s8a.m7.p11 Crystal Structure of L-Chain Mouse Ferritin at 1.2 Å resolution. T. Granier[1], B. Langlois d'Estaintot[1], J. M. Chevalier[1],J. M. Mellado[1], M. Béhuré[1], B. Gallois[1], P. Santambrogio[2], P. Arosio[2], G. Précigoux[1]. [1] *Unité de Biophysique Structurale, Université Bordeaux I, Bât. B8, Avenue des Facultés, 33405 Talence, France*. [2] *Dibit, Scientific Institute H. San Raffaele, Via Olgettina 58, 20132 Milano, Italy*.
Keywords: protein, iron metabolism, high resolution.

Ferritin is the ubiquitous iron storage protein, utilized by most living organisms to reversibly store and release iron ions. At present, more than 50 sequences of ferritins are fully or partially known. Despite a sequence homology sometimes lower than 20%, these proteins show an invariance in their secondary, tertiary and quaternary structures. Apoferritin molecules are made of 24 subunits, each of which consists of a four-helix bundle (A-D) and a fifth short helix (E) at the C-terminus. The 24 bundles pack in a 432 point symmetry and form a hollow shell, whose inner cavity can host a ferrihydrite mineral core composed of up to 4500 iron ions (for a review, see references 1 & 2).

The structures of several eukaryotic ferritins have been already published. Nevertheless, none of these have been determined at a resolution better than 1.9 Å. Recently, looking at the crystal structure of L-chain mouse ferritin (182 a.a per subunit), we have collected data at LURE (beam line DW32) under cryogenic conditions (150K) at a resolution of 1.2 Å. The data set characteristics are: completeness ≈ 98%, multiplicity = 5.1, R_{sym}=0.06, $I/\sigma(I)$ ≈ 2.1 in the last resolution shell.

Refinement of the structure is in progress. Results will be discussed and compared with those previously obtained at room temperature (2.1 Å resolution). High resolution structural details will be highlighted in relation to the biological functions of the protein.

[1] Harrison, P.M & Arosio, P. (1996) Biochim. Biophys. Acta, 1275, 161 – 203.
[2] Chasteen, N.D. & Harrison, P.M. (1999) J. Struct. Biol., 126, 182 – 194.

s8a.m7.p12 [NiFe] Hydrogenase from *Desulfovibrio desulfuricans* ATCC 27774: Gene sequencing, three-dimensional structure determination and refinement at 1.8 Å and modelling studies of its interaction with the tetra-haem cytochrome c_3. P.M. Matias[1], C.M. Soares[1], L.M. Saraiva[1], R. Coelho[1], J. Morais[1], J. Le Gall[1,2] and M.A. Carrondo[1]. [1]*Instituto de Tecnologia Química e Biológica, Universidade Nova de Lisboa, Apartado 127, 2781-901 Oeiras, Portugal;* [2]*Department of Biochemistry and Molecular Biology, University of Georgia, Athens, GA 30602 USA.*
Keywords: hydrogenase crystal structure, molecular modelling, electron transfer.

The primary and three-dimensional structures of a [NiFe] hydrogenase isolated from *D. desulfuricans* ATCC 27774 were determined, by nucleotide analysis and single crystal X-ray crystallography. The three-dimensional structural model was refined to R=0.167 and R-free=0.223 using data to 1.8 Å resolution. The three-dimensional structure of this [NiFe] hydrogenase shows two unique structural features features which may have biological relevance in terms of the enzyme activation process: the [4Fe-4S] cluster nearest the [NiFe] centre has been modified to [4Fe-3S-3O] by loss of one sulphur atom and inclusion of three oxygen atoms; a three-fold disorder was observed for Cys 536 which binds to the nickel atom in the [NiFe] centre. In addition, the bridging sulphur atom that caps the active site in the [NiFe] centre was found to have partial occupancy, thus corresponding to a partly activated enyme. Putative locations for the H_2S molecules produced as the result of the modification of the proximal [4Fe-4S] cluster and the partial un-capping of the active site were also located in the three-dimensional structure.

Molecular modelling studies of the interaction between this [NiFe] hydrogenase from *D. desulfuricans* ATCC 27774 and its physiological partner, the tetra-haem cytochrome c_3 from the same organism were carried out. The lowest energy docking solutions were found to correspond to an interaction between the haem IV region in tetra-haem cytochrome c_3 with the distal [4Fe-4S] cluster in [NiFe] hydrogenase. This interaction should correspond to efficient electron transfer and be physiologically relevant, given the proximity of the two redox centres and the fact that electron transfer decay coupling calculations show high coupling values and a short electron transfer pathway. On the other hand, other docking solutions have been found that, despite showing low electron transfer efficiency, may give clues on possible proton transfer mechanisms between the two molecules.