MS12-03 **Mapping conformational changes of the transcription factor AraR** C.S. Silva[1], D. de Sanctis[2], M. I. Correia[3], M. Petoukhov[4], D. Svergun[4], P.F. Lindley[1], I. de Sá Nogueira[3] and I. Bento[1] *[1]Instituto de Tecnologia Quýmica e Biologica, Universidade Nova de Lisboa, Avenida de Republica, EAN, Oeiras, Portugal; [2]Structural Biology Group, European Synchrotron Radiation Facility, Grenoble, France; [3]Departamento de Ciencias da Vida, Faculdade de Ciencias e Tecnologia, FCT, Universidade Nova de Lisboa, Caparica, Portugal; [4]Biological Small Angle Scattering Group, European Molecular Biology Laboratory, Hamburg.*

AraR is a transcription factor that regulates the utilization of carbohydrates in *Bacillus subtillis* by controlling the expression of at least 13 genes (Franco *et.al.* 2006, Franco *et. al.* 2007). It belongs to the GntR family of regulators and typifies a sub-family within this family, the AraR sub-family. All members of the family show a chimeric organization of two domains: the N-terminal DNA binding domain and a C-terminal effector binding domain. Whereas the N-terminal domain is common to all members of this family, showing a helix-turn-helix (HTH) consensus pattern and a highly conserved topology, the C-terminal effector binding domain shows heterogeneity allowing the sub-division of this family into six sub-families (Hoskisson *et. al.* 2009). In particular, the AraR sub-family shows a C-terminal domain which is homologous to the LacI/GalR family of regulators (Hoskisson *et. al.* 2009). The activation of these regulators, which are functional as dimers, is modulated by the presence and binding of the effector molecule that in the case of AraR is arabinose (Hoskisson *et. al.* 2009). Upon binding the effector, the regulator undergoes a conformational change that prevents the binding of the N-terminal domain to the DNA and thus inhibits gene transcription. Recently, and concurrently with our study, Procházková *et al.* (2012) have determined the crystal structure of a truncated form of the AraR protein, corresponding to the C-terminal effector binding-domain in complex with L-arabinose. In the present work we take a step further and determined the crystal structure of the full-length protein in complex with the effector L-arabinose, which together with SAXS data obtained from the complex in solution, allowed the identification of the conformational changes that underlie the AraR allosteric regulation.

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MS12-04 **Structure of the Hsp47-Collagen Complex.** Ulrich Baumann,[a] Christine Widmer,[b] Jan Gebauer [a]
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Collagen is the most abundant protein in animals and a major component of the extracellular matrix in tissues like skin and bone. Distinctive structural feature of all 28 known collagen types is a unique triple-helical structure formed by tandem repeats of the consensus sequence Xaa-Yaa-Gly, where Xaa and Yaa are frequently proline and hydroxyproline, respectively [1, 2]. Collagen folding and maturation involves interactions with many modifying enzymes and chaperones in the ER. Hsp47/SerpinH1 is a procollagen-specific molecular chaperone belonging to the Serine protease inhibitor family (Serpins) [3]. Contrary to other known chaperones Hsp47 specifically recognizes the folded conformation of its client and is absolutely essential for proper triple helix formation and procollagen maturation. Reduced functional levels of Hsp47 were reported in severe recessive forms of *osteogenesis imperfecta* and homozygous knockout is lethal in mice [4-6]. The mechanism of Hsp47 action is unclear due to a lack of structural information. Here we present crystal structures of Hsp47 in its free form and in complex with homotrimeric synthetic collagen model peptides, each strand comprising one Hsp47 binding site represented by an arginine at the Yaa-position of a Pro-Arg-Gly triplet. Two of these three binding sites in the triple helix are occupied by Hsp47 molecules, which bind in a head-to-head fashion transversally to the collagen triple helix, recognizing the arginine side-chain via a conserved aspartic acid located in sheet C. There are no significant conformational changes upon binding and the characteristic Serpin reactive center loop appears not to be involved in binding. Each Hsp47 molecule makes extensive contacts to at least 2 of the 3 collagen strands, thus binding to the folded structure of the collagen triple helix. The structures explain Hsp47's stabilization of the triple helix as well as the inhibition of collagen bundle formation. In addition, we propose a pH-dependent client release mechanism based on a cluster of histidine residues.

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