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Extending the Shelf-Life of Tomato Using By-Product from Cheese Industry.

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EXTENDING THE SHELF-LIFE OF FRESH-CUT TOMATO USING
BY-PRODUCT FROM CHEESE INDUSTRY

Running Title
Whey permeate on Quality of Fresh-cut tomato

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The effects of three whey permeates were investigated as potential natural washing treatment for fresh-cut tomato and compared with chlorine (120 ppm). Whey permeate treated samples resulted in equivalent or better than chlorine for all these attributes. Whey permeates were equally effective as chlorine to control the micro-organisms of fresh-cut tomato during storage. The microbial counts at day 10 were significantly reduced (~1.0 log CFU/g) in all the treated samples compared to the control (water treated) samples. Moreover whey permeate treated fresh-cut tomatoes showed lower water activity (2 %) and POD activities (21%) than chlorinated samples after 10 days of storage. Whey permeate also inhibited the loss of firmness of treated tomato slices. Sensory scores for aroma were significantly higher in whey permeate treated samples than chlorine treated samples. Among the three types of whey permeate, delactosed permeate (DP) showed the best results in maintaining the quality of fresh-cut tomato.

The market sales of ready-to-eat fresh vegetables have grown rapidly in recent decades as a result of changes in consumer attitudes. The marketing of fresh-cut vegetables is limited by their short shelf-life due to the quick decline in post-processing quality. Many attempts have been made to increase the shelf life of fresh-cut fruit and vegetables and many attempts have been made to increase the use of whey permeate, a valuable by-product of cheese processing industry. This paper takes an interesting approach by attempting to use whey permeate as a preserving agent of fresh-cut tomato quality during storage.

**KEY WORDS:** Whey permeate; Fresh-cut; Tomato; Shelf-life; Quality; Preservation.
Continued growth in the ready-to-eat vegetable industry has been largely driven by increasing demand for convenient, fresh and healthy foods. Increasing the quality retention and shelf-life of these products during storage is an important demand of the industry and consumers (Rico et al. 2007). Chlorinated water (50–200 ppm) is widely used to wash fruits and vegetables as well as fresh-cut produce in order to preserve their quality. However, the possible formation of carcinogenic chlorinated compounds in water (chloramines and trihalomethanes) has called into question the use of chlorine for this purpose (Alegria et al. 2010). As soon as a suitable alternative to chlorine is available to industry, chlorine will be banned throughout the world. In several European countries including Germany, Netherlands, Switzerland and Belgium the use of chlorine in fresh-cut fruits and vegetables is already prohibited. Future regulatory restrictions on the use of chlorine for washing of ‘ready-to-eat’ vegetables are likely and will require the development of safe alternatives (Ölmez and Kretzschmar 2009). Therefore anything that is as cheap as chlorine and performs close to or equal to chlorine is desired by industry. In recent years interest is growing in the use of natural products for the preservation of fresh-cut produce. Research and commercial applications have shown that natural components could replace traditional washing agents (Rojas-Graü et al. 2009). The development of chlorine-free fruit and vegetable products enriched with natural bio-products could contribute greatly to a new and growing market, where the consumer's concerns about their health are met.

Whey permeate is a by-product of the production of whey protein concentrates from cheese whey. The main composition of whey permeate are water, lactose, low molecular peptides and minerals. Whey is used as a fermentation feedstock for the production of lactic acid, acetic acid, propionic acid, ethanol, and single cell protein, etc (Panesar et al. 2007). However, these
applications still do not utilize all the whey produced and new uses for this by-product are needed. Whey and whey ultrafiltrated permeate have been proposed to be used as a natural antioxidant in foods (Contreras et al. 2011). Whey proteins and peptides contain all the essential amino acids (Yalcin 2006). Lactoferrin, α-lactalbumin and β-lactoglobulin are a source of antimicrobial proteins. Casein macro peptide (CMP), α₁– and α₂– caseins are further examples of whey antimicrobial peptides (Rizzello et al. 2005). Martin-Diana et al. (2006) used acidic whey permeate for washing fresh-cut lettuce and carrots during storage. Whey protein has been found to reduce the enzymatic browning of ‘Golden Delicious’ apples (Perez-Gago et al. 2006). Coronadoa et al. (2002) successfully used rosemary extract and whey powder for the oxidative stability of wiener sausages during 10 months frozen storage.

Tomato is one of the most widely used and versatile vegetable crops. It is consumed fresh and also used to manufacture a wide range of processed products. The consumption of tomatoes is currently considered as a nutritional indicator of good dietary habit and healthy lifestyle. This fruit has undoubtedly assumed the status of a food with functional properties, considering the overwhelming epidemiological evidence for its capacity to reduce the risk of chronic diseases such as cardiovascular disease and cancer (Sgherri et al. 2008). This protective action is typically attributed to antioxidant components like lycopene and other carotenoids (pro-vitamin A, beta-carotene), ascorbic acid, vitamin E and flavonoids (Odriozola-Serrano et al. 2008).

Therefore the aim of this study was to examine the effect of whey permeate treatment on the quality retention of fresh-cut tomatoes during storage as an alternative to chlorine.

2. MATERIALS AND METHODS

2.1. Sampling and Treatment Design
Irish vine ripened tomatoes (*Lycopersicon esculentum* L. Mill.) cv. Moneymaker were purchased from a local supermarket (Dunnes Stores). According to the grower, the tomato plants were grown in a greenhouse with a 14 h light period from February until November. The atmosphere of the greenhouse as well as crop irrigation and nutrition were precisely controlled. The temperature of the greenhouse was 16–21 °C which is optimum for lycopene synthesis in tomato fruit. The tomatoes were transported to the food processing lab and stored at 4 °C before processing. An isolated and cleaned minimal-processing room was used for tomato processing. The experiments were carried out between March and November, 2009. Three independent trials were carried out. Each experiment was conducted with 180 fresh-cut tomato packages {4 test days (1, 4, 7 and 10) × 5 treatments × 3 replications × 3 individual batches}.

### 2.2. Preparation of Treatment Solution

Three different types of whey permeate (liquid) were kindly supplied by Glanbia Ltd. Ingredients, Ireland. The permeate concentrate (PC) was pre-concentrated by evaporation before the lactose crystallisation process. Delactosed permeate (DP) was obtained after removal of lactose crystals. The delactosed permeate (DP) was then concentrated further by evaporation to give delactosed concentrate (DC). The main components of the three whey permeates were given in Table 1.

Five washing treatments were conducted in parallel, using the same batch of product. The samples were sanitised with water, chlorinated water (120 ppm) or whey permeates (PC, DP and DC) at 3 % concentration (Ahmed *et al.* 2011). Chlorinated water was prepared by adding sodium hypochlorite solution (≥ 5 % active chlorine, Aldrich Chemical Co., Dublin, Ireland) to distilled water to obtain a final solution containing ~120 ppm free chlorine (pH 8.0) (Delaquis *et al.* 2004). All treatments were prepared using distilled water.
2.3. **Processing and Experimental Setup**

Whole tomatoes were rinsed briefly (1 min) in water prior to washing to remove soil contamination. Washing treatments were performed by immersion of the tomatoes in each treatment solution for 1 min (with agitation). Each treatment was carried out in different baskets (200 g tomatoes/ L). After washing, the tomatoes were dried for 5 min using a salad spinner. Then they were sliced in 6 mm thick transversal slices with a commercial slicing machine (Maxwell chase MCT-25, Baltimore Innovations, UK). Processed tomatoes were then pooled, mixed and ~100 grams placed in a polypropylene tray (180 mm length × 130 mm width × 25 mm depth) from Sharp Interpack Ltd., UK containing one layer of absorbent paper on the bottom (Fresh-R-Pax absorbent pads, Maxwell Chase Technologies, Atlanta). The principal ingredient in fresh-R-Pax absorbent pads is food grade sodium carboxymethyl cellulose (CMC), a common ingredient in ice-cream, sauces, low-fat foods, etc. The trays were then packaged in bags (200 × 320 mm) of 35 µm oriented polypropylene film (OPP) with permeability at 23 °C and 90 % RH of $3.3 \times 10^{-12}$ mol/s/m$^2$/Pa for O$_2$ and $3.1 \times 10^{-9}$ mol/s/m$^2$/Pa for CO$_2$ (Amcor Flexibles Europe-Brighouse, United Kingdom). The packages were then heat-sealed under atmospheric conditions and stored at 4 °C for 10 days (Ahmed *et al.* 2011).

2.4. **Markers Analysis**

Different physico-chemical (headspace gas composition, pH, moisture content, water activity), instrumental analysis (color, texture, sensory), enzymatic activity (browning related enzyme – peroxidase and texture related enzyme – pectin methyl esterase), and microbiological (mesophillic, psychotropic, lactic acid bacteria, yeast and moulds) markers were monitored throughout the storage (10 days) of fresh-cut packaged tomatoes stored at 4 °C.

2.4.1. **Physico-chemical Markers**
Headspace Gas Composition

Changes in O\(_2\) and CO\(_2\) concentration of the headspaces of the fresh-cut tomatoes packages were monitored during the shelf life of the product. A Gaspace analyser (Systech Instruments, UK) was used to monitor levels of O\(_2\) and CO\(_2\). Gas extractions were performed with a hypodermic needle, inserted through an adhesive septum previously fixed to the bags, at a flow rate of 150 mL/min for 10 sec. Three bags per treatment were monitored for each experiment and all bags for other analyses were checked before analysis.

\(pH\)

Ten gram of tomato tissue was blended for 2 min. Then the pH was measured at room temperature using an Orion research pH-meter, UK.

Moisture Content

Moisture content was determined by AOAC method (1990) (Method 925.098). The tomato samples were dried at 105 °C overnight.

Water Activity

The water activity of the treated samples was measured with a fast water activity meter (GBX scientific FA-st/1, Cedex, France). One gram of tomato was placed in a small plastic cup onto the base of the air tight test chamber. The measuring head enclosed the sample and formed an airtight seal with the base. At least 15 samples were measured per treatment.

2.4.2 Instrumental Analysis

Color

For color analysis each piece of tomato in the storage pack was analyzed individually to minimize the variability of the product. Color was quantified using a Color Quest XE colorimeter (HunterLab, Northants, UK). A tomato slice was placed directly on the colorimeter
sensor (3.5 cm of diameter) and measured; 20 – 30 measurements were taken per treatment and day. Before measuring the instrument was calibrated using a white tile (L* = 93.97, a* = 0.88 and b* = 1.21) and a black tile (L* = 56.23, a* = 21.85, b* = 8.31) standards. Hunter color readings were recorded. The L* parameter (lightness index scale) range from 0 (black) to 100 (white). The a* parameter measures the degree of red (+a*) or green (−a*) color and the b* parameter measures the degree of yellow (+b*) or blue (−b*) color. The CIE L* a* b* parameters were converted to Hue (arctan b*/a*) and Chroma (a*²+b*²)⁰⁵. The translucency assessment was conducted visually on the outer pericarp of the tomato slices. The evaluation was done under uniform light conditions.

Texture

Four measurements were made on each slice, two in the outer pericarp and two in the radial pericarp, applying the force in the axial direction. Care was taken to standardize the measurements in the radial pericarp at half radius and in the outer pericarp between two junctions of outer and radial pericarp. The force necessary to cause a deformation of 3mm with a speed of 0.02 mm/s was recorded using an Instron Texture Analyzer (Instron 4302 Universal Testing Machine, Canton MA, USA), with a 3.5 mm diameter flat faced cylindrical probe. Only the central slice in the stack was used in the analyses. The firmness measurement was performed immediately after removing the slice from the storage chamber (at storage temperature). Data were analysed with the Instron series IX software for Windows.

Sensory Analysis

Sensory analysis was performed for tomato samples over 10 days of storage time by a panel with an age range of 25 – 40 years. Fresh appearance, colour, texture, aroma and general acceptability of samples were scored on a hedonic scale of 1 to 9, where a score of one indicated a product of
very poor quality, etc (Ferreira et al. 2008). The evaluation was carried out in the sensory evaluation laboratory. Products were coded using random numbers to avoid bias. Products were placed in plastic cups with lid, on a white surface and judges were isolated from each-other in a booth in an odour-free environment. The sensory analysis was monitored with Compusense Five software (Release 4.4, Ontario, Canada).

2.4.3. Enzymatic Activity

*Browning-related Enzyme - Peroxidase (POD)*

POD enzyme was assayed in homogenates that were prepared as follows: 10 g of tomato puree was placed in a 100 mL beaker in a 1:2 (w:v) ratio with 0.5 M phosphate buffer, pH 6.5, containing 50 g/L polyvinylpyrrolidone. Then homogenization was carried out twice with an Ultra-Turrax T-25 tissue homogenizer at 4 °C and 20,500 rpm, for 1 min each time with a break of 3 min between homogenizations to avoid excess heating of the sample. The homogenate was centrifuged at 12,720 g for 30 min at 4 °C. It was then filtered through Whatman no. 4 filter paper. The resulting crude extract was used without further purification. All the extracts were kept at 4 °C in the dark and used immediately (within 1 h). POD activity was assayed spectrophotometrically by a modified method based on (Martin-Diana et al. 2006). The reaction mixture contained 0.2 mL of extract and 2.7 mL of 0.05 M phosphate buffer, pH 6.5, containing 1.85 mL of hydrogen peroxide (1.5 %, v/v) as oxidant and 3.7 mL of p-phenylenediamine as hydrogen donor. The oxidation of p-phenylenediamine was monitored at 485 nm and 25 °C. A unit of enzyme activity was defined as an increase of 0.1 absorbance units per minute.

*Texture-related Enzyme - Pectin Methyl Esterase (PME)*

PME activity was measured using the method described by Yoo et al. (2009). Ten grams of tomato puree was diluted in an extraction solution (0.2 M sodium phosphate buffer, pH 7.5
containing 1 M sodium chloride and 10 mM dithiothreitol) and homogenized at 4 °C for 2 min at 5,500 rpm. The macerate was incubated at 4 °C for 30 min with agitation and centrifuged at 12,720 g for 30 min at 4 °C. It was then filtered through Whatman no. 4 filter paper. One milliliter of this extract was mixed with 40 mL of substrate solution (1 % pectin). The solution was adjusted to pH 7.0 with 1.0 M NaOH, and the pH of the solution was readjusted to pH 7.5 with 0.05 M NaOH. After the pH reached 7.5; 0.2 mL of 0.05 N NaOH was added. The time required to return to pH 7.5 was recorded. Activity was quantified as carboxyl groups formed by the hydrolysis of methyl esters of pectin and was measured tritrimetrically using a pH electrode to monitor the production of H⁺ (Martin-Diana et al. 2005). The enzymatic activity can be described by the following equation:

\[
PME = \frac{0.2 \text{[mL NaOH]} \times 0.05 \text{[Mol L}^{-1}] \times \text{NaOH} \times X \text{[mL]} \times 10^3 \times \text{[mMol L}^{-1}] \times 10^3 \times \text{[L mL}^{-1}] \times \text{Y [mL]} \times \text{Z [g]} \times \text{time [min]}}{Y \text{[mL]} \times Z \text{[g]} \times \text{time [min]}}
\]

Where, \( X \) = total volume (mL) extracted, \( Y \) = volume used (1 mL) in the assay, \( Z \) = sample used in the assay (10 g). Three macerates per treatment and day were prepared. Triplicates of the enzymatic activity were analyzed.

### 2.4.4. Microbiological Markers

Microbiology analyses were carried out on the samples before and after the treatment at regular intervals through the storage period. 25 g of tomatoes were blended in 225 mL of peptone saline with a Stomacher circulator homogenizer. Enumeration and differentiation of mesophilic bacteria were quantified at 30 °C in plate count agar (PCA) over 72 hrs. Psychrotrophic bacteria were quantified on plate count agar (PCA) at 4 °C over 7 days. Enumeration of lactic acid bacteria was carried out using deMan Rogosa Sharpe Agar (MRS) at 35 °C over 48 hrs. Yeast and moulds were quantified at 25 °C in potato dextrose agar (PDA) over 72 hrs. The results were expressed as log10 colony forming units per gram (CFU/g).
2.5. Statistical Analysis

Data were analyzed by multivariate analysis of variance (MANOVA) using Statgraphics software (version: centurium XV; Statistical Graphics Co., Rockville, USA) for different washing treatments. Analysis of variance one-way (ANOVA) was used to analyze each treatment over storage. In the case of significant differences the LSD range test (p<0.05) was used.

3. RESULTS AND DISCUSSION

3.1. Physico-chemical markers

Headspace Gas Composition

Headspace gas (O₂ and CO₂) composition within fresh-cut tomato packages significantly (p<0.05) changed over storage. Oxygen decreased from atmospheric levels (21% - packaging conditions) to values around 19% at day 1 and levels around 14% by day 10. A sharp increase in carbon dioxide was observed in 24 hours (day 1), from 0 to 3%, reaching values around 7% at the end of storage (day 10). This increase in CO₂ after cutting of fresh tomatoes has been previously described (Gil et al. 2002). Similar gas concentration levels at day 10, with good quality retention on fresh-cut tomatoes, were attained in previous work by Artés et al. (1999) using also passive MAP. The treatments did not show any significant (p<0.05) effect on headspace gas composition as the pattern of change was the same over time.

pH

The pH values of the samples treated with chlorine were significantly (p<0.05) higher than those of samples treated with whey permeate during all storage. The pH increased significantly (p<0.05) over storage in all the samples. This is in agreement with other authors (Artés et al. 1999). At day 10 the lowest pH appeared in samples washed with DP followed by DC and PC,
although the difference among the whey permeate treated samples was not significant (Fig. 1C). The increase in pH during storage might be associated with bacterial growth (Rico et al. 2007).

**Moisture Content**

All the samples in the present study maintained moisture content values around 94%. This was in agreement with findings reported for tomatoes by Hernandez-Suarez et al. (2008). In fact, tomato is one of the highest water containing vegetables. Samples treated with whey permeates had significantly (p<0.05) higher water content values than those treated with chlorine and water (Fig. 1D). The possible reason for the higher moisture content in the whey treated samples could be that whey might form a thin layer on the sample surface which could prevent water loss (Rojas-Graü et al. 2007). Samples treated with water showed the lowest water content at the end of the storage.

**Water Activity**

Significantly (p<0.05) lower water activity was observed in samples treated with whey permeate (no differences among the whey permeates) than those treated with chlorine or water (Fig. 2A). A significant increase was observed in all samples during storage. Water activity is considered an indirect indicator of growth of micro-organisms and most degradation reactions of a chemical, enzymatic and physical nature (Rico et al. 2007). In agreement to this, results obtained for POD activity and microbial counts showed a similar trend (section 3.3 and 3.4). The lower water activity obtained by the permeate treatments positively affected the textural properties of fresh-cut tomatoes during storage, according to data from sensorial and instrumental texture (section 3.2).

**3.2. Instrumental analysis**

**Color**
Fresh-cut tomatoes showed a significant decrease in luminosity during storage (p<0.05). This was in agreement with the work of Lana et al. (2006) using video image analysis of fresh-cut tomato. Luminosity of the samples (L*) was not affected by treatment (Table 2), as well as a* and b* parameters. However, the parameter a* increased significantly (p<0.05) during storage. This is an indicator for red color development and the degree of ripening in tomato (Lana et al. 2006). Hue has a negative correlation with the maturity of the tomato. As tomato matures during storage, Hue decreases. The decreasing trend of b* values throughout the storage showed that the fresh-cut tomatoes did not have any chilling injury stored at 4 °C as it is the optimum storage temperature of fresh-cut fruits and vegetables (Silviera et al. 2010). The authors found that the temperature used during storage did not influence significantly (p<0.05) the development of translucency, indicating that the water soaking of the pericarp tissue is not a result of chilling injury. The intensity of translucency depends on the maturity of the tomatoes.

**Texture**

Texture (firmness) decreased significantly (p<0.05) during storage for all treatments (Fig. 2B). These instrumental results correlated with the sensory panel results (Fig. 3). All three whey permeates treatment maintained significantly (p<0.05) better texture than chlorine or water treatment. The presence of calcium in whey permeates might help to maintain the firmness of tomato during storage (Evans et al. 2010). Different calcium salts have been used for firmness improvement of fresh fruits and vegetables. Calcium carbonate and calcium citrate are the main calcium salts added to foods in order to enhance the nutritional value. Calcium chloride has been widely used as preservative and firming agent in the fruits and vegetables industry for whole and fresh-cut commodities (Chardonnet et al. 2003). Differences between the whey permeate treatments were also observed. Samples washed with DP maintained significantly (p<0.05)
higher texture throughout the storage than DC and PC. There was no significant difference between PC and DC treatments.

Sensory Analysis

All the attributes evaluated (such as, texture, aroma, first impression, general acceptability) except for color (probably due to ripening) decreased significantly (p<0.05) during storage which is associated with a loss of quality (Fig. 3). However, the values at the end of the storage (10 days) were still above the acceptability threshold of 5 for all the attributes scored. The non-hypoxic O$_2$ and CO$_2$ concentration in the packages might have helped to maintain acceptable levels of colour and aroma (Aguayo et al. 2006). The treatments did not affect the visual changes and color parameter of the samples (data not shown). Significant differences were observed between treatments for aroma, texture and general acceptability scores. Lowest scores of texture and general acceptability were observed in water treated samples throughout the storage. In general, whey permeate treatments scored significantly higher (p<0.05) or equivalent to chlorine treatments. Among the whey permeates the general acceptability of delactosed permeate (DP) was the highest, this was in agreement with most of the physico-chemical markers of fresh-cut tomatoes studied in the current research. The use of whey permeate for food preservation has been examined by Nykänen et al. (1998). These authors analyzed the effect of nisin-whey permeate washing solutions on total counts and sensory characteristics in rainbow trout. They found that nisin-whey treatment caused no negative effect on sensory attributes.

3.3. Enzymatic activity

Browning-related Enzyme- Peroxidase (POD)

Fresh-cut tomato treated with whey permeate had lower (p<0.05) POD activity compared to chlorine and water treated samples (Fig. 2C). This lower POD activity might be associated with
the potential antioxidant activity of the whey permeate (Ahmed et al. 2011). A significant (p<0.05) increase in POD activity in all the treatments was observed during storage. The initial increase at day 3 might be due to mechanical stress during minimal processing (Cantos et al. 2001). The depletion of antioxidants might attribute to the sharp increase of POD activity in whey treated samples at the end of storage (day 7 to 10). Differences in POD activity were not observed for different types of whey permeate. Perez-Gago et al. (2006) found that edible composite coatings prepared from whey protein concentrate (WPC) and beeswax (BW) with ascorbic acid or 0.5 % cystein (Cys) reduced the enzymatic browning of ‘Golden Delicious’ apples.

Texture-related Enzyme- Pectin Methyl Esterase (PME)

Water and chlorine-treated samples showed significantly (p<0.05) lower PME activity than those treated with whey permeate (Fig. 2D). Differences between treatments were observed as samples washed with DP had the highest PME activity of all the treatments followed by DC and PC. PME activity increased significantly (p<0.05) during storage for all treatments, although showing fluctuations over storage, which might be due to a wounding response and/or to changes in the solubility of the enzyme during storage (Cantos et al. 2001). Such behaviour has been observed for certain browning related enzymes (Perez-Gago et al. 2006). Other authors have attributed this variability to intrinsic factors and to pre- and postharvest factors, which can affect enzyme activity, vitamin content, etc (Yoo et al. 2009).

3.4. Microbiological Markers

Fresh-cut tomatoes stored at 4 °C had initial loads of total mesophilic bacteria approximately ~ 4 log CFU/g (Fig. 4A). At day 10 samples treated with water reached the highest values (~ 8.5 log CFU/g). The whey permeate treatments were equally effective as chlorine in controlling the
bacterial growth. In fresh-cut tomatoes, total mesophilic counts increased during storage for all the washing treatments. Samples treated with whey permeates and chlorine showed better reduction in the mesophilic counts (~ 1.2 log CFU/g) than samples treated with water, with significant (p<0.05) differences at days 7 and 10 of storage. The values of whey permeate-treated samples at the end of the storage were lower than the recommended $10^8$ CFU/g for consumer consumption of fresh-cut vegetables (Alegria et al. 2009).

Psychrotrophic counts on sliced tomatoes at day 10 were significantly reduced in all the treated samples compared to the control (water treated) samples (Fig. 4B). The highest reduction (1.2 log CFU/g) at day 10 was observed in samples treated with whey permeates followed by samples treated with chlorine (~ 0.8 log CFU/g). Psychrotrophic bacteria initial (day 1) load was approximately 3.5 log CFU/g. The highest microbial load was observed in control samples (water treated), which at day 10 reached ~ 7.0 log CFU/g. The three whey permeate and chlorine treatments resulted in significantly lower psychrotrophic counts than water over storage (p<0.05), and by the end of storage (day 10) whey-treated samples showed lower counts than those treated with water and chlorine (p<0.05).

Fresh-cut tomatoes stored at 4 °C had initial loads of LAB approximately 4 log CFU/g. In all the treatments LAB counts significantly (p<0.05) increased during storage. Chlorine, DP and DC treatments showed a significant reduction (~ 1.0 log CFU/g) in LAB growth compared to water and PC treatments (Fig. 4C). Chlorine treatment maintained the lowest LAB load, followed by DC and DP. PC and water-treated samples showed the highest LAB counts. This might be explained by the presence of lactose in PC which could facilitate the growth of LAB. High LAB loads are associated with fermentation processes (off-odours). However, controlled growth of
these bacteria can have a positive antimicrobial effect on the vegetable due to their production of bacteriocins (Rico et al. 2007).

Fresh-cut tomatoes stored at 4 °C had initial loads of yeast and moulds approximately 4 log CFU/g. This result was in agreement with the finding of Prakash et al. (2002) in diced tomato. Yeast and moulds count increased significantly (p<0.05) with storage time (Fig. 4D). Water showed the highest yeast and moulds growth (~ 8.3 log CFU/g, at day 10). DP treated tomatoes showed significantly (p<0.05) lower counts than chlorine treated samples. DC and PC treatments showed similar effect on yeast and moulds counts to chlorine treatment. Whey permeate and chlorine treatments exhibited significant (p<0.05) reduction (~1.0 log CFU/g) in yeast and moulds growth than the control (water treated) samples.

The antimicrobial capacity of whey permeate can be explained by a variety of factors. The most likely factor is the acid pH of the wash treatment which can have a direct effect on the initial microbial count reduction and on subsequent growth during storage. Another factor can be the presence of lactic acid, which can enter the cells in an un-dissociated form. And finally, the presence of antibacterial peptides in the whey permeate might contribute to its antimicrobial capacity (Clare and Swaisgood 2000). Different antimicrobial peptides have been identified from whey protein hydrolysates. These antimicrobial peptides act against different gram-positive and gram-negative bacteria (Escherichia, Helicobacter, Listeria, Salmonella and Staphylococcus), yeasts and filamentous fungi (Rizzello et al. 2005; Fitzgerald and Murray 2006) and different mechanisms of action have been suggested for whey antimicrobial peptides (Saint-Sauveur et al. 2008; Gauthier et al. 2006).

4. CONCLUSION
In this study, delactosed permeate performed better or similar to the industrial standard chlorine in retaining the quality of fresh-cut tomatoes. Although (DP) treatment showed similar results to chlorine in retaining some of the quality markers, considering the potential harmful effect of chlorine DP could be considered as the best treatment for keeping fresh-cut tomato quality acceptable during 10 days of storage. The quality retention capacity of whey permeate can be explained by a variety of factors. The most likely factor is the acid pH of the wash treatment which can have a direct effect on the initial microbial count reduction and on subsequent growth during storage. Another factor could be the presence of antibacterial peptides (caseinmacropeptide or bacteriocins) in the whey permeate might contribute to its antimicrobial capacity. Although further investigations on pathogens are recommended, whey permeate seems to be a good quality retention method of fresh-cut tomato and to control the growth of bacteria associated with its quality deterioration.

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5. REFERENCES


**TABLE 1. MAIN COMPOSITION OF THE THREE WHEY PERMEATES**

<table>
<thead>
<tr>
<th>Whey Permeates</th>
<th>pH</th>
<th>Total Solid %</th>
<th>Protein %</th>
<th>Dry Matter %</th>
<th>Ash %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Permeate concentrate (PC)</td>
<td>5.48</td>
<td>34.61</td>
<td>1.06</td>
<td>3.06</td>
<td>3.25</td>
</tr>
<tr>
<td>Delactosed concentrate (DP)</td>
<td>5.01</td>
<td>32.90</td>
<td>2.83</td>
<td>8.60</td>
<td>8.14</td>
</tr>
<tr>
<td>Delactosed concentrate (DC)</td>
<td>5.75</td>
<td>53.25</td>
<td>3.77</td>
<td>7.08</td>
<td>10.96</td>
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</table>
TABLE 2. CHANGES IN COLOR IN FRESH-CUT TOMATO STORED AT 4 °C, TREATED WITH 120 PPM CHLORINE (CH), WATER (W) AND WHEY PERMEATE CONCENTRATE (PC), DELACTOSED PERMEATE (DP) AND DELACTOSED CONCENTRATE (DC)\textsuperscript{1}.

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\textsuperscript{1}Values designated by the different letters are significantly different (p<0.05). Lowercase letters are used for comparisons during storage and uppercase letters for treatment comparisons. Three independent trials were carried out in triplicate.
FIG. 1. EFFECT OF TREATMENTS ON O$_2$ (A), CO$_2$ (B), PH (C) AND MOISTURE CONTENT (D) IN FRESH-CUT TOMATO PACKAGES OVER 10 DAYS STORAGE AT 4°C. POINTS DESIGNATED ON ANY CURVE BY THE DIFFERENT LETTERS ARE SIGNIFICANTLY DIFFERENT (P<0.05). LOWER CASE LETTERS ARE USED FOR COMPARISONS DURING STORAGE AND UPPER CASE LETTERS FOR TREATMENT COMPARISONS. THREE INDEPENDENT TRIALS WERE CARRIED OUT IN TRIPLICATE.

FIG. 2. EFFECT OF TREATMENTS ON WATER ACTIVITY (A), TEXTURE (B), POD (C) AND PME ACTIVITY (D) IN FRESH-CUT TOMATO PACKAGES, MONITORED OVER 10 DAYS STORAGE AT 4 °C. POINTS DESIGNATED ON ANY CURVE BY THE DIFFERENT LETTERS ARE SIGNIFICANTLY DIFFERENT (P<0.05). LOWER CASE LETTERS ARE USED FOR COMPARISONS DURING STORAGE AND UPPER CASE LETTERS FOR TREATMENT COMPARISONS. THREE INDEPENDENT TRIALS WERE CARRIED OUT IN TRIPLICATE.

FIG. 3. SENSORY EVALUATION OF FRESH-CUT TOMATOES AFTER TREATMENT STORED AT 4 °C FOR 10 DAYS. VALUES DESIGNATED ON ANY CURVE BY THE DIFFERENT LETTERS ARE SIGNIFICANTLY DIFFERENT (P<0.05). LOWERCASE LETTERS ARE USED FOR COMPARISONS DURING STORAGE AND UPPERCASE LETTERS FOR TREATMENT COMPARISONS. THREE INDEPENDENT TRIALS WERE CARRIED OUT IN TRIPLICATE. FRESH APPEARANCE (9 = EXCELLENT, 1 = POOR); AROMA (9 = FRESH, 1 = ROTTEN); COLOUR (9 = DARK, 1 = LIGHT); TEXTURE (9 = VERY CRISPY, 1 = SOFT); OVERALL ACCEPTABILITY (9 = EXCELLENT, 1 = POOR).

FIG. 4. EFFECT OF WASHING TREATMENTS ON MICROBIAL LOAD: MESOPHILIC (A), PSYCHROTROPHIC (B), LACTIC ACID BACTERIA (C) AND YEAST AND MOULDS (D) DURING 10 DAYS STORAGE OF FRESH-CUT TOMATO AT 4 °C. POINTS DESIGNATED ON ANY CURVE BY THE DIFFERENT LETTERS ARE SIGNIFICANTLY DIFFERENT (P<0.05). LOWERCASE LETTERS ARE USED FOR COMPARISONS DURING STORAGE AND UPPERCASE LETTERS FOR TREATMENT COMPARISONS. THREE INDEPENDENT TRIALS WERE CARRIED OUT IN TRIPLICATE.