

The impact of short-term chokeberry juice intake on selected anthropometric and lipid indicators of women in productive age

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Black chokeberry is used in the prevention of cardiovascular diseases, obesity, hypertension and stress. It is valued for its high antioxidant capacity and, in addition, for its anti-inflammatory and hypolipidemic properties. The aim of this intervention study was to assess the impact of short-term intake of chokeberry juice on chosen anthropometric and lipid indicators of 30 women in productive age (from 45 to 55 years). Volunteers received 50 mL of 100% chokeberry juice daily for one-month period. Anthropometric and lipids parameters were evaluated before and after 4 weeks of intake of chokeberry juice. Analysis of body composition dynamics showed a significant decrease in body weight ($P < 0.01$), positive loss of body fat ($P < 0.05$), especially in visceral fat ($P < 0.01$). The waist to hip ratio was also improved ($P < 0.01$). The lipid profile also changed positively, the level of total cholesterol ($P < 0.05$) and low density lipoprotein decreased ($P < 0.01$). On the contrary, the level of high density lipoprotein increased in a positive direction ($P < 0.01$), while an improvement in the low density lipoprotein/high density lipoprotein ratio was also observed ($P < 0.001$). The average level of C-reactive protein decreased after consumption of chokeberry juice, but without statistical evidence ($P > 0.05$). We did not observe significant changes on kidney and liver markers. The results show that daily short-term intake of chokeberry juice can influence risk factors in the prevention of cardiovascular diseases and induce beneficial effects on health indicators of adult women.

Keywords: chokeberry juice, risk factors, body composition, lipid parameters

1 Introduction

Aronia melanocarpa (Michx.) Elliot is a fruit bush which pertains to the family Rosaceae and the apple subfamily Pomodieae. Chokeberry comes from North America (Platonova et al., 2021; Ekiert et al., 2021). In the United States, there are primary cultivars available for the fruit production: 'Viking' and 'Nero' cultivars (Brand, 2010). Aronia reached to Europe around 1900 through Germany to Russia (Kulling and Rawel, 2008). The significant commercial black chokeberry cultivars in Czech and Slovakia is 'Nero' (Jurikova et al., 2017). The gender name Aronia replaces the common name Aronia. Two species are recognized: *Aronia melanocarpa* (Michx.) Elliot (black chokeberry) and *Aronia arbutifolia* (L.) Elliot (red chokeberry) (Kulling and Rawel, 2008). It has gained a lot of attention due to its rich composition of biologically active substances (Markotić et al., 2017; Cvetanović et

al., 2018; Švarc-Gajić et al., 2019). Many components of aronia are dependent on a series of factors such as cultivar, fertilization, berry ripening, harvest date or habitat/place of harvest. The chemical composition of chokeberry berries, or its freshly squeezed juice, differs from other berries in the high content of carbohydrates, especially sorbitol, cellulose, pectins, anthocyanins (Kulling and Rawel, 2008; Švarc-Gajić et al., 2019; Brand et al., 2022) and the high content of some minerals – calcium, iron, molybdenum, manganese, copper, boron, iodine, cobalt; vitamins – ascorbic acid, riboflavin, pantothenic acid, pyridoxine (Litwińczuk, 2002), β -carotene and other carotenoids: β -cryptoxanthin and violaxanthin (Ekiert et al., 2021). Aronia has the highest antioxidant activity (AA) of all berry varieties, due to its high content of polyphenolic compounds, especially procyanidins and anthocyanins (Downar et al., 2011;

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Nowak et al., 2016; Jurikova et al., 2017; Liu et al., 2021). The content of total phenolic substances in chokeberry fruits ranges from 0.69 to 2.56 mg.g⁻¹ of fresh weight and from 3.44 to 7.85 mg.g⁻¹ of dry weight. The main polyphenols in chokeberry fruit include anthocyanins, proanthocyanidins, flavonoids, and phenolic acids (King and Bolling, 2020). The bioavailability of chokeberry anthocyanins has been analyzed in humans and it has been found that metabolites of anthocyanins produced by intestinal microbiota contribute to their medical benefits (Wangensteen et al., 2014). Almost 40% of the *in vitro* antioxidant activity of chokeberry is due to proanthocyanidins, which are hardly absorbed in the GIT. Their metabolites in the colon are of phenolic nature and exhibit antioxidant activity (Denev et al., 2019). The examine has shown that dietary anthocyanins are effective in obesity control (Jiang et al., 2021). In the berries of *Aronia melanocarpa*, the main four anthocyanins are important: cyanidin-3-O-galactoside (67.5%) and cyanidin-3-O-arabinoside (24.8%) as the main and cyanidin-3-O-glucoside (3.8%) and cyanidin-3-O-xyloside as minor (4.0%) (Goméz et al., 2021). Health benefits of chokeberry have been confirmed *in vitro* and *in vivo*, mainly in cardiovascular diseases (Parzonko et al., 2015). Chokeberry juice contains a considerable amount of vitamin B₃, which can alleviate cardiovascular disease by reducing lipid activity (Zhang et al., 2021). The anti-inflammatory and cardioprotective function of chokeberry has also been studied in patients with cardiovascular diseases (Loo et al., 2016; Banach et al., 2020). In a study by Zhang et al. (2021), chokeberry juice showed greater anti-inflammatory effects than rutin or rutin-magnesium complex. The anti-inflammatory activity of chokeberry has important in the prevention of CVD, diabetes and immune system diseases (Raczowska et al., 2022). The results of Parzonko et al. (2015) suggest that chokeberry extract can protect endothelial progenitor cells from angiotensin II induced dysfunction and have potential function in the prevention of coronary artery disease. The lipid-lowering effect of flavonoids has been demonstrated in many animal and human studies (Kuzmanova et al., 2007; Sidor et al., 2019). Chokeberry juice has been shown to have beneficial effects on total cholesterol and lipid levels (Kardum et al., 2015; Yamane et al., 2020).

The aim of this intervention study was to assess the impact of short-term intake of chokeberry juice on chosen anthropometric and lipid indicators of female in productive age.

2 Material and methods

The research was performed at the Institute of Nutrition and Genomics, Faculty of Agrobiology and Food Resources, SPU in Nitra, with the focus on monitoring the impact of daily consumption of 100% bio chokeberry juice (product of the company ZAMIO from Slovak Republic) on selected anthropometric and biochemical parameters of the probands. The study was approved by the Ethics Committee at the Specialized Hospital St. Zoerardus in Nitra, Slovakia (protocol no. 3/101921/2021).

2.1 Dietary intervention

The clinical trial included 30 women aged 45 and 55 (mean age of 49.23 ±5.44 years), who consumed 50 ml of chokeberry juice daily for 4 weeks as a part of their normal diet. Body composition and biochemical indicators were tested before and after 4 weeks of nutrition intervention. The monitored group of probands consisted of volunteers without health problems and normal biochemistry parameters in the blood. The precondition for the probands in the study was the consent of the participants of the examine and the planned examinations that they had to undergo during the research.

2.2 Measurements and analysis

We determined body composition with chosen parameters – body weight, body fat weight, visceral fat, skeletal muscle mass, fat-free mass using bioelectrical impedance analysis InBody 720 (Biospace Co. Ltd., Seoul, Korea). Biochemical analyses were quantified in an accredited laboratory of the Specialized Hospital St. Zoerardus in Nitra by analyzer BioMajesty JCA-BM6010/C. The level of total cholesterol, high density lipoprotein, triacylglycerols and liver enzymes were quantified by enzymatic method (kit DiaSys). The low density lipoprotein level was determined by the Friedewald equation. The chokeberry juice was made by cold pressing from organic black chokeberry without additives, stabilized only by pasteurization. The concentration of bioactive substances compounds and antioxidant activity with vitamin C were quantified of tested juice, shown in Table 1.

The total phenolic content (TPC) was determined by the spectrophotometric method (Lachman et al., 2006) according to the Folin-Ciocalteu method using spectrophotometer Shimadzu UV/VIS -1,800 and expressed in mg gallic acid equivalents (GAE).g⁻¹. Phenolic compounds were determined by HPLC Agilent 1260 Infinity II (Agilent Technologies GmbH, Waldbronn, Germany) by slightly modified method according by Gabriele et al. (2018). The antioxidant activity

Table 1 Concentration of phytochemicals in chokeberry juice

Parameter	Units	Quantity	Parameter	Units	Quantity
Ferulic acid	mg.L ⁻¹	100.37 ±0.45	Resveratrol	mg.L ⁻¹	4.82 ±0.98
Rutin	mg.L ⁻¹	238.69 ±0.25	Neochlorogenic acid	mg.L ⁻¹	372.36 ±1.89
Benzoic acid	mg.L ⁻¹	204.71 ±1.09	Chlorogenic acid	mg.L ⁻¹	497.86 ±0.52
Caffeic acid	mg.L ⁻¹	38.56 ±0.06	Quercetin	mg.L ⁻¹	7.93 ±0.02
Coumaric acid	mg.L ⁻¹	30.36 ±1.41	Vitamin C	mg.100 g ⁻¹	12.60 ±0.08
Total phenolic content	mg GAE.g ⁻¹	4.90 ±0.51	Antioxidant activity (inhibition of DPPH)	%	83.18 ±0.51

GAE – gallic acid equivalents

of chokeberry juice was detected using the stable radical 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay expressed as percentage of inhibition of the radical (Brand-Williams et al., 1995). Vitamin C content was determined by HPLC system Waters Separations Module 2695 with UV detector 2996 (Hernandez et al., 2006).

2.3 Statistical analysis

We used the Shapiro-Wilk test to determine whether the variables were normally distributed and the parametric variables were compared by the paired *t*-test using software Statistica Cz version 14. All data are expressed as the mean ±standard deviation (SD) and *P*-values of 0.05 or less were considered significant.

3 Results and discussion

The basic characteristics of the probands who participated in the regular intake of 100% chokeberry juice are shown in Table 2. It expresses the average values with standard deviations of somatometric, biochemical parameters and age of women. Dyslipoproteinemia, as one of the main risk factors for cardiovascular diseases, represents a group of metabolic diseases characterized by an increased level of plasma lipids and lipoproteins or

their inappropriate atherogenic composition (Jackuliak et al., 2013). An increased cholesterol level is considered from 5.2 mmol.l⁻¹ and values high above 7.8 mmol.l⁻¹ represent very high risk (NPZ, 2022). In the case of people with a high cardiovascular risk, it is necessary to achieve low density lipoprotein (LDL) levels below 2.5 mmol.l⁻¹. This monitored group of women shows an increased risk of CVD, we recorded 83% of women with total cholesterol higher than 5.2 mmol.l⁻¹ and 97% of them achieve LDL higher than 2.5 mmol.l⁻¹. The optimal value of triacylglycerols considered to be <1.7 mmol.l⁻¹ (Jackuliak et al., 2013) and 20% of women had high triacylglycerols levels >1.7 mmol.l⁻¹. In the group of women, the fasting blood glucose level was higher than the standard (up to 5.2 mmol.l⁻¹) of 27% and the average level was 5.02 ±0.71 mmol.l⁻¹.

In this interventional clinical study, women consumed 50 mL of 100% chokeberry juice daily for 4-week period. Daily consumption of chokeberry juice during four weeks was well accepted and did not cause any changes (*P*>0.05) in the liver and kidney functions of the volunteers (Table 3). The results of Kowalczyk et al. (2003) and Valcheva-Kuzmanova et al. (2007) indicated that anthocyanins in chokeberry have hepatoprotective effects and limits the accumulation of toxins in the kidney and liver. Aronia

Table 2 Basic characteristic of probands

Parameter	Mean ±SD	Min. – Max.	Optimal range
Age (year)	49.23 ±5.44	45–55	–
Body weight (kg)	74.12 ±11.80	53.10–97.80	–
Body mass index (kg.m ²)	27.13 ±3.98	19–36.4	18.5–24.9
Total cholesterol (mmol.l ⁻¹)	6.17 ±0.97	4.79–8.24	<5.19
HDL (mmol.l ⁻¹)	1.73 ±0.37	1.06–2.7	≥1.55
LDL (mmol.l ⁻¹)	3.90 ±0.86	2.29–5.77	<2.6
Triacylglycerols (mmol.l ⁻¹)	1.18 ±0.55	0.52–2.96	<1.7
Glucose (mmol.l ⁻¹)	5.02 ±0.71	4.10–6.80	4.0–5.5

Source: Cleeman et al., 2001

HDL – high density lipoprotein; LDL – low density lipoprotein; SD – standard deviation; Min. – minimum; Max. – maximum; optimal range of monitored parameters were compared with reference values of the NCEP ATP III

Table 3 The impact of short-term intake of chokeberry juice on kidney and liver parameters

Parameter	Baseline	Week 4	P-value
ALT ($\mu\text{kat.L}^{-1}$)	0.29 \pm 0.11	0.27 \pm 0.10	>0.05
AST ($\mu\text{kat.L}^{-1}$)	0.34 \pm 0.10	0.34 \pm 0.08	>0.05
GGT ($\mu\text{kat.L}^{-1}$)	0.56 \pm 1.21	0.55 \pm 1.18	>0.05
Urea (mmol.L^{-1})	4.98 \pm 1.63	5.22 \pm 1.72	>0.05
Creatinine ($\mu\text{mol.L}^{-1}$)	67.51 \pm 9.47	67.16 \pm 8.99	>0.05
Uric acid ($\mu\text{mol.L}^{-1}$)	299.23 \pm 69.31	297.79 \pm 66.37	>0.05

ALT – alanine aminotransferase; AST – aspartate aminotransferase; GGT – gamma glutamyl transferase

juice has beneficial effects on total cholesterol and lipid levels in patients with diabetes mellitus (Simeonov et al., 2002). Chokeberry juice has been shown to have beneficial effects on total cholesterol and lipid levels of patients with diabetes mellitus (Simeonov et al., 2002). Aronia extract also reduced body weight and glucose level in diabetes model KK-Ay mice (Yamane et al., 2016), improved hepatic lipid metabolism and indicators of oxidative stress of mice (Kim et al., 2013). Kardum et al. (2015) evaluated regular consumption of chokeberry juice in 23 subjects, and the results obtained indicate a beneficial effect on the lipid profile. Regular consumption of chokeberry juice has been shown to have a positive effect on the lipid profile (Kardum et al., 2015). Simeonov et al. (2002) and Skoczynska et al. (2007) in addition to the lipid profile, also noted an effect on blood pressure and glucose levels.

The same positive effect of dietary supplement of chokeberry juice on lipids were confirmed in reduction of triacylglycerols, total cholesterol and LDL in men with mild hypercholesterolemia (Skoczynska et al., 2007) as well as in metabolic syndrome in hypercholesterolemic patients (Broncel et al., 2010). In a study by Oprea et al. (2014) chokeberry juice had the ability to reduce fasting glucose level in patients with noninsulin dependent diabetes who received juice daily for three months. We evaluated the following markers of CVD risk, namely the lipid profile, CRP and glucose. Lowering effect was

shown on total cholesterol ($P < 0.05$) and especially on low density lipoprotein ($P < 0.01$). On the contrary, the level of high density lipoprotein increased in a positive direction ($P < 0.01$), while an improvement in the LDL/HDL ratio was also observed ($P < 0.001$). The average level of C-reactive protein decreased after consumption of chokeberry juice, but without statistical evidence ($P > 0.05$). Improvement of LDL/HDL ratio was also observed by Habanova et al. (2022), who studied the impact of apple/ berry juice intake. Effects of short-term regular intake of chokeberry juice on biochemical parameters are shown in Table 4. Duchnowicz et al. (2012) showed that extract of chokeberry lowered the high total cholesterol and lipid peroxidation in erythrocytes in patients with hypercholesterolemia and this extract also enhanced the rheological properties of erythrocytes. Anti-inflammatory and cardioprotective effects of chokeberry were studied in other research at patients with cardiovascular diseases (Ryszawa et al., 2006; Olas et al., 2008; Banach et al., 2020). Obesity is related to an increased risk of cardiovascular diseases occurrence, especially coronary heart diseases. Mechanisms associated with obesity include changes in body composition that affect the hemodynamics and structure of the heart. Atherosclerotic plaque formation is induced by cardiac dysfunction and pro-inflammatory cytokines produced by adipose tissue (Carbone et al., 2019).

Table 4 The impact of short-term intake of chokeberry juice on biochemical indicators

Parameter	Baseline	Week 4	P-value
Total cholesterol (mmol.L^{-1})	6.17 \pm 5.92	5.92 \pm 0.90	<0.05
HDL (mmol.L^{-1})	1.73 \pm 0.37	1.81 \pm 0.36	<0.01
LDL (mmol.L^{-1})	3.90 \pm 0.86	3.61 \pm 0.88	<0.01
Triacylglycerols (mmol.L^{-1})	1.18 \pm 0.55	1.16 \pm 0.51	>0.05
LDL/HDL ratio	2.34 \pm 0.65	2.09 \pm 0.65	<0.001
CRP (mg.L^{-1})	4.96 \pm 1.31	4.76 \pm 1.43	>0.05
Glucose (mmol.L^{-1})	5.02 \pm 0.71	4.96 \pm 0.76	>0.05

HDL – high density lipoprotein; LDL – low density lipoprotein; CRP – C-reactive protein; mean \pm SD – standard deviation

Table 5 The impact of short-term take of chokeberry juice on body composition

Parameter	Baseline	Week 4	P-value
Body weight (kg)	74.12 ±11.80	73.09 ±11.8	<0.01
Body fat mass (kg)	27.05 ±8.03	26.62 ±8.15	<0.05
BMI (kg.m ⁻²)	27.13 ±3.95	26.78 ±3.98	<0.05
Visceral fat (cm ²)	110.06 ±29.27	107.97 ±28.77	<0.01
Skeletal muscle mass (kg)	25.82 ±3.27	25.46 ±3.08	>0.05
Fat-free mass (kg)	47.07 ±5.55	46.47 ±5.24	>0.05
WHR	0.95 ±0.06	0.94 ±0.06	<0.01

WHR – waist to hip ratio; mean ±SD – standard deviation

Table 6 The impact of short-term intake of chokeberry juice on visceral fat of probands with overweight or obesity

BMI (kg.m ⁻²) probands	Baseline VF (cm ²)	Week 4 VF (cm ²)	P-value
BMI 25–29.9, overweight (<i>n</i> = 13)	119.01 ±16.83	115.69 ±16.27	<0.001
BMI 30–40, obesity I. and II. (<i>n</i> = 6)	143.91 ±19.80	142.04 ±20.60	>0.05
Total probands (<i>n</i> = 30)	110.06 ±29.27	107.97 ±28.77	<0.01

BMI – body mass index; VF – visceral fat; mean ±SD – standard deviation

Based on anthropometric measurements, we noted the following differences in the observed group of probands during the duration of the clinical study. During the consumption of chokeberry juice, we found out that the average BMI values of probands decreased ($P < 0.05$) in the evaluation of body fat mass, especially with a beneficial reduction of visceral fat ($P < 0.01$). A WHR index waist-to-hip ratio higher than 0.85 indicates a risk for cardiovascular diseases. During WHR index evaluation we found that in the beginning the average value in the probands was 0.95, which significantly decreased to 0.94 ($P < 0.01$) after the chokeberry juice consumption (Table 5). When evaluating the BMI of the research probands, according to WHO (2021), we recorded 36.7% of the probands in the optimal weight category (18.6–24.9 kg.m⁻²). 43.3% of the probands were overweight (25–29.9 kg.m⁻²). With a BMI of 30–34.9 kg.m⁻², it was in the obesity category I. degree 16.7% of the probands and 3.3% had obesity II. degree. Thus, 20% of the research subjects fall into the risk category for the development of CVD.

Jiao et al. (2021) confirmed hypotheses in the prevention of high-fat obesity, focusing on the potential of cyanidin-3-O-galactoside (C3G) from chokeberry. Supplementation with orlistat and C3G inhibited high-fat diet-induced weight gain, body fat gain, liver damage, lipid metabolism disorder, and proinflammatory factors in rats treated with chokeberry with C3G during an 8-week intervention experiment. Aronia C3G can be used in a therapeutic approach to modulate inflammation in the context of obesity. Yamane et al. (2020) found that daily consumption of chokeberry juice inhibited the adipocyte volume and reduced the weight in obese mice,

this signalize the unidentified anti-obesity inhibitors. Consumption of chokeberry juice also inhibits weight increase, lipid dysmetabolism and inflammation in obese mice (Bhaswant et al., 2017; Jeong et al., 2019).

Therefore, in the evaluation of anthropometric parameters, we mainly focused on the evaluation of changes in the risk visceral fat (VF) of the probands in connection with the consumption of chokeberry juice during the research. The average value of VF in the category of overweight probands decreased from the baseline of 119.01 cm² to 115.69 cm² ($P < 0.001$). In the category of probands with obesity I. and II. degree, we also noted a decrease in visceral fat without a significant statistical change. Statistically significant differences ($P < 0.01$) was confirmed in evaluation of visceral fat in the entire set of probands, shown in Table 6.

4 Conclusions

Black chokeberry is a highly biologically valuable plant, suitable for maintaining health and preventing civilization diseases. It is available on the market in various forms, but the best known is chokeberry juice however is currently not used enough in nutrition due to its bitter taste. In the research, we evaluated the impact of four-week intake of 100% chokeberry juice on chosen anthropometric indicators, focusing on changes in total and visceral fat and lipid indicators in women in productive age. In conclusion, we can confirm that regular intake of chokeberry juice can influence risk factors in the prevention of cardiovascular diseases and induce beneficial effects on health indicators of adult

women. Further larger studies with larger numbers of participants and longer duration of intervention are needed to further investigate the extent to which aronia juice may influence cardiovascular health risk factors.

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