

COMPOSITIONS AND METHODS FOR TREATING LIBERIBACTER DISEASES AND OTHER BACTERIAL DISEASES

BACKGROUND

[0001] Huanglongbing (HLB), also called citrus greening, is one of the most devastating citrus plant diseases. This citrus plant disease causes multibillion-dollar loss annually in the United States alone. According to research from the Institute of Food and Agricultural Sciences at the University of Florida, Florida has lost approximately 162,200 acres of citrus plants and 7,513 jobs since detection of HLB in Florida in 2005. The most recent forecasts from the National Association of Academies of Science predicted that citrus production from 2016 to 2017 is approximately 70% lower than peak production levels from 1997 to 1998. Moreover, HLB also spreads rapidly in Texas and California. Recently, more than 400 confirmed cases of HLB-infected trees have been reported in southern California. HLB is caused by the phloem-limited Gram-negative bacteria of the *Liberibacter* species, *e.g.*, *Candidatus Liberibacter* species (*e.g.*, *Candidatus Liberibacter asiaticus* (*Ca. L. asiaticus*)), which is transmitted by insects of the Psyllidae family, *e.g.*, Asian citrus psyllids (ACP).

[0002] Another important disease cause by the *Liberibacter* species is Potato Zebra Chip (ZC) disease, also called Potato Zebra complex disease. ZC disease is associated with *Candidatus Liberibacter solanacearum* (*Ca. L. solanacearum*), which is transmitted by potato psyllids (*e.g.*, *Bactericera cockerelli*). ZC disease reached epidemic level in northern Texas in 2006 and has spread to Arizona, California, Colorado, Idaho, Oregon, Kansas, Nebraska, and New Mexico. ZC disease has caused millions of dollars loss to the potato industry in the southwestern United States, particularly Texas. In addition to potato, other solanaceous crops, including tomato, eggplant and pepper, can also be infected. There exists a need in the art for innovative compositions and methods to treat diseases in plants caused by *Liberibacter* species (*e.g.*, *Candidatus Liberibacter* species).

SUMMARY

[0003] The disclosure provides stable antimicrobial (*e.g.*, antibacterial or antifungal or both) peptides (SAMPs) that may be used in methods of preventing or treating bacterial diseases, such as those caused by Gram-negative bacteria, *e.g.*, a *Liberibacter* disease (*e.g.*, citrus greening disease (also called Huanglongbing (HLB)) or potato Zebra Chip disease) in plants (*e.g.*, citrus plants or potato plants). The SAMPs disclosed herein may be heat stable, as well

as stable in plant extracts and/or in plant lysates (*e.g.*, citrus lysates). The SAMPs described herein can effectively inhibit/kill different bacteria species, such as Gram-negative bacterial species, *e.g.*, *Liberibacter* species, for example, *Candidatus Liberibacter asiaticus* (*C.Las*) that infects citrus, *Candidatus Liberibacter solanacearum* that infects all solanaceous plants, and *Liberibacter crescens*, which is a culturable bacterium that infects papaya. *Liberibacter crescens* can be used as a surrogate for the unculturable *Candidatus Liberibacter* species (*e.g.*, *C.Las*). Furthermore, the SAMPs disclosed herein can also inhibit and kill other bacterial pathogens (*e.g.*, other Gram-negative bacterial pathogens), such as *Agrobacterium tumefaciens* (also known as *Rhizobium radiobacter*) and *Pseudomonas syringae* strains. *Agrobacterium tumefaciens* can cause crown gall disease or tumors in more than 140 eudicot species. Different strains of *Pseudomonas syringae* can cause bacterial canker or blast diseases on many dicot and monocot crops.

[0004] In one aspect, the disclosure features an isolated stable antimicrobial (*e.g.*, antibacterial or antifungal or both) peptide (SAMP) comprising a sequence that is substantially identical (*e.g.*, at least 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94% 95%, 96%, 97%, 98%, or 99% identical) to a sequence of any one of SEQ ID NOs:1-13 and 35-37. In some embodiments, the peptide comprises a sequence having at least 75% sequence identity (*e.g.*, at least 80%, 85%, 90%, 91%, 92%, 93%, 94% 95%, 96%, 97%, 98%, or 99% sequence identity) to a sequence of any one of SEQ ID NOs:1-13 and 35-37. In particular embodiments, the peptide comprises a sequence having at least 75% sequence identity (*e.g.*, at least 80%, 85%, 90%, 91%, 92%, 93%, 94% 95%, 96%, 97%, 98%, or 99% sequence identity) to a sequence of any one of SEQ ID NOs:1 and 2.

[0005] In another aspect, the disclosure features an isolated stable antimicrobial (*e.g.*, antibacterial) peptide (SAMP) comprising a sequence of

X₁GX₂X₃VSX₄ENX₅X₆QGF₇HX₈FEX₉TFX₁₀SX₁₁EGX₁₂AEYX₁₃X₁₄HPX₁₅HVEX₁₆ANX₁₇X₁₈LX₁₉X₂₀LEKX₂₁LX₂₂X₂₃DYKPX₂₄TX₂₅RV (SEQ ID NO:27), in which X₁ is R, K, or W; X₂ is K or E; X₃ is N or D; X₄ is T or I; X₅ is L, F, or R; X₆ is H or Q; X₇ is P or T; X₈ is I, L, or V; X₉ is S or F; X₁₀ is E or D; X₁₁ is T or L; X₁₂ is V or I; X₁₃ is V or I; X₁₄ is S, A, or D; X₁₅ is S, A, or V; X₁₆ is Y or F; X₁₇ is L or T; X₁₈ is F, M, or L; X₁₉ is A, P, or T; X₂₀ is N or Q; X₂₁ is V or F; X₂₂ is V or I; X₂₃ is V or I; X₂₄ is T, E, or Q; and X₂₅ is V, E, L.

[0006] In some embodiments of this aspect, the isolated SAMP comprising a sequence of SEQ ID NO:27, in which X₁ in SEQ ID NO:27 is R; X₂ is K; X₃ is N; X₄ is I; X₅ is L; X₆ is H;

X₁₀ is E; X₁₁ is T; X₁₂ is V; X₁₃ is V; X₁₆ is Y; X₁₇ is L; X₁₈ is F; X₁₉ is A; X₂₀ is N; X₂₁ is V; X₂₂ is V, X₂₃ is I; and X₂₄ is T.

[0007] In another aspect, the disclosure features an agricultural composition comprising an isolated stable antimicrobial (*e.g.*, antibacterial) peptide (SAMP) described herein. The agricultural composition may further comprise at least one of an herbicide, an herbicide safener, a surfactant, a fungicide, a pesticide, a nematocide, a plant activator, a synergist, a plant growth regulator, an insect repellant, an acaricide, a molluscicide, or a fertilizer.

[0008] In another aspect, the disclosure features a nucleic acid molecule encoding an isolated stable antimicrobial (*e.g.*, antibacterial) peptide (SAMP) described herein. Also provided is a polynucleotide comprising a promoter operably linked to the nucleic acid molecule (optionally where the promoter is heterologous to the nucleic acid molecule). The disclosure also features a cell comprising the nucleic acid molecule of the previous aspect. In some embodiments, the cell is a bacterial, yeast, plant, insect, or mammalian (*e.g.*, human) cell. In particular embodiments, the cell is a plant cell.

[0009] In another aspect, the disclosure features a plant comprising an isolated stable antimicrobial (*e.g.*, antibacterial) peptide (SAMP) described herein or the polynucleotide discussed above or the nucleic acid molecule encoding an isolated SAMP described herein. In some embodiments, the plant is a citrus plant or a solanaceous plant. In some embodiments, the plant is more tolerant to a bacterial pathogen compared to a control plant (otherwise identical) in which the SAMP is absent.

[0010] In another aspect, the disclosure features a plant comprising an *in situ* altered stable antimicrobial (*e.g.*, antibacterial) peptide (SAMP) comprising at least one amino acid substitution corresponding to an amino acid at any one of positions X₁ to X₂₅ as set forth in SEQ ID NO:27, wherein the mutated SAMP provides *Liberibacter* disease (*e.g.*, Huanglongbing (HLB)) resistance or *Liberibacter* disease (*e.g.*, HLB) tolerance, *Pseudomonas* disease (*e.g.*, bacterial canker or blast diseases) resistance or *Pseudomonas* disease tolerance, or *Agrobacterium* disease (*e.g.*, Crown Gall disease or tumors) resistance or *Agrobacterium* disease tolerance to the plant.

[0011] In another aspect, the disclosure features an expression cassette comprising a promoter operably linked to a polynucleotide encoding an isolated stable antimicrobial (*e.g.*, antibacterial) peptide (SAMP) described herein, wherein introduction of the expression

cassette into a plant results in the plant having enhanced *Liberibacter* disease (e.g., HLB) resistance or *Liberibacter* disease (e.g., HLB) tolerance, *Pseudomonas* disease (e.g., bacterial canker or blast diseases) resistance or *Pseudomonas* disease tolerance, or *Agrobacterium* disease (e.g., Crown Gall disease or tumors) resistance or *Agrobacterium* disease tolerance. In some embodiments, the promoter of the expression cassette is heterologous to the polynucleotide. In some embodiments, the promoter is inducible. In some embodiments, the promoter is plant tissue-specific (e.g., phloem-specific, tuber-specific, root-specific, stem-specific, trunk-specific, or leaf-specific). In some embodiments, the phloem-specific promoter is the sucrose transporter protein SUC2 promoter.

[0012] In another aspect, the disclosure features a transgenic plant comprising the expression cassette of the previous aspect, wherein the plant has enhanced *Liberibacter* disease (e.g., HLB) resistance or *Liberibacter* disease (e.g., HLB) tolerance, *Pseudomonas* disease (e.g., bacterial canker or blast diseases) resistance or *Pseudomonas* disease tolerance, or *Agrobacterium* disease (e.g., Crown Gall disease or tumors) resistance or *Agrobacterium* disease tolerance, as compared to a control plant lacking the expression cassette. In some embodiments, the transgenic plant is a citrus plant or a solanaceous plant.

[0013] In another aspect, the disclosure features a method of preventing or treating a *Liberibacter* disease (e.g., HLB), a *Pseudomonas* disease (e.g., bacterial canker or blast diseases), or an *Agrobacterium* disease (e.g., Crown Gall disease or tumors) in a plant by contacting the plant with an isolated stable antimicrobial (e.g., antibacterial) peptide (SAMP) described herein or an agricultural composition comprising an isolated SAMP. In some embodiments of these aspects, the isolated peptide or agricultural composition is injected into the trunk of the plant. In some embodiments, the isolated peptide or agricultural composition is injected into the stem of the plant. In other embodiments, the isolated peptide or agricultural composition is foliar sprayed onto the plant. In other embodiments, the isolated peptide or agricultural composition is applied to the root of the plant. In other embodiments, the isolated peptide or agricultural composition is applied to the plant by dripping irrigation to the roots. In other embodiments, the isolated peptide or agricultural composition is applied by laser ablation.

[0014] In another aspect, the disclosure features a method of preventing or treating potato Zebra Chip (ZC) disease in a plant by contacting the plant with an isolated stable antimicrobial (e.g., antibacterial) peptide (SAMP) described herein or an agricultural composition comprising an isolated SAMP. In yet another aspect, the disclosure features a method of preventing or treating a bacterial infection in a plant caused by bacteria in the genus

Liberibacter (e.g., *Candidatus Liberibacter* species or *Liberibacter crescens*) by contacting the plant with an isolated SAMP described herein or an agricultural composition comprising an isolated SAMP. In still another aspect, the disclosure features a method of preventing or treating a bacterial infection in a plant caused by bacteria in the genus *Agrobacterium* (e.g., *Agrobacterium tumefaciens* species) by contacting the plant with an isolated SAMP described herein or an agricultural composition comprising an isolated SAMP. In still yet another aspect, the disclosure features a method of preventing or treating a bacterial infection in a plant caused by bacteria in the genus *Pseudomonas* (e.g., *Pseudomonas syringae* species) by contacting the plant with an isolated SAMP described herein or an agricultural composition comprising an isolated SAMP.

[0015] In another aspect, the disclosure features a method of inhibiting the growth of bacteria or killing bacteria in the genus *Liberibacter* (e.g., *Candidatus Liberibacter* species or *Liberibacter crescens*) in a plant by contacting the plant with an isolated stable antimicrobial (e.g., antibacterial) peptide (SAMP) described herein or an agricultural composition comprising an isolated SAMP. In yet another aspect, the disclosure features a method of inhibiting the growth of bacteria or killing bacteria in the genus *Agrobacterium* (e.g., *Agrobacterium tumefaciens* species) in a plant by contacting the plant with an isolated stable antimicrobial (e.g., antibacterial) peptide (SAMP) described herein or an agricultural composition comprising an isolated SAMP. In still yet another aspect, the disclosure features a method of inhibiting the growth of bacteria or killing bacteria in the genus *Pseudomonas* (e.g., *Pseudomonas syringae* species) in a plant by contacting the plant with an isolated stable antimicrobial (e.g., antibacterial) peptide (SAMP) described herein or an agricultural composition comprising an isolated SAMP.

[0016] In another aspect, the disclosure features a method of preventing or treating a *Liberibacter* disease (e.g., HLB) in a plant or preventing or treating a bacterial infection in a plant caused by bacteria in the genus *Liberibacter* (e.g., *Candidatus Liberibacter* species (e.g., *Candidatus Liberibacter asiaticus*, *Candidatus Liberibacter africanus*, and *Candidatus Liberibacter americanus*) or *Liberibacter crescens*) by introducing an expression cassette described herein (e.g., an expression cassette comprising a promoter operably linked to a polynucleotide encoding an isolated stable antimicrobial (e.g., antibacterial) peptide described herein) into the plant. In another aspect, the disclosure features a method of preventing or treating potato ZC disease in a plant or preventing or treating a bacterial infection in a plant caused by bacteria in the genus *Liberibacter* (e.g., *Candidatus Liberibacter* species (e.g.,

Candidatus Liberibacter solanacearum (Ca. *L. solanacearum*)) or *Liberibacter crescens*) by introducing an expression cassette described herein (e.g., an expression cassette comprising a promoter operably linked to a polynucleotide encoding an isolated SAMP described herein) into the plant.

[0017] In yet another aspect, the disclosure features a method of preventing or treating a *Pseudomonas* disease (e.g., bacterial canker or blast diseases) in a plant or preventing or treating a bacterial infection in a plant caused by bacteria in the genus *Pseudomonas* (e.g., *Pseudomonas syringae* species) by introducing an expression cassette described herein (e.g., an expression cassette comprising a promoter operably linked to a polynucleotide encoding an isolated stable antimicrobial (e.g., antibacterial) peptide described herein) into the plant.

[0018] In still yet another aspect, the disclosure features a method of preventing or treating a *Agrobacterium* disease (e.g., Crown Gall disease or tumors) in a plant or preventing or treating a bacterial infection in a plant caused by bacteria in the genus *Agrobacterium* (e.g., *Agrobacterium tumefaciens* species) by introducing an expression cassette described herein (e.g., an expression cassette comprising a promoter operably linked to a polynucleotide encoding an isolated stable antimicrobial (e.g., antibacterial) peptide described herein) into the plant.

[0019] In another aspect, the disclosure features a method of inhibiting the growth of bacteria or killing bacteria in the genus *Liberibacter* (e.g., *Candidatus Liberibacter* species or *Liberibacter crescens*) in a plant by introducing an expression cassette described herein (e.g., an expression cassette comprising a promoter operably linked to a polynucleotide encoding an isolated stable antimicrobial (e.g., antibacterial) peptide described herein) into the plant.

[0020] In another aspect, the disclosure features a method of inhibiting the growth of bacteria or killing bacteria in the genus *Agrobacterium* (e.g., *Agrobacterium tumefaciens* species) in a plant by introducing an expression cassette described herein (e.g., an expression cassette comprising a promoter operably linked to a polynucleotide encoding an isolated stable antimicrobial (e.g., antibacterial) peptide described herein) into the plant.

[0021] In yet another aspect, the disclosure features a method of inhibiting the growth of bacteria or killing bacteria in the genus *Pseudomonas* (e.g., *Pseudomonas syringae* species) in a plant by introducing an expression cassette described herein (e.g., an expression cassette

comprising a promoter operably linked to a polynucleotide encoding an isolated stable antimicrobial (*e.g.*, antibacterial) peptide described herein) into the plant.

[0022] In another aspect, the disclosure features a method of producing a plant having enhanced *Liberibacter* disease (*e.g.*, HLB) resistance or *Liberibacter* disease (*e.g.*, HLB) tolerance by introducing an isolated stable antimicrobial (*e.g.*, antibacterial) peptide described herein or an expression cassette comprising a promoter operably linked to a polynucleotide encoding an isolated stable antimicrobial (*e.g.*, antibacterial) peptide into a plurality of plants; and selecting a plant that comprises the isolated peptide or expresses the polynucleotide from the plurality of plants.

[0023] In another aspect, the disclosure features a method of producing a plant having enhanced potato ZC disease resistance or potato ZC disease tolerance by introducing an isolated stable antimicrobial (*e.g.*, antibacterial) peptide (SAMP) described herein or an expression cassette comprising a promoter operably linked to a polynucleotide encoding an isolated SAMP into a plurality of plants; and selecting a plant that comprises the isolated peptide or expresses the polynucleotide from the plurality of plants.

[0024] In another aspect, the disclosure features a method of producing a plant having enhanced *Pseudomonas* disease (*e.g.*, bacterial canker or blast diseases) resistance or *Pseudomonas* disease (*e.g.*, bacterial canker or blast diseases) tolerance by introducing an isolated stable antimicrobial (*e.g.*, antibacterial) peptide described herein or an expression cassette comprising a promoter operably linked to a polynucleotide encoding an isolated stable antimicrobial (*e.g.*, antibacterial) peptide into a plurality of plants; and selecting a plant that comprises the isolated peptide or expresses the polynucleotide from the plurality of plants.

[0025] In another aspect, the disclosure features a method of producing a plant having enhanced *Agrobacterium* disease (*e.g.*, Crown Gall disease or tumors) resistance or *Agrobacterium* disease (*e.g.*, Crown Gall disease or tumors) tolerance by introducing an isolated stable antimicrobial (*e.g.*, antibacterial) peptide described herein or an expression cassette comprising a promoter operably linked to a polynucleotide encoding an isolated stable antimicrobial (*e.g.*, antibacterial) peptide into a plurality of plants; and selecting a plant that comprises the isolated peptide or expresses the polynucleotide from the plurality of plants.

[0026] In another aspect, the disclosure features a method of producing a plant having enhanced *Liberibacter* disease (*e.g.*, HLB) resistance or *Liberibacter* disease (*e.g.*, HLB)

tolerance (*i.e.*, enhanced resistance or tolerance to a bacterial infection caused by bacteria in the genus *Liberibacter* (*e.g.*, *Candidatus Liberibacter* species (*e.g.*, *Candidatus Liberibacter asiaticus*, *Candidatus Liberibacter africanus*, and *Candidatus Liberibacter americanus*) or *Liberibacter crescens*)) by introducing a mutation into a polynucleotide in the plant, wherein the mutated polynucleotide encodes an isolated stable antimicrobial (*e.g.*, antibacterial) peptide described herein (*e.g.*, an isolated SAMP having at least 75% sequence identity to the sequence of SEQ ID NO:1 or 2). In another aspect, the disclosure features a method of producing a plant having enhanced potato ZC disease resistance or potato ZC disease tolerance (*i.e.*, enhanced resistance or tolerance to a bacterial infection caused by bacteria in the genus *Liberibacter* (*e.g.*, *Candidatus Liberibacter* species (*e.g.*, *Candidatus Liberibacter solanacearum* (*Ca. L. solanacearum*)) or *Liberibacter crescens*)) by introducing a mutation into a polynucleotide in the plant, wherein the mutated polynucleotide encodes an isolated stable antimicrobial (*e.g.*, antibacterial) peptide (SAMP) described herein (*e.g.*, an isolated SAMP having at least 75% sequence identity to the sequence of SEQ ID NO:1 or 2). In some embodiments of these aspects, the introducing occurs *in situ* in the genome of a plant cell. In particular embodiments, the introducing comprises clustered regularly interspaced short palindromic repeats (CRISPR)/Cas genome editing. In some embodiments of these aspects, the plant is a citrus plant or a solanaceous plant (*e.g.*, a potato plant).

[0027] In still another aspect, the disclosure features a method of producing a plant having enhanced *Agrobacterium* disease resistance or *Agrobacterium* disease tolerance (*i.e.*, enhanced resistance or tolerance to a bacterial infection caused by bacteria in the genus *Agrobacterium* (*e.g.*, Crown Gall disease or tumors caused by *Agrobacterium* strains)) by introducing a mutation into a polynucleotide in the plant, wherein the mutated polynucleotide encodes an isolated stable antimicrobial (*e.g.*, antibacterial) peptide (SAMP) described herein (*e.g.*, an isolated SAMP having at least 75% sequence identity to the sequence of SEQ ID NO:1 or 2). In some embodiments of these aspects, the introducing occurs *in situ* in the genome of a plant cell. In particular embodiments, the introducing comprises clustered regularly interspaced short palindromic repeats (CRISPR)/Cas genome editing. In some embodiments of these aspects, the plant is eudicot plant.

[0028] In still another aspect, the disclosure features a method of producing a plant having enhanced *Pseudomonas* disease resistance or *Pseudomonas* disease tolerance (*i.e.*, enhanced resistance or tolerance to a bacterial infection caused by bacteria in the genus *Pseudomonas* (*e.g.*, bacterial canker or blast diseases caused by *Pseudomonas* strains)) by introducing a

mutation into a polynucleotide in the plant, wherein the mutated polynucleotide encodes an isolated stable antimicrobial (*e.g.*, antibacterial) peptide (SAMP) described herein (*e.g.*, an isolated SAMP having at least 75% sequence identity to the sequence of SEQ ID NO:1 or 2). In some embodiments of these aspects, the introducing occurs *in situ* in the genome of a plant cell. In particular embodiments, the introducing comprises clustered regularly interspaced short palindromic repeats (CRISPR)/Cas genome editing. In some embodiments of these aspects, the plant is a monocot or dicot plant (*e.g.*, a tomato plant).

[0029] In any of the compositions or methods described in the present disclosure, the plant may species be from the genus *Citrus* (*e.g.*, *Citrus maxima*, *Citrus medica*, *Citrus micrantha*, *Citrus reticulata*, *Citrus aurantiifolia*, *Citrus aurantium*, *Citrus latifolia*, *Citrus limon*, *Citrus limonia*, *Citrus paradise*, *Citrus sinensis*, and *Citrus tangerine*) or species from the family Solanaceae (*e.g.*, *Solanum* spp., *Capsicum* spp., and *Nicotiana* spp.). Species from the genus *Solanum* include, *e.g.*, *Solanum tuberosum*, *Solanum lycopersicum*, *Solanum melongena*, *Solanum aviculare*, *Solanum capsicastrum*, *Solanum crispum*, *Solanum laciniatum*, *Solanum laxum*, *Solanum pseudocapsicum*, *Solanum rantonnetii*, *Solanum seaforthianum*, and *Solanum wendlandii*. Species from the genus *Capsicum* include, *e.g.*, *Capsicum annuum*, *Capsicum baccatum*, *Capsicum campylopodium*, *Capsicum cardenasii*, *Capsicum chacoense*, *Capsicum cornutum*, *Capsicum dusenii*, *Capsicum eximium*, *Capsicum friburgense*, *Capsicum frutescens*, *Capsicum geminifolium*, *Capsicum havanense*, , *Capsicum lanceolatum*, *Capsicum lycianthoides*, *Capsicum minutiflorum*, *Capsicum mositicum*, *Capsicum pubescens*, *Capsicum recurvatum*, *Capsicum schottianum*, *Capsicum spina-alba*, *Capsicum tovarii*, and *Capsicum villosum*. Species from the genus *Nicotiana* include, *e.g.*, *Nicotiana acuminata*, *Nicotiana benthamiana*, *Nicotiana glauca*, *Nicotiana longiflora*, *Nicotiana rustica*, *Nicotiana tabacum*, and *Nicotiana occidentalis*.

[0030] In particular embodiments, the plant is selected from the group consisting of *Citrus reticulata*, *Citrus sinensis*, *Citrus clementina*, *Capsicum annuum*, *Solanum tuberosum*, *Solanum lycopersicum*, *Solanum melongena*, and *Nicotiana benthamiana*. In particular embodiments, the plant is a sweet orange plant (*Citrus sinensis*). In particular embodiments, the plant is a clementine plant (*Citrus clementina*). In particular embodiments, the plant is a potato plant (*Solanum tuberosum*). In some embodiment, the plant is a vegetable- or fruit-producing plant.

[0031] In any of the aspects of the disclosure described herein, in some embodiments, the SAMP is a heat stable (HS) peptide.

[0032] Furthermore, in any of the aspects of the disclosure described herein, in some embodiments, the SAMP may also provide resistance or tolerance to bacterial diseases caused by other bacterial pathogens, such as *Agrobacterium tumefaciens* (also known as *Rhizobium radiobacter*) and *Pseudomonas syringae*.

Definitions

[0033] As used herein, the term “*Liberibacter* disease” refers to a disease, such as an infection, caused by bacteria in the genus *Liberibacter* (e.g., *Candidatus Liberibacter* species or *Liberibacter crescens*). A *Liberibacter* disease may infect plants such as citrus plants (e.g., orange, grapefruit, tangerine, lemon, lime, key lime, papaya, citron, and pomelo) and solanaceous plants (e.g., potato, tomato, eggplant, and pepper). Huanglongbing (HLB) is a type of *Liberibacter* disease that infects citrus plants.

[0034] As used herein, the terms “citrus greening disease” and “Huanglongbing (HLB)” refer to a bacterial infection of plants (e.g., citrus plants) caused by bacteria in the genus *Liberibacter* (e.g., *Candidatus Liberibacter* species (e.g., *Candidatus Liberibacter asiaticus*, *Candidatus Liberibacter africanus*, and *Candidatus Liberibacter americanus*) or *Liberibacter crescens*). The infection is vectored and transmitted by the Asian citrus psyllid, *Diaphorina citri*, and the African citrus psyllid, *Trioza erytreae*. Three different types of HLB are currently known: the heat-tolerant Asian form, and the heat-sensitive African and American forms.

[0035] As used herein, the term “Potato Zebra Chip (ZC) disease” refers to a bacterial infection of plants (e.g., potato plants) caused by bacteria in the genus *Liberibacter* (e.g., *Candidatus Liberibacter* species (e.g., *Candidatus Liberibacter solanacearum* (Ca. *L. solanacearum*)) or *Liberibacter crescens*). The infection is vectored and transmitted by potato psyllids (e.g., *Bactericera cockerelli*).

[0036] As used herein, the term “*Agrobacterium* disease” refers to a disease, such as an infection, caused by bacteria in the genus *Agrobacterium* (e.g., *Agrobacterium tumefaciens* species, also known as *Rhizobium radiobacter*). *Agrobacterium* diseases can comprise Crown Gall disease, or tumors, in more than 140 eudicot species.

[0037] As used herein, the term “*Pseudomonas* disease” refers to a disease, such as an infection, caused by bacteria in the genus *Pseudomonas* (e.g., *Pseudomonas syringae* species). *Pseudomonas* diseases can comprise bacterial canker or blast diseases on many dicot and

monocot crops (e.g., Tomato Bacterial Speck, Tomato Bacterial Spot, and Tomato Bacterial Canker).

[0038] As used herein, the term “disease resistance” refers to the ability of a plant to not be affected by a *Liberibacter* disease (e.g., HLB), *Agrobacterium* disease, or *Pseudomonas* disease; or infection by *Liberibacter* bacteria (e.g., *Candidatus Liberibacter* species or *Liberibacter crescens*), *Agrobacterium* bacteria (e.g., *Agrobacterium tumefaciens* species), or *Pseudomonas* bacteria (e.g., *Pseudomonas syringae* species).

[0039] As used herein, the term “disease tolerance” refers to the ability of a plant to continuously grow and survive despite being infected by bacteria (e.g., gram-negative bacteria, such as *Liberibacter* bacteria, *Agrobacterium* bacteria or *Pseudomonas* bacteria).

[0040] As used herein, the term “*Liberibacter* disease resistance” refers to the ability of a plant to not be affected by a *Liberibacter* disease (e.g., HLB) or infection by *Liberibacter* bacteria (e.g., *Candidatus Liberibacter* species or *Liberibacter crescens*).

[0041] As used herein, the term “*Liberibacter* disease tolerance” refers to the ability of a plant to continuously grow and survive despite being infected by *Liberibacter* bacteria (e.g., *Candidatus Liberibacter* species or *Liberibacter crescens*) or having a *Liberibacter* disease (e.g., HLB). In some embodiments, a plant with a *Liberibacter* disease (e.g., HLB) may show minor symptoms of the disease, such as yellowing of leaves, blotchy mottle of the leaves, zinc-deficiency-like mottle, chlorosis, and reduced fruit yield, but is still able to grow or produce fruit despite the infection.

[0042] As used herein, the term “potato ZC disease resistance” refers to the ability of a plant to not be affected by potato ZC disease or infection by *Liberibacter* (e.g., *Candidatus Liberibacter* species (e.g., *Candidatus Liberibacter solanacearum* (Ca. *L. solanacearum*)) or *Liberibacter crescens*) bacteria.

[0043] As used herein, the term “potato ZC disease tolerance” refers to the ability of a plant to continuously grow and survive despite being infected by *Liberibacter* (e.g., *Candidatus Liberibacter* species (e.g., *Candidatus Liberibacter solanacearum* (Ca. *L. solanacearum*)) or *Liberibacter crescens*) bacteria or having potato ZC disease. In some embodiments, a plant with potato ZC disease may show minor symptoms of potato ZC disease, such as chlorosis, leaf scorching, swollen nodes, vascular tissue browning, curled leaves, collapsed stolons,

enlarged lenticels, vascular tissue browning, medullary ray discoloration, and necrotic flecking of tuber tissue, but is still able to grow or produce potato despite the infection.

[0044] As used herein, the term “*Agrobacterium* disease resistance” refers to the ability of a plant to not be affected by an *Agrobacterium* disease (e.g., Crown Gall disease) or infection by *Agrobacterium* bacteria (e.g., *Agrobacterium tumefaciens* species, also known as *Rhizobium radiobacter*).

[0045] As used herein, the term “*Agrobacterium* disease tolerance” refers to the ability of a plant to continuously grow and survive despite being infected by *Agrobacterium* bacteria (e.g., *Agrobacterium tumefaciens* species, also known as *Rhizobium radiobacter*) or having an *Agrobacterium* disease (e.g., Crown Gall disease).

[0046] As used herein, the term “*Pseudomonas* disease resistance” refers to the ability of a plant to not be affected by a *Pseudomonas* disease (e.g., Tomato Bacterial Speck, Tomato Bacterial Spot, and Tomato Bacterial Canker) or infection by *Pseudomonas* bacteria (e.g., *Agrobacterium tumefaciens* species).

[0047] As used herein, the term “*Pseudomonas* disease tolerance” refers to the ability of a plant to continuously grow and survive despite being infected by *Pseudomonas* bacteria (e.g., *Agrobacterium tumefaciens* species) or having a *Pseudomonas* disease (e.g., Tomato Bacterial Speck, Tomato Bacterial Spot, and Tomato Bacterial Canker).

[0048] As used herein, the term “stable antimicrobial peptides” or “SAMPs” refers to peptides identified in plants that are *Liberibacter* disease-resistant/tolerant (e.g., HLB-resistant/tolerant). Such peptides are expressed at a higher level in *Liberibacter* disease-resistant/tolerant (e.g., HLB-resistant/tolerant) plants than *Liberibacter* disease-susceptible (e.g., HLB-susceptible) plants. These SAMPs may be injected into plants to prevent or treat a *Liberibacter* disease (e.g., HLB). In some embodiments, the isolated SAMPs disclosed herein have antibacterial or antifungal or both properties. In some embodiments, the isolated SAMPs disclosed herein are heat stable (e.g., heat stable (HS) peptides). In some embodiments, the isolated SAMPs disclosed herein are also stable in plant extracts. In further embodiments, the isolated SAMPs disclosed herein are also stable in plant lysates (e.g., citrus lysates).

[0049] As used herein, the term “agricultural composition” refers to a composition formulated for application to a plant or plant part (e.g., seed, cutting, shoots, etc.). An agricultural composition is typically in liquid form, e.g., for application by spraying or soaking,

but can be in a powder for rehydration or application (dusting or dry coating), or gaseous form (*e.g.*, for enclosed environments). The agricultural composition can be concentrated, *e.g.*, for dilution or water or other solvent. An agricultural composition can also include more than one active ingredient, *e.g.*, a SAMP (*e.g.*, an HS peptide) described herein, alone or in combination with a fungicide, herbicide, fertilizer, etc.

[0050] As used herein, the term “treat” or “treating” a *Liberibacter* disease (*e.g.*, an HLB or a potato ZC disease) in plants refers to the reduction or eradication of symptoms caused by the *Liberibacter* disease by methods described herein. Symptoms of HLB include, but are not limited to, yellowing of leaves, blotchy mottle of the leaves, zinc-deficiency-like mottle, severe chlorosis, and reduced fruit yield. Symptoms of potato ZC disease include, but are not limited to, chlorosis, leaf scorching, swollen nodes, vascular tissue browning, curled leaves, collapsed stolons, enlarged lenticels, vascular tissue browning, medullary ray discoloration, and necrotic flecking of tuber tissue. In some embodiments, the disclosed methods may not necessarily result in eradication or cure of the *Liberibacter* disease (*e.g.*, HLB or potato ZC disease), but can significantly reduce the symptoms caused by the disease.

[0051] As used herein, the term “prevent” or “preventing” a *Liberibacter* disease (*e.g.*, an HLB or a potato ZC disease) in plants refers protecting a plant that is at risk for the disease from developing the disease, or decreasing the risk that a plant may develop the disease. A plant may be contacted with a SAMP (*e.g.*, an HS peptide) described herein before the plant develops the disease, or shows signs of the disease.

[0052] As used herein, the term “plant” includes whole plants, shoot vegetative organs/structures (*e.g.*, leaves, stems and tubers), roots, flowers and floral organs/structures (*e.g.*, bracts, sepals, petals, stamens, carpels, anthers and ovules), seed (including embryo, endosperm, and seed coat) and fruit (the mature ovary), plant tissue (*e.g.*, vascular tissue, ground tissue, and the like) and cells (*e.g.*, guard cells, egg cells, and the like), and progeny of same. Plants that can be treated as described herein include, *e.g.*, citrus plants (*e.g.*, orange, grapefruit, tangerine, lemon, lime, key lime, papaya, citron, and pomelo) and solanaceous plants (*e.g.*, potato, tomato, eggplant, and pepper).

[0053] The term “plant” also includes naturally occurring mutants and genetically modified plants. A “genetically modified plant” or “transgenic plant” refers to one whose genome has been manipulated so that it is different than a wild-type plant of the same species, variety or cultivar, *e.g.*, to add a gene or genetic element, remove a gene or genetic element, mutate a gene or genetic element, change chromatin structure, change gene or protein expression levels,

etc. A transgenic plant may contain an expression vector or cassette. The expression cassette typically comprises a polypeptide-encoding sequence or a modulating nucleic acid (*e.g.*, an antisense, an siRNA or ribozyme) operably linked (*i.e.*, under regulatory control of) to an appropriate inducible or constitutive regulatory sequences that allow for the expression of a polypeptide or modulating nucleic acid. The expression cassette can be introduced into a plant by transformation or by breeding after transformation of a parent plant. Such methods can be used in a whole plant, including seedlings and mature plants, as well as to a plant part, such as seed, fruit, leaf, or root, plant tissue, plant cells or any other plant material, *e.g.*, a plant explant, as well as to progeny thereof, and to *in vitro* systems that mimic biochemical or cellular components or processes in a cell. In the context of the present disclosure, genetically modified plants may include genetic modifications in a gene encoding a SAMP (*e.g.*, an HS peptide). In some embodiments, the modified gene in the genetically modified plant may encode a SAMP (*e.g.*, an HS peptide) described herein (*e.g.*, a SAMP having at least 75% sequence identity or at least one amino acid substitution relative to the sequence of any one of SEQ ID NOs:1-13 and 35-37 (*e.g.*, SEQ ID NOs:1 and 2)).

[0054] An "expression cassette" refers to a nucleic acid construct that, when introduced into a host cell, results in transcription and/or translation of an RNA or polypeptide, respectively.

[0055] As used herein, the term "polynucleotide" refers to an oligonucleotide, or nucleotide, and fragments or portions thereof, and to DNA or RNA of genomic or synthetic origin, which may be single- or double-stranded, and represent the sense or anti-sense strand. A single polynucleotide is translated into a single polypeptide.

[0056] As used herein, the terms "peptide" and "polypeptide" are used interchangeably and describe a single polymer in which the monomers are amino acid residues which are joined together through amide bonds. A polypeptide is intended to encompass any amino acid sequence, either naturally occurring, recombinant, or synthetically produced.

[0057] As used herein, the term "substantial identity" or "substantially identical," used in the context of nucleic acids or polypeptides, refers to a sequence that has at least 50% sequence identity with a reference sequence. Alternatively, percent identity can be any integer from 50% to 100%. In some embodiments, a sequence is substantially identical to a reference sequence if the sequence has at least 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity to the reference sequence as determined using the methods described herein; preferably BLAST using standard parameters, as described below. Embodiments of the present disclosure provide for SAMPs (*e.g.*, HS

peptides) that are substantially identical to any of SEQ ID NOs:1-13 and 35-37 (SEQ ID NOs:1 and 2).

[0058] For sequence comparison, typically one sequence acts as a reference sequence, to which test sequences are compared. When using a sequence comparison algorithm, test and reference sequences are entered into a computer, subsequence coordinates are designated, if necessary, and sequence algorithm program parameters are designated. Default program parameters can be used, or alternative parameters can be designated. The sequence comparison algorithm then calculates the percent sequence identities for the test sequences relative to the reference sequence, based on the program parameters.

[0059] A comparison window includes reference to a segment of any one of a number of contiguous positions, *e.g.*, a segment of at least 10 residues. In some embodiments, the comparison window has from 10 to 600 residues, *e.g.*, about 10 to about 30 residues, about 10 to about 20 residues, about 50 to about 200 residues, or about 100 to about 150 residues, in which a sequence may be compared to a reference sequence of the same number of contiguous positions after the two sequences are optimally aligned.

[0060] Algorithms that are suitable for determining percent sequence identity and sequence similarity are the BLAST and BLAST 2.0 algorithms, which are described in Altschul *et al.* (1990) *J. Mol. Biol.* 215: 403-410 and Altschul *et al.* (1977) *Nucleic Acids Res.* 25: 3389-3402, respectively. Software for performing BLAST analyses is publicly available through the National Center for Biotechnology Information (NCBI) web site. The algorithm involves first identifying high scoring sequence pairs (HSPs) by identifying short words of length W in the query sequence, which either match or satisfy some positive-valued threshold score T when aligned with a word of the same length in a database sequence. T is referred to as the neighborhood word score threshold (Altschul *et al, supra*). These initial neighborhood word hits act as seeds for initiating searches to find longer HSPs containing them. The word hits are then extended in both directions along each sequence for as far as the cumulative alignment score can be increased. Cumulative scores are calculated using, for nucleotide sequences, the parameters M (reward score for a pair of matching residues; always >0) and N (penalty score for mismatching residues; always <0). For amino acid sequences, a scoring matrix is used to calculate the cumulative score. Extension of the word hits in each direction are halted when: the cumulative alignment score falls off by the quantity X from its maximum achieved value; the cumulative score goes to zero or below, due to the accumulation of one or more negative-scoring residue alignments; or the end of either sequence is reached. The BLAST algorithm

parameters W, T, and X determine the sensitivity and speed of the alignment. The BLASTN program (for nucleotide sequences) uses as defaults a word size (W) of 28, an expectation (E) of 10, M=1, N=-2, and a comparison of both strands. For amino acid sequences, the BLASTP program uses as defaults a word size (W) of 3, an expectation (E) of 10, and the BLOSUM62 scoring matrix (*see* Henikoff & Henikoff, *Proc. Natl. Acad. Sci. USA* 89:10915 (1989)).

[0061] The BLAST algorithm also performs a statistical analysis of the similarity between two sequences (*see, e.g.,* Karlin & Altschul, *Proc. Nat'l. Acad. Sci. USA* 90:5873-5787 (1993)). One measure of similarity provided by the BLAST algorithm is the smallest sum probability (P(N)), which provides an indication of the probability by which a match between two nucleotide or amino acid sequences would occur by chance. For example, an amino acid sequence is considered similar to a reference sequence if the smallest sum probability in a comparison of the test amino acid sequence to the reference amino acid sequence is less than about 0.01, more preferably less than about 10^{-5} , and most preferably less than about 10^{-20} .

BRIEF DESCRIPTION OF THE DRAWINGS

[0062] FIGS. 1A and 1B are bar graphs showing the expression levels of stable antimicrobial peptides (SAMPs) in HLB-susceptible and HLB-tolerant plants. FIG. 1C shows HLB-tolerant citrus varieties or close relatives express elevated levels of SAMPs.

[0063] FIGS. 2A-2C are peptide structures showing key amino acids in the loop between two α -helices of SAMPs. The prediction of peptide structure is performed by Swiss-model with AtSAMP1 as the template (PDB ID: 1Q4R). The sequences of CghSAMPa (also referred to as SAMPa herein) and CghSAMPb (also referred to as SAMPb herein) are shown in Table 1.

[0064] FIG. 3A shows photographs of *Ca. L. solanacearum*-infected *Nb* plants treated with CghSAMPa (SAMPa) or CghSAMPb (SAMPb) peptide, or mock solution.

[0065] FIG. 3B shows the bacterial titers of *Ca. L. solanacearum*-infected *Nb* plants treated with CghSAMPa (SAMPa) or CghSAMPb (SAMPb) peptide, or mock solution.

[0066] FIG. 4A-B shows SAMP effectively suppresses the growth of *C. Las* (*Candidatus Liberibacter asiaticus*) in HLB-positive citrus plants and the new shoots have no HLB symptoms. FIG. 4A shows HLB-positive citrus plants with similar *C.Las* titer were treated with buffer (mock), or SAMP (at 100 nM or 10 μ M) once by trunk injection. Pictures were taken after 5 weeks of treatment. FIG. 4B shows the titer of *C.Las* was measured by PCR on genomic DNA isolated from 8 new leaves from each mock or SAMP-treated citrus plants.

[0067] FIG. 5A-C shows SAMP pre-treated potato plants grew better and produced more tubers after *C.Lso* (*Ca. L. solanacearum*) infection by potato psyllid feeding. FIG. 5A shows potato plants were pre-treated with SAMP (10 μ M) or buffer only (mock), and then were exposed to *C.Lso* positive potato psyllids for 5 days. Plants are 3 weeks post infection. FIG. 5B shows potato tubers harvested from mock or SAMP pre-treated plants. FIG. 5C shows frying test of potato tubers harvested from SAMP or mock pre-treated *C.Lso* infected plants and healthy plants.

[0068] FIG. 6A-B show SAMP pre-treated tomato plants grew better after *C.Lso* infection by potato psyllid feeding. FIG. 6A shows tomato plants were pre-treated with SAMP or buffer only (mock) and then were exposed to *C.Lso* positive potato psyllids for 5 days. Plants are 4 weeks post infection. FIG. 6B shows above-ground biomass of SAMP pre-treated tomato plants.

[0069] FIG. 7 shows the bacterial titers of *Ca. L. solanacearum*-infected tomato plants treated with CghSAMPa or CghSAMPb peptide, or mock solution.

[0070] FIGS. 8A-8C show that SAMPs have priming effect on citrus plants and can be used to vaccinate the seedlings in the nursery.

[0071] FIGS. 9A and 9B show that SAMPs have low phytotoxic activity on citrus leaves.

[0072] FIG. 10 shows that SAMPs are highly expressed in the fruit of Australian finger lime, Australian desert lime, lemon, and *Poncirus trifoliata* (common root stock).

[0073] FIG. 11 shows that SAMPs are sensitive to human protease pepsin, a major gastric enzyme.

[0074] FIG. 12 shows that SAMPs are stable after storing at room temperature for 24 hours, or 60 °C for 24 hours, or 100 °C for 20 mins, and remain active to kill *Liberibacter crescens* as shown by the viability/cytotoxicity assay for visualizing live and dead bacterial cells (DMAO (green): a membrane-permeable DNA dye for visualizing live bacteria, and EthD-III (red): a membrane-impermeable DNA dye for visualizing dead bacteria).

[0075] FIG. 13 shows that SAMPs are stable in citrus cell lysate, which indicates that they are also stable in trees.

[0076] FIGS. 14A and 14B show that SAMPs exhibit antimicrobial activity against Gram-negative bacterial pathogens *Pseudomonas syringae* and *Agrobacterium tumefaciens* in the agar diffusion assay.

[0077] FIG. 15 shows that SAMPs, after storage for 24 hours at 4 °C, room temperature, or 60 °C, remain active to kill *Agrobacterium tumefaciens* as shown by the viability/cytotoxicity assay for visualizing live and dead bacterial cells (DMAO (green): a membrane-permeable DNA dye for visualizing live bacteria and EthD-III (red): a membrane-impermeable DNA dye for visualizing dead bacteria).

DETAILED DESCRIPTION OF THE EMBODIMENTS

I. Introduction

[0078] Citrus greening disease or “Huanglongbing” (HLB), caused by bacteria *Candidatus Liberibacter*, is a type of *Liberibacter* disease that specifically infects citrus plants. HLB is one of the most destructive diseases of citrus. *Liberibacter* (e.g., *Candidatus Liberibacter* species (*Ca. Liberibacter* or *Ca. L.*) or *Liberibacter crescens*) is a Gram-negative bacterial pathogen restricted to the phloem. HLB has caused in a significant reduction in citrus quality and quantity, resulting in billions of dollars in losses of citrus products every year, and seriously impacts the viability of the citrus industry. Current methods of treating HLB mainly involve removal of infected plants and chemical treatment against the insect vector and only led to partial control of the disease. No sustainable disease control methods for HLB have been found. The expansive and fast spread of the disease to multiple locations has already made complete removal of the infected trees an impractical strategy.

[0079] To identify and characterize important citrus defense regulators, a comparative analysis of small RNAs (sRNAs) and sRNA target genes between HLB-resistant and HLB-tolerant hybrid varieties and HLB-susceptible varieties. Several citrus defense regulators that uniquely respond to *Ca. L.* infection in HLB-resistant and HLB-tolerant hybrid varieties, but not in HLB-susceptible varieties were identified. Among the identified citrus defense regulators, putative antibacterial genes encoding stable antimicrobial proteins were found to express at a much higher level in the HLB-resistant and HLB-tolerant hybrid varieties than in HLB-susceptible varieties. The HLB-resistant and HLB-tolerant hybrid rootstocks are from completely different geographic and genetic backgrounds.

[0080] After the identification of these stable antimicrobial proteins that provide HLB-resistance or HLB-tolerance, a functional analysis of these stable antimicrobial proteins was performed in Solanaceae plant *Nicotiana benthamiana* (*Nb*) that is infected with *Candidatus Liberibacter solanacearum* (*Ca. L. solanacearum*) transmitted by potato psyllid (Example 2).

The result showed that applying SAMPs to infected plants can effectively inhibit or kill *Liberibacter* species (*e.g.*, *Candidatus Liberibacter* species) and achieve plant protection.

[0081] The disclosure includes isolated stable antimicrobial (*e.g.*, antibacterial) peptides (SAMPs) (*e.g.*, heat stable (HS) peptides), agricultural compositions containing such peptides, plants comprising such peptides, transgenic plants expressing such peptides, methods of using such peptides to prevent or treat bacterial diseases, such as those caused by Gram-negative bacteria, *e.g.*, a *Liberibacter* disease (*e.g.*, an HLB or a potato ZC disease), bacterial diseases caused by *Agrobacterium tumefaciens* (also known as *Rhizobium radiobacter*), and bacterial diseases caused by *Pseudomonas syringae*, and methods of producing plants that comprise such peptides or express such peptides. The SAMPs disclosed herein can inhibit and kill bacterial pathogens, such as Gram-negative bacterial pathogens, *e.g.*, *Liberibacter* species, *Agrobacterium tumefaciens* (also known as *Rhizobium radiobacter*), and *Pseudomonas syringae* strains. The SAMPs disclosed herein can also provide plants with resistance or tolerance to bacterial diseases, such as those caused by Gram-negative bacteria, *e.g.*, bacterial diseases caused by *Liberibacter* species, *Agrobacterium tumefaciens*, and *Pseudomonas syringae* strains.

II. Stable Antimicrobial Peptides (SAMPs)

[0082] The disclosure provides stable antimicrobial (*e.g.*, antibacterial) peptides (SAMPs) that may be injected into plants to prevent or treat a bacterial disease, such as a Gram-negative bacterial disease, *e.g.*, a *Liberibacter* disease (*e.g.*, HLB). The SAMPs disclosed herein may also be used to prevent or treat bacterial diseases caused by other bacterial pathogens, such as *Agrobacterium tumefaciens* (also known as *Rhizobium radiobacter*) and *Pseudomonas syringae* strains. The plants may also be genetically modified to express one or more of the SAMPs described herein. The present disclosure identified genes in plants encoding SAMPs that are differentially expressed in *Liberibacter* disease-resistant/tolerant (*e.g.*, HLB-resistant/tolerant) plants and *Liberibacter* disease-susceptible (*e.g.*, HLB-susceptible) plants. In some embodiments, the SAMPs disclosed herein are heat stable (*e.g.*, heat stable (HS) peptides). In some embodiments, the SAMPs disclosed herein are also stable in plant extracts. In further embodiments, the SAMPs disclosed herein are also stable in plant lysates (*e.g.*, citrus lysates). As shown in FIGS. 1A and 1B, these SAMPs expressed at a much higher level in *Liberibacter* disease-resistant/tolerant (*e.g.*, HLB-resistant/tolerant) plants than in *Liberibacter* disease-susceptible (*e.g.*, HLB-susceptible) plants. By comparing the amino acid sequences of SAMPs from different citrus plants and solanaceous plants, it is found that the SAMPs

expressed in some HLB-resistant/tolerant citrus plants and pepper (*Ca: Capsicum annuum*) are shorter while the SAMPs expressed in HLB-susceptible solanaceous plants are longer. Table 1 below provides amino acid sequences of SAMPs in various hybrid plants, citrus plants, and solanaceous plants.

Table 1

SAMP	Plant Source	Amino Acid Sequence	Nucleic Acid Sequence
CghSAM Pa	<i>Eremocitrus glauca</i> x citrus sp.	MCCNRGKNVSIENLHQ GFTHIFESTFESTEGVAE YVSHPSHVEYANLFLA NLEKVLVIDYKPTTVRV (SEQ ID NO:1)	ATGTGCTGCAACAGGGGCAAGAATGTGAGCATTGAGA ATCTTCATCAGGGTTTCACTCATATTTTTGAATCTACC TTTGAGAGCACAGAGGGTGTTCAGAGTATGTATCTC ATCCGTCACATGTTGAATACGCAAACCTGTTCCCTGGCC AACTTGGAGAAAAGTTCTCGTGATTGACTACAAACCGA CAACAGTACGTGTCTGAGAAGGGTGGGCGCGCCGACC CAGCTTTCTTGACAAAAGTTGGCATTATAAGAAAG (SEQ ID NO:14) or ATGTGCTGCAACAGGGGCAAGAATGTGAGCATTGAGA ATCTTCATCAGGGTTTCACTCATATTTTTGAATCTACC TTTGAGAGCACAGAGGGTGTTCAGAGTATGTATCTC ATCCGTCACATGTTGAATACGCAAACCTGTTCCCTGGCC AACTTGGAGAAAAGTTCTCGTGATTGACTACAAACCGA CAACAGTACGTGTCTGA (SEQ ID NO:28)
CghSAM Pb	<i>Eremocitrus glauca</i> x citrus sp.	MCCNRGKNVSIENLHQ GFPHLFESTFESTEGVAE YVSHPAHVEYANLFLA NLEKVLVIDYKPTTVRV (SEQ ID NO:2)	ATGTGCTGCAACAGGGGCAAGAATGTGAGCATTGAGA ATCTTCATCAGGGTTTCCCTCATCTTTTCGAATTTACCT TTGAGAGCACAGAGGGTGTTCAGAGTATGTATCTCA TCCGGCACATGTTGAATACGCAAACCTGTTCCCTGGCC AACTTGGAGAAAAGTTCTCGTGATTGACTACAAACCGA CAACAGTACGTGTCTGAGAAGGGTGGGCGCGCCGACC CAGCTTTCTT (SEQ ID NO:15) or ATGTGCTGCAACAGGGGCAAGAATGTGAGCATTGAGA ATCTTCATCAGGGTTTCCCTCATCTTTTCGAATTTACCT TTGAGAGCACAGAGGGTGTTCAGAGTATGTATCTCA TCCGGCACATGTTGAATACGCAAACCTGTTCCCTGGCC AACTTGGAGAAAAGTTCTCGTGATTGACTACAAACCGA CAACAGTACGTGTCTGA (SEQ ID NO:29)
942SAM P1, US-942	<i>Citrus reticulata</i> x <i>Poncirus trifoliata</i>	MCCNRGKNVSIENLHQ GFTHIFESTFESTEGVAE YVAHPAHVEYANLFLA NLEKVLVIDYKPTTTERV (SEQ ID NO:3)	ATGTGCTGCAACAGGGGCAAGAATGTGAGCATTGAGA ATCTTCATCAGGGTTTCACTCATATTTTTGAATCTACC TTTGAGAGCACAGAGGGTGTTCAGAGTATGTAGCTC ATCCGGCACATGTTGAATACGCAAACCTGTTCCCTGGC CAACTTGGAGAAAAGTTCTCGTGATTGACTACAAACCG ACAACAGAACGTGTCTAAGGGTGGGCGCGCCGACCCA GCTTTCTTGACAA (SEQ ID NO:16) or ATGTGCTGCAACAGGGGCAAGAATGTGAGCATTGAGA ATCTTCATCAGGGTTTCACTCATATTTTTGAATCTACC TTTGAGAGCACAGAGGGTGTTCAGAGTATGTAGCTC ATCCGGCACATGTTGAATACGCAAACCTGTTCCCTGGC CAACTTGGAGAAAAGTTCTCGTGATTGACTACAAACCG ACAACAGAACGTGTCTAA (SEQ ID NO:30)

SAMP	Plant Source	Amino Acid Sequence	Nucleic Acid Sequence
CsSAMP 1	<i>Citrus sinensis</i>	MCCNRGKNVSIENLHQ GFTHIFESTFESTEGVAE YVAHPAHVEYANLFLA NLEKVLVIDYKPTTVRV (SEQ ID NO:4)	ATGTGCTGCAACAGGGGCAAGAATGTGAGCATTGAGA ATCTTCATCAGGGTTTCACTCATATTTTGAATCTACC TTTGAGAGCACAGAGGGTGTTCAGAGTATGTAGCTC ATCCGGCACATGTTGAATACGCAAACCTGTTTCCTGGC CAACTTGGAGAAAGTTCTCGTGATTGACTACAAACCG ACAACAGTACGTGTCTGAGTTGTACTAGTAGGGAA (SEQ ID NO:17) or ATGTGCTGCAACAGGGGCAAGAATGTGAGCATTGAGA ATCTTCATCAGGGTTTCACTCATATTTTGAATCTACC TTTGAGAGCACAGAGGGTGTTCAGAGTATGTAGCTC ATCCGGCACATGTTGAATACGCAAACCTGTTTCCTGGC CAACTTGGAGAAAGTTCTCGTGATTGACTACAAACCG ACAACAGTACGTGTCTGA (SEQ ID NO:31)
CsSAMP 2	<i>Citrus sinensis</i>	MIAELIRSCGLELLAV KYKGKNVSIENLHQGFT HIFESTFESTEGVAEYV AHPAHVEYANLFLANL EKVLVIDYKPTTVRV (SEQ ID NO:5)	ATGGAAGAAGCTAAAGGAGTGGTGAAGCACGTA CTTC TGGCCAAGTTCAAAGAAGGGACTGCTCAAGATCAAAT TGATCAGCTCATCAAAGACTATGCAAATCTTGTGAAT CTCATTGAACCCATGAAGTCTTTCCAATGGGGCAAGA ATGTGAGCATTGAGAATCTTCATCAGGGTTTCACTCAT ATTTTTGAATCTACCTTTGAGAGCACAGAGGGTGTTC AGAGTATGTAGCTCATCCGGCACATGTTGAATACGCA AACTTGTTCCTGGCCAACTTGGAGAAAGTTCTCGTGAT TGACTACAAACCGACAACAGTACGTGTCTGA (SEQ ID NO:18)
CsSAMP 3	<i>Citrus sinensis</i>	MEEAKGVVKHVLLAKF KEGTAQDQIDQLIKDYA NLVNLIEPMKSFQWGK DVSINRHQGFTHIFEST FESTEGVAEYVAHPAH VEYANLFLANLEKVLVI DYKPTTVRV (SEQ ID NO:6)	ATGGAAGAAGCTAAAGGAGTGGTGAAGCACGTA CTTC TGGCCAAGTTCAAAGAAGGGACTGCTCAAGATCAAAT TGATCAGCTCATCAAAGACTATGCAAATCTTGTGAAT CTCATTGAACCCATGAAGTCTTTCCAATGGGGCAAGA ATGTGAGCATTGAGAATCTTCATCAGGGTTTCACTCAT ATTTTTGAATCTACCTTTGAGAGCACAGAGGGTGTTC AGAGTATGTAGCTCATCCGGCACATGTTGAATACGCA AACTTGTTCCTGGCCAACTTGGAGAAAGTTCTCGTGAT TGACTACAAACCGACAACAGTACGTGTCTGA (SEQ ID NO:19)
CsSAMP 4	<i>Citrus sinensis</i>	MGEGEEAAMGEFKHLV IVKFKEGVVVEDIVKG MKKLVEIDAVKSFEW GQDVEGQEMLRQGFTH AFLMTFNKKEDYTFAS HPSHVEFSATFSAAIEKI VLLDFPTVLGKAPAA (SEQ ID NO:7)	ATGGGTGAGGGTGAAGAGGCAGCAATGGGAGAGTTC AAGCACTTGGTGATTGTTAAGTTCAAGGAAGGTGTGG TTGTGGAGGATATTGTCAAAGGGATGAAAAAGCTGGT TTCAGAGATTGATGCTGTCAAATCTTTTGAATGGGGCC AAGATGTAGAAGGGCAGGAGATGCTTAGGCAAGGCT TCACACATGCATTCTTGATGACATTCAACAAGAAGGA AGACTATACAACCTTTGCAAGCCATCCCAGCCACGTC GAATTCTCGGCTACATTTTCAGCTGCTATTGAGAAGAT TGTCTGCTTGATTTCCTACCGTGCTTGGCAAAGCAC CAGCAGCATGA (SEQ ID NO:20)
CsSAMP 1	<i>Citrus clementina</i>	MKAETKGRDMEEAKG VVKHVLLAKFKEGTAQ DQIDQLIKDYANLVNLI EPMKSFQWGKDVSIEN LHQGFTHIFESTFESTEG VAEYVAHPAHVEYANL FLANLEKVLVIDYKPTT VRV (SEQ ID NO:8)	ATGAAAGCCGAAACAAAAGGCAGAGATATGGAAGAA GCTAAAGGAGTGGTGAAGCACGTA CTTCGCGCAAGT TCAAAGAAGGGACTGCTCAAGATCAAATTGATCAGCT CATCAAAGACTATGCAAATCTTGTGAATCTCATTGAA CCCATGAAGTCTTTCCAATGGGGCAAGGATGTGAGCA TTGAGAATCTTCATCAGGGTTTCACTCATATTTTGA TCTACCTTTGAGAGCACAGAGGGTGTTCAGAGATATG TAGCTCATCCGGCACATGTTGAATACGCAAACCTGTTT CTGGCCAACTTGGAGAAAGTTCTCGTGATTGACTACA AACCGACAACGTACGTGTCTGA (SEQ ID NO:21)

SAMP	Plant Source	Amino Acid Sequence	Nucleic Acid Sequence
CaSAMP	<i>Capsicum annuum</i>	MSYGRGKDVSTENLQQ GFTHVFESTFDSTEGVA EYVSHPVHVEFANLML PQLEKVLVIDYKPEKVG P (SEQ ID NO:9)	ATGTCATATGGCAGGGGTAAGGATGTGAGCACAGAG AACCTCCAGCAAGGTTTCACTCATGTTTTTGAGTCAAC GTTTCGACAGTACAGAAGGTGTTGCAGAGTATGTAAGT CATCCGGTTCATGTTGAATTTGCAAATCTAATGCTTCC TCAGCTGGAGAAAGTCCTCGTCATCGACTACAAACCG GAGAAAGTCGGTCCCTAA (SEQ ID NO:22)
NbSAMP	<i>Nicotiana benthamiana</i>	MEGGKVKHILLAKFKD GIPADQIDQLIKQYANL VNLIPEMKAFHWGENV SIENFHQGFTHVFESTFD STEGIAEYIDHPAHVEY ANTLLPQLEKVLVIDYK PEKVG (SEQ ID NO:10)	ATGGAGGGTGGTAAAGTGAAGCACATATTGCTGGCCA AGTTCAAAGATGGAATTCCAGCAGACCAAATCGACCA ACTGATTAAGCAATATGCTAATCTTGTCATCTCATCG AACCAATGAAAGCTTTTCATTGGGGTGAGAATGTGAG CATAGAGAACTTCCACCAAGGTTTCACTCATGTTTTTG AGTCAACGTTTCGACAGTACAGAAGGAATTGCAGAGTA TATAGATCATCCGGCTCATGTTGAATATGCAAATACA TTGCTTCCTCAGCTGGAGAAAGTCCTTGTCATCGACTA CAAACCAGAGAAAGTTGGTCCC (SEQ ID NO:23) or ATGGAGGGTGGTAAAGTGAAGCACATATTGCTGGCCA AGTTCAAAGATGGAATTCCAGCAGACCAAATCGACCA ACTGATTAAGCAATATGCTAATCTTGTCATCTCATCG AACCAATGAAAGCTTTTCATTGGGGTGAGAATGTGAG CATAGAGAACTTCCACCAAGGTTTCACTCATGTTTTTG AGTCAACGTTTCGACAGTACAGAAGGAATTGCAGAGTA TATAGATCATCCGGCTCATGTTGAATATGCAAATACA TTGCTTCCTCAGCTGGAGAAAGTCCTTGTCATCGACTA CAAACCAGAGAAAGTTGGTCCCTAA (SEQ ID NO:32)
SISAMP	<i>Solanum lycopersicum</i>	MEGGKGGVVKHILLAK FKDGIPPEQIDQLIKQYA NLVNLVEPMKAFQWG KDVSIENLHQGFTHVFE STFDSLEGVAEYIAHPV HVEYANTLLPQLEKFLI VDYKPQ (SEQ ID NO:11)	ATGGAGGGTGGCAAAGGAGGAGTTGTGAAGCACATTT TGCTAGCAAAGTTCAAAGATGGGATCCCACCTGAACA GATTGATCAACTCATTAAGCAGTATGCTAATCTTGTC ATCTTGTTGAACCCATGAAGGCTTTTCAATGGGGTAA GGATGTGAGCATAGAAAATCTTCATCAAGGTTTCACT CATGTTTTCGAGTCTACGTTTGACAGTTTAGAAGGTGT TGCAGAGTATATAGCTCATCCTGTTTATGTTGAATATG CAAATACATTGCTTCCTCAGCTGGAGAAATTCCTTATC GTCGACTACAAACCACAG (SEQ ID NO:24) or ATGGAGGGTGGCAAAGGAGGAGTTGTGAAGCACATTT TGCTAGCAAAGTTCAAAGATGGGATCCCACCTGAACA GATTGATCAACTCATTAAGCAGTATGCTAATCTTGTC ATCTTGTTGAACCCATGAAGGCTTTTCAATGGGGTAA GGATGTGAGCATAGAAAATCTTCATCAAGGTTTCACT CATGTTTTCGAGTCTACGTTTGACAGTTTAGAAGGTGT TGCAGAGTATATAGCTCATCCTGTTTATGTTGAATATG CAAATACATTGCTTCCTCAGCTGGAGAAATTCCTTATC GTCGACTACAAACCACAGTAA (SEQ ID NO:33)
SmSAMP	<i>Solanum melongena</i>	MNIAVFLPSSCPALPRS KASRPSPGQFPFLAKN VQLLLVLRYSSTARA MSLRGENVSIENLHQGF THVFESTFDSVEGIAEYI DHPAHVEYANILLTQLE KVLVIDYKPEKLSP (SEQ ID NO:12)	ATGAATATTGCTGTCTTTCTCCCTTCGTCCTGCCCTGC CCTGCCCCGCTCAAAGGCTTCCCGCCCATCCCCACCCG GCCAATTTCCGTTCTAGCCAAGAATGTTAGCTTCTA CTAGTCTTGAGGTCTTATAGTTCCACCGCTCGTGCTAT GTCACCTTAGGGGTGAGAATGTGAGCATAGAGAACCTC CACCAAGGTTTCACTCACGTTTTTCGAGTCAACGTTTGA CAGTGTAGAAGGCATTGCAGAGTATATAGATCATCCT GCTCATGTTGAATATGCAAATATATTGCTTACTCAGCT GGAGAAAGTCCTTGTCATCGACTACAAACCAGAGAAA CTCAGCCCCTAA (SEQ ID NO:25)

SAMP	Plant Source	Amino Acid Sequence	Nucleic Acid Sequence
StSAMP	<i>Solanum tuberosum</i>	MEGGKGGVVKHILLAK FKDGIPPEQIDQLIKQYA NLVNLIEPMKAFQWGK DVSIENTLHQGFTHVFES TFDSLEGVAEYIAHPVH VEFANTMLPQLEKVLII DYKPQ (SEQ ID NO:13)	CTCATACAATCAACCAAATAAAGGACCCTTTTCTCTCC ACTATTTTGTGCTTGTCTAGTCAAGGAAGAAGAGTGAT AAAATAGAAATGGAGGGTGGTAAAGGAGGAGTGGTG AAGCACATTTTGTAGCAAAGTTCAAAGATGGGATCC CACCTGAACAAATTGATCAACTCATTAAAGCAGTATGC TAATCTTGTCAATCTTATTGAACCCATGAAGGCTTTTC AATGGGGCAAGGATGTGAGCATAGAAAAACCTTCACCA AGGTTTCACTCATGTTTTTGAGTCGACGTTTGACAGTT TAGAAGGCGTTGCAGAGTATATAGCTCATCCTGTTCA TGTTGAATTTGCAAATACAATGCTTCCTCAGCTGGAG AAAGTCCTTATCATTGACTACAAACCACAGTAACTCA GTCCTAAACTGGATTCACAAATTGATGCACCTTGATGT AATAGGTATATCAGTTTTACTTTACTGTACTGAAATCC AATAAGAACACAAACTTTTATTAAGGGTGTGTGTCTT GCTTGTGTGCAATTATTGTATTCACTTCGTAGACGCTA ATGCGAGTAACCTTATGGTCAGCTTGGGCTGTTTAAACT CGAGGAAGAATGCTCTTCGTTCTTCTTCCAGGGAG AATGAATGATGAACAACATATAAGTGCATCAATAAAC TCAGATTGGTGTTCCTTCTTCTT (SEQ ID NO:26) or ATGGAGGGTGGTAAAGGAGGAGTGGTGAAGCACATT TTGCTAGCAAAGTTCAAAGATGGGATCCACCTGAAC AAATTGATCAACTCATTAAAGCAGTATGCTAATCTTGT AATCTTATTGAACCCATGAAGGCTTTTCAATGGGGCA AGGATGTGAGCATAGAAAACCTTCACCAAGGTTTCAC TCATGTTTTTGTGAGTCGACGTTTGACAGTTTAGAAGGCG TTGCAGAGTATATAGCTCATCCTGTTTCATGTTGAATT GCAAATACAATGCTTCCTCAGCTGGAGAAAGTCCTTA TCATTGACTACAAACCACAGTAA (SEQ ID NO:34)
PtSAMP 1a	<i>Ponirus trifoliata</i>	MCCNRGKNVSIENLHQ GFTHIFESTFESTEGVAE YVAHPAHVEYANSFLA NLEKVLVIDYKPTTVRV (SEQ ID NO:35)	ATGTGCTGCAACAGGGGCAAGAATGTGAGCATTGAGA ATCTTCATCAGGGTTTCACTCATATTTTGAATCTACC TTTGAGAGCACAGAGGGTGTGTCAGAGTATGTAGCTC ATCCGGCACATGTTGAATACGAAACTCGTTCCTGGC CAACTTGGAGAAAGTTCTCGTGATTGACTACAAACCG ACAACAGTACGTGTCTGA (SEQ ID NO:38)
PtSAMP 1b	<i>Ponirus trifoliata</i>	MCCNRGKNVSIENLHQ GFTHIFESTFESTEGVAE YVAHPAHVEYTNFLA NLEKVLVIDYKPTTVRV (SEQ ID NO:36)	ATGTGCTGCAACAGGGGCAAGAATGTGAGCATTGAGA ATCTTCATCAGGGTTTCACTCATATTTTGAATCTACC TTTGAGAGCACAGAGGGTGTGTCAGAGTATGTAGCTC ATCCGGCACATGTTGAATACGAAACTCGTTCCTGGC CAACTTGGAGAAAGTTCTCGTGATTGACTACAAACCG ACAACAGTACGTGTCTGA (SEQ ID NO:39)
MCaSA MP	<i>Microcitrus australasica</i>	MCCNRGKNVSIENLHQ GFTHIFESTFESTEGVAE YVSHPAHVEYANLFLA NLEKVLVIDYKPTTVRV (SEQ ID NO:37)	ATGTGCTGCAACAGGGGCAAGAATGTGAGTATTGAGA ATCTTCATCAGGGTTTCACTCATATTTTGAATCTACC TTTGAGAGCACAGAGGGTGTGTCAGAGTATGTATCTC ATCCGGCACATGTTGAATACGAAACTTGTTCCTCGCC AACTTGGAGAAAGTTCTCGTGATTGACTACAAACCGA CAACAGTACGTGTCTGA (SEQ ID NO:40)

[0083] Further, SAMPs (e.g., HS peptides) from the *Liberibacter* disease-resistant/tolerant (e.g., HLB-resistant/tolerant) plants contain a few different amino acids within the α - β barrel domain as compared to the corresponding amino acids in the *Liberibacter* disease-susceptible (e.g., HLB-susceptible) plants. Two key amino acid differences between the *Liberibacter* disease-resistant/tolerant (e.g., HLB-resistant/tolerant) plants and the *Liberibacter* disease-

susceptible (*e.g.*, HLB-susceptible) plants are identified in the loop between two α -helices of SAMPs (FIGS. 2A-2C). These two amino acid positions are indicated as bold and underlined in the sequence alignment of SEQ ID NOs:1-13 and 35-37.

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CghSAMPa -----MCC-----NR 5
CghSAMPb -----MCC-----NR 5
942SAMP1 -----MCC-----NR 5
CsSAMP1 -----MCC-----NR 5
CsSAMP2 -----MIAELIRSCC-----GLELLAVKYK 20
CsSAMP3 -----MEEAKGVVKHVLLAKFKEGTAQDQIDQLIKDYANLVN-----LIEPMKSFQW 47
CsSAMP4 -----MGEGEAAAMGEFKHLVIVKFKEGVVVEDIVKGMK-----KLVS-----EIDAVKSFQW 48
CcSAMP1 MKAETKGRDMEEAKGVVKHVLLAKFKEGTAQDQIDQLIKDYANLVN-----LIEPMKSFQW 56
CaSAMP -----MSYGR 5
NbsAMP -----MEGGKVKHILLAKFKDGIPADQIDQLIKQYANLVN-----LIEPMKAFHW 45
SlSAMP -----MEGGKGGVVKHILLAKFKDGIPPEQIDQLIKQYANLVN-----LVEPMKAFQW 48
SmSAMP MNIAV-----FLPSSCPALPRSKASRSPPGQFPFLAKNVQLLLVLRYSSTARAMSLR 54
StSAMP -----MEGGKGGVVKHILLAKFKDGIPPEQIDQLIKQYANLVN-----LIEPMKAFQW 48
PtSAMP1a -----MCC-----NR 5
PtSAMP1b -----MCC-----NR 5
McaSAMP -----MCC-----NR 5

CghSAMPa GKNVSI-ENLHQGFTHIFESTFESTEGVAEYVSHPSHVEYANLFLANLEKVLVIDYKPTT 64
CghSAMPb GKNVSI-ENLHQGFPHLFESTFESTEGVAEYVSHPAHVEYANLFLANLEKVLVIDYKPTT 64
942SAMP1 GKNVSI-ENLHQGFTHIFESTFESTEGVAEYVAHPAHVEYANLFLANLEKVLVIDYKPTT 64
CsSAMP1 GKNVSI-ENLHQGFTHIFESTFESTEGVAEYVAHPAHVEYANLFLANLEKVLVIDYKPTT 64
CsSAMP2 GKNVSI-ENLHQGFTHIFESTFESTEGVAEYVAHPAHVEYANLFLANLEKVLVIDYKPTT 79
CsSAMP3 GKDVSI-ENRHQGFTHIFESTFESTEGVAEYVAHPAHVEYANLFLANLEKVLVIDYKPTT 106
CsSAMP4 GQDVEGQEMLRQGFTHAFLMTFNKKEDYTTFASHPSHVEFSATFSAAIEKIVLLDFPTVL 108
CcSAMP1 GKDVSI-ENLHQGFTHIFESTFESTEGVAEYVAHPAHVEYANLFLANLEKVLVIDYKPTT 115
CaSAMP GKDVST-ENLQGFTHVFESTFDSLEGVAEYVSHPVHVEFANLMLPQLEKVLVIDYKPEK 64
NbsAMP GENVSI-ENFHQGFTHVFESTFDSLEGVAEYIDHPAHVEYANTLLPQLEKVLVIDYKPEK 104
SlSAMP GKDVSI-ENLHQGFTHVFESTFDSLEGVAEYIAHPVHVEYANTLLPQLEKFLIVDYKPQ- 106
SmSAMP GENVSI-ENLHQGFTHVFESTFDSLEGVAEYIDHPAHVEYANILLTQLEKVLVIDYKPEK 113
StSAMP GKDVSI-ENLHQGFTHVFESTFDSLEGVAEYIAHPVHVEFANTMLPQLEKVLIIIDYKPQ- 106
PtSAMP1a GKNVSI-ENLHQGFTHIFESTFESTEGVAEYVAHPAHVEYANSFLANLEKVLVIDYKPTT 64
PtSAMP1b GKNVSI-ENLHQGFTHIFESTFESTEGVAEYVAHPAHVEYTNNSFLANLEKVLVIDYKPTT 64
McaSAMP GKNVSI-ENLHQGFTHIFESTFESTEGVAEYVSHPAHVEYANLFLANLEKVLVIDYKPTT 64
*::*. * :*** * * **: . * . : : ** ***: : : **: .::*:

CghSAMPa VRV---- 67
CghSAMPb VRV---- 67
942SAMP1 ERV---- 67
CsSAMP1 VRV---- 67
CsSAMP2 VRV---- 82
CsSAMP3 VRV---- 109
CsSAMP4 GKAPAA 114
CcSAMP1 VRV---- 118
CaSAMP VGP----- 67
NbsAMP VGP----- 107
SlSAMP ----- 106
SmSAMP LSP----- 116
StSAMP ----- 106
PtSAMP1a VRV---- 67
PtSAMP1b VRV---- 67
McaSAMP VRV---- 67

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[0084] In particular, SAMPs disclosed herein include the following:

Poncirus trifoliata Flying Dragon 1, Kryder 55-5, Nanjing
MCCNRGKNVSIENLHQGFTHIFESTFESTEGVAEYVAHPAHVEYANSFLANLEKVLVIDYKPTTVRV (SEQ ID NO:35)

Poncirus trifoliata Flying Dragon 2

MCCNRGKNVSIENLHQGFTHIFESTFESTEGVAEYVAHPAHVEYTNFLANLEKVLVI
DYKPTTVRV (SEQ ID NO:36)

Microcitrus australasica and *Poncirus trifoliata* (Texas):

MCCNRGKNVSIENLHQGFTHIFESTFESTEGVAEYVSHPAHVEYANLFLANLEKVLVI
DYKPTTVRV (SEQ ID NO:37)

Australian desert lime *Eremocitrus glauca* 1

MCCNRGKNVSIENLHQGFTHIFESTFESTEGVAEYVSHPSHVEYANLFLANLEKVLVID
YKPTTVRV (SEQ ID NO:1)

[0085] The present disclosure provides isolated stable antimicrobial (*e.g.*, antibacterial) peptides (SAMPs) (*e.g.*, HS peptides) comprising a sequence that is substantially identical (*e.g.*, at least 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% identical to) to a sequence of any one of SEQ ID NOs:1-13 and 35-37 (*e.g.*, SEQ ID NOs:1-5 and 9). In some embodiments, the isolated peptides comprise a sequence having at least 75% sequence identity (*e.g.*, at least 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity) to a sequence of any one of SEQ ID NOs:1-13 and 35-37 (*e.g.*, SEQ ID NOs:1-5 and 9). In particular embodiments, the isolated peptides comprise a sequence having at least 75% sequence identity (*e.g.*, at least 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity) to a sequence of any one of SEQ ID NOs:1 and 2.

[0086] The present disclosure provides isolated SAMPs (*e.g.*, HS peptides) comprising a sequence having at least one amino acid substitution (*e.g.*, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, or 15 amino acid substitutions) relative to a sequence of any one of SEQ ID NOs:1-13 and 35-37 (*e.g.*, SEQ ID NOs:1-5 and 9). In particular embodiments, the isolated peptides comprise a sequence having at least one amino acid substitution (*e.g.*, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, or 15 amino acid substitutions) relative to a sequence of any one of SEQ ID NOs:1 and 2.

[0087] Isolated SAMPs (*e.g.*, HS peptides) of the present disclosure may also comprise a sequence of

X₁GX₂X₃VSX₄ENX₅X₆QGF₇HX₈FEX₉TFX₁₀SX₁₁EGX₁₂AEYX₁₃X₁₄HPX₁₅HVEX₁₆ANX₁₇
X₁₈LX₁₉X₂₀LEKX₂₁LX₂₂X₂₃DYKPX₂₄TX₂₅RV (SEQ ID NO:27), in which X₁ is R, K, or W;
X₂ is K or E; X₃ is N or D; X₄ is T or I; X₅ is L, F, or R; X₆ is H or Q; X₇ is P or T; X₈ is I, L,
or V; X₉ is S or F; X₁₀ is E or D; X₁₁ is T or L; X₁₂ is V or I; X₁₃ is V or I; X₁₄ is S, A, or D;
X₁₅ is S, A, or V; X₁₆ is Y or F; X₁₇ is L or T; X₁₈ is F, M, or L; X₁₉ is A, P, or T; X₂₀ is N or Q;

X₂₁ is V or F; X₂₂ is V or I; X₂₃ is V or I; X₂₄ is T, E, or Q; and X₂₅ is V, E, L. In particular embodiments of isolated SAMPs (*e.g.*, HS peptides) comprising a sequence of SEQ ID NO:27, X₁ may be R; X₂ may be K; X₃ may be N; X₄ may be I; X₅ may be L; X₆ may be H; X₁₀ may be E; X₁₁ may be T; X₁₂ may be V; X₁₃ may be V; X₁₆ may be Y; X₁₇ may be L; X₁₈ may be F; X₁₉ may be A; X₂₀ may be N; X₂₁ may be V; X₂₂ may be V, X₂₃ may be I; and/or X₂₄ may be T.

[0088] In certain embodiments, the disclosure also provides methods of producing a plant (*e.g.*, a citrus plant) having enhanced *Liberibacter* disease resistance (*e.g.*, HLB resistance) or *Liberibacter* disease tolerance (*e.g.*, HLB tolerance) (*i.e.*, enhanced resistance or tolerance to a bacterial infection caused by bacteria in the genus *Liberibacter* (*e.g.*, *Candidatus Liberibacter* species) or *Liberibacter crescens*) by introducing a mutation into a polynucleotide in the plant, in which the mutated polynucleotide encodes a SAMP of the present disclosure, such as the SAMPs described above (*e.g.*, SAMPs having at least 75% sequence identity (*e.g.*, at least 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity) or at least one amino acid substitution (*e.g.*, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, or 15 amino acid substitutions) relative to the sequence of any one of SEQ ID NOs:1-13 and 35-37 (*e.g.*, SEQ ID NOs:1-5 and 9). In particular, the mutated polynucleotide may encode a SAMP having at least 75% sequence identity (*e.g.*, at least 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% sequence identity) or at least one amino acid substitution (*e.g.*, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, or 15 amino acid substitutions) relative to the sequence of any one of SEQ ID NOs:1 and 2.

[0089] In other embodiments, methods of producing a plant (*e.g.*, a citrus plant) having enhanced *Liberibacter* disease resistance (*e.g.*, HLB resistance) or *Liberibacter* disease tolerance (*e.g.*, HLB tolerance) (*i.e.*, enhanced resistance or tolerance to a bacterial infection caused by bacteria in the genus *Liberibacter* (*e.g.*, *Candidatus Liberibacter* species) or *Liberibacter crescens*) may be achieved by introducing one or more mutations into a polynucleotide in the plant, in which the mutations specifically alter the amino acids in the native SAMP in the plant that correspond to the two bold and underlined amino acids in the sequence alignment shown above. For example, the positions in a polynucleotide sequence encoding a native SAMP in a plant that correspond to the two bold and underlined amino acids in the sequence alignment shown above may be mutated to encode S, A, or D as the first of two bold and underlined amino acids and S, A, or V as the second of the two bold and underlined amino acids.

[0090] In some embodiments, one of skill in the art may perform sequence alignment of a polynucleotide sequence encoding a native SAMP in a plant and a polynucleotide sequence encoding an effective SAMP (*e.g.*, SEQ ID NO:14 or 15) to determine specific nucleic acids in the polynucleotide of the plant that need to be mutated such that the resulting mutated polynucleotide in the plant encodes the substantially the same polynucleotide sequence as that of an effective SAMP (*e.g.*, SEQ ID NO:14 or 15).

[0091] The disclosure also provides methods of preventing or treating a *Liberibacter* disease (*e.g.*, HLB) and/or preventing or treating a bacterial infection caused by bacteria in the genus *Liberibacter* (*e.g.*, *Candidatus Liberibacter* species or *Liberibacter crescens*) in a plant by contacting the plant with a SAMP described above (*e.g.*, SAMPs having at least 75% sequence identity to or at least one amino acid substitution relative to the sequence of any one of SEQ ID NOs:1-13 and 35-37 (*e.g.*, SEQ ID NOs:1-5 and 9 (*e.g.*, SEQ ID NO:1 and 2)). Without being bound by any theory, SAMPs (*e.g.*, HS peptides) may target and destroy bacterial cells, and/or induce defense response in plants, thus, enhancing the *Liberibacter* disease resistance or *Liberibacter* disease tolerance of plants. An example of a bacterial infection caused by bacteria in the genus *Liberibacter* (*e.g.*, *Candidatus Liberibacter* species or *Liberibacter crescens*) that can be treated or prevented as described herein is potato zebra chip disease. For example, potato zebra disease can be treated or prevented in potato or tomato plants.

III. Agricultural Compositions

[0092] The disclosure also provides agricultural compositions that contain one or more of the SAMPs (*e.g.*, HS peptides) described herein for use in preventing or treating a bacterial disease (*e.g.*, a *Liberibacter* disease (HLB) or potato Zebra Chip disease, and other bacterial diseases such as those caused by *Agrobacterium tumefaciens* (also known as *Rhizobium radiobacter*) and *Pseudomonas syringae*) in a plant. In some embodiments, the agricultural composition further includes at least one of an herbicide, an herbicide safener, a surfactant, a fungicide, a pesticide, a nematocide, a plant activator, a synergist, a plant growth regulator, an insect repellent, an acaricide, a molluscicide, or a fertilizer.

[0093] An agricultural composition comprising one or more SAMPs (*e.g.*, HS peptides) described herein can also include one or more of: a surface-active agent, an inert carrier, a preservative, a humectant, a feeding stimulant, an attractant, an encapsulating agent, a binder, an emulsifier, a dye, a UV protective, a buffer, a flow agent, a fertilizer, a nitrogen fixation agent, micronutrient donors, or other preparations that influence plant growth. The agricultural composition can also include one or more agrochemicals including: herbicides, insecticides,

fungicides, bactericides, nematocides, molluscicides, acaricides, plant growth regulators, harvest aids, and fertilizers, which can also be combined with carriers, surfactants or adjuvants as appropriate for the agrochemical. Suitable carriers and adjuvants can be solid or liquid and correspond to the substances ordinarily employed in formulation technology, *e.g.*, natural or regenerated mineral substances, solvents, dispersants, wetting agents, tackifiers, binders, or fertilizers. The active ingredients of the present disclosure are normally applied in the form of compositions and can be applied to the crop area, plant, or seed to be treated. For example, the compositions of the present disclosure may be applied during growth, seeding, or storage.

[0094] Surface-active agents that can be used with the presently described SAMPs (*e.g.*, HS peptides) include anionic compounds such as a carboxylate of, for example, a metal; carboxylate of a long chain fatty acid; an N-acylsarcosinate; mono- or di-esters of phosphoric acid with fatty alcohol ethoxylates or salts of such esters; fatty alcohol sulfates such as sodium dodecyl sulfate, sodium octadecyl sulfate or sodium cetyl sulfate; ethoxylated fatty alcohol sulfates; ethoxylated alkylphenol sulfates; lignin sulfonates; petroleum sulfonates; alkyl aryl sulfonates such as alkyl-benzene sulfonates or lower alkylnaphthalene sulfonates, *e.g.*, butyl-naphthalene sulfonate; salts of sulfonated naphthalene-formaldehyde condensates; salts of sulfonated phenol-formaldehyde condensates; more complex sulfonates such as the amide sulfonates, *e.g.*, the sulfonated condensation product of oleic acid and N-methyl taurine; or the dialkyl sulfosuccinates, *e.g.*, the sodium sulfonate or dioctyl succinate. Non-ionic agents include condensation products of fatty acid esters, fatty alcohols, fatty acid amides or fatty-alkyl- or alkenyl-substituted phenols with ethylene oxide, fatty esters of polyhydric alcohol ethers, *e.g.*, sorbitan fatty acid esters, condensation products of such esters with ethylene oxide, *e.g.*, polyoxyethylene sorbitar fatty acid esters, block copolymers of ethylene oxide and propylene oxide, acetylenic glycols such as 2,4,7,9-tetraethyl-5-decyn-4,7-diol, or ethoxylated acetylenic glycols. Examples of a cationic surface-active agent include, for instance, an aliphatic mono-, di-, or polyamine such as an acetate, naphthenate or oleate; or oxygen-containing amine such as an amine oxide of polyoxyethylene alkylamine; an amide-linked amine prepared by the condensation of a carboxylic acid with a di- or polyamine; or a quaternary ammonium salt.

[0095] Examples of inert materials or inert carriers that can be used include, but are not limited to, inorganic minerals such as kaolin, phyllosilicates, carbonates, sulfates, phosphates, or botanical materials such as cork, powdered corncobs, peanut hulls, rice hulls, and walnut shells.

[0096] Herbicides that can be used with the presently described SAMPs (*e.g.*, HS peptides) include compounds that kill or inhibit growth or replication of undesired plants, typically a subset of plants that is distinct from the desired plant or crop. There are several modes of action: ACCase inhibition, carotenoid biosynthesis inhibition, cell wall synthesis inhibition, ALS inhibition, ESP synthase inhibition, glutamine synthase inhibition, HPPD inhibition, microtubule assembly inhibition, PPO inhibition, etc. Examples of commercially available herbicides include One-Time®, MSMA, Corvus®, Volunteer®, Escalade®, Q4®, Raptor®, Acumen®, Sencor®, Bullet®, TopNotch®, Valor®, PastureGard®, glyphosate (Roundup®), DSMA, Break-Up®, Hyvar®, Barricade®, etc. Herbicides can be mixed with “herbicide safeners” to reduce general toxicity of the herbicide, as described, *e.g.*, in Riechers *et al.* (2010) *Plant Physiol.* 153:3.

[0097] Pesticides (*e.g.*, nematocides, molluscicides, insecticides, miticide/ acaricides) can be used in combination with the presently disclosed SAMPs (*e.g.*, HS peptides) to kill or reduce the population of undesirable pests affecting the plant. Pesticides can also be used with repellants or pheromones to disrupt mating behavior. Insecticides are directed to insects, and include, *e.g.*, those of botanical origin (*e.g.*, allicin, nicotine, oxymatrine, jasmolin I and II, quassia, rhodajaponin III, and limonene), carbamate insecticides (*e.g.*, carbaryl, carbofuran, carbosulfan, oxamyl, nitrilacarb, CPMC, EMPC, fenobucarb), fluorine insecticides, formamidine insecticides, fumigants (*e.g.*, ethylene oxide, methyl bromide, carbon disulfide), chitin synthesis inhibitors, macrocyclic lactone insecticides, neonicotinoid insecticides, organophosphate insecticides, urea and thiourea insecticides, etc. Nematocides affect nematodes, and include, *e.g.*, organophosphorus nematocides (*e.g.*, diamidafos, fosthiazate, heterophos, phsphamidon, triazophos), fumigant nematocides (*e.g.*, carbon disulfide, methyl bromide, methyl iodide), abamectin, carvacrol, carbamate nematocides (*e.g.*, benomyl, oxamyl), etc. Molluscicides are directed to slugs and snails, and include, *e.g.*, allicin, bromoacetamide, thiocarb, trifenmorph, fentin, copper sulfate, etc. Many pesticides target more than one type of pest, so that one or two can be selected to target insects, mollusks, nematodes, mitogens, etc.

[0098] Fertilizers typically provide macro- and micronutrients in a form that they can be utilized by the plant, or a plant-associated organism. These include, *e.g.*, nitrogen, phosphorus, potassium, sulfur, calcium, boron, chlorine, copper, iron, manganese, molybdenum, zinc, nickel, and selenium. Fertilizers are often tailored to specific soil conditions or for particular crops or plants. Fertilizers that can be used include naturally-occurring, modified, concentrated and/ or chemically synthesized materials, *e.g.*, manure, bone meal, compost, fish meal, wood chips, etc., or can be chemically synthesized, UAN, anhydrous ammonium nitrate,

urea, potash, etc. Suppliers include Scott®, SureCrop®, BCF®, RVR®, Gardenline®, and many others known in the art.

[0099] Fungicides are compounds that can kill fungi or inhibit fungal growth or replication. Fungicides that can be used with the presently disclosed SAMPs (*e.g.*, HS peptides) include contact, translaminar, and systemic fungicides. Examples include sulfur, neem oil, rosemary oil, jojoba, tea tree oil, *Bacillus subtilis*, *Ulocladium*, cinnamaldehyde, etc.

[0100] The agricultural compositions of the disclosure can be in a suitable form for direct application or as a concentrate of primary composition that requires dilution with a suitable quantity of water or other diluent before application. The concentration of the SAMP (*e.g.*, an HS peptide) in the agricultural composition will vary depending upon the nature of the particular formulation, specifically, whether it is a concentrate or to be used directly, the type of plant, and in some cases, on the nature of the use, *e.g.*, for preventing a plant that is at risk of a *Liberibacter* disease (*e.g.*, HLB) or for treating a plant that is already infected with a *Liberibacter* disease (*e.g.*, HLB).

IV. Methods of preventing or treating a bacterial disease

[0101] As described herein, SAMPs may be used to prevent or treat a bacterial disease, *e.g.*, a Gram-negative bacterial disease. A *Liberibacter* disease refers to an infection caused by Gram-negative bacteria in the genus *Liberibacter* (*e.g.*, *Candidatus Liberibacter* species or *Liberibacter crescens*). A *Liberibacter* disease may infect plants such as citrus plants (*e.g.*, orange, grapefruit, tangerine, lemon, lime, key lime, papaya, citron, and pomelo) and solanaceous plants (*e.g.*, potato, tomato, eggplant, and pepper). Huanglongbing (HLB) is a type of *Liberibacter* disease that infects citrus plants. Potato Zebra Chip (ZC) disease is a type of *Liberibacter* disease that infects potato plants. The infection is vectored and transmitted by potato psyllids (*e.g.*, *Bactericera cockerelli*). The methods of utilizing the SAMPs disclosed herein may also be used to prevent or treat other bacterial diseases (*e.g.*, other Gram-negative bacterial diseases), such as those caused by *Agrobacterium tumefaciens* (also known as *Rhizobium radiobacter*) and *Pseudomonas syringae*.

HLB

[0102] The present disclosure also provides methods of preventing or treating HLB in plants. In some embodiments of the methods, the plants with HLB may be contacted with a SAMPs (*e.g.*, an HS peptide) described herein (*e.g.*, a SAMP comprising at least 75% sequence identity to the sequence of SEQ ID NO:1 or 2) or an agricultural composition comprising one or more SAMPs (*e.g.*, HS peptides) described herein. In some embodiments, the SAMP or agricultural

composition may be injected into the trunk of the plant. In other embodiments, the SAMP or agricultural composition may be injected into the stem of the plant. In yet other embodiments, the SAMP or agricultural composition may be foliar sprayed onto the plant. In yet other embodiments, the SAMP or agricultural composition may be applied by dripping irrigation to the plant. Once the plants are contacted with the SAMPs (*e.g.*, HS peptides) described herein, the peptides may enhance HLB resistance or HLB tolerance of the plants, thus, preventing or treating HLB in the plants.

[0103] The methods described herein can be used to reduce symptoms caused by HLB, including yellowing of leaves, blotchy mottle of the leaves, zinc-deficiency-like mottle, severe chlorosis, and reduced fruit yield. It will be understood that symptoms of HLB vary according to the time of infection, stage of the disease, tree species, and tree maturity, among other things. It will be further understood that in some embodiments, the disclosed methods may not necessarily result in eradication or cure of the infection but can significantly reduce the symptoms caused by HLB.

[0104] Thus, in some embodiments, the methods provided herein reduce the symptoms of HLB by reducing the yellowing of leaves, resulting in a greener appearance, increasing the growth rate of the plant, and/or increasing the fruit yield of the plant. Thus, in some embodiments, the fruit yield is improved by 5%, 10%, 20%, 30%, 40%, 50%, 60% 70%, 80%, 90%, 100%, 200%, 500% or more compared to a plant that is not treated according to the methods. In some embodiments, the fruit yield is increased to 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, or 100% of the yield of a similar plant that was not infected by HLB.

[0105] The methods described herein may also be used to prevent HLB-infection in plants. For example, a plant that is not yet infected with HLB, but is at risk for infection (*i.e.*, the plant is located in an area where HLB is identified in surrounding plants) may be contacted by one or more SAMPs (*e.g.*, HS peptides) as described in the methods of the disclosure. The plant at risk for HLB may also be genetically modified to express one or more SAMPs (*e.g.*, HS peptides) described herein to prevent HLB.

Potato ZC disease

[0106] The present disclosure also provides methods of preventing or treating potato ZC disease in potato plants. In some embodiments of the methods, the plants with potato ZC disease may be contacted with a SAMP (*e.g.*, an HS peptide) described herein (*e.g.*, a SAMP comprising at least 75% sequence identity to the sequence of SEQ ID NO:1 or 2) or an agricultural composition comprising one or more SAMPs (*e.g.*, HS peptides) described herein.

In some embodiments, the SAMP or agricultural composition may be injected into the tuber of the plant. In other embodiments, the SAMP or agricultural composition may be applied to the roots of the plants. In yet other embodiments, the SAMP or agricultural composition may be foliar sprayed onto the plant. Once the plants are contacted with the SAMPs (*e.g.*, HS peptides) described herein, the peptides may enhance potato ZC disease resistance or potato ZC disease tolerance of the plants, thus, preventing or treating potato ZC disease in the plants.

[0107] The methods described herein can be used to reduce symptoms caused by potato ZC disease, including chlorosis, leaf scorching, swollen nodes, vascular tissue browning, curled leaves, collapsed stolons, enlarged lenticels, vascular tissue browning, medullary ray discoloration, and necrotic flecking of tuber tissue. It will be understood that symptoms of potato ZC disease vary according to the time of infection, stage of the disease, plant species, and maturity, among other things. It will be further understood that in some embodiments, the disclosed methods may not necessarily result in eradication or cure of the infection, but can significantly reduce the symptoms caused by potato ZC disease.

[0108] Thus, in some embodiments, the methods provided herein reduce the symptoms of potato ZC disease as described above, resulting in a more healthy appearance, increasing the growth rate of the plant, and/or increasing the yield of the plant. Thus, in some embodiments, the yield is improved by 5%, 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 100%, 200%, 500% or more compared to a plant that is not treated according to the methods. In some embodiments, the yield is increased to 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, or 100% of the yield of a similar plant that was not infected by potato ZC disease.

[0109] The methods described herein may also be used to prevent potato ZC disease-infection in plants. For example, a plant that is not yet infected with potato ZC disease, but is at risk for infection (*i.e.*, the plant is located in an area where potato ZC disease is identified in surrounding plants) may be contacted by one or more SAMPs (*e.g.*, HS peptides) as described in the methods of the disclosure. The plant at risk for potato ZC disease may also be genetically modified to express one or more SAMPs (*e.g.*, HS peptides) described herein to prevent potato ZC disease.

V. Production of plants comprising SAMPs

[0110] In another aspect, the present disclosure provides for transgenic plants comprising recombinant expression cassettes for expressing a SAMP (*e.g.*, an HS peptide) as described herein in a plant. In some embodiments, a transgenic plant is generated that contains a complete or partial sequence of a polynucleotide that is derived from a species other than the

species of the transgenic plant. It should be recognized that transgenic plants encompass the plant or plant cell in which the expression cassette is introduced as well as progeny of such plants or plant cells that contain the expression cassette, including the progeny that have the expression cassette stably integrated in a chromosome.

[0111] A recombinant expression vector comprising a SAMP (*e.g.*, an HS peptide) coding sequence driven by a heterologous promoter may be introduced into the genome of the desired plant host by a variety of conventional techniques. For example, the DNA construct may be introduced directly into the genomic DNA of the plant cell using techniques such as electroporation and microinjection of plant cell protoplasts, or the DNA construct can be introduced directly to plant tissue using ballistic methods, such as DNA particle bombardment. An exemplary vector is a viral vector that can express and optionally replicate in the plant. Exemplary viral vectors can include, for example, citrus tristeza virus (CTV) for expressing the peptide in a phloem-limited manner in citrus, or tobacco rattle virus (TRV) to express the antimicrobial peptides in potato or other plants. Alternatively, the DNA construct may be combined with suitable T-DNA flanking regions and introduced into a conventional *Agrobacterium tumefaciens* host vector. The virulence functions of the *Agrobacterium tumefaciens* host will direct the insertion of the construct and adjacent marker into the plant cell DNA when the cell is infected by the bacteria. While transient expression of the constitutively active SAMP is encompassed by the disclosure, generally, expression of a construct of the disclosure will be from insertion of expression cassettes into the plant genome, *e.g.*, such that at least some plant offspring also contain the integrated expression cassette.

[0112] Microinjection techniques are also useful for this purpose. These techniques are well known in the art and thoroughly described in the literature. The introduction of DNA constructs using polyethylene glycol precipitation is described in Paszkowski *et al. EMBO J.* 3:2717-2722 (1984). Electroporation techniques are described in Fromm *et al. Proc. Natl. Acad. Sci. USA* 82:5824 (1985). Ballistic transformation techniques are described in Klein *et al. Nature* 327:70-73 (1987).

[0113] *Agrobacterium tumefaciens*-mediated transformation techniques, including disarming and use of binary vectors, are well described in the scientific literature. See, for example, Horsch *et al. Science* 233:496-498 (1984), and Fraley *et al. Proc. Natl. Acad. Sci. USA* 80:4803 (1983).

[0114] Transformed plant cells derived by any of the above transformation techniques can be cultured to regenerate a whole plant that possesses the transformed genotype and thus the

desired phenotype, *e.g.*, resistance or tolerance to a *Liberibacter* disease (*e.g.*, HLB). Such regeneration techniques rely on manipulation of certain phytohormones in a tissue culture growth medium, typically relying on a biocide and/or herbicide marker which has been introduced together with the desired nucleotide sequences. Plant regeneration from cultured protoplasts is described in Evans *et al.*, Protoplasts Isolation and Culture, Handbook of Plant Cell Culture, pp. 124-176, MacMillan Publishing Company, New York, 1983; and Binding, Regeneration of Plants, Plant Protoplasts, pp. 21-73, CRC Press, Boca Raton, 1985. Regeneration can also be obtained from plant callus, explants, organs, or parts thereof. Such regeneration techniques are described generally in Klee *et al. Ann. Rev. of Plant Phys.* 38:467-486 (1987).

[0115] One of skill in the art will recognize that after the expression cassette is stably incorporated in transgenic plants and confirmed to be operable, it can be introduced into other plants by sexual crossing. Any of a number of standard breeding techniques can be used, depending upon the species to be crossed.

[0116] The expression cassettes and other constructs of the disclosure can be used to confer *Liberibacter* disease resistance or tolerance on essentially any plant. Thus, the disclosure has use over a broad range of plants, including species from the genus *Citrus* (*e.g.*, *Citrus maxima*, *Citrus medica*, *Citrus micrantha*, *Citrus reticulata*, *Citrus aurantiifolia*, *Citrus aurantium*, *Citrus latifolia*, *Citrus limon*, *Citrus limonia*, *Citrus paradise*, *Citrus sinensis*, and *Citrus tangerine*) or species from the family Solanaceae (*e.g.*, *Solanum* spp., *Capsicum* spp., and *Nicotiana* spp.). Species from the genus *Solanum* include, *e.g.*, *Solanum tuberosum*, *Solanum lycopersicum*, *Solanum melongena*, *Solanum aviculare*, *Solanum capsicastrum*, *Solanum crispum*, *Solanum laciniatum*, *Solanum laxum*, *Solanum pseudocapsicum*, *Solanum rantonnetii*, *Solanum seaforthianum*, and *Solanum wendlandii*. Species from the genus *Capsicum* include, *e.g.*, *Capsicum annuum*, *Capsicum baccatum*, *Capsicum campylopodium*, *Capsicum cardenasii*, *Capsicum chacoense*, *Capsicum cornutum*, *Capsicum dusenii*, *Capsicum eximium*, *Capsicum friburgense*, *Capsicum frutescens*, *Capsicum geminifolium*, *Capsicum havanense*, , *Capsicum lanceolatum*, *Capsicum lycianthoides*, *Capsicum minutiflorum*, *Capsicum mositicum*, *Capsicum pubescens*, *Capsicum recurvatum*, *Capsicum schottianum*, *Capsicum spina-alba*, *Capsicum tovarii*, and *Capsicum villosum*. Species from the genus *Nicotiana* include, *e.g.*, *Nicotiana acuminata*, *Nicotiana benthamiana*, *Nicotiana glauca*, *Nicotiana longiflora*, *Nicotiana rustica*, *Nicotiana tabacum*, and *Nicotiana occidentalis*.

[0117] In particular embodiments, the plant is selected from the group consisting of *Citrus reticulata*, *Citrus sinensis*, *Citrus clementina*, *Capsicum annuum*, *Solanum tuberosum*, *Solanum lycopersicum*, *Solanum melongena*, and *Nicotiana benthamiana*. In particular embodiments, the plant is a sweet orange plant (*Citrus sinensis*). In particular embodiments, the plant is a clementine plant (*Citrus clementina*). In particular embodiments, the plant is a potato plant (*Solanum tuberosum*). In some embodiment, the plant is a vegetable- or fruit-producing plant.

[0118] Those of skill will recognize that a number of plant species can be used as models to predict the phenotypic effects of transgene expression in other plants. For example, it is well recognized that both tobacco (*Nicotiana*) and *Arabidopsis* plants are useful models of transgene expression, particularly in other dicots.

[0119] In some embodiments, the plants of the disclosure have enhanced SAMP-mediated phenotypes, for example enhanced bacterial disease (e.g., a *Liberibacter* disease (e.g., HLB and ZC) and other bacterial diseases such as those caused by *Agrobacterium tumefaciens* (also known as *Rhizobium radiobacter*) and *Pseudomonas syringae*) resistance or tolerance, as compared to plants that are otherwise identical except for expression of the SAMP.

CRISPR/Cas

[0120] Plant gene manipulations can now be precisely tailored in non-transgenic organisms using the CRISPR/Cas9 genome editing method. In this bacterial antiviral and transcriptional regulatory system, a complex of two small RNAs – the CRISPR-RNA (crRNA) and the transactivating crRNA (tracrRNA) – directs the nuclease (Cas9) to a specific DNA sequence complementary to the crRNA (Jinek, M., *et al. Science* 337, 816–821 (2012)). Binding of these RNAs to Cas9 involves specific sequences and secondary structures in the RNA. The two RNA components can be simplified into a single element, the single guide-RNA (sgRNA), which is transcribed from a cassette containing a target sequence defined by the user (Jinek, M., *et al. Science* 337, 816–821 (2012)). This system has been used for genome editing in humans, zebrafish, *Drosophila*, mice, nematodes, bacteria, yeast, and plants (Hsu, P.D., *et al., Cell* 157, 1262–1278 (2014)). In this system the nuclease creates double stranded breaks at the target region programmed by the sgRNA. These can be repaired by non-homologous recombination, which often yields inactivating mutations. The breaks can also be repaired by homologous recombination, which enables the system to be used for gene targeted gene replacement (Li, J.-F., *et al. Nat. Biotechnol.* 31, 688–691, 2013; Shan, Q., *et al. Nat. Biotechnol.* 31, 686–688, 2013). In some embodiments of the methods in the present disclosure, a gene encoding a wild-

type or endogenous SAMP in a plant may be modified using the CAS9/CRISPR system to match the polynucleotide sequence encoding a SAMP described herein (*e.g.*, a polynucleotide sequence encoding the SAMP having at least 75% sequence identity or at least one amino acid substitution relative to the sequence of any one of SEQ ID NOs:1-13 and 35-37 (*e.g.*, SEQ ID NOs:1 and 2)). In some embodiments, a gene encoding a wild-type or endogenous SAMP in a plant may be modified using the CAS9/CRISPR system to match the polynucleotide sequence of any one of SEQ ID NOs:14-26, 28-34, and 38-40 (*e.g.*, SEQ ID NO:14 and 15).

[0121] Accordingly, in some embodiments, instead of generating a transgenic plant, a native SAMP coding sequence in a plant or plant cell can be altered *in situ* to generate a plant or plant cell carrying a polynucleotide encoding a SAMP described herein (*e.g.*, a SAMP having at least 75% sequence identity or at least one amino acid substitution relative to the sequence of any one of SEQ ID NOs:1-13 and 35-37 (*e.g.*, SEQ ID NOs:1 and 2)). For example, in some embodiments, CRISPR technology is used to introduce one or more nucleotide changes into a SAMP coding sequence *in situ* to change the appropriate codon to make a change corresponding to positions X₁ to X₂₅ as set forth in the sequence of SEQ ID NO:27. The CRISPR/Cas system has been modified for use in prokaryotic and eukaryotic systems for genome editing and transcriptional regulation. The “CRISPR/Cas” system refers to a widespread class of bacterial systems for defense against foreign nucleic acid. CRISPR/Cas systems are found in a wide range of eubacterial and archaeal organisms. CRISPR/Cas systems include type I, II, and III sub-types. Wild-type type II CRISPR/Cas systems utilize the RNA-mediated nuclease, Cas9 in complex with guide and activating RNA to recognize and cleave foreign nucleic acid. Cas9 homologs are found in a wide variety of eubacteria, including, but not limited to bacteria of the following taxonomic groups: *Actinobacteria*, *Aquificae*, *Bacteroidetes-Chlorobi*, *Chlamydiae-Verrucomicrobia*, *Chloroflexi*, *Cyanobacteria*, *Firmicutes*, *Proteobacteria*, *Spirochaetes*, and *Thermotogae*. An exemplary Cas9 protein is the *Streptococcus pyogenes* Cas9 protein. Additional Cas9 proteins and homologs thereof are described in, *e.g.*, Chylinski, *et al.*, RNA Biol. 2013 May 1; 10(5): 726–737 ; Nat. Rev. Microbiol. 2011 June; 9(6): 467-477; Hou, *et al.*, Proc Natl Acad Sci U S A. 2013 Sep 24;110(39):15644-9; Sampson *et al.*, Nature. 2013 May 9;497(7448):254-7; and Jinek, *et al.*, Science. 2012 Aug 17;337(6096):816-21.

[0122] Accordingly, in one aspect, a method is provided of using CRISPR/CAS9 to introduce at least one mutation into a plant cell is performed. In some embodiments, a method of altering a (*e.g.*, native) nucleic acid encoding SAMP in a plant is provided. In some embodiments, the method comprises introducing into the plant cell containing and expressing a

DNA molecule having a target nucleic acid encoding SAMP an engineered, non-naturally occurring Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)—CRISPR associated (Cas) (CRISPR-Cas) system. In some embodiments, the CRISPR-Cas system comprises one or more vectors comprising: a) a first regulatory element operable in a plant cell operably linked to at least one nucleotide sequence encoding a CRISPR-Cas system guide RNA that hybridizes with the target sequence, and b) a second regulatory element operable in a plant cell operably linked to a nucleotide sequence encoding a Type-II Cas9 protein, wherein components (a) and (b) are located on same or different vectors of the system, whereby the guide RNA targets the target sequence and the Cas9 protein cleaves the DNA molecule, whereby at least one mutation is introduced into the target nucleic acid encoding the SAMP, *i.e.*, one or more mutations are introduced into the target nucleic acid encoding the SAMPs to alter the sequence of the target nucleic acid to match the polynucleotide sequence encoding an antimicrobial (*e.g.*, antibacterial) peptide having the sequence of any one of SEQ ID NOs:1-13 and 35-37 (*e.g.*, SEQ ID NOs:1 and 2). In some embodiments, the SAMP is selected from any of SEQ ID NOs:1-5 and 9, or a substantially identical polypeptide thereof.

[0123] In some embodiments, methods of using CRISPR/CAS9 technology to introduce at least one mutation into a (*e.g.*, native) nucleic acid encoding SAMP may be applied to a broad range of plants, including species from the genus *Citrus* (*e.g.*, *Citrus maxima*, *Citrus medica*, *Citrus micrantha*, *Citrus reticulata*, *Citrus aurantiifolia*, *Citrus aurantium*, *Citrus latifolia*, *Citrus limon*, *Citrus limonia*, *Citrus paradise*, *Citrus sinensis*, and *Citrus tangerine*) or species from the family Solanaceae (*e.g.*, *Solanum* spp., *Capsicum* spp., and *Nicotiana* spp.). Species from the genus *Solanum* include, *e.g.*, *Solanum tuberosum*, *Solanum lycopersicum*, *Solanum melongena*, *Solanum aviculare*, *Solanum capsicastrum*, *Solanum crispum*, *Solanum laciniatum*, *Solanum laxum*, *Solanum pseudocapsicum*, *Solanum rantonnetii*, *Solanum seaforthianum*, and *Solanum wendlandii*. Species from the genus *Capsicum* include, *e.g.*, *Capsicum annuum*, *Capsicum baccatum*, *Capsicum campylopodium*, *Capsicum cardenasii*, *Capsicum chacoense*, *Capsicum cornutum*, *Capsicum dusenii*, *Capsicum eximium*, *Capsicum friburgense*, *Capsicum frutescens*, *Capsicum geminifolium*, *Capsicum havanense*, , *Capsicum lanceolatum*, *Capsicum lycianthoides*, *Capsicum minutiflorum*, *Capsicum mositicum*, *Capsicum pubescens*, *Capsicum recurvatum*, *Capsicum schottianum*, *Capsicum spina-alba*, *Capsicum tovarii*, and *Capsicum villosum*. Species from the genus *Nicotiana* include, *e.g.*, *Nicotiana acuminata*, *Nicotiana benthamiana*, *Nicotiana glauca*, *Nicotiana longiflora*, *Nicotiana rustica*, *Nicotiana tabacum*, and *Nicotiana occidentalis*. In particular embodiments, the plant is selected from the group consisting of *Citrus reticulata*, *Citrus sinensis*, *Citrus clementina*, *Capsicum annuum*, *Solanum*

tuberosum, *Solanum lycopersicum*, *Solanum melongena*, and *Nitotiana benthamiana*. In particular embodiments, the plant is a sweet orange plant (*Citrus sinensis*). In particular embodiments, the plant is a clementine plant (*Citrus clementina*). In particular embodiments, the plant is a potato plant (*Solanum tuberosum*). In some embodiment, the plant is a vegetable- or fruit-producing plant.

[0124] In some embodiments, the mutation(s) introduced to the target nucleic acid sequence change the appropriate codon in the sequence to make change(s) corresponding to positions X₁ to X₂₅ as set forth in the sequence of SEQ ID NO:27. For example, after introducing the mutations to the target nucleic acid to change the appropriate codons, the modified nucleic acid sequence encode, at its corresponding positions, one or more amino acids as set forth in positions X₁ to X₂₅ of SEQ ID NO:27. Also provided as a plant or plant cell resulting from the above-described method. Such a plant will contain a non-naturally-occurring nucleic acid sequence encoding the SAMP.

VI. Expression Cassettes

[0125] In some embodiments, the present disclosure provides expression cassettes comprising a polynucleotide encoding a SAMP (*e.g.*, an HS peptide) of the disclosure, wherein introduction of the expression cassette into a plant results in a transgenic plant expressing the SAMP. In some embodiments, a promoter may be operably linked to the polynucleotide encoding the SAMP. The promoter may be heterologous to the polynucleotide. In some embodiments, the promoter may be inducible. In some embodiments, the promoter may plant tissue-specific (*e.g.*, phloem-specific, tuber-specific, root-specific, stem-specific, trunk-specific, or leaf-specific).

[0126] Any of a number of means well known in the art can be used to drive SAMP expression in plants. Any organ can be targeted, such as shoot vegetative organs/structures (*e.g.* leaves, stems, and tubers), roots, flowers and floral organs/structures (*e.g.* bracts, sepals, petals, stamens, carpels, anthers and ovules), seed (including embryo, endosperm, and seed coat) and fruit. Alternatively, the polynucleotide encoding a SAMP (*e.g.*, an HS peptide) described herein can be expressed specifically in certain cell and/or tissue types within one or more organs (*e.g.*, guard cells in leaves using a guard cell-specific promoter). Alternatively, the polynucleotide encoding a SAMP (*e.g.*, an HS peptide) described herein can be expressed constitutively (*e.g.*, using the CaMV 35S promoter).

[0127] To use a polynucleotide encoding a SAMP (*e.g.*, an HS peptide) described herein in the above techniques, recombinant DNA vectors suitable for transformation of plant cells are

prepared. Techniques for transforming a wide variety of higher plant species are well known and described in the technical and scientific literature. *See, e.g., Weising et al. Ann. Rev. Genet.* 22:421-477 (1988). A DNA sequence coding for the SAMP preferably will be combined with transcriptional and translational initiation regulatory sequences which will direct the transcription of the sequence from the gene in the intended tissues of the transformed plant.

[0128] For example, a plant promoter fragment may be employed to direct expression of the SAMP (*e.g.*, an HS peptide) in all tissues of a transgenic plant. Such promoters are referred to herein as "constitutive" promoters and are active under most environmental conditions and states of development or cell differentiation. Examples of constitutive promoters include the cauliflower mosaic virus (CaMV) 35S transcription initiation region, the 1'- or 2'- promoter derived from T-DNA of *Agrobacterium tumefaciens*, and other transcription initiation regions from various plant genes known to those of skill.

[0129] Alternatively, the plant promoter may direct expression of the SAMP (*e.g.*, an HS peptide) in a specific tissue (tissue-specific promoters) or may be otherwise under more precise environmental control (inducible promoters). Examples of tissue-specific promoters under developmental control include promoters that initiate transcription only in certain tissues, such as phloem, tubers, stems, trunks, leaves, or guard cells. Examples of environmental conditions that may affect transcription by inducible promoters include, but are not limited to, anaerobic conditions, elevated temperature, and the presence of light.

[0130] In some embodiments, a polyadenylation region at the 3'-end of the coding region of the SAMP may be included. The polyadenylation region can be derived from a naturally occurring SAMP gene, from a variety of other plant genes, or from T-DNA.

[0131] The vector comprising the polynucleotide sequences (*e.g.*, promoters or SAMP coding regions) may include a marker gene that confers a selectable phenotype on plant cells. For example, the marker may encode biocide resistance, particularly antibiotic resistance, such as resistance to kanamycin, G418, bleomycin, hygromycin, or herbicide resistance, such as resistance to chlorosulfuron or Basta.

[0132] In some embodiments, the polynucleotide encoding the SAMP (*e.g.*, an HS peptide) is expressed recombinantly in plant cells. A variety of different expression constructs, such as expression cassettes and vectors suitable for transformation of plant cells, can be prepared. Techniques for transforming a wide variety of higher plant species are well known and

described in the technical and scientific literature. *See, e.g., Weising et al. Ann. Rev. Genet.* 22:421-477 (1988). A DNA sequence coding for a SAMP (*e.g.,* an HS peptide) described herein can be combined with *cis*-acting (promoter) and *trans*-acting (enhancer) transcriptional regulatory sequences to direct the timing, tissue type, and levels of transcription in the intended tissues of the transformed plant. Translational control elements can also be used.

[0133] Embodiments of the present disclosure also provide for a polynucleotide encoding the SAMP (*e.g.,* an HS peptide) to be operably linked to a promoter which, in some embodiments, is capable of driving the transcription of the SAMP coding sequence in plants. The promoter can be, *e.g.,* derived from plant or viral sources. The promoter can be, *e.g.,* constitutively active, inducible, or tissue specific. In construction of recombinant expression cassettes, vectors, transgenics, of the disclosure, a different promoter can be chosen and employed to differentially direct gene expression, *e.g.,* in some or all tissues of a plant or animal.

Constitutive promoters

[0134] A fragment can be employed to direct expression of a polynucleotide encoding the SAMP (*e.g.,* an HS peptide) in all transformed cells or tissues, *e.g.,* as those of a transgenic plant. The term "constitutive regulatory element" means a regulatory element that confers a level of expression upon an operatively linked nucleic molecule that is relatively independent of the cell or tissue type in which the constitutive regulatory element is expressed. A constitutive regulatory element that is expressed in a plant generally is widely expressed in a large number of cell and tissue types. Promoters that drive expression continuously under physiological conditions are referred to as "constitutive" promoters and are active under most environmental conditions and states of development or cell differentiation.

[0135] A variety of constitutive regulatory elements useful for ectopic expression in a transgenic plant are well known in the art. The cauliflower mosaic virus 35S (CaMV 35S) promoter, for example, is a well-characterized constitutive regulatory element that produces a high level of expression in all plant tissues (Odell *et al., Nature* 313:810-812 (1985)). The CaMV 35S promoter can be particularly useful due to its activity in numerous diverse plant species (Benfey and Chua, *Science* 250:959-966 (1990); Futterer *et al., Physiol. Plant* 79:154 (1990); Odell *et al., supra*, 1985). A tandem 35S promoter, in which the intrinsic promoter element has been duplicated, confers higher expression levels in comparison to the unmodified 35S promoter (Kay *et al., Science* 236:1299 (1987)). Other useful constitutive regulatory elements include, for example, the cauliflower mosaic virus 19S promoter; the Figwort mosaic

virus promoter; and the nopaline synthase (nos) gene promoter (Singer *et al.*, *Plant Mol. Biol.* 14:433 (1990); An, *Plant Physiol.* 81:86 (1986)).

[0136] Additional constitutive regulatory elements including those for efficient expression in monocots also are known in the art, for example, the pEmu promoter and promoters based on the rice Actin-1 5' region (Last *et al.*, *Theor. Appl. Genet.* 81:581 (1991); Mcelroy *et al.*, *Mol. Gen. Genet.* 231:150 (1991); Mcelroy *et al.*, *Plant Cell* 2:163 (1990)). Chimeric regulatory elements, which combine elements from different genes, also can be useful for ectopically expressing a nucleic acid molecule encoding a SAMP (*e.g.*, an HS peptide) described herein (Comai *et al.*, *Plant Mol. Biol.* 15:373 (1990)).

[0137] Other examples of constitutive promoters include the 1'- or 2'- promoter derived from T-DNA of *Agrobacterium tumefaciens* (see, *e.g.*, Mengiste (1997) *supra*; O'Grady (1995) *Plant Mol. Biol.* 29:99-108); actin promoters, such as the *Arabidopsis* actin gene promoter (see, *e.g.*, Huang (1997) *Plant Mol. Biol.* 1997 33:125-139); alcohol dehydrogenase (Adh) gene promoters (see, *e.g.*, Millar (1996) *Plant Mol. Biol.* 31:897-904); *ACT11* from *Arabidopsis* (Huang *et al.* *Plant Mol. Biol.* 33:125-139 (1996)), *Cat3* from *Arabidopsis* (GenBank No. U43147, Zhong *et al.*, *Mol. Gen. Genet.* 251:196-203 (1996)), the gene encoding stearyl-acyl carrier protein desaturase from *Brassica napus* (Genbank No. X74782, Solocombe *et al.* *Plant Physiol.* 104:1167-1176 (1994)), *Gpc1* from maize (GenBank No. X15596, Martinez *et al.* *J. Mol. Biol.* 208:551-565 (1989)), *Gpc2* from maize (GenBank No. U45855, Manjunath *et al.*, *Plant Mol. Biol.* 33:97-112 (1997)), other transcription initiation regions from various plant genes known to those of skill. See also Holtorf *Plant Mol. Biol.* 29:637-646 (1995).

Inducible promoters

[0138] Alternatively, a plant promoter may direct expression of the polynucleotide encoding the SAMP (*e.g.*, an HS peptide) under the influence of changing environmental conditions or developmental conditions. Examples of environmental conditions that may affect transcription by inducible promoters include anaerobic conditions, elevated temperature, drought, or the presence of light. Such promoters are referred to herein as "inducible" promoters. In some embodiments, an inducible promoter is one that is induced by one or more environmental stressors, including but not limited to, drought, freezing cold, and high salt. For example, the disclosure can incorporate a drought-specific promoter such as a drought-inducible promoter of maize (*e.g.*, the maize rab17 drought-inducible promoter (Vilardell *et al.* (1991) *Plant Mol. Biol.* 17:985-993; Vilardell *et al.* (1994) *Plant Mol. Biol.* 24:561-569)); or alternatively a cold, drought, and high salt inducible promoter from potato (Kirch (1997) *Plant Mol. Biol.*

33:897-909) or from *Arabidopsis* (e.g., the rd29A promoter (Kasuga *et al.* (1999) *Nature Biotechnology* 17:287-291). Other environmental stress-inducible promoters include promoters from the following genes: *Rab21*, *Wsi18*, *Lea3*, *Uge1*, *Dip1*, and *R1G1B* in rice (Yi *et al.* (2010) *Planta* 232:743-754).

[0139] In some embodiments, a plant promoter is a stress-inducible promoter (e.g., a drought-, cold-, or salt-inducible promoter) that comprises a dehydration-responsive element (DRE) and/or an ABA-responsive element (ABRE), including but not limited to the rd29A promoter.

[0140] Alternatively, plant promoters which are inducible upon exposure to plant hormones, such as auxins, are used to express the polynucleotide encoding the SAMP (e.g., an HS peptide). For example, the disclosure can use the auxin-response elements E1 promoter fragment (AuxREs) in the soybean (*Glycine max* L.) (Liu (1997) *Plant Physiol.* 115:397-407); the auxin-responsive *Arabidopsis* GST6 promoter (also responsive to salicylic acid and hydrogen peroxide) (Chen (1996) *Plant J.* 10: 955-966); the auxin-inducible parC promoter from tobacco (Sakai (1996) 37:906-913); a plant biotin response element (Streit (1997) *Mol. Plant Microbe Interact.* 10:933-937); and, the promoter responsive to the stress hormone abscisic acid (Sheen (1996) *Science* 274:1900-1902).

[0141] Plant promoters inducible upon exposure to chemicals reagents that may be applied to the plant, such as herbicides or antibiotics, are also useful for expressing the polynucleotide encoding the SAMP (e.g., an HS peptide). For example, the maize *In2-2* promoter, activated by benzenesulfonamide herbicide safeners, can be used (De Veylder (1997) *Plant Cell Physiol.* 38:568-577); application of different herbicide safeners induces distinct gene expression patterns, including expression in the root, hydathodes, and the shoot apical meristem. A SAMP (e.g., an HS peptide) coding sequence can also be under the control of, e.g., a tetracycline-inducible promoter, e.g., as described with transgenic tobacco plants containing the *Avena sativa* L. (oat) arginine decarboxylase gene (Masgrau (1997) *Plant J.* 11:465-473); or, a salicylic acid-responsive element (Stange (1997) *Plant J.* 11:1315-1324; Uknes *et al.*, *Plant Cell* 5:159-169 (1993); Bi *et al.*, *Plant J.* 8:235-245 (1995)).

[0142] Examples of useful inducible regulatory elements include copper-inducible regulatory elements (Mett *et al.*, *Proc. Natl. Acad. Sci. USA* 90:4567-4571 (1993); Furst *et al.*, *Cell* 55:705-717 (1988)); tetracycline and chlor-tetracycline-inducible regulatory elements (Gatz *et al.*, *Plant J.* 2:397-404 (1992); Röder *et al.*, *Mol. Gen. Genet.* 243:32-38 (1994); Gatz, *Meth.*

Cell Biol. 50:411-424 (1995)); ecdysone inducible regulatory elements (Christopherson *et al.*, *Proc. Natl. Acad. Sci. USA* 89:6314-6318 (1992); Kreutzweiser *et al.*, *Ecotoxicol. Environ. Safety* 28:14-24 (1994)); heat shock inducible regulatory elements (Takahashi *et al.*, *Plant Physiol.* 99:383-390 (1992); Yabe *et al.*, *Plant Cell Physiol.* 35:1207-1219 (1994); Ueda *et al.*, *Mol. Gen. Genet.* 250:533-539 (1996)); and *lac* operon elements, which are used in combination with a constitutively expressed *lac* repressor to confer, for example, IPTG-inducible expression (Wilde *et al.*, *EMBO J.* 11:1251-1259 (1992)). An inducible regulatory element useful in the transgenic plants of the disclosure also can be, for example, a nitrate-inducible promoter derived from the spinach nitrite reductase gene (Back *et al.*, *Plant Mol. Biol.* 17:9 (1991)) or a light-inducible promoter, such as that associated with the small subunit of RuBP carboxylase or the LHCP gene families (Feinbaum *et al.*, *Mol. Gen. Genet.* 226:449 (1991); Lam and Chua, *Science* 248:471 (1990)).

Tissue-specific promoters

[0143] Alternatively, the plant promoter may direct expression of the polynucleotide encoding the SAMP (*e.g.*, an HS peptide) in a specific tissue (tissue-specific promoters). Tissue specific promoters are transcriptional control elements that are only active in particular cells or tissues at specific times during plant development, such as in vegetative tissues or reproductive tissues.

[0144] Examples of tissue-specific promoters under developmental control include promoters that initiate transcription only (or primarily only) in certain tissues, such as vegetative tissues, *e.g.*, roots or leaves, or reproductive tissues, such as fruit, ovules, seeds, pollen, pistils, flowers, or any embryonic tissue, or epidermis or mesophyll. Reproductive tissue-specific promoters may be, *e.g.*, ovule-specific, embryo-specific, endosperm-specific, integument-specific, seed and seed coat-specific, pollen-specific, petal-specific, sepal-specific, or some combination thereof. In some embodiments, the promoter is cell-type specific, *e.g.*, guard cell-specific.

[0145] Epidermal-specific promoters include, for example, the Arabidopsis *LTP1* promoter (Thoma *et al.* (1994) *Plant Physiol.* 105(1):35-45), the *CER1* promoter (Aarts *et al.* (1995) *Plant Cell* 7:2115-27), and the *CER6* promoter (Hooker *et al.* (2002) *Plant Physiol* 129:1568-80), and the orthologous tomato *LeCER6* (Vogg *et al.* (2004) *J. Exp Bot.* 55:1401-10).

[0146] Guard cell-specific promoters include, for example, the *DGPI* promoter (Li *et al.* (2005) *Science China C Life Sci.* 48:181-186).

[0147] Other tissue-specific promoters include seed promoters. Suitable seed-specific promoters are derived from the following genes: *MAC1* from maize (Sheridan (1996) *Genetics* 142:1009-1020); *Cat3* from maize (GenBank No. L05934, Abler (1993) *Plant Mol. Biol.* 22:10131-1038); *viviparous-1* from *Arabidopsis* (Genbank No. U93215); *atmyc1* from *Arabidopsis* (Urao (1996) *Plant Mol. Biol.* 32:571-57; Conceicao (1994) *Plant* 5:493-505); *napA* from *Brassica napus* (GenBank No. J02798, Josefsson (1987) *JBL* 26:12196-1301); and the napin gene family from *Brassica napus* (Sjodahl (1995) *Planta* 197:264-271).

[0148] A variety of promoters specifically active in vegetative tissues, such as leaves, stems, roots and tubers, can also be used to express polynucleotide encoding SAMPs (*e.g.*, HS peptides) described herein. For example, promoters controlling patatin, the major storage protein of the potato tuber, can be used, see, *e.g.*, Kim (1994) *Plant Mol. Biol.* 26:603-615; Martin (1997) *Plant J.* 11:53-62. The ORF13 promoter from *Agrobacterium rhizogenes* that exhibits high activity in roots can also be used (Hansen (1997) *Mol. Gen. Genet.* 254:337-343. Other useful vegetative tissue-specific promoters include: the tarin promoter of the gene encoding a globulin from a major taro (*Colocasia esculenta* L. Schott) corm protein family, tarin (Bezerra (1995) *Plant Mol. Biol.* 28:137-144); the curculin promoter active during taro corm development (de Castro (1992) *Plant Cell* 4:1549-1559) and the promoter for the tobacco root-specific gene TobRB7, whose expression is localized to root meristem and immature central cylinder regions (Yamamoto (1991) *Plant Cell* 3:371-382).

[0149] Leaf-specific promoters, such as the ribulose biphosphate carboxylase (RBCS) promoters, can also be used. For example, the tomato RBCS1, RBCS2 and RBCS3A genes are expressed in leaves and light-grown seedlings, only RBCS1 and RBCS2 are expressed in developing tomato fruits (Meier (1997) *FEBS Lett.* 415:91-95). Ribulose bisphosphate carboxylase promoters expressed almost exclusively in mesophyll cells in leaf blades and leaf sheaths at high levels, described by Matsuoka (1994) *Plant J.* 6:311-319, can be used. Another leaf-specific promoter is the light harvesting chlorophyll a/b binding protein gene promoter, see, *e.g.*, Shiina (1997) *Plant Physiol.* 115:477-483; Casal (1998) *Plant Physiol.* 116:1533-1538. The *Arabidopsis thaliana* myb-related gene promoter (Atmyb5) described by Li (1996) *FEBS Lett.* 379:117-121, is leaf-specific. The Atmyb5 promoter is expressed in developing leaf trichomes, stipules, and epidermal cells on the margins of young rosette and cauline leaves, and in immature seeds. Atmyb5 mRNA appears between fertilization and the 16-cell stage of embryo development and persists beyond the heart stage. A leaf promoter identified in maize by Busk (1997) *Plant J.* 11:1285-1295, can also be used.

[0150] Another class of useful vegetative tissue-specific promoters are meristematic (root tip and shoot apex) promoters. For example, the "*SHOOTMERISTEMLESS*" and "*SCARECROW*" promoters, which are active in the developing shoot or root apical meristems, described by Di Laurenzio (1996) *Cell* 86:423-433; and, Long (1996) *Nature* 379:66-69; can be used. Another useful promoter is that which controls the expression of 3-hydroxy-3- methylglutaryl coenzyme A reductase HMG2 gene, whose expression is restricted to meristematic and floral (secretory zone of the stigma, mature pollen grains, gynoecium vascular tissue, and fertilized ovules) tissues (see, *e.g.*, Enjuto (1995) *Plant Cell*. 7:517-527). Also useful are kn1-related genes from maize and other species which show meristem-specific expression, see, *e.g.*, Granger (1996) *Plant Mol. Biol.* 31:373-378; Kerstetter (1994) *Plant Cell* 6:1877-1887; Hake (1995) *Philos. Trans. R. Soc. Lond. B. Biol. Sci.* 350:45-51. For example, the *Arabidopsis thaliana* KNAT1 promoter (see, *e.g.*, Lincoln (1994) *Plant Cell* 6:1859-1876).

[0151] One of skill will recognize that a tissue-specific promoter may drive expression of operably linked sequences in tissues other than the target tissue. Thus, as used herein a tissue-specific promoter is one that drives expression preferentially in the target tissue, but may also lead to some expression in other tissues as well.

[0152] In another embodiment, the polynucleotide encoding the SAMP (*e.g.*, an HS peptide) is expressed through a transposable element. This allows for constitutive, yet periodic and infrequent expression of the constitutively active polypeptide. The disclosure also provides for use of tissue-specific promoters derived from viruses including, *e.g.*, the tobamovirus subgenomic promoter (Kumagai (1995) *Proc. Natl. Acad. Sci. USA* 92:1679-1683; the rice tungro bacilliform virus (RTBV), which replicates only in phloem cells in infected rice plants, with its promoter which drives strong phloem-specific reporter gene expression; the cassava vein mosaic virus (CVMV) promoter, with highest activity in vascular elements, in leaf mesophyll cells, and in root tips (Verdaguer (1996) *Plant Mol. Biol.* 31:1129-1139).

[0153] In another embodiment, the present disclosure provides for expression vectors comprising an expression cassette of the disclosure (*e.g.*, as described herein).

VII. Plants

[0154] In some embodiments, the plant is a citrus plant. In some embodiments, the citrus plant is an orange tree, a lemon tree, a lime tree, or a grapefruit tree. In one embodiment, the citrus plant is a navel orange, Valencia orange, sweet orange, mandarin orange, or sour orange. In one embodiment, the citrus plant is a lemon tree. In one embodiment, the citrus plant is a lime

tree. In some embodiments, the plant is a relative of a citrus plant, such as orange jasmine, limeberry, and trifoliate orange. In some embodiments, the plant is a potato plant.

[0155] In some embodiments, the present disclosure provides for plants (or a plant cell, seed, flower, leaf, fruit, or other plant part from such plants or processed food or food ingredient from such plants) comprising an expression cassette comprising a promoter operably linked to a polynucleotide encoding a SAMP (*e.g.*, an HS peptide) of the disclosure (*e.g.*, as described herein). In some embodiments, the plant has decreased UBC expression or activity and/or increased expression or activity of Pi transporters.

EXAMPLES

Example 1 – Expression of antimicrobial SAMP genes

[0156] The expression levels of SAMPs in HLB-susceptible and HLB-tolerant plants were detected by quantitative RT-PCR. The expression level of citrus actin was used as an internal control. The expression level of SAMPs is higher in two different HLB-tolerant/resistant varieties from totally different geographic and genetic backgrounds (a hybrid of Cleopatra mandarin (*Citrus reticulata*) and *Poncirus trifoliate* US942, and a *Eremocitrus glauca* hybrid) than in their corresponding HLB-sensitive close relatives (FIGS. 1A and 1B).

Example 2 – Effects of SAMPs in suppressing *Candidatus Liberibacter solanacearum* (*Ca. L. solanacearum*) infection

[0157] Solanaceae plant *Nicotiana benthamiana* (*Nb*) is commonly used for gene function studies against pathogen infections. *Nb* can be infected by *Ca. L. solanacearum* by being infested with *Ca. L. solanacearum*-positive potato psyllid, which is a pathosystem highly similar to potato ZC disease and citrus HLB. The effects of two SAMPs, CghSAMPa and CghSAMPb (SEQ ID NOs:1 and 2, respectively, which are hybrids from crossing *Citrus glauca* with citrus sp.), were tested using the *Nb*/potato psyllid/ *Ca. L. solanacearum* pathosystem. The CghSAMPa and CghSAMPb peptides were expressed and purified in *E.coli*. *Ca. L. solanacearum*-infected *Nb* plants were treated with 30 μ M CghSAMPa or CghSAMPb peptide, or mock solution by stem injection. The photos in FIG. 3A were taken after 3 weeks of treatment. FIG. 3A shows that the infected plants treated with the SAMPs were able to grow much better than the plants that received mock treatment. FIGS. 3B and 7 further show that the plants treated with CghSAMPa and CghSAMPb peptides had much lower bacterial titer compared to the plants treated with mock solution. The results demonstrated that the two

SAMPs from resistant/tolerant citrus rootstocks significantly controlled the titer of *Ca. L. solanacearum* in *Nb* plants and promoted plant growth.

Example 3 – Effects of SAMPs delivered via trunk delivery

[0158] We have tested effective SAMP treatment on HLB-positive citrus plants using trunk delivery method, and the bacterial titer of all the treated trees are largely reduced, about 30-100 folds. No bacteria were detected in the treated trees. All the new shoots and leaves from the treated trees were no longer symptomatic, and the new shoots and leaves from the mock treated plants still have yellow patch symptoms (Figure 4)

Example 4 – Effects of SAMPs in suppressing *Candidatus Liberibacter solanacearum* (*Ca. L. solanacearum*) infection in potato and tomato

[0159] We have also performed the effective SAMP treatment on *C.Lso*-infected potato. The disease symptom was clearly inhibited, and the yield of tuber production was increased. (Figure 5)

[0160] We also tested the effective SAMP on *C.Lso*-infected tomato, and the disease symptom was largely reduced as well. (Figure 6)

Example 5 – Effects of SAMPs in citrus plants

[0161] SAMP was apply by foliar sprayed with 5% Southern Ag Methylated Seed Oil (MSO) on the 1 year old seedlings. The expression level of defense response marker genes CsPR1, CsPR2, and CAPAL were evaluated by qRT-PCR with the ubiquitin gene as an internal control for up to 7 days after treatment. FIGS. 8A-8C show that SAMPs primed the citrus plants to have increased expression of the defense response marker genes.

[0162] Further, different concentrations of SAMP solutions were infiltrated into sweet orange leaves. No cell death was observed with 30 μ M treatment. FIGS. 9A and 9B show that SAMPs have low phytotoxic activity on citrus leaves.

[0163] Moreover, mRNA expression analysis (FIG. 10) demonstrates that SAMPs are highly expressed in the fruit of Australian finger lime, Australian desert lime, lemon, and *Poncirus trifoliata* (common root stock), which have already been consumed by humans for hundreds of years. The mRNA expression was detected by RT-PCR.

Example 6 – Protease digestion of SAMPs

[0164] FIG. 11 shows that SAMPs are sensitive to human protease pepsin, a major gastric enzyme. 20 µg of SAMP were incubated with 0.4% solution of pepsin in 10 mM HCl at 37 °C. The reaction was analyzed with 18% SDS-PAGE gel and visualized with coomassie blue staining. SAMP was completely digested within 45 min.

Example 7 – Stability of SAMPs

[0165] SAMPs were incubated at room temperature (RT) for 24 hours, 60 °C for 24 hours, or 100 °C for 20 mins. Subsequently, the SAMPs were used for viability/cytotoxicity assay. The assay was done by incubating 10⁷ cells/mL of *Liberbacter crescens* with the pre-treated SAMP or buffer only as mock treatment for 2 hours. The samples were then stained with DMAO (green) and EthD-III (orange), which represent live and dead cells, respectively. FIG. 12 shows that SAMPs are stable at up to 100 °C.

[0166] Further, to investigate the stability of SAMPs in citrus plants, 20 µg of SAMPs were incubated with 200 µg fresh citrus lysate in 1x PBS buffer at room temperature. The reaction was analyzed with 18% SDS-PAGE gel and visualized with coomassie blue staining. FIG. 13 shows that SAMPs are stable in citrus cell lysate, which indicates that they are also stable in trees.

Example 8 – Additional antimicrobial activity of SAMPs

[0167] The antimicrobial activity of SAMPs against other Gram-negative bacterial pathogens was tested. Specifically, activity against *Pseudomonas syringae* and *Agrobacterium tumefaciens* was verified by agar diffusion assay, FIG. 14A and FIG. 14B respectively. Each essay was done by applying 10 µL SAMPs on the medium with bacteria, where the concentration for SAMPs was 100 µM for *Pseudomonas syringae* and 150 µM for *Agrobacterium tumefaciens* respectively. The culture plates were incubated at 28 °C and observed after 24 hours. The rings without bacterial growth confirm antimicrobial activity.

[0168] In addition, SAMPs were incubated at 4 °C for 24 hours, RT for 24 hours, or 60 °C for 24 hours. Subsequently, the SAMPs were used for viability/cytotoxicity assay. The assay was done by incubating 10⁷ cells/mL of *Agrobacterium tumefaciens* with the pre-treated SAMP or buffer only as mock treatment for 2 hours. The samples were then stained with DMAO (green) and EthD-III (orange), which represent live and dead cells, respectively. FIG.

15 shows that SAMPs are stable and have antimicrobial activity against *Agrobacterium tumefaciens* up to 60 °C.

[0169] One or more features from any embodiments described herein or in the figures may be combined with one or more features of any other embodiment described herein in the figures without departing from the scope of the disclosure.

[0170] All publications, patents and patent applications cited in this specification are herein incorporated by reference as if each individual publication or patent application were specifically and individually indicated to be incorporated by reference. Although the foregoing disclosure has been described in some detail by way of illustration and example for purposes of clarity of understanding, it will be readily apparent to those of ordinary skill in the art in light of the teachings of this disclosure that certain changes and modifications may be made thereto without departing from the spirit or scope of the appended claims.

WHAT IS CLAIMED:

1. An isolated stable antimicrobial peptide (SAMP) comprising a sequence that is substantially identical to a sequence of any one of SEQ ID NOs:1-13 and 35-37.
2. The isolated SAMP of claim 1, wherein the peptide comprises a sequence having at least 75% sequence identity to a sequence of any one of SEQ ID NOs:1-13 and 35-37.
3. The isolated SAMP of claim 2, wherein the peptide comprises a sequence having at least 75% sequence identity to a sequence of any one of SEQ ID NOs:1 and 2.
4. An isolated stable antimicrobial peptide (SAMP) comprising a sequence of
X₁GX₂X₃VSX₄ENX₅X₆QGF₇HX₈FEX₉TFX₁₀SX₁₁EGX₁₂AEYX₁₃X₁₄HPX₁₅HVEX₁₆ANX₁₇
X₁₈LX₁₉X₂₀LEKX₂₁LX₂₂X₂₃DYKPX₂₄TX₂₅RV (SEQ ID NO:27),
wherein X₁ is R, K, or W; X₂ is K or E; X₃ is N or D; X₄ is T or I; X₅ is L, F, or R; X₆ is H or Q; X₇ is P or T; X₈ is I, L, or V; X₉ is S or F; X₁₀ is E or D; X₁₁ is T or L; X₁₂ is V or I; X₁₃ is V or I; X₁₄ is S, A, or D; X₁₅ is S, A, or V; X₁₆ is Y or F; X₁₇ is L or T; X₁₈ is F, M, or L; X₁₉ is A, P, or T; X₂₀ is N or Q; X₂₁ is V or F; X₂₂ is V or I; X₂₃ is V or I; X₂₄ is T, E, or Q; and X₂₅ is V, E, L.
5. The isolated SAMP of claim 4, wherein X₁ is R; X₂ is K; X₃ is N; X₄ is I; X₅ is L; X₆ is H; X₁₀ is E; X₁₁ is T; X₁₂ is V; X₁₃ is V; X₁₆ is Y; X₁₇ is L; X₁₈ is F; X₁₉ is A; X₂₀ is N; X₂₁ is V; X₂₂ is V; X₂₃ is I; and X₂₄ is T.
6. The isolated SAMP of any one of claims 1 to 5, wherein the isolated SAMP is a heat stable (HS) peptide.
7. The isolated SAMP of any one of claims 1 to 6, wherein the isolated SAMP is stable in plant extracts and/or in plant lysates.
8. An agricultural composition comprising an isolated SAMP of any one of claims 1 to 7.
9. The agricultural composition of claim 8, further comprising at least one of an herbicide, an herbicide safener, a surfactant, a fungicide, a pesticide, a nematocide, a plant activator, a synergist, a plant growth regulator, an insect repellent, an acaricide, a molluscicide, or a fertilizer.
10. A nucleic acid molecule encoding an isolated SAMP of any one of claims 1 to 7.
11. A cell comprising the nucleic acid molecule of claim 10.

12. The cell of claim 11, wherein the cell is a plant cell.
13. A plant comprising an isolated SAMP of any one of claims 1 to 7 or the nucleic acid molecule of claim 10.
14. The plant of claim 13, wherein the plant is a citrus plant.
15. The plant of claim 13, wherein the plant is a solanaceous plant.
16. A transgenic plant comprising an *in situ* mutated stable antimicrobial peptide (SAMP) comprising at least one amino acid substitution corresponding to an amino acid at any one of positions X₁ to X₂₅ as set forth in SEQ ID NO:27, wherein the mutated SAMP provides disease resistance or disease tolerance to the transgenic plant.
17. The transgenic plant of claim 16, wherein the disease is a *Liberibacter* disease.
18. The transgenic plant of claim 16, wherein the disease is an *Agrobacterium* or a *Pseudomonas syringae* disease.
19. The transgenic plant of claim 16, wherein the *Liberibacter* disease is Huanglongbing (HLB).
20. The transgenic plant of claim 16 or 17, wherein the SAMP is a heat stable (HS) peptide.
21. An expression cassette comprising a promoter operably linked to a polynucleotide encoding an isolated SAMP of any one of claims 1 to 7, wherein introduction of the expression cassette into a plant results in the plant having enhanced disease resistance or disease tolerance.
22. The expression cassette of claim 21, wherein the disease is a *Liberibacter* disease.
23. The expression cassette of claim 21, wherein the disease is an *Agrobacterium* or a *Pseudomonas syringae* disease.
24. The expression cassette of claim 22, wherein the *Liberibacter* disease is HLB.
25. The expression cassette of claim 21, wherein the promoter is heterologous to the polynucleotide.
26. The expression cassette of claim 21 or 25, wherein the promoter is inducible.
27. The expression cassette of claim 21 or 25, wherein the promoter is a tissue-specific promoter.

28. The expression cassette of claim 27, wherein the tissue-specific promoter is a phloem-specific promoter.
29. The expression cassette of claim 28, wherein phloem-specific promoter is the sucrose transporter protein SUC2 promoter.
30. A transgenic plant comprising the expression cassette of any one of claims 21 to 29, wherein the plant has enhanced disease resistance or disease tolerance compared to a control plant lacking the expression cassette.
31. The transgenic plant of claim 30, wherein the disease is a *Liberibacter* disease.
32. The transgenic plant of claim 30, wherein the disease is an *Agrobacterium* or a *Pseudomonas syringae* disease.
33. The transgenic plant of claim 31, wherein the *Liberibacter* disease is HLB.
34. The transgenic plant of claim 16 or 30, wherein the plant is a citrus plant.
35. The transgenic plant of claim 16 or 30, wherein the plant is a solanaceous plant.
36. A method of preventing or treating a disease in a plant, comprising contacting the plant with an isolated SAMP of any one of claims 1 to 7 or an agricultural composition of claim 8 or 9.
37. The method of claim 36, wherein the disease is a *Liberibacter* disease.
38. The method of claim 36, wherein the disease is an *Agrobacterium* or a *Pseudomonas syringae* disease.
39. The method of claim 37, wherein the *Liberibacter* disease is HLB.
40. A method of preventing or treating a bacterial infection in a plant caused by bacteria in the genus *Liberibacter*, *Agrobacterium*, or *Pseudomonas*, comprising contacting the plant with an isolated SAMP of any one of claims 1 to 7 or an agricultural composition of claim 8 or 9.
41. The method of claim 40, wherein the genus *Liberibacter* is *Candidatus Liberibacter*.
42. The method of claim 40, wherein the bacteria in the genus *Liberibacter* are *Liberibacter crescens*.
43. The method of claim 36, wherein the isolated SAMP or agricultural composition is injected into the trunk of the plant.

44. The method of claim 36, wherein the isolated SAMP or agricultural composition is injected into the stem of the plant.
45. The method of claim 36, wherein the isolated SAMP or agricultural composition is foliar sprayed onto the plant or is applied to the plant by dripping irrigation to the roots or is applied to the plant by laser ablation.
46. The method of any one of claims 36 to 45, wherein the bacterial infection causes potato zebra chip disease.
47. A method of preventing or treating a *Liberibacter*, a *Agrobacterium*, or a *Pseudomonas* disease in a plant, comprising introducing an expression cassette of any one of claims 21 to 29 into the plant.
48. The method of claim 47, wherein the *Liberibacter* disease is HLB.
49. A method of preventing or treating a bacterial infection in a plant caused by bacteria in the genus *Liberibacter*, *Agrobacterium*, or *Pseudomonas*, comprising introducing an expression cassette of any one of claims 21 to 29 into the plant.
50. The method of claim 49, wherein the genus *Liberibacter* is *Candidatus Liberibacter*.
51. The method of claim 49, wherein the bacteria in the genus *Liberibacter* are *Liberibacter crescens*.
52. A method of producing a plant having enhanced *Liberibacter*, *Agrobacterium*, or *Pseudomonas* disease resistance or *Liberibacter*, *Agrobacterium*, or *Pseudomonas* disease tolerance, comprising:
- introducing an isolated SAMP of any one of claims 1 to 7 or an expression cassette of any one of claims 21 to 29 into a plurality of plants; and
- selecting a plant that comprises the isolated SAMP or expresses the polynucleotide from the plurality of plants.
53. A method of producing a plant having enhanced *Liberibacter*, *Agrobacterium*, or *Pseudomonas* disease resistance or *Liberibacter*, *Agrobacterium*, or *Pseudomonas* disease tolerance, comprising introducing a mutation into a polynucleotide in the plant, wherein the mutated polynucleotide encodes an isolated SAMP of any one of claims 1 to 7.
54. The method of claim 53, wherein the introducing occurs *in situ* in the genome of a plant cell.

55. The method of claim 53 or 54, wherein the introducing comprises clustered regularly interspaced short palindromic repeats (CRISPR)/Cas genome editing.
56. The method of any one of claims 52 to 55, wherein the plant is resistant or tolerant to a bacterial infection caused by bacteria in the genus *Liberibacter*.
57. The method of claim 56, wherein the genus *Liberibacter* is *Candidatus Liberibacter*.
58. The method of claim 56, wherein the bacteria in the genus *Liberibacter* are *Liberibacter crescens*.
59. The method of any one of claims 52 to 58, wherein the *Liberibacter* disease is HLB.
60. The method of any one of claims 36 to 55, wherein the plant is a citrus plant.
61. The method of any one of claims 36 to 55, wherein the plant is a solanaceous plant.