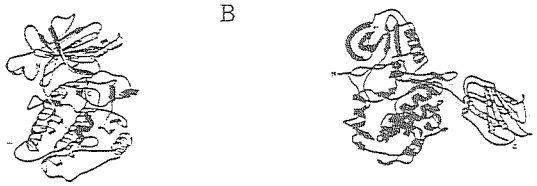
twitchin (B) containing the autoinhibited kinase domain and a C-terminal Ig-like domain shows that the Ig-like domain extends from the kinase domain opposite to the active site and exposes possible myosin interacting surfaces. Our studies of the regulation and domain architecture of giant protein kinases jointly with an analysis of autoinhibitory sequences of other autoinhibited kinases point to possible common and diverse features of the intrasteric regulatory mechanisms.

A B



MS04.10.07 CRYSTAL STRUCTURE OF A HISTIDINE KINASE DOMAIN OF THE ANAEROBIC SENSOR PROTEIN ARCB FROM *E. COLI*. Toshio Hakoshima[1], Masato Kato[1], Toshiyuki Shimizu[1], Kazuya Ishige[2], and Takeshi Mizuno[2]. [1]Department of Molecular Biology, Nara Institute of Science and Technology (NAIST), 8916-5 Takayama, Ikoma, Nara 630-01, Japan and; [2]School of Agriculture, Nagoya University, Chikusa-ku, Nagoya 464, Japan

The *Escherichia coli* ArcB protein is the anaerobic sensor and a member of the large family of the so-called two-component signal transduction proteins. ArcB comprising 778 amino acid residues possesses three phosphorylation sites (His-292, Asp-576, His-717). These multi-phosphorylation sites were suggested to make a phosphotransfer circuit that is crucial for the mechanism underlying signal transduction through ArcB. The C-terminal histidine kinase domain is composed of 125 amino acid residues, in which the active His-717 is located. Here, three-dimensional structure of this domain has been determined at 2.06Å resolution. The crystal belong to the space group $P2_12_12_1$ with unit cell dimensions, a=30.56Å, b=34.93Å, c=1 10.78Å. The structure was solved by MIR methods and was refined. The current model has an R-factor of 23%. The kinase domain of ArcB is found to form an all-α structure consisting of six helices (H1 to H6); 75% of its residues locates on the α helices. The molecules has a kidney-like shape with dimensions, 30Å by 30Å by 45Å. Two helices H3 and H6 are 7- and 8-turns long, respectively, and are bent at the centers of the helices. The helices H4 and H5 form a four-helix bundle subdomain together with the C-terminal half of the helix H3 and the N-terminal half of the helix H6. The active residue, His-64 (corresponding to His-717 in the intact ArcB), locates at the surface of the helix H4 and is surrounded with Glu61, Lys-65 and Lys-67 from the same helix and with Gln-83 and Gln-86 from the helix H5. This active site lies in the internal curvature of the kidney-shaped molecule. The helices H1 and H2 form another subdomain with the N-terminal half of the helix H3 and the C-terminal half of the helix H6.

MS04.10.08 STRUCTURE OF THE PHOSPHORYLATED FORM OF THE CDK2:CYCLIN-A COMPLEX. Philip D. Jeffrey, Alicia A. Russo, Nikola P. Pavletich, Memorial Sloan-Kettering Cancer Center, 1275 York Avenue, New York, NY 10021, USA

Progression through the eukaryotic cell cycle is controlled by the cyclin-dependent kinases (CDKs), which are in turn under the control of multiple levels of regulation. The isolated CDK is inactive, becoming partially activated upon binding of the corresponding cyclin subunit. Full activation of the kinase complex is achieved upon phosphorylation at a site on the CDK subunit (Thr 160 in CDK2).

We have previously determined the structure of a CDK2:CyclinA complex in its unphosphorylated state at 2.3 Angstrom resolution (ref. 1). Comparison of the CDK2 subunit with the structure of the uncomplexed CDK2 (ref. 2) revealed extensive conformational changes in two regions (the PSTAIRE helix and activation loop). In contrast, comparison of the cyclinA subunit with the uncomplexed cyclinA structure (ref. 3) did not indicate any conformational change on complex formation.

Subsequently, we have crystallized the fully active complex containing CDK2 that was phosphorylated at Thr 160. The structure of this complex reveals further conformational changes caused by phosphorylation. The contribution of these changes to the full activation of the kinase will be discussed.

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MS04.10.09 HORMONE BINDING TO THE THYROID HORMONE RECEPTOR. R. Fletterick, R. Wagner, J. Baxter, A. Shiau, J. Apriletti. University of California, San Francisco, Dept. of Biochemistry/Biophysics, San Francisco, CA 94143-0448, USA

This receptor is a central metabolic regulator and is a member of the family of nuclear receptors that regulate transcription of responsive genes. The structures of two isoforms α and β were determined to understand the mechanism of recognition of hormone and the changing relationship of the receptor to its receptor, repressor and activator partners. The ligand binding domain (LBD) of the receptor comprises 12 helices and employs the hormone as a component of the hydrophobic core of the domain. The structures of the LBD with hormone and hormone analogs suggest means for precise recognition. The LBD can form dimers with homologs of the receptor or homodimers. The structures of two different homodimers have been determined to learn whether the means of association is used in transcriptional control.

PS04.10.10 CRYSTAL STRUCTURE OF mTNF. K.J. Baeyens, P. Brouckaert*, A. Raeymaekers*, W. Fiers*, C.J. De Ranter & H.L. De Bondt, Laboratory for Analytical Chemistry and Medicinal Physicochemistry, K.U.Leuven, Leuven, Belgium; *Laboratory of Molecular Biology, VIB and U. of Ghent, Belgium

TNF is a trimer consisting of 17 kDa subunits. The 3D-structure of human (h) TNF at 2.6 Å resolution has been reported. TNF interacts with two types of receptor, TNF-R55 and TNF-R75. Cellular signaling occurs by triggering one or the other, or both receptors, depending on the cell type and conditions. hTNF in the mouse only interacts with TNF-R55, and hence is a specific ligand for the latter. Remarkably, in the normal mouse, hTNF is 50-fold less toxic as compared to murine (m) TNF, indicating that the