Setup of A Continuous Pulsed Electric Field System for Microbial Reduction in Some Liquid Foods

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ABSTRACT---- Pulse electric field (PEF) has been used as a non-thermal process in food production. In this study, a PEF system was set up to use for microbial reduction in some liquid foods. The PEF system comprised a stainless steel chamber, a pulse generator and a pivotal system with auxiliary devices. The set up PEF was evaluated for the specific electro-rheological characteristics in four different kinds of the liquid foods. It was found that the set up PEF had the volume of the treatment zone at 18.85 mL, the length of the entrance zone at 40 mm. At the flow rate of 500 ml/min, the hydraulic diameter was 10 mm. There were differences in the rheological properties and conductivity that provided the electrical resistance of 0.318, 0.692, 0.936 and 0.286 Ω for orange juice, durian juice, mangosteen juice and coconut water, respectively. The set up PEF was significantly effective at 20 - 40 KV for microbial reduction in the coconut water, the durian juice and the mangosteen juice, except for orange juice. In addition, two-round use of the PEF was effective to reduce the microbial contamination in the model of durian juice. The results suggested that the set up PEF would be used for the microbial reduction in the fruit juices and two-round of the PEF-treatment cycles may provide for more effective.

Keywords--- pulsed electric field, microbial reduction, non-thermal processing, liquid foods

1. INTRODUCTION

Recently, consumers have been demanding for fresh-like products with concerning on the nutritional quality and microbiological safety. The products with the original taste, high nutritional values, and abundant of bioactive compounds are attractive to the consumers. The fresh fruit or vegetable derived products are rich in vitamin C, vitamin E, β -carotene, and phenolic compounds [1,2]. However, the products are required the pasteurization to prolong their shelf life. The thermal processes such as ultrahigh temperature and high temperature short time process can do the microbial inactivation. The processes could affect the nutritional qualities, some organoleptic properties, and the stability of the bioactive compounds [1,2,3]. Alternatively, the non-thermal processes such as pulse electric field (PEF), ohmic heating, ultrasound, and high-pressure technique have been innovated for microbial inactivation, which might minimally deteriorate to change the organoleptic characteristics and nutritional qualities of the food products [2,3]. The PEF is an interested non-thermal process that has been used to treat many kinds of biological materials [4]. The PEF affects to the microorganisms by electroporation. The microbial lethal effect of the PEF depends on the intensity of the electric field and the retention time of pulsing. In addition, the intrinsic properties of the liquid foods such as pH, components, dielectric, conductivity, and rheological properties of the food materials, are also influencing on the electrical breakdown of the microbial cells [4,5].

In general, a PEF system should be consisted of a pulse generator, treatment chamber(s), a fluid-handling system, a monitoring system, and auxiliary devices. The PEF chamber is one of the key components in the PEF pasteurization, which composed of an anode and a cathode electrode held in position by insulating material to form an enclosed system. In the design of a treatment chamber, the pivotal factors should be taken to be account including: (i) the uniform of the generated electrical field, (ii) the rate of ohmic heating due to the electrolysis, (iii) the electrical properties of the liquid food (e.g. conductivity, electrical resistance, etc.), (iv) the fluid dynamic during the liquid flows through the chamber, (v) the attained

voltage, and (vi) the safety and the effectiveness in use [2,5,6]. The PEF chamber could be designed as parallel plate, centric cylinder, or co-field configurations. Almost of the homogenous liquid food should be homogeneously treated with PEF [4]. The PEF with high intensities has been used for microbial reduction in some liquid foods such as orange juice, apple juice, carrot-orange juice mix, berry juice, milk, and liquid egg [7,8,9,]. The PEF application was demonstrated for inactivation of some pathogens (e.g. *Escherichia coli, Samonella,* and *Staphyllococus aureus*), yeasts, and molds. In several, food processing accepts for a three or more log reduction of microbial count. Therefore, at least 20 kV/cm of the electrical field intensity is required [10,11].

This research aimed to set up and to evaluate the effectiveness of the design PEF system for microbial reduction in some liquid foods with varied in their electrical and rheological properties.

2. MATERIALS AND METHODS

2.1 Sample preparations

Coconuts (*Cocos nucifera* L.), durians (Mon Thong) (*Durio Kutejensis*), oranges (*Citrus reticulate* Blanco) and mangosteens (*Garcinia mangostana* L.) were purchased from in Chanthaburi Province, Thailand. Coconut water and orange juice were freshly prepared and kept at 4°C until used within 1 h. The durian and mangosteen flesh were freshly isolated and immediately kept in -20°C until used within 3 months.

2.2 Design of the PEF continuous system

A continuous PEF system was designed in a laboratory scale for microbial reduction in liquid foods. The PEF system consisted of a treatment chamber (electrode chamber), an electric pulse generator, a liquid flow system, and accessory devices.

2.3 Determination of the PEF capacities

The set up PEF was determined for the range of flow rate, frequency of pulsing and the electrical voltage. The flow rate was monitored at the inlet and outlet sides by using flow meters and a cylinder-conventional method. The electrical pulsing intensity (frequency) and the voltage were measured using a TBS1064 oscilloscope (Tektronix, USA).

2.4 Specific characteristics of the PEF system

The specific electro-rheological characteristics of the PEF system were tested in four different kinds of liquid foods. The PEF chamber was determined and calculated for the gap between the electrodes (d), electrode width (W_E), electrode surface area (S), electrode path length (L), volume of the treatment zone (v_t), the length of the entrance zone (L_E), the flow rate of the food (F_f) and the hydraulic diameter (D_h) (De Vito, 2006). The density and viscosity of the liquid foods were measured by a DMA 4500 Density meter (Anthon Parr, USA) and a DVII⁺-Pro viscometer (Brookfield, USA), respectively. The conductivity was determined by using an Acumet pH-Conductivity meter (Fisher Scientific, USA). Furthermore, the electrical resistance of the liquid foods in the same PEF system was calculated following the mathematical relation described by De Vito [2]. The specific electrical and rheological properties were determined for the set up PEF system. The electrical intensity (E) of the PEF chamber would be directly determined by the distance (d) between the coupled electrodes with a constant voltage (V) according to the equation 1 [2].

$$E = \frac{V}{D} \tag{1}$$

The electrical resistance (R_c) of the liquid food is also influencing on the effectiveness for microbial reduction in the liquid foods. The R_c is related to the gap between the electrodes, and inversed to the electrode surface area (S) and the electrical resistance (σ_f) (equation 2).

$$R_c = \frac{d}{SS_f} \tag{2}$$

The wetted perimeter (W_p) and the pivot area (A_r) are specific to the PEF pivotal and related to the hydraulic diameter of the pivot dimension (D_h) (equation 3). The rheological properties including the density (ρ) , the viscosity (η) and the rate of fluid flow (F_f) of the liquid foods are also determiners for the rheological behavior of the liquid foods that have usually been explained in the term of Reynold's number (Re)(equation 4). In addition, the length of entrance (L_E) can be derived from the Re and D_h value (equation 5)[2].

$$D_h = \frac{4A_r}{W_p} \tag{3}$$

$$\operatorname{Re} = \frac{F_f \Gamma_f D_h}{m} \tag{4}$$

$$L_E = 0.028 Re D_h \tag{5}$$

2.5 Evaluation of the set up PEF system for microbial reduction

One liter of sample was mixed with 3 L of sterile distilled water before subjected to the PEF system. The treatments were undergone at the electrical voltages of 20 and 40 kV at 30 kHz and the flow rate of 1 L/min. Subsequently, the liquid food samples were decimal diluted in the sterile 0.85% NaCl before spreading on plate count agar (PCA) medium and cultivated at 37°C for 12-24 h [12].

2.6 PEF-circulating cycle on microbial reduction

The durian juice was used as the model to elucidate the number of the PEF–circulating cycle on the microbial reduction. Two-hundred grams of the frozen durian flesh was blend in a sterilized wire blender with addition of 250 ml sterile water, and filtered through a three layers of sterile cloth sheets. Afterwards, the juice was mixed with 3 L of the sterile water. Thereafter, the sample was subjected to the PEF chamber at the minimum efficient condition for microbial reduction (1.0 L/min of the flow rate, 30 kHz of the frequency and 10 kV of the applied voltage). The sample was tested for 0 (control), 1, 2, 3 and 4 rounds of the treating cycle before determination of the viable count on the PCA [12]. The PEF was washed twice with 5 min circulation of 4 L hot water.

2.7 Statistical Analysis

The analysis of variances (ANOVA) were tested for the significant difference at P<0.05 of the triplicate determinations using SPSS packaging program (version 11.5).

3. RESULTS AND DISCUSSION

3.1 Set up of the continuous PEF process

In this study, the PEF was design as a vertical continuous process as shown in Fig. 1a. The liquid food was firstly transferred to the electrode chamber equipped with an automatic controllable pivotal line. The solenoid valve and pump with flow meter were installed for controlling and to ensure the uniform flowing of the liquid foods during the PEF process. The electrode chamber was the parallel plate of the pulsed electric field generator. The draw of the PEF chamber design in each parts and orientations was shown in Fig. 1b. A polyamide spacer was used to insulate the cathode and anode electrodes. The electrodes were connected to a pulse generating circuit to form the pulse shape, frequency, and intensity (Fig. 2).

3.2 The PEF limitations

The set up PEF had a range of the electrical capacity as shown in Table 1. The PEF system could be adjusted to a frequency in the range of 10 - 30 kHz that relevance to the pulsing generating circuit of the astable circuit. The voltage would be applied to the PEF chamber in the range of 10 - 40 kV. The voltage supply could also be determined by the space of the

electrodes in the treatment chamber. In this study the flow rate was held on the continuous flow throughout the flow of water in the pipe (0.5-1.0 L/min).

Table 1 Specific characteristics of the parallel plate chamber of the set up PEF an astable pulsing circuit

Characteristic	Valu	ie	Characteristic	Val	ue
Gap between the electrodes, d	5	mm	The length of the entrance zone, L_E	40	mm
Electrode width, W_E	15	mm^2	Hydraulic diameter, D_h	10	mm
Electrode surface area, S	3,142	mm	The flow rate of the food, F_f	0.5 - 1.0	L/min
Electrode length, L	40	mm	Range of the frequency (apparent)	10 - 30	kHz
Volume of the treatment zone, v_t	6,283	mm^3	Voltage supply	10 - 40	kV/cm

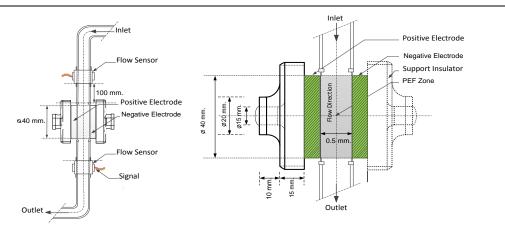


Fig. 1. Schematic of the PEF pivotal line in a cross section (a) and the PEF chamber (b).

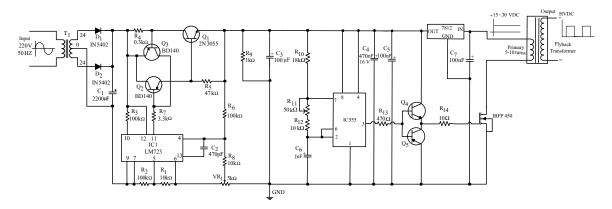


Fig. 2. The pulse generating circuit (astable circuit)

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3.3 PEF on microbial reduction in some liquid foods

Table 1 shows the characteristics of the gap between the electrodes, electrical width, electrode surface area and the electrode length to determine the treatment zone of 18.85 ml with the length of the entrance zone of 40 mm, thus provided the chamber with the hydraulic diameter of 10 mm in the case of the flow rate at 500 ml/min (Table 2). The rheological properties of the liquid foods were density of 1.09, 071, 1.15 and 0.72 kg/L, and the viscosity of 28.05, 336.00, 31.15 and 13.32 cP for orange juice, durian juice, mangosteen juice and coconut water, respectively.

Characteristic	Liquid food					
	Orange juice	Durian juice	Mangosteen juice	Coconut water		
Electrical property						
- Conductivity (mS/cm)	$5.00 \pm 0.08^{\circ}$	2.30 ± 0.01^{b}	1.70 ± 0.02^{a}	$5.57 \pm 0.05^{\circ}$		
- Electrical resistance, Ω	0.318	0.692 0.936		0.286		
Rheological property						
- Density (kg/L)	1.09 ± 0.01^{b}	0.71 ± 0.01^{a}	1.15 <u>+</u> 0.03 ^c	0.72 ± 0.04^{a}		
- Viscosity (cP)	28.05 ± 1.01^{b}	36.00 <u>+</u> 8.98 ^c	1.15 ± 0.47^{b}	13.32 <u>+</u> 0.34 ^a		
At the flow rate 500 ml/min						
- Reynolds number, Re	1,239	67	1,173	1,723		
- Length of entrance, LE (mm)	1.8	0.1	1.7	2.5		
At the flow rate 1,000 ml/min						
- Reynolds number, Re	2,478	135	2,346	3,446		
- Length of entrance, LE (mm)	3.6	0.2	3.4	5.0		

Table 2 Specific electrical and rheological characteristics of the liquid foods

Mean+S.D. from triplicate determinations

The different superscripts in the same row (a-c) denoted the significances (P < 0.05).

The set up PEF chamber was used to reduce the microbial contaminated in some liquid foods. The efficiency of the PEF on the microbial lethal was varied. Durian juice with the initial microbial content of 5.50 log CFU/mL could be reduced to 5.02 and 4.78 log CFU/mL by the PEF process in the frequency of 30 kHz at the voltage of 20 and 40 kV, respectively (Table 3). The PEF-treated coconut water was also effective to reduce the microbial load of 0.96 and 1.48 log CFU/mL at 20 and 40 kV, respectively. Moreover, the set up PEF also had a reduction potential on the microbial contaminated in mangosteen juice of 0.96 and 1.92 log CFU/mL at 20 and 40 kV, respectively. However, the PEF had no reductive effect on the microbial count in the orange juice. The set up PEF might be potentially used to reduce the microorganisms in some liquid foods. However, the PEF efficiency testing was done in one loop of PEF treating. The number of PEF treating cycle should be done further.

	Microbial number (log CFU/mL)				
Sample	Control [†]	PEF^{\ddagger}			
	Control	20 kV	40 kV		
Coconut water	4.32 ± 0.08^{Aa}	3.36 <u>+</u> 0.37 ^{Ab}	2.84 ± 0.58^{Ab}		
Durian juice	5.50 ± 0.15^{Bc}	$5.02\pm0.02^{\mathrm{Bb}}$	$4.78\underline{+}0.01^{ABa}$		
Mangosteen puree	7.77 ± 0.51^{Cc}	6.81 ± 0.56^{Cb}	$5.85\underline{+}0.04^{\mathrm{Ba}}$		
Orange juice	4.06 ± 0.40^{Aa}	3.66 ± 0.45^{Aa}	4.04 ± 0.40^{ABa}		

Table 3 Effect of PEF on the microbial reduction in some liquid foods

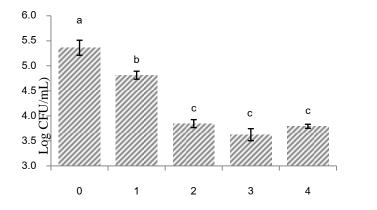
Mean+S.D. from triplicate determinations

The different superscripts in the same row (a-c) and column (A-C) denoted the significances (P < 0.05).

[†]Control is the PEF-untreated samples were measured after final volume adjustment.

[‡]The PEF was run at the frequency of 30 kHz and the flow rate of 1 L/min.

The PEF treatment was evaluated for the microbial reduction using the durian juice as the model. In the test, the sample was treated at 10 kV of voltage with a frequency of 30 kHz and a flow rate of 1 L/min. The testing condition was tried at the lowest efficiency on microbial reduction by the set up PEF system. The initial microbial content of the durian juice was 2.3×10^5 cfu/ml (Fig. 3). Only one round of the PEF treatment, approximately 1.0 log CFU of the microbial number was reduced while the re-circulating of the PEF treatment was more effectively on the microbial reduction. However, the PEF treatment was not increased for further circulation. It could be suggested that the number of circulating cycle of the durian juice passing through the PEF chamber was two cycles under the provided condition.



Circulating cycle (rounds)

Fig. 3. The number of PEF treatment cycles on microbial survival in durian juice.

4. CONCLUSION

The set up PEF was successful to be use as a machine for microbial reduction in the liquid foods. The electro-rheological characteristics of the attained liquid should be taken in to account for the PEF system design. The optimal configuration of the PEF geometric should be more deeply studies for more effective. The number of PEF-circulating cycles might also be put on the further study.

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6. **REFERENCES**

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