

which has been implicated in catalysis. What was completely unsuspected, however, is that two of the ligands to the zinc come from one monomer and two from the other. Indeed, the whole of the active site is formed only on dimerisation. Each monomer contains two structurally very similar domains. Of the eight strands making up the beta-sandwich, four strands are from the N-terminal domain of one monomer and four from the C-terminal domain of the other. What is most remarkable, however, is that if the N- and C-terminal domains are superimposed, the two zinc ligands of one domain overlap almost exactly with the those of the other domain. This has important implications for the evolution of the enzyme.

MS04.02.06 CRYSTAL STRUCTURE OF THE SELENOCYSTEINE, MOLYBDOPTERIN AND 4Fe4S CLUSTER CONTAINING ENZYME FORMATE DEHYDROGENASE-H. Jeffrey C. Boyington¹, Vadim Gladyshev², Thessa C. Stadtman² and Peter D. Sun¹, ¹Laboratory of Molecular Structure, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Rockville, MD 20852, ²Laboratory of Biochemistry, National Heart, Lung and Blood Institute, National Institutes of Health, Bethesda, MD 20892.

Formate dehydrogenase-H is a key redox component of the formate hydrogen lyase complex of *E. coli*, oxidizing formate to carbon dioxide with the release of two protons and two electrons under anaerobic conditions. This 80 kDa protein contains a 4Fe4S cluster and a molybdenum atom that is coordinated by two molybdoterin guanine dinucleotide (MGD) cofactors and an essential selenocysteine residue. The three-dimensional structure of both the reduced (Mo_{IV}, 4Fe4S_{red}) and oxidized (Mo_{VI}, 4Fe4S_{ox}) forms of formate dehydrogenase-H have been determined by x-ray crystallography using multiple isomorphous replacement and multiwavelength anomalous scattering methods to a resolution of 2.3 Å and an R=21.9% for the reduced form and a resolution of 2.8 Å and an R=20.0% for the oxidized form. Both structures are comprised of four Σ/Σ domains including two MGD cofactor binding domains that are related by a pseudo two-fold rotation axis. Examination of the structure of the oxidized form of the enzyme reveals a 27 degree rotation of one of the MGD cofactors bound to the molybdenum relative to the reduced form with comparatively little movement observed in the rest of the protein. This rotation creates a strained molybdenum coordination geometry in the catalytically active oxidized form so that it is poised to accept the substrate formate as an additional molybdenum ligand. This is the first three-dimensional protein structure of a selenium and molybdenum containing enzyme determined to date. A catalytic mechanism is proposed based on both oxidation states of the enzyme, which incorporates all three of the cofactors revealing a probable electron flow.

MS04.02.07 CRYSTAL STRUCTURE OF DIMETHYLSULFOXIDE REDUCTASE DEFINES A NEW FAMILY OF OXOTRANSFERASES CONTAINING THE MOLYBDENUM COFACTOR. H. Schindelin, C. Kisker, D. C. Rees, Division of Chemistry and Chemical Engineering, 147-75CH, California Institute of Technology, Pasadena, CA 91125, USA

Enzymes containing the molybdenum cofactor (Mo-co) constitute a class of oxotransferases which are widespread in organisms ranging from bacteria, to higher plants and animals, including humans. These enzymes catalyze the transfer of an oxogroup between the substrate and water in a two-electron redox reaction. Dimethylsulfoxide (DMSO) reductase is found in a variety of bacteria, including *Escherichia coli*, *Rhodobacter sphaeroides* and *Rhodobacter capsulatus*. In these organisms, the enzyme serves as a terminal electron acceptor during anaerobic growth in the pres-

ence of DMSO. The crystal structure of DMSO reductase from *Rhodobacter sphaeroides* has been determined in the oxidized, Mo(VI), and reduced, Mo(IV), forms and contains a Mo-co comprising two molybdopterine guanosine dinucleotides as organic components. The fold of the polypeptide chain can be roughly divided into four domains, which, with the exception of the first domain, are all involved in cofactor binding. The second and third domains share structural homology, despite the absence of any sequence homology and each coordinates one guanosine. The fourth domain which mostly interacts with the two pterin moieties, has an unusual topology, since it consists of a six-stranded β -barrel containing parallel as well as anti-parallel β -strands. The two molybdopterine guanosine dinucleotides asymmetrically coordinate the molybdenum with their dithiolene sulfurs. Notably, a serine sidechain oxygen is coordinated to the metal in both oxidation states, while in the reduced enzyme, in addition to the absence of the oxo-ligand present in the oxidized form, one of the dithiolene sulfurs no longer remains coordinated to the molybdenum. Differences in the coordination of the molybdenum between the oxidized and the reduced states provide valuable clues about the reaction mechanism of this enzyme. Sequence comparison of DMSO reductase with enzymes containing a molybdopterine guanine dinucleotide suggests similarities in both the protein architecture and the presence of two molybdopterins, thus defining DMSO reductase as the archetype of this group of bacterial oxotransferases.

PS04.02.08 STRUCTURE OF THE ALDEHYDE OXIDOREDUCTASE FROM DESULFOVIBRIO GIGAS: A MEMBER OF THE XANTHINE OXIDASE PROTEIN FAMILY. Archer, M.¹, Romco, M. J.^{1,2}, Duarte, R.³, Moura, I.³, Moura, J.J.G.³, LeGall, J.⁴, Hof, P.⁵, Huber, R.⁵, ITQB, Apt 127, 2780 Oeiras, Portugal, ²IST, 1000 Lisboa, Portugal, ³FCT-UNL, 2785 Monte Caparica, Portugal, ⁴University of Georgia, Athens, GA 30602, USA, ⁵MPI, D-82152 Martinsried, Germany

The crystal structure of the Aldehyde Oxidoreductase (MOP) has been solved by the MIR method at 2.25 Å resolution and refined to an Rfactor of 16.5% [1]. MOP, isolated from the sulfate reducing bacterium *Desulfovibrio gigas* is a homodimer of 907 aminoacid residues per subunit. It contains a molybdopterine cofactor (mo-co) and two different [2Fe-2S] centers and is folded into four domains. The first two bind the two iron sulfur clusters and the others are associated with Mo-co. Mo-co is a molybdopterine cytosine dinucleotide and is deeply buried in the protein and accessible through a 15 Å deep tunnel. The Mo is pentacoordinated by two dithiolene sulfur atoms of the molybdopterine and three oxygen ligands. MOP oxidizes aldehydes to carboxylic acids. It belongs to an electron transfer chain, consisting of four proteins from *D.gigas* - flavodoxin, cytochrome c3 and hydrogenase - so that the oxidation of aldehydes may be linked to the generation of hydrogen. MOP sequence reveals homology to the xanthine oxidases. In xanthine oxidases (ca 1300 residues) the electron transfer occurs from the Mo site to a flavin group, which is absent in MOP. The crystal analysis of MOP shows the structure of Mo-co in detail and allows a first look at a member of the xanthine oxidase family. A structure-based catalytic mechanism for the xanthine oxidase family of molybdenum enzymes is proposed.

1. Romco, M. J., Archer, M., Moura, I., Moura, J. J. G., LeGall, J., Engh, R., Schneider, M., Hof, P., Huber, R., Science 270, 1170 (1995).