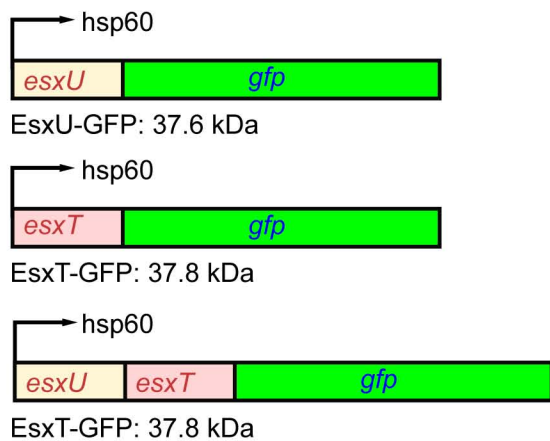
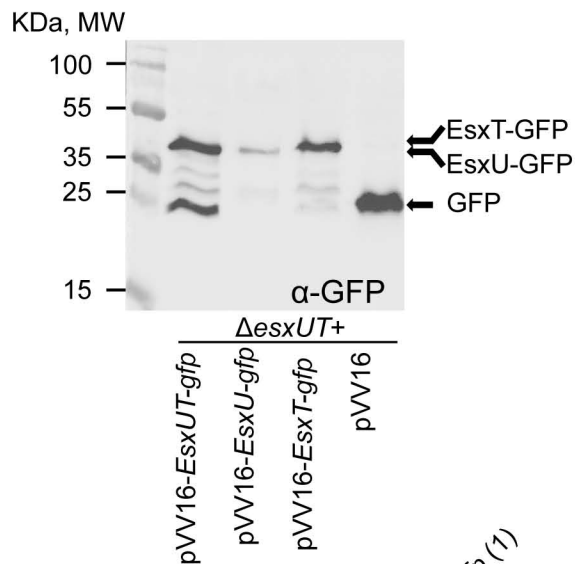


Sup. Figure 1

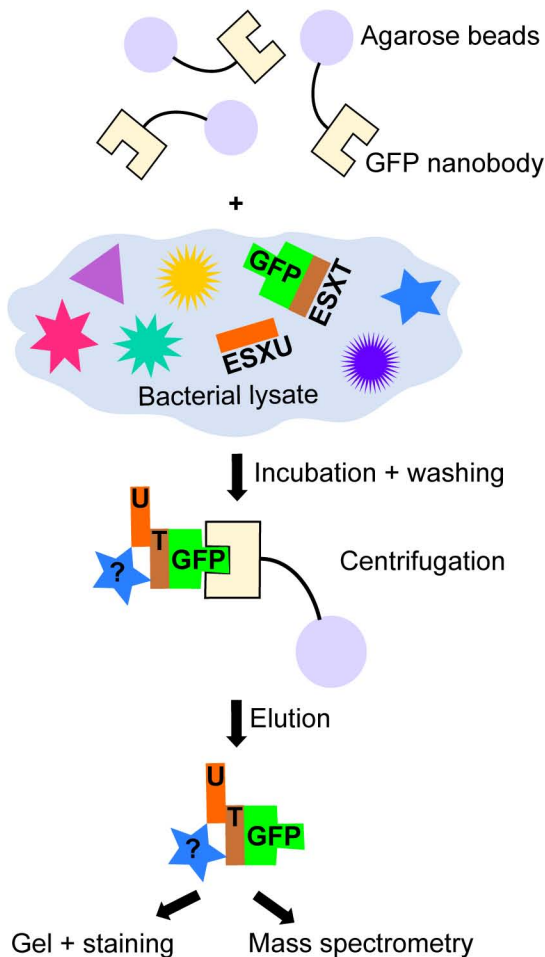
A



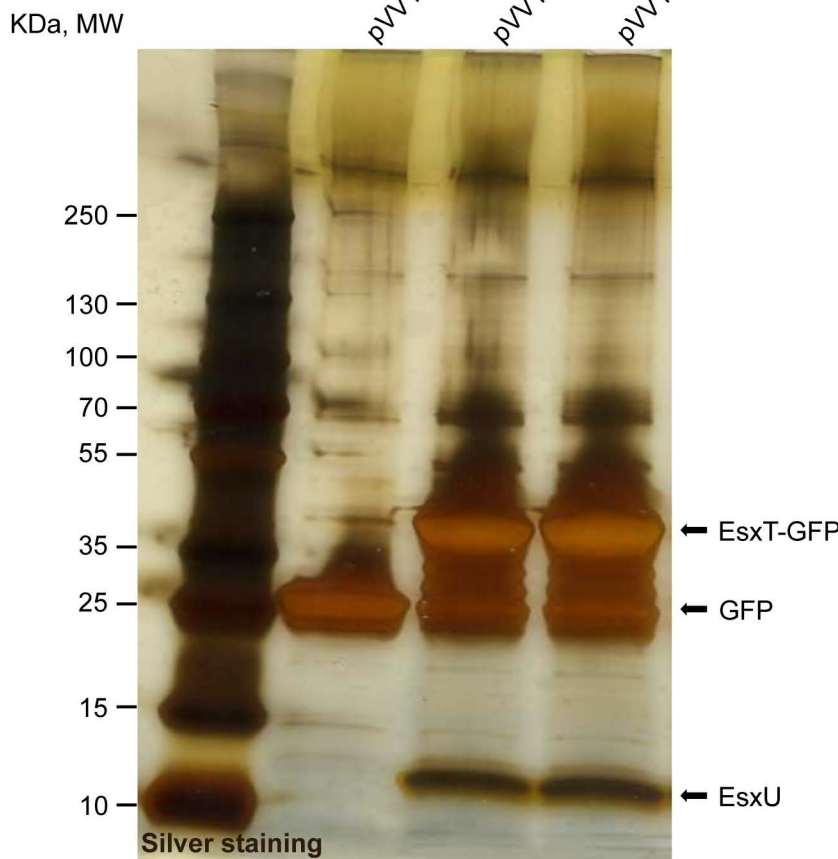
B



C



D

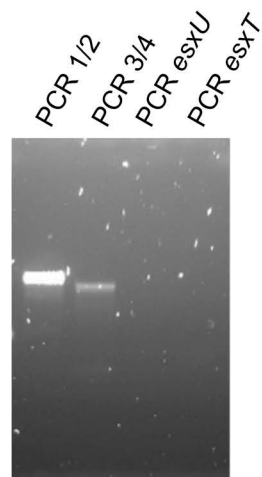
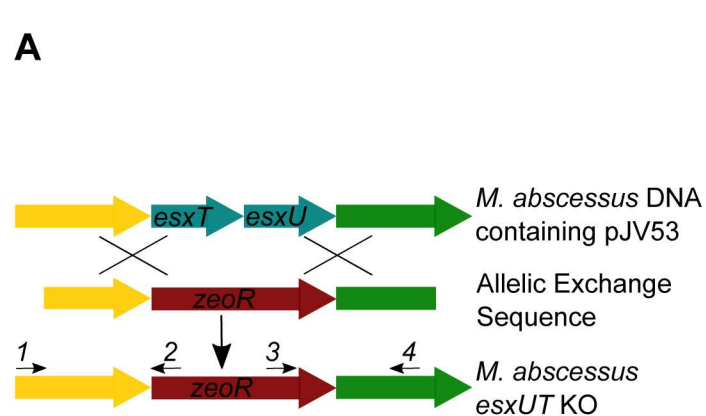


E

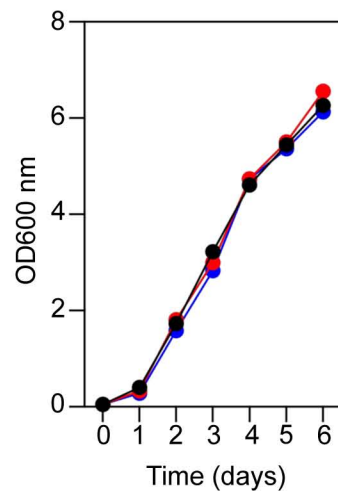
Protein name	Accession number	MW (kDa)	Total number of unique peptides		
			Control	Pull-down-1	Pull-down-2
EsxT	<i>Mab_3753c</i>	10.7	0	72	96
EsxU	<i>Mab_3754c</i>	10.5	0	47	52
rpoC	<i>Mab_3868c</i>	146.5	1	9	14
Cpn60.2	<i>Mab_650</i>	56.4	2	14	6
Ag84	<i>Mab_2017</i>	30	0	2	1

Sup. Figure 2

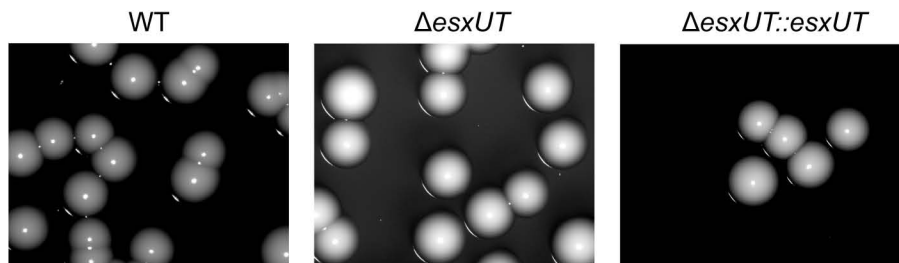
A



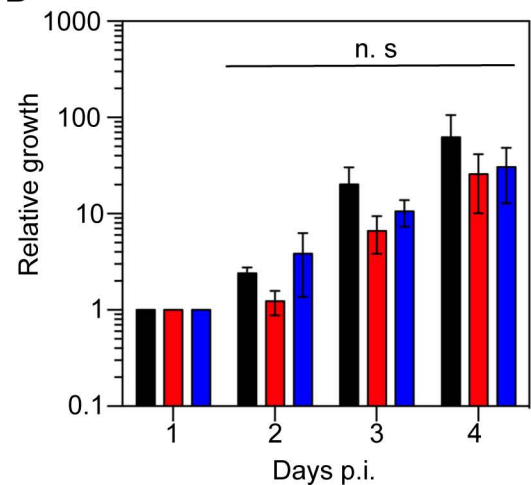
B



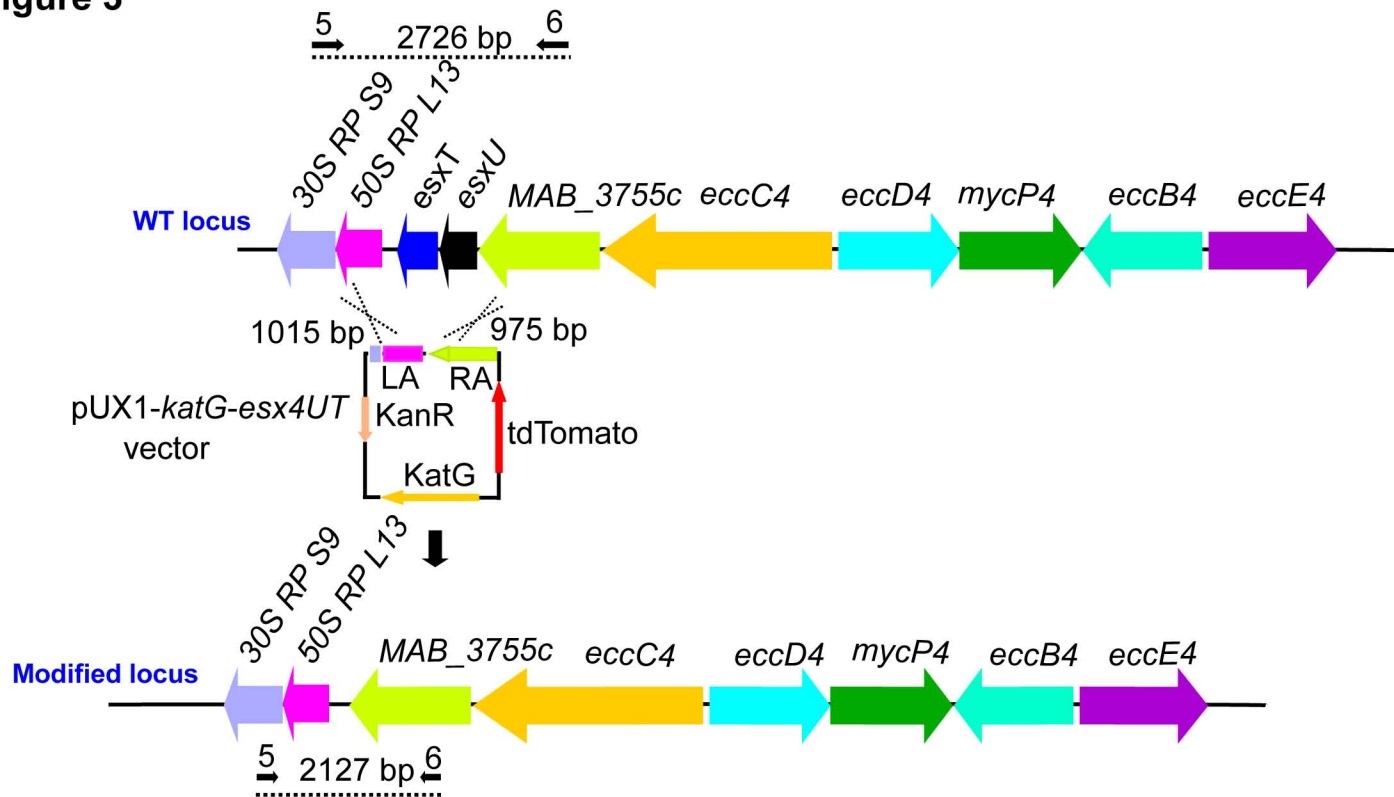
C



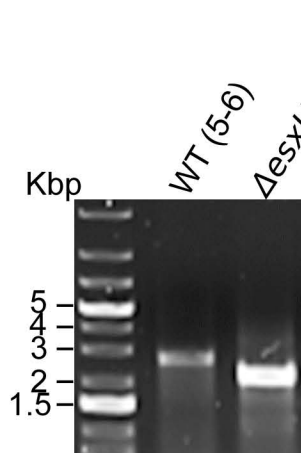
D



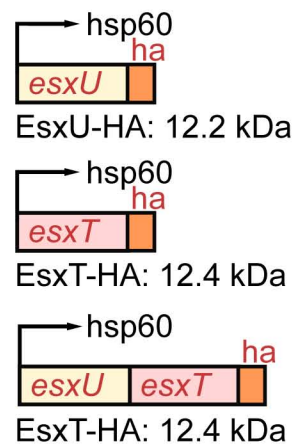
A



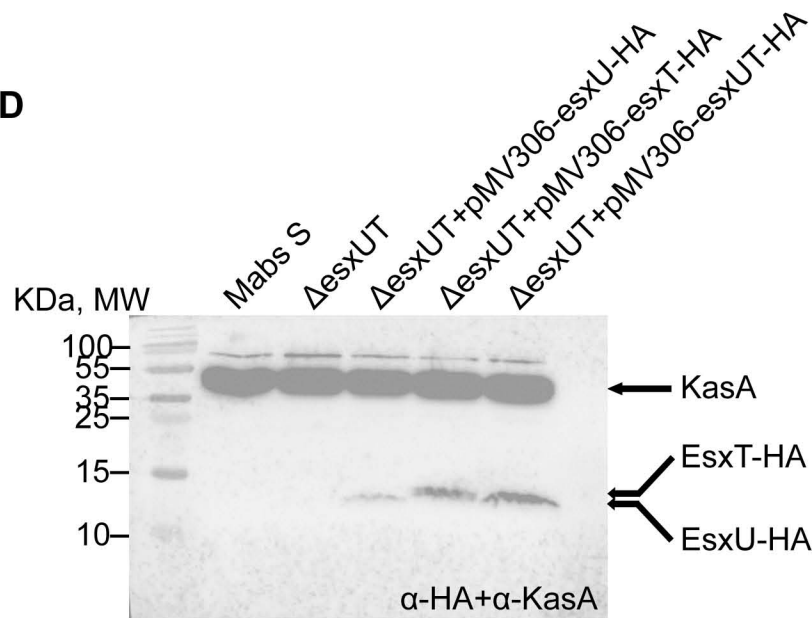
B



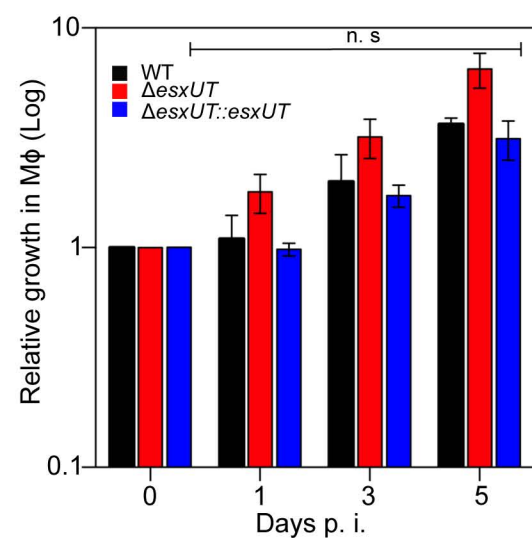
C



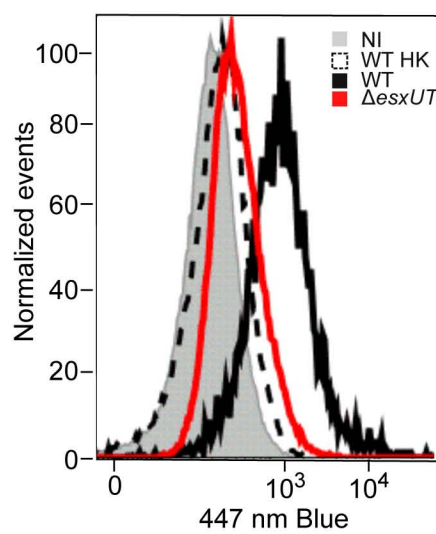
D



E

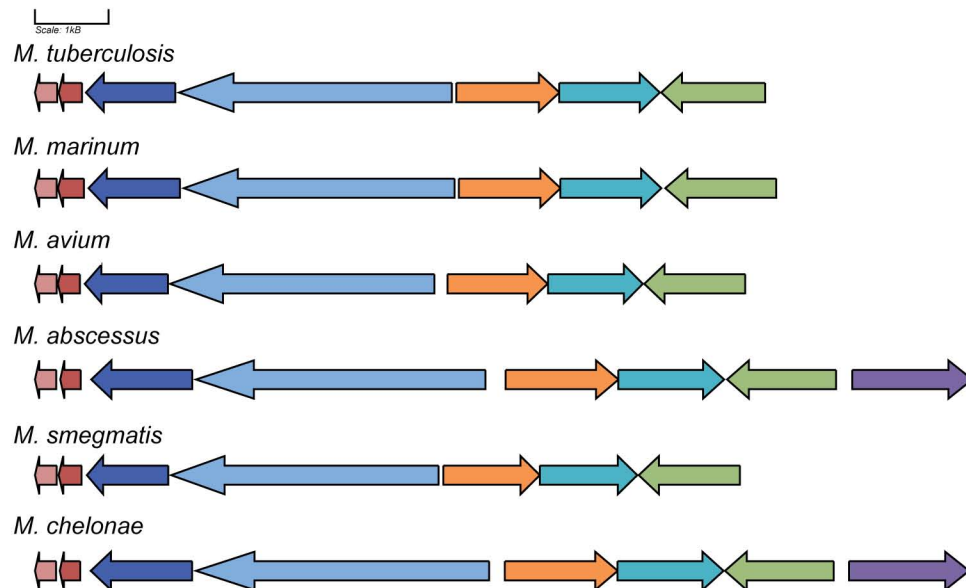


F



Sup. Figure 4

A



B

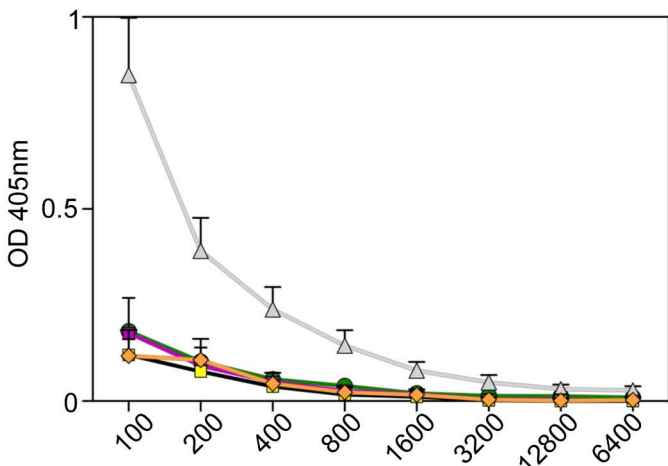
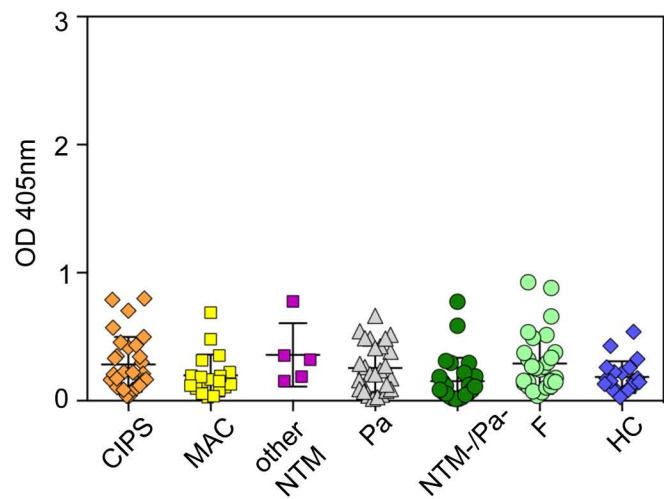
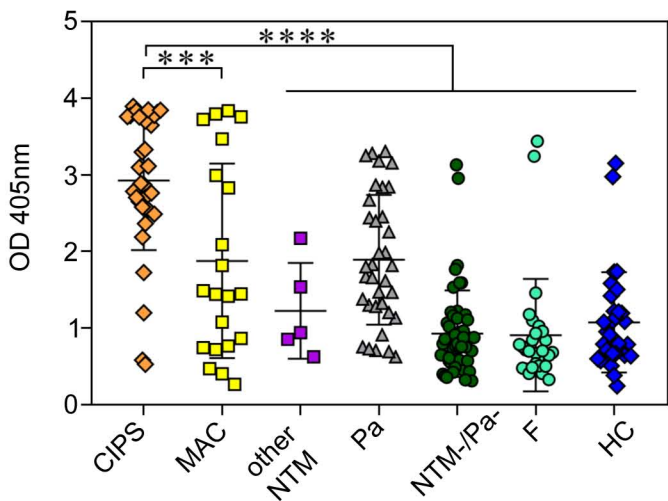
MAB_3753c/1-96	1	-MSQITYNHGEIDALVADVKGIIKFKFQAGLEELQHDIQPLVQQAEGQEATAYQEYQKAWHQSAEDL	65
BB28_19235/1-97	1	MTMQITYNHGEIDALVADVKGIIKFKFQAGLEELQHDIQPLVQQAEGQEATAYQEYQKAWHQSAEDL	66
MAB_3753c/1-96	66	NQILTLQLNAKVDQGNQDAQHTDQSAAGAWHH	96
BB28_19235/1-97	67	NQILTLQLNSKVDQGNQDAQHTDQSAAGAWHG	97

C

MAB_3754c/1-92	1	MAVFQNDLALLDSTAKKIDGKYQEF TAMQSQLRDRVAVGTSTWQGQARHAFDEAMARFDQEMGD	64
BB28_19240/1-92	1	MAVFQNDLALLDSTAKKIDGKYQEF TAMQSQLRDRVAVGTSTWQGQARHAFDEAMARFDQEMGD	64
MAB_3754c/1-92	65	IQKVLIGIHDTMESNKRRIQEMDESQTF	92
BB28_19240/1-92	65	IQKVLVGIHDTMESNKRRIQEMDEGQTF	92

D

MAB_3760/1-544	1	MTSSGAAPEQVRRATMPIQIMVLSQLLVA AVL I SLLFGWPWWAALLIAIVSLALVLGRWGGQTVRHVAVTGFKYLTGRTYRSSGS	86
BB28_19270/1-544	1	MTSSGAAPEQVRRATMPIQIMVLSQLLVA AVL I SLLFGWPWWAALLIAIVSLALVLGRWGGQTVRHVAVTGFKYLTGRTYRSSGS	86
MAB_3760/1-544	87	IADTPSGQDSWTGTRWENGKLI TVLEI SATQRHPSVITPDHCATDSL VPLRVLRECMHQNDIELESIDVINSNGORTAQGTVLRGGY	172
BB28_19270/1-544	87	IADTPSSQDSWTGTRWENGKLI TVLEV SATQRHPSVITPDHCATDSL VPLRVLRECMHQNDIELESIDVINSNGORTAQGTVLRGGY	172
MAB_3760/1-544	173	DRLVGPLPAVAIRTVVWVLRFDPGANVDAVFRGGGRRGAERCAVIA TARVRRALAAAHCASTILQAGQIHRISAEMLRQYAGAPM	258
BB28_19270/1-544	173	DRLVGPLPAVAIRTVVWVLRFDPGANVDAVFRGGGRRGAERCAVIA TARVRRALAAAHCASTILQAGQIHRISAEMLRQYAGAPM	258
MAB_3760/1-544	259	HE DW SGLT L I DSYNVAMG I DPWY I T DAT L TGLWARPTLET S V T V R L R P A A K G R T S V G A L V R W A T D E Q L P V R H S T G M T S L N G S Q R D A	344
BB28_19270/1-544	259	HE EW GGLML I DSYNVAMG I DPWY I T DAT L TGLWARPTLET T V T V R L R P A A K G R T S V G A L V R W A T D E Q L P I R H S T G M T S L N G S Q R D A	344
MAB_3760/1-544	345	FFAGLPVGAIGLEYLTRSIEMSNEELDGIHLPTSGCGQLIGSDMSGRAIATRLSGQGVTVVVRAELYVCQVVLRRAVIGALVVV	430
BB28_19270/1-544	345	FFAGLPVGAIGLEYLTRSIEMSNEELDGIHLPTSGCGQLIGSDMSGRAIARLSGQGVTVVVRAELYVCQVVLRRAVIGALVVV	430
MAB_3760/1-544	431	YTD R P H M W D V M V T S I G D A N R I Q F A S G P G F I D P R C T L A V V D G M Q P T A L P S N C T A L H I S S E Y D A L P A R P D V I I D Q P N G Y D H I L L T A	516
BB28_19270/1-544	431	YTD R P H M W D V M V T S I G D A N R I Q F A S G P G F I D P R C T L A V V D G M Q P T A L P S N C T A L H I S S E Y D A L P A R P D V I I D Q P N G Y D H I L L T A	516
MAB_3760/1-544	517	GGETIQVRLVTVSDelayLGRPIQAF AQ	544
BB28_19270/1-544	517	GGETIQVRLVTVSDelayLGRPIQAF AQ	544

Sup. Figure 5**A****B****C**

S1 Table. Primers used in this study

Number	5'→3' sequence	Restrictio site	F (sense), R (antisense)
Cloning in pUX1-<i>katG</i>			
1	gaga <u>TTAATTA</u> Atagtccgcgatgaa cgtccaggagt	Pacl	To clone the right arm of <i>esxUT</i> in pUX1- <i>katG</i> (F)
2	gaga <u>CAATTG</u> ATTCTGAAAAACAGCC ACttcagttcctccc	Mfel	To clone the right arm of <i>esxUT</i> in pUX1- <i>katG</i> (R)
3	gaga <u>CAATTG</u> cactgacccttccggtcaatctgtag	Mfel	To clone the left arm of <i>esxUT</i> in pUX1- <i>katG</i> (F)
4	gaga <u>GCTAGC</u> ttgatgagctgctggtgcacctgttc	Nhel	To clone the left arm of <i>esxUT</i> in pUX1- <i>katG</i> (R)
Primers to verify double homologous recombination			
5	ttgccaccttgaggatcgatcctg	-	To check double homologou recombination Δ <i>esxUT</i> (F)
6	tcaggtgggcgtaaagtccacgct	-	To check double homologou recombination Δ <i>esxUT</i> (R)
Primers to sequence modified <i>esxUT</i> locus			
7	catggagaaggagcctcaa	-	To sequence Δ <i>esxUT</i> locus (F)
8	attgatgatgatgacgaaatcgc	-	To sequence Δ <i>esxUT</i> locus (R)
Cloning in pMV306-KanR			
9	gaga <u>CAATTG</u> CCGTGGCTGTTTTTCAGAATGACCTGGC	Mfel	To clone the <i>esxU</i> ORF in pMV306 (F)
10	gaga <u>AAGCTT</u> CTAAGCGTAATCTGGAACATCGTATGGT G AAGTCTGGCTCTCGTCCATC	HindIII	To clone the <i>esxU</i> ORF in pMV306 (R)
11	gaga <u>CAATTG</u> CCATGAGCCAGATTACTTACAACCACGGC	Mfel	To clone the <i>esxT</i> ORF in pMV306 (F)
12	gaga <u>AAGCTT</u> CTAAGCGTAATCTGGAACATCGTATGG GTAGTGGTGCCAGGCGCCGG	HindIII	To clone the <i>esxT</i> ORF in pMV306 (R)
13	gaga <u>CAATTG</u> CCGTGGCTGTTTTTCAGAATGACCTGGC	Mfel	To clone the <i>esxUT</i> ORFs in pMV306 (F)
14	gaga <u>AAGCTT</u> CTAAGCGTAATCTGGAACATCGTATGG GTAGTGGTGCCAGGCGCCGG	HindIII	To clone the <i>esx4UT</i> ORFs in pMV306 (R)

Cloning in pVV16-KanR			
15	gaga CATATG GTGGCTGTTTTTCAGAATGACCTGGC	NdeI	To clone the <i>esxU</i> ORF in pVV16 (F)
16	gaga GGATCC GAAGGTCTGGCTCTCGTCCATC	BamHI	To clone the <i>esxU</i> ORF in pVV16 (R)
17	gaga CATATG ATGAGCCAGATTACTTACAACCACGGC	NdeI	To clone the <i>esxT</i> ORF in pVV16 (F)
18	gaga GGATCC GTGGTGCCAGGCGCCG	BamHI	To clone the <i>esxT</i> ORF in pVV16 (R)
19	gaga CATATG GTGGCTGTTTTTCAGAATGACCTGGC	NdeI	To clone the <i>esxUT</i> ORFs in pVV16 (F)
20	gaga GGATCC GTGGTGCCAGGCGCCG	BamHI	To clone the <i>esxUT</i> ORFs in pVV16 (R)
Cloning in pUC19 zeo			
21	gccaaagctgcatgcctgcagATCGATCCTGGGAGACCAG		Gibson_EsxU_Left_Fwd
22	gtgctctagAAAACAGCCACTTCAGTTCC		Gibson_EsxU_Left
23	tggtctgttCTAGAGCAACGAAAGGCTCAG		Gibson_Zeocine_Fwd
24	gtcagtggtGCGGAACGCTCAGTGGAAAC		Gibson_Zeocine_Rev
25	agcgttccgcCACCACTGACCCTTCCGG		Gibson_EsxT_Right_Fwd
26	agtgaattcgagctcgggtaccGATGAGCTGCTGGTGAC		Gibson_EsxT_Right_Rev
27	agtgaattcgagctcgggtaccGATGAGCTGCTGGTGAC		EsxUT_pMVH361_Fwd
28	taactacgtcgacatcgataTCAGTGGTGCCAGGCGC		EsxUT_pMVH361_Rev
Primer to check if the <i>esxUT</i> gene are replaced by zeocine			
29	GTCCGACGGTGATGAACCCGCTGCG		To check double homologous recombination Δ <i>esxUT</i> primer 1
30	GAGCACCGGAACGGCACTGGTCAAC		To check double homologous recombination Δ <i>esxUT</i> primer 2
31	GCATGACCAAATCCCTTAACGTGAGTTTTCG		To check double homologous recombination Δ <i>esxUT</i> primer 3
32	GTGGGCGTAAATGTCCACGCTGTC		To check double homologous recombination Δ <i>esxUT</i> primer 4
Cloning in pET6HN			
33	ATATCCAAGCTTGTGGCTGTTTTTCAGAATGACCT-	HindIII	pET6HN-MAB_3754-HindIII

34	CCGGAATTCGCGAAGGTCTGGCTCTCGTCC	EcoRI	pET6HN-MAB_3754-EcoRI
35	ATATCCAAGCTTATGAGCCAGATTACTTACAACCAC	HindIII	pET6HN-MAB_3753c-HindIII
36	CCGGAATTCGCGTGGTGCCAGGCGCCG	EcoRI	pET6HN-MAB_3753c-EcoRI
qPCR primers			
37	TACCAGGAGTTCACGGCGAT		qRT_MAB_3754_Fwd
38	CATCTCCTGGTCGAAGCGAG		qRT_MAB_3754_Rev
39	CGCTGGTTGCTGATGTCAAG		qRT_MAB_3753_Fwd
40	GTA CTCTGGTACGCAGTGG		qRT_MAB_3753_Rev
41	TCCGAGAAAGACAAGGCTTC		SigA_Fwd
42	CCAGCTCAACTTCCTCTTCG		SigA_Rev