**The Ferredoxin-like Proteins HydN and YsaA Enhance Redox Dye-linked Activity of the Formate Dehydrogenase H Component of the Formate Hydrogenlyase Complex**

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**Supplemental Information**

**Table S1: Oligonucleotides used for cloning of bacterial two hybrid constructs.**

|  |  |  |
| --- | --- | --- |
|  | target vector | |
| gene | pUT18a | pT25a |
| *aegA* | T18-aegAFW\_HindIII  gcg AAGCTT g atgAATCGTTTTATTATGGC  T18-aegARW\_EcoRI  gcg GAATTC ga GTGAGATTTGACTGATTTTAC | T18C/T25-aegAFW\_BamHI  gcg GGATCC c AATCGTTTTATTATGGCCAAC  T18C/T25-aegARW\_KpnI  gcg GGTACC tcaGTGAGATTTGACTGATTTTAC |
| *fdhF* | FdhF\_pUT18FW\_HindIII  gcg AAGCTTg atgAAAAAAGTCGTCACGGT  HycB\_pUT18RW\_EcoRI  gcg GAATTC ga CGCCAGTGCCGCTTCGCGCA | FdhF\_pT25FW\_PstI  gcg CTGCAGgg AAAAAAGTCGTCACGGTTTG  FdhF\_pT25RW\_BamHI  gcg GGATCC ttaCGCCAGTGCCGCTTCGC |
| *fdnH* | T18-fdnHFW\_HindIII  gcg AAGCTT g atgGCTATGGAAACGCAGG  T18-fdnHRW\_EcoRI  gcg GAATTC gaCTCATGATGATCCTCCTCGTC | T18C/T25-fdnHFW\_BamHI  gcg GGATCC cGCTATGGAAACGCAGGACAT  T18C/T25-fdnHRW\_KpnI  gcg GGTACC ttaCTCATGATGATCCTCC |
| *fdx* | T18-fdxFW\_HindIII  gcg AAGCTT g atgCCAAAGATTGTTATTTTG  T18-fdxRW\_EcoRI  gcg GAATTC gaATGCTCACGCGCATGGTTG | T18C/T25-fdxFW\_BamHI  gcg GGATCC c CCAAAGATTGTTATTTTGCC  T18C/T25-fdxRW\_KpnI  gcg GGTACC ttaATGCTCACGCGCATGGT |
| *hycB* | HycB\_pUT18FW\_HindIII  gcg AAGCTTg atgAATCGTTTTGTAATTGCTG  HycB\_pUT18RW\_EcoRI  gcg GAATTC ga TTTAGCCTCTCCACTTTGAG | HycB\_pT25FW\_PstI  gcg CTGCAGgg AATCGTTTTGTAATTGCTGAC  HycB\_pT25RW\_BamHI  gcg GGATCC tcaTTTAGCCTCTCCACTTT |
| *hycF* | HycF\_pUT18\_PstI  gcg CTGCAGg atgTTTACCTTTATCAAAAAAG  HycF\_pUT18\_EcoRI  gcg gaattc gaGATGGCCTCTTTCATATGGC | HycF\_pT25FW\_PstI  gcg CTGCAGgg TTTACCTTTATCAAAAAAGT  HycF\_pT25RW\_BamHI  gcg GGATCC tcaGATGGCCTCTTTCATATG |
| *hydN* | HydN\_pUT18FW\_HindIII  gcg AAGCTTg atgAACCGTTTCATCATTGC  HydN\_pUT18RW\_EcoRI  gcg GAATTCga GAACATCAGCGCCGTACGGC | HydN\_pT25FW\_PstI  gcg CTGCAGgg AACCGTTTCATCATTGCTGAC  HydN\_pUT18CRW\_BamHI  gcg GGATCC ttaGAACATCAGCGCCGTAC |
| *hyfA* | HyfA\_T18FW\_KpnI  gcg GGTACC atgAACCGCTTTGTGGTGGC  HyfA\_T18RW\_EcoRI  gcg GAATTC ga GCGTTGCTCCTGAGTGAGGG | HyfA\_PT25FW\_KpnI  gcg GGTACC t AACCGCTTTGTGGTGGCCGA  HyfA\_pT25RW\_KpnI  gcg GGTACC ttaGCGTTGCTCCTGAGTGAG |
| *narH* | T18-narHFW\_HindIII  gcg AAGCTT g atgAAAATTCGTTCACAAG  T18-narHRW\_EcoRI  gcg GAATTC ga TGGATGCGGCTCCGTTTTGC | T18C/T25-narHFW\_KpnI  gcg GGTACC t AAAATTCGTTCACAAGTCGG  T18C/T25-narHRW\_KpnI  gcg GGTACC tcaTGGATGCGGCTCCGTTTTG |
| *nuoE* | T18-nuoEFW\_HindIII  gcg AAGCTT g atgCACGAGAATCAACAACC  T18-nuoERW\_EcoRI  gcg GAATTC ga TTTATACCGCTCCAGCAGTTC | T18C/T25-nuoEFW\_BamHI  gcg GGATCC c CACGAGAATCAACAACCACA  T18C/T25-nuoERW\_KpnI  gcg GGTACC tta tcaTTTATACCGCTCCAGCAG |
| *ysaA* | T18-ysaAFW\_HindIII  gcg AAGCTT g atgAACCGGTTTATTATTGCG  T18-ysaARW\_EcoRI  gcg GAATTC ga AACAGGCTGCTGCCGTAGCC | T18C/T25-ysaAFW\_BamHI  gcg GGATCC c AACCGGTTTATTATTGCGGAT  T18C/T25-ysaARW\_KpnI  gcg GGTACC tta tcaAACAGGCTGCTGCCGTAGC |

aThe box contains the name of the forward oligonucleotide followed by its sequence in 5’ to 3’ direction and then the same for the reverse oligonucleotide.

**Table S2:** Plasmids of the bacterial two hybrid system constructed here.

|  |  |  |
| --- | --- | --- |
| Plasmid | Genotype | Source |
| pT18-AegA | pT18, AegA-T18 fusion protein, *aegA* cloned HindIII/EcoRI, AmpR | This work |
| pT18-FdhF | pT18, FdhF-T18 fusion protein, *fdhF* cloned HindIII/EcoRI, AmpR | This work |
| pT18-FdnH | pT18, FdnH-T18 fusion protein, *fdnH* cloned HindIII/EcoRI, AmpR | This work |
| pT18-Fdx | pT18, Fdx-T18 fusion protein, *fdx* cloned HindIII/EcoRI, AmpR | This work |
| pT18-HycB | pT18, HycB-T18 fusion protein, *hycB* cloned HindIII/EcoRI, AmpR | This work |
| pT18-HycF | pT18, HycF-T18 fusion protein, *hycF* cloned PstI/EcoRI, AmpR | This work |
| pT18-HydN | pT18, HydN-T18 fusion protein, *hydN* cloned HindIII/EcoRI, AmpR | This work |
| pT18-HyfA | pT18, HyfA-T18 fusion protein, *hyfA* cloned KpnI/EcoRI, AmpR | This work |
| pT18-NarH | pT18, NarH-T18 fusion protein, *narH* cloned HindIII/EcoRI, AmpR | This work |
| pT18-NuoE | pT18, NuoE-T18 fusion protein, *nuoE* cloned HindIII/EcoRI, AmpR | This work |
| pT18-YsaA | pT18, YsaA-T18 fusion protein, *ysaA* cloned HindIII/EcoRI, AmpR | This work |
| pT25-AegA | pT25, T25-AegA fusion protein, *aegA* cloned BamHI/KpnI, CmR | This work |
| pT25-FdhF | pT25, T25-FdhF fusion protein, *fdhF* cloned PstI/BamHI, CmR | This work |
| pT25-FdnH | pT25, T25-FdnH fusion protein, *fdnH* cloned BamHI/KpnI, CmR | This work |
| pT25-Fdx | pT25, T25-Fdx fusion protein, *fdx* cloned BamHI/KpnI, CmR | This work |
| pT25-HycB | pT25, T25-HycB fusion protein, *hycB* cloned PstI/BamHI, CmR | This work |
| pT25-HycF | pT25, T25-HycF fusion protein, *hycF* cloned PstI/BamHI, CmR | This work |
| pT25-HydN | pT25, T25-HydN fusion protein, *hydN* cloned PstI/BamHI, CmR | This work |
| pT25-HyfA | pT25, T25-HyfA fusion protein, *hyfA* cloned KpnI/KpnI, CmR | This work |
| pT25-NarH | pT25, T25-NarH fusion protein, *narH* cloned KpnI/KpnI, CmR | This work |
| pT25-NuoE | pT25, T25-NuoE fusion protein, *nuoE* cloned BamHI/KpnI, CmR | This work |
| pT25-YsaA | pT25, T25-YsaA fusion protein, *ysaA* cloned BamHI/KpnI, CmR | This work |

**Figure S1: Alignment of the HydN group of ferredoxin-like proteins.** The proteins with the given ID’s were aligned with the ClustalO algorithm and visualized with Jalview. Cysteines were highlighted in yellow. Regions where only one protein without cysteines was shown, were omitted from the alignment. The bars under the alignment indicate Conservation, Quality, Consensus and Occupancy of the alignment.