



# Article **Thermal Treatment Influence on Selected Nutritional Values of Common Sea Buckthorn (Hyppophae rhamnoides) Juice**

Ján Mezey<sup>1,\*</sup>, Ondrej Hegedűs<sup>2</sup>, Ivana Mezeyová<sup>1</sup>, Katarína Szarka<sup>2</sup> and Alžbeta Hegedűsová<sup>1</sup>

- <sup>1</sup> Institute of Horticulture, Faculty of Horticulture and Landscape Engineering, Slovak Agriculture University in Nitra, 949 01 Nitra, Slovakia; i.mezeyova@gmail.com (I.M.); alzbeta.hegedusova@uniag.sk (A.H.)
- <sup>2</sup> Department of Chemistry, Faculty of Education, J. Selye University, 945 01 Komárno, Slovakia; hegeduso@gmail.com (O.H.); szarkak@ujs.sk (K.S.)
- \* Correspondence: jan.mezey@uniag.sk; Tel.: +421-905-583-150

Abstract: Since ancient times, sea buckthorn (Hyppophae rhamnoides) (SBT) has been utilized as a medical plant for its ability to extract health-promoting compounds from its fruits, seeds, pulp, skin, bark, roots, and leaves. L-ascorbic acid is primarily found in fruits, and because of this, it can be utilized as a fortification agent to enhance other juices. The study's goal was to look into how the L-ascorbic acid and selected nutritional parameters in common sea buckthorn juice changed over the period of storage and different thermal treatments. The L-ascorbic acid stability in the processed juice in both used varieties ("Hergo" and "Leikora") was ensured by the processing technology with a modified vat (or batch) (low-temperature long-time pasteurization) process. Even after being sterilized at 120 °C for 15 min, the amount of L-ascorbic acid in the processed sea buckthorn juice in both varieties was unaffected and ranged between 1762 and 2058 mg/kg. There was no change in the pH level at the same time; it stayed extremely low (about 2.3), which may have helped the L-ascorbic acid to stabilize. The sterilized juice variant in both varieties had the highest levels of glucose, fructose, total sugar, malic acid, total acid, and total soluble solids (TSS), which were significantly higher than in fresh juice or in either variant after pasteurization. Given this, we advise processing the SBT berries immediately after harvest using thermal processing to prevent ascorbic acid (AA) loss, or storing them under frost conditions until processing.

**Keywords:** L-ascorbic acid; sea buckthorn (*Hyppophae rhamnoides*); pasteurization; nutritional parameters

# 1. Introduction

For centuries, the people of Central and Southeastern Asia have used sea buckthorn (SBT) as an agent of traditional medicine to prevent various ailments. It has been considered to be one of the most important valuable bio-resources, and has been used locally for centuries as fuel, fodder, small timber, food, and medicine. Every part of the plant, such as the fruit, leaf, twig, root, and bark, has been traditionally used as medicine, nutritional supplements, firewood, and fences [1,2]. A wide spectrum of pharmacological effects of SBT have been recently reported, including antioxidant, immunomodulatory, anti-atherogenic, anti-stress, hepatoprotective, radioprotective, and tissue repair [3–5]. The phytochemistry review of SB showed that the nutrition constituents, including flavonoids, vitamins, fatty acids, carotenoids, and phytosterols, as well as mineral elements, were particularly rich in SB, and was followed by detailed information of the main constituents of SB [6]. The bioactive components vary with fruit maturity, fruit size, species, geographic locations, climate, and methods of extraction [7,8].

Sea buckthorn juice is one of the most popular processing products used as raw material for further processing, for example, in the making of syrup. The nutritional content of the juice and syrup depends on the processing technologies and equipment



Citation: Mezey, J.; Hegedűs, O.; Mezeyová, I.; Szarka, K.; Hegedűsová, A. Thermal Treatment Influence on Selected Nutritional Values of Common Sea Buckthorn (*Hyppophae rhamnoides*) Juice. *Agronomy* **2022**, *12*, 1834. https:// doi.org/10.3390/agronomy12081834

Academic Editor: Gerardo Fernández Barbero

Received: 29 June 2022 Accepted: 30 July 2022 Published: 2 August 2022

**Publisher's Note:** MDPI stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.



**Copyright:** © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). used [9–11]. The flavor of sea buckthorn (*Hippophae rhamnoides* L.) berries is perceived as sour and astringent, mainly due to the high acid content; most importantly, malic acid [12,13]. Juice from ripe fruits is extracted by pressing or centrifugal techniques, and can be stored under refrigeration for a few days and requires pasteurization or freezing for long-term storage. For preservation purposes, it is necessary to sterilize/pasteurize the juice. High-temperature–short-time (HTST) processes of 80–90  $^\circ$ C for several seconds are preferred. The storage of the juice beyond 6 months results in browning, but the shelf life of the juice can be extended by storing at  $4 \degree C$  [14]. The high temperature used for processing and preservation (pasteurization, sterilization, blanching) of the juices results in undesirable changes in their organoleptic features (flavor, color, texture), and often results in considerable losses in their valuable bio-active components, such as vitamin C [15]. The total ascorbic acid loss during industrial juice production is about 5–11%. Other researchers observed that the storage of thermal-treated (HTST 90 °C, 45 s) sea buckthorn juice for 7 days at 6 °C resulted in a degradation of vitamin C of about 11 to 12% [9]. The production of concentrated juice results in 50% depletion of the total ascorbic acid [16]. Thermal pasteurization of the sea buckthorn juice at 80°C for 10 min has led to significant losses of vitamin C (approximately 86%) within a period of 3 months of storage at refrigeration conditions [17]. Pectin methyl esterase (PME) is involved in the disintegration of the cell wall, among other functions, and its inactivation results in L-ascorbic acid remaining inside the cells. The thermal inactivation of PME has been the subject of studies, and is usually inactivated by pasteurization at elevated temperatures [18]. Data for the isothermal inactivation of sea buckthorn PME obtained in the temperature range from 40 to 70 °C could be accurately modeled by applying the first-order kinetic model. For higher studied temperatures (80 °C), no residual activity was detected even after 10 s of processing [19].

The study's objective was to evaluate if any L-ascorbic acid was lost during the thermal treatment procedure. The partial aims include finding out how the thermal processing and storage affect sensorially important nutritional values of common sea buckthorn juice within a respond to request of PD Tvrdošovce, the largest manufacturer and processor of SBT in Slovakia.

#### 2. Materials and Methods

## 2.1. Plant Material Handling

Sea buckthorn (*Hippophae rhamnoides* L.) is a berry-bearing, hardy bush belonging to the *Elaeagnaceae* family that grows widely in various regions of Asia, Europe, and North America [20]. It produces orange-yellow berries in the autumn. The botanical definition of a berry is a particular type of small fleshy fruit [21].

In southwest Slovakia (PD Tvrdošovce), a commercial 15-ha organic SBT orchard (Figure 1A,B) produced the berries. In terms of climate, the area is characterized by a warm and dry summer and a mildly warm, dry, or extremely dry winter. The annual mean air temperature and annual rainfall for Tvrdošovce, as determined by the long-term climatic average (1961–2000), are 9.8 °C and 522 mm, respectively. The experimental area's soil type ranges from brown soil to chernozem on loess and loess loams. Bushes are grown as an organic orchard with  $4 \times 2$  m spacing and are 13 years old. About 10 to 15 lateral skeleton branches, from which bearing shoots were developed, are present in each bush. On 21 September 2018, entire branches (Figure 1C) of sea buckthorn berries (*Hippophae rhamnoides*) were collected at the stage of commercial maturity, as determined by evaluating the fruit hardiness with a penetrometer (Fruit test, Wagner Instruments, Greenwich, USA). We used the "Hergo" and "Leikora" varieties (Figure 2). Because these are the ones that are most prevalent in the producer's plantings and juices, the varieties were chosen at the producer's request; both ripen at the end of September or the beginning of October.



**Figure 1.** (**A**) Commercial sea buckthorn planting at PD Tvrdošovce; (**B**,**C**) mature bushes right before harvest; (**D**) industrial deep-freezer with harvested branches in big-boxes; (**E**) tools for separating berries from branches.



Figure 2. Evaluated varieties of sea buckthorn (Hyppophae rhamnoides).

## 2.2. Chemical Parameters

In the context of making juice, sea buckthorn fruits, which are a significant source of nutrients and other intriguing compounds, were assessed analytically. The content of total acids, malic acid, total sugar, glucose, fructose, pH, and total soluble solids (TSS) were analyzed with an FT-IR spectrophotometer (Alpha Wine Analyzer, module for juices, Bruker Optics, Billerica, MA, USA).

The harvest of berries was performed by pruning whole branches, which were placed in 300 L volume plastic bins and placed into an industrial deep-freezer adjusted to -40 °C (Figure 1D). According to the variants, they were picked out from deep-freezer big-boxes and knocked down from branches and naturally melted (Figure 1E). The total volume of processed fruit was 5 kg from various big-boxes placed in the deep-freezer. The berries were cleaned to remove damaged, diseased, or pest-infected fruits. The berries were crushed in a commercial fruit juicer with 1200 rpm without heating (Magimix Duo Le Plus XL, Vincennes, France). The total volume of the obtained juice was 4.25 L from each SBT variety (85% juice yield). A volume of 100 mL of the juice was strained through sieves of 0.2 mm diameter, filled in 8  $\times$  15 mL test tubes, and processed through a centrifuge (Hettich BV-20, Geldermaslen, The Netherlands) for 120 s at 6000 RPM (4032 RCF, or Gforce). Sample injection was performed with a syringe, using 5 mL of the sample positioned in contact with attenuated total reflectance on a diamond crystal, and analyzed in an FT-IR spectrophotometer (Alpha Wine Analyzer, module for juices, Bruker Optics, USA). After three measurements, background measurements were made using double-distilled water. One-hundred and twenty scans were acquired for both the background and sample measurements. At a sample temperature of 40  $^{\circ}$ C and a spectral resolution of 14 cm<sup>-1</sup>, all FT-IR spectra were captured in the range of 4000 to 350 cm<sup>-1</sup>. The measured data were displayed as a g/1 content, except TSS, which was displayed as °BRIX. Each procedure, starting from centrifugation, was repeated three times [22]. In MS Excel and Statgraphic Centurion XVII (StatPoint Inc., Herndon, VA, USA), the data from the three measurements were further processed.

L-ascorbic acid (AA) content was measured by the HPLC method described in the literature [23]. The tested material was homogenized in a solution containing 2% oxalic acid.

The homogenate was extracted in an ultrasonic bath, the extract was clarified and filtered, and then 20 L of the clear extract was dosed for analysis. Waters HPLC system, Milford, MA, USA: Waters 2489 UV/VIS Detector, Waters 1525 Binary HPLC Pump, chromatographic column: RP C-18, 5 m, ID 4.6 mm, length 150 mm, mobile phase: acetonitrile–phosphate buffer solution (pH = 3.5) 5:95, flow rate: 0.5 mL/min, UV detection 264 nm, where AA has an absorption maximum, was used to ensure separation. L-ascorbic acid (CertiPUR, Merck, Darmstadt, Germany) was used for the preparation of standard solutions.

Every measurement underwent three replications, and parallel AA standard quantification was performed. After 23 and 52 days of storage at -20 °C, the same measurements were made.

#### 2.3. Thermal Treatment

In thermal treatment, 3 triplicated variants were made. In the modified VAT pasteurization process (MVPP) 1x, the juice was treated by pasteurization at 85 °C for 5 min. In the modified VAT pasteurization process (MVPP) 2x, the juice was treated by pasteurization at 85 °C for 5 min, then cooled down to 5 °C, and immediately treated again with MVPP at 85 °C for 5 min, while rapid heating to pasteurization temperature and subsequent intense cooling was ensured. This technique simulates the thermal treatment of SBT juice provided by the company, PD Tvrdošovce, which is a manufacturer of commercial SBT products. Because L-ascorbic acid has a relatively high stability in juice at low pH (about 2.3), it was tested even at a sterilizing temperature of 120 °C for 15 min (variant 3—sterilization). In all variants, the thermal treatment of 250 mL of SBT juice was performed in 500 mL glass beakers which were placed in 27 L pasteurizing pot (Warmmaster, Merten und Storck GmbH, Germany) filled with 5 L of tap water, heated to estimated temperature, and kept at an estimated temperature and time with the use of a digital thermostat.

#### 2.4. Statistical Evaluation

A statistical analysis was performed using Statgraphic Centurion XVII (StatPoint Inc. USA). The obtained results were evaluated by analysis of variance (ANOVA), the multifactor analysis of variance, and the multiple range test.

The AA determination method was validated, and the analyses' results were compared to the method's relative expanded uncertainty, which was determined to be Ux = 12 percent ( $\alpha = 0.05$ ) [23].

#### 3. Results and Discussion

In fresh SBT juice, the fructose (Table 1) content ranged, according to variety, between 25.44 and 31.92 g/L. The fructose content in 10 SBT populations in Iran ranged from 22.91 to 132.06 g/L, with a mean value 62.82 g/L [16]. Different elements, such as climatic conditions, variety, level of phenological maturity, etc., might be responsible for the higher fructose concentration. The glucose content (Table 1) ranged between 7.83 and 10.72 g/L. Glucose content in Iranian SBT ranged from 38.14 to 110.70 g/L, with a mean value 65.36 g/L [24], and the sugar content in SBT from two Dagestan regions varied between 51.9 to 57.4 g/L [25]. Similar to the case of fructose, we also ascribe the increased glucose level in this instance to the meteorological conditions, which are very different from those of Central Europe. The TSS (Table 1) ranged between 4.71 to 4.81 °Brix. In Iranian SBT berries, the SSC content ranged from 11.85 to 31.50 g/L [24], and in Russian berries, it ranged between 12.3 and 14.4% [25], and in other studies, from 12.56 to 14.67% [26] and 6.4 to 12.7 °Brix [27]. In the Leikora variety, the TSS was 7.5 °Brix [28], 5.6 °Brix [29], and 7.4–12.6 °Brix, respectively. In Turkish SBT, TSS values between 10.1–14.8 °Brix were measured [30], and in Chinese genotypes, the reported values were between 10.2–22.7 °Brix [31,32]. The total sugar content (Table 1) ranged between 43.06 and 44.26 g/L.

| Variaty | Variant       | Fructose                  | Glucose                   | TSS                      | Total Sugar               |
|---------|---------------|---------------------------|---------------------------|--------------------------|---------------------------|
| vallety | variant       | (g/L)                     | (g/L)                     | (°BRIX)                  | (g/L)                     |
|         | sterilization | $26.53\pm4.03$            | $8.94 \pm 1.6$            | $5.37 \pm 1.01$          | $47.64 \pm 4.25$          |
| Leikora | MVPP 2x       | $23.95\pm3.88$            | $7.81 \pm 1.52$           | $4.66 \pm 1.30$          | $42.15\pm3.19$            |
| Leikula | MVPP 1x       | $22.73\pm3.92$            | $7.58 \pm 1.67$           | $4.62\pm0.99$            | $43.52\pm3.32$            |
|         | fresh         | $25.44 \pm 4.12$          | $7.83 \pm 1.71$           | $4.81\pm0.78$            | $44.26\pm3.56$            |
|         | sterilization | $33.88 \pm 5.98$          | $12.92 \pm 1.58$          | $5.26 \pm 1.12$          | $45.81 \pm 4.01$          |
| Horao   | MVPP 2x       | $29.74 \pm 5.97$          | $11.77 \pm 1.65$          | $4.71\pm0.79$            | $41.68\pm3.25$            |
| Hergo   | MVPP 1x       | $26.59\pm 6.04$           | $11.23 \pm 1.78$          | $4.7\pm0.8$              | $42.15\pm3.28$            |
|         | fresh         | $31.92\pm 6.24$           | $10.72 \pm 1.52$          | $4.71\pm0.85$            | $43.06\pm3.45$            |
| Average | sterilization | $30.21\pm3.68~\mathrm{d}$ | $10.93\pm1.99~\mathrm{b}$ | $5.32\pm0.06~\text{b}$   | $46.73\pm0.91~\mathrm{b}$ |
|         | MVPP 2x       | $26.85\pm2.90b$           | $9.79\pm1.98~\mathrm{a}$  | $4.69\pm0.02~\mathrm{a}$ | $41.92\pm0.23~\mathrm{a}$ |
|         | MVPP 1x       | $24.66\pm0.57~\mathrm{a}$ | $9.41\pm1.83~\mathrm{a}$  | $4.66\pm0.04~\mathrm{a}$ | $42.84\pm0.69~ab$         |
|         | fresh         | $28.68\pm3.24~\mathrm{c}$ | $9.28\pm1.45~\mathrm{a}$  | $4.76\pm0.05$ a          | $43.66\pm0.6~\text{ab}$   |

**Table 1.** Fructose, glucose, TSS, and total sugar content change during thermal treatment variants and their statistical evaluation.

The different letters listed with the SD values in the columns represent statistically significant differences between the observed variants on average at p < 0.05 by LSD in ANOVA (Statgraphic XVII).

The variation in total sugar content, glucose, fructose content, and TSS over a wide range of values is due to many circumstances, e.g., the time of harvesting the fruits and their degree of ripeness (they can be harvested in autumn, at full maturity; or in winter, after the fruit freezes), as well as the origin, population, and genetic background of the plant [33,34].

# 3.1. Acids

The malic acid content (Table 2) varied between 21.34 and 29.98 g/L; another study reported values between 16.4 and 17.4 g/L [8]. Depending on the fruits' origin, variations of the malic acid content in the sea buckthorn juice were between 11 and 60 g/L [34], and 6.3 g/L, respectively [35]. In our study, the pH value (Table 2) varied between 2.31 and 2.38. Similar results were obtained for some Romanian berries: at the initial moment of the analysis, the pH values of the juices varied between 2.7 (the control batch) and 3.25. The pH value varied between 2.45 and 2.61 [25], and 2.5 to 2.73 [27]. The total titrable acids (Table 2) varied between 26.11 and 34.15 g/L, and in other studies, ranged between 31.4 and 47.3 g/L [26].

**Table 2.** Malic acid, pH level, and total acid content change during thermal treatment variants and their statistical evaluation.

| Variaty | Variant       | Malic Acid       | pH            | Total Acid       |
|---------|---------------|------------------|---------------|------------------|
| Vallety |               | (g/L)            |               | (g/L)            |
|         | sterilization | $22.98\pm3.58$   | $2.38\pm0.21$ | $28.06 \pm 4.01$ |
| Leikora | MVPP 2x       | $22.56\pm3.78$   | $2.34\pm0.15$ | $27.26\pm3.9$    |
| Lentoru | MVPP 1x       | $22.04 \pm 3.45$ | $2.36\pm0.18$ | $26.54 \pm 3.81$ |
|         | fresh         | $21.34\pm3.12$   | $2.38\pm0.25$ | $26.11 \pm 4.12$ |
|         | sterilization | $32.66\pm5.41$   | $2.3\pm0.11$  | $37.17 \pm 5.25$ |
| Horgo   | MVPP 2x       | $31.24\pm5.65$   | $2.28\pm0.27$ | $35.28 \pm 5.11$ |
| Tiergo  | MVPP 1x       | $30.56\pm3.64$   | $2.3\pm0.09$  | $34.98 \pm 6.01$ |
|         | fresh         | $29.98 \pm 5.89$ | $2.31\pm0.17$ | $34.15\pm5.54$   |

| Variety | Variant       | Malic Acid                | pH                       | Total Acid                |
|---------|---------------|---------------------------|--------------------------|---------------------------|
|         | variant       | (g/L)                     |                          | (g/L)                     |
|         | sterilization | $27.82\pm4.84~\mathrm{a}$ | $2.34\pm0.04~\mathrm{a}$ | $32.62\pm4.55~\text{b}$   |
| Average | MVPP 2x       | $26.9\pm4.34~\mathrm{a}$  | $2.31\pm0.03~\mathrm{a}$ | $31.27\pm4.01~\mathrm{a}$ |
|         | MVPP 1x       | $26.3\pm4.26~\mathrm{a}$  | $2.33\pm0.03~\mathrm{a}$ | $30.76\pm4.22~\mathrm{a}$ |
|         | fresh         | $25.66\pm4.32~\mathrm{a}$ | $2.35\pm0.03$ a          | $30.13\pm4.02~\mathrm{a}$ |

Table 2. Cont.

The different letters listed with the SD values in the columns represent statistically significant differences between the observed variants on average at p < 0.05 by LSD in ANOVA (Statgraphic XVII).

The raw material's maturity is a crucial consideration, just as it is in the case of the sugar and acid content. The acid content of the fruit decreases and the pH value increases as the degree of berry ripeness increases.

## 3.2. Thermal Treatment—Sugars

Table 1 displays the changing values of selected sugars based on the processing technology. Only the sterilized version had a significant change in the total sugar content compared to the MVPP 2x variant (p < 0.05). The highest fructose content was achieved by sterilization, and the lowest by MVPP 1x. All variants showed a significant change in fructose content (p < 0.05). The variant with sterilization had the highest value for the contents of glucose and TSS, and the content had considerably risen (p < 0.05) in comparison to the other three variants (Table 1). This fact, according to our theory, is caused by more water evaporation at high thermal treatment temperatures (120 °C). During thermal treatment, water evaporation increases with temperature, which also increases the concentration of sugars. Partly different results were achieved in starfruit (Averrhoa carambola): the treatment conditions did not produce any effect on the values of TSS, and only a little increase in TSS occurred, and it was estimated as 7.0 °Brix when the juice was heated at 90 °C for 30–40 min. [36]. Another study proclaimed that there was no significant difference (p > 0.05) in the fructose concentration in sea buckthorn juice treated at 90, 100, and 110 °C, but the content of fructose in the samples treated at 120 °C showed a significant difference (p < 0.05) between the initial and treated samples after 30 min [37]. As a result, we propose that, above all, a higher temperature can be used to clearly alter the fructose content of sea buckthorn juice.

#### 3.3. Thermal Treatment—Acids

The variant with sterilization had the highest total acid content, and its content significantly increased (p < 0.05) in comparison to the other three variants, with values increasing from 34.35 g/L in fresh juice to 36.96 g/L in the sterilized variant. We conclude that thermal treatment has no discernible effect on the change in malic acid content or pH level, since there was no significant (p < 0.05) content change in either (Table 2). We assume that a change in the concentration of other acids present in the juice caused a significant change in the content of total acids following sterilization. In starfruit (*Averrhoa carambola*), with increasing temperature and time, the pH values increased significantly ( $p \le$  acids of star fruit juice may be destroyed during heat treatment, and this ultimately affects the pH value of the juice) [36]. We can infer from this study's authors' varied usage of heat treatment time and temperature (70 °C, 80 °C, and 90 °C for 10, 20, 30, and 40 min.) that changes in acid content become more apparent with longer heat treatment times, whereas with short-term treatment, this was not the case, which was confirmed by no change of acid content after 90 °C/15 s thermal treatment in blueberry juice [38].

According to the results, it can be declared that the thermal treatment of SBT juice has a significant (p < 0.05) influence on the total sugar, glucose, fructose, TSS, and total acid content. No influence on the content of malic acid and the pH level was observed. By all varieties and in all analyzed parameters, the highest values of glucose, fructose, total

sugar, TSS (Table 1), malic acid, and total titratable acid were observed in the variant with sterilized juice (Table 2).

#### 3.4. L-Ascorbic Acid Content

Our results indicated that fresh SBT juice contained L-ascorbic acid in amounts ranging from 1090 to 2588 mg/L (Table 3). Variants with thermal treatment have ascorbic acid contents ranging from 1047 to 2682 mg/L (Table 4).

Table 3. Content of ascorbic acid in sea buckthorn fruit stored under freezing conditions.

| Storago                | Leikora    |                        | Hergo      |                        |
|------------------------|------------|------------------------|------------|------------------------|
| Storage                | AA (mg/kg) | U <sub>x</sub> (mg/kg) | AA (mg/kg) | U <sub>x</sub> (mg/kg) |
| fresh (no storage)     | 1149       | 138                    | 2551       | 306                    |
| 23 days -20 °C         | 1190       | 143                    | 2588       | 311                    |
| 52 days $-20~^\circ C$ | 1090       | 131                    | 2582       | 310                    |

Ux—uncertainty of measurement.

Table 4. Ascorbic acid content in sea buckthorn juice during heat treatment.

| Storage       | Leikora    |                        | Hergo      |                        |  |
|---------------|------------|------------------------|------------|------------------------|--|
| Storage       | AA (mg/kg) | U <sub>x</sub> (mg/kg) | AA (mg/kg) | U <sub>x</sub> (mg/kg) |  |
| fresh         | 1091       | 131                    | 2523       | 303                    |  |
| MVPP 1x       | 1154       | 138                    | 2641       | 317                    |  |
| MVPP 2x       | 1270       | 152                    | 2682       | 322                    |  |
| sterilization | 1047       | 126                    | 2477       | 297                    |  |

U<sub>x</sub>—uncertainty of measurement.

The vitamin C content in Iranian SBT was in the range between 1390 and 10,000 mg/L [24]; in another study, the vitamin C content was between 253 and 1772 mg/L [25]; and vitamin C content in 10 SBT genotypes from Turkey was between 449 and 571 mg/L [26], and 3200 mg/L, respectively [35]. The concentration of vitamin C varies from 0.3 to 3.1 g/kg fruit in the European *H. rhamnoides* subspecies, from 0.4 to 3 g/kg fruit in Russian varieties belonging to the Mongolian subspecies, from 4.6 to 13.3 g/kg fruit in the *H. rhamnoides* subspecies, and from 2 to 25 g/kg fruit in the Chinese *H. rhamnoides* subspecies [39].

Sea buckthorn juice is produced on a large scale by first freezing the fruit on chopped branches for the necessary period of time, shaking the fruit, and then cold-pressing it. After being kept in sea buckthorn juice for a long time at -20 °C, the amount of L-ascorbic acid was assessed using a lab model of this activity (Table 3). The results of the studies demonstrate the high stability of AA during the period of both monitored varieties' whole storage time. We monitored the AA content after the single (MVPP 1x) and double pasteurization (MVPP 2x) of juice for 5 min based on the experience of a large manufacturer (PD Tvrdošovce) of preserved sea buckthorn juice (Table 4). The measured changes in AA content before and after pasteurization are lower than the declared expanded uncertainty of the used determination method. The lowest AA content was observed in the 120 °C sterilization variant, as expected, but even this difference was less than the expanded uncertainty of the method. Because of the thermolability and oxilability of AA [40–42], the results obtained from the heat treatment variants of SBT juice are remarkable. Our results are also confirmed by a study where vitamin C and total carotene content did not alter significantly depending on the pasteurization temperature and duration [43].

We assume that the thermal treatment caused PME to become inactive, and as a result, the AA concentration remained steady, which is supported by other investigations [19,44]. PME inactivation and residual antioxidant activity were both considerably impacted by

thermal treatment. When compared to an untreated sample, processing even under benign circumstances (60 °C for 1 min) reduced the antioxidant activity by 2.5 times, and inactivated PME by 50% [1]. Because all SBT juice varieties have extremely low pH levels, we assume that the AA in SBT fruits has good heat stability. These claims deserve a separate investigation, during which, we will also pay attention to the dynamics of changes in the various examined parameters (e.g., changing pH level/time of thermal treatment/thermal treatment temperature—AA content).

Due to the lack of studies that address the same issues on sea buckthorn, we were unable to find more relevant sources for discussion, but we confirmed the high organic acid content [12,13]; valuable sugars [24,25,45]; relatively high total soluble solid content [24,25,27,29,31,32]; and, in comparison to other fruits, the extremely high vitamin C content [4,24–26,39].

## 4. Conclusions

Because sea buckthorn fruits are typically fairly tart, adding them to mixed juices is an innovative approach to make use of their concentrated antioxidants and nutrients. We can achieve a very pleasant sweet and sour taste enriched with valuable bioactive substances by adding even a small amount of sea buckthorn juice to apple juice, for example. The sterilized juice variant in both varieties had the highest levels of glucose, fructose, total sugar, malic acid, total acid, and TSS, which were significantly higher than in fresh juice or in either variant after pasteurization. The juice's evaporation of water is the cause of this fact.

Significant changes (p < 0.05) between cold storage, the frozen variant, and fresh juice were found in the L-ascorbic acid content change throughout storage. We come to the conclusion that if we immediately freeze the berries after harvest and store them for at least 52 days at -20 °C, there will not be any significant L-ascorbic acid loss. However, if we store the berries or raw juice for 16 days at 5 °C, there will be a significant (p < 0.05) loss of L-ascorbic acid content, in the range of 11 to 15%.

The L-ascorbic acid concentration was stable in both sea buckthorn varieties, and thermal treatments had no impact on it at all. There was no change in the pH level concurrently; it stayed extremely low (about 2.3), which may have helped the L-ascorbic acid stabilize.

Given this, we advise processing the sea buckthorn berries immediately after harvest using thermal processing to prevent L-ascorbic acid loss, or storing them under frost conditions until processing.

With the sea buckthorn berries' 2000 mg/kg L-ascorbic acid content, 50 mL of sea buckthorn juice is all that is required to meet an adult's daily vitamin C requirement of 100 mg.

**Author Contributions:** Conceptualization, J.M. and O.H.; methodology, J.M. and O.H.; validation, A.H., I.M. and O.H.; formal analysis, J.M. and K.S.; investigation, J.M.; resources, J.M.; writing—original draft preparation, J.M.; writing—review and editing, J.M., I.M., A.H. and O.H.; supervision, A.H. and O.H.; funding acquisition, I.M. All authors have read and agreed to the published version of the manuscript.

**Funding:** This research was funded by KEGA, grant number KEGA 004SPU-4/2022, "Interactive Classroom for Horticulture study program in the Context of Innovation of the Current Student's Teaching Process".

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: Not applicable.

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

# References

- Suryakumar, G.; Gupta, A. Medicinal and therapeutic potential of Sea buckthorn (*Hippophae rhamnoides* L.). *J. Ethnopharmacol.* 2011, 138, 268–278. [CrossRef] [PubMed]
- Dhyani, D.R.; Maikhuri, K.; Misra, S.; Rao, K.S. Endorsing the declining indigenous ethnobotanical knowledge system of Seabuckthorn in Central Himalaya. *Indian J. Ethnopharmacol.* 2020, 127, 329–334. [CrossRef] [PubMed]
- Upadhyay, N.K.; Kumar, R.; Mandotra, S.K.; Meena, R.N.; Siddiqui, M.S.; Sawhney, R.C.; Gupta, A. Safety and wound healing efficacy of sea buckthorn (*Hippophae rhamnoides* L.) seed oil in experimental rats. *Food Chem. Toxicol.* 2009, 47, 1146–1153. [CrossRef] [PubMed]
- 4. Vilas-Franquesa, A.; Saldo, J.; Juan, B. Food Production. *Processing Nutr.* 2020, 2, 1–17.
- 5. Wani, T.A.; Wani, S.M.; Ahmad, M.; Ahmad, M.; Gani, A.; Masoodi, F.A. Influence of processing on physicochemical and antioxidant properties of apricot (*Prunus armeniaca* L. variety Narmo). *Cogent Food Agric*. **2016**, *2*, 1–9.
- 6. Ren, R.; Li, N.; Su, C.; Wang, Y.; Zhao, X.; Yang, L.; Li, Y.; Zhang, B.; Chen, J.; Ma, X. The bioactive components as well as the nutritional and health effects of sea buckthorn. *RSC Adv.* **2020**, *10*, 44654. [CrossRef]
- Leskinen, H.M.; Suomela, J.P.; Yang, B.; Kallio, H.P. Regioisomer compositions of vaccenic and oleic acid containing triacylglycerols in sea buckthorn (*Hippophae rhamnoides*) pulp oils: Influence of origin and weather conditions. J. Agric. Food Chem. 2010, 58, 537–545. [CrossRef]
- 8. Pop, R.; Weesepoel, Y.; Socaciu, C.; Pintea, A.; Vincken, J.P. Carotenoid composition of berries and leaves from six Romanian sea buckthorn (*Hippophae rhamnoides*) varieties. *Food Chem.* **2014**, *147*, 1–9. [CrossRef]
- Isrigova, T.A.; Salmanov, M.M.; Isrigova, V.S.; Taibova, D.S.; Sannikova, E.V. Development of a technology for the production of a functional food based on plant raw materials. In Proceedings of the E3S Web of Conferences Cep. International Scientific and Practical Conference "Development of the Agro-Industrial Complex in the Context of Robotization and Digitalization of Production in Russia and Abroad", DAIC, Yekaterinburg, Russia, 15–16 October 2020.
- 10. Mukailov, M.D.; Ulchibekova, N.A.; Isrigova, T.A.; Akhmedov, M.E.; Selimova, U.A. Functional foods produced from strawberries. *Int. J. Adv. Sci. Technol.* **2020**, *9*, 03003.
- 11. Titorenko, K.V.; Zhichkin, K.A. Innovative approaches to breeding in the dairy industry. *IOP Conf. Ser. Earth Environ. Sci.* 2021, 723, 032003. [CrossRef]
- 12. Tiitinen, K.; Vahvaselka, M.; Laakso, S.; Kallio, H. Malolactic fermentation in four varieties of sea buckthorn (*Hippophae rhamnoides* L.). *Eur. Food Res. Technol.* **2007**, 224, 725–732. [CrossRef]
- 13. Walczak-Zeidler, K.; Feliczak-Guzik, A.; Nowak, I. Oleje Roslinne Stosowane Jako Surowce Kosmetyczne-Leksykon; Wydawnictwo Cursiva: Kostrzyn, Poland, 2012; ISBN 9788362108206.
- 14. Stobdan, T.; Korekar, G.; Srivastava, R.B. Nutritional attributes and health application of seabuckthorn (*Hippophae rhamnoides* L.) a review. *Current Nutr. Food Sci.* 2013, *9*, 151–165. [CrossRef]
- 15. Verbeyst, L.; Bogaerts, R.; Plancken, I.; Hendrickx, M.; Loe, A. Modeling of vitamin C degradation during thermal and highpressure treatments of red fruit. *Food Bioprocess Technol.* **2013**, *6*, 1015–1023. [CrossRef]
- Gutzeit, D.; Baleanu, G.; Winterhalter, P.; Jerz, G. Vitamin C content in sea buckthorn berries (*Hippophaë rhamnoides* L. ssp. *rhamnoides*) and related products: A kinetic study on the storage stability and the determination of processing effects. *Food Sci.* 2008, 73, C615–C620. [CrossRef]
- 17. Manea, I.; Buruleanu, L. Mathematical model for the evaluation of the sea-buckthorn juice preservation. *Ovidius Univ. Ann. Chem.* **2009**, *20*, 83–86.
- Sampedro, F.; Geveke, D.J.; Fan, X.; Zhang, H.Q. Effect of PEF, HHP and thermal treatment on PME inactivation and volatile compounds concentration of an orange juice–milk based beverage. *Innov. Food Sci. Emerg. Technol.* 2009, 10, 463–469. [CrossRef]
- Alexandrakis, Z.; Kyriakopoulou, K.; Katsaros, G.; Krokida, M.; Taoukis, P. Selection of Process Conditions for High Pressure Pasteurization of Sea Buckthorn Juice Retaining High Antioxidant Activity. *Food Bioprocess Technol.* 2014, 7, 3226–3234. [CrossRef]
- 20. Bal, L.M.; Meda, V.; Naik, S.N.; Satya, S. Sea buckthorn berries: A potential source of valuable nutrients for neutraceuticals and cosmoceuticals. *Food Res. Int.* 2011, 44, 1718–1727. [CrossRef]
- Barkhuu, B.; Lodonjav, M.; Ganzorig, O.; Tumurtogoo, N. The Physicochemical Composition of Sea Buckthorn (*Hippophae rhamnoides* L.) Oil and Its Treatment Characteristics. In Proceedings of the 5th International Conference on Chemical Investigation and Utilization of Natural Resource (ICCIUNR-2021), Ulaanbaatar, Mongolia, 4–15 October 2021; Atlantis Press: Dordrecht, The Netherlands, 2021; pp. 43–51.
- 22. Mezey, J.; Mezeyová, I. Changes in the levels of selected organic acids and sugars in apple juice after cold storage. *Czech J. Food Sci.* **2018**, *36*, 175–180.
- Hegedűs, O.; Szarka, K.; Hegedűsová, A.; Gódány, Z.; Šlosár, M.; Nechifor, A.C.; Tonk, S. Validation and Quality Assurance of Ascorbic Acid Determination in Agricultural Products. *Rev. De Chim.* 2019, 70, 2308–2314. [CrossRef]
- 24. Kuhkheil, A.; Naghdi Badi, H.; Mehrafarin, A.; Abdossi, V. Chemical constituents of sea buckthorn (*Hippophae rhamnoides* L.) fruit in populations of central Alborz Mountains in Iran. *Res. J. Pharmacogn. RJP* **2017**, *4*, 1–12.
- Isrigova, T.A.; Selimova, U.A.; Ganakaev, A.I.; Taibova, D.S.; Sannikova, E.Y. Nutritional value of fruit and berry raw material for the production of functional food. *IOP Conf. Series Earth Environ. Sci.* 2022, 979, 012073. [CrossRef]

- Ilhan, G.; Gundogdu, M.; Karlovi'c, K.; Židovec, V.; Vokurka, A.; Ercisli, S. Main Agro-Morphological and Biochemical Berry Characteristics of Wild-Grown Sea Buckthorn (*Hippophae rhamnoides* L. ssp. *caucasica Rousi*) Genotypes in Turkey. *Sustainability* 2021, 13, 1198. [CrossRef]
- 27. Gâtlan, A.-M.; Gutt, G. Sea Buckthorn in Plant Based Diets. An Analytical Approach of Sea Buckthorn Fruits Composition: Nutritional Value, Applications, and Health Benefits. *Int. J. Environ. Res. Public Health* **2021**, *18*, 8986. [CrossRef]
- Raffo, A.; Paoletti, F.; Antonelli, M. Changes in sugar, organic acid, flavonol and carotenoid composition during ripening of berries of three sea buckthorn (*Hippophae rhamnoides* L.) cultivars. *Eur. Food Res. Technol.* 2004, 219, 360–368. [CrossRef]
- Ficzek, G.; Mátravölgyi, G.; Furulyás, D.; Rentsendavaa, C.; Jócsák, I.; Papp, D.; Simon, G.; Végvári, G.; Stéger-Máté, M. Analysis of bioactive compounds of three sea buckthorn cultivars (*Hippophae rhamnoides* L. 'Askola', 'Leikora', and 'Orangeveja') with HPLC and spectrophotometric methods. *Eur. J. Hortic. Sci.* 2019, *84*, 31–38. [CrossRef]
- 30. Ercisli, S.; Orhan, E. Chemical composition of white (*Morus alba*), red (*Morus rubra*) and black (*Morus nigra*) mulberry fruits. *J. Food Chem.* **2007**, *103*, 1380–1384. [CrossRef]
- Zhang, W.; Yan, J.; Duo, J.; Ren, B.; Guo, J. Preliminary study of biochemical constitutions of berry of sea buckthorn growing in Shanxi province and their changing trend. In Proceedings of the International Symposium on Sea Buckthorn (*Hippohae rhamnoides* L.), Xi'an, China, 19–23 October 1989; pp. 96–105.
- Tong, J.C.; Zhang, Z.; Zhao, Y.; Yang, T.K. The determination of the physical-chemical constants and sixteen mineral elements in raw sea buckthorn juice. In Proceedings of the International Symposium on Sea buckthorn (*H. rhamnoides* L.), Xi'an, China, 19–23 October 1989; pp. 132–137.
- 33. Pallavee, K.; Ashwani, M. Sea Buckthorn Juice: Nutritional Therapeutic Properties and Economic Considerations. *Int. J. Pharmacogn. Phytochem. Res.* 2017, *9*, 880–884. [CrossRef]
- 34. Tang, X. Breeding in Sea Buckthorn: Genetic of Berry Yield, Quality and Plant Cold Hardiness; University of Helsinki: Helsinki, Finland, 2002.
- Buya, B.; Zheng, H.Z.; Chung, S.K. Chemical Composition of Mongolian Sea-buckthorn (*Hippophae rhamnoides* L.) Fruits. *Agric. Rex. Bull. Kyungpook Natl. Univ.* 2012, 30, 35–39.
- Shourove, J.H.; Zzaman, W.; Chowdhury, R.S.; Hoque, M.M. Effect of thermal treatment on physicochemical stability and antioxidant properties of locally available underutilized star fruit juice. *Asian Food Sci. J.* 2020, 14, 41–53. [CrossRef]
- 37. Jaśniewska, A.; Diowksz, A. Wide Spectrum of Active Compounds in Sea Buckthorn (*Hippophae rhamnoides*) for Disease Prevention and Food Production. *Antioxidants* 2021, 12, 1279. [CrossRef]
- Chen, J.; Tao, X.Y.; Sun, A.D.; Wang, Y.; Liao, X.J.; Li, L.N.; Zhang, S. Influence of pulsed electric field and thermal treatments on the quality of blueberry juice. *Int. J. Food Prop.* 2014, 17, 1419–1427. [CrossRef]
- 39. Zeb, S. Chemical and Nutritional Constituents of Sea Buckthorn Juice. Pak. J. Nutr. 2004, 3, 99–106.
- 40. Igwemmar, N.C.; Kolawole, S.A.; Imran, I.A. Effect of Heating on Vitamin C Content of Some Selected Vegetables. *Int. J. Sci. Technol. Res.* 2013, *2*, 209–212.
- 41. Munyaka, A.W.; Makule, E.E.; Oey, I.; Van Loey, A.; Hendrickx, M. Thermal stability of L-ascorbic acid and ascorbic acid oxidase in broccoli (*Brassica oleracea* var. *italica*). *J. Food Sci.* **2010**, *75*, C336–C340. [CrossRef]
- 42. Rickman, J.C.; Barrett, D.M.; Bruhn, C.M. Review Nutritional comparison of fresh, frozen and canned fruits and vegetables. Part 1. Vitamins C and B and phenolic compounds. *J. Sci. Food Agric.* **2007**, *7*, 930–944. [CrossRef]
- Seglina, D.; Karklina, D. The dynamics of Vitamin C and total carotenes content in peasteurized sea-buckthorn juice. In Proceedings of the International Scientific Conference: Research for Rural Development, International Scientific Conference: Research for Rural Development, Jelgava, Latvia, 19–22 May 2005; pp. 205–207.
- 44. Seglina, D.; Karklina, D.; Ruisa, S.; Krasnova, I. The effect of processing on the composition of sea buckthorn juice. *J. Fruit Ornam. Plant Res.* **2006**, *14*, 257.
- Criste, A.; Urcan, A.C.; Bunea, A.; Pripon Furtuna, F.R.; Olah, N.K.; Madden, R.H.; Corcionivoschi, N. Phytochemical composition and biological activity of berries and leaves from four Romanian sea buckthorn (*Hippophae rhamnoides* L.) varieties. *Molecules* 2020, 25, 1170. [CrossRef]