

Presenter: Rui Zhang

Authors: RUI ZHANG, Yi-Tao Yu

Title: U2 SNRNA PSEUDOURIDYLATION IN THE RECOGNITION OF BRANCH-SITE SEQUENCE DURING PRE-MRNA SPLICING

Abstract

During spliceosome assembly, spliceosomal small nuclear ribonucleoproteins (snRNPs) and splicing factors recognize and interact with the pre-mRNA sequence. Specifically, U2 snRNP recognize the branch site sequence (BSS) of pre-mRNA and the binding last until splicing complete. Pseudouridines (Ψ s) are concentrated in the branch site recognition region (BSRR) of U2 snRNAs and it is proved to be essential for the splicing of pre-mRNAs. However, information is still limited regarding how exactly the Ψ s contribute to recognizing the pre-mRNA BSS during splicing. Our group developed a yeast screening system where a library of reporter pre-mRNAs with randomly mutated BSSs is transformed into different yeast strains containing different numbers and combinations of Ψ s in the U2 BSRR. The BSSs, which can support pre-mRNA splicing under different U2 Ψ background were screened and sequenced, allowing us to decode how Ψ contribute to the recognition of different BSSs. With high throughput sequencing and spot test assay, we identified that Ψ s in the BSRR of U2 snRNA play a role to select different BSSs. By RT-PCR, we confirmed that not only in a reporter gene, but also in the native genes, Ψ s in the BSRR of U2 impact pre-mRNA splicing. With primer-extension assay, we further demonstrated that Ψ s in the BSRR of U2 play a role in the alternative splicing, which is common in other eukaryotes but rare in yeast.