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Synergistic Effect of Slightly Acidic Electrolyzed Water and Ultrasound at Mild Heat Temperature in Microbial Reduction and Shelf-Life Extension of Fresh-Cut Bell Pepper

Ke Luo and Deog-Hwan Oh*

Department of Food Science and Biotechnology and Institute of Bioscience and Biotechnology, Kangwon National University, Chuncheon 200-701, Republic of Korea

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*Corresponding author Phone: +82-33-250-6457; Fax: +82-33-241-0508; E-mail: deoghwa@kangwon.ac.kr

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Introduction

electrolyzed water (SAEW), ultrasound (US), or mild heat (60°C)) on the growth of *Listeria monocytogenes* and *Salmonella enterica* serovar *Typhimurium* in fresh-cut bell pepper, and the shelf-life and sensory quality (color and texture) were followed during storage at 4°C and 25°C. An additional 0.65, 1.72, and 2.70 log CFU/g reduction was achieved by heat treatments at 60°C for 1 min for DW, SAEW, and SAEW+US, respectively. Regardless of the type of pathogen, the combined treatment (SAEW+US+60°C) achieved a significantly (p < 0.05) longer lag time in all treatment groups. This combined treatment also prolonged the shelf-life of bell pepper up to 8 days and 30 h for the storage at 4°C and 25°C, respectively. There was also no significant difference in the color and hardness of treated (SAEW+US+60°C) bell pepper from that of control during the storage. This new hurdle approach is thus expected to improve the microbial safety of bell peppers during storage and distribution.

The objectives of this study were to evaluate the effect of combined treatments (slightly acidic

Keyword: Fresh-cut bell pepper, slightly acidic electrolyzed water, *Listeria monocytogenes*, *Salmonella enterica* serovar *Typhimurium*, ultrasound

Fresh-cut vegetables are an essential part of the human diet and there has been an increasing demand for their consumption in recent years owing to their health effects and changes in people's lifestyles [1]. However, products such as fresh-cut or minimally processed vegetables that have been associated with the outbreak of foodborne illness were commonly contaminated by foodborne pathogens during the various farming or post-harvest stages [2, 24]. Bell pepper is included in the five crops that are regarded as the greatest economical importance in most European and North American countries [35], and more consumers are willing to eat raw bell pepper as salads [9]. It has been reported that fecal coliform has been found in 15% and 85%in each 40 of packed bell pepper and freshly harvested bell pepper, respectively [20], reflecting that bell pepper has a high potential to be contaminated by pathogenic and

food spoilage microorganisms. Among the pathogenic microorganisms, *Listeria monocytogenes* and *Salmonella enterica* are major concerns in terms of annual deaths and hospitalizations [4]. Moreover, bell pepper has been reported to be recalled as a result of the detection of *L. monocytogenes* and *Salmonella* on this product [11, 21]. Therefore, the control and prevention of foodborne pathogens and food spoilage microorganisms in raw bell pepper are a big challenge in the food industry.

Dipping or rinsing of vegetables associated with chemical compounds, such as chlorine, are methods widely applied to reduce the number of pathogenic or spoilage microorganisms on the surface of minimally processed vegetables in order to improve the food safety and to extend the shelf-life of the products. However, some of the chemical compounds, such as the inorganic chlorine compounds, are prohibited in European countries owing to the potential formation of harmful by-products that can damage the environment. Slightly acidic electrolyzed water (SAEW), a novel sanitizer that has a high antimicrobial effect due to the presence of a high concentration of hypochlorous acid (HOCl) [8, 25], has been increasingly applied and studied during the last decade around the world. Owing to the low available chlorine in SAEW, the sanitizer enables to reduce the corrosion of surfaces of fresh products and minimize the potential damage to human health and the environment. It has been reported that electrolyzed water combined with mild heating was significantly more effective in reducing the number of unwanted organisms on food [18, 19, 25].

Ultrasound, a form of energy generated by sound waves, is regarded as an environmentally friendly antimicrobial agent in the food industry. It has been recommended as an alternative sanitization step in the fresh produce industry. A single process of ultrasound treatment does not have a significant impact on decreasing high-load microbial contamination [27]. Therefore, further studies for improving the bactericidal efficiency via combination with a sanitizer or another physical method, such as mild heating, were necessary. It is documented that the extension of shelf-life of fresh-cut produce depends on the washing step [33]. Until now, there has been no report of the application of SAEW or ultrasound to extend the shelf-life of fresh bell pepper. The aim of this study was to evaluate the effects of SAEW combined with ultrasound and mild heat on the microbial safety, shelf-life, and sensory quality of bell pepper.

Materials and Methods

Strains

The three strains of Listeria monocytogenes (ATCC 19114, ATCC 19116, ATCC 19118) and two strains of Salmonella enterica serovar Typhimurium (ATCC 14028, ATCC 26926) used in this study were obtained from the Department of Food Science, University of Georgia (Griffin, GA, USA). For preparation of the inoculum, each suspension (0.1 ml) of the stock cultures was individually transferred into 10 ml of Tryptic Soy Broth (TSB; Becton Dickinson Diagnostic Systems, Sparks, Maryland, USA) and incubated at 37°C for 24 h. The cultures in the late stationary phase were harvested by centrifugation (3,000 × g, 10 min, 4°C) and the pellets were then washed, resuspended in 10 ml of 0.1% sterile peptone water (PW; Difco, MD, USA) to make a final cell concentration of approximately 9.0 log CFU/ml. Each strain was mixed to construct L. monocytogenes and S. Typhimurium culture cocktails. The cocktails were used in the following experiments. The cell count of the mixed culture was confirmed by plating 0.1 ml portions of appropriately diluted cocktail onto TSA plates and incubating the plates at 35°C for 24 h.

Inoculation of Samples

Fresh bell peppers purchased from a local retail store in Chuncheon, South Korea were aseptically transported to the laboratory within 2 h. The external surface of sliced samples ($10 \pm 0.2 \text{ g}$, $3 \times 3 \text{ cm}$) were separately inoculated by the two types of inoculum by depositing droplets at 20 locations using a micropipettor. Initial populations of the two pathogens on the bell pepper samples were approximately 5 log CFU/g. The inoculated samples were aseptically dried in a laminar flow hood for 30 min to guarantee sufficient attachment of the bacteria on the samples, and then immediately exposed to sanitizing treatments.

Preparation of SAEW

The SAEW (pH: 5.0–5.2; oxidation reduction potential (ORP): 930–950 mV; and available chlorine concentration (ACC): 28– 30 mg/l) was produced by electrolysis of a diluted hydrochloric acid (6% HCl) in a chamber without membrane, using an electrolysis device (BC-360; Cosmic Round Korea Co. Ltd., Seongnam, Korea) at a setting of 2.5 A and 22.8 V. The pH and ORP of the SAEW solution were determined by a dual-scale pH meter (Accumet model 15; Fisher Scientific Co., Fair Lawn, NJ, USA) equipped with bearing pH and ORP electrodes. The ACC was determined by a colorimetric method using a digital chlorine test kit (RC-3F; Kasahara Chemical Instruments Corp., Saitama, Japan). After production, the SAEW was stored in polypropylene containers until use.

Experimental Procedure

The samples were divided into three treatment groups and control (only dipping in deionized water) at room temperature $(23 \pm 2^{\circ}C)$. For the combined treatments, the samples immersed in SAEW were placed in a sterile bench-top ultrasonic cleaner (JAC-4020; KODO Technical Research Co., Ltd., Hwaseong, Gyeonggido, South Korea) at a fixed frequency of 40 kHz and acoustic energy density (AED) of 400 W/L [12]. The optimized combination treatment of SAEW and ultrasound at mild heating temperature was based on our previous work that optimized the antimicrobial effect of slightly acidic electrolyzed water in L. monocytogenes and S. Typhimurium on bell pepper by combining ultrasound with mild heat (data not shown). All sanitizing treatments were performed at a dip time of 1 min. Following each sanitizing procedure, SAEW was removed and the tank was sterilized by ultraviolet light irradiation for 10 min, and then serially rinsed with 70% ethanol and left to dry. Group 1 and 2 samples were dip treated in DW and SAEW, respectively, at 60°C (DW+60°C; SAEW+60°C). Group 3 was dip treated in SAEW with ultrasound at 60°C (SAEW+US+60°C). All independent trials were replicated three times.

Shelf-Life Study

After sanitizing, the heat-treated samples were immediately placed into bags containing BPW (4°C) for 1 min in order to terminate the thermal inactivation. The samples were then

aseptically placed in stomacher bags (Nasco Whirl-pak, Janesville, WI, USA) and isothermally stored at 4°C and 25°C. For the enumeration, the samples at appropriate sampling time were immediately mixed with 90 ml of 0.1% sterile BPW, and then pummeled for 2 min (Lab-blender 400; Seward, London, UK). After pummeling, aliquots of the homogenate were serially diluted in BPW and aliquots (0.1 ml) were plated on Oxford medium base agar (Difco) supplemented with modified Oxford antimicrobic supplement (Difco), SS agar (Difco), and plate count agar (Difco) to enumerate L. monocytogenes, S. Typhimurium, and total bacterial counts (TBC), respectively. The populations of targeted pathogens were counted after incubation of the agar plates at 37°C for 24 h, and expressed as log CFU/g. Each experiment was replicated three times, and means of microbial populations (log CFU/g) from different storage temperatures were calculated. The end of the shelf-life arrived when the population of a group of microorganism or the sensory quality reached an unacceptable level.

Model Development

The observed data of the pathogen growth on bell pepper stored at 4°C and 25°C were fitted to the Baranyi model (Eq. (1)) [3] to obtain the kinetic parameters of specific growth rate (SGR [log CFU/h]) and lag time (LT [h]). The growth curves were fitted using the DMFit Add-In software in Excel, Institute of Food Research, Norwich, UK.

$$Y(t) = Y_0 + \mu A(t) - \frac{1}{m} ln \left\{ 1 + \exp[\mu A(t)] - 1 / \exp(Y_{max} - Y_0) \right\} Eq. (1)$$

$$A(t) = t + \frac{1}{\upsilon} \ln[\exp(-\upsilon t) + \exp(-h_0) - \exp(-\upsilon t - h_0)]$$

where Y(t) is the microbial count in units of log CFU/g at time t (h), Y₀ is the logarithm of initial microbial count (log CFU/g), Y_{max} is the logarithm of maximum microbial count (log CFU/g), m characterizes the curvature before the stationary phase, A(t) is a rescaling of t, μ is the specific growth rate (log CFU/h), ν is the rate of increase of the limiting substrate, assumed to be equal to μ , λ is the lag phase duration (h), and h₀ is the physiological state of the microorganism under consideration.

Texture and Color Analyses

In the present study, the TA-XT2i texture analyzer (Texture Technology Corp., Scarsdale, NY, USA) was used to estimate the changes in texture of treated and untreated bell peppers during storage at 4°C and 25°C for 12 and 7 days, respectively. Samples $(3 \times 3 \text{ cm})$ were placed onto the press holder, and a probe (TA18) was moved down at 2 mm/sec. Maximum force was recorded using Texture Expert software (ver. 1.22, Texture Technology Corp.). All experiments were performed three times with independently prepared samples.

The color change of treated and untreated bell peppers were measured with a Minolta colorimeter (model CR300; Minolta Co.,

Osaka, Japan) at three locations on each leaf and expressed as L*, a*, and b* values, which indicate color lightness, redness, and yellowness of the sample, respectively. All analyses were performed three times with independently prepared samples.

Statistical Analysis

The combined effects of SAEW and ultrasound at different temperatures against *L. monocytogenes* and *S. Typhimurium* on bell pepper samples were tested by one factor analysis of variance (ANOVA) using the SPSS statistical package ver. 21 (SPSS lnc., Chicago, lL, USA). Differences between sample means were analyzed according to Tukey's test.

Results and Discussion

The pH, ORP, and ACC of slightly acidic electrolyzed water used in this study were 5.0-5.2, 930-950 mV, and 28-30 mg/l, respectively. HOCl present in electrolyzed water is the main compound that enables to kill the microbial cell through inhibiting glucose oxidation by chlorine-oxidizing sulfhydryl groups of certain enzymes important in carbohydrate metabolism [16]. Ultrasound, a non-thermal method, is a form of energy generated by inaudible sound waves (>16 kHZ) [17]. The bath-type ultrasound at frequency of 20-100 kHz is widely used and generates a powerful cavitation phenomenon, which can destroy and detach microorganisms from surfaces of fresh produce without affecting quality [31, 32]. It has been reported that ultrasound enables to enhance the antimicrobial effect of chemical sanitizers [5, 6, 22, 27, 31, 34]. Therefore, the combined treatment of SAEW and ultrasound at mild heat (60°C) temperature was conducted in this study to evaluate the synergistic effect in microbial safety and sensory quality of bell pepper during storage at 4°C and 25°C.

Effect of Decontamination Treatment on Bacterial Growth During Storage

The effects of different sanitizing treatments on the growth of *Listeria monocytogenes*, *Salmonella enterica* serovar *Typhimurium*, and total bacterial count on each bell pepper during storage at 4°C and 25°C for 18 days and 60 h, respectively, are shown in Figs. 1–3. All the isothermal growth curves clearly exhibited the lag, exponential, and stationary phases. As shown in Figs. 1–3, treatments with DW+60°C, SAEW+60°C, and SAEW+US+60°C for 1 min achieved an additional reduction of 0.65, 1.72, and 2.70 log CFU/g in average among the three types of pathogens, when compared with that of water washing (control). The maximum reduction achieved from SAEW+US+60°C

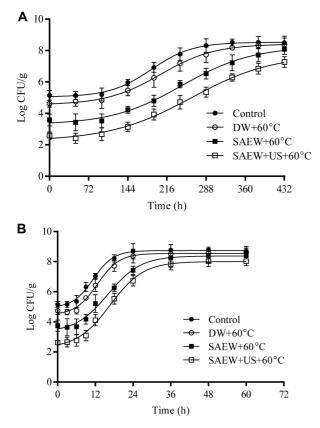


Fig. 1. Effects of different sanitizing treatments on the growth of *L. monocytogenes* on fresh-cut bell pepper stored at $4^{\circ}C$ (**A**) and $25^{\circ}C$ (**B**).

Vertical bars represent the mean ± standard deviation of triplicate assays. DW, deionized water (control); SAEW, slightly acidic electrolyzed water; US, ultrasound.

treatment indicated that an increased antimicrobial effect of SAEW was promoted by ultrasound. It has been reported that microscopic bubbles generated by cavitation are forced to oscillate in size or shape in liquids during ultrasound treatment [29]. The adherent bubbles on every surface of the submerged produce keep growing until they eventually implode and release the energy that causes detachment and inactivation of pathogens from the produce [10]. Gogate and Kabadi [14] reported that ultrasound treatment enables to contribute the penetration of the chemical oxidants through the cellular membrane and assists in the disaggregation of the microorganisms and thus results in an increased efficiency of sanitizing agents. São José et al. [30] reported that the combined treatment of ultrasound (40 kHz) and 1% lactic acid for 3 min at room temperature reduced Salmonella enterica Enteritidis on green pepper by 2.9 log CFU/g. Forghani and Oh [13] reported that the combined treatment of SAEW

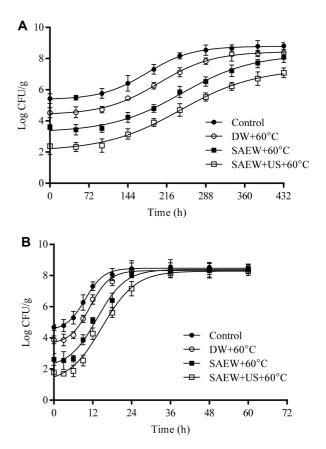


Fig. 2. Effects of different sanitizing treatments on the growth of *S. Typhimurium* on fresh-cut bell pepper stored at $4^{\circ}C$ (**A**) and $25^{\circ}C$ (**B**).

Vertical bars represent the mean ± standard deviation of triplicate assays. DW, deionized water (control); SAEW, slightly acidic electrolyzed water; US, ultrasound.

(pH, 5.2-5.5; ORP, 500-600 mV; ACC, 21-22 mg/l) and ultrasound (40 kHz) followed by water wash for 3 min at room temperature resulted in 2.1 and 2.4 log CFU/g reductions on average for TBC and L. monocytogenes, respectively, on different vegetables (lettuce, sesame leaf, and spinach). Sagong et al. [27] reported that the combined treatment with ultrasound (40 kHz) and 2% organic acid for 5 min at room temperature $(23 \pm 2^{\circ}C)$ achieved 3.18 and 2.87 log CFU/g reductions in L. monocytogenes and S. Typhimurium on lettuce, respectively. It has been reported that combined treatment of low concentration electrolyzed water (LcEW) and ultrasound (40 kHz) at 40°C for 3 min achieved a 2.1 log CFU/g reduction [12]. In comparison with the observed bacterial reductions from the abovereported studies, our results on the bacterial reduction by SAEW+US+60°C were more effective on bacterial reduction by shortening the processing time down to 1 min.

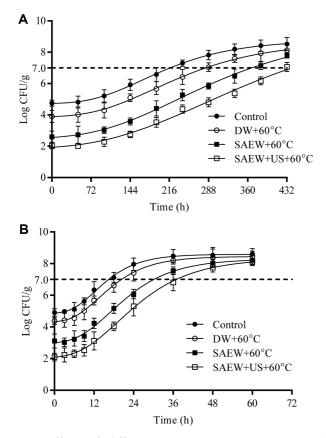


Fig. 3. Effects of different sanitizing treatments on total bacterial counts of bell pepper stored at 4°C (**A**) and 25°C (**B**). DW, deionized water (control); SAEW, slightly acidic electrolyzed water; US, ultrasound; Dash line, the end of shelf-life.

Primary Modeling for Bacterial Growth During Storage

In the present study, the growth data (log CFU/g) were fitted into the Baranyi model to calculate the specific growth rate (SGR, log CFU/h) and lag time (LT, h) of L. monocytogenes and S. Typhimurium growth on treated bell pepper during storage at 4°C and 25°C. The primary model provided a good statistical fitness with a high coefficient of determination (R^2 average > 0.98) (Table 1). Regardless of storage temperature, the SGR values of L. monocytogenes at each treatment group were significantly lower (p < 0.05) than that of S. Typhimurium, whereas the LT values of *L. monocytogenes* were significantly greater (p < 0.05) than that of S. Typhimurium (Table 1), indicating that S. Typhimurium grew faster than L. monocytogenes on bell pepper. It has been reported that Salmonella enterica on lettuce at 25°C grew faster than L. monocytogenes [28]. In addition, the LT value of each pathogen growth on SAEW+US+60°C treated bell pepper was significantly greater (p < 0.05) than that obtained in the SAEW+60°C group, indicating that an efficient control of the pathogen growth on bell pepper was achieved by ultrasound. The significant difference in the LT might be due to the decreased metabolic activity of the pathogens by the sanitizing treatment before the storage. Baranyi and Roberts [3] reported that LT is affected by both the pre- and post-inoculation environment. Therefore, based on the comparison of LT, the combined treatment (SAEW+US+60°C) was more effective in controlling the pathogens growth on bell pepper stored at 4°C and 25°C.

Table 1. Effects of different combination treatments on growth parameters (mean ± standard deviation) of *L. monocytogenes*, *S. Typhimurium*, and total bacterial counts (TBC) on bell pepper at 4°C and 25°C.

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Pathogen	Treatments ^a		Storage at 4°C		St	Storage at 25°C			
ramogen	Treatments	SGR	LT	\mathbb{R}^2	SGR	LT	\mathbb{R}^2		
L. monocytogenes	Control	$0.018\pm0.001^{\rm D}$	101.42 ± 10.19^{G}	0.995	$0.25\pm0.06^{\rm E}$	$5.21 \pm 1.24^{\rm F}$	0.994		
	DW+60°C	$0.017 \pm 0.001^{\circ}$	$104.12 \pm 21.46^{\rm H}$	0.991	$0.23 \pm 0.07^{\circ}$	5.62 ± 0.63^{G}	0.993		
	SAEW+60°C	$0.017 \pm 0.002^{\circ}$	$105.51 \pm 18.17^{\rm H}$	0.986	$0.23 \pm 0.05^{\circ}$	5.72 ± 2.01^{G}	0.991		
	SAEW+US+60°C	$0.017 \pm 0.002^{\circ}$	110.19 ± 15.71^{I}	0.988	$0.24\pm0.04^{\rm D}$	6.26 ± 1.68^{H}	0.993		
S. Typhimurium	Control	0.023 ± 0.001^{G}	$85.81 \pm 10.15^{\rm E}$	0.991	0.33 ± 0.06^{H}	$4.12 \pm 1.21^{\text{B}}$	0.995		
	DW+60°C	$0.021 \pm 0.001^{\rm E}$	$91.84 \pm 18.79^{\rm F}$	0.989	$0.33\pm0.04^{\rm H}$	$4.36 \pm 1.32^{\circ}$	0.991		
	SAEW+60°C	$0.022 \pm 0.002^{\rm F}$	$94.03 \pm 19.85^{\text{F}}$	0.992	$0.32 \pm 0.04^{\rm G}$	$4.85 \pm 1.66^{\rm D}$	0.994		
	SAEW+US+60°C	$0.021 \pm 0.001^{\rm E}$	$98.81 \pm 14.15^{\text{G}}$	0.993	$0.30\pm0.02^{\rm F}$	5.26 ± 2.05^{F}	0.995		
TBC	Control	$0.015 \pm 0.001^{\text{B}}$	$58.69 \pm 10.12^{\text{A}}$	0.996	$0.16 \pm 0.02^{\text{B}}$	$3.75 \pm 1.52^{\text{A}}$	0.995		
	DW+60°C	$0.014 \pm 0.001^{\rm A}$	$65.26 \pm 11.21^{\text{B}}$	0.995	$0.16 \pm 0.02^{\text{B}}$	$4.18 \pm 1.71^{\text{B}}$	0.991		
	SAEW+60°C	$0.015 \pm 0.001^{\text{B}}$	$68.73 \pm 16.08^{\circ}$	0.985	$0.15\pm0.01^{\rm A}$	$4.39 \pm 1.67^{\circ}$	0.993		
	SAEW+US+60°C	$0.014\pm0.001^{\rm A}$	$83.13 \pm 10.34^{\mathrm{D}}$	0.991	$0.16\pm0.01^{\scriptscriptstyle B}$	$5.02 \pm 2.28^{\text{E}}$	0.993		

 $^{A-I}$ Values within the same row with different capital letters indicate significant difference (p < 0.05) according to Tukey's test.

^aDifferent treatment conditions of DW (sterilized deionized water), SAEW (slightly acidic electrolyzed water), and US (ultrasound).

SGR, specific growth rate (log CFU/h); LT, lag time (h).

Shelf-Life and Sensory Quality

Changes in background microbial populations of bell pepper during storage at 4°C and 25°C are shown in Figs. 3A and 3B, respectively. The growth curves of TBC on treated (DW+60°C, SAEW+60°C, and SAEW+US+60°C) bell pepper were all below that of the control, regardless of storage temperature. The end of shelf-life was considered to occur when the TBC reached $\geq 7 \log CFU/g$ [15]. The TBC of the control group reached unacceptable levels within 10 days $(7.30 \pm 0.20 \log CFU/g)$ and 18 h (7.13 ± 0.31) log CFU/g) of storage at 4°C and 25°C, respectively. The TBC on treated (SAEW+US+60°C) bell pepper stored at 4°C and 25°C reached unacceptable levels after 18 days (7.06 \pm 0.29 $\log CFU/g$) and 48 h (7.76 ± 0.28 log CFU/g), respectively, reflecting that the treatment (SAEW+US+60°C) effectively prolonged the shelf-life of bell pepper by approximately 8 days and 30 h during storage at 4°C and 25°C, respectively. In addition, the results in Table 1 show that a significantly (p < 0.05) increased LT was achieved by the combined treatment (SAEW+US+60°C), regardless of storage temperature. These results thus indicate that the combined treatment (SAEW+US+60°C) effectively controlled the growth of aerobic bacteria by prolonging the LT and shelflife of bell pepper, as compared with the treatment without ultrasound. It has been reported that the combined treatment of SAEW (pH, 6.29; OPR, 826 \pm 16 mV; ACC, 30 \pm 0.2 mg/l) and 0.5% fumaric acid with mild heat (40°C) resulted in a longer LT on pork stored at 4°C [23]. Yang et al. [36] also reported that the combination of ultrasound (40 kHz) with salicylic acid was more effective than the individual treatment in controlling postharvest blue mold that is caused by Penicillium expansum in peach fruit. Moreover, it has been reported that the antimicrobial effect of LcEW in combination with ultrasound (40 kHz) was effective in extending the shelf-life of lettuce [12].

During storage and distribution, the changes in quality of bell pepper might be due to the physicochemical stress by SAEW, ultrasound, and mild heat treatment. To investigate this possibility, the effects of different sanitizing treatments (DW+60°C, SAEW+60°C, SAEW+US+60°C) for 1 min on the quality of bell pepper during storage at 4°C and 25°C for 12 and 7 days, respectively, were studied by colorimetric and texture analyses (Tables 2-4). The results in Tables 2 and 3 show that there was no significant change ($p \ge 0.05$) in L (lightness), a (redness), and b (yellowness) values of bell pepper during storage at 4°C and 25°C for 9 and 3 days, respectively, regardless of treatment groups. In addition, there was no significant change ($p \ge 0.05$) in the hardness values of bell pepper stored at 4°C after 12 days, regardless of the type of treatment, whereas the values of bell pepper stored at 25°C were significantly different (p < 0.05) after 7 days, when compared with those of storage within 5 days (Table 4). These results indicate that the combined treatment (SAEW+US+60°C) did not significantly influence the quality of bell pepper during the storage, when compared with the control group. Cao et al. [7] also stated that application of ultrasound (40 kHz) enables to control food spoilage and maintain the quality of fresh produce.

In conclusion, based on the results of the present study, the combined treatment (SAEW+US+60°C) effectively improved the microbial safety by prolonging the shelf-life of bell pepper during storage at 4°C and 25°C. Moreover, the combined treatment did not significantly affect the quality of bell pepper during the storage. The novel sanitizing technique developed in this study could thus be useful to control the growth of undesirable microorganisms on bell pepper during storage and distribution. Further studies will be useful to determine the physicochemical properties of ultrasound that enhance the antimicrobial

Table 2. Color parameters (mean ± standard deviation) for bell pepper stored at 4°C for 12 days following different sanitizing treatments for 1 min.

t ^h		Control ^a			DW+60°C ^b		S	SAEW+60°C	nC	SA	EW+US+60	°C ^d
(days)	Le	a ^f	b ^g	L	а	b	L	а	b	L	a	b
0	$84.7\pm3.0^{\rm A}$	$\textbf{-2.6}\pm0.1^{\rm A}$	$37.4 \pm 4.3^{\text{A}}$	$83.3\pm2.0^{\rm A}$	$-2.7\pm0.2^{\rm A}$	$27.3\pm2.7^{\rm A}$	$88.7\pm4.7^{\rm A}$	$-3.7\pm0.3^{\rm A}$	$30.5\pm1.5^{\scriptscriptstyle A}$	$84.5\pm2.8^{\rm A}$	$-5.1\pm0.2^{\rm A}$	$38.7\pm2.8^{\rm A}$
3	$84.6\pm2.8^{\rm A}$	$-2.6\pm0.3^{\rm A}$	37.2 ± 3.5^{A}	$83.1\pm2.1^{\rm A}$	$\textbf{-2.7}\pm0.1^{\rm A}$	$27.1\pm0.7^{\rm A}$	$88.5 \pm 1.7^{\rm A}$	$-3.7\pm0.1^{\rm A}$	$30.4\pm0.9^{\rm A}$	$84.2\pm3.6^{\rm A}$	$\textbf{-5.1} \pm 0.3^{\text{A}}$	$38.1\pm2.5^{\rm A}$
6	$84.1 \pm 1.9^{\rm A}$	$\text{-}2.5\pm0.2^{\scriptscriptstyle A}$	$37.1 \pm 2.7^{\text{A}}$	$82.7\pm3.0^{\rm A}$	$\text{-}2.7\pm0.2^{\text{A}}$	$27.0\pm0.7^{\rm A}$	$88.2\pm2.9^{\rm A}$	$-3.6\pm0.2^{\rm A}$	$30.2 \pm 2.1^{\text{A}}$	$84.0\pm4.3^{\rm A}$	$\textbf{-4.8} \pm 0.1^{\text{A}}$	$38.2\pm1.4^{\rm A}$
9	$83.3\pm5.0^{\rm A}$	$\text{-}2.4\pm0.2^{\scriptscriptstyle A}$	$36.8 \pm 3.1^{\text{A}}$	$81.8\pm2.9^{\text{AB}}$	$\text{-}2.4\pm0.2^{\scriptscriptstyle B}$	$26.6 \pm 1.1^{\rm A}$	$87.6\pm5.3^{\rm A}$	$-3.5\pm0.2^{\rm A}$	$29.8\pm2.4^{\rm A}$	$83.7\pm5.1^{\rm A}$	$-4.7\pm0.1^{\rm A}$	$37.8 \pm 1.5^{\scriptscriptstyle \rm A}$
12	$81.6\pm4.5^{\rm B}$	$-2.1\pm0.2^{\rm B}$	35.2 ± 2.1^{B}	$81.1 \pm 3.4^{\text{B}}$	$\textbf{-2.3}\pm0.3^{\rm C}$	25.2 ± 2.1^{B}	85.6 ± 2.6^{B}	$-3.1\pm0.2^{\rm B}$	$28.5\pm1.0^{\rm B}$	$83.6\pm4.5^{\scriptscriptstyle B}$	-4.4 ± 0.2^{B}	36.6 ± 2.1^{B}

 $^{A-C}$ Values within the same row with different capital letters indicate significant difference (p < 0.05) according to Tukey's test.

ard Different treatment conditions of DW (sterilized deionized water, control), SAEW (slightly acidic electrolyzed water), and US (ultrasound).

^{e-g}L, lightness; a, redness; b, yellowness.

^hStorage period (3, 6, 9, and 12 days).

ť		Control ^a			DW+60°C ^b		5	SAEW+60°C	c	SA	EW+US+60°	°C ^d
(days)	Le	a ^f	b ^g	L	а	b	L	а	b	L	а	b
0	$87.4\pm3.3^{\rm A}$	$\textbf{-3.3}\pm0.2^{\rm A}$	$36.2\pm4.3^{\scriptscriptstyle A}$	$86.3\pm3.4^{\rm A}$	$-4.3\pm0.5^{\rm A}$	$33.8\pm2.2^{\rm A}$	$89.2\pm4.8^{\rm A}$	$\textbf{-5.2} \pm 0.8^{\rm A}$	$25.7\pm1.3^{\rm A}$	$85.5\pm5.3^{\rm A}$	$\textbf{-3.2}\pm0.8^{\rm A}$	$33.4 \pm 1.3^{\scriptscriptstyle A}$
1	$87.4\pm6.2^{\rm A}$	$\textbf{-3.3} \pm 0.3^{\mathrm{A}}$	$36.1\pm3.5^{\scriptscriptstyle A}$	$86.4\pm3.4^{\rm A}$	$\textbf{-4.3} \pm 0.3^{\mathrm{A}}$	$33.6\pm2.3^{\rm A}$	$89.1\pm3.4^{\scriptscriptstyle A}$	$\textbf{-5.2} \pm 0.3^{\text{A}}$	$25.3 \pm 1.9^{\rm A}$	$85.3\pm4.6^{\rm A}$	$\textbf{-3.2} \pm 0.3^{\text{A}}$	$33.2 \pm 2.9^{\text{A}}$
3	$87.3\pm4.7^{\rm A}$	$\textbf{-3.2}\pm0.2^{\rm A}$	$36.0\pm2.7^{\rm A}$	$86.2\pm4.1^{\rm A}$	$\textbf{-4.2} \pm 0.8^{\rm A}$	$33.5\pm2.9^{\rm A}$	$88.9\pm3.1^{\scriptscriptstyle A}$	$\textbf{-5.2} \pm 0.6^{\text{A}}$	$25.2\pm2.1^{\rm A}$	$85.2\pm3.3^{\rm A}$	$\textbf{-3.0} \pm 0.4^{\mathrm{A}}$	$33.1 \pm 1.1^{\text{A}}$
5	$86.3\pm2.2^{\rm A}$	$\textbf{-2.8} \pm 0.1^{\text{B}}$	$35.8\pm3.3^{\scriptscriptstyle AB}$	$85.5\pm2.4^{\scriptscriptstyle AB}$	$-3.9\pm0.6^{\rm B}$	$33.1\pm1.8^{\rm AB}$	$87.7\pm3.8^{\rm AB}$	$\textbf{-5.1} \pm 0.5^{\text{B}}$	$24.5\pm1.4^{\scriptscriptstyle B}$	$84.3\pm4.1^{\scriptscriptstyle AB}$	$\text{-}2.7\pm0.3^{\text{AB}}$	$32.8 \pm 1.9^{\rm A}$
7	$85.1\pm2.2^{\rm B}$	$\textbf{-2.3} \pm 0.1^{\text{C}}$	$34.6\pm1.2^{\scriptscriptstyle B}$	$84.2\pm2.2^{\scriptscriptstyle B}$	$-2.7\pm0.3^{\rm C}$	$32.3\pm2.4^{\scriptscriptstyle B}$	$86.4\pm4.8^{\scriptscriptstyle B}$	$-4.3\pm0.3^{\rm C}$	$23.4 \pm 1.5^{\rm C}$	$84.1\pm3.9^{\scriptscriptstyle B}$	$-2.5\pm0.5^{\rm B}$	$31.5\pm2.0^{\scriptscriptstyle B}$

Table 3. Color parameters (mean ± standard deviation) for bell pepper stored at 25°C for 7 days following different sanitizing treatments.

 $^{A-C}$ Values within the same row with different capital letters indicate significant difference (p < 0.05) according to Tukey's test.

a-dDifferent treatment conditions of DW (sterilized deionized water, control), SAEW (slightly acidic electrolyzed water), and US (ultrasound).

e-gL, lightness; a, redness; b, yellowness.

^hStorage period (3, 6, 9, and 12 days).

Table 4. Change in hardness (mean ± standard deviation) of bell pepper stored at 4°C and 25°C after different sanitizing treatments.

T ^e	\mathbf{T}^{f}	Hardness (N)							
(°C)	(days)	Control ^a	DW+60 ^b	SAEW+60 ^c	SAEW+US+60 ^d				
4	0	$1,017 \pm 60^{\text{A}}$	$1,083 \pm 82^{\text{A}}$	$1,532 \pm 68^{A}$	$1,015 \pm 42^{\text{A}}$				
	3	975 ± 23^{A}	$1,023 \pm 55^{\text{A}}$	$1,552 \pm 150^{\text{A}}$	$1,033 \pm 146^{A}$				
	6	936 ± 33^{A}	966 ± 56^{A}	$1,320 \pm 168^{A}$	$1,004 \pm 62^{A}$				
	9	957 ± 66^{A}	$966 \pm 24^{\mathrm{A}}$	$1,293 \pm 145^{\text{A}}$	973 ± 24^{A}				
	12	927 ± 66^{A}	933 ± 27^{A}	$1,203 \pm 120^{\text{A}}$	922 ± 13^{A}				
25	0	$1,278 \pm 75^{\text{A}}$	$1,284 \pm 30^{\text{A}}$	$1,041 \pm 85^{A}$	$1,143 \pm 70^{\text{A}}$				
	1	$1,365 \pm 60^{\text{A}}$	$1,260 \pm 55^{\text{A}}$	$1,083 \pm 57^{\text{A}}$	$1,227 \pm 58^{A}$				
	3	$1,128 \pm 81^{\text{A}}$	$1,272 \pm 62^{A}$	$1,014 \pm 45^{A}$	$1,128 \pm 62^{A}$				
	5	$1,194 \pm 66^{A}$	$1,188 \pm 52^{A}$	$987 \pm 45^{\text{A}}$	$1,095 \pm 24^{\text{A}}$				
	7	$1,047 \pm 45^{B}$	$1,074 \pm 90^{B}$	$888\pm74^{\scriptscriptstyle B}$	912 ± 40^{B}				

 $^{A-B}$ Values within each five of the same row with different capital letters indicate significant difference (p < 0.05) according to Tukey's test.

a-dDifferent treatment conditions of DW (sterilized deionized water, control), SAEW (slightly acidic electrolyzed water), and US (ultrasound).

°Storage temperature at 4°C and 25°C.

^fStorage period (3, 6, 9, and 12 days).

effect of chemical sanitizers (*e.g.*, SAEW) and thus to improve the synergistic effect of combining disinfection technologies.

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