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## Nutritional composition, anti-nutritional, and technological properties of flours from Brazilian common bean genotypes

Juliana Aparecida Correia Bento<sup>1,4\*</sup>, Jennifer Vieira Pinto<sup>1</sup>, Solange Guidolin Canniatti Brazaca<sup>2</sup> Rosângela Nunes Carvalho<sup>3</sup>, Priscila Zaczuk Bassinello<sup>3</sup>

- <sup>1</sup> Universidade Federal de Goiás, Goiânia, GO, Brasil. E-mail: julianaap.ufg@gmail.com; vp.jennifer@gmail.com
- <sup>2</sup> Universidade de São Paulo, Piracicaba, SP, Brasil. E-mail: sgcbraza@gmail.com
- <sup>3</sup> Embrapa Arroz e Feijão, Santo Antônio de Goiás, GO, Brasil. E-mail: rosangela.carvalho@embrapa.br; priscila.bassinello@embrapa.br
- <sup>4</sup> Universidade Federal de Mato Grosso, Cuiabá, MT, Brasil

ABSTRACT: This study aimed to evaluate the nutritional and technological quality of the flour from different common bean groups (carioca, black, pink, and purple), to verify its potential application as an ingredient in food products. The carioca bean flours had the greatest content of dietary fiber (15-18 g 100g<sup>-1</sup>), essential amino acids (Met and Cys), Fe content (63 mg kg<sup>-1</sup>) combined with the lowest phytate: Fe molar ratio (6.6). It also presented low water absorption (WAI), high water solubility, and intermediate values of peak viscosity (775 cP). The black beans showed the maximum amount of essential amino acids and lipids, and low content of phytates (0.4%). Black bean flour showed the greatest viscoamylographic values, WAI, foaming capacity (FC), foam stability (FS), emulsifying capacity (EC), and emulsion stability (ES). Instead, the pink and purple beans had the lowest viscoamylographic profile, EC, and ES values. The purple grains presented the highest protein amount (22 g 100g<sup>-1</sup>), and a high molar ratio between phytate to Zn and Fe (18.8 and 9.7, respectively). The pink grains presented high Try and carbohydrates contents, and a low Zn quantity. In general, despite the differences observed, the flours had advantageous nutritional and technological properties for different applications in the food industry.

Key words: amino acids profile; minerals; Phaseolus vulgaris, protein digestibility; trypsin and α-amylase inhibitors

# Composição nutricional, propriedades antinutricionais e tecnológicas de farinhas de genótipos de feijoeiro brasileiro

RESUMO: Este estudo teve como objetivo avaliar a qualidade nutricional e tecnológica da farinha de diferentes grupos de feijoeiros (carioca, preto, rosa e roxo), para verificar seu potencial de aplicação como ingrediente em produtos alimentícios. As farinhas de feijão carioca apresentaram os maiores teores de fibra alimentar (15-18 g 100g¹), aminoácidos essenciais (Met e Cys), teor de Fe (63 mg kg¹) combinados com a menor relação molar fitato:Fe (6.6). Também apresentou baixa absorção de água (WAI), alta solubilidade em água e valores intermediários de pico de viscosidade (775 cP). O feijão preto apresentou o máximo teor de aminoácidos essenciais e lipídios, e baixo teor de fitatos (0,4%). A farinha de feijão preto apresentou os maiores valores viscoamilográficos, WAI, capacidade de formação de espuma (FC), estabilidade da espuma (FS), capacidade emulsificante (CE) e estabilidade da emulsão (ES). Em vez disso, os feijões rosa e roxo tiveram o menor perfil viscoamilográfico, valores de CE e ES. Os grãos roxos apresentaram maior teor de proteína (22 g 100g 1) e alta relação molar entre fitato para Zn e Fe (18,8 e 9,7, respectivamente). Os grãos rosados apresentaram alto teor de Try e carboidratos é baixo teor de Zn. De maneira geral, apesar das diferenças observadas, as farinhas apresentaram propriedades nutricionais e tecnológicas vantajosas para diferentes aplicações na indústria alimentícia.

Palavras-chave: perfil de aminoácidos; minerais; Phaseolus vulgaris; digestibilidade de proteínas; inibidores de tripsina e α-amilase



## Introduction

In the last decade, there has been an increase in the demand for food products that have nutritional appeal, especially in terms of protein content, dietary fiber, and the presence of bioactive compounds. The bean grain meets all these requirements, as it has a high content of protein and fiber (both soluble and insoluble), a good source of minerals, and vitamins. In addition, beans have high amounts of phenolic compounds, anthocyanins, saponins, etc., which are responsible for the antioxidant activity of this food (Los et al., 2018; Bento et al., 2021c). Among the common bean (Phaseolus vulgaris) cultivars available in the Brazilian market, the carioca and black beans are the most cultivated, followed by cowpea (Vigna unguiculata). Other types of beans are also cultivated, such as red, white, yellow, pink, and purple beans. The content of phenolic compounds and anthocyanins is related to the color of the grains so that more pigmented grains (e.g., black and purple) are inclined to have a greater amount of these compounds (Bento et al., 2021b).

Despite the nutritional advantages of beans, they may have some undesirable characteristics that limit their acceptability or nutritional value, such as the low content of sulfur amino acids in their proteins and the presence of antinutritional factors. They may also have low protein bioavailability due to the complexation with antinutrients (Los et al., 2018). The antinutritional factors present in beans - phytates, protease inhibitors, polyphenols (tannins), and  $\alpha$ -amylase inhibitors - reduce the activity of some enzymes, the biological action of various chemical compounds, and the absorption of metabolites (Bento et al., 2023). However, the cooking process can attenuate and/or eliminate part of the antinutrients present in beans and increase their protein digestibility (Worku & Sahu, 2017; Los et al., 2018).

In general, consumers prefer to eat beans cooked in broth, as an accompaniment to rice. Other forms of consumption are tropeiro beans (beans with meat, cabbage, and cassava flour), soup, salads, etc. However, the preparation of beans can be time-consuming, which has reduced the consumption of beans in recent years. One option to increase the consumption of beans is the development of bean-based food products, which meet the perspectives of practicality and healthiness (Bassinello et al., 2020). Bean flour is an alternative use for the development of food products with high nutritional value, or an ingredient that can be incorporated into various food preparations. In addition, bean flour can be used in the preparation of gluten-free foods, giving them high protein and dietary fiber content (Bento et al., 2021a). Moreover, beans are an option for plant-based protein, which is in focus nowadays since animal protein products (such as meat and dairy) could be replaced with plant protein products, because of the low conversion efficiency (15%) from crop protein to animal protein in intensive production systems (Ferreira et al., 2021).

The study of the technological properties of bean flours is important to guide for better application of these

ingredients, once the composition of bean grains interferes with their properties. Studies that explore the nutritional and technological properties of bean flour facilitate the use of this material in food processing and development. Therefore, this study aimed to characterize the nutritional and technological quality of the flour from six common bean cultivars, from different commercial groups (carioca, black, pink, and purple), to verify their potential application as ingredients in food products.

## **Materials and Methods**

#### Plant material and flour preparation

Bean grains were selected from the commercial groups: carioca (Pérola and BRS Estilo), black (IPR Uirapuru and BRS Esteio) and special or colored (BRS Agreste (pink) and BRS Pitanga (purple)), from the Active Germplasm Bank of Embrapa Arroz e Feijão, in Santo Antônio de Goiás, GO, Brazil. The cultivation was carried out in the experimental fields of Embrapa Arroz e Feijão, on the Capivara farm, in Santo Antônio de Goiás, GO, Brazil. The soil was fertilized with 200 kg ha-1 of nitrogen, phosphorus, and potassium, at 11:52:00 concentrations, and harvested between September and October 2014. The experimental design was carried out in randomized blocks, in plots composed of 8 rows of 4 meters, with three replications for each cultivar. After harvesting and drying the beans in the sun (final moisture ± 12.0 g 100g<sup>-1</sup>), the beans underwent cleaning, purging, and selection operations. The samples were split and stored in low-density polyethylene bags, in portions of 2 kg per commercial group, in a cool and dry place until used in the preparation and analysis of the respective flours.

To obtain the flour, the grains of the carioca, black, pink, and purple commercial groups, separately, were sanitized with neutral detergent for laboratory use (Multi-N, manufacturer Izzi Química Ltda) and distilled water, in a proportion of 1:5. The grains were dried in an oven at 45 °C for 5 hours, and after drying, transferred to a cyclone-type mill (Ciclotec 1193/FOSS) to obtain the flour (fineness modulus of 2.6-3.4, with particle size between 28 and 200 mm), whose granulometry was determined with a 100 g sample using a magnetic sieve shaker (series 07.09/Bertel) with a sieve variation of 12 to 270 mesh (at speed 7 for 10 minutes shaking). The granulometric profile of the flours of bean by-products was determined according to the method described by Rios et al. (2018).

#### **Proximal composition**

The moisture content, the ash, the crude protein content, and the lipids were carried out with three repetitions and in triplicate, according to AOAC (2016). To obtain the total dietary fiber, a defatted sample obtained from the Soxhlet extraction was used, following the enzyme-gravimetric method. The total carbohydrate content was estimated by difference (dry matter less protein, total dietary fiber, and lipids) (AOAC, 2016). The total energy value was estimated

using the conversion factors of: 4 kcal g<sup>-1</sup> for proteins, 4 kcal g<sup>-1</sup> for carbohydrates and 9 kcal g<sup>-1</sup> for lipids.

#### In vitro protein digestibility

Protein digestibility was performed according to the methodology proposed by Akeson & Stahmann (1964), with modification, in raw and cooked beans. The cooked samples were cooked for 10 minutes at 121 °C, with the previous maceration of 12 hours with water at a ratio of 1:3 for maceration and 1:2 for cooking. After hydrolysis, the digestibility calculation was performed based on the protein percentage measured by Kjeldahl and the average obtained from the digestibility. The results were expressed as percentage.

#### **Mineral content**

The samples were evaluated for mineral content, according to the methodology described by Millar et al. (2019), which consists of nitroperchloric digestion with a solution of HNO<sub>3</sub>:HCIO<sub>4</sub>. The mineral contents were calculated using a standard curve with known concentrations for each element analyzed.

#### Amino acid profile

To determine the amino acid profile, the samples were subjected to acid hydrolysis of proteins and peptides with an aqueous solution of 6N chloric acid, containing 0.01% phenol (m/v), for 22 hours at 110 °C. The hydrolyzed samples were dried in a rotary evaporator, centrifuged at 2655 g, and resuspended in 1.0mL of Milli-Q water. 50 μL aliquots of the supernatant were dried and subjected to pre-column derivatization reaction of free amino acids with phenylisothiocyanate. The separation of the phenylthiocarbamyl-amino acid derivatives (PTC-aa) was performed on a C18 reverse-phase column (Pico-Tag, Waters,  $3.9 \times 150$  mm) with monitoring at 254nm wavelength. The sample quantification was based on the area of each amino acid peak, taking as a reference the peak area of the amino acid standard with known concentration, and the standard was derived under the same conditions as the samples. The essential amino acid score (EAS) was estimated according to the standard recommended by the World Health Organization, called the standard (WHO, 2007).

#### Antinutritional compounds Phytic acid

The sample (1 g) was dissolved in 50 mL of 0.2 N HCl solution and shaken for two hours at 150 rpm. Then, it was filtered on filter paper (14 to 18  $\mu$ m pore), and 1 mL of iron solution was added to 0.5 mL of each extract obtained, followed by a water bath for 30 minutes with boiling water. Subsequently, it was centrifuged at 2447 g at 25 °C for 30 minutes. Then, biripidine solution (1.5 mL) was added to 1.0 mL of the supernatant, followed by reading in a digital spectrophotometer (Shimadzu UV-1800) at an absorbance of 519 nm. For the bioavailability of Fe and Zn, the phytic

acid versus element molar ratio (AF: Fe and AF: Zn) was used, which was determined by the molar ratios between phytic acid and flour minerals.

#### **Condensed tannins**

The extracts were prepared with samples dissolved in methanol (250 mg in 5 mL of methanol), stirred for 20 minutes on a shaker table, and followed by centrifugation for 20 minutes at 2057 g. The tannin content was determined according to the methodology described by Price et al. (1980). The reading was performed in a UV-visible spectrophotometer (Shimadzu UV-1800) at an absorbance of 500 nm. The results were expressed in mg of catechin  $100g^{-1}$  of a sample.

#### Trypsin inhibitor

The flour extracts (1%, w:v) were prepared in 0.1 mol L<sup>-1</sup> sodium phosphate buffer pH 7.6, under stirring at 4 °C for 30 minutes, followed by centrifugation for 10 minutes at 4000 g. The inhibition assay was performed according to the methodology described by Arnon (1970). A trypsin inhibition unit (TIU) was defined as the 0.1 change in absorbance at 280 nm. The results were expressed as TIU per gram of sample, with an inhibitory unit being responsible for the inhibition of a trypsin unit.

#### α-amylase inhibitor

The determination of  $\alpha$ -amylase inhibitor activity was performed using the methodology proposed by <u>Deshpande (1992)</u>, in which 1 g of the sample with 10 mL of distilled water was left to rest for 12 hours at 4 °C, followed by centrifugation at 3000 g for 20 minutes. The supernatant was evaluated for  $\alpha$ -amylase inhibitory activity. Absorbance was read in a spectrophotometer (Shimadzu UV-1800) at 540 nm. One unit of  $\alpha$ -amylase inhibitor (UAI) was defined as the amount of inhibitor that inhibits one unit of  $\alpha$ -amylase.

#### Foaming capacity and stability

A portion (0.5 g) of bean flour was dispersed in 50 mL of McIlvaine buffer solution (disodium phosphate - citric acid), using pH 2.5, 5.6, and 8.0. The dispersion was vigorously shaken in a blender (Walita®) for five minutes and then transferred to a 50 mL beaker. The volume of the foam formed was recorded as foaming capacity (mL 100mL<sup>-1</sup>), and observations were made after 30 minutes to determine the stability of the foam.

## **Emulsifying capacity and stability**

The emulsifying activity was calculated by dividing the volume of the emulsified layer by the total volume before centrifugation. The stability of the emulsion was determined following the same procedure to determine the emulsifying activity, with few modifications: before centrifuging the samples, they were subjected to heat treatment at 85 °C for 15 minutes and centrifuged after cooling. Emulsion stability

was expressed as the percentage of emulsifying activity remaining after heating.

#### Water absorption and solubility

The water solubility index (WSI) and the water absorption index (WAI) of bean flours were determined according to the method of Okezie & Bello (1988).

#### **Paste properties**

The Rapid Visco Analyzer (Perten, RVA 4500, Huddinge, Sweden) was used, and 3.0 g of the bean flour (d.w.b.) dispersed in 25 mL of distilled water. The dispersion was maintained at 50 °C for 1 minute, heated at 95 °C at a rate of 9.5 °C min<sup>-1</sup>, held at 95 °C for 2.5 minutes, and cooled to 50 °C at a speed of 11.842 °C min<sup>-1</sup>. The peak viscosity, final viscosity, and setback (tendency to retrograde) were determined.

#### Statistical analysis

The experiment was carried out in three replications, with a randomized block design. Data were subjected to Analysis of Variance (ANOVA), followed by a comparison of means by Tukey test, at a 5% level of significance, using the STATISTICA version 7.1 program. To conduct the PCA it was used the standardized or normalized PCA, based on Pearson correlation matrix provided by XLSTAT software (Addinsoft, 2021). The PCA biplot was made based on the Euclidean distance in the p-dimensional variable space (distance biplot). Heatmap was created in ClusVis (available in <a href="https://biit.cs.ut.ee/clustvis/">https://biit.cs.ut.ee/clustvis/</a>) using clustering distance and the method for the row was correlation and average, and the tree was ordered by tightest cluster first.

## **Results and Discussion**

#### Proximal composition, minerals, and amino acids

The protein contents of the different bean flour varied between 19.77 e 22.10 g 100g<sup>-1</sup> (<u>Table 1</u>). The lowest values were found for carioca bean flour (cultivar BRS Estilo) and the black group (cultivars BRS Esteio and IPR Uirapuru). On the other hand, flours from cultivars BRS Pitanga and Pérola had the highest protein values.

The flours showed protein digestibility between 81.02 and 84.30%, with no statistical difference (p > 0.05), except for the flour of cultivar BRS Esteio, which showed a value of 3.6% lower. Protein digestibility in cooked grains was higher, ranging from 88.99 to 90.48% (supplementary Table 1). The increase in protein digestibility after cooking is due to the denatured protein being more susceptible to the action of digestive enzymes. In addition, the cooking process denatures enzymes that decrease protein absorption during digestion. (e.g., trypsin inhibitor) (Shi et al., 2017). Therefore, after cooking the product developed with raw bean flour, the protein digestibility will be increased. For lipid content, the flour of the cultivar BRS Esteio, from the black group, had the highest content (p < 0.05) ( $\underline{\text{Table 1}}$ ). The ash contents of bean flour varied between 4.39 and 4.67 g  $100g^{-1}$  (p > 0.05) (Table 1).

The soluble fiber content, responsible for the increase in the viscosity of the gastrointestinal content, of carioca bean flour (5.14-5.30 g  $100g^{-1}$ ) were higher (p < 0.05) when compared to other flours. The results were comparable with that found by <u>Felker et al. (2018)</u> for pinto bean (5.4 g  $100g^{-1}$ ). Insoluble fibers ranged from 8.48 to 13.13 g  $100g^{-1}$  in the bean flours (<u>Table 1</u>). Results superior to that found in wheat

Table 1. Proximal composition (dry weight) (g 100g<sup>-1</sup>), insoluble (IDF), soluble (SDF), and total (TDF) dietary fiber, energy (kcal), and mineral content (dry weight) of the different bean flours.

Cultivars	Protein	Lipids	Ash	Carbohydrate	SDF	IDF	TDF	Energy
BRS Estilo	19.45 <sup>c</sup> ±0.86	1.51 <sup>c</sup> ±0.09	$4.39^{a}\pm0.09$	56.38 <sup>c</sup> ±0.92	5.14 <sup>a</sup> ±0.59	13.13 <sup>a</sup> ±1.01	18.27 <sup>a</sup> ±1.21	
Pérola	21.48 <sup>ab</sup> ± .25	1.35 <sup>cd</sup> ±0.08	4.48 <sup>a</sup> ±0.05	57.37 <sup>bc</sup> ±0.57	5.30 <sup>a</sup> ±0.33	10.04 <sup>a</sup> ±0.33	15.33 <sup>b</sup> ±0.26	327.52 <sup>d</sup> ±1.20
<b>BRS Agreste</b>	20.43 <sup>bc</sup> ±1.10	$1.22^{de}0.11$	4.67° ±0.21	63.85° ±1.44	1.35° ±0.22	8.48° ±0.81	$9.83^{d} \pm 0.13$	348.08 <sup>a</sup> ±1.21
BRS Pitanga	22.10 <sup>a</sup> ±0.72	1.07 <sup>e</sup> ±0.09	4.50° ±0.22	58.25 <sup>bc</sup> ±1.39	3.64 <sup>b</sup> ±0.36	10.45 <sup>b</sup> ±0.46	14.08 <sup>bc</sup> ±0.46	331.06 <sup>cd</sup> ±2.37
<b>BRS Esteio</b>	19.77 <sup>c</sup> ±1.12	2.31 <sup>a</sup> ±0.11	4.45 <sup>a</sup> ±0.09	60.01 <sup>b</sup> ±1.25	3.24 <sup>b</sup> ±0.33	10.21 <sup>b</sup> ±0.31	13.46 <sup>c</sup> ±0.05	339.93 <sup>b</sup> ±0.45
IPR Uirapuru	19.91° ±0.77	1.82 <sup>b</sup> ±0.26	4.48 <sup>a</sup> ±0.22	60.29 <sup>b</sup> ±0.83	3.40 <sup>b</sup> ±0.36	10.11 <sup>b</sup> ±0.43	13.51° ±0.42	337.15 <sup>bc</sup> ±2.23
				Min	nerals			
	N (g kg	<sup>1</sup> )	P (g kg <sup>-1</sup> )	К (а	g kg <sup>-1</sup> )	Ca (g kg <sup>-1</sup>	<sup>1</sup> )	Mg (g kg <sup>-1</sup> )
BRS Estilo	28.42° ±0.99		4.65ª ±0.13	8.14ª ±0.59		1.19 <sup>a</sup> ±0.05		1.43 <sup>b</sup> ±0.09
Pérola	32.60° ±1.21		4.73 <sup>a</sup> ±0.09	10.00 <sup>a</sup> ±0.06		1.02 <sup>ab</sup> ±0.15		1.71 <sup>a</sup> ±0.04
BRS Agreste	30.37° ±2.45		4.63° ± 0.27	$9.18^{a} \pm 0.84$		0.84 <sup>b</sup> ± 0.11		1.40 <sup>b</sup> ± 0.03
BRS Pitanga	32.81° ±3.30		$4.85^{a} \pm 0.24$	8.92 <sup>a</sup> ± 0.83		$1.10^{ab} \pm 0.$	03	1.52 <sup>b</sup> ± 0.04
<b>BRS Esteio</b>	30.11 <sup>a</sup> ±0.45		$4.59^{a} \pm 0.10$	$8.72^{a} \pm 0.70$		$0.95^{ab} \pm 0.$	03	1.45 <sup>b</sup> ± 0.03
IPR Uirapuru	29.78ª ±1.86		4.84° ± 0.31	9.07 <sup>a</sup> ± 0.72		1.24 <sup>a</sup> ± 0.2	20	1.51 <sup>b</sup> ± 0.09
	Cu (mg k	g <sup>-1</sup> )	Fe (mg kg <sup>-1</sup> )	Mn (ı	ng kg <sup>-1</sup> )	Zn (mg kg	<sup>-1</sup> )	
BRS Estilo	9.86ª ±0.	93	63.71 <sup>a</sup> ±2.78	10.38	a ± 0.65	33.77ª±2.	21	
Pérola	9.96° ± 0	.08	52.22 <sup>b</sup> ±0.93	11.22	a ± 0.87	33.18°±0.	29	
BRS Agreste	9.06° ± 0	.83	51.47 <sup>b</sup> ± 5.28	9.77	± 1.41	29.00 <sup>b</sup> ±1.	43	
BRS Pitanga	8.76 <sup>a</sup> ± 0	.37	48.12 <sup>bc</sup> ±3.39	10.91	a ± 0.57	33.42°±1.	27	
BRS Esteio	9.20° ± 0.54		42.25° ± 0.72	10.38 <sup>a</sup> ± 0.75		33.53°±0.38		
IPR Uirapuru	9.17ª ± 0	.58	52.33 <sup>b</sup> ± 2.52	10.39	<sup>a</sup> ± 1.25	32.98 <sup>a</sup> ±1.0	03	

<sup>\*</sup> Means followed by the same letter in the column do not differ significantly from each other by the Tukey test (p > 0.05).

flour (4.93 g  $100g^{-1}$ ) and similar to fava bean flour (9.07 g  $100g^{-1}$ ) (Millar et al., 2019).

Insoluble fiber is responsible for reducing intestinal transit time, increasing stool weight, slowing glucose absorption, and decelerating starch digestion. The total dietary fiber content of bean flours varied between 9.83 and 18.27 g 100g<sup>-1</sup> for flours of cultivars BRS Agreste and BRS Estilo, respectively (<u>Table 1</u>). Carioca bean flours presented higher TDF than wheat flour (10.08 g 100g<sup>-1</sup>) and fava beans (13.8 g 100g<sup>-1</sup>) (<u>Millar et al., 2019</u>), and lower than pinto bean (23.0 g 100g<sup>-1</sup>) (<u>Felker et al., 2018</u>).

Regarding the mineral composition of bean flours, nitrogen, potassium, phosphorus, magnesium, calcium, iron, zinc, manganese, and copper stood out in higher concentrations (Table 1). Therefore, bean flour is an abundant source of essential minerals for the functioning of the human body, as they are needed in various enzymatic reactions. Zinc has been recognized to provide normal cognitive function as well as fatty acid and carbohydrate metabolism (Millar et al., 2019). Iron is a mineral of great importance in human nutrition, as it is present in hemoglobin, which is essential for breathing. The results found for Fe and P in the carioca bean cultivar, BRS Estilo, were superior to those reported by Ribeiro & Kläsener (2020), 58.41 and 3.23 g kg<sup>-1</sup>, respectively. Furthermore, it is noteworthy that the proximate composition, as well as the concentrations of minerals in the common bean, varies according to genotype (G), environment (E), and G × E interaction (Ribeiro & Kläsener, 2020).

The essential amino acids found in higher concentrations in bean flour were leucine, followed by lysine, phenylalanine, threonine, valine, isoleucine, tyrosine, histidine, methionine, tryptophan, and cysteine (Table 2). Essential amino acids are those that the human body cannot synthesize, so they must be ingested with meals. All samples exhibited higher amounts of threonine, valine, isoleucine, tryptophan, leucine, lysine, and histidine compared to WHO (2007) requirements for children (2-5 years) (Table 2).

The black bean flours (BRS Esteio and IPR Uirapuru) presented higher levels of essential amino acids, followed by the special beans (BRS Agreste and BRS Pitanga), and finally, the carioca beans (BRS Estilo and Pérola). The amino acids methionine and cysteine are usually limiting in legumes. There was no statistical difference (p > 0.05) in the methionine contents (24.72 to 29.57 mg g $^{-1}$  of protein) in the flours of the cultivars BRS Esteio and BRS Estilo; and cysteine (6.02 to 7.10 mg g $^{-1}$  of protein) in the flours of cultivars BRS Agreste and Pérola. According to Ribeiro et al. (2007) this composition is very similar to that observed in bean cultivars marketed in Brazil.

The Essential Amino Acid Score (EAS) establishes a relationship between the content of each indispensable amino acid of the test protein with the corresponding amino acid of a standard or a protein taken as a reference or WHO (2007) standard (Table 2).

The amino acid that has the lowest EAS is considered limiting, and a protein that has a score greater than 1.0 for all amino acids is of high nutritional value. The EAS found

Table 2. Composition of essential and non-essential amino acids (mg g<sup>-1</sup> protein) of different common bean flours.

Amino acids	BRS Estilo	Pérola	BRS Agreste	BRS Pitanga	BRS Esteio	IPR Uirapuru	Standard FAO/WHO/UNU <sup>3</sup>
Tryptophan <sup>1</sup>	19.83 <sup>ab</sup> ± 0.02	17.00 <sup>bc</sup> ± 0.03	22.33° ± 0.02	15.33 <sup>cd</sup> ± 0.02	13.50 <sup>d</sup> ± 0.01	14.50 <sup>cd</sup> ± 0.01	7.4
Histidine <sup>1</sup>	42.83 <sup>ab</sup> ± 0.02	44.67 <sup>ab</sup> ± 0.06	41.33 <sup>b</sup> ± 0.04	45.67 <sup>ab</sup> ± 0.06	49.00 <sup>ab</sup> ± 0.05	51.50° ± 0.08	18.0
Valine <sup>1</sup>	78.83 <sup>b</sup> ± 0.06	85.00 <sup>b</sup> ± 0.05	83.00 <sup>b</sup> ± 0.06	83.50 <sup>b</sup> ± 0.12	87.83 <sup>b</sup> ± 0.09	102.50 <sup>a</sup> ± 0.06	42.0
Valine <sup>1</sup>	103.17 <sup>b</sup> ± 0.06	109.83 <sup>b</sup> ± 0.07	106.67 <sup>b</sup> ± 0.08	103.83 <sup>b</sup> ± 0.11	114.17 <sup>ab</sup> ± 0.15	130.00° ± 0.14	52.0
Isoleucine <sup>1</sup>	66.33 <sup>b</sup> ± 0.05	70.00 <sup>b</sup> ± 0.04	69.50 <sup>b</sup> ± 0.05	68.83 <sup>b</sup> ± 0.10	73.67 <sup>ab</sup> ± 0.08	101.33° ± 0.37	31.0
Leucine <sup>1</sup>	137.83 <sup>b</sup> ± 0.10	145.67 <sup>b</sup> ± 0.09	144.00 <sup>b</sup> ± 0.11	141.33 <sup>b</sup> ± 0.15	152.33 <sup>b</sup> ± 0.16	177.17 <sup>a</sup> ± 0.10	63.0
Phenyl-Tyr <sup>1.4</sup>	161.34 <sup>b</sup> ± 0.09	$169.17^{b} \pm 0.06$	166.50 <sup>b</sup> ± 1.3	163.50 <sup>b</sup> ± 0.16	172.83 <sup>b</sup> ± 0.18	204.00 <sup>a</sup> ± 0.14	46.0
Phenylalanine <sup>1</sup>	99.17 <sup>b</sup> ± 0.09	104.67 <sup>b</sup> ± 0.06	$103.67^{b} \pm 0.08$	101.50 <sup>b</sup> ± 0.10	107.50 <sup>b</sup> ± 0.11	127.17 <sup>a</sup> ± 0.09	-
Tyrosine <sup>1</sup>	62.17 <sup>b</sup> ± 0.04	$64.50^{b} \pm 004$	62.83 <sup>b</sup> ± 0.05	62.00 <sup>b</sup> ± 0.06	65.33 <sup>b</sup> ± 0.07	$76.83^{a} \pm 0.05$	-
Met-Cys <sup>1.4</sup>	36.49 <sup>a</sup> ± 0.06	35.13° ± 0.10	31.72° ± 0.06	34.05° ± 0.05	31.24° ± 0.04	32.67 <sup>a</sup> ± 0.04	26.0
Methionine <sup>1</sup>	29.57 <sup>a</sup> ± 0.05	28.03° ± 0.08	$25.70^{a} \pm 0.05$	$27.22^{a} \pm 0.04$	$24.72^{a} \pm 0.03$	25.67 <sup>a</sup> ± 0.04	-
Cysteine <sup>1</sup>	6.92° ± 0.01	$7.10^{a} \pm 0.02$	6.02 <sup>a</sup> ± 0.01	6.83° ± 0.01	$6.52^{a} \pm 0.01$	$7.00^{a} \pm 0.00$	-
Cysteine <sup>1</sup>	79.67 <sup>b</sup> ± 0.04	86.33 <sup>b</sup> ± 0.05	86.00 <sup>b</sup> ± 0.06	88.00 <sup>b</sup> ± 0.09	87.83 <sup>b</sup> ± 0.10	105.67a ± 0.08	27.0
Total	726.32	762.8	751.05	744.04	782.40	919.34	312.40
Eas (%) <sup>5</sup>	1.40	1.35	1.22	1.31	1.20	1.26	-
Aspartic acid <sup>2</sup>	190.00 <sup>b</sup> ± 0.06	201.00 <sup>b</sup> ± 0.11	204.50 <sup>b</sup> ± 0.17	205.17 <sup>b</sup> ± 0.21	215.67 <sup>ab</sup> ± 0.29	245.17 <sup>a</sup> ± 0.22	-
Serine <sup>2</sup>	105.50 <sup>b</sup> ± 0.06	109.67 <sup>b</sup> ± 0.09	110.67 <sup>b</sup> ± 0.09	111.00 <sup>b</sup> ± 0.10	121.33ab ± 0.13	135.33 <sup>a</sup> ± 0.12	-
Glutamic acid <sup>2</sup>	250.67 <sup>bc</sup> ± 0.09	260.50 <sup>bc</sup> ± 0.18	247.83° ± 0.21	244.33° ± 0.26	291.00 <sup>ab</sup> ± 0.34	326.00° ± 0.28	-
Glycine <sup>2</sup>	77.00 <sup>b</sup> ± 0.8	77.50 <sup>b</sup> ± 0.10	76.00 <sup>b</sup> ± 0.05	75.67 <sup>b</sup> ± 0.09	80.67 <sup>ab</sup> ± 0.09	95.33° ± 0.09	-
Arginine <sup>2</sup>	119.17 <sup>b</sup> ± 0.10	132.50 <sup>ab</sup> ± 0.12	120.33 <sup>b</sup> ± 0.11	132.50 <sup>ab</sup> ±0.09	128.67 <sup>ab</sup> ± 0.15	148.83 <sup>a</sup> ± 0.17	-
Proline <sup>2</sup>	$70.83^{b} \pm 0.08$	74.00 <sup>b</sup> ± 0.06	$70.67^{b} \pm 0.05$	69.33 <sup>b</sup> ± 0.07	80.17 <sup>ab</sup> ± 0.12	89.50 <sup>a</sup> ± 0.07	-
Alanine <sup>2</sup>	68.67 <sup>b</sup> ± 0.03	67.83 <sup>b</sup> ± 0.06	68.83 <sup>b</sup> ± 0.04	64.83 <sup>b</sup> ± 0.07	73.17 <sup>ab</sup> ± 0.10	81.33 <sup>a</sup> ± 0.07	-
Total	881.84	923.10	898.83	902.83	990.68	1121.49	-

<sup>\*</sup> Means followed by the same letter in the row do not differ significantly from each other by the Tukey test (p > 0.05); ¹ Essential amino acid; ² Non-essential amino acids; ³ FAO/WHO/UNU standard is for preschool children 1 to 2 years old (WHO, 2007); ⁴ Phenyl-Tyr = phenylalanine and tyrosine (aromatic amino acids), and Met-Cys = methionine and cysteine (sulfur amino acids); ⁵ EAS - essential amino acid score: proportion of methionine + cysteine (limiting amino acids in bean flour) concerning the needs of preschool children aged 1 to 2 years old; Shaded values correspond to amino acids that are limiting in flour, for school-age children.

for the limiting amino acids of the bean flour in the present study ranged from 1.20, for the bean flour of the cultivar BRS Esteio, to 1.40 for the bean flour of the cultivar BRS Estilo. This indicates that comparing the values obtained with the reference values, the bean grains evaluated have an excellent composition of essential amino acids, considered of high quality, capable of meeting a large part of the needs of schoolchildren and adults.

The similarities in chemical composition (proximate composition, amino acids, and minerals) between the different bean cultivars were evaluated by principal component analysis (Figure 1A) with an explained variance of 73.8%. Black bean flours stand out from the others with higher amino acid content, except for TRY, MET, and CYS. The highest concentration of limiting amino acids MET and CYS was observed in flours of the carioca bean group. These also presented the highest minerals content Mg, K, and Cu (cultivar BRS Pérola) and Fe (cultivar BRS Estilo) (Figure 1). Flour from the cultivar BRS Agreste (pink) did not group with the other grains, due to its higher carbohydrate amount and lower amino acid content (except for TRY), minerals, and total dietary fiber (Figure 1A).

#### **Anti-Nutritionals**

Phytic acid contents in bean flours ranged from 0.428 to 0.551%, with cultivars in the carioca, pink and purple group showing the highest values, and beans in the black group showing the lowest values (p < 0.05) (Table 2). Phytic acid can form insoluble complexes with minerals, negatively interfering with its bioavailability (Bento et al., 2023). This bioavailability has been evaluated through the molar ratio between phytic acid and mineral (phytate: mineral) in the diet, so that the higher the molar ratio, the lower the mineral bioavailability (WHO, 2007; Mayer Labba et al., 2021). The bean flour of cultivar BRS Pitanga presented the highest value for the molar ratio (phytate: iron), being 46.54% higher than

the molar ratio presented by BRS Estilo (<u>Table 2</u>). The bean flours of the cultivars BRS Estilo and IPR Uirapuru were the ones that presented the lowest values for molar ratio when compared to the others. A molar ratio less than 0.4 indicates a high iron bioavailability (<u>Mayer Labba et al., 2021</u>), thus all bean flour presents low iron bioavailability.

The values for molar ratio (phytate: zinc) were between 12.63 and 18.88 (Table 2). According to the World Health Organization (WHO), diets with a phytate: zinc molar ratio above 15 have low Zn bioavailability (10 to 15%), between 5 and 15 have average Zn bioavailability (30 to 35%) and, below 5 have high bioavailability (WHO, 2007). Therefore, bean flours from cultivars BRS Agreste, BRS Pitanga, and Pérola showed low zinc bioavailability. On the other hand, BRS Estilo, BRS Esteio, and IPR Uirapuru flours presented medium bioavailability of Zn. Heat treatment reduces the phytic acid content in pulses. Furthermore, depending on its concentration, phytic acid can be considered beneficial to health as it acts as an anti-cancer, antioxidant, and anti-diabetics (Millar et al., 2019).

The bean flours of the Pérola cultivar had the lowest tannin content when compared to the other cultivars. On the other hand, the highest values were found for the cultivars of the black group, BRS Esteio and IPR Uirapuru (Table 3). The use of proteins in animals and humans is affected by the presence of tannins in the integument of beans, whose content can vary from 0 to 2%, depending on the species and seed color. However, Belmiro et al. (2020) and Bento et al. (2021b) showed that after the thermal treatment of the bean grains, a reduction (90-100%) in the tannin content is observed. Considering that the raw bean flour-based product will undergo thermal treatment, the tannin content should not interfere with protein digestion.

The concentration of trypsin inhibitors in the different bean cultivars ranged from 0.53 to 1.39 UIT mg<sup>-1</sup> (<u>Table</u> 3). The values found for all cultivars are lower than those

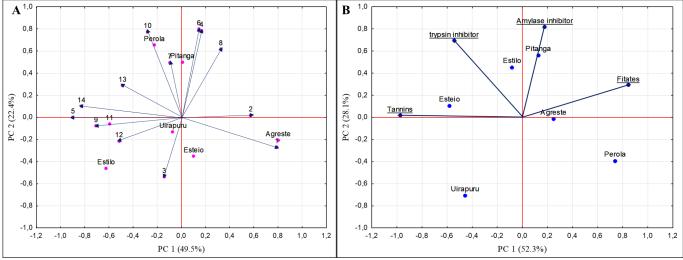


Figure 1. A: PCA (Principal Component Analysis) of the chemical components and minerals present in the different common bean flours (1- Carbohydrate, 2- ashes, 3-lipids, 4-protein, 5-TDF, 6-N, 7-P, 8-K, 9-Ca, 10-Mg, 11-Cu, 12-Fe, 13-Mn, and 14-Zn); B: PCA of the antinutrients content present in the different common bean flours.

**Table 3.** Phytic acid content, tannins, trypsin inhibitors,  $\alpha$ -amylase inhibitors, and molar ratio (Phytate: Iron and Phytate: Zinc) of bean flours.

Cultivars	Phytic acid (PA)	Molar ratio	Molar ratio	Tannins	Trypsin inhibitor	α-amylase
Culturals	(%)	PA: Fe	PA: Zn	(mg catechin g <sup>-1</sup> )	(TIU·g <sup>-1</sup> )	inhibitor (AIU g <sup>-1</sup> )
BRS Estilo	$0.496^{a} \pm 0.03$	$6.61^{bc} \pm 0.62$	$14.57^{bc} \pm 0.77$	$6.15^{\circ} \pm 0.30$	1361.78° ± 78.54	256.34° ± 2.01
Pérola	$0.539^{a} \pm 0.01$	$8.74^{abc} \pm 0.16$	16.10 <sup>ab</sup> ± 0.27	1.03 <sup>e</sup> ± 0.06	1177.78 <sup>b</sup> ± 33.99	232.70° ± 16.65
<b>BRS Agreste</b>	$0.551^{a} \pm 0.04$	$9.16^{ab} \pm 1.49$	$16.27^{a} \pm 0.80$	$4.37^{d} \pm 0.38$	666.67° ± 27.76	170.94 <sup>b</sup> ± 13.87
BRS Pitanga	$0.549^{a} \pm 0.02$	9.69° ± 0.85	18.88 <sup>ab</sup> ± 2.01	6.71° ± 0.57	745.78° ± 70.93	278.95° ± 37.17
<b>BRS Esteio</b>	$0.428^{b} \pm 0.03$	$8.57^{abc} \pm 0.64$	$12.63^{\circ} \pm 0.86$	$11.79^{a} \pm 0.42$	1387.78 <sup>a</sup> ± 82.46	255.18° ± 37.41
IPR Uirapuru	0.443 <sup>b</sup> ± 0.04	$7.16^{bc} \pm 0.80$	13.31 <sup>bc</sup> ± 1.56	9.97 <sup>b</sup> ± 0.43	53.33 <sup>d</sup> ± 27.76	100.33° ± 13.06

<sup>\*</sup> Means followed by the same letter in the column do not differ significantly from each other by the Tukey test (p > 0.05). TIU - Trypsin Inhibitory Unit; AIU - α-Amylase Inhibitory Unit.

presented by fava bean (1.2-23.1 UIT mg¹) and soybean (45.89 UIT mg¹) (Shi et al., 2017; Mayer Labba et al., 2021). Trypsin inhibitors reduce enzyme activity of trypsin and chymotrypsin, thus reducing the levels of digestion and absorption of proteins. The cooking process may provide a total removal of trypsin inhibitory activity (Shi et al., 2017), therefore this antinutrient may not interfere with the nutritional value of cooked products made with bean flours.

The contents of  $\alpha$ -amylase inhibitors ranged from 100.33 to 278.95 UIA  $g^{-1}$  (Table 3). The highest values of  $\alpha$ -amylase inhibitors were for the cultivars BRS Pitanga, BRS Estilo, BRS Esteio, and Pérola, being between 278.95 and 232.70 AIU  $g^{-1}$ . The values found are lower than those reported for soybean (938.7 AIU  $g^{-1}$ ) and navy bean (1079.8 AIU  $g^{-1}$ ) (Shi et al., 2017). The  $\alpha$ -amylase inhibitor is known to reduce the digestibility of starch due to the inhibition of pancreatic and salivary  $\alpha$ -amylase enzymes. On the other hand, amylase inhibitors could be healthy since it prevents dietary starch from being absorbed by the body, in that way, it decreases the peak of glucose levels in postprandial blood. Thus, this antinutrient can be an advantageous tactic in glycemic index control in people with type 2 diabetes (Shi et al., 2017).

Similarities in antinutrient content between different bean cultivars were evaluated by principal component analysis (Figure 1B) with a variance of 80.4%. The flours were separated into two groups (PC1 = 52.3%): grains from cultivars with high phytate content (BRS Pitanga, Pérola, and BRS Agreste), and low phytate content (BRS Estilo, BRS Esteio, and IPR Uirapuru) (Figure 1B). Regarding the other

antinutrients (tannins, trypsin inhibitor, and  $\alpha$ -amylase inhibitor), the flours of cultivars BRS Estilo and IPR Uirapuru stood out with the highest content. On the other hand, the cultivar IPR Uirapuru had the lowest trypsin inhibitor content, corresponding to approximately 3.9% of the levels presented by the cultivars BRS Estilo and BRS Esteio, which had the highest values. However, it is noteworthy that these antinutrients can be reduced or eliminated during the food cooking process (Shi et al., 2017; Belmiro et al., 2020; Bento et al., 2021b; Mayer Labba et al., 2021). Therefore, cooked foods made with bean flour must have a low antinutrient content.

#### **Technological properties**

Flour from cultivars BRS Agreste, BRS Pitanga, and IPR Uirapuru showed the highest percentage of foam formation at pH 2.5 (<u>Table 4</u>). For the cultivar BRS Esteio, foam formation was higher at pH 8.0 and lower at pH 5.6. This variation in the foaming capacity (FC) regarding the pH of the medium is related to the solubility of proteins. Therefore, these results are influenced by the protein composition of each cultivar. In addition, the composition of the flour (e.g., carbohydrate) influences the FC (<u>Gupta et al., 2018</u>), which would justify the higher value (53.3%) in the flour of the cultivar with the highest carbohydrate content (BRS Agreste) (<u>Table 4</u>).

As for foaming stability (FS), in general, the flours were more stable at pH 5.6, with no statistical difference between cultivars (p > 0.05). The stability of the foam is related to the quality of the protein, requiring the formation of cohesive,

**Table 4.** Values of foam formation capacity and stability at pH 2.5; 5.6 and 8.0 and emulsion stability capacity of bean flours from different commercial groups.

Cultivars	BRS Estilo	Pérola	BRS Agreste	BRS Pitanga	BRS Esteio	IPR Uirapuru		
Foaming capacity (%)								
pH 2.5	36.67 <sup>b</sup> ± 1.53	36.00 <sup>b</sup> ± 2.00	53.33 <sup>a</sup> ± 5.77	41.67 <sup>b</sup> ± 5.51	26.67° ± 2.08	37.67 <sup>b</sup> ± 2.08		
pH 5.6	26.67 <sup>bc</sup> ± 0.58	28.00 <sup>bc</sup> ±3.46	$30.33^{ab} \pm 2.08$	$35.00^{a} \pm 1.73$	26.00 <sup>bc</sup> ± 3.61	$25.00^{\circ} \pm 1.73$		
pH 8.0	27.00° ± 1.73	25.00° ± 1.00	30.67 <sup>b</sup> ± 0.58	33.33 <sup>ab</sup> ± 1.53	33.67 <sup>a</sup> ± 1.15	33.33 <sup>ab</sup> ± 1.53		
			Foam stability (%)					
pH 2.5	71.33 <sup>ab</sup> ± 3.25	69.50 <sup>b</sup> ± 3.40	64.56 <sup>b</sup> ± 7.68	67.64 <sup>b</sup> ± 6.18	78.78 <sup>a</sup> ± 1.66	78.71° ± 1.15		
pH 5.6	94.96° ± 2.06	93.11 <sup>a</sup> ± 2.99	$92.57^{a} \pm 6.67$	93.27° ± 4.71	92.06° ± 10.35	96.03° ± 3.87		
pH 8.0	94.02 <sup>ab</sup> ± 3.69	96.05° ± 0.19	91.38 <sup>ab</sup> ± 1.88	96.01 <sup>a</sup> ± 1.81	90.12 <sup>ab</sup> ± 1.97	89.00 <sup>b</sup> ± 1.81		
Emulsifying capacity (%)	51.62 <sup>b</sup> ± 0.09	51.53 <sup>b</sup> ± 2.83	51.32 <sup>b</sup> ± 1.21	50.31 <sup>b</sup> ± 1.23	55.16 <sup>a</sup> ± 0.28	53.27 <sup>ab</sup> ± 2.27		
Emulsion stability (%)	54.64 <sup>bc</sup> ± 1.73	58.10 <sup>ab</sup> ±3.73	54.35 <sup>bc</sup> ± 1.39	49.70° ± 4.12	59.57 <sup>ab</sup> ± 0.82	63.48° ± 1.11		

<sup>\*</sup> Means followed by the same letter in the column do not differ significantly from each other by the Tukey test (p > 0.05).

elastic, continuous, and air-impermeable films. According to Marquezi et al. (2017) a decrease in the attractive hydrophobic forces between protein molecules occurs in the pH regions in the acidic and alkaline range, where the protein molecules become positively and negatively charged, respectively. In the region of the isoelectric point or near, the lack of repulsive interactions promotes favorable protein-protein interactions and the formation of a viscous film at the interface, in addition to an increase in the amount of protein adsorbed to the interface. These two factors increase both FC and FS (Damodaran, 2017). Flours with greater capacity to form foam, as well as greater stability of the formed foam, can be applied in the development of baked products (including gluten free products), as they contribute to better aeration and softness of the final product.

The emulsifying capacity (EC) ranged from 51.32 to 55.16%, and the cultivar BRS Agreste presented the lowest values (p < 0.05) ( $\underline{\text{Table 4}}$ ). These results were lower than those found by Marquezi et al. (2017) for bean flours from different commercial groups (68.28-76.09%). The hydrophilic/hydrophobic balance of amino acids on the protein surface affects the EC. Carbohydrates, notably starch and fiber also assist in catching and binding with oil and/or water improving the EC (Gupta et al., 2018). Thus, the high amount of carbohydrate present in the flours of cultivar Agreste might contribute to its high EC (55.16%). As for the stability of the emulsion, the results found varied between 49.70 and 63.48%, in which the cultivar IPR Uirapuru presented greater stability of the emulsion (p < 0.05) (Table 4). The high emulsion stability of bean flours is due to the nature of their proteins, as albumins are better emulsifiers than globulins (5 mL of phase separation after 780 hours at 21 °C). Therefore, the nature of the protein and the amount of protein influence the stability of the flour emulsion, in addition to the carbohydrate composition. The EC of flours is crucial for bakery products, like cakes, bread, and muffins (Gupta et al., 2018).

The bean flours presented WSI between 26.40% for the cultivar BRS Esteio, and 34.82% for the cultivar BRS Pitanga (Table 5). The cultivars with intermediate values, BRS Estilo, Pérola, and BRS Agreste, did not differ statistically from each other (p > 0.05). The results found are following those reported by Gomes et al. (2015) for raw bean flour (37.46%). The WSI of flour is influenced by factors such as the content of lipids, proteins, and the proportion of amylose and amylopectin. Flours with a higher proportion of amylose have

higher WSI. The presence of proteins with a high degree of hydration, and non-protein substances such as phosphates and lipids also favor the highest WSI (<u>Damodaran, 2017</u>). Therefore, the higher protein content of the Pitanga cultivar flour would justify its higher WSI.

The water absorption index (WAI) of bean flour ranged from 2.93 to 3.93 g g-1 (Table 5). Similar results were found by Gupta et al. (2018). The WAI represents the flour's ability to associate with the water molecule. Water absorption depends on the protein content and fiber content of the sample. Intact protein absorbs the equivalent of its weight in water, and when denatured, it can absorb larger amounts of water due to the alteration of the hydrophilic-hydrophobic balance (Bourre et al., 2019). Fibers, on the other hand, have a high bonding capacity with water and can be responsible for water absorption in up to one-third of the sample's weight (Bourre et al., 2019). On the other hand, the high lipid content in the flour contributes to the reduction of WAI, interrupting the hydration of starch granules with hydrophobic parts. The addition of bean flours in foodstuffs, for example, sausages, custards, and bread may raise the WAI without protein dissolution (Gupta et al., 2018).

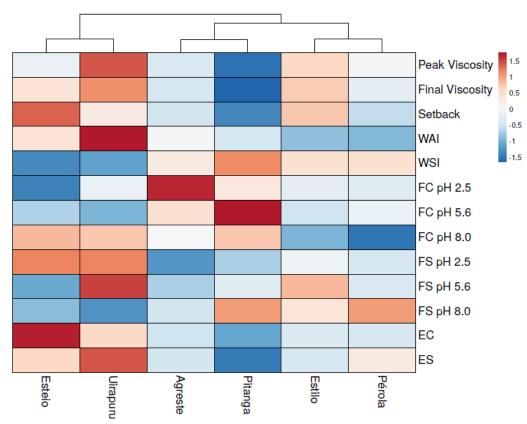
The bean flour of the cultivar IPR Uirapuru had the highest viscosity peak (494.1 cP), when compared to the other cultivars (p < 0.05), and its maximum viscosity was 20.48, 36.08, and 39.32%, respectively, higher than the values presented by the cultivars BRS Estilo and Pérola, do and BRS Esteio (Table 5). On the other hand, cultivar BRS Pitanga presented a viscosity peak 3.6 times lower, when compared to the maximum viscosity presented for the bean flour of the cultivar IPR Uirapuru. Similar values to this cultivar were obtained by Gomes et al. (2015) for raw bean flour, where the authors obtained a maximum viscosity of 415.3 cP. The presence of non-starch components in the bean flours (e.g., proteins, dietary fibers, etc.) reduces the viscosity of the gel because these compounds compete with the starch for water, which results in a weak gel matrix (Romero & Zhang, 2019). This is in accordance with the results since the flours that present high content of protein presented low viscosities (e.g., cultivar Pitanga) (Table 5, Table 1). The maximum viscosity is often correlated with the quality of the final product and also indicates the viscous load that can be encountered during mixing (Damodaran, 2017).

The highest values of final viscosity were observed in the cultivars IPR Uirapuru and BRS Estilo (p < 0.05). On the other hand, cultivar BRS Pitanga presented a final viscosity

Table 5. Solubility in water (WAI), water absorption capacity (WAI), and viscoamylographic properties of bean flours.

Cultivars	WSI (g 100g <sup>-1</sup> )	WAI (g g <sup>-1</sup> )	Peak viscosity (cP)	Final viscosity (cP)	Setback (cP)
BRS Estilo	32.54 <sup>b</sup> ± 0.47	2.96 <sup>de</sup> ± 0.05	392.9 <sup>b</sup> ± 37.9	775.0 <sup>ab</sup> ± 30.6	382.1 <sup>ab</sup> ± 11.8
Pérola	32.54 <sup>b</sup> ± 0.75	2.93 <sup>e</sup> ± 0.07	315.8 <sup>bc</sup> ± 55.9	610.9° ± 67.0	293.7 <sup>cd</sup> ± 25.8
BRS Agreste	$31.90^{b} \pm 0.58$	$3.28^{\circ} \pm 0.10$	270.2° ± 66.7	572.4° ± 83.8	$300.4^{cd} \pm 23.3$
BRS Pitanga	34.82 <sup>a</sup> ± 1.01	$3.11^{d} \pm 0.17$	136.4 <sup>d</sup> ± 43.9	401.6 <sup>d</sup> ± 98.3	264.8 <sup>d</sup> ± 58.6
BRS Esteio	$26.40^{\circ} \pm 0.70$	$3.45^{b} \pm 0.04$	$299.8^{\circ} \pm 65.9$	725.7 <sup>b</sup> ± 92.5	424.1° ± 32.7
IPR Uirapuru	25.00° ± 1.73	3.93° ± 0.08	494.1° ± 96.8	846.0° ± 89.4	350.3 <sup>bc</sup> ± 73.3

<sup>\*</sup> Means followed by the same letter in the column do not differ significantly from each other by the Tukey test (p > 0.05).



**Figure 2.** Heatmap visualization showing the technological properties studied, where red and blue color represent the highest and the lowest values, respectively. WAI: water absorption index, WSI: water solubility index, FC: form capacity, FS: form stability, EC: emulsion capacity, ES: emulsion stability.

value of approximately 2.10 and 1.92 times lower than those presented by cultivars IPR Uirapuru and BRS Estilo (Table 5). The results are comparable with those obtained by Gomes et al. (2015) for raw bean flour (885.7 cP), and smaller than that obtained for raw white bean flour (1294 cP) by Bourre et al. (2019). This difference found in the final viscosity values for bean flours may be due to the difference in the variety of bean grains used, as well as the starch content (mainly amylose content), and non-starch components present in the grains (Romero & Zhang, 2019).

As for the retrogradation trend, the values found ranged from 264.8 to 424.1 cP (<u>Table 5</u>). The cultivars with the greatest tendencies to retrograde were BRS Esteio and BRS Estilo (p < 0.05). Cassava starch has a high tendency to retrograde (2802 cP) (<u>Chandanasree et al., 2016</u>), about six times greater than that of bean flour. Low setback values are desirable when applying to food products such as creamy soups, sauces, bread, and puddings.

The similarities regarding the functional/technological properties between the different bean cultivars were evaluated by principal component analysis with an explained variance of 78.1%. Black bean flours were separated from the others (PC1 = 60%), due to the higher values of viscosity, FS (pH 2.5), EC and ES, and low WSI (Figure 2). Thus, flours of black beans may be suitable for use as a thickening and emulsifying agent in sauces and mayonnaise, for example. On the other hand, the flours made with cultivars BRS Agreste,

BRS Pitanga, and Pérola that presented low viscosity, but high WSI (Figure 2), may be suitable for the development of soups and puddings, for example; because these products need high levels of replacement (with bean flours) without affecting the final texture or the potential of syneresis (Felker et al., 2018).

## Conclusion

All bean flours presented high content of protein (with good digestibility), dietary fibers, and anti-nutritional compounds (phytate, tannins, trypsin inhibitors, and α-amylase inhibitors) might not interfere with their nutritional value since they must be cooked with maceration before consumption. The bean flours are a source of minerals, with highlight to the carioca bean that presented higher amounts of Fe and Zn. The flours also presented various essential amino acids (His, Thr, Tyr, Val, Lys, ILE, Leu, Phe, Met, Cys, and Try). In general, it was possible to identify beans with a high content of essential amino acids (black beans), minerals (carioca beans), protein (purple bean), carbohydrate (pink bean), which reinforce the high nutritional value of common beans and make their flours an ingredient with high nutritional value. The black bean flour showed high viscoamylographic values, WAI, and FC, FS, EC, and ES. On the other hand, the group of special grains (pink and purple beans) had the lowest viscoamylographic values,

EC, and ES. Variation in amylographic profile, WSI, WAI and EC, FC allow the application in the different food systems, such as pasta, biscuits (baked products in general), soup mixes, among others.

## **Compliance with Ethical Standards**

**Author contributions:** Conceptualization: JVP, SGCB, PZB; Formal analysis: JACB; Investigation: JVP, RNC; Resources: SGCB, PZB; Supervision: SGCB, PZB; Validation: JACB, RNC; Visualization: PZB; Writing - original draft: JACB, JVP; Writing - review & editing: JACB, PZB.

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## **Literature Cited**

- Addinsoft. XLSTAT statistical and data analysis solution. New York: Addinsoft, 2021. <a href="https://www.xlstat.com">https://www.xlstat.com</a>. 05 Nov. 2022.
- Akeson, W. R.; Stahmann, M. A. A Pepsin pancreatin digest index of protein quality evaluation. The Journal of Nutrition, v.83, n.3, p.257-261, 1964. https://doi.org/10.1093/jn/83.3.257.
- AOAC International. Official Methods of Analysis of AOAC International. 20.ed. Gaithersburg: AOAC, 2016. 3172p.
- Arnon, R. Papain. In: Perlmann, G. E.; Lorand, L. (Eds.). Proteolytic enzymes: New York, Academic Press, 1970. p.226-244. (Methods in Enzymology, 19).
- Bassinello, P. Z.; Bento, J. A. C.; Gomes, L. D. O. F.; Caliari, M.; Oomah, B. D. Nutritional value of gluten-free rice and bean based cake mix. Ciência Rural, v.50. n.6, e20190653, 2020. https://doi.org/10.1590/0103-8478cr20190653.
- Belmiro, R. H.; Tribst, A. A. L.; Cristianini, M. Effects of high pressure processing on common beans (*Phaseolus vulgaris* L.): cotyledon structure, starch characteristics, and phytates and tannins contents. Starch, v.72, n.3-4, e1900212, 2020. <a href="https://doi.org/10.1002/star.201900212">https://doi.org/10.1002/star.201900212</a>.
- Bento, J. A. C.; Bassinello, P. Z.; Morais, D. K.; Souza Neto, M. A. D.; Bataus, L. A. M.; Carvalho, R. N.; Caliari, M.; Soares Júnior, M. S. Pre-gelatinized flours of black and carioca bean by-products: Development of gluten-free instant pasta and baked snacks. International Journal of Gastronomy and Food Science, v.25, e100383, 2021a. https://doi.org/10.1016/j.ijgfs.2021.100383.
- Bento, J. A. C.; Ribeiro, P. R. V.; Alexandre e Silva, L. M.; Alves Filho, E. G.; Bassinello, P. Z.; Brito, E. S.; Caliari, M.; Soares Júnior, M. S. Chemical profile of colorful bean (*Phaseolus vulgaris* L) flours: Changes influenced by the cooking method. Food Chemistry, v.356, e129718, 2021b. <a href="https://doi.org/10.1016/j.foodchem.2021.129718">https://doi.org/10.1016/j.foodchem.2021.129718</a>.

- Bento, J. A. C.; Ribeiro, P. R. V.; Bassinello, P. Z.; Brito, E. S. D.; Zocollo, G. J.; Caliari, M.; Soares Júnior, M. S. Phenolic and saponin profile in grains of carioca beans during storage. LWT-Food Science Technology, v.139, e110599, 2021c. <a href="https://doi.org/10.1016/j.lwt.2020.110599">https://doi.org/10.1016/j.lwt.2020.110599</a>.
- Bento, J. A. C.; Ribeiro, P. R. V.; Bassinello, P. Z.; Souza Neto, M. A. D.; Carvalho, R. N.; Brito, E. S. D.; Caliari, M.; Soares Júnior, M. S. Functional properties and chemical profile of aged carioca beans and cooked under thesteam of autoclave. Ciência Rural, v.53, n.9, e20220342, 2023. <a href="https://doi.org/10.1590/0103-8478cr20220342">https://doi.org/10.1590/0103-8478cr20220342</a>.
- Bourre, L.; Frohlich, P.; Young, G.; Borsuk, Y.; Sopiwnyk, E.; Sarkar, A.; Nickerson, M. T.; Al, Y. F.; Dyck, A.; Malcolmson, L. Influence of particle size on flour and baking properties of yellow pea, navy bean, and red lentil flours. Cereal Chemistry, v.96, n.4, p.655-667, 2019. https://doi.org/10.1002/cche.10161.
- Chandanasree, D.; Gul, K.; Riar, C. S. Effect of hydrocolloids and dry heat modification on physicochemical, thermal, pasting and morphological characteristics of cassava (*Manihot esculenta*) starch. Food Hydrocolloids, v.52, p.175-182, 2016. <a href="https://doi.org/10.1016/j.foodhyd.2015.06.024">https://doi.org/10.1016/j.foodhyd.2015.06.024</a>.
- Damodaran, S. Amino acids, peptides, and proteins. In: Damodaran, S.; Parkin, K. L. (Eds.). Fennema's food chemistry. 5.ed. Boca Raton: CRC Press, 2017. Chap. 5, 122p. <a href="https://doi.org/10.1201/9781315372914">https://doi.org/10.1201/9781315372914</a>.
- Deshpande, S. S. Food legumes in human nutrition: a personal perspective. Critical Reviews in Food Science and Nutrition, v.32, n.4, p.333-363, 1992. <a href="https://doi.org/10.1080/10408399209527603">https://doi.org/10.1080/10408399209527603</a>.
- Felker, F. C.; Kenar, J. A.; Byars, J. A.; Singh, M.; Liu, S. X. Comparison of properties of raw pulse flours with those of jet-cooked, drum-dried flours. LWT - Food Science and Technology, v.96, p.648-656, 2018. https://doi.org/10.1016/j.lwt.2018.06.022.
- Ferreira, K. C.; Bento, J. A. C.; Caliari, M.; Bassinello, P. Z.; Berrios, J. D. J. Dry bean proteins: Extraction methods, functionality, and application in products for human consumption. Cereal Chemistry, v.99, n. 1, p.67-77, 2021. <a href="https://doi.org/10.1002/cche.10514">https://doi.org/10.1002/cche.10514</a>.
- Gomes, L. D. F.; Santiago, R. D. C.; Carvalho, A. V.; Carvalho, R. N.; Oliveira, I. G.; Bassinello, P. Z. Application of extruded broken bean flour for formulation of gluten-free cake blends. Food Science and Technology, v.35, n.2, p.307-313, 2015. <a href="https://doi.org/10.1590/1678-457x.6521">https://doi.org/10.1590/1678-457x.6521</a>.
- Gupta, S.; Chhabra, G. S.; Liu, C.; Bakshi, J. S.; Sathe, S. K. Functional properties of select dry bean seeds and flours. Journal of Food Science, v.83, n.8, p.2052-2061, 2018. <a href="https://doi.org/10.1111/1750-3841.14213">https://doi.org/10.1111/1750-3841.14213</a>.
- Los, F. G. B.; Zielinski, A. A. F.; Wojeicchowski, J. P.; Nogueira, A.; Demiate, I. M. Beans (*Phaseolus vulgaris* L.): whole seeds with complex chemical composition. Current Opinion in Food Science, v.19, p.63-71, 2018. <a href="https://doi.org/10.1016/j.cofs.2018.01.010">https://doi.org/10.1016/j.cofs.2018.01.010</a>.
- Marquezi, M.; Gervin, V. M.; Watanabe, L. B.; Moresco, R.; Amante, E. R. Chemical and functional properties of different common Brazilian bean (*Phaseolus vulgaris* L.) cultivars. Brazilian Journal of Food Technology, v.20, e2016006, 2017. <a href="https://doi.org/10.1590/1981-6723.0616">https://doi.org/10.1590/1981-6723.0616</a>.

- Mayer Labba, I.-C.; Frøkiær, H.; Sandberg, A.-S. Nutritional and antinutritional composition of fava bean (*Vicia faba* L., var. minor) cultivars. Food Research International, v.140, e110038, 2021. https://doi.org/10.1016/j.foodres.2020.110038.
- Millar, K. A.; Gallagher, E.; Burke, R.; McCarthy, S.; Barry-Ryan, C. Proximate composition and anti-nutritional factors of fava-bean (Vicia faba), green-pea and yellow-pea (*Pisum sativum*) flour. Journal of Food Composition and Analysis, v.82, e103233, 2019. https://doi.org/10.1016/j.jfca.2019.103233.
- Okezie, B. O.; Bello, A. B. Physicochemical and functional-properties of winged bean flour and isolate compared with soy isolate. Journal of Food Science, v.53, n.2, p.450-454, 1988. <a href="https://doi.org/10.1111/j.1365-2621.1988.tb07728.x">https://doi.org/10.1111/j.1365-2621.1988.tb07728.x</a>.
- Price, M. L.; Hagerman, A. E.; Butler, L. G. Tannin content of cowpeas, chickpeas, pigeon peas, and mung beans. Journal of Agricultural and Food Chemistry, v.28, n.2, p.459-461, 1980. https://doi.org/10.1021/jf60228a047.
- Ribeiro, N. D.; Kläsener, G. R. Physical quality and mineral composition of new Mesoamerican bean lines developed for cultivation in Brazil. Journal of Food Composition and Analysis, v.89, e103479, 2020. <a href="https://doi.org/10.1016/j.jfca.2020.103479">https://doi.org/10.1016/j.jfca.2020.103479</a>.
- Ribeiro, N. D.; Londero, P. M. G.; Cargnelutti Filho, A.; Jost, E.; Poersch, N. L.; Mallmann, C. A. Composição de aminoácidos de cultivares de feijão e aplicações para o melhoramento genético. Pesquisa Agropecuária Brasileira, v.42, n.10, p.1393-1399, 2007. https://doi.org/10.1590/S0100-204X2007001000004.

- Rios, M. J. B. L.; Damasceno-Silva, K. J.; Reis Moreira-Araújo, R. S.; DE Figueiredo, E. A. T.; Moura Rocha, M.; Hashimoto, J. M. Chemical, granulometric and technological characterization of integral flours of commercial caupi-beans. Revista Caatinga, v.31, n.1, p.217-224, 2018. <a href="https://doi.org/10.1590/1983-21252018v31n125rc">https://doi.org/10.1590/1983-21252018v31n125rc</a>.
- Romero, H. M.; Zhang, Y. Physicochemical properties and rheological behavior of flours and starches from four bean varieties for gluten-free pasta formulation. Journal of Agriculture Food Research, v.1, e100001, 2019. <a href="https://doi.org/10.1016/j.jafr.2019.100001">https://doi.org/10.1016/j.jafr.2019.100001</a>.
- Shi, L.; Mu, K.; Arntfield, S. D.; Nickerson, M. T. Changes in levels of enzyme inhibitors during soaking and cooking for pulses available in Canada. Journal of food science and technology, v.54, n.4, p.1014-1022, 2017. <a href="https://doi.org/10.1007/s13197-017-2519-6">https://doi.org/10.1007/s13197-017-2519-6</a>.
- World Health Organization WHO. Protein and amino acid requirements in human nutrition: report of a joint WHO/FAO/ UNU expert consultation. Geneva: WHO; FAO; United Nations University, 2007. 265p. (WHO Technical Report Series, 935). https://iris.who.int/handle/10665/43411. 05 Nov. 2022.
- Worku, A.; Sahu, O. Significance of fermentation process on biochemical properties of *Phaseolus vulgaris* (red beans). Biotechnology Reports, v.16, p.5-11, 2017. <a href="https://doi.org/10.1016/j.btre.2017.09.001">https://doi.org/10.1016/j.btre.2017.09.001</a>.