

Original article

Effect of Two Different Blueberry (*Vaccinium Corymbosum* L.) Juice Processing Methods on Polyphenolic and Anthocyanin Contents

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Abstract

Blueberry juice has become a popular beverage due to its nutritional value, including vitamins, minerals, and antioxidants. This study examined the effects of pressing, mashing, enzymatic treatment, and pasteurization on the anthocyanin content, color, and polyphenolic composition of blueberry juice (BJ). Enzymatic treatment caused a significant reduction (30%) in anthocyanin (ACN) content in Direct Juice Extraction (DJE), whereas Mash Treatment Processing (MTP) led to a significant increase. Overall, ACN levels were higher in treated samples after each processing step in MTP, while polyphenolic levels showed a slight increase in both DJE and MTP. However, both ACNs and polyphenolics decreased after pasteurization. BJ is effective for processing fresh juice. Additionally, throughout the processing stages, ACN, phenolic content, and antioxidant activity were higher, and juice yield was greater in comparison to both methods.

Keywords: Anthocyanins, Blueberry, Depectinization, Polyphenols.

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INTRODUCTION

Blueberries have gained popularity in recent years and have been incorporated into various food products, such as baked goods, beverages, and snacks. The increasing demand for blueberries, driven by their nutritional value, has led to a rise in production and crop value over the past decade. Blueberries contain common antioxidants such as vitamin C, along with a significant number of phytochemicals, particularly phenolic compounds. Among these, anthocyanins (ACNs), a specific group of phenolics, are responsible for the characteristic blue-purple color of blueberries and blueberry-based products (Kalt et al. 2001; Howard, Clark and Brownmiller 2003).

ACNs are key quality indicators for red fruits and their juices. The extraction efficiency and yield of blueberry juice (BJ) are highly dependent on the juice extraction method, particularly the enzymatic degradation of cell walls (Weber and Larsen 2017). Furthermore, the potential health benefits of blueberries and their products, such as juice, are closely linked to their high antioxidant capacity, which strongly correlates with their ACN and total phenolic content (TPC) (Lee and Wrolstad 2004). However, the ACN and TPC content of BJ is significantly influenced by processing conditions and methods. It is well known that depectinization, clarification, and pasteurization have varying effects on ACN levels in different fruit juices (Sadilova, Stintzing and Cale 2006; Türkyılmaz, Yemiş and Ozkan 2012; Dereli et al. 2015; Perreault et al. 2021). Therefore, the impact of processing on ACN and TPC levels in each fruit juice should be thoroughly investigated.

MATERIALS and METHODS

Material

Blueberries (*Vaccinium corymbosum*, Bluegold) were obtained from the same orchard in Hayrat, Trabzon. They were transported in a refrigerated vehicle at 4 °C immediately after harvest to the Food Engineering Department at Süleyman Demirel University, where they were promptly processed into juice.

Solvent and Reagents

For enzymatic treatment, Novoferm-61 was obtained from Novozymes A/S (Bagsværd, Denmark). Clarifying agents 110 [bentonite, gelatin (Type A, 80–100 Bloom strength), and kieselsol] were obtained from Erbslöh Geisenheim GmbH (Geisenheim, Germany). All reagents used for chemical analyses were purchased from Merck Co. (Darmstadt, Germany). Freshly prepared ultrapure water (18.2 MΩ·cm) was used for all analyses (Millipore Simplicity UV, Molsheim, France).

Juice Production

In this study, to produce blueberry juice (BJ), two different methods that direct juice extraction (DJE) and mash treatment process (MTP) were applied. The evaluation involved hand-pressing fresh

berries in cheesecloth at the same intervals to compare the differences between these processing methods. Free juice, press cakes, and press bags were weighed, and recoveries of free juice, press cake, juice loss, and total juice were calculated and averaged. Free juice was considered the free-flowing juice that was collected. Press cake was the pulp, skins, and seeds that remained in the cheesecloth. Juice loss was the amount of juice lost in the mashing and pressing stages, including transfers and losses within cheesecloth. The total juice recovery was the free juice plus the juice loss.

Direct Juice Extraction (DJE)

Crude Blueberry Juice (CBJ) Production: The fresh blueberries were placed in a cheesecloth for juice production. The obtained hazy juice was called as “crude blueberry juice (CBJ)”. The CBJ was passed through a 4-layer muslin cloth for removing the residue. The filtered CBJ was bottled in 1.5-L plastic (PET) bottles and then cooled in an icy-water bath and stored at -18 °C until the clarification treatments.

Enzymatic Treatment and Clarification Trials for CBJ: The CBJs were first treated with commercial enzymes to break down the hydrocolloid polysaccharides, mainly pectin and starch. For this, the enzymes, [Novoferm-61 (Novozymes A/S) (2–10 mL/L)], were added at various concentrations to the CBJs in a temperature-controlled water bath (GFL 1004, Hannover, Germany) at 50 °C and the juices were then incubated at this temperature for a period of 2 h. After the incubation, the juice samples were filtered through a coarse filter paper (Isolab, Interlab Co., Istanbul, Turkey), then the transmittance (%) of the samples were determined (Table 1). The highest transmittance (%) was obtained for Novoferm-61 at a concentration of 2 mL/L was 66.53%.

Table 1. Transmittance values (%) for different enzyme dosage treatments of DJE and MTP in depectinization.

Enzyme Dosages	T (%)
<i>DJE Depectinization (mL L⁻¹)</i>	
2.0	66.55
6.0	60.24
8.0	59.43
10.0	64.13
<i>MTP Depectinization (mL kg⁻¹)</i>	
1.0	76.62
2.0	69.44

The main juice mass was then treated with this enzyme at the determined concentration. After enzymatic treatment, clarification trials were applied to the enzymatically treated BJJs with adding various amounts of bentonite (10%, w/v), gelatin (1%, w/v) and kiselsol (15%, v/v). The turbidities of the clarified samples were then determined. However, there was no significant differences between transmittances (%) for samples. The rates and transmittances (%) were represented in Table 1. For that, only enzymatic treatment was applied to the blueberry juice processing (DBJ, depectinized blueberry

juice). For pasteurization of the BJs (PDBJ, pasteurised and depectinized direct blueberry juice), the juice samples were transferred into 200-mL hermetically capped glass bottles.

Enzymatic Treatment and Clarification Trials

The blueberry mash (BM) was also treated with commercial enzyme (Novoferm-61). For that, the enzyme was added at various dosages to the BMs in a temperature-controlled water bath (GFL 1004, Hannover, Germany) at 45 °C and the BMs were then incubated at this temperature for a period of 30 min. The best amount of commercial enzyme (Novoferm-61) was determined as 1 mL kg⁻¹ because of the depectinization trials for mash. The rates and transmittances (%) were represented in Table 1. Negative alcohol precipitation test was used as an indication of depectinization in enzyme treated blueberry mash. After depectinization, the sample was placed in cheesecloth for pressing. After this procedure, the juice samples (DMTP, Depectinized Blueberry Mash Treatment Processing Juice) were filtered through a coarse filter paper (Isolab, Interlab Co., Istanbul, Turkey), and bottled into 200-mL hermetically capped glass bottles until pasteurization (PMTP, pasteurised and depectinized mash treatment blueberry juice).

Juice Pasteurization

For pasteurization of the DJE and MTP samples, which were bottled into 200-mL hermetically capped glass bottles, pasteurized in a water bath (WB 14, Memmert GmbH + Co. KG, Schwabach, Germany) at 95 °C for 3 min once the cold point reached to 95 oC, monitored by a thermocouple thermometer (Digi-Sense Model No:91100-50, Cole-Palmer Instrument Co., Vernon Hills, IL, U.S.A.). Following the pasteurization, the pasteurized juice samples were immediately cooled to room temperature by plunging the samples in an icy-water bath.

Analyses

Compositional Analyses: The pH of the samples was measured potentiometrically using a pH meter (WTW Inolab Level 1, Weilheim, Germany). Titratable acidity was determined according to the method outlined by IFU (1968) and expressed as “g anhydrous citric acid/100 mL or g sample.” Total soluble solid content (TSS) was measured by an automatic digital refractometer (Atago Rx-7000α, Tokyo, Japan) and expressed in brix (oBx, g/100 g) after the samples were filtered through a 0.45 µm PVDF (polyvinylidene fluoride) filter (Millipore Co., Bedford, MA, U.S.A.). The turbidity was measured by a turbidimeter (HACH 2100N, Loveland, CO, USA) as NTU (Nephelometric turbidity unit value). All compositional measurements were carried out at 20 °C.

Total Monomeric Anthocyanins (ACNs): Total monomeric ACN contents were determined using the pH-differential method, as described by Giusti and Wrolstad (2005). Monomeric ACN pigments reversibly change color with a change in pH; the colored oxonium form exists at pH 1.0, and the colorless hemiketal form predominates at pH 4.5. The difference in the absorbance of the pigments

at 520 nm is proportional to the pigment concentration (ACNs). Results are expressed on a malvidin-3-glucoside basis. Degraded ACNs in the polymeric form are resistant to color change regardless of pH and are not included in the measurements because they absorb at pH 4.5 as well as pH 1.0. The dilutions were then allowed to equilibrate for 15 min at room temperature (~22 °C).

In the first instance, the solutions were filtered through a 0.45 µm PVDF filter to remove the haze. The absorbance of equilibrated solutions at 512 nm (λ_{max}) for ACN content and 700 nm for haze correction was measured on a UV-VIS double-beam spectrophotometer (Thermo Spectronic Helios- α , Cambridge, England) with 1-cm path length disposable cuvettes (Brand Gmbh, Wertheim, Germany). All absorbance measurements were carried out at room temperature and made against distilled water as a blank. Pigment content was calculated as malvidin-3-glucoside equivalents, with a molecular weight of 493.5 and extinction coefficient of 28 000 L cm⁻¹ mg⁻¹. The difference in absorbance values at pH 1.0 and pH 4.5 was directly proportional to ACN concentration. All ACN measurements were replicated twice.

Polymeric Colour Content: Percent polymeric colour was determined using the method described by Giusti and Wrolstad (2005). For analysis, 0.2 mL of potassium metabisulfite was added to 2.8 mL of diluted sample (bisulfite-bleached sample) and 0.2 mL of deionized water was added to 2.8 mL of diluted sample (nonbleached, control sample). After equilibrating for 15 min, but not more than 1 h, samples were measured at λ_{max} (512 nm) for monomeric ACNs, 700 nm for haze correction and 420 nm for brown colour. Disposable cuvettes of 1-cm path length were used, the results were expressed as “%” and the measurements were replicated two times.

Total Phenolic Content (TPC): TPC of the samples was determined by the Folin-Ciocalteu method. Gallic acid was used as a standard to obtain a calibration curve. Gallic acid (25 mg) was dissolved in 50 mL absolute ethyl alcohol to get standard gallic acid solutions for solutions in different concentrations (50, 100, 200, 300 and 400 mg gallic acid L⁻¹). For analyse, 1 mL of each sample was added to 75 mL distilled water in a 100 mL volumetric flask. Then, 5 mL of Folin–Ciocalteu reagent was added into the mixture and, it was held at room temperature for 3 min. After 10 mL of saturated Na₂CO₃ solution was added to the mixture, absorbance values were determined at 720 nm after 60 min. Results were calculated and expressed as “mg of gallic acid equivalent per L juice”. TPC measurements were replicated two times.

Antioxidant Activity (AA): The AA was measured according to the ABTS method described by Türkiyeilmaz et al. (2013). ABTS⁺ radical cation was generated by reacting 7 mM ABTS and 2.45 mM potassium persulfate in the dark for 24 h at 20 °C. The phosphate buffer saline (PBS) was chosen as initially blanked solution for spectrophotometer. The samples were diluted 1:100 with PBS. Before the AA measurements, the ABTS⁺ solution was diluted to obtain an absorbance of 0.700 ± 0.020 at 734 nm with PBS at pH 7.4. Prior to analysis, the initial absorbance of the diluted ABTS⁺ solution was recorded

and then performed to the measurements for the different volumes of samples. Then, the absorbances at 734 nm were measured for 10, 20 and 30 µl of diluted samples which added to 990, 980 and 970 µl of the diluted ABTS⁺ solutions, respectively. The absorbances were also recorded after 6 min. Triplicate determinations were performed at each volume of the samples and the average values were calculated and, percentage inhibition (inhibition, %) was determined. Then the volumes of samples were plotted as a function of inhibition (%) to obtain the equation defining the curve with linear regression analysis. Trolox solution was used as a standard and all these values were calculated for trolox solutions (including 5, 10, 15 and 20 µl trolox). Results were calculated from the ratio of the slopes of the curves obtained for samples and trolox. The AA was expressed as “mM of Trolox equivalents mL⁻¹ juice”.

Statistical Analyses

Statistical analyses were performed using SPSS v.17 software. (IBM, NY, USA). Normal distribution and homogeneity of variance were previously tested by Shapiro-Wilk. Levene's test was used to check whether the variances were equal for samples that had a normal distribution. All results were found to be normally distributed with a 95% confidence ($p > 0.05$). Statistical differences between means were determined by analysis of Duncan's multiple range test at the 5% significant level ($p < 0.05$).

RESULTS and DISCUSSION

Changes in compositional measurements during juice processing

Changes in TSS content (°Brix), turbidity, pH and titratable acidity were determined during depectinization and pasteurization of DJE and MTP samples (Table 2). During both processing steps, there were no significant changes in pH, titratable acidity and TSS content ($p > 0.05$). The pH, titratable acidity and TSS content of DJE samples ranged from 3.04 to 3.08, 1.12 to 1.03 g/100 mL (as anhydrous citric acid) and 9.2 to 9.7, respectively (Table 2). The pH, titratable acidity and TSS values of MTP samples ranged from 2.84 to 3.04, 1.40-1.36 g/100 mL (as anhydrous citric acid) and 9.03, respectively (Table 2). These results showed that clarification and pasteurization did not affect the organic acid and sugar content of blueberry juices. Similar results were observed in black mulberry juice and pomegranate juice during clarification and pasteurization (Turfan et al. 2011; Askin et al. 2022).

Table 2. Compositional properties of DJE and MTP samples.

Samples	Soluble solid content (°Bx)	Turbidity (NTU)	pH	Titratable Acidity(%)**
<i>DJE Samples</i>				
CBJ	9.2±0.0000b	7.73±0.0305b	3.04±0.0057b	1.12±0.0077a
DBJ	8.7±0.1154c	52.67±0.5773c	3.09±0.005a	1.03±0.0178a
PDBJ	9.7±0.1000a	0.98±0.0404a	3.08±0.0057a	1.03±0.0067a
<i>MTP Samples</i>				
Mash	9.03±0.0577b	11.49±0.0141b	2.84±0.0152b	1.40±0.0206a
DMTP	10.06±0.057a	3.66±0.0152a	3.03±0.0251a	1.05±0.0357b

PMTP	9.03±0.0577b	3.82±0.1514a	3.04±0.0230a	1.36±0.0255a
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*Values were expressed as mean ± SD

*Different letters in the same column represent significant difference (p<0.05).

**Expressed as anhydrous citric acid.

Effect of processing and pasteurization on ACNs and colour properties of juices

The production of BJ is performed by various procedures. It pressed or processed into purees and enzyme preparation are applied releasing the cell-bound water and promoting the extraction of the fruit. These are cell-wall degrading enzymes such as pectinases and they are applied to pressed juice or mash. In the study, both were studied, and different changes were seen for ACN content in DJE and MTP samples (Figure 1) after depectinization. For DJE samples, there were a decrease in ACN contents from 75.15 to 52.28 (mg L⁻¹). Anthocyanins could not as efficiently be extracted in the pressing process as are sugars, acids, and other water-soluble components. In addition, anthocyanins are mainly found on the skins that is difficult to homogenate for blueberries (Hager, Howard and Prior 2008). Therefore, the press-cake is a potential colour source for following extraction and so causes a loss of anthocyanins in juices. Enzymatic treatment led to significant reductions in ACN content (30%), when polymeric colour had resulted an increasing in DJE processing (Figure 1). In literatures, it was pointed that various proportions of anthocyanin degradation (18 – 55 %) via pressing (Skrede, Wrolstad and Durst 2000; Lee and Wrolstad 2004; Hager, Howard and Prior 2008; 2008; Kulcan et al. 2015). Besides, some endogenous enzymes such as polyphenol oxidase (PPO) from blueberries cause anthocyanin degradation in crushed fresh blueberries (Perreault et al. 2021). Literatures have been shown that blueberry peroxidase, in the presence of chlorogenic acid and H₂O₂, rapidly oxidized ACNs into brown pigments (Hager, Howard and Prior 2008).

In mash process, the reduction of mash viscosity is a critical point for juice production. During mash depectinization, it is released higher content of juice and made easier pressing by depolymerisation of pectin and other cell wall polysaccharides (Weber and Larsen 2017). In MTP samples, depectinization had increasing effect on ACN content, significantly, from 44.49 to 172.69 (mg L⁻¹), as well as the polymeric colour value (Figure 1). There was no significant difference for the interaction between pectin and ACNs of MTP and DJE samples. However, depectinization enzymes interacted to all parts of the fruit (skin, pulp, and juice) in MTP processing. Anthocyanins are most located in the skins of fruits (Kader et al. 1999; Lee and Wrolstad 2004; Moyer et al. 2008). Consequently, it resulted in higher contents of ACNs in MTP samples. It was similar changes with other research (Rommel 1990; Chang 1994; Mikkelsen and Poll 2002; Askin et al. 2022). On the other hand, different enzymes possess various effects on anthocyanin concentration. Some literatures pointed out that some enzymes are responsible for decreased ACN content by the side activities that cleavage of the glycosidic bond of the ACNs. Another important factor is enzyme dosages for ACN content changes. It can be detected free anthocyanins while using excessive enzyme dosages or alteration of ACN profile when elevated dosages

are applied. Briefly, the reason for the difficulties in predicting anthocyanin losses might be found in the dosages, types, and origin of enzymes, and pectin structure of fruit (Landbo and Meyer 2004; Buchert et al. 2005; Hager, Howard and Prior 2008; Koponen et al. 2008).

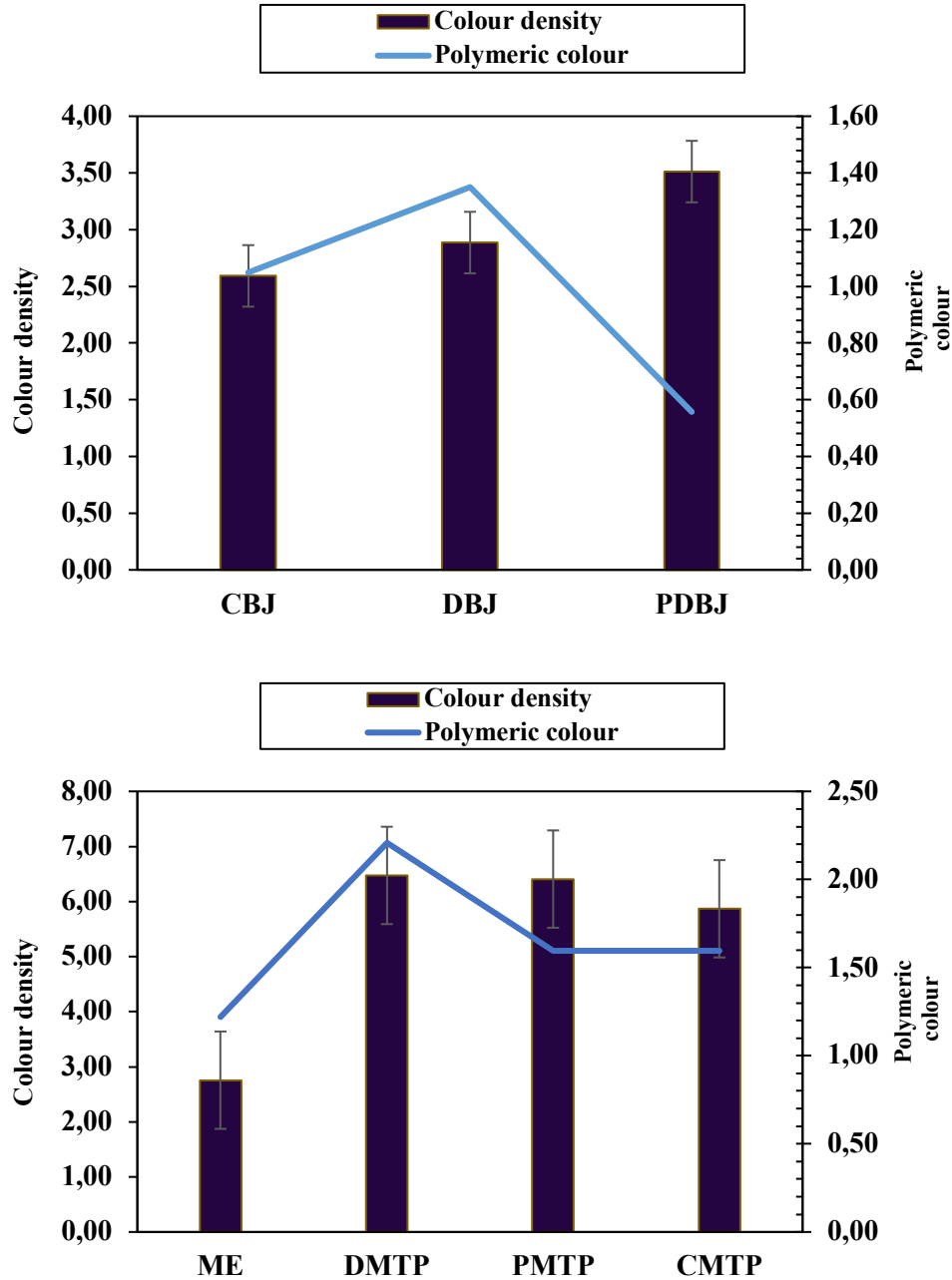


Figure 1. Changes in polymeric colour and colour density of BJ during processing.

The reason for the achieved results may be due to the interaction of pectin and ACN during depectinization. As it known blueberries contains more than about 0.5% pectin, is a complex polysaccharide found in the cell wall and middle lamella of plant tissue (Chevalier et al. 2019). Pectinases degrade the pectin molecules during depectinization and after that the ACNs of blueberries were released from the cells into juice (Askin et al. 2022). These interactions cause the changes in

stability, color properties, extractability, and bioavailability of ACNs (Askin et al. 2022; Fernandes et al. 2014). Many studies showed that the interaction between pectin and ACNs in depectinization resulted in higher ACN content (Askin et al. 2022).

Recent literatures proved that anthocyanin stability is not only a function of the pasteurization temperature, but also it is affected by the intrinsic properties of the product (pH, anthocyanins chemical structure, anthocyanin concentration, endogenous enzymes and other natural compounds called co-pigments, metallic ions, and sugars) and the processing (heating, storage temperature and time, the presence of oxygen and/or light) (Hager, Howard and Prior 2008; Rapeanu et al. 2005; Sulaimana et al. 2015; Terefe et al. 2015; Siddiq and Dolan 2017). There were increasing in ACN contents after pasteurization for both blueberry juice processes (DBJ and MTP).

The anthocyanin content of pasteurized DBJ (PDBJ) and pasteurized MTP (PMTP) were significantly higher (19%) than that of the initial pressed juice. The possible explanation for this phenomenon would be the release of ACNs from the plant cell by the effect of the heat applied during pasteurisation after enzymes were inactivated with pasteurization (Skrede, Wrolstad and Durst 2000; Türkyılmaz et al. 2012). Besides, the effect of pasteurization has high relation with anthocyanin structure, fruit matrix and processing parameters. The individual anthocyanins acted differently under pasteurization conditions (Weber and Larsen 2017).

In addition to all these, another point should be mentioned about anthocyanin changes. Literatures also observed a considerable alteration of the anthocyanin profile in juices by processing method (Skrede, Wrolstad and Durst 2000; Lee and Wrolstad 2004; Hager, Howard and Prior 2008). For instance, it was observed a dramatical changes of ACN profiles with increasing proportions of malvidin glycosides and decreasing proportions of delphinidin derivatives in pressing (Skrede, Wrolstad and Durst 2000; Lee and Wrolstad 2004). The authors also concluded that activity of some enzymes might be responsible for selective degradation at the different steps of processing, enzyme applications and heat treatments, except glycosidases (Threlfall et al. 2006; Dobson et al. 2016; Weber and Larsen 2017).

Effects of processing and pasteurization on total phenolic content of juices

The total phenolic content of the blueberry juices varied widely depending on the method of fruit mash treatment prior to juice pressing used in the process (Table 3). The differences between the processing methods and each step were found to be statistically significant ($p < 0.05$). Different research revealed that the conditions of berry mash conditions greatly affect the final polyphenol concentration in juices (Landbo and Meyer 2004; Koponen et al. 2008). In the study, MTP samples had the lowest content of total phenolic compounds (471 mg L^{-1}). However, the juices DMTP produced from MTP were found to be the richest sources of phenolic compounds (829 mg L^{-1}) (Table 3). Besides, an increase of approximately 10% for DJE samples was detected in the total amount of phenolic substances after

the depectinization process. The pectolytic enzymes that are added to the blueberry juices in the depectinization stage, break down pectin, which binds cells together in plant tissues, and ensures the release of components such as phenolic substances in the cell. This increase in the number of phenolic compounds observed at the end of the depectinization process is due to the release of phenolics because of the breakdown of pectin (Szajdek and Borowska 2008). Similarly, many studies have shown that pectolytic enzymes in different fruit beverages such as black currant, blackberry, plum etc. cause a significant increase in titratable acidity, total soluble solid content (TSS) and anthocyanin contents, especially phenolic compounds (Rommel, Heatherbell and Wrolstad 1990; Chang et al. 1994; Mikkelsen and Poll 2002).

Table 3. Changes in total monomeric anthocyanin content, total phenolic content, antioxidant activity and ascorbic acid content of DJE and MTP during depectinization and pasteurization.

Samples	TPC (mg L ⁻¹)	Antioxidant Activity (mM L ⁻¹)
<i>DBJ</i>		
CBJ	672±1.0000c	6.54±0.0152c
DBJ	738±3.7859b	6.72±0.0251b
PDBJ	787±9.2376a	9.55±0.0251a
<i>MTP Samples</i>		
ME	471±2.8284d	5.56±0.0152d
DMTP	829±4.2426a	11.21±0.0208b
PMTP	779±4.9497b	11.77±0.0152a
CMTP	752±5.6568c	11.17±0.0152c

*Values were expressed as mean ± SD

*Different letters in the same column represent significant difference (p<0.05).

Pasteurized MTP (PMTP) juice had a slightly higher TPC than the unpasteurized juice (Table 3), possibly due to release of Folin-Reactive components during pasteurization. However, DJE samples were similar for pasteurized and unpasteurized juices. On the other hand, pasteurization is defined as deactivate enzymes, and in some fruit juices there was almost complete deactivation of polyphenol oxidase and peroxidase. The deactivation of such enzymes in the pasteurized juices might explain why the loss of phenols was less than in the unpasteurized juices (Szajdek and Borowska 2008; Saeeduddin et al. 2015).

Effects of processing and pasteurization on antioxidant activity

Berry juices are a rich source of biologically active compounds, exhibiting strong antioxidant activity (Yilmaz and Toledo, 2005). To characterize the antioxidant properties of juices, percentage inhibition (inhibition, %) was determined by ABTS radical scavenging assay. Lower values of inhibition indicate lower activity of the juice. The untreated juices (DJE and MTP) exhibited the weakest antioxidant

activity (6.54- and 5.56-mM L⁻¹, respectively) (Table 3). After that depectinization application, there was a statistically significant ($p < 0.05$) increase in activities. As expected, depectinized juices with on the method of mash treatment prior to pressing (DMTP) had higher antioxidant activity than directly depectinized blueberry juice (DDBJ). It is possible that the high concentrations of polymers were significant contributors to the antioxidant levels for DBJ samples. These juices were also the richest sources of polyphenols (Table 3). With the pasteurization application, while a significant increase was detected in PDBJ ($p < 0.05$), no significant difference was observed in PMTP samples ($p > 0.05$) (Table 3). The increase in antioxidant capacity may be due to the formation of Maillard reaction products in response to thermal treatment or the formation of anthocyanin polymers (Yilmaz and Toledo 2005). The antioxidant capacity of polymeric anthocyanins balanced out for the loss of antioxidant capacity because of monomeric anthocyanin degradation.

CONCLUSION

Blueberries have become popular beverages due to their nutritional values such as vitamins, minerals, and antioxidants. This is important for blueberry research that need to be found out chemical composition, processed products, effects of process steps and technology for increasing evaluation choices. Juice is the most popular processed product, and it should be determined that the effects of processing methods on yield and quality. Besides, how each juice processing step can impact the chemical composition can help producers to increasing juice yield and revised the quality of the juice and provided new product options.

Processing, enzymatic treatment and pasteurization revealed significant effects on ACNs and colour of blueberry juice. Although depectinization led to reducing the ACN contents only in DJE samples, colour density for these samples increased. This may be due to that clarification provided appropriate molar ratio of phenolics to ACNs for copigmentation. For MTP samples, there were increasing in ACNs during depectinization, accompanied by an increase in colour intensity along with colour intensity.

We found that anthocyanin, phenolic content, and antioxidant activity were higher in fruit juice in the MTP method compared to the DJE method, and the juice yield was higher. MTP method should be preferred when processing blueberry juice into fruit juice. However, evaluation of kinetic changes and storage stability is also required for DJE and MTP processing. In addition, the mechanism of different new techniques needs to be evaluated for a better understanding of the processing effects on BJ quality. New approaches for thermal and non-thermal applications and their effects on changes and subsequent changes in anthocyanins, phenolics and antiradical activity should be investigated. The most important benefit of improved juice processing, which is a combination of different applications, will be to achieve target quality, reduce harmful conditions and maintain storage stability.

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