S N BOSE NATIONAL CENTRE
FOR BASIC SCIENCES
Block JD, Sector III, Salt Lake, Kolkata 700 106

DEPARTMENTAL SEMINAR
Chemical and Biological Sciences

27th February,2024 **4.00 PM** **ONLINE / FERMION**

SPEAKER

Dr. Alexander Hautke
Postdoctoral Researcher, Ebbinghaus lab,
Ruhr-Universität Bochum and Research Center, Germany

TITLE OF THE TALK

Conformation, Localization and Condensation of CAG Repeat RNAs in vitro and inside living cells

ABSTRACT

In their physiological environment, biomolecules function in crowded, chemically complex surroundings with a plethora of interactions. Still, their behavior is studied in dilute aqueous solutions containing very few species of molecules, limiting the insights to be gained from such experiments. Yet, studies inside living cells are hard to undertake due to a strong background. However, Fast Relaxation Imaging (FReI) allows for studies both in vitro and inside cell. We are studying CAG repeat RNAs, which are connected to Huntington's Disease (HD), in vitro and inside HeLa cells investigating the impacts of macromolecular crowding, transient chemical interactions and water activity on their localization, folding stability and liquid-liquid phase separation (LLPS). Experiments are performed using FReI, Fluorescence Recovery after Photobleaching (FRAP) and fluorescence-based colocalization. CAG repeat RNAs form hairpins that grow in stability with the number of repeats. Further, they are sequence-specifically recruited into nuclear speckles at both physiological and pathological repeat length, retain their folded conformation inside the speckles and sequester transcription and translation factors, which is assumed to have an impact on HD pathology.

Our results reveal that a (CAG)20 hairpin is heavily destabilized by cosolutes, which is caused by nonspecific chemical interactions, reduced water activity or both. In cell, the governing factors seem to be a combination of destabilizing chemical interactions and high-water activity. Further, both hairpin folding stability and mobility inside nuclear speckles are strongly affected by ATP due to preferential interactions between the adenine moiety and the nucleobases in the unfolded state. This finding is quite striking given the fact that declining cellular ATP levels are a frequently occurring symptom in HD patients and that striatal neurons, which are predominantly affected in HD, are especially susceptible to a lack of ATP. Taken together, our experiments provide new insights into the biophysics of CAG repeat RNAs and its impact on HD. They underscore the importance of ATP for the cellular homeostasis of CAG repeat RNAs and will hopefully inspire further research towards a cure for the currently untreatable neurodegenerative disease that is HD.

HOST FACULTY

Prof. Rajib K Mitra
SENIOR PROFESSOR, CHEMICAL and BIOLOGICAL SCIENCES

****