Lima Bean

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Introduction

According to the taxonomy, the bean belongs to the genus *Phaseolus*, which includes approximately 35 species of which 4 are cultivated: *P. vulgaris* L.; *P. lunatus* L.; *P. coccineus* L., and *P. acutifolius* L. (Arias-Restrepo et al. 2007). *Phaseolus lunatus* L. belongs to the Fabaceae family, and there are two domesticated genetic stocks from two different wild forms with two seed morphologies, small and large (Debouk 2019). The small seeds are known as ib., patashete and futuna (Yucatan, Chiapas, and Jalapa, Mexico, respectively), caballero bean (Cuba), ixtapacal (Guatemala), chilipuca (El Salvador), haba (Puerto Rico and Panama), sieva and comba (Colombia), and guaracaro (Venezuela), among others. The large seeds are known as lima, layo and pallar (Peru), torta (Colombia), palato (Bolivia), and manteotto (Argentina) (Debouk 2019).

It is proposed that *P. lunatus* could have originated in the Neotropical region of America, ranging from Mexico to Chile, passing through the Andean region of Peru. It is believed that its origin is found in Guatemala since in this area the wild progenitor of this species was found; on the other hand, molecular studies propose that its origin is found in the Andean zone and that its distribution throughout the Americas was given by domestication (FAO 2018).

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Table 1 Nutrient content in *Phaseolus lunatus* L. (g/100 g)

Components	Values
Moisture	12.0
Protein	10.1–20.7
Fat	0.1–1.2
Total carbohydrates	30.7–62.4
Crude fiber	2.1–4.9
Ash	1.7–3.7
Calcium (mg)	40–113
Phosphorus (mg)	124–330
Iron (mg)	3.5–4.8
Thiamine (mg)	0.27–0.34
Riboflavin (mg)	0.12–0.21
Niacin (mg)	1.66–2.2
Ascorbic acid (mg)	40

The nutrient content according to FAO (2020) for *Phaseolus lunatus* L. is (Table 1):

Other researches have shown its seeds have high protein (21–26%) and carbohydrate (55–64%) contents; high levels of minerals such as K, Zn, Ca, and Fe; and low levels of Na and P (Chel-Guerrero et al. 2012).

In Mexico, *Phaseolus lunatus* L. is widely grown in the tropic and is known as ib (Mayan dialect). It is common to find this legume in backyard home gardens; in Yucatan it is traditionally planted in the system rub-overthrow-burn (nomad agriculture) with corn and wild pumpkins (Debouk 2019). Betancur-Ancona et al. (2009) mentions its seeds are an underexploited protein source and could be a potential ingredient in industrial food systems.

Chronic Noncommunicable Diseases (CNCDs)

These are currently a major public health problem, since these diseases are one of the main causes of death and disability, both in developed countries and in those that are in development. These diseases include cardiovascular diseases, cancer, diabetes mellitus, and chronic respiratory diseases, among others (Gómez et al. 2010). CNCDs share a set of risk factors, including smoking, hypertension, high serum cholesterol levels, obesity, physical inactivity, and diabetes. A strategy to reduce the occurrence of these cases is primary prevention through community programs that promote and modify positively individual and group life habits (PAHO 2002).

Exercise and physical activity are important, due to the positive effects they cause on the improvement of health, increasing functional capacity and improving the quality of life of people (Gómez et al. 2010). Other aspects to take into account are changes in habits, specifically in the consumption of tobacco, alcohol (immoderate), and food. In Mexico, CNCDs are one of the greatest challenges which the health system is facing; due to the large number of cases that occur as they contribute to an increasing general mortality, they are also the most frequent cause of premature disability, together with the complexity and high cost for its treatment (Córdova-Villalobos et al. 2008).

Therefore, in Mexico the government has implemented the program Chécate, Mídete, & Muévete (2019) (Check Yourself, Measure Yourself, and Move), which contains tools and information to create healthy habits; tips are given regarding diets and exercise routines; in this portal people are encouraged to improve their health through the following steps: (1) visit to the doctor at the corresponding health clinic to keep track of the weight and measurements of the waist circumference; (2) BMI calculation and explanation of the relationship between food consumption and daily activity; and (3) calculation of the necessary physical activity based on sex, age, and activity level.

Postharvest Processing

Production

Nowadays, the cultivation of *Phaseolus lunatus* is practiced in different countries of Latin America and the Caribbean. The sieve ecotype is cultivated from Mexico to Argentina, while the Lima is present in a smaller area, comprised of the western zone of the Andes, mainly in Peru (FAO 2018); 7000 ha are sown in this country, and 11,000 tons are harvested annually (López-Alcocer et al. 2016). During the harvest season, harvesting can be done manually; however, it is important not to delay it since the pods can be opened, and most of the grains would be lost. The yield in terms of dry grains is in the order of 800–2000 kg/ha and can reach 3000 kg/ha in some regions depending on the variety (FAO 2018).

Storage Conditions

The first step after harvesting is to ensure that the pods reach a humidity of 12–14%, to subsequently be able to perform the extraction of the seed, always avoiding unnecessary breakage of the grains. Afterward, the storage of the seeds must be done in metal silos, avoiding the proliferation of insects and the increase of humidity (FAO 2018).

Poor handling and storage after the harvest induces the hardening process in the grain. According to the method dictated in the applicable Mexican regulations NMX-FF-038-SCFI-2002, hardened grain is that for which cooking time has increased significantly in relation to cooking freshly harvested grain. The hardening process in the grain is caused by the effects of aging or grain storage under conditions of high relative humidity in combination with high temperatures (temperature >25 °C and relative humidity >65%). Beans are considered hard when their cooking time is over 55 min. The aforementioned is very important because the temperature and relative humidity in the state of Yucatan are the same that induce the hardening effect (temperature >25 °C and relative humidity >65%, respectively), so storage becomes more important to avoid grain losses.

Toxicity

Legumes contain different antinutritional compounds, such as flatulence factors, saponins, protease inhibitors, tannins, phytic acid, lectins, etc. (De Dios et al. 2009); in the case of wild lima bean, plants contain cyanogenic glycosides that are known to defend the plant against leaf herbivores (Cuny et al. 2019); these seeds contain phaseolunatin, a glycoside which unfolds under the influence of an enzyme in glucose, hydrocyanic acid, and acetone. This compound is abundant in wild varieties making them poisonous (FAO 2020). Betancur-Ancona et al. (2004a) mention that all legumes contain antinutritional components and in the case of *P. lunatus* L. contain cyanogenic glycosides in quantities of 0.0369 g/kg, and these can limit its direct consumption in food and feed. During the soaking and cooking time of the grains, the glucoside is not completely eliminated, so the remnant portion of this compound continues to have its toxic properties (FAO 2020); the authors abovementioned proposed a wet-fractionation process for the detoxification of *P. lunatus* L.

In recent research, the lethal dose of hydrolysates and peptide fractions from *P. lunatus* L. have been evaluated (Nuñez-Aragón et al. 2019), founding that there was no mortality due to the administration of such materials at different concentration doses (10, 100, 1000, 1600, 2900, and 5000 mg/kg). The period of test in male ICR mice was for 14 days, and no behavioral alterations, signs of toxicity, or weight loss was detected during the observation period; the estimated LD_{50} was above 5000 mg/kg. Therefore, the wet-fractionation process proposed by Betancur-Ancona et al. (2004a) allows the detoxification of peptide fractions from *P. lunatus* L.

Nutritional Content of P. lunatus *L.*

Proximate Composition of Flour

In our work team, the *P. lunatus* L. legume has been extensively explored since 2000 until today. All lima bean seeds of our researches were purchased in the local market of the city of Merida; specifically, from the major distributor of the entity (supplier of the farmer, downtown, Merida, Yucatan). The harvest of the beans was done in the communities of the neighboring state of Campeche (Mexico).

Table 2 Proximate composition of flours from *P. lunatus* L. (g/100 g) harvested in Mexico

Components	2002	2004	2009	2015	2018
Moisture	14.88	10.24	14.80	11.02	9.30
Protein	24.07	25.50	23.70	23.82	20.86
Crude fiber	5.10	5.90	5.10	5.83	4.60
Fat	3.77	0.75	3.70	1.19	1.42
Ash	3.40	4.90	3.30	4.01	3.83
NFE	63.66	62.90	63.40	65.15	69.29

Chel-Guerrero et al. (2002), Betancur-Ancona et al. (2004a, 2009), Franco-Miranda (2015), Arias-Trinidad (2018)
NFE nitrogen-free extract

Table 2 shows the proximate composition of flours from *P. lunatus* in different years of harvest. It can be seen that despite the different harvest times, the proximal composition of grain has not been modified, and the results are similar to the report by FAO (2020, Table 1), positioning this legume as a stable source of macromolecules such as proteins and carbohydrates.

Giami (2001) reports the proximal composition of three new improved lines of Nigerian lima beans, observing similar values in the protein content (23.8, 24.4, and 27.3%), fat (1.5, 1.8, and 2.1%), ash (3.4, 3.5, and 3.6%), and crude fiber (2.4, 2.5, and 2.7%); the only differences between materials were in the NFE (53.4, 56.8, and 57.3%); these may be due to the genetic variations in the *P. lunatus* analyzed by the aforementioned authors.

Amino Acid Composition

Table 3 shows the amino acid composition of lima bean flour at different years of harvest, observing that the obtained flours from lima bean (ungerminated and germinated) supply the recommended amino acid scoring patterns of histidine, threonine, tryptophan, tyrosine, valine, isoleucine, leucine, phenylalanine, and lysine; but these flours are deficient in methionine and cysteine (FAO 2011). It is important to highlight that the obtention of germinated lima bean flour was made; however, this process does not improve sulfur amino acid deficiencies. An alternative to improve these deficiencies could be improved with a wet-fractionation process as mentioned by Betancur-Ancona et al. (2004a) with the objective to obtain protein isolates.

Wet-Fractionation Process

As mentioned, previously this process would allow obtaining protein isolates, starches, and fibrous residues; Fig. 1 shows the general scheme for this process. Betancur-Ancona et al. (2004a) report different flour/water ratio, pH and time of agitation to extract protein, and starch fractions from *P. lunatus* L. The

Table 3 Amino acid composition (g/100 g) of flours of *Phaseolus lunatus* L. harvested in Mexico

	(2004)	(2009)	(2009) germinated	(2015)	(2018)	FAO (2011)
Essentials						
Histidine	3.20	3.08	3.13	3.00	3.00	2.0
Threonine	4.87	4.46	4.40	4.10	4.36	3.1
Tryptophan	1.32	0.97	1.07	1.66	1.63	0.85
Tyrosine	10.67[a]	3.77	3.72	3.45	3.45	5.2[a]
Valine	5.12	4.40	4.41	5.90	4.54	4.3
Methionine	2.05[b]	0.81	0.63	0.81	1.00	2.7[b]
Cysteine	[b]	0.55	0.53	0.29	1.00	[b]
Isoleucine	4.29	3.86	3.92	5.71	5.28	3.2
Leucine	8.54	8.65	8.49	9.35	7.36	6.6
Phenylalanine	[a]	6.22	6.05	6.18	5.84	[a]
Lysine	7.97	7.10	6.85	5.69	5.94	5.7
Nonessentials						
Asp + Asn	14.43	12.98	13.86	13.16	10.98	
Glu + Gln	14.85	16.70	16.63	13.81	12.89	
Serine	7.99	7.97	7.93	5.44	5.99	
Glycine	4.87	4.46	4.34	2.48	2.74	
Arginine	7.01	6.37	6.42	10.13	12.25	
Alanine	5.01	4.82	4.83	3.31	3.19	
Proline	NR	2.84	2.80	5.43	8.56	

¥FAO (2011), recommended amino acid scoring patterns for child (6 months to older child), teenage and adult group

Betancur-Ancona et al. (2004a), Domínguez-Magaña (2009), Sandoval-Peraza (2015), Arias-Trinidad (2018). *NR* not reported

[a] Tyrosine + phenylalanine

[b] Methionine + cysteine

experimental design used in that study proved advantageous for choosing conditions to obtain starch and protein yield, depending on which of the products is required. For example if the starch is the priority product, the best treatment was with a 1:10 ratio (flour/water), pH 9, and 3 h of agitation; on the other hand, if the protein is the priority product, the best treatment was with a 1:6 ratio (flour/water), pH 11, and 1 h of agitation.

Protein Isolates

Table 4 shows the proximate composition of protein isolates (PI) from *P. lunatus* in different years of harvest. All PI were obtained by wet-fractionation process with the following conditions: 1:6 flour/water ratio, pH 11, and 1 h of stirring. It should be noted that the seeds of all studies were acquired from the same distributor, so the

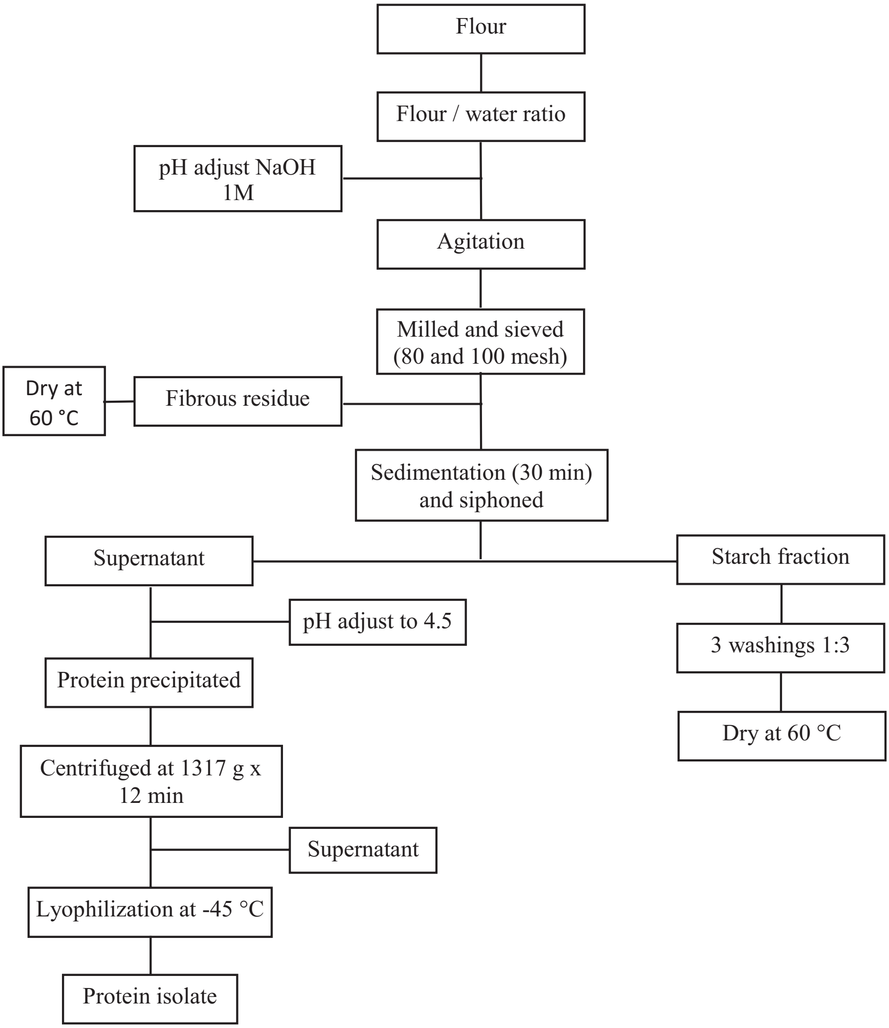

Fig. 1 Wet-fractionation process for integral use of *P. lunatus* L. flours

grains correspond to the same species and area of cultivation, but in different years of harvest, observing small differences in the protein content in the isolates (5%); therefore, these differences may be due to the protein extraction process.

Habitually the consumption of proteins in the human diet comes from animal sources such as meat, milk, eggs, etc.; however, a viable alternative could be the one mentioned by Linnemann and Dijkstra (2002), where proteins of vegetable origin can be used for the production of protein-rich foods, allowing to replace the

Table 4 Proximate composition of protein isolates from *P. lunatus* L. (g/100 g) harvested in Mexico

Components	2002	2009	2012 germinated	2015	2018
Moisture	7.87	8.6	5.4	3.38	8.8
Protein	71.13	69.9	69.14	74.06	62.37
Crude fiber	0.20	0.20	0.96	0.11	1.5
Fat	0.67	0.67	4.16	3.84	3.30
Ash	2.82	2.82	3.68	2.99	4.01
NFE	25.12	26.4	22.06	19	28.82

Chel-Guerrero et al. (2002, 2012), Betancur-Ancona et al. (2009), Franco-Miranda (2015), Arias-Trinidad (2018)
NFE nitrogen-free extract

consumption of meat in the human diet and in this way reduce the excessive exploitation of animals and how it affects the environment. Vioque et al. (2006) mention that in addition to obtaining protein concentrates, the hydrolysis of these serves to improve techno-functional properties or to obtain hydrolysates with biological activity, depending on their degree of hydrolysis.

Table 5 shows the amino acidic profiles of PI from *P. lunatus* obtained in different years of harvest. It can be observed that during the processing of lima bean flour from different harvests until the PI is obtained, there was a change in the amounts of all amino acids in comparison to the initial flours (Table 3). Note: the amino acid profiles shown in Tables 3 and 5 correspond to the flours and their respective PI.

The amino acid composition quantified in the materials of this study can be approached in the compliance with the daily requirements according to FAO (2011) for infants from 6 months to adults. The deficiency in sulfur amino acids continued in some cases even after obtaining the PI; on the other hand, when the amount of sulfur is covered, a decrease in aromatic amino acids was observed; it should be noted that the same behavior was observed in aromatic and sulfurized amino acids for PI obtained from germinated lima bean flours. These deficiencies can be solved by combining materials that are rich in the amino acids that lack IPs. For example, the proteins of corn and beans complement each other, providing significant amounts of each respective limited amino acids (Treviño-Mejía et al. 2016).

The BV calculated for the PI obtained by Sandoval-Peraza (2015) was 43.42 (Table 5) which is higher in comparison to that reported for beach pea (36.5) according to Chavan et al. (2001); also the value of this study is within the range reported by Pastor-Cavada et al. (2011) for 28 different species of beans with values between 18.7 and 67. The PER showed a value of 2.98, being higher than that reported by Betancur-Ancona et al. (2004a) for PI obtained from *Phaseolus lunatus* (2.5); these authors mentioned that the value of PER obtained classifies the PI as a good-quality protein. With the information obtained through the use of these formulas, it can be observed that the PI of *Phaseolus lunatus* can be an alternative source in the formulation of products enriched with protein.

Table 5 Amino acid composition (g/100 g) of protein isolates of *Phaseolus lunatus* L. harvested in Mexico

	(2004)	(2012)	(2012) germinated	(2015)	(2018)	FAO (2011)
Essentials						
Histidine	3.70	3.24	3.17	3.00	3.09	2.0
Threonine	4.71	4.40	4.41	3.95	3.95	3.1
Tryptophan	1.20	0.98	0.79	1.37	2.01	0.85
Tyrosine	11.33[a]	3.54	3.09	3.63	3.63	5.2[a]
Valine	5.80	4.79	4.88	4.65	4.65	4.3
Methionine	2.98[b]	0.35	0.50	0.57	1.60	2.7[b]
Cysteine	[b]	3.87	3.90	0.82	0.82	[b]
Isoleucine	4.78	4.30	4.11	4.35	4.95	3.2
Leucine	9.28	9.19	9.01	8.52	8.52	6.6
Phenylalanine	[a]	0.52	0.65	5.62	5.62	[a]
Lysine	7.55	5.99	5.94	7.07	7.32	5.7
Nonessentials						
Asp + Asn	12.79	12.80	12.47	12.05	10.43	
Glu + Gln	15.21	15.30	16.10	13.62	12.62	
Serine	7.38	7.39	7.26	5.25	5.25	
Glycine	4.70	4.67	4.84	4.55	5.12	
Arginine	6.13	4.96	5.02	9.50	9.50	
Alanine	5.17	6.08	6.19	2.81	2.81	
Proline	NR	7.62	7.68	8.59	8.59	
BV	–	–	–	43.2	–	
c-PER	2.5	–	–	2.98	–	

¥FAO (2011) recommended amino acid scoring patterns for child (6 months to older child), teenage, and adult group

Betancur-Ancona et al. (2004a), Chel-Guerrero et al. (2012), Sandoval-Peraza (2015), Arias-Trinidad (2018)

NR no reported

[a] Tyrosine + phenylalanine

[b] Methionine + cysteine

Starch Isolates

One of the products resulting from wet-fractionation is starch; approximately 10–40% of starch flour can be obtained for every 10 kg of lime beans (Betancur-Ancona et al. 2001, 2004a); Miranda-Villa et al. (2013) reported a 15% recovery for *Phaseolus lunatus*; however, the aforementioned authors did not solubilize the protein by changing the pH, observing that this process during the extraction plays an important role in the starch extraction yield.

Table 6 shows the proximal composition of starch isolates from lima bean noticing the lowest values of protein (under 0.2%); this property allows the use of these starches in the high-glucose syrup industry. 0.35% is the maximum allowed by the FDA in corn syrup to avoid the formation of undesirable dark syrups resulting from Maillard reactions during processing.

Table 6 Proximate composition of starches isolates from *P. lunatus* L. (g/100 g) harvested in Mexico

Components	2003	2005	2008
Moisture	0	11.93	10.9
Protein	0.12	0.10	0.14
Crude fiber	0.67	NR	0.10
Fat	0.54	0.12	0.12
Ash	0.14	0.04	0.65
NFE	98.53	98.49	98.9

Betancur-Ancona et al. (2003), Novelo-Cen and Betancur Ancona (2005), Segura-Campos et al. (2008). *NFE* nitrogen-free extract

It has been reported that the isolated starch of *P. lunatus* grown in Mexico has a composition of 60–70% amylopectin and 30–35% amylose, in addition to a total dietary fiber content of 1.25% (Betancur-Ancona et al. 2003; Novelo-Cen and Betancur-Ancona 2005; Segura-Campos et al. 2008); these authors mention that the values of amylose are similar to the reported in other legume starches like pinto bean, navy bean, and field pea (32–34%). The starch composition extracted from lime beans harvested in Colombia showed amylopectin and amylose values of 78.19 and 21.81%, respectively, presenting a slight difference (Miranda-Villa et al. 2013).

Fibrous Residue

After wet-fractionation it is common to have about a 30% yield of the fibrous fraction, Betancur-Ancona et al. (2004b) report a 35.43% of yield for *P. lunatus*.

Table 7 shows the proximal composition of fibrous residues from lima bean noticing the highest values of NFE and the difference content of crude fiber between samples; this occurs because NFE represents part of the cellulose, hemicellulose, pectin, and other carbohydrates not included in the crude fiber content as a result of the limitations of the method used; therefore a more adequate determination for this estimate would be total dietary fiber combined with the Van Soest fractions. Betancur-Ancona et al. (2004b) report the dietary fiber composition in the *P. lunatus* fibrous residues (29.4% total dietary fiber) observing that the insoluble fraction has the highest value (28.6%) followed by the soluble fraction (0.77%).

Functional Properties

This section shows the functional properties reported in the flour, protein isolates, starch, and fibrous residue obtained from *P. lunatus* L.

Table 7 Proximate composition of fibrous residues from *P. lunatus* L. (g/100 g) harvested in Mexico

Components	2004b	2004a
Moisture	4.72	10.54
Protein	11.1	6.30
Crude fiber	12.88	32.84
Fat	0.66	7.9
Ash	1.77	3.34
NFE	68.9	56.73

Betancur-Ancona et al. (2004a and b)
NFE nitrogen-free extract

Table 8 Water-holding capacity in *P. lunatus* flour and isolated fractions

Material	(g/g sample)	Reference
P. lunatus flour	2.65	Chel-Guerrero et al. (2002)
P. lunatus protein isolates (PI)	3.50	
Total globulin from *P. lunatus* at pHs 5, 7, and 9	1 1.05 1	Chel-Guerrero et al. (2011)
7S globulin fraction from *P. lunatus* at pHs 5, 7, and 9	3.4 1.5 3.3	
11S globulin fraction from *P. lunatus* at pHs 5, 7, and 9	3.3 2.9 3.2	

Water-Holding Capacity (WHC)

Table 8 shows the values reported for WHC in flour and different fractions obtained from *P. lunatus* L.

It is a fact that the best value of WHC was from PI compared to the seed meal; the authors of this research mention that a factor that can affect WHC is the concentration of carbohydrates present in the flour. Regarding PI, WHC may be due to the different protein fractions of this compound (e.g., albumins, globulins, etc.). The previous information was reported by Chel-Guerrero et al. (2011) where the 7S and 11S fractions of globulin had a higher WHC at different pHs compared to the total globulin; these authors demonstrate that extraction conditions and raw material composition play an important role in the resulting WHC properties, coinciding with the reports that at a higher pH, there is a greater interaction with water by proteins.

Oil-Holding Capacity (OHC)

Table 9 shows the values reported for OHC in flour and different fractions obtained from *P. lunatus* L.

Table 9 Oil-holding capacity in *P. lunatus* flour and isolated fractions

Material	(g/g sample)	Reference
P. lunatus flour	1.83	Chel-Guerrero et al. (2002)
P. lunatus protein isolates (PI)	4.59	
Total globulin from *P. lunatus* at pHs 5, 7, and 9	4	Chel-Guerrero et al. (2011)
7S globulin fraction from *P. lunatus* at pHs 5, 7, and 9	2.3	
11S globulin fraction from *P. lunatus* at pHs 5, 7, and 9	4.2	

PI from *P. lunatus* had the highest value of OHC in comparison with their respective flour; the authors of this study attribute the OHC to the high levels of nonpolar residues in the PI protein molecules. This functional capacity makes the PI from lima bean potentially useful in structural interactions in food, especially in flavor retention, improvement of palatability, and extension of shelf life in meat products through reduction of moisture and fat loss.

In accordance with OHC, it can be observed that the total globulin and its 11S fraction had similar values; this may be due to the fact that this fraction conforms approximately 60% of the total globulin. The authors mention that the differences in OHC between the 7S and 11S fractions were probably due to their conformational characteristics, which can influence a proteins capacity to entrap oil.

WHC and OHC of the fibrous fraction of *P. lunatus* obtained after wet-fractionation have also been reported, reporting values of 26.5% and 18%, respectively, observing that the fiber also has functional properties that can be used in the production of rich foods in fiber (Betancur-Ancona et al. 2004b).

Emulsifying Activity (EA) and Emulsion Stability (ES)

Chel-Guerrero et al. (2002) report the EA of lime bean flour in a pH range of 2–10, observing an EA comprised between 48 and 52%, presenting a stability decrease zone (2%) at a pH range of 6–7, increasing again to 50% from pH 8 and ending at 52% at pH 10. According to PI, a higher EA was observed at pH 2 (56%); however, at a pH range from 2 to 5, there was a notable decay of EA (42%), increasing again at pH 6 (52%) and ending at 50% at pH 10. The behavior of decrease and subsequent increase in each percentage presented by PI may possibly be due to the protein composition of the isolate as well as the amino acid content. Regarding ES, it was observed that both emulsions formed with lime bean flour and PI show a greater stability (80–100%) from pH 6. The authors mention that these results indicate that both (flour and PI) materials are effective emulsifiers, making them useful in applications such as the production of sausages, mayonnaise, and seasonings, especially in products that require heating.

A way to improve the emulsifying activity can be by making use of the enzymatic hydrolysis of the PI. The above was reported by Betancur-Ancona et al. (2009) for lime bean PI hydrolysates with the enzyme flavourzyme at 5 and 15 min of reaction. Noting that the highest values of EA were hydrolysates with flavourzyme at 15 min, at pHs 2 and 6 (80% at both pHs) followed by hydrolysates with flavourzyme at 5 min at pHs 2, 8 and 10 (65 and 60%, respectively). It should be noted that both hydrolysates showed an activity decrease at pH 4 (40%). These authors mention that the highest emulsifying capacity observed in the flavourzyme hydrolysates is probably because of the limited hydrolysis attained with this enzymatic complex, which provides a better hydrophobic/hydrophilic balance.

Another aspect that can be observed is that the hydrolysis of PI improves the functional properties; the aforementioned authors reported hydrolysis degrees of 2.8% and 6.3% for lime bean hydrolysates with flavourzyme at 5 and 15 min, respectively. Vioque et al. (2006) mention that obtaining a hydrolysis degree (DH) below 10% improves the functional properties (solubility, physicochemical and sensory properties) of a protein concentrate; values of DH above 10% allow the production of bioactive peptides; these could be used as food supplements or in medical diets.

Polanco-Lugo et al. (2014) report the EA for a limited hydrolysate of a *P. lunatus* PI with an enzymatic sequential system (pepsin-pancreatin), noting that the hydrolysate with a degree of hydrolysis of 1.7% at different pH values (2–10) had higher values of EA (99.3–261.11 m^2/g) compared to those of their respective PI (67.61–180.2 m^2/g). These authors mention that the increase in EA is probably due to suitable solubility of the limited hydrolysate in the system, as well as the increased hydrophobic surface due to limited hydrolysis of the globulin structure. On the other hand, the PI which had less interaction between nonpolar side chains of the proteins and lipidic chains might be attributed to the high extent of protein aggregation in PI caused by the extraction process.

Regarding the fibrous residue obtained from the wet-fractionation of *P. lunatus*, it has been found that it also has EA (49.3%) and ES (28.25%) (Betancur-Ancona et al. 2004b). The aforementioned authors indicate that the use of this fibrous fraction into food will depend on the type of product, for example, in products in which long shelf life is required, and thus higher emulsifying stability and the fibrous fraction from *P. lunatus* might not be so appropriate.

Foaming Capacity (FC)

The values reported to FC in flour, PI, and hydrolysates from *P. lunatus* L. are shown in Table 10.

Starch

In the case of starch from *P. lunatus* L., a high gelatinization temperature (80.16 °C) has been reported; this high gelatinization temperature of lima bean starch may allow use of higher temperatures than those used for other common starches during thermal processing food, in order to achieve complete gelatinization and to assure

Table 10 Foaming capacity in flour, PI, and hydrolysates from *P. lunatus* L.

Material	pH	Values (%)	Reference
Flour from *P. lunatus* (120 min)	2	23	Chel-Guerrero et al. (2002)
	4	18	
	6	16	
	8	15	
	10	15	
PI from *P. lunatus* (120 min)	2	30	
	4	10	
	6	5	
	8	15	
	10	10	
Limited hydrolysates from *P. lunatus* (flavourzyme at 5 and 15 min separately; hydrolysis degree (DH) 2.8 and 6.3, respectively; 120 min)	2	142 and 160	Betancur-Ancona et al. (2009)
	4	138 and 140	
	6	141 and 142	
	8	148 and 159	
	10	150 both	

its thickening effect. The solubility and the swelling power were correlated in a direct way with the temperature; the swelling pattern for *P. lunatus* starch shows swelling resistance at lower temperatures than 75 °C; in the range of 70–90 °C, the granules swell gradually when the temperature increases (Betancur-Ancona et al. 2001).

Campechano-Carrera et al. (2007) report the effect of the pyrodextrinization of starch extracted from *P. lunatus* indicating that this process dramatically reduces viscosity compared to native starch (13.2 and 752 cP, pyrodextrinized and native starch from lima bean, respectively). These authors indicated that the pyroconversion preferentially affects amorphous areas mainly formed of the ramified chains causing a reduction in the molecular weight and conversion to more highly branched molecules that are responsible for imparting viscosity. Another fact which may cause a drastic reduction in viscosity was the addition of HCL during the pyrodextrinization process, because this reagent caused hydrolysis and rearrangement of the starch molecules.

The functional properties that have been reported for flours, protein concentrates, hydrolysates, starches, and fibrous residues can provide a basis for choosing the best terms of use and application of these materials for the production of functional foods or for the improvement of those already present in the market.

Table 11 Bioactive properties for hydrolysates from *P. lunatus* L.

Hydrolysate conditions	Values	Activity	Reference
Sequential hydrolysis (60 min with pepsin-pancreatin), hydrolysis degree (DH) 15.97%	IC_{50} 0.321 mg/mL TEAC 13.2 mM/protein	I-ACE Antioxidant	Polanco-Lugo et al. (2014)
Hydrolysis with alcalase (90 min), DH 32%	IC_{50} 0.056 mg/mL TEAC 9.89 mM/protein	I-ACE Antioxidant	Torruco-Uco et al. (2009)
Hydrolysis with flavourzyme (90 min), DH 22%	IC_{50} 0.0069 mg/mL 8.42 mM/protein		
Hydrolysis of germinated seeds with alcalase (30 min), DH 30.34%	IC_{50} 0.61 mg/mL	I-ACE	Domínguez-Magaña et al. (2015)
Hydrolysis with alcalase (120 min), DH 51.28%	IC_{50} 0.56 mg/mL		
Sequential hydrolysis (60 min with pepsin-pancreatin), DH 32.16	IC_{50} 0.25 mg/mL		
Sequential hydrolysis of germinated seeds (60 min with pepsin-pancreatin), DH 32.16	IC_{50} 0.28 mg/mL		

Bioactive Properties

This section shows the bioactive proprieties reported from *P. lunatus* L. hydrolysates; Table 11 shows these activities.

Within our working group, extensive hydrolysis conditions have been analyzed extensively to obtain hydrolysates with biological activity, among which I-ACE and antioxidant stand out. Within the different bioactivities shown in Table 11, it seems that the best I-ACE values are obtained at 60–120 min of enzymatic action, either the individual or sequential enzyme, flavourzyme being the one that produces hydrolysates with the highest I-ACE activity.

An important aspect is mentioned by Torruco-Uco et al. (2009) that there is no correlation between I-ACE and antioxidant activity. This suggests that the antioxidant activity of the peptides may depend on the specific proteases used to produce them, the degree of hydrolysis attained, the nature of the released peptides (molecular weight, composition, and amino acid sequence), as well as the combined effects of their properties, including their ability to locate free radicals, acting as chelating agents of metal ions or as a hydrogen donor (Tang et al. 2009).

An alternative to improve the biological activity of the hydrolysates generated can be by obtaining peptide fractions of different molecular weight, by ultrafiltration (Cho et al. 2004). In this regard, Ciau-Solís et al. (2017) report the I-ACE activity of a molecular weight fraction >3 kDa from a lime bean hydrolysate obtained by sequential hydrolysis with pepsin-pancreatin at 90 min with a DH of 32.33%. This fraction showed an IC_{50} value of 0.172 mg/mL which is higher for hydrolysates obtained with the same enzymatic sequential system (IC_{50} of 0.321, 0.25, and 0.28 mg/mL Table 11). This shows that obtaining peptide fractions can be an alternative to improve the biological properties that the hydrolysate can present.

Cordova-Lizama et al. (2013) report the antithrombotic and anticariogenic activity in lime bean hydrolysates obtained with the enzyme pepsin, finding an 88% inhibition of platelet aggregation at a hydrolysate concentration of 4.5 mg/mL. Regarding the anticariogenic activity, a reduction in the demineralization of calcium and phosphorus was found in a hydroxypatite matrix by 50 and 55.8%, respectively.

Bojorquez-Balam et al. (2013) report the antimicrobial activity of hydrolysates and peptide fractions (>10 and <10 kDa) of lima beans. Finding that both the hydrolysate and the peptide fractions showed antimicrobial activity against *S. aureus* and *S. flexneri*, the <10 kDa fraction presented the highest inhibitory activity with 392.04 µg/mL for *S. aureus* and 993.17 µg/mL for *S. flexneri*.

Renin-inhibitory activity is another bioactive property that has been found in peptide fractions of *P. lunatus*, with inhibition values of 31.73 and 30.05% being observed for >3 kDa molecular weight fractions obtained with sequential alcalase-flavourzyme and pepsin-pancreatin systems, respectively (Ciau-Solís et al. 2017).

Another fraction that has shown biological activity is the fibrous residue obtained from the wet-fractionation processing of lima beans; it has been found that this portion has antioxidant activity (35.5%) which is slightly lower than those presented by hydrolysates obtained from the same seed (51.43–61.34%) (Torruco-Uco et al. 2009).

The biological properties that can be obtained from protein hydrolysates and fibrous residues from *P. lunatus* could be implemented in the preparation of functional products that provide some benefit to the organism beyond its nutritional value or elaborate nutraceutical products that could be used both in the prevention and treatment of chronic degenerative diseases.

Product Development

Functional Foods

This term was first used in Japan (1984) as a result of a study on the relationships between nutrition, sensory satisfaction, fortification, and modulation of physiological systems in order to define those food products fortified with special constituents that possess advantageous physiological effects (Bigliardi and Galati 2013). Otherwise this term is considered a marketing term only, and there is no consistent definition recognized globally by regulatory institutions. Crowe and Francis (2013) mention that the Academy of Nutrition and Dietetics define functional foods as:

Whole foods along with fortified, enriched, or enhanced foods that have a potentially beneficial effect on health when consumed as part of a varied diet on a regular basis at effective levels.

In Mexico the term functional food is widely used in the scientific field; but to date there are no laws that specifically regulate the use and production of these foods; however, their consumption is aimed at obtaining a health benefit. According to Hartmann and Meisel (2007) in the market, there is a wide variety of functional products added with bioactive peptides, for example, Calpis (Calpis Co., Japan), Evolus (Valio, Finland), BioZate (Davisco, USA), C12 Peption (DMV, the Netherlands), peptide soup (Nippon, Japan), ProDiet F200 (Ingredia, France), Capolac (Arla Foods, Denmark), etc. In Mexico, companies such as LALA® (2019) have de-lactose with fiber within their milk line, which provides more than 10% of the fiber required per day per serving. Bimbo® (2019) commercializes double fiber bread, which provides 25% per serving of the daily fiber requirement. The previously mentioned shows the need of the population to acquire food containing any compound that could provide some benefit to the organism during its consumption.

When talking about functional foods, these are commonly confused with nutraceuticals; however, they differ from each other, since a nutraceutical product is a substance of natural origin that, when ingested, in the organism behaves as a medicine, providing a beneficial effect for health beyond its nutritional value (Cortés et al. 2015). Recently the definition was modified by Health Canada, defining nutraceutical as a product isolated or purified from foods, and generally sold in medicinal forms (pills, capsules, powders, etc.) not usually associated with food and demonstrated to have a physiological benefit or provide protection against chronic disease (Monge 2008).

Foods Added with **Phaseolus lunatus** *L.*

This section shows the different foods that have been prepared with the addition of hydrolysates, starch, and fibrous residue extracted from *P. lunatus*.

Functional Foods

(a) *Flour*

Pérez-Navarrete et al. (2006) report the preparation of extrudates prepared with mixtures of cornmeal and lima beans in relation to 75:25, 50:50, and 25:75, respectively. It was observed that the property of expansion decreases as the content of bean flour increases; according to the density and maximum breaking force by compression, an increase of these parameters was observed as the proportion of bean flour increased. The 50:50 mixture was considered potentially nutritious because it met the requirements of essential amino acids such as lysine and tryptophan in 100%.

(b) *Protein Isolates*

Davalos-Cervera (2003) reports the elaboration of Frankfurt-type sausages added with protein isolates of *P. lunatus* with addition levels of 3.5 and 7%. An increase in the protein content of 46.49 and 51.66%, respectively, was observed, being higher than the control (39.71%); sensory evaluation showed acceptance for sausages with a 3.5% addition and rejection for sausages made with 7%. For both products, minimal concentrations of cyanogenic glycosides (0.188 and 0.559 mg/100 g) were found without jeopardizing their consumption.

(c) *Hydrolysates*

Franco-Miranda et al. (2017) reports the incorporation of protein hydrolysates from lima bean (1 and 3%) in the production of concha-type Mexican sweet bread, observing that the addition of *P. lunatus* hydrolysates to bread dough affected its rheological properties, decreased tenacity, and slightly increased elasticity compared to the control. The I-ACE activity of bread with a 3% hydrolysate addition showed the highest activity (83.10%) compared to that added with 1% and the control (69.61 and 11.27%, respectively); regarding the antioxidant activity, bread with a 3% addition also showed the highest value (TEAC 17.19 μmol trolox/g) compared to that added with 1% and the control (TEAC 15.59 and 5.42 μmol trolox/g, respectively). The bread added with 3% lima beans had a low acceptance due to a slight acidic taste.

(d) *Starch*

Chim-Rodríguez (2000) reports the preparation of cookies added with 50% of lima bean starch; the cookies had a total dietary fiber content of 3.67%, 2.42% insoluble dietary fiber and 1.24% soluble dietary fiber. The sensory test showed that cookies added with *P. lunatus* starch were accepted even over the control cookies.

(e) *Fibrous Residue*

Peraza-Mercado (2000) reports the addition of fibrous residue of *P. lunatus* in an addition percentage of 2.6%. The total, insoluble, and soluble dietary fiber content was 9.23, 7.14 and 2.08%, respectively; the sensory evaluation reported a low acceptance for these cookies, presenting softness compared to the control cookies.

Perspectives

With the information presented in this section, it is shown that both the flour and each of the fractions that can be obtained from *P. lunatus* may well be used in the production of functional foods with sensory acceptance. The aforementioned will depend on choosing the product to be prepared in conjunction with the functional properties of the different fractions of lima beans that were discussed in "Functional Properties" section.

Nutraceuticals

Encapsulation is a process to entrap one substance defined as an active agent within another substance defined as wall material. In the food industry, an encapsulation process can be applied for different reasons, for example, improve delivery of bioactive molecules (e.g., antioxidants, minerals, vitamins, phytosterols, fatty acids, etc.) and living cells (e.g., probiotics) into foods. Encapsulation technology refers to a technique in which the bioactive component is completely enveloped, covered, and protected by a physical barrier, without any protrusion of the bioactive component. Also, microcapsules release their contents at controlled rates over prolonged periods and under specific conditions (Nedovic et al. 2011).

To guarantee the activity of these bioactive peptides, it must remain active and intact during the gastrointestinal digestion and absorption in order to achieve their physiological effects. But once it is in the organism, all peptides go through different barriers that can inactivate them and consequently lose their efficiency (Segura-Campos et al. 2011). In addition to improve the beneficial effect of bioactive peptides, the microencapsulation could be a way to protect the peptides against the environment to which they are exposed. Microencapsulation protects bioactive peptides and ensures the release of an appropriate dosage at a gastric or intestinal pH. In this way the encapsulation of hydrolysates and peptide fractions with I-ACE and antioxidant activity from *P. lunates* were evaluated.

Ruiz-Ruiz et al. (2013) reports an IC_{50} of residual I-ACE in intestinal media of 2.9 mg/mL for encapsulated hydrolysates and released by in vitro digestion; the encapsulation conditions were pH 4, 1 mM $CaCl_{2,}$ and a 50:50 ratio of alginate sodium and carboxymethylated flamboyant gum. It was observed that the conditions selected for the encapsulation of this hydrolysate allowed an efficiency of 71.7%.

Sandoval-Peraza et al. (2014) report the encapsulation of a peptide fraction with a <10 kDa weight obtained from protein hydrolysates of *P. lunatus* by ultrafiltration, by means of the ionic gelation technique using alginate and carboxymethylated flamboyant gum. The treatment with the highest encapsulation efficiency achieved a 33.43% entrapment of the peptide fraction; the encapsulation conditions for this treatment were 50:50 gum ratio (alginate/flamboyan), 0.1 M $CaCl_{2,}$ and 25 min of hardening time. After an in vitro digestion, the antioxidant activity and remnant I-ACE in the intestinal environment were 615.10 mTEAC/mg protein and 0.035 mg/mL of IC_{50}; it should be noted that the IC_{50} value and antioxidant activity of the fraction released were higher compared to the one that was initially encapsulated (0.37 mg/mL of IC_{50} and 26.94 TEAC mM/mg protein, respectively).

Notably hydrolysates as well as their fractions can be used in the preparation of nutraceutical products, and it has been demonstrated that these fractions maintain their biological activity even after being exposed to an in vitro digestion.

Future Trends in Processing and Product Development

Currently, Mexico and the United States occupy the first places of worldwide prevalence of obesity in the adult population (30%), which is ten times higher than that found in countries such as Japan and Korea. Regarding the child population, Mexico ranks fourth in the worldwide prevalence of obesity, approximately 28.1% in boys and 29% in girls, surpassed only by Greece, the United States, and Italy (Dávila-Torres et al. 2015).

In our country, according to ENSANUT (2012), the state of Yucatan is among the first places in obesity nationwide. This is alarming, since the development of obesity is the first step to develop chronic degenerative diseases. Therefore, it is important to implement national strategies such as those mentioned before (Check yourself, Measure yourself, and Move), along with the search for functional and nutraceutical products that can help in the treatment and prevention of chronic degenerative diseases.

This review intends to explore the seeds of *Phaseolus lunatus* (underexploited species) grown and harvested in the region (Yucatan, Mexico) as a possibility to obtain compounds with functional and biological activity and their subsequent implementation in the production of functional foods and nutraceuticals. It should be noted that the data presented here can be used as a starting point and improvement for research involving the use of *Phaseolus lunatus*, both nationally and internationally, since the cultivation, consumption, and use of this legume extend from Mexico to South America (Chile).

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