

## Supplementary Methods

### Primer used for cloning of tRNAs:

tRNA<sup>Tyr</sup>: 5'-TAA TAC GAC TCA CTA TAG GTG GGG TTC CCG AG-3'  
5'-CCT GGT GGT GGG GGA AGG A-3'

tRNA<sub>f</sub><sup>Met</sup>: 5'-TAA TAC GAC TCA CTA TAG GCG GGG TGG AGC AGC C-3'  
5'-CCT GGT TGC GGG GGC CGG AT-3'

tRNA<sup>Met</sup>: 5'-TAA TAC GAC TCA CTA TAG GCT ACG TAG CTC AGT TG-3'  
5'-CCT GGT GGC TAC GAC GGG A-3'

### Primer for PCR amplification of tRNAs, followed by T7 transcription:

#### tRNA<sup>Tyr</sup>:

forward primer for all variants: 5'-TAA TAC GAC TCA CTA TAG GTG GGG TTC CCG AG-3'  
reverse primer for:

WT 5'- TGG TGG TGG GGG AAG GA-3'  
C74G 5'- TGC TGG TGG GGG AAG GA-3'  
C74A 5'- TGT TGG TGG GGG AAG GA-3'  
C74U 5'- TGA TGG TGG GGG AAG GA-3'  
C75G 5'- TCG TGG TGG GGG AAG GA-3'  
C75A 5'- TTG TGG TGG GGG AAG GA-3'  
C75U 5'- TAG TGG TGG GGG AAG GA-3'

#### tRNA<sup>Phe</sup>:

forward primer for all variants: 5'-TAA TAC GAC TCA CTA TAG-3'  
reverse primer for:

WT 5'- TGG TGC GCG GAC TCG GAA TC-3'  
C74G 5'- TGC TGC GCG GAC TCG GAA TC-3'  
C74A 5'- TGT TGC GCG GAC TCG GAA TC-3'  
C74U 5'- TGA TGC GCG GAC TCG GAA TC-3'  
C75G 5'- TCG TGC GCG GAC TCG GAA TC-3'  
C75A 5'- TTG TGC GCG GAC TCG GAA TC-3'  
C75U 5'- TAG TGC GCG GAC TCG GAA TC-3'

#### tRNA<sub>f</sub><sup>Met</sup>:

forward primer for all variants (C1G): 5'-TAA TAC GAC TCA CTA TAG GCG GGG-3'  
reverse primer for:

WT 5'-TGG TTG CGG GGG CCG -3'  
74U 5'-TGA TTG CGG GGG CCG G-3'  
A72C 5'-TGG TGG CGG GGG CCG GAT TTG-3'

#### tRNA<sup>Met</sup>:

forward primer: 5'-TAA TAC GAC TCA CTA TAG GCT ACG TAG CTC AGT TG-3'  
reverse primer: 5'-TGG TGG CTA CGA CGG GA-3'