EsxA membrane-permeabilizing activity plays a key role in mycobacterial cytosolic translocation and virulence: effects of single mutation at glutamine 5

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Table S1. The p-values calculated by Log-Rank analysis of zebra fish embryos survival (*	p <0.05).
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	Mm	Mm	Mm	Mm	Mm
		ΔEsxA/B	ΔEsxA/B(WT)	ΔEsxA/B(Q5K)	ΔEsxA/B(Q5V)
Mm	-	0.00018*	0.178	0.00114*	0.489
Mm		-	0.013*	0.585	1.7e-05*
ΔEsxA/B					
Mm			-	0.0496*	0.0463*
∆EsxA/B(WT)					
Mm				-	0.000127*
ΔEsxA/B(Q5K)					
Mm					-
ΔEsxA/B(Q5V)					

Table S2: Sequences of the primers for esxA/esxB knockout in Mm and for PCR confirmation

Primers	Forward
KO-Left Arm-F (P _{LA} 1)	5'-GAATTCCTGCAGGATGCCTTCAACGAGATGGATTAC-3'
KO-Left Arm-R ($P_{LA}2$)	5'-CTCTAGAGGATCCCCCGGCATCGGTCTTCATCTCT-3'
KO-Right Arm-F $(P_{RA}1)$	5'-CTCTAGAGGATCCCCGAACCTGGCTCGCACCATC-3'
KO-Right Arm-R (P _{RA} 2)	5'-GACTAGAGGGTACCCTTCTTCTGCTATGTCCTGTTG -3'
PCR-Left Arm-F-1	5'-TAGAAGTCAGCGACGGCGCAGCCAG-3'
PCR-Left Arm-R-1	5'- CGTGGTCGAGGCGATGGCGGAG-3'
PCR-Left Arm-F-2	5'-TCGAACTGTACCGGATTTCG-3'

PCR-Right Arm-R-2	5'-GCTGATGGGCTTCGGTTG-3'
PCR-P _{CE1}	5'-CAGCATGGCAGAGATGAAGA-3'
PCR-P _{CE2}	5'-CGGTATTCTACGCGAACGAG-3'

Supplemental Figure 1



Figure S1. Time course of cytosolic translocation of Mtb and MtbΔEsxA/B in THP-1 cells measured by a fluorometer. THP-1 cells were infected with Mtb or MtbΔEsxA/B at MOI of 2. At the 7, 8 and 9 days of post-infection, the cells were loaded with CCF4-AM for 2 h. The cells were washed, detached and fixed by PFA. Blue (450nm)/green (530) ratio was measured in the fluorometer with excitation at 409 nm. The data is

presented as means \pm S.E.