In Silico Exploration of the Structural Impact of Modifications on the Fine Structure of Trna via MD Simulations

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Transfer RNA (tRNA) is the most diversely modified RNA. Although the strictly conserved purine position 37 in the anticodon stem loop (ASL) undergoes a wide assortment of modifications that are phylogenetically distributed, we do not yet fully understand the roles of these modifications. To provide molecular-level details for how such modifications impact the structure and function of tRNA, molecular dynamics (MD) simulations are used to compare the structural dynamics of unmodified and modified tRNAs. A focus is placed on three hypermodified base families that include the parent i6A, t6A, and yW modifications, as well as derivatives formed through the incorporation of additional chemical substituents (such as a hydroxy, methyl, thiomethyl or peroxy group). Our data reveal that the hypermodifications exhibit significant conformational flexibility in tRNA, which can be modulated by additional chemical functionalization. Regardless of the modification family or level of chemical substitution, the hypermodifications do not affect the global tRNA three-dimensional structure or the domain-domain interactions, suggesting that there are no long-range effects of modifying the 37th position. Although the overall structure of the anticodon stem remains intact regardless of the modification considered, the anticodon loop must rearrange to accommodate the bulky, dynamic hypermodifications, which includes changes in the nucleotide glycosidic and backbone conformations and enhanced or completely new nucleobase–nucleobase interactions compared to unmodified tRNA or tRNA containing smaller (m1G) modifications at the 37th position. Importantly, the extent of the changes in the anticodon loop is influenced by the addition of small functional groups to parent modifications, implying each substituent can further finetune tRNA structure. Although the dominant conformation of the ASL is achieved in different ways for each modification, the molecular features of all modified tRNA drive the ASL domain to adopt the functional open-loop conformation. Importantly, the impact of the hypermodifications is preserved in different sequence contexts. These findings highlight the likely role of these modifications in regulating mRNA structure and translation.