

Supplementary data to the manuscript:

"Maltose uptake by the novel ABC-transport system MusEFGK₂I causes increased expression of *ptsG* in *Corynebacterium glutamicum*"

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The supplementary data comprise one table (Table S1) and four figures (FIG. S1, FIG. S2, FIG. S3, FIG. S4).

TABLE S1. Oligonucleotides used in this study.

Name	Sequence (5'-3')	Purpose, restriction site
DelMus_1	<u>GGGGATCCAACCGCGAAGTGTGCTGCGAAGCTCCGATCCGATGCTGAG</u>	Deletion of <i>mus</i> genes, BamHI
DelMus_2	<u>CTTATAAAATTGGAGTGTGAAGGTTATTGCGTGCAGACGATCCGATGCTGAG</u>	Deletion of <i>mus</i> genes
DelMus_3	<u>CACGCAATAACCTTCACACTCCAATTATAAGACGACTACCTGCTGCCATAC</u>	Deletion of <i>mus</i> genes
DelMus_4	<u>GGGGATCCTGCCTTGGCAATGA</u>	Deletion of <i>mus</i> genes, BamHI
ΔMusConF	TAGATGGCGCACAGTGACTC	Verification of <i>mus</i> deletion
ΔMusConR	TAACTACCGCAACACCGATG	Verification of <i>mus</i> deletion
DelPtsG_1	<u>GTCGACGGGCATAATTCTGACAGTGTG</u>	Deletion of <i>ptsG</i> , SalI
DelPtsG_2	<u>CTTATAAAATTGGAGTGTGAAGGTTATTGCGTGGACGCCAAGAACTGATGGG</u>	Deletion of <i>ptsG</i>
DelPtsG_3	<u>CACGCAATAACCTTCACACTCCAATTATAAGCCGCTGACTTCATTGATCC</u>	Deletion of <i>ptsG</i>
DelPtsG_4	<u>GGATCCTAAGGACGCCATGTCAAACC</u>	Deletion of <i>ptsG</i> , BamHI
ΔPtsGConF	TCGTACGGTGTGGTTAAG	Verification of <i>ptsG</i> deletion
ΔPtsGConR	AGTATGCACCGCGTAATC	Verification of <i>ptsG</i> deletion
MusFor	<u>TCTAGATGGCGCACAGTGACTCACTT</u>	Cloning of pXMJ19-musEFGK and pXMJ19-musEFGKI, XbaI
MusRev	<u>ACCGGTCGAGTATGCGATTCTCATGGT</u>	Cloning of pXMJ19-musEFGK, AgeI
MusLoRev	<u>GGGGATCCATGACGTGGATACCACTACC</u>	Cloning of pXMJ19-musEFGKI and pXMJ19-cg2701, BamHI
MusI-For	<u>GGGGATCCTCTCCACGCAGAGGCACAT</u>	Cloning of pXMJ19-cg2701 and pXMJ19-cg2701-strep, BamHI
MusI-Rev	<u>GAGCTCTCAATTTTCGAACTGCGGGTGGCTCCAGCTGC</u> <u>CACCGCTACCGCTGCAAAGGGGCTATCGG</u>	Cloning of pXMJ19-cg2701-Strep, SacI
Cg2703_for	GCTTCTTGGAGGCCACATTG	RT-PCR, cg2703 RNA Probe, inactivation of cg2703
Cg2703_rev	<u>GGGCCCTAATACGACTCACTATAGGGTATCGCGGTTACCGTTGGAG</u>	RT-PCR, cg2703 RNA Probe, inactivation of cg2703

Cg2708_for	ACTGAAGATGCCGGCAAGT	RT-PCR, cg2708 RNA Probe, inactivation of cg2708
Cg2708_rev	<i>GGGCCCTAATACGACTCACTATA</i> AGGGATTATCCTCCGGCGTCATGG	RT-PCR, cg2708 RNA Probe, inactivation of cg2708
Cg2707_for	CCTATTCCGCCTATCTCGTC	RT-PCR, cg2707 RNA Probe, inactivation of cg2707
Cg2707_rev	<i>GGGCCCTAATACGACTCACTATA</i> AGGGCGATAGTCGGTTCGTATT	RT-PCR, cg2707 RNA Probe, inactivation of cg2707
Cg2701_for	TTCGCTGACCTAGTCATCGT	RT-PCR, cg2701 RNA Probe, inactivation of cg2701
Cg2701_rev	<i>GGGCCCTAATACGACTCACTATA</i> AGGGACTGCGAGGAAGAACAGGT	RT-PCR, cg2701 RNA Probe, inactivation of cg2701
PrmusF-for	<u>TCTAGAATGACA</u> ACTGGGCTGCTGAG	musF-Pr probe, cloning of pET2-PmusF, <i>Xba</i> I
PrmusF-TS-for	<u>TCTAGATCT</u> CCCTACCGCCTTCC	Cloning of pET2-PmusF-TS, <i>Xba</i> I
PrmusF-rev	<u>GGATCCTT</u> ATGGCGTTGGTGATAGTGGTG	musF-Pr probe, cloning of pET2-PmusF and pET2-PmusF-TS, <i>Bam</i> HI
ramBp3b_forw	ACCGCTGACGATGTGGCCCCGACCACGCCG	<i>ramBp3b</i> probe
ramBp3b_rev	ACTGAGGTGTTGCAA <u>ACTT</u> GTTGATTTCGCT	<i>ramBp3b</i> probe
RACE-cg2704-SP1	AAGTTGGTACCGCGGAGT	5'-RACE, cDNA synthesis
RACE-cg2704-SP2	AGGC _{GA} ATGTTGACTG	5'-RACE, nested primer
Oligonucleotide anchor primer	dT GACCACCGTATVGATGTCGACTTTTTTTTTTTTV	5'-RACE, amplification of dA-tailed cDNA

^a Restriction sites in the oligonucleotides are underlined, V represents an A, C, or G, linker sequences for cross over PCR are shown in bold, T7-promoter sequences for *in vitro* transcription are shown in italics, the sequences encoding the strep-tag and the linker in the oligonucleotide MusI-rev are shown in red and green, respectively.

FIG. S1. Maltose uptake of *C. glutamicum* WT, different concentrations (0.5 - 30 μM) of [^{14}C]-maltose were tested. Data represent mean values of three independent measurements from 2 independent cultivations and were fitted according to the Michaelis-Menten equation.

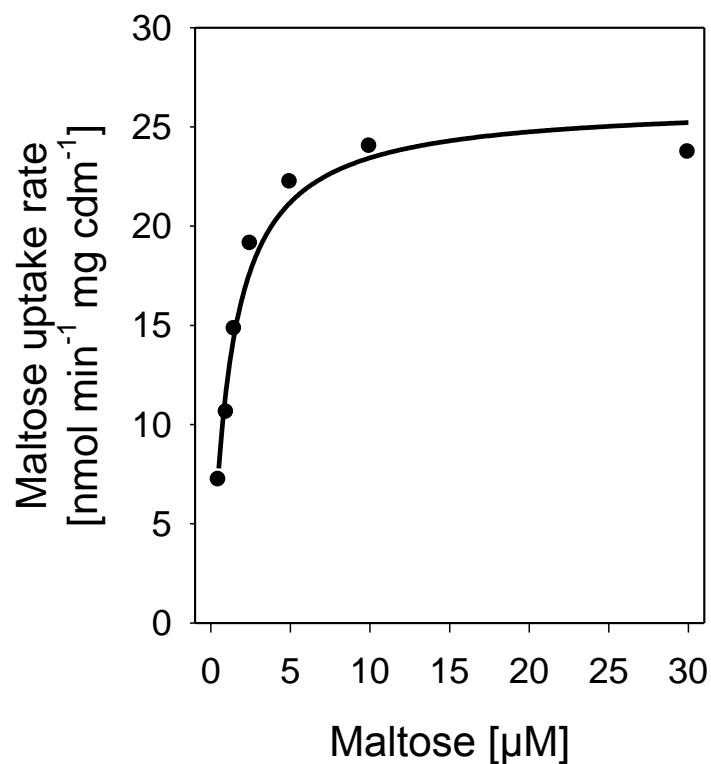


FIG. S2. Genomic locus of the intergenic region of the *C. glutamicum* *musE* and *musF* genes. Coding regions are shaded in grey and shown in italics, the *musE* Stop codon and the *musF* ATG start codon are underlined. The *musF* transcriptional start site (TS_{musF}) is shaded in black, -10 region is shown in bold. Overlapping putative RamA binding sites are shown in half boxes.

	RamA1	RamA2	
1	<i>gctaa</i> agctctat	tcccg tccccct	cgccacactcctccaatagcggag
	' <i>muse</i> '		
51	ggcgaaaaaggaggttaggtgagggttgtggcccgctcgcgagcttgc		
101	gcgagcagttattccaCATCCTcagcctc c tctcctcaccgcctccccg		
	-10	TS _{musF}	
151	ctgggaaaacgtgtggcaccacacctgaaattaaggttcaccacc ATGcaag		
	' <i>musF'</i> '		

FIG. S3. Alignment of *C. glutamicum* MusI with the hypothetical proteins from *B. longum* NCC2705, *S. agalactiae* 2603 and *S. pneumoniae* TIGR4, each contains 5 transmembrane regions, the DUF 624 motif (residues 30 to 101 in the amino acid sequence of SP_1677) and is encoded adjacent to an genes for an ABC transport system. Amino acids identical in three sequences are shaded in black and similar amino acids are shaded in grey.

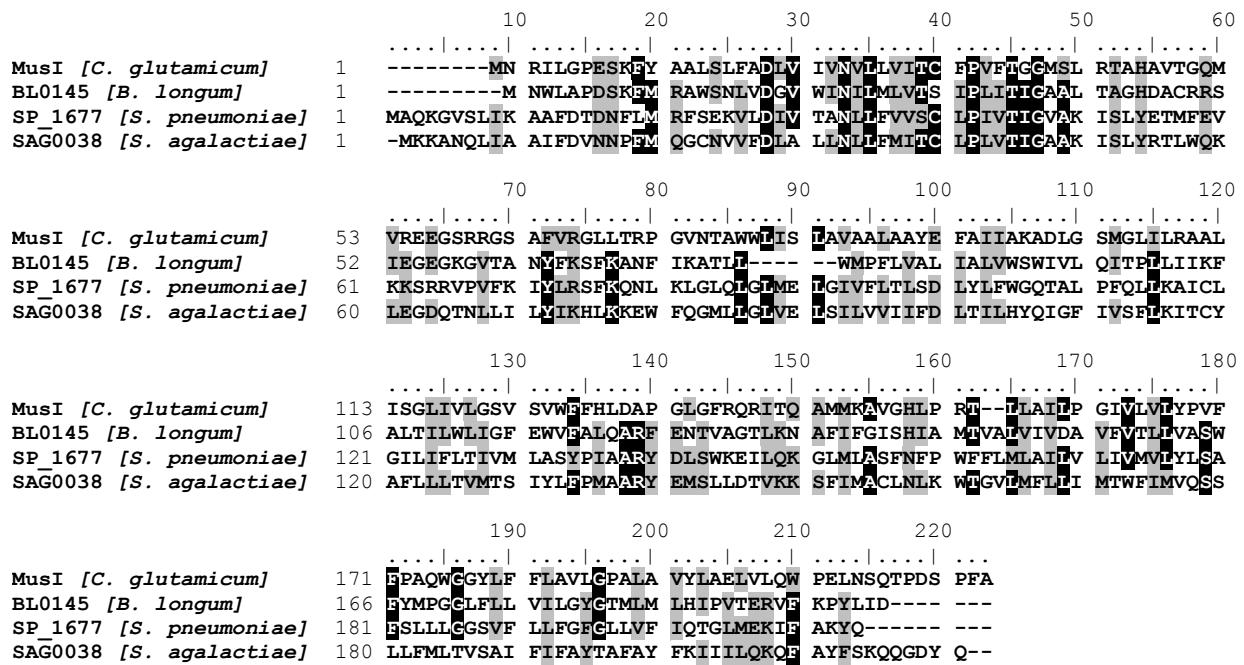


FIG. S4. Genomic organisation of gene clusters from *C. glutamicum* ATCC13032, *B. longum* NCC2705, *S. agalactiae* 2603 and *S. pneumoniae* TIGR4 encoding ABC-transporters. Genes encoding *musI*-like proteins are shown in red, genes for permeases in yellow, substrate binding proteins in green, and ATPase in blue. Genes encoding additional (hypothetical) proteins are coloured in grey.

