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One aspect of the present invention relates to isolated nucleic acid molecules (i) encoding proteins or polypeptides of Pseudomonas CEL and EEL genomic regions, (ii) nucleic acid molecules which hybridize thereto under stringent conditions, or (iii) nucleic acid molecules that include a nucleotide sequence which is complementary to the nucleic acid molecules of (i) and (ii). Expression vectors, host cells, and transgenic plants which include the DNA molecules of the present invention are also disclosed. Another aspect relates to isolated proteins or polypeptides and compositions containing the same. The nucleic acid molecules and proteins of the present invention can be used to imparting disease resistance to a plant, making a plant hypersusceptible to colonization by nonpathogenic bacteria, causing eukaryotic cell death, and treating cancerous conditions.

11 Claims, 11 Drawing Sheets
OTHER PUBLICATIONS


* cited by examiner
Figure 1
Figures 2A-C
Figure 3
Figures 4A-E

Figure 5
Figures 6A-B

Figure 7
Figure 10

Figure 11
Signal from kinetochore

Spindle Checkpoint Components
Mad1, Mad3, Bub1-3, Mps1
Mad2

APC INACTIVE

APC ACTIVE

Anaphase

Pds1
Marked for degradation

Figure 12
Figures 13A-B
Figure 14

Figure 15
Figures 16A-B
Hrp genes are probably universal based on such gene-for-gene (avr-R) interactions may produce either of these reactions in plants appears to be a plant cell with a heterologous DNA molecule of the hypersensitive response (HR), a rapid, defense-associated programmed death of plant cells in contact with the pathogen (Alfano and Collmer, 1997). The ability to produce either of these reactions in plants appears to be directed by hrc (HR and pathogenicity) and hrc (HR and conserved) genes that encode a type III protein secretion pathway and by avr (avirulence) and hop (Hrp-dependent outer protein) genes that encode effectors secreted by the bacteria. Hop effectors may also be secreted by the HR-triggering R-gene surveillance system of potential hosts (hence the avr designation), and plant breeding for resistance based on such gene-for-gene (avr-R) interactions may produce complex combinations of races and differential cultivars (Keen, 1990). hrc genes are probably universal among necrosis-causing gram-negative plant pathogens, and they have been sequenced in P. syringae pv. syringae (Psy) 61, Erwinia amylovora Ea321, Xanthomonas campestris pv. vesicatoria (Xcv) 85-10, andRalstonia solanacearum GMI1000 (Alfano and Collmer, 1997). Based on their distinct gene arrangements and regulatory components, the hrc genes of these four bacteria can be divided into two groups: I (Pseudomonas and Erwinia) and II (Xanthomonas and Ralstonia). The discrepancy between the distribution of these groups and the phylogeny of the bacteria provides some evidence that hrc genes have been horizontally acquired and, therefore, may represent pathogenicity islands (Pais) (Alfano and Collmer, 1997).

Pais have been defined as gene clusters that (i) include many virulence genes, (ii) are selectively present in pathogenic strains, (iii) have different G+C content compared to host bacteria DNA, (iv) occupy large chromosomal regions, (v) are often flanked by direct repeats, (vi) are bordered by tRNA genes and/or cryptic mobile genetic elements, and (vii) are unstable (Hacker et al., 1997). Some Pais have been shown to be linked to the hrc cluster in P. syringae: avrR and several other Hrp-regulated transcriptional units are linked to the hrc border of the hrc cluster in P. syringae pv. tomato (Pto) DC3000 (Lorang and Keen, 1995); avrPphE is adjacent to hrcR in Pseudomonas phaseolicola (Pph) 1302A (Mansfield et al., 1994); and hopPsyA (hopA) is adjacent to hrcK in Psy 61 (Heu and Hutcheson, 1993). Other Pseudomonas avr genes are located elsewhere in the genome or on plasmids (Leach and White, 1996), including a plasmid-borne group of avr genes described as a Pais in Pph 1449B (Jackson et al., 1999).

Because Avr, Hop, Hrp, and Hrc proteins represent promising therapeutic treatments in both plants and animals, it would be desirable to identify other proteins encoded by the Pais in pathogenic bacteria and identify uses for these proteins.

The present invention overcomes these deficiencies in the art.

SUMMARY OF THE INVENTION

One aspect of the present invention relates to isolated nucleic acid molecules (i) encoding proteins or polypeptides of Pseudomonas Conserved Effector Loci ("CEL") and Exchangeable Effector Loci ("EEL") genomic regions, (ii) nucleic acid molecules which hybridize thereto under stringent conditions, or (iii) nucleic acid molecules that include a nucleotide sequence which is complementary to the nucleic acid molecules of (i) and (ii). Expression vectors, host cells, and transgenic plants which include the DNA molecules of the present invention are also disclosed. Methods of making such host cells and transgenic plant are disclosed.

A further aspect of the present invention relates to isolated proteins or polypeptides encoded by the nucleic acid molecules of the present invention. Compositions which contain the proteins are also disclosed.

Yet another aspect of the present invention relates to methods of imparting disease resistance to a plant. According to one approach, this method is carried out by transforming a plant cell with a heterologous DNA molecule of the present invention and regenerating a transgenic plant from the transformed plant cell, wherein the transgenic plant expresses the heterologous DNA molecule under conditions effective to impart disease resistance. According to another approach, this method is carried out by treating a plant with a protein or polypeptide of the present invention under conditions effective to impart disease resistance to the treated plant.

A still further aspect of the present invention relates to a method of making a plant hypersusceptible to colonization by nonpathogenic bacteria. According to one approach, this...
method is carried out by transforming a plant cell with a heterologous DNA molecule of the present invention and generating a transgenic plant from the transformed plant cell, wherein the transgenic plant expresses the heterologous DNA molecule under conditions effective to render the transgenic plant hypersusceptible to colonization by non-pathogenic bacteria. According to an alternative approach, this method is carried out by treating a plant with a protein or polypeptide of the present invention under conditions effective to render the treated plant susceptible to colonization by non-pathogenic bacteria.

Another aspect of the present invention relates to a method of causing eukaryotic cell death by introducing into a eukaryotic cell a cytotoxic *Pseudomonas* protein, where the introducing is performed under conditions effective to cause cell death.

A further aspect of the present invention relates to a method of treating a cancerous condition by introducing a cytotoxic *Pseudomonas* protein into cancer cells of a patient under conditions effective to cause death of cancer cells, thereby treating the cancerous condition.

The benefits of the present invention result from three factors. First, there is substantial and growing evidence that phytopathogenic effector proteins have evolved to elicit exquisite changes in eukaryote metabolism at extremely low levels, and at least some of these activities are potentially relevant to mammals and other organisms in addition to plants. For example, ORF5 in the Psy B728a EEL is similar to *Xanthomonas campestris* pv. *vesicatoria* AvrBsL, a phytopathogen protein that appears to have the same active site as its animal pathogen homolog YopI, which inhibits mammalian MAPKK defense signaling (Orth et al., 2000). Second, the *P. syringae* CEL and EEL regions are enriched in effector protein genes, which makes these regions fertile targets for effector gene bioprospecting. Third, rapidly developing technologies for delivering genes and proteins into plant and animal cells improve the efficacy of protein-based therapies.

**BRIEF DESCRIPTION OF THE DRAWINGS**

FIG. 1 is a diagram illustrating the conserved arrangement of hrp/hrp genes within the Hrp Pairs of Psy 61, Psy B728a, and Pto DC3000. Regions sequenced in B728a and DC3000 are indicated by lines beneath the strain 61 sequence. Known regulatory genes are shaded. Arrows indicate the direction of transcription, with small boxes denoting the presence of a Hrp box. The triangle denotes the 3.6-kb insert with phage genes in the B728a hrp/hrp region.

FIGS. 2A–C show the EEL of Pto DC3000, Psy B728a, and Psy 61, the tgt-queA-rRNA~ew~ locus in *P. aeruginosa* (Pa), and EEL border sequences. FIG. 2A is a diagram of the EELs of three *P. syringae* strains shown aligned by their hrpK sequences and are compared with the tgt-queA-rRNA~ew~ locus in *P. aeruginosa* (Pa) PAO1. Arrows indicate the direction of transcription, with small boxes denoting the presence of a Hrp box. Shaded regions are conserved, striped regions denote mobile genetic elements, and open boxes denote genes that are completely dissimilar from each other. FIG. 2B is an alignment of the sequences of the DC3000 (DC) (SEQ. ID. No. 85), B728a (B7) (SEQ. ID. No. 86), and 61 (SEQ. ID. No. 87) EELs at the border with rRNA~ew~, with conserved nucleotides shown in upper case. FIG. 2C is an alignment of the sequences of the DC3000 (DC) (SEQ. ID. No. 85), B728a (B7) (SEQ. ID. No. 86), and 61 (SEQ. ID. No. 90) EELs at the border with hrpK, with conserved nucleotides shown in upper case.

FIG. 3 is a diagram illustrating the Hrp Pai CEL of *P. syringae*. The Pto DC3000 CEL is shown with the corresponding fragments of Psy B728a that were sequenced aligned below. The nucleotide identity of the sequenced fragments in coding regions ranged from 72% to 83%. Arrows indicate the direction of transcription, with small boxes denoting the presence of a Hrp box.

FIGS. 4A–E illustrate the plant interaction phenotypes of Pto mutants carrying deletions of the EEL (CUCPB5115) and CEL (CUCPB5115). FIG. 14A is a graph illustrating growth in tomato of DC3000 and CUCPB5115 (mean and SD). FIG. 14B is a graph illustrating growth in tomato of DC3000, CUCPB5115, and CUCPB5115(pCPP3016) (mean and SD). FIG. 14C is an image showing HR collapse in tobacco leaf tissue 24 h after infiltration with 10⁷ cfu/ml of DC3000 and CUCPB5115. FIG. 14D is an image showing the absence of disease symptoms in tomato leaf 4 days after inoculation with 10⁴ cfu/ml of CUCPB5115. FIG. 14E is an image showing disease symptoms typical of wild-type in tomato leaf 4 days after inoculation with 10⁴ cfu/ml of CUCPB5115(pCPP3016).

FIG. 5 is an image of the immunoblot analysis showing AvrPto secretion by Pto DC3000 derivatives with deletions affecting the three major regions of the Hrp Pai. Bacteria were grown in Hrp-inducing minimal medium at pH 5.5 and 22°C. An OD₆₅₀ of 0.35 and then separated into cell-bound (C) and supernatant (S) fractions by centrifugation. Proteins were then resolved by SDS-PAGE, blotted, and immunostained with antibodies against AvrPto and β-lactamase as described (Manceau and Harvais, 1997), except that supernatant fractions were concentrated 3-fold relative to cell-bound fractions before loading. Pto DC3000, CUCPB5115 (CEL deletion), CUCPB5114 (hrp/hrc deletion), and CUCPB5110 (EEL deletion) all carried pCPP2318, which expresses β-lactamase without a signal peptide as a cytoplasmic marker.

FIGS. 6A–B illustrate, enlarged as compared to FIG. 1, the organization of the shcA and hopPsyA 'aeroperon in the EEL of the Hrp Pai of Psy 61. In FIG. 6A, the shcA and hopPsyA are depicted as white boxes. At the border of the Hrp Pai are the rRNA~ew~ and queA genes depicted as gray boxes. A 5' truncated hrpK gene is represented as a hatched box. The arrows indicate the predicted direction of transcription and the black box denotes the presence of a putative HrpL-independent promoter upstream of shcA. FIG. 6B illustrates schematically the construction of the deletion mutation in the shcA ORF marker-exchanged into Psy 61. Black bars depict regions that were amplified along with added restriction enzyme sites and each are aligned with the corresponding DNA region represented in FIG. 6A. The striped box depicts the nptII cassette that lacks transcriptional and translational terminators used in making the functionally non-polar shcA Psy 61 mutant. EcoRI; E; EcoRV; V; XbaI; X; and XhoI, Xh.

FIG. 7 is an image of an immunoblot showing that shcA encodes a protein product. pLV9 is a derivative of pFLAG-CTC in which the shcA ORF is cloned and fused to the FLAG epitope and translation is directed by a vector ribosome binding site (RBS). pLV26 contains an amplified product containing the shcA coding region and its native RBS site. Cultures of *E. coli* DH5α carrying either pFLAG-CTC (Control), pLV9, or pLV26 were grown to an OD₆₅₀ of 0.8 and then 100 μl aliquots were taken, centrifuged, resuspended in SDS-PAGE buffer, and then subjected to SDS-PAGE and immunoblot analysis with anti-FLAG antibodies and secondary antibodies conjugated with alkaline phosphatase.
FIG. 8 is an image of an immunoblot showing that Psy 61 shcA mutant UNLV102 does not secrete HopPsyA and shcA provided in trans complements this defect. Psy 61 cultures were grown at 22°C in hpr-depressing medium and separated into cell-bound (C) and supernatant fractions (S). The cell-bound fractions were concentrated 13.4-fold and the supernatant fractions were concentrated 100-fold relative to the initial culture volumes. The samples were subjected to SDS-PAGE and immunoblot analysis, and HopPsyA and β-lactamase (Bla) were detected with either anti-HopPsyA or anti-β-lactamase antibodies followed by secondary antibodies conjugated to alkaline phosphatase as described in the experimental procedures. The image of the immunoblot was captured using the Bio-Rad Gel Doc 2000 UV fluorescent gel documentation system with the accompanying Quantity 1 software.

FIG. 9 is an image of an immunoblot showing that shcA is required for the type III secretion of HopPsyA, but not secretion of HrpZ. P. fluorescens 55 cultures were grown in hpr-depressing medium and separated into cell-bound (C) and supernatant (S) fractions. The cell-bound fractions were concentrated 13.4-fold and the supernatant fractions were concentrated 100-fold relative to the initial culture volumes. The samples were subjected to SDS-PAGE and immunoblot analysis, and HopPsyA and HrpZ were detected with either anti-HopPsyA or anti-HrpZ antibodies followed by secondary antibodies conjugated to alkaline phosphatase as described in experimental procedures. The image of the immunoblot was captured using the Bio-Rad Gel Doc 2000 UV fluorescent gel documentation system with the accompanying Quantity 1 software.

FIG. 10 is a series of four images of tobacco leaves showing that P. fluorescens 55 carrying a pHR111 derivative with a functionally nonpolar shcA mutation is impaired in its ability to translocate HopPsyA into plant cells. P. fluorescens 55 cultures were grown overnight in King’s B and suspended in 5 mM MES pH 5.6 to an OD 600 of 1.0, and infiltrated into tobacco leaf panels. Because the pHR111-induced HR is due to the translocation of HopPsyA inside plant cells, a reduced HR indicates that HopPsyA is not delivered well enough to induce a typical HR. The leaf panels were photographed with incident light 24 hours later.

FIG. 11 is an image of an immunoblot showing that ShcA binds to HopPsyA. Soluble protein samples from sonicated cultures (Sonicate) of Psy 61 shcA mutant UNLV102 carrying pLN1 (HopPsyA) or pLN2 (ShcA-FLAG, HopPsyA) were mixed with anti-FLAG M2 affinity gel (Gel). The gel was washed (Wash) with TBS buffer, mixed with SDS-PAGE buffer, and subjected to SDS-PAGE and immunoblot analysis along with the sonicate and wash samples. HopPsyA and ShcA-FLAG were detected with anti-HopPsyA or anti-FLAG antibodies followed by secondary antibodies conjugated to alkaline phosphatase as described in experimental procedures.

FIG. 12 is a diagram illustrating the spindle checkpoint in S. cerevisiae. The spindle checkpoint is activated by a signal emitted from the kinetochores when there are abnormalities with the microtubules. This signal is somehow received by the spindle checkpoint components, which respond in a variety of ways. Mad2 is thought to bind to Cdc20 at the APC inhibiting its ubiquitin ligase activity. In the absence of Mad2 (and presumably damage to the spindle), the APC is active and it marks Pds1 and other inhibitors of anaphase for degradation via the ubiquitin proteolysis pathway; anaphase ensues.

FIGS. 13A–B illustrate the effects of transgenically expressed HopPsyA on Nicotiana benthamiana, and Arabidopsis thaliana. FIG. 13A shows N. tabacum cv. Xanthi and N. benthamiana leaves infiltrated with Agrobacterium tumefaciens GV3101 with or without pTA7002::hopPsyA. FIG. 13B illustrates Arabidopsis thaliana Col-1 infiltrated with A. tumefaciens +/- pTA7002::hopPsyA. For all plants shown in FIGS. 13A–B, 48 h after Agrobacterium infiltration, plants were sprayed with the glucocorticoid dexamethasone (DEX). Images were collected 24 h after DEX treatment. A.t. = Agrobacterium tumefaciens; pA=pTA7002::hopPsyA.

FIG. 14 is an image of an SDS-PAGE which shows the distribution of HopPsyA and β-lactamase in cultures of Psy 61 (pCPP2318) or a hrp mutant, Psy 61-4 2089 (pCPP2318). Bacterial cultures were grown at 22°C in hpr-depressing medium and separated into cell-bound (C) and supernatant fractions (S). The cell-bound fractions were concentrated 13.4 fold, and the supernatant fractions were concentrated 100 fold relative to initial culture volumes. The samples were subjected to SDS-PAGE and immunoblot analysis and HopPsyA and β-lactamase were detected with either anti-HopPsyA or anti-β-lactamase antibodies followed by secondary antibodies conjugated to alkaline phosphatase. Pss wild-type = Pseudomonas syringae pv. syringae 61 (pCPP2318); Pss hrpC = Pseudomonas syringae pv. syringae 61-2089 (pCPP2318).

FIG. 15 is a graph illustrating the ability of wild-type Pseudomonas syringae pv. syringae and a hopPsyA mutant to multiply in bean leaves. Values represent the average plate counts from crushed plant leaves of two independent inoculations. Wild-type (●), Pseudomonas syringae pv. syringae 61; hopPsyA mutant (○), Pseudomonas syringae pv. syringae 61-2070.

FIGS. 16A–B illustrate the interaction of HopPsyA and Mad2 in a yeast two-hybrid assay. FIG. 16A illustrates cultures of yeast EGY48 strains containing either pLV24 (pEG202::hopPsyA) and pJG4-5 (fish-vector), pLV24 and pLV116 (pJG4-5::mad2), or pEG202 (bait vector) and pLV116 on medium containing 5-bromo-4-chloro-3-indolyl-β-D-galactopyranoside (Xgal) to check for β-galactosidase activity with either glucose (Glc) or galactose (Gal). β-galactosidase activity was indicated only in the presence of both HopPsyA and Mad2. FIG. 16B illustrates cultures of the same yeast strains on minimal medium leucine dropout plates with either Glc or Gal sugars. 1=EGY48 (pLV24, pJG4-5); 2=EGY48 (pLV24, pLV116); 3=EGY48 (pEG202, pLV116).

A DNA molecule which contains the CEL of Pseudomonas syringae pv. tomato DC3000 has a nucleotide sequence (SEQ. ID. No. 1) as follows:
ggtaacgagggc tctgtaaagc agaagcgcgt ccaacagcaac gtaggggaag
ataatcttct gtagaaacta cagctaacttc tgaagcctgct
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tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 20700
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 20760
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

---

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 20820
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 20880
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 20940
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 21000
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 21060
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 21120
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 21180
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 21240
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 21300
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 21360
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 21420
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 21480
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 21540
tggattgtgc gggcgttttg aacagcggt tggatctttgc aatctgcttg

tggagagcc cacagccgcta atacccgttg tttggtggct ttgggtggct 21600
-continued

tctacggaag ttgcaacgca gcagaaggctg acgggctcgta ctccgaagoga ctaaattgctgct 24060
gaaaccaata cagttgcgcgt tcaataacac acggcagcag agggcgaag ctgccgccgt 24120
cgcgcgcttt aacagaggtcg acgggctcgta ctccgaagoga ctaaattgctgct 24180
gaaaccaata cagttgcgcgt tcaataacac acggcagcag agggcgaag ctgccgccgt 24240
gaaaccaata cagttgcgcgt tcaataacac acggcagcag agggcgaag ctgccgccgt 24300
gaaaccaata cagttgcgcgt tcaataacac acggcagcag agggcgaag ctgccgccgt 24360
tgcttccttc gcagaggtcg acgggctcgta ctccgaagoga ctaaattgctgct 24420
gaaaccaata cagttgcgcgt tcaataacac acggcagcag agggcgaag ctgccgccgt 24480
caaagcctgctgcccttc gctggccttgc gctgggcctg cgtgccttgcg ctgctggcctg 24540
caaagcctgctgcccttc gctggccttgc gctgggcctg cgtgccttgcg ctgctggcctg 24600
caaagcctgctgcccttc gctggccttgc gctgggcctg cgtgccttgcg ctgctggcctg 24660
caaagcctgctgcccttc gctggccttgc gctgggcctg cgtgccttgcg ctgctggcctg 24720
caaagcctgctgcccttc gctggccttgc gctgggcctg cgtgccttgcg ctgctggcctg 24780
caaagcctgctgcccttc gctggccttgc gctgggcctg cgtgccttgcg ctgctggcctg 24840
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caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 24960
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25020
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25080
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25140
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25200
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25260
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25320
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25380
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25440
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25500
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25560
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25620
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25680
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25740
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25800
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25860
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25920
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 25980
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 26040
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 26100
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 26160
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 26220
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 26280
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 26340
caaagcctgctgcccttc gctgggcttgac ccggcgtcgt ccggcgtcgt ccggcgtcgt 26400
ctgtcaggtc atgaacggtc atggggtcag atggacagcc ggtaagaacc gaggctcttt
ctgggctgtt ttcgcctgt gcgtcattcg ctgataattc tccagatcgc gctgcaacga
catagtggc agoaacgcga aacaagcttc cttgaataca ggggttatac caagagacgc
gcatcgtggc cagagctgcc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
cgcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
catgtcgtgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
cgcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
catgtcgtgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
cgcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
catgtcgtgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
cgcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
catgtcgtgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
cgcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
catgtcgtgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
cgcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
catgtcgtgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
cgcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
catgtcgtgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
cgcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
catgtcgtgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
cgcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
catgtcgtgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
cgcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
catgtcgtgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
cgcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
catgtcgtgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
cgcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
catgtcgtgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc gcgtcactgc
Several undefined nucleotides exist in SEQ. ID. No. 1, however these appear to be present in intergenic regions. The CEL of *Pseudomonas syringae* pv. *tomato* DC3000 contains a number of open reading frames (ORFs). Two of the products encoded by the CEL are HrpW and AvrE, both of which are known. An additional 10 products are produced by ORF1-10, respectively, as shown in FIG. 3. The nucleotide sequences for a number of these ORFs and their encoded protein or polypeptide products are provided below.

The DNA molecule of ORF3 from the *Pseudomonas syringae* pv. *tomato* DC3000 CEL has a nucleotide sequence (SEQ. ID. No. 2) as follows:

```
acgatcagtt cgcggatcgg cggggccggt ggcgtcaaac tcagccgggt aaaccagcag
cacgatactg ttcccgccca gacagctcac ccaaatgcag tcactgcagg catgaatccg
ccgctgactc ccgatcagtc agggtcacac gcgacagaaa gctcgtctgc cggcgcggcg
cggctgaatg tcgcggctcg acacacacag cttttgcagg ccttcaaggc tgagcatggg
acggctccgg tcagcggcgc gccgatgatc agttcgcgtg ctgcgttgtt gatcggtagt
cctgctgcagg ccgagccttt gccttttgaa gtcatggccg agaaattgtc tcctgagcgc
tatcaactga agcagtttca gggctcggac ttgcagcagc ggctggaaaa attcgcccag
```
The protein or polypeptide encoded by Pto DC3000 CEL ORF3 has an amino acid sequence (SEQ. ID. No. 3) as follows:

```
1  Met Ile Ser Ser Arg Ile Gly Gly Ala Gly Gly Val Lys Leu Ser Arg  
  5  10  15
20  Val Asn Gln Gln His Asp Thr Val Pro Ala Gln Thr Ala His Pro Asn
25  
30  Ala Val Thr Ala Gly Met Asn Pro Pro Leu Thr Pro Asp Gln Glu Asp
35  40  45
50  Ser His Ala Thr Glu Ser Ser Ala Gly Ala Ala Arg Ala Arg Leu Val
55  60
65  Ala Ala Arg His Thr Gln Leu Leu Gln Ala Phe Lys Ala Glu His Gly
70  75  80
85  Thr Ala Pro Val Ser Gly Ala Pro Met Ile Ser Ser Arg Ala Ala
90  95
100 Leu Ile Gly Ser Leu Leu Gln Ala Glu Pro Leu Pro Phe Glu Val Met
105  110
115 Ala Glu Lys Leu Ser Pro Glu Arg Tyr Gln Leu Lys Gln Phe Glu Gly
120  125
```
-continued

Ser Asp Leu Gln Glu Arg Leu Glu Lys Phe Ala Gln Pro Gly Gln Ile
130 135 140

Pro Asp Lys Ala Glu Val Gly Gln Leu Ile Lys Gly Phe Ala Gln Ser
145 150 155 160

Val Ala Asp Glu Leu His Phe Glu Leu Met His Asp Ala Ser Pro
165 170 175

 Ala Thr Val Gly Gln His Ala Lys Ala Asp Lys Ala Thr Leu Ala Val
180 185 190

Ser Gln Thr Ala Leu Gly Glu Tyr Ala Gly Arg Ala Ser Lys Ala Ile
195 200 205

Gly Glu Gly Leu Ser Asn Ser Ile Ala Ser Leu Asp Glu His Ile Ser
210 215 220

 Ala Leu Asp Leu Thr Leu Glu Asp Ala Glu Gly Asn Lys Glu Ser
225 230 235 240

 Leu His Ala Asp Arg Glu Ala Leu Val Asp Ala Lys Thr Thr Leu Val
245 250 255

Gly Leu His Ala Asp Phe Val Lys Ser Pro Gln Ala Lys Arg Leu Ala
260 265 270

Ser Val Ala Ala His Thr Leu Asp Met Val Ala Val Ser Asp Leu Val
275 280 285

Thr Ala Arg Asn Thr Val Gly Gly Trp Lys Gly Ala Gly Pro Ile Val
290 295 300

 Ala Ala Val Pro Gln Phe Leu Ser Ser Met Thr His Leu Gly Tyr
305 310 315 320

Val Arg Leu Ser Thr Ser Asp Lys Leu Arg Asp Thr Ile Pro Glu Thr
325 330 335

Ser Ser Asp Ala Asn Met Leu Lys Ala Ser Ile Ile Gly Met Val Ala
340 345 350

Gly Ile Ala His Glu Thr Val Asn Ser Val Val Lys Pro Met Phe Gln
355 360 365

 Ala Ala Leu Gln Lys Thr Gly Leu Asn Glu Arg Leu Asn Met Val Pro
370 375 380

Met Lys Ala Val Asp Thr Asn Thr Val Ile Pro Asp Pro Phe Glu Leu
385 390 395 400

Lys Ser Glu His Gly Glu Leu Val Lys Thr Pro Glu Glu Val Ala
405 410 415

Gln Asp Lys Ala Phe Val Lys Ser Glu Arg Ala Leu Asn Gln Lys
420 425 430

Lys Val Gln Gly Ser Ser Thr His Pro Val Gly Glu Leu Met Ala Tyr
435 440 445

Ser Ala Phe Gly Gly Ser Glu Ala Val Arg Glu Asp Leu Asn Met Val Pro
450 455 460

His Gly Ile Asn Gly Gln Thr Leu Ser Ala Arg Ala Leu Ala Ser Gly
465 470 475 480

Phe Gly Ala Val Ser Ala Ser Ser Glu Thr Leu Leu Glu Ile Lys
485 490 495

Ser Asn Tyr Val Asp Pro Gln Gly Arg Lys Ile Pro Val Phe Thr Pro
500 505 510

Asp Arg Ala Glu Ser Asp Leu Lys Lys Leu Leu Gly Met Asp
515 520 525

Leu Arg Glu Pro Ser Val Arg Thr Thr Phe Tyr Ser Lys Ala Leu Ser
530 535 540

Gly Ile Gln Ser Ser Ala Leu Thr Ser Ala Leu Pro Pro Val Thr Ala
The DNA molecule of ORF4 from the *Pseudomonas syringae* pv. *tomato* DC3000 CEL has a nucleotide sequence (SEQ. ID. No. 4) as follows:

```
atgaccaca atgaccagta ccacaccctt atcaacgaaa tctgcgcact cagcctgatt
tccacacctg ascgcttctg tgcacgcgct cttacccac tgcgaccgcc ctcgccgccg
tgccccgccc ctgtgcgacc gcgagcgcgcc ggcgcacctt ccagcgtgtg
tgctgccgct cgcgggcgcc ggcgacggtg tgcggtggcc gccgcccttt gcgtgcgttg
tttgcgctgc gcggccgccg gcgcgtggtg cgggtggtgtg tcgccgctgg
tgggccgggg gcgcgcgggg ggcgcggttc gcgtgcgttc gcggccgcgg
tgtgcgcggt gcgcgcgttc gcggtgcggt gcgcgcgttc gcggccgcgg
tgtgcgcggt gcgcgcgttc gcggtgcggt gcgcgcgttc gcggccgcgg
tgggccgggg gcgcgcgggg ggcgcggttc gcgtgcgttc gcggccgcgg
tgtgcgcggt gcgcgcgttc gcggtgcggt gcgcgcgttc gcggccgcgg
tgtgcgcggt gcgcgcgttc gcggtgcggt gcgcgcgttc gcggccgcgg
```

The protein or polypeptide encoded by Pto DC3000 CEL ORF4 has an amino acid sequence (SEQ. ID. No. 5) as follows:

```
Met Thr Asn Asn Asp Gln Tyr His Thr Leu Ile Asn Glu Ile Cys Ala
Leu Ser Leu Ile Ser Thr Pro Glu Arg Phe Tyr Glu Ser Ala Asn Phe
Lys Ile Ser Glu Val Asp Phe Thr Leu Gin Phe Gin Asp Arg Asp Glu
Gly Arg Ala Val Leu Ile Tyr Gly Asp Met Gly Ala Leu Pro Ala Arg
Gly Arg Glu Ser Ala Leu Leu Leu Met Asp Ile Asn Phe His Met
Phe Ala Gly Ala His Ser Pro Ala Phe Ser Phe Asn Ala Gin Thr Gly
Arg Val Leu Leu Met Gly Ser Val Ala Leu Glu Arg Ala Ser Ala Glu
Gly Val Leu Leu Leu Met Lys Ser Phe Ser Asp Leu Ala Lys Glu Trp
Arg Glu His Gly Phe Met Gly Gin Ala Thr Thr Ala Gly Ser Ser Thr
Asp Gin Pro Val Ala Pro Ala Lys Arg Glu Ser Leu Ser Ala Pro
Gly Arg Phe Gin
```
The DNA molecule of ORF5 from the *Pseudomonas syringae* pv. *tomato* DC3000 CEL has a nucleotide sequence (SEQ. ID. No. 6) as follows:

```
atgcacatca accacagctc ccaacacaccct ctgctgactcc ttgggacata 60
ggcggacggc tagctgctcctc cgtctcttgcct cccagctctgt gcgcagcttgctc 120
ttagatgatag cggtagccat ccgtctgatggt cggcagttgtgc cgtcactgctc 180
tccggtggag cgaacctttgct gccacccgct cggcaacggcg gagataaatgctgctc 240
cggcgtcgcac gccgactgtgctgc gccgccctggcc cggagctgatt gcgggtagtgctc 300
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 360
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 420
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 480
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 540
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 600
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 660
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 720
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 780
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 840
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 900
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 960
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 1020
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 1080
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 1140
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 1200
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 1260
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 1320
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 1380
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 1440
tccgggtgggt gcggcggcagc cgcctccggc gggccgacggc ggggcggccc 1500
```

The protein or polypeptide encoded by Pto DC3000 CEL ORF5, now known as HopPtoA, has an amino acid sequence (SEQ. ID. No. 7) as follows:

```
Met His Ile Asn Arg Arg Val Gln Gln Pro Pro Val Thr Ala Thr Asp 1 5 10 15
Ser Phe Arg Thr Ala Ser Asp Ala Ser Leu Ala Ser Ser Ser Val Arg 20 25 30
Ser Val Ser Ser Asp Gln Gln Arg Glu Ile Asp Ala Ile Ala Asp Tyr 35 40 45
Leu Thr Asp His Val Phe Ala Ala His Lys Leu Pro Pro Ala Asp Ser 50 55 60
Ala Asp Gly Gln Ala Ala Val Asp Val His Asn Ala Gln Ile Thr Ala 65 70 75 80
Leu Ile Glu Thr Arg Ala Ser Arg Leu His Phe Glu Gly Glu Thr Pro 85 90 95
Ala Thr Ile Ala Asp Thr Phe Ala Lys Ala Glu Lys Leu Asp Arg Leu
```
Ala Thr Thr Thr Ser Gly Ala Leu Arg Ala Thr Pro Phe Ala Met Ala
115
Ser Leu Leu Gln Tyr Met Gln Pro Ala Ile Asn Lys Gly Asp Trp Leu
130
Pro Ala Pro Leu Lys Pro Leu Thr Pro Leu Ile Ser Gly Ala Leu Ser
145
Gly Ala Met Asp Gln Val Gly Thr Lys Met Met Asp Arg Ala Thr Gly
165
Asp Leu His Tyr Leu Ser Ala Ser Pro Asp Arg Leu His Asp Ala Met
180
Ala Ala Ser Val Lys Arg His Ser Pro Ser Leu Arg Glu Val Leu
195
Asp Thr Gly Val Ala Val Gln Thr Tyr Ser Ala Arg Asn Ala Val Arg
210
Thr Val Leu Ala Pro Ala Leu Ala Ser Arg Pro Ala Val Gin Gly Ala
225
Val Asp Leu Gly Val Ser Met Ala Gly Gly Leu Ala Ala Asn Ala Gly
245
Phe Gly Asn Arg Leu Leu Ser Val Gin Ser Arg Arg His Gin Arg Gly
260
Gly Ala Leu Val Leu Gly Leu Lys Asp Lys Glu Pro Lys Ala Gin Leu
275
Ser Glu Glu Asn Asp Trp Leu Glu Ala Tyr Lys Ala Ile Lys Ser Ala
290
Ser Tyr Ser Gly Ala Ala Leu Asn Ala Gly Lys Arg Met Ala Gly Leu
305
Pro Leu Asp Met Ala Thr Asp Ala Met Gly Ala Val Arg Ser Leu Val
325
Ser Ala Ser Ser Leu Thr Gin Gly Leu Ala Ala Gly Gly Phe
340
Ala Gly Val Gly Lys Leu Gin Met Ala Thr Lys Asn Ile Thr Asp
355
Pro Ala Thr Lys Ala Ala Val Ser Gin Leu Thr Asn Ala Gly Ser
370
Ala Ala Val Phe Ala Gly Trp Thr Thr Ala Ala Leu Thr Thr Asp Pro
385
Ala Val Lys Ala Glu Ser Phe Ile Gin Asp Thr Val Lys Ser Thr
405
Ala Ser Ser Thr Thr Gly Tyr Val Ala Asp Gin Thr Val Lys Leu Ala
420
Lys Thr Val Lys Asp Met Gly Gly Glu Ala Ile Thr His Thr Gly Ala
435
Ser Leu Arg Asn Thr Val Asn Asn Leu Arg Gin Gin Pro Ala Arg Glu
450
Ala Asp Ile Glu Glu Gly Gly Thr Ala Ala Ser Pro Ser Glu Ile Pro
465
Phe Arg Pro Met Arg Ser
485
The DNA molecule of ORF6 from the *Pseudomonas syringae* pv. *tomato* DC3000 CEL has a nucleotide sequence (SEQ. ID. No. 8) as follows:

```
atgtctggtc ctttcgagaa aaaatggcgg tgtttcaccc gaaccgtgac ctacgttggc
tggtcgctgt tctggcttct gctctgggac gtggccgtca ccgtggacgt catgctgata
gaaggcaaag gcatcgactt ccccctgatg cccctcacgt tgctttgctc ggcactgatc
gtgctgatca gctttcgcaa ctcgagtgcc tataaccgtt ggtgggaagc gcgcaccttg
```

The protein or polypeptide encoded by Pto DC3000 CEL ORF6 has an amino acid sequence (SEQ. ID. No. 9) as follows:

```
Met Ser Gly Pro Phe Glu Lys Lys Trp Arg Thr Arg Thr Val 1 5 10 15
Thr Tyr Val Gly Trp Ser Leu Phe Trp Leu Leu Trp Asp Val Ala 20 25 30
Val Thr Val Asp Val Met Leu Ile Gly Lys Ile Asp Phe Pro 35 40 45
Leu Met Pro Leu Thr Leu Cys Ser Ala Leu Ile Val Leu Ile Ser 50 55 60
Phe Arg Ser Ser Ala Tyr Asn Arg Trp Trp Glu Ala Arg Thr Leu 65 70 75 80
Trp Gly Ala Met Val Asn Thr Ser Arg Ser Phe Gly Arg Gln Val Leu 85 90 95
Thr Leu Ile Asp Gly Arg Asp Leu Asn Asn Pro Val Lys Ala 100 105 110
Ile Leu Phe Glu Arg His Val Ala Tyr Leu Arg Ala Leu Arg Ala His 115 120 125
Leu Lys Gly Asp Val Lys Thr Ala Lys Leu Asp Gly Leu Leu Ser Pro 130 135 140
Asp Glu Ile Gln Arg Ala Ser Gln Ser Asn Asn Phe Pro Asp Asp Ile 145 150 155 160
Leu Asn Gly Ser Ala Val Ser Gln Ala Phe Ala Ala Gly Gln 165 170 175
```
The DNA molecule of ORF7 from the *Pseudomonas syringae pv. tomato* DC3000 CEL has a nucleotide sequence (SEQ. ID. No. 10) as follows:

```
agctatatcc agcaatctgg cgcccaatca ggggttgccg ctaagacgca acacgataag
```

The protein or polypeptide encoded by Pro DC3000 CEL ORF7 has an amino acid sequence (SEQ. ID. No. 11) as follows:

```
Phe Asp Ser Ile Arg Leu Thr Arg Leu Glu Ser Thr Met Val Asp Leu Ser Asn Cys Gln Gly Gly Met Glu Arg Ile Ala Asn Thr Pro Leu Pro Tyr Pro Tyr Val Tyr Phe Pro Arg Leu Phe Ser Thr Leu Phe Cys Ile Leu Met Pro Leu Ser Met Val Thr Thr Leu Gly Trp Phe Thr Pro Ala Ile Ser Thr Val Val Gly Cys Met Leu Leu Ala Met Asp Arg Ile Gly Thr Asp Leu Glu Asn Cys Gln Thr Leu Leu Met Phe Ser Glu Asp Leu Cys Asn Thr Ile Glu Lys Asn Asp Leu Leu Asp Leu Ala Asp Leu Pro Leu Ala Asp Leu Gly Leu Ala Ser Met Phe Ser Glu Pro Glu Arg Gln Pro Leu Ala Asp Leu Lys Ser Pro Val Pro Trp Arg Val Ala Asn Ala Ser Ile Gly Leu Ser Arg Gln Lys Asn Arg Leu Gly Gln Ala Arg Leu Ile Ala Ser Glu Leu Leu Ala Pro Phe Arg Ser Val Ala Asp Val Ala Pro Cys His Ala Ser Ala Ile Ser Thr Val Val Gly Cys Met Leu Leu Ala Met Asp Arg Ile Gly Thr Asp Leu Glu Asn Cys Gln
```
Met Tyr Ile Gln Gln Ser Gly Ala Gln Ser Gly Val Ala Ala Lys Thr
1      5
Gln His Asp Lys Pro Ser Ser Gly Leu Ala Pro Gly Ser Ser
20     25
Asp Ala Phe Ala Arg Phe His Pro Glu Lys Ala Gly Ala Phe Val Pro
35     40
Leu Glu Gly His Glu Val Phe Phe Asp Ala Arg Ser Ser Phe Ser
50     55
Ser Val Asp Ala Ala Asp Leu Pro Ser Pro Glu Gln Val Gln Pro Gln
65     70
Leu His Ser Leu Arg Thr Leu Leu Pro Asp Leu Met Val Ser Ile Ala
85     90
Ser Leu Arg Asp Gly Ala Thr Gln Tyr Ile Lys Thr Arg Ile Lys Ala
100    105
Met Ala Asp Asn Ser Ile Gly Ala Thr Ala Asn Ile Glu Ala Lys Arg
115    120
Lys Ile Ala Gln Glu His Gly Cys Glu Val Leu Val Pro Phe His Gln
130    135
Ser Lys Phe Leu Phe Glu Lys Thr Ile Asp Arg Ala Phe Ala Ala
145    150
155
Asp Tyr Gly Arg Ala Gly Asp Gly His Ala Cys Leu Gly Leu Ser
165 170 175
Val Asn Trp Cys Gln Ser Arg Ala Lys Gly Gln Ser Asp Gln Ala Phe
180 185 190
Phe His Lys Leu Glu Asp Tyr Gln Gly Asp Ala Leu Leu Pro Arg Val
195 200 205
Met Gly Phe Gly His Ile Glu Gln Ala Tyr Ser Asn Lys Leu Gln
210 215 220
Asn Ala Ala Pro Met Leu Leu Asp Leu Pro Leu Leu Gly Met Thr
225 230 235 240
Leu Gly Lys Gly Leu Gly Arg Ala Gln His Ala His Tyr Ala Val Ala
245 250 255
Leu Glu Asn Leu Asp Arg Ala Lys Ala Val Leu Gln Pro Gly Lys
260 265 270
Asp Gln Met Leu Leu Phe Leu Ser Asp Ser His Ala Met Ala Leu His
275 280 285
Gln Asp Ser Gln Gly Cys Leu His Phe Asp Pro Leu Phe Gly Val
290 295 300
Val Gln Ala Asp Ser Phe Ser Asn Met Ser His Phe Leu Ala Asp Val
305 310 315 320
Phe Lys Arg Asp Val Gly Thr His Trp Arg Gly Thr Glu Gln Arg Leu
325 330 335
Gln Leu Ser Gln Met Val Pro Arg Ala Asp Phe His Leu Arg
340 345 350 355

The DNA molecule of ORF8 from the *Pseudomonas syringae* pv. *tomato* DC3000 CEL has a nucleotide sequence (SEQ. ID. No. 12) as follows:

atgcggcctg tcgaggcaaa agatcggctt tatcagtggc tgcgcaatcg aggcatcgat
60
ggcgagggt gtcacagcga cacaagttsa aggccgaatg tctggctcgg
120
ttgccagac agggacttc gttgctcctc ttcacacagc tccaagsgct gcagatgccg
180
The protein or polypeptide encoded by Pto DC3000 CEL ORF8 has an amino acid sequence (SEQ. ID. No. 13) as follows:

```
  Met Arg Pro Val Glu Ala Lys Asp Arg Leu Tyr Gln Trp Leu Arg Asn 
  1  5 10 15
 Arg Gly Ile Asp Ala Gln Glu Gly Glu Arg His Asn Val Arg Thr Ala 
  20 25 30
 Asn Gly Ser Glu Cys Leu Leu Trp Leu Pro Glu Glu Asp Thr Ser Leu 
  35 40 45
 Phe Ile Phe Thr Gln Ile Glu Arg Leu Thr Met Pro Gln Asp Aan Val 
  50 55 60
 Ile Leu Ile Leu Ala Met Ala Leu Asn Leu Glu Pro Ala Arg Thr Gly 
  65 70 75 80
 Gly Ala Ala Leu Gly Tyr Asn Pro Asp Arg Glu Leu Leu Leu Arg 
  85 90 95
 Ser Val His Ser Met Ala Asp Leu Asp Gly Thr Gly Leu Asp His Leu 
 100 105 110
 Met Thr Arg Ile Ser Thr Leu Ala Val Ser Leu Gln Arg Tyr Leu Glu 
115 120 125
 Asp Tyr Arg Arg Gln Glu Ala Gly Lys Thr Ala Gln Lys Glu Pro 
130 135 140
 Arg Phe Leu Pro Ala Val His Leu Thr Pro Arg Thr Phe Met Thr 
145 150 155
```

The DNA molecule of ORF9 from the *Pseudomonas syringae* pv. *tomato* DC3000 CEL has a nucleotide sequence (SEQ. ID. No. 14) as follows:

```
atgcttaaaa aatgcctgct actggttata tcaatgtcac ttggcggctg ctggagcctg 60
atgattcatc tggacggcga gcgttgcatc tatcccggca ctcgccaagg ttgggcgtgg 120
```

The protein or polypeptide encoded by Pto DC3000 CEL ORF9 has an amino acid sequence (SEQ. ID. No. 15) as follows:

```
  Met Leu Lys Cys Leu Leu Leu Val Ile Ser Met Ser Leu Gly Gly 
  1  5 10 15
 Cys Trp Ser Leu Met Ile His Leu Asp Gly Glu Arg Cys Ile Tyr Pro 
 20 25 30
```
The DNA molecule of ORFlO from the *Pseudomonas syringae* pv. *tomato* DC3000 CEL has a nucleotide sequence (SEQ. ID. No. 16) as follows:

```
atgaaacagg tagaagtcca gatcattact gaattgcctt gtcaggttct gatcctggag
caagaggcag tagcagaggg cttcaggttt cttacccgct tgatcgagga gtggaggtcc
ggaaagaatc gattcgaggc caagggtgaa tgcctcatgg tcgtacttct ggacggcgct
cgggcaggta tcggaggcct ttcgcgtgat ccgcatgccc ggggtgatat gggcaggcta
cgcctgtatat ccctcggag cgcctgttat ccctcggag cgcctgtatat ccctcggag
cgcctgtatat ccctcggag cgcctgtatat ccctcggag cgcctgtatat ccctcggag
cgcctgtatat ccctcggag cgcctgtatat ccctcggag cgcctgtatat ccctcggag
```

The protein or polypeptide encoded by Pto DC3000 CEL ORFlO has an amino acid sequence (SEQ. ID. No. 17) as follows:

```
Met Lys Gln Val Glu Val Gln Ile Ile Thr Glu Leu Pro Cys Gln Val
1   5  10  15
Leu Ile Leu Glu Glu Glu Ala Val Ala Glu Gly Phe Arg Phe Leu Thr
20  25 30
Arg Leu Ile Glu Glu Trp Arg Ser Gly Lys Asn Arg Phe Glu Ala Lys
35 40 45
Gly Glu Cys Leu Met Val Val Leu Leu Asp Gly Ala Leu Ala Gly Ile
50  55  60
Gly Gly Leu Ser Arg Asp Pro His Ala Arg Gly Asp Met Gly Arg Leu
65  70  75 80
Arg Arg Leu Tyr Val Ala Ser Ala Ser Arg Gly Glu Leu Gly Lys
85  90 95
Thr Leu Val Asn Arg Leu Val Glu His Ala Ala Glu Gly Phe Phe Ala
100 105 110
Val Arg Leu Phe Thr Asp Thr Pro Ser Gly Ala Lys Phe Tyr Leu Arg
115 120 125
Cys Gly Phe Glu Ala Val Asp Glu Val His Ala Thr His Ile Lys Leu
130 135 140
Leu Arg Arg Val
145
```

A DNA molecule which contains the EEL of *Pseudomonas syringae* pv. *tomato* DC3000 has a nucleotide sequence (SEQ. ID. No. 18) as follows:
ggatccagcg gcgtattgtc gtggcgatgg aacgcgttac ggattttcag cacaccggta
tcgatgaaca ggtggccgtt gcgggcgttg cgggtcggca tgacacaatc gaacatatca
acgccacggc gcacaccttc gaccagatct tcgggcttgc ctacacccat caagtaacga
ggtttgtctg ctggcataag gcccggcagg taatccagca ccttgatcat ctcgtgcttg
ggctcgccca ccgacagacc gccaatcgcc aggccgtcaa agccgatctc atccaggcct
tcgagcgaac gcttgcgcag gttctcgtgc atgccaccct gaacaatgcc gaacagcgcg
gcagtgtttt cgccgtgcgc gaccttggag cgcttggccc agcgcaacga cagctccatg
gagacacgtg ctacgtcttc gtcggccggg tacggcgtgc actcatcgaa aatcatcacg
acgtccgaac ccaggtcacg ctggacctgc atcgactctt ccgggcccat gaacaccttg
gcaccatcga ccggagaggc gaaggtcacg ccctcctcct tgatcttgcg catggcgccc
aggctgaaca cctgaaaacc gccagagtcg gtcagaatcg gccctttcca ctgcatgaaa
tcgtgcaggt cgccgtggcc cttgatgacc tcggtgcccg gacgcagcca caagtggaag
gtttgccca gaatcatctg cgcaccggtg gcctcgatat cacgcggcaa catgcccttg
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9480
ttcgagcgc ggtgtggcgt gcgcgagcgc cttttttttt cttttttttt cttttttttt
9540
ttcgagcgc ggtgtggcgt gcgcgagcgc cttttttttt cttttttttt cttttttttt
9600
Several undefined nucleotides exist in SEQ. ID. No. 18, however these appear to be present in intergenic regions. The DNA molecule of Pseudomonas syringae pv. tomato DC3000 EEL contains a number of ORFs. One of the products encoded by The EEL is a homolog of TnpA' from P. stutzeri. An additional four products are produced by ORF1-4, respectively. The nucleotide sequences for a number of these ORFs and their encoded protein or polypeptide products are provided below.

The DNA molecule of ORF1 from the Pseudomonas syringae pv. tomato DC3000 EEL has a nucleotide sequence (SEQ. ID. No. 19) as follows:

```
atgagacccg tcggtggacc ggctccaggc tattatccgc caacctatga agctgagcgt 60
cccactgcgc aagctgcagg aaacgatcgc gcccgatctt cacaggccag ttcctctcca
```

```
gtggccggcc ggctgaccga actgcacgcc ggcttgccca ccgccaccca caggtgtcgg 100
caaggctttg gcctgaggct tgcggccggttgaccggcg gagatgggcaa ccatcggctc
```

```
tccctcancggg antgatccng gaccgnaacc cttannggaa taatccggtt aaancggcta 1140
```

```
tnaanaggg ttcctcata tggaatgug gcgccacacc ccttttggna 11458
```

See the image for the full nucleotide sequence.
The protein or polypeptide encoded by Pto DC3000 EEL ORFl has an amino acid sequence (SEQ. ID. No. 20) as follows:

```
Met Arg Pro Val Gly Gly Pro Ala Pro Gly Tyr Tyr Pro Pro Thr Tyr
5 10 15
Glu Ala Glu Arg Pro Thr Ala Glu Ala Ala Gly Asp Arg Ala Arg
20 25 30
Ser Ser Glu Ala Ser Ser Ser Pro Ala Ala Ser Val Ala Pro Glu Thr
35 40 45
Pro Met Leu Gly Asp Leu Lys Arg Phe Pro Ala Gly Arg Tyr Pro Asp
50 55 60
Met Lys Val Gly Asn Ile Arg Leu Lys Ile Glu Gly Glu Glu Gly Glu
65 70 75 80
Gly Lys Asp Gly Val Lys His Thr Arg Arg Arg Lys Pro Asp Ala Ala
85 90 95
Gly Ser Ser His Val His Gly Gly Ser Val Ala Ser Thr Ser Ala
100 105 110
Ser Ala Glu Ser Lys Ala Leu Glu Asp Thr Asn Phe Lys Ala Ser Asp
115 120 125
Leu Ala Glu Leu Ala Arg Thr Cys Glu Ser Pro His Pro Tyr Ala Leu
130 135 140
Ala Pro Ser Lys Ala Gly Lys Ser Ser Glu Leu Ser Ala Asn Val
145 150 155 160
```
Val Ser Ile Leu Leu Gln Glu Gly Lys His Ala Leu Glu Gln Arg Leu
Glu Ala Gln Gly Leu Lys Leu Ala Asp Val Val Val Ser Glu Gly Arg
Asp His Leu His Ile Asn Leu Asn Tyr Leu Leu Met Asp Ser Cys Leu
Gly Thr Ser Lys Gly Leu Trp Ala Pro Asp Ser Asn Asp Lys Lys Leu
Ile Ala Lys Ala Ala Arg Tyr Phe Asp Asp Phe Asn Ala Gln Lys Leu
Pro Glu Leu Ala Pro Leu Thr Lys Met Lys Ser Lys Asp Ser Leu Gly
Val Met Arg Glu Leu Leu Arg Asp Ala Pro Gly Leu Val Ile Gly Glu
Gly His Asn Ser Thr Ser Lys Arg Glu Leu Ile Asn Asp Met Lys
Ser Leu Lys Ala Ser Gly Val Thr Thr Leu Phe Met Glu His Leu Cys
Ala Glu Ser His Asp Lys Ala Leu Asn Asn Tyr Leu Ser Ala Pro Lys
Gly Ser Pro Met Pro Ala Arg Leu Lys Asp Tyr Leu Leu Met Leu Gly Ser
Gln Gly His Gln Ala Pro Glu Glu Leu His Thr Tyr Asn Phe Thr
Thr Leu Val Glu Ala Ala Lys His Ala Gly Leu Arg Val Val Ser Leu
Aasp Thr Thr Ser Thr Tyr Met Ala Pro Glu Leu Ala Glu Ile Lys Arg
Ala Glu Ala Met Asn Tyr Ala Glu Lys Ile Arg Leu Ser Lys
Pro Glu Gly Lys Trp Val Ala Phe Val Gly Ala Thr His Ala His Thr
Cys Aasp Gly Val Pro Gly Leu Ala Glu His Gly Val Arg Ser Leu
Val Ile Aasp Leu Gly Leu Lys Ser Arg Ala Thr Val Aasp Ile Aan
Val Lys Asn Tyr Gly Gly Leu Leu Aan Pro Aasp Val Arg Leu Ser Tyr
Lyisa

The DNA molecule of ORF2 from the Pseudomonas syringae pv. tomato DC3000 EEL has a nucleotide sequence (SEQ. ID. No. 21) as follows:

atgcaaaaga cgaccctatg ggctttagcc tttgcaatgt tggcagggtg tggggtttcg
60
gggccggcgc cggaagtga tattcagggt gcccaggcag agatgaasac accyytaaa
120
tsaattcgg atgcctacac ctcaaaaaa ctggatgctg tgctggaagc ccgcaccaac
180
aaaagttata tgaataaagg tcagctgatc gaccttgtat caggagcgtt tttaggaaca
240
cacagctgt caacatgtt gtygggctca gcggagggtc tgggtgaagc ggcctagcaac
300
tggagagtc tgagttgct gttctatttc gcggctactcg aagcttttcg aagatcccaa
360
The protein or polypeptide encoded by Pto DC3000 EEL ORF2 has amino acid sequence (SEQ. ID. No. 22) as follows:

```
Met Gln Lys Thr Thr Leu Trp Ala Leu Ala Phe Ala Met Leu Ala Gly
1  5 10 15
Cys Gly Val Ser Gly Pro Ala Pro Gly Ser Asp Ile Gln Gly Ala Gln
20 25 30
Ala Glu Met Lys Thr Pro Val Lys Leu Asn Leu Asp Ala Tyr Thr Ser
35 40 45
Lys Lys Leu Asp Ala Val Leu Glu Ala Arg Thr Asn Lys Ser Tyr Met
50 55 60
Asn Lys Gly Gin Leu Ile Asp Leu Val Ser Gly Ala Phe Leu Gly Thr
65 70 75 80
Pro Tyr Arg Ser Asn Met Leu Val Gly Ser Ala Asn Val Pro Glu Gin
85 90 95
Leu Val Ile Asp Phe Arg Gly Leu Asp Cys Phe Ala Tyr Leu Asp Tyr
100 105 110
Val Glu Ala Phe Arg Ser Thr Ser Gin Gin Asp Phe Val Arg Asn
115 120 125
Leu Val Gin Val Arg Tyr Lys Gly Asp Val Asp Phe Leu Asn Arg
130 135 140
Lys His Phe Phe Thr Asp Trp Ala Tyr Gly Thr Ala Tyr Pro Val Ala
145 150 155 160
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165 170 175
Arg Leu Asn Glu Arg Ala Lys Gly Val Tyr Leu Pro Gly Leu Pro
180 185 190
Val Val Glu Arg Ser Met Thr Tyr Ile Pro Ser Arg Leu Val Asp Ser
195 200 205
Gln Val Ser His Leu Arg Thr Gly Asp Tyr Ile Gly Ile Tyr Thr
210 215 220
Pro Ala Ser Arg Ala Gly Cys Asp Thr Arg Phe Leu Tyr Arg Asp
225 230 235 240
Gly
```

The DNA molecule of ORF3 from the *Pseudomonas syringae pv. tomato* DC3000 EEL has a nucleotide sequence

```
-continued
tcgcagcagg attttgtgag gaatctcgtt caggttcgtt acaagggtgg cgatgttgac
tttttgaatc gcaagcactt tttcacggat tgggcttacg gaacggcata ccctgtggcg
gatgacatta ccgcgcagat aagccccggt gcggtaagtg tcagaaaacg ccttaatgaa
agggccaaag gcaaagtcta tctgccaggg ttgcctgtgg ttgagcgtag catgaagtat
gccgagcc gcctggtgga cagtcaggtg gtcgagccac ccgcaaccc cgctggtgc
ggcata gcctggtgga tggagccac ccgcaaccc gtcgagcgg
ggataa

The DNA molecule of ORF3 from the *Pseudomonas syringae pv. tomato* DC3000 EEL has a nucleotide sequence

```
```
```
The protein or polypeptide encoded by Pto DC3000 EEL ORF3 has an amino acid sequence (SEQ. ID. No. 24) as follows:

```
Met Arg Ala Tyr Lys Asn Leu Thr Ala Lys Ile Gly Phe Leu Leu 1 5 10 15
Ala Leu Thr Ile Ile Gly Thr Ser Leu Pro Ala Phe Ala Val Asn Asp 20 25 30
Cys Asp Leu Asp Asn Ser Thr Gly Ala Thr Cys Gly Gly Asn 35 40
Asp Lys Asp Leu Asp Asn Asp Asn Ser Thr Asp Ala Ala Phe Gly 50 55 60
Asn Asp Lys Asp Met Asp Asn Asp His His Thr Asp Ala Ala Phe 65 70 75 80
Gly Asn Asp Lys Ala Leu Asp Asn Asp His His Thr Asp Ala Ala Phe 85 90 95
Gly Gly Asn Asp Lys Leu Asp Asn Asp Asn Lys Thr Asp Ala Ala 100 105 110
Phe Gly Gly Asn Asp Arg Leu Asp Asn Asn Thr Asp Asn Tyr 115 120 125
Aen Gly Thr Pro Ser Ala Ala Lye Lys 130 135
```

*Ps. syringae pv. tomato* DC3000 EEL ORF3 has now been shown to significantly reduce virulence when mutated. Perhaps more interestingly, overexpression strongly increases lesion size. Hence, this effector is biologically active and appears to have a key role in symptom production.

The DNA molecule of ORF4 from the *Pseudomonas syringae pv. tomato* DC3000 EEL has a nucleotide sequence (SEQ. ID. No. 25) as follows:

```
atgaacaaga tcgtctacgt aaaaagcttcttttcagtcgt ctgcaagctg a 60
```

The protein or polypeptide encoded by Pto DC3000 EEL ORF4 has an amino acid sequence (SEQ. ID. No. 26) as follows:

```
atgacgatsgctcttggtacgccgtcttggtgcgccgccccagggagggagtctcggt 60
```
The EEL of *Pseudomonas syringae* pv. *syringae* B728a contains a number of ORFs. Two of the open reading frames appear to be mobile genetic elements without comparable homologs in EELs of other *Pseudomonas syringae* variants. An additional four products are produced by ORF1-2 and ORF5-6, respectively. The nucleotide sequences for a number of these ORFs and their encoded protein or polypeptide products are provided below.

The DNA molecule of ORF1 from the *Pseudomonas syringae* pv. *syringae* B728a EEL has a nucleotide sequence (SEQ. ID. No. 27) as follows:

```
ATGGGTTCGG TATCGTCAAA AGCATCTGTC ATTTCCTTCG GCACTTTCG CGCATCATA
60
ACAAACTCTC CAGAGGATC CTCAGTCCAT CAACGAGCCA GGACGCCAAG GTGCGGTGAG
120
CTTCAGGGGC CCAAGTGAG CAGATTGATG CCTTACCAGC AGGCCTAATG AGGTGTGGCC
180
CGATGGCCTA ATCCGCATT TAAACAGGAC GATGCACCCC ACCAGATGGG GTATGGAGAA
240
TCGTTCTACC AATAAAACG AGACTGTTG GTGTGCTGCT OAAATGGA GATAAAGCCG
300
TTTGAGGATG TCTGAGATG TCTGGAGGATG GGCTCACTAC TACAGGGACA 420
CGACACCCAAG CGAGTATGTG GCCAAATT ATGCACAATG TGGACATCTT TATAAGGCT
540
CTCGTCCA CCTCTTCACG CGACATATC CTGCATATT TTACTGATG TACCAGGAG 600
CTGCCACAA GGTGTGGCA CCAAGGATGG ACCAATGGGG GTGAGCTAC GTGAGGAGGA
660
ACACACCCG CGAGATATGT GCAAAATT ATGCAACATG TGGACATCTT TATAAGGCC
720
CTCTCGATG CTCAGTGGTG CGAGGGCACT ATGCGCAAG ATATCACTTG 780
TGAGCGGGCA AAGCAGTGGC CGACATATG GCAACTGGG CTAAGGGTAC CTGCGGTC
840
CGATGCTT GACACGCTGG GGGGCTGGCA CTGCGGCTG ATGACCTGG CTCGCTG
900
GATCGTGGCG CTGACATTG CCAATTGAAA GACTTATGGA AGGTTTCTC 960
GAACATAATC GA
972
```
The protein or polypeptide encoded by Psy\textsubscript{B728a EEL} ORF1 has an amino acid sequence (SEQ. ID. No. 28) as follows:

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</table>

As indicated in Table 1 (see Example 2), the DNA molecule encoding this protein or polypeptide bears significant homology to the nucleotide sequence from \textit{Pseudomonas syringae pv. phaseolicola} which encodes \textit{AvrPphC}.

The DNA molecule of ORF2 from the \textit{Pseudomonas syringae pv. syringae} B728a EEL has a nucleotide sequence (SEQ. ID. No. 29) as follows:
The protein or polypeptide encoded by psy B728a EEL ORF2 has an amino acid sequence (SEQ. ID. No. 30) as follows:

```
Met Arg Ile His Ser Ser Gly His Gly Ile Ser Gly Pro Val Ser Ser
  1   5   10   15
Ala Glu Thr Val Glu Lys Ala Val Gln Ser Ser Ala Gln Ala Gln Asn
  20  25  30  35  40
Glu Ala Ser His Ser Gly Pro Ser Glu His Pro Gly Ser Arg Ser Cys
  45  50  55  60
Gln Ala Arg Pro Asn Tyr Pro Tyr Ser Ser Val Lys Thr Arg Leu Pro
  65  70  75  80
Pro Val Ala Ser Ala Gly Gin Ser Leu Ser Glu Thr Pro Ser Ser Leu
  85  90  95
Pro Gly Tyr Leu Leu Arg Arg Leu Asp Arg Arg Pro Leu Asp Gln
  93  98 103 108 113 118
Asp Ala Ile Lys Gly Leu Ile Pro Ala Asp Glu Ala Val Gly Glu Ala
 105 110
Arg Arg Ala Leu Pro Phe Gly Arg Gly Asn Ile Asp Val Ala Glu
 115 120 125
Arg Ser Asn Leu Glu Ser Gly Ala Arg Thr Leu Ala Ala Arg Arg Leu
 130 135 140
Arg Lys Asp Ala Glu Thr Ala Gly His Glu Pro Met Pro Glu Asn Glu
 145 150 155 160
Asp Met Asn Trp His Val Leu Val Ala Met Ser Gly Glu Val Phe Gly
```
As indicated in Table 1 (see Example 2), the DNA molecule encoding this protein or polypeptide bears significant homology to the nucleotide sequence from *Pseudomonas syringae* pv. *phaseolicola* which encodes AvrPphE.

The DNA molecule of ORF5 from the *Pseudomonas syringae* pv. *syringae* B728a EEL has a nucleotide sequence (SEQ. ID. No. 31) as follows:

```plaintext
atgaatatct caggtccgaa cagacgtcag gggactcagg cagagaacac tgaaagcgct
tcgtcatcat cggtaactaa cccaccgcta cagcgtggcg agggcagacg tctgcgacgt
caggatgcgc tgccaacgga tatcagatac aacgccaacc agacagcgac atcaccgcaa
aacgcgcgcg cggcaggaag atatgaatca ggggccagct catccggcgc gaatgatact
cgcaggctg aaggttcaat gccttcgtcg tccgcccttt tacaatttcg cctcgccggc
gggcggaacc attctgagct ggaaaatttt catactatga tgctgaactc accgaaagca
tcacggggag atgctatacc tgagaagccc gaagcaatac ctaagcgcct actggagaag
atggaaccga ttaacctggc ccagttagct ttgcgtgata aggatctgca tgaatatgcc
gtaatggtct gtaaccaagt gaaaaagggt gaaggtccga actccaatat tacgcaagga
gatatcaagt tactgccgct gttcgccaaa gcggaaaata caagaaatcc cggcttgaat
cgcatacat tcgacaaaaa acaagcaatc gctgagtatg cgggttgttt accccccatt caaaaagatg
ccagaccacc atatagcctt ggatatccaa ctgagatacg gccatcgacc gtcgattgtc
```
The protein or polypeptide encoded by Psy B728a EEL ORF5 has an amino acid sequence (SEQ. ID. No. 32) as follows:

```
Met Asn Ile Ser Gly Pro Asn Arg Arg Gln Gly Thr Gln Ala Glu Asn
1  5  10  15
Thr Glu Ser Ala Ser Ser Ser Val Thr Asn Pro Pro Leu Gln Arg
20 25 30
Gly Gly Arg Arg Leu Arg Arg Gln Ala Leu Pro Thr Asp Ile
35 40 45
Arg Tyr Asn Ala Asn Gln Thr Ala Thr Ser Pro Gln Aen Ala Arg Ala
50 55 60
Ala Gly Arg Tyr Glu Ser Gly Ala Ser Gly Ala Asn Asp Thr
65 70 75 80
Pro Gln Ala Glu Gly Ser Met Pro Ser Ser Ala Leu Leu Gln Phe
85 90 95
Arg Leu Ala Gly Gly Arg Asn His Ser Glu Leu Gly Asn Asp His Thr
100 105 110
Met Met Leu Aen Ser Pro Lys Ala Ser Arg Gly Asl Ala Ile Pro Glu
115 120 125
Lys Pro Glu Ala Ile Pro Lys Arg Leu Gly Ile Met Gly Pro Ile
130 135 140
Asn Leu Ala Gln Leu Ala Leu Arg Asp Asp Leu His Gly Tyr Ala
145 150 155 160
Val Met Val Cys Asn Gln Val Val Gly Gly Gly Pro Asn Ser Aen
165 170 175
Ile Thr Gln Gly Asp Ile Lys Leu Leu Pro Leu Phe Ala Lys Ala Glu
180 185 190
Asn Thr Arg Aen Pro Gly Leu Aen Leu His Thr Phe Lys Ser His Lys
195 200 205
Asp Cys Tyr Gln Ala Ile Lys Glu Gln Asn Arg Asp Ile Gln Gly Asn
210 215 220
Lys Gln Ser Leu Ser Met Arg Val Val Tyr Pro Pro Phe Lys Lys Met
225 230 235 240
Pro Asp His His Ile Ala Leu Asp Ile Gln Leu Arg Tyr Gly His Arg
245 250 255
Pro Ser Ile Val Gly Phe Glu Ser Ala Pro Gly Aen Ile Asp Ala
260 265 270
Ala Glu Arg Glu Ile Ser Ala Leu Gly Asn Val Leu Ile Lys Met
275 280 285
Val Gly Aen Phe Leu Gly Tyr Ser Lys Thr Asp Cys Thr Met Phe Ala
290 295 300
```
The DNA molecule of ORF6 from the *pseudomonas syringae* pv. *syringae* B728a EEL has a nucleotide sequence (SEQ. ID. No. 33) as follows:

```
atgcctgtg aacggattga acagcaaaat acgctgtttg tttatctgtg cgtgggcacg
cattatact aacacgctgg aacggattga acagcaaaat acgctgtttg tttatctgtg cgtgggcacg
```

The protein or polypeptide encoded by Psy B728a EEL ORF6 has an amino acid sequence (SEQ. ID. No. 34) as follows:

```
Met Thr Leu Glu Arg Ile Glu Gln Asn Thr Leu Phe Val Tyr Leu
Cys Val Gly Thr Leu Ser Thr Pro Ala Ser Ser Thr Leu Leu Ser Asp
Ile Leu Ala Ala Asn Leu Phe His Tyr Gly Ser Ser Asp Gly Ala Ala
Phe Gly Leu Asp Glu Lys Asn Asn Glu Val Leu Leu Phe Gln Arg Phe
Asp Pro Leu Arg Ile Asp Glu Asp His Phe Val Ser Ala Cys Val Gln
Met Ile Glu Val Ala Ile Trp Arg Ala Lys Leu Leu His Gly His
Ser Ala Pro Leu Ala Ser Ser Thr Arg Leu Thr Lys Ala Gly Leu Met
Leu Thr Met Ala Gly Thr Ile Arg
```
The EEL of *Pseudomonas syringae* pv. *syringae* 61 contains a number of ORFs. One of the open reading frames encodes the outer membrane protein HopPsyA. The DNA molecule which encodes HopPsyA has a nucleotide sequence (SEQ. ID. No. 35) as follows:

```
gtgaacocca tccatgcaacg ototocacgc gtgaagagcgc tcagacatto aacagttgat  60
attcaggca tcmaactcgc ggt cacgtgtag gacacgcga cgaacgtgta cagagatgt  120
ggcggcgtcg acggtgcttc agggctgatg gcacagtacgc aacagagcga  180
tttctacgctg tcctgctgactg ggtacgatcg gacgagcgct gcacactcgc  240
gtcgacatcgc aacagacatt tgcacgggt ctgacattgg gctgagttgc  300
gttgcaacggc acggcgcaag ctgacatcg tgcagttgcct ggtgcgacag  360
tctgagctc gsgccgctgt gcgccgctgt gcctgaaca tcgctgagcc  420
gacacggtg cagacgcgtgc gactacctctg ttcttgtctg actgtgtaca  480
gctggcgtcgc tgcgtggtgc gccgctgtgc gcctgcgctgc gccgtgcgtgc  540
gacggtgctgc gcctgctgac ccacctgtgc gctgctgctgc gcctgcgctgc  600
tgtctgttct cctgctgttc ggttggtctg tgcgtgctgct gcctgctgctg  660
gctgctgctg cctgctgctg cctgctgctg cctgctgctg cctgctgctg  720
gctgctgctg cctgctgctg cctgctgctg cctgctgctg cctgctgctg  780
gctgctgctg cctgctgctg cctgctgctg cctgctgctg cctgctgctg  840
```

HopPsyA has an amino acid sequence (SEQ. ID. No. 36) as follows:

```
Val Asn Pro Ile His Ala Arg Phe Ser Ser Val Glu Ala Leu Arg His
1  5 10 15
Ser Asn Val Asp Ile Gln Ala Ile Lys Ser Glu Gly Gln Leu Glu Val
20 25 30
Asn Gly Lys Arg Tyr Glu Ile Arg Ala Ala Ala Asp Gly Ser Ile Ala
35 40 45
Val Leu Arg Pro Asp Gln Gln Ser Lys Ala Asp Lys Phe Lys Gly
50 55 60
Ala Ala His Leu Ile Gly Gly Ser Gin Arg Ala Gln Ile Ala Gln
65 70 75 80
Val Leu Asn Glu Lys Ala Ala Ala Val Pro Arg Leu Asp Arg Met Leu
85 90 95
Gly Arg Arg Phe Asp Leu Glu Lys Gly Ser Ser Ala Val Gly Ala
100 105 110
Ala Ile Lys Ala Ala Asp Ser Arg Leu Thr Ser Lys Gin Thr Phe Ala
115 120 125
Ser Phe Gln Gln Trp Ala Glu Lys Ala Glu Ala Leu Gly Arg Tyr Arg
130 135 140
Asn Arg Tyr Leu His Asp Leu Gln Gly His Asl Arg His Asn Ala
```
The remaining open reading frame, designated shcA, is a DNA molecule having a nucleotide sequence (SEQ. ID. No. 37) as follows:

atggagatgc ccgccttggc gtttgacgat aagggtgcgt gcaacatgat catcgacaag
60
gcattcgctc tgacgctgtt gcgcgacgac acgcatcaac gtttgttgct gattggtctg
120
cttgagccac acgaggatct acccttgcag cgcctgttgg ctggcgctct caaccccctt
180
gtgaatgccg gccccggcat tggctgggat gagcaaagcg gcctgtacca cgcttaccaa
240
agcatcccgc gggaaaaagt cagcgtggag atgctgaagc tcgaaattgc aggattggtc
300
gaatggatga agtgttggcg agaagcccgc acgtga
336

The encoded protein or polypeptide, ShcA, has an amino acid sequence (SEQ. ID. No. 38) as follows:

Met Glu Met Pro Ala Leu Ala Phe Asp Asp Lys Gly Ala Cys Asn Met
1  5 10 15
Ile Ile Asp Lys Ala Phe Ala Leu Thr Leu Leu Arg Asp Thr His
20 25 30
Gln Arg Leu Leu Ile Gly Leu Leu Glu Pro His Glu Asp Leu Pro
35 40 45
Leu Gln Arg Leu Leu Ala Gly Ala Asn Pro Leu Val Asn Ala Gly
50 55 60
In addition to the above DNA molecules and proteins or polypeptides, the present invention also relates to homologs of various DNA molecules of the present invention which have been isolated from other Pseudomonas syringae pathovars. For example, a number of AvrPphE, AvrPphF, and HopPsyA homologs have been identified from Pseudomonas syringae pathovars. The DNA molecule from Pseudomonas syringae pv. angulata which encodes an AvrPphE homolog has a nucleotide sequence (SEQ. ID. No. 39) as follows:

The amino acid sequence (SEQ. ID. No. 40) for the AvrPphE homolog of Pseudomonas syringae pv. angulata is as follows:
This protein or polypeptide has GC content of about 57 percent, an estimated isoelectric point of about 9.5, and an estimated molecular weight of about 41 kDa. The DNA molecule from Pseudomonas syringae pv. glycinea which encodes an AvrPphE homolog has a nucleotide sequence (SEQ. ID. No. 41) as follows:

atgagatc acagtgctgg tcacagcctg cccgcgccag gccctagcgt ggaaaccact gaaaaggctg ttcaatcatc atcggcccag aaccccgctt cttgcagttc acaaacagaa cgtcctgaag ccggttcgac tcaagtgcga ccgaactacc cttactcatc agtcaagaca cgcttgccac ccgtttcttc cacagggcag gccatttctg acacgccatc ttcattgtcc ggttacctgc tgttacgtcg gctcgaccga cgtccactgg atgaagacag tatcaaggct ctggttccgg cagacgaagc gttgcgtgaa gcacgccgcg cgttgccctt cggcaggggc

The DNA molecule from Pseudomonas syringae pv. glycinea which encodes an AvrPphE homolog has a nucleotide sequence (SEQ. ID. No. 41) as follows:

atgagatc acagtgctgg tcacagcctg cccgcgccag gccctagcgt ggaaaccact gaaaaggctg ttcaatcatc atcggcccag aaccccgctt cttgcagttc acaaacagaa cgtcctgaag ccggttcgac tcaagtgcga ccgaactacc cttactcatc agtcaagaca cgcttgccac ccgtttcttc cacagggcag gccatttctg acacgccatc ttcattgtcc ggttacctgc tgttacgtcg gctcgaccga cgtccactgg atgaagacag tatcaaggct ctggttccgg cagacgaagc gttgcgtgaa gcacgccgcg cgttgccctt cggcaggggc
The amino acid sequence (SEQ. ID. No. 42) for the AvrPphE homolog of Pseudomonas syringae pv. glycinea is as follows:

```
Met Arg Ile His Ser Ala Gly His Ser Leu Pro Ala Pro Gly Pro Ser
Val Glu Thr Thr Glu Lys Ala Val Gln Ser Ser Ser Ala Gln Aen Pro
Ala Ser Cys Ser Ser Gln Thr Glu Arg Pro Glu Ala Gly Ser Thr Gln
Val Arg Pro Aen Tyr Pro Tyr Ser Ser Val Lys Thr Arg Leu Pro Pro
Val Ser Ser Thr Gln Ala Ile Ser Asp Thr Pro Ser Ser Leu Ser
Gly Tyr Leu Leu Arg Leu Arg Asp Arg Arg Pro Leu Asp Glu Asp
Ser Ile Lys Ala Leu Val Pro Ala Asp Glu Ala Leu Arg Glu Ala Arg
Arg Ala Leu Pro Phe Gly Arg Gly Aen Ile Aen Val Aen Ala Gln Arg
Thr His Leu Gln Ser Gly His Arg Ala Val Ala Lys Arg Leu Arg
Lys Asp Ala Glu Arg Ala His Glu Pro Met Pro Glu Aen Asp Glu
Met Aen Trp His Val Leu Val Ala Met Ser Gly Gln Val Phe Gly Ala
Gly Aen Cys Gly Glu His Ala Arg Ile Ala Ser Phe Ala Tyr Gly Ala
Leu Ala Gln Glu Ser Gly Arg Ser Pro Arg Glu Lys Ile His Leu Ala
Glu Glu Pro Gly Lys Aep His Val Trp Ala Glu Thr Aep Aen Ser Ser
Ala Gly Ser Ser Pro Ile Val Met Aep Pro Trp Ser Aen Gly Val Ala
```
This protein or polypeptide has GC content of about 57 percent, an estimated isoelectric point of about 9.1, and an estimated molecular weight of about 41 kDa.

The DNA molecule from *Pseudomonas syringae* pv. *tabaci* which encodes an AvrPphE homolog has a nucleotide sequence (SEQ. ID. No. 43) as follows:

```
agagaattc acaagtgcctg tocaagctct gcgtcgccag gcotagcgt ggaaaccact
```

```
gaaaggtcg ttcactcact atccggccag aacoccgctt ctggcagttc aacacagaa
```

```
cgctcgcac cggcctgttc ttcgcagctct gcgtcgcgtc gcgttcgccg
```

```
cggcaggtct cggagtctgct gttgacggcg gcggcgagct cggcctctct gtcggcagct
```

```
cggcagcggc gcggcggcgg gcccggcagc ggccgagcag cccggaaaag atcacgtctg ggctgaaacg
```

```
gataattcca gcgctggctc ttcgcccatc gtcatggacc cgtggtctaa cggcgcagcc
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```
The amino acid sequence (SEQ. ID. No. 44) for the AvrPphE homolog of Pseudomonas syringae pv. tabaci is as follows:

```
Met Arg Ile His Ser Ala Gly His Ser Leu Pro Ala Pro Gly Pro Ser
1 5 10 15
Val Glu Thr Thr Glu Lys Ala Val Gln Ser Ser Ser Ala Gln Asn Pro
20 25 30
Ala Ser Cys Ser Ser Gln Thr Glu Arg Pro Glu Ala Gly Ser Thr Gln
35 40 45
Val Arg Pro Asn Tyr Pro Tyr Ser Ser Val Lys Thr Arg Leu Pro Pro
50 55 60
Val Ser Ser Thr Gly Gln Ala Ile Ser Asp Thr Pro Ser Ser Leu Pro
65 70 75 80
Gly Tyr Leu Leu Leu Arg Arg Leu Asp Arg Pro Leu Asp Glu Asp
85 90 95
Ser Ile Lys Ala Leu Val Pro Ala Asp Glu Ala Val Arg Gln Ala Arg
100 105 110
Arg Ala Leu Pro Phe Gly Arg Gly Asn Ile Asp Val Asp Ala Gln Arg
115 120 125
Thr His Leu Glu Ser Gly Ala Arg Ala Val Ala Lys Arg Leu Arg
130 135 140
Lys Asp Ala Glu Arg Ala Gly His Glu Pro Met Pro Gly Asn Asp Glu
145 150 155 160
Met Aan Trp His Val Leu Val Ala Met Ser Gly Gln Val Phe Gly Ala
165 170 175
Gly Aan Cys Gly Glu His Ala Arg Ile Ala Ser Phe Ala Tyr Gly Ala
180 185 190
Leu Ala Gln Glu Ser Gly Arg Ser Pro Arg Glu Lys Ile His Leu Ala
195 200 205
Glu Gln Pro Gly Lys Arg His Val Trp Ala Glu Thr Aep Aen Ser Ser
210 215 220
Ala Gly Ser Ser Pro Ile Val Met Aep Pro Trp Ser Aen Gly Ala Ala
225 230 235 240
Ile Leu Ala Glu Asp Ser Arg Phe Ala Lys Asp Arg Ser Ala Val Glu
245 250 255
Arg Thr Tyr Ser Phe Thr Leu Ala Met Ala Ala Glu Ala Gly Lys Val
260 265 270
Thr Arg Glu Thr Ala Glu Aen Val Leu Thr His Thr Ser Arg Leu
275 280 285
Gln Lys Arg Leu Ala Aep Gln Leu Pro Aen Val Ser Pro Leu Gly
290 295 300
Gly Arg Tyr Gin Gin Glu Lys Ser Val Leu Asp Gin Ala Phe Ala Arg
305 310 315 320
Arg Val Ser Aep Lys Leu Aen Ser Asp Aep Pro Arg Aen Gin Ala Leu Gln
325 330 335
Met Glu Ile Gln Ala Val Gin Val Ala Met Ser Leu Gly Ala Gly
340 345 350
Val Lys Thr Val Ala Arg Gin Ala Pro Lys Val Arg Gin Ala Arg
355 360 365
Ser Val Ala Ser Ser Lys Gin Met Pro Pro Pro Arg
370 375 380
```

This protein or polypeptide has GC content of about 57 percent, an estimated isoelectric point of about 9.3, and an estimated molecular weight of about 41 kDa.

Another DNA molecule from Pseudomonas syringae pv. tabaci which encodes a AvrPphE homolog has a nucleotide sequence (SEQ. ID. No. 45) as follows:
The encoded AvrPphE homolog has an amino acid sequence according to SEQ. ID. No. 46 as follows:

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A DNA molecule from *Pseudomonas syringae* pv. *glycinea* race 4 which encodes an avrPphE homolog has a 35 nucleotide sequence (SEQ. ID. No. 47) as follows:

```
atgagaattc acagtgctgg tcacagcctg cccgcgccag gccctagcgt ggaaaccact
gaaagacct tgtcaattct cctcgccagc aaccccgctt cttgcagttc acaaacagaa
cgtcctgaag ccggttcgac tcaagtgcga ccgaactacc cttactcatc agtcaagaca
cgcttgccac ccgtttcttc cacagggcag gccatttctg acacgccatc ttcattgtcc
ggttacctgc tgttacgtcg gctcgaccga cgtccactgg atgaagacag tatcaaggct
cgtggtctgg cagcaagagg tttgccgtag cggcggaggg cggggagggc
gacaggttag acggatcagc atcgggtacc tttgacattg gggctttcag
gaaaagggtg ttcgctaccg ttcacagagc aagcatctac atcagtgccg
gacaggttag acggatcagc atcgggtacc tttgacattg gggctttcag
gaaaagggtg ttcgctaccg ttcacagagc aagcatctac atcagtgccg
gacaggttag acggatcagc atcgggtacc tttgacattg gggctttcag
```
The encoded AvrPphE homolog has an amino acid sequence according to SEQ. ID. No. 48 as follows:

Met Arg Ile His Ser Ala Gly His Ser Leu Pro Ala Pro Gly Pro Ser
Val Glu Thr Thr Glu Lys Ala Val Gln Ser Ser Ser Ala Gln Asn Pro
Ala Ser Cys Ser Ser Gln Thr Glu Arg Pro Glu Ala Gly Ser Thr Gln
Val Arg Pro Asn Tyr Pro Tyr Ser Ser Val Lys Thr Arg Leu Pro Pro
Val Ser Ser Thr Gln Ala Ile Ser Asp Thr Pro Ser Ser Leu Ser
Gly Tyr Leu Leu Leu Arg Leu Asp Arg Pro Leu Asp Glu Asp
Ser Ile Lys Ala Leu Val Pro Ala Asp Glu Ala Leu Arg Glu Ala Arg
Arg Ala Leu Pro Phe Gly Arg Gly Asn Ile Asp Val Asp Ala Gln Arg
Thr His Leu Gln Ser Gly Ala Arg Ala Val Ala Lys Arg Leu Arg
Lys Asp Ala Glu Arg Ala Gly His Glu Pro Met Pro Glu Asp Arg Glu
Met Asn Trp His Val Leu Val Ala Met Ser Gly Gln Val Phe Gly Ala
Gly Asn Cys Gly Glu His Ala Arg Ile Ala Ser Phe Ala Tyr Gly Ala
Leu Ala Gln Glu Ser Gly Arg Ser Pro Arg Glu Lys Ile His Leu Ala
Glu Gln Pro Gly Lys Asp His Val Trp Ala Glu Thr Asp Ser Ser Ser
Ala Gly Ser Ser Pro Ile Val Met Asp Pro Trp Ser Asn Gln Val Ala
Ile Leu Ala Glu Asp Ser Arg Ala Lys Asp Arg Ser Ala Val Glu
Arg Thr Tyr Ser Phe Thr Leu Ala Met Ala Ala Glu Ala Gly Lys Val
Ala Arg Glu Thr Ala Glu Asn Val Leu Thr His Thr Ser Arg Leu
Gln Lys Arg Leu Ala Asp Gln Leu Pro Asn Val Ser Pro Leu Glu Gly
Gly Arg Tyr Gln Pro Gly Lys Ser Val Leu Asp Glu Ala Phe Ala Arg
Arg Val Ser Asp Lys Leu Asn Ser Asp Pro Arg Arg Ala Leu Gln
Met Glu Ile Glu Ala Val Gly Val Ala Met Ser Leu Gly Ala Glu Gly
Val Lys Thr Val Ala Arg Gln Ala Pro Lys Val Val Arg Gln Ala Arg
A DNA molecule from *Pseudomonas syringae* pv. *phaseolicola* strain B130 which encodes AvrPphE has a nucleotide sequence (SEQ. ID. No. 49) as follows:

```
atgagaatc acagtgctgg tcacagcctg cccgcgccag gccctagcgt ggaaaccact
60
gaaaaggctg ttcaatcatc atcggcccag aaccccgctt cttgcagttc acaaacagaa
120
cgtcctgaag ccggttcgac tcaagtgcga ccgaactacc ccggccgtct gcgctggaa
180
cgcttgccac ccgtttcttc cacagggcag gccatttctg acacgccatc ttcattgccc
240
ggttacctgc tgttacgtcg gctgtgaaac gaaagaagtgc gcggccggct ccgccgcttc
300
gcccgcgcg gcgcggccga ccggccggct cggccggcat gcggccggcg gcggccggct
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cggtgggtggt ggtgggtggt ggtgggtggt ggtgggtggt ggtgggtggt ggtgggtggt
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1080
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1140
tas
```

The encoded AvrPphE homolog has an amino acid sequence according to SEQ. ID. No. 50 as follows:

```
Met Arg Ile His Ser Ala Gly His Ser Leu Pro Ala Pro Gly Pro Ser
1 5 10 15
Val Glu Thr Thr Glu Lys Ala Val Glu Ser Ser Ser Ala Gln Asn Pro
20 25 30
Ala Ser Cys Ser Ser Gln Thr Glu Arg Pro Glu Ala Gly Ser Thr Gln
35 40 45
Val Arg Pro Asn Tyr Pro Tyr Ser Val Lys Thr Arg Leu Pro Pro
50 55 60
Val Ser Thr Gln Ala Ile Ser Asp Thr Pro Ser Ser Leu Pro
65 70 75 80
Gly Tyr Leu Leu Arg Arg Leu Asp Arg Arg Pro Leu Asp Gly Asp
85 90 95
Ser Ile Lys Ala Leu Val Pro Ala Asp Gla Ala Leu Arg Glu Ala Arg
100 105 110
```
A DNA molecule from *Pseudomonas syringae pv. angu- lata* strain Pa9 which encodes AvrPphE homolog has a nucleotide sequence (SEQ. ID. No. 51) as follows:

```
ATGAGAATTCAACAGTGCTGGTCACAGCCTGCCCTGCCAGGCGCTAGCCTGGAAACCAC
GAAAGGCTGGTCAATCATCCTCGGCCCAGAACCCCGCTTCTTACAGTTCACAACAGAA
CGTCCTGAAAGCCGGTCACTCACTGACCTCACTCCATGCACAGTACAG
GGTTACGTCACTGACCGATCAGAAAGCTGATAATACATCGACCAGAAGCTTGAGAAA
ATGGATGCACATCGTCACCCCACTGCAAAGCGCGCTCGCAGATCGCTGCACAAAGCGCTTGA
GAAAGATGCGCGCTGGCATGAGCGATGCCCAGGGAATGAGATGAGAATGAACTGGCA
ATGTTCCTGTCTGCATGCAAGGGCAGGTTGGCGCTGGAACATGCCTCCTATACAGCA
CTCGCTTACGGGGCCCTGGCTCAGGAAAGCGGGCGTAGTGGAAAGGTCGCACTTGCG
```

---

**US 6,852,835 B2**

-continued

```
Arg Ala Leu Pro Phe Gly Arg Gly Asn Ile Asp Val Asp Ala Glu Arg 115
Thr His Leu Gln Ser Gly Ala Arg Ala Val Ala Lys Arg Leu Arg 130
Lys Asp Ala Glu Arg Ala His Gly Pro Met Pro Glu Asn Asp Glu 145
Met Asn Trp His Val Leu Val Ala Met Ser Gly Gln Val Phe Gly Ala 165
Gly Asn Cys Gly Glu His Ala Arg Ile Ala Ser Phe Ala Tyr Gly Ala 180
Leu Ala Gln Glu Ser Gly Arg Ser Pro Arg Glu Lys Ile His Leu Ala 195
Glu Gln Pro Gly Lys Asp His Val Thr Asp Arg Ser Ser 210
Ala Gly Ser Ser Pro Ile Val Met Asp Pro Trp Ser Asn Gly Ala Ala 225
Ile Leu Ala Glu Asp Ser Arg Phe Ala Lys Asp Arg Ser Ala Val Glu 240
Arg Thr Tyr Ser Phe Thr Leu Ala Met Ala Ala Glu Ala Gly Lys Val 260
```

---

A DNA molecule from *Pseudomonas syringae pv. angu- lata* strain Pa9 which encodes AvrPphE homolog has a nucleotide sequence (SEQ. ID. No. 51) as follows:

```
atgagaattc acagtgctgg tcacagcctg cctgcgccag gccctagcgt ggaaaccact
```
The encoded AvrPphE homolog has an amino acid sequence according to SEQ. ID. No. 52 as follows:

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A DNA molecule from *Pseudomonas syringae pv. delphinii* strain PDDCC529 which encodes an *AvrPphE* homolog has a nucleotide sequence (SEQ. ID. No. 53) as follows:

```
adgaaaaec ataactgtgg cccactcatt cccgtgctct tgtgagctct gggcaagactg ccgcaatcatc attggctcaa ccgcagagcc aacgagccac ccccgtctcg ccatcagaga cttctgatgc ccgtccgtcc agtgtgcgta cgaactacc cttattcatga gtaaaacac ggttgcctcc cgttgcgctc gcagggcagc cactgtccgg gatgccgtct tcattacccg gctacttgct gttacgtcgg cttgaccatc gtccactgga tcaagacggt atcaaaggtt tgattccagc agatgaagcg gtgggtgaag cacgtcgcgc gttgcctttc ggcaggggca atatcgacgt ggatgcaag ccgcctccaact tggaaagcgg agcccgcaca ctcgcggcta ggcgtttgag aaaagatgcc gaggccgcgg gtcacgaacc aatgcctgca aatgaagata tgaactggca tgttcttgtt gcgatgtcag gacaggttctt tggcgcaggt aactgcgggg aacatgcccg catagcgagt ttcgcctacg gtgcactggc tcaggaaaaa gggcggaacg ccgatgagac tattcatttg gctgcgcaac gcggtaaaga ccacgtctgg gctgaaacgg acaattcaag cgctggatct tcaccggttg tcatggatcc gtggtcgaac ggtcctgcca tttttgcgga ggatagtcgg tttgccaaag atcgaagtac ggtagaacga acggattcct tcacgcttgc aactgctgct gaagcaggca agatcacgcg agagacggcc gagaatgctt tgacacaggc gaccagccgt ttgcagaaac gtcttgctga tcagaaaacg caagtctcgc cgcttgcagg agggcgctat cggcaagaaa attcggtgct tgatgacgcg ttcgcccgac gggcaagtgg caagttgagc aacaaggatc cgcggcatgc attacaggtg gaaatcgagg cggccgcagt tgcaatgtcg ctgggcgccc aaggcgtaaa agcggttgcg gaacaggccc ggacggtagt tgaacaagcc aggaaggtcg catctccca aggcacgcct cagcgagata cgtgaa
```

The encoded *avrPphE* homolog has an amino acid sequence according to SEQ. ID. No. 54 as follows:

```
Met Lys Ile His Asn Ala Gly Pro Ser Ile Pro Met Pro Ala Pro Ser
1  5  10 15
Ile Glu Ser Ala Gly Lys Thr Ala Glu Ser Ser Leu Ala Glu Pro Gln
20 25 30
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-continued
A DNA molecule from *Pseudomonas syringae* pv. *delphinii* strain PDDCC529 which encodes a homolog of *P. syringae* pv. *tomato* DC3000 EEL ORF2 has a nucleotide sequence (SEQ. ID. No. 55) as follows:

```
gtggttgagc gaaccggcac tgcatatcga aggcgtggag cagcctgctc gcgtatcacg
agccaaaatc aggtccgacg acgctttgga attacggtga atcagatgca aaagacgtcc
```

A DNA molecule from *Pseudomonas syringae* pv. *delphinii* strain PDDCC529 which encodes a homolog of *P. syringae* pv. *tomato* DC3000 EEL ORF2 has a nucleotide sequence (SEQ. ID. No. 55) as follows:

```
gtggttgagc gaaccggcac tgcatatcga aggcgtggag cagcctgctc gcgtatcacg
agccaaaatc aggtccgacg acgctttgga attacggtga atcagatgca aaagacgtcc
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The encoded protein or polypeptide has an amino acid sequence according to SEQ. ID. No. 56 as follows:

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</tr>
<tr>
<td>Ser Tyr Val Asn Lys Gly Glu Leu Ile Asp Leu Val Ser Gly Ala Phe</td>
<td>100 105 110</td>
</tr>
<tr>
<td>Leu Gly Thr Pro Tyr Arg Ser Asn Met Leu Val Gly Thr Glu Gln Ile</td>
<td>115 120 125</td>
</tr>
<tr>
<td>Pro Glu Gln Leu Val Ile Asp Phe Arg Gly Leu Asp Cys Phe Ala Tyr</td>
<td>130 135 140</td>
</tr>
<tr>
<td>Leu Asp Tyr Val Glu Ala Leu Arg Arg Ser Thr Ser Gln Glu Asp Phe</td>
<td>145 150 155 160</td>
</tr>
<tr>
<td>Val Arg Asn Leu Val Glu Val Arg Tyr Lys Gly Gly Asp Val Asp Phe</td>
<td>165 170 175</td>
</tr>
<tr>
<td>Leu Asn Arg Lys His Phe Phe Thr Asp Trp Ala Tyr Gly Thr Thr His</td>
<td>180 185 190</td>
</tr>
<tr>
<td>Pro Val Ala Asp Asp Ile Thr Thr Gln Ile Ser Pro Gly Ala Val Ser</td>
<td>195 200 205</td>
</tr>
<tr>
<td>Val Arg Lys Arg Leu Asn Gly Arg Ala Lys Gly Lys Val Tyr Leu Pro</td>
<td>210 215 220</td>
</tr>
<tr>
<td>Gly Leu Pro Val Val Glu Arg Ser Met Thr Tyr Ile Pro Ser Arg Leu</td>
<td>225 230 235 240</td>
</tr>
</tbody>
</table>
A DNA molecule from *Pseudomonas syringae pv. delphini* strain PDDCC529 ORF1 encodes a homolog of AvrPphF and has a nucleotide sequence (SEQ. ID. No. 57) as follows:

```
atgaaaaact catttgatct tcttgtcgac ggtttggcga aagactacag catgccgaat
  60
ttgccgaaca agaaacacga caatgaagtc tattgcttca cattccagag cgggctcgaa
 120
gtaaacattt atcaggacga ctgtcgatgg gtgcatttct ccgccacaat cggacaattt
180
cagacgcca gcataagacac gtcagccac gcacatcaca cggacatatt cagcttggga
240
agcctttct tccctttgg gatgacagga gagaggtcg gcttcttcct cagacgtgtt
300
cgctgtgatt aatgactagt cgctgaagcct tgcgactttt gcgtgactt gctgactga
360
gcgcggcgca tcagacggca atctagcttt agttaa
396```

The encoded AvrPphF homolog has an amino acid sequence according to SEQ. ID. No 58 as follows:

```
Met Lys Asn Ser Phe Asp Leu Leu Val Asp Gly Leu Ala Lys Asp Tyr
  1  5 10 15
Ser Met Pro Asn Leu Pro Asn Lys His Asp Asn Glu Val Tyr Cys
  20 25 30
Phe Thr Phe Gln Ser Gly Leu Glu Val Asn Ile Tyr Gln Asp Asp Cys
  35 40 45
Arg Trp Val His Phe Ser Ala Thr Ile Gly Gln Phe Glu Ala Asp Ser
  50 55 60
Asn Asp Thr Leu Ser His Ala Leu Gln Leu Asn Asp Ser Leu Gly
  65 70 75 80
Lys Pro Phe Phe Thr Phe Gly Met Asn Gly Glu Val Gly Val Leu
  85 90 95
His Thr Arg Val Pro Leu Ile Glu Met Asn Thr Val Glu Met Arg Lys
 100 105 110
Val Phe Glu Asp Leu Leu Asp Val Ala Gly Gly Ile Arg Ala Thr Phe
 115 120 125
Lys Leu Ser
130```

A DNA molecule from *Pseudomonas syringae pv. delphini* strain PDDCC529 ORF1 encodes a homolog of AvrPphF and has a nucleotide sequence (SEQ. ID. No. 59) as follows:
The encoded AvrPphF homolog has an amino acid sequence according to SEQ. ID. No. 60 as follows:

Met Ser Thr Ile Pro Gly Thr Ser Gly Ala His Pro Ile Tyr Ser Ser
1 5 10 15
Ile Ser Ser Pro Arg Asn Met Ser Gly Ser Pro Thr Pro Ser His Arg
20 25 30
Ile Gly Gly Thr Leu Thr Ser Ile His Gln Leu Ser Ala Ser Gln
35 40 45
Arg Glu Gln Phe Leu Asn Thr His Asp Pro Met Arg Lys Leu Arg Ile
50 55 60
Asn Asp Thr Pro Leu Tyr Arg Thr Thr Glu Lys Arg Phe Ile Gln
65 70 75 80
Glu Gly Lys Leu Ala Gly Asn Pro Lys Ser Ile Ala Arg Val Asn Leu
85 90
His Glu Gly Leu Glu Leu Asn Pro Leu Ala Ser Ile Leu Gly Asn Leu
100 105 110
Pro His Glu Ala Ser Ala Tyr Phe Pro Lys Ser Ala Arg Ala Ala Asp
115 120 125
Leu Lys Asp Pro Ser Leu Asn Val Met Thr Gly Ser Arg Ala Lys Asn
130 135 140
Asp Ile Arg Gly Tyr Ala His Asp His Val Ala Val Lys Met Arg
145 150 155 160
Leu Gly Asp Phe Leu Glu Gly Lys Val Tyr Ala Asp Thr Ser
165 170 175
Ser Val Ile Asp Gly Asp Glu Ala Ser Ala Leu Ile Val Thr Leu
180 185 190
Pro Lys Gly Gln Lys Val Pro Val Glu Ile Ile Pro Thr His Asn Asp
195 200 205
Asn Ser Asn Lys Gly Arg Gly
210 215

A DNA molecule from Pseudomonas syringae pv. syringae strain 226 encodes a homolog of HopPsyA and has a nucleotide sequence (SEQ. ID. No. 61) as follows:
The encoded HopPsyAhomolog has an amino acid sequence according to SEQ. ID No. 62 as follows:

Val Asn Pro Ile His Ala Arg Phe Ser Ser Val Glu Ala Leu Arg His
   1   5   10  15
Ser Asn Val Asp Ile Glu Ala Ile Lys Ser Gly Glu Leu Glu Val
   20  25  30
Asn Gly Lys Arg Tyr Glu Ile Arg Ala Ala Asp Gly Ser Ile Ala
   35  40  45
Val Leu Arg Pro Asp Gln Glu Ser Lys Ala Asp Gly Ser Ile Ala
   50  55  60
Ala Ala His Leu Ile Gly Gly Glu Ser Gin Arg Ala Gin Ile Ala Gin
   65  70  75  80
Val Leu Asn Glu Lys Ala Ala Val Pro Arg Leu Asp Arg Met Leu
   85  90  95
Gly Arg Arg Phe Asp Leu Glu Lys Gly Ser Ser Ser Val Gly Ala
  100 105 110
Ala Ile Lys Ala Ala Asp Ser Arg Leu Thr Ser Lys Gin Thr Phe Ala
  115 120 125
Ser Phe Gin Gin Thr Ala Glu Lys Ala Gln Leu Gly Arg Asp Thr
  130 135 140
Glu Ile Gly Ile Tyr Met Ile Tyr Lys Arg Asp Thr Pro Asp Thr Thr
  145 150 155 160
Pro Met Asn Ala Glu Gin Gln His Tyr Leu Glu Thr Leu Gin Ala
  165 170 175
Leu Asp Asn Lys Asn Leu Ile Ile Arg Pro Glu Ile His Asp Asp
  180 185 190
A DNA molecule from *Pseudomonas syringae* pv. *atrobotryae* strain B143 encodes a homolog of HopPsyA and has a nucleotide sequence (SEQ. ID. No. 63) as follows:

```
ATGACCCGTAAGAAGCAGTGGTGGAT
GTACAGGACGACGAAACTTCAATACCTTCTGTTATACAGGTTGAT
GGGTTGTTAATACCCCGGGACTTCAATACCTTCTGTTATACAGGTTGAT
CTGGCACTTCAATACCTTCTGTTATACAGGTTGAT
```

-continued

```
195 200 205
Arg Glu Glu Glu Leu Aep Leu Gly Arg Tyr Ile Ala Glu Aep Arg

210 215
Asn Ala Arg Thr Gyl Phe Phe Aep Met Val Pro Lys Aep Gln Arg Ala

220 225
Pro Glu Thr Aen Ser Gyl Arg Leu Thr Ile Gyl Val Gln Gln

230 235 240
Aep Lys His Leu Aep Ser Val Gln Val Gln Pro Aep Ala Tyr Arg Val Glu Arg

245 250 255
Ser Val Thr Gln Gly Lys Val Val Gly Pro Ala Tyr Gly Gln Gln

260 265 270
Thr Aep Ser Apl Ile Leu Tyr Ile Aen Gly Asp Leu Ala Lys Ala Val

275 280 285
Lys Leu Gly Glu Lys Leu Lys Leu Ser Gyl Ile Epl Pro Gln Gly

290 295 300
Phe Val Glu His Thr Pro Leu Ser Met Gln Thr Gln Leu Gln Val

305 310 315 320
Ser Tyr Ala Glu Ser Val Gln Gln Pro Ser Ser His Gly Gln Ala

325 330 335
Arg Thr His Val Ile Met Aep Leu Leu Gly Lys Pro Met Gln

340 345 350
Asn Arg Leu Lys Met Ala Leu Ala Glu Arg Gly Tyr Aep Pro Gln Aen

355 360 365
Pro Ala Leu Arg Ala Arg Aen

370 375
```
The encoded HopPsyAhomolog has an amino acid sequence according to SEQ. ID. No. 64 as follows:

**Met Asn Pro Ile Gln Thr Arg Phe Ser Ser Val Glu Ala Leu Arg His**

1  5  10  15

**Ser Glu Val Asp Val Gln Glu Leu Lys Ala His Gly Gln Ile Glu Val**

20  25  30

**Gly Gly Lys Cys Tyr Asp Ile Arg Ala Ala Asn Asp Leu Thr**

35  40  45

**Val Gln Arg Ser Asp Lys Gln Met Ala Met Ser Lys Phe Phe Lys Lys**

50  55  60

**Ser Glu Val Asp Val Gln Glu Leu Lys Ala His Gly Gln Ile Glu Val**

65  70  75  80

**Val Leu Asn Asp Lys Arg Gly Ser Ser Val Pro Arg Leu Ile Arg Glu**

85  90  95

**Gly Gln Thr His Leu Gly Arg Met Glu Phe Asn Ile Glu Glu Gly Gln**

100  105  110

**Gly Ser Ser Ala Ala Thr Ser Val Glu Asn Ser Arg Leu Pro Asn Gly**

115  120  125

**Arg Leu Val Asn Ser Ile Leu Gln Trp Val Glu Lys Ala Lys Ala**

130  135  140

**Asn Gly Ser Thr Ser Thr Ser Ala Leu Tyr Gin Ile Tyr Ala Lys Glu**

145  150  155  160

**Leu Pro Arg Val Glu Leu Leu Pro Arg Thr Glu His Arg Ala Cys Leu**

165  170  175

**Ala His Met Tyr Lys Leu Asn Gly Lys Asp Gly Ile Ser Ile Trp Pro**

180  185  190

**Gln Phe Leu Asp Gly Val Arg Gly Leu Gln Leu Lys His Arg Thr Lys**

195  200  205

**Val Phe Met Met Asn Asn Pro Lys Ala Ala Asp Phe Tyr Lys Ile**

210  215  220

**Glu Arg Ser Gly Thr Gin Phe Pro Asp Gin Ile Val Lys Ala Arg Leu**

225  230  235  240

**Thr Ile Asn Val Lys Pro Gin Phe Gin Lys Ala Met Val Asp Ala Ala**

245  250  255

**Val Arg Leu Thr Ala Glu Arg His Asp Ile Ile Thr Ala Lys Val Ala**

260  265  270

**Gly Pro Ala Lys Ile Gly Thr Ile Thr Asp Ala Ala Val Phe Tyr Val**

275  280  285

**Ser Gly Asp Phe Ser Ala Gin Thr Leu Ala Lys Glu Leu Gin Ala**

290  295  300

**Leu Leu Pro Asp Ala Phe Ile Asn His Thr Pro Ala Gly Met Gin**

305  310  315  320

**Ser Met Gly Lys Leu Cys Tyr Ala Gin Arg Thr Pro Gin Asp Arg**

325  330  335

**Thr Ser His Gly Met Ser Arg Ala Ser Ile Ile Glu Ser Ala Leu Ala**

340  345  350

**Asp Thr Ser Arg Ser Leu Glu Lys Leu Arg Asn Ala Phe Lys**

355  360  365
A DNA molecule from *pseudomonas syringae pv. tomato* strain DC3000 encodes a homolog of HopPtoA, identified herein as HopPtoA2, and has a nucleotide sequence (SEQ. ID. No. 65) as follows:

```
atgcacatca accaatccgc ccaacaaccg cctggcgttg caatggagag ttttcggaca
gcttccgacg cgtcccttgc ttcgagttct gtgcggtctg tcagcactac ctcgtgccgc
```

Although hopPtoA2 does not lie within the CEL, it is included here as a homolog of hopPtoA, which corresponds to CEL ORF5 as noted above. The encoded HopPtoA2 protein or polypeptide has an amino acid sequence according to SEQ. ID. No. 66 as follows:

```
Met His Ile Asn Gln Ser Ala Gln Gln Pro Pro Gly Val Ala Met Glu
Ser Phe Arg Thr Ala Ser Asp Ala Ser Leu Ala Ser Ser Ser Ser Val Arg
```

```
1 5 10 15
```

```
Ser Ala Gly Tyr Asn Pro Asp Asn Pro Ala Phe Arg Leu Glu
370 375 380
```

```
Ala Asp Asn Pro Ala Phe Arg Leu Glu
```

```
385 390 395
```
Ser Val Ser Thr Thr Ser Cys Arg Asp Leu Gin Ala Ile Thr Asp Tyr
35 40 45
Leu Lys His His Val Phe Ala Ala His Arg Phe Ser Val Ile Gly Ser
50 55 60
Pro Asp Glu Arg Asp Ala Ala Ala His Asn Glu Gln Ile Asp Ala
65 70 75 80
Leu Val Glu Thr Arg Ala Asn Arg Leu Tyr Ser Glu Gly Glu Thr Pro
85 90 95 95
 Ala Thr Ile Ala Glu Thr Phe Ala Lys Ala Glu Lys Phe Asp Arg Leu
100 105 110
 Ala Thr Thr Ala Ser Ser Ala Phe Glu Asn Thr Pro Phe Ala Ala Ala
115 120 125
 Ser Val Leu Gin Tyr Met Gin Pro Ala Ile Asn Lys Gly Asp Trp Leu
130 135 140
 Ala Thr Pro Leu Lys Pro Leu Thr Pro Leu Ile Ser Gly Ala Leu Ser
145 150 155 160
 Gly Ala Met Asp Gin Val Gly Thr Lys Met Met Asp Arg Ala Arg Gly
165 170 175 175
 Asp Leu His Tyr Leu Ser Thr Ser Pro Asp Lys Leu His Asp Ala Met
180 185 190
 Ala Val Ser Val Lys Arg His Ser Pro Ala Leu Gly Arg Gin Val Val
195 200 205
 Asp Met Gly Ile Ala Val Gin Thr Phe Ser Ala Leu Asn Val Arg
210 215 220
 Thr Val Leu Ala Pro Ala Leu Ala Ser Arg Pro Ser Val Gin Gly Ala
225 230 235 240
 Val Asp Phe Gly Val Ser Thr Ala Gly Gin Leu Val Ala Asn Ala Gly
245 250 255
 Phe Gly Asp Arg Met Leu Ser Val Gin Ser Arg Gin Leu Arg Gly
260 265 270
 Gly Ala Phe Val Leu Gly Met Lys Asp Lys Gin Pro Lys Ala Ala Leu
275 280 285
 Ser Glu Gin Thr Asp Trp Leu Gin Ala Thr Tyr Lys Ala Ile Lys Ser Ala
290 295 300
 Ser Tyr Ser Gly Ala Ala Leu Asn Ala Gly Lys Arg Met Ala Gly Leu
305 310 315 320
 Pro Leu Asp Val Ala Thr Asp Gin Leu Lys Ala Val Arg Ser Leu Val
325 330 335
 Ser Ala Thr Ser Leu Thr Asn Gly Leu Ala Leu Ala Gly Gin Tyr
340 345 350
 Ala Gly Val Ser Lys Leu Gin Gin Met Ala Thr Lys Asn Ile Thr Asp
355 360 365
 Ser Ala Thr Lys Ala Ala Val Ser Gin Leu Ser Asn Leu Val Gly Ser
370 375 380
 Val Gly Val Phe Ala Gly Trp Thr Thr Ala Gly Leu Ala Thr Asp Pro
385 390 395 400
 Ala Val Lys Ala Glu Ser Phe Ile Gin Asp Lys Val Lys Ser Thr
405 410 415
The proteins or polypeptides used in accordance with the present invention are preferably produced in purified form (preferably at least about 80%, more preferably 90%, pure) by conventional techniques. Typically, the protein or polypeptide of the present invention is secreted into the growth medium of recombinant host cells (discussed infra). Alternatively, the protein or polypeptide of the present invention is produced but not secreted into growth medium. In such cases, to isolate the protein, the host cell (e.g., E. coli) carrying a recombinant plasmid is propagated, lysed by sonication, heat, or chemical treatment, and the homogenate is centrifuged to remove bacterial debris. The supernatant is then subjected to sequential ammonium sulfate precipitation. The fraction containing the protein or polypeptide of interest is subjected to gel filtration in an appropriately sized dextran or polyacrylamide column to separate the proteins. If necessary, the protein fraction may be further purified by HPLC.

DNA molecules encoding other EEL and CEL protein or polypeptides can be identified using a PCR-based methodology for cloning portions of the pathogenicity islands of a bacterium. Basically, the PCR-based strategy involves the use of conserved sequences from the hrpK and tRNA<sup>low</sup> genes (or other conserved border sequences) as primers for cloning EEL intervening regions of the pathogenicity island. As shown in FIGS. 2B–C, the hrpK and tRNA<sup>low</sup> genes are highly conserved among diverse Pseudomonas syringae variants. Depending upon the size of EEL, additional primers can be prepared from the originally obtained cDNA sequence, allowing for recovery of clones and walking through the EEL in a step-wise fashion. If full-length coding sequences are not obtained from the PCR steps, contigs can be assembled to prepare full-length coding sequences using suitable restriction enzymes. Similar PCR-based procedures can be used for obtaining clones that encode open reading frames in the CEL. As shown in FIG. 3, the CEL of diverse Pseudomonas syringae pathovars contain numerous conserved domains. Moreover, known sequences of the hrp/phr domain, hrp W, Aavr, or gstA can be used to prepare primers.

Using the above-described PCR-based methods, a number of DNA sequences were utilized as the source for primers. One such DNA molecule is isolated from the tRNA<sup>low</sup> gene of Pseudomonas syringae pv. tomato DC3000, which has a nucleotide sequence (SEQ. ID. No. 67) as follows:

gcctgatgg cggatctggg aagaggggc gattccaaaat cgcttccgga aagaagtggg 60
An additional DNA molecule which can be used to supply suitable primers is from the tRNA<sup>Met</sup> gene of <i>Pseudomonas syringae</i> pv. syringae B728a, which has a nucleotide sequence (SEQ. ID. No. 68) as follows:

| 60 | gcccctgatgg   cggaattggt   agacgcggcg   gattcaaaat   ccgttttcga   aagaagtggg |
| 85 | agttcgattc   tccctcgggg   cacca |

Another DNA molecule is isolated from the queA gene of <i>Pseudomonas syringae</i> pv. <i>tomato</i> DC3000, which has a nucleotide sequence (SEQ. ID. No. 69) as follows:

| 60 | atgcgcgtcg   ctgactttac   cttcgaactc   cccgattccc   tgattgctcg   tcacccgttg   gccgagcgtc   gcagcagtcg   tctgttgacc   cttgatgggc   cgacgggcgc   gctggcacat   cgtcaattca   ccgatttgct   cgacccattg   cgctcgccgc |
| 85 | gcccctgatgg   cggaattggt   agacgcggcg   gattcaaaat   ccgttttcga   aagaagtggg |

This DNA molecule encodes QueA, which has an amino acid sequence (SEQ. ID. No. 70) as follows:

| 1  | Met | Arg | Val | Ala | Asp | Phe | Thr | Phe | Glu | Leu | Pro | Asp | Ser | Leu | Ile | Ala |
| 5  |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |
| 10 |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |
| 15 |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |
| 20 | Arg | His | Pro | Leu | Ala | Glu | Arg | Ser | Ser | Arg | Leu | Leu | Thr | Leu | Aep |
| 25 |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |
| 30 |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |
| 35 | Gly | Pro | Thr | Gly | Ala | Leu | Ala | His | Arg | Gln | Phe | Thr | Aep | Leu | Leu | Glu |
| 40 |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |
| 45 |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |
| 50 | His | Leu | Arg | Ser | Gly | Leu | Met | Val | Phe | Asn | Aan | Thr | Arg | Val | Ile |
| 55 |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |
| 60 |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |
| 65 | Pro | Ala | Arg | Leu | Phe | Gly | Gln | Lys | Ala | Ser | Gly | Gly | Lys | Lys | Leu | Glu | Ile |
DNA molecules encoding other EEL and CEL proteins or polypeptides can also be identified by determining whether such DNA molecules hybridize under stringent conditions to a DNA molecule as identified above. An example of suitable stringency conditions is when hybridization is carried out at a temperature of about 37°C, using a hybridization medium that includes 0.9M sodium citrate ("SSC") buffer, followed by washing with 0.2×SSC buffer at 57°C. Higher stringency can readily be attained by increasing the temperature for either hybridization or washing conditions or decreasing the sodium concentration of the hybridization or wash medium. Nonspecific binding may also be controlled using any one of a number of known techniques such as, for example, blocking the membrane with protein-containing solutions, addition of heterologous RNA, DNA, and SDS to the hybridization buffer, and treatment with RNase. Wash conditions are typically performed at or below stringency. Exemplary high stringency conditions include carrying out hybridization at a temperature of about 42°C to about 65°C for up to about 20 hours in a hybridization medium containing 1M NaCl, 50 mM Tris-HCl, pH 7.4, 10 mM EDTA, 0.1% sodium dodecyl sulfate (SDS), 0.2% ficoll, 0.2% polyvinylpyrrolidone, 0.2% bovine serum albumin, and 50 μg/ml E. coli DNA, followed by washing carried out at between about 42°C to about 65°C in a 0.2×SSC buffer. Also encompassed by the present invention are nucleic acid molecules which contain conserved substitutions as compared to the above identified DNA molecules and, thus, encode the same protein or polypeptides identified above. Further, complementary sequences are also encompassed by the present invention.

The nucleic acid of the present invention can be either DNA or RNA, which can readily be prepared using the above identified DNA molecules of the present invention. The delivery of effector proteins or polypeptides can be achieved in several ways, depending upon the host being treated and the materials being used: (1) as a stable or plasmid-encoded transgene; (2) transiently expressed via Agrobacterium or viral vectors; (3) delivered by the type III secretion systems of disarmed pathogens or recombinant nonpathogenic bacteria which express a functional, heterologous type III secretion system; or (4) delivered via topical application followed by TAT protein transduction domain-mediated spontaneous uptake into cells. Each of these is discussed infra.
The DNA molecule encoding the protein or polypeptide can be incorporated in cells using conventional recombinant DNA technology. Generally, this involves inserting the DNA molecule into an expression system to which the DNA molecule is heterologous (i.e., not normally present). The heterologous DNA molecule is inserted into the expression system or vector in proper sense orientation and correct reading frame. The vector contains the necessary elements for the transcription and translation of the inserted protein-coding sequences.

U.S. Pat. No. 4,237,224 to Cohen and Boyer describes the production of expression systems in the form of recombinant plasmids using restriction enzyme cleavage and ligation with DNA ligase. These recombinant plasmids are then introduced by means of transformation and replicated in unicellular cultures including prokaryotic organisms and eukaryotic cells grown in tissue culture.

Recombinant genes may also be introduced into viruses, such as vaccinia virus. Recombinant viruses can be generated by transfection of plasmids into cells infected with virus. Suitable vectors include, but are not limited to, the following viral vectors such as lambda vector system gt11, gt WES.1B, Charon 4, and plasmid vectors such as pBR322, pBR325, pACYC177, pACYC1844, pUC8, pUC9, pUC18, pUC19, pLG339, pK290, pKCl37, pKC101, SV 40, pBluescript II SK+/- or SK+/- (see "Strategene Cloning Systems" Catalog (1993) from Stratagene, La Jolla, Calif., which is hereby incorporated by reference), pQE, pHE821, pGEX, pET series (see Studier et al., 1990). Recombinant molecules can be introduced into cells via transformation, particularly transduction, conjugation, mobilization, or electroporation. The DNA sequences are cloned into the vector using standard cloning procedures in the art, as described by Sambrook et al., 1989.

A variety of host-vector systems may be utilized to express the protein-encoding sequences. Primarily, the vector system must be compatible with the host cell used. Host-vector systems include, but are not limited to, the following: bacteria transformed with bacteriophage DNA, plasmid DNA, or cosmid DNA; microorganisms such as yeast containing yeast vectors; mammalian cell systems infected with virus (e.g., vaccinia virus, adenovirus, etc.); insect cell systems infected with virus (e.g., baculovirus); and plant cells infected by bacteria. The expression elements of these vectors vary in their strength and specificities. Depending upon the host-vector system utilized, any one of a number of suitable transcription and translation elements can be used.

Different genetic signals and processing events control many levels of gene expression (e.g., DNA transcription and messenger RNA (mRNA) translation).

Transcription of DNA is dependent upon the presence of a promoter which is a DNA sequence that directs the binding of RNA polymerase and thereby promotes mRNA synthesis. The DNA sequences of eukaryotic promoters differ from those of prokaryotic promoters. Eukaryotic promoters and accompanying genetic signals may not be recognized in or may not function in a prokaryotic system and, further, prokaryotic promoters are not recognized and do not function in eukaryotic cells.

Similarly, translation of mRNA in prokaryotes depends upon the presence of the proper prokaryotic signals which differ from those of eukaryotes. Efficient translation of mRNA in prokaryotes requires a ribosome binding site called the Shine-Dalgaro ("SD") sequence on the mRNA. This sequence is a short nucleotide sequence of mRNA that is located before the start codon, usually AUG, which encodes the amino-terminal methionine of the protein. The SD sequences are complementary to the 3'-end of the 16S rRNA (ribosomal RNA) and probably promote binding of mRNA to ribosomes by duplexing with the rRNA to allow correct positioning of the ribosome. For a review on maximizing gene expression, see Roberts and Lauer, 1979.

Promoters vary in their "strength" (i.e., their ability to promote transcription). For the purposes of expressing a cloned gene, it is desirable to use strong promoters in order to obtain a high level of transcription and, hence, expression of the gene. Depending upon the host cell system utilized, any one of a number of suitable promoters may be used. For instance, when cloning in E. coli, its bacteriophages, or plasmids, promoters such as the 17 phase promoter, lac promoter, trp promoter, recA promoter, ribosomal RNA promoter, the P6 and P7 promoters of coliphage lambda and others, including but not limited to, lacUV5, ompF, bla, lpp, and the like, may be used to direct high levels of transcription of adjacent DNA segments. Additionally, a hybrid trp-lacUV5 (tac) promoter or other E. coli promoters produced by recombinant DNA or other synthetic DNA techniques may be used to provide for transcription of the inserted gene.

Bacterial host cell strains and expression vectors may be chosen which inhibit the action of the promoter unless specifically induced. In certain operations, the addition of specific inducers is necessary for efficient transcription of the inserted DNA. For example, the lac operon is induced by the addition of lactose or IPTG (isopropylthio-beta-D-galactoside). A variety of other operons, such as trp, pro, etc., are under different controls.

Specific initiation signals are also required for efficient gene transcription and translation in prokaryotic cells. These transcription and translation initiation signals may vary in "strength" as measured by the quantity of gene specific messenger RNA and protein synthesized, respectively. The DNA expression vector, which contains a promoter, may also contain any combination of various "strong" transcription and/or translation initiation signals. For instance, efficient translation in E. coli requires an SD sequence about 7-9 bases 5' to the initiation codon ("ATG") to provide a ribosome binding site. Thus, any SD-ATG combination that can be utilized by host cell ribosomes may be employed. Such combinations include but are not limited to the SD-ATG combination from the cro gene or the N gene of coliphage lambda, or from the E. coli tryptophan E, D, C, B or A genes. Additionally, any SD-ATG combination produced by recombinant DNA or other techniques involving incorporation of synthetic nucleotides may be used.

Once the isolated DNA molecule encoding the polypeptide or protein has been cloned into an expression system, it is ready to be incorporated into a host cell. Such incorporation can be carried out by the various forms of transformation noted above, depending upon the vector/host cell system. Suitable host cells include, but are not limited to, bacteria, virus, yeast, mammalian cells, insect, plant, and the like.

Because it is desirable for recombinant host cells to secrete the encoded protein or polypeptide, it is preferable that the host cell also possess a functional type III secretion system. The type III secretion system can be heterologous to host cell (Ham et al., 1998) or the host cell can naturally possess a type III secretion system. Host cells which naturally contain a type III secretion system include many pathogenic Gram-negative bacteria, such as numerous Erwinia species, Pseudomonas species, Xanthomonas species, etc. Other type III secretion systems are known and...
still others are continually being identified. Pathogenic bacteria that can be utilized to deliver effector proteins or polypeptides are preferably disarmed according to known techniques, i.e., as described above. Alternatively, isolation of the effector protein or polypeptide from the host cell or growth medium can be carried out as described above.

Another aspect of the present invention relates to a transgenic plant which expresses a protein or polypeptide of the present invention and methods of making the same.

In order to express the DNA molecule in isolated plant cells or tissue or whole plants, a plant expressible promoter is needed. Any plant-expressible promoter can be utilized regardless of its origin, i.e., viral, bacterial, plant, etc. Without limitation, two suitable promoters include the nopaline synthase promoter (Fraley et al., 1983) and the cauliflower mosaic virus 35S promoter (O'Dell et al., 1985). Both of these promoters yield constitutive expression of coding sequences under their regulatory control.

While constitutive expression is generally suitable for expression of the DNA molecule, it should be apparent to those of skill in the art that temporally or tissue regulated expression may also be desirable, in which case any regulated promoter can be selected to achieve the desired expression. Typically, the temporally or tissue regulated promoters will be used in connection with the DNA molecule that are expressed at only certain stages of development or only in certain tissues.

In some plants, it may also be desirable to use promoters which are responsive to pathogen infiltration or stress. For example, it may be desirable to limit expression of the protein or polypeptide in response to infection by a particular pathogen of the plant. One example of a pathogen-inducible promoter is the gus promoter from potato, which is described in U.S. Pat. Nos. 5,750,874 and 5,723,760 to Strittmayer et al., which are hereby incorporated by reference.

Expression of the DNA molecule in isolated plant cells or tissue or whole plants also requires appropriate transcription termination and polyadenylation of mRNA. Any 3' regulatory region suitable for use in plant cells or tissue can be operably linked to the first and second DNA molecules. A number of 3' regulatory regions are known to be operable in plants. Exemplary 3' regulatory regions include, without limitation, the nopaline synthase 3' regulatory region (Fraley et al., 1983) and the cauliflower mosaic virus 3' regulatory region (Odell et al., 1985).

The promoter and a 3' regulatory region can readily be ligated to the DNA molecule using well known molecular cloning techniques described in Sambrook et al., 1989.

One approach to transforming plant cells with a DNA molecule of the present invention is particle bombardment (also known as biolistic transformation) of the host cell. This can be accomplished in one of several ways. The first involves propelling inert or biologically active particles at cells. This technique is disclosed in U.S. Pat. Nos. 4,945,050, 5,036,006, and 5,100,792, all to Sanford, et al. Generally, this procedure involves propelling inert or biologically active particles at the cells under conditions effective to penetrate the outer surface of the cell and to be incorporated within the interior thereof. When inert particles are utilized, the vector can be introduced into the cell by coating the particles with the vector containing the heterologous DNA. Alternatively, the target cell can be surrounded by the vector so that the vector is carried into the cell by the wake of the particle. Biologically active particles (e.g., dried bacterial cells containing the vector and heterologous DNA) can also be propelled into plant cells. Other variations of particle bombardment, now known or hereafter developed, can also be used.

Another method of introducing the DNA molecule into plant cells is fusion of protoplasts with other entities, either minicells, cells, lysosomes, or other fusible lipid-surfaced bodies that contain the DNA molecule (Fraley et al., 1982). The DNA molecule may also be introduced into the plant cells by electroporation (Fromm, et al., 1985). In this technique, plant protoplasts are electroporated in the presence of plasmids containing the DNA molecule. Electrical impulses of high field strength reversibly permeabilize biomembranes allowing the introduction of the plasmids. Electroporated plant protoplasts reform the cell wall, divide, and regenerate.

Another method of introducing the DNA molecule into plant cells is to infect a plant cell with Agrobacterium tumefaciens or Agrobacterium rhizogenes previously transformed with the DNA molecule. Under appropriate conditions known in the art, the transformed plant cells are grown to form shoots or roots, and develop further into plants. Generally, this procedure involves inoculating the plant tissue with a suspension of bacteria and incubating the tissue for 48 to 72 hours on regeneration medium without antibiotics at 25–28°C.

Agrobacterium is a representative genus of the Gram-negative family Rhizobiaceae. Its species are responsible for crown gall (A. tumefaciens) and hairy root disease (A. rhizogenes). The plant cells in crown gall tumors and hairy roots are induced to produce amino acid derivatives known as opines, which are catalyzed only by the bacteria. The bacterial genes responsible for expression of opines are a convenient source of control elements for chimeric expression cassettes. In addition, assaying for the presence of opines can be used to identify transformed tissue. Heterologous genetic sequences such as a DNA molecule of the present invention can be introduced into appropriate plant cells by means of the Ti plasmid of A. tumefaciens or the Ri plasmid of A. rhizogenes. The Ti or Ri plasmid is transmitted to plant cells on infection by Agrobacterium and is stably integrated into the plant genome (Schell, 1987).

Plant tissue suitable for transformation include leaf tissue, root tissue, meristems, zygotic and somatic embryos, and anthers.

After transformation, the transformed plant cells can be selected and regenerated. Preferably, transformed cells are first identified using, e.g., a selection marker simultaneously introduced into the host cells along with the DNA molecule of the present invention. Suitable selection markers include, without limitation, markers coding for antibiotic resistance, such as kanamycin resistance (Fraley et al., 1983). A number of antibiotic-resistance markers are known in the art and are continually being identified. Any known antibiotic-resistance marker can be used to transform and select transformed host cells in accordance with the present invention. Cells or tissues are grown on a selection media containing an antibiotic, whereby generally only those transformants expressing the antibiotic resistance marker continue to grow.

Once a recombinant plant cell or tissue has been obtained, it is possible to regenerate a full-grown plant therefrom. Thus, another aspect of the present invention relates to a transgenic plant that includes a DNA molecule of the present invention, wherein the promoter induces transcription of the first DNA molecule in response to infection of the plant by an oomycete. Preferably, the DNA molecule is stably inserted into the genome of the transgenic plant of the present invention.
Plant regeneration from cultured protoplasts is described in Evans et al., 1983, and Vasili, 1984 and 1986. It is known that practically all plants can be regenerated from cultured cells or tissues, including but not limited to, all major species of rice, wheat, barley, rye, cotton, sunflower, peanut, corn, potato, sweet potato, bean, pea, chicory, lettuce, endive, cabbage, cauliflower, broccoli, turnip, radish, spinach, onion, garlic, eggplant, pepper, celery, carrot, squash, pumpkin, zucchini, cucumber, apple, pear, melon, strawberry, grape, raspberry, pineapple, soybean, tobacco, tomato, sorghum, and sugarcane.

Means for regeneration vary from species to species of plants, but generally a suspension of transformed protoplasts or a petri plate containing transformed explants is first provided. Callus tissue is formed and shoots may be induced from callus and subsequently rooted. Alternatively, embryo formation can be induced in the callus tissue. These embryos germinate as natural embryos to form plants. The culture media will generally contain various amino acids and hormones, such as auxin and cytokinins. It is also advantageous to add glutamic acid and proline to the medium, especially for such species as corn and alfalfa. Efficient regeneration will depend on the medium, on the genotype, and on the history of the culture. If these three variables are controlled, then regeneration is usually reproducible and repeatable.

After the DNA molecule is stably incorporated in transgenic plants, it can be transferred to other plants by sexual crossing or by preparing cultivars. With respect to sexual crossing, any of a number of standard breeding techniques can be used depending upon the species to be crossed. Cultivars can be propagated in accord with common agricultural procedures known to those in the field.

Diseases caused by the vast majority of bacterial pathogens result in limited lesions. That is, even when everything is working in the pathogen’s favor (e.g., no triggering of the hypersensitive response because of R-gene detection of one of the effectors), the parasitic process still triggers defenses after a couple of days, which then stops the infection from spreading. Thus, the very same effectors that enable parasitism to proceed must also eventually trigger defenses. Therefore, premature expression of these effectors is believed to “turn on” plant defense mechanisms (i.e., prior to infection) and make the plant resistant to the specific bacteria from which the effector protein was obtained or many pathogens. An advantage of this approach is that it involves natural products and plants seem highly sensitive to pathogen effector proteins.

According to one embodiment, a transgenic plant is provided that contains a heterologous DNA molecule of the present invention. Preferably, the heterologous DNA molecule is derived from a plant pathogen EEL. When the heterologous DNA molecule is expressed in the transgenic plant, plant defenses are activated, imparting disease resistance to the transgenic plant. The transgenic plant can also contain an R-gene which is activated by the protein or polypeptide product of the heterologous DNA molecule. The R gene can be naturally occurring in the plant or heterologously inserted therein. A number of R genes have been identified in various plant species, including without limitation: RPS2, RPM1, and RPP5 from Arabidopsis thaliana; Cf2, C9, I2, Pto, and Prf from tomato; N from tobacco; L6 and M from flax; Xa21 from rice; and Hs1pr-1 from sugar beet. In addition to imparting disease resistance, it is believed that stimulation of plant defenses in transgenic plants of the present invention will also result in simultaneous enhancement in growth and resistance to insects.

According to another embodiment, a plant, transgenic or non-transgenic, is treated with a protein or polypeptide of the present invention. By treating, it is intended to include various forms of applying the protein or polypeptide to the plant. The embodiments of the present invention where the effector polypeptide or protein is applied to the plant can be carried out in a number of ways, including: 1) application of an isolated protein (or composition containing the same) or 2) application of bacteria which do not cause disease and are transformed with a gene encoding the effector protein of the present invention. In the latter embodiment, the effector protein can be applied to plants by applying bacteria containing the DNA molecule encoding the effector protein. Such bacteria are preferably capable of secreting or exporting the protein so that the protein can contact plant cells. In these embodiments, the protein is produced by the bacteria in plants.

Such topical application is typically carried out using an effector fusion protein which includes a transduction domain, which will afford transduction domain-mediated spontaneous uptake of the effector protein into cells. Basically, this is carried out by fusing an 11-amino acid peptide (YGRKKRRQRRR, SEQ. ID. No. 91) by standard rDNA techniques to the N-terminus of the effector protein, and the resulting tagged protein is taken up into cells by a poorly understood process. This peptide is the protein transduction domain (PTD) of the human immunodeficiency virus (HIV) TAT protein (Schwarze et al., 2000). Other PTDs are known and may possibly be used for this purpose (Prochiantz, 2000).

When the effector protein is topically applied to plants, it can be applied as a composition, which includes a carrier in the form, e.g., of water, aqueous solutions, slurries, or dry powders. In this embodiment, the composition contains greater than about 5 nM of the protein of the present invention.

Although not required, this composition may contain additional additives including fertilizer, insecticide, fungicide, nematicide, and mixtures thereof. Suitable fertilizers include (NH4)2NO3. An example of a suitable insecticide is Malathion. Useful fungicides include Captain.

Other suitable additives include buffering agents, wetting agents, coating agents, and, in some instances, abrading agents. These materials can be used to facilitate the process of the present invention.

According to another aspect of the present invention, a transgenic plant is provided that contains a heterologous DNA molecule that encodes a transcript or a protein or polypeptide capable of disrupting function of a plant pathogenic CEL product. Because the genes in the CELs are particularly important in pathogenesis, disrupting the function of their products in plants can result in broad resistance since CEL genes are highly conserved among Gram negative pathogens, particularly along species lines. An exemplary protein or polypeptide which can disrupt function of a CEL product is an antibody, polyclonal or monoclonal, raised against the CEL product using conventional techniques. Once isolated, the antibody can be sequenced and nucleic acids synthesized for encoding the same. These nucleic acids, e.g., DNA, can be used to transform plants.

Transgenic plants can also be engineered so that they are hypersusceptible and, therefore, will support the growth of nonpathogenic bacteria for biotechnological purposes. It is known that many plant pathogenic bacteria can alter the environment inside plant leaves so that nonpathogenic bacteria can grow. This ability is presumably based on changes in the plant caused by pathogen effector proteins. Thus,
transgenic plants expressing the appropriate effector genes can be used for these purposes.

According to one embodiment, a transgenic plant including a heterologous DNA molecule of the present invention expresses one or more effector proteins, wherein the transgenic plant is capable of supporting growth of compatible nonpathogenic bacteria (i.e., non-pathogenic endophytes such as various *Clavibacter* spp.). The compatible nonpathogenic bacteria can be naturally occurring or it can be recombinant. Preferably, the nonpathogenic bacteria is recombinant and expresses one or more useful products. Thus, the transgenic plant becomes a green factory for producing desirable products. Desirable products include, without limitation, products that can enhance the nutritional quality of the plant or products that are desirable in isolated form. If desired in isolated form, the product can be isolated from plant tissues. To prevent competition between the non-pathogenic bacteria which express the desired product and those that do not, it is possible to tailor the needs of recombinant, non-pathogenic bacteria so that only they are capable of living in plant tissues expressing a particular effector protein or polypeptide of the present invention.

The effector proteins or polypeptides of the present invention are believed to alter the plant physiology by shifting metabolic pathways to benefit the parasite and by activating or suppressing cell death pathways. Thus, they may also provide useful tools for efficiently altering the nutrient content of plants and delaying or triggering senescence. There are agricultural applications for all of these possible effects.

A further aspect of the present invention relates to diagnostic uses of the CEL and EEL. The CEL genes are universal to species of Gram negative bacteria, particularly pathogenic Gram-negative bacteria (such as *P. syringae*), whereas the EEL sequences are strain-specific and provide a “virulence gene fingerprint” that could be used to track the presence, origins, and movement (and restrict the spread through quarantines) of strains that are particularly threatening. Although the CEL and EEL have been identified in various pathovars of *Pseudomonas syringae*, it is expected that most all Gram-negative pathogens can be identified, distinguished, and classified based on the homology of the CEL and EEL genes.

According to one embodiment, a method of determining relatedness between two bacteria is carried out by comparing a nucleic acid alignment or amino acid alignment for a CEL of the two bacteria and then determining the relatedness of the two bacteria, wherein a higher sequence identity indicates a closer relationship. The CEL is particularly useful for determining the relatedness of two distinct bacterial species.

According to another embodiment, a method of determining relatedness between two bacteria which is carried out by comparing a nucleic acid alignment or amino acid alignment for an EEL of the two bacteria and then determining the relatedness of the two bacteria, wherein a higher sequence identity indicates a closer relationship. The EEL is particularly useful for determining the relatedness of two pathovars of a single bacterial species.

Given the methods of determining relatedness of bacteria species and/or pathovars, these methods can be utilized in conjunction with plant breeding programs. By detecting the “virulence gene fingerprint” of pathogens which are prevalent in a particular growing region, it is possible either to develop transgenic cultivars as described above or to identify existing plant cultivars which are resistant to the prevalent pathogens.

In addition to the above described uses, another aspect of the present invention relates to gene- and protein-based therapies for animals, preferably mammals including, without limitation, humans, dogs, mice, rats. The *P. syringae* pv. *syringae* B728a EEL ORF5 protein (SEQ. ID. No. 32) is a member of the *AvrRsv/Yop* protein family. *Yop* is injected into human cells by the *Versinia* type III secretion system, where it disrupts the function of certain protein kinases to inhibit cytokine release and promote programmed cell death. It is believed that the targets of many pathogen effector proteins (i.e., *P. syringae* effector proteins) will be universal to eukaryotes and therefore have a variety of potentially useful functions. In fact, two of the proteins in the *P. syringae* Hrp pathogenicity islands are toxic when expressed in yeast. They are HopPsyA from the *P. syringae* pv. *tomato* EEL and HopPtoA from the *P. syringae* pv. *tomato* DC3000 CEL. This supports the concept of universal eukaryote targets.

Thus, a further aspect of the present invention relates to a method of causing eukaryotic cell death which is carried out by introducing into a eukaryotic cell a cytotoxic *Pseudomonas* protein. The cytotoxic *Pseudomonas* protein is preferably HopPsyA (e.g., SEQ. ID. Nos. 36 (Psy 61), 62 (Psy 226), or 64 (Psy B143)) HopPtoA (SEQ. ID. No. 7), or HopPtoA2 (SEQ. ID. No. 66). The eukaryotic cell which is treated can be either in vitro or in vivo. When treating eukaryotic cells in vivo, a number of different protein- or DNA-delivery systems can be employed to introduce the effector protein into the target eukaryotic cell.

Without being bound by theory, it is believed that at least the HopPsyA effector proteins exert their cytotoxic effects through Mad2 interactions, disrupting cell checkpoint of spindle formation (see infra).

The protein- or DNA-delivery systems can be provided in the form of pharmaceutical compositions which include the delivery system in a pharmaceutically acceptable carrier, which may include suitable excipients or stabilizers. The dosage can be in solid or liquid form, such as powders, solutions, suspensions, or emulsions. Typically, the composition will contain from about 0.01 to 99 percent, preferably from about 20 to 75 percent of active compound(s), together with the carrier, excipient, stabilizer, etc.

The compositions of the present invention are preferably administered in injectable or topical-applied dosages by solution or suspension of these materials in a physiologically acceptable diluent with a pharmaceutical carrier. Such carriers include sterile liquids, such as water and oils, with or without the addition of a surfactant and other pharmaceutically and physiologically acceptable carrier, including adjuvants, excipients or stabilizers. Illustrative oils are those of petroleum, animal, vegetable, or synthetic origin, for example, peanut oil, soybean oil, or mineral oil. In general, water, saline, aqueous dextrose and related sugar solution, and glycols, such as propylene glycol or polyethylene glycol, are preferred liquid carriers, particularly for injectable solutions.

Alternatively, the effector proteins can also be delivered via solution or suspension packaged in a pressurized aerosol container together with suitable propellants, for example, hydrocarbon propellants like propane, butane, or isobutane with conventional adjuvants. The materials of the present invention also may be administered in a non-pressurized form such as in a nebulizer or atomizer.

Depending upon the treatment being effected, the compounds of the present invention can be administered orally, topically, transdermally, parenterally, subcutaneously, intramuscularly, intraperitoneally, by intrana-
sal instillation, by intracavitary or intravesical instillation, intraocularly, intraarterially, intralesionally, or by application to mucous membranes, such as, that of the nose, throat, and bronchial tubes.

Compositions within the scope of this invention include all compositions wherein the compound of the present invention is contained in an amount effective to achieve its intended purpose. While individual needs vary, determination of optimal ranges of effective amounts of each component is within the skill of the art.

One approach for delivering an effector protein into cells involves the use of liposomes. Basically, this involves providing a liposome which includes that effector protein to be delivered, and then contacting the target cell with the liposome under conditions effective for delivery of the effector protein into the cell.

Liposomes are vesicles comprised of one or more concentrically ordered lipid bilayers which encapsulate an aqueous phase. They are normally not leaky, but can become leaky if a hole or pore occurs in the membrane, if the membrane is dissolved or degrades, or if the membrane temperature is increased to the phase transition temperature. Current methods of drug delivery via liposomes require that the liposome carrier ultimately become permeable and release the encapsulated drug at the target site. This can be accomplished, for example, in a passive manner wherein the liposome bilayer degrades over time through the action of various agents in the body. Every liposome composition will have a characteristic half-life in the circulation or at other sites in the body and, thus, by controlling the half-life of the liposome composition, the rate at which the bilayer degrades can be somewhat regulated.

In contrast to passive drug release, active drug release involves using an agent to induce a permeability change in the liposome vesicle. Liposome membranes can be constructed so that they become destabilized when the environment becomes acidic near the liposome membrane (see, e.g., Proc. Natl. Acad. Sci. USA 84:7851 (1987); Biochemistry 28:908 (1989), which are hereby incorporated by reference). When liposomes are endocytosed by a target cell, for example, they can be routed to acidic endosomes which will destabilize the liposome and result in drug release.

Alternatively, the liposome membrane can be chemically modified such that an enzyme is placed as a coating on the membrane which slowly destabilizes the liposome. Since control of drug release depends on the concentration of enzyme initially placed in the membrane, there is no real effective way to modulate or alter drug release to achieve “on demand” drug delivery. The same problem exists for pH-sensitive liposomes in that as soon as the liposome vesicle comes into contact with a target cell, it will be engulfed and a drop in pH will lead to drug release.

This liposome delivery system can also be made to accumulate at a target organ, tissue, or cell via active targeting (e.g., by incorporating an antibody or hormone on the surface of the liposomal vehicle). This can be achieved according to known methods.

Different types of liposomes can be prepared according to Bangham et al., (1965); U.S. Pat. No. 5,653,996 to Hsu et al.; U.S. Pat. No. 5,643,599 to Lee et al.; U.S. Pat. No. 5,885,613 to Holland et al.; U.S. Pat. No. 5,631,237 to Dzau et al.; and U.S. Pat. No. 5,059,421 to Loughrey et al.

An alternative approach for delivery of effector proteins involves the conjugation of the desired effector protein to a polymer that is stabilized to avoid enzymatic degradation of the conjugated effector protein. Conjugated proteins or polypeptides of this type are described in U.S. Pat. No. 5,681,811 to Ekwaruwe.

Yet another approach for delivery of proteins or polypeptides involves preparation of chimeric proteins according to U.S. Pat. No. 5,817,789 to Heartlein et al. The chimeric protein can include a ligand domain and, e.g., an effector protein of the present invention. The ligand domain is specific for receptors located on a target cell. Thus, when the chimeric protein is delivered intravenously or otherwise introduced into blood or lymph, the chimeric protein will adsorb to the targeted cell, and the targeted cell will internalize the chimeric protein, which allows the effector protein to de-stabilize the cell checkpoint control mechanism, affording its cytotoxic effects.

When it is desirable to achieve heterologous expression of an effector protein of the present invention in a target cell, DNA molecules encoding the desired effector protein can be delivered into the cell. Basically, this includes providing a nucleic acid molecule encoding the effector protein and then introducing the nucleic acid molecule into the cell under conditions effective to express the effector protein in the cell. Preferably, this is achieved by inserting the nucleic acid molecule into an expression vector before it is introduced into the cell.

When transforming mammalian cells for heterologous expression of an effector protein, an adenovirus vector can be employed. Adenovirus gene delivery vehicles can be readily prepared and utilized given the disclosure provided in Berkner, 1988, and Rosenfeld et al., 1991. Adenovirus-associated viral gene delivery vehicles can be constructed and used to deliver a gene to cells. The use of adenovirus-associated viral gene delivery vehicles in vitro is described in Chatterjee et al. 1992; Walsh et al. 1992; Walsh et al., 1994; Flotte et al., 1993a; Ponnazhagan et al., 1994; Miller et al., 1994; Einerhand et al., 1995; Luo et al., 1995; and Zhou et al., 1996. In vivo use of these vehicles is described in Flotte et al., 1993b and Kaplitt et al., 1994. Additional types of adenovirus vectors are described in U.S. Pat. No. 6,057,155 to Wickham et al.; U.S. Pat. No. 6,033,908 to Bout et al.; U.S. Pat. No. 6,001,577 to Wilson et al.; U.S. Pat. No. 5,994,132 to Chamberlain et al.; U.S. Pat. No. 5,801,225 to Kochanek et al.; U.S. Pat. No. 5,885,808 to Sporn et al.; and U.S. Pat. No. 5,871,727 to Curiel.

Retroviral vectors which have been modified to form infective transformation systems can also be used to deliver nucleic acid encoding a desired effector protein into a target cell. One such type of retroviral vector is disclosed in U.S. Pat. No. 5,849,586 to Kriegler et al.

Regardless of the type of infective transformation system employed, they should be targeted for delivery of the nucleic acid to a specific cell type. For example, for delivery of the nucleic acid into tumor cells, a high titer of the infective transformation system can be injected directly within the tumor site so as to enhance the likelihood of tumor cell infection. The infected cells will then express the desired effector protein, e.g., HopPhoA, HopPsA, or HopPhoA2, disrupting cellular functions and producing cytotoxic effects.

Particularly preferred is use of the effector proteins of the present invention to treat a cancerous condition (i.e., the eukaryotic cell which is affected is a cancer cell). This can be carried out by introducing a cytotoxic Pseudomonas protein into cancer cells of a patient under conditions effective to inhibit cancer cell division, thereby treating the cancerous condition.

By introducing, it is intended that the effector protein is administered to the patient, preferably in the form of a composition which will target delivery to the cancer cells. 
Alternatively, when using DNA-based therapies, it is intended that the introducing be carried out by administering a target DNA delivery system to the patient such that the cancer cells are targeted and the effector protein is expressed therein.

EXAMPLES

The following Examples are intended to be illustrative and in no way are intended to limit the scope of the present invention.

Materials and Methods

Bacterial Strains, Culture Conditions, Plasmids, and DNA Manipulation Techniques

Three experimentally amenable strains that represent different levels of diversity in P. syringae were investigated: Psy 61, Psy B728a, and Pto DC3000. (i) Psy 61 is a weak pathogen of bean whose hrp gene cluster, cloned on cosmid pHIR11, contains all of the genes necessary for nonpathogenic bacteria like Pseudomonas fluorescens and Escherichia coli to elicit the HR in tobacco and to secrete in culture the HrpZ harpin, a protein with unknown function that is secreted abundantly by the Hrp system (Alfano et al., 1996). The pHIR11 hip cluster has been completely sequenced (FIG. 1) (Alfano and Collmer, 1997), and the hopPsyA gene in the hypervariable region at the left edge of the strain was shown to encode a protein that has an Avr phenotype, travels the Hrp pathway, and elicits cell death when expressed in tobacco cells (Alfano and Collmer, 1997; Alfano et al., 1997; van Dijk et al., 1999). (ii) Psy B728a is in the same pathovar as strain 61 but is highly virulent and is a model for studying the role of the Hrp system in epiphytic fitness and pathogenicity (brown spot of bean) in the field (HiranO et al., 1999). (iii) Pto DC3000 is a well-studied pathogen of Arabidopsis and tomato (causing bacterial speck) that is highly divergent from pathovar syringae strains. Analysis of rRNA operon RFLP patterns has indicated that Pto and Psy are distantly related and could be considered separate species (Manceau and Horvais, 1997). Thus, we were able to compare two strains in the same pathovar with a strain from a highly divergent pathovar.

Conditions for culturing E. coli and P. syringae strains have been described (van Dijk et al., 1999), as have the sources for Psy 61 (Preston et al., 1995), Psy B728a (Hirano et al., 1999), and Pto DC3000 (Preston et al., 1995). Cloning and DNA manipulations were done in E. coli DH5α using pBluescript II (Stratagene, La Jolla, Calif.), pRK415 (Kean et al., 1988), and cosmid pCPP47 (Bauer and Collmer, 1997), according to standard procedures (Aussel et al., 1994). Cosmid libraries of Pto DC3000 and Psy B728a genomic DNA were previously constructed (Charkowski et al., 1998). Oligonucleotide synthesis and DNA sequencing were performed at the Cornell Biotechnology Center. The nucleotide sequence of the Pto DC3000 hrphrc cluster was determined using subclones of pCPP2473, a cosmid selected from a genomic cosmid library based on hybridization with the hrpK gene of Psy 61. The nucleotide sequence of the Psy B728a hrphrc cluster was determined using subclones of pCPP3017. These cosmids were selected from a genomic library based on hybridization with the hrpC operon of 61. The left side of the Psy 61 EEL region was cloned by PCR into pBSKSII+ Xhol and EcoRI sites using the following primers:

SEQ. ID. NO. 71, which primes within queA and contains an Xhol site:

SEQ. ID. NO. 72, which primes within hopPsyA and contains an EcoRI site:

Pfu polymerase was used for all PCR experiments. DNA sequence data were managed and analyzed with the DNAStar Program (Madison, Wis.), and databases were searched with the BLASTX, BLASTP, and BLASTN programs (Altschul et al., 1997).

Mutant Construction and Analysis

Large deletions in the Pto DC3000 Hrp Pshl were constructed by subcloning border fragments into restriction sites on either side of an ωsp6 cassette in pRK415, electroplating the recombinant plasmids into DC3000, and then selecting and screening for marker exchange mutants as described (Alfano et al., 1996). The following left and right side (FIGS. 2 and 3) deletion border fragments were used (with residual gene fragments indicated): for CUCPB5110 left tgt-gua-rrnaL-ORF4 (27 bp of ORF4) and right ORF1-hrpK (396 bp of ORF1); and for CUCPB5115 left hps-avrE (2569 bp of avrE) and right ORF6 (156 bp upstream of ORF6 start codon). The later fragment was PCR-amplified using the following primers:

SEQ. ID. NO. 73, which primes in the ORF5-ORF6 intergenic region and contains an Xhol site:

SEQ. ID. NO. 74, which primes in ORF6 and contains a HindIII site:

Mutant constructions were confirmed by Southern hybridizations using previously described conditions (Charkowski et al., 1998). The ability of mutants to secrete AvrPto was determined with anti-AvrPto antibodies and immunoblot analysis of cell fractions as previously described (van Dijk et al., 1999). Mutant CUCPB5 115 was complemented with pCPP3016, which carries ORF2 through ORF10 in cosmid pCPP47, and was introduced from E. coli DH5α by triparental mating using helper strain E. coli DH5α (pRK600), as described (Charkowski et al., 1998).

T7 Expression Analysis

Protein products of the Pto DC3000 EEL were analyzed by T7 polymerase-dependent expression using vector pET21 and E. coli BL21(DE3) as previously described (Huang et al., 1995). The following primer sets were used to PCR each ORF from pCPP3091, which carries in pBSKSII+ a BamHI fragment containing tgt to hrcV:

ORF1, SEQ. ID. Nos. 75 and 76, respectively:

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agtaggatcc tgaaatgtag gggcoggg 28
agttaaagctt ctatgctgct ttcaogta 28
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ORF2, SEQ. ID. Nos. 77 and 78, respectively:

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agtaggatcc tcttcgaagga atgagaaga 28
agttaaagctt cgtgaagatg catttcgc 28
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ORF3, SEQ. ID. Nos. 79 and 80, respectively:

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agtaggatcc tgcgtcactga tgcagaagta 28
agtacgag caaacaata acacggta 28
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With appropriate antibiotics, suspended in 5 molar boron, comminuting the tissue in 0.5 ml of 5 molar MES pH 5.6, and then infiltrated with a needleless syringe into the leaves of test plants at 50 μg/ml for pathogenicity assays (Charkowski et al., 1998). The difference between Psy strains 61 and B728a in this region was particularly surprising. This region of the P. syringae Hrp Pai was given the EEL designation because it contained completely different effector protein genes (Table 1 below), which appear to be exchanged at this locus at a high frequency. In this regard, it is noteworthy that (i) ORF2 in the B728a EEL is a homolog of avrPphE, which is in a different location, immediately downstream of hrpK (hrpY), in PpH 1302A (Mansfield et al., 1994), (ii) hopPsyA (hrmA) is present in only a few Psy strains (Heu and Hutcheson, 1993; Alano et al., 1997), (iii) ORF5 and ORF1 in the B728a EEL predict a protein that is similar to Xanthomonas AvrBsT and possesses multiple motifs characteristic of the AvrRsv family (Ciesiolka et al., 1999). G+C content different from the genomic average is a hallmark of horizontally transferred genes, and the G+C contents of the ORFs in the three EELs are considerably lower than the average of 59-61% for P. syringae (Palleroni et al., 1984) (Table 1 below). They are also lower than hrpK (60%) and queA (63-64%). The ORFs in the Pto DC3000 EEL predict no products with similarity to known effector proteins, however T7 polymerase-dependent expression revealed products in the size range predicted for ORF 1, ORF3, and ORF4. Furthermore, the ORF1 protein is sequestered in a hrp-dependent manner by E. coli (CPP2156), which expresses an Erwinia chrysanthemi Hrp system that secretes P. syringae Avr proteins (Ham et al., 1998). Several ORFs in these EELs are preceded by Hrp boxes indicative of HrpL-activated promoters (FIG. 1) (Xiao and Hutcheson, 1994), and the lack of intervening Rho-independent terminator sequences or promoters suggests that ORF1 in DC3000 and ORF1 and ORF2 in B728a are expressed from HrpL-activated promoters upstream of the respective hrpK genes. The EELs of these three strains also contain sequences homologous to insertion sequences, transposases, phage integrase genes, and plasmids (FIG. 2 and Table 1 below). The Psy B728a ORF5 and ORF6 operon is bordered on the left side linked to any type III secretion system genes or other genes in the Hrp Pai (FIG. 2). Thus, this is the apparent point of insertion of the Hrp Pai in the ancestral Pseudomonas genome.

Example 3
Identification of a Conserved Effector Locus (CEL) Located on the Right Side of the Hrp Pae in Psy B728a and Pto DC3000
Previous studies of the region to the right of hrpR in DC3000 had revealed the existence of the avrE locus, which...
is comprised of two transcriptional units (Lorang and Keen, 1995), the S sequences for the first 4 transcriptional units beyond hrpR (Lorang and Keen, 1995), and the identity of the fourth transcriptional unit as the hrpW gene encoding a second harpin (Charkowski et al., 1998). The DNA sequence of the first 14 ORFs to the right of hrpR in Pto DC3000 was completed in this investigation and the corresponding region in Psy B728a was partially sequenced (Fig. 3). Like the EEL, this region contains putative effector genes, e.g., avrE (Lorang and Keen, 1995). Unlike the EEL, the ORFs in this region have an average G+C content of 58.0%, which is close to that of the hrp/hrc genes, the region contains no sequences similar to known mobile genetic elements, and it appears conserved between Psy and Pto (Fig. 3). Comparison of the regions sequenced in B728a and DC3000 revealed that the first 7 ORFs are arranged identically and have an average DNA sequence identity of 78%. Hence, this region was given the CEL designation.

The precise border of the CEL remains undefined, and no sequences that were repeated in the EEL border of the Hrp Pai were found. ORF7 and ORFS are likely to be part of the CEL, based on the presence of an upstream Hrp box (Fig. 3). However, the region beyond ORF10 probably is not in the CEL because the product of the next ORF shows homology to a family of bacterial GalA proteins (e.g., 28% identity with E. coli GalA over 204 amino acids; E=1e-8) (Blattner et al., 1995), and glutathione-S-transferase activity is common in nonpathogenic fluorescent pseudomonads (Zablotowicz et al., 1995). The presence of a galP homolog (38% identity over 256 amino acids, based on incomplete sequence, to E. coli GalP; E=2e-42) (Blattner et al., 1997) in this region further suggests that it is beyond the CEL.

Several other features of this region in B728a and DC3000 are noteworthy. (i) Both strains have a 1 kb intergenic region between hrpR and ORF1 that is distinguished by low sequence identity (44%) but which contains three inverted repeats that could form stem loop structures affecting expression of the hrpRS operon. (ii) ORF1 is most similar to E. coli murein lytic transglycosylase Mid (38% identity over 324 amino acids; E=4e-56). (iii) ORF2 is 42% identical over 130 amino acids with E. amylovora DspF (E=9e-24), a candidate chaperone (Bogdanove et al., 1998a; Gaudriault et al., 1997). (iv) The ORF5 protein is secreted in a hrp-dependent manner by E. coli(pCPP2156), but mutation with an OSp’ cassette has little effect on either HR elicitation in tobacco or pathogenicity in tomato (Charkowski, unpublished). (v) Finally, six operons in this region are preceded by Hrp boxes (Lorang and Keen, 1995) (Fig. 3), which is characteristic of known avr genes in P. syringae (Alfano et al., 1996). Thus, the CEL carries multiple candidate effectors.

Example 4
Investigation of EEL and CEL Roles in Pathogenicity

A mutation was constructed in DC3000 that replaced all of the ORFs between hrpK and tRNA^{A2} (EEL) with an OSp’ cassette (Fig. 2). This Pto mutant, CUCPB5110, was tested for its ability to elicit the HR in tobacco and to cause disease in tomato. The mutant retained the ability to elicit the HR and to produce disease symptoms, but it failed to reach population levels as high as the parental strain in tomato (Fig. 4A).

A mutation was constructed in DC3000 that replaced avrE through ORF5 (CEL) with an OSp’ cassette. This deleted all of the CEL ORFs that were both partially characterized and likely to encode effectors. This Pto mutant, CUCPB5115, still elicited the HR in tobacco, but tissue collapse was delayed ca. 5 h (Fig. 4C). The mutant no longer elicited disease symptoms in tomato when infiltrated at a concentration of 10^6 cfu/ml, and growth in planta was strongly reduced (Fig. 4B). However, the mutant elicited an HR dependent on the tomato Pto R gene that was indistinguishable from the wild-type in tests involving PtoS (susceptible) and PtoR (resistant) Rio Grande tomato lines. Plasmid pCPP3016, which carries ORF2 through ORF10, fully restored the ability of CUCPB5115 to cause disease symptoms and partially restored the ability of the mutant to multiply in tomato leaves (Figs. 4B and 4E). Deletion of the hrp/hrc cluster abolishes HR and pathogenicity phenotypes in Pto DC3000 (Collmer et al., 2000). To confirm that the large deletions in Pto mutants CUCPB5110 and CUCPB5115 did not disrupt Hrp secretion functions, we compared the ability of these mutants, the DC3000 hrp/hrc deletion mutant, and wild-type DC3000 to make and secrete AvrPto in culture while retaining a cytoplasmic marker comprised of β-lactamase lacking its signal peptide. AvrPto provided an ideal subject for this test because it is a well-studied effector protein that is secreted in culture and injected into host cells in planta (Alfano and Collmer, 1997; van Dijk et al., 1999). Only the hrp/hrc deletion cluster mutant was impaired in AvrPto production and secretion (Fig. 5).

Based on the above studies, the P. syringae hrp/hrc genes are part of a Hrp Pai that has three distinct loci: an EEL, the hrp/hrc gene cluster, and a CEL. The EEL harbors exchangeable effector genes and makes only a quantitative contribution to parasitic fitness in host plants. The hrp/hrc locus encodes the Hrp secretion system and is required for effector protein delivery, parasitism, and pathogenicity. The CEL makes no discernible contribution to Hrp secretion functions but contributes strongly to parasitic fitness and is required for Pto pathogenicity in tomato. The Hrp Pai of P. syringae has several properties of Pals possessed by animal pathogens (Hacker et al., 1997), including the presence of many virulence-associated genes (several with relatively low G+C content) in a large (ca. 50-kb) chromosomal region linked to a tRNA locus and absent from the corresponding locus in a closely related species. In addition, the EEL portion of the Hrp Pai is unstable and contains many sequences related to mobile genetic elements.

The EEL is a novel feature of known Pals, which is likely involved in fine-tuning the parasitic fitness of P. syringae strains with various plant hosts. By comparing closely- and distantly-related strains of P. syringae, we were able to establish the high instability of this locus and the contrasting high conservation of its border sequences. No single mechanism can explain the high instability, as we found fragments related to phages, insertion sequences, and plasmids in the Pso and Pto EELs, and insertion sequences were recently reported in the corresponding region of three other P. syringae strains (Inoue and Takikawa, 1999). The mechanism or significance of the localization of the EELs between tRNA^{A2} and hrpK sequences in the Hrp Pals also is unclear. Pto DC3000 carries at least one other effector gene, avrPto, that is located elsewhere in the genome (Ronald et al., 1992), many P. syringae avr genes are located on plasmids (Leach and White, 1996), and the EEL ORFs represent a mix of widespread, (e.g., avrRxv family) and seemingly rare (e.g., hopPsyA), effector genes. The G+C content of the EEL ORFs is significantly lower than that of the rest of the Hrp Pai and the P. syringae genome. Although certain genes in the non-EEL portions of the Hrp Pai, such as hrpA, are highly divergent, they have a high G+C content, and there is no evidence that they have been horizontally transferred separately from the rest of the Hrp Pai. The relatively low
G+C content of the ORFs in the EELs (and of other \textit{P. syringae} avr genes) suggests that these genes may be horizontally acquired from a wider pool of pathogenic bacteria than just \textit{P. syringae} (Kim et al., 1998). Indeed, the \textit{avrRxs} family of genes is found in a wide range of plant and animal pathogens (Ciesiolka et al., 1999). The weak effect on parasitic fitness of deleting the \textit{Pto} DC3000 EEL, or of mutating \textit{hopPsyA} (hpmA) in \textit{Psy} 61 (Huang et al., 1991), is typical of mutations in individual \textit{avr} genes and presumably results from redundancy in the effector protein system (Leach and White, 1996).

The functions of \textit{hrpK} and of the CEL ORF1 are unclear but warrant discussion. These two ORFs reside just outside the \textit{hrp}L and \textit{hrpR} delimited cluster of operons containing both \textit{hrp} and \textit{hrc} genes and thereby spatially separate the three regions of the \textit{Hrp Pai} (FIGS. 1–3). \textit{hrpK} mutants have a variable \textit{Hrp} phenotype (Mansfield et al., 1994; Bozso et al., 1999), and a \textit{Psy} B728a \textit{hrpK} mutant still secretes \textit{HrpZ} (Alfano, unpublished), which suggests that \textit{HrpK} may be an effector protein. Nevertheless, the \textit{HrpK} proteins of \textit{Psy} 61 and \textit{Pto} DC3000 are 79% identical and therefore are more conserved than many \textit{Hrp} secretion system components. It is also noteworthy that \textit{hrpK} appears to be in an operon with other effector genes in \textit{Psy} B728a and \textit{Pto} DC3000. In contrast, the CEL ORF1 may contribute (weakly or redundantly) to \textit{Hrp} secretion functions by promoting penetration of the system through the bacterial peptidoglycan layer. The ORF1 product has extensive homology with \textit{E. coli} \textit{MifD} and shares a lysosome-like domain with the product of \textit{ipgF} (Mushgian et al., 1996), a \textit{Shigella flexneri} gene that is also located between loci encoding a type III secretion system and effector proteins (Allaoui et al., 1993). Mutations in these genes in \textit{Pto} and \textit{S. flexneri} have no obvious phenotype (Lorang and Keen, 1995; Allaoui et al., 1993), as is typical for genes encoding peptidoglycan hydrolases (Dijkshoorn and Keck, 1996).

The loss of pathogenicity in \textit{Pto} mutant CUCPB5115, with an \textit{avrE}-ORF5 deletion in the CEL, was surprising because pathogenicity is retained in DC3000 mutants in which the corresponding operons are individually disrupted (Lorang and Keen, 1995; Charkowski et al., 1998). In assessing the possible function of this region and the conservation of its constituent genes, it should be noted that \textit{avrE} is unlike other \textit{avr} genes found in \textit{Pto} in that it confers avirulence to \textit{P. syringae pv. glycinea} on all tested soybean cultivars and it has a homolog (\textit{dspe}) in \textit{E. amylovora} that is required for pathogenicity (Lorang and Keen, 1995; Bogdanove et al., 1998b). Although the CEL is required for pathogenicity, it is not essential for type III effector protein secretion because the mutant still secretes \textit{AvrPto}. It also appears to play no essential role in type III translocation of effector proteins into plant cells because the mutant still elicits the \textit{HR} in nonhost tobacco and in a \textit{PtoR}-resistance tomato line, and \textit{pHrR1}, which lacks this region, appears capable of translocating several \textit{Avr} proteins (Gopalan et al., 1996; Pirhonen et al., 1996). The conservation of this region in the divergent pathovars \textit{Psy} and \textit{Pto}, and its importance in disease, suggests that the products of the CEL may be redundantly involved in a common, essential aspect of pathogenesis.

The similar G+C content and codon usage of the \textit{hrp}/\textit{hrc} genes, the genes in the CEL, and total \textit{P. syringae} genomic DNA suggests that the \textit{Hrp Pai} was acquired early in the evolution of \textit{P. syringae}. Although, the EEL region may have similarly developed early in the radiation of \textit{P. syringae} into its many pathovars, races, and strains, the apparent instability that is discussed above suggests ongoing rapid evolution at this locus. Indeed, many \textit{P. syringae} avr genes are associated with mobile genetic elements, regardless of their location (Kim et al., 1998). Thus, it appears that \textit{Hrp}-mediated pathogenicity in \textit{P. syringae} is collectively dependent on a set of genes that are universal among divergent pathovars and on another set that varies among strains even in the same pathovar. The latter are presumably acquired and lost in response to opposing selection pressures to promote parasitism while evading host R-gene surveillance systems.

Example 5
Role of \textit{ShcA} as a Type III Chaperone for the \textit{HopPsyA} Effector

The \textit{ORF} upstream of \textit{hopPsyA}, tentatively named \textit{shcA}, encodes a protein product of the predicted molecular mass. The \textit{ORF} upstream of the \textit{hopPsyA} gene in \textit{P. s. syringae} 61 (originally designated \textit{ORF1}) shares sequence identity with \textit{exsC} and \textit{ORF7}, which are genes adjacent to type III effector genes in \textit{P. aeruginosa} and \textit{Yersinia pestis}, respectively (Frank and Iglewski, 1991; Perry et al., 1998). Although neither of these \textit{ORFs} have been shown experimentally to encode chaperones, they have been noted to share properties that type III chaperones often possess (Cornellis et al., 1998). One of these properties is the location of the chaperone gene itself (FIGS. 1 and 6). Chaperone genes are often adjacent to a gene that encodes the effector protein with which the chaperone interacts. Furthermore, \textit{shcA} also shares other common characteristics of type III chaperones: its protein product is relatively small (about 14 kDa), it has an acidic \textit{pI}, and it has a C-terminal region that is predicted to be an amphipathic \textit{a}-helix. To begin assessing the function of \textit{shcA}, it was first determined whether \textit{shcA} encodes a protein product. A construct was prepared using PCR that fused \textit{shcA} in-frame to a sequence encoding the \textit{FLAG} epitope. This construct, \textit{pLV26}, contains the nucleotide sequence upstream of \textit{shcA}, including a putative ribosome binding site (\textit{RBS}). \textit{DH5}α\textit{F}°\textit{pLV26} cultures were grown in rich media and induced at the appropriate density with IPTG. Whole cell lysates were separated by SDS-PAGE and analyzed with immunoblots using anti-\textit{FLAG} antibodies. By comparing the \textit{ShcA}-\textit{FLAG} encoded by \textit{pLV26} to a construct that made \textit{ShcA}-\textit{FLAG} from a vector \textit{RBS}, it was concluded that the native \textit{RBS} upstream of \textit{shcA} was competent for translation (FIG. 7). Thus, the \textit{shcA} ORF is a legitimate gene that encodes a protein product.

To test the effects of \textit{shcA} on bacterial-plant interactions, an \textit{shcA} mutation was constructed in the minimalist \textit{hrp}/\textit{hrc} cluster carried on \textit{cosmid pHrR11}. There are distinct advantages to having the \textit{shcA} mutation marker-exchanged into \textit{pHrR11}. The main one is that the \textit{HR} assay can be used as a screen to determine if \textit{HopPsyA} is being translocated into plant cells because the \textit{pHrR11}-dependent \textit{HR} requires the delivery of \textit{HopPsyA} into plant cells (Alfano et al., 1996; Alfano et al., 1997). With the chromosomal \textit{shcA} mutant, other \textit{Hop} proteins would probably be delivered to the interior of plant cells. Some of these proteins would be recognized by the \textit{R} gene-based plant surveillance system and initiate an \textit{HR} masking any defect in \textit{HopPsyA} delivery. \textit{E. coli} \textit{MC}4100 carrying \textit{pLV10}, a \textit{pHrR11} derivative, which contains a nonpolar \textit{nptII} cartridge within \textit{shcA}, was unable to elicit an \textit{HR} on tobacco (FIG. 8). This indicates that \textit{shcA} is required for the translocation of \textit{HopPsyA} into plant cells. To determine if \textit{HopPsyA} was secreted in culture, cultures of the nonpathogen \textit{P. fluorescens} 55 were grown. This bacterium carried either \textit{pHrR11}, \textit{pCPP2089} (a \textit{pHrR11} derivative defective in type III secretion), or \textit{pLV10}. The representative results can be seen in FIG. 8. \textit{shcA} was required for the in-culture type III secretion of the \textit{HopPsyA}}
of HopPsyA secretion in culture directly via the native Hrp system carried in *P. s. syringae* 61 was tested. *P. s. syringae* 61 cultures grown in hrp-depressing fructose minimal medium at 22°C, were subjected to centrifugation. Proteins present in the supernatant fractions were concentrated by TCA precipitation, and the cell-bound and supernatant samples were resolved with SDS-PAGE and analyzed with immunoblots using anti-HopPsyA antibodies. A HopPsyA signal was detected in supernatant fractions from wild type *P. s. syringae* 61 (FIG. 14). Importantly, HopPsyA was not detected in supernatant fractions from *P. s. syringae* 61-2089, which is defective in Hrp secretion, indicating that the HopPsyA signal in the supernatant was due specifically to type III protein secretion (FIG. 14). As a second control, both strains contained pCPP2318, which encodes the mature β-lactamase lacking its N-terminal signal peptide, and provides a marker for cell lysis. β-lactamase was detected only in the cell-bound fractions of these samples, clearly showing that cell lysis did not occur at a significant level (FIG. 14). The fact that HopPsyA is secreted via the type III secretion system in culture and that the avirulence activity of HopPsyA occurs only when it is expressed in plant cells strongly support that HopPsyA is delivered into plant cells via the type III pathway.

HopPsyA contributes in a detectable, albeit minor, way to growth of *P. s. syringae* 61 in bean. The effect of a HopPsyA mutation on the multiplication of *P. s. syringae* 61 in bean tissue has been reported (Huang et al., 1991). These data essentially indicate that HopPsyA contributes little to the ability of *P. s. syringae* 61 to multiply in bean. The *P. s. syringae* 61 hopPsyA mutant does not grow as well in bean leaves as the wild-type strain (FIG. 15). This was unexpected, because these results are in direct conflict with previously reported data. One rationale for the discrepancy is that the previous reports focused primarily on the major phenotype that a hrp mutant exhibits on in planta growth and predicated the discovery that HopPsyA was a type III-secreted protein. Thus, it is quite possible that the earlier experiments missed the more subtle effect that HopPsyA appears to have on the multiplication of *P. s. syringae* 61 in bean tissue (Huang et al., 1991). The data presented here supports that HopPsyA contributes to the pathogenicity of *P. s. syringae* and are consistent with the hypothesis that the majority of Hops from *P. s. syringae* contribute subtly to pathogenicity. The lack of strong pathogenicity phenotypes for mutants defective in different *avr* and hop genes may be due to possible *avr/hop* gene redundancy or a decreased dependence on any one Hop protein through coevolution with the plant. Indeed, the type III-delivered proteins of plant pathogens that are delivered into plant cells may not be virulence proteins per se, but rather they may suppress responses of the plant that are important for pathogenicity to proceed (Jakobek et al., 1993). These responses may be defense responses or other more general processes that maintain the status quo within the plant (e.g., the cell cycle).

**Example 7**

Molecular Interactions of HopPsyA

HopPsyA interacts with the *Arabidopsis* MAD2 protein in the yeast 2-hybrid system. To determine a pathogenic target for HopPsyA, the yeast 2-hybrid system was used with cDNA libraries made from *Arabidopsis* (Fields and Song, 1989; Finley and Brent, 1994). In the yeast 2-hybrid system, a fusion between the protein of interest (the "bait") and the LexA DNA-binding domain was transformed into a yeast tester strain. A cDNA expression library was constructed in a vector that creates fusions to a transcriptional activator
domain. This library was transformed into the tester strain en masse, and clones encoding partners for the “bait” are selected via their ability to bring the transcriptional activator domain into proximity with the DNA binding domain, thus initiating transcription of the LEU2 selectable marker gene. A second round screening of candidates, that activate the LEU2 marker, relies on their ability to also activate a lacZ reporter gene. Bait constructs were initially made with hopPsyA in the yeast vector pEG202 that corresponded to a full-length HopPsyA-LexA fusion, the carboxy-terminal half of HopPsyA fused to LexA, and the amino-terminal half of HopPsyA fused to LexA, and named these constructs pLV23, pLV24, and pLV25, respectively. However, pLV23 was lethal to yeast and pLV25 activated the lacZ reporter gene in relatively high amounts on its own (i.e., without the activation domain present). Thus, both pLV23 and pLV25 were not used to screen for protein interactors via the yeast 2-hybrid system. pLV24, which contains the 3’ portion of hopPsyA fused to LexA, proved to be an appropriate construct to use for bait in the yeast 2-hybrid system, because it did not autoactivate the lacZ reporter gene and, based on van der Schouven et al., 1999, the HopPsyA-LexA fusion produced by pLV24 appeared to localize to the nucleus. In addition, it was confirmed that pLV24 made a protein of the appropriate size that corresponds to HopPsyA by performing immunoblots with anti-HopPsyA antibodies on yeast cultures carrying this vector.

Initial screens with pLV24 and Arabidopsis cDNA libraries in the yeast 2-hybrid vector pLG4-5. From three independent screens, several hundred by sequences similar to those in a Pph plasmid that carries several avr genes (Jackson et al., 1999) and by a sequence homologous to insertion elements that are typically found on plasmids, suggesting plasmid integration via an IS element in this region (Szabo and Mills, 1984). Psy B728a ORF3 and ORF4 show similarity to sequences implicated in the horizontal acquisition of the LEE Pai by pathogenic E. coli strains (Perna et al., 1998). These Psy B728a ORFs are not preceded by Hrp boxes and are unlikely to encode effector proteins.

### Table 1: ORFs and fragments of genetic elements in the EELs of P. syringae DC3000, Psy B728a, and Psy 61 and similarities with known avr genes and mobile genetic elements.

<table>
<thead>
<tr>
<th>ORF or sequence</th>
<th>% G+C</th>
<th>Size</th>
<th>BLAST E value with representative similar sequence(s) in database, or relevant feature</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Psy B728a</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ORF1</td>
<td>51</td>
<td>328 aa</td>
<td>9e-40 Pph AvrPsyC (Yucel et al., 1994)</td>
</tr>
<tr>
<td>ORF2</td>
<td>52</td>
<td>328 aa</td>
<td>1e-154 Pph AvrPsyE (Mansfield et al., 1994)</td>
</tr>
<tr>
<td>ORF3</td>
<td>55</td>
<td>507 aa</td>
<td>2e-63 E. coli L0015 (Perna et al., 1998)</td>
</tr>
<tr>
<td>ORF4</td>
<td>55</td>
<td>118 aa</td>
<td>9e-9 E. coli L0014 (Perna et al., 1998)</td>
</tr>
<tr>
<td>ORF5</td>
<td>49</td>
<td>441 aa</td>
<td>1e-4 Ncv AvrBSt (Ciesiolka et al., 1999)</td>
</tr>
<tr>
<td>ORF6</td>
<td>52</td>
<td>120 aa</td>
<td>None</td>
</tr>
<tr>
<td>IntA</td>
<td>59</td>
<td>49 aa</td>
<td>3e-5 E. coli CP4-like integrase (Perna et al., 1998)</td>
</tr>
<tr>
<td><strong>Psy 61</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HopPsyA</td>
<td>53</td>
<td>375 aa</td>
<td>Hrp-secreted Avr (Alfano et al., 1997; van Dijk et al., 1999)</td>
</tr>
<tr>
<td>ShcA</td>
<td>57</td>
<td>112 aa</td>
<td>6e-4 Y008B (Perry et al., 1998)</td>
</tr>
</tbody>
</table>

### Table 2: Percent Amino Acid Sequence Identity Between Different Mad2 Homologs

<table>
<thead>
<tr>
<th>Mad2 Homolog</th>
<th>Arabidopsis</th>
<th>Corn</th>
<th>Human</th>
<th>Mouse</th>
<th>Frog</th>
<th>Yeast</th>
<th>Bolding Yeast</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arabidopsis</td>
<td>—</td>
<td>91.3</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Corn</td>
<td>—</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Not unexpectedly, the sequence of the *Arabidopsis* Mad2 protein is more closely related to the corn Mad2, the only plant Mad2 homolog represented in the databases.

The corn Mad2 is about 82% identical to the *Arabidopsis* Mad2. FIGS. 16A–B show yeast strains containing either pLV24 and pL+4-S, pLV202 and pLV116, or pLV24 and pLV116 on leucine drop-out plates and plates containing X-Gal, showing that only when both HopPsyA and Mad2 are present, β-galactosidase and LEU2 activity are induced. It is important to note that the cDNA library that yielded mad2 has been used for many different yeast 2-hybrid screens and advances (Elledge, 1996; Glotzer, 1996; Rudner and Murray, 1996). APC, which activates the APC to degrade inhibitors of the spindle checkpoint (Glotzer, 1996; Rudner and Murray, 1996). The overexpression of APC, which activates the APC to degrade inhibitors of the spindle checkpoint when overexpressed in a manner that is dependent on functional Bub1, Bub3, and Mad2 proteins (Hardwick and Murray, 1995). Another required protein in this checkpoint is Mps1, a protein kinase that activates the spindle checkpoint when overexpressed in a manner that is dependent on all of the Bub and Mad proteins, indicating that Mps1 acts very early in the spindle checkpoint (Hardwick et al., 1996).

Based on data from the different Mad2 homologs that have been studied, Mad2 appears to have a central role in the spindle checkpoint. Addition of Mad2 to *Xenopus* egg extracts results in inhibition of cyclin B degradation and mitotic arrest due to the inhibition of the ubiquitin ligase activity of the APC (Li et al., 1997). The overexpression of Mad2 from fission yeast causes mitotic arrest by activating the spindle checkpoint (He et al., 1997). Whereas, introducing anti-Mad2 antibodies into mammalian cell cultures causes early transition to anaphase in the absence of microtubule drugs, indicating that Mad2 is involved in the normal cell cycle. Several reports suggest that different Mad2 homologs directly interact with the APC (Li et al., 1997; Fang et al., 1998; Kallio et al., 1998). Another protein called Cdc20 in *S. cerevisiae* binds to the APC, is required for activation of the APC during certain cell cycles, and Mad2 binds to it (Hwang et al., 1998; Kim et al., 1998; Lorca et al., 1998; Wassmann and Benezra, 1998). The picture that is emerging from all of these exciting findings is that Mad2 acts as an inhibitor of the APC, probably by binding to Cdc20. When Mad2 is not present, the Cdc20 binds to the APC, which activates the APC to degrade inhibitors of the spindle checkpoint.
transition to anaphase. FIG. 12 shows a summary of the spindle checkpoint focusing on Mad2’s involvement and using the names of the spindle checkpoint proteins from *S. cerevisiae*.

The plant spindle checkpoint: A possible target of bacterial pathogens. Many of the cell cycle proteins from animals have homologs in plants (Mironov et al., 1999). In fact, one of the early clues that there existed a spindle checkpoint was first made in plants. The observation noted was that chromosomes that lagged behind in their attachment to the spindle caused a delay in the transition to anaphase (Bajer and Mole-Bajer, 1956). Moreover, mad2 has been recently isolated from corn and the Mad2 protein localization in plant cells undergoing mitosis is consistent with the localization of Mad2 in other systems (Yu et al., 1999). Based on a published meeting report, genes that encode components of the APC from *Arabidopsis* have been recently cloned (Inze et al., 1999). Thus, it appears that a functional spindle checkpoint probably is conserved in plants. The data presented above shows that the *P. syringae* HopPsyA protein interacts with the *Arabidopsis* Mad2 protein in the yeast 2-hybrid system.

It is possible that a pathogenic strategy of a bacterial plant pathogen is to alter the plant cell cycle. Duan et al. recently reported that pthA, a member of the avrBs3 family of avr genes from *X. citri*, is expressed in citrus and causes cell enlargement and cell division, which may implicate the plant cell cycle (Duan et al., 1999). If HopPsyA does target Mad2, at least two possible benefits to pathogenicity can be envisioned. Since plant cells in mature leaves are quiescent, one benefit of delivering HopPsyA into these cells may be that it may trigger cell division through its interaction with Mad2. This is consistent with the observation that anti-Mad2 antibodies cause an early onset of anaphase in mammalian cells (Gorbsky et al., 1998). More plant cells near the pathogen may increase the nutrients available in the apoplast. A second possible benefit may occur if HopPsyA is delivered into plant cells actively dividing in young leaves. Delivery of HopPsyA into plant cells of these leaves may derail the spindle checkpoint through its interaction with Mad2. These cells would be prone to more mistakes segregating their chromosomes; in some cells this would result in death and the cellular contents would ultimately leak into the apoplast providing nutrients for the pathogen.

Example 8

**Cytotoxic Effects of HopPtoA and HopPsyA Expressed in Yeast**

Both hopPtoA (SEQ. ID. No. 6) and hopPsyA (SEQ. ID. No. 35) were first cloned into pFLAG-CTC (Kodak) to generate an in-frame fusion with the FLAG epitope, which permitted monitoring of protein production with anti-FLAG monoclonal antibodies. The FLAG-tagged genes were then cloned under the control of the GALL promoter in the yeast shuttle vector p415GAL1 (Mumberg et al., 1994). These regulatable promoters of *Saccharomyces cerevisiae* allowed comparison of recombinant activity and heterologous expression. The recombinant plasmids were transformed into uracl auxotrophic yeast strains FY833/4, selecting for growth on SC-Ura (synthetic complete medium lacking uracil) based on the presence of the URA3 gene on the plasmid. The transformants were then streaked onto SC-Ura medium plates containing either 2% galactose (which would induce expression of HopPsyA and HopPtoA) or 2% glucose. No growth was observed on the plates supplemented with 2% galactose. This effect was observed with repeated testing and was not observed with empty vector controls, with four other effectors similarly cloned into p415GAL1, or when raffinose was used instead of galactose. FLAG-tagged nontoxic Avr proteins were used to confirm that the genes were differentially expressed, as expected, on plates containing galactose. Importantly, the toxic effect with HopPsyA was observed when the encoding gene was recloned into p415GAL1, which expresses foreign genes at a substantially lower level than p415GAL1.

References

Each of the references cited herein or otherwise listed below are expressly incorporated by reference in their entirety into this specification.


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Although the invention has been described in detail for the purposes of illustration, it is understood that such detail is solely for that purpose, and variations can be made therein by those skilled in the art without departing from the spirit and scope of the invention which is defined by the following claims.

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<223> OTHER INFORMATION: n at any position is undefined
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<211> LENGTH: 623
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae
<400> SEQUENCE: 3

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20 25 30
Ala Thr Ala Gly Met Asn Pro Pro Leu Thr Pro Asp Gln Ser Gly
35 40 45
Ser His Ala Thr Glu Ser Ser Ser Ala Gly Ala Arg Leu Aen Val
50 55 60
Ala Ala Arg His Thr Glu Leu Glu Ala Phe Lys Ala Glu His Gly
65 70 75 80
Thr Ala Pro Val Ser Gly Ala Pro Met Ile Ser Ser Arg Ala Ala Leu
85 90 95
Leu Ile Gly Ser Leu Leu Gln Ala Glu Pro Leu Pro Phe Gln Val Met
100 105 110
Ala Glu Lys Leu Ser Pro Glu Arg Tyr Glu Leu Gln Phe Gln Gly
115 120 125
Ser Asp Leu Gln Glu Arg Leu Gly Lys Ala Glu Pro Gly Glu Ile
130 135 140
Pro Asp Leu Ala Glu Val Gln Leu Ile Gys Gly Phe Ala Glu Ser
145 150 155 160
Val Ala Asp Glu Leu His Phe Glu Leu Met His Asp Ala Ser Pro
165 170 175
Ala Thr Val Gly Glu His Ala Lys Ala Asp Lys Ala Thr Leu Ala Val
180 185 190
Ser Gln Thr Ala Leu Gly Glu Tyr Ala Gly Arg Ala Ser Lys Ala Ile
195 200 205
Gly Glu Gly Leu Ser Asn Ser Ile Ala Ser Leu Asp Glu His Ile Ser
210 215 220
Ala Leu Asp Leu Thr Leu Gln Asp Ala Glu Gly Aen Lys Gly Ser
225 230 235 240
Leu His Ala Asp Arg Glu Ala Leu Val Asp Ala Lys Thr Thr Leu Val
245 250 255
Gly Leu His Ala Asp Phe Val Lys Ser Pro Glu Ala Lys Arg Leu Ala
Ser Val Ala Ala His Thr Gln Leu Asp Asn Val Ser Asp Leu Val 275 280 285
Thr Ala Arg Asn Thr Val Gly Trp Lys Gly Ala Gly Pro Ile Val 290 295 300
Ala Ala Val Pro Gln Phe Leu Ser Ser Met Thr His Leu Gly Tyr 305 310 315 320
Val Arg Leu Ser Thr Ser Asp Leu Arg Asp Thr Ile Pro Glu Thr 325 330 335
Ser Ser Asp Ala Asn Met Leu Lys Ala Ser Ile Ile Gly Met Val Ala 340 345 350
Gly Ile Ala His Glu Thr Val Asn Ser Val Val Lys Pro Met Phe Gln 355 360 365
Ala Ala Leu Gln Lys Thr Gly Leu Asn Glu Arg Leu Asn Met Val Pro 370 375 380
Met Lys Ala Val Asp Thr Aan Thr Val Ile Pro Asp Pro Phe Glu Leu 385 390 395 400
Lys Ser Glu His Gly Glu Leu Val Lys Thr Pro Glu Glu Val Ala 405 410 415
Gln Asp Lys Ala Phe Val Lys Ser Glu Arg Ala Leu Leu Asn Gln Lys 420 425 430
Lys Val Gln Gly Ser Ser Thr His Pro Val Gly Leu Met Ala Tyr 435 440 445
Ser Ala Phe Gly Gly Ser Glu Ala Ala Val Arg Glu Met Leu Asn Asp Val 450 455 460
His Gln Ile Asn Gly Glu Thr Leu Ser Ala Arg Ala Leu Ala Ser Gly 465 470 475 480
Phe Gly Gly Ala Val Ser Ala Ser Ser Gin Thr Leu Leu Gin Leu Lys 485 490 495
Ser Asn Tyr Val Asp Pro Gin Gly Arg Lys Ile Pro Val Phe Thr Pro 500 505 510
Asp Arg Ala Glu Ser Asp Leu Lys Asp Leu Leu Lys Gly Met Asp 515 520 525
Leu Arg Glu Pro Ser Val Arg Thr Phe Tyr Ser Lys Ala Leu Ser 530 535 540
Gly Ile Gln Ser Ser Ala Leu Thr Ser Ala Leu Pro Pro Val Thr Ala 545 550 555 560
Gln Ala Glu Gly Ala Ser Gly Thr Leu Ser Ala Gly Ala Ile Leu Arg 565 570 575
Asn Met Ala Leu Ala Ala Thr Gly Ser Val Ser Tyr Leu Ser Thr Leu 580 585 590
Tyr Thr Asn Gin Ser Val Thr Ala Glu Ala Lys Ala Lys Ala Ala 595 600 605 610 615 620

<210> SEQ ID NO 4
<211> LENGTH: 495
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae
<400> SEQUENCE: 4
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<210> SEQ ID NO 5
<211> LENGTH: 164
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae
<400> SEQUENCE: 5

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Leu Ser Leu Ile Ser Thr Pro Glu Arg Phe Tyr Glu Ser Ala Asn Phe
20 25 30
Lys Ile Ser Glu Val Asp Phe Thr Leu Gln Phe Gln Asp Arg Asp Glu
35 40 45
Gly Arg Ala Val Leu Ile Tyr Gly Asp Met Gly Ala Leu Pro Ala Arg
50 55 60
Gly Arg Glu Ser Ala Leu Leu Ala Leu Met Asp Ile Asn Phe His Met
65 70 75 80
Phe Ala Gly Ala His Ser Pro Ala Phe Ser Phe Asn Ala Gin Thr Gly
85 90 95
Arg Val Leu Leu Met Gly Ser Val Ala Leu Glu Arg Ala Ser Ala Glu
100 105 110
Gly Val Leu Leu Leu Met Lys Ser Phe Ser Asp Leu Ala Lys Glu Trp
115 120 125
Arg Glu His Gly Phe Met Gly Gin Ala Thr Thr Ala Gly Ser Ser Thr
130 135 140
Aas Gln Pro Val Ala Pro Ala Lys Arg Glu Ser Leu Ser Ala Pro
145 150 155 160
Gly Arg Phe Glu
<210> SEQ ID NO 6
<211> LENGTH: 1461
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae
<400> SEQUENCE: 6

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280
tgactgta gctgtgagtg gcaagcgttg tctgtgtaag ccagcctgct cctgcatctg
340
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Asp Thr Gly Val Ala Val Gln Thr Ser Ala Arg Asn Ala Val Arg
210 215 220
Thr Val Leu Ala Pro Ala Leu Ala Ser Arg Pro Ala Val Gln Gly Ala
225 230 235 240
Val Asp Leu Gly Val Ser Met Ala Gly Gly Leu Ala Ala Asn Ala Gly
245 250 255
Phe Gly Asn Arg Leu Leu Ser Val Gln Ser Arg Asp His Gly Arg Gly
260 265 270
Gly Ala Leu Val Leu Gly Leu Lys Asp Lys Glu Pro Lys Ala Glu Leu
275 280 285
Ser Glu Glu Asn Asp Trp Leu Glu Ala Tyr Lys Ala Ile Lys Ser Ala
290 295 300
Ser Tyr Ser Gly Ala Leu Asn Ala Gly Lys Arg Met Ala Gly Leu
305 310 315 320
Pro Leu Asp Met Ala Thr Asp Ala Met Gly Ala Val Arg Ser Leu Val
325 330 335
Ser Ala Ser Ser Leu Thr Glu Asn Leu Ala Gly Asp Leu Val Gln Gly Phe
340 345 350
 Ala Gly Val Gly Lys Leu Gln Glu Met Ala Thr Lys Asn Ile Thr Asp
355 360 365
Pro Ala Thr Lys Ala Val Ser Glu Leu Ala Leu Ala Gly Ser
370 375 380
 Ala Ala Val Phe Ala Gly Trp Thr Thr Ala Leu Thr Thr Asp Pro
385 390 395 400
 Ala Val Lys Ala Glu Ser Phe Ile Glu Asp Thr Val Lys Ser Thr
405 410 415
 Ala Ser Ser Thr Thr Gly Tyr Val Ala Asp Gin Thr Val Lys Leu Ala
420 425 430
 Lys Thr Val Lys Asp Met Gly Gly Ala Ile Thr His Thr Gly Ala
435 440 445
 Ser Leu Arg Asn Thr Val Asn Leu Arg Gin Glu Pro Ala Arg Glu
450 455 460
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Phe Arg Pro Met Arg Ser
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<210> SEQ ID NO 8
<211> LENGTH: 1074
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae

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120
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180
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240
tggggcgcaa tggtcaacac ttcacgcagt tttggccggc aggtactgac gctgatcgat
300
ggcgaacggg atgacctcaa caaccctgtc aaagccatac tctttcaacg tcatgtggct
360	acttgcgtg ccctgcgcgc gcacctcaaa ggcgacgtca aaacagcaaa actcgacggg
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420
ttacctgtgc cggacgagat tcagcgcgcc agccagagca acaacttccc caatgacatc
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540
cgtctgaccc gcctggaatc gaccatggtc gatctgtcca actgtcaggg cggcatggag
600
cgcatcgcca acacgccact gccctacccc tacgtttatt tcccacggct gttcagcacg
660
c tgttctgca tcctgatgcc gctgagcatg gtcaccaccc tgggctggtt caccccggcg
720
atctccacgg tggtaggctg catgctgctg gcaatggacc gcatcggtac agacctgcaa
780
gccccgttcg gcaacagtca gcaccggatc cgcatggaag acctgtgcaa caccatcgaa
840
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900
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960
aggttaggg aagcgcgag cttatgccg gtcgagcag gcagaaaaac
1020
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1074

<210> SEQ ID NO 9
<211> LENGTH: 357
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae
<400> SEQUENCE: 9

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Val Thr Val Asp Val Met Leu Ile Glu Gly Lys Gly Ile Asp Phe Pro
35 40 45
Leu Met Pro Leu Thr Leu Leu Cys Ser Ala Leu Ile Val Leu Ile Ser
50 55 60
Phe Arg Asn Ser Ser Ala Tyr Asn Arg Trp Trp Glu Ala Arg Thr Leu
65 70 75 80
Trp Gly Ala Met Val Asn Thr Ser Arg Ser Phe Gly Arg Gln Val Leu
85 90 95
Thr Leu Ile Asp Gly Glu Arg Asp Leu Asn Asn Pro Val Lys Ala
100 105 110
Ile Leu Phe Gln Arg His Val Ala Tyr Arg Ala Leu Arg Ala His
115 120 125
Leu Lys Gly Asp Val Lys Thr Ala Lys Leu Asp Gly Leu Ser Pro
130 135 140
Asp Glu Ile Gln Arg Ala Ser Ser Asn Asn Phe Pro Asn Asp Ile
145 150 155 160
Leu Asn Gly Ser Ala Val Ile Ser Gln Ala Phe Ala Gly Gln
165 170 175
Phe Asp Ser Ile Arg Leu Thr Arg Leu Glu Ser Thr Met Val Asp Leu
180 185 190
Ser Asn Cys Gln Gly Met Glu Arg Ile Ala Asn Thr Pro Leu Pro
195 200 205
Tyr Pro Tyr Val Tyr Phe Pro Arg Leu Phe Ser Thr Leu Phe Cys Ile
210 215 220
Leu Met Pro Leu Ser Met Val Thr Thr Leu Gly Trp Phe Thr Pro Ala
225 230 235 240
Ile Ser Thr Val Val Gly Met Leu Leu Ala Met Asp Arg Ile Gly
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US 6,852,835 B2

<210> SEQ ID NO 12
<211> LENGTH: 480
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae

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dtg 

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<210> SEQ ID NO 13
<211> LENGTH: 159
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae

<400> SEQUENCE: 13
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20 25 30
Asn Gly Ser Glu Cys Leu Leu Trp Leu Pro Glu Asp Thr Ser Leu
35 40 45
Phe Ile Phe Thr Gln Ile Glu Arg Leu Thr Met Pro Gln Asp Aan Val
50 55 60
Ile Leu Ile Ala Met Ala Leu Aan Leu Glu Pro Ala Arg Gly
65 70 75 80
Gly Ala Ala Gly Tyr Arg Asp Ser Arg Gly Glu Leu Leu Leu Arg
85 90 95
Ser Val His Ser Met Ala Asp Leu Asp Gly Leu Asp His Leu
100 105 110
Met Thr Arg Ile Ser Thr Leu Ala Val Ser Leu Gln Arg Tyr Leu Glu
115 120 125
Asp Tyr Arg Gln Glu Glu Ala Gly Tyr Thr Ala Gln Lys Glu Pro
130 135 140
Arg Phe Leu Pro Ala Val His Leu Thr Pro Arg Thr Phe Met Thr
145 150 155

<210> SEQ ID NO 14
<211> LENGTH: 288
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae

<400> SEQUENCE: 14
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180
ttggacacac tgctgctgcc ctacgacctc accgcttttc tgcccgaaaa tcttggcggt
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288

<210> SEQ ID NO 15
<211> LENGTH: 95
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae

<400> SEQUENCE: 15
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Cys Thr Trp Ser Leu Met Ile His Leu Asp Gly Glu Arg Cys Ile Tyr Pro
20 25 30
Gly Thr Arg Glu Gly Trp Ala Trp Gly Thr His Asn Gly Gly Glu Ser
35 40 45
Trp Pro Ile Leu Ile Asp Val Pro Phe Ser Leu Ala Leu Asp Thr Leu
50 55 60
Leu Leu Pro Tyr Asp Leu Thr Ala Phe Leu Pro Glu Asn Leu Leu Gly Gly
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85  90  95

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<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae
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cgcttgagaccc gcaagaaattt ttcgccgtgc gcctgttcac tgatactccg 360
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<210> SEQ ID NO 17
<211> LENGTH: 148
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae
<400> SEQUENCE: 17
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Leu Ile Leu Glu Glu Ala Val Ala Gly Phe Arg Phe Leu Thr
 20  25  30
Arg Leu Ile Glu Glu Trp Arg Ser Gly Lys Asn Arg Phe Glu Ala Lys
 35  40  45
Gly Glu Cys Leu Met Val Leu Leu Asp Gly Ala Ala Gly Ile
 50  55  60
Gly Gly Leu Ser Arg Asp Pro His Ala Arg Gly Asp Met Gly Arg Leu
 65  70  75  80
Arg Arg Leu Tyr Val Ala Ser Ala Ser Arg Gly Gln Gly Leu Gly Lys
 85  90  95
Thr Leu Val Asn Arg Leu Val Glu His Ala Ala Glu Phe Phe Ala
 100 105 110
Val Arg Leu Phe Thr Asp Pro Ser Gly Ala Lys Phe Tyr Leu Arg
 115 120 125
Cys Gly Phe Gln Ala Val Asp Glu Val His Ala Thr His Ile Lys Leu
 130 135 140
Leu Arg Arg Val
 145

<210> SEQ ID NO 18
<211> LENGTH: 11458
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae
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<221> NAME/KEY: unsure
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<223> OTHER INFORMATION: n at any position is undefined
<400> SEQUENCE: 18
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agcagctgcc gcttttttgg gcttctcttc ttctctcttc ttctctcttc ttctctcttc 4560
atgcctcccg ccccagctctc gatcgtcttc gcagcgtgct gcgtgctctt cggcgtttttg 4620
gcgccgctcc cccagctctc gatcgtcttc gcagcgtgct gcgtgctctt cggcgtttttg 4680
cccgctctcc cccagctctc gatcgtcttc gcagcgtgct gcgtgctctt cggcgtttttg 4740
While the image represents a page from a document, the content appears to be a sequence of nucleotides, which is typical in genetic or molecular biology contexts. However, without a specific context or a question related to the content, it's challenging to provide a meaningful interpretation or transformation into a more natural form. For precise analysis or translation, one would need to know the specific purpose or context in which these nucleotides are used, such as for genetic coding, analysis, or research.
caggcatcgg actgcctttg ggcgcgctca ggtaattatt gagcgccttg tcatgtgact
cggcgcagag gtgctccata aaaagcgtgg tcacgccact ggccttcaag ctcttcatgt
tattgatcag ttcacgcttg ctggacgttg aattgtgacc ctcaccaata acaagccccg
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cagttgactt cggtggtcct gctggtacag ggtgcctggt ggcgttacag 8040
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tggaactggg gggcagaccc aagggggcac ccagcggcacc ggttgacccc 8280
tgcacgttccc aaccgcgcctt aacggttcctc gcggattctc ggcgttacag 8340
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tggaactggg gggcagaccc aagggggcac ccagcggcacc ggttgacccc 9480
atgagacccg tcggtggacc ggctccaggc tattatccgc caacctatga agctgagcgt 60
cccactgcgc aagctgcagg aaacgatcgc gcccgatctt cacaggccag ttcctctcca 120
Mr Arg Pro Val Gly Gly Pro Ala Pro Gly Tyr Tyr Pro Pro Thr Tyr
1 5 10 15
Glu Ala Glu Arg Pro Thr Ala Glu Ala Ala Gly Asn Arg Ala Arg 20 25 30
Ser Ser Glu Ala Ser Ser Ser Pro Ala Ala Ser Val Ala Pro Glu Thr 35 40 45
Pro Met Leu Gly Asp Leu Lys Arg Phe Pro Ala Gly Arg Tyr Pro Asp 50 55 60
Met Lys Val Glu Asn Ile Arg Leu Lys Ile Glu Gly Glu Glu Pro Gly 65 70 75 80
Gly Lys Asp Gly Val Lys His Thr Arg Arg Arg Tyr Arg Pro Ala Ala 85 90 95
Gly Ser Ser His Val His Gly Glu Ser Val Ala Ser Thr Ser Ala 100 105 110
Ser Ala Glu Ser Lys Ala Leu Glu Asp Thr Asn Phe Lys Ala Ser Asp 115 120 125
Leu Ala Glu Leu Ala Arg Trp Cys Glu Ser Pro His Pro Tyr Ala Leu 130 135 140
Ala Pro Ser Lys Ala Ala Gly Lys Ser Ser Gin Leu Ser Ala Asn Val 145 150 155 160
Val Ser Ile Leu Leu Gin Glu Gly Lys His Ala Leu Gin Gin Arg Leu 165 170 175
Glu Ala Gin Gly Leu Lys Leu Ala Asp Val Val Val Ser Glu Gly Arg 180 185 190
Asp His Leu His Ile Asn Leu Asn Tyr Leu Glu Met Asp Ser Cys Leu 195 200 205
Gly Thr Ser Lys Gly Leu Trp Ala Pro Asp Ser Asn Asp Lys Lys Leu 210 215 220
Ile Ala Lys Ala Ala Arg Tyr Phe Asp Asp Phe Asn Ala Gln Lys Leu 225 230 235 240
Pro Glu Leu Ala Pro Leu Thr Lys Met Lys Ser Lys Asp Ser Leu Gly 245 250 255
Val Met Arg Gin Leu Leu Arg Asp Ala Pro Gly Leu Val Ile Gly Glu 260 265 270
Gly His Asn Ser Thr Ser Ser Lys Arg Glu Leu Ile Asn Asn Met Lys 275 280 285
Ser Leu Lys Ala Ser Gly Val Thr Thr Leu Phe Met Glu His Leu Cys 290 295 300
Ala Glu Ser His Asp Lys Ala Leu Asn Asn Tyr Leu Ser Ser Ala Pro Lys 305 310 315 320
Gly Ser Pro Met Pro Ala Arg Leu Asn Tyr Leu Asp Leu Gin Ser 325 330 335
Gln Gly His Gin Ala Pro Glu Leu His Thr Lys Tyr Asn Phe Thr 340 345 350
Thr Leu Val Glu Ala Ala Lys His Ala Gly Leu Arg Val Val Ser Leu 355 360 365
Asp Thr Thr Ser Thr Tyr Met Ala Pro Glu Lys Ala Glu Ile Lys Arg 370 375 380
Ala Gin Ala Met Asn Tyr Tyr Ala Ala Glu Lys Ile Arg Leu Ser Lys 385 390 395 400
Pro Glu Gly Lys Trp Val Ala Phe Val Gly Ala Thr His Ala Thr Ser 405 410 415
Cys Asp Gly Val Pro Gly Leu Ala Glu Leu His Gly Val Arg Ser Leu 420 425 430
Val Ile Asp Asp Leu Gly Leu Lys Ser Arg Ala Thr Val Asp Ile Asn 435 440 445
Val Lys Asn Tyr Gly Gly Gly Leu Asn Pro Aep Val Arg Leu Ser Tyr 450 455 460
Lys Val 465

<210> SEQ ID NO 21
<211> LENGTH: 726
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae
<400> SEQUENCE: 21
atgcacaaga cgacccctag ggcggtagcc tgtgaatgtc tggytggttg 60
gggcccggcg cgggaaagta tatcgcgggt ggcacggcag agctgaaaaa acccgttaas 120
tgaaaatcg atgcctcaac ctgaaaaca tctgatgctg tgcggaaagc cgcacccac 180
aasagttata tgataaaagc tcagctgatc gcctttgtat cagagcgtgt tcgggcaaca 240
ccgtacgcgt caacacagtgtt ggatggcctga gcgaatgtac ctgaacaatt agtcatcgac
ttcagaggtc tggattgttt tgcttatctg gattacgtcg aagcgtttcg aagatcaaca
tgcgacagg gatattgctg catcctcgcc ccaaggtgct cagctgtgac
tttttgaact caacacaccttt tcagaaggtc tcggattgttt tgcttatctg gattacgtcg aagcgtttcg aagatcaaca
tgcgacagg gatattgctg catcctcgcc ccaaggtgct cagctgtgac
tttttgaact caacacaccttt tcagaaggtc tcggattgttt tgcttatctg gattacgtcg aagcgtttcg aagatcaaca
tgcgacagg gatattgctg catcctcgcc ccaaggtgct cagctgtgac
tttttgaact caacacaccttt tcagaaggtc tcggattgttt tgcttatctg gattacgtcg aagcgtttcg aagatcaaca
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tttttgaact caacacaccttt tcagaaggtc tcggattgttt tgcttatctg gattacgtcg aagcgtttcg aagatcaaca
tgcgacagg gatattgctg catcctcgcc ccaaggtgct cagctgtgac
tttttgaact caacacaccttt tcagaaggtc tcggattgttt tgcttatctg gattacgtcg aagcgtttcg aagatcaaca
tgcgacagg gatattgctg catcctcgcc ccaaggtgct cagctgtgac
<210> SEQ ID NO 22
<211> LENGTH: 241
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae
<400> SEQUENCE: 22
Met Gln Lys Thr Thr Leu Trp Ala Ala Phe Ala Met Leu Ala Gly
1  5 10 15
Cys Gly Val Ser Gly Pro Ala Pro Gly Ser Asp Ile Gln Gly Ala Gln
20 25 30
Ala Glu Met Lys Thr Pro Val Lys Leu Asn Leu Asp Ala Tyr Thr Ser
35 40 45
Lys Lys Leu Asp Ala Val Leu Glu Ala Arg Thr Asn Lys Ser Tyr Met
50 55 60
Asp Asp Ala Gly Glu Leu Ile Asp Leu Val Ser Gly Ala Phe Leu Gly Thr
65 70 75 80
Pro Tyr Arg Ser Asn Met Leu Val Gly Ser Ala Asn Val Pro Glu Gin
85 90 95
Leu Val Ile Asp Phe Arg Gly Leu Asp Cys Phe Ala Tyr Leu Asp Tyr
100 105 110
Val Glu Ala Phe Arg Ser Thr Ser Gin Gin Asp Phe Val Arg Asn
115 120 125
Leu Val Glu Val Arg Tyr Lys Gly Gly Asp Val Phe Leu Asn Arg
130 135 140
Lys His Phe Phe Thr Asp Trp Ala Tyr Gly Thr Ala Tyr Pro Val Ala
145 150 155 160
Asp Asp Ile Thr Ala Glu Ile Ser Pro Gly Ala Val Ser Val Arg Lys
165 170 175
Arg Asn Glu Arg Ala Lys Gly Val Tyr Leu Pro Gly Leu Pro
180 185 190
Val Val Glu Arg Ser Met Thr Tyr Ile Pro Ser Arg Leu Val Asp Ser
195 200 205
Gln Val Val Ser His Leu Arg Thr Gly Asp Tyr Ile Gly Ile Tyr Thr
210 215 220
Pro Ala Ser Arg Ala Gly Cys Asp Thr Arg Phe Leu Tyr Arg Asp
225 230 235 240
Gly
<210> SEQ ID NO 23
<211> LENGTH: 417
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae
SEQUENCE: 23
atgcgcgcgt ataaaaacct gacggcaaag atcggcggct ttctgcttgc gctgacgatc
attggcactt cgctacctgc atttgccgta aacgattgtg atctggacaa cgacaacagc
accggtgcca cgtgtggcgg caacgacaag gatctggata acgacaacgt gactgacgc
gcatttggcg gcaacgacaa ggatatggac aatgaccacc acaccgacgc ggcatttggg
ggtaacgaca aggacctgga caacgatcac catacggatg cagcgtttgg cggtaacgac
aaagatctcg acaacgacaa caaaaccgat gcggctttcg gtggaaatga ccgcgatctt
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<210> SEQ ID NO 24
<211> LENGTH: 138
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae

Met Arg Ala Tyr Lys Asn Leu Thr Ala Lys Ile Gly Gly Phe Leu Leu
1 5 10 15
Ala Leu Thr Ile Ile Gly Thr Ser Leu Pro Ala Phe Ala Val Asn Asp
20 25 30
Cys Asp Leu Asp Asn Ser Thr Gly Ala Thr Cys Gly Gly Aen
35 40 45
Asp Lys Asp Asp Asn Ser Thr Gly Ala Thr Cys Gly Gly Aen
50 55 60
Asp Lys Asp Met Asp Asn Ser Thr Thr Asp Ala Ala Phe Gly
65 70 75 80
Gly Aen Asp Lys Asp Leu Asp Asn Ser Thr Thr Asp Ala Ala Phe
85 90 95
Gly Gly Aen Ase Lys Leu Ase Asp Asn Asp Lys Leu Lys Asp Ala Ala
100 105 110
Phe Gly Gly Aen Asp Arg Asp Leu Aen Asp Aen Ase Asp Leu Aen
115 120 125
Tyr Aen Gly Thr Pro Ser Ala Ala Lys Lys
130 135

<210> SEQ ID NO 25
<211> LENGTH: 411
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae

<400> SEQUENCE: 25
atgaacaaga tcgtctacgt aaaaagttac ttttaaccccct ttggggagga agtcgcttc
atggcgttct cacgagtcta ctgggagga aagagctgg forecast gatggctctc
agasggcag cggccggatt tccgagacgc gcacgccg gagggagc ggggaggcat
acgctgacctc gacgccgaga gttgctcag cggcgcgct ggtgtgctg gctttcagc
gagggcggcc gccgggatgg ttttttgtgg cggctggctg ctggtcgggt ggggggggg
<210> SEQ ID NO 26
<211> LENGTH: 136
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae
SEQ ID NO 26
LENGTH: 972
TYPE: DNA
ORGANISM: Pseudomonas syringae

SEQUENCE:

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Met Asn Lys Ile Val Tyr Val Lys Ala Tyr Phe Lys Pro Ile Gly Glu Glu Val Ser Val Lys Val Pro Thr Gly Glu Ile Lys Gly Phe Phe Glu Val Ser Gly Ala Tyr Glu Ile Gln Thr Val Leu Pro Ile Leu Ser Gin Ala Tyr Ala Leu Lys Tyr Arg Tyr Glu Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gin Gi```
Met Gly Cys Val Ser Ser Lys Ala Ser Val Ile Ser Ser Asp Ser Phe 1 5 10 15
Arg Ala Ser Tyr Thr Asn Ser Pro Glu Ala Ser Val His Glu Arg 20 25 30
Ala Arg Thr Pro Arg Cys Gly Glu Gin Gly Pro Gin Val Ser Arg 35 40 45
Leu Met Pro Tyr Gin Gin Ala Leu Val Gly Val Ala Arg Trp Pro Asn 50 55 60
Pro His Phe Asn Arg Asp Ala Pro His Gin Met Glu Tyr Gly Glu 65 70 75 80
Ser Phe Tyr His Lys Ser Arg Glu Leu Gly Ala Ser Val Ala Asn Gly 85 90 95
Glu Ile Glu Thr Phe Gin Glu Leu Trp Ser Glu Ala Arg Asp Trp Arg 100 105 110
Ala Ser Arg Ala Gly Gin Asp Ala Arg Leu Phe Ser Ser Arg Asp 115 120 125
Pro Asn Ser Ser Arg Ala Phe Val Thr Pro Ile Thr Gly Pro Tyr Glu 130 135 140
Phe Leu Lys Aep Arg Phe Ala Asn Arg Lys Aep Gly Glu Lys His Lys 145 150 155 160
Met Met Asp Phe Leu Pro His Ser Asn Thr Phe Arg Phe His Gly Lys 165 170 175
Ile Asp Gly Glu Arg Leu Pro Leu Thr Trp Ile Ser Ser Ser Asp 180 185 190
Arg Arg Ala Asp Arg Thr Lys Asp Pro Tyr Gin Arg Leu Arg Asp Gln 195 200 205
Gly Met Asn Aep Val Gly Glu Pro Aep Val Met Leu His Thr Gin Ala 210 215 220
Glu Tyr Val Pro Lys Ile Met Gin His Val Glu His Leu Tyr Lys Ala 225 230 235 240
Ala Thr Asp Ala Ala Leu Ser Asp Ala Asn Ala Leu Lys Lys Leu Ala 245 250 255
Glu Ile His Trp Trp Thr Val Gin Ala Val Pro Asp Phe Arg Gly Ser 260 265 270
Ala Ala Lys Ala Glu Leu Cys Val Arg Ser Ile Ala Glu Ala Arg Gly 275 280 285
Met Gin Leu Pro Pro Pro Met Arg Leu Gly Ile Val Pro Asp Leu Gin Ala 290 295 300
Leu Thr Met Pro Leu Lys Asp Phe Val Lys Ser Tyr Gin Gly Phe Phe 305 310 315 320
Glu His Asn

<210> SEQ ID NO 29
<211> LENGTH: 1149
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae
<400> SEQUENCE: 29
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gaaaaggccg tgcaatcatc ggcccaagcg cagaatgaag cgtctcacag cggtccatca 120
gaacatcctg aatcccgctc ctgtcaggca cgcccgaact acccttattc gtcagtcaaa 180
acacggttac cccctgttgc gtctgcaggg cagtcgctgt ctgagacacc ctcttcattg 240
cctggctacc tgctgttacg tcggcttgat cgtcgtccgc tggaccagga cgcaataaag 300
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360

30

382

Pseudomonas syringae

<210> SEQ ID NO 30
<211> LENGTH: 382
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae

<400> SEQUENCE:

Met Arg Ile His Ser Ser Gly His Gly Ile Ser Gly Pro Val Ser Ser
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Ala Glu Thr Val Glu Lys Ala Val Gln Ser Ser Ala Gln Ala Gln Asn
20 25 30
Glu Ala Ser His Ser Gly Pro Ser Glu His Pro Ser Arg Ser Cys
35 40 45
Gln Ala Arg Pro Asn Tyr Pro Tyr Ser Ser Val Lys Thr Arg Leu Pro
50 55 60
Pro Val Ala Ser Ala Gly Gin Ser Leu Ser Glu Thr Pro Ser Ser Leu
65 70 75 80
Pro Gly Tyr Leu Leu Leu Arg Arg Leu Asp Arg Arg Pro Leu Asp Gln
85 90
Asp Ala Ile Lys Gly Leu Ile Pro Ala Asp Glu Ala Val Gly Glu Ala
100 105 110
Arg Arg Ala Leu Pro Phe Gly Arg Gly Asn Ile Asp Val Asp Ala Gln
115 120 125
Arg Ser Asn Leu Glu Ser Gly Ala Arg Thr Leu Ala Ala Arg Arg Leu
130 135 140
Arg Lys Asp Ala Glu Thr Ala Gly His Glu Pro Met Pro Glu Asn Glu
145 150 155 160
Asp Met Asn Trp His Val Leu Val Ala Met Ser Gly Gin Val Phe Glu
165 170 175
Ala Gly Asn Cys Gly Glu His Ala Arg Ile Ala Ser Phe Ala Tyr Glu
180 185
Ala Ser Ala Gin Glu Lys Gly Arg Ala Asp Gin Ile His Leu
190 195 200 205
Ala Ala Gin Ser Gly Glu Asp His Val Trp Ala Glu Thr Asp Asp Ser
210 215 220
225 230
235
240 245 250 255
260 265 270 275
280 285 290 295
300 305 310 315
320 325 330 335
340 345 350 355
360 365 370 375
380 385

382

-continued
Ser Ala Gly Ser Ser Pro Ile Val Met Asp Pro Thr Ser Asn Gly Pro
225 230 235 240
Ala Val Phe Ala Glu Asp Ser Arg Phe Ala Lys Asp Arg Ala Val
245 250 255
Glu Arg Thr Asp Ser Phe Thr Leu Ser Thr Ala Ala Lys Ala Gly Lys
260 265 270
Ile Thr Arg Glu Thr Ala Lys Ala Leu Thr Gln Ala Thr Ser Arg
275 280 285
Leu Glu Glu Arg Leu Ala Glu Asp Glu Ala Glu Ala Val Ser Pro Val Glu
290 295 300
Gly Gly Arg Tyr Arg Glu Asn Ser Val Leu Asp Asp Ala Phe Ala
305 310 315 320
Arg Arg Val Ser Asp Met Leu Asn Asn Ala Asp Pro Arg Ala Leu
325 330 335
Gln Val Glu Ile Glu Ala Ser Gly Val Ala Met Ser Leu Gly Ala Gln
340 345 350
Gly Val Lys Thr Val Val Arg Glu Ala Pro Lys Val Arg Glu Ala
355 360 365
Arg Gly Val Ala Ser Ala Lys Gly Met Ser Pro Arg Ala Thr
370 375 380

<210> SEQ ID NO 31
<211> LENGTH: 1236
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae

<400> SEQUENCE: 31

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tcgtcatcat cggtaactaa cccaccgcta cagcgtggcg agggcagacg tctgcgacgt
caggatgcgc tgccaacgga tatcagatac aacgccaacc agacagcgac atcaccgcaa
aacgcgcgcg cggcaggaag atatgaatca ggggccagct catccggcgc gaatgatact
cgcaggctg aaggttcaat gccttcgtcg tccgcccttt tacaatttcg cctcgccggc
ggcggaacc attctgagct ggaaaatttt catactatga tgctgaactc accgaaagca
tcacggggag atgctatacc tgagaagccc gaagcaatac ctaagcgcct actggagaag
atggaaccga ttaacctggc ccagttagct ttgcgtgata aggatctgca tgaatatgcc
gtaatggtct gtaaccaagt gaaaaagggt gaaggtccga actccaatat tacgcaagga
gatatcaagt tactgccgct gttcgccaaa gcggaaaata caagaaatcc cggcttgaat
cgcgcgcgcg ggtcagcacg ttacctctat tgaaggtttc agaatgcagg aaataaagag agcaggtgac
ttccttgccg camaaggt cgggcgaag cttggag

<210> SEQ ID NO 32
<211> LENGTH: 411
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae

<400> SEQUENCE: 32

Met Asn Ile Ser Gly Pro Asn Arg Gln Gly Thr Gln Ala Glu Asn
1 5 10 15
Thr Glu Ser Ala Ser Ser Ser Val Thr Asn Pro Leu Gln Arg
20 25 30
Gly Glu Gly Arg Arg Leu Arg Gln Ala Leu Pro Thr Asp Ile
35 40 45
Arg Tyr Asn Ala Asn Gln Thr Ala Thr Ser Pro Gln Asn Ala Arg Ala
50 55 60
Ala Gly Arg Tyr Glu Ser Gly Ala Ser Ser Gly Ala Asn Asp Thr
65 70 75 80
Pro Gln Ala Glu Gly Ser Met Pro Ser Ser Ala Leu Leu Gln Phe
85 90 95
Arg Leu Ala Gly Gly Arg Asn His Ser Glu Leu Glu Asn Phe His Thr
100 105 110
Met Met Leu Asn Ser Pro Lys Ala Ser Arg Gly Asp Ala Ile Pro Glu
115 120 125
Lys Pro Glu Ala Ile Pro Lys Arg Leu Glu Lys Met Glu Pro Ile
130 135 140
Aan Leu Ala Gln Leu Ala Leu Arg Asp Lys Asp Leu His Gln Tyr Ala
145 150 155 160
Val Met Val Cys Asn Gln Val Lys Gly Gln Gly Gln Pro Asn Ser Asn
165 170 175
Ile Thr Gln Gly Asp Ile Lys Leu Pro Leu Phe Ala Lys Ala Glu
180 185 190
Asn Thr Arg Asn Pro Gly Leu Asn Leu His Thr Phe Lys Ser His Lys
195 200 205
Aas Cys Tyr Gln Ala Ile Lys Gly Gln Asn Arg Asp Ile Gln Lys Aan
210 215 220
Lys Gln Ser Leu Ser Met Arg Val Tyr Pro Pro Phe Lys Met
225 230 235 240
Pro Asp His His Ile Ala Leu Asp Ile Gln Leu Arg Tyr Gly His Arg
245 250 255
Pro Ser Ile Val Gly Phe Glu Ser Ala Pro Gly Asn Ile Asp Ala
260 265 270
Ala Gln Arg Glu Ile Leu Ser Ala Leu Gly Asn Val Lys Ile Lys Met
275 280 285
Val Gly Asn Phe Leu Gln Tyr Ser Lys Thr Asp Cys Thr Met Phe Ala
290 295 300
Leu Aan Asn Ala Leu Lys Ala Phe Lys His His Glu Gln Tyr Thr Ala
305 310 315 320
Arg Leu His Aan Gly Glu Lys Gln Val Pro Ile Pro Ala Thr Phe Leu
325 330 335
Lys His Ala Gln Ser Lys Ser Leu Val Glu Asn His Pro Glu Lys Aas
340 345 350
Thr Thr Val Thr Lys Asp Gly Gln Gly Leu His Met Glu Thr Leu
355 360 365
His Arg Asn Arg Ala Tyr Arg Ala Glu Arg Ser Ala Gly Gln His Val 370 375 380
Thr Ser Ile Glu Gly Phe Arg Met Glu Ile Lys Arg Ala Gly Asp 385 390 395 400
Phe Leu Ala Ala Asn Arg Val Arg Ala Lys Pro 405 410

<210> SEQ ID NO 33
<211> LENGTH: 363
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae

SEQUENCE: 33
atgacgctgg aacggattga acagcaaaat acgctgtttg tttatctgtg cgtgggcacg 60
cttotactc cagcacaag cacacactctg agogatattc tggcggcaca cctotttcctat 120
tatgggcca gcgattggcg ggccttcggg cggcttcagc gcggcttcagc 180
tttcagctgct ggcggcagc gcggcttcagc gcggcttcagc gcggcttcagc 240
atggcgggca aatggcgggca ggcggcagc gcggcttcagc gcggcttcagc gcggcttcagc 300
gccgctggc gcggcttcagc gcggcttcagc gcggcttcagc gcggcttcagc gcggcttcagc 360
gccgctggc gcggcttcagc gcggcttcagc gcggcttcagc gcggcttcagc gcggcttcagc 363
tga

<210> SEQ ID NO 34
<211> LENGTH: 120
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae

SEQUENCE: 34
Met Thr Leu Glu Arg Ile Glu Gln Gln Asn Thr Leu Phe Val Tyr Leu 1 5 10 15
Cys Val Gly Thr Leu Ser Thr Pro Ala Ser Ser Thr Leu Leu Ser Asp 20 25 30
Ile Leu Ala Ala Asn Leu Phe His Tyr Gly Ser Ser Asp Gly Ala Ala 35 40 45
Phe Gly Leu Asp Glu Lys Asn Asn Glu Val Leu Leu Phe Gln Arg Phe 50 55 60
Asp Pro Leu Arg Ile Asp Glu Asp His Phe Val Ser Ala Cys Val Gln 65 70 75 80
Met Ile Glu Val Ala Lys Ile Trp Arg Ala Lys Leu His Gly His 85 90 95
Ser Ala Pro Leu Ala Ser Ser Thr Arg Leu Thr Lys Ala Gly Leu Met 100 105 110
Leu Thr Met Ala Gly Thr Ile Arg 115 120

<210> SEQ ID NO 35
<211> LENGTH: 1128
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae

SEQUENCE: 35
gtgaacccta tccatgcacg cttctccagc gtagaagcgc tcagacattc aaacgttgat 60
atttcgaagcg ccatacgtc gcgtgtcggc gggcggtgct cggcggcttc gcggcttcagc 120
gccgctggc gcccggttc ctctcgcggc gcggcttcagc gcggcttcagc gcggcttcagc 180
tttcagctgct ggcggcagc gcggcttcagc gcggcttcagc gcggcttcagc gcggcttcagc 240
gtactcaacg agaaacgccc gcgcttgcca gattggtgga cagaacgctc 300
gacctggaga agggcgggaag tagcgccttg gccgccgcaas tcacagccctg cagacgcttc 360
cctgactaaa aacagcact tgcagccttc cagcaaaaaag ctagggccttc 420
ggcgcgatacc gaaatcggccc gcgctgttgc ggcgccgcaas tcagcggggc agacacagcgc 480
tatgcattgc gcagcgttcc gaaatcgcct gcagcggggc acagcgggctgataa 540
aaacactcctagcagccc gcagatggtc gatagctggc cagagggaga gctggtcctg 600
ggcgcgatacc gaaatcgcct gcagaacgctc agacacgcttc ttttgtatag gttctctaa 660
gaccaacgct cactcggagc aacatcggga cagccttcga tctgtgtgag acacacttat 720
ggcgcgactg tggccctgac aatggcacaac ctgactgctgc aacgacatac tggcagcaca 780
ggctgaaagtc ggtgctgctg gcacacccgc ccacacacgc ctctctgtgc tctctctctc 840
aatgggatgtg tcagcggggc ctagacgctg cagcggggc ctagcgcggc 900
tctgtgactgc gttctgctgtgc aacatcgcct gcagcggggc acagcggggc aacacgcttc 960
acagcggggc ctagacgctg cagcggggc ctagcgcggc 1020
tctgtgactgc gttctgctgtgc aacatcgcct gcagcggggc acagcggggc aacacgcttc 1080
aacatcgcct gcagcggggc ctagacgctg cagcggggc ctagcgcggc 1140

<210> SEQ ID NO 36
<211> LENGTH: 375
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae
<400> SEQUENCE: 36

Val Asn Pro Ile His Ala Arg Phe Ser Ser Val Glu Ala Leu Arg His 1 5 10 15
Ser Asn Val Aep Ile Gln Ala Ile Lys Ser Glu Gly Gln Leu Glu Val 20 25 30
Asn Gly Lys Arg Tyr Glu Ile Arg Ala Ala Aap Gly Ser Ile Ala 35 40 45
Val Leu Arg Pro Aep Gln Glu Ser Glu Ala Asp Lye Phe Lys Gly 50 55 60
 Ala Ala His Leu Ile Gly Gly Glu Ser Glu Arg Ala Glu Ile Ala Glu 65 70 75 80
Val Leu Aep Glu Lye Ala Ala Val Pro Arg Leu Aep Arg Met Leu 85 90 95
Gly Arg Arg Phe Aep Lue Glu Lye Gly Gly Seer Ser Ala Val Gly Ala 100 105 110
 Ala Ile Lys Ala Ala Aep Ser Arg Leu Thr Ser Lye Gln Thr Phe Ala 115 120 125
Ser Phe Gln Glu Trp Ala Glu Lys Ala Glu Ala Leu Arg Tyr Arg 130 135 140
Asn Arg Tyr Leu His Aep Leu Gln Glu Gly His Ala Arg His Aep Ala 145 150 155 160
Tyr Glu Cye Gly Arg Val Lye Aen Ile Thr Trp Lye Arg Tyc Arg Leu 165 170 175
Ser Ile Thr Arg Lye Thr Leur Ser Tyr Ala Pro Gln Ile His Aep Aep 180 185 190
Arg Glu Glu Glu Leu Aep Lye Arg Tyc Ile Ala Glu Aep Arg 195 200 205
Aep Aen Ala Thr Gly Phe Phe Aep Met Val Pro Lye Aep Glu Arg Ala
Pro Glu Thr Asn Ser Gly Arg Leu Thr Ile Gly Val Glu Pro Lys Tyr
Gly Ala Gln Leu Ala Leu Ala Met Ala Thr Leu Met Asp Lys His Lys
Ser Val Thr Gln Gly Lys Val Gly Pro Ala Lys Tyr Gly Glu Gln
Thr Asp Ser Ala Ile Leu Tyr Ile Asn Gly Asp Leu Ala Lys Ala Val
Lys Leu Gly Glu Lys Leu Lys Leu Ser Gly Ile Pro Pro Glu Gly
Phc Val Glu His Thr Pro Leu Ser Met Gln Ser Thr Gly Leu Gly Leu
Ser Tyr Ala Glu Ser Val Gly Gln Pro Ser Ser His Gly Gln Ala
Arg Thr His Val Ile Met Asp Ala Leu Lys Gly Gln Gly Pro Met Glu
Asn Arg Leu Lys Met Ala Leu Ala Glu Arg Gly Tyr Asp Pro Glu Asn
Pro Ala Leu Arg Ala Arg Asn

<210> SEQ ID NO 37
<211> LENGTH: 336
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae

<400> SEQUENCE: 37
atggagatgc ccgccttggc gtttgacgat aagggtgcgt gcaacatgat catcgacaag  
gcattcgctc tgacgctgtt gcgcgacgac acgcatcaac gtttgttgct gattggtctg  
cttgagccac acgaggatct acccttgcag cgcctgttgg ctggcgctct caaccccctt  
gtaatgccg gccccggcat tggctgggat gagcaaagcg gcctgtacca cgcttaccaa  
agcatcccgc gggaaaaagt cagcgtggag atgctgaagc tcgaaattgc aggattggtc  
gaatggatga agtgttggcg agaagcccgc acgtga

<210> SEQ ID NO 38
<211> LENGTH: 111
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae

<400> SEQUENCE: 38
Met Glu Met Pro Ala Leu Ala Phe Asp Asp Lys Gly Ala Cys Asn Met
Ile Ile Asp Lys Ala Phe Ala Leu Thr Leu Leu Arg Asp Thr His
Gln Arg Leu Leu Ule Gly Leu Glu Pro His Glu Asp Leu Pro
Leu Gln Arg Leu Leu Ala Leu Asn Pro Leu Val Asn Ala Gly
Pro Gly Ile Gly Trp Asp Glu Gln Ser Gly Lys Tyr His Ala Tyr Gln
Ser Ile Pro Arg Glu Lys Val Ser Val Glu Met Leu Lys Leu Glu Ile
Ala Gly Leu Val Glu Trp Met Lys Cys Trp Arg Glu Ala Arg Thr
### Sequence Information

**SEQ ID NO**: 39  
**LENGTH**: 1143  
**TYPE**: DNA  
**ORGANISM**: Pseudomonas syringae pv. syringae

#### Sequence

```
ATGAGAATTC ACAGTGCTGG TCACAGCCTG CCGGCTCAG
GGAACGCTT 60
GAATAAGGCT GTTCAATCAT TGTCGGCCAG GAACCGCGCC CTGTTGCTGG 120
CGTCCTGAAG CCGGCGCCAG CGCAGCGACT CTTACAGTCA CAGGGCGGCT 180
CTGGTCCGG CAGAGGAGAC GGGATTGAGC CGTGGCAGAC CAGGCTTGCT 240
CGCCGCGGCG GTGTTGCTGG TCACAGTCCA CCCGAGGCGG 300
AACATTGATG TGGATGCGGT GGTGGCTTCA GCTGCTGCTG 360
GGCGTGGCTA ATGAGGATG TCAGTGAGGC TGGCTGCTGG 420
```

### Protein Information

**SEQ ID NO**: 40  
**LENGTH**: 380  
**TYPE**: PRT  
**ORGANISM**: Pseudomonas syringae pv. syringae

#### Protein Sequence

```
Met Arg Ile His Ser Ala Gly His Ser Leu Pro Ala Pro Gly Pro Ser 1 5 10 15
Val Glu Thr Thr Glu Lys Ala Val Gln Ser Ser Ser Ala Gln Asn Pro 20 25 30
Ala Ser Tyr Ser Ser Gln Thr Glu Arg Pro Glu Ala Gly Ser Thr Gln 35 40
Val Arg Leu Asn Tyr Pro Tyr Ser Ser Val Lys Thr Arg Leu Pro Pro 50 55 60
Val Ser Ser Thr Gly Gln Ala Ile Ser Ala Thr Pro Ser Ser Leu Pro 65 70 75 80
Gly Tyr Leu Leu Arg Leu Arg Leu Asp Arg Arg Pro Leu Asp Glu Asp 85 90 95
Ser Ile Lys Ala Leu Val Pro Ala Asp Glu Ala Val Arg Glu Ala Arg 100 105 110
```
<210> SEQ ID NO: 41
<211> LENGTH: 1143
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae pv. glycinea

**SEQUENCE: 41**

```
atgagaattc acagtgctgg tcacagcctg cccgcgccag gccctagcgt ggaaaccact
  gaaaaggctg ttcaatcatc atcggcccag aaccccgctt cttgcagttc acaaacagaa
  cgtcctgaag ccggttcgac tcaagtgcga ccgaactacc cttactcatc agtcaagaca
  cgcttgccac ccgtttcttc cacagggcag gccatttctg acacgccatc ttcattgtcc
  ggttacctgc tgttacgtcg gctcgaccga cgtccactgg atgaagacag tatcaaggct
  ctggttccgg cagacgaagc gttgcgtgaa gcacgccgcg cgttgccctt cggcaggggc
  aacattgatg tggatgcaca acgtacccac ctgcaaagcg gcgctcgcgc agtcgctgca
  aagcgcttga gaaaagatgc cgagcgcgct ggccatgagc cgatgcccga gaatgatgag
  atgaactggc atgttct tgtcgccatgt ca gggcaggtgt ttggcgctgg caactgtggc
  gaacatgctc gtatagcaag cttcgcttac ggggccctgg ctcaggaaag cgggcgtagt
```

---continued---
<210> SEQ ID NO 42
<211> LENGTH: 380
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae pv. glycinea

<400> SEQUENCE: 42

Met Arg Ile His Ser Ala Gly His Ser Leu Pro Ala Pro Gly Pro Ser
Val Glu Thr Thr Glu Lys Ala Val Gln Ser Ser Ser Ala Gln Asn Pro
Ala Ser Cys Ser Ser Gln Thr Glu Arg Pro Glu Ala Gly Ser Thr Gln
Val Arg Pro Asn Tyr Pro Tyr Ser Ser Val Lys Thr Arg Leu Pro Pro
Val Ser Ser Thr Gly Gln Ala Ile Ser Asp Thr Pro Ser Ser Leu Ser
Gly Tyr Leu Leu Leu Arg Arg Leu Arg Arg Pro Leu Asp Glu Asp
Ser Ile Lys Ala Leu Val Pro Ala Asp Glu Ala Leu Arg Glu Ala Arg
Arg Ala Leu Pro Phe Gly Arg Gly Asn Ile Asp Val Asp Ala Glu Arg
Thr His Leu Gln Ser Gly Ala Arg Ala Val Ala Lys Arg Leu Arg
Lys Asp Ala Glu Arg Ala Gly His Glu Pro Met Pro Glu Asp Glu
Met Asn Trp His Val Leu Val Ala Met Ser Gly Gln Val Phe Gly Ala
Gly Asn Cys Gly Glu His Ala Arg Ile Ala Ser Phe Ala Tyr Gly Ala
Leu Ala Gln Glu Ser Gly Arg Ser Pro Arg Glu Lys Ile His Leu Ala
Glu Gln Pro Gly Lys Asp His Val Trp Ala Glu Thr Asp Asn Ser Ser
Ala Gly Ser Ser Pro Ile Val Met Asp Pro Trp Ser Asn Gly Val Ala
Ile Leu Ala Glu Asp Ser Arg Phe Ala Lys Asp Arg Ser Ala Val Glu
Arg Thr Tyr Ser Phe Thr Leu Ala Met Ala Ala Glu Ala Gly Lys Val

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### Additional Information:

- **SEQ ID NO:** 43
- **LENGTH:** 1143
- **TYPE:** DNA
- **ORGANISM:** Pseudomonas syringae pv. tabaci

### Sequence:

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atgagaattc acagtgctgg tcacagcctg cctgcgccag gccctagcgt ggaaaccact 60
gaaaaggctg ttcaatcatc atcggcccag aaccccgctt cttgcagttc acaaacagaa 120
cgtcctgaag ccggttcgac tcaagtgcga ccgaactacc cttacttctg acaacgacaa 180
gttaacctgc tgtaacctgc gtctgacgcc ccagaacagc cgaagccagc gcgaagccagc 240
cacaacctgc aggccgaagc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 300
gtctgctgg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 360
ttcctggctg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 420
ttcctggctg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 480
ttcctggctg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 540
ttcctggctg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 600
ttcctggctg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 660
ttcctggctg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 720
ttcctggctg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 780
ttcctggctg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 840
ttcctggctg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 900
ttcctggctg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 960
ttcctggctg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 1020
ttcctggctg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 1080
ttcctggctg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 1140
ttcctggctg gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc gcagaagcgc 1143
```

### Additional Details:

- **SEQ ID NO:** 44
- **LENGTH:** 380
- **TYPE:** PRT
- **ORGANISM:** Pseudomonas syringae pv. tabaci

### Sequence:

```
```
Val Glu Thr Thr Glu Lys Ala Val Gln Ser Ser Ser Ala Gln Asn Pro
20 25 30
Ala Ser Cys Ser Ser Glu Thr Glu Pro Glu Ala Gly Ser Thr Gln
35 40 45
Val Arg Pro Asn Tyr Pro Tyr Ser Ser Val Lys Thr Arg Leu Pro Pro
50 55 60
Val Ser Ser Thr Gly Glu Ala Ile Ser Asp Thr Pro Ser Ser Leu Pro
65 70 75 80
Gly Tyr Leu Leu Leu Arg Arg Leu Asp Arg Arg Pro Leu Asp Gln Asp
85 90 95
Ser Ile Lys Ala Leu Val Pro Ala Asp Glu Ala Val Arg Glu Ala Arg
100 105 110
Arg Ala Leu Pro Phe Gly Arg Gly Arg Ile Asp Val Asp Ala Gln Arg
115 120 125
Thr His Leu Gln Ser Gly Ala Val Ala Lys Arg Leu Arg
130 135 140
Lys Asp Ala Glu Arg Ala Gly His Glu His Glu Pro Met Pro Gly Asp Glu
145 150 155 160
Met Asn Trp His Val Leu Val Ala Met Ser Gly Glu Val Phe Gly Ala
165 170 175
Gly Asn Cys Gly Glu His Ala Arg Ile Ala Ser Phe Ala Tyr Gly Ala
180 185 190
Leu Ala Gln Glu Ser Gly Arg Ser Pro Arg Glu Lys Ile His Leu Ala
195 200 205
Glu Gln Pro Gly Lys Asp His Val Trp Ala Glu Thr Asp Asn Ser Ser
210 215 220
Ala Gly Ser Ser Pro Ile Val Met Asp Pro Trp Ser Asn Gly Ala Ala
225 230 235 240
Ile Leu Ala Gln Ser Arg Ser Arg Ala Lys Asp Arg Ser Asp Ala Val Glu
245 250 255
Arg Thr Tyr Ser Phe Thr Leu Ala Met Ala Ala Glu Ala Gly Lys Val
260 265 270
Thr Arg Glu Thr Ala Glu Val Leu Thr His Thr Thr Ser Arg Leu
275 280 285
Gln Lys Arg Leu Ala Asp Gln Leu Pro Asn Val Ser Pro Leu Glu Gly
290 295 300
Gly Arg Tyr Glu Gln Glu Lys Ser Val Leu Asp Glu Ala Phe Ala Arg
305 310 315 320
Arg Val Ser Asp Lys Leu Asn Ser Asp Pro Arg Arg Ala Leu Gln
325 330 335
Met Glu Ile Gln Ala Val Gly Val Ala Met Ser Leu Gly Ala Glu Gly
340 345 350
Val Lys Thr Val Ala Arg Glu Val Gly Val Ala Pro Lys Val Arg Glu Ala Arg
355 360 365
Ser Val Ala Ser Ser Lys Gly Met Pro Pro Arg Arg
370 375 380
<210> SEQ ID NO 45
<211> LENGTH: 1143
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae pv. tabaci
<400> SEQUENCE: 45
atgagaattc acagtgctgg tcacagcctg cctgc gccag gccctagcgt ggaaaccact
60
gaaaggtctg ttcacatctc atcgccccag aaccccgctt cttgcagttc acaaacagaa

cgtcctgaag ccggttcgac tcaagtgcga ccgaactacc cttactcatc agtcaagaca
cgccttgccac ccgtttcttc tacagggcag gccatttctg acacgccatc ttcattgccc
ggttacctgc tgttacgtcg gctcgaccga cgtccactgg atgaagacag tatcaaggct

cctggttccgg cagacgaagc ggtgcgtgaa gcacgccgcg cgttgccctt cggcaggggc

aacattgatg tggatgcaca acgtacccac ctgcaaagcg gcgctcgcgc agtcgctgca

gaaaagatgc cgagcgcgct ggccatgagc cgatgcccgg gaatgatgag

gttcgcccga cgagtgagcg acaagttgaa tagtgacgat ccacggcgtg cgttgcagat ggaaattgaa
 gcctgttggtg ttgcaatgtc gctgggtgcc gaaggcgtca agacggtcgc ccgacaggcg

caaaggtgg tcaggcaagc cagaagcgtc gcgtcgtcta aaggcatgcc tccacgaaga

taa

<210> SEQ ID NO 46
<211> LENGTH: 380
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae pv. tabaci

<400> SEQUENCE: 46

Met Arg Ile His Ser Ala Gly Ser Leu Pro Ala Pro Gly Pro Ser
1  5 10 15
Val Glu Thr Thr Glu Ala Val Glu Ser Ser Ser Ala Glu Aan Pro
20 25 30
Ala Ser Cys Ser Ser Gln Thr Glu Arg Pro Glu Ala Gly Ser Thr Gln
35 40 45
Val Arg Pro Aan Tyr Pro Tyr Ser Ser Val Lys Thr Arg Leu Pro Pro
50 55 60
Val Ser Ser Thr Gly Gln Ala Ile Ser Asp Thr Pro Ser Ser Leu Pro
65 70 75 80
Gly Tyr Leu Leu Arg Arg Leu Arg Asp Arg Arg Pro Leu Asp Glu Asp
85 90 95
Ser Ile Lys Ala Leu Val Pro Ala Asp Glu Ala Val Arg Glu Ala Arg
100 105 110
Arg Ala Leu Pro Phe Gly Arg Gly Aan Ile Asp Val Aan Leu Glu Arg
115 120 125
Thr His Leu Gln Ser Gly Ala Arg Ala Val Ala Lys Arg Leu Arg
130 135 140
Lys Asp Ala Glu Arg Ala Gly His Glu Pro Met Pro Gly Aan Asp Glu
145 150 155 160
Met Aan Trp His Val Leu Val Ala Met Ser Gly Glu Val Phe Gly Ala
165 170 175

---continued---
Gly Asn Cys Gly Glu His Ala Arg Ile Ala Ser Phe Ala Tyr Gly Ala
180 185 190
Leu Ala Gln Glu Ser Gly Arg Ser Pro Arg Glu Lys His Leu Ala
195 200 205
Glu Glu Pro Gly Lys Asp His Val Trp Ala Glu Thr Asp Asn Ser Ser
210 215 220
Ala Gly Ser Ser Pro Ile Val Met Asp Pro Trp Ser Asn Gly Ala Ala
225 230 235 240
Ile Leu Ala Glu Asp Ser Arg Phe Ala Lys Asp Arg Ser Ala Val Glu
245 250 255
Arg Thr Tyr Ser Phe Thr Leu Ala Met Ala Ala Gly Lys Val
260 265 270
Thr Arg Glu Thr Ala Glu Asn Val Leu Thr His Thr Ser Arg Leu
275 280 285
Gln Lys Arg Leu Ala Asp Gln Leu Pro Asn Val Ser Pro Leu Glu Gly
290 295 300
Gly Arg Tyr Gln Gln Gly Lys Ser Val Leu Asp Glu Ala Phe Ala Arg
305 310 315 320
Arg Val Ser Asp Lys Leu Asn Ser Asp Pro Arg Arg Ala Leu Glu
325 330 335
Met Glu Ile Glu Ala Val Gly Val Ala Met Ser Leu Gly Ala Glu Gly
340 345 350
Val Lys Thr Val Ala Arg Gln Ala Pro Lys Val Arg Glu Glu Ala Arg
355 360 365
Ser Val Ala Ser Ser Lys Gly Met Pro Pro Arg Arg
370 375 380

<210> SEQ ID NO 47
<211> LENGTH: 1143
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae pv. glycinea

<400> SEQUENCE: 47
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60
ggaagtcgcggaaagagaagtcgcgaggtctagatggtctagctgaaacactac
120
gcggtctgctgcatggaccggctggtctacgcgcgagaagtctggaggaagctggtc
180
cgcgcgtctgctggtctacgcgcgagaagtctggaggaagctggtcggagggagc
240
cgcgcgtctgctggtctacgcgcgagaagtctggaggaagctggtcggagggagc
300
cgcgcgtctgctggtctacgcgcgagaagtctggaggaagctggtcggagggagc
360
cgcgcgtctgctggtctacgcgcgagaagtctggaggaagctggtcggagggagc
420
cgcgcgtctgctggtctacgcgcgagaagtctggaggaagctggtcggagggagc
480
cgcgcgtctgctggtctacgcgcgagaagtctggaggaagctggtcggagggagc
540
cgcgcgtctgctggtctacgcgcgagaagtctggaggaagctggtcggagggagc
600
cgcgcgtctgctggtctacgcgcgagaagtctggaggaagctggtcggagggagc
660
cgcgcgtctgctggtctacgcgcgagaagtctggaggaagctggtcggagggagc
720
cgcgcgtctgctggtctacgcgcgagaagtctggaggaagctggtcggagggagc
780
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840
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Val Arg Pro Asn Tyr Pro Tyr Ser Ser Val Lys Thr Arg Leu Pro Pro 50 55 60
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Lys Asp Ala Glu Arg Ala Gly His Glu Pro Met Pro Glu Asp Asp
Met Asn Trp His Val Leu Val Ala Met Ser Gly Gln Val Phe Gly Ala
Gly Asn Cys Gly Glu His Ala Ala Ser Phe Ala Tyr Gly Ala
Leu Ala Gln Glu Ser Gly Arg Ser Pro Arg Glu Lys Ile His Leu Ala
Glu Gin Pro Gly Lys Asp His Val Trp Ala Glu Thr Asp Asn Ser Ser
Gly Ser Ser Pro Ile Val Met Asp Pro Trp Ser Asn Gly Ala Ala
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Arg Thr Tyr Ser Phe Thr Leu Ala Met Ala Ala Glu Ala Gly Lys Val
Ala Arg Glu Thr Ala Glu Asn Val Leu Thr His Thr Ser Arg Leu
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Pro Ser Ser Val Arg Thr Asn Tyr Pro Tyr Ser Ser Val Lys Thr Arg
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Glu Ala Arg Arg Ala Leu Pro Phe Gly Arg Gly Asn Ile Asp Val Asp
Ala Gln Arg Ser Asn Leu Glu Ser Gly Ala Arg Thr Leu Ala Ala Arg
Arg Leu Arg Lys Asp Ala Glu Ala Ala Gly His Glu Pro Met Pro Ala
Asn Glu Asp Met Asn Trp His Val Leu Val Ala Met Ser Gly Glu Val
Phe Gly Ala Gly Asn Cys Gly Glu His Ala Arg Ile Ala Ser Phe Ala
Tyr Gly Ala Leu Ala Gln Glu Gly Arg Asn Ala Asp Glu Thr Ile
His Leu Ala Ala Gln Arg Gly Lys His Val Trp Ala Glu Thr Asp
Aas Ser Ser Ala Gly Ser Ser Pro Val Val Met Asp Pro Trp Ser Asn
Gly Pro Ala Ile Phe Ala Glu Asp Ser Arg Phe Ala Lys Asp Arg Ser
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cagcctgcctg gcgtatcagc aagccaaaatc aggtccgacg acgctttgga attacggtga
atacagatgca aaagacgtcc cttattggctt tggcctttgc aatcctggca gggtgtgggg
agtgatattc agggtgccca ggcagagatg aaaacaccca ttaaagtaga tctggatgcc
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aatgttggtgg gcacagagga aatacctgaac aaggtccgacg acgctttgga attacggtga
atacagatgca aaagacgtcc cttattggctt tggcctttgc aatcctggca gggtgtgggg
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atacagatgca aaagacgtcc cttattggctt tggcctttgc aatcctggca gggtgtgggg
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<210> SEQ ID NO 56
<211> LENGTH: 316
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae pv. delphinii

<400> SEQUENCE: 56

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Ser  Arg  Ile  Thr  Ser  Glu  Asn  Glu  Arg  Arg  Arg  Phe  Gly  Ile  Thr
 20  25  30
Val  Aan  Glu  Met  Glu  Lys  Thr  Ser  Leu  Leu  Ala  Ala  Phe  Ala  Ile
 35  40  45
Leu  Ala  Gly  Cys  Gly  Gly  Ser  Gly  Glu  Ala  Ala  Pro  Gly  Ser  Asp  Ile  Gln
 50  55  60
Gly  Ala  Glu  Ala  Glu  Met  Lys  Thr  Pro  Ile  Lys  Val  Asp  Leu  Asp  Ala
 65  70  75  80
Tyr  Thr  Ser  Lys  Leu  Asp  Ala  Val  Leu  Glu  Ala  Arg  Ala  Aan  Lys
 85  90
Ser  Tyr  Val  Aan  Lys  Gly  Glu  Leu  Ile  Asp  Leu  Val  Ser  Gly  Ala  Phe
100 105 110
Leu  Gly  Thr  Pro  Tyr  Arg  Ser  Aan  Met  Leu  Val  Gly  Thr  Glu  Ile
115 120 125
Pro  Glu  Glu  Leu  Val  Ile  Asp  Phe  Arg  Gly  Leu  Asp  Cys  Phe  Ala  Tyr
130 135 140
Leu  Asp  Tyr  Val  Glu  Ala  Leu  Arg  Arg  Ser  Thr  Ser  Glu  Gln  Asp  Phe
145 150 155 160
Val  Aan  Leu  Val  Glu  Val  Arg  Tyr  Lys  Gly  Gly  Asp  Val  Asp  Phe
165 170 175
Leu Asn Arg Lys His Phe Phe Thr Asp Trp Ala Tyr Gly Thr Thr His
180 185 190
Pro Val Ala Asp Asp Ile Thr Thr Tyr Glu Ile Ser Pro Gly Ala Val Ser
195 200 205
Val Arg Lys Arg Leu Asn Glu Arg Ala Lys Gly Lys Val Tyr Leu Pro
210 215 220
Gly Leu Pro Val Val Glu Arg Ser Met Thr Tyr Ile Pro Ser Arg Leu
225 230 235 240
Val Asp Ser Gln Val Val Ser His Leu Arg Thr Gly Asp Tyr Ile Gly
245 250 255
Ile Tyr Thr Pro Leu Pro Gly Leu Asp Val Thr His Val Gly Phe Phe
260 265 270
Ile Met Thr Asp Lys Gly Pro Val Leu Arg Asn Ala Ser Ser Arg Lys
275 280 285
Glu Asn Arg Lys Val Met Asp Leu Pro Phe Leu Asp Tyr Val Ser Glu
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Lys Pro Gly Ile Val Val Phe Arg Ala Lys Asp Asn
305 310 315

<210> SEQ ID NO 57
<211> LENGTH: 396
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae pv. delphinii

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120
gtacactatt atcgagcga caggtgatcg ggatatctt ccgacgtcat ccgacatattt
180
cgaacgcac gacgtgcgcc gcgtcccttg gcgtcccttc caggtccattt ccgacgtatttt
240
ggcgacgcac gacgtgcgcc gcgtcccttg gcgtcccttc caggtccattt ccgacgtatttt
300
gggtgcgttc gacgacgcac gacgtgcgcc gcgtcccttg gcgtcccttc caggtccattt ccgacgtatttt
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396

<210> SEQ ID NO 58
<211> LENGTH: 131
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae pv. delphinii

<400> SEQUENCE: 58
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Ser Met Pro Asn Leu Pro Asn Lys His Asp Asn Glu Val Tyr Cys
20 25 30
Phe Thr Phe Gln Ser Gly Leu Glu Val Asn Ile Tyr Gln Asp Asp Cys
35 40 45
Arg Trp Val His Phe Ser Ala Thr Ile Gly Gln Phe Gln Asp Ala Ser
50 55 60
Asn Asp Thr Leu Ser His Ala Leu Gln Leu Asn Asn Phe Ser Leu Gly
65 70 75 80
Lys Pro Phe Phe Thr Phe Gly Met Asn Gly Glu Lys Val Gly Val Leu
85 90 95
His Thr Arg Val Pro Leu Ile Glu Met Asn Thr Val Glu Met Arg Lys
100 105 110
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<tr>
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<210> SEQ ID NO 59
<211> LENGTH: 648
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae pv. delphinii

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<210> SEQ ID NO 60
<211> LENGTH: 215
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae pv. delphinii

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Pro Lys Gly Gln Lys Val Pro Val Glu Ile Ile Pro Thr His Asn Asp

Asn Ser Asn Lys Gly Arg Gly

Val Asn Pro Ile His Ala Arg Phe Ser Ser Val Glu Ala Leu Arg His

Ser Asn Val Asp Ile Gln Ala Val Lys Ser Glu Gly Gln Leu Glu Val

Asn Gly Lys Arg Tyr Glu Ile Arg Ala Asa Ala Asp Gly Ser Ile Ala

Val Leu Arg Pro Asp Gln Glu Ser Lys Ala Asp Lys Phe Phe Lys Gly

Ala Ala His Leu Ile Gly Gln Ser Glu Arg Ala Glu Ile Ala Glu

Val Leu Asn Glu Lys Ala Ala Val Pro Leu Aep Arg Met Leu
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gcggctgcca ataacgacct gactgtccag cgttctgaca aacagatggc gatgagcaag
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cagaacagca ggctgcccaa tggccgcttg gtaaacagca gtattttgca atgggtcgaa
aaggcgaaag ccaatggcag cacaagtacc agtgctcttt atcagatcta cgcaaaagaa
cctcccgcgtg tagaactgct gccacgcact gagcaccggg cgtgtctggc gcatatgtat
gtacaggagc tcaaagcaca cggtcaaata gaagtgggtg gcaaatgcta cgacattcgc

gly arg arg phe asp leu glu lys gly gly ser ser ala val gly ala
100 105 110
ala ile lys ala ala asp ser arg leu thr ser lys gly thr phe ala
115 120 125
ser phe gln gln trp ala glu lys ala glu ala leu gly arg asp thr
130 135 140
glu ile gly ile tyr met ile tyr lys arg asp thr pro asp thr thr
145 150 155 160
pro met asn ala ala glu gln glu his tyr leu glu thr leu glu ala
165 170 175
leu asn lys lys asn leu ile arg pro gln ile his asn asp
180 185 190
arg glu glu glu leu asp leu gly tyr ile ala glu asn arg
195 200 205
asn ala arg thr gly phe phe arg met val pro lye asp gln arg ala
210 215 220
pro glu thr asn ser gly arg leu thr ile gly val glu pro lye tyr
225 230 235 240
gly ala gln leu ala leu met ala thr leu met asp lys his lys
245 250 255
ser val thr gln gly lys val val gly pro ala lys tyr gly gln gln
260 265 270
thr asn ser ala ile leu tyr ile asn gly asp leu ala lys ala val
275 280 285
lye leu gly gly leu lye leu ser gly ile pro pro gly lye
290 295 300
phe val glu his thr pro leu ser met gln ser thr gly leu gly leu
305 310 315 320
ser tyr ala glu ser val gly gln pro ser his gly gln ala
325 330 335
arg thr his val ile met asn ala leu lye gly gln gly pro met glu
340 345 350
asn arg leu lys met ala leu ala glu arg gly tyr asp pro glu asn
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pro ala leu arg ala arg asn
370 375
<210> seq id no 63
<211> length: 1149
<212> type: dna
<213> organism: pseudomonas syringae pv. syringae
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gly arg arg phe asp leu glu lys gly gly ser ser ala val gly ala
100 105 110
ala ile lys ala ala asp ser arg leu thr ser lys gly thr phe ala
115 120 125
ser phe gln gln trp ala glu lys ala glu ala leu gly arg asp thr
130 135 140
glu ile gly ile tyr met ile tyr lys arg asp thr pro asp thr thr
145 150 155 160
pro met asn ala ala glu gln glu his tyr leu glu thr leu glu ala
165 170 175
leu asn lys lys asn leu ile arg pro gln ile his asn asp
180 185 190
arg glu glu glu leu asp leu gly tyr ile ala glu asn arg
195 200 205
asn ala arg thr gly phe phe arg met val pro lye asp gln arg ala
210 215 220
pro glu thr asn ser gly arg leu thr ile gly val glu pro lye tyr
225 230 235 240
gly ala gln leu ala leu ala met ala thr leu met asp lys his lys
245 250 255
ser val thr gln gly lys val val gly pro ala lys tyr gly gln gln
260 265 270
thr asn ser ala ile leu tyr ile asn gly asp leu ala lys ala val
275 280 285
lye leu gly gly leu lye leu ser gly ile pro pro gly lye
290 295 300
phe val glu his thr pro leu ser met gln ser thr gly leu gly leu
305 310 315 320
ser tyr ala glu ser val gly gln pro ser his gly gln ala
325 330 335
arg thr his val ile met asn ala leu lye gly gln gly pro met glu
340 345 350
asn arg leu lys met ala leu ala glu arg gly tyr asp pro glu asn
355 360 365
pro ala leu arg ala arg asn
370 375

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<210> SEQ ID NO 64
<211> LENGTH: 382
<212> TYPE: PRT
<213> ORGANISM: Pseudomonas syringae pv. atrofaciens
<400> SEQUENCE: 64
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Ser Glu Val Asp Val Glu Leu Lys Ala His Gly Glu Ile Glu Val
Gly Gly Lys Cys Tyr Asp Ile Arg Ala Ala Asn Asn Asp Leu Thr
Val Glu Arg Ser Asp Lys Glu Met Ala Met Ser Lys Phe Phe Lys
Ala Gly Leu Ser Gly Ser Gly Ser Gly Ser Asp Glu Ile Ala Glu
Val Leu Asn Aasp Lys Arg Gly Ser Ser Val Aasp Val Arg Leu Ile Arg Glu
Gly Glu Thr His Leu Gly Arg Met Gin Phe Asn Ile Glu Glu Gly Glu
Gly Ser Ser Ala Ala Thr Ser Val Glu Asn Ser Aasp Leu Pro Asn Gly
Arg Leu Val Aasp Ser Ile Leu Gln Trp Val Glu Lys Ala Lys Ala
Aasp Gly Ser Thr Ser Thr Ser Ala Leu Tyr Gin Ile Tyr Ala Lys Glu
Leu Pro Arg Val Glu Leu Leu Pro Arg Thr Glu His Arg Ala Cys Leu
 Ala His Met Tyr Lys Leu Aasp Gly Lys Arg Ile Ser Ile Trp Pro
Gln Phe Leu Aasp Gly Val Aasp Gly Leu Glu Leu Lys His Aasp Thr Lys
Val Phe Met Met Aasp Pro Lys Ala Ala Aasp Glu Phe Tyr Lys Ile
Glu Arg Ser Gly Thr Glu Phe Pro Aasp Glu Ala Val Lys Ala Arg Leu
Thr Ile Aasp Val Lys Pro Gin Phe Gin Lys Ala Met Val Aasp Ala
600
660
720
780
840
900
960
1020
1080
1140
1149
Val Arg Leu Thr Ala Glu Arg Arg Asp Ile Ile Thr Ala Lys Val Ala
260 265 270
Gly Pro Ala Lys Ile Gly Thr Ile Thr Asp Ala Asp Val Phe Tyr Val
275 280 285
Ser Gly Asp Phe Ser Ala Ala Gin Thr Leu Ala Lys Glu Leu Gin Ala
290 295 300
Leu Leu Pro Asp Ala Phe Ile Aan His Thr Pro Ala Gly Met Gin
305 310 315 320
Ser Met Gly Lys Gly Leu Cys Tyr Ala Glu Arg Thr Pro Gin Asp Arg
325 330 335
Thr Ser His Gly Met Ser Arg Ala Ser Ile Ile Glu Ser Ala Leu Ala
340 345 350
Asp Thr Ser Arg Ser Leu Glu Lys Leu Arg Asn Ala Phe Lys
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Ser Ala Gly Tyr Asn Pro Asp Asn Pro Ala Phe Arg Leu Glu
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Seq ID NO: 65
Length: 1464
Type: DNA
Organism: Pseudomonas syringae pv. tomato

<400> Sequence: 65

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US 6,852,835 B2

283

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284
ccgttccagc tcaggcgttt gtaa

SEQ ID NO: 66
LENGTH: 487
TYPE: PRT
ORGANISM: Pseudomonas syringae pv. tomato

SEQUENCE: 66

Met His Ile Asn Gln Ser Ala Gln Gln Pro Pro Gly Val Ala Met Glu 1 5 10 15
Ser Phe Arg Thr Ala Ser Asp Ala Ser Leu Ala Ser Ser Ser Val Arg 20 25 30
Ser Val Ser Thr Ser Cys Arg Asp Leu Gln Ala Ile Thr Asp Tyr 35 40 45
Leu Lys His His Val Phe Ala Ala His Arg Phe Ser Val Ile Gly Ser 50 55 60
Pro Asp Glu Arg Asp Ala Leu Ala His Asn Gln Gln Ile Asp Ala 65 70 75 80
Leu Val Glu Thr Arg Ala Asn Arg Leu Tyr Ser Glu Gly Glu Thr Pro 85 90 95
 Ala Thr Ile Ala Glu Thr Phe Ala Lys Ala Glu Phe Asp Arg Leu 100 105 110
Ala Thr Thr Ala Ser Ser Ala Phe Glu Asn Thr Pro Phe Ala Ala Ala 115 120 125
Ser Val Leu Gln Tyr Met Gln Pro Ala Ile Asn Gly Asp Trp Leu 130 135 140
Ala Thr Pro Leu Lys Pro Leu Thr Pro Leu Ile Ser Gly Ala Leu Ser 145 150 155 160
Gly Ala Met Asp Gln Val Gly Thr Lys Met Met Asp Arg Ala Arg Gly 165 170 175
Asp Leu His Tyr Leu Ser Thr Ser Pro Asp Lys Leu His Asp Ala Met 180 185 190
Ala Val Ser Val Lys Arg His Ser Pro Ala Leu Gly Arg Gln Val Val 195 200 205
Aasp Met Gly Ile Ala Val Glu Thr Phe Ser Ala Asn Val Arg 210 215 220
Thr Val Leu Ala Pro Ala Leu Ala Ser Arg Pro Ser Val Gln Gly Ala 225 230 235 240
Val Asp Phe Gly Val Ser Thr Ala Gly Gly Leu Val Ala Asn Ala Gly 245 250 255
Phe Gly Asp Arg Met Leu Ser Val Gln Ser Arg Asp Gln Leu Arg Gly 260 265 270
Gly Ala Phe Val Leu Gly Met Lys Asp Lys Glu Pro Lys Ala Ala Leu 275 280 285
Ser Glu Glu Thr Asp Trp Leu Asp Ala Tyr Lys Ala Ile Lys Ser Ala 290 295 300
Ser Tyr Ser Gly Ala Ala Asn Ala Gly Arg Met Ala Gln Val Leu 305 310 315 320
Pro Leu Asp Val Ala Thr Asp Gly Leu Lys Ala Val Arg Ser Leu Val 325 330 335
Ser Ala Thr Ser Leu Thr Lys Asn Gly Leu Ala Ala Gly Gly Tyr 340 345 350
Ala Gly Val Ser Lys Leu Glu Met Ala Thr Lys Asn Ile Thr Asp 355 360 365
Ser Ala Thr Lys Ala Ala Val Ser Gln Leu Ser Asn Leu Val Gly Ser
Val Gly Val Phe Ala Gly Trp Thr Thr Ala Gly Leu Ala Thr Asp Pro
Ala Val Lys Ala Glu Ser Phe Ile Gln Asp Lys Val Lys Ser Thr
Ala Ser Ser Thr Ser Tyr Val Ala Asp Gin Thr Val Lys Leu Ala
Lys Thr Val Lys Asp Met Ser Gly Glu Ala Ile Ser Ser Thr Gly Ala
Ser Leu Arg Ser Thr Val Asn Leu Arg His Arg Ser Ala Pro Glu
Ala Asp Ile Glu Glu Gly Gly Ile Ser Ala Phe Ser Arg Ser Glu Thr
Pro Phe Gln Leu Arg Arg Leu

<210> SEQ ID NO: 67
<211> LENGTH: 88
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae pv. tomato

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<210> SEQ ID NO: 68
<211> LENGTH: 85
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae pv. syringae

<400> SEQUENCE: 68
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<210> SEQ ID NO: 69
<211> LENGTH: 1065
<212> TYPE: DNA
<213> ORGANISM: Pseudomonas syringae pv. tomato

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acccgtgtca ttcccgcacg tttgttcggg cagaaggcgt ccggcggcaa gctggagatt
ctggtcgagc gcgtgctgga cagccatcgt gtgctggcgc acgtgcgtgc cagcaagtcg
ccaaaagccgg gctcgtcgat cctgatcgat ggcggcggcg aggccgagat ggtggcgcgg
catgacgcgc tgttcgagtt gcgctttgcc gaagaagtgc tgccgttgct ggatcgtgtc
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tatcagaccg tttacgccca gcgcgccggt gctgtggcgg cgccgactgc cggcctgcat
ttcgaccagc cgttgatgga agcaattgcc gccaagggcg tcgagactgc ttttgtcact
cgtgcacgtg gcgcgggtac gttccagccg gtgcgtgtcg agcagatcga agatcaccac
<table>
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<tr>
<th>Codon</th>
<th>Amino Acid</th>
</tr>
</thead>
<tbody>
<tr>
<td>ATG</td>
<td>Met</td>
</tr>
<tr>
<td>GAA</td>
<td>Arg</td>
</tr>
<tr>
<td>GTC</td>
<td>Val</td>
</tr>
<tr>
<td>GGG</td>
<td>Asp</td>
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<td>ACA</td>
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<td>Ala</td>
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<tr>
<td>CAC</td>
<td>Arg</td>
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</table>

**Sequence:**
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ATG GAA GTC GGG ACA GCG CAA GAT TAA TCT GCT CAC
```

**Details:**
- **SEQ ID NO:** 70
- **LENGTH:** 354
- **TYPE:** PRT
- **ORGANISM:** Pseudomonas syringae pv. tomato

**Sequence Description:**
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Met Arg Val Ala Asp Phe Thr Phe Leu Pro Asp Ser Leu Ile Ala
```

**Codon Table:**
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<table>
<thead>
<tr>
<th>Codon</th>
<th>Amino Acid</th>
</tr>
</thead>
<tbody>
<tr>
<td>ATG</td>
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<tr>
<td>CAC</td>
<td>Arg</td>
</tr>
</tbody>
</table>
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**References:**
- US 6,852,835 B2
- **Page:** 289 - 290

**Note:** The sequence and table above cover the nucleotide and amino acid sequences for a specific gene or protein. The table details the codons and their corresponding amino acids, with special attention to the start codon (ATG) and stop codons (TAA, TGA, TAG).
Ser Ala Phe Ala Gly Tyr Pro Glu Thr Met Ala Ala Tyr Ala Ala Ala
305 310 315 320
Ile Glu His Gly Tyr Arg Phe Phe Ser Tyr Gly Asp Ala Met Phe Ile
325 330 335
Thr Arg Aen Pro Ala Pro Thr Ala Pro Gin Glu Ser Ala Pro Glu Asp
340 345 350
His Ala

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<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Description of Artificial Sequence: primer
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agtggtctga ctgaggctgc ga

<210> SEQ ID NO 86
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cccagagcc ggtttttttt g

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gtgccttgat ccaatcctcg
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<210> SEQ ID NO 88
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What is claimed:

1. An isolated protein or polypeptide selected from the group consisting of (i) a protein or polypeptide comprising an amino acid sequence of SEQ ID NO: 7 or SEQ ID NO: 66, and (ii) a protein or polypeptide encoded by a nucleic acid molecule whose complement hybridizes, at a temperature of at least about 37°C, in a medium comprising at most about 0.9M SSC, to a DNA molecule comprising the nucleotide sequence of SEQ ID NO: 6 or SEQ ID NO: 65.

2. The isolated protein or polypeptide according to claim 1, wherein the protein or polypeptide comprises an amino acid sequence of SEQ ID NO: 7 or SEQ ID NO: 66.

3. A composition comprising:
   a carrier and
   a protein or polypeptide according to claim 1.

4. The isolated protein or polypeptide according to claim 1, wherein the protein or polypeptide is a recombinant protein or polypeptide.

5. The isolated protein or polypeptide according to claim 1, wherein the protein or polypeptide is at least about 80% pure.

6. The isolated protein or polypeptide according to claim 1, wherein the protein or polypeptide is at least about 90% pure.

7. The isolated protein or polypeptide according to claim 1, wherein the protein or polypeptide is encoded by a nucleic acid molecule whose complement hybridizes, at a temperature of at least about 37°C, in a medium comprising at most about 0.9M SSC, to a DNA molecule comprising the nucleotide sequence of SEQ ID NO: 6 or SEQ ID NO: 65.

8. The isolated protein or polypeptide according to claim 1, wherein the protein or polypeptide is encoded by a nucleic acid molecule whose complement hybridizes, at a temperature of at least about 42°C, in a medium comprising at most about 0.9M SSC, to a DNA molecule comprising the nucleotide sequence of SEQ ID NO: 6 or SEQ ID NO: 65.

9. The isolated protein or polypeptide according to claim 1, wherein the protein or polypeptide is encoded by a nucleic acid molecule whose complement hybridizes, at a temperature of about 65°C, in a medium comprising at most about 0.9M SSC, to a DNA molecule comprising the nucleotide sequence of SEQ ID NO: 6 or SEQ ID NO: 65.

10. The composition according to claim 3, wherein the protein or polypeptide comprises an amino acid sequence of SEQ ID NO: 7 or SEQ ID NO: 66.

11. The composition according to claim 3, wherein the protein or polypeptide is encoded by a nucleic acid molecule whose complement hybridizes, at a temperature of at least about 37°C, in a medium comprising at most about 0.9M SSC, to a DNA molecule comprising the nucleotide sequence of SEQ ID NO: 6 or SEQ ID NO: 65.

* * * * *