Indian J. Genet., 79(2): 438-443 (2019)
DOI: 10.31742/IJGPB.79.2.7

Variation for seed protein and ODAP content in grass pea (*Lathyrus sativus* L.) germplasm collections

Karthika Rajendran, Ashutosh Sarker[1]*, Murari Singh, Ali M. Abd El-Moneim and Hani Nakkoul

Biodiversity and Integrated Gene Management Program, International Center for Agricultural Research in the Dry Areas (ICARDA), Aleppo, Syria

(Received: February 2018; Revised: December 2018; Accepted: January 2019)

Abstract

Development of grass pea (*Lathyrus sativus* L.) varieties with low seed anti-nutritional factor, β-N-Oxalyl-L-α, β-diaminopropionic acid (ODAP) content but with high seed protein content would be beneficial for human consumption. In this study, a total of 702 germplasm accessions, from the International Center for Agricultural Research in the Dry Areas (ICARDA), Syria, were grouped into seven sets of trials based on their origin, and evaluated for seed ODAP content and seed protein content at two experimental sites, Tel Hadya and Breda over eight years. Significant genotypic differences were found for both the traits in all the germplasm sets except for seed ODAP content in BANG1 and ETH1. The effects associated with genotype × year within locations interactions were larger than genotypic effects for seed protein content in all the germplasm sets except ICARDA and for seed ODAP content in all the sets except ICARDA and PAK. The highest range was found for seed protein content (28.82-30.72%) in ICARDA germplasm set and for seed ODAP content in ETH2 (0.32-0.47%) and PAK (0.38-0.53%) germplasm sets. On the basis of best linear unbiased predictor values, new promising sources with low seed ODAP content such as ILG468, ILG1934, ILG1950 and ILG1951 and for high protein content such as ILG311, ILG670, ILG688, ILG691 and ILG708 and were identified for future grass pea breeding.

Key words: Grass pea, ODAP, protein, variation, BLUP

Introduction

Grass pea (*Lathyrus sativus* L.) is a cool season legume crop. It is a diploid (2n=14) (Talukdar 2009). It has high amount of protein content in seeds (27%) (Hanbury et al. 2000). It is a hardy legume crop; it can tolerate drought, moderate level of soil salinity and water-logging. It is currently cultivated in many countries of Asia (India, Pakistan, Bangladesh, Nepal and China), Middle East (Iran, Iraq, Afghanistan, Syria and Lebanon), Africa (Ethiopia, Ghana, Sudan, Niger, Ivory Coast and Mauritania) and Southern Europe (France, Spain and Italy) for both human consumption and animal feed (Campbell 1997; Vaz Patto et al. 2006; Piergiovanni et al. 2011; Dixit et al. 2016). Nevertheless, presence of an anti-nutritional factor, a neurotoxic non-protein amino acid, β-N-Oxalyl-L-α, β diaminopropionic acid (ODAP) in grass pea seeds causes 'Neuro-Lathyrism", an irreversible paralysis of lower limbs in humans when it is continuously consumed over a period of 3-4 months (Lambien et al. 1996). However, the development of new grass pea cultivars with low ODAP content but with high protein content in its seeds would make the crop much more acceptable for human consumption.

Crop genetic resources contribute source of variability for multiple traits for the development of new cultivars over time. The International Center for Agricultural Research in the Dry Areas (ICARDA) has the mandate for improving the productivity of dry-land agriculture in the West Asia and Sub-Saharan Africa (SSA) where grass pea cultivation is prevailing (FAOSTAT 2017). ICARDA germplasm holds a collection of 1883 *Lathyrus* spp. accessions from different parts of the world (Abd El Moneim et al. 2001; Rajendran et al. 2018). However, the germplasm collection of ICARDA has not been characterized for seed ODAP content and seed protein content and it may contain untapped genetic variability for future breeding progress. Early studies on genetic

*Corresponding author's e-mail: a.sarker@cgiar.org
[1]Present address: Regional Coordinator and Food Legume Breeder, South Asia and China Regional Program, International Center for Agricultural Research in the Dry Areas (ICARDA), 2nd Floor, Office Block-C, NASC Complex, DPS Marg, New Delhi 110 012, India
Published by the Indian Society of Genetics & Plant Breeding, A-Block, F2, First Floor, NASC Complex, IARI P.O., Pusa Campus, New Delhi 110 012; Online management by www.isgpb.org; indianjournals.com

characterization of grass pea revealed the genetic diversity for these two quality traits (Bisignano et al. 2002; Granati et al. 2003; Tamburino et al. 2012). With this background, the present work was carried out to: 1) evaluate grass pea germplasm from Bangladesh, Pakistan, Nepal, Syria and Ethiopia at different locations over several years for seed protein and ODAP content, 2) assess the genetic variability and heritability for seed protein and ODAP contents, and 3) identify promising sources of genetic variation for future breeding.

Materials and methods

A total of 702 grass pea germplasm accessions from the seed bank of International Center for Agricultural Research in the Dry Areas (ICARDA), Aleppo, Syria were grouped into seven sets of trials based on their origin of collection, such as, "BANG1" (100 accessions) and "BANG2" (225 accessions), for the germplasm from Bangladesh; "ETH1" (100 accessions) and "ETH2" (64 accessions), for the germplasm from Ethiopia; "NEP" (49 accessions), for the germplasm from Nepal and "PAK" (64 accessions), for the germplasm from Pakistan. As ICARDA had its head quarters at Syria, the germplasm originated in Syria was named as "ICARDA" (100 accessions). Subsequently, all seven germplasm sets were evaluated for seed ODAP content and seed protein content in seven sets of trials at two experimental station of ICARDA namely, Tel Hadya and Breda over eight years (1998/99-2005/06). It is important to note that the accessions from a given set remained as same over years and locations. In each year, the crop growing season included the period from September to the following May. In each and individual trial, accessions were evaluated in simple lattice designs (Cochran and Cox 1957). The local check, denoted by ILG347 was employed in all trials except in ETH2 and ICARDA; where ILG587 was used as a local check in ETH2 and ILG431 was used as a local check in ICARDA. All sets were evaluated in both locations except BANG2 and ETH2 which were evaluated only in Tel Hadya due to limited quantities of seed. During the crop growing period optimum level of fertilizers, weed management practices, pesticide sprays were done at necessary conditions. At maturity stage, seeds were harvested, cleaned and dried at room temperature. AOAC official method (Kjeldahl method) was used to estimate seed protein concentration (n=6.25) (AOAC, 1970) and seed ODAP content was determined calorimetrically using the *o*-phtalaldehyde method of Rao (1978) which was further modified by Briggs et al. (1983).

Initially, the statistical analysis of traits data from individual trials was carried out while assuming a model with replication effects, block effects within replications and genotypes effects as "random". The test of equality of effects was carried out by fitting a mixed model using REML (Restricted Maximum Likelihood) approach. Further again, to examine how the response of genotypes in a given trial varied with year and location, a combined analysis of data over the location and year was carried out. Components of variance of genotype effects, genotype x location (G×L) interaction and genotype x year within locations (G×YwL) and pooled error were estimated by fitting a mixed effects model using REML. The model also assumed fixed effects for location, random effects of replications and blocks within replications within location – year combinations (i.e. the environments). In order to keep the estimates of variance components in a meaningful range, an option in the REML was set to produce only non-negative estimates of variance components. From the mixed models for the combined analysis the best linear unbiased predictor (BLUP) values were obtained for every individual trait. The broad-sense heritability (h^2_b) was estimated using the following the formula:

$$h^2_b = \sigma^2_g / (\sigma^2_g + \sigma^2_{gl}/l + \sigma^2_{gy(l)}/(yl) + \sigma^2_e/(ylr))$$

where, σ^2_g = genetic variance, σ^2_{gl} = genotype by location interaction variance component, $\sigma^2_{gy(l)}$ = genotype x year interaction within location variance component, σ^2_e = pooled error variance, l = number of locations, y=number of years within a given location and r= number of replications. The heritability estimates were categorized as low (<0.30), moderate (0.31-0.60) and high (>0.60) (Robinson et al. 1949). All statistical analyses were carried out under GenStat software for windows, Release 18, *VSNL International* Ltd, Hemel, Hempstead, Hertfordshire, UK (VSN International. 2015).

Results and discussion

The REML for individual trials found significant variances due to genotypes (Data not presented). Further, the REML analysis of the combined data showed genotypic variability and interaction effect across locations (GxL) and year within locations (GxYwL) to variable degree and their significance is given in terms of P-values in Table 1. Significant genotypic (σ^2_g) differences were found for seed protein content and seed ODAP content in entire germplasm sets except for seed ODAP content in BANG1 and ETH1. Incidence of significant genetic variation for

seed protein content and seed ODAP content indicates great potential of these germplasm in future breeding programs (Table 1).

Significant G×L interaction effect were identified for seed protein content and seed ODAP content only in BANG1. But, significant G×YwL interaction effects were revealed for seed protein content in entire germplasm sets and for seed ODAP content in all germplasm sets except in BANG1 and PAK. Mostly, the effects associated with the G×YwL interaction were larger than the genotypic effects for seed protein content in all germplasm sets except ICARDA and for seed ODAP content in all germplasm sets except ICARDA and PAK (Table 1). Moreover, the same trait has also obtained a low, moderate and high level of heritability and confirmed again the major influence of climatic conditions of the growing seasons in Breda and Tel Hadya across the years (1998/99-2005/06) (Table 1). When compared the two experimental sites, Tel Hadya (360 mm) recorded higher average total growing season (September to June) rainfall than Breda (291mm) across the eight years. The range of total growing season rainfall (from September to the following May) was 261-483 mm at Tel Hadya and 198-386 mm at Breda. These variation in growing season rainfall from one year to the next even within the same location would have affected the crop response and hence the ODAP concentration in grass pea seeds. As has been stated in Piergiovanni and Damascelli (2011) the climatic factors such as rainfall and average temperature during the crop growing season largely affected the amount of anti-

Table 1. Estimates of components of variance and their standard errors, broad sense heritability of seed protein and ODAP content

Trials	Traits	σ^2_g		σ^2_{gl}		$\sigma^2_{gy(l)}$		σ^2_e		h^2_b	
		Estimate	S.E	Estimate	S.E	Estimate	S.E	Estimate	S.E	Estimate	S.E
BANG 1	Protein content	0	Bound[a]	0.0353***	0.0087	0.0509***	0.0156	0.383	0.0186	5.854E-07	6.5685E-08
	ODAP content	0.000081	0.000089	0.000182**	0.000101	0.000166	0.000128	0.00343	0.000167	0.23	0.2176
BANG 2	Protein content	0.051***	0.012	NA[b]		NA[b]		0.427	0.0213	0.44	0.06382
	ODAP content	0.00031***	0.000087	NA[b]		NA[b]		0.00387	0.000191	0.37	0.07094
ETH 1	Protein content	0.0112**	0.0041	0	Bound[a]	0.0524***	0.0121	0.356	0.0144	0.40	0.09145
	ODAP content	0.000017	0.000031	0.000064	0.000041	0.000283***	0.000078	0.00237	0.000095	0.11	0.1887
ETH 2	Protein content	0.1708***	0.0383	NA[b]		NA[b]		0.482	0.0349	0.82	0.03657
	ODAP content	0.000711***	0.000178	NA[b]		NA[b]		0.00242	0.000177	0.74	0.05271
ICARDA	Protein content	0.3576***	0.054	0.0034	0.0047	0.0319**	0.0108	0.34	0.0134	0.96	0.00849
	ODAP content	0.000224***	0.000044	0	Bound[a]	0.000143*	0.000059	0.00189	0.000076	0.76	0.03721
NEP	Protein content	0.0254*	0.0116	0.0085	0.0108	0.0818***	0.0195	0.339	0.0209	0.52	0.1464
	ODAP content	0.000548***	0.000156	0	Bound[a]	0.000762***	0.000189	0.00359	0.000222	0.74	0.05761
PAK	Protein content	0.0282***	0.0087	0	Bound[a]	0.0315*	0.0175	0.433	0.0229	0.60	0.07716
	ODAP content	0.000711***	0.000159	0	Bound[a]	0.000518	0.000319	0.0031	0.000165	0.82	0.0348

BANG1/BANG2, ETH1/ETH2, ICARDA, NEP, PAK are trial codes and had accessions originated from Bangladesh, Ethiopia, Syria, Nepal and Pakistan respectively.
σ^2_g, σ^2_{gl}, $\sigma^2_{gy(l)}$, σ^2_e are variance components due to genotype (G), genotype ×location (G×L) interaction, genotype × year within location (G×YwL) interaction, and plot-errors. *, ** and **** significant at 5%, 1% and 0.1% respectively.
S.E. = Standard Error.
a = Variance components estimates kept at the boundary when restricted to positive range, b = Data not available

nutritional factors accumulation in grass pea seeds.

The mean, range and standard deviation of various germplasm sets are presented in Table 2. The mean seed protein content in Bangladesh (28.95%) and Ethiopian (29.18%) germplasm are observed higher than the previous results by Kaul et al. (1982) and Urga et al. (1995). On the other hand, the germplasm from Syria (ICARDA) recorded lower the mean seed protein content (29.43%) than the earlier findings by Aletor et al. (1994). Among all germplasm sets, the highest range was found for seed protein content (28.82-30.72%) in ICARDA material but with low level of variation (CV = 2.0% in the predicted mean values). The problem of low level of variation (CV=2.58%) for seed protein content in Turkish grass pea landraces was stated by Basaran et al. (2013) in recent times.

In contrary to seed protein content, a relatively higher level of variation for seed ODAP content was revealed across all germplasm sets (Table 2). Among all sets, the highest range was found in ETH2 (0.32-0.47%) and in PAK (0.38-0.53%). As shown by Kumar et al. (2011), our findings also agreed that the germplasm material from Ethiopia possess the highest variability (CV = 7.3%) for seed ODAP content than the germplasm from Bangladesh, Nepal and Pakistan (Table 2). The correlations between protein and ODAP content, based on their BLUPs for the accessions, varied with the trials: 0.57 (P<0.001, BANG1), 0.53 (P<0.001, ICARDA), 0.34 (P<0.001, BANG2), -0.30 (P<0.05, PAK), while the estimates, 0.26 (NEP), -0.19 (ETH2) and 0.11 (ETH1) were non-significant at 5% level. The presence of non-constant correlation indicates the effect of natural selection that changes the genetic constitution of the accessions and their trait expression within each germplasm group which was obtained from different countries.

The five most desirable accessions selected based on BLUP values for seed protein content and seed ODAP content are presented in Table 3 and 4 respectively. All five selected accessions for seed protein content in BANG1, ETH2, ICARDA, NEP and PAK recorded significantly more protein than the local check. Particularly, accessions namely ILG311, ILG670, ILG688, ILG691 and ILG708 in ICARDA had the protein content of more than 30% (Table 3). Similarly, the accession ILG468 in ETH1, all five selected accessions in ETH2 and ICARDA, accessions ILG1934, ILG1950 and ILG1951 in NEP and all five selected accessions in PAK recorded significantly lower ODAP content than the local check (Table 4). The promising accessions identified in the present study could be used as parents in grass pea breeding programmes.

As these two traits are largely influenced by G×YwL interaction, it is also important to note that the success of breeding safe cultivars of grass pea is likely to be depending on the development of genotypes that express low levels of ODAP and high protein content in the given environments. Furthermore, the less genetic variability for seed protein content and the lack of genotypes with low

Table 2. Summary of trial means, standard deviation and the range of the different quantitative traits based on the best linear unbiased predictor (BLUP) values of the accessions from the seven sets of trials

Trial code	PMean		Standard deviation		CV (%)		Minimum		Maximum	
	Protein content	ODAP content	Protein content	ODAP content	Protein content	ODAP content	Protein content	ODAP content	Protein content	ODAP content
BANG1	28.9	0.51	0.095	0.01	0.33	1.96	28.7	0.49	29.2	0.53
BANG2	29.0	0.54	0.17	0.01	0.59	1.85	28.2	0.51	29.4	0.58
ETH1	29.1	0.44	0.09	0.005	0.31	1.14	28.9	0.42	29.3	0.45
ETH2	29.26	0.41	0.38	0.03	1.30	7.32	27.86	0.32	29.86	0.47
ICARDA	29.43	0.49	0.59	0.013	2.00	2.65	28.82	0.45	30.72	0.54
NEP	28.96	0.49	0.155	0.02	0.54	4.08	28.16	0.43	29.18	0.52
PAK	29.04	0.47	0.14	0.025	0.48	5.32	28.72	0.38	29.65	0.53

PMean = Predicted Mean
CV = Coefficient of Variation between the BLUP estimates (Standard deviation/PMean)

Table 3. For seed protein content, the list of five desirable accessions (the highest five in descending order) from the *Lathyrus sativus* core collections with their predicted means combined across the 16 environments

BANG1		BANG2		ETH1		ETH2		ICARDA		NEP		PAK	
ILG No.	PMean	ILG No.	PMean	ILG No.	PMean	ILG No.	PMean	ILG No.	PMean	ILG No.	PMean	ILG No.	PMean
1837	29.2	2936	29.4	438	29.3	ETH9	29.9	691	30.7	1953	29.2	11	29.7
1846	29.1	2814	29.3	483	29.3	PAK100	29.8	688	30.6	1897	29.2	1761	29.2
1842	29.1	2881	29.3	460	29.3	ETH31	29.8	311	30.6	1910	29.1	1760	29.2
1828	29.1	2790	29.3	431	29.3	ETH6	29.8	708	30.6	1923	29.1	1759	29.2
1825	29.1	2895	29.3	453	29.3	ETH12	29.7	670	30.6	1901	29.1	1775	29.2
L.C	28.7	L.C	29.1	L.C	29.1	L.C	29.3	L.C	30.1	L.C	28.9	L.C	28.8
S.E±	0.22	S.E±	0.36	S.E±	0.20	S.E±	0.30	S.E±	0.40	S.E±	0.18	S.E±	0.15

PMean = Predicted Mean
L.C = Local Check. The local check ILG347 was employed in all trials except in ETH2 and ICARDA; where ILG587 was used as a local check in ETH2 and ILG431 was used as a local check in ICARDA.
S.E = Standard Error

Table 4. For seed ODAP content, the list of five desirable accessions (the lowest five in ascending order) from the *Lathyrus sativus* core collections with their predicted means combined across the 16 environments

BANG1		BANG2		ETH1		ETH2		ICARDA		NEP		PAK	
ILG No.	PMean	ILG No.	PMean	ILG No.	PMean	ILG No.	PMean	ILG No.	PMean	ILG No.	PMean	ILG No.	PMean
2090	0.48	3036	0.51	468	0.42	PAK100	0.32	736	0.45	1950	0.43	11	0.38
2191	0.49	2427	0.51	445	0.43	PAK208	0.33	735	0.47	1951	0.44	954	0.43
2206	0.50	2537	0.51	441	0.43	PAK209	0.33	722	0.47	1934	0.44	1804	0.43
2196	0.50	3027	0.52	386	0.43	ETH34	0.37	705	0.47	1935	0.46	1756	0.43
2082	0.50	225	0.52	403	0.44	ETH42	0.39	654	0.47	1989	0.46	958	0.44
L.C	0.50	L.C	0.54	L.C	0.45	L.C	0.47	L.C	0.52	L.C	0.47	L.C	0.51
S.E±	0.06	S.E±	0.08	S.E±	0.02	S.E±	0.05	S.E±	0.03	S.E±	0.02	S.E±	0.03

PMean = Predicted Mean
L.C = Local Check. The local check ILG347 was employed in all trials except in ETH2 and ICARDA; where ILG587 was used as a local check in ETH2 and ILG431 was used as a local check in ICARDA
S.E = Standard Error

level of seed ODAP content in the present study, also suggest to look for new possibilities of broadening the genetic base either through the introgression of alleles from other wild gene pool or mutation breeding techniques.

Authors' contribution

Conceptualization of research (AS, AMA); Designing of the experiments (AS, MS); Contribution of experimental materials (AMA, HN); Execution of field/ lab experiments and data collection (HN, AMA); Analysis of data and interpretation (MS, KR, AS); Preparation of manuscript (KR, MS, AS).

Declaration

The authors declare no conflict of interest.

Acknowledgements

We express our sincere thanks to Dr. Ahmed Amri and Dr. Fouad Maalouf for their valuable suggestions on the manuscript. We express our sincere gratitude to the reviewers for their advice on the manuscript. We would like to thank the gene bank of ICARDA for providing the seed materials and support. The CGIAR Research Program on Grain Legumes is gratefully acknowledged for providing financial and research support.

References

AOAC. 1970. Official Methods of Analysis of the Association Official Analytical Chemists. 11th ed. Methods. Association, Washington, DC.

Abd El Moneim A. M., Van Dorrestein B., Baum M. and Bejiga J. R. 2001. Role of ICARDA in improving the nutritional quality and yield potential of grasspea (*Lathyrus sativus* L.), for subsistence farmers in dry areas. Lathyrus Lathyrism Newslett., **4**: 55-58.

Aletor V. A., Abd El-Moneim A. and Goodchild A. V. 1994. Evaluation of the seeds of selected lines of three *Lathyrus* spp. for b-N-oxalylamino-L-alanine (BOAA), tannins, trypsin inhibitor activity and certain *in vitro* characteristics. J. Sci. Food Agric., **65**(2): 143-151.

Basaran U., Zeki A. C. A. R., Karacan M. and Onar A. N. 2013. Variation and correlation of morpho-agronomic traits and biochemical Contents (Protein and B-ODAP) in Turkish grass pea (*Lathyrus sativus* L.) landraces. Turk. J. Field Crops., **18**(2): 166-173.

Briggs C. J., Parreno N. and Campbell C. G. 1983. Phytochemical Assessment of Lathyrus species for the neurotoxic agent, β-N-oxalyl-L-α-β-Diaminopropionic Acid. Planta Med., **47**: 188-190.

Campbell C. G. 1997. Grasspea (*Lathyrus sativus* L.) promoting the conservation and use of underutilized and neglected crops. Rome, Italy: Institute of Plant Genetics and Crop Plant Research, Gatersleben/ International Plant Genetic Resources Institute.

Cochran W. G. and Cox G. M. 1957. Experimental Designs, 2nd ed. Wiley, New York.

Dixit G. P., Parihar A. K., Bohra A. and Singh N. P. 2016. Achievements and prospects of grass pea (*Lathyrus sativus* L.) improvement for sustainable food production. Crop J., **4**(5): 407-416.

FAOSTAT. 2017. Available at: http://www.fao.org/faostat/en/#data/QC (accessed 05.01.08).

Granati E., Bisignano V., Chiaretti D., Crinò P. and Polignano G. B. 2003. Characterisation of Italian and exotic *Lathyrus* germplasm for quality traits. Genet. Resour. Crop Evol., **50**: 273-280.

Hanbury C. D., White C. L., Mullan B. P. and Siddique K. H. M. 2000. A review of the potential of *Lathyrus sativus* L. and *L. cicera* L. grain for use as animal feed. Anim. Feed Sci. Technol., **87**(1): 1-27.

Kaul A. K., Islam M. Q. and Begum K. 1982. Variability for various agronomic characters and neurotoxin content in some cultivars of khesari (*Lathyrus sativus* L.) in Bangladesh. Bangl. J. Bot., **11**: 158-167.

Kumar S., Bejiga G., Ahmed S., Nakkoul H. and Sarker A. 2011. Genetic improvement of grass pea for low neurotoxin (β-ODAP) content. Food Chem. Toxicol., **49**: 589-600.

Lambein F., Khan J. K., Kuo Y. H., Campbell C. G. and Briggs C. J. 1993. Toxins in the seedlings of some varieties of grass pea (*Lathyrus sativus*). Nat. Toxins., **1**: 246-249.

Piergiovanni A. R. and Damascelli A. 2011. L-homoarginine accumulation in grass pea (*Lathyrus sativus* L.) dry seeds. A preliminary survey. Food Nutr. Sci., **2**(03): 207.

Piergiovanni A. R., Lupo F. and Zaccardelli M. 2011. Environmental effect on yield, composition and technological seed traits of some Italian ecotypes of grass pea (*Lathyrus sativus* L.). J. Sci. Food Agric., **91**(1): 122-129.

Rao S. L. N. 1978. A sensitive and specific calorimetric method for the determination of α, β-diamino-propionic acid and the *Lathyrus sativus* neurotoxin. Analytic Biochem., **96**: 386-395.

Rajendran K., Sarker A., Singh M., Abd El-Moneim A. M. and Nakkoul H. 2018.Characterization of grass pea (*Lathyrus sativus* L.) germplasm collections for the beneficial agronomic traits. Indian J. Genet., **78**(3): 357-363. doi: org/10.31742/IJGPB.78.3.8.

Robinson H. F., Comstock R. E. and Harvey P. H. 1949. Estimation of heritability and the degree of dominance in corn. Agron. J., **41**: 353-359.

Talukdar D. 2009. Reciprocal translocations in grass pea (*Lathyrus sativus* L.): Pattern of transmission, detection of multiple interchanges and their independence. J. Hered., **101**: 169-176.

Tamburino R., Guida V., Pacifico S., Rocco M., Zarelli A., Parente A. and Di Maro A. 2012. Nutritional values and radical scavenging capacities of grass pea (*Lathyrus sativus* L.) seeds in Valle Agricola district, Italy. Aust. J. Crop Sci., **6**(1): 149.

Urga K., Fufa H., Biratu E. and Husain A. 2005. Evaluation of *Lathyrus sativus* cultivated in Ethiopia for proximate composition, minerals, β-ODAP and anti-Nutritional Components. Afr. J. Food Agric. Nutr. Dev., **5**(1): 1-15.

Vaz Patto M. C., Skiba B., Pang E. C. K., Ochatt S. J., Lambein F. and Rubiales D. 2006. Lathyrus improvement for resistance against biotic and abiotic stresses: From classical breeding to marker assisted selection. Euphytica, **147**: 133-147.

VSN International. 2015. The Guide to the Genstat Command Language (Release 18), Part 2 Statistics. VSN International, Hemel Hempstead, UK.

Xu Q., Liu F., Chen P., Jez J. M. and Krishnan H. B. 2017. β-*N*-Oxalyl-l-α, β-diaminopropionic Acid (β-ODAP) Content in *Lathyrus sativus*: The Integration of Nitrogen and Sulfur Metabolism through β-Cyanoalanine Synthase. Int. J. Mol. Sci., **18**(3): 526.