Mechanisms of Rare Diseases

(from genome structural alterations to functional consequences)

Stefan Dimitrov

Institute of Molecular Biology, Sofia/Institute of Advanced Biosciences, Grenoble

Rare diseases are said to be rare when they affect one person in 2,000, i.e. more than 3 million French people and at least 30 millions Europeans. There are 7-8,000 rare diseases identified to date and the vast majority is from unknown origin. More than 90% of rare diseases are without treatment. Rare diseases are a major threat for human health and understanding of the molecular etiology of rare diseases is of primary need.

Here, I will initially present shortly some of our data on the mechanism of Rett Syndrome (RS), a very severe neuro-developmental rare disease. The genetic cause of the disease was defined as loss of function of *methyl CpG binding protein 2* (MeCP2). Nevertheless, the precise mechanism of how loss of function of this protein causes this devastating disease was not very clear. In contrast to the existing dogma claiming that MeCP2 binds to CpG conatining sequences, we observed that MeCP2 specifically recognizes and binds both *in vitro* and *in vivo* hydroxymethylated CA repeats. Moreover, we demonstrated a new function of MeCP2 as a long-range chromosome organiser, especially in chromatin domains associated with the nuclear lamina (LAD) which is the area at the inner face of the nuclear membrane. Therefore, MeCP2, previously described as transcriptional repressor, also organizes 3D chromatin architecture, and Rett Syndrome is, indeed, an epigenetic disease.

Next, I will describe a novel work flow for deciphering the molecular etiology of rare diseases and the application of this workflow on analyzing the Rahman Syndrome (RMNS) molecular origin. RMNS is a recently described developmental disorder caused by frameshift mutations in linker histone H1.4, that produce a truncated C-terminal domain (CTD) with reduced positive charge. We found that the mutation induces nucleosome arrays to adopt a more extended, flexible conformation exhibiting phase separation behavior similar to those lacking H1.4. Molecular dynamics simulations supported by FRET analysis indicate that the mutated CTD recognizes a shorter length of linker DNA, resulting

in a more open nucleosome conformation. Correspondingly, the mutation substantially increases H1.4 mobility within cell nuclei. The combined data suggest that RS mutations alter gene expression during development by promoting a relaxed chromatin state. This suggestion was further supported by a series of experiments at genome-wide level by using a cohort of "omics" approaches including ATAC-seq, RNA-seq and ChIP.

Finally, I will briefly summarize the objectives and preliminary data on a new project focused on Very Early Onset of Inflammatory Bowel Diseases (VEO-IBD). The data point out to the identification of new variants operating in VEO-IBD.

References

1. Boopathi, R. et al, *submitted*
2. Ibrahim A. et al., *Science* 2021, Jun 25;372(6549) :eabd5581.
3. Garcia-Saez I. et al., *Mol. Cell*, 2018 Dec 6;72(5):902-915.e7.
4. Bednar J. et al. , *Mol Cell.*, 2017, May 4;66(3):384-397.e8.