

LECTURE #7: PROTEIN KINASES

1. General considerations
 - The protein kinase super-family
 - Conserved primary structure and core catalytic domain
 - Examples of differing modes of regulation
 - cAMP
 - Lipids
 - Small GTPases
 - Ca²⁺/calmodulin

2. Kinetic analysis of phosphotransferase reaction mechanism
 - Role of Mg²⁺-ATP complex as phosphate donor
 - Phosphoacceptors (substrates)
 - Substrate recognition and selectivity
 - Use of synthetic peptides
 - Use of generic protein substrates
 - Identification of authentic *in vivo* targets
 - Phosphotransfer reaction mechanism

3. Structural basis of regulation
 - Activation loop phosphorylation
 - Use of phospho-specific antibodies
 - Regulation of activity
 - Dissociation of a negative regulatory subunit (e.g. PKA)
 - Displacement of a pseudosubstrate motif (e.g. PKC and CaMK II)
 - Displacement of a negative regulatory domain (e.g. Src)
 - Association with a positive regulatory factor (e.g. Cdks)
 - Phosphorylation-induced conformational change (e.g. MAPKs)
 - Activation via proteolytic cleavage (e.g. MEKK-1)
 - Regulation of localization
 - Tethering to membrane-associated factors or lipids (e.g. PAKs, PDK-1, c-Akt/PKB)
 - Translocation to the nucleus (e.g. PKA)
 - Binding to scaffolding proteins (e.g. Ste5, JIPs, AKAPs)

4. Structural basis for substrate selectivity
 - Ser-/Thr-directed versus Tyr-directed protein kinases
 - Dual-specificity protein kinases
 - Non-canonical protein kinases (TOR, DNA-PK_{cs}, ATM, WNK1, Vps15, ILK, etc.)