How can one tRNA decode multiple codons?

Mechanism of ambiguous codon decoding facilitation by tRNA modifications at the wobble position

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In Short

- tRNA modifications are essential in translation, but the mechanisms of their function are poorly understood.
- Modifications at the wobble position of tRNA (1st anticodon position) are particularly important in ambiguous decoding, when one anticodon decodes multiple codons that differ by their third position.
- This project is focused on the tRNA modifications that promote ambiguous decoding of purine-ending codons.
- Molecular modeling allows to dissect the ambiguous decoding mechanism into the elementary physichochemical effects of the tRNA modifications.
- Hybrid QM/MM scheme is applied to model the ribosome environment and tRNA modifications effects on the proton transfer reactions in base pairs.

Translation of nucleotide sequences of mRNAs into amino acid sequences of proteins is governed by the ribosome – the most ancient molecular machine in all living systems. During this process, the codons of an mRNA are decoded by anticodons of aminoacylated tRNAs, and this codon-anticodon decoding is arguably the most complex recognition mechanism in cells. One of its still poorly understood features common in all known organisms is the ambiguous decoding, when a single anticodon is able to efficiently decode several synonymous codons that differ by their third position.

In most cases, this phenomenon is largely dependent on various modifications of tRNAs, especially at the first (wobble) position of anticodons [1]. It is assumed that the mechanism of ambiguous decoding involves formation of Watson-Crick (WC)-like geometries in non-canonical base pairs, and tRNA modifications facilitate this process. Experiments show increased decoding of both purine (R)-ending codons by anticodons with some modified wobble uracils [2]. Recent studies reveal various effects of wobble tRNA modifications on the anticodon [3], but current models cannot explain the mechanism of ambiguous decoding they ensure, and fail to faithfully explain the experimental observations.

Based on our previous studies of some wobble uracil modifications using simpler models, we suggest two separate mechanisms for enhanced binding to A- and G- ending codons, which stem from the same physical origin – the electron-withdrawing property of the modifications that increases acidity of the modified uracil.

We propose a mechanism involving a proton transfer in the modified A-U base pairs that leads to the increased interaction energy of the base pair, thus the strength of the codon-anticodon binding in the case of full-cognate A-ending codons. The suggested mechanism of the facilitated binding to the G-ending codons involves acceleration of the recently discovered tautomerization reaction, increasing a probability to form a WC-like G-U base pair in a presumably kinetically-restricted codon recognition step during translation.

In this project we set a direction to the more realistic models of ambiguous decoding. The proton transfer reactions in the codon-anticodon base pairs will be modeled in a ribosome A-site environment using hybrid QM/MM simulations. Multi-replica metadynamics will be applied to reveal the free energy landscape of the proton transfer in A-U base pairs. As the tautomerization reaction in G-U base pairs is more complex than a single proton transfer, it will be studied using umbrella sampling method with the utilization of the path collective variables.

Our models produce specific predictions which will be tested in the experimental part of this project.

WWW

https://www.chemie.uni-hamburg.de/institute/bc/arbeitsgruppen/ignatova/personen/mitarbeiter/ignatova-zoya.html

More Information

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[2] S. Kurata *et al*, *J. Biol. Chem.* **283**, 18801-18811 (2008). doi:10.1074/jbc.M800233200

[3] E. Sochacka *et al*, *Nucleic Acids Res.* **45**, 4825-4836 (2017). doi:10.1093/nar/gkw1347

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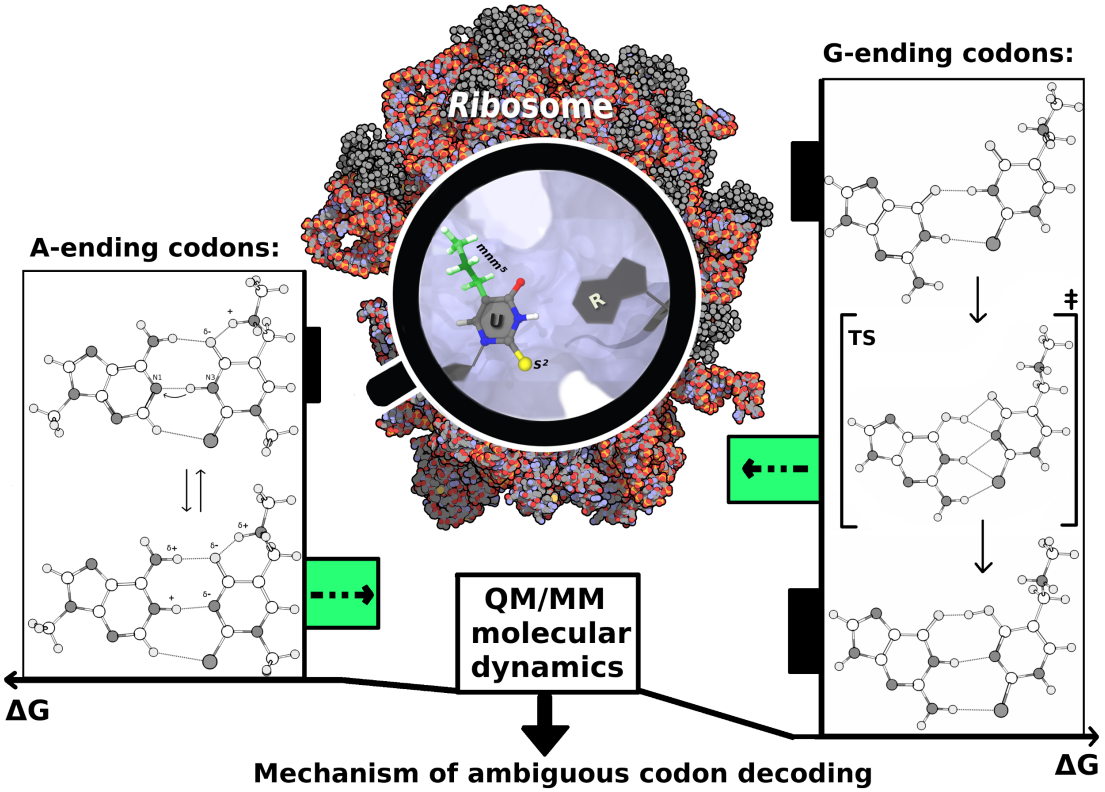

Figure 1: *The scheme of the mechanism studied in this project. When an anticodon with a wobble uridine modification (methylaminomethyl-2-thiouridine (mnm5s2U) in this example) encounters its cognate R-ending codon in the A-site of the ribosome, the modified nucleotide facilitates ambiguous decoding by proton-transfer-mediated charge redistribution in the case of the full-cognate A-ending codon (left), decreasing base pair interaction free energy, and by accelerating the tautomerization reaction leading to the WC-ike geometry of G-U base pair in the case of the G-ending codon. In both cases the effect is due to the increased acidity of the modified nucleotide.*