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Effect of Sprouting on Chemical Composition and Amino Acid Content of Sudanese Lentil Cultivars

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Abstract: The aim of this research was to study the effect of sprouting on the chemical composition, energy and amino acid content of lentil cultivars. Three Sudanese lentil cultivars (Rubatab, Nadi and Selaim) were sprouted for 3 and 6 days. The sprouted seeds were dried and milled. The Proximate composition and amino acids content as affected by sprouting were determined. During sprouting crude fat and fiber were increased, whereas Nitrogen Free Extract (NFE) and food energy were decreased. Sprouting of the seeds was observed to affect amino acid content and showed that it increases partly or all essential and nonessential amino acids with slight variation between cultivars. For Selaim cultivar, sprouting for 3 days increased the proportion of all essential amino acids except methionine, further increase in the period of sprouting to 6 days was observed to decrease amino acid content, this was observed for histidine, lysine and arginine. This result was also observed for Rubatab cultivar in which the content of essential amino acids increased due to sprouting except methionine and lysine. In Nadi cultivar sprouting for 3 days increased the essential and nonessential amino acids. Generally, all lentil cultivars were low in their content of sulfur amino acids (methionine and cystine).

Key words: Sprouting, lentil, cultivars, composition, amino acids

INTRODUCTION

Seed germination is a primary step to generate a new plant. During germination, a series of active and complex biochemical and physiological reactions is taking place, which result in extensive changes in composition and/or morphology. Intensive investigations on compositional changes of plant seed during germination are important because the necessity of understanding the compositional changes and relevant functions from the view point of plant science. When the seeds are destined for food use, an understanding of the compositional changes resulting from germination in relation to food quality is also important (Chioce *et al*., 1997).

In Egypt, it is common to germinate some legume seeds which are rich in protein (20-50%) such as termes (*Lupin termes*), broad bean (*Vicia faba*) and chickpea (*Cicer arietinum*), before direct eating, cooking or used in salad dressing. Germination improves the nutritional value of proteins which are hydrolyzed into easily assimilable polypeptide and essential amino acids and inactivate trypsin inhibitors (Ahmed *et al*., 1995).

Germination is regarded at present as one of the cheapest and most effective procedures to increase the nutritional value of legumes, supposedly through the breakdown of certain anti-nutritional factors such as phytates, proteases inhibitors, lectins and a-glactosides (De la Cuardra *et al*., 1994).

Germination of legume seeds for human consumption has been a common practice in the orient for centuries and appears to be a simple and effective processing method for achieving desirable changes in nutritional quality. At present, germinated legumes are becoming an increasing proportion of the total consumption of food legumes in the world (Ghavidel and Prakash, 2007) and they also used to produce flours of high nutritional value.

Germination causes important changes in the biochemical, nutritional and sensory characteristics of legume seeds. Extensive breakdown of seed storage compounds and synthesis of structural proteins and other cell components takes place during this process. Fats and carbohydrates that are often at surplus levels in western diets are broken down while dietary fibre, which is mostly at a sub-optimal level, increases. Vitamins and secondary compounds, many of which are considered beneficial as anti oxidants, often are altered, dramatically during germination. Phytic acid and dietary fibre both affects the uptake of micro-nutrient in the digestive tract and these

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compounds are altered differently during the germination process. Other anti-nutrient factors, such as the flatulence-producing α galactosidase, trypsin and chymotrypsin inhibitors, which affect the digestion of proteins are also reduced after germination (Frias *et al*., 1995; Vidal-Valverde *et al*., 1994).

MATERIALS AND METHODS

A bulk of healthy and clean seeds of lentil (*Lens culinaris* Medic) cultivars (Rubatab, Nadi and Selaim) used in this study was obtained from Elhudeeba Research Station, Sudan. this study was conducted during the season 2006/2007. The bulk of each cultivar was divided into three equal portions, the first portion was reserved as a control (unsprouted seeds), the second portion was allowed to sprout for 3 days and the third portion was allowed to sprout for 6 days. Prior to sprouting, the seeds were soaked at room temperature in distilled water for 2 h. Sprouting was carried out in sterile petri-dishes lined with wet filter paper for 3 and 6 days in a refrigerator (4°C). At the end of respective sprouting period samples were dried at room temperature, ground to pass through a 0.4 screen for subsequent chemical analysis. The control groups (unsprouted seeds) were ground and kept at 4°C for further analysis. Sample of unsprouted and sprouted seeds were analyzed for total nitrogen, ether extract, crude fiber and ash (AOAC, 1980). Crude protein was calculated by multiplying the percent nitrogen by the factor 6.25. Nitrogen Free Extract (NFE) was estimated by difference. The energy content was determined by multiplying the percentages of crude protein, crude fat and NFE by factors of 4, 9 and 4, respectively (Osborne and Voogt, 1978). Five hundred milligrams of the pulverized sample (0.5 mm) was acid hydrolyzed (6 M HCl) for about 24 h in a closed bottle, then the hydrolyzed sample was divided into two portions one of them was oxidized (hydrogen peroxide/formic acid) for 24 h and then chilled, the other portion was left without oxidation, then the pH was adjusted to 2.2 with NaOH. The volume was completed to 100 mL with citrate buffer pH 2.2. Two milliliters were then filtered (membrane filter) and used for analyzing amino acid by using analyzer/ion exchange chromatography. All amino acids were detected at 570 nm, except proline which was detected using a separate detector channel at 440 nm.

RESULTS AND DISCUSSION

As shown in Table 1, there were remarkable changes in proximate composition and food energy values of lentil due to sprouting. Crude fat and crude fiber increased slightly for the three cultivars as sprouting progressed. Ash content was slightly increased for Selaim cultivar while for Rubatab and Nadi it slightly decreased. The protein content of Nadi and Selaim cultivars slightly increased while that of Rubatab it decreased with sprouting. The Nitrogen Free Extract (NFE) and food energy for all cultivars decreased with the sprouting time. The slight increment in fat content during sprouting compared to other constituents for all cultivars may be due to the fact that fat contains about twice the food energy values of protein and carbohydrate (Osborne and Voogt, 1978), the reduction in food energy value of the sprouted seeds might be attributed to the very slow increment in fat content with increasing sprouting period. Nielson and Liener (1984) and Shastry and John (1991) attributed the reduction in NFE and protein to their utilization during sprouting. Reduction of some storage nutrients of lentil seeds resulted in a concomitant increase in other nutrients.

Changes in nutrient and in anti-nutrient factors occurring during sprouting depend on the type of legume and on the sprouting conditions such as time, temperature and light cycle (Frias *et al*., 1995). This clearly indicates potential for optimization. Kuo *et al*. (2003) who also reported that the growth conditions during the sprouting process can have important effects on the composition of secondary metabolites of nutritional importance.

For Selaim cultivar sprouting produced high increase in all essential amino acids except methionine which was 0.6 g kg^{-1} in raw sample and thereafter it decreased to 0.4 g kg^{-1} after 6 days sprouting (Table 2). Sprouting of the seeds for 3 days increased amino acids content, but sprouting for longer time (6 days) decreased some of them such as threonine, histidine, lysine and arginine. Dramatic increase was seen for the essential amino acids valine, isoleucine, leucine, phenylalanine for Selaim sprouted for 6 days. There was an increment in aspartic acid and proline which were also detected by Kuo *et al*. (2003) in sprouted lentil. In Rubatab cultivar the increment of most amino acids such as threonine, valine, isoleucine, phenylalanine and histidine was observed after 3 days sprouting. Tow of the essential amino acids (methionine and lysine) of this cultivar decreased due to sprouting for 6 days.

For Nadi cultivar sprouting resulted in an increase in amino acids content such as threonine, valine, methionine, isoleucine, leucine, phenylalanine, lysine and arginine after 3 days sprouting; these were. There was a decrease in some amino acids such as aspartic acid, tyrosine and glycine in this cultivar due to sprouting; such a decrease was also observed by Kuo *et al*. (2003) in sprouted lentil. The increment of amino acids during sprouting may be attributed to the fact that seedlings are

Table 1: Proximate composition (%) and energy value (kcal/100 g) of lentil cultivars as affected by sprouting

Cultivars	Sprouting time (days)	Crud fibre	Ash	Crude protein	Fat	Nitrogen free extract	Energy
Rubatab	0	3.7±1.0^{b}	2.50±0.03^{a}	28.2±0.03^{a}	0.60±0.0^{b}	67.5±0.01^{a}	388.2±0.10^{a}
	3	5.2±0.5^{a}	2.50±0.005^{a}	27.0±0.34^{a}	0.60±0.01^{b}	64.7±0.02^{b}	372.2±0.15^{b}
	6	5.8±0.07^{a}	2.40±0.01^{b}	27.6±0.54^{a}	0.67±0.0^{a}	63.5±0.208^{c}	370.7±0.208^{a}
Nadi	0	1.9±0.1^{b}	3.90±0.06^{b}	24.5±0.00^{b}	0.65±0.06^{c}	70.0±0.136^{a}	383.0±0.10^{a}
	3	5.7±0.15^{a}	3.10±0.07^{a}	26.5±1.60^{a}	0.80±0.01^{b}	64.0±0.15^{c}	368.9±0.20^{c}
	6	5.8±0.1^{a}	3.00±1.0^{b}	25.7±0.04^{a}	0.97±0.01^{a}	64.6±0.01^{b}	369.6±0.15^{b}
Selaim	0	1.6±0.03^{b}	2.80±1.0^{b}	27.9±0.15^{a}	0.45±0.01^{c}	67.3±0.15^{a}	384.9±0.15^{a}
	3	5.0±0.11^{a}	3.00±1.0^{b}	28.1±0.13^{a}	0.52±0.05^{b}	63.4±0.20^{c}	370.4±0.15^{c}
	6	6.4±0.076^{a}	3.00±1.0^{b}	28.4±0.08^{a}	0.75±0.01^{a}	64.5±0.10b	378.4±0.15^{b}

Values are means±SD; Means not sharing a common letter in a column are significantly different at (p = 0.05) as assessed by Duncan multiple range test

Table 2: Effect of sprouting on amino acid content (g kg^{-1} US) of lentil cultivars

	Cultivars								
	Selaim			Rubatab			Nadi		
	Sprouting time (days)			Sprouting time (days)			Sprouting time (days)		
Amino acid	0	3	6	0	3	6	0	3	6
Aspartic acid	28.28	35.28	41.56	30.95	32.37	29.69	26.46	27.23	29.48
Threonine	8.88	9.68	9.48	10.01	10.22	9.57	8.26	9.04	7.86
Serine	13.99	14.96	14.61	15.12	15.10	14.55	13.20	12.67	12.38
Glutamic acid	39.59	37.43	35.63	42.01	39.47	42.44	35.99	35.52	33.35
Glycine	9.65	9.95	9.46	10.41	10.38	10.01	9.21	9.03	8.44
Alanine	10.33	11.63	11.79	11.13	11.61	10.68	9.83	1.05	9.62
Cystine	3.25	3.17	3.08	2.75	2.77	2.83	2.93	2.82	2.72
Valine	8.49	9.78	10.54	10.85	10.99	10.01	7.13	9.51	7.27
Methionine	<0.553	<0.334	<0.387	<0.368	<0.365	<0.362	<0.231	<0.718	<0.459
Isoleucine	7.21	8.12	8.60	9.55	9.57	8.72	5.87	8.33	5.75
Leucine	16.44	17.34	16.75	18.96	18.70	17.88	14.71	15.79	13.80
Tyrosine	6.98	6.99	7.02	7.32	7.19	7.46	6.29	5.81	5.77
Phenylalanine	11.09	12.15	12.24	12.99	13.07	12.19	10.16	11.03	9.82
Histidine	7.69	8.25	8.06	8.14	8.16	7.79	7.70	7.11	7.23
Lysine	15.13	15.69	15.17	17.15	16.92	16.28	14.62	15.24	13.38
Arginine	18.65	18.99	18.16	23.02	21.69	21.06	16.81	17.10	14.73
Proline	9.73	11.07	11.75	10.82	10.91	10.22	8.93	9.34	9.59

Values are means of duplicate samples

the site of high amino acid biosynthetic activity, resulting in high content of free amino acid. Also storage proteins can undergo proteolysis and contribute to the increase of free amino acids.

CONCLUSION

Germination resulted in remarkable changes in proximate composition and food energy values of lentil. Lentil was low in sulphur-containing amino acids (methionine, cysteine), germination caused an improvement in content of essential and nonessential amino acids.

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