Acta Sci. Pol., Technol. Aliment. 11(4) 2012, 401-410
pISSN 1644-0730 eISSN 1889-9594 www.food.actapol.net/

EFFECT OF GERMINATION PERIODS AND CONDITIONS ON CHEMICAL COMPOSITION, FATTY ACIDS AND AMINO ACIDS OF TWO BLACK CUMIN SEEDS

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ABSTRACT

Background. Chemical composition changes drastically during seeds germination because biochemical activity produces essential compounds and energy, and some nutrients transform to bioactive components as a part of these changes. There are some reports about the effect of germination on nutrient contents of different seeds, but very little information is available on black cumin seed. Besides, most studies have been conducted using a single set of germination conditions, and reports on the effect of modifying the processing conditions are scarce. The purpose of this work was to study the effect of different germination conditions on the content of chemical composition, fatty acids and amino acids of two black cumin seeds originated from Ethiopia (Africa) and Syria (Asia).
Methods. Germination process was carried out according to the Urbano method. Proximate chemical analysis were estimated using AOAC method. The fatty acid composition was determined by gas liquid chromatography and amino acid composition by procedures described by Mario. Statistical analysis was performed on all obtained values.
Results. The content of moisture, oil, crude protein, ash and total carbohydrates of raw Ethiopian sample were slightly higher than the amounts of raw Syrian sample. Germination of the two samples increased both oil and protein contents while other constituents were decreased. Little change was observed in fatty acids after germination as saturated and total monounsaturated fatty acids were decreased, while the total of polyunsaturated fatty acids was increased. The total amino acids increased during germination and it affected by germination conditions. Germinated seeds showed noticeable decreases in the contents of K, Na, Ca and Fe. The results suggested that germinated black seeds are a good food material with improved nutritional properties

Key words: germination, *Nigella sativa*, fatty acid, amino acid, minerals

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INTRODUCTION

During seeds germination the chemical composition changes drastically because biochemical activity produces essential compounds and energy [Moongngarm and Saetung 2010], some nutrients transform to bioactive components as a part of these changes. During germination lipids, carbohydrates, and storage proteins within the seed are broken down in order to obtain the energy and amino acids necessary for the plants development [Ferreira et al. 1995]. Germination process has been used for centuries for the purpose of softening the kernel structure, improving its nutritional value, reducing anti-nutritional effects and improving the functionality of seed protein [Kaukovirta-Norja et al. 2004, Ijarotimi and Keshinro 2012, Suliburska et al. 2009].

After germination of beans, histidine, glutamate, glycine, arginine, tyrosine and tryptophan contents decreased while, in lentils and peas, free protein amino acids increased after germination. Light germination gave the highest amounts of free protein amino acids in beans and lentils, but the lowest in peas. The highest free non-protein amino acids content was found in peas after dark germination [Kuo et al. 2004].

Seeds can be germinated either by soaking in ethanol then in distilled water, and kept between thick layers of cotton cloth at room temperature for three days, or by soaking in sodium hypochlorite solution for 30 min at room temperature. Then drained and washed to neutral pH, and then soaked in distilled water for 5 h 30 min [Lee and Karunanithy 1990].

Black cumin (*Negilla sativa*) is considered as unconventional oilseed, contains proteins, carbohydrates, and fixed oil (84% fatty acids, including linolenic and oleic) and volatile oils, alkaloids, saponins, crude fibre, as well as minerals, such as calcium, iron, sodium and potassium [Gali-Muhtasib et al. 2006]. Black cumin is one of the most revered medicinal seeds in history. The popularity of the plant was highly enhanced by the ideological belief in it as a cure for multiple diseases. In fact, this plant has occupied special place for the wide range of medicinal value [Ramadan 2006]. The importance of black cumin is attributed to the bioactive compound of thymoquinone which is as high as 25% in the seed oil [Pagola et al. 2004]. Phenolic rich fractions extracted from black cumin seedcake showed a potential value as natural antioxidants and were used to improve oxidative stability of corn oil [Mariod et al. 2009]. Nergiz and Otles [1993] studied the compositions of fatty acids, sterols, total polyphenols and tocopherols of black cumin seed oil besides analysis of water-soluble vitamins and minerals in the seeds. There has been a growing interest in effects of different processing methods e.g. germination, soaking, autoclaving, roasting, and boiling on chemical composition, antinutritional factors, *in-vitro* digestibility and naturally occurring compounds of seeds.

There are some reports about the effect of germination on the nutrient contents of different seeds, but very little information is available for black cumin seed. Besides, most studies have been conducted using a single set of germination conditions, and reports on the effect of modifying the processing conditions are scarce. The purpose of this work was to study the effect of different germination conditions on the content of chemical composition, fatty acids and amino acids of two black cumin seeds originated from Ethiopia (Africa) and Syria (Asia).

MATERIAL AND METHODS

Material

Black cumin seeds. Two samples (Ethiopian and Syrian) of black cumin (*Negilla sativa*) seeds were collected from local market of Khartoum north, Khartoum, Sudan. The obtained seeds were cleaned under running tap water, dried under room temperature and prepared for different analysis.

Germination. Following method of Urbano et al. [2005], the germination process was carried out, where 500 g of Ethiopian and Syrian black cumin seeds were soaked in 2500 ml of 0.7 g/L sodium hypochlorite solution for 30 min at room temperature. Seeds were then drained and washed with tap water to neutral pH, and then soaked in distilled water for 5 h. The soaked seeds were kept between thick layers of cotton cloth allowed to germinate in a seed germinator at 25°C and 80% relative humidity for 2, 4 and 6 days under light (GLD_2, GLD_4, and GLD_6) or in the dark (GDD_2, GDD_4 and GDD_6) at 20°C. Germinated seeds were dried in hot air oven at 60°C till constant weight, and then ground into fine powder to pass through 5 mm sieve and stored at ambient conditions (20°C, 60% RH) for analysis.

Methods

Proximate chemical analysis. Moisture, crude protein, ash, crude fat and crude fibre were estimated using AOAC method [1990]. Carbohydrates were calculated by difference. The proximate principles i.e. moisture, crude protein, ash, crude fat and crude fibre were summed up and subtracted from 100. Total nitrogen was determined by the micro-Kjeldahl method. The nitrogen to protein conversion factor of 6.25 was used to calculate protein content.

Oil extraction. Oil was extracted from dried germinated black cumin seeds by Soxhlet apparatus using hexane 70% and determined following a previously described method [Tian et al. 2010]. The oil content was determined as percentage of the extracted oil to the sample weight (w/w). The sample was analyzed in triplicate, and then mean and standard deviation were calculated. The extracted oil was stored in cold room in dark glass bottle for further analysis.

Mineral analysis. One gram of dried, ground seedcake of black cumin seeds was weighed and placed in a porcelain crucible. Then the sample was placed in a muffle at 500°C overnight. The ash was cooled and dissolved in 5-ml of 20% HCL, the solution was warmed through an acid washed filter paper in to a 50-ml volumetric flask. The filter paper was washed and e the solution was diluted to volume with deionizer water and mixed well [AOAC 1990]. Minerals were determined using a Perkin Elmer PE model 5000 atomic absorption spectrophotometer [Analytical... 1994].

Fatty acid composition. The fatty acid composition of the seed oils were determined by gas liquid chromatography (GLC). The oils were converted to their corresponding methyl esters following a previously described method [AOCS 1993]. BF_3 methanol reagent (14% boron trifluoride) was used for methylation. GLC analysis of the fatty acid methyl esters (FAME) was performed using a Hewlett-packard HP--5890 series II gas chromatograph (GLC) coupled to a flame ionization detector (FID) equipped with an Ultra 2 capillary column (25 m × 0.32 mm × 0.5 µm 5% biphenyl and 95% dimethyl polysiloxane, Hewlett-packard, Waldron Germany), a split injector (split ratio 88:1), and an FID. The column temperature program was 5 min at 150°C, 10°C/min to 275°C and 10 min at 275°C. The injector temperature was 250°C with split ratio of 88:1. The carrier gas was hydrogen at a flow rate of 1.6 mL/min. The detector temperature was 280°C with air and hydrogen flow rate of 460 and 33 mL/min, respectively. The fatty acid peaks were identified by comparing the retention times with those of a mixture of standard FAMEs (Sigma chemicals, Deisenhofer, Germany). Each FAME sample was analysed.

Amino acid composition. The content of dry matter total N was determined according to procedures described [Mariod et al. 2009]. The content of amino acids of defatted black cumin seeds was determined using Amino Acid Analyzer (S 433 Saykam, Germany). Sample containing 200 mg of protein in hydrolysis tube were acid hydrated with 5 ml 6NHCL and tightly closed the tube and incubated at 110°C for 24 hours, solution was filtered through filter paper 125 mm, 200 ml of the filtrate was taken, the filtrate was evaporated at 140°C for about an hour, 1 ml of diluted buffer was add to dried sample. The amino acids composition of the hydrolysed sample was determined with automatic amino acid analyser.

Statistical analysis. The analyses were performed with three replicates. The mean values and standard deviation (mean ±SD) were calculated and tested using the Student's t-test ($P > 0.05$). Statistical analysis of variance (ANOVA) was performed on all values using the statistical program Statgrafics Statistical Graphics System version 4.0 [Statgraphics 1985-1989].

RESULTS AND DISCUSSION

Proximate chemical analysis

The proximate composition of raw and germinated black cumin seeds (Ethiopian & Syrian) under light and dark for 2, 4 and 6 days is given in Table 1. Proximate analysis of the raw Ethiopian black seeds showed that moisture content (7.9%), oil content (29.1%), crude protein (26.1%), ash content (5.7%) and total carbohydrates (31.2%) were slightly higher than the amounts of the raw Syrian black cumin seed variety.

Table 1 shows the results of proximate composition of raw and germinated Syrian black cumin (SBC) seeds, the content of moisture (7.2%), oil (13.2%), crude protein (25.8%), fibre (5.8%), ash (4.3%) and total carbohydrates (43.7%) was affected by germination process, where germination in general increased

Table 1. Chemical composition of raw and germinated Ethiopian black cumin seeds*

Sample	Moisture	Oil	Protein	Ash	Fiber	Carbohydrates
Raw	7.9 ±0.1	29.1 ±0.3	26.1 ±0.3	5.7 ±0.1	8.1 ±0.1	23.1 ±1.2
GLD_2	7.4 ±0.1	29.0 ±0.7	29.0 ±0.2	3.8 ±0.1	7.4 ±0.2	23.4 ±2.1
GLD_4	7.3 ±0.1	30.6 ±0.8	30.0 ±0.2	3.9 ±0.1	7.6 ±0.2	20.6 ±1.1
GLD_6	7.5 ±0.1	31.5 ±0.1	29.0 ±0.1	3.7 ±0.3	7.5 ±0.4	20.8 ±1.1
GDD_2	7.1 ±0.1	31.8 ±0.6	28.1 ±0.2	3.8 ±0.1	7.9 ±0.3	21.3 ±1.2
GDD_4	7.5 ±0.2	32.1 ±0.7	28.7 ±0.1	3.9 ±0.1	6.2 ±0.3	21.6 ±2.0
GDD_6	8.2 ±0.1	32.5 ±0.4	28.8 ±0.1	3.7 ±0.2	7.6 ±0.4	21.0 ±1.1

*The data were reported as means ±standard deviation, n = 3.
GLD_2 – germinated at day light for 2 days. GLD_4 – germinated at day light for 4 days. GLD_6 – germinated at day light for 6 days. GDD_2 – germinated at dark for 2 days. GDD_4 – germinated at dark for 4 days. GDD_6 – germinated at dark for 6 days.

both oil and protein contents while other constituent were decreased. In the same manner as in Ethiopian black cumin (EBC) seeds, the oil content of germinated SBC seeds showed slight increase with increase of germination days, respectively, germination at dark showed higher increase in oil content than at light. Such variation in nutrient concentrations among species and varieties may be related to the variations of cultivated regions, storage conditions and maturity stage. It may also be due to geographical and climatic differences where black cumin seeds had been grown [Atta 2003]. Data on nutrient contents mentioned that black cumin seeds are considered as a beneficent source of oil and many minerals. The germinated EBC were higher in oil and protein contents than raw ones; meaning that germination process significantly ($P > 0.05$) increased oil and protein contents. This could be due to the fact that, during the germination process, several enzymes are activated and some non-protein nitrogen substances, such as nucleic acids, are produced: therefore, these can cause protein levels to be increased [Moongngarm and Saetung 2010]. A significant ($P > 0.05$) decrease was observed amongst EBC samples, in the level of available carbohydrate, fibre and ash as the carbohydrates supply main energy for seed growing. The oil content of germinated EBC showed slight increase with increase of germination days, respectively, germination at dark showed higher increase in oil content than at light. Even though lipids could be hydrolysed during germination and used to produce the necessary energy for the biochemical and physicochemical modifications, which occurred in the seed [Moongngarm and Saetung 2010].

Fatty acid composition

Fatty acid composition of EBC and SBC seeds is given in Tables 2 and 3 which show that linoleic, oleic and palmitic acids account for more than 93.0% of the total fatty acids for the two black cumin oils. This result seem to be higher than 92.5% of those reported by Cheikh-Rouhou et al. [2007] and lower than 94.0% of those reported by AI-Jassir [1992] and Ramadan and Morsel [2002]. The ratio of saturated to unsaturated fatty acids (S/U %) was 21.1% in EBC oil and 20.2% in SBC oil. These ratios were lower than 29.4% of those reported by Cheikh-Rouhou et al. [2007] for Tunisian black seed oil and 25.7% of those reported by Ramadan and Morsel [2003] for Egyptian black cumin seed oil. Again these variations may also be due to geographical and climatic differences.

Tables 2 and 3 show little change in fatty acids of EBC and SBC after the process of germination, from Table 2 saturated fatty acids of EBC were affected by germination as it decreased from 17.4% in raw sample to 15.4% in germinated sample, in the same manner total monounsaturated fatty acids showed little decrease, while the total of polyunsaturated fatty acids was increased from 57.6% in raw seeds to reach up to

Table 2. Chemical composition of raw and germinated Syrian black cumin seeds*

Sample	Moisture	Oil	Protein	Ash	Fiber	Carbohydrates
Raw	7.2 ±0.3	13.2 ±0.4	25.8 ±2.2	4.3 ±0.1	5.8 ±0.6	43.7 ±2.2
GLD_2	7.4 ±0.1	15.6 ±0.5	26.0 ±1.5	4.2 ±0.2	7.0 ±0.5	39.8 ±2.1
GLD_4	7.8 ±0.2	16.0 ±0.6	26.7 ±1.2	4.1 ±0.1	5.8 ±0.3	39.6 ±2.0
GLD_6	8.6 ±0.1	13.9 ±0.2	26.2 ±2.1	4.1 ±0.1	7.3 ±0.5	39.9 ±2.4
GDD_2	7.3 ±0.2	16.3 ±0.3	26.7 ±2.4	4.1 ±0.1	8.1 ±0.6	37.5 ±2.3
GDD_4	7.8 ±0.1	18.6 ±1.2	28.1 ±3.3	4.4 ±0.4	5.5 ±0.4	35.6 ±2.2
GDD_6	8.1 ±0.1	17.5 ±1.1	28.6 ±1.6	4.1 ±0.1	6.8 ±0.5	34.9 ±1.7

*The data were reported as means ±standard deviation, n = 3.
GLD_2 – germinated at day light for 2 days. GLD_4 – germinated at day light for 4 days. GLD_6 – germinated at day light for 6 days. GDD_2 – germinated at dark for 2 days. GDD_4 – germinated at dark for 4 days. GDD_6 – germinated at dark for 6 days.

Table 3. Fatty acid composition (as percentages) of raw and germinated Ethiopian black cumin (EBC) oil*

Fatty acids	Sample				
	control	GDL_4	GDL_6	GDD_4	GDD_6
Myristic 14:0	0.4 ±0.1	0.3 ±0.1	0.3 ±0.1	0.2 ±0.1	0.2 ±0.1
Palmitic 16:0	13.9 ±0.6	13.1 ±0.6	12.4 ±0.5	12.5 ±0.5	12.1 ±0.5
Palmitoleic 16:1	0.2 ±0.1	0.2 ±0.1	0.2 ±0.1	0.2 ±0.1	0.2 ±0.1
Stearic 18:0	2.9 ±0.3	2.8 ±0.3	2.9 ±0.3	3.2 ±0.3	2.9 ±0.3
Oleic 18:1	24.5 ±1.1	24.2 ±1.1	23.8 ±0.8	24.1 ±0.9	24.1 ±1.0
Linoleic 18:2	55.1 ±2.2	56.4 ±2.3	57.2 ±2.4	56.6 ±2.4	57.2 ±2.5
Linolenic 18:3	0.2 ±0.1	0.3 ±0.1	0.4 ±0.1	0.3 ±0.1	0.2 ±0.1
Arachidic 20:0	0.2 ±0.1	0.2 ±0.1	0.2 ±0.1	0.2 ±0.1	0.2 ±0.1
Gadoleic 20:1	0.3 ±0.1	0.3 ±0.1	0.3 ±0.1	0.3 ±0.1	0.4 ±0.2
Eicosadienoic 20:2	2.3 ±0.2	2.2 ±0.2	2.3 ±0.2	2.4 ±0.2	2.5 ±0.3
Saturated	17.4 ±0.6	16.4 ±0.5	15.8 ±0.4	16.1 ±0.5	15.4 ±0.4
Monounsaturated	25.0 ±1.1	24.7 ±1.0	24.3 ±1.0	24.6 ±1.3	24.7 ±1.3
Polyunsaturated	57.6 ±2.1	58.9 ±2.1	59.9 ±2.1	59.3 ±2.1	59.9 ±2.1

*The data were reported as means ±standard deviation, n = 3.
GLD_2 – germinated at day light for 2 days. GLD_4 – germinated at day light for 4 days. GLD_6 – germinated at day light for 6 days. GDD_2 – germinated at dark for 2 days. GDD_4 – germinated at dark for 4 days. GDD_6 – germinated at dark for 6 days. EBC – control.

59.9% in germinated seeds. From Table 3 SBC fatty acids showed little affect as a result of germination. Ethiopian and Syrian black cumin oils are very rich in both oleic and linoleic acids.

Amino acid composition

The amino acid contents of raw and germinated Ethiopian and Syrian black seeds analysed by amino acid analyser are indicated in Tables 4 and 5. In Table 4 the total amino acids of EBC raw seeds were 200.6 mg/g this amount was increased during germination at day light to reach 255.8 mg/g and it also increased during germination at dark to reach 247.2 mg/g. The total amino acids seeds was 200.6 mg/g this amount was increased in seeds germinated at day light for 2, 4 and 6 hrs to reach 245.6, 243.6, and 255.8 mg/g, respectively (Table 4).

Table 5 shows total amino acid of raw and germinated SBC seed, the total amount of raw seed was 199.8 mg/g this amount was increased to 200, 224.1 and 235.8 mg/g, as a result of germination at day light for 2, 4 and 6 days, respectively. This amount was increased to 255.0, 257.4, and 231.9 mg/g, as a result of germination at dark for 2, 4 and 6 days, respectively. The increase of total amino acids, after germination, was a result of the degradation of protein by protease and a synthesis of new enzymes, which helped to liberate the free amino acids [Moongngarm and Saetung 2010]. The amino acid profile did change to a great extent; a noticeable increase in aspartic, glutamic, and arginine acids was observed, whereas there was less increase in the available leucine, glycine and valine acids levels as germination progressed. Generally germination process increased both individual and total amino acids the reason is that the proteins in the raw seeds were degraded and converted into a soluble state after germination [Tian et al. 2010]. Germinated black cumin seeds (EBC, SBC) are an excellent source of different valuable amino acids.

Table 4. Fatty acid composition (as percentages) of raw and germinated Syrian black cumin (SBC) oil*

Fatty acids	Sample						
	control	GLD_2	GLD_4	GLD_6	GDD_2	GDD_4	GDD_6
Myristic 14:0	0.3 ±0.1	0.2 ±0.1	0.3 ±0.1	0.3 ±0.1	0.3 ±0.1	0.2 ±0.1	0.2 ±0.1
Palmitic 16:0	13.1 ±0.3	12.6 ±0.2	14.1 ±0.3	14.5 ±0.2	13.3 ±0.2	13 ±0.2	13.2 ±0.2
Palmitoleic 16:1	0.2 ±0.1	0.2 ±0.1	0.3 ±0.1	0.3 ±0.1	0.2 ±0.1	0.2 ±0.1	0.2 ±0.1
Stearic 18:0	3.2 ±0.2	3.5 ±0.2	3.3 ±0.2	3.3 ±0.2	3.4 ±0.2	3.2 ±0.2	3.4 ±0.2
Oleic 18:1	23.5 ±0.5	24.6 ±0.5	23.7 ±0.4	23.2 ±0.4	23.4 ±0.4	24.0 ±0.5	23.5 ±0.5
Linoleic 18:2	56.5 ±1.2	55.7 ±1.2	54.8 ±1.1	55.6 ±1.1	56.1 ±1.2	56.3 ±1.2	56.3 ±1.2
Linolenic 18:3	0.3 ±0.1	0.2 ±0.1	0.3 ±0.1	0.3 ±0.1	0.3 ±0.1	0.3 ±0.1	0.3 ±0.1
Arachidic 20:0	0.2 ±0.1	0.2 ±0.1	0.1 ±0.1	0.1 ±0.1	0.2 ±0.1	0.2 ±0.1	0.2 ±0.1
Gadoleic 20:1	0.3 ±0.1	0.3 ±0.1	0.2 ±0.1	0.3 ±0.1	0.3 ±0.1	0.3 ±0.1	0.3 ±0.1
Eicosadienoic 20:2	2.4 ±0.2	2.5 ±0.3	2.0 ±0.2	2.2 ±0.2	2.5 ±0.2	2.3 ±0.2	2.4 ±0.2
Saturated	16.8 ±0.4	16.5 ±0.4	17.7 ±0.5	18.1 ±0.2	17.2 ±0.2	16.6 ±0.5	17.0 ±0.5
Monounsaturatd	24.0 ±0.3	25.1 ±0.4	24.2 ±0.3	23.8 ±0.4	23.9 ±0.4	24.5 ±0.5	24.0 ±0.5
Polyunsaturated	59.2 ±0.6	58.4 ±0.2	57.1 ±0.4	58.1 ±0.5	58.9 ±0.4	58.9 ±0.2	59.0 ±0.1

*The data were reported as means ±standard deviation, n = 3.
GLD_2 – germinated at day light for 2 days. GLD_4 – germinated at day light for 4 days. GLD_6 – germinated at day light for 6 days. GDD_2 – germinated at dark for 2 days. GDD_4 – germinated at dark for 4 days. GDD_6 – germinated at dark for 6 days. SBC – control.

Table 5. Amino acid of raw and germinated Ethiopian black cumin (EBC) seed, mg/g*

Amino acid	Sample						
	control	GLD_2	GLD_4	GLD_6	GDD_2	GDD_4	GDD_6
Aspartic acid	15.8 ±0.5	18.8 ±0.2	18.9 ±0.2	20.7 ±0.4	17.3 ±0.3	18.5 ±0.4	19.3 ±0.5
Threonine	8.4 ±0.2	10.6 ±0.3	10.8 ±0.2	11.7 ±0.4	9.8 ±0.3	10.6 ±0.4	10.9 ±0.3
Serine	6.9 ±0.4	8.7 ±0.4	8.7 ±0.4	10.1 ±0.3	8.3 ±0.2	9.1 ±0.4	9.4 ±0.4
Glutamic acid	43.2 ±0.8	50.9 ±0.6	47.4 ±0.6	49.6 ±0.5	46.8 ±0.7	49.2 ±0.6	50.3 ±0.7
Glycine	11.2 ±0.2	12.2 ±0.2	12.2 ±0.2	12.3 ±0.3	11.6 ±0.4	12.2 ±0.4	12.3 ±0.4
Alanine	9.0 ±0.2	10.8 ±0.2	11.3 ±0.4	11.7 ±0.4	10.2 ±0.2	10.8 ±0.2	10.9 ±0.2
Cyctine	3.6 ±0.1	4.8 ±0.3	4.8 ±0.3	4.8 ±0.3	4.3 ±0.3	4.5 ±0.3	5.0 ±0.3
Valine	12.7 ±0.5	15.9 ±0.4	16.4 ±0.3	17.2 ±0.3	14.5 ±0.2	15.7 ±0.3	16.2 ±0.8
Methionine	2.1 ±0.2	3.7 ±0.2	3.5 ±0.1	3.3 ±0.1	3.0 ±0.1	3.1 ±0.2	3.2 ±0.3
Isolucine	9.0 ±0.2	11.2 ±0.4	11.9 ±0.3	12.6 ±0.4	10.3 ±0.3	11.2 ±0.4	11.7 ±0.4
Leucine	13.0 ±0.3	16.1 ±0.2	16.6 ±0.6	17.6 ±0.7	15.1 ±0.7	16.2 ±0.6	16.5 ±0.7
Tyrosine	3.0 ±0.2	4.3 ±0.3	4.1 ±0.3	3.1 ±0.2	4.0 ±0.4	3.5 ±0.5	3.5 ±0.5
Phenylalanine	7.6 ±0.4	10.5 ±0.5	10.1 ±0.4	11.3 ±0.3	9.4 ±0.2	10.1 ±0.3	10.5 ±0.4
Histidine	5.5 ±0.2	6.8 ±0.2	7.0 ±0.3	7.2 ±0.2	6.2 ±0.2	6.7 ±0.3	6.8 ±0.4
Lysine	9.3 ±0.2	10.8 ±0.3	11.1 ±0.3	13.1 ±0.3	10.3 ±0.3	11.1 ±0.3	11.1 ±0.3
Ammoina	11.4 ±0.4	12.4 ±0.5	12.3 ±0.6	12.5 ±0.6	12.1 ±0.7	11.9 ±0.7	12.5 ±0.8
Arginine	18.3 ±0.5	23.0 ±0.8	22.9 ±0.7	22.9 ±0.7	22.2 ±0.5	23.8 ±0.5	22.6 ±0.8
Proline	10.7 ±0.5	14.1 ±0.6	13.8 ±0.2	14.1 ±0.7	12.2 ±0.7	12.9 ±0.6	14.4 ±0.9
Total	200.6	245.6	243.6	255.8	227.6	241.1	247.2

*The data were reported as means ±standard deviation, n = 3.
GLD_2 – germinated at day light for 2 days. GLD_4 – germinated at day light for 4 days. GLD_6 – germinated at day light for 6 days. GDD_2 – germinated at dark for 2 days. GDD_4 – germinated at dark for 4 days. GDD_6 – germinated at dark for 6 days. EBC – control.

Mineral composition

Mineral analyses as essential guarantee the quality of any food product. Table 6 shows mineral content of Ethiopian and Syrian black cumin seeds, SBC raw seed showed lower Ca content (1.74 mg/100 g) than EBC (1.92 mg/100 g), and lower (0.350) amount of iron content than that of EBC (1.59 mg/100 g). SBC also showed lower amount (42.6 mg/100 g) of potassium content than (58.92 mg/100 g) of EBC. A comparison of the mineral content of the two black cumin seeds with Turkish [Nergiz and Otles 1993], and Saudi [AI-Jassir 1992] black cumin seeds from literature indicated that the major differences are in the amounts of calcium, iron, sodium and potassium. These were reported as 188, 57.5, 85.3 and 1180 mg/100 g (Turkish sample), and 0.04, 0.15, 0.75 and 7.6 mg/100 g (Saudi samples), respectively, this may be due to differences in geographic and climatic factors in the areas where black cumin seeds were grown.

From Table 6, 7, 8 germinated EBC and SBC had lower K, Na, Ca and Fe contents than the raw samples. Germinated seeds showed noticeable decreases in the contents of K, Na, Ca and Fe. These decreases

Table 6. Amino acid of raw and germinated Syrian black cumin (SBC) seed, mg/g*

Amino acid	Sample						
	control	GLD_2	GLD_4	GLD_6	GDD_2	GDD_4	GDD_6
Aspartic	16.0 ±0.5	15.9 ±0.3	17.2 ±0.5	18.2 ±0.5	17.3 ±0.5	19.8 ±0.6	18.0 ±0.5
Threonine	7.7 ±0.1	7.9 ±0.2	9.2 ±0.3	9.4 ±0.3	9.2 ±0.3	10.5 ±0.4	9.2 ±0.3
Serine	5.5 ±0.2	6.7 ±0.3	7.3 ±0.4	7.3 ±0.5	7.4 ±0.5	8.2 ±0.5	7.0 ±0.3
Glutamic	41.7 ±1.1	45.4 ±1.2	47 ±1.0	49.3 ±1.3	47.2 ±1.0	53.1 ±1.4	47.5 ±1.0
Glycine	10.2 ±0.3	10.1 ±0.3	10.5 ±0.4	11.0 ±0.5	10.6 ±0.3	11.5 ±0.6	11.0 ±0.4
Alanine	9.6 ±0.3	9.3 ±0.3	10.5 ±0.4	10.7 ±0.4	10.2 ±0.4	11.6 ±0.5	10.9 ±0.4
Cyctine	3.7 ±0.1	3.8 ±0.3	3.5 ±0.2	4.3 ±0.3	3.9 ±0.3	4.7 ±0.4	3.8 ±0.2
Valine	12.0 ±0.4	11.7 ±0.3	14.4 ±0.5	14.8 ±0.4	14.0 ±0.4	16.7 ±0.5	15 ±0.5
Methionine	2.7 ±0.1	2.4 ±0.2	3.1 ±0.3	3.5 ±0.3	3.1 ±0.2	3.5 ±0.3	3.0 ±0.2
Isolucine	9.6 ±0.3	9.3 ±0.3	10.6 ±0.4	10.9 ±0.3	10.5 ±0.3	12.5 ±0.5	11.2 ±0.2
Leucine	13.7 ±0.4	13.2 ±0.4	14.7 ±0.5	15.2 ±0.6	14.4 ±0.4	17 ±0.6	15.5 ±0.5
Tyrosine	4.0 ±0.3	3.8 ±0.2	3.8 ±0.2	3.6 ±0.1	3.5 ±0.1	4.4 ±0.3	4.0 ±0.3
Phenylalanine	6.9 ±0.4	6.0 ±0.4	9.1 ±0.5	9.6 ±0.5	9.3 ±0.4	10.7 ±0.5	9.8 ±0.5
Histidine	5.5 ±0.3	5.5 ±0.3	6.3 ±0.4	6.6 ±0.4	6.3 ±0.2	7.4 ±0.5	6.6 ±0.3
Lysine	8.8 ±0.4	8.6 ±0.3	9.3 ±0.4	10.1 ±0.5	9.6 ±0.4	10.9 ±0.5	9.8 ±0.4
Ammoina	11.0 ±0.5	12.0 ±0.5	11.9 ±0.4	13.4 ±0.6	13.2 ±0.6	12.4 ±0.5	12.6 ±0.5
Arginine	19.9 ±0.7	19.6 ±0.7	23.8 ±0.8	24.7 ±0.8	23.2 ±0.8	27.8 ±0.9	24.2 ±0.8
Proline	11.3 ±0.5	9.1 ±0.4	12.1 ±0.6	13.2 ±0.7	12.1 ±0.6	14.8 ±0.8	12.7 ±0.5
Total	199.8	200	224.1	235.8	225	257.4	231.9

*The data were reported as means ±standard deviation, n = 3.
GLD_2 – germinated at day light for 2 days. GLD_4 – germinated at day light for 4 days. GLD_6 – germinated at day light for 6 days. GDD_2 – germinated at dark for 2 days.

might be attributed to the leaching of such minerals into soaking water. Khalil and Mansour [1995] found that Na and K levels in faba beans were decreased as germination proceeded. These results are in reasonably good agreement with those reported by Lee and Karunanithy [1990].

CONCLUSIONS

The effect of different germination conditions on the content of chemical composition, fatty acids and amino acids of two black cumin seeds originated from Ethiopia (Africa) and Syria (Asia) using raw seeds as controls were investigated. Major nutrients were greatly changed during germination. For example the contents of protein and oil increased significantly ($P > 0.05$), while other constituent were decreased. Germination at dark showed greater effect on composition than at light and germination processes can improve raw black cumin seeds nutritional value, and turn black seeds into a better food material than raw grain.

Table 7. Minerals of raw and germinated Ethiopian black cumin seeds, mg/100 g*

Minerals	Sample						
	raw	GLD_2	GLD_4	GLD_6	GDD_2	GDD_4	GDD_6
Ca	1.927 ±0.5	2.123 ±0.5	2.112 ±0.2	1.911 ±0.2	2.024 ±0.3	1.679 ±0.2	1.634 ±0.5
Co	0.033 ±0.1	0.012 ±0.2	0.026 ±0.1	0.027 ±0.2	0.023 ±0.1	0.026 ±0.2	0.027 ±0.2
Cr	0.009 ±0.01	0.002 ±0.01	0.003 ±0.02	0.001 ±0.01	0.007 ±0.01	0.005 ±0.01	0.004 ±0.01
Cu	0.094 ±0.02	0.118 ±0.2	0.101 ±0.1	0.086 ±0.02	0.146 ±0.1	0.206 ±0.3	0.083 ±0.01
Fe	1.590 ±0.5	0.338 ±0.2	0.300 ±0.1	0.271 ±0.2	0.407 ±0.3	0.256 ±0.1	0.246 ±0.1
K	58.92 ±1.2	57.67 ±0.2	55.01 ±1.0	58.92 ±1.1	57.99 ±0.9	35.26 ±0.8	56.42 ±0.8
Na	22.63 ±0.8	17.45 ±0.6	20.75 ±0.5	20.75 ±0.4	20.13 ±0.8	15.15 ±0.3	17.45 ±0.4
Mn	0.155 ±0.1	0.088 ±0.02	0.110 ±0.1	0.091 ±0.02	0.108 ±0.1	0.083 ±0.02	0.072 ±0.01
Pb	0.044 ±0.01	0.031 ±0.07	0.044 ±0.08	0.003 ±0.01	0.007 ±0.04	0.004 ±0.01	0.032 ±0.01
Zn	0.522 ±0.3	0.516 ±0.3	0.391 ±0.4	0.393 ±0.4	0.561 ±0.2	0.381 ±0.1	0.434 ±0.3

*The data were reported as means ± standard deviation, n = 3.
GLD_2 – germinated at day light for 2 days. GLD_4 – germinated at day light for 4 days. GLD_6 – germinated at day light for 6 days. GDD_2 – germinated at dark for 2 days. GDD_4 – germinated at dark for 4 days. GDD_6 – germinated at dark for 6 days. EBC – control.

Table 8. Minerals of raw and germinated Syrian black cumin seeds, mg/100 g*

Minerals	Sample						
	raw	GLD_2	GLD_4	GLD_6	GDD_2	GDD_4	GDD_6
Ca	1.741 ±0.2	1.703 ±0.02	1.660 ±0.3	1.611 ±0.2	1.724 ±0.3	1.682 ±0.2	1.713 ±0.2
Co	0.012 ±0.1	0.009 ±0.02	0.017 ±0.01	0.020 ±0.02	0.024 ±0.02	0.066 ±0.02	0.038 ±0.02
Cr	0.005 ±0.02	0.001 ±0.01	0.000 ±0.0	0.006 ±0.02	0.000 ±0.00	0.007 ±0.01	0.000 ±0.0
Cu	0.083 ±0.02	0.009 ±0.01	0.102 ±0.2	0.117 ±0.1	0.267 ±0.2	0.136 ±0.1	0.168 ±0.2
Fe	0.350 ±0.1	0.061 ±0.02	0.359 ±0.2	0.283 ±0.1	0.358 ±0.2	0.269 ±0.1	0.395 ±0.2
K	42.63 ±0.4	40.784 ±0.1	36.36 ±0.6	39.65 ±0.5	33.07 ±0.4	40.85 ±0.5	33.23 ±0.7
Na	36.78 ±0.5	36.13 ±0.7	36.25 ±0.8	36.26 ±0.4	28.29 ±0.3	28.24 ±0.6	28.20 ±0.5
Mn	0.099 ±0.02	0.015 ±0.01	0.091 ±0.01	0.083 ±0.01	0.093 ±0.2	0.075 ±0.02	0.084 ±0.2
Pb	0.016 ±0.02	0.020 ±0.01	0.055 ±0.02	0.056 ±0.01	0.248 ±0.3	0.125 ±0.1	0.154 ±0.1
Zn	0.352 ±0.2	0.321 ±0.02	0.372 ±0.2	0.374 ±0.1	0.300 ±0.2	0.335 ±0.1	0.323 ±0.2

*The data were reported as means ± standard deviation, n = 3.
GLD_2 – germinated at day light for 2 days. GLD_4 – germinated at day light for 4 days. GLD_6 – germinated at day light for 6 days. GDD_2 – germinated at dark for 2 days. GDD_4 – germinated at dark for 4 days. GDD_6 – germinated at dark for 6 days. SBC – control.

REFERENCES

Analytical methods for Atomic absorption no 0303-0812. 1994. Perkin Elmer Corporation.

AI-Jassir M.S., 1992. Chemical composition and microflora of black cumin (*Nigella sativa* L.) seeds growing in Saudi Arabia. Food Chem. 45, 239-242.

AOAC. 1990. Association of Official Analytical Chemists. Official Methods of Analysis. Washington, DC, USA.

AOCS. 1993. Official methods and recommended practices of the American Oil Chemists' Society. AOCS Champaign, IL, 13b-45.

Atta M.B., 2003. Some characteristics of nigella (*Nigella sativa* L.) seed cultivated in Egypt and its lipid profile. Food Chem. 83, 63-68.

Cheikh-Rouhou S., Besbes S., Hentati B., Blecker C., Deroanne C., Attia H., 2007. *N. sativa* L.: Chemical composition and physicochemical characteristics of lipid fraction. Food Chem. 101 (2), 673-681.

Ferreira R.B., Melo T.S., Teixeira A.N., 1995. Catabolism of the seed storage proteins from *Lupinus albus*: fate of globulins during germination and seedling growth. Aust. J. Plant Phys. 22, 373-381.

Gali-Muhtasib H., Roessner A., Schneider-Stock R., 2006. Thymoquinone: A promising anti-cancer drug from natural sources. Int. J. Biochem. Cell Biol. 38, 1249-1253.

Ijarotimi O.S., Keshinro O.O., 2012. Comparison between the amino acid, fatty acid, mineral and nutritional quality of raw, germinated and fermented African locust bean (*Parkia biglobosa*) flour. Acta Sci. Pol., Technol. Aliment. 11 (2), 151-165.

Kaukovirta-Norja A., Wilhemson A., Poutanen K., 2004. Germination: A means to improve the functionality of oat. Agric. Food Sci. 13, 100-112.

Khalil A.H., Mansour E.H., 1995. The effect of cooking, autoclaving and germination on the nutritional quality of faba beans. Food Chem. 54, 177-182.

Kuo Y., Rozan P., Lambein F., Frias J., Vidal-Valverde C., 2004. Effect of different germination condition on the contents of free protein and non-protien amino acids of commercial legumes. Food Chem. 86, 537-545.

Lee C.K., Karunanithy R., 1990. Effect of germination on the chemical composition of Glycine and phasealus leans. J. Sci. Food Agri. 51, 437-445.

Mariod A.A., Aseel K.M., 2009. Characterization of the seed oil and meal from *Monechma ciliatum and Prunus mahaleb* seeds. J. Am. Oil Chem. Soc. 86, 749-755.

Mariod A.A., Ibrahim R.M., Ismail M., Ismail N., 2009. Antioxidant activity and phenolic content of phenolic rich fractions obtained from black cumin (*Nigella sativa*) seedcake. Food Chem. 116, 306-312.

Moongngarm A., Saetung N., 2010. Studies on comparison of chemical compositions and bioactive compounds of germinated rough rice and brown rice Thailand. Food Chem. 122, 782-788.

Nergiz C., Otles S., 1993. Chemical composition of *Nigella sativa* L. seeds. Food Chem. 48, 259-261.

Pagola S., Benavente A., Raschi A., Romano E., Molina M.A., Stephens P.W., 2004. Crystal structure determination of thymoquinone by high-resolution X-ray powder diffraction. AAPS Pharm. Sci. Tech. 5, e28.

Ramadan M.F., 2006. Nutritional value, functional properties and nutraceutical applications of black cumin (*Nigella sativa* L.) an overview. Int. J. Food Sci. Technol. 42, 1208-1218.

Ramadan M.F., Morsel J.T., 2002. Characterization of phospholipid composition of black cumin (*Nigella sativa* L.) seed oil. Nahrung/Food 46, 240-244.

Ramadan M.F., Morsel J.T., 2003. Analysis of glycolipids from black cumin (*Nigella sativa* L.), coriander (*Coriandrum sativum* L.) and niger (*Guizotia abyssinca* Cass.) oilseeds. Food Chem. 80, 197-204.

Statgraphics. 1985-1989. Statgraphics statistical graphics systems. V. 4.0. STSC Rockville.

Suliburska J., Krejpcio Z., Lampart-Szczapa E., Wójciak R.W., 2009. Effect of fermentation and extrusion on the release of selected minerals from lupine grain preparations. Acta Sci. Pol., Technol. Aliment. 8 (3), 87-96.

Tian B., Xie B., Shi J., Wua J., Cai Y., Xu, T., Xue S., Deng Q., 2010. Physicochemical change of oat seeds during germination. Food Chem. 119, 1195-1200.

Urbano G., Aranda P., Vılchez A., Aranda C., Cabrera L., Porres J.M., Lopez-Jurado M., 2005. Effects of germination on the composition and nutritive value of protein in *Pisum sativum*. L. Food Chem. 93, 671-679.

Received – Przyjęto: 24.05.2012

Accepted for print – Zaakceptowano do druku: 22.08.2012

For citation – Do cytowania

Mariod A.A., Edris Y.A., Cheng S.F., Abdelwahab S.I., 2012. Effect of germination periods and conditions on chemical composition, fatty acids and amino acids of two black cumin seeds. Acta Sci. Pol., Technol. Aliment. 11(4), 401-410.