

Supplementary Figure 1: Representation of the gene content of the genomic locus harboring the gene encoding phospholipase C (*MAB_0555*) of *M. abscessus* (top panel) and comparisons shown with other *mycobacteria* (bottom panel).

Supplementary Figure 2: **(A)** Time-course hydrolysis of p-NPPC by phospholipases C. rMabs-PLC (50 µg) was incubated at 37°C with 5 mM of p-NPPC in 1 mL (final volume) of Tris-HCl (pH 7.2) buffer with 5 mM CaCl₂ and 1.5% sorbitol. The release of p-nitrophenol was monitored at various times at 410 nm. Fifteen microgram of each PLC from *B. cereus* and *C. perfringens* (Sigma) were used as positive control in similar conditions. **(B)** Temperature effect on rMabs-PLC activity. The optimal temperature was determined by incubating the reaction mixture at different temperatures from 25°C to 55°C in 10 mM Tris-HCl buffer pH 7.2. **(C)** Effect of pH on rMabs-PLC. The optimal pH was determined by measuring the activity of rPLC at 37°C for 48 h incubation between pH 4 and pH 10 using 10 mM sodium acetate (pH 4-5); 10 mM Mes (pH 6-6.5); 10 mM Tris/HCl (pH 7.8) and 10 mM glycine (pH 9.0) and 10 mM sodium bicarbonate (pH 10) using p-NPPC as substrate.

Supplementary Table 1: List of Primers used in this study.

Primer name	Primer sequence	Restriction site
	Construction of KO PLC mutant	
PLCF1	5'-CGGTCGTAGCCTGAAGCGATG-3'	
PLCR1	5'-ACGGCATGAAATGGAAGCCAT-3'	
KMlIF1	5'- <u>CGACGCGTTAAAACGACGGCCAGTGAAT</u> -3'	<i>Mlu</i> I
KMlIR1	5'- <u>CGACGCGTCAGGAAACAGCTATGACCATGA</u> -3'	<i>Mlu</i> I
PLCF2	5'- <u>ATGCCATGGT</u> CAGCCGCTGACCGCCGC-3'	<i>Nco</i> I
PLCR2	5'-AACAT <u>GAAGCTTGGCGCGTGGGTACCGC</u> -3'	<i>Hind</i> III
HyPLCF1	5'- <u>CAGCTGACCTCGAACTCCAACAGGTGA</u> -3'	<i>Pvu</i> II
HyPLCR1	5'- <u>GTTAACACGGCATGAAATGGAAGCCAT</u> -3'	<i>Hpa</i> I
ZeoF1	5'-CCC <u>AAGCTTCAGGCATCAAATAAACGAAA</u> -3'	<i>Hind</i> III
ZeoR1	5'-CC <u>ATCGATTCA</u> GTCTGCTCCTCGGCCAC-3'	<i>Clal</i>
RHF1	5'-ATCTGATGCACGACGAATCA-3'	
RHR1	5'-CGTAGGTGGTCCAGCCATAG-3'	
0555F	5'-CCCGCGGTGT <u>CCTGGCTCATC</u> -3'	
0555R	5'-TGATACCGCCAGCGCCGCAGA-3' Quantitative PCR	
PLCRTF	5'CATTACTCGGCACCCTGTC3'	
PLCRTR	5'ATGAGCAGCCATCCAGTTGT3'	