
# Summary Report: Processed Avocado and Guacamole

FY 2017 – 2019 Microbiological Sampling Assignment

**Office of Compliance**
**Center for Food Safety and Applied Nutrition**

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## AT-A-GLANCE

- **The FDA collected and tested 887 samples of processed avocado and guacamole, both domestic and imported product, for *Salmonella* spp. and *Listeria monocytogenes* from November 2017 to September 2019.**
- **The agency detected *Salmonella* spp. in two samples, which were later determined to be distinct samples of the same brand of domestically manufactured guacamole from different lots. Neither of these samples had received high-pressure processing (HPP) treatment, which is a validated lethality step.**
- **The FDA detected *Listeria monocytogenes* in 15 samples. Of those, eight had not received HPP treatment. The agency could not ascertain the HPP-treatment status of the other seven samples.**
- **The findings of this assignment affirm that pathogens may be present in processed avocado and/or guacamole and appear to align with other research that shows HPP is effective at neutralizing pathogenic microorganisms.**
- **The assignment findings also underscore the need for processors and others in the processed avocado and guacamole supply chain to comply with the FDA's Preventive Controls for Human Food Rule and for importers of these foods to comply with the FDA's Foreign Supplier Verification Programs Rule.**

## EXECUTIVE SUMMARY

The U.S. Food and Drug Administration collected and tested processed avocado, the main ingredient in guacamole, and finished guacamole as part of the agency's proactive and preventive approach to deploying its sampling resources with the ultimate goal of preventing contaminated food from reaching consumers.

### Assignment Overview

The assignment began in November 2017 and ended in September 2019. In total, the FDA collected and tested 887 samples of processed avocado and guacamole (domestic and imported product) for *Salmonella* spp. and *Listeria monocytogenes*. This total is smaller than the initial number of samples the agency set out to collect and test because the agency encountered factors that twice required a reduction of the collection target, as explained in the Sample Collection section of this report (page 6).

As to the design of the assignment, the FDA directed its field staff not to collect products that had undergone high-pressure processing (HPP) or products intended for HPP. HPP is a "kill step" validated to eliminate pathogenic microorganisms in food, and it is often used in the manufacture of processed avocado and guacamole. In seeking to exclude from the assignment products that had been HPP-treated, the FDA's intent was to focus on products that posed the greatest risk to consumers.

The agency learned during its evaluation of the test results that some of the products collected had received HPP treatment but were not labeled as such. FDA staff worked retrospectively with industry to identify the HPP-treatment status of the samples collected but could not determine the status of a number of samples. Those samples were designated as "could not ascertain" for purposes of the data analysis.

### Findings and Follow-up Actions

The FDA detected *Salmonella* spp. in two samples which were later determined to be distinct samples of the same brand of domestically manufactured guacamole from different lots. Neither sample had received HPP treatment. In addition, the agency detected *Listeria monocytogenes* in 15 samples from nine different firms. Of those 15 samples, eight had not been HPP treated. The HPP-treatment status of the other seven samples could not be ascertained.

When the FDA detected a pathogen in a domestic sample, agency personnel worked with the company that owned or distributed the affected product to conduct a voluntary recall in all cases in which product was available, or likely to still be available, to consumers. The FDA also conducted one follow-up inspection of a domestic facility, and state officials in Florida likewise conducted one domestic inspection. As to the imported samples, the agency refused to admit lots associated with the positives and placed the responsible companies on import alert. In all, the agency placed two firms on import alert. In addition, the agency conducted whole genome sequencing (WGS) analysis on the positives but was unable to determine whether processed avocado or guacamole were the food vehicle associated with any known human illnesses.

In addition to affirming that *Salmonella* spp. and *Listeria monocytogenes* may be present in processed avocado and/or guacamole, the assignment data show that the estimated prevalence of these pathogens in the non-HPP-treated samples was higher than in the HPP-treated samples. This finding appears to support other research that shows HPP is effective at neutralizing pathogenic microorganisms,[1] even as this assignment was not designed to compare possible differences based on HPP-treatment status. The findings also underscore the need for processors and others in the processed avocado and guacamole supply chain to comply with the FDA's Preventive Controls for Human Food Rule[2] and for importers of these foods to comply with the FDA's Foreign Supplier Verification Programs Rule.[3]

## BACKGROUND

The FDA Food Safety Modernization Act (FSMA) amended the Federal Food, Drug, and Cosmetic Act (FD&C Act) to provide the agency with additional authority to better prevent food safety problems before they occur. To develop better prevention-based systems, the FDA needs data and other information to help identify hazards that must be addressed and minimized. That is why sampling is an important part of the FDA's preventive approach to food safety and why starting in 2013 the FDA developed a new sampling model to identify patterns that may help prevent contamination by disease-causing microorganisms.

Avocados and avocado products became a heightened focus of the FDA in 2013, following a study published that same year by the Centers for Disease Control and Prevention (CDC). The CDC study found that *Salmonella* spp. contamination of salsa or guacamole had resulted in 26 outbreaks and 1,872 illnesses during the 35-year period examined.[4] During a portion of the same interval, from 2001 to 2013, the FDA collected and tested 429 avocado samples for microbial hazards. Of those 429 samples (which were mostly imported, processed product), 77 of them (or 18%) were found to contain a human pathogen, warranting regulatory follow-up, including recalls and/or the addition of firms to import alerts. The CDC study findings, coupled with FDA's data, indicated a need to gain reliable estimates of the prevalence of *Salmonella* spp. and *Listeria monocytogenes* in avocados and avocado products and to identify common factors among the positive samples, if possible, to help protect consumers.

The FDA sought to address the need (i.e., its data gap) in 2014 by implementing a large-scale sampling assignment focused on whole fresh avocados, which represent the start of the supply chain for guacamole and other products made from avocados. Completed in 2016, the whole fresh avocado assignment estimated that the prevalence of *Listeria monocytogenes* on the fruit's exterior was 17.7%, based on the test results obtained three months into the assignment, prior to an updating of the test method to focus on the edible portion of the fruit. The test method also was updated in view of the fact that no known outbreaks or individual illnesses had been linked

[1] Huang, H.; Wu, S.; Lu, J.; Shyu, Y.; and Wang, C. (2017). Current status and future trends of high-pressure processing in food industry. *Food Control*, Vol. 72, Part A, Feb. 2017, p. 1-8.
[2] The Preventive Controls for Human Food Rule requires food facilities to have a food safety plan in place that includes an analysis of hazards and risk-based preventive controls to minimize or prevent the identified hazards.
[3] The Foreign Supplier Verification Programs Rule requires that importers perform certain risk-based activities to verify that food imported into the U.S. has been produced in a manner that meets applicable U.S. safety standards.
[4] Kendall, M., Mody, R., Mahon, B., Doyle, M., Herman, K. & Tauxe, R. (2013). Emergence of salsa and guacamole as frequent vehicles of foodborne disease outbreaks in the U.S., 1973–2008. *Foodborne Pathogens and Disease*.

to the presence of the pathogen on the fruit's exterior at the time (August 2014). Additionally, having updated the test method, the FDA was able to better evaluate the public health concerns associated with the hazard-commodity pair. The whole fresh avocado assignment found the estimated prevalence of *Listeria monocytogenes* in the avocado pulp samples to be 0.2%. In addition, the assignment found the estimated prevalence of *Salmonella* spp. in the samples to be 0.7%. Upon considering these findings, the agency determined that further sampling was needed to better understand the extent to which these pathogens may be contaminating processed avocado and guacamole.

### Processed Avocado and Guacamole Production

Processed avocado – used to make guacamole, sandwich spreads, and some beverages, among other products – is made from whole fresh avocados. Processors receive shipments of the fruit, typically wash it in a machine with rotating brushes and chlorinated water, and hold it in refrigerated storage for a short time, initially at 5°C, to allow for even ripening. The processing begins with a sorting step during which unsuitable avocados are discarded, and the selected fruit is halved, deseeded, peeled and then placed in a mixer and blended to a soft, smooth preparation. When making guacamole, ingredients such as onion, tomato and jalapeños are commonly added, as well as erythorbic and ascorbic acids, to preserve color and freshness. The resulting product is extruded into vacuum-packed bags, in sizes either for restaurant customers or retail marketing.[5] Once packaged, the product may undergo HPP, during which it is immersed in a vessel filled with cold water and subjected to high levels of isostatic pressure, which kills any pathogens that may be present.

## OBJECTIVES

The objectives of the FDA's FY 2017-2019 processed avocado and guacamole sampling assignment were:

- To estimate the prevalence of *Salmonella* spp. and *Listeria monocytogenes* in processed avocado and guacamole;
- To determine if there were common factors associated with positive findings (such as by origin); and
- To take appropriate regulatory action in response to violations.

## SAMPLE COLLECTION

The FDA had originally planned to collect and test 1,600 samples (800 domestic and 800 of international origin), consistent with the design of the agency's other large-scale microbiological surveillance sampling assignments. However, in July 2018, the FDA adjusted its collection target to 1,200 samples (936 domestic and 264 of international origin) after initial sampling confirmed that a relatively small number of firms – particularly, domestic firms – produce and/or distribute

[5] *Avocado: Post-Harvest & Processing*. International Tropical Fruits Network. (2016, May 3). Retrieved April 20, 2021, from https://www.itfnet.org/v1/2016/05/avocado-post-harvest-processing/.

processed avocado. In adjusting the collection target, the FDA's intent was to avoid biasing the data by oversampling product from the same firms and to minimize the burden on industry. The agency also had learned that an increasing number of processors had begun to use HPP.

In March 2019, the FDA further reduced its collection target to account for a 35-day lapse in federal appropriations that began on December 22, 2018, and the associated impact on the workload of the agency field staff. Similar slight adjustments were made for the same reason to other food sampling assignments as well. Ultimately, the FDA collected and tested 887 processed avocado and guacamole samples (571 domestic; 316 of international origin) from November 2017 to September 2019.

The agency's field staff collected samples one at a time from both individual lots and multiple lots. When the collection site featured multiple lots, the field staff generally collected one sample from each lot. The FDA's approach, which avoided commingling samples from different lots, was designed to help the agency facilitate targeted removal of potentially adulterated product from the food supply.

The FDA collected processed avocado in the form of fresh cut, pureed, refrigerated and frozen product, as well as frozen avocado pulp with additives, and guacamole. The FDA did not collect whole avocados or any product from farms or growers. Each sample was made up of 10 subsamples. Each subsample was a sealed package or container of processed avocado or guacamole weighing a minimum of eight ounces. The agency divided the subsamples evenly for testing purposes, testing half for *Salmonella* spp. and half for *Listeria monocytogenes*. Collecting and testing samples composed of multiple subsamples is more reflective of actual conditions, and it increases the probability of detecting pathogens if present, given that microbial hazards may not be uniformly present. Accordingly, if one subsample tested positive for a target pathogen, the FDA regarded the entire sample as positive for the organism.

The FDA directed its field staff not to collect product that had been subject to HPP or product intended for HPP because the treatment, when properly carried out, is a validated lethality step. However, many of the products collected did not reference HPP on their labeling even though they had undergone the process. As a result, the FDA's dataset ultimately included an unknown number of samples subject to HPP. The FDA adjusted for the uncertainty of HPP treatment by querying industry, as described in Appendix A.

### Domestic Sample Collection

Agency field staff collected 571 domestic samples of processed avocado and guacamole, with most collected at retail (Table 1). Samples were collected in 33 states, Puerto Rico and the District of Columbia, with the largest number collected in California (295), followed by Texas (67), and Florida (42).

**Table 1: Domestic Sample Collection Sites**

| Collection Site | Domestic Samples Collected | Percentage of Domestic Samples* | Percentage of All Samples* |
|---|---|---|---|
| **Distribution Center/Warehouse** | 29 | 5% | 3.3% |
| **Processor** | 14 | 2.5% | 1.6% |
| **Retail** | 525 | 91.9% | 59.2% |
| **Unidentified** | 3 | 0.5% | 0.3% |
| **Total** | 571 | 100% | 64% |

* Numbers do not add up to the percentage totals due to rounding.

### Import Sample Collection

The field staff collected 316 samples of imported product. The FDA used two approaches to collect samples of imported product: collection in "import status" and domestic import (DI) sampling. "Import status" refers to samples collected at ports of entry or other locations where the product was being held prior to its release into domestic commerce. The FDA collected 110 samples at import-status locations, representing about 35% of the total collection of imported product sampled. In addition, 206 samples (about 65%) were collected as DI samples and counted toward the import sample total. DI samples are samples of international origin collected after being released into domestic commerce. They often are collected near the port of entry, usually at a warehouse, but may also be collected from retail stockrooms, prior to consumer handling. Unlike samples collected in import status, DI sampling allows for imported products to be released and sold domestically or to undergo processing. For purposes of this report, DI samples are included as import sample data because they originated outside the United States (Table 2).

**Table 2: Import Sample Collection Sites**

| Collection Site | Import Samples Collected | Percentage of Import Samples | Percentage of All Samples |
|---|---|---|---|
| **Port of Entry/Import** | 110 | 35% | 12% |
| **Domestic Import** | 206 | 65% | 23% |
| **Total** | 316 | 100% | 35% |

The FDA collected samples of imported product from five countries. The large majority originated in Mexico (291), followed by Peru (18), the Dominican Republic (4), Greece (2), and Guatemala (1).

### By Season

The agency collected samples year-round. Avocados are most commonly grown from February through September in the United States, and throughout the year in Mexico, allowing for year-round production of processed avocado and guacamole. The FDA collected most of its samples in the summer (287 samples), followed by the spring (281), the fall (177), and winter (142).

### By Guacamole vs. Processed Avocado

Of the 887 samples collected and tested, 737 were guacamole and 150 were processed avocado.

## PATHOGEN FINDINGS

This section reports the prevalences of *Salmonella* spp. and *Listeria monocytogenes* in the samples tested based on their designation as "not HPP treated," "HPP treated," or "could not ascertain." The test methods the FDA used are described in Appendix B: Test Methods.

The binning of the test data by the three categories noted above reduces each category's sample size in relation to the full dataset, and thus yields a relatively wide confidence interval for some of the bacterial prevalence estimates, as indicated in the tables below.

### *Salmonella* spp.

The FDA detected *Salmonella* spp. in two of the 322 samples of product not HPP treated, an estimated prevalence of 0.6% (Table 3). Serotyping found each organism to be *Salmonella* Muenchen. The FDA isolated the organisms from separate, distinct samples of the same brand of domestically manufactured guacamole.

The agency did not detect *Salmonella* spp. in the samples categorized as "HPP treated" or "could not ascertain."

**Table 3: *Salmonella* spp. Findings**

| Category of HPP Treatment | Samples Collected | Samples Positive | Estimated Prevalence | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|
| **Not HPP Treated** | 322 | 2 | 0.6% | 0.1% | 3.4% |
| **HPP Treated** | 362 | 0 | 0% | – | – |
| **Could Not Ascertain** | 203 | 0 | 0% | – | – |
| **Total** | 887 | 2 | 0.2% | 0.0% | 1.3% |

### *Listeria monocytogenes*

The FDA detected *Listeria monocytogenes* in 15 samples from nine firms. Of those 15 samples, eight were not HPP treated and seven were categorized as "could not ascertain." (Table 4).

**Table 4: *Listeria monocytogenes* Findings**

| Category of HPP Treatment | Samples Collected | Samples Positive | Estimated Prevalence | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|
| **Not HPP Treated** | 323 | 8 | 2.5% | 1.0% | 6.3% |
| **HPP Treated** | 361 | 0 | 0% | – | – |
| **Could Not Ascertain** | 203 | 7 | 3.5% | 1.1% | 10.1% |
| **Total** | 887 | 15 | 1.7% | 0.8% | 3.5% |

The FDA has reported the "could not ascertain" samples and associated confidence intervals in Tables 3 and 4 (above) for informational purposes. The FDA cautions against making inferences about the samples in this category given the uncertainty of the HPP treatment.

The follow-up actions that the FDA took in response to the positives are described in the Public Health Impact and Follow-Up Activities section of this report (page 13).

### By Guacamole vs. Processed Avocado: *Salmonella* spp.

Of the 737 guacamole samples, two that were not HPP treated tested positive for *Salmonella* spp. (Table 5).

**Table 5: *Salmonella* spp. in Guacamole**

| Category of HPP Treatment | Samples Collected | Samples Positive | Estimated Prevalence | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|
| **Not HPP Treated** | 296 | 2 | 0.7% | 0.1% | 3.7% |
| **HPP Treated** | 293 | 0 | 0% | – | – |
| **Could Not Ascertain** | 148 | 0 | 0% | – | – |
| **Total** | 737 | 2 | 0.3% | 0.1% | 1.5% |

Of the 150 processed avocado samples, none tested positive for *Salmonella* spp. (Table 6).

**Table 6: *Salmonella* spp. in Processed Avocado**

| Category of HPP Treatment | Samples Collected | Samples Positive | Estimated Prevalence | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|
| **Not HPP Treated** | 27 | 0 | 0% | – | – |
| **HPP Treated** | 68 | 0 | 0% | – | – |
| **Could Not Ascertain** | 55 | 0 | 0% | – | – |
| **Total** | 150 | 0 | 0% | – | – |

The FDA did not design its assignment to compare bacterial prevalence by guacamole versus processed avocado, and the differing sample sizes limit such evaluation. However, in considering the product-type totals (specifically, the estimated prevalence in each last row of Tables 5 and 6), the difference in the *Salmonella* spp. contamination rate in guacamole versus processed avocado was not statistically significant ($P$-value > .05), 0.3% and 0%, respectively.

### By Guacamole vs. Processed Avocado: *Listeria monocytogenes*

Of the 737 guacamole samples, seven that were not HPP treated and five categorized as "could not ascertain" tested positive for *Listeria monocytogenes* (Table 7).

**Table 7: *Listeria monocytogenes* in Guacamole**

| Category of HPP Treatment | Samples Collected | Samples Positive | Estimated Prevalence | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|
| **Not HPP Treated** | 296 | 7 | 2.4% | 0.8% | 6.6% |
| **HPP Treated** | 293 | 0 | 0% | – | – |
| **Could Not Ascertain** | 148 | 5 | 3.4% | 1.0% | 10.6% |
| **Total** | 737 | 12 | 1.6% | 0.7% | 3.6% |

Of the 150 processed avocado samples, one that was not HPP treated and two categorized as "could not ascertain" tested positive for *Listeria monocytogenes* (Table 8).

**Table 8: *Listeria monocytogenes* in Processed Avocado**

| Category of HPP Treatment | Samples Collected | Samples Positive | Estimated Prevalence | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|
| **Not HPP Treated** | 27 | 1 | 3.7% | 0.7% | 18.5% |
| **HPP Treated** | 68 | 0 | 0.0% | – | – |
| **Could Not Ascertain** | 55 | 2 | 3.6% | 1.0% | 12.4% |
| **Total** | 150 | 3 | 2.0% | 0.7% | 5.7% |

Again, the FDA did not design its assignment to compare bacterial prevalence by guacamole versus processed avocado. However, in considering the estimated prevalence in the last row of Tables 7 and 8, the difference in the *Listeria monocytogenes* contamination rate in guacamole versus processed avocado was not statistically significant (*P*-value > .05), 1.6% and 2.0%, respectively.

### By Origin

The FDA also calculated the bacterial prevalences by origin for informational purposes. The FDA estimated the prevalence of *Salmonella* spp. in domestically produced guacamole and processed avocado to be 0.4% and did not detect the pathogen in any of the import samples. Based on the test results, the agency estimated the prevalence of *Listeria monocytogenes* in domestically produced guacamole and processed avocado to be 2.3%, and in the import samples, to be 0.6%. The FDA cautions against comparing the estimated prevalences by origin because it is not known whether the domestic finished products were uniformly made using domestically grown avocados, as opposed to imported avocados, as the main ingredient. Additional data on origin can be found in Appendix D.

### By Season

The FDA did not detect a statistical difference by season in the contamination rate of either *Salmonella* spp. (*P*-value > .05), or *Listeria monocytogenes* (*P*-value > .05). This data is provided for informational purposes only, as the agency did not design its sample collection to compare bacterial prevalence by season. Additional data on seasonality can be found in Appendix D.

### By 'Repeat Violation' Firms (De-Identified), and Related Actions

For purposes of this subsection, 'repeat violation' firms are defined as physical locations where the agency detected one or more positive samples during each of two or more sample collections. Eleven of the 17 positive samples were associated with 'repeat violation' firms (Table 9).

**Table 9: 'Repeat Violation Firms' (De-Identified), and Related Actions**

| Firm ID | Firm Type | Firm Location | Sample Collection Date * | Pathogen | Action |
|---|---|---|---|---|---|
| **A** | Processor | Western United States | 10/2017 | *Listeria monocytogenes* | Class 1 Recall |
| | | | 10/2017 | *Listeria monocytogenes* | Class 1 Recall |
| | | | 11/2017 | *Listeria monocytogenes* | Class 1 Recall |
| **B** | Processor | Western United States | 6/2018 | *Listeria monocytogenes* | Referred to State, ** Processor Notified† |
| | | | 6/2018 | *Listeria monocytogenes* | Referred to State, ** Processor Notified† |
| | | | 6/2018 | *Listeria monocytogenes* | Referred to State, ** Processor Notified† |
| | | | 7/2019 | *Listeria monocytogenes* | Referred to State, ** Processor Notified† |
| **C** | Processor | Southeastern United States | 7/2018 | *Listeria monocytogenes* | Processor Notified† |
| | | | 7/2018 | *Listeria monocytogenes* | Processor Notified† |
| | | | 10/2018 | *Salmonella* | Processor Notified† |
| | | | 10/2018 | *Salmonella* | Processor Notified† |

* Only months and years are listed to avoid identifying firms.
** Firm was referred to state authorities because the product was manufactured in store for retail sale in the same store, and thus there was no interstate commerce.
† No recall was conducted because the product was no longer available at retail.

### By Grocery Store vs. 'Other'

Of the 17 samples that tested positive, six were collected at grocery stores. The products from which these six samples were collected were made in the prep kitchens of the grocery stores, or at off-site facilities associated with these retail establishments. All six samples were positive for *Listeria monocytogenes*. The rest of the positive samples were collected at processor facilities, distribution hubs, or ports of entry.

### Effectiveness of HPP Treatment

Focusing on the samples for which the HPP treatment status is known, the FDA found an estimated bacterial prevalence for either *Salmonella* spp. or *Listeria monocytogenes* of 3.1% in samples not treated with HPP, and did not detect either pathogen in the HPP-treated samples (Table 10). These findings appear to align with other research that has validated HPP as an effective kill step. However, this assignment was not designed to make such a comparison, and neither did the data originate from a controlled experiment. The analytical results do not account for uncontrolled factors, such as the possible use of different equipment or procedures (e.g., levels of applied pressure and holding times), among other variables.

**Table 10: Pathogen Findings (Combined) by HPP Treatment Status**

| Category of HPP Treatment | Samples Collected | Samples Positive | Estimated Prevalence | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|
| **Not HPP Treated** | 323 | 10 | 3.1% | 1.1% | 8.2% |
| **HPP Treated** | 361 | 0 | 0% | – | – |

## REGULATORY APPROACH

The Federal Food, Drug, and Cosmetic Act (FD&C Act) authorizes the FDA to take regulatory action regarding adulterated food. Regulatory tools at the agency's disposal include warning letters, import alerts, import refusals, administrative detentions, seizures, injunctions, suspension of registration, and mandatory recalls (if a firm does not conduct an adequate voluntary recall).

Processed avocado and guacamole that test positive for *Salmonella* spp. or *Listeria monocytogenes* are adulterated under Section 402(a)(1) of the FD&C Act in that they bear or contain a poisonous or deleterious substance which may render them injurious to health. Such foods may be subject to regulatory action.

## PUBLIC HEALTH IMPACT AND FOLLOW-UP ACTIVITIES

The agency analyzed the pathogens detected in the processed avocado and guacamole samples to identify their genetic patterns and determine whether those pathogens may be linked to human illness. Based on the available data, the FDA was unable to determine whether processed avocado and/or guacamole were the food vehicles involved in any known human illnesses.[6]

Whenever the FDA detected a positive finding under this assignment, the agency sought to remove all affected product from the marketplace. Removal of contaminated products from the marketplace prevents consumption and thus avoids potential illnesses, consistent with the agency's prevention efforts.

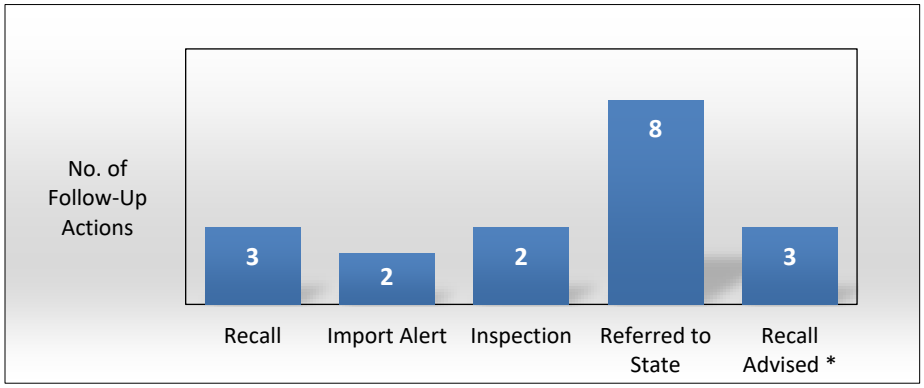
With respect to the domestic samples that tested positive for a target pathogen, the agency worked with the firm that owned or distributed the affected product to conduct a voluntary recall. In some cases, however, there was no product left to recall, or low likelihood of availability of product to recall, because of the commodity's relatively short shelf life. In cases where no recall was carried out, the agency provided the firm with guidance on minimizing microbial hazards and shared its findings with state partners (as is done with samples that result in recalls). In addition, the FDA conducted follow-up inspections as warranted.

With respect to the import samples that tested positive for a target pathogen, the FDA refused to admit the shipments associated with the positive findings into the U.S. and placed the responsible firms and product on Import Alert 21-12, thereby requiring additional controls for future entries. The FDA placed two firms on the import alert during this sampling assignment.

The chart below (Figure 1) reports the follow-up action(s) taken in response to all the positives.

[6] The FDA performed whole genome sequencing (WGS) analysis on the processed avocado and guacamole samples that tested positive for *Salmonella* spp. or *Listeria monocytogenes* and checked the National Center for Biotechnology Information Pathogen Database for possible linkage to clinical illness. In the case of all the positive food samples, either there was no linkage to any clinical illness or the available epidemiological information was inconclusive with respect to the food or other vehicle involved in the illnesses.

**Figure 1: Follow-Up Actions by Type**




* In three instances, the FDA advised firms to conduct a voluntary recall. However, the firms indicated that they were unable to carry out a recall because the product was past its shelf life and no longer on retail shelves. These three instances are separate and distinct from the effectuated recalls reported in the first bar.

## CONCLUSION AND NEXT STEPS

The agency accomplished the objectives of this sampling assignment, the most fundamental being to estimate the *Salmonella* spp. and *Listeria monocytogenes* contamination rates in processed avocado and guacamole. However, the adjustments to the collection target and binning of the data by HPP-treatment status, both required to maintain the integrity of the analysis, have served to reduce the sample sizes and therefore increase the uncertainty associated with the results.

As detailed in the Pathogen Findings section of this report, based on this assignment's test results, the FDA has estimated the *Salmonella* spp. and *Listeria monocytogenes* contamination rates in processed avocado and guacamole as follows:

> ***Salmonella* spp**. The prevalence of *Salmonella* spp. in the samples not subject to HPP was 0.6%, with a 95% confidence interval of 0.1% to 3.4%. The FDA did not detect the pathogen in the samples categorized as "HPP treated" or "could not ascertain."
>
> ***Listeria monocytogenes***. The prevalence of *Listeria monocytogenes* in the samples not HPP treated was 2.5%, with a with a 95% confidence interval of 1.0% to 6.3%. The prevalence of *Listeria monocytogenes* in the samples categorized as "could not ascertain" was 3.5%, with a 95% confidence interval of 1.1% to 10.1%. The agency did not detect positives in any of the HPP-treated samples.

While this assignment was designed primarily to estimate the overall prevalence of the target pathogens associated with processed avocado and guacamole, the agency also evaluated its test results preliminarily and throughout its sampling for signals (i.e., variations in prevalence by origin and season) to determine whether more targeted sampling or further study was warranted, the details of which can be found in Appendix D. The agency did not detect any signals related to origin or season that warranted more targeted sampling or additional study. The FDA also calculated and has provided the breakdowns that follow.

> **Guacamole vs. Processed Avocado:** Based on the test results, the FDA did not detect a difference in the contamination rate of either *Salmonella* spp. or *Listeria monocytogenes* in guacamole versus processed avocado (*P*-value > .05, for each comparison).

**Origin:** Based on the test results, the FDA estimated the prevalence of *Salmonella* spp. in domestically produced guacamole and processed avocado to be 0.4% and did not detect the pathogen in any of the import samples. Based on the test results, the agency estimated the prevalence of *Listeria monocytogenes* in domestically produced guacamole and processed avocado to be 2.3%, and in the import samples, to be 0.6%. The agency cautions against comparing the estimated prevalences by origin because it is not known whether the domestic finished products were uniformly made using domestically grown avocados, as opposed to imported avocados, as the main ingredient.

**Season:** The FDA did not detect a seasonal difference in the contamination rate of either *Salmonella* spp. (*P*-value > .05), or *Listeria monocytogenes* (*P*-value > .05).

In addition, the findings of this assignment appear to be consistent with other research that has shown HPP to be effective at neutralizing pathogenic bacteria, even as this assignment was not designed to compare possible differences in the contamination rate(s) between HPP-treated and non-HPP-treated samples.

The assignment data affirm that *Salmonella* spp. and *Listeria monocytogenes* may be present in processed avocado and/or guacamole and show that the estimated prevalence of the target pathogens in the non-HPP-treated samples was higher than in the HPP-treated samples, underscoring the need for processors of processed avocado and guacamole and others in the supply chain to comply with the agency's Preventive Controls for Human Food Rule, as applicable. The FDA has published draft guidance to help industry comply with its preventive controls rule.

For importers of processed avocado and guacamole, compliance with the agency's Foreign Supplier Verification Programs (FSVP) Rule is an important way to help ensure the safety of imported processed avocado and guacamole. The FDA has published draft guidance regarding its FSVP Rule and began FSVP inspections in 2017.

While the FDA learned that an increasing number of processors are using HPP (and did not detect pathogens in product samples identified as HPP-treated), the agency remains concerned about the potential for contamination in processed avocado and guacamole and so encourages industry attention to its preventive controls and FSVP rules, as described above. The FDA will continue to sample processed avocado and guacamole for pathogens as warranted, consistent with its mission to protect consumers.

## APPENDIX A: RESEARCH TO ASCERTAIN HPP TREATMENT STATUS

The FDA realized that its test data included an unknown number of samples subject to HPP shortly after beginning its analysis. As a result, the agency paused its evaluation, reassessed how to proceed, and subsequently queried industry as part of its effort to clarify its findings.

From July 2020 to December 2020, FDA personnel contacted each firm up to three times by phone or email or both, to ascertain whether the product(s) were subject to HPP at the time of sample collection.

The agency developed simple, parallel scripts for its phone and email outreach. Each script featured brief background information on the assignment and the reason for the correspondence. The FDA's use of the scripts made for a standardized approach that, coupled with the uniform three instances of outreach, served to put all firms on equal footing.

The agency contacted more than 200 manufacturers and/or grocery stores in total. Based on the responses (or lack of a response, in some cases), the FDA categorized all the samples collected and tested in one of three ways: "not HPP treated," "HPP treated," or "could not ascertain." By categorizing the data in this way, the FDA was able to resume its evaluation, as set forth in the body of this report.

## APPENDIX B: TEST METHODS

Analysts tested the samples using aseptic methods specific to each pathogen, as follows:

### *Salmonella* spp.

FDA analysts extracted 75 grams from each of five subsamples, combined them in a pre-enrichment lactose broth and incubated them for 24 hours at 35 degrees Celsius. The analysts used VIDAS *Salmonella* SLM (AOAC Official Method of Analysis [OMA] 2004.03) or VIDAS *Salmonella* Easy (AOAC OMA 2011.03) methods to detect *Salmonella*. The FDA's Bacteriological Analytical Manual (chapter 5) culture method for *Salmonella* was then used to confirm the VIDAS results. Sample enrichments positive for *Salmonella* were plated onto selective agars. Isolates were confirmed, serotyped, and subtyped using single nucleotide polymorphism (SNP) based whole genome sequence analysis.

### *Listeria monocytogenes*

FDA analysts extracted 25 grams from each of five subsamples, combined them in a *Listeria* enrichment broth and analyzed them using VIDAS *Listeria* (AOAC OMA 999.06) immunoassay or the method described in chapter 10 on *Listeria monocytogenes* in the FDA's Bacteriological Analytical Manual to detect the pathogen. Each method employed its own enrichment scheme. The FDA's Bacteriological Analytical Manual culture method for *Listeria monocytogenes* was then used to confirm the VIDAS results.

## APPENDIX C: PATHOGEN FINDINGS BY YEAR/SEASON AND HPP-TREATMENT STATUS

This table categorizes the pathogen findings by year and season, as well as HPP-treatment status.

| Year | Season | HPP Treatment* | Samples Collected | *Salmonella* spp. | *Listeria monocytogenes* |
|---|---|---|---|---|---|
| 2017 | Fall | N | 39 | 0 | 4 |
| 2017 | Fall | U | 19 | 0 | 0 |
| 2017 | Fall | Y | 49 | 0 | 0 |
| 2017 | Winter | N | 0 | 0 | 0 |
| 2017 | Winter | U | 1 | 0 | 0 |
| 2017 | Winter | Y | 1 | 0 | 0 |
| 2018 | Spring | N | 43 | 0 | 0 |
| 2018 | Spring | U | 45 | 0 | 4 |
| 2018 | Spring | Y | 62 | 0 | 0 |
| 2018 | Summer | N | 40 | 0 | 3 |
| 2018 | Summer | U | 28 | 0 | 0 |
| 2018 | Summer | Y | 38 | 0 | 0 |
| 2018 | Fall | N | 22 | 2 | 0 |
| 2018 | Fall | U | 10 | 0 | 0 |
| 2018 | Fall | Y | 21 | 0 | 0 |
| 2018 | Winter | N | 32 | 0 | 0 |
| 2018 | Winter | U | 10 | 0 | 0 |
| 2018 | Winter | Y | 66 | 0 | 0 |
| 2019 | Spring | N | 48 | 0 | 0 |
| 2019 | Spring | U | 34 | 0 | 2 |
| 2019 | Spring | Y | 49 | 0 | 0 |
| 2019 | Summer | N | 80 | 0 | 1 |
| 2019 | Summer | U | 46 | 0 | 1 |
| 2019 | Summer | Y | 55 | 0 | 0 |
| 2019 | Fall | N | 10 | 0 | 0 |
| 2019 | Fall | U | 1 | 0 | 0 |
| 2019 | Fall | Y | 6 | 0 | 0 |
| 2019 | Winter | N | 9 | 0 | 0 |
| 2019 | Winter | U | 9 | 0 | 0 |
| 2019 | Winter | Y | 14 | 0 | 0 |

* N = No (Not HPP Treated)
Y = Yes (HPP Treated)
U = Unknown (i.e., Could Not Ascertain)

## APPENDIX D: PATHOGEN FINDINGS BY PRODUCT ORIGIN AND SEASON

### By Origin: *Salmonella* spp.

Of the 571 domestic samples, two that were not HPP treated tested positive for *Salmonella* spp. (Table D1).

**Table D1*: Salmonella* spp. in Domestic Samples**

| Category of HPP Treatment | Samples Collected | Samples Positive | Estimated Prevalence | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|
| **Not HPP Treated** | 248 | 2 | 0.8% | 0.2% | 4.3% |
| **HPP Treated** | 201 | 0 | 0% | – | – |
| **Could Not Ascertain** | 122 | 0 | 0% | – | – |
| **Total** | 571 | 2 | 0.4% | 0.1% | 2.0% |

Of the 316 import samples, none tested positive for *Salmonella* spp. (Table D2).

**Table 92: *Salmonella* spp. in Import Samples**

| Category of HPP Treatment | Samples Collected | Samples Positive | Estimated Prevalence | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|
| **Not HPP Treated** | 75 | 0 | 0% | – | – |
| **HPP Treated** | 160 | 0 | 0% | – | – |
| **Could Not Ascertain** | 81 | 0 | 0% | – | – |
| **Total** | 316 | 0 | 0% | – | – |

The FDA has provided the analytical results in the two tables directly above for informational purposes. It may seem sensible to seek to compare the estimated contamination rates by product origin, but the FDA cautions against doing so based solely on the findings in Tables D1 and D2. Importantly, such a comparison may be invalid given that the agency does not know whether the domestic finished products were uniformly made using domestically grown avocados and given that some among them may well have been manufactured with imported avocados as the main ingredient.

### By Origin: *Listeria monocytogenes*

Of the 571 domestic samples, seven that were not HPP treated and six categorized as "could not ascertain" tested positive for *Listeria monocytogenes* (Table D3).

**Table 103*: Listeria monocytogenes* in Domestic Samples**

| Category of HPP Treatment | Samples Collected | Samples Positive | Estimated Prevalence | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|
| **Not HPP Treated** | 248 | 7 | 2.8% | 1.0% | 7.6% |
| **HPP Treated** | 201 | 0 | 0.0% | – | – |
| **Could Not Ascertain** | 122 | 6 | 4.9% | 1.4% | 15.7% |
| **Total** | 571 | 13 | 2.3% | 1.0% | 5.2% |

Of the 316 import samples, one that was not HPP treated, and one categorized as "could not ascertain" tested positive for *Listeria monocytogenes* (Table D4).

**Table D4: *Listeria monocytogenes* in Import Samples**

| Category of HPP Treatment | Samples Collected | Samples Positive | Estimated Prevalence | 95% Confidence Interval Lower Bound | 95% Confidence Interval Upper Bound |
|---|---|---|---|---|---|
| **Not HPP Treated** | 75 | 1 | 1.3% | 0.2% | 8.8% |
| **HPP Treated** | 160 | 0 | 0% | – | – |
| **Could Not Ascertain** | 81 | 1 | 1.2% | 0.2% | 6.7% |
| **Total** | 316 | 2 | 0.6% | 0.2% | 2.3% |

The FDA has provided the analytical results in the two tables directly above for informational purposes and again cautions against comparing the estimated contamination rates by product origin based solely on this assignment's findings. If some of the domestic finished products were manufactured using imported avocados as the main ingredient, which may well have been the case, the comparison would be invalid.

*Listeria monocytogenes* may be introduced into processed avocado and/or guacamole either through the presence of the pathogen in the avocados used as an ingredient to make the finished products[7] or by transmission from within the manufacturing environment.[8, 9]

### By Season

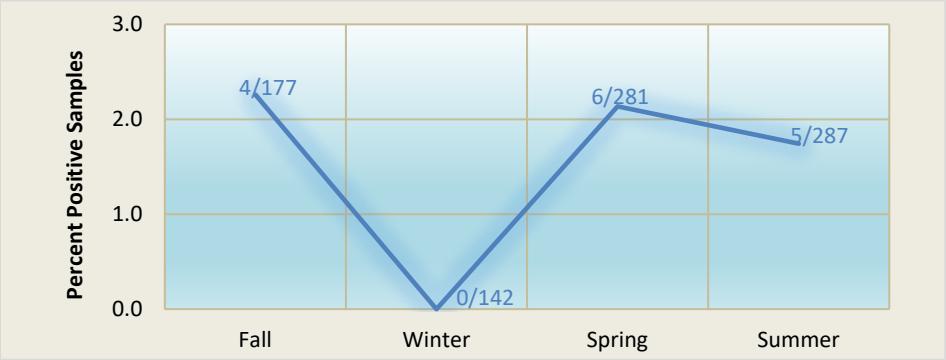
The FDA did not design its sample collection to compare bacterial prevalence by season and therefore cautions against making inferences based solely on the analytical results that follow, which are provided for informational purposes.

The FDA detected *Salmonella* spp. in two out of 177 samples collected in the fall and did not detect the pathogen in the other seasons. Although the FDA only detected *Salmonella spp.* in the samples collected in the fall, a Fisher's exact test did not find a seasonal difference in the *Salmonella* spp. contamination rate (*P*-value > .05).

The FDA detected the 15 *Listeria monocytogenes* positives in the spring (6), summer (5), and fall (4). A Fisher's exact test did not find a seasonal difference in the *Listeria monocytogenes* contamination rate (*P*-value > .05). The graph that follows provides the total (i.e., uncategorized) *Listeria monocytogenes* findings by season (Figure D1).

[7] A 2018 report by the FDA found a *Listeria monocytogenes* contamination rate of 0.2% in the 1,254 samples of avocado pulp tested. The report, titled "Microbiological Surveillance Sampling: FY14-16 Whole Fresh Avocados," is published on FDA.gov.
[8] Tompkin, R.B. (2002). Control of *Listeria monocytogenes* in the Food-Processing Environment. *Journal of Food Protection*, Vol. 65, No. 4, pp. 709–725.
[9] Garner, D., & Kathariou, S. (2016). Fresh Produce–Associated Listeriosis Outbreaks, Sources of Concern, Teachable Moments, and Insights. *Journal of Food Protection*, Vol. 79, No. 2, pp. 337-344.

**Figure D1: *Listeria monocytogenes* Findings by Season**




The fractions in the graph's plot area report the number(s) of samples that tested positive for *Listeria monocytogenes* out of the total number of samples collected for the indicated season.

The FDA also categorized its pathogen findings by year and season into the three HPP categories used throughout this report. Those findings are provided in Appendix C. Of note, the binning by year and season substantially reduces the individual sample sizes, with fewer than 50 samples in each season, for most of the seasons, as shown in Appendix C.