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#### Review

# Calcium for extending the shelf life of fresh whole and minimally processed fruits and vegetables: a review

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The preservation of quality of fresh products is relevant for the industry due to its economic impact. This paper presents a comprehensive review of the use of different sources of calcium to preserve fresh fruits and vegetables in order to extend the shelf life and enhance the nutritional value. Emphasis is on discussing about the best sources of calcium, concentration, temperature and method of application, suitability of the commodities; and some hints for the cost/benefit analysis are presented.

#### Introduction

Fruit and vegetable consumption is growing rapidly in recent years. Associated with the new consumer's profile

"rich in cash/poor in time", there is a demand for readyto-eat products. For this reason, the market of minimally processed fruits and vegetables has grown rapidly in recent decades as a result of changes in consumer attitudes (An Bord Glas, 2002). Leafy vegetables in particular are a rich source of antioxidants such as beta-carotene and ascorbic acid (Arthey & Dennis, 1992; Negi & Roy, 2000). Also, the link between the consumption of these products and the lower risk of suffering certain chronic diseases (Ruowei, Serdula, Bland, Mokdad Bowman, & Nelson, 2000; Subar *et al.*, 1995) has been contributed to the consumer's attitude change during recent decades.

Minimally processed vegetables, also called ready-to-use, fresh-cut or pre-cut produce, are raw fruits and vegetables that have been washed, peeled, sliced, chopped or shredded prior to being packaged for consumption (Barry-Ryan & O'Beirne, 1998). They are also defined as those subjected to some processing techniques of lesser magnitude than canning or freezing but which, nevertheless, add value to the product before distribution and consumption (King & Bolin, 1989). The marketing of minimally processed fruits and vegetables is limited by a short shelf life and rapid deterioration of their components due to tissue damage as a result of processing, e.g. washing or cutting, and the microbial growth (Watada & Qui, 1999). In recent years, the use of physiologically active compounds (PAC) has attracted interest of consumers and the industry (minerals, probiotics, etc.) (Alzamora et al., 2005).

Titchenal and Dobbs (in press) point out some dark green leafy cabbage family vegetables and turnip greens as good calcium sources, and most leafy vegetables as potential calcium sources. The major source of calcium in the United States diet is dairy products, which supply 75% of the intake, and vegetables, fruits and grains which supply the rest (Allen, 1982).

The awareness of consumers on the benefits of calcium is relatively high. The calcium content in the diet is critical in most stages of life (Gras, Vidal, Betoret, Chiralt, & Fito, 2003). Dietary calcium raises concern for consumers and health specialists due to the number of processes it is involved in, the high amount present in the body, and the continuous research highlighting the benefits of an adequate intake. Nowadays, an increasing part of the products in the food industry are fortified, especially dairy products followed by beverages and snacks (Caceres, Garcia, & Selgas, 2006). The interest in calcium has intensified in recent years as

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a result of evidences linking osteoporosis, hypertension and cancer to calcium deficiency. While the cause of these diseases is multifactor and poorly understood, there is some evidence to support the hypothesis that increased calcium intake might reduce the risk of suffering from these diseases (Appel et al., 1997; Cumming et al., 1997). Also, the use of phosphorous-free sources of calcium, such as gluconate, citrate, lactate, acetate and carbonate calcium salts, can help to obtain a good balance of calcium and phosphorous in the diet (Cerklewski, 2005). To give consumers the opportunity to increase their calcium intake without resorting to supplementation, the industry has been encouraged to fortify food and beverages with calcium (Cerklewski, 2005). This opens new ways of supplementing calcium intake by increasing the calcium content in these commodities. For this reason, the use of natural sources of calcium as preservative with a nutritional fortification effect presents an advantage for the industry and for the consumer.

## Calcium sources to maintain the shelf life of fresh vegetables and fruits

Different calcium salts have been studied for decay prevention, sanitation and nutritional enrichment 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 (Brant, 2002). Other forms of calcium used in the food industry are calcium lactate, calcium chloride, calcium phosphate, calcium propionate and calcium gluconate, which are used more when the objective is the preservation and/or the enhancement of the product firmness (Alzamora *et al.*, 2005; Luna-Guzman & Barrett, 2000; Manganaris, Vasilakakis, Diamantidis, & Mignani, 2007). The selection of the appropriate source depends on several factors: bioavailability and solubility are the most significant, followed by flavour change and the interaction with food ingredients.

Calcium chloride has been widely used as preservative and firming agent in the fruits and vegetables industry for whole and fresh-cut commodities. Chardonnet, Charron, Sams, and Conway (2003) and Sams, Conway, Abbott, Lewis, and Ben-Shalom (1993) studied the effect of calcium chloride on fruit firmness and decay after the harvest of whole apples. Saftner, Bai, Abbott, and Lee's (2003) work was also focused on the firming effect of calcium chloride treatment on fresh-cut honeydew. Luna-Guzman and Barrett (2000) compared the effect of calcium chloride and calcium lactate dips in fresh-cut cantaloupe firmness, microbial load, respiration and sensorial evaluation. Other authors (Garcia, Herrera, & Morilla, 1996; Main, Morris, & Wehunt, 1986; Morris, Sistrunk, Sims, Main, & Wehunt, 1985; Rosen & Kader, 1989; Suutarinen, Anakainen, & Autio, 1998) used calcium chloride as firming agent for processed strawberries. Wills and Mahendra (1989) examined the effect of calcium chloride on fresh-cut peaches from a quality point of view, meanwhile Conway and Sams (1984) evaluated the safety of strawberries treated with calcium chloride. Other fruits and vegetables, in which the effect of calcium chloride was studied, showing significant improvement in the quality of the final product, are grapefruit (Baker, 1993), hot peppers (Mohammed, Wilson, & Gomes, 1991) and diced tomatoes (Floros, Ekanayake, Abide, & Nelson, 1992). The use of calcium chloride is associated with bitterness and off-flavours (Bolin & Huxsoll, 1989; Ohlsson, 1994), mainly due to the residual chlorine remaining on the surface of the product.

Calcium lactate, calcium propionate and calcium gluconate have shown some of the benefits of the use of calcium chloride, such as product firmness improvement, and avoid some of the disadvantages, such as bitterness and residual flavour (Yang & Lawsless, 2003). Also, the use of calcium salts other than calcium chloride could avoid the formation of carcinogenic compounds (chloramines and trihalomethanes) linked to the use of chlorine.

Manganaris *et al.* (2007) compared the effect of calcium lactate, calcium chloride and calcium propionate dipping in peaches. Calcium increased in tissues with no dependence on the source used. Calcium incorporation by impregnation with two calcium sources, calcium lactate and calcium gluconate, was studied in fresh-cut apple (Anino, Salvatori, & Alzamora, 2006).

Another source of calcium is the calcium-amino acid chelate formulations which had been patented as nutritionally functional chelates. Lester and Grusak (1999) showed that the use of calcium chelate doubled the shelf life of whole honeydew melon.

### Methods for calcium application on fresh-like minimally processed fruits and vegetables

Two main ways of application of the calcium in fresh fruits and vegetables have been reported: dipping—washing (I) and impregnation (II) processes.

(I) *Dipping treatments* are commonly used for fresh products, especially those more perishable, such as leafy vegetables. It usually consists of the soaking of the product, applying or not applying mechanical agitation, followed by the removal of excess washing solution. This treatment is gentler to the product than the impregnation techniques which can cause tissue damage and metabolic stress. The washing treatment is usually applied to the whole product or followed by other minimal procedures, including washing, peeling, cutting, shredding and/or slicing.

Dipping treatments favour the dispersion of the solution on the surface of the vegetable. This method has an extra benefit since the enzymes and substrates released from the injured cells during the minimal procedure are rinsed, avoiding oxidation reactions that could lead to browning and off-flavours (Soliva-Fortuny & Martin-Belloso, 2003). Different factors (pH, immersion time, temperature, and concentration) can affect product integrity.

The dipping time ranges from 1 to 5 min in most of the published work. Luna-Guzman, Cantwell, and Barrett (1999) used periods of 5 min for immersion of fresh-cut cantaloupe. Martin-Diana *et al.* (2005a) treated fresh-cut

lettuce and carrots for 1-5 min. Manganaris *et al.* (2007) used a time period of 5 min for immersion of whole peach fruits. Longer periods were used by Suutarinen *et al.* (1998) with whole strawberries, lasting for 15 min.

The effect of temperature has been shown to be of major importance in the results of dipping-washing treatment. Cool, room and warm temperatures (4, 20-25, 40-60 °C, respectively) have been used with different calcium solutions. Results showed that the use of warm temperature (40-60 °C) increased the beneficial effects of the treatment due to higher washing solution retention inside the product (Bartolome & Hoff, 1972; Garcia *et al.*, 1996; Rico *et al.*, 2007).

Luna-Guzman *et al.* (1999) compared different temperatures (20, 40 and 60 °C) for treating fresh-cut cantaloupes with calcium chloride. The use of 60 °C improved the beneficial action of the calcium solutions in comparison with 40 or 20 °C. Similar results were observed by Rico *et al.* (2007) who found that the use of higher temperatures increased the diffusion of calcium into carrot tissues and improved the quality, especially related to texture maintenance and browning reduction in comparison with lower temperatures.

The control of the pH in the washing treatments has been considered an important factor. Acid pH has been usually recommended (Wiley, 1994). However, Adams (1991) and Delaquis, Stewart, Toivonen, and Moyls (1999) reported that lowering the pH from 8 to 6 did not significantly improve the antimicrobial effect of cold or warm chlorinated water washing treatment. Thus, they considered that there was little justification for the additional step of pH adjustment of washing water with the aim of reducing microbial load on vegetables. In the case of calcium, most authors used the solution without any pH adjustment, which ranged from 5.5 to 6.5.

The concentrations of the calcium salts used as washing treatments are usually within a range of 0.5-3% (Luna-Guzman & Barrett, 2000; Luna-Guzman *et al.*, 1999; Main *et al.*, 1986; Manganaris *et al.*, 2007; Martin-Diana *et al.*, 2005b; Morris *et al.*, 1985).

(II) Impregnation: the objective of these techniques is the modification of the composition of food material

through partial water removal and impregnation with solutes, without affecting the material's integrity. The process driven forces can be osmotic gradient between the sample and solution, application of vacuum followed by atmospheric condition restoration, or both osmotic gradient and vacuum conditions (Fig. 1) (Zhao & Xie, 2004). Depending on the time (minutes to days) of the process and the magnitude of the vacuum (5–200 mbar) or solution concentration (20-75 °Bx) used, the product obtained might lack the characteristics of a minimally processed product and rather be considered a dehydrated product.

Under vacuum conditions, when the porous tissue is immersed in the solution, the air is extracted from the pores; when the atmospheric pressure is restored, the solution penetrates into the intracellular spaces by capillary and pressure gradients (Alzamora *et al.*, 2005; Fito, 1994). The partial substitution of the internal gas by the new liquid phase allows the reformulation of the food by the modification of the solid matrix, avoiding eventual stress due to long exposure to gradient solute concentration as in osmotic process (Fito & Chiralt, 2000; Mujica-Paz, Valdez-Fragoso, Lopez-Malo, Palou, & Welti-Chanes, 2003). Also, to obtain product stability, impregnation techniques have been used to enhance the textural properties and/or fortify the product (Fito *et al.*, 2001; Gras *et al.*, 2003).

Impregnation provides broad applications in fruits and vegetables processing. Some of the advantages are the development of re-formulated products by impregnation with different sources of calcium, also improving the final product quality (Fito *et al.*, 2001).

Gras *et al.* (2003) applied vacuum impregnation to eggplant, carrot and oyster mushroom obtaining texture improvements. Alzamora *et al.* (2005) and Anino *et al.* (2006) have done extensive work to date applying vacuum impregnation in fresh-cut apple. Calcium lactate and calcium gluconate were selected at 5% for 2-22 h. The final product obtained after the long periods used for the impregnation might not be defined as a minimally processed product. Calcium decreased the force needed to sample rupture



Fig. 1. Schematic diagram of the equipment used for vacuum impregnation experiments (Anino et al., 2006).

and fracture occurred over a period of deformation, as in ductile materials.

Fito *et al.* (2001) modelled the vacuum impregnation in oranges and eggplants and other fruits and vegetables using different calcium sources. The authors considered this method a good choice to produce fresh-cut fruits or vegetables enriched or fortified with PAC.

Coating is also a technique that can be used to enrich minimally processed fruits and vegetables with calcium. Han, Zhao, Leonard, and Traber (2004) working with strawberries and red raspberries used a chitosan-based coating formulated with calcium lactate and calcium gluconate. Adding calcium did not alter the positive effects of coating, which proved to extend the shelf life by decreasing the incidence of decay and weight loss, changes in colour, titratable acidity and pH.

Another technique that might have application for treating fruits and vegetables with calcium solutions is spraying over the product surface. Spraying or fluxing, however, has been little researched to date. Experiments with spray jets for low-pressure cleaning have been carried out by Kaye, Pickles, and Field (1995) and Rose (1997) examining the spray cleaning effect of selected nozzles on vegetable surfaces. However, no conclusions on the application could be derived from these studies.

## Effect of calcium treatments on the commodities: quality, safety and nutritional issues

Calcium is involved in maintaining the textural quality of produce since calcium ions form cross-links or bridges between free carboxyl groups of the pectin chains, resulting in strengthening of the cell wall (Garcia *et al.*, 1996). Calcium complexes to cell wall and middle lamella polygalacturonic acid residues, improving structural integrity (Van-Buren, 1979).

Manganaris *et al.* (2007) suggested 62.5 mM calcium chloride immersion treatment as a potential postharvest treatment for whole peaches, since increased tissue firmness reduced the susceptibility to physiological disorders and reduced the risk of salt-related injures. Saftner *et al.* (2003) found calcium propionate, calcium chloride and calcium chelate treatments more than doubled tissue calcium content and inhibited changes in honeydew chunks firmness. Calcium lactate showed a more lasting effect of firmness preservation than calcium chloride during the storage (Fig. 2) in fresh-cut cantaloupe (Luna-Guzman & Barrett, 2000).

Calcium lactate was applied by vacuum impregnation to eggplant, carrot and oyster mushroom (Gras *et al.*, 2003). Calcium impregnation occurred in the intercellular spaces of eggplant and oyster mushroom and xylem of carrot. Mechanical behaviour of eggplant and carrot was notably affected by calcium, with an increase in stiffness and fragility. Eggplant and oyster mushroom appeared to be highly suitable for obtaining fortified products by using small concentrations of calcium in the impregnation solution.

Iceberg lettuce is highly appreciated by the consumer because of its characteristic crispy texture. Crispness, evaluated using a texturometer, of samples treated with calcium lactate was significantly higher than crispness of samples washed with chlorine (Martin-Diana *et al.*, 2006; Rico *et al.*, 2007). However, when the washing treatment with calcium lactate was at 50 °C (heat-shock), the texture results were even better than at 20 °C, significantly retarding the softening process. Microstructural analysis (Cryo-SEM



**Fig. 2.** Firmness (Nm) of fresh-cut cantaloupe from experiment 1, dipped in water or 2.5% calcium lactate for 1 min at 25 or 60 °C, and stored under air at 5 °C and 95% RH. For each evaluation day, columns with same letter are not significantly different (p < 0.05). N = 14, vertical bars indicate  $2 \times$ S.E. (Luna-Guzman & Barrett, 2000).

micrographs) showed a loss of turgor (shrinkage) of the tissue cells in the lettuce samples washed with chlorine, an effect less evident when using calcium lactate (Fig. 3). The combined treatment of calcium lactate and warm heating also showed more effectiveness in maintaining the turgor of cortex tissue cells and reducing the extent of lignification at cutting-edge areas in carrots (Fig. 4).

Complementing calcium treatments with mild heating  $(\sim 40-70 \ ^{\circ}C, \sim 5 \ ^{min}$  or less) is an interesting alternative. Time and temperature of the heat treatment must not compromise the fresh-like characteristics of the commodities. Heat can be applied with the calcium treatment by



**Fig. 3.** Cryo-scanning electron micrographs of fresh-cut lettuce washed with 120 mg/L chlorine, 15 g/L calcium lactate at room temperature (25 °C) and at 50 °C (heat-shock) after 1 day of storage at 4 °C (Martin-Diana *et al.*, 2006).

increasing the temperature of the calcium solution in contact with the fruit or vegetable, or at a different stage. Mild heat has been reported to activate the pectin methylesterase (PME) (Bartolome & Hoff, 1972). Endogenous and added calcium can make plant tissue firmer by binding to the pectin carboxyl groups that are generated through the action of PME (Stanley, Bourne, Stone, & Wismer, 1995).

Calcium can also help to keep longer the fresh-like appearance of minimally processed fruits and vegetables by controlling the development of browning. Control of the flesh browning has been observed in fruits in different studies, e.g. in peaches (Manganaris *et al.*, 2007) and pineapple (Hewajulige, Wilson-Wijeratnam, Wijesundera, & Abeysekere, 2003). Saftner *et al.* (2003) found treatments with calcium inhibited colour changes and development of tissue translucency in honeydew chunks.

Another benefit derived from the use of calcium treatments is the incorporation of significant quantities of calcium to the fruit or vegetable matrix. This is also one of the objectives and driving forces promoting the research on these types of treatments. Anino et al. (2006) analysed the ability of fresh-cut apple matrix for calcium incorporation by impregnation techniques using calcium lactate and calcium gluconate (Fig. 5). These authors explored the possibility of obtaining a product fortified with calcium. The calcium amount incorporated into the apple matrix would satisfy about 23-63% (200 g of fruit) of the adequate intake. Sensory analysis also showed that the use of calcium improved the textural characteristic of the product during storage. Calcium content of fresh-cut lettuce significantly increased when treated with calcium lactate compared with chlorine treatments (Table 1). A combination of calcium lactate and heat-shock further increased the calcium content of the samples (Rico et al., 2007). Alzamora et al. (2005) regard impregnation as a feasible technology for making fruit and vegetable matrices suitable for fortified food development. Also, these authors emphasise on the high produce-specificity of this process and point to the need for further research in this area.

Sensory analyses of minimally processed products treated with calcium have been reported in some cases, depending on the source of calcium used, bitterness as a side effect of the treatment. Luna-Guzman and Barrett (2000) imparted calcium chloride (~173 mM) in fresh-cut musk melon finding undesirable bitterness, which was avoided when using calcium lactate at the same concentration (Table 2). Saftner et al. (2003) found that sensory evaluations with calcium propionate and calcium chelate (40 mM) were taste free and did not impart a lip feel. Meanwhile, calcium chloride solutions had slightly salty taste and lip feel. Martin-Diana et al. (2005a) did not find significant differences in sensory attributes (browning, texture or off-flavours) between samples treated with calcium lactate and calcium chloride. However, when warm temperatures were used, a significant improvement in sensory attributes (browning and texture) was observed (Fig. 6).



Fig. 4. Lignification observations in Cryo-SEM micrographs of sliced carrot tissues in the 10 peeler-wounded area (orientation 1) at day 10 in samples treated with chlorine (120 mg/L), 11 calcium lactate (15 g/L) at 25 °C and calcium lactate at 50 °C (Rico *et al.*, 2007).

### Importance of calcium in the industry: cost and benefits

Today's consumer has, on average, a busier lifestyle than in previous decades. Income, as measured by Gross National Product (GNP), has been rising over the past two decades, throughout the EU and the US. Both unemployment



**Fig. 5**. Effect of calcium incorporation on the rupture force (*F*<sub>rup</sub>) of apples subjected to impregnation in the isotonic glucose solution with and without Ca<sup>2+</sup>: with calcium (grey); without calcium (white); (A) atmospheric impregnation and (B) vacuum impregnation (Anino *et al.*, 2006).

and interest rates have generally been declining throughout the 1990s, substantially increasing consumer's disposable income. This means that consumers have the ability to spend more on higher value or added-value foods, even though the current percentage spent on food from the total household budget has fallen over the past 30 years. This reflects the fact that people do not consume more food as their income rises, but rather tend to purchase higher value food. Fishbein (2004) considers it important to diversify the sources of calcium due to dynamic dietary changes, which transcend age, gender, ethnicity, geographic, and economic status, and creates the necessity for the consumer to have at their disposal multiple sources of calcium (dairy, non-dairy, fortified foods and supplemental).

The existing methods for application of the calcium treatment in fresh fruits and vegetables have already been studied on a significant number of commodities, showing positive results for the feasibility of this type of products. The fact that the calcium treatment is applied to keep the quality and properties of the product and not only for calcium enrichment purposes is an advantage for the implementation of this type of products. Also, the perspective of growth for the enriched food market makes this a very interesting opportunity from the industry's point of view (Alzamora *et al.*, 2005).

Table 1. Calcium content in fresh-cut lettuce after being treated
with 120 ppm chlorine at 25 °C and 1.5% calcium lactate at two
temperatures (25 and 50 °C) (Rico et al., 2007)

Treatments	Calcium (mg/100 g)		
Calcium lactate			
25 °C	0.162 <sup>a</sup>		
50 °C	0.191 <sup>a</sup>		
Chlorine	0.122 <sup>b</sup>		
Values designated by the same latter are not significantly different			

Values designated by the same letter are not significantly different (p > 0.05).

Table 2. Mean sensory scores of fresh-cut cantaloupe dipped in calcium chloride or calcium lactate solutions (Luna-Guzman & Barrett, 2000)

Treatment	Sensory attribute				
	Bitterness	Flavour	Hardness	Moisture content	
Just-cut 1.0% Calcium chloride 2.5% Calcium chloride 1.0% Calcium lactate 2.5% Calcium lactate	2.6 <sup>c</sup> 4.8 <sup>a,b</sup> 5.8 <sup>a</sup> 2.7 <sup>c</sup> 3.4 <sup>b,c</sup>	$\begin{array}{c} 4.6^{a,b} \\ 3.1^{b} \\ 4.2^{a,b} \\ 5.6^{a} \\ 4.7^{a,b} \end{array}$	3.4 <sup>c</sup> 5.1 <sup>a,b</sup> 4.8 <sup>a,b,c</sup> 4.7 <sup>b,c</sup> 6.0 <sup>a</sup>	5.9 <sup>a,b</sup> 6.4 <sup>a</sup> 4.8 <sup>b,c</sup> 4.5 <sup>c</sup> 4.5 <sup>b,c</sup>	
Under each attribute, means followed by same letter are not significantly different ( $p < 0.05$ ). For each attribute, a higher number represents higher intensity on a 0–10 scale.					

Some of the purified calcium sources might result to be expensive, but the fact that the treatment is also adding value to the product is an advantage to balance the cost/ benefit rate. Other alternatives are calcium chelates, al-though more expensive than other salts (Saftner *et al.*, 2003). Calcium chelates are not corrosive to processing equipment and are more likely to penetrate deeply in plant tissues; however, problems associated with their low solubility have been reported.

#### **Future research needs**

From the point of view of the possibilities in the fortified food market, there is a wide range of opportunities to develop new products using calcium treatments, due to the high and increasing demand of fresh and healthy food. When using impregnation techniques, the behaviour of the vegetable or fruit matrices is highly produce-specific, increasing the amount of research needed. Also, some issues regarding impregnation techniques, especially vacuum impregnation need further



Fig. 6. Texture evaluation by sensory panel of salad-cut lettuce treated with 1.5% calcium lactate and different treatment temperatures and chlorine (CHL) control during storage for 10 days (Martin-Diana *et al.*, 2005b).

research. Differences in density of the solution and the commodity (usually lower) makes keeping in contact sample and solution more difficult. Approaches to solve the problem are the use of stirring, compressing or sinking rods. These methods can cause damage to the product and increase the cost of the process. More research might be necessary in this area to obtain acceptable levels of quality for fresh-like minimally processed products.

An important aspect needing further research is bioavailability of the calcium incorporated to the product. The bioavailability depends on a large number of factors, such as commodity pH or fibre content, and needs to be studied for each particular case when obtaining a fortified product. Also, the antimicrobial effect of the treatments based on calcium salts on fruits and vegetables has been relatively little studied, compared with the amount of reports showing the effects of these treatments on the quality.

The markets for convenient, fresh and fortified foods are continuously growing. This extensive knowledge gathered and the further research proposed can be exploited for the development of novel fresh-like minimally processed fruits and vegetables enriched in calcium that can take advantage of these three market niches.

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