



Neutral and acidic electrolysed water kept microbial quality and health promoting compounds of fresh-cut broccoli throughout shelf life



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ABSTRACT

The effect of neutral (NEW) and acidic (AEW) electrolysed water (EW) sanitising treatments (both with 70 and 100 mg L⁻¹ free chlorine) on the bioactive profile and microbial quality of fresh-cut 'Parthenon' and kailan-hybrid broccoli throughout 19 days at 5 °C was studied. Disinfection with 100 mg L⁻¹ NaClO was used as control. EW treatments, particularly NEW100, achieved the best microbial reductions after shelf life, being a promising alternative to chlorine in both broccoli cvs. In addition, all EW-treated samples, regardless of pH or free chlorine levels, showed up to 30% higher total phenolic (TP) contents than chlorine-disinfected ones, which reported values of 556.5 (kailan-hybrid) and 444.0 mg gallic acid kg⁻¹ fw ('Parthenon'). The best correlation between total antioxidant capacity (TAC) and TP was attained by the FRAP method with an R² = 0.65–0.68. Throughout shelf life, TP and FRAP-analysed TAC kept stable values, or even slightly increased. On the processing day, the APX, GPX, CAT and SOD activities of chlorine-treated samples were 105.6/115.4, 3783.2/6791.0, 359.0/433.0 and 798.0/1489.0 U g⁻¹ protein for kailan-hybrid/'Parthenon', respectively. SOD and CAT activities of EW-treated samples were 13–46% lower than those of the NaOCl-disinfected samples. Myrosinase activity in kailan-hybrid was 1.5-fold higher than in 'Parthenon'. Generally, the myrosinase activity in EW-disinfected samples was kept during shelf life, while in NaOCl-sanitised samples decreased. In general, both EW treatments seem to be promising techniques for keeping good microbial quality in both fresh-cut broccoli cvs. Furthermore, this alternative water sanitising technique showed better bioactive compounds retention in broccoli.

Industrial relevance: The present study shows for the first time the effects of neutral and acidic electrolyzed water treatments (with two different free Cl concentrations) on the bioactive content (antioxidant capacity, phenolics, and antioxidant enzymes) and microbial quality of two different broccoli cvs. (kailan-hybrid and 'Parthenon') throughout 19 days at 5 °C. These results will be useful for the fresh-cut (FC) vegetable industry since the optimum parameters for the EW treatments of two FC broccoli cvs. are hereby provided in order to accomplish the safety and microbial quality aspects. Furthermore, data regarding to enhancing of health-promoting properties of these FC broccoli cvs. after EW treatments are shown. In that way, the FC industries may supply to the consumers a FC vegetable that meet the safety aspects and with improved health-promoting properties.

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1. Introduction

Kailan-hybrid is a new natural hybrid between kailan (*Brassica oleracea*, Albolabrador group, also called kailan, Chinese broccoli or Chinese kale) and conventional broccoli (*B. oleracea*, Italica group). This kailan-hybrid has a long slender stem and a mild sweet taste with a completely edible portion being a suitable product for the minimal processing or fresh-cut (FC) industry (Martínez-Hernández,

Artés-Hernández, Gómez, & Artés, 2013a). However, the processing steps required good result in microbial proliferation and, currently in the FC vegetable industry, washing with 50–150 mg L⁻¹ NaClO is a sanitising method widely used (Artés, Gómez, & Artés-Hernández, 2007). However, chlorine, among other disadvantages, may be potentially harmful for humans and the environment (Hrudey, 2009). Thus, alternative techniques such as water disinfection with electrolysed water (EW), both neutral (NEW) and acidic (AEW) types, seem to be useful for keeping microbial quality and to prevent cross contamination in several FC horticultural products (Abadías, Usall, Oliveira, Alegre, & Viñas, 2008; Artés, Gómez, Aguayo, Escalona, & Artés-Hernández, 2009; Rico et al., 2008; Tomás-Callejas, Martínez-Hernández, Artés, & Artés-Hernández, 2011) and recently have been described to be effective against moth proliferation (Jemni et al., 2014).

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The inactivation mechanism for EW treatments proposed that oxidation–reduction potential (ORP) could first affect and damage the redox state of glutathione disulfide–glutathione couple, and then penetrate the outer and inner membranes of microorganisms resulting in the bacteria necrosis (Liao, Chen, & Xiao, 2007). The main advantage of EW is its safety. In contrast with the risks of using NaClO, such as skin and membrane irritation and toxicity, EW is not corrosive to skin, mucous membranes, or organic material. Additionally, EW is more eco-friendly than NaClO and is not potentially harmful for human health (Huang, Hung, Hsu, Huang, & Hwang, 2008). Among NEW and AEW, NEW does not affect the surface colour and visual appearance of FC produce. Furthermore, NEW is preferable to AEW, since a neutral pH is less aggressive for the processing equipment. NEW showed great sanitising effects against natural microflora and pathogenic bacteria in several FC vegetables, such as kailan-hybrid, carrots, spinach, bell pepper, potato, cucumber, Japanese radish, lettuce and mizuna baby leaves, lowering microbial counts from 0.6 to 2.6 log units (Abadías et al., 2008; Martínez-Hernández, Artés-Hernández, Gómez, Formica, & Artés, 2013; Rico et al., 2008; Tomás-Callejas et al., 2011).

Sanitising treatments (such as EW), wounding, modified atmosphere packaging (MAP) and chilled storage involved during the processing and shelf life of FC produce have been regarded as abiotic stresses (Jacobo-Velázquez et al., 2011; Martínez-Hernández, Artés-Hernández, Gómez, Formica, et al., 2013). Such stresses may increase the accumulation of reactive oxygen species (ROS) in the plant cells and, in order to mitigate the possible plant cell damage, the enzymatic and non-enzymatic antioxidant systems are activated. Kailan-hybrid is rich in polyphenols (Martínez-Hernández, Gómez, Artés, & Artés-Hernández, 2013), which have been widely reported as non-enzymatic antioxidants. Other non-enzymatic antioxidants present in broccoli are vitamins C and E, folic acid and carotenoids. Members of the enzymatic antioxidant defence system include superoxide dismutase (SOD; EC 1.15.1.1), catalase (CAT; EC 1.11.1.6), ascorbate peroxidase (APX; EC 1.11.1.11), guaiacol peroxidase (GPX; EC 1.11.1.7) and glutathione reductase (GR; EC 1.6.4.2). The super-oxide radical (O_2^-) is dismutated to H_2O_2 by SOD, and CAT, APX and GPX metabolize H_2O_2 to H_2O (Kang & Saltveit, 2001). Furthermore, kailan-hybrid is highly rich in glucosinolates (biologically inactive) which are separated in different cell compartments from the enzyme myrosinase (MYR; β -thioglucoside glucohydrolase; EC 3.2.3.1) which is located in the myrosin cells. However, after tissue disruption (cutting, chewing, etc.) glucosinolates come into contact with the MYR which causes rapid hydrolysis to form, among others, chemopreventive compounds known as isothiocyanates. Several studies have focused on the effects of postharvest treatments on the glucosinolate levels, although there is very scarce information about postharvest treatments on the MYR activity, which is crucial to get the chemopreventive benefits from the glucosinolates (Halkier & Gershenzen, 2006; Jones, Frisina, Winkler, Imsic, & Tomkins, 2010).

However, there is no information on the effects of EW treatments on the levels of the above mentioned bioactive compounds in FC products. In this way, the main aim of this work was to study the effects of NEW and AEW as emergent sanitizers versus a control washing with NaClO on the microbial quality of a new hybrid and a conventional broccoli cv. and their impact on their bioactive compound profiles throughout shelf life.

2. Materials and methods

2.1. Plant material

Kailan-hybrid broccoli (*B. oleracea* Italica group \times Alboglabra group, cv. Bimi®) of 15 to 18 cm length and 'Parthenon' cv. (*B. oleracea* Italica group) were hand-harvested in December from open air commercial cultivation in the southeast Mediterranean area of Spain (Lorca, Murcia). The broccoli was grown according to integrated pest management cultural practices. Immediately after harvesting, broccoli

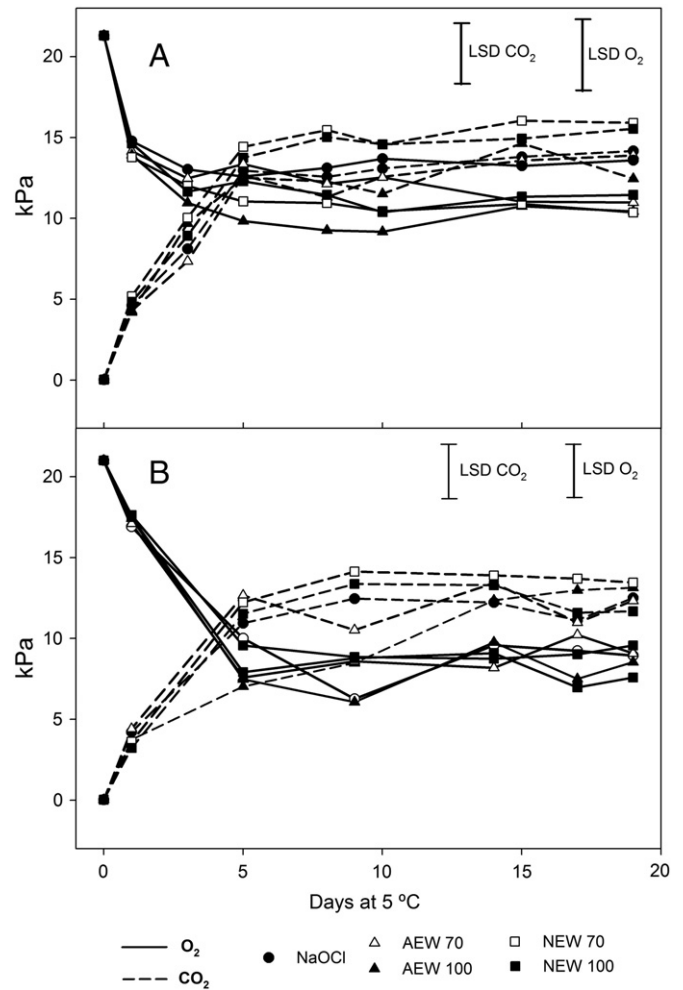


Fig. 1. Gas changes within packages of fresh-cut kailan-hybrid (A) and 'Parthenon' broccoli (B) washed under several disinfection treatments and stored up to 19 days at 4 °C ($n = 5$). Average (CO_2 and O_2) SD values = 1.0 (both A and B). O_2 solid lines; CO_2 dashed lines. Balanced with N_2 .

was selected in a packinghouse for discarding heads and stems with yellowing, decay, cuts or bruises and sound pieces were precooled with crushed ice and transported by car about 80 km to the Technical University of Cartagena. Both kinds of broccoli were then stored at 1 °C and 90–95% RH until the next day, when they were processed.

2.2. Sample preparation, minimal processing and storage conditions

Minimal processing was accomplished in a disinfected cold room at 8 °C. Plant material was inspected, selecting those free from defects and with similar visual appearance. Small leaves of kailan-hybrid broccoli stems (when they were present) were eliminated with a sharp knife. The kailan-hybrid broccoli was cut in about 15-cm-long spears and the 'Parthenon' heads were cut into florets. All broccoli pieces were washed for 1 min with tap water (4 °C) in order to remove organic material. The EW treatment (5 °C; pH 7 ± 0.1 ; ORP = +900 mV) for washing broccoli pieces consisted in two NEW (70 and 100 mg L^{-1} free Cl) and two AEW (70 and 100 mg L^{-1} free Cl). Both EW were produced by an Envirolite EL 400 device (Aquadroja, Madrid, Spain). Subsequently, the broccoli pieces were drained in a perforated basket for 1 min. Contact time was 2 min using a ratio 300-g plant material/5-L disinfectant (w/v). As control, a standard industrial disinfection treatment with NaClO (100 mg L^{-1} ; 5 °C; pH 6.5 ± 0.1) for 2 min was used with the same contact time, plant material/disinfectant ratio and rinsing conditions than EW treatments. During all washing treatments

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