

Supplementary Information

Table S1. Distribution of post-transcriptional modifications in mt-tRNAs from 10 phyla across phylogeny.

Nts	I Pro.	II Fun.	III Nem.	IV Mol.	V Ech.	VI Ins.	VII Uro.	VIII Bir.	IX Mam.	X Pla.	Σ
6	–	–	–	–	–	–	m ² G	–	m ² G	–	m ² G
8	–	–	m ¹ A	–	–	–	–	–	–	–	m ¹ A
9	–	–	m ¹ A	m ¹ A	m ¹ A m ¹ G	m ¹ A m ¹ G	m ¹ A	–	m ¹ A m ¹ G	–	m ¹ A, m ¹ G
10	–	m ² G	–	–	m ² G	m ² G	m ² G	–	m ² G	–	m ² G
13	–	–	–	–	D	–	–	–	–	Ψ	Ψ
15	–	D	–	–	–	–	–	–	–	–	D
16	D	–	–	D	m ¹ A	–	–	–	–	D	D, m ¹ A
17	D	–	–	–	–	–	–	–	–	D	D
18	–	–	–	–	–	–	–	–	–	Gm	Gm
20	D	D	–	–	D	–	–	–	D	–	D
20a	–	D	–	–	–	–	–	–	–	D	D
20b	–	–	–	–	–	–	–	–	–	acp ³ U	acp ³ U
25	–	–	–	Ψ	–	–	Ψ	–	–	–	Ψ
26	m ² G m ² ₂ G	m ² ₂ G	m ² G m ² ₂ G, Ψ	–	–	Ψ	–	–	m ² G m ² ₂ G	–	m ² G m ² ₂ G, Ψ
27	Ψ	–	Ψ	Ψ	Ψ	Ψ	Ψ	Ψ	Ψ	Ψ	Ψ
28	–	–	Ψ	–	Ψ	Ψ	–	–	Ψ	–	Ψ
29	–	Ψ	Ψ, t ⁶ A	–	–	–	–	–	Ψ	–	Ψ, t ⁶ A
31	Ψ	Ψ	Ψ	–	–	Ψ	–	–	Ψ	–	Ψ
32	Ψm Cm	Ψ	Ψ	Ψ	Ψ, m ³ C, m ⁵ C	Ψ	Ψ	–	Ψ, m ³ C	–	Ψ, m ³ C, Cm
33	s ² U	–	–	–	–	–	–	–	–	–	s ² U
34	Ψ, t ⁶ A cmnm ⁵ U cmnm ⁵ s ² U Um, Cm,	cmnm ⁵ U	Ψ, f ⁵ C cmnm ⁵ U cmnm ⁵ s ² U	m ⁷ G	m ⁷ G, Q	f ⁵ C, Q	τm ⁵ U τm ⁵ s ² U (s ² U)	f ⁵ C	f ⁵ C, Q τm ⁵ U τm ⁵ s ² U	I, k ² C	Ψ, t ⁶ A, f ⁵ C, m ⁷ G, Q, cmnm ⁵ U, τm ⁵ s ² U cmnm ⁵ s ² U, τm ⁵ U Um, Cm, I, k ² C
35	–	–	–	–	Ψ	–	–	–	–	–	Ψ
37	i ⁶ A t ⁶ A m ¹ G	i ⁶ A	ms ² i ⁶ A m ¹ G t ⁶ A	t ⁶ A	i ⁶ A t ⁶ A, m ⁶ t ⁶ A m ¹ G	i ⁶ A, t ⁶ A ms ² i ⁶ A	ms ² i ⁶ A	–	i ⁶ A, t ⁶ A ms ² i ⁶ A m ¹ G	m ⁶ A m ¹ G	i ⁶ A, m ⁶ A, t ⁶ A, m ⁶ t ⁶ A, ms ² i ⁶ A, m ¹ G

Table S1. Cont.

Nts	I Pro.	II Fun.	III Nem.	IV Mol.	V Ech.	VI Ins.	VII Uro.	VIII Bir.	IX Mam.	X Pla.	Σ
38	–	–	–	–	Ψ, ms ² i ⁶ A	Ψ	Ψ	–	–	Ψ	Ψ, ms ² i ⁶ A
39	Ψ, Ψ _m	Ψ	–	Ψ	Ψ	Ψ, Ψ _m , Cm	Ψ	–	Ψ	Ψ	Ψ, Ψ _m , Cm
40	–	Ψ	–	–	Ψ	–	Ψ	–	Ψ	–	Ψ
41	Ψ	Ψ	–	–	–	Ψ	–	–	–	–	Ψ
44	Um	Um	–	–	–	Ψ	–	–	–	Um	Um
45	–	–	–	–	–	D	–	–	–	–	D
46	–	–	–	m ⁷ G	–	–	–	–	–	m ⁷ G	m ⁷ G
47	–	–	–	D	–	–	–	–	–	acp ³ U	D, acp ³ U
48	–	–	–	m ⁵ C	–	–	–	–	m ⁵ C	–	m ⁵ C
49	m ⁵ C	–	–	–	–	–	–	–	m ⁵ C	m ⁵ C	m ⁵ C
50	–	–	–	–	–	–	–	–	Ψ	–	Ψ
54	T	T	–	T	Ψ	–	–	T	–	T	T, Ψ
55	Ψ	Ψ	–	Ψ	Ψ	–	–	Ψ	Ψ	Ψ	Ψ
57	–	–	–	–	m ¹ A	–	–	–	Ψ	–	Ψ
58	–	–	–	–	–	–	–	–	m ¹ A	m ¹ A	m ¹ A
64	–	–	–	–	–	–	Ψ	–	–	–	Ψ
65	–	–	–	–	–	Ψ	–	–	–	–	Ψ
66	–	–	–	–	–	Ψ	–	–	–	–	Ψ
67	–	–	–	m ² G	–	Ψ	–	–	Ψ	–	m ² G, Ψ
68	–	–	–	–	–	Ψ	–	–	–	–	Ψ
70	–	–	Ψ	–	–	–	–	–	–	–	Ψ
71	–	–	Ψ	–	–	–	–	–	–	–	Ψ
72	–	–	–	m ⁵ C	–	–	–	–	m ⁵ C	–	m ⁵ C
Nb Seq.	4	25	10	4	7	16	6	1	35	15	123

Most data are retrieved from *tRNADB* data base [125] and additional data (in blue) are taken from refs [50,108,193,194,1,2]. The number of RNA sequences of mt-tRNAs investigated in 10 eukaryotic phyla is indicated. Modified nucleosides (nts) are found in Acceptor stem (nts 6 and 66–72), D-arm with connecting residues (nts 8–26), Anticodon loop (nts 32–38, on yellow background), Anticodon stem (nts 27–31 and 39–41, on light yellow background), Variable region (nts 44–48) and T-arm (nts 49–65); I Pro., unicellular protozoans including alveolates, amoebas and euglenes (F, K, L, W, Y; *Leishmania tarentolae*, *Tetrahymena pyriformis*, *T. thermophila*, *Trypanosoma brucei*); II Fun., fungi (All but five—*C*, *D*, *E*, *N*; *Neurospora crassa*, *Saccharomyces cerevisiae*); III Nem., nematodes (F, E, K, L, M, Q, R, S, W; *Ascaris suum*); IV Mol., molluscs (K, S; *Loligo bleckeri*); V Ech., echinoderms (D, H, K, N, S, Y; *Asterias amurensis*); VI Ins., insects (D, E, G, I, K, M, N, Q, R, S, V, W; *Aedes albopictus*, *Drosophila melanogaster*); VII Uro., urochordates (G, M, W; *Halocynthia roretzi*); VIII Bir., birds (M; *Gallus gallus*); IX Mam., mammals (All; *Bos taurus*, *Didelphis virginiana*, *Homo sapiens*, *Mesocricetus auratus*, *Rattus norvegicus*); X Pla., plants (F, I, L, M, P, R, W, Y; *Oenothera*, *Phaseolus vulgaris*, *Physcomitrella patens*, *Solanum tuberosum*, *Triticum aestivum*). Σ The represented coding specificities are indicated by the amino acid one-letter code (missing specificities are in italics) and are scattered in the indicated species. § Data on *T. brucei* mt-tRNAs (K, L, W) [50] concern imported tRNAs modified in mitochondria to Cm32.

References

1. Tomita, K.; Ueda, T.; Watanabe, K. 7-Methylguanosine at the anticodon wobble position of squid mitochondrial tRNA^{Ser(GCU)}: Molecular basis for assignment of AGA/AGG codons as serine in invertebrate mitochondria. *Biochim. Biophys. Acta* **1998**, *1399*, 78–82.
2. Tomita, K.; Ueda, T.; Ishiwa, S.; Crain, P.F.; McCloskey, J.A.; Watanabe, K. Codon reading patterns in *Drosophila melanogaster* mitochondria based on their tRNA sequences: A unique wobble rule in animal mitochondria. *Nucleic Acids Res.* **1999**, *27*, 4291–4297.