Survival and Death of *Salmonella* Typhimurium and *Campylobacter jejuni* in Processing Water and on Chicken Skin during Poultry Scalding and Chilling

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ABSTRACT

*Salmonella* Typhimurium and *Campylobacter jejuni* were inoculated in scalding water, in chilled water, and on chicken skins to examine the effects of scalding temperature (50, 55, and 60°C) and the chlorine level in chilled water (0, 10, 30, and 50 ppm), associated with the ages of scalding water (0 and 10 h) and chilled water (0 and 8 h), on bacterial survival or death. After scalding at 50 and 60°C, the reductions of *C. jejuni* were 1.5 and 6.2 log CFU/ml in water and <1 and >2 log CFU/cm² on chicken skins; the reductions of *Salmonella* Typhimurium were <0.5 and >5.5 log CFU/ml in water and <0.5 and >2 log CFU/cm² on skins, respectively. The age of scalding water did not significantly (*P* > 0.05) affect bacterial heat sensitivity. However, the increase in the age of chilled water significantly (*P* < 0.05) reduced the chlorine effect. In 0-h chilled water, *C. jejuni* and *Salmonella* Typhimurium were reduced by 3.3 and 0.7 log CFU/ml, respectively, after treatment with 10 ppm of chlorine and became nondetectable with 30 and 50 ppm of chlorine. In 8-h chilled water, the reduction of *C. jejuni* and *Salmonella* Typhimurium was <0.5 log CFU/ml with 10 ppm of chlorine and ranged from 4 to 5.5 log CFU/ml with 50 ppm of chlorine. Chlorination of chilled water did not effectively reduce the bacteria attached on chicken skins. The *D*-values of *Salmonella* Typhimurium and *C. jejuni* were calculated for the prediction of their survival or death in the poultry scalding and chilling.

Poultry products are frequent vehicles of *Salmonella* and *Campylobacter*. The incidence of *Salmonella* in raw poultry carcasses is 30 to 50%, and the number of *Salmonella* ranges from 1 to 30 CFU/carcass (28). The published data for *Campylobacter* vary considerably; the incidence ranged from 30 to 100%, and the number ranged from 10² to 10⁶ CFU/carcass (28). Poultry-borne salmonellosis and campylobacteriosis cost $426 to $814 million annually in the United States (2).

The number and the incidence of *Salmonella* and *Campylobacter* in raw poultry carcasses are greatly affected by the operation conditions of scalding and chilling processes. In commercial scalding, temperature is set in a range of 50 to 60°C for 2 to 2.5 min (5). Some research results (19, 29) showed that high scalding temperature greatly reduced the bacterial survival in the scalders. Another study (10) showed that a higher scalding temperature caused the broiler skin to lose more of the stratum corneum layer, making skin easier for bacteria to adhere. Varying levels of the organic matter in scalding water also affect *D*-values of bacteria (7).

Chlorine (up to 50 ppm) is approved by the U.S. Department of Agriculture (25) and commonly applied in commercial poultry processing for the chlorination of chilled water. Chlorine at the levels of 18 to 100 ppm has been shown to be effective in reducing bacteria in chilled water and on chicken carcasses (9, 11, 14, 23). According to the report of Mead et al. (15), chlorinating the chilled water to a concentration of total residual chlorine less than 30 ppm did not prevent microbial cross-contamination, but it became effective when the concentration was increased to a level greater than 30 ppm. The effectiveness of chlorine is dependent not only on the concentration of chlorine used but also on the chemical composition of chilled water (12, 16), because chlorine reacts not only with microorganisms but also with the inorganic and organic materials present (27).

*D*-values of *Campylobacter* and *Salmonella* on chicken carcasses and in processing water were reported by Humphrey et al. (8) associated with the water pH and Okrend et al. (18) associated with the addition of acetic acid in scalding water. However, there are no *D*-values available to describe the destruction of both bacterial species at different scalding temperatures with different water ages. The effect of chlorination on reducing *Campylobacter* and *Salmonella* in water and on chicken skin was tested by several researchers (1, 13) using the chlorine demand-free water such as tap water or buffer solution. However, no reports have been found for the *D*-values of *Campylobacter* and *Salmonella* on the chicken skins and in chilled water with variable chlorine levels and water ages. Therefore, the objectives of this research were to determine the *D*-values of *Salmonella* Typhimurium and *C. jejuni* in poultry scalding, affected by the scalding temperature and the age of scalding.
water, and in poultry chilling, affected by the chlorine level and the age of chilled water.

**MATERIALS AND METHODS**

**Microorganisms.** A nalidixic acid (N)-resistant mutant of *Salmonella Typhimurium* (ATCC 14028) was obtained from Dr. Amy Waldroup (Department of Poultry Science, University of Arkansas, Fayetteville, Ark.) and maintained on XLT4 (Remel, Lenexa, Kans.) agar slants containing 200 ppm N-sodium salt (Sigma Chemical Co., St. Louis, Mo.) at 4°C. Fresh culture was grown for 18 to 20 h at 37°C in brain heart infusion broth (Difco Laboratories, Detriot, Mich.) containing 200 ppm N-sodium salt. *Campylobacter jejuni* (ATCC 33291) stock culture was maintained in a cryogenic vial at −70°C. Fresh culture was grown using a biphasic system of brucella agar (Remel) overlayed with brain heart infusion broth and cultured at 42°C for 18 to 24 h in a three-gas incubator containing 5% O₂, 10% CO₂, and 85% N₂ (20, 30).

**Processing water and chicken skins.** Tap water from the laboratory was used as 0-h processing water. It was assumed that the solids lost in the overflow water and gained from incoming carcasses reach equilibrium around 5 to 6 h after batch operation started (3, 26). The 8-h chilled water and the 10-h scalding water were collected at the outlet of the overflow from a scaler and a chiller in a commercial poultry processing plant on the test day to represent the processing water that had reached solid equilibrium. Chicken skins, approximately 12 cm² in area, were cut from the breasts of the chicken carcasses obtained from a processing plant on the test day. The skin pieces were held with plastic skin holders to expose an area of 10.75 cm² on the external side of each skin sample for treatment.

**Skin sample inoculation and sampling.** Skin samples were inoculated with two initial levels (×10⁶–7 and ×10³ CFU/cm²) for treatments. For the treatments with the lower inoculum level, the skin samples were irradiated to ~35 kGy with a 10-MeV electron accelerator (Linear Accelerator Facility, Iowa State University, Ames, Iowa) and kept at −20°C until use. For the treatments with the higher inoculum level the skins were used without irradiation.

A skin attachment model has been widely used for evaluating the bactericidal efficacy of potential carcass disinfectants (4, 22, 24). The skin attachment model results in 23 to 44% of inoculated cells attaching on skin. To prepare the skin samples with bacteria, a modified skin attachment model was used. First, 50 µL of bacterial culture was dropped on the exposed area (10.75 cm²) of each skin sample and spread evenly with a sterilized glass rod. The bacterial cultures with 10⁸–9 and 10⁵ CFU/ml were used to obtain the skins with ×10⁶–7 or ×10³ CFU/cm² of bacteria, respectively. Second, all inoculated skin samples were left at room temperature for 30 min to allow the bacterial cells to adhere to the skin surfaces. Then, each skin sample was rinsed with 20 ml of sterilized phosphate-buffered saline (0.05 M, pH 7.4) to wash off loosely attached bacterial cells. After treatments, each skin sample was placed into a sterilized stomaching bag (model 400; Seward Medical Limited, London, UK) with 20 ml of 0.1% peptone water (Remel), and stomached in a stomacher (Stomacher 400; Seward Medical Limited) for 2 min. One milliliter of sample of the stomaching water was collected and decimally diluted to 10⁻² with phosphate-buffered saline for microbial tests.

**Scalding treatment.** The scalding water and the chicken skin samples with *Salmonella Typhimurium* and *C. jejuni* were treated under six combinations of two factors: the scalding temperature (50, 55, and 60°C) and the age of scalding water (0 and 10 h). To determine the bacterial survival in scalding water, three beakers, each containing 100 ml of scalding water, were placed in a water bath (model 25; Precision Scientific, Chicago, Ill.) to maintain the required scalding temperature. One milliliter of bacterial culture (10⁻⁸⁻⁹ CFU/ml) was inoculated into each beaker to obtain an initial population of 10⁻⁶⁻⁷ CFU/ml. One milliliter of water samples from each of these three beakers was taken every minute for a 5-min scalding treatment and decimally diluted to 10⁻⁴ with phosphate-buffered saline for the microbial tests. Bacterial concentration in the scalding water at t = 0 was determined by multiplying the concentration of bacteria in the original culture by the dilution factor (1:100). To determine the bacterial survival on chicken skin, 9 liters of scalding water contained in a stainless steel container was heated to a required scalding temperature and moved into the water bath to maintain the temperature. Fifteen inoculated skin samples were submerged into the scalding water. The temperatures of the scalding water and the chicken skins were monitored by a digital thermometer (model HH23; Omega Engineering Inc., Stamford, Conn.) with T-type thermocouples. Three skin samples were taken every minute within a 5-min treatment. The number of bacteria on the chicken skin at t = 0 was determined using the inoculated skin samples without treatment.

**Chilling treatment.** Chilled water with *Salmonella Typhimurium* and *C. jejuni* was treated under eight combinations of
two factors: the chlorine concentration (0, 10, 30, and 50 ppm) and the age of chilled water (0 and 8 h). Chicken skins with bacteria were treated under combinations of the same factors, except that the chlorine concentration was controlled only at the lowest and highest levels (0 and 50 ppm) because the pretest result showed that 10 and 30 ppm of chlorine had little effect on the bacteria inoculated on the skins. Beakers, each containing 100 ml of chilled water, were placed in an iced-water bath to keep the temperature of the chilled water at 2°C. A hypochlorite stock solution (containing 5.88 mg/ml hypochlorite) from Pristine-M (Ecolab Inc., St. Paul, Minn.) containing 8.4% active ingredient sodium hypochlorite was used to obtain 10, 30, and 50 ppm of initial chlorine levels in the chilled water. The chlorine concentration in the stock solution was calibrated in double deionized water before each test. The residual free chlorine and the total chlorine in chilled water were determined using an Ion Specific Meter (model HI 93711; Hanna Instruments, Woonsoket, R.I.). To determine the bacterial survival in chilled water, 1 ml of bacterial culture (10^8–9 CFU/ml) was inoculated into each of three beakers to get an initial population of 10^6 CFU/ml in water and CFU/cm^2 on skin. The chicken skins were mounted on skin holders and submerged in the scalding water.

### TABLE 1. D-values of C. jejuni and Salmonella Typhimurium in scalding water and on chicken skins during scalding at different temperatures with different water ages

<table>
<thead>
<tr>
<th>Source</th>
<th>Water age (h)</th>
<th>Scalding temperature (°C)</th>
<th>D_\text{main} (min)</th>
<th>D_\text{res} (min)</th>
<th>P_\text{res}</th>
<th>D_\text{main} (min)</th>
<th>D_\text{res} (min)</th>
<th>P_\text{res}</th>
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</thead>
<tbody>
<tr>
<td>Scalding water</td>
<td>0</td>
<td>50</td>
<td>4.0</td>
<td>—*</td>
<td>0</td>
<td>23.6</td>
<td>—*</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>0</td>
<td>55</td>
<td>0.2</td>
<td>13.9</td>
<td>6 × 10^{-6}</td>
<td>1.1</td>
<td>—*</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>0</td>
<td>60</td>
<td>&lt;0.2</td>
<td>—</td>
<td>0</td>
<td>0.2</td>
<td>2.6</td>
<td>4 × 10^{-4}</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>50</td>
<td>13.3</td>
<td>—</td>
<td>0</td>
<td>NC*</td>
<td>—</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>55</td>
<td>0.4</td>
<td>NC*</td>
<td>2 × 10^{-5}</td>
<td>4.1</td>
<td>—</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>60</td>
<td>&lt;0.2</td>
<td>—</td>
<td>0</td>
<td>0.4</td>
<td>9.2</td>
<td>7 × 10^{-6}</td>
</tr>
<tr>
<td>Chicken skin</td>
<td>0</td>
<td>50</td>
<td>2.1</td>
<td>NC*</td>
<td>7 × 10^{-2}</td>
<td>18.9</td>
<td>—*</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>0</td>
<td>55</td>
<td>2.2</td>
<td>19.4</td>
<td>4 × 10^{-2}</td>
<td>5.9</td>
<td>—*</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>0</td>
<td>60</td>
<td>0.5</td>
<td>18.3</td>
<td>7 × 10^{-3}</td>
<td>2.5</td>
<td>—*</td>
<td>0</td>
</tr>
<tr>
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<td>50</td>
<td>1.4</td>
<td>38.0</td>
<td>1 × 10^{-4}</td>
<td>13.1</td>
<td>—*</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>55</td>
<td>1.5</td>
<td>10.2</td>
<td>3 × 10^{-2}</td>
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<td>—*</td>
<td>0</td>
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<tr>
<td></td>
<td>10</td>
<td>60</td>
<td>0.5</td>
<td>NC*</td>
<td>9 × 10^{-3}</td>
<td>1.9</td>
<td>—*</td>
<td>0</td>
</tr>
</tbody>
</table>

* The inoculum level was 10^{6–7} (CFU/ml in water and CFU/cm^2 on skin). The chicken skins were mounted on skin holders and submerged in the scalding water.

* The processing time at which scalding water was obtained.

* D\_\text{main}, the D-values of the main subpopulation; D\_\text{res}, the D-values of the more resistant subpopulation; P\_\text{res}, the proportion of the more resistant subpopulation over the total population.

* No subpopulation.

* No change in cell numbers.

For enumeration of the N-resistant mutant of Salmonella Typhimurium, the samples were plated on tryptic soy agar (Difco Laboratories, Detroit, Mich.) containing 200 ppm N-sodium salt and incubated at 37°C for 48 h (17). The pretest result showed that the injured cells could be recovered on this agar as well as on tryptic soy agar. For enumeration of C. jejuni, the samples were plated on Campy-Cefex agar and incubated at 42°C for 48 h under microaerobic conditions (5% O_2, 10% CO_2, and 85% N_2). Campy-Cefex agar was selected because it contains horse blood and can recover the injured cells effectively (21). A 0.1-ml aliquot of each sample dilution was plated. Additionally, 1 ml of the original sample was divided and spread on four plates; the total colonies on these four plates were counted as CFU/ml when the cell numbers in the diluted samples were too low to be counted.

### Determination of D-values

D-values were determined by the linear regression technique. According to Humpheson’s method (6), if the destruction curve was linear, the bacteria were considered as a single population. The D-value was determined as the reciprocal of the slope of the linear regression line. If the destruction curve was failed, two conjunct linear regression lines with different slopes were obtained. The bacteria were considered as two populations, a main subpopulation and a more resistant subpopulation. The D-values of the main subpopulation and the more resistant subpopulation were determined as the reciprocals of those two regression lines and notated as D\_\text{main} and D\_\text{res}, respectively.

### Statistical analysis

The means and the standard deviations of the cell number of Salmonella Typhimurium and C. jejuni (log CFU/ml or log CFU/cm^2) in the samples were calculated based on the triplicates, and the difference among the means was analyzed using statistical software JMP IN (SAS Institute Inc., Cary, N.C.) via t test (α = 0.05).

### RESULTS AND DISCUSSION

#### Bacterial survival in scalding water

When the scalding temperature was increased from 50 to 60°C, the number of C. jejuni (Fig. 1A) and Salmonella Typhimurium (Fig.
FIGURE 2. The survival of (A) C. jejuni and (B) Salmonella Typhimurium on chicken skins at scalding temperatures of 50 (●), 55 (○), and 60°C (▲). Chicken skins were mounted on skin holders and submerged in 0-h (— —) and 10-h (— — —) scalding water.

FIGURE 3. The temperature profiles of water (— — —) and chicken skin (— — —) during scalding at temperatures of 50 (▲), 55 (○), and 60°C (●).

1B) in the scalding water at the end of a 5-min treatment was reduced by approximately 6 log CFU/ml. C. jejuni was sensitive to the temperature change within the range of 50 to 55°C. At 55°C almost all the cells died, and the reduction was 5 logs more than that at 50°C. The sensitive temperature range for Salmonella Typhimurium was 55 to 60°C, which was 5°C higher than the range for C. jejuni. The reduction of Salmonella Typhimurium after a 5-min treatment increased by 4 log CFU/ml when the scalding temperature increased from 55 to 60°C.

The survival data for C. jejuni in the 0-h scalding water at 50°C exhibited a linear curve (Fig. 1A). No subpopulation existed at this temperature. When the temperature was increased to 55°C, a more resistant subpopulation was apparent in all of the triplicate samples. A tailed survival curve (Fig. 1A) was plotted based on the means of the bacterial survivors in triplicate samples. The proportion of the more resistant subpopulation was 6.3 × 10^(-6) (Table 1), i.e., out of 10^6 cells, 6 cells were more resistant. When the temperature increased to 60°C, the whole population was killed in 1 min (Fig. 1A). Similarly, in 10-h scalding water, C. jejuni was reduced at a single linear rate at 50°C, but had a more resistant subpopulation at 55°C, and became nondetectable in 1 min at 60°C (Fig. 1A). The destruction curves for Salmonella Typhimurium showed basically linear declines at 50 and 55°C (Fig. 1B). The more resistant subpopulation showed up only when the temperature increased to 60°C (Fig. 1B). The D_{main}, D_{res}, and the proportion of the more resistant subpopulation (P_{res}) of Salmonella Typhimurium and C. jejuni in water during scalding are listed in Table 1.

Bacterial survival on chicken skins during scalding.

The temperature effect on bacterial survival was reduced after bacteria attached to the chicken skins. When scalding temperature increased from 50 to 60°C, the bacterial reduction on chicken skins increased by only approximately 2 log CFU/cm² (Fig. 2). One possible reason was that the skin temperature could never reach the level of the water temperature. As shown in Figure 3, after the skins (at room temperature) were immersed into the scalding water, the skin temperature rose rapidly in the first 20 s, reaching a temperature approximately 10°C lower than the temperature of the water. Then skin temperature increased slowly and reached an equilibrium between the water and the skin after 3 min. Thereafter, the skin temperature remained at a temperature of 3°C lower than the water temperature.

Skin test results also showed that C. jejuni was more heat sensitive than Salmonella Typhimurium. At 50°C the numbers of C. jejuni on chicken skins decreased significantly (Fig. 2A), but the number of Salmonella Typhimurium did not (Fig. 2B). Destruction of Salmonella Typhimurium was observed only at a scalding temperature of 55°C or higher (Fig. 2B). An interesting phenomenon was that at all temperature levels, C. jejuni displayed tailed curves that declined rapidly during the first 1 to 3 min and then remained unchanged during the rest of the treatment time (Fig. 2A). Differently, Salmonella Typhimurium was decreasing linearly throughout the 5-min treatments (Fig. 2B). The D_{main}, D_{res}, and P_{res} of Salmonella Typhimurium and C. jejuni on chicken skins during scalding are listed in Table 1.

The tests with the skins inoculated with the bacteria at the two initial levels of ~10^3 and 10^6–7 CFU/cm² were conducted to examine the effect of initial cell number on
A bacterial survival in chilled water. Measured chlorine levels (both free and total chlorine) of the chilled water used in these tests (either the tap water from the laboratory or the 8-h chilled water from the processing plant) were less than 3 ppm. Without chlorine addition C. jejuni and Salmonella Typhimurium did not change significantly \( (P > 0.05) \) in the chilled water during the treatment (Fig. 4). Both bacterial species decreased linearly in the 0-h chilled water with all the tested chlorine levels (Fig. 4). With 10, 30, and 50 ppm of chlorine the \( D \)-values were 17.2, 1.3, and 0.5 min for \( C. \) jejuni and 67.1, 3.4, and 0.9 min for Salmonella Typhimurium (Table 2), respectively. However, in the 8-h chilled water the chlorine effect was diminished. With 10 ppm chlorine, \( D \)-values of both bacterial species were >113 min (Table 2). With 30 and 50 ppm chlorine \( C. \) jejuni and Salmonella Typhimurium both exhibited the more resistant subpopulations persisting in the treatment (Fig. 4). The \( D_{\text{main}}, D_{\text{res}}, \) and \( P_{\text{res}} \) of Salmonella Typhimurium and \( C. \) jejuni in the chilled water are listed in Table 2.

Free chlorine is the key component of the hypochlorite providing bactericidal activity. Residual free chlorine in chilled water depends on the amount of chlorine added, the organic components in the chilled water, as well as the contact time \( (27) \). The changes in the residual free chlorine of the chilled water in relation to the treatment time (contact time) are shown in Figure 5. The residual free chlorine at \( t = 0 \) was the initial chlorine level measured in the double deionized water with the addition of chlorine at a required level. When 10, 30, or 50 ppm of chlorine was added into the 0-h chilled water, the residual free chlorine dropped to approximately 6, 17, or 34 ppm in 1 min and then contin-

### Table 2. \( D \)-values of \( C. \) jejuni and Salmonella Typhimurium in chilled water and on chicken skins during chilling at different chlorine levels with different water ages

<table>
<thead>
<tr>
<th>Source</th>
<th>Water age ( b ) (h)</th>
<th>Chlorine concentration (ppm)</th>
<th>( D_{\text{main}} ) ( c ) (min)</th>
<th>( D_{\text{res}} ) ( c ) (min)</th>
<th>( P_{\text{res}} )</th>
<th>( D_{\text{main}} ) ( c ) (min)</th>
<th>( D_{\text{res}} ) ( c ) (min)</th>
<th>( P_{\text{res}} )</th>
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<tbody>
<tr>
<td>Chilling water</td>
<td>0</td>
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<td>—  ( d )</td>
<td>0</td>
<td>67.1</td>
<td>—  ( d )</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>0</td>
<td>30</td>
<td>1.3</td>
<td>—  ( d )</td>
<td>0</td>
<td>3.4</td>
<td>—  ( d )</td>
<td>0</td>
</tr>
<tr>
<td></td>
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<td>0.5</td>
<td>—  ( d )</td>
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<td>—  ( d )</td>
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<td>—  ( d )</td>
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<td></td>
<td>8</td>
<td>50</td>
<td>6.0</td>
<td>103.0</td>
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<td>41.2</td>
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<td>73.0</td>
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<td></td>
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<td>—  ( d )</td>
<td>0</td>
<td>167.7</td>
<td>—  ( d )</td>
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</table>

\( a \) The inoculum level was \( 10^{6-7} \) (CFU/ml in water and CFU/cm\(^2\) on skin). The chicken skins were mounted on skin holders and submerged in the chilling water.

\( b \) The processing time at which the chilling water was obtained.

\( c \) \( D_{\text{main}} \), the \( D \)-values of the main subpopulation; \( D_{\text{res}} \), the \( D \)-values of the more resistant subpopulation; \( P_{\text{res}} \), the proportion of the more resistant subpopulation over the total population.

\( d \) No subpopulation.
ued decreasing to 3, 10, or 20 ppm in 50 min (Fig. 5). In 8-h chilled water whichever of 10, 30, or 50 ppm chlorine was added, the residual free chlorine dropped to ~0 ppm in only 1 min (Fig. 5). These results were consistent with the report of Tsai et al. (27). The consumption of free chlorine by the organic compounds in the chilled water reduced the free chlorine available for microorganisms, consequently reducing the effect of the chlorine treatments.

In the 0-h chilled water with 30 ppm of chlorine, it took 10 min to reduce C. jejuni by 6 log CFU/ml but 20 min to reach the same reduction of Salmonella Typhimurium (Fig. 4). In the 8-h chilled water with 50 ppm of chlorine, Salmonella Typhimurium showed a reduction of approximately 3 log CFU/ml, while C. jejuni showed a reduction of 5 log CFU/ml after 50 min treatment (Fig. 4).

Bacterial survival on chicken skins during chilling. An addition of 50 ppm of chlorine led to 20 to 30 ppm of residual free chlorine present in the 0-h chilled water during a 50-min treatment (Fig. 5), but the reduction of Salmonella Typhimurium on the chicken skin was less than 1 log CFU/ml (Fig. 6) with a D-value of 78.7 min (Table 2). The reduction of C. jejuni on the chicken skin in the 8-h chilled water was even less (Fig. 6) with a D-value of 167.7 min (Table 2). The possible reasons are that the oily layer on the chicken skin surface prevented chlorine from reaching the bacteria hidden in the cervixes and the follicles, and also the concentration of the free chlorine around the skin was reduced due to the consumption of the chlorine by the organic materials on the skin surface.

Tests with the skins inoculated with bacteria at the two initial levels of ~10^3 and 10^6–7 CFU/cm^2 were conducted to examine the effect of initial cell number on bacterial survival. The survival curves of C. jejuni and Salmonella Typhimurium with these two initial levels were basically in parallel (data not shown). This indicated that the initial bacterial cell number did not affect the reduction of bacteria on the skin during chilling.

**CONCLUSIONS**

Under the experimental conditions defined in this study, raising the scalding temperature and the chlorine concentration of the chilled water effectively blocked cross-contamination through water but had little effect on the bacteria attached on skins. In scalding water, C. jejuni was sensitive to the temperature change from 50 to 55°C, whereas Salmonella Typhimurium was sensitive to the change from 55 to 60°C. The age of the scalding water had no effect on the bacterial heat sensitivity during scalding. However, the increase in the age of chilled water significantly reduced the bactericidal activity of the chlorine.

$D_{\text{main}}$ together with $D_{\text{res}}$ was able to describe the bacterial destruction with tailed curves. However, not all the curves could be fit to two linear lines due to their complexities. A more reliable and accurate method is needed for the prediction of the bacterial survivors. As the next step, predictive microbiology models need to be developed based on the data collected in this study for the prediction of the survival or death of C. jejuni and Salmonella Typhimurium during poultry scalding and chilling, associated with the scalding temperature, the chlorine level in chilled water, and the water age.
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