How do histone modifications and histone variants affect nucleosome structure and dynamics? Single-molecule fluorescence studies

Doctoral or Master's thesis project in the Division Biophysics of Macromolecules

Project description

Covalent modifications of histones play a central role in determining gene expression in normal and malignant states of the cell. We are interested in their molecular basis, i.e., how histone modifications affect nucleosome structure and thus the global packing of the chromatin fiber and DNA accessibility.

Single molecule fluorescence spectroscopy, pioneered by W.E. Moerner (Nobel prize in chemistry 2014) is an ideal tool to study the structure and dynamics of isolated nucleosomes (1-3). By detecting signals from fluorescently labeled single biomolecules, transitions between structural states (e.g., DNA unwrapping, histone dissociation, nucleosome repositioning) can be observed directly in real time. Recently, we have discovered new structural substates during nucleosome opening, giving new insight into the mechanism of chromatin access (1,2). The formation and stability of these substates is strongly affected by histone modifications, such as lysine acetylation (4,5).

A particular interesting recent finding is the central role of two H2A arginines at the H2A/H3 interface in the overall stability of the nucleosome. Computer simulations predicted that these residues take part in a structural transition induced by clipping histone N-terminal tails (6). Mutating these residues strongly destabilizes the nucleosome, inducing a more open structure.

Using single molecule FRET and other biophysical methods established in our group, you will study the effect of modifications in the histone tails on the structure and dynamics of nucleosomes. Currently our focus of interest is in the role of the arginine mutations in nucleosome opening and the dynamics of histone tails. This project will be centered on studying the dynamics of the nucleosome interior and the histone tails using single-molecule FRET. You will prepare defined nucleosome samples, apply and develop procedures for histone modification and fluorescent labeling of DNA and protein. You will then take single molecule fluorescence data from these samples and analyze them to characterize the structure of nucleosomes in various modification states. The experiments will be closely connected to molecular dynamics simulations going on in our group (7).

Students interested in developing optical equipment will also have the opportunity to implement new techniques for single molecule detection and data analysis.

Desired qualifications

Our group is highly interdisciplinary, comprising biologists, physicists, chemists and mathematicians, and using molecular and cell biology, experimental biophysics, and computer modeling. An advantage would be an interest in biochemistry and molecular biology, in particular DNA and protein preparation and modification (i.e. don't be 'column-shy'). If you also like physics and sophisticated optical equipment, and an open and friendly atmosphere, you will be very happy in our group.

References

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