**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\_HindIIIgcg AAGCTT g atgAATCGTTTTATTATGGCT18-aegARW\_EcoRIgcg GAATTC ga GTGAGATTTGACTGATTTTAC | T18C/T25-aegAFW\_BamHIgcg GGATCC c AATCGTTTTATTATGGCCAACT18C/T25-aegARW\_KpnIgcg GGTACC tcaGTGAGATTTGACTGATTTTAC |
| *fdhF* | FdhF\_pUT18FW\_HindIIIgcg AAGCTTg atgAAAAAAGTCGTCACGGTHycB\_pUT18RW\_EcoRIgcg GAATTC ga CGCCAGTGCCGCTTCGCGCA | FdhF\_pT25FW\_PstIgcg CTGCAGgg AAAAAAGTCGTCACGGTTTGFdhF\_pT25RW\_BamHIgcg GGATCC ttaCGCCAGTGCCGCTTCGC |
| *fdnH* | T18-fdnHFW\_HindIIIgcg AAGCTT g atgGCTATGGAAACGCAGGT18-fdnHRW\_EcoRIgcg GAATTC gaCTCATGATGATCCTCCTCGTC | T18C/T25-fdnHFW\_BamHIgcg GGATCC cGCTATGGAAACGCAGGACATT18C/T25-fdnHRW\_KpnIgcg GGTACC ttaCTCATGATGATCCTCC |
| *fdx* | T18-fdxFW\_HindIIIgcg AAGCTT g atgCCAAAGATTGTTATTTTGT18-fdxRW\_EcoRIgcg GAATTC gaATGCTCACGCGCATGGTTG | T18C/T25-fdxFW\_BamHIgcg GGATCC c CCAAAGATTGTTATTTTGCCT18C/T25-fdxRW\_KpnIgcg GGTACC ttaATGCTCACGCGCATGGT |
| *hycB* | HycB\_pUT18FW\_HindIIIgcg AAGCTTg atgAATCGTTTTGTAATTGCTGHycB\_pUT18RW\_EcoRIgcg GAATTC ga TTTAGCCTCTCCACTTTGAG | HycB\_pT25FW\_PstIgcg CTGCAGgg AATCGTTTTGTAATTGCTGACHycB\_pT25RW\_BamHIgcg GGATCC tcaTTTAGCCTCTCCACTTT |
| *hycF* | HycF\_pUT18\_PstIgcg CTGCAGg atgTTTACCTTTATCAAAAAAG HycF\_pUT18\_EcoRIgcg gaattc gaGATGGCCTCTTTCATATGGC | HycF\_pT25FW\_PstIgcg CTGCAGgg TTTACCTTTATCAAAAAAGT HycF\_pT25RW\_BamHIgcg GGATCC tcaGATGGCCTCTTTCATATG |
| *hydN* | HydN\_pUT18FW\_HindIIIgcg AAGCTTg atgAACCGTTTCATCATTGCHydN\_pUT18RW\_EcoRIgcg GAATTCga GAACATCAGCGCCGTACGGC | HydN\_pT25FW\_PstIgcg CTGCAGgg AACCGTTTCATCATTGCTGACHydN\_pUT18CRW\_BamHIgcg GGATCC ttaGAACATCAGCGCCGTAC |
| *hyfA* | HyfA\_T18FW\_KpnIgcg GGTACC atgAACCGCTTTGTGGTGGCHyfA\_T18RW\_EcoRIgcg GAATTC ga GCGTTGCTCCTGAGTGAGGG | HyfA\_PT25FW\_KpnIgcg GGTACC t AACCGCTTTGTGGTGGCCGAHyfA\_pT25RW\_KpnIgcg GGTACC ttaGCGTTGCTCCTGAGTGAG |
| *narH* | T18-narHFW\_HindIIIgcg AAGCTT g atgAAAATTCGTTCACAAGT18-narHRW\_EcoRIgcg GAATTC ga TGGATGCGGCTCCGTTTTGC | T18C/T25-narHFW\_KpnIgcg GGTACC t AAAATTCGTTCACAAGTCGGT18C/T25-narHRW\_KpnIgcg GGTACC tcaTGGATGCGGCTCCGTTTTG |
| *nuoE* | T18-nuoEFW\_HindIIIgcg AAGCTT g atgCACGAGAATCAACAACCT18-nuoERW\_EcoRIgcg GAATTC ga TTTATACCGCTCCAGCAGTTC | T18C/T25-nuoEFW\_BamHIgcg GGATCC c CACGAGAATCAACAACCACAT18C/T25-nuoERW\_KpnIgcg GGTACC tta tcaTTTATACCGCTCCAGCAG |
| *ysaA* | T18-ysaAFW\_HindIIIgcg AAGCTT g atgAACCGGTTTATTATTGCGT18-ysaARW\_EcoRIgcg GAATTC ga AACAGGCTGCTGCCGTAGCC | T18C/T25-ysaAFW\_BamHIgcg GGATCC c AACCGGTTTATTATTGCGGATT18C/T25-ysaARW\_KpnIgcg 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.