Supporting Information

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Fig. S1. Schematic structure of ManLAM, PIM₅ and PIM₆. A representation of the structure of ManLAM shows the mannose-capped nonreducing termini, the mannan core, and the phosphatidylinositolmannoside anchor. Although LAM could have a few arabinan chains (not confirmed with data), only 1 chain is shown for simplicity, and not all arabinan termini are capped. The mannan core is characterized by an $\alpha(1\rightarrow 6)$ -linked mannan, substituted at C2 by $\alpha(1\rightarrow 2)$ Manp residues. ManLAM is characterized by mono-, di, and tri-Manp caps. PIM₅ and PIM₆ contain $\alpha(1\rightarrow 2)$ Manp residues and mono- and di- Manp cap like structures, respectively.



Fig. 52. MALDI-TOF/MS analysis of purified ManLAM from *M. tuberculosis* H37Rv and H37Rv Δ *Rv*2181. Shown is negative mode MS analysis of WT ManLAM (A) and mutant ManLAM (B).



Fig. S3. Comparative partial two-dimensional NMR spectra of LM and ManLAM variants. The NMR ¹H ¹³C spectra of LM isolated from *M. tuberculosis* H37Rv (*A*) and H37Rv Δ *Rv2181* (*B*) were acquired in D₂O. (*C* and *D*) Two-dimensional ¹H ¹³C HSQC spectra of purified ManLAM from H37Rv Δ *Rv2181* (*C*) and *M. tuberculosis* (*D*). Only the expanded anomeric regions are shown. The peaks annotated with a question mark have not been assigned. MTX indicates a cross-peak characteristic of 5-deoxy-5-methyl-5-thio- α -xylofuranose.



Fig. 54. Analysis of LM/LAM from *M. smegmatis* Δ*MSMEG_4247*/pVV16 (lanes1), Δ*MSMEG_4247*/pVV-*Rv2181* (lanes 2), *MSMEG_4247*/pVV-*Rv1635c* (lanes 3), and *MSMEG_4247*/pVV-*Rv2181*-*Rv1635c* (lanes 4). LM/LAM was extracted from cells of recombinant strains by the phenol procedure, separated on a 10–20% Tricine gel, and visualized by PAS staining (A) and Western blot with lectin Con A (B).