MS2-P1 **Structural basis of the protein-protein interactions among ISC proteins involved in *de novo* Fe-S clusters biosynthesis.**

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Iron-sulfur (Fe-S) clusters act as cofactors of various Fe-S proteins that are widely distributed in nature and required to maintain fundamental life processes. Recent studies revealed that the assembly of Fe-S clusters in several bacteria as well as eukaryotic mitochondria is achieved by a multicomponent system, called ISC machinery. This machinery is generally encoded, in bacteria, by the *iscSUA-hscBA-fdx* operon, and consists of six ISC proteins. Among them, the components playing central roles in *de novo* Fe-S cluster biogenesis are IscS and IscU. IscS is a cysteine desulfurase that catalyzes the sulfur atom abstraction from cysteine substrate and provides it for biosynthesis. IscU directly accepts the sulfur atoms from IscS and serves as a scaffold for the assembly of a nascent Fe-S cluster, prior to its delivery to target Fe-S protein. We have so far determined the unique trimeric structure of the [2Fe-2S] cluster-bound form of IscU from the hyperthermophilic bacterium *Aquifex aeolicus* (*Aa*) [1]. This structural informa-tion provided mechanistic implications that the dynamic association/dissociation of IscU must be the critical events to interact with the other ISC components in the assembly for the nascent Fe-S clusters. Here we focus on the protein-protein interactions among ISC proteins, to clarify the detailed mechanism of *de novo* Fe-S cluster biogenesis. Coexpression of *Aa* IscS and *Aa* IscU in *E. coli* resulted in formation of a binary complex, and it was purified and crystallized. A data set was collected to 3.6 Å resolution on beamline BL38B1 at SPring-8. The structure of IscS moiety was solved with the MR method using a model of the dimeric *Aa* IscS as the search prove. Although electron densities derived from IscU were mostly invisible, some secondary structures could be assigned. We modeled the structure of IscU moiety in IscS-IscU complex by fitting the previously determined *Aa* IscU into the assigned fragments. The resultant model revealed that two IscU monomers bound near each C-terminus of dimeric IscS where the catalytic pocket of IscS and the Fe-S cluster binding site of IscU were located away from each other. It is likely that the flexible loop conserved among IscS orthologues mediates the transfer of the sulfur source using invariant cysteine residue at the tip of the loop [2]. In addition, our model indicates the possibility that the positively charged area found in the IscS surface probably acts as the binding site of the other ISC protein such as Fdx exhibiting biased negative charge. The cross-linking assay by using EDC, the cross-linker specific for electorostatic interactions showed the covalent bond formation between *Aa* IscS and *Aa* Fdx. These findings allow us to propose the new scheme of Fe-S cluster assembly: IscS serves not only as a sulfur donor but also as a scaffold for the protein-protein interactions in ISC machinery.

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MS2-P2 **Terahertz Absorption change in Photosynthetic Reaction Center upon photoactivation**

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Photosynthetic reaction center is a membrane protein which plays an important role in converting solar energy into chemical energy. Upon continuous illumination a charge separation occurs within the protein which is stable for several seconds [1]. The stability of this state is very important to protect the protein against the release of excess heat and an instable radical due to charge recombination. Absorption of terahertz radiation corresponds to the excitation of large vibrations in the protein and is thus suitable for studying its different dynamical states. Here we recorded terahertz absorption spectra of the dark and illuminated state of reaction centre from the purple membrane bacteria *Rhodobacter sphaeroides* to investigate the stability of the charge separated state. The difference terahertz absorption spectra show an overall increase in absorption for the illuminated state in the 30-130 cm^{-1} spectral region. By simulating and fitting a theoretical terahertz spectrum of reaction center from an elastic network model we were able to investigate the effect of charge separation and conformational changes within the interaction network. We find that a more loose interaction network in the photoactivated state of the protein is the most probable cause of the absorption increase.

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