

Supplementary Materials to accompany the manuscript:

Exploiting a phage-bacterium interaction system as a molecular switch to decipher macromolecular interactions in the living cell

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SUPPLEMENTARY MATERIALS

Supplementary Table S1. Stl mutants identified with reduced DNA binding ability in the Stl switch system

Number of mutations	Stl mutants
1 mutation	Stl ^{K19K}
	Stl ^{E59K}
	Stl ^{G62Afs*88}
	Stl ^{G66Afs*88}
	Stl ^{I123T}
	Stl ^{Y143Y}
	Stl ^{V144A}
	Stl ^{R177H}
	Stl ^{K214*}
	Stl ^{A236T}
	Stl ^{K238E}
2 mutations	Stl ^{Q6H, S76T}
	Stl ^{G15S, K240R}
	Stl ^{T16S, D142A}
	Stl ^{G92D, V229M}
	Stl ^{I134V, H256R}
	Stl ^{K158N, I220V}
3 mutations	Stl ^{D174V, G185G, K193K}
4 mutations	Stl ^{I18T, Y98H, D142D, R227C}

	Stl ^{V55M, F79L, N137D, I161T}
5 mutations	Stl ^{Y70C, K80K, K93R, L152R, I181Lfs*188}
6 mutations	Stl ^{M1R^a, G54V, L129P, D140E, E186V, H188H}
	Stl ^{I17V, F38S, L72P, G92S, D95G, D108N}
	Stl ^{N41D, E186V, R227R, D235D, K240R, K244E}
7 mutations	Stl ^{I17T, S76S, G92A, I212T, L222P, R227R, I237I}
8 mutations	Stl ^{S30G, Y84H, I134A, Y143H, N168S, L194L, E224E, Q257Q}
10 mutations	Stl ^{F38L, H46Y, N48S, I58T, L65P, P86Q, Y112H, S114N, N135I, N203S}
11 mutations	Stl ^{K31N, I53I, K63K, I67I, R74H, K93K, D155D, K193E, T197A, L245P, Y246*}

^apKW08-Stl vector is containing an AU-tag before the coding sequence of Stl (Hirmondó et al. 2015, DNA Repair), therefore a protein may be translated from this mutant despite the first Met is mutated

Supplementary Table S2. Oligonucleotides used in the present study

Restriction sites are underlined

Used in	Oligo name	5'-3' sequence
Cloning of p2NIL-LacZ ^{Str} -INT plasmid	Sall_str	TTT <u>TAGTCGAC</u> CATATTCTCACCTCCTCGAAC
	HindIII_str	TGTGT <u>AAGCTT</u> CATATTCTCACCTCCTCGAAC
	BglII_LacZ	TGTGT <u>AGATCT</u> GTCGTTGTGGTCACTCG
	Sall_LacZ	TATAT <u>GTCGAC</u> CGCCCAAACATGCATGGAT
	NotI_INT_for	ATATAG <u>CGGCGC</u> GCTGCTCCATAACATCAAACATC
	NotI_INT_rev	ATATAG <u>CGGCGC</u> GGAAGCTTGCATGCCTGC
Cloning of pKW08-Stl ^{C-term}	Stl-Cterm_Au_BamHI_f	ATTAGGATCCATGGATACGTATCGCTACATAAGCCCGACCCTGAACG
	Stl_HindIII_r	ATTA <u>AAAGCTT</u> GCGGCCGCTTAGTTGGTATC
Error prone PCR, Cloning of pKW08-Stl ^{A236T} and pKW08-Stl ^{MUT} , and pKW08-Stl ^{AA}	Stl_Au_BamHI_f	AATTAGGATCCATGGATACGTATCGCTACATAGCTAGCC
	Stl_HindIII_r	ATTA <u>AAAGCTT</u> GCGGCCGCTTAGTTGGTATC
Colony PCR for sequencing	Stl_seq_f	GGTGGTGAGTCATAGTTGC
	Stl_seq_r	CGCTTAATCCAAAGTTCAAACG
EMSA	Stl-Str	TCGTAAACATATTCTCACCTCCTCGAACAAATTATCTCACATCGAGATATTTATTTCAACATTA AATATTGCAAATTGAGATATTTTTTCGATATGATATCATTTGGATGGAAGGAGCTGGTCAAA TGGCAGAATTACCAACACATTACGGCACAATTATTA ^{AA} ACTCTTAGAAAATACATGAAATTAA CTCAAAGCAAATTGAGTGAAAGGACAGGATTTAGGATCC

Supplementary Table S3. Plasmids used in the present study

Plasmid name	Characteristics	Antibiotic Resistance	Reference
p2NIL-LacZ ^{Str} -INT	reporter plasmid; LacZ; L5 integration cassette	Kan ^R	this study
pKW08-Stl	expression of Stl in Mycobacterium; Tet-inducible	Hyg ^R	Hirmondo et al., 2015
pKW08-Stl ^{C-term}	expression of C-terminal part of Stl in Mycobacterium; Tet-inducible	Hyg ^R	this study
pKW08-Stl ^{AA}	expression of AA mutant Stl in Mycobacterium; Tet-inducible	Hyg ^R	this study
pKW08-Stl ^{MUT}	expression of random mutagenized Stl in Mycobacterium; Tet-inducible	Hyg ^R	this study
pKW08-Stl ^{A236T}	expression of A236T mutant Stl in Mycobacterium; Tet-inducible	Hyg ^R	this study
pGex-4T-1-Stl	protein expression of Stl, Gluthation-S-transferase tag	CA ^R	Nyiri et al., 2015
pGex-4T-1-Stl ^{A236T}	protein expression of A236T mutant Stl, Gluthation-S-transferase tag	CA ^R	this study
pSJ27- ϕ dut	expression of ϕ 11 dUTPase in Mycobacterium; Bxb1 integration cassette	Cm ^R	this study