

Application of slightly acidic electrolyzed water and ultraviolet light for *Salmonella enteritidis* decontamination of cell suspensions and surfaces of artificially inoculated plastic poultry transport coops and other facility surfaces

Y. T. Zang, S. Bing,¹ Y. J. Li, and D. Q. Shu

College of Animal Science and Technology, Jiangxi Agricultural University, Jiangxi 330045, China

ABSTRACT The efficiency of combination treatment of slightly acidic electrolyzed water (SAEW) and ultraviolet light (UV) for inactivation of *Salmonella enteritidis* (*S. enteritidis*) on the surface of plastic poultry coops and other facility surfaces was evaluated in the presence of organic matter. The bactericidal activities of SAEW, UV + SAEW, and composite phenol (CP) for inactivating *S. enteritidis* were also compared. Moreover, a model of UV + SAEW treatment of plastic transport coops with different times and available chlorine concentrations (ACC) was developed using multiple linear regression analysis. There are differences between SAEW and CP inactivation of *S. enteritidis* on coops, stainless steel, and glass surfaces ($P < 0.05$), and there are no differences between SAEW and CP on tire surfaces ($P > 0.05$). Disinfection of some rough material surfaces with SAEW treatment alone under feces interference on poultry farms may need a longer treatment

time and/or a higher ACC than smooth surfaces. The combined treatment of UV and SAEW showed higher inactivation efficiency of *S. enteritidis* compared to CP and SAEW treatment alone ($P < 0.05$) in pure cultures or on the facility surfaces. A complete 100% inactivation of *S. enteritidis* on plastic poultry coop surfaces was obtained by using UV + SAEW with an ACC of 90 mg/L for more than 70 s. The established model had a good fit that was quantified by the determination coefficient R^2 (0.93) and a lack of fit test ($P > 0.05$). The bactericidal efficiency of UV + SAEW increased with greater ACC and increasing time. The findings of this study indicate that the combination treatment of UV and SAEW may be a promising disinfection method and could be used instead of SAEW alone, especially on rough materials in the presence of organic matter on poultry farms.

Key words: slightly acidic electrolyzed water, ultraviolet, *Salmonella enteritidis*, facility surfaces

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INTRODUCTION

Contaminated transport coops, iron materials, and other facility surfaces have been reported to be sources of *Salmonella* on poultry farms, and *Salmonella* is an important pathogen for animals and humans and represents a serious public health concern worldwide (Racicot et al., 2012; Totton et al., 2012). Disinfection has been reported as a generally employed method for preventing the introduction of both endemic and epidemic infections in animal production (Totton et al., 2012). Some studies have demonstrated that disinfection can reduce microbial contamination of poultry transport coops and other objects and decrease the prevalence of *Salmonella* spp (DeBenedictis et al., 2007; Zang et al., 2017a,b). At present, most farmers utilize chemical sanitation systems to decontaminate

facility surfaces in the poultry industry. However, the use of chemical disinfectants to eliminate or inactivate pathogens may cause potentially toxic, corrosive, or volatile problems (Gulati et al., 2001; Cao et al., 2009).

In recent years, the use of slightly acidic electrolyzed water (SAEW) as a facility surface decontamination method has been met with increasing interest. SAEW, with a near-neutral pH of 5.0–6.5, is generated by electrolysis of dilute hydrochloric acid or sodium chloride solution in an electrolytic cell without diaphragm separation (Cao et al., 2009; Koide et al., 2009; Sheng et al., 2018). Relative to chemical disinfectants, SAEW has the advantage of being less corrosive for equipment, less irritating for hands, and minimizes human health and safety issues from Cl_2 off-gassing (Cao et al., 2009). Some studies have demonstrated that SAEW could be used as a disinfectant in the poultry industry. Cao et al. (2009) have reported that a reduction of $6.5 \log_{10}$ CFU/g of *Salmonella enteritidis* (*S. enteritidis*) on eggshells was obtained by SAEW at an available chlorine concentration (ACC) of 15 mg/L for 3 min. Hao et al. (2013) reported that treatment with

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¹Corresponding author: zangyitian1@126.com

SAEW with an ACC of 250 mg/L significantly reduced bacteria and fungi in dust, feces, feathers and feed ($P < 0.05$) in a layer house. Zang et al. (2015a) also indicated that a maximum reduction of $3.12 \log_{10}$ CFU/cm² for *S. enteritidis* was obtained for coops treated with tap water for 15 s followed by SAEW treatment for 40 s at an ACC of 50 mg/L. These findings indicate that SAEW may be an alternative disinfectant to reduce the population of pathogens on facility surfaces on poultry farms. However, it was shown that SAEW efficiency could be affected by the presence of organic matter. Zang et al. (2015b) found that under feces soiling interference, a reduction of $1.38 \log_{10}$ CFU/cm² for an *Escherichia coli* (*E. coli*) and *S. enteritidis* mixture was obtained on vehicle tires, after washing with tap water for 4 min followed by SAEW treatment for 5 min at an ACC of 140 mg/L. Zang et al. (2017a) also reported a 2.61-log reduction of bacteria on iron materials, which was obtained after 2 min of treatment with an ACC of SAEW of 200 mg/L. However, in the food industry, SAEW is often used at an ACC of approximately 30 to 100 mg/L (Deza et al., 2005; Jung et al., 2018). Single antimicrobial treatments of SAEW need longer washing and treatment times and/or a higher ACC in the poultry industry than in other industries. Therefore, to overcome this drawback, combining the effects of 2 or more decontamination methods with SAEW in lower quantities and lower treatment times could be applied.

Currently, ultraviolet (UV) light is used as a surface decontamination method, and it is lethal to most microorganisms on hard surfaces and is increasingly preferred and applied in research and the poultry industries (Goerzen and Scott., 1995; Gabriel et al., 2017). UV-C light ($\lambda = 254$ nm) provides effective inactivation of microorganisms by damaging nucleic acids and creating nucleotide dimers, thus leaving the microorganisms unable to perform vital cellular functions (Turtoi and Borda, 2014). De Reu et al. (2006) obtained a 4-log reduction when a strain of *E. coli* and a *Listeria monocytogenes* strain were treated with UV-C light at 0.18 J/cm². Chavez et al. (2002) investigated the effects of UV-C light on the total number of bacterial populations total aerobic plate count (APC) of eggshells and found that the APC was significantly reduced for eggshells exposed to UV-C light with an intensity of 7.35 mW/cm² for 30 and 60 s compared to untreated eggs. UV light has also usually been combined with electrolyzed water to improve the inactivation of microorganisms. Pang and Hung (2016) demonstrated that the combined treatment of SAEW and UV-ozonated water in the spray washing process could more effectively reduce *E. coli* O157:H7 on lettuce. It has also been reported that UV light is more efficient when combined with other disinfectants (Mcdaniel, 2011; Turtoi and Borda, 2014). However, little information is available on the synergistic effects of SAEW and UV-light to decontaminate contaminated facility surfaces in the presence of organic materials.

Hence, the overall objectives of this study were to: (1) to compare the efficiency of SAEW, UV+SAEW, and other disinfectants (composite phenol) to inactivate *S. enteritidis* in cell suspensions under the presence of organic matter, (2) to compare the efficiency of SAEW, UV+SAEW, and other disinfection methods (composite phenol, UV) to inactivate *S. enteritidis* on the surface of plastic poultry transport coops and other facility surfaces in the presence of feces soiling, and (3) to develop a model and to determine the effect of available chlorine treatment time on bactericidal activity of UV+SAEW on the surface of plastic poultry transport coops.

MATERIALS AND METHODS

Bacterial Cultures

The strains of *S. enteritidis* (CVCC 2184) were obtained from the China Veterinary Culture Collection (Beijing, China). The bacterium was hydrated according to the directions of the manufacturer and cultured in tryptic soy broth (TSB; Beijing Land Bridge Technology Company Ltd., Beijing, China) at 37°C for 24 h. Following incubation, a 10 mL culture was pooled into a sterile centrifuge tube and centrifuged at $4000 \times g$ at 4°C for 10 min. The supernatant was decanted, and the pellets were resuspended in 10 mL of 0.1% buffered sterile peptone water (BPW; Beijing Land Bridge Technology Company Ltd., Beijing, China), washed 3 times, and resuspended in 10 mL of the same solution to obtain a final cell concentration of approximately $9 \log$ CFU/mL. The bacterial population in each culture was confirmed by plating 0.1 mL portions of appropriately diluted culture on tryptic soy agar (TSA; Beijing Land Bridge Technology Company Ltd., Beijing, China) plates and then incubating the plates at 37°C for 24 h. The prepared cultures were then used in subsequent experiments.

Inoculation

A 20% solution of liquid feces was prepared by the addition of 100 g of chicken feces (obtained from poultry with no bedding) to 500 mL of sterile distilled water and then inactivated by autoclaving (YXQ-LS-18SI, Shanghai Boxun Industrial Co., Ltd., Shanghai China). The liquid feces solution was shaken and then mixed with equal portions of the prepared culture mixtures to obtain final populations of contaminated culture of approximately 10^8 CFU/mL and 10% concentration (Zang et al., 2015a).

The plastics were obtained from a plastic poultry transport coop (High Density Polyethylene materials, $7.35 \times 5.45 \times 2.60$ cm, Shenzhen Lanhai Co., Ltd., Shenzhen, China). The stainless steel and glass were purchased from commercial suppliers. The tires were obtained from a waste tire (750–16, Qingdao Hongxinyu Rubber Co., Ltd., Qingdao, China). The plastic coop

Table 1. Physicochemical properties of spraying solutions.

Solutions	Concentration of active ingredient (mg/L)	pH	ORP ¹ (mV)
Control (deionized water)	0	6.15 ± 0.02 ⁴	398.7 ± 0.4
SAEW ²	10	6.49 ± 0.03	797.8 ± 0.4
	30	6.51 ± 0.02	803.2 ± 0.2
	50	6.52 ± 0.01	818.3 ± 0.3
	70	6.54 ± 0.01	825.6 ± 0.7
	90	6.56 ± 0.02	835.4 ± 0.6
UV+SAEW ³	10	6.49 ± 0.03	798.5 ± 0.1
	30	6.51 ± 0.02	806.7 ± 0.4
	50	6.52 ± 0.01	819.7 ± 0.7
	70	6.54 ± 0.01	826.5 ± 0.4
	90	6.56 ± 0.02	836.7 ± 0.1
Composite phenol	0.4%	3.93 ± 0.03	432.6 ± 0.2
	0.7%	3.75 ± 0.05	440.8 ± 0.9
	1%	3.57 ± 0.02	478.5 ± 0.5

¹Oxidation reduction potential.

²SAEW = slightly acidic electrolyzed water

³UV+SAEW = Combination treatment of ultraviolet and slightly acidic electrolyzed water

⁴Values are the means ± standard deviation (n = 3).

was washed with tap water to remove soil, trimmed to approximately 1.5 × 1.5 cm² and packed in a polyethylene bag. The tires, stainless steel, and glasses were also washed with tap water to remove soil, trimmed to approximately 5 × 5 cm² in size and packed in a polyethylene bag for the experiment. Before inoculation, the surface samples packed by the kraft papers were inactivated by an autoclave (YXQ-LS-18SI, Shanghai Boxun Industrial Co., Ltd., Shanghai, China) and then air-dried under a biosafety hood (DH-920, Beijing East Union Hall Instrument Manufacturing Co., Ltd., Beijing, China) at room temperature for 60 min to remove water. Each sample piece was inoculated by spreading 0.1 mL onto the front side region of the prepared contaminated culture inoculum. Subsequently, all inoculated pieces were air-dried under the biosafety hood for 30 min at room temperature to allow bacterial attachment. The final concentrations of *S. enteritidis* inoculated on the plastic coop, tires, the stainless steel and glasses samples were approximately 6.67 log₁₀ CFU/cm², 6.01 log₁₀ CFU/cm², 6.64 log₁₀ CFU/cm², and 6.61 log₁₀ CFU/cm², respectively, on average. The samples for each treatment were prepared at least in duplicate, and all treatments were repeated 3 times. The results were reported as the mean values.

Preparation of Treatment Solutions

SAEW at different ACCs (Table 1) was produced using a nonmembrane generator (Ruiande Biosafety Technology Co., Ltd., Beijing, China) to electrolyze NaCl (1 g/L) containing HCl (100 μ/L) solution. Composite phenols (CP; Guangdong Treasure Biological Pharmaceutical Co., Ltd., Guangdong, China) were purchased from commercial suppliers and prepared by dilution with deionized water to obtain the final concentration (Table 1).

The pH, oxidation reduction potential (ORP), and ACC of treatment solutions were measured immedi-

ately before each experiment. The pH and ORP values were measured with a dual scale pH/ORP meter (CON60, Trans-Wiggins, Singapore). The ACC was determined by a digital chlorine test system (RC-2Z, Kasahara Chemical Instruments Co., Saitama, Japan). The detection range was 0 to 320 mg/L.

Preparation of UV light

The UV-C treatments were performed in a chamber (85 cm × 75 cm × 45 cm) equipped with 2 sets of 2 unfiltered germicidal emitting lamps (253.7 nm, Philips, Co., Netherlands). One set of lamps was placed on the left and the other one on the right of the radiation cabinet, and the height of the 2 lamps was both 40 cm. All UV experiments were conducted at a fixed initial UV intensity (10.2 ± 0.3 W/cm²), which was measured by a radiometer (UVX-254, Ultraviolet Products, California, USA). Before each experiment, the UV lamp was turned on for approximately 20 min to achieve stable irradiation intensity. Contaminated samples, prepared as previously described, were aseptically transferred to the base of sterile glass petri plates and placed on a net positioned midway between the UV-C lamps. To achieve the combined effect, the treatments with SAEW were carried out in the order shown in Tables 2 and 4.

Treatment of Pure Culture in the Presence of Organic Matter

A volume of 9 mL of CP (0.4, 0.7, and 1%) or SAEW (containing 10, 30, 50, 70, and 80 mg/L of ACC) was transferred to sterile tubes. One milliliter of each bacterial culture (approximately 8.0 log₁₀ CFU/mL) containing 1% bovine serum albumin (BSA) was added to 9 mL of CP or SAEW (with and without UV light) for 20, 40, 60, and 80 s. Following treatment, 1 mL of each sample was transferred to a tube containing 9 mL of

Table 2. Efficacy of SAEW, UV+SAEW, and CP against pure cultures of *S. enteritidis* with different ACCs and times in the presence of 1% BSA.

Treatment	Concentration of active ingredient ¹ (mg/L)	Surviving population of <i>S. enteritidis</i> (log ₁₀ CFU/mL) ³			
		20 s	40 s	60 s	80 s
SAEW ⁵	30	4.56 ± 0.04 ^A	4.54 ± 0.03 ^A	4.50 ± 0.02 ^A	4.48 ± 0.01 ^A
	50	3.34 ± 0.08 ^A	3.31 ± 0.06 ^A	3.26 ± 0.02 ^A	3.23 ± 0.10 ^A
	70	2.55 ± 0.11 ^A	2.51 ± 0.03 ^A	2.48 ± 0.05 ^A	2.44 ± 0.07 ^A
UV+ SAEW ⁶	30	2.44 ± 0.05 ^B	2.32 ± 0.06 ^B	2.19 ± 0.08 ^B	2.09 ± 0.02 ^B
	50	1.47 ± 0.07 ^B	1.23 ± 0.02 ^B	1.02 ± 0.07 ^B	0.73 ± 0.04 ^B
	70	0.71 ± 0.13 ^B	0.47 ± 0.08 ^B	0.15 ± 0.05 ^B	ND ⁴
CP ²	0.4%	4.88 ± 0.06 ^C	4.86 ± 0.11 ^C	4.85 ± 0.03 ^C	4.84 ± 0.06 ^C
	0.7%	3.47 ± 0.04 ^C	3.44 ± 0.01 ^C	3.42 ± 0.05 ^C	3.39 ± 0.02 ^C
	1%	2.74 ± 0.02 ^C	2.71 ± 0.09 ^C	2.69 ± 0.09 ^C	2.64 ± 0.04 ^C

¹ACC = Available chlorine concentration.

²CP = Composite phenol.

³Values are the means ± standard deviation (n = 3).

⁴ND = No detectable.

⁵SAEW = slightly acidic electrolyzed water.

⁶UV+ SAEW = Combination treatment of ultraviolet and slightly acidic electrolyzed water.

^{A-C}values with different capital-case letters in superscripts, within the same column of different disinfection methods at same concentration of active ingredient mean significantly different ($P < 0.05$).

Table 3. Surviving populations of SAEW, UV+SAEW, and CP against *S. enteritidis* on plastic poultry transport coops and other facility surfaces with ACC of 70 mg/L and treatment time of 80 s.

Treatment	ACC ¹ (mg/L)	Surviving population of <i>S. enteritidis</i> (log ₁₀ CFU/cm ²) ³			
		Tire	Coop	Stainless steel	Glass
Control	0	4.21 ± 0.12 ^{a,A}	3.93 ± 0.05 ^{b,A}	3.52 ± 0.08 ^{c,A}	3.46 ± 0.11 ^{c,A}
SAEW ⁵	70	3.81 ± 0.02 ^{a,B}	1.15 ± 0.01 ^{b,B}	0.91 ± 0.04 ^{c,B}	0.87 ± 0.07 ^{c,B}
UV ⁶	0	4.19 ± 0.09 ^{a,A}	4.01 ± 0.04 ^{a,A}	3.43 ± 0.02 ^{a,B}	2.86 ± 0.11 ^{b,C}
UV+ SAEW	70	3.19 ± 0.05 ^{a,C}	0.79 ± 0.03 ^{b,C}	ND ⁴	ND
CP ²	1%	3.83 ± 0.02 ^{a,B}	1.34 ± 0.07 ^{b,D}	1.17 ± 0.06 ^{c,C}	1.15 ± 0.08 ^{c,D}

¹ACC = Available chlorine concentration.

²CP = Composite phenol.

³Values are the means ± standard deviation (n = 3).

⁴ND = No detectable.

⁵SAEW = slightly acidic electrolyzed water.

⁶UV = Ultraviolet.

^{a-c}values with different lower-case letters in superscripts within a line were significantly different ($P < 0.05$).

^{A-D} values with different capital-case letters in superscripts within a column were significantly different ($P < 0.05$).

neutralizing buffer (0.5% Na₂S₂O₃) and then vortexed using the mixer. After 5 min of neutralization, surviving bacteria were determined by serial dilution in sterile 0.1% peptone water, plated in duplicate (0.1 mL) on tryptic soy agar plates, and then incubated at 37°C for 24 h.

Treatment of Samples and Microbiological Determination

Inoculated sample pieces were sprayed with composite phenol or SAEW (with and without UV light) or sterilization deionized water (control) by an atmospheric pressure manual sprayer to disinfect them under different conditions (Tables 3 and 4). After treatment, moistened sterile swabs were treated with a neutralizing agent (described above) and used to collect surface samples from the plastic pieces. The sterilized cotton swabs, which had been wiped back and forth twenty times on the sample surfaces, were immediately

transferred into 5 mL neutralizing agent tubes for microbiological analyses. The tubes were shaken at 1800 rpm (MIR-S100, Sanyo Electric Biomedical Co., Ltd., Osaka, Japan). The surviving bacteria was determined by serial dilutions in sterile 0.1% peptone water, and 0.1 mL of each dilution was plated onto TSA in triplicate and then incubated at 37°C for 24 h before the colonies were counted. No viable cells in the blank control group were detected in each trial.

Statistical Analysis

All treatments were repeated 3 times, and the results were reported as the mean values. A *t*-test was performed to determine the significance of differences.

Origin (Version 9.0, OriginLab Cor., Hampton, USA) was used for multiple linear regression analysis and to generate the models according to the data from Table 4. The statistical significance and goodness of fit of the models were evaluated using the adjusted

Table 4. Efficacy of UV+SAEW against *S. enteritidis* on plastic poultry transport coops with different ACCs and times.

Treatment	ACC ¹ (mg/L)	Surviving population of <i>S. enteritidis</i> (log ₁₀ CFU/mL) ³				
		10 s	30 s	50 s	70 s	90 s
Control	0	4.69 ± 0.09 ^{a,A}	4.67 ± 0.07 ^{b,A}	4.63 ± 0.06 ^{c,A}	4.61 ± 0.03 ^{d,A}	4.58 ± 0.08 ^{e,A}
UV+ SAEW ²	10	4.59 ± 0.03 ^{a,B}	4.36 ± 0.03 ^{b,B}	4.17 ± 0.06 ^{c,B}	3.95 ± 0.02 ^{d,B}	3.72 ± 0.02 ^{e,B}
	30	4.21 ± 0.01 ^{a,C}	3.79 ± 0.07 ^{b,C}	3.41 ± 0.04 ^{c,C}	3.08 ± 0.04 ^{d,C}	2.61 ± 0.08 ^{e,C}
	50	3.62 ± 0.06 ^{a,D}	3.01 ± 0.08 ^{b,D}	2.45 ± 0.09 ^{c,D}	1.85 ± 0.05 ^{d,D}	1.28 ± 0.01 ^{e,D}
	70	3.09 ± 0.05 ^{a,E}	2.27 ± 0.05 ^{b,E}	1.43 ± 0.04 ^{c,E}	0.80 ± 0.02 ^{d,E}	0.19 ± 0.07 ^{e,E}
	90	2.95 ± 0.04 ^{a,F}	1.94 ± 0.02 ^{b,F}	0.85 ± 0.02 ^{c,F}	ND ⁴	ND

¹ACC = Available chlorine concentration.

²UV+ SAEW = Ultraviolet and slightly acidic electrolyzed water.

³Values are the means ± standard deviation (n = 3).

⁴ND = No detectable.

^{a-e}values with different lower-case letters in superscripts within a line were significantly different ($P < 0.05$).

^{A-F}values with different capital-case letters in superscripts within a column were significantly different ($P < 0.05$).

determination coefficients (R^2). The statistical significance of the model was determined using Fisher's F-test.

RESULTS AND DISCUSSION

Treatment of Pure Cultures of *S. Enteritidis* in the Presence of Organic Matter

Table 2 shows the CP, SAEW, and UV + SAEW with different concentrations and their bactericidal activity for pure *S. enteritidis* cultures at different times in the presence of 1% BSA. The initial population of *S. enteritidis* was approximately 9.2 log₁₀ CFU/mL, and the bactericidal efficiency of all solutions increased with increasing available concentrations and time. The populations of *S. enteritidis* were reduced to undetectable levels with SAEW at an ACC of 70 mg/L in the presence of 1% BSA after 80 s of treatment. The SAEW and CP treatments reduced the population of *S. enteritidis* significantly ($P < 0.05$). Similar results were reported by Ni et al. (2015). They showed that with BSA at a concentration of 3.0%, SAEW at an ACC of 40 to 80 mg/L reduced the population of *S. enteritidis* after 2.5 to 7.5 min of treatment compared to the no treatment group ($P < 0.05$), and the reduction in the bacterial count was 2.60 to 4.96 log₁₀ CFU/mL. However, Cao et al. (2009) reported 100% inactivation of pure cultures of *S. enteritidis* (reduction of approximately 8.2 log₁₀ CFU/mL) using SAEW with a very low ACC of 4 mg/L. Different results in these studies may be due to the absence of BSA. Oomori et al. (2000) reported that the bactericidal activity of acidic electrolyzed water declines in the presence of organic materials. Ni et al. (2015) indicated that as the BSA concentration increased, the activity of SAEW decreased. The absence of organic materials causes SAEW with a high ACC to reach the same sterilization effect.

As shown in Table 2, the combined treatment of UV + SAEW showed higher inactivation of *S. enteritidis* compared to SAEW alone at the same ACC ($P < 0.05$). Our findings indicate that UV in combination with SAEW was a more effective way to inactivate

S. enteritidis in suspension. Although suspension tests are often used as an official method to test disinfectants, the main disadvantage of these tests is that they are unrealistic, because the bacteria in suspensions are usually more susceptible to disinfectants than bacteria dried on surfaces (Gradel et al., 2004; Ni et al., 2015). Therefore, surface tests should also be performed to confirm the disinfection efficiency of UV + SAEW combined treatment on *S. enteritidis*.

Treatment of *S. Enteritidis* on the Facility Surfaces in the Presence of Organic Matter

The effectiveness of various decontamination technologies, including CP, UV, SAEW, and the combination of UV and SAEW for the inactivation of *S. enteritidis* on the surface of facilities is summarized in Table 3. The initial populations of *S. enteritidis* on tire, plastic coop, stainless steel, and glass in the untreated group were 4.52, 4.63, 4.48, and 4.51 log₁₀ CFU/cm², respectively. No differences between the control group and the UV treatment group of tire, plastic coop samples were observed ($P > 0.05$). This result is mainly caused by the absence of feces on the surface of facilities, which shields cells from cramping the UV light penetration. Gomez-Lopez et al. (2007) have reported that the efficacy of using UV light for decontamination of foods is often lower than when tested on clean surfaces. Some studies have also demonstrated that UV light does not penetrate well through organic matter, such as protein and other organic matrices (Unluturk et al., 2007; Mun et al., 2009).

The maximum 3.64 log₁₀ reduction of bacteria in glass materials (initial populations of 4.51 log₁₀ CFU/cm²) was obtained with SAEW at an ACC of 70 mg/L for a treatment time of 80 s. However, only 0.71 log₁₀ was observed for the tire surface (initial populations of 4.52 log₁₀ CFU/cm²) after the same treatment. This difference is mainly because of the materials, which are highly significant factors when disinfecting using SAEW. Several studies have reported similar results. Liu and Su (2006) reported that

L. monocytogenes immersed in EO water (50 mg/L chlorine) for 5 min was reduced in number by 3.73 log₁₀ CFU/25 cm² on stainless steel, and a reduction of only 1.52 log₁₀ CFU/25 cm² was observed on floor tile. Zang et al. (2017a) showed that the inactivation efficiency of *S. enteritidis* sprayed by SAEW treatment was different between iron materials, kits and clothing surfaces (iron > kit > clothing).

When compared to the control (sterilization deionized water, DW) and UV group, spraying with SAEW, CP, and UV + SAEW reduced the populations of *S. enteritidis* on the surface of facilities ($P < 0.05$). There are differences between SAEW and CP on coops, stainless steels and glasses ($P < 0.05$), and there were no differences between SAEW and CP on tires ($P > 0.05$). Deza et al. (2005) and Ni et al. (2016) obtained a similar report. Ni et al. (2016) showed that treatment with SAEW at an ACC of 30 to 90 mg/L was statistically more effective than treatment with CP in reducing the populations of *E. coli*, *S. typhimurium* and *S. aureus* on stainless steel ($P < 0.05$). They also found no significant differences in bactericidal efficiency between SAEW and CP for reducing total aerobic bacteria in the vehicles ($P > 0.05$) under the presence of organic matter. Our findings also found that only 0.71 and 0.69 log₁₀ CFU/cm² reduction on tires was obtained after SAEW (ACC, 70 mg/L) and CP (1%) single treatment for 80 s, respectively. It was speculated that SAEW efficiency could be affected by the presence of feces. Moreover, low bacterial reduction on tire surfaces might be explained by an insufficient short time (80 s, in this study) or a low ACC (70 mg/L) of treatment and that tire surfaces are a rough material for decontamination, which strongly attach bacterial cells and feces compared to smooth surfaces (stainless steel, glass, coop). These findings also demonstrate that single treatment with SAEW on some rough material surfaces under feces interference in poultry farm may need a longer treatment time and/or a higher ACC than on other smooth surfaces. Zang et al. (2015b) also reported that in the presence of the feces soiling, to obtain the reduction of 1.38 log₁₀ CFU/cm² for *E. coli* and *S. enteritidis* mixture on the vehicle tire, the surface needed to be treated with tap water washing for 4 min followed by SAEW treatment for 5 min at an ACC of 140 mg/l.

However, the synergistic effect of SAEW combined with UV gave a higher *S. enteritidis* inactivation than SAEW single treatment ($P < 0.05$). These findings showed that UV+SAEW treatment was effective in disinfection of *S. enteritidis* on the facility surfaces. Some studies have also demonstrated that ultraviolet light is more efficient when used in combination with other disinfectants. Ekundayo (2011) have investigated the efficacy of combining electrolyzed oxidizing water and UV light on the microbiological quality of fresh jalapeño peppers. They found that peppers treated with UV and EO water produced the best microbial inhibition. Turtoi and Borda (2014) reported that *Salmonella* was effectively inactivated on egg shells in a short time and at

low temperature with the use of a combination of UV light and ozone treatment or UV light and H₂O₂ treatment. Pang and Hung (2016) also demonstrated that the combined treatment of SAEW and UV-ozonated water in the spray washing process could more effectively reduce *E. coli* O157: H7 on lettuce. These findings suggested that the combination treatment of UV and SAEW may be a promising disinfection method and could be used instead of SAEW alone, especially on rough materials in the presence of organic matter.

Model Fitting

Multiple linear regression analysis was applied to analyze the influence of treatment time and ACC on the inactivation of *S. enteritidis* by UV + SAEW on the surfaces of plastic coops. The variables used in the experimental design are listed in Table 4. Multiple regressions were performed to model the equation. The general model equation was:

$$y = -0.039x_1 - 0.027x_2 + 5.87 \quad (1)$$

where y is the surviving population value in log₁₀ CFU/cm²; x₁ is the ACC in mg/L; and x₂ is the treatment time in s. The adjusted determination R² was 0.93. The adjusted R² higher than 0.90 indicated that no significant terms were missed by the model (Myers, 1976). Moreover, the quality of fitness models were assessed by a lack of fit test ($P > 0.05$), which determines model accuracy. The linear coefficients (x₁ and x₂) were significant ($P < 0.05$).

The significantly linear coefficients of x₁ and x₂ mean that the bactericidal efficiency of UV + SAEW is significantly affected by ACC and time ($P < 0.05$). As shown in Table 4, the bactericidal efficiency of UV + SAEW increased with increasing available chlorine and increasing time. The initial population of *S. enteritidis* on the surface of coops was approximately 4.63 log₁₀ CFU/cm². 100% inactivation of *S. enteritidis* was obtained by using UV + SAEW with an ACC of 90 mg/L for more than 70 s.

CONCLUSION

In conclusion, our findings indicated that the combination treatment of UV and SAEW may improve the microbiological quality of *S. enteritidis* in pure cultures or on the facility surfaces compared to the application of SAEW alone in the presence of organic matter. The bactericidal efficiency of UV + SAEW is significantly affected by ACC and time ($P < 0.05$) and is increased with increasing ACC and increasing time. The lower ACC and the reduced treatment time of UV + SAEW treatment compared to SAEW alone on facility surfaces make it a suitable option in controlling microbial contamination on the facility surfaces in poultry farms, especially on the rough material surfaces in the presence of organic matter.

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CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest.

REFERENCES

- Cao, W., Z. Zhu, Z. Shi, C. Wang, and B. Li. 2009. Efficiency of slightly acidic electrolyzed water for inactivation of *Salmonella enteritidis* and its contaminated shell eggs. *Int. J. Food Microbiol.* 130:88–93.
- Chavez, C., K. D. Knape, C. D. Coufal, and J. B. Carey. 2002. Reduction of eggshell aerobic plate counts by ultraviolet irradiation. *Poult. Sci.* 81:1132–1135.
- DeBenedictis, P., M. S. Beato, and I. Capua. 2007. Inactivation of avian influenza viruses by chemical agents and physical conditions: a review. *Zoonoses Pub. Health* 54:51–68.
- Deza, M., M. Araujo, and M. Garrido. 2005. Inactivation of *Escherichia coli*, *Listeria monocytogenes*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* on stainless steel and glass surfaces by neutral electrolyzed water. *Lett. Appl. Microbiol.* 40:341–346.
- De Reu, K., K. Grijspeerdt, L. Herman, M. Heyndrickx, M. Uyttendaele, J. Debevere, F. F. Putirulan, and N. M. Bolder. 2006. The effect of a commercial UV disinfection system on the bacterial load of shell eggs. *Lett. Appl. Microbiol.* 42:144–148.
- Gabriel, A. A., D. D. Vera, O. M. Y. Lazo, V. B. Azarcon, C. G. De Ocampo, J. C. Marasigan, and G. T. Sandel. 2017. Ultraviolet-C inactivation of *Escherichia coli* O157: H7, *Listeria monocytogenes*, *Pseudomonas aeruginosa*, and *Salmonella enterica* in liquid egg white. *Food Control.* 73:1303–1309.
- Goerzen, P. R., and T. A. Scott. 1995. Ultraviolet light sanitation for broiler hatching eggs. *Poult. Sci.* 74:83.
- Gomez-Lopez, V. M., P. Ragaert, J. Debevere, and F. Devlieghere. 2007. Pulsed light for food decontamination: a review. *Trends Food Sci. Technol.* 18:464–473.
- Gulati, B. R., P. B. Allwood, C. W. Hedberg, and S. M. Goyal. 2001. Efficacy of commonly used disinfectants for the inactivation of calicivirus on strawberry, lettuce, and a food-contact surface. *J. Food Prot.* 64:1430–1434.
- Gradel, K. O., A. R. Sayers, and R. H. Davies. 2004. Surface disinfection tests with *Salmonella* and a putative indicator bacterium, mimicking worst-case scenarios in poultry houses. *Poult. Sci.* 83:1636–1643.
- Hao, X. X., B. M. Li, C. Y. Wang, and W. Cao. 2013. Application of slightly acidic electrolyzed water for inactivating microbes in a layer breeding house. *Poult. Sci.* 92:2560–2566.
- Jung, S., B. S. Ko, H. Jang, H. J. Park, and S. Oh. 2018. Effects of slightly acidic electrolyzed water ice and grapefruit seed extract ice on shelf life of brown sole (*Pleuronectes herzensteini*). *Food Sci. Biotech.* 27:261–267.
- Koide, S., J. Takeda, J. Shi, H. Shono, and G. G. Atungulu. 2009. Disinfection efficacy of slightly acidic electrolyzed water on fresh cut cabbage. *Food Control.* 20:294–297.
- Liu, C. C., and Y. C. Su. 2006. Efficiency of electrolyzed oxidizing water on reducing *Listeria monocytogenes* contamination on seafood processing gloves. *Int. J. Food Microbiol.* 110:14–15.
- Mcdaniel. 2011. Hatchability of broiler breeder eggs sanitized with a combination of ultraviolet light and hydrogen peroxyde. *Int. J. Poult. Sci.* 10:320–324.
- Myers, R. H. 1976. Response Surface Methodology. Ann Arbor, Michigan: Edwards Brothers Inc. Distributors.
- Mun, S., S. H. Cho, T. S. Kim, B. T. Oh, and J. Yoon. 2009. Inactivation of *Ascaris* eggs in soil by microwave treatment compared to UV and ozone treatment. *Chemosphere.* 77:285–290.
- Ni, L., W. Cao, W. C. Zheng, Q. Zhang, and B. M. Li. 2015. Reduction of microbial contamination on the surfaces of layer houses using slightly acidic electrolyzed water. *Poult. Sci.* 94:2838–2848.
- Ni, L., W. Cao, W. C. Zheng, Q. Zhang, and B. M. Li. 2016. Application of slightly acidic electrolyzed water for decontamination of stainless steel surfaces in animal transport vehicles. *Prev. Vet. Med.* 133:42–51.
- Ekundayo, O. R. 2011. Efficacy of electrolyzed oxidizing (EO) water combined with UV-C light or gas exchange on sanitization of Jalapeno pepper using *Serratia marcescens* as surrogate for *Salmonella*. https://ir.library.oregonstate.edu/concern/graduate_thesis_or_dissertations/6969z3692
- Oomori, T., T. Oka, T. Inuta, and Y. Araka. 2000. The efficacy of disinfection of acidic electrolyzed water in the presence of organic materials. *Anal. Sci.* 16:465–469.
- Pang, Y., and Y. Hung. 2016. Efficacy of slightly acidic electrolyzed water and UV-ozonated water combination for inactivating *Escherichia coli* O157:H7 on Romaine and iceberg lettuce during spray washing process. *J. Food Sci.* 81:1585–1592.
- Raccicot, M., D. Venne, A. Durivage, and J. P. Vaillancourt. 2012. Evaluation of strategies to enhance biosecurity compliance on poultry. *Prev. Vet. Med.* 103:208–218.
- Sheng, X. W., D. Shu, X. Tang, and Y. T. Zang. 2018. Effects of slightly acidic electrolyzed water on the microbial quality and shelf life extension of beef during refrigeration. *Food Sci. Nutr.* 6:1975–1981.
- Turtoi, M., and D. Borda. 2014. Decontamination of egg shells using ultraviolet light treatment. *World Poult. Sci. J.* 70:265–278.
- Totton, S. C., A. M. Farrar, W. Wilkins, O. Bucher, L. A. Waddell, B. J. Wilhelm, S. A. McEwen, and A. Rajic. 2012. A systematic review and meta-analysis of the effectiveness of biosecurity and vaccination in reducing *Salmonella* spp. in broiler chickens. *Food Res. Int.* 45:617–627.
- Unluturk, S., M. R. Atilgan, H. Baysal, and C. Tari. 2007. Use of UV-C radiation as non-thermal process for liquid egg products (LEP). *J. Food Eng.* 85:561–568.
- Zang, Y. T., B. M. Li, Sh. Bing, and W. Cao. 2015a. Modeling disinfection of plastic poultry transport coops inoculated with *Salmonella enteritidis* by slightly acidic electrolyzed water using response surface methodology. *Poult. Sci.* 94:2059–2065.
- Zang, Y. T., X. Sh. Li, B. M. Li, and W. Cao. 2015b. Simulation of disinfection optimization of vehicle tire surface using slightly acidic electrolyzed water. *Transactions CSAE.* 31:199–204.
- Zang, Y. T., B. M. Li, Z. X. Shi, X. W. Sheng, H. X. Wu, and D. Q. Shu. 2017a. Inactivation efficiency of slightly acidic electrolyzed water against microbes on facility surfaces in a disinfection channel. *Inter. J. Agr. Biol. Eng.* 10:23–30.
- Zang, Y. T., B. M. Li, W. Zheng, H. X. Wu Sheng, and D. Q. Shu. 2017b. Influence of droplet size and deposition on slightly acidic electrolyzed water spraying disinfection effect on livestock environment. *Transactions CSAE.* 33:224–229.