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Nutritional, rheological and sensory evaluation of *Lupinus mutabilis* food products – a Review

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Abstract: This review presents an overview of the state-of-art on uses of *Lupinus mutabilis*. This valuable legume is cheap, eco-friendly, has good taste and could be used to increase the protein content and to improve the fat and protein profile of more than fifty processed and fresh products (*i.e.* spaghetti, lasagne, snacks, bread, hamburgers, sweets, soups, and salads). *L. mutabilis* might also be used to prepare meat, milk and yoghurt substitutes with good sensory evaluation. Sensory evaluation of specific fermented sausage and jelly ranked better than the control. Specific *L. mutabilis* spaghetti had similar rheological behaviour like the control. Bread with 10% of *L. mutabilis* flour had a protein efficiency ratio (76%) higher than the control (28%) and similar acceptability. *L. mutabilis* jelly could reduce postprandial glucose in people with non-insulin dependent diabetes and *L. mutabilis* purée could be eaten by people with celiac disorders (especially babies). Data on each product is critically evaluated to infer conclusions and to make suggestions to improve the sensory, rheological and nutritional quality of lupin products.

Keywords: grain products; new foods; nutrition; rheology

Lupins (*Lupinus spp.*) are very important legumes used for hundreds of years as a protein source in human and animal nutrition (GÜEMES-VERA *et al.* 2008; CARVAJAL-LARENAS *et al.* 2015b). The FAO (2019) reports that 1,610,969 tonnes were produced worldwide in 2017 (Oceania 64.0%, Europe 27.5%, Africa 4.7%, and Americas 3.8%). From the four major cultivated species – *Lupinus albus*, *Lupinus luteus*, *Lupinus angustifolius*, and *Lupinus mutabilis*, the latter shows the highest average content of protein 43.3 g/100 g of DW and of fat 18.9 g/100 g of DW (CARVAJAL-LARENAS *et al.* 2014), which is comparable to the contents in soya bean (Table 1). Debittered lupin can be eaten directly as a snack (VILLACRÉS *et al.* 2003), or can be used as an ingredient in many different products (VILLACRÉS *et al.* 2003; GÜEMES-VERA *et al.* 2008) such as fresh salads, soups, cakes, hamburgers, bread, sausages, pasta, etc. Debittered

lupins can also be used to prepare meat, milk and yoghurt substitutes (JIMÉNEZ-MARTÍNEZ *et al.* 2003; VILLACRÉS *et al.* 2003; VILLACRÉS *et al.* 2006; GÜEMES-VERA *et al.* 2008). The replacement of meat and cow milk (totally or partially) with lupin would be advantageous because the production of grains in general and lupins in particular uses less resources and therefore it is cheap and eco-friendly (DE VRIES & DE BOER 2010; MALAV *et al.* 2015; OSEN *et al.* 2014; JONES 2015). *L. mutabilis* products seem to meet another important requirement for acceptance, a good taste (JACOBSEN & MUJICA 2006). In addition, some of these products might have nutraceutical applications (VALLEY & SIPSAS 2010; BALDEÓN *et al.* 2012). However, and despite the fact that lupins in general and their products in particular can be very nutritious, they are much less studied when compared with soya bean and its products.

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This study has been conducted to critically evaluate studies on lupin-based products, giving special attention to the chemical composition and nutritional, sensory and instrumental evaluation while it is also possible to compare information from different authors. Gaps of information and research needs are also presented. This study has included products obtained from full-fat lupin flour (protein 44.8%, fat 26.3%, fibre 9%, humidity 3.6%, ash 2.1%, and carbohydrates 14.2%), defatted flour (protein 32.5%, fat 4.5%, carbohydrates 48.1% and humidity 6.4%), lupin protein concentrates (protein 66.1%, moisture 7.8%, fibre 0.0%, and ash 1.0%) and lupin isolates (protein 86.3% DM, fat 2.1% DM, fibre 0.4% DM, ash 2.3% DM and carbohydrates 8.7% DM), and wheat flour (protein 10.4% DM, fat 1.1% DM, fibre 2.2% DM, ash 1.0% DM, and carbohydrates 85.3% DM).

Bread

Several authors have used wheat-lupin mixtures containing up to 20% of lupin flour or up to 10% of lupin protein concentrate or up to 4% of lupin isolate in bread-making, obtaining a product with higher protein (Table 2) and improved amino acid content (JACOBSEN & MUJICA 2006; GÜEMES-VERA *et al.* 2008; KOHAJDOVÁ *et al.* 2011). Thus, the protein efficiency ratio (PER) of bread made with 10% of *L. mutabilis* flour was claimed to rise from 28% (in bread from 100% wheat flour) to 56% (GUEGUEN & CERLETTI 1994) (standard = 100% casein).

Regarding sensory evaluation of lupin breads, the results are different. Some authors claimed to be accept-

able (or with an evaluation similar to the control) when the mix had up to 10% of lupin flour or up to 5.0% of lupin protein concentrate or up to 2.0% of isolated lupin protein (DERVAS *et al.* 1999; GÜEMES-VERA *et al.* 2008). On the other hand, other authors reported an inferior quality of bread when compared to the control (100% wheat bread (GÜEMES-VERA *et al.* 2008; ROSELL *et al.* 2009) (Tables 3 and 4). This difference in results might be explained by variations in formulations (bread type) and processes. For example, favourable variations in volume could be associated with lupin endogenous enzymes which could produce additional gas. Moreover, the addition of starch (and amylose specifically) is important during pasting (gelling and recrystallization) by its positive influence on volume. On the other hand, smaller volume is suggested to be influenced by the process (inadequate fermentation time), as well as by inadequate energy applied – like mixing and heating conditions – to the dough) which in turn would affect the hydration level, heat-induced aggregation and unfolding of its proteins (ROSELL *et al.* 2009). Moreover, the action of yeast could be influenced by formulation and matrix composition. Thus, the addition of lupin flour, protein concentrate or isolate means increment of globulin proteins and decrement of starch content, which could interfere with the formation and quality (consistency) of gluten network (ROSELL *et al.* 2009). The resulting effect of adding lupin to formulations without adjusting the formula and/or process could generate a matrix with lower (or weaker) interconnection of gluten proteins resulting in a decrease of trapped CO₂ (ROSELL *et al.*

Table 1. Average composition of whole raw and debittered lupin seeds (g/100 g dry weight)

Material	<i>L. albus</i>	<i>L. luteus</i>	<i>L. angustifolius</i>	<i>L. mutabilis</i>	<i>L. mutabilis</i> ¹	Source
Macronutrients						
Moisture	8.6	9.4	9.0	8.1	74.3	
Proteins	38.2	42.2	33.9	43.3	57.5	
Lipids	11.2	5.5	6.3	18.9	16.6	CARVAJAL-LARENAS <i>et al.</i>
Fibre	8.9	15.8	16.0	8.2	7.2	(2015b)
Ash	3.4	3.8	3.0	3.9	2.9	
Carbohydrates	38.3	32.7	40.8	25.7	15.8	calculated ²
Unsaturated fatty acids						
Oleic (C18:1)	6.0	1.6	2.1	8.8	8.7	calculated according to
Linoleic (C18:2)	2.1	2.7	2.5	6.3	4.7	CARVAJAL-LARENAS <i>et al.</i>
Linolenic (C18:3)	1.0	0.3	0.4	0.5	0.5	(2015b)

¹debittered, ²calculated by difference (exclude fibre)

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Table 2. Chemical composition of some products made with lupin flour or lupin isolated (%)

Product	Moisture	Protein	Ash	Fat	Carbohydrates	Starch	Fibre	Reference
Bread								
100% WF	–	13.4		10.3	–	–	2.0	JACOBSEN & MUJICA (2006)
87.5% WF + 12.5% LF	–	17.2		13.1	–	–	2.7	
Spaghetti								
100% WS	13.5	12	0.9	2.2	–	73.9	–	DOXASTAKIS <i>et al.</i> (2007)
95% WS, 5% ILP1	12.4	15.4	1.1	1.8	–	71.2	–	
90% WS, 10% ILP1	11.9	18.7	1.3	1.6	–	63.6	–	
85% WS, 15% ILP1	11.6	21.9	1.4	1.5	–	61.4	–	
80% WS, 20% ILP1	11.9	25.4	1.4	1.3	–	57.4	–	
Meat substitute								
Grilled beef (100% meat)	54.7	21.2	–	17.3	0.0	–	0.0	USDA (2019)*
<i>L. mutabilis</i> meat (dehulled)	–	57.9	–	25.1	–	–	3.5	VILLACRÉS <i>et al.</i> (2006)
<i>L. mutabilis</i> meat (whole) ¹	–	50.8	–	19.8	–	–	10.0	
Sausages								
FRA normal (0% ILP2)	56.5	11.3	2.3	29.8	–	–	–	ALAMANOU <i>et al.</i> (1996)
FRA 1% ILP2 of product	57.2	11.8	2.5	28.3	–	–	–	
FRA 2% ILP2 of product	57.4	12.6	2.5	27.4	–	–	–	
FRA 3% ILP2 of product	58.0	13.5	2.5	25.9	–	–	–	
FER normal (0% ILP2 + 0% LSF)	52.6	15.3	3.3	28.2	0.3	–	–	PAPAVERGOU <i>et al.</i> (1999)
FER 2% LSF of meat	52.6	15.3	–	–	1.0	–	–	
FER 2% ILP3 of meat	52.6	15.3	4.4	26.7	–	–	–	
Milk and yoghurt substitutes								
100% CM	87.0	3.5	–	3.3	4.8	–	–	VILLACRÉS <i>et al.</i> (2006)
LM	87.5	3.5	–	1.6	1.0	–	–	
CM yoghurt	87.9	3.5	–	3.2	4.7	–	0.0	USDA (2019)**
<i>L. mutabilis</i> yoghurt	–	3.7	1.2	2.2	–	–	–	VILLACRÉS <i>et al.</i> (2006)
(CM 80% + LM 20%) yoghurt	78.0	3.9	0.7	3.0	14.1	–	0.2	CASTAÑEDA-CASTAÑEDA <i>et al.</i> (2008)
(CM 70% + LM 30%) yoghurt	78.0	3.9	0.7	2.9	14.0	–	0.3	
Jelly								
BJ	44.8	0.9	0.5	–	–	–	–	ROMERO & MEDINA (2004)
BJ + 7.5% LF	45.3	5.6	0.5	–	–	–	–	
BJ + 15% LF	47.6	9.1	1.0	–	–	–	–	
Purée								
Potato Babyfood	86.6	1.0	–	0.1	11.7	–	0.9	USDA (2019)***
25% O + 25% WC + 50% <i>L. mutabilis</i>	–	25.3	–	14.2	16.0	–	4.4	LEÓN-MARROÚ <i>et al.</i> (2011)

WF – wheat flour; LF – *L. mutabilis* flour; WS – wheat semolina; ILP1 – isolated lupin protein and the dehulled product riboflavin 1.7, niacin 4.2, pyridoxine 0.12, and cobalamine 0.02 (mg/100 g); ¹riboflavin 2.4, niacin 23.2, pyridoxine 0.42, cobalamine 0.03 mg/100 g, phosphorus 570 mg/100 g, manganese 9.52 mg/100 g and the dehulled product riboflavin 1.7, niacin 4.2, pyridoxine 0.12, cobalamine 0.02 (mg/100 g); FRA – Frankfurter sausages; ILP2 – *Lupinus albus* ‘Graecus’ protein isolated; FER – fermented sausages; LSF – lupin seed flour; ILP3 – isolated lupin protein; CM – Cow’s milk; LM – *L. mutabilis* milk; BJ – blackberry jelly; LF – lupin flour; O – oca; WC – white carrot; USDA – Food composition databases available at: <https://ndb.nal.usda.gov/ndb/foods/search/list>; *basic report 23141; **basic report 01116; ***basic report 03112

Table 3. Sensory and/or instrumental evaluation of some products made with lupin flour or lupin isolated

Used in	Evaluator	Evaluation time	Formula	Evaluation	Reference
Bread	n.a.	n.a.	90% WF + 10% LF	acceptability 93% compared with 100% wheat bread	(GROSS <i>et al.</i> 1983)
Loaf bread	1 trained and 35 untrained	n.a.	95% WF + 5% LF	most acceptable	
Sweet bread	1 trained and 35 untrained	n.a.	90% WF + 10% LF	most acceptable	GÜEMES-VERA <i>et al.</i> (2004)
Flat bread	n.a.	n.a.	40% WF + 40% CF +20% LF	successful recipe	CREMER (1983)
Spaghetti	instrumental	n.a.	97% WS + 3% ILP3	hardness, similar to control, extensibility reduced 50%	LÓPEZ-SANTOS <i>et al.</i> (2006)
Spaghetti	instrumental	n.a.	1kg blend (5% ILP3 + 95% WS) + 350 ml water	similar to control	DOXASTAKIS <i>et al.</i> (2007)
Spaghetti	instrumental	n.a.	1kg blend (20% ILP3 + 80% WS) + 350 ml water	bad rheological and cooking performance	
FRI	n.a.	n.a.	fermented lupin (24h, <i>R. oligosporus</i>)	like very much	CHÁVEZ & PEÑALOZA (1988)
FRA	15 untrained	1 week	1–2% of ILP2	like	
FRA	15 untrained	1 week	3% of ILP2	dislike	ALAMANOU <i>et al.</i> (1996)
FER	7 trained	after 1 month	20 g of ILP2 per kg	like more than control	PAPAVERGOU <i>et al.</i> (1999)
Flavoured LM	n.a.	n.a.	31% LF + 62% water + 7% sugar + stabilizer +flavour	good organoleptic valorisation	VILLACRÉS <i>et al.</i> (2006)
Vainilla FLY	38 untrained	n.a.	80% CM + 20% LM	like a little	CASTAÑEDA-CASTAÑEDA <i>et al.</i> (2008)
FLY	65 untrained	n.a.	1l LCM +15 g lactose + 30 g sucrose+30 g started culture	between likes and likes much	JIMÉNEZ-MARTÍNEZ <i>et al.</i> (2003)
BJ	80 untrained	n.a.	53.5% blackberry + 46.5% fructose	between like a lot and like	
BLJ	80 untrained	n.a.	53.5% blackberry + 39% fructose +7.5% LF	like	ROMERO & MEDINA (2004)
BLJ	80 untrained	n.a.	53.5% blackberry + 31.5% fructose +15% LF	indifferent	

WF – wheat flour; LF – *L. mutabilis* flour; CF – cassava flour; WS – wheat semolina; ILP3 – isolated *Lupinus mutabilis* protein; FRI – fried lupin meat; FRA – Frankfurter sausages; ILP2 – isolated *L. albus* ‘Graecus’ protein; FER – fermented sausages; LM – *L. mutabilis* milk; FLY – flavoured lupin yoghurt; CM – cow milk; LCM – *L. campestri* milk; BJ – blackberry jelly; BLJ – blackberry lupin jelly

2009; KOHAJDOVÁ *et al.* 2011) and a poor bread texture (GÜEMES-VERA *et al.* 2004).

Differences in bread texture might be explained by thermomechanical variations during processing. For example, ROSELL *et al.* (2009) performed a study on five wheat – *L. mutabilis* flour blends (0, 12.5, 25, 50, and 100% lupin; mixing 6 min at 30°C, heating rate 4°C/min until 90°C, holding 7 min, cooling rate 4°C per min until 55°C and holding 5 min). Results showed that the increasing lupin content in dough (up to 25%) did not affect the dough consistency significantly, (probably) due to lupin proteins masking the effect of starch dilution.

The cooling period showed again that up to 25% of substitution, the consistency of lupin doughs was not extremely different from those made with 100% wheat flour. Perhaps that would be the effect of interactions between wheat amylose and lupin lipids, which could be acting as a surfactant combination. Lupin blends (50 and 100% lupin) had a very different behaviour. In order to confirm these results, the authors made lupin breads. However, only the samples containing up to 12.5% of lupin had an acceptable sensory performance, but lower than the control, pointing out that the texture might also be affected by the lupin variety

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Table 4. Details of sensory and/or instrumental evaluation of some products made with lupin flour or lupin isolated showed in Table 3

Used in	Details of evaluation	Reference
Bread	(100% WF). Specific volume (SV) 14.4; colour of crust (CC) 10; cutting consistency (CO) 4.0; symmetry (S) 4.0; colour of crumb (CR) 8; grain (G) 8; texture (T) 8; smell (S) 9, taste (Ta) 13. Total 74.8; Comparison 100%. (90% WF + 10% LF). SV = 13.2; CC = 9; CO = 3.5; S = 4.0; CR = 8; G = 8; T = 8; S = 8; Ta = 12. Total 72.7; Comparison 93%. (90% WF + 10% <i>L. albus</i> flour). SV=13.1; CC = 9; CO = 3.5; S = 5.0; CR = 6; G = 8; T = 7; S = 8; Ta = 12. Total 71.6; Comparison 91%. (90% WF + 10% Soya bean flour). SV = 7.0.1; CC = 9; CO = 3.5; S = 2.5; CR = 6; G = 7; T = 7; S = 8; Ta = 11. Total 61.0; Comparison 78%.	GROSS et al. (1983)
Spaghetti	(100% WS) Hardness 5.5 N; extensibility 2.8 cm. (97% WS + 3% <i>L. mutabilis</i> protein concentrate). Hardness 5.3 N; extensibility 1.4 cm.	LÓPEZ-SANTOS et al. (2006)
Spaghetti	(100% WS); maximum consistency (MC) in farinograph units (F.U) 520; radio resistance to deformation (R_{50})/extensibility (E) at 135 min (R_{50}/E) 6.50; cooking loss (CL) 7.50%; firmness of cooked spaghetti (F) 45.7 g. (5% ILP3, 95% WS); (MC) 500 (F.U); (R_{50}/E) 6.92; (CL) 8.71%; (F) 44.4 g. (20% ILP3, 80% WS); (MC) 470 (F.U); (R_{50}/E) 10.3; (CL) 16.93%; (F) 66.9 g.	DOXASTAKIS et al. (2007)
FRI	Hedonic evaluations. General acceptability (4.3/5).	CHÁVEZ & PEÑALOZA (1988)
FRA	Control (0% ILP2) first- bite hardness (FBH) = 10.8 kg; fracturability (Fr) 7.6 kg; chewiness (CH) 424; colour (C) 4.3/6; overall acceptability (OA) \approx 4.8/6. (1% ILP2). (FBH) = 8.6 kg, (Fr) 8.1 kg, (CH) 344, (C) 4.0 / 6, (OA) \approx 4/6. (2% ILP2). (FBH) = 7.0 kg, (Fr) 7.4 kg, (CH) 465, (C) 3.9 / 6, (OA) \approx 4/6. (3% ILP2). (FBH) = 4.5 kg, (Fr) 5.0 kg, (CH) 283, (C) 2.8/6, (OA) \approx 2/6.	ALAMANOU et al. (1996)
FER	Control Firmness (F) (Zwick units) 70.3; appearance (A) 5.3/6; colour (C) 5.5/6; taste and odour (T&O) 4.7/6. Fermented sausage (F) 66.6, (A) 5.1/6, (C) 5.8/6, (T&O) 5.2/6.	PAPAVERGOU et al. (1999)
Vanilla FLY	<i>L. mutabilis</i> yoghurt 5-points hedonic scale; (80 CM : 20 LM); aroma 4.1; taste 3.8; general acceptability 3.9. (70 CM : 30 LM) aroma 4.0, taste 3.6, general acceptability 3.6.	CASTAÑEDA-CASTAÑEDA et al. (2008)
FLY	<i>L. campestris</i> yoghurt 7-point hedonic scale; colour (C) 5.5; aroma (Ar) 5.5; flavour (Fl) 5.3; texture (T) 4.8; general acceptability (GA) 5.8. Control, cow's milk yoghurt (C) 6.2; (Ar) 6.2; (Fl) 6.4; (T) 5.9; (GA) 6.2.	JIMÉNEZ-MARTÍNEZ et al. (2003)
BJ	Control (0% LF) colour (C) (bright 100% of respondents; characteristic 9 5%); odour (O) (characteristic 96.2%); taste (Ta) (moderately sweet 72.5%, acid 45%); texture (T) (soft 57.5%). Jelly (7.5% LF): C (bright and characteristic 73.8%); O (characteristic 80%); Ta (moderately sweet 72.5%; acid 70%); T (soft 75%). Jelly (15% LF): C (matt 87.5% non characteristic 91.2%); O (non characteristic 67.5%); Ta (little sweet and acid 56.2%), T (hard 65%).	ROMERO & MEDINA (2004)

WF – wheat flour; LF – *L. mutabilis* flour; CF – cassava flour; WS – wheat semolina; ILP3 – isolated *Lupinus mutabilis* protein; FRI – fried lupin meat; FRA – Frankfurter sausages; ILP2 – isolated *L. albus* 'Graecus' protein; FER – fermented sausages; FLY – flavoured lupin yoghurt; CM – cow milk; LM – *L. mutabilis* milk; BJ – blackberry jelly

and the processes of obtaining the lupin flour, concentrated or isolated. For example, when obtaining these by-products, the ionic strength, pH and drying temperature will affect the properties of that by-product (*i.e.* solubility and emulsifying capacity) and, as a consequence, the behaviour of the dough matrix (CARVAJAL-LARENAS *et al.* 2015b). To improve the bread quality, researchers should try to include the lupin proteins as part of the dough matrix, perhaps as

colloidal dispersion or colloidal solution. Processes such as solvation, acylation, succinylation, enzymatic hydrolysis and protein denaturation are options that should be explored besides the matrix composition.

The results of these studies point out several conclusions. First, it seems that the increasing protein content might also imply to apply the increasing mixing time because that could engage better protein hydration and unfolding, facilitating the kneading

and later the dough strength. Second, the impact of starch (and amylose specifically) is important during pasting (gelling and recrystallization) by its influence on volume. Therefore, the amylose content is another ingredient that should be considered during formulation. Increasing the lupin (protein) content perhaps needs variations in the mixing and kneading time as well as in the amylose content.

Spaghetti and pasta

Lupinus mutabilis was reported by LÓPEZ-SANTOS *et al.* (2006) to be suitable to elaborate spaghetti. In this study, the authors used defatted isolated lupin protein to replace up to 3% of semolina. The best results were reported for 3% replacement with hardness similar to the control (0% isolated protein) but with the half extensibility of that in the control (Tables 3 and 4). This amount of substitution is similar to the value reported by DOXASTAKIS *et al.* (2007) in a study on white lupin. In this study, the authors made spaghetti with several blends of wheat semolina and white lupin isolated protein (Table 2). However, they found satisfactory results only up to 5% replacement. The instrumental evaluation of lupin blends in both studies shows an inverse relationship between the amount of added isolated lupin protein and dough development time, maximum consistency, tolerance index, elasticity and extensibility. This behaviour could be explained as a consequence of the gluten structure dilution by the increment of isolated lupin protein, which means the increment of protein content, mostly β -conglutin (7S globulin) and conglutin (11S globulin), which make the dough more compact and rigid.

On the other hand, these results contrast with those of LINSBERGER-MARTIN *et al.* (2010), who made pasta by replacing 50% and 100% of buckwheat with lupin, white bean (*Phaseolus vulgaris*) and pea (*Pisum sativum*) flours. Textures of bean and pea pasta were comparable to wheat pasta but different for lupin pasta. This difference might be due to the twofold protein content of lupins compared to peas and beans. On the other hand, peas and beans have the threefold starch content in comparison with lupins. As for the protein chemical composition, peas and beans have about 20% lower globulin content than lupin seeds. In addition, glutelins are present in peas and beans (12–15%) but they are not in lupins. Finally, the β -conglutin to α -conglutin ratio is close to 1 : 2 in peas and beans, and 1.3 : 1 in lupins (VAN BARNEVELD 1999). All these characteristics would make the lupin flour less elastic and extensible than pea and bean flours.

Meat substitute, meat balls, hamburgers and sausages

Meat substitute, also called meat analogue, imitation meat, mock meat, is the product that is structurally similar in texture, appearance, chemical composition and flavour to meat but of different composition (MALAV *et al.* 2015). Most of the meat analogues can be obtained by using high-moisture extrusion cooking (of slurries of wheat gluten, soy or pea protein), bioreactor-grown fungi and traditional high-protein preparations, such as fermented soy cake (tempeh), cooked wheat gluten (seitan) and pressed soy protein (tofu) (OSEN *et al.* 2014; JONES 2015).

In the case of *L. mutabilis* meat substitutes tempeh is the most common product (VILLACRÉS *et al.* 2006). In the tempeh products, debittered *L. mutabilis* was inoculated with *Rhizopus oligosporus* types 'NRRL 2710', 'Amsterdam' or 'BoBogor' in flour rice and then incubated. The macronutrient composition of the product can be seen in Table 2. Note that the product is a very good source of protein, fat, fibre, vitamins of B complex, phosphorus, and manganese. In addition, the authors reported that the fried product had a good taste similar to meat (Tables 3 and 4) and a 12-day shelf life under refrigeration. The soft texture, resulted from fermentation, could be ideal for elderly and young consumers.

L. mutabilis Sweet has also been used to elaborate meat balls and hamburgers (JACOBSEN & MUJICA 2006). In both cases whole lupin seeds are debittered, ground and mixed with meat and other ingredients (eggs and species). The *L. mutabilis* seeds replaced 50 and 56% of meat used in making the meat balls and hamburgers. The addition of eggs will have a functional purpose to act as a binder (MALAV *et al.* 2015), and to improve the nutritional value of hamburgers and meat balls by complementation.

These studies suggest that *L. mutabilis* meat substitutes could be used as an alternative to meat both from the nutritional composition and sensory point of view. In addition, and considering that these lupin products (tempeh, hamburgers and meat balls) are easy to make, they do not need any special infrastructure or technology, they could be fabricated easily and incorporated into human diet.

On the other hand, the lupin-meat substitutes still need research. Thus, the relationship between ingredient properties, matrix composition and rheological behaviour (chewiness, hardness, texture, palatability, etc.) remains practically unknown. Moreover, it will

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be worth to investigate lupin-meat substitutes using high-moisture extrusion cooking, extensional flow or high pressure because these technologies would lead to stabilization of the three-dimensional network and produce a meat analogue with better meat-like texture (OSEN *et al.* 2014).

Lupins have also been used to prepare sausages. For example *Lupinus albus* 'Graecus' isolated protein added up to 3% of the product was used to increase the protein content of frankfurter sausages (ALAMANOU *et al.* 1996) (Table 2). Sensory evaluation showed that sausages made with 1% or 2% of protein isolates were liked by the judges and scored both higher than the control (fermented sausages) and lower than the control (without fermentation) (Tables 3 and 4). But they did not like the product made with 3% of protein isolate. The addition of 1% or 2% of isolated lupin protein to the sausage formula could be considered low. However, the importance of such addition is founded on the replacement capacity that these amounts of isolated lupin protein have over the amount of used meat and this in turn is based on water absorption capacity and emulsifying capacity of specific lupin isolates – up to 6 g of water/g DM and 2 l of oil/g of lupin protein, respectively (CARVAJAL-LARENAS *et al.* 2015b). For example, based on the protein content of meats and other ingredients PAPAVERGOU *et al.* (1999) were able to replace 95 g of a meat mixture (beef 25%, pork 45% and pork backfat 30%) with 20 g of *L. albus* isolated protein plus 74 g of water per kilogram of fermented sausages keeping the equal protein content (Table 2).

The replacement (total or partial) of meat by lupin would also be important because that could improve the fat profile and would reduce the cholesterol content of the diet (BERTI *et al.* 2013).

Lupin milk and yoghurt substitutes

Lupin milk substitutes have been prepared from *L. mutabilis* and *Lupinus campestris* by mixing chopped debittered lupin and water at a ratio 1:2–9 kg/l (JIMÉNEZ-MARTÍNEZ *et al.* 2003; VILLACRÉS *et al.* 2006; CASTAÑEDA-CASTAÑEDA *et al.* 2008). Then, the mix is filtered and the aqueous fraction is used to prepare lupin milk. Sugar, flavour, stabilizer, isolated lupin protein and vegetable fat have been added to improve the sensory attributes and to reach a composition similar to whole or skimmed cow's milk. From 1 kg of fresh and debittered lupin were obtained 2.2 litres

of lupin milk (VILLACRÉS *et al.* 2006). The chemical composition and sensory evaluation can be seen in Tables 2 and 3. Lupin milk had a good organoleptic evaluation but only when it was flavoured.

Regarding composition and stability, they seem adjustable by added materials and process. For example CARVAJAL-LARENAS *et al.* (2015b) suggested that if water pH is about 8–10 with an ionic strength of 1 (adjusted with sodium chloride), the lupin protein solubility could be enhanced because the electrostatic repulsion will be high at those conditions and the net result will be an increment of product performance. Lupin milk substitutes have also been used to obtain yoghurt-like products. In order to do so, the lupin milk was enriched with powder (cow) milk and *L. mutabilis* protein isolate (VILLACRÉS *et al.* 2006) or it was enriched with lactose and sucrose (JIMÉNEZ-MARTÍNEZ *et al.* 2003). Then, the mix is pasteurized and inoculated with *Streptococcus thermophilus* and *Lactobacillus delbrueckii* 'bulgaricus', and fermented as a cow milk mix.

The macronutrient composition of lupin yoghurt-like product is shown in Table 2. Note that this composition is similar to that reported by CASTAÑEDA-CASTAÑEDA *et al.* (2008), who also made yoghurt from mixtures of 80% (70%) of cow's milk and 20% (30%) of *L. mutabilis* milk.

The sensory evaluation of the unflavoured lupin yoghurt showed that it was unacceptable and tasteless. However, after flavouring, the taste turned to be between like a little and like much (Tables 3 and 4) but worse than that of cow's milk yoghurt (JIMÉNEZ-MARTÍNEZ *et al.* 2003) showing that taste affects the liking directly (PALA & ATAKISI 2012). Consistency is another important characteristic of yoghurt that depends on the matrix capability to absorb water and to form a stable gel. The lupin yoghurt-like consistency was reported to be similar to cow's milk yoghurt (JIMÉNEZ-MARTÍNEZ *et al.* 2003). This consistency and its stability would be based on the effect of pH, heat and solids on the lupin yoghurt-like product in a similar way like they do on other systems. For instance, in cow-milk yoghurt, protein fortification (with bisulphite) and heat treatments are cited as the most important features to reach a good consistency (AKALIN *et al.* 2012) because of the protein aggregation and disulphide bonding (CARVAJAL-LARENAS *et al.* 2015b) in addition to the protein aggregation by isoelectric pH. In addition and in order to improve consistency, it has also been suggested to set up total solids content (to 12–14 g/100 g), protein content (up

to 40–50 g/ kg) and the addition of calcium caseinate and sodium caseinate (AKALIN *et al.* 2012). Moreover, the process used to obtain the lupin isolate could also affect the gel properties. For example, it is stated that the lupin isolate obtained at the acidic side of isoelectric pH helps to form stable gels of globulins because at this pH carboxylic groups are less dissociated and interactions between protein molecules and water are increased (CARVAJAL-LARENAS *et al.* 2015b). In addition, it has been proposed in soy bean that removing phenolic compounds from bean could improve the gel texture because phenolics interfere with non-covalent networking interactions between proteins (JONES 2015). Finally, it should be considered that lupin has peptic substances (β -1,4-galactan) which could also improve the texture (VAN BARNEVELD 1999). Then, in order to control the consistency of lupin yoghurt-like products all these features should be taken into account.

Researchers are encouraged to study meticulously the elaboration, characterization and standardization of lupin milk and yoghurt substitutes because this can help to improve the nutritional status of the population that cannot include cow's milk in their diets, for example those with high cholesterol levels. This kind of products could also be suitable for those who live in areas where cow's milk production is not possible.

Functional lupin products

L. mutabilis has also been used to prepare food for special groups of population. For example, ROMERO and MEDINA (2004) and VILLAROEL *et al.* (1996) prepared lupin jellies using between 0 and 15% of lupin flour. The use of lupins in making jellies is important because this product might generate a reduction of postprandial glucose in people with non-insulin dependent diabetes (VILLAROEL *et al.* 1996).

The chemical composition and sensory evaluation of some lupin jellies are shown in Tables 2 and 3. Note that, as expected, the addition of lupin flour increased substantially the protein content of jellies when compared with the control.

As for the sensory evaluation, the best ranked lupin jellies were those with a replacement level between 5 and 10%. However, these jellies scored lower than the control (ROMERO & MEDINA 2004). The authors did not report using pectins and acids nor did they have any inconveniences with the gelling process; that might be because of both the probable protein unfolding carried out during the

cooking, and the acid pH given by the fruit used (blackberry and plum) contributing to increase the binding interactions and therefore forming stable gels. In addition, the values of lupin flour used in these studies agreed with the last gelation concentration of lupin flours reported in literature (between 6 and 14%) (CARVAJAL-LARENAS *et al.* 2015b).

Another possible functional product prepared with *L. mutabilis* is a sort of purée that could be used for people with celiac disorders (especially babies). In this study, lupin was mixed with oca (*Oxalis tuberosa*) and white carrot (parsnip) (*Arracaccia xanthorriza*) (LEÓN-MARROÚ *et al.* 2011). The authors prepared seven (gluten-free) mixtures and found that the combination of oca, white carrot and lupin (25 : 25 : 50) had the best protein content (Table 2).

The results of these studies agree with others that show that mixing lupin with other grains, cereals and foods increases the nutritional content of the mixtures by complementation (LEÓN-MARROÚ *et al.* 2011; BERTI *et al.* 2013). This research also agrees with other studies that show that *L. mutabilis* might have nutraceutical applications that should be investigated deeply (VALLEY & SIPSAS 2010; BALDEÓN *et al.* 2012).

CONCLUSIONS

Nowadays, lupins in general are already used to obtain lupin-based products. However, the *Lupinus mutabilis* variety is perhaps the only one employed to elaborate both meals and processed products. Some reasons would explain its wide use: (1) its chemical composition (especially high protein content), (2) lupin is cheaper than meat and (3) this seed has good taste which is better than that of soya bean. These features would help to obtain a good nutritional product, tasty and at low cost, which in turn could contribute to improve the nutritional status of the lower-income population (compared for instance to meat-based products). This is an important insight that could justify the production of lupin products/dishes. However, and in order to do that, this research found some information gaps that need to be filled in as follows:

In all studied products, the chemical composition (protein content) was improved when adding lupin. It is also expected that substitution of animal fat by lupin fat improves the product fat profile. However, it was hardly possible to find studies that show chemical composition in more details. For example, protein or fat profile of elaborated product.

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As for the formulations and procedures, there is a need to develop research that will reach a deep understanding of the relationship between the procedure to obtain lupin by-products (flour, protein isolate or concentrate), their composition (specially protein, fat and fibre content), other ingredients and their influence on the spatial configuration of the matrix mix, its physical properties and rheological behaviour. The generated knowledge will be useful for the development of better products.

Moreover, hardly any of the studies determined the shelf life of products and no study has been devoted to its relationship to the chemical composition and/or procedure used to obtain that product.

As for the sensory evaluation and rheological behaviour there is valuable but limited information in most studies that lupin products scored lower or slice lower than control products. Just few products scored better. In addition, most studies about sensory and rheological behaviour only make one of them, and just very few included both. This means that there is plenty of room for setting up studies on sensory and rheological behaviour jointly.

Regarding the nutritional composition, there is limited information about lupin products. Most of the studies only report chemical composition and/or caloric density. Therefore, there is a lack of deeper information about nutritional quality of lupin products, *i.e.* true digestibility, protein efficiency ratio, protein digestibility, corrected amino acid score and biological value. In addition, the nutraceutical behaviour of lupin products remains practically unknown.

In short, lupin products have an enormous potential opening the space for a plenty of possibilities to conduct research about lupin products.

Recommendations to improve the nutritional value of lupin products

The nutritional quality of lupin products could be improved by fortification with methionine (KOHAJDOVÁ *et al.* 2011; CARVAJAL-LARENAS *et al.* 2015b). Thus, the protein efficiency ratio (PER) of *L. mutabilis* has been reported to increase from about 1 to that of casein (2.5) (PETERSON 1998) by adding 0.2% DL-methionine. In addition, the PER value of lupin products could be increased if the formula contained complementary protein carriers rich in sulphur-containing amino acids (KOHAJDOVÁ *et al.* 2011; CARVAJAL-LARENAS *et al.* 2015b) such as cereal proteins, fish products, and hen (whole) eggs.

Cooking could also improve the nutritional quality of lupin derivatives (BALDEÓN *et al.* 2012). Moreover,

the addition of specific lupin derivatives might have health benefits. For example, lupin phytochemicals may be responsible for the beneficial cardiovascular effects (KOHAJDOVÁ *et al.* 2011) and lupin γ -conglutination might reduce the postprandial glucose (BALDEÓN *et al.* 2012).

Recommendations to improve sensory properties of lupin products

VILLACRÉS *et al.* (2000) and CARVAJAL-LARENAS *et al.* (2015a) showed that sensory properties of debittered lupin seem to be affected by the processing (debittering) conditions. Changing the water three times a day would be the preferred treatment (CARVAJAL-LARENAS *et al.* 2015a). Fermentation combined with frying would also improve the taste and texture of lupin-meat, making it very similar to fried beef (VILLACRÉS *et al.* 2006). Fermentation would increase the protein content and would have a proteolytic effect (VILLACRÉS *et al.* 2006) while the frying process would develop a browning effect. Texture seems to be also adjustable by controlling the formula (*i.e.* protein content and its composition, solids, starch and methionine content, pH, ionic force) and by controlling the procedure (the type of extruder, mixing times and processing temperatures). Combination of lupin with other foods (*i.e.* shrimps, onions, tomato, tuna fish, etc.), spices and flavours enhances the lupin acceptance.

Recommendations to improve rheological properties of lupin products

Rheological behaviour of lupin products can be affected by protein-containing and non-protein ingredients (GÜEMES-VERA *et al.* 2008), processing conditions and technology used. In addition, the processes of defatting lupin, concentration and isolation of its proteins (pH, heat, ionic strength) as well as chemical and enzymatic treatments can modify the protein structure at different levels affecting the rheological behaviour of mixtures and products. Nevertheless, in order to improve the rheological quality of lupin products some general recommendations can be made – when compared with the control – as follows:

Lupin bread volume might be enhanced by controlling (increasing) the fermentation time (and mixing time), and adding (increasing) starch (specifically amylose) content. In addition, processes such as solvation, acylation, succinylation, enzymatic hydrolysis and protein

denaturation are also options that would help to increase lupin bread quality.

The texture of lupin spaghetti and pasta might also be improved by monitoring the total protein content (keeping it similar to the control, at least at the beginning) and simultaneously both decreasing the globulin content and increasing starch and glutelin content. This can be done by mixing lupin by-products with pea and bean by-products, and/or with other legumes or cereals, or mixing directly lupin by-products with glutelin isolates and starch.

The quality of meat analogues could be improved by using extensional flow, high-pressure processing and high-moisture extraction cooking, since these technologies would produce muscle-like textures (OSEN *et al.* 2014; JONES 2015).

As for the milk analogue, the solubility of lupin proteins might be enhanced if the protein extraction is done at pH 8–10 with an ionic strength of 1. In addition, in order to increase the yoghurt analogue consistency, it would help to improve its protein quality (by adding bisulphite). Moreover, the solids content in milk analogue used to make yoghurt should be at about 14% (being 4 or 5% of that value protein). In addition, the addition of calcium or sodium caseinate would improve the quality of the product.

Finally, the complexity of the seed proteins would require efforts in understanding the extraction processes and characterization of grains of individual protein fractions (JONES 2015) that might have specific applications. For example, it seems that the addition of specific amounts of: (1) lupin globulins (α - and β -conglutin) might increase crunchiness, and (2) starch might increase elasticity. Moreover, the extraction of phenolic compounds in lupin by-products might alter the elasticity of lupin product as this process has done in soy and flaxseed (OSEN *et al.* 2014) products.

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