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Diversity for vitamin and amino acid content in grass pea (*Lathyrus sativus* L.)

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ABSTRACT

This study was conducted to determine vitamin A, B, C, β-carotene and amino acid profile in 18 genotypes and four grass pea (*Lathyrus sativus* L.) varieties which have low β-ODAP. Present results indicated that retinol, β-carotene, thiamine, riboflavin, niacin, pantothene, pyridoxine, folic acid and ascorbic acid ranged from 25.6 to 44.1 µg/kg; 240.8 to 410.1 µg/kg, 3.74 to 5.44; 1.86 to 2.76; 12.37 to 20.25; 14.43 to 22.41; 4.92 to 6.62; 4.04 to 6.77 and 33.4 to 58.2 mg/kg, respectively in seeds. In addition to, the amino acid profile of the genotypes differed significantly and total amino acid amounts were found to be 19.69 to 23.48 g/100 g seeds. A large and significant variation was observed among these genotypes with low β-ODAP content in respect to the quality of the nutrient content. This variability will be useful to breeders for utilization in grass pea improvement.

Key words: Amino acids, β-ODAP, Diversity, Grass pea, Lathyrus sativus, Vitamins.

INTRODUCTION

Due to serious apprehension about food security and agricultural sustainability especially due to global climatic changes, the explotation of under utilized plants is gaining importance. The under-utilized plants, grass pea (*Lathyrus sativus* L.), which is nutritionally rich, well adapted to adverse environments and suited for low input agriculture have been thrown attention and its importance is now well recognized in the world (Campbell 1997; Kumar *et al.,* 2011).

The genus Lathyrus, which belongs to Fabaceae family includes 187 species (Jackson and Yunus, 1984; Plitmann *et al.,* 1995). Grass pea is a well adapted species to adverse environments such as drought, flood, insect attacks (Campbell 1997). Grass pea has high nutritional value and is a good source of minerals. Its protein and carbohydrate content values are up to 31 and 65% respectively (Tamburino *et al.*, 2012; Mondal and Puteh, 2014).

Grass pea, providing an economic yield under adverse environmental conditions and with a great potential for use in marginal low-rainfall areas is a popular crop in subsistence farming in certain developing countries (VazPatto *et al.*, 2006: Arslan, 2016). The chemical components and their amount in grass pea may vary according to varieties/genotype, geographical region of their cultivation, maturity and environmental factors (Wiryavan and Dingle, 1999).

However, β-ODAP (β-*N*-oxalyl-L-α, β-diaminop-ropioni c acid) is a neurotoxin compound which is the non-protein amino acid and found abundantly in the seeds of grass pea (Arslan *et al.*, 2017; Mondal and Puteh, 2014; Fikre *et al.*, 2008) and can lead to lathyrism (Hanbury *et al.*, 1999). β-ODAP compound amounts of grass pea vary from 0.04 up to 1.6% in seed depending on genotype and environment (Hanbury *et al.,* 1999: Tamburino *et al.,* 2012), and its critical value is less than 0.2% (Abd El-Moneim *et al.,* 2010). This toxic compound is the main limiting factor in the utilization of grass pea (Sharma *et al.*, 2000).

Vitamins, present in minute amounts in natural foodstuffs are a group of complex organic compounds, essential for normal metabolism. They are classified not on chemical characteristics but on function. There are 15 vitamins which are identified and classified as either fat or water-soluble. However, the number of compounds that can justifiably be classified as vitamins is controversial because of being ascribed the term vitamin to many substances that do not meet the criteria for vitamin identification (McDowell, 2000; Xu *et.al,* 2017).

Vitamins which are related to many degenerative diseases, such as cancer and diabetes have been determined to play an important role in the prevention of diseases. (Diplock, 1991; Sahin *et al.,* 2005). Free radicals which form under normal physiological conditions as a result of increasing stress are made ineffective by antioxidant vitamin and enzymes in cells. When antioxidant vitamins, which are barrier against free radicals in both animals and plants decrease in cells, free radical molecules destroy the cell components. The construction of a connection between the

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increase in free radicals and degenerative diseases has increased the importance of antioxidant vitamin and enzymes (Jenkins and Hidiroglou, 1972; Sahin *et al.,* 2005).

Legumes have an important place in human and animal nutrition. Legumes and their seeds contain many vitamins such as vitamin K, vitamin B1, B2, B6, vitamin C, vitamin E, niacin are good sources of vitamins. Determining the vitamin content and amount of food is important to standardize the quantity of food that must be taken daily for human and animal nutrition. Proteins are made up of various combinations up to 26 amino acids. The amino acids which are used principally as building blocks for the synthesis of proteins are the required and vital nutrients to maintain physiological activities of human and animals. (Ates *et al.,* 2010).

Amino acids are classified as either essential or nonessential. Nonessential amino acids can be synthesized by most animals. The essential amino acids are arginine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan, and valine. Because Monogastric animals are not capable of synthesising amino acids, their diets should be supplemented with proteins containing the 10 essential amino acids (Galili *et al.,* 2005; Galili and Amir, 2013).

Legumes and their seeds are all valuable sources of protein. The average protein content in legume seeds is 21-25%. Legume seeds are rich in lysine and characteristically poorer in sulfur-containing amino acids (methionine and cysteine) in comparison to cereals. Legumes are an excellent complementary protein source for cereals. (Fikre *et al.,* 2008; Aniszewski *et al.,* 2013).

In the light of information the aim of this study is to determine of vitamin and amino acid profile and content in seeds of 18 grass pea genotypes and four varieties with low β-ODAP contents (1.55 to 3.52 mg/g), which were selected from 173 genotypes with known β-ODAP content (Arslan *et al.,* 2017) in earlier study.

MATERIALS AND METHODS

Plant materials: Eighteen genotypes and four varieties with low β-ODAP contents (1.55 to 3.52 mg/g) were selected from 173 genotypeswith known β-ODAP content. Analyses of vitamins and amino acids were conducted at Akdeniz University, Food and Agricultural Research Laboratory. Each sample was analyzed in triplicate and results were given as mg/kg in the dry material. The plant extracts required for the analyzes were prepared according to the method described in the article by Arslan *et al.* (2017).

Vitamin analyses

B group vitamins and vitamin C analysis: Quantification of water-soluble B vitamins (vitamin B1, vitamin B2, vitamin B3, vitamin B5, vitamin B6, vitamin B7, vitamin B9, vitamin B12) and vitamin C (ascorbic acid) in grass pea seeds was performed according to the method described by Kývrak (2015) and using UHPLC-MS/MS device.

According to this method, 3 g of homogenised sample was mixed with extraction solvent (30 mL of water–acetonitrile (80:20; v/v)) and the mixture was extracted and waited at 4 ºC for 6 h, then placed in an ultrasonic bath for 15 min, centrifuged at 4000 rpm at 20 ºC for 10 min. The supernatant obtained after centrifugation was filtered through 0.22µm PTFE membrane and then injected into UHPLC-MS / MS.

Vitamin A (Retinol) and β-carotene analysis: Extraction of vitamin A (Retinol) and carotenoids from grass pea seed samples was performed according to the method described by Duriot *et al.* (2010). According to this method, 3 g of the homogenized sample were mixed with extraction solvents (10 ml of acetone and 2 ml of diethylether). The mixture was vortexed for 5 minutes, centrifuged at 4000 rpm at 20 ºC for 10 min. After centrifugation, The extract liquid phase, obtained was transferred to another tube and evaporated to dryness under nitrogen, and the dry residue was dissolved in 1 ml of acetonitrile and filtered through a 0.22 µm PTFE membrane and then injected into UHPLC-MS / MS.

Amino acid analysis: The total amount of amino acids in grass pea (Glycine, Alanine, Serine, Proline, Valine, Threonine, Cysteine, Isoleucine, Leucine, Aspargine, Lysine, Glutamine, Methionine, Histidine, Phenylalanine, Arginine, Trozin, Tryptophan and Cystine) were identified in the LC-MS/MS device *by modifying the methods* described by Lee and Hwang (2017), Chan and Matanjun (2017). According to this method, 0.2 g of the homogenized sample was weighed and 10 mL of 6 N HCl solution was added. The obtained mixture was tightly capped and mixed with the vortex for 5 minutes in the test tube and then placed in a 110 °C oven for 24 hours to complete hydrolysis. The mixture cooled to room temperature was centrifuged at 4000 rpm at 4°C for 15 minutes. The supernatant obtained after centrifugation was filtered through a 0.45 µm PTFE membrane and then injected into LC-MS/MS. Analysis was carried out by making necessary modifications to device conditions reported by Kivrak (2015).

Statistical analysis: All analyses were performed in triplicate and their means were reported. Pearson's correlation coefficient and ANOVA for comparisons were calculated using SAS 9.1 software (SAS Institute Inc., 2003).

RESULTS AND DISSUSSION

Variability in vitamin contents: In the present study, vitamins and amino acid contents of 22 grass pea genotypes with low β-ODAP contents were determined. The levels of vitamins A, B, C and β-carotene in seeds of grass pea genotypes were calculated based on dry weight (Table 1). Large and significant variation was observed among the genotypes for vitamin content. The result indicate that the

Table 1: Vitamin contents in grass pea genotypes

Genotypes	Retinol (vitamin A) µg/kg	β-Carotene µg/kg	Thiamine (B1)mg/kg	Riboflavin (B2)mg/kg	Niacin (B3)mg/kg	Pantothenic cid (B5) mg/kg	Pyridoxine (B6) mg/kg	Folic acid(B9) mg/kg	Ascorbic acid (vitamin C) mg/kg
LS-89	27.4	246.4	5.28	2.68	19.59	21.65	6.46	6.51	53.8
LS-96	26.5	243.8	5.36	2.72	19.97	22.03	6.54	6.64	56.5
LS-105	44.1	410.1	3.74	1.86	12.37	14.43	4.92	4.04	33.4
LS-106	39.7	373.2	4.15	2.08	14.27	16.33	5.32	4.69	37.2
LS-113	33.5	305.2	4.71	2.38	16.93	18.99	5.89	5.60	41.9
LS-115	41.5	393.9	3.98	1.99	13.51	15.57	5.16	4.43	35.3
LS-118	31.8	298.7	4.87	2.46	17.69	19.75	6.05	5.86	43.3
LS-122	42.3	381.1	3.90	1.95	13.13	15.19	5.08	4.30	34.9
LS-124	29.1	268.1	5.12	2.59	18.83	20.89	6.30	6.25	48.4
LS-140	30.0	282.2	5.04	2.55	18.45	20.51	6.22	6.12	46.7
LS-141	37.1	341.0	4.39	2.21	15.41	17.47	5.57	5.08	39.1
LS-142	38.8	361.0	4.23	2.12	14.65	16.71	5.41	4.82	37.7
LS-152	35.3	331.8	4.55	2.29	16.17	18.23	5.73	5.34	40.5
LS-153	36.2	329.2	4.47	2.25	15.79	17.85	5.65	5.21	39.8
LS-156	37.9	360.4	4.31	2.16	15.03	17.09	5.49	4.95	38.4
LS-165	43.2	397.6	3.82	1.90	12.75	14.81	5.00	4.17	34.8
LS-166	30.9	287.4	4.96	2.51	18.07	20.13	6.14	5.99	44.4
LS-169	34.4	323.5	4.63	2.33	16.55	18.61	5.81	5.47	41.2
Ceora	28.3	257.2	5.20	2.64	19.21	21.27	6.38	6.38	50.1
Eren	40.6	385.5	4.06	2.03	13.89	15.95	5.24	4.56	36.8
İptaş	25.6	240.8	5.44	2.76	20.25	22.41	6.62	6.77	58.2
Karadağ	32.7	293.9	4.79	2.42	17.31	19.37	5.97	5.73	42.6
Means	34.9	323.3	4.60	2.30	16.40	18.40	5.80	5.40	42.5
LSD	**	**	**	**	**	**	**	**	**

**: Statistically significant at P = 0.01 significance level.

least amount of vitamin was retinol and β-carotene which were in the range of 25.6 - 44.1 and 240.8 - 410.1 µg/kg, respectively in genotypes. The highest level of vitamin was found to be pantothenic acid or pantothene (14.43 - 22.41 mg/kg) was followed by niacin (12.37 - 20.25 mg/kg).

Among the genotypes, LS-105 genotype was found to have the maximum vitamin A and β-carotene content and İptaþ variety with lowest level of vitamin A and β-carotene content. The lowest vitamin B and vitamin C content were found in LS-105 genotype while the highest vitamin B and vitamin C were found in İptaþ variety (Figure 1).

Sahin *et al.* (2009) reported that in *Lathyrus* species, retinol and ascorbic acid contents were in the range of 0.03 to 1.05 µg/g and 0.794 to 23.779 µg/g respectively. Chavan *et al.* (1999) found that the contents of ascorbic acid, β-carotene, folic acid, riboflavin, thiamine and niacin in seeds of *Lathyrus maritimus* L. were 1.60; 12.17; 0.08; 0.06; 0.59 and 3.44 mg/100 g, respectively. Korus *et al.* (2002) found that in the raw seeds of *Lathyrus* cultivar, the mean contents of Vitamin C, riboflavin, β-carotene were 31.5, 0.131, 0.31 mg/100g fresh matter respectively.

The finding of our study indicate that retinol contents were lower than those reported for *Lathyrus* species studied by Sahin *et al.* (2009). Ascorbic acid contents were higher than that reported for *Lathyrus maritimus* by Chavan *et al.* (1999) but were similar to findings of Sahin *et al.* (2009) and β-carotene contents were found higher than that of Chavan *et al* (1999). The vitamin content of plant species is related to certain factors, such as genetic features and ecological conditions (Munzuroglu *et al.*, 2000).

Table 2: Correlation coefficients of vitamin content in grass pea genotypes.

	Retinol	β-Carotene	Thiamine	Folic acid	Pyridoxine	Niacin	Pantothenic acid	Riboflavin
β-Carotene	0.994**							
Thiamine	-1.000*	-0.994**						
Folic acid	-1.000*	-0.994**	1.000*					
Pyridoxine	-1.000*	-0.994**	1.000*	1.000*				
Niacin	-1.000*	-0.994**	1.000**	1.000**	1.000**			
Pantothenic acid	-1.000*	-0.994**	1.000*	1.000*	1.000*	1.000**		
Riboflavin	-1.000*	-0.994**	1.000*	1.000*	1.000*	1.000**	1.000*	
Ascorbic acid	-0.954**	-0.949**	0.954**	0.954**	0.954**	0.953**	0.954**	0.954**

* Significant at the 0.05 probability level, ** Significant at the 0.01 probability level.

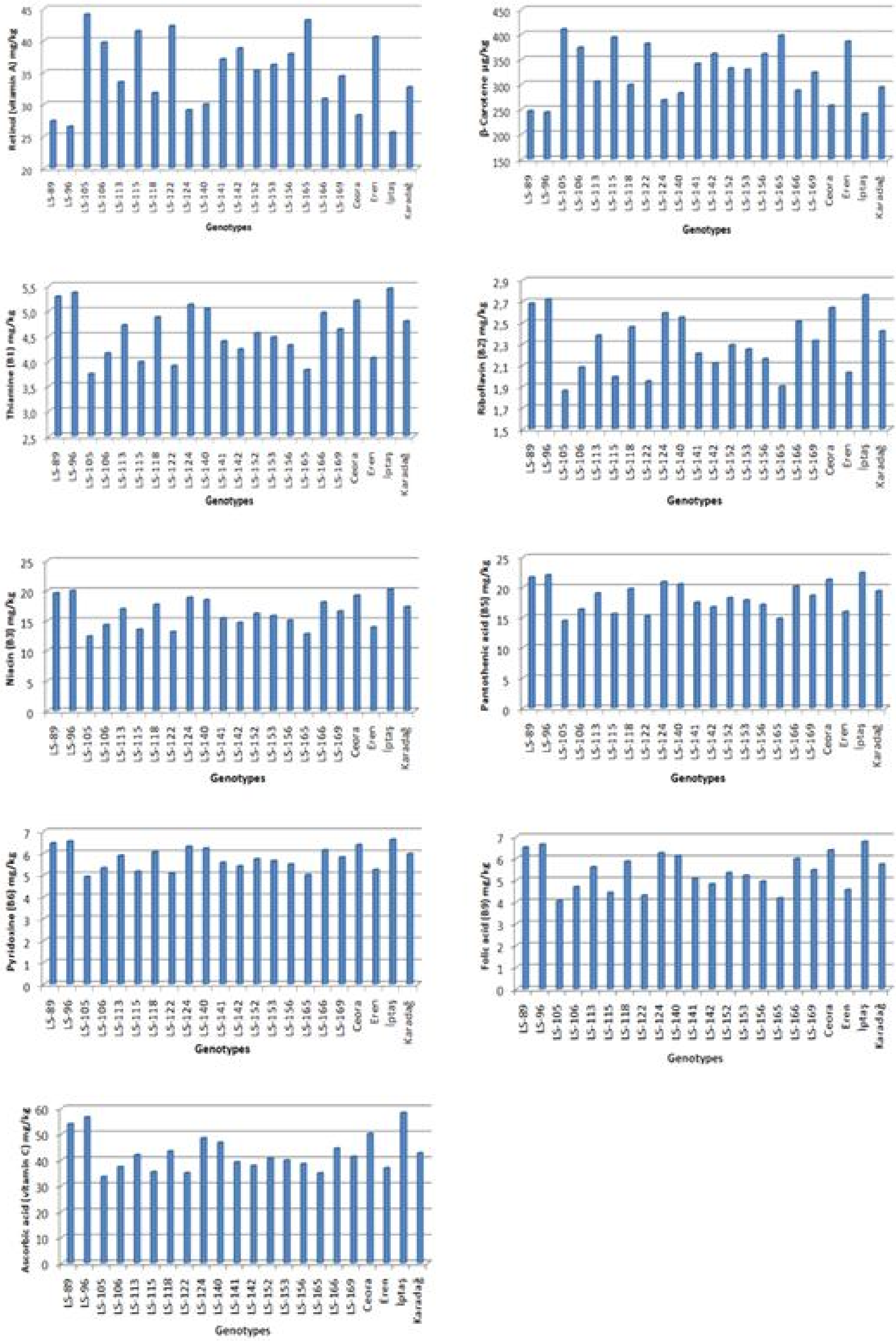

Fig-1: Vitamin contents of grass pea genotypes

Table 3: Amino acid contents in grass pea genotypes (g/100g seeds)

	Essential amino acids								Non essential amino acids								
Genotypes	**Valine**	**Isoleucine+ leucine**	**Threonine**	**Methionine**	**Lysine**	**Phenylalanine**	**Histidine**	**Tyrosine**	**Arginine**	**Alanine**	**Glycine**	**Serine**	**Proline**	**Aspartic**	**Glutamic acid**	**Cystine acid**	**Total protein amino acid**
LS-89	0.89	2.41	0.79	0.24	1.30	1.19	0.58	0.51	1.79	0.955	0.88	1.06	0.82	2.36	3.70	0.29	19.75
LS-96	0.87	2.29	0.77	0.27	1.39	1.12	0.57	0.48	1.97	0.894	0.87	0.95	0.86	2.45	3.65	0.28	19.69
LS-105	1.04	2.83	0.99	0.32	1.65	1.33	0.72	0.62	2.24	1.017	1.04	1.25	1.02	2.82	4.22	0.38	23.48
LS-106	1.13	2.74	0.90	0.31	1.60	1.29	0.66	0.60	2.11	0.958	1.14	1.21	0.99	2.75	4.25	0.33	22.96
LS-113	0.96	2.62	0.86	0.29	1.53	1.09	0.63	0.57	2.16	0.981	0.96	1.16	0.95	2.69	4.11	0.31	21.88
LS-115	1.01	2.77	0.91	0.31	1.62	1.36	0.67	0.64	2.22	1.008	1.02	1.23	1.12	2.77	4.22	0.33	23.20
LS-118	0.95	2.57	0.84	0.34	1.50	1.21	0.62	0.56	2.12	0.966	0.88	1.13	0.93	2.65	3.94	0.31	21.52
LS-122	1.14	2.79	0.91	0.31	1.63	1.32	0.61	0.66	2.22	1.011	1.02	1.23	1.01	2.63	4.16	0.33	23.01
LS-124	0.91	2.47	0.81	0.28	1.45	1.17	0.60	0.54	2.04	0.93	0.91	0.89	0.90	2.55	3.80	0.28	20.51
LS-140	0.92	2.50	0.82	0.28	1.46	1.18	0.65	0.55	2.07	0.942	0.92	1.11	0.91	2.59	3.84	0.30	21.05
LS-141	0.99	2.69	0.80	0.30	1.57	1.27	0.63	0.59	2.18	0.993	0.99	1.26	0.97	2.73	4.13	0.32	22.40
LS-142	1.07	2.72	0.89	0.31	1.68	1.20	0.66	0.59	2.20	0.935	0.98	1.20	0.99	2.74	4.18	0.32	22.67
LS-152	0.98	2.65	0.87	0.30	1.55	1.25	0.64	0.56	2.17	0.987	0.92	1.17	0.91	2.63	4.07	0.36	22.02
LS-153	0.94	2.67	0.87	0.35	1.78	1.26	0.61	0.52	2.12	0.99e	0.98	1.18	0.97	2.72	4.10	0.32	22.39
LS-156	1.00	2.70	0.81	0.30	1.58	1.27	0.65	0.53	2.19	0.996	0.99	1.19	0.98	2.74	4.15	0.30	22.40
LS-165	1.03	2.72	1.05	0.32	1.72	1.37	0.68	0.61	2.23	1.045	1.03	1.18	1.02	2.78	4.31	0.34	23.44
LS-166	0.85	2.54	0.83	0.29	1.42	1.26	0.61	0.50	2.10	0.954	0.93	1.12	0.92	2.62	3.89	0.30	21.14
LS-169	0.97	2.55	0.86	0.34	1.54	1.24	0.64	0.58	2.16	0.984	0.97	1.16	0.95	2.65	3.81	0.31	21.73
Ceora	0.90	2.44	0.80	0.27	1.43	1.15	0.59	0.53	2.02	0.918	0.90	1.08	0.88	2.52	3.75	0.29	20.46
Eren	1.02	2.76	0.90	0.31	1.76	1.36	0.67	0.72	2.21	1.005	1.01	1.22	0.97	2.76	4.23	0.34	23.23
Ýptaþ	0.85	2.45	0.71	0.26	1.37	1.03	0.57	0.50	1.89	0.882	0.86	1.02	0.84	2.42	3.54	0.28	19.47
Karadað	0.91	2.60	0.85	0.29	1.52	1.23	0.63	0.57	2.15	0.978	0.95	1.15	0.94	2.69	3.85	0.31	21.61
Means	0.97	2.61	0.86	0.30	1.55	1.23	0.63	0.57	2.12	0.969	0.96	1.14	0.95	2.65	4.00	0.32	21.82
LSD	0.0079**	0.0118**	0.0035**	0.0116**	0.0077**	0.0101**	0.0101**	0.0035**	0.0077**	0.0104**	0.0118**	0.0035**	0.0069**	0.0077**	0.002**	0.0104**	0.0077**

**: Statistically significant at P = 0.01 significance level.

β-carotene, which is a precursor of vitamin A is converted into vitamin A in the intestine and the liver. It is an important antioxidant that suppresses the formation of free radicals by preventing the oxidation of unsaturated fats. Free radicals have a very effective role in the degeneration of enzymes, proteins and lipids in tissues and cellular membranes. Vitamin A, which ranks among fat-soluble vitamins, is not much found in nature but β-carotene is found widely in plants. Ayaşan and Karakozak (2010), Wang *et al.* (1988) reported that consuption of β-carotene in abundant quantities will cater the requirement of vitamin A.

Association analysis of vitamins: Correlations of vitamin content of grass pea genotypes (Table 2) indicate that retinol and β-carotene were strongly associated with the other vitamins at the significance level of 0.01 and 0.05 to the negative direction, while other vitamins seem to have significant associations with each other at the significance level of 0.01 to the positive direction.

Variability in amino acid profiles: The amino acid composition in food determines mainly the nutritional quality of food proteins (Pastor-Cavada *et al.*, 2011). In the present study, the levels of amino acid in seeds of 22 grass pea genotypes with low β-ODAP contents were calculated based on dry weight (Table 3). Significant differences were observed among the genotypes for the trait.

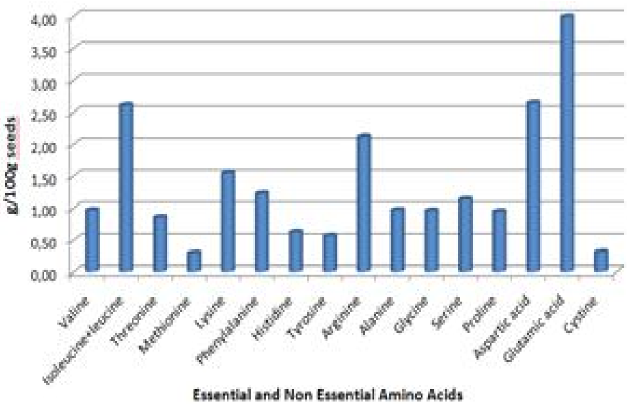

Fig-2: Amino acid profile in grass pea genotypes (mean of 22 genotypes)

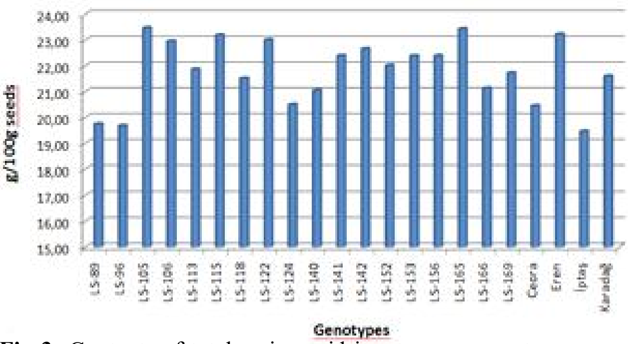

Fig-3: Contents of total amino acid in grass pea genotypes

Table 4: Correlation coefficients of amino acid contents in grass pea genotypes.

	Alanine	Arginine	Aspartic acid	Cystine	Glutamic acid	Glycine	Histidine	Isoleucine+ lanine	Lysine	Methio-nine	Phenyla-leucine	Proline	Serine	Threonine	Tyrosine	Valine
Arginine	0.665**															
Aspartic acid	0.678**	0.923**														
Cystine	0.715**	0.737**	0.744**													
Glutamic acid	0.761**	0.833**	0.899**	0.786**												
Glycine	0.617**	0.673**	0.796**	0.675**	0.844**											
Histidine	0.673**	0.779**	0.868**	0.814**	0.810**	0.737**										
Isoleucine+ leucine	0.754**	0.841**	0.885**	0.837**	0.927**	0.840**	0.810**									
Lysine	0.653**	0.814**	0.862**	0.719**	0.860**	0.732**	0.699**	0.846**								
Methionine	0.554**	0.763**	0.766**	0.575**	0.634**	0.539**	0.551**	0.665**	0.790**							
Phenylalanine	0.834**	0.702**	0.730**	0.726**	0.787**	0.741**	0.723**	0.779**	0.716**	0.574**						
Proline	0.692**	0.846**	0.873**	0.677**	0.850**	0.794**	0.772**	0.875**	0.771**	0.683**	0.775**					
Serine	0.735**	0.746**	0.788**	0.800**	0.811**	0.732**	0.723**	0.878**	0.720**	0.633**	0.708**	0.759**				
Threonine	0.771**	0.728**	0.759**	0.793**	0.804**	0.723**	0.803**	0.751**	0.756**	0.595**	0.786**	0.743**	0.613**			
Tyrosine	0.635**	0.697**	0.663**	0.702**	0.730**	0.681**	0.658**	0.790**	0.709**	0.474*	0.703**	0.710**	0.670**	0.681**		
Valine	0.570**	0.684**	0.687**	0.695**	0.831**	0.858**	0.658**	0.839**	0.713**	0.545**	0.653**	0.740**	0.718**	0.699**	0.766**	
Total protein amino acid	0.794**	0.901**	0.939**	0.842**	0.963**	0.866**	0.857**	0.963**	0.900**	0.734**	0.845**	0.907**	0.862**	0.846**	0.801**	0.843**

* Significant at the 0.05 probability level, ** Significant at the 0.01 probability level.

Glutamic acid is found to be the most abundant amino acid in the grass pea genotypes (3.54 to 4.31 g/100 g) followed by aspartic acid, arginine, isoleucine+leucine and lysine. Sulfur amino acid methionine (0.24 - 0.35 g/100 g) and cystine (0.28 - 0.38 g/100 g) were found at lower levels. These amino acids were followed sequentially by tyrosine, histidine, and threonine. The total amino acid content in the grass pea genotypes range from 19.47 to 23.48 g/100 g seeds. The amino acid profile and total amino acid of grass pea seeds are presented in Figure 2 and Figure 3.

In respect to the amino acid content of grass pea seeds, obtained results were in compliance with the findings of Fikre *et al.* (2008) in study of grass pea seeds belonging to different regions and in grass pea seeds grown in Italy (Tamburino *et al.* 2012).

Association analysis of amino acid: Correlation studies of amino acid content of grass pea genotypes (Table 4) indicate significant positive associations (0.01) among the amino acids. Only the positive association between tyrosine and methionine was determined at a moderate significance level of 0.05.

CONCLUSION

Agricultural strategies,which are set for increasing substances such as micronutrients minerals, amino acids etc in foods to meet daily dietary requirements are being assessed as sustainable and long-term solutions (Karaköy *et al.*, 2012). There are different approaches such as producing micronutrient-enriched cultivars (biofortification), which are considered to bepromising and costeffective method or as developing new cultivars from natural variants that have favorable traits. Soybean which is used by the animal feed industry to meet the animal requirement of crude protein and essential amino acids is quite expensive as compared to grass pea. Grass pea has enough potential to meet these requirements. as a source of protein to lower the cost of animal feeds. The production in grass pea should be increased as it can be well adapted to a wide range of environmental conditions (Hanbury *et al.*, 2000). To improve grass pea varieties, breeding studies aim to increase vitamins and amino acid concentrations and knowledge on the variability of the traits among available germplasm is the need of hour.

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