Supplementary Information

CONTENT

Supplementary Materials and Methods

Sequencing of strain *Pseudomonas* sp. CMR5c Bioinformatics Injection assay with *Galleria mellonella* Feeding assay with *Spodoptera littoralis* Bacterial colonization of *Spodoptera littoralis* larvae Identification of biocontrol activity In vitro inhibition of plant pathogens Statistics

Supplementary Results

Phylogeny of sequenced isolates Plant-beneficial effects and antifungal compounds

Supplementary Figures

Figure S1. Phylogenetic tree of the *P. fluorescens* group based on the four-gene MLSA

- scheme of Mulet et al (2012)
- Figure S2. Oral activity against Spodoptera littoralis larvae
- Figure S3. In-vitro inhibition of Pythium ultimum and Fusarium oxysporum
- Figure S4. *P. protegens* CHA0 deficient for a specific phospholipase C, but not for *aprX* or the *reb* cluster is reduced in oral activity against insect larvae

Figure S5. Repetition of experiments depicted in Figure 4 and Supplementary Figure S4.

Supplementary Tables

 Table S1. Full list of gene clusters associated with biocontrol or insecticidal activity for all

strains included in Figure 1. Supplementary to Figure 2.

- Table S2. Strains, plasmids and primers used in this study
- **Table S3.** Mean amino acid identities (AAI) and Genome-to-Genome Distance Calculator(GGDC) values for all genomes related to *P. brassicacearum, P. kilonensis* and *P. thivervalensis*.

Table S4. Oral activity against Plutella xylostella larvae (repetition of experiment depicted in

Figure 3 and Table 2)

Table S5. Biocontrol activity against *Pythium ultimum* on cucumber plants

 Table S6.
 Genomic features

Table S7. Genes specific to insecticidal strains

References

Supplementary Materials and Methods

Sequencing of strain Pseudomonas sp. CMR5c

Paired-end sequence reads of genomic DNA of *Pseudomonas* CMR5c were generated using the Illumina HiSeq2500 system. The *de novo* assembly analysis was performed using the "*de novo* assembly" option of the CLC Genomics Workbench version 7.0.4. The scaffolding analysis was performed using the SSPACE Premium scaffolder version 2.3 (Boetzer et al 2011). Automated gap closure analysis was done using GapFiller version 1.10 (Boetzer and Pirovano 2012). No further manual assembly was performed.

Bioinformatics

Housekeeping genes of sequenced strains were collected from the annotated genomes, cropped to the size of the fragments used for phylogeny and concatenated according to Mulet et al (2012). Alignments were done using Muscle in MEGA v6.0, and a phylogenetic analysis was done with the maximum likelihood method.

Pairwise average amino acid identities (AAI) were calculated in EDGAR (Blom et al 2009). GGDC values are calculated using the Genome-to-Genome Distance Calculator Version 2 and reported according to formula 2, best suited when including draft genomes (Auch et al 2010, Meier-Kolthoff et al 2013).

Injection assay with Galleria mellonella

Washed bacterial cells (10 µl) suspended in 0.9% sterile NaCl solution and adjusted to the desired concentration were injected into the hemolymph of ultimate-instar *Galleria mellonella* larvae (Hebeisen Fishing, Zürich, Switzerland) using a 1-ml syringe with a 27-gauge needle in a repetitive dispensing Tridak Stepper (Intertronic, Oxfordshire, UK). Sterile 0.9% NaCl solution served as control. Three times ten larvae were injected per treatment and kept in Petri dishes at 24°C in the dark. Larvae were scored as live or dead regularly over two days. Mortality was defined as the inability of larvae to react to poking.

Feeding assay with Spodoptera littoralis

Food pellet assays with *Spodoptera littoralis* were performed as described by Ruffner et al. (2013). Briefly, third instar larvae of *S. littoralis* (Syngenta Crop Protection AG, Stein) were exposed to modified insect diet (Gupta et al 2005, Ruffner et al 2013) inoculated with 4×10^7 colony forming units per food pellet. For control treatments, pellets were treated with $10 \mu l$ sterile 0.9% NaCl solution. Instead of using petri dishes (Ruffner et al 2013), pellets were

placed into Greiner six-well plates and presented to one larva per well. Five plates were prepared per bacterial strain (30 larvae per treatment). Larvae were incubated in the dark at room temperature and were fed with fresh, bacteria free diet when necessary. Survival rates were recorded daily. Larvae were considered to be dead when they did not react to repeated poking.

Bacterial colonization of Spodoptera littoralis larvae

Bacteria were extracted from surviving larvae at the end of the experiment. Larvae were surface-disinfested for 30 s in 70% ethanol, rinsed with sterile water and homogenized in 10 ml sterile 0.9% saline solution with a Polytron PT-DA 2112 blender (Kinematica, Littau, Switzerland). Serial dilutions of the resulting homogenate were then plated onto King's B agar (King et al 1954) supplemented with ampicillin (40 μ g ml⁻¹), chloramphenicol (13 μ g ml⁻¹), and cycloheximide (100 μ g ml⁻¹) to select for the bacterial strains fed to the larvae. Plates for bacterial quantification were incubated for two days at 27°C before colony counting.

Identification of biocontrol activity

Biocontrol of *Pythium* damping-off of cucumber was assessed for each strain adapted after Sharifi-Tehrani et al (1998). Aliquots of 200 μ l of over-night cultures of bacteria grown in LB were plated on King's B agar (King et al 1954) and incubated for 24 h at 24°C. Bacteria were then scraped off the plate and washed in sterile distilled H₂O. Each bacterial strain was added to five pots filled with 120 g of TREF go PP7000 plant substrate (Gvz Rossat AG, Otelfingen, Switzerland) to a final concentration of 5 × 10⁷ cfu per g soil. Each pot was inoculated with 0.3 g of *Pythium* inoculum grown on millet seeds and planted with three pre-germinated cucumber seeds. Pots were incubated at 70% humidity for 16 h with light (15 klux) at 22°C, followed by an 8-h dark period at 20°C. After 12 d, shoot weight per pot was recorded. Biocontrol activity was calculated after Rezzonico et al (2007) as:

 $(1 - ((W_c - W_I)/(W_c - W_P))) \times 100$

using shoot weight obtained in the control with neither bacterial nor pathogen inoculum (W_c) , in the unprotected control with the pathogen alone (W_P) and in presence of the tested bacterial strain and the pathogen (W_I) . A total of seven experiments was conducted and each strain was tested at least twice. The model strain *P. protegens* CHA0 was included as a reference in each experiment.

In vitro inhibition of plant pathogens

F. oxysporum Schlecht. f. sp. *radicis-lycopersici* strain Forl22 (Forl) and *P. ultimum* Trow strain 67-1 (Pu) were cultivated on malt agar as described by Sharifi-Tehrani et al (1998). In vitro inhibition of Pu and Forl was assessed on malt agar (MA) and GCY (Tambong and Höfte 2001) plates. Mycelial plugs were placed at the center of the plates either one day before (for Pu) or one day after (for Forl) adding the bacteria. Bacterial strains were grown overnight in LB. Cells were washed with sterile distilled H_2O and a suspension of an OD_{600} of 1.0 was prepared. The suspension was streaked out in a square around the mycelial plug using an inoculation loop. Plates were inoculated at 24°C. The mycelial diameter was measured after 2 d for Pu and after 8 d for Forl. Each strain was tested twice on each medium with four replicates.

Statistics

Data analysis was performed in R version 3.1.1. (http://www.r-project.org). Mortality rates of the insect toxicity tests with wild-type strains and data on in-vitro inhibition of plant pathogens were analysed by multiple comparisons using Kruskal-Wallis adjusted by Bonferroni-Holm. LT₅₀ values were estimated based on the generalized linear model using the MASS package in R (Venables and Ripley 2002). To test for significant differences in insect toxicity tests between *P. protegens* CHA0 and its mutant strains the Log-Rank test of the Survival package of R was used. To identify strains with significant biocontrol activity, a t-test was performed testing each strain against the respective unprotected control with pathogen alone.

Supplementary Results

Phylogeny of sequenced isolates

Many strains of the *Pseudomonas fluorescens* group were classified years ago, and their taxonomic status was not updated since then. We performed a comparative systematic study to correctly assign the isolates that were sequenced in this study to the phylogeny within the genus *Pseudomonas* (Mulet et al 2012). A recent study (Gomila et al 2015) has performed similar work with other published genomes, some of which were also included in our study.

The core genome tree (Figure 1), practically a core genome Multilocus Sequence Analysis (MLSA) (Blom et al 2009), confirmed the phylogenetic position of a range of isolates that we included in sequencing. For publicly available genomes, the position is corresponding to the study of Gomila et al (2015), whereas we confirmed the position of some isolates that already had a unconfirmed status in phylogeny. The phylogenetic position of the isolates could now be confirmed using digital DNA-DNA hybridization (Supplementary Table S3)(Meier-Kolthoff et al 2014), average amino acid identities (Supplementary Table S3) of the core genome (Konstantinidis et al 2006) and the four-gene MLSA (Supplementary Figure S1) (Gomila et al 2015, Mulet et al 2012).

Five strains that were sequenced in this study are now included in *P. protegens* (Ramette et al 2011): strains CHA0^T, PGNR1, BRIP, K94.41 and PF, while two isolates (PCL1391 and CD), already included in *P. chlororaphis* (Chin-A-Woeng et al 1998, Ruffner et al 2015), can now be assigned to the respective subspecies as we also included three of the subspecies type strains in the genome analysis. Strain PCL1391 belongs to *P. chlororaphis* subsp. *piscium*, whereas strain CD is a *P. chlororaphis* subsp. *aureofaciens*. By searching the annotations for genes coding for the differential phenotypes as described in literature (Burr et al 2010, Peix et al 2007), we could confirm these designations.

Using the data generated in this study, strain TM1A3 is confirmed as member of the species *P. brassicacearum* by its relationship to type strain *P. brassicacearum* subsp. *brassicacearum* NFM421^T, but this strain cannot yet be assigned to a subspecies, as MLSA or genomic data for the other subspecies are missing. Based on AAI and DNA-DNA hybridization strains P12 and PITR2 are closely related to the type strains of *P. kilonensis and P. thivervalensis*, respectively, which were also sequenced in this study and can thus be assigned to these species (Supplementary Table S3).

We cannot assign a species name to the other species. Three strains (Q12-87, P97.38 and Pf153) belong to the *P. corrugata* subgroup, while *Pseudomonas* sp. MIACH is included in

the *P. fluorescens* subgroup (Supplementary Figure 1). This is in agreement with the core genome tree (Figure 1). *Pseudomonas* sp. P1.8 is, based on the MLSA, belonging to the *P. jessenii* subgroup, while *Pseudomonas* sp. P1.31 is a member of the *P. koreensis* subgroup. The closest related genome-sequenced strains included in Figure 1 were also assigned to the corresponding subgroups (Gomila et al 2015). However, none of these can be assigned to a known *Pseudomonas* species, indicating that these strains represent novel species within the genus.

Plant-beneficial effects and antifungal compounds

In a pot experiment all strains were tested for their biocontrol activity against the oomycete pathogen *Pythium ultimum* on cucumber roots. In all sub-clades, several strains were found to display effective plant protection whereas others had no significant biocontrol activity. Thus, biocontrol activity seems to be phylogenetically less predictable than insecticidal activity. The presence of biosynthetic genes for the two important antifungal metabolites 2,4-diacetylphloroglucinol (DAPG) (*phl*) and phenazine (Phz) (*phz*) was not necessarily linked to *P. ultimum* biocontrol since strains *P. chlororaphis* subsp. *aureofaciens* LMG 5004 (*phz*⁺), *P. kilonensis* DSM 13647^T (*phl*⁺), and *Pseudomonas* sp. CMR5c (*phz*⁺, *phl*⁺) did not provide any protection against the root pathogen (Figure 2). In contrast, all *P. protegens* strains displayed significant biocontrol activity (Keel et al 1996, Perneel et al 2007) did not protect cucumber plants against *P. ultimum* might be explained by the different experimental conditions used in this study, such as the plant as well as the pathogen species or the substrate.

Similar to the results on insecticidal activity, no connection between the original habitat and the degree of plant protection was observed. Thus, also the strains *P. chlororaphis* subsp. *piscium* DSM 21509^T or *P. protegens* BRIP recently isolated from fish and cyclops, respectively, provided significant biocontrol activity (Supplementary Table S5). In general, antifungal metabolite production appears to be less an adaptation to life on roots than a universal defence mechanism against microbial competitors. For instance the fish isolate *P. chlororaphis* subsp. *piscium* DSM 21509^T was also found to have *in vitro* activity towards the oomycete fish pathogen *Saprolegnia parasitica*, which causes significant losses in fish hatcheries and breeding units (data not shown). Thus, an isolate from a certain habitat could also be used as biocontrol agent in a completely different ecological context.

7

A subset of 15 strains of sub-clade 1 and 2 was further tested for *in vitro* inhibition of mycelial growth of *P. ultimum* and a second plant pathogen, *Fusarium oxysporum*, on MA and GCY medium, favouring the production of DAPG or Phz, respectively (Figure 2, Supplementary Figure S3). Throughout both phylogenetic groups, all strains except P1.8 were found to exhibit *in vitro* pathogen inhibition with DAPG and Phz producing strains performing best on media conducive to metabolite biosynthesis.

Supplementary Figures



Supplementary Figure S1: Maximum Likelihood phylogenetic tree of strains belonging to the *P*. *fluorescens* group based on the four-gene (16S rRNA, *gyrB*, *rpoB* and *rpoD*) MLSA scheme of Mulet et al (2012). Strains investigated in this study are indicated in bold. Bootstrap values over 50% are indicated in the tree.



Supplementary Figure S2. Sub-clade 1 strains cause lethal oral infections in *Spodoptera littoralis* larvae while sub-clade 2 strains, although some of them are able to persist in the insect, do not kill the larvae.

Survival (A) and colonization (B) rates of *S. littoralis* larvae upon feeding on artificial diet inoculated with either 4×10^7 cells of the indicated *Pseudomonas* strains or 0.9% NaCl (control). A) Survival of larvae after 5 d. Bars show means (± se) of five replicates with six larvae each. Asterisks indicate bacterial treatments that were significantly different from the control based on multiple comparisons by Kruskal-Wallis adjusted by Bonferroni-Holm (p ≤ 0.05). Each strain was tested in an independent second experiment with highly similar results.

B) Some strains of sub-clade 2 are able to persist in *S. littoralis* larvae whereas numbers of others strongly decline within a few days. To get an estimate of the capacity of inoculants to persist and multiply within *S. littoralis* larvae upon ingestion, six surviving larvae were extracted and colonization levels were assessed by plating serial dilutions on selective medium at the end of the experiment. Data derived from two independent experiments. Strains of sub-clade 1, here represented by *P. protegens* CHA0, generally multiply to levels of about 10^8 cfu/larva. Colonization levels in control larvae represent bacterial background levels on King's B agar (King et al 1954) supplemented with ampicillin (40 µg ml⁻¹), chloramphenicol (13 µg ml⁻¹), and cycloheximide (100 µg ml⁻¹).

10



Supplementary Figure S3. Inhibition of mycelial growth of *Pythium ultimum* and *Fusarium* oxysporum on MA and GCY medium. Bacteria were streaked out in a square around a plug of oomycete/fungal mycelium and mycelial diameter was measured after 2 days for *P. ultimum* A) and 8 days for *F. oxysporum* B). Strains with different letters were significantly different from each other based on multiple comparisons by Kruskal-Wallis adjusted by Bonferroni-Holm ($p \le 0.05$). Each strain was tested in an independent second experiment with highly similar results.



Supplementary Figure S4. Deletion of *plcN* (encoding phospholipase C), but not of *aprX* or the *reb* cluster, reduces oral activity of *P. protegens* CHA0 against insect larvae.

A, C) Systemic activity against *Galleria mellonella*. 30 larvae per treatment were injected with 2 x 10³ bacterial cells and survival was recorded every hour.

B, D) Oral activity against *Plutella xylostella*. Larvae were fed on artificial diet inoculated with 4 x 10⁶ bacterial cells.

B) The virulence of the phospholipase C-negative ($\Delta plcN$) mutant was slightly reduced compared to the wild type strain (p-value<0.01, Log-Rank test, Survival Package in R). Although the effect was not significant in all experiments, the tendency of slower killing was always observed. Each mutant was tested at least three times with similar results. One repetition is depicted in Supplementary Figure S5. CHA0, wild type; $\Delta plcN$, phospholipase *C*-negative mutant; $\Delta gacA$, GacA-negative mutant; $\Delta rebB1-3$, mutant for the *rebB*-cluster; $\Delta aprX$, metallopeptidase AprX-negative mutant; 0.9% NaCl served as negative control.



Supplementary Figure S5. Repetition of experiments depicted in Figure 4 and Supplementary Figure S4.

Oral activity against *Plutella xylostella*. Larvae were exposed to artificial diet inoculated with 4×10^{6} bacterial cells.

A) Significant differences according to a Log-Rank test (Survival Package in R) between treatments with the wild type CHA0 and the chitinase C-negative mutant are indicated with ***(p-value <0.0001). CHA0, wild type; $\Delta chiC$, chitinase C-negative mutant; $\Delta gacA$, GacA-negative mutant; $\Delta plcN$, phospholipase C-negative mutant; 0.9% NaCl served as negative control.

Supplementary Tables

Supplementary Table S1. Full list of gene clusters associated with biocontrol or insecticidal activity for all strains shown in Figure 1. Supplementary to overview Figure 2.

							Gen	e or i	meta	bol	lite name (Locus t)					
		fit	chiC	chitinase	plcN	aprX	aprA	- psl	rebB		DAPG	PCA	PCN	2-OH-PCA	HCN	PRN	PLT	HPR	rhizoxin	CLP
Accession number	Strain	PFL_2980- PFL_2987	PCL1391_1854- PCL1391_1855	PCL1391_3057-	°CL1391_2966	°CL1391_2141	°CL1391_3021	PCL1391_4983,498 1994	CL1391_0072, 0073, 0075, 0076		PFL_5951- PFL_5958	CL1391_4880- CL1391_4888	°CL1391_4889	ochIO6_5227	PFL_2577- PFL_2579	PFL_3604- PFL_3607	PFL_2784- PFL_2800	^o chIO6_4242- ^o chIO6_4244	PFL_2989- PFL_2997	PFL_2145- PFL_2147
CP003190.1	P. protegens CHAOT	+	+	-	+	+	+	+	+		+	-	-	-	+	+	+	-	-	+
LHUV0000000	P. protegens PGNR1	+	+		+	+	+	+	+		+	-	-	-	+	+	+	-	-	+
LHUW0000000	P. protegens BRIP	+	+	-	+	+	+	+	+		+	-	-	-	+	+	+	-	-	+
LHUU00000000	P. protegens K94.41	+	+	-	+	+	+	+	+		+	-	-	-	+	+	+	-	-	+
CP000076.1	P. protegens Pf-5	+	+	-	+	+	+	+	+		+	-	-	-	+	+	+	-	+	+
LHUX0000000	P. protegens PF	+	+	-	+	+	+	+	+		+	-	-	-	+	+	+	-	+	+
AP014522.1	P. protegens Cab57	+	+	-	+	+	+	+	+		+	-	-	-	+	+	+	-	-	+
LHUY0000000	Pseudomonas sp. CMR5c	+	+	-	+	+	+	+	+		+	+	+	-	+	+	+	-	-	+
LHUZ0000000	P. chlororaphis subsp. piscium JF3835T	+	+	+	+	+	+	+	+		-	+	+	-	+	-	-	-	-	+
ATBG0000000	P. chlororaphis HT66	+	+	+	+	+	+	+	+		-	+	+	-	+	-	-	-	-	+
LFUT0000000	P. chlororaphis subsp. piscium PCL1391	+	+	+	+	+	+	+	+		-	+	+	-	+	-	-	-	-	+
AYUD0000000.1	P. chlororaphis subsp. aurantiaca PB-St2	±	+	+	+	-	+	+	±		-	+	-	+	+	+	-	+	-	-
CP009290.1	P. chlororaphis subsp. aurantiaca JD37	±	+	+	+	+	+	+	+			+	-	+	+	+	-	+	-	-
CP008696.1	P. chlororaphis PA23	+	+	+	+	+	+	+	+		-	+	-	+	+	+	-	+	-	-
LHVB0000000	P. chlororaphis CD	+	+	+	+	+	+	+	+		-	+	-	+	+	+	-	+	-	-
LHVA0000000	P. chlororaphis subsp. aureofaciens LMG 1245T	+	+	±	+	+	+	+	+		-	+	-	+	+	+	-	+	-	-
AWWJ0000000	P. chlororaphis YL-1	+	+	+	+	+	+	+	±		-	+	-	+	+	+	-	+	-	-
CM001490.1	P. chlororaphis O6	+	+	+	+	+	+	+	+		1	+	-	+	+	+	-	+	-	
CM001559.1	P. chlororaphis subsp. aureofaciens 30-84	+	+	+	+	+	+	+	+		-	+	-	+	+	+	-	+	-	
LHVC00000000	P. chlororaphis subsp. chlororaphis LMG 5004T	+	+	+	+	+	+		+			+	_+		+	+		+		
AF0Y0000000	Pseudomonas sp. HK44	<u> </u>	L				<u> </u>		<u> </u>				<u> </u>							
LHVD00000000	P. brassicacearum TM1A3	- 7					+	-	-		+	1	-	-	+	-	-	-		-
AZOC00000000.1	P. brassicacearum 51MFCVI2.1		-	-	-	-	-	-	-		-	1	-	-	+	-	-	-	-	-
CP002585.1	P. brassicacearum subsp. brassicacearum NFM421		-	-	-	-	+	-	-		+	1	-	-	+	-	-	-	-	-
AHP000000000	P. brassicacearum Q8r1-96		-	-	-	-	+	-	-		+	1	-	-	+	-	-	-	-	-
	P. kilonensis DSWI 136471		-	-	-	-	1	-	-		÷.		-	-	÷.	-	-	-	-	-
CD0031E0.1	P. KIIOIIEIISIS P12	-	-	-	-	-	Ť	-	-		Ť	-	-	-	Ť	-	-	-	-	-
LHVE0000000	P thivenuclensis DSM12194T	-	-	-	-	-	Ţ	-	-			-	-	-	1	-	-	-	-	-
LHVE0000000	P thivervalensis DITR?						1				1				11					
LHV10000000	Pseudomonas sp. 012-87						1				1				11					
AGBM0000000	Pseudomonas sp. Q12 07		_	_		-	÷.	-			÷			-	÷	-	-		_	
CP007410.1	Pseudomonas sp. Q2 of	-	-	-	-	-	+	- I	-		÷.		-	-	+	-	-	-	-	-
LHVJ00000000	Pseudomonas sp. P97.38	-	-	-	-	-	+	-	-		+	-	-	-	+	-	-	-	-	-
ATKI0000000	P. corrugata CFBP5454	-	-	-	-	-	+	-	-		-	-	-	-	+	-	-	-	-	-
LHVK00000000	P corrugata DSM7228 T	-	-	-	-	-	+	-	-		-	-	-	-	+	-	-	-	-	-
AUPB00000000.1	P. mediterranea CFBP 5447	-	-	-	-	-	+	-	-		-	-	-	-	+	-	-	-	-	-
LHVL0000000	Pseudomonas sp. Pf153	-	-	-	-	-	+	-	-		-	-	-	-	+	-	-	-	-	-
CP003880.1	Pseudomonas sp. UW4	-	-	-	-	-	-	-	-		-	-	-	-	-	-	-	-	-	-
LHVM0000000	Pseudomonas sp. P1.8	-	-	-	-	-	-	-	-		-	-	-	-	-	-	-	-	-	-
ARLP00000000	P. mandelii 36MFCvi1.1	±	-	-	-	-	+	-	-		-	-	-	-	+	-	-	-	-	-
AZQQ0000000	P. mandelii PD30	-	-	-	-	-	+	-	-		-	-	-	-	-	-	-	-	-	-
CP005960.1, CP005961.1	P. mandelii JR-1	-	-	+	-	-	+	-	-		-	-	-	-	+	-	-	-	-	-
CP000094.2	Pseudomonas sp. Pf0-1	-	-	+	-	+	+	-	-		-	-	-	-	+	-	-	-	-	+
LHVN0000000	Pseudomonas sp. P1.31	-	-	-	-	-	+	-	-		-	-	-	-	+	-	-	-	-	
ALYL0000000	Pseudomonas sp. R124	-	-	+	-	+	+		-		-	-	-	-	+	-	-	-	-	+
CP008896.1	Pseudomonas sp. UK4	-	-	-	-	-	+	+	-		-	-	-	-	-	-	-	-	-	-
JENC0000000.1	Pseudomonas sp. ATCC 17400	-			-		+	+	-				-	-	-	-	-	-		-
AKXH0000000	Pseudomonas sp. BBc6R8		-	-	+	-	+	±	-		1	1	-	-	-	-	-	-	-	-
CP004045.1	P. poae RE*1-1-14			- 7			+	±	-				-	-		-	-	-		+
ANIZW0100000	P. pode BRIP34879	-	-			-	+	±	-				-	-	-	-	-	-	-	+
AUUH0000000	P. Veronii 1YdB1EX2	±	-			-	+	-	-				-	-	-	-	-	-	-	-
LP006852.1	Pseudomonas sp. TKP		-	-	-	-	+	±	-				-	-	-	-	-	-	-	-
	Pseudomonas so MIACH			Ť				+												+
NC 013660 1	Pseudomonas sp. SPW25		-	-	-	-	Ţ	÷	-					-	-	-	-		-	
AV0600000000	Pseudomonas sp. 56W25						+	+												
CM001025.1	Pseudomonas sp. WH6	-	-		-	-	+	Ť.	-					-	-			-	-	-
AMZG0000000.1	Pseudomonas sp. BS2	-	-	-	-	-	+	-	-				+	-	-	-	-	-	-	-
CM001514.1	P. synxantha BG33R	-	-	+	-	-	+	±	±		-	-	-	-	-	-	-	-	-	+
CM001513.1	Pseudomonas sp. SS101	-	-	+	-	-	+	±	±		-	-	-	-	-	-	-	-	-	+
AOJA00000000	Pseudomonas sp. FH5	-	-	+	-	-	+	±	±		-	-	-	-	-	-	-	-	-	-
NC_017911.1; NC_021361.1	Pseudomonas sp. A506	-	-	+	-	-	+	±	±		-	-	-	-	-	-	-	-	-	-
AHZX00000000.1	P. fragi B25		-	-		+	-		-		-		-	-	±		-	-		

Loci were defined as being present when showing 70% similarity over 70% of gene length to the loci indicated in the table. As none of the strains harbors all loci, three different reference strains (PCL1391, Pf-5, O6) were used.

+, gene/s present; -, gene/s absent; ±, gene cluster partially present

P. fluorescens insecticidal toxin-cluster (*fit*); chitinase C (*chiC*); phospholipase C (*plcN*); metallopeptidase *aprX* (*aprX*); alkaline metalloprotease *aprA* (*aprA*); *rebB*-cluster (*rebB*); *psl*-cluster (*psl*); 2,4-diacetylphloroglucinol (DAPG); phenazines: phenazine-1-carboxamide (PCN), phenazine-1-carboxylic acid (PCA), 2-hydroxy-PCA (2-OH-PCA); hydrogen cyanide (HCN); pyrrolnitrin (Prn), pyoluteorin (Plt); 2-hexyl-5-propyl-alkylresorcinol (HPR); cyclic lipopeptide (CLP)

As most genomes consist of several contigs, genes might be found to be absent in a certain strain although they are in fact present, but are located at the border of contigs.

Name	Relevant characteristics ¹ or sequence $(5' \rightarrow 3')^2$	Reference or comment
Pseudomonas		
protegens		
CHA0	Wild type	(Jousset et al 2014, Stutz et al 1986)
CHA5099	Δ <i>chiC</i> (deletion of PFLCHA0_c21380)	This study
CHA5221	Δ <i>rebB1-3</i> (deletion of PFLCHA0_c01820 through PFLCHA0_c01860)	This study
CHA5222	$\Delta a pr X$ (deletion of PFLCHA0_c25470)	This study
CHA5223	Δ <i>plcN</i> (deletion of PFLCHA0_c31570)	This study
Escherichia coli		
DH5α, DH5α λpir	Laboratory strains	(Sambrook and Russel 2001)
Plasmids		
pEMG	pSEVA212S; <i>ori</i> R6K, <i>lacZα</i> MCS flanked by two I-SceI sites; Km ^r , Ap ^r	(Martinez-Garcia and de Lorenzo 2011)
pME8327	pEMG-Δ <i>chiC</i> ; suicide plasmid for the in-frame deletion of PFLCHA0_c21380 (<i>chiC</i>) in CHA0; Km ^r	This study
pME11026	pEMG-Δ <i>rebB1-3</i> ; suicide plasmid for the deletion of the PFLCHAO_c01820 to PFLCHA0_c01860 region (<i>rebB1-3</i> cluster) in CHA0; Km ^r	This study
pME11027	pEMG-Δ <i>aprX</i> ; suicide plasmid for the in-frame deletion of PFLCHA0_c25470 (<i>aprX</i>) in CHA0; Km ^r	This study
pME11028	pEMG-Δ <i>plcN</i> ; suicide plasmid for the in-frame deletion of PFLCHA0_c31570 (<i>plcN</i>) in CHA0; Km ^r	This study
pSW-2	oriRK2, xylS, P _m ::I-scel; Gm ^r	(Martinez-Garcia and de Lorenzo 2011)
Primers		
aprX-del-1	G <u>GAATTC</u> GATGGGCCTGTTCTGAGAGG, EcoRI	Deletion of CHA0 aprX
aprX-del-2	CCC <u>AAGCTT</u> TGCTTCCGAGAGTGCTTTTGAC, HindIII	Deletion of CHA0 aprX
aprX-del-3	CCC <u>AAGCTT</u> AGCCTGATGATCGACCTGAC, HindIII	Deletion of CHA0 aprX
aprX-del-4	CG <u>GGATCC</u> TACCAGCAGTTCTGCAACCAG, BamHI	Deletion of CHA0 aprX
chiD-1	CG <u>GAATTC</u> GCCACAGGCTCAACTAAAACAT, EcoRI	Deletion of CHA0 chiC
chiD-2	GG <u>GGTACC</u> AATGCTCGGCATCAGGGAAGCA, Kpnl	Deletion of CHA0 chiC
chiD-3	GG <u>GGTACC</u> CATGGCTGAGTTGTGACGGCCA, Kpnl	Deletion of CHA0 chiC
chiD-4	CG <u>GGATCC</u> CGCTTACCAATGATTACAACTG, BamHI	Deletion of CHA0 chiC
plcC-del-1	G <u>GAATTC</u> ATAACGCCACCCATTTCAGC, EcoRI	Deletion of CHA0 plcN
plcC-del-2	CCC <u>AAGCTT</u> ACTGGGCATGGGTTATTGAGTC, HindIII	Deletion of CHA0 plcN
plcC-del-3	CCCAAGCTTGCATGAAGACCTTGGCAAAAATG, HindIII	Deletion of CHA0 plcN
plcC-del-4	CG <u>GGATCC</u> GGCCTATGCACGAAAGTTGT, BamHI	Deletion of CHA0 plcN
reb-del-1	G <u>GAATTC</u> GTATTGCCCGGTTTGCAGC, EcoRI	Deletion of CHA0 reb cluster
reb-del-2	CCC <u>AAGCTT</u> ACTGGGCATGGGTTATTGAGTC, HindIII	Deletion of CHA0 reb cluster
reb-del-3	CCC <u>AAGCTT</u> GCATGAAGACCTTGGCAAAAATG, HindIII	Deletion of CHA0 reb cluster
reb-del-4	CG <u>GGATCC</u> CGCTTACCAATGATTACAACTG, BamHI	Deletion of CHA0 reb cluster

Supplementary Table S2. Strains, plasmids and primers used in this study

¹ Ap^r, ampicillin; Gm^r, gentamicin; and Km^r, kanamycin resistance, respectively. ² Specified restriction sites are underlined.

Supplementary Table S3. Mean amino acid identities (AAI) and Genome-to-Genome Distance Calculator (GGDC) values for all genomes related to *P. brassicacearum*, *P. kilonensis* and *P. thivervalensis*.

Α	AAI values												
		1	2	3	4	5	6	7	8	9	10	11	12
1	<i>P. brassicacearum</i> subsp. <i>brassicacearum</i> NFM 421^{T}		99.82	99.81	99.85	97.74	97.74	97.82	96.44	96.39	94.67	94.67	94.87
2	P. brassicacearum TM1A3	99.82		99.79	99.84	97.73	97.73	97.82	96.44	96.38	94.66	94.67	94.85
3	P. brassicacearum 51MCFVI21	99.81	99.79		99.81	97.72	97.72	97.78	96.42	96.37	94.66	94.65	94.84
4	Pseudomonas sp. Q8r1-96	99.85	99.84	99.81		97.73	97.73	97.82	96.43	96.38	94.66	94.65	94.86
5	P. kilonensis DSM 13647 ^T	97.74	97.73	97.72	97.73		99.49	98.33	96.61	96.59	94.54	94.54	94.73
6	P. kilonensis P12	97.74	97.73	97.72	97.73	99.49		98.33	96.61	96.57	94.50	94.50	94.72
7	Pseudomonas sp. F113	97.82	97.82	97.78	97.82	98.33	98.33		96.62	96.58	94.58	94.58	94.71
8	P. thivervalensis DSM 13194 ^T	96.44	96.44	96.42	96.43	96.61	96.61	96.62		99.48	94.43	94.43	94.41
9	P. thivervalensis PITR2	96.39	96.38	96.37	96.38	96.59	96.57	96.58	99.48		94.40	94.40	94.37
10	Pseudomonas sp. Q12-87	94.67	94.66	94.66	94.66	94.54	94.50	94.58	94.43	94.40		99.79	94.48
11	Pseudomonas sp. Q2-87	94.67	94.67	94.65	94.65	94.54	94.50	94.58	94.44	94.40	99.79		94.47
12	Pseudomonas sp. DF41	94.87	94.85	94.84	94.86	94.73	94.72	94.71	94.41	94.37	94.48	94.47	
-													
в	GGDC values		•			-		-	•	•	40		40
В	GGDC values	1	2	3	4	5	6	7	8	9	10	11	12
B 1	GGDC values <i>P. brassicacearum</i> subsp. <i>brassicacearum</i> NFM 421^{T}	1	2 96.0	3 96.9	4 96.6	5 59.1	6 59.1	7 61.0	8 46.2	9 45.9	10 36.5	11 36.7	12 38.6
B 1 2	GGDC values <i>P. brassicacearum</i> subsp. <i>brassicacearum</i> NFM 421 ^T <i>P. brassicacearum</i> TM1A3	1 96.0	2 96.0	3 96.9 96.3	4 96.6 96.1	5 59.1 59.2	6 59.1 59.1	7 61.0 60.9	8 46.2 46.1	9 45.9 46.0	10 36.5 36.3	11 36.7 36.4	12 38.6 38.4
B 1 2 3	GGDC values P. brassicacearum subsp. brassicacearum NFM 421 [™] P. brassicacearum TM1A3 P. brassicacearum 51MCFVI21 P. documentary of the second seco	1 96.0 96.8	2 96.0 96.3	3 96.9 96.3	4 96.6 96.1 97.0	5 59.1 59.2 59.1	6 59.1 59.1 59.2	7 61.0 60.9 61.1	8 46.2 46.1 46.2	9 45.9 46.0 46.0	10 36.5 36.3 36.5	11 36.7 36.4 36.5	12 38.6 38.4 38.4
B 1 2 3 4	GGDC values <i>P. brassicacearum</i> subsp. <i>brassicacearum</i> NFM 421 ^T <i>P. brassicacearum</i> TM1A3 <i>P. brassicacearum</i> 51MCFVI21 <i>Pseudomonas</i> sp. Q8r1-96	1 96.0 96.8 96.6	2 96.0 96.3 96.1	3 96.9 96.3 97.0	4 96.6 96.1 97.0	5 59.1 59.2 59.1 59.0	6 59.1 59.2 58.9	7 61.0 60.9 61.1 60.9	8 46.2 46.1 46.2 46.1	9 45.9 46.0 46.0 46.0	10 36.5 36.3 36.5 36.6	11 36.7 36.4 36.5 36.7	12 38.6 38.4 38.4 38.5
B 1 2 3 4 5	GGDC values <i>P. brassicacearum</i> subsp. <i>brassicacearum</i> NFM 421^{T} <i>P. brassicacearum</i> TM1A3 <i>P. brassicacearum</i> 51MCFVI21 <i>Pseudomonas</i> sp. Q8r1-96 <i>P. kilonensis</i> DSM 13647 ^T	1 96.0 96.8 96.6 59.1	2 96.0 96.3 96.1 59.2	3 96.9 96.3 97.0 59.1	4 96.6 96.1 97.0	5 59.1 59.2 59.1 59.0	6 59.1 59.2 58.9 88.5	7 61.0 60.9 61.1 60.9 64.5	8 46.2 46.1 46.2 46.1 47.4	9 45.9 46.0 46.0 46.0 47.0	10 36.5 36.3 36.5 36.6 36.4	11 36.7 36.4 36.5 36.7 36.5	12 38.6 38.4 38.4 38.5 38.8
B 1 2 3 4 5 6	GGDC values <i>P. brassicacearum</i> subsp. <i>brassicacearum</i> NFM 421 ^T <i>P. brassicacearum</i> TM1A3 <i>P. brassicacearum</i> 51MCFVI21 <i>Pseudomonas</i> sp. Q8r1-96 <i>P. kilonensis</i> DSM 13647 ^T <i>P. kilonensis</i> P12	1 96.0 96.8 96.6 59.1 59.1	2 96.0 96.3 96.1 59.2 59.1	3 96.9 96.3 97.0 59.1 59.2	4 96.6 96.1 97.0 59.0 58.9	5 59.1 59.2 59.1 59.0 888.5	6 59.1 59.2 58.9 88.5	7 61.0 60.9 61.1 60.9 64.5 64.5	8 46.2 46.1 46.2 46.1 47.4 47.2	9 45.9 46.0 46.0 46.0 47.0 46.9	10 36.5 36.3 36.5 36.6 36.4 36.5	11 36.7 36.4 36.5 36.7 36.5 36.5	12 38.6 38.4 38.4 38.5 38.8 38.3
B 1 2 3 4 5 6 7	GGDC values <i>P. brassicacearum</i> subsp. <i>brassicacearum</i> NFM 421 ^T <i>P. brassicacearum</i> TM1A3 <i>P. brassicacearum</i> 51MCFVI21 <i>Pseudomonas</i> sp. Q8r1-96 <i>P. kilonensis</i> DSM 13647 ^T <i>P. kilonensis</i> P12 <i>Pseudomonas</i> sp. F113	1 96.0 96.8 96.6 59.1 59.1 61.0	2 96.0 96.3 96.1 59.2 59.1 60.9	3 96.9 96.3 97.0 59.1 59.2 61.1	4 96.6 96.1 97.0 59.0 58.9 60.9	5 59.1 59.2 59.1 59.0 88.5 64.5	6 59.1 59.2 58.9 88.5 64.5	7 61.0 60.9 61.1 60.9 64.5 64.5	8 46.2 46.1 46.2 46.1 47.4 47.2 46.7	9 45.9 46.0 46.0 46.0 47.0 46.9 46.3	10 36.5 36.3 36.5 36.6 36.4 36.5 36.2	11 36.7 36.4 36.5 36.7 36.5 36.5 36.5 36.3	12 38.6 38.4 38.5 38.8 38.3 38.3 38.4
B 1 2 3 4 5 6 7 8	GGDC values <i>P. brassicacearum</i> subsp. <i>brassicacearum</i> NFM 421 ^T <i>P. brassicacearum</i> TM1A3 <i>P. brassicacearum</i> 51MCFVI21 <i>Pseudomonas</i> sp. Q8r1-96 <i>P. kilonensis</i> DSM 13647 ^T <i>P. kilonensis</i> P12 <i>Pseudomonas</i> sp. F113 <i>P. thivervalensis</i> DSM 13194 ^T	1 96.0 96.8 96.6 59.1 59.1 61.0 46.2	2 96.0 96.3 96.1 59.2 59.1 60.9 46.1	3 96.9 96.3 97.0 59.1 59.2 61.1 46.2	4 96.6 96.1 97.0 59.0 58.9 60.9 46.1	5 59.1 59.2 59.1 59.0 88.5 64.5 47.4	6 59.1 59.2 58.9 88.5 64.5 47.2	7 61.0 60.9 61.1 60.9 64.5 64.5 46.7	8 46.2 46.1 46.2 46.1 47.4 47.2 46.7	9 45.9 46.0 46.0 46.0 47.0 46.9 46.3 88.0	10 36.5 36.3 36.5 36.6 36.4 36.5 36.2 35.4	11 36.7 36.4 36.5 36.7 36.5 36.5 36.3 35.5	12 38.6 38.4 38.5 38.8 38.3 38.4 37.2
B 1 2 3 4 5 6 7 8 9	GGDC values P. brassicacearum subsp. brassicacearum NFM 421 ^T P. brassicacearum TM1A3 P. brassicacearum 51MCFVI21 Pseudomonas sp. Q8r1-96 P. kilonensis DSM 13647 ^T P. kilonensis P12 Pseudomonas sp. F113 P. thivervalensis DSM 13194 ^T P. thivervalensis PITR2	1 96.0 96.8 96.6 59.1 59.1 61.0 46.2 45.9	2 96.0 96.3 96.1 59.2 59.1 60.9 46.1 46.0	3 96.9 96.3 97.0 59.1 59.2 61.1 46.2 46.0	4 96.6 96.1 97.0 59.0 58.9 60.9 46.1 46.0	5 59.1 59.2 59.1 59.0 88.5 64.5 47.4 47.0	6 59.1 59.2 58.9 88.5 64.5 47.2 46.9	7 61.0 60.9 61.1 60.9 64.5 64.5 64.5 46.7 46.3	8 46.2 46.1 46.2 46.1 47.4 47.2 46.7 88.0	9 45.9 46.0 46.0 46.0 47.0 46.9 46.3 88.0	10 36.5 36.3 36.5 36.6 36.4 36.5 36.2 35.4 35.3	11 36.7 36.4 36.5 36.7 36.5 36.5 36.3 35.5 35.3	12 38.6 38.4 38.5 38.8 38.3 38.4 37.2 37.0
B 1 2 3 4 5 6 7 8 9 10	GGDC values P. brassicacearum subsp. brassicacearum NFM 421 ^T P. brassicacearum TM1A3 P. brassicacearum 51MCFVI21 Pseudomonas sp. Q8r1-96 P. kilonensis DSM 13647 ^T P. kilonensis P12 Pseudomonas sp. F113 P. thivervalensis DSM 13194 ^T P. thivervalensis PITR2 Pseudomonas sp. Q12-87	1 96.0 96.8 96.6 59.1 59.1 61.0 46.2 45.9 36.5	2 96.0 96.3 96.1 59.2 59.1 60.9 46.1 46.0 36.3	3 96.9 96.3 97.0 59.1 59.2 61.1 46.2 46.0 36.5	4 96.6 96.1 97.0 59.0 58.9 60.9 46.1 46.0 36.6	5 59.1 59.2 59.1 59.0 88.5 64.5 47.4 47.0 36.4	6 59.1 59.2 58.9 88.5 64.5 47.2 46.9 36.5	7 61.0 60.9 61.1 60.9 64.5 64.5 46.7 46.3 36.2	8 46.2 46.1 46.2 46.1 47.4 47.2 46.7 888.0 35.4	9 45.9 46.0 46.0 46.0 46.9 46.3 88.0 35.3	10 36.5 36.3 36.5 36.6 36.4 36.5 36.2 35.4 35.3	11 36.7 36.4 36.5 36.7 36.5 36.5 36.3 35.5 35.3 96.0	12 38.6 38.4 38.5 38.8 38.3 38.4 37.2 37.0 37.1
B 1 2 3 4 5 6 7 8 9 10 11	GGDC values <i>P. brassicacearum</i> subsp. <i>brassicacearum</i> NFM 421 ^T <i>P. brassicacearum</i> TM1A3 <i>P. brassicacearum</i> 51MCFVI21 <i>Pseudomonas</i> sp. Q8r1-96 <i>P. kilonensis</i> DSM 13647 ^T <i>P. kilonensis</i> P12 <i>Pseudomonas</i> sp. F113 <i>P. thivervalensis</i> DSM 13194 ^T <i>P. thivervalensis</i> PITR2 <i>Pseudomonas</i> sp. Q12-87 <i>Pseudomonas</i> sp. Q2-87	1 96.0 96.8 96.6 59.1 59.1 61.0 46.2 45.9 36.5 36.7	2 96.0 96.3 96.1 59.2 59.1 60.9 46.1 46.0 36.3 36.4	3 96.9 96.3 97.0 59.1 59.2 61.1 46.2 46.0 36.5 36.5	4 96.6 96.1 97.0 59.0 58.9 60.9 46.1 46.0 36.6 36.7	5 59.1 59.2 59.1 59.0 88.5 64.5 47.4 47.0 36.4 36.5	6 59.1 59.2 58.9 88.5 64.5 47.2 46.9 36.5 36.5	7 61.0 60.9 61.1 60.9 64.5 64.5 46.7 46.3 36.2 36.3	8 46.2 46.1 46.2 46.1 47.4 47.2 46.7 88.0 35.4 35.5	9 45.9 46.0 46.0 46.0 47.0 46.9 46.3 88.0 35.3 35.3	10 36.5 36.3 36.5 36.6 36.4 36.5 36.2 35.4 35.3 96.0	11 36.7 36.4 36.5 36.7 36.5 36.5 36.3 35.5 35.3 96.0	12 38.6 38.4 38.5 38.8 38.3 38.4 37.2 37.0 37.1 37.2

Sub-clade	Strain	LT50 (d)	survival (%) at 3 dpi ±sdev
Sub-clade 1	P. protegens CHA0 ^T	1.6 (1.5; 1.8) ^{abc}	3.1 ± 6.3 *
	P. protegens PGNR1	1.6 (1.4; 1.8) ^{abc}	0.0 ± 0.0 *
	P. protegens BRIP	1.3 (1.1; 1.5) ^a	0.0 ± 0.0 *
	P. protegens K94.41	1.0 (-19.1; 21.2) ^{abcdef}	0.0 ± 0.0 *
	P. protegens PF	1.4 (1.2; 1.6) ^{ab}	0.0 ± 0.0 *
	Pseudomonas sp. CMR12a	1.9 (1.7; 2.2) ^{cde}	6.3 ± 7.2 *
	<i>P. chl. piscium</i> DSM 21509^{T}	1.6 (1.4; 1.8) ^{abc}	3.1 ± 6.3 *
	P. chl. piscium PCL1391	1.7 (1.5; 1.9) ^{bcd}	0.0 ± 0.0 *
	P. chl. aureofaciens CD	1.5 (1.3; 1.7) ^{ab}	3.1 ± 6.3 *
	<i>P. chl. aureofaciens</i> LMG 1245^{T}	2.1 (1.9; 2.4) ^{def}	21.9 ± 12.0 *
	<i>P. chl. chlororaphis</i> LMG 5004^{T}	2.4 (2.1; 2.8) ^{ef}	31.3 ± 12.5 *
Sub-clade 2	P. brassicacearum TM1A3	NA	81.3 ± 21.7
	<i>P. kilonensis</i> DSM 13647^{T}	NA	93.8 ± 7.2
	P. kilonensis P12	NA	87.5 ± 10.2
	<i>P. thivervalensis</i> DSM 13194^{T}	NA	84.4 ± 12.0
	P. thivervalensis PITR2	NA	90.6 ± 12.0
	Pseudomonas sp. Q12-87	NA	81.3 ± 7.2
	Pseudomonas sp. P97.38	NA	87.5 ± 17.7
	<i>P. corrugata</i> DSM 7228 ^{T}	NA	75.0 ± 21.7
	Pseudomonas sp. Pf153	NA	93.8 ± 7.2
	Pseudomonas sp. P1.8	NA	87.5 ± 10.2
	Pseudomonas sp. P1.31	NA	84.4 ± 12.0
Sub-clade 3	<i>P. fluorescens</i> DSM 50090^{T}	NA	65.6 ± 12.0 *
	Pseudomonas sp. MIACH	2.7 (1.8; 3.5) ^{def}	45.8 ± 26.0 *
	Pseudomonas sp. SS101	2.8 (2.3; 3.3) ^f	46.9 ± 27.7 *
control	0.9% NaCL	NA	96.9 ± 6.3

Supplementary Table S4. Lethal time (LT₅₀) and survival of *Plutella xylostella* larvae upon oral uptake of *Pseudomonas* strains.

Repetition of the experiment depicted in Figure 3 and Table 2. *Plutella xylostella* larvae were exposed to food pellets inoculated with 8 x 10^7 bacterial cells. LT_{50} values are estimates based on the generalized linear model using the MASS package in R (Venables and Ripley 2002). Numbers in brackets depict 95% confidence intervals for LT_{50} and significantly different values within the same column are followed by different letters.

NA = no LT_{50} value was calculated, because end mortality was less than 50%.

Asterisks indicate significant differences compared to control larvae treated with 0.9% NaCl based on multiple comparisons by Kruskal-Wallis adjusted by Bonferroni-Holm ($p \le 0.05$).

Sub-clade	Strain	Biocontrol Activity relative to P. protegens CHA0						
		repetitio	n 1			repetition 2		
Sub-clade 1	<i>P. protegens</i> $CHA0^{T}$	1.00			*	1.00	*	
	P. protegens PGNR1	1.02	±	0.14	*	0.76 ± 0.05	*	
	P. protegens BRIP	0.94	±	0.24	*	1.07 ± 0.09	*	
	P. protegens K94.41	0.43	±	0.21	*	0.50 ± 0.23	*	
	P. protegens PF	0.35	±	0.16	*	0.49 ± 0.33	*	
	Pseudomonas sp. CMR5c	0.02	±	0.04		0.00 ± 0.00		
	Pseudomonas sp. CMR12a	0.51	±	0.25	*	0.00 ± 0.00		
	<i>P. chl. piscium</i> DSM 21509^{T}	0.54	±	0.28	*	0.21 ± 0.30		
	P. chl. piscium PCL1391	0.72	±	0.15	*	0.55 ± 0.30	*	
	<i>P. chl. aureofaciens</i> LMG 1245^{T}	0.15	±	0.23		0.39 ± 0.56		
	P. chl. aureofaciens CD	0.89	±	0.11	*	0.51 ± 0.11	*	
	<i>P. chl. chlororaphis</i> LMG 5004 ^{T}	0.00	±	0.00		0.00 ± 0.00		
Sub-clade 2	P. brassicacearum TM1A3	0.72	±	0.22	*	0.31 ± 0.31		
	P. kilonensis DSM 13647^{T}	0.00	±	0.00		0.00 ± 0.00		
	P. kilonensis P12	0.23	±	0.29		0.18 ± 0.20		
	<i>P. thivervalensis</i> DSM 13194^{T}	0.31	±	0.30		0.28 ± 0.28		
	P. thivervalensis PITR2	0.70	±	0.15	*	0.74 ± 0.07	*	
	Pseudomonas sp. Q12-87	0.67	±	0.21	*	0.58 ± 0.32	*	
	Pseudomonas sp. P97.38	0.61	±	0.12	*	0.51 ± 0.17	*	
	<i>P. corrugata</i> DSM 7228 ^{T}	0.00	±	0.00		0.06 ± 0.14		
	Pseudomonas sp. Pf153	0.52	±	0.12	*	0.25 ± 0.38		
	Pseudomonas sp. P1.8	0.05	±	0.12		0.33 ± 0.34		
	Pseudomonas sp. P1.31	0.61	±	0.11	*	0.54 ± 0.21	*	
Sub-clade 3	P. fluorescens DSM 50090 [™]	0.00	±	0.00		0.00 ± 0.00		
	Pseudomonas sp. MIACH	0.39	±	0.47		1.02 ± 0.47	*	
	Pseudomonas sp. SS101	0.82	±	0.25	*	0.00 ± 0.00		

Supplementary	/ Table S5. Biocontro	l activity against	Pythium ultimum	on cucumber plants

Biocontrol activity was calculated after Rezzonico et al (2007) as:

 $(1 - ((W_{c} - W_{I})/(W_{c} - W_{P}))) \times 100$

using shoot weight obtained in the control with neither bacterial nor pathogen inoculum (W_c), in the unprotected control with the pathogen alone (W_p) and in presence of the tested bacterial strain and the pathogen (W_l). Due to the large number of strains, not all strains could be tested in the same experiment. Therefore, biocontrol activity is shown relative to the biocontrol activity of our model strain *P. protegens* CHAO, which was included as a reference in all experiments. A total of seven experiments was performed and each strain was tested at least twice (repetition 1 and repetition 2). Biocontrol activity for *P. protegens* CHAO ranged between 63% and 100%.

Means of five replicates \pm sdev are shown. Statistics was performed for each experiment separately on absolute biocontrol activity values. Asterisks indicate that strains displayed significant biocontrol activity based on a t-test (p = 0.05) against the unprotected control with pathogen alone (W_P).

Sub-clade	Strain	# Reads * 10 ⁶	# Contigs	Genome size (Mbp)	N50 (kb)	Coverage
Sub-clade 1	P. protegens PGNR1	1.96	15	6.86	871	75
	P. protegens BRIP	1.12	19	6.89	701	44
	P. protegens K94.41	1.28	17	6.99	582	50
	P. protegens PF	1.41	14	7.07	1051	52
	Pseudomonas sp. CMR5c	22.35	44	6.76	502	37
	<i>P. chlororaphis</i> subsp. <i>piscium</i> DSM21509 ^T	1.35	36	7.04	414	51
	P. chlororaphis subsp. piscium PCL1391	1.29	17	6.86	820	51
	<i>P. chlororaphis</i> subsp. <i>aureofaciens</i> LMG 1245^{T}	1.43	45	7.02	311	54
	P. chlororaphis subsp. aureofaciens CD	1.92	32	6.8	388	75
	<i>P. chlororaphis</i> subsp. <i>chlororaphis</i> LMG 5004^{T}	1.13	15	6.79	875	44
Sub-clade 2	P. brassicacearum TM1A3	2.16	29	6.69	552	86
	<i>P. kilonensis</i> DSM 13647 ^{T}	1.60	44	6.39	281	66
	P. kilonensis P12	1.98	44	6.39	277	81
	<i>P. thivervalensis</i> DSM 13194 ^{T}	1.65	25	6.58	445	67
	P. thivervalensis PITR2	1.75	26	6.77	661	68
	Pseudomonas sp. Q12-87	1.33	45	6.30	261	56
	Pseudomonas sp. P97.38	2.12	36	6.06	278	92
	<i>P.</i> corrugata DSM 7228 ^{T}	1.63	31	6.13	374	71
	Pseudomonas sp. Pf153	1.70	30	5.98	577	75
	Pseudomonas sp. P1.8	1.87	43	6.36	325	79
	Pseudomonas sp. P1.31	2.18	48	6.27	262	92
Sub-clade 3	<i>P. fluorescens</i> DSM 50090^{T}	1.43	17	6.39	973	59
	Pseudomonas sp. MIACH	1.37	73	6.82	236	54

Supplementary Table S6. Genomic features

		tegens .	lomonas sp. CMR	roraphis	lomonas sp. SS101	orescens DSM50090 ^T	<i>lomnas</i> sp. MIACH	ade 2
Locus Tags	Gene	P. pro	Pseud	P.chlc	Pseud	P. fluc	Pseud	sub-cl
PCL1391_0010	Thioesterase	+	+	+	-	-	-	-
PCL1391_0029	Hypothetical protein	+	+	+	+	+	+	-
PCL1391_0030	Hypothetical protein	+	+	+	+	+	+	-
PCL1391_0072	RebB like protein	+	+	+	+	-	-	-
PCL1391_0073	Hypothetical protein	+	+	+	+	-	-	-
PCL1391_0075	RebB protein	÷	+	+	+	-	-	-
PCL1391_0076	RebB protein	+	+	+	+	-	-	-
PCL1391_0101	Hypothetical protein	+	+	+	+	+	-	-
PCL1391_0108	Cyanate transport protein CynX	+	+	+	+	+	+	-
PCL1391_0109	CMP deaminase	+	+	+	+	+	-	-
PCL1391_0110	Putative ankyrin-containing lipoprotein	+	+	+	-	-	-	-
PCL1391_0111	LysR family transcriptional regulator	+	+	+	+	+	+	-
PCL1391_0170	Hypothetical protein	+	+	+	+	+	+	-
PCL1391_0171	Polysaccharide deacetylase	+	+	+	-	-	-	-
PCL1391_0195	Ketosteroid isomerase	+	+	+	-	-	-	-
PCL1391_0279	Histidine-specific permease	+	+	+	+	+	+	-
PCL1391_0332	Hypothetical protein	+	+	+	+	+	+	-
PCL1391_0603	Kynurenine formamidase	+	+	+	+	+	+	-
PCL1391_0604	Tryptophan 2,3-dioxygenase	+	+	+	+	+	+	-
PCL1391_0605	Aromatic amino acid transport protein AroP	+	+	+	+	+	+	-
PCL1391_0610	AsnC family transcriptional regulator	+	+	+	+	+	+	-
PCL1391_0611	Kynureninase	+	+	+	+	+	+	-
PCL1391_0612	Amino acid permease	+	+	+	+	+	+	-
PCL1391_0639	Alpha/beta hydrolase	+	+	+	-	-	-	-
PCL1391_0640	membrane protein	+	+	+	+	+	+	-
PCL1391_0733	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_0734	Phosphatidylcholine hydrolyzing phospholipase	+	+	+	+	+	+	-
PCL1391_0828	Serine transporter	+	+	+	+	+	-	-
PCL1391_0938	Signal transduction histidine kinase	+	+	+	+	-	-	-
PCL1391_0939	Hisitdine kinase	+	+	+	+	-	-	-
PCL1391_0940	LuxR family transcriptional regulator	+	+	+	+	+	+	-
PCL1391 0949	GNAT family acetyltransferase	+	+	+	-	-	-	-

Supplementary Table S7. Genes specific to insecticidal strains. Locus tags (prefix PCL1391_) and gene names are indicated for *Pseudomonas chlororaphis* subsp. *piscium* PCL1391.

PCL1391_1183	Membrane protein	+	+	+	-	-	-	-
PCL1391_1217	TonB-denpendent receptor	+	+	+	+	+	+	-
PCL1391_1218	Iron dicitrate transporter FecR	+	+	+	+	+	+	-
PCL1391_1219	RNA polymerase sigma factor	+	+	+	+	+	+	-
PCL1391_1245	Oxidoreductase	+	+	+	-	-	-	-
PCL1391_1247	GNAT family acetyltransferase	+	+	+	-	-	-	-
PCL1391_1251	Cyclic diguanylate phosphodiesterase	+	+	+	-	-	-	-
PCL1391_1352	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_1354	LysR family transcriptional regulator	+	+	+	-	-	-	-
PCL1391_1370	AraC family transcriptional regulator	+	+	+	-	-	-	-
PCL1391_1510	Endoribonuclease L-PSP	+	+	+	+	-	-	-
PCL1391_1588	Probable sugar efflux transporter	+	+	+	+	+	+	-
PCL1391_1733	Extradiol dioxygenase	+	+	+	-	+	+	-
PCL1391_1817	MFS transporter	+	+	+	-	-	-	-
PCL1391_1854	Chitin-binding protein	+	+	+	+	+	-	-
PCL1391_1855	Chitinase	+	+	+	+	+	-	-
PCL1391_1901	HxIR family transcriptional regulator	+	+	+	+	-	+	-
PCL1391_1903	Heme transporter CcmD	+	+	+	-	-	-	-
PCL1391_1904	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_1905	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_1906	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_1910	Haloacid dehalogenase	+	+	+	+	+	+	-
PCL1391_1978	IclR family transcriptional regulator	+	+	+	+	-	-	-
PCL1391_1979	ABC transporter substrate-binding protein	+	+	+	+	-	-	-
PCL1391_1980	Amino acid ABC transporter permease	+	+	+	+	-	-	-
PCL1391_1982	FAD-dependent oxidoreductase	+	+	+	+	-	-	-
PCL1391_2008	Hypothetical protein	+	+	+	+	+	+	-
PCL1391_2011	RNA-binding protein	+	+	+	-	-	-	-
PCL1391_2016	RNA 3'-terminal phosphate cyclase	+	+	+	-	-	-	-
PCL1391_2021	Diaminopimelate decarboxylase	+	+	+	-	-	-	-
PCL1391_2037	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_2051	RNA polymerase subunit sigma-70	+	+	+	+	-	+	-
PCL1391_2053	Putative TonB-dependent receptor	+	+	+	+	-	-	-
PCL1391_2076	Methyl-accepting chemotaxis protein	+	+	+	-	-	-	-
PCL1391_2141	Serralysin	+	+	+	-	-	-	-
PCL1391_2164	Alpha/beta hydrolase	+	+	+	-	+	+	-
PCL1391_2185	GNAT family acetyltransferase	+	+	+	+	+	-	-
PCL1391_2193	ABC transporter permease	+	+	+	-	+	+	-
PCL1391_2194	ABC transporter substrate-binding protein	+	+	+	-	+	+	-
PCL1391_2195	Methionine sulfoxide reductase A	+	+	+	-	-	-	-
PCL1391_2197	RND transporter	+	+	+	+	+	+	-

PCL1391_2199	Hypothetical protein	+	+	+	+	+	+	-
PCL1391_2220	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_2221	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_2281	Acyl-CoA dehydrogenase	+	+	+	-	+	+	-
PCL1391_2405	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_2433	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_2479	Hypothetical protein	+	+	+	+	+	-	-
PCL1391_2481	Na/Pi cotransporter	+	+	+	-	-	-	-
PCL1391_2482	Hypothetical membrane protein	+	+	+	-	-	-	-
PCL1391_2483	Magnesium-transporting ATPase, P-type 1	+	+	+	+	+	+	-
PCL1391_2484	Conserved hypothetical protein	+	+	+	+	+	-	-
PCL1391_2556	MFS transporter	+	+	+	+	-	-	-
PCL1391_2557	L-2-hydroxyglutarate oxidase LhgO	+	+	+	+	-	-	-
PCL1391_2558	GntR family transcriptional regulator	+	+	+	+	-	-	-
PCL1391_2605	AraC family transcriptional regulator	+	+	+	-	-	-	-
PCL1391_2609	Sulfite reductase	+	+	+	-	-	-	-
PCL1391_2610	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_2645	Polyketide cyclase	+	+	+	+	+	+	-
PCL1391_2659	MFS transporter	+	+	+	+	-	+	-
PCL1391_2660	Oxidoreductase	+	+	+	-	-	-	-
PCL1391_2673	Transcriptional activator protein CzcR	+	+	+	+	+	+	-
PCL1391_2790	Glycosyltransferase	+	+	+	+	+	+	-
PCL1391_2887	Biopolymer transporter ExbD	+	+	+	+	+	-	-
PCL1391_2966	Non-hemolytic phospholipase C	+	+	+	-	-	-	-
PCL1391_2967	Membrane protein	+	+	+	-	-	-	-
PCL1391_2972	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_2973	Cyclic diguanylate phosphodiesterase	+	+	+	-	-	-	-
PCL1391_2987	Putative ABC transporter, permease subunit	+	+	+	+	+	-	-
PCL1391_2988	Putative ABC transporter, substrate-binding protein	+	+	+	+	+	-	-
PCL1391_2989	Putative ABC transporter, ATP-binding protein	+	+	+	+	+	-	-
PCL1391_2990	Acyl-CoA dehydrogenase	+	+	+	+	+	-	-
PCL1391_2992	AraC family transcriptional regulator	+	+	+	+	+	-	-
PCL1391_3032	Aminotransferase	+	+	+	-	+	+	-
PCL1391_3062	Amino acid transporter	+	+	+	-	-	-	-
PCL1391_3089	MFS transporter	+	+	+	-	-	-	-
PCL1391_3117	4-Hydroxyphenylacetate 3-monooxygenase oxygenase component	+	+	+	-	-	-	-
PCL1391_3126	(R,R)-Butanediol dehydrogenase	+	+	+	-	-	-	-
PCL1391_3130	Hypothetical protein	+	+	+	-	+	+	-
PCL1391_3144	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_3145	LysR family transcriptional regulator	+	+	+	+	+	+	-

PCL1391_3234	Transporter	+	+	+	-	-	-	-
PCL1391_3422	Hypothetical protein	+	+	+	+	+	+	-
PCL1391_3423	Conserved hypothetical protein	+	+	+	+	+	+	-
PCL1391_3454	Response regulator FitH	+	+	+	-	-	-	-
PCL1391_3455	Transcriptional regulator FitG	+	+	+	-	-	-	-
PCL1391_3456	Sensor histidine kinase FitF	+	+	+	-	-	-	-
PCL1391_3457	Channel protein FitE	+	+	+	-	-	-	-
PCL1391_3458	Cytotoxin FitD	+	+	+	-	-	-	-
PCL1391_3459	Type I secretion system ATPase FitC	+	+	+	-	-	-	-
PCL1391_3479	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_3513	Putative glucosidase	+	+	+	+	+	+	-
PCL1391_3514	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_3515	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_3565	Molybdenum cofactor biosynthesis protein MoaA	+	+	+	-	-	-	-
PCL1391_3566	Molybdenum cofactor biosynthesis protein B	+	+	+	-	-	-	-
PCL1391_3569	Molybdopterin synthase catalytic subunit	+	+	+	-	-	-	-
PCL1391_3570	Molybdenum cofactor biosynthesis protein MoaD	+	+	+	-	-	-	-
PCL1391_3571	Molybdenum cofactor biosynthesis protein MoaC	+	+	+	-	-	-	-
PCL1391_3574	Molybdopterin-dependent oxidoreductase alpha subunit	+	+	+	-	-	-	-
PCL1391_3575	Cytochrome D ubiquinol oxidase subunit I	+	+	+	-	-	-	-
PCL1391_3576	Ubiquinol oxidase subunit II, cyanide insensitive	+	+	+	-	-	-	-
PCL1391_3600	TonB-denpendent receptor	+	+	+	+	+	+	-
PCL1391_3671	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_3751	Nucleoside 2-deoxyribosyltransferase	+	+	+	-	-	-	-
PCL1391_3843	Conserved hypothetical protein	+	+	+	+	+	-	-
PCL1391_3855	DNA polymerase subunit beta	+	+	+	+	+	+	-
PCL1391_3876	Hypothetical protein	+	+	+	+	+	+	-
PCL1391_3932	HAD family hydrolase	+	+	+	-	-	-	-
PCL1391_3935	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_3937	Amidohydrolase	+	+	+	-	-	-	-
PCL1391_3989	Methyltransferase	+	+	+	-	-	-	-
PCL1391_4028	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_4037	Hemolysin secretion/activation protein, ShIB family	+	+	+	+	+	-	-
PCL1391_4083	LuxR family transcriptional regulator	+	+	+	-	-	-	-
PCL1391_4176	Glutathione S-transferase	+	+	+	+	+	+	-
PCL1391_4307	Hypothetical protein	+	+	+	+	+	-	-
PCL1391_4350	AraC family transcriptional regulator	+	+	+	-	-	-	-
PCL1391_4351	Fatty acid hydroxylase	+	+	+	-	-	-	-

PCL1391_4367	TetR family transcriptional regulator	+	+	+	-	-	-	-
PCL1391_4386	Lysine transporter LysE	+	+	+	-	-	-	-
PCL1391_4387	LysR family transcriptional regulator	+	+	+	-	-	-	-
PCL1391_4607	Putative arginase	+	+	+	+	-	+	-
PCL1391_4608	Transporter	+	+	+	+	-	+	-
PCL1391_4609	Transporter	+	+	+	+	-	+	-
PCL1391_4610	Fatty acid desaturase	+	+	+	-	-	ŀ	-
PCL1391_4611	Structural protein MipA	+	+	+	+	+	+	-
DCI 1201 4612	2,3-Diketo-5-methylthio-1-phosphopentane	+	+	+	+	+	+	-
FCL1391_4012	Adenosylmethionine-8-amino-7-oxo-nanoate							
PCL1391_4613	aminotransferase	+	+	+	+	+	+	-
PCL1391_4614	Esterase	+	+	+	+	+	+	-
PCL1391_4615	Hypothetical protein	+	+	+	+	+	+	-
PCL1391_4616	Sensor histidine kinase	+	+	+	+	+	+	-
PCL1391_4617	Fis family transcriptional regulator	+	+	+	+	+	+	-
PCL1391_4626	50S ribosomal protein L31	+	+	+	+	+	-	-
PCL1391_4642	TonB-denpendent receptor	+	+	+	+	+	+	-
PCL1391_4646	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_4723	HIT family hydrolase	+	+	+	-	-	-	-
PCL1391_4735	Serine hydroxymethyltransferase	+	+	+	-	+	+	-
PCL1391_4798	Hypothetical membrane protein	+	+	+	+	-	-	-
PCL1391_4800	XRE family transcriptional regulator	+	+	+	+	-	-	-
PCL1391_4801	Histidine kinase	+	+	+	+	-	-	-
PCL1391_4891	GNAT family acetyltransferase	+	+	+	-	-	-	-
PCL1391_4896	D-alanyl-alanine synthetase	+	+	+	-	+	+	-
PCL1391_4904	MFS transporter	+	+	+	-	-	-	-
PCL1391_4905	LysR family transcriptional regulator	+	+	+	-	-	-	-
PCL1391_4907	Short-chain dehydrogenase	+	+	+	+	-	-	-
PCL1391_4917	Cobalt-zinc-cadmium resistance protein CzcD	+	+	+	+	+	-	-
PCL1391_4925	Acid phosphatase	+	+	+	+	+	+	-
PCL1391_4982	Hypothetical protein	+	+	+	-	+	+	-
PCL1391_4983	Glycosyl transferase PsIA	+	+	+	+	+	+	-
PCL1391_4985	Glycosyl transferase PsIC	+	+	+	+	+	+	-
PCL1391_4986	Polysaccharide biosynthesis/export protein PsID	+	+	+	+	+	+	-
PCL1391_4987	Polysaccharide biosynthesis/export protein PsIE	+	+	+	+	+	+	-
PCL1391_4988	glycosyl transferase PsIF	+	+	+	+	+	+	-
PCL1391_4989	Glycosyl hydrolase PslG	+	+	+	+	+	+	-
PCL1391_4990	Glycosyl transferase PsIH	+	+	+	+	+	+	-
PCL1391_4991	Glycosyl transferase Psll	+	+	+	+	+	+	-
PCL1391_4992	Membrane protein PslJ	+	+	+	+	+	+	-
PCL1391_4993	Acetyltransferase	+	+	+	+	-	-	-

PCL1391_4994	Membrane protein PslK	+	+	+	+	+	+	-
PCL1391_5052	Hypothetical protein	+	+	+	-	+	+	-
PCL1391_5077	Benzoate transporter	+	+	+	+	-	+	-
PCL1391_5099	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_5179	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_5182	Copper-containing nitrite reductase	+	+	+	-	-	-	-
PCL1391_5360	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_5397	Aminoglycoside N(6')-acetyltransferase	+	+	+	-	-	-	-
PCL1391_5511	Putative membrane protein	+	+	+	-	-	-	-
PCL1391_5577	UDP-4-amino-4-deoxy-L-arabinose oxoglutarate ami-transferase	+	+	+	+	+	+	-
PCL1391_5659	Membrane protein	+	+	+	-	+	+	-
PCL1391_5765	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_5770	Hypothetical protein	+	+	+	-	-	-	-
PCL1391_5773	Membrane protein	+	+	+	+	+	+	-
PCL1391_5799	TraR family zinc finger protein	+	+	+	+	+	-	-
PCL1391_5806	Phosphoribosyl-AMP cyclohydrolase 2	+	+	+	-	+	+	-

+, gene present; -, gene absent Loci shaded in grey are discussed in the text.

References

Auch AF, von Jan M, Klenk H-P, Goeker M. (2010). Digital DNA-DNA hybridization for microbial species delineation by means of genome-to-genome sequence comparison. *Stand Genomic Sci* **2**: 117-134.

Blom J, Albaum SP, Doppmeier D, Pühler A, Vorhölter F-J, Zakrzewski M *et al.* (2009). EDGAR: a software framework for the comparative analysis of prokaryotic genomes. *BMC Bioinformatics* **10**: 154.

Boetzer M, Henkel CV, Jansen HJ, Butler D, Pirovano W. (2011). Scaffolding pre-assembled contigs using SSPACE. *Bioinformatics* **27**: 578-579.

Boetzer M, Pirovano W. (2012). Toward almost closed genomes with GapFiller. *Genome Biol* **13**: R56.

Burr SE, Gobeli S, Kuhnert P, Goldschmidt-Clermont E, Frey J. (2010). *Pseudomonas chlororaphis* subsp. *piscium* subsp. nov., isolated from freshwater fish. *Int J Syst Evol Microbiol* **60**: 2753-2757.

Chin-A-Woeng TFC, Bloemberg GV, van der Bij AJ, van der Drift K, Schripsema J, Kroon B *et al.* (1998). Biocontrol by phenazine-1-carboxamide-producing *Pseudomonas chlororaphis* PCL1391 of tomato root rot caused by *Fusarium oxysporum* f. sp. *radicis-lycopersici*. *Mol Plant-Microbe Interact* **11**: 1069-1077.

Gomila M, Peña A, Mulet M, Lalucat J, García-Valdés E. (2015). Phylogenomics and systematics in *Pseudomonas*. *Front Microbiol* **6**: 214.

Gupta GP, Rani S, Birah A, Raghuraman M. (2005). Improved artificial diet for mass rearing of the tobacco caterpillar, *Spodoptera litura* (Lepidoptera: Noctuidae). *Int J Trop Insect Sci* **25**: 55-58.

Jousset A, Schuldes J, Keel C, Maurhofer M, Daniel R, Scheu S *et al.* (2014). Full-genome sequence of the plant growth-promoting bacterium *Pseudomonas protegens* CHA0. *Genome Announc* **2**: e00322-00314.

Keel C, Weller DM, Natsch A, Defago G, Cook RJ, Thomashow LS. (1996). Conservation of the 2,4-diacetylphloroglucinol biosynthesis locus among fluorescent *Pseudomonas* strains from diverse geographic locations. *Appl Environ Microbiol* **62**: 552-563.

King EO, Ward MK, Raney DE. (1954). 2 simple media for the demonstration of pyocyanin and fluorescin. *Journal of Laboratory and Clinical Medicine* **44**: 301-307.

Konstantinidis KT, Ramette A, Tiedje JM. (2006). The bacterial species definition in the genomic era. *Phil Trans R Soc B* **361:** 1929-1940.

Martinez-Garcia E, de Lorenzo V. (2011). Engineering multiple genomic deletions in Gramnegative bacteria: analysis of the multi-resistant antibiotic profile of *Pseudomonas putida* KT2440. *Environ Microbiol* **13**: 2702-2716. Meier-Kolthoff JP, Auch AF, Klenk HP, Goker M. (2013). Genome sequence-based species delimitation with confidence intervals and improved distance functions. *BMC Bioinformatics* **14:** 60.

Meier-Kolthoff JP, Klenk H-P, Göker M. (2014). Taxonomic use of DNA G+C content and DNA-DNA hybridization in the genomic age. *Int J Syst Evol Microbiol* **64**: 352-356.

Mulet M, Gomila M, Scotta C, Sánchez D, Lalucat J, García-Valdéz E. (2012). Concordance between whole-cell matrix-assisted laser-desorption/ionization time-of-flight mass spectrometry and multilocus sequence analysis approaches in species discrimination within the genus *Pseudomonas*. *Syst Appl Microbiol* **35**: 455-464.

Peix A, Valverde A, Rivas R, Igual JM, Ramírez-Bahena M-H, Mateos PF *et al.* (2007). Reclassification of *Pseudomonas aurantiaca* as a synonym of *Pseudomonas chlororaphis* and proposal of three subspecies, *P. chlororaphis* subsp. *chlororaphis* subsp. nov., *P. chlororaphis* subsp. aureofaciens subsp. nov., comb. nov. and *P. chlororaphis* subsp. *aurantiaca* subsp. nov., comb. nov. and *P. chlororaphis* subsp. aurantiaca subsp. nov., comb. nov. and *P. chlororaphis* subsp. aurantiaca subsp. nov., comb. nov. Int J Syst Evol Microbiol **57**: 1286-1290.

Perneel M, Heyrman J, Adiobo A, De Maeyer K, Raaijmakers JM, De Vos P *et al.* (2007). Characterization of CMR5c and CMR12a, novel fluorescent *Pseudomonas* strains from the cocoyam rhizosphere with biocontrol activity. *J Appl Microbiol* **103**: 1007-1020.

Ramette A, Frapolli M, Fischer-Le Saux M, Gruffaz C, Meyer J-M, Défago G *et al.* (2011). *Pseudomonas protegens* sp. nov., widespread plant-protecting bacteria producing the biocontrol compounds 2,4-diacetylphloroglucinol and pyoluteorin. *Int J Syst Evol Microbiol* **34**: 180-188.

Rezzonico F, Zala M, Keel C, Duffy B, Moenne-Loccoz Y, Defago G. (2007). Is the ability of biocontrol fluorescent pseudomonads to produce the antifungal metabolite 2,4-diacetylphloroglucinol really synonymous with higher plant protection? *New Phytol* **173**: 861-872.

Ruffner B, Péchy-Tarr M, Ryffel F, Hoegger P, Obrist C, Rindlisbacher A *et al.* (2013). Oral insecticidal activity of plant-associated pseudomonads. *Environ Microbiol* **15**: 751-763.

Ruffner B, Péchy-Tarr M, Höfte M, Bloemberg G, Grunder J, Keel C *et al.* (2015). Evolutionary patchwork of an insecticidal toxin shared between plant-associated pseudomonads and the insect pathogens *Photorhabdus* and *Xenorhabdus*. *BMC Genomics* **16**: 609.

Sambrook J, Russel DW (2001). *Molecular Cloning: A Laboratory Manual*. Cold Spring Harbor Laboratory Press: New York.

Sharifi-Tehrani A, Zala M, Natsch A, Moenne-Loccoz Y, Defago G. (1998). Biocontrol of soilborne fungal plant diseases by 2,4-diacetylphloroglucinol-producing fluorescent pseudomonads with different restriction profiles of amplified 16S rDNA. *Eur J Plant Pathol* **104:** 631-643.

Stutz EW, Defago G, Kern H. (1986). Naturally-occuring fluorescent pseudomonads involved in suppression of black root-rot of tobacco. *Phytopathology* **76**: 181-185.

Tambong JT, Höfte M. (2001). Phenazines are involved in biocontrol of *Pythium myriotylum* on cocoyam by *Pseudomonas aeruginosa* PNA1. *Eur J Plant Pathol* **107**: 511-521.

Venables WN, Ripley BD (2002). *Modern Applied Statistics with S*, Fourth edn. Springer: New York.