Evolution of TAG codon in Methanosarcina

Margarita Meer[a], Inna Povolotskaya[a], Peter Vlasov[a], Viktoria Shcherbakova[b],

Fyodor Kondrashov[a]

a. Centre for Genomic Regulation (CRG) and Universitat Pompeu Fabra (UPF), Barcelona, Spain

b. Skryabin Instiute of Biochemistry and Physiology of Microorganisms, Pushchino, Russia

margarita.meer@crg.eu

Pyrrolysine (Pyl, O) is the 22nd amino acid known to be included into proteins during translation. It can be found mostly in methanogenic archaea of the genus Methanosarcina. Pyrrolysine is encoded by TAG codon [1]. At the same time TAG still retains the stop codon function in this genus [2].

In contrast to the Selenocysteine (Sec) coding system, where the SECIS structure determines the Sec-coding function of a specific TGA codon, such system has not been found for Pyl-coding TAG codons [3].

Thus, TAG codon in Methanosarcina is bi-functional with the Pyl-coding and stop codon functions being in a competitive inhibition-like state. Thereby from a single mRNA at every read through, a TAG codon has a chance of being recognized by translation machinery as a stop codon or as a Pyl codon.

Consistent with such an inefficient system, the Methanosarcina genomes contain very few TAG codons, suggesting them to be deleterious. Our goal was to investigate the evolution of TAG codons in this genus.

To this end, we have sequenced genomes of 14 closely related strains of three Methanosarcina species in addition to several genomes found in GenBank. Using a phylogenetic approach we have estimated the rate of TAG gain and loss in these genomes.

TAG codons in Methanosarcina can be broadly classified into two classes. First, stop-like TAG codons that are at the end of ORFs and correspond to stop-codons in other strains and species. And second, Pyl-like codons are found in the middle of the ORFs and correspond to amino acid coding codons.

We find that Methanosarcina genomes are accumulating Pyl-like TAG codons. At the same time stop-like codons evolve faster than either TAA or TGA codons, indicating that Methanosarcina genomes are not in equilibrium for TAG codon content.

The recent evolution of the Pyl-coding Methanisarcina genomes provides a remarkable example of evolutionary dynamics associated with an ongoing change of the genetic code. Our data suggest that a switch in the coding function of a codon can proceed through a bi-functional state of the codon that gradually increases the prevalence of the new function while loosing the propensity to perform the old one.

1. Hao B. et al. (2002) A new UAG-encoded residue in the structure of a methanogen methyltransferase, *Science*, **296(5572):**1462-6.

2. Alkalaeva E. et al. (2009) Translation termination in pyrrolysine-utilizing archaea, *FEBS Lett.*, **583(21):**3455-60.

3. Namy O. et al. (2007) Adding pyrrolysine to the Escherichia coli genetic code, *FEBS Lett.*, **581(27):**5282-8.