**Structural studies of phase-separating human gene regulatory proteins and their role in the structure and formation of membraneless organelles**

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The eukaryotic cell nucleus is exquisitely organised in order to facilitate and choreograph the complex manoeuvres required to decode the genome and regulate the production of its products. Within the nucleus are a number of non-membrane-bound bodies which self-assemble on cell division and adapt and reorganise throughout the cell cycle. Paraspeckles are an example of such subnuclear bodies that form when a specific group of nuclear RNA binding proteins are brought into close proximity by binding a 23000 nucleotide long noncoding RNA, NEAT1. Paraspeckle formation occurs coordinated and dynamic process of liquid-liquid phase condensation, where high concentrations of soluble paraspeckle proteins in the nucleus are poised to condense onto NEAT1 as soon as it is transcribed.

Proteins that build paraspeckles are enriched in various types of intrinsically disordered domain, including coiled-coil domains and ‘prion like domains’, typified by the proteins SFPQ, NONO, RBM14, FUS and HNRNPA1. We have been carrying out studies in vitro to characterise the biophysical properties of proteins required for paraspeckle formation in order to learn more about the mechanisms involved. In addition to gel- and liquid-formation assays and crystallographic studies, we have also been using small-angle neutron scattering to seek structural detail on the essential paraspeckle protein HNRNPK which forms a variety of aggregates, droplets and fibrils when made recombinantly. These insights have helped us build a better understanding of the dynamics and molecular structure of paraspeckle components.

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