

## **Determinants of translation termination fidelity in eukaryotes**

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### **Abstract**

Eukaryotic translation process is generally terminated when the ribosomal A site is populated by a stop codon. A release factor recognizes it, and the polypeptide chain is released from the ribosome. In some probability, however, translation continues past this point, into the 3'UTR region. Such events, referred to as readthrough events, happen when, instead of the release factor, a tRNA recognizes the stop codon. The causes for these events are not completely understood yet and it is therefore hard to predict their probability for a given mRNA.

Understanding the different mRNA features effecting termination fidelity can be used in biotechnological objectives to adjust termination probability. One example could be in heterologous expression, when the target gene is linked to a homologous essential gene to improve stability of the system. In this case, enhancing readthrough probability would result in two products: one where both genes are translated together, and one where only the target gene is translated. This way the desired stability is maintained, while a clean protein product of the target gene is produced.

Previous studies include both reporter systems, showing termination fidelity is determined by the identity of the stop codon and flanking nucleotides, and prediction models confirming it genome-wide with additional features. Reporter systems have the advantage of showing proved and measurable readthrough events but lack genome-wide perspective. There are only a few existing prediction models, so there is room to explore new features.

Here we try to better estimate readthrough probability genome-wide using Ribo-Seq data, and test additional features striving to get better prediction. Some of the feature we examine are related to codon usage bias, folding energy of mRNA, and stop codons in all reading frames. Furthermore, we will analyze all features statistically genome-wide, hoping to understand the nature of each feature's individual effect, and how they work together. To demonstrate our results, we are conducting an experiment where several sequences are designed with appropriate modifications, based on Mcherry gene linked to ECFP gene, with stop codon in between.