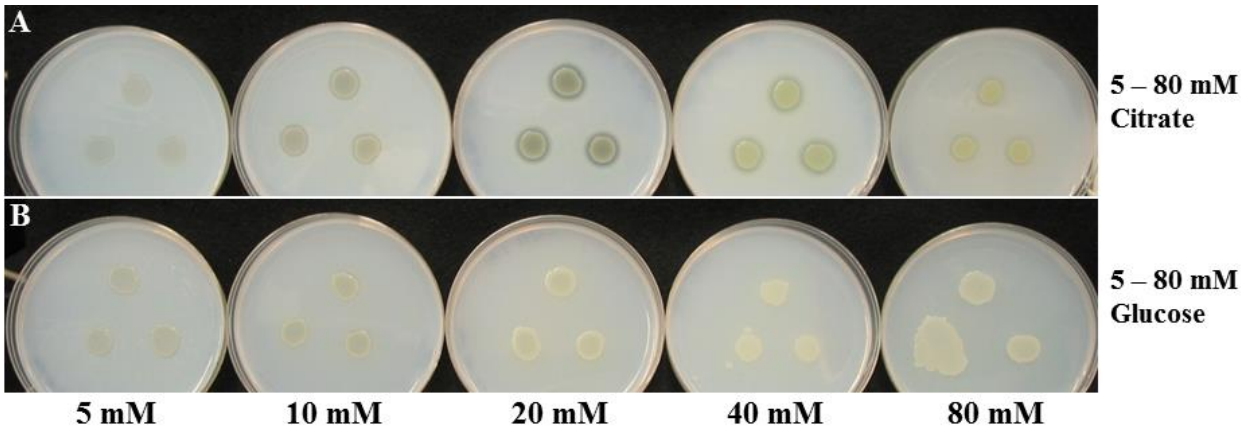
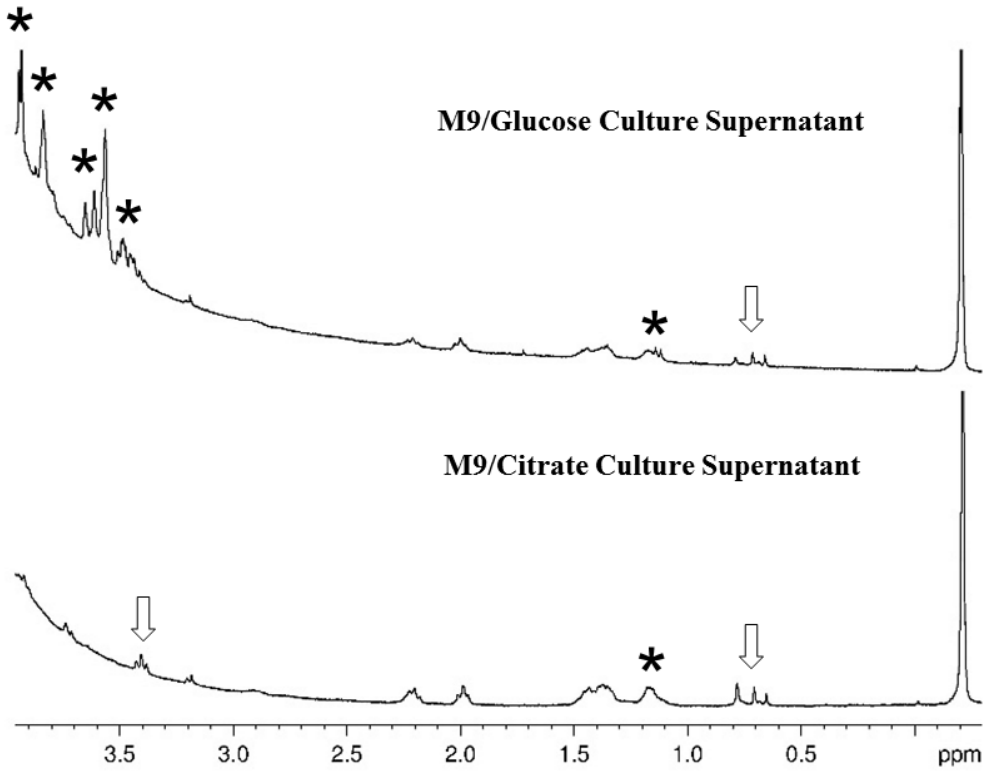
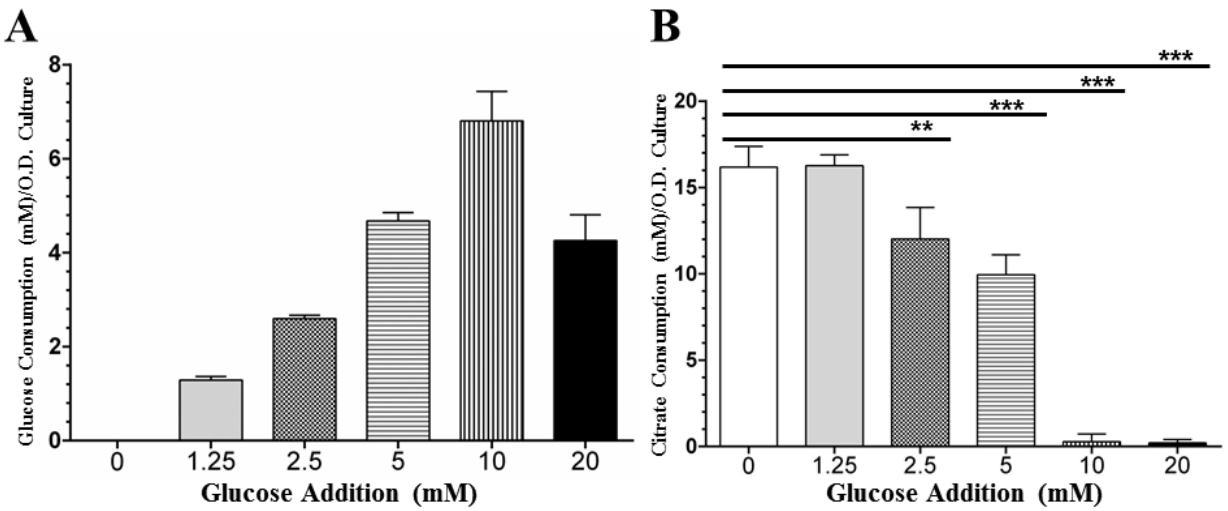
Supplemental Material



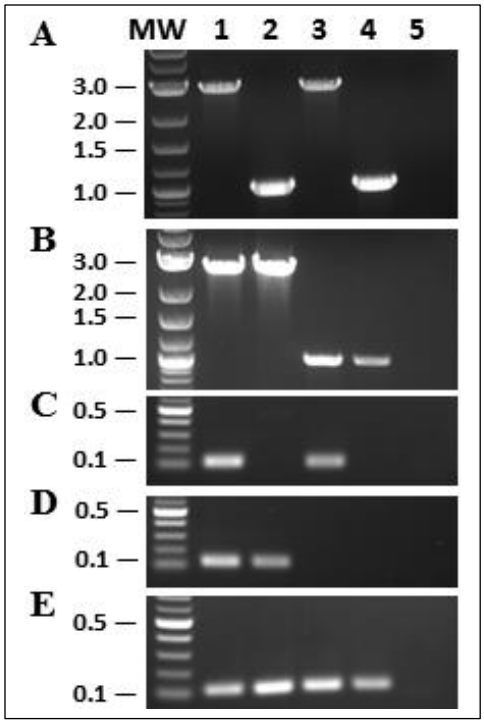
SUPPLEMENTAL FIGURE S1. Impact of citrate and glucose concentrations on Impranil clearing. *P. protegens* strain Pf-5 grown on M9 agar with 3 g/L Impranil containing **(A)** 5 to 80 mM citrate and **(B)** 5 to 80 mM glucose after 48 hours of growth at 27 °C.



SUPPLEMENTAL FIGURE S2. Identification of Impranil degradation products in Pf-5 culture supernatants post-Impranil clearing assays. Cell-free culture supernatants of *P. protegens* strain Pf-5 grown in M9 medium supplemented with different carbon sources were used in Impranil clearing assay. Post-Impranil clearing samples were subjected to NMR analysis to quantify concentration of soluble PU hydrolysis products. Representative ^{1}H NMR data from the degradation of Impranil using *P. protegens* Pf-5 cultures supernatants grown in M9 media supplemented with either 20 mM glucose (top panel) or 20 mM citrate (bottom panel) are shown to the same ordinate scale. Peaks used for integrated area calculations are indicated with open arrows. * represents signals that are also present in the initial supernatant without exposure to Impranil.



SUPPLEMENTAL FIGURE S3. Glucose and citrate consumption after 22 hours growth in M9 + 20 mM citrate media supplemented with varying concentrations of glucose. Cultures of strain Pf-5 were grown for 22 hours in M9/citrate medium supplemented with varying concentrations of glucose. Cell-free supernatants of these cultures were subjected to HPLC analysis to determine levels of glucose **(A)** and citrate **(B)** consumption. **, $p<0.001$; ***, $p<0.0001$.



SUPPLEMENTAL FIGURE S4. Verification of mutants by PCR. Genomic DNA of *P. protegens* strain Pf-5 wild type (Lane 1), *pueA* deletion mutant (Pf5Δ*pueA*, Lane 2), *pueB* deletion mutant (Pf5Δ*pueB*, Lane 3), and *pueA/pueB* double deletion mutant (Pf5Δ*pueAB*, Lane 4) were analyzed by PCR to confirm their identities. Negative control samples (Lane 5) in which water was used as template were included for each analysis. **(A)** PCR amplification using primers corresponding to regions outside of *pueA* gene (Pf5pueATestF and Pf5pueATestR) resulted in the appearance of a 2.85 kB or a 1.0 kB product for wild type sequence or *pueA* deletion, respectively. **(B)** Similarly, using primers corresponding to regions outside of *pueB* gene (Pf5pueBTestF and Pf5pueBTestR) resulted in the appearance of a 2.69 kB or a 1.0 kB product for wild type sequence or *pueB* deletion, respectively. **(C-D)** Primers corresponding to *pueA* (pueAF and pueAR) (C) and *pueB* (pueBF and pueBR) (D) gene sequences were also used to further confirm either the presence (0.1 kB band) or the removal (lack of PCR products) of the respective genes. (E) Primers corresponding to *ropD* gene (rpoDF and rpoDR) were used as positive control for PCR reaction, which yield a 0.1 kB product band.

Identification of putative Cbr/crc catabolite repression system in *P. protegens* Pf-5.

Using the blastp algorithm (http://blast.ncbi.nlm.nih.gov/Blast.cgi), the amino acid sequence from Crc of *P. aeruginosa* strain PAO1 (AAC44428.1) was compared to protein sequences derived from the *P. protegens* strain Pf-5 genome. Highest similarity was found with exodeoxyribonuclease III (YP_263115.1). A pairwise alignment of both proteins using the Stretcher software resulted in 88.8 % identity.

Using blastn algorithm (http://blast.ncbi.nlm.nih.gov/Blast.cgi), we found two stretches of nucleotide sequences not yet annotated in the Pf-5 genome that possibly encode regulatory RNAs similar to CrcZ from *Pseudomonas aruginosa* PAO1 (GeneID 9793394: 63-469). Highest identities (76 % and 69 %) were found with a nucleotide sequence (6040962-6041319) located between the genes encoding the two component response regulator CbrB and the poly(A) polymerase PcnB, and with a nucleotide sequence (4546200 – 4546573) positioned between the genes encoding a possible hydroxymethylglutaryl-CoA lyase (LiuE) and a potential long-chain-fatty-acid-CoA ligase, respectively.

Supplemental Table S1. Mascot Database Search Results of Band 6A.

Protein Best Hit

gi|4558791 **Mass:** 64784 **Score:** 390 **Queries matched:** 6

polyurethanase esterase A [*Pseudomonas chlororaphis*]

gi|70730566 **Mass:** 64900 **Score:** 390 **Queries matched:** 6

polyurethanase A [*Pseudomonas fluorescens* Pf-5]

Peptide Summary Report

Start - End[a]	Observed	Mr(expt)	Mr(calc)	ppm	Miss	Score	Expect	Rank	Peptide[b]
127 - 141	1676.7279	1675.7206	1675.8318	-66.34	0	32	50	1	K.YDAQGHLESIGIAFR.G
247 - 259	1535.6846	1534.6773	1534.7780	-65.59	0	52	0.41	1	K.VLNIGYENDPVFR.A
337 - 350	1457.6620	1456.6547	1456.7522	-66.89	0	75	0.0023	1	K.DSTIVVANLSDPAR.A
351 - 359	1189.5187	1188.5114	1188.5887	-65.04	0	69	0.0077	1	R.ASTWVQDLNR.N
367 - 391	2642.0713	2641.0640	2641.2256	-61.18	0	81	0.00068	1	K.GSTFIIGSDGNDLIQGGSGNDYLEGR.A
480 - 491	1337.5940	1336.5867	1336.6735	-64.94	0	84	0.00023	1	K.AGGQLTQYASSVR.G

[a]"Start – End" numbering correspond to the amino acid residue numbers of Pf-5 PueA protein that matches the corresponding peptide.

[b]Matching tryptic-fragment peptide sequences are underlined.

Supplemental Table S2. Mascot Database Search Results of Band 1B.

Protein Best Hit

gi|346642991 **Mass:** 59059 **Score:** 389 **Queries matched:** 6

polyurethanase B [Pseudomonas fluorescens Pf-5]

Peptide Summary Report

Start - End[a]	Observed	Mr(expt)	Mr(calc)	ppm	Miss	Score	Expect	Rank	Peptide[b]
79 - 93	1626.7526	1625.7453	1625.8413	-59.04	0	41	5.3	1	K.YDASGQLLSIGISFR.G
100 - 119	2159.8811	2158.8738	2158.9920	-54.72	0	49	0.84	1	K.DGINDLQAAFVSGFADNYSR.L
268 - 279	1443.6277	1442.6204	1442.7041	-58.03	0	92	3.8e-05	1	R.LINSDFYDLTSR.D
280 - 293	1504.7078	1503.7005	1503.7893	-59.02	1	45	2.2	1	R.DSTVVISNLSEGKR.D
328 - 340	1324.4631	1323.4558	1323.5327	-58.12	0	75	0.00064	1	K.GNDYLDGGAGDDR.F
367 - 381	1655.7068	1654.6995	1654.7951	-57.75	0	87	0.00014	1	K.NFSIANDGDGTLYIR.D

[a]"Start – End" numbering correspond to the amino acid residue numbers of Pf-5 PueB protein that matches the corresponding peptide.

[b]Matching tryptic-fragment peptide sequences are underlined.