TITLE: Role of nucleic acids dynamics in folding, function and disease

Summary: In the recent years, several fundamental cellular processes have been recognized to be driven by intrinsic dynamics of nucleic acids, putting them under the spotlight. Features of highly polymorphic nucleic acid motifs found in promoters of several genes indicate that dynamic structural equilibria must be at the heart of their role as transcriptional regulators to provide response to cellular cues and protein binding partners. The high energy, low populated conformational states of nucleic acids that are either targeted by binding partners or are the initiator of conformational change are challenging to detect with traditional methods, but can be analysed with a subset of methods in NMR spectroscopy. With this specific arsenal of NMR techniques, we plan to connect motions coded in nucleic acids sequence with structural fluctuations and rearrangements during re-folding and protein binding to illuminate how DNA transcription and gene expression rely on structural dynamics of nucleic acids.

Research techniques used: NMR is a primary research technique used, as it is particularly well suited for detection and characterization of conformational changes. NMR makes it possible to uncover exchange in time range from nanoseconds to hours, offers detailed structural information in form of chemical shifts and can report on alternative conformers present at very low abundance. The increasing size and number of interacting partners of the macromolecular systems of interest will require creative labelling schemes for nucleic acids and will be achieved by an introduction of isotopic labelling, methylated modified nucleotides or ^{19}F modified backbone. SAXS and cryoEM will be used to complement structural information obtained with NMR. A broad set of accessory methods (EMSA, microscale thermophoresis, CD and UV spectroscopy, enzymatic and computational methods) will be employed to establish functional consequences of structural dynamics.

The reason why the topic is innovative: The challenge for regulatory nucleic acid motifs in promoters is to obtain structural information for dynamic ensemble of each species that can exist and understand how accessibility of different parts of motif affects binding with cellular proteins and changes functional outcome. With NMR methods dedicated to detection of dynamic behaviour finding low abundant initiators of structural changes should be possible. Exploring structural dynamics in the context of long, double-stranded constructs should offer an alternative to otherwise fragmented insights and explain how small changes in primary sequence lead to disrupted gene expression and disease phenotype.

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