Supplemental Material

The Trk potassium transporter is required for RsmB-mediated activation of virulence in the phytopathogen *Pectobacterium wasabiae*

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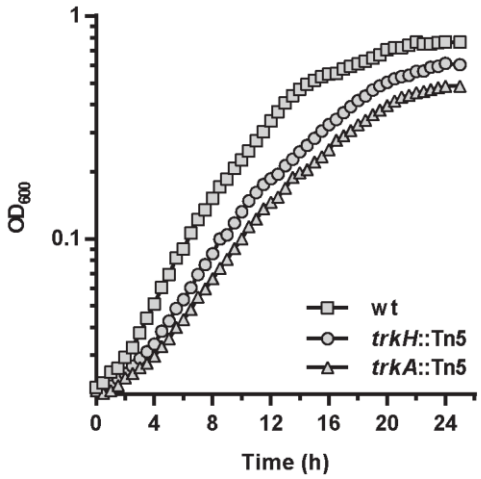

Fig. S1. Growth of wt and *trk* mutant *P. wasabiae*. Optical Density at 600 nm (OD_{600}) was measured throughout growth of wt (squares), *trkH*::Tn5 (RSV141, circles) and *trkA*::Tn5 (RSV236, triangles) *P. wasabiae* in MM.

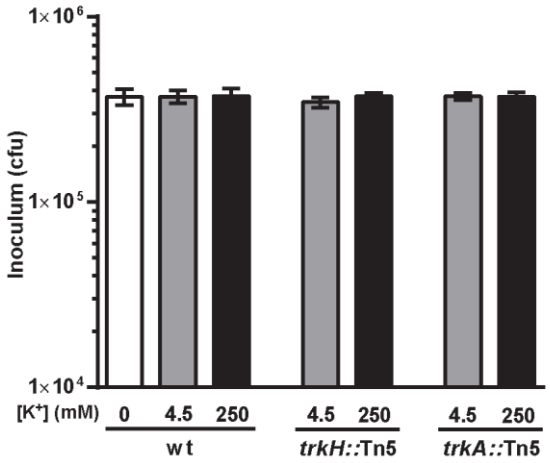

Fig. S2. Extracellular potassium concentration does not affect inoculum viability. The inoculum of wt, *trkH*::Tn5 and *trkA*::Tn5 when re-suspended in potassium-free PBS (0 mM of potassium, white bar), PBS (4.5 mM of potassium, grey bars) or PBS supplemented with potassium at a final concentration of 250 mM (black bars), was quantified by serial dilution and plating of the bacterial suspension before inoculation of potato tubers. Error bars represent SD, n = 3.

Table S1 – Strains and plasmids used in this study

Strains and Plasmids	Relevant Genotype	Source
P. wasabiae		
SCC3193	Wild-type	(14)
RSV206	SCC3193 carrying pRSV206	This study
RSV470	Δ*gacA*::*str*	This study
RSV503	RSV470 carrying pRSV206	This study
RSV124	*trkA*::Tn5*kan*	This study
RSV236	*trkA*::Tn5*kan*	This study
RSV302	RSV236 carrying pRSV206	This study
RSV562	Δ*gacA*::*str* in RSV236 background carrying pRSV206	This study
RSV108	*trkH*::Tn5*kan*	This study
RSV154	*trkH*::Tn5*kan*	This study
RSV141	*trkH*::Tn5*kan*	This study
RSV187	RSV141 carrying pRSV206	This study
RSV713	RSV206 carrying pOM18	This study
RSV711	RSV302 carrying pOM18	This study
RSV580	RSV302 carrying pRSV526	This study
RSV808	RSV187 carrying pOM18	This study
RSV802	RSV187 carrying pRSV781	This study
E. coli		
DH5α	DH5α	Laboratory stock
Plasmids		
pCMW1	Promoterless-*gfp* reporter vector, Kan^r	(16)

pUC18	Cloning vector, Amp^r	(19)
pKD46	λ Red recombinase expressing vector, Amp^r	(17)
pKD3	Vector containing chloramphenicol cassette, Cm^r	(17)
pKNG101	Vector containing streptomycin cassette, Str^r	(20)
pOM18	Cloning vector, $Spec^r$	This study
pRSV59	pCMW1 vector with chloramphenicol cassette, cm^r, kan^r	This study
pRSV206	pRSV59 containing $Promoter_{rsmB}$-*gfp*, cm^r, kan^r	This study
pRSV526	$trkA^+$	This study
pRSV781	$trkH^+$	This study

Table S2 – Primers used in this study.

Primers Name	Sequence
P0276-pKD3/4(BamHI)	ACGCGGATCCGTGTAGGCTGGAGCTGCTTC
P0277-pKD3/4(BamHI)	ACGCGGATCCCATATGAATATCCTCCTTAGT
P0528-rsmB(SphI)	GAGCATGCTGTCCTGGAAGAATTCGGCACG
P0529-rsmB(SalI)	CTGTCGACATCCGAGTGCAGTAACAGTG
P1106-pKNG101(XhoI)	AGCCTCGAGAGAGTCTTTGTTTTGACGCCAT
P1107-pKNG101(XhoI)	CTACTCGAGATCCTCTACGCCGGACGCATC
P0918-gacA(SalI)	GCAGTCGACGACACGACAGGCAGAATCAGA
P0919-gacA(SacI)	TAGGAGCTCAGAGCGGTTGCGATAGACG
P1097-Del.gacA(XhoI)	CGCCTCGAGAGATAATTCTCCAAAAAAGGG
P1098-Del.gacA(XhoI)	AGGCTCGAGGTGAGTGAGAGTTTCGATGC
P1148-trkA(SalI)	AGTGTCGACCGTACTCGCTGTCGATATTG
P1149-trkA(SacI)	CGTGAGCTCTCTTCCTGTTTACGACGCA
P1150-trkA(XhoI)	GGCCTCGAGTCCGGGAGCTAATTTATTACT
P1151-trkA(XhoI)	GGCCTCGAGAATAAGCCATGCCGTTTATTG
P1314-trkH(PstI)	TATCTGCAGGGGTAAACCGATGTTGTCGCA
P1306-trkH(XbaI)	AGCTCTAGAGGGACTATTCCCGCCAGAAG
P0052-Arb1K	GGCCACGCGTCGACTAGTACNNNNNNNNNNGATAT
P0053-Arb2K	GGCCACGCGTCGACTAGTAC
P0054-Arb6K	GGCCACGCGTCGACTAGTACNNNNNNNNNNACGCC
P0057-Kan_SP2	ACCTACAACAAAGCTCTCATCAACC
P0058-Kan_SP1	TGGCCTGTTGAACAAGTCTG

Table S3 – Mutants identified in this study

Mutant	Gene disrupted[a]	Gene name or putative homologue
RSV117	W5S_0007[b]	*ascG*
RSV126	W5S_4216	*damX*
RSV221	W5S_4216[b]	*damX*
RSV109	W5S_4607	*expI*
RSV135	W5S_4607	*expI*
RSV112	W5S_4607	*expI*
RSV101	W5S_4607	*expI*
RSV133	W5S_4607	*expI*
RSV138	W5S_4607	*expI*
RSV107	W5S_4607	*expI*
RSV224	W5S_4088	*rsgA*
RSV238	W5S_2585	*sapC*
RSV124	W5S_4132	*trkA*
RSV236	W5S_4132	*trkA*
RSV108	W5S_4400	*trkH*
RSV154	W5S_4400	*trkH*
RSV141	W5S_4400	*trkH*
RSV125	W5S_4168	*tusB*
RSV168	W5S_4218	*aroK*[c]
RSV241	W5S_3085[b]	*cydA*[c]
RSV231	W5S_4215	*dam*[c]
RSV144	W5S_2225	*galU*[c]
RSV267	W5S_2225	*galU*[c]
RSV240	W5S_1079	*hemL*[c]

RSV230	W5S_0715	*pnp*[c]
RSV119	W5S_0715	*pnp*[c]
RSV157	W5S_3391	*recB*[c]
RSV149	W5S_2530	Hypothetical protein
RSV113	W5S_4091	Hypothetical protein

[a] Genes numbered according to SCC3193 complete genome sequence NCBI taxid: 1166160

[b] Transposon inserted upstream of the gene

[c] Gene name according to the *E. coli* putative homologue

Table S4 – Growth Rates in Minimal Potassium Medium from Fig. S2.

Strain	Potassium Concentration			
	0.25 mM	**2.5 mM**	**25 mM**	**250 mM**
wt	0.18 h^{-1}	0.23 h^{-1}	0.20 h^{-1}	0.17 h^{-1}
trkH::Tn5	-	0.18 h^{-1}	0.18 h^{-1}	0.17 h^{-1}
trkA::Tn5	-	0.14 h^{-1}	0.15 h^{-1}	0.14 h^{-1}