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Development of a multifunctional yogurt-like product from germinated brown rice



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ARTICLE INFO ABSTRACT The suitability of germinated brown rice (GBR) for developing novel multifunctional yogurt-like products was Keywords: Brown rice evaluated. Crude brown rice, soaked brown rice and GBR for 48 h and 96 h were fermented (F-CBR, F-SBR, F-Oryza sativa GBR48 and F-GBR96, respectively). The viability of the starter culture, acidification pattern, techno-functional Germination properties, content of bioactive compounds [phenolic compounds, y-aminobutyric acid (GABA) and y-oryzanol], Fermentation biological activity [antioxidant and angiotensin I-converting enzyme (ACE) inhibitory activities] and sensory Functional foods attributes were evaluated. Fermentation did not modify proximate composition but improved phenolic and GABA contents as well as ACE-inhibitory activity and consistency index of yogurt-like products. Among them, F-GBR96 exhibited the highest phenolic (15.2 mg GAE/100 g) and GABA (1.9 mg/100 g) concentrations, anti-

oxidant activity (46.9 µg TE/100 g) and ACE-inhibition (61.5%) and was well accepted by panellists.

1. Introduction

The increasing consumer demand for healthy and high-quality foods has led both industry and scientific community to develop new functional foods. Whole grains are gaining popularity due to their high nutritional value and bioactive compounds involved in protective effects against chronic diseases (McRae, 2017), making them valuable ingredients for the development of functional foods.

Rice (*Oryza sativa* L.) is one of the most consumed cereals worldwide, but it is mostly consumed as white rice. Brown rice (BR), containing endosperm, embryo and bran, is nutritionally more complete and provides phytochemicals with health-promoting relevance as γoryzanol, γ-aminoburyric acid (GABA) and ferulic acid located mainly in the germ and bran layers (Gong et al., 2017). However, BR consumption is limited due to its poor textural and sensory properties. BR germination has been demonstrated to be a cost-effective strategy to improve textural and organoleptic quality, nutrient and phytochemical bioavailability and biological activity of this cereal (Cáceres, Martínez-Villaluenga, Amigo, & Frias, 2014). Germinated BR (GBR) is consumed in salads, boiled, or even it is incorporated as an ingredient in bakery products (Chung, Cho, & Lim, 2014; Cornejo, Caceres, Martínez-Villaluenga, Rosell, & Frias, 2015). The development of innovative products is very challenging for food industry to fulfil social demands for natural, gluten-free and vegan products. In this context, ready-to-eat yogurt-like formulations including GBR may expand the range of nondairy fermented products in the market, which is quite limited in western countries so far. Fermentation with lactic acid bacteria (LAB) may enhance GBR health-promoting properties since LAB are cell factories producing nutrients and bioactive compounds that improve functional features of cereals (Waters, Mauch, Coffey, Arendt, & Zannini, 2015). Diverse types of cereals are substrates of lactic-acid fermented beverages, such as oat, maize, rice, barley and sorghum, or a mixture of them (Freire, Ramos, & Schwan, 2017; Salmerón, Thomas, & Pandiella, 2015). Germinated whole grains have been demonstrated to be better substrates for LAB growth compared with non-malted cereals (Nsogning Dongmo, Procopio, Sacher, & Becker, 2016). Therefore, the aim of the present study was to explore the suitability of GBR for the production of a healthy and nutritious fermented vogurt-like product. topic that has not been explored so far. Proximate composition, content of bioactive compounds (phenolic compounds, GABA, and γ -oryzanol), biological activities (antioxidant and ACE-inhibitory activities) and sensory quality were evaluated on the GBR products. Non-fermented formulations were also prepared to evaluate the effect of fermentation on nutritional and health-promoting properties of GBR yogurt-like

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products.

2. Materials and methods

2.1. Chemicals

Unless otherwise stated, all reagents were obtained from Sigma-Aldrich (Barcelona, Spain). MRS agar was obtained from Pronadisa (Madrid, Spain).

2.2. Rice

Commercial BR (*Oryza sativa* subssp. Indica, var. SLF09) was provided by Productora Nacional de Alimentos C.A., INDIA–PRONACA (Ecuador).

2.3. Starter culture

A commercial thermophilic starter culture FD-DVS YC-180 Yo-Flex^{*} containing diverse LAB strains was purchased from Chr. Hansen (Guayaquil, Ecuador). The starter culture was grown in de Man Rogosa Sharpe (MRS) broth at 37 °C for 18 h, and then, bacterial cells were harvested by centrifugation ($8000 \times g$, 10 min), washed twice, and suspended in sterile water, before inoculation in GBR substrates.

2.4. BR germination and flour preparation

BR was soaked in 0.1% sodium hypochlorite for 30 min and then grains were rinsed with distilled water until reaching neutral pH. Then, BR was soaked (SBR) in distilled water for 24 h at 28 °C and germinated for 48 h (GBR48) and 96 h (GBR96) as previously described (Cáceres et al., 2014). These samples were dried at 50 \pm 1 °C for 24 h in a temperature-controlled cabin (Memmert SM200) and milled in a cyclone mill (UDY Corporation, USA). Flours from crude BR (CBR) were also obtained and used as reference. Three germination batches were prepared for each experimental condition.

2.5. Manufacture of fermented products

Yogurt-like products were manufactured from CBR, SBR, GBR48 and GBR96. Briefly, rice flour samples were supplemented with 7% sucrose (w/v), 5% glucose (w/v) and 0.5% stabilizer gelatin (w/v) and mixed in distilled water at 1:4 ratio (w:v) for 1 h at 20 °C, cooked at 95 °C for 30 min, and vacuum-filtered. The resultant slurry fractions were cooled to 42 °C, placed in sterile Erlenmeyer flasks, and then, the starter culture was inoculated into the slurry fractions to get an initial cell count of 2×10^7 CFU/g. Lactic-acid fermentations were performed at 42 °C in agitation until reaching pH 4.4 \pm 0.2 and quickly cooled at 4 °C. Fermentation experiments were performed in duplicate for each germination condition. Fermented products were coded as follows: F-CBR for that prepared with crude BR, F-SBR for that prepared with soaked BR, and F-GBR48 and F-GBR96 for those obtained from BR germinated for 48 and 96 h, respectively. Non-fermented products were also produced from crude (NF-CBR), soaked (NF-SBR) and germinated BR (NF-GBR48 and NF-GBR96).

2.6. Bacterial growth and pH determination

Bacterial growth was determined in fermented products by plating decimal buffered peptone water dilutions $(10^{6}-10^{8})$ in triplicate onto MRS agar and counting the viable cells after incubation under anaerobic conditions (37 °C for 48 h). The pH was monitored using a pH-meter Basic 20 (Crison Instruments S. A., Barcelona, Spain).

2.7. Proximate composition

Chemical composition of non-fermented and fermented products was determined following AOAC (2016) methods for moisture, protein, fat and ash. Carbohydrates were calculated by difference. Food energy was calculated using standardized conversion factors (4.0 kcal/g for proteins and carbohydrates, and 9.0 kcal/g for fats) (FAO, 2002).

2.8. Techno functional properties

2.8.1. Consistency and flow behaviour index

The consistency index in Pa.sⁿ (*K*) and the flow behaviour index (n) were determined using a rotational viscometer Brookfield DV-II + with spindle for non-Newtonian fluids through the logarithmic linearization of the apparent viscosity (η) curve in Pa.s vs shear rate in s⁻¹ (γ) (Steffe, 1996).

2.8.2. Total soluble solids and density

Total soluble solids and density were determined by refractometry and gravimetric methods, respectively (AOAC, 2016).

2.9. Determination of total phenolic compounds

Total phenolic content (TPC) was determined using Folin-Ciocalteu's reagent (Cáceres et al., 2014). Results were expressed as mg of gallic acid equivalents (GAE)/100 g product.

2.10. Determination of GABA content

GABA was extracted with methanol and further analyzed by RP-HPLC as in Caceres at al. (2014). Results were expressed as mg GABA/100 g product.

2.11. Determination of γ -oryzanol content

The analysis of γ -oryzanol in BR products was performed by RP-HPLC according to Cáceres, Peñas, Martinez-Villaluenga, Amigo, and Frias (2017). Results were expressed in mg γ -oryzanol/100 g product.

2.12. Determination of oxygen radical absorbance capacity (ORAC)

Antioxidant activity was determined as ORAC by fluorescence as described recently in Caceres et al. (2014). Results were expressed as mg of Trolox equivalents (TE)/100 g product.

2.13. Determination of ACE-inhibitory activity

It was determined in all BR formulations as in Cáceres et al. (2017). Results were presented as % inhibition.

2.14. Sensory evaluation

BR yogurt-like products were submitted to sensory analysis by 8 trained panellists (50% male, 50% female) recruited at Facultad de Ingeniería Mecánica y Ciencias de la Producción (ESPOL, Guayaquil, Ecuador), selected by their regular consumption of yogurt-like products. Panellists evaluated typical attributes for fermented products such as astringency, bitterness, sourness, cereal-type flavour, fermented odour, creaminess, white colour and overall acceptability, using a 9-point hedonic scale, ranging from dislike extremely (1) to like extremely (9). Each panelist received 2 samples of each type of experimental fermented products. F-CBR was considered the reference product, and received a score of 5 for all descriptors. Panellists evaluated the other formulations in comparison with F-CBR.

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