

Phase diagram of nucleosome core particles

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The compaction of chromatin in the nucleus is highly dynamic and subject to reversible changes in higher order folding and nucleosome positioning. The crystallographic structure of the nucleosome core particle has been resolved at high resolution (^{1,2}), but we seriously lack structural data about the multiple local changes of chromatin organization that occur locally inside the living cell. To overcome the difficulty of analysing the structural details of chromatin organization, we chose to use a simplified experimental model consisting of isolated nucleosome core particles, prepared under conditions (salt and particle concentrations) that are biologically relevant (from about 100 to 500 mg/ml).

By controlling the ionic conditions (3 to 160 mM monovalent salt, no divalent ions) and the applied osmotic pressure, multiple dense phases of nucleosomes were obtained. We present here an X-ray diffraction study coupled with cryo-electron microscopy of the different phases observed. For low salt concentration (below 25 mM), a lamello-columnar phase is found: NCP stack into columns that align to form bilayers, kept separated from one another by a layer of solvent. NCPs form a monoclinic lattice in the plane of the bilayer. For higher salt concentration (above 50mM), NCPs order into either a 2D columnar hexagonal phase or 3D orthorhombic (quasi-hexagonal) crystals (³). The lamellar and hexagonal (or quasi-hexagonal) organizations coexist in the intermediate salt range; their demixing requires a long time. An isotropic (and in some cases, a nematic) phase of columns is observed in the moderate concentration range. The phase diagram will be presented (⁴). The relevance of these phases in the biological context will be discussed.

¹ Lüger K., Mäder A.W., Richmond R.K., Sargent D.F., Richmond T.J. (1997) Crystal structure of the nucleosome core particle at 2.8Å resolution. *Nature* **389**, 251-260.

² Davey C.A., Sargent D.F., Luger K., Maeder A.W., Richmond T.J. (2002) Solvent mediated interactions in the structure of the nucleosome core particle at 1.9Å resolution. *J. Mol. Biol.* **319**, 1097-1113.

³ Mangenot S., Leforestier A., Durand D., Livolant F. (2003) *Biophys. J.* **84**, 2570-2584.

⁴ Mangenot S., Leforestier A., Durand D., Livolant F. (2003) Phase diagram of nucleosome core particles. *J. Mol. Biol.* **333**, 907-916.