Guidance for Industry: Standards for the Growing, Harvesting, Packing, and Holding of Sprouts for Human Consumption

You may submit electronic or written comments regarding this guidance at any time. Submit electronic comments to https://www.regulations.gov. Submit written comments to the Dockets Management Staff (HFA-305), Food and Drug Administration, 5630 Fishers Lane, rm. 1061, Rockville, MD 20852. All comments should be identified with Docket No. FDA-2017-D-017517.

For questions regarding this document contact FDA's Technical Assistance Network by submitting your question at https://www.fda.gov/food/food-safety-modernization-act-fsma/fsma-technical-assistance-network-tan.

U.S. Department of Health and Human Services
Food and Drug Administration
Center for Food Safety and Applied Nutrition

Month and Year of Publication

Table of Contents

Guidance for Industry: Standards for the Growing, Harvesting, Packing, and Holding of Sprouts for Human Consumption

I. Introduction

This guidance is intended for those persons ("you") who grow, harvest, pack and/or hold sprouts covered by Subpart M of our final rule, published in the *Federal Register* (80 FR 74353) on November 27, 2015, entitled, "Standards for the Growing, Harvesting, Packing, and Holding of Produce for Human Consumption" (the Produce Safety Rule or the Rule).

The Produce Safety Rule established for the first time U.S. Federal requirements for the growing, harvesting, packing, and holding of produce for human consumption, including sprouts (Title 21 Code of Federal Regulations part 112 (21 CFR part 112)). The Rule focuses on certain conditions and practices identified as common routes of contamination of produce (similar to the areas covered by the 1998 Guidance, "Guide to Minimize Microbial Food Safety Hazards for Fresh Fruits and Vegetables" (GAPs Guide)) (Ref. 1). The Rule establishes requirements addressing common routes of microbial contamination, including: agricultural water; biological soil amendments of animal origin; worker health and hygiene; equipment, tools, buildings and sanitation; and domesticated and wild animals.

Sprouts represent a distinct food safety concern because the conditions under which sprouts are produced (i.e., temperature, water activity, pH and available nutrients) are also ideal for the growth of pathogens, if present (Ref. 2). Between 1996 and 2020 in the United States, FDA observed 52 reported outbreaks of foodborne illness associated with sprouts. Together, it is estimated that these outbreaks resulted in at least 2700 cases of illness, 200 hospitalizations, and three deaths (Ref. 3, Ref. 4, Ref. 5, Ref. 6, Ref. 7, and Ref. 8). During this timeframe, sprouts have been associated with outbreaks of several different pathogens, including *Salmonella* spp., *Listeria monocytogenes*, *E. coli* O157:H7, and several types of non-O157:H7 pathogenic *E. coli* (*i.e.*, *E. coli* O157:NM (H-), *E. coli* O104:H4, *E. coli* O26, *E. coli* O121, *E. coli* O103) (Ref. 9 and Ref. 10). In foodborne illness outbreaks associated with sprouts where the source of

contamination was identified, epidemiological investigations often identify the most likely source of contamination as seeds used for sprouting (Ref. 2 and Ref. 11). However, poor sanitation and unhygienic practices at the sprout operation have also contributed to the contamination of sprouts (Ref. 2, Ref. 6, Ref. 12, Ref. 13, and Ref. 14).

Because the distinctive practices and conditions for growing sprouts present unique risks, we established sprout-specific requirements in Subpart M (Sprouts) of the Produce Safety Rule. Subpart M of the Rule builds on sprout production practices similar to areas covered in our 1999 Sprout Guidances (discussed further below, now withdrawn). Sprout operations subject to the Produce Safety Rule must comply with all applicable requirements in the Rule, including, but not limited to, all applicable requirements in Subpart M.

The requirements in the Produce Safety Rule are directed specifically to covered farms (as that term is defined in the Rule) that grow, harvest, pack or hold covered produce, including sprouts. Covered farms that grow, harvest, pack or hold sprouts are referred to in this guidance as "sprout operations," or "you." Produce that is covered by the Rule is referred to as "covered produce."

Neither the Produce Safety Rule nor this guidance document is directed to growing, conditioning, or distributing seed for sprouting or to the handling of sprouts at a retail food establishment. However, as noted in our prior Sprout Guidances and our May 2009 letter to suppliers and distributors of seed for sprouting and sprout operations (Ref. 15 and Ref. 16), everyone in the sprout supply chain has a responsibility to help ensure food safety. FDA has also issued the final guidance document regarding good agricultural practices for seed for sprouting, entitled "Reducing Microbial Food Safety Hazards in the Production of Seed for Sprouting: Guidance for Industry" (Ref. 17). The Produce Safety Rule does not address chemical or physical hazards. However, you have a responsibility to ensure that your sprouts are not adulterated or misbranded under the Federal Food, Drug, and Cosmetic Act (FD&C Act) (21 U.S.C. §§ 301 *et seq.*) and are produced in compliance with all applicable laws and regulations. Under section 402(a)(1) of the FD&C Act, a food is adulterated if it bears or contains any added poisonous or deleterious substance which may render it injurious to health, and such substances may include or otherwise result from physical and chemical (including radiological) contamination.

This guidance provides our current thinking and recommendations to assist sprout operations subject to the Produce Safety Rule primarily in complying with the sprout-specific requirements in Subpart M. This guidance also briefly discusses certain requirements (in Subparts E and O of the Rule relating to Agricultural Water and Records) of particular relevance to a sprout operation. Some requirements of Subpart M and other subparts to which sprouts are subject (e.g., Subpart M, §112.147, related to spent sprout irrigation water; Subpart C, related to training; and Subpart L, related to equipment, tools, and buildings) are covered in the draft reissued sections (Ref. 18). In addition, this guidance may also be useful to sprout operations that are not subject to the Produce Safety Rule that voluntarily choose to follow the standards established in the Rule. In the development of this guidance, we particularly considered industry and international documents related to food safety and hygienic production of sprouts (Ref. 19, Ref. 20, and Ref. 21). We have incorporated aspects of these documents that are consistent with our laws, regulations, and existing policies.

Some of the material in this guidance relates to regulatory requirements of the Produce Safety Rule that are also covered in the draft "Standards for the Growing, Harvesting, Packing, and Holding of Produce for Human Consumption: Guidance for Industry" (general produce compliance and implementation draft guidance (83 FR 53196) issued in October 2018) (Ref. 22). At the present time, the material on these overlapping topics (e.g., records, cleaning and sanitizing) is consistent between the two guidance documents. This final guidance focuses specifically on insights drawn from FDA's experience with sprout operations, such as from inspections and sprout-associated foodborne illness outbreak investigations and elaborates on how the broader records and cleaning and sanitizing standards in part 112 could apply to and be implemented in a sprout operation.

In general, FDA's guidance documents do not establish legally enforceable responsibilities. Instead, guidances describe the Agency's current thinking on a topic and should be viewed only as recommendations, unless specific regulatory or statutory requirements are cited. The use of the word should in Agency guidances means that something is suggested or recommended, but not required.

II. Background

A. Other FDA Efforts Related to Sprouts

On October 27, 1999, we published a notice of availability in the *Federal Register* (64 FR 57893) for two guidance documents to inform all parties involved in the production of sprouts (i.e., producers, conditioners, and distributors of seeds used for sprouting, and sprout operations) that sprouts had been recognized as an important cause of foodborne illness and to provide recommendations for preventive controls that we believed should be taken immediately to reduce the likelihood of sprouts serving as a vehicle for foodborne illness. We refer to these prior (now withdrawn) guidance documents collectively as the 1999 Sprout Guidances.

FDA and our food safety partners in the public and private sectors have engaged in education and outreach to the sprout industry to promote adoption of our recommendations. We have also worked with the sprout industry to advance the scientific knowledge applicable to enhancing the safety of sprouts. For example, in 2000, we collaborated with the California Department of Public Health, in cooperation with the sprout industry and academia, to develop an educational video series entitled, "Safer Processing of Sprouts" (Ref. 23). In addition, we have provided technical assistance to the Illinois Institute of Technology's Institute for Food Safety and Health (IIT IFSH) Sprout Safety Taskforce in developing their "US Sprout Industry Production Best Practices" (Ref. 20).

We also have been working with the Sprout Safety Alliance (SSA), a public-private partnership, since 2012 to develop a standardized curriculum and training and outreach programs for stakeholders in the sprout industry to enhance the industry's understanding and implementation of the requirements of the Produce Safety Rule, and of best practices for improving sprout safety (Ref. 24, Ref. 25, and Ref. 26). Additionally, in 2023, the SSA developed a series of educational videos entitled, "Food Safety Best Practices for Sprout Production". These videos focus on demonstrating key sprout safety components of the Produce Safety Rule (cleaning and sanitizing; environmental sampling; seed treatment; and sampling and testing spent sprout irrigation water)

within a sprout operation (Ref. 27). The SSA is composed of representatives from the sprout industry, retailers, academia, and federal, state, and local food safety agencies. The SSA is funded by a grant from FDA to IIT IFSH.

B. Coverage of the Produce Safety Rule

Under § 112.1 of the Produce Safety Rule, unless specifically excluded under § 112.2, food that is produce (as that term is defined in the Rule), and that is a raw agricultural commodity (RAC), is covered by the Rule. This includes a produce RAC that is grown domestically and a produce RAC that will be imported or offered for import into any State or territory of the United States, the District of Columbia, or the Commonwealth of Puerto Rico. Covered farms (i.e., those subject to the Produce Safety Rule) must comply with all applicable requirements of the Rule when conducting a covered activity on covered produce (see § 112.4). There are certain exclusions, exemptions and limitations on which farms are "covered farms" (see § 112.4). For example, sprout operations for which, on a rolling basis, the average annual monetary value of produce sold during the previous 3-year period, adjusted for inflation, is less than or equal to $25,000, are not covered by this regulation (§ 112.4(a)). See https://www.fda.gov/food/food-safety-modernization-act-fsma/fsma-inflation-adjusted-cut-offs for information on FDA Food Safety Modernization Act (FSMA) inflation adjusted cut-offs. See also "Standards for Produce Safety, Coverage and Exemptions/Exclusions for 21 Part 112", available at https://www.fda.gov/media/94332/download to assist you in determining which provisions of the Produce Safety Rule, if any, apply to you.

Sprouts are produce as we defined that term in the Produce Safety Rule (see § 112.3). Sprouts are also RACs when they are in their raw or natural state (see § 112.3 and section 201(r) of the FD&C Act, defining "raw agricultural commodity" as "any food in its raw or natural state, including all fruits that are washed, colored, or otherwise treated in their unpeeled natural form prior to marketing."). Therefore, sprouts in their raw or natural state are covered produce, except as otherwise provided in § 112.2.

If sprouts are made into processed food(s), those processed foods are not covered by the Produce Safety Rule (see § 112.2(a)(3)). An example of a processed food made using sprouts is sprouted seed butter. This does not mean that sprout RACs that will be made into processed food are themselves exempt from the Produce Safety Rule simply because those RACs will later be transformed into processed food. Rather, it means that the Produce Safety Rule only applies during the time that the sprouts are RACs. Other requirements, such as those in 21 CFR Part 117, may apply to any manufacturing/processing of the sprout RACs into processed food, depending on the circumstances.

Sprouts do not qualify for the exemption from the Produce Safety Rule in § 112.2(a)(1) for produce that is rarely consumed raw. This provision contains a list of produce commodities (such as potatoes) for which we have analyzed dietary consumption patterns and determined such commodities are rarely consumed raw. This exemption applies only to the commodities identified in § 112.2(a)(1), none of which are sprouts.

Some sprouts may be eligible for exemption from the Produce Safety Rule under § 112.2(b), if they will receive commercial processing that adequately reduces the presence of microorganisms

of public health significance. Examples of commercial processing that adequately reduces the presence of microorganisms of public health significance appear in § 112.2(b)(1). This exemption requires covered farms to establish and maintain certain documentation and disclosures set forth in § 112.2(b)(2)-(3): 1) disclosures to customers; and 2) annual written assurances obtained from customers. Sprouts that receive commercial processing (e.g., canned, shelf-stable mung bean sprouts) could potentially qualify for this exemption from the Produce Safety Rule, but only if the commercial processing adequately reduces the presence of microorganisms of public health significance and all documentation and disclosure requirements are met.[1] We note that simply drying/dehydrating sprouts may not adequately reduce the presence of microorganisms of public health significance (Ref. 28 and Ref. 29) and therefore would not make them eligible to receive the commercial processing exemption from the Rule.

Some sprout operations may be eligible for a qualified exemption and associated modified requirements in a calendar year if, during the previous 3-year period preceding the applicable calendar year, the average annual monetary value of the food (as defined in § 112.3) the sprout operation sold directly to qualified end-users (as defined in § 112.3) during such period exceeded the average annual monetary value of the food the sprout operation sold to all other buyers during that period; and the average annual monetary value of all food (as defined in § 112.3) the sprout operation sold during the 3-year period preceding the applicable calendar year was less than $500,000, adjusted for inflation. (FDA makes available the updated inflation-adjusted figures at https://www.fda.gov/food/food-safety-modernization-act-fsma/fsma-inflation-adjusted-cut-offs.) Section 112.6(b) describes the modified requirements applicable to a qualified exempt operation (e.g., labeling). Section 112.7 requires the sprout operation eligible for the qualified exemption, provided for in § 112.5, to establish and keep adequate records necessary to demonstrate that the sprout operation satisfies the criteria for a qualified exemption (e.g., dated sales receipts), including a written record reflecting that the owner, operator, or agent in charge of the sprout operation has performed an annual review and verification of the sprout operation's continued eligibility for the qualified exemption.

C. Coverage of Subpart M

The requirements in 21 CFR part 112, Subpart M apply to the growing, harvesting, packing and holding of all sprouts except sprouts that are grown in soil or non-soil substrates (e.g., mats, perlite or other growth media) and that are harvested above the soil or substrate line without their

[1] In the *Federal Register* of January 5, 2018 (83 FR 598), we published a notification of availability of a guidance document titled "Policy Regarding Certain Entities Subject to the Good Manufacturing Practice and Preventive Controls, Produce Safety, and/or Foreign Supplier Verification Programs." In that guidance document, we stated that we intend to exercise enforcement discretion regarding certain written assurance requirements, including those in 21 CFR part 112 (the Produce Safety Rule). We intend to exercise such discretion until we can complete a rulemaking process to consider options for the assurance requirements. https://www.fda.gov/regulatory-information/search-fda-guidance-documents/guidance-industry-policy-regarding-certain-entities-subject-current-good-manufacturing-practice-and. See also the *Federal Register* of March 14, 2022 (87 FR 14169), in which we published a notification of availability of a guidance document titled "Current Good Manufacturing Practice and Preventive Controls, Foreign Supplier Verification Programs, Intentional Adulteration, and Produce Safety Regulations: Enforcement Policy Regarding Certain Provisions." In that guidance document, we restated our intent to exercise enforcement discretion with respect to the Produce Safety Rule assurance requirements, as previously indicated in the 2018 guidance document described immediately above. https://www.fda.gov/regulatory-information/search-fda-guidance-documents/guidance-industry-current-good-manufacturing-practice-and-preventive-controls-foreign-supplier.

roots (see § 112.141). We determined that soil- or substrate-grown sprouts that are harvested above the soil or substrate line, such that their roots are not harvested for human consumption, do not present the same risks as other types of sprouts and, therefore, we excluded them from the sprout-specific requirements in Subpart M (80 FR 74353 at 74497). However, the requirements of Subpart M do apply to soil- or substrate-grown sprouts that are harvested with the roots. If you use soil or substrate as a growth medium in your operation, in addition to all other relevant requirements of the Produce Safety Rule, you must comply with the applicable requirements in 21 CFR part 112, Subpart F (Biological Soil Amendments of Animal Origin and Human Waste). We discuss the requirements of Subpart F in the draft compliance and implementation guidance (Ref. 22).

We recognize that sprout operations may sell certain soil- or substrate-grown sprout types, such as wheatgrass, in a tray used for growing, with the soil/substrate and root intact, to commercial entities, including facilities that are required to register with FDA under section 415 of the FD&C Act, restaurants, retail food establishments, and non-profit food establishments. In such cases, the operation that has harvested the sprouts with the roots and these sprouts are ordinarily subject to Subpart M (see § 112.141). However, we understand that, in some cases, such sprouts may then be cut above the soil and/or substrate line at the commercial entity immediately before use or sale to the consumer. When a sprout operation sells sprouts with the roots intact in soil or substrate, and the commercial entity that receives them will cut the sprouts above the soil or substrate line before use, we intend to exercise enforcement discretion for the requirements of Subpart M if the sprout operation annually collects written assurances from the commercial entity stating that the sprouts will be cut above the soil or substrate line before use (see also Section VII.C.1).

Note that soil- or substrate-grown sprouts harvested above the soil line are still considered covered produce and, unless exempt or excluded under the provisions of 21 CFR part 112, Subpart A, are subject to all other applicable requirements of the Produce Safety Rule. The standards in Subpart M may be useful to sprout operations that produce soil- or substrate-grown sprouts that are harvested above the soil or substrate line that voluntarily choose to follow these standards, in addition to complying with the required provisions of all other subparts in the Produce Safety Rule.

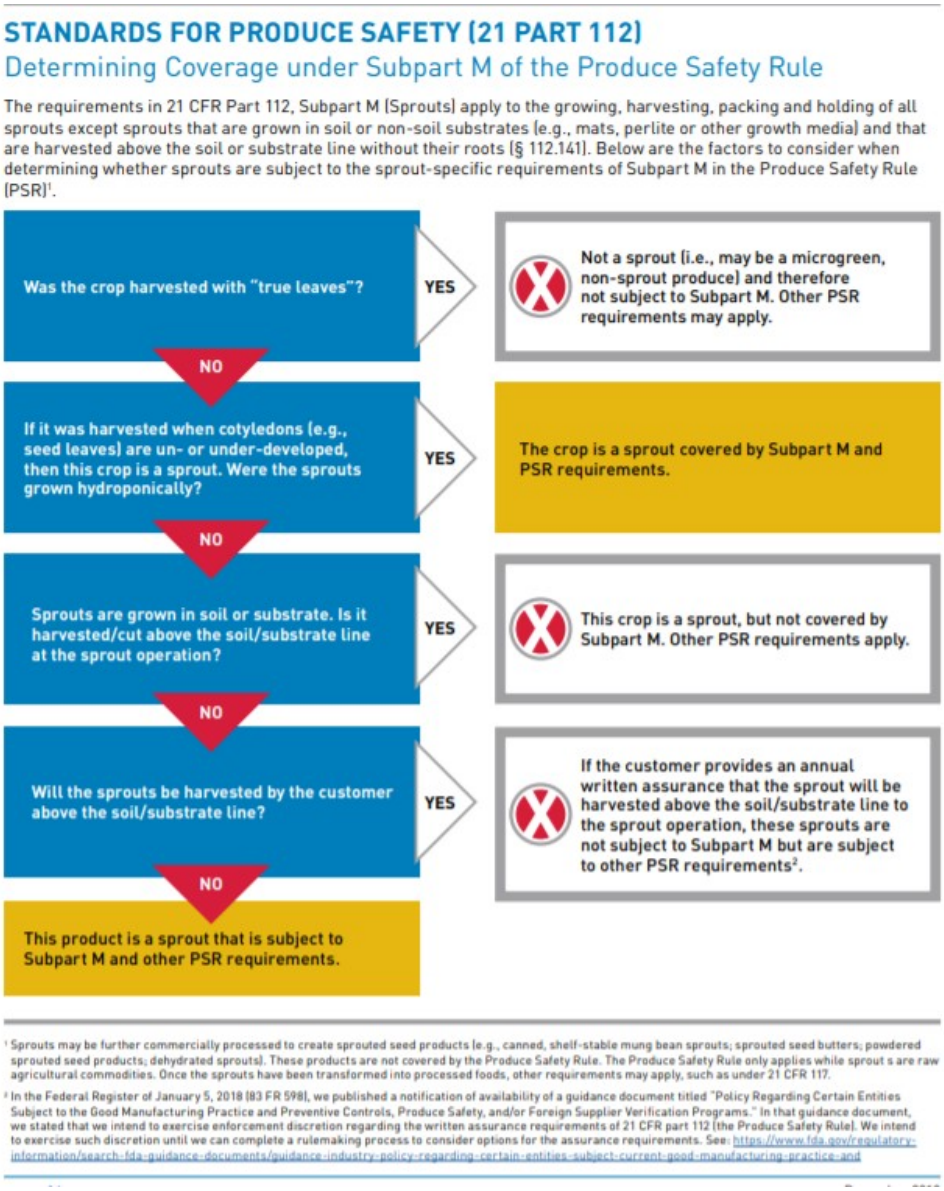
We are aware that certain sprouts are grown hydroponically in trays, and then harvested without the root at either the sprout operation or by the customer. As we previously stated in the preamble to the final rule (80 FR 74353 at 74497, comment/response 364), all hydroponically grown sprouts are subject to the requirements of Subpart M. Under typical conditions for growing hydroponic sprouts, water circulates between sprouts in the same growing unit, such that any pathogens present in the seed or introduced during sprouting can spread throughout the production lot of sprouts (Ref. 2, Ref. 30, and Ref. 31).

We note that microgreens and sprouts are different products. This interpretation is consistent with our prior Sprout Guidances and with other public and private standards (e.g., IIT IFSH Sprout Taskforce US Sprout Industry Production Best Practices document (Ref. 20), Food Safety Australia New Zealand (FSANZ) standards for sprouts (Ref. 32)). Historically, the primary criterion we have used to distinguish between the two product categories has been the growth stage of the leaves. Sprouts are usually harvested when the cotyledons (i.e., seed leaves) are still

un- or under-developed and true leaves have not begun to emerge. In contrast, microgreens reach a later stage of growth, typically associated with the emergence of "true" leaves. Unlike sprouts covered by Subpart M, microgreens are typically grown in soil or substrate and harvested above the soil or substrate line. Because microgreens are not sprouts, they are not subject to the requirements in Subpart M (Ref. 33). However, microgreens are considered covered produce for the purposes of the Produce Safety Rule and, unless exempt or excluded under the provisions in Subpart A, microgreens are subject to the other subparts of the Produce Safety Rule (80 FR 74353 at 74497, comment/response 363).

The following figure provides factors to consider when determining whether sprouts are subject to the sprout-specific requirements of Subpart M of the Produce Safety Rule.

Figure 1. Determining Coverage under Subpart M of the Produce Safety Rule



Note that throughout this guidance, for the ease of the reader, we often refer collectively to everything, including beans, sprouted to produce sprouts for human consumption simply as "seeds." In the Rule, we used the phrase "seeds or beans" to remove any potential confusion as to whether beans for sprouting were included. References to "seeds" in this guidance should be read to include things other than seeds that are sprouted to produce sprouts for human consumption, such as beans.

In addition, the definition of "produce" includes sprouts irrespective of seed source (see § 112.3). We consider sprouts to be a distinct category of produce that includes many different varieties (e.g., alfalfa, mung). Sprouts (i.e., alfalfa, mung and soybean sprouts) were evaluated to determine whether they are rarely consumed raw, and therefore exempt from the Produce Safety Rule (see 80 FR 74353 at 74392, comment/response 68). Sprouts did not meet the threshold criteria for the rarely consumed raw exemption and are therefore considered covered produce. The mature plant and/or the seeds used to grow sprouts may or may not fall under the definition of "produce." For example, wheatgrass sprouts and soybean sprouts have long been considered sprouts by the food industry and FDA and are covered produce under the Produce Safety Rule. However, for both wheat and soybeans, the mature crops are considered food grains, which do not fall under the definition of "produce" and are not subject to the Produce Safety Rule. Sprouts may also be produced from seeds of produce that is considered to be rarely consumed raw, such as beet or pumpkin sprouts. While the mature produce may be rarely consumed raw, and therefore exempt from the Produce Safety Rule, the sprouts produced from the seeds of these crops are still covered produce under the Produce Safety Rule because we consider sprouts to be a distinct commodity from the mature form, and sprouts have not met the threshold criteria for the rarely consumed raw exemption.

D. Compliance Dates for Sprouts

Sprout operations covered by the Produce Safety Rule and subject to Subpart M must be in compliance with all applicable provisions of the Produce Safety Rule at this time. The extended compliance dates provided for covered farms producing other produce commodities to comply with the agricultural water requirements of Subpart E do not apply to the production of sprouts subject to Subpart M. Sprout operations eligible for the qualified exemption from the Produce Safety Rule are also expected to be in compliance with the applicable modified requirements at this time.

The same compliance dates that apply to all other covered produce apply to sprouts that are not subject to Subpart M but are otherwise subject to the Produce Safety Rule (i.e., soil- or substrate-grown sprouts harvested without their roots).

E. General Sprout Production

This section discusses common production practices for growing, harvesting, packing, packaging and holding of sprouted seeds. Later sections of this guidance describe the requirements of the Produce Safety Rule and provide recommendations for sprouts operations in conducting the different steps in sprout production.

Typically, sprout production consists broadly of the steps depicted in Figure 2. Your operation may add to or omit some of these practices (or do them in a different order) depending on a number of factors, including: the type of seeds you sprout; whether the required seed treatment is applied by you, your seed supplier, or both; and the size and resources of your operation.

Figure 2. Typical Sprout Production Processes (Adapted from Ref. 2)

Seed Receipt → Seed Storage → Initial Seed Rinse → Seed Treatment → Pre-germination Seed Soak → Germination and Growth → Microbial Testing of Spent Sprout Irrigation Water (or in-process sprouts) → Harvest →Wash/Drain Sprouts →Bulk Cool/Spin Dry → Pack and/or Package → Cooling & Storage →Distribution

III. Cleaning and Sanitizing

Cleaning and sanitizing are different, and important, steps that are critical to the safety of your finished sprouts. Cleaning refers to the practice of removing organic material and other debris from surfaces. The Produce Safety Rule defines "sanitize" to mean "to adequately treat cleaned surfaces by a process that is effective in destroying vegetative cells of microorganisms of public health significance, and in substantially reducing numbers of other undesirable microorganisms, but without adversely affecting the product or its safety for the consumer" (§ 112.3). When cleaning and sanitizing is required, you must properly clean surfaces prior to sanitizing because many sanitizers will not be effective unless food and dirt have been removed from the surface first (see definition of "sanitize" in § 112.3, "to adequately treat *cleaned* surfaces …" (emphasis added)). Sanitizing surfaces is usually achieved through chemical means.

This section discusses cleaning and sanitizing policies and procedures, frequencies, verification activities, recordkeeping, and corrective actions to take in response to suspected or known contamination.

A. Frequency of Cleaning and Sanitizing

Section 112.123(d)(1) is a general requirement that applies to all covered farms, and requires you to inspect, maintain, clean and, when necessary and appropriate, sanitize all "food contact surfaces" (FCSs) of equipment and tools used in covered activities as frequently as reasonably necessary to protect against contamination of covered produce. For operations growing sprouts covered by Subpart M, we determined that it is "necessary and appropriate" to sanitize all such FCSs used to grow, harvest, pack, or hold sprouts after cleaning the FCSs and therefore we specifically require both cleaning and sanitizing for such FCSs prior to contact with sprouts or seeds used to grow sprouts, as reflected in § 112.143(b).

"Food contact surfaces" means "those surfaces that contact human food and those surfaces from which drainage, or other transfer, onto the food or onto surfaces that contact the food ordinarily occurs during the normal course of operations. 'Food contact surfaces' includes food contact surfaces of equipment and tools used during harvest, packing, and holding" (§ 112.3). "Food contact surfaces" also includes FCSs of equipment and tools used in growing sprouts, such as those that contact seeds for sprouting (see e.g., § 112.143(b)). In a sprouting operation, FCSs include, for example: trays or drums used for sprouting; interior surfaces of containers used for

seed rinsing, seed treatment, and pre-germination seed soaking; and counters that come into contact with sprouts during packing and/or packaging.

While the appropriate frequency of cleaning and sanitizing may vary based on your specific production practices, in general, we recommend cleaning and sanitizing of FCSs used in sprout operations at least daily to meet the requirements of §§ 112.123(d)(1) and 112.143(b). For example, you should clean and sanitize all FCSs in the production environment at the end of each production day. We recommend that initial sanitizing, of any surface, occur following the completion of cleaning. You may determine that there are some situations where it is necessary to re-sanitize an FCS in the morning after cleaning and sanitizing it the night before. To illustrate the requirement in § 112.143(b) that FCSs be cleaned and sanitized "before contact" with sprouts or seeds used to grow sprouts, we provide the following non-exhaustive list of examples of when you must incorporate a cleaning and sanitizing step for FCSs:

- Before contact with a different production batch of sprouts (e.g., when more than one production batch of sprouts, including seeds for a different production batch of sprouts, will contact the same FCSs on the same day). Cleaning and sanitizing FCSs between production batches of sprouts minimizes the potential for cross-contamination from one production batch to the next, and also could reduce the extent of corrective actions needed in response to a positive pathogen (or indicator organism) test result (e.g., from a spent sprout irrigation water, sprouts, or environmental sample), because it could serve as a point at which the risk of contamination of subsequent production batches is substantially reduced (see Section V (Sampling and Testing of Spent Sprout Irrigation Water (or In-Process Sprouts)) in the draft re-issued sections (Ref. 18)) and Section VI (Environmental Monitoring) in this document);

- Before the first time the FCS is used in your sprout operation to contact seeds for sprouting or sprouts. FCSs of such tools or equipment may have been contaminated during manufacturing, storage, or transport, or, if they were repurposed from another use, may have been contaminated during that use;

- Before contact with seeds or sprouts after storage for an extended period (e.g., several days, weeks, or months) since the FCSs of the equipment and tools may have become contaminated during storage;

- Before contact with seeds or sprouts after the FCS has been exposed to a source of contamination (e.g., contaminated water, human or animal fecal material, animal or insect activity); and

- Before contact with seeds or sprouts after contacting food other than sprouts, regardless of whether the food is produce that is covered by the Produce Safety Rule, because of the risk that pathogens transferred from these other foods may multiply in the unique conditions in which sprouts are grown.

Surfaces that are used continuously for more than a day, such as bins or drums that contain a single production batch of sprouts during germination and growth periods lasting more than a day, do not need to be cleaned or sanitized on a daily basis while they continuously contain or

otherwise contact only one production batch of growing sprouts. We consider such uses to be a single, continuous instance of contact. The end of one production batch and the beginning of the next should be considered a break in the single, continuous instance of contact, warranting cleaning and sanitizing of the surfaces between batches.

Additionally, you must maintain and clean all non-FCSs of equipment and tools subject to Subpart L that are used during harvesting, packing, and holding as frequently as reasonably necessary to protect against contamination of sprouts (§ 112.123(d)(2)). You may consider sanitizing your non-FCSs on the same schedule as your FCSs.

While the rule does not require sanitizing of non-FCSs, we recommend that sprout operations sanitize even non-FCSs on the same schedule that we recommend for cleaning FCSs because of the particularly high-risk nature of sprout production. We recommend cleaning and sanitizing all non-FCSs in accordance with the frequencies in Table 1 below. The recommendations in the following table are adapted from a 1999 publication by Tompkin et al. (Ref. 34). If the results of your environmental monitoring, spent sprout irrigation water testing, or product testing indicate a food safety concern, you should consider increasing the frequency of cleaning and sanitizing as part of an overall corrective action plan. If the manufacturer of equipment used in your operation recommends cleaning on a more frequent basis, we recommend that you increase this frequency to match the recommendations of the manufacturer.

Table 1. Recommended Frequency of Cleaning and Sanitizing Non-Food Contact Surfaces for Sprout Operations

Surface, Area, or Equipment	**Frequency of Cleaning and Sanitizing[a]**
Drains and floors	Daily
Waste containers	Daily
Cleaning tools (e.g., mops, brushes)	Daily
Surfaces that have a greater potential to become a source of *L. monocytogenes* contamination (e.g., surfaces likely to be touched by employees who touch product or FCSs during operations, or areas where there may be a build-up of moisture or product residue)	Daily
Condensate drip pans	Monthly
Motor housings, external surfaces of enclosed processing systems	Monthly
Overhead piping, ceilings and walls[b]	Semi-annually
Refrigerators (e.g., coolers) containing exposed RTE food	Quarterly
Interiors of Ice Makers	Semi-annually

[a] Since production environments vary, it may be appropriate to increase cleaning frequencies depending on the specific circumstances of the product area.

[b] We recommend that that you clean and sanitize walls and ceilings in close proximity to a production line at the same time as the production equipment (e.g., daily).

B. Cleaning and Sanitizing Policies and Recordkeeping

You should create policies that describe cleaning and sanitation procedures. These include schedules, needed materials, tools, and chemicals for specific cleaning and sanitizing tasks. You may need several policies describing various tasks throughout the production environment. Each piece of equipment, as well as the production environment itself, should be considered when developing your policies. While not required by the Produce Safety Rule, we recommend establishing these policies to promote consistent applications of your intended cleaning and sanitizing procedures by different workers. The following details should be included in each policy and periodically reviewed by management:

- For what piece(s) of equipment or area(s) of the production environment the policy was created;
- Who is responsible for the cleaning and sanitizing tasks (see discussion on § 112.22(a)(3) in Section III. A. 1 (Training and Supervision of Personnel who Conduct Cleaning and Sanitizing Activities) of the draft re-issued sections (Ref. 18));
- When and how often each task is to be completed;
- What tools and chemicals (for both cleaning and sanitizing) are needed;
- How the chemicals are to be prepared, as applicable;
- How the chemicals are to be used (including what precautions need to be taken), as applicable; and
- How to properly clean and sanitize the piece(s) of equipment or work area(s), including any equipment disassembly that is necessary.

You must establish and keep records of the date and method of cleaning and sanitizing of equipment used during growing operations for sprouts and all covered harvesting, packing, or holding activities (§ 112.140(b)). The records should be sufficient to demonstrate that your established cleaning procedures have been achieved (e.g., through the use of a cleaning and sanitizing checklist). Maintaining records of the performed activities not only helps you ensure activities are appropriately performed, but also serves as documentation to show auditors or inspectors, as necessary, that the activities were properly performed. These records should be maintained each time cleaning and sanitizing is performed, even if performed more than once per day.

C. Cleaning

You should establish your cleaning procedures based on the characteristics of the soils and debris; the type, design and age of equipment or tools; surface materials (e.g., stainless steel, plastic, concrete); the general effectiveness of your cleaning agent(s); label instructions; the results of sanitation verification activities (see Section III. E (Verification of Cleaning and Sanitizing)); and environmental monitoring results (see Section VI (Environmental Monitoring)).

Factors that influence the effectiveness of cleaning procedures include, but are not limited to, the following:

- Characteristics of the organic matter, sprouts, and other debris that would impact its removal from the equipment;
- The surface material (e.g., plastic, stainless steel, concrete, etc.) to be cleaned and its condition and construction. You should consider the surface material to be cleaned when selecting cleaning agents/detergents;
- Contact time of the cleaning agent/detergent with the surface(s) being cleaned; and
- The duration and force of physical scrubbing and the pressure of water or air used during the removal of the organic material.

Mops, brushes and other equipment used for cleaning and sanitizing should be durable and replaced immediately if damaged, cracked, or worn, to prevent the colonization of those tools by pathogens that could lead to contamination of product or FCSs. These tools and equipment should also be stored appropriately to prevent them from becoming soiled, and they should be cleaned and sanitized as needed.

The following steps are an example of a typical wet cleaning procedure (Ref. 35 and Ref. 36); some steps could be inapplicable to your operation or could be modified (Ref. 35 and Ref. 37):

1. *Pre-Clean*: personnel use tools such as brushes, brooms, scrapers, and collection pans to remove heavy residue, plant material or debris from the equipment or tools;
2. *Initial Rinse*: personnel rinse the equipment or tools with water to remove additional organic material and residue that was loosened during the pre-clean step; scrubbing could be necessary during this step;
3. *Washing with Cleaning Agent and Mechanical Action*: personnel apply a cleaning agent in one of several forms, such as by spray or foam; personnel should ensure full coverage of the relevant surface with the cleaning agent for the time and at the temperature specified by the cleaning agent manufacturer; small equipment or tools or components of these can be washed in sinks or tanks containing a solution of the cleaning agent; mechanical action (e.g., scrubbing) ordinarily should be applied, using tools such as brushes or scouring pads, in order to fully remove organic material and residues;

4. *Post-Rinse*: personnel apply water to remove loosened soil and debris along with the cleaning agent; after the post-rinse the equipment and tools could be allowed to dry prior to further activities; and

5. *Inspection*: personnel check the equipment or tools for any remaining soil or residue.

D. Sanitizing

Sanitizing is generally not effective unless it is preceded by a cleaning step, because residual organic material can insulate pathogens from the action of the sanitizing treatment (Ref. 35, Ref. 37, Ref. 38, Ref. 39, and Ref. 40).

The following should be considered when developing your sanitizing procedures and when sanitizing your equipment, tools, or production environment:

- The surfaces should be cleaned as described above in Section III. C (Cleaning) before sanitizing;

- Sanitizing approaches should take into account the type, duration, application method, temperature, and concentration of the sanitizing or antimicrobial treatment (Ref. 37 and Ref. 41);

- Verification activities, described below, should be conducted to evaluate the effectiveness of your cleaning and sanitizing activities; and

- Chemical sanitizing agents must be used according to label directions, in accordance with the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA).

The following should be considered when selecting sanitizing agents:

- The material of the surface that is to be sanitized;

- The contact time of the sanitizing or antimicrobial treatment with the surface being treated;

- Characteristics of water (e.g., hardness, pH, temperature) used to prepare the sanitizing agent;

- The compatibility of the sanitizing agent with the cleaning chemical(s); and

- Appropriateness of the sanitizing agent for use on food or FCSs.

Consulting with cleaning and/or sanitizing agent suppliers, academia, industry associations, scientific literature, and government agencies may help you select the products that are best suited for your operation.

E. Verification of Cleaning and Sanitizing

You should verify the chemical concentration of the sanitizer(s) that you use each time you use them, with an appropriate test kit or analytical instrument. If analytical instruments are used to measure sanitizer concentrations, they must be accurate and precise as necessary and appropriate in keeping with their purpose (§ 112.124(a)), adequately maintained (§ 112.124(b)), and adequate in number for their designated use (§ 112.124(c)).

To verify the effectiveness of your cleaning and sanitizing procedures, we recommend that you use at least one, or more, of the available test methods. Some of these tests, which include bioluminescence, adenosine triphosphate (ATP), and protein-based technologies, provide rapid results and allow for follow-up or intensified cleaning and sanitizing activities if results are above your established thresholds. However, it is critical to recognize that none of these tests is an appropriate substitute for environmental monitoring of *Listeria* spp. or *L. monocytogenes*, as required for sprout operations by § 112.145 (see Section VI (Environmental Monitoring)).

F. Cleaning and Sanitizing Conducted in Response to Suspected or Known Contamination

In addition to the cleaning and sanitizing required by §§ 112.123(d) and 112.143(b), you must clean and sanitize FCSs and the production environment as a corrective action measure if:

- Sprouts or spent sprout irrigation water test positive for the pathogens *E. coli* O157:H7, *Salmonella*, or any pathogens meeting the criteria in § 112.144(c). In such a situation, you must clean and sanitize the affected surfaces and surrounding areas (§ 112.148(c)); or

- *Listeria* spp. or *L. monocytogenes* is detected in the growing, harvesting, packing, or holding environment. In such a situation, you must clean and sanitize the affected surfaces and surrounding areas (§ 112.146(b)).

Further, you should clean and sanitize FCSs and the production environment as a corrective action measure when:

- You have used a lot of seeds that you have been subsequently notified has tested positive for a pathogen or has been implicated in a foodborne illness outbreak;

- Your sprout operation is involved in a foodborne illness outbreak; or

- You become aware of an occurrence of known or suspected contamination of your sprouts or sprout operation.

In such situations you should consider the following actions as a means of “intensified” cleaning and sanitizing:

- Dismantling equipment further than is described in your cleaning and sanitizing procedures or is your normal practice;

- Intensified (e.g., more vigorous, longer duration) scrubbing of surfaces;
- Identifying, cleaning, and sanitizing possible harborage sites for pathogens and possible cross-contamination routes;
- Soaking of equipment parts in an appropriate sanitizing agent for an appropriate duration;
- Increased frequency of cleaning and sanitizing activities for all surfaces that are not already cleaned or sanitized daily;
- Heat treatment of equipment or parts, as appropriate;
- Replacement or repair of tools, equipment, or production operation surfaces, as needed.

Additional information on corrective action measures required and/or recommended in response either to positive pathogen results in spent sprout irrigation water or sprouts (§ 112.148), or to findings of *Listeria* spp. or *Listeria monocytogenes* in your sprout production environment (§ 112.146) is discussed in Section V (Sampling and Testing of Spent Sprout Irrigation Water (or In-Process Sprouts)) in the draft re-issued sections (Ref. 18) and Section VI (Environmental Monitoring) and Section V (Seeds for Sprouting) in this document.

IV. Agricultural Water in Sprout Operations

In the Produce Safety Rule, we define "agricultural water" as "water used in covered activities on covered produce where water is intended to, or is likely to, contact covered produce or food contact surfaces, including water used in growing activities (including irrigation water applied using direct water application methods, water used for preparing crop sprays, and water used for growing sprouts) and in harvesting, packing, and holding activities (including water used for washing or cooling harvested produce and water used for preventing dehydration of covered produce)" (§ 112.3).

Several uses of water typical to sprouting operations meet the definition of "agricultural water," including water used to:

- Irrigate sprouts;
- Prepare ice that will contact sprouts;
- Wash finished product;
- Wash hands during and after harvest activities; and
- Contact food contact surfaces (FCSs) (including water used to prepare seed treatments, or to rinse, wash, or soak seeds prior to sprouting).

As stated above, "food contact surfaces" means "those surfaces that contact human food and those surfaces from which drainage, or other transfer, onto the food or onto surfaces that contact the food ordinarily occurs during the normal course of operations. 'Food contact surfaces'

includes food contact surfaces of equipment and tools used during harvest, packing, and holding" (§ 112.3). "Food contact surfaces" also includes FCSs of equipment and tools used in growing sprouts, such as those that contact seeds for sprouting (see § 112.143(b)). While seeds used to grow sprouts are not themselves "covered produce," seeds for sprouting are considered "food" under Section 201(f) of FD&C Act and as defined in § 112.3. Requirements relating to "food contact surfaces" in part 112 apply not only to surfaces that contact (or drain onto, or otherwise transfer onto) sprouts themselves but also to those that contact seeds used to grow sprouts. In a sprouting operation, FCSs include, for example: trays or drums used for sprouting; interior surfaces of containers used for seed rinsing, seed treatment, and pre-germination seed soaking; and counters that come into contact with sprouts during packing and/or packaging.

It is possible that not all of your water uses meet the definition of "agricultural water." For example, water that you use to wash the exterior of your delivery vehicle would not be considered "agricultural water" (assuming that the vehicle exterior does not contact the sprouts, and water from the exterior of the vehicle does not drain or transfer to the sprouts).

This section of the guidance is intended to help you understand and comply with some of the agricultural water requirements of the Produce Safety Rule, where water is intended or likely to contact sprouts and FCSs. Here, we provide a limited discussion of only certain provisions of part 112 related to agricultural water as they relate to sprout production, most notably the requirements in Subpart E that agricultural water must be safe and of adequate sanitary quality for its intended use (§ 112.41); the numerical microbial quality criterion that is relevant to sprouting operations (§ 112.44(a)); and the related requirements for agricultural water testing frequency (§ 112.46).[2]

A. Numerical Microbial Quality Criterion for Agricultural Water Used in a Sprouting Operation (§ 112.44(a))

Certain uses of "agricultural water" (including water used in growing sprouts) are subject to the microbial water quality requirement of no detectable generic *E. coli* per 100 mL prescribed by § 112.44(a). We are not aware of any commonly used agricultural water applications in sprouting operations that are not subject to this microbial water quality requirement. Agricultural water uses in sprouting operations that are subject to this standard include instances where water is:

- Used as sprout irrigation water (§ 112.44(a)(1));
- Applied in any manner that directly contacts covered produce during or after harvest activities (e.g., water that is applied to covered produce for washing or cooling activities, such as at entry to a wash tank, and water that is applied to harvested crops to prevent dehydration before cooling), including when used to make ice that directly contacts covered produce during or after harvest activities (§ 112.44(a)(2));

[2] In the *Federal Register* on December 6, 2021, we issued a proposed rule that, if finalized, would amend certain pre-harvest agricultural requirements for covered produce (other than sprouts) (86 FR 69120). To ensure that interested parties can readily view the proposed pre-harvest agricultural water revisions, we also proposed to reorganize and replace subpart E in its entirety. Of note, this proposed rule would not substantively alter the standards established in part 112, subpart E, for agricultural water used for sprouts, for which the compliance dates have passed, or for agricultural water used during harvesting, packing, and holding activities, or for treatment of agricultural water.

- Used to contact FCSs of equipment and tools used in growing, harvesting, packing and holding activities of sprouts (see § 112.143(b)), or to make ice that will contact FCSs (§ 112.44(a)(3)); and

- Used for washing hands (i.e., during and after harvest activities (§ 112.44(a)(4)), and supplied to hand-washing facilities at sprouting operations, including water used for hand-washing during growing activities (see § 112.130(a) and (b)(2); see also § 112.143(a))).

For sprout operations, § 112.143(a) requires that growing (as well as harvesting, packing, and holding) take place in a fully-enclosed building. Sections 112.130(a) and (b)(2) require that, for growing that takes place in a fully-enclosed building, adequate and readily accessible hand-washing facilities must be provided and must be furnished with water that satisfies the § 112.44(a) microbial quality criterion. The rule requires that water used for hand washing during all phases of sprout operations must meet the § 112.44(a) microbial quality criterion.

Sprout operations use different types of growing units, such as rotating drums, bins, tanks, and racks of trays. These growing units may use different methods to irrigate the sprouts, such as overhead intermittent sprays. Water for all of the irrigation methods used to irrigate sprouts must meet the microbial quality criterion in § 112.44(a), and related requirements elsewhere in Subpart E (§ 112.44(a)(1)).

After sprouts are removed from the growing unit, some sprout operations wash sprouts with cool water to remove hulls and/or to help lower the temperature of the sprouts. Some operations use a bubbling water bath to loosen and float off hulls. Occasionally, sprouts are washed in recirculating water in a flume system. The water used in all of these applications must meet the microbial quality criterion in § 112.44(a), and related requirements elsewhere in Subpart E. There are also additional requirements at § 112.48 for maintaining and monitoring the quality of water used during harvest, packing, and holding activities.

Ice may also be used during packing and shipping. Water used to make such ice must meet the standard prescribed by § 112.44(a)(3). If using ice, you should take steps, such as physical separation, to prevent melting ice from spreading from one production batch of sprouts to other production batches of sprouts, or from other products to sprouts, as it may serve as a source or route of contamination. Additionally, water used for cleaning FCSs used for packing and holding sprouts must meet the standard in § 112.44(a)(3).

B. Agricultural Water Source Type and Treatment Status

You must identify the type(s) of agricultural water source(s) used in your operation (e.g., ground water, water from a public water supply), and must inspect your agricultural water sources and distribution systems, to the extent that they are under your control, to identify conditions that are reasonably likely to introduce known or reasonably foreseeable hazards into or onto covered produce or FCSs in light of your covered produce, practices, and conditions (§ 112.42(a)). You must adequately maintain these systems (§ 112.42(b)).

Understanding the types of agricultural water sources that you use will help you to determine which requirements of Subpart E are applicable. As explained above, we are not aware of any uses of agricultural water (as defined in § 112.3) common in sprouting operations that are not subject to the microbial quality requirement in § 112.44(a). Therefore, our discussion below focuses on § 112.44(a) and related provisions.

You may not use untreated surface water for purposes covered by § 112.44(a), such as for sprout irrigation water (see § 112.44(a)). The definition of "surface water" appears in § 112.3 and includes, for example, water from rivers.

You may use untreated ground water for purposes covered by § 112.44(a). The definition of "ground water" appears in § 112.3 and includes, for example, water from wells that does not meet the definition of "surface water" (e.g., wells that are not influenced by surface water). In addition to all other applicable requirements of Subpart E that apply to agricultural water generally, if you use untreated ground water for purposes covered by § 112.44(a), you must:

- Sample and test water from each untreated ground water source used for such purposes, at the frequency established in § 112.46(c), and in compliance with the methodology requirements established for both sampling and testing in §§ 112.47(b) and 112.151 (see also Section IV. E (Agricultural Water Testing Frequency, Sampling, and Test Methods) below);

- If the microbial quality criterion (no detectable generic *E. coli* in 100 mL) is not met, immediately discontinue using water from the affected water source and/or distribution system for any purpose covered by § 112.44(a), and take appropriate corrective measures before using the affected water source and/or distribution system again for any such purpose (§ 112.45(a)); and

- Maintain records related to agricultural water testing, corrective actions, and test methodology, as required by §§ 112.50(b)(2), (6), and (9). For example, if you determine that water you use for a purpose listed in § 112.44(a) does not meet the microbial quality criterion established in that section, § 112.45(a) requires that you take certain steps as a result, and § 112.50(b)(6) requires you to keep records documenting the steps that you took.

You may use treated water (i.e., water that is treated in accordance with § 112.43) for purposes covered by §112.44(a). This is true regardless of the source from which the water was taken. For example, you may use water that originated from a surface water source and has been subsequently treated in accordance with the requirements of § 112.43. Water that has been treated in accordance with the requirements of § 112.43 is not required to be tested to ensure compliance with the microbial quality criterion (§ 112.46(a)(3)). In addition to all other applicable requirements of Subpart E that apply to use of agricultural water generally by sprout operations, if you use treated water for purposes covered by § 112.44(a), you must:

- Comply with all applicable requirements for treating water (e.g., the treatment must be effective to achieve the no detectable generic *E. coli* per 100 mL microbial quality criterion (§ 112.43(a)(1)), delivered in a manner that ensures it consistently meets that

criterion (§ 112.43(a)(2)), and monitored at a frequency adequate to ensure it consistently meets that criterion (§ 112.43(b)). For example, an effective monitoring program when using a chemical treatment method would measure the level of active compound as well as those factors that may affect its activity, such as pH, temperature, and contact time;

- Establish and keep documentation of scientific data or information you relied on to support the adequacy of the method you used to satisfy the requirements of §§ 112.43(a)(1) and (a)(2) for treating agricultural water (§ 112.50(b)(3)); and

- Maintain records related to such treatment (§ 112.50(b)(4)).

You also may use water from a public water system or supply as described in § 112.46(a)(1) and § 112.46(a)(2) for purposes covered by §112.44(a). Such water is not required to be tested to ensure compliance with the microbial quality criterion (§§ 112.46(a)(1) and (2)). In addition to all other applicable requirements of Subpart E that apply to agricultural water generally, if you use water from a public water supply for purposes covered by § 112.44(a), you must:

- Maintain annual documentation of the results or certificates of compliance from the public water system or supply that demonstrate that the water meets the microbial quality criterion of § 112.44(a) (i.e., no detectable generic *E. coli* per 100 mL) (§ 112.50(b)(7)).

If you hold water received from a public water system or supply in a covered tank indoors, or otherwise protected from potential contamination from the environment, it can continue to be considered public water and the applicable requirements from Subpart E described above apply.

If you hold water received from a public water system or supply in a pond, an uncovered tank outdoors where it is open to the atmosphere and therefore exposed to potential contamination in a manner similar to the exposure of other surface water sources, the water becomes subject to the requirements and restrictions related to a "surface water" source. For example, the prohibition from using untreated surface water for uses covered by § 112.44(a) would apply to such water.

If you use water received from a public water system or supply to recharge a well or otherwise expose the water to potential contamination in a manner similar to that of ground water sources, it becomes subject to the requirements related to a "ground water" source. For example, the testing requirements in § 112.46(c) applicable to untreated ground water would apply to such water.

C. Safe and of Adequate Sanitary Quality for Its Intended Use (§ 112.41)

In addition to the specific numerical microbial water quality criterion in § 112.44(a) that applies to sprout operations as discussed above, a general agricultural water quality requirement applies. Section 112.41 requires that all agricultural water must be safe and of adequate sanitary quality for its intended use. The principle of "safe and of adequate sanitary quality for its intended use" contains elements related both to the attributes of the source water and how it will be used.

A test result indicating the agricultural water does not meet the applicable microbial water quality requirement in § 112.44(a) demonstrates that the water is not safe or of adequate sanitary

quality for use as agricultural water in a sprout operation. However, the converse is not necessarily true. That is, agricultural water that was tested and found to meet the § 112.44(a) microbial quality criterion may not be safe or of adequate sanitary quality for such use if, for example, pathogens are present in the water. For example, when conducting a routine inspection, under § 112.42, of a well that you use as a source of untreated ground water for sprout irrigation, you might find a dead animal in the well. You would now have reason to believe that the agricultural water from that well is not safe or of adequate sanitary quality for its intended use. If you have determined, or have reason to believe, that your agricultural water source is not safe or of adequate sanitary quality for its intended use as required by § 112.41, you must immediately discontinue that use(s), and before you may use the agricultural water source and/or distribution system again for that use(s), you must take appropriate corrective actions, as set forth in § 112.45(a).

D. Reuse of Sprout Irrigation Water

We recommend against re-using the spent sprout irrigation water collected from one production batch to irrigate subsequent batches of sprouts (e.g., applying spent water from batch 1 to irrigate batch 2) without any form of water treatment in between batches. The conditions that are used to produce sprouts allow microorganisms, including those of public health significance, to grow. Using spent sprout irrigation water from one production batch of sprouts to irrigate another production batch of sprouts can lead to cross contamination between batches, increasing the amount of product exposed to any contamination that may be present. Moreover, depending on the circumstances, such water may not be safe and of adequate sanitary quality for its intended use, in which case the use would be prohibited by § 112.41 (Ref. 31 and Ref. 42).

Separate from the requirements for agricultural water under Subpart E, we note that the requirements in Subpart M for testing spent sprout irrigation water for *Salmonella* spp., *E. coli* O157:H7 (and any other pathogen meeting the requirements of § 112.144(c)) apply to each individual production batch of sprouts (§ 112.147). Re-using spent sprout irrigation water from one production batch of sprouts to irrigate another production batch of sprouts does not relieve you of the requirement to conduct testing in accordance with § 112.147 for spent sprout irrigation water from each individual production batch of sprouts (see Section V (Sampling and Testing of Spent Sprout Irrigation Water (or In-Process Sprouts)) in the draft re-issued sections (Ref. 18)).

Similarly, we also recommend that you not reuse spent sprout irrigation water to irrigate the same production batch of sprouts over time without any form of water treatment in between uses. Reusing spent sprout irrigation water for subsequent irrigation of the same production batch of sprouts over time could reintroduce any pathogens present in the water into the growing sprouts, which could contaminate a greater proportion of a given batch of sprouts. Any pathogens that may be present in the reused spent sprout irrigation water could then be added to the growing sprouts and could multiply to higher numbers during sprouting (Ref. 43).

E. Agricultural Water Testing Frequency, Sampling, and Test Methods

You are required to test your agricultural water source(s) to ensure compliance with the microbial quality criterion in § 112.44(a) if you use untreated ground water as agricultural water

(see Section IV. B (Agricultural Water Source Type and Treatment Status) above). In this circumstance, you are initially required to test each such water source a minimum of four times throughout the growing season or over a period of one year (§112.46(c)). If all four of the initial samples tested meet the microbial quality criterion in § 112.44(a), you may test once annually thereafter. You must resume testing at least four times per growing season or year if any annual test fails to meet the microbial quality criterion in § 112.44(a).

Section 112.46(c) requires all such samples to be representative of the intended use(s). By "representative of the intended use(s)" we mean collected from a location, and at a time of year, such that the sample can reasonably be expected to represent the quality of the agricultural water as it is normally used. For example, if you normally use untreated ground water for agricultural water uses only from April through August of each year, you should collect samples during that part of the year so that the samples are representative of the intended use(s). You should assess your production schedule to determine an appropriate sampling scheme for collecting the four initial samples.

With respect to sampling location, sampling at the point of your actual use is ideal (e.g., sampling where water comes out of a faucet or hose in your building and enters your sprout growing unit to be used for irrigation). You may also sample at other points along the water distribution system, from the water source itself to the point of use, as long as there is no reasonably likely point of contamination in the water distribution system between your chosen sampling point and the point of use, such that the sample can reasonably be expected to represent the quality of the water when it is used. For example, if untreated ground water is drawn from a well that is not under your control, you might choose to collect samples from a point that is under your control, such as a valve on a pipe at the edge of your property line. As another example, if the well is under your control and the piping from the well head to the points of use is in good condition, you may choose to collect samples from the well head. In either case, such samples can be considered "representative of the intended use(s)" (with respect to the location of the samples), provided that there is no reasonably likely point of contamination in the water distribution system between your chosen sampling point and the point of use. Further, § 112.42(a) requires you to conduct an inspection of your agricultural water system, to the extent under your control, to identify conditions that are reasonably likely to introduce known or reasonably foreseeable hazards. This inspection is required at the beginning of a growing season, as appropriate, but at least annually. This inspection should assist you in selecting your sampling points by determining what sampling locations are, or are not, representative of your intended use(s). Section 112.50(b)(1) requires you to establish and keep records of the agricultural water system inspection findings required by § 112.42(a).

Agricultural water samples to be tested for compliance with the § 112.44(a) microbial quality criterion must be collected aseptically, in accordance with § 112.47(b). You must use sterile equipment and tools and aseptic technique when collecting samples. Note that cleaning and sanitizing of sampling equipment and tools may not result in their being sterile (See Appendix 1 (Aseptic Sampling)).

Agricultural water samples must be analyzed for compliance with the § 112.44(a) microbial quality criterion using a method set forth in § 112.151 (§ 112.47(b)). We reviewed publicly

available test methods for water and determined that EPA Method 1603 is appropriate for testing water quality for this purpose (§ 112.151(a)).

However, we recognize that other scientifically valid analytical methods may be available or may become available in the future. Therefore, we provide flexibility for you to use any other scientifically valid analytical method that is at least equivalent to the prescribed analytical method (i.e., EPA Method 1603) in accuracy, precision in detecting the relevant organism or indicator (i.e., generic *E. coli*) and precision in the relevant sample matrix (i.e., ground water), and sensitivity (§ 112.151(b)(1)). We have identified a number of testing methodologies that meet the requirements of § 112.151(b)(1), and have posted a list of these methods on our website. See "Equivalent Testing Methodologies for Agricultural Water" at https://www.fda.gov/food/laboratory-methods-food/equivalent-testing-methodology-agricultural-water.

Section 112.50(b)(9) requires you to establish and keep documentation of any analytical methods that you choose to use for agricultural water testing in lieu of the method that is incorporated by reference in § 112.151(a). We recommend that records established and kept in compliance with § 112.50(b)(9) include 1) name and identification number of the method validated by third-party methods validation organization, or 2) detailed analytical procedures, an explanation of why the alternate method is at least equivalent to the reference method (such as a scientific study, or data or information regarding the sensitivity, specificity, and precision of the alternate method compared to the reference method), and any other relevant information supporting the use of the alternate method. We note that you are not required to notify us or submit information about such methods of analysis for our review or approval prior to use.

You should choose a laboratory that is qualified to test agricultural water for generic *E. coli*. Testing is typically contracted to a third-party testing laboratory; however, testing may also be performed by a sprout operation's own laboratory (e.g., an operation's own "in-house" laboratory). Using an accredited laboratory (e.g., a laboratory accredited to International Organization for Standardization (ISO) Standard 17025) is one way to have confidence that the laboratory will provide reliable, accurate test results.

F. Post-Harvest Water Management

Section 112.48(a) requires you to manage the water used during harvest, packing, and holding activities for sprouts, as necessary, including by establishing and following water-change schedules for re-circulated water to maintain its safety and adequate sanitary quality and minimize the potential for contamination of covered produce and FCSs with known or reasonably foreseeable hazards. Section 112.48(b) requires you to visually monitor the quality of water that you use during harvest, packing, and holding activities. For example, as a sprout operation:

- You must visually monitor water used for washing and cooling sprouts for buildup of organic material (such as hulls and plant debris). We recommend against the reuse of untreated post-harvest water in multiple production batches of sprouts because of the potential risk associated with its use.

- You should consider using a water treatment system as part of managing your post-harvest water. If you choose to treat your water, those treatments should be applied in a manner that is consistent with § 112.43.

- The specific level and frequency of post-harvest water treatment, the point at which treatment should be applied, and the time intervals for monitoring treatments necessary to align with § 112.43 may vary. An effective monitoring program for use of a chemical treatment method should measure the level of active compound as well as those factors that may affect its activity, such as pH, temperature, contact time, and organic load. For example, adequate monitoring of water treated with hypochlorite in a postharvest wash for sprouts should include monitoring the level of active antimicrobial (free available chlorine) and pH, since hypochlorite activity is reduced both by organic material (e.g., soil, plant debris) and pH values outside its effective range (pH 6.0–7.5) (Ref. 44, Ref. 45, Ref. 46, and Ref. 47).

V. Seeds for Sprouting

The requirements in § 112.142 are designed to reduce the likelihood that seeds for sprouting serve as a vehicle for introducing contamination in sprouts. This section of the guidance provides recommendations to help you comply with these requirements for receiving, storing, and treating seeds for sprouting. As previously mentioned, throughout this guidance, for the ease of the reader, we often refer collectively to everything sprouted to produce sprouts for human consumption, including beans, simply as "seeds." In the Rule, we used the phrase "seeds or beans" to remove any potential confusion as to whether beans for sprouting were included. References to "seeds" in this guidance should not be read to exclude other things that are sprouted to produce sprouts for human consumption, such as beans or nuts.

While poor sanitation and unhygienic practices at the sprout operation can contribute to contamination of sprouts, studies indicate that contaminated seed is the likely source of most sprout-related outbreaks (Ref. 2, Ref. 11, Ref. 48, and Ref. 49). Seed contamination can occur at various points in the supply chain, including the seed farm, seed conditioner, seed supplier, transportation, or the sprout operation. The sprout growing process provides opportunities for contamination that may be present in or on a few seeds to spread through the entire sprout production batch.

Only a small portion of seed produced in the United States is destined for sprouting, while seed is more frequently used as planting stock for forages for livestock or for field cultivation of human food (Ref. 2). Although we encourage you to purchase seeds that were produced specifically for sprouting and grown under Good Agricultural Practices (GAPs) (i.e., FDA's guidance titled "Reducing Microbial Food Safety Hazards in the Production of Seed for Sprouting") (Ref. 17), we are aware that this does not always occur. For this reason, seeds that you use for sprouting may be grown, milled, and/or stored under conditions where contamination is likely to occur. Seed may become contaminated by potential sources of fecal contamination, such as contaminated water, inadequately treated or raw manure used as fertilizer, wild or domesticated animals, unclean equipment, or inadequate worker hygiene. These conditions may contribute to the presence of human pathogens on seed for sprouting.

The steps of sorting, cleaning, storing, packaging, and shipping of seeds by seed mills may spread contamination between seed lots or within a seed lot, especially if GAPs are not followed. During these steps, seeds from multiple lots of different origins may be mixed together, providing opportunity for cross contamination between lots to occur, and complicating any corrective actions and/or traceback of the origin of the lots should they be involved in a contamination event or illness (Ref. 2). Use of unclean equipment may introduce contamination to a seed lot or spread contamination to other seed lots if the same equipment is used with multiple lots and not cleaned in between uses with different seed lots. Damage to the seeds during these steps may also affect the overall safety of the seeds; such damage may occur inadvertently or deliberately (i.e., scarification). Cracks, crevices, and other types of damage to seeds may facilitate the internalization of pathogens into seeds and make it difficult to remove pathogens during subsequent treatment and handling (Ref. 2).

A. Seed Sourcing, Receiving, Handling and Storage

1. Seed Sourcing

You must take measures reasonably necessary to prevent the introduction of known or reasonably foreseeable hazards into or onto seeds that you will use for sprouting (§ 112.142(a)). We are aware that some sprout seed suppliers seek written assurances from their seed growers and handlers that their seeds were produced under GAPs and handled according to food safety best practices throughout harvesting, conditioning, storage, and transportation. Sprout operations can request this information from the entity which sells their seeds. While not requirements of the Produce Safety Rule, we recommend these practices as prudent when sourcing the seeds used for sprouting. In addition, we are aware that production of seed for sprouting under GAPs is required by some foreign governments (Ref. 21) and is accepted as an international food safety best practice (Ref. 19). We have published guidance titled, "Reducing Microbial Food Safety Hazards in the Production of Seed for Sprouting" (Ref. 17) which recommends GAPs for seed growers and handlers to produce seed for sprouting in accordance with food safety practices. We recommend that you source seeds for sprouting that have been grown, packed, and held according to this guidance.

Knowing your seed supplier(s) and establishing specifications for the seeds you receive (such as being grown under GAPs and handled under sanitary conditions during storage and distribution or transport) can help improve the safety of the seeds you receive. Specifications also provide standards against which you can assess the acceptability of the seeds you receive for the production of finished sprouts.

We further recommend having a proactive discussion with your seed supplier to learn what its practices are in the event that a portion of a master lot of seed tests positive for a pathogen prior to distribution (e.g., does the supplier divert the entire "master lot" to a use other than seeds for sprouting?). You should also ask whether seed from different sources is generally commingled before a master lot is broken down into sub-lots, to determine the relevance of any future positive test results on multiple sub-lots. We also recommend having a discussion with your seed supplier about how you should notify them of any positive test results you receive (e.g., of spent sprout irrigation water as required under § 112.142(b)(2)), including what information they would need and under what circumstances they would accept a return of a seed lot that had tested

positive for a pathogen (e.g., results of confirmatory testing). It is prudent to obtain this information up front, rather than waiting to obtain it after receiving a positive test result. We also recommend you ask the seed supplier what actions they take when notified of contamination (e.g., by regulators or other sprout operations) associated with a seed lot that has already entered commerce.

2. Seed receiving

We recommend you develop a seed receiving program setting out the standard operating procedures (SOPs) you follow to receive and inspect seed upon receipt and the criteria you use in determining whether to accept a shipment. The seed receiving SOP should describe the objectives, description of procedures for receiving seeds, including parameters that should be monitored or verified (e.g., in the form of a checklist), documentation that should be received, who is responsible for each task, materials or supplies needed (e.g., black light), and actions to be taken in the event that non-conformities are identified. An example of a seed receiving and inspection checklist can be found in the next section (Section V. A. 3 (Visual Inspection of Seeds and Their Packaging)). Your seed receiving program should include specific procedures for how you will handle any issues identified while inspecting a shipment of seed.

Once seeds arrive at your operation, you should verify that the transport vehicle is clean and sanitary. You should also check the seed tag, package labeling and other documentation to ensure that the seeds are what you ordered (e.g., they match the information on the purchase order), that they are from a known supplier, that they meet any specifications you may have established with your seed supplier, and that any information or documentation that you plan to keep for your own records (such as documentation of the seed lot number, or of prior treatment of seed) has been provided. If requested information or documentation pertinent to the seed lot is missing, you may consider following up with your seed supplier to obtain that information. If a shipment does not meet your requirements, you should evaluate the deviation(s) and, based on your criteria for accepting seeds, determine whether the safety of the seed may be compromised and whether the lot should be rejected. You should be able to account for lots rejected due to evidence of contamination or for other reasons (e.g., in the event of a future recall).

You should not use seed that is labeled “Not for Human Consumption” for growing sprouts for human consumption, as these seeds may have been grown, handled, or treated in such a way that makes them inappropriate for human food. As you are sourcing seed, we recommend asking your seed supplier if the lot of seed you are purchasing has tested positive for a pathogen or if the seed lot came from a larger master lot, and if so, whether any test done on any other part of that master lot was positive for a pathogen. We recommend against using seed from sub-lots of a master lot that has tested positive for a pathogen unless you have information that the source of the contamination to the part of the lot that tested positive is not applicable to your sub-lot (see also Section V. A. 4 (Seed Testing), below, for a discussion of § 112.142(c)). This is because of the likelihood that other portions of the master lot could also be contaminated, and the limitations of seed testing methods to detect sporadic contamination.

3. Visual inspection of seeds and their packaging

You must visually examine seeds, and packaging used to ship seeds, for signs of potential contamination with known or reasonably foreseeable hazards (§ 112.142(d)). Each bag should be examined for physical damage that could result in contamination with such hazards (e.g., holes from rodents) and other signs of contamination (e.g., stains, insects, presence of insect, bird, or rodent feces, presence of rodent urine, or foreign material) upon arrival. This can be as simple as using a black light in a dark room to examine seed packaging for signs of rodent urine, and visually examining the seeds and packaging for rodent feces, rodent gnawed holes, or excessive dirt, debris, and damage. In addition to looking for signs of rodent damage, you should also look for damaged seeds. Damage to seeds can provide small, hard to reach places for pathogens to lodge in and make any treatment to reduce pathogens less effective. We also recommend that if you store the seeds for any length of time after they arrive at your operation, you should re-inspect the bags of seeds for signs of contamination before using them for sprout production.

Procedures for receiving and inspecting seed (e.g., that could be included in a seed receiving and inspection checklist) should include the following information included in Table 2.

Table 2. Suggested Procedures for Receiving and Inspecting Seed

Check the transportation vehicle for conditions that could lead to contamination;
Verify supplier name and address on packages to see if they match those on the purchase order;
Check date of shipping and volume of products shipped;
Verify conformance to your seed specifications (e.g., seed type, origin, grown in accordance with GAPs for seeds (i.e., FDA's guidance titled "Reducing Microbial Food Safety Hazards in the Production of Seed for Sprouting") (Ref. 17), prior seed treatment);
Check lot information was provided (e.g., lot size, lot number, master lot number (if applicable), name of establishments from which seed originated as well as establishments that handled seed, harvest date);
Check condition of bags of seed (e.g., water damage, off odor, presence of insect, bird, or rodent feces, presence of rodent urine using a blacklight, mechanical damage, mold), ensuring that packaging does not allow for contamination and is intact;
Check seed testing record, or letter of guarantee or certificate of conformance for such testing, if available, including lot number tested, name of laboratory, test results, and details of tests performed (e.g., test method used, information to support scientific validity of test method, date tested);
Check seed treatment record, or letter of guarantee or certificate of conformance for such treatment, if available; and
Account for lots rejected due to evidence of contamination or for other reasons.

If you observe obvious signs of seed contamination upon receipt and you conclude the contamination occurred prior to the seed arriving at your sprout operation, you should report the information to the seed grower, distributor, supplier, or other entity from whom you received the seed, and return the shipment to the supplier. You should also discontinue use of the affected part of the seed lot, and we recommend that you conduct intensified cleaning and sanitizing of any surfaces that may have become contaminated by the seed lot (see Section III (Cleaning and Sanitizing)) and take any other actions necessary to prevent reoccurrence of contamination. If you know or have reason to believe that a lot of incoming seeds may be contaminated with a pathogen, either because of a microbial test result or because it has been associated with foodborne illness, there are steps that you must take (§ 112.142(b) and (c)), and you must document those actions as required under §112.150(b)(6). These steps are discussed in the section of this guidance document, Section V. C (Corrective Actions for Seeds That May Be Contaminated with a Pathogen).

4. Seed testing

The Produce Safety Rule does not require microbial testing of seeds for sprouting by you or your supplier. However, if you or your supplier choose to conduct seed testing, we recommend that you use a sampling plan that results in a sample that is representative of the entire lot of seed, collect the sample aseptically, and test the sample using a scientifically valid method. Because microbial contamination in seeds, when present, is often at low levels and not uniformly distributed throughout a seed lot, such contamination is often difficult to detect (Ref. 2). If you or your seed supplier test a lot of seeds for pathogens, you should be aware that the absence of a positive test result for pathogens does not mean that the lot of seeds is necessarily pathogen-free.

A positive test result for a pathogen indicates that the lot of seeds is contaminated. In such a case, you must take certain actions under § 112.142(b), including discontinuing use of that seed lot for sprout production, except as provided for in certain limited circumstances in § 112.142(c), and reporting the information to the seed grower, distributor, supplier, or other entity from whom you received the seeds. One such circumstance is if you treat the lot of seeds with a process that is reasonably certain to achieve destruction or elimination of the most resistant microorganisms of public health significance that are likely to occur in the seeds (§ 112.142(c)(1)). We understand that some in the food industry may refer to such a treatment as a “pasteurization step.” However, we note that processes that meet the description in § 112.142(c)(1) are not currently commonly used in the sprouting industry, in part, because such processes would be likely to impact the germination ability of the seed. Such processes are far more robust than seed treatments described in § 112.142(e), which typically only reduce the numbers of microorganisms of public health significance, rather than eliminate them. For more information, see the seed treatment section (Section V.B (Seed Treatment)) of this document below. If you choose to conduct such a treatment (i.e., “pasteurization step”) on your seed lot so that you may use those seeds for sprouting, we note that you must still comply with the requirement in § 112.142(b)(2) to report the positive test result to your seed grower, distributor, or supplier.

You are not required to take the steps set forth in § 112.142(b)(1) and (2) if you later reasonably determine, through appropriate follow-up actions, that the lot of seeds is not the source of

contamination (§ 112.142(c)(2)). However, we consider that there are very few circumstances in which it would be appropriate to apply the exception at § 112.142(c)(2) in the event there has been a positive pathogen test result in an incoming lot of seed.

It is also important to note that if a positive pathogen test result is obtained from a seed lot, re-testing a new sample from the same seed lot and obtaining a negative pathogen test result does not negate the previous positive test result. In such a case, the required follow-up actions must still be taken.

5. Seed storage

Seeds can become contaminated during storage. Storing seed for sprouting is considered part of the process of growing sprouts, and therefore the building in which your seed is stored is subject to the applicable requirements for buildings in Subpart L (§ 112.122(a)). Further, seed for sprouting must be stored in a fully-enclosed building (§112.143(a)), which should be of sound construction and in good repair. You must take measures reasonably necessary to prevent the introduction of known or reasonably foreseeable hazards into or onto seeds that you will use for sprouting (§ 112.142(a)). Many pests are attracted to seeds, and these pests can serve as a source of, or vector for, spreading contamination, especially when seeds are in storage. In order to prevent the introduction of known or reasonably foreseeable hazards into or onto seeds that will be used for sprouting, you should store seeds in a protected manner. Your seed storage area should be clean, dry, and dedicated to seed storage. You must take measures to exclude pests from the building (§§ 112.143(a), 112.126, and 112.128(b), and see Section IV (Equipment, Tools, and Buildings) in the draft re-issued sections (Ref. 18)) and you must routinely monitor for pests, as necessary and appropriate (§ 112.128, and see Section IV (Equipment, Tools, and Buildings) in the draft re-issued sections (Ref. 18)). You should store seeds off the floor and away from walls to facilitate pest control inspection. You should regularly inspect seed packaging and containers (see Section IV (Equipment, Tools, and Buildings) in the draft re-issued sections (Ref. 18)).

Once the original seed packaging is opened, the remaining seed must be stored in closed containers with tight-fitting lids or otherwise protected from contamination (§ 112.142(a)). If you use containers other than the original packaging to hold seeds, their food contact surfaces (FCSs) must be cleaned and sanitized prior to use (see § 112.143(b)). Containers should also be labeled to maintain the identity of the seed lot they contain. If the seeds have received prior antimicrobial treatment by a grower, supplier or distributor, this should be indicated on the label.

If you intend to rely solely on a seed treatment conducted by an entity earlier in the supply chain (e.g., you will not re-treat the seeds yourself), you must store seeds that have received that prior treatment by the seed grower, distributor, or supplier under conditions that will prevent them from becoming contaminated (see § 112.142(e)(2)(ii)). We recommend that seeds that received prior treatment be stored separately from untreated seeds, in airtight containers/packages that are as small as practicable to minimize the number of times any particular container/package is opened and closed to remove seed from that container (because each such event presents the possibility of contamination). If you reuse containers/packaging, you must clean and sanitize the FCSs on these containers before contact with seeds used to grow sprouts (§ 112.143(b)).

B. Seed Treatment

Research indicates that seed contamination, when it occurs, may be at low levels, intermittent, and unequally distributed within seed lots (Ref. 2). However, even low levels of pathogens on seed for sprouting are a concern, because the conditions present during sprouting are ideal to support the growth of pathogens. There is some evidence that sprout operations associated with outbreaks often did not apply seed treatments correctly, consistently, or at all in the production of sprouts associated with the outbreak (Ref. 2). While treating seeds used for sprouting does not guarantee pathogen-free sprouts, seed treatment has been shown to reduce the percentage of contaminated batches of sprouts (Ref. 50, Ref. 51, Ref. 52, Ref. 53, Ref. 54, and Ref. 55). For example, a quantitative risk assessment model predicts that, without any seed treatment or spent sprout irrigation water testing, 5.22% of sprout batches will be contaminated with *Salmonella* (Ref. 55). Following seed treatment, the predicted fraction of contaminated sprout batches was reduced to 2.32%, 0.0320%, and 0.000320% when 1-log, 3-log, or 5-log seed treatment was applied, respectively. When seed treatment was combined with spent sprout irrigation water testing, the fraction of positive sprout batches predicted by this model was reduced further. (This risk assessment is discussed in more detail below.) Therefore, seed treatment is a critical part of a multi-hurdle approach to reduce the public health risks associated with the consumption of sprouts.

Section 112.142(e) requires that the seeds you use to grow sprouts be treated using a scientifically valid method to reduce microorganisms of public health significance. We use the term "scientifically valid" to mean an approach that is based on scientific information, data, or results published in, for example, scientific journals, references, textbooks, or proprietary research. We note that the term "scientifically valid" has a different meaning than "validated" with regard to seed treatment, for the purposes of the Produce Safety Rule. The term "validation study" is often used to refer to a systematic process of obtaining and evaluating scientific and technical evidence that a control measure or a combination of control measures is capable of effectively controlling the identified hazard(s). The Produce Safety Rule does not require you to perform a validation study to evaluate a seed treatment; however, we note that a validation study can help to evaluate whether a particular treatment is "scientifically valid".

To meet the seed treatment requirement, you must adopt one of the following two approaches:

- Treat seeds that you use to grow sprouts using a scientifically valid method to reduce microorganisms of public health significance (§ 112.142(e)(1)). If you perform treatment of seeds used for sprouting in accordance with § 112.142(e)(1), you must establish and keep documentation of this treatment (§112.150(b)(1)); or

- Alternatively, you may rely on prior treatment of seeds conducted by a grower, distributor, or supplier of the seeds (whether to fulfill this requirement completely or for the purpose of you considering such prior treatment when applying appropriate additional treatment of the seeds at your operation immediately before sprouting), provided that you obtain documentation (such as a Certificate of Conformance) from the grower, distributor, or supplier that: (i) the prior treatment was conducted using a scientifically valid method to reduce microorganisms of public health significance; and (ii) the treated

seeds were handled and packaged following the treatment in a manner that minimizes the potential for contamination (§ 112.142(e)(2)). Such records should include documentation of the scientifically valid method used to treat the seeds, references to scientific data supporting the method, the level of log reduction achieved, the type of seeds used for any validation study that may have been done, the pathogens targeted, and information on how the seeds were handled and packaged following the treatment to minimize the potential for contamination (e.g., seeds were packaged immediately after treatment, type of packaging).

1. Choosing a seed treatment

A successful seed treatment reduces microbial pathogens while preserving seed viability, germination, and vigor. Seed types can vary in sensitivity to antimicrobial agents and other types of treatments, which can affect how well the seeds germinate and grow after treatment. Additionally, the varying surface features of different types of seeds can influence how well a treatment can access and inactivate pathogens on or in the seed. Therefore, an antimicrobial treatment that is effective for one type of seed may not be as appropriate for other types.

When reviewing the options available for seed treatment, especially if you plan to treat seeds at your operation (as opposed to, or in addition to, purchasing pre-treated seeds), you should consider the feasibility of applying the treatment at your operation. For example, irradiation[3] is an option for seed treatment to reduce microorganisms of public health significance that may not be feasible for you to apply on-site. In addition, hot water treatments have been demonstrated to reduce pathogens on seeds by more than 5 log CFU/g in one study (Ref. 56) and to undetectable levels in another study (Ref. 57). However, these treatments can require use of equipment, such as industrial-sized hot water pasteurization machines (Ref. 58), that might be cost prohibitive for a small sprout operation.

Nothing in the Produce Safety Rule requires or authorizes you to take measures in conflict with existing federal, State, or local regulations. We expect any seed treatment that is used to be applied in accordance with all applicable federal, state, tribal, or local requirements. For example, sources of radiation used to treat food are considered food additives, under the law, and require an authorizing regulation by FDA. Also, in compliance with the Federal Insecticide, Fungicide and Rodenticide Act (FIFRA), pesticide chemicals used as seed treatments must be registered with the Environmental Protection Agency (EPA) and labeled for use to reduce microorganisms of public health significance on seeds for sprouting. Unlike pesticide chemicals, equipment that works to control microorganisms by physical means (e.g., heat) are classified by EPA as "pesticide devices" and do not require registration with EPA under FIFRA (Ref. 59). Note also that some States require registration of pesticide devices, and we refer you to the

[3] 21 CFR § 179.26(b)(10) allows seed for sprouting to be treated with ionizing radiation up to a maximum dose of 8 kGy. We note that the codified language in the regulation states "seeds," but for the purposes of the regulation, the FDA recognizes that beans for sprouting (e.g., mung bean seeds used for sprouting) would also be included. For information about the use of non-ionizing sources of radiation (such as UV light, radio frequency, microwaves, and pulsed light) we refer the reader to 21 CFR Part § 179 more generally. We note that while seeds that have been treated with ionizing radiation must be labeled with a radura symbol along with either the statement "Treated with radiation" or the statement "Treated by irradiation," sprouts that are grown using irradiated seeds need not be so labeled where the sprouts themselves have not been irradiated (see 21 CFR § 179.26(c)(2); 65 FR 64605, at 64606-7 (Oct. 30, 2000)).

appropriate State pesticide regulatory agency for more information on a particular State's requirements related to such devices (Ref. 60).

Rather than establishing a specific type or method of seed treatment that you must use to treat seeds, the Produce Safety Rule allows you to use any scientifically valid method to reduce microorganisms of public health significance on seeds that you use to grow sprouts (§ 112.142(e)). This approach provides you with flexibility in choosing a seed treatment. In choosing a treatment(s) appropriate to your operation and your sprouts, and in compliance with the Produce Safety Rule, there are a number of things to consider. In this guidance, we highlight certain treatments that have been studied in the literature and discuss important considerations related to using those treatments.

Known seed treatment methods include those that work by chemical means (e.g., liquid or gas), physical means (e.g., heat), or a combination of these. Based on a review of available literature, physical and combination type treatments have been reported to be the most effective for reducing the numbers of pathogens on seeds for sprouting. Physical treatments, such as heat (dry heat or hot water), high pressure, and irradiation are reported to have better penetration characteristics for reaching bacteria on microscopically rough surfaces, as well in as the interior of the seed, compared to chemical treatments (Ref. 61). In some studies, physical treatments have been reported to achieve a 5-log or greater reduction in pathogens on seeds (Ref. 58, Ref. 61, Ref. 62, and Ref. 63). Combination treatments (i.e., applying two or more methods sequentially or simultaneously) may be more effective than using a single treatment alone. Examples of such combination treatments include: chemical plus heat; irradiation plus dry heat; and high pressure plus dry heat. Literature suggests many combination treatments may be able to achieve a 5-log or greater reduction in pathogens on seeds (Ref. 58, Ref. 61, Ref. 62, Ref. 63, Ref. 64, Ref. 65, and Ref. 66).

Previous studies suggest that the following chemicals, when used at appropriate concentrations, may be able to achieve at least a 3-log reduction in pathogens on seeds: acidified sodium chlorite (Ref. 67); calcium hydroxide (Ref. 68), calcium hypochlorite[4] (Ref. 61); caprylic acid (Ref. 69); gaseous acetic acid (Ref. 70 and Ref. 71); hydrogen peroxide (Ref. 68 and Ref. 72); lactic acid (Ref. 65); monocaprylin (Ref. 69); oxalic acid (Ref. 65); phytic acid (Ref. 65); and sodium hypochlorite (Ref. 73). This is not intended to be an exhaustive list. We note, however, that such treatments require EPA approval and pesticide registration under FIFRA before they may be used for such purposes. We refer readers to EPA for further information on the registration process. Information regarding currently registered pesticide products is available on EPA's website at: https://iaspub.epa.gov/apex/pesticides/f?p=PPLS:1.

2. Seed treatment efficacy

Many seed treatments reduce, but do not eliminate, microorganisms of public health significance that may be present on the seeds. Pathogens that are not eliminated by seed treatment can be amplified during the sprouting process; therefore, adequate seed treatment prior to sprouting is a

[4] In our prior Sprout Guidances, we cited a 20,000 ppm calcium hypochlorite treatment as an example of a seed treatment. The EPA registration for this treatment has since been updated and we refer readers to EPA for further information on that registration.

key component of the multi-hurdle risk reduction framework for sprouts established in the Produce Safety Rule.

The use of seed treatment in a multi-hurdle risk reduction framework is supported by the findings of a 2018 quantitative risk assessment conducted by researchers at FDA's Center for Food Safety and Applied Nutrition (Ref. 55). They assessed the risk of salmonellosis associated with the consumption of alfalfa sprouts to evaluate the public health impact of treating seeds and testing spent sprout irrigation water for *Salmonella* cells washed off during production. If there were no interventions, the assessment estimates that there would be 76,000 (95% confidence interval (CI) 15,400-248,000)[5] cases of salmonellosis every year tied to the consumption of contaminated sprouts. The model indicates that the risk reduction from using a seed treatment that can achieve a 1-log reduction in *Salmonella* alone (i.e., without spent sprout irrigation water testing) is comparable to spent sprout irrigation water testing alone, and each additional 1-log reduction in *Salmonella* achieved by the seed treatment was predicted to provide additional reduction (i.e., greater than spent sprout irrigation water testing alone).

Among the approaches that the researchers evaluated, the one they found to have the greatest public health impact, is using a seed treatment that obtains a 5-log reduction of *Salmonella* combined with testing spent sprout irrigation water. Together, these two interventions would result in an estimated 99.9994% reduction in the number of salmonellosis cases per year – essentially no illnesses (95% CI <1-1.5). A 3-log reduction treatment in combination with effective spent sprout irrigation water testing (i.e., collecting and testing a sample that is representative of 100% of your batch of sprouts) would reduce the number of cases per year to 45 (95% CI 10-146). Both approaches provide substantial public health benefits. However, the researchers estimate that there would be hundreds, or thousands, of illnesses if spent sprout irrigation water is tested in combination with treatments that achieve less than a 3-log reduction. For example, testing spent sprout irrigation water with a treatment that can obtain a 1-log reduction of *Salmonella* is predicted to only reduce the number of cases per year to 3,560 (95% CI 821-11,400). Testing spent sprout irrigation water alone would only reduce the cases to an estimated 12,100 (95% CI 2,400-41,200) per year.

Based on the results of this risk assessment, we recommend the use of a seed treatment that achieves at least a 3-log reduction in microorganisms of public health significance. As more efficacious seed treatments become readily available on the market and registered for this use, it is likely that we will update this recommendation in the future. For more information on spent sprout irrigation water testing, see Section V (Sampling and Testing of Spent Sprout Irrigation Water (or In-Process Sprouts)) in the draft re-issued sections (Ref. 18).

We recommend that you use the most efficacious seed treatment available that is practical for your operation to reduce the presence of microorganisms of public health significance on seeds for sprouting, with primary consideration given to reduction of *Salmonella* spp. and *E. coli* O157:H7. You should not rely only on a treatment that achieves a low level of reduction if other, more effective, treatments that are practical for your operation are available. We understand that a 3-log reduction is the minimum level of reduction of pathogens the EPA will consider when

[5] The confidence interval (CI) numbers are the lower and upper range of the estimate. The estimates in this risk assessment do not factor in interventions that industry is currently conducting; they are designed to compare the impact of different approaches.

registering an antimicrobial treatment for seeds that includes a public health claim. Because using a treatment that provides a higher level of pathogen elimination before spent sprout irrigation water testing is undertaken is expected to reduce the number of production batches of sprouts that test positive for a pathogen (Ref. 55), such an approach would minimize the likelihood that you will invest time and resources to produce a production batch of sprouts that is later determined to be contaminated through routine testing required under § 112.147 (and which must be discarded as a result, as required by § 112.148).

The validity and efficacy of any treatment is dependent on the specific parameters governing how the treatment is applied. You should carefully monitor those variables that impact the overall efficacy of the treatment, including, for example, seed type, treatment concentration, treatment time, temperature, pH, pressure, radiation dose, seed-to-treatment solution ratio, pre- and post-treatment rinsing, and agitation (as appropriate to maximize contact between seed and the treatment solution or gas). You must document observations of treatment and key parameters monitored during preparation and application of treatments (e.g., chemical composition of treatment solution, heating temperature, equipment used, concentration of treatment solution, treatment time, pH of treatment solution, presence of agitation)) (§ 112.161)(a)(1)(ii)). If you are utilizing a seed treatment described in scientific literature, you should ensure that your use of the treatment is within the parameters used in the study. In other words, you should ensure that the conditions under which the treatment was applied in the study are compatible with how you will be applying the treatment at your operation. For example, if the treatment you are considering has only been tested on alfalfa seeds, and you plan to use the treatment on mung beans, you should not assume that the treatment will be equally effective at reducing microorganisms of public health significance on mung beans. You should, instead, obtain a study that was conducted on the type of seeds that you intend to treat.

The Produce Safety Rule does not prohibit the use of proprietary seed treatments (e.g., treatments developed based on a firm's own scientific research not published in scientific literature). However, your use of such a treatment method must meet the same requirements for scientific validity as must be met if you use a treatment based on published literature.

If you choose to treat seeds using a scientifically valid method to reduce microorganisms of public health significance at your operation (i.e., instead of relying on prior seed treatment applied by an entity earlier in the supply chain), you should develop a written seed treatment SOP as a best practice to help you improve the safety of your sprouts. The SOP should include:

- Objectives;
- Methods used;
- Who is responsible for each task;
- Materials needed;
- Treatment procedures (e.g., how to prepare, how to monitor, how and when to discard);
- Parameters to be measured or monitored; and

- Relevant records to be established and kept (in compliance with the requirements of § 112.150(b)(1) and otherwise).

3. Using pre-treated seeds

There are several seed treatment methods that can be effectively applied by a grower, handler, or distributor of seeds such that, when followed by good handling and packaging practices, they can eliminate the need for you to treat seeds at your operation immediately before sprouting. However, you may, nonetheless, decide to re-treat pre-treated seeds. Using pre-treated seeds has several benefits, including increased flexibility for you and availability of treatment options that may be suitable for your supplier but cost-prohibitive to you (e.g., irradiation). We note that if you choose to purchase seeds that have been pre-treated with ionizing radiation, if you re-treat those same seeds with ionizing radiation, the cumulative dose must not exceed the maximum dose allowed by § 179.26(b)(10).

If you choose to rely, in whole or in part, on using pre-treated seeds, you must obtain documentation from your seed supplier that the treatment was conducted using a scientifically valid method to reduce microorganisms of public health significance (§ 112.142(e)(2)(i)) and that the treated seeds were handled and packaged, following the treatment, in a manner that minimizes the potential for contamination (§ 112.142(e)(2)(ii)). If you choose to rely solely on using pre-treated seeds, you must obtain documents of this type from your supplier that are specific to each lot of seeds you receive from that supplier. One such type of document you may obtain is a Certificate of Conformance. Such a certificate (or other form of documentation used for this purpose) should include the name or brief description of the scientifically valid method used to treat the seeds, the level of log reduction that the method is capable of achieving, the type of seeds used for any relevant validation study, and the pathogen(s) targeted by the study. We recommend that you also ask your seed supplier for a written explanation of the treatment parameters that were applied, and the basis for their conclusion that the treatment is scientifically valid. In the event of an investigation or inspection of your sprout operation, we may ask to review the science supporting the seed treatment(s) you rely on, including proprietary treatment(s), to ensure it is scientifically valid.

If you intend to rely solely on a seed treatment conducted by an entity earlier in the supply chain (e.g., you will not re-treat the seeds yourself), you must store seeds that have received that prior treatment by the seed grower, distributor, or supplier under conditions that will prevent them from becoming contaminated (See § 112.142(e)(2)(ii) and above Section V. A. 5 (Seed Storage)).

4. Additional considerations for treating seeds for sprouting

a. Water Use

If you use water to prepare your seed treatment, the water is agricultural water and therefore must meet the microbial quality criteria in § 112.44(a) (i.e., no detectable generic *E. coli* in 100 mL water), and related requirements elsewhere in Subpart E. Similarly, if you rinse or soak seeds before, during, or after seed treatment, water used for these purposes must also meet this microbial quality criterion, and related requirements elsewhere in Subpart E, because such water

contacts FCSs (surfaces of the rinse or soak container that then contact seeds used for sprouting) (See § 112.44(a)(3)) and see Section IV (Agricultural Water in Sprout Operations)).

b. Equipment, tools, and storage

You must visually examine seeds, and packaging used to ship seeds, for signs of potential contamination with known or reasonably foreseeable hazards (§ 112.142(d)). If you have stored the seeds for any length of time after their arrival at your operation and prior to sprouting, before bringing seeds into the treatment area, you should re-inspect bags of seeds for signs of contamination. Any containers or utensils that will come into contact with the seeds as part of the treatment process must be cleaned and sanitized before such contact (§ 112.143(b)). In addition, if you rinse or soak seeds prior to sprouting, you must clean and sanitize the FCSs of tools and equipment used during this activity before they contact the seeds (§ 112.143(b) and see Section III (Cleaning and Sanitizing)).

c. Pre-treatment rinse, post-treatment rinse, and pre-germination seed soak

Seeds are typically rinsed before treatment, and may also be rinsed after treatment. While the Produce Safety Rule does not require pre- or post-treatment rinsing, we recommend that seeds be rinsed thoroughly before treatment to remove dirt, and to increase the efficiency of the treatment. If you rinse seeds before treatment, you should repeat the rinsing process with new water until the visible dirt is removed and the rinse water runs clear. We also recommend that you conduct your rinse in such a way as to maximize seed surface contact with the water (e.g., by mixing). If you use a surfactant to help remove soil and debris during seed rinsing, it should be rinsed out completely before the next step.

Depending on production practices and the type of seeds used, some sprout growers choose to use a pre-germination soak to improve germination. Soaking causes seeds to swell and softens their hulls to allow the sprout to grow out of the seed. Pre-germination soaking is not required by the Produce Safety Rule. If you rinse or soak seeds prior to sprouting, you must take any other measures that are reasonably necessary to prevent the introduction of known or reasonably foreseeable hazards into or onto the seeds during that process (§ 112.142(a)). Containers used to rinse or soak seeds should be large enough to allow thorough mixing. We recommend that you change the water each time you change the seeds you are rinsing or soaking (e.g., for each new production batch of sprouts) to ensure that it meets the agricultural water quality standards in §§ 112.41 and 112.44(a).

d. Chemical seed treatment

If you apply a chemical seed treatment, you should use clean containers designated for mixing the treatment chemicals. You should take care when preparing the treatment to ensure that the chemical is present at the desired concentration. You should determine the quantity of treatment solution needed based on the quantity of the seeds to be treated. You should refer to the chemical label instructions to calculate the amount of chemical needed to achieve the desired concentration in the desired volume of treatment solution. You should use a scale to weigh dry chemicals before adding them to the water (or other diluent). You should stir the solution to mix it completely and dissolve all solids. After mixing, you should verify the treatment solution

concentration according to the label directions (e.g., with a test kit), since the concentration can impact treatment effectiveness.

Once the chemical treatment is prepared, you should carefully combine it with the seeds. You should agitate the seeds and the chemical treatment at the correct temperature and for the appropriate amount of time according to your established procedures and the chemical's label instructions. It is important to use the correct amount of treatment for a known quantity of seeds; too much seed and/or too little chemical may decrease the effectiveness of the treatment. The same batch of seed treatment solution should not be used to produce more than one production batch of sprouts; you should prepare a new treatment solution before each production batch of sprouts. You should rinse seeds thoroughly after treatment to remove any residual treatment solution, if a rinse is recommended on the chemical label.

C. Corrective Actions for Seeds That May Be Contaminated with a Pathogen

If you learn that a lot of seeds has been associated with foodborne illness, or if you learn that a lot of seeds may be contaminated with a pathogen based on microbial test results (including those required under § 112.144(b)), you are required to take certain actions with respect to that seed lot and sprouts grown from that seed lot (§ 112.142(b)). If your seed supplier notifies you to stop using a lot of seed, we recommend you clarify with your supplier whether the request is due to potential contamination of the seed with a pathogen. You are required to maintain records related to corrective actions taken on seed for sprouting (§ 112.150(b)(6)). You should maintain a record to support your decision on whether to take a corrective action on potentially contaminated seed, including, for example, a record of the discussion with your seed supplier, the reason given for asking you to not use the lot of seed, and the date on which this information was communicated.

Section 112.142(b) requires that in such circumstances, except as provided in § 112.142(c), you must discontinue use of all seeds from that lot for sprout production (§112.142(b)(1)), must ensure that sprouts grown from that lot of seeds do not enter commerce (§112.142(b)(1)), and must report the information to the seed grower, distributor, supplier, or other entity from whom the seed was purchased (§112.142(b)(2)). Research indicates that seed or bean contamination, when it occurs, may be at low levels, intermittent, or unequally distributed within lots. Even low levels of human pathogens on seeds or beans used for sprouting are a concern, due to the ideal growth conditions present during sprouting. Once you know or have reason to believe that a lot of seeds or beans is contaminated, for example through microbial testing, there is reason to believe that other parts of that lot may also be contaminated. Even if you subdivide the original lot you receive, corrective actions are applicable to the entire original lot of seeds or beans.

You are required to report the findings to whomever supplied the seeds to you (e.g., the seed grower, distributor, or supplier) so that entity can then take appropriate follow-up actions. These actions may include informing other buyers about the contamination of the suspected lot of seeds, destroying or diverting any remaining seeds from that lot to non-food uses, and/or investigating the potential source of contamination, as necessary. Additionally, your seed grower, distributor, or supplier may be required to submit a report to the Reportable Food Registry (RFR), which requires food facilities to report certain information to the FDA when

there is a reasonable probability that the use of, or exposure to, an article of food will cause serious adverse health consequences or death to humans or animals.

Depending on the circumstances, it may also be appropriate for you to recall sprouts that have already entered commerce that were produced from the affected seed lot. You should recall any sprouts that are adulterated.

When your belief that a lot of seeds may be contaminated is based solely on microbial test results (e.g., spent sprout irrigation water test result), you would not have to take the steps described in §112.142(b)(1) (i.e., discontinuing use of the suspect lot of seeds and ensuring that sprouts made from them do not enter commerce) if you treat the suspect lot with a process that is reasonably certain to achieve destruction or elimination of the most resistant microorganisms of public health significance that are likely to occur in the seeds (§112.142(c)(1)). (We emphasize that such processes are far more robust than seed treatments described in §112.142(e), which typically only reduce the numbers of microorganisms of public health significance, rather than eliminating them.) This option exists to allow you flexibility in responding to a finding of a pathogen in a seed lot that would otherwise mean you would have to discontinue use of the seeds, and to encourage future innovation in seed treatment processes. Processes that meet the requirements of §112.142(c)(1) are not, at this time, commonly used in the sprouting industry.

Further, when your belief that a lot of seeds may be contaminated is based solely on microbial test results (e.g., spent sprout irrigation water or in-process sprout test result), and if you reasonably determine, through appropriate follow-up actions, that the lot of seeds is not the source of contamination, you do not have to take the steps in § 112.142(b)(1) or (2) (i.e., discontinuing use of that seed lot and ensuring that sprouts grown from that lot do not enter commerce), nor do you need to inform the seed grower, distributor or supplier of the positive test result). We acknowledge that a spent sprout irrigation water/in-process sprout sample that tests positive for a pathogen could be caused by contamination that originated either in the seed or elsewhere during the sprout growing process. Nonetheless, you must take the corrective actions described in § 112.142(b)(1), unless you can reasonably determine, through appropriate follow-up actions, that the lot of seeds is not the source of contamination. However, we expect that the situations in which you could take follow-up actions that would be adequate for you to reasonably determine that the lot of seeds was not the source of contamination are quite limited. For examples of scenarios in which we believe such a determination might be appropriate, see 80 FR 74353 at 74500-74501 (November 27, 2015).

Section 112.150(b)(6) requires that you establish and keep records of certain corrective actions including those corrective actions taken in accordance with §§ 112.142(b) and (c) (i.e., required actions if you know or have reason to believe that a lot of seeds may be contaminated with a pathogen) and § 112.148 (i.e., additional required actions where your knowledge or reason to believe that the lot of seeds is contaminated came from required spent sprout irrigation water or sprout pathogen testing pursuant to § 112.144(b)). Such records must comply with all applicable requirements for records in Subpart O.

In the event that your sprouts are associated with foodborne illness, you are required to discontinue use of all seeds from the affected lot for sprout production and ensure that sprouts grown from that lot of seeds do not enter commerce (§ 112.142(b)(1)). Also, you must report the

information (i.e., association of the seed lot with illness) to your grower, distributor, supplier, or other entity from whom you received that seed lot (§ 112.142(b)(2)). We are not aware of actions that you could take to demonstrate that the lot of seeds was not the source of contamination following an outbreak of foodborne illness associated with your sprouts. Regulators may make a determination that the seeds were not associated with the foodborne illness outbreak, but it is unlikely that you would have adequate information and records (e.g., epidemiological data and traceback information) to make that determination independently. Therefore, in such circumstances you must take the actions required in §§ 112.142(b)(1) and (2).

To facilitate taking any necessary corrective actions associated with your seed, we recommend that you establish and keep sufficient records to allow you to maintain the lot identity of the seeds you receive and the sprouts you produce, by which we mean that you would be able to determine which seed lot was used to grow each production batch of sprouts, and which container(s) of seed is part of a lot of seed. For each seed lot, your records should allow you to determine the identity of the supplier, variety of the seed, date of receipt, batch or lot number of the seed that you received (including all identifying numbers assigned to the seed, both by the supplier and by you, if different), and the date(s) that you used the seed from that lot for sprouting). This information will facilitate traceback to your seed supplier and (when possible) to the seed grower as needed. Maintaining records that provide traceability by connecting each production batch of sprouts to the lot of seeds used to grow them can help minimize the scope of product potentially affected in the event of a problem with a particular production batch of sprouts or a particular lot of seeds. For example, if you receive a positive test result on a spent sprout irrigation water test, you will need to know which seed lot(s) you used to grow that batch of sprouts, in order to comply with the corrective actions required in § 112.142(b). If your records do not allow you to identify which batch of seed was used to grow the contaminated product, you will need to take the required corrective actions on all seed of that variety at your operation (e.g., if your records do not identify which lot of clover seed was used to produce a batch of clover sprouts that tested positive for *Salmonella*, you would need to discontinue use of all lots of clover seed at your operation in order to comply with the requirements of § 112.142(b), unless you can reasonably determine, through appropriate follow-up actions, that the lot of seeds is not the source of contamination). We recommend that you maintain such records related to each lot of seeds for at least two years after your last use of the seed lot.

VI. Environmental Monitoring

This section of the guidance is intended to help you comply with the requirements of the Produce Safety Rule for environmental monitoring of the sprout growing, harvesting, packing, and holding environment by sampling and testing environmental samples for *Listeria* species (spp.) or *Listeria monocytogenes* (*L. monocytogenes*). For the purposes of this guidance, environmental samples are samples collected from a surface or area of the growing, harvesting, packing, or holding environment for the purpose of testing the surface or area for the presence of *Listeria* spp. or *L. monocytogenes*, in accordance with the requirements of §§ 112.144(a) and 112.145. To accomplish this, you must establish and implement a written environmental monitoring plan that is designed to identify *L. monocytogenes* if it is present in the growing, harvesting, packing, or holding environment (§ 112.145(a)). As part of your environmental monitoring plan, you must develop a sampling plan (§ 112.145(c)) that includes how often, when, and where you will sample the environment, and identifies the microorganism for which you will test (i.e., *Listeria*

spp. or *L. monocytogenes*). You must collect environmental samples aseptically and test them using a method as set forth in § 112.152 (§ 112.145(d)).

The written environmental monitoring plan must also include a corrective action plan that, at a minimum, requires you to take the actions in § 112.146, and details when and how you will accomplish those actions, if the growing, harvesting, packing, or holding environment tests positive for *Listeria* spp. or *L. monocytogenes* (§ 112.145(e)). The presence of *Listeria* spp. on a surface indicates the potential for contamination of that surface with *L. monocytogenes* and suggests that conditions are suitable for survival and/or growth of *L. monocytogenes* (Ref. 74). Section 112.146 describes the corrective actions that you must take if the growing, harvesting, packing, or holding environment tests positive for *Listeria* spp. or *L. monocytogenes*, which include:

- Conducting additional testing of surfaces and areas surrounding the area where *Listeria* species or *L. monocytogenes* was detected to evaluate the extent of the problem (§ 112.146(a)) (we refer to this type of testing in this document as "exploratory testing");
- Cleaning and sanitizing the affected surfaces and surrounding areas (§ 112.146(b));
- Conducting additional sampling and testing to determine whether the *Listeria* spp. or *L. monocytogenes* has been eliminated (§ 112.146(c)) (we refer to this type of testing in this document as "cleaning verification testing");
- Conducting finished product testing when appropriate (§ 112.146(d));
- Performing any other actions necessary to prevent recurrence of the contamination (§ 112.146(e)); and
- Taking appropriate action to prevent any food that is adulterated under section 402 of the FD&C Act from entering into commerce (§ 112.146(f)).

A. Principles for Developing an Environmental Monitoring Plan

L. monocytogenes is the target pathogenic microorganism of concern for environmental monitoring in a sprout operation. There are several species of *Listeria*, but *L. monocytogenes* is the primary species known to cause disease in humans. This pathogen is among the leading causes of death from foodborne illness in the United States, and predominantly affects older adults, pregnant women, newborns and those with weakened immune systems (Ref. 75, Ref. 76, Ref. 77, and Ref. 78). *L. monocytogenes* has been associated with foodborne illness outbreaks as a result of a strain of the pathogen that is transient in a food establishment. However, more commonly, *L. monocytogenes* foodborne illness outbreaks are associated with a strain of the pathogen that becomes established in a food establishment (i.e., a resident strain). In such cases, it can result in repeated product contamination (Ref. 79). Several foodborne illness outbreaks and recalls involving sprouts have occurred due to contamination with *L. monocytogenes*, and these outbreaks have included fatalities (Ref. 3, Ref. 4, and Ref. 80).

L. monocytogenes is widespread in the environment. It is found in soil, water, sewage, and decaying vegetation (Ref. 81, Ref. 82, Ref. 83, and Ref. 84). It can be readily isolated from

humans, domestic animals, raw agricultural commodities, and food processing environments (particularly in cool, damp areas). *L. monocytogenes* can multiply slowly at refrigeration temperatures, thereby challenging an important defense against proliferation of foodborne pathogens, refrigeration (Ref. 85, Ref. 86, Ref. 87, Ref. 88, and Ref. 89). However, *L. monocytogenes* grows more quickly at warmer temperatures. While *L. monocytogenes* may occasionally be found almost anywhere in a sprout production environment, it is most likely to become established in areas and on surfaces that are not only wet, but are relatively undisturbed and that may trap organic material. These areas and surfaces include drains, cooling units, drip pans, areas of condensation on walls or ceilings, and areas that are difficult to access or difficult to clean (e.g., cracks in metal, plastic or concrete surfaces, and hollow rollers). *L. monocytogenes* is known to form biofilms (i.e., communities of microbes embedded in an organic polymer matrix, adhering to a surface) on food contact surfaces (FCSs) and non-FCSs and, as a result, it can persist on these surfaces (Ref. 90). Once *L. monocytogenes* has established a niche, it may persist in the environment for long periods of time, serving as a potential source of repeated contamination, until and unless the niche is identified and eliminated (Ref. 89 and Ref. 91).

As stated above, "food contact surfaces" means "those surfaces that contact human food and those surfaces from which drainage, or other transfer, onto the food or onto surfaces that contact the food ordinarily occurs during the normal course of operations. 'Food contact surfaces' includes food contact surfaces of equipment and tools used during harvest, packing, and holding" (§ 112.3). "Food contact surfaces" also includes FCSs of equipment and tools used in growing sprouts, such as those that contact seeds for sprouting (see 112.143(b)). Requirements relating to "food contact surfaces" in part 112 apply not only to surfaces that contact (or drain onto, or otherwise transfer onto) sprouts themselves but also those that contact seeds used to grow sprouts. In a sprouting operation, FCSs include, for example: trays or drums used for sprouting; interior surfaces of containers used for seed rinsing, seed treatment, and pre-germination seed soaking; and counters that come into contact with sprouts during packing and/or packaging. The term "non-food contact surfaces" refers to any surfaces that, under normal operating procedures, do not contact either food or FCSs, and from which drainage, or other transfer, to food or FCSs does not occur. Non-FCSs may include equipment, vents, fixtures, drains, walls, floors, and employee clothing, shoes, and accessories.

The goals of an environmental monitoring program should be to:

- Find *L. monocytogenes* and its harborage sites if present in your operation;
- Ensure that corrective actions have eliminated *L. monocytogenes* and harborage sites when found in your operation; and
- Determine that FCSs that are used to grow, harvest, pack or hold sprouts have been cleaned and sanitized prior to contact with sprouts.

B. The Written Environmental Monitoring Plan

The mandatory contents of your environmental monitoring plan are prescribed in § 112.145, as discussed above. We recommend that you periodically review and assess your written environmental monitoring plan and update it as needed in response to new information, relevant

changes in your operation, or corrective actions that you have taken. For example, we recommend updating your environmental monitoring plan when you purchase new equipment, add new product lines, make significant changes to production flow or cleaning and sanitizing protocols, identify new sampling locations, or identify new concerns relative to your operation as a result of your environmental monitoring or corrective actions taken in response to such monitoring. We also recommend updating your environmental monitoring plan when you become aware of new science relative to the control of *L. monocytogenes.*

In the sections below, we describe in more detail the components of the sampling plan section of the environmental monitoring plan.

C. Developing a Sampling Plan

As part of your environmental monitoring plan, you must have a written sampling plan. Your written sampling plan must:

- Specify what you will test collected samples for (i.e., *Listeria* species or *L. monocytogenes*) (§ 112.145(c)(1)) (we refer to this microorganism as the "test microorganism" in the remainder of this document);
- Specify the frequency of sample collection, which must be no less than monthly (§ 112.145(c)(2));
- Specify at what point during production you will collect the samples (§ 112.145(c)(2)); and
- Specify the number and location of sample collection sites, which must include appropriate FCS sites and non-FCS sites of equipment, and other surfaces within the growing, harvesting, packing, and holding environment (§ 112.145(c)(3)).

Your written sampling plan should:

- Include a list of all FCS and non-FCS sites identified for testing at a level of detail that facilitates your identification of sampling sites;
- Identify the specific test method by which collected samples will be tested for the test microorganism (you are required to use a method set forth in § 112.152);
- Identify the person(s) responsible for sample collection in your operation and any specific training that the person(s) should have (§112.22(a)(3));
- Specify how you will meet the requirement in § 112.145(d) that samples must be collected aseptically. Your plan should include procedures for aseptic sample collection, including appropriate materials and steps to prepare for sample collection;
- Specify the sample collection method used (e.g., sponge v. swab sampling, whether any samples will be composited) and sample sizes to be collected at the various sample collection sites;

- Identify the laboratory you are using to conduct the testing; and
- Identify the records you will keep for each sample collected, including the documentation of the results of your analytical tests.

D. Testing for *Listeria* spp. or *L. monocytogenes*

Your sampling plan must specify what you will test collected samples for (i.e., the test microorganism, *Listeria* spp. or *L. monocytogenes)* (§ 112.145(c)(1)). Testing for *Listeria* spp. will detect multiple species of *Listeria*, including *L. monocytogenes*. A positive test result for the presence of *Listeria* spp. on an FCS or non-FCS indicates the potential for contamination of that surface with *L. monocytogenes* and suggests that conditions are suitable for survival and/or growth of *L. monocytogenes* (Ref. 74). However, such a test result does not establish the presence of *L. monocytogenes* on that surface. Nonetheless, testing for either the pathogen directly (*L. monocytogenes*) or the indicator organism (*Listeria* spp.) facilitates accomplishing the objectives for environmental monitoring stated above. If you test for *Listeria* spp. and take appropriate corrective actions when you find it on an FCS or non-FCS, you should eliminate *Listeria* spp. regardless of whether or not the initial finding of *Listeria* spp. was, in fact, *L. monocytogenes*. For this reason, except in certain circumstances described further below (such as when product testing of sprouts is appropriate as a corrective action), you are not required to take the added step of determining whether the identified *Listeria* spp. is, in fact, *L. monocytogenes,* although you may voluntarily choose to do so. Further, except in the circumstances described below (such as when testing for *L. monocytogenes* in a growing unit as a corrective action), we recommend that you test environmental samples for *Listeria* spp. because doing so will detect both *L. monocytogenes* as well as species of *Listeria* that are more common than *L. monocytogenes* and allow you to correct situations that could potentially lead to contamination with *L. monocytogenes*. See also the discussion in Section VI. K (Documenting and Interpreting Test Results) below for an explanation of how you should react to positive test results depending upon whether you identify *Listeria* spp. or *L. monocytogenes* as the test microorganism in your plan, to help you make this decision.

When testing product (e.g., as part of corrective actions), we recommend testing for *L. monocytogenes* rather than for *Listeria* spp. because of the risk to public health from *L. monocytogenes* in food. If you choose to test food for *Listeria* spp. and find it to be positive, we recommend that you either determine whether the *Listeria* spp. is *L. monocytogenes* or treat the food as if it were contaminated with *L. monocytogenes*. We further recommend that a lot of tested product be held until the test results are received, to prevent the need for a recall, should *L. monocytogenes* be detected in the product.

E. Person(s) Collecting Samples

You should identify in your sampling plan the person(s) that performs sample collection, at least by title. Sample collection may be performed by, for example, employees or contracted personnel. For a larger operation, we recommend you assemble a trained Sampling Team to undertake this activity. Training should be specified in your sampling plan, and your records should indicate that that training was successfully completed (see § 112.22(a)(3)). See Section III (Personnel Qualification, Training, and Hygienic Practices) in the draft re-issued sections (Ref.

18) for additional information on training related to sample collection for environmental samples. Samples must be collected aseptically, in accordance with § 112.145(d), and, therefore, training in aseptic techniques may be useful.

F. Establishing Sample Collection Locations and Frequency

1. Identifying sample collection locations

You should take a risk-based approach in determining where to sample and test the environment for the presence of *Listeria* spp. or *L. monocytogenes*. This can be accomplished by characterizing the areas in your operation according to the potential for product contamination (e.g., using a zone system). Zone designations for surfaces or areas reflect how close those surfaces or areas are to a ready-to-eat food (such as sprouts), and the risk that the surfaces or areas pose to the food if the surfaces or areas are contaminated with *L. monocytogenes* (Ref. 92, Ref. 93, and Ref. 94). For example, you could characterize your operation with four zones as shown in Appendix 3 (Potential Sources of *L. monocytogenes* for Sampling in a Sprout Operation). Zone 1 is all FCSs, zone 2 is non-FCSs in close proximity to food and FCSs, zone 3 is more remote non-FCSs that are in or near the production areas and could lead to contamination of zones 1 and 2, and zone 4 includes non-FCSs in remote areas outside of the production area, from which environmental pathogens can be introduced into the production environment.

Establishing a zone system is not a requirement of the Produce Safety Rule, but doing so may be helpful in designing an environmental monitoring plan and determining the frequency for sampling different surfaces in different areas. If you do not establish a zone-based system, you should otherwise characterize areas where you will collect environmental samples according to their potential for contamination or, at a minimum, distinguish between FCSs and non-FCSs.

2. Deciding on the number of food contact surface and non-food contact surface sampling sites

You must specify sample collection sites in your sampling plan, and these must include both FCS and non-FCS sites, which you may have designated by zone as described above (§ 112.145(c)(3)). You should make a list of FCS and non-FCS sites for potential sampling at a level of detail that facilitates your identification of sampling sites and sampling frequencies and include this list as part of your sampling plan. For examples of FCS and non-FCS sampling sites, see Appendix 3 (Potential Sources of *L. monocytogenes* for Sampling in a Sprout Operation).

We also recommend that you assign identifiers to each of your sample sites in your sampling plan, particularly if your operation has more than one possible location that could meet a site description. For example, if you have three rotary drum growing units, you should consider assigning unique identifiers (e.g., Growing Unit A, Growing Unit B, Growing Unit C) to facilitate sample collection and so that you can associate test results with the correct sample site (i.e., to facilitate trend analysis and corrective actions). In addition, you may consider developing a diagram of your operation that identifies the FCSs and non-FCSs that you have selected as sampling sites.

To determine the appropriate number of FCS and non-FCS sites to sample, you should consider the size of your operation (e.g., square footage), features of your operation (e.g. building layout), design of your equipment (e.g. hard to reach/disassemble/clean areas), product flow, production methods you use to produce the sprouts, your cleaning procedures, and any previous sampling results. The number of sampling sites must be sufficient to determine whether your measures are effective (§ 112.145(c)(3)). To accomplish this requirement, your selection of sampling sites should be adequate to verify the implementation and effectiveness of your sanitation measures for controlling the presence of *L. monocytogenes* in the sprout production environment by finding *Listeria* spp. or *L. monocytogenes* if it remains in the sprouting operation after your routine cleaning and sanitizing procedures. We recommend you develop a full listing of FCS and non-FCS sampling locations that you will sample within a specified frequency, and that the plan rotate through different sites from that list over time such that all sites that you have identified for testing are tested within the time period that you have determined is appropriate (e.g., quarterly). If you are testing a representative subset of FCS and non-FCS sites during each sampling event (e.g., monthly), each surface should be assigned a priority for sampling based on risk. For example, if you are using a four-zone system, we recommend that the sampling sites in zones 1 and 2 be sampled at a higher frequency than those in zones 3 and 4, in keeping with the higher risk of *L. monocytogenes* contamination of sprouts that zones 1 and 2 present.

While larger sprout operations will likely have more sampling sites than smaller operations, even the smallest sprout growers should collect samples from at least 5 sites of FCS (that directly contact the food) and 5 sites of non-FCS from each production area (e.g., each growing room) per sampling event. Your plan should focus on those sites that are in wet production areas (such as growing and packing areas) that pose the greatest risk.

As discussed in Section VI. A (Principles for Developing an Environmental Monitoring Plan), *L. monocytogenes* can be widespread in the environment, has been isolated from sprout production environments, and has been shown to persist on equipment and in the production environment in harborage sites. As a result, even if you are cleaning and sanitizing frequently, you should expect that you will occasionally have positive environmental samples. We discuss in subsequent sections of this guidance the need to take appropriate corrective actions and to reassess your sampling plan for possible revision to address the occurrence of positive pathogen or indicator organism findings. Further, if you consistently have negative test results from the sites you are sampling, you should reassess your sampling plan to determine whether you should add and/or substitute other sites to ensure that you are not missing a possible source of contamination.

3. Identifying sampling frequency

Your sampling plan must specify how often you will collect environmental samples, which must be no less than monthly. Frequency of sampling should be based on risk, which, in part, depends on the size and complexity of your operation. The timing of the collection of the environmental samples during production is an important addition to the sampling plan to ensure that sampling is conducted in a manner to optimize detection of *Listeria*, if present. While environmental sampling must be performed at least monthly (§ 112.145(c)(2)), each sampling event need not include samples from all your identified sampling sites, which may be spread out over a longer period of time (e.g., quarterly). When developing your sampling plan, you first need to determine: 1) how often you will conduct environmental sampling (i.e., collect samples from

some of your identified sampling sites), and 2) how frequently your periodic sampling will ensure that you have collected samples from of all your identified sampling sites (i.e., all sites in your site inventory. There are various ways to achieve this, and we provide distinct examples below.

a. Sampling frequency example #1

You evaluate your operation and identify 90 sites for sampling (30 FCS, 60 non-FCS, focusing on non-FCSs in close proximity to FCSs), of which a representative number will be sampled monthly. You might determine, for example, that all FCS sites that you have identified for testing will be tested at least every other month and all non-FCS sites that you have identified for testing will be tested at least quarterly.

In such a case you could choose to, for example, collect samples monthly, with 35 samples (from 15 FCS and 20 non-FCS sites) at each sampling event (e.g., first production day of every month), as shown in Table 3.

Table 3. Sampling Frequency Example #1

Month	FCS Sites Sampled	Non-FCS sites Sampled
1	Sites 1 – 15	Sites 1 – 20
2	Sites 16 – 30	Sites 21 – 40
3	Sites 1 – 15	Sites 41 – 60

b. Sampling frequency example #2

Like in Example 1, you again identify 90 sites for sampling (30 FCS, and 60 non-FCS). You might determine that both FCS and non-FCS will be sampled quarterly. To accomplish this you could choose to collect a proportion of those samples every week, for a total of 30 samples collected over the course of each month (e.g., 5-10 samples each week), as shown in Table 4.

Table 4. Sampling Frequency Example #2

Month / Week	FCS Sites Sampled	Non-FCS sites Sampled
Month 1 / Week 1	Sites 1 – 3	Sites 1 – 5
Month 1 / Week 2	Sites 4 – 6	Sites 6 – 10
Month 1 / Week 3	Sites 7 – 8	Sites 11 – 15
Month 1 / Week 4	Sites 9 – 10	Sites 16 – 20
Month 2 / Week 1	Sites 11 – 13	Sites 21 – 25

Month / Week	FCS Sites Sampled	Non-FCS sites Sampled
Month 2 / Week 2	Sites 14 – 16	Sites 26 – 30
Month 2 / Week 3	Sites 17 – 18	Sites 31 – 35
Month 2 / Week 4	Sites 19 – 20	Sites 36 – 40
Month 3 / Week 1	Sites 21 – 23	Sites 41 – 45
Month 3 / Week 2	Sites 24 – 26	Sites 46 – 50
Month 3 / Week 3	Sites 27 – 28	Sites 51 – 55
Month 3 / Week 4	Sites 29 – 30	Sites 56 – 60

You could not wait, however, for the end of each quarter to collect all 90 samples (e.g., sampling in January and then waiting until April to sample all 90 sites again) as this practice would not meet the monthly minimum sampling frequency requirement (§ 112.145(c)(2)).

c. Sampling frequency example #3

You establish a 4-zone system that prioritizes sampling from Zones 1 and 2. There are various ways to achieve this.

You might determine, for example, that all FCS sites that you identify for testing will be tested at least once a month and all non-FCS sites you identify for testing will be tested at least quarterly, recognizing that the FCS and non-FCS sites that you identify for testing are representative of the total number of FCS and non-FCS that you have identified in your operation.

In such a case you could choose to, for example, specify sample collection from specific FCS sites at least once every week, such that all FCS sites that you identify for testing are tested at least once each month.

As part of such an approach, you might choose to, for example, specify sample collection from representative sets of Zone 2 non-FCS sites every two weeks and sample collection from Zone 3 and 4 sites monthly, such that all non-FCS sites that you identify for testing are tested at least once each quarter.

G. Timing of Sample Collection

Your sampling plan must specify the point(s) during production at which you will collect environmental samples (§ 112.145(c)(2)). We recommend you collect environmental samples several hours after the start of production (e.g., 3 to 4 hours), preferably towards the end of

production, just prior to cleanup. Collecting your samples toward the end of production allows *L. monocytogenes* (if present) to work its way out of harborage sites and into the environment where it can more easily be detected.

Environmental samples should not be taken immediately after surfaces have been sanitized, as the sanitizer may affect the test results. The timing of collection of environmental samples for identifying *L. monocytogenes* should not be confused with that of other types of samples collected for verification of cleaning and sanitizing, which are typically collected immediately after sanitizing. For example, ATP hygiene monitoring involves the use of a device called a luminometer to measure the combined total ATP of organic material (food residues and microbial populations) collected from a swabbed surface. See discussion on verification of cleaning and sanitizing in Section III (Cleaning and Sanitizing) of this guidance for more information.

H. Sample Collection and Shipping

While not required by the Produce Safety Rule, we recommend your sampling plan specify the sampling method that you will use and detail your sample collection procedures. Use of appropriate materials and equipment is particularly important to ensure that sample collection is conducted aseptically, as required by § 112.145(d). For more details on aseptic sampling and information on sampling methods and sample collection, see Appendix 1 (Aseptic Sampling) and Appendix 2 (Collection Procedures – Environmental Sampling).

Prior to and during delivery or shipment to a laboratory, samples should be kept refrigerated. Sealed coolant packs should be used in lieu of ice, as needed during delivery or shipment, to avoid the possibility of melting ice contaminating the sample. Samples should not be frozen. Samples should be shipped so that they are received by the laboratory within 24 hours after sample collection. We recommend that the maximum timeframe between environmental sampling and analysis be 48 hours (Ref. 95).

Prior to sending the samples to a laboratory for testing, you should verify that your samples are clearly identified with the date and location of collection of the samples and the name of your sprouting operation. You should also specify the microorganism for testing.

I. Choosing a Laboratory

You should choose a laboratory that is qualified to test environmental samples for *Listeria* spp. or *L. monocytogenes* (whichever is your test microorganism). Testing is typically contracted to a third-party testing laboratory, but may also be performed by your own laboratory (e.g., your own "in-house" laboratory). If you use an "in-house" laboratory, you may be able to complete the screening step of the test method, but may need to send the enrichment broth out to a third-party testing laboratory for any confirmatory testing (if it cannot be conducted "in-house"). If using a third-party testing laboratory, you should use a laboratory that employs scientifically valid laboratory methods and procedures that can provide reliable, accurate test results. Using an accredited laboratory (e.g., a laboratory accredited to International Organization for Standardization (ISO)/IEC 17025:2017, General requirements for the competence of testing and calibration laboratories, Third edition, November 2017) is one way to have confidence that the

laboratory will provide reliable, accurate test results. Regardless of which laboratory you use, testing must be done using a method as set forth in § 112.152. You should ask the laboratory which test methods will be used when analyzing environmental samples for *Listeria* spp. or *L. monocytogenes*. We recommend you use a laboratory that either utilizes the method listed in § 112.152(a) or an alternate method reviewed by FDA and found to be equivalent that has been made available on our website (available at https://www.fda.gov/food/laboratory-methods-food/equivalent-testing-methodologies-listeria-species-and-listeria-monocytogenes-environmental-samples). If the laboratory test methods do not meet the requirements of § 112.152, you may consider selecting another laboratory to analyze your samples or request that the laboratory utilize a different method.

In addition, FDA published a final rule to establish the laboratory accreditation for analyses of foods (LAAF) program as required by FSMA. This final rule outlines the procedures and standards that accreditation bodies and laboratories will need to follow to participate in the LAAF program, as well as procedures for how FDA will manage and oversee the LAAF program. Once fully implemented, the final rule will require sprout operations subject to Subpart M to use a LAAF-accredited laboratory for corrective action testing of environmental samples (§ 112.146(a) and (c)) and finished product (§ 112.146(d)). See 21 CFR 1.1107(a)(1)(i).

J. Choosing a Test Method for *Listeria* spp. or *L. monocytogenes*

In accordance with § 112.152, the growing, harvesting, packing, and holding environment must be tested for *Listeria* spp. or *L. monocytogenes* using "Testing Methodology for *Listeria* species or *L. monocytogenes* in Environmental Samples" (currently available at https://www.fda.gov/media/94358/download[6] (§ 112.152(a))) or a scientifically valid method that is at least equivalent to FDA's method in accuracy, precision, and sensitivity (§ 112.152(b)). If your laboratory utilizes a different method to test your environmental samples, we recommend you use the questions included in the decision tree for test methods requirements for *Salmonella* and *E. coli* O157:H7 in spent sprout irrigation water (or sprouts) (available at https://www.fda.gov/media/132369/download) when you are determining whether the method may be equivalent to the method listed in § 112.152(a).

If you test sprouts for *Listeria monocytogenes* as part of a corrective action, we recommend that you use the procedures described in FDA's Bacteriological Analytical Manual Online (BAM), Chapter 10 – "*Listeria monocytogenes*," "Detection and Enumeration of *Listeria monocytogenes* in Foods" (currently available at: https://www.fda.gov/food/laboratory-methods-food/bam-chapter-10-detection-listeria-monocytogenes-foods-and-environmental-samples-and-enumeration).[6]

If you plan to use a test method other than the one listed in § 112.152(a) to test the growing, harvesting, packing, and holding environment for *Listeria* spp. or *L. monocytogenes*, it must be a

[6] Because websites are subject to change, it is possible that this specific website address will change. If you cannot access this document at that website, alternative websites where you currently can access this document include https://www.fda.gov/food/science-research-food/laboratory-methods-food, and https://www.fda.gov/food/guidance-regulation-food-and-dietary-supplements/food-safety-modernization-act-fsma. Alternatively, you can search on part or all of the title of the method, in the search box on FDA's Web site at https://www.fda.gov/ or using a generally available search engine.

scientifically valid method that is at least equivalent to the method in § 112.152(a) in accuracy, precision, and sensitivity (§ 112.152(b)). We use the term "scientifically valid" to mean an approach that is based on scientific information, data, or results published in, for example, scientific journals, references, textbooks, or proprietary research. Although you are not required to notify or submit information to FDA prior to using such an alternate method, you must establish and keep records of any such alternate methods that you use (§ 112.150(b)(5)). For example, your records could specify that you are using one of the methods that FDA listed in "Equivalent Testing Methodologies for *Listeria* species and *Listeria monocytogenes* in Environmental Samples" on our website (available at https://www.fda.gov/food/laboratory-methods-food/equivalent-testing-methodologies-listeria-species-and-listeria-monocytogenes-environmental-samples). If the alternate method that you use is not listed by FDA, your records regarding alternate methods should include the 1) name and identification number of the method validated by a third-party methods validation organization, or 2) detailed analytical procedures, an explanation of why the alternate method is at least equivalent to the reference method (such as a scientific study, or data or information regarding the sensitivity, specificity, and precision of the alternate method compared to the reference method), and any other relevant information supporting the use of the alternate method.

As noted in the list of "Equivalent Testing Methodologies for *Listeria* species and *Listeria monocytogenes* in Environmental Samples," FDA's method in the BAM for testing *Listeria* spp. and *L. monocytogenes* in environmental samples (available at https://www.fda.gov/food/laboratory-methods-food/bam-chapter-10-detection-listeria-monocytogenes-foods-and-environmental-samples-and-enumeration) is another method that is considered to be at least equivalent to the method in § 112.152(a) in accuracy, precision, and sensitivity and can be used as an alternate test method (as permitted in § 112.152(b)).

We recommend use of methods validated through a collaborative study (such methods are currently available at https://www.fda.gov/media/83812/download)[7], for example per AOAC Appendix J or ISO 16140:2016. Alternate methods should be compared to FDA's reference method (§ 112.152(a)) in environmental samples and demonstrated to meet the requirements for alternate methods in § 112.152(b) (i.e., demonstrated to be at least equivalent to the reference method in accuracy, precision, and sensitivity). Methods validated by third-party methods validation organizations such as AOAC Official Methods of Analysis (OMA), MicroVal, and Association Française de Normalisation (AFNOR) may meet the requirements in § 112.152(b); however, FDA does not automatically consider methods validated by third-party organizations such as the listed organizations to be equivalent to the reference method. Information on alternate methods reviewed by FDA and found to be equivalent will be made available on our website, such as at https://www.fda.gov/food/guidance-regulation-food-and-dietary-supplements/food-safety-modernization-act-fsma and https://www.fda.gov/food/science-research-food/laboratory-methods-food.

[7] Alternative websites where you can currently access this document include: https://www.fda.gov/science-research/field-science-and-laboratories/method-validation-guidelines and https://www.fda.gov/food/guidance-regulation-food-and-dietary-supplements/food-safety-modernization-act-fsma. Alternatively, you can search on part or all of the title of the method, in the search box on FDA's Web site at https://www.fda.gov/ or using a generally available search engine.

K. Documenting and Interpreting Test Results

Section 112.150(b)(4) requires that you establish and keep records of test results from environmental testing for *Listeria* spp. or *L. monocytogenes* you perform in compliance with §§ 112.144(a) and 112.145 or any additional testing conducted if you detect *Listeria* spp. or *L. monocytogenes* in the environment in compliance with § 112.146 (a), (c), (d), and (e). The results of all analytical tests must be documented, regardless of whether they were conducted by your own (e.g., in-house) laboratories or third-party laboratories.

Testing for *Listeria* spp. or *L. monocytogenes* in environmental samples using the FDA reference method (§ 112.152(a)) can yield one of the following results:

- A positive result for *Listeria* spp., which for purposes of the FDA reference method, means finding the presence of typical colonies on a *Listeria* specific agar (i.e., completing Steps I.A-I.F in the FDA reference method with respect to *Listeria*).[8]

- A positive result for *L. monocytogenes*, which can be obtained if a positive result for *Listeria* spp., or a presumptive positive result for *L. monocytogenes*, is followed by confirmatory steps that result in a confirmed *L. monocytogenes* cultural isolate (i.e., completing Step I.I in the FDA reference method).

- One of two types of negative results:

    - *Listeria* specific agars do not yield a positive result for *Listeria* spp., indicating a negative finding for *Listeria* spp.; or

    - Additional confirmatory steps after a positive result for *Listeria* spp. or a presumptive positive result for *L. monocytogenes* do not result in confirmation of the presence of *L. monocytogenes*, indicating that while *Listeria* spp. were found, *L. monocytogenes* was not found.

1. Sampling plan that specifies testing for *Listeria* spp. (recommended approach)

If you choose to establish a sampling plan that specifies testing for *Listeria* spp., you are not required to perform confirmatory testing for *L. monocytogenes* after obtaining a positive test result for *Listeria* spp., although you may voluntarily choose to do so.

If your plan specifies that you will test for *Listeria* spp. and you conduct testing using the FDA reference method (recommended approach):

- A positive result for *Listeria* spp. triggers the requirement to take corrective actions under § 112.146;

[8] The FDA reference method provides options for testing for both *Listeria* spp. and *L. monocytogenes*. They are described in the reference method in parallel, such that, for example, step I.E.1 describes part of the procedure for testing for *L. monocytogenes* while step I.E.2 describes part of the procedure for testing for *Listeria* spp. Thus, a "positive result for *Listeria* spp." refers to completion of the reference method up to step I.F (or G, if applicable) including all steps relevant to *Listeria* spp.

- A negative result for *Listeria* spp. does not trigger the requirement to take corrective actions under § 112.146;

- If you voluntarily choose to perform confirmatory testing for *L. monocytogenes* on an environmental sample that yields a positive result for *Listeria* spp., a positive result for *L. monocytogenes* also triggers the requirement to take corrective actions under § 112.146; and

- If you voluntarily choose to perform confirmatory testing for *L. monocytogenes* on an environmental sample that yields a positive result for *Listeria* spp., a negative result for *L. monocytogenes* does not eliminate the requirement to take corrective actions under § 112.146 based on your positive result for *Listeria* spp. A positive test result for the presence of *Listeria* spp. on an FCS or non-FCS indicates the potential for contamination of that surface with *L. monocytogenes* and suggests that conditions are suitable for survival and/or growth of *L. monocytogenes* (Ref. 74). Therefore, if your tests identify the presence of *Listeria* spp., you should eliminate *Listeria* spp., regardless of whether or not the initial finding of *Listeria* spp. was, in fact, *L. monocytogenes*.

2. Sampling plan that includes testing for *L. monocytogenes*

If you choose to establish a plan that specifies that you will test for *L. monocytogenes* and you conduct testing using the FDA reference method:

- A positive result for *Listeria* spp. triggers the requirement to take corrective actions under § 112.146, and you must continue with confirmatory testing to identify *L. monocytogenes* as provided in your sampling plan;

- A negative result for *Listeria* spp. does not trigger the requirement to take corrective actions under § 112.146;

- A positive result for *L. monocytogenes* triggers the requirement to take corrective actions under § 112.146; and

- A negative result for *L. monocytogenes* does not eliminate the requirement to take corrective actions under § 112.146 based on your positive result for *Listeria* spp. A positive test result for the presence of *Listeria* spp. on an FCS or non-FCS indicates the potential for contamination of that surface with *L. monocytogenes* and suggests that conditions are suitable for survival and/or growth of *L. monocytogenes* (Ref. 74). Therefore, if you identify *Listeria* spp., you should eliminate *Listeria* spp., regardless of whether or not the initial finding of *Listeria* spp. was, in fact, *L. monocytogenes*.

L. Developing a Corrective Action Plan and Taking Corrective Actions

Your written environmental monitoring plan must also include a corrective action plan that, at a minimum, requires you to take the actions in § 112.146, and details when and how you will accomplish those actions, if the growing, harvesting, packing, or holding environment tests positive for *Listeria* spp. or *L. monocytogenes* (§ 112.145(e)).

1. Corrective action plan

Having a corrective action plan in place at your operation will help ensure that you take corrective actions quickly and consistently in response to a finding of *Listeria* spp. or *L. monocytogenes* in the environment. Your corrective action plan must (§ 112.145(e)):

- Specify when and how you will conduct additional testing of surfaces and areas surrounding the area where the positive test result was detected to evaluate the extent of the problem, including the potential for *Listeria* spp. or *L. monocytogenes* to have become established in a niche (§ 112.146(a)) (i.e., "exploratory testing");
- Specify how and when you will clean and sanitize the affected surfaces and surrounding areas (§ 112.146(b));
- Specify when and how you will conduct additional sampling and testing to determine whether the *Listeria* spp. or *L. monocytogenes* has been eliminated (§ 112.146(c)) (i.e., "cleaning verification testing");
- Specify when and how you will conduct finished product testing when appropriate (§ 112.146(d));
- Indicate that you will perform any other actions necessary to prevent recurrence of the contamination (§ 112.146(e)) and should specify what those actions may be. For example, you should specify additional steps you will take to determine the source and route of contamination if your exploratory testing or cleaning verification testing yields positive results for *Listeria* spp. or *L. monocytogenes* and steps you will take to eliminate such sources and routes of contamination; and
- Specify when and how you will take appropriate action to prevent any food that is adulterated under section 402 of the FD&C Act from entering into commerce (§ 112.146(f)).

2. Implementing corrective actions

We recommend that you consider what corrective action is appropriate in a specific set of circumstances based, in part, on whether or not you detected *Listeria* spp. or *L. monocytogenes*, whether or not you detected the organism on an FCS or a non-FCS site, and whether or not this was the first time you detected the organism on the surface. Based on the criteria described above, Table 5 provides a key to the corrective actions described in the sections below.

Table 5. Corrective Actions When *Listeria* Species or *L. monocytogenes* is Found in an Environmental Sample

Test Microorganism	Type of Surface or Product where Positive was Found	Positive*	Sprout Guidance Section
Listeria spp.	FCS	1st positive	L.3.a.

Listeria spp.	FCS	2nd positive	L.3.b.
*Listeri*a spp.	FCS	3rd or subsequent positive	L.3.c.
L. monocytogenes	Product	Any	L.3.c.
Listeria spp.	FCS in a growing unit	Any	L.3.d.
L. monocytogenes	FCS or non-FCS	Any	L.4.
Listeria spp.	Non-FCS	1st positive	L.5.a.
Listeria spp.	Non-FCS	2nd positive	L.5.b.
Listeria spp.	Non-FCS	3rd or subsequent positive	L.5.c.

*For the purposes of this table, 2nd, 3rd, etc. positives refer to positives identified during corrective actions in response to the 1st positive identified through routine monitoring.

3. Corrective actions if you detect *Listeria* spp. on a food contact surface

We describe appropriate corrective actions for a positive test result for *Listeria* spp. from an environmental sample collected during your routine sampling of an FCS site in this section. These steps are also summarized in Figure 3 (FCS* Testing and Follow-Up Activities Decision Tree).

a. *Listeria* spp. positive on an FCS (1st positive)

You should examine the area surrounding the site of the positive test result in all directions for potential sources of *Listeria* spp. as described in Appendix 3 (Potential Sources of *L. monocytogenes* for Sampling in a Sprout Operation) of this guidance. You should pay particular attention to possible niches that provide harborage for *L. monocytogenes.*

i. Exploratory testing

You must conduct additional testing of surfaces and areas surrounding the area where *Listeria* spp. was detected to evaluate the extent of the problem, including the potential for *Listeria* spp. or *L. monocytogenes* to have become established in a niche (§ 112.146(a)). Consider the following:

- Exploratory testing provides you with information about the extent of the problem represented by the initial positive test result (e.g., whether the presence of *Listeria* spp. is isolated or more extensive). You do not need to wait for the results of the exploratory testing before doing any cleaning and sanitizing, but you should use the results of

exploratory testing to inform your decision about the extent of cleaning and sanitizing that you will perform, as required by § 112.146(b) (see next subheading);

- You should conduct exploratory sampling and testing of sites that represent a potential source of contamination to the contaminated FCS site. Exploratory testing should include at least 3 samples from sites in close proximity to the positive site and those upstream in the operation (i.e., locations in the operation that the product contacts earlier in the product flow) to help identify the source of contamination;

- Exploratory testing while in production: If you receive a positive *Listeria* spp. result for a routine sample from an FCS site while you are in production (e.g., you are either still growing the production batch of sprouts that was growing when you took your routine sample, or you have started another production batch of sprouts), you should conduct exploratory testing during that production cycle;

- Exploratory testing when not in production: If you receive a positive result for a routine sample from an FCS site when you are not in production (e.g., the production batch of sprouts that was growing when you took your routine sample has been completed and you have not started the next production batch), you should conduct exploratory testing during the production of the next batch; and

- If you receive any positive results from your exploratory testing, you should conduct intensified cleaning and sanitizing and intensified sampling and testing (discussed below as a recommended response to a 2nd positive).

ii. Cleaning and sanitizing

You must clean and sanitize the affected surfaces, including FCSs and non-FCSs, as well as surfaces from the surrounding area where the positive sample was taken (§ 112.146(b); see also §§ 112.123(d)(1)-(2); 112.143(b)). Consider the following:

- At a minimum, the site of the initial positive *Listeria* spp. result and the sites of any additional positive results found during exploratory testing should all be cleaned and sanitized before your next production run; and

- You should also consider verifying the efficacy of your cleaning and sanitizing using additional methods (e.g., ATP testing) beyond the required testing (§ 112.146(c)) before your next production run; see Section III (Cleaning and Sanitizing), and

- If you receive negative test results from all of your exploratory testing, and you have cleaned and sanitized the affected FCSs and non-FCSs from the surrounding area where the original positive sample was taken, you may consider the actions required by §§112.146(b) and 112.146(c) to have been accomplished while conducting the actions required by § 112.146(a).

b. *Listeria* spp. positive on an FCS during exploratory testing (2nd positive)

i. Three production days of intensified cleaning and sanitizing

If any of the exploratory testing samples are positive for *Listeria* spp., as an action to prevent recurrence of the contamination, you must clean and sanitize the affected surfaces and surrounding areas (§ 112.146(b)). The cleaning and sanitizing that you perform under these circumstances should be intensified beyond your routine efforts and should be performed in the affected areas for the next three production days. Intensified cleaning and sanitizing include sanitation measures that you perform in addition to your normal sanitation procedures. See also Section III.F. (Cleaning and Sanitizing Conducted in Response to Suspected or Known Contamination). You should escalate the intensity of these measures in response to additional positive *Listeria* spp. samples. Intensified cleaning and sanitizing can include:

- Increasing the frequency of cleaning and sanitizing for certain pieces of equipment;
- Breaking down the equipment into its parts for further cleaning;
- The use of more effective or different cleaning and sanitizing methods or agents (e.g. more aggressive scrubbing, applying heat (where possible)) (see Section III (Cleaning and Sanitizing));
- Intensified (e.g., more vigorous, longer duration) scrubbing of surfaces;
- Identifying, cleaning, and sanitizing of possible harborage sites and possible cross-contamination routes; and
- Soaking of equipment parts in an appropriate sanitizing agent for an appropriate duration.

ii. Three production days of intensified testing

If any of the exploratory testing samples are positive for *Listeria* spp., as an action to prevent recurrence of the contamination (§§ 112.146(c) and 112.146(e)), you should conduct a round of intensified testing during each of the next three production days, both to verify the effectiveness of your intensified cleaning and sanitizing, and to look for possible harborage sites in the affected area. To look for possible harborage sites, you should disassemble equipment as appropriate, and sample and test areas of the equipment exposed by disassembly prior to cleaning and sanitizing the equipment. Each round of intensified testing should include at least 3 samples, including the initial *Listeria* spp. positive site, surrounding positive sites that tested positive for *Listeria* spp. exploratory testing, and surrounding FCS and non-FCS sites in close proximity to any positive sites.

iii. Finished product testing and other product actions

You must conduct finished product testing when appropriate (§ 112.146(d)). In the circumstances described here, where you have identified a second positive result on an FCS, you should:

- Test the production batch(es) of sprouts from the production day associated with the second positive for *Listeria* spp. on the FCS. You should test the sprouts for *L.*

monocytogenes using a statistically-based sampling protocol and analytical methods that will provide an appropriate level of confidence in the results (e.g., 95% confidence that you will detect *L. monocytogenes* in the sample if it is present). The International Commission on Microbiological Specifications for Foods (ICMSF) provides additional information on scientifically-based sampling plans that can be used to provide statistical confidence for results of product testing (Ref. 96). While these test results are pending, and while you are taking the other steps recommended in this section (i.e., three production days of intensified cleaning and sanitizing, three production days of intensified sampling and testing, receiving results from such testing, conducting a comprehensive investigation), you should not allow the production batch(es) of sprouts to enter commerce.

iv. Comprehensive investigation

Following a second positive finding of *Listeria* spp. on an FCS, as an action to prevent recurrence of the contamination (§ 112.146(e)), you should conduct a comprehensive investigation to identify and address *Listeria* sources, and modify your sanitation procedures, where appropriate. You may need to stop production at your sprout operation in order to conduct the comprehensive investigation or to take actions based on the comprehensive investigation (e.g., repair or replace equipment). Such an investigation should include:

- Checking maintenance records for modifications or repairs to relevant equipment;
- Interviewing and observing sanitation, maintenance, and production employees to determine whether appropriate procedures are being followed, including hygienic practices;
- Reviewing production, maintenance, and sanitation procedures to determine whether modification of the procedures is necessary to prevent contamination; and
- Reviewing traffic patterns and equipment layout.

Based on the results of the comprehensive investigation, you should correct any identified problems (e.g., re-train personnel, revise sanitation procedures, repair equipment, update maintenance program) to prevent recurrence of contamination (§§ 112.21(d) and 112.146(e)).

v. Negative results from intensified testing and finished product testing

If all results from your three production days of intensified testing are negative, and your finished product testing is also negative, you should return to routine environmental monitoring. It would be reasonable to allow all sprouts from all three production days to enter commerce at this point, provided there is no other reason for concern (e.g., all other testing requirements in § 112.147 have been satisfied for these batches). However, you should target those surfaces where positive test results were found for sampling and testing during your next routine environmental sampling event.

c. *Listeria* spp. positive during intensified testing (3rd or subsequent positive) or *L. monocytogenes* positive product

i. Stop production and investigate

If your intensified sampling and testing again detects *Listeria* spp. on an FCS and/or a non-FCS site (third or subsequent positive), and/or you detect *L. monocytogenes* in your product, you should assume that you have a harborage site. As an action to prevent recurrence of the contamination (§ 112.146(e)), you should stop production and consult a food safety expert familiar with troubleshooting *L. monocytogenes* contamination problems to conduct a comprehensive investigation and make recommendations for appropriate actions for you to take based upon that investigation. You could choose to seek more information from sources such as cleaning and sanitation suppliers, equipment manufacturers, academia, extension services, and industry associations to assist you with these corrective action steps.

ii. Product actions

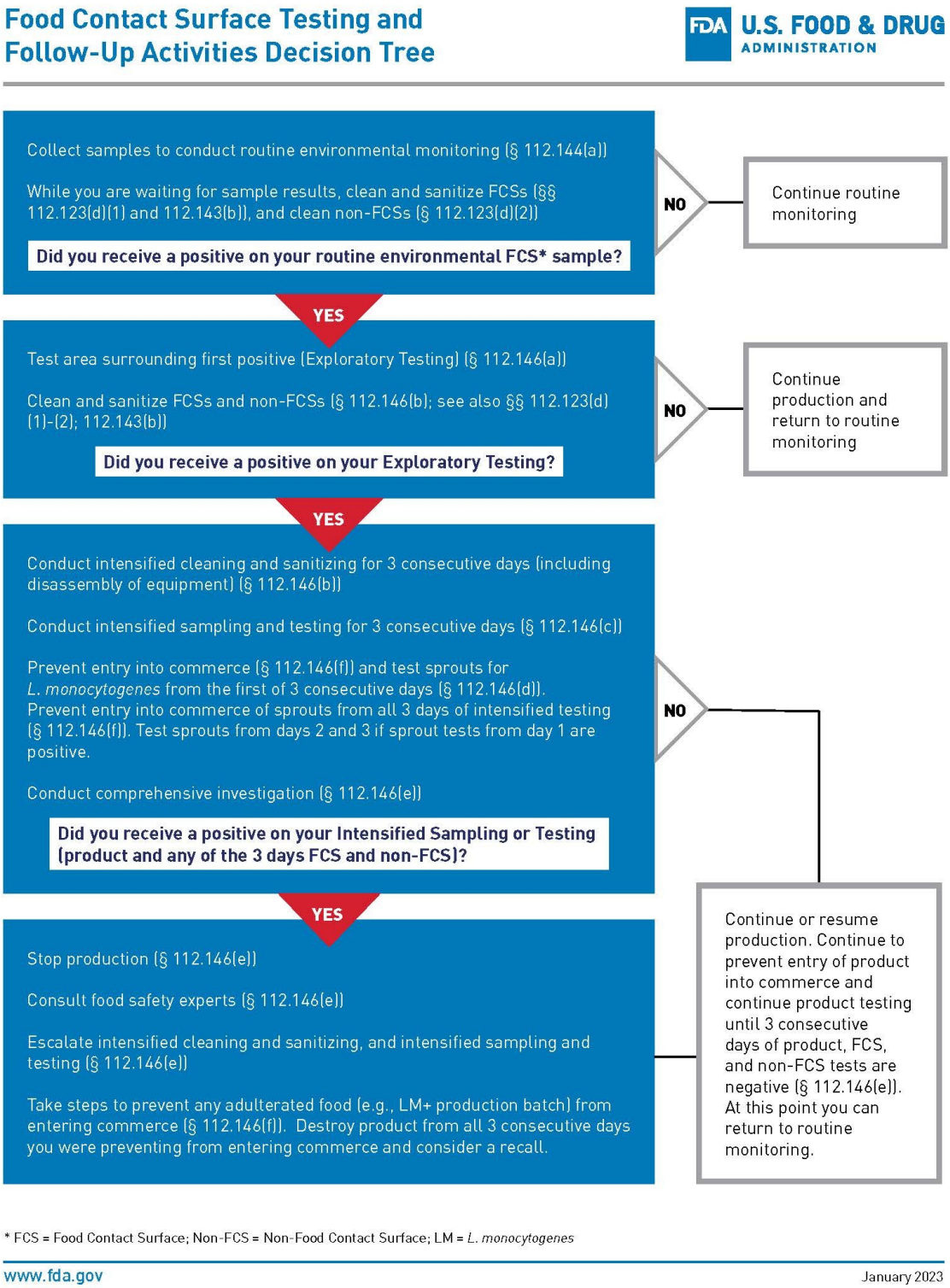
You also must take appropriate action to prevent any food that is adulterated under section 402 of the FD&C Act from entering into commerce (§ 112.146(f)). If a production batch of sprouts tests positive for *L. monocytogenes*, § 112.146(f) requires you to prevent it from entering into commerce. We recommend that you destroy any such production batch. If you are following the "three production days of intensified testing" described above in L.3.b, you also should consider the production batches during all three days of intensified testing to be adulterated, considering the positive finding of *L. monocytogenes* in the first batch, combined with the earlier positive findings in your environment. Moreover, we recommend that you destroy any such batches if you have held them or recall them if you have already shipped them. You should also evaluate whether any other production batches of sprouts (either at your operation or in distribution) should be recalled or destroyed.

iii. Returning to production

After all of these corrective actions have been taken and production begins again, you should prevent your new production batches of sprouts from entering commerce until you test each production batch of sprouts for *L. monocytogenes* and conduct intensified testing of the environment on each production day, until you have received three consecutive days of negative test results for FCSs, non-FCSs, and sprouts. After receiving these negative test results for three consecutive days, you would not need to continue to test each production batch of sprouts for *L. monocytogenes*.

The example in Figure 3 addresses testing and follow-up actions for a single positive finding of *Listeria* spp. on an FCS during one sampling period. Detecting *Listeria* spp. at several FCS sampling locations during the same sampling period could indicate that your routine sanitation procedures are inadequate and could indicate that *Listeria* spp. has more broadly established in your operation. The risk associated with cross-contamination from contaminated FCS sites to food increases as the number of contaminated FCS sites increases. Therefore, when more than one FCS site sample tests positive for *Listeria* spp. during one sampling period, you should immediately review your written cleaning and sanitation procedures to identify and implement more effective measures and escalate your corrective actions until you have identified and eliminated the *Listeria* spp. source(s).

Figure 3. FCS* Testing and Follow-Up Activities Decision Tree



d. Additional considerations if you detect *Listeria* spp. on an FCS in a growing unit (e.g., rotary drum growing units)

If you obtain a positive for *Listeria* spp. from an FCS in a growing unit (e.g., FCS on a rotary drum growing unit) during growing, there is a heightened risk that your sprouts may be contaminated with *L. monocytogenes*. As a result, we recommend that you take additional corrective actions in response to the finding, beyond those already discussed above.

If you receive notification of a positive test result for *Listeria* spp. from an FCS in a growing unit collected during your routine sampling (i.e., a first environmental positive from such a site), you should take all of the corrective actions described above relative to such a finding and should also take action to prevent the production batch(es) of sprouts associated with the positive sample site (i.e., the production batch of sprouts that was grown in that growing unit in which the positive *Listeria* spp. sample was collected at the time it was collected and any other potentially affected product) from entering commerce while you take the following steps:

1) Conduct exploratory testing as described above (§ 112.146(a)). If your exploratory testing yields additional positive samples for *Listeria* spp., you should conduct intensified cleaning and sanitizing, and intensified testing, for three consecutive production days (as described above as a recommended response to a 2nd *Listeria* spp. positive on an FCS site). If any of your intensified testing yields a positive result, you should proceed to the corrective actions recommended above for a 3rd *Listeria* spp. positive on an FCS site.

2) Further analyze the sample that was positive for *Listeria* spp. to determine whether the *Listeria* identified is *L. monocytogenes.* If you determine the sample is positive for *L. monocytogenes*, § 112.146(f) requires you to take appropriate action to prevent the production batch of sprouts grown in the affected growing unit from entering commerce. We recommend that you destroy any such product.

3) If all of your intensified testing of the environment for *Listeria* spp. for three consecutive production days, and *L. monocytogenes* finished product testing is negative, it would be reasonable at that time to allow the production batch of sprouts at issue to enter commerce, and to return to routine sampling and testing.

4. Corrective actions if you detect *Listeria monocytogenes* on a food contact surface or non-food contact surface

In general, we anticipate that sprout operations will elect to test FCS and non-FCS sites for *Listeria* spp., rather than *L. monocytogenes*. Ordinarily, there is minimal value in determining whether *Listeria* spp. is *L. monocytogenes* because, typically, you should focus on eliminating the *Listeria* spp. regardless of whether it is, in fact, *L. monocytogenes*. However, in certain cases you should consider conducting further tests to determine whether the *Listeria* spp. positive in your environmental samples is *L. monocytogenes.* One example of such a situation is described above in Section VI. L. 3. d (Additional considerations if you detect *Listeria* spp. on an FCS in a growing unit (e.g., rotary drum growing units)) for positive *Listeria* spp. findings on an FCS in a growing unit.

If you detect *L. monocytogenes* on an FCS, § 112.146(f) requires to you take appropriate action to prevent any food that is adulterated under section 402 of the FD&C Act from entering into commerce. Depending on the circumstances, you may have production batches of sprouts that are adulterated under section 402(a)(4) of the FD&C Act because of their association with the affected FCS location. We recommend that you destroy any potentially affected production batch of sprouts (or other food) associated with the contaminated FCS (as part of a recall, if applicable) and follow procedures outlined above in the subsection titled "returning to production" in Section VI. L. 3. c. (*Listeria* spp. positive during intensified testing (3rd or subsequent positive) or *L. monocytogenes* positive product).

Section 112.150(b)(6) requires that you establish and keep records of corrective actions conducted in accordance with the requirements of § 112.146. Such records must include:

- Documentation of additional testing of surfaces and areas surrounding the area where the positive test result was detected to evaluate the extent of the problem, including the potential for *Listeria* spp. or *L. monocytogenes* to have become established in a niche (§ 112.146(a)) (i.e., "exploratory testing");

- Documentation of cleaning and sanitizing the affected surfaces and surrounding areas (§ 112.146(b));

- Documentation of additional sampling and testing to determine whether the *Listeria* spp. or *L. monocytogenes* has been eliminated (§ 112.146(c));

- Documentation of finished product testing when appropriate (§ 112.146(d));

- Documentation of any other actions necessary to prevent recurrence of the contamination (§ 112.146(e)); and

- Documentation of appropriate action to prevent any food that is adulterated under section 402 of the FD&C Act from entering into commerce (§ 112.146(f)).

5. Corrective actions if you detect *Listeria* spp. on a non-food contact surface

We describe appropriate corrective actions for a positive test result for *Listeria* spp. from an environmental sample collected during your routine sampling of a non-FCS site in this section. These steps are also summarized in Figure 4 (Non-FCS* Testing and Follow-Up Activities for Zone 2 Decision Tree). We focus on corrective actions for positives in Zone 2, which are in close proximity to food and FCS, because, when contaminated, they present a greater risk of contamination of the food than do non-FCS in other zones. However, as required by § 112.146, you must also take corrective actions if a positive result(s) is obtained in Zones 3 or 4. Corrective actions for non-FCS positives in Zones 3 and 4 may be less rigorous than those for non-FCS positives in Zone 2 provided that all relevant requirements are met. For example, you must conduct additional testing (i.e., "exploratory testing," see Section VI. L. 3. a (*Listeria* spp. positive on an FCS (1st positive))) of surfaces and areas surrounding the area where *Listeria* species or *L. monocytogenes* was detected (§ 112.146(a)). However, it would be reasonable to take fewer exploratory samples after a positive test result in Zone 3 or 4 compared to the number of exploratory samples taken after a positive test result in Zone 2. You must clean and sanitize

the affected surfaces and surrounding areas after obtaining a positive test result in Zone 3 or 4 (§ 112.146(b)), however the cleaning and sanitizing conducted might be more aggressive after a positive test result in Zone 2 compared to cleaning and sanitizing after a positive test result in Zone 3 or 4. You must then conduct additional sampling and testing (see Section VI. L. 3. a (*Listeria* spp. positive on an FCS (1st positive)) to determine whether the *Listeria* species or *L. monocytogenes* has been eliminated after obtaining a positive test result in Zone 3 or 4 (§ 112.146(c)). However, it would be reasonable to take more samples as a follow-up for a positive test result in Zone 2 compared to the number of samples taken after a positive test result in Zone 3 or 4. Finished product testing is also more likely to be necessary in response to a positive test result in Zone 2 compared to after a positive test result in Zone 3 or 4.

The discussion in this section relates only to positive environmental samples on non-FCSs. If you obtain a positive environmental sample (e.g., *Listeria* spp.) on an FCS during follow-up testing to an original positive on a non-FCS (e.g., during exploratory testing), you should immediately switch to taking corrective actions appropriate to positive findings on FCSs, which is covered in Section VI. L. 3. b (*Listeria* spp. positive on FCS during exploratory testing (2nd positive)) above.

a. *Listeria* spp. positive on non-FCS (1st positive)

You should examine the area surrounding the site of the positive test result in all directions for potential sources of *Listeria* spp. as described in Appendix 3 (Potential Sources of *L. monocytogenes* for Sampling in a Sprout Operation) of this guidance. You should pay particular attention to possible niches that provide harborage for *L. monocytogenes.*

i. Exploratory testing

You must conduct additional testing of surfaces and areas surrounding the area where *Listeria* spp. was detected to evaluate the extent of the problem, including the potential for *Listeria* spp. or *L. monocytogenes* to have become established in a niche (§ 112.146(a)). Consider the following:

- Exploratory testing provides you with information about the extent of the problem represented by the initial positive test result (e.g., whether the presence of *Listeria* spp. is isolated or more extensive). You do not need to wait for the results of the exploratory testing before doing any cleaning and sanitizing. However, you should use the results of exploratory testing to inform your decision about the extent of cleaning and sanitizing that you will perform as required by § 112.146(b) (see next subheading);

- If the original positive test result is from a composite sample, you should either conduct additional follow-up testing to identify the specific non-FCS(s) that is contaminated with *Listeria* spp. or, alternatively, conduct your exploratory testing as if each non-FCS site represented by the composite is positive (i.e., conduct exploratory testing of surfaces and areas surrounding all of the sites represented by the composite);

- Exploratory testing should include at least 3 samples from surrounding FCS and non-FCS sites in close proximity to the positive site;

- Exploratory testing while in production: If you receive a positive *Listeria* spp. result for from a routine sample for a non-FCS site while you are in production (e.g., you are either still growing the production batch of sprouts that was growing when you took your routine sample, or you have started another production batch of sprouts), you should conduct exploratory testing during that production cycle;

- Exploratory testing when not in production: If you receive a positive *Listeria* spp. result for a routine sample for a non-FCS site when you are not in production (e.g., the production batch of sprouts that was growing when you took your routine sample has been completed and you have not started the next production batch), you should conduct exploratory testing during the production of the next batch; and

- If you receive any positive results from your exploratory testing, you should conduct intensified cleaning and sanitizing and intensified sampling and testing (discussed below as a recommended response to a 2nd positive)

ii. Cleaning and sanitizing

You must clean and sanitize the affected surfaces, including FCSs and non-FCSs as well as surfaces from the surrounding area where the positive sample was taken (§ 112.146(b); see also §§ 112.123(d)(1)-(2); 112.143(b)). Consider the following:

- At a minimum, the site of the initial positive *Listeria* spp. result and the sites of any additional positive results found during exploratory testing should all be cleaned and sanitized before your next production run; and

- You should also consider verifying the efficacy of your cleaning and sanitizing using additional methods (e.g., ATP testing) beyond the required testing before your next production run; See Section III (Cleaning and Sanitizing); and

- If you receive negative test results from all of your exploratory testing, and you have cleaned and sanitized the affected FCSs and non-FCSs from the surrounding area where the original positive sample was taken, you may consider the actions required by §§112.146(b) and 112.146(c) to have been accomplished while conducting the actions required by § 112.146(a).

b. *Listeria* spp. positive on non-FCS during exploratory testing or cleaning verification testing (2nd positive)

i. Intensified cleaning and sanitizing

If any of the exploratory testing samples are positive for *Listeria* spp., as an action to prevent recurrence of the contamination, you must clean and sanitize the affected surfaces and surrounding areas (§ 112.146(b)). The cleaning and sanitizing that you perform under these circumstances should be intensified beyond your routine efforts. Intensified cleaning and sanitizing include sanitation measures that you perform in addition to your normal sanitation procedures. See also Section III.F. (Cleaning and Sanitizing Conducted in Response to Suspected

or Known Contamination). You should escalate the intensity of these measures in response to additional positive samples. Intensified cleaning and sanitizing can include:

- Increasing the frequency of cleaning and sanitizing for certain pieces of equipment;
- Breaking down the equipment into its parts for further cleaning;
- The use of more effective or different cleaning and sanitizing methods or agents (e.g. more aggressive scrubbing, applying heat (where possible)) (see Section III (Cleaning and Sanitizing));
- Intensified (e.g., more vigorous, longer duration) scrubbing of surfaces;
- Identifying, cleaning, and sanitizing of possible harborage sites and possible cross-contamination routes; and
- Soaking of equipment parts in an appropriate sanitizing agent for an appropriate duration.

ii. Intensified testing

If any of the exploratory testing samples are positive for *Listeria* spp., you should, as an action to prevent recurrence of the contamination (§§ 112.146(c) and 112.146(e)), conduct another round of sampling and testing ("intensified testing"), both to verify the effectiveness of your intensified cleaning and sanitizing, and to look for possible harborage sites in the affected area.

To look for possible harborage sites, you should disassemble equipment as appropriate and sample and test areas of the equipment exposed by disassembly prior to cleaning and sanitizing the equipment. Consider the following:

1) The follow-up samples should include at least 3 samples, including the initial positive site, surrounding sites that tested positive in exploratory testing, and surrounding FCS and non-FCS sites in close proximity to any positive sites; and

2) If your intensified sampling and testing results are all negative, you should return to routine environmental monitoring. However, we recommend that you target those surfaces where *Listeria* spp. positives were previously found for sampling and testing during your next routine environmental sampling event. If you find another *Listeria* spp. positive result at the site of the initial positive or in any surrounding areas, you should take further steps as described below.

c. *Listeria* spp. positive on non-FCS during intensified testing (3rd or subsequent positive)

i. Additional activities/comprehensive investigation

If your intensified testing results in an additional positive *Listeria* spp. sample(s), as an action to prevent recurrence of the contamination (§ 112.146(e)), you should conduct additional activities to determine the source and route of contamination, including a comprehensive investigation, as

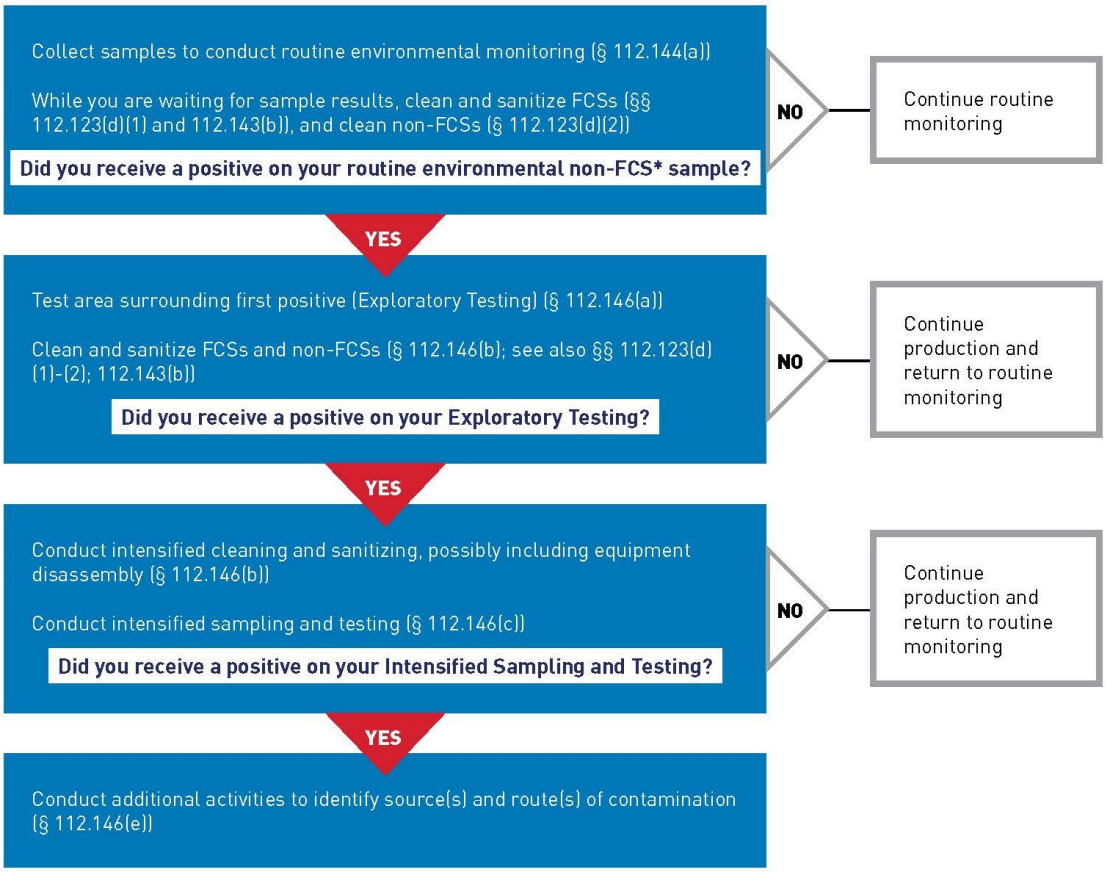
discussed in Section VI. L. 3. b (*Listeria* spp. positive on FCS during exploratory testing (2nd positive). These actions may vary depending on the risk that an FCS or food could become contaminated from the positive non-FCS site. Examples of such actions include escalating mitigation efforts to identify and eliminate the *Listeria* spp. source and consultation with a *Listeria* control expert.

The example in Figure 4 addresses testing and follow-up actions for a single positive finding of *Listeria* spp. on a Zone 2 non-FCS during one sampling period. Detecting *Listeria* spp. at several Zone 2 non-FCS sampling locations during the same sampling period could indicate that your routine sanitation procedures are inadequate and could indicate that *Listeria* spp. has become established more broadly in harborages in Zone 2. The risk associated with cross-contamination to an FCS (Zone 1) or food increases as the number of contaminated Zone 2 non-FCS sites increases. When several Zone 2 non-FCS sites test positive for *Listeria* spp. during one sampling period, we recommend that you review your written sanitation procedures to identify and implement more effective measures and escalate your corrective actions until the situation is resolved.

Figure 4. Non-FCS* Testing and Follow-Up Activities for Zone 2 Decision Tree

Non-Food Contact Surface Testing and Follow-Up Activities for Zone 2 Decision Tree

U.S. FOOD & DRUG ADMINISTRATION



* Non-FCS = Non-Food Contact Surface

M. Analysis of Data for Trends

To make the best use of the data that you collect through your environmental monitoring program, we recommend that you analyze the data (e.g., sample results, corrective actions, findings from comprehensive investigations) from the program over time for trends that can help you to continuously improve sanitation conditions and reduce the overall percentage of positive environmental samples in your operation. This trend analysis could reveal that *L. monocytogenes* is not being controlled in your operation (e.g., a resident strain has become established in a niche environment), triggering you to take additional steps to control it.

Examples of trends that could indicate that *L. monocytogenes* is not being controlled are:

- Finding *Listeria* spp. in the same area on multiple, but non-consecutive, sampling occasions (e.g., positive one week and negative the next, and then positive again a couple of weeks later, appearing to be isolated positives);
- An increase in the overall percentage of *Listeria* spp. positive samples in your operation; and
- Increases in the percentage of *Listeria* spp. positive environmental samples in particular areas of your operation.

Even if you have taken appropriate corrective actions for individual positive test results from a particular site, the continued finding of *Listeria* spp. positives at that site or in that area over time may indicate a continuing problem, such as an unidentified harborage site. If your analysis of data indicates a potential problem, such as an increased incidence of *Listeria* spp. in your operation, you should conduct a more complete investigation to determine if further actions are warranted and take appropriate corrective actions to reduce the incidence of *Listeria* spp. in your operation. A trend analysis may also lead you to update your written environmental monitoring plan.

VII. Recordkeeping

Recordkeeping is an essential component of safely growing, harvesting, packing, and holding food, including sprouts. Records can be used to ascertain safety of products, monitor storage conditions, document good agricultural practices, and track receipt and distribution of product(s). Records also offer written evidence that required conditions are being consistently met and your established procedures are being properly followed and are under control (e.g., by facilitating trend analyses). They may also be used to help determine the cause of underlying problems in the event of a foodborne illness outbreak or recall.

In this section, we provide a brief overview of the recordkeeping requirements in the Produce Safety Rule for sprout operations covered by Subpart M.

A. Recordkeeping Overview

1. General requirements

Except as otherwise specified, the requirements of Subpart O apply to all records that are required under the Produce Safety Rule. You must include the following in all required records, as applicable and unless otherwise specified:

- The name and location of your sprout operation (§ 112.161(a)(1)(i));
- The actual values and observations obtained during monitoring (§ 112.161(a)(1)(ii));
- An adequate description (such as the commodity name, or the specific variety or brand name of a commodity, and, when available, any lot number or other identifier) of covered produce applicable to the record (§ 112.161(a)(1)(iii));
- The location of a growing area or other area (for example, a specific growing bin) applicable to the record (§ 112.161(a)(1)(iv)); and
- The date and time of the activity documented (e.g., date and time of sample collection) (§112.161(a)(1)(v)).

Required records must also:

- Be created at the time an activity is performed or observed (§ 112.161(a)(2));
- Be accurate, legible, and indelible (§ 112.161(a)(3));
- Be dated, and signed or initialed by the person who performed the activity documented (§ 112.161(a)(4)); and
- Be reviewed, dated, and signed, within a reasonable time after the records required under §§112.7(b), 112.30(b)(2), 112.50(b)(2), (4), and (6), 112.60(b)(2), 112.140(b)(1) and (2), and 112.150(b)(1), (4), and (6) are made, by a supervisor or responsible party (§112.161(b)).

Records can take many forms, including photographs. However, the photos alone do not have sufficient information to meet the general requirements for a record, and the additional information listed above must be captured (see § 112.161).

a. Actual values and observations

Section 112.161(a)(1)(ii) requires records to include, as applicable, the actual values and observations obtained during monitoring. “Actual values and observations” mean that: (a) the value or observation, as applicable, itself is written (e.g., 96°F) rather than a summary term, such as “OK” or “Passed”; and (b) truthful information is recorded. For example, if you are documenting a repair as a corrective action, we recommend taking pictures before and after the repair. Another example of the recording of actual values and observations is the listing of food

and/or food contact surfaces (FCSs) that may have been affected by *Listeria* contamination, as identified during a comprehensive investigation as a corrective action following repeated *Listeria spp*. positive results on an FCS.

b. Date and time

As applicable, you must include the date and time of the documented activity on the records (§ 112.161(a)(1)(v)). Records must also be created at the time that an activity is performed or observed (§ 112.161(a)(2)). You must not pre-fill records, nor rely on your memory to write down the information later. For example, pertinent information, such as the date and time your seed treatment was applied, spent sprout irrigation water sample was collected, and spent sprout irrigation water sample was received and analyzed by the testing lab must be recorded at the time that the action is taken.

c. Adequate description of covered produce applicable to the record

You must include an adequate description of covered produce to which the record applies (§ 112.161(a)(1)(iii)). For sprouts, we recommend including a production batch lot number that includes the product name (e.g., sandwich blend), variety (e.g., alfalfa sprouts and/or clover sprouts), and brand name that your operation uses to identify the product. We recommend that you assign a unique identifier for each individual production batch of sprouts (as that term is defined in § 112.3). Using unique production batch numbers and including them on all applicable records enables you to track a production batch of sprouts internally and through distribution in the event there is a problem with that production batch.

If you use an identification system for product after it has been packaged for sale that is different from your production batch code, your ability to track the product will be enhanced if you are able to link the packed product code to the production batch of sprouts used to produce that packed product. For example, you may combine product from Production Batch "A" with product from Production Batch "B" during packing and assign product identifier #1234 to the final packed product. In this example, your ability to track the product would be enhanced if you are able to link packed product identifier #1234 to Production Batches A and B.[9]

We recommend that your records include the following information, as applicable:

- The seed lot number(s) for the seeds used to grow the production batch of sprouts (so that the lot of seeds that were used to grow each production batch of sprouts can be easily and reliably identified in the event of a problem with the production batch of sprouts);
- Container size/type;
- Date packaged;

[9] FDA published a rule to establish additional traceability recordkeeping requirements (beyond what is already required in existing regulations) for persons who manufacture, process, pack, or hold foods the Agency has designated for inclusion on the Food Traceability List. The rule will affect some sprout operations. For further information, see "Food Traceability List" at https://www.fda.gov/food/food-safety-modernization-act-fsma/food-traceability-list.

- Number of units packaged;
- Holding area; and
- Any other comments or information that may be useful in the event of a problem with the production batch of sprouts.

We also recommend that, for records of seed treatment that you perform at your operation to reduce microorganisms of public health significance (as required by §§ 112.142(e)(1) and 112.150(b)(1)), you include a description of the seeds to which the record relates. Such a description should include the type of seeds treated, and the seed lot number.

d. Location of growing area or other area applicable to the record

You must include the location of the growing area or other area applicable to the record (§ 112.161(a)(1)(iv)). For example, you could include: in your records for testing spent sprout irrigation water, the specific sprouting room and/or growing unit in which you grew the batch of sprouts being tested (e.g., Bean Sprouting Room #2, Cabin 1); in your records for seed treatment, the specific seed treatment area in which you treated the seeds; or, in your records of environmental monitoring, the particular packing line from which the sample was collected. If you have a diagram of your sprout operation, you may find it helpful for the names of the locations in the records to match the names in your diagrams.

e. Accuracy, legibility, indelibility

Required records must be accurate, legible, and indelible (§ 112.161(a)(3)). For example, if you make a mistake on a record, you should mark through the mistake with a single line, add the correct information as close as possible to the information entered in error and initial and date the correction. You should not write over the existing information, obscure it by scratching it out, or use liquid correction fluid. While handwriting styles may vary, the information on a required record must be legible so that company officials and regulatory agencies, as appropriate, can review it. You must document required information in an indelible manner (e.g., in ink) so that it cannot be erased.

f. Dated, and signed or initialed by the person who performed the activity

The individual who performed the activity documented must date, and sign or initial the required record (§ 112.161(a)(4)). Examples of such individuals include the person inspecting the agricultural water system (for records required under § 112.50(b)(1)), the worker who sanitized equipment used for growing sprouts (for records required under § 112.140(b)(1)), or the laboratory analyst who tested spent sprout irrigation water samples for microbiological contamination (for records required under § 112.150(b)(4)).

2. Duplication not required

Section 112.163(a) provides that you are not required to duplicate any existing records if those records contain all of the required information and satisfy the requirements of the Produce Safety Rule. Similarly, if you have records containing some but not all of the required information, §

112.163(b) provides you the flexibility to keep any new information required either separately or combined with your existing records, even where the formats for the records may not be the same. For example, if you already receive an annual report of your municipal water quality from your county/city, this record may meet the requirement at § 112.46(a)(2) for such public water system results, and you would not need obtain further certification.

3. Record retention and availability

You must keep required records for at least 2 years past the date the record was created (§ 112.164(a)(1)).

You must keep records that relate to the general adequacy of equipment or processes or records that relate to analyses, sampling or action plans that you use, including the results of scientific studies, tests, and evaluations, for at least 2 years after you discontinue use of such equipment or processes, or records related to analyses, sampling, or action plans (§ 112.164(b)). Examples of such records for a sprout operation include: the written sampling plan for spent sprout irrigation water or sprouts, including the corrective action plan (§§ 112.147(a) and (c) and 112.150(b)(3)); the written environmental monitoring plan including the corrective action plan (§§ 112.145(a) and (e) and 112.150(b)(2)); and documentation of scientific data or information relied on to support the adequacy of a method used to satisfy the requirements of §§ 112.43(a)(1) and (a)(2) for treating agricultural water (§ 112.50(b)(3)).

If you are eligible for the qualified exemption in accordance with § 112.5, you must retain records that you rely on during the 3-year period preceding the applicable calendar year to satisfy the criteria for a qualified exemption, in accordance with §§ 112.5 and 112.7, as long as necessary to support your exemption status during the applicable calendar year (§ 112.164(a)(2)).

In addition, you must have all required records readily available and accessible during the retention period for inspection and copying by FDA upon oral or written request, except that you have 24 hours to obtain records you keep offsite and make them available and accessible for inspection and copying (§ 112.166(a)). Offsite storage of required records is permissible, provided such records can be retrieved and provided onsite within 24 hours of request for official review (§ 112.162(a)). Electronic records are considered to be onsite at your sprout operation if they are accessible from an onsite location at your farm (§ 112.162(b)).

4. Format

As required by § 112.165, you must keep records as: (1) original records; (2) true copies; or (3) electronic records. “Original records” generally refers to the document where the information was first recorded. “True copies” include, for example, photocopies, pictures, scanned copies, microfilm, microfiche or other accurate reproductions of the original records. True copies of records should be of sufficient quality to detect whether the original record was changed in a manner that obscured an original entry (e.g., through the use of liquid correction fluid). “Electronic records” are subject to the same requirements as paper records. We do not require electronic records, nor are we specifying the form or format of the records that must be established and maintained, except as otherwise set forth in Subpart O (e.g., certain content is required when applicable as discussed in Section VII. A. 1 (General Requirements) above). To

satisfy the requirements of the Produce Safety Rule, paper or electronic records, or a combination of the two, may be used.

Records that are established or maintained to satisfy the requirements of part 112 and that meet the definition of electronic records in § 11.3(b)(6) are exempt from the requirements of part 11 (Electronic Records; Electronic Signatures). Records that satisfy the requirements of part 112, but that also are required under other applicable statutory provisions or regulations, remain subject to part 11 (§ 112.165(c)).

Although part 11 does not apply, except for records otherwise subject to part 11 (as provided in § 112.165(c)), you should take appropriate measures to ensure that electronic records are trustworthy, reliable, and generally equivalent to paper records with handwritten signatures executed on paper.

B. Supervisory Review of Records

Certain required records must be reviewed, dated, and signed by a supervisor or a responsible party within a reasonable time after the records are created (§ 112.161(b)). These records include, as applicable:

- Records related to eligibility for the qualified exemption (§ 112.7(b));
- Records related to required training of personnel (§ 112.30(b));
- Documentation of the results of all agricultural water testing (§ 112.50(b)(2));
- Documentation of the results of water treatment monitoring (§ 112.50(b)(4));
- Documentation of actions you take when your agricultural water does not meet the water quality requirements (§112.50(b)(6));
- Documentation that process controls were achieved for a treated biological soil amendment of animal origin you produce for your own covered sprout operation(s) (§ 112.60(b)(2));
- Documentation of cleaning and sanitizing of equipment (§§ 112.140(b)(1) and (2));
- Records related to seed treatment (§ 112.150(b)(1));
- Documentation of analytical test results for all sprout-specific testing (including spent irrigation water testing and environmental monitoring) done under Subpart M (§ 112.150(b)(4)); and,
- Documentation of sprout-specific corrective actions you take under Subpart M (§ 112.150(b)(6)).

The person reviewing records should first make sure that all the required records are kept and are complete, ensure that they were completed accurately and in a timely manner, consider whether

the records suggest any problems that need to be corrected, and institute any corresponding corrective actions, as necessary. The reviewer should also ensure that the observations and results are consistent with the expected range for the activity, look for any trend that could lead to problems in the future if not adequately addressed, and institute preventive measures as necessary. For example, a supervisor reviews a seed treatment record for seed to which 19,000 ppm sodium hypochlorite should be applied and available chlorine concentration monitored to ensure proper treatment. However, the supervisor notes that the recorded value of sodium hypochlorite is 500 ppm. The supervisor should follow up with the worker who made the recording to determine whether there was a malfunction with the monitoring instrument, a difficulty by the worker in reading the instrument, a recording error (i.e., the record is not accurate), or an issue of not following the seed treatment protocol.

C. Sprouts-Specific Record Requirements and Recommendations

1. Enforcement Discretion for Requirements of Subpart M (applies to soil or substrate grown sprout types that are harvested by the customer before use)

If you produce sprouts that will be sold to a commercial entity in a tray that was used for growing the sprouts, that contains the soil/substrate and roots intact, and such sprouts are then to be cut above the soil and/or substrate line at the retail or other establishment immediately before use, we intend to exercise enforcement discretion for the requirements of Subpart M provided you annually collect written assurances from your customers, stating that the sprouts will be cut above the soil and/or substrate line before use.

2. Records for Seeds for Sprouting

a. Required records for seed for sprouting

Section 112.150(b)(1) requires that you establish and keep documentation of your treatment of seeds to reduce microorganisms of public health significance in the seeds, at your sprout operation; or alternatively, documentation (such as a Certificate of Conformance) from your seed supplier that seeds are treated to reduce microorganisms of public health significance and are appropriately handled and packaged following the treatment, in accordance with the requirements of § 112.142(e). Like all records required by the Produce Safety Rule, seed treatment records must comply with all applicable requirements for records in Subpart O. For example, as applicable, these records must include:

- The name and location of your sprout operation (§ 112.161(a)(1)(i));
- Actual values and observations obtained during monitoring (§ 112.161(a)(1)(ii)) (e.g., observations of treatment conditions and key parameters monitored during preparation and application of treatments (e.g., chemical composition of treatment solution, heating temperature, equipment used, concentration of treatment solution, treatment time, pH of treatment solution, presence of agitation));

- An adequate description of the covered produce (§ 112.161(a)(1)(iii)) (e.g., the type of seed being treated, and, when available, any lot number or other identifier of the seed applicable to the record);

- The location of the area applicable to that record (§ 112.161(a)(1)(iv)) (e.g., the seed treatment room);

- The date and time of the activity documented (§ 112.161(a)(1)(v)) (e.g., the date and time the treatment was applied); and

- This record must be dated and signed or initialed by the person who performed the activity documented (§ 112.161(a)(4)) and must be reviewed, dated and signed within a reasonable time after the records are made by a supervisor or responsible party (§ 112.161(b)).

If you are relying on prior treatment of seeds by someone in the supply chain prior to your operation (e.g., a grower, distributor, or supplier of seeds), whether to fulfill the treatment requirement completely or for the purpose of considering such prior treatment when applying appropriate additional treatment at your operation in accordance with § 112.142(e)(2), you must obtain documentation (such as a Certificate of Conformance) from the grower, distributor, or supplier of seeds that the treatment was conducted using a scientifically valid method to reduce microorganisms of public health significance and that the treated seeds were handled and packaged, following the treatment, in a manner that minimizes the potential for contamination (§ 112.150(b)(1). Such records must comply with all applicable requirements for records in Subpart O. For example, as applicable, these records must:

- Be dated, and signed or initialed by the person who performed the activity documented (§ 112.161(a)(4)), e.g., a person from the grower, distributor or supplier of seeds that applied the seed treatment; and

- Be reviewed, dated and signed within a reasonable time after the records are made by a supervisor or responsible party (§ 112.161(b)). For example, a supervisor or responsible party at the sprout operation must review, date and sign the Certificate of Conformance received for prior treatment of seeds by the grower, distributor or supplier of seeds.

Section 112.150(b)(6) requires that you establish and keep records of certain corrective actions. These include those corrective actions taken in accordance with §§ 112.142(b) and (c) in response to contaminated seed and § 112.148. Such records must comply with all applicable requirements for records in Subpart O. For example, as applicable, these records must include:

- The name and location of your sprout operation (§ 112.161(a)(1)(i));

- Actual values and observations obtained during monitoring (§ 112.161(a)(1)(ii)) (e.g., date/time, and person who provided the notification to the seed supplier, the information provided, and responses from the seed supplier after notification); if conducting follow-up actions to investigate the potential source of contamination, the actual findings of those follow-up actions; if you treat an affected lot of seeds with a process that is reasonably certain to achieve destruction or elimination in seeds of the most resistant

microorganisms of public health significance, the records you keep to document this action must include the same content required for records documenting routine seed treatment (see above in this same section). Further, our recommended content for records related to routine seed treatment is also applicable to these records;

- An adequate description of the covered produce (§ 112.161(a)(1)(iii)) (e.g., the type of seed affected, and, when available, any lot number or other identifier of the seed to which the record is applicable);

- The location of the area applicable to that record (§ 112.161(a)(1)(iv)) (e.g., the seed treatment room if treating the seeds in compliance with § 112.142 (e)(1), or the areas of the sprout operation that were included in an investigation as part of a follow-up action undertaken in compliance with § 112.142(c)(2)); and

- The date and time at which you performed each activity (§ 112.161(a)(1)(v)) (e.g., notification of seed supplier, returning the affected seed lot to your supplier, destroying sprouts grown from the affected seed lot, treating seeds with a process reasonably certain to achieve destruction or elimination in seeds of the most resistant microorganisms of public health significance, or conducting follow up actions to investigate the potential source of the contamination).

Section 112.150(b)(4) requires that you establish and keep records of all analytical tests conducted for purposes of compliance with Subpart M. This includes records of test results from any testing conducted as part of follow-up actions related to suspected contamination of a seed lot with a pathogen in compliance with § 112.142(c)(2) (see also § 112.150(b)(6)).

b. Recommended records for seeds for sprouting

While not requirements of the Produce Safety Rule, we recommend these practices as prudent for seeds used for sprouting.

- To ensure seed treatments (as required under § 112.142(e)) are consistently applied correctly, if you treat seeds at your sprout operation, you should develop a written seed treatment Standard Operating Procedure (SOP). See Section V (Seeds for Sprouting) for more information.

- We recommend you develop a seed receiving program setting out the SOPs you follow to receive and inspect seed upon receipt and the criteria you use in determining whether to accept a shipment. We recommend that you use a seed receiving and inspection checklist for all incoming seed shipments to record and maintain information about the lot. An example of a seed receiving and inspection checklist can be found in Section V. A. 3 (Visual Inspection of Seeds and Their Packaging).

- To minimize the risk of contamination associated with seeds for sprouting, we recommend that you request copies of assurances from your seed suppliers that the seeds they provide were produced under GAPs and handled according to food safety best practices throughout harvesting, conditioning, storage, and transportation (Ref. 17). We recommend receiving and maintaining copies of assurances from your seed suppliers as a

prudent practice when sourcing the seeds used for sprouting and as a practice to support § 112.142(a).

- We recommend that you establish and keep sufficient records to allow you to maintain the lot identity of the seeds you receive and the sprouts you produce. We recommend that you maintain such records related to each lot of seeds for at least two years after your last use of the seed lot. See subsection C (Corrective Actions for Seeds That May be Contaminated with a Pathogen) of Section V (Seeds for Sprouting) for more information.

Seed lot numbers are typically assigned by seed suppliers. The seed lot number may appear on seed packages, or on seed shipment records. The seed lot number allows both the seed supplier and you to track seeds. Although not required by the Produce Safety Rule we recommend that you keep sufficient records to connect your test results and corrective actions to specific seed lots whenever possible. In addition, FDA published a rule to establish additional traceability recordkeeping requirements (beyond what is already required in existing regulations) for persons who manufacture, process, pack, or hold foods the Agency has designated for inclusion on the Food Traceability List. The rule will affect some sprout operations. For further information, see "Food Traceability List" at https://www.fda.gov/food/food-safety-modernization-act-fsma/food-traceability-list.

3. Required Records for Spent Sprout Irrigation Water

Sections 112.150(b)(3) and 112.147(a) require that you establish and implement a written sampling plan that identifies the number and location of samples (of spent sprout irrigation water or sprouts) to be collected for each production batch of sprouts to ensure that the collected samples are representative of the production batch when testing for contamination.

- The plan should identify: the sample type (e.g., spent sprout irrigation water, sprouts); pathogens to be tested; specific test methods to be used; person(s) responsible for collecting the sample; timing of sample collection (e.g., number of hours after start of growing cycle); procedures for aseptic technique; sampling tools to be used; laboratory to be used; and records to be kept).

- Your sampling plan must include a corrective action plan, which must, at a minimum, require you to take the actions specified in § 112.148, and must detail when and how you will accomplish those actions, if the samples of spent sprout irrigation water or sprouts test positive for *E. coli* O157:H7, *Salmonella* spp., or a pathogen meeting the criteria in § 112.144(c). (see Section V (Sampling and Testing of Spent Sprout Irrigation Water (or In-Process Sprouts)) of the draft re-issued sections (Ref. 18) for additional information on the written sampling plan)

Section 112.150(b)(5) requires you to establish and keep documentation of any analytical methods used to test for *E. coli* O157:H7 or *Salmonella* species in lieu of the methods for sprout production batch testing that are incorporated by reference in §§ 112.152 and 112.153.

- Such records should include: 1) the name and identification number of the method validated by a third party methods validation organization, or 2) the detailed analytical

procedures and information relevant to the determination of the scientific validity of the alternate method;

- In addition, if you test spent sprout irrigation water (or in-process sprouts) for any other pathogen(s) meeting the criteria in § 112.144(c), you should keep records of the scientifically valid test method you used for such testing (§ 112.153(b)).

Section 112.150(b)(4) requires that you establish and keep records of all analytical tests conducted of spent sprout irrigation water or in-process sprouts for *Salmonella* spp., *E. coli* O157:H7 as required under § 112.144(b), and any additional pathogen required under § 112.144(c). The results must be documented whether they were conducted by a third party or your own (e.g., in-house) laboratory. Records of required pathogen test results for spent sprout irrigation water (or sprouts) must comply with all applicable requirements for records in Subpart O. For example, as applicable, these records must include:

- The name and location of your sprout operation (§ 112.161(a)(1)(i));
- Actual values and observations obtained (e.g., test results from spent sprout irrigation water (or sprouts) for each pathogen, including screening and confirmatory test results (if conducted)) (§ 112.161(a)(1)(ii));
- An adequate description of covered produce applicable to the record (e.g., production batch number) (§ 112.161(a)(1)(iii));
- Location of the growing area from which the sample was collected (e.g., growing unit and growing area/room information) (§ 112.161(a)(1)(iv));
- Date and time of activity documented (e.g., date and time of sample collection, and sample receipt and analysis by the testing lab) (§ 112.161(a)(1)(v)); and
- In addition, such records must be dated, and signed or initialed by the person who performed the activity documented (e.g., the individual who conducted sample analysis (§ 112.161(a)(4)), and reviewed, dated, and signed, within a reasonable time after the records are made, by a supervisor or responsible party (§ 112.161(b)).

Section 112.150(b)(6) requires that you establish and keep records of corrective actions you take in accordance with §§ 112.142(b) and (c), 112.146 and 112.148. See subsection V. G. 3 (Documenting your corrective actions) in Section V (Sampling and Testing of Spent Sprout Irrigation Water (or In-Process Sprouts)) in the draft re-issued sections (Ref. 18).

Such records also must comply with all applicable requirements for records in Subpart O. For example, as applicable, these records must include:

- The name and location of your sprout operation (§ 112.161(a)(1)(i));
- Actual values and observations (e.g., see above Required Records for Seed for Sprouting for examples related to the seed lot; sanitizer concentration, post-cleaning and sanitizing ATP test results, observations related to actions taken to prevent reoccurrence of

contamination (e.g., employee retraining (see § 112.21(d)) and observation for compliance with procedures)) (§ 112.161(a)(1)(ii));

- An adequate description of covered produce applicable to the record (e.g., affected sprout production batch number) (§ 112.161(a)(1)(iii));

- Location of a specific growing area or other area applicable to the record (e.g., location or other identifiers for growing units and other surfaces cleaned and sanitized as part of corrective actions) (§ 112.161(a)(1)(iv));

- Date and time of activity documented (e.g., when cleaning and sanitizing or employee re-training corrective actions were conducted) (§ 112.161(a)(1)(v)); and

- In addition, such records must be dated, and signed or initialed by the person who performed the activity documented (e.g., the individual who reported positive test findings to the seed supplier; the individual who took steps to ensure a contaminated production batch of sprouts did not enter commerce (§ 112.161(a)(4)) and reviewed, dated, and signed within a reasonable time after the records are made, by a supervisor or responsible party (§ 112.161(b)).

4. Required records for environmental monitoring

Sections 112.150(b)(2) requires that you establish and keep a written environmental monitoring plan, in accordance with section 112.145, which is designed to identify *Listeria monocytogenes* if it is present in the growing, harvesting, packing, or holding environment.

- Your environmental monitoring plan must be directed to sampling and testing for either *Listeria* species or *L. monocytogenes* and your written environmental monitoring plan must include a sampling plan, including what you will test collected samples for, how often you will collect environmental samples, and a list of sample collection sites §§ 112.145(b), 112.145(c)). Your written environmental monitoring plan must include a corrective action plan that, at a minimum, requires you to take the actions in § 112.146, and details when and how you will accomplish those actions, if the growing, harvesting, packing, or holding environment tests positive for *Listeria* species or *L. monocytogenes*.

- The plan should identify the specific test methods to be used, the person(s) responsible for collecting the sample, sample collection method type (e.g., sponge vs. swab sampling, whether any samples will be composited), procedures for aseptic technique, laboratory to be used, and records to be kept.

- Section 112.150(b)(5) requires that you establish and keep records of any analytical methods you use in lieu of the methods that are incorporated by reference in §112.152(a). For example, your records could specify that you are using one of the methods that FDA listed in "Equivalent Testing Methodologies for *Listeria* species and *Listeria monocytogenes* in Environmental Samples" on our Internet site (https://www.fda.gov/food/laboratory-methods-food/equivalent-testing-methodologies-listeria-species-and-listeria-monocytogenes-environmental-samples). If the alternate method that you use is not listed by FDA, your records of any alternate analytical

methods you use in lieu of the methods that are incorporated by reference in § 112.152(a) (§ 112.150(b)(5)) should include 1) name and identification number of the method validated by a third party methods validation organization, or 2) the detailed analytical procedures, an explanation of why the alternate method is at least equivalent to the reference method (such as a scientific study, or data or information regarding the sensitivity, specificity, and precision of the alternate method compared to the reference method), and any other relevant information supporting the use of the alternate method. (see Section VI (Environmental Monitoring) of this guidance for additional information on the written environmental monitoring plan).

Section 112.150(b)(4) requires that you establish and keep records of test results from environmental testing for *Listeria* spp. or *L. monocytogenes* you perform in compliance with §§ 112.144(a) and 112.145 or any additional testing conducted if you detect *Listeria* spp. or *L. monocytogenes* in the environment in compliance with § 112.146 (a), (c), (d), and (e). The results of all analytical tests must be documented, regardless of whether they were conducted by your own (e.g., in-house) laboratories or third-party laboratories. Records of environmental monitoring tests must comply with all applicable requirements for records in Subpart O. For example, as applicable, these records must include:

- The name and location of your sprout operation (§ 112.161(a)(1)(i));
- Actual values and observations obtained (e.g., test results, including screening and confirmatory test results (if conducted), and any results for *Listeria* spp. and *L. monocytogenes*) (§ 112.161(a)(1)(ii));
- An adequate description of covered produce applicable to the record (e.g., production batch number(s) of sprouts in production at the time of the environmental sample) (§ 112.161(a)(1)(iii));
- Location of the growing area or other area(s) applicable to the record (e.g., identification of each sampling site, including whether it is an FCS or non-FCS site, description of the room or area in the sprout operation) (§ 112.161(a)(1)(iv)); and
- Date and time of the activity documented (e.g., date/time of sample collection, and sample receipt and analysis by the testing lab) (§ 112.161(a)(1)(v)).

Section 112.150(b)(6) requires that you establish and keep records of corrective actions conducted in accordance with the requirements of § 112.146. See Section VI (Environmental Monitoring) for more information. Such records must include:

- The name and location of your sprout operation (112.161(a)(1)(i));
- Actual values and observations obtained (e.g., observations and information related to a comprehensive investigation following repeated *Listeria* spp. positive results on an FCS, such as the food and FCSs potentially affected by the contamination event) (§ 112.161(a)(1)(ii));

- An adequate description of covered produce applicable to the record (e.g., production batch number of sprouts in production at the time of the corrective action) (§ 112.161(a)(1)(iii));
- Location of a growing area or other area(s) applicable to the record (e.g., identification of locations sampled or subjected to intensified cleaning and sanitizing) (§ 112.161(a)(1)(iv)); and
- Date and time of the activity documented (e.g., date/time of testing or intensified cleaning and sanitizing) (§ 112.161(a)(1)(v)).

D. Records Required by Provisions Other Than Subpart M

The records required under the Produce Safety Rule are dependent, in part, on the nature of the practices and procedures related to the covered activities in your sprout operation and are listed here under the applicable section of the Produce Safety Rule. Other required records that are relevant to sprout operations are discussed below.

Table 6. Records Required by Provisions Other Than Subpart M

Record Type	Description
Records relating to commercial processing exemption	Section 112.2(b) requires sprout operations relying on the exemption for produce that receives commercial processing that adequately reduces the presence of microorganisms of public health significance, provided for in § 112.2(b), to establish and maintain documentation of: 1) their required disclosures to customers; and 2) annual written assurances obtained from customers.
Records relating to eligibility for qualified exemption	Section 112.7 requires sprout operations eligible for the qualified exemption, provided for in § 112.5, to establish and keep adequate records necessary to demonstrate that the sprout operation satisfies the criteria for a qualified exemption (e.g., dated sales receipts), including a written record reflecting that the owner, operator, or agent in charge of the sprout operation has performed an annual review and verification of the sprout operation's continued eligibility for the qualified exemption.
Records relating to training	Section 112.30 requires you to establish and keep records that document required training of personnel, required by §§ 112.21 and 112.22, including the date of training, topics covered, and the persons(s) trained.
Records related to Agricultural water	Section 112.50(b)(1) requires you to establish and keep records of your Agricultural water system inspection findings, required by § 112.42(a); Section 112.50(b)(2) requires you to establish and keep documentation of the results of all analytical tests conducted on agricultural water for purposes of compliance with Subpart E;

Record Type	Description
	Section 112.50(b)(3) requires you to establish and keep documentation of scientific data or information you rely on to support the adequacy of a method used to satisfy the requirements of §§ 112.43(a)(1) and (a)(2) for treating agricultural water. Section 112.50(b)(4) requires you to establish and keep documentation of results of water treatment monitoring under § 112.43(b). Section 112.50(b)(6) requires you to establish and keep documentation of actions you take in accordance with§ 112.45 (measures taken if agricultural water does not meet the requirements of § 112.41 or § 112.44). Section 112.50(b)(7) requires you to establish and keep annual documentation of the results or certificates of compliance from a Public Water System required under § 112.46(a)(1) or (a)(2), if applicable.; and Section 112.50(b)(9) requires you to establish and keep documentation of any analytical methods that you use for water testing in lieu of the method that is incorporated by reference in § 112.151(a).
Records related to biological soil amendments of animal origin	Section 112.60 requires you to establish and keep certain documentation relating to any treated biological soil amendments of animal origin (BSAAO) you use (e.g., substrates). The required records differ based on whether treatment was conducted by you or by a third party. For a treated BSAAO supplied by a third party, you are required to establish and keep documentation (such as a Certificate of Conformance) at least annually that the process used to treat the BSAAO is a scientifically valid process that has been carried out with appropriate process monitoring, and that the BSAAO has been handled, conveyed, and stored in a manner and location to minimize the risk of contamination by an untreated or in process BSAAO (§ 112.60(b)(1)). For a treated BSAAO you produce for your own covered sprout operation(s), you are required to establish and keep documentation that process controls were achieved (§ 112.60(b)(2)).
Records of cleaning and sanitizing	Section 112.140 requires you to establish and keep documentation of the date and method of cleaning and sanitizing of equipment subject to Subpart L used in growing operations for sprouts and harvesting, packing, or holding for sprouts and other covered produce.

VIII. Appendices

Appendix 1. Aseptic Sampling

You must aseptically collect environmental samples (§ 112.145(d)) and samples of spent sprout irrigation water or sprouts (§ 112.147(b)). If you are required to test your agricultural water (§§ 112.44(a), 112.46(c)), you must aseptically collect agricultural water samples as well (§ 112.47(b)). Aseptic sampling is a sampling technique used to assure that the microbial load of a sample is not affected by the sampling method and/or the sample collector does not contaminate the source from which the sample is collected (including cross-contamination between sample sites). The use of sterile sampling implements and containers, and a prescribed sampling method, defines aseptic sampling (See 80 FR 74450 and references cited therein). Collected samples should also be handled in a manner to ensure samples are not contaminated during storage or during transportation to the laboratory.

Sterile Equipment

The requirements in §§ 112.47(b),112.145(d) and 112.147(b) to collect samples "aseptically" mean that you must use sterile sampling equipment to collect the required samples. Note that "sterile" is not equivalent to "clean and sanitized." Sterilization achieves a higher standard than cleaning and sanitizing. "Sterilization" refers to a validated process used to render a product free of all forms of viable microorganisms. In many cases, thermal methods, such as steam, are used to achieve sterilization (See Liquid Chemical Sterilization) (Ref. 97). "Sterile" refers to the end point achieved by a sterilizing process. You may purchase pre-packaged sterilized tools/equipment to use in sampling, or you may use (and re-use) tools and equipment for sampling that have been properly sterilized, such as in an autoclave or a dry heat oven. An autoclave is a piece of equipment that sterilizes instruments and equipment by using highly pressurized saturated steam to achieve temperatures that effectively kill microorganisms. If you decide to use an autoclave, any responsible personnel should receive adequate training prior to the use of the autoclave. When used properly, autoclaves are safe and highly effective to use to sterilize sample containers or sampling equipment (e.g., cups or tongs) and can be cost-effective. Sampling equipment, such as stainless steel forceps, spatulas, and sample containers, can be sterilized using an autoclave (steam heat), for example, at 121 °C (250 °F) for 30 minutes at 15 psi, or for heat-resistant, dry materials in a dry-heat oven, for example, at 140 °C (284°F) for 3 hours (Ref. 98). If you choose to use an autoclave, you should follow the instructions provided by the manufacturer to ensure that sterilizing of sampling tools and equipment is effective. If you choose to sterilize your own sampling tools and equipment, you should package them prior to sterilization in a manner to prevent contamination post-sterilization (e.g., wrap them with aluminum foil or Kraft paper, or placed in other suitable containers), and should only open their packaging immediately prior to use.

We recommend that you include your plans regarding use of sterilized sampling equipment in your written sampling plan for spent sprout irrigation water (or sprouts) (see Section V (Sampling and Testing of Spent Sprout Irrigation Water (or In-Process Sprouts)) in the draft re-issued sections (Ref. 18)) and your written environmental monitoring plan (see Section VI (Environmental Monitoring)).

General Aseptic Sample Collection Procedures

The requirements in §§ 112.47(b), 112.145(d), and 112.147(b) to collect samples "aseptically" also mean that, in addition to using sterile equipment for sampling, you must use a sampling method that does not affect the microbial load of the sample collected and does not contaminate the source from which the sample is collected. We recommend that you include your procedures for aseptic technique in your written sampling plan for spent sprout irrigation water (or sprouts) (see Section V (Sampling and Testing of Spent Sprout Irrigation Water (or In-Process Sprouts)) in the draft re-issued sections (Ref. 18)) and your written environmental monitoring plan (see Section VI (Environmental Monitoring)). We recommend the following aseptic techniques, which are generally applicable to any kind of sampling:

1. For activities in fully enclosed buildings, a sample collector should wear a clean lab coat, sterile gloves, and a hair net to ensure he or she does not contaminate the samples.

2. Hands should be washed immediately before sampling, and prior to putting on sterile gloves. Gloves should be put on in a manner that does not contaminate the outside of the glove. Gloves should be properly disposed of after use.

3. To prevent cross contamination, gloves should be changed between samples. In addition, you should change gloves if you touch any surfaces other than the sample sites, such as garbage, drains, or the floor.

4. Hands should be kept away from mouth, nose, eyes, and face while collecting samples. You should not cough or sneeze in the vicinity of the samples, and if you do, discard the affected sample(s).

5. Sampling instruments should be protected from contamination at all times before and during use. Either use them only once or sterilize them in between uses, so each sample will be taken by fresh and sterile utensils.

6. The type of sample containers used (e.g., bags, tubes, cups, flasks) should depend on the type of sample collected. You should use containers that are dry, leak-proof, wide-mouthed, and of a size suitable for the type of sample collected.

    - The container should be properly labeled, such as with a marked strip of masking tape, prior to sampling to identify the sample information, such as the sample production batch, time, and the date of sampling. The container should be opened only sufficiently to collect the sample directly in the container, and then immediately closed and sealed.

    - Containers such as plastic jars or metal cans that are leak-proof may be sealed.

    - If collecting samples in a container with a lid, the lid should not be placed on a counter.

    - Whenever possible, avoid using glass containers, which may break and contaminate the sample, the source from which the sample was taken, and/or covered produce,

including sprouts.

7. Sample containers for water samples (i.e., agricultural water and spent sprout irrigation water) should not be overfilled, and you should leave an air space of 1 to 2 inches at the top to prevent overflow.

8. For samples collected indoors, samples and sampling equipment should not be exposed to unfiltered air currents. When opening sterile sampling containers, you should work rapidly, open sterile sampling containers only to admit the sample and close it immediately.

9. You should not touch the inside of the sterile sample container, lip or lid. You should not allow fingers or anything except the sample to contact the inside of the sample container.

10. You should not use a sample container that has fallen on the floor.

11. You should not expose covered produce, including sprouts, or food contact surfaces to samples or hands after sampling. You must wash hands with soap and water following sample collection, because during sampling hands may have become contaminated in a manner reasonably likely to lead to contamination of covered produce (§ 112.32(b)(3)(vi)).

12. Samples should be delivered to the laboratory promptly. You should ship the samples so that they are received by the laboratory within 24 hours from the time of sample collection and analyzed promptly. A delay of more than 24 hours between sample collection and the lab's receipt may make the test results inaccurate (Ref. 99).

13. Prior to and during delivery or shipment to a laboratory, you should hold your samples between 0 and 4.4 ºC (between 32 and 40 ºF). You should use sealed coolant packs in lieu of ice, as needed during delivery or shipment, to avoid the possibility of melting ice contaminating the sample. You should not freeze the samples.

Appendix 2. Collection Procedures – Environmental Sampling

The two most common methods to collect environmental samples to test for *Listeria* spp. or *Listeria monocytogenes* are "surface sponging" and "swabbing." Another sampling method used is the "rinse" method. In general, the preferred method of sampling is surface sponging, but certain areas could be more appropriately sampled using a swab (e.g., head screws, small water collection points, screw holes, threaded surfaces or interior corners of equipment) or rinse method. For wet surfaces, you should wipe and absorb moisture and wet product and residue with a sponge. For dry surfaces, you should wipe the sample site area with a sponge or swab moistened with Dey-Engley (D/E) broth. The sample size of each site should be consistent with the testing methodology and the sample collection method being used. The recommended sample size for swabs is generally smaller (e.g., 1 inch by 1 inch) compared to sponges (e.g., 4 inches by 4 inches up to 12 inches by 12 inches). However, if collecting from surfaces with visible residue (e.g., dust, dirt, buildup of organic material), we recommend increasing the number of sponges and collecting from a smaller surface area for each sponge to improve the likelihood of detection.

If Sponge Sampling:

1. You should wash and sanitize hands to the mid-forearm before you begin sampling. Aseptically place a glove on the hand used for swabbing.

2. Using the ungloved hand, open the bag containing the sponge on a stick by pulling off the clear perforated strip at the top of the bag.

3. Pull apart the white tabs to open the mouth of the bag.

4. If not using pre-moistened sponges, aseptically pour 9-10 ml of sterile D/E or other neutralizing broth into the bag to hydrate the sponge, being careful not to contaminate the broth or sponge during the transfer.

5. Close the bag and evenly moisten sponges by hand massage.

6. Position the sponge so that the handle is sticking out of the bag and close the bag around the stick.

7. Through the bag, squeeze the excess broth gently out of the sponge. Do not let your hand go past the thumb stop on the stick.

8. Carefully take the sponge-stick out of the bag by grasping the stick and swab the area selected using firm and even pressure. Be careful to maintain sanitary conditions when sampling. Do not let your hand go past the thumb stop on the handle.

9. Swab the chosen area (e.g., between 4 inches by 4 inches up to 12 inches by 12 inches square of food contact or environmental surface area):

    - Sponge vertically (approximately 10 times); then

    - Flip the sponge and use the other side to swab horizontally (approximately 10 times);

    - Then move the sponge diagonally, using the same surface side as you used for horizontal (approximately 10 times).

10. Open the bag and insert the sponge portion into the bag.

11. Grip the sponge through the bag and bend the stick of the sponge back and forth with slight force, while gripping the sponge through the bag. The stick should break easily within the sponge. (Do not break the stick at the thumb stop.) Discard the broken stick. If the stick is sticking out above the sponge, discard this sample.

12. Squeeze as much air out of the bag as possible and fold the top of the bag down at least 3 times until it is folded all the way down to the sponge. Fold in the tabs to lock the fold in place.

13. Label the bag with the date and location of the sample.

14. Take each new sample following the same steps in 1 – 13, starting by changing the glove on the gloved hand between samples.

15. Prior to and during delivery or shipment to a laboratory, samples should be kept refrigerated. Sealed coolant packs should be used in lieu of ice, as needed during delivery or shipment, to avoid the possibility of melting ice contaminating the sample. Samples should not be frozen. Samples should be shipped so that they are received by the laboratory within 24 hours after sample collection. We recommend that the maximum timeframe between environmental sampling and analysis be 48 hours (Ref. 95).

If Swab Sampling:

1. You should wash and sanitize hands to the mid-forearm before you begin sampling. Aseptically place a glove on the hand used for swabbing.

2. Using the ungloved hand, open the tube containing the swab.

3. If not using pre-moistened swabs, aseptically pour 9-10 ml of sterile D/E or other neutralizing broth into the tube to hydrate the swab or use sampling swabs that are already pre-moistened in 9-10 ml of D/E broth. If the D/E broth is not purple, discard the tube.

4. Close the cap of the tube and wait until the swab is moistened.

5. Carefully take the swab out of the tube and swab the area selected using firm and even pressure. Be careful to maintain sanitary conditions when sampling.

6. Swab the chosen area (e.g., 1 inch by 1inch square of food contact or environmental surface area, or insert the swab into the crack, crevice or hole) using firm and even pressure:

    - Swab the 1 inch by 1inch surface vertically (approximately 10 times); then

    - Rotate the swab and use the other side to swab horizontally (approximately 10 times); then

    - Move the swab diagonally, using the same surface side as you used for horizontal (approximately 10 times).

    - For cracks, crevices and holes, rotate the swab in the area as you move it around the area.

7. Do not touch the outside of the opening of the tube; insert the swab portion into the tube.

8. Close the cap of the tube.

9. Label the bag with the date and location of the sample.

10. Take each new sample following the same steps in 1 – 9, starting by changing the glove on the gloved hand between samples.

11. Prior to and during delivery or shipment to a laboratory, samples should be kept refrigerated. Sealed coolant packs should be used in lieu of ice, as needed during delivery or shipment, to avoid the possibility of melting ice contaminating the sample. Samples should not be frozen. Samples should be shipped so that they are received by the laboratory within 24 hours after sample collection. We recommend that the maximum timeframe between environmental sampling and analysis be 48 hours (Ref. 95).

Collecting Rinse Samples

To collect samples using a rinse technique, you should add small pieces from equipment (such as screws, nuts or gaskets) directly to the bag containing D/E broth and hand massage the bag for sufficient time to remove soil and residues (approximately 1 minute). Then aseptically remove the items from the bag and subject the broth to analysis.

In some situations, involving small cracks and crevices, it may help to use a plastic bulb transfer pipette. You should use tubes containing 10 mL sterile D/E broth in this procedure. You should pull the D/E broth into the pipette bulb and transfer the D/E broth to the crack or crevice, and then you should pull it back into the bulb. You should repeat this several times to thoroughly rinse the crack or crevice.

Collecting Liquid Samples (Including Floor Drain Effluents)

We recommend that you use a sterile plastic beaker or similar container to collect 110 + 5 ml of liquids, where possible, such as drainage effluents, standing water, melt water from thawed processing ice, and vacuum or drip pan condensate. We recommend that you immediately transfer the collected sample into a sterile screw-capped bottle and then chill and store the bottle at 5 degrees C (41 degrees F), including during transport to the testing laboratory.

Preparing Samples Collected from Liquids

For larger samples (e.g., 100 mL or greater), we recommend that you filter 100 ml of the collected liquid through one or more sterile 0.45 micron pore-diameter filters as soon as possible after sample collection. If particulate content is high (e.g., judging from the sample turbidity), we recommend that you pass the liquid through a sterile glass pre-filter before the 0.45 micron filter. You should rinse a retained substance on the filter plus any pre-filter with 5-10 ml of D/E broth to remove any residual inhibitory substances. If necessary, you should cut the filters from the funnel devices, using sterile scalpels. You should put each filter and the pre-filter, if any, in a sterile bag (if you will use a Stomacher) or in a sterile container (such as a blender jar, if you use a blender). You should add 225 mL of UVM broth, and follow procedures in "Testing Methodology for *Listeria* species of *L. monocytogenes* in Environmental Samples" (version 1, Oct 2015) (currently available at: https://www.fda.gov/media/94358/download) beginning with incubation of the primary enrichment.

For small volumes of liquid samples, we recommend that you add the liquid sample to 225 mL of UVM broth and follow procedures in "Testing Methodology for *Listeria* species of *L.*

monocytogenes in Environmental Samples" (version 1, Oct 2015) beginning with incubation of the primary enrichment.

Appendix 3. Potential Sources of *L. monocytogenes* for Sampling in a Sprout Operation

This table provides examples of possible food contact and non-food contact surfaces for use in developing a *Listeria* environmental monitoring program. The list is not all-inclusive and is not intended to be determinative; rather, we provide the examples below as an aid. We recommend that firms develop their own robust assessments of the food contact and non-food contact surfaces in their operations, as well as an understanding of appropriate zone designations.

Table 7. Potential Sources of *L. monocytogenes* in a Sprout Operation[10]

Sample Location	Zone 1	Zone 2	Zone 3	Zone 4	Notes
Aprons	X	X			Aprons reasonably likely to contact sprouts would be considered Zone 1.
Conveyors	X	X			Fibrous and porous-type conveyor belts, hollow rollers for conveyances are of particular concern.
Ceilings and shields from which condensate or drainage can drip onto the product	X				For the purpose of your environmental monitoring program, we recommend you focus on the surfaces that directly contact the food
Ceilings that are not above sprouts, overhead structures, and catwalks		X	X		Condensate drip pans are of particular concern.
Gloves	X	X			Gloves that are reasonably likely to contact sprouts would be considered Zone 1.
Growing bins or trays	X				
Rotary drums	X				

[10] The table has been added to the document to facilitate a better understanding of food contact surfaces (FCSs) and non-FCSs, and zone designations. The contents are based on information that was available in the draft "Guidance for Industry: Compliance with and Recommendations for Implementation of the Standards for the Growing, Harvesting, Packing, and Holding of Produce for Human Consumption for Sprout Operations" (Ref. 100), and are now presented in a more user-friendly format.

Sample Location	Zone 1	Zone 2	Zone 3	Zone 4	Notes
Spin dryer (for sprouts)	X				
Tools for cleaning equipment		X			e.g., Brushes and scouring pads, cracked hoses
Utensils	X				
Work table	X	X			The surface that is reasonably likely to contact sprouts is Zone 1; the underside and supports are Zone 2.
Walls		X	X	X	Zone depends on proximity to food contact surfaces and food.
Equipment housing or framework		X	X		Wet, rusting, or hollow framework, open bearings within equipment, motor housings, on/off switches are areas of particular concern.
Pallets		X	X		
Wash areas	X	X	X		e.g., Washing tubs, sinks
Maintenance tools		X	X		e.g., Wrenches and screw drivers
Coolers		X	X		Wet insulation in walls or around pipes and cooling units, rubber seals around doors are of particular concern.
Floors and drains in the immediate vicinity of FCSs		X			
Floors or drains in or near the production areas but not in the immediate vicinity of FCSs			X		
Racks for trays		X			

Sample Location	Zone 1	Zone 2	Zone 3	Zone 4	Notes
Surfaces employees handle prior to handling the product		X			
Forklifts			X	X	
Vacuum cleaners and floor scrubbers			X	X	
Trash cans			X	X	
Hand trucks and carts that move in the production area			X		
Cafeterias				X	
Hallways outside the production area				X	
Locker rooms				X	
Areas outside of the production area in which raw materials (e.g., seeds) or finished products are stored or transported		X	X	X	

IX. References

The following references marked with an asterisk (*) are on display at the Dockets Management Staff (HFA-305), Food and Drug Administration, 5630 Fishers Lane, Rm. 1061, Rockville, MD 20852, 240-402-7500, and are available for viewing by interested persons between 9 a.m. and 4 p.m., Monday through Friday; they also are available electronically at https://www.regulations.gov. References without asterisks are not on public display at https://www.regulations.gov because they have copyright restriction. Some may be available at the website address, if listed. References without asterisks are available for viewing only at the Dockets Management Staff. FDA has verified the website addresses, as of the date this document publishes in the Federal Register, but websites are subject to change over time.

Ref. 1. Food and Drug Administration. 1998. "Guidance for Industry: Guide to Minimize Microbial Food Safety Hazards for Fresh Fruits and Vegetables." Accessed October 14, 2021. (http://www.fda.gov/Food/GuidanceRegulation/GuidanceDocumentsRegulatoryInformation/ucm064574.htm)*

Ref. 2. National Advisory Committee on Microbiological Criteria for Foods. 1999. "Microbiological Safety Evaluations and Recommendations on Sprouted Seeds." *International Journal of Food Microbiology* 52 (3):123-153.

Ref. 3. D'Lima, C., et al. "Memorandum to the File: Produce Related Outbreaks and Illnesses, December 2011." Food and Drug Administration.*

Ref. 4. Merriweather, S., et al. "Memorandum to the File: 2011-2014 Produce Related Outbreaks and Illnesses, May 2015." Food and Drug Administration.*

Ref. 5. Gubernot, D., et al. "Memorandum to the File: 2015-2016 Sprout and Sprout-Derived Product Related Outbreak Data, July 2016." Food and Drug Administration.*

Ref. 6. Obenhuber, D., et al. "Memorandum to the File: August 2018-August 2020 Sprout Related Outbreak Data, October 13, 2020." Food and Drug Administration.*

Ref. 7. Fatica, Marianne K., et al. "Memorandum to the File: July-December 2016 Sprout Related Outbreak Data, February 16, 2017." Food and Drug Administration.*

Ref. 8. Fatica, Marianne K., et al. "Memorandum to the File: January 2017-August 2018 Sprout Related Outbreak Data, September 12, 2018." Food and Drug Administration.*

Ref. 9. Centers for Disease Control and Prevention (CDC). "National Outbreak Reporting System (NORS)." Last updated December 7, 2018. Accessed on October 7, 2021. (https://wwwn.cdc.gov/norsdashboard/)*

Ref. 10. Foley, Catherine, et al. "Outbreak of *Escherichia coli* O104: H4 infections associated with sprout consumption—Europe and North America, May–July 2011." *MMWR. Morbidity and mortality weekly report* 62.50 (2013):1029-1031.

Ref. 11. Obenhuber, D., et al. "Memorandum to the file: 2012-2020 Sprout-Related Outbreak Data, July 20, 2020." Food and Drug Administration.*

Ref. 12. Food and Drug Administration. 2014. "FDA Form 483, Inspectional Observations: Wholesome Soy Products Inc." Accessed August 24, 2021. (https://www.fda.gov/media/90409/download)*

Ref. 13. Food and Drug Administration. 2020. "Warning letter: Sprout Unlimited Inc." Accessed August 24, 2021. (https://www.fda.gov/inspections-compliance-enforcement-and-criminal-investigations/warning-letters/sprouts-unlimited-inc-603883-02212020)*

Ref. 14. Food and Drug Administration. 2020. "Waning letter: Chicago Indoor Garden, Inc." Accessed August 24, 2021. (https://www.fda.gov/inspections-compliance-enforcement-and-criminal-investigations/warning-letters/chicago-indoor-garden-inc-606992-07302020)*

Ref. 15. Food and Drug Administration. 2009. "Letter from FDA to Seed Suppliers, Distributors, and Sprouters." Accessed October 18, 2021. (http://wayback.archive-it.org/7993/20180908120136/https://www.fda.gov/Food/GuidanceRegulation/GuidanceDocumentsRegulatoryInformation/ProducePlantProducts/ucm078778.htm)*

Ref. 16. Food and Drug Administration, 1999. "Guidance for Industry: Sampling and Microbial Testing of Spent Irrigation Water During Sprout Operation." (withdrawn) Accessed February 14, 2023. (https://www.ifsh.iit.edu/sites/ifsh/files/departments/ssa/pdfs/fda1999_spent_irrigation_water.pdf)*

Ref. 17. Food and Drug Administration. 2022. "Guidance for Industry: Reducing Microbial Food Safety Hazards in the Production of Seed for Sprouting." Accessed October 28, 2022. (https://www.fda.gov/media/127972/download)*

Ref. 18. Food and Drug Administration. 2023. "Draft Guidance for Industry: Standards for the Growing, Harvesting, Packing, and Holding of Sprouts for Human Consumption." Accessed XXXXX. (Website)*

Ref. 19. Codex. 2010. "Code of Hygienic Practice for Fresh Fruits and Vegetables, CAC/RCP 53-2003, Annex II, Annex for Sprout Production." Accessed February 14, 2023. (https://www.fao.org/fao-who-codexalimentarius/sh-proxy/en/?lnk=1&url=https%253A%252F%252Fworkspace.fao.org%252Fsites%252Fcodex%252FStandards%252FCXC%2B53-2003%252FCXC_053e.pdf)

Ref. 20. IFSH Sprout Safety Task Force Best Practices Sub-committee. 2019. "US Sprout Industry Production Best Practices." Accessed October 13, 2021.

(https://www.ifsh.iit.edu/sites/ifsh/files/departments/Copyrighted%20US%20Sprout%20Industry%20Production%20Best%20PractiRef.ces%20052619.pdf)

Ref. 21. The European Sprouted Seeds Association (ESSA). 2017. "ESSA Hygiene Guideline for the Production of Sprouts and Seeds for Sprouting (2017/(220/ 03), Section 2. Production of Seeds." Official Journal of the European Union. Accessed October 13, 2021. (https://eur-lex.europa.eu/legal-content/EN/TXT/HTML/?uri=CELEX:52017XX0708(01)&from=EN)*

Ref. 22. Food and Drug Administration. 2018. "Guidance for Industry: Standards for the Growing, Harvesting, Packing, and Produce for Human Consumption." Last modified October 22, 2018. Accessed October 27, 2022. (https://www.fda.gov/regulatory-information/search-fda-guidance-documents/draft-guidance-industry-standards-growing-harvesting-packing-and-holding-produce-human-consumption)*

Ref. 23. California Department of Public Health and Food and Drug Administration. 2000. "Safer Processing of Sprouts" Video Series. Accessed October 18, 2021. (https://www.ifsh.iit.edu/ssa/resources)

Ref. 24. Sprout Safety Alliance. 2017. "Safer Sprout Production for Produce Safety Rule Compliance. Version 2.3." Accessed October 13, 2021. (https://d1vy0qa05cdjr5.cloudfront.net/c6f30ca0-84ae-4613-bec0-5439702d4b9e/FSPCA%20-%20Sprouts/SSA%20curriculum%20V2.3%20-%20For%20PRINT%20watermark%20optimized.pdf)

Ref. 25. Institute for Food Safety and Health. "IIT IFSH Launches FDA Funded Sprout Safety Alliance." Accessed October 14, 2021. (https://www.ifsh.iit.edu/news/2012/02/iit-ifsh-launches-fda-funded-sprout-safety-alliance)*

Ref. 26. Food and Drug Administration. 2015. "FDA's Strategy for FSMA Training." Accessed October 14, 2021. (http://www.fda.gov/Food/GuidanceRegulation/FSMA/ucm461513.htm)*

Ref. 27. Sprout Safety Alliance. 2023. "Food Safety Best Practices for Sprout Production" Video Series. Accessed XX. (https://www.ifsh.iit.edu/ssa/resources)

Ref. 28. Gradl, D.R., et al. 2015. "Survival of *Salmonella* during Drying of Fresh Ginger Root (Zingiber officinale) and Storage of Ground Ginger." *Journal of Food Protection* 78 (11):1954-1959.

Ref. 29. Wang, C., et al. 2016. "Growth and Survival of *Salmonella* in Partially Sprouted Pumpkin, Sunflower, and Chia Seeds Subsequently Dried for Direct Consumption." Poster presented at the Institute for Food Technologists Conference in Chicago, IL, July.

Ref. 30. Liu, B., et al. 2007. "Mathematical Modeling and Assessment of Microbial Migration during the Sprouting of Alfalfa in Trays in a Nonuniformly Contaminated Seed Batch Using Enterobacter aerogenes as a Surrogate for *Salmonella* Stanley." *Journal of Food Protection* 70 (11):2602–2605.

Ref. 31. Hora, R., et al. 2005. "Spatial Distribution of *Salmonella*, *Escherichia coli* O157:H7, and Other Bacterial Populations in Commercial and Laboratory-Scale Sprouting Mung Bean Beds." *Journal of Food Protection* 68 (12):2510–2518.

Ref. 32. Food Standards Australia New Zealand. 2011. "Proposal P1004, Primary Production and Processing Standard for Seed Sprouts, Approval Report."

Ref. 33. Smith, M.A. 2014. "Evolution of FDA's Policy: Sprouts Versus Microgreens." Presentation in Symposium S31, in Microgreens and Sprouts under Microscope: Similarities and Differences in Botanic Structure, Agricultural Practices, and Food Safety Risks. Annual Meeting of the International Association for Food Protection, August 5. Indianapolis, IN.

Ref. 34 Tompkin, R.B., et al. 1999. "Guidelines to Prevent Post-Processing Contamination from *Listeria monocytogenes*." *Dairy, Food and Environmental Sanitation* 19 (8):551-562.

Ref. 35. Marriott, N. and R.B. Gravani. 2006. "Fruit and Vegetable Processing Plant Sanitation." *Principles of Food Sanitation, 5th edition,* edited by N.G. Marriott and R.B. Gravani, pp 336-349. New York, New York: Springer.

Ref. 36. Stanfield. P. 2002. "Cleaning and Sanitizing a Food Plant." *Food Plant Sanitation, 1st edition*, edited by Y.H. Hui et al., pp 101-114. New York, New York: Marcel Dekker, Inc.

Ref. 37. Dunsmore, D.G., et al. 1981. "Design and Performance of Systems for Cleaning." *Journal of Food Protection* 44 (3):220-240.

Ref. 38. Dvorak, G. 2008. "Disinfection 101." Center for Food Security and Public Health. Accessed February 13, 2023. (https://www.cfsph.iastate.edu/Disinfection/Assets/Disinfection101.pdf)

Ref. 39. Kopper, G., et al. 2013. "Hygiene in Primary Production." *Food Safety Management, 1st edition*, edited by Y. Motarjemi and H. Lelieveld, pp 559-621. Waltham, Massachusetts: Academic Press.

Ref. 40. Marriott, N. and R.B. Gravani. 2006. "Sanitizers." *Principles of Food Sanitation, 5th edition*, edited by N.G. Marriott and R.B. Gravani, pp 165-189. New York, New York: Springer.

Ref. 41. Birkenhauer, E., et al. 2015. "Prevention and Control of Biofilms in the Food Industry and Bio-Nanotechnology Approaches." *Biofilms in the Food Environment, 2nd edition*, edited by A.L. Pometto III and A. Demirci, pp 84-130. United Kingdom: Wiley Blackwell.

Ref. 42. McEgan, R., et al. 2008. "Composite versus Single Sampling of Spent Irrigation Water to Assess the Microbiological Status of Sprouting Mung Bean Beds." *Journal of the Science of Food and Agriculture*, 88 (9):1549-1553.

Ref. 43. Hara-Kudo, Y., et al. 1997. "Potential Hazard of Radish Sprouts as a Vehicle of *Escherichia coli* O157:H7." *Journal of Food Protection* 60 (9):1125-1127.

Ref. 44. Suslow, T.V. 1997. "Postharvest Chlorination: Basic Properties and Key Points for Effective Disinfection." University of California, Division of Agriculture and Natural Resources." Publication 8003. Accessed October 15, 2021. (https://ucfoodsafety.ucdavis.edu/sites/g/files/dgvnsk7366/files/inline-files/26414.pdf)

Ref. 45. Suslow, T.V. 2001. "Water Disinfection: A Practical Approach to Calculating Dose Values for Preharvest and Postharvest Applications." The University of California, Division of Agriculture and Natural Resources. Publication 7256. Accessed October 15, 2021. (https://ucanr.edu/sites/Postharvest_Technology_Center_/files/232177.pdf)

Ref. 46. Ritenour, M.A., et al. 2002. "Chlorine Use in Produce Packing Lines." Institute of Food Safety and Agricultural Sciences, University of Florida. HS-761. Accessed October 14, 2021. (https://journals.flvc.org/edis/article/view/108301/103581)

Ref. 47. University of Florida IFAS Extension. "Citrus Postharvest and Handling." Accessed October 15, 2021. (http://edis.ifas.ufl.edu/topic_citrus_post_harvest)

Ref. 48. EFSA Panel on Biological Hazards (BIOHAZ). 2011. "Scientific Opinion on the Risk Posed by Shiga Toxin Producing *Escherichia coli* (STEC) and Other Pathogenic Bacteria in Seeds and Sprouted Seeds." EFSA Journal, 9(11):2424.

Ref. 49. Bazaco, M.C., et al. 2021. "An Overview of Historic Foodborne Illness Outbreak Investigations Linked to the Consumption of Sprouts: 2012-2020." Food Safety Magazine. June/July. Accessed September 23, 2021. (https://www.food-safety.com/articles/7202-an-overview-of-historic-foodborne-illness-outbreak-investigations-linked-to-the-consumption-of-sprouts-20122020)*

Ref. 50. Winthrop, K.L., et al. 2003. "Alfalfa Sprouts and Salmonella Kottbus Infection: A Multistate Outbreak following Inadequate Seed Disinfection with Heat and Chlorine." *Journal of Food Protection* 66 (1):13-17.

Ref. 51. Gill, C.J., et al. 2003. "Alfalfa Seed Decontamination in a *Salmonella* Outbreak." *Emerging Infectious Diseases* 9 (4):474-479.

Ref. 52. Brooks, J.T., et al. 2001. "*Salmonella* Typhimurium Infections Transmitted by Chlorine-pretreated Clover Sprout Seeds." *American Journal of Epidemiology* 154 (11):1020-1028, 2001.

Ref. 53. Montville, R., et al. 2005. "Monte Carlo Simulation of Pathogen Behavior During the Sprout Production Process." *Applied and Environmental Microbiology* 71 (2):746-753.

Ref. 54. Fett, W.F. 2002. "Factors Affecting the Efficacy of Chlorine against *Escherichia coli* O157:H7 and *Salmonella* on Alfalfa Seed." *Food Microbiology* 19:135-149.*

Ref. 55. Chen, Y., et al. 2018. "Risk Assessment of Salmonellosis from Consumption of Alfalfa Sprouts and Evaluation of the Public Health Impact of Sprout Seed Treatment and Spent Irrigation Water Testing." *Risk Analysis* 38 (8): 1738-1757.*

Ref. 56. Weiss, A., et al. 2005. "Efficacy of Heat Treatment in the Reduction of *Salmonellae* and *Escherichia coli* O157:H– on Alfalfa, Mung bean and Radish Seeds Used for Sprout Production." *European Food Research and Technology* 221 (1):187-191.

Ref. 57. Bari, M.L., et al. 2010. "Practical Evaluation of Mung Bean Seed Pasteurization Method in Japan." *Journal of Food Protection* 73 (4):752-757.

Ref. 58. Bari, L., et al. 2010. "Scale-Up Seed Decontamination Process to Inactivate *Escherichia coli* O157:H7 and *Salmonella* Enteritidis on Mung Bean Seeds." *Foodborne Pathogen and Disease* 7 (1):51-56.

Ref. 59. Environmental Protection Agency. "Pesticide Registration Manual: Chapter 13 - Devices." Accessed October 14, 2021. (https://www.epa.gov/pesticide-registration/pesticide-registration-manual-chapter-13-devices)*

Ref. 60. National Pesticide Information Center. "State Pesticide Regulatory Agencies." Accessed October 14, 2021. (http://npic.orst.edu/reg/state_agencies.html)

Ref. 61. Ding, H., et al. 2013. "Microbial Contamination in Sprouts: How Effective Is Seed Disinfection Treatment?" *Journal of Food Science* 78 (4):R495-R501.*

Ref. 62. Neetoo, H., et al. 2011. "Individual and Combined Application of Dry Heat with High Hydrostatic Pressure to Inactivate *Salmonella* and *Escherichia coli* O157:H7 on Alfalfa Seeds." *Food Microbiology* 28 (1):119-127.

Ref. 63. Johanson, J., et al. "Memorandum to the File: Review of Literature on Methods for Treatment of Seed Intended for Sprouting, October 2015." Food and Drug Administration.*

Ref. 64. Rajkowski, K.T., et al. 2012. "Irradiation of Seeds and Sprouts." *Food Irradiation Research and Technology, 2nd Edition*, edited by X. Fan and C.H. Sommers, pp 295-316. Hoboken, NJ, USA: John Wiley & Sons, Inc.

Ref. 65. Sikin, A.M., et al. 2013. "Current Intervention Strategies for the Microbial Safety of Sprouts." *Journal of Food Protection* 76 (12):2099-2123.

Ref. 66. Trząskowska, M., et al. 2018. "Pathogen reduction on mung bean reduction of *Escherichia coli* O157: H7, *Salmonella enterica* and *Listeria monocytogenes* on Mung Bean Using Combined Thermal and Chemical Treatments with Acetic Acid and Hydrogen Peroxide." *Food Microbiology* 76:62-68.

Ref. 67. Liao, C.H. 2009. "Acidified Sodium Chlorite as an Alternative to Chlorine for Elimination of Salmonella on Alfalfa Seeds." *Food Microbiology and Safety* 74 (4):M159-M164.*

Ref. 68. Holliday, S.L., et al. 2001. "Efficacy of Chemical Treatments in Eliminating *Salmonella* and *Escherichia coli* O157:H7 in Scarified and Polished Alfalfa Seeds." *Journal of Food Protection* 64 (10):1489-1495.

Ref. 69. Chang, S., et al. "Inactivation of *Escherichia coli* O157:H7 and *Salmonella* spp. on Alfalfa Seeds by Caprylic acid and Monocaprylin." *International Journal of Food Microbiology* 144:141-146, 2010.

Ref. 70. Nei, D., et al. 2011. "Disinfection of Radish and Alfalfa Seeds Inoculated with *Escherichia coli* O157:H7 and *Salmonella* by a Gaseous Acetic Acid Treatment." *Foodborne Pathogens and Disease* 8 (10):1089-1094.

Ref. 71. Nei, D., et al. 2014. "Large-Scale Gaseous Acetic Acid Treatment to Disinfect Alfalfa Seeds Inoculated with *Escherichia coli*." *Foodborne Pathogens and Disease* 11 (4):332-334.

Ref. 72. Beuchat, L.R. 1997. "Short Communication: Comparison of Chemical Treatments to Kill *Salmonella* on Alfalfa Seeds Destined for Sprout Production." *International Journal of Food Microbiology* 34:329-333.

Ref. 73. Environmental Protection Agency (EPA). 2018. Pesticide Product Label, Registration Number 5813-114. Accessed October 14, 2021. (https://www3.epa.gov/pesticides/chem_search/ppls/005813-00114-20181030.pdf)*

Ref. 74. Buchanan, R.L. 2000. "Acquisition of Microbiological Data to Enhance Food Safety." *Journal of Food Protection* 63 (6):832-838.*

Ref. 75. Goulet, V., et al. 2012. "Incidence of Listeriosis and Related Mortality Among Groups at Risk of Acquiring Listeriosis." *Clinical Infectious Diseases* 54 (5):652-660.

Ref. 76. Pouillot, R., et al. 2012. "Relative Risk of Listeriosis in Foodborne Diseases Active Surveillance Network (FoodNet) Sites According to Age, Pregnancy and Ethnicity." *Clinical Infectious Diseases* 54 (S5):S405-S410.*

Ref. 77. Pouillot, R., et al. 2015. "*Listeria monocytogenes* Dose Response Revisited - Incorporating Adjustments for Variability in Strain Virulence and Host Susceptibility." *Risk Analysis* 35 (1):90-108.*

Ref. 78. Food and Drug Administration, and USDA Food Safety and Inspection Service. 2003. "Quantitative Assessment of Relative Risk to Public Health from Foodborne *Listeria monocytoge*nes Among Selected Categories of Ready to Eat Foods." Accessed October 14, 2021. (https://www.fda.gov/food/cfsan-risk-safety-assessments/quantitative-assessment-relative-risk-public-health-foodborne-listeria-monocytogenes-among-selected)*

Ref. 79. Centers for Disease Control and Prevention. 2015. "Wholesome Soy Products, Inc. Sprouts and Investigation of Human Listeriosis Cases (Final Update)." Accessed October 14, 2021. (https://www.cdc.gov/listeria/outbreaks/bean-sprouts-11-14/index.html)*

Ref. 80. Food and Drug Administration. "Recalls: *Listeria monocytogenes* in Sprouts." FDA Recall Enterprise System.*

Ref. 81. Weller, D., et al. 2016. "Validation of a Previously Developed Geospatial Model that Predicts the Prevalence of *Listeria monocytogenes* in New York State Produce Fields." *Applied and Environmental Microbiology* 82 (3):797-807.

Ref. 82. Weller, D., et al. 2015. "Spatial and temporal factors associated with an increased prevalence of *Listeria monocytogenes* in New York State spinach fields." *Applied and Environmental Microbiology* 81 (17):6059-6069.

Ref. 83. Weller, D., et al. 2015. "Irrigation is Significantly Associated with an Increased Prevalence of *Listeria monocytogenes* in Produce Production Environments in New York State." *Journal of Food Protection* 78 (6):1132-1141.

Ref. 84. National Advisory Committee on Microbiological Criteria for Foods. 1991. "*Listeria monocytogenes*: Recommendations by the National Advisory Committee on Microbiological Criteria for Foods." *International Journal of Food Microbiology* 14:185-246.

Ref. 85. International Commission on Microbiological Specifications for Foods (ICMSF). 1996. "Chapter 8. *Listeria monocytogenes*." *Microorganisms in Foods 5 - Characteristics of Microbial Pathogens*, pp 141-182. London: Blackie Academic & Professional.

Ref. 86. Lado, B.H., et al. 2007. "Characteristics of *Listeria monocytogenes* Important to Food Processors." *Listeria, Listeriosis, and Food Safety, 3rd edition*, edited by E.T. eds Ryser and E.H. Marth, pp 157-213. Boca Raton, FL: CRC Press Taylor & Francis Group.

Ref. 87. El-Kest, S.E., et al. 1992. "Freezing of *Listeria monocytogenes* and Other Microorganisms: A Review." *Journal of Food Protection* 55 (8):639-648.

Ref. 88. Institute for Food Technologists. 2001. "Evaluation and Definition of Potentially Hazardous Foods, IFT/FDA Contract No. 223-98-2333." Accessed October 14, 2021. (https://www.fda.gov/files/food/published/Evaluation-and-Definition-of-Potentially-Hazardous-Foods.pdf)

Ref. 89. Codex Alimentarius Commission. 2007. "Guidelines on the Application of General Principles of Food Hygiene to the Control of *Listeria monocytogenes* in Foods CAC/GL 61." Accessed February 14, 2023. (https://www.fao.org/fao-who-codexalimentarius/sh-proxy/en/?lnk=1&url=https%253A%252F%252Fworkspace.fao.org%252Fsites%252Fcodex%252FStandards%252FCXG%2B61-2007%252FCXG_061e.pdf)

Ref. 90. Carpentier, B., et al. 1993. "Biofilms and Their Consequences, with Particular Reference to Hygiene in the Food Industry." *Journal of Applied Bacteriology* 75:499-511.

Ref. 91. Carpentier, B., et al. 2011. "Review - Persistence of *Listeria monocytogenes* in Food Industry Equipment and Premises." *International Journal of Food Microbiology* 145:1-8.

Ref. 92. United Fresh Produce Association. 2013. "Guidance on Environmental Monitoring and Control of *Listeria* for the Fresh Produce Industry." Accessed October 14, 2021. (http://www.centerforproducesafety.org/amass/documents/document/263/Listeria%20Guidance%20UFPA%202013.pdf)

Ref. 93. Grocery Manufacturers Association. 2014. "*Listeria monocytogenes* Guidance on Environmental Monitoring and Corrective Actions in At-risk Foods." Accessed October 14, 2021. (http://ucfoodsafety.ucdavis.edu/files/208833.pdf)

Ref. 94. Tompkin, R.B. 2002. "Control of *Listeria monocytogenes* in the Food-Processing Environment." *Journal of Food Protection*, 65 (4):709–725.

Ref. 95. Food and Drug Administration. "BAM Chapter 10: Detection of *Listeria monocytogenes* in Foods and Environmental Samples, and Enumeration of *Listeria monocytogenes* in Foods." Accessed October 14, 2021. (https://www.fda.gov/food/laboratory-methods-food/bam-chapter-10-detection-listeria-monocytogenes-foods-and-environmental-samples-and-enumeration)*

Ref. 96. The International Commission on Microbiological Specifications for Foods (ICMSF). 2002. "Chapter 8. Selection of cases and attributes plans." *Microorganisms in Foods 7 - Microbiological Testing in Food Safety Management*. pp. 145-165. New York, NY: Kluwer Academic/Plenum.

Ref. 97. Food and Drug Administration. 2017. "Liquid Chemical Sterilization." Accessed October 14, 2021. (http://www.fda.gov/MedicalDevices/ProductsandMedicalProcedures/GeneralHospitalDevicesandSupplies/ucm208018.htm)*

Ref. 98. World Health Organization. 2004. "Laboratory Biosafety Manual, Third Edition." Geneva: WHO. Accessed October 14, 2021. (http://www.who.int/csr/resources/publications/biosafety/en/Biosafety7.pdf)

Ref. 99. Food and Drug Administration. 2015. "Chapter 4 - Sampling in Investigations Operations Manual 2015." Accessed October 14, 2021. (http://www.fda.gov/downloads/ICECI/Inspections/IOM/UCM123507.pdf)*

Ref. 100. Food and Drug Administration. 2017. "Draft Guidance for Industry: Compliance with and Recommendations for Implementation of the Standards for the Growing, Harvesting, Packing, and Holding of Produce for Human Consumption for Sprout Operations." Access December 9, 2021. (https://www.fda.gov/regulatory-information/search-fda-guidance-documents/draft-guidance-industry-compliance-and-recommendations-implementation-standards-growing-harvesting)*