Decontamination by ultrasound application in fresh fruits and vegetables

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ABSTRACT

Changes in consumer eating habits, health concerns, and convenient and practical foods have led to an increased demand for fruit and vegetable products. Food safety is essential considering that there are reports of outbreaks involving the consumption of fruits and vegetables contaminated with pathogens. Washing associated with sanitizer procedure is considered as a critical step to satisfy hygienic and sanitary requirements and maintain the sensory and nutritional characteristics of fruits and vegetables. Chemical compounds are widely applied to clean and sanitize fresh fruits and vegetables, and some of these chemicals, such as the inorganic chlorine compounds, produce by-products that are dangerous to human health. The use of ultrasound is a technology that is gaining ground in the food industry. Ultrasound is a form of energy generated by sound waves at frequencies that are too high to be detected by the human ear. In ultrasound, the removal of dirt and food residues from surfaces and the inactivation of microorganisms occur as a consequence of cavitation, which is the formation, growth and collapse of bubbles that generate a localized mechanical and chemical energy. There are indications that this technology can be used in the food industry, alone or associated with chemical sanitizers. In this paper, we discuss the principles, mechanisms and effects of ultrasound on fruits and vegetables as a sanitization technology.

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1. Introduction

The increased consumption of fruits and vegetables has led to an increased concern about the high quality of nutritional, sensory and microbiological aspects of these foods (Chemat, Zill-e-huma, & Khan, 2011; Fava et al., 2011; Martínéz-Sánchez, Allende, Bennett, Ferreres, & Gil, 2006; Plaza et al., 2011; Vandekinderen et al., 2009). Safety and quality are important terms in the food industry. Recently, there has been great interest in new methods to ensure the preservation of food without the use of additives and maintenance of the food’s nutritional value and sensory aspects with low-energy consumption, at a competitive cost, and using environment-friendly products with a high degree of safety.

To minimize the risk of microbial contamination, potential sources should be identified, and specific preventive measures should be implemented.

To prepare ready-to-eat fruits and vegetables, the produce must be clean, safe, and free of soil residue, insects, metals and weeds. Fruits and vegetables should be carefully cleaned before processing because fresh-cut produce is processed from material grown in contact with soil and without any antimicrobial treatments.

The inclusion of a washing and sanitization steps before cutting and during processing effectively reduces the risk of pathogen and residue contamination on fruits and vegetables from harvest and handling conditions. During sanitization, fruits and vegetables are subjected to an effective treatment to destroy or reduce the number of pathogenic microorganisms, without affecting the quality, safety or product. The effectiveness of washing to remove soil impurities and microbial contaminations is related to numerous factors, such as raw material spoilage, the duration of washing, the water temperature, the method of washing (dipping, rinsing, or dipping/blowing), the type and concentration of the sanitizer and the type of fresh-cut fruit or vegetable. The selection of a sanitizer for fresh-cut fruit or vegetable. The selection of a sanitizer for fresh-cut fruit or vegetable. The selection of a sanitizer for fresh-cut fruit or vegetable. The selection of a sanitizer for fresh-cut fruit or vegetable.

Currently, commercial operations use wash treatments with antimicrobials as the only step to reduce microbial populations on fresh fruits and vegetables. Sanitizing with chemical solutions, which might include chlorine, primarily influences the safety and preservation of these products (Allende, Selma, Lópeze-Gálvez, Vilaescusa, & Gil, 2008; Artés & Allende, 2005; Pérez-Gregorio, González-Barreiro, Rial-Otero, & Simal-Gándara, 2011; Ruiz-Cruz, Félix, Díaz-Cinco, Ibáñez-Osuna, & González-Aguilar, 2007; Vandekinderen et al., 2009).

However, chlorine-based compounds are corrosive and cause skin and respiratory tract irritations to food handlers (Alvaro et al., 2009; Fava et al., 2011; Pérez-Gregorio et al., 2011; Vandekinderen et al., 2009). Water hyperchlorination generates high concentrations of trihalomethanes (trichloromethane, bromodichloromethane, dibromochloromethane, tribromomethane and chloroform) and other disinfectant by-products that are potentially carcinogenic (Fava et al., 2011; Rico, Martín-Diana, Barat, & Barry-Ryan, 2007; Ruiz-Cruz, Félix, et al., 2007; Selma, Ibáñez, Allende, Cantwell, & Suslow, 2008; Pérez-Gregorio et al., 2011; Vandekinderen et al., 2009). Chlorinated compounds have been the focus of environmental concerns, and some environmental groups have suggested terminating the use of these products worldwide. In European countries, such as the Netherlands, Sweden, Germany and Belgium, the use of chlorine on minimally processed vegetable products is prohibited (Pérez-Gregorio et al., 2011; Rico et al., 2007). There have also been studies on emerging pathogens that can be more tolerant to chlorinated compounds, which raises further concerns about the effectiveness and use of chlorine in the minimally processed food industry (Allende et al., 2008; Alvaro et al., 2009).

In recent years, emerging technologies, such as high pressure, pulsed electric field, electrolysed water, irradiation, ozone and ultrasound treatments, have been widely studied for application in food industry. Studies evaluating the application of ultrasound in food science and technology have been expanded due to its promising effects in food processing and preservation (Adekunle, Tiwari, Cullen, et al., 2010; Adekunle, Tiwari, Scannell, Cullen, & O’Donnell, 2010; Arzeni et al., 2012; Birmma, Sfika, & Vantakaris, 2013; Cameron, McMaster, & Britz, 2008; Cao et al. 2010; Chemat & Hoaraou, 2004; Fava et al., 2011; Gabriel, 2012; Knorr, Zenker, Heinz, & Lee, 2004; Motihile, Zhang, Nsor-Atindana, & Wang, 2011; Nascimento et al. 2008; Patil, Bourke, Kelly, Frías, & Cullen, 2009; São José & Vanetti, 2012; Seymour, Burfoot, Smith, Cox, & Lockwood, 2002; Soria & Villamiel, 2010; Tiwari, Patras, Brunton, Cullen, & O’Donnell, 2010). In this context, this review focuses on application of ultrasound on fruit and vegetables.

2. History of ultrasound

Ultrasound has been used with different objectives, such as communication with animals, the location of flaws in concrete buildings, chemical synthesis and the diagnosis or treatment of diseases (Dolatowski, Stadnik, & Stasiak, 2007; Mason, 2003).

The discovery of ultrasound came with Pierre Curie in 1880 in his studies of the piezoelectric effect (Shankar & Pagel, 2011). In 1894, Thornycroft and Barnaby observed that vibrations were generated in the propulsion of missiles launched by a destroyer, which produced implosion bubbles and/or cavities in the water, a phenomenon known as cavitation (Martines, Davolos, & Júnior, 2000).

Before World War II, applications of ultrasound were being developed for a range of technologies, including surface cleaning procedures. In the 1960s, ultrasound technology was well established and used in cleaning and plastic welding (Mason, 2003). Despite the diverse applications and great development, ultrasound science is still considered a recent technology. Sixty years ago, low-intensity ultrasound methods were used to characterize foods, but it is only recently that the potential of the method has been evaluated (Dolatowski et al., 2007; Ulusoy, Colak, & Hampikyan, 2007).

3. Principles of ultrasound

Ultrasound is a form of energy generated by sound waves at frequencies that are too high to be detected by the human ear. These waves participate in a region of the sound spectrum that is divided into three main parts: infrasound (ν < 16 Hz), band sound (16 Hz < ν < 16 kHz) and ultrasound (ν > 16 kHz). The ultrasound band is also divided into low frequency (16 kHz < ν < 1 MHz) and
high frequency (ν > 1 MHz) bands. High-frequency ultrasound bands are typically used for medical and industrial imaging purposes (Butz & Tauscher, 2002; Cárcel, García-Pérez, Benedito, & Mulet, 2012; Demirdöven & Baysal, 2009; Dolatowski et al., 2007; Kentish & Ashokkumar, 2011; Leadley & Williams, 2006; Sala, Raso, Pagán, & Condón, 1998).

Ultrasound can be classified by the energy amount used: Sound power (W), sound intensity (W m⁻²) and sound energy density (Ws m⁻³) are the most significant measures used for categorizing ultrasound applications (Knorr et al. 2004). Notably, the applications of ultrasound are classified into low-energy (low power or low intensity) and high-energy (high power or high intensity) ultrasound (Kentish & Ashokkumar, 2011).

The low-energy group includes frequencies higher than 1 MHz and these ultrasonic waves cause no physical or chemical alterations in the properties of the material through which the wave passes. Low-energy ultrasound produces non-destructive effects and provides information about the physicochemical properties of foods, such as their composition, structure and physical state (Knorr et al. 2004; Leadley & Williams, 2006). High-power ultrasound includes frequencies between 18 kHz and 100 kHz and can produce physical, mechanical or chemical effects, such as the physical disruption and acceleration of certain chemical reactions (Golmohamadi, Möller, Powers, & Nindo, 2013; Jayasooriya, Bhandari, Torley, & D’Arcy, 2004; Mason, 2003; São José & Vanetti, 2012).

High-intensity ultrasound has been used for many years to produce emulsions, disrupt cells and disperse aggregated materials. Diverse areas have been recognized for potential future development, for example, the adjustment and control of crystallization processes; the measurement of texture, viscosity, and content of solids and fluids; the degassing of liquid foods; the inactivation of enzymes; enhanced drying, filtration and induction of oxidation reactions and the inactivation of microorganisms (Knorr et al. 2004; Suslick et al., 1999). The advantage using ultrasound is a consequence of the various effects on the medium through which it is transmitted (Dolatowski et al., 2007).

Ultrasound is a technology used in the electronic industry to decontaminate surfaces and has been recommended for use in the food industry (Cao et al., 2010; Chen & Zhu, 2011; Dolatowski et al., 2007; Kentish & Ashokkumar, 2011; Nascimento et al., 2008; Patist & Bates, 2008). The physical effect can be the main action that is involved on microbial cell death. However, is known that the production of reactive compounds as peroxide hydrogen can be promoted the microbial disruption either (Cao, Lewis, Ashokkumar, & Hemar, 2014).

In food processing, high-intensity ultrasound at low frequencies, from 20 to 100 kHz, is useful in inactivating microorganisms (Piyasena, Mohareb, & McKellar, 2003). When propagated through a biological structure, ultrasound promotes the compression and expansion of the medium particles, resulting in the production of a high amount of energy (Butz & Tauscher, 2002; Dolatowski et al., 2007; Kentish & Ashokkumar, 2011; Piyasena et al., 2003). The inactivation of microorganisms is a consequence of cavitation. Cavitation (Fig. 1) is the formation, growth and collapse of bubbles that generate a localized mechanical and chemical energy (Gogate & Kabadi, 2009; Rastogi, 2011).

When ultrasound waves pass through a liquid medium, mechanical vibrations and acoustic streaming are generated within the liquid. Liquid media can contain dissolved gas, and they can expand, promoting bubble formation and collapse through ultrasound (Kentish & Ashokkumar, 2011). In addition, the violent collapse events that occur during transient and repetitive transient cavitation can generate enormous localized temperatures (5500 K) and pressures (1000 MPa), or so-called hot spots, and release free radicals as a result of the dissociation of vapour trapped in the bubbles. The occurrence of hot spots is limited in duration (Gabriel, 2012; Knorr et al., 2004; Rastogi, 2011; Riener, Noci, Cronin, Morgan, & Lyng, 2009). It is important to discuss the hot spots because if the treatment conditions are not well adjusted rather than to achieve a safe decontamination without damage to the plant, can be obtain rupture plant cell which culminates in loss of quality original of the sanitized product (São José & Vanetti, 2012).

Increasing temperature and pressure changes can induce many chemical changes within both the vapour phase inside the cavitation bubble and the surrounding fluid (Kentish & Ashokkumar, 2011; Piyasena et al., 2003). This phenomenon also generates turbulence-located microcirculation. The implosions are asymmetric when produced near a solid surface, creating a microjet that hits the solid (Cárcel et al., 2012), and these changes contribute to the mechanism used for cleaning surfaces (Bermúdez-Aguirre, Mobbs, & Barbosa-Cánovas, 2011; Gogate & Kabadi, 2009;
The amount of energy released by cavitation depends on the kinetics of the growth and collapse of the bubbles. Probably, these microjets contributed to remove the adhered bacteria cells on fruit and vegetable surfaces. This fact can occur because the higher population of bacteria and the production of exopolysaccharides became a barrier from sanitizers solutions (São José & Vanetti, 2012).

The ultrasound alone can provide powerful disinfection, but its use for large-scale microbiological decontamination should be further evaluated, and in combination with other technologies, it could even provide excellent results (Joyce, Phull, Lorimer, & Mason, 2003). The application of ultrasound individually or in combination with peracetic acid was effective in removing Salmonella from the surface of cherry tomatoes (São José & Vanetti, 2012). The use of ultrasound has also been implemented in the disaggregation of biofilms and the inactivation of microorganisms, which can help in the preservation of foods. Fig. 2 show that ultrasound can remove adhered cells of Salmonella Typhimurium on cherry tomato surface.

Ultrasonic waves generate and distribute cavitation implosions in liquid medium. The energy released during cavitation allows surfaces that are generally difficult to access using other cleaning methods to be reached (Gao et al., 2014). It is known that chemical sanitizers are occasionally unsuccessful against microorganisms.
that are present on biofilms or tightly adhered on surfaces. In this case, physical methods might be more helpful. Ultrasound is a consistent and uniform method to remove contaminants. The use of ultrasound in the food industry is promising because a microbial biofilm can occur on the surfaces of fruit and vegetables.

Ultrasound offers advantages in terms of cost, productivity and selectivity, with better processing time, improved quality, and reduced chemical damage and physical risks. Currently, the application of this technology has attracted attention for its role in environmental sustainability without causing damage, and it is therefore applicable to the concept of green technology (Chemat et al., 2011).

3.1. Generation of power ultrasound

A power ultrasound system consists of three basic parts: an electrical power generator, a transducer and a coupler/emitter (Mothibe et al., 2011).

The electrical generator is the source of energy for the ultrasound system. Most generators allow the power to be set only directly through voltage at current settings. These generators are typically designed for industrial cleaning, therapeutic and disinfecting applications (Leadley & Williams, 2006).

Transducers convert electrical energy through mechanical vibrations at ultrasound frequencies. The main types of transducers include liquid-driven, magnetostrictive and piezoelectric transducers (Bermudez-Aguirre et al., 2011). Liquid-driven transducers conduct liquid across a thin metal blade, causing it to vibrate at ultrasonic frequencies. With this transducer, rapidly alternating pressure and cavitation effects occur in the liquid, which generates a high degree of mixing. This device is commonly used for the mixing and homogenization process. Magnetostrictive transducers are electro-mechanical devices that use magnetostriction, an effect derived from some ferromagnetic materials, which modifies dimension in response to the application of a magnetic field. The frequency range is typically restricted to below 100 kHz, and 60% of the electrical energy was found to be re-allocated to acoustic energy because of the energy loss due to heat (Leadley & Williams, 2006). Piezoelectric transducers are electrostrictive devices that use ceramic materials. Piezoceramic elements are the most common and efficient (80–95% transfer to acoustic energy) transducers (Leadley & Williams, 2006). Piezoelectric transducers are not able to resist long exposures to high temperatures, for example, they cannot tolerate a temperature of 85 °C. These transducers are also used in diagnosis ultrasound equipment (Rastogi, 2011).

The coupler or emitter, which is also called the reactor or ultrasonic cell, is responsible for emitting the ultrasound waves from the transducer into the medium. The most important forms of couplers are bath couplers and horns/probes (Leadley & Williams, 2006).

3.2. Available equipment

Many ultrasonic systems are available that primarily differ in the power generator design, the transducer type used and the reactor to which it is coupled. The ultrasonic transducer (design, format and method) is important to define its effectiveness and efficiency, which is an important variable. Any differences between laboratory and pilot plant designs and applications of ultrasound can produce different results (Bermudez-Aguirre et al., 2011).

Two different types of ultrasound equipment are typically used in laboratories: ultrasonic cleaning baths and ultrasonic probes (Cárcel et al., 2012; Chemat et al., 2011; Mothibe et al., 2011).

Ultrasonic cleaning baths are commonly used for solid dispersion, degassing solutions or cleaning materials (Fig. 3). They are less used for chemical reactions, although they are inexpensive and easy to use. Different capacities of this equipment are available. Typical tank sizes range from 10 to 2500 L (Awad, 2011). Ultrasonic baths have transducers located on the walls and/or base of the tank and the ultrasonic energy is delivered directly to the liquid. Usually, this type of ultrasound operates at approximately 40 kHz and produces high intensities at fixed levels. The depth of the liquid is significant for maintaining these intensities and should not be less than half the wavelength of the ultrasound in the liquid. In the ultrasonic bath, the frequencies are released through a mechanism that produces a more uniform cavitation field and reduces stationary wave zones (Cárcel et al., 2012; Chemat et al., 2011).

In the case of ultrasonic probes or horn systems, a rod-shaped metal horn is used to amplify and conduct the high power acoustic vibration in the medium. The horns or probes are frequently half or multiple wavelengths in length. The gain in amplitude depends upon the shape and variation in the diameter of the horn between the driven face and the emitting face. This system is considered more powerful because the ultrasonic intensity is released from a small surface at the extreme of the probe, and the probe can be immersed into reaction flasks. However, this system can only be used on samples with small volumes, and careful attention must be paid to the rapidly increasing temperature of the sample (Cárcel et al., 2012; Chemat et al., 2011).

However, laboratory studies can be adapted to allow large-scale application using bath and flow ultrasound systems. Many ultrasound systems have been designed for use in food processing. For example, an ultrasound bath can be used as a reactor for chicken shackles to avoid cross-contamination. Ultrasonic flow systems are used for liquid food, which is passed through a vibrating tube. The sound energy generated for transducers bound to the outside of the tube is transferred directly into the flowing liquid (Mason, Riera, Vercet, & Lopez-Buesa, 2005).

Ultrasound processing is becoming an important food-processing technology with a large commercial scale-up and sufficient returns on capital investments. Depending on the application, the energy required per litre of material treated (often defined as kWh/L) is similar to any other equipment in the industry, such as that required for homogenization. Moreover, the equipment is easy to use because of the absence of moving parts (Patist & Bates, 2008).

Riera et al. (2004) discuss about use of airborne ultrasound and conclude that applications do exist provided that source must be very powerful and efficient. This may be an option for fruits and vegetables that are highly perishable and should not be immersed in solutions.

Lin, Xu, and Hu (2011) proposed a new type of high power composite ultrasonic transducer that can be used in ultrasonic cleaning due the necessity of high power ultrasonic radiators with large radiating surfaces and powers. This transducer has a longitudinal sandwich transducer, two metal end masses, and a metal hollow cylinder that can radiate high power ultrasonic wave in the longitudinal and the radial directions.

3.3. Factors affect ultrasound application in fresh fruit and vegetables

It is known that the extent of physical and chemical effects of ultrasound treatment are strongly associated with the amplitude of ultrasonic waves, exposure time, volume processed, food composition and treatment temperature (Ananta, Voigt, Zenker, Heinz, & Knorr, 2005; Gómez-López, Orosolán, Martínez-Vépe, & Tapia, 2010; Koda, Miyamoto, Toma, Matsuoka, & Maebayashi, 2009;
Piyasena et al., 2003; Salleh-Mack & Roberts, 2007; Su, Zivanovic, & D’Souza, 2010; Tiwari et al., 2010).

Depending on the frequency used and sound wave amplitude applied, a number of physical, chemical and biochemical effects can be observed with a variety of applications (Knorr et al., 2004). The frequency selected for the application affects the cavitation activity. The bubble size is inversely proportional to the frequency. Lower frequencies produce larger bubbles, and a higher energy is liberated. Ultrasonic waves with large amplitudes facilitate the displacement of molecules and collapse pressure (Povey & Mason, 1998).

The effects produced by high power ultrasound when passed through a medium are diverse, and their relative importance depends on the characteristics of the food. The viscosity of the medium can affect cavitation. In highly viscous foods, ultrasound diffusion is difficult, thus reducing the frequency at which cavitation occurs. Lower frequencies are more effective in this case, as the ultrasound waves can pass through the viscous product (Bermúdez-Aguirre et al., 2011).

The pH of the medium or food is another condition that can affect ultrasound efficiency. At a lower pH, inactivation rate of microorganisms is increased. Salleh-Mack and Roberts (2007) studied the effect of pH on the inactivation of *Escherichia coli* and observed a significant effect on the ultrasound inactivation (24 kHz/9 min), where a lower pH caused a major reduction in the count of microorganisms. The same authors evaluated the presence of soluble solids at higher concentrations and observed that longer ultrasound treatments were necessary to obtain the same reduction as observed in the treatment control. Then, the concentration of soluble solids in the medium had a protective effect.

Gera and Doores (2011) studied the kinetics and mechanism of bacterial inactivation and the sonoprotective effect of milk components during the application of ultrasound (24 kHz at 30–35 °C) and observed that this food matrix exerts a protective effect on bacteria. Among the milk components tested, the presence of lactose showed higher D values, suggesting that lactose exerted a protective effect on bacteria. These results suggest that when fresh-cut products are sanitized with ultrasound, some of the compounds of these foods, such as exudates, can be released into the medium and exert a protective effect on bacteria. Moreover, solid particles can reduce the efficiency of cavitation.

The optimum temperature should be determined to enhance the formation of cavitation. Salleh-Mack and Roberts (2007) observed that ultrasound (24 kHz/9 min) increased the sensitivity of *E. coli* to thermal inactivation at increased temperatures. The temperature increase is not appropriate for minimally processed fruits and vegetables. Higher temperatures generated structural and physiological alterations on vegetable tissue, whereas lower temperatures reduced the respiration rate of fresh produce and minimised the loss of texture and other factors related to quality.

### 4. Microbial inactivation

The contamination of farm-raised vegetables and fruits might occur as a result of inadequate water irrigation, fertilizers, harvesting, packaging, transport or handling at the point of sale. *Salmonella* spp., *Listeria monocytogenes*, *E. coli* O157: H7, *Aeromonas* and the hepatitis A virus are some of the microorganisms associated with contamination in foods, such as fruits and vegetables, which indicates the importance of controlling these pathogens as a public health concern (Abadias, Usall, Anguera, Solsona, & Viñas, 2008; Lapidot, Romling, & Yaron, 2006). The complete removal and/or inactivation of pathogens from the surfaces of fresh products are a challenge for the food industry (São José & Vanetti, 2012). Thereby, new strategies to decontamination fruits and vegetable have to be studied.

Several researches have been conducted to investigate the mechanism of ultrasonic disruption in biological systems, and cavitation effects are considered to be a major factor. On Table 1 are exposed some causes and effects of ultrasound on microbial cell.

The mechanisms involved in cellular disruption are multifactorial and might include shear forces generated during the movement of bubbles or sudden localized temperature and pressure changes caused by bubble collapse (Salleh-Mack & Roberts, 2007; Ulusoy et al., 2007).

<table>
<thead>
<tr>
<th>Causes</th>
<th>Effect</th>
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<tbody>
<tr>
<td>Cavitation</td>
<td>Cell wall structures are disrupted</td>
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<td></td>
<td>Thinning of cells</td>
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<td>Pore formation, cell membrane disruption,</td>
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<td>and cell breakage</td>
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<td>Free radical production</td>
<td>DNA injures which produce breakages and</td>
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<td>fragmentation</td>
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There are two types of cavitation, which have been observed to have different effects. The first is stable cavitation, which occurs due to oscillations of ultrasound waves, inducing the formation of tiny bubbles in the liquid. The collapse of bubbles does not occur in this process, but requires thousands of cycles of oscillating ultrasound waves to increase the bubble size (Cárcel et al., 2012). As the ultrasonic wave passes through the medium, the bubbles vibrate, and cavitation sets up currents in the fluid adjacent to the vibrating bubbles; the currents in turn exert a twisting and rotational motion on near areas. Small bubbles travel through the sonic field and create microcurrents. This effect is called microstreaming. Nyborg (1982) describes microstreaming as a phenomenon that participates in a class of acoustical occurrences, such as movements, forces, strains and stresses, in which the geometrical scale is small. This manifestation provides extensive forces, which rub against the surface of cells and disrupt the cell membrane to induce the breakdown of the cell wall. Nearby vibrating gas bubbles, rotational forces and stresses affect intracellular organelles (Leadley & Williams, 2006). The second type is transient cavitation, where bubbles increase in size within a few oscillatory cycles and then collapse, causing cellular stress. Pressure can disrupt cell wall structures, leading to cell leakage and disruption. Occasional collapse, causing cellular stress. Pressure can disrupt cell wall breakdown. Nearby vibrating gas bubbles, rotational forces and stresses affect intracellular organelles (Leadley & Williams, 2006). Other proposed mechanisms of ultrasound on microbial inactivation occur by free radical formation. These compounds can help facilitate different applications of ultrasound. During bubble collapse, energy is released, which can contribute to the sonolysis of water. In this reaction, OH⁻, H⁺ and hydrogen peroxide are generated, which have important bactericidal effects. DNA is the primary target of these free radicals in the bacterial cell, which produce breakages and fragmentation along the extension of the DNA (Bermúdez-Aguirre et al., 2011).

Stanley, Golden, Williams, and Weiss (2004) observed that an increased hydrogen peroxide concentration induces a strong oxidation of lipid membranes, a disturbance in the enzymatic activity of protein complexes responsible for ATP production, and the maintenance of solute concentration in the cell interior, affecting the genetic material. Disruption cell facilitates input of other sanitizers as chlorine compounds that accelerates cell death.

The performance of ultrasound treatment is affected by the form, type or diameter of the microorganisms. It is important to know what the target microorganism to choose the best treatment conditions that will guarantee the method efficiency. Larger cells have a larger surface area and are more sensitive than smaller ones (Ananta et al., 2005; Piyasena et al., 2003). Gram-positive bacteria are recognised as more resistant than gram-negative bacteria because they have thicker cell walls that protect against the effects of ultrasound. According to the literature, the differences in the sensitivity of the cells might also be due to a more tightly adherent layer of peptidoglycan in gram-positive cells (Ananta et al., 2005; Piyasena et al., 2003; Ulusoy et al., 2007). Ultrasonic inactivation has currently been associated with damage to the cell wall and cell wall structures, which is supported by the fact that some bacteria are more resistant to cavitation than others under the same treatment conditions. The thinning of cell membranes together with heating and free radical production is essential for inactivation (Knorr et al., 2004; Ulusoy et al., 2007).

The outer membrane of gram-negative bacteria acts as a barrier and protects the cell. However, Ananta et al. (2005) observed that ultrasound (20 kHz/1746 W) destabilizes the outer membrane and facilitates the penetration of compounds. Moreover, ultrasound has been associated with the enhanced inactivation of microorganisms in chemical solutions.

The effects of ultrasound on different bacteria remain questionable. Scherba, Weigel, and O’Brien (1991) showed evidence of no significant difference between the percentage of gram-positive and gram-negative cells that are inactivated using ultrasound (26 kHz). Differences in the observed results mentioned could be related to treatment conditions, such as the frequency or time of application. The shape of cells can influence the effectiveness of ultrasound. Alliger (1975) studied the effects of ultrasound on cells and found that spherical cells (cocci) are more resistant to ultrasound than rod-shaped cells.

Pagán, Mañás, Alvarez, and Condón (1999) demonstrated the potential of ultrasound (20 kHz) to inactivate pathogenic microorganisms, such as L. monocytogenes, which are found in outbreaks of food poisoning. However, Ulusoy et al. (2007) commented that pathogens are resistant to different conditions of ultrasound treatment, particularly when used alone. Comparisons of different research results are complicated, considering that conditions of ultrasound treatment can be different. Thus, it is important to obtain a careful and detailed evaluation of all treatment parameters before determining the effectiveness of a particular method on fruits and vegetables.

Although most studies are performed with ultrasound in liquid systems and Guerrero, Lópezmalo, & Alzamora, 2001; Munkacsi & Elbasa, 1976; San Martin, Harte, Lieveld, Barbosa-Cánovas, & Swanson, 2001; Zenker, Heinz, & Knorr, 2003), it is known that microorganism contaminants in solid foods can also be inactivated using this technology (Mason et al., 2005).

Limitations in reducing microorganisms from the surface of fruits and vegetables might be associated with the occurrence of the multi-layered hydrophobic cuticle composed of cutin and wax molecules that cover the epidermis, and this cuticle is highly repellent (Velázquez, Barbini, Escudero, & Estrada, 2009). Thus, ultrasound has the potential to produce a deestabilization of bacteria biofilms and can be used for the inactivation of bacteria (São José & Vanetti, 2012). However, Sengül, Erkaya, Baslar, and Ertugay (2011) proposed that this method was not effective in killing microorganisms in food at room or sublethal temperatures. Some researches show that the use of ultrasound alone can contribute to decontaminate. In fresh Tuber aestivum truffles, Rivera, Venturini, Oria, and Blanco (2011) observed that ultrasound (35 kHz/10 min) promotes the elimination of 1 log CFU g⁻¹ of mesophilic microorganisms. Furthermore, others reports indicate that that ultrasound is effective to inhibit the incidence of deterioration and preserves the quality of post-harvest of fruits, as strawberries (Cao et al., 2010).

Ultrasound may be useful in the decontamination surface when applied in association with other method. Heat, high pressure, ultraviolet radiation, pulsed electric field or chemical methods are often used for cleaning and disinfection and can be applied in combination with ultrasound. In the case of fruits and vegetables, heat and pressure are not recommended for drastically altering the tissue structure of the plant. This review only discusses the combination of ultrasound with chemical compounds, such as commercial sanitizers, organic acids and others antimicrobials (Table 1).

Seymour et al. (2002) reported that the combination of ultrasound (32–40 kHz/10 min) with sodium hypochlorite at 50 mg L⁻¹ there was a further 1 log CFU g⁻¹ reduction of S. Typhimurium in lettuce compared with ultrasound or chlorine solution alone. Kim, Feng, Kushad, and Fan (2006) studied the effects of ultrasound (40 kHz) on the germination of broccoli seeds and E. coli O157:H7 and observed that none of the ultrasound treatments achieved more than a 2 log cycle reduction in the E. coli population without lowering germination to below 85%.

Ferrante, Guerrero, and Alzamora (2007) evaluated the additional efficiency of the combination of vanillin, citral, and
ultrasound to inactivate \textit{L. monocytogenes} in orange juice. The combination of high intensity ultrasound (20 kHz) with vanillin (500–1500 mg L\(^{-1}\)) and citral (25–100 mg L\(^{-1}\)) significantly increased \textit{L. monocytogenes} inactivation.

Scouten and Beuchat (2002) found that the combination of ultrasound (40 kHz) and \textit{Ca(OH)}\(_2\) might be an alternative for the decontamination of alfalfa sprouts inoculated with \textit{Salmonella} and \textit{E. coli} O157:H7. Apples treated with ultrasound at a 170 kHz frequency combined with 20 mg L\(^{-1}\) chlorine dioxide showed a 4-log CFU g\(^{-1}\) reduction in the population of \textit{Salmonella} and \textit{E. coli} O157:H7 (Huang et al., 2006). In addition, the population of \textit{S. Typhimurium} ATCC 14028 adhered to the surface of cherry tomatoes was reduced by approximately 4 log CFU g\(^{-1}\) in combination when ultrasound (40 kHz) and peracetic acid were used (São José & Vanetti, 2012).

Zhou, Feng, and Luo (2009) observed that use of ultrasound (200W/L) contributed to a reduction of 0.7–1.1 log CFU g\(^{-1}\) of \textit{E. coli} O157: H7 in spinach in all treatments compared with treatments using only chemical sanitizers. Ayyildiz, Sanık, and Ileri (2011) observed that a sequential combination of ultrasound (20 kHz) and chlorine dioxide provided a 3.2–3.5 log reduction in the number of \textit{E. coli} and total coliforms in raw wastewater, while the sum of the log reductions using individual treatments were 1.4–1.9 log CFU ml\(^{-1}\). Huang et al. (2006) investigated the decontamination efficiency of the combination of chlorine dioxide and ultrasound on apples and lettuce. Combining 10 min of ultrasound at 170 kHz and chlorine dioxide at 20 mg L\(^{-1}\) and 40 mg L\(^{-1}\) increased \textit{Salmonella} reduction by more than 2 log compared with the use of chlorine dioxide alone in apples. These results indicated that the association of ultrasound with other methods can result in a greater efficiency of sanitization.

Chen and Zhu (2011) demonstrated that the use of chlorine dioxide in combination with ultrasound (40 kHz) allows the maintenance of the post-harvest quality in the Japanese plum (\textit{Prunus salicina} L). Sagong et al. (2011) observed a synergistic effect in the use of lactic acid (2%), citric acid (2%) and malic acid (2%) combined with ultrasound (40 kHz) for 5 min in the inactivation of \textit{E. coli} O157:H7, \textit{S. Typhimurium} and \textit{L. monocytogenes} inoculated in organic lettuces, without significantly affecting the colour and texture.

These results indicate that ultrasound associated with other sanitizers is able to enhance the reduction of microbial flora in different products. However, the overall inactivation observed when ultrasound treatment is combined with antimicrobials depends on several factors, such as the type of antimicrobial agent, concentration and contact time. These details make it difficult to obtain general conclusions for other foods.

Other condition that ultrasound can be used for is inactivation of endospores or spores. It is known that few species of bacteria have the ability to produce highly these resistant structures. These structures can resist to a range of hazardous conditions, such as high temperatures, osmotic pressures, extremes pH values, and mechanical shocks. Spores protect against heat, radiation and dehydration (Joyce et al., 2003). Some species, as \textit{Bacillus cereus} is commonly found on fruits and vegetables. \textit{B. cereus} is a common soil inhabitant that can contaminate a variety of foods, such as rice, spices, milk and dairy products, vegetables, meat, cakes and other desserts. These bacteria produce several toxins and have been isolated from sprouts and sprouting seeds (Kim et al., 2006) and from raw vegetables used to prepare minimally processed refrigerated foods. The use of sanitizers to reduce spore bacteria levels in fruits and vegetables has been proposed as a safety strategy. \textit{Bacillus} and \textit{Clostridium} spores are more resistant to sonication than vegetative bacteria, and many of the heat-resistant bacteria are similarly resistant to ultrasound (Sanz, Palacios, Lopez, & Ordonez, 1985). Sagong et al. (2013) observed that the most efficient treatment for reducing \textit{B. cereus} spores was the combination of ultrasound (40 kHz) and 0.1% Tween 20, yielding reductions of 2.49 and 2.22 log CFU/g on lettuce and carrots, respectively, without causing deterioration of quality.

Ultrasound treatment, however, can induce changes in some characteristics of spores, such as swelling, surface erosion and growth stimulation (Burgos, Ordoñez, & Sala, 1972).

Studies have been conducted (Burgos et al., 1972) to evaluate the effect of heat, pressure and ultrasound waves on \textit{B. subtilis}. These studies were conducted in liquid medium and used other methods of preservation that are not appropriate for fruits and vegetables. It is well understood that these methods can produce diverse undesirable alterations in the natural characteristics of foods. Thus, more research is needed to evaluate the occurrence of spores on fruits and vegetables and propose new technologies of decontamination.

Fruits and vegetables can also be contaminated with moulds and yeasts. \textit{Fusarium}, \textit{Alternaria} and \textit{Phoma} are toxigenic moulds that affect plants, and these organisms can be transferred from diseased plant parts to contaminate healthy plants. After harvesting, other moulds could contaminate and spoil plant materials and produce toxic metabolites. Some of them could also produce mycotoxins while growing on these products, while others are pathogens that produce health hazards for the consumer (Tounj, 2005).

Synthetic chemical fungicides have been primarily used for reducing postharvest disease, mainly to control moulds on fruits and vegetables. However, consumer concerns about pesticide residues on foods, along with pathogen resistance, have increased the need to develop new methods to control postharvest diseases (Yang, Cao, Cai, & Zheng, 2011).

As Cao et al. (2010) showed, ultrasound (40 kHz) could be applied to control decay and maintain the quality of fresh products. It is known that large postharvest losses occur as a consequence of contamination by moulds and yeast.

Yang et al. (2011) investigated the effect of ultrasound (40 kHz/10 min) and salicylic acid (0.05 mM) individually or in combination on blue mould, which is caused by \textit{Penicillium expansum} in peach fruit. The results showed that the combination of ultrasound with salicylic acid was more effective than either individual treatment in controlling postharvest blue mould in peach fruit.

Guerrero et al. (2001) conducted studies with ultrasound (20 kHz) for 20 min in \textit{Sacccharomyces cerevisiae} cells and observed that the treatment punctured cells walls with a leakage of content and damage at the inner cellular level. In this same study, the cells appeared damaged in different forms, where some cells exhibited a disorganized inner structure or/rupture of the wall and plasma membrane, while others cells did not. Cameron et al. (2008) evaluated the effect of power ultrasound (20 kHz) on the survival of three microorganisms types in UHT milk and observed damage to the outer cell wall and inner cell membranes, and in the case of yeast, internal damage to cell organelles was also observed.

Raw fruits and vegetables have frequently been involved in the food-borne transmission of enteric viruses to humans. Among them, the Norovirus (NoV) and hepatitis A virus (HAV) are currently recognized as some of the most important human food-borne pathogens with regard to the number of outbreaks and people affected in the world (Fraissé et al., 2011). Although viral contamination can occur during all steps of food processing, primary production is a decisive stage at which prevention measures must be focused to minimize the risk of infection to consumers.

Postharvest sanitization can be an important technological solution for decreasing the bacterial load on fresh raw material (Fraissé et al., 2011; Koopmans & Duizer, 2004). Thus, it is important to evaluate new methods, such as ultrasound, that can eliminate viruses.
Reports that evaluate the inactivation of viruses on fruits and vegetables using new technologies are scarce. Su et al. (2010) evaluated the effect of high intensity ultrasound on the infectivity of murine norovirus, feline calicivirus and MS2 bacteriophage inoculated in phosphate-buffered saline and orange juice. These authors verified that the ultrasound effectiveness depended on the virus type, the initial virus titre and the suspension solution. They also concluded that ultrasound should be used in association with other methods to inactivate virus activity in food.

One justification for the difficult inactivation could be the small cell size and genetic make-up (particularly single-stranded RNA) of the viruses studied; thus, the viruses were more resistant to the methods to inactivate virus activity in food.

In Table 2 are presented the results of several studies that applied ultrasound on fruits and vegetables to reduce microorganism count.

5. Effect of ultrasound on the properties of fruits and vegetables

Fruits and vegetables undergo a series of changes after harvest due to their environment, nutrient supply and injury during the harvesting process. Metabolism is modified but continues to function in live plant tissues. The intention of food preservation methods is to extend the food’s shelf life through the inhibition of deleterious enzymatic reactions and microbial growth and the preservation of nutritional and sensory aspects. Thus, it is important to evaluate the consequences of new methods on the quality of fruits and vegetables.

5.1. Inactivation of enzymes

The evaluation of enzymes and intermediate compounds of the main metabolic pathways or secondary routes can be excellent indicators of product quality, stress condition or physiological disorder. The most important enzymes in fruits and vegetables are involved in taste and odour, such as lipoxigenase, lipases and proteases; texture is related to pectinases, β-glucosidases, cellulases, hemicellulases and peroxidase; colour is related to anthocyanases, peroxidase, lipoxigenase, chlorophyllase and polyphenoloxidase; and nutritional value is related to ascorbic acid oxidase, thiaminase and lipoxigenase (Lamikanra, 2002).

Studies evaluating the effects of ultrasound on the enzymes present in fruits and vegetables are scarce. Most studies are

<table>
<thead>
<tr>
<th>Microorganism</th>
<th>Conditions</th>
<th>Food</th>
<th>Time/min</th>
<th>Reduction</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Salmonella Typhimurium</td>
<td>US + 50 mg L⁻¹ NaOCl</td>
<td>Lettuce</td>
<td>10 min</td>
<td>1.2 log CFU g⁻¹</td>
<td>Seymour et al. (2002)</td>
</tr>
<tr>
<td>E. coli O157:H7</td>
<td>US 40 kHz + Ca(OH)₂ 1% treatment in bag</td>
<td>Alfalfa seeds</td>
<td>2 min</td>
<td>1.02</td>
<td>Scouen and Beuchat (2002)</td>
</tr>
<tr>
<td>Salmonella</td>
<td>US 40 kHz + Ca(OH)₂ 1% treatment in bag</td>
<td>Alfalfa seeds</td>
<td>2 min</td>
<td>1.61</td>
<td></td>
</tr>
<tr>
<td>L. monocytogenes</td>
<td>US 40 kHz + Ca(OH)₂ 1% treatment in bag</td>
<td>Alfalfa seeds</td>
<td>2 min</td>
<td>2.28</td>
<td></td>
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<tr>
<td>E. coli O157:H7</td>
<td>US 40 kHz + Ca(OH)₂ 1% treatment in bag</td>
<td>Alfalfa seeds</td>
<td>2 min</td>
<td>3.27</td>
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<tr>
<td>Salmonella</td>
<td>US 40 kHz + Ca(OH)₂ 1% treatment in bag</td>
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<td>2 min</td>
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</tr>
</tbody>
</table>

conducted using liquid mediums or foods, such as fruit juices and milk. Nevertheless, this article will discuss the effect of ultrasound on important enzymes that are also present in fruits and vegetables.

The mechanism of enzyme inactivation by ultrasound is associated with cavitation through mechanical and chemical effects (Mañas, Muñoz, Sanz, & Condón, 2006; Raviyan, Zhang, & Feng, 2005; Rawson, Tiwari, Patras, et al., 2011; Vercet, Burgos, Crelier, & Lopez-Buesa, 2001). When the bubbles collapse, high temperatures and shockwave pressures are generated. In addition to these effects, microstreaming is associated with high shear forces. Under these intense conditions, ultrasound could induce the disruption of hydrogen bonding and van der Waals interactions in polypeptide chains, leading to alterations of the secondary and tertiary structures of the protein. The biological activities of enzymes are lost by these modifications. The hot spots also lead to water molecule cleavage, generating high-energy intermediates, such as hydroxyl and hydrogen-free radicals. The free radicals formed might react with some amino acid residues that participate in enzyme stability, substrate binding or catalytic function that consequently modify biological activity (Mawson, Gamage, Terefe, & Knoerzer, 2011; Raviyan et al., 2005; Rawson, Tiwari, Patras, et al., 2011).

Some enzymes in food need to be inactivated to ensure them stabilization. The ultrasound inactivation of enzymes depends on properties, such as frequency and power, and factors, such as enzyme type, concentration, medium pH, and temperature (Mawson et al., 2011; Raviyan et al., 2005; Tarun, Kurchenko, & Metelitsa, 2006). Mawson et al. (2011) proposed that hydroxyl radicals produced by cavitation events are less effective on enzymes immobilized on the surfaces or within cells, such as those of vegetable and fruit tissues. However, there is no evidence of the genotoxic potential of ultrasound, as it is unclear whether extracellular or intracellular cavitation occurs.

Despite doubts about the effect of ultrasound in food enzymes, the possible effect of this method of food preservation in enzymatic activity will be discussed. Lipooxygenase (LOX) is present in most plant tissues, and in the presence of oxygen, it catalyses the oxidation of polyunsaturated fatty acids. Sterols and fatty acids are constituents of cell membranes, and any change in the composition of these compounds causes loss of fluidity. Lipid peroxidation can induce the loss of membrane integrity. LOX causes undesirable effects, such as the destruction of chlorophyll and carotenoids, the development of flavours and odours, and oxidative damage to proteins and vitamins (Lopez et al., 1994). The undesirable odour in vegetables is closely associated with the enzymatic peroxidation of unsaturated fatty acids.

Lopez et al. (1994) evaluated the effect of ultrasound (20 kHz) on soybean lipooxygenase buffer and observed a synergistic inactivation effect between heat conditions and ultrasound. Minimally processed fruits and vegetables are not subjected to heat treatment; therefore, it is useful to study the effect of ultrasound alone or in combination with other non-thermal treatments on the inactivation of enzymes, such as lipooxygenase. It is important that the treatment does not cause changes in taste and odour in food. López and Burgos (1995) reported a synergistic inactivation effect that increased with amplitude with ultrasound treatment in soybean lipooxygenase buffer. These same authors showed that hydroxyl radicals have a short half-life and that their range of action is short but can undergo radical—radical combination reactions to generate hydrogen peroxide. Hydrogen peroxide is an efficient LOX-inactivating factor, even at low concentrations and at room temperature.

Enzyme inactivation is a requisite for the stabilization of some food materials (Vercet et al., 2001). Fruit enzymes can cause the development of brown colour and the loss of vitamin C, which typically occurs in fruits that are served raw and are not subjected to blanch treatment like vegetables.

Peroxidase (POD) is an enzyme commonly found in vegetables (Lamikanra, 2002). The presence of this enzyme is associated with off-flavours and off-colours in raw and unblanched frozen vegetables (González-Aguilar et al., 2005; Ruiz-Cruz, Islas-Osuna, Sotelo-Mundo, Vázquez-Ortiz, & González-Aguilar, 2007). POD inactivation increases the shelf life of vegetables during frozen storage and can be an index for blanching adequacy. Heat conditions can be used for enzyme inactivation, but these enzymes were heat resistant, and this treatment can modify several food properties, such as flavour, colour or nutritional value (Cruz, Vieira, & Silva, 2006; Lamikanra, 2002).

Ercan and Soysal (2011) used ultrasound to inactivate peroxidase in tomatoes. When ultrasound was applied for 150 s at 50% ultrasonic power, the inactivation of tomato POD was 100%. Polyphenoloxidase (PPO) is an enzyme that participates in reactions that cause browning in fruits and vegetables (Ruiz-Cruz, Islas-Osuna, et al., 2007). Also known as catechol oxidase, diphenol oxidase, o-diphenolase or phenolase tironase, PPO is found in most fruits and vegetables, and the location of this enzyme in plant cells depends on the species, age and grade maturity. In green leaves, the enzyme is primarily found in chloroplasts (Lamikanra, 2002).

Softening is one of the most important changes usually observed during fruit ripening (Ketsa & Daengkanit, 1999). The loss of firmness, however, is most often attributed to the enzymatic breakdown of the middle lamella and cell wall (Ketsa & Daengkanit, 1999). A large number of enzymes participate in the degradation of pectic substances, such as pectinmethylesterase, polygalacturonase, β-galactosidase and cellulase.

Pectinmethylesterase (PME) is an endogenous pectic enzyme that has been identified primarily in the cell walls of vegetables cell that de-esterifies the methyl group of pectin and converts it into low methoxy pectin or pectic acids (Ketsa & Daengkanit, 1999; Lamikanra, 2002). The methoxy pectin or pectic acids can be depolymerized and hydrolysed by PG, resulting in viscosity and texture losses. To avoid quality losses, a partial or complete inactivation of PME together with the inactivation of PG is necessary during tomato processing (Raviyan et al., 2005). Pectin solubilization might contribute to loss from the cells walls, resulting in softening during ripening. Raviyan et al. (2005) observed that ultrasound (cavitation was measured by hydrogen peroxide yield rate 0.004–0.020 mg L min$^{-1}$) treatment effectively increased tomato PME inactivation compared with a thermal treatment at the same temperature. These authors related the increase of PME inactivation with an increase in the cavitation intensity expressed by H$_2$O$_2$ yield. This conclusion is consistent with Mawson et al. (2011), who reported the importance of free radicals for enzyme inactivation. The development of polygalacturonase (PG) activity and softening occurred simultaneously in cherry, mango, papaya, pear and tomato (Ketsa & Daengkanit, 1999; Lamikanra, 2002). PG hydrolyses glycosidic bonds between adjacent galacturonic acids, which are the monomeric units of pectin (Vercet, Sánchez, Burgos, Montañés, & Buesa, 2002).

5.2. Effect on food components

There has been increasing consumer interest in functional food with basic nutrient functions and properties that can promote health and prevent diseases. Fruits and vegetables have these properties.

The content of these food compounds is strongly associated with their processing, manipulation and storage. Some studies have shown that these operations have significant effects on the content of bioactive compounds (Plaza et al., 2011; Rawson, Tiwari, Patras,
et al., 2011; Rawson, Tiwari, Tuohy, et al., 2011; Ruiz-Cruz, Islas-Osuna, et al., 2007; Soria & Villamiel, 2010; Vandekinderen et al., 2009). Fresh-cut produce deteriorates faster than intact produce, and this deterioration affects not only microbiological quality but also nutritional and sensory quality. Therefore, we know that bioactive compounds decrease as a consequence of deterioration.

It is known that to control microbial contamination in fruits and vegetables, it is necessary to use chemical or physical agents, such as sanitizers. However, it is important that these processing steps and postharvest treatments do not cause a significant reduction in the produce’s nutrient content (Vandekinderen et al., 2008, 2009). These characteristics are relevant to a complete evaluation of the effectiveness of the decontamination step.

The food industry is constantly searching for processing technologies that allow the microbiological control of their products without changes in the nutritional and sensory characteristics of fresh product. Studies assessing the effect on nutritional content using ultrasound are typically performed with fruit juices. However, it is important to evaluate these parameters for products subject to new processing technologies to allow a more accurate evaluation of the potential for application in food industry.

Some researchers have demonstrated the effect of common sanitizers on the nutritional quality of fresh fruits and vegetables. Ruiz-Cruz et al. (2010) observed the reduction of vitamin C concentration in fresh cut Jalapeno peppers after treatment with chlorinated solutions. Vandekinderen et al. (2008) observed that gaseous chlorine dioxide reduced both α and β-carotene in grated carrots. Pérez-Gregorio et al. (2011) studied sanitizing technologies on the loss of flavonoids and anthocyanins in onion slices and concluded that onion flavonoids decreased using chemically disinfectant products and that ultrasound both maintained the initial levels of flavonoids and subsequently increased them. Ruiz-Cruz, Islas-Osuna, et al., (2007) showed that different sanitizers decrease carotene contents continuously during the storage of carrots. These results provided insight into the efficacy of physical methods, such as ultrasound, on these compounds.

During cavitation, hydroxyl radicals are produced, and these compounds can react with food compounds, but this interaction can be beneficial or not, depending on the process and food matrix (Soria & Villamiel, 2010). It was reported that L-ascorbic acid was degraded by ultrasound treatment, possibly due to the generation of free radicals. However, during storage, ascorbic acid retention in ultrasound-treated orange juice was shown to be better than that in a thermally processed juice (Lee & Feng, 2011). Aadil, Zeng, Han, and Sun (2013) observed different results when evaluate effect of sonication treatments on the contents of vitamin C in grapefruit juice. In that research, was observed a significant increase in vitamin C in all the sonicated juice samples (28 kHz, power set at 70%) as compared to control. This increase in vitamin C can be attributed to the elimination of entrapped oxygen due to cavitation (Cheng, Soh, Liew, & Teh, 2007).

The degree of hydroxylation influences the activity of antioxidants in food and biological systems. Radical formation is considered as a disadvantage for preserving the bioactivity of food components, such as phenolic compounds. Nevertheless, compounds such as flavonoids might enhance antioxidant activity by increasing the extent of hydroxylation (Soria & Villamiel, 2010). It is an important to make the appropriate choice of ultrasound parameters and conditions to treat fruit and vegetables without affecting the nutrients.

Alexandre, Brandão, and Silva (2012) treated strawberries with non-thermal technologies, such as ultrasound (35 kHz), and observed higher anthocyanin contents than in samples washed with chemical solutions when stored at room temperature for 6 days. Tiwari, O’Donnell, Patras, Brunton, and Cullen (2009) studied the stability of anthocyanins in sonicated (20 kHz) strawberry juice during storage and observed that this compound presented high levels of retention. Tiwari et al. (2010) observed the significant retention of anthocyanins in grape juices after treatment with ultrasound (at a constant frequency of 20 kHz and pulse durations of 5 s on and 5 s off): 97.5% AC (cyanidin), 48.2% MA (malvanidina) and 80.9% DA (delphinidin). Tiwari et al. (2009) commented that the degradation of the anthocyanins might be related to oxidation reactions promoted by the interaction of free radicals formed during sonication.

Vitamin C is easily destroyed during the processing and storage of foods. The vitamin C content indicated a better quality of fruit and vegetables because it is a quality parameter and should be maintained at an appropriate level (González-Aguillar et al., 2005). Ercan and Soysal (2011) observed that tomato juice treated with ultrasound (23 kHz at 75% power for 90 s) and 12% of initial vitamin C is lost.

Tiwari et al. (2009) observed the retention of ascorbic acid in strawberry juice treated with ultrasound (20 kHz using a 1500 W ultrasonic processor). This result was attributed to the elimination of dissolved oxygen during cavitations, which is essential for ascorbic degradations.

Ascorbic acid retention reduced colour change during storage, which is appropriate because colour is associated with the selection of fresh vegetables. The stability of vitamins is different from food to food, even when they are subjected to similar processing and storage.

Rawson, Tiwari, Patras, et al. (2011) studied the effect of ultrasound (20 kHz) on vitamin C and carotenoids and concluded that this treatment was consistent with the retention of these compounds in carrots discs. Ashokkumar et al. (2008) suggested that the hydroxyl radicals generated during ultrasound treatment could be used to improve the degree of hydroxylation in food materials and hence increase the antioxidant activity of foods. However, it should be noted that the generation of OH· radicals might affect the quality of some foods by reducing the antioxidant capacity. Intense sonication is also known to generate off-flavours. Therefore, it is important to choose adequate conditions for applying ultrasound treatment to maintain microbiological safety and nutritional quality.

Bhat, Kamaruddin, Min-Tze, and Karim (2011) observed an increase in the total phenolic content in sonicated (25 kHz) kasturi juice samples compared with the control. Some studies, such as that of Arzeni et al. (2012), evaluated the individual effect of ultrasound on food components by applying high-intensive ultrasound to food proteins. The results of that author indicated that ultrasound (20 kHz) induced modifications on functional properties, such as viscosity and solubility, and those changes were believed to be associated with molecular modifications, such as a hydrophobicity increase and particle size variation. These changes depend of the nature of protein and denaturation degree.

5.3. Sensory aspects

Some studies, such as the work of Vandekinderen et al. (2009), treated vegetables with chemical sanitizers and did not observe any change in the sensory quality immediately after treatment. Ultrasound has been studied as a method for the sanitization of fruits and vegetables (Ajouni, Sibriani, Premier, & Tomkins, 2006; Cao et al., 2010; Sagong et al., 2013; Säo José & Vanetti, 2012; Seymour et al., 2002; Zhou et al., 2009). Data that can be found in the literature explore the inactivation/removal of microorganisms and do not discuss their effects on the nutritional and sensory characteristics of fresh produce. Much of the data showed that ultrasound and related effects on nutritional and sensory characteristics are the result of studies in liquid foods, such as juices and
milk. However, we discuss these results to understand the possible changes that could occur through the use of this method in fruit and vegetable sanitization.

The quality of fruits and vegetables is based on several properties: texture, colour, flavour, and nutritional and functional characteristics. Biochemical transformations during the post-harvest development are primarily responsible for changes in the nutritional and sensory attributes of fruits and vegetables.

Fruit firmness is an important attribute that defines food quality. Texture is a physical characteristic that describes the responses to the deformation of a liquid or solid food product. As a key food quality attribute, texture is essentially determined by the microstructure of the product. The texture of food treated by ultrasound can be determined by the structure or property changes of proteins and enzymes during sonication.

In peppers, ultrasound (47 kHz) might injure the cell wall structure and induce changes in the texture (Gabaldón-Leyva et al., 2007). However, Cao et al. (2010) observed that ultrasound treatment (40 kHz) inhibited the decrease of firmness in strawberries. Alexandre et al. (2012) treated strawberries with different sanitizers and observed that samples treated with ultrasound (35 kHz) had 16% more firmness retention than the water-washed samples. These different observations should be due to different treatment conditions as frequency, time, amplitude.

A restricted number of reports discuss the flavour changes in food treated with ultrasound. The free radicals produced by cavitation might catalyse the degradation of flavour compounds. In other cases, such as wine, ultrasound can decrease the off-flavours (Lee & Feng, 2011).

The increase in volatile compounds is almost certainly due to reactions that take place during sonication treatment. It is uncertain that the nature of chemical reactions involved is specifically associated with only one or two of the components in a matrix chemically complex as milk (Riener et al., 2009).

Riener et al. (2009) observed that the long sonication (24 kHz) of milk produced volatile organic compounds that can be responsible for a “rubbery” aroma. This result shows that ultrasound-induced reactions must be considered towards possibilities successful introduction of this technology in food industry. The composition of food and parameters of ultrasound influence the potential changes in the flavour of the final product.

The colour of a food product is an important freshness-related attribute for the consumer to use in evaluating the quality of the product. Disinfection agents possess strong oxidising properties associated with deleterious effects on the colour of vegetables by inducing browning or bleaching of the vegetable tissue. Thermosonication (20 kHz) used in the blanching of watercress resulted in darker and less yellowish colours than in raw watercress (Cruz, Vieira, & Silva, 2006). Fava et al. (2011) treated grape berry fruits with ultrasound (20 kHz) and observed that this treatment induced significant differences in the colour parameters compared with untreated fruit. Alexandre et al. (2012) treated strawberries with ultrasound (35 kHz) for 2 min and observed better colour retention than in other treatments.

Aday, Temizkan, Büyükcan, and Caner (2013) verified that all ultrasound treatments were effective to reduce mould growth and 30 W and 60 W treatments maintained better textural properties compared with 90 W. As a result, it was concluded that power levels between 30 W and 60 W had improved quality and can be used to extend shelf life of strawberry.

6. Consumer acceptance

Consumers have been interested in products that have microbiological safety during production, a long shelf life, and minimal changes in the nutritional and sensory quality of the food (Rollin, Kennedy, & Wills, 2011). Such technologies, distinct of traditional processes, are aimed at reducing the loss of sensitive components, responsible for these sensory and nutritional qualities.

Furthermore, new trends in technology should also seek to protect the environment, showing concern for the balance between the production and consumption of food. It is important that technologies do not impact the environment, such as in the form of toxic by-products. These technologies are called “emerging technologies” or “clean technologies”.

Sound waves are generally considered safe, non-toxic, and environmentally friendly, and this gives the use of ultrasound a major advantage over other techniques. Studies confirming the potential application in food industry show that these new technologies and products should be evaluated and accepted for consumers. This evaluation will determine the commercial success or failure of these products. Many factors can influence the consumer’s acceptance of food innovations. According to Rollin et al. (2011), the comprehension of risk-benefit perceptions of consumers, socio-demographic and economic factors, knowledge and information will be essential to the realization and success of technological advances.

7. Industrial uptake, status update and challenges faced by the food industry

There is a great interest in ultrasound due to the fact that industries can be provided with practical and consistent ultrasound equipment. Currently, it is important to use a green novel technology that has a role in the environment sustainability. Fruit and vegetables are washed typically with chlorine solutions. However, there is a tendency in eliminating chlorine based compounds from the decontamination process and applying novel and emerging technologies in the food industry. Researches indicated that ultrasound can play an important role in fruit and vegetable decontamination. The adequate conditions, frequency, contact time and combination of treatments for different decontamination technologies are a challenge for the commercial application of ultrasound. More studies are required to use ultrasound technology for decontamination purposes in food industry.

8. Conclusion

Ultrasound has potential applications for fresh fruits and vegetables to control microbial contamination without changing their quality aspects. Some studies have shown that ultrasound treatment could inactivate microorganisms and some enzymes. The application of ultrasound can be effective when parameters (frequency, power, treatment time etc.) are adjusted to permit the efficiency of sanitization without changing the nutritional and sensory quality of the food. The chemical and physical energy generated by acoustic cavitation promotes severe damage to the cell wall, resulting in the inactivation of microorganisms. Thus, ultrasound can be a viable technique for quality assurance and food safety. Studies evaluating the use of ultrasound on fresh fruits and vegetables should be encouraged to provide a better understanding of the process and promote its application.

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sensorial quality, and nutrient content of grated carrots (Daucus carota L.). Journal of Agricultural and Food Chemistry, 56(14), 5723–5731.


