Novel taurine-containing uridine derivatives and mitochondrial human diseases

doi: 10.1093/nass/1.1.257

Nucleic Acids Research Supplement No. 1 257-258

Novel taurine-containing uridine derivatives and mitochondrial human diseases

Tsutomu Suzuki[1,2,*], Takeo Suzuki[2,*], Takeshi Wada[1,2], Kazuhiko Saigo[1,2] and Kimitsuna Watanabe[1,2]
[1]Dept of Integrated Biosciences, Graduate School of Frontier Sciences, [2]Dept of Chemistry and Biotechnology, Graduate School of Engineering, University of Tokyo, 5-1-5 Kashiwanoha, Kashiwa, Chiba Prefecture, 277-8562, Japan *These authors contributed equally to this work.

ABSTRACT

Two novel modified uridines were identified from mammalian mitochondrial (mt) tRNAs. Mass spectrometric analysis revealed that they are modified uridines possessing a sulfonic acid group derived from taurine; 5-taurinomethyl-uridine from mt tRNAs for Trp and Leu(UUR), and 5-taurinomethyl-2-thiouridine from mt tRNAs for Lys, Gln and Glu. We have found lack of modification of these taurine-containing uridines in mutant mt tRNAs for Leu(UUR) and Lys from pathogenic cells of mitochondrial encephalomyopathies, MELAS and MERRF, respectively.

INTRODUCTION

A characteristic structural feature of tRNA is the presence of post-transcriptionally modified nucleosides in the anticodon first position (wobble position), which participate in codon–anticodon pairing. It is known that mammalian mitochondrial decoding system requires 22 mitochondrial (mt) tRNA species, which is the minimum set capable of decoding all the sense codons throughout all living organisms and organelles (1). According to the mitochondrial wobble rule, mt tRNAs with unmodified uridine at the wobble position are able to decode all four codons in the family box, so that, a single species of such tRNAs is responsible for decoding each family box (2). In two-codon sets, it is known that purine-ending codons are read by tRNAs possessing modified uridines with unknown structures. However, it was shown that only AUR (R; purine) codons were read by mt $tRNA^{Met}$ with 5-formyl cytidine (f^5C) at the wobble position(3). Thus, post-transcriptional modification at the anticodon wobble position plays a significant role to maintain translational efficiency and accuracy in mammalian mitochondria. Recently, we discovered that mt $tRNA^{Leu}$ and $tRNA^{Lys}$ respectively obtained from human pathogenic cells of the mitochondrial encephalomyopathies MELAS and MERRF are deficient in the normal uridine modification at the wobble position (4,5). Since unmodified uridine could cause either misreading on the basis of mitochondrial wobble rule or decoding deficiency, it has been proposed that a decoding disorder resulted from the modification defect might be one of the main causes of mitochondrial dysfunction. The chemical structures of the unknown modified uridines in mammalian mt tRNAs should be required to comprehend the minimum decoding system in its entirety, and to investigate the molecular mechanism of pathogenesis of mitochondrial encephalomyopathies.

RESULTS AND DISCUSSION

Analysis by LC/MS using an ESI/iontrap mass spectrometer revealed that the two previously unknown nucleosides are a uridine derivative (U*: molecular mass, 381 Da) in the tRNAs for Leu(UUR) and Trp, and its 2-thio derivative (s^2U*: 397 Da) in the tRNAs for Lys, Gln and Glu. The atomic composition of s^2U*

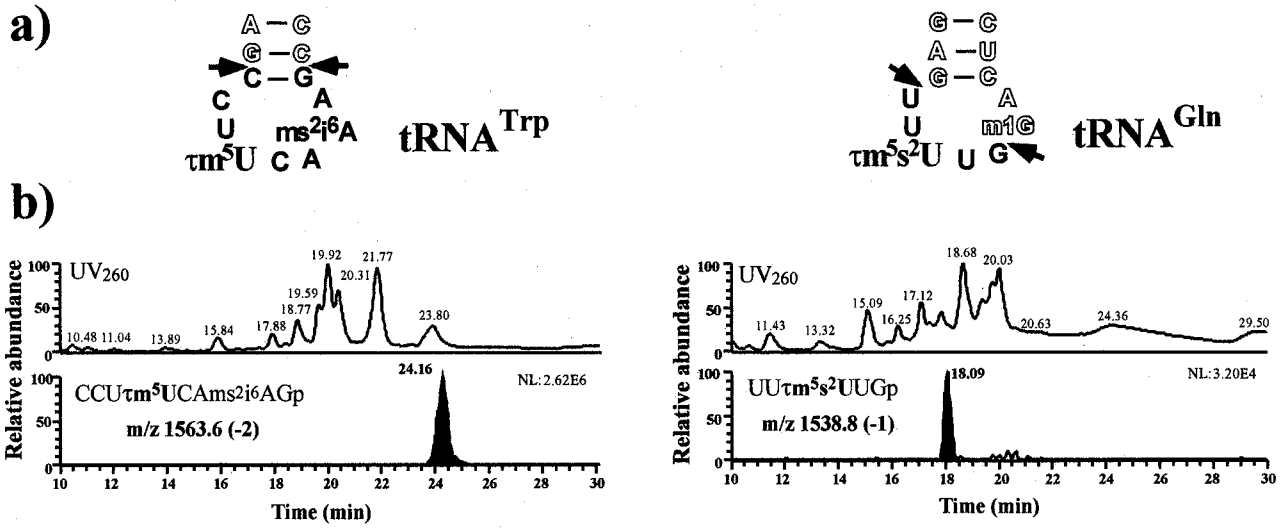

Fig.1 Mass spectrometric analysis of RNA fragments having novel taurine-containing uridines a) RNase T1 cleavage sites of mt tRNAs for Trp and Gln. b) Mass chromatogram for RNase T1 digests of each tRNA.

was determined to be $C_{12}H_{19}N_3O_8S_2$ by high-resolution FT-ICR mass spectrometry. These findings taken together with results of NMR and fragmentation analysis using collision-induced dissociation (CID) indicated that the modification occurs at position 5 of the uracil base, with the most plausible structure in both cases being a taurinomethyl possessing a sulfonic acid group derived from taurine. Hence, the two nucleosides were named 5-taurinomethyluridine (τm^5U) and 5-taurinomethyl-2-thiouridine (τm^5s^2U), the chemical structure of which are verified by comparison with the synthetic τm^5U. As shown in Fig. 1, each taurine-containing uridine was identified in the RNA fragment including anticodon region.

This is the first reported case of any modified nucleoside possessing a sulfonic acid group in a side chain. The decoding properties of these nucleosides in relation to the strong electrostacity of sulfonic acid should be clarified in future work. From our results, it can be concluded that only four kinds of modification (τm^5U, τm^5s^2U, Q and f^5C) are required to discriminate all sense codons in the minimum decoding system. Since we have also found these modified uridines in human and ascidian mt tRNA counterparts, it is likely that they are common to vertebrate and protochordate mitochondria (4, 6).

Further study of the biosynthesis of these nucleosides will shed light on the new metabolic pathway of taurine and, hopefully, on therapeutic measures for mitochondrial diseases.

Novel taurine-containing uridines

REFERENCES

1. Osawa, S. *Evolution of the Genetic Code* (1995) Oxford University Press, Oxford.
2. Watanabe, K. & Osawa, S. (1995) in *tRNA: Structure, Biosynthesis, and Function* (eds Söll, D. & RajBhandary, U. L.) 225 (American Society for Microbiology, Washington, D.C.).
3. Moriya, J., Yokogawa, T., Wakita, K., Ueda, T., Nishikawa, K., Crain, P. F., Hashizume, T., Pomerants, S. C., McCloskey, J. A., Kawai, G., Hayashi, N., Yokoyama, S. & Watanabe, K. (1994) *Biochemistry*, **33**, 2234.
4. Yasukawa, T., Suzuki, T., Suzuki, T., Ueda, T., Ohta, S. & Watanabe, K. (2000) *J. Biol. Chem.*, **275**, 4251.
5. Yasukawa, T., Suzuki, T., Ishii, N., Ueda, T., Ohta, S. & Watanabe, K.(2000) *FEBS Lett.*, **467**, 175.
6. Kondow, A., Suzuki, T., Yokobari, S., Ueda, T. & Watanabe, K. (1999)*Nucleic Acids Res.*, **27**, 2554.