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Comparative chemical composition of seeds of amaranth varieties introduced in Uzbekistan

Shuhratjon S. Olimjonov[1], Jamolitdin F. Ziyavitdinov[1], Soyibjon S. Bozorov[1,2], Uchqun J. Ishimov[1], Nodir Sh. Berdiev[1], Nadjiya N. Abrekova[1], Muydinjon M. Muminov[3] and Akmal M. Asrorov[1,4,✉]

[1]*Laboratory of Proteins and Peptides Chemistry, Institute of Bioorganic chemistry, Academy of Sciences of Uzbekistan, 83, M. Ulughbek Str., Tashkent 100125, Uzbekistan*

[2]*Laboratory of Physical and Chemical Methods of Research, Center for Advanced Technologies of Ministry of Innovative Development of Uzbekistan, 3A, Talabalar shaharchasi, Tashkent 100125, Uzbekistan*

[3]*Andijan Experimental Exhibition Area Functioning at the Disposal of the scientific and Practical Centre for Implementation of Innovative Projects, Ministry of Innovative Development of Uzbekistan, Bobur massif, Andijan Region, 170800, Uzbekistan*

[4]*State Key Laboratory of Drug Research, Shanghai Institute of Materia Medica, Chinese Academy of Sciences, 501 Haike Road, Shanghai 201203, China*

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Abstract

Amaranth is one of ancient cultured plants possessing valuable food quality that is still important in modern agriculture. In this work, we studied the seed chemical compositions of four plant varieties of *Amaranthus cruentus* and *Amaranthus hypochondriacus* species, acclimatized in Uzbekistan. Quantity of free amino acids, amino acids compositions of proteins, vitamins, oligo- and polysaccharides were established using chemical and physical methods. Our results on protein content suggest that introduction of these varieties into eco-climate of Andijan region, Uzbekistan, was not very favorable, though proteins resembled up to 9.4 – 13.4 % (w/w) of seed biomass. Of the water-soluble vitamins tested, the vitamin B1 was most abundant (0.81 – 1.14 $\mu g.g^{-1}$ of seed) with significant differences among the acclimatized varieties. Impact of feeding rats with seeds flour of studied Helios variety was established. Blood levels of triglycerides, high-, low- and very low-density lipoproteins, catalase activity under hyperlipidemia were analyzed. We found Helios variety seed flour, possibly because of high saccharides content, significantly lowered the levels of total cholesterol (-26 %) and low-density lipoproteins (-21 %). The acclimatized varieties were identified as potentially valuable food source possessing antihyperlipidemic property.

Introduction

The increasing number of world population necessitates to use plant resources, having nutritive quality, more and efficiently. In this regard, amaranth plant seed is a suitable raw material providing high nutrition value. The plant genus belonging to *Amaranthacea* family contains around 70 species (Berghofer and Schoenlechner 2002). The species *Amaranthus hypochondriacus*, *Amaranthus cruentus* and *Amaranthus caudatus* are cultivated for their seeds in Latin American countries (Bressani 2003). It is grown in India for both leaves and seeds (Prakash and Pal 1991).

✉ *Corresponding author:* akmal84a@gmail.com

Slovakia, Hungary and Italy are the European producers of amaranth. Approximately 100 000 ha in Russia is expected as amaranth growing area (Moudry *et al.* 1999). Four new varieties belonging to *A. hypochondriacus* (Kharkov and Lera) and *A. cruentus* (Andijan and Helios) species have been recently acclimatized in Uzbekistan (Bozorov *et al.* 2018).

Amaranth seed has been widely investigated. Updated reviews on its chemical compositions have shown its high nutritive quality: oils, fatty acids, proteins and peptides, free amino acids, squalene, tocopherols and tocotrienols, sterols and other lipophilic compounds, carbohydrates, dietary fiber and other constituents (Singh and Singh 2011; Venskutonis and Kraujalis 2013). These papers report highly differing amounts of compounds in various amaranth varieties. Therefore, in terms of the same growing conditions, studying comparative chemical compositions of different amaranth varieties was considered significant in terms of application in bakery processes. Acclimatization of amaranth started in Uzbekistan a decade ago.

In previous work, we demonstrated oil composition of four amaranth varieties, introduced in the country (Bozorov *et al.* 2018). In this work, we compared the quantity of water-soluble, acidic and basic polysaccharides, free and bound amino acids, some major monosaccharides as well as water-soluble vitamins. Besides, we studied the effects of amaranth seed flour on lipid profile of rats.

Experimental

Chemicals and reagents

All the used chemicals and reagents were of analytical grade. For HPLC analysis, chemical from Sigma were used.

Total Nitrogen

Total nitrogen in 1 g defatted seed flour was determined by modified Kjeldahl method. Ammonium salts formed in boiling sulfuric acid and further ammonium was quantified titrimetrically.

Extraction and quantification of polysaccharides

The flour samples were ground in a laboratory grinder to a size of 2 to 6 mm. To remove low molecular weight impurities and proteins, 100 grams of raw material was extracted in a Soxhlet apparatus with 95 % ethyl chloroform-alcohol mixture.

To isolate water-soluble polysaccharides, fat-freed flour samples were extracted with boiling water under reflux condenser, for two hours, twice. Obtained extracts were filtered and concentrated.

We, further, extracted pectin fractions with 1.1 % hydrochloric acid (1:10, w/v) using reflux condenser in water bath at 95 °C, for 2 hours. The extracts were filtered, concentrated and further was dialyzed against distilled water overnight, and centrifuged (10 min; 6,000 rpm). The supernatant was used for quantification.

We carried alkaline extraction using 5 % NaOH solution (1:10, w/v) at 60 °C in oil bath, for 1 hour. The extracts were filtered, centrifuged (10 min; 6,000 rpm) and neutralized with acetic acid. Crude extracts were concentrated at 50 °C and dialyzed against distilled water overnight. Freeze-dried extracts were quantified.

The concentrations of polysaccharides in obtained fractions were determined by phenol-vitriolic method (Saha and Brewer 1994).

Protein quantification

Quantity of soluble proteins was determined by Lowry method (Lowry *et al.* 1951).

Extraction of Amino Acids and Their Modification

One gram seed samples were extracted in 3 mL water for 4 h stirring. The supernatant was isolated after centrifugation for 10 min (3,000 rpm). Higher molecular compounds were precipitated adding 10 % trichloroacetic acid (1:1, v/v) for 15 min (8,000 rpm). 200 µL aliquot was taken and lyophilized. Dry extract was dissolved in 200 µL water acetonitrile-triethylamine (1:7:1) then the extract was dried. The process was twice repeated. In order to obtain phenylthiocarbamoyl (FTC) derivatives purified samples and standard amino acid were modified with water-ethanol-

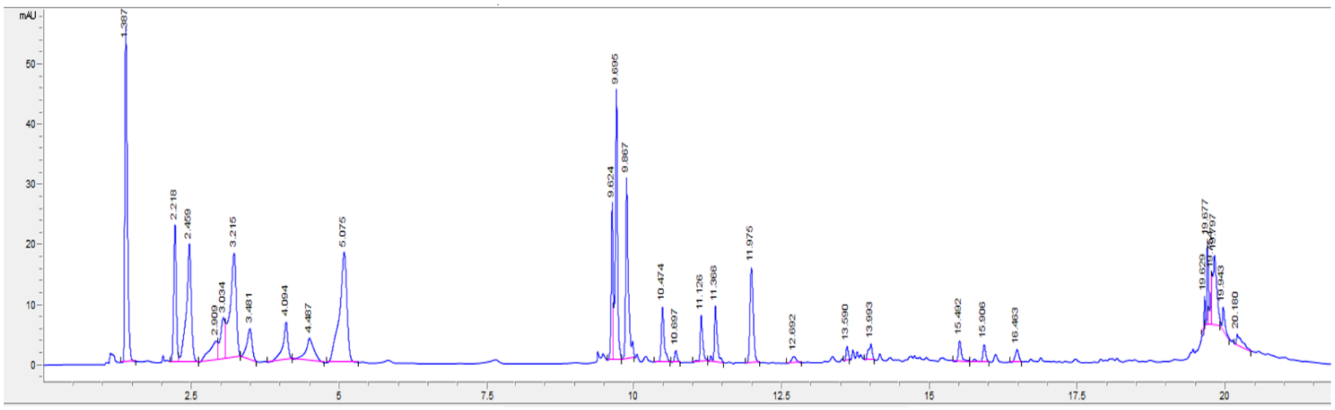

Fig. 1. HPLC chromatogram for vitamins of seeds of Helios variety at 290 nm.

triethylamine-phenylisothiocyanate (1:7:1:1) for 30 min (Cohen and Strydom 1988).

Preparation of total amino acid samples

Ground seed samples (1 g each) were hydrolyzed in 1 mL HCl (5.7 M) and concentrated trifluoroacetic acid mixture (2:1). The ampules were sealed in vacuum. The reaction was carried at 166 °C for 70 min. Further the hydrolysates were freeze-dried. Further FTC derivatives were obtained as described above.

HPLC analysis of amino acids

Quantity analyses of FTC derivatives were conducted in Agilent technologies 1200 (Column: Supelco Discovery HS C18, Cat 56925/-U 7.5 cm / 4.6 mm, 3 µm), with buffer: B – 0.14 M CH3COONa + 0.05 % tetraethyl ammonium, pH 6.4, A–MeCN, gradient % B/min, 0 – 6 %, 5 min, 6 – 30 %, 30 min, 30 – 60 % 5 min, 60 – 100 % 5 min. Amino acid derivatives were detected with a flow rate 1.2 mL.min^{-1} at 269 nm. RSD values for the mean values of amino acids' quantity, were used for statistical evaluation. A representative chromatogram from the extract of Helios seeds is depicted in Fig. 1, indicating the adequacy of the method.

Analysis of vitamins

Vitamins were extracted with ten-fold 40 % ethanol. The process was carried boiling, using reflux condenser for an hour. Further, the process lasted for 2 h at room temperature stirring. The extract was taken out. Seed samples were extracted with five-fold volume of 40 % ethanol twice more. The sum of extracts were centrifuged (7,000 rpm) and used for HPLC analysis.

The vitamins were identified and quantified by HPLC (Agilent-1200, reverse phase Eclipse XDB C18 column, 5µm, 4.6 x 150mm) at 254 and 290 nm, diode-array detector. The flow rate made 1 mL.min^{-1}, A: 0.5 % acetic acid (pH 1.7); B: acetonitrile; the gradient of acetate buffer and acetonitrile (6 – 8 min 90:10, 9 – 15 min 80: 20, 15 – 17 min 96:4) at 25 °C, 5 µL of aliquot was used for analysis. We identified and quantified vitamins by calibration curves that were created by using standards of vitamins.

Animal experiments

Animal experiments were carried in compliance with requirements of Pharmaceutical Agency of Uzbekistan developed in accordance with standards of research involving animals. We carried animal experiments on 50 white outbred male rats with a body weight 250 – 300 g in the vivarium of the Institute of Bioorganic chemistry. Animals were kept in the standard condition with a balanced diet. On the sixth day of the treatment the blood samples of the animals were used for the quantification of total cholesterol (TC), triglycerides, low-density lipoproteins LDL, high density lipoproteins (HDL), very low-density lipoproteins (VLDL) and catalase activity. These parameters were calculated by Friedewald equation according to state standards. The atherogenic coefficient was calculated using the following formula: AC = (total cholesterol – HDL) / HDL.

Table 1. Total and remaining oiliness of see of amaranth varieties (% in relation to total mass).

Species	Amaranth varieties	Seed Yield [t.ha^{-1}]	Total oiliness	Cold compressed	Remaining oiliness after cold compression
A. hypochondriacus	Kharkov	3 – 5 t	11.86±0.32	7.81±0.17	4.05±0.15
	Lera	1.5 – 2 t	11.20±0.40	7.55±0.23	3.65±0.17
A. cruentus	Andijan	1 – 1.5 t	10.07±0.14	6.39±0.06	3.68±0.08
	Helios	2 – 2.5 t	11.87±0.34	7.68±0.16	4.19±0.18

Results and Discussion

Obtained results show that oiliness of these amaranth seed varieties range from 10.07 to 11.87 % (v/w). During cold compression almost two third of the oil was found extracted (65.9, 67.4, 63.4 and 64.7 % respectively, v/w). The highest oiliness values were determined in Kharkov and Helios varieties. Remaining oiliness was established by extracting with petrol (Table 1).

Total nitrogen in amaranth seeds was established between 15.7 – 17.5 % (w/w). There is a lack of significant differences in total nitrogen in Kharkov, Lera and Helios seeds. Andijan variety sample has the lowest total nitrogen, equaled 15.7 % (w/w) in relation to defatted seeds. Soluble proteins' quantity found reversely correlated with total nitrogen; the highest quantity belonged to Andijan variety that was equal to 5.1 % (w/w). Kharkov and Lera seeds contained 47.6 – 47.8 mg soluble proteins per one g defatted seeds (Table 2).

We found glucose as the major monosaccharide among the studied samples with inconsiderable differences. Around 5 mg glucose was calculated per 100 mg amaranth seed flour samples. With no significant differences, fructose and mannose were determined as minor monosaccharides in all four samples. Water-soluble polysaccharides made 22 – 24 % (w/w) of the seed biomass. Identical mass ratios of acidic and alkali-soluble polysaccharides (1:3) were observed in Lera, Andijan and Helios varieties. Unlikely, Kharkov seed flour contained higher level of acidic carbohydrates. Thus, we established identical content of total carbohydrates of these four seed samples (Table 2).

Our results indicate that, more than two third of the free amino acids in the seed composition belong to nonessential ones. Aspartic acid, glutamic acid and glutamine were determined to be major amino acids among all. Nonessential free amino acids in *Amaranthus hypochondriacus* varieties were found significantly higher than the varieties of *Amaranthus cruentus* species (Table 3).

We found, that histidine was the most widespread essential amino acid in the seeds of these studied amaranth varieties that its quantity is at least twice higher than valine, isoleucine, leucine and tryptophan prevailing remaining essential amino acids. Differing from nonessential amino acids, species-dependent distinctions were not found among these varieties. However, Andijan variety of *A. cruentus* contained remarkably lower quantities of valine, methionine, tryptophan

Table 2. Weight fractions (%) of saccharides and proteins in amaranth seeds, acclimatized in Uzbekistan.

	A. hypochondriacus		***A. cruentus***	
	Kharkov	Lera	Andijan	Helios
Glucose	5.24±0.08	5.03±0.06	4.98±0.05	5.22±0.07
Fructose	0.41±0.02	0.39±0.03	0.38±0.02	0.40±0.01
Sucrose	0.23±0.01	0.21±0.01	0.22±0.01	0.23±0.01
Sum	5.88	5.63	5.58	5.85
Soluble polysaccharides	23.90±0.98	23.70±1.04	22.10±0.99	23.50±1.01
Acidic polysaccharides	19.60±0.88	8.40±0.34	8.20±0.34	8.40±0.34
Alkali-soluble polysaccharides	11.70±0.55	22.10±0.99	24.80±1.16	25.30±1.11
Sum	55.2	54.20	55.10	57.20
Total saccharides	61.08	59.83	60.68	63.05
Total nitrogen	17.30±0.5	17.50±0.3	15.70±0.70	17.10±0.60
Soluble proteins	4.76±0.02	4.78±0.05	5.10±0.02	5.00±0.01

Table 3. Contents of free amino acids in the seeds of amaranth varieties (mg.g^{-1} dry mass, RSD ≤ 3 %).

No.	Amino acids	*A. hypochondriacus*		*A. cruentus*	
		Kharkov	**Lera**	**Andijan**	**Helios**
Nonessential amino acids					
1	Aspartic acid	**0.332**	**0.487**	**0.326**	**0.312**
2	Glutamic acid	**0.617**	**0.611**	**0.203**	**0.498**
3	Serine	0.132	0.067	0.045	0.170
4	Glycine	0.061	0.083	0.060	0.073
5	Asparagine	0.062	0.083	0.059	0.074
6	Glutamine	**0.307**	**0.414**	**0.095**	**0.213**
7	Proline	0.040	0.044	0.042	0.041
8	Tyrosine	0.084	0.101	0.051	0.079
9	Alanine	0.160	0.104	0.089	0.167
	Sum	***1.795***	***1.994***	***0.970***	***1.627***
Essential amino acids					
10	Threonine	0.048	0.036	0.033	0.067
11	Arginine	0.057	0.068	0.066	0.054
12	Valine	**0.078**	**0.088**	**0.056**	**0.081**
13	Methionine	0.040	0.047	0.024	0.040
14	Isoleucine	**0.090**	**0.093**	**0.084**	**0.088**
15	Leucine	**0.088**	**0.090**	**0.089**	**0.090**
16	Histidine	**0.177**	**0.186**	**0.167**	**0.169**
17	Tryptophan	**0.071**	**0.082**	**0.053**	**0.076**
18	Phenylalanine	0.039	0.053	0.042	0.046
19	Lysine	0.060	0.087	0.041	0.068
	Sum	***0.748***	***0.830***	***0.655***	***0.779***
	Total	**2.543**	**2.824**	**1.625**	**2.406**

Amino acids, the concentration of which are significantly higher than others, are given in bold.

and lysine (Table 3).

The sum amounts of free amino acids were determined to make 0.16 – 0.28 % (w/w) of the seed. Total amino acids concentrations were calculated as around 11.4 ± 2 % (w/w) of the seed flour (Table 4). These results make enable to conclude that the seeds of these introduced amaranth varieties is a rich protein source but not of free amino acids.

Total amino acids contents studied in our work revealed that glutamic acid/glutamine sum were major nonessential amino acids. During the hydrolysis, glutamine and asparagine convert their acidic forms. Thus, the sum of glutamic acid and glutamine, aspartic acid and asparagine are given together. At least twice higher concentrations of glutamine and glutamic acid were determined compared to other nonessential amino acids. Among other nonessential amino acids, no notable differences were found, except for proline (Table 4).

Phenylalanine was found the most widespread amino acid in the amaranth seeds. We determined thrice higher its quantity than other prevailing amino acids in Kharkov, Lera and Andijan varieties. The lowest total amino acids belonged to Helios variety. Notably lower quantity of alanine, proline, arginine, methionine and phenylalanine were defined in this sample compared to other varieties (Table 4).

These results indicate that, studied Amaranth seeds are a rich source of proteins with 9.4 – 13.4 % (w/w) related to seed mass. Our results demonstrate similar amino acid ratios in these four varieties. However, highly different molar ratios of bound amino acids were reviewed in amaranth seeds (Venskutonis and Kraujalis 2013). Thus, in terms of nutritious quality of seeds, it is possible to expect various molar ratios in different species and varieties. For instance, arginine and lysine were of the highest quality in *A. cruentus* (Escudero *et al.* 2004) and *A. hypochondriacus* (Dodok *et al.* 1997), whereas they were detected as low-content amino acids compared to others in another *A. hypochondriacus* variety (Nimbalkar *et al.* 2012). The threonine molar content was

Table 4. Total amino acid contents in the seeds of amaranth varieties (mg.g^{-1} dry mass, RSD ≤ 3 %).

No.	Amino acids	*A. hypochondriacus*		*A. cruentus*	
		Kharkov	**Lera**	**Andijan**	**Helios**
Nonessential amino acids					
1	Aspartic acid/asparagine	10.24	10.09	7.96	7.21
2	Glutamic acid/glutamine	**27.16**	**26.26**	**18.04**	**16.15**
3	Serine	4.80	4.49	3.53	3.68
4	Glycine	5.62	5.44	4.40	4.00
5	Tyrosine	5.53	4.43	6.75	5.96
6	Alanine	4.97	5.11	1.84	3.58
7	Proline	3.98	3.98	3.43	2.97
	Sum	**62.30**	**59.80**	**45.95**	**43.55**
Essential amino acids					
8.	Threonine	5.53	4.43	6.75	5.96
9.	Arginine	6.82	7.63	4.48	4.62
10.	Valine	5.09	4.87	6.08	5.57
11.	Methionine	3.11	2.22	1.98	1.33
12.	Isoleucine	**7.23**	**6.49**	**6.28**	**5.51**
13.	Leucine	**7.79**	**7.02**	**6.93**	**6.26**
14.	Histidine	5.20	5.20	6.00	7.35
15.	Phenylalanine	**23.69**	**19.11**	**18.41**	**8.17**
16.	Lysine	**7.15**	**6.26**	**6.03**	**5.70**
	Sum	**71.61**	**63.23**	**62.94**	**50.47**
	Total	**133.91**	**123.03**	**108.89**	**94.02**

Amino acids, the concentration of which are remarkably higher than others, are given in bold.

reverse in these three amaranth varieties.

Our results revealed that B1 and B2 vitamins are the main vital components in the seed of all varieties with almost no statistical differences except for vitamin B1 in Lera variety. Vitamin B1 was determined in the range of 0.81 – 1.14 μg.g^{-1}. 0.33 – 0.38 μg vitamin B2 was observed in 1 g of amaranth seeds. B5, B6, B9 and PP vitamins were demonstrated as minor vitamin components in the seeds with statistical differences with the exception for B9. We found that among the studied components, vitamin PP was the most minor one in Kharkov and Lera varieties belonging to *A. hypochondriacus*, and Andijan and Helios varieties of *A. cruentus* contained the smallest amount B5 (Fig. 2). Data on vitamin contents in amaranth do not correspond with the obtained results in this work. Several times higher contents of vitamin B2, B5 and B6 were reported in amaranth seeds. Similarity belonged to B1 only (Pavlík 2012). Vitamin B2 and vitamin B6 were determined as the main vitamins raw amaranth seeds, that equaled 1.47 and 4.54 mg.g^{-1}, respectively (Murakami *et al.* 2014). Vitamin B1 (thiamine), B2 (riboflavin) and B6 (pyridoxine) were found in similar quantities in amaranth seeds grown in Kenya, that ranged 5 – 6 mg.g^{-1} (Mburu *et al.* 2012). Mihhalevski *et al.* (2013) demonstrated contents of vitamins in rye and wheat seeds and flours. Vitamin B1 content in red rye malt variety was similar to values we measured in our samples. Its content in other rye flours was defined a few-fold times higher (differing from 1.85 to 3.76 μg.g^{-1}). The contents of B5, B6 and PP vitamins were several-fold higher in rye varieties than our findings. Thus, we established that amaranth flour concedes rye and wheat flour by its vitamins contents. Several-fold differences of our vitamin quantity data from these results could be linked to acclimatization in hot climate that may not be favorable for the plant. Further research will require to compare annual results.

A. hypochondriacus is considered to be the hybrid of a wild type *A. powelli* and progenitor species – *A. cruentus* (Brenner *et al.* 2010). Possibly, due to this relationship, previous research shows no remarkable differences in protein and/or carbohydrate contents (Table 5). However, in acclimatized varieties, we observed differences in protein percentage. Besides, Obtained results in this work revealed significantly lower percentage of proteins in the seeds compared to other varieties

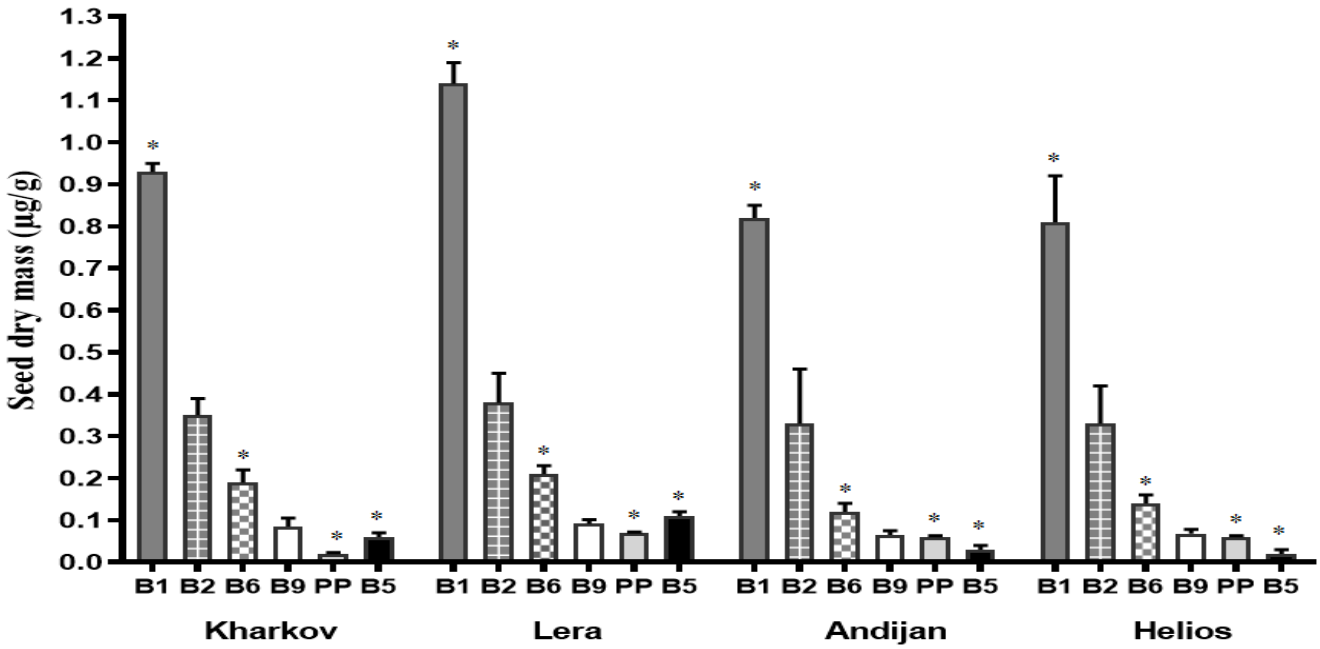

Fig. 2. Water-soluble vitamins contents in acclimatized amaranth varieties. Data indicate average value of 3 replicates ± RSD. Asterisks indicate significant difference among varieties at $P < 0.05$.

in both species (Table 5). Possibly, it could be resulted by drier season in Uzbekistan. For instance, the Lera variety created by individual selection from *A. hypochondriacus* was reported to contain 20.6 % in the seed (Goptsiy *et al.* 2008). It could be concluded that drier and hotter climate would not be a favorable condition for amaranth in terms of protein.

Total carbohydrates of these four varieties were determined to be average compared with available data: 60 – 63 % (w/w) of the seed mass belonged to saccharides (Table 5). The seed yield in Kharkov and Helios make 2.0 – 2.5 and 3 – 5 tons per hectare, respectively. Therefore, these varieties have been grown for the seed. Andijan and Lera varieties seed yield make 1 – 1.5 and 1.5 – 2.0 ton per hectare. They were chosen for their above-ground biomass (Table 1).

We further studied the pharmacologic effects of amaranth seed flour (Helios) on lipids level in experimental rats (Table 6). Obtained results in twin model was accompanied with significantly

Table 5. Comparison of proteins and carbohydrates content in *A. cruentus* and *A. hypochondriacus* seeds.

Variety	Protein % (w/w)	Carbohydrate % (w/w)	Reference
A. cruentus	16.21	58.00	Capriles 2008
A. cruentus (82s-1011)	14.70		Bressani *et al.* 1987
A. cruentus (823434)	15.07		Bressani *et al.* 1987
A. cruentus (82s-1034)	16.00		Bressani *et al.* 1987
A. cruentus	15.68	55.41	Babor *et al.* 1994
A. cruentus	16.89	66.09	Uriyapongson and Rayas-Duarte 1994
A. hypochondriacus hyb.	14.02	68.23	Uriyapongson and Rayas-Duarte 1994
A. hypochondriacus	14.23		Tömösközi et al 2009
A. hypochondriacus (A-718)	13.70		Bressani *et al.* 1987
A. hypochondriacus (A-720)	14.75		Bressani *et al.* 1987
A. hypochondriacus (82s-1023)	15.32		Bressani *et al.* 1987
A. hypochondriacus (82s-1024)	14.75		Bressani *et al.* 1987
A. cruentus, Andijan	**10.90**	**60.68**	This work
A. cruentus, Helios	**9.40**	**63.05**	This work
A. hypochondriacus, Kharkov	**13.40**	**61.08**	This work
A. hypochondriacus, Lera	**12.30**	**59.83**	This work

Table 6. Effects of Helios seed flour on lipid profile of rats carried in twin model (n = 10).

Groups	Total cholesterol [mg.L^{-1}]	Triglycerides [mg.L^{-1}]	High density lipoproteins [mg.L^{-1}]	Low density lipoproteins [mg.L^{-1}]	Very low density lipoproteins [mg.L^{-1}]	Catalase activity [mCat.L^{-1}]
Intact animals	5.20±0.03	6.00±0.06	3.10±0.05	1.20±0.04	1.10±0.03	0.079±0.002
Control	8.75±0.03	10.60±0.04	1.60±0.05	3.15±0.03	2.12±0.05	0.447±0.030
(Hyperlipidemia + H_2O)	+68 %	+77 %	-48 %	+74 %	+91 %	>5,6-times
Hyperlipidemia + amaranth	6.50±0.03	9.20±0.06	2.00±0.05	2.49±0.04	1.84±0.03	0.24±0.01
flour (Helios), 200 mg.kg^{-1}	-26 %*	-13 %*	+25 *	-21 *	-13 %*	<1.9-times

* stands for significant differences of the indicated parameters compared to control samples.

decreased levels of total cholesterol (-26 %), triglycerides (-13 %), low density (-21 %) and very low density lipoproteins (-13 %) in comparison with hyperlipidemia model. Almost twice lower catalase activity was determined in animal fed with amaranth flour compared with control. These deviations in the lipoproteins quantity indicate the positive effect of the flour. A directly proportional solid link between elevated cholesterol/LDL and coronary heart disease (Nelson 2013) should serve as basis to conclude that the amaranth seed flour can be used as antihyperlipidemic nutrition (Table 6).

HDL, increased by 25 %, can also be estimated as positive point. More recent literature review, based on genetic analysis and clinical research, conclude that higher HDL are always beneficial for coronary heart diseases, while their lower level is always detrimental (Kosmas *et al.* 2018).

Conclusions

In this work we compared the chemical compositions of the seed flour of four amaranth varieties belonging to *A. hypochondriacus* and *A. cruentus* series, acclimatized in Uzbekistan. It has been established that, total amino acids make 9.4 – 13.4 % (w/w) of the seed flour, more than of half of which belong to essential ones. Phenylalanine, glutamic acid and glutamine were determined as major bound amino acids. Dry and hot climate of Uzbekistan caused total protein content to be significantly lower, compared to progenitors. Contents of water-soluble vitamins in the seeds of amaranth varieties have been established. We found vitamin B1 and B3 as major compositions of this class of compounds in all of these varieties. Total carbohydrates quantities were determined not to significantly differ. The detailed study of the chemical composition amaranth seeds, determining nutrition value, revealed that it can be used as rich carbohydrate source. We demonstrated important biological value of amaranth flour with antihyperlipidemic property.

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Conflict of Interest

The authors declare that they have no conflict of interest.

References

Babor K, Halásová G, Dodok L, Géciová R, Lokaj J (1994) Characterization of starch from *Amaranthus cruentus* L. Chem Pap. 48: 58-63.

Berghofer E, Schoenlechner R (2002) Grain amaranth. *In* Belton PS, Taylo JRN (Eds.), Pseudocereals and less common cereals. Grain properties and utilization potential, Springer-Verlag, Berlin, Heidelberg, pp. 219-253.

Bozorov SS, Berdiev NSh, Ishimov UJ, Olimjonov ShS Ziyavitdinov JF, Asrorov AM, Salikhov ShI (2018) Chemical composition and biological activity of seed oil of amaranth varieties. Nova Biotechnol. Chim. 17: 66-73.

Brenner DM, Baltensperger DD, Kulakow PA, Lehmann JW, Myers RL, Slabbert MM (2010). Genetic resources and breeding of amaranthus. *In* Janick J (Ed.), Plant

breeding reviews, Wiley-Blackwell, New Jersey, USA, pp. 227-285.

Bressani R, Gonzhles JM, Zuiiga J, Breuner M, Elias LG (1987) Yield, selected chemical composition and nutritive value of 14 selections of amaranth grain representing four species. J. Sci. Food Agric. 38: 347-356.

Bressani R (2003) Amaranth. *In* Caballero B. (Ed.), Encyclopedia of food sciences and nutrition, Elsevier/Academic Press, Oxford, pp. 66-173.

Capriles VD, Coelho KD, Guerra-Matias AC, Areas JAG (2008) Effects of processing methods on amaranth starch digestibility and predicted glycemic index. J. Food Sci. 73, H160-4.

Cohen SA, Strydom DJ (1988) Amino acid analysis utilizing phenylisothiocyanate derivatives. Anal. Biochem. 174: 1-16.

Dodok L, Modhir AA, Buchtová V, Halásová G, Poláček I (1997) Importance and utilization of amaranth in the food industry. Part 2. Composition of amino acids and fatty acids. Nahrung 41: 108-110.

Escudero NL, De Arellano ML, Luco JM, Gimenez MS, Mucciarelli SI (2004) Comparison of the chemical composition and nutritional value of *Amaranthus cruentus* flour and its protein concentrate. Plant Food Hum. Nutr. 59: 15-21.

Goptsiy T. Voroncov N, Popov V, Zhyravel D, Gromenko S (2008). Grain varieties of amaranth developed by selection at Kharkiv National Agrarian University and the perspectives of their use. *In* "Amaranth – Plant for the Future" 5th International Symposium of the European Amaranth Association Nitra, Slovak Republic. 2008, pp: 97-100

Kosmas CE, Martinez I, Sourlas A, Bouza KV, Campos FN, Torres V, Montan PD, Guzman E (2018) High-density lipoprotein (HDL) functionality and its relevance to atherosclerotic cardiovascular disease. Drugs Context. 7: 212525.

Lowry OH, Rosenbrough NJ, Farr AL, Randall RJ (1951) Protein measurement with the Folin phenol reagent. J. Biol. Chem. 193: 265-275.

Mihhalevski A, Nisametdinov I, Hälvin K, Oseka A, Paalme T (2013) Stability of B-complex vitamins and dietary fiber during rye sourdough bread production. J. Cereal Sci. 57: 30-38.

Moudry J, Pejcha J, Peterka J (1999). The effect of genotype and farming technology on the yield of amaranth *Amaranthus* sp. *In* Collection of Scientific Papers, Faculty of Agriculture in České Budějovice. Series for Crop Sciences. 16: 93-98.

Murakami T, Yutani A, Yamano T, Iyota H, Konishi Y (2014) Effects of popping on nutrient contents of amaranth seed. Plant Foods Hum. Nutr. 69: 25-29.

Mburu MW, Gikonyo NK, Kenji GM, Mwasaru AM (2012) nutritional and functional properties of a complementary food based on Kenyan amaranth grain (*Amaranthus cruentus*). Afr. J. Food Agric. Nutr. Dev. 12: 5958-5977.

Nelson RH (2013) Hyperlipidemia as a risk factor for cardiovascular disease. Prim. Care. 40: 195-211.

Nimbalkar MS, Pai SR, Pawar NV, Oulkar D, Dixit GB (2012) Free amino acid profiling in grain Amaranth using LC-MS/MS. Food Chem. 134: 2565-2569.

Pavlík V (2012) The revival of Amaranth as a third-millennium food. Neuro. Endocrinol. Lett. 33: 3-7.

Prakash D, Pal M (1991) Nutritional and antinutritional composition of vegetable and grain amaranth leaves. J. Sci. Food Agric. 57: 573-583.

Saha SK, Brewer CF (1994) Determination of the concentrations of oligosaccharides, complex type carbohydrates, and glycoproteins using the phenol-sulfuric acid method. Carbohydr. Res. 254: 157-167.

Singh N, Singh P (2011). Amaranth: Potential source for flour enrichment. *In* Preedy VR, Watson RR, Patel VB. (Eds.), Flours and breads and their fortification in health and disease prevention, Academic Press, Elsevier, Oxford, UK, pp: 101-111.

Tömösközi S, Baracskai I, Schoenlechner R, Berghofer E, Lasztity R (2009). Comparative study of composition and technological quality of amaranth: I. Gross chemical composition, amino acid and mineral content. Acta Aliment. 38: 341-347.

Uriyapongson J, Rayas-Duarte P (1994) Comparison of yield and properties of Amaranth starches using wet and dry-wet milling processes'. Cereal Chem. 71: 571-577.

Venskutonis PR, Kraujalis P (2013). Nutritional components of amaranth seeds and vegetables: A review on composition, properties, and uses. Compr. Rev. Food Sci. Food Saf. 12: 381-412.