

*Supplementary Information*

Ornithine Transcarbamylase ArgK Plays a Dual role for the Self-  
defense of Phaseolotoxin Producing *Pseudomonas syringae* pv.  
*phaseolicola*

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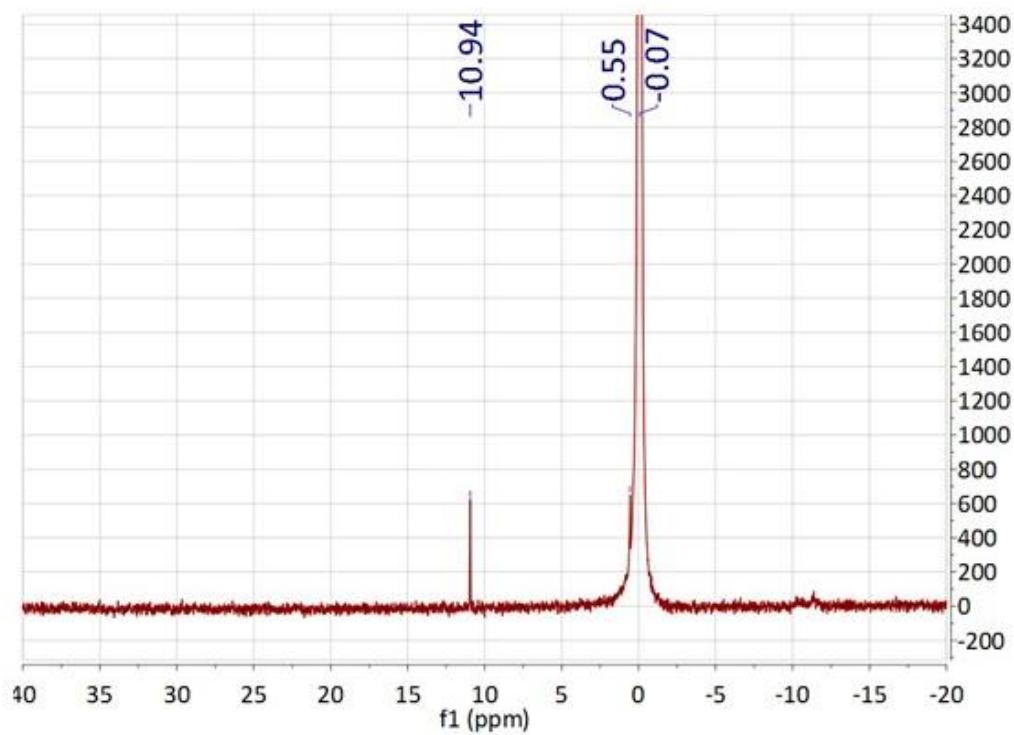
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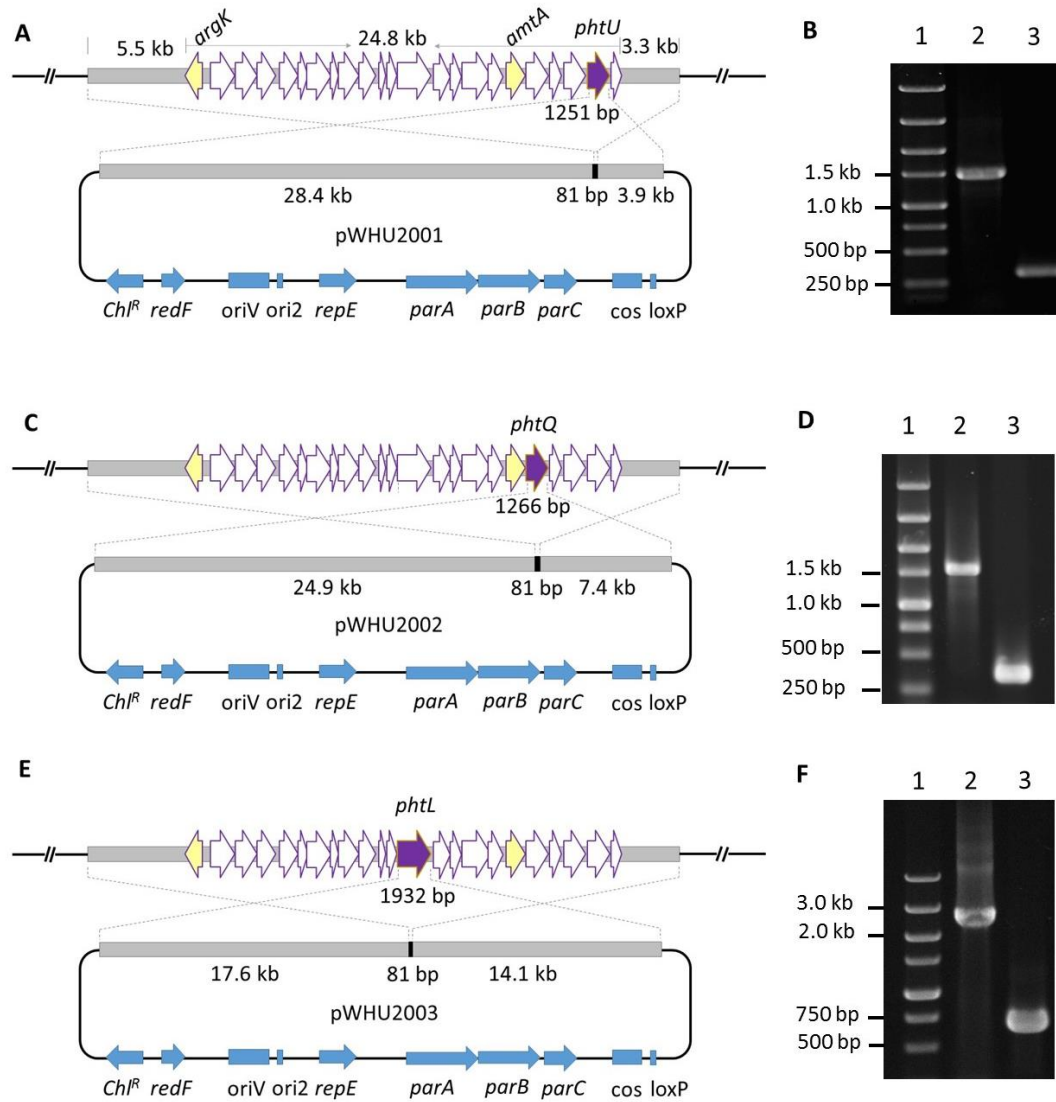
Running Title: ArgK's role in PHT producers' self-defense

**Table S1** PCR Primers used in this study

Primer name	Primer sequence
DUF1	GCTTTTATCAGAAAAATGAAAGCTAATTTGAATTGGGGAGTCATTCCGGG GATCCGTCGACC
DUR2	AGGAATTTCAAAGCTTCCTCACACCTGAATGTTAAGAGTGATTGTAGGCT GGAGCTGCTTC
VUF1	TTACGCGATGTGCAGTAGGATA
VUR2	AATGGCGATACCTGACACCT AT
DQF1	GATCTGGTTATTTTTGAATATTGAATTTACGAGAGGCAGTTCATTCCGGGG ATCCGTCGACC
DQR2	ATGAACACTCGGAGGGCGCTAGGTATGCCTGCGGCTGCCATTGTAGGCT GGAGCTGCTTC
VQF1	CGCAGGGTTTGAGCCGATTCTA
VQR2	AGTCGCCATTACTCCGTACACG
DLF1	TGGGTTCTTGTGCCGCAATTTAAAGGAAATTAACCGGTGATTCCGGGGA TCCGTCGACC
DLR2	TGCAAAGCTGATCTGAAAGGCATCGGCGAAACACTTTTCATGTAGGCTG GAGCTGCTTC
VLF1	TGGACCATTCAACTTTTGGCCGTCA
VLR2	GCGAATGAGATGAGGTATCTGGCAA
P <sub>phtA</sub> F1	TAAGATTAGCGGATCCCATTAGTGTAGGCAGGGC
P <sub>phtA</sub> R2	ACTTATTACTTCCTTTTTTCGT
PhtUF1	AAGGAAGTAATAAGTATGACGAATATATTAATGG
PhtUR2	CGACTCTAGAGGATCCTTAGTATGAAAGAGATACA
PhtQF1	AAGGAAGTAATAAGTGTGCCAATGAAAAAATAATAT
PhtQR2	CGACTCTAGAGGATCCTCAAAGTGGTTCAGAACTA
OKF1	CGCGGATCCATGAAGATTACAAGCCTGAAAAACC
OKR2	CCGCTCGAGTCAGGGGACGACTGTCTCCAGCATC



**Figure S1**  $^{31}\text{P}$  NMR spectrum of PHTs produced by *Pseudomonas syringae* pv. *phaseolicola* 1448A.



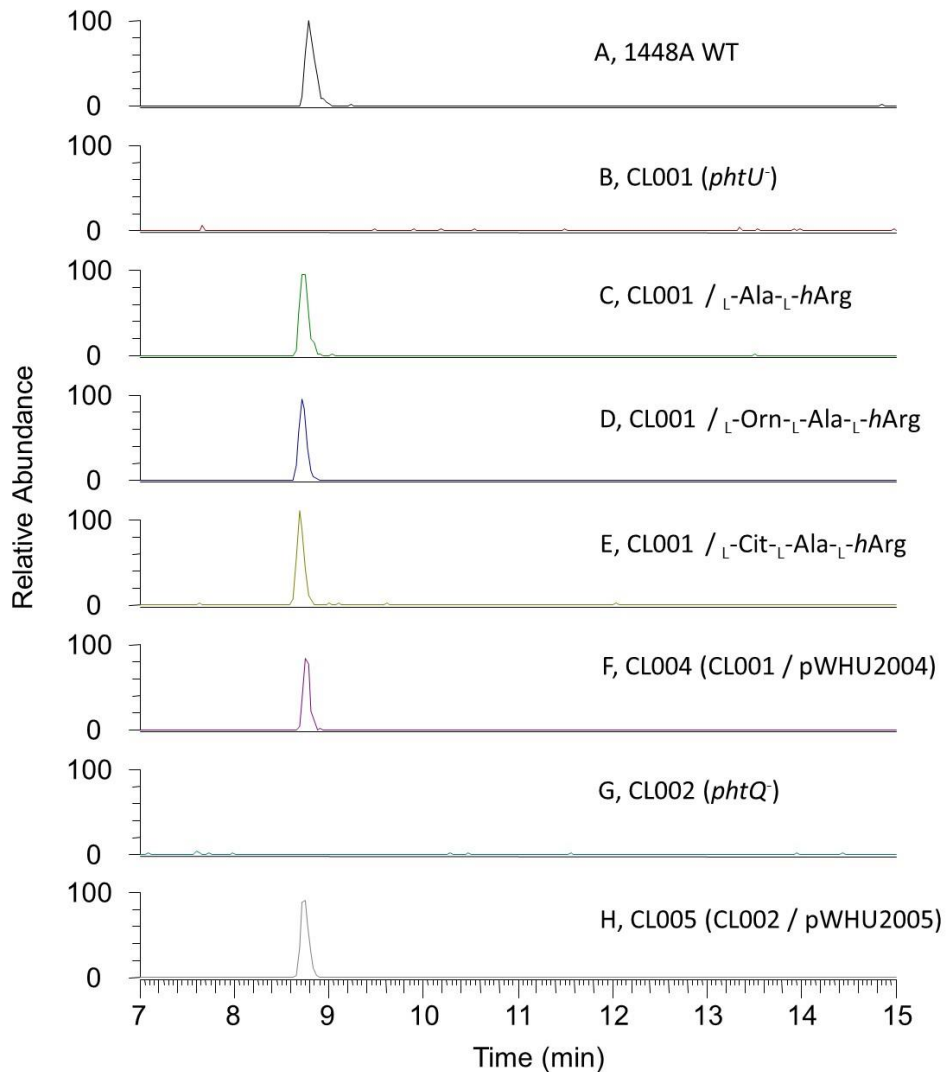
**Figure S2** In-frame deletion of PHT biosynthesis related gene *phtU* (A), *phtQ* (C)

and *phtL* (E), and PCR validation of *phtU*<sup>-</sup> (B), *phtQ*<sup>-</sup> (D) and *phtL*<sup>-</sup> (F) mutants.

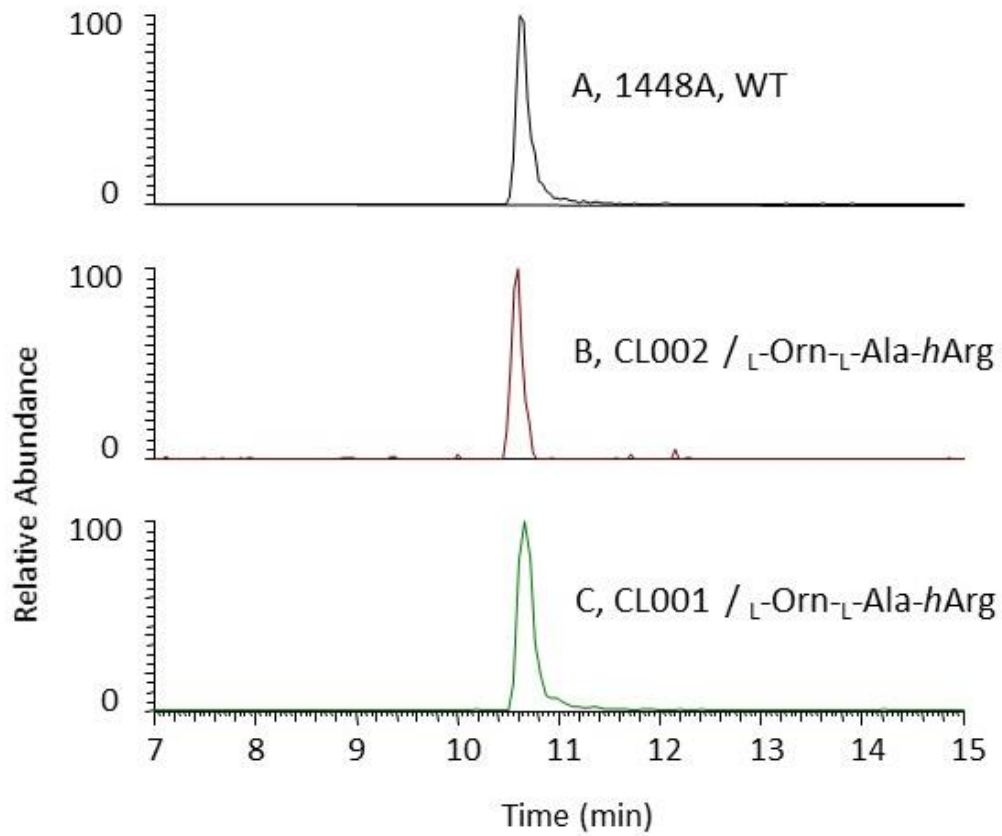
B, lane 1, DNA marker; lane 2, wild type (1471 bp); lane 3, *phtU*<sup>-</sup> mutant (301 bp).

D, lane 1, DNA marker; lane 2, wild type (1531 bp); lane 3, *phtQ*<sup>-</sup> mutant (265 bp).

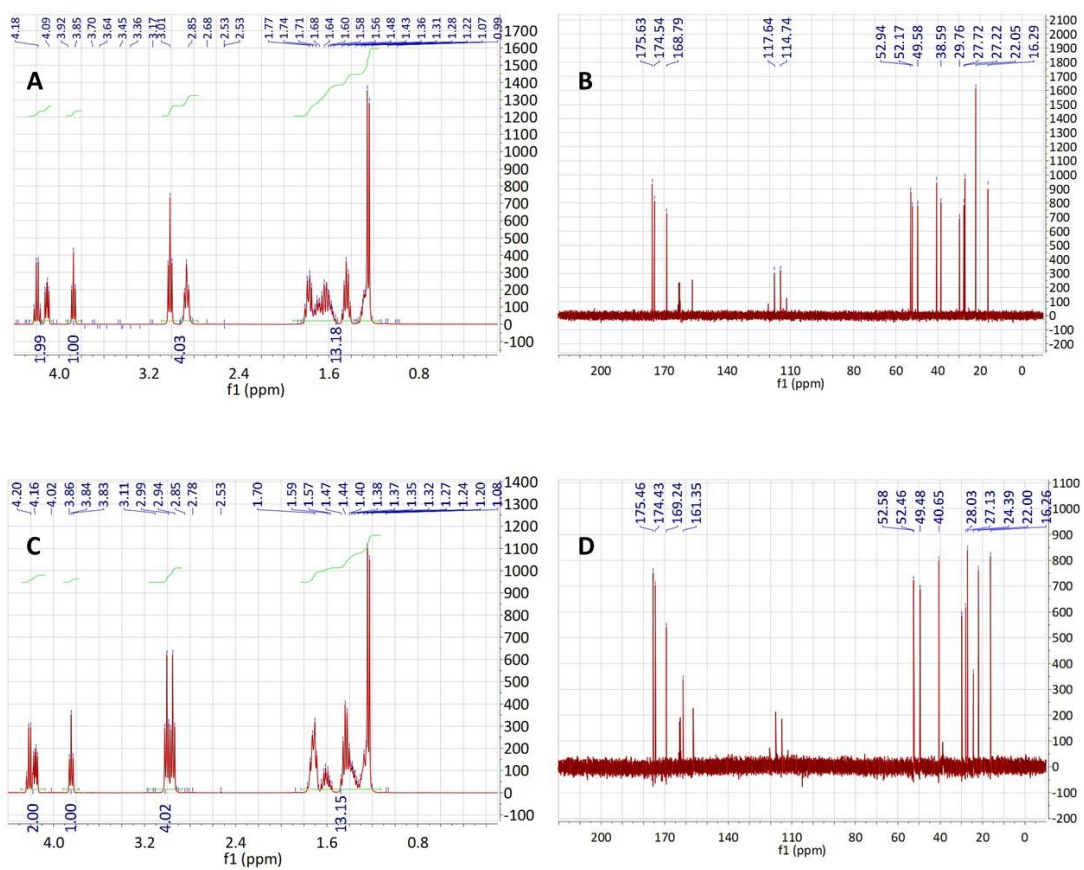
F, lane 1, DNA marker; lane 2, wild type (2499 bp); lane 3, *phtL*<sup>-</sup> mutant (648 bp).



**Figure S3** Extracted ion chromatograms of PHTs from culture supernatants of gene in-frame deletion mutants. Extracted with a tolerance of 0.5 Da. A, 1448A (PHT producer, wild type); B, *phtU* mutant strain CL001; C, *phtU* mutant strain CL001 fed with L-Ala -L-hArg; D, *phtU* mutant strain CL001 fed with L-Orn- L-Ala- L-hArg; E, *phtU* mutant strain CL001 fed with L-Cit- L-Ala- L-hArg; F, *phtU* mutant gene complementation strain CL004, with infusion gene  $P_{phtA}$ -ORF $_{phtU}$  in pWHU2004; G, *phtQ* mutant strain CL002; H, *phtQ* mutant gene complementation strain CL005, with infusion gene  $P_{phtA}$ -ORF $_{phtQ}$  in pWHU2005.



**Figure S4** Extracted ion chromatograms of  $L$ -Cit-  $L$ -Ala-  $L$ - $h$ Arg from mutant strains culture supernatants. Extracted with a tolerance of 0.5 Da. A, 1448A (PHT producer, wild type); B, *phtQ* mutant strain CL002 fed with  $L$ -Orn-  $L$ -Ala-  $L$ - $h$ Arg as substrate; C, *phtU* mutant strain CL001 fed with  $L$ -Orn-  $L$ -Ala-  $L$ - $h$ Arg as substrate.



**Figure S5**  $^1\text{H}$  spectra of tripeptide standards  $\text{L-Orn-L-Ala-L-hArg}$  (A) and  $\text{L-Cit-L-Ala-L-hArg}$  (C) and  $^{13}\text{C}$  NMR spectra of  $\text{L-Orn-L-Ala-L-hArg}$  (B) and  $\text{L-Cit-L-Ala-L-hArg}$  (D).