1 **Running Title:** *Listeria* transfer during slicing of onions

2

3 **Title:** Transfer of *Listeria monocytogenes* during mechanical slicing of onions

4

5 Andrew Scollon, Haiqiang Wang, and Elliot T. Ryser*

6

7 Department of Food Science and Human Nutrition, Michigan State University, East

8 Lansing, Michigan 48824, USA

9

10

11

12 **Key words:** *Listeria monocytogenes*, onion, transfer, cross-contamination, hand slicer,
13 mechanical

14

15 ***Author for correspondence.** Tel: (517) 355-8474 ext. 185; Fax: (517) 353-8963; E-
16 mail: ryser@msu.edu

17

18

19

20

21

22

23

ABSTRACT

A series of recalls in 2012 involving diced yellow onions contaminated with *Listeria monocytogenes* from one manufacturer has heightened public health concerns surrounding the preparation, handling and use of raw onions. In response, this study aimed to quantify the extent of *L. monocytogenes* transfer during mechanical slicing of onions. Spanish yellow onions (*Allium cepa*) were dip-inoculated in a 3-strain avirulent *L. monocytogenes* cocktail (M3, J22F and J29H) to contain ~ 8.6, 6.8, or 5.9 log CFU/onion. After air-drying, one inoculated onion was sliced using a mechanical hand slicer, followed by twenty uninoculated onions with the first, fourth, and last slices collected for *Listeria* analysis. Each sample was added to UVM medium, homogenized by stomaching, appropriately diluted and then plated, with or without prior membrane filtration, on Modified Oxford Agar with the plates incubated at 35°C for 48 h. All UVM samples were enriched for 48 h and then streaked to MOX if *Listeria* was not quantifiable by direct plating. After slicing one onion inoculated at 8.6 log CFU/onion followed by 20 uninoculated onions, onions 1, 10 and 20 yielded average *Listeria* populations of 6.7, 3.8, and 2.7 log compared to 3.2, 1.6, and 1.7 log CFU/onion and 3.6, 1.3, and 0.9 log CFU/onion when the initial onion contained 6.8 and 5.9 CFU/onion, respectively. *Listeria* transfer during onion slicing was best described using an exponential decay model which should help to further the understanding of *Listeria* transfer during onion processing and provide the industry with guidelines to improve end product safety.

Increased consumption of raw, minimally processed, fresh-cut produce is raising new safety concerns regarding the risk of foodborne illness. In 2012, the safety of raw onions came under scrutiny following the recall of more than 900 kg of diced onions from one major producer. Due to the widespread use of onions as an ingredient in many ready-to-eat foods, this recall caused a ripple effect through the industry with eleven additional recalls issued across the United States and Canada, all without incident (Food and Drug Administration, 2012). In follow-up investigations, the implicated strain of *L. monocytogenes* was recovered from multiple sites within the incriminated processing facility, including a chute under an inspection table, a shroud on a peeling machine, and the loading hopper of a peeling machine (California Department of Public Health, 2012). Based on these locations, the pathogen may have entered the facility on the incoming intact onions and then been spread to subsequent onions during processing.

The incidence of *Listeria* in fresh produce is typically quite low with multiple studies failing to recover *Listeria* from onions sold at retail (Kaneko, et al., 1999). Various salads containing onions have tested positive for *Listeria*, however no direct link was shown to onions (Beuchat, 1996; Sizmur & Walker, 1988). Nonetheless, contamination of onions via contact with soil and food processing equipment remains a possibility due to the presence of *Listeria* in food processing facilities (Aguado et al., 2004, and Beuchat & Brackett, 1990) as supported by the aforementioned recall.

Commercial onion processing begins with “topping and tailing” where the root and sprout ends of the onion bulb are simultaneously removed by blades either before or after the outer skin is peeled using a corkscrew-type conveyer. Once peeled, onions are typically washed in water containing 50 – 200 ppm chlorine (Suslow, 1997), and

conveyed to a slicer or dicer. In contrast to diced onions which are washed again, sliced onions are packaged directly without further washing in order to keep the slices intact. On-site slicing of onions via manual slicers, similar to that used in this study, is a common practice in restaurants.

A series of previous studies have addressed the transfer of *L. monocytogenes* as well as *Escherichia coli* O157:H7 and *Salmonella* during grinding of beef (Gilbert 1969, Florez 2002), slicing of delicatessen meats, (Lin, et al., 2006, Pérez-Rodríguez et al., 2007, Sheen & Hwang, 2011, Vorst et al., 2004), shredding of leafy greens (Beuchat & Brackett, 1990; Buchholz et al., 2012a, Buchholz et al., 2012b, Nou & Lou, 2010), and slicing/dicing of tomatoes (Chen et al., 2014). In several reports, log-linear, Weibull, and exponential decay models (Pérez-Rodríguez, et al., 2007; Sheen & Hwang, 2010) were used to better describe the trends in bacterial transfer for different processing scenarios.

The presence of *Listeria* in onions resulting in a recall prompted the current study which aimed to quantify the extent of *L. monocytogenes* transfer during mechanical slicing of onions. The ability of *L. monocytogenes* to transfer from one inoculated onion to different slicer components and then to sequentially sliced onions was also assessed along with the impact of inoculation level on transfer.

MATERIALS AND METHODS

Experimental design. Three avirulent and five virulent *L. monocytogenes* strains were first assessed for attachment, growth characteristics, and survival on onions during

storage. Thereafter, transfer of the three avirulent strains from one inoculated to multiple uninoculated onions during mechanical slicing was quantified by direct plating, with these findings then subjected to mathematical modeling to predict the extent of transfer.

Bacterial strains. Three avirulent *L. monocytogenes* strains - M3 serotype 1/2a (Hly⁻, parent strain Mackaness), J22F serotype 4b (Hly⁺, purB mariner-based mutant of H7550- Cd^S, parent strain NCTC 10527), and J29H serotype 4b (Hly⁻, parent strain NCTC 10527) (obtained from Dr. Sophia Karthariou, North Carolina State University, Raleigh, NC) were used in all slicing experiments. In addition, five virulent strains - R2-499 (serotype 1/2a, 2000 deli turkey outbreak), N3-008 (serotype 4b, 1981 coleslaw outbreak), N3-031 (serotype 1/2a, 1988 hot dog outbreak) , J1-110 (serotype 4b, 1985 Mexican-style cheese outbreak 1985) and J1-177 (serotype 1/2b, 1997 clinical isolate) previously obtained from Dr. Martin Wiedmann (Cornell University, Ithaca, NY) were used to compare attachment, growth, and survival of the avirulent strains. All strains were stored at -80°C in trypticase soy broth containing 0.6% (w/v) yeast extract (TSBYE, Becton, Dickinson and Company, Sparks, MD) and 10% (v/v) glycerol. Each strain was initially streaked for isolation to plates of trypticase soy agar containing 0.6% (w/v) yeast extract (TSAYE, Becton, Dickinson and Company) and incubated for 24 h at 35°C. Thereafter, an isolated colony of each strain underwent two consecutive 24 h/35°C transfers in TSBYE. When used as cocktails, the three avirulent strains were combined in equal volumes and appropriately diluted to obtain populations of ~ 8.9, 6.4, or 5.3 log CFU/ml for onion inoculation, with these levels confirmed by plating appropriate dilutions on Modified Oxford Agar (MOX, Neogen Corp., Lansing, MI).

Onions. Multiple lots of Spanish yellow onions (*Allium cepa*) were purchased from a local supplier (Stan Setas Produce Company, Lansing, MI) over a period of four months with each lot stored at 4°C for no more than 7 d before use. The root and sprout ends of each onion were removed using a sterile kitchen knife. After hand peeling the outer skin, the onions were tempered to room temperature (23°C ± 2°C) and weighed prior to slicing.

Attachment, growth, and survival. Attachment was assessed using the microtiter plate assay developed by Stepanović et al. (2004). Triplicate wells of a 96 well untreated polystyrene microtiter tissue culture plate (Flat Bottom, BD Falcon, Franklin Lakes, NJ) were separately filled with 24 h cultures of the virulent and avirulent strains previously diluted in sterile TSBYE to ~10³ CFU/ml with three wells containing TSBYE alone serving as negative controls. Following 48 h of incubation at 23°C, the wells were emptied, rinsed 3 times with sterile 0.1% Phosphate Buffered Solution (PBS) to remove unattached cells, and air dried. Methanol (Fisher Chemicals, Fair Lawn, NJ) was then added to fix the attached cells followed by 200 µl of 2% Crystal Violet (Remel, Lenexa, KS) for staining. After emptying, thoroughly rinsing and treating the wells with 200 µl of 33% (v/v) glacial acetic acid (Sigma Chemical Company, St. Louis, MO) to solubilize the crystal violet, bacterial adherence was quantified by measuring optical density at 550 nm with a microplate reader (Bio-Tek Instruments Inc. Model SIAFR, Winooski, VT).

Growth was assessed by inoculating 200 ml of TSBYE with ~10⁴ CFU in triplicate (3 flasks/ culture). After 0, 3, 6, 9, 12, and 24 h of incubation at 35°C, 1 ml aliquots were appropriately diluted and plated on TSAYE containing esculin 0.1% (w/v) and ferric ammonium citrate 0.05% (w/v) (TSAYE-EF) with the plates counted after 24 h

of incubation at 35°C. Generation times were determined using values obtained during logarithmic growth according to Eq. (1):

$$G = \frac{t}{3.3 \log^b / B} \quad (1)$$

where t = time in minutes, b = *L. monocytogenes* population at 3 h, and B = *L. monocytogenes* population at 9 h.

Survival was assessed by immersing whole peeled onions in either a 3-strain avirulent or 5-strain virulent *L. monocytogenes* cocktail containing ~ 6.4 log CFU/ml for 2 min to achieve ~ 10⁶ CFU/onion, followed by air-drying in a biosafety cabinet for 90 min. Initially and after 4, 24, 48, 120, and 168 h of storage at 4°C, three onions were placed in individual Whirl-pakTM bags containing 100 ml of PBS, hand-rubbed for 90 s, appropriately diluted, and plated on MOX to quantify *Listeria*.

Onion slicer. A manual onion slicer (NEMCO model 56750-2, Hicksville, IN) yielding nine 0.5 cm-thick slices was used for all experiments. In order to identify the product contact areas on the slicer for subsequent sampling, Glo Germ (Glo-Germ Company, Moab, UT) was used as reported previously (Buchholz, et al., 2012a, Vorst et al., 2006). One onion was fully submerged in 0.5% (w/v) Glo-Germ solubilized in 5% ethanol, dried for 90 min, and then manually sliced, after which the components of the slicer were viewed under UV light (352 nm, Sankyo Denki Co., Ltd, Tokyo, Japan). Using this procedure, two 100 cm² product contact areas of the slicer – the pusher plates and blades- were identified for subsequent sampling (Figure 1).

Sampling protocol. Initially, the distribution of *L. monocytogenes* on the different onion slices was determined by dip inoculating one onion to achieve ~ 8.9 log CFU/onion, slicing the inoculated onion, adding each of the 8 to 9 slices to an individual

Whirl-pakTM bag, and enumerating each on MOX. Following the inoculated onion, one uninoculated onion was then sliced, sampled, and enumerated similarly. Based on statistical similarity ($P < 0.05$), a 3-slice method was used where slices 1, 4, and the last slice of at least half the full thickness were composited for testing in the following slicing experiments.

***Listeria* transfer during slicing.** Whole peeled onions were immersed in the 3-strain avirulent cocktail containing ~8.9, 6.4, or 5.3 log CFU/ml for 2 min and then air-dried in a biosafety cabinet for 90 min, giving populations of ~8.6, 6.8, or 5.9 log CFU/onion. Although unusually high, these inoculum levels were necessary to obtain quantifiable results for modeling in the subsequent slicing experiments. Initial populations were confirmed by hand-rubbing two inoculated onions in separate Whirl-pakTM bags containing 100 ml of University of Vermont Medium (UVM, Neogen) for 60 s followed by plating on MOX.

Transfer of *L. monocytogenes* to the slicer was assessed by slicing one inoculated onion, after which 100 cm² areas of the pusher plates and blades were disassembled and then sampled using the 1-ply composite tissue method developed by Vorst et al. (2004). Sequential *L. monocytogenes* transfer to previously uncontaminated onions was then assessed by slicing one inoculated onion immediately followed by twenty uninoculated onions with the first, middle and last slice from each onion composited and examined for numbers of *Listeria*. After slicing twenty uninoculated onions, the pusher plate and blades were disassembled with 100 cm² areas again sampled using the 1-ply composite tissue method. All slicing experiments were conducted in triplicate. After each experiment, the slicer was disassembled, washed with a brush to physically remove

debris, and then sanitized with 200 ppm Quorum V (Ecolab, St. Paul, MN), rinsed in tap water, and sprayed with 70% ethanol followed by drying in a biosafety cabinet under UV light for 10 min before reassembly.

Microbial analysis. All samples were added to Whirl-pak™ bags containing 50 ml of UVM, homogenized by stomaching (Stomacher 400 Circulator, Seward USA, Davie, FL) for 1 min at 300 rpm, appropriately diluted in PBS, and plated, with or without prior filtration (0.45 µm filter, Millipore Corporation, Billerica, MA) using a vacuum pump (Model E46046, EMD Millipore Corporation) on MOX with the UVM-diluted samples enriched at 35°C. All colonies resembling *Listeria* were counted after 48 h of incubation at 35°C. After slicing and enumeration in the first of three experiments, multiple colonies were selected and confirmed as *Listeria* using the Neogen Reveal® 2.0 *Listeria* test kit (Neogen). When the MOX plates were negative for *Listeria*, the UVM enrichments were streaked to plates of MOX which were then incubated at 35°C for 48 h and examined for the presence or absence of *Listeria*.

Evaluation of transfer model. An exponential decay model from a previous study (Sheen & Hwang, 2010) was used to describe the *L. monocytogenes* transfer pattern during onion slicing. The model used to fit the data is shown in Eq. (2):

$$Y = A \cdot e^{X/B} \quad (2)$$

where Y (dependent variable) is the log CFU/onion transferred and X (independent variable) is the number of the specific uninoculated onion that was sliced. A and B are transfer model parameters. The above equation was fitted using the *nlinfit* algorithm of MATLAB (R2012a, MathWorks, Natick, MA). The estimated parameters, normalized

root mean squared errors (NRMSE) of the model, shown in Eq. (3), and asymptotic 95% confidence intervals of the parameters were then estimated:

$$\text{NRMSE} = \frac{\text{RMSE}}{(Y_{\max} - Y_{\min})} \quad (3)$$

where NRMSE is the normalized root mean squared error, RMSE is the root mean squared error, $Y_{\max}$ is the maximum value taken by Y , and $Y_{\min}$ is the minimum value taken by Y .

Statistical analysis. All experiments were performed in triplicate. *Listeria* populations were reported as log CFU/onion $\pm$ SE for the inoculated and uninoculated onions or log CFU/100 cm² $\pm$ SE for the slicer surface samples. For subsequent analysis of the *Listeria* transfer data, onion samples positive by enrichment were assumed to contain 1 CFU, whereas surface samples negative by enrichment were assumed to contain 0.5 CFU. Onion samples negative by enrichment were not included in the analysis. The Tukey-Kramer HSD test was performed using JMP 10 (SAS Institute Inc., Cary, NC). Statistical significance was set at $P < 0.05$.

RESULTS

***Listeria* attachment, growth and survival.** In the microtiter plate assay, avirulent strain M3 exhibited significantly greater attachment (average OD of 0.62) ($P < 0.05$) compared to the other seven strains (average OD of 0.11 – 0.13). All strains except M3 grew similarly ($P > 0.05$) with generation times of 50.9 ± 1.7 , 33.7 ± 1.0 , 32.6 ± 0.6 , 36.2 ± 0.8 , 33.5 ± 0.3 , 39.1 ± 1.3 , 40.9 ± 3.0 , and 36.8 ± 0.7 min for M3, J22F, J29H, R2-499, N3-031, N3-008, J1-110, and J1-177, respectively. Survival of *L. monocytogenes* on

whole peeled onions was similar ($P > 0.05$) for both the avirulent and virulent cocktails, with average populations of 5.2 ± 0.3 and 5.6 ± 0.2 log CFU/onion after 7 d of storage, respectively (Table 1). Avirulent *Listeria* populations decreased significantly ($P < 0.05$) after 7 d of storage at 4°C.

***Listeria* distribution on onions.** After slicing one inoculated onion, two uninoculated onions were sliced with the numbers of *Listeria* individually determined for each slice and then compiled to obtain the population per onion (Figure 2). Estimated *Listeria* populations per onion were not significantly ($P > 0.05$) different when the first, fourth and last slices were composited, as compared to sampling each individual slice for determining the population per onion.

***Listeria* transfer to the slicer.** After slicing one inoculated onion, *L. monocytogenes* was recovered from the pusher plates and blades (Figure 3). Except for the highest inoculum, the pusher plates yielded similar ($P > 0.05$) *Listeria* populations after slicing one inoculated onion followed by twenty uninoculated onions as compared to *Listeria* populations recovered after slicing one inoculated onion. At all inoculation levels, the blades yielded significantly ($P < 0.05$) lower *Listeria* populations after slicing one inoculated onion followed by twenty uninoculated onions as compared to *Listeria* populations recovered after slicing one inoculated onion.

***Listeria* transfer to uninoculated onions from the slicer.** After slicing one inoculated followed by twenty uninoculated onions, all but one sample at the lowest inoculation level yielded detectable levels of *Listeria* in one or more replicates. At the highest inoculation level of ~ 8.6 log CFU/onion, onions 1, 10 and 20 yielded average *Listeria* populations of 6.7 ± 0.2 , 3.8 ± 0.1 , and 2.7 ± 0.4 log CFU/onion, respectively

(Figure 4). *Listeria* was sporadically detected by enrichment in one or more replicates for all onions when the inoculated onion contained ~ 6.8 log CFU/onion. Similarly, *Listeria* was sporadically recovered after enrichment in one or more replicates from all but the 17th onion which was negative by enrichment in all three replicates when the inoculated onion contained ~ 5.9 log CFU/onion.

Transfer model and parameter estimation. The parameter estimations after fitting the equation for each transfer scenario are shown in Table 2. The model was a better fit at the higher inoculation levels as shown by the lower NRMSE values (Table 2). The observed values and predicted lines for transfer were then plotted, along with the confidence band (CB) for the prediction line (Figure 4). Although the prediction lines generally reflected the trends in *Listeria* transfer, the CBs were relatively wide, with the CB width between 0.6 – 1.4, 0.5 – 1.1, and 0.7 – 1.5 log CFU/onion at inoculation levels of ~ 8.6 , 6.8, and 5.9 log CFU/onion, respectively. In addition, at the two lower onion inoculation levels, the model over-predicted the low numbers of *Listeria* observed after continued slicing as shown in Figure 5.

DISCUSSION

This research clearly shows the potential for cross-contamination from inoculated to uninoculated onions during sequential slicing. After slicing twenty onions, detectable levels of *L. monocytogenes* remained on both the pusher plates and the blades, allowing for further transfer. These observations are consistent with previous cross-contamination studies in which foodborne pathogens were shown to readily transfer between slicers or mechanical shredders and deli meats (Vorst et al., 2006), lettuce (Buchholz, et al., 2012a;

274 Buchholz, et al., 2012b), and celery (Kaminski et al., 2014) during simulated commercial
275 processing.

276 In this study, after slicing one inoculated onion followed by twenty uninoculated
277 onions, *Listeria* populations decreased significantly ($P < 0.05$) on the blade while the
278 pusher plates retained a greater proportion of the initial *Listeria* population. This may be
279 partially explained by the shear force on the blade surface which readily removed loosely
280 attached cells. The limited reduction of *Listeria* on the pusher plate surfaces suggests that
281 fewer cells were transferred due to static contact with such transfer likely to continue if
282 additional onions had been sliced.

283 Once sliced, onions have a shelf life of approximately 14 days at 4°C, which may
284 be sufficient time for small numbers of *Listeria* to reach potentially hazardous levels
285 (Farber et al., 1998). When an onion is sliced, various sulfur compounds possessing
286 antimicrobial activity, including *S*-alk(en)yl cysteine sulfoxide metabolites, are released
287 and converted to other inhibitory compounds (Lancaster & Collin, 1981, Ramos, et al.,
288 2006). While onion juice is known to possess antimicrobial activity (Kim et al., 2004,
289 Ramos et al., 2006, Ye et al. 2013), *L. monocytogenes* was reportedly able to grow on
290 sliced onions when stored at 10°C (Farber et al., 1998), and in the current study, persisted
291 on the surface of whole peeled onions stored at 4°C. Inconsistent temperature control
292 throughout cold-chain distribution is also raising concern over microbial growth in fresh-
293 cut produce (Nunes et al., 2009, Zeng, et al., 2014).

294 Appropriate use of Good Agricultural Practices (GAPs), Good Manufacturing
295 Practices (GMPs), and sanitation programs as set forth by the FDA is critical for
296 minimizing contamination prior to processing, reducing cross-contamination during

processing, and removing contaminants after processing. Transfer to and from equipment surfaces, resulting in cross-contamination of product is clearly possible during mechanical slicing of onions. Onions can either be manually pushed by a plate as in this study or fed into a chute, which acts as a stationary pusher plate while the blades move perpendicular to the chute. Regardless of the type of slicer, thorough cleaning and sanitizing of both the blades and the pusher plates/chute after use are equally important as evidenced by the continued presence of *Listeria* on component surfaces after slicing twenty uninoculated onions. Due to difficulties associated with cleaning closely spaced (0.64 cm separation) parallel plates, it is important to follow manufacturer cleaning protocols that recommend complete disassembly prior to cleaning and sanitizing. Neal (2013) reported that including both a visual demonstration and written cleaning/sanitizing instructions in worker training programs increased the overall cleanliness of delicatessen slicers, reinforcing the importance of proper staff training.

The exponential decay model applied in this study was able to describe the transfer behavior of *L. monocytogenes* during slicing of onions, which is consistent with previous reports (Aarnisalo et al., 2007; Sheen & Hwang, 2010; Sheen & Hwang, 2011). However, the model was less accurate using the low inoculum, due to variability in the transfer data as seen by the relatively high NRMSE (Table 2). Other studies were conducted by our group to evaluate the transfer behavior of *Salmonella* during slicing/pilot-scale dicing of tomatoes (Wang & Ryser, 2014), for which this same exponential decay model was successfully applied. In addition, an analysis of the impact of various different processing variables (e.g., temperature, slice thickness, and tomato variety) on *Salmonella* transfer during slicing showed the importance of properly

controlling certain conditions during slicing to minimize bacterial cross-contamination (unpublished data). Thus, in future studies it will be necessary to more closely examine those factors that impact *Listeria* transfer during onion slicing. When the prediction lines were compared for the different inoculation levels, the highest inoculation level (8.6 log CFU/onion) yielded a much steeper decay pattern compared to the other two inoculation levels due in part to the nearness of the initial inoculation to the limit of detection for the lower inoculations. Variation in observed results can also be attributed to differences in onion diameter and density.

In conclusion, this is the first study to quantitatively assess *L. monocytogenes* transfer during slicing of onions and demonstrate the spread of contamination from a single onion to sequentially sliced product. Since *L. monocytogenes* has been shown to survive in refrigerated onions and grow during unintentional temperature abuse, vegetable slicers should be properly cleaned and sanitized during prolonged use to minimize the risk of cross-contamination and possible product recalls, with maintenance of the cold chain also needed to minimize pathogen growth in the finished product.

ACKNOWLEDGEMENTS

Financial support for this work was provided by the U.S. Department of Agriculture under grant number 2011-51110-31027.

References

- 342 Aarnisalo, K., Sheen, S., Raaska, L., & Tamplin, M. (2007). Modelling transfer of
 343 *Listeria monocytogenes* during slicing of ‘gravad’ salmon. *International Journal of Food*
 344 *Microbiology*, 118, 69–78.
- 345 Aguado, V., Vitas, A. I., & I. García-Jalón, I. (2004). Characterization of *Listeria*
 346 *monocytogenes* and *Listeria innocua* from a vegetable processing plant by RAPD and
 347 REA. *International Journal of Food Microbiology*, 90, 341–347.
- 348 Beuchat, L. R., and Brackett, R.E. (1990). Survival and growth of *Listeria*
 349 *monocytogenes* on lettuce as influenced by shredding, chlorine treatment, modified
 350 atmosphere packaging, and temperature. *Journal of Food Science* 55, 755–758.
- 351 Beuchat, L.R. (1996). Pathogenic microorganisms associated with fresh produce. *Journal*
 352 *of Food Protection*, 59, 204–216
- 353 Buchholz, A. L., Davidson, G. R., Marks, B. P., Todd, E. C., & Ryser, E. T. (2012a).
 354 Quantitative transfer of *Escherichia coli* O157:H7 to equipment during small-scale
 355 production of fresh-cut leafy greens, *Journal of Food Protection*, 75, 1184–1197.
- 356 Buchholz, A. L., Davidson, G. R., Marks, B. P., Todd, E. C., & Ryser, E. T. (2012b).
 357 Transfer of *Escherichia coli* O157:H7 from equipment surfaces to fresh-cut leafy greens
 358 during processing in a model pilot-plant production line with sanitizer-free water. *Journal*
 359 *of Food Protection*, 75, 1920–1929.
- 360 California Department of Public Health (2012). Gills Onions *Listeria* Mono. Available at:
 361 <https://www.cdph.ca.gov/HealthInfo/Documents/fdbEIRGO2012.pdf> Accessed January
 362 18, 2015

- 363 Chen, D., Zhao, T., & Doyle, M.P. (2014). Transfer of foodborne pathogens during
364 mechanical slicing and their inactivation by levulinic acid-based sanitizer on slicers.
365 *Food Microbiology*, 38, 263–269.3.
- 366 Farber, J.M., Wang, S.L., Cai, Y. & Zhang, S. (1998). Changes in populations of *Listeria*
367 *monocytogenes* inoculated on packaged fresh-cut vegetables. *Journal of Food Protection*,
368 61, 192–195.
- 369 Florez, R. A. & Tamplin, M.L. (2002). The distribution patterns of *Escherichia coli*
370 O157:H7 in ground beef produced by a laboratory-scale grinder. *Journal of Food*
371 *Protection*, 65, 1894–1902
- 372 Food and Drug Administration. (2012). Gills onions expands voluntary recall of diced
373 and slivered red and yellow onions, and diced onion and celery mix because of possible
374 health risk, July 25, 2012. Available at: [http://www.fda.gov/](http://www.fda.gov/Safety/Recall/ucm313399.htm)
375 [Safety/Recall/ucm313399.htm](http://www.fda.gov/Safety/Recall/ucm313399.htm) Accessed 20 May 2013
- 376 Food and Drug Administration. (2010). Commodity specific food safety guidelines for
377 the production, harvest, post-harvest, and valued-added unit operation of green onions.
378 February 26, 2010. Available at:
379 [http://www.fda.gov/food/guidanceregulation/guidancedocumentsregulatoryinformation/p](http://www.fda.gov/food/guidanceregulation/guidancedocumentsregulatoryinformation/produceplantproducts/ucm203094.htm)
380 [roduceplantproducts/ucm203094.htm](http://www.fda.gov/food/guidanceregulation/guidancedocumentsregulatoryinformation/produceplantproducts/ucm203094.htm). Accessed 20 February 2014
- 381 Gilbert, R. J. (1969). Cross contamination by cooked-meat slicing machines and cleaning
382 cloths. *Journal of Hygiene*, 67, 249–254.

- 383 Kaminski, C. N., Davidson, G. R., & Ryser, E. T. 2014. *Listeria monocytogenes* transfer
384 during mechanical dicing of celery and growth during subsequent storage. *Journal of*
385 *Food Protection*, 77, 765–71.
- 386 Kaneko, K., Hayashidani, H., Takahashi, K., Shiraki, Y., Limawongpranee, S. & Ogawa,
387 M. (1999). Bacterial Contamination in the environment of food factories processing
388 ready-to-eat fresh vegetables. *Journal of Food Protection*, 62, 800–804
- 389 Kim, J., Kim, Y., & Kyung, K. (2004). Inhibitory activity of essential oils of garlic and
390 onion against bacteria and yeasts. *Journal of Food Protection*, 67, 499–504.
- 391 Lancaster, J. E., & Collin, H. A. (1981). Presence of alliinase in isolated vacuoles and of
392 alkyl cysteine sulfoxides in the cytoplasm of bulbs of onion *allium cepa*. *Plant Science*
393 *Letters*, 22, 169–176.
- 394 Lin, C., Takeuchi, K., Zhang, L., Dohm, C. B., Meyer, J. D., Hall, P. A., & Doyle M. P.
395 (2006). Cross-contamination between processing equipment and deli-meats by *Listeria*
396 *monocytogenes*. *Journal of Food Protection*, 69, 71–79.
- 397 Neal, J.A. (2013). Comparative analysis of training delivery methods for new employees
398 cleaning and sanitizing retail deli slicers: An exploratory study. *Food Control* 29, 149–
399 155.
- 400 Nou, X., & Luo, L. (2010). Whole leaf wash improves chlorine efficacy for microbial
401 reductions and prevents cross-contamination during fresh-cut lettuce processing. *Journal*
402 *of Food Science*, 75, M283–M290.

- 403 Nunes, M., Edmond, J.P., Rauth, M., Dea, S., & Chau, K. (2009). Environmental
404 conditions encountered during typical consumer retail display affect fruit and vegetable
405 quality and waste. *Postharvest Biology and Technology*, 51, 232–241
- 406 Painter J., & Slutsker, L. (2007). Listeriosis in humans. In: E. T. Ryser & E. H. Marth,
407 editor. *Listeria, Listeriosis and Food Safety*, 3rd ed. Taylor and Francis, Boca Raton, FL.
- 408 Pérez-Rodríguez, F., Valero, A., Todd, E. C. D., Carrasco, E., García-Gimeno, R. M. &
409 Zurera, G. 2007. Modeling transfer of *Escherichia coli* O157:H7 and *Staphylococcus*
410 *aureus* during slicing of a cooked meat product. *Meat Science*, 76, 692–699.
- 411 Ramos, F., Takaishi, Y., Shirotori, M., Kawaguchi, Y., Tsuchiya, K., Shibata, H., Higuti,
412 T., Tadokoro, T., & Taeuchi, M. (2006). Antibacterial and antioxidant activities of
413 quercetin oxidation products from yellow onion (*Allium cepa*) skin. *Journal of*
414 *Agricultural. Food Chemistry*, 54, 3551–3557.
- 415 Sheen, S., & Hwang, C.A. (2010). Mathematical modeling the cross-contamination of
416 *Escherichia coli* O157:H7 on the surface of ready-to-eat meat product while slicing.
417 *Food Microbiology*, 27, 37–43.
- 418 Sheen, S., & Hwang, C.A. (2011). Modeling the surface cross-contamination of
419 *Salmonella spp.* on ready-to-eat meat via slicing operation. *Food and Nutritional*.
420 *Sciences*, 2, 916–924.
- 421 Sizmur, K., & Walker, C.W. (1988). *Listeria* in prepacked salads. *The Lancet*, 331, 1167

- 422 Stepanović, S., Cirković, I., Ranin, L., & Svabić-Vlahović, M. (2004). Biofilm formation
423 by *Salmonella* spp. and *Listeria monocytogenes* on plastic surface. *Letters in Applied*
424 *Microbiology*, 38, 428–432.
- 425 Suslow, T. (1997). Postharvest chlorination: basic properties and key points for effective
426 distribution. Oakland: University of California, Division of Agriculture and Natural
427 Resources. Publication 8003.
- 428 United States, California Department of Public Health, ERU Activity Summary Report
429 Gills Onions 2012. Gills Onions_Onions_ListeriaMono. Available at:
430 <http://www.cdph.ca.gov/HealthInfo/Documents/fdbEIRGO2012.pdf> Accessed 1 February
431 2014
- 432 Vorst, K.L, Todd, E.C.D., & Ryser, E.T. (2004). Improved quantitative recovery of
433 *Listeria monocytogenes* from stainless steel surfaces using a one-ply composite tissue.
434 *Journal of Food Protection*, 67, 2212–2217.
- 435 Vorst, K.L, Todd, E.C.D., & Ryser, E.T. (2006). Transfer of *Listeria monocytogenes*
436 during mechanical slicing of turkey breast, bologna, and salami. *Journal of Food*
437 *Protection*, 69, 619–626.
- 438 Wang, H.Q., & Ryser, E.T. (2014). Quantitative Transfer and Sanitizer Inactivation of
439 *Salmonella* during Simulated Commercial Dicing of Tomatoes. *Annual Meeting of*
440 *Institute of Food Technologists*. Available at: [http://www.ift.org/Meetings-and-](http://www.ift.org/Meetings-and-Events/Past-Meeting-)
441 [Events/Past-Meeting-](http://www.ift.org/Meetings-and-Events/Past-Meeting-)

[Resources/Technical%20Abstract%20Search%20Details.aspx?id=60926](#) Accessed 1

January 2016.

Ye, C.-L., Dai, D.-H., & Hu, W.-L. (2013). Antimicrobial and antioxidant activities of the essential oil from onion (*Allium cepa* L.). *Food Control*, 30, 48–53.

Zeng, W., Vorst, K., Brown, W., Marks, B. P., Jeong, S., Pérez-Rodríguez, F., & Ryser, E. T. (2014). Growth of *Escherichia coli* O157:H7 and *Listeria monocytogenes* in packaged fresh-cut romaine mix at fluctuating temperatures during commercial transport, retail storage, and display. *Journal of Food Protection*, 77, 197–206.

464 Table 1: Populations (log CFU/onion) of *Listeria* on whole peeled onions during 7 d of
 465 storage at 4°C.

466
 467

<i>Listeria</i> strain	Day 0	Day 1	Day 2	Day 5	Day 7
Avirulent	6.5 ± 0.1^a	6.3 ± 0.1^a	5.9 ± 0.1^a	5.6 ± 0.2^a	5.2 ± 0.3^b
Virulent	6.1 ± 0.1^a	6.1 ± 0.2^a	5.7 ± 0.3^a	6.1 ± 0.1^a	5.6 ± 0.2^a

468

469 Populations within each strain were analyzed for changes during storage; values with
 470 different letters are significantly ($P < 0.05$) different.

471 Table 2. Model parameters A and B for *L. monocytogenes* transfer from one inoculated
 472 onion to 20 uninoculated onions during slicing.

Inoculated onion level (log CFU/onion)	A (95% CI) ^a	B (95% CI)	NRMSE ^b (log CFU/onion)
5.9	3.05 (2.40, 3.70)	-29.24 (-49.73, -8.76)	0.26
6.8	3.33 (2.82, 3.83)	-27.72 (-39.66, -15.78)	0.19
8.6	6.13 (5.51, 6.76)	-21.28 (-26.32, -16.25)	0.18

473 ^aCoefficient value (95% confidence interval).

474 ^bNormalized root mean squared error for the transfer model.

475

Figure Legends

Figure 1. Onion hand slicer: A) Pusher plates, B) Blades

Figure 2. Comparison of the resulting total *Listeria* population (log CFU/onion) transferred after calculating the total *Listeria* population per onion based on the first, fourth, and last onion slice as the average for the total onion. Statistical significance ($P < 0.05$) denoted by different letter for bars at same sequentially sliced onion.

Figure 3. Mean *Listeria* populations (log CFU/100 cm²) on the pusher plates and blades after slicing one inoculated followed by 20 uninoculated onions. Data represent means $\pm$ SE ($n = 3$). Columns with asterisks are significantly different ($P < 0.05$) from the corresponding component.

Figure 4. Predicted *L. monocytogenes* transfer from one inoculated onion (A: 5.9 log CUF/onion; B: 6.8 log CFU/onion; C: 8.6 log CFU/onion) to 20 uninoculated onions. ypred is the line of prediction; yobs is the observed line; CB is the confidence band for the line of prediction.

Figure 5. Correlation between predicted and observed values for all three inoculation levels.

Figure 1

Figure 2

Figure 3

508
509
510
511
512

Figure 4

A)

B)

C)

581

Figure 5

582