

# SUPPLEMENTARY MATERIAL

## The HaloTag: Improving Soluble Expression and Applications in Protein Functional Analysis

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**Supplementary Table 1.** Expression and solubility of proteins of interest with various HaloTag expression vectors

Locus ID	Protein name	Species	pFN18A		pGW-nHalo		pFN19A		pFC20A		pFC14A	
			Exp	Sol	Exp	Sol	Exp	Sol	Exp	Sol	Exp	Sol
b0071	LeuC	<i>Escherichia coli str. K12 substr. W3110</i>					1	1	1	1		
b0072	LeuD	<i>Escherichia coli str. K12 substr. W3110</i>					1	1	1	1		
b2023	HisF	<i>Escherichia coli str. K12 substr. W3110</i>					0	0	0	0		
b2025	HisH	<i>Escherichia coli str. K12 substr. W3110</i>					1	1	1	1		
b2231	RpoA	<i>Escherichia coli str. K12 substr. W3110</i>					1	1	1	1		
b3295	RpoB	<i>Escherichia coli str. K12 substr. W3110</i>					1	1	0	0		
b3699	GyrA	<i>Escherichia coli str. K12 substr. W3110</i>					1	1	1	1		
b3987	GyrB	<i>Escherichia coli str. K12 substr. W3110</i>					1	1	1	1		
BMA_0267	site-specific recombinase, phage integrase family, truncation	<i>B. mallei</i> ATCC23344			1	0						
BMA_0429	cytidylate kinase	<i>B. mallei</i> ATCC23344			1	1						
BMA_2469	transketolase	<i>B. mallei</i> ATCC23344			1	1						
BMA_3281	flagellar M-ring protein FliF	<i>B. mallei</i> ATCC23344			0	0						
BMA_A0079_26_486	plcN	<i>B. mallei</i> ATCC23344			0	0						
BMA_A0079_519_718	plcN	<i>B. mallei</i> ATCC23344			1	1						
BMA_A0238	hypothetical protein	<i>B. mallei</i> ATCC23344			1	0						
BMA_A0410_1_519	vgrG3	<i>B. mallei</i> ATCC23344			0	0						
BMA_A0446	vgrG2.2	<i>B. mallei</i> ATCC23344			1	1						
BMA_A0553	Ser/Thr protein phosphatase family protein	<i>B. mallei</i> ATCC23344			0	0						
BMA_A0679	phosphatase, CheC family	<i>B. mallei</i> ATCC23344			1	1						
BMA_A0728	type VI secretion system protein TssN	<i>B. mallei</i> ATCC23344			1	1						

**Supplementary Table 1. Contd....**

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			Exp	Sol	Exp	Sol	Exp	Sol	Exp	Sol	Exp	Sol
PB1	PB1	Influenza A (A/New York/1682/2009(H1N1))									1	N/A
PB2	PB2	Influenza A (A/New York/1682/2009(H1N1))									1	N/A
SP_0058	transcriptional regulator, GntR family	<i>S. pneumoniae TIGR4</i>					0	0	1	1		
SP_0064	PTS system; IIA component	<i>S. pneumoniae TIGR4</i>			1	1						
SP_0238	ACT domain protein	<i>S. pneumoniae TIGR4</i>							1	0		
SP_0291	GTP cyclohydrolase I (foLE)	<i>S. pneumoniae TIGR4</i>			1	1			1	1		
SP_0308	PTS system; IIA component	<i>S. pneumoniae TIGR4</i>			1	1			1	1		
SP_0321	PTS system; IIA component	<i>S. pneumoniae TIGR4</i>			1	1			1	1		
SP_0360	UDP-N-acetylglucosamine 2-epimerase (cps4L)	<i>S. pneumoniae TIGR4</i>			1	1	1	1	1	1		
SP_0360	UDP-N-acetylglucosamine 2-epimerase	<i>S. pneumoniae TIGR4</i>					1	1				
SP_0385	conserved hypothetical protein	<i>S. pneumoniae TIGR4</i>			1	1						
SP_0416	transcriptional regulator, MarR family	<i>S. pneumoniae TIGR4</i>					0	0				
SP_0435	translation elongation factor P (efp)	<i>S. pneumoniae TIGR4</i>			1	1			0	0		
SP_0488	conserved hypothetical protein	<i>S. pneumoniae TIGR4</i>			0	0			0	0		
SP_0501	transcriptional regulator, MerR family	<i>S. pneumoniae TIGR4</i>					1	1	1	1		
SP_0526	response regulator BlpR	<i>S. pneumoniae TIGR4</i>					1	1	0	0		
SP_0604	sensor histidine kinase VncS (vncS)	<i>S. pneumoniae TIGR4</i>			0	0			1	1		
SP_0694	conserved domain protein	<i>S. pneumoniae TIGR4</i>			1	0			0	0		
SP_0723	conserved domain protein	<i>S. pneumoniae TIGR4</i>			1	0			0	0		
SP_0741	conserved hypothetical protein	<i>S. pneumoniae TIGR4</i>					0	0				
SP_0789	conserved hypothetical protein	<i>S. pneumoniae TIGR4</i>			1	1			1	1		
SP_0798	DNA-binding response regulator CiaR	<i>S. pneumoniae TIGR4</i>					1	1	1	1		
SP_0805	hydrolase; haloacid dehalogenase-like family	<i>S. pneumoniae TIGR4</i>			1	1						
SP_0845	lipoprotein	<i>S. pneumoniae TIGR4</i>			1	1			0	0		
SP_0851	conserved hypothetical protein	<i>S. pneumoniae TIGR4</i>							1	0		
SP_0937	conserved hypothetical protein	<i>S. pneumoniae TIGR4</i>			1	1			1	1		
SP_0945	ribosome recycling factor (frr)	<i>S. pneumoniae TIGR4</i>			1	1						
SP_0954	competence protein CelA	<i>S. pneumoniae TIGR4</i>			1	0			0	0		
SP_0979	oligoendopeptidase F (pepF)	<i>S. pneumoniae TIGR4</i>			1	1			1	1		
SP_0998	hypothetical protein	<i>S. pneumoniae TIGR4</i>					1	1				
SP_1050	putative transcriptional regulator	<i>S. pneumoniae TIGR4</i>					1	1	1	1		

Supplementary Table 1. Contd.....

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			Exp	Sol	Exp	Sol	Exp	Sol	Exp	Sol	Exp	Sol
SP_1102	conserved hypothetical protein TIGR00103	<i>S. pneumoniae</i> TIGR4			1	1			1	1		
SP_1118	pullulanase; putative	<i>S. pneumoniae</i> TIGR4			0	0						
SP_1130	transcriptional regulator	<i>S. pneumoniae</i> TIGR4					1	1	1	1		
SP_1131	putative transcriptional regulator	<i>S. pneumoniae</i> TIGR4					0	0	1	0		
SP_1132	hypothetical protein	<i>S. pneumoniae</i> TIGR4							1	1		
SP_1227	DNA-binding response regulator	<i>S. pneumoniae</i> TIGR4					0	0	1	1		
SP_1249	guanosine monophosphate reductase (guaC)	<i>S. pneumoniae</i> TIGR4			1	1			0	0		
SP_1362	putative transcriptional regulator MecA	<i>S. pneumoniae</i> TIGR4					1	1	1	1		
SP_1372	conserved hypothetical protein	<i>S. pneumoniae</i> TIGR4			0	0			0	0		
SP_1423	putative transcriptional repressor	<i>S. pneumoniae</i> TIGR4					1	1	0	0		
SP_1504	TPR domain protein	<i>S. pneumoniae</i> TIGR4			1	1			1	1		
SP_1524	aminotransferase; class II	<i>S. pneumoniae</i> TIGR4			1	1						
SP_1572	non-heme iron-containing ferritin	<i>S. pneumoniae</i> TIGR4			1	1			1	1		
SP_1631	threonyl-tRNA synthetase (thrS)	<i>S. pneumoniae</i> TIGR4			1	1			1	1		
SP_1638	iron-dependent transcriptional regulator	<i>S. pneumoniae</i> TIGR4					1	1	0	0		
SP_1650	manganese ABC transporter; manganese-binding adhesion lipoprotein (psaA)	<i>S. pneumoniae</i> TIGR4			1	1			1	1		
SP_1671	D-alanine--D-alanine ligase (ddlA)	<i>S. pneumoniae</i> TIGR4			1	1			1	1		
SP_1699	holo-(acyl-carrier protein) synthase (acpS)	<i>S. pneumoniae</i> TIGR4			1	1	1	1	0	0		
SP_1714	transcriptional regulator, GntR family	<i>S. pneumoniae</i> TIGR4					1	1	1	1		
SP_1752	mechanosensitive ion channel; putative	<i>S. pneumoniae</i> TIGR4			1	1			0	0		
SP_1774	putative transcriptional regulator	<i>S. pneumoniae</i> TIGR4					1	1	1	1		
SP_1797	ABC transporter, permease protein	<i>S. pneumoniae</i> TIGR4			1	1						
SP_1799	sugar-binding transcriptional regulator, LacI family	<i>S. pneumoniae</i> TIGR4					1	1	1	1		
SP_1802	hypothetical protein	<i>S. pneumoniae</i> TIGR4			1	1			1	0		
SP_1802	hypothetical protein	<i>S. pneumoniae</i> TIGR4					1	1				
SP_1813	N-(5'-phosphoribosyl)-anthranilate isomerase (trpF)	<i>S. pneumoniae</i> TIGR4			1	1			0	0		
SP_1823	MgtC/SapB family protein	<i>S. pneumoniae</i> TIGR4			1	1			0	0		
SP_1863	transcriptional regulator, MarR family	<i>S. pneumoniae</i> TIGR4					0	0				
SP_1910	conserved hypothetical protein	<i>S. pneumoniae</i> TIGR4			0	0			1	1		
SP_1925	hypothetical protein	<i>S. pneumoniae</i> TIGR4			1	1			1	1		
SP_1959	nucleoside diphosphate kinase (ndk)	<i>S. pneumoniae</i> TIGR4			1	1			1	1		

Supplementary Table 1. Contd.....

Locus ID	Protein name	Species	pFN18A		pGW-nHalo		pFN19A		pFC20A		pFC14A	
			Exp	Sol	Exp	Sol	Exp	Sol	Exp	Sol	Exp	Sol
SP_1978	diaminopimelate decarboxylase (lysA)	<i>S. pneumoniae TIGR4</i>			1	1			1	1		
SP_1980	cmp-binding-factor 1	<i>S. pneumoniae TIGR4</i>			1	1			1	1		
SP_1999	catabolite control protein A	<i>S. pneumoniae TIGR4</i>					1	1	1	1		
SP_2039	conserved hypothetical protein	<i>S. pneumoniae TIGR4</i>			1	1			1	1		
SP_2077	putative transcriptional repressor	<i>S. pneumoniae TIGR4</i>					0	0	0	0		
SP_2105	hypothetical protein	<i>S. pneumoniae TIGR4</i>			1	1	0	0	0	0		
SP_2105	hypothetical protein	<i>S. pneumoniae TIGR4</i>					0	0				
SP_2119	putative transcriptional regulator	<i>S. pneumoniae TIGR4</i>					1	1	1	1		
SP_2209	conserved hypothetical protein	<i>S. pneumoniae TIGR4</i>			1	1			1	1		
SP_2235	response regulator ComE	<i>S. pneumoniae TIGR4</i>					1	1	1	1	1	
y0011	Hypothetical protein	<i>Y. pestis Kim10</i>					1	1				
Y0018	putative IS element protein	<i>Y. pestis Kim10</i>	1	1								
Y0043	Yop secretion and targeting control protein	<i>Y. pestis Kim10</i>	0	0								
Y0045	hypothetical protein	<i>Y. pestis Kim10</i>	1	1								
Y0046	hypothetical protein	<i>Y. pestis Kim10</i>	1	1								
Y0048	low calcium response protein R	<i>Y. pestis Kim10</i>	1	0								
Y0049	low calcium response protein G	<i>Y. pestis Kim10</i>	1	1								
Y0050	secreted effector protein	<i>Y. pestis Kim10</i>	0	0								
Y0052	secreted effector protein	<i>Y. pestis Kim10</i>	1	1								
Y0078	secretion chaperone	<i>Y. pestis Kim10</i>	1	1								
Y0082	secretion chaperone	<i>Y. pestis Kim10</i>	1	1								
y0549	Hypothetical protein	<i>Y. pestis Kim10</i>					1	1				
y0636	Hypothetical protein	<i>Y. pestis Kim10</i>					1	1				
y0931	Hypothetical protein	<i>Y. pestis Kim10</i>					1	1				
y0974	Hypothetical protein	<i>Y. pestis Kim10</i>					0	0	0	0		
y1125	Hypothetical protein	<i>Y. pestis Kim10</i>					1	1				
y1333	Hypothetical protein	<i>Y. pestis Kim10</i>					0	0	0	0		
y1724	Hypothetical protein	<i>Y. pestis Kim10</i>					1	1				
y1726	Hypothetical protein	<i>Y. pestis Kim10</i>					1	1				
y2136	Hypothetical protein	<i>Y. pestis Kim10</i>					0	0				
y2245	Hypothetical protein	<i>Y. pestis Kim10</i>					1	1				
y2259	Hypothetical protein	<i>Y. pestis Kim10</i>					1	0				
y2737	Hypothetical protein	<i>Y. pestis Kim10</i>					1	0				
y3130	Hypothetical protein	<i>Y. pestis Kim10</i>					1	1				

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Locus ID	Protein name	Species	pFN18A		pGW-nHalo		pFN19A		pFC20A		pFC14A	
			Exp	Sol	Exp	Sol	Exp	Sol	Exp	Sol	Exp	Sol
y3658	Hypothetical protein	<i>Y. pestis Kim10</i>	0	0								
y3659	Hypothetical protein	<i>Y. pestis Kim10</i>	0	0								
y3660	Hypothetical protein	<i>Y. pestis Kim10</i>	1	1								
y3663	Hypothetical protein	<i>Y. pestis Kim10</i>	1	1								
y3664	Hypothetical protein	<i>Y. pestis Kim10</i>	0	0								
y3665	Hypothetical protein	<i>Y. pestis Kim10</i>	1	1								
y3668	Hypothetical protein	<i>Y. pestis Kim10</i>	0	0								
y3669	putative ATP-dependent protease	<i>Y. pestis Kim10</i>	1	1								
y3671	Hypothetical protein	<i>Y. pestis Kim10</i>	1	1								
y3673	Hypothetical protein	<i>Y. pestis Kim10</i>	1	0								
y3674	Hypothetical protein	<i>Y. pestis Kim10</i>	1	0								
y3675	Hypothetical protein	<i>Y. pestis Kim10</i>	1	1								
y3676	Hypothetical protein	<i>Y. pestis Kim10</i>	1	0								
y3714	Hypothetical protein	<i>Y. pestis Kim10</i>							1	0	0	0
y3978	Hypothetical protein	<i>Y. pestis Kim10</i>							1	1		

Proteins were expressed in HEK293T (pFC14A) and in BL21(DE3)/pMagic (pFN18A, pFN19A, pFC20A and pGW-nHalo)

0: No expression or Insoluble

1: Expression or Soluble