Supporting Information

Table S1. Summary of homologues for each *pqq* gene product.

Products of *pqq* genes	Average length of seeds [1] (amino acids)	Pfam domain	Number of global homologues	Minimum identity in homologues (%)
PqqA	27	PqqA	124	22.7
PqqB	302	No domain from Pfam, but a PQQ_syn_pqqB domain from CDD [2]	797	16.6
PqqC	246	TENA_THI-4	518	9.2
PqqD	93	PqqD	518	7.4
PqqE	372	Radical_SAM	662	18
PqqF	576	peptidase_M16, peptidase_M16_C	572	13.4

[1] See **Table 1** for the description of seeds.
[2] CDD: conserved domain database of NCBI.

Table S2. Species that contain PqqB-E.[a]

Species	Pathogenicity	Set of Pqq synthesis genes
**Acetobacter pasteurianus* IFO 3283-01-42C		ABCDE
**Acidiphilium cryptum* (strain JF-5)		ABCDE
**Acidobacterium sp.* MP5ACTX8		ABCDE
**Acinetobacter baumannii* ATCC 19606	Opportunistic human pathogen	ABCDE
**Acinetobacter calcoaceticus*		ABCDE
Acinetobacter haemolyticus ATCC 19194	Rare human pathogen	BCDE
Acinetobacter sp. RUH2624		ABCDE
**Agrobacterium radiobacter* (strain K84/ATCC BAA-868)		ABCDE
**Arcobacter nitrofigilis* (strain ATCC 33309/DSM 7299/LMG 7604/NCTC 12251/CI)		ABCDEF
**Azospirillum sp.* (strain B510)		ABCDEF
**Azotobacter vinelandii* (strain DJ/ATCC BAA-1303)		ABCDEF
**Azoarcus sp*. (strain BH72)	Plant symbiont	ABCDEF
**Beijerinckia indica subsp. indica* (strain ATCC 9039/DSM 1715/NCIB 8712)		ABCDEF
[+]*beta proteobacterium* KB13		BCDEF
**Bradyrhizobium japonicum*	Plant symbiont	ABCDEF
**Bradyrhizobium sp.* (strain BTAi1/ATCC BAA-1182)	Plant symbiont	ABCDEF
**Burkholderia ambifaria* MC40-6		ABCDE
**Burkholderia cenocepacia* (strain HI2424)	Human cystic fibrosis pathogen	ABCDE
Burkholderia cepacia (strain J2315/LMG 16656)	Animal pathogen in Mammalia and Insecta, Plant pathogen	ABCDE
Burkholderia dolosa AUO158		CDE [b]
**Burkholderia glumae* (strain BGR1)	Plant pathogen	ABCDE
**Burkholderia graminis* C4D1M		BCDE
**Burkholderia multivorans* (strain ATCC 17616/ 249)	Animal pathogen in Mammalia	ABCDE
**Burkholderia phymatum* (strain DSM 17167/STM815)	Plant symbiont	ABCDE
**Burkholderia sp.* CCGE1001		ABCDE
**Burkholderia vietnamiensis* (strain G4/LMG 22486)		ABCDE
**Burkholderia xenovorans* (strain LB400)		ABCDE
**Chromohalobacter salexigens* (strain DSM 3043/ATCC BAA-138/NCIMB 13768)		ABCDE
Chthoniobacter flavus Ellin428		BCDEF
Citreicella sp. SE45		BCDEF
**Colwellia psychrerythraea* (strain 34H/ATCC BAA-681)	Plant pathogen	ABCDE
**Cronobacter turicensis* (strain DSM 18703/LMG 23827/z3032)	Neonatal pathogen	ABCDEF
**Cupriavidus taiwanensis* (strain R1/LMG 19424)	Plant pathogen	ABCDE
**Dechloromonas aromatica* (strain RCB)		ABCDEF
**Dinoroseobacter shibae* (strain DFL 12)	Animal symbiont	ABCDEF
**Enterobacter intermedius*	Rare opportunistic pathogen	ABCDEF
**Enterobacter sakazakii* (strain ATCC BAA-894)	Neonatal pathogen	ABCDEF

**Erwinia amylovora* (strain ATCC 49946/CCPPB 0273/Ea273/27-3)	Plant pathogen	ABCDEF
**Erwinia pyrifoliae*	Plant pathogen	ABCDEF
**Erwinia tasmaniensis* (strain DSM 17950/Et1/99)	Plant commensal	ABCDEF
Fulvimarina pelagi HTCC2506		ABCDE
+gamma proteobacterium HTCC2207		BCDE
**Geodermatophilus obscurus* (strain ATCC 25078/DSM 43160/JCM 3152/G-20)		ABCDE
**Gluconacetobacter diazotrophicus* (strain ATCC 49037/DSM 5601/PAl5)	Plant symbiont	ABCDE
Gluconacetobacter hansenii ATCC 23769		BCDE
**Gluconobacter oxydans*		ABCDE
Gluconobacter sp. H24		ABCDE
**Granulibacter bethesdensis* (strain ATCC BAA-1260/CGDNIH1)	First reported case of invasive human disease caused by any of the Acetobacteraceae	ABCDE
Grimontia hollisae CIP 101886	Potential agent of gastroenteritis and bacteraemia in the Mediterranean area	BCDE
**Hydrogenobacter thermophilus* (strain DSM 6534/ IAM 12695/TK-6)		BCDEF
**Hyphomicrobium denitrificans* (strain ATCC 51888/DSM 1869/NCIB 11706/TK 0415)		ABCDEF
**Klebsiella pneumoniae*	Rare opportunistic pathogen	ABCDEF
Klebsiella sp. 1_1_55	Opportunistic pathogen	BCDEF
**Klebsiella variicola* (strain At-22)		ABCDEF
**Leptothrix cholodnii* (strain ATCC 51168/LMG 8142/SP-6)		ABCDE
**Limnobacter sp*. MED105		ABCDEF
Lutiella nitroferrum 2002		BCDEF
marine gamma proteobacterium HTCC2143		BCDE
Marinobacter algicola DG893	Plant symbiont	ABCDE
Marinobacter sp. ELB17		BCDE
**Methylacidiphilum infernorum* (isolate V4)		ABCDE
**Methylibium petroleiphilum* (strain PM1)		ABCDE
**Methylobacillus flagellatus* (strain KT/ATCC 51484/DSM 6875)		ABCDEF
**Methylobacterium chloromethanicum* (strain CM4/NCIMB 13688)		ABCDEF
**Methylobacterium extorquens* (strain ATCC 14718/DSM 1338/AM1)		ABCDEF
**Methylobacterium nodulans* (strain ORS2060/ LMG 21967)	Plant saprophyte and symbiont	ABCDEF
**Methylobacterium populi* (strain ATCC BAA-705/ NCIMB 13946/BJ001)	Plant endophyte	ABCDEF
**Methylobacterium radiotolerans* (strain ATCC 27329/DSM 1819/JCM 2831)	Plant symbiont	ABCDEF
**Methylobacterium sp.* (strain 4-46)		ABCDEF
**Methylocella silvestris* (strain BL2/DSM 15510/ NCIMB 13906)		ABCDEF
**Methylococcus capsulatus*		ABCDEF
+Methylophaga thiooxidans DMS010		BCDE

[+]*Methylosinus trichosporium* OB3b		BCDEF
**Methylotenera mobilis* (strain JLW8/ATCC BAA-1282/DSM 17540)		ABCDEF
**Methylotenera sp.* (strain 301)		ABCDEF
**Methylovorus sp.* (strain SIP3-4)		ABCDEF
**Mycobacterium smegmatis* (strain ATCC 700084/ mc(2, #5076)155)	Animal commensal in Mammalia	ABCDE
Neptuniibacter caesariensis		BCDE
**Nitrobacter winogradskyi* (strain Nb-255/ATCC 25391)		ABCDEF
Nitrococcus mobilis Nb-231		ABCDEF
**Nitrosococcus halophilus* (strain Nc4)		ABCDEF
**Nitrosococcus oceani* ATCC19707		ABCDEF
**Pantoea ananatis*	Plant pathogen	ABCDEF
**Pantoea sp.* At-9b		ABCDEF
**Paracoccus denitrificans* (strain Pd 1222)		ABCDEF
**Polaromonas naphthalenivorans* (strain CJ2)		ABCDEF
**Pseudomonas aeruginosa*	Opportunistic human pathogen	ABCDEF
**Pseudomonas entomophila* (strain L48)	Insect pathogen	ABCDEF
**Pseudomonas fluorescens*	Potential pathogen to birds	ABCDEF
**Pseudomonas mendocina* (strain ymp)	Rare human pathogen	ABCDEF
**Pseudomonas putida* (strain KT2400)		ABCDEF
[+]*Pseudomonas savastanoi pv. savastanoi* NCPPB 3335	Tumor-inducing pathogen	ABCDEF
**Pseudomonas syringae pv. phaseolicola* (strain 1448A/Race 6)	Plant symbiont	ABCDEF
Rahnella aquatilis	Rarely opportunistic pathogen	ABCDEF
[+]*Ralstonia eutropha* (strain ATCC 17699/H16/ DSM 428/Stanier 337)		ABCDE
**Ralstonia pickettii* (strain 12D)	Opportunistic nocosomial pathogen	ABCDEF
[+]*Rhizobium meliloti*	Plant symbiont	ABCDE
**Rhizobium sp.* (strain NGR234)	Plant symbiont	ABCDEF
**Rhodobacter sphaeroides* (strain ATCC 17023/ 2.4.1/NCIB 8253/DSM 158)		ABCDEF
**Rhodobacterales bacterium* HTCC 2654		ABCDEF
**Rhodomicrobium vannielii* ATCC 17100		ABCDE
**Rhodopseudomonas palustris*		ABCDEF
Rickettsiella grylli	Athropod pathogen	BCDEF
**Roseobacter denitrificans* (strain ATCC 33942/ OCh 114) (*Erythrobacter sp.* (strain OCh 114))		ABCDEF
**Roseobacter litoralis* Och 149		ABCDEF
**Roseobacter sp.* AzwK-3b		ABCDEF
**Roseovarius sp.* HCC2601		ABCDEF
**Saccharopolyspora erythraea* (strain NRRL 23338)		ABCDEF
**Sagittula stellata* E-37		ABCDEF
Serratia marcescens	Opportunistic human pathogen	ABCDE
Serratia odorifera 4Rx13	Rare opportunistic pathogen	ABCDEF
**Shewanella woodyi* (strain ATCC 51908 / MS32)		ABCDEF

**Silicibacter pomeroyi*		ABCDEF
**Sinorhizobium medicae* (strain WSM419)		ABCDE
**Stackebrandtia nassauensis* (strain DSM 44728/NRRL B-16338/NBRC 102104/LLR-40K-21)		ABCDE
**Starkeya novella* (strain ATCC 8093/DSM 506/CCM 1077/IAM 12100/NBRC 12443/NCIB 9113)		ABCDEF
**Streptomyces bingchenggensis* (strain BCW-1)		ABCDEF
[+]*Streptomyces rochei*		ABCDE
Thiomonas sp. (strain 3As)		ABCDE
**Variovorax paradoxus* (strain S110)		ABCDEF
**Verminephrobacter eiseniae* (strain EF01-2)	Animal endosymbiont in *Eisenia foetida*	ABCDEF
Vibrio sp. RC586		BCDE
**Xanthobacter autotrophicus* (strain ATCC BAA-1158/Py2)		ABCDEF
**Xanthomonas axonopodis pv. citri* (Citrus canker)	Plant pathogen	ABCDE
**Xanthomonas campestris pv. Campestris*	Plant pathogen	ABCDE
**Xanthomonas oryzae pv. Oryzae*	Rice bacterial blight pathogen	ABCDE

[a] Species with complete genome sequence available are labeled with *. Species without a completely sequenced genome but with the PQQ operon gene order information are labeled with +.

[b] This is the sole entry lacking B.

Table S3. Location of PqqA homologues mis-annotated as intergenic regions in the NCBI database.

Genome	Start position in genome	End position in genome	Distance to PqqB
Acidiphilium cryptum (strain JF-5)	2308383	2308324	2221863bp
Acidobacterium sp. MP5ACTX8	3186608	3186667	22 bp
Agrobacterium radiobacter (strain K84)	1497718	1497771	62 bp
Arcobacter nitrofigilis (strain ATCC 33309 / DSM 7299 / LMG 7604 / NCTC 12251 / CI)	2810583	2810524	124 bp
Azoarcus sp. (strain BH72)	1279504	1279563	151 bp
Azospirillum sp. (strain B510)	1287841	1287900	148 bp
Azotobacter vinelandii (strain DJ / ATCC BAA-1303)	4201776	4201708	49 bp
Beijerinckia indica subsp. indica (strain ATCC 9039 / DSM 1715 / NCIB 8712)	3779214	3779140	35 bp
Burkholderia ambifaria (MC40-6)	438611	438682	68 bp
Burkholderia cenocepacia (strain HI2424)	2155918	2155847	65 bp
Burkholderia multivorans (strain ATCC 17616/249)	227954	228025	67 bp
Burkholderia phymatum (strain STM815)	1842594	1842665	750928 bp
Burkholderia vietnamiensis (strain G4 / LMG 22486)	1170047	1169976	68 bp
Burkholderia xenovorans (strain LB400)	3030290	3030361	2369427 bp
Chromohalobacter salexigens (strain DSM 3043 / ATCC BAA-138 / NCIMB 13768)	945885	945956	14 bp
Cronobacter turicensis (strain DSM 18703 / LMG 23827 / z3032)	944075	944010	69bp
Dechloromonas aromatica (strain RCB)	1826581	1826640	79bp
Enterobacter sakazakii (strain ATCC BAA-894)	2948005	2948070	61 bp
Granulibacter bethesdensis (strain CGDNIH1)	2162621	2162550	129 bp
Methylibium petroleiphilum (strain PM1)	3236227	3236295	476661bp
Methylobacterium chloromethanicum (strain CM4 / NCIMB 13688)	2254953	2254867	51bp
Methylobacterium populi (strain ATCC BAA-705 / NCIMB 13946 / BJ001)	1905787	1905701	52bp
Methylocella silvestris (strain BL2 / DSM 15510 / NCIMB 13906)	1231994	1232068	49bp
Nitrobacter winogradskyi (strain Nb-255 / ATCC 25391)	771485	771414	69bp
Nitrosococcus oceani ATCC19707	2976128	2976193	77 bp
Paracoccus denitrificans (strain Pd 1222)	2358331	2358260	88 bp
Polaromonas naphthalenivorans (strain CJ2)	772866	772801	86 bp
Ralstonia eutropha (strain H16)	1188199	1188134	143 bp
Rhodopseudomonas palustris (strain CGA009)	98346	98414	131 bp
Shewanella woodyi (strain ATCC 51908 / MS32)	2764866	2764798	79 bp
Sinorhizobium medicae (strain WSM419)	370213	370136	60 bp
Starkeya novella (strain DSM 506)	1091028	1091102	42 bp
*Verminephrobacter eiseniae (*strain EF01-2)	4421713	4421778	177 bp
Xanthobacter autotrophicus (strain ATCC BAA-1158 /	2031196	2031125	59 bp

Py2)

Xanthomonas axonopodis pv. citri (Citrus canker)	3661812	3661871	96 bp
*Xanthomonas campestris pv. Campestri*s str. ATCC 33913	3492588	3492647	79 bp
Xanthomonas oryzae pv. Oryzae	3323436	3323495	111 bp

Table S4. Result of Discern for PqqB from *K. pneumoniae* and PhnP from *E. coli* sequences. Homologous residues in PqqB and PhnP are based on the multiple sequence alignment and are plotted in the structure shown in **Figure 4**.

PqqB KP				PhnP EC			
Residue	Position	Rank from Discern	Comments	Residue	Position	Rank from Discern	Comments
His	93	1	putative metal binding	His	81	4	ligand MnB
Asp	88	2		His	76	6	ligand MnA
His	269	3	putative metal binding	His	222	7	ligand MnB
Arg	37	4		Arg	37	15	
Asp	297	5		Asp	245	13	
Asp	92	6	putative metal binding	Asp	80	8	ligand MnB
Asp	59	7		Asp	59	60	
Ser	41	8		Ser	41	21	
Asp	220	9		Asp	187	10	
Gln	39	10		Pro	39	70	
His	244	11					
Gln	90	18		His	78	2	ligand MnA
Asn	55	19		Asp	54	3	
Ser	163	37		His	143	5	ligand MnA
Gly	199	48		Asp	164	1	ligand MnA

Table S5. Result of Discern for PqqC from *K. pneumoniae* and TenA from *B. subtilis* sequences. Homologous residues in PqqC and TenA are based on the multiple sequence alignment and are plotted in the structure shown in **Figure 5**.

PqqC KP				TenA BS		
Residue	Position	Rank from Discern	Comments	Residue	Position	Rank from Discern
His	27	1		His	21	11
His	24	2		Ser	18	27
Lys	214	3		Ser	201	14
His	84	4	proton donor	Thr	78	44
Glu	147	5		Tyr	136	21
Arg	50	6		Asp	44	1
Arg	179	7	O_2 core	Gly	164	180
Tyr	29	8		Phe	23	18
Arg	80	9		His	74	34
His	154	10	O_2 core	Gly	143	77
Tyr	53	11		Tyr	47	5
His	30	12		Val	24	68
Tyr	128	13		His	115	12
Gln	54	14		Leu	48	28
Arg	157	15		Leu	146	98
Asp	223	16		Gly	212	109
Tyr	23	17		Trp	14	160
Trp	46	20		Tyr	40	7
Tyr	175	26	O_2 core	Trp	167	169
Pro	151	53		Cys	135	3
Gln	155	117		Tyr	139	4
Ser	220	126		Glu	205	2
Val	125	46		Tyr	112	6
Met	221	47		Phe	208	9
Ser	178	168		Tyr	163	8

Table S6. Result of Discern for PqqD from *K. pneumoniae.*

Residue	Position	Rank from Discern
Glu	30	1
Ala	80	2
Lys	34	3
Arg	11	4
Arg	14	5
Tyr	13	6
Gly	31	7
Tyr	28	8
Trp	85	9
Asn	36	10

Table S7. Result of Discern for PqqE from *K. pneumoniae* and NirJ from *Sulfurovum sp.* sequences. Homologous residues in PqqE and NirJ are based on the multiple sequence alignment and are plotted in the structure shown in **Figure 7**.

PqqE				NirJ			
Residue	Position	Rank from INTREPID	Comments	Residue	Position	Rank from INTREPID	Comments
Cys	26	1	N-terminal Fe-S cluster ligand	Cys	37	2	N-terminal Fe-S
Cys	22	2	N-terminal Fe-S cluster ligand	Cys	33	4	N-terminal Fe-S
Cys	29	3	N-terminal Fe-S cluster ligand	Cys	40	5	N-terminal Fe-S
Gly	67	7	SAM stabilizing residues	Gly	77	11	SAM stabilizing
Gly	260	9		Gly	28	14	
Gly	66	10	SAM stabilizing residues	Gly	76	13	SAM stabilizing
Glu	68	11	SAM stabilizing residues	Glu	78	18	SAM stabilizing
Tyr	28	12		His	39	9	
Cys	309	20	C-terminal Fe-S cluster ligand	Cys	33	3	C-terminal Fe-S
Cys	306	21	C-terminal Fe-S cluster ligand	Cys	33	1	C-terminal Fe-S
Cys	337	24	C-terminal Fe-S cluster ligand	Cys	36	6	C-terminal Fe-S
Cys	319	25					

Figure S1. Maximum-likelihood trees of PqqB and its homologues. Homologues of each Pqq protein seed sequence were retrieved using FlowerPower (*1*) (See **Methods**). The datasets were filtered to reduce redundancy, so that only ~100 sequences are shown in this figure. Numbers at each node are bootstrap values (only those >50 are shown). PqqB seed sequences were labeled in blue. From the PhnP subfamily, we selected six sequences annotated as PhnP and with the highest identity to *Klebsiella* PqqB. These selected PhnP are labeled in purple. The upper box indicates the orthologues of PhnP (P11692) and the lower one indicates PqqB (B5XX61) orthologues. Trees were rooted using PqqB orthologues as the outgroup. See **Figure S6** for their alignments.

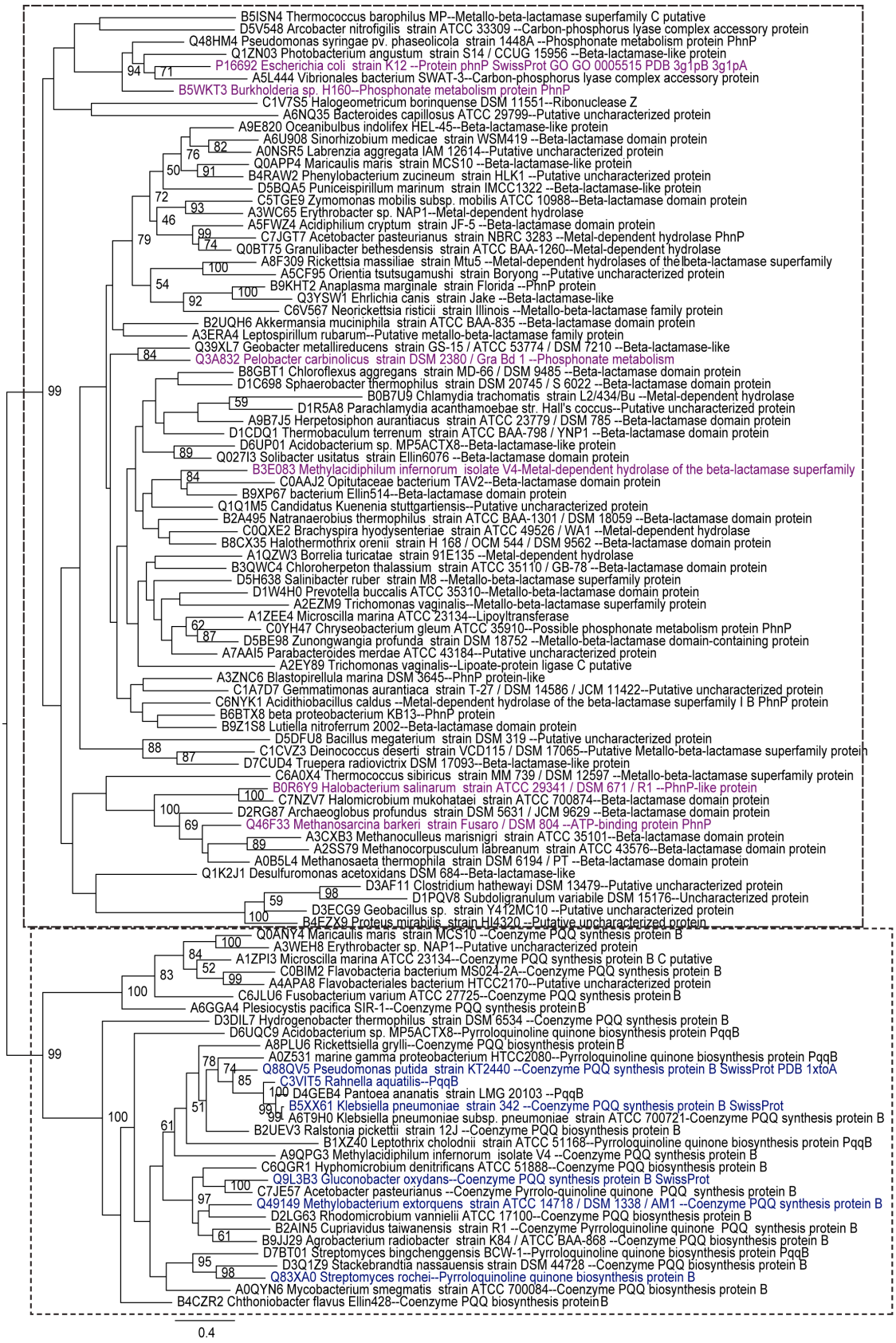

B5ISN4 Thermococcus barophilus MP--Metallo-beta-lactamase superfamily C putative
D5V548 Arcobacter nitrofigilis strain ATCC 33309 --Carbon-phosphorus lyase complex accessory protein
Q48HM4 Pseudomonas syringae pv. phaseolicola strain 1448A --Phosphonate metabolism protein PhnP
Q1ZN03 Photobacterium angustum strain S14 / CCUG 15956 --Beta-lactamase-like protein
P16692 Escherichia coli strain K12 --Protein phnP SwissProt GO GO 0005515 PDB 3g1pB 3g1pA
A5L444 Vibrionales bacterium SWAT-3--Carbon-phosphorus lyase complex accessory protein
B5WKT3 Burkholderia sp. H160--Phosphonate metabolism protein PhnP
C1V7S5 Halogeometricum borinquense DSM 11551--Ribonuclease Z
A6NQ35 Bacteroides capillosus ATCC 29799--Putative uncharacterized protein
A9E820 Oceanibulbus indolifex HEL-45--Beta-lactamase-like protein
A6U908 Sinorhizobium medicae strain WSM419 --Beta-lactamase domain protein
A0NSR5 Labrenzia aggregata IAM 12614--Putative uncharacterized protein
Q0APP4 Maricaulis maris strain MCS10 --Beta-lactamase-like protein
B4RAW2 Phenylobacterium zucineum strain HLK1 --Putative uncharacterized protein
D5BQA5 Puniceispirillum marinum strain IMCC1322 --Beta-lactamase-like protein
C5TGE9 Zymomonas mobilis subsp. mobilis ATCC 10988--Beta-lactamase domain protein
A3WC65 Erythrobacter sp. NAP1--Metal-dependent hydrolase
A5FWZ4 Acidiphilium cryptum strain JF-5 --Beta-lactamase domain protein
C7JGT7 Acetobacter pasteurianus strain NBRC 3283 --Metal-dependent hydrolase PhnP
Q0BT75 Granulibacter bethesdensis strain ATCC BAA-1260--Metal-dependent hydrolase
A8F309 Rickettsia massiliae strain Mtu5 --Metal-dependent hydrolases of the beta-lactamase superfamily
A5CF95 Orientia tsutsugamushi strain Boryong --Putative uncharacterized protein
B9KHT2 Anaplasma marginale strain Florida --PhnP protein
Q3YSW1 Ehrlichia canis strain Jake --Beta-lactamase-like
C6V567 Neorickettsia risticii strain Illinois --Metallo-beta-lactamase family protein
B2UQH6 Akkermansia muciniphila strain ATCC BAA-835 --Beta-lactamase domain protein
A3ERA4 Leptospirillum rubarum--Putative metallo-beta-lactamase family protein
Q39XL7 Geobacter metallireducens strain GS-15 / ATCC 53774 / DSM 7210 --Beta-lactamase-like
Q3A832 Pelobacter carbinolicus strain DSM 2380 / Gra Bd 1 --Phosphonate metabolism
B8GBT1 Chloroflexus aggregans strain MD-66 / DSM 9485 --Beta-lactamase domain protein
D1C698 Sphaerobacter thermophilus strain DSM 20745 / S 6022 --Beta-lactamase domain protein
B0B7U9 Chlamydia trachomatis strain L2/434/Bu --Metal-dependent hydrolase
D1R5A8 Parachlamydia acanthamoebae str. Hall's coccus--Putative uncharacterized protein
A9B7J5 Herpetosiphon aurantiacus strain ATCC 23779 / DSM 785 --Beta-lactamase domain protein
D1CDQ1 Thermobaculum terrenum strain ATCC BAA-798 / YNP1 --Beta-lactamase domain protein
D6UP01 Acidobacterium sp. MP5ACTX8--Beta-lactamase-like protein
Q027I3 Solibacter usitatus strain Ellin6076 --Beta-lactamase domain protein
B3E083 Methylacidiphilum infernorum isolate V4-Metal-dependent hydrolase of the beta-lactamase superfamily
C0AAJ2 Opitutaceae bacterium TAV2--Beta-lactamase domain protein
B9XP67 bacterium Ellin514--Beta-lactamase domain protein
Q1Q1M5 Candidatus Kuenenia stuttgartiensis--Putative uncharacterized protein
B2A495 Natranaerobius thermophilus strain ATCC BAA-1301 / DSM 18059 --Beta-lactamase domain protein
C0QXE2 Brachyspira hyodysenteriae strain ATCC 49526 / WA1 --Metal-dependent hydrolase
B8CX35 Halothermothrix orenii strain H 168 / OCM 544 / DSM 9562 --Beta-lactamase domain protein
A1QZW3 Borrelia turicatae strain 91E135 --Metal-dependent hydrolase
B3QWC4 Chloroherpeton thalassium strain ATCC 35110 / GB-78 --Beta-lactamase domain protein
D5H638 Salinibacter ruber strain M8 --Metallo-beta-lactamase superfamily protein
D1W4H0 Prevotella buccalis ATCC 35310--Metallo-beta-lactamase domain protein
A2EZM9 Trichomonas vaginalis--Metallo-beta-lactamase superfamily protein
A1ZEE4 Microscilla marina ATCC 23134--Lipoyltransferase
C0YH47 Chryseobacterium gleum ATCC 35910--Possible phosphonate metabolism protein PhnP
D5BE98 Zunongwangia profunda strain DSM 18752 --Metallo-beta-lactamase domain-containing protein
A7AAI5 Parabacteroides merdae ATCC 43184--Putative uncharacterized protein
A2EY89 Trichomonas vaginalis--Lipoate-protein ligase C putative
A3ZNC6 Blastopirellula marina DSM 3645--PhnP protein-like
C1A7D7 Gemmatimonas aurantiaca strain T-27 / DSM 14586 / JCM 11422--Putative uncharacterized protein
C6NYK1 Acidithiobacillus caldus --Metal-dependent hydrolase of the beta-lactamase superfamily I B PhnP protein
B6BTX8 beta proteobacterium KB13--PhnP protein
B9Z1S8 Lutiella nitroferrum 2002--Beta-lactamase domain protein
D5DFU8 Bacillus megaterium strain DSM 319 --Putative uncharacterized protein
C1CVZ3 Deinococcus deserti strain VCD115 / DSM 17065--Putative Metallo-beta-lactamase superfamily protein
D7CUD4 Truepera radiovictrix DSM 17093--Beta-lactamase-like protein
C6A0X4 Thermococcus sibiricus strain MM 739 / DSM 12597 --Metallo-beta-lactamase superfamily protein
B0R6Y9 Halobacterium salinarum strain ATCC 29341 / DSM 671 / R1 --PhnP-like protein
C7NZV7 Halomicrobium mukohataei strain ATCC 700874--Beta-lactamase domain protein
D2RG87 Archaeoglobus profundus strain DSM 5631 / JCM 9629 --Beta-lactamase domain protein
Q46F33 Methanosarcina barkeri strain Fusaro / DSM 804 --ATP-binding protein PhnP
A3CXB3 Methanoculleus marisnigri strain ATCC 35101--Beta-lactamase domain protein
A2SS79 Methanocorpusculum labreanum strain ATCC 43576--Beta-lactamase domain protein
A0B5L4 Methanosaeta thermophila strain DSM 6194 / PT --Beta-lactamase domain protein
Q1K2J1 Desulfuromonas acetoxidans DSM 684--Beta-lactamase-like
D3AF11 Clostridium hathewayi DSM 13479--Putative uncharacterized protein
D1PQV8 Subdoligranulum variabile DSM 15176--Uncharacterized protein
D3ECG9 Geobacillus sp. strain Y412MC10 --Putative uncharacterized protein
B4EZX9 Proteus mirabilis strain HI4320 --Putative uncharacterized protein
Q0ANY4 Maricaulis maris strain MCS10 --Coenzyme PQQ synthesis protein B
A3WEH8 Erythrobacter sp. NAP1--Putative uncharacterized protein
A1ZPI3 Microscilla marina ATCC 23134--Coenzyme PQQ synthesis protein B C putative
C0BIM2 Flavobacteria bacterium MS024-2A--Coenzyme PQQ synthesis protein B
A4APA8 Flavobacteriales bacterium HTCC2170--Putative uncharacterized protein
C6JLU6 Fusobacterium varium ATCC 27725--Coenzyme PQQ synthesis protein B
A6GGA4 Plesiocystis pacifica SIR-1--Coenzyme PQQ synthesis protein B
D3DIL7 Hydrogenobacter thermophilus strain DSM 6534 --Coenzyme PQQ synthesis protein B
D6UQC9 Acidobacterium sp. MP5ACTX8--Pyrroloquinoline quinone biosynthesis protein PqqB
A8PLU6 Rickettsiella grylli--Coenzyme PQQ biosynthesis protein B
A0Z531 marine gamma proteobacterium HTCC2080--Pyrroloquinoline quinone biosynthesis protein PqqB
Q88QV5 Pseudomonas putida strain KT2440 --Coenzyme PQQ synthesis protein B SwissProt PDB 1xtoA
C3VIT5 Rahnella aquatilis--PqqB
D4GEB4 Pantoea ananatis strain LMG 20103 --PqqB
B5XX61 Klebsiella pneumoniae strain 342 --Coenzyme PQQ synthesis protein B SwissProt
A6T9H0 Klebsiella pneumoniae subsp. pneumoniae strain ATCC 700721-Coenzyme PQQ synthesis protein B
B2UEV3 Ralstonia pickettii strain 12J --Coenzyme PQQ biosynthesis protein B
B1XZ40 Leptothrix cholodnii strain ATCC 51168--Pyrroloquinoline quinone biosynthesis protein PqqB
A9QPG3 Methylacidiphilum infernorum isolate V4 --Coenzyme PQQ synthesis protein B
C6QGR1 Hyphomicrobium denitrificans ATCC 51888--Coenzyme PQQ biosynthesis protein B
Q9L3B3 Gluconobacter oxydans--Coenzyme PQQ synthesis protein B SwissProt
C7JE57 Acetobacter pasteurianus --Coenzyme Pyrrolo-quinoline quinone PQQ synthesis protein B
Q49149 Methylobacterium extorquens strain ATCC 14718 / DSM 1338 / AM1 --Coenzyme PQQ synthesis protein B
D2LG63 Rhodomicrobium vannielii ATCC 17100--Coenzyme PQQ biosynthesis protein B
B2AIN5 Cupriavidus taiwanensis strain R1 --Coenzyme Pyrroloquinoline quinone PQQ synthesis protein B
B9JJ29 Agrobacterium radiobacter strain K84 / ATCC BAA-868 --Coenzyme PQQ biosynthesis protein B
D7BT01 Streptomyces bingchenggensis BCW-1--Pyrroloquinoline quinone biosynthesis protein PqqB
D3Q1Z9 Stackebrandtia nassauensis strain DSM 44728 --Coenzyme PQQ biosynthesis protein B
Q83XA0 Streptomyces rochei--Pyrroloquinoline quinone biosynthesis protein B
A0QYN6 Mycobacterium smegmatis strain ATCC 700084--Coenzyme PQQ biosynthesis protein B
B4CZR2 Chthoniobacter flavus Ellin428--Coenzyme PQQ biosynthesis protein B
0.4

Figure S2. Maximum-likelihood trees of PqqC and its homologues. Homologues of each Pqq protein seed sequence were retrieved using FlowerPower (*1*) (See **Methods**). The datasets were filtered to reduce redundancy, so that only ~100 sequences are shown in this figure. Numbers at each node are bootstrap values (only those > 50 are shown). PqqC seed sequences were labeled in blue. From the TenA subfamily, we picked representative a TenA sequence with experimental evidence and solved 3-D structure, in this case the sequence P25052. And we chose another four sequences with highest identities to it and *Klebsiella* PqqC as representative TenA proteins, and labeled the sequences in purple. Numbers at each node are bootstrap values. The upper box indicates the orthologues of TenA (P25052) and the lower one indicates PqqC (B5XX60) orthologues. See **Figure S7** for their alignments.

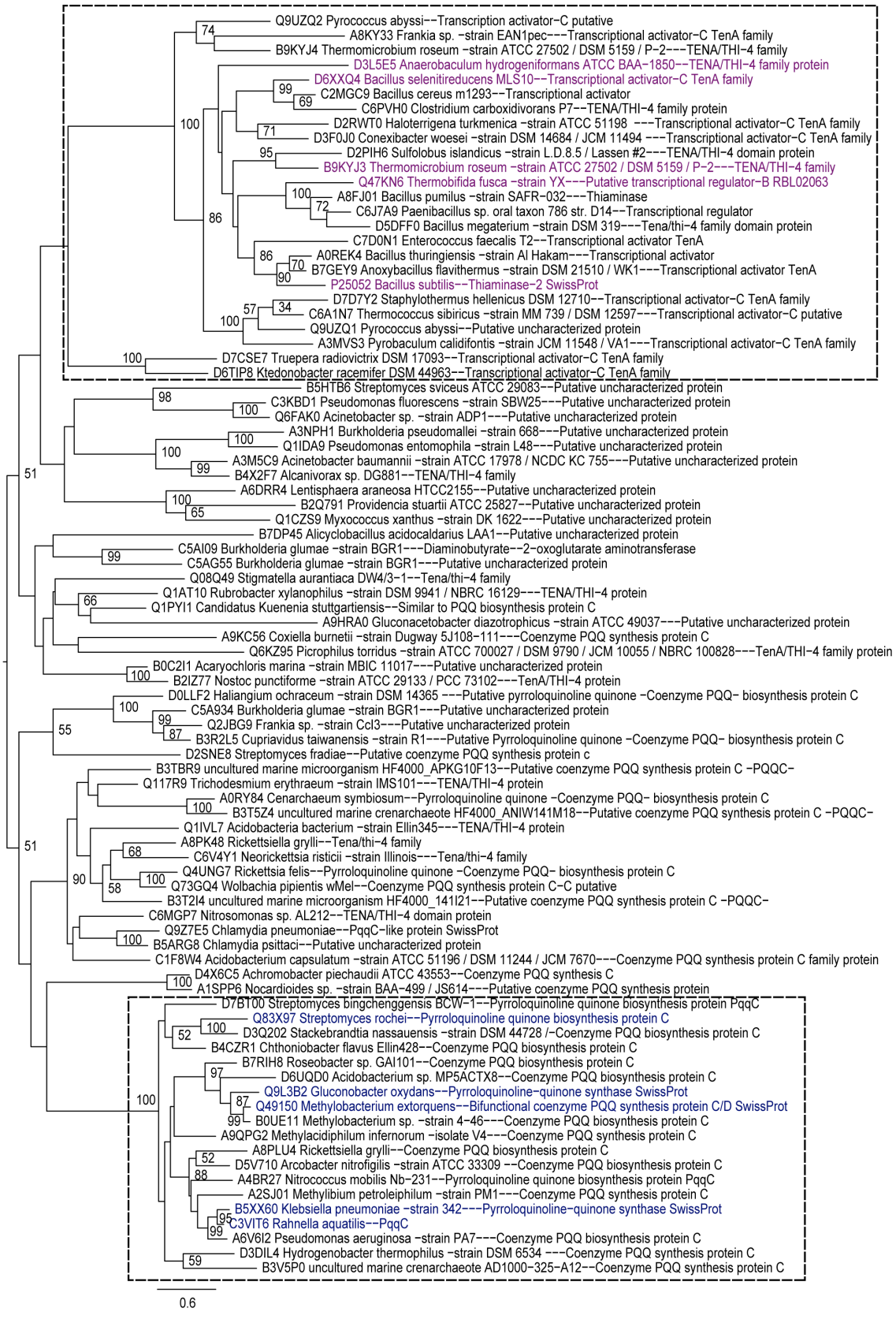

Q9UZQ2 Pyrococcus abyssi--Transcription activator-C putative
A8KY33 Frankia sp. -strain EAN1pec---Transcriptional activator-C TenA family
B9KYJ4 Thermomicrobium roseum -strain ATCC 27502 / DSM 5159 / P-2---TENA/THI-4 family
D3L5E5 Anaerobaculum hydrogeniformans ATCC BAA-1850--TENA/THI-4 family protein
D6XXQ4 Bacillus selenitireducens MLS10--Transcriptional activator-C TenA family
C2MGC9 Bacillus cereus m1293--Transcriptional activator
C6PVH0 Clostridium carboxidivorans P7--TENA/THI-4 family protein
D2RWT0 Haloterrigena turkmenica -strain ATCC 51198 ---Transcriptional activator-C TenA family
D3F0J0 Conexibacter woesei -strain DSM 14684 / JCM 11494 ---Transcriptional activator-C TenA family
D2PIH6 Sulfolobus islandicus -strain L.D.8.5 / Lassen #2---TENA/THI-4 domain protein
B9KYJ3 Thermomicrobium roseum -strain ATCC 27502 / DSM 5159 / P-2---TENA/THI-4 family
Q47KN6 Thermobifida fusca -strain YX---Putative transcriptional regulator-B RBL02063
A8FJ01 Bacillus pumilus -strain SAFR-032---Thiaminase
C6J7A9 Paenibacillus sp. oral taxon 786 str. D14--Transcriptional regulator
D5DFF0 Bacillus megaterium -strain DSM 319---Tena/thi-4 family domain protein
C7D0N1 Enterococcus faecalis T2--Transcriptional activator TenA
A0REK4 Bacillus thuringiensis -strain Al Hakam---Transcriptional activator
B7GEY9 Anoxybacillus flavithermus -strain DSM 21510 / WK1---Transcriptional activator TenA
P25052 Bacillus subtilis--Thiaminase-2 SwissProt
D7D7Y2 Staphylothermus hellenicus DSM 12710--Transcriptional activator-C TenA family
C6A1N7 Thermococcus sibiricus -strain MM 739 / DSM 12597---Transcriptional activator-C putative
Q9UZQ1 Pyrococcus abyssi--Putative uncharacterized protein
A3MVS3 Pyrobaculum calidifontis -strain JCM 11548 / VA1---Transcriptional activator-C TenA family
D7CSE7 Truepera radiovictrix DSM 17093--Transcriptional activator-C TenA family
D6TIP8 Ktedonobacter racemifer DSM 44963--Transcriptional activator-C TenA family
B5HTB6 Streptomyces sviceus ATCC 29083--Putative uncharacterized protein
C3KBD1 Pseudomonas fluorescens -strain SBW25---Putative uncharacterized protein
Q6FAK0 Acinetobacter sp. -strain ADP1---Putative uncharacterized protein
A3NPH1 Burkholderia pseudomallei -strain 668---Putative uncharacterized protein
Q1IDA9 Pseudomonas entomophila -strain L48---Putative uncharacterized protein
A3M5C9 Acinetobacter baumannii -strain ATCC 17978 / NCDC KC 755---Putative uncharacterized protein
B4X2F7 Alcanivorax sp. DG881--TENA/THI-4 family
A6DRR4 Lentisphaera araneosa HTCC2155--Putative uncharacterized protein
B2Q791 Providencia stuartii ATCC 25827--Putative uncharacterized protein
Q1CZS9 Myxococcus xanthus -strain DK 1622---Putative uncharacterized protein
B7DP45 Alicyclobacillus acidocaldarius LAA1--Putative uncharacterized protein
C5AI09 Burkholderia glumae -strain BGR1---Diaminobutyrate--2-oxoglutarate aminotransferase
C5AG55 Burkholderia glumae -strain BGR1---Putative uncharacterized protein
Q08Q49 Stigmatella aurantiaca DW4/3-1--Tena/thi-4 family
Q1AT10 Rubrobacter xylanophilus -strain DSM 9941 / NBRC 16129---TENA/THI-4 protein
Q1PYI1 Candidatus Kuenenia stuttgartiensis--Similar to PQQ biosynthesis protein C
A9HRA0 Gluconacetobacter diazotrophicus -strain ATCC 49037---Putative uncharacterized protein
A9KC56 Coxiella burnetii -strain Dugway 5J108-111---Coenzyme PQQ synthesis protein C
Q6KZ95 Picrophilus torridus -strain ATCC 700027 / DSM 9790 / JCM 10055 / NBRC 100828---TenA/THI-4 family protein
B0C2I1 Acaryochloris marina -strain MBIC 11017---Putative uncharacterized protein
B2IZ77 Nostoc punctiforme -strain ATCC 29133 / PCC 73102---TenA/THI-4 protein
D0LLF2 Haliangium ochraceum -strain DSM 14365 ---Putative pyrroloquinoline quinone -Coenzyme PQQ- biosynthesis protein C
C5A934 Burkholderia glumae -strain BGR1---Putative uncharacterized protein
Q2JBG9 Frankia sp. -strain CcI3---Putative uncharacterized protein
B3R2L5 Cupriavidus taiwanensis -strain R1---Putative Pyrroloquinoline quinone -Coenzyme PQQ- biosynthesis protein C
D2SNE8 Streptomyces fradiae--Putative coenzyme PQQ synthesis protein c
B3TBR9 uncultured marine microorganism HF4000_APKG10F13--Putative coenzyme PQQ synthesis protein C -PQQC-
Q117R9 Trichodesmium erythraeum -strain IMS101---TENA/THI-4 protein
A0RY84 Cenarchaeum symbiosum--Pyrroloquinoline quinone -Coenzyme PQQ- biosynthesis protein C
B3T5Z4 uncultured marine crenarchaeote HF4000_ANIW141M18--Putative coenzyme PQQ synthesis protein C -PQQC-
Q1IVL7 Acidobacteria bacterium -strain Ellin345---TENA/THI-4 protein
A8PK48 Rickettsiella grylli--Tena/thi-4 family
C6V4Y1 Neorickettsia risticii -strain Illinois---Tena/thi-4 family
Q4UNG7 Rickettsia felis--Pyrroloquinoline quinone -Coenzyme PQQ- biosynthesis protein C
Q73GQ4 Wolbachia pipientis wMel--Coenzyme PQQ synthesis protein C-C putative
B3T2I4 uncultured marine microorganism HF4000_141I21--Putative coenzyme PQQ synthesis protein C -PQQC-
C6MGP7 Nitrosomonas sp. AL212--TENA/THI-4 domain protein
Q9Z7E5 Chlamydia pneumoniae--PqqC-like protein SwissProt
B5ARG8 Chlamydia psittaci--Putative uncharacterized protein
C1F8W4 Acidobacterium capsulatum -strain ATCC 51196 / DSM 11244 / JCM 7670---Coenzyme PQQ synthesis protein C family protein
D4X6C5 Achromobacter piechaudii ATCC 43553--Coenzyme PQQ synthesis C
A1SPP6 Nocardioides sp. -strain BAA-499 / JS614---Putative coenzyme PQQ synthesis protein
D7B100 Streptomyces bingchenggensis BCW-1--Pyrroloquinoline quinone biosynthesis protein PqqC
Q83X97 Streptomyces rochei--Pyrroloquinoline quinone biosynthesis protein C
D3Q202 Stackebrandtia nassauensis -strain DSM 44728 /-Coenzyme PQQ biosynthesis protein C
B4CZR1 Chthoniobacter flavus Ellin428--Coenzyme PQQ biosynthesis protein C
B7RIH8 Roseobacter sp. GAI101--Coenzyme PQQ biosynthesis protein C
D6UQD0 Acidobacterium sp. MP5ACTX8--Coenzyme PQQ biosynthesis protein C
Q9L3B2 Gluconobacter oxydans--Pyrroloquinoline-quinone synthase SwissProt
Q49150 Methylobacterium extorquens--Bifunctional coenzyme PQQ synthesis protein C/D SwissProt
B0UE11 Methylobacterium sp. -strain 4-46---Coenzyme PQQ biosynthesis protein C
A9QPG2 Methylacidiphilum infernorum -isolate V4---Coenzyme PQQ synthesis protein C
A8PLU4 Rickettsiella grylli--Coenzyme PQQ biosynthesis protein C
D5V710 Arcobacter nitrofigilis -strain ATCC 33309 --Coenzyme PQQ biosynthesis protein C
A4BR27 Nitrococcus mobilis Nb-231--Pyrroloquinoline quinone biosynthesis protein PqqC
A2SJ01 Methylibium petroleiphilum -strain PM1---Coenzyme PQQ synthesis protein C
B5XX60 Klebsiella pneumoniae -strain 342---Pyrroloquinoline-quinone synthase SwissProt
C3VIT6 Rahnella aquatilis--PqqC
A6V6I2 Pseudomonas aeruginosa -strain PA7---Coenzyme PQQ biosynthesis protein C
D3DIL4 Hydrogenobacter thermophilus -strain DSM 6534 ---Coenzyme PQQ synthesis protein C
B3V5P0 uncultured marine crenarchaeote AD1000-325-A12--Coenzyme PQQ synthesis protein C
0.6

Figure S3. Maximum-likelihood trees of PqqD and its homologues. Homologues of each Pqq protein seed sequence were retrieved using FlowerPower (*1*) (See **Methods**). The datasets were filtered to reduce redundancy, so that only ~100 sequences are shown in this figure. Numbers at each node are bootstrap values (only those >50 are shown). Orthologues of PqqD are boxed in the figure. The five PqqD seed sequences were labeled in blue. See **Figure 7B** for their alignments. Bootstrap values are low for most nodes due to the short length of PqqD proteins (~90 amino acids).

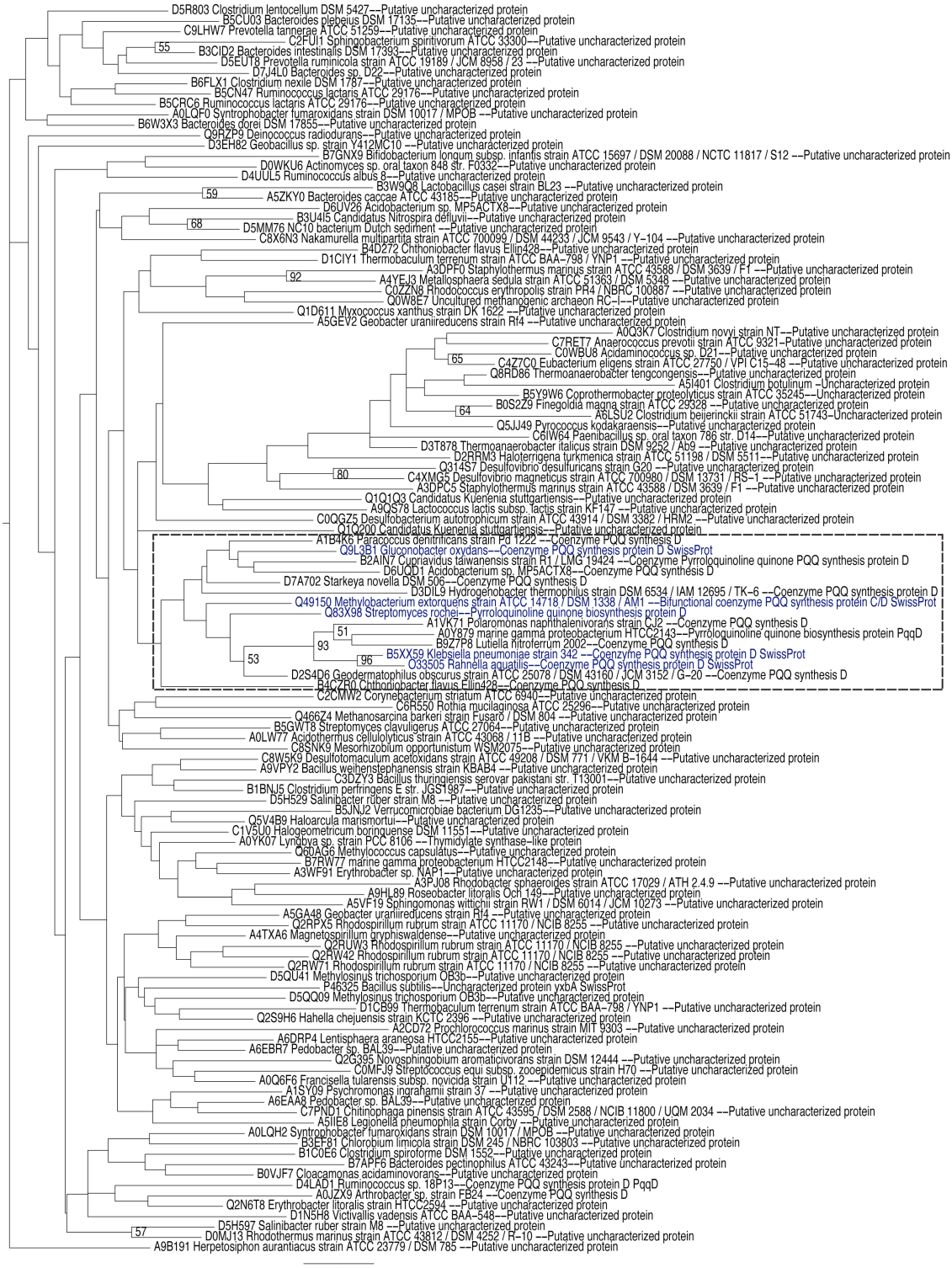

D5R803 Clostridium lentocellum DSM 5427--Putative uncharacterized protein
B5CU03 Bacteroides plebeius DSM 17135--Putative uncharacterized protein
C9LHW7 Prevotella tannerae ATCC 51259--Putative uncharacterized protein
C2FUI1 Sphingobacterium spiritivorum ATCC 33300--Putative uncharacterized protein
55
B3CID2 Bacteroides intestinalis DSM 17393--Putative uncharacterized protein
D5EUT8 Prevotella ruminicola strain ATCC 19189 / JCM 8958 / 23 --Putative uncharacterized protein
D7J4L0 Bacteroides sp. D22--Putative uncharacterized protein
B6FLX1 Clostridium nexile DSM 1787--Putative uncharacterized protein
B5CN47 Ruminococcus lactaris ATCC 29176--Putative uncharacterized protein
B5CRC6 Ruminococcus lactaris ATCC 29176--Putative uncharacterized protein
A0LQF0 Syntrophobacter fumaroxidans strain DSM 10017 / MPOB --Putative uncharacterized protein
B6W3X3 Bacteroides dorei DSM 17855--Putative uncharacterized protein
Q9RZP9 Deinococcus radiodurans--Putative uncharacterized protein
D3EH82 Geobacillus sp. strain Y412MC10 --Putative uncharacterized protein
B7GNX9 Bifidobacterium longum subsp. infantis strain ATCC 15697 / DSM 20088 / NCTC 11817 / S12 --Putative uncharacterized protein
D0WKU6 Actinomyces sp. oral taxon 848 str. F0332--Putative uncharacterized protein
D4UUL5 Ruminococcus albus 8--Putative uncharacterized protein
B3W9Q8 Lactobacillus casei strain BL23 --Putative uncharacterized protein
59
A5ZKY0 Bacteroides caccae ATCC 43185--Putative uncharacterized protein
D6UV26 Acidobacterium sp. MP5ACTX9--Putative uncharacterized protein
B3U4I5 Candidatus Nitrospira defluvii--Putative uncharacterized protein
68
D5MM76 NC10 bacterium Dutch sediment --Putative uncharacterized protein
C8X6N3 Nakamurella multipartita strain ATCC 700099 / DSM 44233 / JCM 9543 / Y-104 --Putative uncharacterized protein
B4D272 Chthoniobacter flavus Ellin428--Putative uncharacterized protein
D1CIY1 Thermobaculum terrenum strain ATCC BAA-798 / YNP1 --Putative uncharacterized protein
A3DPF0 Staphylothermus marinus strain ATCC 43588 / DSM 3639 / F1 --Putative uncharacterized protein
92
A4YEJ3 Metallosphaera sedula strain ATCC 51363 / DSM 5348 --Putative uncharacterized protein
C0ZZN6 Rhodococcus erythropolis strain PR4 / NBRC 100887 --Putative uncharacterized protein
Q0W8E7 Uncultured methanogenic archaeon RC-I--Putative uncharacterized protein
Q1D611 Myxococcus xanthus strain DK 1622 --Putative uncharacterized protein
A5GEV2 Geobacter uraniireducens strain Rf4 --Putative uncharacterized protein
A0Q3K7 Clostridium novyi strain NT--Putative uncharacterized protein
C7RET7 Anaerococcus prevotii strain ATCC 9321--Putative uncharacterized protein
C0WBU8 Acidaminococcus sp. D21--Putative uncharacterized protein
65
C4Z7C0 Eubacterium eligens strain ATCC 27750 / VPI C15-48 --Putative uncharacterized protein
Q8RD86 Thermoanaerobacter tengcongensis--Putative uncharacterized protein
A5I401 Clostridium botulinum --Uncharacterized protein
B5Y9W6 Coprothermobacter proteolyticus strain ATCC 35245--Uncharacterized protein
B0S2Z9 Finegoldia magna strain ATCC 29328 --Putative uncharacterized protein
64
A6LSU2 Clostridium beijerinckii strain ATCC 51743--Uncharacterized protein
Q5JJ49 Pyrococcus kodakaraensis--Putative uncharacterized protein
C6IW64 Paenibacillus sp. oral taxon 786 str. D14--Putative uncharacterized protein
D3T878 Thermoanaerobacter italicus strain DSM 9252 / Ab9 --Putative uncharacterized protein
D2RRM3 Haloterrigena turkmenica strain ATCC 51198 / DSM 5511--Putative uncharacterized protein
Q314S7 Desulfovibrio desulfuricans strain G20 --Putative uncharacterized protein
80
C4XMG5 Desulfovibrio magneticus strain ATCC 700980 / DSM 13731 / RS-1 --Putative uncharacterized protein
A3DPC5 Staphylothermus marinus strain ATCC 43588 / DSM 3639 / F1 --Putative uncharacterized protein
Q1Q1Q3 Candidatus Kuenenia stuttgartiensis--Putative uncharacterized protein
A9QS78 Lactococcus lactis subsp. lactis strain KF147 --Putative uncharacterized protein
C0QGZ5 Desulfobacterium autotrophicum strain ATCC 43914 / DSM 3382 / HRM2 --Putative uncharacterized protein
Q1Q200 Candidatus Kuenenia stuttgartiensis--Putative uncharacterized protein
A1B4K6 Paracoccus denitrificans strain Pd 1222 --Coenzyme PQQ synthesis D
Q9L3B1 Gluconobacter oxydans--Coenzyme PQQ synthesis protein D SwissProt
B2AIN7 Cupriavidus taiwanensis strain R1 / LMG 19424 --Coenzyme Pyrroloquinoline quinone PQQ synthesis protein D
D6UQD1 Acidobacterium sp. MP5ACTX8--Coenzyme PQQ synthesis D
D7A702 Starkeya novella DSM 506--Coenzyme PQQ synthesis D
D3DIL9 Hydrogenobacter thermophilus strain DSM 6534 / IAM 12695 / TK-6 --Coenzyme PQQ synthesis protein D
Q49150 Methylobacterium extorquens strain ATCC 14718 / DSM 1338 / AM1 --Bifunctional coenzyme PQQ synthesis protein C/D SwissProt
Q83X98 Streptomyces rochei--Pyrroloquinoline quinone biosynthesis protein D
A1VK71 Polaromonas naphthalenivorans strain CJ2 --Coenzyme PQQ synthesis D
51
A0Y879 marine gamma proteobacterium HTCC2143--Pyrroloquinoline quinone biosynthesis protein PqqD
93
B9Z7P8 Lutiella nitroferrum 2002--Coenzyme PQQ synthesis D
53
B5XX59 Klebsiella pneumoniae strain 342 --Coenzyme PQQ synthesis protein D SwissProt
96
O33505 Rahnella aquatilis--Coenzyme PQQ synthesis protein D SwissProt
D2S4D6 Geodermatophilus obscurus strain ATCC 25078 / DSM 43160 / JCM 3152 / G-20 --Coenzyme PQQ synthesis D
B4C7R0 Chthoniobacter flavus Ellin428--Coenzyme PQQ synthesis D
C2CMW2 Corynebacterium striatum ATCC 6940--Putative uncharacterized protein
C6R550 Rothia mucilaginosa ATCC 25296--Putative uncharacterized protein
Q466Z4 Methanosarcina barkeri strain Fusaro / DSM 804 --Putative uncharacterized protein
B5GWT8 Streptomyces clavuligerus ATCC 27064--Putative uncharacterized protein
A0LW77 Acidothermus cellulolyticus strain ATCC 43068 / 11B --Putative uncharacterized protein
C8SNK9 Mesorhizobium opportunistum WSM2075--Putative uncharacterized protein
C8W5K9 Desulfotomaculum acetoxidans strain ATCC 49208 / DSM 771 / VKM B-1644 --Putative uncharacterized protein
A9VPY2 Bacillus weihenstephanensis strain KBAB4 --Putative uncharacterized protein
C3DZY3 Bacillus thuringiensis serovar pakistani str. T13001--Putative uncharacterized protein
B1BNJ5 Clostridium perfringens E str. JGS1987--Putative uncharacterized protein
D5H529 Salinibacter ruber strain M8 --Putative uncharacterized protein
B5JNJ2 Verrucomicrobiae bacterium DG1235--Putative uncharacterized protein
Q5V4B9 Haloarcula marismortui--Putative uncharacterized protein
C1V5U0 Halogeometricum borinquense DSM 11551--Putative uncharacterized protein
A0YK07 Lyngbya sp. strain PCC 8106 --Thymidylate synthase-like protein
Q6DAG6 Methylococcus capsulatus--Putative uncharacterized protein
B7RW77 marine gamma proteobacterium HTCC2148--Putative uncharacterized protein
A3WF91 Erythrobacter sp. NAP1--Putative uncharacterized protein
A3PJ08 Rhodobacter sphaeroides strain ATCC 17029 / ATH 2.4.9 --Putative uncharacterized protein
A9HL89 Roseobacter litoralis Och 149--Putative uncharacterized protein
A5VF19 Sphingomonas wittichii strain RW1 / DSM 6014 / JCM 10273 --Putative uncharacterized protein
A5GA48 Geobacter uraniireducens strain Rf4 --Putative uncharacterized protein
Q2RPX5 Rhodospirillum rubrum strain ATCC 11170 / NCIB 8255 --Putative uncharacterized protein
A4TXA6 Magnetospirillum gryphiswaldense--Putative uncharacterized protein
Q2RUW3 Rhodospirillum rubrum strain ATCC 11170 / NCIB 8255 --Putative uncharacterized protein
Q2RW42 Rhodospirillum rubrum strain ATCC 11170 / NCIB 8255 --Putative uncharacterized protein
Q2RW71 Rhodospirillum rubrum strain ATCC 11170 / NCIB 8255 --Putative uncharacterized protein
D5QU41 Methylosinus trichosporium OB3b--Putative uncharacterized protein
P46325 Bacillus subtilis--Uncharacterized protein yxbA SwissProt
D5QQ09 Methylosinus trichosporium OB3b--Putative uncharacterized protein
D1CB99 Thermobaculum terrenum strain ATCC BAA-798 / YNP1 --Putative uncharacterized protein
Q2S9H6 Hahella chejuensis strain KCTC 2396 --Putative uncharacterized protein
A2CD72 Prochlorococcus marinus strain MIT 9303 --Putative uncharacterized protein
A6DRP4 Lentisphaera araneosa HTCC2155--Putative uncharacterized protein
A6EBR7 Pedobacter sp. BAL39--Putative uncharacterized protein
Q2G395 Novosphingobium aromaticivorans strain DSM 12444 --Putative uncharacterized protein
C0MFJ9 Streptococcus equi subsp. zooepidemicus strain H70 --Putative uncharacterized protein
A0Q6F6 Francisella tularensis subsp. novicida strain U112 --Putative uncharacterized protein
A1SY09 Psychromonas ingrahamii strain 37 --Putative uncharacterized protein
A6EAA8 Pedobacter sp. BAL39--Putative uncharacterized protein
C7PND1 Chitinophaga pinensis strain ATCC 43595 / DSM 2588 / NCIB 11800 / UQM 2034 --Putative uncharacterized protein
A5IIE8 Legionella pneumophila strain Corby --Putative uncharacterized protein
A0LQH2 Syntrophobacter fumaroxidans strain DSM 10017 / MPOB --Putative uncharacterized protein
B3EF81 Chlorobium limicola strain DSM 245 / NBRC 103803 --Putative uncharacterized protein
B1C0E6 Clostridium spiroforme DSM 1552--Putative uncharacterized protein
B7APF6 Bacteroides pectinophilus ATCC 43243--Putative uncharacterized protein
B0VJF7 Cloacamonas acidaminovorans--Putative uncharacterized protein
D4LAD1 Ruminococcus sp. 18P13--Coenzyme PQQ synthesis protein D PqqD
A0JZX9 Arthrobacter sp. strain FB24 --Coenzyme PQQ synthesis D
Q2N6T8 Erythrobacter litoralis strain HTCC2594 --Putative uncharacterized protein
D1N5H8 Victivallis vadensis ATCC BAA-548--Putative uncharacterized protein
D5H597 Salinibacter ruber strain M8 --Putative uncharacterized protein
57
D0MJ13 Rhodothermus marinus strain ATCC 43812 / DSM 4252 / R-10 --Putative uncharacterized protein
A9B191 Herpetosiphon aurantiacus strain ATCC 23779 / DSM 785 --Putative uncharacterized protein
06

Figure S4. Maximum-likelihood trees of PqqE and its homologues. Homologues of each Pqq protein seed sequence were retrieved using FlowerPower (*1*) (See **Methods)**. The datasets were filtered to reduce redundancy, so that only ~100 sequences are shown in this figure. Numbers at each node are bootstrap values (only those >50 are shown). PqqE seed sequences were labeled in blue. From the NirJ subfamily, we picked five NirJ sequences that have the highest identity of PqqE as representatives and labeled them in purple. The upper box indicates the orthologues of NirJ (A6Q6Z2) and the lower one indicates PqqE (B5XX58) orthologues. See **Figure S8** for their alignments.

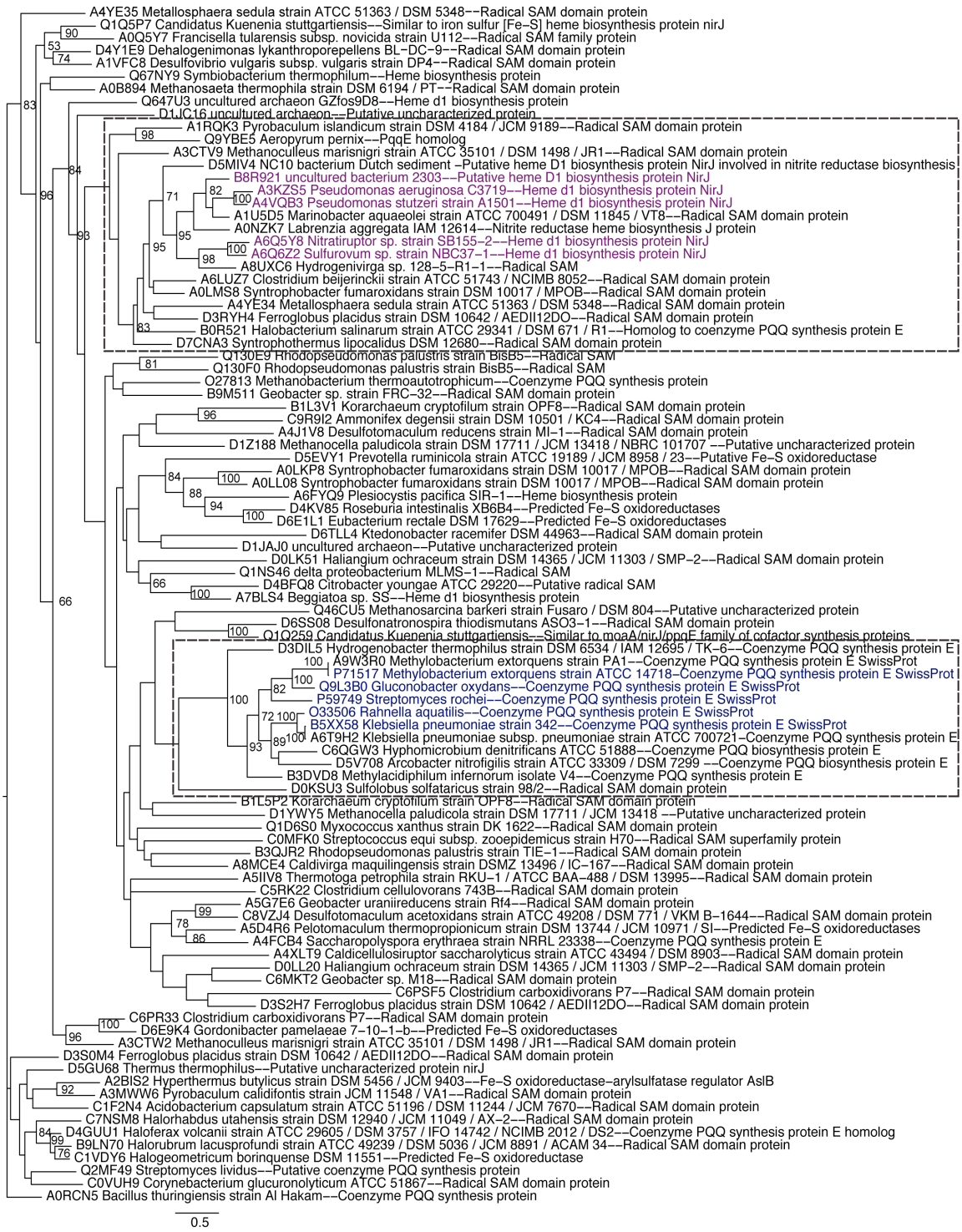

A4YE35 Metallosphaera sedula strain ATCC 51363 / DSM 5348--Radical SAM domain protein
Q1Q5P7 Candidatus Kuenenia stuttgartiensis--Similar to iron sulfur [Fe-S] heme biosynthesis protein nirJ
A0Q5Y7 Francisella tularensis subsp. novicida strain U112--Radical SAM family protein
D4Y1E9 Dehalogenimonas lykanthroporepellens BL-DC-9--Radical SAM domain protein
A1VFC8 Desulfovibrio vulgaris subsp. vulgaris strain DP4--Radical SAM domain protein
Q67NY9 Symbiobacterium thermophilum--Heme biosynthesis protein
A0B894 Methanosaeta thermophila strain DSM 6194 / PT--Radical SAM domain protein
Q647U3 uncultured archaeon GZfos9D8--Heme d1 biosynthesis protein
D1JC16 uncultured archaeon--Putative uncharacterized protein
A1RQK3 Pyrobaculum islandicum strain DSM 4184 / JCM 9189--Radical SAM domain protein
Q9YBE5 Aeropyrum pernix--PqqE homolog
A3CTV9 Methanoculleus marisnigri strain ATCC 35101 / DSM 1498 / JR1--Radical SAM domain protein
D5MIV4 NC10 bacterium Dutch sediment –Putative heme D1 biosynthesis protein NirJ involved in nitrite reductase biosynthesis
B8R921 uncultured bacterium 2303--Putative heme D1 biosynthesis protein NirJ
A3KZS5 Pseudomonas aeruginosa C3719--Heme d1 biosynthesis protein NirJ
A4VQB3 Pseudomonas stutzeri strain A1501--Heme d1 biosynthesis protein NirJ
A1U5D5 Marinobacter aquaeolei strain ATCC 700491 / DSM 11845 / VT8--Radical SAM domain protein
A0NZK7 Labrenzia aggregata IAM 12614--Nitrite reductase heme biosynthesis J protein
A6Q5Y8 Nitratiruptor sp. strain SB155-2--Heme d1 biosynthesis protein NirJ
A6Q6Z2 Sulfurovum sp. strain NBC37-1--Heme d1 biosynthesis protein NirJ
A8UXC6 Hydrogenivirga sp. 128-5-R1-1--Radical SAM
A6LUZ7 Clostridium beijerinckii strain ATCC 51743 / NCIMB 8052--Radical SAM domain protein
A0LMS8 Syntrophobacter fumaroxidans strain DSM 10017 / MPOB--Radical SAM domain protein
A4YE34 Metallosphaera sedula strain ATCC 51363 / DSM 5348--Radical SAM domain protein
D3RYH4 Ferroglobus placidus strain DSM 10642 / AEDII12DO--Radical SAM domain protein
B0R521 Halobacterium salinarum strain ATCC 29341 / DSM 671 / R1--Homolog to coenzyme PQQ synthesis protein E
D7CNA3 Syntrophothermus lipocalidus DSM 12680--Radical SAM domain protein
Q130E9 Rhodopseudomonas palustris strain BisB5--Radical SAM
Q130F0 Rhodopseudomonas palustris strain BisB5--Radical SAM
O27813 Methanobacterium thermoautotrophicum--Coenzyme PQQ synthesis protein
B9M511 Geobacter sp. strain FRC-32--Radical SAM domain protein
B1L3V1 Korarchaeum cryptofilum strain OPF8--Radical SAM domain protein
C9R9I2 Ammonifex degensii strain DSM 10501 / KC4--Radical SAM domain protein
A4J1V8 Desulfotomaculum reducens strain MI-1--Radical SAM domain protein
D1Z188 Methanocella paludicola strain DSM 17711 / JCM 13418 / NBRC 101707 --Putative uncharacterized protein
D5EVY1 Prevotella ruminicola strain ATCC 19189 / JCM 8958 / 23--Putative Fe-S oxidoreductase
A0LKP8 Syntrophobacter fumaroxidans strain DSM 10017 / MPOB--Radical SAM domain protein
A0LL08 Syntrophobacter fumaroxidans strain DSM 10017 / MPOB--Radical SAM domain protein
A6FYQ9 Plesiocystis pacifica SIR-1--Heme biosynthesis protein
D4KV85 Roseburia intestinalis XB6B4--Predicted Fe-S oxidoreductases
D6E1L1 Eubacterium rectale DSM 17629--Predicted Fe-S oxidoreductases
D6TLL4 Ktedonobacter racemifer DSM 44963--Radical SAM domain protein
D1JAJ0 uncultured archaeon--Putative uncharacterized protein
D0LK51 Haliangium ochraceum strain DSM 14365 / JCM 11303 / SMP-2--Radical SAM domain protein
Q1NS46 delta proteobacterium MLMS-1--Radical SAM
D4BFQ8 Citrobacter youngae ATCC 29220--Putative radical SAM
A7BLS4 Beggiatoa sp. SS--Heme d1 biosynthesis protein
Q46CU5 Methanosarcina barkeri strain Fusaro / DSM 804--Putative uncharacterized protein
D6SS08 Desulfonatronospira thiodismutans ASO3-1--Radical SAM domain protein
Q1Q259 Candidatus Kuenenia stuttgartiensis--Similar to moaA/nirJ/pqqE family of cofactor synthesis proteins
D3DIL5 Hydrogenobacter thermophilus strain DSM 6534 / IAM 12695 / TK-6--Coenzyme PQQ synthesis protein E
A9W3R0 Methylobacterium extorquens strain PA1--Coenzyme PQQ synthesis protein E SwissProt
P71517 Methylobacterium extorquens strain ATCC 14718-Coenzyme PQQ synthesis protein E SwissProt
Q9L3B0 Gluconobacter oxydans--Coenzyme PQQ synthesis protein E SwissProt
P59749 Streptomyces rochei--Coenzyme PQQ synthesis protein E SwissProt
O33506 Rahnella aquatilis--Coenzyme PQQ synthesis protein E SwissProt
B5XX58 Klebsiella pneumoniae strain 342--Coenzyme PQQ synthesis protein E SwissProt
A6T9H2 Klebsiella pneumoniae subsp. pneumoniae strain ATCC 700721-Coenzyme PQQ synthesis protein E
C6QGW3 Hyphomicrobium denitrificans ATCC 51888--Coenzyme PQQ biosynthesis protein E
D5V708 Arcobacter nitrofigilis strain ATCC 33309 / DSM 7299 --Coenzyme PQQ biosynthesis protein E
B3DVD8 Methylacidiphilum infernorum isolate V4--Coenzyme PQQ synthesis protein E
D0KSU3 Sulfolobus solfataricus strain 98/2--Radical SAM domain protein
B1L5P2 Korarchaeum cryptofilum strain OPF8--Radical SAM domain protein
D1YWY5 Methanocella paludicola strain DSM 17711 / JCM 13418 --Putative uncharacterized protein
Q1D6S0 Myxococcus xanthus strain DK 1622--Radical SAM domain protein
C0MFK0 Streptococcus equi subsp. zooepidemicus strain H70--Radical SAM superfamily protein
B3QJR2 Rhodopseudomonas palustris strain TIE-1--Radical SAM domain protein
A8MCE4 Caldivirga maquilingensis strain DSMZ 13496 / IC-167--Radical SAM domain protein
A5IIV8 Thermotoga petrophila strain RKU-1 / ATCC BAA-488 / DSM 13995--Radical SAM domain protein
C5RK22 Clostridium cellulovorans 743B--Radical SAM domain protein
A5G7E6 Geobacter uraniireducens strain Rf4--Radical SAM domain protein
C8VZJ4 Desulfotomaculum acetoxidans strain ATCC 49208 / DSM 771 / VKM B-1644--Radical SAM domain protein
A5D4R6 Pelotomaculum thermopropionicum strain DSM 13744 / JCM 10971 / SI--Predicted Fe-S oxidoreductases
A4FCB4 Saccharopolyspora erythraea strain NRRL 23338--Coenzyme PQQ synthesis protein E
A4XLT9 Caldicellulosiruptor saccharolyticus strain ATCC 43494 / DSM 8903--Radical SAM domain protein
D0LL20 Haliangium ochraceum strain DSM 14365 / JCM 11303 / SMP-2--Radical SAM domain protein
C6MKT2 Geobacter sp. M18--Radical SAM domain protein
C6PSF5 Clostridium carboxidivorans P7--Radical SAM domain protein
D3S2H7 Ferroglobus placidus strain DSM 10642 / AEDII12DO--Radical SAM domain protein
C6PR33 Clostridium carboxidivorans P7--Radical SAM domain protein
D6E9K4 Gordonibacter pamelaeae 7-10-1-b--Predicted Fe-S oxidoreductases
A3CTW2 Methanoculleus marisnigri strain ATCC 35101 / DSM 1498 / JR1--Radical SAM domain protein
D3S0M4 Ferroglobus placidus strain DSM 10642 / AEDII12DO--Radical SAM domain protein
D5GU68 Thermus thermophilus--Putative uncharacterized protein nirJ
A2BIS2 Hyperthermus butylicus strain DSM 5456 / JCM 9403--Fe-S oxidoreductase-arylsulfatase regulator AslB
A3MWW6 Pyrobaculum calidifontis strain JCM 11548 / VA1--Radical SAM domain protein
C1F2N4 Acidobacterium capsulatum strain ATCC 51196 / DSM 11244 / JCM 7670--Radical SAM domain protein
C7NSM8 Halorhabdus utahensis strain DSM 12940 / JCM 11049 / AX-2--Radical SAM domain protein
D4GUU1 Haloferax volcanii strain ATCC 29605 / DSM 3757 / IFO 14742 / NCIMB 2012 / DS2--Coenzyme PQQ synthesis protein E homolog
B9LN70 Halorubrum lacusprofundi strain ATCC 49239 / DSM 5036 / JCM 8891 / ACAM 34--Radical SAM domain protein
C1VDY6 Halogeometricum borinquense DSM 11551--Predicted Fe-S oxidoreductase
Q2MF49 Streptomyces lividus--Putative coenzyme PQQ synthesis protein
C0VUH9 Corynebacterium glucuronolyticum ATCC 51867--Radical SAM domain protein
A0RCN5 Bacillus thuringiensis strain Al Hakam--Coenzyme PQQ synthesis protein
0.5

Figure S5. Identified Pfam domains in PqqE, NirJ, and MoaA: three radical SAM proteins with two 4Fe-4S clusters. The C-terminal region is not conserved. Numbers indicate the beginning and end of identifiable Pfam domains in the primary sequence of each enzyme.

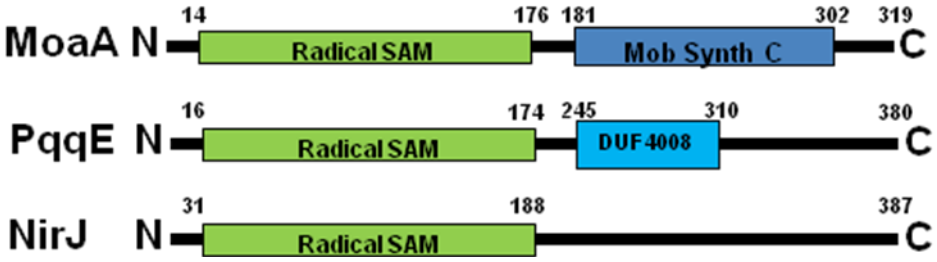


Figure S6. **MSA of selected PqqB orthologues and PhnP orthologues.** Six representative sequences of each orthologue groups were included in the MSA. Five-seed PqqB sequences and an additional PqqB with solved 3D structure were selected. From the PhnP subfamily, we picked a representative PhnP sequence with experimental evidence of its function and solved 3-D structure. We also selected other five sequences annotated as PhnP and with the highest identity to *Klebsiella* PqqB.

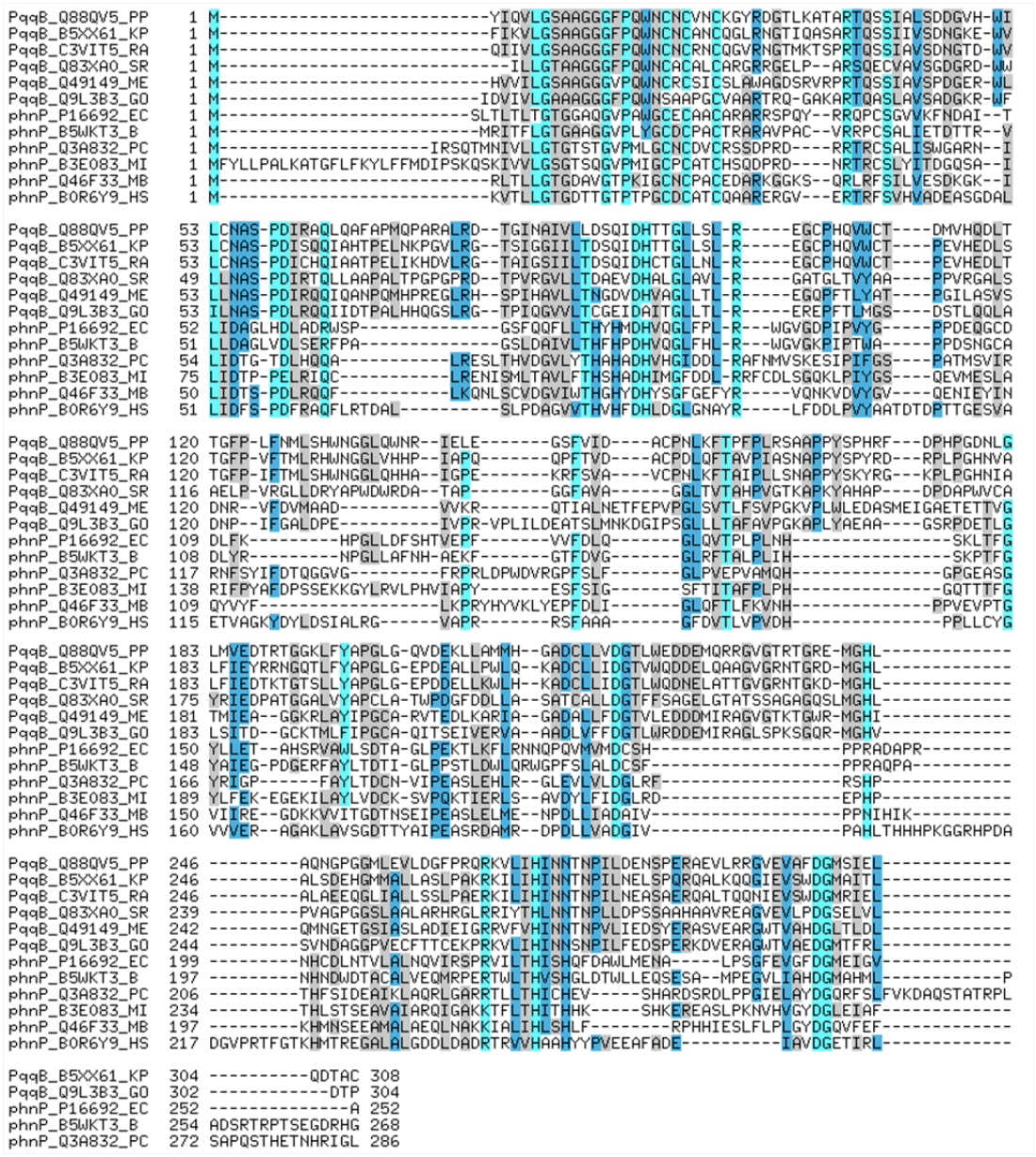

PqqB_Q88QV5_PP
PqqB_B5XX61_KP
PqqB_C3VIT5_RA
PqqB_Q83XA0_SR
PqqB_Q49149_ME
PqqB_Q9L3B3_GO
phnP_P16692_EC
phnP_B5WKT3_B
phnP_Q3A832_PC
phnP_B3E083_MI
phnP_Q46F33_MB
phnP_B0R6Y9_HS

Figure S7. **MSA of selected PqqC orthologues and TenA orthologues.** Five representative sequences of each orthologue groups shown in **Figure S4** were included in the MSA.

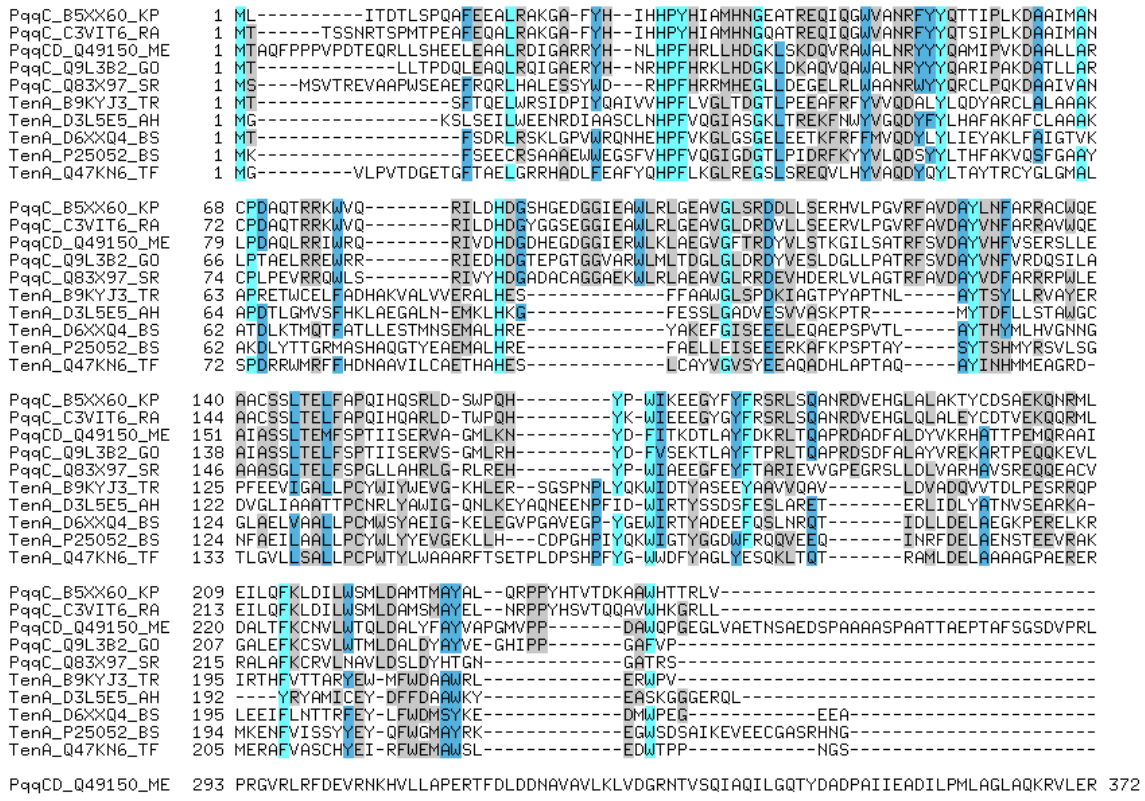
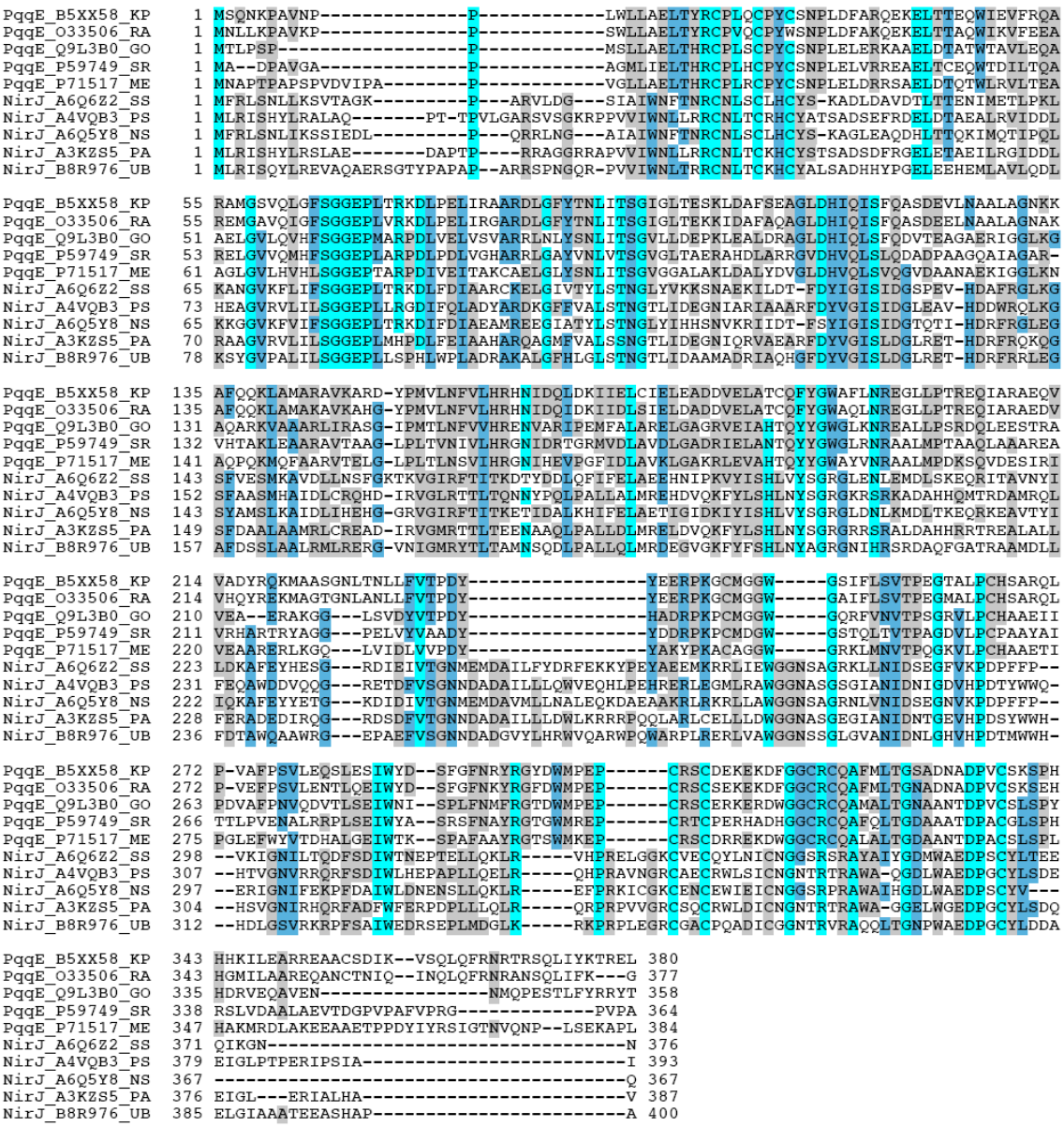


Figure S8. **MSA of selected PqqE orthologues and NirJ orthologues.** Five representative sequences of each orthologue groups shown in **Figure S8** were included in the MSA.



References

1. Krishnamurthy, N., Brown, D., and Sjolander, K. (2007) FlowerPower : clustering proteins into domain architecture classes for phylogenomic inférence of protein function. *BMC Evol. Biol. 7 Suppl 1,* S12.