*Journal of Food Protection, Vol. 76, No. 9, 2013, Pages 1530–1538*
doi:10.4315/0362-028X.JFP-13-098


# Quantifying Transfer Rates of *Salmonella* and *Escherichia coli* O157:H7 between Fresh-Cut Produce and Common Kitchen Surfaces

**DANE A. JENSEN,[1] LORETTA M. FRIEDRICH,[2] LINDA J. HARRIS,[3] MICHELLE D. DANYLUK,[2] AND DONALD W. SCHAFFNER[1]***

[1]*Department of Food Science, Rutgers University, 65 Dudley Road, New Brunswick, New Jersey, 08901-8520;* [2]*Department of Food Science and Human Nutrition, Citrus Research and Education Center, University of Florida, 700 Experiment Station Road, Lake Alfred, Florida 33850; and* [3]*Department of Food Science and Technology, University of California, One Shields Avenue, Davis, California 95616-8598, USA*

MS 13-098: Received 12 March 2013/Accepted 3 May 2013

## ABSTRACT

Cross-contamination between foods and surfaces in food processing environments and home kitchens may play a significant role in foodborne disease transmission. This study quantifies the cross-contamination rates between a variety of fresh-cut produce and common kitchen surfaces (ceramic, stainless steel, glass, and plastic) using scenarios that differ by cross-contamination direction, surface type, produce type, and drying time/moisture level. A five-strain cocktail of rifampin-resistant *Salmonella* was used in transfer scenarios involving celery, carrot, and watermelon, and a five-strain cocktail of rifampin-resistant *Escherichia coli* O157:H7 was used in transfer scenarios involving lettuce. Produce or surface coupons were placed in buffer-filled filter bags and homogenized or massaged, respectively, to recover cells. The resulting solutions were serially diluted in 0.1% peptone and surface plated onto tryptic soy agar with 80 μg/ml rifampin and bismuth sulfite agar with 80 μg/ml rifampin for *Salmonella* or sorbitol MacConkey agar with 80 μg/ml rifampin for *E. coli* O157:H7. When the food contact surface was freshly inoculated, a high amount (>90%) of the inoculum was almost always transferred to the cut produce item. If the inoculated food contact surfaces were allowed to dry for 1 h, median transfer was generally >90% for carrots and watermelon but ranged from <1 to ~70% for celery and lettuce. Freshly inoculated celery or lettuce transferred more bacteria (~2 to ~25% of the inoculum) compared with freshly inoculated carrots or watermelon (approximately <1 to 8%). After 1 h of drying, the rate of transfer from inoculated celery, carrot, and lettuce was <0.01 to ~5% and <1 to ~5% for watermelon. Surface moisture and direction of transfer have the greatest influence on microbial transfer rates.

Fresh-cut produce is increasingly popular for many Americans; fresh and fresh-cut produce have been linked to several outbreaks over the past 20 years *(2, 12, 20, 29)*. U.S. outbreaks with cantaloupe and lettuce occurred in 2012, with a multistate *Escherichia coli* O157:H7 outbreak occurring in fresh-cut leafy greens and a multistate *Salmonella* outbreak occurring in whole cantaloupe in which a recall was extended to watermelons from the same farm *(4, 5)*. Carrot and celery outbreaks are less common, but they do occur, with a 2010 Texas *Listeria monocytogenes* outbreak that was linked to precut celery and a 2004 Finland *Yersinia pseudotuberculosis* outbreak in carrots *(13, 35)*.

Cross-contamination in kitchen environments may play a role in the microbial safety of fresh-cut produce *(39)*. Studies have been done to estimate transfer rates between kitchen utensils, food, cutting boards, and hands *(6, 15, 18, 36)*. Other studies have shown that common kitchen objects (e.g., sponges) may contain many types of bacteria at high levels, which many transfer to foods *(23, 28)*. Furthermore, higher levels of contamination have shown increased attachment to surfaces, when compared to lower inoculum levels *(33)*.

Complicated and multifaceted factors influence both microbial association with and movement to and from surfaces. Surface charges, Van der Waals forces, as well as repulsive electrostatic forces may be involved with initial attachment and subsequent transfer *(34, 37)*. Extracellular matrices composed of proteinaceous and starchy polymers *(1, 30)*, production of surface lipopolysaccharides that have hydrophilic properties *(32)*, and cell surface molecules with nonpolar sites *(8)* have all been thought to influence attachment and transfer. The type *(11, 15, 22, 37, 38)* and the roughness *(8)* of a surface can also effect attachment. Finally, bacteria display a preference for cut/bruised biotic surfaces over intact biotic surfaces, which can increase the risk of outbreaks from fresh-cut produce *(33)*.

This study quantifies the cross-contamination rates between four types of fresh-cut produce (carrots, celery, lettuce, and melon) and four types of common kitchen surfaces (ceramic, glass, plastic, and stainless steel), under

* Author for correspondence. Tel: 732-982-7475; Fax: 732-932-6776; E-mail: schaffner@aesop.rutgers.edu.

freshly inoculated, moist conditions and after a short drying period (1 h postinoculation). All four produce types used in this study are commonly eaten raw and may also come into contact with kitchen surfaces during preparation. The four surfaces used are all found in industrial and home kitchens.

## MATERIALS AND METHODS

**Preparation of domestic food contact surfaces.** Four different food contact surfaces typical of those found in domestic kitchens: glass (3/32 in. [2.4 mm] thick), ceramic tile (glazed), plastic (styrene), and stainless steel (type 304, 18 gauge; onlinemetals.com, Seattle, WA) were ordered online or purchased from a local home improvement store (Winter Haven, FL). Surface materials were cut into coupons (5 by 5 cm). The coupons were disinfected before use by immersion in a solution of 30% sodium hypochlorite (Clorox, Oakland, CA) overnight. The coupons were scrubbed using boiling water, an anionic active detergent, and a sterilized household sponge and then rinsed with boiling water. The coupons were then soaked in 70% ethanol for 1 h, removed, and air dried.

**Produce.** Fresh-cut produce, including mini-peeled carrots, celery, watermelon, and romaine lettuce, in resealable bags or plastic containers were purchased from a local supermarket (Winter Haven, FL). Prior to starting the experiment, produce was stored at 4°C and brought to ambient temperature (~23°C) by placing in a laminar flow hood, without the blower running, for approximately 1 h. Mini-peeled carrots, celery, and cubed watermelon (flesh only) were cut into 10-g pieces. Lettuce was cut into pieces (approximately 3 by 3 cm).

**Selection of strains.** Cocktails of five strains that had been isolated from produce, produce-related commodities, or clinical isolates from produce-associated outbreaks were used. *Salmonella* serovars were used for transfer studies with watermelon, carrots, and celery. Their sources and designations are as follows: Enteritidis (ATCC BAA-1045, raw almonds), Agona (LJH 517, alfalfa sprouts), Gaminara (F2712, orange juice), Michigan (LJH 521, clinical isolate from a cantaloupe outbreak), and Montevideo (G4639, clinical isolate from a tomato outbreak).

A cocktail of five strains of *E. coli* O157:H7 was used for the lettuce transfer studies, their designation and sources are Odwalla outbreak (223), clinical isolate from a cantaloupe outbreak (F658), clinical isolate from a lettuce outbreak (H1730), clinical isolate from an alfalfa sprout outbreak (F4546), and clinical isolate from a spinach outbreak (EC4042). All strains were adapted to grow in the presence of 80 μg/ml rifampin (Thermo FisherScientific, Waltham, MA) through stepwise exposure *(21)*.

**Inoculum preparation.** Prior to each experiment, frozen cultures of each strain were streaked onto tryptic soy agar (Difco, BD, Sparks, MD) with 80 μg/ml rifampin, and incubated at 37°C for 24 h. One isolated colony from each plate was transferred to 10 ml of tryptic soy broth (Difco, BD) with 80 μg/ml rifampin and incubated at 37°C for 24 h. Cultures were then subcultured twice by transferring 0.1 ml of culture to 10 ml of fresh tryptic soy broth with 80 μg/ml rifampin and incubating at 37°C for 24 h. Each strain was centrifuged at 0.6 × *g* for 10 min (Allegra X-12, Beckman Coulter, Fullerton, CA). Cells were washed twice by removing the supernatant and suspending the cell pellet in 10 ml of 0.1% peptone (Difco, BD). Washed cells were suspended in 0.1% peptone at half the original culture volume. Strains were diluted and combined in equal volumes to a concentration of $10^8$ CFU/ml. Final concentrations were verified for each strain by enumeration on tryptic soy agar with 80 μg/ml rifampin.

**Transfer between fresh-cut produce and surfaces.** Sixteen different transfer scenarios were evaluated for each produce type, considering the contact surface type, direction of transfer, and inoculation conditions, for a total for 64 individual scenarios. Each scenario consisted of 20 separate replicates, for a total of 1,280 experiments. For each produce type, 10 μl of inoculum was deposited on the surface in five to eight droplets to obtain a final population of 6 log CFU per coupon. The produce was either immediately touched (approximately 1 to 2 s) to one of the four food contact surfaces (wet) or allowed to dry for 1 h in a biosafety cabinet and then touched (approximately 1 to 2 s) to the contact surface. A contact time of 1 to 2 s was selected based on ease of experimental implementation.

Bacterial concentrations were determined both before and after drying. Transfer from the food contact surface to produce was evaluated by inoculating the food contact surface and subsequently touching the produce surface (also 1 to 2 s). Transfer from produce to the food contact surface was evaluated by inoculating the produce and subsequently touching the food contact surface.

Produce or contact surfaces were placed into a sterile 207-ml Whirl-Pak filter bag (Nasco, Fort Atkinson, WI), 40 ml of DE (Dey/Engley, Thermo Fisher Scientific) was added, and the samples were macerated (Smasher, AES Laboratories, Chemunex, France) for 1 min, or for contact surfaces, massaged with the buffer for 1 min. Samples were serially diluted in 0.1% peptone and surface plated (0.1 ml) onto tryptic soy agar with 80 μg/ml rifampin and bismuth sulfite agar (Difco, BD) with 80 μg/ml rifampin for *Salmonella* or sorbitol MacConkey agar (Difco, BD) with 80 μg/ml rifampin for *E. coli* O157:H7. For low concentrations, 1 ml was surface plated across four plates to decrease the limit of detection to 40 CFU per coupon or piece of produce. Plates were incubated at 37°C for 24 h (tryptic soy agar with 80 μg/ml rifampin and sorbitol MacConkey agar with 80 μg/ml rifampin) or 48 h (bismuth sulfite agar with 80 μg/ml rifampin). Colonies were counted, and *E. coli* O157:H7 or *Salmonella* population levels were expressed as CFU per produce piece or CFU per coupon.

**Data analysis.** Data were compiled, log transformed, and used to create histograms, using Microsoft Excel (Microsoft, Redmond, WA). Our prior laboratory research has shown that percent transfer data must be log transformed to produce normal, or approximately normal, distributions *(6, 26)*. The number of times a particular transfer rate occurred within a target data set (i.e., its frequency) was plotted as a function of log percent transfer rates during the different transfer events. The *x* axis bin width used to create these histograms was in increments of 0.25 log percent transfer because past laboratory experience indicates that bin widths of 0.25 to 0.50 are generally satisfactory *(6, 17, 27)*. Optimal bin size is determined by multiple factors, but generally, the fewer observations available, the larger the bin needs to be to visualize meaningful trends.

The inoculated source is defined as the sum of the amount on both surfaces after the transfer has taken place, so

$$\text{total CFU} = \text{CFU/produce item} + \text{CFU/surface coupon}$$

When the source of contamination is the kitchen surface (coupon),

$$\text{transfer rate (\%)} = (\text{CFU/produce item})/(\text{total CFU}) \times 100$$

When the source of contamination is the produce,

$$\text{transfer rate (\%)} = (\text{CFU/surface coupon})/(\text{total CFU}) \times 100$$

TABLE 1. *Percent transfer of bacteria from inoculated kitchen surfaces to fresh-cut produce*

| Inoculated surface | Drying time (h) | Recipient surface | $\bar{x}$ | σ | Median | Maximum | Minimum | Range |
|---|---|---|---|---|---|---|---|---|
| Ceramic | 0 | Carrot | 79.57 | 4.27 | 80.33 | 86.65 | 71.57 | 15.07 |
| | | Celery | 90.40 | 2.24 | 90.76 | 93.05 | 85.86 | 7.20 |
| | | Lettuce | 91.38 | 2.51 | 91.44 | 94.85 | 86.38 | 8.47 |
| | | Watermelon | 92.55 | 2.24 | 92.52 | 96.27 | 88.62 | 7.65 |
| | 1 | Carrot | 69.58 | 25.05 | 90.91 | 90.91 | 25.00 | 65.91 |
| | | Celery | 48.81 | 11.19 | 52.63 | 65.79 | 22.22 | 43.57 |
| | | Lettuce | 4.13 | 4.31 | 2.54 | 16.67 | 0.49 | 16.18 |
| | | Watermelon | 86.91 | 14.86 | 91.08 | 95.98 | 40.67 | 55.31 |
| Glass | 0 | Carrot | 96.84 | 2.02 | 97.63 | 98.97 | 92.49 | 6.49 |
| | | Celery | 89.88 | 2.06 | 89.49 | 93.07 | 86.31 | 6.75 |
| | | Lettuce | 84.52 | 4.10 | 85.54 | 89.43 | 76.39 | 13.04 |
| | | Watermelon | 86.62 | 2.22 | 86.73 | 90.20 | 82.84 | 7.36 |
| | 1 | Carrot | 99.17 | 1.58 | 99.72 | 99.86 | 94.39 | 5.47 |
| | | Celery | 4.93 | 4.31 | 3.23 | 16.39 | 1.43 | 14.96 |
| | | Lettuce | 1.97 | 2.04 | 1.32 | 9.09 | 0.37 | 8.72 |
| | | Watermelon | 92.52 | 2.33 | 92.13 | 96.97 | 88.69 | 8.28 |
| Plastic | 0 | Carrot | 97.70 | 0.97 | 97.68 | 99.24 | 95.06 | 4.18 |
| | | Celery | 87.72 | 2.06 | 87.78 | 90.84 | 84.00 | 6.84 |
| | | Lettuce | 86.19 | 11.44 | 90.01 | 94.11 | 47.09 | 47.02 |
| | | Watermelon | 97.41 | 0.54 | 97.42 | 98.30 | 95.66 | 2.64 |
| | 1 | Carrot | 99.91 | 0.05 | 99.93 | 99.98 | 99.78 | 0.20 |
| | | Celery | 17.44 | 3.43 | 16.78 | 27.59 | 12.86 | 14.72 |
| | | Lettuce | 18.64 | 25.83 | 4.77 | 73.77 | 2.27 | 71.50 |
| | | Watermelon | 89.39 | 3.22 | 90.05 | 93.56 | 83.33 | 10.23 |
| Stainless | 0 | Carrot | 91.75 | 5.44 | 93.61 | 97.15 | 76.48 | 20.67 |
| | | Celery | 83.36 | 6.72 | 85.61 | 89.59 | 62.13 | 27.46 |
| | | Lettuce | 91.28 | 1.68 | 91.48 | 93.98 | 87.85 | 6.13 |
| | | Watermelon | 94.64 | 2.14 | 95.18 | 97.69 | 89.00 | 8.68 |
| | 1 | Carrot | 84.75 | 6.71 | 86.30 | 95.41 | 70.08 | 25.33 |
| | | Celery | 0.74 | 0.46 | 0.67 | 2.17 | 0.20 | 1.97 |
| | | Lettuce | 39.46 | 19.45 | 40.83 | 71.96 | 7.09 | 64.87 |
| | | Watermelon | 80.00 | 13.67 | 83.40 | 95.60 | 34.88 | 60.72 |

## RESULTS

**Statistical analysis of transfer rates.** Transfer of bacteria from inoculated surfaces to produce and inoculated produce to surfaces is summarized in Tables 1 and 2, respectively. Each table shows five different statistical parameters that were used to characterize the transfer rate (mean [$\bar{x}$], median, standard deviation, minimum transfer observed, maximum transfer observed, and range of transfer rates observed). Many scenario results were indistinguishable (see Fig. 1 for essentially identical transfer rates from freshly inoculated ceramic coupons to carrots, celery, lettuce, and watermelon, for example), and as such, not every one of the 64 specific observations will be noted. The tables will be referenced as needed to augment the discussion of the figures.

**Bacteria transfer from inoculated kitchen surface to produce.** The transfer of *Salmonella* from inoculated ceramic (both freshly inoculated and after drying for 1 h) to carrots, celery, or watermelon or similarly *E. coli* O157:H7 from ceramic to lettuce is shown in Figure 1. When freshly inoculated ceramic surfaces came into contact with any produce type, a very high transfer of bacteria from inoculated ceramic to produce always occurred. After the inoculated ceramic had dried for 1 h, transfer of bacteria to watermelon samples still showed high transfer ($\bar{x}$ = 86.91%; Table 1), but transfer of bacteria to carrots was 25.00 to 91.91% ($\bar{x}$ = 69.58%; Table 1), transfer to celery was 22.22 to 65.79% ($\bar{x}$ = 48.81%; Table 1), and transfer of bacteria to lettuce, 0.49 to 16.67% ($\bar{x}$ = 4.13%; Table 1), was markedly less.

*Salmonella* transfer from inoculated glass (both freshly inoculated and after drying for 1 h) to carrots, celery, or watermelon or *E. coli* O157:H7 to lettuce is shown in Figure 2. When freshly inoculated glass surfaces were touched to any of the produce, a high transfer of bacteria from the glass to the produce was observed. After the inoculated glass had dried for 1 h, transfer to carrot and watermelon samples still displayed high transfer rates (carrot $\bar{x}$ = 99.17%, watermelon $\bar{x}$ = 92.52%; Table 1). After the glass had dried for 1 h, transfer of bacteria to celery was between 1.43 and 16.39% ($\bar{x}$ = 4.93%; Table 1) and *E. coli* O157:H7 transfer of bacteria to lettuce was between 0.37 and 9.09% ($\bar{x}$ = 1.97%).

*Salmonella* transfer from inoculated plastic (both freshly inoculated and after drying for 1 h) to carrots, celery, or watermelon or *E. coli* O157:H7 to lettuce is

TABLE 2. *Percent transfer of bacteria from inoculated fresh-cut produce to kitchen surfaces*

| Recipient surface | Drying time (h) | Inoculated surface | $\bar{x}$ | σ | Median | Maximum | Minimum | Range |
|---|---|---|---|---|---|---|---|---|
| Ceramic | 0 | Carrot | 0.90 | 0.64 | 0.72 | 1.97 | 0.11 | 1.86 |
| | | Celery | 7.72 | 1.47 | 8.28 | 9.77 | 5.23 | 4.54 |
| | | Lettuce | 13.77 | 2.04 | 13.97 | 16.98 | 10.24 | 6.74 |
| | | Watermelon | 1.48 | 0.55 | 1.46 | 2.87 | 0.69 | 2.19 |
| | 1 | Carrot | 0.01 | 0.02 | 0.01 | 0.07 | <0.01 | 0.07 |
| | | Celery | 0.01 | <0.01 | 0.01 | 0.02 | <0.01 | 0.01 |
| | | Lettuce | 0.01 | 0.01 | <0.01 | 0.03 | <0.01 | 0.02 |
| | | Watermelon | 1.65 | 0.47 | 1.61 | 2.71 | 1.04 | 1.67 |
| Glass | 0 | Carrot | 2.32 | 2.33 | 1.48 | 8.25 | 0.21 | 8.05 |
| | | Celery | 8.85 | 1.05 | 9.12 | 10.58 | 6.74 | 3.84 |
| | | Lettuce | 8.86 | 5.93 | 8.46 | 19.19 | 1.59 | 17.60 |
| | | Watermelon | 0.21 | 0.02 | 0.21 | 0.24 | 0.18 | 0.06 |
| | 1 | Carrot | <0.01 | <0.01 | <0.01 | 0.01 | <0.01 | 0.01 |
| | | Celery | 2.96 | 0.44 | 3.02 | 3.54 | 1.94 | 1.60 |
| | | Lettuce | 0.47 | 0.71 | 0.04 | 1.98 | <0.01 | 1.98 |
| | | Watermelon | 0.74 | 0.35 | 0.68 | 1.44 | 0.25 | 1.19 |
| Plastic | 0 | Carrot | 2.47 | 1.37 | 2.52 | 5.04 | 0.56 | 4.48 |
| | | Celery | 8.77 | 2.38 | 8.12 | 13.56 | 5.00 | 8.56 |
| | | Lettuce | 6.40 | 1.50 | 6.66 | 8.92 | 3.86 | 5.06 |
| | | Watermelon | 2.64 | 0.56 | 2.63 | 4.31 | 1.65 | 2.66 |
| | 1 | Carrot | 0.01 | <0.01 | 0.01 | 0.01 | <0.01 | 0.01 |
| | | Celery | 0.01 | 0.01 | 0.01 | 0.03 | 0.01 | 0.03 |
| | | Lettuce | 0.41 | 0.58 | 0.04 | 1.99 | <0.01 | 1.99 |
| | | Watermelon | 0.73 | 0.18 | 0.67 | 1.17 | 0.56 | 0.61 |
| Stainless | 0 | Carrot | 1.24 | 0.40 | 1.28 | 1.86 | 0.53 | 1.32 |
| | | Celery | 15.47 | 4.11 | 15.50 | 23.73 | 6.45 | 17.28 |
| | | Lettuce | 13.97 | 2.54 | 13.81 | 18.23 | 10.13 | 8.10 |
| | | Watermelon | 2.86 | 2.14 | 2.30 | 8.31 | 0.63 | 7.68 |
| | 1 | Carrot | 0.10 | 0.07 | 0.08 | 0.31 | 0.03 | 0.28 |
| | | Celery | 0.29 | 0.61 | <0.01 | 2.02 | <0.01 | 2.02 |
| | | Lettuce | 1.54 | 0.98 | 1.17 | 4.47 | 0.64 | 3.83 |
| | | Watermelon | 2.47 | 0.95 | 2.12 | 4.44 | 1.17 | 3.27 |

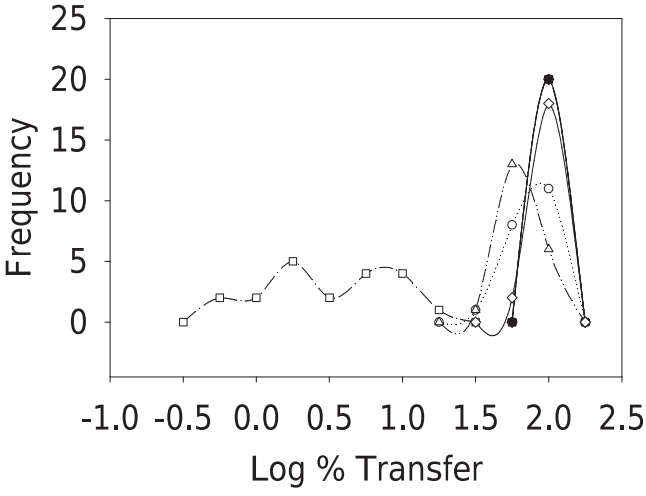


FIGURE 1. *Transfer frequency from inoculated ceramic coupons to produce, with freshly inoculated coupons (solid) and coupons after 1 h of drying (open). Carrot (●), carrot 1 h (○), celery (▼), celery 1 h (△), lettuce (■), lettuce 1 h (□), watermelon (◆), watermelon 1 h (◇). Frequency is the number of times a particular transfer rate occurred within a target data set.*

shown in Figure 3. A high transfer rate of bacteria to produce occurred when freshly inoculated plastic was touched to any of the produce samples. After the plastic had dried for 1 h, a high amount of bacteria still transferred to carrots and watermelon (carrot $\bar{x} = 97.70\%$, watermelon $\bar{x} = 97.41\%$; Table 1). After the inoculated plastic had dried for 1 h, transfer to celery was between 12.86 and 27.59% ($\bar{x} = 17.44\%$; Table 1) and *E. coli* O157:H7 transfer to lettuce ranged from 2.27 to 73.77% ($\bar{x} = 18.64\%$; Table 1).

Figure 4 shows the transfer of *Salmonella* from inoculated stainless steel (both freshly inoculated and after drying for 1 h) to carrots, celery, or watermelon or *E. coli* O157:H7 to lettuce. When freshly inoculated stainless steel was touched to produce, a high rate of bacterial cross-contamination from stainless steel to produce was observed. After the inoculated stainless steel had dried for 1 h, transfer of bacteria to watermelon and carrots still occurred at a high rate. After the stainless steel had dried for 1 h, transfer of bacteria to celery ranged from 0.20 to 2.17% ($\bar{x} = 0.74\%$; Table 1) and transfer of *E. coli* O157:H7 to lettuce ranged from 7.09 to 71.96% ($\bar{x} = 39.46\%$; Table 1).

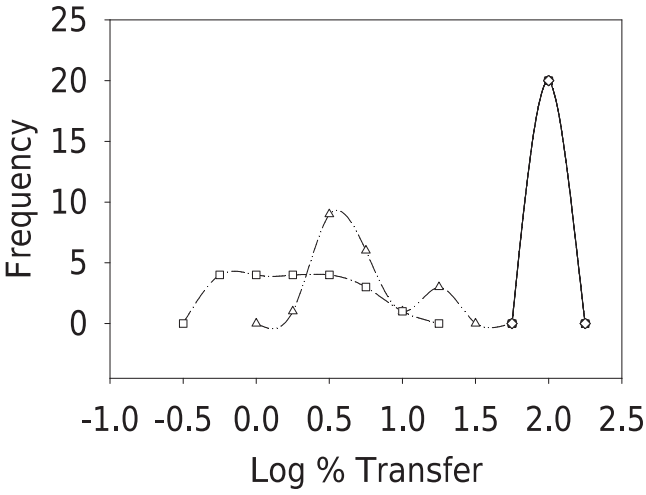


FIGURE 2. *Transfer frequency from inoculated glass coupons to produce, with freshly inoculated coupons (solid) and coupons after 1 h of drying (open). Carrot (●), carrot 1 h (○), celery (▼), celery 1 h (△), lettuce (■), lettuce 1 h (□), watermelon (◆), and watermelon 1 h (◇). Frequency is the number of times a particular transfer rate occurred within a target data set.*

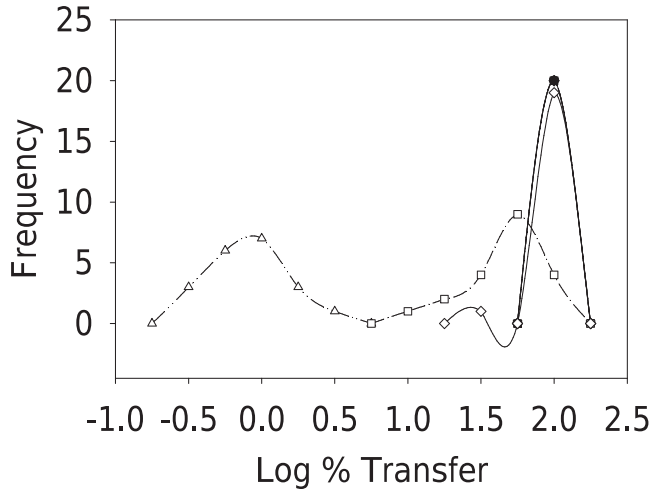


FIGURE 4. *Transfer frequency from inoculated stainless steel coupons to produce, with freshly inoculated coupons (solid) and coupons after 1 h of drying (open). Carrot (●), carrot 1 h (○), celery (▼), celery 1 h (△), lettuce (■), lettuce 1 h (□), watermelon (◆), and watermelon 1 h (◇). Frequency is the number of times a particular transfer rate occurred within a target data set.*

**Bacteria transfer from inoculated produce to kitchen surfaces.** *Salmonella* transfer from inoculated carrots (both freshly inoculated and after drying for 1 h) to four different kitchen surfaces is shown in Figure 5. When freshly inoculated carrots were touched to a ceramic coupon, the transfer rates were highly variable, with a transfer rate range of 0.11 to 1.97% ($\bar{x}$ = 0.90%, $\sigma$ = 0.64), as shown in Table 2. The transfer rate to glass ranged from 0.21 to 8.25%, with a peak around 2% ($\bar{x}$ = 2.32%; Table 2); the transfer rates to plastic ranged from 0.56 to 5.04% ($\bar{x}$ = 2.46%; Table 2); and transfer to stainless steel was 0.53 to 1.86% ($\bar{x}$ = 1.24%; Table 2). After the carrots had dried for 1 h, transfer to all surfaces was noticeably less. Bacteria transfer from inoculated carrots to ceramic was from <0.01 to 0.07% ($\bar{x}$ = 0.01%; Table 2). Bacteria transfer from inoculated carrots to glass, in many cases, had transfer rates below the detection limit (12 of 20 samples), while the remaining transfer rates from carrots to glass varied from <0.01 to 0.01% ($\bar{x}$ < 0.01%; Table 2). After the carrots had dried for 1 h, transfer to plastic was similarly low ($\bar{x}$ < 0.01%; Table 2), while transfer to stainless steel ranged from 0.03 to 0.31% ($\bar{x}$ = 0.10%; Table 2).

The transfer of *Salmonella* from inoculated celery (both freshly inoculated and after drying for 1 h) to four different kitchen surfaces is presented in Figure 6. The transfer rates of *Salmonella* to surfaces were similar for ceramic, glass,

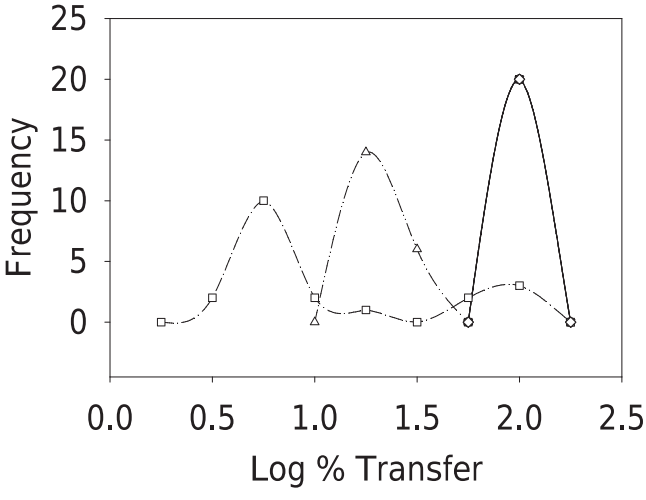


FIGURE 3. *Transfer frequency from inoculated plastic coupons to produce, with freshly inoculated coupons (solid) and coupons after 1 h of drying (open). Carrot (●), carrot 1 h (○), celery (▼), celery 1 h (△), lettuce (■), lettuce 1 h (□), watermelon (◆), and watermelon 1 h (◇). Frequency is the number of times a particular transfer rate occurred within a target data set.*

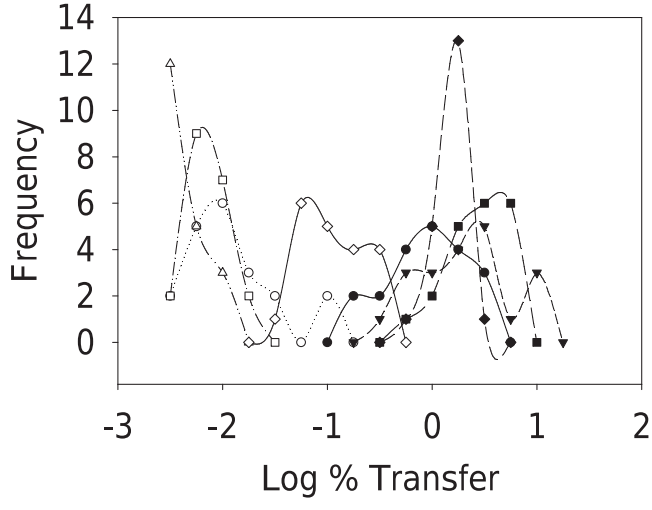


FIGURE 5. *Transfer frequency from inoculated carrots to kitchen surfaces, with freshly inoculated carrots (solid) and carrots after 1 h of drying (open). Ceramic (●), ceramic 1 h (○), glass (▼), glass 1 h (△), plastic (■), plastic 1 h (□), stainless steel (◆), and stainless steel 1 h (◇). Frequency is the number of times a particular transfer rate occurred within a target data set.*

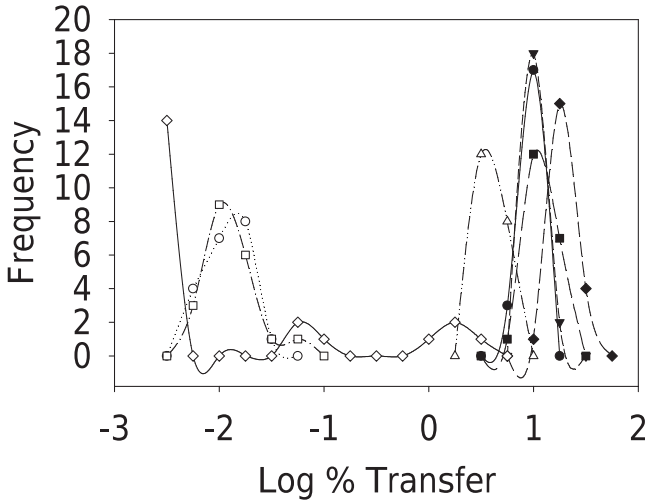


FIGURE 6. *Transfer frequency from inoculated celery to kitchen surfaces, with freshly inoculated celery (solid) and celery after 1 h of drying (open). Ceramic (●), ceramic 1 h (○), glass (▼), glass 1 h (△), plastic (■), plastic 1 h (□), stainless steel (◆), and stainless steel 1 h (◇). Frequency is the number of times a particular transfer rate occurred within a target data set.*

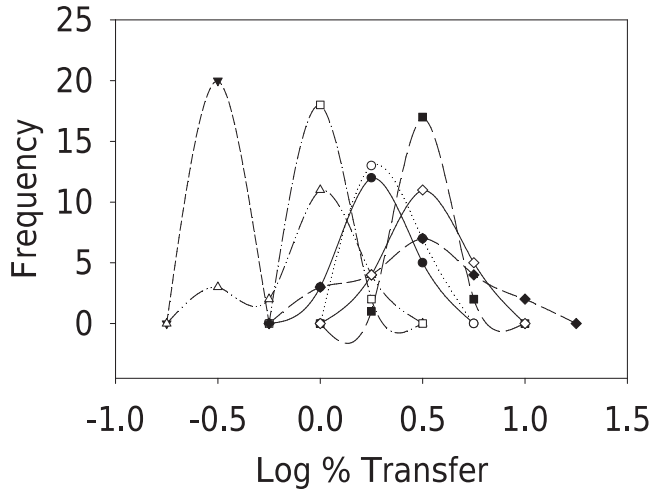


FIGURE 8. *Transfer frequency from inoculated watermelon to kitchen surfaces, with freshly inoculated watermelon (solid) and watermelon after 1 h of drying (open). Ceramic (●), ceramic 1 h (○), glass (▼), glass 1 h (△), plastic (■), plastic 1 h (□), stainless steel (◆), and stainless steel 1 h (◇). Frequency is the number of times a particular transfer rate occurred within a target data set.*

and plastic, but the transfer to stainless steel appeared to have a highest transfer rate of the four surfaces tested. *Salmonella* transfer rates from celery to the four surfaces were the following: ceramic 5.23 to 9.77% ($\bar{x}$ = 7.72%), glass 6.74 to 10.58% ($\bar{x}$ = 8.85%), plastic 5.00 to 13.56% ($\bar{x}$ = 8.77%), and stainless steel 6.45 to 23.73% ($\bar{x}$ = 15.47%; Table 2). After the inoculated celery had dried for 1 h, the transfer of bacteria to three surfaces was quite low: ceramic from <0.01 to 0.02% ($\bar{x}$ = 0.01), plastic from 0.01 to 0.03 ($\bar{x}$ = 0.01), and when *Salmonella* transfer to stainless steel was measured, 14 (70%) of 20 samples were below the detection limit, and the remaining samples showed transfer rates from <0.01 to 2.02% ($\bar{x}$ = 0.29%; Table 2). Curiously, transfer to glass was noticeably higher ($\bar{x}$ = 2.96%, ranging from 1.60 to 1.94%; Table 2), which is also quite evident in Figure 6.

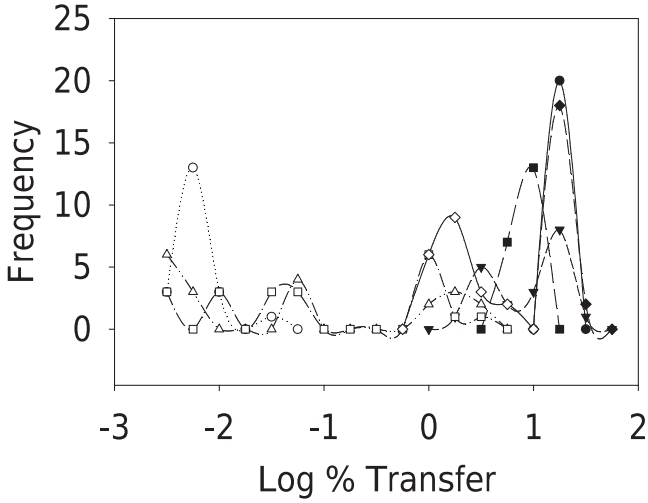


FIGURE 7. *Transfer frequency from inoculated lettuce to kitchen surfaces, with freshly inoculated lettuce (solid) and lettuce after 1 h of drying (open). Ceramic (●), ceramic 1 h (○), glass (▼), glass 1 h (△), plastic (■), plastic 1 h (□), stainless steel (◆), and stainless steel 1 h (◇). Frequency is the number of times a particular transfer rate occurred within a target data set.*

Figure 7 shows the transfer of *E. coli* O157:H7 from inoculated lettuce (both freshly inoculated and after drying for 1 h) to four different kitchen surfaces. When lettuce was freshly inoculated with *E. coli* O157:H7, the transfer rates to ceramic (10.24 to 16.98%, $\bar{x}$ = 13.77%) and stainless steel (10.13 to 18.23%, $\bar{x}$ = 13.97%) were both ~13%. The transfer rates of *E. coli* O157:H7 from inoculated lettuce to glass (1.59 to 19.19%, $\bar{x}$ = 8.86) and to plastic (3.86 to 8.92%, $\bar{x}$ = 6.40%) were lower. When inoculated lettuce was allowed to dry for 1 h, the transfer of bacteria to the ceramic coupon was low and variable (<0.01 to 0.03%), with 3 of 20 samples below the detection limit ($\bar{x}$ = 0.01%; Table 2). Transfer of bacteria from dried inoculated lettuce to stainless steel (0.64 to 4.47%, $\bar{x}$ = 1.54%) was the highest. Transfer of bacteria from dried lettuce to glass ranged from <0.01 to 1.98% ($\bar{x}$ = 0.47%; Table 2), and 6 of 20 samples were below the detection limit. Transfer of bacteria from dried lettuce to plastic was similar to transfer to glass and ranged from <0.01 to 1.99% ($\bar{x}$ = 0.41%; Table 2), with 3 of 20 samples below the detection limit.

*Salmonella* transfer from inoculated watermelon (both freshly inoculated and after drying for 1 h) to four different kitchen surfaces is presented in Figure 8. When freshly inoculated watermelon was touched to ceramic coupons, the transfer rate ranged from 0.69 to 2.87% ($\bar{x}$ = 1.48%; Table 2). When freshly inoculated watermelon was touched to glass coupons, the transfer rate ranged from 0.18 to 0.24% ($\bar{x}$ = 0.21%; Table 2). When freshly inoculated watermelon was touched to plastic coupons, the transfer rate ranged from 1.65 to 4.31% ($\bar{x}$ = 2.64%; Table 2). When

freshly inoculated watermelon was touched to stainless steel coupons, the transfer ranged from 0.63 to 8.31% ($\bar{x}$ = 2.86%; Table 2). Inoculated watermelon that had dried for 1 h had a transfer rate of the *Salmonella* to ceramic coupons from 1.04 to 2.71% ($\bar{x}$ = 1.65%; Table 2). Transfer of bacteria from dried inoculated watermelon to glass ranged from 0.25 to 1.44% ($\bar{x}$ = 0.74; Table 2). Transfer of bacteria from watermelon 1 h after inoculation to plastic ranged from 0.56 to 1.17% ($\bar{x}$ = 0.73%). Transfer to stainless steel ranged from 1.17 to 4.44% ($\bar{x}$ = 2.47%), which was similar to freshly inoculated watermelon transfer rates.

## DISCUSSION

Our study reveals that bacterial transfer is dependent on produce type, surface moisture, and drying time. Surface moisture is related to drying time (where surface moisture decreases with increasing drying time) but is also dependent on produce type, so for very moist items like watermelon, visible surface moisture remains high even after 1 h drying. Our study shows that transfer is less dependent upon kitchen surface type, despite the importance of inert surfaces as suggested in the literature *(8, 11, 22, 24, 37, 38)*. These differences are due, at least in part, to the range of experimental conditions among published studies and perhaps also differences in experimental procedures. Much of the literature investigating the attachment of bacteria and formation of biofilms on surfaces uses incubation periods much longer that the 1 h drying times used here *(8, 24)*. Furthermore, many studies look at attachment to a surface but not cross-contamination from one surface to another *(11, 22, 37, 38)*.

The pressure applied may also play a role in facilitating transfer, although that was not a variable we explicitly considered. Mbithi et al. *(16)* used pressures of 200 and 1,000 g/cm$^2$, both with and without friction, and differences in transfer were small (a ~0.5-log percent transfer difference when pressure is applied). Similarly, data from Kusumaningrum et al. *(14)* indicate that while more transfer generally occurred when slight pressure (20 g/cm$^2$) versus no pressure was used, the differences were also minor (~0.3-log percent transfer difference). Research by D'Sousa et al. *(9)* showed that pressures changes from ~1 to 100 g/cm$^2$ had no effect on virus transfer, while later research from the same laboratory, using similar methods *(10)* showed more transfer at higher (~100 g/cm$^2$) pressures compared with lower pressures (~10 g/cm$^2$), especially under conditions where the inoculum was likely drier.

The contact time may influence transfer, although that was also not a variable we explicitly considered. We used a contact time of 1 to 2 s based on ease of experimental implementation. The literature is unfortunately silent on the distribution of contact times that might occur when produce (or any other food) is handled. Some research *(7)* has shown that contact time generally has little effect on transfer, and other research *(19)* indicates that small (~5 s) differences in hand washing time has little practical effect on contamination reduction on hands. A more systematic study of the influence of contact time is likely warranted.

The most critical observation from this work is that transfer of the bacteria is greater when the direction of transfer is from kitchen surface to produce rather than from produce to kitchen surface. When the transfer was from an inoculated produce item after 1 h of drying to a kitchen surface, we observed the lowest bacterial transfer rates, and the greatest transfer rate (~100%) was observed from a freshly inoculated kitchen surface to produce. The presence of moisture is the most likely explanation for this observation *(10, 25)*, but bacteria may have formed a stronger attachment to produce, due to the presence of complex carbohydrates, which have been shown to increase such attachment *(1, 30, 31, 40)*. Similarly, the higher transfer from kitchen surfaces, regardless of drying time, could be because these abiotic surfaces contain few nutrients, a factor that has been shown to induce detachment *(3)* or that produce surfaces are more complex and ''rougher'' on the microbial scale, thus protecting microbes from transfer.

The high transfer rate to watermelon (Fig. 4), regardless of drying time, may be the result of several factors. Cut watermelon is extremely moist, which may facilitate transfer, even when the originating kitchen surface is dry. After 1 h of drying, the surface of the watermelon pieces were still visibly moist, while the surfaces of carrots, celery, and lettuce were visibly dry. Watermelon cubes possess flat, regular surfaces that likely provided greater surface area than the irregular surfaces of the lettuce, carrot, or celery pieces. A cross-contamination study done by Kusumaningrum et al. *(14)* measured the transfer rates of *Staphylococcus aureus, Salmonella* Enteritidis, and *Campylobacter jejuni* from stainless steel to cucumber slices under no pressure or 20 g/cm$^2$. These researchers observed that all (~100%) of the applied *Salmonella* and *C. jejuni* transferred from the stainless steel to cucumber regardless of pressure applied. Cucumber slices, much like watermelon cubes, have a moist surface facilitating bacterial transfer, and as shown in Figure 4, high transfer rates were seen for transfer from stainless steel for all freshly inoculated conditions, and even after 1 h of drying for carrot and watermelon.

This study highlights the importance of using a higher number of replicates to determine transfer rates (~20), whereas much of the literature reports only the mean and standard deviation from three observations. This is especially important under dryer conditions in which transfer rates may be much more variable. This study also underscores the importance of using clean and dry kitchen surfaces to manage microbial cross-contamination in the kitchen environment. Furthermore, because bacteria readily transfer to moist, fresh-cut produce items, this research further underscores the importance of using separate cutting boards, bowls, and utensils for raw meat and fresh-cut produce. While the causes of foodborne disease are often complex, this research has quantified cross-contamination rates between kitchen surfaces and fresh-cut produce items, which should prove useful in assessing and managing the risks posed by cross-contamination.

## ACKNOWLEDGMENTS

The authors thank Gwen Lundy and Luis Martinez for technical support. The authors also thank the U.S. Food and Drug Administration for their support via grant 1R01FD003672-01 ''Quantifying Risks from Consumer Handling of Fresh-Cut Produce.''

## REFERENCES

1. Barnhart, M. M., and M. R. Chapman. 2006. Curli biogenesis and function. *Annu. Rev. Microbiol.* 60:131–147.
2. Berger, C. N., S. V. Sodha, R. K. Shaw, P. M. Griffin, D. Pink, P. Hand, and G. Frankel. 2010. Fresh fruit and vegetables as vehicles for the transmission of human pathogens. *Environ. Microbiol.* 12:2385–2397.
3. Boyd, A., and A. M. Chakrabarty. 1995. *Pseudomonas aeruginosa* biofilms: role of the alginate exopolysaccharide. *J. Ind. Microbiol. Biotechnol.* 15:162–168.
4. Centers for Disease Control and Prevention. 2012. Multistate outbreak of *Salmonella* Typhimurium and *Salmonella* Newport infections linked to cantaloupe (final update). Available at: http://www.cdc.gov/salmonella/typhimurium-cantaloupe-08-12/index.html. Accessed 4 March 2013.
5. Centers for Disease Control and Prevention. 2012. Multistate outbreak of shiga toxin-producing *Escherichia coli* O157:H7 infections linked to organic spinach and spring mix blend. Available at: http://www.cdc.gov/ecoli/2012/O157H7-11-12/index.html. Accessed 4 March 2013.
6. Chen, Y., K. M. Jackson, F. P. Chea, and D. W. Schaffner. 2001. Quantification and variability analysis of bacterial cross-contamination rates in common food service tasks. *J. Food Prot.* 64:72–80.
7. Dawson, P., I. Han, M. Cox, C. Black, and L. Simmons. 2007. Residence time and food contact time effects on transfer of *Salmonella* Typhimurium from tile, wood and carpet: testing the five-second rule. *J. Appl. Microbiol.* 102:945–953.
8. Donlan, R. M. 2002. Biofilms: microbial life on surfaces. *Emerg. Infect. Dis.* 8:881–890.
9. D'Souza, D. H., A. Sair, K. Williams, E. Papafragkou, J. Jean, C. Moore, and L. Jaykus. 2006. Persistence of caliciviruses on environmental surfaces and their transfer to food. *Int. J. Food Microbiol.* 108:84–91.
10. Escudero, B. I., H. Rawsthorne, C. Gensel, and L. A. Jaykus. 2012. Persistence and transferability of noroviruses on and between common surfaces and foods. *J. Food Prot.* 75:927–935.
11. Fletcher, M., and G. I. Loeb. 1979. Influence of substratum characteristics on the attachment of a marine pseudomonad to solid surfaces. *Appl. Environ. Microbiol.* 37:67–72.
12. Harris, L. J., J. N. Farber, L. R. Beuchat, M. E. Parish, T. V. Suslow, E. H. Garrett, and F. F. Busta. 2006. Outbreaks associated with fresh produce: incidence, growth, and survival of pathogens in fresh and fresh-cut produce. *Compr. Rev. Food Sci. Food Saf.* 2:78–141.
13. Kangas, S., J. Takkinen, M. Hakkinen, U.-M. Nakari, T. Johansson, H. Henttonen, L. Virtaluoto, A. Siitonen, J. Ollgren, and M. Kuusi. 2008. *Yersinia pseudotuberculosis* O:1 traced to raw carrots, Finland. *Emerg. Infect. Dis.* 14:1959–1961.
14. Kusumaningrum, H. D., G. Riboldi, W. C. Hazeleger, and R. R. Beumer. 2003. Survival of foodborne pathogens on stainless steel surfaces and cross-contamination to foods. *Int. J. Food Microbiol.* 85:227–236.
15. Lynch, M. F., R. V. Tauxe, and C. W. Hedberg. 2009. The growing burden of foodborne outbreaks due to contaminated fresh produce: risks and opportunities. *Epidemiol. Infect.* 137:307–315.
16. Mbithi, J. N., V. S. Springthorpe, J. R. Boulet, and S. A. Sattar. 1992. Survival of hepatitis A virus on human hands and its transfer on contact with animate and inanimate surfaces. *J. Clin. Microbiol.* 30:757–763.
17. Montville, R., Y. Chen, and D. W. Schaffner. 2001. Glove barriers to bacterial cross-contamination between hands to food. *J. Food Prot.* 64:845–849.
18. Montville, R., and D. W. Schaffner. 2003. Inoculum size influences bacterial cross contamination between surfaces. *Appl. Environ. Microbiol.* 69:7188–7193.
19. Montville, R., and D. W. Schaffner. 2011. A meta-analysis of the published literature on the effectiveness of antimicrobial soaps. *J. Food Prot.* 74:1875–1882.
20. Painter, J. A., R. M. Hoekstra, T. Ayers, R. V. Tauxe, C. R. Braden, F. J. Angulo, and P. M. Griffin. 2013. Attribution of foodborne illnesses, hospitalizations, and deaths to food commodities by using outbreak data, United States, 1998–2008. *Emerg. Infect. Dis.* 19:507–515.
21. Parnell, T. L., L. J. Harris, and T. V. Suslow. 2005. Reducing *Salmonella* on cantaloupes and honeydew melons using wash practices applicable to postharvest handling, foodservice, and consumer preparation. *Int. J. Food Microbiol.* 99:59–70.
22. Pringle, J. H., and M. Fletcher. 1986. Influence of substratum hydration and adsorbed macromolecules on bacterial attachment to surfaces. *Appl. Environ. Microbiol.* 51:1321–1325.
23. Rusin, P., P. Orosz-Coughlin, and C. Gerba. 1998. Reduction of faecal coliform, coliform and heterotrophic plate count bacteria in the household kitchen and bathroom by disinfection with hypochlorite cleaners. *J. Appl. Microbiol.* 85:819–828.
24. Ryu, J. H., and L. R. Beuchat. 2005. Biofilm formation by *Escherichia coli* O157:H7 on stainless steel: effect of exopolysaccharide and curli production on its resistance to chlorine. *Appl. Environ. Microbiol.* 71:247–254.
25. Sattar, S. A., S. Springthorpe, S. Mani, M. Gallant, R. C. Nair, E. Scott, and J. Kain. 2001. Transfer of bacteria from fabrics to hands and other fabrics: development and application of a quantitative method using *Staphylococcus aureus* as a model. *J. Appl. Microbiol.* 90:962–970.
26. Schaffner, D. W. 2003. Challenges in cross contamination modelling in home and food service settings. *Food Aust.* 55:583–586.
27. Schaffner, D. W., and K. M. Schaffner. 2007. Management of risk of microbial cross-contamination from uncooked frozen hamburgers by alcohol-based hand sanitizer. *J. Food Prot.* 70:109–113.
28. Scott, E., and S. Bloomfield. 1990. The survival and transfer of microbial contamination via cloths, hand and utensils. *J. Appl. Bacteriol.* 68:271–277.
29. Sivapalasingam, S., C. R. Friedman, L. Cohen, and R. V. Tauxe. 2004. Fresh produce: a growing cause of outbreaks of foodborne illness in the United States, 1973 through 1997. *J. Food Prot.* 67:2342–2353.
30. Stepanovic, S., I. Cirkovic, V. Mijac, and M. Svabic-Vlahovic. 2003. Influence of the incubation temperature, atmosphere and dynamic conditions on biofilm formation by *Salmonella* spp. *Food Microbiol.* 20:339–343.
31. Stepanovic, S., I. Cirkovic, L. Ranin, and M. Svabic-Vlahovic. 2004. Biofilm formation by *Salmonella* spp. and *Listeria monocytogenes* on plastic surface. *Lett. Appl. Microbiol.* 38:428–432.
32. Stevenson, G., K. Andrianopoulos, M. Hobbs, and P. R. Reeves. 1996. Organization of the *Escherichia coli* K-12 gene cluster responsible for production of the extracellular polysaccharide colanic acid. *J. Bacteriol.* 178:4885–4893.
33. Takeuchi, K., C. M. Matute, A. N. Hassan, and J. F. Frank. 2000. Comparison of the attachment of *Escherichia coli* O157:H7, *Listeria monocytogenes, Salmonella typhimurium*, and *Pseudomonas fluorescens* to lettuce leaves. *J. Food Prot.* 63:1433–1437.
34. Ukuku, D. O., and W. F. Fett. 2002. Effectiveness of chlorine and nisin-EDTA treatments of whole melons and fresh-cut pieces for reducing native microflora and extending shelf-life. *J. Food Saf.* 22:231–253.
35. U.S. Food and Drug Administration. 2010. FDA lab results positive for *Listeria* at SanGar Fresh Cut Produce. Available at: http://www.fda.gov/Food/FoodSafety/Product-SpecificInformation/FruitsVegetablesJuices/ucm232237.htm. Accessed 6 December 2012.
36. van Asselt, E. D., A. E. de Jong, R. de Jonge, and M. J. Nauta. 2008. Cross-contamination in the kitchen: estimation of transfer rates for cutting boards, hands and knives. *J. Appl. Microbiol.* 105:1392–1401.

37. Van Loosdrecht, M. C., J. Lyklema, W. Norde, G. Schraa, and A. J. Zehnder. 1987. Electrophoretic mobility and hydrophobicity as a measured to predict the initial steps of bacterial adhesion. *Appl. Environ. Microbiol.* 53:1898.
38. Van Loosdrecht, M. C., J. Lyklema, W. Norde, G. Schraa, and A. J. Zehnder. 1987. The role of bacterial cell wall hydrophobicity in adhesion. *Appl. Environ. Microbiol.* 53:1893.
39. Wachtel, M. R., L. C. Whitehand, and R. E. Mandrell. 2002. Association of *Escherichia coli* O157:H7 with preharvest leaf lettuce upon exposure to contaminated irrigation water. *J. Food Prot.* 65:18–25.
40. White, A. P., D. L. Gibson, W. Kim, W. W. Kay, and M. G. Surette. 2006. Thin aggregative fimbriae and cellulose enhance long-term survival and persistence of *Salmonella*. *J. Bacteriol.* 188:3219–3227.