

Recommendations for Handling Fresh-cut Leafy Green Salads by Consumers and Retail Foodservice Operators

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SUMMARY

A panel of scientists with expertise in microbial safety of fresh produce was convened to review recent research and re-evaluate guidelines for foodservice and restaurant operators, regulatory agencies with oversight over food facilities, and consumers for handling prewashed bagged salads. The guidelines developed by the panel, together with materials reviewed by the panel to develop the guidelines, are presented. The background materials reviewed include published research and recent recommendations made by other authoritative sources. The panel concluded that leafy green salad in sealed bags labeled “washed” or “ready-to-eat” that are produced in a facility inspected by a regulatory authority and operated under cGMPs, does not need additional washing at the time of use unless specifically directed on the label. The panel also advised that additional washing of ready-to-eat green salads is not likely to enhance safety. The risk of cross contamination from food handlers and food contact surfaces used during washing may outweigh any safety benefit that further washing may confer.

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INTRODUCTION

Fresh-cut (minimally processed) fruit and vegetable sales have grown to approximately \$15 billion per year in the North American foodservice and retail market and account for nearly 15% of all produce sales. The largest portion of US fresh-cut produce sales at retail are fresh-cut salads, with sales of \$2.7 billion per annum (24). While the incidence of foodborne illness associated with fresh-cut salads is very low relative to the quantity consumed, the increased use of these products has been accompanied by an increase in reported outbreaks associated with their consumption. Since 1995, FDA records indicate that 22 US outbreaks of foodborne illness caused by *Escherichia coli* O157:H7 have been associated with consumption of fresh or fresh-cut lettuce and two with pre-washed spinach (9). In 2006, a large *E. coli* O157:H7 outbreak associated with pre-washed spinach affected over 200 people in more than 20 states (10). This outbreak was followed by two restaurant-associated outbreaks linked to consumption of pre-washed lettuce. An outbreak of *E. coli* O157:H7 in 2005, in Minnesota, was epidemiologically associated with pre-washed bagged salad products containing romaine lettuce (7). Similar outbreaks in 2003 were associated with bagged pre-washed spinach and romaine-iceberg mix (5, 6). An increase in the incidence of hepatitis A in Los Angeles County between August and December 2005 led to an epidemiological study of one cluster of illnesses that implicated two food products, one of which was a leafy green salad (18). Following these outbreaks, the question of possible recommendations for consumers was posed by local regulatory authorities. Specifically, it was proposed that consumers and foodservice operators be advised to re-wash bagged, pre-washed salad greens prior to use. To answer these questions, a panel of food safety experts with particular expertise in produce safety was convened to review recently published research and current recommendations on use of packaged leafy green salads. The panel then met to produce guidelines for foodservice operators and for consumers.

The issue

Does washing of ready-to-eat fresh-cut produce immediately before consumption at retail, restaurant or by consumers significantly enhance, reduce or have no effect on the risk of foodborne illness?

Survival and growth of human pathogens on leafy vegetables and internalization of cells

Studies on survival and growth of pathogens on lettuce and parsley have shown that *Shigella sonnei* and *E. coli* O157:H7 will decrease in numbers when the produce is stored at 4–5°C/39–41°F but increase at 12°C/54°F (*E. coli* O157:H7) and 21°C/70°F (both pathogens) (1, 28). Seo and Frank (20) inoculated lettuce by immersion in a suspension of *E. coli* O157:H7 overnight at 7°C/45°F, after which it was rinsed with sterile distilled water and then treated with a 20 ppm chlorine solution. In a separate experiment, lettuce leaves were first immersed in a suspension of *Pseudomonas fluorescens* for 48 h at 16°C/61°F to allow biofilm formation. The leaves were then rinsed with sterile water and transferred to a suspension of *E. coli* O157:H7 for 24 h at 7°C/45°F. Examination of inoculated lettuce leaf surfaces by confocal scanning laser microscopy showed that *Pseudomonas* (predominant psychrotrophic spoilage organism) adhered to and grew mainly on the intact leaf surface, whereas *E. coli* O157:H7 was entrapped 20 to 100 µm below the surface in stomata and cut edges. Many live *E. coli* O157:H7 cells were found in stomata and on cut edges following the chlorine treatment. This indicates the probability that subsequent washing probably will not be effective in removing the cells. Takeuchi et al. (23) allowed attachment of cells of *E. coli* O157:H7, *Listeria monocytogenes*, *Salmonella* Typhimurium, and *P. fluorescens* to lettuce leaves for 18 h at 4°C/39°F. The cut edges were physically separated from the remainder of the leaf section, and populations were enumerated on appropriate selective media. In addition, the inoculated lettuce sections were examined by confocal scanning laser microscopy. Results confirmed the preferential attachment of *E. coli* O157:H7 to cut surfaces, and showed that *L. monocytogenes* has an even greater preference for cut surfaces, whereas *S. Typhimurium* attached equally to both sites. *L. monocytogenes* also has been shown to grow on lettuce.

Effect of wash procedures on subsequent growth or survival during storage

Delaquis et al. (11) inoculated cut iceberg lettuce with *E. coli* O157:H7 and

L. monocytogenes before and after washing for 3 min in cold (4°C/39°F) and warm (47°C/117°F) water containing 100 ppm total chlorine, then stored the product at 1 and 10°C/50°F under aerobic conditions. Populations of *E. coli* O157:H7 declined over 14 days storage at 1°C/34°F under both washing conditions as well as at 10°C/50°F when washed in cold chlorine solution (current industry practice). Populations increased when stored at 10°C/50°F after a warm chlorine solution wash. However, this is not the procedure currently used in commercial operations. Similar results were obtained with *L. monocytogenes*, which showed about a 1 log CFU/g increase in the inoculated control when stored at 10°C/50°F but a 2 log CFU/g increase when the lettuce was washed with warm chlorine solution. Li et al. (16) also studied the survival and growth of *E. coli* O157:H7 on lettuce treated with 20 ppm chlorine at either 20 or 50°C/68 or 122°F then stored at 5°C/41°F for 18 days or at 15°C/59°F for 7 days. Populations declined throughout storage at 5°C/41°F but increased by 2.3 to 3.2 log CFU/g within 2 days at 15°C/59°F, and then continued to increase at a slower rate through the 7 days of storage at that temperature.

Home or foodservice washing procedures

Vijayakumar and Wolf-Hall (26) evaluated “household sanitizers” for their effectiveness in reducing levels of inoculated *E. coli* and naturally present aerobic mesophilic bacteria on iceberg lettuce. Treatments tested were diluted solutions of apple cider vinegar, 5% (0.3% acetic acid); household bleach, 4% (180 ppm available chlorine); lemon juice, 13% (0.6% citric acid); and white vinegar, diluted 35:65 with water (1.9% acetic acid). The white vinegar solution, used at 21°C/70°F for 10 min without agitation, or 5 min with agitation, produced a 5.4 log CFU/g reduction in *E. coli*, compared to a 0.9 log CFU/g reduction achieved with distilled water at the same temperature. However, sensory evaluation of the lettuce treated with white vinegar showed that it was significantly less acceptable than samples treated with the other sanitizers. Lemon juice (at 4°C/39°F) and cider vinegar (at 21°C/68°F) gave reductions of 2.1 and 2.7 log CFU/g, respectively, compared

to 0.9 log CFU/g for distilled water. The bleach solution gave a reduction of 1.6 log CFU/g when used at 4°C/39°F with agitation for 10 min.

Kilonzo-Nthenge, Chen, and Godwin (13) evaluated home washing methods for reducing surface contamination of lettuce with *L. monocytogenes*. Washing with running tap water for 15 s achieved a 1.4 log CFU/g reduction, compared to the following treatments: 2 min soak in tap water followed by 15 s rinse (1.8 log CFU/g reduction); 2 min soak in vinegar (5%) followed by 15 s rinse in water (1.9 log CFU/g reduction); 2 min soak in Veggie Wash (2.0 oz/gal of water) followed by 15 s rinse in water (1.7 log CFU/g reduction).

Several researchers have shown that washing lettuce with chlorine solutions (20 to 200 mg/l) reduces the microbial load (either naturally occurring microflora or inoculated pathogen) more than washing with water. However, the difference is relatively small, and neither treatment eliminates pathogens or spoilage bacteria. For example, Lang, Harris and Beuchat (15) obtained average reductions of *E. coli* O157:H7 on lettuce of 0.6 log CFU/ml with water and 1.4 log CFU/ml with chlorine (200 ppm) when the lettuce was submerged with agitation for 5 min. An inoculated sample that contained 5.1 log CFU before treatment contained 4.6 log CFU after washing with water and 3.7 log CFU after treatment with chlorine. Weissinger, Chantarapanont, and Beuchat (27) inoculated *Salmonella* Baildon onto shredded lettuce at low (0.6 log CFU/g) and high (3.6 log CFU/g) level and treated the inoculated lettuce with cold (4°C/39°F) sodium hypochlorite (NaClO) solution (120 and 200 ppm) immediately after inoculation for 40 s. The test organism was recovered from all samples by enrichment, and populations on the lettuce treated with the high inoculum level was found to be reduced by 1.1 log CFU/g with 120 ppm free chlorine and 1.1 log CFU/g with 200 ppm free chlorine. Washing with cold deionized water (control) reduced the population by 0.3 log CFU/g. Kondo, Murata, and Isshiki (14) inoculated iceberg lettuce with *Staphylococcus aureus*, *E. coli* O157:H7, and *S. Typhimurium* DT104 by immersing leaves in cell suspensions for 5 min or 1 h. In addition, some leaves inoculated

for 1 h were wrapped in plastic film and stored at 4°C/39°F for 2 days. Inoculated leaves were washed five times with 0.85% NaCl. Washing was most effective (2.9% residual cells for *E. coli* O157:H7) on leaves inoculated for 5 min and least effective (13.6% residual cells for *E. coli* O157:H7) when 2 days storage occurred before washing. Inoculated leaves were immersed in treatment solutions for 10 min at room temperature or for 1 min at 50°C/122°F, and then cooled in 0.85% NaCl solution at 4°C/39°F for 30 s, followed by three washes in 0.85% NaCl solution. Treatment solutions included fumaric acid (5 mM and 50 mM), NaClO (200 ppm, pH 6.0), and distilled water. For leaves inoculated and held for 1 h and stored for 2 days, treatment with NaClO reduced populations of *E. coli* O157:H7 to 6.4% of the pretreatment cell population, compared with 17.8% residual cells when treatment was with distilled water. Treatment with 50 mM fumaric acid at room temperature was not significantly more effective than 200 ppm NaClO, leaving 4.0% residual cells.

Singh et al. (21) used aqueous chlorine dioxide (10 mg/L for 10 min), ozonated water (10 mg/L for 10 min), and thyme oil (0.1% for 5 min) to wash shredded romaine lettuce inoculated with *E. coli* O157:H7. When sprinkle-inoculated lettuce samples were held for 24 h at 5°C/41°F before washing, log reductions achieved by washing were 1.6, 1.5, and 1.9 log CFU/ml (respectively), compared to a log reduction of 0.9 log CFU/ml by sterile deionized water wash. A multistage washing treatment improved efficacy somewhat. Using treatment times of 5 min for de-ionized water, aqueous chlorine dioxide, ozonated water, and 2 min for thyme oil, log reductions after the first wash were 0.5, 1.2, 1.1 and 1.5 log CFU/ml, respectively. After the second wash, total log reductions were 0.6, 1.7, 1.6, and 2.2 log CFU/ml, respectively. A third wash did not result in significant improvement. The authors speculate that this may be because the remaining microorganisms have penetrated the cut surfaces and stomata and are not accessible to the sanitizers.

Smith et al. (22) evaluated the effect of a commercial peroxyacetic acid produce wash on the natural microflora in a food service setting and found that when the initial contamination was greater than 100 CFU/g, use of the commercial wash

resulted in about a 1 log CFU/g greater reduction than water alone. Sapers (19) reviewed washing treatments for home or foodservice use and found that use of alternatives to chlorine for produce washes may avoid disadvantages of chlorine such as formation of toxic reaction products, but differences in antimicrobial efficacy are small. He also observed that “safe and uniform application may be problematic without the controls available for large-scale applications.”

Escudero et al. (12) evaluated the effects of chlorine and chlorine combined with surface active agents and organic acids on *Yersinia enterocolitica* on fresh lettuce. The combination of 100 ppm chlorine and 0.5% lactic acid (pH 2.28, 22°C/72°F, 1 min treatment) produced a reduction of more than 6 log CFU/g of the target organism. The authors did not address potential hazards to workers of using this solution in a foodservice setting.

Studies on washing produce and general food handling by consumers and foodservice establishments

Li-Cohen and Bruhn in 2002 (17) studied consumer handling of fresh produce from the time of purchase to the plate via a national mail survey of 624 respondents. Six percent of respondents replied that they never or seldom wash fresh produce before consumption. Approximately 53% of all respondents did not wash their hands before handling fresh produce; 56% report that they always wash the sink before handling fresh produce; and of those that wash the sink, 11% use water only. Ninety-seven percent of all respondents reported that they always washed food preparation surfaces after contact with raw meat products. However, washing was inefficient, since 5% of respondents only dry wipe, and 24% of respondents wash these potentially contaminated food preparation surfaces with water only. This survey also found that many respondents did not separate produce from raw meat, poultry or fish in their refrigerators. This data indicates that the possibility of re-contaminating a previously washed product in the consumer's kitchen is fairly high.

In 2003, the US Food and Drug Administration (FDA) collected data

TABLE 1. Percent of facilities out of compliance with assessment criteria based on 1997 Food Code

Type of facility	Contaminated equipment/protection from contamination ¹	Surfaces/utensils cleaned and sanitized	Poor personal hygiene ²	Proper hand-washing
Fast food restaurant	21.9	50.9	31.2	53.8
Full serve restaurant	37.3	56.6	41.7	72.7
Retail stores/produce	20.5	44.4	22.3	33.3

¹Contaminated equipment/protection from contamination is a multi-factor category that includes surfaces/utensils cleaned and sanitized.

²Poor personal hygiene is a multi-factor category that includes proper hand washing.

Source: USHHS-FDA (25).

via site-visits to over 900 establishments representing nine distinct facility types including restaurants, institutional food-service operations and retail food stores (25). Direct observations of produce handling practices were supplemented with information gained from discussions with management and food workers and were used to document the establishments' compliance status based on provisions in the 1997 Model FDA Food Code. Failure to control product holding temperatures, poor personal hygiene, use of contaminated equipment/failure to protect food handling equipment from contamination and risk of potential chemical contamination were the risk factors found to be most often out of compliance with the 1997 FDA Model Food Code. The percentages of "out of compliance" observations for each of these risk factors were found to be: improper holding/time temperature (49.3%), poor personal hygiene (22.3%), contaminated equipment (20.5%) and chemical contamination (13.5%). Specifically, for the improper holding/time and temperature risk factor, it was found that maintaining cold holding temperatures at or below 5°C/41°F for produce items that are classified as potentially hazardous foods (PHF) did not occur in 70.2% of the observed situations. Holding PHFs at or below 5°C/41°F is critical to preventing the potential growth of human

pathogens, which may rapidly proliferate on inadequately refrigerated PHFs. Date marking of refrigerated ready-to-eat, PHFs is also an important component of any food safety system, and it is designed to promote proper food rotation and limit the growth of *L. monocytogenes* during cold storage. However, appropriate date marking of ready-to eat, PHF produce items made on-site did not occur in 34.0% of the observations.

The personal hygiene risk factors associated with produce that are most in need of attention at retail and foodservice operations include adequate, available and accessible handwashing facilities. These personal hygiene risk factors were found by the survey to be not in compliance with the 1997 FDA Model Food Code 33.3%, 26.2%, and 20.6% of the time, respectively. Hands are a very common vehicle for the transfer of human pathogens to food products, and food handlers' hands may become contaminated when they engage in activities such as handling raw meat products, using the restroom, coughing or handling soiled tableware.

Food safety procedures for cleaning and sanitizing food contact surfaces and utensils for handling produce were found to be not in compliance with the 1997 FDA Model Food Code in 44.4% of the observations in this study. Proper cleaning and sanitization of food contact

surfaces is essential to preventing cross contamination. Results for selected types of facilities and selected assessment criteria are shown in Table 1.

Many fresh-cut fruit and vegetable products are "ready-to-eat" food products that require no further preparation. These products are no different from any other ready-to-eat food product. The fresh-cut produce industry was established to provide convenient ready-to-eat foods to food service establishments and the consumer in a form that reduced the risk of food product contamination by placing preparation of fresh-cut produce in a controlled food manufacturing environment.

Current recommendations regarding re-washing of fresh-cut produce

Advice to consumers contained in current publications such as the "Fight BAC" materials from the Partnership for Food Safety Education (2), the 2005 report of the Produce for Better Health Foundation (8), California Department of Health Services document (3, 4) and the 2005 Dietary Guidelines Advisory Committee Report may be summarized as follows.

1. Consumers should first read the label to determine if the product is ready-to-eat. Packaged salad mixes labeled "ready-to-eat,"

“washed,” or “triple-washed” need not be washed again by the user if they are kept refrigerated and used by the “use-by” date.

2. If desired, pre-washed packaged salads may be rewashed without harming product quality. Since improper handling in the home or restaurant during preparation is a leading cause of foodborne illness, it is important to protect the product from cross contamination from raw foods, contaminated equipment, or inadequately washed hands.
3. Antibacterial agents may be used on raw produce if they are approved for food contact and used according to directions. However, these products do not completely remove bacterial pathogens or disease-causing viruses.

After reviewing all of the above information, the panel drafted the following recommendations for (a) retail and food service operators and (b) consumers.

Recommendations to retail and food service operators regarding rewashing ready-to-eat lettuce/leafy green salads

1. Carefully read labels to determine whether a product is a raw agricultural commodity (e.g. hearts of Romaine) that should be washed before consumption or a ready-to-eat (RTE) food product (e.g. pre-washed lettuce/leafy green salad). If the product is not labeled “washed”, “triple washed” or “ready-to-eat”, the product needs to be washed before consumption.
2. If a RTE lettuce/leafy green salad is received in sealed bags labeled “washed”, “triple washed” or “ready-to-eat” from a facility inspected by a regulatory authority and operated under cGMPs, it does not need additional washing at the time of use unless specifically directed on the label.
3. Additional washing of RTE lettuce/leafy green salads is not likely to enhance safety.
 - Current research suggests that if harmful microorganisms are present after commercial washing treatments, they are

likely to resist removal or inactivation by further washing.

- If appropriate practices are not followed, there is a risk of cross contamination from food handlers and food-contact surfaces such as sinks, colanders and pans used during washing. This may outweigh any safety benefit that further washing may confer in bagged, pre-washed, RTE salads.
4. If the end-user chooses to wash the RTE lettuce/leafy green salads before use:
 - Wash hands thoroughly with soap and warm water before handling RTE lettuce/leafy green salads. Rewash hands as necessary.
 - Clean and sanitize the sink, colander, and any equipment or utensils that will contact the product.
 - Use cold running water to wash RTE lettuce/leafy green salads to reduce the potential for cross contamination.
 - If product is soaked, reduce the potential for cross contamination by using a registered (US EPA, US FDA, state and local jurisdictions) and appropriately labeled antimicrobial products as per manufacturer’s directions. Antimicrobial concentrations should be monitored to ensure appropriate concentrations are maintained during soaking or washing. Household bleach is generally not acceptable for this application.
 5. Additional Considerations
 - Wash hands thoroughly for 20 s with soap and warm water before handling RTE lettuce/leafy green salads. Rewash hands as necessary.
 - Use a barrier such as clean, intact gloves and/or an appropriate clean and sanitized utensil (changed with sufficient frequency to prevent cross contamination) to handle or dispense fresh-cut

lettuce/leafy green salads. This does not alleviate the need for proper hand-washing, so hands should be washed for 20 s before gloves are used.

- RTE lettuce/leaf green salads should be shipped, stored and displayed under refrigeration.
- RTE lettuce/leafy green salad shipping containers may become contaminated during transport and storage. Therefore:
 - Inspect product cartons or bags upon receipt and reject any product that shows evidence of mishandling or tampering (e.g., dirty, wet, open or crushed boxes or bags, etc.).
 - Ensure that storage practices do not subject the product to potential cross contamination (e.g., do not store raw meats above RTE lettuce/leafy green salad cartons or bags).
- Discard the product if it appears spoiled or has exceeded its labeled use-by date.

Recommendations to consumers regarding washing ready-to-eat lettuce/leafy green salads

1. Carefully read labels to determine whether a product is one that should be washed before consumption (e.g. hearts of Romaine) or is a ready-to-eat (RTE) food product (e.g. pre-washed lettuce/leafy green salad). If the product is not labeled “washed”, “triple washed” or “ready-to-eat”, the product needs to be washed before consumption.
2. If a RTE lettuce/leafy green salad is received in either a sealed bag or rigid plastic containers labeled “washed”, “triple washed” or “ready-to-eat” it does not need additional washing before you eat it unless specifically directed on the label.
3. Additional washing treatments are not likely to enhance the safety of RTE lettuce/leafy green salads.

- Harmful bacteria are rarely found on RTE lettuce/leafy green salads.
 - In the unlikely event that harmful bacteria are present on a RTE lettuce/leafy greens salad after commercial washing, they are likely to resist removal or inactivation by further washing.
 - If the following instructions for washing are not followed, there is a risk of cross contamination from hands sinks, colanders, pans and utensils that may be used during washing. This may outweigh any safety benefit that further washing may provide to pre-washed, ready-to-eat salads.
4. If you choose to wash the RTE lettuce/leafy green salads before use, you should:
- Wash your hands thoroughly with soap and warm water for at least 20 s before handling RTE lettuce/leafy green salads. Rewash hands as necessary.
 - Clean with hot soapy water, the sink, colander, salad spinner and any utensils that will contact the lettuce/leafy greens salad.
 - Use cold running water to wash RTE lettuce/leafy green salads to reduce the potential for cross contamination.
 - Dry RTE lettuce/leafy green salad with a clean salad spinner or paper towel not previously used for another purpose.
 - Never use detergent or bleach to wash fresh vegetables. These products are not intended for consumption.
5. Follow FightBAC![™] procedures to protect RTE lettuce/leafy green salads from contamination.

Check

- Check to be sure that RTE lettuce/leafy green salads you buy are not bruised or damaged.
- Check that RTE lettuce/leafy green salads are refrigerated at the store before buying.

Do not buy RTE lettuce/leafy green salads that are not refrigerated.

Clean

- Wash hands with warm water and soap for at least 20 s before handling RTE lettuce/leafy green salads.
- Use hot water and soap to clean all surfaces and utensils, including counter tops and salad spinners, that will touch RTE lettuce/leafy green salads.
- Use a clean utensil to serve RTE lettuce/leafy green salads.

Separate

- When shopping, be sure fresh produce is separated from household chemicals and raw foods such as meat, poultry and seafood in your cart and in bags at checkout.
- Keep RTE lettuce/leafy green salads separate and protect from contact with raw meat, poultry or seafood or their juices in your refrigerator. Do not allow raw meat, poultry or seafood juices to drip onto RTE lettuce/leafy green salads.

Chill

- Store RTE lettuce/leafy green salads in the refrigerator.

Throw Away

- Throw away RTE lettuce/leafy green salad if it has touched raw meat, poultry or seafood.
- Discard the product if it appears spoiled or has exceeded its labeled use-by date.

More information regarding safe produce handling may be found on the FightBAC![™] Web site at: <http://portal.fightbac.org/pfse/toolstheycanuse/phec/>.

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