

Structure of the human Fe-S cluster assembly sub-complex: implications in Friedreich's ataxia and primary metabolism

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Iron-sulfur (Fe-S) clusters are small, inorganic cofactors required for most living organisms.¹ These cofactors play roles in many essential processes including oxidative respiration and DNA replication and repair. In eukaryotes, Fe-S clusters are synthesized and distributed in the mitochondria by a multi-protein complex. A tightly bound sub-complex of this system consists of the cysteine desulfurase, NFS1, and essential eukaryotic specific adaptor proteins ISD11 and acyl-carrier protein (ACP). This sub-complex catalyzes the conversion of cysteine to alanine and persulfide sulfur. This persulfide sulfur can be transferred to the scaffold protein, ISCU2, where in the presence of Fe²⁺ and electrons, a [2Fe-2S] cluster can be formed. Interestingly, the protein associated with Friedrich's ataxia, FXN, has been shown to accelerate cysteine desulfurase activity, sulfur transfer, and Fe-S cluster assembly.¹⁻⁴ In order to elucidate mechanistic details which lead to the activation properties of FXN, we determined 3.09 Å x-ray crystal and 15 Å electron microscopy structures of the sub-complex consisting of human NFS-ISD11 in complex with ACP from *E. coli*. Along with *in vitro* characterization, we also conducted mutagenesis studies on the NFS1-ISD11-ACP sub-complex in *S. cerevisiae* in order to validate interaction surfaces. Overall, this structure reveals a new architecture for cysteine desulfurases that clearly demonstrates the essential nature of ISD11 and ACP in Fe-S cluster biosynthesis and provides a mechanistic hypothesis for FXN based activation. In addition, we also demonstrate that ACP threads its lipid-bound cofactor into ISD11 generating new links among Fe-S cluster biosynthesis, primary metabolism, and mitochondrial fatty-acid synthesis.⁵

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