Nanoarchitectural Engineering of RNA For Structural Determination Using Cryo-EM

Prof. Peng Yin[1], Di Liu[1]
[1]*Harvard University*
py@hms.harvard.edu

High-resolution structural studies are essential for understanding the folding and function of diverse RNAs. Herein, we present a nanoarchitectural engineering strategy for efficient structural determination of RNA-only structures using single-particle cryo-EM. This strategy—ROCK (RNA oligomerization-enabled cryo-EM via installing kissing loops)—involves installing kissing-loop sequences onto the functionally nonessential stems of RNAs for homomeric self-assembly into closed rings with multiplied molecular weights and mitigated structural flexibility. ROCK enables cryo-EM reconstruction of the *Tetrahymena* group I intron at 2.98-Å resolution overall (2.85 Å for the core), allowing de novo model building of the complete RNA, including the previously unknown peripheral domains. ROCK is further applied to two smaller RNAs—the *Azoarcus* group I intron and the FMN riboswitch, revealing the conformational change of the former and the bound ligand in the latter. ROCK holds promise to greatly facilitate the use of cryo-EM in RNA structural studies.