

# **Salmonella Transfer Potential during Hand Harvesting of Tomatoes under Laboratory Conditions**

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MS 13-048: Received 4 February 2013/Accepted 12 April 2013

## **ABSTRACT**

Tomato good agricultural practices, mandatory guidelines in Florida, do not have specific regulations for glove use during tomato harvesting. The objective of the research reported here was to evaluate *Salmonella* transfer from contaminated gloves to tomatoes and vice versa upon single and subsequent touches. Experiments were performed using mature, green, round tomatoes with two types of gloves (reusable and single use) and two hygienic conditions of reusable glove (clean and dirty [fouled with tomato leaves]). The transfer scenarios used during experiments were glove to tomato, tomato to glove, and glove to up to 25 subsequently touched tomatoes. The inoculated surface (6 log CFU per surface), after drying for 24 h, touched the uninoculated surface for 5 s. *Salmonella* populations from gloves and tomatoes were enumerated on nonselective and selective agar supplemented with 80 µg/ml rifampin. Enrichments were performed when counts fell below the detection limit. The rates of *Salmonella* transfer to tomatoes during a single touch were similar for single-use and reusable gloves; transfer from tomatoes to gloves was higher to single-use gloves than to reusable gloves under wet (0 h) inoculation conditions. Dirty reusable gloves did not transfer more *Salmonella* than clean reusable gloves during single contact under any conditions. When a single glove was sequentially touched to multiple tomatoes, clean reusable gloves transferred higher levels of *Salmonella* to the first few tomatoes touched than did single-use gloves and dirty reusable gloves. As workers' gloves became dirty over time during harvest, the risk of *Salmonella* transfer to tomatoes did not increase.

Florida tomatoes are normally hand harvested at the mature green stage of development (19) by workers typically wearing reusable gloves. Even at the mature green stage, tomatoes are sensitive to mechanical damage, and thus, the harvest crew must be trained and supervised to ensure gentle handling while transferring tomatoes into the bins and gondolas (19). Upon arrival at the packinghouse, tomatoes are dumped into dump tanks with sanitizers (typically chlorine), moved by conveyor belts, and manually graded by workers wearing single-use gloves. Sized and graded tomatoes are packed into fiberboard cartons for distribution (19).

The Florida tomato industry implemented mandatory state inspection and auditing of tomato production, handling, and packing to verify adherence to Florida tomato good agricultural practices (T-GAPs) (production program) and tomato best management practices (packing program) in 2008 (2). T-GAPs and tomato best management practices are mandatory practices in Florida to ensure the safety of fresh tomatoes produced, packed, repacked, distributed, and sold in or from Florida (2). Currently, there is no guidance in T-GAPs and tomato best management practices for glove use during harvesting or packing.

Tomatoes have been implicated in U.S. *Salmonella* outbreaks (4, 5, 7, 10, 11), with the majority of tomatoes traced to production in the U.S. states of Virginia and Florida. The *Salmonella enterica* serovars involved in these outbreaks include Anatum, Baildon, Braenderup, Javiana, Newport, and Typhimurium (4, 5). Traceback investigations rarely identify the source of the pathogen, although preharvest conditions are often targeted as a probable source of *Salmonella* contamination. Suspected routes of preharvest *Salmonella* contamination in tomatoes include water, soil, manure, insects, and human sources (3). Subsequent cross-contamination during harvesting and packing may result in an increased number of contaminated tomatoes.

Cross-contamination, the transfer of pathogens from one surface to another, has been identified as a risk factor in a number of food processing and handling environments. The movement of pathogens between foods and surfaces is dependent upon the type of bacteria (14), type of surface (6), moisture level (9), and inoculation dose (15). Despite it being a critical point in understanding pathogen movement and contamination, studies on cross-contamination during hand harvesting of produce remain relatively scarce.

An understanding of the potential movement of *Salmonella* from workers' gloved hands to and from tomatoes during harvest and packing remains a critical data

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gap for tomato food safety. An evaluation of how cross-contamination risks may change over the course of a harvesting shift as workers' gloves become increasingly fouled has not been conducted. In this study, the *Salmonella* transfer risk associated with clean (single-use and reusable) and dirty reusable gloves, considered a potential route of cross-contamination during tomato harvest and packing, are quantified. The data generated in this study are essential to generate model scenarios of produce contamination during harvesting with gloved hands and to devise appropriate intervention to manage risk.

## MATERIALS AND METHODS

**Tomatoes.** Round, mature, green tomatoes were purchased from local sources and stored at 4°C for up to 5 days before use. After washing with tap water, tomatoes were stored overnight at room temperature (ca. 22°C) prior to inoculation. A circle (2 to 3 cm in diameter) was drawn on the surface of tomatoes using a permanent marker on the day of inoculation.

**Gloves.** Clean gloves, reusable (latex gloves, Publix, Lakeland, FL) and single use (powder-free latex exam gloves, Fisher Scientific, Fair Lawn, NJ), were cut into square pieces (5 by 5 cm) with sterile scissors.

**Dirty reusable gloves.** In order to consistently foul reusable gloves in the laboratory in a manner similar to that in which gloves are naturally fouled from tomato harvesting, various quantities of local soil (0.1, 0.3, and 0.5 g), internal tomato tissue (3.75 g, 1.875 g, and a drop), and a tomato leaf were sequentially rubbed on a glove piece for up to 30 s in 5-s intervals. Glove pieces made dirty using the various combinations of fouling materials were inoculated immediately after fouling or after 30 min of drying. *Salmonella* transfer between the gloves that had been fouled in various ways and tomatoes was evaluated. A standardized protocol of a single tomato leaf with 20 s of rubbing was used as the standard fouling technique.

***Salmonella* strains.** A five-strain cocktail of *Salmonella* was used in all experiments. These strains and their sources included *Salmonella* Michigan (strain LJH 615, cantaloupe outbreak), *Salmonella* Montevideo (strain LJH 614, tomato outbreak, human isolate), *Salmonella* Newport (isolate from tomato outbreak, environmental), *Salmonella* Poona (strain LJH 631, cantaloupe outbreak, human isolate), and *Salmonella* Saintpaul (strain BAC# 158, orange juice). Strains were adapted to grow in the presence of 80 µg/ml rifampin (Fisher Scientific) through the use of a stepwise exposure to increasing (1:2) concentrations of antibiotic (16).

**Inoculum.** *Salmonella* cultures stored at -80°C in cryogenic vials were thawed and then were streaked onto tryptic soy agar (TSA; nonselective, Difco, BD, Sparks, MD) plates supplemented with rifampin (80 µg/ml) (TSAR) followed by overnight incubation (24 ± 2 h) at 37 ± 2°C. One isolated colony was transferred to 10 ml of tryptic soy broth (Difco, BD) supplemented with rifampin (80 µg/ml) and incubated for 24 h at 37 ± 2°C. Following this incubation, a 10-µl loopful was transferred into 10 ml of fresh tryptic soy broth with rifampin and incubated for 24 h at 37 ± 2°C. Cultures were collected by centrifugation (Allegra X-12, Beckman Coulter, Fullerton, CA) at 3,000 × g for 10 min. The cells were washed twice by removing the supernatant and suspending in 10 ml of Butterfield's phosphate buffer (BPB;

Hardy Diagnostics, CA). Washed cells were suspended in half the initial volume (5 ml) of BPB, resulting in ca.  $1.0 \times 10^9$  CFU/ml bacterial populations. Serial dilutions were performed with 0.1% peptone water (Difco, BD) to achieve populations of ca.  $10^7$  CFU/ml. Equal volumes of each *Salmonella* strain were combined into the cocktail. The *Salmonella* cocktail was transferred to 1-ml-size micropipettes and microcentrifuged (MiniSpin, Eppendorf, Hauppauge, NY) at  $13,400 \times g$  for 10 min. The supernatant was discarded, 1 ml of 0.1% peptone water added as a carrier medium, and the mixture stored on ice for up to 1 h prior to inoculating gloves or tomatoes.

The cocktail (100 µl) of *Salmonella* cells was distributed in 6 to 8 drops on the glove pieces or circled area of tomatoes set on lead rings to prevent rolling (12). The inoculum was dried for 0, 1, or 24 h in a biosafety cabinet. All transfer experiments were conducted on the laboratory bench at ambient temperature (22 ± 3°C) and humidity (37% ± 10%).

**Transfer from clean (single-use or reusable) or dirty (reusable) gloves to tomatoes.** Transfer from gloves was evaluated after the drying times of 0 (wet inoculum), 1, and 24 h. The 24-h drying time was not used for dirty gloves, as gloves would be washed at the end of each harvest day to remove fouling materials; it is an unlikely situation that a *Salmonella* inoculum would be able to dry for 24 h on a dirty glove prior to reuse. Uninoculated tomatoes were touched with the inoculated glove surfaces by applying light pressure (no impression on the tomato surface) for less than 5 s and placed into a sterile sampling bag (17.78 by 30.48 cm; Fisher Scientific, Pittsburg, PA). Each replication consisted of three inoculated-glove samples for each drying time and three samples for control. Three additional glove samples were placed in each group and sampled with a protocol similar to that used for tomatoes to determine the population of *Salmonella* on the inoculated surface. A total of six replications were performed ( $n = 18$ ).

**Transfer from tomatoes to clean (single-use or reusable) gloves.** Inoculated tomatoes with drying times of 0, 1, and 24 h were touched with glove pieces (glove pieces on top of tomatoes) by applying light pressure (no impression on the tomato surface) for less than 5 s and then placed in sterile sampling bags as described above. The sampling methods and number of replications were also as described above in the glove-to-tomato transfer scenario.

**Transfer from clean (single-use or reusable) or dirty (reusable) gloves to multiple sequential tomatoes.** Inoculated gloves with drying times of 0 and 1 h were touched sequentially with 25 (clean gloves) or 10 (dirty gloves) tomatoes one after the other and placed in sampling bags as described above. The experiments were replicated three times with three samples at one time point ( $n = 9$ ).

***Salmonella* enumeration.** Twenty milliliters of BPB was added to sampling bags containing the originally inoculated or transfer surface. For all dirty-glove samples, 0.1% Tween 20 (Fisher Scientific, Fairlawn, NJ) was added to the 20 ml of BPB to help in removal of cells from fouled surfaces (17). Tomato samples were subjected to a gentle rub-shake-rub for 1 min, and glove samples were macerated (Smasher blender, AES Chemunex, Cranbury, NJ) for 1 min prior to enumeration. Serial dilutions were prepared in 0.1% peptone water (9 ml), and 0.1 ml was surface plated onto TSAR and bismuth sulfite agar (BSA; selective, Difco, BD) supplemented with rifampin (80 µg/ml)

(BSAR) in duplicate. To improve the limit of detection, 0.25 ml of the liquid in the sampling bags (i.e., the lowest dilution) was surface plated onto both TSAR and BSAR in quadruplicate. The colonies present after incubation at  $37 \pm 2^\circ\text{C}$  for  $24 \pm 2$  h (TSAR) or  $48 \pm 2$  h (BSAR) were counted by hand.

When counts fell below the limit of detection (1.3 log CFU per surface), enrichment for *Salmonella* was conducted by the U.S. Food and Drug Administration *Bacteriological Analytical Manual* protocol for produce (10, 21). Briefly, 20 ml of double-strength lactose (Difco, BD) was added to the sterile sampling bags and incubated at  $37 \pm 2^\circ\text{C}$  for 24 h. One hundred microliters and 1 ml of the mixture were then transferred to 9.9-ml tubes of Rappaport-Vassiliadis R10 (Difco, BD) and tetrathionate (Difco, BD) broths, respectively. The test tubes were incubated for 48 h at  $42 \pm 2^\circ\text{C}$  for Rappaport-Vassiliadis broth and 24 h at  $37 \pm 2^\circ\text{C}$  for tetrathionate broth. Ten-microliter loopfuls were streaked onto BSA, xylose lysine deoxycholate agar (Difco, BD), and Hektoen enteric agar (Difco, BD) and incubated at  $37 \pm 2^\circ\text{C}$  for 24 h. Presumptive *Salmonella*-positive colonies were confirmed by using triple sugar iron agar (Difco, BD) and lysine iron agar (Difco, BD) slants.

**Transfer coefficients.** Transfer coefficients (TCs) were calculated with the equation  $\text{TC} = P_t/P_i$ , where  $P_t$  is the *Salmonella* population on a touched surface, which can be tomato or glove (CFU per surface), and  $P_i$  is the *Salmonella* population on an inoculated surface (CFU per surface).

**Statistics.** The averages of the results, including *Salmonella* populations and TCs, were analyzed using Statistical Analysis System (SAS 9.2, SAS Institute Inc., Cary, NC) for analysis of variance, and significance was determined by using the least-square significance test at a  $P$  value of  $<0.05$ . The results obtained following *Salmonella* enrichment were analyzed using Fisher's exact method in SAS.

## RESULTS

**Dirty reusable gloves.** The levels of *Salmonella* transfer to tomatoes touched with inoculated dirty gloves and dirty gloves touched with inoculated tomatoes immediately after inoculation were not significantly different based on fouling type ( $P \geq 0.05$ ; data not shown). After 1 h of inoculum drying, the populations of *Salmonella* recovered from tomatoes touched with inoculated dirty gloves were below the detection limit in all cases ( $P \geq 0.05$ ; data not shown). As the composition of the fouling material and drying did not significantly alter transfer in all subsequent experiments, gloves were made dirty by rubbing a tomato leaf on the glove piece. The use of Tween 20 significantly improved the recovery of *Salmonella* from dirty glove surfaces after 1 h of drying ( $P \leq 0.05$ ; data not shown) but did not significantly influence *Salmonella* recovery either from tomato surfaces touched with dirty gloves after 1 h or from clean gloves ( $P \geq 0.05$ ; data not shown). Tween 20 was added to the recovery buffer for all experiments with dirty gloves.

***Salmonella* transfer between tomatoes and gloves during a single touch.** The results of the statistical analyses performed on the TCs obtained for TSAR and BSAR at different inoculum drying times were not significantly different ( $P \geq 0.05$ ); all tables and figures were created from

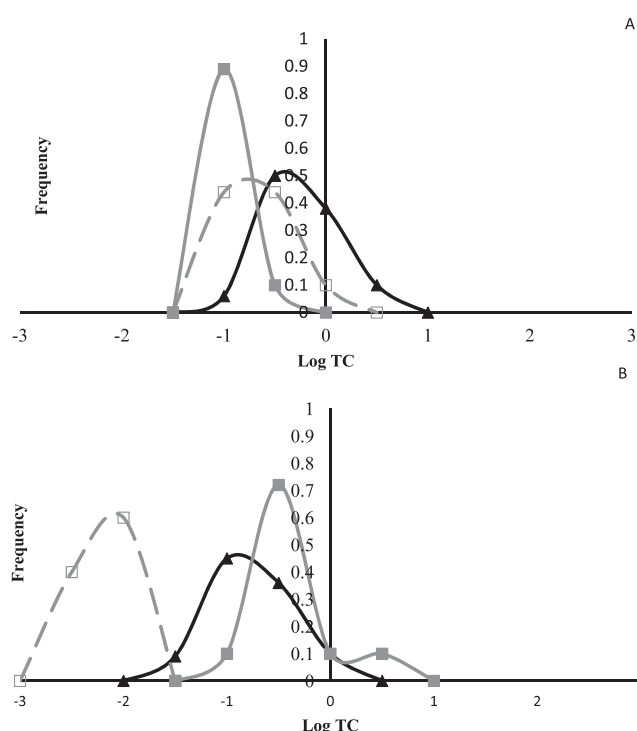


FIGURE 1. Distribution of coefficients of *Salmonella* transfer from gloves to tomatoes (A, B) with wet (A) or dry (1 h) (B) inoculum for clean single-use gloves (black line), clean reusable gloves (grey line), and dirty reusable gloves (dashed grey line) ( $n = 9$  to 18).

data generated on TSAR. The distributions of log TCs are shown in Figures 1 and 2.

***Salmonella* transfer from gloves to tomatoes.** The *Salmonella* transfer to tomatoes from gloves inoculated immediately previously (0 h of drying) did not differ significantly between single-use, clean reusable, and dirty reusable gloves ( $P \geq 0.05$ ) (Fig. 1A, black line [single-use], grey line [clean reusable], and dashed grey line [dirty reusable]). The TCs obtained from inoculated single-use gloves, clean reusable gloves, and dirty reusable gloves when the inoculum was wet (0 h of drying) were  $0.32 \pm 0.1$ ,  $0.25 \pm 0.1$  and  $0.41 \pm 0.3$ , respectively. After drying of the inoculum on glove pieces for 1 h, the rates of *Salmonella* transfer did not differ significantly between single-use and clean reusable gloves ( $P \geq 0.05$ ) (Fig. 1B, black line [single use] and grey line [clean reusable]). The TCs or positive enrichments obtained after 1 h of inoculum drying for inoculated single-use gloves, clean reusable gloves, and dirty reusable gloves were  $0.29 \pm 0.2$ ,  $0.48 \pm 0.5$ , and 5 of 9 tomatoes *Salmonella* positive upon enrichment, respectively. Dirty reusable gloves transferred significantly less *Salmonella* to tomatoes than single-use and clean reusable gloves after 1 h of inoculum drying ( $P \leq 0.05$ ); no significant differences in the percentages of positive gloves were seen between any of the glove types (Fisher's exact method) (Fig. 1B, black line [single use], grey line [clean reusable], and dashed grey line [dirty reusable]).

After 1 h of inoculum drying, the rates of *Salmonella* transfer to tomatoes from single-use gloves decreased

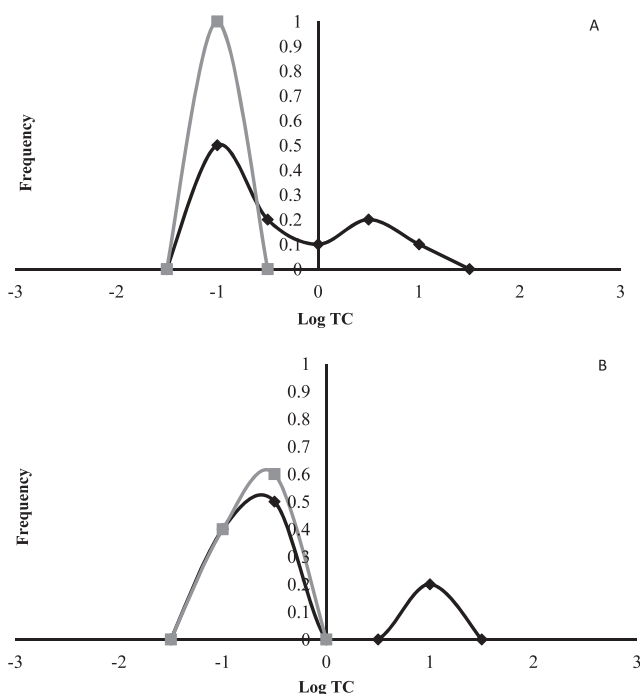


FIGURE 2. Distribution of coefficients of *Salmonella* transfer from tomatoes to gloves (A, B) with wet (A) or dry (1 h) (B) inoculum for clean single-use gloves (black line) and clean reusable gloves (grey line) ( $n = 9$  to 18).

significantly (Fig. 1B, black distribution shifts toward the left, indicating less transfer) and the rates of transfer for clean reusable gloves increased significantly (Fig. 1B, grey distribution shifts toward the right, indicating more transfer) compared with the rates of transfer with 0 h of drying (wet) ( $P \leq 0.05$ ) (Fig. 1A, black lines and grey lines). Dirty gloves transferred significantly less *Salmonella* (Fig. 1B, dashed grey distribution shifts toward the left) upon 1 h of drying ( $P \leq 0.05$ ) (Fig. 1B, dashed grey line, distribution shift toward the left) than was transferred with 0 h of drying (wet) ( $P \leq 0.05$ ) (Fig. 1A, dashed grey line).

No counts were obtained from dirty gloves after 1 h of drying, and so enrichments were performed to confirm the presence or absence of *Salmonella*; for this reason, in addition to the reasons described in “Materials and Methods” related to the unlikelihood of a dirty glove being used following 24 h without being washed, no experiments were performed following 24 h of drying on dirty gloves. When the inoculum was dried for 24 h on single-use and clean reusable gloves, the *Salmonella* populations transferred to tomatoes fell below the limit of detection but were detectable upon enrichment of tomatoes for 9 of 9 replications with single-use gloves and 6 of 9 replications with clean reusable gloves (data not shown).

***Salmonella* transfer from tomatoes to gloves.** The rates of *Salmonella* transfer from inoculated tomatoes to single-use gloves were significantly higher than the rates of transfer from inoculated tomatoes to clean reusable gloves when the inoculum was wet (0 h of drying) ( $P \leq 0.05$ ) (Fig. 2A, black line [single-use] and grey line [clean reusable]); the TCs were  $0.37 \pm 0.2$  for single-use gloves

and  $0.18 \pm 0.0$  for clean reusable gloves. However, after 1 h of drying, no significant differences were observed between the rates of *Salmonella* transfer to single-use gloves and clean reusable gloves ( $P \geq 0.05$ ) (Fig. 2B, black line [single-use] and grey line [clean reusable]); the TCs were  $0.39 \pm 0.2$  for single-use gloves and  $0.38 \pm 0.2$  for clean reusable gloves.

The rates of *Salmonella* transfer to single-use gloves from tomatoes at 0 and 1 h of drying were not significantly different ( $P \geq 0.05$ ) (Fig. 2A and B, black lines). Similarly, there were no significant differences in the rates of *Salmonella* transfer from tomatoes to clean reusable gloves after 0 or 1 h of drying ( $P \geq 0.05$ ) (Fig. 2A and B, grey lines). Drying the inoculum on the tomato surface for 24 h significantly reduced the populations transferred to single-use glove pieces compared with the rates of transfer for wet inoculum (0 h drying) and those dried for 1 h; no *Salmonella* populations were recovered from the single-use gloves after 24 h of inoculum drying on tomatoes ( $P \leq 0.05$ ). When transferred from tomatoes to clean reusable gloves after 24 h of drying, *Salmonella* populations were observed in 7 of 9 replications (data not shown).

***Salmonella* transfer from gloves to multiple tomatoes.** When tomatoes were subsequently touched with the same inoculated single-use, clean, or dirty reusable gloves, *Salmonella* transferred to up to 25, 23, and 10 tomatoes, respectively. Statistically, no significant differences were observed in the rates of *Salmonella* transfer under wet (0 h) and dry (1 h) inoculum conditions for single-use, clean reusable, or dirty reusable gloves ( $P \geq 0.05$ ) (Tables 1 and 2). The enrichment results obtained from the 10th tomato to the 25th tomato touched with single-use gloves and clean reusable gloves were not significantly different from each other ( $P \geq 0.05$ ) (Table 1).

For single-use gloves, the TCs were significantly less for the fourth tomato touched than for the first tomato ( $P \leq 0.05$ ) (Table 1). For clean reusable gloves, the TCs were significantly less for the second tomato touched than for the first tomato ( $P \leq 0.05$ ) (Table 1). The rates of *Salmonella* transfer to the first tomatoes touched with clean reusable gloves were significantly higher than the rates of transfer to all the tomatoes touched subsequently with inoculated single-use gloves ( $P \leq 0.05$ ) (Table 1). The single-use gloves transferred less *Salmonella* to more tomatoes touched, and the clean reusable gloves transferred more *Salmonella* to fewer tomatoes. After drying of the inoculum for 1 h, the first three tomatoes touched with single-use gloves differed significantly from each other in the rates of *Salmonella* transfer ( $P \leq 0.05$ ) (Table 2). For clean reusable gloves, significant differences were observed from the first tomato to the fifth tomato after 1 h of drying ( $P \leq 0.05$ ) (Table 2).

There were no significant differences in the rates of *Salmonella* transfer to the 10 tomatoes touched with dirty reusable gloves ( $P \geq 0.05$ ) (Table 1). The rates of *Salmonella* transfer to the first tomatoes touched with clean reusable gloves were significantly higher than the rates of transfer to any of the tomatoes touched with dirty reusable



TABLE 1. *Salmonella* transfer from inoculated gloves (single-use, clean reusable, and dirty reusable) with wet inoculum to subsequently touched tomatoes

Tomato no.	Single use			Clean reusable			Dirty reusable		
	Population <sup>a</sup>	TC <sup>b</sup>		Population	TC		Population	TC	
		Avg	Range		Avg	Range		Avg	Range
Glove	5.8 ± 0.5	—	—	5.7 ± 0.4	—	—	5.7 ± 0.2	—	—
T-1	5.1 ± 0.1	0.33 ± 0.4 A <sup>c</sup>	0.04–1.2	5.3 ± 0.2	0.61 ± 0.6 A	0.11–1.8	4.9 ± 0.5	0.22 ± 0.2 A	0.01–0.69
T-2	4.8 ± 0.4	0.18 ± 0.2 A	0.01–0.17	4.6 ± 1.2	0.46 ± 0.6 B	0.00–1.8	4.8 ± 0.4	0.16 ± 0.1 A	0.10–0.39
T-3	4.6 ± 0.4	0.13 ± 0.1 A	0.01–0.35	4.3 ± 1.2	0.27 ± 0.3 B	0.00–5.1	4.5 ± 0.5	0.09 ± 0.1 A	0.02–0.29
T-4	4.3 ± 0.5	0.05 ± 0.0 B	0.00–0.09	4.0 ± 1.2	0.26 ± 0.6 B	0.00–1.8	4.5 ± 0.2	0.07 ± 0.0 A	0.03–0.15
T-5	4.0 ± 1.1	0.05 ± 0.1 C	0.00–0.09	3.5 ± 1.2	0.06 ± 0.1 C	0.00–0.34	4.0 ± 0.5	0.03 ± 0.0 A	0.00–0.05
T-6	3.8 ± 0.8	0.02 ± 0.0 D	0.00–0.03	3.1 ± 1.4	0.03 ± 0.1 C	0.00–0.06	4.1 ± 0.3	0.03 ± 0.0 A	0.00–0.06
T-7	3.5 ± 1.0	0.01 ± 0.0 D	0.00–0.02	3.1 ± 1.1	0.02 ± 0.0 C	0.00–0.02	3.4 ± 1.1	0.02 ± 0.0 A	0.00–0.10
T-8	3.0 ± 1.4	0.01 ± 0.0 D	0.00–0.04	2.5 ± 1.5	0.02 ± 0.0 C	0.00–0.08	3.7 ± 0.3	0.02 ± 0.0 A	0.00–0.05
T-9	3.0 ± 1.0	0.01 ± 0.0 D	0.00–0.03	2.6 ± 1.2	0.01 ± 0.0 C	0.00–0.08	<3.1 ± 0.9 <sup>d</sup>	8/9 <sup>e</sup>	
T-10	2.5 ± 1.0	0.01 ± 0.0 D	0.00–0.06	<2.1 ± 0.9 <sup>f</sup>	6/9		<2.7 ± 1.1 <sup>d</sup>	8/9	
T-11	5/9	5/9		<1.8 ± 1.1 <sup>f</sup>	7/9				
T-12	<1.4 ± 0.8 <sup>d</sup>	8/9		<1.6 ± 0.5 <sup>f</sup>	7/9				
T-13	<1.8 ± 0.8 <sup>d</sup>	7/9		<1.9 ± 0.9 <sup>f</sup>	7/9				
T-14	<1.7 ± 0.8 <sup>d</sup>	7/9		<1.8 ± 0.7 <sup>f</sup>	7/9				
T-15	<1.7 ± 0.8 <sup>d</sup>	6/9		<2.1 ± 0.8 <sup>f</sup>	7/9				
T-16	4/9	4/9		5/9	5/9				
T-17	2/9	2/9		4/9	4/9				
T-18	2/9	2/9		3/9	3/9				
T-19	4/9	4/9		5/9	5/9				
T-20	2/9	2/9		4/9	4/9				
T-21	2/9	2/9		2/9	2/9				
T-22	3/9	3/9		2/9	2/9				
T-23	3/9	3/9		2/9	2/9				
T-24	1/9	1/9		0/9	0/9				
T-25	1/9	1/9		0/9	0/9				

<sup>a</sup> Average *Salmonella* population (log CFU per tomato or glove;  $n = 9$ ) ± standard deviation, or number of positive enrichments.

<sup>b</sup> TC, transfer coefficient, average ± standard deviation and range, or number of positive enrichments.

<sup>c</sup> Letters represent significant differences within columns only; same letters represent no significant differences ( $P \geq 0.05$ ).

<sup>d</sup> Replications were occasionally below the limit of detection.

<sup>e</sup> Number of positive enrichments of nine total enrichments.

<sup>f</sup> Replications were occasionally negative upon enrichment.

gloves ( $P \leq 0.05$ ) (Table 1). Clean reusable gloves transferred more *Salmonella* to the first tomatoes touched, while the rates of *Salmonella* transfer to the second to the eighth tomatoes touched with clean reusable gloves and all 10 tomatoes touched with dirty reusable gloves were not statistically different ( $P \geq 0.05$ ) (Table 1). Drying the inoculum on the dirty glove surface for 1 h reduced the population transferred to below the limit of detection from the 1st tomato to the 10th tomato, and no significant differences were observed between all 10 tomatoes ( $P \geq 0.05$ ) (Table 2).

## DISCUSSION

Cross-contamination between workers' gloved hands and tomatoes is a potential route for *Salmonella* contamination of tomatoes and may be influenced by the fouling of workers' gloves over the course of the day. *Salmonella* can transfer from gloves (single use and reusable) to tomatoes and from tomatoes to gloves under both wet (0 h of drying) and dry (1 h of drying) conditions. However, for single-use

gloves, *Salmonella* transfer was not detected after the inoculum had dried for 24 h. Single-use gloves are made of thin (4 to 8 mils) material (1); in our studies, the inoculum dried out quickly on the glove surface and the rates of *Salmonella* transfer from single-use gloves decreased as the inoculum drying time increased. Unlike single-use gloves, reusable gloves are made of strong material where drying times were observed to take longer; higher TCs were obtained from reusable gloves after 1 h of inoculum drying than when the inoculum was not dried. The rates of *Salmonella* population decline seen on reusable glove surfaces after 1 h of drying were larger than the rates of decline seen on single-use gloves, affecting the calculated TCs by decreasing the value of the denominator. The transfer of *Salmonella* to tomatoes was less under dry conditions for all scenarios except reusable gloves dried for 1 h.

The method of calculating TCs was dependent upon on the *Salmonella* populations recovered from both the touched surface and initially inoculated surface. In the experiments

TABLE 2. *Salmonella* transfer from inoculated gloves (single use, clean reusable, and dirty reusable) with 1-h-dried inoculum to subsequently touched tomatoes

Tomato no.	Single use			Clean reusable			Dirty reusable		
	Population <sup>a</sup>	TC <sup>b</sup>		Population	TC		Population	TC	
		Avg	Range		Avg	Range		Avg	Range
Glove	5.4 ± 0.1	—	—	5.3 ± 0.3	—	—	4.3 ± 0.3	—	—
T-1	5.0 ± 0.3	0.49 ± 0.3 A <sup>c</sup>	0.1–1.1	4.6 ± 0.7	0.86 ± 1.7 A	0.02–4.9	4/9 <sup>d</sup>	4/9 A	
T-2	4.0 ± 1.3	0.30 ± 0.4 B	0.03–0.98	3.5 ± 1.7	0.87 ± 2.2 A	0.00–2.2	3/9	3/9 A	
T-3	4.0 ± 1.0	0.26 ± 0.5 c	0.00–1.5	3.6 ± 1.2	0.16 ± 0.3 AB	0.00–0.85	3/9	3/9 A	
T-4	3.5 ± 1.3	0.37 ± 0.6 c	0.00–1.4	3.3 ± 1.6	0.35 ± 0.7 A	0.00–1.9	2/9	2/9 A	
T-5	2.9 ± 1.0	0.05 ± 0.1 c	0.00–0.31	2.8 ± 1.5	0.12 ± 0.3 BC	0.00–0.78	3/9	3/9 A	
T-6	2.5 ± 1.1	0.01 ± 0.0 c	0.00–0.05	<2.3 ± 1.6 <sup>e</sup>	7/9		3/9	3/9 A	
T-7	2.2 ± 1.1	0.01 ± 0.0 c	0.00–0.01	<2.3 ± 1.5 <sup>e</sup>	7/9		5/9	5/9 A	
T-8	2.2 ± 1.1	0.01 ± 0.0 c	0.00–0.05	<2.1 ± 1.3 <sup>e</sup>	6/9		2/9	2/9 A	
T-9	8/9	8/9		4/9	4/9		3/9	3/9 A	
T-10	6/9	6/9		5/9	5/9		3/9	3/9 A	
T-11	3/9	3/9		4/9	4/9				
T-12	4/9	4/9		4/9	4/9				
T-13	3/9	3/9		4/9	4/9				
T-14	3/9	3/9		3/9	3/9				
T-15	4/9	4/9		4/9	4/9				
T-16	1/9	1/9		3/9	3/9				
T-17	3/9	3/9		2/9	2/9				
T-18	2/9	2/9		2/9	2/9				
T-19	2/9	2/9		2/9	2/9				
T-20	1/9	1/9		0/9	0/9				
T-21	1/9	1/9		0/9	0/9				
T-22	1/9	1/9		1/9	1/9				
T-23	1/9	1/9		0/9	0/9				
T-24	1/9	1/9		0/9	0/9				
T-25	1/9	1/9		0/9	0/9				

<sup>a</sup> Average *Salmonella* population (log CFU per tomato or glove;  $n = 9$ ) ± standard deviation, or number of positive enrichments.

<sup>b</sup> TC, transfer coefficient, average ± standard deviation and range, or number of positive enrichments.

<sup>c</sup> Letters represent significant differences within columns only; same letters represent no significant differences ( $P \geq 0.05$ ).

<sup>d</sup> Number of positive enrichments of nine total enrichments.

<sup>e</sup> Replications were occasionally negative upon enrichment.

described here, the glove's surface was flat, while the tomatoes were round or curved. This difference in shapes might have influenced the inoculum drying and may not be representative of the way a glove conforms to the shape of a hand and the tomato during harvesting. Significant differences between wet (0 h) and dry (1 h) conditions were observed when gloves were the inoculated surface, while no significant differences were obtained under the same conditions when tomatoes were the inoculated surface. The tomatoes selected for these experiments had no skin injuries; bacteria may detach more easily from the tomato surface than would be expected if injuries were present (18).

Similar studies by Jimenez et al. (13) and Sharps et al. (20) used alternative methods in an attempt to quantify cross-contamination, by calculating percent transfer efficiency and using logarithmic conversion of data for calculating transfer efficiencies, respectively. The highest transfer event reported by Jimenez et al. (13) was the transfer of *Salmonella* Typhimurium from inoculated and dried bell pepper to gloved hands (46.56%). In this study, TCs of *Salmonella* from gloved hands to tomatoes, after

drying the inoculum for 1 h, were comparatively lower for both single-use (0.39) and reusable (0.38) gloves. However, the TCs from inoculated and dried (1 h) single-use (0.29) and reusable (0.48) gloves reported here are higher than those reported from inoculated dried gloves to bell peppers (0.84%) (13). Unfortunately, the type of gloves used and the time of inoculum drying were not reported by Jimenez et al. (13), which may help to explain the differences in rates of transfer in the studies. Sharps et al. (20) quantified the transfer potential of viruses to surfaces under wet and dry conditions. The viruses (norovirus genogroups I and II and murine norovirus 1), when used individually or as a three-virus cocktail, transferred better to the recipient surface (different fruit surfaces or stainless steel) under wet conditions. The results of this study vary depending upon the transfer scenario, and the rates of transfer from tomatoes to both types of glove do not differ between wet (0 h of drying) and dry (1 h) inoculum conditions. *Salmonella* transfer was higher from clean reusable gloves and lower from single-use gloves and dirty reusable gloves upon 1 h of drying. This difference may be explained by differences in

transfer surfaces studied and microorganisms used. Cross-contamination during handling of tomatoes by workers' gloved hands may be minimized when conditions are dry. Wet conditions, including high moisture during harvest and incorrect hand washing and drying, may increase the risk for pathogen movement, and efforts should be made to control the presence of moisture during tomato handling.

The rate of transfer from tomatoes to clean reusable gloves was significantly less than that from tomatoes to clean single-use gloves under wet (0 h) conditions. As high-moisture conditions are more likely to occur during harvest than postharvest handling, a lower risk of *Salmonella* transfer will be provided by reusable glove use. Single-use gloves, common in packinghouses for handling washed, waxed, and dried tomatoes, are not practical for use during harvesting, where chances of tearing exist. The potential for *Salmonella* transfer from clean reusable gloves to tomatoes, even after 24 h of drying, emphasizes the need to clean and sanitize reusable gloves between harvest days to avoid the chances of cross-contamination of more tomatoes from the same glove.

The gloved hands of workers harvesting tomatoes become dirty during the course of the day. There are no recommendations under the current Florida T-GAPs regarding glove changes or cleaning during a harvest day (2). To better understand how the potential risks of cross-contamination change over a typical tomato harvesting day, the rates of *Salmonella* transfer from dirty gloves, as would be seen at the end of the day after fouling material build up, and from clean gloves, typical at the beginning of the day, were compared. *Salmonella* could transfer from dirty reusable gloves to tomatoes under wet and dry inoculum conditions, with significantly less transfer when the inoculum was dry. Under no conditions was *Salmonella* transfer higher from dirty reusable gloves than from clean reusable gloves. Under actual harvesting conditions, the fouling material that builds up on the surface of a glove during harvest will vary depending on a number of soil, physical, and environmental conditions and may further influence the transfer of *Salmonella*. The presence of organic matter on a glove surface makes it porous and rough and may bind *Salmonella* and reduce or prevent its transfer to tomatoes (18). The transfer of *Escherichia coli* O157:H7 between beef and cutting boards under both wet and dry (5 min of drying) inoculation conditions was similarly influenced by the presence of organic matter (8); the TCs from cutting boards to beef and beef to cutting boards decreased with increased amounts of beef tissue present on high-density polyethylene cutting board surfaces.

Determining the number of tomatoes *Salmonella* may transfer to from a contaminated glove improves our understanding of the movement of *Salmonella* from a point source to multiple tomatoes. *Salmonella* transferred to up to 25 tomatoes from single-use gloves, up to 23 tomatoes from clean reusable gloves, and up to 10 tomatoes from dirty reusable gloves; the different numbers of tomatoes contaminated from the different gloves may be influenced by the material of the glove and by the presence of organic matter. The risk of multiple subsequently harvested tomatoes

becoming contaminated during tomato harvest does not increase as gloves become dirty over the course of the day.

The rates of transfer of *Salmonella* to tomatoes did not differ significantly between single-use and reusable gloves. Single-use gloves are generally used in packinghouses and changed frequently upon touching other surfaces, while reusable gloves are used during harvest and get dirty during use. Dirty reusable gloves did not pose a higher risk of *Salmonella* transfer than clean reusable gloves. *Salmonella* survival on glove surfaces was not studied in this project and may be influenced by the presence of organic matter. Drying decreased *Salmonella* transfer to uninoculated surfaces in all transfer scenarios except from reusable gloves dried for 1 h to tomatoes, but drying the inoculum for 1 day is not sufficient to completely prevent transfer from clean reusable gloves to tomatoes. Washing reusable gloves after 1 day of use is an important step to prevent cross-contamination from one day to the next and should not be neglected.

## ACKNOWLEDGMENTS

This research was supported by U.S. Department of Agriculture CSREES NIFSI 2008-51110-0688. We are grateful for the technical assistance of Loretta M. Friedrich, Gwen Lundy, Luis Martinez, Laura Strawn, Lenin O. Interiano-Villeda, Rachel McGegan, and Joshua Vandamm.

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