Poster Presentation

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Structural analysis of tRNA(His) guanylyltransferase comlexed with tRNA

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tRNA(His) guanylyltransferase (Thg1) of eukaryote adds a guanylate to the 5’ end of immature or incorrectly processed tRNAs (3'-5' polymerization) by three reaction steps: adenylylation; guanylylation and dephosphorylation. This additional guanylate provides the major identity element for histidyl-tRNA synthetase to recognize its cognate substrate tRNA(His) and differentiates tRNA(His) from the pool of tRNAs present in the cell (1). Previous studies indicate that Thg1 is a structural homolog of canonical 5'-3' polymerases in the catalytic core with no obvious conservation of the amino acid sequence(2). However, the substrate binding of Thg1 is unclear and requires information on the three-dimensional structure in complex with tRNA. In this study, we determined the crystal structures of Thg1 from Candida albicans (CaThg1) in tRNA-bound (CaThg1-tRNA), ATP-bound (CaThg1-ATP), and GTP-bound (CaThg1-GTP) form, and elucidated how Thg1 functions as a reverse polymerase to add nucleotide(3). The crystal structures of CaThg1-tRNA complex shows that two tRNAs are bound to tetrameric Thg1 in parallel orientation which is consistent with SAXS (Small angle X-ray scattering) and gel filtration analysis. One tRNA interacts with three monomers for its positioning, anticodon recognition, and catalytic activation. The end of the acceptor stem and the anticodon loop are both recognized by the same sub-domain belonging to the different monomers. Moreover, the structural comparison of Thg1-tRNA with canonical 5'-3' polymerase shows that the domain architecture of Thg1 is reversed to that of canonical 5'-3' polymerase.

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