The 3D cryoelectron ultrastructure of translating polyribosomes.

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Abstract

Cryoelectron tomography (CET) allows for the three-dimensional localization and structural characterization of large complexes in vitrified samples.
A template matching approach developed in our group was used to map 70S ribosomes within the cryoelectron tomogram of a bacterial cell-free protein synthesis system translating a luciferase model construct. Positively identified 70S particles were found both as isolated species and as rather ordered arrangements that resemble 'two rowed' polysomes.
The classification of polysomal vs. monosomal particles by assessing relative spatial orientation and distance *in silico* is an alternative to biochemical separation of translating from non-translating ribosomes for structural investigations.
Beyond the possibility to obtain structural information of classified isolated ribosomes as entities, the topological examination of polysomal particles could provide us with insights into the 'nano-environment' for the folding of nascent polypeptides. We show that the peptide tunnels of adjacent 70S particles on a polysome have a certain preferred orientation to each other. This observation might have implications on co-translational events of assembly or aggregation of nascent peptide chains.