SUPPLEMENTARY INFORMATION

Genetic tools for the industrially promising methanotroph *Methylomicrobium buryatense*

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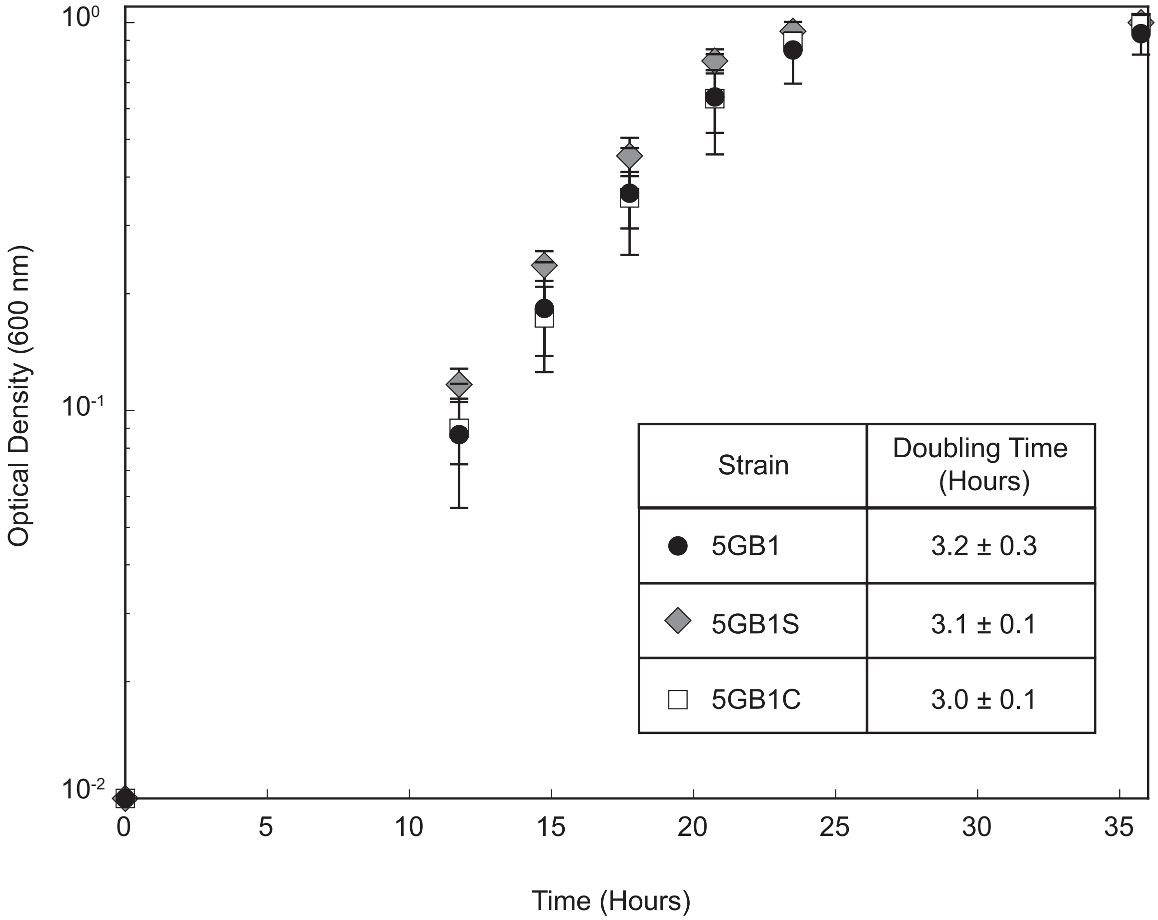
Supplementary Figures

Fig S1. Growth curves and doubling times for *M. buryatense* strains 5GB1, 5GB1S, and 5GB1C. Data represents the mean of 3 replicates ± standard deviation.



Strain	Doubling Time (Hours)
● 5GB1	3.2 ± 0.3
◆ 5GB1S	3.1 ± 0.1
□ 5GB1C	3.0 ± 0.1

Fig S2. Diagnostic PCR showing native plasmid loss in *M. buryatense* 5GB1S and 5GB1C compared to 5GB1. PCR A: Chromosomal locus positive control (expected size: 621 bp), PCRs B-D: 3 native plasmid loci (expected sizes: 590 bp, 640 bp, 635 bp respectively). Primer sequences are listed in Table S1.

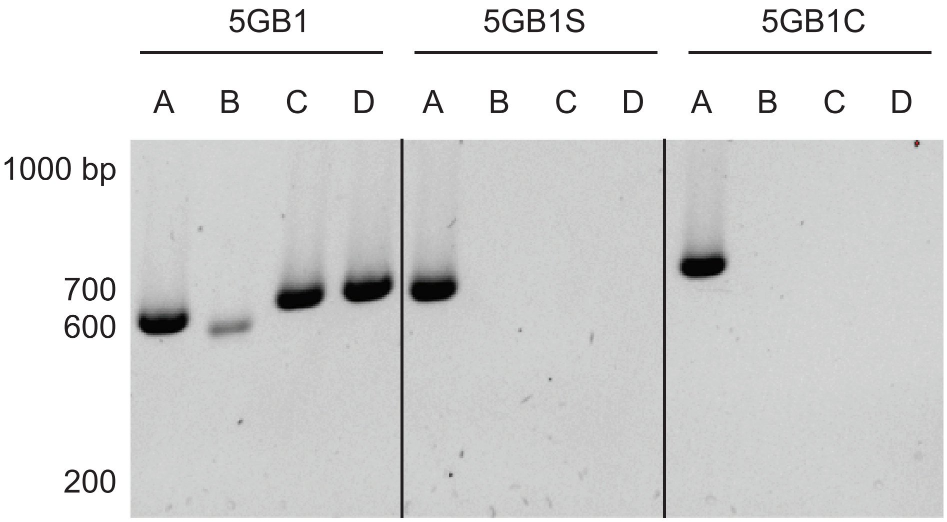


Fig S3. IncP-based broad host range vector pCM66. Leftover pieces of antibiotic resistance genes from restriction based cloning methods are shown in red.

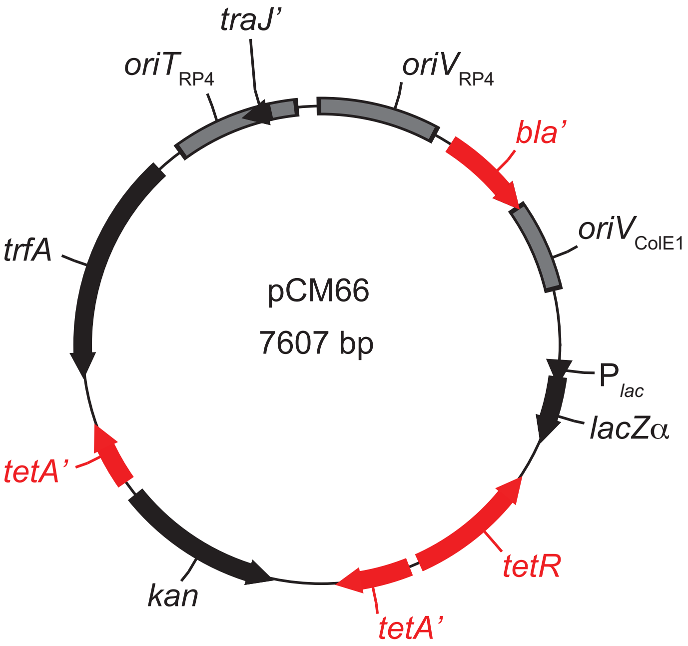
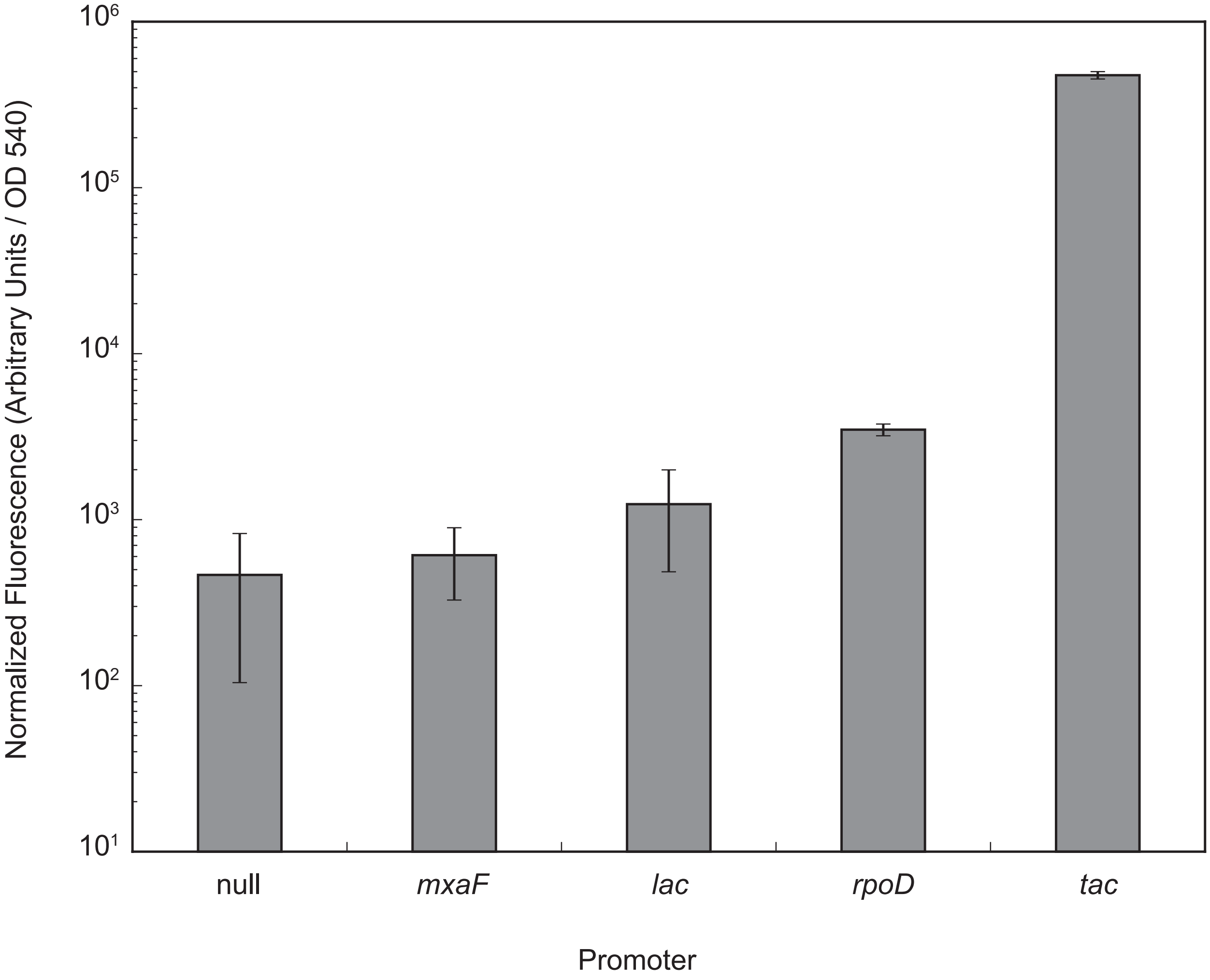


Fig S4. Relative promoter strengths in *E. coli*. Fluorescence (Ex 535nm/Em 590nm) was measured in whole *E. coli* TOP10 cells containing probe vectors with promoter-dTomato fusions and normalized to optical density (540 nm). Data represents the mean of 3 replicates ± standard deviation.



Supplementary Tables

Table S1. Selected primers used in this study. Homology regions used for Gibson Assembly are underlined.

Primer Name	**Sequence (5' to 3')**	**Description**
AP186_pCM433kanT_fwd1	ATGTGCAGGTTGTCGGTGTC	For amplifying pCM433kanT backbone.
AP187_pCM433kanT_rev1	TGGTAACTGTCAGACCAAGTTTACTC	
AP259_pAWP78_fwd1	TTGTCGGGAAGATGCGTGAT	For amplifying pAWP78 backbone.
AP254_pAWP78_rev1	CAGCTCACTCAAAGGCGGTA	
AP88_433kanRvect_fwd1	TACGTACGCGTGTTAACCGG	For amplifying *glgA* knockout vector backbones.
AP89_433kanRvect_rev1	AGCTGCAGCATATGCATCCA	
AP90_glgA1_Up_fwd1	TGGATGCATATGCTGCAGCTGAGTTCGGGCAAGGTACAGA	For amplifying flanks to knock out glycogen synthase *glgA1* (METBUDRAFT_3833 / MBURv2_210177) with pAWP45.
AP91_glgA1_Up_rev1	GAACTCTCCAGGCAGTCAAAATTCGGTTCTAG	
AP92_glgA1_Dwn_fwd1	TTTGACTGCCTGGAGAGTTCGGTAACTCGATTG	
AP93_glgA1_Dwn_rev1	CCGGTTAACACGCGTACGTATGTGGAATTACCGGAAGGCA	
AP94_glgA2_Up_fwd1	TGGATGCATATGCTGCAGCTAGTTGGGGCGAACAAGGCTA	For amplifying flanks to knock out glycogen synthase *glgA1* (METBUDRAFT_3841 / MBURv2_210185) with pAWP47.
AP95_glgA2_Up_rev1	ATCGGCCTCTATTTATCATCCCGCTGCCCTTAG	
AP96_glgA2_Dwn_fwd1	GATGATAAATAGAGGCCGATTGATGAGGAT	
AP97_glgA2_Dwn_rev1	CCGGTTAACACGCGTACGTAGCAAATAAACGAGGCGTATC	
AP339_repAB_Up_fwd1	ATATGAGTAAACTTGGTCTGACAGTTACCAAGATCTGCGGATTTAGGTCATG	For amplifying flanks to knock out *repA* (METBUDRAFT_0076 / MBURv2_10076) to *repB* (METBUDRAFT_0086 / MBURv2_10086)
AP340_repAB_Up_rev1	TCGGAACCTGCCAATGGCAATTTCCTATCTTTTGG	
AP341_repAB_Dwn_fwd1	AGATAGGAAATTGCCATTGGCAGGTTCCGACCCAT	
AP342_repAB_Dwn_rev1	CGTGCATCACGACACCGACAACCTGCACATGGTATTAAGTTCCGCATC	

	AATAGCTTC	native plasmid locus using pAWP93.
AP1_plasdiagPCRA_fwd1	GAGGACGTCCACAGCGGCTTTGACTGGATCG	For native plasmid loss diagnostic PCR (Figure S2).
AP4_plasdiagPCRA_rev1	GAGGAGCTCCGTCAAAGGACGCCGTGAGCCCAG	
AP346_ plasdiagPCRB_fwd1	AATCCGTCCCATGAGCCTTCCC	
AP347_ plasdiagPCRB_rev1	CCCAACAAGGCTAATGCAGC	
AP348_ plasdiagPCRC_fwd1	TTATCCCGAAGGCGAAACATGC	
AP349_ plasdiagPCRC_rev1	GCACTCTAGCGACAAATATCCGA	
AP350_ plasdiagPCRD_fwd1	AAGGCCGCACAGGAAGTTCG	
AP351_ plasdiagPCRD_rev1	GACGGTTACATCCCTGACGCGA	

Table S2. Promoter sequences for dTomato probe vectors. The ATG start codon of dTomato occurs immediately following these sequences in each plasmid.

Promoter	**Sequence (5' to 3')**	**dTomato reporter plasmid containing promoter**
lac	CCAGGCTTTACACTTTATGCTTCCGGCTCGTATGTTGTGTGGAGGTATTCACACAGGAAACAGCT	pAWP88
tac	CTCTGAAATGAGCTGTTGACAATTAATCATCGGCTCGTATAATGTGTGGAGGTATTCACACAGGAAACAGCT	pAWP89
300 bp upstream of *rpoD* (METBUDRAFT_3024 / MBURv2_160101)	GGACGACACAGCGAGAAAGTTGTGAAGCAACTGGTTTTTTGGATGTGATGGTTCCCGAGGAAGGGGTTCGTGCCGAATTTTCAGACGCTTTGGAACGATTGAATGCGCAAGCGAGAGAAGGTCGTATTGAGCAGTTATTAGAAAAAGAGTCGGCGGAAGGCTTAAGTCGAGAGGAGCGGGAAATATTAGGGAGGTTACTGGTAAGCCGATAAAAGGGAATTCGTATTAACATAAAAAAGAATATGTTGTATAATTTCCTGTTCAGCGTAATTGTGCTGATCGTATTCTGTGAGTGAGTG	pAWP92
300 bp upstream of *mxaF* (METBUDRAFT_3941 / MBURv2_210291)	AATTAAACCGGGAATGATGTCGGATATTTAACGGCAAAGCCATGGGAGCTTTTCCCGAATTTGAATGCCGACATACTCTCGGGATATTTTCCCTGTTTTTTCTTAGCGCTTTTCCCGTCATCTGGGTGCTGTATTCCGTAACGTCGCATCCCGCTCCTTCCGTATGATTACCGTCCGTGCGCTGCCCTCTATGAATGATTCGTTATGCGCCTTGATCAAGCTAAGCCGGTTGTAACAACAAACACCGCAATCAATAGGGGGCCGCGCCGACATTATGCGAAAAATCAATCTGGAGGAATT	pAWP87

Table S3. Genotypic differences of *Methylomicrobium buryatense* 5GB1 compared to published *Methylomicrobium buryatense* 5G genome (NCBI accession AOTL01000000).

In addition to the listed genotypic differences, we also discovered that the 5GB1 genome contains a 309 bp transposon insertion in the sigma factor gene *rpoS*, leading to a truncated 218 amino acid RpoS product.

See next page for table.

breseq version 0.24rc6
mutation predictions | marginal predictions | summary statistics | genome diff | command line log

Predicted mutations

evidence	position	mutation	annotation	gene	description
JC	140,502	+GGAAAGA	intergenic (+16/-404)	*MBURv2_20047* → / → *MBURv2_20048*	Peptidase S1 and S6, chymotrypsin/Hap/Undecaprenyl-phosphate galactosephosphotransferase
RA	393,809	G→T	Q341K (CAA→AAA)	*MBURv2_20291* ←	conserved exported protein of unknown function
JC	431,236	+ACCGCTT	coding (162/279 nt)	*MBURv2_20329* →	membrane protein of unknown function
RA	580,112	+TTCACTA	intergenic (+135/+104)	*MBURv2_20468* → / ← *pgm*	Zinc/iron permease/Phosphoglucomutase
JC	667,699	Δ21 bp	intergenic (+70/-72)	*MBURv2_30052* → / → *MBURv2_30053*	conserved protein of unknown function/conserved hypothetical protein
JC	676,649	56 bp x 2	duplication	*MBURv2_30060* → / → *MBURv2_30061*	GTP-binding protein HSR1-related protein/protein of unknown function
JC	770,914	Δ21 bp	intergenic (+799/-119)	*MBURv2_30133* → / → *cas2*	conserved exported protein of unknown function/CRISPR-associated endoribonuclease Cas2 1
RA	780,024	C→T	A198V (GCC→GTC)	*MBURv2_30144* →	conserved protein of unknown function
RA	810,254	T→G	F1000L (TTT→TTG)	*MBURv2_30169* →	Linear gramicidin synthase subunit D [Includes: ATP-dependent D-leucine adenylase ; Leucine racemase [ATP-hydrolyzing] ; ATP-dependent tryptophan adenylase ; ATP-dependent glycine adenylase ; Linear gramicidin--PCP reductase] (fragment)
RA	863,269	A→G	intergenic (+65/-471)	*MBURv2_50037* → / → *rbgA*	conserved protein of unknown function/Ribosome biogenesis GTPase A
RA	863,356	G→T	intergenic (+152/-384)	*MBURv2_50037* → / → *rbgA*	conserved protein of unknown function/Ribosome biogenesis GTPase A
RA	863,363	G→T	intergenic (+159/-377)	*MBURv2_50037* → / → *rbgA*	conserved protein of unknown function/Ribosome biogenesis GTPase A
JC	924,720	Δ7 bp	intergenic (+214/-72)	*MBURv2_50095* → / → *MBURv2_50096*	conserved protein of unknown function/Sugar transferase, PEP-CTERM/EpsH1 system associated
RA	949,136	C→T	G57G (GGC→GGT)	*MBURv2_50113* →	OmpA family protein
RA	949,141	2 bp→TC	coding (176-177/831 nt)	*MBURv2_50113* →	OmpA family protein
RA	949,146	G→C	D61H (GAT→CAT)	*MBURv2_50113* →	OmpA family protein
RA	949,148	3 bp→GAA	coding (183-185/831 nt)	*MBURv2_50113* →	OmpA family protein
RA	949,163	T→G	D66E (GAT→GAG)	*MBURv2_50113* →	OmpA family protein
RA	997,352	N→G	?89K (AAN→AAG)	*MBURv2_50154* →	transposase
RA	1,105,229	N→C	?172P (CNG→CCG)	*MBURv2_50241* →	conserved protein of unknown function
RA	1,421,698	Δ1 bp	coding (9/144 nt)	*MBURv2_60051* →	protein of unknown function
JC	1,516,562	Δ12 bp	coding (783-794/993 nt)	*MBURv2_60136* ←	conserved protein of unknown function
RA	1,517,657	Δ1 bp	coding (55/156 nt)	*MBURv2_60137* →	protein of unknown function
RA	1,519,665	C→T	E310E (GAG→GAA)	*MBURv2_60142* ←	transposase
RA	1,519,668	G→A	I309I (ATC→ATT)	*MBURv2_60142* ←	transposase
RA	1,536,111	N→T	?422G (GGN→GGT)	*dxs* →	1-deoxyxylulose-5-phosphate synthase, thiamine-requiring, FAD-requiring
RA	1,536,141	A→C	A432A (GCA→GCC)	*dxs* →	1-deoxyxylulose-5-phosphate synthase, thiamine-requiring, FAD-requiring
RA	1,536,252	N→T	?469G (GGN→GGT)	*dxs* →	1-deoxyxylulose-5-phosphate synthase, thiamine-requiring, FAD-requiring
RA	1,684,299	Δ1 bp	intergenic (+82/-477)	*MBURv2_60281* → / → *MBURv2_60282*	Retron-type reverse transcriptase (fragment)/transposase
RA	1,712,205	N→T	?151L (CTN→CTT)	*umuC* →	DNA polymerase V, subunit C
RA	1,712,263	N→T	?171L (NTN→TTA)	*umuC* →	DNA polymerase V, subunit C
RA	1,712,265	N→A	?171L (NTN→TTA)	*umuC* →	DNA polymerase V, subunit C
MC JC	1,815,401	Δ118 bp	intergenic (-227/+445)	*MBURv2_60409* ← / ← *MBURv2_70001*	protein of unknown function/Plasmid stabilization system
MC JC	1,817,604	Δ150 bp		*[MBURv2_80001]*	*[MBURv2_80001]*
RA	2,126,951	N→G	?191C (TNT→TGT)	*MBURv2_120134* →	conserved protein of unknown function
RA	2,228,653	C→T	G199G (GGC→GGT)	*yhhI* →	H repeat-associated protein YhhI
RA	2,254,116	N→C	?386C (TNT→TGT)	*MBURv2_130091* ←	conserved protein of unknown function
JC	2,271,543	Δ97 bp	intergenic (-562/+115)	*MBURv2_130105* ← / ← *MBURv2_130106*	Multicopper oxidase type 3/Disulfide bond isomerase, DsbC/G-like protein
RA	2,430,526	Δ1 bp	intergenic (-118/+114)	*MBURv2_130232* ← / ← *fliI*	Flagellar export protein FliJ/flagellum-specific ATP synthase
					Flagellar export protein FliJ/flagellum-specific ATP

RA	2,430,528	Δ1 bp	intergenic (-120/+112)	*MBURv2_130232* ← / ← *fliI*	synthase
RA	2,430,531	2 bp→G	intergenic (-123/+108)	*MBURv2_130232* ← / ← *fliI*	Flagellar export protein FliJ/flagellum-specific ATP synthase
RA	2,430,534	3 bp→GCG	intergenic (-126/+104)	*MBURv2_130232* ← / ← *fliI*	Flagellar export protein FliJ/flagellum-specific ATP synthase
RA	2,430,538	A→C	intergenic (-130/+102)	*MBURv2_130232* ← / ← *fliI*	Flagellar export protein FliJ/flagellum-specific ATP synthase
RA	2,430,541	2 bp→TT	intergenic (-133/+98)	*MBURv2_130232* ← / ← *fliI*	Flagellar export protein FliJ/flagellum-specific ATP synthase
RA	2,430,544	G→A	intergenic (-136/+96)	*MBURv2_130232* ← / ← *fliI*	Flagellar export protein FliJ/flagellum-specific ATP synthase
RA	2,430,546	A→C	intergenic (-138/+94)	*MBURv2_130232* ← / ← *fliI*	Flagellar export protein FliJ/flagellum-specific ATP synthase
RA	2,430,549	G→C	intergenic (-141/+91)	*MBURv2_130232* ← / ← *fliI*	Flagellar export protein FliJ/flagellum-specific ATP synthase
RA	2,430,551	Δ3 bp	intergenic (-143/+87)	*MBURv2_130232* ← / ← *fliI*	Flagellar export protein FliJ/flagellum-specific ATP synthase
RA	2,430,555	Δ1 bp	intergenic (-147/+85)	*MBURv2_130232* ← / ← *fliI*	Flagellar export protein FliJ/flagellum-specific ATP synthase
RA	2,430,558	G→C	intergenic (-150/+82)	*MBURv2_130232* ← / ← *fliI*	Flagellar export protein FliJ/flagellum-specific ATP synthase
RA	2,460,123	C→A	intergenic (-369/+110)	*bamA* ← / ← *MBURv2_130262*	Outer membrane protein assembly factor BamA/AFG1-family ATPase
JC	2,477,643	Δ49 bp	intergenic (-68/+240)	*MBURv2_130276* ← / ← *MBURv2_130277*	conserved protein of unknown function/putative virion core protein (Lumpy skin disease virus)-like protein
RA	2,593,133	Δ1 bp	coding (775/795 nt)	*MBURv2_130386* →	protein of unknown function
RA	2,663,596	G→T	H532N (CAC→AAC)	*rpoB* ←	RNA polymerase, beta subunit
JC	2,703,485	Δ49 bp	intergenic (-347/+336)	*MBURv2_130501* ← / ← *ectC*	Tetratricopeptide repeat domain protein/L-ectoine synthase
RA	2,718,814	N→T	intergenic (-599/-275)	*MBURv2_130515* ← / → *MBURv2_130516*	conserved exported protein of unknown function/Retron-type reverse transcriptase
JC	2,758,218	Δ49 bp	intergenic (+325/-123)	*MBURv2_130543* → / → *MBURv2_130544*	conserved protein of unknown function/conserved protein of unknown function
RA	2,939,318	Δ2 bp	intergenic (+162/+98)	*ispH* → / ← *MBURv2_130718*	1-hydroxy-2-methyl-2-(E)-butenyl 4-diphosphate reductase, 4Fe-4S protein/putative TrkA-N domain protein
RA	3,243,751	Δ1 bp	intergenic (+15/-154)	*umuC* → / → *MBURv2_130990*	DNA polymerase V, subunit C/conserved protein of unknown function
RA	3,402,031	G→C	P136A (CCT→GCT)	*MBURv2_160132* ←	conserved protein of unknown function
RA	3,908,877	N→T	?309I (ATN→ATT)	*MBURv2_170001* →	transposase
RA	3,908,880	N→A	?310E (GAN→GAA)	*MBURv2_170001* →	transposase
RA	3,912,896	A→G	I46I (ATT→ATC)	*MBURv2_170005* ←	protein of unknown function
RA	3,912,899	G→C	V45V (GTC→GTG)	*MBURv2_170005* ←	protein of unknown function
RA	3,912,921	A→G	I38T (ATA→ACA)	*MBURv2_170005* ←	protein of unknown function
RA	3,939,410	C→G	T1285T (ACG→ACC)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,464	T→G	S1267S (TCA→TCC)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,466	2 bp→TA	coding (3798-3799/3963 nt)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,482	C→G	T1261T (ACG→ACC)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,497	G→A	D1256D (GAC→GAT)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,503	G→A	D1254D (GAC→GAT)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,509	G→A	D1252D (GAC→GAT)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,512	C→G	T1251T (ACG→ACC)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,524	C→G	T1247T (ACG→ACC)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,566	T→G	T1233T (ACA→ACC)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,569	G→A	D1232D (GAC→GAT)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,590	C→G	T1225T (ACG→ACC)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,593	G→A	D1224D (GAC→GAT)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,635	G→A	D1210D (GAC→GAT)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,659	G→A	D1202D (GAC→GAT)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,701	G→A	D1188D (GAC→GAT)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,939,968	T→G	T1099T (ACA→ACC)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,940,037	G→A	D1076D (GAC→GAT)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,940,049	G→A	D1072D (GAC→GAT)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,940,061	G→A	D1068D (GAC→GAT)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)

RA	3,940,091	G→A	D1058D (GAC→GAT)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,940,144	A→T	S1041T (TCA→ACA)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,940,738	A→T	S843T (TCC→ACC)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	3,941,854	A→T	S471T (TCA→ACA)	*MBURv2_190001* ←	LPXTG-motif cell wall anchor domain protein (fragment)
RA	4,028,796	Δ7 bp	intergenic (+89/+611)	*yraL* → / ← *MBURv2_190083*	putative methyltransferase/protein of unknown function
RA	4,090,457	G→A	intergenic (+464/-828)	*MBURv2_200002* → / → *MBURv2_210001*	conserved protein of unknown function/protein of unknown function
RA	4,099,325	+T	intergenic (+194/-634)	*MBURv2_210008* → / → *MBURv2_210009*	conserved exported protein of unknown function/conserved exported protein of unknown function
RA	4,099,625	3 bp→GTG	intergenic (+494/-332)	*MBURv2_210008* → / → *MBURv2_210009*	conserved exported protein of unknown function/conserved exported protein of unknown function
RA	4,262,607	G→C	intergenic (+111/-40)	*MBURv2_5s_rRNA_1* → / → *MBURv2_210142*	ribosomal RNA 5s_rRNA/protein of unknown function
RA	4,319,976	N→C	?292C (TNT→TGT)	*MBURv2_210192* ←	conserved protein of unknown function
RA	4,335,952	A→T	H104Q (CAT→CAA)	*MBURv2_210207* ←	conserved protein of unknown function
RA	4,343,534	N→G	?11L (NTG→CTG)	*MBURv2_210214* ←	transposase (fragment)
RA	4,363,037	C→A	A183A (GCC→GCA)	*MBURv2_210234* →	conserved protein of unknown function
RA	4,371,038	T→C	K267E (AAA→GAA)	*MBURv2_210244* ←	protein of unknown function
JC	4,439,764	+TTCACTA	coding (80/276 nt)	*MBURv2_210305* →	protein of unknown function
JC	4,466,694	Δ4 bp	intergenic (-76/-271)	*MBURv2_210339* ← / → *speE*	protein of unknown function/Spermidine synthase
RA	4,516,410	2 bp→GC	coding (584-585/1581 nt)	*MBURv2_210383* →	transposase
RA	4,529,979	T→C	R35R (CGA→CGG)	*MBURv2_210391* ←	transposase
RA	4,530,076	Δ1 bp	coding (8/1104 nt)	*MBURv2_210391* ←	transposase
RA	4,530,105	Δ2 bp	intergenic (-22/-138)	*MBURv2_210391* ← / → *MBURv2_210392*	transposase/conserved protein of unknown function
RA	4,530,126	2 bp→AT	intergenic (-43/-117)	*MBURv2_210391* ← / → *MBURv2_210392*	transposase/conserved protein of unknown function
RA	4,530,130	Δ1 bp	intergenic (-47/-114)	*MBURv2_210391* ← / → *MBURv2_210392*	transposase/conserved protein of unknown function
RA	4,530,166	Δ1 bp	intergenic (-83/-78)	*MBURv2_210391* ← / → *MBURv2_210392*	transposase/conserved protein of unknown function
RA	4,530,739	+C	coding (496/519 nt)	*MBURv2_210392* →	conserved protein of unknown function
RA	4,925,409	C→G	intergenic (+100/+153)	*MBURv2_250059* → / ← *MBURv2_250060*	conserved exported protein of unknown function/conserved membrane protein of unknown function
RA	4,925,434	Δ14 bp	intergenic (+125/+115)	*MBURv2_250059* → / ← *MBURv2_250060*	conserved exported protein of unknown function/conserved membrane protein of unknown function
JC	4,992,169	182 bp x 2	duplication	*MBURv2_250100* → / → *MBURv2_250101*	conserved protein of unknown function/conserved protein of unknown function
RA	5,007,736	G→A	G42D (GGC→GAC)	*MBURv2_250115* →	protein of unknown function
RA	5,020,996	Δ3 bp	intergenic (-372/-138)	*MBURv2_260006* ← / → *MBURv2_260007*	protein of unknown function/Molybdopterin biosynthesis protein MoeB
RA	5,044,973	C→T	A354A (GCG→GCA)	*dxs* ←	1-deoxyxylulose-5-phosphate synthase, thiamine-requiring, FAD-requiring

Unassigned missing coverage evidence

	seq id	start	end	size	←reads	reads→	gene	description
* * ÷	MBURv2	82732	82831	100	236 [0]	[0] 196	*[MBURv2_10087]*	[MBURv2_10087]
* * ÷	MBURv2	609333–609427	613315–613053	3627–3983	118 [114]	[0] 245	*[MBURv2_20501]–MBURv2_30007*	[MBURv2_20501], MBURv2_30001, MBURv2_30002, MBURv2_30003, MBURv2_30004, MBURv2_30005, MBURv2_30006, MBURv2_30007
* * ÷	MBURv2	779034–779175	779286–779178	4–253	116 [114]	[112] 115	*[MBURv2_30143]*	[MBURv2_30143]
* * ÷	MBURv2	819847–819961	820061	101–215	117 [111]	[0] 1398	*[MBURv2_30179]*	[MBURv2_30179]
* * ÷	MBURv2	820833	821033–820933	101–201	711 [0]	[113] 115	*mbtH/MBURv2_50001*	Protein MbtH/RNA polymerase, sigma-24 subunit, ECF subfamily
* * ÷	MBURv2	1353350	1353550–1353452	103–201	275 [0]	[113] 116	*[MBURv2_60001]*	[MBURv2_60001]
* * ÷	MBURv2	1422819–1422897	1423331–1423035	139–513	116 [112]	[113] 115	*[MBURv2_60052]–[MBURv2_60054]*	[MBURv2_60052], MBURv2_60053, [MBURv2_60054]
* * ÷	MBURv2	1680105–1680275	1680404–1680311	37–300	116 [113]	[114] 115	*MBURv2_60275*	transposase
* * ÷	MBURv2	1960799	1960974	176	116 [114]	[0] 438	*MBURv2_80115/MBURv2_90001*	putative Glycosyl transferase family 2/conserved protein of unknown function
* * ÷	MBURv2	1964952	1965274–1965139	188–323	116 [113]	[113] 117	*[MBURv2_90005]–[MBURv2_100001]*	[MBURv2_90005], [MBURv2_100001]
* * ÷	MBURv2	1967033	1967223	191	442 [1]	[114] 116	*[MBURv2_100003]*	[MBURv2_100003]

* * ÷	MBURv2	1967488–1967597	1967778–1967771	175–291	116 [114]	[114] 115	*[MBURv2_120001]*	[MBURv2_120001]
* * ÷	MBURv2	2139320–2140548	2141155–2140652	105–1836	116 [114]	[114] 115	*MBURv2_120145–MBURv2_120146*	MBURv2_120145, MBURv2_120146
* * ÷	MBURv2	3249728–3250761	3250930–3250862	102–1203	115 [114]	[114] 115	*[MBURv2_130999]–[MBURv2_140001]*	[MBURv2_130999], MBURv2_131000, [MBURv2_140001]
* * ÷	MBURv2	3252480	3253310–3252580	101–831	180 [0]	[114] 118	*[MBURv2_150001]*	[MBURv2_150001]
* * ÷	MBURv2	3264171–3264319	3264823–3264420	102–653	115 [113]	[113] 115	*[MBURv2_150015]–[MBURv2_160002]*	[MBURv2_150015], MBURv2_160001, [MBURv2_160002]
* * ÷	MBURv2	3439232–3439434	3439984–3439745	312–753	115 [113]	[114] 115	*MBURv2_160164*	transposase
* * ÷	MBURv2	3907685–3907828	3908084	257–400	115 [114]	[114] 116	*[MBURv2_160578]–[MBURv2_170001]*	[MBURv2_160578], [MBURv2_170001]
* * ÷	MBURv2	3913037–3913043	3913143	101–107	125 [113]	[0] 183	*[MBURv2_180001]*	[MBURv2_180001]
* * ÷	MBURv2	3919334–3920026	3920114–3920071	46–781	117 [113]	[114] 118	*[MBURv2_180001]–[MBURv2_180002]*	[MBURv2_180001], [MBURv2_180002]
* * ÷	MBURv2	3939119–3939197	3939334–3939306	110–216	121 [108]	[114] 118	*[MBURv2_180009]–[MBURv2_190002]*	[MBURv2_180009], [MBURv2_180010], [MBURv2_190001], [MBURv2_190002]
* * ÷	MBURv2	4088265–4088333	4088434	102–170	116 [114]	[0] 773	*[MBURv2_190133]*	[MBURv2_190133]
* * ÷	MBURv2	4090655	4090819–4090755	101–165	1040 [0]	[113] 118	*MBURv2_200002/MBURv2_210001*	conserved protein of unknown function/protein of unknown function
* * ÷	MBURv2	4611954–4612178	4613338–4612575	398–1385	115 [114]	[114] 115	*[MBURv2_210490]–[MBURv2_210493]*	[MBURv2_210490], MBURv2_210491, MBURv2_210492, [MBURv2_210493]
* * ÷	MBURv2	4710530–4712611	4713681–4712713	103–3152	115 [113]	[112] 116	*[MBURv2_210584]–MBURv2_220001*	[MBURv2_210584], MBURv2_210585, MBURv2_210586, MBURv2_220001
* * ÷	MBURv2	4744116	4744548	433	115 [113]	[113] 116	*[ruvB]*	[ruvB]
* * ÷	MBURv2	4755115–4755142	4755674	533–560	136 [97]	[0] 260	*[MBURv2_230012]*	[MBURv2_230012]
* * ÷	MBURv2	4866712–4866866	4867422–4867095	230–711	115 [113]	[113] 115	*[MBURv2_240107]–[MBURv2_250001]*	[MBURv2_240107], [MBURv2_250001]
* * ÷	MBURv2	5012842–5013596	5015094–5014316	721–2253	117 [114]	[114] 116	*MBURv2_250123–MBURv2_260001*	MBURv2_250123, MBURv2_260001

Unassigned new junction evidence...

		seq id	position	reads (cov)	reads (cov)	score	skew	freq	annotation	gene	product
*	?	MBURv2	1 =	0 (0.000)	232 (0.820)	179/566	0.4	100%	intergenic (–/-115)	*–/MBURv2_10001*	–/protein of unknown function
	?	MBURv2	= 82731	0 (0.000)					coding (4563/4566 nt)	*MBURv2_10087*	conserved protein of unknown fur
*	?	MBURv2	82832 =	0 (0.000)	134 (0.700)	101/382	1.0	57.0%	intergenic (+98/-92)	*MBURv2_10087/waaA*	conserved protein of unknown function/3-deoxy-D-manno-octulo acid transferase
	?	MBURv2	4562967 =	300 (1.050)					intergenic (+222/-408)	*MBURv2_210430/MBURv2_210431*	protein of unknown function/cons protein of unknown function
*	?	MBURv2	613316 =	0 (0.000)	203 (0.870)	141/466	0.6	68.7%	intergenic (-1091/+123)	*MBURv2_30007/MBURv2_30008*	protein of unknown function/Coby acid ac-diamide synthase
	?	MBURv2	1705944 =	226 (0.790)					intergenic (+257/-137)	*MBURv2_60309/MBURv2_60310*	Glutathione peroxidase/protein of unknown function
*	?	MBURv2	819638 =	NA (NA)	283 (0.990)	208/568	0.1	96.9%	intergenic (+27/+28)	*MBURv2_30178/MBURv2_30179*	conserved protein of unknown function/protein of unknown funct
	?	MBURv2	= 935633	9 (0.030)					coding (1663/2583 nt)	*MBURv2_50104*	putative Sensor histidine kinase/response regulator
*	?	MBURv2	820062 =	0 (0.000)	141 (0.860)	116/328	0.2	67.0%	intergenic (-139/+413)	*MBURv2_30179/mbtH*	protein of unknown function/Prote MbtH
	?	MBURv2	4754855 =	139 (0.840)					coding (54/777 nt)	*MBURv2_230012*	transposase (fragment)
*	?	MBURv2	= 820832	0 (0.000)	162 (1.020)	118/316	0.1	100%	intergenic (-139/+319)	*mbtH/MBURv2_50001*	Protein MbtH/RNA polymerase, sigma-24 subunit, ECF subfamily
	?	MBURv2	= 3254366	NA (NA)					intergenic (-450/+554)	*MBURv2_150001/MBURv2_150002*	putative reverse transcriptase/pro of unknown function
*	?	MBURv2	884705 =	0 (0.000)	266 (0.930)	199/568	0.2	100%	coding (621/984 nt)	*rpoS*	RNA polymerase, sigma S (sigma factor
	?	MBURv2	= 945544	NA (NA)					intergenic (+745/-233)	*hpsi/MBURv2_50110*	Hexulose-6-phosphate synthase isomerase/putative transport prot (MFS superfamily)
*	?	MBURv2	= 884706	4 (0.010)	266 (0.940)	200/562	0.2	98.5%	coding (622/984 nt)	*rpoS*	RNA polymerase, sigma S (sigma factor
	?	MBURv2	945238 =	NA (NA)					intergenic (+439/-539)	*hpsi/MBURv2_50110*	Hexulose-6-phosphate synthase isomerase/putative transport prot (MFS superfamily)
	?	MBURv2	935626 =	11 (0.040)					coding (1656/2583 nt)	*MBURv2_50104*	putative Sensor histidine

*					195 (0.680)	155/568	0.9	94.7%			kinase/response regulator
	?	MBURv2	= 1479206	NA (NA)					intergenic (-67/-136)	*MBURv2_60098/ndh*	transposase (fragment)/NADH dehydrogenase
*	?	MBURv2	1055534 =	NA (NA)	307 (1.080)	241/568	0.0	66.9%	intergenic (+204/-928)	*MBURv2_50193/MBURv2_50194*	AAA ATPase central domain protein/conserved protein of unkn function
	?	MBURv2	= 2592714	152 (0.530)					coding (356/795 nt)	*MBURv2_130386*	protein of unknown function
*	?	MBURv2	= 1055842	NA (NA)	275 (0.960)	204/568	0.2	100%	intergenic (+512/-620)	*MBURv2_50193/MBURv2_50194*	AAA ATPase central domain protein/conserved protein of unkn function
	?	MBURv2	= 1353349	0 (0.000)					intergenic (-42/+96)	*MBURv2_50460/MBURv2_60001*	conserved protein of unknown function/protein of unknown funct
*	?	MBURv2	= 1521363	320 (1.120)	358 (1.290)	242/552	0.0	58.3%	intergenic (+43/+261)	*MBURv2_60143/MBURv2_60144*	conserved protein of unknown function/putative NAD-dependent epimerase/dehydratase
	?	MBURv2	2718373 =	202 (0.730)					intergenic (-158/-716)	*MBURv2_130515/MBURv2_130516*	conserved exported protein of unknown function/Retron-type rev transcriptase
*	?	MBURv2	1960975 =	0 (0.000)	435 (1.550)	226/560	0.0	100%	intergenic (+434/-27)	*MBURv2_80115/MBURv2_90001*	putative Glycosyl transferase fam 2/conserved protein of unknown function
	?	MBURv2	= 1967032	0 (0.000)					coding (102/111 nt)	*MBURv2_100003*	protein of unknown function
*	?	MBURv2	2077070 =	NA (NA)	235 (0.860)	172/542	0.4	100%	intergenic (-17/+30)	*MBURv2_120096/MBURv2_120097*	conserved protein of unknown function/transposase
	?	MBURv2	4755675 =	0 (0.000)					intergenic (+97/-666)	*MBURv2_230012/MBURv2_240001*	transposase (fragment)/conserve protein of unknown function
*	?	MBURv2	2227988 =	260 (0.910)	311 (1.100)	221/562	0.1	69.8%	intergenic (-32/-69)	*MBURv2_130069/yhhI*	conserved protein of unknown function/H repeat-associated prot YhhI
	?	MBURv2	3098491 =	12 (0.040)					coding (160/1404 nt)	*fumC*	fumarate hydratase (fumarase C),aerobic Class II
*	?	MBURv2	2401865 =	292 (1.020)	121 (0.720)	76/336	1.7	58.4%	intergenic (-180/+29)	*MBURv2_130208/MBURv2_130209*	CheW protein/protein of unknown function
	?	MBURv2	= 5067378	0 (0.000)					intergenic (+84/–)	*MBURv2_260042/–*	HemY domain protein/–
*	?	MBURv2	2742602 =	NA (NA)	305 (1.070)	223/568	0.1	93.3%	coding (3259/3300 nt)	*MBURv2_130533*	Putative Serine/threonine protein kinase (fragment)
	?	MBURv2	= 3098498	22 (0.080)					coding (167/1404 nt)	*fumC*	fumarate hydratase (fumarase C),aerobic Class II
*	?	MBURv2	= 3252479	0 (0.000)	174 (0.620)	106/558	2.8	52.4%	intergenic (-715/+187)	*MBURv2_140002/MBURv2_150001*	exported protein of unknown function/putative reverse transcrip
	?	MBURv2	3442789 =	316 (1.130)					intergenic (+768/+616)	*MBURv2_160166/MBURv2_160167*	conserved protein of unknown function/conserved protein of unk function
*	?	MBURv2	3435297 =	618 (2.170)	315 (1.110)	207/566	0.1	50.4%	coding (756/1581 nt)	*MBURv2_160159*	transposase
	?	MBURv2	4343881 =	5 (0.020)					coding (748/1017 nt)	*MBURv2_210215*	transposase (fragment)
*	?	MBURv2	3912891 =	272 (0.950)	650 (2.340)	247/554	0.0	63.8%	coding (143/489 nt)	*MBURv2_170005*	protein of unknown function
	?	MBURv2	= 3913670	485 (1.700)					coding (6340/6870 nt)	*MBURv2_180001*	protein of unknown function
*	?	MBURv2	4088435 =	0 (0.000)	764 (2.750)	172/554	0.5	100%	intergenic (+133/+118)	*MBURv2_190133/MBURv2_200001*	protein of unknown function/prote unknown function
	?	MBURv2	= 4090654	0 (0.000)					intergenic (+661/-631)	*MBURv2_200002/MBURv2_210001*	conserved protein of unknown function/protein of unknown funct
*	?	MBURv2	= 4090654	0 (0.000)	143 (0.520)	115/546	2.2	55.0%	intergenic (+661/-631)	*MBURv2_200002/MBURv2_210001*	conserved protein of unknown function/protein of unknown funct
	?	MBURv2	4095078 =	234 (0.850)					intergenic (+71/-266)	*MBURv2_210005/MBURv2_210006*	conserved protein of unknown function/General secretion pathwa protein E (Modular protein)