65 ± 0.08^{a}	1.58 ± 0.03^{a}
Y&M ($\log_{10}$ CFU/g)	0	3.16 ± 0.62	ND	ND	ND
	3–24	—	ND	ND	ND
pH	0	3.98 ± 0.01^{A}	3.99 ± 0.01^{Ab}	3.89 ± 0.02^{Bb}	3.88 ± 0.02^{Bb}
	3	—	3.92 ± 0.02^{d}	3.93 ± 0.03^{b}	3.89 ± 0.01^{b}
	6	—	3.95 ± 0.01^{c}	4.00 ± 0.02^{a}	3.94 ± 0.02^{b}
	9	—	3.91 ± 0.01^{d}	3.91 ± 0.01^{b}	3.90 ± 0.02^{b}
	14	—	4.03 ± 0.01^{a}	4.01 ± 0.01^{a}	4.02 ± 0.01^{a}
	19	—	3.95 ± 0.01^{c}	3.99 ± 0.01^{a}	4.04 ± 0.02^{a}
	24	—	4.07 ± 0.01^{a}	3.97 ± 0.02^{a}	4.08 ± 0.02^{a}

Note. "—," the samples were not tested due to spoilage; "ND," not detected (<1 CFU/g); all data were means ± SD, $n = 6$; values with different uppercase (lowercase) letters within one row (column) were significantly different ($p < 0.05$).

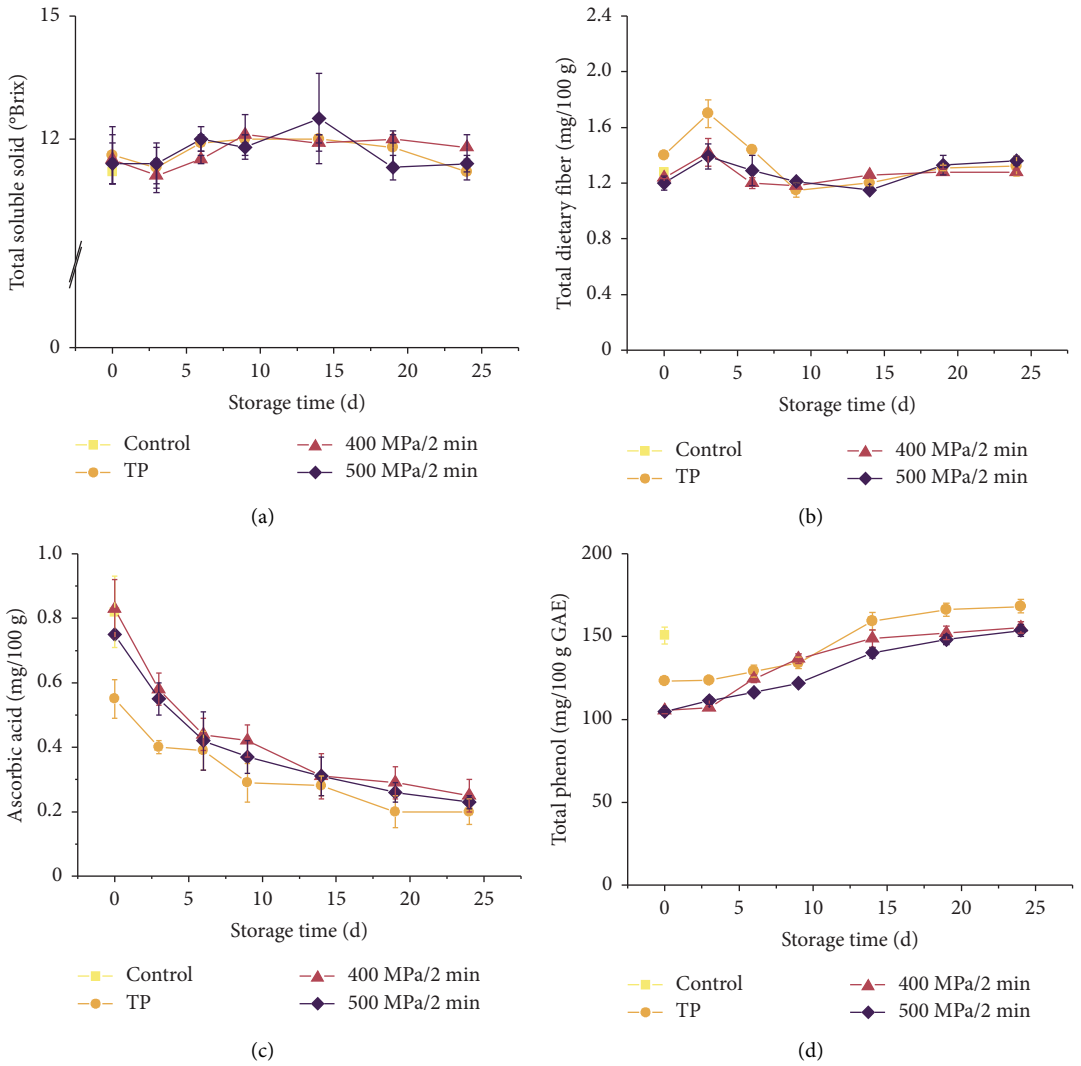

Figure 1: Continued.

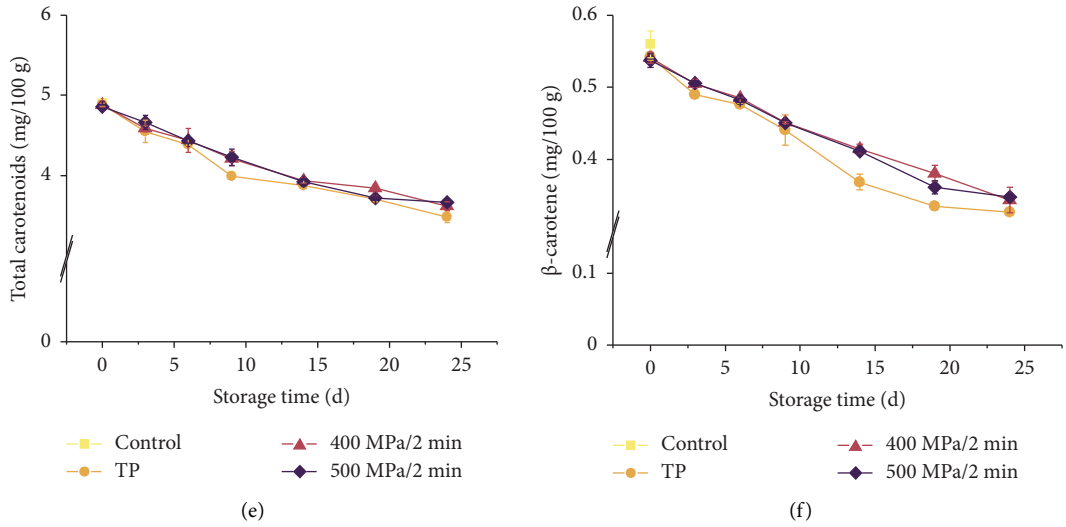

Figure 1: The content of total soluble solid (a), total dietary fiber (b), ascorbic acid (c), total phenol content (d), total carotenoids (e), and β-carotene (f) in puree blends after processing and during storage.

Table 2: The color parameters of puree blends after processing and during storage.

	Storage time (d)	Control	TP	400 MPa/2 min	500 MPa/2 min
L^*	0	39.46 ± 1.57^{A}	40.80 ± 1.77^{Ab}	38.96 ± 1.49^{Abc}	38.89 ± 0.24^{Abc}
	3	—	40.92 ± 1.34^{b}	38.80 ± 1.45^{bc}	39.12 ± 0.99^{bc}
	6	—	40.24 ± 1.82^{b}	39.63 ± 1.41^{b}	39.26 ± 1.50^{b}
	9	—	44.20 ± 1.94^{a}	44.36 ± 0.60^{a}	43.29 ± 0.32^{a}
	14	—	39.54 ± 0.24^{b}	38.62 ± 0.44^{bc}	37.47 ± 0.63^{c}
	19	—	40.75 ± 1.53^{b}	37.55 ± 0.83^{c}	38.31 ± 0.31^{bc}
	24	—	40.07 ± 0.40^{b}	39.47 ± 0.56^{bc}	39.82 ± 1.36^{b}
a^*	0	8.17 ± 0.69^{A}	9.05 ± 0.87^{Aa}	8.34 ± 0.76^{Aabc}	7.77 ± 0.20^{Abc}
	3	—	8.51 ± 0.68^{a}	8.03 ± 0.82^{bc}	8.38 ± 0.70^{ab}
	6	—	8.24 ± 0.97^{a}	8.59 ± 0.72^{ab}	8.00 ± 1.08^{abc}
	9	—	8.92 ± 1.92^{a}	9.31 ± 0.78^{a}	9.22 ± 0.36^{a}
	14	—	7.41 ± 0.41^{a}	8.02 ± 0.28^{bc}	7.01 ± 0.10^{c}
	19	—	8.86 ± 1.01^{a}	7.19 ± 0.67^{c}	7.71 ± 0.27^{bc}
	24	—	8.94 ± 0.15^{a}	8.38 ± 0.30^{abc}	8.85 ± 1.12^{ab}
b^*	0	16.03 ± 1.85^{A}	17.52 ± 2.45^{Aab}	15.26 ± 1.43^{Aa}	14.94 ± 0.28^{Aa}
	3	—	17.65 ± 1.18^{a}	16.10 ± 1.47^{a}	16.29 ± 0.92^{a}
	6	—	15.56 ± 2.26^{ab}	15.19 ± 1.71^{a}	14.11 ± 1.65^{ab}
	9	—	15.13 ± 3.92^{abc}	15.62 ± 2.16^{a}	15.65 ± 1.69^{a}
	14	—	12.76 ± 0.45^{c}	12.78 ± 0.32^{b}	11.84 ± 0.23^{b}
	19	—	14.64 ± 1.73^{abc}	11.92 ± 1.13^{b}	11.99 ± 0.63^{b}
	24	—	14.84 ± 0.26^{b}	14.44 ± 0.65^{a}	16.00 ± 1.94^{a}
ΔE	0	0	5.09 ± 0.66^{Aa}	2.50 ± 0.47^{Bbcd}	1.86 ± 2.01^{Bcd}
	3	—	2.30 ± 1.21^{bc}	0.76 ± 0.28^{d}	1.10 ± 0.20^{d}
	6	—	3.69 ± 2.37^{abc}	2.46 ± 0.63^{bcd}	3.21 ± 0.68^{abcd}
	9	—	5.76 ± 1.74^{a}	5.26 ± 1.21^{a}	5.09 ± 0.61^{a}
	14	—	3.53 ± 1.73^{abc}	3.45 ± 1.90^{abc}	4.78 ± 2.20^{ab}
	19	—	2.05 ± 0.26^{c}	4.65 ± 1.75^{ab}	4.33 ± 1.80^{abc}
	24	—	2.73 ± 1.09^{bc}	1.90 ± 1.28^{cd}	2.22 ± 1.34^{bcd}

Note. "—," the samples were not tested due to spoilage; all data were means ± SD, $n = 6$; values with different uppercase (lowercase) letters within one row (column) were significantly different ($p < 0.05$).

value of TP-treated sample was 5.09 ± 0.66, more than the critical value of 2.0 and significantly greater than the HHP-treated ones, which meant that the TP treatment induced more visible chromatic aberrations than the HHP treatments. The treatment of TP could cause browning reactions more easily and produce some Maillard products [38], which

could explain why the higher ΔE value appeared in the TP-treated samples and was consistent with the above-mentioned results of TDF. During storage, the L^*, a^*, and b^* values of all treated samples showed fluctuations, but there were no significant differences within the group. As for the ΔE value, all treated samples showed significant increases at day 9 but no significant changes during the whole 24 days of storage. The increase of ΔE value at day 9 may be due to the significant increase of L^* value; Wang et al. [39] found a similar relevance when the purple sweet potato nectar was treated by HHP and stored at 4°C or 25°C. In conclusion, the HHP-treated samples exhibited better color stability than the TP-treated ones.

3.4. PPO and PME Activity. The effects of HPP and TP treatments on the activity of PPO and PME in the samples were shown in Figures 2(a) and 2(b). After treated at 400 MPa/2 min or 500 MPa/2 min, the PPO activity in the samples decreased to 64.54% and 56.57%, respectively, while that of the TP samples decreased to 38.05% (Figure 2(a)). HHP treatment could only do damage to the partial secondary bonds inside the protein, such as hydrogen bonds and hydrophobic bonds, and researchers already reported the pressure resistance of PPO at low temperature [40]. However, due to heat effects, thermal processing always exhibited better inhibition of enzymes in puree [38]. During the storage, the PPO activity of the treated samples decreased significantly. At day 24, the PPO activities of the 400 MPa/2 min and 500 MPa/2 min treated samples exhibited a decrease to 16.66% and 13.46%, respectively, while that of the TP samples decreased to 6.81%. These results meant that the HHP inhibited less PPO activity in the samples than TP treatment, which could explain the lower content of total phenol contents in HHP treated samples during storage.

At day 0, compared with the untreated sample, the activity of PME in the TP and 400 MPa/2 min treated samples reduced significantly, while showing no differences in the 500 MPa/2 min treated samples (Figure 2(b)). During storage, the PME activities in all treated samples were gradually reactivated in the initial 6 days of storage and reached the highest values, thereafter, decreased again, but there was no significant difference between HHP- and TP-treated samples. The PME activity in Valencia orange juice treated at 600 MPa/1 min showed similar results during 12 weeks of storage at 4°C which attributed to the variation content of pulp in the samples [31].

3.5. Antioxidant Capacity. As shown in Figures 2(c) and 2(d), the DPPH removal ability of all treated samples occurred with slight decreases compared with the untreated samples at day 0, but there were no obvious differences among the HHP- and TP-treated samples. Moreover, 500 MPa/2 min treated samples demonstrated higher FRAP reduction ability than TP or 400 MPa/2 min treated ones and displayed no significant difference in comparison with untreated samples. During 24 days of storage, the antioxidant capacity of all treated samples decreased slowly, and the FRAP antioxidant capacity of TP-treated samples was significantly lower than HPP-treated purees. The decrease in total antioxidant capacity may be the result of a combination of a reduction in total phenolic, ascorbic acid, and carotene levels in the samples. It has been proven that the DPPH removal capacity and FRAP reduction ability of fruit and vegetable products are highly correlated with the content of ascorbic acid and total phenol [37, 39, 41], while carotenoids are usually considered with strong antioxidant capacity.

3.6. Particle Size Distribution. In view of the particle diameters of all samples were distributed in a large range of values, the volume mean diameters of D [4, 3] values were chosen for further analyzation as shown in Table 3. It could be seen that the D [4, 3] value of samples after treated decreased at day 0, and HHP treated samples showed less decrease compared with TP-treated ones. Due to the ineffective inhibition of PME, the decrease of particle diameter after processed might be caused by the depolymerization and degradation of the long molecular chains of the original pectin [42]. The particle diameters of all treated samples manifested volatility within a certain range during refrigerated storage, but the samples treated under 400 MPa/2 min seemed to be more stable. Similar better stability was obtained when treated with banana puree at 500 MPa/10 min compared to TP treatment at 90°C/2 min during 30 days of refrigerated storage [43].

3.7. Rheology Properties. As shown in Figure 3, at day 0, the G' and G'' values of samples decreased after processed at 400 MPa/2 min or TP but increased in the 500 MPa/2 min treated samples, compared with the untreated samples. The viscosity decrease of samples might be related to the particle size reduce in results of D [4, 3] for the larger and irregular particle always causing a higher hindrance to flow [44], and the increase could be attributed to the compacting effects, or protein-tissue coagulation under HHP [45]. During the refrigeration period, the samples treated at TP or 500 MPa/2 min reached the highest G′ and G″ values on the 3rd day of storage (Figures 3(b) and 3(d)), while 400 MPa/2 min treatment delayed this kind of change and reached the peak values on the 6th day (Figure 3(c)). These results were basically in accordance with the results of PSD and D [4, 3]. Since the PME was not effectively inhibited or even activated, the produced soluble pectin could bind Ca^{2+} ions to form water-retaining gelatinous structures [45], so the overall viscoelasticity of the system was enhanced. Subsequently, the residual PME activity causes the continuous degradation of macromolecules such as pectin and polysaccharides, eventually leading to a decrease in G' and G'' values again.

3.8. Correlation and Principal Component Analysis (PCA) Analysis. The linear dependence among antioxidant compounds, endogenous enzyme activity, and antioxidant capacity of TP or HHP treated samples are shown in Table 4. The content of AA, total carotenoids, and β-carotene all showed an extremely significant correlation ($p < 0.001$) with

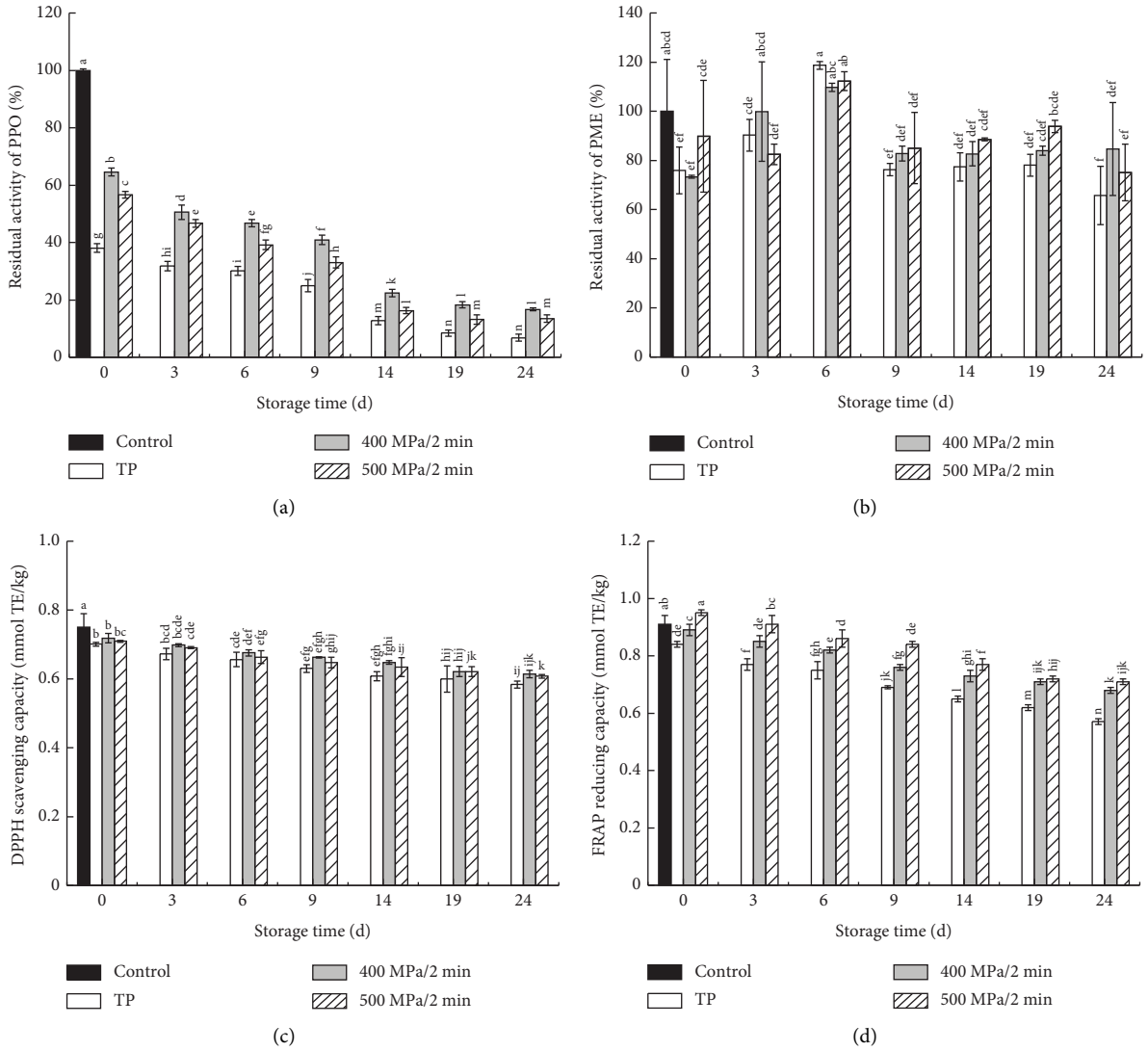

Figure 2: The antioxidant capacity of DPPH (a) and FRAP (b) and the relative residual activity of PPO (c) and PME (d) of puree blends after processing and during storage.

Table 3: The particle parameters of D [4, 3] values of the puree blends after processing and during storage.

	Storage time (d)	Control	TP	400 MPa/2 min	500 MPa/2 min
D [4, 3]	0	906.93 ± 19.56^{A}	813.70 ± 17.73^{Cb}	828.40 ± 9.02^{BCc}	851.50 ± 10.60^{Bb}
	3	—	846.20 ± 6.24^{a}	851.20 ± 15.65^{a}	850.20 ± 2.75^{b}
	6	—	815.23 ± 15.17^{b}	847.93 ± 10.64^{ab}	852.30 ± 17.27^{b}
	9	—	783.43 ± 9.30^{c}	826.70 ± 17.94^{c}	793.93 ± 7.81^{c}
	19	—	830.77 ± 2.76^{ab}	831.90 ± 4.00^{c}	907.10 ± 4.98^{a}
	24	—	839.37 ± 3.32^{a}	817.07 ± 5.96^{c}	841.40 ± 12.57^{b}

Note. "—," the samples were not tested due to spoilage; all data were means ± SD, $n = 6$; values with different uppercase (lowercase) letters within one row (column) were significantly different ($p < 0.05$).

the FRAP reduction ability of samples. None of the four phytochemicals displayed a linear correlation with DPPH removal capacity because the DPPH method was sensitive to all antioxidant components and thus affected by the opposite changes in total phenolic content during storage. Total phenolic content was significantly negatively correlated with PPO enzyme activity and all phytochemicals showed significant correlation with color parameters, which was consistent with the existing conclusions. These results indicated that the retention of phytochemicals after processing and during refrigeration greatly affects the preservation of the original color and antioxidant capacity of the sample.

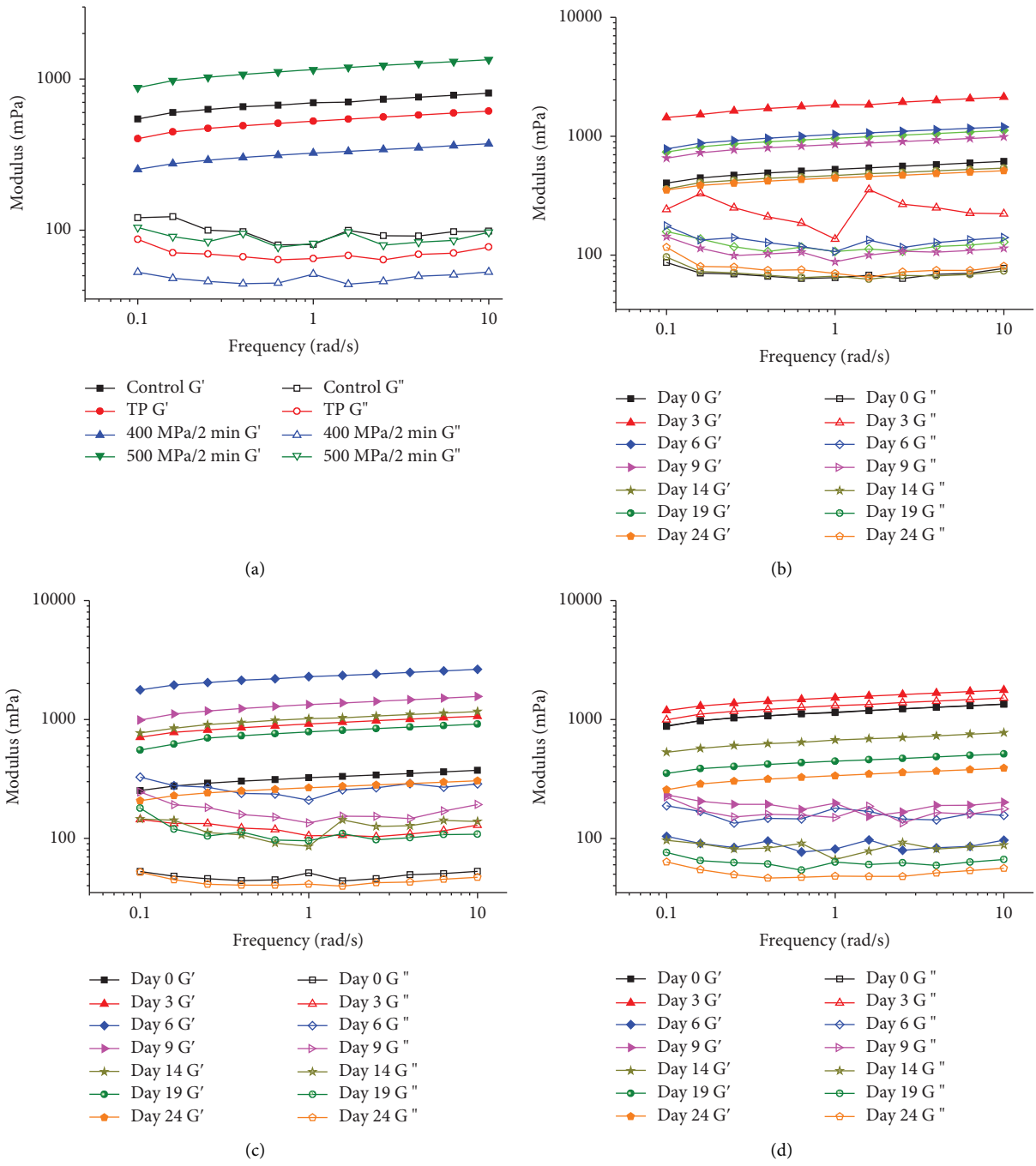

Figure 3: The storage modulus (G') and loss modulus (G'') of puree blends after processing and during storage ((a) samples at day 0; (b) 90°C/2 min; (c) 400 MPa/2 min; (d) 500 MPa/2 min).

The quality parameters are used for PCA analysis to better understand the comprehensive impact of HHP and TP treatment on samples. The projected canonical plots of composite puree samples with various treatments are illustrated in Figure 4. The intercorrelation between the control and HPP treated samples was observed, whereas the traditional thermal treated samples were highly discriminated from them (Figure 4). The first two principal components clearly explained the variation and cumulatively accounted for 98.7% of the total variance. In the Y-axis direction, the control sample was completely separated from the TP sample, while there was an intersection with the HHP sample. Moreover, the HHP sample overlapped with the TP sample in the X-axis direction. This indicated that although HHP samples displayed some similarities with TP samples; overall, their attributes were still much closer to those of untreated samples.

Table 4: Correlation analysis among phytochemicals, color parameters, antioxidant capacity, and endogenous enzyme activity of puree blends subjected to high hydrostatic pressure (HHP) or thermal processing (TP).

	AA	TPC	Total carotenoids	β-carotene	L^*	a^*	b^*	DPPH	FRAP	PPO	PME
AA	1	−0.64***	0.86***	0.85***	0.47***	0.36**	−0.44***	−0.06	0.89***	0.93***	0.27*
TPC		1	−0.60***	−0.61***	−0.35**	−0.30*	0.43***	0.08	−0.67***	−0.69***	−0.11
Total carotenoids			1	0.98***	0.57***	0.56***	−0.53***	0.03	0.88***	0.91***	0.28*
β-carotene				1	0.50***	0.53***	−0.60***	0.05	0.88***	0.92***	0.30*
L^*					1	0.66***	0.28*	−0.08	0.34**	0.38**	−0.20
a^*						1	−0.06	0.36**	0.31*	0.39**	0.22
b^*							1	−0.05	−0.58***	−0.65***	−0.37**
DPPH								1	0.08	−0.11	0.34**
FRAP									1	0.89***	0.33**
PPO										1	0.27*
PME											1

Note:"*," "**"and "***" means significant at $p \leq 0.05$, $p \leq 0.01$, and $p \leq 0.001$, respectively.

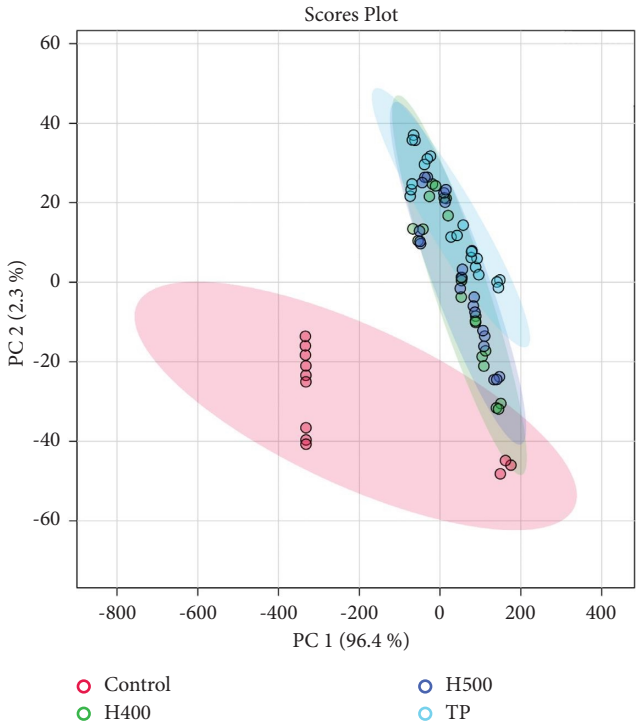

Figure 4: Principal component analysis (PCA) between high hydrostatic pressure (HHP) treatments and thermal processing (TP) of puree blends (TP: 90°C/2 min; H400: 400 MPa/2 min; H500: 500 MPa/2 min).

4. Conclusion

HHP and TP treatment could both ensure the microbiological safety of the apple-kiwi-carrot puree blend for at least 24 days when stored at 4°C. Compared with the TP treatments, the HHP treatments were more beneficial to the preservation of AA, carotenoids, and antioxidant capacity of samples. Moreover, the particle diameters of samples treated under 400 MPa/2 min seemed to be more stable. These results revealed that HHP treatment could preserve more of the original quality of the samples, while there was a significant difference in the overall sensory and nutritional properties between TP and control samples. In summary, the HHP technology could be a good alternative to apply in the puree blend and showed a vast potential for guaranteeing the high quality and nutritive value of fresh materials. Also, HHP might perform better if other technologies, such as natural inhibitor addition, were combined to inactivate the activity of endogenous enzymes for actual commercial applications in the future.

Data Availability

The datasets used to support the findings of this study are available from the corresponding author upon reasonable request.

Conflicts of Interest

The authors declare that they have no known competing financial interests.

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Supplementary Materials

The data related to sensory evaluation of the puree blends of different compositions (Table S1) and the particle size distribution (PSD) of puree blends after processing and storage (Figure S1) are presented in the supplementary file (Supplementary Materials). (*Supplementary Materials*)

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