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Influence of juice processing factors on quality of black chokeberry pomace as a future resource for colour extraction

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ABSTRACT

Aronia melanocarpa berries are a rich source of anthocyanins and its pomace, a by-product of juice processing, could be efficiently used for extraction of natural colours for the food industry. This study evaluated the influence blanching, freezing, maceration temperatures (2 °C, 50 °C) and enzyme treatments before juice pressing on the yield and anthocyanin composition of both juice and pomace.

Total anthocyanin levels in pomace were affected mostly by enzyme treatment followed by maceration temperature. The pre-heating of the mash prior to processing increased juice yield and retention of anthocyanins in the pomace. Cold maceration of frozen berries without enzyme addition gave the highest concentrations of anthocyanins in the pomace, and both cold and hot maceration of fresh unblanched berries with enzyme the lowest. The results support future exploitation of natural colours from pomace side streams of *Aronia*, thus increasing competitiveness of *Aronia* berry production.

1. Introduction

In recent years, the use of natural colour additives in food has substantially increased following demands by consumers who desire colourants from natural (plant) sources, reinforced on the belief that they are better from a health perspective (Oplatowska-Stachowiak & Elliott, 2015). This was spurred; following concerns that synthetic colours may cause adverse health effects e.g., behavioral and neurological problems, especially in children (Stevens et al., 2013). As a consequence, food industries tend to make efforts to replace their synthetic colouring agents with natural colourants (Bearth, Cousin, & Siegrist, 2014).

Anthocyanins – a group of phenolic compounds, with reddish-purple hues, are well known natural alternatives to synthetic colours, with potential application in colouring of food products (Rodriguez-Amaya, 2016). Besides their colouring properties, anthocyanins are also bioactive components of interest in human health (Rodriguez-Mateos, Heiss, Borges, & Crozier, 2014).

Berry fruits are well documented among the widely recognized plant sources of anthocyanins (Rodriguez-Mateos et al., 2014; Sosnowska et al., 2015). For instance, black chokeberries (*Aronia melanocarpa* (Michx.) Elliot) have been highlighted as a suitable source for production of natural food colourants (Jeppsson, 1996), and are one among the richest known sources of proanthocyanidins and anthocyanins, in comparison to other berries e.g., elderberries and black currant (Kulling & Rawel, 2008; Veberic, Slatnar, Bizjak, Stampar, & Mikulic-Petkovsek, 2015; Wu, Gu, Prior, & McKay, 2004). Furthermore, there is convincing evidence that *Aronia* extracts exhibit significant health promoting properties such as anti-inflammatory, anti-diabetic effects as well as combating heart diseases and other cardiovascular diseases (Chrubasik, George, & Sigrun, 2010; Kulling & Rawel, 2008; Sosnowska et al., 2015). This places *Aronia* in the top end of fruits and berry species that could be a promising industrial raw material, and increased popularity of *Aronia* products is expected in the future.

Aronia processing into juice generates by-products such as pomace (press cake residue). As a by-product of processing, pomace is an under utilized product, and often discarded. Recent studies have, however, shown that *Aronia* pomace is a good source of phenolic compounds, especially anthocyanins (Sójka, Kołodziejczyk, & Milala, 2013; Wilkes, Howard, Brownmiller, & Prior, 2013). In order for the industry to exploit this resource more information is needed on how the quality of *Aronia* pomace varies depending on the methods used in juice pressing and what quality of the colour extract can be obtained from pomace. In the natural colourant field, production of concentrated powdered form of anthocyanin extracts is done using freezer drying or spray drying. In addition, several methods employing solvents such as ethanol, methanol and acetone followed by technologies such as

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pressurized liquid extraction, supercritical fluid extraction and ultrasound or microwave-assisted extraction are used for the extraction of anthocyanin from the pomace (Struck, Plaza, Turner, & Rohm, 2016).

The processing of fresh berries to juice is basically dependent on processing parameters, such as fruit pre-treatment, grinding, mash heating, mash maceration with enzyme preparations, pressing, pasteurization and filtration (Kårlund, Moor, Sandell, & Karjalainen, 2014). In addition to processing, subsequent storage of the product can alter the quality and content of anthocyanins, resulting in significant losses in the product (Patras, Brunton, O'Donnell, & Tiwari, 2010; Tiwari & Cummins, 2013). For example, blanching at 95 °C/3 min in combination with pasteurization during processing of *Aronia* into juice resulted in significant loss of anthocyanins, when compared to original levels found in fresh berries (Wilkes et al., 2013). During processing of blueberries, it has been reported that cold press extracted blueberry juices had a light blue colour, while hot press juice with enzyme treatment produced juice with a dark purple-blue colour (Bates, Morris, & Crandall, 2001). Enzymatic processing of black currants increased the juice yield by 10–22% and the content of various phenolic compounds in juice by 4–10-fold as compared to the non-enzymatic process (Laaksonen, Mäkilä, Tahvonen, Kallio, & Yang, 2012a). Furthermore, freezing of berries prior to processing could have a profound effect on the stability and final quality of anthocyanins (Jensen, unpublished results).

Although some studies, on the effect of processing on *Aronia* polyphenols in juice have been performed (Mayer-Miebach, Adamiuk, & Behsnilian, 2012; Wilkes et al., 2013), there is still limited information regarding the influence of processing on *Aronia* juice yield and quality characteristics of the pomace.

Hence the aim of this study was to understand the influence of juice processing steps and factors on yield, anthocyanin retention and profile in *Aronia* pomace. Providing a detailed characterization of pomace from different pomace sources and knowledge on processing effects will allow development of recommendations to the industry to make well informed decisions on how to exploit pomace from *Aronia* for colour production in future.

Parameters tested were (1) pre-processing treatment (blanching, no blanching, freezing); (2) temperature during maceration (cold maceration (2 °C) and hot maceration (50 °C); and (3) enzyme treatments (enzyme or no-enzyme addition). In addition, anthocyanin retention during processing of frozen berries to juice and pomace was determined.

2. Materials and methods

2.1. Chemicals and reagents

Acetonitrile (isocratic grade, purity >99.8%), methanol (HPLC grade, purity >99.8%), sodium acetate (CH_3COONa), sodium hydroxide (NaOH) and potassium chloride (KCl), hydrochloric acid (HCl) were obtained from Sigma-Aldrich (Darmstadt, Germany). Trifluoroacetic acid was purchased from VWR (Leuven, Belgium). Cyanidin-3-galactoside, cyanidin-3-glucoside and cyanidin-3-arabinoside were purchased from Extrasynthese (Genay, France). Pectinex Ultra SP-L® enzyme was purchased from Novozymes (Bagsvaerd, Denmark).

2.2. Plant material

Fully ripe berries (21 kg) from the cultivar *A. melanocarpa* 'Viking' were manually harvested at the Department of Food Science, Aarhus University (Årslev, Denmark) on September 22, 2015 at optimal maturity when the berries were at 19–21 °Brix. The *Aronia* planting was 14 years old and grown organically with 80 kg N supplied per year. The fresh berries were separated from stalks and leaves, washed with cold water and dried in air at room temperature for approximately 1 h. The fresh berries were then divided into three batches, which were pre-treated separately prior to juice processing. To inactivate the polyphenol oxidase known to degrade anthocyanins and facilitate softening of skins, to allow better extraction of anthocyanins (Patras, Brunton, O'Donnell, & Tiwari, 2010), the first batch (A) was subjected to a blanching step by dipping small batches (500 g) of berries with a mesh sieve in water heated to 95 °C for 10 s and then cooled to 30 °C. In the second batch (B), the berries were not blanched, while in the third batch (C) the berries were frozen by transferring to a freezer (−20 °C) within 4 h of harvest. The fresh berries were processed within 48 h of harvesting, while the frozen berries were processed after 4 days of storage at −20 °C.

2.3. Processing treatment

In Denmark, hot pressing (involving heat and enzyme) are widely used commercial scale juicing methods for fruit and berries including *Aronia,* but also cold pressing (involving no heat or enzyme) are used commercially. Therefore, in order to simulate these processes in the range from low to high intensity juice processing, an optimized 'lab scale' procedure was developed in this study. A flow chart representing the processing scheme and processing treatments are given in Fig. 1 and Table 1, respectively.

The pre-treated berries–fresh blanched, fresh unblanched and frozen (thawed overnight at 3 °C), were mashed at low speed bursts for 15 s at room temperature using a blender (Kenwood multipro) and divided into 12 samples/treatments (500 g each sample), and transferred to 1.5 L glass beakers. The following treatments (Table 1) were then applied to the pre-treated berry samples: cold treatment (1, 2, 3), combined cold and enzyme treatment (4, 5, 6), heat treatment (7, 8, 9), and combined heat and enzyme treatment (10, 11, 12). Each treatment was repeated three times.

For the cold enzyme aided treatments, the mash was maintained at 0–2 °C (referred to as 2 °C in this study) in an ice bath and 0.2 mL/kg Pectinex Ultra SP-L enzyme was added and incubated for 1 h. The mash was stirred at regular intervals during the process. Afterwards, the mash was transferred to 2 L polyethylene bags, which was positioned between two parallel porous metal plates (distance 1.1 cm, to facilitate rapid heating of the mash) and pectinase enzyme deactivation was done by heating to 90 °C for 5 min in a temperature controlled water bath, followed by rapid cooling in an ice bath. Juice and pomace was extracted from the mash by pressing with an hydraulic juice extractor (Enerpac, Denmark) through a cotton cheesecloth operated at 400 Ba for 5 min.

For the heat enzyme aided treatments, the mash was preheated to a controlled temperature of 50 °C in a water bath for 30 min and Pectinex Ultra SP-L enzyme was added (0.2 mL/kg) and incubated at 50 °C for 1 h. Enzyme deactivation and pressing was done as described for the cold enzyme treated mash.

For the cold and heat non-enzymatic treatments, the mashes were maintained at 2 °C and 50 °C, respectively and thereafter directly pressed.

After pressing, the weights of the resulting juice and pomace were determined, and weight yields calculated. The samples were stored frozen at −20 °C until further analysis. Each treatment consisted of three independent repeatings.

2.4. Extraction of anthocyanins

The extraction was carried out as described by Dinkova et al. (2014) with slight modifications. Extraction of both fruit and