

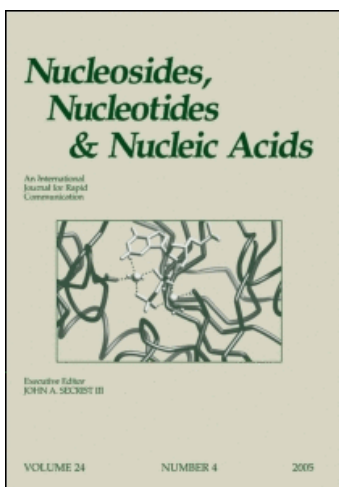
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THE CONFORMATIONAL SPACE OF NASCENT PEPTIDE-ACCEPTING 3'-AMINOACYL RIBONUCLEOS(T)IDES: ¹H NMR DATA AND *AB INITIO* CALCULATIONS OF PUROMYCIN AND SOME SYNTHETIC ANALOGS

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Puromycin is a codon-independent acceptor of nascent peptide chains in ribosomes and is widely used *in vitro* as protein growth terminator and, more recently, as the crucial 3'-terminus of synthetic combinatorial mRNA libraries for the *in vitro* synthesis and selection of covalent protein-mRNA fusion molecules [1].

Now that the structural details of a 50 S ribosomal subunit advanced to 2.4 Å resolution [2] a more reliable knowledge of the ribosomal peptidyl transfer reaction seems within close reach. Studies on the intrinsic conformations of the peptide-accepting 3'-terminus of aminoacyl-tRNA (aa-tRNA) and puromycin using modern spectroscopic and computational techniques should allow for a better understanding of the 3D course of this reaction.

New puromycin analogs were synthesised¹ and tested² for *in vitro* peptide release activity. pH- and T-dependent ¹H NMR spectra¹ in H₂O let us gain insight into the conformational preferences of 3'-aminoacylamino-3'-deoxyadenosine (3'-aa-NH-3'-dA) derivatives. The experimental data was supplemented with a systematic *ab initio* study on 8 conformers of 3'-Ala-NH-3'-dA and on related 3'-amide and 3'-ester variants including 5'-O- and/or 2'-O-methyl, 5'-O-methylphosphate, and

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2'-fluoro groups (42–49 atoms, 24–29 non-H, calculated with *Gaussian 98* using HF, B3LYP, MP2 methods and 6-31G**, 6-31 + G** and 6-311 ++ G** basis sets).

Consistent with the NMR experiments, the calculations suggest that 3'-aminoacyl *amides* with free 5'-hydroxyls as well as the 2'-fluoro-2'-deoxypuromycin analogs prefer a ribofuranose pucker bearing a Ψ -equatorial extension of the aminoacyl moiety (North type sugar pucker) and *syn* periplanar orientation of C=O and α -NH₂ rel. to H-C3', while in *esters* such as 3'-O-alanyladenine-5'-O-methylphosphate the competing (South type) pucker bearing a Ψ -axial extension is of similar stability.

Since the governing stabilising interactions in all found conformers are intramolecular H-bridges, we conclude that the present understanding of ribonucleoside conformation equilibria through the balancing effect between the X3'/C5' *gauche* effect and the N9 anomeric effect needs to be reconsidered. The preference for intra- vs. intermolecular H-bridges is not only a possible competing phenomenon, but also a likely means of control by the ribosomal peptidyl transferase pocket. A 3D reactional pathway of the aa-tRNA \Rightarrow pept-tRNA addition begins to emerge.

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REFERENCES

1. a) Nemoto, N.; Miyamoto-Sato, E.; Husimi, Y.; Yanagawa, H. *FEBS Letters* **1997**, *414*, 405–408; b) Roberts, R.W.; Szostak, J.W. *Proc. Natl. Acad. Sci. USA*, **1997**, *94*, 12297–12302; c) Liu, R.; Barrick, J.E.; Szostak, J.W.; Roberts, R.W. *Methods Enzymol.* **2000**, *318*, 268–93.
2. a) Ban, N.; Nissen, P.; Hansen, J.; Moore, P.B.; Steitz, T.A.. *Science* **2000**, *289*, 905; b) Nissen, P.; Hansen, J.; Ban, N.; Moore, P.B.; Steitz, T.A.. *Science* **2000**, *289*, 920.



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