

SUPPLEMENTARY INFORMATION

The lytic transglycosylase MltB connects membrane homeostasis and *in vivo* fitness of *Acinetobacter baumannii*

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Fig. S1. Validation that Δkm mutant colonizes the bloodstream as well as AB0057.

CBA/J mice were infected with 10^7 CFU of either strain AB0057 or the *km* mutant (AB0057^{Km}) by tail vein injection. At 24 hpi, mice were sacrificed, spleen and liver were harvested, and the bacterial burden was determined by CFU enumeration on LB agar. Bacterial numbers are presented as the \log_{10} CFU g⁻¹ of tissue. Each data point represents a sample from an individual mouse, and horizontal bars indicate the median values. Statistical significance was calculated by the Mann-Whitney test. NS, not significant.

Fig. S2. Colonization of the neutropenic mouse model of bloodstream infection by the transposon pools.

Recovery of the transposon mutants (pools #1 to 5) during bloodstream infection. CBA/J mice were infected with 10^7 CFU of either wild type strain AB0057^{Km} (57) or the transposon pools (Tn pool #1 to #5) by tail vein injection. At 24 hpi, mice were sacrificed, spleen and liver were harvested, and the bacterial burden was determined by CFU enumeration on LB agar. Bacterial numbers are presented as the \log_{10} CFU g⁻¹ of tissue. Each data point represents a sample from an individual mouse, and horizontal bars indicate the median values.

Abbreviation: 57, WT (AB0057^{Km}); Tn, transposon.

Fig. S3. Capsule production in strain AB0057.

A. Maneval's staining of bacteria grown 24 h on LB-agar plate. Images are representative of three independent experiments.

B. Mucoviscosity measurement. Mucoviscosity was used as an indicator of capsule production by measuring optical density (OD₆₀₀) of the supernatant after centrifugation for 5 min at 1,000 × g from a starting turbidity of 1.0.

All results are the mean values and standard deviations of three independent

experiments. Statistical significance was calculated by One-way Anova and the Tukey's multiple comparisons test *, $P < 0.05$; ***, $P < 0.0005$; ****, $P < 0.0001$.

Abbreviation: 57, WT (AB0057^{Km}); compl.: complemented (pABBR_Km-*mrdB*-*mltB*).

Fig. S4. Stress resistance of the *mltB* mutant in strain AB5075.

Resistance to stresses was determined by incubating 10^7 CFU ml⁻¹ of the AB5075 (75) and its isogenic *mltB* mutant to different stressors.

A. Survival in 90% human active serum (HS) and growth in 90% heat-inactivated human serum (HI). The number of surviving CFUs was quantified by CFU enumeration on LB agar every hour. For ease of reading, standard deviations were removed.

B. Survival in the presence of 1 $\mu\text{g ml}^{-1}$ of polymyxin B. The number of CFUs recovered at 60 min (T_{60}) was determined by CFU enumeration on LB agar and compared to time 0 (T_0).

C. Survival to oxidative stress (2.5 mM H₂O₂). The percent survival to 2.5 mM H₂O₂ was determined by dividing the number of CFU recovered at 30 min (T_{30}) post-inoculation by the number of CFUs at time 0 (T_0).

D. Survival in an acidic environment. The percent survival to acidic environment was determined by dividing the number of CFU recovered at 60 min (T_{60}) post-inoculation in LB-pH5 by the number of CFUs recovered in LB-pH7.

E. Survival of osmotic shock. The number of CFUs recovered in LB containing 8.85 mM or 2.5 M NaCl at 2 h (T_2) post-inoculation was determined by CFU enumeration on LB agar.

Results are the mean values and standard deviations of three biological experiments. Statistical significance was calculated by the Student's *t*-test (*, $P < 0.05$; NS: not significant).

Fig. S5. Capsule production in strain AB5075.

A. Maneval's staining of bacteria grown 24 h on LB-agar plate. Images are representative of three independent experiments.

B. Mucoviscosity measurement. Mucoviscosity was used as an indicator of capsule production by measuring optical density (OD₆₀₀) of the supernatant after centrifugation for 5 min at 1,000 × *g* from a starting turbidity of 1.0.

All results are the mean values and standard deviations of three independent experiments. Statistical significance was calculated by the Student's *t*-test (***, *P*<0.001). Abbreviation: 75, WT (AB5075).

Fig. S6. Growth of strain AB0057 in presence of NaCl.

A-D. The WT strain (57) and its derivative strains were grown in presence of different concentration of NaCl. (A) 100 mM. (B) 250 mM. (C) 500 mM. (D) 750 mM. Results are the mean values and standard deviations of three biological experiments. For ease of reading, standard deviations were removed from graphs. Statistical significance was calculated by the two-way ANOVA and Tukey's multiple comparisons test (****, *P*<0.0001).

Abbreviations: 57, WT (AB0057^{Km}); eV, empty vector (pABBR_Km); compl.: complemented (pABBR_Km-*mrdB-mltB*).

Fig. S7. Growth of strain AB5075 in presence of NaCl.

The WT strain AB5075 (75) and its isogenic *mltB* mutant were grown in presence of different concentration of NaCl. (A) 500 mM. (B) 750 mM. (B) Results are the mean values and standard deviations of three biological experiments. For ease of reading, standard deviations were removed from graphs. Statistical significance was calculated by the two-way ANOVA. No differences were observed between the AB5075 and its isogenic *mltB* mutant.

Fig. S8. Adhesion of the *mltB* mutant of strain AB5075.

A. Biofilm formation in LB at 30°C for 24 h under static conditions. Crystal violet binding assay was used to monitor biofilm formation.

B. Adherence onto A549 lung epithelial cells.

Results are the mean values and standard deviations of three biological experiments.

Statistical significance was calculated by the Student's *t*-test (*, $P < 0.05$).

Abbreviation: 75: AB5075.

Fig. S9. Adhesion to polystyrene surface.

A. Adhesion of AB0057 to and its derivative strains to polystyrene.

B. Adhesion of AB5075 and its isogenic *mltB* mutant to polystyrene.

Results are the mean values and standard deviations of three biological experiments.

Statistical significance was calculated by the Student's *t*-test (NS: Not significant).

Abbreviation: 57: WT (AB0057^{Km}); eV: empty vector (pABBR_Km);

compl.: complemented (pABBR_Km-*mrdB-mltB*); 75: AB5075.

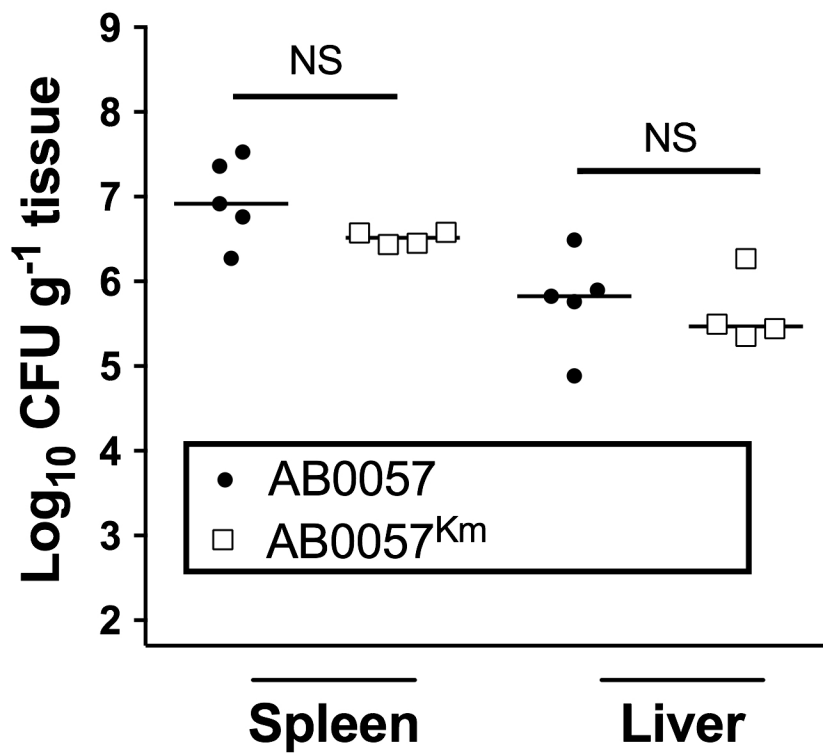


Fig. S1.

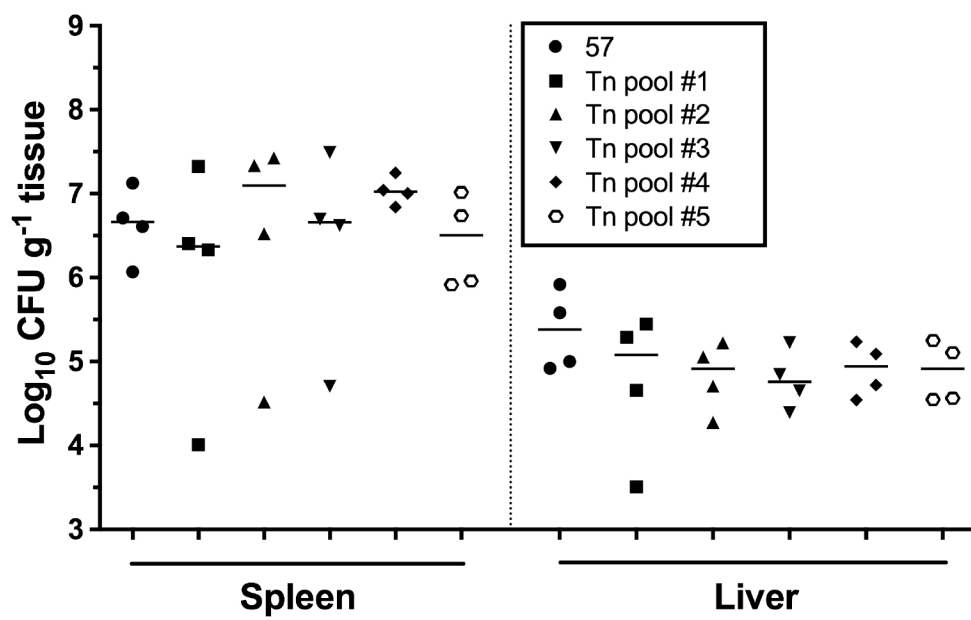


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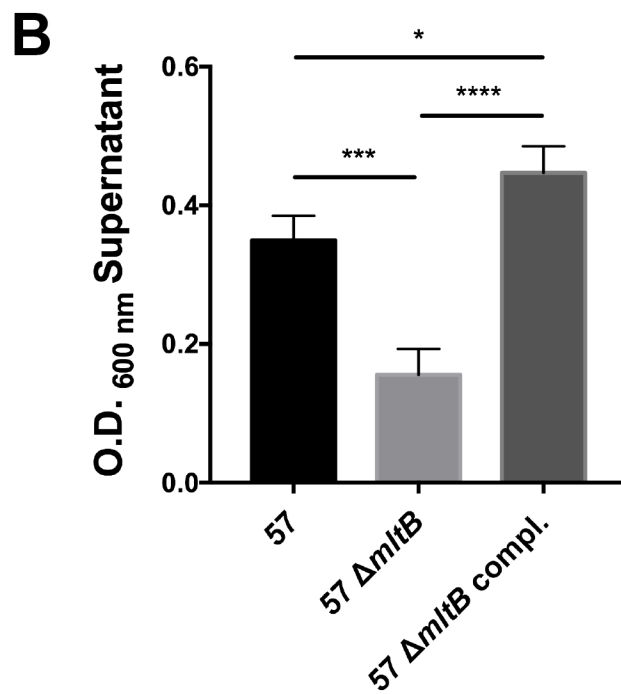
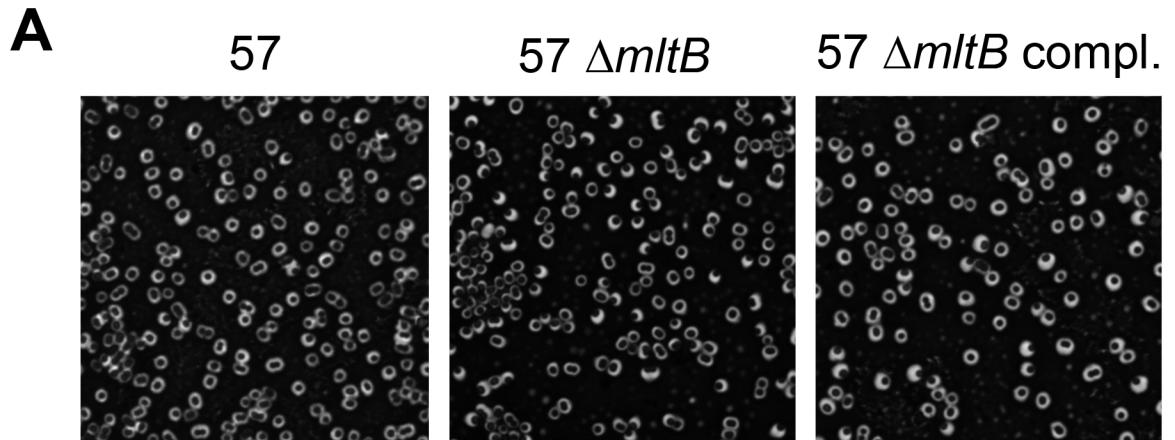


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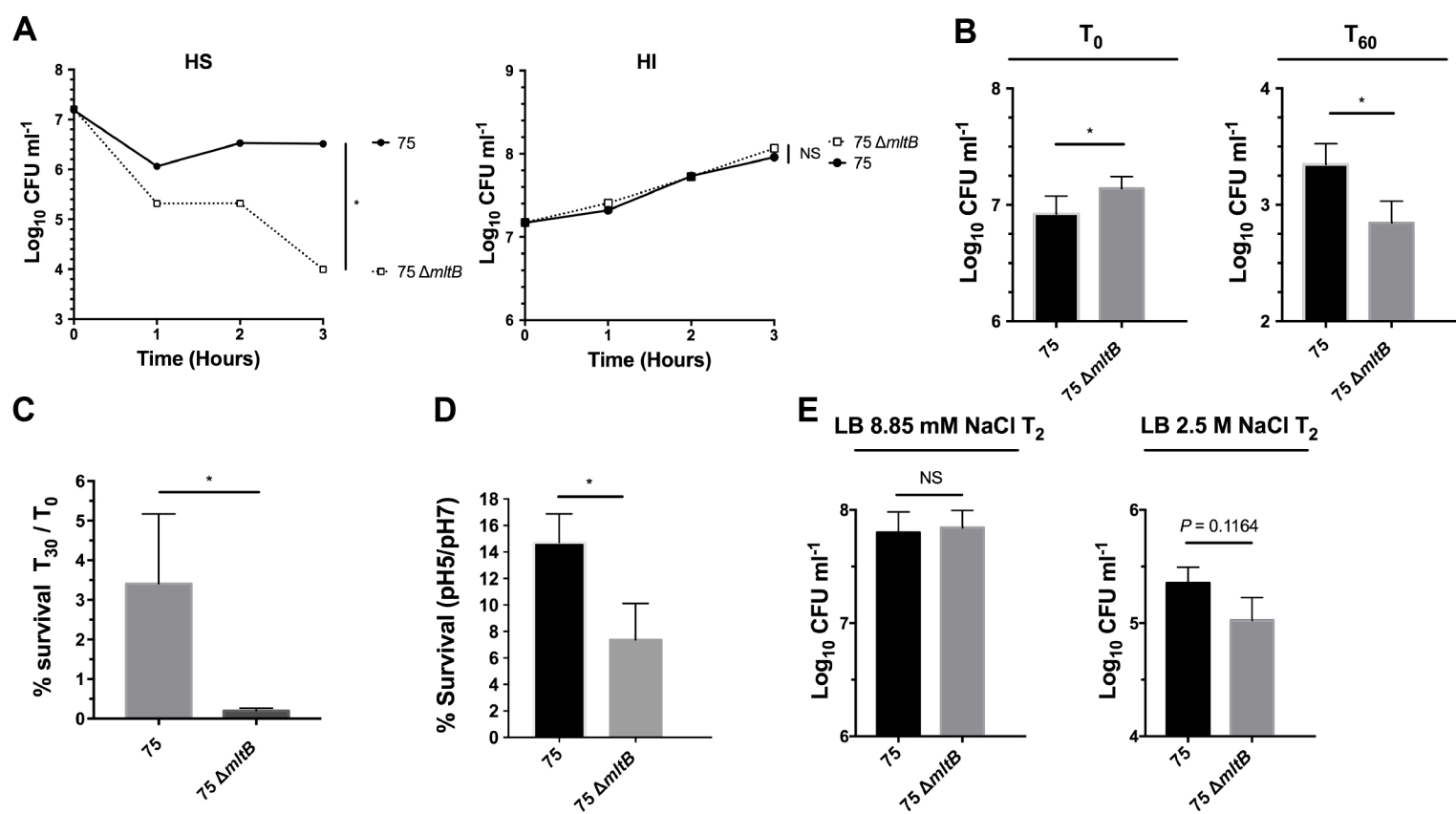


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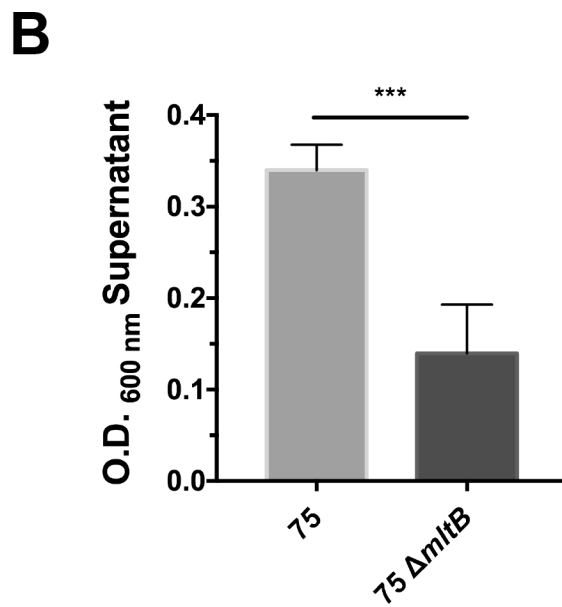
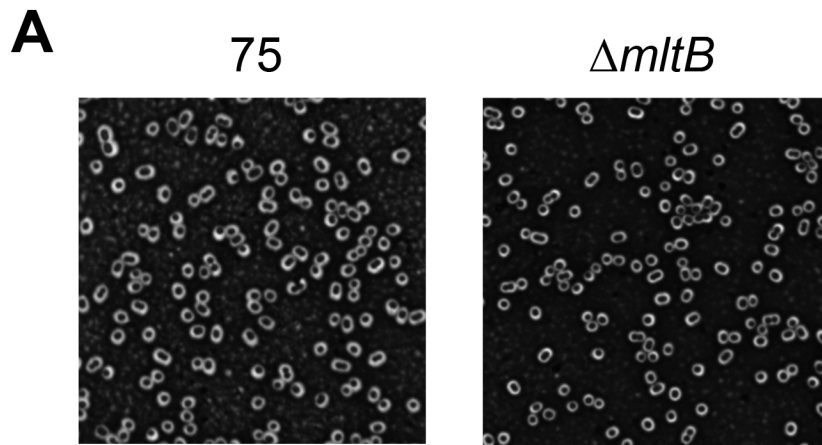


Fig. S5.

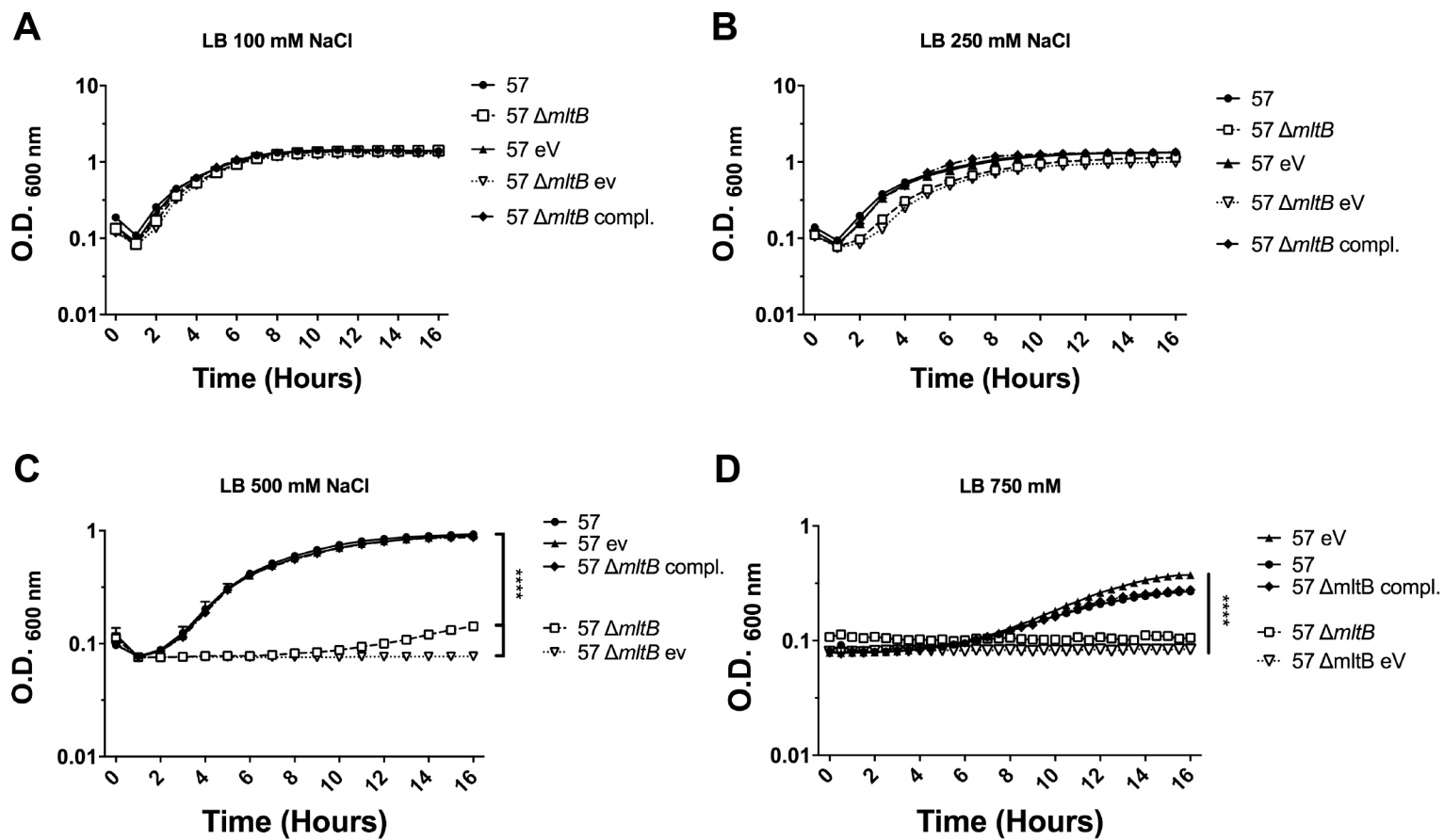


Fig. S6.

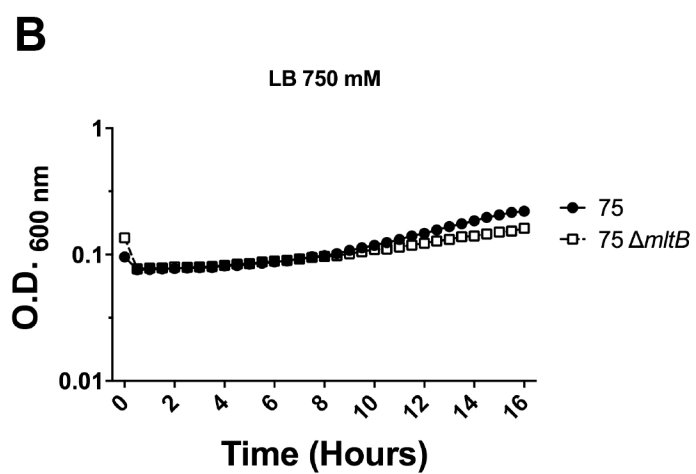
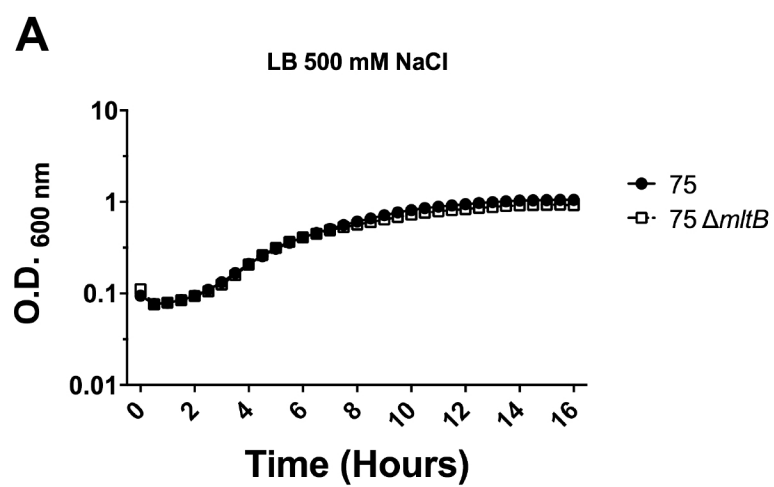


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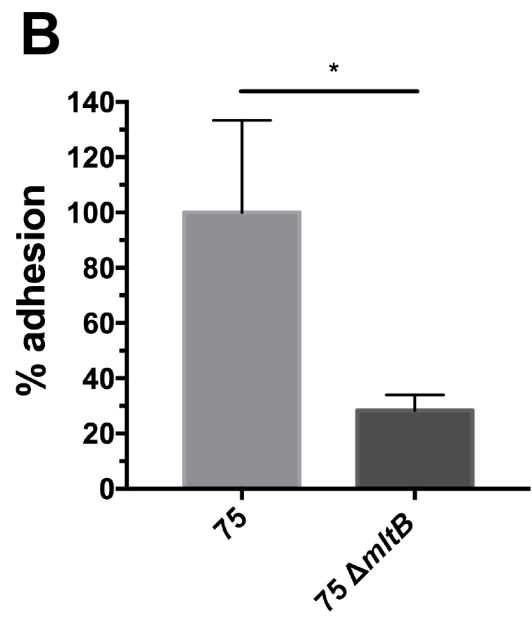
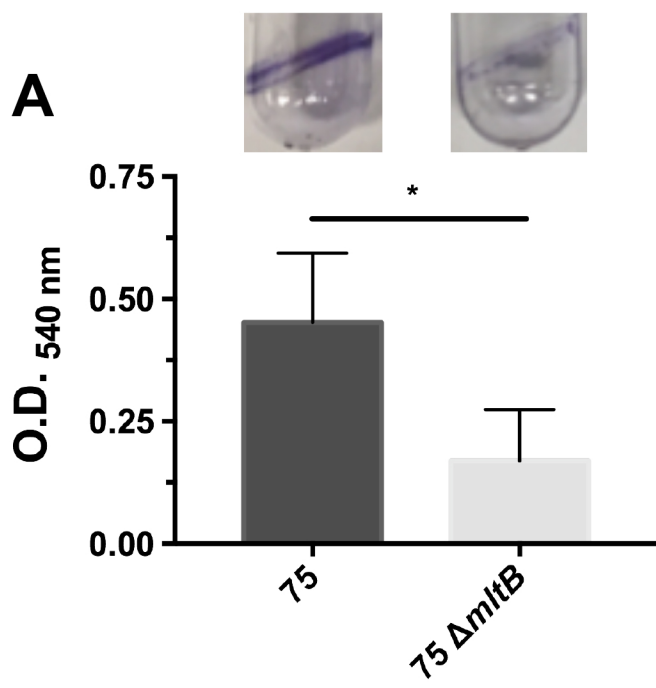


Fig. S8.

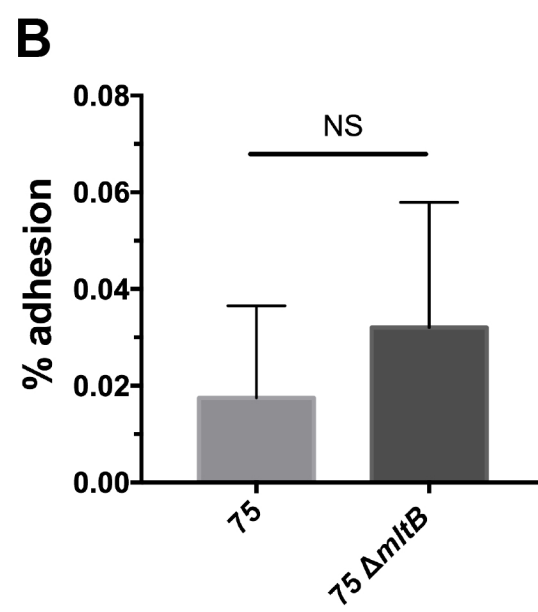
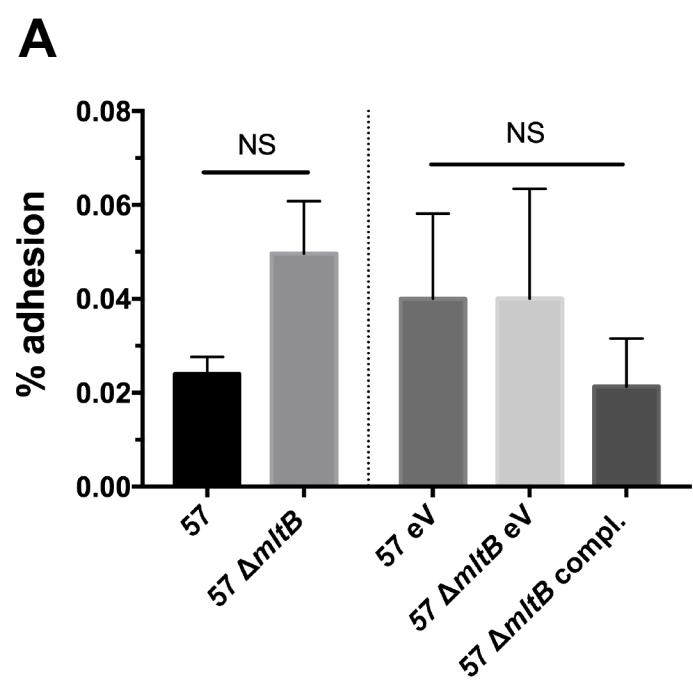


Fig. S9.

Table S1. Random distribution of the *Tn5* across the chromosome.

| Mutant # | Integration site | Mutant # | Integration site |
|-----------------|-------------------------|-----------------|--|
| 1 | <i>insB3</i> | 11 | <i>AB57_0054</i> |
| 2 | <i>macB</i> | 12 | <i>AB57_0519</i> |
| 3 | <i>tonB</i> | 13 | <i>AB57_3074</i> |
| 4 | <i>AB57_1775</i> | 14 | Intergenic region between <i>pilG</i> and <i>AB57_3316</i> |
| 5 | <i>esvE1</i> | 15 | <i>AB57_0369</i> |
| 6 | <i>AB57_0857</i> | 16 | <i>AB57_2259</i> |
| 7 | <i>aph1</i> | 17 | <i>AB57_3071</i> |
| 8 | <i>AB57_3085</i> | 18 | <i>AB57_3092</i> |
| 9 | <i>AB57_3329</i> | 19 | <i>AB57_0291</i> |
| 10 | <i>AB57_1502</i> | 20 | <i>AB57_0091</i> |

Sequencing approach was used to identify the integration site of selected *Tn5* mutants.

Table S2. Candidate fitness factors during bloodstream infection.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS16925 | AB57_3427 | sulfurtransferase | -10.82 | 0.0096 |
| AB57_RS03515 | AB57_0688 | 2-amino-4-hydroxy-6-hydroxymethyldihydropteridine pyrophosphokinase | -10.31 | 0.0078 |
| AB57_RS12865 | AB57_2595 | LuxR family transcriptional regulator | -10.18 | 0.0003 |
| AB57_RS16615 | AB57_3365 | hypothetical protein | -10.11 | 0.0034 |
| AB57_RS16225 | AB57_3288 | hypothetical protein | -9.66 | 0.0091 |
| AB57_RS00885 | AB57_0173 | hypothetical protein | -9.63 | 0.0052 |
| AB57_RS17590 | AB57_3551 | toluene tolerance protein | -9.60 | 0.0062 |
| AB57_RS16280 | AB57_3300 | hypothetical protein | -9.57 | 0.0001 |
| AB57_RS09270 | AB57_1860 | MULTISPECIES: DNA-binding protein HU-beta | -9.52 | 0.0044 |
| AB57_RS05825 | AB57_1154 | hypothetical protein | -9.48 | 0.0075 |
| AB57_RS14300 | AB57_2881 | diadenosine tetraphosphatase | -9.47 | 0.0031 |
| AB57_RS18240 | AB57_3680 | acetyltransferase | -9.37 | 0.0062 |
| AB57_RS13640 | AB57_2749 | lytic transglycosylase | -9.36 | 0.0000 |
| AB57_RS14220 | AB57_2865 | NUDIX hydrolase | -9.24 | 0.0022 |
| AB57_RS02755 | AB57_0530 | MULTISPECIES: 50S ribosomal protein L33 | -9.24 | 0.0001 |
| AB57_RS02760 | AB57_0531 | MULTISPECIES: 50S ribosomal protein L28 | -9.24 | 0.0001 |
| AB57_RS02405 | AB57_0459 | hypothetical protein | -9.21 | 0.0052 |
| AB57_RS01760 | AB57_0336 | glutamyl-Q tRNA(Asp) ligase | -9.17 | 0.0000 |
| AB57_RS19005 | AB57_3836 | phosphoserine phosphatase | -9.16 | 0.0035 |
| AB57_RS12520 | AB57_2526 | phosphoribosylamine--glycine ligase | -9.15 | 0.0001 |
| AB57_RS18340 | AB57_3700 | endonuclease | -9.15 | 0.0050 |
| AB57_RS18195 | | hypothetical protein | -9.09 | 0.0094 |
| AB57_RS13620 | AB57_2745 | membrane protein | -9.08 | 0.0000 |
| AB57_RS00880 | AB57_0172 | PadR family transcriptional regulator | -9.08 | 0.0094 |
| AB57_RS03540 | AB57_0693 | hypothetical protein | -9.07 | 0.0038 |
| AB57_RS08135 | AB57_1626 | allantoin permease | -9.07 | 0.0003 |
| AB57_RS19160 | AB57_3871 | 4-hydroxyphenylpyruvate dioxygenase | -9.07 | 0.0078 |
| AB57_RS00935 | AB57_0183 | MULTISPECIES: Fur family transcriptional regulator | -9.02 | 0.0043 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS04245 | AB57_0829 | methionine--tRNA ligase | -9.02 | 0.0012 |
| AB57_RS19435 | AB57_3924 | membrane protein | -9.00 | 0.0003 |
| AB57_RS00340 | AB57_0065 | TetR family transcriptional regulator | -8.97 | 0.0038 |
| AB57_RS01915 | AB57_4828 | tRNA-Trp | -8.97 | 0.0052 |
| AB57_RS11190 | AB57_2247 | N-acetyl-gamma-glutamyl-phosphate reductase | -8.95 | 0.0034 |
| AB57_RS03840 | AB57_0752 | hypothetical protein | -8.95 | 0.0094 |
| AB57_RS18715 | AB57_3777 | pyruvate dehydrogenase | -8.92 | 0.0019 |
| AB57_RS04780 | AB57_0935 | phosphomannomutase | -8.90 | 0.0084 |
| AB57_RS14655 | AB57_RS14655 | ribonuclease 3 | -8.90 | 0.0001 |
| AB57_RS18200 | AB57_3672 | heme ABC transporter ATP-binding protein | -8.86 | 0.0091 |
| AB57_RS08130 | AB57_1625 | MULTISPECIES: AsnC family transcriptional regulator | -8.86 | 0.0001 |
| AB57_RS19460 | AB57_3929 | hypothetical protein | -8.85 | 0.0002 |
| AB57_RS17790 | AB57_3593 | hypothetical protein | -8.85 | 0.0001 |
| AB57_RS17115 | AB57_3463 | TetR family transcriptional regulator | -8.84 | 0.0016 |
| AB57_RS19230 | AB57_3885 | dienelactone hydrolase | -8.84 | 0.0009 |
| AB57_RS05910 | AB57_1169 | MULTISPECIES: GntR family transcriptional regulator | -8.82 | 0.0000 |
| AB57_RS16190 | | rod shape-determining protein MreD | -8.81 | 0.0058 |
| AB57_RS02165 | AB57_0412 | preprotein translocase subunit SecG | -8.79 | 0.0023 |
| AB57_RS00085 | AB57_0017 | DNA gyrase subunit B | -8.79 | 0.0049 |
| AB57_RS14295 | AB57_2880 | 2,4-diaminobutyrate decarboxylase | -8.79 | 0.0013 |
| AB57_RS05665 | AB57_1125 | cobalt transporter | -8.78 | 0.0000 |
| AB57_RS00485 | AB57_0095 | NAD-dependent dehydratase | -8.78 | 0.0024 |
| AB57_RS14750 | AB57_2981 | 7-carboxy-7-deazaguanine synthase | -8.78 | 0.0091 |
| AB57_RS00750 | AB57_0148 | enoyl-CoA hydratase | -8.77 | 0.0024 |
| AB57_RS00830 | AB57_0162 | BolA family transcriptional regulator | -8.76 | 0.0000 |
| AB57_RS02660 | AB57_0511 | lauroyl acyltransferase | -8.76 | 0.0046 |
| AB57_RS16810 | AB57_3405 | LPS biosynthesis protein | -8.75 | 0.0027 |
| AB57_RS00210 | AB57_0043 | phosphoribosylaminoimidazole carboxylase | -8.74 | 0.0037 |
| AB57_RS15740 | AB57_3188 | hypothetical protein | -8.73 | 0.0030 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS17745 | AB57_3583 | succinylglutamate desuccinylase | -8.72 | 0.0000 |
| AB57_RS18725 | | hypothetical protein | -8.71 | 0.0003 |
| AB57_RS00220 | AB57_0045 | hypothetical protein | -8.69 | 0.0008 |
| AB57_RS03490 | AB57_0683 | GTP pyrophosphokinase | -8.68 | 0.0095 |
| AB57_RS00030 | AB57_0006 | RND transporter | -8.67 | 0.0020 |
| AB57_RS03280 | AB57_0637 | NAD(P)-binding enzyme | -8.66 | 0.0009 |
| AB57_RS19190 | AB57_3877 | hypothetical protein | -8.65 | 0.0013 |
| AB57_RS00255 | AB57_0052 | MULTISPECIES: hypothetical protein | -8.62 | 0.0000 |
| AB57_RS01690 | AB57_0322 | peptidase S41 | -8.61 | 0.0006 |
| AB57_RS16835 | AB57_3410 | glutamine-synthetase adenylyltransferase | -8.60 | 0.0021 |
| AB57_RS00010 | AB57_0002 | anhydro-N-acetylmuramic acid kinase | -8.60 | 0.0048 |
| AB57_RS02895 | AB57_0558 | universal stress protein A | -8.60 | 0.0008 |
| AB57_RS06935 | AB57_1373 | membrane protein | -8.58 | 0.0062 |
| AB57_RS17620 | AB57_3557 | inositol monophosphatase | -8.58 | 0.0051 |
| AB57_RS02385 | AB57_0454 | hypothetical protein | -8.57 | 0.0030 |
| AB57_RS16370 | AB57_3312 | chemotaxis protein | -8.56 | 0.0000 |
| AB57_RS13015 | AB57_2622 | thiamine biosynthesis protein ThiS | -8.55 | 0.0000 |
| AB57_RS11540 | AB57_2319 | ribosome-recycling factor | -8.53 | 0.0002 |
| AB57_RS02775 | AB57_0534 | formyltetrahydrofolate deformylase | -8.53 | 0.0015 |
| AB57_RS11345 | AB57_2279 | branched-chain amino acid transporter 2 carrier protein BrnQ | -8.53 | 0.0001 |
| AB57_RS01695 | AB57_0323 | two-component system response regulator | -8.52 | 0.0001 |
| AB57_RS04785 | AB57_0936 | acetylglutamate kinase | -8.52 | 0.0047 |
| AB57_RS01205 | AB57_0236 | pyrophosphatase | -8.51 | 0.0062 |
| AB57_RS00740 | AB57_0146 | AMP-binding protein | -8.49 | 0.0000 |
| AB57_RS11305 | AB57_2270 | hypothetical protein | -8.49 | 0.0034 |
| AB57_RS18225 | AB57_3677 | MULTISPECIES: DNA-binding response regulator | -8.48 | 0.0017 |
| AB57_RS17855 | AB57_3607 | D-3-phosphoglycerate dehydrogenase | -8.48 | 0.0027 |
| AB57_RS08750 | AB57_1755 | DNA-binding protein | -8.48 | 0.0002 |
| AB57_RS00215 | AB57_0044 | lytic transglycosylase | -8.47 | 0.0000 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS00420 | AB57_0081 | fatty acid desaturase | -8.46 | 0.0062 |
| AB57_RS09275 | AB57_1861 | peptidylprolyl isomerase | -8.45 | 0.0019 |
| AB57_RS11480 | AB57_2307 | DNA recombination/repair protein RecA | -8.43 | 0.0044 |
| AB57_RS19235 | AB57_3886 | hydrolase | -8.42 | 0.0076 |
| AB57_RS12950 | | colicin V biosynthesis protein | -8.42 | 0.0059 |
| AB57_RS01375 | | MULTISPECIES: transcriptional regulator | -8.41 | 0.0000 |
| AB57_RS02120 | AB57_0403 | hypothetical protein | -8.41 | 0.0000 |
| AB57_RS18840 | AB57_3803 | 5-hydroxyisourate hydrolase | -8.40 | 0.0031 |
| AB57_RS09090 | AB57_1824 | hypothetical protein | -8.40 | 0.0000 |
| AB57_RS16195 | AB57_3282 | rod shape-determining protein MreC | -8.39 | 0.0000 |
| AB57_RS15870 | AB57_3216 | hypothetical protein | -8.39 | 0.0075 |
| AB57_RS00930 | AB57_0182 | zinc ABC transporter ATP-binding protein | -8.38 | 0.0007 |
| AB57_RS00015 | AB57_0003 | hydrolase | -8.38 | 0.0034 |
| GenBank:repeat_r egion:NC_011586: 315546:317142 | | | -8.36 | 0.0030 |
| AB57_RS08335 | AB57_1669 | hypothetical protein | -8.36 | 0.0075 |
| AB57_RS13805 | AB57_2778 | MULTISPECIES: hypothetical protein | -8.35 | 0.0003 |
| AB57_RS06615 | AB57_1310 | integrase | -8.35 | 0.0002 |
| AB57_RS08475 | AB57_1697 | L-asparaginase | -8.35 | 0.0032 |
| AB57_RS03635 | AB57_0712 | tRNA(Ile)-lysidine synthase | -8.34 | 0.0003 |
| AB57_RS15230 | AB57_3083 | 3',5'-cyclic-nucleotide phosphodiesterase | -8.34 | 0.0000 |
| AB57_RS16760 | AB57_3395 | phospholipase D | -8.34 | 0.0000 |
| AB57_RS16360 | AB57_3310 | hypothetical protein | -8.33 | 0.0046 |
| AB57_RS02255 | AB57_0427 | 2-octaprenylphenol hydroxylase | -8.33 | 0.0037 |
| AB57_RS00070 | AB57_0014 | glutathione ABC transporter ATP-binding protein | -8.32 | 0.0020 |
| AB57_RS15470 | AB57_3133 | hypothetical protein | -8.32 | 0.0003 |
| AB57_RS15265 | AB57_3090 | methyltransferase | -8.29 | 0.0001 |
| AB57_RS00415 | AB57_0080 | oxidoreductase | -8.29 | 0.0011 |
| AB57_RS00185 | AB57_0038 | hypothetical protein | -8.29 | 0.0020 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS02510 | AB57_0480 | hypothetical protein | -8.28 | 0.0002 |
| AB57_RS01380 | AB57_0271 | MULTISPECIES: mercuric reductase | -8.28 | 0.0027 |
| AB57_RS16720 | AB57_3387 | DNA-binding response regulator | -8.28 | 0.0030 |
| AB57_RS17985 | AB57_3633 | hypothetical protein | -8.28 | 0.0029 |
| AB57_RS01565 | AB57_0297 | DNA invertase Pin | -8.27 | 0.0000 |
| AB57_RS04405 | AB57_0863 | SMC-Scp complex subunit ScpB | -8.27 | 0.0050 |
| AB57_RS17585 | AB57_3549 | MULTISPECIES: toluene-tolerance protein (Ttg2E) | -8.27 | 0.0000 |
| AB57_RS00840 | AB57_0164 | chromosome partitioning protein ParA | -8.26 | 0.0033 |
| AB57_RS05145 | AB57_1012 | hypothetical protein | -8.26 | 0.0002 |
| AB57_RS13735 | AB57_2767 | adenosylhomocysteinase | -8.25 | 0.0029 |
| AB57_RS01775 | AB57_0339 | NUDIX hydrolase | -8.25 | 0.0001 |
| AB57_RS00165 | AB57_0034 | hypothetical protein | -8.25 | 0.0012 |
| GenBank:repeat_r egion:NC_011586: 2653756:2653874 | | | -8.24 | 0.0030 |
| AB57_RS04255 | AB57_0831 | ATP-binding protein | -8.24 | 0.0001 |
| AB57_RS03525 | AB57_0690 | pantothenate synthetase | -8.24 | 0.0009 |
| AB57_RS02085 | AB57_0396 | transcriptional regulator | -8.24 | 0.0051 |
| AB57_RS01390 | | MULTISPECIES: hypothetical protein | -8.24 | 0.0044 |
| AB57_RS15310 | AB57_3096 | MULTISPECIES: ATP-dependent metalloprotease | -8.23 | 0.0094 |
| AB57_RS15620 | AB57_3163 | hypothetical protein | -8.22 | 0.0003 |
| AB57_RS14380 | AB57_2904 | type I-F CRISPR-associated protein Csy1 | -8.22 | 0.0000 |
| AB57_RS12490 | AB57_2520 | tRNA uridine 5-carboxymethylaminomethyl modification enzyme MnmG | -8.21 | 0.0000 |
| AB57_RS15415 | | hypothetical protein | -8.20 | 0.0001 |
| AB57_RS18920 | AB57_3819 | glutamate--cysteine ligase | -8.19 | 0.0006 |
| AB57_RS02575 | AB57_0493 | membrane protein | -8.19 | 0.0003 |
| AB57_RS05115 | AB57_1006 | MULTISPECIES: choline transporter BetT | -8.19 | 0.0018 |
| AB57_RS19025 | AB57_3840 | N utilization substance protein B homolog | -8.18 | 0.0027 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS18720 | AB57_3778 | peptidase M23 | -8.18 | 0.0000 |
| AB57_RS03155 | AB57_0611 | MULTISPECIES: glutaredoxin | -8.18 | 0.0029 |
| AB57_RS01295 | AB57_0255 | MULTISPECIES: arsenate reductase | -8.17 | 0.0002 |
| AB57_RS19065 | AB57_3852 | hypothetical protein | -8.17 | 0.0044 |
| AB57_RS02060 | AB57_0391 | UvrABC system protein C | -8.17 | 0.0015 |
| AB57_RS14820 | AB57_2995 | Holliday junction DNA helicase RuvB | -8.16 | 0.0035 |
| AB57_RS01665 | AB57_0317 | DNA polymerase III subunit chi | -8.16 | 0.0039 |
| AB57_RS19145 | AB57_3868 | maleylacetoacetate isomerase | -8.15 | 0.0000 |
| AB57_RS17035 | AB57_3446 | MULTISPECIES: L-rhamnonate dehydratase | -8.15 | 0.0083 |
| AB57_RS04905 | AB57_0960 | MULTISPECIES: hypothetical protein | -8.15 | 0.0000 |
| AB57_RS03530 | AB57_0691 | glmZ(sRNA)-inactivating NTPase | -8.13 | 0.0037 |
| AB57_RS02145 | AB57_0408 | N-methyltransferase | -8.13 | 0.0008 |
| AB57_RS04205 | AB57_0821 | MULTISPECIES: hypothetical protein | -8.13 | 0.0030 |
| AB57_RS16120 | AB57_3266 | oxidoreductase | -8.13 | 0.0015 |
| AB57_RS04170 | AB57_0814 | cold-shock protein | -8.12 | 0.0002 |
| AB57_RS01370 | | MULTISPECIES: mercury resistance protein | -8.11 | 0.0037 |
| AB57_RS07340 | AB57_1460 | hypothetical protein | -8.10 | 0.0002 |
| AB57_RS06905 | AB57_1367 | cold-shock protein | -8.10 | 0.0002 |
| AB57_RS14015 | AB57_2822 | acinetobactin utilization protein | -8.10 | 0.0008 |
| AB57_RS16800 | AB57_3404 | hypothetical protein | -8.10 | 0.0040 |
| AB57_RS10150 | AB57_2035 | hypothetical protein | -8.09 | 0.0052 |
| AB57_RS06010 | AB57_1189 | MULTISPECIES: hypothetical protein, partial | -8.09 | 0.0003 |
| AB57_RS08950 | AB57_1795 | cobyric acid synthase CobQ | -8.09 | 0.0001 |
| AB57_RS14245 | AB57_2870 | surface adhesion protein | -8.09 | 0.0055 |
| AB57_RS14230 | AB57_2867 | MULTISPECIES: High frequency lysogenization protein HflD homolog | -8.08 | 0.0034 |
| AB57_RS00645 | AB57_0128 | hypothetical protein | -8.08 | 0.0000 |
| AB57_RS00390 | AB57_0075 | MULTISPECIES: phosphoglycolate phosphatase | -8.08 | 0.0000 |
| AB57_RS17850 | AB57_3606 | FAD-linked oxidase | -8.08 | 0.0031 |
| AB57_RS15240 | AB57_3085 | phosphoenolpyruvate carboxykinase | -8.05 | 0.0004 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|--|---|----------------|
| AB57_RS01875 | AB57_0359 | pseudogene | -8.05 | 0.0039 |
| AB57_RS05150 | AB57_1013 | peptidylprolyl isomerase | -8.05 | 0.0012 |
| AB57_RS12555 | AB57_2533 | TetR family transcriptional regulator | -8.05 | 0.0075 |
| AB57_RS15525 | AB57_3144 | HxlR family transcriptional regulator | -8.04 | 0.0001 |
| AB57_RS08995 | AB57_1805 | deoxyribonuclease | -8.04 | 0.0000 |
| AB57_RS14185 | AB57_2858 | LysR family transcriptional regulator | -8.04 | 0.0016 |
| AB57_RS18730 | AB57_RS18730 | pseudogene | -8.03 | 0.0031 |
| AB57_RS09025 | AB57_1811 | 6-carboxy-5,6,7,8-tetrahydropterin synthase | -8.03 | 0.0075 |
| AB57_RS04765 | AB57_0932 | outer membrane protein | -8.03 | 0.0001 |
| AB57_RS13090 | AB57_2639 | pseudogene | -8.02 | 0.0048 |
| AB57_RS01645 | AB57_0313 | MULTISPECIES: restriction endonuclease subunit M | -8.02 | 0.0040 |
| AB57_RS00585 | AB57_0116 | L-lactate permease | -8.01 | 0.0018 |
| AB57_RS19470 | | alcohol dehydrogenase | -8.01 | 0.0049 |
| AB57_RS03470 | AB57_RS03470 | pseudogene | -8.00 | 0.0075 |
| AB57_RS05550 | AB57_1099 | methyltransferase | -8.00 | 0.0000 |
| AB57_RS03450 | AB57_0674 | MULTISPECIES: ribosome silencing factor RsfS | -8.00 | 0.0013 |
| AB57_RS07335 | AB57_1459 | hypothetical protein | -8.00 | 0.0094 |
| AB57_RS02355 | AB57_0448 | type II secretion system protein GspG | -7.99 | 0.0025 |
| AB57_RS19380 | AB57_3917 | membrane protein | -7.99 | 0.0094 |
| AB57_RS03610 | AB57_0707 | hypothetical protein | -7.99 | 0.0002 |
| AB57_RS16805 | AB57_3403 | glycosyl transferase | -7.99 | 0.0017 |
| GenBank:repeat_r egion:NC_011586: 383486:384688 | | | -7.98 | 0.0048 |
| AB57_RS14650 | AB57_2960 | GTPase Era | -7.98 | 0.0060 |
| AB57_RS12195 | AB57_2458 | membrane protein | -7.97 | 0.0030 |
| AB57_RS19280 | AB57_3896 | 2,5-diketo-D-gluconic acid reductase | -7.97 | 0.0053 |
| AB57_RS08620 | AB57_1728 | amino acid ABC transporter ATP-binding protein | -7.97 | 0.0030 |
| AB57_RS01910 | AB57_0362 | elongation factor Tu | -7.97 | 0.0048 |
| AB57_RS11155 | AB57_2240 | hypothetical protein | -7.97 | 0.0008 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS03250 | AB57_0630 | hypothetical protein | -7.97 | 0.0000 |
| AB57_RS01570 | | MULTISPECIES: hypothetical protein | -7.97 | 0.0018 |
| AB57_RS03845 | AB57_0753 | MULTISPECIES: D-ribulose-5-phosphate 3-epimerase | -7.97 | 0.0008 |
| AB57_RS01195 | AB57_0234 | membrane protein | -7.95 | 0.0017 |
| AB57_RS18465 | AB57_3725 | MULTISPECIES: hypothetical protein | -7.94 | 0.0001 |
| AB57_RS19385 | AB57_3918 | glutathione S-transferase | -7.94 | 0.0008 |
| AB57_RS17595 | AB57_3552 | mammalian cell entry protein | -7.94 | 0.0009 |
| AB57_RS01920 | AB57_0363 | preprotein translocase subunit SecE | -7.93 | 0.0030 |
| AB57_RS00315 | AB57_0060 | MULTISPECIES: lipoprotein signal peptidase | -7.93 | 0.0038 |
| AB57_RS16200 | AB57_3283 | MULTISPECIES: rod shape-determining protein MreB | -7.93 | 0.0010 |
| AB57_RS02905 | AB57_0559 | MULTISPECIES: hypothetical protein | -7.93 | 0.0008 |
| AB57_RS03070 | AB57_0594 | hypothetical protein | -7.93 | 0.0044 |
| AB57_RS05490 | AB57_1086 | MULTISPECIES: LysR family transcriptional regulator | -7.91 | 0.0027 |
| AB57_RS16115 | AB57_3265 | molecular chaperone | -7.91 | 0.0044 |
| AB57_RS03645 | AB57_0714 | MULTISPECIES: membrane protein | -7.91 | 0.0017 |
| AB57_RS17740 | AB57_3582 | hypothetical protein | -7.91 | 0.0027 |
| AB57_RS00335 | AB57_0064 | 5'-nucleosidase | -7.90 | 0.0038 |
| AB57_RS18305 | AB57_3692 | homoserine kinase | -7.90 | 0.0052 |
| AB57_RS01100 | AB57_0213 | cysteine synthase A | -7.90 | 0.0000 |
| AB57_RS00755 | AB57_0149 | enol-CoA hydratase | -7.89 | 0.0035 |
| AB57_RS01165 | AB57_0228 | membrane protein | -7.89 | 0.0000 |
| AB57_RS17605 | AB57_3554 | ABC transporter ATP-binding protein | -7.89 | 0.0004 |
| AB57_RS02900 | | MULTISPECIES: hypothetical protein | -7.89 | 0.0006 |
| AB57_RS14970 | AB57_3029 | RND transporter | -7.88 | 0.0094 |
| AB57_RS09630 | AB57_1934 | MULTISPECIES: hypothetical protein | -7.88 | 0.0000 |
| AB57_RS06170 | AB57_1221 | hypothetical protein | -7.87 | 0.0001 |
| AB57_RS01660 | AB57_0316 | hypothetical protein | -7.87 | 0.0046 |
| AB57_RS02795 | AB57_0538 | peptide-methionine (S)-S-oxide reductase | -7.87 | 0.0001 |
| AB57_RS18180 | AB57_3668 | RND transporter | -7.87 | 0.0034 |
| AB57_RS01620 | AB57_0308 | hypothetical protein | -7.86 | 0.0002 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS18030 | AB57_3642 | 3-dehydroquinase synthase | -7.86 | 0.0032 |
| AB57_RS18945 | AB57_3824 | MATE family efflux transporter | -7.86 | 0.0025 |
| AB57_RS13875 | AB57_2792 | xanthine dehydrogenase | -7.86 | 0.0003 |
| AB57_RS16575 | AB57_3356 | 3-ketoacyl-CoA thiolase | -7.86 | 0.0000 |
| AB57_RS15140 | AB57_3065 | hypothetical protein | -7.86 | 0.0016 |
| AB57_RS16520 | AB57_3344 | membrane protein | -7.84 | 0.0024 |
| AB57_RS00810 | AB57_0160 | hypothetical protein | -7.84 | 0.0041 |
| AB57_RS04860 | AB57_0951 | hypothetical protein | -7.84 | 0.0003 |
| AB57_RS08600 | AB57_1724 | glutathione S-transferase | -7.82 | 0.0031 |
| AB57_RS14805 | AB57_2992 | phosphoribosylformylglycinamide synthase | -7.82 | 0.0046 |
| AB57_RS14670 | AB57_2964 | elongation factor 4 | -7.82 | 0.0002 |
| AB57_RS02360 | AB57_0449 | hypothetical protein | -7.82 | 0.0002 |
| AB57_RS05460 | AB57_1080 | MULTISPECIES: sulfate adenylyltransferase subunit 2 | -7.81 | 0.0026 |
| AB57_RS15345 | AB57_3103 | transcription elongation factor GreA | -7.81 | 0.0034 |
| AB57_RS00330 | AB57_0063 | C4-dicarboxylate ABC transporter | -7.81 | 0.0003 |
| AB57_RS18710 | AB57_3776 | dihydrolipoamide acetyltransferase | -7.80 | 0.0015 |
| AB57_RS14385 | AB57_2905 | type I-F CRISPR-associated helicase Cas3 | -7.80 | 0.0094 |
| AB57_RS03650 | AB57_0715 | DNA polymerase I | -7.79 | 0.0036 |
| AB57_RS04080 | AB57_0796 | MULTISPECIES: DNA-binding response regulator | -7.79 | 0.0080 |
| AB57_RS14180 | AB57_2857 | ATP-dependent protease | -7.79 | 0.0040 |
| AB57_RS02340 | AB57_0445 | MULTISPECIES: potassium transporter | -7.78 | 0.0000 |
| AB57_RS16785 | AB57_3400 | glycosyl transferase | -7.78 | 0.0027 |
| GenBank:repeat_r egion:NC_011586: 308009:309004 | | | -7.77 | 0.0036 |
| AB57_RS18940 | AB57_3823 | aminotransferase | -7.77 | 0.0034 |
| AB57_RS08520 | AB57_1706 | phosphoribosylglycinamide formyltransferase | -7.77 | 0.0005 |
| AB57_RS12945 | AB57_2607 | amidophosphoribosyltransferase | -7.76 | 0.0013 |
| AB57_RS14215 | AB57_2864 | hypothetical protein | -7.76 | 0.0000 |
| AB57_RS17825 | AB57_3600 | multidrug transporter | -7.75 | 0.0018 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS12920 | AB57_2602 | GatB/Yqey domain protein | -7.75 | 0.0001 |
| AB57_RS13780 | AB57_2776 | Nif3-like dinuclear metal center hexameric protein | -7.75 | 0.0075 |
| AB57_RS03705 | AB57_0727 | hypothetical protein | -7.75 | 0.0008 |
| AB57_RS12515 | AB57_2525 | purine biosynthesis protein purH | -7.75 | 0.0031 |
| AB57_RS03085 | AB57_0597 | glycosyl transferase | -7.73 | 0.0002 |
| AB57_RS06015 | AB57_1190 | hypothetical protein | -7.73 | 0.0002 |
| AB57_RS03160 | AB57_0612 | MULTISPECIES: rhodanese | -7.73 | 0.0010 |
| AB57_RS15300 | AB57_3094 | hypothetical protein | -7.73 | 0.0030 |
| AB57_RS03285 | AB57_0639 | LysR family transcriptional regulator | -7.73 | 0.0038 |
| AB57_RS00690 | AB57_0136 | hypothetical protein | -7.71 | 0.0000 |
| AB57_RS17510 | AB57_3534 | ATP-binding protein | -7.71 | 0.0001 |
| AB57_RS03510 | AB57_0687 | poly(A) polymerase | -7.71 | 0.0091 |
| AB57_RS17910 | AB57_3618 | MULTISPECIES: 30S ribosomal protein S16 | -7.71 | 0.0049 |
| AB57_RS06030 | AB57_1193 | feruloyl-CoA synthase | -7.70 | 0.0011 |
| AB57_RS12145 | AB57_2448 | DNA mismatch repair protein MutL | -7.70 | 0.0009 |
| AB57_RS19080 | AB57_3855 | hypothetical protein | -7.69 | 0.0044 |
| AB57_RS13715 | AB57_2763 | lipoyl synthase | -7.69 | 0.0015 |
| AB57_RS07430 | AB57_1480 | type VI secretion protein EvpB | -7.69 | 0.0008 |
| AB57_RS14645 | | hypothetical protein | -7.68 | 0.0046 |
| AB57_RS01220 | AB57_0240 | dihydrodipicolinate synthase | -7.68 | 0.0000 |
| AB57_RS07030 | AB57_1393 | hypothetical protein | -7.67 | 0.0030 |
| AB57_RS00615 | AB57_0122 | MULTISPECIES: 2-methylisocitrate lyase | -7.67 | 0.0038 |
| AB57_RS04250 | AB57_0830 | hypothetical protein | -7.67 | 0.0002 |
| AB57_RS18680 | AB57_3770 | inosine-5-monophosphate dehydrogenase | -7.67 | 0.0065 |
| AB57_RS16350 | AB57_3308 | lysophospholipase | -7.66 | 0.0015 |
| AB57_RS02125 | AB57_0404 | pseudogene | -7.66 | 0.0019 |
| AB57_RS11785 | AB57_2372 | phosphotyrosine protein phosphatase | -7.65 | 0.0030 |
| AB57_RS11775 | AB57_2370 | hypothetical protein | -7.65 | 0.0002 |
| AB57_RS03170 | AB57_0614 | MULTISPECIES: oligoribonuclease | -7.65 | 0.0055 |
| AB57_RS00925 | AB57_0181 | DNA repair protein HhH-GPD | -7.64 | 0.0026 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS02110 | AB57_0401 | MULTISPECIES: hypothetical protein | -7.64 | 0.0022 |
| AB57_RS14910 | AB57_3014 | MULTISPECIES: phosphoribosylformylglycinamide cyclo- ligase | -7.63 | 0.0011 |
| AB57_RS00205 | AB57_0042 | MULTISPECIES: phosphoribosylaminoimidazole carboxylase | -7.63 | 0.0026 |
| AB57_RS16955 | AB57_3434 | oxidoreductase | -7.63 | 0.0012 |
| AB57_RS19195 | AB57_3878 | MULTISPECIES: phosphoribosylaminoimidazole- succinocarboxamide synthase | -7.62 | 0.0031 |
| AB57_RS02690 | AB57_0517 | hypothetical protein | -7.62 | 0.0001 |
| AB57_RS01270 | AB57_0250 | MULTISPECIES: thioredoxin reductase | -7.61 | 0.0011 |
| AB57_RS18625 | AB57_3758 | hypothetical protein | -7.61 | 0.0008 |
| AB57_RS16590 | AB57_3359 | patatin family protein | -7.61 | 0.0075 |
| AB57_RS03150 | AB57_0610 | protein-export protein SecB | -7.61 | 0.0042 |
| AB57_RS00720 | AB57_0142 | alanine glycine permease | -7.60 | 0.0035 |
| AB57_RS01700 | AB57_0324 | histidine kinase | -7.59 | 0.0032 |
| AB57_RS01385 | AB57_0272 | MULTISPECIES: mercury transporter MerC | -7.59 | 0.0028 |
| AB57_RS14710 | AB57_2972 | ABC transporter permease | -7.59 | 0.0003 |
| AB57_RS05185 | AB57_1020 | MULTISPECIES: IclR family transcriptional regulator | -7.59 | 0.0003 |
| AB57_RS00775 | AB57_0153 | LuxR family transcriptional regulator | -7.58 | 0.0020 |
| AB57_RS01535 | AB57_0293 | MULTISPECIES: ANT(3 ^{IV})-Ia family aminoglycoside nucleotidyltransferase AadA1 | -7.58 | 0.0026 |
| AB57_RS05260 | AB57_1035 | MFS transporter permease | -7.57 | 0.0003 |
| AB57_RS07540 | AB57_1502 | hypothetical protein | -7.56 | 0.0022 |
| AB57_RS00680 | AB57_0134 | polyketide cyclase | -7.56 | 0.0011 |
| AB57_RS02470 | AB57_0472 | MATE family efflux transporter | -7.54 | 0.0021 |
| AB57_RS00425 | AB57_0082 | MULTISPECIES: ribonuclease PH | -7.53 | 0.0001 |
| AB57_RS00785 | AB57_0155 | acyl-CoA dehydrogenase | -7.53 | 0.0000 |
| AB57_RS03825 | AB57_0749 | methyltransferase | -7.52 | 0.0001 |
| AB57_RS03695 | AB57_0725 | MULTISPECIES: anti-anti-sigma factor | -7.52 | 0.0009 |
| AB57_RS17010 | AB57_3441 | phosphohydrolase | -7.51 | 0.0003 |
| AB57_RS14205 | AB57_2862 | hypothetical protein | -7.51 | 0.0001 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS03505 | AB57_0686 | transporter | -7.51 | 0.0001 |
| AB57_RS00080 | AB57_0016 | cytochrome B | -7.50 | 0.0031 |
| AB57_RS18175 | AB57_3667 | hemolysin D | -7.50 | 0.0006 |
| AB57_RS03425 | AB57_0669 | transporter | -7.49 | 0.0001 |
| AB57_RS18165 | AB57_3665 | cation transporter | -7.49 | 0.0003 |
| AB57_RS14680 | AB57_2966 | serine protease | -7.49 | 0.0000 |
| AB57_RS14480 | AB57_RS14480 | pseudogene | -7.49 | 0.0075 |
| AB57_RS02130 | AB57_0405 | 23S rRNA (guanosine-2'-O-)-methyltransferase | -7.49 | 0.0043 |
| AB57_RS02335 | AB57_0444 | heat-shock protein Hsp33 | -7.49 | 0.0000 |
| AB57_RS03255 | AB57_0631 | peptide chain release factor 3 | -7.48 | 0.0044 |
| AB57_RS08925 | AB57_1790 | peptidylprolyl isomerase | -7.48 | 0.0045 |
| AB57_RS16545 | AB57_3349 | hypothetical protein | -7.48 | 0.0026 |
| AB57_RS18985 | AB57_3832 | nicotinate phosphoribosyltransferase | -7.48 | 0.0005 |
| AB57_RS03775 | AB57_0739 | protein FilA | -7.48 | 0.0041 |
| AB57_RS01425 | | MULTISPECIES: hypothetical protein | -7.47 | 0.0030 |
| AB57_RS02930 | AB57_0565 | MULTISPECIES: endonuclease | -7.46 | 0.0011 |
| AB57_RS17305 | AB57_3493 | ribonuclease R | -7.46 | 0.0049 |
| AB57_RS05165 | AB57_1016 | poly-beta-1,6-N-acetyl-D-glucosamine biosynthesis protein PgaD | -7.45 | 0.0001 |
| AB57_RS19420 | AB57_3921 | MULTISPECIES: DNA replication protein | -7.45 | 0.0016 |
| AB57_RS17665 | AB57_3566 | membrane protein | -7.45 | 0.0001 |
| AB57_RS05605 | AB57_1111 | lon protease | -7.45 | 0.0013 |
| AB57_RS16655 | AB57_3373 | tryptophan synthase subunit alpha | -7.45 | 0.0052 |
| AB57_RS15725 | AB57_4816 | tRNA-Arg | -7.45 | 0.0014 |
| AB57_RS04730 | AB57_0925 | threonine transporter RhtB | -7.45 | 0.0003 |
| AB57_RS14800 | AB57_2991 | Bcr/CflA family drug resistance efflux transporter | -7.45 | 0.0000 |
| AB57_RS18025 | AB57_3641 | hypothetical protein | -7.45 | 0.0018 |
| AB57_RS05410 | AB57_1069 | polyphosphate kinase | -7.44 | 0.0008 |
| AB57_RS01650 | AB57_0314 | riboflavin synthase subunit alpha | -7.44 | 0.0060 |
| AB57_RS17600 | AB57_3553 | ABC transporter permease | -7.43 | 0.0023 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS16480 | AB57_3334 | hypothetical protein | -7.43 | 0.0000 |
| AB57_RS04545 | AB57_0888 | competence protein ComL | -7.42 | 0.0003 |
| AB57_RS02865 | AB57_RS02865 | pseudogene | -7.42 | 0.0015 |
| AB57_RS16475 | AB57_3333 | peptidyl-prolyl cis-trans isomerase | -7.42 | 0.0003 |
| AB57_RS01185 | AB57_0232 | DNA topoisomerase IV subunit A | -7.42 | 0.0075 |
| AB57_RS17180 | AB57_3472 | hypothetical protein | -7.41 | 0.0044 |
| AB57_RS00815 | AB57_0161 | phosphopantetheine-protein transferase | -7.41 | 0.0025 |
| AB57_RS02620 | AB57_0502 | tRNA pseudouridine synthase A | -7.41 | 0.0040 |
| AB57_RS15445 | AB57_3128 | succinate dehydrogenase iron-sulfur subunit | -7.41 | 0.0032 |
| AB57_RS05455 | AB57_1078 | hypothetical protein | -7.40 | 0.0021 |
| AB57_RS16920 | AB57_3428 | tRNA 5-methylaminomethyl-2-thiouridine biosynthesis bifunctional protein MnmC | -7.40 | 0.0026 |
| AB57_RS06775 | AB57_1343 | MULTISPECIES: 3-oxoacyl-ACP synthase | -7.40 | 0.0069 |
| AB57_RS13745 | AB57_2769 | RNA methyltransferase | -7.40 | 0.0001 |
| AB57_RS11325 | AB57_2275 | MULTISPECIES: glutaredoxin | -7.40 | 0.0026 |
| AB57_RS05425 | AB57_1072 | alpha/beta hydrolase | -7.39 | 0.0038 |
| AB57_RS16275 | AB57_3299 | membrane protein | -7.38 | 0.0031 |
| AB57_RS13405 | AB57_2702 | head morphogenesis protein | -7.38 | 0.0030 |
| AB57_RS07145 | AB57_1417 | hypothetical protein | -7.38 | 0.0010 |
| AB57_RS03685 | AB57_0723 | hypothetical protein | -7.38 | 0.0026 |
| AB57_RS11375 | AB57_2286 | 1-phosphofructokinase | -7.37 | 0.0009 |
| AB57_RS08755 | AB57_1756 | N-acetyltransferase | -7.36 | 0.0017 |
| AB57_RS16665 | AB57_3375 | pseudogene | -7.36 | 0.0012 |
| AB57_RS01445 | | MULTISPECIES: hypothetical protein | -7.36 | 0.0002 |
| AB57_RS11335 | AB57_2277 | LysR family transcriptional regulator | -7.35 | 0.0001 |
| AB57_RS17575 | AB57_3547 | hypothetical protein | -7.35 | 0.0001 |
| AB57_RS13630 | AB57_2747 | hypothetical protein | -7.35 | 0.0040 |
| AB57_RS17835 | AB57_3603 | NADPH quinone reductase MdaB | -7.34 | 0.0003 |
| AB57_RS18930 | AB57_3821 | hypothetical protein | -7.34 | 0.0045 |
| AB57_RS02135 | AB57_0407 | hypothetical protein | -7.34 | 0.0000 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS13645 | AB57_2750 | hypothetical protein | -7.33 | 0.0003 |
| AB57_RS00535 | AB57_0106 | acetyltransferase | -7.33 | 0.0049 |
| AB57_RS17080 | AB57_3455 | hypothetical protein | -7.33 | 0.0006 |
| AB57_RS13610 | AB57_2742 | integrase | -7.33 | 0.0030 |
| AB57_RS04590 | AB57_0896 | transcriptional regulator | -7.33 | 0.0001 |
| AB57_RS06350 | AB57_1257 | pseudogene | -7.33 | 0.0003 |
| AB57_RS03800 | AB57_0744 | protein FilF | -7.33 | 0.0079 |
| AB57_RS05565 | AB57_1102 | membrane protein | -7.32 | 0.0021 |
| AB57_RS18875 | AB57_3810 | esterase | -7.31 | 0.0031 |
| AB57_RS00195 | AB57_0040 | UDP-N-acetylmuramate:L-alanyl-gamma-D-glutamyl-meso-diaminopimelate ligase | -7.31 | 0.0006 |
| AB57_RS02050 | AB57_0389 | MULTISPECIES: aspartyl/asparaginyl beta-hydroxylase | -7.31 | 0.0000 |
| AB57_RS17965 | AB57_3629 | hypothetical protein | -7.30 | 0.0083 |
| AB57_RS15540 | AB57_3147 | serine--tRNA ligase | -7.30 | 0.0003 |
| AB57_RS02105 | AB57_0400 | MULTISPECIES: Holliday junction DNA helicase | -7.30 | 0.0016 |
| AB57_RS17050 | AB57_3449 | dimethyladenosine transferase | -7.30 | 0.0050 |
| AB57_RS04335 | AB57_0848 | MULTISPECIES: GntR family transcriptional regulator | -7.30 | 0.0003 |
| AB57_RS18205 | AB57_3673 | O-acetylhomoserine aminocarboxypropyltransferase | -7.29 | 0.0012 |
| AB57_RS15615 | AB57_3162 | MULTISPECIES: oxidoreductase | -7.29 | 0.0026 |
| AB57_RS10540 | AB57_2113 | NUDIX hydrolase | -7.29 | 0.0008 |
| AB57_RS03210 | AB57_0622 | leucine--tRNA ligase | -7.29 | 0.0039 |
| AB57_RS00520 | AB57_0103 | glycosyl transferase family 1 | -7.29 | 0.0094 |
| AB57_RS00155 | AB57_0032 | hypothetical protein | -7.28 | 0.0047 |
| AB57_RS03835 | AB57_0751 | esterase | -7.28 | 0.0026 |
| AB57_RS16910 | AB57_RS16910 | pseudogene | -7.28 | 0.0001 |
| AB57_RS14260 | AB57_2873 | high frequency lysogenization protein HflD | -7.28 | 0.0062 |
| AB57_RS18475 | AB57_3727 | NAD-dependent succinate-semialdehyde dehydrogenase | -7.28 | 0.0035 |
| AB57_RS18630 | AB57_3760 | transporter | -7.28 | 0.0022 |
| AB57_RS16890 | AB57_3420 | hypothetical protein | -7.28 | 0.0002 |
| AB57_RS05095 | AB57_1002 | oxygen-dependent choline dehydrogenase | -7.27 | 0.0057 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS12180 | AB57_2455 | membrane protein | -7.27 | 0.0011 |
| AB57_RS00005 | AB57_0001 | tyrosine--tRNA ligase | -7.26 | 0.0094 |
| AB57_RS00100 | AB57_0020 | chromosomal replication initiator protein DnaA | -7.26 | 0.0023 |
| AB57_RS03245 | AB57_0629 | TetR family transcriptional regulator | -7.25 | 0.0072 |
| AB57_RS00250 | AB57_0051 | hypothetical protein | -7.25 | 0.0032 |
| AB57_RS01190 | AB57_0233 | long-chain fatty acid--CoA ligase | -7.25 | 0.0004 |
| AB57_RS02995 | AB57_0579 | trigger factor | -7.24 | 0.0001 |
| AB57_RS15750 | AB57_3190 | hypothetical protein | -7.23 | 0.0094 |
| AB57_RS05470 | AB57_1082 | hypothetical protein | -7.22 | 0.0049 |
| AB57_RS03765 | AB57_0737 | histidinol-phosphate aminotransferase | -7.22 | 0.0004 |
| AB57_RS00075 | AB57_0015 | membrane protein | -7.22 | 0.0022 |
| AB57_RS16170 | AB57_3277 | O-acetyltransferase | -7.22 | 0.0007 |
| AB57_RS03025 | AB57_0585 | phosphate acetyltransferase | -7.22 | 0.0001 |
| AB57_RS04830 | AB57_0945 | type IV pili twitching motility protein PilT | -7.22 | 0.0005 |
| AB57_RS18620 | AB57_3756 | acetyl-coenzyme A synthetase | -7.22 | 0.0000 |
| AB57_RS12500 | AB57_2522 | ribosomal protein L11 methyltransferase | -7.21 | 0.0008 |
| AB57_RS00410 | AB57_0079 | TetR family transcriptional regulator | -7.21 | 0.0019 |
| AB57_RS17915 | AB57_3619 | pilus assembly protein PilE | -7.21 | 0.0002 |
| AB57_RS02785 | AB57_0536 | MULTISPECIES: biopolymer transporter ExbB | -7.20 | 0.0030 |
| AB57_RS02915 | AB57_0562 | MULTISPECIES: hypothetical protein | -7.20 | 0.0002 |
| AB57_RS09220 | AB57_1850 | MULTISPECIES: guanylate cyclase | -7.20 | 0.0001 |
| AB57_RS01715 | AB57_0327 | threonine synthase | -7.19 | 0.0039 |
| AB57_RS05730 | AB57_1138 | microcin C ABC transporter permease YejB | -7.19 | 0.0008 |
| AB57_RS12460 | AB57_2513 | alanine racemase | -7.18 | 0.0002 |
| AB57_RS16670 | AB57_3377 | hypothetical protein | -7.18 | 0.0015 |
| AB57_RS18575 | AB57_3747 | acetate permease | -7.18 | 0.0052 |
| AB57_RS05795 | AB57_1149 | MULTISPECIES: ribonuclease T | -7.18 | 0.0002 |
| AB57_RS05330 | AB57_1052 | D-serine dehydratase | -7.17 | 0.0038 |
| AB57_RS17645 | AB57_3562 | hypothetical protein | -7.17 | 0.0050 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS14925 | AB57_3017 | peptidase S49 | -7.17 | 0.0011 |
| AB57_RS07695 | AB57_1533 | helicase | -7.17 | 0.0075 |
| AB57_RS13775 | AB57_2775 | 2-alkenal reductase | -7.17 | 0.0005 |
| AB57_RS01065 | AB57_RS01065 | pseudogene | -7.17 | 0.0035 |
| AB57_RS15735 | AB57_3186 | hypothetical protein | -7.17 | 0.0055 |
| AB57_RS17560 | AB57_3544 | ATP-dependent DNA helicase RecG | -7.17 | 0.0047 |
| AB57_RS09370 | AB57_1880 | siderophore biosynthesis protein | -7.16 | 0.0030 |
| AB57_RS02645 | AB57_0508 | endonuclease | -7.16 | 0.0000 |
| AB57_RS08930 | AB57_1791 | LPS-assembly protein LptD | -7.16 | 0.0094 |
| AB57_RS04165 | AB57_0813 | uracil phosphoribosyltransferase | -7.16 | 0.0048 |
| AB57_RS03165 | AB57_0613 | GTPase | -7.16 | 0.0031 |
| AB57_RS01150 | AB57_RS01150 | pseudogene | -7.15 | 0.0000 |
| AB57_RS07180 | AB57_1424 | DNA mismatch repair protein MutS | -7.14 | 0.0016 |
| AB57_RS07140 | AB57_1416 | secretion protein HlyD | -7.14 | 0.0000 |
| AB57_RS15325 | AB57_3099 | hypothetical protein | -7.13 | 0.0026 |
| AB57_RS01825 | AB57_0349 | DNA-binding response regulator | -7.13 | 0.0023 |
| AB57_RS04840 | AB57_0947 | ATP-dependent dsDNA exonuclease | -7.12 | 0.0009 |
| AB57_RS07100 | | cysteinyl-tRNA synthetase | -7.12 | 0.0016 |
| AB57_RS19200 | AB57_3879 | hypothetical protein | -7.12 | 0.0003 |
| AB57_RS18790 | AB57_3793 | arsenate reductase | -7.12 | 0.0006 |
| AB57_RS15425 | AB57_3123 | MULTISPECIES: succinyl-CoA ligase subunit beta | -7.12 | 0.0053 |
| AB57_RS17655 | AB57_3564 | hypothetical protein | -7.12 | 0.0093 |
| AB57_RS18960 | AB57_3827 | MULTISPECIES: DNA-binding response regulator | -7.11 | 0.0028 |
| AB57_RS12495 | AB57_2521 | MULTISPECIES: hypothetical protein | -7.11 | 0.0002 |
| AB57_RS03115 | AB57_0603 | DNA-binding protein | -7.11 | 0.0066 |
| AB57_RS16555 | AB57_3351 | glucose dehydrogenase | -7.10 | 0.0004 |
| AB57_RS13000 | | oxidoreductase | -7.10 | 0.0011 |
| AB57_RS05085 | | hypothetical protein | -7.10 | 0.0008 |
| AB57_RS03175 | AB57_0615 | membrane protein | -7.10 | 0.0020 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|--|---|----------------|
| AB57_RS06735 | AB57_1335 | glutathionine S-transferase | -7.10 | 0.0016 |
| AB57_RS01455 | | transposase | -7.10 | 0.0006 |
| AB57_RS03080 | AB57_0596 | peptidase S41 | -7.09 | 0.0000 |
| AB57_RS04005 | AB57_0781 | MULTISPECIES: hypothetical protein | -7.09 | 0.0030 |
| AB57_RS15235 | AB57_3084 | diacylglycerol kinase | -7.09 | 0.0001 |
| AB57_RS15890 | AB57_3220 | methyltransferase | -7.08 | 0.0008 |
| AB57_RS16915 | AB57_3426 | hypothetical protein | -7.08 | 0.0006 |
| AB57_RS08965 | AB57_1798 | MULTISPECIES: biopolymer transporter ExbD | -7.08 | 0.0030 |
| AB57_RS17045 | AB57_3448 | diadenosine tetraphosphatase | -7.08 | 0.0027 |
| AB57_RS12875 | AB57_4792 | | -7.08 | 0.0006 |
| AB57_RS00310 | AB57_0059 | MULTISPECIES: peptidylprolyl isomerase | -7.07 | 0.0002 |
| AB57_RS02970 | AB57_0574 | MULTISPECIES: methionine biosynthesis protein MetW | -7.07 | 0.0019 |
| AB57_RS17320 | AB57_3496 | hypothetical protein | -7.06 | 0.0003 |
| AB57_RS08875 | AB57_1780 | hypothetical protein | -7.06 | 0.0010 |
| AB57_RS17550 | AB57_3542 | RND transporter | -7.06 | 0.0003 |
| AB57_RS19035 | | phosphatidylglycerophosphatase A | -7.06 | 0.0002 |
| AB57_RS14815 | AB57_2994 | Holliday junction DNA helicase RuvA | -7.05 | 0.0000 |
| AB57_RS16700 | AB57_3383 | glycerophosphoryl diester phosphodiesterase | -7.05 | 0.0001 |
| AB57_RS12870 | AB57_4829 | phosphohistidine phosphatase | -7.05 | 0.0006 |
| AB57_RS15610 | AB57_3160 | polyketide cyclase | -7.05 | 0.0039 |
| AB57_RS00685 | AB57_0135 | ligand-gated channel protein | -7.05 | 0.0004 |
| GenBank:STS:NC _011586:1492875: D1S3688 1494163 | | | -7.05 | 0.0038 |
| AB57_RS12185 | AB57_2456 | transcriptional regulator | -7.04 | 0.0000 |
| AB57_RS03040 | AB57_0588 | 2-dehydro-3-deoxyphosphogluconate aldolase | -7.04 | 0.0055 |
| AB57_RS04485 | AB57_0880 | peptidyl-tRNA hydrolase | -7.04 | 0.0059 |
| AB57_RS01555 | | MULTISPECIES: hypothetical protein | -7.04 | 0.0030 |
| AB57_RS16485 | AB57_3335 | MULTISPECIES: large-conductance mechanosensitive channel | -7.03 | 0.0052 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS09215 | AB57_1849 | MULTISPECIES: hypothetical protein | -7.03 | 0.0002 |
| AB57_RS02395 | AB57_0456 | hypothetical protein | -7.03 | 0.0001 |
| AB57_RS18910 | AB57_3817 | type VI secretion system protein | -7.02 | 0.0002 |
| AB57_RS00675 | AB57_0133 | hypothetical protein | -7.02 | 0.0002 |
| AB57_RS11395 | AB57_2290 | hypothetical protein | -7.02 | 0.0002 |
| AB57_RS03855 | AB57_0755 | hypothetical protein | -7.01 | 0.0000 |
| AB57_RS17100 | AB57_3459 | acetyl-CoA carboxylase carboxyltransferase subunit | -7.00 | 0.0010 |
| AB57_RS02585 | AB57_0495 | 3-isopropylmalate dehydratase small subunit | -7.00 | 0.0012 |
| GenBank:repeat_r egion:NC_011586: 275891:279266 | | | -7.00 | 0.0034 |
| AB57_RS16130 | AB57_3268 | undecaprenyl-diphosphatase | -7.00 | 0.0055 |
| AB57_RS18980 | AB57_3831 | thiosulfate sulfurtransferase | -6.99 | 0.0000 |
| AB57_RS14210 | AB57_2863 | D-alanyl-D-alanine carboxypeptidase | -6.99 | 0.0035 |
| AB57_RS01365 | | MULTISPECIES: hypothetical protein | -6.99 | 0.0002 |
| AB57_RS17075 | AB57_3454 | peptidase | -6.98 | 0.0010 |
| AB57_RS01490 | AB57_0287 | transposase | -6.98 | 0.0007 |
| AB57_RS07015 | | phage head morphogenesis protein | -6.97 | 0.0017 |
| AB57_RS14830 | AB57_2997 | MULTISPECIES: thioesterase | -6.96 | 0.0008 |
| AB57_RS09990 | AB57_2007 | MULTISPECIES: ferrous iron transporter B | -6.96 | 0.0094 |
| AB57_RS17880 | AB57_3612 | tRNA pseudouridine synthase B | -6.96 | 0.0047 |
| AB57_RS13845 | AB57_2786 | anthranilate synthase subunit II | -6.96 | 0.0046 |
| AB57_RS03005 | AB57_0581 | ATP-dependent Clp protease ATP-binding subunit ClpX | -6.96 | 0.0000 |
| AB57_RS02705 | AB57_0520 | MULTISPECIES: hypothetical protein | -6.96 | 0.0016 |
| AB57_RS04940 | AB57_0967 | hypothetical protein | -6.96 | 0.0087 |
| AB57_RS00945 | AB57_0185 | glutathione peroxidase | -6.96 | 0.0062 |
| AB57_RS19075 | AB57_3854 | haloacid dehalogenase | -6.96 | 0.0044 |
| AB57_RS19250 | AB57_3889 | MULTISPECIES: hypothetical protein | -6.95 | 0.0000 |
| AB57_RS19165 | AB57_3872 | hypothetical protein | -6.95 | 0.0018 |
| AB57_RS19245 | AB57_3888 | peptidase M23 | -6.95 | 0.0000 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|--|------------------|--|---|----------------|
| AB57_RS18130 | AB57_4798 | tRNA-Glu | -6.95 | 0.0007 |
| AB57_RS11125 | AB57_2233 | hypothetical protein | -6.95 | 0.0046 |
| AB57_RS01850 | AB57_0354 | thioesterase | -6.94 | 0.0046 |
| AB57_RS17660 | AB57_3565 | PspC family transcriptional regulator | -6.94 | 0.0017 |
| AB57_RS07020 | AB57_1391 | hypothetical protein | -6.93 | 0.0094 |
| AB57_RS02565 | AB57_0491 | haloacid dehalogenase | -6.92 | 0.0000 |
| AB57_RS00550 | AB57_RS00550 | pseudogene | -6.92 | 0.0044 |
| AB57_RS12560 | AB57_2534 | mutlidrug resistance protein | -6.91 | 0.0081 |
| AB57_RS17625 | AB57_3558 | 1-deoxy-D-xylulose-5-phosphate synthase | -6.91 | 0.0070 |
| AB57_RS09625 | AB57_1933 | 2,3-butanediol dehydrogenase | -6.90 | 0.0002 |
| AB57_RS00940 | AB57_0184 | MULTISPECIES: DNA repair protein HhH-GPD | -6.90 | 0.0016 |
| AB57_RS04875 | AB57_0954 | delta-aminolevulinic acid dehydratase | -6.90 | 0.0058 |
| AB57_RS15605 | AB57_3161 | metal-binding protein | -6.90 | 0.0000 |
| AB57_RS01210 | AB57_0237 | membrane protein | -6.90 | 0.0009 |
| AB57_RS15185 | AB57_3073 | protease HtpX | -6.90 | 0.0013 |
| AB57_RS04550 | AB57_0889 | 23S rRNA pseudouridine synthase D | -6.89 | 0.0014 |
| AB57_RS08955 | AB57_1796 | chromosome partitioning protein | -6.88 | 0.0026 |
| AB57_RS00455 | AB57_0089 | peptidyl-prolyl cis-trans isomerase | -6.88 | 0.0027 |
| AB57_RS14735 | AB57_2978 | CysB family transcriptional regulator | -6.87 | 0.0022 |
| AB57_RS18435 | AB57_3719 | MFS transporter | -6.87 | 0.0050 |
| AB57_RS16325 | AB57_3303 | peptidase | -6.87 | 0.0001 |
| AB57_RS00820 | AB57_4835 | tRNA-Gly | -6.87 | 0.0054 |
| AB57_RS00170 | AB57_0035 | carboxylate-amine ligase | -6.87 | 0.0025 |
| AB57_RS19370 | AB57_3914 | hypothetical protein | -6.87 | 0.0011 |
| AB57_RS17315 | AB57_3495 | metal-binding protein | -6.87 | 0.0014 |
| GenBank:repeat_r egion:NC_011586: 8613:12194 | | | -6.87 | 0.0000 |
| AB57_RS16635 | AB57_3369 | protein-tyrosine phosphatase | -6.86 | 0.0000 |
| AB57_RS04720 | AB57_0922 | thioredoxin reductase | -6.86 | 0.0011 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS09250 | AB57_1856 | cysteine desulfurase IscS | -6.85 | 0.0030 |
| AB57_RS18445 | AB57_3721 | aldehyde-activating protein | -6.85 | 0.0022 |
| AB57_RS04535 | AB57_0887 | glutamyl-tRNA reductase | -6.85 | 0.0055 |
| AB57_RS04885 | AB57_0956 | MULTISPECIES: hemolysin D | -6.85 | 0.0026 |
| AB57_RS14335 | AB57_2891 | membrane protein | -6.84 | 0.0048 |
| AB57_RS04315 | AB57_0844 | signal recognition particle protein | -6.84 | 0.0030 |
| AB57_RS03190 | AB57_0618 | MacB family efflux pump subunit | -6.84 | 0.0038 |
| AB57_RS18885 | AB57_3812 | hypothetical protein | -6.84 | 0.0003 |
| AB57_RS03380 | AB57_0660 | MULTISPECIES: two-component system sensor histidine kinase | -6.83 | 0.0094 |
| AB57_RS12915 | AB57_2601 | <u>30S ribosomal protein S21</u> | -6.83 | 0.0003 |
| AB57_RS18125 | AB57_4826 | tRNA-Ala | -6.83 | 0.0006 |
| AB57_RS05480 | AB57_1084 | citrate transporter | -6.82 | 0.0000 |
| AB57_RS14455 | AB57_2921 | isocitrate dehydrogenase | -6.82 | 0.0031 |
| AB57_RS17955 | AB57_3627 | guanylate kinase | -6.82 | 0.0003 |
| AB57_RS07480 | AB57_1490 | ATPase | -6.82 | 0.0052 |
| AB57_RS15420 | AB57_3122 | MULTISPECIES: succinyl-CoA synthetase subunit alpha | -6.81 | 0.0001 |
| AB57_RS07090 | | integrase | -6.81 | 0.0010 |
| AB57_RS14465 | AB57_2923 | D-alanyl-D-alanine carboxypeptidase | -6.80 | 0.0000 |
| AB57_RS18845 | AB57_3804 | uracil permease | -6.80 | 0.0025 |
| AB57_RS01955 | AB57_0370 | DNA-directed RNA polymerase subunit beta' | -6.80 | 0.0003 |
| AB57_RS03195 | AB57_0619 | MacA family efflux pump subunit | -6.80 | 0.0007 |
| AB57_RS18105 | AB57_3657 | ribosomal RNA small subunit methyltransferase H | -6.80 | 0.0033 |
| AB57_RS16610 | AB57_3364 | MFS transporter | -6.80 | 0.0034 |
| AB57_RS05355 | AB57_1057 | membrane protein | -6.79 | 0.0002 |
| AB57_RS18260 | AB57_3682 | MULTISPECIES: imidazoleglycerol-phosphate dehydratase | -6.79 | 0.0028 |
| AB57_RS18815 | AB57_3798 | hypothetical protein | -6.79 | 0.0041 |
| AB57_RS13070 | AB57_2634 | C4-dicarboxylate ABC transporter | -6.79 | 0.0042 |
| AB57_RS17615 | AB57_3556 | DEAD/DEAH box helicase | -6.78 | 0.0015 |
| AB57_RS03790 | AB57_0742 | protein FilD | -6.78 | 0.0027 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS17265 | AB57_3489 | hypothetical protein | -6.77 | 0.0005 |
| AB57_RS15135 | | hypothetical protein | -6.77 | 0.0030 |
| AB57_RS18020 | AB57_3640 | glutamate synthase subunit alpha | -6.77 | 0.0012 |
| AB57_RS13290 | AB57_2681 | hypothetical protein | -6.77 | 0.0028 |
| AB57_RS03975 | AB57_0775 | long-chain fatty acid transporter | -6.77 | 0.0062 |
| AB57_RS00670 | AB57_0132 | pseudogene | -6.77 | 0.0026 |
| AB57_RS10845 | AB57_2176 | MULTISPECIES: GntR family transcriptional regulator | -6.77 | 0.0018 |
| AB57_RS01865 | AB57_0357 | general secretion pathway protein | -6.76 | 0.0002 |
| AB57_RS18300 | AB57_3691 | cold-shock protein | -6.76 | 0.0026 |
| AB57_RS17165 | AB57_3469 | CDP-diacylglycerol--serine O-phosphatidyltransferase | -6.76 | 0.0001 |
| AB57_RS13540 | AB57_2730 | hypothetical protein | -6.76 | 0.0014 |
| AB57_RS18590 | AB57_3750 | hypothetical protein | -6.76 | 0.0003 |
| AB57_RS00225 | AB57_0046 | hypothetical protein | -6.76 | 0.0055 |
| AB57_RS00800 | AB57_0158 | RND transporter | -6.75 | 0.0045 |
| AB57_RS04185 | AB57_0817 | ferredoxin-NADP reductase | -6.75 | 0.0001 |
| AB57_RS16640 | AB57_3370 | cell division protein | -6.75 | 0.0014 |
| AB57_RS19330 | AB57_3906 | phosphoenolpyruvate carboxylase | -6.75 | 0.0010 |
| AB57_RS03020 | AB57_0584 | fumarate hydratase | -6.74 | 0.0020 |
| AB57_RS12505 | AB57_2523 | hypothetical protein | -6.74 | 0.0001 |
| AB57_RS19205 | AB57_3880 | formate transporter | -6.74 | 0.0000 |
| AB57_RS05335 | AB57_1053 | hypothetical protein | -6.74 | 0.0020 |
| AB57_RS00045 | AB57_0009 | class C beta-lactamase | -6.74 | 0.0058 |
| AB57_RS07095 | AB57_4838 | cysteine--tRNA ligase | -6.73 | 0.0010 |
| AB57_RS05485 | AB57_1085 | hypothetical protein | -6.73 | 0.0030 |
| AB57_RS01805 | AB57_0345 | MULTISPECIES: transcriptional regulator | -6.73 | 0.0003 |
| AB57_RS15985 | AB57_3238 | nuclease | -6.73 | 0.0003 |
| AB57_RS11575 | AB57_2328 | hypothetical protein | -6.73 | 0.0002 |
| AB57_RS05580 | AB57_1106 | iron-sulfur protein | -6.72 | 0.0054 |
| AB57_RS15670 | AB57_3174 | lipid A phosphoethanolamine transferase | -6.72 | 0.0022 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS04465 | AB57_0876 | MULTISPECIES: 3,4-dihydroxy-2-butanone-4-phosphate synthase | -6.72 | 0.0062 |
| AB57_RS18695 | AB57_3773 | ligand-gated channel protein | -6.71 | 0.0008 |
| AB57_RS02630 | AB57_0504 | hypothetical protein | -6.71 | 0.0054 |
| AB57_RS00025 | AB57_0005 | hypothetical protein | -6.71 | 0.0038 |
| AB57_RS07675 | AB57_1529 | MULTISPECIES: PaaX family transcriptional regulator | -6.71 | 0.0003 |
| AB57_RS12790 | AB57_2580 | sodium:glutamate symporter | -6.70 | 0.0020 |
| AB57_RS11470 | AB57_2305 | haloacid dehalogenase | -6.70 | 0.0030 |
| AB57_RS17960 | AB57_3628 | MULTISPECIES: DNA-directed RNA polymerase subunit omega | -6.69 | 0.0023 |
| AB57_RS02485 | AB57_0475 | short-chain dehydrogenase | -6.69 | 0.0056 |
| AB57_RS18665 | AB57_3767 | recombination protein RecJ | -6.69 | 0.0002 |
| AB57_RS04810 | AB57_0941 | hypothetical protein | -6.68 | 0.0094 |
| AB57_RS03760 | AB57_0734 | pseudogene | -6.68 | 0.0002 |
| AB57_RS01200 | AB57_0235 | membrane protein | -6.68 | 0.0006 |
| AB57_RS04215 | AB57_0823 | 1-aminocyclopropane-1-carboxylate deaminase | -6.68 | 0.0058 |
| AB57_RS18390 | AB57_3710 | HxIR family transcriptional regulator | -6.68 | 0.0015 |
| GenBank:repeat_r egion:NC_011586: 6676:8609 | | | -6.67 | 0.0058 |
| AB57_RS00265 | AB57_0054 | esterase | -6.67 | 0.0000 |
| AB57_RS11230 | AB57_2256 | peptidase M16 | -6.67 | 0.0026 |
| AB57_RS18155 | AB57_3663 | hypothetical protein | -6.67 | 0.0017 |
| AB57_RS01050 | AB57_0206 | TonB-dependent receptor | -6.67 | 0.0006 |
| AB57_RS18925 | AB57_3820 | membrane protein | -6.67 | 0.0013 |
| AB57_RS00355 | AB57_0068 | MULTISPECIES: alkanesulfonate monooxygenase | -6.67 | 0.0010 |
| AB57_RS18525 | AB57_3737 | thiaminase II | -6.66 | 0.0008 |
| AB57_RS02590 | AB57_0496 | hypothetical protein | -6.66 | 0.0062 |
| AB57_RS16025 | AB57_3246 | DNA helicase | -6.66 | 0.0030 |
| AB57_RS12355 | AB57_2491 | hypothetical protein | -6.66 | 0.0001 |
| AB57_RS16705 | AB57_3384 | hypothetical protein | -6.66 | 0.0023 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS17815 | AB57_3598 | hypothetical protein | -6.66 | 0.0003 |
| AB57_RS12140 | AB57_2447 | tRNA dimethylallyltransferase | -6.65 | 0.0002 |
| AB57_RS16465 | | formamidopyrimidine-DNA glycosylase | -6.65 | 0.0008 |
| AB57_RS08725 | AB57_1749 | hypothetical protein | -6.65 | 0.0002 |
| AB57_RS01345 | AB57_RS01345 | pseudogene | -6.65 | 0.0000 |
| AB57_RS03830 | AB57_0750 | hypothetical protein | -6.65 | 0.0024 |
| AB57_RS17610 | | hypothetical protein | -6.65 | 0.0003 |
| AB57_RS04845 | AB57_0948 | ATP-dependent dsDNA exonuclease | -6.65 | 0.0052 |
| AB57_RS15595 | AB57_3158 | adeC/adeK/oprM family multidrug efflux complex outer membrane factor | -6.65 | 0.0017 |
| AB57_RS16185 | AB57_3280 | septum formation inhibitor Maf | -6.64 | 0.0012 |
| AB57_RS00735 | AB57_0145 | 3-hydroxyisobutyrate dehydrogenase | -6.64 | 0.0006 |
| AB57_RS05370 | AB57_1060 | hypothetical protein | -6.63 | 0.0003 |
| GenBank:STS:NC _011586:297629:2 97780 | D13S22 | | -6.63 | 0.0000 |
| AB57_RS00440 | AB57_0085 | nicotinate-nucleotide pyrophosphorylase | -6.63 | 0.0008 |
| AB57_RS12880 | AB57_4830 | nitroreductase | -6.62 | 0.0016 |
| AB57_RS17775 | AB57_3590 | S-methylmethionine transporter | -6.62 | 0.0011 |
| AB57_RS17505 | AB57_3533 | cystathionine beta-lyase | -6.61 | 0.0000 |
| AB57_RS13740 | AB57_2768 | 5,10-methylenetetrahydrofolate reductase | -6.61 | 0.0026 |
| AB57_RS00065 | AB57_0013 | RND transporter | -6.60 | 0.0054 |
| AB57_RS02570 | AB57_0492 | MULTISPECIES: LysR family transcriptional regulator | -6.60 | 0.0008 |
| AB57_RS15250 | AB57_3087 | glycerol kinase | -6.60 | 0.0023 |
| AB57_RS04865 | AB57_0953 | nucleoside-diphosphate sugar epimerase | -6.60 | 0.0028 |
| AB57_RS11210 | AB57_2252 | threonine dehydratase | -6.60 | 0.0009 |
| AB57_RS06690 | AB57_1325 | ribosomal RNA large subunit methyltransferase K/L | -6.59 | 0.0000 |
| AB57_RS18895 | AB57_RS18895 | pseudogene | -6.59 | 0.0009 |
| AB57_RS16355 | AB57_3309 | coproporphyrinogen III oxidase | -6.58 | 0.0016 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS05790 | AB57_4797 | tRNA-Glu | -6.57 | 0.0075 |
| AB57_RS01265 | AB57_0249 | pyridine nucleotide-disulfide oxidoreductase | -6.57 | 0.0015 |
| AB57_RS18380 | AB57_3708 | hypothetical protein | -6.57 | 0.0011 |
| AB57_RS01110 | AB57_0215 | FMN reductase | -6.57 | 0.0030 |
| AB57_RS15490 | AB57_3137 | RNA polymerase sigma factor RpoD | -6.57 | 0.0001 |
| AB57_RS01710 | AB57_0326 | D-alanyl-D-alanine endopeptidase | -6.57 | 0.0011 |
| AB57_RS15295 | AB57_3093 | hypothetical protein | -6.57 | 0.0022 |
| AB57_RS02610 | AB57_0500 | MULTISPECIES: translation initiation factor IF-1 | -6.57 | 0.0094 |
| AB57_RS04880 | AB57_0955 | hypothetical protein | -6.56 | 0.0047 |
| AB57_RS12360 | | hypothetical protein | -6.55 | 0.0004 |
| AB57_RS02695 | AB57_0518 | Zn-dependent oxidoreductase | -6.55 | 0.0036 |
| AB57_RS19325 | AB57_3905 | TetR family transcriptional regulator | -6.55 | 0.0039 |
| AB57_RS16180 | AB57_3279 | ribonuclease G | -6.55 | 0.0018 |
| AB57_RS02410 | AB57_0460 | ferrochelatae | -6.55 | 0.0050 |
| AB57_RS15745 | AB57_3189 | hypothetical protein | -6.55 | 0.0012 |
| AB57_RS14545 | AB57_2940 | glutamate 5-kinase | -6.55 | 0.0003 |
| AB57_RS16940 | AB57_3431 | acyl-CoA thioesterase | -6.55 | 0.0043 |
| AB57_RS07280 | AB57_1449 | hypothetical protein | -6.54 | 0.0030 |
| AB57_RS01440 | | MULTISPECIES: hypothetical protein | -6.54 | 0.0024 |
| AB57_RS04480 | AB57_0879 | aspartate 1-decarboxylase | -6.54 | 0.0050 |
| AB57_RS01410 | AB57_0273 | MULTISPECIES: transposase | -6.54 | 0.0013 |
| AB57_RS19365 | AB57_3913 | 16S rRNA methyltransferase | -6.54 | 0.0031 |
| AB57_RS14860 | AB57_3004 | hypothetical protein | -6.54 | 0.0028 |
| AB57_RS12165 | AB57_2453 | acetyltransferase | -6.54 | 0.0016 |
| AB57_RS02605 | AB57_0499 | hypothetical protein | -6.54 | 0.0094 |
| AB57_RS03350 | AB57_0654 | MULTISPECIES: hypothetical protein | -6.53 | 0.0002 |
| AB57_RS16675 | AB57_3378 | MULTISPECIES: hypothetical protein | -6.52 | 0.0020 |
| AB57_RS03625 | AB57_0710 | exopolyphosphatase | -6.52 | 0.0002 |
| AB57_RS12455 | AB57_2512 | DNA helicase | -6.52 | 0.0003 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS02370 | AB57_0451 | apolipoprotein N-acyltransferase | -6.52 | 0.0030 |
| AB57_RS04705 | AB57_0919 | alanine acetyltransferase | -6.52 | 0.0029 |
| AB57_RS16580 | AB57_3357 | 3-hydroxyacyl-CoA dehydrogenase | -6.51 | 0.0031 |
| AB57_RS16470 | | formamidopyrimidine-DNA glycosylase | -6.51 | 0.0094 |
| AB57_RS17570 | AB57_3546 | hypothetical protein | -6.51 | 0.0013 |
| AB57_RS01450 | AB57_0280 | MULTISPECIES: chloramphenicol acetyltransferase CAT | -6.51 | 0.0005 |
| AB57_RS15730 | AB57_3187 | integrase | -6.51 | 0.0034 |
| AB57_RS02445 | AB57_0467 | hypothetical protein | -6.50 | 0.0026 |
| AB57_RS13750 | AB57_2770 | MULTISPECIES: transposase | -6.50 | 0.0030 |
| AB57_RS03675 | AB57_0720 | organic hydroperoxide resistance protein | -6.50 | 0.0002 |
| AB57_RS18425 | AB57_3718 | permease | -6.50 | 0.0004 |
| AB57_RS11790 | AB57_2373 | lactonase | -6.50 | 0.0020 |
| AB57_RS17540 | AB57_3540 | acyl-CoA thioesterase II | -6.49 | 0.0028 |
| AB57_RS12350 | AB57_2490 | LuxR family transcriptional regulator | -6.49 | 0.0025 |
| AB57_RS02180 | AB57_4843 | tRNA-Met | -6.49 | 0.0046 |
| AB57_RS06725 | AB57_1333 | tRNA (guanine-N(7)-)-methyltransferase | -6.49 | 0.0063 |
| AB57_RS18560 | AB57_3744 | membrane protein | -6.49 | 0.0042 |
| AB57_RS03885 | AB57_0761 | N-acetylglutamate synthase | -6.49 | 0.0012 |
| AB57_RS11900 | AB57_2395 | universal stress protein | -6.48 | 0.0094 |
| AB57_RS14540 | AB57_2939 | phosphoserine phosphatase | -6.48 | 0.0001 |
| AB57_RS06755 | AB57_1339 | O-methyltransferase | -6.48 | 0.0018 |
| AB57_RS01845 | AB57_0353 | membrane protein | -6.48 | 0.0010 |
| AB57_RS15555 | AB57_3150 | MULTISPECIES: 50S ribosomal protein L27 | -6.47 | 0.0044 |
| AB57_RS01560 | | MULTISPECIES: hypothetical protein | -6.47 | 0.0015 |
| AB57_RS16960 | AB57_3435 | hypothetical protein | -6.47 | 0.0012 |
| AB57_RS19425 | AB57_3922 | MULTISPECIES: hypothetical protein | -6.47 | 0.0016 |
| AB57_RS00385 | AB57_0074 | MULTISPECIES: oxoacyl-ACP reductase | -6.47 | 0.0024 |
| AB57_RS04055 | AB57_0791 | membrane protein | -6.46 | 0.0022 |
| AB57_RS14175 | AB57_2856 | transporter | -6.46 | 0.0019 |
| AB57_RS05135 | AB57_1010 | cobalamin biosynthesis protein | -6.46 | 0.0001 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS07505 | AB57_1495 | D-Ala-D-Ala carboxypeptidase | -6.46 | 0.0030 |
| AB57_RS02290 | AB57_0435 | exonuclease V subunit beta | -6.46 | 0.0000 |
| AB57_RS19220 | AB57_3883 | hypothetical protein | -6.45 | 0.0018 |
| AB57_RS00910 | AB57_0178 | arginine--tRNA ligase | -6.45 | 0.0032 |
| AB57_RS15590 | AB57_3157 | AdeB family multidrug efflux RND transporter permease subunit | -6.45 | 0.0001 |
| AB57_RS16390 | AB57_3316 | hypothetical protein | -6.45 | 0.0008 |
| AB57_RS05445 | AB57_1076 | esterase | -6.45 | 0.0062 |
| AB57_RS07320 | AB57_1457 | voltage-gated chloride channel protein | -6.45 | 0.0064 |
| AB57_RS19185 | AB57_3876 | 4-hydroxy-tetrahydrodipicolinate synthase | -6.45 | 0.0046 |
| AB57_RS15340 | AB57_3102 | carbamoyl phosphate synthase large subunit | -6.45 | 0.0000 |
| AB57_RS02425 | AB57_0463 | SAM-dependent methyltransferase | -6.44 | 0.0018 |
| AB57_RS02990 | AB57_0578 | ligand-gated channel protein | -6.44 | 0.0022 |
| AB57_RS11330 | AB57_2276 | hypothetical protein | -6.44 | 0.0004 |
| AB57_RS01640 | AB57_0312 | deoxycytidylate deaminase | -6.44 | 0.0008 |
| AB57_RS15810 | AB57_3205 | hypothetical protein | -6.44 | 0.0003 |
| AB57_RS07220 | AB57_1434 | hypothetical protein | -6.44 | 0.0008 |
| GenBank:repeat_region:NC_011586:137219:138259 | | | -6.44 | 0.0051 |
| AB57_RS10695 | | MULTISPECIES: hypothetical protein | -6.44 | 0.0002 |
| AB57_RS02350 | AB57_0447 | type II secretion system protein GspF | -6.43 | 0.0002 |
| AB57_RS04345 | AB57_0850 | cell division protein ZipA | -6.43 | 0.0029 |
| AB57_RS05430 | AB57_1073 | pyridine nucleotide-disulfide oxidoreductase | -6.43 | 0.0033 |
| AB57_RS07315 | AB57_1456 | acetyl/propionyl-CoA carboxylase subunit alpha | -6.43 | 0.0044 |
| AB57_RS14690 | AB57_2968 | thioesterase | -6.42 | 0.0008 |
| AB57_RS07620 | AB57_1518 | enoyl-CoA hydratase | -6.42 | 0.0075 |
| AB57_RS03065 | AB57_0593 | hydrolase | -6.41 | 0.0054 |
| AB57_RS00805 | AB57_0159 | porin | -6.41 | 0.0033 |
| AB57_RS15065 | AB57_3049 | membrane protein | -6.41 | 0.0001 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS11550 | AB57_2321 | ribosomal protein S12 methylthiotransferase RimO | -6.41 | 0.0068 |
| AB57_RS14340 | AB57_2892 | RNA polymerase-associated protein RapA | -6.41 | 0.0003 |
| AB57_RS00780 | AB57_0154 | acyl-CoA synthetase | -6.40 | 0.0056 |
| AB57_RS16135 | AB57_3269 | PaaX family transcriptional regulator | -6.40 | 0.0085 |
| AB57_RS19180 | AB57_3875 | bifunctional pyrazinamidase/nicotinamidase | -6.40 | 0.0037 |
| AB57_RS18880 | AB57_3811 | hypothetical protein | -6.40 | 0.0039 |
| AB57_RS16765 | AB57_3396 | TonB-dependent receptor | -6.39 | 0.0071 |
| AB57_RS11220 | AB57_2254 | potassium transporter TrkH | -6.39 | 0.0001 |
| AB57_RS08230 | AB57_1646 | hypothetical protein | -6.38 | 0.0001 |
| AB57_RS00640 | AB57_0127 | MULTISPECIES: transposase | -6.38 | 0.0068 |
| AB57_RS09665 | AB57_1942 | AraC family transcriptional regulator | -6.38 | 0.0014 |
| AB57_RS00370 | AB57_0071 | N-acetylglutamate synthase | -6.38 | 0.0008 |
| AB57_RS18215 | AB57_3675 | sulfate permease | -6.37 | 0.0045 |
| AB57_RS15600 | AB57_3159 | MULTISPECIES: hypothetical protein | -6.37 | 0.0002 |
| AB57_RS19450 | AB57_3927 | MULTISPECIES: hypothetical protein | -6.36 | 0.0010 |
| AB57_RS18120 | AB57_3660 | glutamate--tRNA ligase | -6.36 | 0.0002 |
| AB57_RS16265 | AB57_3297 | P-nitrobenzoate reductase NfnB | -6.36 | 0.0002 |
| AB57_RS02980 | AB57_0576 | pseudogene | -6.36 | 0.0023 |
| AB57_RS19440 | AB57_3925 | MULTISPECIES: hypothetical protein | -6.35 | 0.0006 |
| AB57_RS07175 | AB57_1423 | GCN5 family N-acetyltransferase | -6.35 | 0.0008 |
| AB57_RS17755 | AB57_3585 | N-succinylglutamate 5-semialdehyde dehydrogenase | -6.35 | 0.0033 |
| AB57_RS14375 | AB57_RS14375 | pseudogene | -6.35 | 0.0039 |
| AB57_RS02730 | AB57_0525 | MULTISPECIES: RNA-binding protein | -6.34 | 0.0048 |
| AB57_RS02875 | AB57_0554 | hypothetical protein | -6.34 | 0.0007 |
| AB57_RS19170 | AB57_3873 | MATE family efflux transporter | -6.33 | 0.0022 |
| AB57_RS15560 | AB57_3151 | MULTISPECIES: 50S ribosomal protein L21 | -6.33 | 0.0062 |
| AB57_RS01800 | AB57_0344 | RND transporter | -6.33 | 0.0027 |
| AB57_RS02805 | AB57_0540 | MULTISPECIES: diacylglycerol kinase | -6.33 | 0.0069 |
| AB57_RS01360 | AB57_0268 | MULTISPECIES: transposase | -6.33 | 0.0015 |
| AB57_RS16340 | AB57_3306 | bacteriolytic lipoprotein entericidin B | -6.33 | 0.0002 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS00905 | AB57_0177 | cell division protein | -6.33 | 0.0020 |
| AB57_RS18835 | AB57_3802 | OHCU decarboxylase | -6.33 | 0.0048 |
| AB57_RS18330 | AB57_3698 | hypothetical protein | -6.32 | 0.0030 |
| AB57_RS16740 | AB57_3391 | MULTISPECIES: acyl-CoA dehydrogenase | -6.32 | 0.0000 |
| AB57_RS18295 | AB57_3690 | MULTISPECIES: membrane protein | -6.32 | 0.0066 |
| AB57_RS19095 | AB57_3858 | permease | -6.31 | 0.0016 |
| AB57_RS14430 | AB57_2914 | LysR family transcriptional regulator, partial | -6.31 | 0.0015 |
| AB57_RS15320 | AB57_3098 | MULTISPECIES: RNA-binding protein | -6.31 | 0.0002 |
| AB57_RS16845 | AB57_4796 | tRNA-Leu | -6.31 | 0.0025 |
| AB57_RS08510 | AB57_1704 | succinyldiaminopimelate transaminase | -6.31 | 0.0001 |
| AB57_RS11475 | AB57_2306 | RNA-binding protein | -6.30 | 0.0030 |
| AB57_RS05100 | AB57_1003 | NAD/NADP-dependent betaine aldehyde dehydrogenase | -6.29 | 0.0001 |
| AB57_RS02505 | AB57_0478 | ribonuclease E | -6.29 | 0.0032 |
| AB57_RS15260 | AB57_3089 | MULTISPECIES: hypothetical protein | -6.28 | 0.0015 |
| AB57_RS01260 | AB57_0248 | MULTISPECIES: hypothetical protein | -6.28 | 0.0004 |
| AB57_RS11350 | AB57_2280 | hypothetical protein | -6.28 | 0.0008 |
| AB57_RS05495 | AB57_1087 | isocitrate lyase | -6.28 | 0.0016 |
| AB57_RS18315 | AB57_3694 | hypothetical protein | -6.28 | 0.0005 |
| GenBank:STS:NC _011586:297629:2 97780 | D13S22 | | -6.27 | 0.0014 |
| AB57_RS07550 | AB57_1503 | transcriptional regulator | -6.27 | 0.0030 |
| AB57_RS10780 | | transcriptional regulator | -6.27 | 0.0014 |
| AB57_RS00715 | AB57_0141 | alanine glycine permease | -6.27 | 0.0012 |
| AB57_RS11690 | AB57_2351 | nitrate reductase | -6.27 | 0.0001 |
| AB57_RS05725 | AB57_1137 | peptide transporter | -6.27 | 0.0013 |
| AB57_RS14305 | AB57_2882 | hypothetical protein | -6.26 | 0.0001 |
| AB57_RS14450 | AB57_2920 | pseudouridine synthase | -6.26 | 0.0063 |
| AB57_RS06155 | AB57_1218 | adenine permease | -6.26 | 0.0034 |
| AB57_RS14005 | AB57_2819 | acinetobactin biosynthesis protein | -6.25 | 0.0003 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS00380 | AB57_0073 | MULTISPECIES: hypothetical protein | -6.25 | 0.0000 |
| AB57_RS17700 | AB57_3574 | hypothetical protein | -6.25 | 0.0016 |
| AB57_RS18280 | AB57_3687 | MULTISPECIES: 1-(5-phosphoribosyl)-5 | -6.25 | 0.0032 |
| AB57_RS18335 | AB57_3699 | amino acid transporter LysE | -6.25 | 0.0041 |
| AB57_RS03045 | AB57_0589 | permease | -6.24 | 0.0003 |
| AB57_RS18650 | AB57_3763 | ArsR family transcriptional regulator | -6.24 | 0.0046 |
| AB57_RS00890 | AB57_0174 | quercetin 2,3-dioxygenase | -6.24 | 0.0028 |
| AB57_RS01575 | | MULTISPECIES: conjugal transfer protein TrbI | -6.24 | 0.0008 |
| AB57_RS14200 | AB57_2861 | hypothetical protein | -6.23 | 0.0008 |
| AB57_RS19355 | AB57_3911 | dihydroxy-acid dehydratase | -6.23 | 0.0007 |
| AB57_RS08880 | AB57_1781 | serine/threonine transporter SstT | -6.23 | 0.0009 |
| AB57_RS00465 | AB57_0091 | tyrosine-protein kinase | -6.23 | 0.0024 |
| AB57_RS13905 | AB57_2798 | hypothetical protein | -6.22 | 0.0006 |
| AB57_RS18870 | AB57_3809 | DNA topoisomerase IV subunit B | -6.22 | 0.0008 |
| AB57_RS16855 | AB57_3413 | membrane protein | -6.22 | 0.0000 |
| AB57_RS07165 | AB57_1421 | hypothetical protein | -6.21 | 0.0050 |
| AB57_RS14255 | AB57_2872 | phosphate ABC transporter ATP-binding protein | -6.21 | 0.0049 |
| AB57_RS08485 | AB57_1699 | aminotransferase AlaT | -6.21 | 0.0028 |
| AB57_RS15675 | AB57_3175 | porin | -6.21 | 0.0025 |
| AB57_RS04280 | AB57_0836 | hypothetical protein | -6.21 | 0.0001 |
| AB57_RS02550 | AB57_0488 | catalase-peroxidase | -6.21 | 0.0044 |
| AB57_RS01525 | AB57_0292 | MULTISPECIES: GCN5 family acetyltransferase | -6.20 | 0.0003 |
| AB57_RS13840 | AB57_2785 | peptidase M24 | -6.20 | 0.0015 |
| AB57_RS05080 | AB57_0999 | homocysteine methyltransferase | -6.20 | 0.0055 |
| AB57_RS02515 | AB57_0481 | 23S rRNA pseudouridine synthase | -6.20 | 0.0051 |
| AB57_RS03030 | AB57_0586 | acetate kinase | -6.20 | 0.0056 |
| AB57_RS14640 | AB57_2958 | DNA repair protein RecO | -6.19 | 0.0018 |
| AB57_RS08535 | AB57_1709 | amino acid transporter | -6.19 | 0.0030 |
| AB57_RS05160 | AB57_1015 | poly-beta-1,6 N-acetyl-D-glucosamine synthase | -6.19 | 0.0035 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS18565 | AB57_3745 | beta-aspartyl peptidase | -6.18 | 0.0001 |
| AB57_RS06650 | AB57_1317 | aldo-keto reductase | -6.18 | 0.0030 |
| AB57_RS07250 | AB57_1442 | type VI secretion system protein | -6.17 | 0.0021 |
| AB57_RS03485 | AB57_0682 | 23S rRNA (uracil(1939)-C(5))-methyltransferase RImD | -6.17 | 0.0014 |
| AB57_RS06940 | AB57_1375 | HxlR family transcriptional regulator | -6.17 | 0.0037 |
| AB57_RS07905 | AB57_1578 | enoyl-CoA hydratase | -6.16 | 0.0019 |
| AB57_RS04265 | AB57_0833 | hypothetical protein | -6.16 | 0.0016 |
| AB57_RS13020 | AB57_2623 | MULTISPECIES: thiazole synthase | -6.16 | 0.0040 |
| AB57_RS16945 | AB57_3432 | thioredoxin | -6.16 | 0.0008 |
| AB57_RS16540 | AB57_3348 | MULTISPECIES: quaternary ammonium transporter | -6.16 | 0.0008 |
| AB57_RS16245 | AB57_3292 | threonine dehydratase | -6.16 | 0.0017 |
| AB57_RS16290 | AB57_RS16290 | pseudogene | -6.15 | 0.0002 |
| AB57_RS15575 | AB57_3154 | recombinase | -6.15 | 0.0014 |
| AB57_RS09000 | AB57_1806 | MULTISPECIES: TetR family transcriptional regulator | -6.15 | 0.0051 |
| AB57_RS05940 | AB57_1175 | alanine glycine permease | -6.15 | 0.0075 |
| AB57_RS01230 | AB57_0242 | GntR family transcriptional regulator | -6.14 | 0.0022 |
| AB57_RS05465 | AB57_1081 | sulfate adenylyltransferase | -6.14 | 0.0000 |
| AB57_RS18375 | AB57_3707 | HxlR family transcriptional regulator | -6.14 | 0.0006 |
| AB57_RS04990 | AB57_0978 | membrane protein | -6.14 | 0.0030 |
| AB57_RS00245 | AB57_0050 | LysR family transcriptional regulator | -6.14 | 0.0004 |
| AB57_RS11000 | AB57_2208 | LysR family transcriptional regulator | -6.13 | 0.0030 |
| AB57_RS14930 | AB57_3019 | lipid A biosynthesis acyltransferase | -6.13 | 0.0017 |
| AB57_RS16550 | AB57_3350 | sulfite reductase | -6.12 | 0.0023 |
| AB57_RS16600 | AB57_3362 | pseudogene | -6.12 | 0.0004 |
| AB57_RS19350 | AB57_3910 | hypothetical protein | -6.12 | 0.0052 |
| AB57_RS19155 | AB57_3870 | IclR family transcriptional regulator | -6.12 | 0.0021 |
| AB57_RS15305 | AB57_3095 | dihydropteroate synthase | -6.12 | 0.0038 |
| AB57_RS02975 | AB57_0575 | homoserine O-acetyltransferase | -6.11 | 0.0000 |
| AB57_RS17040 | AB57_3447 | signal peptidase | -6.10 | 0.0035 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS14425 | AB57_2913 | MULTISPECIES: L-cystine transporter tcyP | -6.10 | 0.0008 |
| AB57_RS16840 | AB57_3411 | two-component system sensor histidine kinase | -6.10 | 0.0008 |
| AB57_RS19475 | | flagellar protein | -6.10 | 0.0001 |
| AB57_RS03680 | AB57_RS03680 | pseudogene | -6.09 | 0.0033 |
| AB57_RS15945 | AB57_3232 | hypothetical protein | -6.09 | 0.0014 |
| AB57_RS05320 | AB57_1048 | B12-dependent methionine synthase | -6.08 | 0.0040 |
| AB57_RS00160 | AB57_0033 | guanine deaminase | -6.08 | 0.0015 |
| AB57_RS11025 | AB57_2213 | MULTISPECIES: hypothetical protein | -6.08 | 0.0018 |
| AB57_RS16850 | AB57_3412 | S-adenosylmethionine:tRNA ribosyltransferase-isomerase | -6.08 | 0.0001 |
| AB57_RS00175 | AB57_0036 | hypothetical protein | -6.08 | 0.0052 |
| AB57_RS13010 | AB57_2621 | Uncharacterized membrane protein YgdD | -6.08 | 0.0012 |
| AB57_RS01510 | | MULTISPECIES: transposase | -6.08 | 0.0004 |
| AB57_RS05710 | AB57_1134 | chemotaxis protein | -6.07 | 0.0001 |
| AB57_RS13435 | AB57_2708 | MULTISPECIES: hypothetical protein | -6.07 | 0.0029 |
| AB57_RS03050 | AB57_0590 | gluconokinase | -6.07 | 0.0022 |
| AB57_RS14935 | AB57_3020 | pseudogene | -6.07 | 0.0043 |
| AB57_RS14550 | AB57_2941 | GTPase Obg | -6.06 | 0.0049 |
| AB57_RS06000 | AB57_1187 | MULTISPECIES: GntR family transcriptional regulator | -6.06 | 0.0009 |
| AB57_RS01010 | AB57_0198 | | -6.06 | 0.0052 |
| AB57_RS18580 | AB57_3748 | MULTISPECIES: hypothetical protein | -6.06 | 0.0004 |
| AB57_RS05705 | AB57_1133 | NADH pyrophosphatase | -6.06 | 0.0006 |
| AB57_RS18070 | AB57_3650 | hypothetical protein | -6.06 | 0.0045 |
| AB57_RS03135 | AB57_0607 | membrane protein | -6.05 | 0.0033 |
| AB57_RS02345 | AB57_0446 | primosomal protein Nv' | -6.05 | 0.0017 |
| AB57_RS18210 | AB57_3674 | fatty acyl-CoA reductase | -6.05 | 0.0008 |
| AB57_RS01635 | AB57_0311 | MULTISPECIES: NrdR family transcriptional regulator | -6.05 | 0.0041 |
| AB57_RS14705 | AB57_2971 | 4-amino-4-deoxychorismate lyase | -6.05 | 0.0010 |
| AB57_RS05105 | AB57_1004 | MULTISPECIES: BetI family transcriptional regulator | -6.05 | 0.0026 |
| AB57_RS05800 | AB57_1150 | MULTISPECIES: dihydroorotase | -6.05 | 0.0001 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS05375 | AB57_1061 | pseudogene | -6.04 | 0.0001 |
| AB57_RS14265 | AB57_2874 | phosphate ABC transporter permease | -6.04 | 0.0044 |
| AB57_RS19445 | AB57_3926 | MULTISPECIES: hypothetical protein | -6.03 | 0.0018 |
| AB57_RS14000 | AB57_2817 | MULTISPECIES: iron ABC transporter permease | -6.03 | 0.0002 |
| GenBank:repeat_r egion:NC_011586: 263572:269774 | | | -6.03 | 0.0026 |
| AB57_RS06345 | AB57_1256 | MULTISPECIES: hypothetical protein | -6.03 | 0.0036 |
| AB57_RS17280 | AB57_3491 | membrane protein | -6.03 | 0.0002 |
| AB57_RS01325 | AB57_RS01325 | pseudogene | -6.03 | 0.0007 |
| AB57_RS06765 | AB57_1341 | alkyl hydroperoxide reductase subunit C | -6.02 | 0.0016 |
| AB57_RS01300 | AB57_0256 | pseudogene | -6.02 | 0.0028 |
| AB57_RS14475 | AB57_2925 | peptidyl-prolyl cis-trans isomerase | -6.01 | 0.0023 |
| AB57_RS01055 | AB57_0207 | hypothetical protein | -6.01 | 0.0022 |
| AB57_RS06035 | AB57_1194 | salicylaldehyde dehydrogenase | -6.01 | 0.0014 |
| AB57_RS04970 | AB57_0974 | hypothetical protein | -6.01 | 0.0003 |
| AB57_RS03850 | AB57_0754 | hydrolase | -6.01 | 0.0031 |
| AB57_RS14875 | AB57_3007 | DNA-directed RNA polymerase sigma-70 factor | -6.01 | 0.0014 |
| AB57_RS01615 | AB57_0307 | pseudogene | -6.01 | 0.0028 |
| AB57_RS17070 | AB57_3453 | starvation protein A | -6.00 | 0.0006 |
| AB57_RS16885 | AB57_3419 | phosphopantothenoylecysteine decarboxylase | -6.00 | 0.0017 |
| AB57_RS18780 | AB57_3791 | orotate phosphoribosyltransferase | -6.00 | 0.0000 |
| AB57_RS14855 | AB57_3003 | peptidoglycan-binding protein | -6.00 | 0.0002 |
| AB57_RS16285 | AB57_3301 | membrane protein | -5.99 | 0.0003 |
| AB57_RS18660 | AB57_3766 | outer membrane protein | -5.99 | 0.0000 |
| AB57_RS11650 | AB57_2341 | HxlR family transcriptional regulator | -5.99 | 0.0035 |
| AB57_RS14345 | AB57_2893 | RNA pseudouridine synthase | -5.99 | 0.0001 |
| AB57_RS17525 | AB57_3537 | GTP-binding protein | -5.98 | 0.0053 |
| AB57_RS05625 | AB57_1116 | cell division protein ZapA | -5.98 | 0.0006 |
| AB57_RS17750 | AB57_3584 | N-succinylarginine dihydrolase | -5.98 | 0.0029 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS16530 | AB57_3346 | acetyl-CoA acetyltransferase | -5.98 | 0.0003 |
| AB57_RS03215 | AB57_0623 | hypothetical protein | -5.98 | 0.0000 |
| AB57_RS07555 | AB57_1504 | hemolysin | -5.97 | 0.0017 |
| AB57_RS08935 | AB57_1792 | phosphotransferase | -5.97 | 0.0048 |
| AB57_RS16500 | AB57_RS16500 | pseudogene | -5.97 | 0.0056 |
| AB57_RS14155 | AB57_2852 | pseudogene | -5.97 | 0.0044 |
| AB57_RS09965 | AB57_2002 | S-(hydroxymethyl)glutathione dehydrogenase | -5.97 | 0.0001 |
| AB57_RS18420 | AB57_3716 | hypothetical protein | -5.97 | 0.0010 |
| AB57_RS03795 | AB57_0743 | protein FilE | -5.97 | 0.0002 |
| AB57_RS17185 | AB57_3473 | hypothetical protein | -5.96 | 0.0030 |
| AB57_RS11845 | AB57_2384 | hypothetical protein | -5.96 | 0.0030 |
| AB57_RS04305 | AB57_0842 | tRNA-dihydrouridine synthase C | -5.96 | 0.0006 |
| AB57_RS16330 | AB57_3304 | hypothetical protein | -5.96 | 0.0006 |
| AB57_RS03300 | AB57_0644 | integrase | -5.95 | 0.0012 |
| AB57_RS00660 | AB57_0130 | short-chain dehydrogenase | -5.95 | 0.0023 |
| AB57_RS15510 | AB57_3141 | alcohol dehydrogenase | -5.95 | 0.0015 |
| AB57_RS17675 | AB57_3569 | pseudogene | -5.94 | 0.0084 |
| AB57_RS19375 | AB57_3915 | SNF family Na ⁺ -dependent transporter | -5.94 | 0.0006 |
| AB57_RS17805 | AB57_3596 | chromosome partitioning protein ParA | -5.94 | 0.0014 |
| AB57_RS18520 | AB57_3736 | membrane protein | -5.94 | 0.0005 |
| AB57_RS00590 | AB57_0117 | MULTISPECIES: transcriptional regulator | -5.94 | 0.0001 |
| AB57_RS18540 | AB57_3740 | NADPH:quinone oxidoreductase | -5.93 | 0.0004 |
| AB57_RS03055 | AB57_0591 | aldehyde dehydrogenase | -5.93 | 0.0000 |
| AB57_RS03560 | AB57_0697 | glutathione S-transferase, partial | -5.93 | 0.0063 |
| AB57_RS09585 | AB57_1925 | hypothetical protein | -5.93 | 0.0024 |
| AB57_RS14415 | AB57_2911 | protease | -5.92 | 0.0001 |
| AB57_RS09085 | AB57_1823 | AraC family transcriptional regulator | -5.92 | 0.0019 |
| AB57_RS16460 | AB57_3330 | DNA-3-methyladenine glycosylase | -5.92 | 0.0000 |
| AB57_RS02615 | AB57_0501 | AraC family transcriptional regulator | -5.92 | 0.0000 |
| AB57_RS04340 | AB57_0849 | chromosome segregation protein SMC | -5.91 | 0.0029 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS14785 | AB57_2989 | enol-CoA hydratase | -5.91 | 0.0030 |
| AB57_RS04355 | AB57_0852 | MULTISPECIES: bacterioferritin | -5.91 | 0.0001 |
| AB57_RS16895 | AB57_3422 | hypothetical protein | -5.91 | 0.0008 |
| AB57_RS04415 | AB57_0865 | hypothetical protein | -5.90 | 0.0022 |
| AB57_RS14330 | AB57_2890 | TetR family transcriptional regulator | -5.90 | 0.0014 |
| AB57_RS00725 | AB57_0143 | MULTISPECIES: LysR family transcriptional regulator | -5.89 | 0.0039 |
| AB57_RS17230 | AB57_3482 | hypothetical protein | -5.89 | 0.0001 |
| AB57_RS19060 | AB57_3851 | DNA-cytosine methyltransferase | -5.89 | 0.0012 |
| AB57_RS03620 | AB57_0709 | thioredoxin | -5.89 | 0.0030 |
| AB57_RS18230 | AB57_3678 | two-component system sensor histidine kinase | -5.89 | 0.0046 |
| AB57_RS17240 | AB57_3484 | tryptophan repressor-binding protein | -5.88 | 0.0003 |
| AB57_RS04180 | AB57_0816 | LysR family transcriptional regulator | -5.88 | 0.0032 |
| AB57_RS03945 | AB57_0769 | tricarballylate utilization protein B | -5.88 | 0.0030 |
| AB57_RS16050 | AB57_3253 | pseudogene | -5.88 | 0.0047 |
| AB57_RS19340 | AB57_3908 | transcriptional regulator | -5.87 | 0.0017 |
| AB57_RS12065 | AB57_2429 | helicase | -5.87 | 0.0000 |
| AB57_RS11595 | AB57_2331 | hypothetical protein | -5.87 | 0.0030 |
| AB57_RS17770 | AB57_3588 | glutamate dehydrogenase | -5.87 | 0.0045 |
| AB57_RS03295 | AB57_0641 | tRNA-dihydrouridine synthase A | -5.87 | 0.0015 |
| AB57_RS03860 | AB57_0756 | copper oxidase | -5.86 | 0.0025 |
| AB57_RS13765 | AB57_2773 | malic enzyme | -5.86 | 0.0025 |
| AB57_RS04015 | AB57_0783 | MULTISPECIES: flavin reductase | -5.86 | 0.0001 |
| AB57_RS17935 | AB57_3623 | pilus assembly protein PilW | -5.85 | 0.0006 |
| AB57_RS00120 | AB57_0024 | insertase | -5.85 | 0.0030 |
| AB57_RS14190 | AB57_2859 | peptidoglycan-binding protein | -5.85 | 0.0040 |
| AB57_RS16040 | AB57_3251 | repressor | -5.85 | 0.0001 |
| AB57_RS16155 | AB57_3274 | long-chain fatty acid transporter | -5.85 | 0.0021 |
| AB57_RS00575 | AB57_0114 | UDP-glucose 4-epimerase | -5.85 | 0.0046 |
| AB57_RS16900 | AB57_3423 | phosphoesterase | -5.85 | 0.0041 |
| AB57_RS02260 | AB57_0428 | hypothetical protein | -5.83 | 0.0020 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|--|---|----------------|
| AB57_RS19300 | AB57_3900 | hypothetical protein | -5.83 | 0.0044 |
| AB57_RS01520 | AB57_0291 | MULTISPECIES: aminoglycoside 3-N-acetyltransferase | -5.82 | 0.0001 |
| AB57_RS05700 | AB57_1132 | esterase | -5.82 | 0.0038 |
| AB57_RS03220 | AB57_0624 | acetolactate synthase | -5.82 | 0.0030 |
| AB57_RS00460 | AB57_0090 | peptidyl-prolyl cis-trans isomerase | -5.82 | 0.0004 |
| AB57_RS04310 | AB57_0843 | cytochrome C assembly protein | -5.81 | 0.0005 |
| AB57_RS03750 | AB57_0732 | UDP-N-acetylglucosamine 1-carboxyvinyltransferase | -5.80 | 0.0007 |
| AB57_RS16045 | AB57_3252 | hypothetical protein | -5.80 | 0.0001 |
| GenBank:repeat_r egion:NC_011586: 3356919:3357108 | | | -5.80 | 0.0001 |
| AB57_RS11340 | AB57_2278 | methionine aminopeptidase | -5.80 | 0.0006 |
| AB57_RS04475 | AB57_0878 | hypothetical protein | -5.79 | 0.0030 |
| AB57_RS14920 | AB57_3016 | hydrolase | -5.78 | 0.0002 |
| AB57_RS18800 | AB57_3795 | esterase | -5.78 | 0.0055 |
| AB57_RS17870 | AB57_3610 | MULTISPECIES: hemerythrin | -5.78 | 0.0003 |
| AB57_RS11235 | AB57_RS11235 | pseudogene | -5.77 | 0.0057 |
| AB57_RS15480 | AB57_3135 | MULTISPECIES: hypothetical protein | -5.77 | 0.0009 |
| AB57_RS08710 | AB57_1746 | pilus assembly protein | -5.77 | 0.0039 |
| AB57_RS10690 | AB57_2145 | TetR family transcriptional regulator | -5.77 | 0.0003 |
| AB57_RS15660 | AB57_3172 | two-component system sensor histidine kinase | -5.76 | 0.0003 |
| AB57_RS12420 | AB57_2505 | MULTISPECIES: cytochrome O ubiquinol oxidase | -5.76 | 0.0014 |
| AB57_RS12540 | AB57_2530 | hydroxyacylglutathione hydrolase | -5.76 | 0.0030 |
| AB57_RS17810 | AB57_3597 | membrane protein | -5.76 | 0.0009 |
| AB57_RS15175 | AB57_3071 | tRNA 2-thiocytidine biosynthesis protein TtcA | -5.76 | 0.0028 |
| AB57_RS15245 | AB57_3086 | glycerol-3-phosphate dehydrogenase | -5.75 | 0.0040 |
| AB57_RS18190 | AB57_3670 | hypothetical protein | -5.75 | 0.0055 |
| AB57_RS18395 | AB57_3711 | hypothetical protein | -5.74 | 0.0008 |
| AB57_RS04065 | AB57_0793 | hypothetical protein | -5.74 | 0.0003 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS15815 | AB57_3206 | hypothetical protein | -5.74 | 0.0002 |
| AB57_RS18935 | AB57_3822 | membrane protein | -5.74 | 0.0043 |
| AB57_RS11885 | AB57_2392 | hypothetical protein | -5.74 | 0.0005 |
| AB57_RS07485 | AB57_1491 | type VI secretion protein | -5.73 | 0.0018 |
| AB57_RS11185 | AB57_2246 | hypothetical protein | -5.73 | 0.0043 |
| AB57_RS08495 | AB57_1701 | glutathione peroxidase | -5.72 | 0.0008 |
| AB57_RS02195 | AB57_0415 | translation initiation factor IF-2 | -5.72 | 0.0051 |
| AB57_RS04955 | AB57_0970 | hypothetical protein | -5.72 | 0.0009 |
| AB57_RS14835 | AB57_2998 | gramicidin S synthase | -5.72 | 0.0016 |
| AB57_RS14355 | AB57_2895 | MULTISPECIES: glutamate-1-semialdehyde 2,1-aminomutase | -5.72 | 0.0030 |
| AB57_RS02270 | AB57_0430 | hypothetical protein | -5.71 | 0.0040 |
| AB57_RS01530 | | MULTISPECIES: hypothetical protein | -5.71 | 0.0003 |
| AB57_RS15765 | AB57_3193 | hypothetical protein | -5.71 | 0.0085 |
| AB57_RS09445 | AB57_1895 | hypothetical protein | -5.71 | 0.0001 |
| AB57_RS16735 | AB57_3390 | acyl-CoA dehydrogenase, partial | -5.71 | 0.0014 |
| AB57_RS18585 | AB57_3749 | histidine kinase | -5.71 | 0.0027 |
| AB57_RS03545 | AB57_0694 | acyl-CoA synthetase | -5.70 | 0.0015 |
| AB57_RS18085 | AB57_3653 | UDP-N-acetylmuramoyl-tripeptide--D-alanyl-D-alanine ligase | -5.70 | 0.0008 |
| AB57_RS04555 | AB57_0890 | pseudogene | -5.70 | 0.0003 |
| AB57_RS04980 | AB57_0976 | hypothetical protein | -5.70 | 0.0001 |
| AB57_RS02415 | AB57_0462 | hypothetical protein | -5.69 | 0.0021 |
| AB57_RS01215 | AB57_0239 | hypothetical protein | -5.69 | 0.0003 |
| AB57_RS03100 | AB57_0600 | MULTISPECIES: nucleoside diphosphate kinase | -5.69 | 0.0000 |
| AB57_RS10145 | AB57_2034 | hypothetical protein | -5.69 | 0.0008 |
| AB57_RS16595 | AB57_3360 | GNAT family acetyltransferase | -5.68 | 0.0001 |
| AB57_RS01870 | AB57_0358 | type II secretion system protein GspD | -5.68 | 0.0025 |
| AB57_RS08465 | AB57_1695 | RNA methyltransferase | -5.68 | 0.0001 |
| AB57_RS18500 | AB57_3732 | branched-chain amino acid ABC transporter permease | -5.67 | 0.0005 |
| AB57_RS18595 | AB57_3751 | MULTISPECIES: DNA-binding response regulator | -5.67 | 0.0035 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS01435 | | hypothetical protein | -5.67 | 0.0040 |
| AB57_RS18325 | AB57_3696 | aquaporin Z | -5.66 | 0.0030 |
| AB57_RS11850 | AB57_2385 | LysR family transcriptional regulator | -5.66 | 0.0030 |
| AB57_RS13510 | AB57_2723 | replication protein | -5.66 | 0.0030 |
| AB57_RS13295 | AB57_2682 | phage tail tape measure protein | -5.66 | 0.0000 |
| AB57_RS15315 | AB57_3097 | MULTISPECIES: ribosomal RNA large subunit methyltransferase E | -5.66 | 0.0040 |
| AB57_RS01400 | | MULTISPECIES: MerR family transcriptional regulator | -5.66 | 0.0002 |
| AB57_RS05770 | | hypothetical protein | -5.66 | 0.0034 |
| AB57_RS08480 | AB57_1698 | glutamate symport protein | -5.66 | 0.0001 |
| AB57_RS00190 | AB57_0039 | hypothetical protein | -5.65 | 0.0078 |
| AB57_RS05110 | AB57_1005 | choline transporter | -5.65 | 0.0065 |
| AB57_RS18365 | AB57_3705 | AraC family transcriptional regulator | -5.65 | 0.0036 |
| AB57_RS14460 | AB57_2922 | serine protease | -5.64 | 0.0001 |
| AB57_RS06005 | AB57_1188 | chlorogenate esterase | -5.64 | 0.0030 |
| AB57_RS03640 | AB57_0713 | pyrroline-5-carboxylate reductase | -5.64 | 0.0017 |
| AB57_RS12525 | AB57_2527 | hypothetical protein | -5.64 | 0.0001 |
| AB57_RS16335 | AB57_3305 | hypothetical protein | -5.63 | 0.0015 |
| AB57_RS19360 | AB57_3912 | methionyl-tRNA formyltransferase | -5.63 | 0.0003 |
| AB57_RS02210 | AB57_0418 | pesticidal protein Cry5Aa | -5.63 | 0.0008 |
| AB57_RS15940 | AB57_3231 | hypothetical protein | -5.63 | 0.0005 |
| AB57_RS06100 | AB57_1207 | AraC family transcriptional regulator | -5.62 | 0.0004 |
| AB57_RS13850 | AB57_2787 | aminopeptidase N | -5.62 | 0.0003 |
| AB57_RS13320 | AB57_2685 | hypothetical protein | -5.62 | 0.0025 |
| AB57_RS16680 | AB57_3379 | tryptophan synthase subunit beta | -5.61 | 0.0026 |
| AB57_RS04300 | AB57_0841 | hypothetical protein | -5.61 | 0.0002 |
| AB57_RS01670 | AB57_0318 | cytosol aminopeptidase | -5.61 | 0.0033 |
| AB57_RS01840 | AB57_0352 | hypothetical protein | -5.61 | 0.0009 |
| AB57_RS14150 | AB57_2850 | GTPase | -5.61 | 0.0022 |
| AB57_RS12970 | AB57_2613 | phosphohistidine phosphatase | -5.61 | 0.0009 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS04330 | AB57_0847 | membrane protein | -5.61 | 0.0003 |
| AB57_RS14400 | AB57_2908 | metal-binding protein | -5.60 | 0.0018 |
| AB57_RS13625 | AB57_2746 | NADPH-dependent 7-cyano-7-deazaguanine reductase | -5.60 | 0.0002 |
| AB57_RS09410 | AB57_1889 | 3-deoxy-7-phosphoheptulonate synthase | -5.60 | 0.0021 |
| AB57_RS03980 | AB57_0776 | amidohydrolase | -5.60 | 0.0075 |
| AB57_RS00865 | AB57_0169 | hypothetical protein | -5.59 | 0.0000 |
| AB57_RS10630 | AB57_2133 | GntR family transcriptional regulator | -5.59 | 0.0003 |
| AB57_RS18635 | AB57_3759 | 16S rRNA methyltransferase | -5.59 | 0.0038 |
| AB57_RS00545 | AB57_0108 | UDP-glucose 4-epimerase | -5.58 | 0.0004 |
| AB57_RS04560 | AB57_0891 | flavodoxin | -5.58 | 0.0005 |
| AB57_RS08825 | AB57_1770 | bifunctional proline dehydrogenase/pyrroline-5-carboxylate dehydrogenase | -5.58 | 0.0000 |
| AB57_RS11510 | AB57_2313 | membrane protein | -5.58 | 0.0001 |
| GenBank:repeat_r egion:NC_011586: 286568:286690 | | | -5.58 | 0.0034 |
| AB57_RS12290 | AB57_2477 | kinase | -5.57 | 0.0004 |
| AB57_RS19100 | AB57_3859 | histidine ammonia-lyase | -5.57 | 0.0000 |
| AB57_RS11740 | AB57_2361 | hypothetical protein | -5.57 | 0.0015 |
| AB57_RS01160 | AB57_0227 | MULTISPECIES: hypothetical protein | -5.56 | 0.0001 |
| AB57_RS18515 | AB57_3735 | MFS transporter | -5.56 | 0.0048 |
| AB57_RS14165 | AB57_2854 | bile acid:sodium symporter | -5.55 | 0.0062 |
| AB57_RS13080 | AB57_2636 | hypothetical protein | -5.55 | 0.0003 |
| AB57_RS19275 | AB57_3894 | LysR family transcriptional regulator | -5.54 | 0.0002 |
| AB57_RS19070 | AB57_3853 | glutamate racemase | -5.54 | 0.0080 |
| AB57_RS16865 | AB57_3415 | queuine tRNA-ribosyltransferase | -5.54 | 0.0000 |
| AB57_RS05845 | AB57_1158 | type VI secretion system protein | -5.54 | 0.0034 |
| AB57_RS18440 | AB57_4854 | tRNA-Thr | -5.54 | 0.0028 |
| AB57_RS03270 | AB57_0635 | hydrolase | -5.54 | 0.0028 |
| AB57_RS19090 | AB57_3857 | imidazolonepropionase | -5.54 | 0.0047 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS04225 | AB57_0825 | MFS transporter | -5.53 | 0.0001 |
| AB57_RS04090 | AB57_0798 | hypothetical protein | -5.53 | 0.0020 |
| GenBank:STS:NC_011586:736648:7 | D1S3689 | | -5.53 | 0.0029 |
| 37073 | | | | |
| AB57_RS11380 | AB57_2287 | PTS fructose transporter subunit IIBC | -5.53 | 0.0010 |
| AB57_RS05575 | AB57_1105 | endonuclease IV | -5.53 | 0.0027 |
| AB57_RS19175 | AB57_3874 | sodium transporter | -5.53 | 0.0023 |
| AB57_RS18265 | AB57_3683 | imidazole glycerol phosphate synthase subunit HisH | -5.53 | 0.0006 |
| AB57_RS13085 | AB57_2637 | NAD kinase | -5.53 | 0.0004 |
| AB57_RS10400 | AB57_2082 | AdeA/Adel family multidrug efflux RND transporter periplasmic adaptor subunit | -5.52 | 0.0031 |
| AB57_RS06645 | AB57_1316 | type I deoxyribonuclease HsdR | -5.52 | 0.0033 |
| AB57_RS18220 | AB57_3676 | RNA-binding transcriptional accessory protein | -5.52 | 0.0027 |
| AB57_RS17175 | AB57_3471 | oxidoreductase | -5.52 | 0.0053 |
| AB57_RS19225 | AB57_3884 | MULTISPECIES: membrane protein | -5.52 | 0.0006 |
| AB57_RS11490 | AB57_2309 | hypothetical protein | -5.52 | 0.0014 |
| AB57_RS18270 | AB57_3684 | hypothetical protein | -5.52 | 0.0004 |
| GenBank:repeat_region:NC_011586:283687:285283 | | | -5.51 | 0.0002 |
| AB57_RS18060 | AB57_3648 | pilus assembly protein PilM | -5.51 | 0.0006 |
| AB57_RS09895 | AB57_1987 | polyphosphate kinase | -5.51 | 0.0003 |
| AB57_RS08785 | AB57_1762 | transketolase | -5.51 | 0.0008 |
| AB57_RS05180 | AB57_1019 | hypothetical protein | -5.51 | 0.0014 |
| AB57_RS09550 | AB57_1918 | pseudogene | -5.50 | 0.0002 |
| AB57_RS17680 | AB57_3570 | tetracycline resistance MFS efflux pump | -5.50 | 0.0008 |
| AB57_RS02835 | AB57_0546 | phosphatase | -5.50 | 0.0024 |
| AB57_RS00795 | AB57_0157 | non-ribosomal peptide synthetase | -5.50 | 0.0030 |
| AB57_RS04295 | AB57_0840 | heme utilization protein | -5.49 | 0.0017 |
| AB57_RS05630 | AB57_1117 | hypothetical protein | -5.49 | 0.0003 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS18005 | AB57_3637 | hypothetical protein | -5.49 | 0.0001 |
| AB57_RS17310 | AB57_3494 | peptidase M3 | -5.49 | 0.0018 |
| AB57_RS08325 | AB57_1666 | ubiquinol oxidase subunit II, cyanide insensitive | -5.49 | 0.0069 |
| AB57_RS08895 | AB57_1784 | cupin | -5.48 | 0.0026 |
| AB57_RS02870 | AB57_0553 | hypothetical protein | -5.48 | 0.0033 |
| AB57_RS05785 | AB57_1148 | TonB-dependent receptor | -5.48 | 0.0010 |
| AB57_RS03035 | AB57_0587 | phosphogluconate dehydratase | -5.47 | 0.0004 |
| AB57_RS19285 | AB57_3897 | MFS transporter | -5.47 | 0.0000 |
| AB57_RS19255 | AB57_3890 | DNA-3-methyladenine glycosylase | -5.47 | 0.0000 |
| AB57_RS18160 | AB57_3664 | membrane protein | -5.47 | 0.0002 |
| AB57_RS04410 | AB57_0864 | serine protease | -5.47 | 0.0036 |
| GenBank:repeat_r egion:NC_011586: 588254:589436 | | | -5.47 | 0.0002 |
| AB57_RS02880 | AB57_0555 | MULTISPECIES: transposase | -5.46 | 0.0002 |
| AB57_RS16715 | AB57_3386 | hypothetical protein | -5.46 | 0.0009 |
| AB57_RS00620 | AB57_0123 | methylcitrate synthase | -5.46 | 0.0002 |
| AB57_RS06355 | AB57_1258 | MULTISPECIES: antirepressor | -5.45 | 0.0046 |
| AB57_RS07805 | AB57_1557 | carnitine dehydratase | -5.45 | 0.0023 |
| AB57_RS19115 | AB57_3862 | histidine utilization repressor | -5.45 | 0.0003 |
| AB57_RS00130 | AB57_0027 | transcriptional regulator | -5.45 | 0.0058 |
| AB57_RS01330 | AB57_0262 | MULTISPECIES: DNA topoisomerase I | -5.45 | 0.0017 |
| AB57_RS11170 | AB57_2243 | membrane protein | -5.44 | 0.0000 |
| AB57_RS19265 | AB57_3892 | GntR family transcriptional regulator | -5.44 | 0.0021 |
| AB57_RS09415 | AB57_1890 | hypothetical protein | -5.44 | 0.0036 |
| AB57_RS08885 | AB57_1782 | ArsR family transcriptional regulator | -5.43 | 0.0043 |
| AB57_RS03590 | AB57_0703 | hypothetical protein | -5.43 | 0.0001 |
| AB57_RS13300 | | hypothetical protein | -5.43 | 0.0053 |
| AB57_RS01180 | AB57_0231 | MFS transporter | -5.42 | 0.0020 |
| AB57_RS01795 | AB57_0343 | membrane protein | -5.42 | 0.0008 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS18485 | AB57_3729 | decarboxylase | -5.42 | 0.0027 |
| AB57_RS03805 | AB57_0745 | ADP-ribose pyrophosphatase | -5.42 | 0.0001 |
| AB57_RS17970 | AB57_3630 | reactive intermediate/imine deaminase | -5.42 | 0.0030 |
| AB57_RS01630 | AB57_0310 | ammonia channel protein | -5.42 | 0.0018 |
| AB57_RS14195 | AB57_2860 | 5'-nucleotidase surE | -5.42 | 0.0026 |
| AB57_RS01415 | AB57_0275 | MULTISPECIES: tetracycline resistance MFS efflux pump | -5.42 | 0.0004 |
| AB57_RS01790 | AB57_0342 | transcriptional regulator | -5.41 | 0.0005 |
| AB57_RS11205 | AB57_2251 | ribose-5-phosphate isomerase A | -5.41 | 0.0008 |
| AB57_RS16795 | AB57_3402 | hypothetical protein | -5.41 | 0.0005 |
| AB57_RS02580 | AB57_0494 | 3-isopropylmalate dehydratase large subunit | -5.40 | 0.0000 |
| AB57_RS19040 | AB57_3844 | bifunctional protein GlnU | -5.40 | 0.0016 |
| AB57_RS09065 | AB57_1819 | membrane protein | -5.40 | 0.0008 |
| AB57_RS19240 | AB57_3887 | A/G-specific adenine glycosylase | -5.39 | 0.0013 |
| AB57_RS04700 | AB57_0918 | metal-dependent hydrolase | -5.39 | 0.0027 |
| AB57_RS19130 | AB57_3865 | hypothetical protein | -5.38 | 0.0001 |
| AB57_RS04565 | AB57_0892 | MULTISPECIES: SsrA-binding protein | -5.38 | 0.0017 |
| AB57_RS03970 | AB57_0774 | MULTISPECIES: LysR family transcriptional regulator | -5.37 | 0.0003 |
| AB57_RS02100 | AB57_0399 | secretion protein HlyD | -5.37 | 0.0018 |
| AB57_RS15570 | AB57_3153 | solanesyl diphosphate synthase | -5.36 | 0.0015 |
| AB57_RS03330 | AB57_0649 | MULTISPECIES: hypothetical protein | -5.36 | 0.0036 |
| AB57_RS02700 | AB57_0519 | MarR family transcriptional regulator | -5.36 | 0.0022 |
| AB57_RS13400 | AB57_2701 | hypothetical protein | -5.35 | 0.0051 |
| AB57_RS17285 | AB57_3492 | hydrolase | -5.35 | 0.0050 |
| AB57_RS01810 | AB57_0346 | Fe(2+)-trafficking protein | -5.35 | 0.0047 |
| AB57_RS14130 | AB57_2846 | MULTISPECIES: hypothetical protein | -5.35 | 0.0009 |
| AB57_RS05090 | AB57_1001 | malate:quinone oxidoreductase | -5.35 | 0.0003 |
| AB57_RS18810 | AB57_3796 | thiol:disulfide interchange protein | -5.35 | 0.0039 |
| AB57_RS00400 | AB57_0077 | alkali-inducible disulfide interchange protein | -5.35 | 0.0016 |
| AB57_RS00835 | AB57_0163 | hypothetical protein | -5.35 | 0.0014 |
| AB57_RS16125 | | AraC family transcriptional regulator | -5.34 | 0.0025 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|--|---|----------------|
| AB57_RS17000 | AB57_3439 | membrane protein | -5.34 | 0.0011 |
| AB57_RS00560 | AB57_0111 | UTP--glucose-1-phosphate uridylyltransferase | -5.34 | 0.0006 |
| AB57_RS03480 | AB57_0681 | 3\'-5\' exonuclease | -5.34 | 0.0054 |
| AB57_RS02250 | AB57_0426 | oxidoreductase | -5.34 | 0.0007 |
| AB57_RS03275 | AB57_0636 | aconitate hydratase | -5.34 | 0.0006 |
| AB57_RS12170 | AB57_2452 | 16S rRNA pseudouridine(516) synthase | -5.34 | 0.0015 |
| AB57_RS17650 | AB57_3563 | acyl-CoA dehydrogenase | -5.33 | 0.0008 |
| GenBank:repeat_r egion:NC_011586: 2911053:2911200 | | | -5.33 | 0.0023 |
| AB57_RS16560 | AB57_3353 | porin | -5.33 | 0.0005 |
| AB57_RS10625 | AB57_2132 | calcium-binding protein | -5.32 | 0.0030 |
| AB57_RS19260 | AB57_3891 | alcohol dehydrogenase | -5.32 | 0.0039 |
| AB57_RS15395 | AB57_3116 | neutral zinc metallopeptidase | -5.32 | 0.0001 |
| AB57_RS14760 | AB57_2983 | membrane protein | -5.32 | 0.0027 |
| AB57_RS18610 | AB57_3754 | transcriptional regulator | -5.32 | 0.0030 |
| AB57_RS00180 | AB57_0037 | membrane protein | -5.31 | 0.0030 |
| AB57_RS18285 | AB57_3688 | Cro/C1 family transcriptional regulator | -5.31 | 0.0000 |
| AB57_RS13115 | AB57_2644 | MULTISPECIES: DNA-binding response regulator | -5.31 | 0.0003 |
| AB57_RS16260 | AB57_3296 | hypothetical protein | -5.31 | 0.0005 |
| AB57_RS02965 | AB57_0573 | hypothetical protein | -5.30 | 0.0008 |
| AB57_RS10570 | AB57_2121 | LysR family transcriptional regulator | -5.30 | 0.0016 |
| AB57_RS00150 | AB57_0031 | MULTISPECIES: hypoxanthine phosphoribosyltransferase | -5.29 | 0.0041 |
| AB57_RS06105 | AB57_1208 | transferase | -5.29 | 0.0014 |
| AB57_RS00825 | AB57_4836 | tRNA-Gly | -5.29 | 0.0030 |
| AB57_RS08125 | AB57_1624 | lysine transporter LysE | -5.29 | 0.0030 |
| AB57_RS18355 | AB57_4813 | tRNA-Ser | -5.28 | 0.0010 |
| AB57_RS14615 | AB57_2954 | hypothetical protein | -5.28 | 0.0030 |
| AB57_RS17215 | AB57_3479 | endoribonuclease YbeY | -5.28 | 0.0022 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS03465 | AB57_0677 | enoyl-CoA hydratase | -5.27 | 0.0001 |
| AB57_RS02315 | AB57_0440 | magnesium transporter | -5.27 | 0.0017 |
| AB57_RS12545 | AB57_2531 | MULTISPECIES: hypothetical protein | -5.27 | 0.0041 |
| AB57_RS03615 | AB57_0708 | MULTISPECIES: transcription termination factor Rho | -5.27 | 0.0030 |
| AB57_RS01060 | AB57_0208 | hypothetical protein | -5.26 | 0.0001 |
| AB57_RS15010 | AB57_3037 | DNA gyrase subunit A | -5.26 | 0.0006 |
| AB57_RS07135 | AB57_1415 | ATP-binding protein | -5.26 | 0.0000 |
| AB57_RS02560 | AB57_0490 | RNA pyrophosphohydrolase | -5.26 | 0.0001 |
| AB57_RS15690 | AB57_3178 | esterase | -5.25 | 0.0048 |
| AB57_RS16065 | AB57_3257 | hypothetical protein | -5.24 | 0.0030 |
| AB57_RS04085 | AB57_0797 | two-component system sensor histidine kinase | -5.24 | 0.0037 |
| AB57_RS17945 | AB57_3625 | fimbrial protein | -5.24 | 0.0006 |
| AB57_RS02500 | AB57_0477 | long-chain acyl-CoA synthetase | -5.23 | 0.0008 |
| AB57_RS04425 | AB57_0867 | pseudogene | -5.22 | 0.0027 |
| AB57_RS14980 | AB57_3031 | multidrug ABC transporter permease | -5.21 | 0.0006 |
| AB57_RS01145 | AB57_0223 | DNA polymerase IV | -5.21 | 0.0022 |
| AB57_RS10965 | AB57_2200 | MarR family transcriptional regulator | -5.21 | 0.0030 |
| AB57_RS00920 | AB57_0180 | pseudogene | -5.21 | 0.0002 |
| AB57_RS17005 | AB57_3440 | MULTISPECIES: membrane protein | -5.20 | 0.0002 |
| AB57_RS09905 | AB57_1990 | MULTISPECIES: pyrroloquinoline-quinone synthase | -5.20 | 0.0003 |
| AB57_RS14810 | AB57_2993 | deoxyguanosinetriphosphate triphosphohydrolase | -5.20 | 0.0025 |
| AB57_RS16685 | AB57_3380 | N-(5'-phosphoribosyl)anthranilate isomerase | -5.19 | 0.0007 |
| AB57_RS06760 | AB57_1340 | hypothetical protein | -5.19 | 0.0008 |
| AB57_RS06750 | AB57_1338 | MULTISPECIES: hypothetical protein | -5.19 | 0.0043 |
| AB57_RS07160 | AB57_1420 | O-methyltransferase | -5.18 | 0.0058 |
| AB57_RS13190 | AB57_2660 | transcriptional regulator | -5.18 | 0.0018 |
| AB57_RS11735 | AB57_2360 | acetyl-CoA carboxylase biotin carboxylase subunit | -5.18 | 0.0020 |
| AB57_RS18550 | AB57_3742 | ABC-ATPase UvrA | -5.17 | 0.0044 |
| AB57_RS05350 | AB57_1056 | arylsulfatase | -5.16 | 0.0030 |
| AB57_RS18450 | AB57_3722 | methyltransferase | -5.15 | 0.0026 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS02480 | AB57_0474 | radical SAM protein | -5.15 | 0.0053 |
| AB57_RS01980 | AB57_0375 | sodium:proton antiporter | -5.15 | 0.0000 |
| AB57_RS16930 | AB57_3429 | nucleotide-binding protein | -5.15 | 0.0033 |
| AB57_RS10545 | AB57_2114 | nicotinate phosphoribosyltransferase | -5.14 | 0.0015 |
| AB57_RS19455 | AB57_3928 | MULTISPECIES: hypothetical protein | -5.14 | 0.0007 |
| AB57_RS05695 | AB57_1131 | MULTISPECIES: hypothetical protein | -5.14 | 0.0051 |
| AB57_RS15820 | AB57_3207 | hypothetical protein | -5.14 | 0.0006 |
| AB57_RS13870 | AB57_2791 | MULTISPECIES: hypothetical protein | -5.13 | 0.0001 |
| AB57_RS18470 | AB57_3726 | LysR family transcriptional regulator | -5.13 | 0.0015 |
| AB57_RS16015 | AB57_3243 | hypothetical protein | -5.13 | 0.0015 |
| AB57_RS07990 | AB57_1595 | MFS transporter | -5.13 | 0.0002 |
| AB57_RS19430 | AB57_3923 | MULTISPECIES: hypothetical protein | -5.13 | 0.0010 |
| AB57_RS17015 | AB57_3442 | phospholipase | -5.13 | 0.0013 |
| AB57_RS18820 | AB57_3799 | hypothetical protein | -5.13 | 0.0046 |
| AB57_RS00610 | AB57_0121 | MULTISPECIES: GntR family transcriptional regulator | -5.13 | 0.0003 |
| AB57_RS07025 | AB57_1392 | hypothetical protein | -5.12 | 0.0049 |
| AB57_RS00095 | AB57_0019 | DNA polymerase III subunit beta | -5.12 | 0.0050 |
| AB57_RS11820 | AB57_2379 | TetR family transcriptional regulator | -5.11 | 0.0056 |
| AB57_RS09095 | AB57_1825 | malate synthase G | -5.11 | 0.0021 |
| AB57_RS17225 | AB57_3481 | hypothetical protein | -5.11 | 0.0001 |
| AB57_RS04600 | AB57_0898 | betaine/carnitine/choline BCCT transporter | -5.10 | 0.0015 |
| AB57_RS16450 | AB57_3328 | oxidoreductase | -5.10 | 0.0058 |
| AB57_RS17190 | AB57_3474 | malate dehydrogenase | -5.09 | 0.0002 |
| AB57_RS03205 | AB57_0621 | hypothetical protein | -5.09 | 0.0017 |
| AB57_RS09555 | AB57_1919 | MULTISPECIES: ATPase AAA | -5.09 | 0.0016 |
| AB57_RS16775 | AB57_3398 | MULTISPECIES: aspartate--tRNA(Asp/Asn) ligase | -5.09 | 0.0030 |
| AB57_RS01580 | | MULTISPECIES: MerR family transcriptional regulator | -5.09 | 0.0016 |
| AB57_RS19210 | AB57_3881 | ATPase AAA | -5.09 | 0.0002 |
| AB57_RS05690 | AB57_1130 | MULTISPECIES: hypothetical protein | -5.08 | 0.0027 |
| AB57_RS14675 | AB57_2965 | thioesterase | -5.08 | 0.0016 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS08455 | AB57_1693 | hypothetical protein | -5.08 | 0.0006 |
| AB57_RS15530 | AB57_3145 | RNA methyltransferase | -5.08 | 0.0003 |
| AB57_RS12485 | AB57_2518 | transcriptional regulator | -5.07 | 0.0035 |
| AB57_RS05435 | AB57_1074 | MULTISPECIES: rubredoxin | -5.07 | 0.0042 |
| AB57_RS07910 | AB57_1579 | nitronate monooxygenase | -5.07 | 0.0048 |
| AB57_RS00730 | AB57_0144 | methylmalonate-semialdehyde dehydrogenase | -5.06 | 0.0001 |
| AB57_RS05850 | AB57_1159 | type IV secretion protein Rhs | -5.06 | 0.0000 |
| AB57_RS12190 | AB57_2457 | hypothetical protein | -5.06 | 0.0030 |
| AB57_RS03095 | AB57_0599 | MULTISPECIES: FeS assembly protein IscX | -5.06 | 0.0056 |
| AB57_RS14135 | AB57_2847 | carbon starvation protein A | -5.05 | 0.0013 |
| AB57_RS02525 | AB57_0483 | hypothetical protein | -5.05 | 0.0002 |
| AB57_RS04900 | AB57_0959 | integrase | -5.05 | 0.0009 |
| AB57_RS12150 | AB57_2449 | tRNA threonylcarbamoyladenosine biosynthesis protein TsaE | -5.05 | 0.0051 |
| AB57_RS05075 | AB57_0998 | arginine:ornithine antiporter | -5.05 | 0.0005 |
| AB57_RS18995 | AB57_3834 | hypothetical protein | -5.04 | 0.0003 |
| AB57_RS06700 | AB57_1327 | aspartate carbamoyltransferase | -5.04 | 0.0052 |
| AB57_RS18795 | AB57_3794 | hypothetical protein | -5.03 | 0.0017 |
| AB57_RS03410 | AB57_0666 | MULTISPECIES: hypothetical protein | -5.03 | 0.0023 |
| AB57_RS03395 | AB57_0663 | hypothetical protein | -5.03 | 0.0008 |
| AB57_RS17055 | AB57_3450 | 4-hydroxythreonine-4-phosphate dehydrogenase | -5.03 | 0.0005 |
| AB57_RS06095 | AB57_1206 | 4-hydroxyacetophenone monooxygenase | -5.03 | 0.0009 |
| AB57_RS06770 | AB57_1342 | ATP-dependent helicase | -5.03 | 0.0000 |
| AB57_RS05555 | AB57_1100 | penicillin-binding protein 2 | -5.03 | 0.0017 |
| AB57_RS18000 | AB57_3636 | hypothetical protein | -5.02 | 0.0036 |
| AB57_RS18320 | AB57_3695 | AraC family transcriptional regulator | -5.01 | 0.0042 |
| AB57_RS12775 | AB57_2577 | hypothetical protein | -5.01 | 0.0030 |
| AB57_RS09515 | AB57_1909 | MULTISPECIES: hypothetical protein | -5.01 | 0.0054 |
| AB57_RS00570 | AB57_0113 | glucose-6-phosphate isomerase | -5.01 | 0.0001 |
| AB57_RS19335 | AB57_3907 | pyrimidine permease | -5.01 | 0.0003 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS16505 | AB57_3341 | lysine transporter | -5.01 | 0.0058 |
| AB57_RS17220 | AB57_3480 | cell envelope biogenesis protein TonB | -5.00 | 0.0001 |
| AB57_RS04400 | AB57_0862 | ribosomal large subunit pseudouridine synthase B | -5.00 | 0.0012 |
| AB57_RS18490 | AB57_3730 | GABA permease | -5.00 | 0.0039 |
| AB57_RS14685 | AB57_2967 | L-aspartate oxidase | -5.00 | 0.0016 |
| AB57_RS00540 | AB57_0107 | hypothetical protein | -5.00 | 0.0000 |
| AB57_RS16030 | AB57_3247 | DNA replication protein DnaD | -5.00 | 0.0022 |
| AB57_RS14510 | AB57_2933 | UDP-N-acetyl glucosamine 2-epimerase | -4.99 | 0.0019 |
| AB57_RS05175 | AB57_1018 | nucleoside transporter | -4.99 | 0.0002 |
| AB57_RS18400 | AB57_3712 | TetR family transcriptional regulator | -4.99 | 0.0008 |
| AB57_RS19125 | AB57_3864 | hypothetical protein | -4.98 | 0.0004 |
| AB57_RS15055 | AB57_3046 | alanine glycine permease | -4.98 | 0.0002 |
| AB57_RS16175 | AB57_3278 | MULTISPECIES: hypothetical protein | -4.98 | 0.0005 |
| AB57_RS01420 | AB57_0276 | MULTISPECIES: multidrug transporter | -4.98 | 0.0027 |
| AB57_RS14850 | AB57_3002 | translocation protein TolB | -4.98 | 0.0011 |
| AB57_RS02945 | AB57_0569 | preprotein translocase subunit TatB | -4.98 | 0.0041 |
| AB57_RS18555 | AB57_3743 | diguanylate cyclase | -4.97 | 0.0018 |
| AB57_RS00405 | AB57_0078 | TetR family transcriptional regulator | -4.97 | 0.0015 |
| AB57_RS02595 | AB57_0497 | 3-isopropylmalate dehydrogenase | -4.96 | 0.0033 |
| AB57_RS18310 | AB57_3693 | MULTISPECIES: imidazole glycerol phosphate synthase subunit HisF | -4.96 | 0.0002 |
| AB57_RS15585 | AB57_3156 | AdeA/Adel family multidrug efflux RND transporter periplasmic adaptor subunit | -4.96 | 0.0003 |
| AB57_RS05560 | AB57_1101 | hypothetical protein | -4.96 | 0.0013 |
| AB57_RS05440 | AB57_1075 | ATPase | -4.96 | 0.0055 |
| AB57_RS15100 | AB57_3057 | TetR family transcriptional regulator | -4.96 | 0.0020 |
| AB57_RS07130 | AB57_1414 | RND transporter | -4.95 | 0.0077 |
| AB57_RS17795 | AB57_3594 | hypothetical protein | -4.95 | 0.0044 |
| AB57_RS04380 | AB57_0857 | adenosylmethionine--8-amino-7-oxononanoate aminotransferase BioA | -4.95 | 0.0008 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS00350 | AB57_0067 | MULTISPECIES: sulfonate ABC transporter | -4.95 | 0.0003 |
| AB57_RS18010 | AB57_3638 | metal-dependent hydrolase | -4.95 | 0.0001 |
| AB57_RS18135 | AB57_3661 | pseudogene | -4.94 | 0.0032 |
| AB57_RS14905 | AB57_3013 | permease | -4.94 | 0.0044 |
| AB57_RS17545 | AB57_3541 | glycerol-3-phosphate acyltransferase | -4.94 | 0.0030 |
| AB57_RS03955 | AB57_0771 | LysR family transcriptional regulator | -4.94 | 0.0036 |
| AB57_RS05660 | AB57_1124 | MerR family transcriptional regulator | -4.94 | 0.0030 |
| AB57_RS14500 | AB57_2931 | hypothetical protein | -4.93 | 0.0031 |
| AB57_RS16695 | AB57_3382 | ATP--cobalamin adenosyltransferase | -4.93 | 0.0014 |
| AB57_RS12780 | AB57_2578 | LysR family transcriptional regulator | -4.93 | 0.0039 |
| AB57_RS17020 | AB57_3443 | acyltransferase | -4.93 | 0.0002 |
| AB57_RS15705 | AB57_3181 | hypothetical protein | -4.93 | 0.0011 |
| AB57_RS04210 | AB57_0822 | hypothetical protein | -4.92 | 0.0050 |
| AB57_RS02940 | | preprotein translocase | -4.92 | 0.0050 |
| AB57_RS01745 | AB57_0333 | hypothetical protein | -4.92 | 0.0003 |
| AB57_RS08970 | AB57_1799 | ATP-binding protein | -4.92 | 0.0030 |
| AB57_RS05655 | AB57_1123 | hypothetical protein | -4.90 | 0.0002 |
| AB57_RS05585 | AB57_1107 | glutamate dehydrogenase | -4.89 | 0.0000 |
| AB57_RS15915 | AB57_3226 | hypothetical protein | -4.89 | 0.0030 |
| AB57_RS15060 | AB57_3048 | DNA repair protein RadA | -4.89 | 0.0014 |
| AB57_RS04760 | AB57_0931 | acyltransferase | -4.89 | 0.0001 |
| AB57_RS02390 | AB57_0455 | electron transfer flavoprotein-ubiquinone oxidoreductase | -4.89 | 0.0008 |
| AB57_RS15535 | AB57_3146 | sirohydrochlorin ferrochelatase | -4.88 | 0.0014 |
| AB57_RS13065 | AB57_2633 | acyl-CoA dehydrogenase | -4.88 | 0.0030 |
| AB57_RS15195 | AB57_3075 | multidrug efflux RND transporter permease subunit | -4.87 | 0.0012 |
| AB57_RS00625 | AB57_0124 | MULTISPECIES: Fe/S-dependent 2-methylisocitrate dehydratase AcnD | -4.87 | 0.0009 |
| AB57_RS17205 | AB57_3477 | tRNA-2-methylthio-N(6)-dimethylallyladenosine synthase | -4.87 | 0.0002 |
| AB57_RS12155 | AB57_2450 | pseudouridylate synthase | -4.87 | 0.0050 |
| AB57_RS17085 | AB57_3456 | 3-methylcrotonyl-CoA carboxylase subunit alpha | -4.87 | 0.0030 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|--|---|----------------|
| AB57_RS06535 | AB57_1294 | 11 methyltransferase | -4.86 | 0.0003 |
| AB57_RS13535 | AB57_2729 | hypothetical protein | -4.86 | 0.0008 |
| AB57_RS02430 | AB57_0464 | flavoprotein | -4.86 | 0.0038 |
| AB57_RS14600 | AB57_2951 | aspartate aminotransferase | -4.85 | 0.0001 |
| AB57_RS04890 | AB57_0957 | multidrug resistance protein B | -4.85 | 0.0008 |
| GenBank:repeat_r egion:NC_011586: 2802623:2802812 | | | -4.84 | 0.0030 |
| AB57_RS06435 | AB57_1272 | pseudogene | -4.84 | 0.0006 |
| AB57_RS01335 | | MULTISPECIES: hypothetical protein | -4.84 | 0.0006 |
| AB57_RS03090 | AB57_0598 | phosphatidylglycerophosphatase | -4.84 | 0.0004 |
| AB57_RS14975 | AB57_3030 | putative RND family drug transporter | -4.84 | 0.0008 |
| AB57_RS16725 | AB57_3388 | two-component system sensor histidine kinase | -4.83 | 0.0003 |
| AB57_RS18645 | AB57_3762 | alkene reductase | -4.83 | 0.0001 |
| AB57_RS10395 | AB57_2081 | AdeB family multidrug efflux RND transporter permease subunit | -4.83 | 0.0002 |
| AB57_RS14630 | AB57_2956 | hypothetical protein | -4.83 | 0.0068 |
| AB57_RS13635 | AB57_2748 | cell wall shape-determining protein | -4.82 | 0.0017 |
| AB57_RS04395 | AB57_0861 | dithiobiotin synthetase | -4.82 | 0.0025 |
| AB57_RS02275 | AB57_0431 | pseudogene | -4.81 | 0.0003 |
| AB57_RS13710 | AB57_2762 | pyridine nucleotide-disulfide oxidoreductase | -4.81 | 0.0012 |
| AB57_RS05675 | AB57_1127 | LysR family transcriptional regulator | -4.80 | 0.0036 |
| AB57_RS19055 | AB57_RS19055 | pseudogene | -4.80 | 0.0001 |
| AB57_RS18350 | AB57_3703 | pseudogene | -4.80 | 0.0051 |
| AB57_RS06140 | AB57_1215 | short-chain dehydrogenase | -4.80 | 0.0030 |
| AB57_RS03930 | AB57_0766 | acyl-CoA dehydrogenase | -4.80 | 0.0030 |
| AB57_RS18825 | AB57_3800 | MULTISPECIES: hypothetical protein | -4.79 | 0.0017 |
| AB57_RS04190 | AB57_0818 | hypothetical protein | -4.79 | 0.0003 |
| AB57_RS11370 | AB57_2285 | PTS fructose transporter subunit IIA | -4.78 | 0.0012 |
| AB57_RS06060 | AB57_1199 | Vanillate O-demethylase oxidoreductase | -4.78 | 0.0030 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS00595 | AB57_0118 | MULTISPECIES: alpha-hydroxy-acid oxidizing enzyme | -4.78 | 0.0017 |
| AB57_RS03060 | AB57_0592 | gamma-glutamyl phosphate reductase | -4.78 | 0.0000 |
| AB57_RS02280 | AB57_0433 | hypothetical protein | -4.77 | 0.0011 |
| AB57_RS12340 | AB57_2488 | peptide chain release factor 1 | -4.77 | 0.0030 |
| AB57_RS17500 | AB57_3532 | glutamine amidotransferase | -4.77 | 0.0030 |
| AB57_RS15090 | AB57_3054 | hypothetical protein | -4.77 | 0.0017 |
| AB57_RS07420 | AB57_1478 | hypothetical protein | -4.76 | 0.0008 |
| AB57_RS15190 | AB57_3074 | hypothetical protein | -4.76 | 0.0002 |
| AB57_RS08505 | AB57_1703 | amino acid transporter | -4.76 | 0.0030 |
| AB57_RS15285 | AB57_3092 | alkaline phosphatase | -4.76 | 0.0025 |
| AB57_RS09135 | AB57_1833 | heme-binding protein | -4.75 | 0.0008 |
| AB57_RS01720 | AB57_0328 | homoserine dehydrogenase | -4.75 | 0.0001 |
| AB57_RS13480 | AB57_2715 | hypothetical protein | -4.75 | 0.0008 |
| AB57_RS13485 | AB57_2716 | hypothetical protein | -4.75 | 0.0008 |
| AB57_RS18075 | AB57_3651 | hypothetical protein | -4.74 | 0.0030 |
| AB57_RS18145 | AB57_4799 | tRNA-Glu | -4.74 | 0.0030 |
| AB57_RS14825 | AB57_2996 | N-acyl-L-amino acid amidohydrolase | -4.74 | 0.0003 |
| AB57_RS03320 | AB57_0647 | hypothetical protein | -4.74 | 0.0037 |
| AB57_RS17800 | AB57_3595 | endonuclease | -4.74 | 0.0001 |
| AB57_RS05155 | AB57_1014 | poly-beta-1,6-N-acetyl-D-glucosamine N-deacetylase PgaB | -4.74 | 0.0001 |
| AB57_RS02265 | AB57_0429 | SAM-dependent methyltransferase | -4.74 | 0.0069 |
| AB57_RS18170 | AB57_3666 | cation transporter | -4.74 | 0.0015 |
| AB57_RS00600 | AB57_0119 | lactate dehydrogenase | -4.73 | 0.0013 |
| AB57_RS11855 | AB57_2386 | transaldolase | -4.73 | 0.0005 |
| AB57_RS05315 | AB57_1047 | transketolase, partial | -4.73 | 0.0030 |
| AB57_RS04385 | AB57_0859 | pseudogene | -4.73 | 0.0034 |
| AB57_RS02160 | AB57_0411 | triosephosphate isomerase | -4.72 | 0.0003 |
| AB57_RS16690 | AB57_3381 | ligand-gated channel protein | -4.72 | 0.0031 |
| AB57_RS02045 | AB57_0388 | hypothetical protein | -4.72 | 0.0038 |
| AB57_RS11175 | AB57_2244 | ATP-dependent Clp protease ATP-binding subunit ClpA | -4.72 | 0.0022 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|--|---|----------------|
| AB57_RS19135 | AB57_3866 | amino acid transporter | -4.71 | 0.0005 |
| AB57_RS11515 | AB57_2314 | membrane protein | -4.70 | 0.0038 |
| AB57_RS17555 | AB57_3543 | nucleoside-diphosphate sugar epimerase | -4.70 | 0.0093 |
| AB57_RS05985 | AB57_1183 | glucarate dehydratase | -4.70 | 0.0030 |
| AB57_RS04960 | AB57_0971 | hypothetical protein | -4.69 | 0.0025 |
| AB57_RS03105 | AB57_0601 | 23S rRNA (adenine(2503)-C(2))-methyltransferase RImN | -4.69 | 0.0006 |
| AB57_RS01340 | | MULTISPECIES: hypothetical protein | -4.68 | 0.0009 |
| AB57_RS09980 | AB57_2005 | fimbrial protein | -4.68 | 0.0011 |
| AB57_RS06055 | AB57_1198 | MULTISPECIES: GntR family transcriptional regulator | -4.68 | 0.0030 |
| AB57_RS04230 | AB57_0826 | TetR family transcriptional regulator | -4.67 | 0.0091 |
| AB57_RS19215 | AB57_3882 | glucose dehydrogenase | -4.67 | 0.0007 |
| AB57_RS14470 | AB57_2924 | NADH dehydrogenase | -4.67 | 0.0017 |
| AB57_RS14290 | AB57_2879 | aldehyde dehydrogenase | -4.67 | 0.0030 |
| GenBank:repeat_r egion:NC_011586: 285536:285658 | | | -4.67 | 0.0008 |
| AB57_RS10640 | AB57_2135 | hydrolase | -4.66 | 0.0030 |
| AB57_RS01995 | AB57_0378 | heat-shock protein Hsp90 | -4.66 | 0.0010 |
| AB57_RS04070 | AB57_0794 | ribonucleotide-diphosphate reductase subunit alpha | -4.66 | 0.0003 |
| AB57_RS02285 | AB57_0434 | exonuclease V subunit gamma | -4.66 | 0.0003 |
| AB57_RS01830 | AB57_0350 | porphobilinogen deaminase | -4.65 | 0.0008 |
| AB57_RS11570 | AB57_2326 | ornithine carbamoyltransferase | -4.65 | 0.0094 |
| AB57_RS18685 | AB57_3771 | ferredoxin | -4.65 | 0.0034 |
| AB57_RS19085 | AB57_3856 | formimidoylglutamase | -4.65 | 0.0023 |
| AB57_RS15045 | AB57_3044 | diaminopimelate decarboxylase | -4.65 | 0.0013 |
| AB57_RS18950 | AB57_3825 | short-chain dehydrogenase | -4.64 | 0.0010 |
| AB57_RS14505 | AB57_2932 | hypothetical protein | -4.64 | 0.0030 |
| AB57_RS16255 | AB57_3294 | transporter | -4.64 | 0.0020 |
| AB57_RS01175 | AB57_0230 | membrane protein | -4.64 | 0.0003 |
| AB57_RS18150 | AB57_4800 | tRNA-Glu | -4.64 | 0.0030 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS13755 | AB57_2771 | MULTISPECIES: transposase | -4.64 | 0.0090 |
| GenBank:repeat_r egion:NC_011586: 2837509:2838692 | | | -4.64 | 0.0090 |
| AB57_RS09510 | AB57_1908 | hypothetical protein | -4.64 | 0.0030 |
| AB57_RS01000 | AB57_0196 | MFS transporter | -4.63 | 0.0032 |
| AB57_RS01020 | AB57_0200 | pseudogene | -4.63 | 0.0003 |
| AB57_RS05505 | AB57_1089 | hypothetical protein | -4.63 | 0.0017 |
| AB57_RS03290 | AB57_0640 | hypothetical protein | -4.62 | 0.0015 |
| AB57_RS18040 | AB57_3644 | membrane protein | -4.62 | 0.0000 |
| AB57_RS13415 | AB57_2704 | hypothetical protein | -4.62 | 0.0030 |
| AB57_RS14955 | AB57_3024 | hypothetical protein | -4.62 | 0.0004 |
| AB57_RS19105 | AB57_3860 | urocanate hydratase | -4.62 | 0.0015 |
| AB57_RS14715 | AB57_2974 | esterase | -4.61 | 0.0006 |
| AB57_RS16455 | AB57_3329 | TonB-dependent receptor | -4.61 | 0.0013 |
| AB57_RS15070 | AB57_3050 | elongation factor P--(R)-beta-lysine ligase | -4.61 | 0.0016 |
| AB57_RS18850 | AB57_3805 | ion channel protein Tsx | -4.60 | 0.0003 |
| AB57_RS13760 | AB57_2772 | histidine kinase | -4.60 | 0.0046 |
| AB57_RS18065 | AB57_3649 | penicillin-binding protein 1A | -4.60 | 0.0002 |
| AB57_RS17325 | AB57_3497 | membrane protein | -4.59 | 0.0009 |
| AB57_RS04105 | AB57_0801 | MULTISPECIES: NADH-quinone oxidoreductase subunit B | -4.59 | 0.0055 |
| AB57_RS09545 | AB57_1916 | hypothetical protein | -4.59 | 0.0073 |
| AB57_RS03400 | AB57_0664 | MULTISPECIES: copper resistance protein CopB | -4.59 | 0.0005 |
| AB57_RS12785 | AB57_2579 | polyketide cyclase | -4.58 | 0.0002 |
| AB57_RS19290 | AB57_3898 | hypothetical protein | -4.58 | 0.0003 |
| AB57_RS02115 | AB57_0402 | DNA repair protein RecN | -4.58 | 0.0091 |
| AB57_RS03655 | AB57_0717 | membrane protein | -4.57 | 0.0030 |
| AB57_RS15400 | AB57_3117 | membrane protein | -4.57 | 0.0022 |
| AB57_RS11610 | AB57_2334 | fumarate hydratase | -4.57 | 0.0008 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS03950 | AB57_0770 | tricarballoylate dehydrogenase | -4.57 | 0.0069 |
| AB57_RS18865 | AB57_3808 | ureidoglycolate hydrolase | -4.57 | 0.0009 |
| AB57_RS08835 | AB57_1772 | proline:sodium symporter | -4.56 | 0.0042 |
| AB57_RS15715 | AB57_3183 | 3-methylitaconate isomerase | -4.56 | 0.0002 |
| AB57_RS14950 | AB57_3023 | hypothetical protein | -4.56 | 0.0001 |
| AB57_RS19150 | AB57_3869 | glyoxalase | -4.55 | 0.0001 |
| AB57_RS00635 | AB57_0126 | hypothetical protein | -4.55 | 0.0009 |
| AB57_RS19140 | AB57_3867 | fumarylacetoacetase | -4.55 | 0.0028 |
| AB57_RS15375 | AB57_RS15375 | pseudogene | -4.55 | 0.0008 |
| AB57_RS03500 | AB57_0685 | nucleoside triphosphate pyrophosphohydrolase | -4.55 | 0.0037 |
| AB57_RS01985 | AB57_0376 | membrane protein | -4.54 | 0.0026 |
| AB57_RS06630 | AB57_1314 | ATP phosphoribosyltransferase regulatory subunit | -4.54 | 0.0007 |
| AB57_RS15485 | AB57_3136 | MULTISPECIES: hypothetical protein | -4.54 | 0.0041 |
| AB57_RS01515 | AB57_0290 | class I integron integrase | -4.52 | 0.0001 |
| AB57_RS02540 | AB57_0486 | RpiR family transcriptional regulator | -4.52 | 0.0043 |
| AB57_RS05275 | AB57_1039 | AraC family transcriptional regulator | -4.52 | 0.0037 |
| AB57_RS14765 | AB57_2984 | ATPase | -4.52 | 0.0046 |
| AB57_RS09505 | AB57_1907 | O-succinylhomoserine sulfhydrylase | -4.51 | 0.0003 |
| AB57_RS14325 | AB57_RS14325 | pseudogene | -4.51 | 0.0030 |
| AB57_RS06425 | AB57_1271 | hypothetical protein | -4.51 | 0.0030 |
| AB57_RS03585 | | hypothetical protein | -4.51 | 0.0005 |
| AB57_RS12465 | AB57_2514 | peptidase | -4.51 | 0.0012 |
| AB57_RS15720 | AB57_3185 | MULTISPECIES: amino acid permease | -4.50 | 0.0007 |
| AB57_RS01965 | AB57_0372 | hypothetical protein | -4.50 | 0.0027 |
| AB57_RS19295 | AB57_3899 | 4-hydroxy-tetrahydrodipicolinate reductase | -4.49 | 0.0073 |
| AB57_RS18915 | AB57_3818 | dihydrolipoamide acetyltransferase | -4.49 | 0.0007 |
| AB57_RS05960 | AB57_1178 | coniferyl aldehyde dehydrogenase | -4.49 | 0.0016 |
| AB57_RS00710 | AB57_0140 | MULTISPECIES: hypothetical protein | -4.48 | 0.0030 |
| AB57_RS14410 | AB57_2910 | hypothetical protein | -4.48 | 0.0005 |
| AB57_RS04660 | AB57_0910 | hypothetical protein | -4.46 | 0.0015 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS01740 | AB57_0332 | iron transporter FeoB | -4.46 | 0.0024 |
| AB57_RS05805 | AB57_1151 | diguanylate cyclase | -4.46 | 0.0001 |
| AB57_RS16790 | AB57_3401 | galactosyltransferase Lgt5 | -4.44 | 0.0038 |
| AB57_RS17245 | AB57_3485 | membrane protein | -4.44 | 0.0006 |
| AB57_RS11840 | AB57_2383 | MULTISPECIES: acetyl-CoA acetyltransferase | -4.43 | 0.0006 |
| AB57_RS07270 | AB57_1447 | type IV secretion protein Rhs | -4.43 | 0.0030 |
| AB57_RS00695 | AB57_0137 | MULTISPECIES: AsnC family transcriptional regulator | -4.43 | 0.0050 |
| AB57_RS10140 | AB57_2033 | hypothetical protein | -4.43 | 0.0004 |
| AB57_RS13530 | AB57_2728 | alkaline phosphatase | -4.43 | 0.0030 |
| AB57_RS02520 | AB57_0482 | phosphoglycolate phosphatase | -4.41 | 0.0048 |
| AB57_RS11225 | AB57_2255 | phospholipase | -4.41 | 0.0002 |
| AB57_RS05720 | AB57_1136 | 1,4-beta-N-acetylmuramidase | -4.41 | 0.0014 |
| AB57_RS04895 | AB57_0958 | gamma-glutamyltransferase | -4.40 | 0.0039 |
| AB57_RS15290 | AB57_4795 | tRNA-Leu | -4.39 | 0.0029 |
| GenBank:repeat_r egion:NC_011586: 3201913:3202051 | | | -4.39 | 0.0029 |
| AB57_RS01355 | AB57_0267 | MULTISPECIES: acetyltransferase | -4.39 | 0.0030 |
| AB57_RS18855 | AB57_3806 | monooxygenase | -4.39 | 0.0025 |
| AB57_RS00430 | AB57_0084 | phospholipase C | -4.39 | 0.0042 |
| GenBank:repeat_r egion:NC_011586: 593000:599202 | | | -4.39 | 0.0008 |
| GenBank:STS:NC _011586:297629:2 D13S22 97780 | | | -4.38 | 0.0030 |
| AB57_RS06380 | AB57_1262 | hypothetical protein | -4.38 | 0.0030 |
| AB57_RS14285 | AB57_2878 | MULTISPECIES: AsnC family transcriptional regulator | -4.37 | 0.0030 |
| AB57_RS17195 | AB57_3475 | hypothetical protein | -4.37 | 0.0035 |
| AB57_RS17260 | AB57_3488 | fumarylacetoacetate hydrolase | -4.36 | 0.0039 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS03335 | AB57_0651 | MULTISPECIES: molecular chaperone DnaJ | -4.36 | 0.0024 |
| AB57_RS10375 | AB57_2077 | transcriptional regulator | -4.35 | 0.0010 |
| AB57_RS03870 | AB57_0758 | cation transporter | -4.35 | 0.0001 |
| AB57_RS00450 | AB57_0088 | membrane protein | -4.34 | 0.0030 |
| AB57_RS14310 | AB57_2883 | TetR family transcriptional regulator | -4.34 | 0.0015 |
| AB57_RS06815 | AB57_1351 | MULTISPECIES: LysR family transcriptional regulator | -4.33 | 0.0029 |
| AB57_RS02150 | AB57_0409 | type II secretion system protein F | -4.33 | 0.0024 |
| AB57_RS15580 | AB57_3155 | phosphatidylglycerophosphatase | -4.32 | 0.0031 |
| AB57_RS04110 | AB57_0802 | NADH-quinone oxidoreductase subunit C/D | -4.32 | 0.0055 |
| AB57_RS07155 | AB57_1419 | universal stress protein | -4.32 | 0.0077 |
| AB57_RS08435 | AB57_1689 | chromate transporter | -4.32 | 0.0003 |
| AB57_RS04985 | AB57_0977 | TatD family hydrolase | -4.32 | 0.0011 |
| AB57_RS07490 | AB57_1492 | type VI secretion protein | -4.32 | 0.0045 |
| AB57_RS00445 | AB57_0087 | MULTISPECIES: N-acetyl-anhydromuranmyl-L-alanine amidase | -4.31 | 0.0033 |
| GenBank:repeat_r egion:NC_011586: 324399:326560 | | | -4.30 | 0.0006 |
| AB57_RS16535 | AB57_3347 | 3-demethylubiquinone-9 3-methyltransferase | -4.30 | 0.0036 |
| AB57_RS15700 | AB57_3180 | membrane protein | -4.29 | 0.0062 |
| AB57_RS18785 | AB57_3792 | catabolite repression control protein | -4.29 | 0.0038 |
| AB57_RS05915 | AB57_1170 | polyketide cyclase | -4.29 | 0.0030 |
| AB57_RS03230 | AB57_0626 | MULTISPECIES: ketol-acid reductoisomerase | -4.29 | 0.0005 |
| AB57_RS03130 | AB57_0606 | hypothetical protein | -4.28 | 0.0014 |
| AB57_RS15825 | AB57_3208 | MULTISPECIES: DNA-binding protein | -4.28 | 0.0035 |
| AB57_RS19320 | AB57_3904 | RND transporter | -4.28 | 0.0012 |
| AB57_RS04965 | AB57_RS04965 | pseudogene | -4.27 | 0.0008 |
| AB57_RS14960 | AB57_3025 | transporter | -4.27 | 0.0005 |
| AB57_RS13280 | AB57_2679 | hypothetical protein | -4.26 | 0.0025 |
| AB57_RS13305 | AB57_2683 | hypothetical protein | -4.25 | 0.0018 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS00140 | AB57_0029 | thiol:disulfide interchange protein | -4.25 | 0.0000 |
| AB57_RS03865 | AB57_0757 | copper resistance protein B | -4.25 | 0.0014 |
| AB57_RS06310 | AB57_1249 | MULTISPECIES: hypothetical protein | -4.25 | 0.0030 |
| AB57_RS15205 | AB57_3077 | hydrolase | -4.24 | 0.0002 |
| AB57_RS12055 | AB57_2427 | hydroxyethylthiazole kinase | -4.23 | 0.0002 |
| AB57_RS05235 | AB57_1030 | MULTISPECIES: short-chain dehydrogenase | -4.23 | 0.0077 |
| AB57_RS15795 | AB57_3202 | DNA-binding protein | -4.22 | 0.0030 |
| AB57_RS14985 | AB57_3032 | multidrug ABC transporter permease | -4.22 | 0.0025 |
| AB57_RS10655 | AB57_RS10655 | pseudogene | -4.22 | 0.0003 |
| AB57_RS02000 | AB57_0379 | selenocysteine synthase | -4.22 | 0.0002 |
| AB57_RS15095 | AB57_3056 | pseudogene | -4.22 | 0.0017 |
| AB57_RS09520 | AB57_1910 | MULTISPECIES: recombinase RecR | -4.21 | 0.0034 |
| AB57_RS05420 | AB57_1071 | LysR family transcriptional regulator | -4.21 | 0.0046 |
| AB57_RS16365 | AB57_3311 | two-component system sensor histidine kinase/response regulator | -4.20 | 0.0032 |
| AB57_RS05590 | AB57_1108 | hypothetical protein | -4.19 | 0.0006 |
| AB57_RS11240 | AB57_2259 | sugar kinase | -4.19 | 0.0040 |
| AB57_RS03455 | AB57_0675 | hydroxypyruvate isomerase | -4.19 | 0.0004 |
| AB57_RS02325 | AB57_0442 | DNA-binding protein | -4.18 | 0.0005 |
| AB57_RS02245 | AB57_0425 | phosphoribosyl-ATP diphosphatase | -4.17 | 0.0038 |
| AB57_RS03690 | AB57_0724 | carbon-nitrogen hydrolase | -4.17 | 0.0006 |
| AB57_RS16515 | AB57_3343 | hypothetical protein | -4.17 | 0.0009 |
| AB57_RS16375 | AB57_3313 | twitching motility protein PilT | -4.16 | 0.0025 |
| AB57_RS19000 | AB57_3835 | hypothetical protein | -4.16 | 0.0049 |
| AB57_RS16565 | AB57_3354 | carnitine dehydratase | -4.16 | 0.0004 |
| AB57_RS05325 | AB57_1050 | nuclease PIN | -4.15 | 0.0037 |
| AB57_RS05760 | | hypothetical protein | -4.14 | 0.0077 |
| AB57_RS11195 | AB57_2250 | hypothetical protein | -4.14 | 0.0030 |
| AB57_RS12040 | AB57_2424 | aminopeptidase N | -4.14 | 0.0001 |
| AB57_RS09590 | AB57_1926 | MULTISPECIES: transcriptional regulator, partial | -4.14 | 0.0030 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS01130 | AB57_0220 | radical SAM protein | -4.13 | 0.0023 |
| AB57_RS16385 | AB57_3315 | MULTISPECIES: transcriptional regulator | -4.12 | 0.0049 |
| AB57_RS08730 | AB57_1750 | biotin synthase | -4.11 | 0.0010 |
| AB57_RS03565 | AB57_0698 | hypothetical protein | -4.10 | 0.0053 |
| AB57_RS05070 | AB57_RS05070 | pseudogene | -4.09 | 0.0008 |
| AB57_RS19305 | AB57_3901 | MULTISPECIES: molecular chaperone DnaJ | -4.09 | 0.0011 |
| AB57_RS00525 | AB57_0104 | UDP-glucose 4-epimerase | -4.08 | 0.0001 |
| AB57_RS12295 | AB57_2478 | molybdopterin biosynthesis protein | -4.08 | 0.0030 |
| AB57_RS01275 | AB57_0251 | MULTISPECIES: FMN reductase | -4.07 | 0.0008 |
| AB57_RS05525 | AB57_1094 | urease subunit alpha | -4.07 | 0.0030 |
| AB57_RS08540 | AB57_1710 | hypothetical protein | -4.07 | 0.0017 |
| AB57_RS01170 | AB57_0229 | preprotein translocase SecA | -4.06 | 0.0000 |
| AB57_RS02650 | AB57_0509 | proton glutamate symport protein | -4.06 | 0.0001 |
| AB57_RS16395 | AB57_3317 | hemolysin D | -4.06 | 0.0000 |
| AB57_RS17210 | AB57_3478 | ATP-binding protein | -4.05 | 0.0001 |
| AB57_RS14845 | AB57_3000 | pseudogene | -4.04 | 0.0022 |
| AB57_RS05450 | AB57_1077 | lysine--tRNA ligase | -4.04 | 0.0069 |
| AB57_RS14250 | AB57_2871 | peptidase S49 | -4.04 | 0.0041 |
| AB57_RS14585 | AB57_2948 | hypothetical protein | -4.04 | 0.0037 |
| AB57_RS00850 | AB57_0166 | threonine transporter RhtB | -4.03 | 0.0030 |
| AB57_RS16270 | AB57_3298 | Bcr/CflA family drug resistance efflux transporter | -4.02 | 0.0004 |
| AB57_RS09735 | AB57_1955 | aspartate ammonia-lyase | -4.02 | 0.0010 |
| AB57_RS13100 | AB57_2641 | hypothetical protein | -4.02 | 0.0008 |
| AB57_RS00270 | AB57_0055 | peptidyl-prolyl cis-trans isomerase | -4.02 | 0.0002 |
| AB57_RS01610 | AB57_0306 | hypothetical protein | -4.01 | 0.0014 |
| GenBank:repeat_r egion:NC_011586: 319168:322540 | | | -4.01 | 0.0003 |
| AB57_RS03820 | AB57_0748 | tRNA threonylcarbamoyladenosine biosynthesis protein TsaB | -4.01 | 0.0077 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS17930 | AB57_3622 | type IV fimbrial biogenesis protein | -4.01 | 0.0049 |
| AB57_RS08545 | AB57_1711 | hypothetical protein | -4.00 | 0.0009 |
| AB57_RS16220 | AB57_3287 | glutamine amidotransferase | -3.99 | 0.0020 |
| AB57_RS17640 | AB57_3561 | shikimate dehydrogenase (NADP+) | -3.99 | 0.0008 |
| AB57_RS18700 | AB57_3774 | membrane protein | -3.99 | 0.0010 |
| AB57_RS05240 | AB57_1031 | MULTISPECIES: L-aspartate dehydrogenase | -3.98 | 0.0091 |
| GenBank:repeat_r egion:NC_011586: 2264424:2265754 | | | -3.98 | 0.0069 |
| AB57_RS00135 | AB57_0028 | cation diffusion facilitator transporter | -3.98 | 0.0003 |
| AB57_RS03890 | AB57_0762 | MULTISPECIES: quinolinate synthetase | -3.98 | 0.0002 |
| AB57_RS03225 | AB57_0625 | MULTISPECIES: acetolactate synthase small subunit | -3.98 | 0.0029 |
| AB57_RS00090 | AB57_0018 | DNA recombination protein RecF | -3.98 | 0.0000 |
| AB57_RS10875 | AB57_2182 | 3-oxoadipate CoA-transferase subunit A | -3.98 | 0.0069 |
| AB57_RS16150 | AB57_3273 | NAD(P)H dehydrogenase | -3.97 | 0.0004 |
| AB57_RS02230 | AB57_0422 | cation:proton antiporter | -3.96 | 0.0008 |
| AB57_RS01880 | AB57_0360 | phosphoglycolate phosphatase | -3.95 | 0.0017 |
| AB57_RS16830 | AB57_3409 | MULTISPECIES: branched-chain amino acid aminotransferase | -3.95 | 0.0033 |
| AB57_RS04285 | AB57_RS04285 | pseudogene | -3.94 | 0.0020 |
| AB57_RS10700 | AB57_2147 | N-acyl-L-amino acid amidohydrolase | -3.94 | 0.0008 |
| AB57_RS12530 | AB57_2528 | methionine ABC transporter ATP-binding protein | -3.94 | 0.0084 |
| AB57_RS07005 | AB57_1388 | helicase | -3.94 | 0.0005 |
| AB57_RS09030 | AB57_1812 | uracil-DNA glycosylase | -3.93 | 0.0006 |
| AB57_RS07255 | AB57_1443 | type IV secretion protein Rhs | -3.93 | 0.0037 |
| AB57_RS00200 | AB57_0041 | membrane protein | -3.93 | 0.0004 |
| AB57_RS16935 | AB57_3430 | hypothetical protein | -3.90 | 0.0022 |
| AB57_RS02175 | AB57_4842 | tRNA-Met | -3.89 | 0.0017 |
| AB57_RS18055 | AB57_3647 | membrane protein | -3.89 | 0.0010 |
| AB57_RS01885 | AB57_0361 | anthranilate synthase | -3.89 | 0.0010 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS15360 | AB57_3106 | methyltransferase | -3.88 | 0.0055 |
| AB57_RS16410 | AB57_3320 | hypothetical protein | -3.88 | 0.0055 |
| AB57_RS05810 | AB57_1152 | argininosuccinate synthase | -3.88 | 0.0038 |
| AB57_RS18890 | AB57_3813 | hypothetical protein | -3.87 | 0.0013 |
| AB57_RS05270 | AB57_1038 | glutaminase | -3.87 | 0.0018 |
| AB57_RS18015 | AB57_3639 | glutamate synthase subunit beta | -3.87 | 0.0006 |
| AB57_RS04800 | AB57_0939 | MULTISPECIES: hemerythrin | -3.86 | 0.0008 |
| AB57_RS13970 | AB57_2811 | peptide synthetase | -3.86 | 0.0030 |
| AB57_RS11245 | AB57_2260 | Rieske | -3.86 | 0.0015 |
| AB57_RS14515 | AB57_2934 | hypothetical protein | -3.86 | 0.0077 |
| AB57_RS15145 | AB57_3066 | peptide synthetase | -3.86 | 0.0030 |
| AB57_RS19015 | AB57_3838 | 3,4-dihydroxy-2-butanone-4-phosphate synthase | -3.85 | 0.0011 |
| AB57_RS15785 | AB57_3197 | tail protein | -3.84 | 0.0014 |
| AB57_RS02740 | AB57_0527 | hypothetical protein | -3.84 | 0.0043 |
| AB57_RS19050 | AB57_RS19050 | pseudogene | -3.83 | 0.0034 |
| AB57_RS04715 | AB57_0921 | leucyl/phenylalanyl-tRNA--protein transferase | -3.83 | 0.0024 |
| AB57_RS00565 | AB57_0112 | UDP-glucose 6-dehydrogenase | -3.83 | 0.0055 |
| AB57_RS15565 | | hypothetical protein | -3.82 | 0.0030 |
| AB57_RS05670 | AB57_1126 | amino-acid transporter Mb2008 | -3.82 | 0.0003 |
| AB57_RS18675 | AB57_3769 | phosphoglucosamine mutase | -3.82 | 0.0047 |
| AB57_RS17255 | AB57_3487 | AraC family transcriptional regulator | -3.82 | 0.0008 |
| AB57_RS16160 | AB57_3275 | phosphoserine phosphatase | -3.80 | 0.0007 |
| AB57_RS13110 | AB57_2643 | two-component system sensor histidine kinase | -3.80 | 0.0055 |
| AB57_RS02710 | AB57_0521 | DNA topoisomerase I | -3.80 | 0.0022 |
| AB57_RS14025 | AB57_RS14025 | pseudogene | -3.79 | 0.0055 |
| AB57_RS14665 | AB57_2963 | S26 family signal peptidase | -3.79 | 0.0030 |
| AB57_RS06320 | AB57_1251 | crossover junction endodeoxyribonuclease RusA | -3.79 | 0.0030 |
| AB57_RS02685 | AB57_0516 | pseudogene | -3.79 | 0.0016 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS08905 | AB57_1786 | hypothetical protein | -3.78 | 0.0002 |
| AB57_RS18455 | AB57_3723 | hypothetical protein | -3.78 | 0.0001 |
| AB57_RS02725 | AB57_0524 | ATP-dependent DNA helicase PcrA | -3.77 | 0.0026 |
| AB57_RS18235 | AB57_3679 | acetyl-CoA hydrolase | -3.76 | 0.0006 |
| AB57_RS00705 | AB57_0139 | alanine racemase | -3.75 | 0.0031 |
| AB57_RS02320 | AB57_0441 | MULTISPECIES: MerR family transcriptional regulator | -3.75 | 0.0016 |
| AB57_RS06660 | AB57_1319 | MULTISPECIES: osmotically inducible protein C | -3.75 | 0.0047 |
| AB57_RS06335 | AB57_1254 | MULTISPECIES: hypothetical protein | -3.75 | 0.0030 |
| AB57_RS05050 | AB57_0991 | heme oxygenase | -3.74 | 0.0030 |
| AB57_RS02530 | AB57_0484 | glutathione S-transferase, partial | -3.74 | 0.0055 |
| AB57_RS13975 | AB57_2812 | Rhizobactin siderophore biosynthesis protein rhbE | -3.73 | 0.0030 |
| AB57_RS00760 | AB57_0150 | MFS transporter | -3.73 | 0.0017 |
| AB57_RS01155 | AB57_0226 | MFS transporter | -3.72 | 0.0040 |
| AB57_RS17515 | AB57_3535 | flavohepotein | -3.71 | 0.0026 |
| AB57_RS01405 | | MULTISPECIES: transposase | -3.71 | 0.0003 |
| AB57_RS11180 | AB57_2245 | MULTISPECIES: ATP-dependent Clp protease adaptor ClpS | -3.70 | 0.0081 |
| AB57_RS08460 | AB57_1694 | MULTISPECIES: serine O-acetyltransferase | -3.68 | 0.0013 |
| AB57_RS17875 | AB57_3611 | membrane protein | -3.68 | 0.0029 |
| AB57_RS17250 | AB57_3486 | membrane protein | -3.68 | 0.0042 |
| AB57_RS13695 | AB57_2759 | MFS transporter | -3.66 | 0.0030 |
| AB57_RS14280 | AB57_2877 | pyruvate decarboxylase | -3.66 | 0.0070 |
| AB57_RS01605 | AB57_0305 | MULTISPECIES: sulfate transporter | -3.65 | 0.0058 |
| AB57_RS17940 | AB57_3624 | hypothetical protein | -3.65 | 0.0037 |
| AB57_RS06675 | AB57_1322 | chaperone protein ClpB | -3.65 | 0.0010 |
| AB57_RS06300 | AB57_1247 | hypothetical protein | -3.65 | 0.0029 |
| AB57_RS13865 | AB57_2790 | indole-3-glycerol phosphate synthase | -3.63 | 0.0026 |
| AB57_RS15710 | AB57_3182 | MULTISPECIES: geranyltranstransferase | -3.61 | 0.0001 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS14580 | AB57_2947 | excinuclease ABC subunit B | -3.61 | 0.0004 |
| AB57_RS14795 | | hypothetical protein | -3.61 | 0.0030 |
| AB57_RS19315 | AB57_3903 | ACR family transporter | -3.61 | 0.0002 |
| AB57_RS00700 | AB57_0138 | MULTISPECIES: D-amino acid dehydrogenase | -3.61 | 0.0004 |
| AB57_RS15475 | AB57_3134 | hypothetical protein | -3.60 | 0.0065 |
| AB57_RS18430 | AB57_3717 | LysR family transcriptional regulator | -3.60 | 0.0021 |
| AB57_RS17760 | AB57_3586 | arginine N-succinyltransferase | -3.59 | 0.0013 |
| AB57_RS16085 | AB57_3260 | hypothetical protein | -3.58 | 0.0030 |
| AB57_RS16710 | AB57_3385 | fatty acid desaturase | -3.58 | 0.0007 |
| AB57_RS00605 | AB57_0120 | MULTISPECIES: aromatic amino acid aminotransferase | -3.58 | 0.0013 |
| AB57_RS08330 | AB57_1667 | cytochrome D ubiquinol oxidase subunit I | -3.57 | 0.0030 |
| AB57_RS03110 | AB57_0602 | pilus assembly protein PilW | -3.55 | 0.0000 |
| AB57_RS02925 | AB57_0564 | MULTISPECIES: transposase | -3.55 | 0.0013 |
| AB57_RS02765 | AB57_0532 | coniferyl aldehyde dehydrogenase | -3.55 | 0.0022 |
| AB57_RS12960 | AB57_2610 | Type II secretion system | -3.55 | 0.0002 |
| AB57_RS13285 | AB57_2680 | hypothetical protein | -3.55 | 0.0049 |
| AB57_RS13125 | AB57_2646 | type II secretion system protein E | -3.54 | 0.0069 |
| AB57_RS08740 | AB57_1752 | hypothetical protein | -3.54 | 0.0069 |
| AB57_RS05265 | AB57_1037 | peptide deformylase | -3.54 | 0.0036 |
| AB57_RS11520 | AB57_2315 | peptidase | -3.53 | 0.0008 |
| AB57_RS14790 | AB57_2988 | NIF3 1 | -3.53 | 0.0030 |
| AB57_RS14730 | AB57_2977 | sulfate ABC transporter ATP-binding protein | -3.53 | 0.0001 |
| AB57_RS02420 | AB57_0461 | MULTISPECIES: DNA gyrase inhibitor | -3.53 | 0.0013 |
| AB57_RS01035 | AB57_0203 | peptidoglycan-binding protein LysM | -3.51 | 0.0021 |
| AB57_RS13215 | AB57_2667 | hypothetical protein | -3.50 | 0.0015 |
| AB57_RS06305 | AB57_1248 | DNA replication protein | -3.49 | 0.0015 |
| AB57_RS18045 | AB57_3645 | hypothetical protein | -3.47 | 0.0049 |
| AB57_RS11525 | AB57_2316 | 1-deoxy-D-xylulose 5-phosphate reductoisomerase | -3.47 | 0.0019 |
| AB57_RS13920 | AB57_2801 | peptidase | -3.47 | 0.0027 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS14045 | AB57_2829 | hypothetical protein | -3.46 | 0.0014 |
| AB57_RS06330 | AB57_1253 | hypothetical protein | -3.46 | 0.0069 |
| AB57_RS09350 | AB57_1876 | ligand-gated channel protein | -3.43 | 0.0014 |
| AB57_RS02860 | AB57_0551 | MULTISPECIES: carbapenem-hydrolyzing class D beta-lactamase OXA-23 | -3.42 | 0.0016 |
| AB57_RS14040 | AB57_2828 | hypothetical protein | -3.41 | 0.0006 |
| AB57_RS05400 | AB57_1067 | peptidase S54 | -3.40 | 0.0016 |
| AB57_RS05615 | AB57_1113 | hypothetical protein | -3.39 | 0.0057 |
| AB57_RS17530 | AB57_3538 | metal-dependent hydrolase | -3.39 | 0.0022 |
| AB57_RS07435 | AB57_1481 | MULTISPECIES: hypothetical protein | -3.38 | 0.0030 |
| AB57_RS18245 | AB57_4839 | tRNA-Phe | -3.36 | 0.0030 |
| AB57_RS01305 | | MULTISPECIES: MerR family transcriptional regulator | -3.33 | 0.0055 |
| AB57_RS05855 | AB57_1160 | hypothetical protein | -3.33 | 0.0022 |
| AB57_RS01725 | AB57_0329 | thiol:disulfide interchange protein | -3.33 | 0.0014 |
| AB57_RS05125 | AB57_1008 | MULTISPECIES: acyl-CoA thioesterase | -3.33 | 0.0000 |
| AB57_RS17235 | AB57_3483 | xanthine phosphoribosyltransferase | -3.32 | 0.0005 |
| AB57_RS15430 | AB57_3124 | dihydrolipoamide dehydrogenase | -3.31 | 0.0077 |
| AB57_RS19110 | AB57_3861 | hypothetical protein | -3.31 | 0.0006 |
| AB57_RS08005 | AB57_1598 | 3-methylcrotonyl-CoA carboxylase subunit alpha | -3.31 | 0.0006 |
| AB57_RS04755 | AB57_0930 | preprotein translocase subunit SecA | -3.31 | 0.0038 |
| AB57_RS08850 | AB57_1775 | AraC family transcriptional regulator | -3.29 | 0.0050 |
| AB57_RS18275 | AB57_3685 | pseudogene | -3.28 | 0.0055 |
| AB57_RS04095 | AB57_0799 | diguanylate cyclase | -3.28 | 0.0002 |
| AB57_RS07770 | AB57_1550 | cytochrome B561 | -3.28 | 0.0033 |
| AB57_RS17980 | AB57_3632 | MULTISPECIES: bacterioferritin | -3.26 | 0.0033 |
| AB57_RS18600 | AB57_3752 | FMN reductase | -3.26 | 0.0049 |
| AB57_RS02155 | AB57_0410 | type IV-A pilus assembly ATPase PilB | -3.25 | 0.0001 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS17920 | AB57_3620 | pilin | -3.23 | 0.0005 |
| AB57_RS04270 | AB57_0834 | multidrug transporter MatE | -3.23 | 0.0015 |
| AB57_RS02985 | AB57_0577 | MULTISPECIES: hydroxylase | -3.20 | 0.0000 |
| AB57_RS17765 | AB57_3587 | acetylornithine aminotransferase | -3.20 | 0.0011 |
| AB57_RS12980 | AB57_2615 | Arsenite_oxidase; nitroreductase | -3.20 | 0.0017 |
| AB57_RS15050 | AB57_3045 | MULTISPECIES: hypothetical protein | -3.19 | 0.0003 |
| AB57_RS03460 | AB57_0676 | 2-hydroxy-3-oxopropionate reductase | -3.19 | 0.0001 |
| AB57_RS02625 | AB57_0503 | L-asparaginase 1 | -3.16 | 0.0049 |
| AB57_RS02065 | AB57_0392 | acyl-CoA thioesterase | -3.15 | 0.0003 |
| AB57_RS02735 | AB57_0526 | RNA-binding protein | -3.13 | 0.0015 |
| AB57_RS07460 | AB57_1486 | type VI secretion protein | -3.11 | 0.0030 |
| AB57_RS09970 | AB57_2003 | hypothetical protein | -3.10 | 0.0030 |
| AB57_RS17520 | AB57_3536 | MULTISPECIES: hypothetical protein | -3.10 | 0.0015 |
| AB57_RS18290 | AB57_3689 | transporter | -3.09 | 0.0033 |
| AB57_RS14095 | AB57_2840 | aldehyde dehydrogenase | -3.08 | 0.0030 |
| AB57_RS16995 | AB57_3438 | hypothetical protein | -3.07 | 0.0046 |
| AB57_RS03145 | AB57_0609 | ACP synthase | -3.06 | 0.0008 |
| AB57_RS02935 | AB57_0567 | alkaline phosphatase | -3.05 | 0.0033 |
| AB57_RS17925 | AB57_3621 | pilus assembly protein PilY | -3.03 | 0.0034 |
| AB57_RS11560 | AB57_2324 | nitrogen regulation protein NR(I) | -3.02 | 0.0052 |
| AB57_RS15225 | AB57_3081 | pseudogene | -2.99 | 0.0073 |
| AB57_RS11555 | AB57_2323 | PAS domain-containing sensor histidine kinase | -2.99 | 0.0003 |
| AB57_RS09425 | AB57_1891 | peptidase | -2.99 | 0.0014 |
| AB57_RS14740 | AB57_2979 | membrane protein | -2.92 | 0.0009 |
| AB57_RS02040 | AB57_0387 | fatty acid oxidation complex subunit alpha | -2.91 | 0.0003 |
| AB57_RS08890 | AB57_1783 | TetR family transcriptional regulator | -2.90 | 0.0005 |
| AB57_RS02555 | AB57_0489 | phosphoenolpyruvate-protein phosphotransferase | -2.89 | 0.0011 |
| AB57_RS15910 | AB57_3225 | head morphogenesis protein | -2.88 | 0.0019 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|---|---|----------------|
| AB57_RS01135 | AB57_0221 | hypothetical protein | -2.86 | 0.0037 |
| AB57_RS13225 | AB57_2669 | hypothetical protein | -2.85 | 0.0069 |
| AB57_RS00580 | AB57_0115 | hypothetical protein | -2.82 | 0.0008 |
| AB57_RS17200 | AB57_3476 | lytic transglycosylase | -2.81 | 0.0026 |
| AB57_RS12470 | AB57_2515 | transglutaminase | -2.80 | 0.0014 |
| AB57_RS15370 | AB57_RS15370 | pseudogene | -2.78 | 0.0037 |
| AB57_RS01140 | AB57_0222 | hypothetical protein | -2.78 | 0.0042 |
| AB57_RS14700 | AB57_2970 | hypothetical protein | -2.75 | 0.0003 |
| AB57_RS06040 | AB57_1195 | MULTISPECIES: crotonase | -2.68 | 0.0017 |
| AB57_RS12965 | AB57_RS12965 | pseudogene | -2.66 | 0.0036 |
| AB57_RS09985 | AB57_2006 | pilus assembly protein | -2.65 | 0.0003 |
| AB57_RS02075 | AB57_0394 | methylated-DNA--protein-cysteine methyltransferase | -2.65 | 0.0016 |
| AB57_RS07780 | AB57_1552 | MULTISPECIES: GntR family transcriptional regulator | -2.65 | 0.0010 |
| AB57_RS13685 | AB57_2757 | methionine aminopeptidase | -2.64 | 0.0041 |
| AB57_RS04010 | AB57_0782 | 5-methyltetrahydropteroyltriglutamate--homocysteine methyltransferase | -2.59 | 0.0035 |
| AB57_RS18115 | AB57_3659 | peptide signal protein | -2.57 | 0.0045 |
| AB57_RS18460 | AB57_3724 | quercetin 2,3-dioxygenase | -2.56 | 0.0009 |
| AB57_RS02635 | AB57_RS02635 | pseudogene | -2.56 | 0.0000 |
| AB57_RS01785 | AB57_0341 | hypothetical protein | -2.55 | 0.0006 |
| AB57_RS01255 | AB57_0247 | protein TniQ | -2.54 | 0.0055 |
| GenBank:repeat_r egion:NC_011586: 3659118:3659357 | | | -2.54 | 0.0029 |
| AB57_RS01505 | AB57_0289 | MULTISPECIES: transposase | -2.52 | 0.0010 |
| AB57_RS01780 | AB57_0340 | MULTISPECIES: phosphomethylpyrimidine synthase | -2.52 | 0.0018 |
| AB57_RS16630 | AB57_3368 | hypothetical protein | -2.51 | 0.0061 |
| AB57_RS02035 | AB57_0386 | 3-ketoacyl-CoA thiolase | -2.51 | 0.0028 |
| AB57_RS01030 | AB57_0202 | DNA-binding protein | -2.50 | 0.0020 |
| AB57_RS15365 | | metalloprotease | -2.49 | 0.0039 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|---|------------------|--|---|----------------|
| AB57_RS15980 | AB57_3237 | serine/threonine-protein phosphatase 1 | -2.49 | 0.0029 |
| AB57_RS01815 | AB57_0347 | argininosuccinate lyase | -2.48 | 0.0001 |
| AB57_RS08515 | AB57_1705 | uridylyltransferase | -2.46 | 0.0030 |
| GenBank:repeat_r egion:NC_011586: 310258:311077 | | | -2.45 | 0.0004 |
| AB57_RS00915 | AB57_0179 | NAD-dependent malic enzyme | -2.43 | 0.0000 |
| AB57_RS07775 | AB57_1551 | MULTISPECIES: hypothetical protein | -2.42 | 0.0014 |
| AB57_RS03495 | AB57_0684 | short-chain dehydrogenase | -2.42 | 0.0030 |
| AB57_RS04275 | AB57_0835 | MULTISPECIES: deoxycytidine triphosphate deaminase | -2.41 | 0.0001 |
| AB57_RS08960 | AB57_1797 | biopolymer transporter ExbB | -2.37 | 0.0006 |
| AB57_RS17030 | AB57_3445 | membrane protein | -2.37 | 0.0002 |
| AB57_RS18140 | AB57_3662 | hypothetical protein | -2.34 | 0.0025 |
| AB57_RS18545 | AB57_3741 | TetR family transcriptional regulator | -2.31 | 0.0001 |
| AB57_RS00110 | AB57_0022 | ribonuclease P protein component | -2.31 | 0.0001 |
| AB57_RS04440 | AB57_0871 | MULTISPECIES: hypothetical protein | -2.23 | 0.0077 |
| AB57_RS13860 | AB57_2789 | MULTISPECIES: anthranilate phosphoribosyltransferase | -2.21 | 0.0003 |
| AB57_RS01820 | AB57_0348 | alginate biosynthesis protein | -2.17 | 0.0028 |
| AB57_RS18415 | AB57_3715 | hypothetical protein | -2.16 | 0.0036 |
| AB57_RS12645 | AB57_2551 | DNA-binding response regulator | -2.14 | 0.0030 |
| AB57_RS18830 | AB57_3801 | allantoinase | -2.13 | 0.0009 |
| AB57_RS16495 | AB57_3338 | xanthine permease | -2.11 | 0.0049 |
| AB57_RS08770 | AB57_1759 | hypothetical protein | -2.10 | 0.0052 |
| AB57_RS15835 | AB57_3210 | hypothetical protein | -2.07 | 0.0008 |
| AB57_RS11385 | AB57_2288 | hypothetical protein | -2.07 | 0.0008 |
| AB57_RS07915 | AB57_1580 | MFS transporter | -1.99 | 0.0069 |
| AB57_RS18255 | AB57_3681 | GNAT family acetyltransferase | -1.99 | 0.0009 |
| AB57_RS10450 | AB57_2093 | GCN5 family N-acetyltransferase | -1.98 | 0.0030 |
| AB57_RS16510 | AB57_3342 | RTCB protein | -1.97 | 0.0041 |
| AB57_RS02240 | AB57_0424 | DNA helicase | -1.94 | 0.0003 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|---|---|----------------|
| AB57_RS08765 | AB57_1758 | FxsA cytoplasmic membrane protein | -1.91 | 0.0003 |
| AB57_RS01735 | AB57_0331 | iron transporter | -1.90 | 0.0053 |
| AB57_RS17025 | AB57_3444 | GTPase HflX | -1.87 | 0.0001 |
| AB57_RS18955 | AB57_3826 | TetR family transcriptional regulator | -1.87 | 0.0038 |
| AB57_RS14270 | AB57_2875 | phosphonate ABC transporter substrate-binding protein | -1.85 | 0.0008 |
| AB57_RS16240 | AB57_3291 | microcin B17 transporter | -1.85 | 0.0002 |
| AB57_RS01285 | AB57_0253 | arsenate reductase | -1.81 | 0.0058 |
| AB57_RS15975 | | MULTISPECIES: hypothetical protein | -1.79 | 0.0001 |
| AB57_RS02010 | AB57_0381 | dithiol-disulfide isomerase | -1.78 | 0.0002 |
| AB57_RS08025 | AB57_1602 | TetR family transcriptional regulator | -1.77 | 0.0028 |
| AB57_RS16380 | AB57_3314 | MULTISPECIES: two-component system response regulator | -1.74 | 0.0012 |
| AB57_RS00505 | AB57_0099 | hypothetical protein | -1.71 | 0.0070 |
| AB57_RS09190 | AB57_1844 | MULTISPECIES: ribonuclease | -1.71 | 0.0077 |
| AB57_RS00240 | AB57_0049 | protein GrpE | -1.66 | 0.0077 |
| AB57_RS14605 | AB57_2952 | S-(hydroxymethyl)glutathione synthase | -1.65 | 0.0030 |
| AB57_RS19480 | AB57_RS19480 | r01 | -1.63 | 0.0003 |
| AB57_RS19480 | | RNase_P_RNA | -1.63 | 0.0003 |
| AB57_RS00145 | AB57_0030 | MULTISPECIES: transglycosylase | -1.63 | 0.0027 |
| AB57_RS06420 | AB57_1270 | MULTISPECIES: terminase | -1.60 | 0.0069 |
| AB57_RS07765 | AB57_1549 | catalase | -1.60 | 0.0030 |
| AB57_RS08635 | AB57_1731 | transcriptional regulator | -1.55 | 0.0006 |
| AB57_RS03310 | AB57_0645 | MULTISPECIES: hypothetical protein | -1.55 | 0.0069 |
| AB57_RS18965 | AB57_3828 | histidine kinase | -1.48 | 0.0001 |
| AB57_RS05780 | | hypothetical protein | -1.42 | 0.0014 |
| AB57_RS15515 | AB57_3142 | putrescine/spermidine ABC transporter | -1.38 | 0.0042 |
| AB57_RS15440 | AB57_3126 | 2-oxoglutarate dehydrogenase subunit E1 | -1.37 | 0.0008 |
| AB57_RS18050 | AB57_3646 | hypothetical protein | -1.37 | 0.0008 |
| AB57_RS01290 | AB57_0254 | MULTISPECIES: arsenic resistance protein | -1.36 | 0.0077 |
| AB57_RS09455 | AB57_1897 | hypothetical protein | -1.33 | 0.0030 |
| AB57_RS02090 | AB57_0397 | fusaric acid resistance protein | -1.30 | 0.0004 |

Table S2. continued.

| Locus tag | Locus tag | Function | Log₂ Fitness defect in spleen | P value |
|------------------|------------------|--|---|----------------|
| AB57_RS12535 | AB57_2529 | methionine ABC transporter permease | -1.30 | 0.0024 |
| AB57_RS12310 | AB57_2481 | MULTISPECIES: butyryl-CoA dehydrogenase | -1.28 | 0.0030 |
| AB57_RS02770 | | TetR family transcriptional regulator | -1.27 | 0.0006 |
| AB57_RS02300 | AB57_0437 | EstA family serine hydrolase | -1.27 | 0.0008 |
| AB57_RS13355 | AB57_2692 | hypothetical protein | -1.26 | 0.0016 |
| AB57_RS03605 | AB57_0706 | MULTISPECIES: integration host factor subunit alpha | -1.24 | 0.0062 |
| AB57_RS08760 | AB57_1757 | carbapenem-hydrolyzing class D beta-lactamase OXA-69 | -1.23 | 0.0069 |
| AB57_RS07690 | AB57_1532 | endonuclease | -1.21 | 0.0012 |
| AB57_RS18370 | AB57_3706 | NAD(P)H oxidoreductase | -1.16 | 0.0030 |
| AB57_RS18495 | AB57_3731 | DNA-binding protein | -1.11 | 0.0002 |
| AB57_RS02435 | AB57_0465 | MULTISPECIES: aldehyde-activating protein | -1.10 | 0.0055 |
| AB57_RS14530 | AB57_2937 | hypothetical protein | -1.10 | 0.0001 |
| AB57_RS01625 | AB57_0309 | MULTISPECIES: nitrogen regulatory protein P-II 1 | -1.06 | 0.0003 |
| AB57_RS17290 | AB57_4819 | tRNA-Arg | -1.05 | 0.0030 |
| AB57_RS17295 | AB57_4820 | tRNA-Arg | -1.05 | 0.0030 |
| AB57_RS18765 | AB57_3788 | glutathione synthetase | -1.03 | 0.0068 |
| AB57_RS06790 | AB57_1346 | benzoate transporter | -1.02 | 0.0091 |
| AB57_RS06825 | AB57_1353 | ATPase | -1.02 | 0.0030 |

Table S3. *A. baumannii* lytic transglycosylases and their closest homolog.

| Gene | Predicted function | Pfam ^a | Fitness defect ^b | Homolog (species) | % identity / positive | Homolog with AB5075 ^c |
|------------------------------|--------------------------|---|-----------------------------|--|-----------------------|----------------------------------|
| AB57_0044 | Lytic transglycosylase | SLT_2; PG_binding_1; PG_binding_3; SLT | -355 | PA1171 (<i>Pseudomonas aeruginosa</i>) | 60 / 75 | ABUW_3883 |
| AB57_1136 | Lytic transglycosylase | SLT; SLT_2 | -21 | <i>mltD</i> (<i>Pseudomonas aeruginosa</i>) | 48 / 67 | ABUW_2840 |
| AB57_2749 (<i>mltB</i>) | Lytic transglycosylase B | SLT_2 | -655 | <i>mltB</i> (<i>Escherichia coli</i>) | 41 / 56 | ABUW_1243 (<i>mltB</i>) |
| AB57_3476 | Lytic transglycosylase | SLT; SLT_L | -7 | <i>mltE</i> (<i>Neisseria gonorrhoeae</i>) | 42 / 58 | ABUW_0465 |

^a **Pfam**

SLT_2: Transglycosylase SLT domain.
 PG_binding_1: Putative peptidoglycan binding domain.
 PG_binding_3: Predicted Peptidoglycan domain.
 SLT: Transglycosylase SLT domain.
 SLT_L: Soluble lytic transglycosylase L domain.

^b Tn-seq calculated fitness defect in the spleen at 24 hpi.

^c All genes share 100% identity with strain AB5075.

Table S4. *A. baumannii* peptidoglycan composition.

| Peak Name | | Relative % of each Muropeptide | | | | | |
|------------------------------------|---|--------------------------------|------------------|-------------------------|---------------------------|------------------|-------------------------|
| | | LB | | | 50% HI serum ^b | | |
| | | 57 | 57 Δ mltB | 57 Δ mltB compl. | 57 | 57 Δ mltB | 57 Δ mltB compl. |
| 1 | Tri | 0.96 ± 0.05 | 2.11 ± 0.09 | 1.60 ± 0.27 | 2.22 ± 0.26 | 3.54 ± 0.03 | 2.36 ± 0.16 |
| 2 | TetraGly4 | 1.41 ± 0.01 | 1.25 ± 0.36 | 1.52 ± 0.27 | 0.64 ± 0.41 | 0.52 ± 0.23 | 0.48 ± 0.16 |
| 3 | Tetra | 22.82 ± 3.18 | 20.12 ± 0.11 | 24.90 ± 3.26 | 21.06 ± 0.30 | 21.94 ± 1.19 | 22.91 ± 0.01 |
| 4 | TetraTri | 4.71 ± 0.04 | 4.78 ± 0.21 | 4.23 ± 0.24 | 4.33 ± 0.54 | 4.04 ± 0.58 | 3.03 ± 0.07 |
| 5 | TetraTetra | 41.51 ± 0.80 | 41.82 ± 1.58 | 44.38 ± 0.04 | 35.16 ± 0.57 | 33.57 ± 0.62 | 37.32 ± 0.38 |
| 6 | TetraTetraTri | 1.01 ± 0.31 | 1.16 ± 0.09 | 0.71 ± 0.40 | 0.81 ± 0.12 | 0.43 ± 0.20 | 0.19 ± 0.39 |
| 7 | TetraTetraTetra | 17.3 ± 1.60 | 17.60 ± 1.10 | 15.44 ± 2.47 | 18.80 ± 0.21 | 16.11 ± 0.55 | 18.53 ± 0.99 |
| 8 | TetraTetraTetraTri (~30%) / TetraTriAnh (~70%) | 3.38 ± 1.28 | 4.17 ± 0.11 | 2.00 ± 1.11 | 5.46 ± 0.35 | 4.28 ± 0.41 | 4.64 ± 0.20 |
| 9 | TetraTetraAnh I | 0.46 ± 0.92 | 0.56 ± 0.03 | 0.00 ± 0.00 | 1.45 ± 0.17 | 1.26 ± 0.03 | 0.98 ± 0.39 |
| 10 | TetraTetraAnh II | 1.57 ± 0.29 | 1.10 ± 0.02 | 1.19 ± 0.02 | 1.57 ± 0.33 | 1.50 ± 0.07 | 1.55 ± 0.46 |
| 11 | TetraTetraTetraAnh | 0.99 ± 0.04 | 0.83 ± 0.00 | 0.73 ± 0.17 | 1.46 ± 0.22 | 1.17 ± 0.06 | 1.35 ± 0.53 |
| Sum of known peaks | | 96.12 ± 1.19 | 94.72 ± 2.94 | 96.71 ± 0.52 | 92.95 ± 0.93 | 88.35 ± 1.15 | 93.35 ± 2.02 |
| Monomers | | 26.23 ± 3.56 | 24.80 ± 0.35 | 28.97 ± 4.10 | 25.73 ± 0.78 | 29.42 ± 0.67 | 27.59 ± 0.26 |
| Dimers | | 52.65 ± 1.5 | 53.45 ± 0.08 | 52.95 ± 0.72 | 49.85 ± 0.10 | 49.08 ± 1.02 | 49.42 ± 0.52 |
| Trimers | | 20.07 ± 1.67 | 20.68 ± 0.43 | 17.46 ± 3.05 | 22.66 ± 0.56 | 20.05 ± 0.52 | 21.51 ± 0.75 |
| Tetramers | | 1.05 ± 0.39 | 1.07 ± 0.00 | 0.62 ± 0.34 | 1.76 ± 0.13 | 1.45 ± 0.16 | 1.49 ± 0.03 |
| % peptides in cross linkage | | 73.77 ± 3.56 | 75.20 ± 0.35 | 71.03 ± 4.10 | 74.27 ± 0.79 | 70.58 ± 0.67 | 72.41 ± 0.26 |
| Average chain length | | 38.6 ± 8.57 | 41.29 ± 0.21 | 63.93 ± 17.69 | 23.88 ± 2.95 | 27.06 ± 2.10 | 27.98 ± 1.70 |

^aPercentages calculated as per (Glauner *et al.*) and are mean ± variation of two independent experiments.

^b HI : Heat-inactivated.

Table S5. Comparative analysis of candidate fitness factors with other Tn-seq analyses.

| With ATCC17978 (pneumonia infection model) ^a | | | | | |
|---|-------------|--------------|-------------|--------------|-------------|
| AB0057 | ATCC17978 | AB0057 | ATCC17978 | AB0057 | ATCC17978 |
| Locus_tag | Locus_tag | Locus_tag | Locus_tag | Locus_tag | Locus_tag |
| AB57_RS12945 | AUO97_00155 | AB57_RS18310 | AUO97_05240 | AB57_RS03160 | AUO97_10075 |
| AB57_RS12950 | AUO97_00160 | AB57_RS18780 | AUO97_05760 | AB57_RS03205 | AUO97_10120 |
| AB57_RS12970 | AUO97_00180 | AB57_RS18815 | AUO97_05795 | AB57_RS03225 | AUO97_10145 |
| AB57_RS13635 | AUO97_00500 | AB57_RS00945 | AUO97_06465 | AB57_RS03230 | AUO97_10150 |
| AB57_RS13645 | AUO97_00510 | AB57_RS00930 | AUO97_06480 | AB57_RS03510 | AUO97_10340 |
| AB57_RS13685 | AUO97_00550 | AB57_RS00580 | AUO97_06870 | AB57_RS03605 | AUO97_10455 |
| AB57_RS13845 | AUO97_00695 | AB57_RS00545 | AUO97_06875 | AB57_RS03640 | AUO97_10490 |
| AB57_RS13860 | AUO97_00710 | AB57_RS00575 | AUO97_06875 | AB57_RS03705 | AUO97_10555 |
| AB57_RS13865 | AUO97_00715 | AB57_RS00570 | AUO97_06880 | AB57_RS03765 | AUO97_10615 |
| AB57_RS14000 | AUO97_00845 | AB57_RS00380 | AUO97_07045 | AB57_RS03850 | AUO97_10700 |
| AB57_RS14545 | AUO97_01375 | AB57_RS00210 | AUO97_07215 | AB57_RS04005 | AUO97_10860 |
| AB57_RS14550 | AUO97_01380 | AB57_RS00205 | AUO97_07220 | AB57_RS04010 | AUO97_10865 |
| AB57_RS14735 | AUO97_01580 | AB57_RS19355 | AUO97_07450 | AB57_RS04080 | AUO97_10935 |
| AB57_RS14805 | AUO97_01835 | AB57_RS19330 | AUO97_07475 | AB57_RS04085 | AUO97_10940 |
| AB57_RS14910 | AUO97_01940 | AB57_RS19195 | AUO97_07610 | AB57_RS04245 | AUO97_11095 |
| AB57_RS15340 | AUO97_02405 | AB57_RS19190 | AUO97_07615 | AB57_RS04255 | AUO97_11105 |
| AB57_RS16130 | AUO97_02820 | AB57_RS19005 | AUO97_07800 | AB57_RS04275 | AUO97_11130 |
| AB57_RS16180 | AUO97_02865 | AB57_RS01710 | AUO97_08630 | AB57_RS05460 | AUO97_12215 |
| AB57_RS16325 | AUO97_03015 | AB57_RS01715 | AUO97_08635 | AB57_RS05465 | AUO97_12220 |
| AB57_RS16680 | AUO97_03355 | AB57_RS01720 | AUO97_08640 | AB57_RS05605 | AUO97_12370 |
| AB57_RS16685 | AUO97_03360 | AB57_RS01805 | AUO97_08725 | AB57_RS05720 | AUO97_12500 |
| AB57_RS16810 | AUO97_03485 | AB57_RS01815 | AUO97_08735 | AB57_RS06700 | AUO97_13355 |
| AB57_RS16830 | AUO97_03505 | AB57_RS01885 | AUO97_08805 | AB57_RS08485 | AUO97_14850 |
| AB57_RS16900 | AUO97_03620 | AB57_RS02050 | AUO97_08975 | AB57_RS08785 | AUO97_15155 |
| AB57_RS17190 | AUO97_04105 | AB57_RS02245 | AUO97_09165 | AB57_RS08930 | AUO97_15295 |
| AB57_RS17560 | AUO97_04475 | AB57_RS02565 | AUO97_09495 | AB57_RS09030 | AUO97_15395 |
| AB57_RS17590 | AUO97_04505 | AB57_RS02580 | AUO97_09510 | AB57_RS09505 | AUO97_16165 |
| AB57_RS17595 | AUO97_04510 | AB57_RS02585 | AUO97_09515 | AB57_RS11570 | AUO97_17785 |
| AB57_RS17605 | AUO97_04520 | AB57_RS02590 | AUO97_09520 | AB57_RS12185 | AUO97_18735 |
| AB57_RS17750 | AUO97_04660 | AB57_RS02595 | AUO97_09525 | AB57_RS12515 | AUO97_19060 |
| AB57_RS17850 | AUO97_04760 | AB57_RS02620 | AUO97_09550 | AB57_RS12520 | AUO97_19065 |
| AB57_RS17855 | AUO97_04765 | AB57_RS02705 | AUO97_09620 | | |
| AB57_RS17960 | AUO97_04870 | AB57_RS02965 | AUO97_09785 | | |
| AB57_RS18015 | AUO97_04925 | AB57_RS02970 | AUO97_09790 | | |
| AB57_RS18020 | AUO97_04930 | AB57_RS02975 | AUO97_09795 | | |
| AB57_RS18200 | AUO97_05110 | AB57_RS03060 | AUO97_09880 | | |
| AB57_RS18260 | AUO97_05185 | AB57_RS03100 | AUO97_09920 | | |
| AB57_RS18265 | AUO97_05190 | AB57_RS03110 | AUO97_09930 | | |
| AB57_RS18280 | AUO97_05205 | AB57_RS03155 | AUO97_10070 | | |

Table S5. continued.

| With ATCC17978 (bloodstream infection model)^b | | | |
|---|------------------|------------------|------------------|
| AB0057 | ATCC17978 | AB0057 | ATCC17978 |
| Locus_tag | Locus_tag | Locus_tag | Locus_tag |
| AB57_RS13635 | AUO97_00500 | AB57_RS05825 | AUO97_12645 |
| AB57_RS14210 | AUO97_01065 | AB57_RS06100 | AUO97_12885 |
| AB57_RS14805 | AUO97_01835 | AB57_RS07165 | AUO97_13700 |
| AB57_RS15395 | AUO97_02465 | AB57_RS11515 | AUO97_17730 |
| AB57_RS15660 | AUO97_02735 | AB57_RS11885 | AUO97_18455 |
| AB57_RS16130 | AUO97_02820 | AB57_RS12140 | AUO97_18690 |
| AB57_RS16365 | AUO97_03045 | AB57_RS12530 | AUO97_19075 |
| AB57_RS16710 | AUO97_03385 | | |
| AB57_RS16775 | AUO97_03450 | | |
| AB57_RS17315 | AUO97_04230 | | |
| AB57_RS17325 | AUO97_04240 | | |
| AB57_RS18030 | AUO97_04940 | | |
| AB57_RS18435 | AUO97_05405 | | |
| AB57_RS18810 | AUO97_05790 | | |
| AB57_RS00865 | AUO97_06545 | | |
| AB57_RS00835 | AUO97_06575 | | |
| AB57_RS00310 | AUO97_07115 | | |
| AB57_RS00250 | AUO97_07175 | | |
| AB57_RS19355 | AUO97_07450 | | |
| AB57_RS18935 | AUO97_07885 | | |
| AB57_RS00640 | AUO97_08400 | | |
| AB57_RS01670 | AUO97_08590 | | |
| AB57_RS01690 | AUO97_08610 | | |
| AB57_RS01865 | AUO97_08785 | | |
| AB57_RS01870 | AUO97_08790 | | |
| AB57_RS02000 | AUO97_08920 | | |
| AB57_RS02590 | AUO97_09520 | | |
| AB57_RS02765 | AUO97_09680 | | |
| AB57_RS02945 | AUO97_09765 | | |
| AB57_RS03025 | AUO97_09845 | | |
| AB57_RS04295 | AUO97_11150 | | |
| AB57_RS04305 | AUO97_11160 | | |
| AB57_RS04465 | AUO97_11320 | | |
| AB57_RS04475 | AUO97_11330 | | |
| AB57_RS04845 | AUO97_11700 | | |
| AB57_RS05125 | AUO97_11875 | | |
| AB57_RS05185 | AUO97_11935 | | |
| AB57_RS05490 | AUO97_12250 | | |
| AB57_RS05495 | AUO97_12255 | | |

Table S5. continued.

| With AB5075 (<i>Galleria mellonella</i> infection model)^c | | | | | |
|---|------------------|------------------|------------------|------------------|------------------|
| AB0057 | AB5075 | AB0057 | AB5075 | AB0057 | AB5075 |
| Locus_tag | Locus_tag | Locus_tag | Locus_tag | Locus_tag | Locus_tag |
| AB57_RS00095 | ABUW_0002 | AB57_RS17225 | ABUW_0460 | AB57_RS15870 | ABUW_0786 |
| AB57_RS00005 | ABUW_0014 | AB57_RS17195 | ABUW_0466 | AB57_RS15835 | ABUW_0792 |
| AB57_RS19385 | ABUW_0020 | AB57_RS17165 | ABUW_0472 | AB57_RS15820 | ABUW_0796 |
| AB57_RS19200 | ABUW_0058 | AB57_RS17100 | ABUW_0486 | AB57_RS15815 | ABUW_0797 |
| AB57_RS19190 | ABUW_0060 | AB57_RS17070 | ABUW_0492 | AB57_RS15810 | ABUW_0798 |
| AB57_RS19185 | ABUW_0061 | AB57_RS17050 | ABUW_0496 | AB57_RS15735 | ABUW_0814 |
| AB57_RS19035 | ABUW_0091 | AB57_RS17045 | ABUW_0497 | AB57_RS15730 | ABUW_0815 |
| AB57_RS18915 | ABUW_0115 | AB57_RS17030 | ABUW_0500 | AB57_RS15670 | ABUW_0827 |
| AB57_RS18890 | ABUW_0120 | AB57_RS16960 | ABUW_0514 | AB57_RS15660 | ABUW_0829 |
| AB57_RS18885 | ABUW_0121 | AB57_RS16915 | ABUW_0523 | AB57_RS15600 | ABUW_0841 |
| AB57_RS18870 | ABUW_0124 | AB57_RS16890 | ABUW_0528 | AB57_RS15595 | ABUW_0842 |
| AB57_RS18815 | ABUW_0135 | AB57_RS16885 | ABUW_0529 | AB57_RS15590 | ABUW_0843 |
| AB57_RS18715 | ABUW_0155 | AB57_RS16840 | ABUW_0585 | AB57_RS15585 | ABUW_0844 |
| AB57_RS18710 | ABUW_0156 | AB57_RS16810 | ABUW_0591 | AB57_RS15540 | ABUW_0852 |
| AB57_RS18675 | ABUW_0163 | AB57_RS16805 | ABUW_0592 | AB57_RS15535 | ABUW_0853 |
| AB57_RS18625 | ABUW_0174 | AB57_RS16800 | ABUW_0593 | AB57_RS15485 | ABUW_0863 |
| AB57_RS18370 | ABUW_0228 | AB57_RS16795 | ABUW_0594 | AB57_RS15470 | ABUW_0866 |
| AB57_RS18365 | ABUW_0229 | AB57_RS16775 | ABUW_0598 | AB57_RS15415 | ABUW_0877 |
| AB57_RS18330 | ABUW_0236 | AB57_RS16670 | ABUW_0619 | AB57_RS15400 | ABUW_0882 |
| AB57_RS18200 | ABUW_0262 | AB57_RS16635 | ABUW_0626 | AB57_RS15365 | ABUW_0888 |
| AB57_RS18190 | ABUW_0264 | AB57_RS16615 | ABUW_0630 | AB57_RS15360 | ABUW_0889 |
| AB57_RS18140 | ABUW_0274 | AB57_RS16595 | ABUW_0634 | AB57_RS15340 | ABUW_0893 |
| AB57_RS18120 | ABUW_0278 | AB57_RS16550 | ABUW_0643 | AB57_RS15305 | ABUW_0900 |
| AB57_RS18065 | ABUW_0289 | AB57_RS16545 | ABUW_0644 | AB57_RS15295 | ABUW_0902 |
| AB57_RS17985 | ABUW_0305 | AB57_RS16540 | ABUW_0645 | AB57_RS15285 | ABUW_0904 |
| AB57_RS17955 | ABUW_0311 | AB57_RS16480 | ABUW_0657 | AB57_RS15265 | ABUW_0908 |
| AB57_RS17870 | ABUW_0328 | AB57_RS16460 | ABUW_0660 | AB57_RS15250 | ABUW_0911 |
| AB57_RS17855 | ABUW_0331 | AB57_RS16390 | ABUW_0677 | AB57_RS15245 | ABUW_0912 |
| AB57_RS17700 | ABUW_0364 | AB57_RS16355 | ABUW_0684 | AB57_RS15235 | ABUW_0914 |
| AB57_RS17640 | ABUW_0376 | AB57_RS16350 | ABUW_0685 | AB57_RS15230 | ABUW_0915 |
| AB57_RS17625 | ABUW_0379 | AB57_RS16325 | ABUW_0690 | AB57_RS15195 | ABUW_0922 |
| AB57_RS17620 | ABUW_0380 | AB57_RS16285 | ABUW_0698 | AB57_RS15190 | ABUW_0923 |
| AB57_RS17595 | ABUW_0385 | AB57_RS16240 | ABUW_0707 | AB57_RS15145 | ABUW_0932 |
| AB57_RS17560 | ABUW_0392 | AB57_RS16190 | ABUW_0717 | AB57_RS15140 | ABUW_0933 |
| AB57_RS17545 | ABUW_0395 | AB57_RS16180 | ABUW_0719 | AB57_RS15135 | ABUW_0934 |
| AB57_RS17525 | ABUW_0399 | AB57_RS16170 | ABUW_0721 | AB57_RS15070 | ABUW_0948 |
| AB57_RS17310 | ABUW_0443 | AB57_RS16135 | ABUW_0728 | AB57_RS14980 | ABUW_0966 |
| AB57_RS17305 | ABUW_0444 | AB57_RS16115 | ABUW_0732 | AB57_RS14975 | ABUW_0967 |
| AB57_RS17285 | ABUW_0448 | AB57_RS15975 | ABUW_0763 | AB57_RS14970 | ABUW_0968 |
| AB57_RS17250 | ABUW_0455 | AB57_RS15940 | ABUW_0771 | AB57_RS14950 | ABUW_0973 |

Table S5. continued.

| With AB5075 (<i>Galleria mellonella</i> infection model)^c | | | | | |
|---|------------------|------------------|------------------|------------------|------------------|
| AB0057 | AB5075 | AB0057 | AB5075 | AB0057 | AB5075 |
| Locus_tag | Locus_tag | Locus_tag | Locus_tag | Locus_tag | Locus_tag |
| AB57_RS14905 | ABUW_0982 | AB57_RS14260 | ABUW_1117 | AB57_RS13320 | ABUW_1311 |
| AB57_RS14875 | ABUW_0988 | AB57_RS14245 | ABUW_1120 | AB57_RS13295 | ABUW_1316 |
| AB57_RS14860 | ABUW_0991 | AB57_RS14230 | ABUW_1123 | AB57_RS15765 | ABUW_1323 |
| AB57_RS14825 | ABUW_0998 | AB57_RS14215 | ABUW_1126 | AB57_RS13225 | ABUW_1330 |
| AB57_RS14820 | ABUW_0999 | AB57_RS14210 | ABUW_1127 | AB57_RS13215 | ABUW_1332 |
| AB57_RS14815 | ABUW_1000 | AB57_RS14205 | ABUW_1128 | AB57_RS13125 | ABUW_1351 |
| AB57_RS14800 | ABUW_1003 | AB57_RS14180 | ABUW_1133 | AB57_RS13115 | ABUW_1353 |
| AB57_RS14790 | ABUW_1005 | AB57_RS14175 | ABUW_1134 | AB57_RS13110 | ABUW_1354 |
| AB57_RS14785 | ABUW_1006 | AB57_RS14165 | ABUW_1136 | AB57_RS13100 | ABUW_1356 |
| AB57_RS14760 | ABUW_1011 | AB57_RS14150 | ABUW_1139 | AB57_RS13070 | ABUW_1362 |
| AB57_RS14735 | ABUW_1016 | AB57_RS14135 | ABUW_1142 | AB57_RS13065 | ABUW_1363 |
| AB57_RS14710 | ABUW_1021 | AB57_RS14130 | ABUW_1143 | AB57_RS13010 | ABUW_1374 |
| AB57_RS14705 | ABUW_1022 | AB57_RS14095 | ABUW_1150 | AB57_RS16015 | ABUW_1409 |
| AB57_RS14680 | ABUW_1027 | AB57_RS14045 | ABUW_1161 | AB57_RS12960 | ABUW_1443 |
| AB57_RS14670 | ABUW_1029 | AB57_RS14040 | ABUW_1162 | AB57_RS12950 | ABUW_1445 |
| AB57_RS14655 | ABUW_1032 | AB57_RS14015 | ABUW_1168 | AB57_RS12865 | ABUW_1462 |
| AB57_RS14640 | ABUW_1035 | AB57_RS14005 | ABUW_1170 | AB57_RS12785 | ABUW_1478 |
| AB57_RS14615 | ABUW_1040 | AB57_RS14000 | ABUW_1173 | AB57_RS12775 | ABUW_1480 |
| AB57_RS14605 | ABUW_1042 | AB57_RS13970 | ABUW_1179 | AB57_RS12645 | ABUW_1506 |
| AB57_RS14585 | ABUW_1046 | AB57_RS13875 | ABUW_1198 | AB57_RS12560 | ABUW_1523 |
| AB57_RS14580 | ABUW_1047 | AB57_RS13860 | ABUW_1201 | AB57_RS12545 | ABUW_1526 |
| AB57_RS14550 | ABUW_1053 | AB57_RS13850 | ABUW_1203 | AB57_RS12540 | ABUW_1527 |
| AB57_RS14540 | ABUW_1055 | AB57_RS13845 | ABUW_1204 | AB57_RS12515 | ABUW_1532 |
| AB57_RS14530 | ABUW_1057 | AB57_RS13805 | ABUW_1212 | AB57_RS12505 | ABUW_1534 |
| AB57_RS14515 | ABUW_1060 | AB57_RS13780 | ABUW_1217 | AB57_RS12500 | ABUW_1535 |
| AB57_RS14510 | ABUW_1061 | AB57_RS13765 | ABUW_1220 | AB57_RS12490 | ABUW_1537 |
| AB57_RS14505 | ABUW_1062 | AB57_RS13715 | ABUW_1228 | AB57_RS12485 | ABUW_1539 |
| AB57_RS14500 | ABUW_1063 | AB57_RS13695 | ABUW_1232 | AB57_RS12470 | ABUW_1542 |
| AB57_RS14455 | ABUW_1072 | AB57_RS13645 | ABUW_1242 | AB57_RS12465 | ABUW_1543 |
| AB57_RS14450 | ABUW_1073 | AB57_RS13640 | ABUW_1243 | AB57_RS12460 | ABUW_1544 |
| AB57_RS14400 | ABUW_1085 | AB57_RS13635 | ABUW_1244 | AB57_RS12355 | ABUW_1564 |
| AB57_RS14385 | ABUW_1088 | AB57_RS13620 | ABUW_1247 | AB57_RS12350 | ABUW_1565 |
| AB57_RS14380 | ABUW_1089 | AB57_RS13510 | ABUW_1270 | AB57_RS12340 | ABUW_1567 |
| AB57_RS14355 | ABUW_1098 | AB57_RS16025 | ABUW_1271 | AB57_RS12310 | ABUW_1573 |
| AB57_RS14335 | ABUW_1102 | AB57_RS13480 | ABUW_1278 | AB57_RS12295 | ABUW_1576 |
| AB57_RS14305 | ABUW_1108 | AB57_RS13435 | ABUW_1288 | AB57_RS12195 | ABUW_1597 |
| AB57_RS14290 | ABUW_1111 | AB57_RS13415 | ABUW_1292 | AB57_RS12190 | ABUW_1598 |
| AB57_RS14285 | ABUW_1112 | AB57_RS13405 | ABUW_1294 | AB57_RS12180 | ABUW_1600 |
| AB57_RS14280 | ABUW_1113 | AB57_RS15910 | ABUW_1294 | AB57_RS12170 | ABUW_1602 |
| AB57_RS14265 | ABUW_1116 | AB57_RS13355 | ABUW_1304 | AB57_RS12165 | ABUW_1603 |

Table S5. continued.

| With AB5075 (<i>Galleria mellonella</i> infection model) ^c | | | | | |
|--|-----------|--------------|-----------|--------------|-----------|
| AB0057 | AB5075 | AB0057 | AB5075 | AB0057 | AB5075 |
| Locus_tag | Locus_tag | Locus_tag | Locus_tag | Locus_tag | Locus_tag |
| AB57_RS12155 | ABUW_1605 | AB57_RS11230 | ABUW_1802 | AB57_RS09190 | ABUW_2214 |
| AB57_RS12145 | ABUW_1607 | AB57_RS11225 | ABUW_1803 | AB57_RS09135 | ABUW_2225 |
| AB57_RS12140 | ABUW_1608 | AB57_RS11220 | ABUW_1804 | AB57_RS09095 | ABUW_2234 |
| AB57_RS12065 | ABUW_1625 | AB57_RS11210 | ABUW_1806 | AB57_RS09090 | ABUW_2235 |
| AB57_RS12055 | ABUW_1627 | AB57_RS11195 | ABUW_1809 | AB57_RS09085 | ABUW_2236 |
| AB57_RS12040 | ABUW_1630 | AB57_RS11190 | ABUW_1811 | AB57_RS09000 | ABUW_2253 |
| AB57_RS11900 | ABUW_1661 | AB57_RS11185 | ABUW_1812 | AB57_RS08995 | ABUW_2254 |
| AB57_RS11885 | ABUW_1664 | AB57_RS11170 | ABUW_1815 | AB57_RS08965 | ABUW_2260 |
| AB57_RS11855 | ABUW_1671 | AB57_RS11025 | ABUW_1844 | AB57_RS08960 | ABUW_2261 |
| AB57_RS11850 | ABUW_1672 | AB57_RS11000 | ABUW_1849 | AB57_RS08955 | ABUW_2262 |
| AB57_RS11845 | ABUW_1673 | AB57_RS10875 | ABUW_1874 | AB57_RS08930 | ABUW_2267 |
| AB57_RS11790 | ABUW_1685 | AB57_RS10700 | ABUW_1910 | AB57_RS08925 | ABUW_2268 |
| AB57_RS11785 | ABUW_1686 | AB57_RS10640 | ABUW_1922 | AB57_RS08895 | ABUW_2274 |
| AB57_RS11775 | ABUW_1688 | AB57_RS10625 | ABUW_1925 | AB57_RS08875 | ABUW_2278 |
| AB57_RS11740 | ABUW_1695 | AB57_RS10570 | ABUW_1936 | AB57_RS08850 | ABUW_2282 |
| AB57_RS11735 | ABUW_1696 | AB57_RS10545 | ABUW_1943 | AB57_RS08835 | ABUW_2285 |
| AB57_RS11690 | ABUW_1706 | AB57_RS10450 | ABUW_1964 | AB57_RS08785 | ABUW_2295 |
| AB57_RS11650 | ABUW_1714 | AB57_RS10395 | ABUW_1975 | AB57_RS08770 | ABUW_2298 |
| AB57_RS11610 | ABUW_1722 | AB57_RS09985 | ABUW_2053 | AB57_RS08765 | ABUW_2299 |
| AB57_RS11575 | ABUW_1729 | AB57_RS09980 | ABUW_2054 | AB57_RS08760 | ABUW_2300 |
| AB57_RS11560 | ABUW_1732 | AB57_RS09970 | ABUW_2056 | AB57_RS08755 | ABUW_2301 |
| AB57_RS11555 | ABUW_1733 | AB57_RS09965 | ABUW_2057 | AB57_RS08750 | ABUW_2302 |
| AB57_RS11550 | ABUW_1734 | AB57_RS09905 | ABUW_2069 | AB57_RS08740 | ABUW_2305 |
| AB57_RS11525 | ABUW_1739 | AB57_RS09895 | ABUW_2072 | AB57_RS08725 | ABUW_2308 |
| AB57_RS11520 | ABUW_1740 | AB57_RS09665 | ABUW_2118 | AB57_RS08710 | ABUW_2311 |
| AB57_RS11515 | ABUW_1741 | AB57_RS09630 | ABUW_2125 | AB57_RS08620 | ABUW_2330 |
| AB57_RS11510 | ABUW_1742 | AB57_RS09625 | ABUW_2126 | AB57_RS08545 | ABUW_2345 |
| AB57_RS11490 | ABUW_1746 | AB57_RS09590 | ABUW_2133 | AB57_RS08535 | ABUW_2347 |
| AB57_RS11480 | ABUW_1748 | AB57_RS09585 | ABUW_2134 | AB57_RS08520 | ABUW_2350 |
| AB57_RS11475 | ABUW_1749 | AB57_RS09555 | ABUW_2140 | AB57_RS08510 | ABUW_2352 |
| AB57_RS11470 | ABUW_1750 | AB57_RS09510 | ABUW_2149 | AB57_RS08505 | ABUW_2353 |
| AB57_RS11395 | ABUW_1767 | AB57_RS09505 | ABUW_2150 | AB57_RS08495 | ABUW_2355 |
| AB57_RS11380 | ABUW_1770 | AB57_RS09455 | ABUW_2161 | AB57_RS08480 | ABUW_2358 |
| AB57_RS11375 | ABUW_1771 | AB57_RS09445 | ABUW_2163 | AB57_RS08475 | ABUW_2359 |
| AB57_RS11370 | ABUW_1772 | AB57_RS09425 | ABUW_2167 | AB57_RS08465 | ABUW_2361 |
| AB57_RS11350 | ABUW_1776 | AB57_RS09410 | ABUW_2170 | AB57_RS08460 | ABUW_2362 |
| AB57_RS11345 | ABUW_1777 | AB57_RS09370 | ABUW_2178 | AB57_RS08455 | ABUW_2363 |
| AB57_RS11335 | ABUW_1779 | AB57_RS09350 | ABUW_2182 | AB57_RS08435 | ABUW_2367 |
| AB57_RS11330 | ABUW_1780 | AB57_RS09220 | ABUW_2208 | AB57_RS08335 | ABUW_2387 |
| AB57_RS11240 | ABUW_1799 | AB57_RS09215 | ABUW_2209 | AB57_RS08330 | ABUW_2389 |

Table S5. continued.

| With AB5075 (<i>Galleria mellonella</i> infection model)^c | | | | | |
|---|------------------|------------------|------------------|------------------|------------------|
| AB0057 | AB5075 | AB0057 | AB5075 | AB0057 | AB5075 |
| Locus_tag | Locus_tag | Locus_tag | Locus_tag | Locus_tag | Locus_tag |
| AB57_RS08325 | ABUW_2390 | AB57_RS07090 | ABUW_2652 | AB57_RS05780 | ABUW_2829 |
| AB57_RS08230 | ABUW_2409 | AB57_RS07030 | ABUW_2664 | AB57_RS05760 | ABUW_2832 |
| AB57_RS08135 | ABUW_2428 | AB57_RS07020 | ABUW_2666 | AB57_RS05730 | ABUW_2838 |
| AB57_RS08130 | ABUW_2429 | AB57_RS07015 | ABUW_2667 | AB57_RS05725 | ABUW_2839 |
| AB57_RS08125 | ABUW_2430 | AB57_RS07005 | ABUW_2669 | AB57_RS05720 | ABUW_2840 |
| AB57_RS08005 | ABUW_2455 | AB57_RS06935 | ABUW_2683 | AB57_RS05710 | ABUW_2842 |
| AB57_RS07990 | ABUW_2458 | AB57_RS06905 | ABUW_2690 | AB57_RS05705 | ABUW_2843 |
| AB57_RS07915 | ABUW_2473 | AB57_RS06815 | ABUW_2709 | AB57_RS05690 | ABUW_2846 |
| AB57_RS07910 | ABUW_2474 | AB57_RS06790 | ABUW_2714 | AB57_RS05675 | ABUW_2849 |
| AB57_RS07905 | ABUW_2475 | AB57_RS06775 | ABUW_2717 | AB57_RS05670 | ABUW_2850 |
| AB57_RS07805 | ABUW_2496 | AB57_RS06770 | ABUW_2718 | AB57_RS05665 | ABUW_2851 |
| AB57_RS07775 | ABUW_2502 | AB57_RS06760 | ABUW_2720 | AB57_RS05660 | ABUW_2852 |
| AB57_RS07770 | ABUW_2503 | AB57_RS06755 | ABUW_2721 | AB57_RS05630 | ABUW_2858 |
| AB57_RS07765 | ABUW_2504 | AB57_RS06750 | ABUW_2722 | AB57_RS05615 | ABUW_2862 |
| AB57_RS07695 | ABUW_2521 | AB57_RS06690 | ABUW_2734 | AB57_RS05585 | ABUW_2869 |
| AB57_RS07690 | ABUW_2522 | AB57_RS06650 | ABUW_2742 | AB57_RS05580 | ABUW_2870 |
| AB57_RS07620 | ABUW_2536 | AB57_RS06170 | ABUW_2752 | AB57_RS05575 | ABUW_2871 |
| AB57_RS07550 | ABUW_2555 | AB57_RS06140 | ABUW_2758 | AB57_RS05565 | ABUW_2874 |
| AB57_RS07505 | ABUW_2564 | AB57_RS06105 | ABUW_2765 | AB57_RS05560 | ABUW_2875 |
| AB57_RS07490 | ABUW_2567 | AB57_RS06100 | ABUW_2766 | AB57_RS05555 | ABUW_2876 |
| AB57_RS07485 | ABUW_2568 | AB57_RS06095 | ABUW_2767 | AB57_RS05550 | ABUW_2877 |
| AB57_RS07480 | ABUW_2569 | AB57_RS06055 | ABUW_2775 | AB57_RS05525 | ABUW_2882 |
| AB57_RS07460 | ABUW_2573 | AB57_RS06035 | ABUW_2779 | AB57_RS05485 | ABUW_2890 |
| AB57_RS07435 | ABUW_2578 | AB57_RS06030 | ABUW_2780 | AB57_RS05480 | ABUW_2892 |
| AB57_RS07430 | ABUW_2579 | AB57_RS06015 | ABUW_2783 | AB57_RS05465 | ABUW_2895 |
| AB57_RS07340 | ABUW_2598 | AB57_RS06010 | ABUW_2784 | AB57_RS05460 | ABUW_2896 |
| AB57_RS07335 | ABUW_2599 | AB57_RS06005 | ABUW_2785 | AB57_RS05455 | ABUW_2898 |
| AB57_RS07320 | ABUW_2602 | AB57_RS05985 | ABUW_2789 | AB57_RS05445 | ABUW_2900 |
| AB57_RS07315 | ABUW_2603 | AB57_RS05960 | ABUW_2794 | AB57_RS05425 | ABUW_2904 |
| AB57_RS07280 | ABUW_2611 | AB57_RS05940 | ABUW_2798 | AB57_RS05410 | ABUW_2907 |
| AB57_RS07255 | ABUW_2617 | AB57_RS17765 | ABUW_2800 | AB57_RS05400 | ABUW_2909 |
| AB57_RS07250 | ABUW_2618 | AB57_RS05915 | ABUW_2803 | AB57_RS05370 | ABUW_2915 |
| AB57_RS07220 | ABUW_2626 | AB57_RS05855 | ABUW_2815 | AB57_RS05350 | ABUW_2920 |
| AB57_RS07175 | ABUW_2635 | AB57_RS05850 | ABUW_2816 | AB57_RS05335 | ABUW_2923 |
| AB57_RS07165 | ABUW_2637 | AB57_RS05845 | ABUW_2817 | AB57_RS05330 | ABUW_2924 |
| AB57_RS07160 | ABUW_2638 | AB57_RS05825 | ABUW_2820 | AB57_RS05275 | ABUW_2935 |
| AB57_RS07155 | ABUW_2639 | AB57_RS05805 | ABUW_2824 | AB57_RS05265 | ABUW_2937 |
| AB57_RS07145 | ABUW_2641 | AB57_RS05800 | ABUW_2825 | AB57_RS05260 | ABUW_2939 |
| AB57_RS07135 | ABUW_2643 | AB57_RS05795 | ABUW_2826 | AB57_RS05240 | ABUW_2943 |
| AB57_RS07130 | ABUW_2644 | AB57_RS05785 | ABUW_2828 | AB57_RS05175 | ABUW_2956 |

Table S5. continued.

| With AB5075 (<i>Galleria mellonella</i> infection model) ^c | | | | | |
|--|-----------|--------------|-----------|--------------|-----------|
| AB0057 | AB5075 | AB0057 | AB5075 | AB0057 | AB5075 |
| Locus_tag | Locus_tag | Locus_tag | Locus_tag | Locus_tag | Locus_tag |
| AB57_RS05155 | ABUW_2960 | AB57_RS04005 | ABUW_3198 | AB57_RS02370 | ABUW_3506 |
| AB57_RS05145 | ABUW_2962 | AB57_RS03980 | ABUW_3204 | AB57_RS02345 | ABUW_3511 |
| AB57_RS05135 | ABUW_2964 | AB57_RS03975 | ABUW_3205 | AB57_RS02335 | ABUW_3513 |
| AB57_RS05085 | ABUW_2974 | AB57_RS03955 | ABUW_3209 | AB57_RS02325 | ABUW_3515 |
| AB57_RS05080 | ABUW_2975 | AB57_RS03830 | ABUW_3234 | AB57_RS02290 | ABUW_3522 |
| AB57_RS05075 | ABUW_2976 | AB57_RS03820 | ABUW_3236 | AB57_RS02285 | ABUW_3523 |
| AB57_RS04905 | ABUW_3016 | AB57_RS03800 | ABUW_3240 | AB57_RS02210 | ABUW_3538 |
| AB57_RS04900 | ABUW_3017 | AB57_RS03790 | ABUW_3242 | AB57_RS02085 | ABUW_3563 |
| AB57_RS04865 | ABUW_3024 | AB57_RS03775 | ABUW_3245 | AB57_RS01955 | ABUW_3589 |
| AB57_RS04840 | ABUW_3029 | AB57_RS03750 | ABUW_3250 | AB57_RS01850 | ABUW_3610 |
| AB57_RS04830 | ABUW_3031 | AB57_RS03705 | ABUW_3259 | AB57_RS01830 | ABUW_3614 |
| AB57_RS04780 | ABUW_3041 | AB57_RS03695 | ABUW_3261 | AB57_RS01720 | ABUW_3636 |
| AB57_RS04765 | ABUW_3045 | AB57_RS03655 | ABUW_3269 | AB57_RS01710 | ABUW_3638 |
| AB57_RS04755 | ABUW_3047 | AB57_RS03615 | ABUW_3277 | AB57_RS01660 | ABUW_3648 |
| AB57_RS04730 | ABUW_3052 | AB57_RS03590 | ABUW_3282 | AB57_RS01650 | ABUW_3650 |
| AB57_RS04715 | ABUW_3055 | AB57_RS03585 | ABUW_3283 | AB57_RS01645 | ABUW_3651 |
| AB57_RS04705 | ABUW_3057 | AB57_RS03565 | ABUW_3287 | AB57_RS00945 | ABUW_3740 |
| AB57_RS04700 | ABUW_3058 | AB57_RS03515 | ABUW_3297 | AB57_RS00940 | ABUW_3741 |
| AB57_RS04590 | ABUW_3078 | AB57_RS03490 | ABUW_3302 | AB57_RS00935 | ABUW_3742 |
| AB57_RS04565 | ABUW_3083 | AB57_RS03400 | ABUW_3320 | AB57_RS00930 | ABUW_3743 |
| AB57_RS04550 | ABUW_3086 | AB57_RS03380 | ABUW_3324 | AB57_RS00580 | ABUW_3815 |
| AB57_RS04545 | ABUW_3087 | AB57_RS03285 | ABUW_3344 | AB57_RS00570 | ABUW_3817 |
| AB57_RS04475 | ABUW_3101 | AB57_RS03270 | ABUW_3347 | AB57_RS00565 | ABUW_3818 |
| AB57_RS04340 | ABUW_3129 | AB57_RS03255 | ABUW_3350 | AB57_RS00560 | ABUW_3819 |
| AB57_RS04335 | ABUW_3130 | AB57_RS03245 | ABUW_3352 | AB57_RS00540 | ABUW_3820 |
| AB57_RS04315 | ABUW_3134 | AB57_RS03205 | ABUW_3360 | AB57_RS00465 | ABUW_3833 |
| AB57_RS04295 | ABUW_3138 | AB57_RS03170 | ABUW_3367 | AB57_RS00450 | ABUW_3836 |
| AB57_RS04280 | ABUW_3141 | AB57_RS03135 | ABUW_3374 | AB57_RS00425 | ABUW_3841 |
| AB57_RS04275 | ABUW_3142 | AB57_RS03100 | ABUW_3381 | AB57_RS00400 | ABUW_3846 |
| AB57_RS04270 | ABUW_3143 | AB57_RS03080 | ABUW_3385 | AB57_RS00315 | ABUW_3863 |
| AB57_RS04265 | ABUW_3144 | AB57_RS02975 | ABUW_3406 | AB57_RS00240 | ABUW_3878 |
| AB57_RS04185 | ABUW_3160 | AB57_RS02965 | ABUW_3408 | AB57_RS00090 | ABUW_4003 |
| AB57_RS04180 | ABUW_3161 | AB57_RS02755 | ABUW_3431 | AB57_RS00085 | ABUW_4004 |
| AB57_RS04110 | ABUW_3175 | AB57_RS02660 | ABUW_3447 | AB57_RS00080 | ABUW_4005 |
| AB57_RS04095 | ABUW_3178 | AB57_RS02620 | ABUW_3455 | AB57_RS00075 | ABUW_4006 |
| AB57_RS04085 | ABUW_3180 | AB57_RS02605 | ABUW_3458 | AB57_RS00030 | ABUW_4009 |
| AB57_RS04080 | ABUW_3181 | AB57_RS02590 | ABUW_3461 | AB57_RS00010 | ABUW_4013 |
| AB57_RS04070 | ABUW_3183 | AB57_RS02560 | ABUW_3467 | AB57_RS19380 | ABUW_4021 |
| AB57_RS04055 | ABUW_3186 | AB57_RS02525 | ABUW_3474 | AB57_RS19370 | ABUW_4023 |
| AB57_RS04010 | ABUW_3197 | AB57_RS02505 | ABUW_3478 | AB57_RS19360 | ABUW_4025 |

Table S5. continued.

With AB5075 (*Galleria mellonella* infection model)^c

| AB0057 | AB5075 |
|------------------|------------------|
| Locus_tag | Locus_tag |
| AB57_RS19355 | ABUW_4026 |
| AB57_RS19350 | ABUW_4027 |
| AB57_RS19340 | ABUW_4029 |
| AB57_RS19335 | ABUW_4030 |
| AB57_RS19330 | ABUW_4031 |
| AB57_RS19325 | ABUW_4033 |
| AB57_RS19305 | ABUW_4037 |
| AB57_RS19280 | ABUW_4042 |
| AB57_RS19135 | ABUW_4074 |
| AB57_RS19130 | ABUW_4075 |
| AB57_RS19110 | ABUW_4079 |
| AB57_RS19105 | ABUW_4080 |
| AB57_RS19100 | ABUW_4081 |
| AB57_RS19095 | ABUW_4082 |
| AB57_RS19090 | ABUW_4083 |
| AB57_RS19065 | ABUW_4090 |
| AB57_RS19060 | ABUW_4091 |
| AB57_RS19040 | ABUW_4095 |
| AB57_RS19015 | ABUW_4100 |
| AB57_RS19005 | ABUW_4102 |
| AB57_RS18995 | ABUW_4104 |
| AB57_RS18985 | ABUW_4106 |
| AB57_RS18980 | ABUW_4107 |
| AB57_RS18960 | ABUW_4111 |
| AB57_RS18955 | ABUW_4112 |
| AB57_RS18950 | ABUW_4113 |
| AB57_RS18940 | ABUW_4115 |
| AB57_RS18925 | ABUW_4118 |

Reference

^a Wang, N., Ozer, E.A., Mandel, M.J., and Hauser, A.R. (2014) Genome-wide identification of *Acinetobacter baumannii* genes necessary for persistence in the lung. *MBio* 5: e01163-01114.

^b Subashchandrabose, S., Smith, S., DeOrnellas, V., Crepin, S., Kole, M., Zahdeh, C., and Mobley, H.L. (2016) *Acinetobacter baumannii* genes required for bacterial survival during bloodstream infection. *mSphere* 1: e00013-15.

^c Gebhardt, M.J., Gallagher, L.A., Jacobson, R.K., Usacheva, E.A., Peterson, L.R., Zurawski, D.V., and Shuman, H.A. (2015) Joint transcriptional control of virulence and resistance to antibiotic and environmental stress in *Acinetobacter baumannii*. *MBio* 6: e01660-01615.

Table S6. Strains and plasmids used in this study.

| Strains | Characteristic(s) ^a | Source or reference |
|----------------------------------|--|----------------------------------|
| <i>E. coli</i> | | |
| MGN-617 | <i>hi thr leu tonA lacY glnV supE ΔasdA4 recA::RP4 2-Tc::Mu [pir]; Km^r</i> | (Dozois <i>et al.</i> , 2000) |
| DY330 | W3110 <i>ΔlacU169 gal490 λcl857 Δ(cro-bioA)</i> | (Yu <i>et al.</i> , 2000) |
| DH5 α λ pir | <i>sup E44, ΔlacU169 (ΦlacZΔM15), recA1, endA1, hsdR17, thi-1, gyrA96, relA1, λpir</i> phage lysogen | Laboratory collection |
| <i>A. baumannii</i> | | |
| AB0057 | MDR bloodstream isolate | (Hujer <i>et al.</i> , 2006) |
| AB0057 ^{Km} ; 57; WT | AB0057 <i>Δkm::FRT</i> ; Km susceptible | This study |
| <i>ΔhcaR</i> | AB0057 ^{Km} <i>ΔhcaR</i> | This study |
| <i>Δ0044</i> | AB0057 ^{Km} <i>ΔAB57_0044</i> | This study |
| <i>ΔvipAB</i> | AB0057 ^{Km} <i>ΔvipAB</i> | This study |
| <i>ΔfilA</i> | AB0057 ^{Km} <i>ΔfilA</i> | This study |
| <i>ΔbfmR</i> | AB0057 ^{Km} <i>ΔbfmR</i> | This study |
| <i>ΔgltP</i> | AB0057 ^{Km} <i>ΔgltP</i> | This study |
| 57 <i>ΔmltB</i> (AB57_2749) | AB0057 ^{Km} <i>ΔmltB</i> | This study |
| 57 eV | AB0057 ^{Km} + pABBR_Km; Km ^R | This study |
| 57 <i>ΔmltB</i> eV | AB0057 ^{Km} <i>ΔmltB</i> + pABBR_Km; Km ^R | This study |
| 57 <i>ΔmltB</i> compl. | AB0057 ^{Km} <i>ΔmltB</i> + pABBR_Km- <i>mrdb-mltB</i> ; Km ^R | This study |
| AB5075 | MDR Tibia/osteomyelitis isolate | (Jacobs <i>et al.</i> , 2014) |
| 75 <i>ΔmltB</i> | ABUW <i>Δ1243::T26 tnaB1_kr130917p02q159</i> | (Gallagher <i>et al.</i> , 2015) |
| Plasmids | | |
| pKD3 | Template plasmid for the amplification of the Cm cassette bordered by FRT sites; Ap ^r Cm ^r | (Datsenko & Wanner, 2000) |
| pKD4 | Template plasmid for the amplification of the Km cassette bordered by FRT sites; Ap ^r Km ^r | (Datsenko & Wanner, 2000) |
| pSU2719 | P15A replicon, <i>lacZα</i> , Cm ^r | (Martinez <i>et al.</i> , 1988) |

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|---------------------|--|-------------------------------|
| pKD_Zeo | Template plasmid for the amplification of the Zeo cassette bordered by FRT sites; Ap ^r Zeo ^r | This study |
| pSUABKm | pSU2719-AB57_0288; Cm ^r , Km ^r | This study |
| pSU ΔABKm::zeo | pSU2719-ΔAB57_0288::Zeo; Cm ^r , Km ^r | This study |
| pAT03 | pMMB67EH with FLP recombinase; Ap ^r | (Tucker <i>et al.</i> , 2014) |
| pAT03_Km | pAT03_Km; Km ^r | This study |
| pCVD442 | Suicide vector containing <i>sacB</i> ; Ap ^r | (Donnenberg & Kaper, 1991) |
| pCVD442_MCS | pCVD442 + MCS; The IS1 element was swapped by a MCS; Ap ^r , R6K, <i>sacB</i> | This study |
| pCVD_Amk | pCVD442_MCS_Amk; Amk ^r , R6K, <i>sacB</i> | This study |
| pCVD_AmK_ΔhcaR | Δ <i>hcaR</i> in pCVD_AmK; Amk ^r , R6K, <i>sacB</i> | This study |
| pCVD_AmK_ΔAB57_0044 | Δ <i>AB57_0044</i> in pCVD_AmK; Amk ^r , R6K, <i>sacB</i> | This study |
| pCVD_AmK_ΔvipAB | Δ <i>vipAB</i> in pCVD_AmK; Amk ^r , R6K, <i>sacB</i> | This study |
| pCVD_AmK_ΔfilA | Δ <i>filA</i> in pCVD_AmK; Amk ^r , R6K, <i>sacB</i> | This study |
| pCVD_AmK_ΔbfmR | Δ <i>bfmR</i> in pCVD_AmK; Amk ^r , R6K, <i>sacB</i> | This study |
| pCVD_AmK_ΔgltP | Δ <i>gltP</i> in pCVD_AmK; Amk ^r , R6K, <i>sacB</i> | This study |
| pABBR_MCS | <i>ori</i> of pWH1266 cloned into pBR322; Tet ^r , Ap ^r | (Tucker <i>et al.</i> , 2014) |
| pABBR_Km | pABBR_MCS_Km; Km ^r | This study |
| pABBR_Km-mrdB-mltB | Genes <i>mrdB-mltB</i> cloned into pABBR_Km; Km ^r | This study |

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Table S7. Primers and oligonucleotides.

| Name | Sequence 5' → 3' | Purpose |
|--------------------|---|---|
| TraDIS | | |
| Tn-specific primer | AATGATACGGCGACCACCGAGATCTACAA GGCATGCAAGCTTCAGGGTTGAGATGTGT ATAAG | Tn-gDNA junction amplification |
| Pri_Adap_#1_R | CAAGCAGAAGACGGCATA <u>CGAGACGTGAT</u> GTGACTGGA | Tn-gDNA junction amplification of input #1 from pool #1. Used with Tn-specific primer. |
| Pri_Adap_#2_R | CAAGCAGAAGACGGCATA <u>CGAGATACATC</u> GGTGACTGGA | Tn-gDNA junction amplification of input #2 from pool #1. Used with Tn-specific primer. |
| Pri_Adap_#3_R | CAAGCAGAAGACGGCATA <u>CGAGATGCCTA</u> AGTGACTGGA | Tn-gDNA junction amplification of input #1 from pool #2. Used with Tn-specific primer. |
| Pri_Adap_#4_R | CAAGCAGAAGACGGCATA <u>CGAGATTGGTC</u> AGTGACTGGA | Tn-gDNA junction amplification of input #2 from pool #2. Used with Tn-specific primer. |
| Pri_Adap_#5_R | CAAGCAGAAGACGGCATA <u>CGAGATCACTG</u> TGTGACTGGA | Tn-gDNA junction amplification of input #1 from pool #3. Used with Tn-specific primer. |
| Pri_Adap_#6_R | CAAGCAGAAGACGGCATA <u>CGAGATATTGG</u> CGTGACTGGA | Tn-gDNA junction amplification of input #2 from pool #3. Used with Tn-specific primer. |
| Pri_Adap_#7_R | CAAGCAGAAGACGGCATA <u>CGAGATGATCT</u> GGTGACTGGA | Tn-gDNA junction amplification of input #1 from pool #4. Used with Tn-specific primer. |
| Pri_Adap_#8_R | CAAGCAGAAGACGGCATA <u>CGAGATTCAAG</u> TGTGACTGGA | Tn-gDNA junction amplification of input #2 from pool #4. Used with Tn-specific primer. |
| Pri_Adap_#9_R | CAAGCAGAAGACGGCATA <u>CGAGATCTGAT</u> CGTGACTGGA | Tn-gDNA junction amplification of input #1 from pool #5. Used with Tn-specific primer. |
| Pri_Adap_#10_R | CAAGCAGAAGACGGCATA <u>CGAGATAAGCT</u> AGTGACTGGA | Tn-gDNA junction amplification of input #2 from pool #5. Used with Tn-specific primer. |
| Pri_Adap_#11_R | CAAGCAGAAGACGGCATA <u>CGAGATGTAGC</u> CGTGACTGGA | Tn-gDNA junction amplification of output #1 from pool #1. Used with Tn-specific primer. |
| Pri_Adap_#12_R | CAAGCAGAAGACGGCATA <u>CGAGATTACAA</u> GGTGACTGGA | Tn-gDNA junction amplification of output #2 from pool #1. Used with Tn-specific primer. |
| Pri_Adap_#13_R | CAAGCAGAAGACGGCATA <u>CGAGATTGTTG</u> ACTGTGACTGGA | Tn-gDNA junction amplification of output #3 from pool #1. Used with Tn-specific primer. |
| Pri_Adap_#14_R | CAAGCAGAAGACGGCATA <u>CGAGATACGGA</u> ACTGTGACTGGA | Tn-gDNA junction amplification of output #4 from pool #1. Used with Tn-specific primer. |

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|----------------|--|---|
| Pri_Adap_#15_R | CAAGCAGAAGACGGCATA <u>CGAGATTCTGA</u> <u>CATGTGACTGGA</u> | Tn-gDNA junction amplification of output #1 from pool #2. Used with Tn-specific primer. |
| Pri_Adap_#16_R | CAAGCAGAAGACGGCATA <u>CGAGATCGGGA</u> <u>CGGGTGACTGGA</u> | Tn-gDNA junction amplification of output #2 from pool #2. Used with Tn-specific primer. |
| Pri_Adap_#17_R | CAAGCAGAAGACGGCATA <u>CGAGATCTCTA</u> <u>CGTGACTGGA</u> | Tn-gDNA junction amplification of output #3 from pool #2. Used with Tn-specific primer. |
| Pri_Adap_#18_R | CAAGCAGAAGACGGCATA <u>CGAGATGTGCG</u> <u>GACGTGACTGGA</u> | Tn-gDNA junction amplification of output #4 from pool #2. Used with Tn-specific primer. |
| Pri_Adap_#19_R | CAAGCAGAAGACGGCATA <u>CGAGATCGTTT</u> <u>CACGTGACTGGA</u> | Tn-gDNA junction amplification of output #1 from pool #3. Used with Tn-specific primer. |
| Pri_Adap_#20_R | CAAGCAGAAGACGGCATA <u>CGAGATAAGGC</u> <u>CACGTGACTGGA</u> | Tn-gDNA junction amplification of output #2 from pool #3. Used with Tn-specific primer. |
| Pri_Adap_#21_R | CAAGCAGAAGACGGCATA <u>CGAGATTCCGA</u> <u>AACGTGACTGGA</u> | Tn-gDNA junction amplification of output #3 from pool #3. Used with Tn-specific primer. |
| Pri_Adap_#22_R | CAAGCAGAAGACGGCATA <u>CGAGATTACGT</u> <u>ACGGTGACTGGA</u> | Tn-gDNA junction amplification of output #4 from pool #3. Used with Tn-specific primer. |
| Pri_Adap_#23_R | CAAGCAGAAGACGGCATA <u>CGAGATATCCA</u> <u>CTCGTGACTGGA</u> | Tn-gDNA junction amplification of output #1 from pool #4. Used with Tn-specific primer. |
| Pri_Adap_#24_R | CAAGCAGAAGACGGCATA <u>CGAGATGCTAC</u> <u>CGTGACTGGA</u> | Tn-gDNA junction amplification of output #2 from pool #4. Used with Tn-specific primer. |
| Pri_Adap_#25_R | CAAGCAGAAGACGGCATA <u>CGAGATATATCA</u> <u>GTGTGACTGGA</u> | Tn-gDNA junction amplification of output #3 from pool #4. Used with Tn-specific primer. |
| Pri_Adap_#26_R | CAAGCAGAAGACGGCATA <u>CGAGATGCTCA</u> <u>IGTGACTGGA</u> | Tn-gDNA junction amplification of output #4 from pool #4. Used with Tn-specific primer. |
| Pri_Adap_#27_R | CAAGCAGAAGACGGCATA <u>CGAGATAAAGG</u> <u>AATGTGACTGGA</u> | Tn-gDNA junction amplification of output #1 from pool #5. Used with Tn-specific primer. |
| Pri_Adap_#28_R | CAAGCAGAAGACGGCATA <u>CGAGATCTTTTG</u> <u>GTGACTGGA</u> | Tn-gDNA junction amplification of output #2 from pool #5. Used with Tn-specific primer. |
| Pri_Adap_#29_R | CAAGCAGAAGACGGCATA <u>CGAGATTAGTI</u> <u>GGTGACTGGA</u> | Tn-gDNA junction amplification of output #3 from pool #5. Used with Tn-specific primer. |
| Pri_Adap_#30_R | CAAGCAGAAGACGGCATA <u>CGAGATCCGGT</u> <u>GGTGACTGGA</u> | Tn-gDNA junction amplification of output #4 from pool #5. Used with Tn-specific primer. |

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| Sequencing primer | GCATGCAAGCTTCAGGGTTGAGATGTGTAT AAG | Sequencing the Tn-gDNA junction |
| Plasmids | | |
| pKD_R_BamH1 | GGGGGGATCCGCGCGCCATTTAAA TGAAGTTCCT | Inverted PCR of pKD3 to remove the <i>cat</i> cassette |
| pKD_F_for_Bpu10I | GAGCAACTGACTGAAATGCCTCAA* | |
| sh_ble_F(2)_Bam H1 | CATAGGATCCGCTACCCGTGATATTGC TGAAGAG | Amplification of sh_ble (Zeo) from pCR-BluntII-TOPO and cloned into the pKD- Δ <i>cat</i> , creating of pKD_Zeo |
| sh_ble_R_Bpu10I | TCTTTTCGCTAAGGTCTGACGCTCAGT GGAACGAAA | |
| AB57_0288_5'_F_ Kpn1 | ACCGGTACCTGCGTGTGCTGCAAGGA AATC | Amplification of AB57_0288 and cloning into pSU2719, creating pSU_ABKm |
| AB57_0288_3'_R_ Sph1 | TGCGCATGCTCGCATGGTGTTCGGAAT CT | |
| AB57_0288_F_KO | GAACAATAAACTGTCTGCTTACATAAA CAGTAATACAAGGGGTGTTATGTGTGT AGGCTGGAGCTGCTTC | KO of AB57_0288 in pSU_ABKm by recombineering, creating pSU_ Δ ABKmssh_ble |
| AB57_0288_R_KO | CAGGTTTGTGTCTCTACAAAGACTAACT ATCAGAAAACTCATCGAGCATTGGTC CATATGAATATCCTCCTTAGTTCC | |
| Km_F_pvuI | CAAGCGATCGTAAAGGAAGCGGAACA CGTAGAAAG | Amplification of the <i>kan</i> cassette from pKD4 and cloning into pABBR_MCS, creating pABBR_Km |
| Km_R_SalI | GTCCGTCGACACGATTCCGAAGCCCAA CCTTTCA | |
| Amk_F_AatII | ATCGGACGTCAGGACAGAAATGCCTC GACTTC | Amplification of the amikacin cassette (gene <i>ABAYE3630</i>), and cloned into pCVD_MCS |
| Amk_R_BsaI | CAATGATACCGCGAGACCCACCGGGC TTTAAACTCGCCTCTAT | |
| 2749_F_compl_ PspX1 | AACTAACTCGAGGAGTCTATCGGAAA CCTGGTGAGAG | Cloning the <i>mdrB-mltB</i> locus into pABBR_Km vector. |
| 2749_R_compl_ NruI | GGGTGGTCCGGATTCATCGCTTACGG AGAGTGTCT | |
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| Mutants | | |
|---|--|---|
| KO_AB57_0044_F_for_Xbal_screen_(frag_5') | ACACCTGTTGCAGATGTAGATGTGTT* | Amplification of ~ 1 Kb upstream of <i>AB57_0044</i> |
| KO_AB57_0044_R_PvuI-HF_(frag_5') | ACAAT CGATCG AAAACGTCGCATATTATCC TAGTTCAATTT | |
| KO_AB57_0044_F_PvuI-HF_(frag_3') | GCCAAC CGATCG GCACGAAAAATGATGCAA TAAAATATTAATAAAAAAGG | Amplification of ~ 1 Kb downstream of <i>AB57_0044</i> |
| KO_AB57_0044_R_Sac1_(frag_5') | CGTT GAGCTC CTTTGTTATTAGGCATGGCC TTTGC | |
| KO_AB57_0044_R_screen | ACTTGCTCATGAGTTTGGACATGC | Screening to confirm the <i>AB57_0044</i> mutation; used with KO_AB57_0044_F_for_Xbal_s creen_(frag_5') |
| KO_AB57_2749_F_Sac1_(frag_5') | CGTAG GAGCTC AGTCTATCGGAAACCTGGT GAGAG | Amplification of ~ 1 Kb upstream of <i>AB57_2749</i> |
| KO_AB57_2749_R_MluI-Hf_(frag_5') | TTAAC ACGCGT AGACAACATATCTTTTTTAA ATTCCAGATTATCGAT | |
| KO_AB57_2749_F_MluI-HF_(frag_3') | GGTT ACGCGT ACAGAAGTAGCTAGCCGA TAGGCT | Amplification of ~ 1 Kb downstream of <i>AB57_2749</i> |
| KO_AB57_2749_R_for_Xbal_screen_(fr ag_3') | AGTGGTACTAGGGTAGCGGAATTAT* | |
| KO_AB57_2749_F_screen | CTAAAGCAGTTCATATTGCGATCGACA | Screening to confirm the <i>AB57_0044</i> mutation; used with KO_AB57_2749_R_for_Xbal_s creen_(frag_3') |
| KO_vipAB_F_MluI_(frag_5') | CGGC ACGCGT CATAAAAGTTAGCCTTGTTCC TTAACAAATTGT | Amplification of ~ 1 Kb upstream of <i>vipAB</i> |
| KO_vipAB_R_Sal1_(frag_5') | TTTTT GTCGACT CGCGTGTACTGACATAAA TAGTGTC | |
| KO_vipAB_R_MluI_(frag_3') | GTATC ACGCGT GCAATCAACTGGTATAGTA CTAAAATTTAGTCTTTGAAA | Amplification of ~ 1 Kb downstream of <i>vipAB</i> |
| KO_vipAB_F_Sac1_(frag_3') | CGCT GAGCTC TCCCTTCGGCCTGTATCAAC AT | |
| KO_vipAB_screen_F | TATCCTTTGTTTCATCTTCGCAATTTAC | Screening to confirm the <i>vipAB</i> mutation |

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|-------------------------------|--|---|
| KO_vipAB_screen_R | TAAATACGACTGGCGCTTGCA | |
| KO_RRcsu_fim_R_MluI_(Frag_5) | TTTC ACGCGT GTCTCATATCATTGCCCTATAAATCTCATT | Amplification of ~ 1 Kb downstream of <i>bfmR</i> |
| KO_RRcsu_fim_F_PvuI_(Frag_5) | TGCC ACGATCGT GGTGGTAGCAATATCGCTTTCTTT | |
| KO_RRcsu_fim_F_Xba1_(Frag_3') | TATCTTTATCT CTAGAGT CGACCTGATGCA TACGATGATT | Amplification of ~ 1 Kb upstream of <i>bfmR</i> |
| KO_RRcsu_fim_R_MluI_(Frag_3') | ACGCGT GTTAAAGAAACCAATGGATTGTAA AATCTGATTAACCTT | |
| KO_RRcsu_fim_F_screen | ATCTAGTGAAGGTGTACCCTGCTC | Screening to confirm the <i>bfmR</i> mutation |
| KO_RRcsu_fim_R_screen | CTCATCACGAAGCAAACGTGCA | |
| KO_FilA_F_MluI_(Frag_5) | GTGAA ACGCGT CATTGAGCACTCTCCCAA GAGC | Amplification of ~ 1 Kb upstream of <i>filA</i> |
| KO_FilA_R_Sac1_(Frag_5) | GTAT GAGCTC TTAGACCAGCGAGGATGAAT ATTTTGTTCCG | |
| KO_FilA_R_MluI_(Frag_3) | ATGTG ACGCGT TACAACATATCACAATATCTG GTCATTAATTTAAATATAAAAAAAGC | Amplification of ~ 1 Kb downstream of <i>filA</i> |
| KO_FilA_F_Xba1_(Frag_3) | CAACAAAAGCT CTAGA AATTGACTGTTGCAT CCTCTACATGAC | |
| KO_FilA_F_screen | CGCATCGTAGAAGCCTTATTTGCT | Screening to confirm the <i>filA</i> mutation |
| KO_FilA_R_screen | ATAACCAGAATACGGTGCATTGGT | |
| KO_hcaR_R_MluI_(frag_5) | AGATG ACGCGT TTCCATAAAGACTAATCCAA AAAATGTCTTACTATGAT | Amplification of ~ 1 Kb upstream of <i>hcaR</i> |
| KO_hcaR_F_Sal1_(frag_5) | ATCC GTCGAC AGGCGATCTAGGCTGCTGATTA | |
| KO_hcaR_F_MluI_(frag_3) | ACAAA ACGCGT CATCTTGATAAAAATCTAG ATTAAGCACCTTAGGT | Amplification of ~ 1 Kb downstream of <i>hcaR</i> |
| KO_hcaR_R_Sac1_(frag_3) | TGTTG GAGCTC TCAAACAGCGGAAGGCT TAAAAC | |
| KO_HcaR_screen_F | ATAAGACTCACGAGTGACTAACCAACG | |

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|----------------------|--|---|
| KO_HcaR_screen_R | GGA CTGAGTGAAGCAAATCCGTATG | Screening to confirm the <i>hcaR</i> mutation |
| KO_1698_F_Mlu1_Reg5' | aatttctt ACGCGT cataggaacctctatccttgggttc | Amplification of ~ 1 Kb upstream of <i>gltP</i> |
| KO_1698_R_Sac1_Reg5' | ttgtca GAGCTC atccgaacaaccaactggttc | |
| KO_1698_F_Xba1_Reg3' | gagcagtt TCTAGA cg cattgagccaacaagtacga | Amplification of ~ 1 Kb downstream of <i>gltP</i> |
| KO_1698_R_Mlu1_Reg3' | taaa ACGCGT caagaacaacaatcgcttaagtttct | |
| KO_1698_F_screen | atcagaagcggccaatgcaa | Screening to confirm the <i>gltP</i> mutation |
| KO_1698_R_screen | tcattccgatccggactatcc | |
| qRT-PCR | | |
| ESR | | |
| degP_F_qPCR | GTAATCAGAGCACCTTCCGGTTTAG | Amplification of a portion of <i>degP</i> |
| degP_R_qPCR | TGACTCGTTCATACCTAGGCGTTAT | |
| baeR_F_qPCR | TGTTACAGTACCTTACGGCAAATC | Amplification of a portion of <i>baeR</i> |
| baeR_R_qPCR | GGTCAGGATGCTTACACGAAC TTT | |
| rstA_F_qPCR | CGCCCACTATCATCAACCAA | Amplification of a portion of <i>rstA</i> |
| rstA_R_qPCR | GCACCCATTTCCAGACCAAGTA | |
| dsbA_F_qPCR | GGGTAGTGTAATGGCAGCAGATTT | Amplification of a portion of <i>dsbA</i> |
| dsbA_R_qPCR | GGTACTTCCACTTTGCCTGGATTG | |
| LT | | |
| LT_0044_F_qPCR | GAAGGACGCCGCAAAGTTATT | Amplification of a portion of <i>AB57_0044</i> |
| LT_0044_R_qPCR | CCAAATGCTCCTGCCCATGAA | |
| LT_1068_F_qPCR | GTGAAGTTGTTGCTGGTGGTTCTACC | Amplification of a portion of <i>AB57_1068</i> |
| LT_1068_R_qPCR | ATGCGCTCCATCATCCATGT | |
| LT_1136_F_qPCR | CCGACCACAACAAC TTTCAACCT | Amplification of a portion of <i>AB57_1136</i> |
| LT_1136_R_qPCR | TACCGTTACCGAGCTGCTACTT | |
| LT_2801_F_qPCR | GCCAGCGTTACATGGACATTAG | Amplification of a portion of <i>AB57_2801</i> |
| LT_2801_R_qPCR | ATAACGTGAGCGCGTGCATAAA | |
| LT_3476_F_qPCR | CGTTGTACGATCTGCCGATAAA | Amplification of a portion of <i>AB57_3476</i> |
| LT_3476_R_qPCR | ACCCAACCTATAACCAACCGAGAG | |
| PBP | | |
| PBP_0326_F_qPCR | TCCAATCCGTGATACTGTGGGTTAT | |

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|-----------------|--------------------------|---|
| PBP_0326_R_qPCR | CGGCACTGATGCGTTGGTATTT | Amplification of a portion of <i>AB57_0326</i> |
| pbp2_F_qPCR | CCGGTTTAATTCCAGCGGTCAT | Amplification of a portion of <i>pbp2</i> |
| pbp2_R_qPCR | TCAGCAGCCCAACTTGCTAAAC | |
| PBP_2186_F_qPCR | AGCTCAAGCTCGCATTGCTTTA | Amplification of a portion of <i>AB57_2186</i> |
| PBP_2186_R_qPCR | CATCCTTGCTGTGGGCCATTAT | |
| pbp1B_F_qPCR | CGGCTTTAACCGTGCCTTAGAT | Amplification of a portion of <i>pbp1B</i> |
| pbp1B_R_qPCR | GGAACGCTAATTGGAGCGTCTT | |
| PBP_2863_F_qPCR | CTGACCCAAGTGTTGATGGCTT | Amplification of a portion of <i>AB57_2863</i> |
| PBP_2863_R_qPCR | CACGTTTCAGACATGCTTGGTGTA | |
| pbp1A_F_qPCR | CGTGGTGCAGAAGCACATGATA | Amplification of a portion of <i>pbp1A</i> |
| pbp1A_R_qPCR | CACGGTACTGTTGAGCCATCTT | |
| | | |

Underlined nucleotides denote barcode sequence

Bold nucleotides denote restriction site

*: Restriction site is natively encoded within the amplified product