s8a.m1.p5 **Structure and mechanism of pteridine reductase 1 from *Leishmania*.** A. Schüttelkopf, D.G. Gourley and W.N. Hunter. *Department of Biochemistry, University of Dundee, MSI/WTB Complex, Dow St., DUNDEE, DD1 5EH, Scotland.*
Keywords: trypanosomatids, pteridine reductase, drug resistance.

Pteridine reductase 1 (PTR1) is a novel member of the short-chain dehydrogenase/reductase (SDR) family found in trypanosomatids, where it effects the NADPH-dependent two-step reduction of a wide variety of pterin compounds from the oxidized (e.g. biopterin) to the fully reduced (e.g. tetrahydrobiopterin) form. Trypanosomatids are pterin auxotrophs, therefore they rely on the uptake of oxidized pterins to satisfy their pterin/folate requirements, and PTR1 constitutes a validated target for drugs against these protozoan parasites. In addition PTR1 has been implied in antifolate resistance in these organisms.

PTR1 from *Leishmania major* has been crystallized in complex with cofactor and a number of substrates/inhibitors in two orthorhombic crystal forms with a=80Å, b=81Å, c=90Å (space group $P2_12_12$) and a=94Å, b=104Å, c=137Å (space group $P2_12_12_1$). The structures provide insight into the catalytic mechanism, which, for the second reduction step, differs significantly from the established generic SDR mechanism. Together, the substrate/ inhibitor structures demonstrate various substrate binding modes in the active site. This information is currently used in the computer-aided design of potential high-specificity inhibitors for PTR1.

s8a.m1.p6 **Crystal structure of an 'half-and-half' three-fingers chimeric protein.** M.H. le Du[1#], A. Ricciardi[2], M. Khayati[1], R. Ménez[1], J.C. Boulain[1], A. Ménez[1], and F. Ducancel[1#]. *[1] : Département d'Ingénierie et d'Etude des Protéines, Commissariat l'Energie Atomique, CE Saclay, 91191 Gif-sur-Yvette Cedex, France. [2] : Instituto de Investigaciones Biologicas, Clemente Estable, Montevideo, Uruguay 11600.*
Keywords: chimeric protein, three-fingers, structural scaffold.

Similarly to antibodies, three-fingers proteins form a structurally related family of compounds which exert a large variety of different function. To address the question of the prediction of functional areas onto the surface of three-fingers snake toxins, we confered the acetylcholinesterase inhibitory activity of fasciculins to the short-chain curaremimetic toxin α. Their relative functional regions belong to loop I and II, but the superimposition of the two structures suggests that these functional regions also correspond to structurally distinct zones. Loop I, half of loop II and the C-terminal residue of fasciculin 2 were therefore transferred into the toxin α. The resulting chimera has an inhibition constant which is only 15-fold lower than that of fasciculin 2, and the potency of binding to the toxin α target has been lost. In order to understand the structure-function relationship between the chimera and its 'parent' molecules, we solved its structure by X-ray crystallography. The protein crystallized in space group $P3_121$ with a=b=58.5 Å, and c=62.3 Å. The crystal structure was solved by molecular replacement and refined to 2.1 Å resolution. The superimposition of the model of the chimera on fasciculin 2 or toxin α revealed an overall fold intermediate between those of the two parent molecules. The regions corresponding to toxin α and to fasciculin 2 retained their respective geometries. In addition, the chimera protein displayed a structural behavior similar to that of fasciculin 2 : dimerization in the crystal structure of fasciculin 2 ; the geometry of the region that binds to acetylcholinesterase. In conclusion, this structure shows that the chimera retains the structural characteristics of toxin α, and the structural specificity of the transferred function from fasciculin.