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A practical method for isolation of phenolic compounds from black carrot utilizing pressurized water extraction with in-site particle generation in hot air assistance

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ABSTRACT

Anthocyanins and related phenolic compounds can be extracted from highly pigmented garden vegetables and their by-products using subcritical water with or without a secondary co-solvent, ethanol. Carrot, being one of the richest vegetables for its natural components, is well known for its high phenolic content. The present study is concerned with the study of bioactive compound profile of black carrot. Extracted phenolic compounds were recovered in powdered form by establishing a novel system capable of doing subcritical water extraction along with powdered particle production. Pressurized water extraction was carried out at 10 and 20 MPa and at temperatures ranging from 40 to 100 °C. The extraction efficiencies in terms of anthocyanin recovery were compared with respect to the extraction parameters employed and the product was fed to air assisted particle formation system to investigate the process functions, the effect of temperature and pressure on the resultant particle as well.

1. Introduction

In recent years, number of studies on public health, environmental protection, solvent contamination and controlled food production using organic solvents have been significantly increased [1–3]. The cost of organic solvents for food processing applications is reasonably high and there have been strict regulations for waste management. For the reasons expressed above, researches provided alternative directions for their studies for production of pure, high value added food products using processes, i.e., green techniques [4–7].

Supercritical fluid technology has advantages of being cheap, safe and is applicable to wide range of food materials [8–11]. Other advantages in using diffusivity characteristics being close to gases and being at least two orders of magnitude larger than liquids [6,9]. Extraction of flavor essences, fats, oils, antioxidants, polysaccharides, triterpenoids and other food ingredient substances from natural plants are examples of the application of supercritical fluids [10,11].

Black carrot is a good source for phenolic compounds as a rich source due to their nutraceutical value., as well as there being fortified in many other constituents such as anthocyanins, calcium, iron and zinc (Table 1). Also, antioxidant activity of black carrot is four times more than the red carrot [12]. Black carrots are a good choice among natural colorants for pastry industry and also used as appetizer in salad, fruit and vegetable juices, candies, jellies, etc [13–16]. Among natural pigments in plants, anthocyanins are one of the best belonging to polyphenolic class of compounds. These antioxidants can prevent cancer, improve visual functions and prevent cardiovascular diseases [17–21].

The extraction of anthocyanins from natural matrices, conventional techniques are generally used. In these classical methods mostly ethanol is used. Moreover, conventional methods are time- and solvent-consuming and may have a destructive effect on anthocyanins due to the use of elevated temperature and long time during the extraction [22–24]. High temperature values have been reported to increase anthocyanin degradation. Besides, solvents that are eluted from extraction system upsets the balance for environmental protection [25–27].

Studies performed on black carrots to extract phenolic compounds have made use of acidified water in using citric or lactic acid at low temperatures starting from 80 °C up to 100 °C. Extraction pressures were set at constant level between 10 and 20 MPa. These types of acid-aided solvent studies has led to solute deterioration at temperatures above 100 °C. Lately, improved extraction using ethanol in water (40%-80%) instead of acidified water at temperatures ranging from 70 °C to 130 °C from grapes [28].

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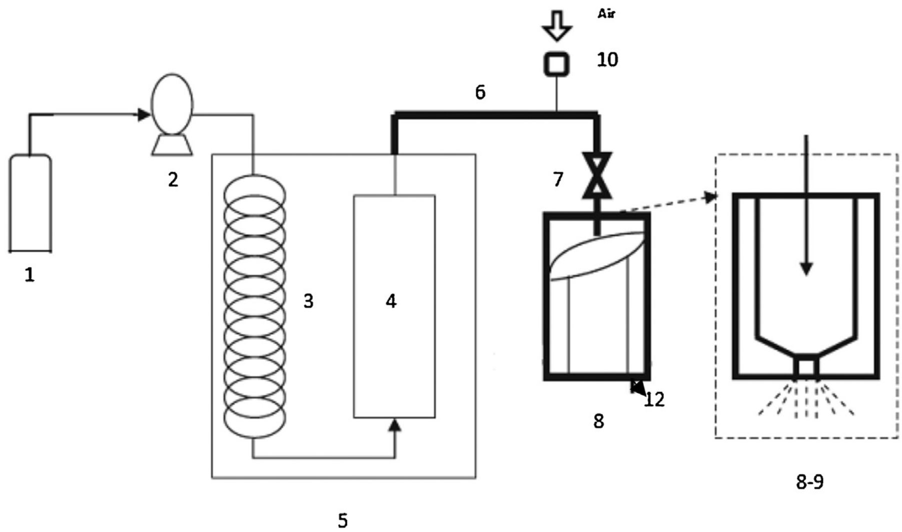

Fig. 1. Combined extraction/particle production apparatus: (1) Distilled water reservoir; (2) HPLC pump; (3) pre-heater; (4) extraction vessel; (5) oven; (6) extract outlet pipe line; (7) ball valve; (8) collection chamber; (9) air atomizing expansion nozzle; (10) air inlet-needle valve; (12) vacuum-air exit.

Table 1
Nutrients present in black carrot (per 100 g, w/w, http://www.natureword.com/).

Nutrient name	Milligrams (mg)	Percentage (%)
Folates	19	5
Niacin	0.983	6
Pantothenic acid	0.273	5.5
Pyridoxine	0.138	10
Riboflavin	0.058	4
Thiamin	0.066	6
Vitamin A	16706 IU	557
Vitamin C	5.9	10
Vitamin K	13.2	11
Sodium	69	4.5
Potassium	320	6.5
Calcium	33	3
Copper	0.045	5
Iron	0.30	4
Magnesium	12	3
Manganese	0.143	6
Phosphorus	35	5
Selenium	0.0001	≤1
Zinc	0.24	2

Being an interdisciplinary research, this study combines the importance of healthy diet by creating an alternative method for extraction and new functional food development. Therefore, the purpose of this study was to determine the applicability of subcritical water extraction for extraction of anthocyanins from black carrot and to investigate the effects of temperature and pressure on the anthocyanin and phenolic content total extraction yield. The technique couples subcritical water extraction with air assisted particle formation technology.

Examples of different particle formation methods have arisen from the use of carbon dioxide as an extraction solvent over the last 20 years [19–21]. In these cases, the polarity of supercritical fluids can be changed by using co-solvents appropriate to the polarity of target compound to be extracted [27–30]. Hence, an alternative can be offered using compounds, and by linking the extraction process directly with the particle formation process [22–26].

Specifically, dried black carrot samples were extracted at different temperatures ranging from 40 to 100 °C using subcritical water assisted with 80% (v/v) ethanol. The selected extracts were completely dried and fed to the process to obtain fine particles that can be directly used food products as natural coloring agents.

2. Materials and methods

2.1. Materials

Black carrots were purchased from the local market (Konya, Turkey). Anthocyanin standards: Cyanidin-3-diglucoside-5 glucoside and Cyanidin 3,5 diglucoside were obtained from Carl Roth GmbH D-76185 Karlsruhe, Germany.

Glucose standard solution containing 0.1% (w/v) benzoic acid and having 1 mg/mL concentration was purchased from Sigma Aldrich Co. LLC Munich, Germany.

2.2. Methods

The black carrot samples were cut into smaller particles and then dried at 50 °C in an oven (Memmert, Germany). After drying, samples were ground to nearly 3 mm particle sizes. The ground and dried black carrot particles were then placed in the extraction column. The bottom and upper parts of column was filled with glass beads to reduce the void volume in the parts and to form compact extraction area. Extract was obtained by performing subcritical water extraction (SWE). Relevant experimental procedures were performed using replication method. The treatment combinations were set to correlate and to compare matches with the parallel sets of experiments. Extracts were kept at +4 °C until analyzed. Following the extraction, particle production was applied using hot air assistance given in the sections below.

2.2.1. Combined subcritical water extraction (SWE)/Particle formation system

The selectivity of subcritical water extraction allows for manipulation of the composition of the extracts by changing primarily the experimental temperature. Two different extraction mechanisms were used: [1] adding acid to water and [2] using neat water. These kinds of aqueous-based treatments were done in order to emphasize the effect of water diffusivity in the sub-critical extraction process [30,31]. Both water-soluble and insoluble polysaccharide fractions were isolated from black carrot samples.

A schematic representation of the combined extraction/particle production experimental apparatus that was used to produce powders rich in phenolics and anthocyanins was shown in Figs. 1 and 2, respectively. This system has exhibited a novel property of being