

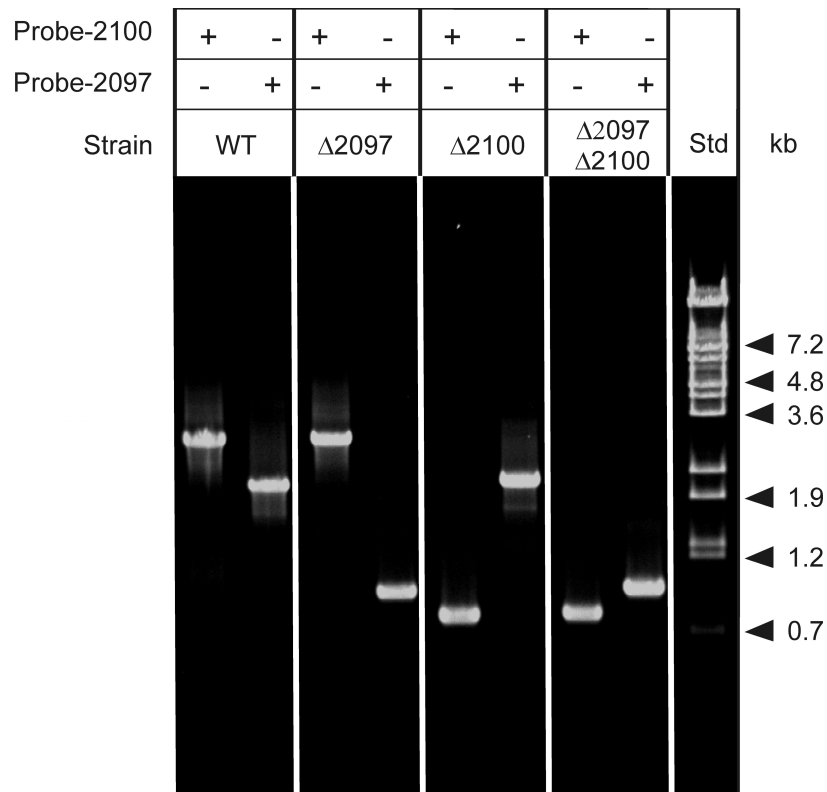
SUPPLEMENTARY MATERIAL

Supplementary Table S1: Primers used for plasmid and strain construction. Restriction sites are in bold, and overhangs required for hybridisation in the second PCR in italics. Primers are given in their 5' to 3' direction.

Primer	Sequences
<i>A_{mptC}</i>	CGTT AAGCTT TGTGTGCTAACTGCGTAATACTCGCGAC
<i>B_{mptC}</i>	<i>CCCATCCACTAAACTTAAACATCACCGTGCATGAAAAATAGTGTATCCG</i>
<i>C_{mptC}</i>	<i>TGTTTAAGTTTAGTGGATGGGTGGTTACTGCCGTATTTAGTTGTG</i>
<i>D_{mptC}</i>	GTTT GGATCC ATAACCGTAGTCAAATGCAGAGTCTTGAGC
<i>E_{mptC}</i>	CATTTGTGCCAACAACACGTGGTCATATGCG
<i>F_{mptC}</i>	GCGGATGCGATCACCGTCAACGC
<i>A_{mptD}</i>	CGCT TCTAGAGGATC ATTCCACAATTCCTACCCTCAG
<i>B_{mptD}</i>	<i>CCCATCCACTAAACTTAAACAATGTGCTGTCTGGGGGTTC</i>
<i>C_{mptD}</i>	<i>TGTTTAAGTTTAGTGGATGGGTGGGTCTTGGGCTGAAAG</i>
<i>D_{mptD}</i>	GCGG GAATTCCAGGCGGAATCC CACATTGAGTTC
<i>E_{mptD}</i>	GTGATCATCGCAGTCGCAGTTGCTGC
<i>F_{mptD}</i>	GCATGGGCCACGACAAGGTTCGC
Ex-2100 RBS for	GATTAT GTCGACAAGGAGATATAGATGATGCACGGT GAAAACTTG TGG
Ex-2100 rev	GATATT GAATTC TCAAATGAGCGCTTTGTTTTGATTG
Ex-2097 RBS for	GATTAT GTCGACAAGGAGATATAGATGTTGACGAACCC CAGACAG C
Ex-2097 rev	GATATT GAATTC CCTATTTGGATATTTTGGGAAATGC
Ex-Rv2181- for	GTGCTGCAGAAGGAGATATAGATATGAATTCGCCCTTGGTGGTCCG G
Ex-Rv2181- rev	CACGGATCCTTAGACGGTCACGGTCAGGCTG

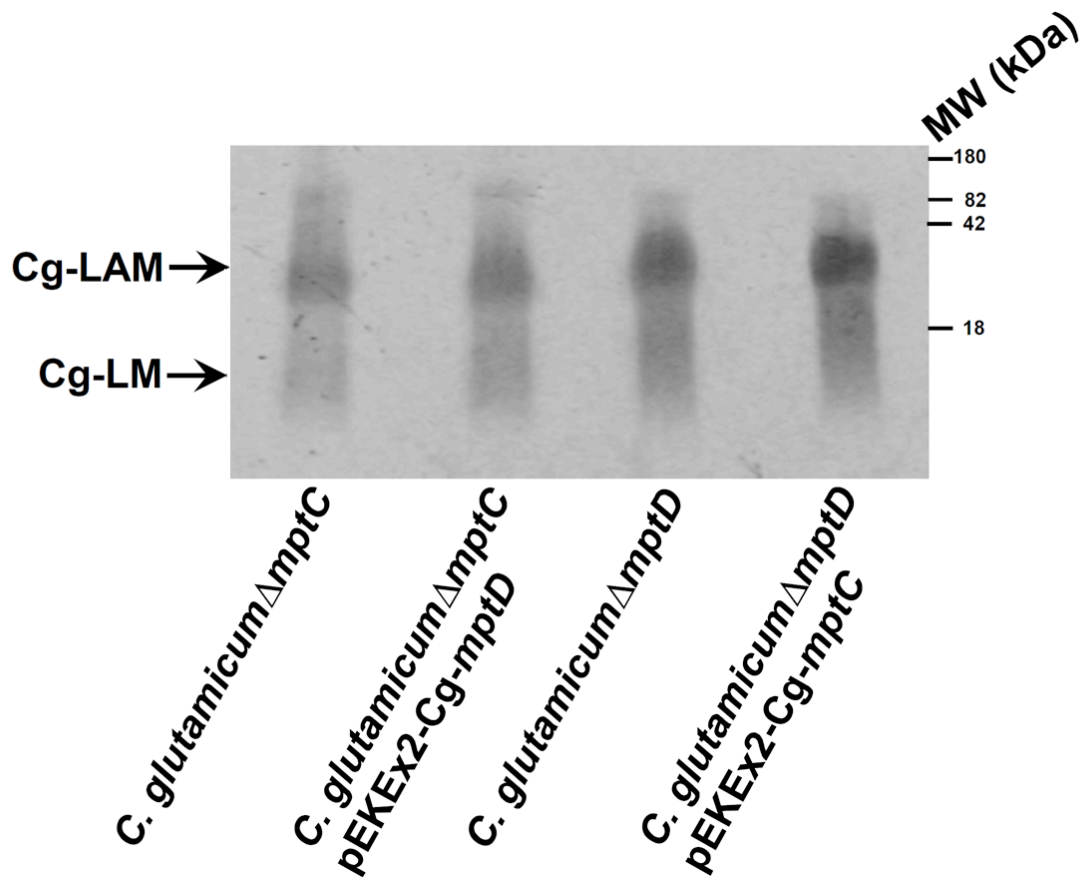
SUPPLEMENTARY MATERIAL

Supplementary Figure S1: Characterization of chromosomal deletions in three different *C. glutamicum* mutants. Chromosomal DNA was prepared from the wild-type (WT), the deletion mutant *C. glutamicum* $\Delta mptD$ (D2097), the deletion mutant *C. glutamicum* $\Delta mptC$ (D2100), and the double deletion mutant *C. glutamicum* $\Delta mptC \Delta mptD$ (D2100D20970). This served as template for PCR amplifications with primers specific for *mptC* (Probe-2100), and *mptD* (Probe-2097). A “+” denotes amplification with the specific primer pair. On the right standards are given with their size in kb.



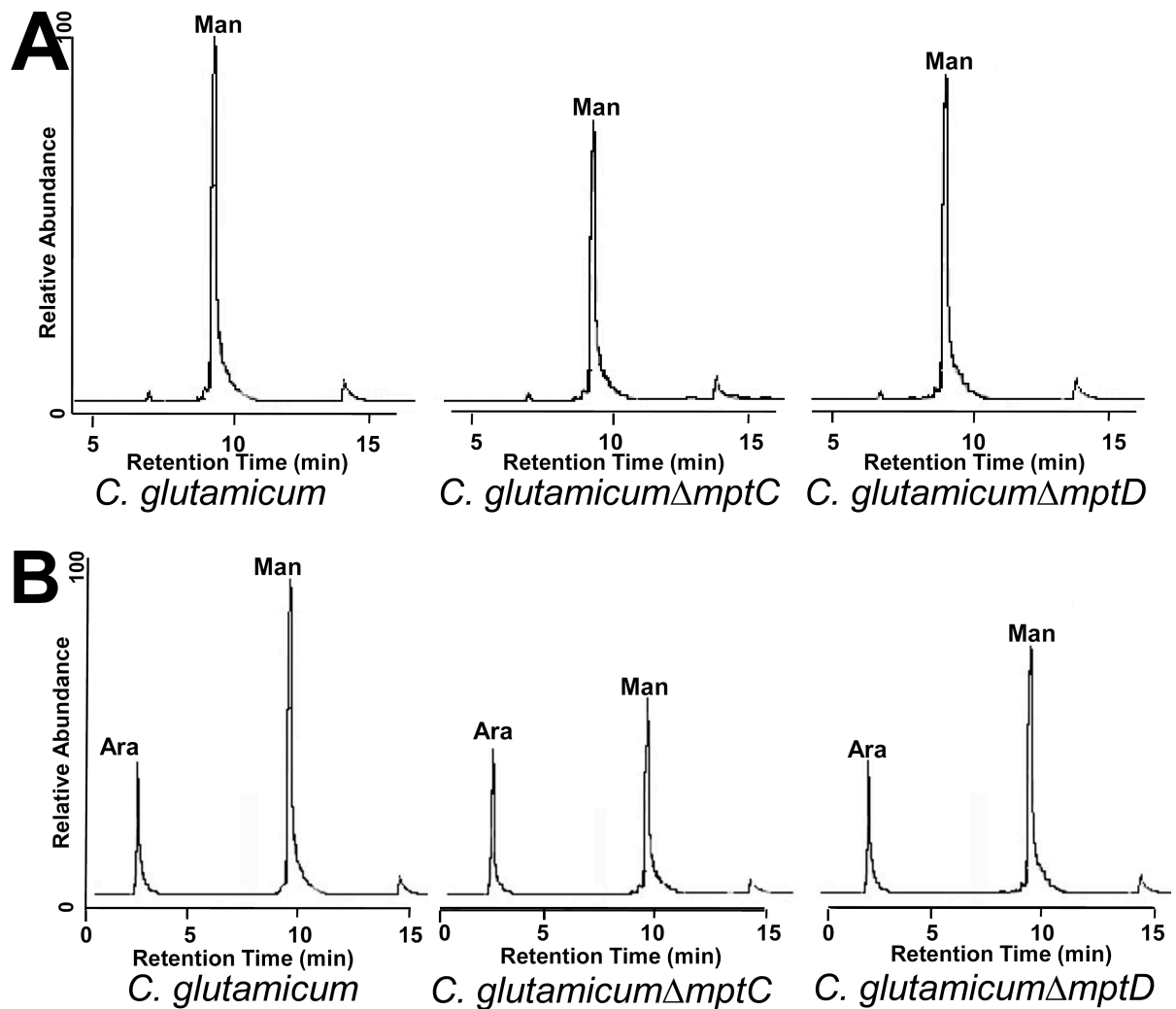
SUPPLEMENTARY MATERIAL

Supplementary Figure S2: Lipoglycan profiles of mutant and complemented strains of *C. glutamicum*. Lipoglycans were extracted from *C. glutamicum* Δ mptC, *C. glutamicum* Δ mptC pEKEX2-Cg-mptD, *C. glutamicum* Δ mptD, *C. glutamicum* Δ mptD pEKEX2-Cg-mptC, and analyzed using SDS-PAGE and visualized using a Pro-Q emerald glycoprotein stain (Invitrogen). Lipoglycan profiles are represented with standard molecular weight markers of glycoproteins of 180, 82, 42, and 18 kDa.



SUPPLEMENTARY MATERIAL

Supplementary Figure S3: Glycosyl compositional analysis of purified Cg-LM (A) and Cg-LAM (B) from different strains of *C. glutamicum*. Samples of individually purified lipoglycans were hydrolysed with 2M TFA, reduced and per-*O*-acetylated and subjected to GC analysis.



SUPPLEMENTARY MATERIAL

Supplementary Figure S4: GC-MS of per-*O*-methylated alditol acetate derivatives from Cg-LAM purified from *C. glutamicum*Δ*mptC*Δ*mptD* (A) and the identification of 2,3,4-tri-*O*-CH₃-1,5,6-tri-*O*-COCH₃-mannitol with the characteristic fragment alditol cleavage ions *m/z* 102, 118, 129, 162, 189, and 233 (B). GC-MS peaks marked (*) are non-carbohydrate impurities from the per-*O*-methylation and alditol acetate preparation procedures.

