

vapor diffusion method at 20°C. The crystals of azurin-I belong to the monoclinic crystal system and have space group *C2* with unit cell parameters of $a=130.6\text{Å}$, $b=54.4\text{Å}$, $c=74.7\text{Å}$, and $\beta=96.1^\circ$. Four molecules are in the asymmetric unit. The crystals of azurin-II belong to the tetragonal crystal system and have space group *P4122* with unit cell parameters $a=b=52.6\text{Å}$, $c=100.7\text{Å}$. Only one molecule is in the asymmetric unit. They diffract up to 2.8Å and 2.0Å resolutions, respectively. Their crystal structures were solved by molecular replacement method using AMoRe in the CCP4. Molecular structure of Azurin II from *Alcaligenes xyloxydans* NCIB 11015 was chosen as a starting model for both azurins. The refinement for both azurins is being carried out by XPLOR.

PS02.07.11 A NOVEL IRON CENTER IN DESULFOFERRODOXIN FROM *D. DESULFURICANS* ATCC 27774: CRYSTAL STRUCTURE AT 1.8 Å RESOLUTION. Pedro M. Matias¹, Ana Coelho^{1,2}, Maria A. Carrondo^{1,3}, Vilmos Fülöp⁴, Ana Gonzalez⁵ and Andy Thompson⁶. ¹ITQB, Universidade Nova de Lisboa, 2780 OEIRAS, Portugal; ²Universidade de Évora, 7000 ÉVORA, Portugal; ³IST, Universidade Técnica de Lisboa, 1000 LISBOA, Portugal; ⁴LMB and OCMS, University of Oxford, Oxford OX1 3QU, UK ⁵ESRF, BP-220, 38043 GRENOBLE CEDEX France; ⁶EMBL Grenoble Outstation, BP-156, 38042 GRENOBLE CEDEX France

A novel iron centre in desulfoferrodoxin from sulfate reducing bacteria *D. desulfuricans* ATCC 27774 (DFX) has been structurally characterized: this center contains a single Fe atom octahedrally coordinated to four equatorial histidines, one axial cysteine and one axial water molecule, and unlike most other iron centres in proteins, it is exposed to the solvent, rather than buried within the bulk of the polypeptide chain. The three dimensional structure of DFX contains two domains linked by a short stretch of amino acid residues. The larger domain is associated with the novel Fe centre. The smaller domain contains a desulfoferrodoxin-like Fe-S₄ centre (DX). This presentation will focus on the structural details that can be perceived from the current model and will include a comparison between DX and the DX-like domain of DFX. The crystal structure of DFX has been determined to 2.5 Å by the MAD method using data measured at the ESRF. The current R-factor value is 26.0 % (R_{free} 34.0 %). Refinement against 1.8 Å data measured at SRS-Daresbury is in progress.

PS02.07.12 CRYSTAL STRUCTURES OF CYTOCHROME c' AND ITS n-BUTYL-ISOCYANIDE-BOUND FORM FROM PURPLE PHOTOTROPHIC BACTERIUM. Tahir H. Tahirov, Shintaro Misaki, Yoshiki Higuchi and Noritake Yasuoka, Department of Life Science, Faculty of Science, Himeji Institute of Technology, Hyogo 678-12, Japan and Terry E. Meyer, and Michael A. Cusanovich, Department of Biochemistry, University of Arizona, Tucson, AZ 85721, U.S.A.

The structures of cytochrome c' from *Rhodobacter capsulatus* (RCCP) strain M110 have been determined by the molecular replacement method. Iron anomalous scattering data were also used to confirm the molecular replacement solution. The structures are refined at 1.72 Å and 2.0 Å resolutions to R values of 15.0% and 16.3% respectively. The RCCP molecule is a dimer and each of the identical 129 residue subunits folds as a four-helical bundle with a covalently bound heme group in the center. This structural motif resembles that of cytochromes c' reported from *Rhodospirillum rubrum* (RMCP), *Rhodospirillum rubrum* (RRCP), and *Chromatium vinosum* (CVCP). However, the architecture of the RCCP dimer, that is, the mode of association of subunits, differs substantially from that of the other cytochromes c'.

In the RCCP dimer, the subunits are roughly parallel to each other and only helix B of each subunit participates in formation of the dimer interface. In RMCP, CVCP and RRCP the subunits cross each other to form an X shape, and two helices, A and B, of each subunit are involved in the interactions across the dimer interface. Structural comparison of four cytochromes c' reveal that they can be divided into two groups. In group 1 cytochromes c', CVCP and RRCP, the amino acid sequences and the folding of subunits are arranged in such a way to allow the formation of a large groove between helices B and C with direct solvent accessibility to the heme sixth ligand position. There is no such groove in group 2 cytochromes c', RMCP and RRCP. This may account in part for the differences in carbon monoxide binding.

An X-ray structure analysis of n-butylisocyanide-bound *Rhodobacter capsulatus* cytochrome c' was also carried out at 2.4 Å resolution. A comparison of the ligand bound structure with that of the native protein reveals that a significant conformational change of amino acid residues occurs in the heme vicinity, accompanied by a rearrangement of the hydrogen bonding pattern. The results suggest that heme puckering in cytochrome c' is a unique mechanical mechanism which may control the dissociation of the dimer to monomers upon ligand binding.

PS02.07.13 CRYSTAL STRUCTURES OF CLOSTRIDIUM PASTEURIANUM RUBREDOXIN MUTANTS V8A AND V8N. Tom J. Brett*, John J. Stezowski*, Kathy M. Selbo*, Charles R. Ross II*, Qiangdong Zeng†, Donald M. Kurtz†, and Robert A. Scott†, *Department of Chemistry, University of Nebraska-Lincoln, Lincoln, NE 68588-0304 †Center for Metalloenzyme Studies, University of Georgia, Athens, GA 30602-2556

Crystal structures of V8A and V8N mutants of *Clostridium pasteurianum* rubredoxin (Rd) have been determined at 1.68Å and 1.82Å resolution data giving final R-values of 14.10% and 14.40%, respectively. The structure determinations were pursued to enhance observations from recent studies in which redox potentials of several Rd mutants were determined¹. In these studies, site-directed mutagenesis was used to study the effects of mutations resulting in surface charge changes near the Fe(Cys)₄ site of Rd. As predicted by simple electrostatic considerations, Rd mutants with positively charged arginine residues in place of neutral surface residues (V8R and L41R) exhibit significant increases in the Fe(II/III) reduction potential. However, significant increases in reduction potential were also seen in mutants where the local negative charge was increased (V8D and L41D), and also when neutral residues were replaced by other neutral residues (V8A and V8N). These findings suggest that reduction potential changes are not dominated by electrostatic effects. Rather, it has been proposed that these reduction potential changes are due to increases in the local dielectric at the Fe(Cys)₄ center. Under this hypothesis, increasing the local dielectric, e.g., by increasing the solvent accessibility of the Fe(Cys)₄ site, would increase the reduction potential. In comparison with the Rd wild-type structure, the V8A and V8N mutants show increased solvent accessibility at one of the cysteine sulfurs (S-Cys42).

1 Q. Zeng, E.T. Smith, D.M. Kurtz, Jr., R.A. Scott. *Inorg. Chim. Acta*, in press.