


## RESEARCH ARTICLE

# Effect of high hydrostatic pressure and thermal treatment on polyphenolic compounds and the antioxidant capacity of *Phaseolus coccineus* L.

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## Abstract

**Background and Objectives:** High hydrostatic pressure (HHP) is a nonthermal technology that has been applied to several innovative food products, extending shelf life while preserving sensory characteristics and nutritional value. This technology is feasible to apply to legumes, like beans, to soften them and reduce preparation time. Traditionally, before bean consumption, soaking and cooking processes are carried out to develop palatability; however, this involves a loss of compounds with high biological activity such as dietary fiber, oligosaccharides, and polyphenols, which are of interest due to their antioxidant capacity and related benefits linked to a reduction in the risk of several diseases. Thus, the aim of the study was to evaluate the impact of HHP against traditional cooking on polyphenols and the antioxidant capacity of four *Phaseolus coccineus* L. cultivars: three Mexican (purple, brown, and white) and one Spanish (white).

**Findings:** There was a higher loss of polyphenols in beans where only heat treatment was applied compared to those treated by HHP. The highest antioxidant capacity (1,1-diphenyl-2-picrylhydrazyl and ferric-reducing antioxidant power) was observed in colored samples with a higher content of total polyphenols.

**Conclusions:** The use of HHP decreases the cooking time of beans by 15 min while preserving the polyphenol content and antioxidant properties.

**Significance and Novelty:** HHP can be an alternative for the legume food industry, to reduce cooking time, while preserving or improving their composition and functionality.

## KEYWORDS

antioxidant capacity, high-hydrostatic pressure, *Phaseolus coccineus*, polyphenols

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## 1 | INTRODUCTION

Legume seeds have been an indispensable component of the human diet for centuries. Most cultivated legumes used for food are consumed as grain seeds named pulses. However, some legume species are cultivated to be consumed as vegetables. Both types are also a traditional and prominent component of the “Mediterranean Diet,” one of the characteristics of which is the abundance of plant-based foods (Goñi & Carbajal, 2017).

Among pulses, the runner scarlet bean (*Phaseolus coccineus* L.) is perhaps the second most significant species of *Phaseolus* in the world (Labuda, 2010). *Phaseolus coccineus* L. also known as Ayocote bean is native to central Mexico. In the 18th century, it was introduced into Europe from America. Runner bean is used as dry seeds approximately 2 cm long and comes in white, purple, brown, black, red, and pinto colors. Its cultivation is of importance in some regions of Austria, Spain, Greece, Italy, and Poland; in these countries, the main color of the seed is white. In Spain, the main cultivar, Judi3n de la Granja, is found in La Granja de San Ildefonso (Segovia); it has outstanding culinary qualities, like the softness of its cover and its creamy taste. Its size is bigger than the Mexican Ayocotes, they are white, black, and purple; those with white color are the most commonly used in human nutrition (Urrialde et al., 2019).

Dry seeds of legumes are a remarkable source of many nutrients including proteins or starch, as well as compounds with an important biological activity such as dietary fiber, oligosaccharides, minerals, and vitamins. In addition to their important nutritional value, pulses have an outstanding polyphenol content, which allows them to be considered as possible functional foods due to their antioxidant potential. In vitro and in vivo studies have shown that a diet rich in polyphenols reduces the risk of several chronic diseases (Zhang & Tsao, 2016). Most of the phenolic compounds are concentrated in the seed coats of pulses (Amarowicz & Pegg, 2008; Dueñas et al., 2006; Gan et al., 2016). Phenolic compounds in beans have been reported to show health-positive effects such as antioxidant, anti-inflammatory, antihypertensive, antiatherosclerosis, antitumor, and antiaging (Garcia-Lafuente et al., 2014).

Polyphenols are secondary metabolites of edible plants derived from phenylalanine and tyrosine with the basic structure of phenol (Hervert-Hernández & Goñi, 2011). These show structural diversity ranging from simple phenolics to complex as well as highly polymerized compounds. The high-molecular-weight phenolic compounds having a complex structure are often referred to as polyphenols. Polyphenols, mainly flavonoids and phenolic acids, are bound covalently to

cell wall structure materials such as fermentable fibers as pectin and hemicelluloses phenol (Hervert-Hernández & Goñi, 2011; Siemińska-Kuczer et al., 2022). Part of the ingested polyphenols remains unabsorbed along the gastrointestinal tract, reaching the large intestine being a substrate for the intestinal microbiota metabolism (Wan et al., 2021). This can result in a prebiotic-like effect with positive health repercussions (González-Sarriás et al., 2017). They also can exhibit antioxidant capacity reducing oxidative cell damage. They can act directly by neutralizing free radicals or indirectly by inhibiting the activity of free radical-generating enzymes or by enhancing the activity of intracellular enzymes related to reactive oxygen species. Besides, polyphenols are able to interact with proteins, and at the organoleptic level, they are key determinants of the color, flavor, and taste of foods.

Traditionally, before consumption, pulses undergo a soaking process overnight to improve cooking and to eliminate or reduce some anti-nutritional components (Fernandes et al., 2010). Various heat treatment methods are applied in the preparation of beans to obtain an appealing taste, color, and texture and to enhance their nutritional characteristics (Redondo-Cuenca et al., 2017). Therefore, the choice of preparation method is a major factor that affects the composition and properties of the beans.

In terms of new technological processing methods, high hydrostatic pressure (HHP) is a cold pasteurization technique that involves subjecting products to high levels of pressure through a carrier liquid, usually water. This technology is commonly used because of its great advantages, such as the destruction of pathogens and the increase of the shelf life of the products. In addition, it is known to preserve organoleptic characteristics and the presence of high-value temperature-sensitive compounds. Likewise, it can reduce undesirable alterations affecting the nutritional and sensorial properties of foods, normally produced during traditional heat treatment processes, and helps to preserve the organoleptic characteristics of fresh foods for a longer period (Ferrari et al., 2010). HHP treatment is adjusted by combining three fundamental process parameters such as pressure, temperature, and time; combining all three in different ways allows for great flexibility in the process design to be applied to each food that needs to be treated (Heinz & Buckow, 2010).

The use of HHP before traditional food processing is an innovative technology to keep health-related compounds, improve health attributes of foods by increasing the bioavailability of micronutrients and phytochemicals, reduce allergenic potential, preserve healthy lipids, and reduce salt intake by increasing salt perception (Barba et al., 2017). In addition, the application of HHP to pulses

can reduce processing time, save energy, and avoid the risk of over-processing some parts (Rastogi et al., 2007). In cereals, HHP has been used as an alternative to reduce the allergenicity of rice (Kato et al., 2000). In soybeans, HHP treatment has been effective in reducing the microbial population as well as inactivating enzymes (Préstamo et al., 2000). Recently, Belmiro et al. (2018) demonstrated that the use of HHP in common beans represents a technological improvement in bean processing, affecting both industrial processing (shorter drying time) and consumer processing (shorter hydration and cooking time), without negative effects in the final texture of the grains.

Considering the importance of pulses in human nutrition and their content in polyphenols, the aim of the study was to evaluate the impact of HHP and cooking against traditional cooking on polyphenols content and antioxidant capacity of four *Phaseolus coccineus* L. varieties, three Mexican and one Spanish.

## 2 | MATERIALS AND METHODS

### 2.1 | Materials and chemicals

Four types of *Phaseolus coccineus* L. were tested: three Ayocote beans from Puebla (Mexico), white (WA), purple (PA), and black (BA), and one from La Granja de San Ildefonso (Segovia, Spain), white Judi3n de La Granja (JG) (Figure 1). The beans were kept in closed recipients, protected from light and humidity.

All reagents were of analytical quality.

### 2.2 | Sample preparation and experimental design

Conditions for sample processing were those previously selected by Redondo-Cuenca et al. (2022). Raw beans were soaked in water at a 1:4 w/v ratio for 10 h at room temperature. After removing the soaking water with a sieve,

half of the beans were boiled with 400 mL of boiling water at atmospheric pressure (98°C) for 60 min to obtain the cooked samples (C), and the rest of the soaked seeds were taken to Hiperbaric's facilities (Burgos, Spain) to be submitted to HHP. The optimum conditions for treatment were 600 MPa, 5 min at 25°C. Beans were placed in polyvinyl containers and vacuum-sealed to avoid bubble formation during HHP. Following the HHP step, the samples were boiled for 45 min. The pressure and cooking time conditions for the samples with and without HHP treatment were determined in previous work (Redondo-Cuenca et al., 2022). From five different treatments, they were chosen as optimal once a texture test was carried out, observing statistically similar results in the hardness value (2.68–3.84 N).

After the cooking stage, the beans were drained from the boiling water, lyophilized using a Telstar freeze-dryer (model Lyo Quest), and milled until they passed through a 1 mm sieve.

The samples were labeled according to the treatment applied to each in the following way: R (raw), C (cooked), and HHP + C (HHP + cooked).

### 2.3 | Extractable polyphenols (EP), hydrolyzable polyphenols (HP), and condensed tannins (CT)

The extraction of polyphenols was performed following the procedure of Pérez-Jiménez et al. (2008). 0.5 g of each sample was treated with methanol/water (50:50, v/v; pH 2) and acetone/water (70:30, v/v). Methanolic and acetic extracts were combined and used to determine EP and the antioxidant capacity associated with extractable antioxidants. The residues of these extractions were subjected to two different acidic treatments: methanol/sulfuric acid (20:2, v/v) to determine hydrolyzable tannins and HCl/butanol (5:95, v/v) to know CT (proanthocyanidins). In the extracts obtained previously, the phenolic content and antioxidant activity were determined as described in Sections 2.4 and 2.5, respectively.



FIGURE 1 Image of the beans studied in this work. (a) Judi3n de La Granja (JG). (b) White Ayocote (WA). (c) Purple Ayocote (PA). (d) Black Ayocote (BA). [Color figure can be viewed at [wileyonlinelibrary.com](https://onlinelibrary.wiley.com)]

## 2.4 | Total phenolic content

The content in total phenolic compounds of each extract (EP, HP, and CT) was quantified by the Folin–Ciocalteu colorimetric method (Singleton et al., 1999), and the absorbance was read (Synergy HTX Absorbance Microplate Reader BioTek Co.). Results were expressed as milligrams of gallic-acid equivalents in dry matter (mg GAE/g dm).

## 2.5 | Antioxidant capacity

It was analyzed in the extracts obtained previously following two procedures, 1,1-diphenyl-2-picrylhydrazyl (DPPH) and ferric-reducing antioxidant power (FRAP).

### 2.5.1 | DPPH radical scavenging activities

The DPPH radical scavenging capacity of extracts was measured as described by Ho et al. (2008). Twenty microliters of sample extract was pipetted into wells of a 96-well plate; 200  $\mu$ L of 0.2 mM DPPH methanolic solution was added to each well and the plate was shaken with a plate shaker for 5 min. After 30 min of scavenging reaction at room temperature, the absorbance was measured at 540 nm (Synergy HTX Microplate Reader, BioTek Co.). A standard curve of Trolox was used as control to estimate the antioxidant capacity of the samples. Results were expressed as Trolox equivalents on dry matter ( $\mu$ mol TE/g dm).

### 2.5.2 | Ferric-reducing antioxidant power assay

Benzie and Strain (1996) was used with some modifications (Mengibar et al., 2013; Pulido et al., 2000). Nine hundred microliters of FRAP reagent, containing 2,4,6-tri(2-pyridyl)-s-triazine (TPTZ),  $\text{FeCl}_3$ , and acetate buffer, were mixed with 90  $\mu$ L of distilled water and 30  $\mu$ L of the sample or the blank (solvents used for extraction). Absorbance at 595 nm was measured at 37°C after 30 min (Synergy HTX Microplate Reader, BioTek Co.). A standard curve of Trolox was used to estimate the antioxidant capacity of the samples, and it was expressed as Trolox equivalents ( $\mu$ mol TE/g dm).

## 2.6 | Statistical analysis

All analyses were performed in triplicate and data are shown as mean  $\pm$  standard deviation. A two-factor and a one-factor ANOVA, Duncan's multiple range test, were

performed to test the statistically significant differences ( $p < .05$ ) in the results (SAS version 9 software). The correlation analysis between phenolics and antioxidant capacity was performed with Pearson's correlation test (SAS version 9 software).

## 3 | RESULTS AND DISCUSSION

The nutritional composition of raw, cooked, and HHP + C ayocote beans was studied in previous work (Redondo-Cuenca et al., 2022). It was observed that cooking and HHP + C raised the protein level in beans. Nevertheless, total carbohydrates, minerals,  $\alpha$ -galactosides, and ciceritol were decreased. Fat was without changes after cooking but decreased with HHP + C. In the case of dietary fiber, the observed behavior was different according to the treatment applied and the type of bean. Considering the effect on the nutritional composition, it was of interest to assess the influence of this HHP treatment on the polyphenol level of the samples and their antioxidant capacity.

It is known that the amounts of phenolic compounds with antioxidant properties are generally higher in beans with colored seed coats. The results obtained here confirm this fact in ayocote samples, as indicated below.

### 3.1 | Impact of processing on polyphenol content

The results (Table 1) in raw samples show that the dark-colored varieties, PA and BA beans, had a higher content of extractable phenols (EP) with a lower degree of polymerization, (6.73 and 6.69 mg/g, respectively) than the white varieties, JG (1.56 mg/g) and WA raw beans (1.60 mg/g) in agreement with Corzo-Ríos et al. (2020). The nonextractable polyphenol content with a high degree of polymerization (condensed tannins, CT, plus hydrolyzable polyphenols, HP, that comprise hydrolyzable tannins, phenolic acids, and hydroxycinnamic) was much higher than the EP content in all samples. The nonextractable polyphenol content with a high degree of polymerization as CT, plus HP that comprises hydrolyzable tannins, phenolic acids, and hydroxycinnamic was much higher than the EP content in all samples. CT was also higher in the dark samples as was expected because it is known that legumes with colored seed coats such as lentils, red beans, and black beans have high levels of CT (Amarowicz & Pegg, 2008).

Although the levels of polyphenols in raw legumes are considerable, it is important to know the contents in cooked samples as they are consumed because the

**TABLE 1** Polyphenolic content in the analyzed beans (mg GAE/g dry basis).

Sample	Extractable polyphenols	Hydrolysable polyphenols	Condensed tannins	Total
JG <sub>R</sub>	1.56 ± 0.35 <sup>bA</sup>	4.54 ± 0.29 <sup>bB</sup>	16.4 ± 1.20 <sup>bA</sup>	20.30 ± 2.43 <sup>bA</sup>
WA <sub>R</sub>	1.60 ± 0.34 <sup>bA</sup>	2.57 ± 0.16 <sup>cC</sup>	16.30 ± 0.80 <sup>bA</sup>	20.51 ± 0.94 <sup>bA</sup>
PA <sub>R</sub>	6.73 ± 0.73 <sup>aA</sup>	4.98 ± 0.30 <sup>aB</sup>	19.21 ± 0.40 <sup>aA</sup>	30.70 ± 0.97 <sup>aA</sup>
BA <sub>R</sub>	6.69 ± 0.69 <sup>aA</sup>	5.02 ± 0.10 <sup>aB</sup>	20.40 ± 0.54 <sup>aA</sup>	32.08 ± 1.28 <sup>aA</sup>
JG <sub>C</sub>	0.70 ± 0.10 <sup>cB</sup>	6.32 ± 0.33 <sup>aA</sup>	2.61 ± 0.10 <sup>cC</sup>	9.67 ± 0.43 <sup>cC</sup>
WA <sub>C</sub>	1.20 ± 0.20 <sup>bA</sup>	5.71 ± 0.40 <sup>bA</sup>	2.12 ± 0.25 <sup>dC</sup>	9.09 ± 0.45 <sup>cB</sup>
PA <sub>C</sub>	1.54 ± 0.11 <sup>aC</sup>	6.56 ± 0.16 <sup>aA</sup>	7.11 ± 0.34 <sup>aC</sup>	15.29 ± 0.26 <sup>aC</sup>
BA <sub>C</sub>	1.12 ± 0.07 <sup>bC</sup>	6.07 ± 0.20 <sup>abA</sup>	6.28 ± 0.38 <sup>bC</sup>	13.45 ± 0.42 <sup>bC</sup>
JG <sub>HHP + C</sub>	1.06 ± 0.30 <sup>cB</sup>	6.37 ± 0.27 <sup>aA</sup>	4.90 ± 0.63 <sup>cB</sup>	12.28 ± 1.27 <sup>cB</sup>
WA <sub>HHP + C</sub>	0.40 ± 0.10 <sup>dC</sup>	3.70 ± 0.70 <sup>cB</sup>	4.20 ± 0.70 <sup>cB</sup>	8.26 ± 1.32 <sup>dB</sup>
PA <sub>HHP + C</sub>	2.49 ± 0.42 <sup>aB</sup>	6.59 ± 0.35 <sup>aA</sup>	8.80 ± 0.29 <sup>bB</sup>	17.89 ± 0.14 <sup>bB</sup>
BA <sub>HHP + C</sub>	2.00 ± 0.17 <sup>bB</sup>	5.31 ± 0.23 <sup>bB</sup>	12.00 ± 0.67 <sup>aB</sup>	19.26 ± 0.60 <sup>aB</sup>

Note: Mean ± SD ( $n = 3$ ). Capital letter in superscript per column and variety indicates statistically significant differences, Duncan's test ( $p < .05$ ). Lowercase letter in superindex by column indicates statistically significant differences in the treatment, Duncan's test ( $p < .05$ ). R (raw), C (cooked), and HHP + C (HHP + cooked). Abbreviations: BA, black ayocote; JG, Judión de la Granja; PA, purple ayocote; WA, white ayocote.

physiological effects of polyphenolic compounds are closely related to their degree of bioavailability. Polyphenols in food must be somehow bioavailable to have biological effects (Saura-Calixto et al., 2007), so possibly, the heat treatment promotes the enzymatic release from the food matrix and promotes the bioavailability of polyphenols in the small intestine. The effect of both treatments on the levels found in the extracts is related below.

When cooking and HHP + C treatments were applied, a reduction in the values of EP and CT was observed; the decrease was greater in the case of cooking (55%–83% for EP and between 63% and 87% for CT) compared to the values after HHP + C treatment (32%–70% EP and 42%–74% CT), except in the case of WA subjected to cooking where there was no significant decrease. These results indicate that the HHP process added to a shorter cooking time promotes the rupture of the cell wall of the beans, causing a migration of fluid from the cell to the extracellular medium, extracting a lower amount of water-soluble compounds such as EP and CT, in comparison with the application of prolonged cooking. In other investigations, Belmiro et al. (2020) observed a reduction in tannin content of 30%–40% in common beans after applying pressures of 50 and 600 MPa for 1 and 10 min, while, in another study, the HHP treatment at 200 MPa in applied chickpeas contributed to the reduction of tannin content by up to more than 24% (Alsaman & Ramaswamy, 2020).

On the other side, in HP, an increase in their value was detected in all cases after both treatments. In relation

to the content of the total polyphenol fraction, there was a statistically significant decrease in all samples, being higher after the cooking treatment compared to HHP + C, except for WA, where the decrease was the same in both treatments. López-Martínez et al. (2017) found that the level of phenolic compounds in legumes decreases during the cooking process.

The contents obtained indicate that phenolic compounds were preserved by HHP + C compared to conventional cooking. The results are in accordance with those reported by Akond et al. (2011) and Kan et al. (2017). These losses can be due to the long heat exposure during thermal processing or to the leaching of some of the phenols into the broth due to the softening of the cell walls that occurs during cooking. Aguilera et al. (2011) and Granito et al. (2008) attributed this reduction to the fact that high temperatures can alter the chemical structure of polyphenols, promoting polymerization and/or decomposition of their aromatic structures. The increase in HP could be due to the release of compounds from the food matrix.

The physiological effects derived from polyphenol consumption depend on the quantity and sort of polyphenolic compounds present in the food. Accordingly, phenolic compounds could be physiologically classified based on their solubility in the intestinal environment. A significant part of the EPs is usually soluble in the intestinal environment and would be absorbed in the small intestine. On the contrary, those

with a higher degree of polymerization and, therefore not extractable, reach the colon where they could interact with the colonic microbiota and produce some active metabolites. From the nutritional point of view, as indicated by Saura-Calixto et al. (2007), it is important to consider both fractions (low and high degrees of polymerization) and evaluate how the culinary treatment affects each of them.

### 3.2 | Impact of processing on antioxidant capacity of beans

The potential health benefits of common beans are attributed to the presence of secondary metabolites such as phenolic compounds that possess antioxidant properties. Their antioxidant activity is directly related to their chemical structures such as the degree of glycosylation and number as well as the position of hydroxyl groups attached in relation to the carboxyl functional group. These compounds contribute to antioxidant activity due to their radical scavenging as well as metal-chelating activity (Aguilera et al., 2011).

To assess the antioxidant capacity of the bean samples, two methods were selected, DPPH shows the free radical scavenging abilities of food, while FRAP deals with the reducing power of foods. The results of the

DPPH scavenging activities and FRAP values for the ayocotes are shown in Table 2.

All beans showed antioxidant efficacy assessed by DPPH assay (ranging from 3.87 to 8.38  $\mu\text{mol TE/g dm}$ ), the sample raw purple ayocote has the highest antioxidant activity. The antioxidant activity varied significantly in the order: PA > BA > JG > WA. These results are consistent with those of Golam Masu et al. (2011), who reported that dark beans showed relatively higher levels of antioxidant activity in common beans. Another study (Alvarado-López et al., 2019) found that the highest radical scavenging activity was exhibited by the purple variety followed by black, brown, and white varieties. The process of cooking and HHP + C of colored beans caused significant decreases in DPPH values for EP. JG maintained the values after both treatments, while white ayocote showed a slight increase only after the cooking process. When studying the antioxidant activity of HP and CT, there was an increase in DPPH values in all varieties and both treatments ( $p < .05$ ) except for PA, which was only affected by cooking.

Regarding FRAP values, in the case of EP, the values found were from 12.14 to 88.3  $\mu\text{mol TE/g dm}$ . The results are in accordance with Kan et al. (2017), who analyzed 26 kidney bean cultivars. In all beans, there was a decrease after the treatments applied and this decrease was higher after the cooking treatment ( $p < .05$ ). It was confirmed

TABLE 2 Antioxidant capacity by DPPH and FRAP in raw and cooked beans ( $\mu\text{mol TE/g}$  dry basis).

Sample	DPPH			FRAP		
	Extractable polyphenols	Hydrolysable polyphenols	Condensed tannins	Extractable polyphenols	Hydrolysable polyphenols	Condensed tannins
JG <sub>R</sub>	5.95 ± 0.22 <sup>bA</sup>	5.85 ± 0.08 <sup>cA</sup>	4.13 ± 0.01 <sup>aC</sup>	28.37 ± 1.41 <sup>bA</sup>	4.98 ± 0.14 <sup>cB</sup>	7.74 ± 0.90 <sup>bAB</sup>
WA <sub>R</sub>	5.49 ± 0.13 <sup>cB</sup>	5.54 ± 0.11 <sup>dC</sup>	4.13 ± 0.01 <sup>aC</sup>	23.54 ± 1.98 <sup>bA</sup>	4.38 ± 1.50 <sup>cB</sup>	7.53 ± 0.28 <sup>bB</sup>
PA <sub>R</sub>	8.38 ± 0.04 <sup>aA</sup>	6.95 ± 0.07 <sup>bB</sup>	3.90 ± 0.01 <sup>bC</sup>	88.30 ± 1.41 <sup>aA</sup>	26.99 ± 2.12 <sup>aA</sup>	44.15 ± 0.01 <sup>aA</sup>
BA <sub>R</sub>	8.34 ± 0.01 <sup>aA</sup>	7.09 ± 0.09 <sup>aB</sup>	3.87 ± 0.03 <sup>cB</sup>	80.47 ± 1.70 <sup>aA</sup>	23.18 ± 0.42 <sup>bC</sup>	45.39 ± 2.47 <sup>aA</sup>
JG <sub>C</sub>	6.02 ± 0.10 <sup>aA</sup>	6.46 ± 0.06 <sup>bC</sup>	4.31 ± 0.01 <sup>aA</sup>	12.14 ± 0.40 <sup>dC</sup>	7.47 ± 0.60 <sup>dA</sup>	6.78 ± 0.80 <sup>dB</sup>
WA <sub>C</sub>	6.28 ± 0.44 <sup>aA</sup>	6.43 ± 0.11 <sup>bA</sup>	4.29 ± 0.01 <sup>bA</sup>	17.77 ± 0.80 <sup>cC</sup>	8.99 ± 0.99 <sup>cA</sup>	10.05 ± 0.99 <sup>cA</sup>
PA <sub>C</sub>	5.89 ± 0.43 <sup>aC</sup>	7.86 ± 0.15 <sup>aA</sup>	4.23 ± 0.02 <sup>cA</sup>	20.70 ± 0.85 <sup>bC</sup>	23.51 ± 0.28 <sup>bB</sup>	32.88 ± 2.12 <sup>bB</sup>
BA <sub>C</sub>	5.84 ± 0.10 <sup>aC</sup>	7.89 ± 0.25 <sup>aA</sup>	4.16 ± 0.03 <sup>dA</sup>	23.88 ± 0.60 <sup>aC</sup>	31.58 ± 0.14 <sup>aA</sup>	41.44 ± 1.50 <sup>aB</sup>
JG <sub>HHP + C</sub>	5.84 ± 0.24 <sup>cA</sup>	6.22 ± 0.17 <sup>cB</sup>	4.18 ± 0.01 <sup>bB</sup>	20.13 ± 1.13 <sup>bB</sup>	4.37 ± 0.71 <sup>cB</sup>	8.63 ± 0.28 <sup>cA</sup>
WA <sub>HHP + C</sub>	5.85 ± 0.16 <sup>cAB</sup>	5.73 ± 0.07 <sup>dB</sup>	4.23 ± 0.01 <sup>aB</sup>	21.55 ± 0.28 <sup>bB</sup>	4.28 ± 0.01 <sup>cB</sup>	8.04 ± 0.14 <sup>dB</sup>
PA <sub>HHP + C</sub>	6.67 ± 0.11 <sup>aB</sup>	7.06 ± 0.17 <sup>bB</sup>	4.13 ± 0.02 <sup>cB</sup>	29.24 ± 1.20 <sup>aB</sup>	19.45 ± 0.28 <sup>bC</sup>	45.62 ± 0.49 <sup>aA</sup>
BA <sub>HHP + C</sub>	6.36 ± 0.14 <sup>bB</sup>	7.74 ± 0.22 <sup>aA</sup>	4.18 ± 0.01 <sup>bA</sup>	30.96 ± 1.90 <sup>aB</sup>	24.17 ± 0.70 <sup>aB</sup>	31.72 ± 0.30 <sup>bC</sup>

Note: Mean ± SD ( $n = 3$ ). Capital letter in superscript per column and variety indicates statistically significant differences, Duncan's test ( $p < .05$ ). Lowercase letter in superindex by column indicates statistically significant differences in the treatment, Duncan's test ( $p < .05$ ). R (raw), C (cooked), and HHP + C (HHP + cooked). Abbreviations: BA, black ayocote; DPPH, 1,1-diphenyl-2-picrylhydrazyl; FRAP, ferric-reducing antioxidant power; JG, Judi6n de la Granja; PA, purple ayocote; WA, white ayocote.

that the behavior observed in the colored beans with respect to EP was the same as when the antioxidant capacity was determined by the DPPH. In relation to the FRAP values determined in HP and CT, it was observed that in the white samples after the cooking process, there was a significant increase in both samples, except in the CT of the JG which remained stable. However, the HHP + C treatment retained the initial values except in the JG, where a statistically significant increase was detected. Regarding colored samples, when HP was evaluated, their behavior was different between the two beans; so in the case of PA, there was a reduction in them after both processes, being higher in HHP + C, while in BA, they increased their value, mainly during cooking. For CT, in PA, they only decreased with cooking, and in BA, they decreased in both cases, but especially in HHP + C ( $p < .05$ ). The antioxidant potential of the complex food system is linked to the composition of the active components that contribute to the antioxidant potential and the processing conditions used. Greater stability of bioactive compounds has been observed at high pressure compared to heat treatment, possibly because HHP affects only noncovalent bonds and exerts a minor effect on chemical constituents associated with desirable food quality components, such as flavor, color, and nutritional compounds (Hogan et al., 2005). The results agree with Lee et al. (2018), who observed greater retention of the total content of phenol, flavonoids, and proanthocyanidin by HHP in red beans, which can be attributed to the short duration and residence time of HHP processing. On the contrary, the heat treatment showed a 52.19% decrease in total phenolic compounds and, therefore, lower antioxidant activity.

The highest antioxidant capacity assessed by both methods was observed in the colored samples, with a higher content of total polyphenols. Correlations between the antioxidant activities (DPPH and FRAP) and phenolic contents were analyzed by using Pearson's correlation coefficient ( $R^2$ ). A good correlation was observed in both cases; the correlation values were  $R^2 = 0.9837$  for DPPH and  $R^2 = 0.9881$  for FRAP.

### 3.3 | Relationship between polyphenolic content and antioxidant activity

To illustrate the potential contribution to the antioxidant activity of the total polyphenols in the *Phaseolus coccineus* L., Pearson's correlation coefficients were determined on the relationships between DPPH/FRAP radical scavenging ability and the contents of the metabolite groups (Supporting Information S1: Table 1).

From the analysis, strong direct correlations are observed between the content of EP and the antioxidant activity by both methods,  $R^2 = 0.962$  for DPPH and  $R^2 = 0.983$  for FRAP, and a direct correlation for HP in relation to the DPPH radical ( $R^2 = 0.632$ ). In contrast, the fraction of HP that exhibits high antioxidant activity toward FRAP does not present the same behavior in the correlation analysis, indicating a weak relationship with antioxidant activity ( $R^2 = 0.389$ ). Regarding CT, these decrease after HHP treatment and mostly during cooking. This behavior generates a weak positive correlation with antioxidant activity by FRAP ( $R^2 = 0.405$ ) and a strong negative one for DPPH ( $R^2 = -0.872$ ). The results show that the total phenolic content and CT are not the only contributors to the antioxidant activity. The presence of other compounds in the extracts, such as flavonoids and anthocyanins, can also react straight with free radicals in addition to polyphenolic compounds (Chávez-Mendoza and Sánchez, 2017).

Alvarado-López et al. (2019) studied the antioxidant capacity of an aqueous extract of ayocote beans (purple, black, brown pigmented seed coat, and white seed coat) using the oxygen radical absorbance capacity and DPPH assays in vitro model systems. The results indicated a direct correlation between antioxidant activity with the content of total phenolic content. Juárez-López and Aparicio-Fernández (2012) reported that thermal treatment affected the amount of CT on both studied Mexican varieties ("Flor de junio" and "Peruano"). This may be caused by the destruction of these compounds or by changes in their structure or solubility during thermal treatment causing a decrease in antioxidant activity.

After the cooking and HHP processes, the conservation of polyphenols is sought, which due to their known radical scavenging and transition metal chelating capacity, as Huber (2016) indicated, can counteract the onset and propagation of oxidative processes related to chronic degenerative diseases.

## 4 | CONCLUSIONS

Colored samples had the highest content of polyphenolic compounds compared to white beans. Phenolic compounds were preserved by HHP + C compared to heat treatment alone. The highest antioxidant capacity assessed by DPPH and FRAP was observed in the colored samples, with a higher content of total polyphenols. This activity was affected in different ways depending on the processing and the sample. HHP, at 600 MPa for 5 min at a temperature of 25°C before cooking, decreased cooking time while preserving or enhancing the polyphenolic profile of the beans and their

antioxidant properties. The use of HHP can be an alternative for food enterprises since this technology is useful for reducing cooking time and, therefore, costs and energy.

## ACKNOWLEDGMENTS

The authors acknowledge “Marca de Garantía-Judión de La Granja” (Segovia, Spain) for bean samples and Hiperbaric (Burgos, Spain) for the HHP treatments. A. A. L. thanks to Programa Investigadores e Investigadoras Cátedras COMECYT EDOMEX.

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**How to cite this article:** García-Alonso, A., Alvarado López, A. N., & Redondo-Cuenca, A. (2024). Effect of high hydrostatic pressure and thermal treatment on polyphenolic compounds and the antioxidant capacity of *Phaseolus coccineus* L. *Cereal Chemistry*, 1–9. <https://doi.org/10.1002/cche.10776>