RNA sequencing analysis in iPSCs derived Rett neurons

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Mutations in *MECP2* gene are responsible for the large majority of classic Rett syndrome cases and for a percentage of variant patients. *MECP2* encodes for a transcriptional regulator that plays essential roles in different biological processes. To study MeCP2-related pathways directly in human neurons, we established a neuronal model of the disease by genetic reprogramming of patient fibroblasts into induced Pluripotent Stem Cells (iPSCs) and subsequent neuronal differentiation. Specifically, isogenic clones from the same patient (classic Rett; mutation p.Thr158Met) that express either the wild type or the mutant *MECP2* allele as a consequence of X chromosome inactivation have been employed. RNA-seq analysis has been performed on total ribodepleted RNA isolated from mutated and control cells and data have been analyzed by DESeq tool. Gene Ontology and Kegg pathway analyses (DAVID) on differentially expressed mRNAs with a fold change ≥2 (P-value ≤0,01) revealed up-regulation of genes involved in neuronal differentiation and down-regulation of genes involved in cell cycle regulation and glial differentiation. This is in line with the functions of MeCP2 as a regulator of cell proliferation and of the proliferation/differentiation balance in neuronal precursors. Interestingly, between up-regulated genes we also observed transcripts involved in extracellular matrix (ECM) organization, consistent with the emerging role of ECM in the regulation of neuronal maturation and plasticity. In conclusion, this study confirms the relevance of known MeCP2-related pathways and identifies new altered signaling pathways in a human neuronal model of classic Rett syndrome, providing information useful for the design of new therapeutic strategies.