

**Supplementary Tables and
Caption to Supplementary Figures**

Table S1. Summary of *P. aeruginosa* *ppk1* mutant gain and lost phenotypes.

Test	Difference	Mode of action
	Gain phenotype	
L-Aspartic acid	68	C-Source, amino acid
L-Lactic acid	64	C-Source, amino acid
L-Arginine	50	C-Source, carboxylic acid
His-Met	61	N-Source, peptide
Tyr-His	53	N-Source, peptide
L-Cysteine	51	N-Source, amino acid
pH 9.5 + Glycine	69	pH, deaminase
pH 9.5 + L-Phenylalanine	66	pH, deaminase
pH 9.5 + L-Serine	61	pH, deaminase
Carbenicillin	158	Wall, lactam
Cefsulodin	80	Wall, cephalosporin
Ruthenium red	78	Respiration, mitochondrial Ca ²⁺ porter
Erythromycin	67	Protein synthesis, 50S ribosomal subunit, macrolide
	Lost phenotype	
D-Gluconic acid	-112	C-Source, carboxylic acid
D-Mannitol	-68	C-Source, carbohydrate
L-Isoleucine	-57	C-Source, amino acid
L-Valine	-60	N-Source, amino acid
L-Methionine Sulfone	-69	S-Source, organic
Orphenadrine	-78	Anti-cholinergic
Patulin	-534	Microtubulin polymerization inhibitor, antifungal
2,2'-Dipyridyl	-401	Chelator, lipophilic
Sodium Arsenite	-332	Toxic anion
Sodium Arsenate	-305	Toxic anion, P04 analog
Dichlofluanid	-271	Fungicide, phenylsulphamide
Thiamphenicol	-269	Protein synthesis, amphenicol
Chloramphenicol	-209	Protein synthesis, amphenicol
Chlorodinitrobenzene	-182	Oxidizes sulphhydryls, depletes glutathione
8-Hydroxyquinoline	-181	Chelator, lipophilic
Nafcillin	-163	Wall, lactam
Antimony (III) chloride	-158	Toxic cation
1,10-Phenanthroline monohydrate	-152	Chelator, lipophilic
Captan	-152	Fungicide, carbamate
Sulfathiazole	-148	Folate antagonist, PABA analog
Gallic acid	-141	Respiration, ionophore, H ⁺
Streptomycin	-140	Protein synthesis, 30S ribosomal subunit, aminoglycoside
Poly-L-lysine	-140	Membrane, detergent, cationic
Protamine sulfate	-127	Membrane, nonspecific binding
Sulfadiazine	-124	Folate antagonist, PABA analog
Oxacillin	-113	Wall, lactam
Myricetin	-113	DNA & RNA synthesis, polymerase inhibitor
Nickel chloride	-110	Toxic cation
Diamide	-106	Oxidizes sulphhydryls, depletes glutathione
Plumbagin	-99	Oxidizing agent
Oxophenylarsine	-98	Tyrosine phosphatase inhibitor
Trans-Cinnamic acid	-67	Respiration, ionophore, H ⁺
EDTA	-66	Chelator, hydrophilic
Glycine hydroxamate	-63	tRNA synthetase
L-Glutamic acid g-monohydroxamate	-63	tRNA synthetase

Table S2. Summary of *P. aeruginosa* *ppk2* mutant gain and lost phenotypes.

Test	Difference	Mode of action
	Gain phenotype	
L-Arginine	75	C-Source, amino acid
His-Met	56	N-Source, peptide
His-His	51	N-Source, peptide
Phospho-Glycolic acid	57	P-Source, organic
pH 9.5 + L-Histidine	84	pH, deaminase
pH 9.5 + L-Norvaline	62	pH, deaminase
Ruthenium red	110	Respiration, mitochondrial Ca ²⁺ porter
Sodium Selenite	82	Toxic anion
Erythromycin	80	Protein synthesis, 50S ribosomal subunit, macrolide
Carbenicillin	71	Wall, lactam
Carbenicillin	40	Wall, lactam
	Lost phenotype	
D-Gluconic acid	-106	C-Source, carboxylic acid
D-Mannitol	-57	C-Source, carbohydrate
L-Valine	-58	N-Source, amino acid
L-Methionine Sulfone	-95	S-Source, organic
pH 4.5 + 5-Hydroxy-L-Lysine	-61	pH, decarboxylase
Patulin	-495	Microtubulin polymerization inhibitor, antifungal
2,2'-Dipyridyl	-397	Chelator, lipophilic
Sodium Arsenate	-321	Toxic anion, P04 analog
Sodium Arsenite	-242	Toxic anion
8-Hydroxyquinoline	-230	Chelator, lipophilic
Thiamphenicol	-193	Protein synthesis, amphenicol
Chlorodinitrobenzene	-187	Oxidizes sulphydryls, depletes glutathione
Chloramphenicol	-173	Protein synthesis, amphenicol
Nafcillin	-172	Wall, lactam
1,10-Phenanthroline monohydrate	-171	Chelator, lipophilic
Oxophenylarsine	-161	Tyrosine phosphatase inhibitor
Antimony (III) chloride	-153	Toxic cation
Orphenadrine	-146	Anti-cholinergic
Sulfathiazole	-142	Folate antagonist, PABA analog
Acriflavine	-140	DNA intercalator, inhibits RNA
Glycine hydroxamate	-136	tRNA synthetase
Oxacillin	-130	Wall, lactam
Myricetin	-122	DNA & RNA synthesis, polymerase inhibitor
Dichlofluanid	-119	Fungicide, phenylsulphamide
Plumbagin	-118	Oxidizing agent
Proflavine	-116	DNA intercalator, inhibits RNA
Gallic acid	-115	Respiration, ionophore, H ⁺
Captan	-113	Fungicide, carbamate
Protamine sulfate	-109	Membrane, nonspecific binding
Sulfadiazine	-107	Folate antagonist, PABA analog
Dequalinium chloride	-102	Ion channel inhibitor, K ⁺
2,4-Dinitrophenol	-93	Respiration, ionophore, H ⁺
trans-Cinnamic acid	-90	Respiration, ionophore, H ⁺
EDTA	-88	Chelator, hydrophilic
Nickel chloride	-80	Toxic cation
Poly-L-lysine	-63	Membrane, detergent, cationic

Table S3. Summary of unique *P. aeruginosa* *ppk1* mutant gain and lost phenotypes.

Test	Difference	Mode of action
		Gain phenotype
L-Aspartic acid	68	C-Source, amino acid
L-Lactic acid	64	C-Source, amino acid
Tyr-His	53	N-Source, peptide
L-Cysteine	51	N-Source, amino acid
pH 9.5 + Glycine	69	pH, deaminase
pH 9.5 + L-Phenylalanine	66	pH, deaminase
pH 9.5 + L-Serine	61	pH, deaminase
Cefsulodin	80	Wall, cephalosporin
Test	Difference	Mode of action
		Lost phenotype
L-Isoleucine	-57	C-Source, amino acid
Streptomycin	-140	Protein synthesis, 30S ribosomal subunit,
Diamide	-110	aminoglycoside
L-Glutamic acid g-monohydroxamate	-63	Oxidizes sulphydryls, depletes glutathione tRNA synthetase

Table S4. Summary of unique *P. aeruginosa* *ppk2* mutant gain and lost phenotypes.

Test	Difference	Mode of action
		Gain phenotype
His-His	51	N-Source, peptide
Phospho-Glycolic acid	57	P-Source, organic
pH 9.5 + L-Histidine	84	pH, deaminase
pH 9.5 + L-Norvaline	62	pH, deaminase
Sodium Selenite	82	Toxic anion
Carbenicillin	71	Wall, lactam
Test	Difference	Mode of action
		Lost phenotype
pH 4.5 + 5-Hydroxy-L-Lysine	-61	pH, decarboxylase
Acriflavine	-140	DNA intercalator, inhibits RNA
Proflavine	-116	DNA intercalator, inhibits RNA
Dequalinium chloride	-102	Ion channel inhibitor, K ⁺
2,4-Dinitrophenol	-93	Respiration, ionophore, H ⁺

Table S5. Summary of antimicrobial susceptibility testing (M.I.C.EvaluatorTM (Oxoid) and Etest® (Biomerieux) for *P. aeruginosa* *ppk1* and *ppk2* mutant strains.

Strain	Antibiotics ¹									
	E	IPM	CIP	VA	CN	AMC	SM	CL	KM	RI
PAO1 WT	16	2	0.75	>256	1	>256	8	>256	32	>32
PAO1 <i>ppk1</i>	16	0.5	0.12	>256	1	>256	3	32	24	16
PAO1 <i>ppk2</i>	16	0.5	0.12	>256	1	>256	3	32	32	3

¹ MIC (Minimal inhibitory concentration) parameter for antibiotics Erythromycin (E), Imipenen (IPM), Ciprofloxacin (CIP), Vancomycin (VA), Gentamicin (CN), Amoxicillin (AMC), Streptomycin (SM), Chloramphenicol (CL), Kanamycin (KM) and Rifampicin (RI).

Supplementary Figures

Figure S1. Consensus graphical profile of metabolic and sensitivity tests of *P. aeruginosa* *ppk1* (A) and *ppk2* (B) mutants.

Significant changes are enclosed in boxes. Yellow indicates that respiration rate of the wild type and mutant strains were similar. Red indicates faster respiration rate of the wild type (lost phenotype). Green indicates faster respiration rate of the mutant (gain phenotype). The quantitative difference values are shown in Table 1 and Supplementary tables.

Figure S2. Clustering analysis of metabolic tests (PM1-PM8) and pH response (PM10) from *P. aeruginosa* PAO1 *ppk1* and *ppk2* mutants.

Gain (blue) and lost (yellow) phenotypes were standardized by dividing the respiration value of the mutants by the value of the wild type strain, for each phenotype. The results are shown separately for the different categories: carbon (C) sources, nitrogen (N) sources, phosphorus (P) and sulfur sources (S), peptide nitrogen (N) sources and pH response.

Figure S3. Venn diagrams of phenotypic microarrays results from polyP synthesis mutants from *E. coli* K12 and *P. aeruginosa* PAO1.

The numbers indicate the total phenotypes gained (A) or lost (B) between *P. aeruginosa* *ppk1* and *ppk2* mutants and *E. coli* Δ *ppk1* mutant. Phenotypic microarray results from *E. coli* Δ *ppk1* mutant were performed in a previous work (Unpublished results).