REGULATION OF TAU EXON 10 SPLICING: A SHRNA SCREEN OF THE HUMAN SPLICEOSOME

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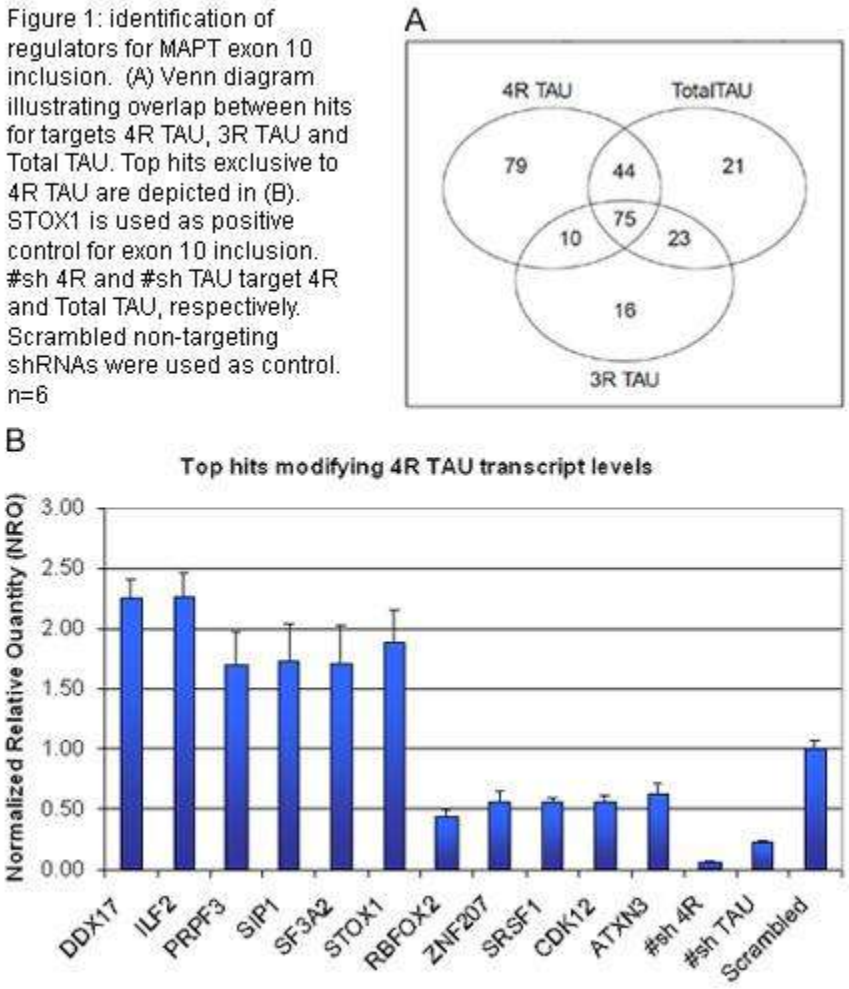
Mutations in *MAPT* are a major cause of frontal temporal dementia (FTD) of which a subgroup alters alternative splicing of TAU mRNA, primarily resulting in excess inclusion of exon 10. In this study we aim to find regulators of TAU exon 10 inclusion, which might provide us with potential targets for FTD therapeutics. To identify regulators of TAU exon 10 splicing, we performed a short hairpin RNA (shRNA) library screen of the human spliceosome in BE(2)M17 neuroblastoma cell lines. The library contains ~330 shRNAs, targeting spliceosomal small nuclear ribonucleoproteins (snRNPs), snRNP-associated and non-snRNP splicing factors. Alterations in exon 10 inclusion have been determined by quantitative PCR, which has been optimized to specifically detect endogenous 4R and 3R TAU transcript levels. The screen was conducted with n=6, giving us >90% power to detect 40% changes in 4R TAU transcript levels. We found several novel inhibitors as well as enhancers of TAU exon 10 inclusion; results are depicted in figure 1. Next we will deconvolve the shRNA pools and confirm hits that rescore with multiple shRNAs. Primary cortical neurons of humanized *MAPT* mice and human induced pluripotent stem cells differentiated into neurons will be used for confirmation of hits and detailed molecular follow-up, to test if confirmed hits could be potential targets for FTD therapeutics.

Figure 1: identification of regulators for MAPT exon 10 inclusion. (A) Venn diagram illustrating overlap between hits for targets 4R TAU, 3R TAU and Total TAU. Top hits exclusive to 4R TAU are depicted in (B). STOX1 is used as positive control for exon 10 inclusion. #sh 4R and #sh TAU target 4R and Total TAU, respectively. Scrambled non-targeting shRNAs were used as control. n=6



[Results from shRNA screen of the human spliceosome]