

## Comparative study on chemical and lipid composition of two varieties of quinoa seeds (*Chenopodium quinoa* L.)

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The seeds of two varieties of quinoa (*Chenopodium quinoa* L.), white and black, were analysed for their chemical composition and a detailed study of their lipids was carried out as well. The chemical composition of the seeds was as follows: protein content - 15.7% and 14.5%, lipids - 5.9% and 6.6%, carbohydrates - 65.6% and 64.6%, starch - 40.2% for both varieties, water soluble sugars - 7.2% and 5.5%, and minerals - 2.4% and 2.3%, respectively. The oil content was about 6.0%, but high amounts of biologically active compounds in the seed oils of black quinoa were noted (tocopherols - 1102 mg/kg, phospholipids - 7.6% and sterols - 3.4%), while in the seed oils of white quinoa tocopherols were 365 mg/kg, phospholipids - 11.9% and sterols - 2.1%. Linoleic and oleic acids dominated in both oils, followed by palmitic and linolenic acid. Unsaturated fatty acids predominated in the lipid fraction where the content of polyunsaturated fatty acids was 53.1% (white quinoa) and 49.4% (black quinoa), followed by monounsaturated fatty acids - 30.7% and 33.5%, respectively. In the tocopherol fraction of the oils the main component was  $\gamma$ - tocopherol, followed by  $\alpha$ - tocopherol. The main component of the sterol fraction was  $\beta$ - sitosterol (80.1% and 75.7%). Phospholipids in the seeds of the two varieties of quinoa had similar composition. Despite some differences in the chemical and lipid composition, these quinoa seeds were established to be a valuable source of proteins, carbohydrates and healthy lipid-soluble bioactive components for human nutrition.

**Keywords:** white and black quinoa (*Chenopodium quinoa* L.), bioactive components, proteins, lipids

### INTRODUCTION

Quinoa (*Chenopodium quinoa* Willd., genus *Chenopodium*, *Chenopodiaceae* family) is widely distributed worldwide, with around 200 - 250 varieties. Quinoa is a typical crop for Andean and can be found on the territories of Colombia, Ecuador, Peru, Bolivia, Argentina and Chile [1] but high yields are seen in East Asia, Europe, Africa, and America. Nowadays, the food industry needs new gluten-free products which are beneficial for the normal metabolic processes in the human body and satisfy the psycho-physical well-being [2]. The grains of amaranth, quinoa and buckwheat pseudocereals are free of gluten and have excellent nutritional properties [3]. Quinoa seeds have a good nutritional value and they can be used in baking, as well as for replacing rice in main courses [4].

The interest in quinoa is sparked mainly by the nutritional composition. The grains are rich in macronutrients (protein and carbohydrates), biologically active compounds including dietary fibres, minerals, amino acids, phenolics, phytosterols and vitamins (ascorbic acid, thiamin, riboflavin) [5-9]. Quinoa is rich in polyphenolic compounds (mainly vanillic acid, ferulic acid and

their derivatives, as well as main flavonoids quercetin, kaempferol and their glycosides) which are good antioxidants. Some of them have anti-inflammatory properties and can reduce the risk of chronic diseases [10-13]. The quinoa seeds can be coloured in white, black and red [7] as the antioxidant activity and phenolic profiles depend by the color. The darker quinoa seeds (black quinoa) are rich of phenolic compounds and have better antioxidant activity than the white ones.

Quinoa has different composition depending by the variety of the plant. Lipid content can vary (5.94% - 10.71%) [1]. The quantity of protein is determined to be 14.2% and starch content - 47.22% - 59.72% [14]. Quinoa can be consumed as a functional food, both seeds and oil. The oil is rich of unsaturated fatty acids and they are most important for the nutritional value of the quinoa seed oil. The content of oleic acid is 33% [15]. Tocopherols are biologically active components with good protection against oxidative damage.  $\alpha$ -Tocopherol is the major component of quinoa seed oil as the total content of tocopherol is reported to be 37.88 - 77.73 mg kg<sup>-1</sup> [16].

The color of quinoa is variable. White and black seeds are most commonly consumed but a detailed

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comparative study of these two varieties of quinoa seeds is not available. The aim of this research is to examine the chemical and lipid composition, as well as biologically active composition of both white and black quinoa seeds and oils.

## EXPERIMENTAL

All analyses were carried out with quinoa seeds purchased from the local market with country of origin Peru.

### Proximate composition analysis

The glyceride oil was isolated from the seeds with n-hexane by Soxhlet extraction [17]. The contents of proteins, ash and moisture were analysed according to AOAC [18] and that of carbohydrates was calculated using a formula [9].

### Determination of fatty acid composition

Gas chromatography was used for determination of fatty acid composition [19]. Transesterification of the oil with sulfuric acid in methanol was the way to obtain the fatty acid methyl esters (FAMES) [20]. Determination was performed on Agilent 8860 gas chromatograph. The equipment was with a capillary column DB Fast FAME (30 m x 0.25 mm x 0.25 µm (film thickness)) and a flame ionization detector (FID). The column temperature was from 70 °C (1 min), at 6 °C/min to 180 °C, and at 5 °C/min to 250 °C; the injector temperature was 270 °C and that of detector - 300 °C; nitrogen was the carrier gas. Identification was done by comparison with the retention times of a standard mixture of FAME.

### Analysis of sterols

The unsaponifiable matter was extracted with n-hexane after saponification of glyceride oil [21]. Sterols were isolated from the unsaponifiable fraction by thin-layer chromatography (TLC) [22], after that they were determined spectrophotometrically at 597 nm. The composition of sterols was analysed on a HP 5890 gas chromatograph equipped with DB 5 capillary column (25 m x 0.25 mm x 0.25 µm (film thickness)) and FID. Temperature gradient was from 90 °C (3 min) up to 290 °C at a rate of 15 °C/min and then up to 310 °C at a rate of 4 °C/min (10 min); detector temperature: 320 °C; injector temperature: 300 °C and carrier gas was hydrogen. Identification was confirmed by comparison with the retention times of a standard mixture of sterols [23].

### Analysis of tocopherols

The composition of tocopherols was determined in the oil by high performance liquid chromatography with Nucleosil Si 50-5 column (250

x 4 mm, particle size: 5 mm), fluorescence detection at 290 nm excitement and 330 nm emission. The operating conditions were mobile phase of hexane:dioxane = 96:4 (v/v) and flow rate of 1 mL/min [24].

### Analysis of phospholipids

Phospholipids were isolated from the seeds according to Folch *et al.* [25] using extraction with a mixture of chloroform and methanol (2:1, v/v). Two-dimensional TLC was used to determine the individual phospholipids [26]. Identification was performed by comparing the Rf values with those of standards. The spots of phospholipids were scrapped and mineralized with a mixture of perchloric and sulfuric acid, 1:1 (v/v), and the quantification was performed spectrophotometrically at 700 nm [27]. The total content of phospholipids was calculated.

### Statistics

All measurements were performed in triplicate (n = 3), the results were presented as mean value ± standard deviation (SD) and the significant differences were determined by one-way ANOVA (Duncan test, p < 0.05).

## RESULTS AND DISCUSSION

The chemical composition of white quinoa (WQ) and black quinoa (BQ) seeds is presented in Table 1.

**Table 1.** Chemical composition of white and black quinoa seeds

Compounds, %	Quinoa	
	White	Black
Glyceride oil	5.9±0.4 <sup>a</sup>	6.6±0.3 <sup>a</sup>
Protein	15.7±0.2 <sup>a</sup>	14.5±0.1 <sup>b</sup>
Carbohydrates:	65.6±0.6 <sup>a</sup>	64.6±0.4 <sup>a</sup>
- starch	40.2±0.3 <sup>a</sup>	40.2±0.2 <sup>a</sup>
- water soluble sugars	7.2±0.4 <sup>a</sup>	5.5±0.3 <sup>b</sup>
Ash	2.4±0.1 <sup>a</sup>	2.3±0.1 <sup>a</sup>
Moisture	10.4±0.2 <sup>a</sup>	12.0±0.5 <sup>b</sup>

<sup>a,b</sup> - Different letters mean statistical difference (Duncan test, p < 0.05).

The oil content differed among the both varieties of quinoa and it was in the range between 5.9% – 6.6%. The results were close to those reported by Nowak *et al.* [28] and Chauhan *et al.* [29]. The amount of glyceride oil was higher for BQ. Shen *et al.* [30] reported the opposite results – the content of glyceride oil in WQ was found to be 6.19% and in BQ – 5.68%. The quinoa seeds are very good sources of proteins as the quantities are close between seeds of white and black quinoa. The protein content was between 14% – 16% which is in agreement with

Diaz-Valencia *et al.* [14], while Ando *et al.* [31], Marmouzi *et al.* [32], Nowak *et al.* [28] declared about 13.0%. The content of carbohydrates was found to be 65.6% (WQ) and 64.6% (BQ) as the amount of starch was equal in both analysed types. These results are lower than reported earlier by Diaz-Valencia *et al.* [14] where the starch content was found to be 47.22% - 59.72%, but close to Ando *et al.* [31] and Marmouzi *et al.* [32] (63.7%). The quantity of water - soluble sugars was established to be 7.2% (WQ) and 5.5% (BQ). The ash content in the two analysed quinoa seeds was found to be similar (2.4%) as well as the moisture content (10.4% - WQ and 12.0% - BQ) which corresponds to the data by Przybylski *et al.* [33].

Fatty acid composition was one of the main parameters which were used for characterization of the oils in terms of their nutritional value. Fatty acid composition of white and black quinoa seed oil was established and the results are given in Table 2.

**Table 2.** Fatty acid composition of quinoa seed oil.

Fatty acids, %		Quinoa	
		White	Black
Lauric	C <sub>12:0</sub>	0.1±0.02	-*
Myristic	C <sub>14:0</sub>	0.4±0.1 <sup>a</sup>	0.2±0.05 <sup>b</sup>
Myristoleic	C <sub>14:1</sub>	0.1±0.01 <sup>a</sup>	0.1±0.02 <sup>a</sup>
Pentadecanoic	C <sub>15:0</sub>	0.1±0.03 <sup>a</sup>	0.1±0.02 <sup>a</sup>
Pentadecenoic	C <sub>15:1</sub>	0.2±0.05	-
Palmitic	C <sub>16:0</sub>	14.6±0.2 <sup>a</sup>	15.6±0.3 <sup>b</sup>
Palmitoleic	C <sub>16:1</sub>	0.3±0.1 <sup>a</sup>	0.2±0.05 <sup>a</sup>
Margaric	C <sub>17:0</sub>	0.1±0.01 <sup>a</sup>	0.1±0.02 <sup>a</sup>
Heptadecenoic	C <sub>17:1</sub>	0.1±0.01	-
Stearic	C <sub>18:0</sub>	0.2±0.05 <sup>a</sup>	0.9±0.1 <sup>b</sup>
Oleic	C <sub>18:1</sub>	28.5±0.5 <sup>a</sup>	33.2±0.3 <sup>b</sup>
Linoleic	C <sub>18:2</sub>	47.5±0.4 <sup>a</sup>	47.5±0.3 <sup>a</sup>
Linoleic (trans)	C <sub>18:2 (trans)</sub>	0.2±0.05	-
Linolenic	C <sub>18:3</sub>	5.0±0.3 <sup>a</sup>	1.9±0.2 <sup>b</sup>
Arachidic	C <sub>20:0</sub>	0.3±0.05 <sup>a</sup>	0.2±0.03 <sup>a</sup>
Eicosadienoic	C <sub>20:2</sub>	0.3±0.02	-
Eicosatrienoic	C <sub>20:3</sub>	0.1±0.01	-
Behenic	C <sub>22:0</sub>	0.4±0.1	-
Erucic	C <sub>22:1</sub>	1.5±0.3	-
Saturated fatty acids		16.2	17.1
Unsaturated fatty acids		83.8	82.9
- Monounsaturated fatty acids		30.7	33.5
- Polyunsaturated fatty acids		53.1	49.4

\* - Not identified; <sup>a,b</sup> - Different letters mean statistical difference (Duncan test,  $p < 0.05$ ).

Predominating fatty acids in the oil of WQ and BQ were the unsaturated ones (UFAs). The major part of them was polyunsaturated fatty acids (PUFA) – 53.1% for the oil of WQ and 49.4% for BQ. Two essential fatty acids, linoleic acid (C18:2) and oleic

(C18:1), were in the highest quantity in both analysed quinoa seed oils. The amount of linoleic acid was determined to be the same (47.5%) in WQ and BQ while the contents of oleic acid were different. The quantity of oleic acid (C18:1) in BQ was higher (33.2%) than that in the oil from white seeds (28.5%). The results regarding the content of linoleic acid are in agreement with these reported by Shen *et al.* [30] where the content in BQ was found to be higher than in the white one – 51.75% and 43.74%, respectively, while our results regarding the content of linolenic acid (WQ – 5.0% and BQ – 1.9%) were significantly lower (WQ – 8.24% and BQ – 4.59%). The fraction of saturated fatty acids was presented by palmitic acid (C16:0) as follows – 14.6% in WQ and 15.6% in BQ. The data are higher than those reported by Shen *et al.* [30] (13.16% and 9.77%). The rest of the fatty acids were identified in minimal quantities.

The biologically active compounds in quinoa seed oil were determined and the results are presented in Table 3.

**Table 3.** Biologically active compounds in white and black quinoa seed oils

Compounds, %	Quinoa	
	White	Black
Unsaponifiable matter, %		
- in the oil	7.3±0.3 <sup>a</sup>	4.8±0.4 <sup>b</sup>
- in the seeds	0.4±0.0 <sup>a</sup>	0.3±0.0 <sup>b</sup>
Sterols, %		
- in the oil	2.1±0.1 <sup>a</sup>	3.4±0.2 <sup>b</sup>
- in the seeds	0.1±0.0 <sup>a</sup>	0.2±0.0 <sup>b</sup>
Tocopherols, mg/kg		
- in the oil	365±17 <sup>a</sup>	1102±21 <sup>b</sup>
- in the seeds	22±1 <sup>a</sup>	73±1 <sup>b</sup>
Phospholipids, %		
- in the oil	11.9±0.5 <sup>a</sup>	7.6±0.3 <sup>b</sup>
- in the seeds	0.7±0.0 <sup>a</sup>	0.5±0.0 <sup>b</sup>

<sup>a,b</sup> - Different letters mean statistical difference (Duncan test,  $p < 0.05$ ).

The information about the content of unsaponifiable matter of quinoa seed oil was missing in the literature. Its amount in the oil from WQ was 7.3% and in BQ – 4.8%, respectively. The results are higher than the reported in Codex Stan 19 [34] for other vegetable oils where the quantity of unsaponifiable matter is 1.0% (for peanut oil), 1.5% (soybean oil) and 2.0% (rapeseed oil). The sterols were the major part of the unsaponifiable matter. There was a difference between the contents of sterol in the oils of WQ (2.1%) and BQ (3.4%). Tocopherols (known as Vitamin E) are an excellent natural antioxidant and they are important for human health. The data about the tocopherol content of oil

from WQ and BQ were notably different. The total amount of tocopherols in the oil of BQ was 4 times higher than that of WQ. The content of phospholipids in the oils of white and black quinoa was found to be 11.9% (WQ) which is notably higher than 7.6% (BQ).

The individual composition of the sterol fraction is presented in Table 4.

**Table 4.** Individual sterol composition of quinoa seed oils

Sterols, %	Quinoa	
	White	Black
Campesterol	0.5±0.1 <sup>a</sup>	1.1±0.2 <sup>b</sup>
Stigmasterol	2.5±0.2 <sup>a</sup>	2.9±0.3 <sup>a</sup>
Δ <sup>7</sup> - Campesterol	1.1±0.1 <sup>a</sup>	1.2±0.3 <sup>a</sup>
β - Sitosterol	80.1±0.5 <sup>a</sup>	75.7±0.4 <sup>b</sup>
Δ <sup>5</sup> - Avenasterol	2.8±0.2 <sup>a</sup>	3.2±0.1 <sup>b</sup>
Δ <sup>7,25</sup> - Stigmastadienol	0.9±0.1 <sup>a</sup>	7.8±0.3 <sup>b</sup>
Δ <sup>7</sup> - Stigmasterol	6.9±0.2 <sup>a</sup>	3.6±0.2 <sup>b</sup>
Δ <sup>7</sup> - Avenasterol	5.2±0.1 <sup>a</sup>	4.5±0.3 <sup>b</sup>

<sup>a,b</sup> - Different letters mean statistical difference (Duncan test, p < 0.05).

The individual composition of the sterol fraction of quinoa seed oils covers all major phytosterols. β – Sitosterol dominated, as its quantity in the oil of WQ is 80.1% and of BQ – 75.7%. The amounts of β – sitosterol reported by Shen *et al.* [30] are lower than our results as follows - 61.4% (WQ) and 58.0% (BQ). There were considerable differences between the amounts of Δ<sup>7,25</sup> – stigmastadienol and Δ<sup>7</sup> – stigmasterol. Black quinoa seed oil was rich of Δ<sup>7,25</sup> – stigmastadienol (7.8%) while in the oil of WQ its quantity was minimal (under 1%). The level of Δ<sup>7</sup> – stigmasterol in the oil of WQ was almost 2 times higher than in the oil of BQ. The other sterol components were in similar quantities in both varieties of quinoa. The content of stigmasterol (2.5%, WQ and 2.9%, BQ) was considerably different than the data by Shen *et al.* [30] (36.9%, WQ and 40.1%, BQ) while the content of campesterol (1.7% for WQ and 1.9% for BQ) is close to our results.

The individual composition of the tocopherol fraction of quinoa seed oil is presented in Table 5.

In the analysed quinoa seed oils α–, β– and γ– tocopherols were detected. The content of γ– tocopherol was similar (approximately 75.0%) in the oil of WQ and BQ and it was in the highest amount compared with α– and β– tocopherol. WQ seed oil had a slightly larger amount of α– tocopherol than BQ. β– Tocopherol was detected only in BQ seed oil.

**Table 5.** Individual tocopherol composition of quinoa seed oils

Tocopherols, %	Quinoa	
	White	Black
α- tocopherol	24.9±1.6 <sup>a</sup>	19.5±0.9 <sup>b</sup>
β- tocopherol	-*	6.0±0.5
γ- tocopherol	75.1±1.6 <sup>a</sup>	74.5±1.3 <sup>a</sup>

\* - Not identified; <sup>a,b</sup> - Different letters mean statistical difference (Duncan test, p < 0.05).

The results are in agreement with other researchers who reported γ– tocopherol as the major component of the tocopherol fraction (53.7% for WQ and 67.5% for BQ) followed by a significant content of α– tocopherol (30.9% for WQ and 21.8% for BQ) [30]. The latter authors declare a total tocopherol content of 123.09 mg/kg (WQ) and 156.67 mg/kg (BQ). The results are considerably different and much lower than those presented in the current study – 365 mg/kg in the WQ and 1102 mg/kg in the BQ.

All classes of individual phospholipids were determined in the white and black quinoa seeds. The results are given in Table 6.

**Table 6.** Individual phospholipid composition of quinoa seeds

Phospholipids, %	Quinoa	
	White	Black
Lysophosphatidylcholine	15.6±0.2 <sup>a</sup>	13.5±0.5 <sup>b</sup>
Lysophosphatidylethanolamine	9.4±0.4 <sup>a</sup>	9.0±0.3 <sup>a</sup>
Sphingomyelin	8.8±0.3 <sup>a</sup>	8.4±0.2 <sup>a</sup>
Phosphatidylinositol	18.6±0.5 <sup>a</sup>	12.8±0.3 <sup>b</sup>
Phosphatidylcholine	10.5±0.3 <sup>a</sup>	9.2±0.2 <sup>b</sup>
Phosphatidylethanolamine	8.2±0.2 <sup>a</sup>	8.5±0.1 <sup>a</sup>
Phosphatidylserine	7.4±0.4 <sup>a</sup>	8.4±0.2 <sup>b</sup>
Monophosphatidylglycerol	-*	8.1±0.1
Diphosphatidylglycerol	8.3±0.3 <sup>a</sup>	9.2±0.1 <sup>b</sup>
Phosphatidic acids	13.2±0.2 <sup>a</sup>	12.9±0.4 <sup>a</sup>

\* - Not identified; <sup>a,b</sup> - Different letters mean statistical difference (Duncan test, p < 0.05).

Lysophosphatidylcholine (LPC), phosphatidylinositol (PI), phosphatidic acids (PA) and phosphatidylcholine (PC) predominated in both quinoa seed varieties. Their quantities were similar for WQ and BQ except the phosphatidylinositol where the amount in WQ (18.6%) was notably higher than in BQ (12.8%). All other individual phospholipids (lysophosphatidylethanolamine (LPEA), sphingomyelin (SM), phosphatidylcholine (PC), phosphatidylethanolamine (PEA), phosphatidylserine (PS), monophosphatidylglycerol

(MPG) and diphosphatidylglycerol (DPG)) were observed in quantities between 7% – 11% in both quinoa seeds. The data of Przybylski *et al.* [33] are considerably different from our results: PA (1.1%), PS (4.0%), PEA (18.5%), PI (10.5%), LPEA (43.2%), PC (12.3%) and LPC (3.6%).

### CONCLUSION

The comparative analysis of white and black quinoa seeds and oils shows similar lipid compositions and contents of bioactive substances. Quinoa seeds are recognised as a rich source of essential fatty acids, nutrients, high level of protein. They are free of gluten and can be considered as a food with positive effects on the human health. With that said, white and black quinoa seeds are recommended to be consumed as functional food in the human diet.

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