Supplemental data

Fig. S1. Experimental scheme for the DNA vaccination. Each animal was i.m. injected following a prime/boost scheme on day 0, 21 and 42. On day 56, mice were aerosolically challenged with *M. abscessus.* On day 57, 63, 70 and 77, mice were sacrificed, blood samples were collected and lungs, spleen and liver *M. abscessus* CFU were counted.

Fig. S2. Phylogenetic analysis of *M. abscessus* MgtC-like proteins. Phylogenetic tree includes MgtC proteins from opportunist bacterial pathogens found in CF patients (*P. aeruginosa*, *B. cenocepacia*, *M. avium*, *S. maltophilia*, *A. fumigatus*) as well as other bacterial pathogens for which MgtC role in virulence has been studied (*S.* Typhimurium, *M. tuberculosis*, *Brucella* species, *Y. pestis*). Phylogenetic analyses were carried out with the "Phylogeny.fr" web server (http://www.phylogeny.fr) using MUSCLE, PhyML and TreeDyn softwares. *Escherichia coli* YhiD is a distantly related MgtC-like protein used to root the tree.

Fig. S3. Complementation of the growth defect of *ΔmgtC*$_{MAB}$ in Mg^{2+} deprived liquid medium. Growth curves of wild-type, *ΔmgtC*$_{MAB}$ and *ΔmgtC*$_{MAB}$ + *mgtC* strains grown in Sauton's medium without magnesium (left panel) or in Sauton's medium supplemented with magnesium (right panel). OD_{600} is indicated over the growth period. The experiment was independently repeated three times.

Fig. S4. Replication of *M. abscessus* wild-type strain and *mgtC* mutant in THP1 macrophages cell line after 48 h. Results are expressed as means + SD from three independent experiments.

The difference of replication rate between the two strains is not significant (P=0.25, Student's test).

Fig. S5. Specificity of antibodies present in serum of mice immunized with *mgtC*$_{MAB}$ DNA. Sera were tested in western-blot against a total lysate of *M. smegmatis* overexpressing the recombinant MgtC$_{MAB}$ protein tagged with a C-terminal poly-Histidine (lanes M) or another *M. abscessus* recombinant protein His-tagged, MAB_2545c (lanes N). Control blot was carried out with an anti-poly-Histidine antibody (Sigma), lane His; Blotting results of sera diluted at 1:1000 from three mice (M1, M2 and M3); Molecular standard, lane S. Mice sera revealed a single band, co-migrating with the band revealed by anti-His antibody, thus indicating that mice sera recognized specifically the His-tagged MgtC$_{MAB}$ antigen.

Table S1. Primer list

Primer name	Primer sequence
MAB3593-Sph-F	5’- ACATGCATGCGCCATTGGGATGTCGTC -3’
MAB3593-Hind-R	5’- CCCAAGCTTTTACCATTCGCTGGCTTCG -3’
MAB0146-Sph-F	5’- ACATGCATGCCACTTGCGGATTCTGTG -3’
MAB0146-Hind-R	5’- CCCAAGCTTCTACTCGATATCGTCGCCAAG -3’
MAB3593-W227A-F	5’- GTCTCGTCGGTGCGCGCGCATATCGATCGCGAG -3’
MAB3593-W227A-R	5’- CTCGCGATCGATATGCGCGCGCACCGACGAGAC -3’
AM-MgtC F	5’- ACGGCGTTGATCCAGATATT -3’
AM-MgtC R	5’- TTTCGACTGAGCCTTTCGTTGCCGAATCCGAGTGCTATG -3’
AV-MgtC F	5’- AGTTTTCGTTCCACTGAGCGCCGCGTATCTCTTCGAGGT -3’
AV-MgtC R	5’- GCACGCTGACAGAGAAGTTG -3’
Zeo-MgtC F	5’- CATAGCACTCGGATTCGGCAACGAAAGGCTCAGTCGAAA -3’
Zeo-MgtC R	5’- ACCTCGAAGAGATACGCGGCGCTCAGTGGAACGAAAACT -3’
screening 7	5’- CATAGCACTCGGATTCGGC -3’
screening 8	5’- ACCTCGAAGAGATACGCGG -3’
MgtC-reg-PvuII	5’- GTGCA CAGCTG CCCGAATGTCCGACGATGTA -3’
MgtC-reg-HindIII	5’- ATGGG AAGCTT TTACCATTCGCTGGCTTCGAC– 3’