



# Combination of pulsed electric field processing and antimicrobial bottle for extending microbiological shelf-life of pomegranate juice<sup>☆</sup>



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## ABSTRACT

Pomegranate juice was processed using bench top (7.2 L/h flow rate, 35 kV/cm field strength, 72  $\mu$ s total treatment time) and pilot scale (100 L/h flow rate, 35 kV/cm field strength, 281  $\mu$ s total treatment time) continuous pulsed electric field (PEF) processing systems. The treated juice was packaged in PET bottles or PET bottles coated with potassium sorbate and sodium benzoate, and stored at 4 °C for 84 days. Samples were assessed every 7 days for total aerobic bacteria and yeast and mold. Untreated juice had less than one week of shelf-life, while untreated juices in antimicrobial bottles had 56 days. Juices treated with PEF alone had a shelf-life of 21 days (bench scale) and over 84 days (pilot scale). Juices treated with PEF and stored in antimicrobial bottles had a shelf-life over 84 days for both scale tests, which significantly extended the microbiological shelf-life of pomegranate juice.

**Industrial relevance:** Pulsed electric field (PEF), one of novel non-thermal processing technologies, has been studied intensively worldwide for the last decades. However, most of them were done at laboratory scale and few were at pilot or commercial scale. In addition, PEF processing alone may not provide enough shelf-life of juice as juice industry expects. The work in this paper shows the side-by-side comparison of PEF processing at lab and pilot scales and demonstrates that the combination of PEF with antimicrobial bottle packaging significantly extended the shelf-life of juice. The use of a large scale PEF processing system and the combination of antimicrobial packaging provide juice manufacturers an innovative approach for enhancing the safety and extending the shelf-life of juice products.

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## 1. Introduction

Pulsed electric field (PEF) processing has been of growing interest owing to its potential to provide consumers with microbiologically-safe and fresh-like quality foods. Inactivation by PEF is dependent on multiple factors relating to the process conditions, medium, and microbial species (Aronsson, Lindgren, Johansson, & Ronner, 2001; Wouters & Smelt, 1997). These factors may limit the application of PEF as a sole preservation method against pathogenic bacteria under acid conditions, and especially against *Escherichia coli* O157:H7 (Ait-Ouazzou et al., 2012; Garcia, Gomez, Raso, & Pagan, 2005; Garcia, Hassani, Manas, Condon, & Pagan, 2005; Iu, Mittal, & Griffiths, 2001). PEF may therefore be optimally used in combination with other antimicrobial interventions.

The hurdle approach, as described by Leistner (1992), is used to produce minimally processed food by applying several sub-lethal

treatments to achieve microbial stability, rather than relying solely on one lethal preservation method. The microbial stability is achieved by combining these hurdles to increase destruction of the microbial cytoplasmic membrane as well as preventing cell repair of survivors from treatments (e.g. PEF), such as sub-lethally injured cells or bacterial endospores (Galvez, Abriouel, Lopez, & Omar, 2007; Leistner, 2000). Previous studies reported that combining PEF with natural antimicrobials such as bacteriocins, antifungal peptides, essential oils, spices and organic acids can enhance its killing effect on microorganisms in fruit juices (Liang, Mittal, & Griffiths, 2002; Mosqueda-Melgar, Raybaudi-Massilia, & Martin-Belloso, 2008; Nguyen & Mittal, 2007). It has been suggested that the combination of either organic acids or nisin and non-thermal technologies could be effective in the control of undesirable microorganisms in foods (Galvez et al., 2007; Mosqueda-Melgar et al., 2008).

Antimicrobial packaging, which releases antimicrobials into foods from packaging materials, has been widely investigated for various foods. In our previous study (Jin, 2010), glass jars coated with polylactic acid (PLA) containing 250 mg nisin completely inactivated the cells of *Listeria monocytogenes* in skim milk after 3 days and throughout the 42 day storage period at 4 °C. The same coating treatments rapidly reduced the cell numbers of *Listeria* in liquid egg white to undetectable

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level after 1 day, and then remained undetectable throughout the 48 day storage period at 10 °C and the 70 day storage period at 4 °C. In another study, the PLA coating with 500  $\mu$ L AIT completely inactivated 3 and 7 log CFU/mL of *Salmonella* after 7 and 21 days of storage, respectively (Jin & Gurtler, 2011). However, to the best of the authors' knowledge, there are no reported studies combining PEF with antimicrobial bottles. Therefore, the objective of this study was to develop a new approach for pasteurizing and extending shelf-life of juice by combining PEF processing with antimicrobial packaging, using pomegranate juice as a model.

## 2. Materials and methods

### 2.1. Juice

Frozen bulk packaged (20 L per bag) and untreated pomegranate juice were provided by the AMC Group, Spain. The frozen juice was shipped at refrigerated temperature and received within 2 days then stored at  $-20$  °C. The untreated juice was thawed at 4 °C for 3 days prior to PEF processing.

### 2.2. Pulsed electric field processing system and treatment conditions

A bench scale PEF continuous processing system (OSU-4H Model) and a commercial scale PEF continuous processing system (OSU-6 Model) located at Eastern Regional Research Center, Agricultural Research Service USDA (Wyndmoor, PA, USA) were used for this study. Both systems provided bipolar square waveform pulses with a maximum peak voltage of  $\pm 11$  kV and 60 kV, respectively. The high voltage pulse generator operated at a maximum repetition rate of 2000 pulses per second (pps) and pulse width of 1–10  $\mu$ s. Pulses were monitored with a high voltage probe (VD-60; Northstar, Albuquerque, NM, USA), current monitors (Model 110; Pearson, Palo Alto, CA, USA) and oscilloscopes (TDS-210; Tektronix, Beaverton, OR, USA). For the benchtop system, the treated sample was cooled by passing through a cooling coil submerged in a water bath (MultitempWater Bath III, Pharmacia Biotech, AB, Uppsala, Sweden) after passing through each pair of treatment chambers in order to control the final outlet temperature. The inlet and outlet temperatures were monitored by type K thermocouples attached to a dual input digital thermometer (Omega HH509, Omega Engineering Inc., Stamford, CT). For the commercial system, counter flow heat exchangers, controlled by independent PID controllers, maintained the outlet temperature of each chamber at 55 °C. Fig. 1 shows an overview of each system. The treatment conditions are listed in Table 1.

### 2.3. Antimicrobial bottle coatings

Nine hundred micrograms of potassium sorbate (99%, Fisher Scientific, Fairlawn, NJ) and 1500 mg of sodium benzoate (99%, Fisher Scientific, Fairlawn, NJ) were accurately weighed and added to 100 mL methylene chloride (Fisher Scientific, Fairlawn, NJ), stirring with a magnetic stir bar under chemical hood over night until solid compounds were completely dissolved. Ten microliters of this mixture was taken into a vial and 1 g of PLA resin (NatureWorks, Minnetonka, MN) was added to the mixture, stirring for 6 h. The mixture with PLA was transferred into pre-cleaned 8 oz PET bottle (U.S. Plastic Corp, Lima, OH, USA), which were rolled horizontally on a hot dog roaster machine, allowing antimicrobial mixtures to coat the inside wall of each bottle for 30 min. The methylene chloride was evaporated during the bottle's rolling and after the coating at room temperature (ca. 22 °C) under a chemical hood for 24 h, sealed with caps and stored until time of use (within 3 days). Fig. 2 shows the bottles before juice packaging.

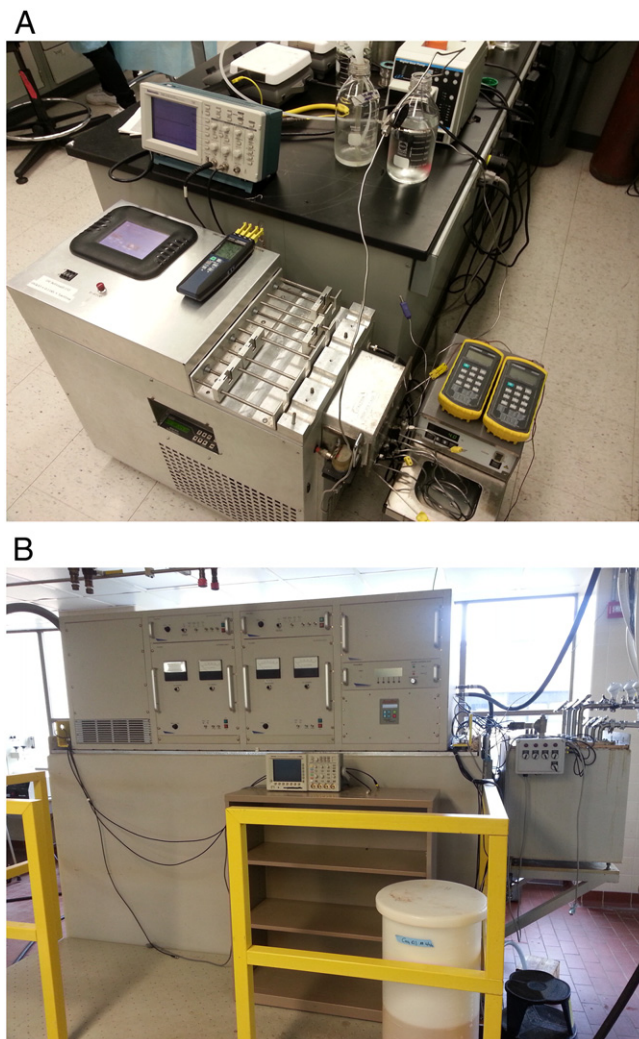


Fig. 1. Overview of PEF processing system. A: Bench top system; B: commercial scale system.

### 2.4. Juice packaging and storage

PEF-processed pomegranate juices were collected inside a sanitary laminar hood using connection tubing from PEF outlet to the hood where juices (200 mL) were packaged into regular PET bottles or antimicrobial-coated PET bottles. Untreated juices were packaged in the same way and used as controls. All juice samples were stored at 4 °C.

Table 1  
PEF processing parameters.

Processing parameter	Pilot scale	Lab scale
Flow rate (L/h)	100	7.2
Gap distance (cm)	1.27	0.29
Inner diameter (cm)	0.807	0.23
Electrical conductivity of juice (s/m)	0.358	0.358
Maximal outlet temperature (°C)	55	55
PEF treatment time ( $\mu$ s)	281	72
Electric field strength (kV/cm)	35	35
Pulse repetition rate (pps*)	2000	2000
Pulse duration ( $\mu$ s)	1	1
Number of PEF treatment chambers	6	6
Initial temperature of pomegranate juice (°C)	4	4

\* pps = pulse per second.

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