Cytosolic help for mitochondrial defects. A novel method for importing tRNA into mitochondria in order to suppress mutations

Created: December 4, 2000

The mitochondrion has cut back its genome substantially since taking up residence in cells as a symbiont 1.5 billion years ago, but it retains its personal transcription, translation and protein-assembling systems, including its tRNA genes.

Even so, the mitochondrion is not fully self-sufficient — to varying extents yeast, plants and protozoan cells can borrow nuclear-encoded tRNA molecules to ease the task of translating transcripts of their mitochondrial genes. New data indicate that nuclear-encoded tRNAs can even be used to salvage errors in mitochondrial transcripts.

In the yeast *Saccharomyces cerevisiae,* only one tRNA ($^{Lys}_{CUU}$) is carried into the mitochondrion, something it can do only if charged with an amino acid, and only if aided by cytosolic import factors. Among these factors is the precursor of the mitochondrial lysyl-tRNA synthetase (pre-MSK).

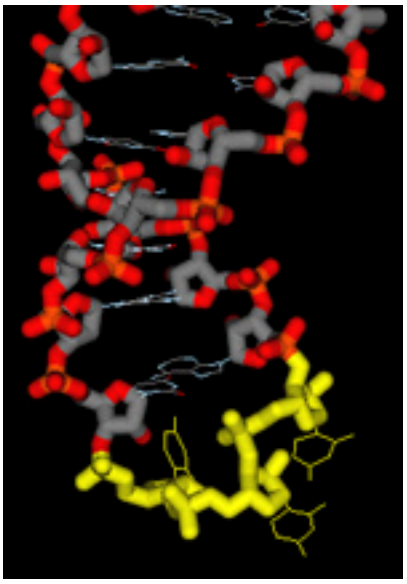
In a recent publication, researchers altered the aminoacylation identity of $tRNA^{Lys}_{CUU}$ so that it was charged with methionine rather than lysine. Both in live yeast cells and in isolated mitochondria, the engineered tRNA could enter the mitochondrion, where the radiolabelled methionine charged on the imported tRNA was incorporated normally into mitochondrial proteins. A second, modified $tRNA^{Lys}$ version with alanine identity was also successfully used *in vivo* to suppress an *amber* (UAG) stop codon (a nonsense mutation) in the mitochondrial *COX2* gene.

Defects in mitochondrial (mt) DNA, caused by base substitutions or rearrangements in genes that encode proteins or tRNAs underlie a range of human pathologies (as discussed in the previous highlight).

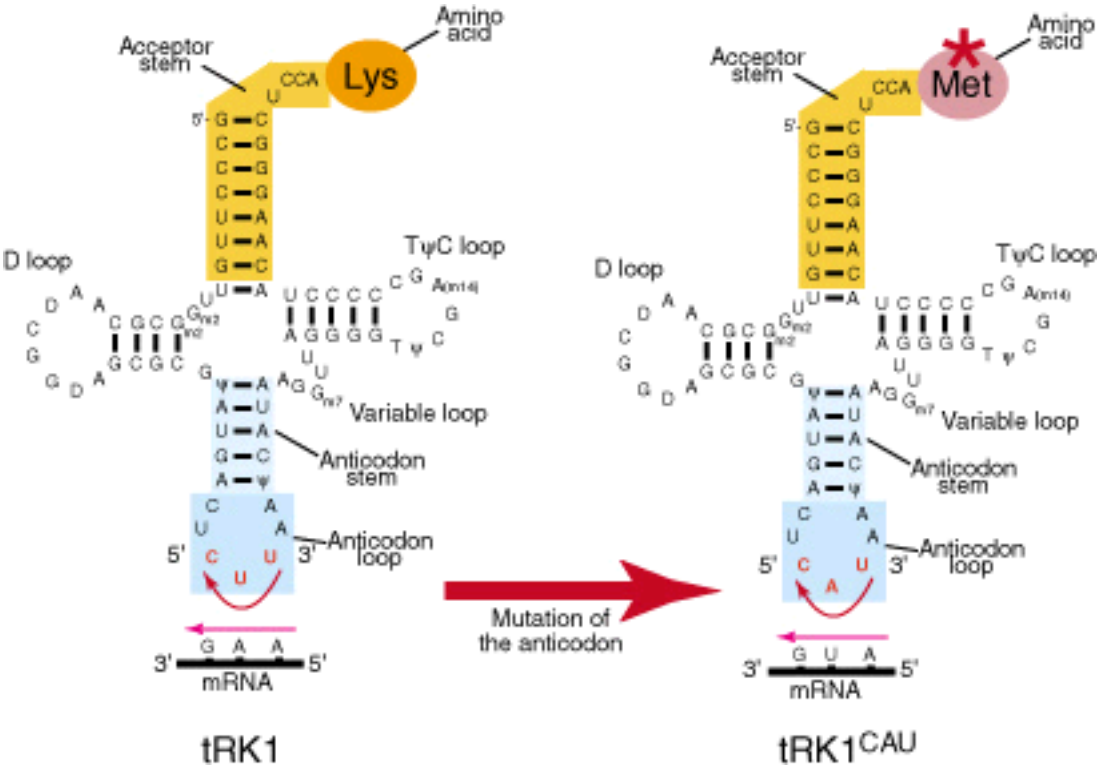
Could the technique used to modify mitochondrial mutations be adapted for use in humans, given that import of nuclear-encoded tRNAs into mammalian mitochondria has never been seen? It seems so, because isolated human mitochondria imported the yeast $tRNA^{Lys}_{CUU}$ and its derivatives, provided that the human cytosolic extracts were supplemented with the yeast pre-MSK. The foreign tRNA was functional on the translational apparatus of human mitochondria, just as in yeast.

This recent innovation might be useful for replacing non-functional tRNAs or for suppressing nonsense mutations in mtDNA.

Story contributed by Tanita Casci, Nature Reviews Genetics [http://www.nature.com/nrg/index.html]



Tertiary structure of tRNA The anticodon loop and stem of $tRNA^{Lys}$ is depicted in the image to the left. The three bases that compose the anticodon (in this example, "U-U-U") are highlighted in yellow.



Mutation of the anticodon of tRNA Outside of the mitochondria, $tRNA^{Lys}_{CUU}$ is charged with lysine, its cognate amino acid. However, by changing the anticodon from C-U-U to C-A-U, the mutated tRNA is subsequently charged with methionine. To be able to track mutant tRNAs, an ^{35}S-radiolabelled methionine was used. This experiment demonstrated that aminoacylation was necessary for transport of tRNA into the mitochondria, although the identity of the amino acid that was charged onto the tRNA was less important.

Important Links

Live PubMed searches

(1) Importation of tRNA

(2) Mitochondrial lysyl-tRNA

(3) REVIEWS

Additional NCBI resources

Genetic codes [http://www.ncbi.nlm.nih.gov/Taxonomy/Utils/wprintgc.cgi?mode=c]

Code chart (pdf) [http://www.ncbi.nlm.nih.gov/Coffeebreak/CB17_TRNA/stdcode.pdf]

Yeast genome [http://www.ncbi.nlm.nih.gov/PMGifs/Genomes/4932.html]

Yeast mitochondrion genome [http://www.ncbi.nlm.nih.gov/genomes/framik.cgi?gi=105&db=Genome]

Box: Search Organelle Genome Resources

Click on the link below to start an html tutorial.

Evolution and the mitochondrial gene order of tRNAs [http://www.ncbi.nlm.nih.gov/Coffeebreak/CB17_TRNA/org1.html]