

Effect of grapefruit seed extract on polyphenol, anthocyanin and colour properties of aronia juice during heat treatment

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ABSTRACT

This study examined the effect of grapefruit seed extract (GSE) (0%–3%) on the stability of total phenolic content (TPC), anthocyanins, and colour in aronia juice under heat treatments (60 °C, 80 °C for up to 120 min). TPC and anthocyanins were measured using spectrophotometry and HPLC. The highest TPC (8545.02 ± 355.55 GAE mg L⁻¹) was measured in aronia juice with 1% GSE after 60 min at 80 °C. The highest anthocyanin retention (3178.75 mg L⁻¹) was detected in the sample with 1% GSE after 5 min at 60 °C. Cyanidin-3-galactoside was the most abundant anthocyanin, followed by cyanidin-3-xyloside and cyanidin-3-arabinoside. Cyanidin-3-arabinoside showed the lowest heat stability, while cyanidin-3-galactoside was relatively more stable. This study, the first to evaluate anthocyanin half-life values in chokeberry juice, suggests that 1% GSE enhances colour stability during heat treatment, supporting its use as a natural food additive.

KEYWORDS

aronia, anthocyanin, grapefruit seed extract, heat treatment, degradation kinetic

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INTRODUCTION

Natural pigments are widely used in the food industry as colouring agents. Anthocyanins are preferred as the replacement of synthetic food colourants, because they are water-soluble plant pigments, they have different, attractive colours, and possible health benefits with antioxidant effect (Cruz et al., 2021). Moreover, due to the health-conscious consumer habits, using natural ingredients in the foods is an increasing trend (Martins et al., 2016).

However, the commercial use of anthocyanins is limited due to their low stability. They are very sensitive to light, temperature, oxygen, salt stress, enzymes or acids and bases present in the matrix (Mbarki et al., 2018; Chen et al., 2023).

The most commonly applied method for prolonging the shelf-life of the food product is heat treatment, which can be applied at temperatures between 50 and 180 °C, depending on the manufacturer. Anthocyanins can rapidly degrade while exposed to heat, due to different mechanisms such as glycosylation, nucleophilic attack of water, cleavage and polymerization, which can have a great impact on the colour properties and the nutritional values as well (Enaru et al., 2021; Rodriguez-Amaya, 2019). However, it should be taken into account that both the colour and the stability of anthocyanin molecules depend on the substitution of coloured aglycones and their forms (acylated or non-acylated) (Alappat and Alappat, 2020).

There are different studies focusing on the thermal effect on pigment stability in fruits or vegetables. Pigments of grumixama berries (*Eugenia brasiliensis* Lam.) above 30 °C degraded by 50% after 10 h at 60 °C, 1.9 h at 80 °C and 0.96 h at 100 °C (Modesto Junior et al., 2023). The total anthocyanin of black goji berry and purple sweet potato was significantly reduced after heat treatment, nearly 30% loss of anthocyanin content was observed at 80 °C 30 min treatment (Gamage and Choo, 2023). However, the anthocyanins in açai pulp were relatively stable because the loss did not reach 1%, confirming its high thermal stability (Marangoni Júnior et al., 2020). In the case of red-purple-flesh potato, total anthocyanin content was generally higher at lower pasteurization (65 °C) after 5 min, however, higher values were also observed at 75 °C after 5 min for some varieties (Tkaczyńska et al., 2024a,b).

Black chokeberry (*Aronia melanocarpa* (Michx.) Elliot) can be a good source of natural food colourant in the food industry due to its high anthocyanins content (Klisurova et al., 2019; Catalkaya et al., 2022). Black chokeberry is a plant indigenous to North America that was introduced to Europe nearly a century ago. This shrub belongs to the Rosaceae family (Zhang et al., 2021). Black chokeberry and its processed products are highly regarded for their abundance of bioactive compounds that provide exceptional biological and nutritional value including flavonoids (primarily anthocyanins, proanthocyanidins, flavanols, and flavonols), phenolic acids (such as chlorogenic, neochlorogenic, and caffeic acids), and tannins. These polyphenolic compounds are the predominant components of black *A. melanocarpa* berries, with concentrations ranging from 10.6 to 197.0 mg g⁻¹ in dry weight (dw) and 6.9–25.6 mg g⁻¹ in fresh weight (fw), expressed as gallic acid equivalents (Piras et al., 2024; Oziembłowski et al., 2022). The deep blue colour of chokeberry fruit results from its high anthocyanin content, which can vary between 307 and 1,480 mg/100 g fw, and is primarily composed of cyanidin derivatives, including cyanidin-3-glucoside, 3-galactoside, 3-xyloside, and 3-arabinoside. Additionally, a minor fraction of anthocyanins consists of pelargonidin-3-galactoside and pelargonidin-3-arabinoside (Sidor and Gramza-Michałowska, 2019; Oszmiański and Wojdyło, 2005; Klisurova et al., 2019; Jurendić and Ščetar, 2021). Furthermore, the potent antioxidant properties of chokeberries

and their derived products are primarily attributed to their high polyphenol content. The antioxidant activity, measured by the ability to scavenge DPPH and ABTS radicals, varied among fresh fruit, juice, and pomace. Fresh fruit exhibited values of 279.38 (DPPH) and 439.49 (ABTS) μM Trolox/100 g dw, while juice showed lower activity at 127.45 (DPPH) and 314.05 (ABTS). Pomace demonstrated the highest values, with 301.89 (DPPH) and 779.58 (ABTS), indicating its strong antioxidant potential (Jurendić and Ščetar, 2021; Oszmiański and Wojdyło, 2005).

The bitterness and tartness of chokeberry are primarily attributed to its high content of polyphenols, particularly densely polymerized proanthocyanins. These procyanidin have a strong affinity for proteins, leading to their denaturation, which in turn influences the perception of tartness, causing a sensation of dryness in the mouth and throat (Trenka et al., 2020; Deng et al., 2021). Although fresh unprocessed aronia fruits are seldom consumed because it has an astringent and bitter flavour, chokeberries have emerged as a substantial industrial crop, playing a vital role in the manufacturing of juices, nectars, wines, jams, and notably serving as natural foodgrade colouring agents (Sidor and Gramza-Michałowska, 2019; Klisurova et al., 2019).

To reduce the decreases of natural colourants during processing, manufacturers nowadays try to use natural materials. Anthocyanin stability can improve through the copigmentation process with different compounds such as amino acids, organic acids, flavonoids and metals among others (Li et al., 2019). Some antioxidants, mostly ascorbic acid, are commonly added to juices to prevent oxidation of natural pigments. However, it has been reported to negatively impact anthocyanin stability, accelerating their degradation especially in the oxygen presence. Although the degradation mechanism is unclear as of yet, it was observed that the presence of ascorbic acid increases the rate of anthocyanin decomposition (Levy et al., 2019; Enaru et al., 2021). An alternative can be the use of Grapefruit Seed Extract (GSE), derived from *Citrus paradisi* L. and traditionally recognized for its antimicrobial properties. GSE is used as an antimicrobial agent in foods (Zayed et al., 2021; Magurano et al., 2021) because it has high concentration of bioactive compounds and antioxidants including polyphenols, ascorbic acid, tocopherol, flavonoids (naringenin), limonoids (limonin) and trace elements (Kim et al., 2021; Roy and Rhim, 2021). Furthermore, GSE can be used in edible packaging film or as a coating material (Roy et al., 2023; Jha, 2020).

The aim of this study was to reveal the effect of GSE on total polyphenol content, anthocyanin concentration and colour properties during heating at 60 °C and 80 °C for 5, 10, 30, 60, 120 min. Furthermore, to determine some kinetic parameters (rate constant and half-life time) of the different anthocyanin compounds during the experiment. As we know this is the first study that evaluated the heat degradation kinetic of individual anthocyanin molecules of chokeberry juice.

MATERIAL AND METHODS

Plant and reagents material

Aronia (variety of Nero) was collected near Lajosmizse (47°02'44.4"N 19°35'14.8"E), Hungary in 2022 in full ripening stage. The sample was stored at -18 °C until further processing and analysis.

The Grapefruit seed extract (GSE) liquid concentrate (33%), 2 fl oz (59 mL) was purchased from NutriBiotic company (Lakeport, CA, USA), which is the most concentrated commercially available GSE product. In the case of the total polyphenol content TPC measurement, methanol, Folin-Ciocalteu phenol reagent, anhydrous sodium carbonate (Na_2CO_3) and for the anthocyanin measurement, the HPLC Gradient-grade acetonitrile and water were obtained from Sigma Aldrich (St. Louis, MO, USA). Formic acid was purchased from Fluka (Buch, Switzerland). Gallic acid, cyanidin-3-arabinoside, cyanidin-3-glucoside, and cyanidin-3-galactoside were obtained from Merk (Darmstadt, Germany).

Samples preparation

Aronia juice was prepared using laboratory pressing machine. The GSE was added to the prepared juice in given percentages: 0%, 1%, 2%, and 3%. The tubes were placed in the water bath (Foss Tecator 1024, Hillerød, Denmark) at controlled different temperature (60 °C and 80 °C) at different durations (5, 10, 30, 60, 120 min). After sampling, tubes were cooled in ice water to avoid the further thermal degradation. Subsequently, the samples underwent analysis for total polyphenol content (TPC), colour parameters, and individual anthocyanin concentration using HPLC method. All treatments and measurements were done in triplicate.

Total phenolic content (TPC)

Total polyphenol concentration was measured using Singleton and Rossi (1965) colorimetric method. The absorbance of the sample solution was detected at 760 nm. The results are expressed in mg gallic acid equivalents per L (mg GAE/L).

High-performance liquid chromatography (HPLC)

For the determination of individual anthocyanin molecules, Kavela et al. (2023) chromatographic method was used. The Shimadzu HPLC system (Shimadzu Corporation, Kyoto, Japan) was applied with a 150×4.6 mm, C18, 3 μm particle size column (Phenomenex Torrance, California, USA). In the elution, 0.5% (V/V) formic acid in high purity water (mobile phase A) and 0.5% formic acid in acetonitrile (mobile phase B) were used with 0.5 mL min^{-1} flow rate. The total gradient program was 30 min and started with 5% solvent B. Solvent B was increased linearly to 25% in 5 min and it was increased further to 100% from 5 to 10 min. Finally, 100% solvent B was held constant for another 5 min, then it was immediately reduced to 5% and the system was run for an additional 10 min at the initial solvent composition. The HPLC measurement was done. The retention times of the different anthocyanin standards were used for the qualification of the peaks. Quantification was done using four-point external calibration, and the results were given in mg L^{-1} .

Calculation of kinetic parameters of anthocyanins

In most cases anthocyanin degradation occurs based on the first-order kinetics under both aerobic and anaerobic conditions. The two important kinetic parameters are the first-order reaction rate constant (k) and half-life ($t_{1/2}$). These were calculated based on the following equations (Kirca and Özkan, 2007):

$$\ln\left(\frac{\gamma_t}{\gamma_0}\right) = -k * t$$

$$t_{1/2} = -\ln 0.5 / k$$

where γ_0 is the initial anthocyanin concentration and γ_t is the anthocyanin concentration after time t (min) of heating at a given temperature (depending on the given experiment).

Colour measurement

Colour parameters were measured according to C.I.E.LAB system using a digital colorimeter (Konica Minolta CR 410, Minolta Canada Inc.). The colour difference (ΔE^*) was calculated based on this equation:

$$\Delta E_{1;2}^* = \sqrt{(L_1^* - L_2^*)^2 + (a_1^* - a_2^*)^2 + (b_1^* - b_2^*)^2}$$

where L_1^* and L_2^* are lightness of sample and control, respectively; a_1^* and a_2^* are redness of sample and control, respectively; b_1^* and b_2^* are yellowness of sample and control, respectively.

The colour difference value (ΔE^*) was evaluated by Lukács (1982) as follows: 0–0.5 = not noticeable, 0.5–1.5 = slightly noticeable, 1.5–3.0 = noticeable, 3.0–6.0 = clearly visible, and 6.0–12.0 = great visibility.

Statistical analysis

The obtained results underwent statistical analysis using IBM SPSS statistics software version 27 (IBM Corp., New York, NY 10022, USA, 2020), and mean differences between factors were assessed using two-way analysis of variances (ANOVA). Significant differences were considered if P -value < 0.05.

RESULTS AND DISCUSSION

Total polyphenol content (TPC)

The Total Phenolic Content (TPC) of aronia juice samples treated with varying concentrations of GSE (0%, 1%, 2%, 3%) and subjected to different heat treatments (60 °C, 80 °C) at various durations (5, 10, 30, 60, 120 min) is shown in Figs 1 and 2.

Based on the results, at 60 °C the highest values of TPC were mostly observed in samples treated with 0% GSE, whereas the highest TPC value was recorded after 60 min of heat treatment 7906.79 ± 354.95 GAE mg L⁻¹. The 0% of GSE indicated a total yield of TPC significantly higher ($P < 0.05$) compared to all other concentrations across each time, except at 120 min, where 1% of GSE showed significantly higher results (6639.81 ± 241.84 GAE mg L⁻¹) compared to 0%, 2%, 3% (4519.75 ± 210.32 ; 5082.15 ± 293.12 ; 4200.63 ± 195.25 GAE mg L⁻¹, respectively). Moreover, there was no significant difference ($P > 0.05$) between the samples treated with 0% GSE across different times (7720.37 ± 75.82 ; 7856.24 ± 482.04 ; 7590.83 ± 296.32 ; 7906.79 ± 354.95 GAE mg L⁻¹, at 5, 10, 30, 60 min); except at 120 min of heat treatment (4519.74 ± 210.31 GAE mg L⁻¹). The 1% of GSE showed values ranging from 6453.40 ± 109.85 to

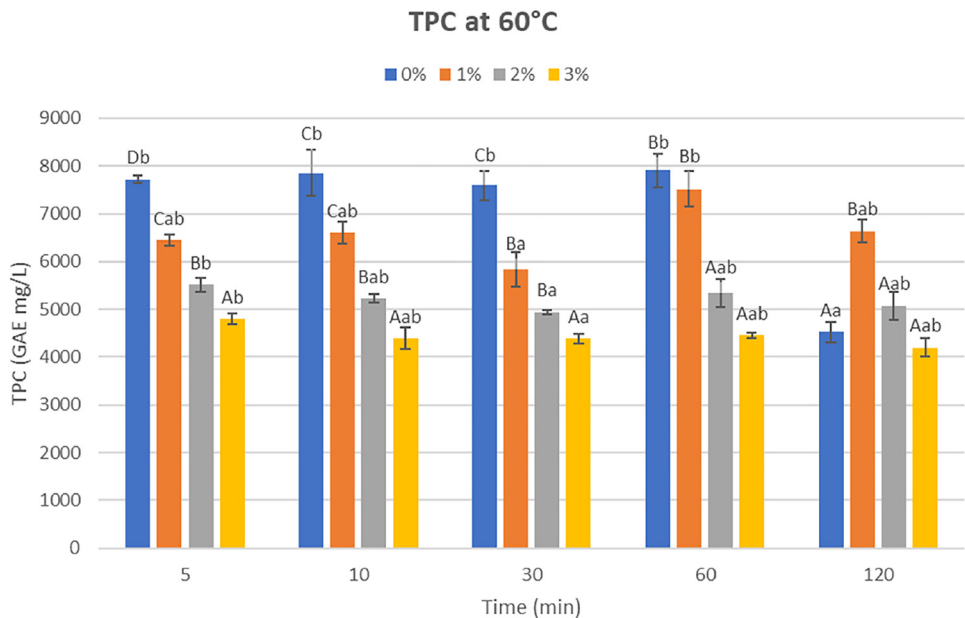


Fig. 1. TPC content of aronia juice samples incorporated with different concentrations of GSE at 60 °C in different times (5, 10, 30, 60, 120 min).

Big letters indicate significant differences between different concentrations at each time. Small letters indicate significant differences of each concentration at different times. Results are indicated in mean values, and the error bars represent the standard deviation

7518.17 ± 374.65 GAE mg L⁻¹, followed by 2% of GSE ranging from 5515 ± 142.28 to 5344.39 ± 290.66 GAE mg L⁻¹. The 3% GSE showed significantly the lowest (*P* < 0.05) TPC results at 80 °C. The highest TPC values were observed in the samples treated with 1% of GSE ranging from 8238.55 ± 270.82 to 8440.76 ± 133.71 GAE mg L⁻¹. The 1% GSE indicated a total yield of TPC significantly higher (*P* < 0.05) compared to all other concentrations across each time. Where the highest TPC value was recorded after 60 min of heat treatment 8545.02 ± 355.55 GAE mg L⁻¹ with 1% of GSE. Furthermore, there is no significant difference (*P* > 0.05) between the TPC values of the samples treated with 1% of GSE across different durations of heat treatment (8238.54 ± 270.82; 8440.75 ± 209.17; 8434.43 ± 323.43; 8545.02 ± 355.54; 8440.75 ± 133.71 GAE mg L⁻¹ at 5, 10, 30, 60, 120 min, respectively). This was followed by 0% (ranging from 6093.21 ± 137.47 to 8484.99 ± 880.72 GAE mg L⁻¹). Then 2% (ranging from 6788.31 ± 321.76 to 7973.17 ± 20 GAE mg L⁻¹) and lastly, 3% of GSE (ranging from 6832.54 ± 194.33 to 7189.57 ± 177.08 GAE mg L⁻¹).

Total phenolics are generally sensitive to high temperatures and heat treatment, in literature, [Ghafoor et al. \(2019\)](#) reported that both the total phenolic content and antioxidant activity in plum and mahaleb were decreased during heating. In a related study, [Putsakum et al. \(2023\)](#) investigated the effects of thermosonication (TS) (a method combining heat and ultrasound), on blackberry juice, finding that TPC was highest at lower temperatures and shorter treatment

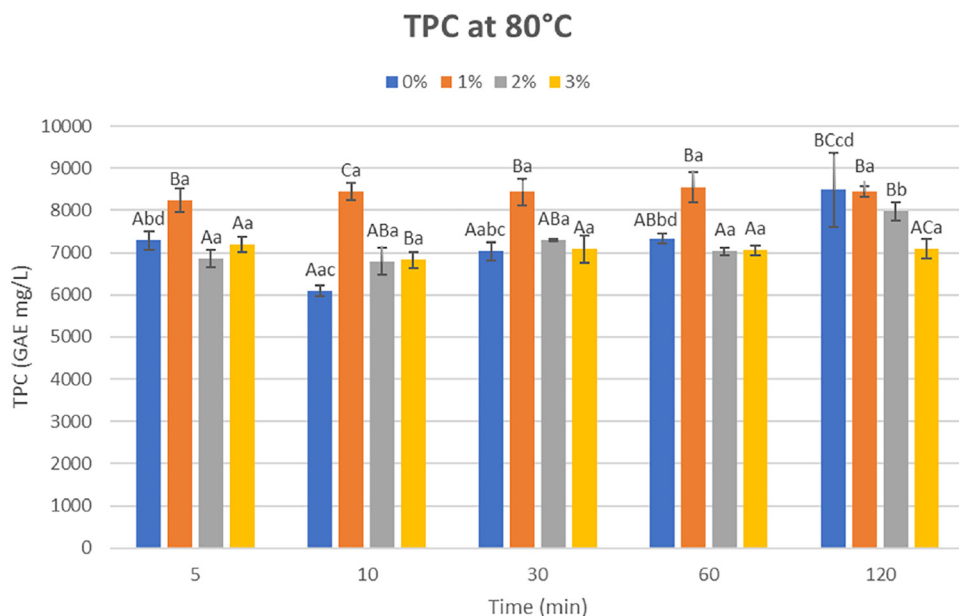


Fig. 2. TPC content of aronia juice samples incorporated with different concentrations of GSE at 80 °C in different times (5, 10, 30, 60, 120 min).

Big letters indicate significant differences between different concentrations at each time. Small letters indicate significant differences of each concentration at different times. Results are indicated in mean values, and the error bars represent the standard deviation

times. Similarly, [Dars et al. \(2019\)](#) observed that high-temperature TS treatments in mango juice significantly impacted phenolic content, primarily due to degradative or oxidative effects.

From our results, we found that adding 1% of grapefruit seed extract to the aronia juice consistently increased the TPC values, particularly at 80 °C compared to 60 °C, reflecting both increased phenolic compounds at high temperatures and GSE's protective effect against thermal degradation. Furthermore, the highest TPC results were observed after 60 min of heat treatment with 1% GSE at both temperatures, indicating that lower concentration of GSE is optimal for preserving and stabilizing polyphenols during thermal processing, while higher concentration may lead to polyphenolic instability. Although GSE substance is traditionally recognized for its antimicrobial effect and its effective use in packaging ([Kanmani and Rhim 2014](#); [Kim et al., 2021](#); [Wang et al., 2021](#)), these findings may contribute to expand its application to enhance and protect the nutritional quality of processed aronia juice.

Anthocyanin concentration

In the aronia juice, three main anthocyanin compounds could be detected in all of the examined samples (Fig. 3). The most abundant compound (peak 1) was identified as cyanidin-3-galactoside, which eluted first. Peak 2 was the cyanidin-3-arabinoside, while cyanidin-3-xyloside was detected as peak 3. These are in accordance with some studies ([Klisurova et al., 2019](#); [Oszmiański and Lachowicz, 2016](#); [L. Zhang et al., 2012](#)).

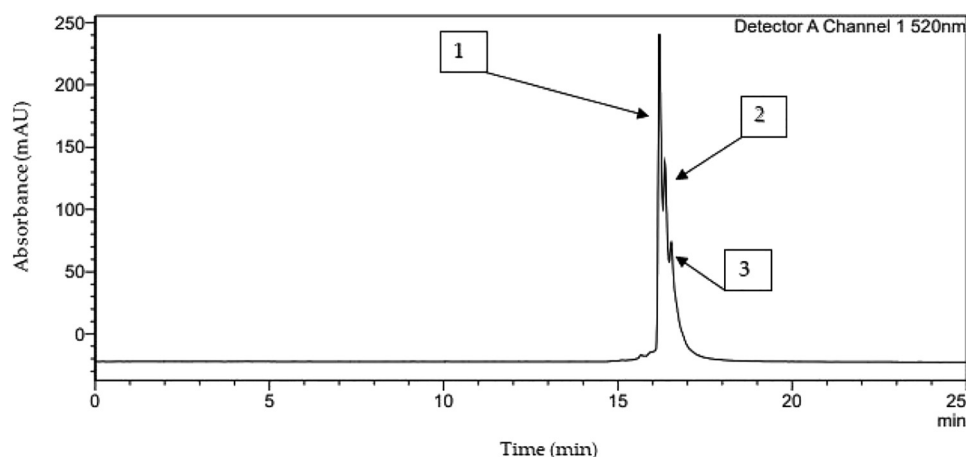


Fig. 3. UV/Vis chromatogram of aronia juice detected at 520 nm. Peak 1 = cyanidin-3-galactoside, 2 = cyanidin-3-arabinoside and 3 = cyanidin-3-xyloside

According to our data, the heat treatment at both temperatures led to a significant decrease in the levels of anthocyanin components, with the highest degradation occurring in samples without GSE. In contrast, samples treated with GSE showed less degradation, as showed in the figures. Based on the results, the average sum of individual anthocyanins ranges from 3178.75 to 1801.96 mg L⁻¹. The highest total anthocyanins yield 3178.75 mg L⁻¹ measured with HPLC analysis was obtained from samples treated with 1% GSE at 60 °C after 5 min of heat treatment. This was followed by 3173.16 mg L⁻¹ from samples treated with 1% GSE at 60 °C after 60 min. The 3rd highest yield, 3170.06 mg L⁻¹ was obtained from samples treated with 2% GSE also at 60 °C after 10 min of heat treatment. The lowest total anthocyanins yield 1801.96 mg L⁻¹ was obtained from samples that had 0% GSE after 120 min of heat treatment at 60 °C. At 80 °C, although differences among samples treated with varying GSE concentrations were minimal, the presence of GSE effectively stabilized the sum of individual anthocyanins across different time points and concentrations. Furthermore, the concentration of cyanidin compounds slightly increased with the temperature (60 °C and 80 °C) and with the addition of the GSE substance. This suggests that new anthocyanins can be formed during heating and that anthocyanin compounds were more stable under these conditions. This effect may be the copigmentation, where the interaction between the GSE polyphenols and aronia anthocyanins results in the formation of new, more stable anthocyanins during heat treatment. Similar findings were reported by [Zhu et al. \(2020\)](#), who observed that the copigmentation with three different phenolic acids was effective in stabilized bayberry anthocyanins in thermal treatment. Other studies have also documented similar results, including [Xue et al. \(2019\)](#), [Klisurova et al. \(2019\)](#), [Fan et al. \(2019\)](#). Copigmentation technique is widely used in the food industry to enhance the colour, helping to preserve the intensity of natural colours by stabilizing the anthocyanin compounds ([Gençdağ et al., 2022](#); [Fan et al., 2019](#); [Tan et al., 2021](#)).

In the case of anthocyanins, the most abundant and stable pigment compound is cyanidin-3-galactoside at both temperatures 60 °C and 80 °C, its concentration ranged from

654.66 ± 2.73 to 1208.1 ± 4.10 mg L⁻¹ at 60 °C, and from 689.01 ± 6.78 to 1172.27 ± 7.67 mg L⁻¹ at 80 °C. Followed by cyanidin-3-xyloside, which ranged from 618.10 ± 16.71 to 1141.84 ± 8.19 mg L⁻¹ at 60 °C, and from 639.35 ± 2.82 to 1122.01 ± 19.56 mg L⁻¹ at 80 °C. The least stable and abundant compound is cyanidin-3-arabinoside, with values ranging from 529.20 ± 22.26 to 889.29 ± 10.12 mg L⁻¹ at 60 °C, and from 551.55 ± 1.85 to 889.29 ± 10.12 mg L⁻¹ at 80 °C (Figs 4 and 5). The statistical evaluation of the anthocyanin concentration can be seen in Tables S1 and S2 for both temperatures. Based on our results the addition of 1% GSE appears to be the most effective concentration for enhancing colour stability under heat treatment.

Anthocyanin stability of aronia juice during heat treatment

As regards the degradation of total anthocyanins without GSE, it strongly depended on temperature (Tables 1 and 2). Results of half-life time ($t_{1/2}$) confirm this observation, as the $t_{1/2}$ data of TA at 60 °C were higher than at 80 °C, which means the pigments were more stable to heat at lower temperature. This is in agreement with the study of Tkaczyńska et al. (2024a,b), where pasteurization of purple-fleshed potato varieties showed better pigment retention at lower temperatures. Similarly, Askin et al. (2022) observed that the pasteurization (95 °C at 3 min) significantly led to the decrease of pigment concentration of both unclarified (39–51%) and clarified (11–18%) black mulberry juice. Furthermore, a study by Loypimai et al. (2016) examined the thermal and pH degradation kinetics of anthocyanin pigments in a natural food colorant derived from black rice bran, finding that anthocyanins and colour stability were better preserved at lower temperatures (60 °C). Similarly, Verbeyst et al. (2010) reported that anthocyanin degradation in strawberry paste is highly temperature-dependent, with higher

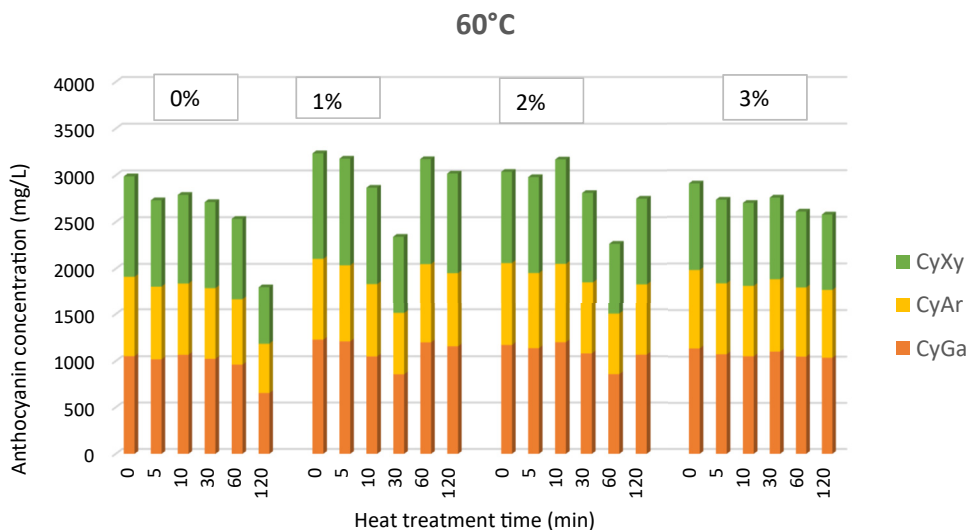


Fig. 4. Individual anthocyanin concentration of aronia juice during heat treatment. CyXy = cyanidin-3-xyloside, CyAr = cyanidin-3-arabinoside, CyGa = cyanidin-3-galactoside

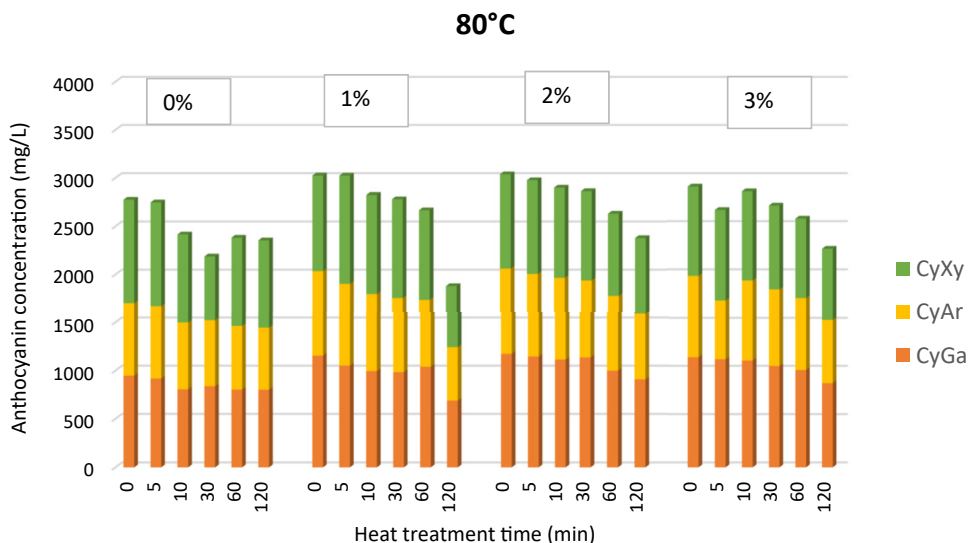


Fig. 5. Individual anthocyanin concentration of aronia juice during heat treatment. CyXy = cyanidin-3-xyloside, CyAr = cyanidin-3-arabinoside, CyGa = cyanidin-3-galactoside

temperatures accelerating the decline in anthocyanin concentration. These findings reinforce the widely reported trend that increased heat exposure accelerates anthocyanin degradation, reducing their stability in food matrices.

Addition of GSE in different ratios (1%, 2%, 3%) can modify the anthocyanin stability at the different temperatures. The highest half-life time values were observed in the case of samples with 3% GSE, which predicts that addition of 3% GSE can improve the stability of pigments in aronia juice during heat treatment. This means that degradation caused by long lasting high temperatures can be reduced using GSE extract as a copigmentation agent. The ability of GSE to stabilize anthocyanins can be attributed to its high polyphenol content, which likely interacts with anthocyanins through copigmentation mechanisms, thereby reducing their susceptibility to degradation.

The heat sensitivity of the individual cyanidin-based anthocyanin component was different and showed differences at the treatment temperatures. Cyanidin-3-arabinoside (CyAr) had lower half-life time values at almost each heat treatment time and GSE% at 60 °C (Tables 1 and 2). Cyanidin-3-galactoside (CyGa) seemed more stable, while cyanidin-3-xyloside (CyXy) was the least susceptible to the heat degradation. This is in agreement with the study of bilberry extract heating (Yue and Xu, 2008), where the thermal stability of anthocyanin with arabinoside was lower than the anthocyanin with glucoside or galactoside. As regard the different GSE ratios, the protective effect in lower concentration (1%) was observed in the case of long-term heating, more exactly after 60 and 120 min especially at 60 °C. In the case of the higher temperature (80 °C), 2–3% GSE can protect the anthocyanins from the degradation during the treatment.

Table 1. Kinetic parameters for the heat degradation of anthocyanins at 60 °C

		60 °C							
GSE ratio	Heat treatment time (min)	CyGa		CyAr		CyXy		TA	
		k·10 ³ /min ⁻¹	t _{1/2} /h	k·10 ³ /min ⁻¹	t _{1/2} /h	k·10 ³ /min ⁻¹	t _{1/2} /h	k·10 ³ /min ⁻¹	t _{1/2} /h
0%	5	-0.007	1.70	-0.017	0.69	-0.030	0.38	-0.003	3.58
	10	0.000	33.16	-0.010	1.10	-0.013	0.92	0.000	11.13
	30	-0.001	12.39	-0.004	3.09	-0.005	2.23	-0.001	15.03
	60	-0.002	7.62	-0.003	3.66	-0.004	3.10	-0.002	7.56
	120	-0.004	2.91	-0.004	2.86	-0.005	2.50	-0.004	3.20
1%	5	-0.003	3.80	-0.012	0.98	0.002	-6.21	0.010	-1.16
	10	-0.010	1.15	-0.011	1.02	-0.009	1.26	-0.003	3.49
	30	-0.012	0.97	-0.010	1.18	-0.010	1.12	-0.009	1.35
	60	0.000	28.36	-0.001	21.91	0.000	14.82	0.001	-14.49
	120	0.000	24.32	-0.001	13.63	0.000	23.46	0.000	10.54
2%	5	-0.006	2.01	-0.017	0.66	0.010	-1.14	-0.004	3.07
	10	0.002	-5.16	-0.004	2.84	0.014	-0.85	0.004	-2.71
	30	-0.003	4.36	-0.005	2.45	-0.001	17.25	-0.003	4.48
	60	-0.005	2.22	-0.005	2.19	-0.004	2.78	-0.005	2.37
	120	-0.001	15.10	-0.001	9.10	-0.001	22.32	-0.001	13.96
3%	5	-0.011	1.02	-0.020	0.58	-0.007	1.67	-0.012	0.93
	10	-0.008	1.49	-0.011	1.08	-0.004	2.70	-0.007	1.55
	30	-0.001	11.45	-0.003	4.24	-0.002	5.94	-0.002	6.43
	60	-0.001	8.73	-0.002	5.46	-0.002	5.28	-0.002	6.33
	120	-0.001	15.07	-0.001	9.53	-0.001	10.07	-0.001	11.39

CyGa: cyanidin-3-galactoside, CyAr: cyanidin-3-arabinoside, CyXy: cyanidin-3-xyloside, TA: total anthocyanin, k: first order reaction rate constant, t_{1/2}: half-life time of anthocyanin.

Colour measurement

Figure 6 shows the colour difference values (ΔE*) values, calculated from L*, a* and b* between the control and the samples with different concentrations of GSE at both temperatures (60 °C and 80 °C) for different durations (5, 10, 30, 60, and 120 min). The colour differences were evaluated according to the noticeability criteria outlined in section 2.7. The results show that at 60 °C, there was a slightly noticeable difference between the control samples and those treated with 1% GSE after 60 and 120 min of heat treatment between the control and 2% treated samples after 30, and 120 min. Similarly, a slight noticeable difference was observed between the control and the 3% treated samples at 5, 10, 60 min. At 80 °C, slight differences were observed between the control and most GSE treated samples across all concentrations, except for the samples treated with 3% GSE after 5 min, also between the control and the samples treated with 2% GSE after 30 and 120 min of heat treatment. A more noticeable difference occurred with 1% of GSE treated samples after 5 min, also the 3% treated samples after 120 min of heat treatment.

Table 2. Kinetic parameters for the heat degradation of anthocyanins at 80 °C

		80 °C							
GSE ratio	Heat treatment time (min)	CyGa		CyAr		CyXy		TA	
		k·10 ³ /min ⁻¹	t _{1/2} /h	k·10 ³ /min ⁻¹	t _{1/2} /h	k·10 ³ /min ⁻¹	t _{1/2} /h	k·10 ³ /min ⁻¹	t _{1/2} /h
0%	5	−0.007	1.30	0.000	42.50	0.001	−14.24	0.00	4.20
	10	−0.016	0.54	−0.009	1.00	−0.016	0.55	−0.01	0.62
	30	−0.004	2.11	−0.003	2.70	−0.016	0.54	−0.01	1.09
	60	−0.003	3.14	−0.002	3.87	−0.003	3.33	0.00	3.39
	120	−0.001	6.24	−0.001	6.47	−0.001	6.25	0.00	6.30
1%	5	−0.019	0.46	−0.007	1.30	0.025	−0.35	0.00	171.30
	10	−0.015	0.58	−0.009	0.95	0.004	−2.35	−0.01	1.26
	30	−0.005	1.62	−0.004	1.98	0.001	−7.12	0.00	3.10
	60	−0.002	4.95	−0.004	2.21	−0.001	8.68	0.00	4.14
	120	−0.004	2.01	−0.004	2.23	−0.004	2.38	0.00	2.19
2%	5	−0.004	2.03	−0.005	1.76	−0.004	2.31	0.00	2.15
	10	−0.005	1.72	−0.003	2.66	−0.006	1.47	0.00	1.86
	30	−0.001	8.65	−0.003	2.74	−0.002	3.88	0.00	4.39
	60	−0.003	3.27	−0.002	4.21	−0.002	3.53	0.00	3.62
	120	−0.002	4.09	−0.002	4.12	−0.002	4.51	0.00	4.25
3%	5	−0.004	2.24	−0.064	0.14	0.002	−4.49	−0.02	0.50
	10	−0.003	2.93	−0.001	6.37	0.000	20.18	0.00	5.14
	30	−0.003	3.11	−0.002	4.70	−0.002	3.78	0.00	3.68
	60	−0.002	4.22	−0.002	4.33	−0.002	4.31	0.00	4.28
	120	−0.002	3.88	−0.002	4.21	−0.002	4.45	0.00	4.14

CyGa: cyanidin-3-galactoside, CyAr: cyanidin-3-arabinoside, CyXy: cyanidin-3-xyloside, TA: total anthocyanin, k: first order reaction rate constant, t_{1/2}: half-life time of anthocyanin.

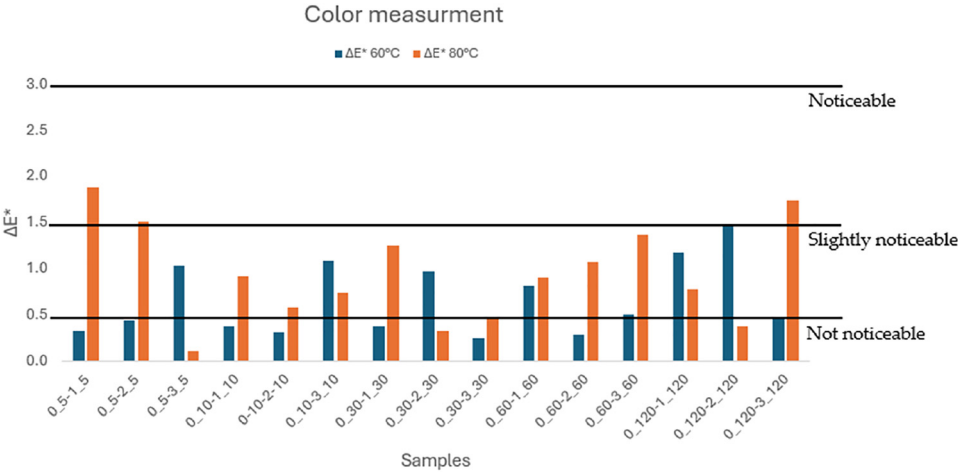


Fig. 6. Colour differences between the control samples (0% GSE) and the treated samples containing 1%, 2%, and 3% GSE at various temperatures (60 °C and 80 °C) and time intervals (5, 10, 30, 60, and 120 min)

CONCLUSION

Aronia melanocarpa fruit is mainly used for processing due to its astringent taste; therefore, several technological steps affect different bioactive compounds during production, like heat treatment and pasteurization. Results of this study showed that 1% GSE was particularly effective in enhancing TPC, especially at 80 °C after 60 min, suggesting that moderate GSE levels stabilize polyphenols under heat. GSE also reduced anthocyanin loss, with cyanidin-3-galactoside being the most stable anthocyanin pigment, and likely promoted copigmentation effects that protected the anthocyanins pigments. In terms of colour stability, GSE at 1% preserved the color of aronia juice, more than higher GSE concentrations, particularly under prolonged heating. These findings support the use of GSE as a natural additive to improve nutritional quality and color retention. However, further examination is necessary to explore the copigmentation effect of GSE in more detail to understand the interaction and its role in pigment stabilization. The results of this study can be used in the food industry where aronia juice as a food colourant is applied for high temperature extruded or baked foods to maximally maintain, polyphenols, anthocyanins and colour during processing.

SUPPLEMENTARY MATERIAL

Supplementary data to this article can be found online at <https://doi.org/10.1556/446.2025.00169>.

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