Real time imaging of mRNA expression dynamics in live cells using protein complementation methods

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Traditional methods for mRNA quantification in cells, such as northern blots, quantitative PCR or microarrays assays, require cell lysis and therefore do not preserve its dynamics. These methods cannot be used to probe the spatio-temporal localization of mRNA in cells, which provide useful information for a wide range biomolecular process, including RNA metabolizim, expression kinetics and RNA interference. To probe mRNA dynamics in live prokaryotic and eukaryotic cells, we develop a method, which exploit the strong affinity of the eukaryotic initiation factor 4A (eIF4A) to specific RNA aptamers. Two parts of the eIF4A are fused to a split Green Fluorescence Protein (GFP), and are expressed in the cells at high abundance. However, only when the RNA apatmer is also present, the two protein parts complement and become fluorescent. Thus, the fluorescent background remains low, allowing us to directly image the expression of mRNA molecules in live e-coli cells from its early onset, over hours. We find that the expression kinetics can be classified in one out of at least three forms, which also display distinct spatial distributions. I will discuss the possible biological origin for these distributions and their time evolution.

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