Supporting Information

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Fig. S1. Analysis of the protein composition of purified FDH1 by SDS/PAGE. GF, protein solution after the final gel filtration step; $\times 20$, same solution concentrated 20-fold. Band 1, 97.8-kDa subunit (sequences gi|16750131 and 116750132); band 2, 65.8-kDa subunit (gi|116750130); ch, minor contaminant identified as a chaperonin that is highly homologous to GroEL from *Escherichia coli* (gi|16747572); band 3, 16.6-kDa subunit (gi|116750129). Proteins were identified by tandem mass spectrometry, using Mascot (www.matrixscience.com) to search the NCBInr database. The search tolerance was 70 ppm, and the peptides detected typically scored above the individual ion score threshold (41 points, representing P < 0.05). The highest individual ion scores were 140, 93, 133, and 66 for 1, 2, 3, and ch, respectively, and the cumulative ion scores for 1–3 were exceptional. Conditions: 4–12% NuPAGE BisTris polyacrylamide gel with Mops/SDS running buffer (Invitrogen), stained with Coomassie blue.

	MDNNIITLKV	NGQSVRGKKG	QTVLEICREN	GIHIPTLCYH	PKMPPYGGCR	7
[2Fe-2S]	LCIVEIENMR	GLPPSCTTPA	~ VDGMSVHTHT	DKVMGVRKTV	LELLLAYGNH	
? [4Fe-4S]	NCLLCEQTGN	CELQDLVYEH	GIDHVRFK SD	FVPQPLDDAN	F AMIIRDQNKC	FeS
2 [4Fe-4S]	VLCGRCVRGC	LEVQVNGVID	IAMRGSDSFI	TTFDNTELKE	SNCVFCGECV	domain
	ASCPTGALTY	KQARFKGRPW	DLKKVVTTCT	YCGVGCQVEL	NVKDNKVVK T	ון
[4Fe-4S]	TSSFDIPGPN	RGSLCVKGRF	GNDFIDSPDR	LKTPLIKENG	GFR EASWDEA	
	LGLVAK RLGE	IKAK SGPDAV	AFFSSAR CTN	EENYLLNKFA	RAVIGTNNID	
SeCys	HCARLX	VVGLAAAFGS	GAMTNSIEEF	ENTDLILVTG	SNTKEMHPVI	
	SSYMKRGVKK	GKTKLIVVDP	R KTELAEFAE	VWLR QKPGTD	VAWLNGMMNV	
	IIAEGLYDKE	YVANRTEGFE	Elkk avaayt	PERVEEISGI	PK DDLIAAAR	
97 8 kDa	LYAKAPAASI	AYAMGITQHI	NGTEAVKSVA	NLAMLCGNIG	IEGGGVNPLR	W(MGD) ₂
97.0 KDa	GQNNVQGACD	MGALPNVFPA	YQTVTSPEIR	EKFAKAWGVK	DLPAKAGLTI	domain
	VEAVNAASEG	KLKALFVMGE	NPMVSDPDLH	HVKDGLTKLD	FLVVQDIFLT	
	ETAALADVVL	PAACFAEK EG	TFSNTER RVQ	LVRKAVDPPG	DALPDWQIIC	
	GISKRMGYPM	DYPDAEAVFD	EITK VTPSYA	GMDYHR LANG	GLQWPCPTKE	
	HPGTKVLHKD	KFVRGKGLFS	AIEWIPPAES	PDAEYPFMLI	TGRVLYQYHT	
	GTMTRKSIGL	NER YPECLIE	INSR DAANMG	IQDMDDIRLV	TRRGSITAKA	
	KIADVVEKGM	VFVPFHFAEA	AANNLTIAAL	DPLAKIPEYK	VCAVRVEKAA	
	G				-	
		_				
[a]	MQPQASKKKK	PAKERQVLVC	RGTGCESQKA	KILFDNLEHE	lk mlgldddi	
[2Fe-2S]	EVKFTGCHGF	CQQGPTVIVM	PAGTFYCNVQ	PEDADEIVKI	DIKDGGKVER	
	LLFIDPKTKE	RVLSYKDMKF	FSPQRRIVLK	NCGFINPEDI	DDYIAVGGYQ	
	GIQKALGGSR	MDVINEIKKS	GLRGRGGGGF	PTGMKWEFCH	NSPGDQK yli	
65.8 kDa	CNADEGDPGA	FMDR AVLEGD	PHAVLEGMMI	AAYAIGASKA	YIYCR aeypm	
	AYER SMIAID	QATKRGFLGK	KIFGSDMDFE	IKLKLGAGAF	VCGEETALMA	
	SIEGKR GMPM	PRPPFPAVK G	LFGKPTNINN	VETFGNIATI	MTKGGDWFAS	
	VGTEKSKGTK	VFALAGK isy	SGLVEIPMGT	PLREIIDEIG	GGIPNKR KFK	
	AAQTGGPSGG	CIPSEHFDIP	MDYENLTQVG	SIMGSGGLIV	TDETTCMVDM	
[4Fe-4S]	AK FFLGFTQK	ESCGKCVPCR	LGTKKMLEVL	GDISKGKATM	EDLEQLLELA	
2 [4E= 48]	QDVK GGSLCG	LGQTAPNPVL	STVRYFRDEY	EAHILR KECP	ARVCVDLIKF	
2 [4Fe-45]	EVNEENCQKC	GLĊFKAĊPAG	AVSWEKKQTA	KIDVSKĊIKĊ	RSCILACRFN	
	AID					
16.6 kDa	MKDKLQRIFT	RHDRKR DALI	PVLQDIQGEF	GYLPPHAMQA	AAR HCR TSAV	
[2E2 20]	EVYGVSTFYA	QFKFSPVGRH	TVTVĊQGTAĊ	HVMGGHRILE	ECKSQLGVQP	
[266-28]	GQTTPDGMFT	LETVAĊIGAĊ	ALSPAVVVDK	DTYGRMKPER	ITEILNAATS	
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Fig. S2. Sequences of the three subunits of FDH1. The three subunits have protein masses of 97.8, 65.8, and 16.6 kDa. The peptides identified by mass spectrometry are highlighted in bold. Protein domains, based on the homology with proteins of known structure, and cofactor binding motifs are indicated. SeCys, selenocysteine residue; FeS, iron sulfur; W(MGD)₂, tungstopterin cofactor.

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