Novel human Rett syndrome in vitro models demonstrate differential impact of specific MeCP2 mutations on neuronal functions and the non-cell autonomous effects of glial cells

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Abstract

Rett syndrome (RTT) is an X-linked dominant neurodevelopmental disorder caused by mutations in the methyl CpG-binding protein 2 (MECP2) gene. MeCP2 activity is critical for early brain development and adult and aging brain functions. Although clinical studies show genotype-phenotype correlations, the role of specific MeCP2 mutations on neural functions has not been yet fully elucidated. Recent data suggest a role of glial cells in the RTT pathogenesis in a non-cell autonomous manner. We analyzed the impact of MeCP2 silencing and over expressed R168X, T158M, R133C and R306C mutants on neuronal functions and on non-cell-autonomous effects of astrocytes and microglia using human primary immortalized neural cells. Silencing of MeCP2 in human NSCs inhibited □3-tubulin and increased GFAP expression. Primary neuronal cultures silenced for MeCP2 or overexpressing R168X and T158M mutants exhibited lower degree of neurite outgrowth, impaired mitochondrial function and decreased BDNF secretion compared to control and R133C mutant expressing cells. Human astrocytes silenced for MeCP2 expressed lower levels of GFAP, glutamate transporter EAAT2 and IGF-1 secretion. MeCP2 silenced microglia showed increased glutamate secretion, decreased BDNF secretion and M1 phenotypes. Analyzing miRNA expression in MeCP2 silenced astrocytes and microglia identified novel altered cellular and exosomal miRNAs and long non-coding RNA expression. We conclude that immortalized human neural cells expressing different MeCP2 mutants can serve as a novel model system for studying molecular mechanisms associated with the pathogenesis of RTT in both neurons and glial cells and for analyzing the impact of specific MeCP2 mutations on various aspects of cell function and interactions.