
Pulsed Electric Fields Effects on Health-Related Compounds and Antioxidant Capacity of Tomato Juice

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Abstract

A significant number of compounds found in plant-based foods such as vitamins, phenolic compounds and carotenoids exhibit health promoting biological functions. It is known that thermal treatment is effective in pasteurizing fruits and vegetable derivatives but the concentration and biological activity of most health-related compounds are dramatically reduced as thermal treatment intensity increases. Thus, a number of alternative technologies allowing low temperature processing such as pulsed electric field treatment (PEF) have emerged and many research efforts have been put towards their development and optimization. Exploratory studies have suggested that PEF have the ability to inactivate microorganisms and enzymes, while avoiding degradation of heat-labile components and, consequently, preserving the nutritional quality of the fresh-like food products. Hence, this processing technology could help not only to obtain safe and stable food products, but also to produce food commodities with high antioxidant properties. The aim of this review is to illustrate the ability of PEF technology as a non-thermal technology for keeping/improving health-related compounds and antioxidant capacity in tomato juices.

Keywords

Pulsed electric fields • Tomato juice • Lycopene • Vitamin C • Phenolic compounds antioxidant capacity

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Introduction

Regular intake of tomatoes and tomato based products has been associated with lower incidence of various forms of cancer, in particular prostate cancer, and heart diseases (Ames et al. 1993). This beneficial effect is believed to be due, at least partially, to the action of tomato antioxidant compounds, which could reduce oxidative damage in the body. Tomato is the predominant source of carotenoids such as lycopene, which exhibits high oxygen-radical scavenging and quenching capacities, and β -carotene. In addition, other antioxidant compounds such as phenolics and vitamin C also contribute to the beneficial effects of tomato. Phenolics possess reducing character, capacity of sequestering reactive oxygen species (ROS) and several electrophiles, tendency to self-oxidation and capacity to modulate the activity of some cell enzymes (Robards et al. 1999). Experimental studies have shown that vitamin C plays an important role in human health, including effects on immune system and the risk of Alzheimer diseases (Sánchez-Moreno et al. 2005). Despite health-related compounds can be affected by abusive temperatures, thermal processing remains as the most commonly used technology for inactivating microorganisms and enzymes in processed juices. In this context, non-thermal technologies such as pulsed electric fields (PEF) are being introduced by food processors as alternative or complementary to conventional thermal treatments. PEF processing is thought to be advantageous in front of conventional thermal treatments because of the limited changes in food quality attributes, which can be explained through the non-thermal nature of the technology. PEF have been also studied as possible treatments to enhance the generation of secondary plant metabolites by inducing stress reactions in fruit products. It has been described that PEF-induced stress affects metabolism with the consequent generation of reactive oxygen species (Galindo et al. 2009). In the last years, the impact of non-thermal technologies on health-related compounds and antioxidant capacity has been extensively evaluated. Based on these results, the effect of PEF processing on the main health-related compounds (carotenes, vitamin C and phenolic compounds) as well as antioxidant activity of tomato juices will be reviewed.

Health-Related Compounds in Tomato Juices

In the last years, a large number of epidemiological studies have related the consumption of a diet rich in fruits and vegetables with less predisposition to certain diseases such as cancer and cardiovascular disease (Ames et al. 1993). Although these beneficial effects have been initially associated with increased consumption of dietary fiber, at present there are a larger number of constituents of plant products that are partnering with this protective effect and are called, in general, bioactive compounds. Bioactive compounds are defined as nutrients that are in very low concentrations in food involved in secondary metabolism of plants, and can have a significant impact on human health. Recently, some nutrients have been included as bioactive compounds because, in addition to its essential function in the body, they have a beneficial health effect (Jeffery et al. 2003). Moreover, these compounds can be grouped according to their chemical structure in antioxidant vitamins, phenolic compounds, carotenoids, sulfur compounds and phytoestrogens, among others. In general, most bioactive compounds are characterized by a marked antioxidant activity. This activity is evidenced in their ability to trap oxygen radicals, nitrogen and organic radicals.

Carotenoids

Carotenoid compounds are a large group of fat-soluble pigments that are widespread in plants. They are divided into two groups, carotenes (hydrocarbon compounds) and xanthophylls (oxygenated derivatives of carotenes). Both are formed by long chains of conjugated double bonds presenting an isoprenoid structure. Thus, carotenoid compounds have the characteristic of having a bilateral symmetrical skeleton of 40 carbon atoms, consisting of four linked isoprene units. Lycopene is the most abundant carotenoid in the ripened tomato, accounting for approximately 80–90% of the total pigments. The amount of lycopene in tomato have been reported to varied from 18 to 170 mg/kg fresh weight since the concentration varied considerably among cultivars, stages of maturity and growing condition (Martínez-Valverde et al. 2002). In addition to lycopene, β -carotene, γ -carotene, ξ -carotene, δ -carotene, lutein, phytoene and phytofluene are also present in tomato and tomato products in a much smaller amount. β -carotene and γ -carotene are very important from the nutritional point of view due to their activity as precursors of vitamin A. In general, carotenoids protect plant cells from oxidation and therefore their decomposition. In the human body, they act as antioxidants, protecting cell membranes from free radical action. An inverse association between carotenoid intake and the risk of certain cancers (lung and stomach) and against arteriosclerosis has been reported.

Vitamin C

The term vitamin C includes ascorbic acid, dehydroascorbic acid and ascorbate salts.

Ascorbic acid is a lactone with an enediol group on carbons two and three (cyclic ester of a hydroxyl carboxylic acid). These two enol hydrogen atoms are those who

give to this compound its acidity and provide electrons for its antioxidant function. The stability properties of vitamin C vary markedly as a function of environmental conditions such as pH, light and temperature, as well as the concentration of trace metal ions, oxygen and degradative enzymes. The first product of ascorbic acid oxidation is the radical monodehydroascorbate, also known as semihydroascorbate, or ascorbate-free radical. If allowed to persist though, two molecules of monodehydroascorbate will also spontaneously disproportionate to dehydroascorbic acid. Dehydroascorbic acid itself is unstable and undergoes irreversible hydrolytic ring cleavage to 2,3-diketogulonic acid in aqueous solution, which possesses no vitamin C activity (Davey et al. 2000). Further reactions in ascorbic acid degradation beyond 2,3-diketogulonic acid are of no nutritional consequence, but contribute to the flavor and color changes associated with browning reactions. Vitamin C is essential for humans because it can not synthesized by the body due to the lack gulonolactone oxidase enzyme. Vitamin C is widely known for its role in preventing scurvy. In addition, modulate a number of important enzymatic reactions. Vitamin C plays also a vital role in the conversion of 3,4-dihydroxyphenylethylamine to noradrenaline. Vitamin C increases the absorption of iron by converting Fe^{3+} into Fe^{2+} that is absorbed more readily and is required in the metabolism of the amino acid tyrosine (Burini 2007). Vitamin C is also an important antioxidant that can reduce or eliminate superoxide, hydroxyl radical, hypochlorous acid and other free radical and oxidants. The major sources of vitamin C are fruits and vegetables such as tomatoes, although the amount of vitamin C that contain depends on the variety, soil conditions, climate, maturity, storage conditions and processing.

Phenolic Compounds

Phenolic compounds are a group of bioactive compounds that are synthesized by plants during their development, in response to various adverse conditions such as infections, wounds, filings, etc. secondary metabolites (Dixon and Paiva 1995). There are approximately 8000 known natural compounds, which are characterized by having at least one phenolic ring in them molecular structure. Phenolic compounds are synthesized through the malonic or shikimic acid pathways, or both as it is the case of flavonoids. Phenolic compounds are divided into two large groups, flavonoids and non-flavonoids compounds. Flavonoids is the generic term given to polyphenolic compounds characterized by a chemical structure based on a C₆-C₃-C₆ skeleton, which consists of two phenyl rings and heterocyclic ring. Flavonoids generally are bound to sugar molecules or partially polymerized resulting in dimers, trimers, etc., to form multi-linked complex as condensed tannins. Flavonoids comprise several thousands of compounds and are divided into flavonols, flavones, flavanones, anthocyanins, flavanols and isoflavones. Among flavonols, tomatoes are characterized by having high contents of kaempferol, quercetin and myricetin; however the concentration depends on several factors such as variety, fruit growing conditions, geographical origin and harvesting time, among others. Within the group of

non-flavonoids, phenolic acids and stilbenes are the main compounds. The group of phenolic acids includes the hydroxybenzoic and hydroxycinnamic acid derivatives. Some examples of hydroxybenzoic acid derivatives are p-hydroxybenzoic, ellagic and gallic acid. Within hydroxycinnamic acids, chlorogenic, p-coumaric, caffeic and ferulic acids are found in greater proportion in fruits. Martínez-Valverde et al. (2002) reported a concentration of phenolic compounds between 272.6 to 498.6 mg/kg fresh weight in different cultivars of tomatoes. In addition, chlorogenic acid has been suggested to be the major phenolic compound present in tomato juices in concentrations of 4 mg/100 ml (Odriozola-Serrano et al. 2009).

Antioxidant Capacity

Most bioactive compounds have a marked antioxidant capacity trapping oxygen, nitrogen and organic radicals. A free radical is defined as a chemical species having in its structure one or more unpaired electrons, which makes it a very unstable compound, with high capacity of forming other free radicals and damage cellular structures (Kaur and Kapoor 2001). Free radicals are generated naturally during metabolism by the partial reduction of the oxygen molecule, thereby forming reactive species such as hydrogen hydroperoxide (H_2O_2), superoxide ($O_2^{\bullet-}$), hydroperoxyl (HO_2^{\bullet}) and hydroxyl (OH^{\bullet}), among others. Free radicals are capable of damaging reversibly or irreversibly all kinds of biochemical compounds. The accumulation of these species leads to the appearance of oxidative deoxyribonucleic acid (DNA) damage, as well as proteins and lipids of cell membranes (lipid peroxidation), events closely related to aging processes of tissue and the onset of degenerative diseases (Ames et al. 1993). Free radicals and other highly reactive oxygen compounds are believed to contribute to cause a variety of diseases, especially chronic diseases related to age, such as cancer, Alzheimer's, Parkinson's, cataracts, arteriosclerosis and others.

Effect of Pulsed Electric Field-Processing Critical Parameters on Health-Related Compounds

Carotenoids

Studies on tomato indicate higher carotenoids concentrations in PEF-processed tomato juices than in the untreated samples (Odriozola-Serrano et al. 2007, 2008a; Vallverdú-Queralt et al. 2013) (Fig. 1).

In this way, Vallverdú-Queralt et al. (2013) suggested that treated tomato fruit with moderate-intensity PEF at 1.2 kV/cm and 5 monopolar pulses of 4 μ s with frequency of 0.1 Hz led to maximum increases of α -carotene, 9- and 13-*cis*-lycopene, relative content by 93%, 94% and 140%, respectively. An enhancement of up to 46.2% in the lycopene relative concentration of tomato juices after applying PEF

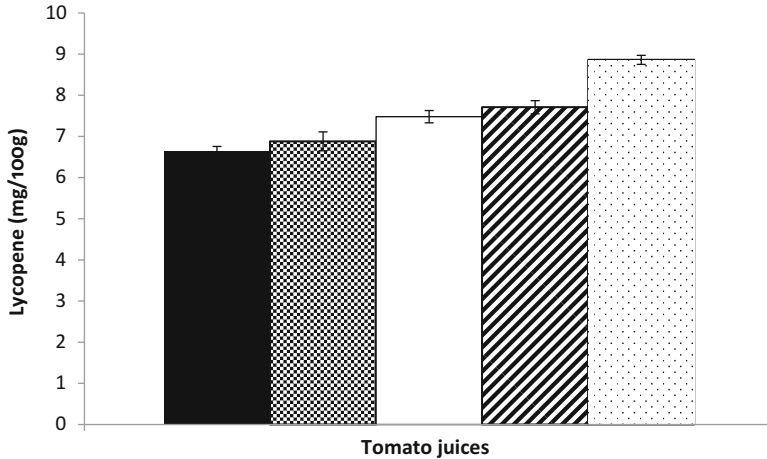


Fig. 1 Effects of PEF treatment on lycopene content of tomato juices: (■) untreated, (▨) 20 kV/cm for 1000 μ s in bipolar mode using 1- μ s pulses at 250 Hz, (□) 25 kV/cm for 1000 μ s in bipolar mode using 1- μ s pulses at 250 Hz, (▩) 30 kV/cm for 1000 μ s in bipolar mode using 1- μ s pulses at 250 Hz and (◻) 35 kV/cm for 1000 μ s in bipolar mode using 1- μ s pulses at 250 Hz. Data shown are mean \pm standard deviation

treatments with pasteurization purpose (35 kV/cm for 1000 μ s, $T < 40$ °C) has been reported by Odriozola-Serrano et al. (2007). Consistently, Sánchez-Moreno et al. (2005) observed that the content of total carotenoids in “gazpacho”, a cold vegetable soup where tomato is the major component, increased approximately a 62% after applying bipolar 4- μ s pulses of 35 kV/cm for 750 μ s at 800 Hz. Some arguments have been proposed to justify the relative increase of carotenoids in PEF-processed juices. Nguyen and Schwartz (1999) suggested that homogenization and heat treatment disrupt cell membranes and protein-carotenoids complex, making carotenoids more accessible for extraction. Rodríguez-Amaya (1997) hypothesized that thermal treatment may lead to an increase in some individual carotenoids, owing to greater stability, inactivation of oxidative and hydrolytic enzymes, and unaccounted moisture loss, which concentrates the sample. However, several authors (Torregrosa et al. 2005; Odriozola-Serrano et al. 2009; Vallverdú-Queralt et al. 2013) suggested that the changes in the relative amounts of carotenoids after PEF-treatment are not consistent for similar compounds. In this way, Odriozola-Serrano et al. (2009) reported that the increase in lycopene content just after PEF processing coincided with a depletion of phytoene and neurosporene content compare to the untreated juice. Although the reason for these results is not well understood, it was speculated that carotenoid conversions could be triggered by the PEF treatments. This would explain the increase in lycopene at the expense of its precursors. PEF processing may accelerate lycopene synthesis in tomatoes, involving the conversion of geranylgeranyl diphosphate (GGPP) to phytoene by phytoene synthase and the conversion

of phytoene to phytofluene, β -carotene, and lycopene by phytoene desaturase (Fraser et al. 1994).

Process parameters such as electric field strength and treatment time are the most important variables to be controlled in order to optimize the inactivation of microorganisms and enzymes by PEF and have been also shown to play a key role in carotenoids retention (Cortés et al. 2006). Odriozola-Serrano et al. (2008b) suggested that the greater the electric field strength and treatment time the higher the relative lycopene content when tomato juice is subjected to electric field strength from 20 to 35 kV/cm for up to 2000 μ s using bipolar 1- μ s pulses at 250 Hz. Consistently, carotenoid concentration rose as treatment time increased when PEF treatments at 25 or 30 kV/cm were applied to orange-carrot juice (Torregrosa et al. 2005). Furthermore, other treatment variables such as pulse frequency, pulse width and polarity could affect the carotenoid retention in tomato juices. Odriozola-Serrano et al. (2007) observed that higher frequency and pulse width resulted in a greater lycopene relative content. The simultaneous increase in the two variables from 50Hz, 1- μ s to 250Hz, 7- μ s resulted in an increment of lycopene of 43%, whereas rising one variable and keeping the other constant lycopene content rose between 8.4% (50Hz, 7- μ s) and 26.4% (250Hz, 1- μ s). The use of bipolar pulses raises lycopene in tomato (Odriozola-Serrano et al. 2007) and watermelon juice (Oms-Oliu et al. 2009) more than monopolar treatments. A difference of 7.6% in lycopene was observed in tomato juices when using bipolar over monopolar pulses after applying a PEF treatment set up at 35 kV/cm for 1000 μ s at 150Hz and 7 μ s pulse width (Odriozola-Serrano et al. 2007).

Several models have been used to describe the microbial destruction and enzymatic inactivation as a function of the PEF critical parameters. However, a few mathematical models have been proposed to predict the variation of the antioxidant potential of tomato juice as affected by key parameters involved in PEF treatments. Odriozola-Serrano et al. (2007) proposed a second-order response function ($R^2_{\text{adj}} = 0.94$) to fit lycopene retention after the application of electric field treatments set at 35 kV/cm for 1000 μ s using squared wave pulses, frequencies from 50 to 250 Hz, and pulse widths from 1 to 7 μ s, in monopolar or bipolar mode. Frequency, pulse width and pulse polarity affected the lycopene content linearly, whereas only the quadratic term of frequency was significant. The combined effects of frequency and pulse width, as well as frequency and pulse polarity, were included in the model as interaction terms. On the other hand, Odriozola-Serrano et al. (2008b) suggested that a model proposed by Peleg can be used to relate the kinetics of lycopene changes in tomato juice as affected by PEF electric field strength and time. The fitting performance of Peleg model was good irrespective of the electric field strength ($R^2_{\text{adj}} = 0.759\text{--}0.992$).

Some authors have studied the changes of some carotenoids in PEF-treated tomato juices during storage (Odriozola-Serrano et al. 2007; Vallverdú-Queralt et al. 2013). Odriozola-Serrano et al. (2009) observed a decrease in the amounts of all individual carotenoids in tomato juice over time, with the exception of β -carotene

and phytoene content, which were maintained for 56 days, regardless of the treatment applied. *Trans*-lycopene decreased much more considerably than other carotenoids through the storage period as a result of isomerization phenomena, since *trans*-lycopene can be converted to 13-*cis*-lycopene, which can be transformed into other *cis*-isomer Vallverdú-Queralt et al. (2013). Odriozola-Serrano et al. (2007) suggested that the concentration of lycopene decreased as storage time increased regardless of the processing treatment applied following first-order kinetic models ($R^2 \geq 0.866$) with rate constants from 1.6×10^{-2} to 2.3×10^{-2} days⁻¹. Oxidation of the highly unsaturated carotenoid structure may occur by autooxidation, which is a spontaneous free-radical chain reaction in the presence of oxygen, or by photooxidation produced by oxygen in the presence of light (Kidmose et al. 2002). The severity of oxidation depends on the structure of carotenoids and the environmental conditions, whereas the compounds being formed depend on the oxidation process and the carotenoids structure (Ramakrishnan and Francis 1980).

On the other hand, PEF-processed tomato juices kept higher amounts of carotenoids than heat-treated juices for 56 days at 4 °C (Odriozola-Serrano et al. 2008a, 2009; Vallverdú-Queralt et al. 2013). Accordantly, Cortés et al. (2006) reported higher stability of these health-related compounds in comparison to thermally-pasteurized juices. Most differences between PEF and heat treatments can be explained in terms of the temperature/time binomious.

Vitamin C

The retention of vitamin C ranged from 60.6 to 99.0% in PEF-treated juices in comparison to the fresh juice when tomato juices were subjected to an electric field strength set at 35 kV/cm for 1000 μ s, monopolar or bipolar square-wave pulses from 1 to 7 μ s, and frequencies from 50 to 250 Hz (Odriozola-Serrano et al. 2007).

Higher vitamin C retentions (72.2–99.9%) were reported in PEF-treated watermelon juices at similar conditions (Oms-Oliu et al. 2009). Torregrosa et al. (2005) reported vitamin C retentions between 87.5 and 97% in orange-carrot juice treated at different electric field strengths (25–40 kV/cm) for different treatment times (from 30 to 340 μ s) using 2.5- μ s bipolar pulses. Applying the same PEF conditions, differences in vitamin C retention among PEF-treated juices could be due to their different pH; since more acidic conditions are known to stabilize vitamin C (Davey et al. 2000).

Although specific PEF conditions that assure microbial inactivation have been used to pasteurize fruit juices, few studies have aimed at evaluating the effect of PEF critical parameters on vitamin C. Vitamin C content significantly depended on the PEF processing parameters, pulses applied in monopolar mode, as well as decreasing electric field strength, treatment time, frequency and pulse width led to increasingly high vitamin C retention in tomato juice (Odriozola-Serrano et al. 2007, 2008b). An increase of vitamin C degradation when electric field strength, treatment time and pulse width rise in PEF-processed gazpacho has been reported by Elez-Martínez and Martín-Belloso (2007). Degradation of ascorbic acid

depends upon many factors such as oxygen, light, processing temperature and time (Davey et al. 2000). In general, the milder the treatment, the better the vitamin C retention. Up to now, some authors have studied the kinetics of vitamin C degradation under PEF conditions and have stated that this vitamin depletion can be approached through simple first-order models (Torregrosa et al. 2006). However, Odriozola-Serrano et al. (2008b) proposed a model based on the Weibull distribution function ($R^2_{\text{adj}} \geq 0.747$) to better prediction of the kinetic degradation of vitamin C regardless the treatment conditions. Changes in vitamin C of tomato juice treated by PEF have been also modelled as a function of pulse width, frequency and polarity using a second-order regression model ($R^2_{\text{adj}} = 0.84$) (Odriozola-Serrano et al. 2007).

A number of studies have proven the effectiveness of PEF in achieving higher vitamin C contents in tomato juices comparison with heat treatments. Vitamin C retention just after treatment in heat-treated (90 °C, 60 s) tomato juice was 79.2%, whereas in PEF-processed juice (35 kV/cm for 1500 μ s in bipolar 4- μ s pulses at 100 Hz, $T < 40$ °C) a 86.5% retention was attained (Odriozola-Serrano et al. 2008a). Similarly, PEF-processing resulted into greater vitamin C retention than equivalent thermal treatments in terms of microbial inactivation in other fruit juices (Odriozola-Serrano et al. 2009). Most differences between PEF and heat treatments can be explained by the different temperatures reached through processing. Ascorbic acid is a heat-sensitive bioactive compound in the presence of oxygen. Thus, high temperatures during processing can greatly affect the rates of its degradation through an aerobic pathway. It must be emphasized that the reported treatments were conducted while cooling the juice, so that its average temperature at the exit of the treatment chamber did not increase above 40 °C even when treatment conditions exceeding those required for microbial inactivation were applied. Some studies have suggested that the concentration of vitamin C decreased with the storage time in both heat-treated and PEF-treated juices following an exponential trend. In addition, it has been demonstrated that vitamin C is better retained in PEF-treated juices than in those thermally-processed after 56 days at 4 °C (Fig. 2). A first-order kinetic model adequately fitted vitamin C depletion ($R^2 = 0.9680\text{--}0.838$) as a function of the storage time in tomato juices with degradation rates (k_t) between 2.4×10^{-2} to 3.0×10^{-2} days⁻¹ (Odriozola-Serrano et al. 2008a). Storage conditions such as temperature or oxygen concentration may have a significant influence on the rates of vitamin C degradation.

Few studies have been carried out to assess the effects of consuming fruit and vegetable products treated by PEF on vitamin C bioavailability in a healthy human population. Sánchez-Moreno et al. (2005) suggested that drinking two glasses of PEF-treated gazpacho soup (500 mL/day) containing approximately 73 mg of vitamin C was associated with a significant increase in plasma vitamin C concentration and a decrease in plasma levels of 8-epiPGF2 α (biomarkers of lipid peroxidation) and uric acid. PEF-treated gazpacho soup daily maintained the vitamin C bioavailability and antioxidant properties of fresh products with a longer shelf-life, increasing plasma vitamin C, and decreasing oxidative stress and inflammation biomarkers in healthy humans.

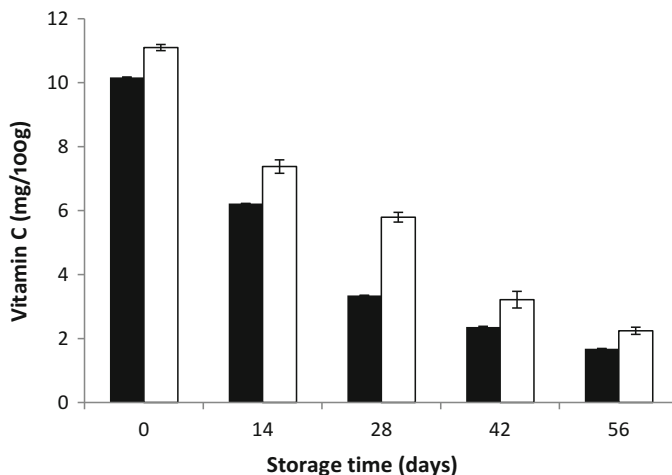


Fig. 2 Effects of PEF and heat pasteurization on vitamin C content of tomato juice throughout storage at 4 °C. Tomato juices: (■) PEF at 35 kV/cm for 1500 μ s in bipolar 4- μ s pulses at 100 Hz, $T < 40$ °C and (□) heat pasteurized at 90 °C for 60s. Data shown are mean \pm standard deviation

Phenolic Compounds

Literature provides scarce evidence of changes in the content of phenolic compounds in juices as affected by PEF treatments. Pulsed electrical discharge treatments have been reported to degrade phenolic compounds in aqueous media. Previous studies have reported a dramatic degradation of phenolic compounds in aqueous solutions with high iron concentration by applying exponential-wave pulsed corona discharges of high intensity (46 kV) (Grymonpré et al. 2001). In complex food systems, such degradation after the application of PEF treatments under common conditions of operation (electric fields less than 40 KV/cm) is not usually observed. This obeys to the fact that the effect of PEF treatments regarding the phenolic contents of juices appears to be influenced by complex mechanisms in which other molecular and radical species are involved. Hence, whereas thermal treatments, even with pasteurizing intensity, have been found to cause a considerable loss of the phenolic content of fruit juices, the phenolic content of PEF treated juices has not been reported to substantially change, at least in a quantitative basis. Odriozola-Serrano et al. (2008a) could not observe any significant difference between the overall phenolic content of fresh and PEF-treated tomato juices, just after processing, even after prolonged treatment conditions (35 kV/cm for 1500 μ s with 4- μ s bipolar pulses at 100 Hz). In their study, tomato juices preserved using mild heat and PEF were found to maintain their total phenolic content, just after processing and throughout 90 days of refrigerated storage. A plausible explanation for such outstanding preservation is the complete inactivation of oxidative enzymes such as peroxidase and polyphenol oxidase. Peroxidase is the main enzyme involved in the loss of quality of tomato juice. As well, peroxidase is involved in the oxidative degradation of phenolic

compounds. The low effect of these mild treatments on phenolics is supported by the fact that both mild heat and PEF conditions have been shown to inactivate peroxidase enzymes in juices. In the specific case of tomato, Aguiló-Aguayo et al. (2010) reported a 97% reduction of the initial peroxidase activity in PEF-treated tomato juices. However, the extent of this depletion could strongly depend on the intensity of the applied treatments.

On the other hand, qualitative data show slight differences in the amount of individual phenolic compounds as affected by PEF treatments. In this regard, the content of chlorogenic acid, which is the most important phenolic compound in tomato juice, has been reported to be greater than in tomato juices treated at 90 °C for 1 min over storage (Odriozola-Serrano et al. 2009). The initial chlorogenic acid concentrations in PEF-treated tomato juice were kept for almost 50 days under refrigeration (4 °C). The content of other phenolic acids such as quercetin and kaempferol, which are also present in important quantities in tomato, were not affected by the treatments but exhibited a slight but significant decrease through storage. However, this decrease was always less important than that observed in heat-treated juices, regardless the treatment intensity, throughout 56 days at 4 °C. In contrast, PEF-treated tomato juices underwent a substantial depletion of p-coumaric acid during storage, probably as a consequence of the hydroxylation of this phenolic compound to caffeic acid, a phenolic compound with higher antioxidant power, whose content slightly increased over time. This is usually an enzymatically-catalysed process, and could reveal the presence of residual monophenol monooxygenase activity in the treated samples. However, it must be noted that differences between PEF-treated and heat-treated juices were not significant, although the highest increase in caffeic acid concentrations was found in the PEF-treated juice.

More recently, some researchers have proposed the application of mild PEF treatments as a way for eliciting an increase in the amount of antioxidant compounds in fruits prior to processing. Phenolic compounds have been shown to be particularly affected by PEF treatments. In this case, the treatments must not be understood as a preservation alternative, but as a source of oxidative stress in the intact fruit, which responds by activating complex metabolic pathways that end with the production of phenolics and other secondary metabolites with antioxidant power. Vallverdú-Queralt et al. (2012) reported a 44.6% increase in the total phenolic content of tomatoes after a treatment of 30 exponential-wave pulses of 1.2 kV/cm and a subsequent incubation time of 24 h. The extent of this stress was shown to be strongly dependent on the intensity of the treatment. An overall increase in phenolics was observed as the number of pulses rose. Authors found that a faster metabolic response took place within the range of 25–30 pulses at 0.7–11 kV/cm. The extent of this response seems to be related to the generation of pores in the cell membrane as well as to the alteration of the normal function of voltage-gated ion channels, which are a specific type of transmembrane ion channel of the cell membrane. Under certain conditions these pores can be reversed by the cell and deregulation of the ion channels could lead to the influx of calcium ions, thus activating the production of a protein that, in turn, phosphorylates polyphenol ammonia liase, an enzyme involved in the production of phenolic compounds by plant tissues. Furthermore,

PEF treatments have been found to elicit a different response when considering different phenolic compounds. A study carried out by Vallverdú-Queralt et al. (2013) found that, whereas an increase in hydroxycinnamic acids and flavanones was found in tomatoes 24 h after the treatments, other phenolic compounds such as flavonols, coumaric and ferulic acids were not affected. These findings suggest the use of mild PEF treatments for enhancing the bioactive concentration of raw materials that can be transformed into healthier juices.

Antioxidant Capacity

PEF treatments do not lead to dramatic changes in the overall antioxidant content of tomato juices. A high retention of the antioxidant capacity was reported by Elez-Martínez and Martín-Belloso (2007) in a PEF-treated cold vegetable soup in which tomato was the main component. Odriozola-Serrano et al. (2007) carried out a screening study aimed at evaluating the impact of different critical parameters on the retention of antioxidant compounds in tomato juice. The optimal conditions for achieving maximal antioxidant capacity (92.3%) in the treated juice were 35 kV/cm for 1000 μ s at 150 Hz with 4 μ s square-wave bipolar pulses. It was shown that pulse polarity was the most significant parameter affecting the antioxidant capacity retention. In this sense, treatments in bipolar mode have been shown to produce tomato juices with greater antioxidant capacity. On the other hand, an increase in frequency or pulse width was generally deleterious for the antioxidant capacity of the juices. In a parallel work, Odriozola-Serrano et al. (2008b) confirmed that, within the 15–35 kV/cm range, field intensity does not seem to play the most significant role regarding antioxidant capacity. In addition, these authors proposed a first order model and a model based on the Weibull function to fit the changes in antioxidant capacity values as affected by PEF. Modelling allowed estimating the mean treatment time to achieve complete loss of antioxidant capacity, which ranged from 3271 to 15,970 μ s, depending on the electric field strength applied. These values are within the range established by Elez-Martínez et al. (2006) for achieving the complete inactivation of peroxidase enzyme, which allows stating that the antioxidant capacity of tomato juice is in a similar range of sensitivity to PEF treatments than peroxidase enzymes. Therefore, processing conditions need to be cautiously selected in order to obtain maximum inactivation of enzymes without causing a great loss of antioxidant potential.

In a later study, tomato juices subjected to selected PEF conditions (35 kV/cm for 1500 μ s in bipolar 4- μ s pulses at 100 Hz) and mild heat treatments (90 °C for 30/60 s) were compared throughout storage (Odriozola-Serrano et al. 2008a). It was found that antioxidant capacity, determined on the basis of the 2,2-diphenyl-1-picrylhydrazyl stable radical (DPPH), did not significantly change as a consequence of the PEF treatment application. Antioxidant capacity values decreased throughout refrigerated storage for either heat-treated or PEF-treated juices, although a slightly lower decrease was reported in the latter case. Correlations between antioxidant capacity and other antioxidant compounds seemed to indicate a strong correlation of the antioxidant capacity with vitamin C and lycopene contents.

To sum up it can be concluded that PEF treatments might be used to obtain tomato juices and other tomato-based products with high antioxidant characteristics. Conditions for optimal retention of antioxidant compounds need to be carefully selected in order to inactivate microorganisms and enzymes without compromising the health-related content of tomato juice.

Conclusions

PEF, as non-thermal technology, allows obtaining tomato juices with minor changes or increased content in health-related compounds just after the treatment and through the storage period. Future research should focus on identifying the role of biochemical transformations of health-related compounds precursors as influenced by processing. The evaluation of the effects of PEF critical parameters on bioactive compounds as well as on the antioxidant capacity of tomato juice is useful to achieve the optimal processing conditions to obtain tomato juice with high nutritional quality. Some proposed mathematical models may help to predict the variation of the antioxidant potential of tomato juice as affected by key parameters involved in PEF treatments. New applications of PEF technology should be further explored not only to obtain safe and stable juices with high content of health-related compounds but also to assess their bioavailability.

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