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## Nucleosides, Nucleotides and Nucleic Acids

Publication details, including instructions for authors and subscription information:

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### Hydrolytic Reactions of an RNA Dinucleotide Containing a 3'-S-Phosphorothiolate Linkage

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**To cite this Article** Elzagheid, Mohamed , Oivanen, Mikko , Colin, B. , Jones, N. M. , Cosstick, Richard and Lönnberg, Harri(1999) 'Hydrolytic Reactions of an RNA Dinucleotide Containing a 3'-S-Phosphorothiolate Linkage', *Nucleosides, Nucleotides and Nucleic Acids*, 18: 6, 1265 – 1266

**To link to this Article:** DOI: 10.1080/07328319908044687

**URL:** <http://dx.doi.org/10.1080/07328319908044687>

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## HYDROLYTIC REACTIONS OF AN RNA DINUCLEOTIDE CONTAINING A 3'-S-PHOSPHOROTHIOLATE LINKAGE

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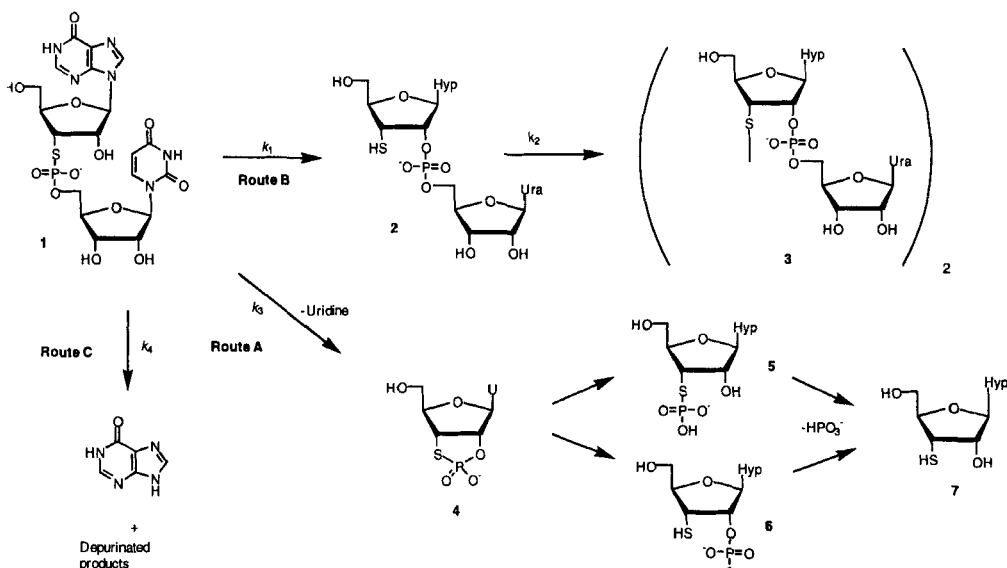
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**ABSTRACT:** The pH-rate profiles (pH 0.2 to 9 at 90° C) for the competing hydrolytic reactions of (3'-deoxy-3'-thioinosyl)-3',5'-uridine (IspU) have been determined by an HPLC method.

Nucleoside phosphorothioates are well known and widely used analogs of nucleoside phosphoesters in nucleic acid chemistry and biochemistry, especially in studying the mechanisms of cleavage of RNA by enzymes and ribozymes. More recently, nucleoside phosphorothiolate diesters having a bridging oxygen replaced by sulfur, have received considerable interest for related purposes. Since this kind of use of phosphorothiolates as mechanistic probes requires a detailed knowledge of the effects of the structural modification on the intrinsic chemical reactivity, we have now studied the kinetics and mechanisms of the non-enzymatic hydrolytic reactions of an RNA dinucleoside analog, (3'-deoxy-3'-thioinosyl)-3',5'-uridine **1** (IspU).<sup>1</sup> The pH-rate profiles (pH 0.2 to 9 at 90°C) for the competing reactions were determined by HPLC methods described previously<sup>2</sup>.

Under alkaline conditions (pH > 8.5), only one reaction takes place: cleavage of the phosphorothiolate linkage results in the accumulation of uridine and 3'-thioinosine-3'-monophosphorothiolate as the only detectable products. The cleavage is first-order in [OH<sup>-</sup>]. Under these conditions, IspU is degraded more than 2 orders of magnitude faster than the native dinucleoside monophosphates (**Route A**).

On going to neutral conditions, isomerization of IspU to its 2',5'-phosphorodiester counterpart starts to compete with the cleavage of the phosphorothiolate linkage.



The rate of isomerization is pH-independent at  $\text{pH} > 3$ , and this is the only reaction detected at  $\text{pH} 3 - 6$ . No sign of the reverse reaction was detected, showing that the inosine 3'-thio function does not act as a nucleophile towards phosphorus. Instead, the 2',5'-dimer formed tends to dimerize (*i. e.* to form a "tetranucleoside") by disulfide formation between two inosine 3'-mercapto groups (**Route B**). Isomerization of IspU is about 60 times as fast as the pH-independent isomerization of 3',5'-UpU to 2',5'-UpU<sup>2</sup>.

Under acidic conditions at  $\text{pH} < 3$ , the product mixture of hydrolysis of IspU is more complicated. Both the diester cleavage and isomerization appear to be acid-catalyzed (first-order in  $[\text{H}^+]$ ), and even acid-catalyzed depurination of the inosine moiety competes with the reactions of the phosphorothiolate function (**Route C**).

## REFERENCES

1. Weinstein, B.; Earnshaw, D.J.; Cosstick, R.; Cech, T.R. *J. Am. Chem. Soc.* **1996**, 118, 10341-10350.
2. Järvinen, P.; Oivanen, M.; Lönnberg, H. *J. Org. Chem.* **1991**, 56, 5396-5401.