

Supporting Information

Title: The non-enantioselectivity property of human deoxycytidine kinase explained by structures of the enzyme in complex with L- and D-nucleosides

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Content: Fig. S1 and Table S1

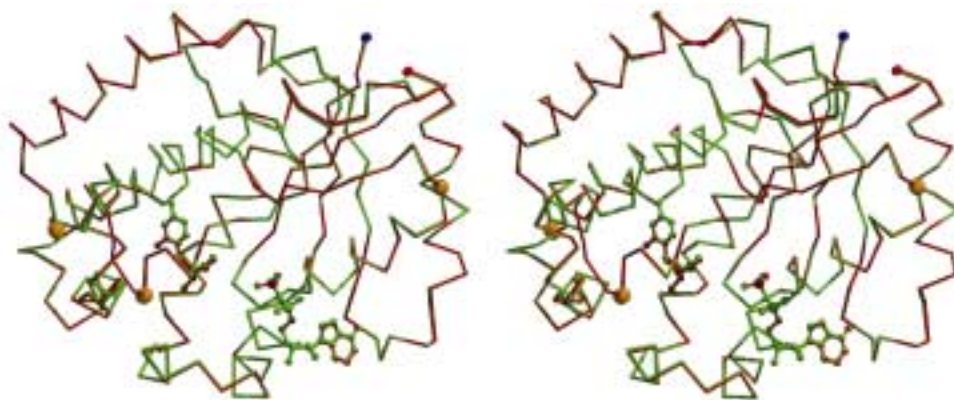


Fig. S1. Stereo representation of the superposition of the structures of the WT-dCK (red) and the C₄S-variant (green) in complex with D-dC and ADP. The overlay reveals that the overall fold of dCK is identical between the C₄S mutant and the WT enzyme. This demonstrates that C₄S is a reliable substitute of the WT dCK. The cysteine residues mutated to serines in the C₄S construct (Cys45, Cys59 and Cys146) are represented as yellow balls. Cys9 is not shown because for all structures the electron density preceding residue 20 does not allow model building. The blue ball corresponds to the N-terminus (N₂₀) and the red ball to the C-terminus (C₂₆₀) of the enzyme. Note that the positions of both the main chain residues and of the nucleoside/nucleotides are preserved.

| Enzyme | Nucleoside | ATP as phosphoryl donor | | | UTP as phosphoryl donor | | |
|------------------|-------------------|-------------------------|--|--|-------------------------|--|--|
| | | K _m (μM) | k _{cat} (sec ⁻¹) | k _{cat} /K _m (sec ⁻¹ M ⁻¹) | K _m (μM) | k _{cat} (sec ⁻¹) | k _{cat} /K _m (sec ⁻¹ M ⁻¹) |
| WT | FTC | 4.4±0.6 | 0.036 ± 0.001 | 8.2 × 10 ³ | 6.2±0.5 | 0.100 ± 0.002 | 16.2 × 10 ³ |
| C ₄ S | FTC | 4.9±0.4 | 0.040 ± 0.001 | 8.2 × 10 ³ | 14.9±0.6 | 0.112 ± 0.001 | 7.4 × 10 ³ |
| WT | 3TC ^b | 3.4 ± 1.0 | 0.030 ± 0.001 | 8.8 × 10 ³ | 8.0 ± 1.0 | 0.102 ± 0.001 | 12.8 × 10 ³ |
| C ₄ S | 3TC ^b | 8.4 ± 1.4 | 0.028 ± 0.002 | 3.3 × 10 ³ | 14.9 ± 1.3 | 0.072 ± 0.002 | 4.8 × 10 ³ |
| WT | L-dC ^b | <3 | 0.012 ± 0.001 | >4 × 10 ³ | <3 | 0.010 ± 0.001 | >3 × 10 ³ |
| C ₄ S | L-dC ^b | <3 | 0.036 ± 0.001 | >12 × 10 ³ | <3 | 0.042 ± 0.001 | >14 × 10 ³ |
| WT | D-dC ^b | <3 | 0.033 ± 0.001 | >11 × 10 ³ | <3 | 0.049 ± 0.001 | >16 × 10 ³ |
| C ₄ S | D-dC ^b | 3.0 ± 0.2 | 0.173 ± 0.002 | 57.7 × 10 ³ | <3 | 0.217 ± 0.005 | >72 × 10 ³ |

^a Values shown are the averages of at least two experiments, and standard deviations are shown.

^b Taken from Sabini, E., Hazra, S., Burley, S.K., Konrad, M. and Lavie, A. Nucleic Acids Research, Vol. 35, Issue 1, 2007, page 186-192.