SUPPLEMENTAL INFORMATION

A Non-Natural Nucleoside with Combined Therapeutic and Diagnostic Activities against Leukemia

Edward A. Motea[a], Irene Lee[a], and Anthony J. Berdis[b,#]

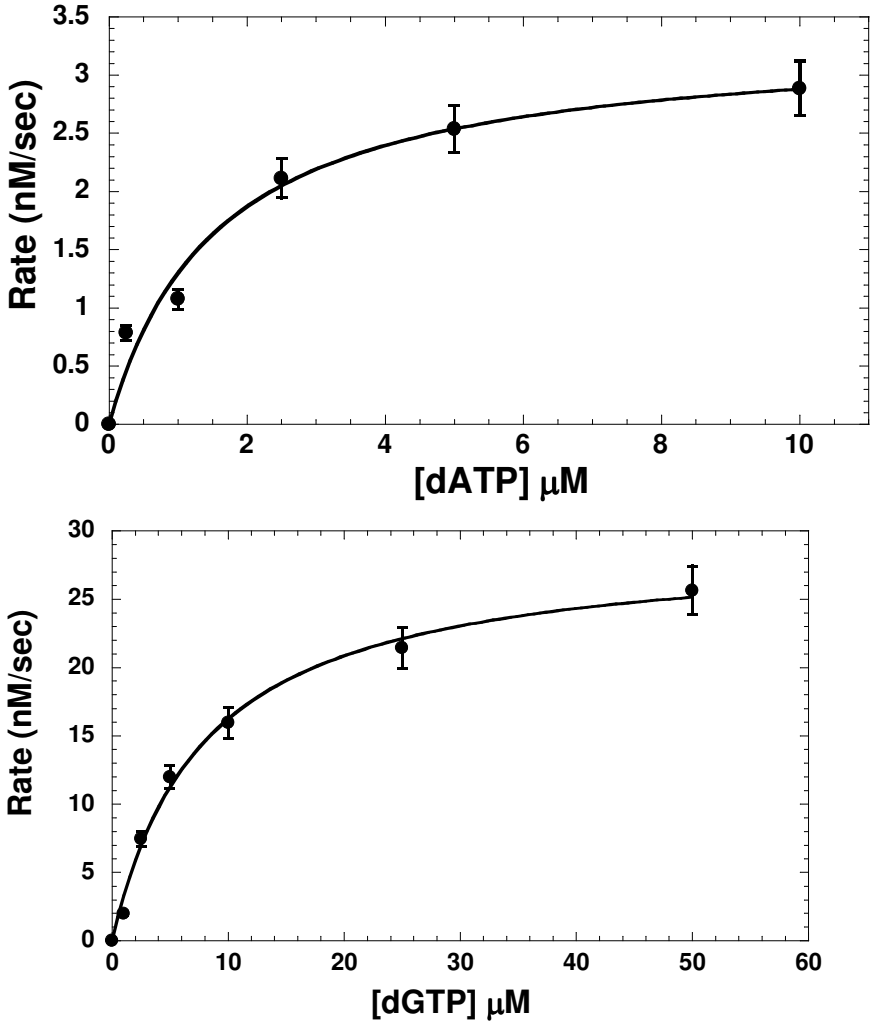
Departments of [a]Chemistry and [b]Pharmacology

Case Western Reserve University, 10900 Euclid Avenue, Cleveland, Ohio 44106

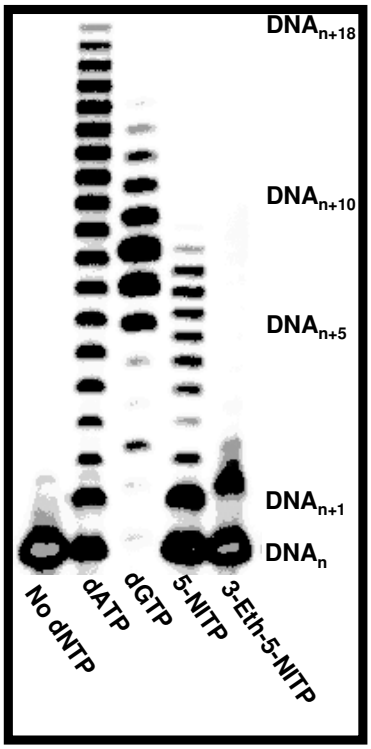
[#]***Corresponding Author Information:*** Telephone (216)-368-4723, Fax (216) 368-3395,

e-mail: ajb15@cwru.edu

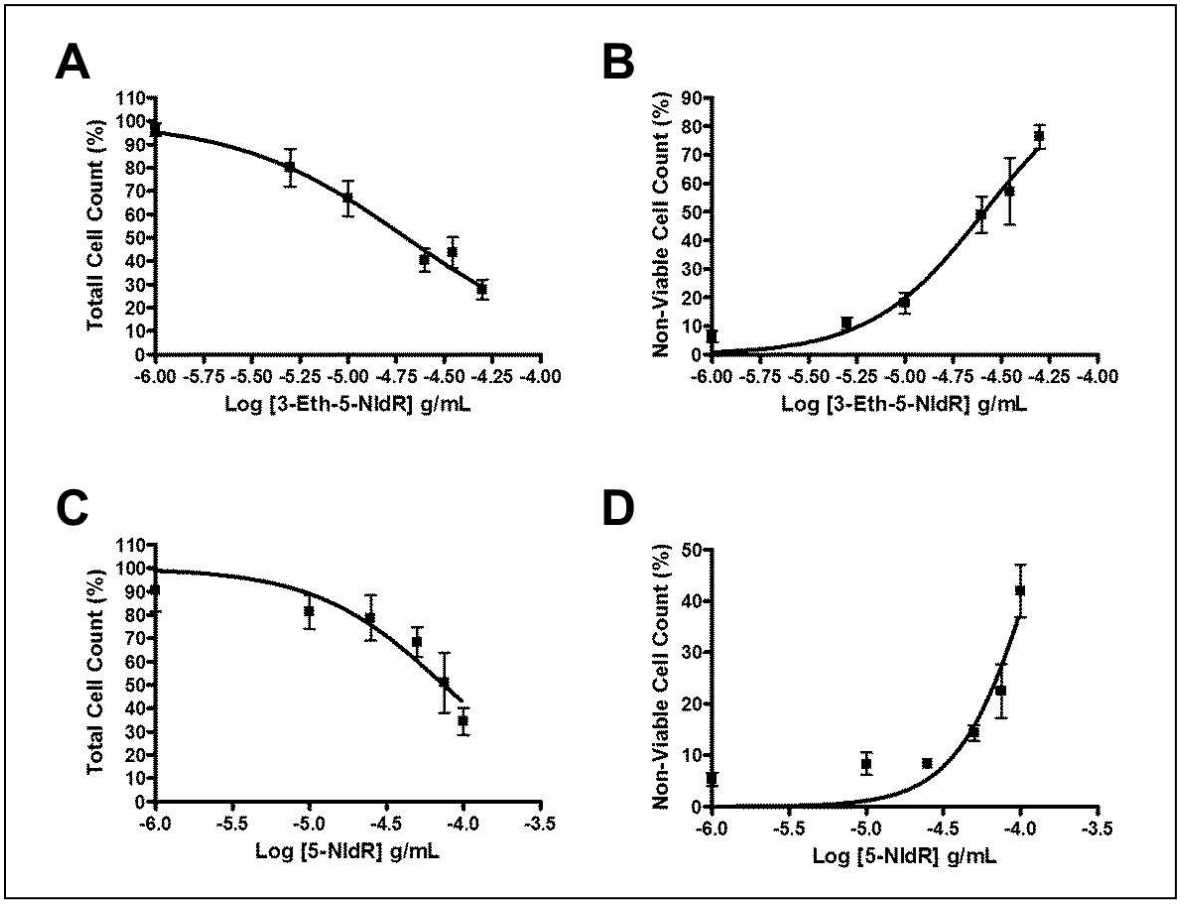
SUPPLEMENTAL FIGURE 1: Michaelis-Menten plots for the incorporation of dATP and dGTP by terminal deoxynucleotidyl transferase (TdT).



SUPPLEMENTAL FIGURE 2: Nucleotide incorporation by terminal deoxynucleotidyl transferase (TdT) up to 20 minutes. Note that 3-Eth-NITP is not elongated beyond DNA_{n+1} whereas 5-NITP is extended to obtain products ranging in size from DNA_{n+1} to DNA_{n+10}.



SUPPLEMENTAL FIGURE 3: Dose-response curves for the non-natural nucleosides in acute lymphoblastic (ALL) cell lines after treatment with non-natural nucleosides for three days. Dimethyl sulfoxide (DMSO) was used as the vehicle and the highest nucleoside concentration used in each experiment was 100 µg/mL. GraphPad Prism was used to obtain a (A) IC_{50} value of 14.1 ± 2.4 µg/mL and (B) LD_{50} value of 27.7 ± 1.7 µg/mL for 3-Eth-5-NIdR against TdT-positive MOLT4 cells. (C) IC_{50} value of 36.4 ± 5.8 µg/mL and (D) LD_{50} value of approximately 100 µg/mL were obtained for 5-NIdR against TdT-positive MOLT4 cells.



SUPPLEMENTAL FIGURE 4. Agarose gel shows DNA laddering as a result of apoptosis in MOLT4 cells treated with non-natural nucleosides. DNA was extracted from MOLT 4 cells treated with (**I**) DMSO [Vehicle], (**II**) 100 µg/mL 5-NIdR, (**III**) 40 µg/mL 3-Eth5-NIdR, and (**IV**) 100 µg/mL 3-Eth-5-NIdR for 48 h. The 1 Kb DNA Marker was from New England Biolabs, Inc. DNA isolation was performed using the DNeasy® Blood & Tissue kit from QIAGEN.