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(54) Title: COMPOUNDS WITH ONE OR MORE FUNCTIONAL GROUPS AND USE THEREOF IN LIQUID DISINFECTANTS

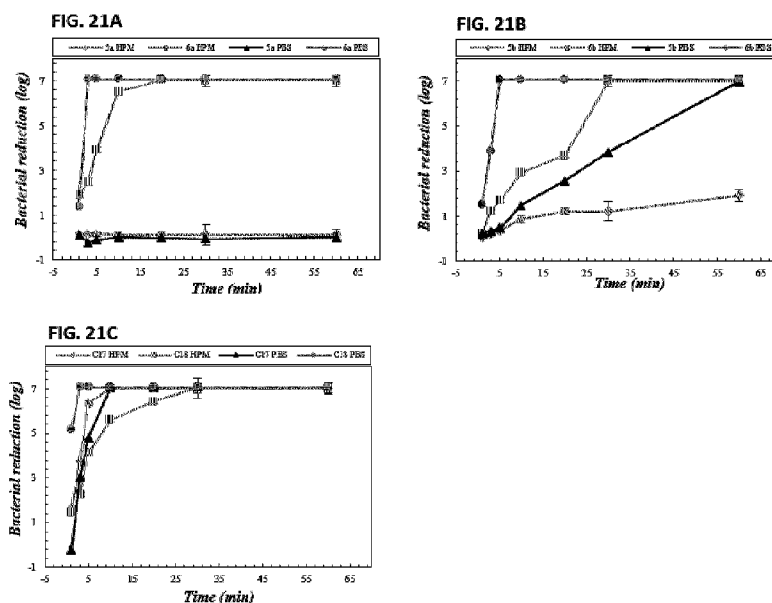


FIG. 21

(57) Abstract: Embodiments of the present disclosure relate to compounds that have one, two or more function groups, where the functional groups may be selected from a group consisting of a piperidine, a hydantoin, a quaternary ammonium cation or combinations thereof. The compounds may have biocidal activity and the compounds may subsequently be chemically modified to enhance or provide biocidal activity. The chemical modification may be performed in situ and repeated once or multiple times to extend the time-frame over which the compounds have the desired biocidal activity. The functional groups may be physically separated from one another by other atoms within the compound and this physical separation may provide a desired compound- stability and influence the compound's biocidal activity. The compounds disclosed herein may form part or all of a liquid disinfectant.



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COMPOUNDS WITH ONE OR MORE FUNCTIONAL GROUPS AND USE THEREOF IN LIQUID DISINFECTANTS

TECHNICAL FIELD

The present disclosure relates to the field of biocidal compounds and precursors thereof. In particular, the present disclosure relates to biocidal compounds, and precursors thereof, that can be used in a liquid application, such as a liquid disinfectant.

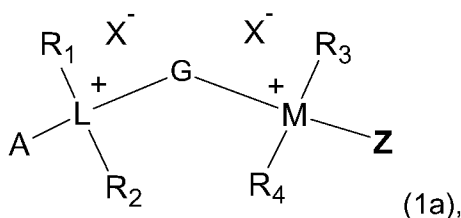
BACKGROUND

Microbial resistance to biocidal compounds poses a large and growing threat to human health during the 21st century. Various circumstances and applications call for the use of liquid disinfectants that include biocidal compounds. Currently, there are different broad-spectrum biocidal compounds extensively employed during disinfection applications, including: silver, hydrogen peroxide, nitrogen oxide, sodium hypochlorite, quaternary ammonium compounds (QAC) and *N*-halamine compounds.

It is known that the combination of QACs with *N*-halamine provides a greater biocidal activity when compared to the activity of these components alone. However, *N*-halamine compounds may have a range of different stabilities in environments that comprise organic content, for example biological fluids and other protein-based contaminants. This stability range may be based on the dissociation constant of each *N*-halamine compound which permits the chloronium ion to transfer to other amines within the organic content. The loss of the chloronium ion can cause the *N*-halamine compound to lose some or all of its biocidal activity. For example, organic content within an environment with may quench the functionality of the *N*-halamine. This instability presents a challenge to using *N-halamine* compounds in various applications where it is desirable to kill microbes within an environment with organic content. The organic content can also decrease the biocidal activity of QACs through other mechanisms.

SUMMARY

Embodiments of the present disclosure comprise compounds that are selected from a group of compounds that have the general formula (**Formula 1a**):



wherein L is nil or nitrogen,

when L is nil:

both R_1 and R_2 are nil,

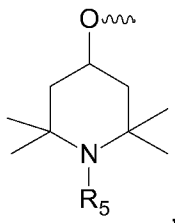
M is nitrogen or phosphorous,

when M is nitrogen, R_3 and R_4 are each $C_nH_{(2n+1)}$,

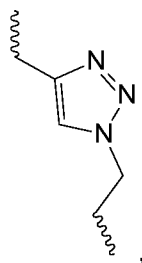
n is either 1 or 2,

when M is phosphorous, R_3 and R_4 are both one of methyl, ethyl and phenyl,

A is:



G is:



Z is $C_{na}H_{(2na+1)}$,

where $na = 12$ to 24

and R_5 is one of hydrogen, chlorine, bromine, iodine and CH_3CO ;

when L is nitrogen:

M is nitrogen or phosphorous,

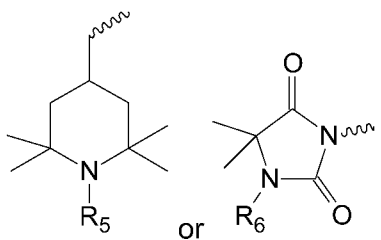
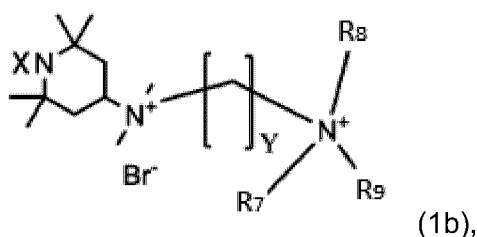
when M is nitrogen, each of R_1, R_2, R_3 and R_4 are $C_{nb}H_{(2nb+1)}$,

nb is one of 1 or 2,

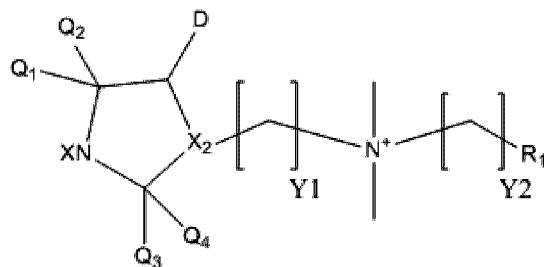
when M is phosphorous, R_3 and R_4 are both one of methyl, ethyl and phenyl,

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A is:

G is $-(CH_2)_{nc}-$ and where nc is 0 or an integer between 1 and 12Z is $C_{nd}H_{(2nd+1)}$, nd is an integer between 1 and 16, R_5 and R_6 are each one of hydrogen, chlorine, bromine, iodine and CH_3CO ; and X^- is an anionic counter-charge selected from the group of Cl^- , Br^- and PO_4^{3-} , when X^- is PO_4^{3-} the anionic counter-charge balances with respect to the cation.Other embodiments of the present disclosure comprise compounds that are selected a group of compounds that have the general formula (**Formula 1b**):wherein R_9 is $C_{ne}H_{2ne+1}$ and n is an integer between 10 and 20 or R_4 is $C_pH_{2p}NH_2$ and p is an integer between 1 and 10;wherein R_7 and R_8 are each independently one of CH_3 or CH_2CH_3 ;wherein Y is an integer between 4 and 20; andwherein X is one of hydrogen, chlorine, bromine or iodine.Embodiments of the present disclosure comprise compounds that are selected from a group of compounds that have the general formula (**Formula 2**):wherein C is one of $C_3H_6(COOH)_2$, $C_4H_8(COOH)_2$, $C_5H_{10}(COOH)_2$, $C_6H_{12}(COOH)_2$ or combinations thereof; andwherein D is V_3W^{3-} ,

wherein V is



wherein when X_2 is C then Q_1 , Q_2 , Q_3 and Q_4 are all H_2 and D is H_2 or when X_2 is N then Q_1 and Q_2 are each CH_3 , Q_3 and Q_4 are collectively $=O$ and D is $=O$;

wherein Y1 and Y2 are each independently an integer between 2 and 12;

wherein W is PO_4 , SO_4 , HPO_4 or W^{3-} is $3F^-$, $3Cl^-$ or $3Br^-$;

wherein R1 is CH_3 or $=CH_2$; and

wherein X is one of hydrogen, chlorine, bromine or iodine.

Some further embodiments of the present disclosure relate to the use of the above compounds as biocides that are delivered in a liquid state and may be used in environments with or without a relative-protein content. In some embodiments of the present disclosure, the compounds have biocidal activity or the potential for increased biocidal activity. In some embodiments of the present disclosure, the compounds may be modified by chemically connecting, attaching or bonding a halogen moiety to the biocide compound to provide the biocidal activity or to increase the biocidal activity.

Embodiments of the present disclosure may be dissolved, either completely or partially, into aqueous-based solutions. The compound or a combination of compounds disclosed herein may be added into the aqueous-based solution in a non-halogenated form and then halogenated within the aqueous-based solution. Optionally, the compound or compounds may be added into the aqueous-based solution in an already halogenated form and the compound or compounds may be halogenated again or repeatedly to extend the time-frame over which the compound has biocidal activity while the aqueous-based solution is being used in an application.

Further embodiments of the present disclosure relate to the use of the above compounds in combination with one or more potentiator compounds that alter the biocidal activity of the above compounds. Examples of potentiator compounds may include an inorganic compound, a surfactant or combinations thereof.

Further embodiments of the present disclosure relate to a liquid biocide with a formulation that comprises: one or more of the above compounds in combination with one or more potentiator compounds, or not, and further additive ingredients.

BRIEF DESCRIPTION OF THE DRAWINGS

5 These and other features of the present disclosure will become more apparent in the following detailed description in which reference is made to the appended drawings, wherein:

FIG. 1 is a schematic representation of an example of a chemical reaction scheme for synthesizing compounds according to an embodiment of the present disclosure;

FIG. 2 is another schematic representation of an example of a chemical reaction scheme for synthesizing compounds according to another embodiment of the present disclosure;

10 **FIG. 3** is another schematic representation of an example of a chemical reaction scheme for synthesizing compounds according to another embodiment of the present disclosure, wherein FIG. 3A shows a synthesis reaction and FIG. 3B shows a halogenation reaction;

FIG. 4 is another schematic representation of an example of a chemical reaction scheme for synthesizing compounds according to another embodiment of the present disclosure;

15 **FIG. 5** is another schematic representation of an example of a chemical reaction scheme for synthesizing compounds according to another embodiment of the present disclosure, wherein FIG. 5A shows a chemical reaction scheme for producing a compound with a six-carbon chain between cationic centers and FIG. 5B shows a chemical reaction scheme for producing a compound with a four-carbon chain between cationic centers;

20 **FIG. 6** is a line graph showing an example of experimental bacterial killing data that compares some of the embodiments of the present disclosure in 0 % fetal bovine serum (FBS);

FIG. 7 is a line graph showing an example of experimental bacterial killing data that compares some of the embodiments of the present disclosure in 5 % FBS;

25 **FIG. 8** is a line graph showing an example of experimental bacterial killing data that compares some of the embodiments of the present disclosure in 20 % FBS;

FIG. 9 is a line graph showing an example of experimental bacterial killing data that compares some of the embodiments of the present disclosure before and after *in situ* halogenation;

FIG. 10 is a line graph showing an example of experimental bacterial killing data that compares some of the embodiments of the present disclosure in Mueller Hinton broth;

FIG. 11 is a line graph showing an example of experimental bacterial killing data that compares some of the embodiments of the present disclosure in 0 % FBS;

FIG. 12 is a line graph showing an example of experimental bacterial killing data that compares some of the embodiments of the present disclosure in 5 % FBS;

FIG. 13 is a line graph showing an example of experimental bacterial killing data that compares some of the embodiments of the present disclosure in 20 % FBS;

FIG. 14 is NMR data that demonstrates the chemical composition of one of the compounds disclosed herein;

FIG. 15 is NMR data that demonstrates the chemical composition of another compound disclosed herein;

FIG. 16 is another schematic representation of an example of a chemical reaction scheme for synthesizing compounds according to another embodiment of the present disclosure;

FIG. 17 is another schematic representation of an example of a chemical reaction for synthesizing compounds according to another embodiment of the present disclosure;

FIG. 18 is two bar graphs that each show an example of experimental bacterial killing data that compares some embodiment of the present disclosure, wherein FIG. 18A shows a comparison of different potentiator ingredients; and FIG. 18B shows a comparison different ratios of one potentiator to a compound of the present disclosure;

FIG. 19 is another schematic representation of an example of a chemical reaction scheme for synthesizing compounds according to another embodiment of the present disclosure.

FIG. 20 is a schematic representation of an exemplary chemical reaction for synthesizing an exemplary biocidal compound according to an embodiment of the present disclosure;

FIG. 21 shows three line-graphs that each depict biocidal-activity data of compounds according to embodiments of the present disclosure in high protein media or phosphate buffered saline, wherein FIG. 21A shows exemplary data obtained from experiments with a first bacterial specie and two compounds; FIG. 21B shows exemplary data obtained from experiments with the first bacterial specie and two other compounds; and FIG. 21C shows exemplary data obtained from experiments with the first bacterial specie and two further compounds;

FIG. 22 shows three line-graphs that each depict biocidal-activity data of compounds according to embodiments of the present disclosure in high protein media or phosphate buffered saline, wherein FIG. 22A shows exemplary data obtained from experiments with a second bacterial specie and two

compounds; FIG. 22B shows exemplary data obtained from experiments with the second bacterial specie and two other compounds; and FIG. 22C shows exemplary data obtained from experiments with the second bacterial specie and two further compounds;

5 **FIG. 23** shows three line-graphs that each depict biocidal-activity data of compounds according to embodiments of the present disclosure in high protein media or phosphate buffered saline, wherein FIG. 23A shows exemplary data obtained from experiments with a third bacterial specie and two compounds; FIG. 23B shows exemplary data obtained from experiments with the third bacterial specie and two other compounds; and FIG. 23C shows exemplary data obtained from experiments with the third bacterial specie and two further compounds; and

10 **FIG. 24** is a line graph that depicts the stability of C18 (amide based *N-halamine*), 6a and 6b (amine based *N-halamines*) in the presence of 5% FBS (Fetal bovine serum) over a 120-minutes time-course.

DETAILED DESCRIPTION

15 Embodiments of the present disclosure relate to compounds that have one, two or more function groups, where the functional groups may be selected from a group consisting of an *N*-halamine precursor, a piperidine, a hydantoin, a cationic center with an ammonium or phosphonium or combinations thereof. The compounds may have biocidal activity and the compounds may subsequently be chemically modified to enhance or provide biocidal activity. The chemical modification may be performed *in situ* and repeated once or multiple times to extend the time-frame over which the
20 compounds have the desired biocidal activity. The functional groups may be physically separated from one another by other atoms within the compound and this physical separation may provide a desired compound-stability and influence the compound's biocidal activity.

25 Embodiments of the present disclosure relate to compounds that have the potential for biocidal activity or that have actual biocidal activity and that biocidal activity will persist in a liquid state in an environment with various degrees of relative protein content, such as a low-relative protein content, a medium-relative protein content or a high-relative protein content. Relative-protein content may also be referred to herein as organic load. The degree of the relative-protein content may be based upon various factors within the environment, such as temperature, pH, the concentration or total mass of the
30 protein(s), the types of proteins and/or the inhibitory effect of the protein(s) within the environment. Without being bound by any particular theory, compounds that comprise multiple functional groups or moieties may have different relative stabilities in the liquid state when in the presence of proteins or other inhibitory organic content. These relative stabilities may be based on the dissociation constant of the functional groups or moieties within the compounds. For example, the stability of an *N*-halamine

with an amine may be more stable than an *N*-halamines with an amide. An *N*-halamine with an amide may be more stable than an *N*-halamine with an imide.

Definitions

5 Unless defined otherwise, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this disclosure belongs.

As used herein, the term "about" refers to an approximately +/-10% variation from a given value. It is to be understood that such a variation is always included in any given value provided herein, whether or not it is specifically referred to.

10 As used herein, the term "activity" refers to biocidal activity that kills, inhibits the growth of or otherwise renders a microbe harmless.

The terms "biocide" as used herein means a chemical compound, a chemical composition or a chemical formulation, such as a disinfectant, that has biocidal activity and can kill or render harmless one or more microbes.

15 The terms "halo" or "halogen" by themselves or as part of another substituent, as used herein, have the same meaning as commonly understood by one of ordinary skill in the art, and refer to chlorine, bromine or iodine.

The term "liquid" as used herein means an incompressible fluid that may be in the form of a bulk phase, a surface phase, a spray, a droplet, a micro droplet or a nano droplet.

20 As used herein, the terms "microbe" and "microbes" refer to one or more single celled, or multi-cellular, microorganisms exemplified by at least one of bacterium, archaea, yeast or fungi.

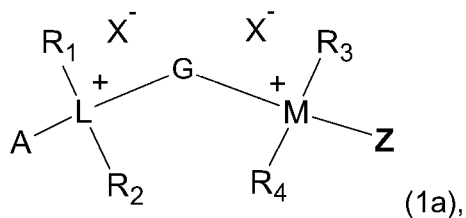
25 The term "*N*-halamine" as used herein refers to a compound containing one or more nitrogen-halogen covalent bonds that is normally formed by the halogenation of imide, amide or amine groups of a compound. The presence of the halogen on an *N*-halamine moiety may render the compound biocidal or enhance the compound's biocidal activity. *N*-halamines, as referred to in the present disclosure, include both cyclic, acyclic *N*-halamine compounds and precursors to *N*-halamine compounds.

30 The terms "quaternary ammonium cation", "quaternary ammonium compound", "quaternary ammonium salt", "QAC", and "quat" may be used interchangeably throughout the present disclosure to refer to ammonium compounds in which four organic groups are linked to a nitrogen atom that produces a positively charged ion (cation) of the structure NR_4^+ .

Embodiments of the present disclosure will now be described by reference to the figures, FIG. 1 to FIG. 24. The figures show examples of reactions that may be used to synthesize various

embodiments of the present disclosure. The figures also show examples of experimental data that demonstrate biocidal activity and the potential thereof of embodiments of the present disclosure.

Embodiments of the present disclosure comprise compounds that are selected from a group of compounds that have the general formula (**Formula 1a**):



wherein L is nil or nitrogen,

when L is nil:

both R₁ and R₂ are nil,

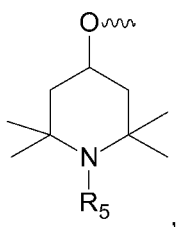
M is nitrogen or phosphorous,

when M is nitrogen, R₃ and R₄ are each C_nH_(2n+1),

n is either 1 or 2,

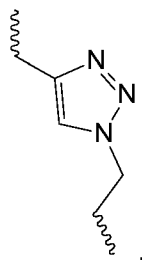
when M is phosphorous, R₃ and R₄ are both one of methyl, ethyl and phenyl,

A is:



G is:

10



Z is $C_{na}H_{(2na+1)}$,

where $na = 12$ to 24

and R_5 is one of hydrogen, chlorine, bromine, iodine and CH_3CO ;

5

when L is nitrogen:

M is nitrogen or phosphorous,

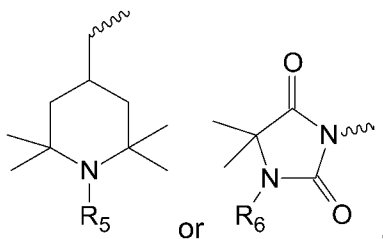
when M is nitrogen, each of R_1, R_2, R_3 and R_4 are $C_{nb}H_{(2nb+1)}$,

nb is one of 1 or 2,

when M is phosphorous, R_3 and R_4 are both one of methyl, ethyl and phenyl,

10

A is:



G is $-(CH_2)_{nc}-$ and where nc is 0 or an integer between 1 and 12

Z is $C_{nd}H_{(2nd+1)}$,

nd is an integer between 1 and 16,

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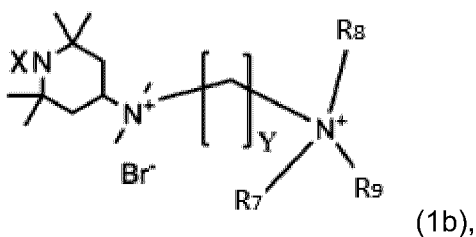
R_5 and R_6 are each one of hydrogen, chlorine, bromine, iodine and CH_3CO ; and

X^- is an anionic counter-charge selected from the group of Cl^- , Br^- and PO_4^{3-} , when X^- is PO_4^{3-} the anionic counter-charge balances with respect to the cation.

Other embodiments of the present disclosure comprise compounds that are selected a group of compounds that have the general formular (**Formula 1b**):

20

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wherein R9 is $C_{ne}H_{2ne+1}$ and n is an integer between 10 and 20 or R4 is $C_pH_{2p}NH_2$ and p is an integer between 1 and 10;

wherein R7 and R8 are each independently one of CH_3 or CH_2CH_3 ;

wherein Y is an integer between 4 and 20; and

wherein X is one of hydrogen, chlorine, bromine or iodine.

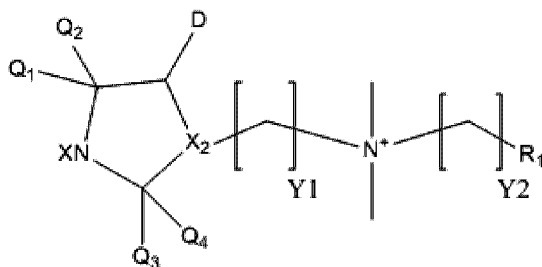
Other embodiments of the present disclosure comprise compounds that are selected from a group of compounds that have the general formula (**Formula 2**):



wherein C is one of $C_3H_6(COOH)_2$, $C_4H_8(COOH)_2$, $C_5H_{10}(COOH)_2$, $C_6H_{12}(COOH)_2$ or combinations thereof; and

wherein D is V_3W^{3-} ,

wherein V is



wherein when X2 is C then Q1, Q2, Q3 and Q4 are all H2 and D is H2 or when X2 is N then Q1 and Q2 are each CH3, Q3 and Q4 are collectively =O and D is =O;

wherein Y1 and Y2 are each independently an integer between 2 and 12;

wherein W is PO_4 , SO_4 , HPO_4 or W^{3-} is $3F^-$, $3Cl^-$ or $3Br^-$;

wherein R1 is CH_3 or $=CH_2$; and

wherein X is one of hydrogen, chlorine, bromine or iodine.

The above compounds can be delivered in a liquid state while demonstrating biocidal activity or the potential for increased biocidal activity. For example the above compounds may be introduced as a liquid such as an aqueous-based liquid or a non-aqueous based liquid.

Some embodiments of the present disclosure relate to the compounds described herein forming all of or part of a liquid disinfectant that has biocidal activity or the potential for increased biocidal activity.

Some embodiments of the present disclosure relate to the compounds described herein forming all or part of a liquid disinfectant and the compounds have the potential for biocidal activity. These embodiments with the potential for biocidal activity may be chemically modified to provide the biocidal activity. Optionally, the compounds described herein that have the potential for biocidal activity may be chemically modified once or multiple times to provide the biocidal activity.

Some embodiments of the present disclosure relate to a liquid disinfectant formulation that comprises one or more of compounds described herein, wherein the disinfectant formulation has biocidal activity or the potential for biocidal activity.

Some embodiments of the present disclosure relate to a formula for a liquid disinfectant that comprises one or more of the compounds described herein and a potentiator compound. The potentiator compound may enhance the biocidal activity of the liquid disinfectant either following a chemical modification of the liquid disinfectant or not. In one embodiment of the present disclosure the potentiator compound is an inorganic compound, for example ammonium chloride. In another embodiment of the present disclosure the potentiator compound is a surfactant. Other embodiments of the present disclosure relate to a formula for a liquid disinfectant that comprises one or more of the compounds described herein and one or more potentiator compounds.

Some embodiments of the present disclosure relate to a liquid disinfectant that comprises one or more of the compounds that have biocidal activity and/or the potential for increased biocidal activity. The liquid disinfectants may be used for cleaning, disinfecting or for controlling microbe levels in at least the following applications or products: animal housing facilities including at least veterinarian or agricultural facilities; transportation equipment; horticultural facilities; aqua-culture facilities; laboratory equipment; water-based building materials including at least paint, joint cements, spackling and grouting, adhesives and polymer emulsions; water processing facilities where the liquid disinfectant may be recirculated and will not necessarily be disposed of following disinfection including at least non-potable fountain solutions, recirculating water cooling towers, closed loop water cooling systems and industrial baths; pulp and paper processing facilities including at least pulp slurries, paper mill process waters, pigments, filler slurries and mineral slurries; industrial or manufacturing facilities including at least metal or ceramic-working fluids, process waters, cooling fluids and hydraulic fluids;

5 air washer systems; oil and/or gas well injection fluids; oil & gas transport facilities; oil and gas storage facilities; brewery pasteurizing facilities; food and beverage canning facilities; industrial and consumer cleaning and disinfection for at least residential homes, hospitals, institutions, commercial facilities, commercial and consumer product preservation; hard surface disinfection in food and beverage
10 processing facilities including at least utensils, walls and floors in poultry and animal dressing plants, offal rooms, exterior walls and loading platforms of dressing plants, preparation surfaces and mechanized preparation equipment, processing, transportation and storage equipment and surfaces; food preservation including at least a fresh-produce wash; preservation of household or commercial cleaning products for at least liquid detergents, soaps, fabric softeners, floor care products and
15 cleaning wipes; and preservation of personal hygiene and care products including at least shampoo, hair conditioners, soap, body wash, moisturizing lotion, sunscreen, cosmetics and cleaning wipes.

Some embodiments of the present disclosure relate to a liquid disinfectant that comprises one or more of the compounds described herein and that has biocidal activity and/or the potential for
20 increased biocidal activity. The liquid disinfectant may be useful for cleaning, disinfecting or for controlling microbe levels in at least the following agricultural applications: swine farrowing units; nurseries, finisher houses; processing plants, agricultural equipment; personal agricultural equipment such as rubber boots; poultry hatchers; setters; evaporative coolers; humidifying systems; ceiling fans; chicken coops; trays and plastic chick boxes; hard and non-porous surfaces and equipment used in
25 veterinary facilities; stables; foot dips; kennels; horse boxes; feed rooms; tack; feedlot facilities; quarantine pens; cages; animal transportation vehicles and holding facilities; irrigation tanks and lines; fogging operations; facilities for commercial dairy operations including milk extraction and storage equipment and all non-porous surfaces; and equipment, instruments and utensils employed in animal husbandry.

Some embodiments of the present disclosure relate to a liquid disinfectant that comprises one or more of the biocide compounds described herein and that has biocidal activity and/or the potential
30 for increased biocidal activity. The liquid disinfectant may be useful for cleaning, disinfecting or for controlling microbe levels in at least the following emergency disease control and animal biosecurity applications: agricultural facilities; agricultural equipment including trucks; tractors; wheels; livestock trailers; harvesters; loading equipment; slaughter equipment; quarantine facilities; foot dips; shipping facilities; personal protective equipment such as rubber boots, protective suits, respirator and air
35 filtering equipment; transport facilities such as rail, airport, port and related equipment; military facilities and equipment; non-porous surfaces and equipment used in veterinary facilities; stables; kennels; horse boxes feed rooms; tack; feedlot facilities; quarantine pens; cages; animal transportation vehicles; irrigation tanks and lines; fogging operations and all non-porous surfaces; and equipment, instruments and utensils employed in animal husbandry.

5 Some embodiments of the present disclosure relate to a liquid disinfectant that comprises one or more of the biocide compounds described herein and that has biocidal activity and/or the potential for increased biocidal activity. The liquid disinfectant may be useful for cleaning, disinfecting or for controlling microbe levels in at least the following horticultural biocide applications: indoor horticultural facilities and structures such as non-porous surfaces including glasshouse structures, walls, floors, storage rooms; vehicles within horticultural structures; equipment; containers; trays; water systems; ventilation systems and evaporative coolers; irrigation lines and tanks before introduction or reintroduction of any soil, seeds or plants. Alternatively, soil, plants and seeds may be covered or moved to another location to avoid or reduce contact between the liquid disinfectant and the soil, plants or seeds.

15 Some embodiments of the present disclosure relate to a liquid disinfectant that comprises one or more of the biocide compounds described herein and that has biocidal activity and/or the potential for increased biocidal activity. The liquid disinfectant may be useful for cleaning, disinfecting or for controlling microbe levels in at least the following aqua-culture biocide applications: aquaculture facility surfaces and equipment including at least hoses, wetsuits, hip waders, boots, nets, tanks, testing equipment and vehicles.

Examples

Example 1 – Synthesis Reaction A

20 FIG. 1 shows one example of a set of reactions for synthesizing some compounds according to embodiments of the present disclosure. This set of reactions comprises at least 2 steps, labelled as A and B respectively in FIG. 1.

25 Step A comprises a first step of mixing together the compounds identified in FIG. 1 as Compound 1 and Compound 2, respectively. The next step is to permit the mixed compounds to react for about 4.5 to about 5.5 hours at about 50 °C to produce the compound identified as Compound 3 in FIG. 1.

Step B comprises the step of adding either of the compounds identified in FIG. 1 as Compound 4a and Compound 4b, respectively to produce the compounds identified in FIG 1 as Compound 5a and Compound 5b, respectively.

Example 2 –Synthesis Reaction B

30 FIG. 2 shows another example of a set of reactions for synthesizing additional compounds according to embodiments of the present disclosure. This set of reactions comprises at least one step, labelled as C in FIG. 1.

Step C comprises mixing together the compounds identified in FIG. 2 as Compound 6 and Compound 7, respectively to produce the compound identified in FIG. 2 as Compound 8.

Example 3 – Synthesis Reaction C

FIG. 3A shows another example of a reaction for synthesizing additional compounds according to embodiments of the present disclosure. This reaction comprises mixing together Compound 6 with one of three precursor compounds, identified as Compound 9, 10 and 11 in FIG. 3, to produce three respective compounds identified in FIG. 3 as Compound 12, Compound 13 and Compound 14.

Example 4 – Halogenation Reaction D

FIG.3B shows an example of a halogenation reaction for synthesizing additional compounds according to embodiments of the present disclosure. This reaction comprises reacting the reaction products of FIG.3 further reacted with ^tBuOCl in an acetone and water solution (4:1 v/v) for 24 hours at room temperature. This reaction produced three respective further compounds identified in FIG.4 as Compound 15, Compound 16 and Compound 17. Optionally, halogens other than chlorine may be added to either of Compounds 15, 16 and 17 for example by using a different halogen-based reactant with the acetone and water solution.

Example 5 – Synthesis Reaction E

FIG. 4 shows another example of a reaction for synthesizing additional compounds according to embodiments of the present disclosure. The reaction comprises reacting Compounds 18 and 22 with acetonitrile (ACN) under reflux conditions for about an hour. Then Compound 20 is added and the resultant solvent is extracted under oil and then under vacuum to produce the example Compound 21. The purity of Compound 21 is shown in the NMR data shown in FIG. 14.

Example 6 – Synthesis Reaction F and F (i)

FIG. 5A shows another example of a reaction for synthesizing additional compounds according to embodiments of the present disclosure. The reaction comprises reacting Compounds 18 and 22 with ACN under reflux conditions for about an hour. Then Compound 20 is added and the resultant solvent is extracted under oil and then under vacuum to produce the example Compound 23.

FIG. 5B shows another example of a reaction for synthesizing additional compounds according to embodiments of the present disclosure. The reaction comprises reacting Compounds 18(i) and 22 with ACN under reflux conditions for about an hour. Then Compound 20 is added and the resultant solvent is extracted under oil and then under vacuum to produce the example Compound 23(i).

Example 7 – Synthesis Reaction G

FIG. 16 shows another example of a reaction (Synthesis Reaction G) for synthesizing additional compounds according to embodiments of the present disclosure. The reaction comprises reacting Compound 24 (3.0 eq) and Compound 25 (1.0 eq) to produce the example Compound 26.

Example 8 – Synthesis Reaction H

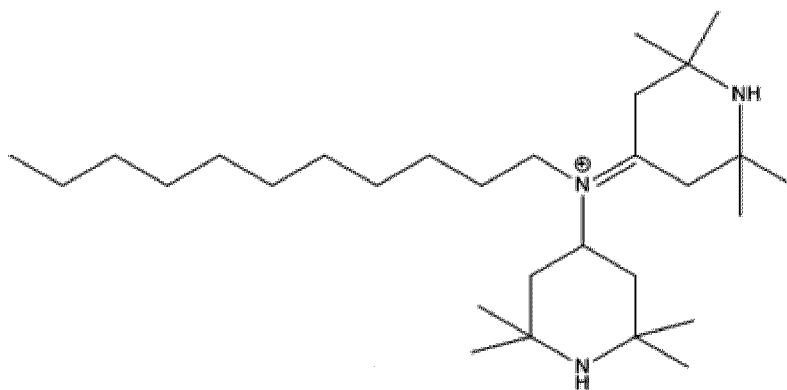
FIG. 17 shows another example of a reaction (Synthesis Reaction H) for synthesizing additional compounds according to embodiments of the present disclosure. The reaction comprises reacting Compound 27 with Compound 25 to produce the example Compound 28.

Example 9 – Synthesis Reaction I

FIG. 19 shows another example of a reaction (Synthesis Reaction I) for synthesizing additional compounds according to embodiments of the present disclosure.

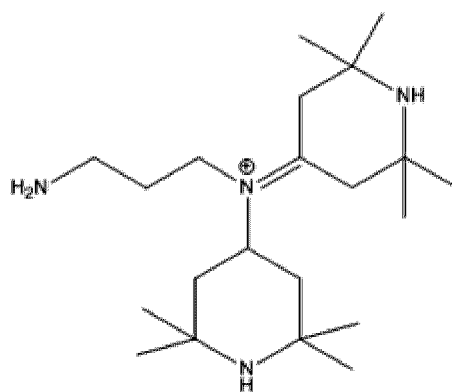
Example 10 – Compounds

An embodiment of the present disclosure is referred to in FIG. 1 as Compound 5a and it has the following general formula (**Formula 3**):



(3).

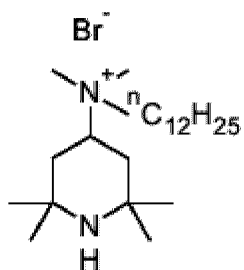
Another embodiment of the present disclosure is referred to in FIG. 1 as Compound 5b and it has the following general formula (**Formula 4**):



(4).

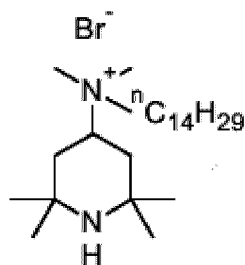
Another embodiment of the present disclosure is referred to as Compound 12 in FIG. 3A and it has the following general formula (**Formula 5**):

17



(5).

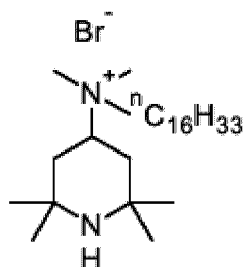
Another embodiment of the present disclosure is referred to as Compound 13 in FIG. 3A and it has the following general formula (**Formula 6**):



(6).

5

Another embodiment of the present disclosure is referred to as Compound 14 in FIG. 3A and it has the following general formula (**Formula 7**):

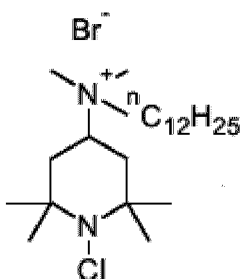


(7).

10

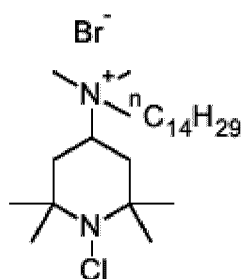
Another embodiment of the present disclosure may be synthesized according to one of the reactions shown in FIG. 3B, this embodiment is a compound that is referred to as Compound 15 in FIG. 3B and it has the following general formula (**Formula 8**):

18



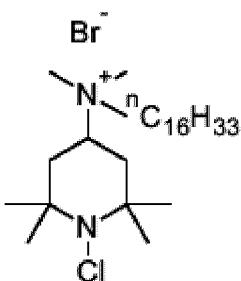
(8).

Another embodiment of the present disclosure may be synthesized according to one of the reactions shown in FIG. 3B, this embodiment is a compound that is referred to as Compound 16 in FIG. 3B and it has the following general formula (**Formula 9**):



(9).

Another embodiment of the present disclosure may be synthesized according to one of the reactions shown in FIG. 3B, this embodiment is a compound that is referred to as Compound 17 in FIG. 3B and it has the following general formula (**Formula 10**):



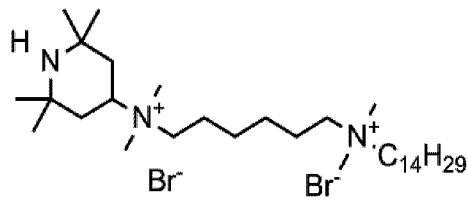
(10).

Another embodiment of the present disclosure may be synthesized according to a reaction similar to that shown in FIG. 4, and it has the following general formula (**Formula 11**):

5

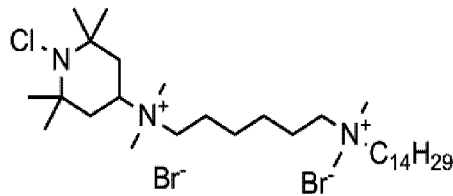
10

19



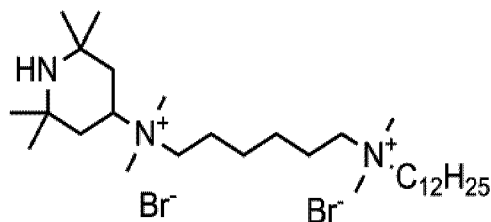
(11).

Another embodiment of the present disclosure may be synthesized according to a reaction similar to that shown in FIG. 4, and then halogenating that reaction product, for example, by a reaction similar to the reaction shown in FIG. 3B. This embodiment is a compound that has the following general formula (**Formula 12**):

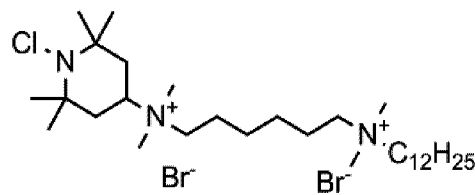


(12).

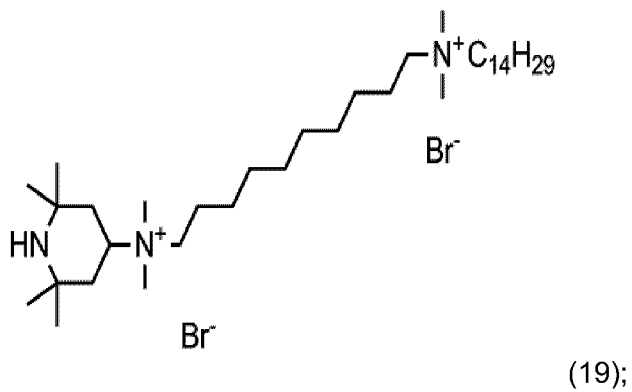
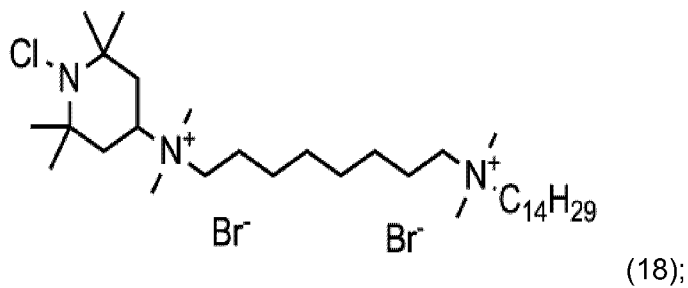
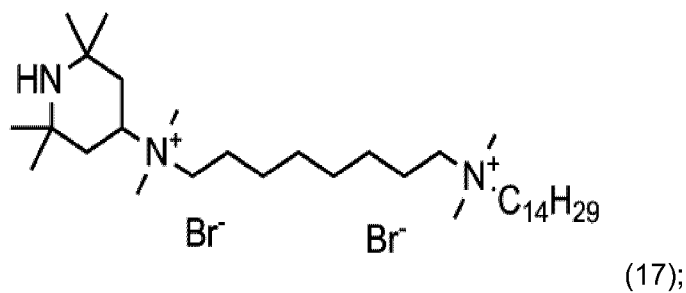
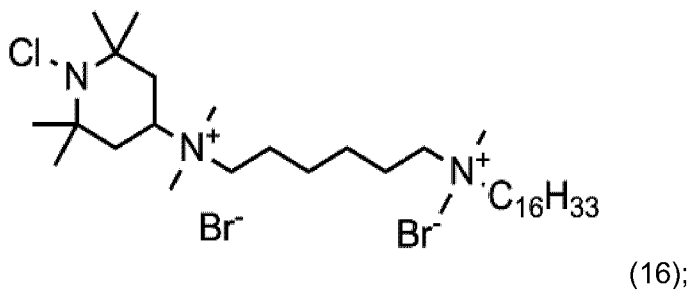
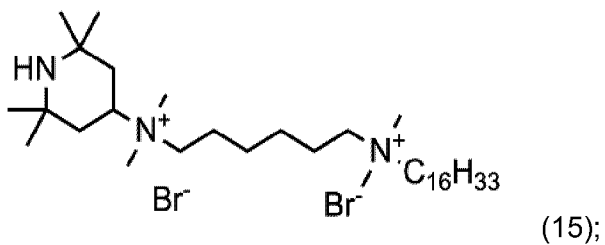
Other embodiments of the present disclosure may be synthesized according to a similar reaction as the reaction shown in FIG. 4 and then optionally chlorinating that reaction product for example by a reaction similar to the reaction shown in FIG. 3B. These embodiments may each be a compound that has the following general formulae:

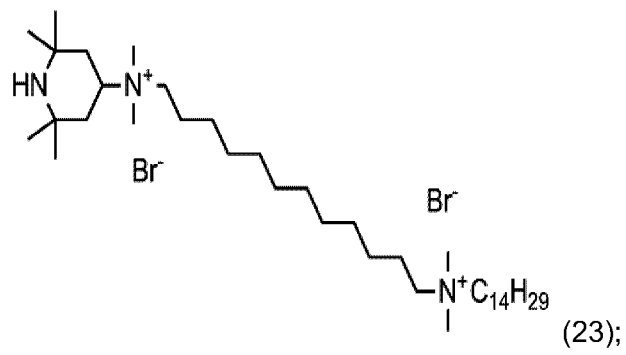
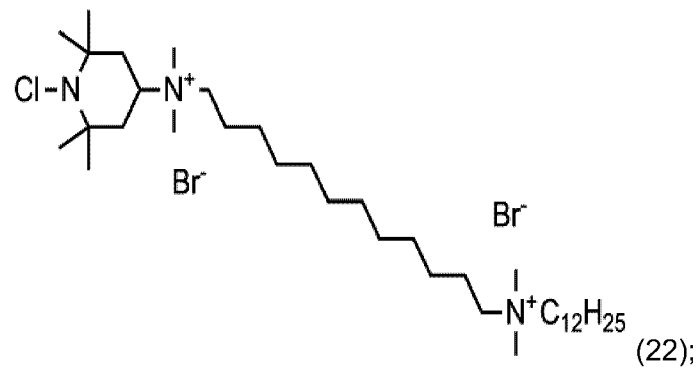
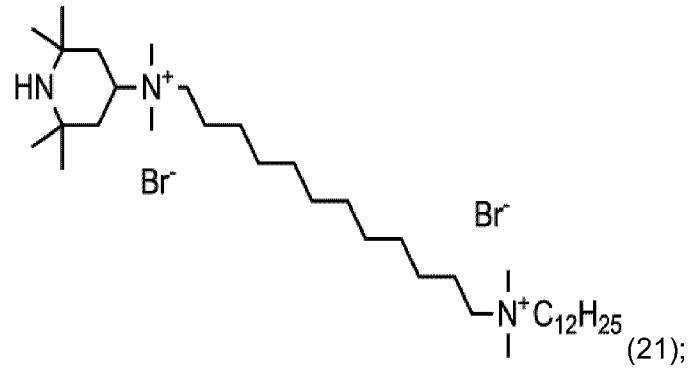
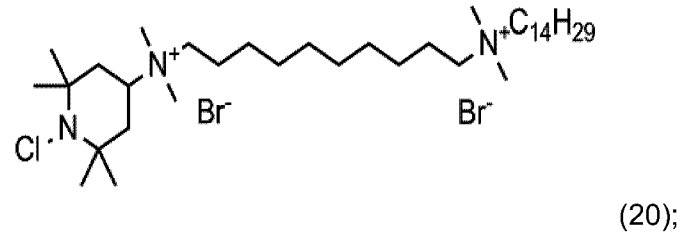


(13);

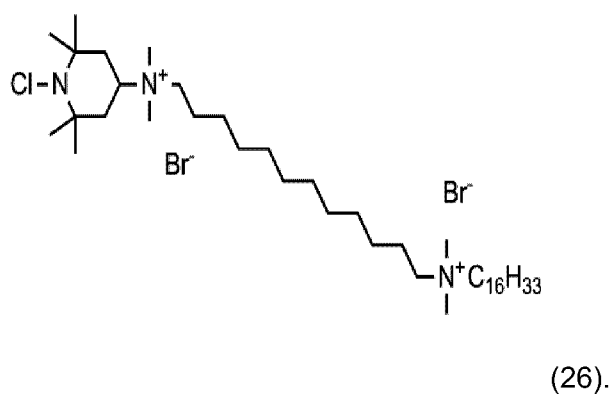
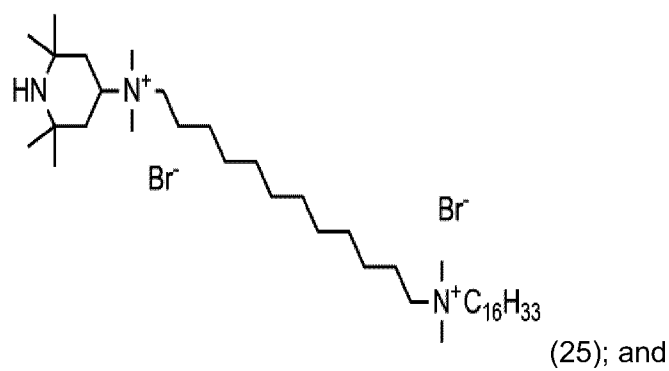
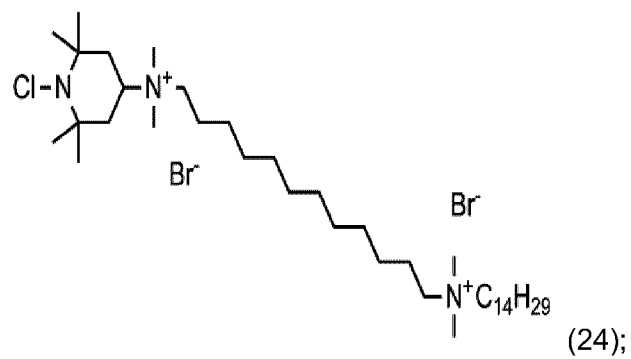


(14);



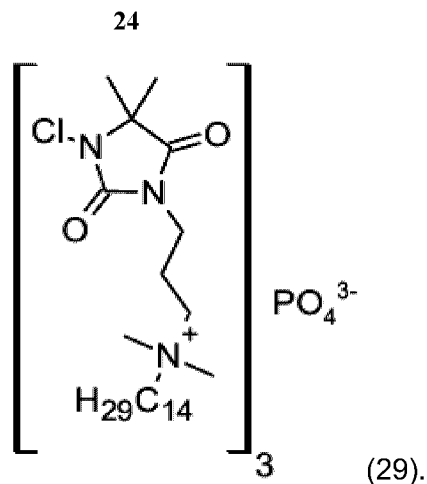


22



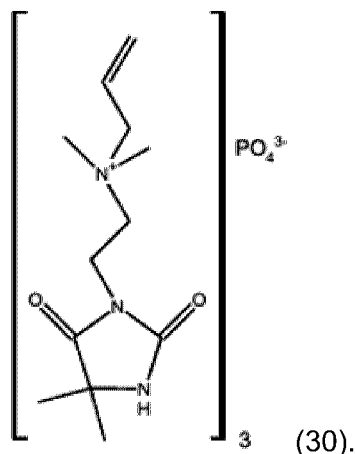
5

Another embodiment of the present disclosure may be synthesized according to the reaction shown in FIG. 5, this embodiment is a compound that is referred to in FIG. 5 as Compound 23 and it has the following general formula (**Formula 27**):



5 Optionally, a different counter-ion may be used in place of the phosphate ion (PO_4^{3-}). For example, a sulphate ion (SO_4^{2-}), a monohydrogen phosphate ion (HPO_4^{2-}), a fluoride ion (F^-), a chloride ion (Cl^-), a bromide ion (Br^-) or a nitrate ion (NO_3^-).

Another embodiment of the present disclosure may be synthesized according to the reaction shown in FIG. 17. One embodiment of the present disclosure is a compound that is referred to in FIG. 17 as Compound 28 and it has the following general formula (**Formula 30**):

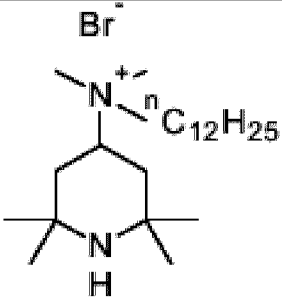
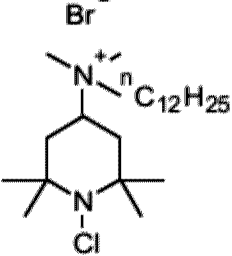
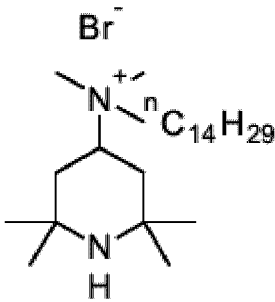
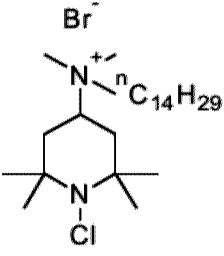


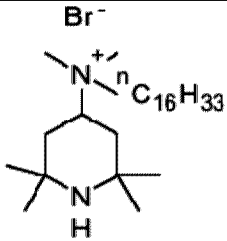
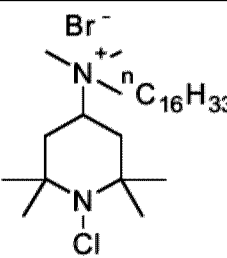
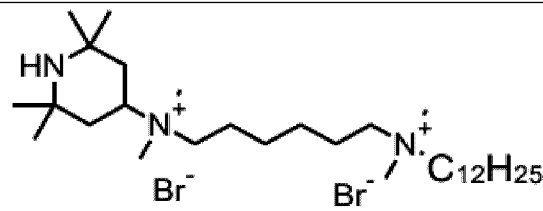
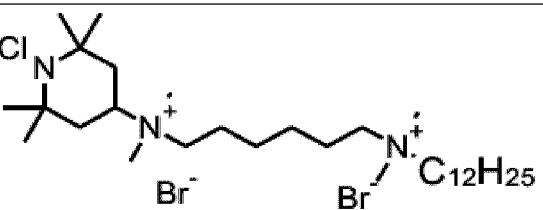
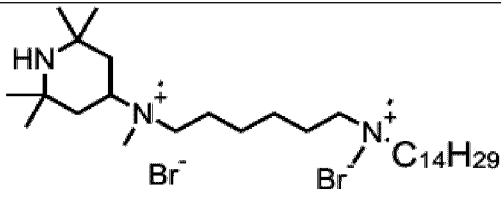
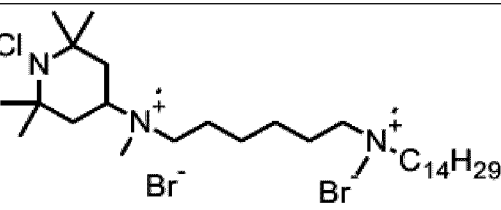
10 Optionally, a counterion may be used to synthesis any of the embodiments of the present disclosure that have the Formula 28, 29 or 30. Examples of a counterion include $\text{C}_3\text{H}_6(\text{COOH})_2$, $\text{C}_4\text{H}_8(\text{COOH})_2$, $\text{C}_5\text{H}_{10}(\text{COOH})_2$, $\text{C}_6\text{H}_{12}(\text{COOH})_2$, $\text{HOOC}(\text{CH}_2)_3\text{COOH}$, $\text{HOOC}(\text{CH}_2)_4\text{COOH}$, $\text{HOOC}(\text{CH}_2)_5\text{COOH}$, $\text{HOOC}(\text{CH}_2)_6\text{COOH}$ or combinations thereof.

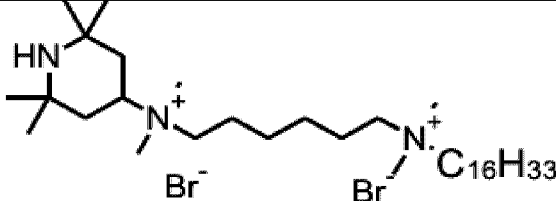
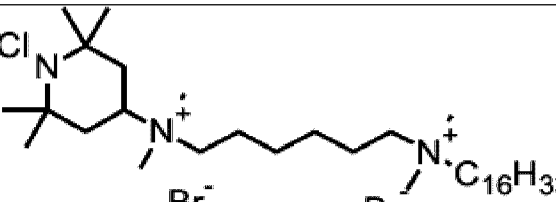
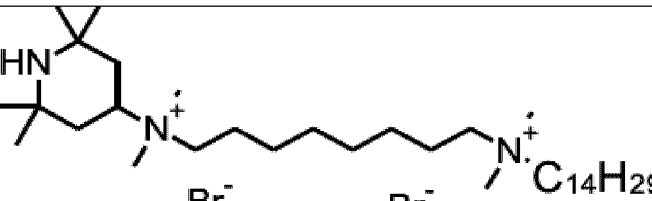
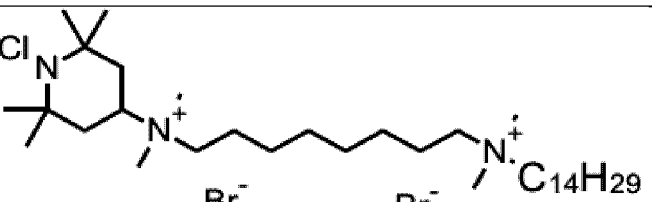

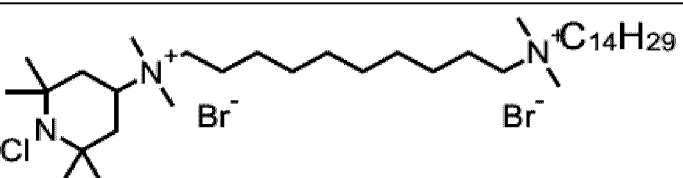
15 The compounds with the general formulae 1 to 30 may be useful in liquid-based biocidal agents, such as a liquid disinfectant, for use in environments with a low, medium or high protein content.

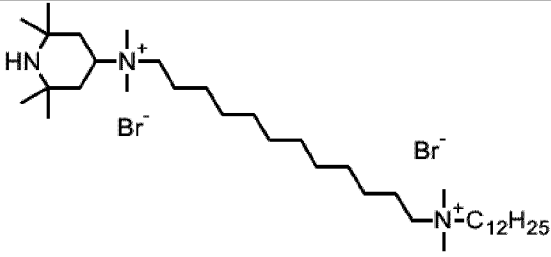
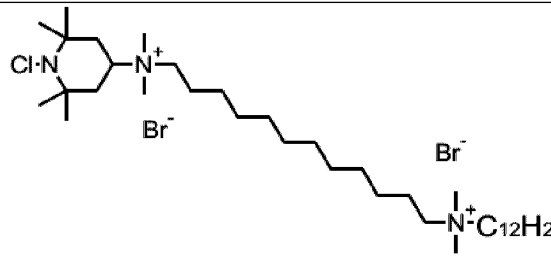
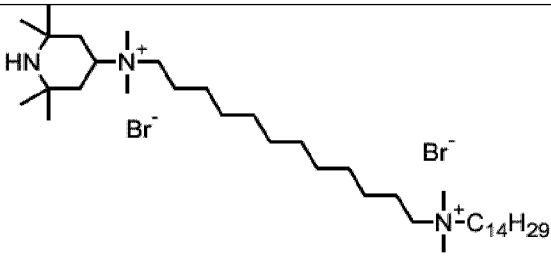
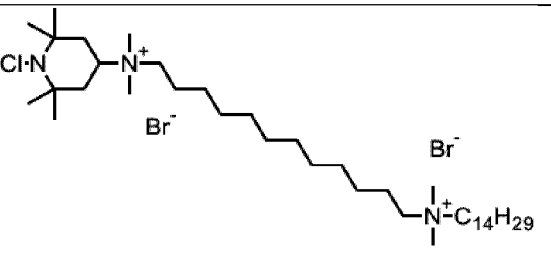
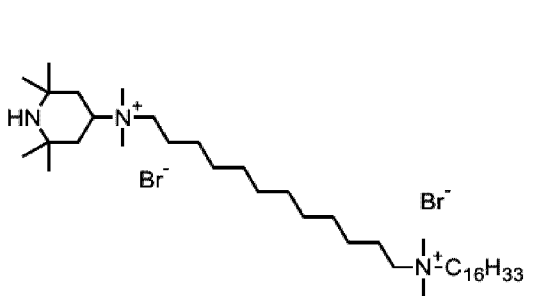

Table 1 below is a nomenclature concurrence table that matches the formulae provided above with a name used to describe the biocide compounds referred to in the experimental data provided in further examples below.

Table 1. A Nomenclature Concurrence Table For Compounds of the Present Disclosure.

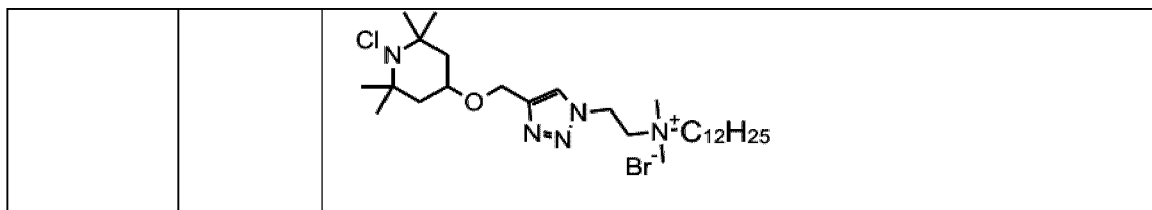
Biocide Name	Formula	General Structure
PIP-C12	5	
PIP-C12Cl	8	
PIP-C14	6	
PIP-C14Cl	9	

PIP-C16	7	
PIP-C16Cl	10	
PIP-C6-C12	13	
PIP-C6-C12-Cl	14	
PIP-C6-C14	11	
PIP-C6-C14-Cl	12	

<p>PIP-C6-C16</p>	<p>15</p>	
<p>PIP-C6-C16-Cl</p>	<p>16</p>	
<p>PIP-C8-C14</p>	<p>17</p>	
<p>PIP-C8-C14-Cl</p>	<p>18</p>	
<p>PIP-C10-C14</p>	<p>19</p>	
<p>PIP-C10-C14-Cl</p>	<p>20</p>	

<p>PIP-C12-C12</p>	<p>21</p>	
<p>PIP-C12-C12-Cl</p>	<p>22</p>	
<p>PIP-C12-C14</p>	<p>23</p>	
<p>PIP-C12-C14-Cl</p>	<p>24</p>	
<p>PIP-C12-C16</p>	<p>25</p>	
<p>PIP-C12-C16-Cl</p>	<p>26</p>	

C18Cl	47	
PIP-C6-C2-NH2	27	
HYD-C3-C14Cl phosphate	29	
P12	N/A	
P12Cl	N/A	



Example 11 –Experimental Data

Minimum Inhibitory Concentration

5 Minimum inhibitory concentration (MIC) is the highest dilution, or smallest concentration, of a compound that exhibits biocidal activity by killing microbes or inhibiting the growth of microbes. In these examples, *Escherichia coli* (E. coli) ATCC 25922 were grown overnight in Mueller Hinton (MH) broth at 37 °C. The concentration of the bacteria was regulated at a density equivalent to about a 0.5 McFarland standard of 1×10^8 CFU/mL by diluting in MH broth. Stock solutions of compounds were prepared in water and diluted (2 fold) in broth in a 96-well plate. A two-fold dilution of a first column was used to prepare column two and so on for 11 total columns.

10 Once the 96-well plate was loaded with the diluted series, about 10 μ L of the bacterial suspension was added to each well that contained the compounds to a final bacterial concentration of 1×10^7 CFU/mL. A growth control was established by adding 10 μ L of the bacterial inoculum into a well with 100 μ L of broth.

15 The plates incubated for 24 hours at 37 °C. A microplate reader was then utilized at 570 nm to confirm the presence of bacteria. Table 2 below provides the MIC data for the compounds tested.

Table 2. Examples of MIC data from bacterial killing experiments.

Compound	MIC value (ppm)
P12	400
P12Cl	200
PIP-C12	200
PIP-C12Cl	200
PIP-C14	50
PIP-C14Cl	50
PIP-C16	25
PIP-C16Cl	25
C18Cl	50
HYD-C3-C14ClPhosphate	50

Bactericidal Testing

Logarithmic-phase cultures of *E. coli* were prepared after culturing in the incubator at 37 °C overnight. A 4000 ppm stock solution of each biocide was prepared in water. Clean conditions were simulated by the addition of 5% fetal bovine serum (FBS). The presence of organic load was simulated by adding 20% FBS and Mueller Hinton broth (MH Broth). A control was also prepared that did not include any biocide.

The concentration of bacteria was regulated at a density equivalent to a 0.5 McFarland standard of 1×10^8 CFU/mL by diluting in PBS. One mL of the bacterial suspension and 625 μ L of each biocide solution were added in to 8.375 mL of one of: water, 5% FBS, 20% FBS and MHB. This created a final concentration of 1×10^7 CFU/mL of bacteria and 250 ppm of each biocide in a 10 mL solution. Following the different experimental contact times of 0 minutes, 1 minute, 5 minutes, 10 minutes, 30 minutes and 60 minutes 1 mL of each sample was removed and transferred to a neutralizing solution. Active chlorine was quenched by adding 0.02 M sodium thiosulfate. Long alkyl chains were quenched with a solution of PBWS with 1.4 % wt/v of lecithin and 10% wt/v of Tween 80. About 100 μ L of each bacterial suspension was then removed and diluted to one of 1×10^1 , 1×10^2 , 1×10^3 in sequence.

Next 100 μ L of each bacterial solution and the three diluted solutions were each loaded into a quadrant of tryptone soya agar plates, which were then incubated at 37 °C for 18 to 20 hours. This procedure was repeated as blank controls that included the bacterial solutions but no biocide. The number of viable bacteria on the four quadrants of the plates were counted with the controls shown as A in CFU/mL and the bacterial solutions that were treated with a biocide shown as B in CFU/mL. The total number of bacteria was calculated using the number of viable bacteria multiplied by the dilution factor.

The percent reduction of bacterial reduction was calculated as follows:

Reduction % = $(A-B)/A \times 100$ and the logarithm reduction = $\log (A/B)$. These bacterial tests were performed at least three times.

FIG. 6 shows an example of bacterial reduction data from experiments run in 0 % FBS. FIG. 7 shows an example of bacterial reduction data from experiments run in 5% FBS. FIG. 8 shows an example of bacterial reduction data from experiments run in 20% FBS. In FIG.6 to FIG. 8 the biocides used were P12, P12Cl, PIP-C12, PIP-C12Cl, PIP-C14, PIP-C14Cl, PIP-C16, PIP-C16Cl, C18Cl and HYD-C3-C14ClPhosphate which were present at a concentration of 250 ppm.

FIG. 9 shows an example of bacterial reduction data from experiments run with 5% FBS with sodium hypochlorite (NaOCl). The biocides used in the experiments that generated the data in FIG. 8

were P12, P12 and NaOCl (1:1), P12 and NaOCl (1:1.5), PIP-C14, PIP-C14 and NaOCl (1:1) and PIP-C14 and NaOCl (1:1.5).

FIG. 10 shows an example of bacterial reduction data from experiments run in MH Broth. The biocides used in FIG. 10 were P12Cl, C18Cl and HYD-C3C14ClPhosphate.

FIG. 11 shows an example of bacterial reduction data from further experiments run in 0% FBS with the following biocides: PIP-C6-C12, PIP-C6-C12Cl, PIP-C6-C14, PIP-C6-C14Cl, PIP-C6-C16, PIP-C6-C16Cl, PIP-C8-C14, PIP-C8-C14Cl, PIP-C10-C14, PIP-C10-C14Cl, PIP-C12-C12, PIP-C12-C12Cl, PIP-C12-C14, PIP-C12-C14Cl, PIP-C12-C16 and PIP-C12-C16Cl. FIG. 12 shows another example of bacterial reduction data using these biocides in 5% FBS. FIG. 13 shows another example of bacterial reduction data using these biocides in 20% FBS.

Cytotoxicity

To test the cytotoxicity, neonatal human dermal fibroblast cells (ATCC-PCS-201) were evaluated using some of the biocides of the present disclosure in an MTT assay. The fibroblasts were grown in fibroblast basal media that was supplemented with a fibroblast growth kit and incubated in 5% CO₂ in humidified conditions at 37 °C. After the fibroblasts reached 80% confluency, the cells were tryponized, quantified with a hemocytometer, seeded onto tissue culture treated polystyrene 96 well plates at a final density of 10⁴ cell/mL and further incubated at 37 °C for 24 hours. The supernatant was then removed and replaced with either medium (control) or one of the biocide solutions at various concentrations. Table 3 below summarizes examples of cytotoxicity data shown as half maximal inhibitory concentration (IC₅₀) values in ppm.

Table 3. Half maximal inhibitory concentration (IC₅₀) values.

Compound	IC ₅₀ Value (ppm)
P12	500
P12-Cl	200
PIP-C12	100
PIP-C12Cl	50
PIP-C14	20
PIP-C14Cl	50
PIP-C16	10
PIP-C16Cl	10
PIP-C18Cl	20
HYD-C3-C14ClPhosphate	50

Example 12 – Liquid Disinfectant

5 Some embodiments of the present disclosure relate to a liquid disinfectant that comprises one or more of the biocide compounds described herein and other additive ingredients that are suitable for liquid disinfectants. Optionally, the biocide compound may be provided as a concentrate and the other additive ingredient may be a diluent, such as water, to provide a liquid disinfectant that is a suitable concentration for a given application.

10 Some embodiments of the present disclosure relate to a liquid disinfectant that comprises one or more of the biocide compounds described herein, a potentiator and optionally another additive ingredient. The potentiator compounds may enhance the biocidal activity of the biocide compounds that comprise the liquid disinfectant. Examples of suitable potentiators include ammonium chloride, a non-ionic surfactant or combinations thereof. Suitable non-ionic surfactants include at least Surfynol W485 and Tween 20.

Table 4 below summarizes an example set of data from experiments that compared the biocidal activity of a compound C18Cl but the unchlorinated form.

15 **Table 4.** A summary of examples of bacterial reduction data from experiments that tested a biocide compound and a potentiator.

Bacteria ^a		Synthetic compounds ^b	Bacteria reduction at various contact times (min)					
			1	10	30	60	90	120
			Log ₁₀	Log ₁₀	Log ₁₀	Log ₁₀	Log ₁₀	Log ₁₀
Gram-negative	<i>E. coli</i> ATCC 25922	Compound 18	0.78	1.95	2.84	2.96	3.76	4.20
		Compound 18 + 0.1% NH₄Cl	0.64	1.31	1.78	1.80	2.16	2.63
		Compound 18 + 1% NH₄Cl	1.18	2.79	3.96	7.41	7.41	7.41
		Compound 18 + 10% NH₄Cl	0.14	0.80	3.06	4.03	4.51	7.41

20 Without being bound by any particular theory, the inventors postulate that when the example of a potentiator, ammonium chloride, was present between 1 to about 10% wt/wt the biocide demonstrated higher killing of *E. coli* at 120 minutes when compared to the biocide with 0.1% of ammonium chloride and the biocide alone.

FIG. 18 shows two examples of bacterial killing data from experiments that combined compound C18 (Formula 47) with two non-ionic surfactants Tween 20 (T) and Surfynol W485 (S) to form a liquid disinfectant for killing bacteria present on a hard surface.

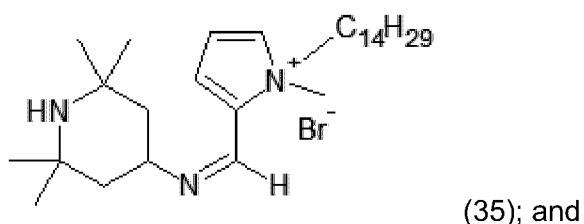
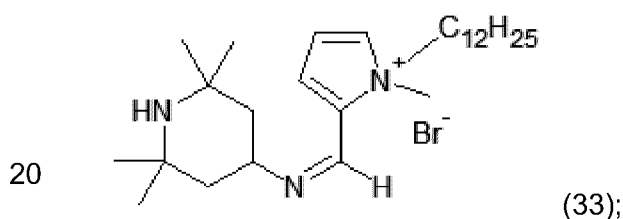
5 FIG. 18A shows that T and S had the same biocidal killing effectiveness as the control (C). The compound C18 was present at 150 ppm and demonstrated a higher relative percentage of dead cells to live cells than the control, T and S. The compound C18 alone had lower bacterial killing than either of the compound C18 plus T and the compound C18 plus S.

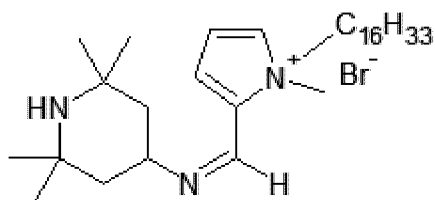
10 FIG. 18B shows that increasing the ratio of T to the compound C18 demonstrated a higher relative percentage of dead cells to live cells.

Without being bound by any particular theory, the presence of a non-ionic surfactant potentiator may have enhanced the penetration of the compound C18 into the bacteria or possibly a biofilm that formed on the hard surface and that comprised bacteria.

Example 13 – Further Biocide Compounds

15 FIG. 19 shows a chemical reaction another embodiment of the present disclosure that may be synthesized according to a reaction similar to that shown in FIG. 19. Briefly, Compounds 29 and 30 are mixed and react with about three drops of AcOH to form Compound 31. Compound 31 reacts with Compound 32, 34 or 36 to produce Compound 33, 35 or 37, respectively. Optionally Compounds 33, 35 or 37 may then be halogenated. This embodiment is a compound that has the following general formula (**Formula 33, Formula 35, Formula 37**):





(37).

Example 14 – Further Synthesis Reaction J

FIG. 20 depicts a synthesis reaction J for synthesizing compounds according to embodiments of the present disclosure. The synthesis reaction J comprises at least five steps, labelled as A, B, C, D and E respectively in FIG. 20.

Step A comprised the steps of dissolving about 20 g (about 139 mmol) of 2-chloro-N,N-dimethylethylamine hydrochloride (shown as Compound 1 in FIG. 20) in about 50 mL water, then adding about 45.13 (5 equiv) of sodium azide to the solution, and then continuing the reaction overnight at reflux. Then, about 7 g of sodium hydroxide was added to the mixture, followed by extraction three times with about 100 mL of dichloromethane (DCM). After a solvent evaporation step, about 10.3 g (95 mmol, 65%) of (2-azido-ethyl)-dimethyl-amine (this compound is identified as Compound 2 in FIG. 20) was recovered.

Step B comprised the steps of dissolving about 5 g (44 mmol) of 2-azido-N,N-dimethylethanamine (Compound 2) in about 50 mL of acetonitrile, followed by the addition of about 48.4 mmol of either 1-bromoalkane, 1-bromotetradecane (to produce Compound 3a) or 1-bromotetradecane (to produce Compound 3b) to the solution, and then the reactions continued overnight at reflux. After a solvent evaporation step, the remaining compound was dissolved in about 5 mL of methanol followed by precipitation in about 150 mL of 50/50 ethyl acetate/ hexane. These steps produced about 11.2 g (35.11 mmol) of Compound 3a with about a 79% recovery or about 11.6 g (33.4 mmol) of Compound 3b with about a 76% recovery.

Step C comprised the steps of adding about 9 g (57 mmol) of 2,2,6,6-tetramethylpiperidin-4-ol (Compound 4 in FIG. 20) to 100 mL anhydrous tetrahydrofuran, and then stirring at ambient room temperature under nitrogen atmosphere for about 30 minutes, followed by the addition of about 2.26 g (57 mmol) of NaH (60%). After mixing for about 30 minutes, the reaction flask was placed in an oil bath set at about 60 °C. About 6.356 mL (57 mmol) of propargyl bromide (80%) was added to the mixture, and the reaction allowed continued for overnight at about 60 °C. Afterward, salts were removed by filtration after which, the solvents were removed using a rotary evaporator. Then, about 50 mL of a 1N HCl solution was used to protonate NH on the piperidine to improve its water solubility. Then, the acid was washed three times with 50 mL of dichloromethane. Then, sodium hydroxide was used to

deprotonate the NH group and make it organic-soluble (in an ice bath). Then, the solution was washed again three times with 50 mL of dichloromethane followed by washing by water. The organic layer was dried on sodium sulfate and the solvent was evaporated. About 5.1 g (26.2 mmol, 46%) of Compound 5 (as shown in FIG. 20) is recovered.

5 Step D comprised the steps of dissolving about 2.91 g (15 mmol) of Compound 5 in about 15 mL of methanol after which, 12.5 mmol of either Compound 3a (4.62 g) or Compound 3b (4.92 g) was added to the solution. About 0.312 g (10%) of copper (II) sulfate was dissolved in about 2 mL water and then added to the solution. Then, about 2.38 g of copper was added to the solution. The reactions were continued overnight at ambient room temperature. The reaction product was then filtered using DIAION®
10 CR20-01 polyamine resin beads to remove the copper particles and to produce either Compound 6a or Compound 6b (as shown in FIG. 20).

Step E comprised the steps of dissolving about 500 mg of either Compound 6a or Compound 6b in 2 mL of water and 8 mL of acetone (10 mL total) after which, about 450 μ l of tert-butyl hypochlorite was added to the reaction vessel. The reaction vessel was then completely wrapped with aluminum foil and stored at 0 °C. This reaction was allowed to continue for about 60 minutes during which time, a
15 pressurized air flow was used to remove acetone and any unreacted tert-butyl hypochlorite. Water was removed using a vacuum. The reaction products of Step E are shown as either Compound 7a or 7b in FIG. 20.

20 Example 15 – Mass Spectrometry Analysis

Nuclear Magnetic Resonance (NMR) spectra were recorded at room temperature in 5 mm NMR tubes on a Bruker Avance 300 MHz NMR spectrometer. Accurate mass measurements were performed using a PERKINELMER® SCIEX® PROTOF 2000 MALDI-OTOF Mass Spectrometer.

25 Compound 2: $^1\text{H-NMR}$ (CDCl_3 , 300 Hz) 3.35 (t, 2H), 2.5 (t, 2H), 2.27 (s, 6 H); $^{13}\text{C-NMR}$ (CDCl_3 , 75 Hz); HRMS (MALDI-TOF) m/z: $[\text{M-Cl}]^+$ calculated for $\text{C}_{24}\text{H}_{48}\text{N}_3\text{O}_2^+$, 410.3741; found: 410.3746.

Compound 3a: $^1\text{H-NMR}$ (D_2O , 300 Hz) 4.04 (t, 2H), 3.62 (t, 2H), 3.43 (t, 2H), 3.22 (s, 6 H), 1.73-1.9 (m, 2H), 1.23-1.48 (m, 18H), 0.91 (t, 3H); $^{13}\text{C-NMR}$ (D_2O , 75 Hz); HRMS (MALDI-TOF) m/z: $[\text{M-Cl}]^+$ calculated for $\text{C}_{24}\text{H}_{48}\text{N}_3\text{O}_2^+$, 410.3741; found: 410.3746.

30 Compound 3b: $^1\text{H-NMR}$ (D_2O , 300 Hz) 4.04 (t, 2H), 3.62 (t, 2H), 3.43 (t, 2H), 3.22 (s, 6 H), 1.73-1.9 (m, 2H), 1.23-1.48 (m, 22H), 0.91 (t, 3H); $^{13}\text{C-NMR}$ (D_2O , 75 Hz); HRMS (MALDI-TOF) m/z: $[\text{M-Cl}]^+$ calculated for $\text{C}_{24}\text{H}_{48}\text{N}_3\text{O}_2^+$, 410.3741; found: 410.3746.

Compound 4: $^1\text{H-NMR}$ (CDCl_3 , 300 Hz) 4.17 (s, 2H), 3.83-3.96 (m, 1H), 2.39 (s, 1H), 1.97-1.96 (d, 1H), 1.93-1.92 (d, 1H), 1.17 (s, 6H), 1.12 (s, 6H), 0.99 (t, 2H); $^{13}\text{C-NMR}$ (D_2O , 75 Hz); HRMS (MALDI-TOF) m/z: $[\text{M-CI}]^+$ calculated for $\text{C}_{24}\text{H}_{48}\text{N}_3\text{O}_2^+$, 410.3741; found: 410.3746.

5 Compound 5a (Formula 40, shown below) : $^1\text{H-NMR}$ (D_2O , 300 Hz) 8.22 (s, 1H), 5.057 (s, 2H), 4.72 (t, 2H), 4.01 (t, 2H), 4.09-4.24 (m, 1H), 3.31 (t, 2H), 3.18 (s, 6H), 2.30 (d, 1H), 2.25 (d, 1H), 1.61 (m, 2H), 1.50 (s, 6H), 1.48 (s, 6H), 1.23-1.38 (m, 18H), 1.18 (t, 2H), 0.88 (t, 3H); $^{13}\text{C-NMR}$ (D_2O , 75 Hz); HRMS (MALDI-TOF) m/z: $[\text{M-CI}]^+$ calculated for $\text{C}_{24}\text{H}_{48}\text{N}_3\text{O}_2^+$, 410.3741; found: 410.3746.

Purity QNMR > 99%.

10 Compound 5b (Formula 41, shown below): $^1\text{H-NMR}$ (D_2O , 300 Hz) 8.22 (s, 1H), 5.057 (s, 2H), 4.72 (t, 2H), 4.01 (t, 2H), 4.09-4.24 (m, 1H), 3.31 (t, 2H), 3.18 (s, 6H), 2.30 (d, 1H), 2.25 (d, 1H), 1.61 (m, 2H), 1.50 (s, 6H), 1.48 (s, 6H), 1.23-1.38 (m, 22H), 1.18 (t, 2H), 0.88 (t, 3H); $^{13}\text{C-NMR}$ (D_2O , 75 Hz); HRMS (MALDI-TOF) m/z: $[\text{M-CI}]^+$ calculated for $\text{C}_{24}\text{H}_{48}\text{N}_3\text{O}_2^+$, 410.3741; found: 410.3746.

Purity QNMR > 96%.

15 Compound 6a (Formula 38, shown below): $^1\text{H-NMR}$ (DMSO, 300 Hz) 8.38 (s, 1H), 4.99 (t, 2H), 4.56 (s, 2H), 3.93 (t, 2H), 3.72-3.88 (m, 1H), 3.12 (s, 8H), 2.1 (d, 1H), 2.06 (d, 1H), 1.55 (m, 2H), 1.4 (t, 2H), 1.32-1.0 (m, 30H), 0.84 (t, 3H).

Purity QNMR > 98%.

20 Compound 6b (Formula 39, shown below): $^1\text{H-NMR}$ (DMSO, 300 Hz) 8.38 (s, 1H), 4.99 (t, 2H), 4.56 (s, 2H), 3.93 (t, 2H), 3.72-3.88 (m, 1H), 3.12 (s, 8H), 2.1 (d, 1H), 2.06 (d, 1H), 1.55 (m, 2H), 1.4 (t, 2H), 1.32-1.0 (m, 34H), 0.84 (t, 3H).

Purity QNMR > 95%.

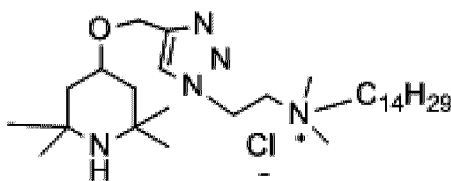
Example 16 – Biocidal Activity Assay

25 Bacterial suspensions of three different bacteria were prepared in phosphate-buffered saline (PBS, 0.1 M, pH 7.4) at a density equivalent to a 0.5 McFarland standard of 1×10^8 colony forming units (CFU)/mL. The first bacteria specie used was Community-associated (CA)-MRSA #40065 which is also referred to as the first bacteria. The second bacterial specie used was multi-drug resistant *Pseudomonas aeruginosa* #73104 (MDR) which is also referred to as the second bacteria. Both bacteria are clinically relevant strains obtained from the CANWARD (Canadian Ward Surveillance) study
30 assessing antimicrobial resistance in Canadian hospitals (online: <http://www.canr.ca>). The third bacteria specie used was *Escherichia coli* (E. coli) (ATCC 25922), which is also referred to as the second bacteria.

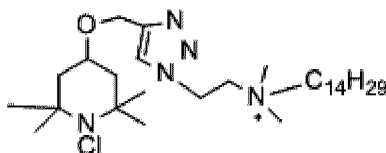
Following a 100 - times dilution to 1×10^6 CFU/mL, a 20 μ L aliquot of the each diluted bacterial suspension was added to about 60 mL of a tryptone soya broth followed by an 18-hour incubation at 37 $^{\circ}$ C. Then, a 50 μ L aliquot of the diluted and incubated bacterial suspensions were added to a solution of 5 ppm (Cl⁻) to 20 mL of PBS (0.1 M, pH 7.4) and 15 ppm (Cl⁻) to a solution of 5% Fetal Bovine Serum in PBS, which is referred to as the high protein media "HPM" in the figures. These mixtures were incubated on an orbital shaker at 37 $^{\circ}$ C.

The following compounds are embodiments of the present disclosure that were tested in the biocidal assays:

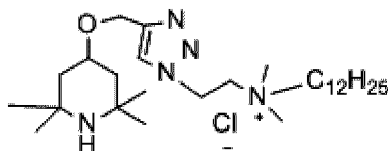
One embodiment of the present disclosure is referred to in the figures as P14 and it has the following general formula (**Formula 38**):



Another embodiment of the present disclosure is referred to in the figures as P14Cl and it has the following general formula (**Formula 39**):

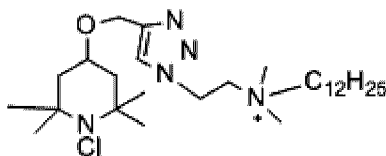


Another embodiment of the present disclosure is referred to in the figures as P12 and it has the following general formula (**Formula 40**):

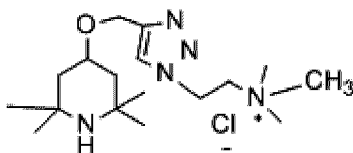


Another embodiment of the present disclosure is referred to in the figures as P12Cl and it has the following general formula (**Formula 41**):

39

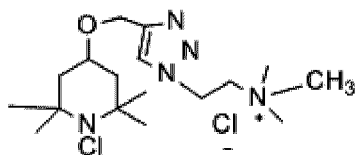


Another embodiment of the present disclosure is referred to in the figures as P1 and it has the following general formula (**Formula 42**):

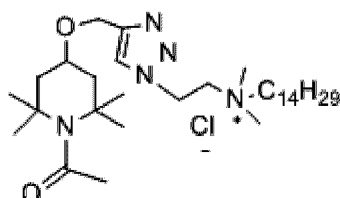


5

Another embodiment of the present disclosure is referred to in the figures as P1Cl and it has the following general formula (**Formula 43**):



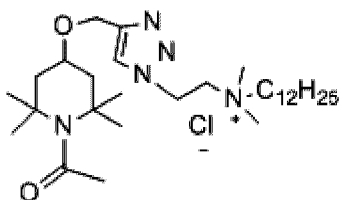
Another embodiment of the present disclosure is referred to in the figures as P14Me and it has the following general formula (**Formula 44**):



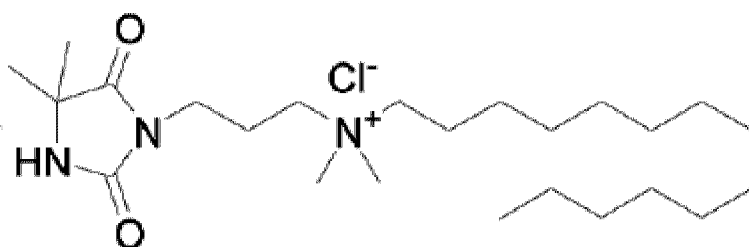
10

Another embodiment of the present disclosure is referred to in the figures as P12Me and it has the following general formula (**Formula 45**):

40

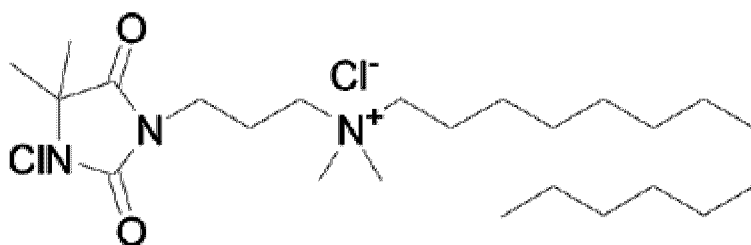


Another embodiment of the present disclosure is referred to in the figures as C17 and it has the following general formula (**Formula 46**):

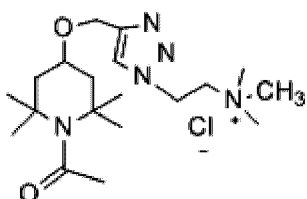


5

Another embodiment of the present disclosure is referred to in the figures as C18 and it has the following general formula (**Formula 47**):



Another embodiment of the present disclosure is referred to in the figures as P1Me and it has the following general formula (**Formula 48**):



10

The inventors used P14Me, P12Me and P1Me because these compounds have similar polarities to the compounds of Formula 39, Formula 41 and Formula 43, respectively.

5 The compounds with the general formulae 38 to 48 were used to produce a stock solution of each compound with a concentration of about 10,000 ppm (based on the concentration of Cl⁻) in PBS. Then, about 30 µL of each stock solution were added to about 20 µL of each of the diluted bacterial suspensions in either PBS or the HPM. These mixtures were then vortexed to produce a final concentration of 15 ppm (Cl⁻).

10 At 0, 2.5, 5, 10, 20, 30 and 60 minutes, a 150-µl aliquot of the mixtures was mixed with about 150 µl of a neutralizer solution (PBS buffer consisting of 1.4% [w/v] lecithin and 10% [w/v] Tween 80) to deactivate the biocidal activity of the exemplary compounds. A 30-µl aliquot of the bacterial suspension was diluted and plated onto tryptone soya agar and incubated for 24 hours, after which the bacterial colonies were counted.

15 FIG. 21A shows the biocidal-activity results of Compound 5a (C12/Formula 40) and Compound 6a (C14/Formula 38) on the first bacteria (MRSA) in PBS or HPM. FIG. 21B shows the biocidal-activity results of Compound 5b (C14/Formula 41) and Compound 6b (P14Cl/Formula 39) on the first bacteria (MRSA) in PBS or HPM. FIG. 21C shows the biocidal-activity results of Compound C17 and C18 on the first bacteria (MRSA) in PBS or HPM.

20 FIG. 22A shows the biocidal-activity results of Compound 5a (C12/Formula 40) and Compound 6a (C14/Formula 38) on the third bacteria (*E. coli*) in PBS or HPM. FIG. 22B shows the biocidal-activity results of Compound 5b (C14/Formula 41) and Compound 6b (P14Cl/Formula 39) on the third bacteria (*E. coli*) in PBS or HPM. FIG. 22C shows the biocidal-activity results of Compound C17 and C18 on the third bacteria (*E. coli*) in PBS or HPM.

25 FIG. 23A shows the biocidal-activity results of Compound 5a (C12/Formula 40) and Compound 6a (C14/Formula 38) on the second bacteria (MDR) in PBS or HPM. FIG. 23B shows the biocidal-activity results of Compound 5b (C14/Formula 41) and Compound 6b (P14Cl/Formula 39) on the second bacteria (MDR) in PBS or HPM. FIG. 23C shows the biocidal-activity results of Compound C17 and C18 on the second bacteria (MDR) in PBS or HPM.

Without being bound by any particular theory, the data from these figures may demonstrate that the P14Cl and P12Cl are effective at killing bacteria while in a liquid state and while in a HPM environment.

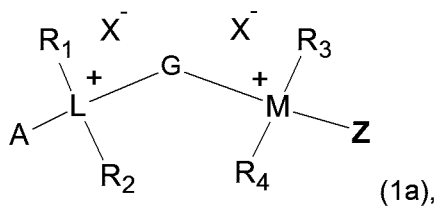
30 Example 17 – Amine and Amide Analysis

About 121 mg of the compound C18 (Formula 47) was added into 10 ml (0.0271 M) ultrapure water, 119 mg of compound 7a (as shown in FIG. 20) was added into 8 ml ultrapure water (0.0271 M)

and 125 mg of compound 7b (as shown in FIG. 20) was added in 8 ml water (0.0271 M). About 50 μ L of FBS was added to these three prepared solutions for a 120 minute time-course with titrations performed at 1, 5, 20, 60, 90 and 120 minute intervals. FIG. 24 shows the observed results from experiments run in triplicate. Without being bound by any particular theory, it was postulated that the observed drop of [Cl⁻] is caused by the presence of the FBS because no similar decrease of [Cl⁻] was detected when the compounds were kept in PBS (i.e. without protein) for 60 minutes (data not shown). The compounds 7a and 7b appear to be more stable than the C18 compound in the presence of proteins within the FBS.

I claim

1. A compound comprising the formula:



wherein L is nil or nitrogen,

when L is nil:

both R_1 and R_2 are nil,

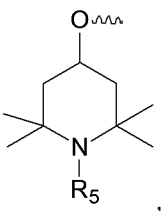
M is nitrogen or phosphorous,

when M is nitrogen, R_3 and R_4 are each $C_nH_{(2n+1)}$,

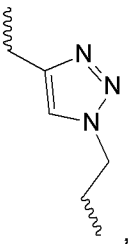
n is either 1 or 2,

when M is phosphorous, R_3 and R_4 are both one of methyl, ethyl and phenyl,

A is:



G is:



Z is $C_{na}H_{(2na+1)}$,

where $na = 12$ to 24

and R_5 is one of hydrogen, chlorine, bromine, iodine and CH_3CO ;

when L is nitrogen:

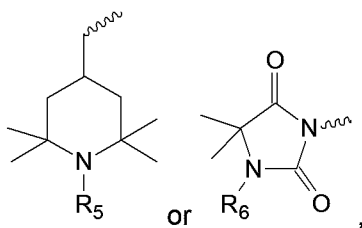
M is nitrogen or phosphorous,

when M is nitrogen, each of R_1, R_2, R_3 and R_4 are $C_{nb}H_{(2nb+1)}$,

nb is one of 1 or 2,

when M is phosphorous, R_3 and R_4 are both one of methyl, ethyl and phenyl,

A is:



G is $-(CH_2)_{nc}-$ and where nc is 0 or an integer between 1 and 12

Z is $C_{nd}H_{(2nd+1)}$,

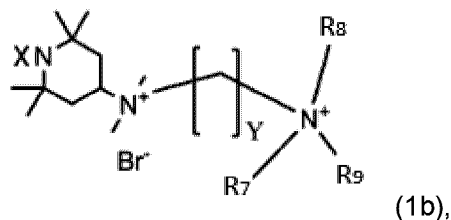
nd is an integer between 1 and 16,

R_5 and R_6 are each one of hydrogen, chlorine, bromine, iodine and CH_3CO ; and

X^- is an anionic counter-charge selected from the group of Cl^- , Br^- and PO_4^{3-} , when X^- is PO_4^{3-} the anionic counter-charge balances with respect to the cation.

2. A compound comprising the formula:

45



wherein R₉ is C_nH_{2n+1} and n is an integer between 10 and 20 or R₄ is C_pH_{2p}NH₂ and p is an integer between 1 and 10;

wherein R₇ and R₈ are each independently one of CH₃ or CH₂CH₃;

wherein Y is an integer between 4 and 20; and

wherein X is one of hydrogen, chlorine, bromine or iodine.

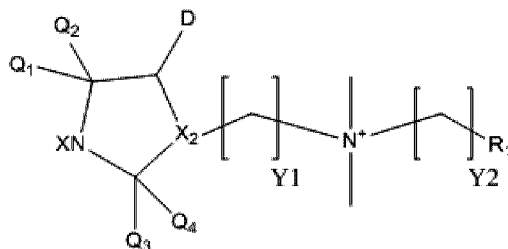
3. A compound comprising the formula:



wherein C is one of C₃H₆(COOH)₂, C₄H₈(COOH)₂, C₅H₁₀(COOH)₂, C₆H₁₂(COOH)₂ or combinations thereof; and

wherein D is V₃W³⁻,

wherein V is



wherein when X₂ is C then Q₁, Q₂, Q₃ and Q₄ are all H₂ and D is H₂ or when X₂ is N then Q₁ and Q₂ are each CH₃, Q₃ and Q₄ are collectively =O and D is =O;

wherein Y₁ and Y₂ are each independently an integer between 2 and 12;

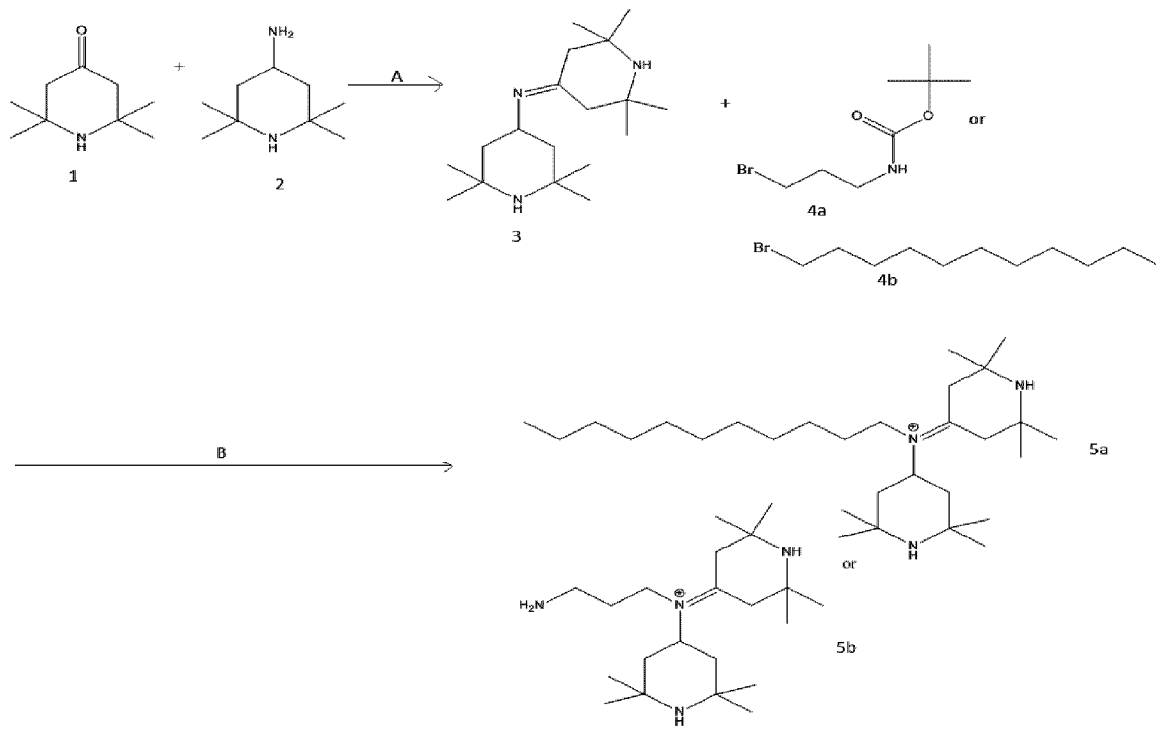
wherein W is PO₄, SO₄, HPO₄ or W³⁻ is 3F⁻, 3Cl⁻ or 3Br⁻;

wherein R₁ is CH₃ or =CH₂; and

wherein X is one of hydrogen, chlorine, bromine or iodine.

4. A liquid disinfectant comprising the compound of claim 1, 2, 3 or combinations thereof.
5. The liquid disinfectant of claim 4 further comprising a potentiator.
6. Use of the liquid disinfectant of either of claims 4 or 5 for cleaning, disinfecting or controlling microbe growth.
7. Use of the liquid disinfectant of either of claims 4 or 5 in an environment with a protein content.
8. The use of claim 7 wherein the environment is at least one of: an animal housing facility; an agricultural facility; transportation equipment; a horticultural facility; an aqua-culture facility; laboratory equipment; a water-based building material; a water-processing facility; a pulp and paper processing facility; an industrial, metal-working facility; an industrial manufacturing facility; an air washer system; an oil and/or gas well; an oil and gas transport facility; an oil and gas storage facility; a brewery pasteurizing facility; a food and beverage canning facility; a residential home; a hospital; a commercial facility; a commercial and consumer product preservation facility; and a hard surface within a food or beverage processing facility.
9. The use of claim 6 or 7 as a preservative for food preservation; preservation of household or commercial cleaning products; and preservation of personal hygiene and care products.
10. The use of claim 7 wherein the environment is one of: a swine farrowing unit; a nursery, a finisher house; a processing plant, agricultural equipment; personal agricultural equipment; an evaporative cooler; a humidifying system; a ceiling fan; a poultry coop; a chick tray; a plastic chick box; hard and non-porous surfaces and equipment used in a veterinary facility; a stable; a foot dip; a kennel; a horse box; a feed room; tack; a feedlot facility; a quarantine pen; a cage; an animal transportation vehicle; an animal holding facility; an irrigation tank; an irrigation line; a fogging operation; a facilities for commercial dairy operations; and equipment, instruments and utensils employed in animal husbandry.
11. The use of claim 7 wherein the environment is one of an emergency disease control environment and a animal biosecurity environment.

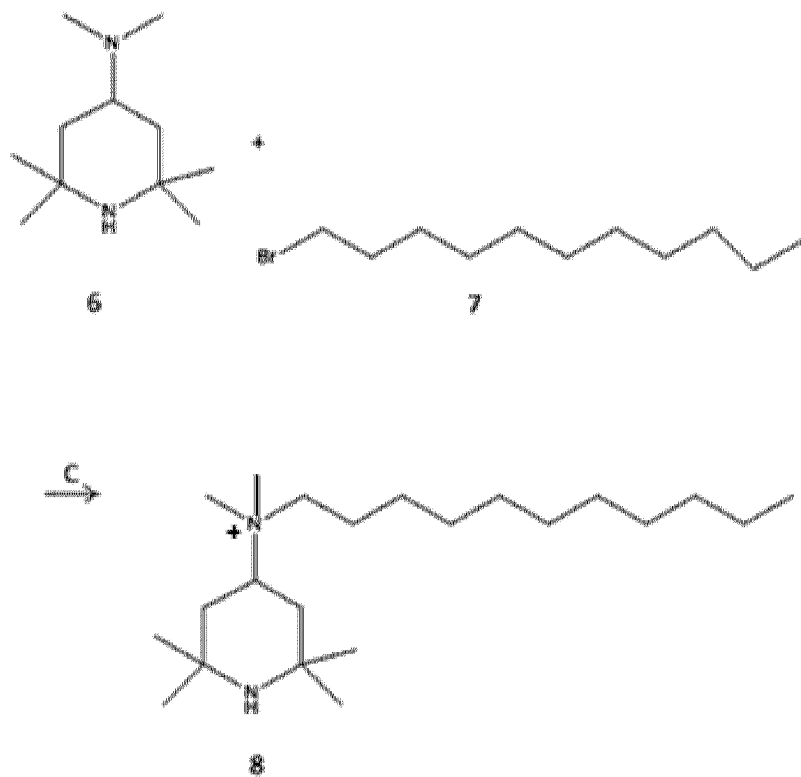
12. The use of claim 7 wherein the environment is horticultural.
13. The use of claim 7 wherein the environment is an aqua-culture facility or aqua-culture vehicle.



Synthesis Reaction A

FIG. 1

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Synthesis Reaction B

FIG. 2

Fig. 3A: Synthesis Reaction C

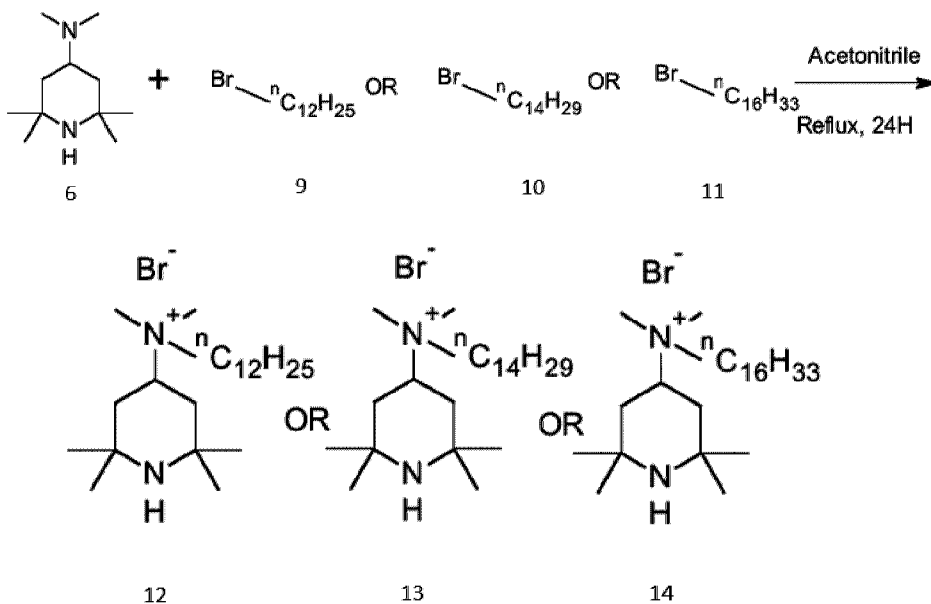


Fig. 3B: Halogenation Reaction D

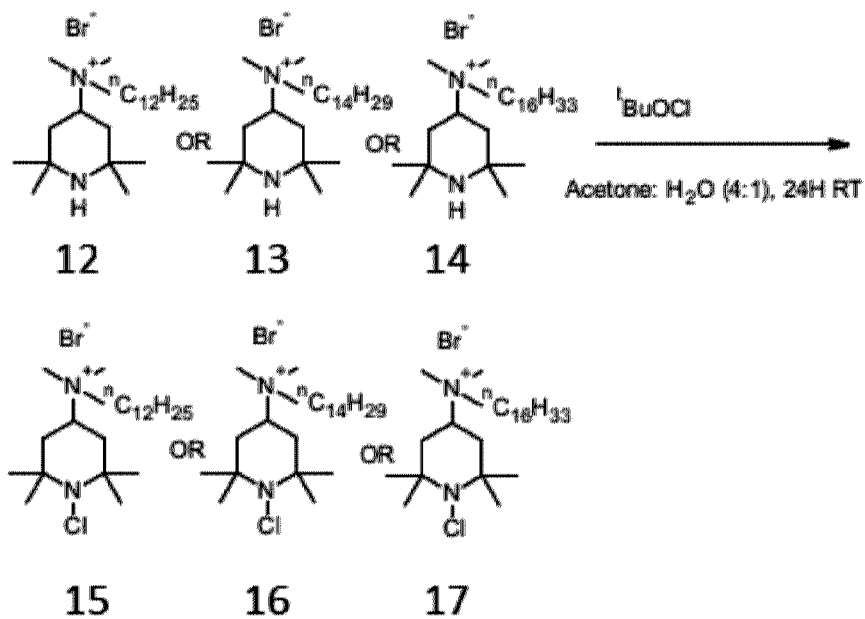
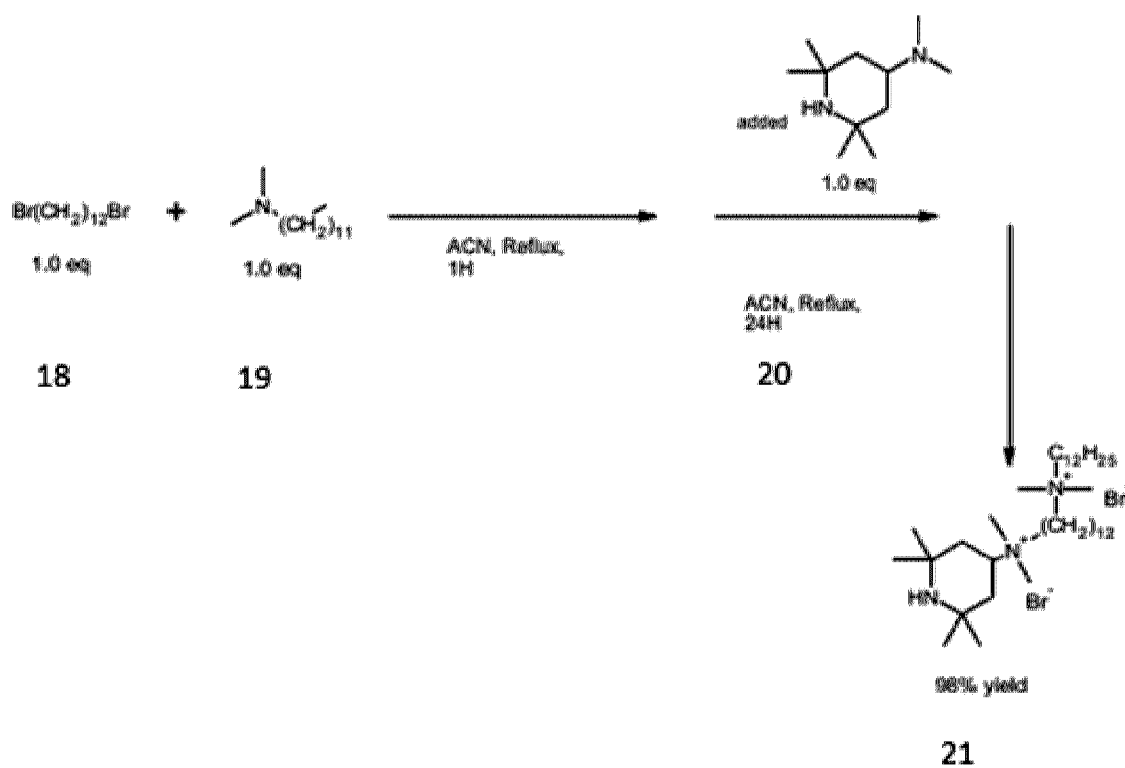


FIG. 3

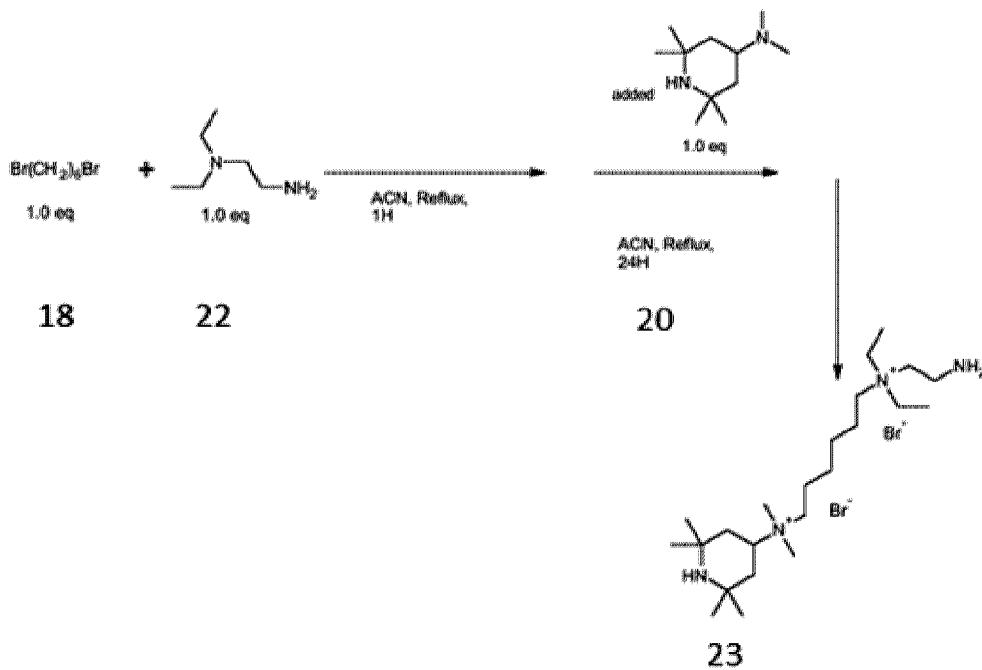
4 / 24



Synthesis Reaction E

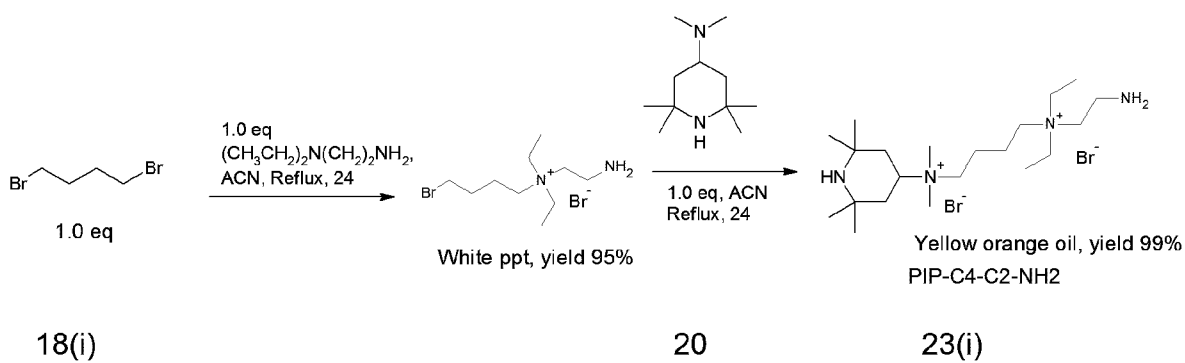
FIG. 4

Fig. 5A



Synthesis Reaction F

Fig. 5B



Synthesis Reaction F (i)

FIG. 5

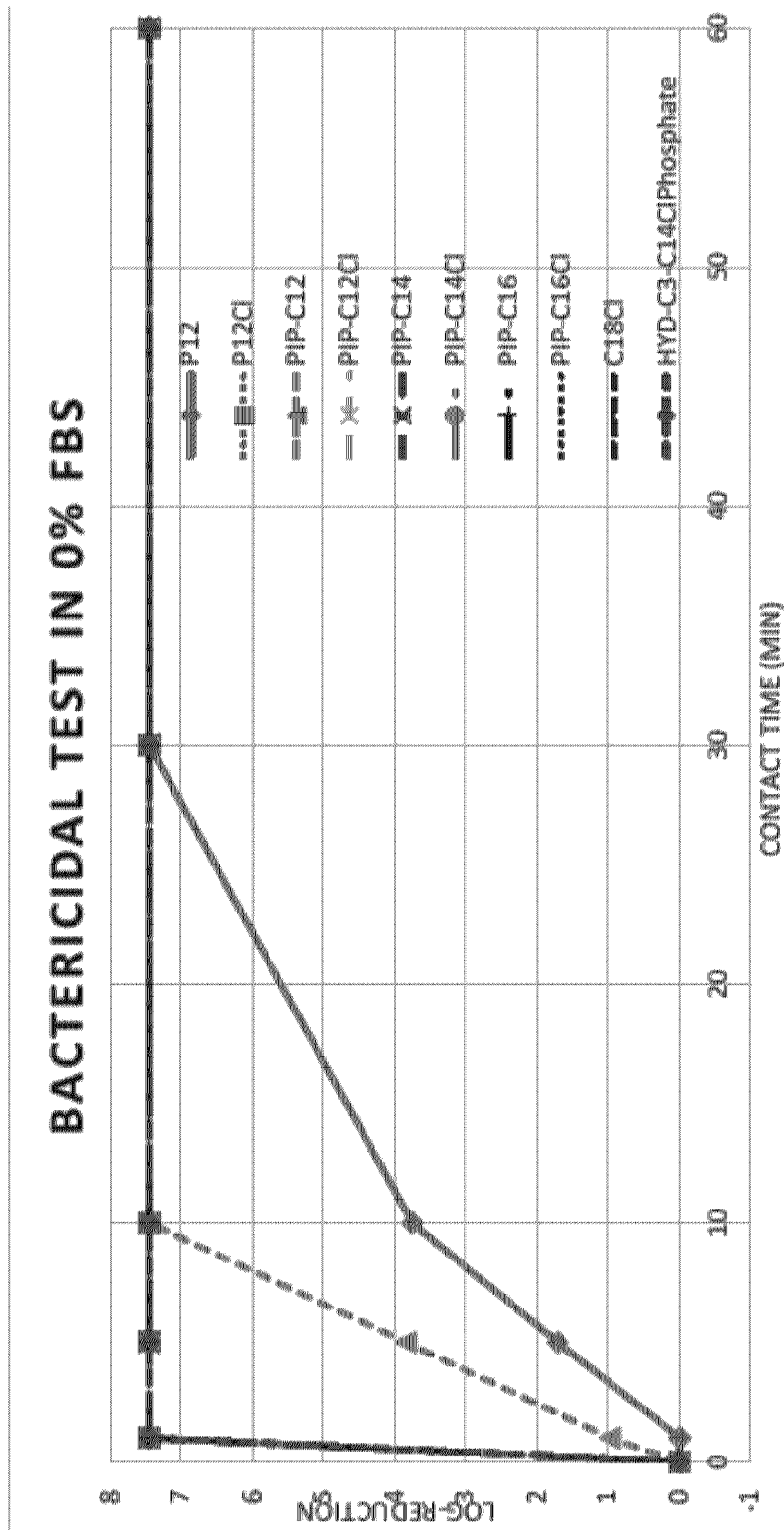


FIG. 6

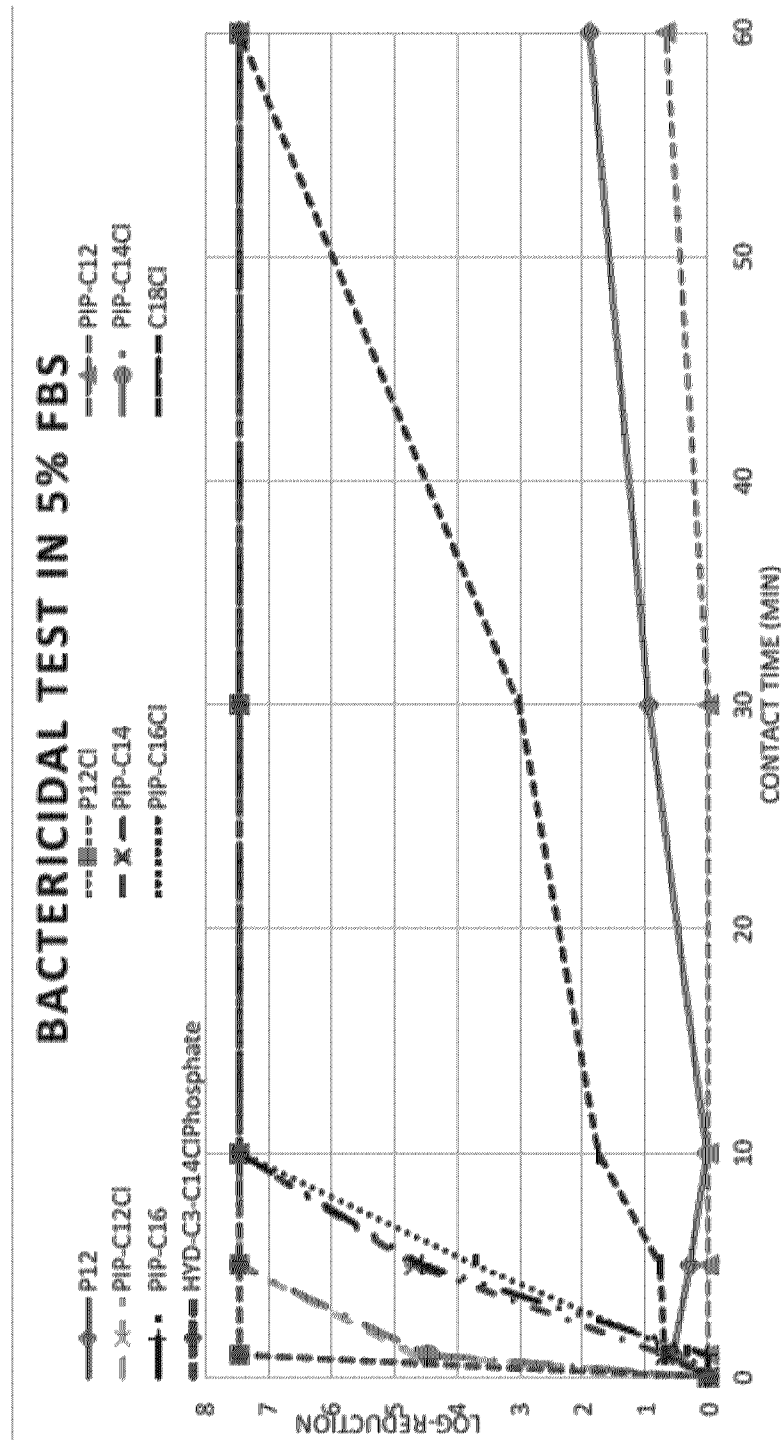


FIG. 7

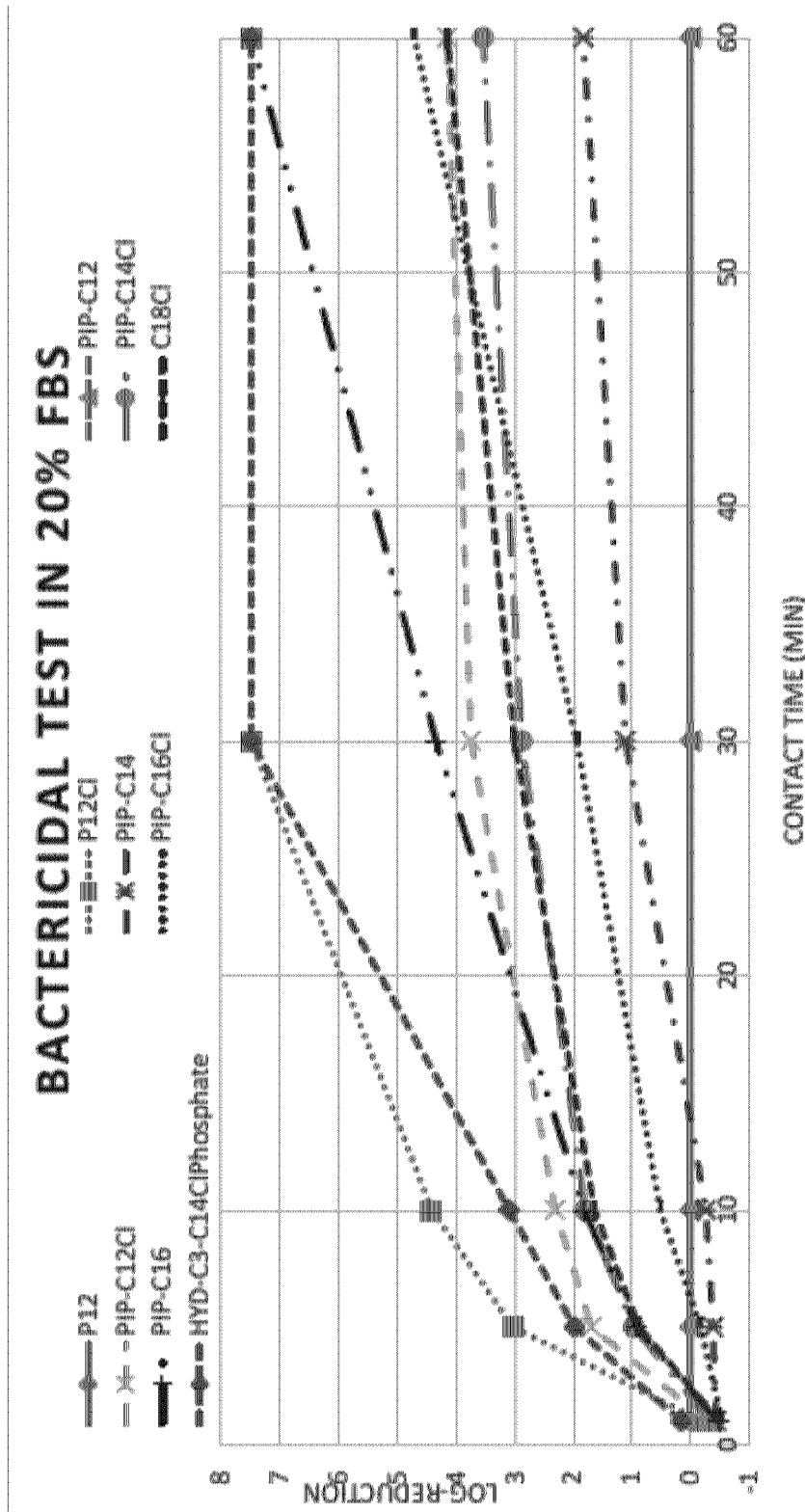


FIG. 8

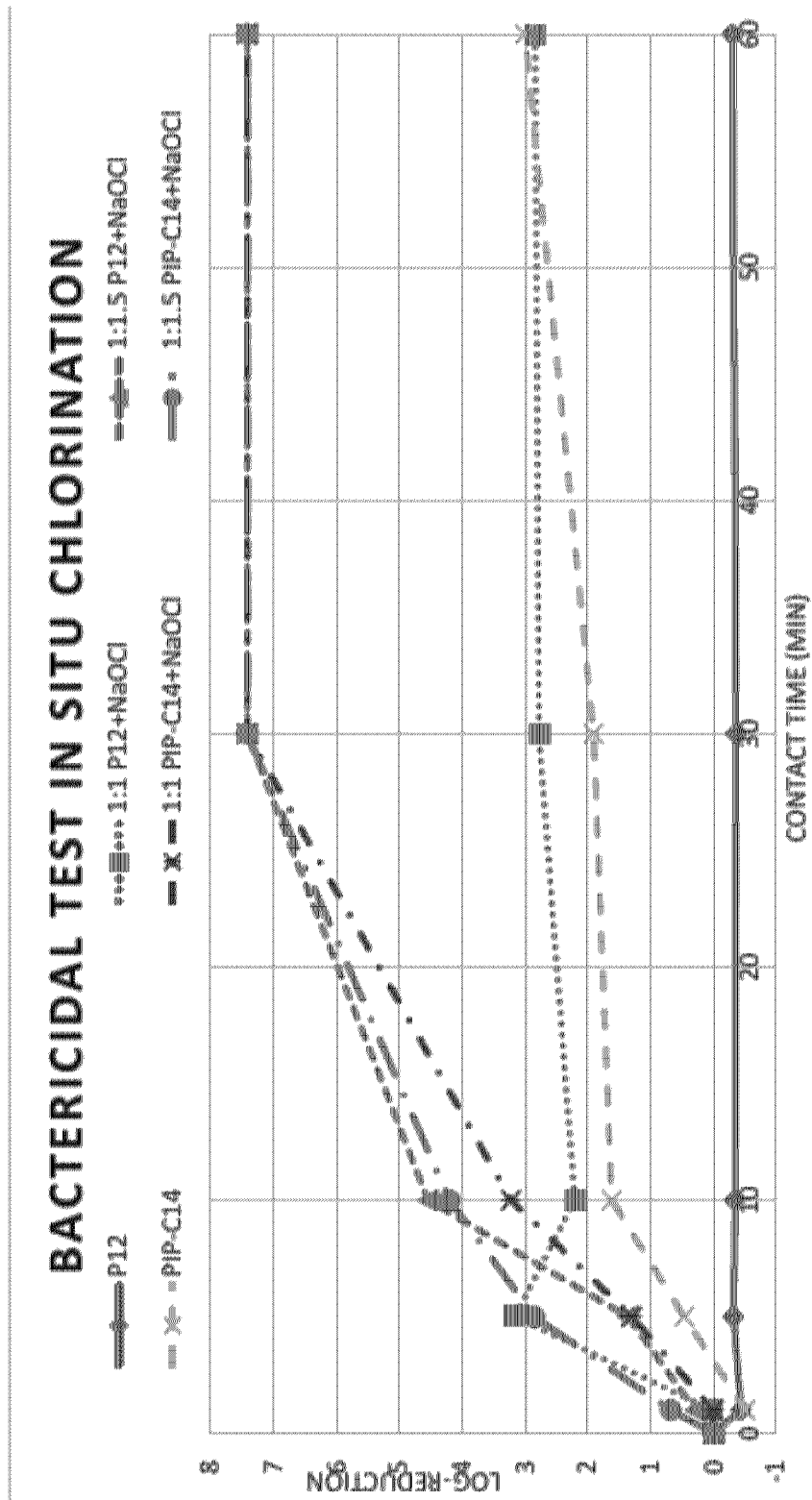


FIG. 9

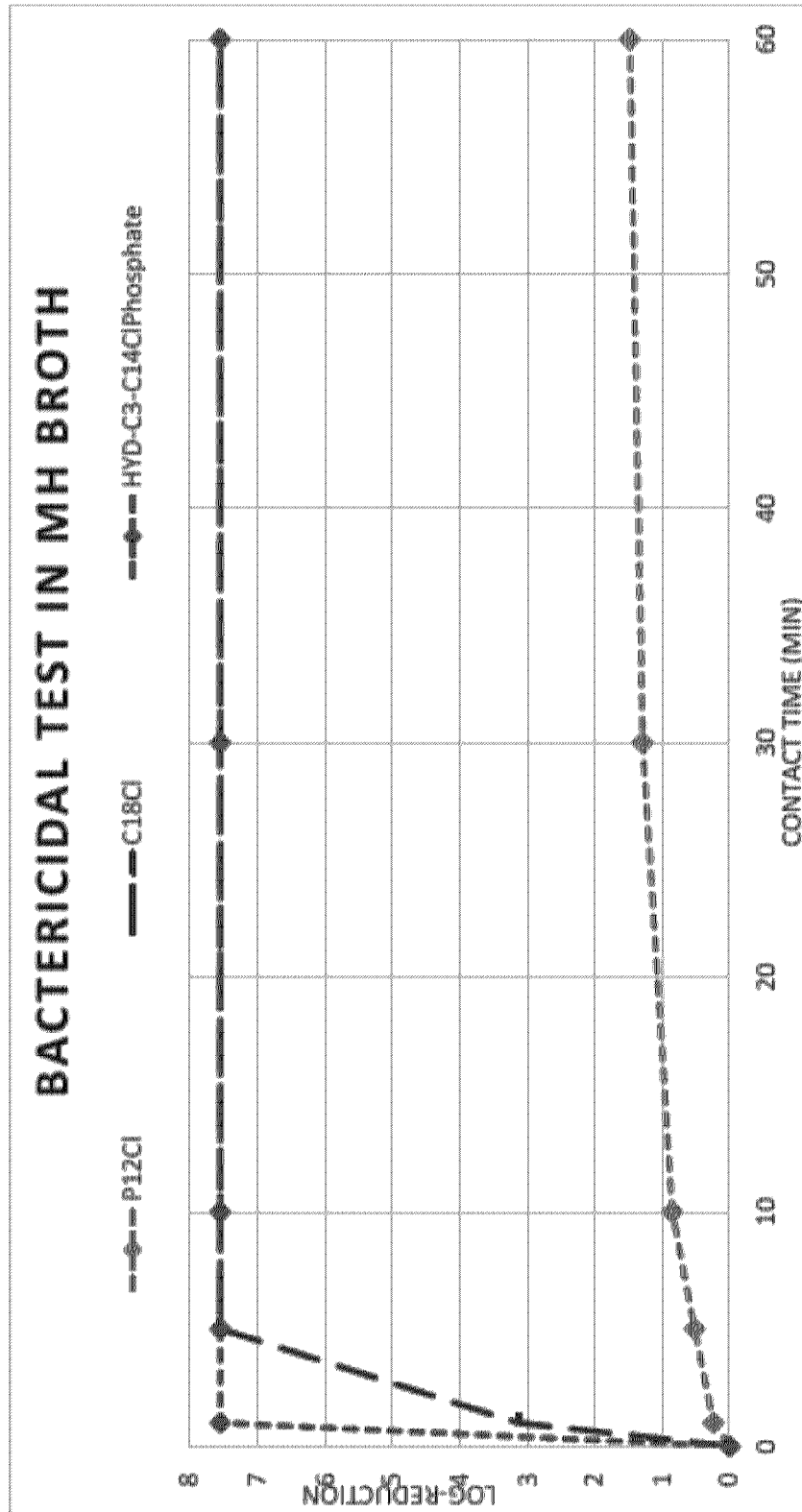


FIG. 10

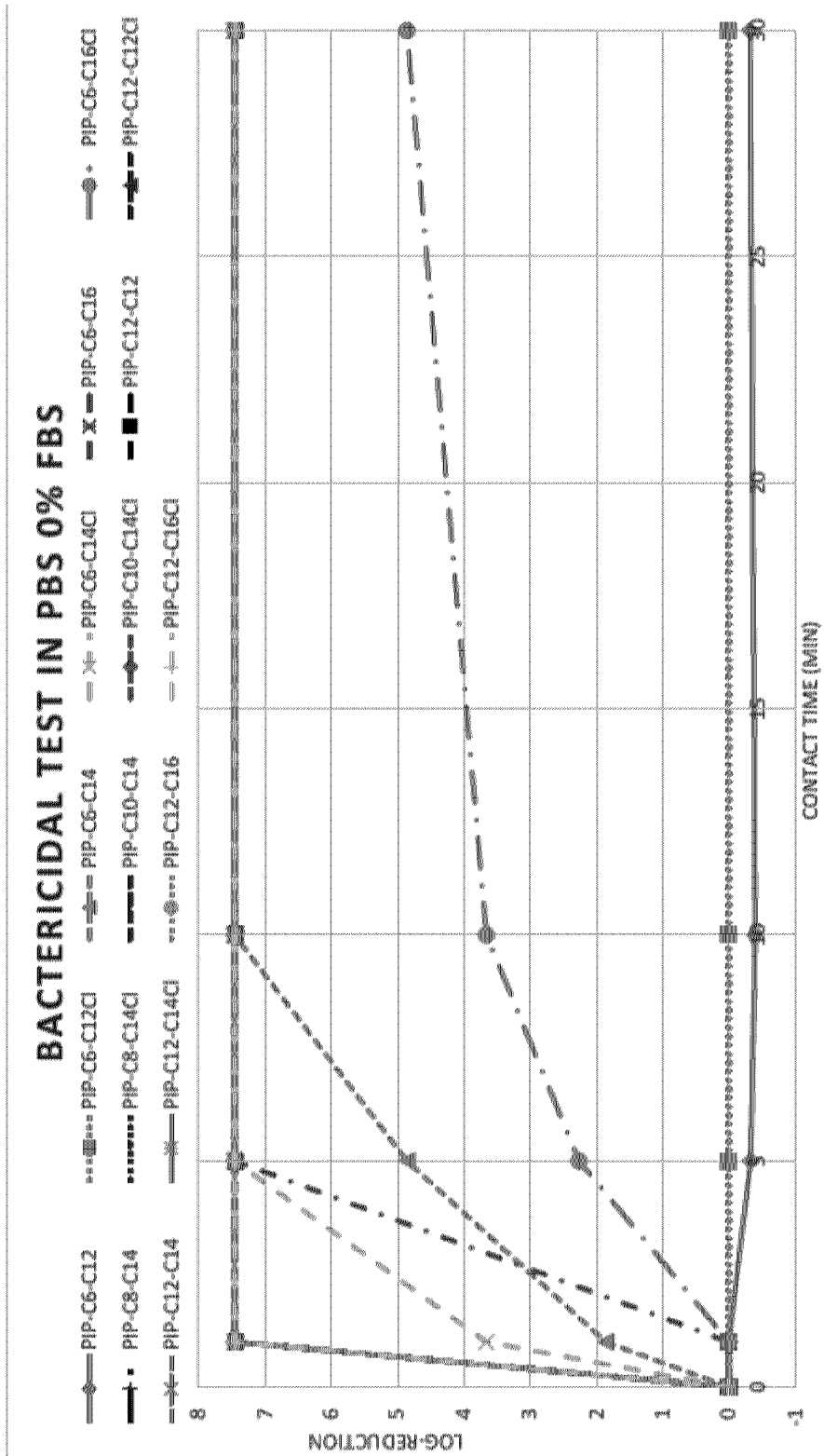


FIG. 11

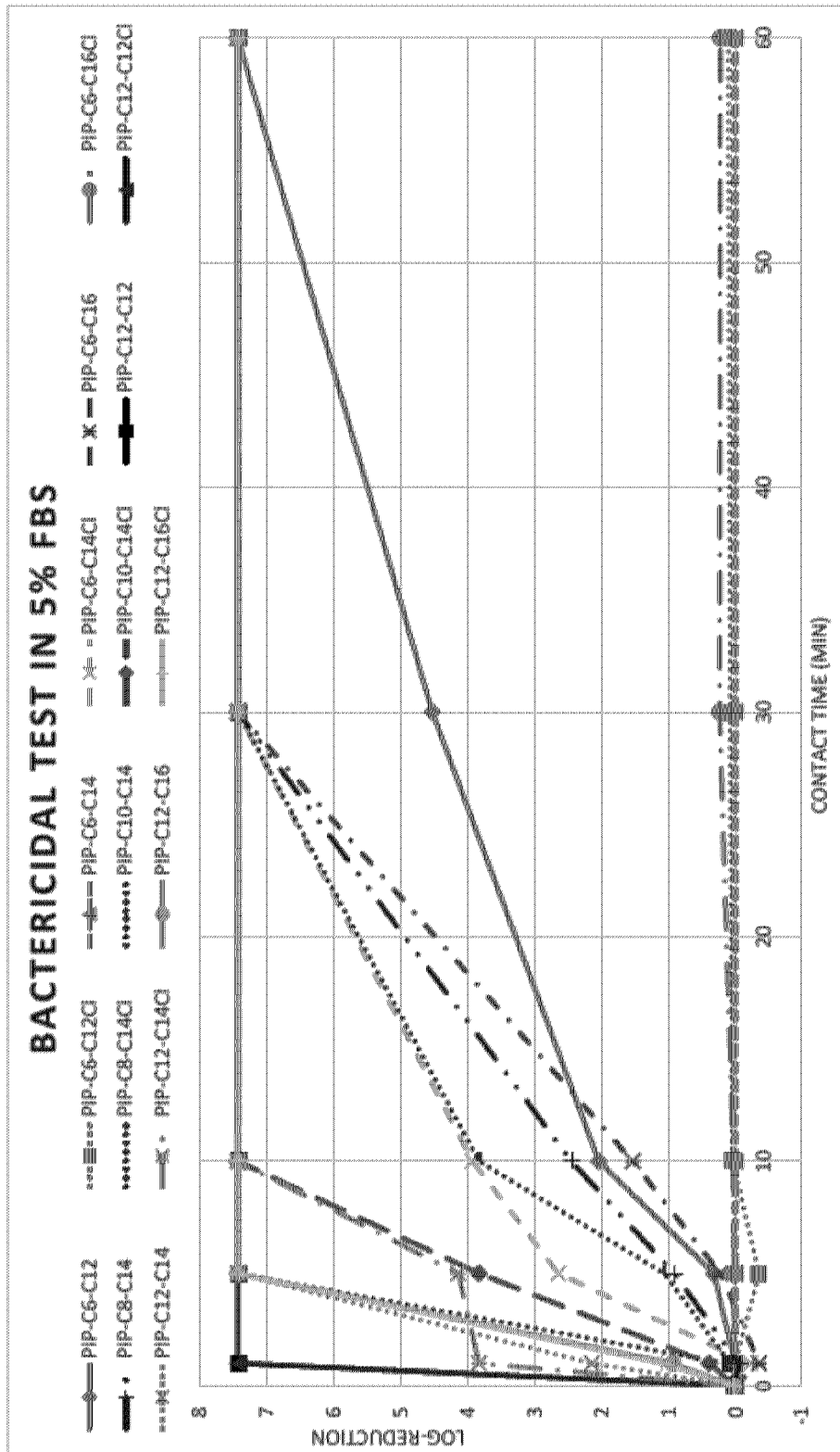


FIG. 12

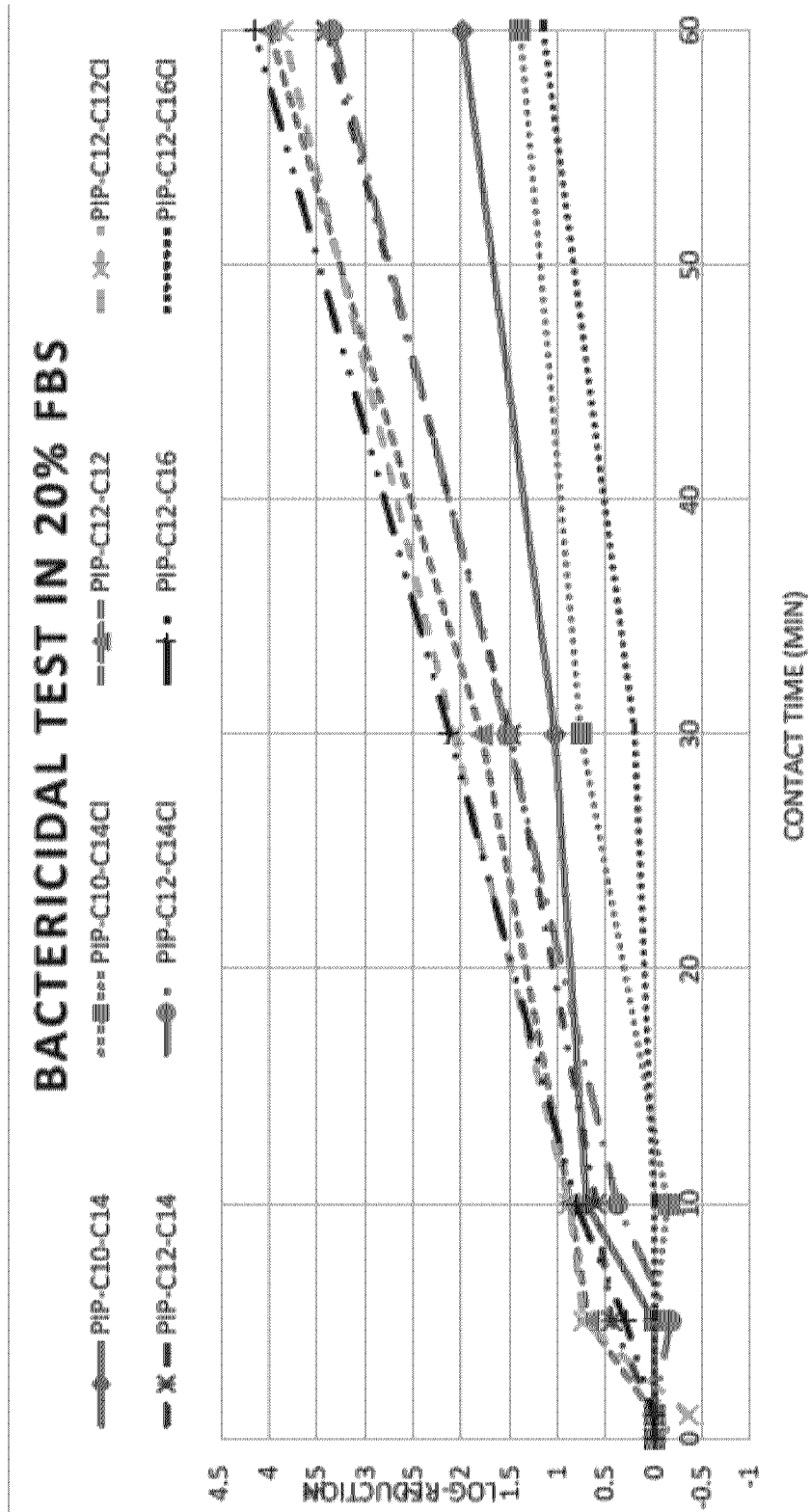


FIG. 13

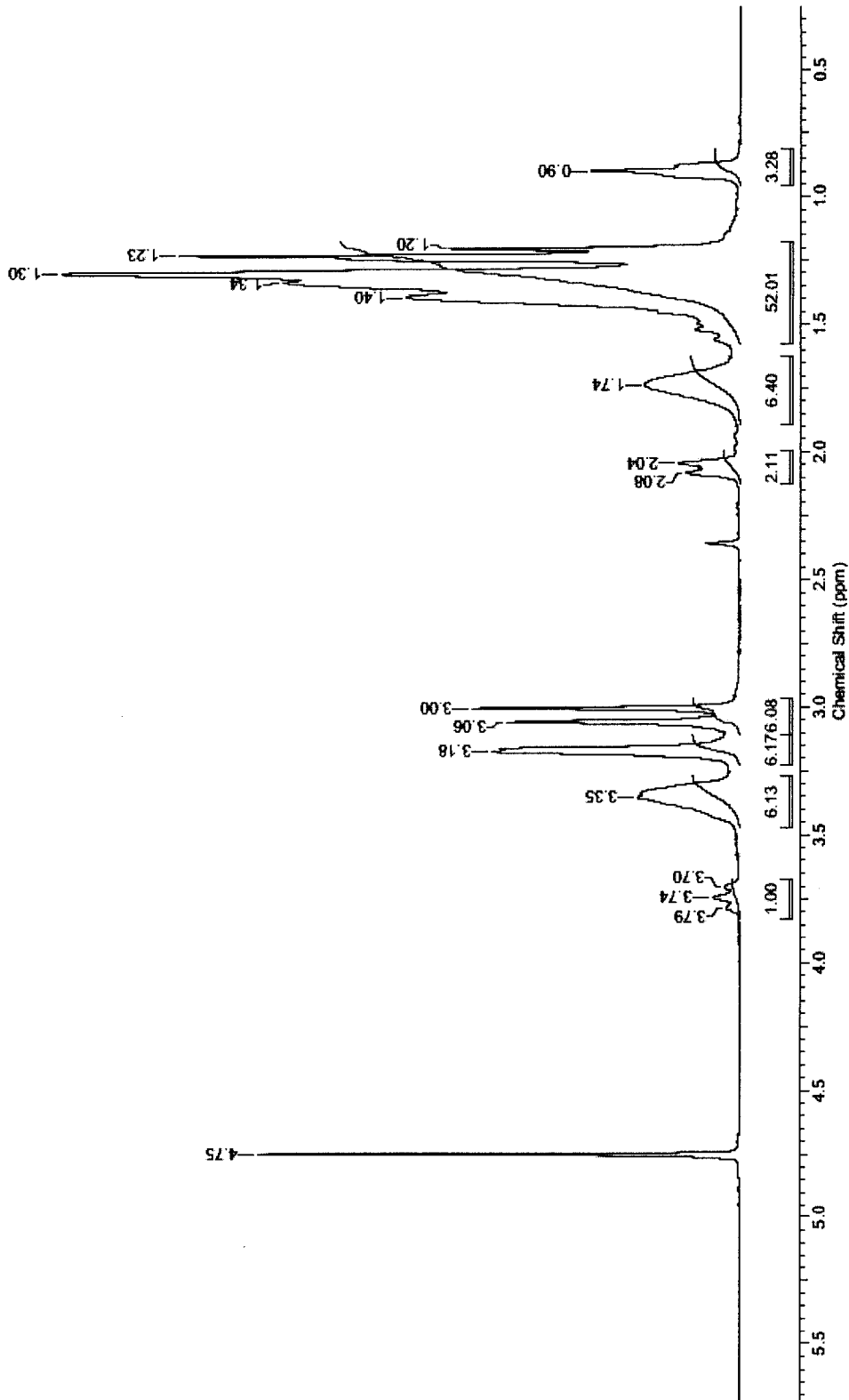


FIG. 14

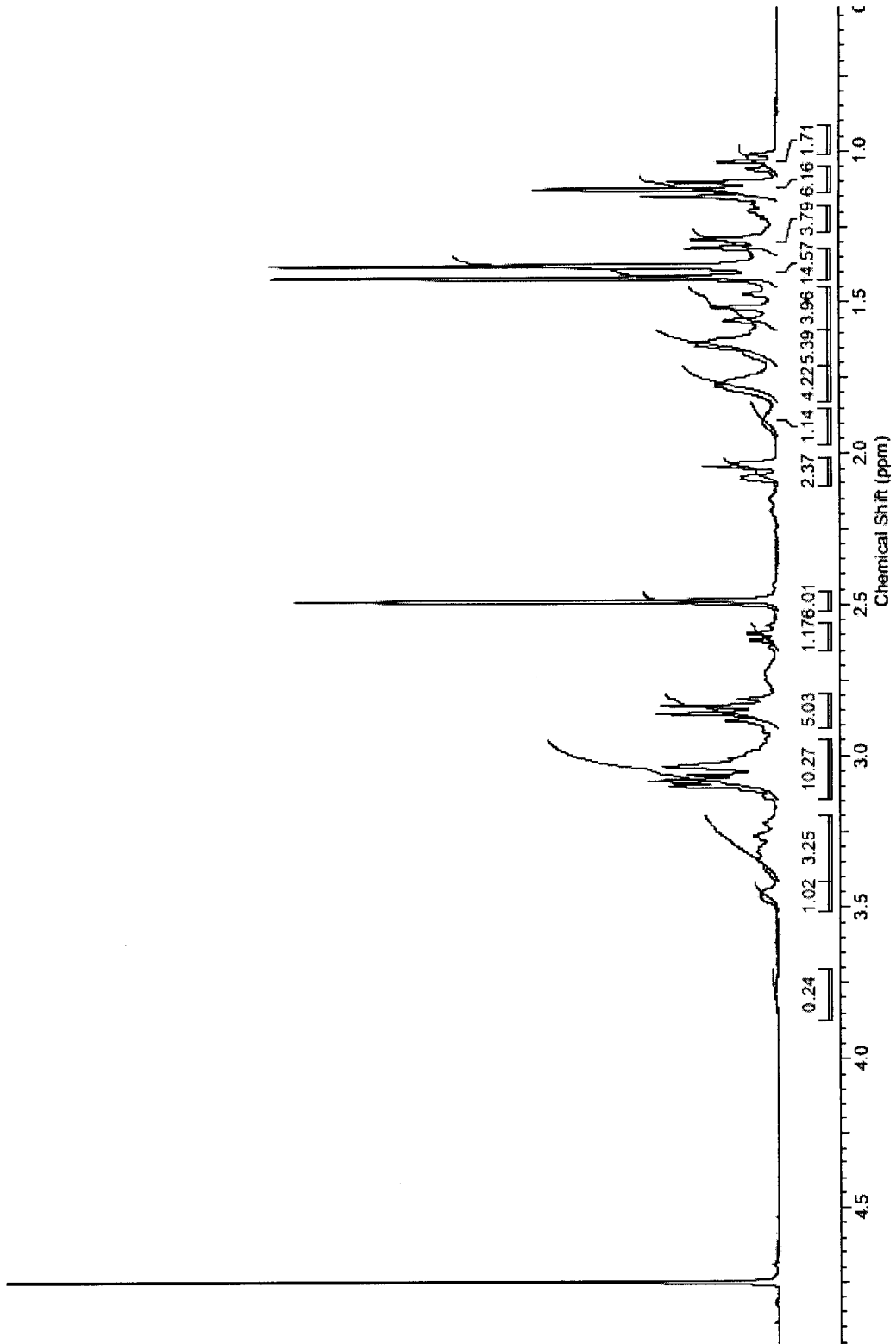
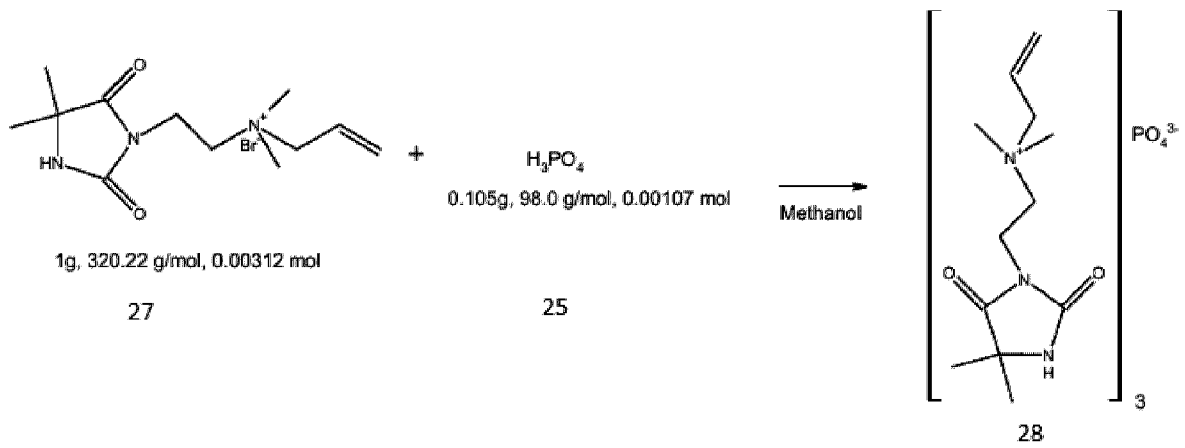


FIG. 15



Synthesis Reaction H

FIG. 17

FIG. 18A

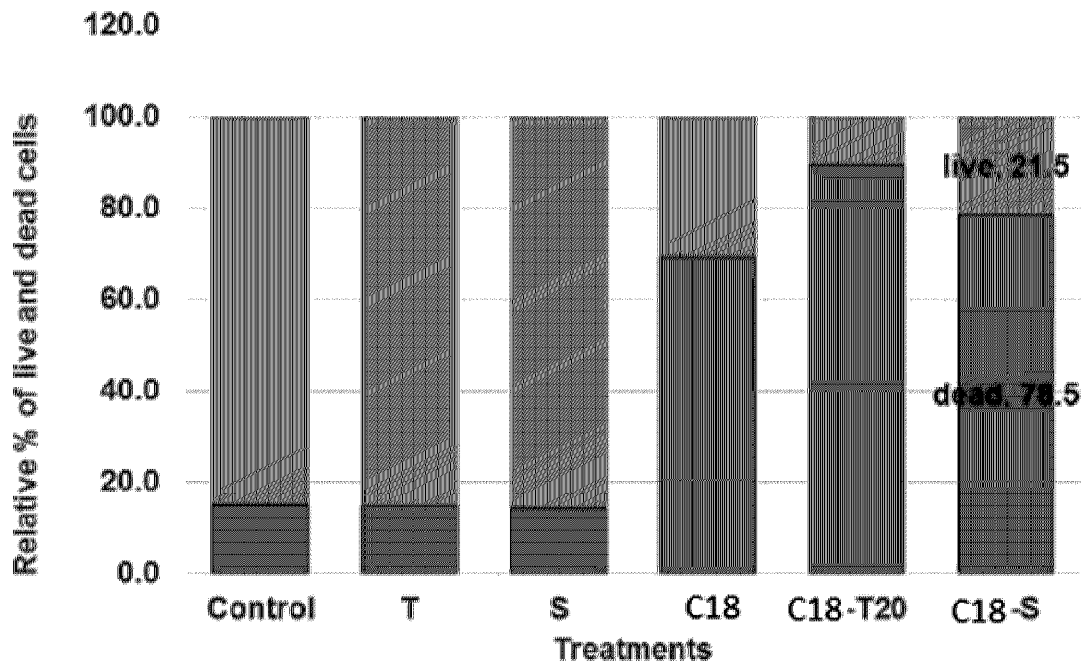


FIG. 18B

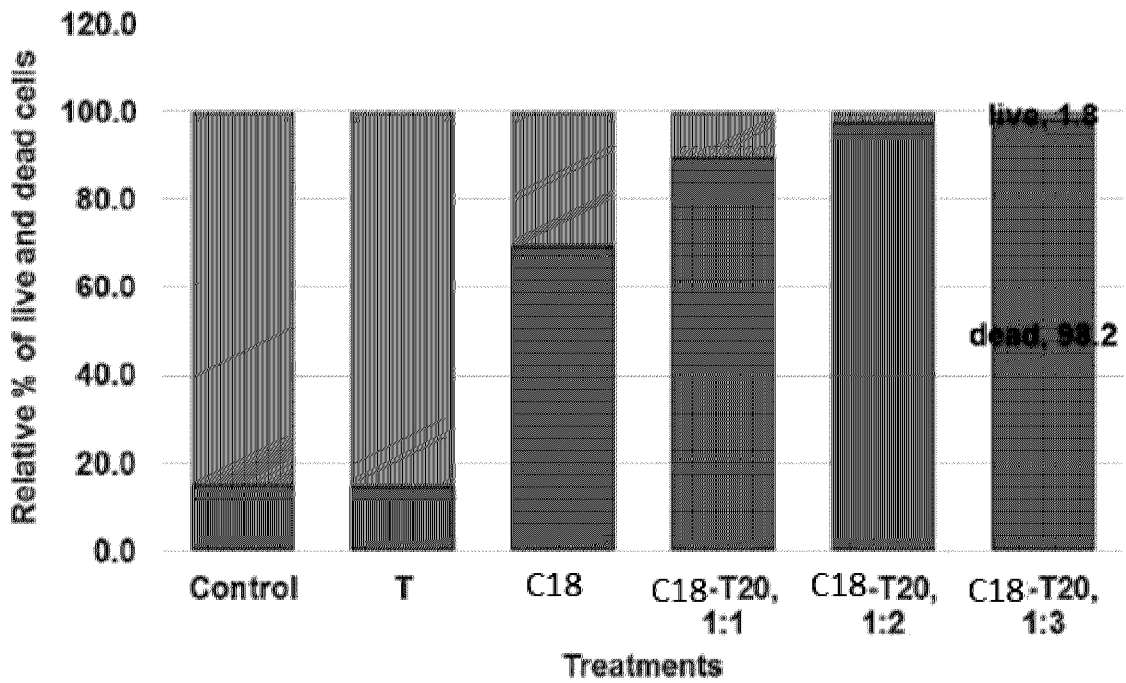
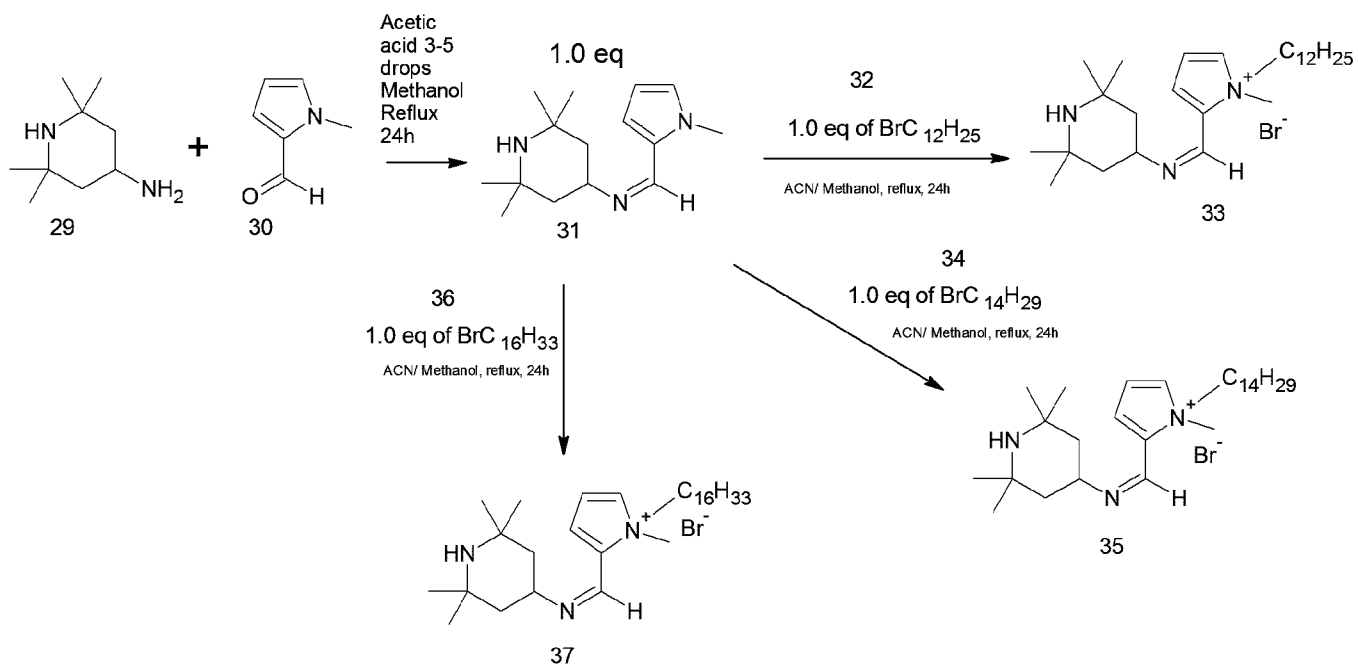
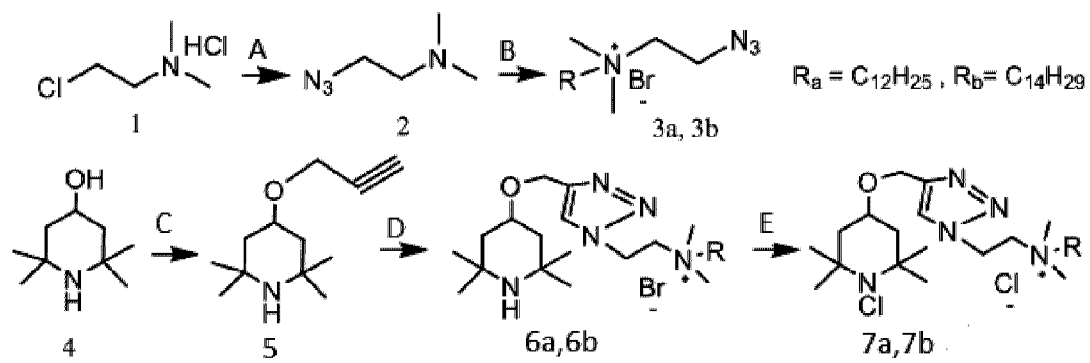


FIG. 18



Synthesis Reaction I

FIG. 19



Synthesis Reaction J

FIG. 20

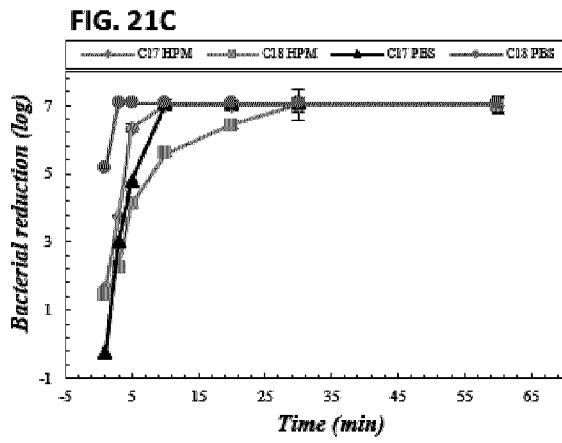
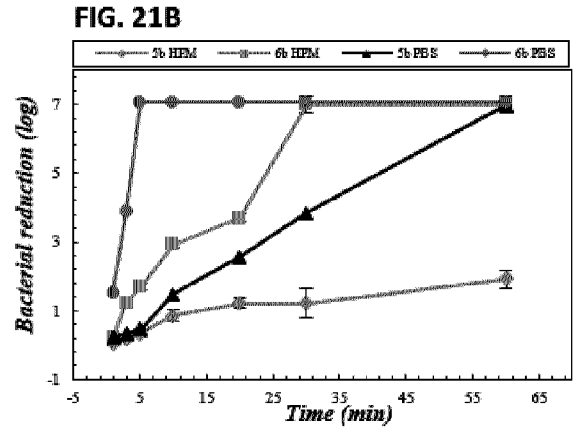
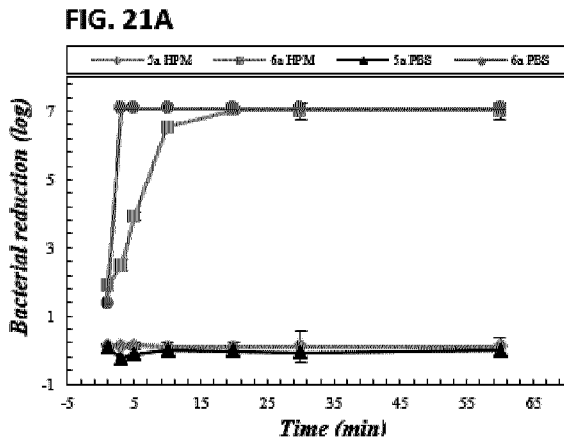


FIG. 21

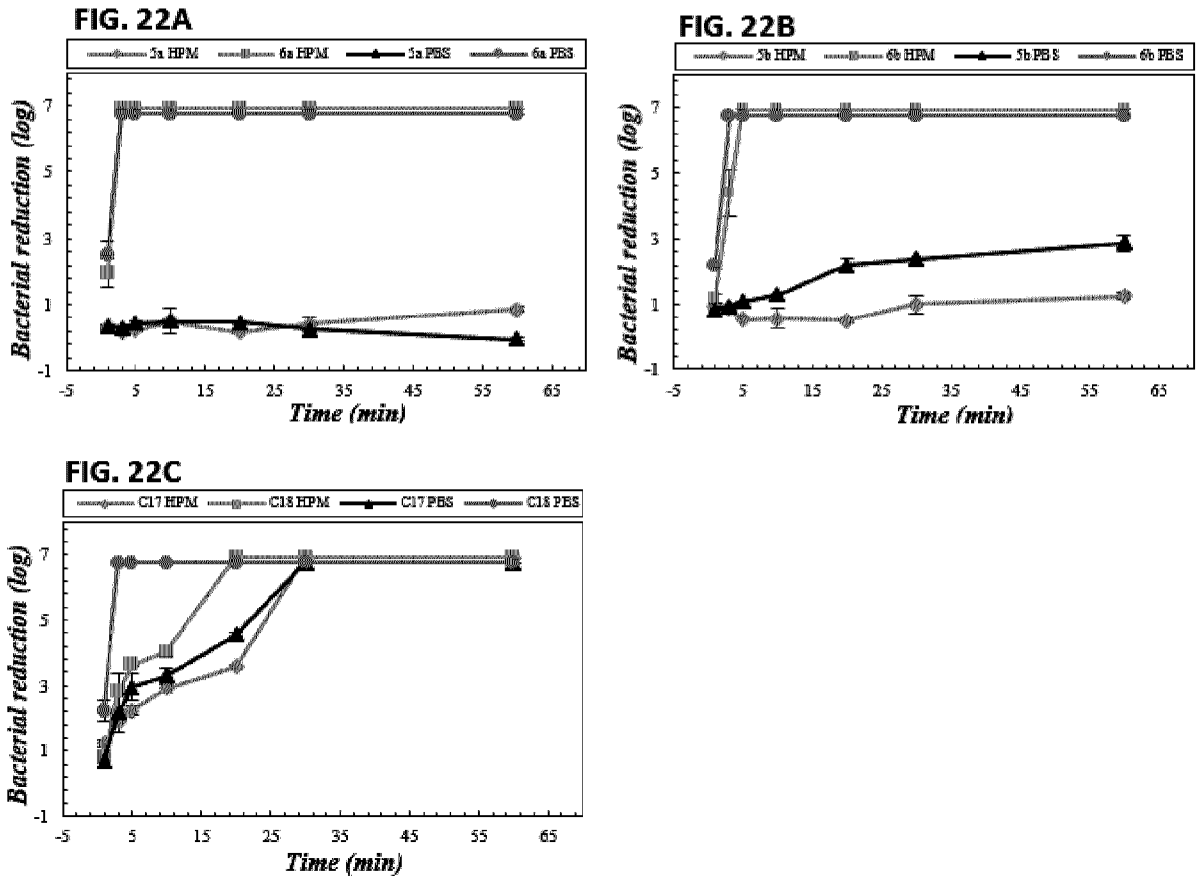


FIG. 22

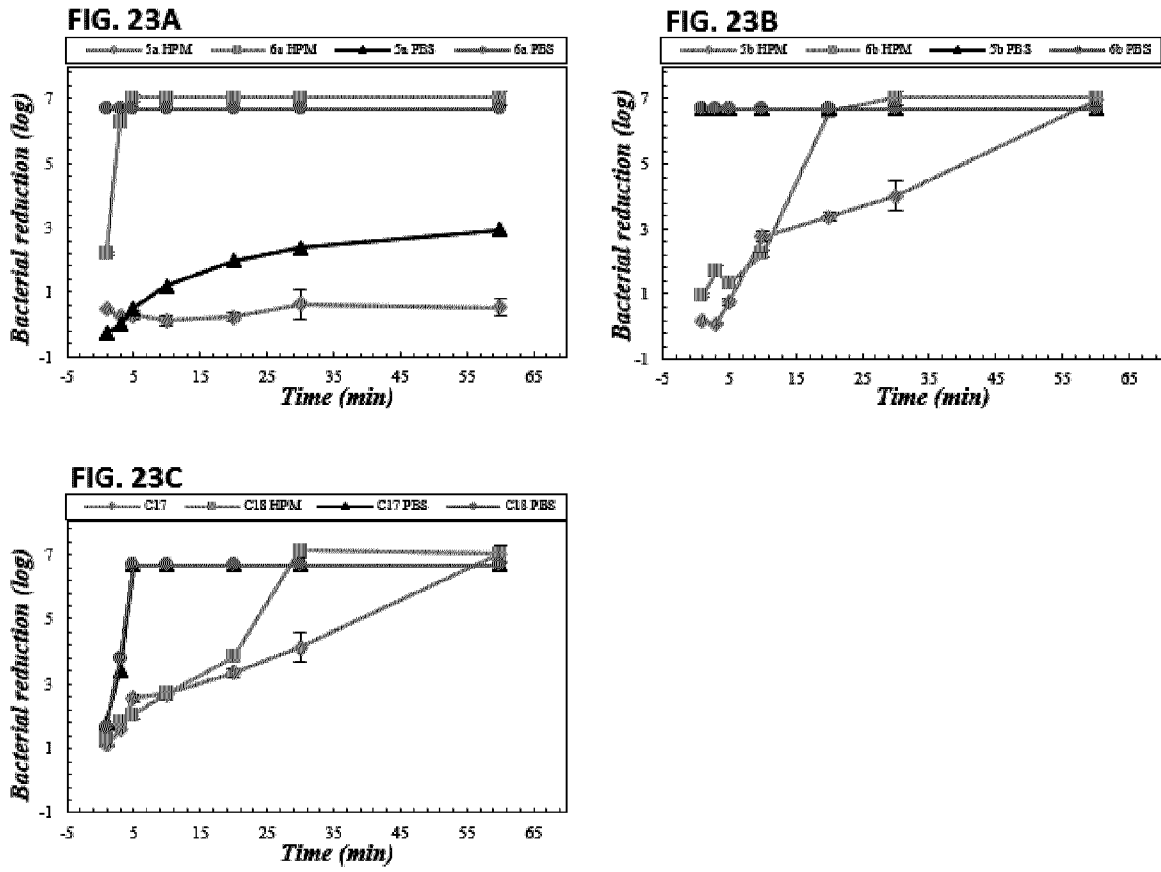


FIG. 23

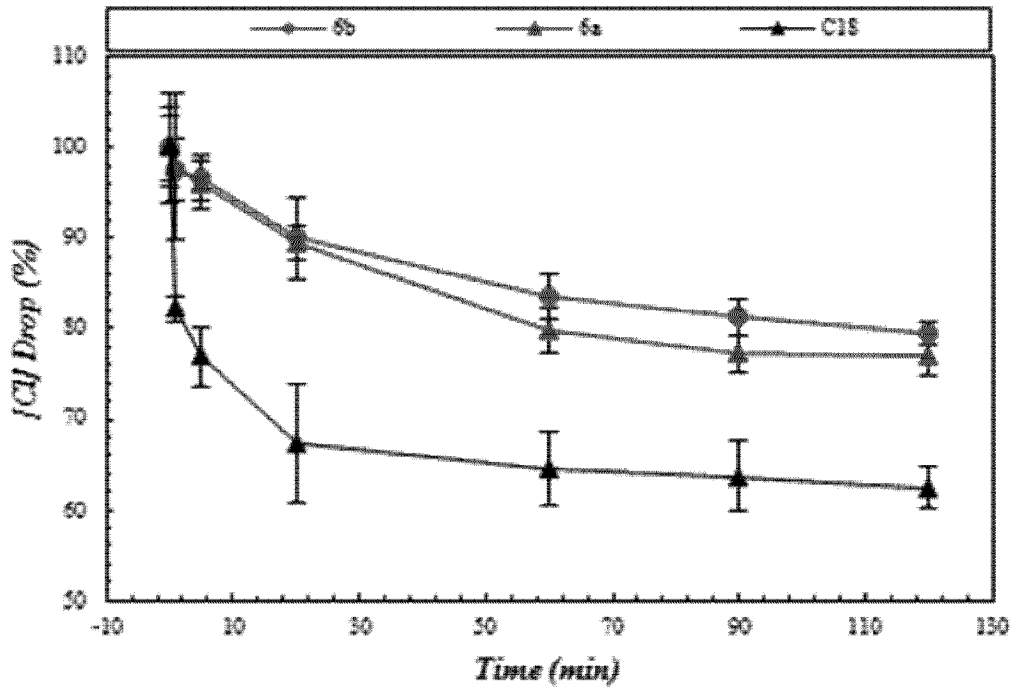


FIG. 24

INTERNATIONAL SEARCH REPORT

International application No.
PCT/CA2017/050598

A. CLASSIFICATION OF SUBJECT MATTER

IPC: **C07D 211/58** (2006.01), **A01N 43/40** (2006.01), **A01N 43/50** (2006.01), **A01N 43/647** (2006.01),
A01N 57/34 (2006.01), **A01P 1/00** (2006.01) (more IPCs on the last page)

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

C07D 211/58 (2006.01), **A01N 43/40** (2006.01), **A01N 43/50** (2006.01), **A01N 43/647** (2006.01), **A01N 57/34** (2006.01), **A01P 1/00** (2006.01)
A23L 3/3463 (2006.01), **A23L 3/3526** (2006.01), **A61L 2/18** (2006.01), **C07D 211/46** (2006.01),
C07D 211/72 (2006.01), **C07D 233/74** (2006.01), **C07D 401/12** (2006.01)

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic database(s) consulted during the international search (name of database(s) and, where practicable, search terms used)

STN Structure search; Questel (Exigence, Song, disinfectant, biocide, liquid disinfectant, halamine)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	CA2869634 A1 SONG, L. 28 November 2013 (28-11-2013)	1 and 4-13 (in part)
A	CA2814378 A1 JAIN, R. et al. 26 April 2012 (26-04-2012)	1 and 4-13 (in part)
A	US20100303930 A1 CAREY, T. C. et al. 02 December 2010 (02-12-2010)	1 and 4-13 (in part)
A	EP328697 A1 AKABANE, Y. et al. 23 August 1989 (23-08-1989)	1 and 4-13 (in part)

 Further documents are listed in the continuation of Box C. See patent family annex.

* Special categories of cited documents:	“T” later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
“A” document defining the general state of the art which is not considered to be of particular relevance	“X” document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
“E” earlier application or patent but published on or after the international filing date	“Y” document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
“L” document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	“&” document member of the same patent family
“O” document referring to an oral disclosure, use, exhibition or other means	
“P” document published prior to the international filing date but later than the priority date claimed	

Date of the actual completion of the international search
29 August 2017 (29-08-2017)Date of mailing of the international search report
08 September 2017 (8 8-09-2017)Name and mailing address of the ISA/CA
Canadian Intellectual Property Office
Place du Portage I, C114 - 1st Floor, Box PCT
50 Victoria Street
Gatineau, Quebec K1A 0C9
Facsimile No.: 819-953-2476

Authorized officer

Karla Randell (819) 635-5133

Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of the first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. Claim Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. Claim Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. Claim Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:
See extra sheet.

1. As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2. As all searchable claims could be searched without effort justifying additional fees, this Authority did not invite payment of additional fees.
3. As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claim Nos.:
4. No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claim Nos.:

Claims 1 and 4-13 (in part) as detailed in Group A.

- Remark on Protest**
- The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.
 - The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.
 - No protest accompanied the payment of additional search fees.

The claims are directed to a plurality of inventive concepts as follows:

Group A - Claims 1 and 4-13 (in part) are directed to compounds of formula 1a wherein L is nil and M is nitrogen or phosphorous, A is a substituted piperidine ring bound via an oxygen atom, G is a triazolyl group and Z is an alkyl chain, and their use as liquid disinfectants;

Group B - Claims 1, 2 and 4-13 (in part) are directed to compounds of formula 1a wherein L is nitrogen, M is nitrogen or phosphorous, A is a substituted piperidine ring bound via a carbon atom and G and Z are alkyl chains, and their use as liquid disinfectants;

Group C - Claims 1 and 4-13 (in part) are directed to compounds of formula 1a wherein L is nitrogen, M is nitrogen or phosphorous, A is a substituted hydantoin ring bound via a carbon atom and G and Z are alkyl chains, and their use as liquid disinfectants;

Group D - Claims 2 and 4-13 (in part) are directed to compounds of formula 1a wherein L is nitrogen M is nitrogen, A is a substituted piperidine ring bound via a carbon atom, G is an alkyl chain and R⁹ is C₁H_{2n}NH₂, and their use as liquid disinfectants;

Group E - Claims 3-13 (in part) are directed to compounds of formula C+D- and their use as liquid disinfectants.

The claims must be limited to one inventive concept as set out in PCT Rule 13.

A23L 3/3463 (2006.01), *A23L 3/3526* (2006.01), *A61L 2/18* (2006.01), *C07D 211/46* (2006.01),
C07D 211/72 (2006.01), *C07D 233/74* (2006.01), *C07D 401/12* (2006.01)

INTERNATIONAL SEARCH REPORT
Information on patent family members

International application No.
PCT/CA2017/050598

Patent Document Cited in Search Report	Publication Date	Patent Family Member(s)	Publication Date
CA2869634A1	28 November 2013 (28-11-2013)	CA2869634A1 AU2013265971A1 CN104968655A EP2850077A1 EP2850077A4 IN9443DEN2014A JP2015523331A MX2014013991A US2015118179A1 US2017204085A1 WO2013173905A1	28 November 2013 (28-11-2013) 18 December 2014 (18-12-2014) 07 October 2015 (07-10-2015) 25 March 2015 (25-03-2015) 26 August 2015 (26-08-2015) 17 July 2015 (17-07-2015) 13 August 2015 (13-08-2015) 06 August 2015 (06-08-2015) 30 April 2015 (30-04-2015) 20 July 2017 (20-07-2017) 28 November 2013 (28-11-2013)
CA2814378A1	26 April 2012 (26-04-2012)	CA2814378A1 AU2011317223A1 BR112013009279A2 CN103260402A EP2629607A2 EP2629607A4 IL225838D0 JP2013545728A KR20130123383A MX2013004311A SG189857A1 US2012129793A1 US9248117B2 WO2012054521A2 WO2012054521A3 WO2012054521A8	26 April 2012 (26-04-2012) 23 May 2013 (23-05-2013) 26 July 2016 (26-07-2016) 21 August 2013 (21-08-2013) 28 August 2013 (28-08-2013) 20 August 2014 (20-08-2014) 27 June 2013 (27-06-2013) 26 December 2013 (26-12-2013) 12 November 2013 (12-11-2013) 02 December 2013 (02-12-2013) 28 June 2013 (28-06-2013) 24 May 2012 (24-05-2012) 02 February 2016 (02-02-2016) 26 April 2012 (26-04-2012) 19 July 2012 (19-07-2012) 30 August 2012 (30-08-2012)
US2010303930A1	02 December 2010 (02-12-2010)	US2010303930A1 AR076932A1 TW201105369A UY32672A WO2010138852A2 WO2010138852A3	02 December 2010 (02-12-2010) 20 July 2011 (20-07-2011) 16 February 2011 (16-02-2011) 30 September 2010 (30-09-2010) 02 December 2010 (02-12-2010) 15 September 2011 (15-09-2011)
EP0328697A1	23 August 1989 (23-08-1989)	EP0328697A1 DE3731506A1 JPH093020A JPS649298A JPH0753520A JP2609996B2 JPS63270800A JPH0813996B2 US4820437A US4931562A	23 August 1989 (23-08-1989) 24 March 1988 (24-03-1988) 07 January 1997 (07-01-1997) 12 January 1989 (12-01-1989) 28 February 1995 (28-02-1995) 14 May 1997 (14-05-1997) 08 November 1988 (08-11-1988) 14 February 1996 (14-02-1996) 11 April 1989 (11-04-1989) 05 June 1990 (05-06-1990)