

Table S5. Oligonucleotides used to create knockout vectors for allelic exchange mutagenesis

Gene(s)	Internal deletion size (bp)	Knockout vector	Oligonucleotide pairs used for vector construction	
			Sequence (5'-3') ^a	Region amplified
<i>M. tuberculosis</i>				
Rv0595c	325	p2Δ0595cKO	GCAGA <u>AAGCTT</u> ACGAAAAGATCGGGC GGCT <u>AGATCT</u> GTACGTTACCAACCGCAC	1990 bp amplicon containing 10 bp of the 5' end of Rv0595c and 1980 bp of upstream sequence
			GAGC <u>AGATCT</u> GACCCAAGACAACGACTA GTAT <u>GCGGCCGCCTAACCTT</u> CACCAGAC	2027 bp amplicon containing 58 bp of the 3' end of Rv0595c and 1969 bp of downstream sequence
Rv2545- Rv2550c	2415	p2Δ2545_50cKO	TGATA <u>AAGCTT</u> GATGACGATCTCGCGCAG TAAT <u>AGATCT</u> GAGATATATGCATTGGA	2052 bp amplicon containing 131 bp of the 5' end of Rv2545 and 1921 bp of upstream sequence
			ATGT <u>AGATCT</u> GCAGCCTTTTCACA TTAT <u>GGTACCAGGGACTAT</u> CAG	1776 bp amplicon containing 42 bp of the 3' end of Rv2550c and 1734 bp of downstream sequence
<i>M. smegmatis</i>				
MSMEG_1283- MSMEG_1284	378	p2ΔSM1283_84KO	TGATA <u>AAGCTT</u> GCTCATAACGGCCAGGC TAAT <u>AGATCT</u> CTGGCCAGCCGGTCCG	965 bp amplicon containing 23 bp of the 5' end of MSMEG1283 and 932 bp of upstream sequence
			CGCCG <u>AGATCT</u> GTGGTTGCATCGC TGAT <u>GGTACCGAGCAGTGGCTACT</u> GG	1099 bp amplicon containing 219 bp of the 3' end of MSMEG1284 and 880 bp of downstream sequence

a. Restriction sites used for cloning are underlined