



Effects of Ultrasonic Treatment on Biologically Active Ingredients of Perilla Drink

NGUYEN TRI YEN CHI^{1*}, NHAT ANH DUONG^{1,2},
NGOC NHU Y HUYNH¹ and BA NHAT TAN NGUYEN²

¹Faculty of Applied Biological Sciences, Department of Biological Technology,
Vinh Long University of Technology Education, Vinh Long, Vietnam.

²Department of Traditional Medicine and Pharmacy, Vinh Long
General Hospital, Vinh Long, Vietnam.

Abstract

Ultrasound can be applied as an efficient and green technology for various food processes. This study focused on evaluating the influence of ultrasonication and thermal treatments on the stability of antioxidant components in perilla drinks. Specifically, it compared the impact of ultrasonic treatments lasting 5, 10, and 15 minutes at frequencies of 58 kHz, 132 kHz, and 192 kHz with thermal treatment ranging from 70°C to 90°C, aiming to explore the effects of ultrasonic treatment on biologically active compounds in perilla drink, including polyphenols, flavonoids, anthocyanins, reducing sugars, DPPH, total soluble solids (TSS), and pH. Findings revealed no significant differences in TSS and pH among the sonicated, heat-treated, and untreated samples. However, the increase in flavonoid content under ultrasound and heat treatment for 10 minutes was statistically significant compared to the control sample ($p < 0.05$). In addition, the experimental results indicate that as ultrasonic treatment time and temperature increased, the reduced sugar and polyphenol content also increased. However, the ability to capture DPPH free radicals decreased with increasing ultrasonic treatment time and temperature ($p < 0.05$). Perilla drink pasteurized using ultrasonic treatment at 132 kHz for 10 minutes maintains anthocyanin, flavonoid, polyphenol, and reducing sugar content, as well as free radical scavenging ability (DPPH), at levels equivalent to the control (no treatment) and remains the most stable among all treatments. These insights are valuable for beverage producers aiming to preserve product quality during processing and storage.



Article History

Received: 06 March 2025

Accepted: 03 May 2025

Keywords

Anthocyanin;
Antioxidant Activity;
Non-Thermal;
Perilla Juice;
Ultrasound.

CONTACT Nguyen Tri Yen Chi ✉ chinty@vlute.edu.vn 📍 Faculty of Applied Biological Sciences, Department of Biological Technology, Vinh Long University of Technology Education, Vinh Long, Vietnam.



© 2025 The Author(s). Published by Enviro Research Publishers.

This is an  Open Access article licensed under a Creative Commons license: Attribution 4.0 International (CC-BY).

Doi: <https://dx.doi.org/10.12944/CRNFSJ.13.2.34>

Introduction

Perilla frutescens (L.) Britton, commonly known as Shiso in Japan, is a purple mint plant from the Lamiaceae family, believed to have originated in the mountainous regions of East Asia. This plant is widely cultivated across Asia, particularly in countries such as China, India, Japan, Korea, Thailand, and Vietnam. *Perilla* serves as a valuable source of biologically active antioxidant compounds, including polyphenols, flavonoids, and anthocyanins.¹ Contemporary biological and pharmacological studies on *Perilla frutescens* have demonstrated its wide range of biological effects, such as anti-inflammatory, antibacterial, antifungal, and antidepressant properties.² These antioxidants are linked to numerous health advantages, including cardiovascular support, diabetes management, cancer prevention, reduced risk of atherosclerosis, and the slowing of age-related degeneration.³

Drinks from fruits and vegetables have high water content and are rich in nutrients, providing a good environment for enzymes and microorganisms to grow. To prolong the preservation time of fruit and vegetable drinks, it is necessary to kill microorganisms and deactivate the enzyme system in fruit and vegetable juice. The oldest and most popular method is long-term heat treatment. However, this method often reduces the quality of fruit and vegetable juices, causes loss of vitamins and biologically active substances, and changes the flavor and color of the product.⁴ Nowadays, customer awareness of product quality is increasingly high. Consumers require the quality of canned fruit and vegetable juice products to have natural flavors, and colors, and preserve natural chemical components and biologically active ingredients.⁵ Therefore, modern pasteurization technology is applied to producing bottled fruit and vegetable juices to meet the above requirements. Ultrasound is used in many applications, including food processing and food analysis.⁶ Ultrasonic processing is one of the non-temperature processing technologies that replaces heat pasteurization. Ultrasonic processing mechanisms are simple, have no chemical residues, are highly safe, have low energy consumption, are environmentally safe, and cause little deterioration in product quality.⁷ Ultrasound treatment reduces the population of pathogenic microorganisms in fruit juice, retaining biologically active compounds of fruit juice such as antioxidants, vitamin C,

polysaccharides, pectin, total phenol content, and enzyme activity of juice. This technology has been tested for red grape juice treatment,⁸ mango juice,⁹ grapefruit juice,¹⁰ orange juice,¹¹ cranberry juice,¹² sugarcane juice,¹³ lily juice,¹⁴ kiwi juice,¹⁵ and different fruit and vegetable juices.¹⁶ *Perilla* drink has many anthocyanins are colored valuable biocompounds, However, anthocyanins have a major disadvantage, namely their low stability. Temperature is a critical parameter of food industrial processing that impacts on the food matrix, particularly affecting heat-sensitive compounds such as anthocyanins.¹⁷ However, there have been no studies examining the effects of ultrasound on *perilla* drink. Therefore, this study aims to investigate the impact of ultrasonic treatment on *perilla* drink, providing a theoretical foundation for its use in the processing and production of *perilla* beverages.

Materials and Methods

Beverages Samples

Perilla drink made with *Perilla frutescens* (L.) extract and *Chrysanthemum indicum* (L.) flowers according to research by Chi *et al.*¹⁸ The experiment was conducted to investigate pasteurization time (5 minutes, 10 minutes, 15 minutes) at temperature (70-90°C) in water baths (Model: WNB7, Memmert, Germany) and ultrasonic wave treatment at frequencies of 58 kHz, 132 kHz, 192 kHz for 10 minutes, 20 minutes, 30 minutes in the ultrasonic cleaning tank (Model: 4-UDS-2618-24, CREST ULTRASONICS CORP., New Jersey, USA) compared with the control sample without heat and ultrasound treatment.

Chemicals and Reagents

DPPH (2,2-diphenyl-1-picrylhydrazyl; TCI, Japan), hydrochloric acid (HCl; Merck, Germany), ethanol (C₂H₅OH; Merck, Germany), quercetin (Merck, Germany), Folin-Ciocalteu reagent (Merck, Germany), gallic acid (C₇H₆O₅; Sigma, USA), 3,5-dinitrosalicylic acid (C₇H₄N₂O₇; Fisher, USA), sodium carbonate (Na₂CO₃; Xilong Scientific, China), potassium sodium tartrate (C₄H₄O₆KNa; Xilong Scientific, China), sodium hydroxide (NaOH; Xilong Scientific, China), glucose (C₆H₁₂O₆; Xilong Scientific, China), potassium chloride (KCl; Xilong Scientific, China), sodium acetate trihydrate (CH₃COONa·3H₂O; Xilong Scientific, China), sodium nitrite (NaNO₂; Xilong Scientific, China), aluminum chloride (AlCl₃; Xilong Scientific, China), methanol

(CH₄O; Xilong Scientific, China), and ascorbic acid (C₆H₈O₆; Xilong Scientific, China).

Determination of Total Polyphenol Content (TPC)

The total polyphenol content (TPC) was determined using the Folin-Ciocalteu method.¹⁹ Initially, 0.2 mL of diluted sample solution, add 0.2 mL of distilled water, then add 0.5 mL of Folin-Ciocalteu 10%, shake well for 3-5 minutes, 0.4 mL of 7.5% sodium carbonate (Na₂CO₃) solution was added. The mixture was incubated in the dark at room temperature for 30 minutes. The absorbance was then measured at 765 nm using a UV/Vis spectrophotometer (UV/Vis 200 nm–830 nm, Eppendorf BioSpectrometer® Basic, Germany). Gallic acid was used as the standard, with concentrations of 0, 0.02, 0.04, 0.06, 0.08, and 0.1 mg/mL. A calibration curve was constructed by plotting absorbance values against gallic acid concentrations.

Determination of Reducing Sugars

The reducing sugar content was determined using the method described by Miller.²⁰ Initially, 2 mL of diluted sample solution mixed with 2 mL of DNS reagent (3,5 dinitrosalicylic acid) then boil in a water bath for 5 minutes to bring to room temperature. The reducing sugar content was measured at wavelength $\lambda = 540$ nm and glucose was used as the standard curve for the concentrations: 0, 0.1, 0.2, 0.3, 0.4, 0.5 mg/mL.

Determination of Total Anthocyanin (TAC)

The total anthocyanin content (TAC) was determined using the UV-Visible spectroscopy method outlined by Giusti *et al.*,²¹ based on the principle that anthocyanin colorants change with pH. The colored oxonium form predominates at pH 1.0 and the colorless hemiketal form at pH 4.5.

Determination of Antioxidant Capacity (DPPH)

The antioxidant capacity (DPPH) was determined using the method developed by Brand-Williams *et al.*²² Prepare a 0.004% DPPH solution in methanol. Initially, 0.6 mL of diluted sample solution into a test tube, add 0.8 mL of 0.004% DPPH and incubated for 30 min in the dark covered with aluminum foil. Then measure the absorbance of the solution at wavelength $\lambda = 517$ nm. The activity was calculated using the following formula:

DPPH radical scavenging activity (%) = $(A_0 - A_1)/A_0 \times 100$

where: where A_0 is an absorbance of the blank (without sample extract), and A_1 is an absorbance of sample extract.

Determination of Total Flavonoid Content (TFC)

The total flavonoid content was determined using the method described by Zhishen *et al.*²³ Initially, 0.5 mL of the appropriately diluted sample was transferred into a test tube, add 3.2 mL of distilled water, then add 0.15 mL of 5% NaNO₂, shake well for 5 minutes, then add 0.15 mL of 10% AlCl₃ and shake well for 5 minutes. Finally, add 1 mL of 1 M NaOH. Then measure the absorbance of the solution at wavelength $\lambda = 510$ nm. The total flavonoid content was quantified using a quercetin calibration curve, which was constructed using standard solutions with concentrations of 0, 0.1, 0.2, 0.3, 0.4, and 0.5 mg/mL. These standards were subjected to the same procedure as the samples, and a standard curve was generated by plotting absorbance against quercetin concentration.

Determination of Total Soluble Solids (TSS) and pH Values

The pH of the samples was measured at room temperature using a pH meter (inoLab pH 7110, WTW, Germany) by directly immersing the electrode into the sample. Total soluble solids (TSS) were determined using a refractometer (ALLA, France) under the same conditions.

Statistical Analysis

All experiments were conducted in triplicate to generate data for statistical evaluation. Statistical analyses were carried out using the Statgraphics software (Statgraphics Centurion XV, Statgraphics Technologies, Inc., Old Tavern Rd, The Plains, VA 20198, USA). Results are presented as mean values \pm standard deviation (SD). Differences among independent variables were assessed using parametric analysis of variance (ANOVA), followed by mean comparisons based on the least significant difference (LSD) test at a significance level of $p < 0.05$.

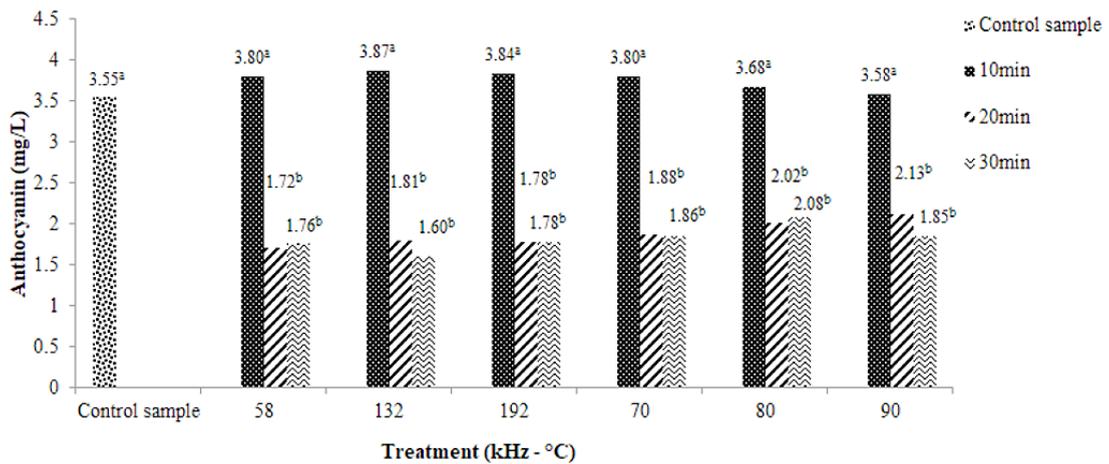
Results

Effect of Ultrasonic Pasteurization Method on Anthocyanin Content

Figure 1 illustrates the impact of ultrasonic pasteurization on the total anthocyanin content.

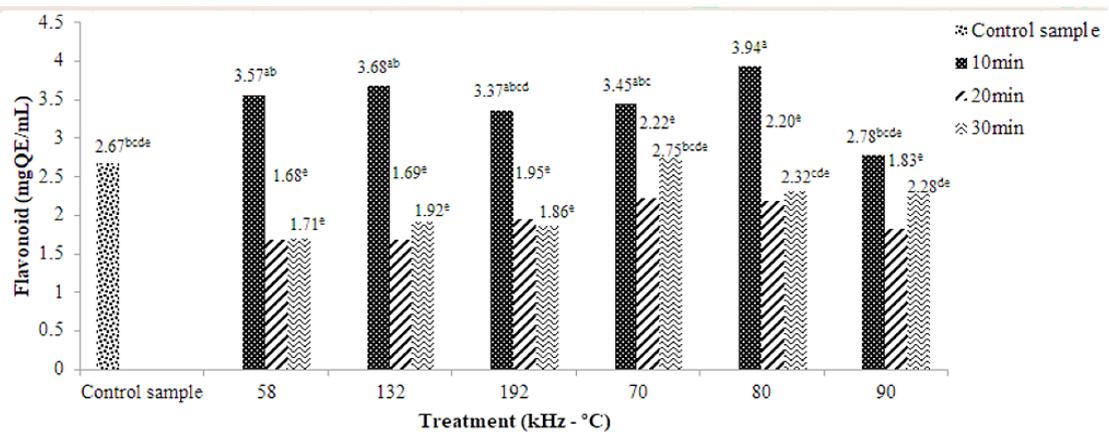
Figure 1 shows that the anthocyanin content when treated with heat and ultrasound for 10 minutes both increased higher than that of the sample control.

Ultrasound treatment at 3 frequencies 58 kHz, 132 kHz, and 192 kHz are 3.80 mg/L, 3.87 mg/L, and 3.84 mg/L respectively, and at temperatures, 70°C, 80°C and 90°C are 3.80 mg/L respectively. 3.68 mg/L and 3.58 mg/L compared to 3.55 mg/L (the sample control). However, the ultrasonic treatment method gave a higher anthocyanin content than the heat treatment and there was no statistically significant difference ($p > 0.05$).



Different letters in the same column indicate statistically significant differences ($p < 0.05$) by LSD test

Fig. 1: Effects of ultrasonic treatment on anthocyanin of perilla drink.



Different letters in the same column indicate statistically significant differences ($p < 0.05$) by LSD test

Fig. 2: Effects of ultrasonic treatment on flavonoids of perilla drink.

Effect of Ultrasonic Pasteurization Method on Flavonoid Content

Figure 2 illustrates the impact of ultrasonic pasteurization on the total flavonoid content.

Flavonoids are essential natural bioactive compounds known for their antioxidant properties. Interestingly, the flavonoid content increased significantly as the ultrasound process progressed. The increase

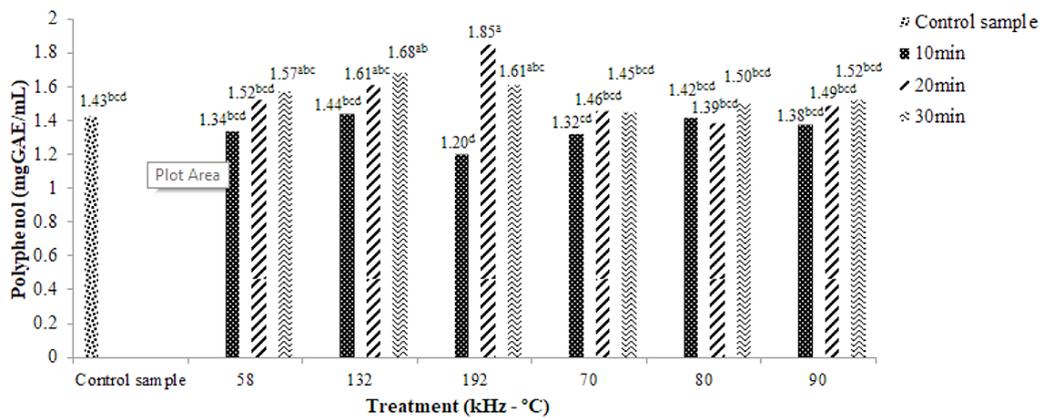
in flavonoid content when treated with ultrasound and heat for 10 minutes was statistically different compared to the control sample. Sample treatment at frequencies of 58 kHz, 132 kHz, 192 kHz and temperatures of 70°C, 80°C, 90°C were 3.57 mgQE/mL, 3.68 mgQE/mL, 3.37 mgQE/mL, 3.45 mgQE/mL, 3.94 mgQE/mL, 2.78 mgQE/mL compared to 2.67 mgQE/mL (control sample).

Effect of Ultrasonic Pasteurization Method on Polyphenol Content

Figure 3 illustrates the impact of ultrasonic pasteurization on the total polyphenol content.

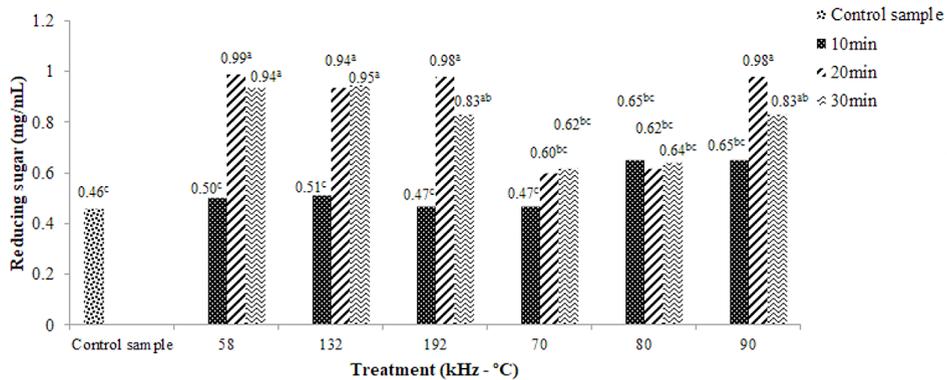
the polyphenol content when ultrasonic treated for 10 minutes at a frequency of 192 kHz (1.20±0.29 mgGAE/mL) is the lowest compared to the control

sample (1.43±0.18 mgGAE/mL) in ultrasonic and temperature samples treatment this difference is statistically significant. In addition, the survey results also showed that as the ultrasonic treatment time increased, the polyphenol content increased. Polyphenol content when treated for 10 minutes at 3 different frequencies is 1.34 mgGAE/mL (58 kHz), 1.44 mgGAE/mL (132kHz), and 1.43 mgGAE/mL (192kHz), respectively, lower than when increasing the time process up to 20 minutes and 30 minutes at the same frequency. When treating perilla water with ultrasound at a frequency of 58 kHz for 20 minutes, the polyphenol content increased to 1.52 mgGAE/mL, when treated for 30 minutes it was 1.57 mgGAE/mL. At frequencies of 132 kHz and 192 kHz, similar results were recorded.



Different letters in the same column indicate statistically significant differences (p < 0.05) by LSD test

Fig. 3: Effects of ultrasonic treatment on polyphenols of perilla drink



sonic treatment on reducing sugar of perilla drink.

Different letters in the same column indicate statistically significant differences (p < 0.05) by LSD test

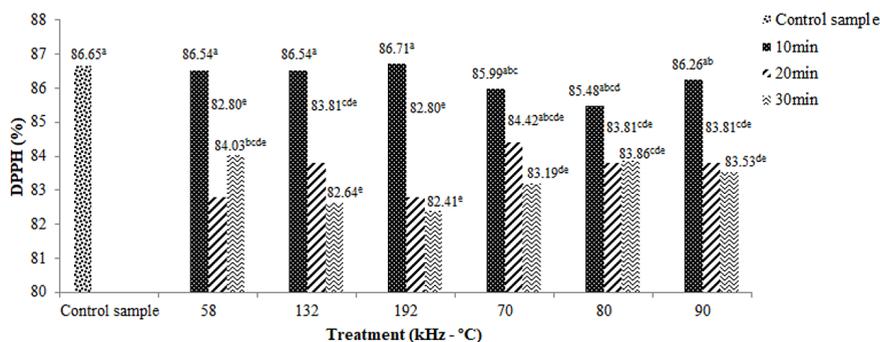
Fig. 4: Effects of ult

Effect of Ultrasonic Pasteurization Method on Reducing Sugar Content

Figure 4 illustrates the impact of ultrasonic pasteurization on the total reducing sugar content.

Reducing sugar content when treated with heat and ultrasound for 10 minutes at frequencies of 58 kHz, 132 kHz, 192 kHz (0.50 mg/mL, 0.51 mg/mL and

0.47 mg/mL) and at temperatures of 70°C, 80°C and 90°C (0.47 mg/mL, 0.65 mg/mL and 0.65 mg/mL) were not statistically different from the untreated control sample (0.46 mg/mL). In addition, the survey results also show that when increasing the ultrasonic treatment time and temperature, the reduced sugar content increases.



Different letters in the same column indicate statistically significant differences ($p < 0.05$) by LSD test

Fig. 5: Effects of ultrasonic treatment on the antioxidant capacity of perilla drink

Effect of Ultrasonic Pasteurization Method on Antioxidant Capacity (Dpph)

Figure 5 illustrates the impact of ultrasonic pasteurization on antioxidant capacity (DPPH) content.

The ability to capture DPPH free radicals when treated with ultrasound recorded at frequencies of 58 kHz, 132 kHz, 192 kHz (86.54%, 86.54%, 86.71%) and temperatures of 70°C, 80°C, 90°C were 85.99%, 85.48%, 86.26% and no statistically significant difference with the control sample (86.65%) at 10 minutes. In addition, the survey results also showed that when increasing ultrasonic treatment time and temperature, the ability to capture DPPH free radicals decreased. The ability to capture DPPH free radicals when treated for 10 minutes at different frequencies is 86.54% (58kHz), 86.54% (132kHz), and 86.71% (192kHz), respectively, higher than when the treatment time is increased 20 minutes and 30 minutes at the same frequency. When treating perilla water with ultrasound waves at 58kHz for 20 minutes, the ability to capture DPPH free radicals decreased to 82.8%, and when treated for 30 minutes, it was 84.03%. At frequencies of 132kHz and 192kHz, similar results were recorded. DPPH free radical scavenging ability recorded at 132kHz frequency was 83.81% (20 minutes) and

82.64% (30 minutes). At the frequency of 192kHz, the ability to capture DPPH free radicals recorded after 20 minutes of treatment was 82.8%, and after 30 minutes was 82.41%.

Effect of Ultrasonic Pasteurization Method on pH Value and TSS

Table 1 presents the variations in pH and total soluble solids (TSS) resulting from heat and ultrasound treatments.

Discussion

In addition, the survey results also showed that when increasing the ultrasonic and heat treatment time, the anthocyanin content decreased.²⁴ Anthocyanin content when treated for 10 minutes at 3 different frequencies is 3.80 mg/L (frequency 58 kHz), 3.87 mg/L (frequency 132 kHz), and 3.84 mg/L (frequency 192 kHz), respectively twice as high as when increasing the processing time to 20 minutes and 30 minutes at the same frequency. The reason for the above phenomenon is that when the ultrasonic treatment time is increased, the time it takes for the ultrasound waves to impact the cell walls increases, and as a result, the number of cell walls is destroyed and the decomposition of anthocyanin also depends on the individual anthocyanin components.²⁵ At temperatures of 70°C, 80°C, and 90°C, similar

results were recorded. When heat treated, the combination of temperature and time applied has a significant influence on anthocyanin stability. Kinetic studies on the effects of heat treatment on phenolic compounds by Turturică and colleagues (2016) concluded that anthocyanins underwent the greatest degradation among polyphenols within the temperature range of 70°C and 110°C.

26 Anthocyanin content tends to decrease as temperature increases because anthocyanin belongs to the group of water-soluble flavonoids that are easily changed by heat treatment. This research result is similar to the results of the survey on the effects of ultrasound on the pasteurization process high intensity, sterilization, and pasteurization on the stability of anthocyanins in jamun fruit juice.²⁴

Table 1: Effect of ultrasonic pasteurization method on pH value and TSS

Treatment	TSS	pH
Control sample	7.00±0.00	3.97±0.20 ^a
58kHz-10min	7.00±0.00	3.71±0.01 ^b
58kHz-20min	7.00±0.00	3.72±0.01 ^b
58kHz-30min	7.00±0.00	3.72±0.08 ^b
132kHz-10min	7.00±0.00	3.71±0.10 ^b
132kHz-20min	7.00±0.00	3.72±0.02 ^b
132kHz -30min	7.00±0.00	3.73±0.05 ^b
192kHz -10min	7.00±0.00	3.71±0.02 ^b
192kHz -20min	7.00±0.00	3.72±0.01 ^b
192kHz -30min	7.00±0.00	3.73±0.03 ^b
70°C-10min	7.00±0.00	3.73±0.05 ^b
70°C-20min	7.00±0.00	3.71±0.02 ^b
70°C-30min	7.00±0.00	3.71±0.03 ^b
80°C-10min	7.00±0.00	3.71±0.02 ^b
80°C-20min	7.00±0.00	3.72±0.01 ^b
80°C-30min	7.00±0.00	3.70±0.01 ^b
90°C-10min	7.00±0.00	3.71±0.06 ^b
90°C-20min	7.00±0.00	3.71±0.04 ^b
90°C-30min	7.00±0.00	3.70±0.08 ^b

Data are presented as mean ± SD of triplicate analyses.

Different letters in the same column indicate statistically significant differences ($p < 0.05$) by LSD test.

The increase in flavonoids is associated with the formation of free radicals, and the release of bioflavonoids because during ultrasonic treatment, the cavitation effect generated by ultrasonic bubbles disrupts cell walls, promoting hydroxylation reactions in the aromatic rings of phenolic compounds. This facilitates the conversion of phenolics from their bound forms into free forms.²⁷ In addition, the results in this study also show that when the ultrasound and heat treatment time is increased, the flavonoid content decreases. Flavonoid content when treated for 10 minutes at 3 different frequencies is 3.57 mgQE/

mL (frequency 58 kHz), 3.68 mgQE/mL (frequency 132 kHz), and 3.37 mgQE/mL (frequency 192 kHz), twice as high as when increasing the processing time to 20 minutes and 30 minutes at the same frequency. Samples treated at temperatures of 70°C, 80°C, and 90°C also recorded similar results. The decrease in anthocyanin, flavonoid, and polyphenol content may be due to heat inactivation kinetics of mixed fruit drinks similar to the study by Bhalerao *et al.*²⁸ Active antioxidants (anthocyanins, polyphenols, flavonoids) are affected by high temperatures, which can lead to a decrease in

bioflavonoids and the greatest degradation is anthocyanins, followed by polyphenols and finally flavonoids.²⁹ Research on plant bioactive content and antioxidant properties as affected by heat treatment suggests that flavonoids are common secondary polyphenolic metabolites in plants affected by heat treatment (50 – 100°C) using quantitative spectroscopic and kinetic methods.³⁰ Depending on the nutritional composition of the food and the treatment conditions, either an increase or decrease in the levels of plant bioactive compounds may occur. Overall, ultrasound treatment demonstrated a positive impact on total flavonoid content.¹⁶

The more the ultrasound time increases, the stronger the gas cavitation phenomenon becomes, so the cell wall structures are broken down more, removing oxygen molecules, inserting -OH groups into the aromatic ring structure, making phenolic compounds have in the juice in its bound form with other molecules begins to release bound phenolics into free molecules. This process effectively enhances the diffusion rate of phenolic compounds, thereby contributing to the overall increase in their content.³¹ The occurrence of phenolic acids varies depending on the type of food matrix. In certain processed star fruit juice samples, a reduction or degradation of specific phenolic acids was observed, whereas in other cases, thermal pasteurization led to the emergence or increased levels of phenolic acids that were not initially present in the fresh juice.¹⁶

The increase in sugar content may be due to acid hydrolysis of sugar or disaccharide decomposition into monosaccharides.¹³ The reduced sugar content when treated for 10 minutes at different frequencies is 0.50 mg/mL (58kHz), 0.51 mg/mL (132 kHz), and 0.47 mg/mL (192kHz), lower than when increasing the time. Process up to 20 minutes and 30 minutes at the same frequency. At temperatures of 70°C, 80°C and 90°C, similar results were recorded. An increase in total sugar content after pasteurization was also observed for sugarcane juice³² and apple juice on the effects of processing and storage on the quality of the juice.³³

Both the temperature and duration of heating significantly influence the levels of bioactive

compounds.³⁴ The decrease in DPPH free radical scavenging ability may be because ascorbic acid has low thermal stability, it is easily oxidized during heat treatment, so prolonging the temperature and pasteurization time will reduce the scavenging ability of DPPH free radical.³⁵ The results are similar to studies on the effects of heating leading to phytochemical degradation such as loss or decomposition of certain types of phenolic compounds and other compounds responsible for DPPH free radical scavenging during processing heat treatment.¹⁶

Heat and ultrasound treatment did not affect TSS possibly due to the level of ultrasound energy, and the heat applied to the sample does not change the macromolecular structure related to these physicochemical properties at the microscopic level.³⁶ The slight change in pH reduction of the treated samples compared to the control sample may be due to the interaction of nutritional components in the sample leading to chemical reactions that create new chemical components. The results of this study are similar to studies on ultrasonic heat and pasteurization of red grape juice, *Opuntia ficus-indica* juice.^{8,37}

Conclusion

Pasteurized by ultrasonic treatment at a frequency of 132 kHz for 10 minutes, the content of anthocyanins, flavonoids, polyphenols, reducing sugars, and free radical scavenging ability (DPPH) in perilla water are all equivalent to the control (no treatment) and the most stable of all treatments. Overall, this study provides valuable information for beverage manufacturers to control the quality properties of perilla drink from *Perilla frutescens* (L.) Britton and *Chrysanthemum indicum* L. Moreover, the study can be used as a basis for further research on the effects of ultrasound treatments on the stability of antioxidant active ingredients as well as the rheological, antioxidant, and microbiological properties of perilla drinks packed in different packaging materials under different storage conditions.

Acknowledgement

This work was supported with equipment funding from Vinh Long University of Technology Education.

Funding Sources

The authors received no financial support for the research, authorship, and publication of this article.

Conflict of Interest

The authors do not have any conflict of interest.

Data Availability Statement

This statement does not apply to this article.

Ethics Statement

This research did not involve human participants, animal subjects, or any material that requires ethical approval.

Informed Consent Statement

This study did not involve human participants, and therefore, informed consent was not required.

Clinical Trial Registration

This research does not involve any clinical trials.

Permission to Reproduce Materials from Other Sources

Not Applicable.

Author Contributions

- **Nguyen Tri Yen Chi:** Conceptualization, Methodology, Writing – Review & Editing.
- **Nhat Anh Duong:** Writing – Original Draft, Supervision, Reviewing the manuscript.
- **Ngoc Nhu Y Huynh:** Analysis, Data Collection.
- **Ba Nhat Tan Nguyen:** Interpretation, Supported the manuscript writing process.

References

1. Aochen C, Kumar A, Jaiswal S, *et al.* *Perilla frutescens* L.: a dynamic food crop worthy of future challenges. *Front Nutr.* 2023;10:1130927. doi:10.3389/fnut.2023.1130927
2. Wu X, Dong S, Chen H, Guo M, Sun Z, Luo H. *Perilla frutescens*: A traditional medicine and food homologous plant. *Chinese herbal medicines.* Jul 2023;15(3):369-375. doi:10.1016/j.chmed.2023.03.002
3. Hou T, Netala VR, Zhang H, Xing Y, Li H, Zhang Z. *Perilla frutescens*: A Rich Source of Pharmacological Active Compounds. *Molecules.* 2022;27(11):3578. doi:10.3390/molecules27113578
4. Dolas R, Saravanan C, Kaur BP. Emergence and era of ultrasonic's in fruit juice preservation: A review. *Ultrasonics sonochemistry.* Nov 2019;58:104609. doi:10.1016/j.ultsonch.2019.05.026
5. Liu Y, Deng J, Zhao T, Yang X, Zhang J, Yang H. Bioavailability and mechanisms of dietary polyphenols affected by non-thermal processing technology in fruits and vegetables. *Current research in food science.* 2024;8:100715. doi:10.1016/j.crfs.2024.100715
6. Lauteri C, Ferri G, Piccinini A, Pennisi L, Vergara A. Ultrasound Technology as Inactivation Method for Foodborne Pathogens: A Review. *Foods* (Basel, Switzerland). Mar 13 2023;12(6)doi:10.3390/foods12061212
7. Li X, Zhang L, Peng Z, *et al.* The impact of ultrasonic treatment on blueberry wine anthocyanin color and its In-vitro anti-oxidant capacity. *Food Chem.* Dec 15 2020;333:127455. doi:10.1016/j.foodchem.2020.127455
8. Margean A, Lupu MI, Alexa E, *et al.* An Overview of Effects Induced by Pasteurization and High-Power Ultrasound Treatment on the Quality of Red Grape Juice. *Molecules.* Apr 4 2020;25(7)doi:10.3390/molecules25071669
9. Santhirasegaram V, Razali Z, Somasundram C. Effects of thermal treatment and sonication on quality attributes of Chokanan mango (*Mangifera indica* L.) juice. *Ultrason Sonochem.* Sep 2013;20(5):1276-82. doi:10.1016/j.ultsonch.2013.02.005
10. Aadil RM, Zeng XA, Zhang ZH, *et al.* Thermosonication: A potential technique that influences the quality of grapefruit juice. *International Journal of Food Science Technology.* 2015;50(5):1275-1282. doi:10.1111/ijfs.12766
11. Khandpur P, Gogate PR. Effect of novel

- ultrasound based processing on the nutrition quality of different fruit and vegetable juices. *Ultrasonics sonochemistry*. Nov 2015;27:125-136. doi:10.1016/j.ultsonch.2015.05.008
12. Gomes WF, Tiwari BK, Rodriguez Ó, de Brito ES, Fernandes FAN, Rodrigues S. Effect of ultrasound followed by high pressure processing on prebiotic cranberry juice. *Food Chem*. Mar 1 2017;218:261-268. doi:10.1016/j.foodchem.2016.08.132
13. Mukhtar K, Nabi BG, Arshad RN, *et al.* Potential impact of ultrasound, pulsed electric field, high-pressure processing and microfluidization against thermal treatments preservation regarding sugarcane juice (*Saccharum officinarum*). *Ultrason Sonochem*. Nov 2022;90:106194. doi:10.1016/j.ultsonch.2022.106194
14. Han SH, Zhu JK, Shao L, *et al.* Effects of Ultrasonic Treatment on Physical Stability of Lily Juice: Rheological Behavior, Particle Size, and Microstructure. *Foods* (Basel, Switzerland). Apr 21 2024;13(8)doi:10.3390/foods13081276
15. Bhutkar S, Brandão TRS, Silva CLM, Miller FA. Application of Ultrasound Treatments in the Processing and Production of High-Quality and Safe-to-Drink Kiwi Juice. *Foods* (Basel, Switzerland). Jan 20 2024;13(2) doi:10.3390/foods13020328
16. Saikia S, Mahnot NK, Mahanta CL. A comparative study on the effect of conventional thermal pasteurisation, microwave and ultrasound treatments on the antioxidant activity of five fruit juices. *Food Sci Technol Int*. Jun 2016;22(4):288-301. doi:10.1177/1082013215596466
17. Lin Y, Li C, Shi L, Wang L. Anthocyanins: Modified New Technologies and Challenges. *Foods* (Basel, Switzerland). Mar 23 2023;12(7)doi:10.3390/foods12071368
18. Anh DN, Chi NTY, Y HNN, Khoa NV. Effects of pasteurization process on biological active ingredients from herbal supplemented perilla drink. *TNU J Sci Technol*. 2024;229(01):325-332. doi:10.34238/tnu-jst.8801
19. Singleton VL, Rossi JA. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. *Am J Enol Vitic* 1965;16(3):144-158. doi:10.5344/ajev.1965.16.3.144
20. Miller GL. Use of dinitrosalicylic acid reagent for determination of reducing sugar. *Anal Chem*. 1959;31(3):426-428. doi:10.1021/ac60147a030
21. Giusti MM, Wrolstad RE. Characterization and measurement of anthocyanins by UV-visible spectroscopy. *Curr Protoc Food Anal Chem*. 2001;(1):F1. 2.1-F1. 2.13. doi:10.1002/0471142913.faf0102s00
22. Brand-Williams W, Cuvelier, M. E., Berset, C. Use of a free radical method to evaluate antioxidant activity *LWT-Food Sci Technol*. 1995;28(1):25-30. doi:10.1016/S0023-6438(95)80008-5
23. Zhishen J, Mengcheng T, Jianming W. The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. *Food Chem*. 1999;64(4):555-559. doi:10.1016/S0308-8146(98)00102-2
24. Shaheer C, Hafeeda P, Kumar R, Kathiravan T, Dhananjay K, Nadasabapathi S. Effect of thermal and thermosonication on anthocyanin stability in jamun (*Eugenia jambolana*) fruit juice. *Int Food Res J*. 2014;21(6):2189-2194.
25. Tiwari BK, Patras A, Brunton N, Cullen PJ, O'Donnell CP. Effect of ultrasound processing on anthocyanins and color of red grape juice. *Ultrason Sonochem*. Mar 2010;17(3):598-604. doi:10.1016/j.ultsonch.2009.10.009
26. Turturică M, Stănciuc N, Bahrim G, Râpeanu G. Investigations on Sweet Cherry Phenolic Degradation During Thermal Treatment Based on Fluorescence Spectroscopy and Inactivation Kinetics. *Food Biopro Technol*. 2016/10/01 2016;9(10):1706-1715. doi:10.1007/s11947-016-1753-7
27. Saeeduddin M, Abid M, Jabbar S, *et al.* Physicochemical parameters, bioactive compounds and microbial quality of sonicated pear juice. *Int J Food Sci Technol*. 2016;51(7):1552-1559. doi:10.1111/ijfs.13124
28. Bhalerao PP, Chakraborty S. Integrated calculation of pasteurization time: A case study for thermal inactivation kinetics of a mixed fruit beverage. *J Food Process Eng*. 2021;44(8):e13761. doi:10.1111/jfpe.13761
29. Turturică M, Stănciuc N, Bahrim G, Râpeanu G. Effect of thermal treatment on phenolic compounds from plum (*Prunus domestica*) extracts—A kinetic study. *J Food*

- Eng.* 2016;171:200-207. doi:10.1016/j.jfoodeng.2015.10.024
30. Ursache FM, Ghinea IO, Turturică M, Aprodu I, Râpeanu G, Stănciuc N. Phytochemicals content and antioxidant properties of sea buckthorn (*Hippophae rhamnoides* L.) as affected by heat treatment - Quantitative spectroscopic and kinetic approaches. *Food Chem.* Oct 15 2017;233:442-449. doi:10.1016/j.foodchem.2017.04.107
31. Das PS, Das P, Nayak PK, Islary A, Krishnan Kesavan R. Process optimization of thermosonicated modhusuleng (*polygonum microcephalum*) leaf juice for quality enhancement using response surface methodology. *Measurement: Food.* 2024:100181. doi:10.1016/j.meafoo.2024.100181
32. Bulegon R, de Almeida Gomes G, Rigo E. Influence of the pasteurization conditions on sugarcane juice packaged in glass packaging. *Rev Do Congr Sul Bras Eng Aliment.* 2018;4(1):13. doi:10.5965/24473650412018012
33. Bianchi F, Pünsch M, Venir E. Effect of Processing and Storage on the Quality of Beetroot and Apple Mixed Juice. *Foods* (Basel, Switzerland). May 11 2021;10(5) doi:10.3390/foods10051052
34. Chi NTY, Duong NA, Huynh NNY, Nguyen VK, Nguyen TTK. Effects of Storage Temperature on Biologically Active Ingredients of Perilla Drink. *Food Chem Adv.* 2025:100897. doi:10.1016/j.focha.2025.100897
35. Mieszczakowska-Fraç M, Celejewska K, Płocharski W. Impact of Innovative Technologies on the Content of Vitamin C and Its Bioavailability from Processed Fruit and Vegetable Products. *Antioxidants* (Basel, Switzerland). Jan 5 2021;10(1)doi:10.3390/antiox10010054
36. Qiu X, Su J, Nie J, *et al.* Effects of Thermosonication on the Antioxidant Capacity and Physicochemical, Bioactive, Microbiological, and Sensory Qualities of Blackcurrant Juice. *Foods* (Basel, Switzerland). Mar 6 2024;13(5)doi:10.3390/foods13050809
37. Ferreira RM, Amaral RA, Silva AM, Cardoso SM, Saraiva JA. Effect of High-Pressure and Thermal Pasteurization on Microbial and Physico-Chemical Properties of *Opuntia ficus-indica* Juices. *Beverages.* 2022;8(4):84. doi:10.3390/beverages8040084.