

W0410

Crystal Structures, Reactivity and Inferred Acylation Transition States for 2'-Amine Substituted RNA. C.M. Gherghe, J.M. Krahn, K.M. Weeks, Dept. of Chemistry, Univ. of North Carolina, Chapel Hill, NC 27599.

Nucleic acid ribose 2'-amine substitutions are broadly useful as structural probes. In addition, selective chemical reaction with 2'-NH₂ substituted nucleotides using an activated ester robustly and site-selectively maps local nucleotide flexibility in RNA and DNA. We determined four RNA crystal structures of self-complementary duplexes containing 2'-amino cytidine (C^N) residues that form either canonical C^N-G base pairs or C^N-A mismatches. The 2'-NH₂ substitution causes no significant structural perturbation to the RNA duplexes. A 2'-amide product structure (see Figure) was visualized directly by acylating the C^N-A mismatch in intact crystals and is readily accommodated within an A-form geometry. To visualize conformations able to facilitate formation of the amide-forming transition state, in which the 2'-amine carries a large positive partial charge, we obtained crystals of the C^N-A duplex at pH 5, where the 2'-NH₂ is protonated. The protonated amine moves to form a strong electrostatic interaction with the 3'-phosphodiester. Taken together with solution phase biochemistry, the structural data suggest that 2'-NH₂ acylation proceeds via either of two transition states, both involving precise positioning of atoms at the adjacent 3'-phosphodiester group.

