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Nutritional and Bioactive Compounds in Dried Sea-Buckthorn Pomace

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Abstract

The purpose of this work was to study the nutritional and bioactive quality of dried sea buckthorn pomace by proximate analysis and evaluation of total phenolics, total flavonoids, total carotenoids content and ABTS antioxidant activity. In addition, the carotenoid, amino acid and fatty acid profiles were assessed using chromatographic methods while mineral content was determined using graphite furnace atomic absorption spectrometry. Dried sea buckthorn pomace presented high values of the fat (20.05%), crude protein (14.89%) and crude fiber (19.86%) content. The essential amino acids represented 38.42% of total amino acids, the most abundant being leucine, followed by phenylalanine and lysine. The fatty acid profile revealed a high concentration of monounsaturated fatty acids (53.08% of total fatty acids), as a result of the high content of oleic and palmitoleic acids, and a low ratio of *n*-6/*n*-3 polyunsaturated fatty acids (1.42). Total carotenoids showed average contents of 245.6 mg/100 g, of which the major ones were β-carotene (80.76 mg/kg) and zeaxanthin (69.60 mg/kg). The results demonstrated that dried sea buckthorn by-products are valuable sources of nutritional and bioactive compounds and have potential to be used as nutraceutical for feed, as ingredient for functional food, as well as for the pharmaceutical industry.

Keywords Fatty acids · Amino acids · Carotenoids · Minerals · Antioxidant activity · Sea-buckthorn pomace · Nutritional

Ernährungsphysiologische und bioaktive Verbindungen in getrocknetem Sanddorn-Trester

Schlüsselwörter Fettsäuren · Aminosäuren · Carotinoide · Mineralien · Antioxidative Aktivität · Sanddorn-Trester · Ernährungsphysiologisch

Introduction

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 (Bal et al. 2011). The female plants produce waxy skinned yellow to orange—red berries, with diameters between 3 and 8 mm, containing a single sheathed seed and a juice filled cellular structure (Beveridge et al. 2002; Li 2003).

Sea buckthorn berries are currently of great interest thanks to their nutraceutical properties and high antioxidant contents, already proven by the ancient traditional uses in medicine for the treatment of asthma, skin diseases, gastric ulcers and lung disorders (Beveridge et al. 1999; Suryakumar and Gupta 2011), and abundantly studied especially at the beginning of the 21st century (Johansson et al. 2000; Süleyman et al. 2001; Eccleston et al. 2002; Yang and Kallio 2002; Zeb 2004a). The studies revealed various pharmacological activities of sea buckthorn fruits, such as cytoprotective, anti-stress, immunomodulatory, hepatoprotective, radioprotective, anti-atherogenic, anti-tumor, anti-

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microbial and tissue regeneration (Zeb 2004b; Suryakumar and Gupta 2011).

In the last decades many medicinal products of wild and cultivated sea buckthorn have been used in Asian and European countries (Chen et al. 2013) and the amount of sea buckthorn fruits used for production of nutritional supplements and functional food ingredients is steadily increasing (Stahl and Sies 2003).

Although the fruits are very acidic and astringent, sea buckthorn is a food resource with great potential. Sea buckthorn juice and pulp are often used as food or beverages as they are very rich in vitamins (C, E, A, B1, B2, F, K and P), carotenoids, flavonoids, tocopherols and other potentially health-beneficial components (Zeb 2004a; Suryakumar and Gupta 2011). The fruits of sea buckthorn have an appreciable content of oil containing among others, two essential fatty acids, linolenic acid (*n*-3) and linoleic acid (*n*-6) (Beveridge et al. 1999; Gao et al. 2000; Yang and Kallio 2001; Kallio et al. 2002; Rösch et al. 2003). The high contents of tocopherols, tocotrienols and carotenoids in the oil (Zadernowski et al. 2003) confer antioxidant properties, demonstrated in numerous studies in humans and in vitro (Gao et al. 2003; Eccleston et al. 2002). The high content of lipids of these carotenoid-rich fruits helps to increase the carotenoids bioavailability and to enhance their absorption in humans (Ranjith et al. 2006; Kruczek et al. 2012).

The processing of sea buckthorn berries for juice extraction leads to a large amount of residues, accounting for 20% of the total fruit weight, consisting of pulp, seed and skin which are known to be rich in carotenoids, polyphenols, fatty acids and sterols (Rösch et al. 2004; Dulf et al. 2012; Radenkovs et al. 2018). In order to reduce the wastes, sea buckthorn pomace is generally utilized as animal feed or for the extraction of biologically active compounds, providing beneficial food constituents, antioxidants, and cosmetics products (Périno-Issartier et al. 2011). Other food applications targeted the addition of sea buckthorn pomace to bread and other bakery products in order to increase their nutritive value (Lougas et al. 2005; Kant et al. 2012) and the direct enrichment of edible oils with sea buckthorn carotenoids (Chemat et al. 2012).

The present study was aimed to evaluate the nutritional and antioxidant properties of sea buckthorn pomace in order to promote the consumption and the use of this extremely valuable residual resource in the food industry.

Materials and Methods

Plant Material

Samples of sea buckthorn pomace (peels, seeds, and residual pulp) were collected from Biocat Prod S.R.L., a commercial producer and processor of sea buckthorn from Gradina, Constanta county, South-Est Romania. The sea buckthorn pomace was the by-products resulted from berries after the juice had been extracted by pressing. As soon as obtained, by-products were packed in plastic bags and frozen at –25 °C. Subsequently, sea buckthorn by-products were subjected to drying in an industrial automated forced hot air dryer (Blue Spark Systems S.R.L., Romania) at 60 °C. The dried material was powdered using an electric grinder, packed in aluminium coated polyethylene bags and stored at the room temperature (18–22 °C) till further analysis.

Chemicals and Reagents

Carotenoid standards (lutein, zeaxanthin, canthaxanthin, astaxanthin, lycopene, β-carotene and trans-β-apo-8′-carotenal) were purchased from Sigma-Aldrich (Chemie, Steinheim, Germany). Fatty acid standards were purchased from Supelco (Supelco 37 component FAME mix, Supelco, Bellefonte, PA).

The solvents used for extraction were of analytical grade while the solvents used for chromatographic analysis (acetonitrile, ethyl-acetate and methanol) were of HPLC grade (Merck, Darmstadt, Germany). Trolox (6-hydroxy-2,5,7,8,-tetramethyl-chroman-2-carboxylic acid), butylated hydroxyanisole, 2,2′-azinobis-3-ethylbenzothiazoline-6-sulfonic acid (ABTS) and Folin-Ciocalteu reagent were obtained from Sigma Aldrich (Germany). All other reagents were of analytical grade (Merck, Germany).

Proximate Composition

The dry-matter content was determined by drying the samples in the Ecocell Comfort drying oven (MMM Medcenter, Germany) at 105 °C until constant weight was obtained. The analysis of crude protein content was conducted according to the Kjeldahl method using a Kjeltec 2300 analyzer unit (Tecator, Sweden).

The crude fat content of the samples was measured by extracting the fat with petroleum ether in a Soxtec 2055 extraction unit (Tecator, Sweden), while the crude fiber content was estimated by the organic residue remaining after digestion with acid and alkali using a Fibertec 2010 (Tecator, Sweden) automatic analyser.

The ash content was determined as the residue after calcination at 550 °C in a Caloris CL 1206 oven (Romania) to constant weight.

Fatty Acid Composition

Dried sea buckthorn pomace was characterized through standard fatty acid methyl esters (FAME) gas chromatog-

raphy method. Fatty acids from the total lipid extracts were converted to their methyl esters using methanol containing 3% concentrated sulfuric acid at 80 °C as a reagent for transesterification. Methyl esters of fatty acids were analyzed in a Perkin Elmer-Clarus 500 gas chromatograph equipped with flame ionization detector (FID) and fitted with a BPX70 capillary column (60 m × 0.25 mm i. d., 0.25 µm film thickness). Column temperature was programmed at 5 °C/min^{-1} from 180 °C to 220 °C. The carrier gas was hydrogen (35 cm/s linear velocity at 180 °C) and the spliting ratio was 1:100. The injector and detector temperatures were 250 and 260 °C, respectively. FAME identification was done by comparison with retention times of the known standards. The results were expressed as g fatty acid per 100 g total fatty acids.

Amino Acid Composition

The amino acid composition was determined by high performance liquid chromatography and gradient elution following acid hydrolysis of samples and derivatization with ortho-phthalaldehyde (OPA)/9-fluorenylmethyloxycarbonyl chloride (FMOC) as described by Varzaru et al. (2013). Sulfur-containing amino acids were converted into cysteic acid and methionine sulfone by pre-oxidation with performic acid prior to hydrolysis and derivatization. The chromatographic separation was performed in a HPLC Finningan Surveyor Plus system (Thermo-Electron Corporation, Waltham, MA) equipped with a diode array detector (DAD) and a Hypersil BDS C18 column (250 × 4.6 mm, particle size 5 µm) (Thermo-Electron Corporation, Waltham, MA). The column operated at 45 °C with a flow rate of 1.7 mL/min using 50 mM phosphate buffer (pH 7.5) as eluent A and water/acetonitrile/methanol (20/20/60) as eluent B. The following linear gradient elution was performed: 2 min at 0% B; 0–57% B in 23 min; 57–100% B in 1 min; 3 min at 100% B; 100–0% B in 1 min and 5 min at 0% B. The DAD was set at 338 nm to monitor the derivatised amino acids. Stock solution of the standard amino acid mixture was prepared in 0.1 M hydrochloric acid and contained 500 µg/mL for each amino acid. Quantitation was based on the external standard method using calibration curves fitted by linear regression analysis. Data were acquired and processed with ChromQuest software.

Mineral Content

Calcium, copper, iron, manganese and zinc were determined by graphite furnace atomic absorption spectrometry (GF-AAS, SOLAAR M, Thermo Electron Corporation, USA) after acid high-pressure digestion in a microwave oven (MWS-2 Microwave System Speedwave, Berghof, Eningen, Germany). The mineral content was expressed in mg per kg of sample.

Total Phenolic Content

Total phenolic content was quantified in the extracts of dried seabuckthorn pomace by using the Folin-Ciocalteu's phenol reagent as described by Singleton and Rossi (1965). For preparing the extracts, 0.3 g of sample were mixed with 5 mL of methanol in the ultrasonic bath for 50 min, then the resulting mixture was centrifuged for 5 min at 4200 rpm and filtered through a 0.45 µm polyamide membrane.

Aliquots of the sample extracts (0.1 mL) or gallic acid standard solutions were transferred into test tubes and mixed with 5 mL of distilled water and 500 µL of Folin-Ciocalteu reagent. After a 5 min reaction time, 1.5 mL of 20% sodium carbonate solution and 2.9 mL of distilled water were added. After incubation for 30 min at 40 °C, the absorbance was read at 765 nm using a Varian Cary 50 UV-Vis spectrophotometer (Varian Co., USA). Results were expressed in mg of gallic acid equivalents (GAE) per kg of sample.

Total Flavonoid Content

The aluminium nitrate method was used to determine the total flavonoid content of dried sea buckthorn pomace (Mohammadzadeh et al. 2007). Methanolic extract (0.5 mL) was added to a test tube containing 0.1 mL of 10% aluminum nitrate, 0.1 mL of 1 M aqueous potassium acetate and 4.3 mL methanol. After 40 min reaction time at room temperature, the absorbance was measured at 415 nm using a Varian Cary 50 UV-Vis spectrophotometer (Varian Co., USA). Quercetin was used as standard and results were expressed in milligrams of quercetin equivalents (QE) per kg of sample.

ABTS Free Radical Scavenging Activity

The free radical scavenging activity was determined using the ABTS (2,29-azinobis-3-ethylbenzothiazoline-6-sulfonic acid) assay described by Re et al. (1999). The ABTS cation radical solution (ABTS+) was prepared by reacting 5 mL of a 7.0 mM ABTS solution and 88 µL of a 145 mM potassium persulfate solution. The mixture was allowed to react at room temperature, in the dark for 16 h. The ABTS+ solution was then diluted with 80% ethanol to obtain an absorbance of 0.700 ± 0.05 at 734 nm. Twelve milliliters of ABTS+ solution were added to 120 µL of sample extract and vigorously mixed in a Vortex. After 6 min, the absorbance at 734 nm was read using ethyl alcohol as blank. Lower levels of absorbance indicate higher antioxidant activity. The calibration curve was constructed

using standard solutions of Trolox (100–2000 µM Trolox/L) in ethanol and the results were expressed in mM Trolox per kg of sample.

Carotenoid Composition

Carotenoids were extracted from 0.5 g sample with 10 mL of petroleum ether:methanol:ethyl acetate (1:1:1, v/v/v) containing 0.1% butyl hydroxytoluene (BHT) by homogenizing for 5 min at 2500 rpm in a Vortex. After centrifugation for 6 min at 6000 rpm the supernatant was collected. The residue was extracted following the same procedure until the supernatant was colorless. The combined supernatants were washed by adding 10 mL of 5% NaCl solution, mixing vigorously and incubating for 30 min until two layers were separated. The upper layer was collected, evaporated to dryness under N2 flow and then re-dissolved in 2 mL of acetonitrile:methanol:ethyl acetate (60:20:20, v/v/v) containing butylated hydroxytoluene (BHT) (1% w/v) for the HPLC analysis. The final solution was filtered through 0.45 µm membrane filters for HPLC injection.

HPLC analyses were performed on a Finningan Surveyor Plus system (Thermo Electron Corporation, San Jose, CA). Separation was achieved by a reversed-phase Hypersil Gold C18 column (5 mm particle size, 250 × 4.6 mm) provided by Thermo Electron Corporation termostatated at 20 °C. The mobile phase system comprised acetonitrile:methanol (95:5, v/v) (A), acetonitrile:methanol:ethyl acetate (60:20:20, v/v/v) (B) and water (C). Carotenoids were eluted at a flow rate of 1.5 mL/min with the following gradient: 96% A and 4% C in the beginning, maintained for 10 min, changed linearly to 100% B in 13 min, maintained 5 min and returned to 96% A and 4% C in 2 min. The detector was set at 450 nm. Quantification was performed using Chrom Quest 4.2 software by comparing peak area with those of known standards.

The total carotenoid content was estimated spectrophotometrically in the extracts obtained as described above using β-carotene for the standard curve drawing. The results were expressed as mg of β-carotene per kg of sample.

Statistical Analysis

The measurements were performed in triplicate and results were expressed as mean value ± standard deviation. Statistical analysis was performed using Statgraphic Centurion XVI software (StatPoint Technologies, Warrenton, VA, USA).

Results and Discussion

The data on the proximate composition of dried sea buckthorn pomace are presented in Table 1. One of the main characteristics of sea buckthorn fruits is their high fat content. Unlike other fruits, sea buckthorn synthesizes and accumulates lipids in all parts of the fruit, and therefore it is possible to obtain three types of oil, depending on whether it is extracted from the pulp, seed or skin (Li and Beveridge 2003). However, given the difficulty of separating the skin from the pulp, normally these two oils are not distinguished, and they are called oil of the pulp or oil of the soft parts.

According to Yang and Kallio (2005), the oil content of the seeds is generally constant (about 10%) and independent of morphological characteristics and origins, although higher values (up to 15–16%) have been reported for some cultivars. On the other hand, the lipid content of the soft tissues (pulp and skin) varies considerably (from 1 to 35%) depending on the origin and other factors such as the time of harvesting, the application of inorganic fertilizers, the state of maturity of the fruits and the climate (Kallio et al. 1999; Yang and Kallio 2002).

The results from this study showed higher crude fat content (20.05%) than reported by Nuernberg et al. (2015) (14.5%) or by Ben-Mahmoud et al. (2014) (17.14%) in sea buckthorn fruit residues obtained after pressing the berries to extract the juice and then dried. Differences in the lipid content of dried sea buckthorn pomace may be attributed to the different subspecies, geographical and climate factors, harvesting time and processing of the berries, as well as the extraction and the drying methods (Yang and Kallio 2002).

Results on crude protein content (14.89%) were less than those reported by Ben-Mahmoud et al. (2014) (20.87%) but in good agreement with data reported by Nuernberg et al. (2015) (14.6%). However, results on crude fiber (19.86%) and ash (1.84%) content agreed well with data reported by Ben-Mahmoud et al. (2014) (18.13 and 2.02%, respectively).

According to Chen (1988), sea buckthorn fruits contain 18 kinds of free amino acids. Of these, eight amino acids (threonine, valine, methionine, leucine, lysine, tryptophan, isoleucine, and phenilalanine) are essential for the human body. The amino acid content of dried sea buckthorn pomace is given in Table 2. Glutamic acid was the most abun-

Table 1 Proximate composition of dried sea buckthorn pomace

Component (g/kg)	Dried sea buckthorn pomace
Dry matter	926.6 ± 11.8
Crude protein	148.9 ± 6.5
Crude fat	200.5 ± 5.3
Crude fiber	198.6 ± 8.9
Ash	18.4 ± 0.8

Table 2 Amino acids content of dried sea buckthorn pomace (g/kg)

Amino acids	Dried sea buckthorn pomace
Aspartic acid	17.2 ± 0.5
Glutamic acid	23.7 ± 1.1
Serine	8.5 ± 0.3
Glycine	5.1 ± 0.2
Threonine	5.2 ± 0.2
Arginine	13.1 ± 0.4
Alanine	6.8 ± 0.3
Tyrosine	4.4 ± 0.2
Valine	6.4 ± 0.3
Phenylalanine	7.9 ± 0.3
Isoleucine	7.1 ± 0.2
Leucine	11.6 ± 0.3
Lysine	7.2 ± 0.2
Cystine	1.5 ± 0.1
Methionine	4.7 ± 0.2
Total amino acids	130.4 ± 4.8

dant in dried sea buckthorn pomace (23.7 g/kg) followed by aspartic acid (17.2 g/kg). The essential amino acids represented 38.42% of total amino acids, the most abundant being leucine, followed by phenylalanine and lysine. Results of lysine content (7.2 g/kg) were in agreement with data reported by Ben-Mahmoud et al. (2014) (7.85 g/kg) for dried sea buckthorn fruit residues, but differed in content of methionine (4.7 g/kg as against 2.82 g/kg).

Table 3 Fatty acids profile of dried sea buckthorn pomace (% of total fatty acids)

Fatty acids		Dried sea buckthorn pomace
Capric	C 10:0	0.05 ± 0.01
Myristic	C 14:0	0.62 ± 0.02
Pentadecanoic	C 15:0	0.11 ± 0.03
Palmitic	C 16:0	29.22 ± 1.25
Palmitoleic	C 16:1	28.75 ± 1.13
Heptadecanoic	C 17:0	0.13 ± 0.01
Heptadecenoic	C 17:1	0.10 ± 0.01
Stearic	C 18:0	1.05 ± 0.08
Oleic cis	C 18:1	24.23 ± 0.94
Linoleic cis	C 18:2n6	8.96 ± 0.41
Linolenic α	C 18:3n3	6.02 ± 0.28
Octadecatetraenoic	C18:4n3	0.46 ± 0.02
Eicosadienoic	C20(2n6)	0.26 ± 0.01
Other fatty acids		0.04 ± 0.01
Fatty acids profile		
Saturated fatty acids (SFA)		31.18 ± 1.40
Monounsaturated fatty acids (MUFA)		53.08 ± 2.08
Polyunsaturated fatty acids (PUFA), of which:		15.70 ± 0.72
■ *n*-3		6.48 ± 0.30
■ *n*-6		9.22 ± 0.42
n-6/*n*-3		1.42 ± 0.12

The composition of fatty acids of dried sea buckthorn pomace is shown in Table 3. This composition is determined by the fatty acid content of seed oil and that of the oil from the soft parts (peel and residual pulp) that are significantly different. Thus, the seed oil is characterized by its high content of unsaturated fatty acids (85–90%), including two essential fatty acids, linoleic acid (18: 2*n*-6) and α-linolenic acid (18: 3*n*-3), together representing up to 70%. The proportions of these two fatty acids are generally 30–40% and 20–35%, respectively (Yang and Kallio 2002). Other fatty acids normally found in seeds are oleic (18: 1*n*-9, 13–30%), palmitic (16: 0, 7–20%), stearic (18: 0, 2–9%) and vaccenic (18: 1*n*-7, 2–4%) acids (Yang and Kallio 2002, 2005).

The oil from the soft parts is characterized by its high content of saturated and monounsaturated fatty acids. It mainly comprises palmitoleic (16: 1*n*-7, 16–54%), palmitic (16: 0, 17–47%), oleic (18: 1*n*-9, 2–35%) acids, and small proportions of linoleic acid (18: 2*n*-6, <10%), α-linolenic (18: 3*n*-3, <3%) and stearic (18: 0, 0.2–3%) acids (Yang and Kallio 2002; Ranjith et al. 2006; Cenkowski et al. 2006).

Dried sea buckthorn pomace had a low ratio of *n*-6/*n*-3 polyunsaturated fatty acids (PUFA) of 1.42% and a high concentration of monounsaturated fatty acids (MUFA, 53.08% of total fatty acids), as a result of the high content of oleic and palmitoleic acids. These results are in good agreement with available data from previous studies (Nuernberg et al. 2015). An increased intake of palmitoleic acid in the diet could have hypocholesterolemic and hypotriglyceridemic effects, and reduce the stroke risk (Yang and Kallio 2002). In addition, the high intake of oleic acid can induce lowered blood fat, improved HDL to LDL cholesterol ratios and inhibition of inflammatory processes in blood vessels (Baum et al. 2012; Miura et al. 2013).

Sea buckthorn berries are a good source of mineral elements like Ca, P, Fe and especially K which is the most abundant (Bal et al. 2011). The mineral elements investigated in this study were calcium, iron, manganese, copper and zinc (Table 4). Calcium content found in dried sea buckthorn pomace (724 mg/kg) was higher than values reported by Arif et al. (2010) (40–100 mg/kg) in fresh berries. Similarly, the levels found for iron (62.9 mg/kg), manganese (12.6 mg/kg) and zinc (22.3 mg/kg) were higher than those

Table 4 Minerals content of dried sea buckthorn pomace (mg/kg)

Mineral	Dried sea buckthorn pomace
Calcium	724 ± 15.6
Iron	62.9 ± 2.6
Manganese	12.6 ± 0.5
Copper	8.3 ± 0.3
Zinc	22.3 ± 1.0

Table 5 Total phenolics, total flavonoids, total carotenoid content and free radical scavenging activity of dried sea buckthorn pomace

Component	Dried sea buckthorn pomace
Total phenolic content (mg GAE/kg)	2791.2 ± 26.6
Total flavonoid content (mg QE/kg)	482.5 ± 20.2
Total carotenoid content (mg/kg)	245.6 ± 11.5
Antioxidant activity (mmol Trolox/kg)	82.96 ± 3.6

reported in fresh sea buckthorn berries (Bal et al. 2011). However, results are in agreement with those reported by Sabir et al. (2005) who found calcium and iron contents of 700–1250 mg/kg and 40–225 mg/kg, respectively in dried berries.

Sea buckthorn berries are rich sources of phenolic compounds. The major polyphenol groups identified in berries are flavonols and condensed tannins (Rösch et al. 2003; Teleszko et al. 2015), compounds that give them a very high antioxidant potential. A high phenolic content was found in our study (2.79 g GAE/kg) (Table 5), in agreement with the results reported by Kitrytė et al. (2017) in dried sea buckthorn pomace. However, the antioxidant capacity of sea buckthorn berries is attributed to the combined action of ascorbic acid, polyphenols (phenolic acids and flavonoids) and carotenoids (Gao et al. 2000).

Carotenoids are the molecules responsible for the color of sea buckthorn berries that can vary from yellow to red. The quantity of carotenoids is subject to numerous variations, particularly depending on the variety, climate and degree of ripening of the fruit (Andersson et al. 2008). Carotenoids present in the berry, pulp and seed oil, and in sea buckthorn residues, are found in the form of carotenes, xanthophylls but also in the form of esterified carotenes (Raffo et al. 2004; Pintea et al. 2005).

Pulp oils are richer in carotenoids than seed oils, which usually contain small amounts of carotenoids (20 to 85 mg/100 g of oil) (Li and Beveridge 2003). According to Yang and Kallio (2002), β-carotene represents approximately 15–55% of total carotenoids, varying its typical contents in the range 100–500 and 20–100 mg/100 g in pulp and seed oils respectively.

In the present study, a total carotenoid content of 245.6 mg/100 g was found in dried sea buckthorn pomace

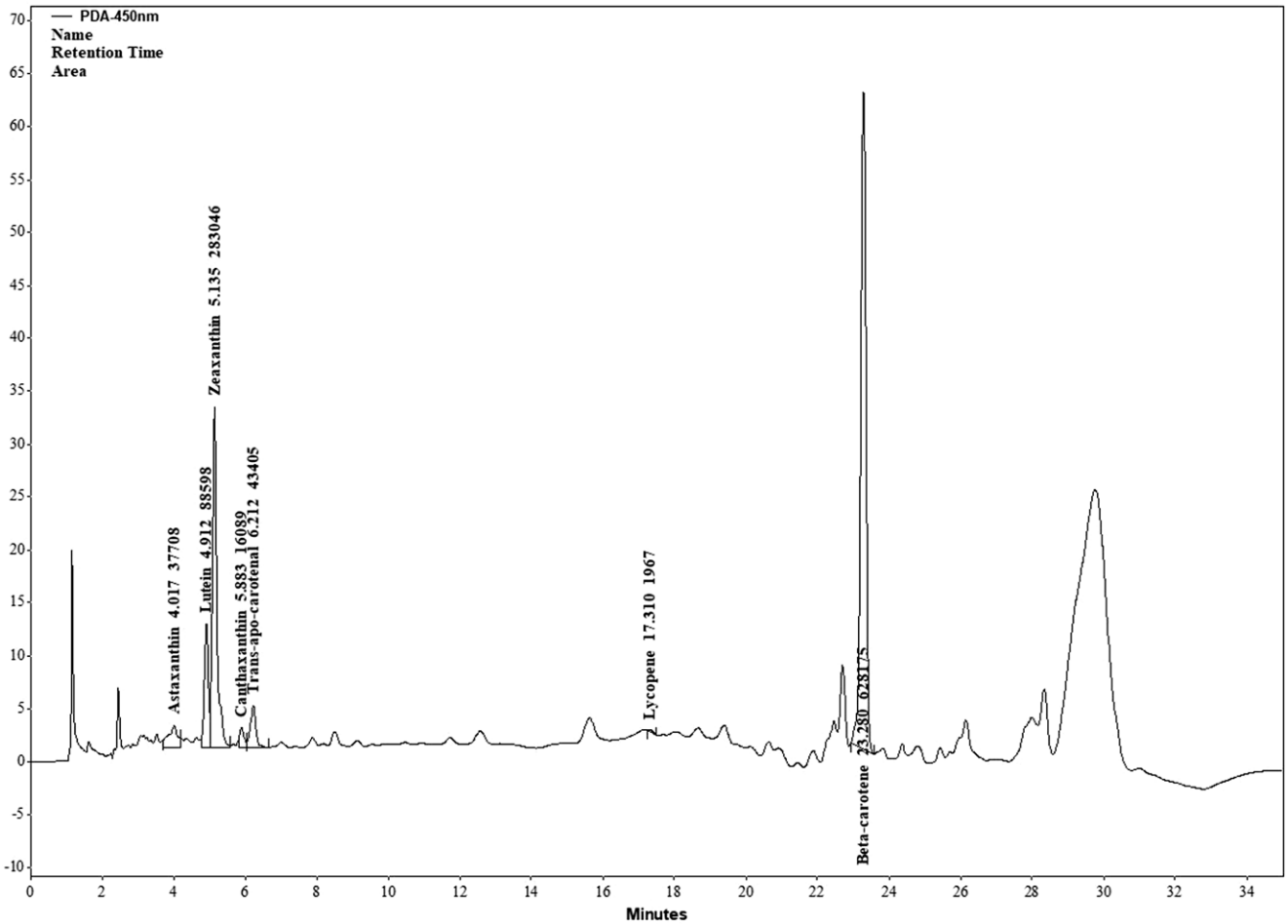

Fig. 1 HPLC chromatogram at λ = 450 nm of carotenoids from a sample of dried sea buckthorn pomace

Table 6 Carotenoid content of dried sea buckthorn pomace (mg/kg)

Phenolic compounds	Dried sea buckthorn pomace
Astaxanthin	4.78 ± 0.23
Lutein	6.96 ± 0.41
Zeaxanthin	69.60 ± 1.56
Canthaxanthin	1.36 ± 0.22
Trans-β-apo-8′-carotenal	2.36 ± 0.12
Lycopene	0.91 ± 0.06
β-Carotene	80.76 ± 3.58
Total carotenoids	166.73 ± 6.18

(Table 5). Teleszko et al. (2015) reported a total carotenoids content between 6.19 and 23.91 mg/100 g fresh weight in sea buckthorn (*Hippophae rhamnoides* L.) berries. Fig. 1 shows an HPLC chromatogram at 450 nm of carotenoids from a sample of dried sea buckthorn pomace. The major pigments were β-carotene (80.76 mg/kg) and zeaxanthin (69.60 mg/kg), but other ubiquitous carotenoids such as lutein, astaxanthin, trans-β-apo-8′-carotenal, canthaxanthin and lycopene were also quantified (Table 6). Raffo et al. (2004) found also that the main carotenoids in seabuckthorn berries were zeaxanthin (30–150 mg/kg) and β-carotene (3–50 mg/kg), but the extent of carotenoid accumulation and the carotenoid profile was affected by genotype.

Conclusions

The results of this study demonstrated that sea buckthorn pomace has a high nutritional value given by its high fat and protein content, its provision of essential amino acids (38.42% of total amino acids) and its fatty acid profile, characterized by the predominance of the monounsaturated fatty acids (53.08% of total fatty acids), the high content of polyunsaturated fatty acids (15.70%), and by the low ratio of *n*-6/*n*-3 fatty acids (1.42). These findings suggest that sea buckthorn pomace may be a valuable food ingredient or could be used as a nutritional feed supplement. In addition, sea buckthorn by-products were found to contain high levels of phenolic compounds and carotenoids and demonstrated high antioxidant activity. The content of bioactive compounds makes sea buckthorn pomace applicable for use as functional food supplement as a source of natural antioxidants in the medical and pharmaceutical industries.

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Conflict of interest V. Nour, T.D. Panaite, A.R. Corbu, M. Ropota and R.P. Turcu declare that they have no competing interests.

References

Andersson SC, Olsson ME, Johansson E, Rumpunen K (2008) Carotenoids in sea buckthorn (Hippophae rhamnoides L.) berries during ripening and use of pheophytin a as a maturity marker. J Agric Food Chem 57(1):250–258

Arif S, Ahmed SD, Shah AH, Hassan L, Awan SI, Hamid A, Batool F (2010) Determination of optimum harvesting time for vitamin C, oil and mineral elements in berries sea buckthorn (Hippophae rhamnoides). Pak J Bot 42:3561–3568

Bal LM, Meda V, Naik SN, Satya S (2011) Sea buckthorn berries: a potential source of valuable nutrients for neutraceuticals and cosmoceuticals. Food Res Int 44:1718–1727

Baum SJ, Kris-Etherton PM, Willett WC, Lichtenstein AH, Rudel LL, Maki KC, Whelan J, Ramsden CE, Block RC (2012) Fatty acids in cardiovascular health and disease: a comprehensive update. J Clin Lipidol 6:216–234

Ben-Mahmoud Z, Mohamed MS, Bláha J, Lukešová D, Kunc P (2014) The effect of sea buckthorn (Hippophae rhamnoides L.) residues in compound feeds on the performance and skin color of broilers. Ind J Anim Res 48(6):548–555

Beveridge T, Harrison JE, Drover J (2002) Processing effects on the composition of sea buckthorn juice from Hippophae rhamnoides L. cv. Indian Summer. J Agric Food Chem 50:113–115

Beveridge T, Li TSC, Oomah D, Smith A (1999) Sea buckthorn products: manufacture and composition. J Agric Food Chem 47(9):3480–3488

Cenkowski S, Yakimishen R, Przybylski R, Muir WE (2006) Quality of extracted sea buckthorn seed and pulp oil. Can Biosyst Eng 48:3.9–3.16

Chemat F, Périno-Issartier S, Loucif L, Elmaataoui M, Mason TJ (2012) Enrichment of edible oil with sea buckthorn by-products using ultrasound-assisted extraction. Eur J Lipid Sci Tech 114:453–460

Chen T (1988) Studies of the biochemical composition of Hippophae and its quality assessment in Gansu Province. Hippophae 1:19–26

Chen C, Xu XM, Chen Y, You MY, Wen FY, Zhan Y (2013) Identification, quantification, and antioxidant activity of acylated flavonol glycosides from sea buckthorn (Hippophae rhamnoides ssp. sinensis). Food Chem 141:1573–1579

Dulf FV, Andrei S, Bunea A, Socaciu C (2012) Fatty acids and phytosterol contents of some Romanian wild and cultivated berry pomaces. Chem Papers 66:925–934

Eccleston C, Baoru Y, Tahvonen R, Kallio H, Rimbach GH, Minihane AM (2002) Effects of an antioxidant rich juice (Sea buckthorn) on risk factors for coronary heart disease in humans. J Nutr Biochem 13:346–354

Gao X, Ohlander M, Jeppsson N, Bjork L, Trajkovski V (2000) Changes in antioxidant effects and their relationship to phytonutrients in fruits of sea buckthorn (Hippophae rhamnoides L.) during maturation. J Agric Food Chem 48(5):1485–1490

Gao ZL, Gu XH, Cheng FT, Jiang FH (2003) Effect of Sea buckthorn on liver fibrosis: a clinical study. World J Gastroenterol 9:1615–1617

Johansson AK, Korte H, Yang B, Stanley JC, Kallio HP (2000) Sea buckthorn berry oil inhibits platelet aggregation. J Nutr Biochem 11:491–495

Kallio H, Yang B, Peippo P, Tahvonen R, Pan R (2002) Triacylglycerols, glycerophospholipids, tocopherols, and tocotrienols in berries and seeds of two subspecies (ssp. sinensis and mongolica) of sea buckthorn (Hippophae rhamnoides). J Agric Food Chem 50(10):3004–3009

Kallio H, Yang B, Tahvonen R, Hakala M (1999) Composition of sea buckthorn berries of various origins. In: Proceedings of International Workshop on Sea Buckthorn Beijing, 28 August-2 September, 1999, pp 13–19

Kant V, Mehta M, Varshneya C (2012) Antioxidant potential and total phenolic contents of seabuckthorn (Hippophae rhamnoides) pomace. Free Radic Antiox 2(4):79–86

Kitrytė V, Povilaitis D, Kraujalienė V, Šulniūtė V, Pukalskas A, Venskutonis PR (2017) Fractionation of sea buckthorn pomace and seeds into valuable components by using high pressure and enzyme-assisted extraction methods. LWT Food Sci Technol 85:534–538

Kruczek M, Świderski A, Mech-Nowak A, Król K (2012) Antioxidant capacity of crude extracts containing carotenoids from the berries of various cultivars of Seabuckthorn (Hippophae rhamnoides L.). Acta Biochim Pol 59(1):135–137

Li TSC (2003) Taxonomy, natural distribution and botany. In: Li TSC, Beveridge T (eds) Sea buckthorn (Hippophae rhamnoides L.): Production and utilization. NRC Research Press, Ottawa, pp 7–11

Li TSC, Beveridge THJ (2003) Sea buckthorn (Hippophae rhamnoides L.): production and utilization. National Research Press, Ottawa, p 133

Lougas T, Veskus T, Martverk K, Täht R, Rada K, Vokk R (2005) Sea buckthorn and its new field use. IntraFood—EFFOST Conference, pp 719–722

Miura K, Stamler J, Brown IJ, Ueshima H, Nakagawa H, Sakurai M, Chan Q, Appel LJ, Okayama A, Okuda N, Curb JD, Rodriguez BL, Robertson C, Zhao L, Elliott P (2013) Relationship of dietary monounsaturated fatty acids to blood pressure: the international study of macro/micronutrients and blood pressure. J Hypertens 31:1144–1150

Mohammadzadeh S, Sharriatpanahi M, Hamedi M, Amanzadeh Y, Ebrahimi SES, Ostad SN (2007) Antioxidant power of Iranian propolis extract. Food Chem 103(3):729–733

Nuernberg K, Nuernberg G, Priepke A, Dannenberger D (2015) Sea buckthorn pomace supplementation in the finishing diets of pigs—are there effects on meat quality and muscle fatty acids? Arch Anim Breed 58:107–113

Périno-Issartier S, Human Z, Abert-Vian M, Chemat F (2011) Solvent free microwave-assisted extraction of antioxidants from sea buckthorn (Hippophae rhamnoides) food by-products. Food Bioprocess Technol 4:1020–1028

Pintea A, Varga A, Stepnowski P, Socaciu C, Culea M, Diehl HA (2005) Chromatographic analysis of carotenol fatty acid esters in Physalis alkekengi and Hippophae rhamnoides. Phytochem Anal 16(3):188–195

Radenkovs V, Püssa T, Juhnevica-Radenkova K, Anton D, Seglina D (2018) Phytochemical characterization and antimicrobial evaluation of young leaf/shoot and press cake extracts from Hippophae rhamnoides L. Food Biosci 24:56–66

Raffo A, Paoletti F, Antonelli M (2004) Changes in sugar, organic acid, flavonol and carotenoid composition during ripening of berries of three seabuckthorn (Hippophae rhamnoides L.) cultivars. Eur Food Res Technol 219(4):360–368

Ranjith A, Kumar KS, Venugopalan VV, Arumughan C, Sawhney RC, Singh V (2006) Fatty acids, tocols and carotenoids in pulp oil of three Seabuckthorn species (Hippophae rhamnoides, H. salicifolia and H. tibetana) grown in the Indian Himalayas. J Am Oil Chem Soc 83:359–364

Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, Rice-Evans C (1999) Antioxidant activity applying an improved ABTS radical cation decolorization assay. Free Radic Biol Med 26(9–10):1231–1237

Rösch D, Bergmann M, Knorr D, Kroh LW (2003) Structure-antioxidant efficiency relationships of phenolic compounds and their contribution to the antioxidant activity of sea buckthorn juice. J Agric Food Chem 51(15):4233–4239

Rösch D, Krumbein A, Kroh L-W (2004) Antioxidant gallocatechins, dimeric and trimeric proanthocyanidins from sea buckthorn (Hippophae rhamnoides) pomace. Eur Food Res Technol 219:605–613

Sabir SM, Maqsood H, Ahmed SD, Shah AH, Khan MQ (2005) Chemical and nutritional constituents of sea buckthorn (Hippophae rhamnoides ssp. turkestanica) berries from Pakistan. Ital J Food Sci 17:455–462

Singleton VL, Rossi JA (1965) Colorimetry of total phenolics with phosphomolybdic–phosphotungstic acid reagent. Am J Enol Viticult 16:144–158

Stahl W, Sies H (2003) Antioxidant activity of carotenoids. Mol Aspects Med 24:345–351

Süleyman H, Demirezer LÖ, Büyükokuroglu ME, Akcay MF, Gepdiremen A, Banoglu ZN, Göçer F (2001) Antiulcerogenic effect of Hippophae rhamnoides L. Phytother Res 15(7):625–627

Suryakumar G, Gupta A (2011) Medicinal and therapeutic potential of Sea buckthorn (Hippophae rhamnoides L.). J Ethnopharmacol 138:268–278

Teleszko M, Wojdylo A, Rudzinska M, Oszmianski J, Golis T (2015) Analysis of lipophilic and hydrophilic bioactive compounds content in sea buckthorn (Hippophae rhamnoides L.) berries. J Agric Food Chem 63:4120–4129

Varzaru I, Untea AE, Martura T, Olteanu M, Panaite TD, Schitea M, Van I (2013) Development and validation of an RP-HPLC method for methionine, cystine and lysine separation and determination in corn samples. Rev Chim 67(7):673–679

Yang B, Kallio H (2002) Composition and physiological effects of sea buckthorn (Hippophae) lipids. Trends Food Sci Technol 13(5):160–167

Yang B, Kallio H (2005) Lipophilic components of sea buckthorn (Hippophae rhamnoides L.) seeds and berries. In: Singh V (ed) Sea buckthorn (Hippophae L.): a multipropose wonder plant, vol 2. Daya Publishing House, New Delhi, pp 70–97

Yang B, Kallio HP (2001) Fatty acid composition of lipids in sea buckthorn (Hippophae rhamnoides L.) berries of different origins. J Agric Food Chem 49:1939–1947

Zadernowski R, Naczk M, Amarowicz R (2003) Tocopherols in sea buckthorn (Hippophae rhamnoides L.) berry oil. J Am Oil Chem Soc 80:55–58

Zeb A (2004a) Chemical and nutritional constituents of sea buckthorn juice. Pak J Nutr 3(2):99–106

Zeb A (2004b) Important therapeutic uses of sea buckthorn (Hippophae): a review. J Biol Sci 4(5):687–693