

Fatty Acids and Sugars in Lablab Seed Produced in Virginia (A Non-traditional Location)[†]

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Abstract

Lablab [*Lablab purpureus* (L.) Sweet], a relatively unknown crop in the United States of America, is understood to be adapted to Southern USA. Even though, previous studies conducted in Virginia have indicated that Lablab can be produced in Virginia as a forage crop, composition of lablab seed produced in Virginia is unknown. To alleviate this limitation, seeds of seventeen lablab lines from a replicated field study, that was conducted for two years, were analyzed for concentrations of fatty acids and sugars. Results indicated that genotypes had mostly significant effects on concentrations of fatty acids and sugars. Prominent fatty acids in lablab seeds, grown in Virginia (USA), were linoleic (53.5%), palmitic (15.8%), and linolenic (14.1%). Mean saturated and total unsaturated fatty acids in lablab seeds were 22.2 and 77.6%, respectively. Mean concentrations of sucrose, fructose, and glucose concentrations in lablab seed were 1.45, 0.42, and 0.78 g per 100 g meal. Mean concentrations of total non-nutritive sugars (Raffinose+Stachyose+Verbascose) in lablab seed were 4.96 g per 100 g meal. Correlations between several nutritional quality traits in lablab seed were observed to be significant. A comparison of nutritional quality of lablab seed with literature values of black bean, navy bean, kidney bean, pinto bean, and pea indicated that lablab has potential as a new food legume for United States of America.

Keywords: *Lablab purpureus*, fatty acids, sugars, raffinose, stachyose, verbascose, seed composition

1. Introduction

Lablab [*Lablab purpureus* (L.) Sweet] is a relatively unknown crop in the United States of America. From a historical perspective, seeds of lablab were planted in 1819 in the Botanical Garden of Sydney, Australia which eventually led to the first improved cultivar, “Rongai” released in Australia in 1962. “Rongai” seed was imported to the US in late 1960s and marketed as supplemental forage for white-tailed deer. Lablab cultivar “Rio Verde” was developed by Texas A&M AgriLife Research and Extension Center in 2006. This was the first lablab cultivar developed in the US for tolerance to defoliation, forage and seed production (Houck, 2013). Lablab has been a novelty garden plant in the U.S. for generations. This vigorous twining vine is characterized by larger alternate purple-green leaves and purple petioles. The vines produce hundreds of spikes of lavender flowers in late summer followed by long-lasting deep lavender-purple pods. It is primarily an ornamental annual vine in the US (Anderson et al., 1996). During 1970s, lablab was labelled as “neglected” but worthy of more intensive studies and further developments (NAS, 1979). Maass et al., (2010) indicated that more than 3000 accessions of lablab have been collected worldwide. Lablab is a native of Asia and Africa (Pengelly & Maas, 2001). As per the information available at University of Agricultural Sciences (Bangalore, India), a site well known as source of information for this crop, lablab is usually known as Dolichos bean, Hyacinth bean, Bonavist bean, Seim bean, Lablab bean, Egyptian kidney bean, Indian bean, Common bean, Field bean, Pendal bean, Pole bean, Waby bean (<http://www.lablablab.org>). It is one of the most ancient crops among cultivated plants, found in archaeo-botanical finds in India prior to 1500 BC

(Fuller, 2003) and in Egyptian Nubia from the 4th century AD (Clapham & Rowley-Conwy, 2007). It is a bushy, semi-erect, perennial herb, showing no tendency to climb. It is one of the major sources of protein in the human diets in southern states of India. The consumer preference varies with pod size, shape, color and aroma (pod fragrance).

Lablab has been noted for decades as being one of the most agro-morphologically diverse (Piper and Morse 1915, Pengelly and Maass 2001; Islam 2008) and versatile tropical legume species through its roles as pulse (also used as 'dhal'), vegetable (green bean, pod, leaf), forage/green manure, herbal medicine, and even ornamental (NRC, 2006). Sheahan (2012) observed that lablab will produce seed in Southern United States (Florida, Georgia, and Texas) and listed three lablab cultivars for USA: two from Australia (Rongai and Highworth), and one from USA ('Rio Verde'—Smith et al., 2008). Bio-functional properties of lablab for use as pharmaceutical or nutraceutical have also been reviewed (Morris, 2009). In Indonesia, seeds serve as raw materials for 'tempeh', a traditional fermented food typically made from soybeans (Subagio & Morita, 2008). However, nutritional quality of lablab seed produced in Virginia and other non-traditional environments is unknown. The overall goal of research with lablab was to expand the menu of alternative crops for erstwhile tobacco farmers in a non-traditional area. Specifically, objective of the current research with lablab was to characterize nutritional quality of lablab seed produced in Virginia to its' potential use as food and feed relative to other food legumes.

2. Materials and Methods

2.1 Plant Material

Sixteen lablab accessions (received from Plant Genetic Conservation Unit, Agricultural Research Service, United States Department of Agriculture, Griffin, GA 30223, USA) and one cultivar "Rio Verde" (Received from Texas A&M University, Overton, Texas, 75684, USA) constituted the plant materials for this study (Table 1).

Table 1. Mean saturated fatty acids* in seeds of seventeen lablab lines grown at Petersburg, Virginia (USA) during 2011 and 2012

Genotype	Source	Oil%	14:0	16:0	18:0	20:0	22:0	24:0	TS**
PI 164772	India	1.01	0.15	16.3	3.42	0.72	0.94	2.17	23.7
PI 183451	India	1.13	0.09	15.5	3.32	0.69	0.94	1.50	22.1
PI 284802	China	0.84	0.15	16.5	3.59	0.77	0.94	0.87	22.9
PI 288466	India	1.05	0.10	15.3	3.29	0.71	0.90	1.32	21.6
PI 288467	India	0.85	0.13	15.7	3.30	0.69	0.96	1.53	22.4
PI 388003	Australia	1.01	0.13	15.1	3.18	0.66	0.90	1.29	21.3
PI 388012	Australia	1.03	0.11	15.2	3.01	0.63	0.84	1.73	21.2
PI 388013	Australia	1.08	0.09	14.7	3.05	0.63	0.81	1.76	21.0
PI 388017	Australia	0.82	0.12	15.6	3.37	0.70	0.93	1.40	22.1
PI 388018	Australia	0.64	0.17	15.8	3.36	0.70	0.93	1.48	22.5
PI 542609	India	0.54	0.15	15.6	2.99	0.70	0.92	1.54	22.9
PI 593055	USA	0.71	0.13	17.7	3.48	0.74	1.16	1.75	24.9
PI 639277	China	0.66	0.13	16.2	2.92	0.60	0.91	1.59	22.3
PI 639279	China	0.94	0.09	15.4	2.79	0.60	0.83	1.39	21.1
PI 639280	China	0.74	0.14	15.7	2.94	0.65	0.74	1.42	21.6
PI 653615	USA	0.87	0.09	15.8	3.22	0.62	0.78	1.32	21.9
Rio Verde	USA	0.77	0.17	16.1	3.33	0.69	0.86	1.34	22.5
Mean		0.87	0.13	15.8	3.22	0.68	0.90	1.49	22.2
LSD (5%)		0.27	0.06	1.3	0.36	0.10	0.17	0.81	1.93

Note. * Means over two years and two replications per year. ** Total saturated fatty acids (14:0+16:0+18:0+20:0+22:0+24:0).

2.2 Production and Field Data Recording

The seventeen lablab entries were grown during 2011 and 2012 in the field (Abel sandy loam-Fine Loamy mixed thermic Aquatic Hapridult soil) at Randolph Farm of Virginia State University in Ettrick, Virginia. The planting dates during both years were May 23 and May 15, respectively using two row plots with rows spaced 1.2 m apart. The field design was a RCBD with two replications. Each plot consisted of four rows with approximately 50 seeds

planted in 2.5 m row length—two rows were harvested for forage characterization of forage yield and quality whereas two rows were harvested to record seed yield. These plots did not receive any herbicide, insecticide, or fertilizer treatments. The plots were kept weed free manually during the early growth. After about 30 days, the lablab canopy spread enough to crowd-out the weeds.

Two rows (each 3 m long) from each plot were harvested manually and threshed after plants were effectively killed by a killing frost during late November during each year.

2.3 Determination of Oil Content and Fatty Acid Spectra in Seeds

The oil concentration was determined in the Common Laboratory of Agricultural Research Station of Virginia State University. The oil was extracted from ground lablab seeds (5 g) three times at room temperature by homogenization for 2 min in 20 mL hexane/isopropanol (3:2, v/v) with a Biospec Model 985-370 Tissue Homogenizer (Biospec Products, Inc. Racine, WI, USA) and centrifuged at 4000 g for 5 min, as described by Hamama et al. (2003). The three extractions were combined and the hexane-lipid layer was separated from the combined extract after shaking with 10 mL of 1% solution of equal amounts of CaCl_2 and NaCl in 50% methanol. The hexane lipid layer was removed by aspiration and dried over anhydrous Na_2SO_4 . The oil percentage (g/100 g dry basis) was determined gravimetrically after drying under vacuum at 40 °C and stored under nitrogen at 10 °C until analysis.

2.4 Determination of Sugar Content in Seeds

Sugars were extracted from ground sample (1 g) and analyzed by HPLC following the methods optimized by Johansen et al. (1996). Sugars in the extracts were identified by comparing their retention times with standard sugars. For quantification, trehalose was used as internal standard and the sugar concentration was expressed as g/100 g meal (Bhardwaj & Hamama, 2016).

2.5 Data Analysis

All data were analyzed using version 9.1 of SAS (SAS Institute, Inc., 2014) using ANOVA with 5% level of significance.

3. Results and Discussion

Results indicated significant effects of genotypes but not of production years on concentrations of all fatty acids and sugars (Tables 1, 2, and 3). Predominant fatty acids in oil of lablab seeds was C18:2 (Linoleic acid: 53.5%), followed by C16:0 (Palmitic acid: 15.8%) and C18:3 (Linolenic acid: 14.1%). Total saturated fatty acids in lablab seed varied from 21.0 to 24.9% with a mean of 22.2%. Total unsaturated fatty acids in lablab seed varied from 74.8 to 78.9% with a mean of 77.6%. Concentration of Linolenic acid (14.1%) indicates that lablab seed's oil is healthy for human consumption but this oil will be unstable due to susceptibility to enhance potential for reaction with oxygen (Hamama et al., 2003). Lablab seeds also contained small concentrations of C22:0 (Erucic acid) which varied from 0.08 to 0.28%. Based on our previous results with canola oil, this concentration of Erucic acid should not be of concern (Bhardwaj & Hamama, 2016).

Table 2. Mean unsaturated fatty acids* in seeds of seventeen lablab lines grown at Petersburg, Virginia (USA) during 2011 and 2012

Genotype	Source	Oil%	18:1	18:2	18:3	20:1	22:1	TUS**
PI 164772	India	1.01	10.6	51.6	13.4	0.41	0.16	76.2
PI 183451	India	1.13	8.5	55.5	13.3	0.36	0.18	77.8
PI 284802	China	0.84	10.0	51.8	14.5	0.39	0.08	76.7
PI 288466	India	1.05	8.0	55.4	14.2	0.34	0.14	78.2
PI 288467	India	0.85	8.0	54.3	14.5	0.38	0.19	77.3
PI 388003	Australia	1.01	8.3	55.1	14.7	0.34	0.11	78.6
PI 388012	Australia	1.03	9.9	53.7	14.5	0.39	0.13	78.7
PI 388013	Australia	1.08	9.7	54.5	14.2	0.37	0.16	78.9
PI 388017	Australia	0.82	9.9	53.1	14.1	0.41	0.15	77.7
PI 388018	Australia	0.64	9.4	52.8	14.6	0.41	0.16	77.4
PI 542609	India	0.54	9.7	52.4	15.1	0.42	0.20	77.9
PI 593055	USA	0.71	10.1	51.8	12.1	0.41	0.28	74.8
PI 639277	China	0.66	9.9	52.9	13.9	0.39	0.12	77.3
PI 639279	China	0.94	9.4	55.1	13.8	0.37	0.15	78.8
PI 639280	China	0.74	8.9	54.2	14.6	0.39	0.15	78.2
PI 653615	USA	0.87	9.5	54.3	13.6	0.35	0.17	78.0
Rio Verde	USA	0.77	10.1	51.7	15.0	0.39	0.09	77.3
Mean		0.87	9.43	53.5	14.1	0.39	0.15	77.6
LSD (5%)		0.27	2.0	2.3	1.6	0.06	0.13	1.96

Note. * Means over two years and two replications per year. ** Total unsaturated fatty acids (18:1+18:2+18:3+20:1+22:1).

Table 3. Mean concentrations of sugars* in seeds of seventeen lablab lines grown at Petersburg, Virginia (USA) during 2011 and 2012

Genotype	Sucrose	Fructose	Glucose	Raffinose	Stachyose	Verbascose	TNN**
PI 164772	1.19	0.26	0.46	0.40	2.34	0.22	4.14
PI 183451	1.75	0.38	1.39	0.45	3.25	0.41	5.85
PI 284802	1.64	0.23	0.19	0.37	2.25	0.19	3.99
PI 288466	1.98	0.62	1.60	0.50	3.32	0.55	6.35
PI 288467	1.33	0.38	0.25	0.40	2.62	0.21	4.57
PI 388003	2.60	0.78	1.49	0.62	4.06	0.57	7.87
PI 388012	1.49	0.26	0.24	0.47	2.50	0.39	4.85
PI 388013	1.39	0.35	0.91	0.49	2.72	0.33	4.94
PI 388017	2.04	0.77	1.55	0.56	3.35	0.33	6.29
PI 388018	1.14	0.32	0.32	0.41	2.38	0.19	4.14
PI 542609	1.21	0.54	1.03	0.51	2.57	0.19	4.48
PI 593055	1.00	0.30	0.32	0.36	1.96	0.22	3.54
PI 639277	0.95	0.23	0.27	0.38	2.27	0.24	3.85
PI 639279	1.39	0.41	1.11	0.47	3.09	0.27	5.21
PI 639280	1.33	0.38	0.30	0.42	2.84	0.23	4.82
PI 653615	1.39	0.30	0.93	0.42	2.93	0.22	4.97
Rio Verde	1.31	0.69	0.96	0.44	2.66	0.13	4.55
Mean	1.45	0.42	0.78	0.45	2.77	0.29	4.96
LSD (5%)	0.73	0.33	1.32	0.17	1.03	0.23	2.01

Note. * Means over two years and two replications per year. ** TNN: Total non-nutritive sugars (Raffinose+Stachyose+Verbascose).

Sucrose, fructose, and glucose concentrations in lablab seeds varied from 0.95 to 2.60 with a mean concentration of 1.45 g/100 g meal, 0.23 to 0.78 with a mean concentration of 0.42 g/100 g meal, and 0.25 to 1.49 with a mean concentration of 0.78 g/100 g meal, respectively. Concentrations of non-nutritive sugars

(Raffinose+Stachyose+Verbascose) in lablab seed varied from 3.54 to 7.87% with a mean concentration of 4.96 g/100 g meal. It is worth noting that first lablab cultivar released in USA (“Rio Verde”) contained the least concentration of Verbascose, one of the predominant non-nutritive sugars. Relative to concentration of total non-nutritive sugars in lablab seed, highest concentration was observed in PI-388003 which originated from Australia whereas several lines including “Rio Verde” contained minimal total non-nutritive sugars.

Statistical analysis indicated that correlations between several nutritional quality traits in lablab seed were significant. Significant positive correlations existed between concentration of C18:3 and concentrations of Stachyose sugars (0.25*) and total unsaturated fatty acids (0.73**) whereas significant negative correlations existed between concentration of C18:3 and concentrations of C22:0 (-0.63**), C22:1 (-0.54**), C24:0 (-0.42**), and total sugars (-0.74**). Total sugar concentration in lablab seeds was significantly and positively correlated with concentration of C18:2 (0.33**) and significantly and negatively correlated with concentrations of C14:0 (-0.34**), C18:1 (-0.32**) and C20:1 (-0.45**). Concentration of non-nutritive sugars was significantly and positively correlated with concentrations of fructose (0.70**), sucrose (0.96**), and total sugars (0.96**). More importantly concentration of non-nutritive sugars was significantly and negatively correlated with concentrations of C18:1 (-0.37**), C20:1 (-0.45**), and total saturated fatty acids (-0.26*) indicating that efforts to reduce non-nutritive sugars should not negatively affect concentrations of these fatty acids.

We compared the nutritional quality with several other food legumes based on information available from the literature (Table 4). In general, lablab seeds had comparable nutritional quality to that of black bean, navy bean, kidney bean, pinto bean, and pea. However, our results are based on only studies with a few lines researched over only two years. We suggest that additional studies might be needed for establishment and utilization of lablab as a food legume crop in United States.

Table 4. Nutritional quality traits of lablab, grown in Virginia, in comparison to literature values for other food legumes.

Trait	Lablab ¹	Black bean	Navy bean	Kidney bean	Pinto bean	Peas	
Fatty acids% of oil	C14:0	0.13				-	
	C16:0	15.8			12.8*	20.3*	16.0*
	C18:0	3.22			1.7*	0.4*	1.75*
	C20:0	0.68					
	C22:0	0.90					
	C24:0	1.49					
	C18:1	9.43			7.7*	20.3*	8.75*
	C18:2	53.5			21.5*	15.0*	38.0*
	C18:3	14.1			33.8*	21.0*	8.75*
	C20:1	0.39			0.0*	0.0*	n/a
	C22:1	0.15					0.00*
	Total saturated	22.2	25.8	22.3*	14.5*	20.8*	17.7*
	Total un-saturated	77.6	74.2	77.7*	85.5*	79.2*	82.2*
Sugars g/100 g meal	Sucrose	1.45	2.29**	2.44**			4.99*
	Fructose	0.42	0.84**	0.86**			0.39*
	Glucose	0.78	0.44**	0.55**			0.12*
	Raffinose	0.45	0.39**	0.48**			
	Stacyose	2.77	1.80**	2.31**			
	Verbascose	0.29	0.00**	0.01**			
	Total sugars	6.16	5.75**	6.64**			5.67*

Note. ¹Means from seventeen lablab lines grown in Virginia (USA) during 2011 and 2012 (This study). * USDA (2018). ** Smith et al. (2018).

4. Conclusions

We conclude that lablab has potential to be a new alternative food and food crop for Virginia and adjoin areas.

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