MS10-P5 **Auto-inhibitory assembly of protein kinase CK2 revealed by X-ray crystallography.** Graziano Lolli,[ab] Lorenzo A. Pinna,[c] Roberto Battistutta, [ab] [a]*Department of Chemical Sciences, University of Padua, Padova, Italy,* [b]*Venetian Institute for Molecular Medicine (VIMM), Padova, Italy,* [c]*Department of Biological Chemistry, University of Padua, Padova, Italy*
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The enzyme CK2 is a pleiotropic, acidophilic, highly conserved Ser/Thr protein kinase essential for cell viability, involved in many cellular processes such as cell cycle progression, gene expression, cell growth and differentiation, embryogenesis, circadian rhythms, and apoptosis. Abnormal high levels of CK2 enzymatic activity have been found in a large variety of solid and hematological tumors. Its oncogenic potential depends mainly on the capacity to act as an antiapoptotic agent. In vivo, the fully functional form of CK2 is considered the α2β2 holoenzyme, a heterocomplex of 140 kDa composed of two catalytic α-subunits and two regulatory β-subunits. This enzyme is not regulated by mechanisms common to other protein kinases, and how its activity is controlled is still unclear. We solved the structure of the tetrameric holoenzyme updating some features that were previously poor defined (PDB: 4DGL). We show that the holoenzyme can self-assemble in trimers of tetramers and subsequently in filaments postulating an auto-inhibitory model for CK2 regulation [1], which is in accordance with a plethora of biochemical and biophysical data available in literature. The core of the trimeric organization is constituted by the electrostatic interaction between the acidic loop of the β-subunit (Asp55-Asp64) and the P+1 loop of the α-subunit (Arg191-Lys198) of another tetramer (acidic/P+1 interaction). This ring organization is then in accordance with the well documented existence of the electrostatic acidic/P+1 interaction and with its dependence on salt concentration [2]. The α subunit in contact with the β acidic loop can simultaneously accommodate the C- terminus of the β subunit from a third neighboring tetramer in its ATP-binding site. The β C-terminus competes with ATP for the binding to the α subunit. Further, once inserted in the ATP-binding pocket, it stabilizes a nonproductive conformation of residues involved in catalysis. β-Arg215 replaces α-Lys68 in its interaction with α-Glu81 from the αC helix (the Lys68-Glu81 ion-pair is essential for an efficient catalysis and is fully conserved in protein kinases), while β-Lys212 interacts with α-Asp175, which is not more available for Mg2+ binding as in the fully functional state. This new interaction is fundamental for a piling organization of trimers, with each trimer rotated of 60° with respect to the preceding one, to give a final hexagonal geometry along the piling axes. In this filamentous organization, half of the α-subunits are inhibited by the presence of the C-terminus and the acidic loop of the β subunit in the α active site, while the remaining half are inhibited by steric hindrance. This polymeric form is therefore expected to be completely inactive and can well represent the filamentous CK2 aggregates described in literature [3-4].

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MS10-P6 **Crystal structure of Staphylococcal enterotoxin B in complex with two human immunoreceptors.** Karin Rödström, Karin Lindkvist-Petersson, *Department of experimental Medical Science, Lund University, Sweden*
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Superantigens are toxins capable of triggering an extreme immune response. They are produced by bacteria, e.g. Staphylococcus aureus and Streptococcys pyogenes, and are involved in causing a variety of diseases. These include food poisoning, toxic shock syndrome and scarlet fever, and a role in autoimmune diseases has also been suggested. Superantigens act by cross-linking receptors in the immune system, more precisely the major histocompatibility complex (MHC) class II and the T cell receptor (TCR) [1]. We have determined the X-ray structure between staphylococcal enterotoxin B (SEB), a superantigen, with bound receptors, MHC class II and TCR to a resolution of 2.9 Å. Judging from the structure, it is evident that the purpose of SEB is to bind in a wedge-like fashion between MHC and TCR, preventing the variable β-domain of TCR (TRBV) to reach the peptide. Three distinct interfaces are formed in the structure, the SEB-MHC interface, the SEB-TRBV interface, and an additional one between MHC and the variable á-domain of TCR (TRAV). This interface between MHC and TRAV is smaller than in an ordinary TCR-MHC complex, and the variable loops of TRAV are displaced and the CDR3 loop is not able to contact the peptide. Staphylococcal enterotoxin B has previously been crystallized in complex with a murine TCR β-chain [2], and together with this structure, it is possible to get a more detailed knowledge of the TRBV preference of SEB. This work is a step towards understanding the complex nature of superantigens, and also to understand what governs protein-protein interactions.

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