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Functional and textural properties of a dehulled oat (*Avena sativa* L) and pea (*Pisum sativum*) protein isolate cracker

E. Morales-Polanco [a], R. Campos-Vega [a], M. Gaytán-Martínez [a], L.G. Enriquez [b], G. Loarca-Piña [a, *]

[a] Posgrado en Ciencia y Tecnología de los Alimentos, Research and Graduate Studies in Food Science, School of Chemistry, Universidad Autónoma de Querétaro, Centro Universitario, Santiago de Queretaro, C.P. 76010, Qro, Mexico
[b] Roquette Food Company Mexico S.A. de C.V., Queretaro, C.P. 76042, Mexico

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ABSTRACT

In this study, the nutritional, antioxidant and physical properties of a cracker made from dehulled oat flour (*Avena sativa* L) and pea (*Pisum sativum*) protein isolate (COP) was investigated. The COP was compared against two commercial crackers, showing a higher nutritional content, emphasizing its high value of protein (24.66 g/100 g cracker), total fiber (18.45 g/100 g cracker) insoluble fiber (13.05 g/100 g cracker), vanillin (0.932 μg/100 g cracker), *p*-cumaric (0.861 μg/100 g cracker) and avenantramide (1.160 μg/100 g cracker) as well as the low content of lipids (9.07 g/100 g cracker), carbohydrates (62.13 g/100 g cracker), total phenolic compounds (0.42 mgGAE/g cracker) antioxidant capacity DPPH (26.93 μmol eq. Trolox/g cracker) and ABTS (171.61 μmol eq. Trolox/g cracker). Certain differences were also found in textural properties, the COP exhibited lower hardness (19.04 N), and gumminess (4.07 N), and higher values of cohesiveness (0.35), springiness (0.45 mm), and chewiness (0.35). Based on these results, dehulled oat and pea protein isolated crackers have the potential to confer health benefits.

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1. Introduction

The manufacturing of processed foods (i.e. snacks) requires the development of new products according to the consumer's preferences and needs, such being an opportunity to incorporate bioactive compounds related to human health benefits as part of the formulations. Cookies and crackers are considered some of the most popular low-moisture baked goods made with soft wheat flour in the United States and Mexico (Kweon, Slade, Levine, & Gannon, 2014). Cookies and crackers have become the most consumed snacks amongst young and adult people due to their low manufacturing cost, convenience, long shelf-life and ability to serve as a vehicle of important nutrients. Furthermore, the consumption of bakery products has been increasing as a result of urbanization and growth of the working female population (Thivani, Mahendran, & Kanimoly, 2016). Nowadays, consumers are more concerned about their health and demand food products that provide health benefits with reduced calories, high protein, dietary fiber content; there is also a trend to increase the intake of natural products rather than foods that contain synthetic additives (Stöckli, Stämpfli, Messner, & Brunner, 2016).

Moreover, evidence of diseases such as high blood pressure, diabetes, cardiovascular diseases (CVDs) and among other illnesses due to lifestyle changes, are on the rise (Xavier *et al.*, 2016). The combination of cereals and legumes has been related to the prevention and/or reduction of non-transmissible diseases due to their bioactive compounds such as dietary fiber, phenolic compounds, protein, phytosterols, among others. These compounds have shown antimutagenic, hypocholesterolemic, hypoglycemic and anticarcinogenic properties (Patil, Brennan, Mason, & Brennan, 2016). Besides protein concentrates and isolates have been recently used by the food industry, mostly derived from soybeans, barley, and wheat. However, due to dietary restriction such as allergies and consumer preferences, the food industry is looking for alternate sources of proteins (Toews & Wang, 2013). Pea protein as a concentrate or as an isolate can be an alternative because of its nutritional quality and ability to provide desirable sensory properties such as structure, texture, taste, and color to formulate food products (O'Sullivan, Murray, Flynn, & Norton, 2014). Noteworthy, oat (*Avena sativa* L.) is the only food recognized as nutraceutical by the U.S. Food and

* Corresponding author.
E-mail address: loarca@uaq.mx (G. Loarca-Piña).

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Drugs Administration (FDA) due to its role in the prevention of coronary diseases mainly (Shah, Masoodi, Gani, & Ahmad Ashwar, 2016). According the FDA a functional food is defined as: "… a part of the usual diet to have helpful effects that go beyond elementary nutritional role …" (Martirosyan & Singh, 2015). Functional food can be enriched with ingredients that usually are not present in that particular food. Similarly, the European Food Safety Authority (EFSA) authorizes the labeling of food products with health claims (Mannarino, Ministrini, & Pirro, 2014). Bakery products are considered acceptable diet agents for health and strength (Deepika, 2016). There are some reports related to gluten-free baked products; however, the information on gluten-free crackers made from pea protein and oat flour is rarely found. The information at hand is still limited, particularly regarding the comparative research with commercial products. Knowing the differences between our formulation and the commercially available products provides relevant information about the potential acceptability by the consumer. The aim of this study was to examine physical hardness, resilience, fracturability, cohesiveness, springiness, gumminess, chewiness, physicochemical characteristics, protein, lipids, and carbohydrate and nutritional (TFD, IDF, SDF, RS, antioxidant capacity and free phenolic compounds) properties of an oat (*Avena sativa* L) and pea protein (*Pisum sativum*) isolate crackers as a novel alternative functional snack.

2. Materials and methods

2.1. Ingredients and cracker preparation

Dehulled oat grain (*Avena sativa* L.) was obtained from a local market of Queretaro (Mexico). One hundred grams of dehulled oat grain was milled to a fine powder using a coffee grinder (KRUPS GX4100, Mexico) and sieved through a Montinox 40 mesh (0.42 mm) screen, this material was called oat flour (OCF). Commercial pea (*Pisum sativum*) protein isolate (PPI) Nutralys®F85M was supplied by Roquette (Frères S.A., Lestrem, France). The preparation of the crackers was made based on the standard method AACC (10-50D) (Gaines & Tsen, 1980). To obtain the base formulation, OCF (80%) and PPI (20%) were used as major ingredients in the formulation. The dough was kneaded and sheeted to a uniform thickness and cut into square shapes (10 cm^2). The baking was done at 180 °C for 10 min in a forced-air convection oven; the oven was equipped with a turbo fan system for heat distribution and the multi-flame system. The characteristics from the oat/pea protein isolate crackers (COP) were compared to two types of wheat crackers: commercial crackers (CC1: carbohydrate: 63 g/100 g cracker, lipids: 20 g/100 g cracker and protein: 7 g/100 g cracker; according to the commercial label) and commercial crackers with reduced lipid content (CC2: carbohydrate: 72 g/100 g cracker, lipids: 6 g/100 g cracker and protein: 11 g/100 g cracker; according to the commercial label).

2.2. Proximate composition

AOAC procedures were used to determine moisture (method 925.10), lipid (method 920.39), ash (method 923.03), and nitrogen (method 920.87) contents of the OCF, PPI, COP, CC1 and CC2 samples (AOAC, 2002). The moisture was determined by the method AACC 44–16.01. The nitrogen content was determined by using the Kjeldahl method, with sodium sulfate as a catalyst. The protein content was calculated as nitrogen x 5.83 for OCF and 6.25 for PPI, OCP, CC1 and CC2. The lipid content was obtained from Soxhlet extraction for 6 h with petroleum ether and the ash content was calculated accordingly by the method AACC 30–25.01, 942.05, respectability (AACC, 1995).

2.3. Nutraceutical composition

2.3.1. Total dietary fiber (TDF)

The dietary fiber fractions, containing soluble dietary fractions (SDF) and insoluble dietary fractions (IDF) were determined following AOAC method 991.43 (AOAC, 2002). One gram of each sample (OCF, PPI, COP, CC1, and CC2) was added to 50 mL phosphate buffer pH 6. The samples were placed in a water bath at 100 °C, and 0.1 mL of α-amylase solution was added, and incubated for 30 min stirring every 5 min. The samples were rapidly cooled and added with 0.1 mL of protease, and placed in a water bath at 60 °C for 30 min. Afterwards, the pH was adjusted at 4, the samples were placed in a water bath at 60 °C for 30 min, and 0.3 mL of amyloglucosidase was added. The samples were incubated for 30 min under constant agitation and then diluted with 95% of ethanol (1:4 ratios), then the mixtures were left at room temperature for 24 h. Such samples were filtered at a constant weight and the residues washed three times with 10 mL of distilled water. The residues were placed in an oven at 90 °C for 2 h and weighed. The TDF was determined gravimetrically and considered as polysaccharide extract (PE). To quantify IDF, the ethanol was not added. The SDF was calculated by subtracting the IDF proportion from TDF.

2.3.2. Resistant starch (RS)

The resistant starch (RS) content was measured following the gravimetric method of Saura-Calixto, Goñi, Bravo, and Mañas (1993). The PE (0.1 g) was homogenized with 6 mL of 2 mol/L of KOH and placed in a shaker (Maxi Mix II, Thermolyne type 37600 mixer, San Francisco, Calif., U.S.A.) for 30 min at 25 °C under constant agitation. Acetate buffer and 2 mol/L HCl were added and the pH adjusted to 4.75. Subsequently, 60 μL of amyloglucosidase were added and the tube placed in a shaking bath at 60 °C for 30 min. The sample was centrifuged (15 min at 3000×*g*) after the incubation. The pellet was re-suspended in 10 mL distilled water and centrifuged twice, freeze dried and weighed. The fraction obtained corresponded to the RS.

2.4. Free phenolic compounds (PCs)

2.4.1. Methanolic extraction of PCs

The PCs were extracted according to Cardador-Martinez, Loarca-Piña, and Oomah (2002) procedure.

2.4.2. Total PCs

The total PCs content was determined by the Folin-Ciocalteu spectrophotometric method adjusted for 96-well plates (Djeridane *et al.*, 2006). The results were expressed as milligrams of gallic acid equivalents per gram of crackers (mg GAE/g cracker).

2.4.3. Analysis of PCs by HPLC-DAD

A High-performance liquid chromatography-diode array detection (HPLC-DAD) analysis was conducted on an Agilent 1100 Series HPLC system (Agilent Technologies, Palo Alto, CA, USA) using a Zorbax Eclipse XDB-C18 column (Agilent Technologies, 4.6 × 250 mm, 5.0 l m). The column was thermostatically controlled at 35 °C ± 0.6 and the flow rate was set to 1 mL/min. The mobile phase consisted of two solvents: solvent A (0.1% v/v acetic acid) and solvent B (100% acetonitrile). A linear gradient was used as follows: 90–78.5% of solvent A, held for 2 min, 78.5–76% for 6 min, 76–60% for 2 min, 60–50% for 4 min and 50–90% for 2 min. The detection was performed at 280 nm at 1 s velocity. A volume of 20 μL was injected the samples were analyzed in duplicate. Quantification was carried out using the external standard method with commercial standards of (+)-catechin, rutin, quercetin, vanillin and ellagic, caffeine, *p*-coumaric, ferulic, gallic, chlorogenic, and sinapic acids.

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