Poster Presentation

MS93.P02

Structural and functional studies of the decapping exoribonucleases

J. Chang[1,2], L. Tong[2]

[1]Kyungpook National University, Department of Biology, Teachers College, Daegu, South Korea, [2]Columbia University, Department of Biological Sciences, New York, NY, USA

Recent studies showed that two homologous yeast proteins, Rai1 and Dxo1, function in a quality control mechanism to clear cells of incompletely 5' end-capped messenger RNAs (mRNAs). Rai1 possesses a novel decapping activity that can remove the entire cap structure dinucleotide from an mRNA. This activity is targeted preferentially towards mRNAs with unmethylated caps in contrast to the canonical decapping enzyme, Dcp2, which targets mRNAs with a methylated cap. Dxo1 also has robust decapping activity on RNAs with unmethylated caps, but it has no detectable pyrophosphohydrolase activity. Unexpectedly, we found that Dxo1 also possesses distributive, 5'-3' exoribonuclease activity, and we named Dxo1 (originally Ydr370C) for this new eukaryotic enzyme with both decapping and exonuclease activities. Studies of yeast in which both Dxo1 and Rai1 are disrupted reveal that mRNAs with incomplete caps are produced even under normal growth conditions, in sharp contrast to current understanding of the capping process. Here, we introduce that their mammalian homolog, Dom3Z (referred to as DXO), possesses pyrophosphohydrolase, decapping, and 5'-3' exoribonuclease activities. Surprisingly, we found that DXO preferentially degrades defectively capped pre-mRNAs in cells. Additional studies show that incompletely capped pre-mRNAs are inefficiently spliced at all introns, a fact that contrasts with current understanding, and are also poorly cleaved for polyadenylation. Crystal structures of DXO in complex with substrate mimic and products at a resolution of up to 1.5 Å provide elegant insights into the catalytic mechanism and molecular basis for their three apparently distinct activities. Our data reveal a pre-mRNA 5' end capping quality control mechanism in mammalian cells, indicating DXO as the central player for this mechanism, and demonstrate an unexpected intimate link between proper 5' end capping and subsequent pre-mRNA processing.

[1] *X. Jiao, J.H. Chang, T. Kilic, L. Tong, M. Kiledjian, Molecular Cell, 2013, 50, 104-115,* ***[2]*** *J.S. Mugridge, J.D. Gross, Molecular Cell, 2013, 50, 2-4,* ***[3]*** *J.H. Chang, X. Jiao, K. Chiba, C. Oh, C.E. Martin, M. Kiledjian, L. Tong, Nature Structural & Molecular Biology, 2012, 19, 1011-1017*

Keywords: DXO, pre-mRNA quality control, 5' end capping