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Wheat bread supplemented with potato peel flour: color, molecular organization, texture and *in vitro* starch digestibility

Pan de trigo suplementado con harina de cáscara de papa: color, organización molecular, textura y digestibilidad *in vitro* del almidón

A. Godoy-Ramirez[1], M. E. Rodriguez-Huezo[2], V.H. Lara-Corona[3], E. J. Vernon-Carter[4], J. Alvarez-Ramirez[4]*
[1] *Departamento de Biotecnología, Universidad Autónoma Metropolitana-Iztapalapa, Apartado Postal 55-534, Iztapalapa, CDMX, 09340 México.*
[2] *Tecnológico Nacional de México, TES de Ecatepec, Departamento de Ingeniería Química y Bioquímica, Av. Tecnológico s/n esq. Av. Central, Col. Valle de Anáhuac, Ecatepec, C.P. 55210, Estado de México, México.*
[3] *Departamento de Química, Universidad Autónoma Metropolitana-Iztapalapa, Apartado Postal 55-534, Iztapalapa, CDMX, 09340 México.*
[4] *Departamento de Ingeniería de Procesos e Hidráulica, Universidad Autónoma Metropolitana-Iztapalapa, Apartado Postal 55-534, Iztapalapa, CDMX, 09340 México.*

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Abstract

In this work the physical characteristics and *in vitro* digestibility of breads made from blends of wheat flour and potato peel flour (PPF), with weight percentage of PPF of 0, 5, 10, 15 and 20 g/100 g were studied. Thermal gravimetric analysis (TGA) showed that the PPF addition accelerated the weight loss with the temperature. Fourier transform infrared (FTIR) analysis revealed PPF induced changes in the bread molecular organization, with an increase of cluster-like water structures, a decrease of the β-sheet protein structures and an increase of amorphous starch arrangements. PPF addition also led to bread with decreased relative volume and increased hardness, which was linked to the disruption of the starch and protein structures. In contrast, starch digestibility showed a marked decrease, of the rapidly digestible starch fraction. A multivariate Principal Component Analysis (PCA) showed that the *in vitro* starch digestibility is linked to the water and starch structures. Overall, the results showed that the addition of up to 10 g of PPF/100 g wheat flour is a viable option for obtaining a bread with decreased starch digestibility and similar overall quality properties of a full wheat bread control.

Keywords: Bread; FTIR; *in vitro* digestibility; potato peel flour.

Resumen

En este trabajo se estudiaron las características físicas y digestibilidad *in vitro* de pan hecho a partir de mezclas de harina de trigo y harina de cáscara de papa (PPF), con porcentajes de PPF de 0, 5, 10, 15 y 20 g/100 g. El análisis termogravimétrico (TGA) mostró que la adición de PPF aceleró la pérdida de peso con la temperatura. El análisis infrarrojo por transformada de Fourier (FTIR) reveló cambios inducidos por PPF en la organización molecular del pan, con un aumento de estructuras de agua en forma de racimos, una disminución de las estructuras de proteínas de la hoja β y un aumento de disposiciones amorfas de almidón. La adición de PPF también dio lugar a un pan con un volumen relativo reducido y una mayor dureza, lo que se relacionó con la alteración de las estructuras de almidón y proteínas. Por el contrario, la digestibilidad del almidón mostró una marcada disminución, principalmente de la fracción de almidón rápidamente digerible. Un Análisis de Componentes Principales (PCA) multivariado mostró que la digestibilidad del almidón *in vitro* está ligada a las estructuras del agua y del almidón. En general, los resultados mostraron que la adición de hasta 10 g de PPF/100 g de harina de trigo es una opción viable para obtener un pan con una digestibilidad reducida del almidón y propiedades de calidad general similares a las de un pan control hecho con solo harina de trigo.

Palabras clave: Pan; FTIR; digestibilidad *in vitro*; harina de cáscara de papa.

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* *Corresponding author. E-mail*: jjar1963@gmail.com;

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1 Introduction

Potato (*Solanum tuberosum* L.) is one of the most important vegetable crops worldwide with a production of about 376 million metric tons in 2021. Potato is consumed in most countries and is eaten in a large diversity of forms, including boiled, cooked, crisped and fried. Commonly, the processing of the raw tuber, particularly the processing chips industry, generates an enormous volume of potato peel (PP) waste, whose improper handling can lead to contamination of soils and water sources due to its microbiological spoilage (Wu, 2016).

Several approaches have been proposed to give a solution to the generation of potato peel (PP) residues. Arapoglou *et al.* (2010) reported that PP wastes have a high potential for ethanol production via enzymatic fermentation. Recently, Ebrahimian *et al.* (2022) showed that PP waste is a reliable alternative to fossil-based products. Awogbemi *et al.* (2022) made a critical review of the usage of PP waste as a low-cost and readily available catalyst and feedstock for biofuel synthesis. Mushtaq *et al.* (2017) noted that PP can be an important source for amylase production. Diverse reports have drawn attention to the rich bioactive compounds of PP waste (Wu, 2016; Sampaio *et al.*, 2020). Elkahoui *et al.* (2018) reported that the relative high contents of proteins (11-18 g/100 g) and relatively low content of fats (non-higher than 2 g/100 g) make the PP an affordable by-product for food and pharmaceutical applications. The content of phenolic compounds in PP, mainly chlorogenic, caffeic and coumaric acids, can be up to ten times higher than in the potato flesh (Rytel *et al.*, 2014; Akyol *et al.*, 2016). Sampaio *et al.* (2020) and Makori *et al.* (2022) highlighted that PP is rich in dietary fiber, phenolic compounds and glycoalkaloids, making the PP waste quite suitable for incorporation in a wide range of food products. Perez-Chabela *et al.* (2022) explored the feasibility of PP flour as a bioactive ingredient for the yogurt production. Fradinho *et al.* (2020) found that potato peel autohydrolysis is a viable way to improve the nutritional characteristics of gluten-free pasta. Azizi *et al.* (2021) proposed the incorporation of PP in the preparation of low-fat and high-fiber potato snacks. Durmaz and Yuksel (2021) incorporated PP flour (PPF) in the formulation of deep-fried wheat chips, finding that total dietary fiber content increased, and the *in vitro* glycemic index decreased. Jacinto *et al.* (2020) reported that PPF is a viable alternative for improving the sensory and nutritional characteristics of gluten-free bread. Ghorbani *et al.* (2022) investigated the incorporation of PPF on biscuit dough rheology and properties. They found that the protein content and color indices increased with the PPF addition, although the cohesion of the biscuit was decreased. Soltan *et al.* (2023) reported that bread fortified by PPF improved lipid profiles in hyperlipidemia.

Despite the proven potential of PP to provide nutritional improvements, studies on the use of PP as a food additive are still scarce. Results in this line should assess the real potential of PP within a sustainable economy plagued by food price inflation in the recent years. In this way, the present work aims to supplement wheat bread with PPF, assessing its impact on the molecular organization, *in vitro* starch digestibility, texture, color and protein content in comparison to a wheat bread control made with only wheat flour.

2 Materials and methods

2.1 Materials

Soft wheat flour (13.1 g moisture, 10.2 g protein, 0.5 g dietary fiber, 0.4 g lipids, 0.8 g ash, and 75 g carbohydrate by difference per 100 g flour), sugar, salt, baking dry yeast (TradiPan S.A. de C.V., CDMX, Mexico) were purchased at a local supermarket (Walmart, CDMX, Mexico). Mature and uniformly sized potatoes (*Solanum tuberasum*, Alpha variety) were collected at a farm located at Metepec, State of Mexico, Mexico. All reagents used were analytical grade. Distilled water was used in all experiments.

2.2 Potato flour preparation

Potatoes were washed with tap water, ensuring that sticking soil and dirt were removed by rubbing with a cloth the potato surface. A Victorinox potato peeler (Ibach, Switzerland) was used for manually peeling the potatoes. The potato peels were washed and soaked in a sodium hypochlorite solution (250 mg/L) for 12 minutes. Afterwards, the peels were dried in a static convection oven (Rational AG, Landsberg, Germany) at 65 ºC for 12 hours to constant weight, and subsequently ground into a fine powder and sieved (Jaipan CM/L-7360065 grinding machine, Japan) to obtain a potato peel flour (PPF) with a particle size of less than 0.2 mm) (Akter *et al.*, 2023). The proximal analysis of the potato peel flour gave the following results: 12.8 g moisture, 6.17 g ashes, 60.06 g total carbohydrates, 13.18 g protein, 2.05 g fat and 5.74 g raw fiber per 100g. The sieved PPF was packed in high-density polyethene (HDPE) bags in an airtight condition and kept in the desiccator at room temperature (25 °C) until required for experiments.

2.3 Bread preparation

Bread preparation was carried out by mixing ingredients in a single step according to the methods of analysis 10-09.01 and 10-10.03 (AACC International, 2010). A homogeneous dry mix made up by 1000 g of flour blend made of wheat and potato peel flours, 75 g of sugar, 15 g of salt and 45 g of bread yeast, were added to 600 g of water was used for obtaining the dough. The ingredients were mixed (Laboratory Spiral Mixer, SP-800-J Alpha Simet Group, Germany) at low speed for 5 min and kneaded for 10 minutes. The dough was allowed to stand for 5 minutes, divided into 300 g portions, placed in silicone containers and leavened in a fermenting chamber for 1 h at 28 °C and 85% relative humidity (Alvarez-Ramirez *et al.*, 2019). Afterwards, the leavened dough was distributed in silicone containers (size 25×10×10 cm) and placed into the aforementioned static convection oven (Rational AG, Landsberg, Germany) at 210 °C, 20% relative humidity for 25 min. After baking, the bread was allowed to cool down at room temperature and stored in hermetic bags. The resulting bread was coded as B_x, where the sub-index "x" denotes the weight percentage of potato peel flour (0, 5, 10, 15 and 20 g/100 g) used to form the flour blend. All breads were made by duplicate.

2.4 Specific volume and moisture content

The breads (B_x) were weighed before being stored in the hermetic bags and had their volume estimated via the rapeseed displacement AACC method 10-05.01. The specific volume was obtained from the ratio between the volume and the weight of the bread. Experiments were performed on three breads of each batch. Moisture content of bread samples was determined before being stored in the hermetic bags using the AACC-method 44-15.02 (González *et al.*, 2021).

2.5 Thermogravimetric analysis (TGA)

TGA analysis was carried out with a TGA Q-50 (TA Instruments, Dallas, TX, USA). The measurements were performed on samples of 10 mg, using a ramp temperature of 5 ºC/min were used for each sample. Weight measurements were made from 25 to 175 °C under a nitrogen flux of 60 mL/min.

2.6 Fourier-transform infrared

Fourier-transform infrared (FTIR) measurements of B_x samples were carried out by following the procedure Reyes *et al.* (2023), with some modifications. A Perkin Elmer spectrophotometer (Spectrum 100, Perkin Elmer, Waltham, MA, USA) endowed with a crystal diamond universal ATR sampling accessory was used. The FTIR spectrum was reported as the mean value of five measurements for each sample. A numerical deconvolution procedure with Gaussian functions (half-width of 15 cm^{-1}, resolution enhancement 1.5) was carried out to obtain individual contributions for distinctive bands.

2.7 Color

The color parameters L*, a* and b*, which are measures of lightness, redness/greenness and yellowness/blueness, respectively, were measured directly on crust samples of B_x with a portable colorimeter in reflectance mode (CR 300, Konica Minolta, Osaka, Japan). Instrument calibration was carried out with a ceramic white tile (L* = 92.49, a* = 1.25, b*= -1.92) and set for illuminant D65, and 2º observer angle (Casas-Godoy *et al.*, 2023).

2.8 Texture

The textural characteristics of hardness, chewiness and adhesiveness of the different B_x were assessed by using a Brookfield CT3-4500 texturometer (AMETEK Brookfield, Middleborough, MA, US) coupled to a TA25/1000 acrylic cylinder probe with size 50.8 mm diameter and 20 mm length. Cylindrical samples of 2.0 cm diameter were obtained from the geometrical center of the bread loaves and were compressed to reach 50% deformation in two cycles (González *et al.*, 2021).

2.9 Starch in vitro digestibility

The *in vitro* digestibility of the B_x was done as described by Alvarez-Ramirez *et al.* (2020). The rapidly digestible starch (RDS), slowly digestible starch (SDS) and resistant starch (RS) contents were analyzed based on the following formula:

$$RDS = \frac{G_{20} - FG}{TS} \times 0.9 \tag{1a}$$

$$SDS = \frac{G_{120} - G_{20}}{TS} \times 0.9 \tag{1b}$$

$$RS = \frac{TS - RDS - SDS}{TS} \times 100 \tag{1c}$$

Here, G_{20} and G_{120} denote the content of glucose released at 20 and 120 min, respectively, FG is the free glucose content and TS is the total glucose released after complete (i.e., very long times) hydrolysis.

2.10 Statistical analysis

Experimental data was analyzed by means of one-way analysis of variance (ANOVA) and a Tukey's test for a statistical significance ($p < 0.05$). The analysis was obtained with the aid of the package SPSS Statistics 19.0. All the experimental measurements were done in triplicate, unless otherwise stated.

Table 1. Relative volume, moisture content and color parameters of the bread with different potato peel flour contents.

Bread Code (Bx)	Relative Volume	Moisture (%)	L*	a*	b*
B0	1.0 ± 0.00^a	17.83 ± 1.25^a	28.99 ± 1.98^a	28.59 ± 0.23^a	24.38 ± 2.16^a
B5	0.96 ± 0.03^b	16.82 ± 1.47^a	26.73 ± 1.75^b	26.44 ± 0.27^b	23.74 ± 1.51^a
B10	0.94 ± 0.02^c	16.59 ± 1.38^a	24.98 ± 1.67^c	22.42 ± 0.77^{bc}	22.34 ± 1.27^{ab}
B15	0.91 ± 0.02^c	16.28 ± 1.14^a	22.59 ± 0.64^c	22.01 ± 0.91^{bc}	20.34 ± 0.33^b
B20	0.87 ± 0.02^c	14.76 ± 1.26^b	21.26 ± 0.29^{cd}	20.15 ± 1.09^c	19.15 ± 0.24^b

Values are reported as means ± standard deviation. Column with different lower-case letters in columns indicate significant differences (p<0.05). Bx means bread with "x" % of potato peel flour (PPF).

3 Results

3.1 Specific volume, moisture content and color

As the content of PPF in the B$_x$ increased the specific volume decreased (Table 1), with B$_{20}$ experimenting a decrease of about 13%. This effect may be due to the *de facto* reduction of the gluten content in the blended flour, as potato starch is gluten free (Ali *et al.*, 2023), causing a relative weaking of the network formed between proteins and carbohydrates. Although the PPF contains a relatively high content of proteins (13.18 g/100 g), its characteristics are not as those of gluten, such that the resulting ability for bread expansion during baking is negatively affected. The moisture content also decreased with the PPF addition, ranging from of 17.83 g/100 g for B$_0$ to 14.76 g/100 g for B$_{20}$. The moisture content and the specific volume were strongly correlated (0.91, significance = 0.031), which suggests that the decrement of the specific volume is likely linked to the loss of moisture in the bread structure. The color parameters of the bread crust of B$_x$ are given in Table 1. The color parameters L*, a* and b* tended to exhibit significant decreases as the PPF content in B$_x$ increased. The decrease in the color parameters between B$_0$ and B$_{20}$ was of 28.99 to 26.73 for L*, 28.59 to 26.44 for a*, and 24.38 to 23.74 for b*, respectively. This shift in the color parameters towards lower values can be ascribed to the relatively large fraction of ashes (6.17 g/100 g) contained by PPF. The ash content for all purpose wheat flour has been reported as being on average much lower (0.4 g/100 g) than for potato flour (3.6 g/100 g) (Pu *et al.*, 2017).

3.2 TGA

The weight loss profile of the B$_x$ in the temperature range from 25 to 175 ºC is presented in Figure 1.a. The weight loss showed an exponential-like decay, with a faster decay when the PPF was incorporated in the bread formulation. The weight loss trajectory reflects the loss of moisture during the baking process (Fessas and Schiraldi, 2001). Water molecules are bound to the bread structure, where the gluten plays an important role. The reduction of the effective content of gluten by dilution in the flour blends, weakened the ability of the doughs obtained from them to retain the moisture as the temperature was increased. Zhou *et al.* (2016) identified three different regimens of water loss in the TGA profile of wheat bread. The first one takes place at relatively low temperatures of 40-60 ºC and is attributed to the loss of easily removable water. The second regime is at about 80-100 ºC and is ascribed to more physically entrapped water, whereas a third regime at 100-140 ºC is linked to water that is more tightly combined with gluten and therefore requires more energy to be removed (Fessas and Schiraldi, 2001; Vodovotz *et al.*, 1996). The first derivative of the weight loss profile (Figure 1.b) indicates that the higher moisture content loss was exhibited in the first baking stage, at relatively low temperatures, and that the weight loss was more pronounced as the PPF content was increased.

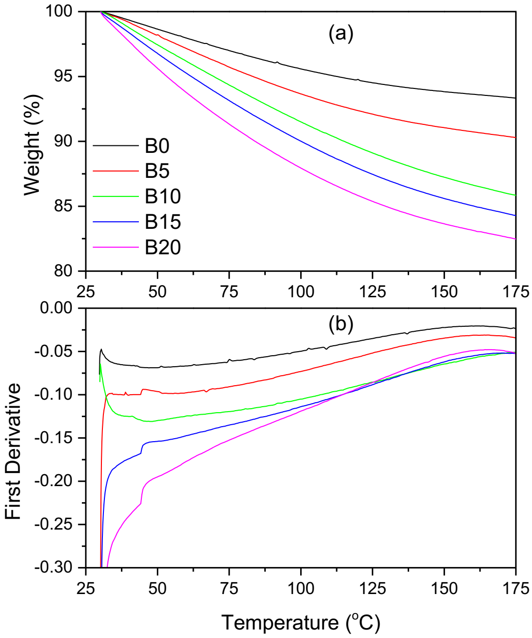

Figure 1. Weight loss of breads (Bx) for a heating rate of 5 °C/min (a). First derivative of the weight loss profile (b). Bx means bread with "x" % of potato peel flour (PPF).

Table 2. Texture parameters for the bread with different potato peel flour contents.

Bread Code (Bx)	Hardness (N)	Chewiness (mJ)	Adhesiveness (mJ)
B_0	4.12 ± 0.20^{d}	15.74 ± 1.25^{cd}	0.22 ± 0.01^{d}
B_5	4.32 ± 0.16^{cd}	16.73 ± 1.47^{c}	0.24 ± 0.01^{cd}
B_{10}	4.72 ± 0.21^{b}	18.46 ± 1.38^{b}	0.28 ± 0.01^{c}
B_{15}	4.86 ± 0.22^{ab}	19.22 ± 1.14^{ab}	0.39 ± 0.01^{b}
B_{20}	5.09 ± 0.21^{a}	23.76 ± 1.26^{a}	0.47 ± 0.02^{a}

Values are reported as means ± standard deviation. Column with different lowercase letters in columns indicate significant differences ($p<0.05$). Bx means bread with "x" % of potato peel flour (PPF).

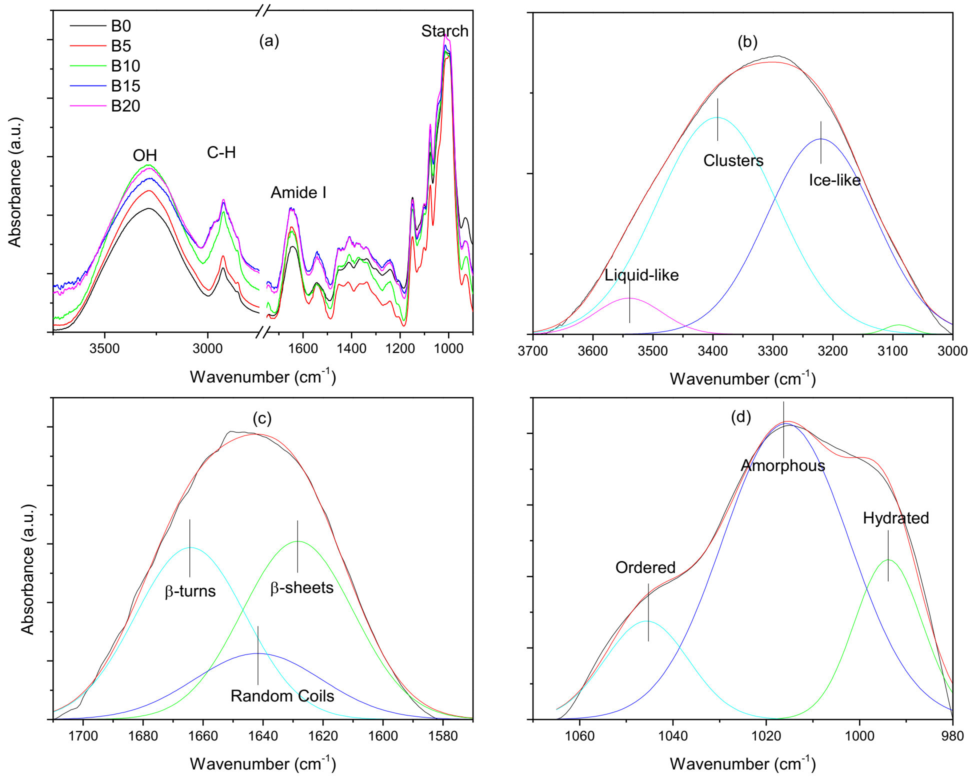

Figure 2. FTIR spectra of the breads (Bx) with different potato peel flour contents (a). Illustrative example of the numerical deconvolution of the control bread (B0) made with only wheat flour: (b) the OH, (c) the Amide I, and (d) the starch regions. Bx means bread with "x" % of potato peel flour (PPF).

3.3 *Texture*

Hardness, chewiness, and adhesiveness (Table 2) increased significantly as PPF content in the B_x was higher. This behavior may be attributed to the decreasing moisture content and relative volume loss undergone by the B_x as PPF content was increased. Hardness was correlated (0.87, significance = 0.032) with the moisture content. Meral and Karaoğlu (2020) found that the addition of stale bread flour increased bread hardness due to the aggregation of flour particles with the concomitant reduction of moisture content. In our case, we hypothesized that the decreased value in the measured textural properties is more likely to be associated with the reduced gluten content in the flour blends, which resulted in a less homogeneous network formation and a decreased moisture content (Wang *et al.*, 2015).

3.4 *FTIR analysis*

The FTIR spectrum of the crumb for the B_x are presented in Figure 2.a. The band 3750-3000 cm^{-1} is linked to the OH band and reflects the interaction of water molecules with bread components (proteins and carbohydrates). The region 1700-1600 cm^{-1} corresponds to the Amide I band caused by the stretching of the C=O group. The fingerprint region with a large peak at about 1020 cm^{-1} is commonly considered a fingerprint region of the starch molecular organization. The above-mentioned bands provide valuable insights on the molecular organization and the impact of the PPF addition of the bread, as is discussed in more detail below.

3.4.1 Water structure

Water molecules interact with the bread components, largely determining the properties of the bread. Water molecules in hydrogels can be present in different forms, and can be classified as free, freezing bound and non-freezing bound structures (Garcia *et al.*, 2004). Walrafen *et al.* (1986) proposed a model structure of water molecules, which can be detected by an analysis of the OH band in the FTIR spectrum. It was considered that the OH-band was composed of five overlapping individual factors detected at about 3090, 3220, 3393, 3540 and 3625 cm^{-1}. The band at 3090 cm^{-1} was linked to a Fermi resonance of the overtone of OH-in-plane bending with the OH-vibration of strongly hydrogen-bonded structured water. The presence of the peak at this band reflects the existence of highly structured water, and the reduction of its intensity reflects the decomposition of such structures in smaller ones with weakened hydrogen binding. Figure 2.b illustrates the results for B_0 of the numerical deconvolution of the OH band in five contributions having Gaussian shape. The band at 3220 cm^{-1} was ascribed to fully bonded water of low density exhibiting an ice-like structure (coordination number close to four), while the band at 3400 cm^{-1} was linked to water clusters displaying an average degree of connection greater than for dimers and trimers. The contribution at 3540 cm^{-1} reflects the presence of water molecules with a poor binding to their surrounding liquid-like structures. Finally, the peak at the highest wavenumber of 3625 cm^{-1} indicates disorganized molecules with a vapor-like structure (Baumgartner *et al.*, 2019). Figure 3.a presents the variations of the water structure for the different bread formulations incorporating PPF. The PPF addition decreased the strongly structured water molecules (ice-like structures) and increased the formation of clusters. Significant differences ($p < 0.05$) were exhibited among the different bread formulations, which suggests that PPF impacted the molecular organization of water probably via disruptions of the gluten organization and to the interaction of the PPF components with the starch. The liquid-like structures vanished for relatively high fractions (B_{15} and B_{20}) of PPF. A strong correlation (0.943, significance = 0.004) was found between bread hardness and cluster relative content.

3.4.2 Protein secondary structure

The secondary structure of proteins has an important impact in the properties of wheat bread (Sivam *et al.*, 2012). The Amide I band located at 1700-1600 cm^{-1} can be numerically deconvoluted to extract information on the protein secondary structure. Figure 2.c shows that the Amide I band can decomposed in three contributions linked to β-structures and random coils. Bock *et al.* (2013) observed that the β-structures are determinants of the bread quality. Figure 3.b shows that the addition of the PPF decreased the content of β-structures while increasing the presence of random coils. Bread containing PPF exhibited increased values of hardness and chewiness (Table 2), which were negatively correlated (-0.876, significance = 0.008) with the decrease of the β-structures. Wang *et al.* (2001) and Miñarro *et al.* (2012) reported that the β-structures have a positive impact in the elasticity of gluten. The decrease of these structures with the PPF addition could be linked to the increased values of bread hardness. On the other hand, the increase of the random structures may be related to the reduced water mobility during the baking process and weakened gluten structure. In this way, the addition of PPF had the effect of weaking the gluten structure, resulting in compact bread morphologies like that exhibited by bread made with gluten-free flours (Singh *et al.*, 2016).

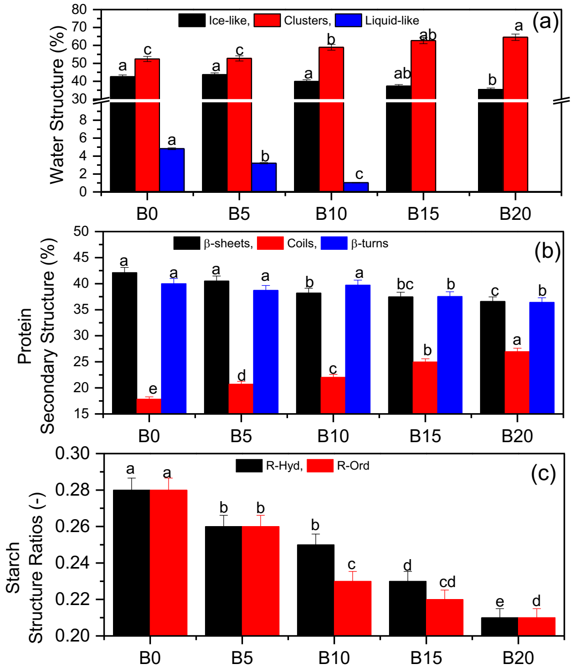

Figure 3. Molecular organization of the breads (Bx) with different content of potato peel flour. Bx means bread with "x" % of potato peel flour (PPF).

3.4.3 Starch structure

The FTIR band with a large peak at about 1020 cm^{-1} is commonly considered a fingerprint of the molecular organization of starch chains. van Soest *et al.* (1995) postulated that the starch band reflects the overlapped contribution of three different structures.

Table 3. *In vitro* starch digestibility of the bread with different potato peel flour contents.

Bread Code (Bx)	Total Starch (g/100 g)	RDS (%)	SDS (%)	RS (%)
B_0	63.24 ± 1.54^a	53.74 ± 0.34^a	24.12 ± 0.27^c	22.14 ± 0.18^b
B_5	61.45 ± 1.72^a	52.19 ± 0.37^{ab}	24.23 ± 0.28^c	23.58 ± 0.22^{ab}
B_{10}	59.32 ± 1.33^{ab}	51.14 ± 0.36^b	25.64 ± 0.25^c	23.22 ± 0.36^{ab}
B_{15}	57.78 ± 1.06^b	46.32 ± 0.44^c	27.88 ± 0.22^b	23.70± 0.28^a
B_{20}	55.14 ± 1.15^c	46.51 ± 0.47^{cd}	29.48 ± 0.25^a	24.01 ± 0.31^a

Values are reported as means ± standard deviation. Column with different lower-case letters in columns indicate. RDS: Rapidly digestible starch, SDS: Slowly digestible starch; RS: Resistant starch. Bx means bread with "x" % of potato peel flour (PPF).

The band at 995 cm^{-1} is ascribed to the presence of hydrated amylose domains, the shoulder at 1022 cm^{-1} is due to amorphous and poorly ordered starch arrangements, and the band at about 1047 cm^{-1} indicates the presence of ordered (e.g., double- and triple-helix) structures. Figure 2.d illustrates the deconvolution of the large peak into the three mentioned individual peaks. van Soest *et al.* (1995) proposed that, rather than the individual contribution of these bands, the starch structure can be better represented by the ratios 995/1022 (R_{Hyd}) and 1047/1022 (R_{Ord}), which represent the fraction of hydrated and ordered structures relative to amorphous ones. Figure 3.c shows that both ratios R_{Hyd} and R_{Ord} decreased with the PPF content. That is, the addition of PPF disrupted the molecular organization of starch by increasing the relative content of amorphous and poorly ordered structures. It was found that the ratios R_{Hyd} and R_{Ord} were positively correlated (0.923, significance = 0.002) with the content of protein random structures. That is, the addition of PPF deteriorated the organization of both proteins and starch in the bread formulations. The formation of such complex structures limits the accessibility of amylolytic enzymes to the starch chains, reducing in this form the starch digestibility (Giuberti *et al.*, 2020).

3.5 *Starch in vitro digestibility*

The total starch showed a slight decrease with the addition of PPF (Table 3), from values of 63.24 g for B_0 to about 55.14 for B_{20} per 100 g. This result was expected since the starch content of PFF is relatively low. The RDS content also decreased, from 53.74% for B_0 to about 46.51% for B_{20}. In contrast, the SDS fraction increased with the PFF content. However, the RS fraction was scarcely impacted by the addition of the PFF. The results in Table 3 show that the PFF interfered with the digestibility of starch in the bread formulations by slowing enzymatic hydrolysis. The components of the PFF acted as fiber additives, which acted as physical barrier that inhibited starch digestion limiting enzyme mobility. Zhu *et al.* (2022) reported that cellulose acts as a fiber that inhibits the access of amylolytic enzymes to starch chains, leading to a moderate decrease of the starch digestibility in wheat bread. On the other hand, PP has a rich content of phenolic compounds, which interact with amylose to form inclusion complexes (Zhu, 2015). Recently, Chen *et al.* (2019) reported that mango peel had the ability of reducing the digestibility of bread, ascribing this effect to the relative high content of polysaccharides and polyphenols of mango peel. Giuberti *et al.* (2020) stated that starch digestibility of products derived from flours with high polyphenol contents could be modulated either by direct inhibition of amylolytic enzymes and/or the formation of inclusion and non-inclusion complexes with starch.

3.6 *Multivariate analysis*

The results described above indicated that the addition of PFF impacted textural and digestibility properties of wheat bread. A Principal Component Analysis (PCA) was carried out to gain insights on how the different response variables were correlated. Seventeen response variables were considered: ice-like (IL) and cluster (CL) water structures, β-sheets (BS) and coils (CO) protein secondary structures, hydrated (RH) and ordered (OR) starch FTIR ratios, relative volume (RV), moisture (MO), lightness (L), redness (A) and yellowness (B), hardness (HA), chewiness (CH) and adhesiveness (AD), total starch (TS), rapidly digestible (RDS) and slowly digestible (SDS) starch. Some variables were not considered, for instance the resistant starch (RS) fraction, as it was obtained as RS = 100 - RDS - SDS, such that its impact in the multivariate analysis is already considered in the RDS and SDS variables. The first and second principal components accounted for 71.65 and 12.74% of the total variance, and together represented 84.39% of the total variance (Figure 4.a). This means that the seventeen response variables are strongly correlated and the impact of the PPF addition can be assessed by few response variables. For instance, ice-like water structures, lightness, total starch, relative volume, and hydrated starch structures are strongly aligned with the rapidly digestible starch fraction. This suggests that water molecules in the hydrated starch structures are arranged in the form of ice-like structures. On the other hand, adhesiveness,

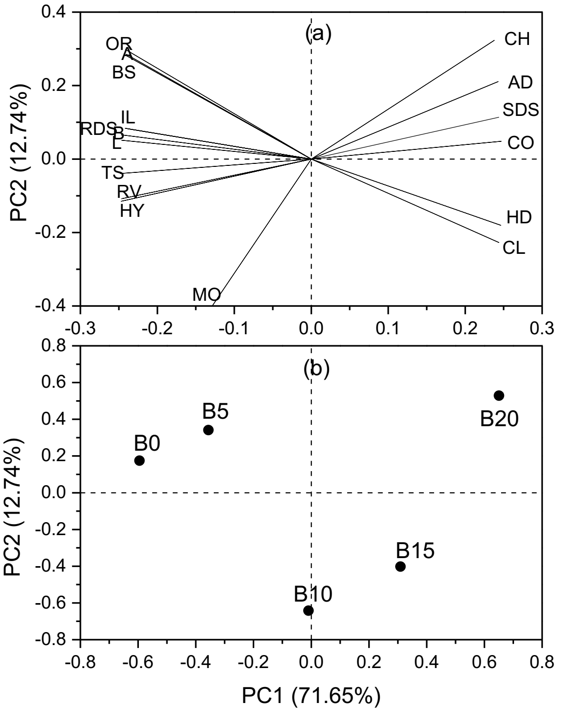

Figure 4. Principal component analysis of the response variables for the breads (Bx). (a) Score plot of the response variables. (b) Score plot of the bread formulations. Bx means bread with "x" % of potato peel flour (PPF).

protein coils and chewiness are aligned with the slowly digestible starch fraction. Hardness is aligned with water cluster structures. Three clusters can be observed in the bread formulations (Figure 4.b). B_5 which contained a relatively small PPF content exhibited properties that were very close to those exhibited by the wheat control bread (B_0). Breads with intermediate PPF contents (B_{10} and B_{15}) exhibited properties that were significantly (84.39%) different from that of the control bread (B_0) The characteristics of the bread B_{20} exhibited important deviations from the other four formulations.

Conclusions

This work studied the impact of PPF on the characteristics and digestibility of wheat bread. The results showed that low additions (5 g/100 g) of PPF produced a bread with texture, color and digestibility similar to the whole-wheat bread. Higher addition of PPF led to bread with marked differences with the control bread, which was ascribed to the several factors which included a lower gluten content in the flour blends which resulted in the formation of a relative weaker bread microstructure, which resulted in breads with lower moisture content. This on turn affected the way in which water interacted with the bread components, inducing changes in the secondary structure of proteins and the molecular organization of starch, impacting the bread properties such as the textural characteristics and starch *in vitro* digestibility. In addition to this, the relative high content of polyphenols of PPF, may contribute to modulate starch digestibility by either inhibiting directly the action amylolytic enzymes and/or by forming inclusion and non-inclusion complexes with starch. In general, the PPF addition increased hardness, chewiness and adhesiveness, but decreased total starch and increased the relative content of slowly digestible starch fraction at the expense of the rapidly digestible starch fraction. Overall, this study demonstrated that PPF has a good potential for its use as a nutritional improver in the preparation of bakery products.

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