Physiochemical and nutritional characteristics, bioaccessibility, and sensory acceptance of baked crackers containing broccoli co-products

Tomás Lafarga [a], Ariadna Bademunt [b], Eimear Gallagher [c], Gloria Bobo [a], Gemma Echeverria [a], Inmaculada Viñas [b], and Ingrid Aguiló-Aguayo [a]*

[a] IRTA, XaRTA-Postharvest, Parc Científic i Tecnològic Agroalimentari de Lleida, Parc de Gardeny, Edifici Fruitcentre, 25003, Lleida, Catalonia, Spain.

[b] Food Technology Department, University of Lleida, XaRTA-Postharvest, Agrotecnio Center, Lleida, Spain

[c] Teagasc Food Research Centre Ashtown, Department of Food Chemistry and Technology, Dublin 15, Ireland.

*__Corresponding author__:
Dr Aguiló-Aguayo, phone: (+34) 973003431 Ext. 1551, email: Ingrid.Aguilo@irta.cat

Running title: Broccoli co-products as novel ingredients in baked crackers

Abbreviations

a_w: Water activity; DW: Dry weight; SDF: Soluble dietary fibre; IDF: Insoluble dietary fibre, TDF: Total dietary fibre, TGC: Total glucosinolate content, DPPH: 2,2-diphenyl-1-picrylhydrazyl; C^*_{ab}: Chroma; δE: Difference from the control; S.D.: Standard deviation; ANOVA: Analysis of variance.

Abstract

The effects of the inclusion of broccoli co-products into crackers on the bioaccessibility as well as their overall physical and nutritional quality were evaluated. Crackers were formulated using a 12.5 or 15.0% flour substitution level. Broccoli-containing crackers presented higher specific volume and spread ratio and lower weight and specific volume than control crackers (p<0.05). Crackers containing broccoli co-products showed an increased green hue and a higher colour intensity (p<0.05). Incorporation of broccoli co-products into crackers significantly increased the total phenolic content and antioxidant capacity (p<0.05). A simulated gastrointestinal digestion suggested that the amount of phenolic and antioxidant compounds released during digestion might be higher than what could be expected from common water-organic extracts. The incorporation of broccoli co-products into baked crackers would not only reduce the amount of food discarded as waste but also promote health and open novel commercial opportunities to food processors.

Keywords: functional foods, antioxidant activity, baked goods, broccoli co-products, bioaccessibility, phenolic compounds

Introduction

Unfortunately, the generation of waste in the food processing industry is unavoidable. A large amount of the food and food co-products currently discarded as waste or used for low value purposes are rich in valuable compounds which could be reincorporated into the food chain. The utilization of edible co-products rich in health-promoting compounds as novel ingredients for the development of functional foods would not only reduce the amount of food discarded as waste, or used for low value purposes, but also promote health and open novel commercial opportunities for food processors. Functional foods deliver additional or enhanced benefits over their basic nutritional value and the functional foods market is currently one of the top trends in the food industry. In addition, the global market for snack foods is projected to exceed US$630 billion by 2020, driven by robust demand for functional snacks and the rising popularity of organic and natural ingredients-based snacks (INC. 2015). Crackers represent an important share of the snack market and provide a large number of opportunities for new product development, especially in the area of functional foods (Millar *et al,*. 2017).

Broccoli (*Brassica oleracea italica*) contains health-promoting substances including phenolic compounds and glucosinolates. Large amounts of co-products including leaves, stems, and stalks are generated during processing of broccoli. Recent studies suggested that broccoli stems and leaves contain high levels of total phenolics and high antioxidant and anticarcinogenic activities. Moreover, Lafarga *et al,*. (2018b) reported comparable nutritional profiles and a similar resistance to thermal processing between the florets and stems of several *Brassica* vegetables including varied broccoli varieties. Although health-promoting compounds found in cruciferous vegetables can be heavily lost during thermal processing (Lafarga *et al,*. 2018a; Sarvan *et al,*. 2012), previous studies suggested that heat-sensible ingredients could be resistant to thermal processing when incorporated into baked products (Lafarga *et al*,. 2016).

The aim of this work was to produce functional crackers with enhanced concentrations of fibre, glucosinolates, and phenolic compounds using broccoli co-products and to study the influence of their inclusion on the physicochemical parameters of the product including weight, density, colour, firmness, moisture content, and fibre content as well as on the overall quality and acceptance of the end product. The effects of broccoli inclusion into crackers on parameters including colour, texture, moisture, water activity (a_w), antioxidant capacity, acceptance, and total glucosinolate (TGC) were studied over a 14-day period.

Materials and methods

Preparation of crackers

Broccoli stems were cut into 10 × 10 × 10 mm cubes, sanitized in 100 ppm sodium hypochlorite for 2 min, rinsed with tap water, and left to dry at room temperature to reduce surface contamination. The co-products were frozen, freeze-dried, milled to a thin powder, vacuum sealed, and stored at -20 ºC until further use. The doughs were prepared for mixing according to the formulations listed in Table S1 and following the methodology described in Supplementary File S1. Control crackers without powdered freeze-dried broccoli were labelled as F00.0. Crackers containing broccoli at a flour substitution level of 12.5 and 15.0% (w/w) were labelled as F12.5 and F15.0, respectively.

Dimensions, weight, and chemical composition of crackers

The weight and dimensions of 30 crackers were averaged for each formulation and replicate. Length, width, and thickness were measured with Verner calipers and the spread ratio, specific volume, and density were calculated for each sample. Weight, length, width, and thickness measurements were taken at day 1 post-baking.

Moisture content was determined using AACC method 44-15.02. Soluble (SDF), insoluble (IDF), and total (TDF) dietary fibre were determined according to AOAC Method 991.43, using the ANKOM dietary fibre analyser (ANKOM technology, NY, USA) and expressed as percentage.

Colour and texture

Colour recordings were taken using a Minolta CR-200 colorimeter (Minolta INC, Tokyo, Japan). CIE values were recorded in terms of $L*$ (lightness), $a*$ (redness/greenness), and $b*$ (yellowness/blueness). Calibration was carried out using a standard white tile (Y:92.5, x:0.3161, y:0.3321) provided by the manufacturer and the D65 illuminant, which approximates to daylight. Chroma (C^*_{ab}) and difference from the control (δE) were calculated following the methodology described by Wibowo *et al,.* (2015). Results are the average of 10 measurements per formulation and replicate taken on day 1 post-baking. Texture characteristics were assessed using a TA.XT2 Texture Analyzer (Stable Micro Systems Ltd., Surrey, England) connected to Exponent software v. 5.0.6.0. Hardness was determined using the hardness measurement by cutting test provided by the manufacturer and a knife edge with slotted insert probe (HDP/BS). Ten samples were taken for each formulation and replicate and measurements were carried out on day 1 post-baking.

Water activity and pH

The a_w of all samples was measured using an AquaLab meter (Decagon Devices Inc., WA, USA) and approximately 2 g of ground sample. Three measurements were taken for each formulation and replicate on days 1, 7, and 14 post-baking.

The pH of 1g of ground crackers added to 10 g of distilled water was measured in a Basic 20 pH meter (Crison Instruments S.A., Barcelona, Spain) as previously described by O'Shea *et al,.* (2017). pH measurements were carried out in triplicate for each formulation and replicate at day 1 post-baking.

Total phenolic content

The TPC was determined by the Folin Ciocalteu method following the modifications described by Altisent, Plaza, Alegre, Viñas, and Abadias (2014). Briefly, samples were homogenized with 70% (w/w) methanol at a sample to solvent ratio or 3:10 (w/v) at 4 ºC for 1 min using a T-25 digital ULTRA-TURRAX® homogenizer (IKA, Staufen, Germany) at 14,000 rpm. Extraction was held at 4 ºC and constant shaking in an ice bath for 20 min. Absorbance was measured at 760 nm using a GENESYS™ 10S-UV Vis spectrophotometer (Thermo Fisher Scientific, MA, USA). The TPC was determined in triplicate for each formulation and replicate on days 1, 7, and 14 post-baking. Results were expressed on a dry weight (DW) basis as mg of gallic acid equivalents per 100 g.

Antioxidant activity

Antioxidant activity was measured using two different methods: the FRAP and the 2,2-diphenyl-l-picrylhydrazyl radical (DPPH·) scavenging activity assays following the methodologies previously described by Altisent, Plaza, Alegre, Viñas, and Abadias (2014) and using the same extract used for TPC determination. Absorbance was measured at 593 and 515 nm for FRAP and DPPH· assays respectively. Antioxidant activity was determined in triplicate for each formulation and replicate on days 1, 7, and 14 post-baking. Results were expressed as g of ascorbic acid equivalents per 100 g of DW.

Total glucosinolate content

The TGC of the broccoli stems and of the control and broccoli-containing crackers were determined following the methodologies described by Mawlong *et al*,. (2017). Briefly, spectrophotometric estimation was done using methanolic extract prepared from by homogenizing 0.1 g defatted sample with 80% (v/v) methanol. The homogenized sample was centrifuged at 3,000 rpm for 4 min after keeping overnight at room temperature. The supernatant was collected and made up to 2 ml with 80% (v/v) methanol. Absorbance was measured at 425 nm using a GENESYS^{TM} 10S-UV Vis spectrophotometer (Thermo Fisher Scientific, MA, USA). TGC was determined in triplicate for each broccoli-containing formulation and replicate on days 1, 7, and 14 post-baking. TGC was expressed as mg of glucoraphanin equivalents per 100 g DW.

Sensory evaluation

Sensory evaluation was undertaken at day 1 post-baking with 40 untrained panellists recruited from IRTA Fruitcentre. Sensory evaluation was conducted in a sensory laboratory with separate booths following the methodology described by Millar *et al*,. (2017) with some modifications. Briefly, samples were placed on white polystyrene plates labelled with random codes and presented to consumers in a randomised order. A 60-s time laps was employed between each sensory palate, to reduce sensory fatigue. Each panellist assessed all three samples and was asked to indicate his or her opinion on the firmness, crunchiness, overall visual appearance, and overall acceptability of the products using a 9-point hedonic scale (from 1: dislike extremely to 9: like extremely).

***In vitro* gastrointestinal digestion**

A simulated gastrointestinal digestion of the control and broccoli-containing crackers was performed following the methodology previously described by Zudaire *et al*,. (2017). The methodology consists of three sequential stages including an oral (α-amylase, pH 7.0), gastric (pepsin, pH 3.0), and intestinal (pancreatin and fresh bile, pH 7.0) phase. Determinations of TPC and antioxidant capacity were carried out after both gastric and intestinal phases by substituting the methanolic extract for the same amount of the digestive enzymatic extracts.

Statistical analysis

Results are expressed as mean ± standard deviation (S.D.). Differences between samples were analyzed using analysis of variance (ANOVA) with JMP 13 (SAS Institute Inc., NC, USA). Where significant differences were present, a Tukey pairwise comparison of the means was conducted to identify where the sample differences occurred (p<0.05).

Results and discussion

In the current study broccoli stems showed an antioxidant potential of 317 ± 29 and 239 ± 14 mg/100 g DW calculated using the FRAP and DPPH· methods, respectively. The TPC of the broccoli co-products used in the current study was calculated as 312 ± 41 mg/100 g DW. Results compared well in terms of TPC and antioxidant bioactivity to those recently obtained by Lafarga *et al*,. (2018b) who assessed the TPC and antioxidant capacity of the stems of different broccoli varieties.

Physical quality

The inclusion of broccoli co-products into the cracker formulations significantly affected the colour parameters of the baked crackers listed in Table 1. The L^* parameter which denotes lightness and varies from 0 (black) to 100 (white) was significantly lower in both broccoli-containing crackers, F12.5% and F15.0%, when compared to the control (p<0.05). This denotes a lighter appearance of the control samples. A negative correlation was observed between L^* values and broccoli content (r^2=-0.837). Similar results were obtained previously after inclusion of coloured ingredients into baked products such as seaweed (Fitzgerald *et al*,. 2014) or blackcurrant pomace (Schmidt *et al*,. 2018). As expected, inclusion of broccoli co-products into crackers increased the green hue of the final product (data not shown) and the C^*_{ab} value, a quantitative indicator of colourfulness (p<0.05). This indicates that the broccoli-containing crackers had a higher colour intensity. A positive correlation was observed between L^* values and broccoli content (r^2=0.947). Similar increases in C^*_{ab} were observed in baked crackers after substitution of flour with pulses (Millar *et al*,. 2017). The δE combines the change in L^*, a^*, and b^* values to quantify the colour deviation from a standard reference sample, in this case, wheat flour crackers. Those samples with δE > 3 display a visible colour deviation

(Wibowo *et al*,. 2015). As expected, broccoli-containing crackers had a $\delta E > 3$, exhibiting a visible colour deviation when compared to the control.

Broccoli-containing crackers showed a lower pH at day 1 post-baking when compared to the control crackers ($p<0.05$). Overall, physical characteristics of the crackers were significantly affected by the inclusion of broccoli at 12.5 and 15.0% (w/w; $p<0.05$). Both broccoli-containing formulations presented higher specific volume and spread ration and lower weight and specific volume than control crackers ($p<0.05$). A positive correlation was observed between the spread ratio and broccoli content ($r^2=0.966$). Higher spread ratios are considered more desirable at industrial level (Tiwari *et al*,. 2011). In addition, the moisture content of the crackers was comparable to that of previous formulations and was higher when compared to other snacks such as legume-containing crackers (Colla and Gamlath 2015). Both broccoli-containing crackers had a lower moisture content than the control ($p<0.05$) and a negative correlation was observed between the broccoli content and water content ($r^2=-0.975$). The moisture content of the three cracker formulations was significantly lower at day 7 when compared to day 1 ($p<0.05$) and no differences were observed between the moisture content at day 7 and day 14 suggesting a stable product.

Inclusion of broccoli at a concentration of 15.0% (w/w) resulted in increased hardness ($p<0.05$). Although an increase in hardness was also appreciated for samples containing broccoli at a concentration of 12.5% (w/w), results were not significantly different. Previous studies suggested a direct relationship between hardness of crackers and total fibre content and a negatively correlation between hardness and moisture content (Millar *et al*,. 2017). Therefore, the observed increase in hardness observed after broccoli inclusion in the current study could be caused by a reduced moisture and an increased fibre content when compared to control samples.

Nutritional characteristics of baked crackers

Previous studies obtained increased TDF content in crackers and other baked goods after substitution of wheat flour with other vegetable-derived sources (Millar *et al*,. 2017). In the current study, the TDF and IDF of broccoli-containing crackers was higher when compared to the control. However, the observed increase in fibre content was not statistically significant.

The TPC of the baked crackers was higher in both broccoli-containing crackers when compared to the control ($p<0.05$; Figure 1). Similar results were observed previously after inclusion of broccoli into bread at a concentration of 2% (w/w) (Gawlik-Dziki *et al*,. 2014). TPC at day 1 was positively correlated with broccoli content (r^2 = 0.919). Moreover, the TPC of all cracker formulations was significantly lower at day 7 when compared to day 1 ($p<0.05$). Previous studies observed a decrease in the TPC of foods during storage (Patras *et al*,. 2011, Howard *et al*,. 2010). In the current study, no differences were observed between the TPC measured at days 7 and 14 for all cracker formulations suggesting stable products.

Due to the high TPC measured in both broccoli-containing formulations, an increased antioxidant activity was expected. The expected increase in antioxidant activity was achieved in both broccoli-containing crackers ($p<0.05$). Gawlik-Dziki *et al*,. (2014) obtained increased antioxidant capacity after inclusion of broccoli sprouts into wheat bread formulations at concentrations ranging from 1 to 5% (w/w). Similar results were obtained by Lee (2015). In the current study, a positive correlation was observed between the antioxidant potential at day 1 and broccoli content when assessed using the DPPH (r^2 = 0.760) and FRAP (r^2 = 0.911) methods. Other food products such as soups have shown increased antioxidant capacity after incorporation of broccoli co-products into their recipe (Alvarez-Jubete *et al*,. 2014). Antioxidant activity assessed using the DPPH· method was lower at day 7 and 14 when compared to day 1. However, the observed decrease was only statistically significant for cF12.5 (p<0.05). When assessed using the FRAP method, the antioxidant capacity of F00.0 and F12.5 was significantly lower at days 7 and 14 when compared to day 1 (p<0.05).A positive correlation was observed between the antioxidant capacity as assessed using the FRAP method and the TPC at day 1 (r^2 = 0.985). Similar results were observed after inclusion of vegetable co-products such as mango peel into baked goods (Ajila *et al*,. 2010) .The TGC of F12.5 and F15.0 is shown in Figure 1. No differences were observed between the TGC of F12.5 and F15.0. In addition, the TGC of both F12.5 and F15.0 was affected by storage and decreased at days 7 and 14 (p<0.05). A decrease in the TGC of formulation F12.5 was observed from day 7 to day 14 but it was not statistically significant.

Sensory analysis

Scores obtained after sensory analysis for overall appearance, overall acceptability, firmness, and crunchiness are shown in Figure S1. Inclusion of broccoli into the crackers formulation resulted in increased overall acceptability (p<0.05). Previous studies which assessed the effect of the incorporation of fruit and vegetable co-products into baked goods obtained acceptability scores comparable to the control (Schmidt *et al*,. 2018, Chareonthaikij *et al*,. 2016). Results are comparable to previous studies which obtained high acceptability scores after inclusion of broccoli powder into bread (Gawlik-Dziki *et al*,. 2014). The overall visual appearance score obtained for F12.5 was significantly high when compared to F00.0 and F15.0 (p<0.05). Although a decrease in crunchiness and firmness was perceived after inclusion of broccoli, results were not statistically different.

Simulated gastrointestinal digestion

Bioaccessibility, which has is defined as the release of compounds from their natural food matrix to be available for intestinal absorption, is one of the main limiting factors for bioavailability (Stahl *et al*., 2002). The *in vitro* gastrointestinal digestion strategy suggests which compounds survive the gastrointestinal tract conditions and are likely to reach the colon where they can act or be absorbed into the blood stream (McDougall *et al*,. 2007). Overall, the results shown in Table 2 were comparable in magnitude for each cracker formulation. Both the TPC and antioxidant potential increased after the gastric stage, when compared to the initial stage, for all cracker formulations (p<0.05). An increase in the TPC was also observed for all cracker formulations after the intestinal stage (p<0.05). Strong pH variations and pepsin may affect the integrity of cell walls, facilitating the liberation of phenolic and antioxidant compounds not detected in initial phases, and hydrolysed wheat- and broccoli-derived proteins resulting in peptides with antioxidant properties (Niu *et al*,. 2013, Cian *et al*,. 2015). The longer extraction process, if compared to values prior to digestion (those obtained after the methanol:water extraction), may partially explain these findings. Results obtained for TPC and antioxidant activity were not comparable as the antioxidant capacity assessed using both the FRAP and DPPH· assays was significantly lower after the intestinal phase when compared to the digestive stage (p<0.05). Different pH values can affect racemization of molecules creating two different chiral enantiomer. This could alter their biological activities and may render antioxidant more reactive early in the digestive process, particularly at acidic pH values (Jamali *et al*,. 2008). Similar results were published byGawlik-Dziki *et al*,. (2009) observed a significant increase in the TPC of the breads containing either 2.5 or 5.0% (w/w) buckwheat flavones during the different stages of digestion. The TPC of these breads varied from 0.50 and 0.72 mg/mL at the initial stage to 0.78 and 0.79 mg/mL, and 0.72 and 0.90 mg/mL after the gastric and intestinal phases, respectively. Pérez-Jiménez and Saura-Calixto (2005) also reported that TPC and antioxidant activity of the digestive enzymatic extracts was significantly higher when compared to that of the water-organic extracts.

Conclusions

Broccoli co-products could be incorporated into baked products such as crackers at relatively high concentrations in order to increase their nutritional and physicochemical quality without affecting their overall acceptance. Incorporation of broccoli into cracker formulations increased the content of dietary fibre, total phenolics, and glucosinolates as well as the antioxidant capacity when compared to the flour-only crackers. Overall, a decrease in the TPC and the antioxidant activity of all cracker formulations was observed during storage, especially during the first week. Results obtained herein suggest that the amount of phenolic and antioxidant compounds released during digestion may be higher than what could be expected from common water-organic extracts.

Acknowledgements

This work was supported by the CERCA Programme of *Generalitat de Catalunya*. T. Lafarga is in receipt of *Juan de la Cierva* contract awarded by the Spanish Ministry of Economy and Competitiveness (FJCI-2016-29541). I. Aguiló-Aguayo thanks the Spanish Ministry of Economy and Competitiveness and to the European Social Fund for the Postdoctoral Senior Grant *Ramon y Cajal* (RYC-2016-19949). Authors thank Congelados de Navarra S.A.U (Navarra, Spain) for providing broccoli processing co-products and Silvia Villaró for her technical assistance.

Conflict Statements

The authors declare no conflict of interests.

References

Ajila, C. M., Aalami, M., Leelavathi, K. & Rao, U. J. S. P. (2010). Mango peel powder: A potential source of antioxidant and dietary fiber in macaroni preparations. *Innovative Food Science & Emerging Technologies,* **11,** 219-224.

Chareonthaikij, P., Uan-On, T. & Prinyawiwatkul, W. (2016). Effects of pineapple pomace fibre on physicochemical properties of composite flour and dough, and consumer acceptance of fibre-enriched wheat bread. *International Journal of Food Science & Technology,* **51,** 1120-1129.

Cian, R. E., Vioque, J. & Drago, S. R. (2015). Structure–mechanism relationship of antioxidant and ACE I inhibitory peptides from wheat gluten hydrolysate fractionated by pH. *Food Research International,* **69,** 216-223.

Colla, K. & Gamlath, S. (2015). Inulin and maltodextrin can replace fat in baked savoury legume snacks. *International Journal of Food Science & Technology,* **50,** 2297-2305.

Fitzgerald, C., Gallagher, E., Doran, L., Auty, M., Prieto, J. & Hayes, M. (2014). Increasing the health benefits of bread: Assessment of the physical and sensory qualities of bread formulated using a renin inhibitory Palmaria palmata protein hydrolysate. *LWT - Food Science and Technology,* **56,** 398-405.

Gawlik-Dziki, U., Dziki, D., Baraniak, B. & Lin, R. (2009). The effect of simulated digestion in vitro on bioactivity of wheat bread with Tartary buckwheat flavones addition. *LWT-Food Science and Technology,* **42,** 137-143.

Gawlik-Dziki, U., Świeca, M., Dziki, D., Sęczyk, Ł., Złotek, U., Różyło, R., Kaszuba, K., Ryszawy, D. & Czyż, J. (2014). Anticancer and antioxidant activity of bread enriched with broccoli sprouts. *BioMed research international,* **2014**.

Howard, L. R., Castrodale, C., Brownmiller, C. & Mauromoustakos, A. (2010). Jam processing and storage effects on blueberry polyphenolics and antioxidant capacity. *Journal of agricultural and food chemistry,* **58,** 4022-4029.

INC., G. I. A. (2015). Snack Foods: A Global Strategic Business Report. Pp. 1-1073.

Jamali, B., Bjørnsdottir, I., Nordfang, O. & Hansen, S. H. (2008). Investigation of racemisation of the enantiomers of glitazone drug compounds at different pH using chiral HPLC and chiral CE. *Journal of pharmaceutical and biomedical analysis,* **46,** 82-87.

Lafarga, T., Bobo, G., Viñas, I., Collazo, C. & Aguiló-Aguayo, I. (2018a). Effects of thermal and non-thermal processing of cruciferous vegetables on glucosinolates and its derived forms. *Journal of Food Science and Technology*.

Lafarga, T., Gallagher, E., Aluko, R. E., Auty, M. A. & Hayes, M. (2016). Addition of an enzymatic hydrolysate of bovine globulins to bread and determination of hypotensive effects in spontaneously hypertensive rats. *Journal of agricultural and food chemistry,* **64,** 1741-1750.

Lafarga, T., Viñas, I., Bobo, G., Simó, J. & Aguiló-Aguayo, I. (2018b). Effect of steaming and sous vide processing on the total phenolic content, vitamin C and antioxidant potential of the genus Brassica. *Innovative Food Science & Emerging Technologies,* **47,** 412-420.

Lee, S.-H. (2015). Quality and antioxidant properties of white breads enhanced with broccoli (Brassica oleracea L.) powder. *Korean journal of food and cookery science,* **31,** 614-622.

Mawlong, I., Sujith Kumar, M., Gurung, B., Singh, K. & Singh, D. (2017). A simple spectrophotometric method for estimating total glucosinolates in mustard de-oiled cake. *International Journal of Food Properties***,** 1-8.

McDougall, G. J., Fyffe, S., Dobson, P. & Stewart, D. (2007). Anthocyanins from red cabbage – stability to simulated gastrointestinal digestion. *Phytochemistry,* **68,** 1285-1294.

Millar, K. A., Barry-Ryan, C., Burke, R., Hussey, K., McCarthy, S. & Gallagher, E. (2017). Effect of pulse flours on the physiochemical characteristics and sensory acceptance of baked crackers. *International Journal of Food Science & Technology,* **52,** 1155-1163.

Niu, L.-Y., Jiang, S.-T. & Pan, L.-J. (2013). Preparation and evaluation of antioxidant activities of peptides obtained from defatted wheat germ by fermentation. *Journal of Food Science and Technology,* **50,** 53-61.

O'Shea, N., Kilcawley, K. N. & Gallagher, E. (2017). Aromatic Composition and Physicochemical Characteristics of Crackers Containing Barley Fractions. *Cereal Chemistry,* **94,** 611-618.

Patras, A., Brunton, N. P., Tiwari, B. & Butler, F. (2011). Stability and degradation kinetics of bioactive compounds and colour in strawberry jam during storage. *Food and bioprocess technology,* **4,** 1245-1252.

Pérez-Jiménez, J. & Saura-Calixto, F. (2005). Literature data may underestimate the actual antioxidant capacity of cereals. *Journal of agricultural and food chemistry,* **53,** 5036-5040.

Sarvan, I., Verkerk R., & Dekker, M. (2012). Modelling the fate of glucosinolates during thermal processing of *Brassica* vegetables. *LWT – Food Science and Technology,* **49,** 178-183.

Schmidt, C., Geweke, I., Struck, S., Zahn, S. & Rohm, H. (2018). Blackcurrant pomace from juice processing as partial flour substitute in savoury crackers: dough characteristics and product properties. *International Journal of Food Science & Technology,* **53,** 237-245.

Stahl, W., van den Berg, H., Arthur, J., Bast, A., Dainty, J., Faulks, R. M., Gärtner, C., Haenen, G., Hollman, P., & Holst, B. (2002). Bioavailability and metabolism. *Molecular aspects of medicine,* **23**, 39-100.

Tiwari, B. K., Brennan, C. S., Jaganmohan, R., Surabi, A. & Alagusundaram, K. (2011). Utilisation of pigeon pea (Cajanus cajan L) byproducts in biscuit manufacture. *LWT - Food Science and Technology,* **44,** 1533-1537.

Wang, T., He, F., & Chen, G. (2014). Improving bioaccessibility and bioavailability of phenolic compounds in cereal grains through processing technologies: A concise review. *Journal of Functional Foods, 7*, 101-111.

Wibowo, S., Vervoort, L., Tomic, J., Santiago, J. S., Lemmens, L., Panozzo, A., Grauwet, T., Hendrickx, M. & Van Loey, A. (2015). Colour and carotenoid changes of pasteurised orange juice during storage. *Food chemistry*, **171,** 330-340.

Zudaire, L., Viñas, I., Abadias, M., Simó, J., Echeverria, G., Plaza, L. & Aguiló-Aguayo, I. (2017). Quality and bioaccessibility of total phenols and antioxidant activity of calçots (Allium cepa L.) stored under controlled atmosphere conditions. *Postharvest Biology and Technology,* **129,** 118-128.

Legends to figure

Figure 1. (A) TPC, antioxidant activity measured using the (B) FRAP and (C) DPPH methods, and (D) TGC

Values represent the mean of three independent experiments ± S.D. Capital letters indicate significant differences between different formulations at the same sampling day. Lower case letters indicate significant differences between sampling days for the same cracker formulation.

Supplementary items

Table S1. Cracker formulations

Supplementary File S1: Baking procedure

Figure S1. Sensory evaluation of broccoli-containing crackers assessed at day 1 post baking