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(54) Title: RHEOLOGY MODIFIED LOW FOAMING LIQUID ANTIMICROBIAL COMPOSITIONS AND METHODS OF USE THEREOF

(57) Abstract: The present disclosure relates to rheology modified, low foaming aqueous antimicrobial compositions. The compositions are phase stable under acidic conditions, and do not need to be rinsed from the surface to which they are applied. The present disclosure further relates to methods of use thereof.

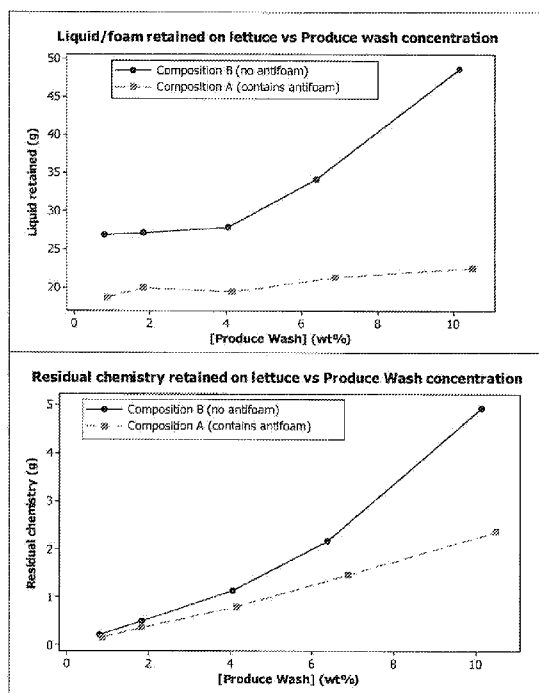


Figure 1



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## **RHEOLOGY MODIFIED LOW FOAMING LIQUID ANTIMICROBIAL COMPOSITIONS AND METHODS OF USE THEREOF**

### **FIELD**

5           The present disclosure relates to aqueous low foaming antimicrobial compositions, and methods of use thereof.

### **BACKGROUND**

During processing, preparation, packaging and serving, food products may  
10   encounter microorganisms that may make the food unsuitable for consumption. The microorganisms may come from the food itself, food contact surfaces, and/or the surrounding environment. The microorganisms can range from pathogenic microorganisms (e.g., *Listeria monocytogenes*, enterohemorrhagic *Escherichia coli*, *Salmonella* and the like) to spoilage organisms that can affect the taste, color, and/or  
15   smell of the final food product (e.g. *Pseudomonas spp*, *Erwinia carotovora*, *Fusarium spp* and the like). Microorganisms can affect a wide variety of food products including meat, poultry, fish and shellfish, cheese, fruits and vegetables, and pre-prepared foods. At certain levels, the presence of microorganisms on a food product may cause everything from a consumer's perception of a lower quality product, to regulatory  
20   investigations and sanctions, to food borne illness or death.

Food processors, grocery retailers, full service restaurants (FSR) and quick service restaurants (QSR) use a variety of methods to treat food products during processing to reduce the presence of microorganisms on food products. These methods

include cleaning and sanitizing the food processing plant and food handling environment, applying or incorporating antimicrobials to or in the food product, irradiating the food product, applying heat, and others. Applying or incorporating an antimicrobial composition to the food product, or to the water used in washing or  
5 transporting the food product, is a preferred way of reducing microorganisms. However, it is difficult to formulate a composition that is effective at reducing microorganisms using ingredients that are acceptable for direct food contact according to government regulations. Further, it is difficult to formulate a composition that can be applied directly to a food product without adversely affecting the color, taste, or smell  
10 of the food product. Finally, many antimicrobials used in the treatment of food products in the retail grocery, FSR and QSR markets may have worker safety concerns, material compatibility (corrosion) concerns or must be generated onsite requiring more costly equipment to dispense and use the antimicrobial.

15

## **BRIEF SUMMARY OF THE INVENTION**

In some aspects, the present disclosure relates to single phase aqueous low foaming antimicrobial concentrate compositions. The compositions comprise at least about 25 wt% water; an antimicrobial agent comprising an anionic surfactant; a defoaming agent; an acidulant; a stabilizing agent; and a thickening agent. The  
20 compositions have a viscosity of between about 50 centipoise and about 3500 centipoise, and the thickening agent is present in an amount effective such that the composition is phase stable under acidic conditions.

In some embodiments, the anionic surfactant is selected from the group consisting of alkyl sulfates, alkyl sulfonates, alkyl aryl sulfonates, sulfonated fatty acids, sulfonated fatty acid esters, sulfonated carboxylic acid esters, and mixtures thereof. In other embodiments, the composition is substantially free of a C5 to C11  
5 carboxylic acid, a chlorite, and an oxidizing agent, and mixtures thereof.

In some embodiments, the defoaming agent comprises a silicone defoaming agent. In other embodiments, the thickening agent is selected from the group consisting of natural polysaccharide or cellulose thickeners, plant exudates, seaweed extracts, water soluble polymers, poly acrylic acid based thickeners, polyacrylamide based  
10 thickeners, inorganic clay based thickeners, and mixtures thereof.

In other embodiments, the acidulant comprises a food additive ingredient. In still yet other embodiments, the acidulant is selected from the group consisting of citric acid, sodium bisulfate, acetic acid, adipic acid, tartaric acid, propionic acid, malic acid, lactic acid, sulfuric acid, and derivatives and mixtures thereof. In some embodiments,  
15 the composition comprises: between about 0.01 wt% and about 50 wt% of the antimicrobial agent; between about 0.01 wt% and about 10.0 wt% defoaming agent; between about 0.1 wt% and about 10 wt% thickening agent; and between about 1 wt% to about 50 wt% of the acidulant.

In some embodiments, the composition has a pH of less than 3.2. In other  
20 embodiments, the composition further comprises an additional ingredient selected from the group consisting of a surfactant, a processing aid, a dye, a colorant, an odorant, and mixtures thereof. In still yet other embodiments, the composition comprises between about 0.1 wt% and about 5.0 wt% stabilizing agent.

In other embodiments, the stabilizing agent is present at an amount effect to prevent the formation of a precipitate for at least 24 hours when the composition is diluted with 500 ppm hard water. In still yet other embodiments, the stabilizing agent comprises a block copolymer of ethylene oxide and propylene oxide. In other  
5   embodiments, the copolymers have a molecular weight of about 5,000 to about 10,000 and the percentage of ethylene oxide is between about 70 to about 90.

In some aspects, the present disclosure relates to methods for reducing a population of microorganisms on a surface. The methods comprise providing a single phase aqueous low foaming antimicrobial concentrate composition. The composition  
10   comprises at least about 25 wt% water; an antimicrobial agent comprising an anionic surfactant; a defoaming agent; an acidulant selected from the group consisting of sulfates, sulfonates, and combinations thereof; a stabilizing agent; and a thickening agent. The composition has a viscosity of between about 50 centipoise and about 3500 centipoise, and the thickening agent is present in an amount effective such that the  
15   composition is phase stable under acidic conditions. The method further comprises diluting the composition; and contacting the surface with the diluted composition such that the population of microorganisms is reduced, wherein the composition does not need to be rinsed from the surface.

In some aspects, the present disclosure relates to methods for washing produce.  
20   The methods comprise contacting the produce with a phase stable aqueous acidic antimicrobial composition comprising at least about 25wt% water; an antimicrobial agent comprising an anionic surfactant; a defoaming agent; an acidulant; a stabilizing agent; and a thickening agent. The composition is diluted prior to contacting, and has a

viscosity of between about 50 centipoise and about 3500 centipoise, and the thickening agent is present in an amount effective such that the composition is phase stable under acidic conditions, and the produce does not need to be rinsed after being contacted by the composition.

5

### **BRIEF DESCRIPTION OF THE DRAWINGS**

Figure 1 is a graphical depiction of liquid retention and residual chemistry carryover onto lettuce versus produce wash concentration.

### **DETAILED DESCRIPTION OF THE INVENTION**

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In some aspects, the present disclosure relates to aqueous low foaming antimicrobial compositions, and methods of use thereof. The compositions are stable, single phase compositions that can be applied directly to food surfaces, and food contact surfaces, without the need to be rinsed. That is, the compositions can be applied directly to a food surface, e.g., fruit or vegetable surface, cutting board, and do not need to be rinsed off prior to consuming or using the surface. Further, the compositions do not substantially alter the organoleptic properties, e.g., color, texture, odor, or taste, of a food surface that they contact. Thus, the compositions provide a no-rinse method for cleaning food surfaces.

15

### **Definitions**

So that the invention maybe more readily understood, certain terms are first defined. For the following defined terms, these definitions shall be applied, unless a different definition is given in the claims or elsewhere in this specification.

As used herein, the phrases “objectionable odor,” “offensive odor,” or “malodor,” refer to a sharp, pungent, or acrid odor or atmospheric environment from which a typical person withdraws if they are able to. Hedonic tone provides a measure of the degree to which an odor is pleasant or unpleasant. An “objectionable odor,”  
5 “offensive odor,” or “malodor” has an hedonic tone rating it as unpleasant as or more unpleasant than a solution of 5 wt-% acetic acid, propionic acid, butyric acid, or mixtures thereof.

As used herein, the term "microorganism" refers to any noncellular or unicellular (including colonial) organism. Microorganisms include all prokaryotes.  
10 Microorganisms include bacteria (including cyanobacteria), spores, lichens, fungi, protozoa, virinos, viroids, viruses, phages, and some algae. As used herein, the term “microbe” is synonymous with microorganism.

As used herein, the phrase "food product" includes any food substance that might require treatment with an antimicrobial agent or composition and that is edible  
15 with or without further preparation. Food products include meat (e.g. red meat and pork), seafood, poultry, produce (e.g., fruits and vegetables), eggs, living eggs, egg products, ready to eat food, wheat, seeds, roots, tubers, leafs, stems, corns, flowers, sprouts, seasonings, or a combination thereof. The term "produce" refers to food products such as fruits and vegetables and plants or plant-derived materials that are  
20 typically sold uncooked and, often, unpackaged, and that can sometimes be eaten raw.

As used herein, the phrase “plant” or "plant product" includes any plant substance or plant-derived substance. Plant products include, but are not limited to,

seeds, nuts, nut meats, cut flowers, plants or crops grown or stored in a greenhouse, house plants, and the like. Plant products include many animal feeds.

As used herein, the phrase "meat product" refers to all forms of animal flesh, including the carcass, muscle, fat, organs, skin, bones and body fluids and like components that form the animal. Animal flesh includes, but is not limited to, the flesh of mammals, birds, fishes, reptiles, amphibians, snails, clams, crustaceans, other edible species such as lobster, crab, etc., or other forms of seafood. The forms of animal flesh include, for example, the whole or part of animal flesh, alone or in combination with other ingredients. Typical forms include, for example, processed meats such as cured meats, sectioned and formed products, minced products, finely chopped products, ground meat and products including ground meat, whole products, and the like.

As used herein the term "poultry" refers to all forms of any bird kept, harvested, or domesticated for meat or eggs, and including chicken, turkey, ostrich, game hen, squab, guinea fowl, pheasant, quail, duck, goose, emu, or the like and the eggs of these birds. Poultry includes whole, sectioned, processed, cooked or raw poultry, and encompasses all forms of poultry flesh, by-products, and side products. The flesh of poultry includes muscle, fat, organs, skin, bones and body fluids and like components that form the animal. Forms of animal flesh include, for example, the whole or part of animal flesh, alone or in combination with other ingredients. Typical forms include, for example, processed poultry meat, such as cured poultry meat, sectioned and formed products, minced products, finely chopped products and whole products.

As used herein, the phrase "poultry debris" refers to any debris, residue, material, dirt, offal, poultry part, poultry waste, poultry viscera, poultry organ,

fragments or combinations of such materials, and the like removed from a poultry carcass or portion during processing and that enters a waste stream.

As used herein, the phrase “food processing surface” refers to a surface of a tool, a machine, equipment, a structure, a building, or the like that is employed as part of a food processing, preparation, or storage activity. Examples of food processing surfaces include surfaces of food processing or preparation equipment (e.g., slicing, canning, or transport equipment, including flumes), of food processing wares (e.g., utensils, dishware, wash ware, and bar glasses), and of floors, walls, or fixtures of structures in which food processing occurs. Food processing surfaces are found and employed in food anti-spoilage air circulation systems, aseptic packaging sanitizing, food refrigeration and cooler cleaners and sanitizers, ware washing sanitizing, blancher cleaning and sanitizing, food packaging materials, cutting board additives, third-sink sanitizing, beverage chillers and warmers, meat chilling or scalding waters, autodish sanitizers, sanitizing gels, cooling towers, food processing antimicrobial garment sprays, and non-to-low-aqueous food preparation lubricants, oils, and rinse additives.

As used herein, the term “ware” refers to items such as eating and cooking utensils, dishes, and other hard surfaces such as showers, sinks, toilets, bathtubs, countertops, windows, mirrors, transportation vehicles, and floors. As used herein, the term “warewashing” refers to washing, cleaning, or rinsing ware. Ware also refers to items made of plastic. Types of plastics that can be cleaned with the compositions according to the invention include but are not limited to, those that include polycarbonate polymers (PC), acrylonitrile-butadiene-styrene polymers (ABS), and polysulfone polymers (PS). Another exemplary plastic that can be cleaned using the

compounds and compositions of the invention include polyethylene terephthalate (PET).

As used herein, the phrase “air streams” includes food anti-spoilage air circulation systems. Air streams also include air streams typically encountered in hospital, surgical, infirmity, birthing, mortuary, and clinical diagnosis rooms.

As used herein, the term “waters” includes food process or transport waters. Food process or transport waters include produce transport waters (e.g., as found in flumes, pipe transports, cutters, slicers, blanchers, retort systems, washers, misting equipment and the like), belt sprays for food transport lines, boot and hand-wash dip-pans, third-sink rinse waters, and the like. Waters also include domestic and recreational waters such as pools, spas, recreational flumes and water slides, fountains, and the like.

As used herein, the phrase “health care surface” refers to a surface of an instrument, a device, a cart, a cage, furniture, a structure, a building, or the like that is employed as part of a health care activity. Examples of health care surfaces include surfaces of medical or dental instruments, of medical or dental devices, of electronic apparatus employed for monitoring patient health, and of floors, walls, or fixtures of structures in which health care occurs. Health care surfaces are found in hospital, surgical, infirmity, birthing, mortuary, and clinical diagnosis rooms. These surfaces can be those typified as “hard surfaces” (such as walls, floors, bed-pans, etc.), or fabric surfaces, e.g., knit, woven, and non-woven surfaces (such as surgical garments, draperies, bed linens, bandages, etc.), or patient-care equipment (such as respirators, diagnostic equipment, shunts, body scopes, wheel chairs, beds, etc.), or surgical and

diagnostic equipment. Health care surfaces include articles and surfaces employed in animal health care.

As used herein, the term “instrument” refers to the various medical or dental instruments or devices that can benefit from cleaning with a composition according to  
5 the present invention.

As used herein, the phrases “medical instrument,” “dental instrument,” “medical device,” “dental device,” “medical equipment,” or “dental equipment” refer to instruments, devices, tools, appliances, apparatus, and equipment used in medicine or dentistry. Such instruments, devices, and equipment can be cold sterilized, soaked or  
10 washed and then heat sterilized, or otherwise benefit from cleaning in a composition of the present invention. These various instruments, devices and equipment include, but are not limited to: diagnostic instruments, trays, pans, holders, racks, forceps, scissors, shears, saws (e.g., bone saws and their blades), hemostats, knives, chisels, rongeurs, files, nippers, drills, drill bits, rasps, burrs, spreaders, breakers, elevators, clamps,  
15 needle holders, carriers, clips, hooks, gouges, curettes, retractors, straightener, punches, extractors, scoops, keratomes, spatulas, expressors, trocars, dilators, cages, glassware, tubing, catheters, cannulas, plugs, stents, scopes (e.g., endoscopes, stethoscopes, and arthoscopes) and related equipment, and the like, or combinations thereof.

As used herein, “agricultural” or “veterinary” objects or surfaces include animal  
20 feeds, animal watering stations and enclosures, animal quarters, animal veterinarian clinics (e.g., surgical or treatment areas), animal surgical areas, and the like.

As used herein, the term “phosphorus-free” or “substantially phosphorus-free” refers to a composition, mixture, or ingredient that does not contain phosphorus or a

phosphorus-containing compound or to which phosphorus or a phosphorus-containing compound has not been added. Should phosphorus or a phosphorus-containing compound be present through contamination of a phosphorus-free composition, mixture, or ingredients, the amount of phosphorus shall be less than 0.5 wt %. More preferably, the amount of phosphorus is less than 0.1 wt%, and most preferably the amount of phosphorus is less than 0.01 wt %.

For the purpose of this patent application, successful microbial reduction is achieved when the microbial populations, e.g., microbial populations on surfaces or in water, are reduced by at least about 50%, or by significantly more than is achieved by a wash with water. Larger reductions in microbial population provide greater levels of protection.

As used herein, the term “sanitizer” refers to an agent that reduces the number of bacterial contaminants to safe levels as judged by public health requirements. In an embodiment, sanitizers for use in this invention will provide at least a 99.999% reduction (5-log order reduction). These reductions can be evaluated using a procedure set out in *Germicidal and Detergent Sanitizing Action of Disinfectants*, Official Methods of Analysis of the Association of Official Analytical Chemists, paragraph 960.09 and applicable sections, 15th Edition, 1990 (EPA Guideline 91-2). According to this reference a sanitizer should provide a 99.999% reduction (5-log order reduction) within 30 seconds at room temperature, 25±2°C, against several test organisms.

As used herein, the term “disinfectant” refers to an agent that kills all vegetative cells including most recognized pathogenic microorganisms, using the procedure described in *A.O.A.C. Use Dilution Methods*, Official Methods of Analysis of the

Association of Official Analytical Chemists, paragraph 955.14 and applicable sections, 15th Edition, 1990 (EPA Guideline 91-2). As used herein, the term “high level disinfection” or “high level disinfectant” refers to a compound or composition that kills substantially all organisms, except high levels of bacterial spores, and is effected with a chemical germicide cleared for marketing as a sterilant by the Food and Drug Administration. As used herein, the term “intermediate-level disinfection” or “intermediate level disinfectant” refers to a compound or composition that kills mycobacteria, most viruses, and bacteria with a chemical germicide registered as a tuberculocide by the Environmental Protection Agency (EPA). As used herein, the term “low-level disinfection” or “low level disinfectant” refers to a compound or composition that kills some viruses and bacteria with a chemical germicide registered as a hospital disinfectant by the EPA.

As used in this invention, the term “sporicide” refers to a physical or chemical agent or process having the ability to cause greater than a 90% reduction (1-log order reduction) in the population of spores of *Bacillus cereus* or *Bacillus subtilis* within 10 seconds at 60° C. In certain embodiments, the sporicidal compositions of the invention provide greater than a 99% reduction (2-log order reduction), greater than a 99.99% reduction (4-log order reduction), or greater than a 99.999% reduction (5-log order reduction) in such population within 10 seconds at 60° C.

Differentiation of antimicrobial “-cidal” or “-static” activity, the definitions which describe the degree of efficacy, and the official laboratory protocols for measuring this efficacy are considerations for understanding the relevance of antimicrobial agents and compositions. Antimicrobial compositions can affect two

kinds of microbial cell damage. The first is a lethal, irreversible action resulting in complete microbial cell destruction or incapacitation. The second type of cell damage is reversible, such that if the organism is rendered free of the agent, it can again multiply. The former is termed microbiocidal and the later, microbiostatic. A sanitizer and a disinfectant are, by definition, agents which provide antimicrobial or microbiocidal activity. In contrast, a preservative is generally described as an inhibitor or microbiostatic composition

As used herein, the term "alkyl" or "alkyl groups" refers to saturated hydrocarbons having one or more carbon atoms, including straight-chain alkyl groups (e.g., methyl, ethyl, propyl, butyl, pentyl, hexyl, heptyl, octyl, nonyl, decyl, etc.), cyclic alkyl groups (or "cycloalkyl" or "alicyclic" or "carbocyclic" groups) (e.g., cyclopropyl, cyclopentyl, cyclohexyl, cycloheptyl, cyclooctyl, etc.), branched-chain alkyl groups (e.g., isopropyl, tert-butyl, sec-butyl, isobutyl, etc.), and alkyl-substituted alkyl groups (e.g., alkyl-substituted cycloalkyl groups and cycloalkyl-substituted alkyl groups).

Unless otherwise specified, the term "alkyl" includes both "unsubstituted alkyls" and "substituted alkyls." As used herein, the term "substituted alkyls" refers to alkyl groups having substituents replacing one or more hydrogens on one or more carbons of the hydrocarbon backbone. Such substituents may include, for example, alkenyl, alkynyl, halogeno, hydroxyl, alkylcarbonyloxy, arylcarbonyloxy, alkoxycarbonyloxy, aryloxy, aryloxycarbonyloxy, carboxylate, alkylcarbonyl, arylcarbonyl, alkoxycarbonyl, aminocarbonyl, alkylaminocarbonyl, dialkylaminocarbonyl, alkylthiocarbonyl, alkoxyl, phosphate, phosphonato, phosphinato, cyano, amino (including alkyl amino, dialkylamino, arylamino, diarylamino, and alkylarylamino), acylamino (including

alkylcarbonylamino, arylcarbonylamino, carbamoyl and ureido), imino, sulfhydryl, alkylthio, arylthio, thiocarboxylate, sulfates, alkylsulfinyl, sulfonates, sulfamoyl, sulfonamido, nitro, trifluoromethyl, cyano, azido, heterocyclic, alkylaryl, or aromatic (including heteroaromatic) groups.

5           In some embodiments, substituted alkyls can include a heterocyclic group. As used herein, the term “heterocyclic group” includes closed ring structures analogous to carbocyclic groups in which one or more of the carbon atoms in the ring is an element other than carbon, for example, nitrogen, sulfur or oxygen. Heterocyclic groups may be saturated or unsaturated. Exemplary heterocyclic groups include, but are not limited to,  
10   aziridine, ethylene oxide (epoxides, oxiranes), thiirane (episulfides), dioxirane, azetidine, oxetane, thietane, dioxetane, dithietane, dithiete, azolidine, pyrrolidine, pyrroline, oxolane, dihydrofuran, and furan.

          All numeric values are herein assumed to be modified by the term “about,” whether or not explicitly indicated. The term “about” generally refers to a range of  
15   numbers that one of skill in the art would consider equivalent to the recited value (i.e., having the same function or result). In many instances, the term “about” may include numbers that are rounded to the nearest significant figure.

          Weight percent, percent by weight, % by weight, wt %, and the like are synonyms that refer to the concentration of a substance as the weight of that substance  
20   divided by the weight of the composition and multiplied by 100.

          The recitation of numerical ranges by endpoints includes all numbers subsumed within that range (e.g. 1 to 5 includes 1, 1.5, 2, 2.75, 3, 3.80, 4 and 5).

As used in this specification and the appended claims, the singular forms “a,” “an,” and “the” include plural referents unless the content clearly dictates otherwise. Thus, for example, reference to a composition containing “a compound” includes a mixture of two or more compounds. As used in this specification and the appended  
5 claims, the term “or” is generally employed in its sense including “and/or” unless the content clearly dictates otherwise.

The use of the terms “antimicrobial” and “biocide” in this application does not mean that any resulting products are approved for use as an antimicrobial agent or biocide.

10 The compositions and methods disclosed herein may comprise, consist of, or consist essentially of the listed ingredients, or steps. As used herein the term “consisting essentially of” refers to a composition or method that includes the disclosed ingredients or steps, and any other ingredients or steps that do not materially affect the novel and basic characteristics of the compositions or methods. For example,  
15 compositions that consist essentially of the listed ingredients do not contain additional ingredients that would affect, for example, the antimicrobial or rheological properties of the compositions.

### Compositions

20 The good detergency and broad biocidal efficacy of Generally Regarded As Safe (GRAS) listed anionic surfactants make them ideal to be used as active ingredients in a produce wash formula. However, the high foam profile of these surfactants makes it hard for them to be used in a convenient liquid form suitable for an automated

dispensing system, such as an Oasis Pro aspirator dispenser available from Ecolab.

Dispensing systems often generate large amounts of stable foam during the dispensing process from the concentrate form. The presence of foam is detrimental when used in a

no rinse formulation, e.g., a no rinse produce wash formulation, as it not only impacts  
5 the visual appearance of the contacted surface, but it can also change the taste, odor, texture and appearance of the contacted surface, e.g., produce.

As a result, it is preferred to eliminate the foam in formulas employing such ingredients. Incorporation of an appropriate defoamer into the composition is one approach to eliminate foam. Indeed, some commercial antimicrobial produce wash

10 products employing anionic surfactant as active ingredients do contain defoamer, such as silicon defoamer. However, these products are in the form of powder, making them much easier to formulate incorporating a defoamer. On the other hand, the silicon defoamer poses significant technical challenges when incorporated into a primarily aqueous liquid form, as these silicon particles tend to aggregate in the composition, and  
15 cause phase stability issues.

In some aspects, the present disclosure relates to single phase, aqueous, low foaming antimicrobial compositions. The compositions are rheology modified such that they remain as a single phase in liquid form, and are low or no foaming, even when agitated, e.g., dispensed through an aspirator, or applied in an agitated bath, e.g., flume  
20 waters. Further, the compositions do not need to be rinsed from the surface they contact prior to consumption or use.

The compositions include at least an antimicrobial agent, an acidulant, a defoaming agent, and a thickening agent. The compositions further include at least

about 25 wt% water. The compositions are phase stable liquid compositions. That is, they are not solids, e.g., powders, tablets, granules, etc. In some embodiments, the compositions do not include oxidizing agents, chlorites, or mixtures thereof. Nor do the compositions include volatile solvents, e.g., monohydric alcohol volatile solvents.

5           *Antimicrobial Agents*

In some aspects, the compositions include an antimicrobial agent. In some embodiments, the antimicrobial agents suitable for use with the compositions of the invention are GRAS antimicrobial agents or food grade compositions.

10           In some embodiments, the antimicrobial agent is selected from the group consisting of anionic surfactants, quaternary ammonium compounds and mixtures thereof. In some embodiments, the compositions can include more than one antimicrobial agent. For example, in some embodiments, the compositions can include one, two, three or four antimicrobial agents.

15           In some embodiments, the compositions include an anionic surfactant as an antimicrobial agent. The anionic surfactant can include, but is not limited to, sulfates, sulfonates, and combinations thereof. Exemplary anionic surfactants for use with the present compositions include alkyl sulfates, alkyl sulfonates, alkyl aryl sulfonates, sulfonated fatty acids, sulfonated fatty acid esters, sulfonated carboxylic acid esters, and mixtures thereof. Sulfonated fatty acids suitable for use include, but are not limited to, 20   sulfonated oleic acid, sulfonated linoleic acid, sulfonated palmitoleic acid and sulfonated stearic acid.

Additional exemplary antimicrobial agents include, but are not limited to, sodium dodecylbenzene sulfonate, sodium dodecyl sulfate, sodium dioctyl

sulfosuccinate, sulfonated oleic acid,  $\alpha$  – sulfonated carboxylic acid, dialkyl dimethyl ammonium chloride, alkylbenzyl dimethyl ammonium chloride and combinations thereof. In some embodiments, the compositions are free or substantially free of a chlorite and/or an oxidizing agent.

5           In some embodiments, the compositions include between about 0.01 to about 20 wt% of one or more antimicrobial agent, about 0.01 wt% to about 10 wt% of one or more antimicrobial agent, or about 1.0 wt% to about 5 wt% of one or more antimicrobial agent. It is to be understood that all values and ranges between these values and ranges are included in the present disclosure.

10           *Defoaming Agents*

The compositions further include a defoaming agent. In some embodiments, the compositions can include defoamers which are of food grade quality. To this end, silicones can be used as effective defoaming agents. The compositions are designed such that silicones can be incorporated as defoaming agents even though the  
15       compositions are in liquid form. Silicones such as dimethyl silicone, glycol polysiloxane, methylphenol polysiloxane, trialkyl or tetraalkyl silanes, hydrophobic silica defoamers and mixtures thereof can all be used as defoamers in the compositions. Commercial defoamers commonly available include silicones such as Ardefoam® from Armour Industrial Chemical Company which is a silicone bound in an organic  
20       emulsion; Foam Kill® or Kresseo® available from Krusable Chemical Company which are silicone and non-silicone type defoamers as well as silicone esters; and Anti-Foam A® and DC-200 commercially available from Dow Corning Corporation. Other additional defoaming agents suitable for use with the compositions include, but are not

limited to, FG10, and 1520, commercially available from Dow Corning Corporation, and Hodag FD-82K, commercially available from Lambent. A defoaming agent can be present at a concentration range from about 0.01 wt% to 20 wt%, from about 0.1 wt% to 10 wt%, or from about 0.01 wt% to about 2 wt%. In some embodiments, the  
5 compositions include more than zero but less than 1 wt% defoaming agent. It is to be understood that all values and ranges between these values and ranges are incorporated in the present disclosure.

#### *Acidulants*

10 In some embodiments, the compositions include an acidulant. The acidulant can be present at an amount effective to form a concentrate composition with pH of about 1 or less. In other embodiments, the acidulant can be present at an amount effective to form a use composition with pH of about 5, about 5 or less, about 4, about 4 or less, about 3, about 3 or less, about 2, about 2 or less, or the like. In some embodiments, the  
15 composition has a pH of between about 0 to about 6, about 1 to about 5, or about 2 to about 4. In other embodiments, the composition has a pH of less than about 3.2.

In some embodiments, the acidulant includes an inorganic acid. Suitable inorganic acids include, but are not limited to, sulfuric acid, sodium bisulfate, phosphoric acid, nitric acid, hydrochloric acid, and mixtures thereof. In some  
20 embodiments, the acidulant includes an organic acid. Suitable organic acids include, but are not limited to, methane sulfonic acid, ethane sulfonic acid, propane sulfonic acid, butane sulfonic acid, xylene sulfonic acid, benzene sulfonic acid, formic acid, acetic acid, mono, di, or tri-halocarboxylic acids, picolinic acid, dipicolinic acid,

levulinic acid and mixtures thereof. In some embodiments, the compositions of the present invention are free or substantially free of a phosphorous based acid.

The compositions may also include a carboxylic acid. In some embodiments, the carboxylic acid includes, but is not limited to, an alpha hydroxy carboxylic acid, sulfonated carboxylic acids, and mixtures thereof. As used herein, the term “alpha hydroxy carboxylic acid” refers to chemical compounds that include a carboxylic acid substituted with a hydroxyl group on the adjacent carbon. Alpha hydroxy carboxylic acids include, for example, glycolic acid, lactic acid, malic acid, citric acid and tartaric acid.

10 In some embodiments, the carboxylic acid for use with the compositions of the present invention includes a C1 to C22 carboxylic acid. In some embodiments, the carboxylic acid for use with the compositions is a C5 to C11 carboxylic acid. In some embodiments, the carboxylic acid for use with the compositions is a C1 to C4 carboxylic acid. Examples of suitable carboxylic acids include, but are not limited to, formic, acetic, propionic, butanoic, pentanoic, hexanoic, heptanoic, octanoic, nonanoic, 15 decanoic, undecanoic, dodecanoic, as well as their branched isomers, lactic, maleic, ascorbic, citric, hydroxyacetic, neopentanoic, neoheptanoic, neodecanoic, oxalic, malonic, succinic, glutaric, adipic, pimelic subric acid, and mixtures thereof. In some embodiments, the composition is substantially free, or free of a C5 to C11 carboxylic acid. In other embodiments, the composition is substantially free, or free of a C1 to C4 20 carboxylic acid. In some embodiments, the composition is substantially free, or free of both a C1 to C4 carboxylic acid, and a C5 to C11 carboxylic acid.

In some embodiments, the compositions include more than one acidulant. For example, in some embodiments, the compositions include one, two, three or four acidulants.

In some embodiments, the acidulant selected can also function as a stabilizing agent. Thus, the compositions of the present invention can be substantially free of an additional stabilizing agent.

In certain embodiments, the present composition includes about 0.5 to about 80 wt-% acidulant, about 1 to about 50 wt%, about 5 to about 30 wt-% acidulant, or about 10 wt% to about 30 wt-% acidulant. It is to be understood that all values and ranges between these values and ranges are encompassed by the compositions disclosed herein.

#### *Stabilizing Agent*

In some embodiments, the compositions also include a stabilizing agent. In some embodiments, the stabilizing agent is a food grade or GRAS stabilizing agent. The stabilizing agents aid in maintaining stability of the compositions. For example, in some embodiments, the stabilizing agents aid in maintaining the stability of the compositions in hard water. In some embodiments, the stabilizing agent is present at an amount effect to prevent the formation of a precipitate for at least 24 hours when the composition is diluted with 500 ppm hard water.

Any of a variety of stabilizing agents can be used in the disclosed compositions. In some embodiments, the stabilizing agent includes an ethylene oxide/propylene oxide block copolymer, mixtures thereof, or the like. In some embodiments, the copolymers have a molecular weight of about 5000 to about 10,000 and the percentage of ethylene

oxide is between about 70 to about 90. Suitable ethylene oxide/propylene oxide block copolymers include for example, those sold under the Pluronic tradename (e.g., Pluronic F108 and Pluronic F68) and commercially available from BASF Corporation. Other exemplary stabilizers for use in the present invention include nonionic surfactants and emulsifiers, for example, polysorbate 80.

In some embodiments, one or more stabilizing agent may be present. In some embodiments, the stabilizing agent is present at between about 0.1 wt% and about 5.0 wt% of the composition. In other embodiments, the stabilizing agent is present between about 0.1 wt% and about 1.0 wt%. It is to be understood that all values and ranges between these values and ranges are encompassed by the present disclosure.

#### *Thickening Agents*

The compositions also include a thickening agent. The thickening agents may be food grade or GRAS. The thickening agents increase the viscosity of the compositions, and allow for the incorporation of a defoaming agent into the compositions under acidic conditions. That is, the thickening agent is present at an amount effective to maintain a phase stable composition when the defoaming agent is present.

In some embodiments, the thickening agent is selected from the group consisting of natural polysaccharide or cellulose thickeners, plant exudates, seaweed extracts, water soluble polymers, polyacrylic acid based thickeners, polyacrylamide based thickeners, inorganic clay based thickeners, and mixtures thereof. Natural polysaccharide or cellulose thickeners include, but are not limited to, guar gum, locust

bean gum, xanthan gum, pectin, and gellan gum. Plant exudates include, but are not limited to, acacia, ghatti, and tragacanth. Seaweed extracts include, but are not limited to, sodium alginate, and sodium carrageenan. Suitable polymers include, but are not limited to, hydroxyethyl cellulose, hydroxypropyl cellulose, hydroxypropyl methylcellulose, and dextrans.

In some embodiments, the composition has a viscosity of between about 50 centipoise and about 3,500 centipoise when tested with spindle 3 at 50 rpm on a Brookfield RVDVI+ viscometer, and the thickening agent is present in an amount effective such that the composition is phase stable under acidic conditions. In other embodiments, the composition includes between about 0.1 wt% and about 10 wt% thickening agent. In other embodiments, the composition includes less than about 1.0 wt% of a thickening agent.

#### *Carrier*

In some embodiments, the compositions include a carrier. The carrier provides a medium which dissolves, suspends, or carries the other components of the composition. The carrier concentration and type will depend upon the nature of the composition as a whole, the environmental storage, and method of application including concentration of the antimicrobial agent, among other factors. Notably the carrier should be chosen and used at a concentration which does not inhibit the antimicrobial efficacy of the active agent in the composition of the invention. When used to wash a food contact surface, viz. produce, the carrier selected should be one that would not adversely affect the food contact surface.

In some embodiments, the carrier includes water. In some embodiments, the carrier does not include volatile solvents. For example, in some embodiments, the compositions do not include volatile monohydric alcohol solvents, including, but not limited to, aliphatic alcohols or a glycol ether.

5           The carrier can be included in the compositions at a concentration of at least about 25 wt%, at least 25 wt%, at least about 50 wt%, at least 50 wt%, from about 20 wt% to about 90 wt%, or from about 40 wt% to about 70 wt%. It is to be understood that all values and ranges between these values and ranges are to be encompassed by the present disclosure.

10

#### *Dye*

In some embodiments, the compositions include one or more dyes. The dyes can be food grade or GRAS dyes. The dyes aid in providing evidence that the antimicrobial composition is present in the treatment solution, e.g., in the sink or wash  
15   water. The dyes for use with the compositions must be stable at a low pH, e.g., pH less than about 2. Dyes suitable for use with the disclosed compositions include, but are not limited to FD&C Green 3, FD& C Yellow 5, and mixtures thereof.

If included, a dye is present at an amount effective to be visible when the composition is diluted to use level. In some embodiments, the dye is present at between  
20   about 0.0001 wt% and about 1.0 wt%, or between about 0.001 wt% and about 0.01 wt%. It is to be understood that all values and ranges between these values and ranges are encompassed by the present disclosure.

*Additional Ingredients*

The compositions of the invention can include additional ingredients. The additional ingredients can include, but are not limited to, a surfactant, a processing aid, a colorant, an odorant, an anti-browning agent and mixtures thereof.

5

*Use Compositions and Methods of Use thereof*

The compositions include concentrate compositions and use compositions. For example, a concentrate composition can be diluted, for example with water, to form a use composition. In an embodiment, a concentrate composition can be diluted to a use solution before to application to an object. For reasons of economics, the concentrate can be marketed and an end user can dilute the concentrate with water or an aqueous diluent to a use solution. In other embodiments, a use composition can include about 0.01 to about 10 wt% of a concentrate composition and about 90 to about 99.99 wt% diluent; or about 0.1 to about 2 wt% of a concentrate composition and about 98 to about 99.9 wt-% diluent.

15

In some embodiments, the compositions are diluted with water. The compositions can be diluted at a ratio of between about 1:1 to about 1:512. In other embodiments, the compositions can be diluted at a ratio of between about 1:64 to about 1:256. The dilution ratio can vary depending on the diluent being used.

20

In some aspects, the present disclosure includes methods of using the disclosed antimicrobial compositions. For example, the methods include a method for reducing a microbial population, a method for reducing the population of a microorganism on skin, a method for treating a disease of skin, and/or a method for reducing an odor. These

methods can operate on an article, surface, in a body or stream of water or a gas, or the like, by contacting the article, surface, body, or stream with a composition. Contacting can include any of numerous methods for applying composition, such as spraying the compositions, immersing the article in the compositions or a combination thereof.

5           In some aspects, the methods include a composition present at an amount effective for killing one or more of the food-borne pathogenic bacteria associated with a food product, including, but not limited to, *Salmonella typhimurium*, *Salmonella javiana*, *Salmonella enterica*, *Campylobacter jejuni*, *Listeria monocytogenes*, and *Escherichia coli* O157:H7, yeast, and mold. In some embodiments, the methods  
10   include applying compositions at an amount effective for killing one or more of the pathogenic bacteria associated with a health care surfaces and environments including, but not limited to, *Salmonella typhimurium*, *Staphylococcus aureus*, methicilin resistant *Staphylococcus aureus*, *Salmonella choleraesuis*, *Pseudomonas aeruginosa*, *Escherichia coli*, mycobacteria, yeast, and mold. The compositions have activity  
15   against a wide variety of microorganisms such as Gram positive (for example, *Listeria monocytogenes* or *Staphylococcus aureus*) and Gram negative (for example, *Escherichia coli* or *Pseudomonas aeruginosa*) bacteria, yeast, molds, bacterial spores, viruses, etc. The compositions of the present invention, as described above, have activity against a wide variety of human pathogens. The present compositions can kill a  
20   wide variety of microorganisms on a food processing surface, on the surface of a food product, in water used for washing or processing of food product, on a health care surface, or in a health care environment.

The compositions can be applied in a variety of areas including kitchens, bathrooms, factories, hospitals, dental offices and food plants, and can be applied to a variety of hard or soft surfaces having smooth, irregular or porous topography. Suitable hard surfaces include, for example, architectural surfaces (e.g., floors, walls, windows, sinks, tables, counters and signs); eating utensils; hard-surface medical or surgical instruments and devices; and hard-surface packaging. Such hard surfaces can be made from a variety of materials including, for example, ceramic, metal, glass, wood or hard plastic. Suitable soft surfaces include, for example paper; filter media; hospital and surgical linens and garments; soft-surface medical or surgical instruments and devices; and soft-surface packaging. Such soft surfaces can be made from a variety of materials including, for example, paper, fiber, woven or nonwoven fabric, soft plastics and elastomers. The compositions can also be applied to soft surfaces such as food and skin (e.g., a hand). The compositions can be included in products such as sterilants, sanitizers, disinfectants, preservatives, deodorizers, antiseptics, fungicides, germicides, sporicides, virucides, detergents, bleaches, hard surface cleaners.

The compositions can also be used in veterinary products such as mammalian skin treatments or in products for sanitizing or disinfecting animal enclosures, pens, watering stations, and veterinary treatment areas such as inspection tables and operation rooms. The present compositions can be employed in an antimicrobial foot bath for livestock or people.

In some aspects, the compositions can be employed for reducing the population of pathogenic microorganisms, such as pathogens of humans, animals, and the like. The compositions exhibit activity against pathogens including fungi, molds, bacteria,

spores, and viruses, for example, *S. aureus*, *E. coli*, *Streptococci*, *Legionella*, *Pseudomonas aeruginosa*, mycobacteria, tuberculosis, phages, or the like. Such pathogens can cause a variety of diseases and disorders, including mastitis or other mammalian milking diseases, tuberculosis, and the like. The compositions can reduce the population of microorganisms on skin or other external or mucosal surfaces of an animal. In addition, the present compositions can kill pathogenic microorganisms that spread through transfer by water, air, or a surface substrate. The compositions need only be applied to the skin, other external or mucosal surfaces of an animal water, air, or surface.

10           The antimicrobial compositions can also be used on foods and plant species to reduce surface microbial populations; used at manufacturing or processing sites handling such foods and plant species; or used to treat process waters around such sites. For example, the compositions can be used on food transport lines (e.g., as belt sprays); boot and hand-wash dip-pans; food storage facilities; anti-spoilage air circulation systems; refrigeration and cooler equipment; beverage chillers and warmers, blanchers, 15 cutting boards, third sink areas, and meat chillers or scalding devices. The compositions can be used to treat produce transport waters such as those found in flumes, pipe transports, cutters, slicers, blanchers, retort systems, washers, and the like. Particular foodstuffs that can be treated with compositions of the invention include 20 eggs, meats, seeds, leaves, fruits and vegetables. Particular plant surfaces include both harvested and growing leaves, roots, seeds, skins or shells, stems, stalks, tubers, corms, fruit, and the like. The compositions may also be used to treat animal carcasses to reduce both pathogenic and non-pathogenic microbial levels.

The compositions can also be employed by dipping food processing equipment into the use solution, soaking the equipment for a time sufficient to sanitize the equipment, and wiping or draining excess solution off the equipment, The compositions may be further employed by spraying or wiping food processing surfaces with the use solution, keeping  
5 the surfaces wet for a time sufficient to sanitize the surfaces, and removing excess solution by wiping, draining vertically, vacuuming, etc.

The compositions may also be used in a method of sanitizing hard surfaces such as institutional type equipment, utensils, dishes, health care equipment or tools, and other hard surfaces. The antimicrobial compositions can be applied to microbes or to  
10 soiled or cleaned surfaces using a variety of methods. These methods can operate on an object, surface, in a body or stream of water or a gas, or the like, by contacting the object, surface, body, or stream with a compound of the invention. Contacting can include any of numerous methods for applying a compound, such as spraying the compound, immersing the object in the compound, or a combination thereof.

15 A concentrate or use concentration of a composition can be applied to or brought into contact with an object by any conventional method or apparatus for applying an antimicrobial or cleaning compound to an object. For example, the object can be wiped with, sprayed with, foamed on, and/or immersed in the compound, or a use solution made from the composition. The composition can be sprayed, foamed, or  
20 wiped onto a surface; the composition can be caused to flow over the surface, or the surface can be dipped into the composition. Contacting can be manual or by machine.

#### *Methods for Contacting a Food Product*

In some aspects, the present disclosure provides methods for contacting a food product with an antimicrobial composition employing any method or apparatus suitable for applying such a composition. Contacting the food product can occur in any location in which the food product might be found, such as field, processing site or plant,  
5 vehicle, warehouse, store, restaurant, or home. These same methods can also be adapted to apply the compositions to other objects.

The present methods require a certain minimal contact time of the composition with food product for occurrence of significant antimicrobial effect. The contact time can vary with concentration of the use composition, method of applying the use  
10 composition, temperature of the use composition, amount of soil on the food product, number of microorganisms on the food product, type of antimicrobial agent, or the like. The exposure time can be at least about 5 to about 15 seconds. In some embodiments, the exposure time is about 15 to about 30 seconds. In other embodiments, the exposure time is at least about 30 seconds. In still yet other embodiments, the exposure time is  
15 about one minute, about two minutes, or about four minutes.

In other embodiments, the exposure time is the amount of time it takes for the composition to dry on the contacted surface. For example the treated surface may be exposed to the compositions for about 20 to about 60 minutes, or about 30 to about 45 minutes.

20 In some embodiments, the method for washing a food product employs a pressure spray including the disclosed compositions. During application of the spray solution on the food product, the surface of the food product can be moved with mechanical action, e.g., agitated, rubbed, brushed, etc. Agitation can be by physical

scrubbing of the food product, through the action of the spray solution under pressure, through sonication, or by other methods. In some embodiments, the agitation occurs in a sink, e.g., a PowerSoak® sink. Agitation increases the efficacy of the spray solution in killing micro-organisms, perhaps due to better exposure of the solution into the  
5 crevasses or small colonies containing the micro-organisms. Further, the compositions are formulated such that despite high agitation, they remain low to no foaming.

The spray solution, before application, can also be heated to a temperature of about 15 to 20 °C, for example, about 20 to 60 °C to increase efficacy. The spray stabilized composition can be left on the food product for a sufficient amount of time to  
10 suitably reduce the population of microorganisms, and then rinsed, drained, or evaporated off the food product.

Application of the material by spray can be accomplished using a manual spray wand application, an automatic spray of food product moving along a production line using multiple spray heads to ensure complete contact, or other spray apparatus. One  
15 automatic spray application involves the use of a spray booth. The spray booth substantially confines the sprayed compound to within the booth. The production line moves the food product through the entryway into the spray booth in which the food product is sprayed on all its exterior surfaces with sprays within the booth. After a complete coverage of the material and drainage of the material from the food product  
20 within the booth, the food product can then exit the booth. The spray booth can include steam jets that can be used to apply the stabilized compositions of the invention. These steam jets can be used in combination with cooling water to ensure that the treatment reaching the food product surface is less than 65°C, e.g., less than 60°C. The

temperature of the spray on the food product is important to ensure that the food product is not substantially altered (cooked) by the temperature of the spray. The spray pattern can be virtually any useful spray pattern.

Immersing a food product in a liquid composition can be accomplished by any  
5 of a variety of methods known to those of skill in the art. For example, the food product can be placed into a tank or bath containing the stabilized composition. Alternatively, the food product can be transported or processed in a flume of the stabilized composition. The washing solution can be agitated to increase the efficacy of the solution and the speed at which the solution reduces micro-organisms accompanying  
10 the food product. Agitation can be obtained by conventional methods, including ultrasonics, aeration by bubbling air through the solution, by mechanical methods, such as strainers, paddles, brushes, pump driven liquid jets, or by combinations of these methods. The washing solution can be heated to increase the efficacy of the solution in killing micro-organisms. After the food product has been immersed for a time  
15 sufficient for the desired antimicrobial effect, the food product can be removed from the bath or flume and the stabilized composition can be rinsed, drained, or evaporated off the food product.

The following examples are provided for the purpose of illustration, not limitation.

## EXAMPLES

*Example 1*

A test was performed to evaluate the impact of the addition of a defoaming agent to an exemplary produce wash formulation. Three formulations were tested.

- 5 Compositions A and B were prepared in accordance with embodiments of the present disclosure. Compositions A and B are shown in the table below.

<b>Agent</b>	<b>Composition A</b>	<b>Composition B</b>
Anionic Surfactant	6.84	6.84
C5-C11 Carboxylic Acid	1.00	1.00
Defoaming Agent	0.50	0.00
Lactic Acid	19.65	19.65
Thickening Agent	0.40	0.40
Processing Aid	1.20	1.20
Stabilizing Agent	0.64	0.64
Miscellaneous	0.055	0.055
DI Water	To 100	To 100

- Composition A also included a defoaming agent. Composition B included the same antimicrobial composition as included in Composition A, but did not include a
- 10 defoaming agent. Comparative Composition 1 included a commercially available produce wash, Fit® Fruit and Produce Wash, commercially available from HealthPro Brands, Inc.

- Each composition was diluted to a ratio of 1 ounce of produce wash to 1 gallon of water (5 grain). Then 40 milliliters (mL) of each of the diluted solutions was added
- 15 to a 250 mL graduated cylinder. The samples were then spun on a foam test machine for 4 minutes. The samples were removed from the machine and the foam height was immediately recorded. The graduated cylinders were also immediately photographed.

The change in foam height after two minutes was also measured. The test was then repeated 2 more times. The results from this test are shown in the table below.

Table 1.

Composition	Replicate	Foam Height (mL)	
		Time = 10 seconds	Time = 120 seconds
Composition A	1	0	0
	2	0	0
	3	0	0
Composition B	1	71	66
	2	76	71
	3	71	66
Comparative Composition 1	1	21	16
	2	26	16
	3	21	16

5

As can be seen from these results, Composition A, which included a defoamer, had no foam present at either time point tested. Composition B, which included the same produce wash as Composition A, but did not include a defoamer had a very high and very stable foam. The commercially available composition tested, Comparative  
 10 Composition 1 also formed a stable foam, however, it was not as high as the foam formed from Composition B. Overall, it was shown that the presence of a defoamer

was important for reducing the foam height and stability in compositions in accordance with embodiments of the present invention.

*Example 2*

5           A study using Compositions A and B, as described above in Example 1, was run to evaluate the carryover of residual chemistry onto lettuce as a function of the foam level and the concentration of various produce washes in accordance with the present invention.

          For this study, Dole brand cut and prepackaged romaine lettuce hearts were  
10       used. The lettuce was weighed and placed in a strainer and the vessel was filled with six liters (L) of either Composition A or B. The compositions were dispensed into the vessel using an Oasis® Cleaning System, commercially available from Ecolab Inc. The dispense time was 35 seconds, at a flow rate of approximately 10.3 liters/minute.

          Thirty seconds after the composition was applied, a picture was taken of the  
15       foam and the strainer was removed from the liquid. The strainer was allowed to drain for 30 seconds and another photo was taken to visually document the amount of foam present. The lettuce was then transferred by hand to a plastic zip-lock bag, and weighed. The initial weight of the bag was 11.6 grams.

          This procedure was repeated using varying sized dispenser aspirator tips to  
20       change the composition concentration, and the produce usage and residual carryover weights were recorded. The results from this study are shown in the table below and in Figure 1.

Table 2.

Composition B				
Sample	Antifoam (Y/N)	Weight percent of Composition (wt%)	Liquid retained on lettuce (g)	Residual chemistry (g)
1	N	0.78%	26.85	0.21
2	N	1.81%	27.18	0.49
3	N	4.05%	27.91	1.13
4	N	6.39%	34.16	2.18
5	N	10.15%	48.74	4.95
Composition A				
6	Y	0.86%	18.7	0.16
7	Y	1.81%	20.01	0.36
8	Y	4.15%	19.47	0.81
9	Y	6.88%	21.37	1.47
10	Y	10.50%	22.65	2.38

The results indicate that compositions that generate foam during washing or processing can carryover significantly more liquid / foam and residual chemistry than compositions that do not generate significant amounts of foam. The inclusion of antifoam to Composition A resulted in minimal foam generation and much less carryover of residual chemistry than the control Composition B prepared without an antifoaming agent.

10

### Example 3

A study was performed to measure the ability of an antimicrobial produce wash composition in accordance with embodiments of the present disclosure to reduce *Listeria monocytogenes*, *Escherichia coli* O157:H7 and *Salmonella enterica* in

processing waters for fruit and vegetables. The formula of the produce wash is also shown in the table below.

Table 3

Produce Wash	
Composition	Amount (weight %)
DI Water	67.95
Anionic Surfactant	1.37
Combination of Acidulants	26.32
Stabilizing Agent	3.06
Thickening Agent	0.70
Defoaming agent	0.60

5

The produce wash composition was tested against three different test systems.

Test System 1 included a *Listeria monocytogenes* mixture that included: *L.*

*monocytogenes* ATCC 49594, *L. monocytogenes* ATCC 19114, and *L. monocytogenes*

ATCC 19116. Test System 2 included a *Escherichia coli* O157:H7 mixture that

10 included *E. coli* O157:H7 ATCC 43895, *E. coli* O157:H7 ATCC 35150, and *E. coli*

O157:H7 ATCC 43890. Test System 3 included a *Salmonella enterica* mixture that

included *S. enterica* subsp. *enterica* ATCC 10721, *S. enterica* subsp. *enterica* ATCC

6962, and *S. enterica* subsp. *enterica* ATCC 13311.

For this study, the produce wash was diluted to various concentrations using

15 varying diluents, as shown in the table below.

Table 4.

Test Substance	Desired Concentration	Diluent	Test Solution (Volume of Test Substance / Total Volume)	pH
Produce Wash	1:171	405ppm Synthetic Hard Water (pH 7.90)	4.68g / 800.02g	2.82
	1:143		5.59g / 800.06g	2.74
Test Substance	Desired Concentration	Diluent	Test Solution (Volume of Test Substance / Total Volume)	pH

Produce Wash	1:256	410ppm Synthetic Hard Water (pH 7.89)	2.0g / 500g	3.07
<b>Test Substance</b>	<b>Desired Concentration</b>	<b>Diluent</b>	<b>Test Solution (Volume of Test Substance / Total Volume)</b>	<b>pH</b>
Produce Wash	1:256	200ppm Synthetic Hard Water (pH 7.89)	3.9g / 1000g	3.12
		400ppm Synthetic Hard Water (pH 7.89)		3.10
<b>Test Substance</b>	<b>Desired Concentration</b>	<b>Diluent</b>	<b>Test Solution (Volume of Test Substance / Total Volume)</b>	<b>pH</b>
Produce Wash	1:128	Sterile MilliQ Water (DI Water)	1.17g / 300g	2.77
	1:192		1.56g / 300g	2.67
	1:256		2.34g / 300g	2.55
	1:128	210ppm Synthetic Hard Water (pH 7.95)	1.17g / 300g	3.18
	1:192		1.56g / 300g	2.94
	1:256		2.34g / 300g	2.72
	1:128	420ppm Synthetic Hard Water (pH 7.91)	1.17g / 300g	3.03
	1:192		1.56g / 300g	2.87
	1:256		2.34g / 300g	2.67
<b>Test Substance</b>	<b>Desired Concentration</b>	<b>Diluent</b>	<b>Test Solution (Volume of Test Substance / Total Volume)</b>	<b>pH</b>
Produce Wash	1:200	200ppm Synthetic Hard Water (pH 7.85)	3.5g / 700g	2.95
	1:220		3.18g / 700g	2.95
	1:200	410ppm Synthetic Hard Water (pH 7.83)	3.5g / 700g	3.03
	1:220		3.18g / 700g	3.01

For this study, 99 mL of the selected test substance (produce wash dilution) was dispensed into a sterile 250 mL Erlenmeyer flask. Duplicate flasks with 99 mL of sterile phosphate buffered dilution water (PBDW) were also prepared for determination of the inoculum populations. The test flasks were vigorously swirled. While the liquid in the flask was still in motion, the tip of a pipette containing 1 mL of the selected Test System suspension was immersed in the test substance midway between the center and the edge of the flask. One (1) mL of the Test System suspension was dispensed into 99 mL of the test substance (produce wash dilution).

After the selected exposure time, 1 mL of the test substance mixture was transferred into 9mL of an inactivating agent using a sterile pipette and vortex to mix. This tube was considered to contain a  $10^{-1}$  dilution of the test solution. For the test substance use solution test samples, 1mL and 0.1 mL from the  $10^{-1}$  inactivating agent tube were plated. For the inoculum population tests, a  $10^{-5}$  and a  $10^{-7}$  dilution in PBDW were prepared. 1mL and 0.1mL from these dilutions were plated. The plates were inverted and allowed to incubate at  $35 \pm 2^\circ\text{C}$  for  $48 \pm 4$  hours. The results from this study are shown in the tables below.

Table 5. Inoculum Numbers

Test System	CFU/mL	Log <sub>10</sub> Growth	Average Log <sub>10</sub> Growth
<i>Listeria monocytogenes</i> mixture	118 x 10 <sup>6</sup> , 121 x 10 <sup>6</sup>	8.07, 8.08	8.08
<i>Escherichia coli</i> O157:H7 mixture	141 x 10 <sup>6</sup> , 146 x 10 <sup>6</sup>	8.15, 8.04	8.10
<i>Salmonella enterica</i> mixture	116 x 10 <sup>6</sup> , 102 x 10 <sup>6</sup>	8.06, 8.01	8.04

Table 6. *Listeria monocytogenes* Mixture

Test Substance	Exposure Time	Survivors (CFU/mL)	Log <sub>10</sub> Survivors	Average Log <sub>10</sub> Survivors	Log Reduction
1:171 in 400ppm hard water	90 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.08
	120 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.08
1:143 in 400ppm hard water	90 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.08
	120 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.08

Table 7. *Escherichia coli* O157:H7 Mixture

Test Substance	Exposure Time	Survivors (CFU/mL)	Log <sub>10</sub> Survivors	Average Log <sub>10</sub> Survivors	Log Reduction
1:171 in 400ppm hard water	90 sec	1 x 10 <sup>1</sup> , 4 x 10 <sup>1</sup>	1.00, 1.60	1.30	6.80
	120 sec	2 x 10 <sup>1</sup> , 1 x 10 <sup>1</sup>	1.30, 1.00	1.15	6.95
1:143 in 400ppm hard water	90 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.10
	120 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.10

\* *E. coli* O157:H7 ATCC 43895 appeared contaminated before testing, so was not included in the mixture.

Table 8. *Salmonella enterica* Mixture

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Test Substance	Exposure Time	Survivors (CFU/mL)	Log <sub>10</sub> Survivors	Average Log <sub>10</sub> Survivors	Log Reduction
1:171 in 400ppm hard water	90 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.04
	120 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.04
1:143 in 400ppm hard water	90 sec	1 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	1.00, <1.00	<1.00	>7.04
	120 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.04

Table 9. Inoculum Numbers

Test System	CFU/mL	Log <sub>10</sub> Growth	Average Log <sub>10</sub> Growth
<i>Listeria monocytogenes</i> mixture	134 x 10 <sup>6</sup> , 110 x 10 <sup>6</sup>	8.13, 8.04	8.09
<i>Escherichia coli</i> O157:H7 mixture	125 x 10 <sup>6</sup> , 127 x 10 <sup>6</sup>	8.10, 8.10	8.10
<i>Salmonella enterica</i> mixture	147 x 10 <sup>6</sup> , 148 x 10 <sup>6</sup>	8.17, 8.17	8.17

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Table 10. *Listeria monocytogenes* Mixture

Test Substance	Exposure Time	Survivors (CFU/mL)	Log <sub>10</sub> Survivors	Average Log <sub>10</sub> Survivors	Log Reduction
1:256 in 400ppm hard water	90 sec	0 x 10 <sup>1</sup>	<1.00	<1.00	>7.09
	120 sec	0 x 10 <sup>1</sup>	<1.00	<1.00	>7.09

Table 11. *Escherichia coli* O157:H7 Mixture

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Test Substance	Exposure Time	Survivors (CFU/mL)	Log <sub>10</sub> Survivors	Average Log <sub>10</sub> Survivors	Log Reduction
1:256 in 400ppm hard water	90 sec	585 x 10 <sup>3</sup>	5.77	5.77	2.33
	120 sec	176 x 10 <sup>3</sup>	5.25	5.25	2.85

Table 12. *Salmonella enterica* Mixture

Test Substance	Exposure Time	Survivors (CFU/mL)	Log <sub>10</sub> Survivors	Average Log <sub>10</sub> Survivors	Log Reduction
1:256 in 400ppm hard water	90 sec	74 x 10 <sup>3</sup>	4.87	4.87	3.30
	120 sec	306 x 10 <sup>1</sup>	3.49	3.49	4.68

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Table 13. Inoculum Numbers

Test System	CFU/mL	Log <sub>10</sub> Growth	Average Log <sub>10</sub> Growth
<i>Listeria monocytogenes</i> mixture	99 x 10 <sup>6</sup> , 105 x 10 <sup>6</sup>	7.99, 8.02	8.01
<i>Escherichia coli</i> O157:H7 mixture	92 x 10 <sup>6</sup> , 92 x 10 <sup>6</sup>	7.96, 7.96	7.96
<i>Salmonella enterica</i> mixture	104 x 10 <sup>6</sup> , 126 x 10 <sup>6</sup>	8.02, 8.10	8.06

Table 14. *Listeria monocytogenes* Mixture

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Test Substance	Exposure Time	Survivors (CFU/mL)	Log <sub>10</sub> Survivors	Average Log <sub>10</sub> Survivors	Log Reduction
1:256 in 200ppm hard water	60 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.01
	90 sec	0 x 10 <sup>1</sup> , 124 x 10 <sup>1</sup>	<1.00, 3.09	2.05	5.96
	120sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.01
	5 min	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.01
	10 min	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.01
1:256 in 400ppm hard water	5 min	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.01
	10 min	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.01

Table 15. *Escherichia coli* O157:H7 Mixture

Test Substance	Exposure Time	Survivors (CFU/mL)	Log <sub>10</sub> Survivors	Average Log <sub>10</sub> Survivors	Log Reduction
1:256 in 200ppm hard water	60 sec	328 x 10 <sup>5</sup> , 316 x 10 <sup>5</sup>	7.51, 7.50	7.51	0.45
	90 sec	194 x 10 <sup>5</sup> , 212 x 10 <sup>5</sup>	7.29, 7.33	7.31	0.65
	120sec	106 x 10 <sup>5</sup> , 124 x 10 <sup>5</sup>	7.03, 7.09	7.06	0.90
	5 min	134 x 10 <sup>3</sup> , 178 x 10 <sup>3</sup>	5.13, 5.25	5.19	2.77
	10 min	47 x 10 <sup>1</sup> , 108 x 10 <sup>1</sup>	2.67, 3.03	2.85	5.11
1:256 in 400ppm hard water	5 min	341 x 10 <sup>1</sup> , 369 x 10 <sup>1</sup>	3.53, 3.57	3.55	4.41
	10 min	2 x 10 <sup>1</sup> , 3 x 10 <sup>1</sup>	1.30, 1.48	1.39	6.57

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Table 16. *Salmonella enterica* Mixture

Test Substance	Exposure Time	Survivors (CFU/mL)	Log <sub>10</sub> Survivors	Average Log <sub>10</sub> Survivors	Log Reduction
1:256 in 200ppm hard water	60 sec	258 x 10 <sup>5</sup> , 227 x 10 <sup>5</sup>	7.41, 7.36	7.39	0.67
	90 sec	79 x 10 <sup>5</sup> , 81 x 10 <sup>5</sup>	6.90, 6.91	6.91	1.15
	120sec	24 x 10 <sup>5</sup> , 19 x 10 <sup>5</sup>	6.38, 6.28	6.33	1.73

	5 min	209 x 10 <sup>1</sup> , 25 x 10 <sup>1</sup>	3.32, 2.40	2.86	5.20
	10 min	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.06
1:256 in 400ppm hard water	5 min	0 x 10 <sup>1</sup> , 4 x 10 <sup>1</sup>	<1.00, 1.60	1.30	6.76
	10 min	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.06

Table 17. Inoculum Numbers

Test System	CFU/mL	Log <sub>10</sub> Growth	Average Log <sub>10</sub> Growth
<i>Escherichia coli</i> O157:H7 mixture	80 x 10 <sup>6</sup> , 110 x 10 <sup>6</sup>	7.90, 8.04	7.97

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Table 18. *Escherichia coli* O157:H7 Mixture

Test Substance	Exposure Time	Survivors (CFU/mL)	Log <sub>10</sub> Survivors	Average Log <sub>10</sub> Survivors	Log Reduction
1:256 in DI water	90 sec	5 x 10 <sup>1</sup> , 7 x 10 <sup>1</sup>	1.70, 1.84	1.77	6.20
1:256 in 200ppm HW		218 x 10 <sup>5</sup> , 212 x 10 <sup>5</sup>	7.34, 7.33	7.34	0.63
1:256 in 400ppm HW		46 x 10 <sup>5</sup> , 45 x 10 <sup>5</sup>	6.66, 6.65	6.66	1.31
1:192 in DI water		0 x 10 <sup>1</sup> , 1 x 10 <sup>1</sup>	<1.00, 1.00	<1.00	>6.97
1:192 in 200ppm HW		20 x 10 <sup>1</sup> , 8 x 10 <sup>1</sup>	2.30, 1.90	2.10	5.87
1:192 in 400ppm HW		40 x 10 <sup>1</sup> , 18 x 10 <sup>1</sup>	2.60, 2.26	2.43	5.54
1:128 in DI water		1 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	1.00, <1.00	<1.00	>6.97
1:128 in 200ppm HW		0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>6.97
1:128 in 400ppm HW		0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>6.97

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Table 19. Inoculum Numbers

Test System	CFU/mL	Log <sub>10</sub> Growth	Average Log <sub>10</sub> Growth
<i>Listeria monocytogenes</i> mixture	108 x 10 <sup>6</sup> , 113 x 10 <sup>6</sup>	8.03, 8.05	8.04
<i>Escherichia coli</i> O157:H7 mixture	94 x 10 <sup>6</sup> , 130 x 10 <sup>6</sup>	7.97, 8.11	8.04
<i>Salmonella enterica</i> mixture	183 x 10 <sup>6</sup> , 183 x 10 <sup>6</sup>	8.26, 8.26	8.26

Table 20. *Listeria monocytogenes* Mixture

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Test Substance	Exposure Time	Survivors (CFU/mL)	Log <sub>10</sub> Survivors	Average Log <sub>10</sub> Survivors	Log Reduction
1:200 in 200ppm hard water	90 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.04
	120 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.04
1:200 in 400ppm hard water	90 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.04
	120 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.04
1:220 in 200ppm hard water	90 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.04
	120 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.04
1:220 in 400ppm hard water	90 sec	10 x 10 <sup>1</sup> , 10 x 10 <sup>1</sup>	2.00, 2.00	2.00	6.04
	120 sec	74 x 10 <sup>1</sup> , 12 x 10 <sup>1</sup>	2.87, 2.08	2.47	5.57

Table 21. *Escherichia coli* O157:H7 Mixture

Test Substance	Exposure Time	Survivors (CFU/mL)	Log <sub>10</sub> Survivors	Average Log <sub>10</sub> Survivors	Log Reduction
1:200 in 200ppm hard water	90 sec	183 x 10 <sup>3</sup> , 72 x 10 <sup>3</sup>	5.26, 4.86	5.06	2.98
	120 sec	9 x 10 <sup>3</sup> , 4 x 10 <sup>3</sup>	3.95, 3.60	3.78	4.26
1:200 in 400ppm hard water	90 sec	58 x 10 <sup>3</sup> , 71 x 10 <sup>3</sup>	4.76, 4.85	4.81	3.24
	120 sec	4 x 10 <sup>3</sup> , 276 x 10 <sup>1</sup>	3.60, 3.44	3.52	4.52
1:220 in 200ppm hard water	90 sec	11 x 10 <sup>5</sup> , 15 x 10 <sup>5</sup>	6.04, 6.18	6.11	1.93
	120 sec	323 x 10 <sup>3</sup> , 185 x 10 <sup>5</sup>	5.51, 5.27	5.39	2.66
1:220 in 400ppm hard water	90 sec	13 x 10 <sup>5</sup> , 17 x 10 <sup>5</sup>	6.11, 6.23	6.17	1.87
	120 sec	240 x 10 <sup>3</sup> , 6 x 10 <sup>5</sup>	5.38, 5.78	5.58	2.46

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Table 22. *Salmonella enterica* Mixture

Test Substance	Exposure Time	Survivors (CFU/mL)	Log <sub>10</sub> Survivors	Average Log <sub>10</sub> Survivors	Log Reduction
1:200 in 200ppm hard water	90 sec	1 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	1.00, <1.00	<1.00	>7.26
	120 sec	0 x 10 <sup>1</sup> , 6 x 10 <sup>1</sup>	<1.00, 1.78	1.39	6.87
1:200 in 400ppm hard water	90 sec	1 x 10 <sup>1</sup> , 5 x 10 <sup>1</sup>	1.00, 1.70	1.35	6.91
	120 sec	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>7.26
1:220 in 200ppm hard water	90 sec	230 x 10 <sup>1</sup> , 178 x 10 <sup>1</sup>	3.36, 3.25	3.31	4.96
	120 sec	1 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	1.00, <1.00	<1.00	7.26
1:220 in 400ppm hard water	90 sec	15 x 10 <sup>3</sup> , 7 x 10 <sup>3</sup>	4.18, 3.84	4.01	4.25
	120 sec	31 x 10 <sup>1</sup> , 40 x 10 <sup>1</sup>	2.49, 2.60	2.55	5.72

The results are also summarized in the tables below.

5 Table 23. *Listeria monocytogenes*

Produce Wash in 200ppm Hard Water Log Reduction					
Dilution	60 sec	90 sec	120 sec	5 min	10 min
1:256	>7.01	5.96	>7.01	>7.01	>7.01
1:220		>7.04	>7.04		
1:200		>7.04	>7.04		
1:128					
Produce Wash in 400ppm Hard Water Log Reduction					
Dilution	90 sec	120 sec	5 min	10 min	
1:256	>7.09	>7.09	>7.01	>7.01	
1:220	6.04	5.57			
1:200	>7.04	>7.04			
1:171	>7.08	>7.08			
1:143	>7.08	>7.08			
1:128					

10 Table 24. *Escherichia coli* O157:H7

Produce Wash in DI Water Log Reduction	
Dilution	90 sec
1:256	6.20
1:192	>6.97
1:128	>6.97

Produce Wash in 200ppm Hard Water Log Reduction					
Dilution	60 sec	90 sec	120 sec	5 min	10 min
1:256	0.45	0.65/0.63	0.90	2.77	5.11
1:220		1.93	2.66		
1:200		2.98	4.26		
1:192		5.54/5.87			
1:128		>6.97			
Produce Wash in 400ppm Hard Water Log Reduction					
Dilution	90 sec	120 sec	5 min	10 min	
1:256	2.33/1.31	2.85	4.41	6.57	
1:220	1.87	2.46			
1:200	3.24	4.52			
1:192	5.54				
1:171	6.80	6.95			
1:143	>7.10	>7.10			
1:128	>6.97				

Table 25. *Salmonella enterica*

Produce Wash in 200ppm Hard Water Log Reduction					
Dilution	60 sec	90 sec	120 sec	5 min	10 min
1:256	0.67	1.15	1.73	5.20	>7.06
1:220		4.96	7.26		
1:200		>7.26	6.87		
1:128					
Produce Wash in 400ppm Hard Water Log Reduction					
Dilution	90 sec	120 sec	5 min	10 min	
1:256	3.30	4.68	6.76	>7.06	
1:220	4.25	5.72			
1:200	6.91	>7.26			
1:171	>7.04	>7.04			
1:143	>7.04	>7.04			
1:128					

5

As can be seen from these results, the exemplary formulations tested achieved significant antimicrobial performance at multiple concentrations in the wash water solution. Antimicrobial reductions achieved will reduce cross contamination that could occur during the washing of produce and maintain a sterile wash basin for additional

food processing. In some applications in which extended contact times are allowed, the exemplary formulations tested showed expanded antimicrobial performance over tested performance intervals at the full range of concentrations tested.

5 *Example 4.*

A study was performed to evaluate the residual efficacy of an exemplary produce wash composition in accordance with embodiments of the present disclosure to reduce *Listeria monocytogenes*, *Escherichia coli* O157:H7 and *Salmonella enterica* on the surface of spinach leaves. The following produce wash composition was used  
 10 for this study:

Table 26

Produce Wash	
Composition	Amount (weight %)
DI Water	67.95
Anionic Surfactant	1.37
Combination of Acidulants	26.32
Stabilizing Agent	3.06
Thickening Agent	0.70
Defoaming agent	0.60

For this study, pre-washed spinach leaves were purchased from a local grocery store. The leaves were sorted, and those that were broken or looked damaged were  
 15 discarded. Five leaves per treatment and control were picked. The leaves were placed spine side down on a rack. Each leaf was inoculated with 8-10 spots, equaling a total of 100  $\mu$ L of prepared  $10^{-8}$  Colony forming units (CFU)/mL of Test System culture. The following Test Systems were used: Test System 1 included a *Listeria monocytogenes* mixture that included *L. monocytogenes* ATCC 49594, *L. monocytogenes* ATCC 19114,

and *L. monocytogenes* ATCC 19116. Test System 2 included an *Escherichia coli* O157:H7 mixture that included *E. coli* O157:H7 ATCC 43895, *E. coli* O157:H7 ATCC 35150, and *E. coli* O157:H7 ATCC 43890.

After the leaves were inoculated, they were dried at room temperature for 45 minutes, or until the inoculum was visibly dry. Then, 300 mL of the test substance (diluted produce wash) was dispensed into a 600 mL beaker. The test substance dilutions used in this study are shown below.

Table 27.

Desired Concentration	Diluent	Test Solution (Volume of Test Substance / Total Volume)	pH
0.75oz/gal	380ppm Synthetic Hard Water (pH 7.71)	10.4g / 1951.4g	n/a
Desired Concentration	Diluent	Test Solution (Volume of Test Substance / Total Volume)	pH
0.75oz/gal	400ppm Synthetic Hard Water (pH 7.71)	10.4g / 2000.0g	2.98

Samples were exposed to the bulk test substance for 90 seconds then removed and allowed a dwell time equal to sample times listed in table 29 below, viz., pre-exposure, immediately post exposure, 2 minutes post-exposure, 5 minutes post-exposure, 10 minutes post-exposure, 15 minutes post-exposure, and 30 minutes post-exposure. At the end of the desired evaluation time, extended activity profile was measured post neutralization vs. an equivalent water control at each time point. Extended activity is a measurement of cumulative kill independent of reduction observed in bulk solution.

The results from this study are shown in the tables below.

Table 28. Inoculum Numbers

Test System	CFU/mL	Log <sub>10</sub> Growth	Average Log <sub>10</sub> Growth
<i>Listeria monocytogenes</i> mixture	73 x 10 <sup>6</sup> , 81 x 10 <sup>6</sup>	7.86, 7.91	7.89

Table 29. *Listeria monocytogenes* Mixture

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Sample Time	Survivors (CFU/Surface)	Log <sub>10</sub> Growth	Avg. Log <sub>10</sub> Growth	Log Reduction
Pre-exposure	1.00 x 10 <sup>6</sup> , 2.20 x 10 <sup>6</sup> , 1.40 x 10 <sup>6</sup> , 8.00 x 10 <sup>5</sup> , 2.40 x 10 <sup>6</sup>	6.00, 6.34, 6.15, 5.90, 6.38	6.15	N/A
Immediately Post-exposure	1.88 x 10 <sup>3</sup> , 2.40 x 10 <sup>2</sup> , 1.60 x 10 <sup>2</sup> , <2.00 x 10 <sup>1</sup> , 4.20 x 10 <sup>2</sup>	3.27, 2.38, 2.20, <1.30, 2.62	2.34	3.81
2 minutes Post-exposure	8.80 x 10 <sup>2</sup> , 2.32 x 10 <sup>3</sup> , 4.00 x 10 <sup>1</sup> , 2.20 x 10 <sup>2</sup> , 8.00 x 10 <sup>1</sup>	2.94, 3.37, 1.60, 2.34, 1.90	2.43	3.72
5 minutes Post-exposure	8.00 x 10 <sup>1</sup> , 8.00 x 10 <sup>1</sup> , 1.20 x 10 <sup>4</sup> , 1.60 x 10 <sup>2</sup> , 4.00 x 10 <sup>1</sup>	1.90, 1.90, 4.08, 2.20, 1.60	2.34	3.81
10 minutes Post-exposure	4.40 x 10 <sup>4</sup> , 8.60 x 10 <sup>2</sup> , <2.00 x 10 <sup>1</sup> , 3.62 x 10 <sup>3</sup> , <2.00 x 10 <sup>1</sup>	4.64, 2.93, <1.30, 2.56, <1.30	2.75	3.40
15 minutes Post-exposure	<2.00 x 10 <sup>1</sup> , <2.00 x 10 <sup>1</sup> , 3.00 x 10 <sup>2</sup> , <2.00 x 10 <sup>1</sup> , <2.00 x 10 <sup>1</sup>	<1.30, <1.30, 2.48, <1.30, <1.30	1.54	4.61
30 minutes Post-exposure	2.00 x 10 <sup>2</sup> , 1.00 x 10 <sup>2</sup> , <2.00 x 10 <sup>1</sup> , 2.00 x 10 <sup>1</sup> , <2.00 x 10 <sup>1</sup>	2.30, 2.00, <1.30, 1.30, <1.30	1.64	4.51

Table 30. Inoculum Numbers

Test System	CFU/mL	Log <sub>10</sub> Growth	Average Log <sub>10</sub> Growth
<i>Escherichia coli</i> O157:H7 mixture	176 x 10 <sup>6</sup> , 201 x 10 <sup>6</sup>	8.25, 8.30	8.27

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Table 31. *Escherichia coli* O157:H7 Mixture

Sample Time	Survivors (CFU/Surface)	Log <sub>10</sub> Growth	Avg. Log <sub>10</sub> Growth	Log Reduction
Pre-exposure	1.62 x 10 <sup>5</sup> , 1.34 x 10 <sup>5</sup> , 1.04 x 10 <sup>5</sup> , 1.48 x 10 <sup>5</sup> , 1.88 x 10 <sup>5</sup>	5.21, 5.13, 5.02, 5.17, 5.27	5.16	N/A
Immediately Post-exposure	6.00 x 10 <sup>3</sup> , 2.00 x 10 <sup>3</sup> , 2.00 x 10 <sup>3</sup> , 5.00 x 10 <sup>4</sup> , 4.82 x 10 <sup>3</sup>	3.78, 3.30, 3.30, 4.70, 3.68	3.75	1.41
2 minutes Post-exposure	1.80 x 10 <sup>4</sup> , 1.40 x 10 <sup>4</sup> , 4.00 x 10 <sup>3</sup> , 1.20 x 10 <sup>4</sup> , 1.60 x 10 <sup>4</sup>	4.25, 4.15, 3.60, 4.08, 4.20	4.06	1.10
5 minutes Post-exposure	6.00 x 10 <sup>3</sup> , 1.04 x 10 <sup>3</sup> , 9.60 x 10 <sup>2</sup> , 1.20 x 10 <sup>3</sup> , 7.20 x 10 <sup>2</sup>	3.78, 3.02, 2.98, 3.08, 2.86	3.14	2.02

10 minutes Post-exposure	$1.00 \times 10^2$ , $<2.00 \times 10^1$ , $2.04 \times 10^3$ , $4.80 \times 10^2$ , $8.00 \times 10^1$	2.00, $<1.30$ , 3.31, 2.68, 1.90	2.24	2.92
15 minutes Post-exposure	$<2.00 \times 10^1$ , $8.00 \times 10^1$ , $1.28 \times 10^3$ , $<2.00 \times 10^1$ , $4.00 \times 10^1$	$<1.30$ , 1.90, 3.11, 1.30, 1.60	1.84	3.32
30 minutes Post-exposure	$2.00 \times 10^1$ , $4.00 \times 10^1$ , $<2.00 \times 10^1$ , $8.80 \times 10^2$ , $3.00 \times 10^2$	1.30, 1.60, $<1.30$ , 2.94, 2.48	1.92	3.24

As can be seen from these results, the exemplary produce wash composition carried over on the surface of the treated materials achieved expanded kill performance for at least 45 minutes post bulk solution exposure. This expanded kill on the surface of produce enhances food safety benefits to the formulation and falls within many field applications.

#### Example 6 –

A study was performed to compare the effect of different acids on exemplary antimicrobial produce wash compositions. For this study, lactic acid, and sodium acid sulfate (SAS) were used. The produce wash compositions tested are shown in the table below.

Table 32.

Formula A (Lactic Acid)		Formula B (SAS)	
Raw Material	Amt (g)	Raw Material	Amt (g)
Anionic Surfactant	0.64	Anionic Surfactant	0.64
Acidulant	0.50	Acidulant	1.50
Defoaming Agent	0.25	Defoaming Agent	0.25
Lactic Acid, 88%	8.92	Sodium Bisulfate	2.65
Thickener	0.75	Thickener	0.75
Stabilizing Agent	2.57	Stabilizing Agent	2.57
Grapefruit Oil	0.05	Grapefruit Oil	0.05
DI Water	86.32	DI Water	91.59
Total	100.00	Total	100.00

pH after dilution	3.12	pH after dilution	3.04
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Both Formula A and Formula B were diluted at 1:64 ratio in 205 ppm synthetic hard water, and were tested against *Escherichia coli* ATCC 11229 at 1 and 2 minute exposure times. For this study, 99 mL of the selected test substance (Formula A or B diluted) was dispensed into a sterile 250 mL Erlenmeyer flask. Duplicate flasks with 99 mL of sterile phosphate buffered dilution water (PBDW) were also prepared for determination of the inoculum populations. The test flasks were vigorously swirled. While the liquid in the flask was still in motion, the tip of a pipette containing 1 mL of the selected Test System suspension was immersed in the test substance midway between the center and the edge of the flask. One (1) mL of the Test System suspension was dispensed into 99 mL of the test substance (Formula A or B diluted). There was also a 10% vegetable soil in the hard water.

After the selected exposure time, 1 mL of the test substance mixture was transferred into 9 mL of an inactivating agent using a sterile pipette and vortex to mix. This tube was considered to contain a  $10^{-1}$  dilution of the test solution. For the test substance use solution test samples, 1 mL and 0.1 mL from the  $10^{-1}$  inactivating agent tube were plated. For the inoculum population tests, a  $10^{-5}$  and a  $10^{-7}$  dilution in PBDW were prepared. 1 mL and 0.1 mL from these dilutions were plated. The plates were inverted and allowed to incubate at  $35 \pm 2^\circ\text{C}$  for  $48 \pm 4$  hours. The results from this study are shown in the tables below.

Table 33. Inoculum Numbers

Test System	CFU/mL	Log <sub>10</sub> Growth	Average Log <sub>10</sub> Growth
<i>Escherichia coli</i> ATCC 11229	$78 \times 10^6$	7.89	7.89

	77 x 10 <sup>6</sup>	7.89	
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Table 34. *Escherichia coli* ATCC 11229

Test Substance	Exposure Time	Survivors (CFU/mL)	Log <sub>10</sub> Growth	Average Log <sub>10</sub> Growth	Log Reduction
Formula A (Lactic Acid)	1 min	110 x 10 <sup>1</sup> , 44 x 10 <sup>1</sup>	3.04, 2.64	2.84	5.05
	2 min	0 x 10 <sup>1</sup> , 0 x 10 <sup>1</sup>	<1.00, <1.00	<1.00	>6.89
Formula B (SAS)	1 min	68 x 10 <sup>3</sup> , 66 x 10 <sup>3</sup>	4.83, 4.82	4.82	3.07
	2 min	123 x 10 <sup>1</sup> , 29 x 10 <sup>3</sup>	3.09, 4.46	3.78	4.11

5 As can be seen from these results, Formula A which contained lactic acid had about a 5 log reduction after only 1 minute. Formula B which contained SAS only had about a 3 log reduction after 1 minute. Thus, Formula A passed with greater than the recommended 4 log reduction after 1 minute and 2 minutes, against *E. coli*. Formula B (the SAS produce wash) only passed with greater than a 4 log reduction after 2 minutes, 10 against *E. coli*. Overall, it was found that the lactic acid produce wash performed better than the SAS produce wash.

The foregoing summary, detailed description, and examples provide a sound basis for understanding the disclosure, and some specific example embodiments of the disclosure. Since the disclosure can comprise a variety of embodiments, the above 15 information is not intended to be limiting. The invention resides in the claims.

## CLAIMS

1. A single phase aqueous low foaming antimicrobial concentrate composition comprising:
  - (a) at least about 25 wt% water;
  - 5 (b) an antimicrobial agent comprising an anionic surfactant;
  - (c) a defoaming agent;
  - (d) an acidulant;
  - (e) a stabilizing agent; and
  - (f) a thickening agent,
- 10 wherein the composition has a viscosity of between about 50 centipoise and about 3500 centipoise, and the thickening agent is present in an amount effective such that the composition is phase stable under acidic conditions.
2. The composition of claim 1, wherein the anionic surfactant is selected from the group consisting of alkyl sulfates, alkyl sulfonates, alkyl aryl sulfonates, sulfonated
- 15 fatty acids, sulfonated fatty acid esters, sulfonated carboxylic acid esters, and mixtures thereof.
3. The composition of claim 1, wherein the composition is substantially free of a C5 to C11 carboxylic acid.
4. The composition of claim 1, wherein the defoaming agent comprises a silicone
- 20 defoaming agent.
5. The composition of claim 1, wherein the thickening agent is selected from the group consisting of natural polysaccharide or cellulose thickeners, plant exudates,

seaweed extracts, water soluble polymers, poly acrylic acid based thickeners, polyacrylamide based thickeners, inorganic clay based thickeners, and mixtures thereof.

6. The composition of claim 1, wherein the acidulant comprises a food additive ingredient.

5 7. The composition of claim 1, wherein the acidulant is selected from the group consisting of citric acid, sodium bisulfate, acetic acid, adipic acid, tartaric acid, propionic acid, malic acid, lactic acid, sulfuric acid, and derivatives and mixtures thereof.

8. The composition of claim 1, wherein the composition comprises:  
10 between about 0.01 wt% and about 50 wt% of the antimicrobial agent;  
between about 0.01 wt% and about 10.0 wt% defoaming agent;  
between about 0.1 wt% and about 10 wt% thickening agent; and  
between about 1 wt% to about 50 wt% of the acidulant

9. The composition of claim 1, wherein the composition has a pH of less than  
15 about 3.2.

10. The composition of claim 1, wherein the composition further comprises an additional ingredient selected from the group consisting of a surfactant, a processing aid, a dye, a colorant, an odorant, and mixtures thereof.

11. The composition of claim 1, wherein the composition is substantially free of a  
20 chlorite, and oxidizing agent, and mixtures thereof.

12. The composition of claim 1, wherein the composition comprises between about 0.1 wt% and about 5.0 wt% stabilizing agent.

13. The composition of claim 1, wherein the stabilizing agent is present at an amount effective to prevent the formation of a precipitate for at least 24 hours when the composition is diluted with 500 ppm hard water.

14. The composition of claim 1, wherein the stabilizing agent comprises a block  
5 copolymer of ethylene oxide and propylene oxide.

15. The composition of claim 14, wherein the copolymers have a molecular weight of about 5,000 to about 10,000 and the percentage of ethylene oxide is between about 70 to about 90.

16. A method for reducing a population of microorganisms on a surface comprising:

10 (a) providing a single phase aqueous low foaming antimicrobial concentrate composition comprising:

(i) at least about 25 wt% water;

(ii) an antimicrobial agent comprising an anionic surfactant;

(iii) a defoaming agent;

15 (iv) an acidulant selected from the group consisting of sulfates, sulfonates, and combinations thereof;

(v) a stabilizing agent; and

(vi) a thickening agent,

wherein the composition has a viscosity of between about 50 centipoise and  
20 about 3500 centipoise, and the thickening agent is present in an amount effective such that the composition is phase stable under acidic conditions;

(b) diluting the composition; and

(c) contacting the surface with the diluted composition such that the population of microorganisms is reduced, wherein the composition does not need to be rinsed from the surface.

17. The method of claim 16, wherein the anionic surfactant is selected from the group consisting of alkyl sulfates, alkyl sulfonates, alkyl aryl sulfonates, sulfonated fatty acids, sulfonated fatty acid esters, sulfonated carboxylic acid esters, and mixtures thereof.

18. The method of claim 16, wherein the defoaming agent comprises a silicone defoaming agent.

19. The method of claim 16, wherein the thickening agent is selected from the group consisting of natural polysaccharide or cellulose thickeners, plant exudates, seaweed extracts, water soluble polymers, poly acrylic acid based thickeners, polyacrylamide based thickeners, inorganic clay based thickeners, and mixtures thereof.

20. The method of claim 16, wherein the acidulant comprises a food additive ingredient.

21. The method of claim 16, wherein the acidulant is selected from the group consisting of lactic acid, citric acid, sodium bisulfate, acetic acid, adipic acid, tartaric acid, propionic acid, malic acid, lactic acid, sulfuric acid, and derivatives and mixtures thereof.

22. The method of claim 16, wherein the composition comprises:

between about 0.01 wt% and about 50 wt% of the antimicrobial agent;

between about 0.01 wt% and about 10.0 wt% defoaming agent;

between about 0.1 wt% and about 10 wt% thickening agent; and

between about 1 wt% to about 50 wt% of the acidulant..

23. The method of claim 16, wherein the composition is diluted at about a 1:64 to about a 1:128 ratio.

24. The method of claim 16, wherein the composition is phase stable for at least  
5 about one year at a temperature of about 25°C.

25. The method of claim 16, wherein the microorganism is selected from the group consisting of spores, bacteria, mold, yeast, viruses, and combinations thereof.

26. The method of claim 16, wherein the step of contacting reduces the population of the microorganism by at least about 4 log reduction after 1 minute.

10 27. The method of claim 16, wherein the surface comprises produce selected from the group consisting of fruits, vegetables and mixtures thereof.

28. The method of claim 16, wherein the surface comprises a food contact surface.

29. The method of claim 28, wherein the food contact surface comprises a food processing tool selected from the group consisting of utensils, trays, strainers, cutting  
15 boards, dishware, pots, pans, and combinations thereof.

30. A method for washing produce comprising:

(a) contacting the produce with a phase stable aqueous acidic antimicrobial composition comprising:

- (i) at least about 25wt% water;
- 20 (ii) an antimicrobial agent comprising an anionic surfactant;
- (iii) a defoaming agent;
- (iv) an acidulant;
- (v) a stabilizing agent; and

(v) a thickening agent present in an amount effective such that the composition is phase stable under acidic conditions,

wherein the composition is diluted prior to contacting and has a viscosity of between about 50 centipoise and about 3500 centipoise, and the produce does not need  
5 to be rinsed after being contacted by the composition.

31. The method of claim 30, wherein the composition does not substantially affect the organoleptic properties of the produce after contacting the produce.

32. The method of claim 30, wherein the step of contacting comprises placing the produce in an agitated bath containing a water source and the composition.

10 33. The method of claim 30, wherein the step of contacting comprises diluting the composition through an aspirator and spraying it on to a surface of the produce.

34. The method of claim 30, wherein after the step of contacting the composition remains on the surface of the produce for between about 30 to about 60 minutes prior to drying.

15 35. The method of claim 34, wherein the composition reduces the population of the microorganism on the surface by about 4 log after remaining on the surface for between about 30 and about 60 minutes.

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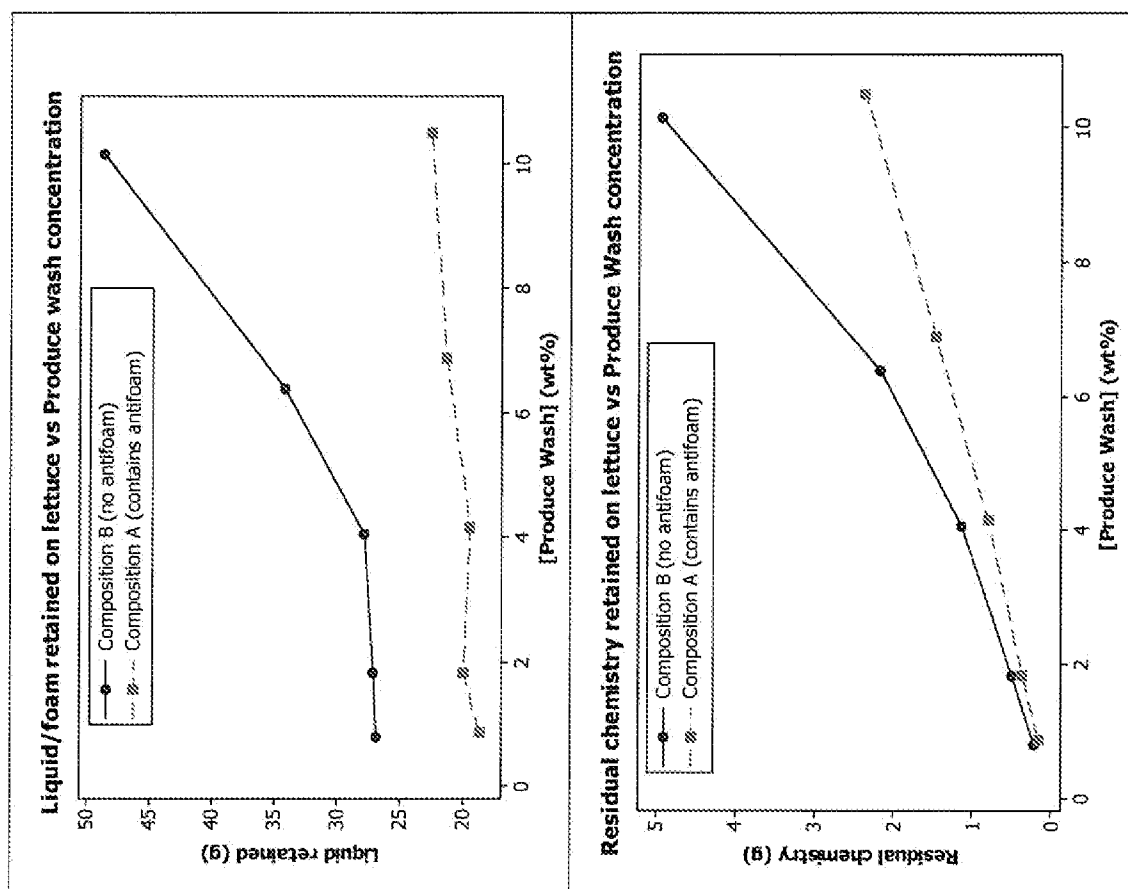


Figure 1