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ORIGINAL ARTICLE

BENEFITS OF SEA BUCKTHORN JUICE CONSUMPTION IN WOMEN OF PRODUCTIVE AGE WITH HYPERCHOLESTEROLEMIA

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ABSTRACT

Background. Sea buckthorn contains almost 200 nutrients and bioactive substances, including phenolic compounds such as flavonoids, vitamins, proteins, amino acids, minerals, alkaloids, chlorophyll derivatives, amines, organic acids, fatty acids and phytosterols. Human and animal studies suggest that sea buckthorn may have a variety of beneficial effects: cardioprotective, antiatherogenic, antioxidant, anticancer, immunomodulatory, antibacterial, antiviral and anti-inflammatory.

Objective. The aim of this study was to evaluate the effect of regular consumption of 100% sea buckthorn juice on the risk factors of cardiovascular diseases in women of productive age with hypercholesterolemia.

Material and Methods. A clinical study involved 19 women with a mean age of 54.06 ±2.97 years who consumed 50 mL of sea buckthorn juice daily for 8 weeks. Anthropometric and biochemical parameters in blood serum were monitored before the start of sea buckthorn consumption and after 8 weeks of consumption. Body composition was determined using an multifrequency analyzer InBody720. Routine biochemical analyzes were performed by standard methods in an accredited laboratory of the University Hospital by automatic biochemical analyzer BioMajesty JCA-BM6010/C. Statistical comparison between individual measurements was performed using a paired t-test, using Statistica Cz version 10 (TIBCO Software, Inc., Palo Alto, CA, USA).

Results. We observed significant decrease of body weight, body mass index ($P<0.05$), body fat and visceral fat ($P<0.001$) after 8 weeks of consumption of 100% sea buckthorn juice. In this intervention study, we observed a significant decrease in low-density cholesterol ($P<0.05$) and a significant increase in high-density cholesterol ($P<0.001$). The level of triglycerides was similar at the end of the study ($P>0.05$). After the intervention, we observed decrease of orosomucoid, immunoglobulin A, immunoglobulin G, immunoglobulin M ($P<0.001$) and C-reactive protein ($P<0.01$).

Conclusions. The obtained results support the hypothesis that the daily consumption of sea buckthorn juice for eight weeks could contribute to the beneficial effects of reducing the risk of cardiovascular diseases, such as the reduction of body and visceral fat, LDL-C, CRP and the increase of HDL-C.

Key words: *sea buckthorn juice, cardiovascular diseases, hypercholesterolemia, obesity, cholesterol*

INTRODUCTION

Cardiovascular diseases (CVD) have emerged as a major cause of mortality and morbidity worldwide [47]. Dyslipidemia is a major modifiable contributor to cardiovascular diseases [31], elevated blood cholesterol accounts for nearly one third of ischemic heart diseases [13, 47]. Reduction in excess calories and improvement in dietary composition may prevent many primary and secondary cardiovascular events [56]. In recent years, research has focused primarily on natural products with beneficial effects, and increased attention is being paid to foods with demonstrable and effective antioxidant activity [41].

Many studies demonstrate that the fruits and leaves of some berry plants biosynthesize phytochemicals with antioxidant activity and can be used as a natural source of free radical scavenging compounds [26, 27, 40]. A diet rich in berries is thought to play an important role in preventing metabolic diseases associated with obesity [42]. The beneficial effects of berries may be mediated by the antioxidant and anti-inflammatory properties of polyphenols [23].

Sea buckthorn (*Hippophae* in Latin) belongs to the *Elaeagnaceae* family [54]. Around 150 species, subspecies and varieties of sea buckthorn have been identified within Europe and Asia. They differed in the habitat of the bush, the appearance of the berries

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and their utility value [7]. The most important and widespread in Europe is the sea buckthorn *Hippophae rhamnoides* [55]. *Hippophae* fruits are called third generation fruits [18]. Sea buckthorn fruits, leaves, oil and other products are a source of many bioactive substances, including phenolic compounds, such as flavonoids, i.e. rutin, quercetin, kaempferol, or myricetin [6], vitamins (tocopherols, carotenoids, ascorbic acid, folate, vitamins B_1, B_2 and K), proteins, amino acids and minerals (Fe, Ca, P and K) [2, 14, 24, 59]. In addition, the plant contains organic acids (quinic acid, malic acid, oxalic acid and tartaric acid) [5, 20], fatty acids, especially unsaturated fatty acids (oleic acid, linoleic acid, linolenic acid) and phytosterols [5, 32]. The main group of phenolic compounds are flavonols, a group of flavonoids that have been identified in fruits, with an average content of 311.5 mg/100 g fresh weight [46]. One of the most interesting features is that sea buckthorn contains high concentrations of vitamin C [50]. The content of vitamin C per 100 g of sea buckthorn fruit is 600 mg, which is significantly more than in rose hips (250-800 mg), black currants (120-215 mg) or raspberries (15-30 mg) [24]. Chemical compounds in different parts of *Hippophae rhamnoides* may vary depending on climatic conditions, their origin and extraction methods [35]. Sea buckthorn fruits are interesting not only from a chemical point of view, but also from a biological and therapeutic point of view for their antioxidant, antitumor, hepato-protective and immunological properties [59]. Human and animal studies suggest that sea buckthorn may have a variety of beneficial effects: cardioprotective, antiatherogenic, antioxidant, anticancer, immunomodulatory, antibacterial, antiviral, and anti-inflammatory [4, 51]. The antioxidants present in sea buckthorn activate the transcription factor nuclear factor E2 (Nrf-2) and inhibit the nuclear factor kappa B (NF-kB) redox signaling pathway, which in turn activates antioxidant enzymes that are responsible for antioxidant activity and are considered one of the mechanisms of action antioxidant activity of sea buckthorn [19]. The anti-inflammatory activity of sea buckthorn can be attributed to ursolic acid, oleanolic acid, citric acid derivatives and flavonoids. Its anti-inflammatory mechanism of action may be related to the inhibition of the expression of pro-inflammatory cytokines and the reduction of the production of pro-inflammatory mediators [51]. *Jaśniewska* and *Diowksz* [17] report that sea buckthorn flavonoids help lower cholesterol levels, improve cardiac function, and protect endothelial cells from damage caused by oxidized low-density lipoproteins. Sea buckthorn polyphenols suppress the expression of cyclins, thereby arresting the cell cycle in the G1 phase and affecting the further proliferation of colon cancer [53]. Biochemical and histopathological studies have shown that sea buckthorn flavonoid extract significantly improves biomarkers including triglycerides aspartate aminotransferase and alanine aminotransferase in serum and liver [51]. Recent attention has focused on the use of fruit juices as a concentrated source of antioxidants. Drinking juice is an effective way to promote fruit and vegetable consumption and is very popular in many countries [3, 10, 43, 45]. A high amount of evidence shows that juice, as part of a balanced diet, contributes to a significant reduction in the risk of many diseases, such as cancer, neurodegenerative diseases and cardiovascular diseases [3, 33, 38].

The aim of this study was to evaluate the effect of regular consumption of 100% sea buckthorn juice (SBJ) on the risk factors of cardiovascular diseases in women of productive age with hypercholesterolemia.

MATERIAL AND METHODS

The study included 19 hypercholesterolemic women of productive age from 50 to 61 years old, with a mean age of 54.06 ± 2.97 years, who participated in an 8-week intervention program. Body composition (body weight – BW, body fat mass – BFM, body mass index – BMI, visceral fat area – VFA, skeletal muscle mass – SMM, fat-free mass – FFM, waist to hip ratio – WHR, intracellular water – ICW, extracellular water – ECW and total body water – TBW), blood pressure (systolic blood pressure – SBP and diastolic blood pressure – DBP), lipid profile (total cholesterol – T-C, low density cholesterol – LDL-C, high density cholesterol – HDL-C, triglycerides – TG), inflammation markers (C-reactive protein – CRP, interleukin-6 – IL-6, orosomucoid – ORM, immunoglobulin A – IgA, immunoglobulin G – IgG and immunoglobulin M – IgM), kidney and liver markers (alanine aminotransferase – ALT, aspartate aminotransferase – AST, gamma glutamyl transferase – GGT, urea, creatinine and uric acid) were monitored before the start of SBJ consumption and after 8 weeks of consumption.

Ethical aspects

The study was conducted in accordance with the Declaration of Helsinki and approved by the Ethics Committee at the Specialized Hospital St. Zoerardus Zobor, NPO Nitra, Slovak Republic (protocol number 3/101921/2021).

Dietary intervention

Volunteers consumed 50 mL of commercial 100% SBJ as recommended by the manufacturer for 8 weeks as part of their regular diet. The juice was provided by ZAMIO Ltd., Trhovište, Slovakia. Juice composition (g/100 mL): fats – 3.2; of which saturated fatty acids –

0.7; carbohydrates – 5.1; of which sugars – 4.7; proteins – 1.0. Total phenolic content – 1.56 mg gallic acid equivalents (GAE)/g, vitamin C – 385 mg/100 g and total carotenoid – 64.79 mg/100 g. The participants were instructed to maintain their normal eating habits during the study, to refrain from consuming dietary supplements and not to modify their physical activity.

Anthropometric parameters

Body height was measured on a Tanita WB-300 ambulatory electronic scale in an upright position, without shoes. We used multi-frequency bioelectrical impedance analysis (MFBIA) – InBody 720 (Biospace Co. Ltd., Seoul, Korea) to diagnose body composition. Blood pressure was determined with a digital electronic sphygmomanometer Omron M7 Intelli IT, HEM-7361T-EBK (Omron Healthcare, Tokyo, Japan).

Biochemical parameters

Venous blood was collected in the morning after 8 hours of fasting in a standard way. After blood serum separation, routine biochemical analyzes were performed in an accredited laboratory of the University Hospital with a BioMajesty JCA-BM6010/C automatic biochemical analyzer using commercial DiaSys kits (Diagnostic Systems GmbH, Holzheim, Germany) according to the manufacturer's instructions.

Statistical analysis

Statistica Cz version 10 (TIBCO Software, Inc., Palo Alto, CA, USA) and MS Excel 2007 (Microsoft Corporation, Redmond, WA, USA) were used for statistical analysis. All data were expressed as mean ± standard deviation (SD). Statistical comparison between individual measurements was performed using a paired t-test, a value of $P < 0.05$ was considered statistically significant.

RESULTS AND DISCUSSION

Characteristics of study participants

The observed group consisted of 19 women of productive age from 50 to 61 years, with an average age of 54.06 ± 2.97 years. All women had an elevated level of total cholesterol, the average 6.41 ± 1.08 mmol.L^{-1}. From the individual values the basic statistical characteristics of the probands were calculated (Table 1).

Effect of SBJ consumption on anthropometric and biochemical parameters

Anthropometric measurements

Worldwide, the burden of morbidity and mortality from diet-related chronic diseases is increasing, driven by poor diet quality and overconsumption of calories. Fortunately, shifting current global dietary patterns towards high-quality, plant-based diets could alleviate these health and environmental burdens [15, 37] not only because of the low content of saturated fat and cholesterol, but also because of the considerable amount of micronutrients and bioactive compounds [12, 48]. Obesity is induced by chronic low-grade inflammation, which can act synergistically with oxidative stress. Thus, intake of fruits and plant extracts high in antioxidant phytochemicals has important anti-obesity activity [48]. *Huang* et al. [16] found that a vegetarian diet can be of considerable importance in weight reduction. The results of anthropometric characteristics and body structure after consumption of SBJ are shown in Table 2. Body weight and BMI of women significantly decreased during the study ($P<0.05$). The body mass index

Table 1. Basic characteristics of study participants

Characteristic	Average ± SD	Min. – max.
Age (yrs)	54.06 ± 2.97	50 – 61
BW (kg)	72.44 ± 14.59	49.2 – 100.8
BMI (kg.m^{-2})	26.13 ± 5.27	19.96 – 38.41
WC (cm)	93.10 ± 14.57	72.40 – 120.60
T-C (mmol.L^{-1})	6.41 ± 1.08	5.06 – 8.33
GLU (mmol.L^{-1})	4.92 ± 0.34	4.30 – 5.40

Data are expressed as average ± standard deviation (SD), min. – max.; BW, body weight; BMI, body mass index; WC, waist circumference; T-C, total cholesterol; GLU, glucose

Table 2. Anthropometric characteristics and blood pressure of study participants

Parameter	Baseline	Week 8	*p*-value
BW (kg)	72.43 ± 14.59	71.95 ± 14.72	0.0450
BFM (kg)	25.17 ± 11.08	24.26 ± 11.07	<0.001
BMI (kg.m^{-2})	26.13 ± 5.27	25.96 ± 5.32	0.0400
VFA (cm^2)	103.08 ± 39.90	99.46 ± 40.25	<0.001
SMM (kg)	25.89 ± 2.51	26.17 ± 2.63	0.0080
FFM (kg)	47.26 ± 4,33	47.69 ± 4,52	0.0130
ICW (kg)	21.39 ± 1.91	21.59 ± 2.03	0.0139
ECW (kg)	13.26 ± 1.29	13.36 ± 1.29	>0.05
TBW (kg)	34.65 ± 3.18	34.96 ± 3.31	0.0140
WHR	0.94 ± 0.08	0.93 ± 0.08	>0.05
SBP (mmHg)	130.60 ± 14.01	130.87 ± 14.40	>0.05
DBP (mmHg)	85.33 ± 9.62	85.80 ± 9.50	>0.05

Data are expressed as average ± standard deviation (SD); SBJ, sea buckthorn juice; BW, body weight; BFM, body fat mass; BMI, body mass index; VFA, visceral fat area; SMM, skeletal muscle mass; FFM, fat-free mass; ICW, intracellular water; ECW, extracellular water; TBW, total body water; WHR, waist to hip ratio; SBP, systolic blood pressure; DBP, diastolic blood pressure

does not express the distribution of fat in the body. In the assessment of anthropometric indicators, we also focused on the assessment of visceral fat (VFA). We consider VFA as another indicator of health and obesity status. It is one of the important factors in the assessment of cardiometabolic risk, which correlates with the components of the metabolic syndrome in men and women, even with a normal BMI indicating the absence of obesity [1]. Abdominal obesity was observed in 7 women (36.8%) in the monitored group. In the whole group, after consumption of SBJ there was a significant decrease of VFA ($P<0.001$). Likewise, a WHR index higher than 0.85 reflects a risk for the development of metabolic diseases. In our monitored group, the WHR index was similar after 8 weeks of nutritional intervention ($P>0.05$). Weight reduction is necessary for the treatment of obesity. However, some treatments may cause a reduction on the muscular mass, reducing metabolic waste, and compromising weight loss and/or maintenance of the lost weight [29]. The current study demonstrated that taking SBJ for 8 weeks caused a significant increase of SMM and FFM ($P<0.05$). From the results of *Lehtonen* et al. [22], who observed the effect of consumption of different sea buckthorn fractions (equivalent to 100 g of fresh berries/day), showed that sea buckthorn supplementation for 33–35 days had a positive effect on the occurrence of metabolic diseases in overweight and obese women. *Larmo* et al. [21] also reported a significant effect of sea buckthorn fruit consumption on the metabolic profiles of overweight women. Recent studies have classified high systolic blood pressure (SBP) as the leading risk factors for death and disability worldwide, as well as hypertension being the largest risk factor for cardiovascular disease [44, 57]. The phytochemical and bioactive compounds of berries such as flavonoids, ellagitannins, and anthocyanins are potentially showing better effects in the risk reduction of various CVD such as reduce blood pressure, decreased endothelial dysfunction, which ultimately increases cardiovascular and brain health in an individual [50]. In our study, consumption of SBJ resulted in similar SBP and DBP values at the end of the study ($P>0.05$).

Kidney and liver markers

In this intervention study, volunteers consumed 50 mL of 100% bio commercial SBJ every day for 8-week period. In general, despite its bitter and sour nature, the juice was well accepted. SBJ consumption was well tolerated by participants and did not show changes ($P>0.05$) in liver and kidney function in serum following 8 weeks of SBJ intake (Table 3).

Lipid profile

The effect of 100% SBJ consumption on the lipid profile is shown in Table 4. Although there was no significant decrease in T-C in this intervention study, on the other hand there was a significant decrease in LDL-C ($P<0.05$) and a significant increase in HDL-C ($P<0.001$). The level of TG was similar at the end of the study ($P>0.05$). *Eccleston* et al. [11] found that SBJ with a flavonoid concentration of 1180 $mg.L^{-1}$ had protective properties against hypertension and coronary heart disease. The results of a study with 229 volunteers consuming 28 g of sea buckthorn berries for 3 months indicated increased blood concentrations of quercetin and isohamnetin, but consumption of sea buckthorn berries did not affect T-C, LDL-C, HDL-C and TG [21].

Inflammation markers

Increasing evidence demonstrates that inflammation plays an important role in the development of atherosclerosis [9, 34, 39]. Mainly, the crucial role of inflammation in atherosclerosis is particularly reflected by the overexpression of nuclear factor kappa B, C-reactive protein (CRP),

Table 3. Kidney and liver markers of study participants

Parameter	Baseline	Week 8	*p*-value
ALT ($\mu kat.L^{-1}$)	0.28 ± 0.10	0.32 ± 0.15	>0.05
AST ($\mu kat.L^{-1}$)	0.32 ± 0.06	0.34 ± 0.06	>0.05
GGT ($\mu kat.L^{-1}$)	0.36 ± 0.16	0.43 ± 0.27	>0.05
Urea ($mmol.L^{-1}$)	4.88 ± 1.45	4.98 ± 1.39	>0.05
Creatinine ($\mu mol.L^{-1}$)	64.75 ± 9.66	67.63 ± 9.63	>0.05
Uric acid ($\mu mol.L^{-1}$)	285.00 ± 63.04	302.67 ± 72.07	>0.05

Data are expressed as average ± standard deviation (SD), SBJ, sea buckthorn juice; ALT, alanine aminotransferase; AST, aspartate aminotransferase; GGT, gamma glutamyl transferase

Table 4. Lipid profile of study participants

Parameter	Baseline	Week 8	*p*-value
T-C ($mmol.L^{-1}$)	6.49 ± 1.07	6.45 ± 1.06	0.6894
HDL-C ($mmol.L^{-1}$)	1.70 ± 0.24	1.82 ± 0.27	<0.001
LDL-C ($mmol.L^{-1}$)	3.91 ± 1.32	3.38 ± 0.58	0.0257
TG ($mmol.L^{-1}$)	1.29 ± 0.70	1.27 ± 0.69	0.1857

Data are expressed as average ± standard deviation (SD); SBJ, sea buckthorn juice; T-C, total cholesterol; HDL-C, high density cholesterol; LDL-C, low density cholesterol; TG, triglycerides

interleukin-6 (IL-6), IL-18, tumor necrosis factor-alpha (TNF-α), and other inflammatory mediators [30]. The CRP is a nonspecific positive acute-phase protein that immediately rises after initiating an inflammatory state [8] and it could be an indicator of endothelial dysfunction that is also linked to a higher risk of CVD [58]. Our results (Table 5) show, that regular 8 week-long consumption of 50 mL of SBJ per day resulted significant reduction of CRP, IgA, IgG, IgM, orosomucoid ($P<0.001$) and similar values of IL-6 ($P>0.05$). In the study of *Larmo* et al. [21] the participants daily took 28 g of frozen sea buckthorn puree or placebo product similar in appearance, taste and smell to the active product and consumption of sea buckthorn berries reduced serum concentrations of CRP. Flavonoids and vitamin C are likely to be among the bioactive compounds responsible for the anti-inflammatory effect of fruit, possibly through synergetic mechanisms [25, 28, 36, 52].

Table 5. Inflammation markers of study participants

Parameter	Baseline	Week 8	*p*-value
CRP ($mg.L^{-1}$)	5.72 ± 2.39	4.75 ± 1.60	0.0038
IL-6 ($ng.L^{-1}$)	7.88 ± 0.93	7.32 ± 0.78	>0.05
ORM ($g.L^{-1}$)	0.89 ± 0.22	0.56 ± 0.17	<0.001
IgA ($g.L^{-1}$)	1.71 ± 0.68	1.57 ± 0.69	<0.001
IgG ($g.L^{-1}$)	10.52 ± 2.65	9.99 ± 2.66	<0.001
IgM ($g.L^{-1}$)	1.25 ± 0.63	1.15 ± 0.60	<0.001

Data are expressed as average ± standard deviation (SD);
SBJ, sea buckthorn juice; CRP, C-reactive protein; IL-6, interleukin-6;
ORM, orosomucoid; IgA, immunoglobulin A;
IgG, immunoglobulin G; IgM, immunoglobulin M

CONCLUSION

Sea buckthorn fruits are called fruits of the third generation. One of the most interesting properties is that sea buckthorn contains high concentrations of vitamin C, carotenoids, tocopherols and other bioactive compounds. The aim of this study was to evaluate the effect of regular consumption of 100% sea buckthorn juice on the risk factors of cardiovascular diseases in women of productive age with hypercholesterolemia. The obtained results support the hypothesis that the daily consumption of SBJ could contribute to the beneficial effects of reducing the risk of cardiovascular diseases, such as the reduction of body and visceral fat, LDL-C, inflammation markers and the increase of HDL-C. However, longer studies with a larger number of volunteers are needed to more thoroughly investigate the consumption of SBJ in the prevention and treatment of cardiovascular diseases.

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Conflict of interest

The authors declare no conflict of interest.

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