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Cryo-electron microscopy (cryo-EM) studies of the ribonucleoprotein complexes: group II intron and ribosomes

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Abstract

Single-particle cryo-EM is an emerging technique in the field of structural biology, with the recent advancement in electron detection technology and improved image processing algorithms, it is now possible to achieve atomic resolution structure of macromolecular complexes. We have been applying cryo-EM to illustrate the architecture of ribonucleoprotein complexes, such as bacterial group II introns and ribosomes. Bacterial group II introns are large ribonucleoprotein complex comprising the intron ribozyme and an intron-encoded protein. They self-splice, yielding mature RNA, and integrate into DNA as retroelements. Recently, the first cryo-EM structure of *Lactococcuslactis* group IIA intron, in complex with an intron-encoded protein, was resolved at 3.8 Å. Molecular analysis of the cryo-EM structure of the group II intron revealed functional coordination between the intron RNA and protein. In addition, the protein structure revealed a close relationship between its reverse transcriptase catalytic domain and telomerase, while its splicing center resembles the spliceosomal Prp8 protein, suggesting a complex ancestral relationship among these proteins.

Ribosomes, the protein synthesis machineries of the cell, are large macromolecular complexes. The mammalian mitochondria contain its own protein synthesis machinery including ribosomes (mitoribosomes). The mitoribosome contains significantly smaller rRNAs and a larger mass of mitochondrial ribosomal proteins (MRPs), including additional 35 mito-specific MRPs, as compared to the bacterial ribosome. Most of the mammalian mitochondrial mRNAs are leaderless, i.e., lacking a 5'UTR and therefore also lacking a Shine-Dalgarno sequence. This situation has some similarity to that in *Mycobacterium smegmatis*, where about one-third of mRNAs are leaderless. How these leaderless mRNAs are recruited by the ribosomes to initiate protein synthesis is unknown. While the cryo-EM structure of the mitoribosome have yielded the architecture of ribosomal small subunit and existence of the previously eluded one of the tRNA-bindign sites, the E-site, on the mitoribosome, our structural studies of the *M. smegmatis* 70S ribosome has also revealed some of the unique features of the mycobacterial ribosome. I will summarize results of above studies that I have been currently involved in, and will outline my future research plan that I would like to pursue as an independent investigator.