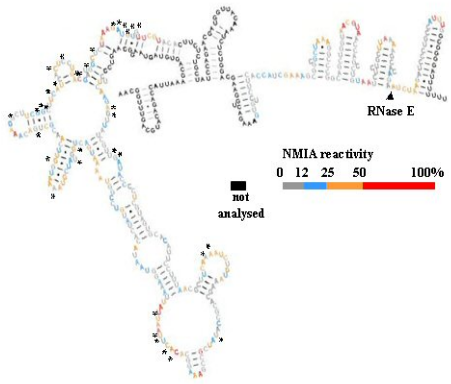
A new perspective on the initiation of bacterial mRNA degradation

Louise Kime, Stefanie S. Jourdan, Jonathan A. Stead and Kenneth J. McDowall

In current models of mRNA degradation, the process is initiated by "decapping" to generate a 5'-monophosphate group that stimulates cleavage by RNase E, an endonuclease required for the rapid degradation of many, if not most transcripts in *E. coli*. A key assumption is the commonly held view that RNase E is only able to cleave efficiently those RNAs that have a 5'-monophosphate group. The results presented here show that this is not in fact the case.

In previous reports, we have described the structure and function of the pocket in RNase E and related enzymes that can bind a 5' monophosphate. Last year, we suggested that *E. coli* RNase E was capable of the rapid cleavage of a quadruplexed oligonucleotide lacking a 5'-monophosphate group. We can now report that 5'-biotinylated RNA can also be cleaved rapidly when conjugated to streptavidin prior to incubation with the NTH of RNase E.

RNase E is a tetramer, more precisely a dimer of dimers, and modelling studies have indicated that two RNA-binding channels in a principal dimer could simultaneously contact single-stranded regions in the context of either the quadruplexes or streptavidin conjugates. This raised the possibility that the requirement for the rapid cleavage of substrates that lack a 5'-monophosphate could be as simple as multiple single-stranded regions accessible to RNase E. Michaelis-Menten analysis is consistent with the duplication of contacts increasing the affinity of RNase E for substrates.



Secondary structure model of *cspA* shown with RNase E recognition sites(*).

Having found that model substrates can be cleaved rapidly by RNase E independent of interaction with a 5'-monophosphate, we extended our analysis to transcripts of *E. coli* and found that *cspA* mRNA can be cleaved rapidly when it has a 5' triphosphate group. We have found similar results for other transcripts including *epd-pgk* mRNA. Moreover, we have probed the structure of *cspA* mRNA using selective acylation of 2'-hydroxyl groups and have confirmed that multiple single-stranded regions are indeed recognised by RNase E.

We propose that the initiation of the decay of many transcripts in *E. coli* is not dependent on "decapping", but rather RNase E cleavages that are facilitated by interaction with multiple single-stranded regions. This model provides a simple explanation for how a change in the rate of translation, as mediated by non-coding RNAs for example, often has an inverse effect on the rate of mRNA turnover. It also explains the recent finding that the decapping enzyme is not essential and only influences the decay of a relatively small proportion of the mRNA pool.

Publications

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