Brain gene regulation in 3D: long-range promoter-enhancer functional interaction networks in murine Neural Stem Cells, and their relevance for neurodevelopmental disease

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Keywords: Chromatin, long-range interactions, enhancer, NDD

Abstract: The structure of the genome has always been described as a one-dimensional phenomenon, but recent studies demonstrated that chromatin presents a highly coordinated 3D organization within the mammalian cell nucleus. This 3D network involves the establishment of physical long-range interactions between elements, like promoters and/or enhancers, located at a long distance on the linear genome map.
Recent experiments of our laboratory [1] showed that when an important transcription factor, SOX2, is deleted in murine Neural Stem Cells (NSCs), chromatin connectivity is significantly altered genome-wide, resulting in an overall reduction of gene expression. This points to the importance of the maintenance of a correct 3D chromatin organization for transcriptional regulation. Furthermore, many 'epigenetic enhancers' have been identified in murine NSCs genome, by ChIP-seq experiments for specific patterns of histone modifcations [1]. Many of these enhancers are conserved in humans [2]. Starting from these data, we generated a list of putative enhancers, functionally connected to mouse genes whose human orthologs are involved in neurodevelopmental diseases (NDD), such as Olig1 and Olig2. Olig1 and Olig2 are involved in diseases caused by Copy Number Variation (CNV), like Down Syndrome, and their expression is altered in many others. Therefore, we hypothesized that it may be possible to reduce excess expression of these genes through negative modulation of the activity of one or more enhancers. We found that targeted silencing of specific putative Olig1 and Olig2 enhancers, using a CRISPR/dCas9 system, causes decreased expression of these connected genes. Olig1 and Olig2 are at the center of a complex interaction network involving many adjacent regions, including both genes and intergenic territories; so, it is possible that interfering with a specific interaction might affect the function of other genes. For this reason, we are now investigating the activity of genes involved in these interactions, such as Paxbp1 and Synj1. The identification of enhancers regulating specific genes associated to NDD is a key step in the discovery of extracellular signals and pathways involved in enhancer activity, leading to the possibility to also use new drugs, instead of CRISPR/dCas9 based approach, for therapeutic purposes.

References:
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