Genetic and epigenetic mechanisms deregulated in Rett syndrome

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Background
Rett Syndrome (RTT; OMIM 312750) is a progressive postnatal neurodevelopmental disorder, characterized by motor dysfunction, autistic−like features and mental retardation. RTT is one of the leading causes of mental disability in girls, with an incidence of 1/10,000 female live births. More than 95% of RTT patients carry mutations in the X-linked Methyl-CpG-binding Protein 2 (MECP2) gene, encoding a key epigenetic factor involved in both transcriptional regulation and in the organization of higher-order chromatin structure$^{1,2}$. MECP2 encodes two splicing isoforms, MeCP2A and MeCP2B, both ubiquitously expressed but enriched in neurons. In mouse, MeCP2 accumulates at pericentric heterochromatin (PCH), which is organized to form repressive nuclear compartments known as chromocenters.

The ablation of Mecp2 in mice recapitulates many aspects of RTT. Importantly, neurological defects in Mecp2−/y mice can be rescued by restoration of the gene, establishing the principle of reversibility in RTT.

Preliminary studies highlighted altered glycosphingolipid (GSL) levels in brain of RTT patients, although these data are still controversial. So far, the gap between MeCP2 dysfunctions and RTT clinical manifestations has not yet been bridged and no effective therapy is currently available.

Our research is focused to understand genetic and epigenetic mechanisms deregulated in RTT by using both cellular and animal models.

Results
In recent years, we have been studying the function of MeCP2 in the organization of chromatin architecture in neurons. We highlighted a key role of MeCP2 in the chromocenter clustering during neural differentiation$^3$, with the contribution of Alpha-thalassemia/mental retardation syndrome X-linked (ATRX)$^4$, a chromatin remodeling factor mutated in ATR-X syndrome, another disease characterized by intellectual disability. Moreover, MeCP2 and ATRX are reciprocally dependent for their expression and targeting to PCH$^4$. Furthermore, we demonstrated that MeCP2 and major satellite forward (MajSat-fw) non-coding RNA (ncRNA), transcribed from the PCH satellite repeats$^5,6$, are mutually dependent for their targeting to chromocenters and cooperate for PCH organization in neurons. Moreover, MeCP2B and its methyl-binding domain are the major players for higher-order PCH organization. Finally, we demonstrated that one of the most common MeCP2 RTT mutation, T158M, impairs higher-order PCH organization$^7$. These findings contribute to clarify the function of MeCP2 and its partners (proteins and ncRNAs) in the formation of repressive nuclear compartments and allow to hypothesize that alterations in this process might concur to RTT pathogenesis.

With a parallel project, we studied the function of MeCP2 in the regulation of metabolism of GSLs, glycosylated lipids abundant in the nervous system. We found altered distribution and levels of several GSLs in Mecp2−/y mouse brain that parallel a global deregulation of genes encoding key enzymes involved in GSL metabolism (glycogenes)$^8$. MeCP2 bound the promoter of several glycosogenes, thus suggesting a direct role in their regulation. In addition, MeCP2 represses the expression of AUTS2$^8$, an epigenetic factor mutated in autism, which is, in turn, involved in GSL reprogramming in neurons$^9$. Globally, the alterations in GSL metabolism were confirmed in murine neurons carrying T158M and R306C RTT missense mutations$^8$. 
Altogether, these findings suggest that MeCP2 controls GSL content in brain by modulating the expression of Auts2 and of a subset of glycogenes. In light of this, we hypothesize that a deregulation of the MeCP2/AUTS2/GSL axis contributes to RTT clinical manifestations.

**Future objectives:** An imbalance of AUTS2 dosage, caused by MeCP2 dysfunctions, might contribute to RTT pathogenesis through an aberrant regulation of GSL metabolism. In this frame, we will investigate the role of MeCP2/AUTS2 crosstalk in the regulation of GSL metabolism and the impact of AUTS2 dosage in RTT clinical manifestation, through the generation of a novel double mutant mouse model, MeCP2<sup>−/−</sup>;Auts2<sup>−/−</sup>, and a new MECP2-null/Auts2 knock-down human neuronal cell line. In these models, we will study the effect of the "normalization" of AUTS2 expression in a MeCP2-null context, by evaluating the rescue of RTT phenotype (in mice) and GSL deregulation.

**References:**


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