

SYNTHESIS AND BIOLOGICAL ACTIVITY OF A TUBERCIDIN ANALOG OF 2-5A

Jean-Claude Jamouille, Jiro Imai and Paul F. Torrence
 Laboratory of Chemistry, National Institute of Arthritis, Diabetes and
 Digestive and Kidney Diseases. U.S. National Institutes of Health,
 Bethesda, Maryland 20205

Previous studies from this laboratory have demonstrated that oligonucleotide binding to the 2-5A-dependent endonuclease (RNase L) of mouse L cells decreases dramatically upon substitution of the adenine rings of 2-5A by uracil, cytosine, or hypoxanthine. To explore the structural requirements for base binding more closely, we prepared the tubercidin analog of 2-5A [ppp5'-(c⁷A)2'p5'(c⁷A)2'p5'(c⁷A)] by lead ion-catalyzed polymerization of 5'-phosphoroimidazolide of c⁷A to give 2',5'-(pc⁷A)₃ which was converted to the triphosphate via pyrophosphate displacement on the phosphoroimidazolide. This material and intermediates were thoroughly characterized by enzymatic digestions and NMR. The 5'-monophosphate, 2',5'(pc⁷A)₃, as well as the 5'-triphosphate, 2',5'-pp(pc⁷A)₃, were bound to RNase L of mouse L cells approximately as well as 2-5A itself as judged by displacement of radiolabeled ppp5'A2'p5'A2'p5'A2'p5'A3'p5'Cb from the nuclease. Nonetheless, the 5'-triphosphate, 2',5'-pp(pc⁷A)₃ did not activate RNase L as judged by its ability to inhibit protein synthesis in encephalomyocarditis viral RNA-programmed extracts of L cells under conditions where 2-5A caused a 50% inhibition of translation at 10⁻⁹M. These results suggest that while the N-7 atoms of the adenine rings of 2-5A may not be involved in binding to RNase L, they are critical for the activation of the enzyme.

