interactions. NMR shows that there is a reduction in the backbone mobility on the regions of the PHD that participate in the peptide binding, and binding affinities differ depending on histone tail lengths. Thermodynamic analysis reveals that the discrimination in favor of methylated lysine is entropy driven, contrary to what has been described for chromodomains. The molecular basis of H3K4me3 recognition by ING4 differs from that of ING2, which is consistent with their different affinities for methylated histone tails. These differences suggest a distinct role in transcriptional regulation for these two ING family members due to the antagonistic effect of the complexes that they recruit onto chromatin. Our results illustrate the versatility of PHD fingers as readers of the histone code.

Keywords: ING, inhibitor of growth, PHD, plant homeodomain, histone 3

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A new type of precipitant, metal cyanide complex

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Transition metal salts, such as Fe, Ni, Co, and Cr salt, are generally avoided in protein crystallization because of fear of protein inactivation or denaturation. However, if transition metal is incorporated in crystal structure, they are useful for SAD, or MAD phasing. Here we cast spotlight on metal cyanide complex salts as protein precipitant, because metal cyanide complex has relatively low affinity for protein and chemically stable over a wide range of pH. We tested five metal cyanide complex salts, $K_3Cr(CN)_6$, $K_3Fe(CN)_6$, $K_4Fe(CN)_6$, $K_3Co(CN)_6$, and $K_2Ni(CN)_4$. Three proteins, lysozyme, proteinase K, and trypsin were tried to crystallize using each salt as a precipitant by batch method. No additional compound added to mother liquid without buffer component (from pH 4 to pH 9). Diffraction data from grown crystals were collected using CuKα radiation. Metal sites were located using anomalous signal by the program *SHELXD*. Phase calculation was performed by SAD method using program *MLPHARE* and automatic model-building by *ARP/wARP* were performed. All tested protein can be crystallized by metal cyanide complexes. The crystal form was isomorphous to that grown with non-metal precipitant except in the case of lysozyme with $K_4Fe(CN)_6$, where we obtained a new crystal form. In the crystal, metal complexes bind to positively-charged surfaces of the protein. In the case of Cr, Fe, and Co salt, location of metal sites, phasing by SAD method, and automatic model-building were accomplished easily and successfully. But, in the case of Ni salt, location of metal sites was difficult, and phasing power on SAD phasing is as low as that of native (non-metal) crystal because of low f" value of Ni at the wavelength of CuKα radiation.

Keywords: new type of precipitant, transition metal complex, SAD method

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Crystallization and crystallographic analysis in a microfluidic chip

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Microfluidic technology opened new possibilities for the crystallization of biological macromolecules. Indeed, microfluidic systems offer a lot of advanges for crystal growth: they enable easy handling of nano-volumes of solutions and, thus, extreme miniaturization and parallelization of crystallization assays. In addition they provide a convection-less environment a priori favorable to the growth of high quality crystals. Pioneer examples implementing free interface diffusion [1] and nano-batch [2] crystallization in microfluidic chips have already demonstrated the value of this technology, especially for high throughput screening applications in structural genomics. We will present the results of a collaborative work initiated in 2004, that is focused on the design of a versatile, low cost and easy-to-use crystallization chip. A novel chip based on the counter-diffusion of solute molecules playing the role of crystallization agents will be described [3]. The chip is made of rigid polymers (e.g. PMMA, COC) that are impermeable to gases and compatible with crystal examination and monitoring in polarized light. Selected materials are also transparent to X-rays, and three-dimensional protein structures can be determined from crystals contained inside this device (image on the left) using X-ray diffraction data collected on a synchrotron source (middle). The outstanding quality of the electron-density maps (right) demonstrates that on-chip crystal analysis is feasible.

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Keywords: biomacromolecule crystallization, microsystems, *in situ* diffraction

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Improving protein crystallization: A large-scale evaluation of protein reductive methylation

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Obtaining diffraction quality crystals is a major bottleneck in protein X-ray crystallography. Chemical modification of proteins can alter their surface properties and crystallization behavior and influence crystallization success. We evaluated how the methylation of lysine residues in proteins may improve crystallization on a set of 370 unique proteins (<30% sequence identity) that failed to produce diffraction quality crystals. Applying well-established methods, the proteins were methylated and screened using standard crystallization procedures. Crystal structures of 26 new proteins were determined, four in both native and methylated states. Crystals of