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Use of postharvest UV-B and UV-C radiation treatments to revalorize broccoli byproducts and edible florets

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ABSTRACT

UV-B radiation (5, 10 and 15 kJ m^{-2}) treatments, single or combined with UV-C (9 kJ m^{-2}), were hereby firstly studied as bioprocessing tools to enhance bioactive compounds of broccoli byproducts (leaves and stalks) compared to edible florets during storage (72 h/15 °C). Leaves showed similar total phenolic content (TPC) and antioxidant capacity (TAC) than florets, and 2.5/14.5 higher glucoraphanin/glucobrassicin contents than florets. UV increased initial TPC and TAC of leaves/stalks up to 31–97/30–75 and 20–120/170–420%, respectively. UV-B10 + C induced the highest TPC increase (110%) in leaves while UV-B10 and UV-B10 + C led to the highest TPC of stalks after 48 h. UV-B10 + C increased (34%) glucobrassicin levels of leaves while UV-B15 and UV-B15 + C induced the highest glucoraphanin levels (131 and 117 mg kg^{-1}) in florets after 72 h. Conclusively, single or combined postharvest UV-B and UV-C treatments can revalorize such agricultural byproducts and also add value to edible broccoli parts.

Industrial relevance: This work demonstrates the high potential of the use of postharvest treatments with UV-B and UV-C, optimized as single or combined treatments, to be used as bioprocessing tools to enhance the bioactive compounds of broccoli byproducts (leaves and stalks) being compared to edible florets. Thought this innovative application of this technology such agricultural wastes may be highly revalorized with a cheap technology leading to high production of health-promoting compounds with such eco-sustainable technology whose benefits can be even reach to the pharmaceutical industry.

1. Introduction

Approximately 1.3 billion tons of food is worldwide wasted or lost per year being 10–20% attributed to preharvest losses as recently published (FAO, 2015). Use of plant byproducts supports the low carbon economy using renewable resources, offering environmental and economic benefits and improve efficiency in food industry (O'Shea, Arendt, & Gallagher, 2012; Sanz-Puig et al., 2016; Tarazona-Diaz & Aguayo, 2013). Broccoli leaves and stalks represent a high proportion of the total above-ground plant biomass which leads to high amount of waste with the correspondent negative environment impact. Therefore, broccoli byproducts revalorization may be an important source of nutritional/bioactive compounds for the food and pharmaceutical industries reducing, at the same time, the agricultural wastes on the fields.

Bimi broccoli is a new natural hybrid between Chinese broccoli (*Brassica oleracea*, Alboglabra group), also called kailan or gailan, and conventional broccoli (*B. oleracea*, Italica group). This broccoli is characterized by a small floret with a long (15–18 cm) slender stem which has a mild sweeter taste compared to conventional broccoli varieties, being completely edible (raw or cooked) (Martínez-Hernández, Gómez, Pradas, Artés, & Artés-Hernández, 2011). The leaves and stalks of Bimi broccoli represent a 75.5% (in dry weight basis) of total above-ground plant biomass. This constitutes a high amount of waste, with a negative effect on the agricultural environment in the Región de Murcia (Southeast of Spain) that is the main European Bimi broccoli producer with 150 ha cultivated in the last campaign (data supplied by Sakata Seeds Ibérica S.L.U.). Bimi broccoli is rich in phenolic compounds, glucosinolates, vitamin C and other antioxidant compounds (Martínez-Hernández et al., 2013). Glucoraphanin and glucobrassicin are the main glucosinolates present in broccoli being their cognate isothiocyanates, sulforaphane and indol-3-carbinol, extensively studied for their potent induction of mammalian detoxication (phase 2) enzyme activity and anti-cancer agent (Traka & Mithen,

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2008). The non-edible parts of Bimi plant (stalks and leaves) may have high contents of bioactive compounds like similarly found in conventional broccoli varieties (Aguiló-Aguayo et al., 2014; Dosz, Ku, Juvik, & Jeffery, 2014). Accordingly, the use of Bimi plant byproducts wasted during preharvest stage appears as an interesting source of health-promoting compounds for the food and pharmaceutical industries.

Plant products have been proposed as biofactories of bioactive compounds through different induced abiotic stresses-mechanisms such as UV radiation, wounding, etc. (Cisneros-Zevallos, 2003; Formica-Oliveira, Martínez-Hernández, Díaz-López, Artés, & Artés-Hernández, 2017). The effects of UV-B radiation prior to harvest growing crops has been widely studied being also characterized the role of this radiation on plant physiology and fitness (Schreiner, Krumbein, Mewis, Ulrichs, & Huyskens-Keil, 2009). Nevertheless, this may highly differ from the metabolic responses occurring in harvested organs (Darré et al., 2017; Müller, Noack, Greiner, Stahl, & Posten, 2014). The potential use of UV-B as a postharvest treatment to enhance the health-promoting compounds of fruit and vegetables has been previously reported just in a few studies (Alegria, 2015; Darré et al., 2017; Du, Avena-Bustillos, Breksa, & McHugh, 2012; Formica-Oliveira et al., 2017; Scattino et al., 2014). UV-C radiation has a high germicidal effect which may reduce microbial growth during storage of fresh-cut products (Santhirasegaram, Razali, George, & Somasundram, 2015).

However, application of UV-B, single or combined with UV-C, has not been still studied as a revalorization tool to increase the main health-promoting compounds in broccoli byproducts. Consequently, this work studied the single and combined effects of UV-B and UV-C on total phenolic content (TPC), total antioxidant capacity (TAC) and main glucosinolates (glucoraphanin and glucobrassicin) of Bimi broccoli leaves and stalks, being compared to the edible part (floret), during 72 h at 15 °C.

2. Materials and methods

2.1. Plant material

Bimi broccoli plants were grown under open air cultivation in fields located in the Region of Murcia, in the Southeast Mediterranean Spanish area, in the spring growing cycle (planting in March) and were harvested randomly at the end of May (average temperature intervals of 4–27 °C). Plant material was grown according to integrated pest management cultural practices. Broccoli plants were hand-harvested at commercial ripening stage of florets (head diameters of 3–5 cm and stem lengths of 15–18). Immediately after harvesting, broccoli plants were pre-cooled with crushed ice and transported by car about 80 km to the Pilot Plant of our Research Group in the Universidad Politécnica de Cartagena, where it was stored at 4 °C and 90–95% relative humidity (RH) until next day.

2.2. Sample preparation

Preparation of plant material was conducted in a disinfected cold room at 8 °C. Leaves were removed from the plant main stalk using a sharp knife. Bimi florets were cut in about 15-cm-long spears. The obtained three broccoli parts (leaves, stalks and florets) were then washed with chlorinated water (150 mg L^{-1} free chlorine; pH 6.5; 5 °C) for 2 min and rinsed with tap water at 5 °C for 1 min. Once drained, plant material was carefully dried with towel paper and disposed in plastic trays until UV treatments were applied.

2.3. Radiation treatments and storage conditions

The radiation chamber consisted of a reflective stainless steel chamber with two lamp banks (one bank suspended horizontally over the radiation vessel and the other one placed below it) being fitted to each bank 6 UV-B and 7 UV-C (alternatively positioned) unfiltered emitting lamps (TL 40W/01 RS and TUV 36W/G36 T8, respectively; Philips, Eindhoven, The Netherlands). UV-B and UV-C radiations were separately applied controlled by two general keys that switched all UV-C or UV-B at the same time. The radiation chamber also had a ventilator continuously switched on during treatments to renovate the air from inside of the chamber with the cold air from the cold room (8 °C). Plant material was placed between the two lines of lamps at 17.5 cm above and below over a bi-oriented polypropylene film (thickness: 35 mm) mounted on a polystyrene (PS) net (130 × 68 cm) that minimized blockage of the radiation. The applied UV-B and UV-C intensities of 9.27 and 25.21 W m^{-2}, respectively, were calculated as the mean of 18 readings on each side of the net using LP 471 UV-B (Delta OHM, Italy) and VLX 254 (Vilber Lourmat, Marne la Vallee, France) radiometers, respectively. Thus, both sides received the same radiation intensities. The radiation chamber is based on that previously described (Artés-Hernández, Escalona, Robles, Martínez-Hernández, & Artés, 2009). The light intensities were kept constant and the applied doses were varied by altering the exposure time at the fixed distance. Applied treatments were:

- CTRL: No radiation treatment used as control.
- Single UV-B treatments: 5 (540 s), 10 (1080 s) and 15 (1619 s) kJ m^{-2} UV-B. Such UV-B doses were selected based on previous experiments and according to Avena-Bustillos et al. (2012) in order to obtain maximum phenolic accumulation while minimizing heating and evaporation processes during UV-B treatment.
- Combined UV-B + UV-C treatments (UV-Bi + C): the previous UV-B doses were applied followed by a UV-C dose of 9 kJ m^{-2} (357 s). Such UV-C was selected based on Martínez-Hernández et al. (2011) and previous tests in order to obtain maximum bioactive increases while preserving food safety.

The treated plant material was then placed in rectangular plastic trays and covered with a black polyethylene bag to reduce water loss. Samples were stored at 15 °C (90–95% RH) up to 72 h (sampling times: 0, 24, 48 and 72 h). Three replicates per treatment and sampling time were prepared. Samples were frozen in liquid nitrogen at every sampling time and stored at − 80 °C until further TPC, TAC and glucosinolates analyses were conducted. Frozen samples were ground to fine powder prior to analyses using liquid N_2 with a mincer (IKA, A 11 basic, Berlin, Germany) at 12,700 × *g* for 10 s.

2.4. Total phenolic content and antioxidant capacity

Extraction to determine TPC and TAC was conducted by homogenization (Ultra Turrax® model 18T, IKA-Werke GmbH & Co. KG, Germany) of 0.5 (floret), 0.25 (leaves) or 1 g (stalk) of ground frozen sample in 3 mL of methanol for 20 s in an ice-water bath. Subsequently, extracts were centrifuged at 13,500 × *g* for 20 min at 4 °C and supernatants were collected and analysed. TPC was analysed by the Folin–Ciocalteu reagent method (Singleton & Rossi, 1965) but with modifications (Cefola et al., 2010; Martínez-Hernández et al., 2011; Serrano, Martinez-Romero, Guillén, Castillo, & Valero, 2006. Briefly, 19 μL of TPC extract was placed on a PS flat-bottom 96-well plate (Greiner Bio-One, Frickenhausen, Germany) and 29 μL of 1 N Folin–Ciocalteu reagent (Sigma, St Louis, MO, USA) was added. Samples were incubated for 3 min in darkness at room temperature. Then, 192 μL of a solution containing Na_2CO_3 (4 g L^{-1}) and NaOH (20 g L^{-1}) was added and the reaction was carried out for 1 h at room temperature in darkness. The absorbance was measured at 750 nm after incubation using a Multiscan plate reader (Tecan Infininte M200, Männedorf, Switzerland). TPC was expressed as chlorogenic acid equivalents in mg kg^{-1} (expressed on a fresh weight basis). Each of the three replicates was analysed in triplicate.

TAC extracts were analysed based on Brand-Williams, Cuvelier, and

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