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Effectiveness of Electric Field strength and treatment time on quality properties of Papaya (*Carica papaya*) Fruit Juice

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Papaya fruit was an underutilized agricultural crop containing high content of nutritional and bioactive components ideal for juice production. Pulsed electric field was an novel technique ideal for juice expression. This research evaluated the influence of pulsed electric field (PEF) in different electric field strength (1.2÷1.8 kV/cm) and treatment time (2÷10 ms) to the quality properties of papaya fruit juice. Results showed that there was a significant increase in carotenoid content in papay juice treated with different electric field strengths and durations as compared to control treatment. Polyphenol oxidase activity was significantly decreased by PEF treatment compared with that of the control. Ascorbic acid and total soluble solid in PEF-papay juice were stable during storage. Pulsed electric field treatment at 1.4 kV/cm electric field strength in 6 ms treatment duration was appropriate for papaya juice production. The PEF-papaya juice had stability for 15 weeks of preservation at ambient environment.

Keywords: Ascorbic acid, carotenoid, electric field strength, papaya, polyphenol oxidase, pulsed electric field, total soluble solid, treatment duration

INTRODUCTION

Papaya fruit was a rich source of carbohydrates, vitamins, proteins, vitamins A and minerals (Bari et al. 2006).It contained numerous bioactive components like saponin, alkaloid, tannin, β-carotene, lycopene, anthocyanin, flavonoid and phenolic beneficial for human health (Chukwuka et al. 2013; Mahendra and Nikhil, 2016; Pavithra et al. 2017). This fruit had antioxidant, antimicrobial, anticarminative, anticancer, hepato-protective, immunological and other therapeutic properties (Farhan et al. 2014).

The polyphenoloxidase was a copper-containing enzyme contributing to enzymatic browning of vegetables and fruits. Its catalizing activity could be effectively retarded by chemical substances, however the residual chemical agents also generated harmful risks to human health. The conventional thermal treatment by hot water blanching successfully retarded the enzymatic browning appearance in fruits and vegetables. However, this physical thermal method was not always appropriate for polyphenoloxidase inactivation of fruit juice because temperature would create negative impact to physicochemical quality. Physical treatment without thermal treatment was very necessary to resolve this matter.

Pulsed electric field (PEF) was an innovative technique capable to permeabilize cells of fruits and vegetables without rising temperature as well as damaging tissue while maintaining food basic structure (Vorobiev and Lebovka 2008; Donsi et al. 2010; Puertolas et al. 2012). This method was applied for solid–liquid extraction, separation, dehydration, freezing, peeling to improve process yield, process velocity, food quality, cost efficiency. PEF involved the intermittent (<300 Hz) application of direct current high voltage pulses (< 50 kV) of very short width (microseconds to milliseconds) to a target foodstuff via at least one electrode and its consequent discharge via at least one grounding. This generated a potential difference between the electrodes (Puertolas et al. 2016). There were three levels of electric field strengths: low intensity (<0.1 kV/cm), medium intensity (0.1÷1.0 kV/cm) and high intensity (> 1.0 kV/cm). Critical electric field strength in the range of 1–2 kV/cm was reported for plant cells while 12–20 kV/cm was suitable for microorganisms (Puertolas et al. 2016). In soft plant tissues, as the mesocarp or pericarp of most fruits, electric field strengths between 0.1 and 10 kV/cm were enough to maximize PEF-induced cell membrane permeabilization. Higher electric fields up to 20 kV/cm were appropriate for effective permeabilization of hard vegetable materials, as grape or sesame seeds (Puertolas et al. 2016). PEF accelerated trans-membrane potential due to charging process at the membrane interfaces (Vorobiev and Lebovka, 2008). When an electrical field was connected, it induced in reversible and irreversible electroporation at the membrane. Reversible electroporation was adequate for biotechnological and medical applications. Meanwhile irreversible electroporation was suitable for improving disintegration and permeabilization processes. The effectiveness of PEF to irreversible permeabilization of cell membranes depended on process parameters, food characteristics and cell characteristics (Puertolas et al. 2012; Vorobiev and Lebovka 2008). Electric field strength was believed the key process variable. Treatment time was the second most important process parameter which defined the PEF efficiency. Objective of our study investigated the effectiveness of electric field strength and treatment time on carotenoid content, polyphenol oxidase activity, ascorbic acid content and total soluble solid content of papaya juice.

MATERIALS AND METHODS

Material

Ripen papaya fruits were harvested from SocTrang province, Vietnam. After collecting, they were rinsed in clean water with 10 ppm peracetic acid for sanitation. The fruits were peeled to get pulp out, mashed and filtered to get the juice. Chemical reagents were all analytical grade.

Researching method

Lab-scale pulsed electric field was used for treatment of papaya juice. The 1.2÷1.8 kV/cm electric field strengths with 2÷10 ms duration were investigated for papaya juice. The juice was also pasteurized at 85^{o}C for 2 minutes as control. The control and treated samples were kept at ambient temperature for 15 weeks. In 3 week-interval, sample was taken to examine carotenoid content, polyphenol oxidase activity, vitamin C and total soluble solid.

Physicochemical determination

Carotenoid content (µg/L) was quantified by high-performance liquid chromatographyfollowing the methodology reported by Khachik et al. (1992). Polyphenol oxidase activity (U/ml/min) was determined by spectrophotometer according to the method ofZhang et al., (2018). Vitamin C (mg/100 ml) was determined by using a 2,6-dichlorophenol indophenol visual titration method. Total soluble solid (^{o}Brix) was measured by hand-held refractometer.

Statistical analysis

The experiments were run in triplicate with different groups of samples. The data were presented as mean±standard deviation. Statistical analysis was performed by the Statgraphics Centurion version XVI..

RESULTS AND DISCUSSION

Carotenoid

Carotenoid was a useful pigment in a great number of fruits and vegetables. The carotenoid content of papaya juice under electric field strength (kV/cm) and treatment time (ms) was clearly presented in table 1. There was a significant increase in carotenoid content in papay juice treated with different electric field strengths and durations as compared to control treatment. Similar results were reported on orange juice (Cortes et al. 2006), orange–carrot juice (Torregrosa et al. 2005), grapefruit juice (Ranaet al.2015), tomato juice (Vallverdu-Queralt et al. 2013), carrot puree (Shahin et al. 2014), date palm extract (Azhari et al. 2019) treated by PEF. The significant increase in carotenoid content on PEF could be due to the distraction of cell walls, which improved free carotenoid in the juice.

Polyphenol oxidase (PPO)

Polyphenol oxidase was mainly located in the chloroplast of thylakoid membranes while phenolics were existed in vacuoles (Spagna et al. 2005). Browning phenomenon occured naturally due to reaction of the enzyme polyphenoloxidase in the existing of oxygen on phenolics and inducing in a brown substance known *o*-Quinones (Al-Amrani et al. 2020). *o*-Quinones polymerized nonenzymatically to produce heterogeneous deep dark polymers called melanin (Li et al. 2016). Polyphenoloxidase operated as a promoter for peroxidase activity because hydrogen peroxide which was a product of polyphenoloxidase reaction with phenolics was essential for peroxidase action. Peroxidase catalyzed oxidation of phenolics in the presence of hydrogen peroxide to form brown compounds (Tomas-Barberan and Espin, 2001). PPO activity was significantly decreased by PEF treatment compared with that of the control. The lowest PPO activity was noticed at electric field strength 1.4 kV in 6 milliseconds (table 2). Electrical fields might provoke a deformation or structural modification of a protein due to the interaction between the external electric field and the functional groups of the protein inducing its unfolding (Elez-Martinez et al., 2006; Freedman et al., 2013). Giner et al. (2001) found that the residual activity of apple PPO decreased by up to 3.15% at 24.6 kV/cm for 6 ms of treatment time, whereas the maximum inactivation obtained for pear PPO was up to 38.0% at 22.3 kV/cm for the same treatment time. Liang et al. (2006) found a significant decrease (33%) in PPO activity in freshly squeezed apple juice when preheated at 50◦C and treated with PEF at 27 kV/cm for 58.7 µs. Schilling et al. (2008) achieved a maximum PPO deactivation of 48% by PEFtreated at 30 kV/cm. Sanchez-Vega et al. (2009) realized a 70% reduction of residual PPO activity in apple juice using PEF treatment at 38.5 kV/cm and 300 pulses per second. Ertugay et al. (2013) proved that PPO activity became completely inactive at 40 kV/cm. Wendy et al. (2014) optimized the combined pulsed electric fields and mild temperature processing conditions for red apple juice polyphenol oxidase inactivation. Treatment perforemd at 30 kV/cm, 1000 µs and 60°C led to red apple juice with the lowest residual PPO enzyme activity. Cincia et al. (2019) concluded that PPO inactivation was more effective by a combination of temperature and PEF.

Ascorbic acid

Ascorbic acid was water soluble vitamin essential for human health (Gallie, 2011; Carr and Vissers, 2013). It's thermally labile, pH-, metal-ion-, and light-sensitive, and degradable by ascorbic acid oxidase (Uddin et al., 2001; Cruz et al., 2006). Ascorbic acid retention was a vital variable nutrients in fruit storage (Lin et al., 1998; Marfil et al., 2008). Consumers were depended on fruits and vegetables in their diet to supply ascorbic acid (Fenoll et al., 2011; Shashirekha et al., 2015). It's beneficial for its antioxidant power and stimulation of immune system (Davey et al., 2000; Cui et al., 2011; Valente et al., 2011). In our research, the pulsed electric field showed the significant retention of ascorbic acid content compared to the conventional thermal treatment (control). Ascorbic acic in the PEF-papaya juice was stable during storage. Meanwhile there was a dramatically decreased of vitamin C in control sample during 15 weeks of storage (table 3). Ade-Omowaye et al. (2003) reported that the ascorbic acid retention of red bell pepper after PEF treatment (50 pulses at 2 kV/cm, pulse duration 400 ms) ranged from 89.6% to 96.5%. Odriozola-Serrano et al. (2009) concluded that the ascorbic acid retention of strawberry juice was 98% after high intensity pulse electric field (HIPEF) (35 kV/cm, 1000 µs, and monopole mode). Marselles-Fontanet et al. (2013) reported that retention of ascorbic acid in grape juice was higher after PEF treatment than after thermal treatment. Zhang et al. (2010) found that the longan juice treated by PEF at an intensity of 32 kV/cm retained greater amounts of vitamin C than thermally treated longan juice. Zhi-Hong et al. (2015) indicated that the PEF treatment didn't cause damage to ascorbic acid.

Total soluble solid

In our research, the pulsed electric field showed the significant retention of total soluble solid content compared to the conventional thermal treatment (control). Ascorbic acic in the control and PEF-papaya juice was stable during storage (table 4). Rivas et al. (2006) indicating negligible change in total soluble solid of PEF processed blended orange and carrot juice. Longan juice was treated by a pulsed electric field (PEF) with an intensity of 32 kVcmand compared with a conventional thermal pasteurization method. Total soluble solids was not statistically significant difference between the untreated and both thermally pasteurized and PEF-treated samples (Zhang et al. 2010). Blueberry juice treated by PEF at 35 kV/cm and 82 µs had no significant change in total soluble solid compared to heat sterilization (Jianet al.2014).

Table 1: Carotenoid content (µg/L) of papaya juice by electric field strength (kV/cm) and treatment time (milliseconds)

Storage (weeks)	Control	Electric field strength (kV/cm)/ Treatment time (milliseconds)				
		1.0/10	1.2/8	1.4/6	1.6/4	1.8/2
3	17.31±0.01^{d}	20.68±0.02^{c}	24.31±0.00^{b}	27.59±0.03^{a}	25.93±0.01^{ab}	22.57±0.00^{bc}
6	14.82±0.03^{d}	19.32±0.00^{c}	23.66±0.02^{b}	27.03±0.00^{a}	25.24±0.02^{ab}	22.05±0.03^{bc}
9	12.24±0.00^{d}	18.69±0.01^{c}	23.02±0.03^{b}	26.54±0.01^{a}	24.86±0.00^{ab}	21.64±0.01^{bc}
12	11.77±0.02^{d}	18.01±0.02^{c}	22.54±0.01^{b}	26.11±0.02^{a}	24.17±0.03^{ab}	21.13±0.02^{bc}
15	10.03±0.01^{d}	17.52±0.00^{c}	22.06±0.03^{b}	25.72±0.00^{a}	23.69±0.01^{ab}	20.78±0.00^{bc}

Note: the values were expressed as the mean of twenty two samples; the same characters (denoted above), the difference between them was not significant (α = 5%).

Table 2: Polyphenol oxidase activity (U/ml/min) of papaya juice by electric field strength (kV/cm) and treatment time (milliseconds)

Storage (weeks)	Control	Electric field strength (kV/cm)/ Treatment time (milliseconds)				
		1.0/10	1.2/8	1.4/6	1.6/4	1.8/2
3	0.0057±0.0001^{a}	0.0036±0.0003^{b}	0.0025±0.0000^{c}	0.0019±0.0002^{d}	0.0022±0.0003^{cd}	0.0030±0.0000^{bc}
6	0.0052±0.0000^{a}	0.0031±0.0002^{b}	0.0021±0.0001^{c}	0.0016±0.0000^{d}	0.0018±0.0001^{cd}	0.0025±0.0002^{bc}
9	0.0047±0.0003^{a}	0.0027±0.0000^{b}	0.0017±0.0003^{c}	0.0013±0.0001^{d}	0.0015±0.0002^{cd}	0.0021±0.0001^{bc}
12	0.0041±0.0002^{a}	0.0022±0.0001^{b}	0.0012±0.0002^{c}	0.0009±0.0000^{d}	0.0011±0.0001^{cd}	0.0017±0.0003^{bc}
15	0.0036±0.0001^{a}	0.0017±0.0000^{b}	0.0008±0.0001^{c}	0.0006±0.0002^{d}	0.0008±0.0000^{cd}	0.0012±0.0001^{bc}

Note: the values were expressed as the mean of twenty two samples; the same characters (denoted above), the difference between them was not significant (α = 5%).

Table 3: Ascorbic acid content (mg/100 ml) of papaya juice by electric field strength (kV/cm) and treatment time (milliseconds)

Storage (weeks)	Control	Electric field strength (kV/cm)/ Treatment time (milliseconds)				
		1.0/10	1.2/8	1.4/6	1.6/4	1.8/2
3	12.51±0.02^{d}	16.45±0.01^{c}	18.06±0.03^{b}	21.13±0.02^{a}	20.57±0.03^{ab}	17.19±0.01^{bc}
6	10.43±0.01^{d}	16.32±0.03^{c}	17.93±0.00^{b}	21.02±0.01^{a}	20.34±0.00^{ab}	17.03±0.02^{bc}
9	8.67±0.03^{d}	16.21±0.00^{c}	17.82±0.01^{b}	20.89±0.00^{a}	20.17±0.01^{ab}	16.87±0.03^{bc}
12	7.10±0.00^{d}	16.07±0.01^{c}	17.71±0.02^{b}	20.63±0.03^{a}	20.06±0.02^{ab}	16.62±0.00^{bc}
15	6.02±0.02^{d}	15.98±0.03^{c}	17.60±0.00^{b}	20.51±0.01^{a}	19.85±0.00^{ab}	16.41±0.01^{bc}

Note: the values were expressed as the mean of twenty two samples; the same characters (denoted above), the difference between them was not significant (α = 5%).

Table 4: Total soluble solid content (ºBrix) of papaya juice by electric field strength (kV/cm) and treatment time (milliseconds)

Storage (weeks)	Control	Electric field strength (kV/cm)/ Treatment time (milliseconds)				
		1.0/10	1.2/8	1.4/6	1.6/4	1.8/2
3	10.27±0.00^{d}	14.80±0.03^{c}	16.03±0.02^{b}	17.83±0.03^{a}	16.45±0.03^{ab}	15.29±0.00^{bc}
6	10.22±0.03^{d}	14.73±0.01^{c}	15.97±0.00^{b}	17.79±0.02^{a}	16.37±0.02^{ab}	15.23±0.03^{bc}
9	10.15±0.01^{d}	14.69±0.02^{c}	15.90±0.03^{b}	17.72±0.01^{a}	16.30±0.03^{ab}	15.17±0.01^{bc}
12	10.08±0.02^{d}	14.61±0.00^{c}	15.83±0.01^{b}	17.65±0.00^{a}	16.24±0.01^{ab}	15.11±0.02^{bc}
15	10.03±0.00^{d}	14.54±0.01^{c}	15.76±0.00^{b}	17.58±0.03^{a}	16.19±0.02^{ab}	15.04±0.00^{bc}

Note: the values were expressed as the mean of twenty two samples; the same characters (denoted above), the difference between them was not significant (α = 5%).

Mango nectar was subjected to pulsed electric fields with pulse frequency (70-120 Hz) and pulse width (15- 24 µs). The total soluble solids of mango nectar did not showed much change after PEF processing (Kumar et al. 2015).

CONCLUSION

Pulsed electric field (PEF) was considered a friendly gentle technology with multiple application fields in replacement to conventional physical techniques. The impact of pulsed electric field in different electric field strength and treatment time to the quality properties of papaya fruit juice was verified. The electric field strength and treatment time significantly improved papaya juice quality properties during production as well as better retention of these values during storage. Pulsed electric field could be used for other underutilized crops to inhance their economic value.

CONFLICT OF INTEREST

The authors declared that present study was performed in absence of any conflict of interest.

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AUTHOR CONTRIBUTIONS

Nguyen Phuoc Minh arranged the experiments and also wrote the manuscript.

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