| Name | Description | Source |
|-----------------------------|---|------------------|
| <u>Strains</u> | | |
| M. tuberculosis | | |
| H37Rv | ATCC 25618, virulent laboratory strain | Laboratory stock |
| H37Ra | ATCC25177, avirulent laboratory strain | Laboratory stock |
| $\Delta Rv0595c$ | Derivative of H37Rv carrying an unmarked deletion in the vapC gene Rv0595c | This work |
| $\Delta Rv2009$ - $Rv2010$ | Derivative of H37Rv carrying an unmarked deletion in the vapBC operon Rv2009-Rv2010 | This work |
| $\Delta Rv2545$ - $Rv2550c$ | Derivative of H37Rv carrying an unmarked deletion spanning from Rv2545 to Rv2550c | This work |
| M. smegmatis | | |
| $mc^2 155$ | <i>ept-1</i> , efficient plasmid transformation mutant of mc^26 | [1] |
| Δ <i>MSMEG1283-1284</i> | Derivative of mc ² 155 carrying an unmarked deletion in the <i>vapBC</i> operon <i>MSMEG_1283-MSMEG_1284</i> | This work |
| $mc^{2}4517$ | ept-1, efficient plasmid transformation mutant of mc ² 6, expression strain with T7 RNA polymerase | [2] |
| Yeast | | |
| AH109 | HIS3, ADE2, lacZ, MEL1 containing reporter strain for screening protein interactions, MATa, trp1- 901, leu2-3, 112, ura3-52, his3-200, gal4_, gal80_, LYS2::GAL1uas-GAL1TATA-HIS3, MEL1 GAL2uasGAL2TATA-ADE2, URA3::MEL1uas-MEL1TATA-lacZ | Clontech |
| <u>Plasmids</u> | | |
| p2NIL | <i>E. coli</i> cloning vector; Km ^R | [3] |
| pGEM3Z(+)f | <i>E. coli</i> cloning vector; Ap ^R | Promega |
| pGOAL17 | Plasmid carrying the <i>lacZ</i> and <i>sacB</i> genes as a <i>PacI</i> cassette; Ap^{R} | [3] |
| pGOAL19 | Plasmid carrying the <i>lacZ</i> , <i>sacB</i> and <i>hyg</i> genes as a <i>PacI</i> cassette; Ap^{R} | [3] |

Table S4. Strains and plasmids used in this study

| р2Δ0595сКО | Knockout vector for creating an unmarked deletion in <i>M. tuberculosis Rv0595c</i> ; Km ^R , Hyg ^R | This work |
|----------------|--|-----------|
| p2∆2009_10KO | Knockout vector for creating an unmarked deletion in <i>M. tuberculosis Rv2009-Rv2010</i> ; Km ^R , Hyg ^R | This work |
| p2Δ2545_50cKO | Knockout vector for creating a <i>hyg</i> -marked deletion in <i>M. tuberculosis</i> $Rv2545$ - $Rv2550c$; Km ^R , Hyg ^R | This work |
| p2∆SM1283_84KO | Knockout vector for creating an unmarked deletion in <i>M. smegmatis MSMEG_1283-MSMEG_1284</i> ; Km ^R , Hyg ^R | This work |
| pOLYG | <i>E. coli</i> -Mycobacterium shuttle vector; Hyg ^R | [4] |
| pSE100 | <i>E. coli</i> -Mycobacterium shuttle vector carrying $P_{myc1}tetO$; Hyg ^R | [5] |
| pSE0549c | pSE100 carrying $Rv0549c$ under control of $P_{myc1}tetO$; Hyg ^R | This work |
| pSE0627 | pSE100 carrying $Rv0627$ under control of $P_{myc1}tetO$; Hyg^{R} | This work |
| pSE1953 | pSE100 carrying $Rv1953$ under control of $P_{myc1}tetO$; Hyg^{R} | This work |
| pSE2010 | pSE100 carrying <i>Rv2010</i> under control of P _{myc1} tetO; Hyg ^R | This work |
| pSE2829c | pSE100 carrying $Rv2829c$ under control of $P_{myc1}tetO$; Hyg^{R} | This work |
| pSE2546 | pSE100 carrying $Rv2546$ under control of $P_{myc1}tetO$; Hyg ^R | This work |
| pSE2548 | pE100 carrying $Rv2548$ under control of $P_{myc1}tetO$; Hyg ^R | This work |
| pSE2549c | pSE100 carrying $Rv2549c$ under control of $P_{myc1}tetO$; Hyg^{R} | This work |
| pSE3320c | pSE100 carrying <i>Rv3320c</i> under control of P _{myc1} tetO; Hyg ^R | This work |
| pSE0550c_49c | pSE100 carrying <i>Rv0550c-Rv0549c</i> under control of P _{myc1} tetO; Hyg ^R | This work |
| pSE0596c_95c | pSE100 carrying <i>Rv0596c-Rv0595c</i> under control of P _{myc1} tetO; Hyg ^R | This work |
| pSE2550c_49c | pSE100 carrying <i>Rv2550c-Rv2549c</i> under control of P _{myc1} tetO; Hyg ^R | This work |
| pSE2830c_29c | pSE100 carrying <i>Rv2830c-Rv2829c</i> under control of P _{myc1} tetO; Hyg ^R | This work |
| pSESM1284 | pSE100 carrying MSMEG_1284 under control of P _{myc1} tetO; Hyg ^R | This work |
| pSE2546_FLAG | pSE100 carrying a C-terminally FLAG-tagged Rv2546 under control of P _{myc1} tetO; Hyg ^R | This work |
| pSE2549c_FLAG | pSE100 carrying a C-terminally FLAG-tagged Rv2549c under control of P _{myc1} tetO; Hyg ^R | This work |
| pMC1s | L5-based integration vector carrying P_{smyc} - <i>tetR</i> ; Km ^R | [5] |
| | | |

| pMC1r | Derivative of pMC1s; Km ^R | This work |
|---------------|--|------------------------------|
| pMC2m | L5-based integration vector carrying P_{imyc} -tetR; Km ^R | D. Schnappinger & S. Ehrt |
| pTTP1B | Tweety-based integration vector; Ap ^R ; Km ^R | [6] |
| pTTP1BG | Derivative of pTTP1B with Km ^R marker replaced by Gm ^R marker from pML10 [7]; Gm ^R | This work |
| pTTP1BGs | Derivative of pTTP1BG carrying P _{smyc} - <i>tetR</i> from pMC1s; Gm ^R | This work |
| pTT2549c | pTTP1BGs derivative carrying P _{myc1} tetO::Rv2549c; Gm ^R | This work |
| pTT0595c | pTTP1BGs derivative carrying P _{myc1} tetO::Rv0595c; Gm ^R | This work |
| pOLYGaa | pOLYG derivative carrying <i>M. smegmatis</i> acetamidase promoter (P _{ami}) from pAGAN11 [8]; Hyg ^R | D. Warner |
| pMAP | Derivative of pMC1r carrying P _{ami} cloned as a 4.4-kb <i>PvuII/Cla</i> I fragment from pOLYGaa; Km ^R | This work |
| pMAP2550c | Integration vector carrying P _{ami} :: <i>Rv2550c</i> ; Km ^R | This work |
| pMAP2830c | Integration vector carrying P _{ami} :: <i>Rv2830c</i> ; Km ^R | This work |
| pMAP0596c | Integration vector carrying P _{ami} :: <i>Rv0596c</i> ; Km ^R | This work |
| pGADT7 | Y2H vector to produce AD fusions, GAL4(768-881)AD, LEU2, HA epitope tag; Ap ^R | Clontech |
| pGAD0595c | pGADT7 containing Rv0595c fused to GAL4 AD | This work |
| pGAD0596c | pGADT7 containing Rv0596c fused to GAL4 AD | This work |
| pGAD2549c | pGADT7 containing Rv2549c fused to GAL4 AD | This work |
| pGAD2550c | pGADT7 containing Rv2550c fused to GAL4 AD | This work |
| pGAD2830c | pGADT7 containing Rv2830c fused to GAL4 AD | This work |
| pGBKT7 | Y2H vector to produce BD fusions, GAL4(1-147)DNA-BD, TRP1, c-MYC epitope tag; Km ^R | Clontech |
| pGBK0595c | pGBKT7 containing <i>Rv0595c</i> fused to <i>GAL4</i> BD | This work |
| pGBK0596c | pGBKT7 containing Rv0596c fused to GAL4 BD | This work |
| pGBK2549c | pGBKT7 containing Rv2549c fused to GAL4 BD | This work |
| pYUB28b | E. coli-Mycobacterium shuttle vector with T7 promoter and C-terminal His-tag; Hyg ^R | [9] |
| pYUBRv0065A-5 | pYUB28b derivative carrying the Rv0065A-Rv0065 operon for protein expression; Hyg ^R | This work |
| pYUBRv0616A-6 | pYUB28b derivative carrying the Rv0616A-Rv0617 operon for protein expression; Hyg ^R | This work |

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