Nature Reviews Molecular Cell Biology | AOP, published online 6 August 2014; doi:10.1038/nrm3856

SPLICING

Catalytic nulls keep busy

> “splice variants abrogated the … catalytic domain but retained … the later-gained domains”

Aminoacyl-tRNA synthetases (aaRSs) catalyse the fusion of amino acids to tRNAs for protein synthesis, but during evolution they have also gained and retained 13 protein domains that can confer other functions. Some of these domains are generated by alternative splicing, which led Schimmel and colleagues to characterize the alternative splice variants of the aaRS genes. Their analysis revealed a set of catalytically inactive (‘null’) aaRSs that have various biological functions that are independent of tRNA aminoacylation.

Daniil Chetverikov / Alamy

The authors carried out a comprehensive search for alternative splice variants of 37 human aaRSs in brain tissues and in leukocytes, using gene-specific multiplex PCR of exon–exon junctions, followed by deep sequencing. They discovered hundreds of novel splice variants and, surprisingly, found that 60 of the 70 identified internal in-frame splice variants abrogated the highly conserved catalytic domain but retained at least one of the later-gained domains. These variants are therefore catalytic nulls in regard to the canonical function of aaRSs, but they may have other biological functions. In support of this, the observed loss of specific exons could potentially lead to extensive structural changes and thus create new protein interactions.

Many of the splice variants were found to be tissue-specific, which suggests that they encode expressed proteins. To explore this possibility, the authors examined 48 of the catalytic null mRNAs and found that all were associated with polysomes and thus potentially translated. Western blot analysis using antibodies specific for five aaRSs detected the presence of the aaRS protein fragments that were expected to have lost the catalytic domain but to have retained the gained domains, and this was corroborated by mass spectrometry. In total, 38 of the 48 transcripts were differentially expressed across a panel of adult and foetal tissues or cells, whereas the parent aaRS genes were evenly expressed.

In order to investigate the potential functions of catalytically null aaRSs, the authors expressed and purified 94 recombinant aaRS protein fragments (as well as a few parental aaRSs as controls) and tested them in various phenotypic cell-based assays, including for proliferation, differentiation, acute inflammatory responses and transcriptional regulation. Whereas the parental proteins were inactive or had a single activity that differed from any of their splice variants, 83 of the aaRS protein fragments tested positive for at least one activity and most tested positive for a set of activities that was specific to the variant.

In summary, the unique characteristics of the alternative splicing of aaRS transcripts enabled the generation of a large ensemble of previously unknown proteins that have various non-canonical functions. It will be interesting to see to what extent this mechanism is shared by other enzyme families.

Eytan Zlotorynski

ORIGINAL RESEARCH PAPER Lo, W.-S. *et al.* Human tRNA synthetase catalytic nulls with diverse functions. *Science* **345**, 328–332 (2014)
FURTHER READING Guo, M., Yang, X.-L. & Schimmel, S. New functions of aminoacyl-tRNA synthetases beyond translation. *Nature Rev. Mol. Cell Biol.* **11**, 668–674 (2010)