

Supplementary Materials: Genomic Analysis of a Newly Isolated *Acidithiobacillus ferridurans* JAGS Strain Reveals Its Adaptation to Acid Mine Drainage

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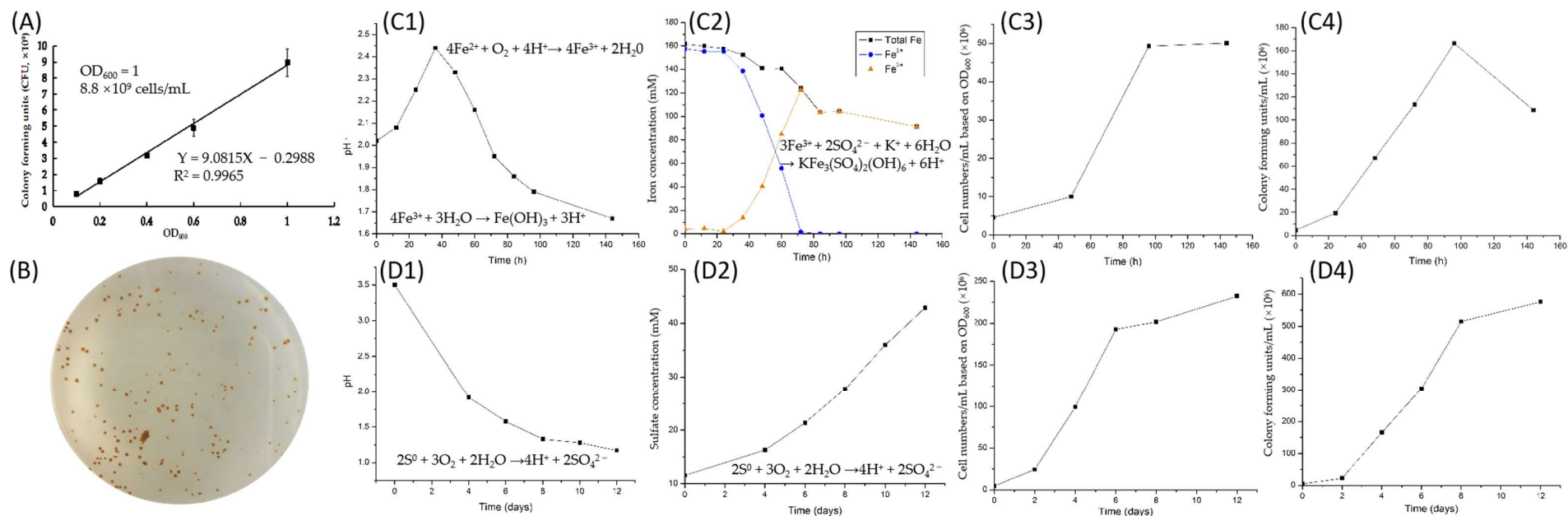


Figure S1. Growth features of *A. ferridurans* JAGS. **(A)** Standard curve of optical density (OD_{600}) versus cell numbers obtained by plate count method. **(B)** Colonies on 2:2 solid medium for cell count. **(C)** Growth on 9K- Fe^{2+} medium, **(C1)** pH value; **(C2)** Fe oxidation; **(C3)** cell numbers by OD_{600} ; **(C4)** cell numbers by plate count method. **(D)** Growth on 9K- S^0 medium, **(D1)** pH value; **(D2)** sulfur oxidation; **(D3)** cell count by OD_{600} ; **(D4)** cell numbers by plate count method.

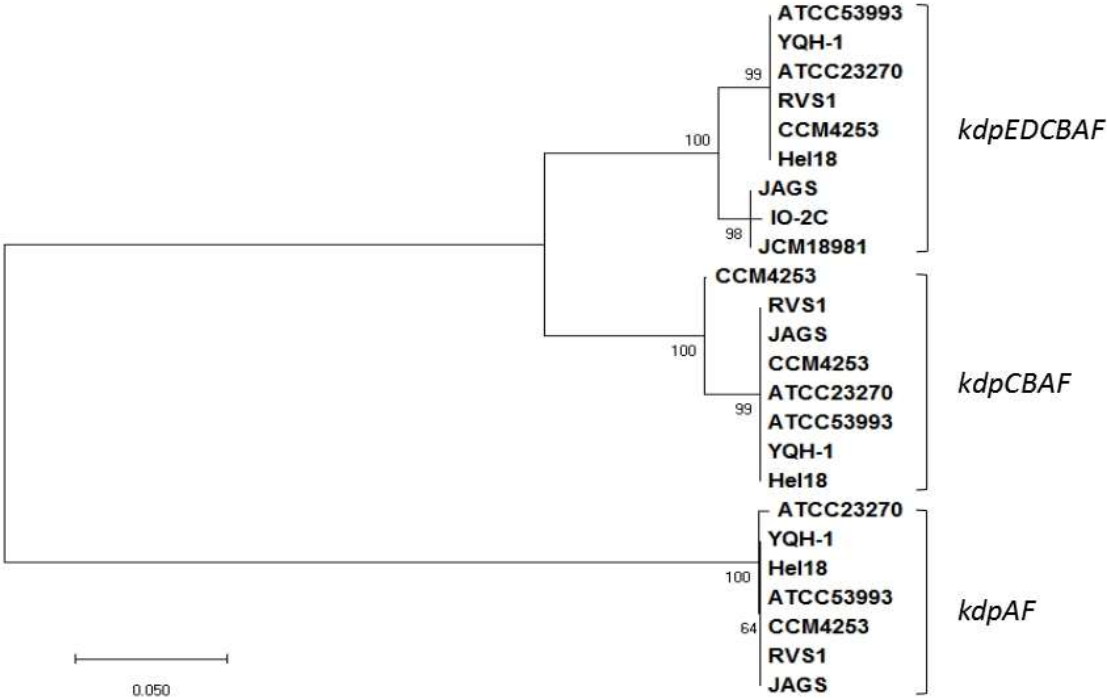


Figure S2. Neighbor-joining (NJ) phylogenetic tree of the *kdpA* protein sequences derived from nine *Acidithiobacillus* strains. Bootstrap values indicated at each node are based on a total of 500 bootstrap replicates.

Table S1. Average nucleotide identity (ANI) (%) based on whole-genome alignments among *Acidithiobacillus* strains by JSpeciesWS.

| * | <i>A. ferriidurans</i> JAGS | <i>A. ferriidurans</i> JCM18981 | <i>A. ferrooxidans</i> IO-2C | <i>A. ferrooxidans</i> ATCC53993 | <i>A. ferrooxidans</i> RVS1 | <i>A. ferrooxidans</i> CCM4253 | <i>A. ferrooxidans</i> YQH1 | <i>A. ferrooxidans</i> Hel18 | <i>A. ferrooxidans</i> ATCC23270 |
|----------------------------------|--------------------------------|------------------------------------|---------------------------------|-------------------------------------|--------------------------------|-----------------------------------|--------------------------------|---------------------------------|-------------------------------------|
| <i>A. ferriidurans</i> JAGS | * | 99.66 | 99.23 | 95.51 | 95.33 | 95.32 | 95.34 | 95.34 | 95.37 |
| <i>A. ferriidurans</i> JCM18981 | 99.13 | * | 99.22 | 95.13 | 95.08 | 95.07 | 95.05 | 95.06 | 95.12 |
| <i>A. ferrooxidans</i> IO-2C | 98.69 | 98.18 | * | 95.32 | 95.27 | 95.26 | 95.26 | 95.26 | 95.52 |
| <i>A. ferrooxidans</i> ATCC53993 | 94.98 | 94.11 | 94.89 | * | 99.86 | 99.78 | 99.8 | 99.8 | 99.87 |
| <i>A. ferrooxidans</i> RVS1 | 94.68 | 93.97 | 94.79 | 99.57 | * | 99.94 | 99.95 | 99.95 | 99.84 |
| <i>A. ferrooxidans</i> CCM4253 | 94.70 | 93.83 | 94.79 | 99.49 | 99.92 | * | 99.97 | 99.98 | 99.82 |
| <i>A. ferrooxidans</i> YQH1 | 94.71 | 93.83 | 94.8 | 99.5 | 99.93 | 99.97 | * | 99.99 | 99.83 |
| <i>A. ferrooxidans</i> Hel18 | 94.69 | 93.83 | 94.77 | 99.49 | 99.91 | 99.97 | 99.97 | * | 99.82 |
| <i>A. ferrooxidans</i> ATCC23270 | 94.76 | 94.00 | 94.81 | 99.64 | 99.56 | 98.93 | 99.1 | 99.04 | * |

Note: lower matrix ANIb, upper matrix ANIm.

Table S2. Genes predicted to be involved in acid and heavy metal tolerance in *A. ferriidurans* JAGS.

| Protein | Gene ID |
|--|-------------------|
| Acid Stress Resistance | - |
| OMP40, outer membrane protein. | F6A13_00370 |
| HpnB, glycosyltransferase. | F6A13_01630 |
| HpnMHNKJIAG cluster, hopanoid-associated proteins. | F6A13_09105-09150 |
| Shc, squalene-hopene cyclase (EC 5.4.99.17). | F6A13_09155 |
| Sqs, squalene synthase (EC 2.5.1.21). | F6A13_09160 |
| kdpD, osmosensitive K ⁺ channel histidine kinase | F6A13_02550 |
| KdpEDFABC cluster, a kdp-type potassium uptake ATPase system. | F6A13_09580-09605 |
| KdpCBFAFAF cluster, a kdp-type potassium uptake ATPase system. | F6A13_11005-11030 |
| TrkA, Voltage-gated potassium channel protein. | F6A13_13725 |
| NhaA, Na ⁺ /H ⁺ antiporter. | F6A13_09475 |
| putative Na ⁺ /H ⁺ antiporter. | F6A13_04755 |
| SpeA, biosynthetic arginine decarboxylase (EC 4.1.1.19). | F6A13_06090 |
| PanD, aspartate 1-decarboxylase (EC 4.1.1.11). | F6A13_14595 |
| Psd, phosphatidylserine decarboxylase (EC 4.1.1.65). | F6A13_04545 |
| ClpB, chaperone protein ClpB (ATP-dependent unfoldase). | F6A13_08410 |
| DnaJK, chaperone proteins. | F6A13_12450-12455 |
| GrpE, heat shock protein. | F6A13_12460 |
| RadA, DNA repair protein. | F6A13_13190 |
| DnaB, replicative DNA helicase (DnaB) (EC 3.6.4.12). | F6A13_13200 |

| | |
|--|---|
| RecA, DNA repair protein. | F6A13_05400 |
| LexA-ImuAB-DnaE cluster: LexA, SOS-response repressor and protease LexA (EC 3.4.21.88); ImuA, RecA/RadA recombinase; ImuB, DNA polymerase IV-like protein; DnaE, error-prone repair homolog of DNA polymerase III alpha subunit (EC 2.7.7.7). | F6A13_07785-07800 |
| Heavy-Metal Resistance | - |
| Lead, cadmium, zinc and mercury transporting ATPase (EC 3.6.3.3) (EC 3.6.3.5); Copper-translocating P-type ATPase (EC 3.6.3.4). MgtA, Magnesium (Mg ²⁺) transport ATPase, P-type (EC 3.6.3.2) CopZ, Copper(I) chaperone RND efflux system (Ni/Co/Cd/Zn). | F6A13_04860, 04905, 04925, 04945, 08625, 10865, 10890, 11740 F6A13_09840 F6A13_04870, 07760, 10900 F6A13_08730-08740, 15065-15075 F6A13_00815-00825, 07835-07845, 08240-08250, 08695-08705, 10835-10845, 11760-11770 |
| CzcABC family RND transporter system (Ni/Mn/Fe/Mo). | F6A13_05895, 06205, 08215, 10785 |
| CzcA/D, Co/Zn/Cd resistance proteins. | F6A13_03815-03830 |
| ZnuACBB, Zinc ABC transporter system. | F6A13_14780-14795 |
| CorA, Mg/Co transport proteins. | F6A13_03800, 08900 |
| CorC, Mg/Co transport proteins | F6A13_13815 |
| Cation-transporting ATPase | F6A13_00710-00725 |
| MerRTPA, Hg resistance. | F6A13_11040-11055 |
| MerCAD, Hg resistance. | F6A13_09070 |
| ArsM, Arsenite methyltransferase (EC 2.1.1.137) | F6A13_10625-10635; |
| ArsADC, As resistance. | F6A13_13380-13405 |
| ArsHBRCD, As resistance. | F6A13_13430, 13435 |
| ArsRC, As resistance. | F6A13_00800, 05345 |
| MntH, Manganese transport protein. | |