

Characterization of a Ran gene from *Puccinia striiformis* f. sp. *tritici* involved in fungal growth and cell death

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Target	Score (Bits)	Expect	Alignment Length	Identities	Positives
▶ P. triticina 1-1 BBBB Race 1 (V2): PTTG_02103.4: GTP-binding nuclear protein spi1	331.257	0.0	184	156	168
▶ P. graminis tritici: PGTG_04433.4: GTP-binding nuclear protein spi1	328.946	0.0	184	155	167
▶ P. striiformis PST-78: PSTG_13752.1: GTP-binding nuclear protein spi1	328.946	0.0	184	156	166
▶ P. graminis tritici: PGTG_17558.4: GTP-binding protein ypt1	110.538	1.03353E-24	161	58	91
▶ P. triticina 1-1 BBBB Race 1 (V2): PTTG_05460.4: GTP-binding protein ypt1	110.538	1.10487E-24	161	58	91
▶ P. triticina 1-1 BBBB Race 1 (V2): PTTG_10008.4: GTP-binding protein ypt5	109.768	1.54262E-24	175	62	98
▶ P. graminis tritici: PGTG_09844.4: rab family protein	108.612	3.55322E-24	173	61	97
▶ P. striiformis PST-78: PSTG_11722.1: GTP-binding protein ypt5	108.612	3.83032E-24	162	58	94
▶ P. graminis tritici: PGTG_15757.4: rab family protein	102.064	3.86465E-22	182	56	97
▶ P. triticina 1-1 BBBB Race 1 (V2): PTTG_04650.4: GTP-binding protein ryh1	101.293	5.53258E-22	182	56	97
▶ P. graminis tritici: PGTG_00987.4: Ras-like protein Rab-8A	99.7525	1.59636E-21	161	51	88
▶ P. striiformis PST-78: PSTG_12637.1: GTP-binding protein ryh1	99.3673	2.03337E-21	185	58	100
▶ P. striiformis PST-78: PSTG_11158.1: Ras-like protein Rab-8A	99.3673	2.48418E-21	161	50	88
▶ P. triticina 1-1 BBBB Race 1 (V2): PTTG_07767.4: GTP-binding protein ypt3	98.9821	2.70035E-21	154	54	82

⇒ Ran proteins

⇒ Rab proteins

Figure S1. A BLAST search using the know Ran protein GSP1 from *S. cerevisiae* as the query in the Broad Institute *Puccinia* database. *PSTG_13752.1* indicates *PsRan*.

Target	Score (Bits)	Expect	Alignment Length	Identities	Positives
▶ P. triticina 1-1 BBBB Race 1 (V2): PTTG_02103.4: GTP-binding nuclear protein spi1	345.51	0.0	195	161	175
▶ P. graminis tritici: PGTG_04433.4: GTP-binding nuclear protein spi1	343.584	0.0	195	160	174
▶ P. striiformis PST-78: PSTG_13752.1: GTP-binding nuclear protein spi1	343.584	0.0	195	161	173
▶ P. graminis tritici: PGTG_17558.4: GTP-binding protein ypt1	110.153	1.16146E-24	161	57	91
▶ P. triticina 1-1 BBBB Race 1 (V2): PTTG_05460.4: GTP-binding protein ypt1	109.768	1.76274E-24	161	57	91
▶ P. graminis tritici: PGTG_09844.4: rab family protein	107.457	7.91486E-24	162	58	94
▶ P. triticina 1-1 BBBB Race 1 (V2): PTTG_10008.4: GTP-binding protein ypt5	107.457	8.74836E-24	162	58	94
▶ P. striiformis PST-78: PSTG_11722.1: GTP-binding protein ypt5	107.457	9.04527E-24	162	58	94
▶ P. graminis tritici: PGTG_15757.4: rab family protein	100.523	9.20281E-22	183	57	97
▶ P. triticina 1-1 BBBB Race 1 (V2): PTTG_04650.4: GTP-binding protein ryh1	100.523	9.51514E-22	183	57	97
▶ P. graminis tritici: PGTG_00987.4: Ras-like protein Rab-8A	99.7525	1.87038E-21	161	51	88
▶ P. striiformis PST-78: PSTG_12637.1: GTP-binding protein ryh1	99.3673	2.44279E-21	183	56	98
▶ P. striiformis PST-78: PSTG_11158.1: Ras-like protein Rab-8A	98.5969	3.73846E-21	161	50	88
▶ P. triticina 1-1 BBBB Race 1 (V2): PTTG_05367.4: Ras-like protein Rab-8A	98.5969	3.96331E-21	161	50	88
▶ P. triticina 1-1 BBBB Race 1 (V2): PTTG_07767.4: GTP-binding protein ypt3	98.2117	4.84201E-21	154	54	82

⇒ Ran proteins

⇒ Rab proteins

Figure S2. A BLAST search using the know Ran protein GSP2 from *S. cerevisiae* as the query in the Broad Institute *Puccinia* database. *PSTG_13752.1* indicates *PsRan*.

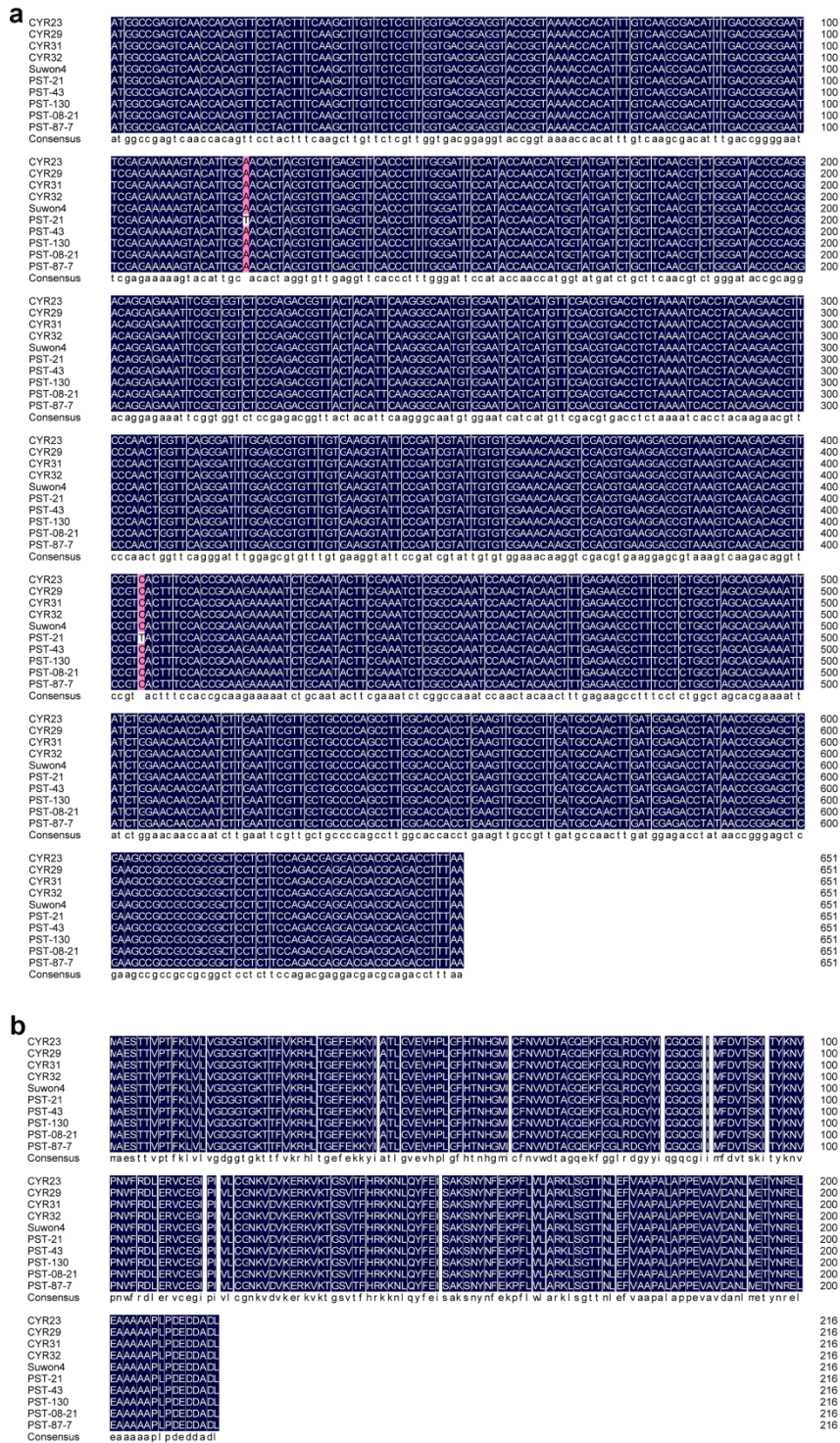


Figure S3. Sequence alignment of *PsRan* among different *Pst* isolates in the nucleotide level (a) and amino acid level (b). Ten *Pst* isolates were used, including five Chinese isolates (CYR23, CYR29, CYR31, CYR32, and Su11-4), three US isolates (PST-21, PST-43, and PST-130) and two UK isolates (PST-08-21 and PST-87-7).

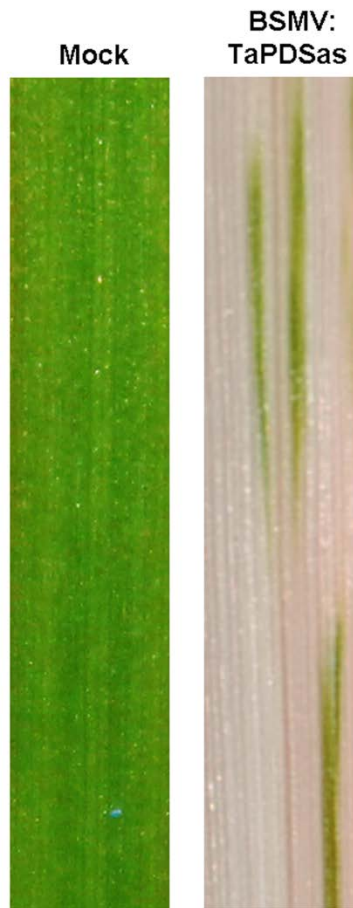


Figure S4. Phenotypes of wheat plants 10 dpi with FES buffer (Mock) and BSMV:TaPDS, respectively.

Table S1 Primers used in this study.

Primer name	Primer 5'-3'	Purpose
PsEF1-qRT-F	TTCGCCGTCCGTGATATGAGACAA	The endogenous reference for normalization in qRT-PCR
PsEF1-qRT-R	ATGCGTATCATGGTGGTGGAGTGA	The endogenous reference for normalization in qRT-PCR
PsRan-qRT-F	ATTTGGAGCGTGTGGTGTGAA	qRT-PCR for validation of the expression patterns of PsRan
PsRan-qRT-R	GACGGAACCTGTCTTGACTTTA	qRT-PCR for validation of the expression patterns of PsRan
PsRan-PacI-F1	CCTTAATTAAGACCCAGATTCCGTTTACG	Clone PsRan-1as to the r vector for HIGS
PsRan-NotI-R1	TATGCGGCCGCTACCGGTACCTCCGTCAC	Clone PsRan-1as to the r vector for HIGS
PsRan-PacI-F2	CCTTAATTAAGCCTTGGCACCACCTGA	Clone PsRan-2as to the r vector for HIGS
PsRan-NotI-R2	TATGCGGCCGCGTCTGCGTCGTCCTCGT CT	Clone PsRan-2as to the r vector for HIGS
PsRan-SpeI-F	GACTAGTATGGCCGAGTCAACCACAGT	Clone PsRan to pCAMBIA-1302 for expression in <i>N. benthamiana</i>
PsRan-AvrII-R	TTCTAGGAAGTCTGCGTCGTCCTCG	Clone PsRan to pCAMBIA-1302 for expression in <i>N. benthamiana</i>
PsRan-ClaI-F	GGCATCGATATGGCCGAGTCAACCACAGT	Clone PsRan to PVX for expression in <i>N. benthamiana</i>
PsRan-SmaI-R	GTACCCGGGTTAAAGGTCTGCGTCGTCCT C	Clone PsRan to PVX for expression in <i>N. benthamiana</i>
GFP-XhoI-F	CCGCTCGAGATGGTGAGCAAGGGCGA	Clone GFP to pREP3X for expression in <i>S. pombe</i>
GFP-BamHI-R	GCGGATCCCTTGACAGCTCGTCCATGC	Clone GFP to pREP3X for expression in <i>S. pombe</i>
Fusion-XhoI-F1	CCGCTCGAGATGGTGAGCAAGGGCGA	Overlapping PCR
Fusion-R1	GGTTGACTCGGCCATCTTGACAGCTCGT C	Overlapping PCR
Fusion-F2	GACGAGCTGTACAAGATGGCCGAGTCAA CC	Overlapping PCR
Fusion-BamHI-R2	GCGGATCCCTAAAGGTCTGCGTCGTCCT	Overlapping PCR
TaEF1-qRT-F	TGGTGTCAATCAAGCCTGGTATGGT	Calculation of fungal biomass
TaEF1-qRT-R	ACTCATGGTGCATCTCAACGGACT	Calculation of fungal biomass