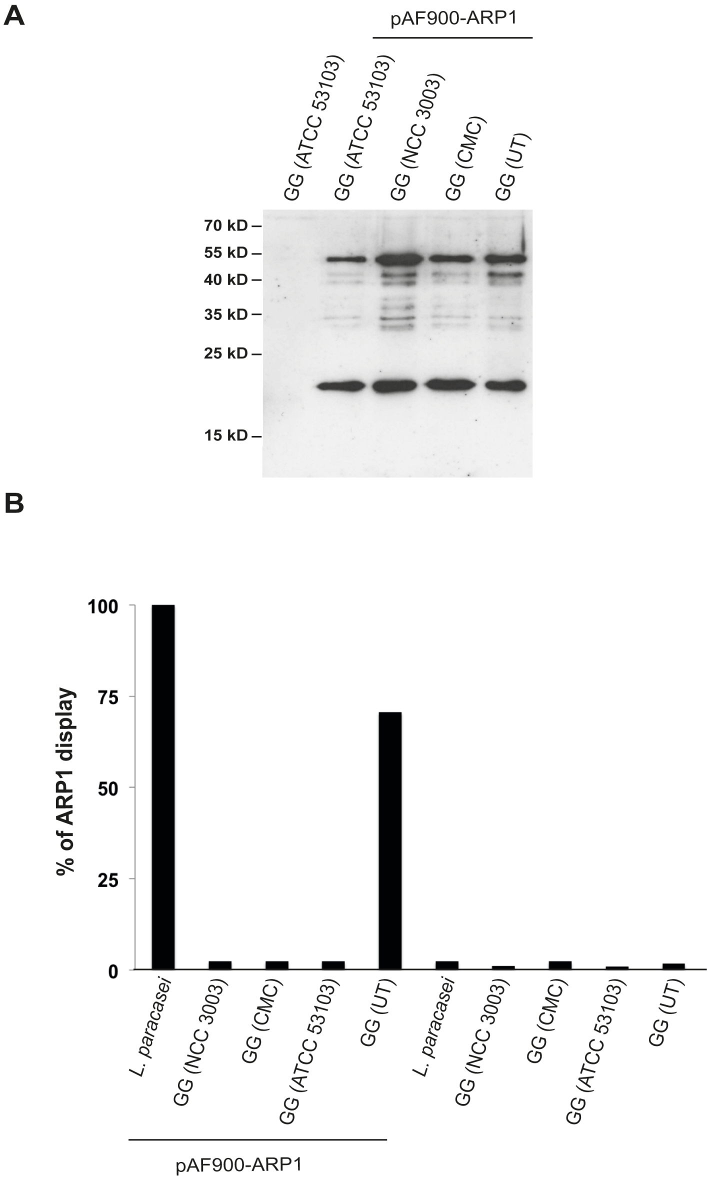

Fig S1. Production and display of ARP1 in the different GG strains with the plasmid pAF900-ARP1. (A) Production of ARP1 determined in culture cell extracts by Western blot using monoclonal mouse anti-E-tag antibodies and HRP labelled goat anti-mouse antibodies as primary and secondary antibodies, respectively. The signal was detected by chemiluminescence using the ECL Plus^{TM} Western blotting detection system (GE Healthcare). The theoretical molecular weight of the recombinant ARP1-PrtP protein is 39.7 kDa. (B) Display of ARP1 on the surface of the cells determined with a fluid based assay. The results are represented as percentage of display taking the display of ARP1 on the surface of *L. paracasei* pAF900-ARP1 as 100 %. Non-transformed Lactobacilli strains were used as negative controls.

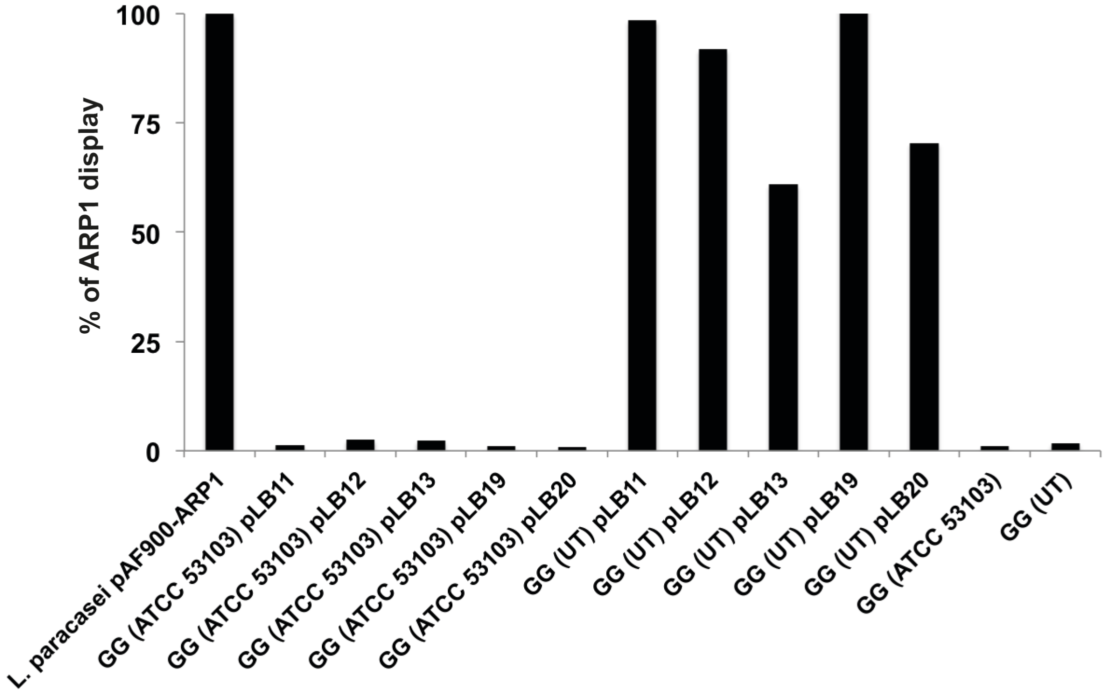

Fig S2. Display of ARP1 on the surface of the cells of the strains GG (ATCC 53103) and GG (UT) transformed with different constructs to produce ARP1 with different anchor domain lengths. The display was determined with a fluid based assay. The results are represented as percentage of display taking the display of ARP1 on the surface of *L. paracasei* pAF900-ARP1 as 100 %. Non-transformed GG strains were used as negative controls.

Table S1. Primers used for cloning purposes. The restriction sites added to the sequence are underlined.

Primer	Sequence (5'-3')	Purpose; relevant features
PrtPGG_450_Fw	CGCAGCTAGCAAGGCGGCAGCAGGTGAGACC	Cloning of 450 aa anchor domain; *Nhe*I site
PrtPGG_500_Fw	CGCAGCTAGCAAGAACGTCTTAACCGTTGCGGC	Cloning of 500 aa anchor domain; *Nhe*I site
PrtPGG_550_Fw	CGCAGCTAGCCGTATCACCGGAACAGCGACAG	Cloning of 550 aa anchor domain; *Nhe*I site
PrtPGG865etag_F	AAGCGCGGCCGCAGGTGCGCCGGTGCCGTATCCGGATCCGCTGGAACCGCGTGCCGCAGCTAGCGCTTATGATCTGATTATTAATGGTGGTGG	Cloning of 865 aa anchor domain; *Not*I site, E-tag-coding sequence, *Nhe*I site
PrtPGG1275etag_F	AAGCGCGGCCGCAGGTGCGCCGGTGCCGTATCCGGATCCGCTGGAACCGCGTGCCGCAGCTAGCTTGGAGAAGAATCCATCAACGGTTG	Cloning of 1275 aa anchor domain; *Not*I site, E-tag-coding sequence, *Nhe*I site
PrPGG_Rv	GTTGAGAGCTCTTAGCTTTGCTTGCGTTTACGCTT	Cloning of anchor domains; *Sac*I site
RmlA2_Eco_F	GCTCGAATTCGCCAACATCATGGTTGACAG	Cloning of *welF*-*welE* into pIAV7; *Eco*RI site
WelG_ Sal_R	GCTCGTCGACGTGGGGTTTCGGGAGATTAT	Cloning of *welF*-*welE* into pIAV7; *Sal*I site

Table S2. Oligonucleotides used for quantification of expression by qPCR

Gene/region	Primer	Sequence (5’ to 3’)	Amplicon lenght in bp
LGG_00053	LGG_00053-Fw	AATTTGCGCCCAGCACTATG	84
	LGG_00053-Rv	CAGCACGATCGATGCACTTT	
LGG_00208	LGG_00208-Fw	CGATTCTACCAGGCAGTTGAT	105
	LGG_00208-Rv	GCGGAATACCGCTACATTCT	
LGG_00234	LGG_00234-Fw	GGGTGTTTGTTCCAAGCGTT	114
	LGG_00234-Rv	CCACAATCAATGAGACCGCC	
LGG_01436	LGG_01436-Fw	CACCGAGGTCATTGTTGCAG	119
	LGG_01436-Rv	TCTGATCAAGGGAGATGCCG	
LGG_01443	LGG_01443-Fw	TTTGTCACAAGCAGAGCACG	118
	LGG_01443-Rv	CTTACGCGGCAAACTTAGCA	
LGG_01444	LGG_01444-Fw	AGAACAAGGCCATCAGCAGT	88
	LGG_01444-Rv	CGGATCAAAGTCGGGTGTTG	
LGG_01445	LGG_01445-Fw	ACTCATTGCGCCATTACGTG	106
	LGG_01445-Rv	CAACAGTTGGGCAAGCTGAG	
LGG_01446	LGG_01446-Fw	CGACTTACCCGGAGTACCAG	89
	LGG_01446-Rv	ATCAATCAAACCGGTTGCGG	
LGG_01501	LGG_01501-Fw	AATCATTGCCTGGCTGCTTG	101
	LGG_01501-Rv	GATTGCCATCTGCCCATTCC	

LGG_01502	LGG_01502-Fw	CAAACGGTGGATTACGCCTC	105
	LGG_01502-Rv	CCCGATGAACCTTATTGGCG	
LGG_01503	LGG_01503-Fw	CTGTGGTCCTTACACGCAGA	111
	LGG_01503-Rv	ATGTGATGAGCGGAGTTGGT	
LGG_01504	LGG_01504-Fw	TGGCTATGTCAGTGCAGGAT	109
	LGG_01504-Rv	CACTGGCACTAATGGTTGGC	
WelE	WelE-Fw	CCCGGTCATTGAAGACGAGT	100
	WelE-Rv	TCGACCACTAGCTTGCCATT	
Upstr_WelF	Upstr_WelF-Fw	CGCGAGAGTTATTTCTAAAGGCT	112
	Upstr_WelF-Rv	GTGGTCGCATTTGGAAAGGT	
Downstr_WelF	Downstr_WelF-Fw	GCTGCAGTGCATATTCTGCC	90
	Downstr_WelF-Fw	TGATGTAACAACAGGAATTTCGCA	

Table S3. Genomic differences among the strains GG (ATCC 53103), GG (Gefilus®) and GG (UT) in comparison to the reference sequence (accession no. FM179322).

	Genomic variations			
Position of the ref. sequence	**GG (ATCC 53103)**	**GG (Gefilus®)**	**GG (UT)**	**Description**
57657	Ref.[a]	Ref.	ins 1585bp	Locus tag LGG_00053 (gene *kdgK;* product: 2-dehydro-3-deoxygluconokinase). Insertion.
240673	del 871bp	Ref.	Ref.	Locus tag LGG_00234 (product: hypothetical protein). Partial deletion. Loci tag LGG_00235 (gene *is7*; product: transposase is7) and LGG_00236 (gene *is8*; product: transposases is8). Deletion
264278	SNP G >A	Ref.	Ref.	Locus tag LGG_000261 (gene *iolT*; product : transporter major facilitator superfamily MFS_1). Amino acid change A420T.
656561	ins CGC	Ref.	Ref.	Locus tag LGG_00643 (gene *glpD*; product: alpha-glycerophosphate oxidase). Codon insertion G163-164ins.
1241467	SNP A->G	Ref.	Ref.	Locus tag LGG_01248 (gene *is39*; product: transposase, IS5 family

				protein). Amino acid change: C250R.
1994717	SNP T->A	Ref.	Ref.	Locus tag LGG_01953 (product: conserved protein). Amino acid change D903V.
2095506	Ref.	Ref.	SNP A->T	Locus tag LGG_02043 (gene *welE*; product: UDP-phosphate beta-glucosephosphotransferase). Amino acid change to stop codon: L177*.
2096942	Ref.	Ref.	ins 855 bp	Locus tag LGG_02044 (gene *welF*; product: glycosyl transferase, group 1). Insertion of is with two transposases.

[a] Ref., no difference with the reference sequence (accession no. FM179322).

Table S4. Genes with differential expression among *L. rhamnosus* GG strains as measured by microarray-based transcriptome analysis

GG (ATCC 53103) vs GG (Gefilus®)		**GG (ATCC 53103) vs GG (UT)**		**GG (Gefilus®) vs GG (UT)**			
Fold change[1]	**p-value**[2]	**Fold change**	**p-value**	**Fold change**	**p-value**	**Posible loci**[3]	**Gene product /description**
-1,30256379 -1,11498229 -1,12777148 -1,13955693	0,457472405 0,377996719 0,571051806 0,585961233	1,022262301 **-4,886399172** **-4,37344236** -1,386667327	0,959257016 **0,000240499** **0,004681272** 0,319650398	1,331561858 **-4,38249039** **-3,87795083** -1,21684778	0,323859406 **0,000276856** **0,005865375** 0,5173084	LGG_00152 LGG_00460 LGG_00461 LGG_01024 LGG_00457	Transposase
-1,12923092 -1,11599493	0,54191247 0,617717466	**-4,456677539** -1,374981157	**0,003303284** 0,315391192	**-3,94664853** -1,23206757	**0,004136994** 0,516997919	LGG_00153 LGG_00461 LGG_01023 LGG_01584 LGG_00457	Transposase
-1,00853061 -1,01677769	0,968939415 0,941849182	1,44391591 **-4,365380843**	0,149247972 **0,000357749**	1,456233396 **-4,29334837**	0,165106398 **0,000364026**	Intergenic region (LGG_01506 / LGG_01507)	13 bp at 5' side: 4-oxalocrotonate tautomerase and 400 bp at 3' side: ABC transporter, ATP-binding protein
-1,15191474 -1,20258081 -1,07725479 -1,13755898	0,476022183 0,46625983 0,740578086 0,366087732	-1,506199918 **-4,632497086** 1,204187583 **-5,015400418**	0,194362167 **0,001895731** 0,546424233 **0,000154675**	-1,30756198 **-3,85212957** 1,297216842 **-4,4089146**	0,37094104 **0,002521001** 0,332487072 **0,000184028**	LGG_00152 LGG_00457 LGG_00461 LGG_01024 LGG_01584	Transposase
-1,22428938 -1,18727539 -1,06330431 -1,16670804	0,440729323 0,497295235 0,64992799 0,009258703	-1,474009991 **-4,308960307** 1,037402897 **-5,242316734**	0,315765619 **0,002564362** 0,877305132 **1,05677E-05**	-1,20397188 **-3,62928461** 1,103074974 **-4,49325502**	0,596910764 **0,003359434** 0,669544017 **1,31552E-05**	LGG_00152 LGG_01584 LGG_00461 LGG_01024 LGG_00457	Transposase
-1,1947184 1,064313564 -1,04651865	0,623943702 0,129093692 0,664248404	**-3,589619186** **-4,362402503** -1,139739043	**0,027529411** **4,35561E-06** 0,601724761	**-3,00457345** **-4,64296416** -1,08907667	**0,036681336** **3,92639E-06** 0,717470046	LGG_00153 LGG_00461 LGG_01025	Transposase , IS4 family protein

-1,04168016	0,712872243	**-3,590533136**	**0,000505078**	**-3,44686715**	**0,000534201**	LGG_00458	
-1,08087368	0,510879754	-1,011144187	0,977787098	1,06896098	0,860940319		
1,030173979	0,92868417	-1,396312841	0,57043429	-1,43844516	0,530564403		
-4,38551179	**1,25369E-05**	**-4,180905506**	**5,96911E-05**	1,04893827	0,621807295	LGG_00236 LGG_00581 LGG_02160 LGG_02165	Transposase
2,126296256	0,249641023	4,441071718	0,094432438	2,088642025	0,164177562		
-3,541204615	**0,000114368**	**-4,178881487**	**8,89039E-06**	-1,180073433	0,11431372		
-4,505082369	**8,33901E-06**	**-5,312359691**	**6,44213E-08**	-1,179192578	0,025401288		
1,916282234	0,001712897	2,058539254	0,001350889	1,074235944	0,695958244		
-3,786489339	**0,00992647**	-2,937455427	0,009434152	1,289037207	0,35446701	LGG_00236	Transposase, IS4 family protein
1,455417423	0,063550548	1,366516927	0,103220222	-1,065056271	0,705959443	LGG_00581	
-2,414765335	1,78375E-05	-2,55181984	0,000724284	-1,056756863	0,610759693	LGG_02160	
1,806945298	0,100363228	1,884783312	0,080820138	1,043077128	0,901539369	LGG_02166	
1,053742604	0,761128215	2,301365176	0,005223161	2,18399177	0,005958459	LGG_00053*	2-dehydro-3-deoxygluconokinase
1,050333452	0,503537783	**3,717647226**	**6,15658E-06**	**3,539492357**	**9,91573E-06**		
1,014611503	0,974490235	-1,245784275	0,51073417	-1,26398706	0,470984151		
-1,1411888	0,689181282	-1,410842645	0,20619192	-1,23629205	0,428387985		
-1,00512619	0,989782576	-1,374618576	0,334137179	-1,33092662	0,334237605		
-2,6237764	0,020857579	-2,818210553	0,01230683	-1,0741047	0,788196361	LGG_00208*	Conserved hypothetical protein
-2,37705506	0,085096044	-2,10710234	0,128624588	1,12811562	0,754059386		
-2,62132016	0,006948493	-2,889171548	0,006252509	-1,10218187	0,653186535		
-2,30642079	0,002415726	-2,972176756	0,002542378	-1,2886533	0,168182339		
-2,57139599	0,029904663	-2,9996609	0,016370001	-1,16654957	0,605850816		
-2,63442065	0,000266002	**-3,283704759**	**4,1804E-05**	-1,24646182	0,067604243		
1,5145655	0,001249791	1,343293939	0,01693384	-1,127501179	0,266157576	LGG_00234*	Putative protein without homology
-1,216785328	0,548591046	-1,160915285	0,609953063	1,048125857	0,842382904		
1,32612977	0,292774794	1,38665235	0,255333229	1,045638505	0,722009226		
-1,331768017	0,703134188	-1,185303538	0,818439988	1,123567065	0,869012594		
-13,90558608	**1,74343E-07**	**-15,48328312**	**6,75035E-06**	-1,113457788	0,20390891		
1,307881557	0,210917451	1,416821138	0,168132402	1,083294684	0,813530316		
-1,182930563	0,642833674	**-3,590353892**	0,038174421	**-3,035134947**	0,049791598	LGG_00458	Transposase, IS4 family protein
-1,023834232	0,839265045	-1,156739651	0,568032451	-1,129811462	0,623070854		
-1,057595816	0,784199823	**-3,756996305**	**0,001273333**	**-3,552393313**	**0,001091365**		
1,00300609	0,985193006	2,139826384	0,007406187	2,133413153	**0,008119082**		
-1,011269562	0,883307379	**-3,709753524**	**0,000596917**	**-3,668412127**	**0,000626199**		
-1,170826612	0,520688958	-1,656340722	0,408761724	-1,414676353	0,551613584		
-1,166267101	0,709080442	**-3,271309728**	0,028891283	-2,804940417	0,039740542	LGG_00460	Transposase , IS4 family

-1,010786787	0,881915709	-1,162072452	0,503801962	-1,149671193	0,529215673		protein
1,080355846	0,086635933	**-4,389593654**	**3,1288E-05**	**-4,742323166**	**2,77712E-05**		
-1,081232612	0,452564341	-1,027133284	0,93975676	1,052670212	0,880377497		
-1,00018044	0,99825333	**-3,65622502**	**0,000497671**	**-3,655565411**	**0,000491405**		
1,030315183	0,926766786	-1,399324696	0,56795582	-1,44174548	0,532522535		
2,697318352	0,01311332	**3,084617243**	0,00940663	1,143586644	0,418050577	LGG_01436*	Phosphoribosyl-ATP pyrophosphatase
-1,132413159	0,248880091	-1,03328275	0,340664789	1,095937349	0,393421968		
2,618388444	0,003226106	2,388934563	0,005404877	-1,096048625	0,647810116		
1,908890543	0,049214466	1,771142564	0,115248397	-1,077773512	0,843125398		
1,925533798	0,118589244	1,833120329	0,196016696	-1,050413204	0,911590478		
1,544647824	0,155376412	1,390495523	0,200904871	-1,110861415	0,58515157		
3,021175534	0,022756009	**3,477754998**	0,017537965	1,151126427	0,517065603	LGG_01443*	Histidinol dehydrogenase
3,081353962	0,040214565	**4,504362548**	0,023335072	1,461812763	0,143824809		
2,96985535	0,011578311	**3,35396382**	0,009014873	1,12933575	0,546198754		
1,349853571	0,293420905	1,313876418	0,27484732	-1,027382448	0,90117453		
1,455714738	0,049665203	1,361906546	0,073647472	-1,068880051	0,6229206		
2,993164502	0,174889103	4,414862711	0,117217948	1,474981648	0,304596546	LGG_01444*	ATP phosphoribosyltransferase catalytic subunit
3,003355391	**0,005599601**	**4,279925982**	**0,002564343**	1,425048129	0,13233526		
3,207148048	0,069928142	**5,362088123**	0,038419978	1,671917867	0,126865575		
1,74815267	0,009317087	1,931701547	0,014512359	1,104995908	0,670381842		
2,754352747	0,027084486	**4,34144726**	0,012552324	1,576213237	0,092552649	LGG_01445*	ATP phosphoribosyltransferase catalytic subunit
1,644854409	0,037527026	-1,247653638	0,695073227	-2,052208588	0,321000224		
3,311039168	0,008512674	**4,937636288**	**0,004076865**	1,491264838	0,082105458		
-1,052610961	0,925258359	1,309554611	0,561450052	1,378451537	0,513479721		
3,146965067	0,023891656	**4,554285275**	0,012587814	1,447199183	0,204019471		
1,234379803	0,446213207	-3,067243401	0,2953112	-3,786143305	0,256422954		
3,082330386	0,057133519	**4,985480773**	0,027208092	1,617438804	0,279341027	LGG_01446*	Histidinol-phosphate aminotransferase
2,521812276	0,013035841	2,610541418	0,033600012	1,035184674	0,933264132		
3,843246457	**0,00034997**	**4,02937715**	**0,00032422**	1,04843059	0,8311056		
2,6226731	0,059611234	2,324475117	0,077863416	-1,128286159	0,571061639		
3,176853674	**2,88246E-05**	**3,196854497**	**7,52717E-05**	1,006295796	0,968189609		
3,280996185	0,098198077	3,128073275	0,104478386	-1,048887253	0,879100047		
-1,010519757	0,983457659	**6,278570944**	0,059967109	6,344619981	0,04199355	LGG_01501*	tRNA and rRNA cytosine-C5-methylases
-1,048537733	0,81084965	2,559486129	0,005324847	2,683717782	0,004927546		
-1,011304138	0,939697245	**4,017239436**	**0,000300331**	**4,062650866**	**0,000451339**		

1,007006388	0,972395924	-1,084138986	0,731775044	-1,091734884	0,705566061		
1,048791832	0,720319657	**3,697207959**	**0,000556798**	**3,525206668**	**6,60923E-05**		
1,012238715	0,969933676	1,152141944	0,624400591	1,138211696	0,634812572		
-1,083206879	0,410771405	**3,715386533**	**0,000124546**	**4,024532252**	**1,02922E-05**	LGG_01502*	Conserved membrane protein
-1,115677021	0,665994295	**15,3270839**	**0,00065531**	**17,1000753**	**0,002910899**		
1,016527831	0,909501052	**3,528345791**	**0,000704689**	**3,470978051**	**0,000457503**		
-1,192665751	0,304660373	2,092600326	**0,000475591**	2,495772739	0,007107846		
1,013261413	0,840822942	**3,457452196**	**1,53946E-05**	**3,412201581**	**3,5102E-05**		
-1,03068072	0,944834966	1,994145308	0,161684829	2,055327122	0,126362938		
-1,01862374	0,855337511	-1,039357113	0,713687374	-1,020354299	0,856486059	LGG_01503*	Conserved protein
-1,157991087	0,302493532	-1,380701556	0,034460838	-1,192324856	0,17894424		
-1,007939898	0,869874965	1,23334967	0,019943139	1,243142341	0,0161553		
-1,00508806	0,979560299	**-4,439615325**	**0,000116874**	**-4,417140649**	**0,000113195**		
1,013521548	0,931550385	-1,097826528	0,481895101	-1,112670842	0,380645197		
-1,02423886	0,910937025	-1,20390548	0,227339324	-1,175414766	0,279841085		
-1,22074681	0,639030474	**-3,215787214**	0,031410877	-2,634278613	0,04718469	LGG_01584	Transposase, IS4 family protein
-1,074514563	0,469489231	1,047072567	0,899625355	1,125094722	0,739002112		
1,07172963	0,146073825	**-4,375614271**	**5,87923E-06**	**-4,689475463**	**5,12654E-06**		
-1,010001088	0,975665123	-1,446661112	0,54346983	-1,432336192	0,546772518		
-1,041763005	0,676913954	**-3,591177789**	**0,000345367**	**-3,447211864**	**0,000396443**		
-3,769963024	**0,000743643**	**-4,262903842**	**0,00014831**	-1,130754815	0,387472306	LGG_01962	Transposase
-1,711248706	0,186627567	**-3,2247528**	0,12538614	-1,884444259	0,253231792		
1,139370067	0,481514895	-1,089866002	0,623661785	-1,241760701	0,198156037		
-2,243533991	0,007984363	**-3,203572563**	0,034512663	-1,42791354	0,277323617		
1,143142063	0,50703659	-1,311712965	0,329245784	-1,499474265	0,154588157		
1,172806112	0,474366414	-1,19378986	0,490319229	-1,400084044	0,205267811		

[1] The loci presenting a fold change in transcription level of ≥ 3 ($p \leq 0.05$) between pair of strains using at least one probe are indicated in bold.

[2] The expression levels between pair of strains were compared using two-tailed Student's t-test.

[3] The expression of loci marked with (*) was further analysed by real-time PCR.