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# Germinated *Cajanus cajan* seeds as ingredients in pasta products: Chemical, biological and sensory evaluation

Alexia Torres<sup>a</sup>, Juana Frias<sup>b</sup>, Marisela Granito<sup>a</sup>, Concepción Vidal-Valverde<sup>b,\*</sup>

<sup>a</sup> Simón Bolívar University, Department of Food Science, 1080A Caracas, Venezuela

<sup>b</sup> Instituto de Fermentaciones Industriales (C.S.I.C.), Juan de la Cierva 3, 28006 Madrid, Spain

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#### Abstract

Pigeon peas (*Cajanus cajan*) seeds were germinated for 4 days at 20 °C in darkness in order to improve the nutritional quality of seeds. Germination brought about a sharp reduction of  $\alpha$ -galactosides, phytic acid and trypsin inhibitor activity (83%, 61% and 36%, respectively) and an increment of vitamin B<sub>2</sub> (145%), vitamin C (from negligible amounts to 14 mg/100 g d.m.), vitamin E (108%) and total antioxidant capacity (28%). These flours were used as ingredients to produce pasta products in a proportion of 5%, 8% and 10%. The supplemented pasta products had shorter cooking time and higher water absorption, cooking and protein losses in water than had control pasta (100% semolina). From sensory evaluation, fortified pasta generally had acceptability similar to control pasta. Cooked pasta with the highest level of substitution (semolina:germinated pigeon pea flour at 10%) was chemically and biologically evaluated and results showed that protein, fat, dietary fibre and mineral contents were improved. Fortified pasta provided more vitamin B<sub>1</sub>, B<sub>2</sub>, E and antioxidant capacity than did control pasta. Biological assessment of fortified, cooked pasta indicated that true TD and PER value increased by 12% and 64%, respectively, in comparison with control. The germinated pigeon pea flour can be an excellent ingredient to increase the nutritional value of semolina pasta without affecting the sensory properties.

Keywords: Pigeon pea; Germination; Fortified pasta; Sensory evaluation; Nutritional quality

## 1. Introduction

Germinated dry beans are receiving increasing attention due to enhanced flavour and nutritional qualities, particularly throughout the breakdown of certain antinutrients, such as phytate, and flatulence factors (Ghorpade & Kadam, 1989). The process involves complex changes that propitiate the breakdown of macromolecules, which increases the rate of starch and protein digestibility, improving the content of amino acids and, consequently, this results in more digestible foodstuffs and it is one of the reasons why legume sprouts have been used in the preparation of legume-based, low cost weaning foods (Martin-Cabrejas et al., 2003; Urooj & Putt-

E-mail address: ificv12@ifi.csic.es (C. Vidal-Valverde).

araj, 1994). Germination is also known to improve the vitamin and mineral contents. It has been reported that vitamin C and riboflavin are synthesized during germination (Henry & Massey, 2001).

Pigeon pea (*Cajanus cajan*), among legumes, has an important place in the diet of many people in the world. It is one of the oldest food crops. India alone contributes over 90% of the world pigeon pea production. It is also a food crop in many other tropical countries and is commercially important in East Africa, the Caribbean and Latin America. It has low concentrations of fat, moderate amount of fibre, good amount of proteins and starch and a reasonably balanced range of all dietary essential minerals. Besides these nutrients, pigeon peas also contain antinutritional factors, such as phytic acid,  $\alpha$ -galactosides, tannins, saponins and trypsin inhibitors (Duhan, Khetarpaul, & Bishnoi, 2001; Salunkhe, Chavan, & Kadam, 1986).

<sup>\*</sup> Corresponding author. Tel.: + 34 915622900x241; fax: + 34 915644853.

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Pasta products are well accepted worldwide because of their low cost, ease of preparation, versatility, sensory attributes and long shelf life (Bergman, Gualberto, & Weber, 1994). Although pasta is traditionally manufactured using only durum wheat flour, it is possible to use non-durum wheat ingredients to produce specifically blended pasta (Brennan, Kuri, & Tudorica, 2004). Pasta products have been fortified to enhance their nutritional properties with supplements from various high-protein sources, such as fish protein concentrates, soy flours, soy isolates, milk and milk products, cottonseed meal, egg albumin, whey proteins and yeast protein concentrates (Nielsen, Summer, & Whalley, 1980). Torres, Frías, Granito, Guerra, and Vidal-Valverde (2006) developed pasta products with  $\alpha$ galactoside free lupin flours and found an increase in their nutritional quality. Salunkhe et al. (1986) reported synergistic effects of pigeon pea, on the protein quality of various cereals. The incorporation of pigeon pea to the extent of 15%, in cereal diets based on ragi, kaffir corn, pearl millet or wheat improved the PER of cereal diets.

The aim of the present study was to increase the nutritional value of pigeon peas by germination and to incorporate the obtained pigeon pea sprout flour to supplement durum semolina wheat flour by making pasta products with higher nutritive value than the pasta without supplementation.

## 2. Materials and methods

# 2.1. Legumes

Pigeon pea seeds (*Cajanus cajan*, var. Aroíto), were obtained from The National Agricultural Research Institute, Yaracuy, Venezuela.

# 2.2. Germinated seeds

Pigeon peas were germinated, following the procedure described by Frías, Miranda-Zarate, Doblado, and Vidal-Valverde (2005). Three hundred grams of clean seeds were soaked in sodium hypochlorite solution (0.07%, w/v) in a relationship of 1:5 (w/v), in dark conditions for 30 min at room temperature. After that time, seeds were washed (until neutral pH) and soaked in distilled water (1:5, w/v) in dark conditions for 5 and a half hours at room temperature. The water was then drained off and samples were germinated in a climatic cabinet (ASL Snijders Sci. International S.L., Tiburg, Holland) at 20 °C for 4 days in darkness. Germination was carried out in triplicate. The sprouts were freezedried (Labconco freeze dryer, Missouri, United States) and ground to 0.5 mm particle size for analysis.

# 2.3. Pasta preparation

The following pasta products, with different levels of germinated pigeon pea flours (GPPF), were prepared.

- Semolina 100% (control)
- Semolina: GPPF (95:5)
- Semolina: GPPF (92:8)
- Semolina: GPPF (90:10)

Pasta products were prepared in triplicate, as follows: homogenized flours were mixed with water to a moisture content of 31.5% and the mix was blended (Kitchen Aid classic model) for 2 min and stood for 15 min. Following this, dough was stretched and extruded (single screw pasta machine Columbus, model Marchio Depositati) and spaghetti was formed (on laboratory scale), pre-dried at room temperature for 1 h and then dried (convection forced air oven) at 50 °C for 2 h. Dry pasta was ground to 0.5 mm and kept until analyzed.

## 2.4. Chemical analysis

#### 2.4.1. Water, nitrogen, fat and ash content

These analyses were carried out according to AOAC (1990). Conversion factor (from nitrogen content to protein content) was 6.25.

# 2.4.2. Mineral determination

Specific minerals presented in the pasta products were analysed by inductively coupled plasma-atomic emission spectroscopy (ICP-AES), according to AOAC (1990) method 984.27.

## 2.4.3. Soluble carbohydrate content

Monosaccharides, disaccharides and  $\alpha$ -galactosides were determined by HPLC, following the procedure described by Granito et al. (2002).

# 2.4.4. Starch content

The content of starch in raw and germinated pigeon pea flour was determined as in Sotomayor et al. (1999).

#### 2.4.5. Energy value

The energy value of legume seeds and pasta products was calculated by the Atwater system (Watt & Merrill, 1963). Factors applied to ingested nutrients were: 4 kcal/g for protein and carbohydrates and 9 kcal/g for fat.

## 2.4.6. Dietary fibre content

Soluble, insoluble and total dietary fibre contents were determined according to Prosky, Asp, Schweiser, De Vries, and Furda (1988).

#### 2.4.7. Vitamin content

Vitamins  $B_1$  and  $B_2$  were determined by HPLC according to Prodanov, Sierra, and Vidal-Valverde (1997). Vitamin C was determined by capillary electrophoresis, following the method described by Frias et al. (2005). Tocopherol isomers and vitamin E activity were determined by HPLC according to Frias et al. (2005).

# 2.4.8. Tannin content

The content of tannins was determined by the Folin– Ciocalteu method, as in Kuiters (1987). Galic acid (Sigma Chemicals) was used as standard.

## 2.4.9. Total antioxidant capacity

The antioxidant capacity was determined as trolox equivalent antioxidant capacity (TEAC) according to Re et al. (1999) modified by Frias et al. (2005).

## 2.4.10. Inositol phosphate content

Inositol phosphates [hexa- (IP<sub>6</sub>), penta- (IP<sub>5</sub>) and tetrainositol phosphate (IP<sub>4</sub>)] were extracted according to Kozlowska, Honke, Sadowska, Frías, and Vidal-Valverde (1996) and the quantification was carried out by HPLC according to Lerhfeld (1994).

#### 2.4.11. Trypsin inhibitor activity (TIA)

TIA was determined as in Vidal-Valverde et al. (1997).

## 2.4.12. Pasta cooking quality

Parameters related to the pasta cooking quality were determined according to Abecassis, Faure, and Feillet (1989) and Matsou, Malcomson, Edwards, and Dexter (1992).

- (a) Cooking time: 10 g of pasta product were dispersed in 100 ml boiling water. From time to time a piece of pasta was held between two glass plates and was compressed. Optimum cooking time (min) was established when no white core was observed after the compressing.
- (b) *Cooking water absorption*: The cooked pasta was drained and weighed to determine water absorption.
- (c) Cooking loss: solids extracted from the cooking water were calculated by concentrating the cooking water to dryness in an oven at 100 °C.
- (d) *Protein loss in water*: This was determined in the cooking water by the Biuret method, as described by Robinson and Hodgen (1940).

#### 2.4.13. Sensorial analysis

Pasta products were cooked in boiling water without addition of salt, drained and kept in warm conditions prior to testing. A 19-member semi-trained panel evaluated pasta products for overall quality (colour, texture and flavour). First, panellists were asked to evaluate the pasta products and choose the acceptability on a non-structured scale (15 cm) (left extreme, 1 = extremely unacceptable and right extreme, 15 = extremely acceptable) (Larmond, 1977). After this evaluation, panellists evaluated differences of supplemented pasta in reference to the control in parameters such as colour, flavour and texture, employing a structured hedonic test (scale from 1 = extreme difference by inferiority and 7 = extreme difference by superiority; control pasta had a fixed score of 4).

#### 2.4.14. Biological analysis

Protein true digestibility (TD) and protein efficiency ratio (PER) were determined according to Allison (1955). The experimental diets were prepared by adding the appropriate amount of cooked pasta products (semolina 100% or semolina fortified with GPPF) to commercial maize starch to provide 10% protein content to the diet. Minerals mix (3.5% from Harland Teklad), maize oil (5.0%), vitamin mix AIN-76 A (1.0% from Harland Teklad) and choline bitartrate (0.2% from Sigma C-1629) were also added. Six-week old Sprague–Dawley rats, weighing about 50 g, were selected, at random by weight, into 5 groups of 6 animals (3 male and 3 female) and housed in individual cages in an environmentally-controlled room. The animals were fed with a control diet from weaning, until the study began. During a 14-day testing period, experimental diets, a protein free diet, a casein diet and water were supplied. Every second day, the animals and their feed intake were weighed. In order to calculate protein digestibility, the faeces of each animal were collected during the last 7 days of the experiment. Nitrogen consumed by each animal for this period was quantified. The faeces were oven-dried at 100 °C for 24 h. The dried samples were ground to 20 mesh. The experimental diet and fecal powder were analyzed for protein  $(N \times 6.25)$  by the micro-Kjeldahl method.

## 2.4.15. Statistical analyses

All data were expressed as means  $\pm$  standard deviations and were subjected to a multiple comparison test with the use of the least significant difference test. Statistical analyses were performed using the Statgraphic 5.0 programme for Windows (Statistical Graphics Corporation, Rockville, Md. USA).

# 3. Results and discussion

Nutrient composition of raw and germinated pigeon pea flours is presented in Table 1. Protein, fat and total starch content were not significantly ( $P \leq 0.05$ ) affected by the germination process. However, available starch, soluble and insoluble dietary fibre and ash content decreased significantly ( $P \le 0.05$ ) by 10%, 62%, 35% and 19%, respectively, after pigeon pea germination. Also, important minerals such as calcium, potassium, zinc and iron, also showed a significant decrease. By contrast, a noticeable increase ( $P \leq 0.05$ ) in total available soluble carbohydrates (glucose, fructose and sucrose) and in resistant starch was obtained. Glucose, a monosaccharide absent in the raw seeds, was detected in germinated pigeon pea flours in a relative high amount (4.66%). Fructose showed an increment of a 1276% whilst sucrose rose by 15%. As a consequence of germination, food energy value also increased significantly ( $P \le 0.05$ ) (12%).

Ghorpade and Kadam (1989) reported that germination had very little effect on the crude protein content in many legumes. They found, in moth beans and horse gram, a progressive increase in the free amino acid content and also

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Table 1

Germination effect on chemical composition of pigeon pea seeds (*Cajanus cajan* var. Aroíto)

Composition	Raw pigeon pea	Germinated pigeon pea
Protein <sup>A</sup>	$29.3\pm0.24^{\rm a}$	$28.9\pm0.10^{\rm a}$
Fat <sup>A</sup>	$2.36\pm0.38^{\rm a}$	$2.19\pm0.10^{\rm a}$
Glucose <sup>A</sup>	nd <sup>a</sup>	$4.66\pm0.07^{\rm b}$
Fructose <sup>A</sup>	$0.29\pm0.04^{\rm a}$	$3.99\pm0.11^{ m b}$
Sucrose <sup>A</sup>	$3.87\pm0.01^{\rm a}$	$4.46\pm0.14^{\rm b}$
Total available soluble sugars <sup>A</sup>	$4.16\pm0.04^{\rm a}$	$13.16\pm0.15^{\rm b}$
Available starch <sup>A</sup>	$39.5\pm1.31^{\text{b}}$	$35.6\pm0.36^{\rm a}$
Resistant starch <sup>A</sup>	$2.80\pm0.60^{\rm a}$	$7.24\pm0.48^{\rm b}$
Total starch <sup>A</sup>	$41.3\pm1.12^{\rm a}$	$42.6\pm0.61^{\rm a}$
Insoluble dietary fibre <sup>A</sup>	$34.9\pm1.02^{b}$	$22.8\pm1.58^{\rm a}$
Soluble dietary fibre <sup>A</sup>	$4.23\pm0.69^{b}$	$1.60\pm0.06^{\rm a}$
Total dietary fibre <sup>A</sup>	$39.1 \pm 0.32^{\mathrm{b}}$	$24.4\pm1.52^{\rm a}$
Ash <sup>A</sup>	$3.99\pm0.07^{\rm b}$	$3.24\pm0.04^{\rm a}$
Calcium <sup>B</sup>	$201\pm1.52^{\rm b}$	$159\pm1.40^{\rm a}$
Sodium <sup>B</sup>	$89.7\pm1.55^{\rm a}$	$88.2\pm4.27^{\rm a}$
Potassium <sup>B</sup>	$1290\pm41.0^{\rm b}$	$1245\pm2.98^{\rm a}$
Magnesium <sup>B</sup>	$110.49 \pm 2.64^{ m b}$	$94.7\pm0.60^{\rm a}$
Zinc <sup>B</sup>	$7.85\pm0.05^{\rm b}$	$4.41\pm0.05^{\rm a}$
Iron <sup>B</sup>	$5.30\pm0.15^{\rm b}$	$3.94\pm0.03^{\rm a}$
Energy (Kcal/100 g)	320.00	358.19
Vitamin B1 <sup>B</sup>	$0.40\pm0.02^{\mathrm{b}}$	$0.22\pm0.03^{\mathrm{a}}$
Vitamin B2 <sup>B</sup>	$0.31\pm0.02^{\rm a}$	$0.76\pm0.01^{ m b}$
Vitamin C <sup>B</sup>	nd <sup>a</sup>	$13.9\pm1.65^{\mathrm{b}}$
α-Tocopherol <sup>B</sup>	$1.06\pm0.03^{\rm a}$	$2.50\pm0.16^{\rm b}$
β-Tocopherol <sup>B</sup>	$0.06\pm0.01^{\mathrm{b}}$	$0.04\pm0.01^{\mathrm{a}}$
γ-Tocopherol <sup>B</sup>	$9.31\pm0.34^{\rm a}$	$16.3 \pm 0.17^{\rm b}$
δ-Tocopherol <sup>B</sup>	$0.27\pm0.01^{\rm a}$	$1.96\pm0.02^{\rm b}$
α-TE Vitamin E	$2.02\pm0.03^{\rm a}$	$4.21\pm0.17^{\rm b}$
(Units/100 g d.m)		
Tannins (galic acid) <sup>A</sup>	$0.39\pm0.02^{\rm b}$	$0.32\pm0.02^{\rm a}$
TEAC (µmol trolox/g)	$33.2\pm0.67^{a}$	$42.6\pm1.18^{\rm b}$

nd = non detected.

The same superscript in the same raw means no significant difference  $(P \le 0.05)$ .

<sup>A</sup> Values in g/100 g dry matter.

<sup>B</sup> Values in mg/100 g dry matter.

a better protein digestibility due to the hydrolysis. King and Puwastien (1987) reported a small decrease in protein nitrogen after a 72 h germination of winged beans and an increase in non protein nitrogen and free amino acids. Sangronis, Camacho, and Cava (2004) found that germination had no effect on the protein content of seeds of Phaseolus vulgaris and Cajanus cajan in contrast with raw seeds. Kylen and McCready (1975) and Camacho, Sierra, Campos, Guzmán, and Marcus (1992) found a decrease in ash and fat contents after germination of chickpeas, peas, beans, lentils and alfalfa. Kavas and Nehir (1992) and Oloyo (2004) reported significant increases in mineral content. Our results showed that the mineral content of cowpea sprouts were lower than those of the raw seeds. This loss could be attributable to leaching of these micronutrients due to the hydration system applied. Similar results were reported by Khalil and Mansour (1995) after germination of faba bean. Khalil (2001), in germinated guar faba bean found a noticeable decrease in sodium, manganese and magnesium but a slight increase in iron and zinc in comparison with raw seeds. Kylen and McCready (1975) found that glucose and fructose increased ten-fold and sucrose doubled after germination of alfalfa, lentils, mung beans and soybeans. Vidal-Valverde et al. (2002) also reported an increase in total available sugars after germination of beans, lentils and peas. Oloyo (2004) found a negative correlation between duration of germination and significant reduction of soluble carbohydrates. Soluble sugar rise was a consequence of hydrolysis of oligosaccharides which are mobilized and used during the process of germination of the seeds (Dey, 1990).

Starch values obtained in this work coincide with those reported by Veena, Urooj, and Puttaraj (1995). These authors also reported that germination negatively affected the soluble dietary fibre fraction. Chitra, Singh, and Rao (1996) showed a decrease of total dietary fibre after germination of chickpeas, pigeon peas, mung beans and soybean. Ologhobo and Fetuga (1986) reported decreases in hemicellulose and cellulose content in germinated cowpeas. Differently, Martin-Cabrejas et al. (2003) reported an increase in dietary fibre of peas after germination.

Table 1 also collects the content of some vitamins in raw and germinated pigeon peas. Germination leads to a marked decrease ( $P \le 0.05$ ) in thiamine (45%) whilst riboflavin increased significantly (145%) ( $P \le 0.05$ ). Vitamin C was not detected in raw pigeon pea and germination for 4 days brought about the presence of a large amount (14 mg/ 100 g d.m.). According to Messina (1999), vitamin C content in germinated seeds could increase iron absorption. The vitamin E activity also increased after germination (108%) that was mainly due to the rise observed in  $\alpha$ -,  $\delta$ - and  $\gamma$ -tocopherols, whilst  $\beta$ -tocopherol, decreased significantly ( $P \le 0.05$ ).

Taking in consideration that the RDAs of vitamin  $B_1$ , B<sub>2</sub>, vitamin C and vitamin E are 1.5 mg/day, 1.7 mg/day, 60 mg/day and 10 mg/day, respectively, for adult men and 1.1 mg/day, 1.3 mg/day, 60 mg/day and 8 mg/day, respectively, for adult women (RDA, 1989), the germinated seeds would supply 15% and 20% of thiamin, 45% and 58% of riboflavin, 23% of vitamin C and 42% and 53% of  $\alpha$ tocopherol of RDAs for men and women, respectively. Nnanna and Dixon (1989) reported an increase in thiamin and riboflavin contents after germination of cowpea, probably because of the liberation of these vitamins' hitherto complexes with proteins. Tannin content was slightly affected by the germination process and a decrease of 18% was observed (Table 1). Our results agree with those reported by Khalil and Mansour (1995), who found that germination had a negligible effect on the reduction of tannins in faba bean seeds, while Verma and Mehta (1988) reported a significant reduction in tannin content after sprouting of rice bean and mung bean. Losses of tannins in beans during germination may be attributable to the presence of polyphenol oxidase and enzymatic hydrolysis can occur. Total antioxidant capacity, measured as TEAC, increased significantly ( $P \leq 0.05$ ) (28%) after germination, in agreement with results reported by Frias et al. (2005) in lupin seeds during germination.

Table 2 shows the content and composition of  $\alpha$ -galactosides, inositol phosphates and trypsin inhibitor activity for raw and germinated pigeon peas. Raw seeds presented raffinose, stachyose and verbascose and germination brought about a drastic decrease of these  $\alpha$ -galactosides (79, 81 and 87%, respectively). These oligosaccharides decline rapidly during the first days of germination and the decrease in the level of these oligosaccharides during germination was attributed to the  $\alpha$ -galactosidase activity (Lower & Kou, 1989). During germination, a mobilization of reserve nutrients occurs and  $\alpha$ -galactosides are used as a source of energy (Dev, 1990). Sathe and Salunkhe (1984) reported a reduction of 90% of verbascose, stachyose and raffinose in chickpeas after 4 days of germination. Ghorpade and Kadam (1989) indicated that over 70% of the raffinose family of oligosaccharides could be removed during germination. Vidal-Valverde and Frías (1992) reported the total elimination of raffinose, stachyose and ciceritol in lentils after 96 h of germination. Vidal-Valverde, Frías, Lambein, and Kuo (2001) found, after 6 days of germination of lentils and beans, the total removal of  $\alpha$ -galactosides, and the presence of soluble sugars. Vidal-Valverde et al. (2002) also reported the complete elimination of ciceritol and stachyose in lentils after 2 days of germination and after 4 days in beans and peas. Raffinose was completely eliminated in lentils and beans after 6 days of germination. Similarly, Siddhuraju and Becker (2001) found a 90% reduction of  $\alpha$ -galactosides in white and black varieties of mucuna beans after 72 h of germination. The remaining  $\alpha$ -galactosides could act as prebiotic agents in prepared food, when germinated pigeon peas are used as ingredients.

Changes in inositol phosphate content during germination are collected in Table 2. In raw seeds the presence of inositol hexa- (IP<sub>6</sub>), penta- (IP<sub>5</sub>) and tetra- (IP<sub>4</sub>) phosphates was observed. IP<sub>6</sub> represented 64% of the total inositol phosphate content in raw pigeon pea seeds, while IP<sub>5</sub> and IP<sub>4</sub> represented 24 and 17%, respectively. After 4 days of germination, IP<sub>6</sub> and IP<sub>5</sub> decreased by 61% and 35%,

 Table 2

 Germination effect on antinutritional factors of pigeon pea seeds (*Cajanus cajan* var. Aroíto)

Composition	Raw pigeon pea	Germinated pigeon pea
Raffinose <sup>A</sup>	$1.23\pm0.08^{\rm b}$	$0.26\pm0.02^{\mathrm{a}}$
Stachyose <sup>A</sup>	$2.35\pm0.20^{\rm b}$	$0.44\pm0.07^{\rm a}$
Verbascose <sup>A</sup>	$1.94\pm0.24^{\rm b}$	$0.26\pm0.06^{\rm a}$
Total α-galactosides <sup>A</sup>	$5.52\pm0.22^{\rm b}$	$0.96\pm0.08^{\mathrm{a}}$
Phytic acid <sup>A</sup> (IP <sub>6</sub> )	$0.46\pm0.02^{\rm b}$	$0.18\pm0.02^{\rm a}$
IP <sub>5</sub> <sup>A</sup>	$0.17\pm0.01^{\rm b}$	$0.11\pm0.01^{\mathrm{a}}$
$IP_4^A$	$0.12\pm0.01^{\rm b}$	nd <sup>a</sup>
IP total <sup>A</sup>	$0.72\pm0.05^{\rm b}$	$0.30\pm0.03^{\rm a}$
TIA	$24.7\pm0.45^{\rm b}$	$15.9\pm0.06^{\rm a}$

The same superscript in the same raw means no significant difference  $(P \leq 0.05)$ .

IP6 = Inositol hexaphosphate: IP5 = Inositol pentaphosphate; IP4 = Inositol tetraphosphate; IP total = Total inositol phosphates; TIA = Trypsin inhibitor activity (values in TIU/mg dry matter).

<sup>A</sup> Values in g/100 g dry matter.

respectively and IP<sub>4</sub> was not detectable. Inositol phosphates are used as a phosphorus source during seed germination plant growth and development. Hydrolysis of IP<sub>6</sub> to lower inositol phosphates and free phosphorus is catalyzed by phytase. The activity of this enzyme depends on seed species and variety and increases during sprouting. As a result of the IP<sub>6</sub> degradation, mineral bioavailability and protein and starch digestibility increase in germinated seeds (Honke, Kozlowska, Vidal-Valverde, Frías, & Górecki, 1998). Messina (1999) and Urbano et al. (1999) report that, as a consequence of phytate reduction after germination, zinc and calcium bioavailability increased. As Sathe and Venkatachalam (2002) reported, the degree of phytate hydrolysis during germination depends on the type of seed, conditions and duration of germination, variable amounts of phytases and phosphatases capable of hydrolyzing phytate and germination temperature, because most phytases work at optimum temperatures over 40 °C. Vidal Valverde et al. (1994) found a reduction of 44-66% in IP<sub>6</sub> content in two lentil seed varieties after 6 days of germination. Similarly, Khalil and Mansour (1995) reported a reduction in phytic acid of 54% in germinated faba beans, and Mulimani, Nanda, and Thippeswamy (2003) observed that, after germination of pigeon peas for 4 days, phytic acid showed a loss of 8-20%.

Germination induced a significant ( $P \le 0.05$ ) decrease (36%) in the trypsin inhibitor activity (Table 2). The advantages of decreased trypsin inhibitory activity during germination have been correlated with an improvement in the quality of the seed proteins (Kavas & Nehir, 1992). Sathe and Salunkhe (1981) not only found a significant reduction (62–69%) in trypsin inhibitory activity, but also observed an appreciable reduction in chymotrypsin and  $\alpha$ -amylase inhibitor activities. Frias, Diaz-Pollan, Hedley, and Vidal-Valverde (1995) observed drastic reduction of the trypsin inhibitor activity in germinated lentils.

The germinated cowpeas, as has been indicated above, presented higher nutritive value than the raw cowpeas, since some vitamin content, available sugars and antioxidant capacity increased while some antinutritional factors decreased. The sprout seeds also have high level of protein, starch and dietary fibre. Thus, the germinated cowpeas flours are an excellent ingredient to use in the pasta supplementation.

Table 3 shows the cooking quality parameters of pasta products prepared with semolina and different proportions of germinated pigeon pea flours. Cooking time of pasta products supplemented with germinated flour at 5%, 8% and 10% was significantly ( $P \le 0.05$ ) smaller than pasta control (100% semolina). Similarly, Ferreira, Wang, de Souza, and Ramírez (2004) reported a diminution of cooking time in pasta products made from wheat and soya. The higher the substitution level the shorter was the cooking time. Taha (1992) verified that there were higher cooking losses in noodles from defaulted soy-supplemented whole durum wheat. Although cooking water absorption and cooking loss were higher in all supplemented macaroni,

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Pasta	Cooking time (min)	Cooking water absorption (%)	Cooking loss (%)	Protein loss in water (mg/100 g)
Semolina 100% (Control)	$15.00\pm1.00^{\rm c}$	$152.00\pm2.58^a$	$3.00\pm0.02^a$	$2.71\pm0.03^{\rm a}$
Germinated pigeon pea pastas Semolina:GPPF (95:5) Semolina:GPPF (92:8) Semolina:GPPF (90:10)	$\begin{array}{c} 12.50 \pm 0.50^{b} \\ 10.40 \pm 0.50^{a} \\ 12.00 \pm 1.00^{b} \end{array}$	$\begin{array}{c} 198.00 \pm 7.10^{c} \\ 194.30 \pm 8.30^{bc} \\ 189.30 \pm 4.20^{b} \end{array}$	$\begin{array}{c} 4.29 \pm 0.16^{\rm b} \\ 5.45 \pm 0.07^{\rm c} \\ 5.51 \pm 0.86^{\rm c} \end{array}$	$\begin{array}{c} 7.53 \pm 0.35^{b} \\ 11.5 \pm 0.20^{c} \\ 12.7 \pm 1.78^{d} \end{array}$

Cooking quality parameters of pasta products with 100% semolina and semolina supplemented with germinated pigeon pea (Cajanus cajan var. Aroíto) flour

Values are the mean  $\pm$  standard deviation of three determinations.

Table 3

The same superscript in the same column means no significant difference

these values still remained within an acceptable range, as described by Morales de León, Mercado, and Cecin (1997). Increased cooking water absorption could be explained as changes in the nature of the interaction of legume starch with fibre and/or with proteins, as a consequence of the germination process and subsequent cooking, rendering it more digestible (Urooj & Puttaraj, 1994). Cooking losses could be attributable to the structural changes in the protein network because of the substitution of wheat protein by legume protein. Cooking losses, although higher ( $P \leq 0.05$ ) than control pasta, were similar to those found by Granito, Torres, and Guerra (2003) in pasta products made with 100% semolina and supplemented with defatted corn, cassava, cowpea flour and gluten. Our results were smaller than those reported by Hoseney (1991), who defined undesirable cooking losses above 9% in pasta making. Protein losses in cooking water were negatively affected ( $P \leq 0.05$ ) by the substitution level of germinated pigeon pea flour in the pasta product. As Edwards, Izydorczyk, Dexter, and Biliaderis (1993) reported, besides cooking time, protein content of pasta products could affect soluble solids losses. Results obtained with pasta products prepared from semolina and different proportions of germinated pigeon pea flours are in accordance with those reported by Dexter and Matsuo (1979) who found a significantly greater protein extractability in spaghetti made from poor quality wheat than products from better quality wheat. This was attributable to a greater proportion of extractable gluten protein for the poorer quality wheats. They also found, in cooked spaghetti, a faster diminution of soluble proteins as albumins, globulins, gliadins and some glutenins (higher levels in cooking water) and consequently an increase in insoluble proteins when cooking time was near 12 minutes. These results show the feasibility of using semolina and germinated pigeon pea flours in pasta making.

General acceptability score of cooked pasta products is presented in Table 4 and no significant effect ( $P \leq 0.05$ ) was observed in the general acceptability score of supplemented pasta products in comparison with the pasta made only with semolina.

The hedonic test on parameters such as colour, flavour and texture of cooked pasta products is presented in Table 5. There was no significant difference ( $P \leq 0.05$ ) between control pasta (100% semolina) and supplemented pastas

#### Table 4

General acceptability of cooked pasta products made with 100% semolina			
or semolina supplemented with germinated pigeon pea flour (Cajanus			
<i>cajan</i> var. Aroíto)			

Pasta	General acceptability score	
Semolina 100% (control)	$9.65\pm3.18^a$	
Germinated pigeon pea pastas		
Semolina:GPPF (95:5)	$10.6 \pm 3.11^{a}$	
Semolina:GPPF (92:8)	$8.90 \pm 3.21^{\mathrm{a}}$	
Semolina:GPPF (90:10)	$9.87\pm3.42^{\rm a}$	

Hedonic scale (non structured) test.

Scale 1-15 points: 1 = extremely unacceptable; 15 = extremely acceptable. Values are the mean  $\pm$  standard deviations

The same superscript in the same column means no significant difference between the pasta products ( $P \leq 0.05$ ).

GPPF = germinated pigeon pea flour.

#### Table 5

Hedonic test of cooked pasta products made with 100% semolina or semolina supplemented with germinated pigeon pea flour (Cajanus cajan var. Aroíto)

Pasta	Colour	Flavour	Texture
Germinated pigeon pea pa	stas		
Semolina:GPPF (95:5)	$5.10\pm1.28^{\rm a}$	$4.75\pm1.47^{\rm a}$	$4.27\pm1.19^{\rm a}$
Semolina:GPPF (92:8)	$5.63\pm1.51^{\rm a}$	$5.09\pm0.89^{\rm a}$	$5.00\pm1.34^{\rm a}$
Semolina:GPPF (90:10)	$5.13\pm1.81^{\rm a}$	$4.67\pm2.50^{\rm a}$	$4.40\pm1.43^{\rm a}$

Semolina 100% pasta had a fixed value of 4. Hedonic scale test. Seven point scale: 1 = extremely different by inferiority; 7 = extremely

different by superiority.

GPPF = Germinated pigeon pea flour.

with germinated pigeon pea flours at 5%, 8% and 10% referred to colour, flavour and texture. In accordance with these results, pasta with the highest level of supplementation (10%) was analyzed for chemical and nutritional composition.

Table 6 collects the chemical composition of cooked pasta spaghetti made from 90:10 semolina: germinated pigeon pea flour. The presence of germinated flour as an ingredient in pasta composition, significantly improved  $(P \leq 0.05)$  the contents of nutrients such as protein, fat, total available sugars, total, available and resistant starch, insoluble and soluble dietary fibre, ash, calcium, sodium, potassium, magnesium and iron in comparison with the pasta made with 100% semolina (Fig. 1). Increased values

Table 6

Chemical composition of cooked pasta products made with 100% semolina or semolina supplemented with germinated pigeon pea flour (*Cajanus cajan var.* Aroíto)

Composition	Semolina 100%	Semolina: GPPF
	(Control)	(90:10) pasta
Protein <sup>A</sup>	$14.8\pm0.37^{\rm a}$	$17.3\pm0.07^{\rm b}$
Fat <sup>A</sup>	$0.07\pm0.01^{\rm a}$	$0.22\pm0.02^{\rm b}$
Fructose <sup>A</sup>	$0.27\pm0.01^{\rm a}$	$0.27\pm0.01^{\rm a}$
Glucose <sup>A</sup>	$0.73\pm0.01^{\rm a}$	$1.14\pm0.04^{\rm b}$
Galactose <sup>A</sup>	nd <sup>a</sup>	$0.20\pm0.02^{\rm b}$
Sucrose <sup>A</sup>	$0.86\pm0.02^{\rm a}$	$1.16\pm0.03^{\rm b}$
Total available soluble sugars <sup>A</sup>	$1.86\pm0.03^{\rm a}$	$2.77\pm0.05^{\rm b}$
Available Starch <sup>A</sup>	$71.2\pm1.99^{\rm a}$	$73.4\pm0.24^{\rm b}$
Resistant Starch <sup>A</sup>	$3.85\pm0.81^{\rm a}$	$4.04\pm0.99^{\rm a}$
Total Starch <sup>A</sup>	$73.9\pm1.09^{\rm a}$	$76.5 \pm 1.89^{\mathrm{b}}$
Insoluble fibre <sup>A</sup>	$3.96\pm0.03^{\rm a}$	$4.21\pm0.39^{\rm a}$
Soluble fibre <sup>A</sup>	$0.74\pm0.08^{\rm a}$	$1.35\pm0.18^{\rm b}$
Total dietary fibre <sup>A</sup>	$4.73\pm0.05^{\rm a}$	$5.56\pm0.58^{\rm b}$
Ash <sup>A</sup>	$0.56\pm0.03^{\rm a}$	$0.96\pm0.03^{\rm b}$
Energy (kcal/100 g)	362.71	388.06
Calcium <sup>B</sup>	$51.7\pm0.82^{\rm a}$	$70.7 \pm 1.41^{ m b}$
Sodium <sup>B</sup>	$12.9\pm0.36^{\rm a}$	$25.6\pm2.23^{\mathrm{b}}$
Potassium <sup>B</sup>	$134\pm5.71^{\rm a}$	$172.2\pm5.67^{\rm b}$
Magnesium <sup>B</sup>	$52.9\pm2.30^{\rm a}$	$55.9\pm0.44^{\rm b}$
Zinc <sup>B</sup>	$3.78\pm0.23^{\rm a}$	$2.91\pm0.71^{\rm b}$
Iron <sup>B</sup>	$2.63\pm0.41^{\rm a}$	$2.70\pm0.27^{\rm a}$
Vitamin B1 <sup>B</sup>	$0.11\pm0.00^{\rm a}$	$0.12\pm0.00^{\rm a}$
Vitamin B2 <sup>B</sup>	$0.10\pm0.01^{\rm a}$	$0.11\pm0.01^{\rm a}$
Alfa Tocopherol <sup>B</sup>	nd	$0.74\pm0.02^{\mathrm{b}}$
Beta Tocopherol <sup>B</sup>	nd	nd
Gamma Tocopherol <sup>B</sup>	$1.30\pm0.02^{\rm a}$	$4.47\pm0.14^{\rm b}$
Delta Tocopherol <sup>B</sup>	nd	nd
α-TE Vitamin E (Units/100 g d.m.)	$0.13\pm0.01^{\rm a}$	$1.18\pm0.02^{\rm b}$
Phytic acid <sup>A</sup> (IP <sub>6</sub> )	$0.10\pm0.01^{\mathrm{a}}$	$0.19\pm0.00^{ m b}$
$IP_5^A$	$0.03\pm0.00^{\rm a}$	$0.04\pm0.01^{\rm a}$
$IP_4^A$	nd <sup>a</sup>	$0.03\pm0.00^{ m b}$
IP total <sup>A</sup>	$0.13\pm0.01^{\rm a}$	$0.27\pm0.00^{ m b}$
TEAC (µmol/g)	$2.24\pm0.33^{\rm a}$	$5.80\pm0.31^{\rm b}$
TIA (TIU/mg d.m.)	nd <sup>a</sup>	$1.57\pm0.04^{\rm b}$
Tannin content (gallic acid) <sup>A</sup>	$0.37\pm0.01^{\rm b}$	$0.19\pm0.01^{\rm a}$

Values are the mean  $\pm$  standard deviation of three determinations. The same superscript in the same column means no significant difference ( $P \le 0.05$ ). GPPF = Germinated pigeon pea flour.

<sup>A</sup> Values in g/100 g dry matter.

<sup>B</sup> Values in mg/100 g dry matter.

of protein, ash and fibre content have been reported by Bahnassey, Kan, and Harrold (1986) in spaghetti fortified with 15% legume flour, by Bergman et al. (1994) in pasta products made from soft wheat flour and cowpea meal and spaghetti pasta and by Granito et al. (2003) from semolina fortified with defatted corn, cassava, cowpea flour and gluten. Total, available and resistant starch values found in pasta fortified with germinated pigeon pea flour are similar to those reported by Brighenti, Casiraghi, and Baggio (1998) in wheat pasta and Goñi and Valentín-Gamazo (2003) in spaghetti fortified with chickpea flour. Wittig et al. (2002) found an important increase in dietary fibre in spaghettis made with semolina, sweet lupin flour and gluten, the soluble:insoluble dietary fibre (SDF/IDF) ratio being 1/4.5. Goñi and Valentín-Gamazo (2003) reported a similar proportion (1/2.23). Results in pasta

made from semolina:germinated pigeon pea flour kept the SDF/IDF ratio to 1/3, a value similar to that reported by previous authors. According to Martin-Cabrejas et al. (2004), the SDF/IDF ratio is an important variant related to structural and sensorial properties, and is also of importance from both a dietary and a functional perspective, and the optimal proportion should be 1/2.

Fortified pasta with germinated pigeon pea showed the presence of a large content of minerals, such as calcium, sodium, potassium, magnesium and iron, than control pasta in similar amounts as those reported by Bahnassey et al. (1986) and Granito et al. (2003) in spaghettis fortified with edible legumes. According to Ranhotra, Gelroth, Novak, Bock, and Matthews (1985), pasta products are a good source of minerals but, since cooking water is discarded after use, minerals can be lost. However they found, in cooked commercial pasta products (spaghettis, egg noodles and macaroni), a good retention of minerals, such as iron, calcium, phosphorus, magnesium, zinc, copper and manganese; also, they reported that pasta products were an excellent source of the evaluated minerals, except calcium, in accordance with RDA daily needs.

Energy value of cooked pasta products is also shown in Table 6 and spaghetti fortified with germinated pigeon pea flour showed an increase of 6.98% in the energy value compared with control pasta, which means that it is a nutrient-dense product (388 kcal/100 g d.m.).

Cooked pasta products, enriched with 10% germinated pigeon pea flour, showed a significant increase ( $P \le 0.05$ ) in vitamin E, due to the presence of a large amount of  $\gamma$ tocopherol in germinated flours. Although value of vitamin E activity was below that reported by Wittig et al. (2002) in spaghetti fortified with sweet lupin and gluten (4.52 units TE/100 g), pasta products prepared with 90:10 semolina:germinated pigeon pea flours still satisfy 12–15% of the RDA for vitamin E.

The total antioxidant capacity was also determined in the studied pasta products (Table 6) and it was found that the supplementation with germinated flour doubled the antioxidant capacity compared with control pasta. Such pasta presented twice the amount of phytic acid and lower inositol phosphates (IP<sub>5</sub>, IP<sub>4</sub>) than 100% semolina pasta. From the nutritional point of view, this means that this supplemented pasta could have some additional value because of the presence of antioxidant compounds. Tannin content in fortified pasta was below that obtained for control pasta (Table 6) and this probably could be explained by a higher leaching effect of these compounds into cooking water.

Supplemented pasta presented a trypsin inhibitory activity of 1.57 (TIU/mg), which is a negligible amount when compared with the TIA value found in the germinated flour (15.9 TIU/mg). Our results agree with data reported by Bahnassey et al. (1986) who obtained TIA values of 1.12– 2.41 in spaghettis fortified with 10–15% of legumes flours.

Biological values of fortified pasta are collected in Table 7. True digestibility and PER values had an increase of

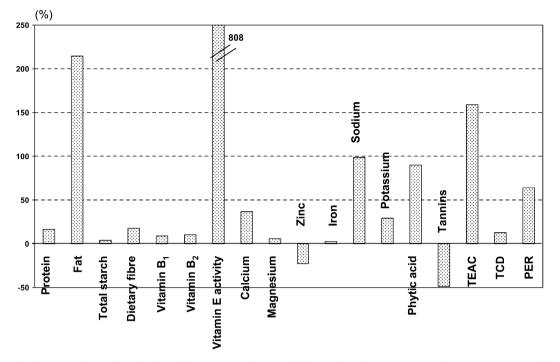


Fig. 1. Nutritional value comparison of cooked semolina pasta supplemented with germinated Cajanus cajan flours (90:10) vs. cooked semolina pasta.

Table 7 Biological values of casein and cooked pasta products made with 100% semolina or semolina supplemented with germinated pigeon pea flour (*Cajanus cajan* var. Aroíto)

	True digestibility (TD)	Protein efficiency ratio (PER)
Casein diet	$98.0\pm0.30^{\rm c}$	$2.98\pm0.13^{\rm c}$
Semolina 100% (control) diet	$84.5\pm3.88^a$	$1.11\pm0.30^{\rm a}$
Pasta Semolina:GPPF (90:10) diet	$95.1\pm0.55^{\rm b}$	$1.82\pm0.28^{\rm b}$

Values are the mean  $\pm$  standard deviations of six determinations. The same superscript in the same column means no significant difference ( $P \leq 0.05$ ).

GPPF = Germinated pigeon pea flour.

12% and 64%, respectively, which demonstrates better nutritional balance of legume-cereal blends. Khalil (2001) reported an increase in biological parameters (PER, true digestibility and biological value) of faba bean after germination. Obizoba (1990) found a PER value of 1.8 in blends of germinated pigeon pea and corn (dietary protein 70% from pigeon pea and 30% from corn). Bergman, Gualberto, and Weber (1996) reported similar results in pasta products with semolina:cowpea meal (90:10), and Torres et al. (2006) in pasta supplemented with  $\alpha$ -galactoside-free *L. angustifolius* flour of varieties Emir and Troll (8–10% substitution of semolina wheat, respectively), and found an increase of PER of 86% and 73%, respectively, in comparison with the pasta made only with semolina.

## 4. Conclusion

Germination of pigeon peas appears to be an effective process for enhancement of chemical and nutritional parameters in this legume, particularly the increase in vitamins B<sub>2</sub>, E and C and the reduction of antinutrients, such as  $\alpha$ -galactosides, inositol phosphates and trypsin inhibitor activity. Germinated seeds, by these benefits, were incorporated as high-protein ingredients (up to 10%) in pasta making resulting in products with good acceptability and larger amounts of protein, total available sugars, dietary fibre, micronutrients, vitamins and PER than pasta made from 100% semolina.

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#### References

- Abecassis, J., Faure, J., & Feillet, P. (1989). Improvement of cooking quality of maize pasta products by heat treatment. *Journal of Science* and Food Agriculture, 47, 475–485.
- Allison, J. B. (1955). Biological evaluation of protein. *Physiological Review*, 35, 664–700.
- AOAC (1990). Official Methods of the Association of Official Analytical Chemists (15th ed.). Arlington, VA: AOAC, pp 152–169.
- Bahnassey, Y. K., Kan, K., & Harrold, R. (1986). Fortification of spaghetti with edible legumes. I. Physicochemical, antinutritional, amino acid and mineral composition. *Cereal Chemistry*, 63, 210–215.
- Bergman, C., Gualberto, D., & Weber, C. (1994). Development of hightemperature-dried soft wheat pasta supplemented with cowpea (*Vigna* unguiculata L. Walp). Cooking quality, color and sensory evaluation. *Cereal Chemistry*, 71, 523–527.
- Bergman, C. J., Gualberto, D. G., & Weber, C. W. (1996). Nutritional evaluation of a high-temperature-dried soft wheat pasta supplemented

with cowpea (Vigna unguiculata (L) Walp). Archivos Latinoamericanos de Nutrición, 46, 146–153.

- Brennan, C. S., Kuri, V., & Tudorica, C. M. (2004). Inulin-enriched pasta: effects on textural properties and starch degradation. *Food Chemistry*, 86, 189–193.
- Brighenti, F., Casiraghi, M. C., & Baggio, C. (1998). Resistant starch in the Italian diet. *British Journal of Nutrition*, 80, 333-341.
- Camacho, L., Sierra, C., Campos, R., Guzmán, E., & Marcus, D. (1992). Cambios nutricionales inducidos por la germinación de leguminosas de consumo habitual en Chile. Archivos Latinoamericanos de Nutrición, 42, 283–290.
- Chitra, U., Singh, U., & Rao, P. V. (1996). Phytic acid, in vitro protein digestibility, dietary fiber and mineral of pulses as influenced by processing methods. *Plant Foods and Human Nutrition*, 49, 307–316.
- Dey, P. M. (1990). Oligosaccharides. In P. M. Dey & J. B. Harborne (Eds.). *Methods in plant biochemistry* (vol. 2, pp. 189–218). Sand Diego, CA: Academic Press.
- Dexter, J. E., & Matsuo, R. R. (1979). Changes in spaghetti protein solubility during cooking. *Cereal Chemistry*, 56, 394–398.
- Duhan, A., Khetarpaul, N., & Bishnoi, S. (2001). Effect of soaking, germination and cooking on phytic acid and hydrochloric acid extractability of a pigeon pea cultivar. *Journal of Food Science and Technology*, 38, 374–378.
- Edwards, N. M., Izydorczyk, M. S., Dexter, J. E., & Biliaderis, C. G. (1993). Cooked pasta textura comparison of dynamic viscoelastic properties. *Cereal Chemistry*, 70, 122–126.
- Ferreira, M., Wang, S., de Souza, C. P., & Ramírez, J. L. (2004). Qualidade de cozimento de massas de trigo e soja pré-cozidas por extrusão. *Pesquisa Agropecuária Brasileira*, 39, 501–507.
- Frias, J., Miranda-Zarate, M. L., Doblado, R., & Vidal-Valverde, C. (2005). Effect of germination and fermentation in the antioxidant vitamin content and antioxidant capacity of *Lupinus albus* L. var. Multolupa.. *Food Chemistry*, 92, 211–220.
- Frias, J., Diaz-Pollan, C., Hedley, C. L., & Vidal-Valverde, C. (1995). Evolution of trypsin inhibitor activity during germination of lentils. *Journal of Agricultural and Food Chemistry*, 43, 2231–2234.
- Ghorpade, V. M., & Kadam, S. S. (1989). Germination. In D. K. Salunkhe & S. S. Kadam (Eds.). CRC Handbook of world Food Legumes: Nutritional Chemistry, Processing Technology and Utilization (vol. III, pp. 165–176). Boca Raton, FL: CRC Press.
- Goñi, I., & Valentín-Gamazo, C. (2003). Chickpea flour ingredient slows glycemic response to pasta in healthy volunteers. *Food Chemistry*, 81, 511–515.
- Granito, M., Frías, J., Doblado, R., Guerra, M., Champ, M., & Vidal-Valverde, C. (2002). Nutritional improvement of beans (*Phaseolus vulgaris*) by natural fermentation. *European Food Research and Technology*, 214, 226–231.
- Granito, M., Torres, A., & Guerra, M. (2003). Desarrollo y evaluación de una pasta a base de trigo, maíz, yuca y frijol. *Interciencia*, 28, 372–379.
- Henry, C. J. K., & Massey, D. (2001). Micronutrient changes during food processing and storage. Crop post harvest programme. Issues paper 5: 1–8. Available from http://www.cphp.uk.com.
- Honke, J., Kozlowska, H., Vidal-Valverde, C., Frías, J., & Górecki, R. (1998). Changes in quantities of inositol phosphates during maturation and germination of legume seeds. *Zeitschrift für Lebensmittel-Untersuchung und-Forschung A*, 206, 279–283.
- Hoseney, C., (1991). Principios de Ciencia y Tecnología de los Cereales. Ed. Acribia, Zaragoza, España, pp. 269–274.
- Kavas, A., & Nehir El, S. (1992). Changes in nutritive value of lentils and mung beans during germination. *Chemie, Mikrobiologie, Technologie der Lebensmittel, 14*, 3–9.
- Khalil, A. H., & Mansour, E. H. (1995). The effect of cooking, autoclaving and germination on the nutritional quality of faba beans. *Food Chemistry*, 54, 177–182.
- Khalil, M. M. (2001). Effect of soaking, germination, autoclaving and cooking on chemical and biological value of guar compared with faba bean. *Die Nahrung*, 45, 246–250.

- King, R. D., & Puwastien, P. (1987). Effect of germination on the proximate composition and nutritional quality of winged beans (*Psophocarpus tetragonolbo*) seeds. *Journal of Food Science*, 52, 106–108.
- Kylen, A. M., & McCready, R. M. (1975). Nutrients in seeds and sprouts of alfalfa, lentils, mung beans and soybeans. *Journal of Food Science*, 40, 1008–1009.
- Kozlowska, H., Honke, J., Sadowska, J., Frías, J., & Vidal-Valverde, C. (1996). Natural fermentation of lentils. Influence of time, concentration and temperature on the kinetics of hydrolysis of inositol phosphates. *Journal of the Science and Food Agriculture*, 71, 367–375.
- Kuiters, L. (1987). Phenolic acids and plant growth in forest ecosystems. PhD Thesis. Vrije Universiteit, Amsterdam, pp. 19–32.
- Larmond, E. (1977). Laboratory Methods for Sensory Evaluation of Food. Publ. 1637, Canada Department of Agriculture, Ottawa.
- Lerhfeld, J. (1994). HPLC separation and quantitation of phytic acid and some inositol phosphates in foods: problems and solutions. *Journal of Agricultural and Food Chemistry*, 42, 2726–2731.
- Lower, C. A., & Kou, T. M. (1989). Oligosaccaride metabolism and accumulation in development soybean seeds. *Crop Science*, 29, 459–465.
- Matsou, R. R., Malcomson, L. J., Edwards, N. M., & Dexter, J. E. (1992). A colorimetric method for estimating spaghetti cooking losses. *Cereal Chemistry*, 69, 27–29.
- Martin-Cabrejas, M. A., Ariza, N., Esteban, R., Mollá, E., Waldron, K., & López-Andreu, F. (2003). Effect of germination on the carbohydrate composition of the dietary fiber of peas (*Pisum sativum L.*). Journal of Agriculture and Food Chemistry, 51, 1254–1259.
- Martin-Cabrejas, M., Sanfiz, B., Vidal, A., Mollá, E., Esteban, R., & López-Andréu, J. (2004). Effect of fermentation and autoclaving on dietary fiber fractions and antinutritional factors of beans (*Phaseolus* vulgaris L). Journal of Agriculture and Food Chemistry, 52, 61–266.
- Messina, M. J. (1999). Legumes and soybeans. Overview of their nutritional profits and health effects. *American Journal Clinical Nutrition*, 70, 439–450.
- Morales de León, J., Mercado, M. P., & Cecin, P. (1997). Desarrollo de una pasta para sopa diseñada de acuerdo a los gustos y recomendaciones nutricias para los ancianos. Archivos Latinoamericanos de Nutrición, 47, 152–156.
- Mulimani, V. H., Nanda, S. K., & Thippeswamy, S. (2003). Effect of processing on phytic acid content in different red gram (*Cajanus cajan* L.) varieties. *Journal of Food Science and Technology*, 40, 371–373.
- Nielsen, M. A., Summer, A. K., & Whalley, L. L. (1980). Fortification of pasta with pea flours and air classified pea protein concentrates. *Cereal Chemistry*, 57, 203–206.
- Nnanna, I. A., & Dixon, P. (1989). Amino acid composition protein quality and water-soluble vitamin content of germinated cowpeas (Vigna unguiculata). Plant Foods for Human Nutrition, 39, 187–200.
- Obizoba, I. C. (1990). Nutritive quality of blends of corn with germinated cowpeas (Vigna unguiculata), pigeon pea (*Cajanus cajan*) and Bambara groundnut (*Voandzeia subterranea*). Cereal Chemistry, 67, 230–232.
- Ologhobo, A. D., & Fetuga, B. L. (1986). Changes in carbohydrate contents of germinating cowpea seeds. *Food Chemistry*, 20, 117–125.
- Oloyo, R. A. (2004). Chemical and nutritional quality changes in germinating seeds of *Cajanus cajan* L. *Food Chemistry*, 85, 497–502.
- Prosky, L., Asp, N. G., Schweiser, T., De Vries, J. W., & Furda, I. (1988). Determination of insoluble, soluble and total dietary fiber in foods and foods products: interlaboratory study. *Journal of the AOAC International*, 71, 1017–1023.
- Prodanov, M., Sierra, I., & Vidal-Valverde, C. (1997). Effect of germination on the thiamine, riboflavin and niacin content in legumes. *Zeitschrift für Lebensmittel-Untersuchung und-Forschung A*, 205, 48–52.
- Ranhotra, G. S., Gelroth, J. A., Novak, F. A., Bock, M. A., & Matthews, R. H. (1985). Retention of selected minerals in enriched pasta products during cooking. *Cereal Chemistry*, 62, 117–119.

- Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M., & Rice-Evans, C. (1999). Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biology and Medicine*, 26, 1231–1237.
- RDA (1989). Recommended dietary allowances. Food and Nutrition Board. (10th ed.). Washingthon, DC: National Academy Press.
- Robinson, H. W., & Hodgen, C. G. (1940). The biuret reaction in the determination of serum protein. I.A study on the condition necessary for the production of the stable which bears a quantitative relationship to the protein concentration. *Journal of Biological Chemistry*, 135, 707–725.
- Sangronis, E., Camacho, C. J., & Cava, R. (2004). Propiedades funcionales de las harinas de leguminosas (*Phaseolus vulgaris* y *Cajanus cajan*) germinadas. *Interciencia*, 29, 80–85.
- Salunkhe, D. K., Chavan, J. K., & Kadam, S. S. (1986). Pigeonpea as an important food source. *Critical Review in Nutrition and Food Science*, 23, 103–140.
- Sathe, S. K., & Salunkhe, D. K. (1981). Studies on trypsin and chymotrypsin inhibitor activities, hemagglutinating activity and sugars in the Great Northern beans (*Phaseolus vulgaris* L.). Journal of Food Science, 46, 626–631.
- Sathe, S. K., & Salunkhe, D. K. (1984). Technology of removal of unwanted compounds of dry beans. *Critical Review in Nutrition and Food Science*, 21, 263–287.
- Sathe, S. K., & Venkatachalam, M. (2002). Influence of processing technologies on phytate and its removal. In: R. Reddy, & S. K. Sathe, (Eds.), Food Phytates. Washinton D.C.: pp. 157–188.
- Siddhuraju, P., & Becker, K. (2001). Effect of various indigenous processing methods on the α-galactoside and mono and disaccharide content of an Indian tribal pulse, *Mucuna pruriens* var. utilis. *Journal* of Science and Food Agriculture, 81, 718–725.
- Sotomayor, C., Frías, J., Fornal, J., Sadowska, J., Urbano, G., & Vidal-Valverde, C. (1999). Lentil starch content and its microscopical structure as influenced by natural fermentation. *Starch/Starke*, 51, 152–156.
- Taha, S. A. (1992). Biochemical, rheological, cooking quality and acceptability of deffated soy-supplemented whole durum meal noodles. *Acta Alimentaria*, 21, 229–238.
- Torres, A., Frías, J., Granito, M., Guerra, M., & Vidal-Valverde, C. (2006). Free α-galactosides lupine flour as pasta ingredient: Chemical,

biological and sensory evaluation. Journal of the Science and Food Agriculture, in press.

- Urbano, G., Lopez-Jurado, M., Fernandez, M., Moreu, M. C., Porres-Foulquie, J., Frias, J., et al. (1999). Ca and P bioavailability of processed lentils as affected by dietary fiber and phytic acid content. *Nutrition Research*, 19, 49–64.
- Urooj, A., & Puttaraj, S. (1994). Effect of processing on starch digestibility in some legumes. An in vitro study. Die Nahrung, 38, 38–46.
- Veena, A., Urooj, A., & Puttaraj, S. (1995). Effect of processing on the composition of dietary fiber and starch in some legumes. *Die Nahrung*, 39, 132–138.
- Verma, P., & Mehta, U. (1988). Study of physical characteristics, sensory evaluation and the effect of sprouting, cooking and dehulling on the antinutritional factors of rice bean (*Vigna umbellate*). Journal of Food Science and Technology, 25, 197–200.
- Vidal-Valverde, C., & Frías, J. (1992). Changes in carbohydrates during germination of lentils. Zeitschrift für Lebensmittel-Untersuchung und-Forschung A, 19, 461–464.
- Vidal Valverde, C., Frías, J., Estrella, I., Gorospe, M. J., Ruíz, R., & Bacon, J. (1994). Effect of processing on some antinutritional factors of lentils. *Journal of Agricultural and Food Chemistry*, 42, 2291–2295.
- Vidal-Valverde, C., Frías, J., Díaz-Pollan, C., Fernández, M., López-Jurado, M., & Urbano, G. (1997). Influence of processing on trypsin inhibitor activity of faba beans and its physiological effect. *Journal of Agricultural and Food Chemistry*, 45, 3559–3564.
- Vidal-Valverde, C., Frías, J., Lambein, F., & Kuo, Y. (2001). Increasing the functionality of legumes by germination. In: 4th European Conference on grain Legumes, Cracow.
- Vidal-Valverde, C., Frías, J., Sierra, I., Blázquez, I., Lambein, F., & Kuo, Y. (2002). New functional legume foods by germination: effect on the nutritive value of beans, lentils and peas. *European Food Research and Technology*, 215, 472–477.
- Watt, B. K., & Merrill, A. L. (1963). Composition of foods. Agriculture Handbook No 8. ARS. US Department of Agriculture, Washington, D.C.
- Wittig, E., Serrano, L., Bunger, A., Soto, D., López, L., Hernández, N., et al. (2002). Optimización de una formulación de espaguetis enriquecidos con fibra dietética y micronutrientes para el adulto mayor. Archivos Latinoamericanos de Nutrición, 52, 91–100.