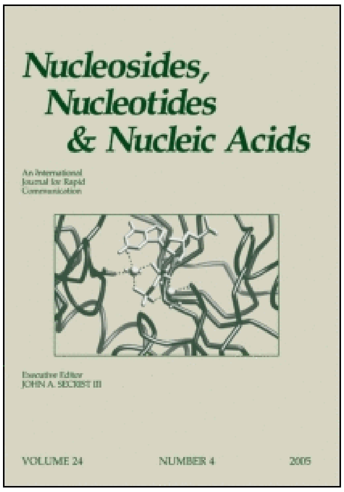

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SYNTHESIS OF P-THIOADENYLYL (2'-5') ADENOSINE AND P-THIOADENYLYL (2'-5')-P-THIOADENYLYL-(2'-5')-ADENOSINE

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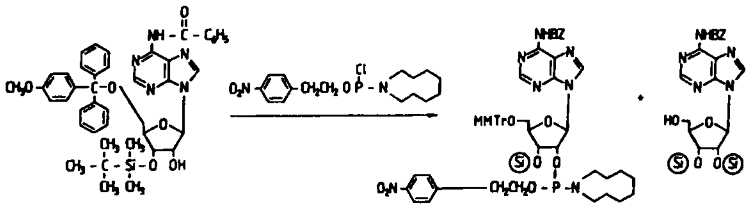
Summary: Dimer and trimer adenylates with 2'-5' phosphorothioate linkages were synthesized via the phosphoramidite method using p-nitrophenylethyl group for phosphate protection and followed by sulfur oxidation. The various diastereoisomers were separated and characterized.

In recent years, analogues of the antiviral and antitumor agent pppA2'p5'A2'p5'A (2-5A) and its core have been subject of several syntheses (1). It is also well established mainly due to the pioneering work of Eckstein et al. (2) that phosphorothioate analogues of oligonucleotides, which contain at a specific location a chiral phosphorothioate instead of the naturally occurring phosphodiester bond, are valuable compounds for elucidation of certain stereochemical aspects of the mechanism of the enzyme catalysed phosphoryl and nucleotidyl transfer reactions. An analogue of 2-5A core such as phorphorothiate, with added chirality of the two phosphorothioate groups which result four stereoisomers, may prove beneficial as mechanistic probes of the action of 2-5 A.

Apart from that 2-5 A is rapidly digested by a phosphodiesterase which shortened its duration of action, the phosphorothioate group is more stable to hydrolysis by phosphodiesterase. P. Nelson (3) et al. have recently published the synthesis of dimer and trimer phosphorothioate using the phosphite triester method.

We successfully achieved the syntheses of four stereoisomeric P-thioadenylyl-(2'-5')-P-thioadenylyl-(2'-5')-adenosine starting from N^6-benzoyl-3'-O-tert.butyldimethylsilyl-5'-O-monomethoxytrityladenosine 1 (4). Treatment with chloro-p-nitrophenylethoxy-N-octahydroazoninophosphane (5), in presence of Hünig's base in dry CH_2Cl_2 gave the corresponding 2'-phosphoramidite 2, which could be purified by column chromatography in good yield and could be stored at -10°C for several months. This was then condensed with N^6-benzoyl-2',3'-di-O-tert.butyl-dimethylsilyladenosine 3, in presence of

3-nitro-1,2,4-triazole and subsequent oxidation by sulfur to give the dimer. Chromatographical separation yielded the two diastereomers 4A and 4B, in 1:0.71 ratio in 39 and 28 % yields respectively. Each of them was detritylated with p-TsOH to give the corresponding 5'-hydroxy compounds 5A ans 5B which were then condensed with phosphoramidite 2 as described for the dimer, to give two sets of trimeric pairs of diastereomers. Resolution and purification of the individual trimers 6A, 6B and 7A and 7B were achieved by silicagel chromatography and characterized by physical means.



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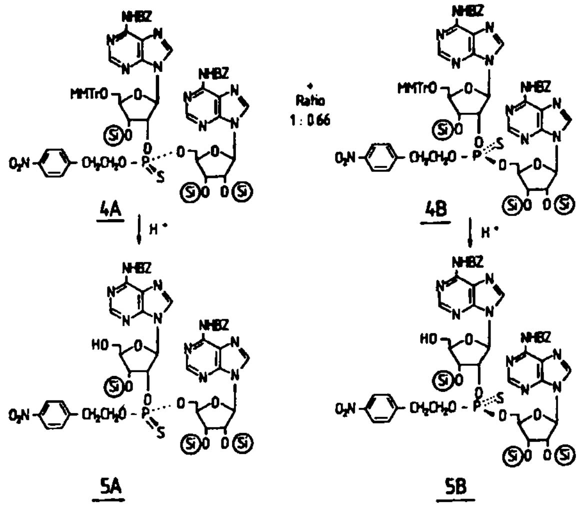
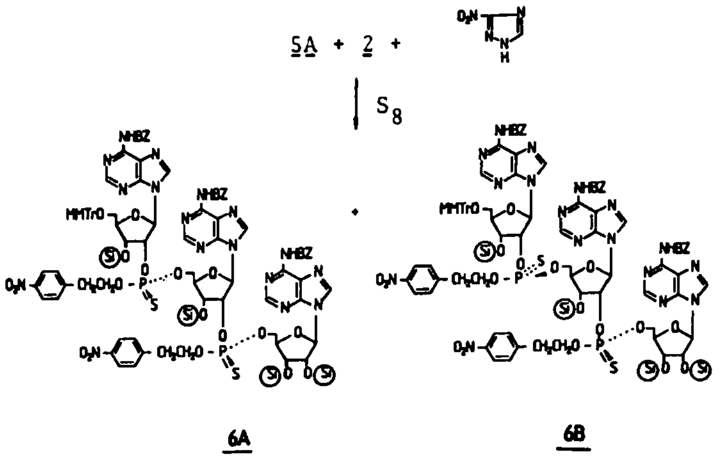
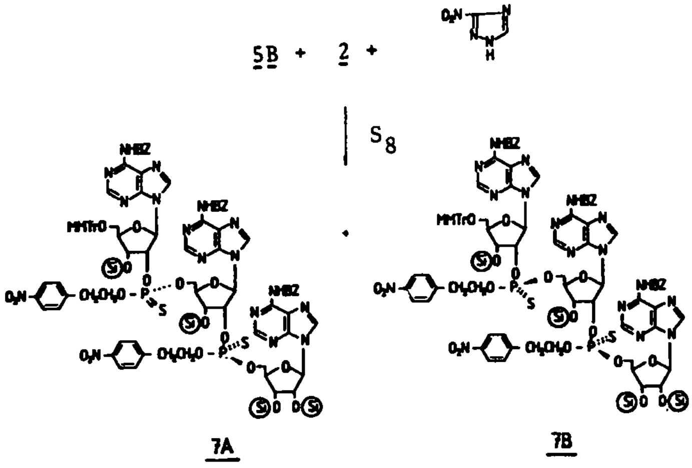
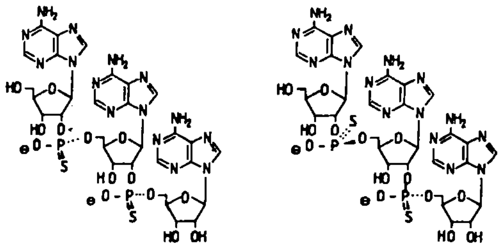


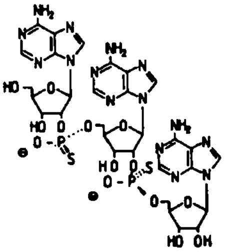
The dimer diastereomers and the trimer diastereomers were deblocked using 0.5 M DBU in pyridine, to remove the p-nitrophenylethyl groups and conc. ammonia to cleave benzoyl group, 1 M TBAF in THF to deprotect the silyl function and finally 80 % AcOH to

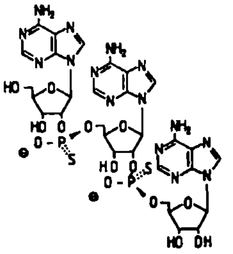




deblock the 5'-MMTr group. The crude product was purified by DEAE Sephadex A 25 column chromatography using a linear gradient of TEAB buffer. Each isomer was found to be pure according to cellulose, and PEI cellulose chromatography, HPLC and ^{31}P-NMR.







The absolute configurations of the isomers were determined by enzymatic studies.

REFERENCES

1. R. Charubala and W. Pfleiderer, Tetrahedron Lett. 1982, 4789;
   b) R. Charubala and W. Pfleiderer, Tetrahedron Lett. 1980, 4077;
   c) J. Engels, Tetrahedron Lett. 1980, 4339;
   d) M. Kwiatkowski, C. Gioeli, J.B. Chattopadhyaya, Chemica Scripta 19, 49 (1982);
   e) R. Crea, J.Org. Chem. 46, 2242 (1981);
   f) G. Gosselin, J.L. Imbach, Tetrahedron Lett. 1981, 4699;
   g) D.A. Eppstein, Y.V. Marsh, B.B. Schryver, M.A. Larsen, J.W. Barnet, J.P.H. Verheyden, E.J. Prisbe, J.Biol.Chem. 257, 13390 (1982);
2. F. Eckstein, Angew.Chem. 95, 431 (1983); Angew.Chem.Int.Ed. Engl. 22, 423 (1983);
3. P.S. Nelson, C.T. Bach and P.H. Verheyden, J.Org.Chem. 49, 2314 (1984);
4. F. Flockerzi, G. Silber, R. Charubala, W. Schlosser, R.S. Varma, F. Creegan, and W. Pfleiderer, Liebigs Ann.Chem. 1981, 2392;
5. M.W. Schwarz and W. Pfleiderer, Tetrahedron Lett. 1984, 5315.