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Peracetic acid (PAA) exhibits higher antimicrobial efficacy compared to chlorine-based disinfectants against *Salmonella* Typhimurium on chicken skin and food-contact surfaces

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ABSTRACT

Outbreaks of *Salmonella*, especially in the poultry industry, pose significant food safety and public health challenges. These outbreaks are often intensified by the limited effectiveness of widely used disinfectants, including chlorine-based agents like chlorine dioxide (ClO_2) and sodium hypochlorite (NaOCl), particularly against bacterial biofilms. Biofilm formation on food-contact surfaces shields *Salmonella* from sanitizers, leading to persistent contamination and increased resistance, emphasizing the need for more effective disinfection strategies. This study aimed to evaluate and compare the bactericidal and antibiofilm efficacy of peroxyacetic acid (PAA) with ClO_2 and NaOCl against *Salmonella enterica* serovar Typhimurium (*S.* Typhimurium) on various food-contact surfaces, including stainless steel, plastic, silicon rubber, and chicken skin. PAA demonstrated the highest bactericidal and antibiofilm effectiveness, requiring lower concentrations than ClO_2 and NaOCl, even in organic load-rich environments. Confocal microscopy and protein leakage assays demonstrated that PAA induces significant cell wall disruption of *S.* Typhimurium planktonic cells, unlike ClO_2 and NaOCl, which were less effective under similar conditions. These findings suggest that PAA is a superior alternative for controlling *Salmonella* in poultry processing facilities, offering greater applicability for sanitation in processing facilities to enhance overall food safety.

Introduction

The rapid expansion of global poultry production and trade has increased opportunities for contamination with various foodborne pathogens, leading to outbreaks worldwide. Among these pathogens, *Salmonella* poses a particularly serious public health risk, often resulting in infections such as salmonellosis or gastroenteritis (Tan et al., 2022). According to the World Health Organization (WHO), over 2 billion cases of diarrheal disease occur globally each year, with approximately 1.3 billion cases attributed specifically to salmonellosis (Popa and Papa, 2021; Sun et al., 2021). In the United States alone, salmonellosis impacts about 1.35 million people annually, leading to 26,500 hospitalizations and 420 fatalities (Centers for Disease Control and Prevention [CDC], 2024). In Europe, *Salmonella* outbreaks continue to be significant, with 1,067 outbreaks reported in 2016, impacting over 9,000 individuals across EU Member States (European Food Safety Authority, 2017).

Among *Salmonella enterica* serovars, *S. enterica* serovar Typhimurium is particularly concerning, accounting for 19 % of human salmonellosis cases (Seo et al., 2020). Its broader host range, including poultry and human, along with the rapid acquisition of antimicrobial resistance, has led the pathogenicity status of this serovar strain to alarming levels (Wang et al., 2019). Furthermore, in food processing facilities, *S.* Typhimurium readily forms biofilms on various surfaces, such as stainless steel, plastic, glass, and rubber. These biofilms act as persistent sources of contamination in the poultry processing industry, facilitating cross-contamination during processing and contributing to the spread of antimicrobial resistance genes (Chia et al., 2009; Kim et al., 2022; Mahamud et al., 2022). Chicken skin, in particular, is a hotspot for contamination and provides an ideal environment for bacterial adherence and growth. Its microstructures conducive to bacterial adherence,

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including bushy feather follicles, deep channels, and cracks, as well as direct exposure to air and equipment surfaces, make it highly susceptible to cross-contamination during slaughtering or processing (Byun et al., 2022; Rouger et al., 2017; Zhang et al., 2020). Consequently, controlling foodborne pathogens and understanding their survival mechanisms in poultry processing facilities have become essential to maintaining safety and hygiene in global meat-based food production systems (Byun et al., 2021).

To combat these risks, poultry processing industries widely use chemical disinfectants, including chlorine-based chlorine dioxide (ClO_2) and sodium hypochlorite (NaOCl), due to their low cost, availability, and effectiveness against *Salmonella* (Byun et al., 2021). However, the effectiveness of these chlorine-based disinfectants is often questioned due to the probability of losing effectiveness after interacting with organic matter (Byun et al., 2022). Additionally, the corrosiveness, consumer avoidance, and residual/carcinogenic effects of chlorine-based disinfectants on human health, along with alterations in chicken appearance, raise the demand for safer and more effective alternative options in poultry processing industries (Ae Kim et al., 2017; Domínguez Henao et al., 2018). In light of the challenges, peroxyacetic acid (PAA), a combination of acetic acid and hydrogen peroxide, has emerged as a promising agent for poultry processing industries to combat *Salmonella* effectively (Kataria et al., 2020). According to the U. S. Food and Drug Administration (FDA) and the EFSA Panel on Biological Hazards (BIOHAZ), peroxyacetic acid (PAA) is considered a safer and more environmentally friendly antimicrobial option. Under FDA regulation (21 CFR 173.370), it is approved for use on meat and poultry carcasses, parts, trim, and organs at a maximum concentration of 220 ppm, effectively addressing concerns about chemical residues and toxicity in food applications (EFSA Panel on Biological Hazards [BIOHAZ], 2014; U.S. Food and Drug Administration [USFDA], 2017). Besides, the strong oxidation capacity, less prone to organic matters, cost-effectiveness, and short exposure time make it a promising choice for large-scale applications, particularly in poultry carcass processing (Bauermeister et al., 2008; Domínguez Henao et al., 2018).

However, while PAA, ClO_2, and NaOCl are well-established for general use in food processing, limited knowledge exists about their efficacy under varied processing conditions, particularly against *S.* Typhimurium biofilms on chicken skin and other food-contact surfaces. Therefore, this study aims to assess the bactericidal and antibiofilm effectiveness of PAA compared to ClO_2 and NaOCl against *S.* Typhimurium. Specifically, it evaluates the efficacy of each disinfectant under different processing conditions and on various surfaces, including stainless steel, plastic, and chicken skin. This research addresses a critical gap in knowledge, providing insights into optimizing disinfection practices to enhance food safety in poultry processing.

Materials and methods

Strain selection and inoculum preparation

Salmonella enterica serovar Typhimurium ATCC 14028, NCCP 14772, and a wild strain (#Wild 3) were used for this study. We previously isolated six wild strains of *S.* Typhimurium from raw chicken skin samples purchased at a local market in Anseong-si, Gyeonggi-do, Korea. Among them, the #Wild 3 (isolated from breast skin) was selected based on its relatively higher planktonic growth in Tryptic soy broth (TSB, BD Difco, Franklin Lakes, NJ, USA) and stronger biofilm formation on stainless steel, as determined by preliminary screening experiments (Table 1). Planktonic and biofilm data were obtained using the standard spread plate count method on xylose lysine deoxycholate (XLD) agar (Oxoid, Thermo Fisher Scientific Inc., Waltham, MA, USA) agar plates. Bacterial stocks in 15 % glycerol were stored at −80°C for an extended period of use.

To create a wild-like condition by cohabiting multiple strains, we prepared a bacterial cocktail by co-culturing the selected strains of *Salmonella (S.* Typhimurium ATCC 14028, NCCP 14772, and wild strain #Wild 3). Strain compatibility was assessed in preliminary planktonic growth experiments (data not shown), confirming no dominance or inhibitory effects among the strains. For cocktail preparation, 40 µL of each strain was added to 10 mL of TSB and incubated overnight at 37 °C. Following incubation, a standardized suspension of the resulting cocktail (10^5 CFU/mL) was prepared in 0.1 % buffered peptone water (BPW; Oxoid) to execute further experiments.

Preparation of the disinfectants

In the study, the different concentrations of each disinfectant were prepared before each experiment by diluting the commercially available concentrated forms with sterile distilled water (DW): a 16 % solution of peroxyacetic acid (PAA, Daesung C&S Co., Seoul, South Korea), a 200 ppm chlorine dioxide (ClO_2, LifeClean, Uddevalla, Sweden), and a 4 % solution of sodium hypochlorite (NaOCl, Yuhan Clorox Co. Ltd, Seoul, Korea). The chlorine content in ClO_2 and NaOCl was measured and calibrated by the Hanna portable photometer (Hanna Instruments, Inc., Woonsocket, RI, USA) before diluting them with distilled water (DW).

Anti-bacterial efficacy assessments

Susceptibility tests of the disinfectants: The efficacy of three different disinfectants was analyzed against the planktonic cocktail culture of *Salmonella* Typhimurium by following an established laboratory protocol (Byun et al., 2021). Briefly, overnight cultured standardized suspension of *S.* Typhimurium cocktail (10^5 CFU/mL; see 'Strain Selection and Inoculum Preparation' section) was incubated with different concentrations of ClO_2, NaOCl, and PAA for 1 and 5 min, followed by neutralization with Dey–Engley neutralizing broth (D/E broth; Difco, Becton Dickinson, Franklin Lakes, NJ, USA). The concentrations of the disinfectants were chosen until the bacterial load reached an undetectable level. Then, each sample was incubated under two different conditions: clean, containing 0.3 % bovine albumin serum (BSA), and dirty, containing 3.0 % bovine albumin serum conditions to mimic real-world scenarios in poultry processing environments (Byun et al., 2021; Kim et al., 2023). The concentrations used in clean conditions were as follows: ClO_2 (10, 20, 30, 40, 50 ppm), NaOCl (50, 100, 150, 200 ppm), and PAA (10, 20, 30, 40, 50 ppm). For dirty conditions, the concentrations were ClO_2 (40, 60, 100, 160, 180 ppm), NaOCl (75, 150, 225, 300 ppm), and PAA (10, 20, 30, 40, 50 ppm).

Confocal Laser Scanning Microscopy (CLSM) analysis: The antimicrobial activities of the sanitizers were visually observed for the treated (ClO_2: 40 ppm, NaOCl: 100 ppm, PAA: 30 ppm; exposure time: 5 min) and non-treated cultures by CLSM analysis for both clean and dirty environment conditions, by following the established laboratory protocols (Mahamud et al., 2024; Nahar et al., 2025). The FilmTracer™

Table 1
Planktonic and biofilm formation of different strains of *Salmonella* Typhimurium.

Condition	*Salmonella* Typhimurium strains							
	ATCC 14028	NCCP 14772	Wild 1	Wild 2	Wild 3	Wild 4	Wild 5	Wild 6
Planktonic	9.4 ± 0.13	9.4 ± 0.02	8.3 ± 0.01	8.6 ± 0.02	9.2 ± 0.05	8.5 ± 0.09	8.5 ± 0.02	8.6 ± 0.07
Biofilm	5.3 ± 0.07	4.1 ± 0.15	4.4 ± 0.17	4.0 ± 0.11	4.8 ± 0.02	4.1 ± 0.10	4.8 ± 0.04	4.7 ± 0.01

LIVE/DEAD® Biofilm Viability Kit (Thermo Fisher Scientific Inc., USA) was used to check the live and dead bacteria stained with SYTO 9 and propidium iodide (PI). Stained samples were imaged by a confocal microscope (LSM800Airy; Carl Zeiss, Oberkochen, Germany) equipped with the 40 × (1.2-mm-wide) water immersion objective.

Protein leakage assay: To determine the bactericidal actions of disinfectants via cell membrane damage-mediated intracellular protein leakage was tested against planktonic cells of *S.* Typhimurium, with slightly modified protocols (Mahamud et al., 2024; Zeng et al., 2010). Briefly, overnight cultured cocktail *S.* Typhimurium cells were centrifuged at 10,000 rpm for 10 min at 4°C and washed twice with 10 mL PBS to remove cell debris. A 250 µL bacterial suspension (10^9 CFU/mL) was treated with 1 mL PAA at different concentrations (10, 20, 30, and 50 ppm) for 5 min. Then, treated samples were neutralized with an equal volume (1:1, v/v) neutralizer (0.1 N Sodium thiosulfate diluted in PBS) to prevent extended membrane-damaging actions of disinfectants. After 5 min of neutralization, samples were centrifuged at 12,000 rpm for 3 min, and 100 µL of supernatant was stained with 200 µL of diluted protein assay dye reagent concentrate (Bio-Rad, Hercules, CA, USA) for 5 min. After 5 min, 200 µL of stained samples were taken into 96-well plates and measured for absorbance with a microplate reader at 595 nm. Untreated bacterial supernatants that received only protein-dye were marked as the control group, while protein dye alone was marked as blank.

Antibiofilm efficacy assessments

Food-contact surface and chicken skin preparation: For biofilm growth, food-contact surfaces and chicken skin surfaces were prepared according to the established laboratory protocols (Kim et al., 2023; Mahamud et al., 2024). Three commonly used food-contact surfaces, ultra-high-weight polyethylene (plastic), silicon rubber, and stainless steel, were selected for this study. Each surface (2 × 2 cm^2) was washed with detergent and washed twice with DW. All surfaces were autoclaved (121°C) and dried in a hot air oven (60°C) until use.

Fresh chicken skin was purchased from a local grocery store (Anseong-si, Korea) and prepared for the study according to the established laboratory protocols (Byun et al., 2022; Kim et al., 2023). The chicken skin was washed thrice with DW, and the extra fat content was removed from the skin. Surfaces were cut into (2 × 2 cm^2) using sterile scissors and dipped in 70 % alcohol for 30 s, washed with DW, and dried in the safety cabinet under the UV light (Sankyo UV Co., Ltd., Seoul, South Korea) for 15 min, both sides to minimize the background microbial community. Of note, while this combination of decontamination steps is based on established methods and is generally considered effective in reducing surface microbiota, quantitative validation of microbial elimination was not performed in this study.

Biofilm disintegration assay: Antibiofilm assessment was done with different concentrations of ClO_2, NaOCl, and PAA on food contact surfaces (plastic, stainless steel, rubber) and chicken skin surfaces. For food contact surfaces, disinfectant concentrations were ClO_2 (10, 30, 50, 100 ppm), NaOCl (50, 100, 150, 300), and PAA (10, 20, 30, 50 ppm). For chicken skin surfaces, disinfectant concentrations were: ClO_2 (5, 10, 20, 30, 50, 100 ppm), NaOCl (5, 10, 20, 30, 50, 100, 150, 300), and PAA (10, 20, 30, 50 ppm) were applied.

Briefly, sterile plastic, silicon rubber, and stainless steel were submerged in 10 mL of TSB containing a cocktail culture of *S.* Typhimurium (10^5 CFU/mL). The food-contact surfaces were incubated for 24 h at 25°C in a static environment. After incubation, the food-contact surfaces were removed and washed gently with sterile DW. Treatments with different concentrations of disinfectants were applied, followed by neutralization with D/E broth. Biofilms from the surfaces were detached using glass beads (1 g, ∮2 mm; Glastechnique, Seoul, Korea), according to the previously mentioned protocol (Nahar et al., 2021).

For chicken skin surfaces, biofilms were developed using a spot inoculation method, and the formed biofilms were detached following established laboratory protocols (Byun et al., 2021, 2022; Chowdhury et al., 2024). The chicken skin surface was inoculated with 100 µL of bacterial suspension (10^5 CFU/mL) and incubated at 4 °C for 2 hours to allow bacterial adherence, followed by incubation in a 10 °C chamber for 24 hours to facilitate biofilm formation. After incubation, skin surfaces were rinsed with sterile DW (no neutralizing solution was used). Biofilms formed on the chicken skin were detached by placing samples into sterile Whirl-Pak® bags (Nasco, Fort Atkinson, WI, USA) containing 10 mL of 0.1 % BPW, followed by mechanical maceration for 2 minutes at speed level 4 using a stomacher (BagMixer 400, Interscience Co., Saint Nom., France). Detached suspensions from both coupons and chicken skin were serially diluted and plated on XLD agar for enumeration. Microbial data were analyzed using the standard plate count method.

Field Emission Scanning Electron Microscopy (FE-SEM): Antibiofilm activities were visualized for the biofilms grown on stainless steel, plastic, and chicken skin surfaces. The representative concentration of each disinfectant [ClO_2 (40 ppm), NaOCl (100 ppm), and PAA (30 ppm)] was chosen to conduct this analysis following the established laboratory protocols (Byun et al., 2021; Nahar et al., 2021). Biofilms of *Salmonella* on food contact surfaces and chicken skin were treated with disinfectants for 5 min. Samples were fixed in 2.5 % glutaraldehyde (Sigma-Aldrich, St. Louis, MO, USA) at room temperature for 4 hours, washed with PBS, and then post-fixed with 2 % osmium tetroxide (Sigma-Aldrich, St. Louis, MO, USA) for 2 hours. After fixation, dehydration was conducted using a graded ethanol series (50-100 %) and hexamethyldisilazane (HMDS; Sigma-Aldrich, St. Louis, MO, USA) solutions (33-100 %), followed by 3 hours of air drying in a fume hood. The samples were mounted on aluminium stubs using carbon tape, sputter-coated with platinum or gold-palladium, and observed using an FE-SEM (Carl Zeiss, Germany) with a 5 kV acceleration voltage and a 5-mm working distance.

Statistical analysis

All experiments were conducted in triplicate, with at least three independent biological replicates. Data from the three replicates ($n = 3$) are presented as the mean ± standard error of the mean (SEM). The graphs were built using GraphPad Prism (ver. 5.03; GraphPad Software, Boston, MA, USA). One-way ANOVA was used for statistical data analysis by the IBM SPSS statistical software (ver. 25; Endicott, NY, USA). Tukey's Post hoc test measured the level of significance ($P < 0.05$).

Results

Bactericidal efficacy of disinfectants against S. Typhimurium planktonic cells under clean and dirty conditions

The susceptibility assay aimed to evaluate the bactericidal efficacy of three disinfectants (ClO_2, NaOCl, and PAA) at different concentrations and exposure times under two different conditions: clean (no organic load) and dirty (with organic load) against the planktonic population of *S.* Typhimurium cocktail. In both clean and dirty conditions, all disinfectants showed significant ($P < 0.05$) bactericidal activity against *S.* Typhimurium, with increasing disinfectant concentrations and exposure times. In clean conditions, 40 ppm of ClO_2, 150 ppm of NaOCl, and 40 ppm of PAA were sufficient for completely eliminating the planktonic population of *S.* Typhimurium in 1 min (Fig. 1A, B, and C). While in dirty conditions, ClO_2, NaOCl, and PAA required 180, 300, and 40 ppm, respectively, to completely kill the planktonic populations of *S.* Typhimurium (Fig. 1D, E, and F). Interestingly, with the increase of exposure time from 1 min to 5 min, all disinfectants drastically reduced the planktonic population of *S.* Typhimurium and required lower disinfectant concentrations in both conditions. It was observed that at 5 min exposure time, the required bactericidal concentrations of ClO_2, NaOCl, and PAA were 30, 100, and 20 ppm, respectively, in clean conditions (Fig. 1A, B, and C), while in dirty conditions, they required 140, 225,

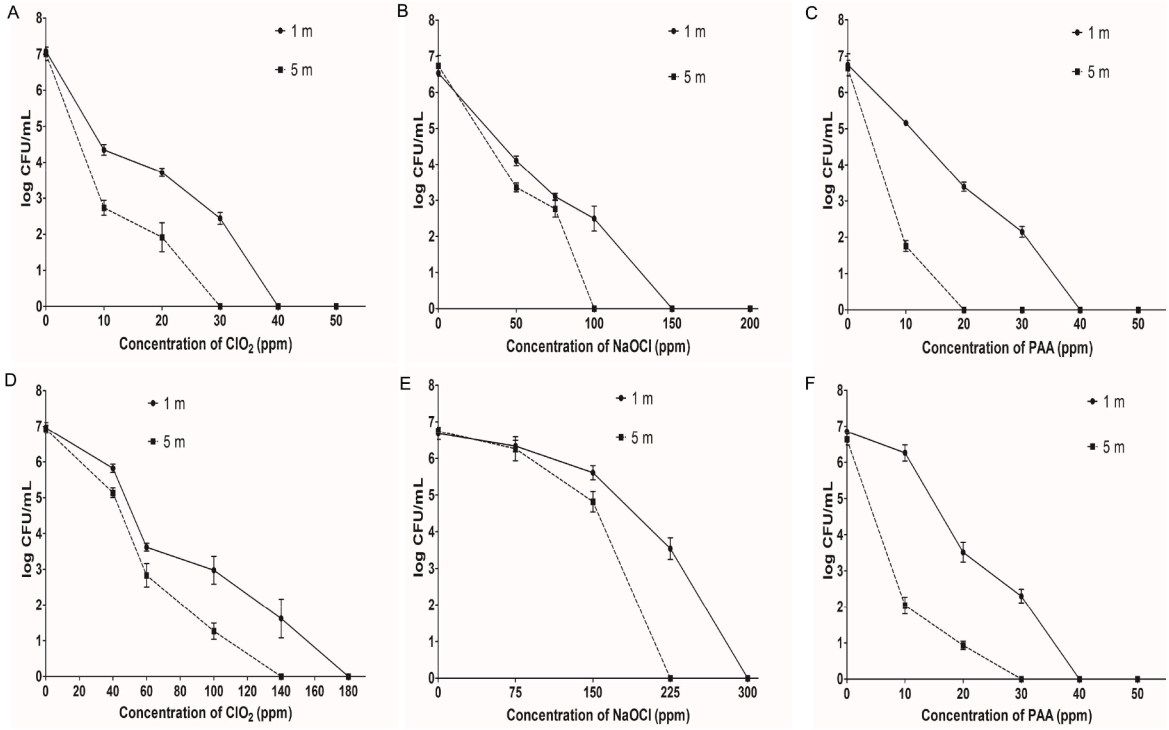


Fig. 1. Time- and concentration-dependent bactericidal activity of disinfectants against *S.* Typhimurium planktonic cells under clean and dirty conditions. Panels A, C, and E represent treatments under clean conditions with ClO_2, NaOCl, and PAA, respectively. Panels B, D, and F represent treatments under dirty conditions with the same disinfectants. Each panel shows the log CFU/mL reduction of *S.* Typhimurium planktonic cells following disinfectant treatment for 1 minute (●) and 5 minutes (■) at increasing concentrations.

and 30 ppm, respectively to completely kill the planktonic *S.* Typhimurium cells (Fig. 1D, E, and F). In summary, although ClO_2 and NaOCl showed a similar pattern of significant ($P < 0.05$) bactericidal actions in both cases, both disinfectants were less efficient in dirty conditions compared to clean conditions. In contrast, PAA showed an expeditious rate of bactericidal actions against *S.* Typhimurium planktonic cells in clean and dirty conditions by requiring lower concentrations than other disinfectants.

CLSM illustrations of bactericidal efficacy of disinfectants against S. Typhimurium planktonic cells under clean and dirty conditions

The CLSM analysis illustrated the bactericidal actions of the disinfectants on treated planktonic populations of *S.* Typhimurium compared to the non-treated control group under clean and dirty environmental conditions with a 5 min exposure period. Green fluorescence represents the viable or survived *S.* Typhimurium planktonic population (stained

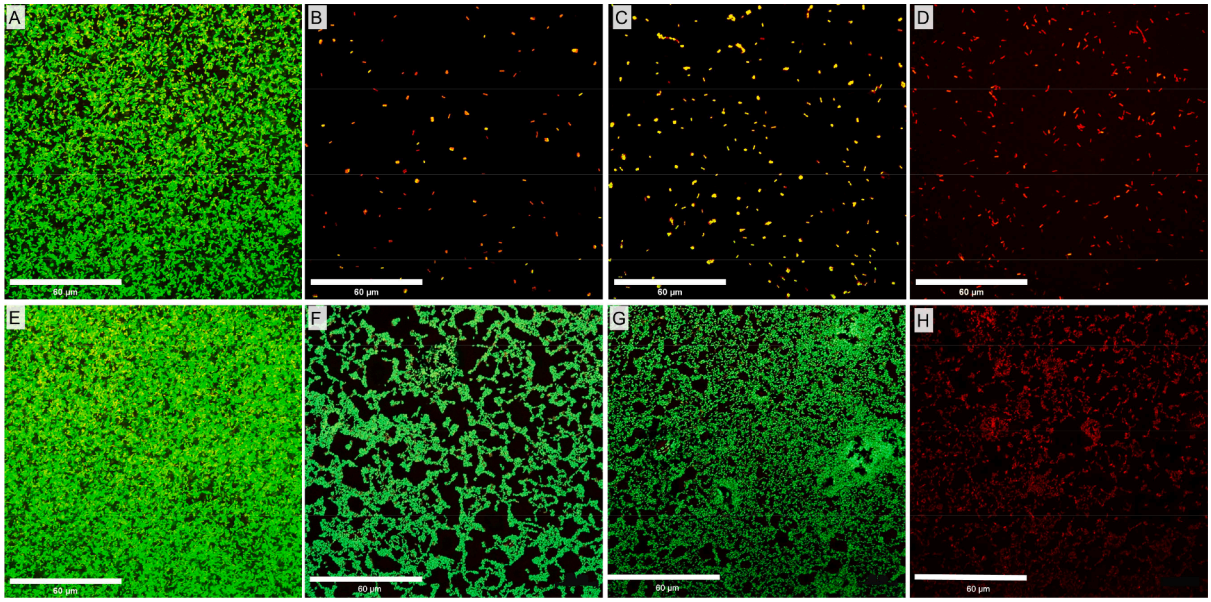


Fig. 2. Confocal laser scanning microscopic illustrations of *S.* Typhimurium planktonic cells treated with disinfectants for 1 min. Panels A and D denote untreated (control) cells under clean and dirty conditions, respectively; whereas disinfectants (ClO_2, NaOCl, and PAA)-treated planktonic cells under clean conditions are denoted by B, C, and D, respectively, and under dirty conditions are denoted by F, G, and H, respectively.

with SYTO 9), whereas injured or dead cells (stained with PI) with damaged membranes demonstrated yellow to red fluorescence. The untreated cells are exhibited as highly-dense populations in both conditions (Fig. 2A and E), while *S.* Typhimurium planktonic cells were significantly injured or killed by all selected disinfectants (ClO_2, NaOCl, and PAA) under the clean conditions within 5 min exposure time (Fig. 2B, C, and D). However, in dirty conditions, both ClO_2 (Fig. 2F) and NaOCl (Fig. 2G) showed negligible bactericidal activities against *S.* Typhimurium planktonic populations. In contrast, in dirty conditions, PAA also showed the most prominent bactericidal actions similar to clean conditions and killed all planktonic cells of *S.* Typhimurium after 5 min exposure (Fig. 2H).

Leakage of intracellular protein

Because of the potent bactericidal actions of PAA observed in the previous experiments, intracellular component leakage due to membrane-damaging effects of PAA was assessed via spectrophotometric assay to understand the mechanistic actions of PAA against planktonic cells. It was observed that at 5 min exposure, the percentages of intracellular protein leakage were significantly ($P < 0.05$) increased in planktonic cells of *S.* Typhimurium with the increment of PAA concentrations (Fig. 3). PAA, with the minimum concentration (10 ppm), showed the lowest membrane-damaging effects, while at the maximum concentration (50 ppm), the percentage of protein leakage was above 90 %. Hence, the result indicates that PAA at increasing concentrations with an increased exposure period has detrimental effects on the cell wall membranes of *S.* Typhimurium planktonic cells.

Removal efficacy of disinfectants against S. Typhimurium biofilms on different food-contact surfaces

The survivability of residential cells of *S.* Typhimurium biofilms on different food contact surfaces (i.e., SS, P, and SR) was assessed. Results showed significant ($P < 0.05$) biofilm reduction on SS, P, and SR surfaces by all selected disinfectants with the increment of disinfectant concentrations and exposure time. Amongst, ClO_2 (100 ppm), at 1 and 5 min exposure time, showed notable reduction efficacy against biofilms on the SS surface (3.6 ± 0.06 and 4.8 ± 0.03 log CFU/cm^2) (Fig. 4A), while the disinfectant showed the maximum actions against biofilms on the P (5.06±0.08 and 5.8 ± 0.01 log CFU/cm^2) (Fig. 4B), followed by SR surface (4.43±0.14 and 4.92±0.04 log CFU/cm^2) (Fig. 4C). In contrast, the resident population of *S.* Typhimurium biofilms on the SS (Fig. 4D) and P surface (Fig. 4E) showed resistance to NaOCl, even though the concentrations and exposure time were increased. However, at 5 min exposure time, NaOCl (300 ppm) reduced biofilms by 4.6 ± 0.01 log CFU/cm^2 on the SR surface (Fig. 4F).

On the other hand, at 1 min exposure time, PAA (50 ppm) significantly ($P < 0.05$) reduced biofilms on SS and P surfaces (5.3 ± 0.01 and 4.7 ± 0.16 log CFU/cm^2, respectively), while biofilms on SR showed resistance to PAA (Fig. 4I). With the increment of exposure time from 1 min to 5 min, PAA completely eradicated biofilms on SS and P surfaces (Fig. 4G, H), with a significant ($P < 0.05$) reduction rate (5.1 ± 0.36 log CFU/cm^2) in the case of SR surfaces. To sum up, the results suggest that PAA was the most potent antibiofilm agent compared to chlorine-based disinfectants, and the increasing exposure time has a greater effect on the efficacy of all disinfectants.

FE-SEM illustrations of biofilms on food contact surfaces

This study has further chosen PAA as the representative disinfectant due to its most potent antibiofilm activities and its effects on biofilm structure and adhered bacterial cells by field emission scanning electron microscopy (FE-SEM). FE-SEM illustrations of biofilms on different food contact surfaces (SS, P, and SR surfaces) demonstrated a significant difference between cell aggregation in untreated control groups (Fig. 5A, C, and E) and PAA-treated (Fig. 5B, D, and F, respectively). Particularly, it was observed that PAA markedly detached the aggregated community of *S.* Typhimurium cells on SS (Fig. 5B) and P surfaces (Fig. 5D) after 5 min exposure. However, among the three food contact surfaces, the density of attached cells on SR surfaces (Fig. 5F) was reduced by the maximum number after treating with PAA for 5 min. The FE-SEM illustrations also show evidence that PAA could remarkably damage the cell membrane structure of biofilm cells on these food contact surfaces, which indicates its potential in controlling biofilms in food industry settings.

Removal efficacy of disinfectants against S. Typhimurium biofilms on chicken skin surfaces

This study also evaluated the biofilm removal performance of the disinfectants against *S.* Typhimurium biofilms on chicken skin surfaces to explain the applications of these disinfectants in chicken processing settings. It was observed that, at 1 min exposure time, ClO_2 (100 ppm)

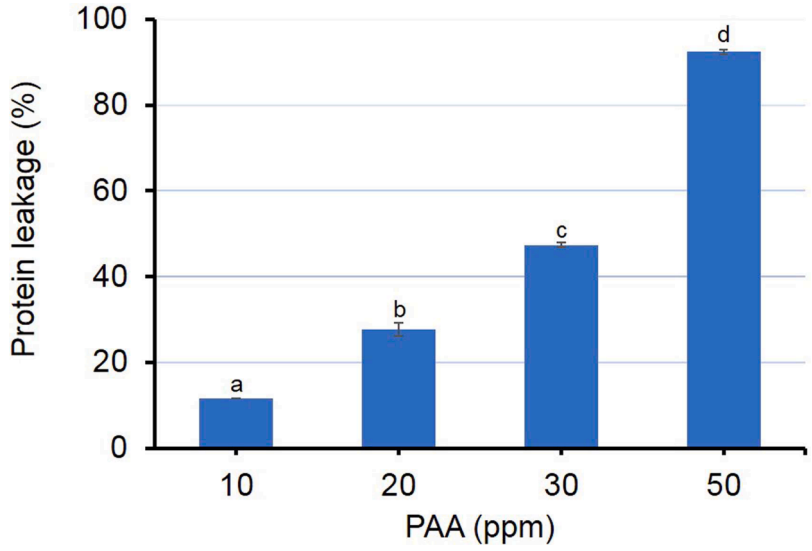


Fig. 3. Assessment of intracellular protein leakage due to PAA treatment. Different lowercase letters ($a - d$) indicate significant differences compared to the control ($P < 0.05$).

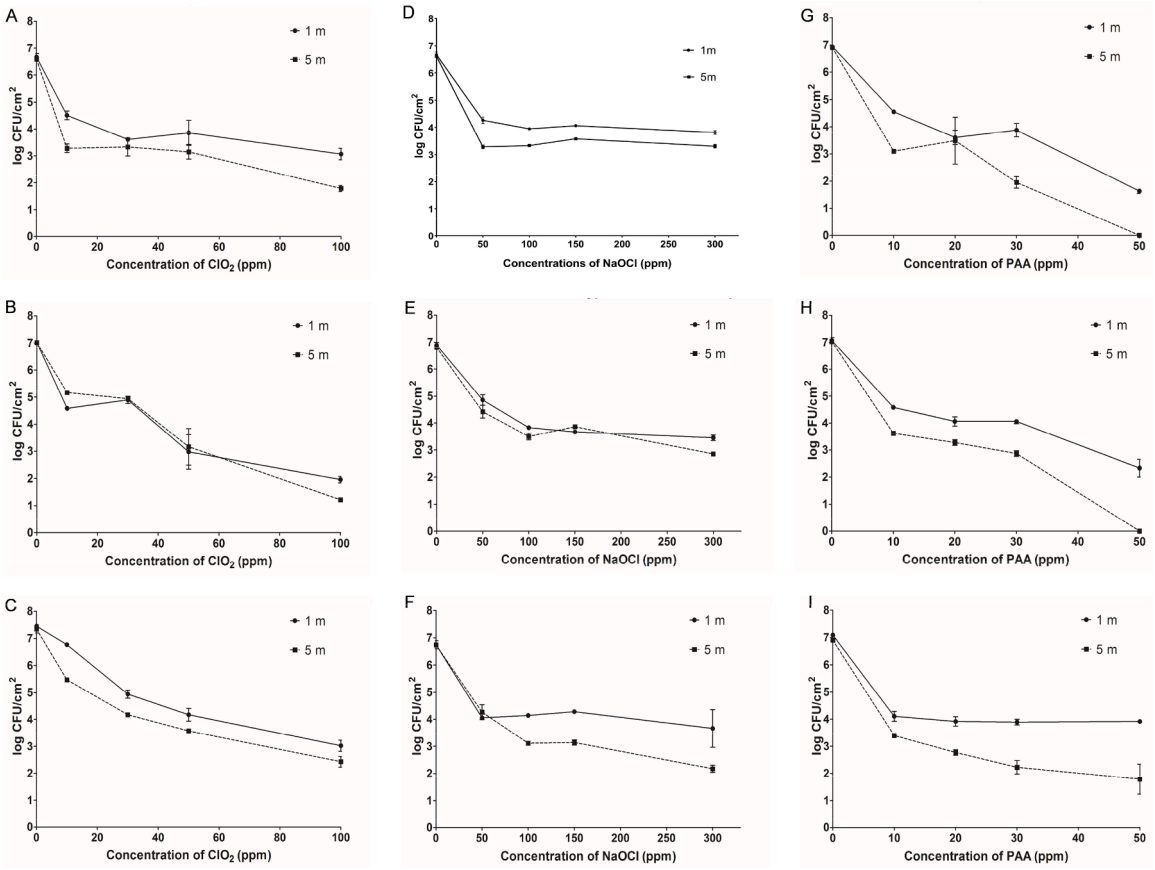


Fig. 4. Time and concentration-dependent biofilm-reducing efficacy of disinfectants against *S.* Typhimurium biofilms on different food contact surfaces. Panels A, D, and G represent biofilm reduction by ClO_2, NaOCl, and PAA, respectively, on stainless steel surfaces. Panel B, E, and H correspond to biofilm treatment on plastic surfaces, and Panels C, F, and I show biofilm reduction on silicon rubber surfaces using the same disinfectants, respectively. Each plot displays log CFU/cm^2 reductions following disinfectant treatment for 1 minute (●) and 5 minutes (■) at increasing concentrations.

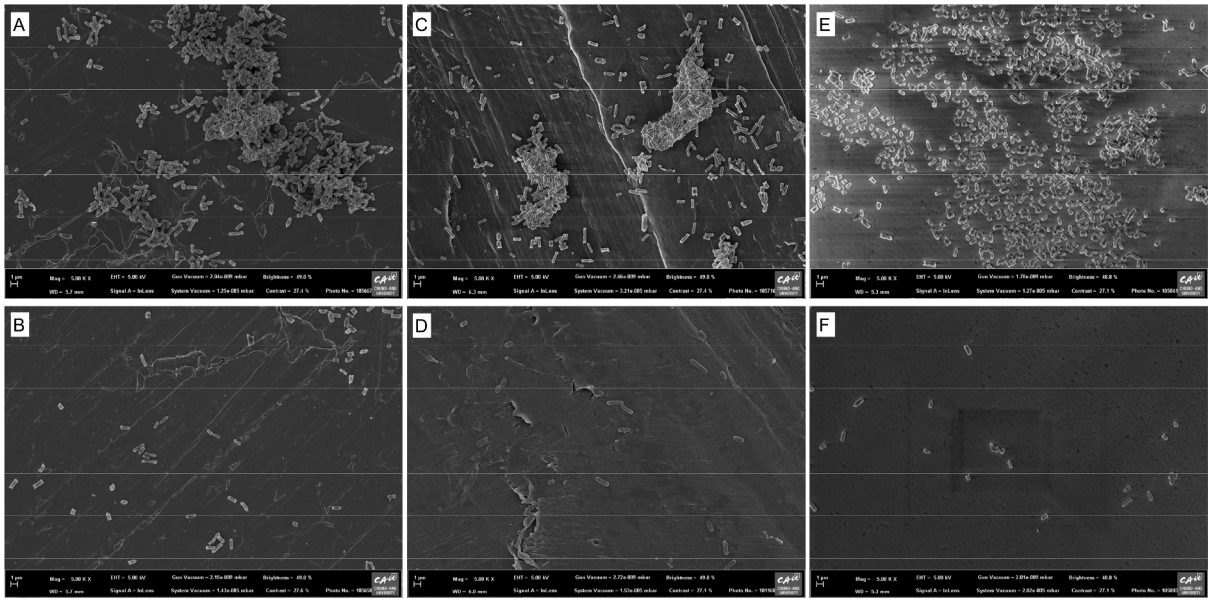


Fig. 5. Field emission scanning electron microscopy illustrations of *S.* Typhimurium biofilms formed on different food contact surfaces treated with PAA. Untreated biofilms on stainless steel, plastic, and silicone rubber surfaces are denoted by A, C, and E, respectively, while PAA-treated (5 min exposure time) *S.* Typhimurium biofilms on stainless steel, plastic, and silicone rubber are denoted by B, D, and F, respectively. Biofilms were observed at × 5000 magnification.

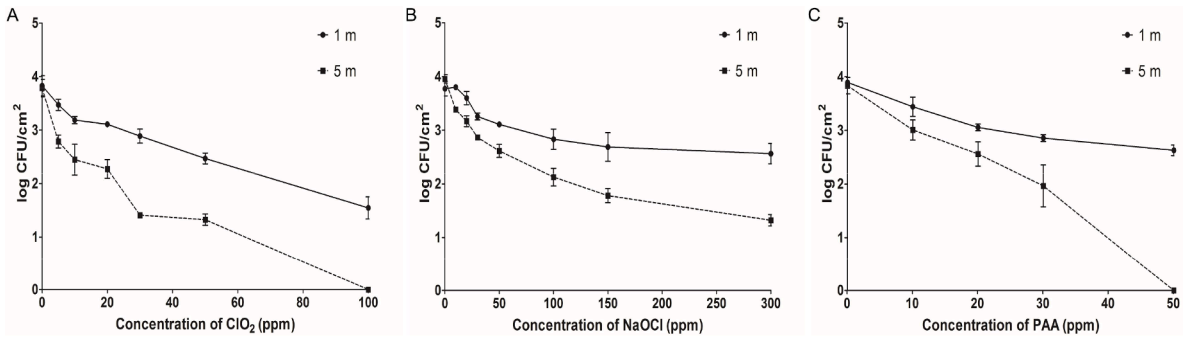


Fig. 6. Time and concentration-dependent biofilm-reducing efficacy of disinfectants against *S.* Typhimurium biofilms on chicken skin surfaces. The disinfectants (ClO_2, NaOCl, and PAA) treated biofilms on the chicken skin surfaces are denoted by A, B, and C, respectively. Each plot displays log CFU/cm^2 reductions following disinfectant treatment for 1 minute (●) and 5 minutes (■) at increasing concentrations.

significantly ($P < 0.05$) eradicated (2.3 ± 0.02 log CFU/cm^2) *S.* Typhimurium biofilms on chicken skin surfaces, while with the increasing exposure time (5 min), it completely removed biofilms from the chicken skin surfaces (Fig. 6A). In contrast, at 1 min exposure time, NaOCl (300 ppm) and PAA (50 ppm) poorly removed *S.* Typhimurium biofilms from the chicken skin surfaces (Fig. 6B and C). While with the increment of exposure time (5 min), PAA completely removed biofilms from the chicken skin surfaces; meanwhile, NaOCl could remove 2.6 ± 0.02 log CFU/cm^2 biofilms from the food surfaces. Therefore, these results suggest that increased exposure times have significant ($P < 0.05$) effects on upregulating the antibiofilm efficacy of all disinfectants in the cases of food surfaces.

FE-SEM illustrations of biofilms on chicken skin surfaces

Disinfectant-treated biofilms on chicken skin surfaces were analyzed with FE-SEM illustrations to elucidate the anti-biofilm and bactericidal actions of all disinfectants. The untreated biofilms on chicken skin surfaces were highly densely populated with undamaged cell membrane structures (Fig. 7A). At the same time, it was observed that all

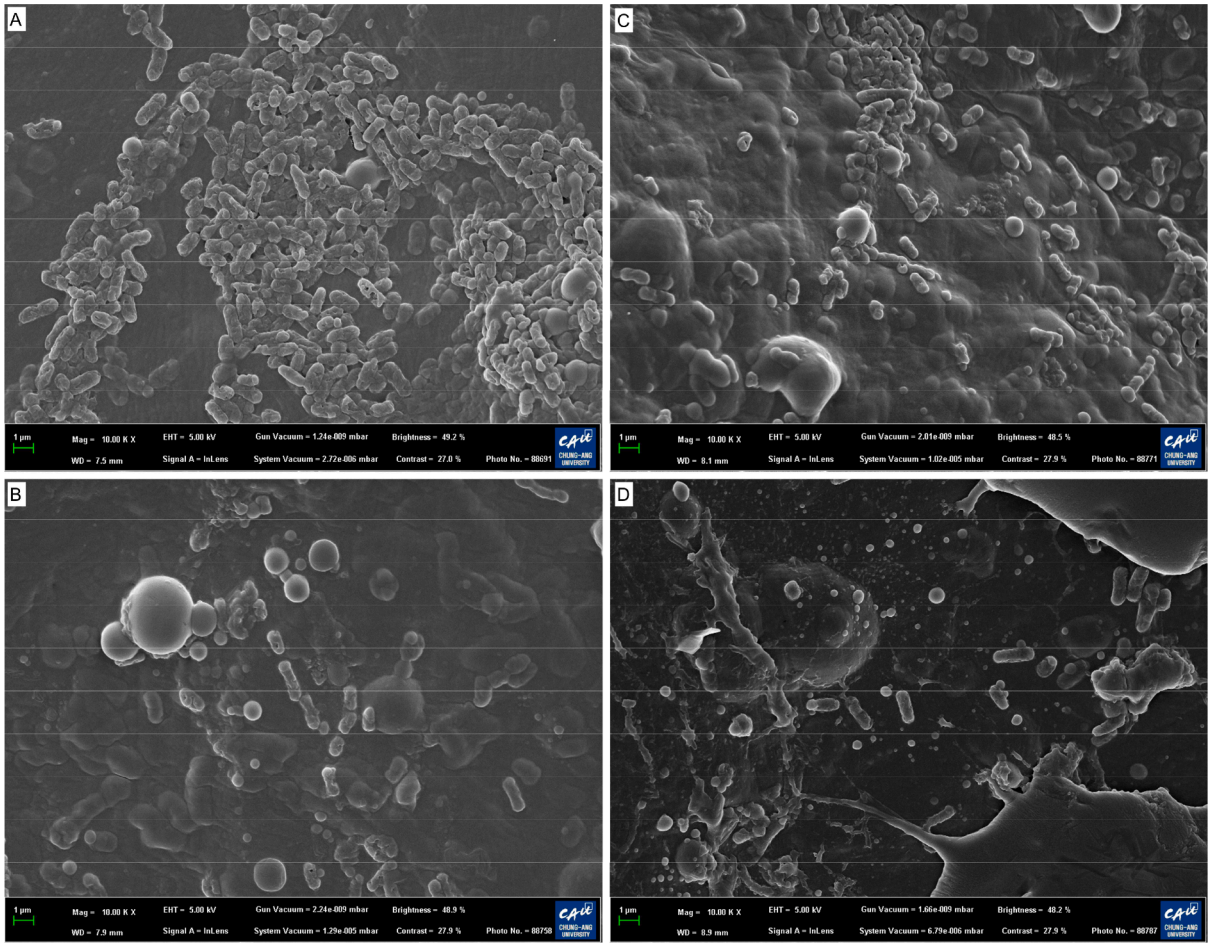


Fig. 7. Field emission scanning electron microscopy illustrations of *S.* Typhimurium biofilms formed on chicken skin surfaces treated with disinfectants for 5 minutes. Panel A denotes untreated biofilms on the chicken skin surface, whereas disinfectants (ClO_2, NaOCl, and PAA) treated biofilms on chicken skin surfaces are denoted by B, C, and D, respectively. Biofilms were observed at × 5000 magnification.

disinfectants (ClO_2, NaOCl, and PAA) remarkably disrupted biofilm structures and significantly ($P < 0.05$) reduced the number of biofilm formers after 5 min exposure period (Fig. 7B, C, and D). ClO_2 showed maximum biofilm-disrupting abilities with cell membrane-damaging actions between the two chlorine-based disinfectants. In contrast, though NaOCl partially damaged the cell structure of individually adhered cells, it had poor detachment effects on biofilms or aggregated cells on the chicken skin surfaces (Fig. 7B and C). However, compared to chlorine-based disinfectants, PAA had tremendously broken down the EPS structure of biofilms on chicken skin surfaces and showed drastic membrane-damaging effects on the resident population of the biofilms (Fig. 7D).

Discussion

Salmonellosis, a zoonotic gastrointestinal disease often caused by consuming *Salmonella*-contaminated poultry meat and eggs, presents significant challenges to global public health (Antunes et al., 2016; Djebbi-Simmons et al., 2019). The presence of *Salmonella* in poultry processing facilities indicates insufficient cleanliness and can facilitate biofilm formation, which in turn promotes cross-contamination and antimicrobial resistance (Damrongsaktrakul et al., 2021; Chowdhury et al., 2024). Therefore, effective control measures are needed to combat *Salmonella* and biofilms on food and food-contact surfaces in food processing environments.

This study evaluated the performance of various commercial disinfectants, such as PAA, ClO_2, and NaOCl, against *S.* Typhimurium in both planktonic states and biofilms on different food-contact and chicken skin surfaces. Results showed that the oxidative disinfectant PAA had the highest bactericidal and antibiofilm activity compared to the chlorine-based disinfectants ClO_2 and NaOCl, consistent with findings from a previous comparative study between PAA and NaOCl (Kim et al., 2023). Confocal laser scanning microscopy (CLSM) and protein leakage assays indicated that PAA exerts strong bactericidal effects, potentially due to cell wall denaturation or increased permeability, leading to intracellular leakage or coagulation (Ao et al., 2021; Cano et al., 2022; Dery et al., 2021; Kitis, 2004). PAA, as a potent oxidative disinfectant, generates reactive oxygen species (ROS) either directly or through reactions with other reactive chemical species. These ROS include hydroxyl radicals ($HO^{\bullet}$), acetoxyl radicals ($CH_3COO^{\bullet}$), and peroxyacetoxyl radicals ($CH_3COOO^{\bullet}$), along with the formation of acetaldehyde (CH_3CHO) (Chen et al., 2021). Thereby, this disinfectant poses oxidative stress on bacterial cells, causing structural damage to cell wall proteins or lipoprotein bilayer cytoplasmic membrane, subsequently altering cell wall permeability and leading to the oxidation of sulfhydryl and sulfur linkages in intracellular proteins, enzymes, and other essential macromolecules (Carletto et al., 2022; Shen et al., 2019).

In terms of antibiofilm efficacy, PAA, followed by ClO_2, has shown the most potent antibiofilm actions against *S.* Typhimurium biofilms on various food contact surfaces. However, our investigation found PAA and ClO_2 less effective against biofilms on silicon rubber surfaces compared to stainless steel or plastic surfaces. This might be happened due to differences in surface topography, hydrophobicity, or roughness of these food contact surfaces, as explained by another similar investigation on PAA and ClO_2 against *Listeria monocytogenes* biofilms on different food contact surfaces, such as stainless steel, low-density polyethylene, polyvinyl chloride, polyester, and rubber surfaces (Hua et al., 2019). Additional studies support these findings, showing that PAA is more effective than NaOCl against *S.* Typhimurium biofilms on various surfaces, including stainless steel, plastic, nylon, and rubber (Kim et al., 2023; Megahed et al., 2019), and is recommended for decontaminating food processing surfaces in other industries as well (Mathew et al., 2018).

Besides food contact surfaces, our study found PAA in parallel, even better in exhibiting antibiofilm actions on the food matrix. For instance, its effects on *S.* Typhimurium biofilms on beef trimmings and chicken breast fillets were found to be significant (Ellebracht et al., 2005; Kumar et al., 2020), and another study reported a notable eradication (4.8 log reduction) of pre-formed *S.* Typhimurium biofilms on chicken neck skin (Acar et al., 2022). Our previous work also found PAA as a potent antibiofilm agent against biofilms of another *Salmonella* strain (*S.* Thompson) on chicken skin (Nahar et al., 2022).

The better antibiofilm efficacies of PAA compared to chlorine-based disinfectants are mainly manifested by the differences in chemical properties, mechanisms of action, and influence of external factors. In addition, PAA shows slower interactions with organic matter or biofilm matrix constituents than free chlorine, which facilitates the oxidative disinfectant to get sufficient time to show the maximum bactericidal efficacy against planktonic cells under organic constituents-rich conditions. Likewise, PAA can easily penetrate biofilms without interacting with the organic biofilm matrix and thus shows biofilm removal or bactericidal activities against biofilm residents (Cano et al., 2022; Zhang et al., 2019). In addition, the delayed interaction feature of PAA with organic components may also contribute to effective biofilm control on chicken skin surfaces without reducing antibiofilm efficacy (Zhang et al., 2019). In contrast, chicken skin surfaces are characterized by a fat-rich, unorthodoxly pitted, and creased surface, which facilitates strong attachment of bacteria to the skin surface (Nahar et al., 2022).

On the other hand, chlorine-based disinfectants, including ClO_2 and NaOCl, usually show bactericidal actions by interacting with sulfur-containing amino acids-rich proteins of the cell membrane, leading to cell wall protein disruption, membrane permeability alterations, and intracellular oxidative stress (Bridges et al., 2022). However, their affinity for organic matter reduces their efficacy, especially under conditions rich in organic matter (World Health Organization [WHO], 2008). Thereby, in our study, both ClO_2 and NaOCl showed negligible bactericidal actions against *S.* Typhimurium planktonic populations and required higher concentrations to kill the planktonic cells in the presence of organic matter compared to clean conditions. The findings were also supported by a study on PAA against *Pseudomonas aeruginosa* PAO1, where they found that compared to NaOCl, PAA could more effectively inhibit the regrowth of biofilm cells, even in the presence of organic matter (Zhang et al., 2019).

In addition, the low diffusivity of chlorine within biofilms might be attributed to their lower antibiofilm and bactericidal actions on residential populations, as observed in other studies (Clayton et al., 2021; Gkana et al., 2017). Therefore, the study observed that both ClO_2 and NaOCl could not completely eradicate biofilms from food and food-contact surfaces. Nonetheless, in the present study, ClO_2 showed better bactericidal and antibiofilm actions against *S.* Typhimurium than NaOCl, as observed in previous studies. For instance, a study observed that even though NaOCl was effective against planktonic cells, it could not effectively remove the pre-formed biofilms of *S.* Typhimurium (Corcoran et al., 2014). Similarly, another study reported that NaOCl completely failed to eradicate *S.* Typhimurium and *Staphylococcus aureus* biofilms from stainless steel surfaces (Gkana et al., 2017). Likewise, in our previous study, ClO_2 showed comparatively higher efficacy than NaOCl against *Salmonella enterica* serovar Enteritidis planktonic cells and established biofilms on different food contact surfaces and chicken skin surfaces (Byun et al., 2021). The difference between these two chlorine-based disinfectants might be due to the difference in their structural and functional groups. For instance, ClO_2 can act in a wide range of pH due to its higher oxidation and diffusion ability (Banach et al., 2015), while NaOCl can only act within a specific pH range and shows a reduced diffusivity due to rapid NaOCl decomposition, caused by interactions with organic components of biofilms (Bridges et al., 2022; Petridis et al., 2019).

Therefore, considering the findings of the study, PAA can be recommended for eliminating *S.* Typhimurium biofilms in chicken processing plants. Nonetheless, this study has some limitations, such as the impacts of these commercial disinfectants on the EPS matrix, surface corrosiveness, and organoleptic properties of chicken skin. Therefore,

future studies should investigate these issues to suggest that disinfectants, particularly PAA, are effective antibiofilm cleaning agents in poultry slaughterhouses or the poultry processing industry.

Conclusion

This study demonstrates that PAA is a highly effective disinfectant for combating *S*. Typhimurium on both food and food-contact surfaces, outperforming chlorine-based disinfectants such as ClO_2 and NaOCl. The strong oxidative properties of PAA allow it to disseminate biofilms more effectively, even in the presence of organic material that typically reduces the efficacy of disinfectants. In contrast, ClO_2 and NaOCl, while commonly used, showed limitations under similar conditions, especially on surfaces with complex topographies where biofilms can adhere strongly.

The findings suggest that PAA could play a crucial role in enhancing hygiene practices in poultry processing plants, where persistent biofilms pose serious food safety challenges. Despite its effectiveness, this study acknowledges that further research is needed to evaluate the impact of PAA on factors such as equipment durability and the sensory qualities of treated poultry. Addressing these aspects will help confirm the potential of PAA as a leading disinfectant for safer, cleaner poultry processing environments, supporting better public health outcomes.

Data availability statement

Data sharing is not applicable as no new data were generated or analyzed during this study.

Ethical approval statement

This article does not include any studies with human or animal subjects.

Author contributions

Mahamud, A. U.: conceptualization, investigation, methodology, data curation, formal analysis, validation, visualization, and manuscript drafting. **Nahar, N.:** conceptualization, investigation, methodology, validation, visualization, and manuscript review and editing. **Ha, S. D.:** conceptualization, supervision, validation, manuscript review and editing, funding acquisition, project administration, and resource management.

Declaration of competing interest

The authors have no financial or personal conflict of interest with any person or organization reported in this work.

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References

Acar, B.O., Cengiz, G., Gülendağ, E., Göncüoğlu, M., Diker, K., 2022. Efficacy of peroxyacetic acid against *Salmonella* biofilms and as a decontamination agent in poultry meat. J. Hellenic. Vet. Med. Soc. 73, 4007–4014. https://doi.org/10.12681/jhvms.26066.

Ae Kim, S., Hong Park, S., In Lee, S., Owens, C.M., Ricke, S.C., 2017. Assessment of chicken carcass microbiome responses during processing in the presence of commercial antimicrobials using a next generation sequencing approach. Sci. Rep. 7, 43354. https://doi.org/10.1038/srep43354.

Antunes, P., Mourão, J., Campos, J., Peixe, L., 2016. Salmonellosis: the role of poultry meat. Clin. Microbiol. Infect. 22, 110–121. https://doi.org/10.1016/j.cmi.2015.12.004.

Ao, X.W., Eloranta, J., Huang, C.H., Santoro, D., Sun, W.J., Lu, Z.D., Li, C., 2021. Peracetic acid-based advanced oxidation processes for decontamination and disinfection of water: a review. Water Res. 188, 116479. https://doi.org/10.1016/j.watres.2020.116479.

Banach, J.L., Sampers, I., Van Haute, S., van der Fels-Klerx, H.J., 2015. Effect of disinfectants on preventing the cross-contamination of pathogens in fresh produce washing water. Int. J. Environ. Res. Public Health 12, 8658–8677. https://doi.org/10.3390/ijerph120808658.

Bauermeister, L.J., Bowers, J.W.J., Townsend, J.C., McKee, S.R., 2008. The microbial and quality properties of poultry carcasses treated with peracetic acid as an antimicrobial treatment. Poult. Sci. 87, 2390–2398. https://doi.org/10.3382/ps.2008-00087.

Bridges, D.F., Lacombe, A., Wu, V.C.H., 2022. Fundamental differences in inactivation mechanisms of *Escherichia coli* O157:H7 between chlorine dioxide and sodium hypochlorite. Front. Microbiol. 13. https://doi.org/10.3389/fmicb.2022.923964.

Byun, K.H., Han, S.H., Yoon, J.W., Park, S.H., Ha, S.D., 2021. Efficacy of chlorine-based disinfectants (sodium hypochlorite and chlorine dioxide) on *Salmonella* Enteritidis planktonic cells, biofilms on food contact surfaces and chicken skin. Food Control 123, 107838. https://doi.org/10.1016/j.foodcont.2020.107838.

Byun, K.H., Na, K.W., Ashrafudoulla, M., Choi, M.W., Han, S.H., Kang, I., Park, S.H., Ha, S.D., 2022. Combination treatment of peroxyacetic acid or lactic acid with UV-C to control *Salmonella* Enteritidis biofilms on food contact surface and chicken skin. Food Microbiol. 102, 103906. https://doi.org/10.1016/j.fm.2021.103906.

Cano, C., Sadat, R., Chaves, B.D., 2022. Peroxyacetic acid effectiveness against *Salmonella* on raw poultry parts is not affected by organic matter. J. Food Prot. 85, 1446–1451. https://doi.org/10.4315/jfp-22-123.

Carletto, D., Furtado, F., Zhang, J., Asimakopoulos, A.G., Eggen, M., Verstege, G.C., Faggio, C., Mota, V.C., Lazado, C.C., 2022. Mode of application of peracetic acid-based disinfectants has a minimal influence on the antioxidant defences and mucosal structures of Atlantic salmon (*Salmo salar*) Parr. Front. Physiol. 13, 900593. https://doi.org/10.3389/fphys.2022.900593.

CDC, 2024. *Salmonella* Infection (Salmonellosis). Centers for Disease Control and Prevention (CDC), United States. Accessed Dec. 2024. https://www.cdc.gov/salmonella/index.html.

Chen, J., Liu, X., Pavlostathis, S.G., 2021. Long-term evaluation of the effect of peracetic acid on a mixed aerobic culture: organic matter degradation, nitrification, and microbial community structure. Water Res. 190, 116694. https://doi.org/10.1016/j.watres.2020.116694.

Chia, T.W., Goulter, R.M., McMeekin, T., Dykes, G.A., Fegan, N., 2009. Attachment of different *Salmonella* serovars to materials commonly used in a poultry processing plant. Food Microbiol. 26, 853–859. https://doi.org/10.1016/j.fm.2009.05.012.

Chowdhury, M.A.H., Ashrafudoulla, M., Mevo, S.I.U., Mizan, M.F.R., Park, S.H., Ha, S.D., 2024. Current and future interventions for improving poultry health and poultry food safety and security: a comprehensive review. Compr. Rev. Food Sci. Food Saf. 22, 1555–1596. https://doi.org/10.1111/1541-4337.13121.

Chowdhury, M.A.H., Ashrafudoulla, M., Mevo, S.I.U., Mahamud, A.U., Reem, C.S.A., Ha, A.J.W., Ha, S.D., 2024. Efficacy of orange terpene against *Escherichia coli* biofilm on beef and food contact surfaces. Food Res. Int. 197, 115204. https://doi.org/10.1016/j.foodres.2024.115204.

Clayton, G.E., Thorn, R.M.S., Reynolds, D.M., 2021. The efficacy of chlorine-based disinfectants against planktonic and biofilm bacteria for decentralised point-of-use drinking water. npj Clean Water 4, 48. https://doi.org/10.1038/s41545-021-00139-w.

Corcoran, M., Morris, D., De Lappe, N., O'Connor, J., Lalor, P., Dockery, P., Cormican, M., 2014. Commonly used disinfectants fail to eradicate *Salmonella enterica* biofilms from food contact surface materials. Appl. Environ. Microbiol. 80, 1507–1514. https://doi.org/10.1128/aem.03109-13.

Damrongsaktrakul, P., Ruengvisesh, S., Rahothan, A., Sukhumrat, N., Tuitemwong, P., Phung-On, I., 2021. Removal of *Salmonella* Typhimurium biofilm from food contact surfaces using *Quercus infectoria* Gall extract in combination with a surfactant. J. Microbiol. Biotechnol. 31, 439–446. https://doi.org/10.4014/jmb.2101.01014.

Dery, J.L., Choppakatla, V., Sughroue, J., Rock, C., 2021. Minimizing risks: use of surface water in pre-harvest agricultural irrigation. Part III: Peroxyacetic Acid (PAA) Treatment Methods in Cooperative Extension. The University of Arizona, Tucson, AZ, USA.

Djebbi-Simmons, D., Xu, W., Janes, M., King, J., 2019. Survival and inactivation of *Salmonella enterica* serovar Typhimurium on food contact surfaces during log, stationary and long-term stationary phases. Food Microbiol. 84, 103272. https://doi.org/10.1016/j.fm.2019.103272.

Domínguez Henao, L., Delli Compagni, R., Turolla, A., Antonelli, M., 2018. Influence of inorganic and organic compounds on the decay of peracetic acid in wastewater disinfection. Chem. Eng. J. 337, 133–142. https://doi.org/10.1016/j.cej.2017.12.074.

Ellebracht, J.W., King, D.A., Castillo, A., Lucia, L.M., Acuff, G.R., Harris, K.B., Savell, J.W., 2005. Evaluation of peroxyacetic acid as a potential pre-grinding treatment for control of *Escherichia coli* O157:H7 and *Salmonella* Typhimurium on beef trimmings. Meat Sci. 70, 197–203. https://doi.org/10.1016/j.meatsci.2005.01.003.

European Food Safety Authority, European Centre for Disease Prevention and Control, 2017. The European Union summary report on trends and sources of zoonoses, zoonotic agents and food-borne outbreaks in 2016. EFSA J. 15, e05077. https://doi.org/10.2903/j.efsa.2017.5077.

Gkana, E.N., Giaouris, E.D., Doulgeraki, A.I., Kathariou, S., Nychas, G.J.E., 2017. Biofilm formation by *Salmonella* typhimurium and *Staphylococcus aureus* on stainless steel

under either mono- or dual-species multi-strain conditions and resistance of sessile communities to sub-lethal chemical disinfection. Food Control 73, 838–846. https://doi.org/10.1016/j.foodcont.2016.09.038.

EFSA Panel on Biological Hazards (BIOHAZ), 2014. Scientific opinion on the evaluation of the safety and efficacy of peroxyacetic acid solutions for reduction of pathogens on poultry carcasses and meat. EFSA J. 12, 3599. https://doi.org/10.2903/j.efsa.2014.3599.

Hua, Z., Korany, A.M., El-Shinawy, S.H., Zhu, M.J., 2019. Comparative evaluation of different sanitizers against *Listeria monocytogenes* biofilms on major food-contact surfaces. Front. Microbiol. 10, 2462. https://doi.org/10.3389/fmicb.2019.02462.

Kataria, J., Vaddu, S., Rama, E.N., Sidhu, G., Thippareddi, H., Singh, M., 2020. Evaluating the efficacy of peracetic acid on *Salmonella* and *Campylobacter* on chicken wings at various pH levels. Poult. Sci. 99, 5137–5142. https://doi.org/10.1016/j.psj.2020.06.070.

Kim, J.M., Zhang, B.Z., Park, J.M., 2023. Comparison of sanitization efficacy of sodium hypochlorite and peroxyacetic acid used as disinfectants in poultry food processing plants. Food Control 152, 109865. https://doi.org/10.1016/j.foodcont.2023.109865.

Kim, Y., Nahar, S., Cho, A.J., Mahamud, A.U., Park, S.H., Ha, S.D., 2023. Synergistic antibacterial effect of DNase I and eugenol against *Salmonella* Enteritidis biofilm on smoked duck and food contact surfaces. Food Control 150, 109764. https://doi.org/10.1016/j.foodcont.2023.109764.

Kim, Y.K., Roy, P.K., Ashrafudoulla, M., Nahar, S., Toushik, S.H., Hossain, M.I., Mizan, M.F.R., Park, S.H., Ha, S.D., 2022. Antibiofilm effects of quercetin against *Salmonella enterica* biofilm formation and virulence, stress response, and quorum-sensing gene expression. Food Control 137, 108964. https://doi.org/10.1016/j.foodcont.2022.108964.

Kitis, M., 2004. Disinfection of wastewater with peracetic acid: a review. Environ. Int. 30, 47–55. https://doi.org/10.1016/s0160-4120(03)00147-8.

Kumar, S., Singh, M., Cosby, D.E., Cox, N.A., Thippareddi, H., 2020. Efficacy of peroxy acetic acid in reducing *Salmonella* and *Campylobacter* spp. Populations on chicken breast fillets. Poult. Sci. 99, 2655–2661. https://doi.org/10.1016/j.psj.2019.12.045.

Mahamud, A.U., Ashrafudoulla, M., Nahar, S., Chowdhury, M.A.H., Park, S.H., Ha, S.D., 2024. Luteolin exhibits antimicrobial actions against *Salmonella* Typhimurium and *Escherichia coli*: impairment of cell adhesion, membrane integrity, and energy metabolism. Food Control 166, 110734. https://doi.org/10.1016/j.foodcont.2024.110734.

Mahamud, A.U., Nahar, S., Ashrafudoulla, M., Park, S.H., Ha, S.D., 2022. Insights into antibiofilm mechanisms of phytochemicals: prospects in the food industry. Crit. Rev. Food Sci. Nutr. 64, 1736–1763. https://doi.org/10.1080/10408398.2022.2119201.

Mathew, E.N., Muyyarikkandy, M..S., Bedell, C., Amalaradjou, M.A., 2018. Efficacy of chlorine, chlorine dioxide, and peroxyacetic acid in reducing *Salmonella* contamination in wash water and on mangoes under simulated mango packinghouse washing operations. Front. Sustain. Food Syst. 2, 18. https://doi.org/10.3389/fsufs.2018.00018.

Megahed, A., Aldridge, B., Lowe, J., 2019. Comparative study on the efficacy of sodium hypochlorite, aqueous ozone, and peracetic acid in the elimination of *Salmonella* from cattle manure contaminated various surfaces supported by Bayesian analysis. PLoS One 14, e0217428. https://doi.org/10.1371/journal.pone.0217428.

Nahar, S., Mahamud, A.U., Cho, A.J., Ashrafudoulla, M., Yu, J., Park, S.H., Ha, S.D., 2025. Flavourzyme suppresses *Pseudomonas aeruginosa* biofilms by targeting motility, quorum sensing, and virulence genes. Curr. Microbiol. 82, 1–13. https://doi.org/10.1007/s00284-025-04200-6.

Nahar, S., Ha, A.J., Byun, K.H., Hossain, M.I., Mizan, M.F.R., Ha, S.D., 2021. Efficacy of flavourzyme against *Salmonella* Typhimurium, *Escherichia coli*, and P*seudomonas aeruginosa* biofilms on food-contact surfaces. Int. J. Food Microbiol. 336, 108897. https://doi.org/10.1016/j.ijfoodmicro.2020.108897.

Nahar, S., Jeong, H.L., Cho, A.J., Park, J.H., Han, S., Kim, Y., Park, S.H., Ha, S.D., 2022. Efficacy of ficin and peroxyacetic acid against *Salmonella enterica* serovar Thompson biofilm on plastic, eggshell, and chicken skin. Food Microbiol. 104, 103997. https://doi.org/10.1016/j.fm.2022.103997.

Petridis, X., Busanello, F.H., So, M.V.R., Dijkstra, R.J.B., Sharma, P.K., van der Sluis, L.W.M., 2019. Chemical efficacy of several NaOCl concentrations on biofilms of different architecture: new insights on NaOCl working mechanisms. Int. Endod. J. 52, 1773–1788. https://doi.org/10.1111/iej.13198.

Popa, G.L., Papa, M.I., 2021. *Salmonella* spp. infection - a continuous threat worldwide. Germs 11, 88–96. https://doi.org/10.18683/germs.2021.1244.

Rouger, A., Tresse, O., Zagorec, M., 2017. Bacterial contaminants of poultry meat: sources, species, and dynamics. Microorganisms 5, 50. https://doi.org/10.3390/microorganisms5030050.

Seo, M.K., Na, K.W., Han, S.H., Park, S.H., Ha, S.D., 2020. Inhibitory effect of ethanol and thiamine dilaurylsulfate against loosely, intermediately, and tightly attached mesophilic aerobic bacteria, coliforms, and *Salmonella* Typhimurium in chicken skin. Poult. Sci. 99, 1571–1580. https://doi.org/10.1016/j.psj.2019.10.058.

Shen, X., Sheng, L., Gao, H., Hanrahan, I., Suslow, Trevor V., Zhu, M.J., 2019. Enhanced efficacy of peroxyacetic acid against *Listeria monocytogenes* on fresh apples at elevated temperature. Front. Microbiol. 10, 1196. https://doi.org/10.3389/fmicb.2019.01196.

Sun, T., Liu, Y., Qin, X., Aspridou, Z., Zheng, J., Wang, X., Li, Z., Dong, Q., 2021. The prevalence and epidemiology of *Salmonella* in retail raw poultry meat in China: a systematic review and meta-analysis. Foods 10, 2757. https://doi.org/10.3390/foods10112757.

Tan, S.J., Nordin, S., Esah, E.M., Mahror, N., 2022. *Salmonella* spp. In chicken: prevalence, antimicrobial resistance, and detection methods. Microbiol. Res. 13, 691–705. https://doi.org/10.3390/microbiolres13040050.

USFDA, 2017. Environmental Assessment for Food Contact Notification FCN 1811. U.S. Food & Drug Administration, Silver Spring, MD, USA.

Wang, X., Biswas, S., Paudyal, N., Pan, H., Li, X., Fang, W., Yue, M., 2019. Antibiotic resistance in *Salmonella* Typhimurium isolates recovered from the food chain through national antimicrobial resistance monitoring system between 1996 and 2016. Front. Microbiol. 10, 985. https://doi.org/10.3389/fmicb.2019.00985.

WHO, 2008. Benefits and risks of the use of chlorine-containing disinfectants in food production and food processing. In: Report of a joint FAO/WHO expert meeting. World Health Organization, Ann Arbor, MI, USA.

Zeng, X., Tang, W., Ye, G., Ouyang, T., Tian, L., Ni, Y., Li, P., 2010. Studies on disinfection mechanism of electrolyzed oxidizing water on *E. coli* and *Staphylococcus aureus*. J. Food Sci. 75, M253–M260. https://doi.org/10.1111/j.1750-3841.2010.01649.x.

Zhang, C., Brown, P.J.B., Miles, R.J., White, T.A., Grant, D.G., Stalla, D., Hu, Z., 2019. Inhibition of regrowth of planktonic and biofilm bacteria after peracetic acid disinfection. Water Res. 149, 640–649. https://doi.org/10.1016/j.watres.2018.10.062.

Zhang, X., Peng, Z., Li, P., Mao, Y., Shen, R., Tao, R., Diao, X., Liu, L., Zhao, Y., Luo, X., 2020. Complex internal microstructure of feather follicles on chicken skin promotes the bacterial cross-contamination of carcasses during the slaughtering process. Front. Microbiol. 11, 571913. https://doi.org/10.3389/fmicb.2020.571913.