

October 12, 2016

Ms. Michelle Arsenault National Organic Standards Board USDA-AMS-NOP 1400 Independence Ave., SW Room 2648-S, Mail Stop 0268 Washington, DC 20250-0268

#### Re. HS: Chlorine dioxide gas

These comments to the National Organic Standards Board (NOSB) on its Fall 2016 agenda are submitted on behalf of Beyond Pesticides. Founded in 1981 as a national, grassroots, membership organization that represents community-based organizations and a range of people seeking to bridge the interests of consumers, farmers and farmworkers, Beyond Pesticides advances improved protections from pesticides and alternative pest management strategies that reduce or eliminate a reliance on pesticides. Our membership and network span the 50 states and the world.

The NOSB must heed the comments below regarding the petition process. Because the petitioner has created confusion around the petitioned substance, important information about the hazards of chlorine dioxide gas and its regulation by EPA have been hidden from the HS in its deliberations.

In reviewing this substances, the NOSB must apply the criteria in the Organic Foods Production Act (OFPA), that its use—

(i) would not be harmful to human health or the environment;

(ii) is necessary to the production or handling of the agricultural product because of the unavailability of wholly natural substitute products; and

(iii) is consistent with organic farming and handling.<sup>1</sup>

The regulations also require:<sup>2</sup>

In addition to the criteria set forth in the Act, any synthetic substance used as a processing aid or adjuvant will be evaluated against the following criteria:

(1) The substance cannot be produced from a natural source and there are no organic substitutes;

(2) The substance's manufacture, use, and disposal do not have adverse effects on the environment and are done in a manner compatible with organic handling;

<sup>&</sup>lt;sup>1</sup> OFPA §6517(c)(1)(A). Further details at OFPA §6518(m).

<sup>&</sup>lt;sup>2</sup> §205.600(b).

(3) The nutritional quality of the food is maintained when the substance is used, and the substance, itself, or its breakdown products do not have an adverse effect on human health as defined by applicable Federal regulations;

(4) The substance's primary use is not as a preservative or to recreate or improve flavors, colors, textures, or nutritive value lost during processing, except where the replacement of nutrients is required by law;

(5) The substance is listed as generally recognized as safe (GRAS) by Food and Drug Administration (FDA) when used in accordance with FDA's good manufacturing practices (GMP) and contains no residues of heavy metals or other contaminants in excess of tolerances set by FDA; and
(6) The substance is essential for the handling of organically produced agricultural products.

# The material petitioned should be chlorine dioxide gas, not sodium chlorite.

The HS proposal suggests some degree of confusion, partly resulting from the mistaken subject of the petition. We submitted early comments –apparently not early enough– regarding the petition process. They are attached at the end of these comments in order to keep together all the issues surrounding this substance.

Sodium chlorite, which has been petitioned for use in handling, is an intermediate in the manufacturing process of chlorine dioxide gas. Chlorine dioxide gas is the material that is proposed to be used, and hence the material that should be petitioned.

# The NOSB should not approve more sanitizers –particularly chlorine-based sanitizers—until performing a comprehensive review of sanitizers and the need for sanitizers in organic production.

We addressed this issue at length in our comments on hypochlorous acid in spring 2016. Those comments are attached. We have discussed many alternatives that are available for use by organic producers and handlers. Rather than simply proposing another chlorine-based material, the NOSB subcommittees should commission a TR that (1) determines what disinfectant/sanitizer uses are required by law and by need to meet practical conditions of cleanliness, and (2) comprehensively examines more organically-compatible methods and materials to determine whether chlorine-based materials are actually needed for any uses. In doing so, the TR authors should consult with EPA's Safer Choice Program and investigate materials on the Safer Chemical Ingredients List. If there are uses for which chlorine is necessary, then the NOSB should include them in the National List and limit the use to those particular uses.

# Chlorine dioxide is hazardous to humans and the environment.

# Chlorine dioxide is hazardous to workers and consumers.

The HS proposal states, "CIO2 gas is not persistent and not a known bio-accumulative substance. When used in the intended use in an enclosed environment and allowed to degrade to CIO2 or the chlorite ion, there is no toxicity." This is absolutely incorrect. Toxicity is a property of a material, not of a use.<sup>3</sup> The HS is also incorrect when it says, "As noted in the petition, the primary concern of exposure to the substance is acute toxicity related to inhalation where the substance is a known irritant to eyes and mucal membranes. Severe exposure (beyond amounts available by petitioned product) can result in chemically induced pneumonia and or death."

<sup>&</sup>lt;sup>3</sup> For example, "The degree to which a substance (a toxin or poison) can harm humans or animals."

<sup>&</sup>lt;u>http://www.medicinenet.com/script/main/art.asp?articlekey=34093</u> "The quality of being poisonous, or the degree to which something is poisonous." <u>http://dictionary.cambridge.org/us/dictionary/english/toxicity</u>.

The toxic action of chlorite is primarily in the form of oxidative damage to red blood cells at doses as low as 10 mg/kg of body weight.<sup>4</sup> Chlorine dioxide is a severe respiratory and eye irritant. According to the Occupational Safety and Health Administration (OSHA), inhalation can produce coughing, wheezing, respiratory distress, and congestion in the lungs. Irritating effects in humans were intense at concentration levels of 5 ppm. OSHA has set a limit of 0.1 parts of chlorine dioxide or chlorite per million parts of air (0.1 ppm) in the workplace during an 8-hour shift, 40-hour workweek.<sup>5</sup>

OFPA §6518(I) requires the NOSB to "review available information from the Environmental Protection Agency, the National Institute of Environmental Health Studies, and such other sources as appropriate, concerning the potential for adverse human and environmental effects." Among the sources that should have been considered concerning the toxicity of chlorine dioxide are the *EPA Registration Eligibility Document (RED) for Chlorine Dioxide and Sodium Chlorite*<sup>6</sup> and the *Toxicological Profile for Chlorine Dioxide and Chlorite* produced by the Agency for Toxic Substances and Disease Registry (ATSDR). We will use those sources, as well as Technical Reviews produced for the NOSB.

According to the 2006 Chlorine Technical Report, "[D]uring the 'activation' of chlorine dioxide (i.e., activating dilute aqueous solutions of sodium chlorite with an acid to produce chlorine dioxide), the release of gas to the air or 'off gassing' can be a safety hazard to users."<sup>7</sup> The Agency for Toxic Substances and Disease Registry (ATSDR) identified respiratory effects of inhalation exposure including lymphocytic infiltration of the alveolar spaces, alveolar vascular congestion, hemorrhagic alveoli, epithelial erosions, and inflammatory infiltrations of the bronchi; in addition to slightly decreased body weight gain, decreased erythrocyte levels, and increased leukocyte levels, relative to controls.<sup>8</sup>

Other toxicological impacts mentioned by ATSDR include neurodevelopmental effects, which appear to be of greatest toxicological concern. These effects have been seen in both epidemiological and animal studies. They include delays in neurodevelopment, as evidenced by delayed brain growth, decreased locomotor and exploratory behavior, and altered auditory startle response in animals exposed during critical periods of neurodevelopment. ATSDR considers that adequate animal cancer bioassays for cancer have not been performed and that genotoxicity testing has produced mixed results.<sup>9</sup>

EPA says,

For most of the bystander/post application occupational scenarios, the inhalation risks for the bystander/post application occupational exposures are of concern using the EPA's selected inhalation toxicological endpoint (RfC). The occupational RfC, 0.003 ppm, is below the limit of detection for chlorine dioxide. Based on OSHA's Integrated Management Information System (IMIS) data available for chlorine dioxide, all air concentration measurements, even those that were undetectable, are above the RfC.<sup>10</sup>

 <sup>5</sup> 2006 Chlorine TR. Lines 284-289. See also <u>http://www.osha.gov/SLTC/healthguidelines/chlorinedioxide/recognition.html.</u>
 <sup>6</sup> EPA, 2006. *Registration Eligibility Document (RED) for Chlorine Dioxide and Sodium Chlorite* <u>https://archive.epa.gov/pesticides/reregistration/web/pdf/chlorine\_dioxide\_red.pdf</u>. P. 61.

<sup>7</sup> 2006 Chlorine TR. Lines 240-242.

<sup>&</sup>lt;sup>4</sup> 2006 Chlorine TR. Lines 263-264.

<sup>&</sup>lt;sup>8</sup> ATSDR, 2004. Toxicological Profile for Chlorine Dioxide and Chlorite. P. 11. Available at: <u>http://www.atsdr.cdc.gov/toxprofiles/tp160.pdf</u>.

<sup>&</sup>lt;sup>9</sup> ATSDR, 2004. Toxicological Profile for Chlorine Dioxide and Chlorite. Pp. 15-55. Available at: <u>http://www.atsdr.cdc.gov/toxprofiles/tp160.pdf</u>.

<sup>&</sup>lt;sup>10</sup> EPA, 2006. Registration Eligibility Document for Chlorine Dioxide and Sodium Chlorite. P. 35.

This means that chlorine dioxide levels cannot be determined to be at a safe level by measurement because the safe level is lower than the lowest level that can be measured.

# Chlorine dioxide must be removed from produce.

The petition addendum states, "Gas applications are different than water chemistry described above, and so, unlike traditional water interventions, precautionary potable rinses are not required. (see the directions page for EPA Reg. # 79814-5 and FCN 000949 - attached below)." A few points need to be made about these references. First, EPA Reg. #79814-5 is a <u>conditionally</u> registered product. Conditional registrations are temporarily granted while the registrant collects and supplies missing data. Environmentalists and consumer advocates unanimously oppose the issuance of conditional registrations because they allow use of –and promote dependence on– products that are not fully tested.

Second, the excerpt provided by the petitioner does not make it clear that the product is labeled only for use "in controlling microbiological growth such as late blight, brown rot, and others on <u>potatoes</u> during storage and shipment."<sup>11</sup> Although Section 2(ee) of the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) allows application "against any target pest not specified on the labeling if the application is to the crop, animal, or site specified on the labeling, unless the Administrator has required that the labeling specifically state that the pesticide may be used only for the pests specified on the labeling after the Administrator has determined that the use of the pesticide against other pests would cause an unreasonable adverse effect on the environment," this provision does not apply in this case because the crop (potatoes) and site (potato storage facilities) are not the same as on the label. Even if allowed, the only supportive residue data come from two studies on tomatoes and cantaloupes, not reviewed by EPA, that were supported by the pesticide registrant (which is also the petitioner for this material.) In addition, the residues of chlorate and perchlorate found, while small, nonetheless would not be allowed because there are no tolerances or exemptions from tolerance for chlorine dioxide, chlorite, chlorate, or perchlorate on tomatoes or tomato products or cantaloupes.<sup>12</sup>

Third, FCN 000949 (as provided in the petitioner's addendum) states that it "is not applicable to usage of the FCS in the field, in facilities that only handle raw agricultural commodities, or in transportation from the field to such facilities." It is thus irrelevant to this petition.

In its Registration Eligibility Document (RED), EPA requires that for a chlorine dioxide fruit and vegetable treatment, the following appear on the label: "Fruits and vegetables treated with chlorine dioxide must be blanched, cooked, or canned before consumption or distribution in commerce." This is based on the need to reduce residues below 3 ppm.<sup>13</sup>

# Chlorine dioxide is hazardous to the environment.

The HS repeatedly refers to the degradation of chlorine dioxide to "CIO<sub>2</sub>." However, CIO<sub>2</sub> is chlorine dioxide.

- https://www.gpo.gov/fdsys/pkg/CFR-2007-title40-vol23/pdf/CFR-2007-title40-vol23-part180.pdf.
- <sup>13</sup> EPA, 2006. Registration Eligibility Document (RED) for Chlorine Dioxide and Sodium Chlorite

<sup>&</sup>lt;sup>11</sup> https://www3.epa.gov/pesticides/chem\_search/ppls/079814-00005-20130827.pdf.

<sup>&</sup>lt;sup>12</sup> EPA, Index to Pesticide Chemical Names, Part 180 Tolerance Information, and Food and Feed Commodities (By Commodity) <u>https://www.epa.gov/sites/production/files/2015-01/documents/tolerances-commodity.pdf</u>. 40 CFR Part 180.

https://archive.epa.gov/pesticides/reregistration/web/pdf/chlorine\_dioxide\_red.pdf. P. 61.

When chlorine dioxide gas is released into the environment, it readily dissociates into chlorine gas and oxygen.<sup>14</sup> Toxic Release Inventory (TRI) data includes 5.7 million pounds of chlorine per year released by facilities making and using chlorine,<sup>15</sup> and 1.7 million pounds of chlorine dioxide released by facilities making and using chlorine dioxide.<sup>16</sup> Most of the reported chlorine dioxide released goes into the atmosphere, where degradation products are chlorine gas, oxygen, and chlorine trioxide.<sup>17</sup>

If released into water or soil, chlorine dioxide or sodium chlorite would have effects ranging from slightly to very highly toxic on a wide range of organisms. It is slightly acutely toxic to estuarine/marine fish and ranges from highly acutely toxic to slightly acutely toxic to estuarine/marine invertebrates. It is acutely toxic to freshwater and marine/estuarine fish and invertebrates and aquatic plants. It is moderately toxic to aquatic plants. For terrestrial plants, results of toxicity studies indicate that chlorine dioxide/sodium chlorite are moderately toxic to terrestrial plants. For aquatic plants, toxicity study results.<sup>18</sup> EPA says, "Chronic risks to listed aquatic organisms cannot be assessed at this time; this risk will be assessed when required chronic toxicity data are submitted to and evaluated by the Agency."<sup>19</sup> (The lack of data is a consequence of conditional registration.)

# Chlorine dioxide gas is not necessary.

The petitioner gives this reason for the need for chlorine dioxide gas: "The substance will improve the shelf life and safety of treated articles. It represents a significant new tool to advance current food safety directives and needs." The HS gives this reason: "There are some fluid alternatives such as sodium hypochlorite or chlorine dioxide in liquid form, the latter of which is already listed on the National List. However, presently there are no anti-microbial pesticides, sanitizers or disinfectants in gas form on the National List." Neither of these reasons is a reason that, as required by OFPA, chlorine dioxide gas "is necessary to the production or handling of the agricultural product because of the unavailability of wholly natural substitute products." Ozone is an antimicrobial pesticide, sanitizer, and disinfectant gas listed on the National List, by the way,

In view of the statement by the petitioner that no residue should be expected because of the high reactivity of chlorine dioxide and that produce surfaces are "naturally rich in reducing matter," it is surprising that the HS merely stated without substantiation, "Neither the nutritional quality of the food nor human health is impacted with use of CIO2 gas or its breakdown products of CIO2 or chlorite ions."

In addition, Technical Reviews performed for the NOSB have repeatedly pointed out alternative practices and materials. Water, acetic acid, vinegar, citric acid, and other naturally occurring acids were mentioned in the 2006 Crops Chlorine TR, for example.<sup>20</sup> Materials currently on the National List include hydrogen peroxide, acidified sodium chlorite, and peracetic acid. The NOSB recently voted to add hypochlorous acid. The TR for hydrogen peroxide also mentions alternative practices including UV light and temperature control.<sup>21</sup> Other

<sup>&</sup>lt;sup>14</sup> ATSDR, 2004. Toxicological Profile for Chlorine Dioxide and Chlorite. P. 9. Available at: <u>http://www.atsdr.cdc.gov/toxprofiles/tp160.pdf</u>.

<sup>&</sup>lt;sup>15</sup> ATSDR Tox Profile Chlorine, p. 162.

<sup>&</sup>lt;sup>16</sup> ATSDR, 2004. Toxicological Profile for Chlorine Dioxide and Chlorite. P. 88. Available at: <u>http://www.atsdr.cdc.gov/toxprofiles/tp160.pdf</u>.

<sup>&</sup>lt;sup>17</sup> ATSDR, Tox Profile for Chlorine Dioxide and Chlorite. Pp. 88, 90.

<sup>&</sup>lt;sup>18</sup> EPA, 2006. Registration Eligibility Document for Chlorine Dioxide and Sodium Chlorite. Pp. 38-39.

<sup>&</sup>lt;sup>19</sup> EPA, 2006. Registration Eligibility Document for Chlorine Dioxide and Sodium Chlorite. Pp. 39, 40.

<sup>&</sup>lt;sup>20</sup> 2006 Chlorine TR Crops lines 260-470.

<sup>&</sup>lt;sup>21</sup> 2015 TR for hydrogen peroxide in crops, lines 729-740.

practices leading to better control of spoilage include care in handling produce to avoid injury to fruits and vegetables, avoiding contact with raw manure, and avoidance of sanitizers that kill beneficial microbes.<sup>22</sup>

# Chlorine dioxide gas is not compatible with organic and sustainable production.

The petitioned product has a conditional registration from EPA, meaning that not all essential data have been submitted. It is not labeled for this use. The necessary tolerances or exemptions from tolerances do not exist. It is a hazardous chemical used to take the place of care in handling and less hazardous materials. It does not enhance life and properties, is not made from renewable resources, and has a negative impact on biodiversity.<sup>23</sup>

# Conclusion

We are frankly appalled that the HS would propose adding chlorine dioxide gas to the National List. We assume that the confusion over the petitioned substance may be partially responsible for this regrettable action. However, it is also apparent that the HS did not carry out its due diligence in examining the statements made by the petitioner. The petition should be rejected because it fails to meet all OFPA criteria.

Thank you for your consideration of these comments.

Sincerely,

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Terry Shistar, Ph.D. Board of Directors

Attachments: Comments on petition process, August 1, 2016. Comments on hypochlorous acid petition, April 11, 2016.

<sup>&</sup>lt;sup>22</sup> See comments on hypochlorous acid.

<sup>&</sup>lt;sup>23</sup> 2011 TR lines 270-271, 278-279, 349-352.

August 1, 2016

Ms. Michelle Arsenault National Organic Standards Board USDA-AMS-NOP 1400 Independence Ave. SW., Room 2648-S, Mail Stop 0268 Washington, DC 20250-0268

#### **Re. Petition process**

These comments to the National Organic Standards Board (NOSB) on items of its Fall 2016 agenda are submitted on behalf of Beyond Pesticides. These are preliminary comments that will hopefully inform the subcommittees' deliberations. They should not be taken as our final comments on the issue.

These comments relate most immediately to ammonium glycinate and ammonium citrate, which have been petitioned for use in crop production, and sodium chlorite, which has been petitioned for use in handling. In all of these cases, an intermediate in the manufacturing process is the subject of the petition.

§6504 of the Organic Foods Production Act (OFPA) says,

To be sold or labeled as an organically produced agricultural product under this chapter, an agricultural product shall—

(1) have been produced and handled without the use of synthetic chemicals, except as otherwise provided in this chapter; ...

#### §6517 of OFPA says,

#### (a) In general

The Secretary shall establish a National List of approved and prohibited substances that shall be included in the standards for organic production and handling established under this chapter in order for such products to be sold or labeled as organically produced under this chapter.

#### (b) Content of list

The list established under subsection (a) shall contain an itemization, by specific use or application, of each synthetic substance permitted under subsection (c)(1) or each natural substance prohibited under subsection (c)(2).

And then gives guidelines and procedures for producing the National List.

Substances on the National List are thus those allowed to be <u>used in</u> organic production and handling. The three petitioned substances mentioned above are not meant to be used in organic production and handling. They are meant to be used in <u>producing substances</u> that would be used in organic production and handling.

In laying out factors to be considered by the NOSB, §6518(m) of OFPA includes "the probability of environmental contamination during manufacture, use, misuse or disposal of such substance." Thus, it is required for the NOSB to examine these intermediates as part of their review of the substance that is actually

used in organic production and handling. In some cases, the NOSB may decide to limit the substance used to one produced by a specific manufacturing method or raw material. Thus, at its spring 2016 meeting, the NOSB decided to list "hypochlorous acid generated via electrolyzed water."

Although there are situations in which the line between manufacture and use is unclear –for example, in the case of smoke bombs used to release sulfur dioxide in rodent tunnels—these are not among them. In the case of these materials, the petitioning of an intermediate clouds the application of OFPA criteria, and could possibly lead to unintended consequences.

OFPA criteria apply to the substances <u>as used</u> in organic production and handling. Since the chelated minerals and chlorine dioxide gas are not petitioned, the NOSB does not have a framework for considering the following issues, among others:

- The need for additional forms of chelated minerals, including the efficacy of citrates and glycinates;
- The impacts of the chelated minerals on soil, plants, and consumers;
- The health and environmental impacts of chlorine dioxide;
- EPA requirements of chlorine dioxide use.

In addition, listing these three substances on the National List opens the door to their direct use, which would have very different consequences from those studied by the NOSB.

Therefore, we request that the subcommittees request petitioners to reformulate their petitions to request use of the materials actually proposed to be used. (In the case of chlorine dioxide, this would be the original petition.)

Thank you for your consideration of these comments.

Sincerely,

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Terry Shistar, Ph.D. Board of Directors

# **Comments on Hypochlorous Acid Petition**

April 11, 2016

Ms. Michelle Arsenault National Organic Standards Board USDA-AMS-NOP 1400 Independence Ave. SW., Room 2648-S, Mail Stop 0268 Washington, DC 20250-0268

#### Re. CS, HS, LS: Hypochlorous Acid

These comments to the National Organic Standards Board (NOSB) on its Spring 2016 agenda are submitted on behalf of Beyond Pesticides. Founded in 1981 as a national, grassroots, membership organization that represents community-based organizations and a range of people seeking to bridge the interests of consumers, farmers and farmworkers, Beyond Pesticides advances improved protections from pesticides and alternative pest management strategies that reduce or eliminate a reliance on pesticides. Our membership and network span the 50 states and the world.

Beyond Pesticides opposes the listing of new sanitizers/disinfectants –especially those containing chlorine—until a thorough review of the need for these products in organic production and handling is performed. This review should identify uses that require chlorine and should also look at more environmentally friendly materials, including those on EPA's Safer Chemical Ingredients List.

# The petition

# 205.601, 205.603, and 205.605. The petitioner asks that the current listings for chlorine materials be amended to include hypochlorous acid.

The petitioner claims that because sodium and calcium hypochlorite, which are currently on the National List in all three sections, in the dilute aqueous form in which they are used, exist in solution as hypochlorous acid, hypochlorous acid is essentially allowed now. In issuing Policy Memorandum 15-4 on September 11, 2015, the NOP is apparently accepting the petitioner's argument. Meanwhile, the NOP has asked the NOSB to review the material and the process used to make it. This process does not respect the role of the NOSB as gatekeeper of the National List, and therefore we ask that the NOP rescind NOP PM 15-4 until the NOSB takes action on hypochlorous acid.

Nevertheless, although we are convinced by the chemistry that hypochlorous acid is indeed allowed to be used under the current listings of sodium hypochlorite and calcium hypochlorite, we ask that the NOSB delay recommending the petitioned change until it performs a thorough review of all sanitizers/disinfectants and their uses.

In considering the inclusion of hypochlorous acid, the NOSB should evaluate the need for disposal of the sodium hydroxide that is co-generated with hypochlorous acid.

The comments below argue that the NOSB and NOP should eliminate use of chlorine-based materials and develop guidance for the appropriate use of alternative materials and practices.

# Sanitizers, disinfectants, and so forth

Often we see the NOSB *assuming* a need for strong chemicals as cleaners or disinfectants when none may be needed. We have seen this in our own investigations with personal care products using the biocide triclosan. Research has shown that washing with ordinary soap and water is as effective as using soap containing triclosan. Furthermore, as pointed out by a 2010 report of EPA's Office of Inspector General (OIG), this problem is widespread —the OIG found that approximately 40% of all antimicrobial products have not been tested for efficacy, and one third of all products tested each year fail, without notification of users.<sup>24</sup> We need research into effective means of cleaning food contact surfaces and food containers with organic and natural cleaning methods, such as hot water or steam or materials more compatible with organic processing, including hydrogen peroxide. We need research on organic systems, including growing, harvesting, storing, and transporting crops in ways that avoid the need for rinsing in highly chlorinated water. However, it is very likely that we currently have all the non-chlorine tools we need.

The NOSB and NOP need to clarify whether chlorine is required by other statutes. In our informal conversations, we have been told that other laws require the use of chlorine in higher concentrations than those listed on the National List. If other laws specifically require the use of chlorine, then it must be allowed under the organic program, but if it is, the use should be specifically delineated on the National List.

# Some definitions

The following definitions are quoted from a guidance document produced by the Centers for Disease Control and Prevention for health care facilities.<sup>25</sup>

*Sterilization* describes a process that destroys or eliminates all forms of microbial life and is carried out in health-care facilities by physical or chemical methods.

*Disinfection* describes a process that eliminates many or all pathogenic microorganisms, except bacterial spores, on inanimate objects.

*Cleaning* is the removal of visible soil (e.g., organic and inorganic material) from objects and surfaces and normally is accomplished manually or mechanically using water with

<sup>&</sup>lt;sup>24</sup> U.S. EPA Office of Inspector General, 2010. EPA Needs to Assure Effectiveness of Antimicrobial Pesticide Products, <u>http://www.epa.gov/oig/reports/2011/20101215-11-P-0029.pdf</u>

<sup>&</sup>lt;sup>25</sup> Guideline for Disinfection and Sterilization in Healthcare Facilities, 2008. <u>http://www.cdc.gov/hicpac/pdf/guidelines/Disinfection\_Nov\_2008.pdf</u>

detergents or enzymatic products. Thorough cleaning is essential before high-level disinfection and sterilization because inorganic and organic materials that remain on the surfaces of instruments interfere with the effectiveness of these processes.

*Sanitizer*: agent that reduces the number of bacterial contaminants to safe levels as judged by public health requirements. Commonly used with substances applied to inanimate objects. According to the protocol for the official sanitizer test, a sanitizer is a chemical that kills 99.999% of the specific test bacteria in 30 seconds under the conditions of the test.

The NOP regulations use these terms as if they are synonymous. Since organic practices depend on having a healthy balance of microbes rather than eliminating them, growers, certifiers, NOSB, and NOP all need to be clear about when sanitizing is necessary and when cleaning is sufficient. Removal of all microbial life leaves surfaces available for colonization by spoilage or pathogenic organisms. If strong residual sanitizers are used, strong selection pressure is applied for the development of resistance to materials that may be needed in emergency medical situations.

# Implications of Microbial Ecology for the Use of Sanitizers and Disinfectants

Research on microbial communities calls into question routine use of antimicrobial soaps, as well as sanitizers in food handling. It suggests that we may prevent disease better by preserving natural microbial communities than by exterminating them.

# **Ecological Processes**

Ecological communities are structured by processes that include colonization, succession, competition, and predation. This applies to microbial communities as well as communities of macroorganisms. When a hurricane strikes an island, it may wipe out most of the vegetation, setting in motion processes leading to the re-establishment of plant and animal communities, which may be different from the original communities, depending on the colonizers and the relationships among them. Colonization by pioneer organisms leads to changes in the environment that make it favorable for others, beginning the process of succession to a more stable community.

Similarly, when a microbial community is wiped out by application of an antibiotic, disinfectant, or antimicrobial soap, the habitat is available for colonization by new microorganisms. Just as organic agriculture is based on the maintenance of healthy ecosystems and ecological communities, organic approaches to food safety and personal hygiene should be based on ecological processes. Here we look at implications of microbial ecology on human skin and plant surfaces.

# Microbiota on the Skin

Much of the recent research on microbial ecology has been stimulated by the Human Microbiome Project (HMP),<sup>26</sup> which is designed to bring new methods of studying microorganisms to bear on the properties and functioning of microbial communities – specifically those in habitats in and on humans.<sup>27,28,29</sup> It is well known that a human individual contains approximately ten times as many bacteria as human cells.<sup>30</sup> The extensive sampling by the HMP of the human microbiome across many individuals and habitats on their bodies helps to characterize the normal microbiota of healthy adults in a Western population, resulting in a concept of an individual human as a "supraorganism." In addition, it supports the concept of disease as "dysbiosis," an imbalance of the natural biota.<sup>31</sup>

The skin is the human body's largest organ and performs a diverse and complex variety of innate and adaptive immune functions. <sup>32</sup> It is an inhospitable environment for microbial life, a somewhat acid environment exposed to the effects of drying, friction, washing, and various chemicals. <sup>33</sup>

The most practical issue arising from studies of the human microbiome is the extent to which the microbiome affects our health. The role of the gastrointestinal microbiome in supporting immunity is becoming certain, though details are complicated by its role in processing food. The skin, through its resident microbial communities, plays an active role in immunity beyond its function as a physical barrier. The skin microbiota contributes to immune system function by inhibiting the growth of pathogenic microbes—by means of competition for nutrients and space and by restricting the growth of competitors through the production of antimicrobial compounds, called bacteriocins, which can inhibit the growth of other species of bacteria.<sup>34</sup>

<sup>&</sup>lt;sup>26</sup> The websites <u>http://nihroadmap.nih.gov/hmp/</u> and <u>http://hmpdacc.org/</u>, and links therein, provide additional information about the HMP and access to HMP data.

<sup>&</sup>lt;sup>27</sup> Turnbaugh, P. J., Ley, R. E., Hamady, M., Fraser-Liggett, C., Knight, R., & Gordon, J. I. (2007). The human microbiome project: exploring the microbial part of ourselves in a changing world. *Nature*, *449*(7164), 804.

 <sup>&</sup>lt;sup>28</sup> Peterson, J., Garges, S., Giovanni, M., McInnes, P., Wang, L., Schloss, J. A., ... & NIH HMP Working Group. (2009).
 The NIH human microbiome project. *Genome research*, *19*(12), 2317-2323.

<sup>&</sup>lt;sup>29</sup> Human Microbiome Project Consortium. (2012). Structure, function and diversity of the healthy human microbiome. *Nature*, *486*(7402), 207-214.

<sup>&</sup>lt;sup>30</sup> Scharschmidt, T. C., & Fischbach, M. A. (2013). What lives on our skin: ecology, genomics and therapeutic opportunities of the skin microbiome. *Drug Discovery Today: Disease Mechanisms*, *10*(3), e83-e89.

<sup>&</sup>lt;sup>31</sup> Robles-Alonso, V., & Guarner, F. (2014). From basic to applied research: lessons from the human microbiome projects. *Journal of clinical gastroenterology, 48*, S3-S4.

<sup>&</sup>lt;sup>32</sup> Sanford, J. A., & Gallo, R. L. (2013, November). Functions of the skin microbiota in health and disease. In *Seminars in immunology* (Vol. 25, No. 5, pp. 370-3.

<sup>&</sup>lt;sup>33</sup> Scharschmidt, T. C., & Fischbach, M. A. (2013). What lives on our skin: ecology, genomics and therapeutic opportunities of the skin microbiome. *Drug Discovery Today: Disease Mechanisms*, *10*(3), e83-e89.

<sup>&</sup>lt;sup>34</sup> Sanford, J. A., & Gallo, R. L. (2013, November). Functions of the skin microbiota in health and disease. In *Seminars in immunology* (Vol. 25, No. 5, pp. 370-3.

Among those with damaged skin, certain bacteriocin producers proliferate and dominate the bacterial community (Roth and James, 1988)."<sup>35</sup>

# Microbial Ecology of the Skin

The dermal environment is a complex system of cell layers, nerves, and glands.<sup>36</sup> Harmful disturbances of the skin microbial community may arise from changes in the composition of the community from acquisition of non-resident microorganisms or the removal of dominant microorganisms, handwashing and other behaviors, environmental factors varying with geography and indoor environments, and host genetics and demographic characteristics.<sup>37</sup>

As our ability to respond to pathogens with antimicrobial chemicals is compromised through the development of antibiotic resistance, the importance of maintaining health and responding to infection through encouragement of commensal microbiota is becoming more widely understood.

There are many ways that the skin microbiota can be disturbed, possibly leading to dysbiosis. For example, sealing skin abrasions with a bandage or other airtight barrier may promote growth of potentially pathogenic anaerobes. *Staphylococcus aureus*, once believed to be a "transient colonizer during abnormal conditions," is now known to be a resident bacterium that may become pathogenic upon disturbance of the individual's skin microbiota.<sup>38</sup>

Hands can be thought of as either carriers of transient infectious organisms and/or as vectors that harbor established, endogenous microorganisms with the potential to be transmitted from one person to another. Despite the benefits of hand washing in reducing disease transmission by removing transients, the effects of hand washing on the longer term resident biota are still unknown. Such impacts can be compared to the disturbance caused by hurricanes and forest fires. Hand washing is meant to remove transient microorganisms to decrease self-inoculation when we eat or reduce transmission of our disease to others, but researchers do not necessarily see a reduction in bacteria after hand washing. Disease results from not just an increase in bacteria, but also a change in the microbial community of the individual and the resulting interaction with host immunity.

# Microbiota of the Phyllosphere

The phyllosphere is comprised of the aboveground portions of plants that are available for colonization by microorganisms. In many ways, the phyllosphere is analogous to human skin. The phyllosphere microbial community includes a large and diverse microbiota of bacteria,

 <sup>&</sup>lt;sup>35</sup> Rosenthal, M., Goldberg, D., Aiello, A., Larson, E., & Foxman, B. (2011). Skin microbiota: microbial community structure and its potential association with health and disease. *Infection, Genetics and Evolution*, *11*(5), 839-848.
 <sup>36</sup> Rosenthal, M., Goldberg, D., Aiello, A., Larson, E., & Foxman, B. (2011). Skin microbiota: microbial community structure and its potential association with health and disease. *Infection, Genetics and Evolution*, *11*(5), 839-848.
 <sup>37</sup> Rosenthal, M., Goldberg, D., Aiello, A., Larson, E., & Foxman, B. (2011). Skin microbiota: microbial community structure and its potential association with health and disease. *Infection, Genetics and Evolution*, *11*(5), 839-848.
 <sup>38</sup> Rosenthal, M., Goldberg, D., Aiello, A., Larson, E., & Foxman, B. (2011). Skin microbiota: microbial community structure and its potential association with health and disease. *Infection, Genetics and Evolution*, *11*(5), 839-848.
 <sup>38</sup> Rosenthal, M., Goldberg, D., Aiello, A., Larson, E., & Foxman, B. (2011). Skin microbiota: microbial community structure and its potential association with health and disease. *Infection, Genetics and Evolution*, *11*(5), 839-848.

fungi, yeast, archaea, and other microorganisms that have commensal, pathogenic, and mutualistic interactions with the plant host. While the phyllosphere contains plant pathogens (and human pathogens), it also contains microorganisms that can act as biocontrols for those pathogens. Commensal microbiota on leaves can play a role in pathogen exclusion, contribute to plant health and productivity, and have practical applications in disease prevention.<sup>39</sup>

Bacteria are considered the most numerous of phyllosphere organisms, including those from the phyla Actinobacteria, Bacteroidetes, Firmicutes, and Proteobacteria.<sup>40</sup> Researchers have suggested that like humans, plants can be considered as supra organisms harboring diverse microbial communities providing specific functions. The combined rhizosphere and phyllosphere microbial communities improve nutrient acquisition and growth, sustain plant growth under stress, induce resistance against pathogens, interact with plant or human pathogens, and interact with herbivores and parasites. Thus, like the skin microbiota, the plant microbiota is a key element in health. There is an interplay between microbiota and plants – while the microbiota affects plants, plants also affect microbiota selection through leaf or root anatomy and morphology or production of exudates.<sup>41</sup>

Phyllosphere, rhizosphere, and soil microbial communities are significantly different in terms of species composition, abundance, and diversity. The main determinants of the rhizosphere microbiome are soil type and plant genotype, while the phyllosphere microbiome is principally influenced by plant species and genotype. Key factors are the chemical and structural composition of the cuticle. The genotype is also particularly important –a single mutation in a plant gene can modify the microbiome. In addition, environmental factors, including UV exposure, air humidity, and geographical location, also influence microbiome composition. Geographical location has been identified as important in a lettuce field, but not in trees. Cropping system, growing season, nitrogen fertilization, and pesticide application also affect community composition. <sup>42, 43</sup>

The human pathogens *Salmonella* and *Escherichia coli* have been found on fresh vegetables, which increases interest in understanding their interactions with the other inhabitants of the phyllosphere. Phyllosphere microorganisms may also provide an ecosystem service to human health. It has been proposed that exposure to environmental microbiota in the air, soil and on plants, is essential for regulating the human immune system. Most epiphytic bacteria are

<sup>&</sup>lt;sup>39</sup> Rastogi, G., Coaker, G. L., & Leveau, J. H. (2013). New insights into the structure and function of phyllosphere microbiota through high-throughput molecular approaches. *FEMS microbiology letters*, *348*(1), 1-10.

<sup>&</sup>lt;sup>40</sup> Rastogi, G., Coaker, G. L., & Leveau, J. H. (2013). New insights into the structure and function of phyllosphere microbiota through high-throughput molecular approaches. *FEMS microbiology letters*, *348*(1), 1-10.

<sup>&</sup>lt;sup>41</sup> Massart, S., Martinez-Medina, M., & Jijakli, M. H. (2015). Biological control in the microbiome era: Challenges and opportunities. *Biological Control, 89*, 98-108.

<sup>&</sup>lt;sup>42</sup> Massart, S., Martinez-Medina, M., & Jijakli, M. H. (2015). Biological control in the microbiome era: Challenges and opportunities. *Biological Control*, *89*, 98-108.

<sup>&</sup>lt;sup>43</sup> Karlsson, I. (2015). Diversity of wheat phyllosphere fungi in different agricultural production systems. Doctoral Thesis Swedish University of Agricultural Sciences Uppsala 2015.

commensal. Some provide specific ecosystem services including phytoremediation of toxic pollutants and cycling of important elements. Others contribute to pathogen exclusion. <sup>44,45</sup>

#### In the field

Like the skin, the phyllosphere is considered a hostile environment for survival and colonization by microorganisms because of fluctuations in solar radiation, temperature, humidity, and heterogeneous availability of nutrients.<sup>46</sup>

Phyllosphere microbes often have a direct positive influence in altering plant surface properties, where they may be involved in nitrogen fixation, promoting the growth of plants, the control of plant pathogens, and the degradation of organic pollutants. However, some phyllosphere microbes have negative effects upon the host –as when plant pathogens result in disease. Phyllosphere microbes may also include human pathogens, thus compromising the safety of plant food grown for consumption.<sup>47</sup>

The phyllosphere community is dynamic. Leaves of both annual and perennial deciduous plants are colonized by microorganisms each year. Successional patterns throughout the growing season generally begin with initial colonization by bacteria, followed by yeasts, then filamentous fungi. <sup>48</sup> Although the contributions of different sources are not well understood, microbial colonizers of the phyllosphere can originate from different sources including soil, air, seed, and other plants.<sup>49</sup>

Plant leaf surfaces are colonized in large part through immigration of bacteria, fungi, and other microorganisms from air, soil, water, seed, or through animal-borne sources. Only a small fraction of the phyllosphere microbiota is shared with the soil.<sup>50</sup> In addition to microbes on the plant surface, it is likely that every plant species is colonized by at least one endophytic bacterial species. Although most endophytes appear to be non-pathogenic to humans, a number of pathogenic bacteria can become internalized as at least temporary endophytes

<sup>&</sup>lt;sup>44</sup> Karlsson, I. (2015). Diversity of wheat phyllosphere fungi in different agricultural production systems. Doctoral Thesis Swedish University of Agricultural Sciences Uppsala 2015.

<sup>&</sup>lt;sup>45</sup> Rastogi, G., Coaker, G. L., & Leveau, J. H. (2013). New insights into the structure and function of phyllosphere microbiota through high-throughput molecular approaches. *FEMS microbiology letters*, *348*(1), 1-10.

<sup>&</sup>lt;sup>46</sup> Rastogi, G., Coaker, G. L., & Leveau, J. H. (2013). New insights into the structure and function of phyllosphere microbiota through high-throughput molecular approaches. *FEMS microbiology letters*, *348*(1), 1-10.

<sup>&</sup>lt;sup>47</sup> Zhang, B., Bai, Z., Hoefel, D., Tang, L., Wang, X., Li, B., ... & Zhuang, G. (2009). The impacts of cypermethrin pesticide application on the non-target microbial community of the pepper plant phyllosphere. *Science of the Total Environment*, *407*(6), 1915-1922.

<sup>&</sup>lt;sup>48</sup> Karlsson, I. (2015). Diversity of wheat phyllosphere fungi in different agricultural production systems. Doctoral Thesis Swedish University of Agricultural Sciences Uppsala 2015.

<sup>&</sup>lt;sup>49</sup> Karlsson, I. (2015). Diversity of wheat phyllosphere fungi in different agricultural production systems. Doctoral Thesis Swedish University of Agricultural Sciences Uppsala 2015.

<sup>&</sup>lt;sup>50</sup> Rastogi, G., Coaker, G. L., & Leveau, J. H. (2013). New insights into the structure and function of phyllosphere microbiota through high-throughput molecular approaches. *FEMS microbiology letters*, *348*(1), 1-10.

within leaves, and no amount of washing or vegetable preparation will remove them, which may be a problem for the consumption of raw vegetables.<sup>51</sup>

As on the skin, the structure of the phyllosphere microbial community affects the survival and impacts of both plant and human pathogens. For example, *Enterobacter asburiae* reduced the survival of *E. coli* O157:H7 on lettuce leaves by a factor of 20–30, while *Wausteria paucula* increased survival by a factor of 6; bacterial isolates belonging to Firmicutes and Enterobacteriaceae reduced the growth rate of *E. coli* O157:H7 on detached spinach leaves; Salmonella introduced onto tomatoes pre- or postharvest altered the composition of the microbial community; *Enterobacter* and *Bacillus* species reduced the persistence of *Salmonella* on preharvest tomatoes; native plant-associated microorganisms acted as competitors to *Salmonella* and *E. coli* O157:H7 on lettuce and alfalfa sprouts.<sup>52, 53, 54</sup>

#### Organic vs. chemical intensive production

Microbial populations on foliage in agricultural settings are influenced by management practices such as organic vs. chemical-intensive farming, use of antibiotics, pesticide application, and nitrogen fertilization.<sup>55, 56</sup> Otteson et al. concluded, "The fact that organic and conventional phyllosphere bacterial communities were significantly different at numerous time points suggests that crop management methods may influence the bacterial consortia associated with the surfaces of fruits and vegetables."<sup>57</sup>

In spite of the differences in microbial communities between the phyllosphere on plants grown organically vs. those grown in a chemical-intensive system, and in spite of the microbially-active inputs into organic production (e.g., compost and manure), there is evidence that the phyllosphere on organic plants does not harbor more plant or human pathogens. Leff and Fierer found that vegetables labeled as conventional had a greater relative abundance of potentially pathogenic Enterobacteriaceae taxa across several produce types, including spinach, lettuce, tomatoes, and peaches, than those labeled organic.<sup>58</sup> Otteson et al. found no

<sup>&</sup>lt;sup>51</sup> Jackson, C. R., Stone, B. W., & Tyler, H. L. (2015). Emerging perspectives on the natural microbiome of fresh produce vegetables. *Agriculture*, *5*(2), 170-187.

<sup>&</sup>lt;sup>52</sup> Shi, X., Wu, Z., Namvar, A., Kostrzynska, M., Dunfield, K., & Warriner, K. (2009). Microbial population profiles of the microflora associated with pre-and postharvest tomatoes contaminated with Salmonella typhimurium or Salmonella montevideo. *Journal of applied microbiology*, *107*(1), 329-338.

<sup>&</sup>lt;sup>53</sup> Rastogi, G., Coaker, G. L., & Leveau, J. H. (2013). New insights into the structure and function of phyllosphere microbiota through high-throughput molecular approaches. *FEMS microbiology letters*, *348*(1), 1-10.

<sup>&</sup>lt;sup>54</sup> Jackson, C. R., Stone, B. W., & Tyler, H. L. (2015). Emerging perspectives on the natural microbiome of fresh produce vegetables. *Agriculture*, *5*(2), 170-187.

<sup>&</sup>lt;sup>55</sup> Rastogi, G., Coaker, G. L., & Leveau, J. H. (2013). New insights into the structure and function of phyllosphere microbiota through high-throughput molecular approaches. *FEMS microbiology letters*, *348*(1), 1-10.

<sup>&</sup>lt;sup>56</sup> Leff, J. W., & Fierer, N. (2013). Bacterial communities associated with the surfaces of fresh fruits and vegetables. *PLoS One*, *8*(3), e59310.

<sup>&</sup>lt;sup>57</sup> Ottesen, A. R., White, J. R., Skaltsas, D. N., Newell, M. J., & Walsh, C. S. (2009). Impact of organic and conventional management on the phyllosphere microbial ecology of an apple crop. *Journal of Food Protection*<sup>®</sup>, *72*(11), 2321-2325.

<sup>&</sup>lt;sup>58</sup> Leff, J. W., & Fierer, N. (2013). Bacterial communities associated with the surfaces of fresh fruits and vegetables. *PLoS One*, *8*(3), e59310.

detectable differences in the presence of potential enteric pathogens between organic and chemical-intensive apples, and neither *Salmonella* nor *Escherichia* were found.<sup>59</sup> Marine et al. found an association between *Salmonella* on leafy greens sampled in the field and growing season but not farming system.<sup>60</sup> Distinct fungal communities and a higher proportion of antagonistic fungal isolates against *Botrytis cinerea* were found on organically grown grapes than on those grown in a chemical-intensive system.<sup>61</sup> Several European studies have shown that *Fusarium* and mycotoxin contamination is lower in organic cereal production than in chemical-intensive production.<sup>62</sup> Xu found that more *Salmonella* introduced on tomato leaves survived on plants grown in a chemical-intensive system than in an organic system. She also reported, "Endophytic bacterial diversities of tomato plants grown in conventional soils were significantly lower than those in organic soils. All contaminated fruit (1%) were from tomato plants grown in conventional soil."<sup>63</sup>

#### Mulches

Xu found that different mulches had different effects on the microbial levels. Straw mulch reduced levels of center rot on sweet onion, while black plastic mulches had the opposite effect. Plastic mulch resulted in more coliforms, yeast and mold, as well as mesophilic, psychrotrophic and lactic acid bacteria before storage."<sup>64</sup>

#### Post-harvest

The true phyllosphere microbiome associated with a plant is the microbial community present on or in plants growing in the field. However, from the viewpoint of consumer safety, the microbial populations present at the point of sale or consumption are more relevant. Both epiphytic and endophytic phyllosphere microorganisms may differ at these different time points. <sup>65</sup> Consumers may be exposed to 50 or more species of bacteria while consuming raw vegetables. While many of these bacteria are likely to be plant symbionts or pathogens, some are human pathogens.<sup>66</sup> The pathogens of greatest public health concern are *Shigella* spp., *Salmonella*, enterotoxigenic and enterohemorrhagic *Escherichia coli*, *Campylobacter* spp., *Listeria monocytogenes*, *Yersinia enterocolitica*, *Bacillus cereus*, *Clostridium botulinum*, viruses,

<sup>&</sup>lt;sup>59</sup> Ottesen, A. R., White, J. R., Skaltsas, D. N., Newell, M. J., & Walsh, C. S. (2009). Impact of organic and conventional management on the phyllosphere microbial ecology of an apple crop. *Journal of Food Protection*<sup>®</sup>, *72*(11), 2321-2325.

<sup>&</sup>lt;sup>60</sup> Marine, S. C., Pagadala, S., Wang, F., Pahl, D. M., Melendez, M. V., Kline, W. L., ... & Micallef, S. A. (2015). The Growing Season, but Not the Farming System, Is a Food Safety Risk Determinant for Leafy Greens in the Mid-Atlantic Region of the United States. *Applied and environmental microbiology*, *81*(7), 2395-2407.

<sup>&</sup>lt;sup>61</sup> Karlsson, I. (2015). Diversity of wheat phyllosphere fungi in different agricultural production systems. Doctoral Thesis Swedish University of Agricultural Sciences Uppsala 2015.

<sup>&</sup>lt;sup>62</sup> Karlsson, I. (2015). Diversity of wheat phyllosphere fungi in different agricultural production systems. Doctoral Thesis Swedish University of Agricultural Sciences Uppsala 2015.

 <sup>&</sup>lt;sup>63</sup> Xu, A. (2014). Microbiological assessment of organic produce pre-and post-harvest on Maryland farms and impact of growing and handling methods on epiphytic bacteria. MS thesis, University of Maryland, College Park.
 <sup>64</sup> Xu, A. (2014). Microbiological assessment of organic produce pre-and post-harvest on Maryland farms and impact of growing and handling methods on epiphytic bacteria. MS thesis, University of Maryland, College Park.
 <sup>65</sup> Jackson, C. R., Stone, B. W., & Tyler, H. L. (2015). Emerging perspectives on the natural microbiome of fresh produce vegetables. *Agriculture*, *5*(2), 170-187.

<sup>&</sup>lt;sup>66</sup> Jackson, C. R., Stone, B. W., & Tyler, H. L. (2015). Emerging perspectives on the natural microbiome of fresh produce vegetables. *Agriculture*, *5*(2), 170-187.

and parasites such as *Giardia lamblia*, *Cyclospora cayetanensis*, and *Cryptosporidium parvum*. Fruits and vegetables can become contaminated with pathogenic microorganisms while growing in fields, orchards, vineyards, or greenhouses, or during harvesting, post-harvest handling, processing, distribution, and preparation in food service or home settings."<sup>67</sup> From a food safety standpoint, it makes more sense to sample at point of sale, rather than in the field.<sup>68</sup>

Research looking at the microbiota in the field and post-harvest has found that the post-harvest phyllosphere microbial community shifts in the relative abundance of different species, becoming less diverse and containing species that do well under storage conditions. <sup>69, 70</sup> Although relatively few of the microbial species found after storage are members of the field phyllosphere, the pre-existing community does affect the success of newly-introduced microbes. <sup>71, 72, 73</sup> More potentially pathogenic groups of microbes are found in the field in tomatoes, but the opposite is true of leafy greens and peppers. <sup>74</sup>

Post-harvest handling operations can cause disturbances in the microbiota and select for microbes that survive under storage conditions. The process of harvesting tomatoes alone seems to be enough to shift the community composition (reducing the number of *E. coli* positive samples). Washed post-harvest produce had higher risks than unwashed and pre-harvest organic produce, as measured by indicator organisms. Although adding a sanitizer to rinse water resulted in produce with no significant difference from pre-harvest samples, it did not decrease indicator microbes. Allende et al. showed that while washing reduces microbial loads initially, the difference is no longer significant after 5 days of storage.<sup>75</sup>

<sup>&</sup>lt;sup>67</sup> Beuchat, L. R. (2002). Ecological factors influencing survival and growth of human pathogens on raw fruits and vegetables. *Microbes and infection*, *4*(4), 413-423.

<sup>&</sup>lt;sup>68</sup> Jackson, C. R., Stone, B. W., & Tyler, H. L. (2015). Emerging perspectives on the natural microbiome of fresh produce vegetables. *Agriculture*, *5*(2), 170-187.

<sup>&</sup>lt;sup>69</sup> Jackson, C. R., Stone, B. W., & Tyler, H. L. (2015). Emerging perspectives on the natural microbiome of fresh produce vegetables. *Agriculture*, *5*(2), 170-187.

<sup>&</sup>lt;sup>70</sup> Leff, J. W., & Fierer, N. (2013). Bacterial communities associated with the surfaces of fresh fruits and vegetables. *PLoS One*, *8*(3), e59310.

<sup>&</sup>lt;sup>71</sup> Shi, X., Wu, Z., Namvar, A., Kostrzynska, M., Dunfield, K., & Warriner, K. (2009). Microbial population profiles of the microflora associated with pre-and postharvest tomatoes contaminated with Salmonella typhimurium or Salmonella montevideo. *Journal of applied microbiology*, *107*(1), 329-338.

<sup>&</sup>lt;sup>72</sup> Jackson, C. R., Stone, B. W., & Tyler, H. L. (2015). Emerging perspectives on the natural microbiome of fresh produce vegetables. *Agriculture*, *5*(2), 170-187.

<sup>&</sup>lt;sup>73</sup> Leff, J. W., & Fierer, N. (2013). Bacterial communities associated with the surfaces of fresh fruits and vegetables. *PLoS One*, *8*(3), e59310.

 <sup>&</sup>lt;sup>74</sup> Xu, A. (2014). Microbiological assessment of organic produce pre-and post-harvest on Maryland farms and impact of growing and handling methods on epiphytic bacteria. MS thesis, University of Maryland, College Park.
 <sup>75</sup> Xu, A. (2014). Microbiological assessment of organic produce pre-and post-harvest on Maryland farms and impact of growing and handling methods on epiphytic bacteria. MS thesis, University of Maryland, College Park.

Storage temperature affects the microbial community, selecting for cold tolerant species. <sup>76, 77</sup> For example, both Enterobacteriaceae and Pseudomonadaceae increased at least 1000-fold over 12 days in fresh-cut spinach stored at 10 °C. Refrigerated storage also reduced the diversity and richness of the phyllosphere community, and the temperature of storage influenced the extent of community changes in storage, with larger changes at colder temperatures. Microbiota in bagged lettuce mixes also changed in storage at 10 °C, experiencing an increase in the relative abundance of Enterobacteriaceae and a decrease in the relative abundance of Pseudomonas, but when the bagged lettuce mixes were stored at refrigerator temperature (4 °C), the decrease in *Pseudomonas* was less pronounced. Thus, Jackson et al concluded, "[R]efrigerated storage might help retain the natural microbiome, while extended storage at cool, but not cold, temperatures might be more likely to promote shifts in the phyllosphere community, and potentially favor pathogenic strains."<sup>78</sup>

Another handling measure that affects the microbial community on post-harvest produce is enclosure in air-tight packages. Commercially pre-bagged, refrigerated lettuce samples showed evidence of the presence of additional bacterial populations, including *Pseudomonas libaniensis*, a species first found in Lebanese spring water.<sup>79</sup> Herbs packaged in plastic containers sealed with polymer contained a high proportion of anaerobic microbes.<sup>80</sup>

#### Implications

Researchers are just beginning to grasp the diversity and complexity of epiphytic and endophytic communities of microbes in the phyllosphere. As we have seen, disturbing these communities –e.g., by washing produce—can result in greater exposure to human pathogens. In addition to the stabilizing effects of the natural microbial community, augmenting phyllosphere microbiota can result in reduction of human pathogens and biocontrol of plant pathogens.

#### Biocontrol

Natural members of the plant phyllosphere can reduce the growth of human pathogens. For example, *Pseudomonas syringae* reduced the growth of *E. coli* O157:H7 from wounded apples by a factor of 10-1000. *Pseudomonas fluorescens* 2-79, inhibited *S. enterica* and reduced the growth of *Salmonella* on alfalfa sprouts by a factor of approximately 100,000. *Enterobacter* 

<sup>79</sup> http://www.tgw1916.net/Pseudomonas/libanensis.html

<sup>&</sup>lt;sup>76</sup> Leff, J. W., & Fierer, N. (2013). Bacterial communities associated with the surfaces of fresh fruits and vegetables. *PLoS One*, *8*(3), e59310.

<sup>&</sup>lt;sup>77</sup> Jackson, C. R., Stone, B. W., & Tyler, H. L. (2015). Emerging perspectives on the natural microbiome of fresh produce vegetables. *Agriculture*, *5*(2), 170-187.

<sup>&</sup>lt;sup>78</sup> Jackson, C. R., Stone, B. W., & Tyler, H. L. (2015). Emerging perspectives on the natural microbiome of fresh produce vegetables. *Agriculture*, *5*(2), 170-187.

<sup>&</sup>lt;sup>80</sup> Jackson, C. R., Stone, B. W., & Tyler, H. L. (2015). Emerging perspectives on the natural microbiome of fresh produce vegetables. *Agriculture*, *5*(2), 170-187.

*asburiae* reduced *E. coli* O157:H7 and *Salmonella* on lettuce."<sup>81</sup> However, plant pathogens, along with physical damage, can provide entry and increased risk from human pathogens.<sup>82</sup>

Biocontrol of plant pathogens is an exciting application of the knowledge of plant microbiota that has been practiced by organic growers for many years. The mechanisms involved include resource competition, antibiosis, parasitism, and induced resistance. It can be practiced either by adding antagonistic microorganisms to the phyllosphere or by stimulating naturally occurring antagonists.<sup>83</sup>

Martin reviewed the evidence showing that according to the preponderance of the research, various types of compost tea and/or compost-based liquid preparations can suppress phytopathogens and plant diseases.<sup>84</sup> He cited works by Stindt, and Samerski and Weltzien suggesting that the theoretical basis for effectiveness of compost tea in controlling aerial plant disease is its ability to alter the microbiota of the phyllosphere and to induce resistance in plant hosts. Martin reported that Evans et al. found that multiple applications of aerated compost tea made from various animal manure and green waste composts were consistently as effective as standard fungicide spray programs for managing grapevine powdery and Botrytis bunch rot. Compost with a high diversity of microbes is generally considered best for the production of compost tea to suppress plant disease, with growing support for teas produced from vermicompost or vermicasting.<sup>85</sup>

# Conclusions from Examining Microbial Ecology

Research on microbial communities suggests that we may prevent disease better by preserving or augmenting natural microbial communities. An ecological approach to microbiota in humans and plants calls into question the routine use of antimicrobial soaps, as well as sanitizers in food handling, to attempt to exterminate microbes.

# Chlorine-based disinfectants

Chlorine is a strong oxidizer and hence does not occur naturally in its pure (gaseous) form. Nearly all naturally occurring chlorine occurs as chloride, the ionic form found in salts such as sodium chloride. Gaseous chlorine is formed by running an electric current through salt brine.<sup>86</sup>

The high oxidizing potential of chlorine leads to its use for bleaching, biocides, and as a chemical reagent in manufacturing processes. Because of its reactivity, chlorine and many of its compounds bind with organic matter. In the case of bleaches, the reaction with chlorine

 <sup>&</sup>lt;sup>81</sup> Xu, A. (2014). Microbiological assessment of organic produce pre-and post-harvest on Maryland farms and impact of growing and handling methods on epiphytic bacteria. MS thesis, University of Maryland, College Park.
 <sup>82</sup> Xu, A. (2014). Microbiological assessment of organic produce pre-and post-harvest on Maryland farms and impact of growing and handling methods on epiphytic bacteria. MS thesis, University of Maryland, College Park.
 <sup>83</sup> Karlsson, I. (2015). Diversity of wheat phyllosphere fungi in different agricultural production systems. Doctoral Thesis Swedish University of Agricultural Sciences Uppsala 2015.

<sup>&</sup>lt;sup>84</sup> Martin, C. C. S. Potential of compost tea for suppressing plant diseases. CAB Reviews 2014: 9, No. 032 <u>http://www.cabi.org/cabreviews</u>

<sup>&</sup>lt;sup>85</sup> Martin, C. C. S. Potential of compost tea for suppressing plant diseases. CAB Reviews 2014: 9, No. 032 <u>http://www.cabi.org/cabreviews</u>

<sup>&</sup>lt;sup>86</sup> <u>http://en.wikipedia.org/wiki/Chlorine</u>

destroys chemicals responsible for color. When used as a disinfectant, chlorine reacts with microorganisms and other organic matter. Similarly, the toxicity of chlorine to other organisms comes from its power to oxidize cells.<sup>87</sup>

# Alternatives to chlorine disinfection

To the extent that organic production requires a disinfectant other than the level of residual in finished drinking water, the NOSB should be looking at non-chlorine alternatives. The above-cited 2003 NOSB recommendation stated:

The TAP reviews pointed out many ways in which chlorine is unsatisfactory for organic handling. Chlorine compounds and other halogens have been shown to produce trihalomethanes. It was the NOSB's opinion that while chlorine needs to be allowed in the handling of organic food out of concern for public health and safety, its use needs to be minimized and operators need incentives and clear guidance to develop viable alternatives that protect the public as effectively as chlorine, but are less harmful to food handlers and the environment.

Toward that end, the NOSB has recommended other methods for disinfecting water in crop contact, including ozone, hydrogen peroxide, and periacetic acid. The review of chlorine should be prioritized in the re-review process in light of new information about alternatives, food safety, health effects, and application procedures. To the extent possible, the NOSB encourages the adoption of non-chemical and less toxic methods of disinfection of wash and chill water. This should be done with the full support and cooperation of the Food and Drug Administration's Center for Food Safety and Nutrition, and the Food Safety Inspection Service.

EPA's Safer Choice (formerly Design for the Environment) program has been investigating alternative disinfectants.<sup>88</sup> A Safer Choice label on a disinfectant means that the product meets the following criteria:

- It is in the least-hazardous classes (i.e. III and IV) of EPA's acute toxicity category hierarchy;
- It is unlikely to have carcinogenic or endocrine disruptor properties;
- It is unlikely to cause developmental, reproductive, mutagenic, or neurotoxicity issues;
- It has no outstanding "conditional registration" data issues;
- EPA has reviewed and accepted mixtures, including inert ingredients;
- It does not require the use of Agency-mandated personal protective equipment;
- It has no unresolved or unreasonable adverse effects reported;
- It has no unresolved efficacy failures (associated with the Antimicrobial Testing Program or otherwise);
- It has no unresolved compliance or enforcement actions associated with it; and

<sup>&</sup>lt;sup>87</sup> <u>http://en.wikipedia.org/wiki/Chlorine</u>

<sup>&</sup>lt;sup>88</sup> http://www.epa.gov/pesticides/regulating/labels/design-dfe-pilot.html

 It has the identical formulation as the one identified in the Safer Choice application reviewed by EPA.<sup>89</sup>

EPA has approved the following for use in Safer Choice disinfectant products: citric acid, hydrogen peroxide, I-lactic acid, ethanol, and isopropanol.<sup>90</sup> Safer Choice disinfectant product formulations and "inert" ingredients must also meet the Safer Choice standard for safer cleaning products.<sup>91</sup> All of the approved synthetic Safer Choice disinfectant active ingredients are on the National List. Citric and lactic acids are considered nonsynthetic, are listed on §205.605(a), and do not need to be listed in order to be used in crop or livestock production. In addition, the need for equipment to be clean must be distinguished from a need for disinfection is difficult to accomplish if a surface is not clean.<sup>92</sup>

EPA's Safer Choice has approved **I-lactic acid and citric acid** as meeting its criteria for use as disinfectants.<sup>93</sup> While the Safer Choice criteria are not the same as OFPA criteria, they do require that the materials be low-hazard and efficacious. Lactic acid and citric acid are both considered nonsynthetic and are listed on §205.605(a) with no restrictions as to use.

**Essential oils** are often cited as a class of natural disinfectants. The TR for hydrogen peroxide refers to the following essential oils and extracts: clove oil, melaleuca (tea tree) oil, and oregano oil, pine oil, basil oil, cinnamon oil, eucalyptus oil, helichrysum oil, lemon and lime oils, peppermint oil, tea tree oil, and thyme oil. Aloe vera contains six antiseptic agents active against fungi, bacteria and viruses. There is considerable research on essential oils as disinfectants that could be useful to organic producers. For example, an early review by Janssen et al described methods for screening.<sup>94</sup> A more recent review by Kalemba and Kunicka gave an updated review of screening methods and an overview of the susceptibility of human and foodborne bacteria and fungi towards different essential oils and their constituents.<sup>95</sup> Deans and Ritchie compared the potency of 50 different essential oils and the range of their antibacterial action against 25 genera of bacteria.<sup>96</sup> A review of the literature should be encouraged by the NOSB to encourage the use of safer materials more compatible with organic principles.

# Practices that eliminate the need for disinfectants

Technical reviews have mentioned practices that eliminate the need for disinfectant materials. They include: hot water, steam, UV radiation, slow filtration for cleaning water. As pointed out

<sup>&</sup>lt;sup>89</sup> http://www.epa.gov/pesticides/regulating/labels/design-dfe-pilot.html

<sup>&</sup>lt;sup>90</sup> http://www.epa.gov/pesticides/regulating/labels/design-dfe-pilot.html

<sup>&</sup>lt;sup>91</sup> <u>http://www.epa.gov/dfe/pubs/projects/formulat/dfe\_criteria\_for\_cleaning\_products\_10\_09.pdf</u>

<sup>&</sup>lt;sup>92</sup> Guideline for Disinfection and Sterilization in Healthcare Facilities, 2008. <u>http://www.cdc.gov/hicpac/pdf/guidelines/Disinfection\_Nov\_2008.pdf</u>

<sup>&</sup>lt;sup>93</sup> http://www.epa.gov/pesticides/regulating/labels/design-dfe-pilot.html

<sup>&</sup>lt;sup>94</sup> Janssen, A. M., Scheffer, J. J. C., & Svendsen, A. B. (1987). Antimicrobial activities of essential oils. *Pharmaceutisch Weekblad*, *9*(4), 193-197.

<sup>&</sup>lt;sup>95</sup> Kalemba, D., & Kunicka, A. (2003). Antibacterial and antifungal properties of essential oils. *Current medicinal chemistry*, *10*(10), 813-829.

<sup>&</sup>lt;sup>96</sup> Deans, S. G., & Ritchie, G. (1987). Antibacterial properties of plant essential oils. *International journal of food microbiology*, *5*(2), 165-180.

at the beginning of these comments, "cleaning" is not synonymous with disinfection, and it is possible that in some cases, disinfection is not necessary at all. And, as indicated above, disinfection is sometimes unhealthy.

#### Conclusion: Other sanitizers and disinfectants

While the uses of disinfectants vary so that no one method or material is likely to be effective in all cases, there are numerous alternative methods and materials that should allow organic producers and handlers to avoid the use of the most toxic materials –in particular, those containing chlorine. Regarding alternative materials for teat dips, the iodine TR says, "The available information suggests that commercial antimicrobial products containing oxidizing chemicals (e.g., sodium chlorite, hypochlorite, iodophor), natural products composed of organic acids (e.g., lactic acid), and homemade products using vinegar (i.e., acetic acid) as the active ingredient may all be equally effective teat dip treatments." The active ingredients identified by the Safer Choice are safer and effective alternatives.

#### Conclusion

We have discussed many alternatives that are available for use by organic producers and handlers. Rather than simply proposing another chlorine-based material, the NOSB subcommittees should commission a TR that (1) determines what disinfectant/sanitizer uses are required by law or are otherwise necessary, and (2) comprehensively examines more organically-compatible methods and materials to determine whether chlorine-based materials are actually needed for any uses. In doing so, the TR authors should consult with EPA's Safer Choice Program and investigate materials on the Safer Chemical Ingredients List. If there are uses for which chlorine is necessary, then the NOSB should include them in the National List and limit the use to those particular uses. In addition, in considering the inclusion of hypochlorous acid, the NOSB should evaluate the need for proper disposal of the sodium hydroxide and hydrogen gas that is co-generated with hypochlorous acid.

Thank you for your consideration of these comments.

Sincerely,

Jeresalha Hit

Terry Shistar, Ph.D. Board of Directors