

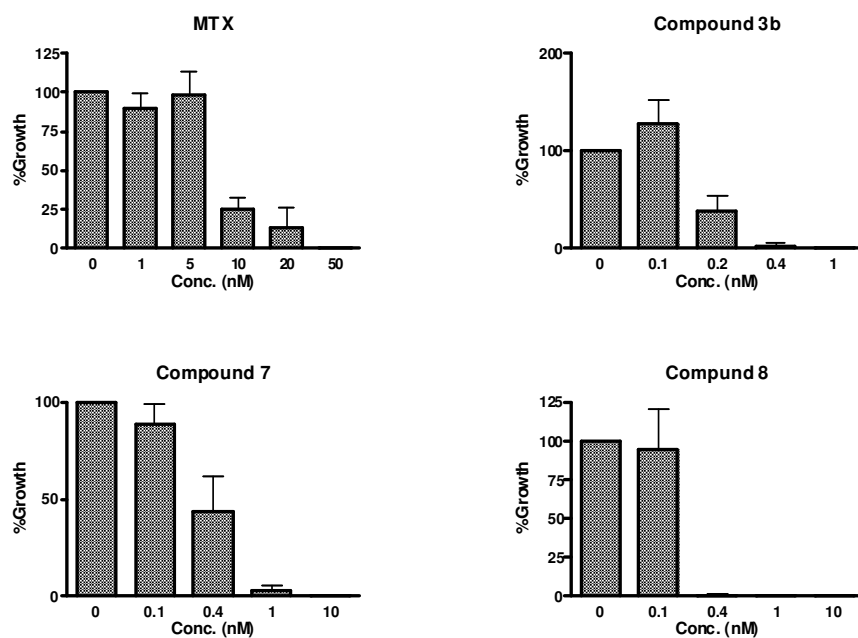
## SUPPORTING INFORMATION

**Synthesis and biological activity of 6-substituted pyrrolo[2,3-*d*]pyrimidine thienoyl regioisomers as inhibitors of *de novo* purine biosynthesis with selectivity for cellular uptake by high affinity folate receptors and the proton-coupled folate transporter over the reduced folate carrier<sup>†</sup>**

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**Figure 1S. Colony formation assay**

**Elemental Analysis**



**Figure 1S. Colony formation assay.** KB cells were inoculated into 60 mm dishes (500 cells per dish), in the presence or absence of a range of concentrations of compound **3b**, compound **7**, compound **8**, or MTX. Colonies were enumerated and results are presented as percent of control treated identically but without drugs, as mean values from 3 experiments (plus/minus SEM). IC<sub>50</sub>s were as follows: **3b**, 0.20 nM; **7**, 0.39 nM; **8**, 0.16 nM; and MTX, 8.5 nM.

## Elemental Analysis

		Calcd, %				Found, %			
Cpd	Formula	C	H	N	S	C	H	N	S
<b>4</b>	$\text{C}_{20}\text{H}_{23}\text{N}_5\text{O}_6\text{S} \cdot 1.85 \text{ H}_2\text{O}$	48.55	5.44	14.15	6.48	48.60	5.09	13.90	6.23
<b>5</b>	$\text{C}_{20}\text{H}_{23}\text{N}_5\text{O}_6\text{S} \cdot 1.0 \text{ H}_2\text{O}$	50.10	5.26	14.61	6.69	50.12	5.18	14.41	6.43
<b>6</b>	$\text{C}_{20}\text{H}_{23}\text{N}_5\text{O}_6\text{S} \cdot 0.4 \text{ CHCl}_3$	48.11	4.63	13.75	6.30	48.15	4.79	13.66	5.95
<b>7</b>	$\text{C}_{20}\text{H}_{23}\text{N}_5\text{O}_6\text{S} \cdot 0.31 \text{ CHCl}_3$	48.93	4.71	14.04	6.43	49.01	5.04	13.64	6.31
<b>8</b>	$\text{C}_{20}\text{H}_{23}\text{N}_5\text{O}_6\text{S} \cdot 1.5 \text{ H}_2\text{O}$	49.17	5.36	14.34	6.56	48.85	4.96	14.00	6.49
<b>9</b>	$\text{C}_{20}\text{H}_{19}\text{N}_5\text{O}_6\text{S} \cdot 1.0 \text{ CH}_3\text{COOH}$	51.06	4.48	13.53	6.20	50.66	4.22	13.57	6.25
<b>10</b>	$\text{C}_{20}\text{H}_{19}\text{N}_5\text{O}_6\text{S} \cdot 0.3 \text{ CHCl}_3$	49.43	3.94	14.20	6.50	49.71	4.30	14.01	6.31
<b>11</b>	$\text{C}_{20}\text{H}_{19}\text{N}_5\text{O}_6\text{S} \cdot 0.3 \text{ CHCl}_3$	49.43	3.94	14.20	6.50	49.80	4.14	13.95	6.15
<b>12</b>	$\text{C}_{20}\text{H}_{19}\text{N}_5\text{O}_6\text{S} \cdot 0.37 \text{ CH}_2\text{Cl}_2$	50.04	4.07	14.33	6.56	50.40	4.13	13.95	6.53
<b>13</b>	$\text{C}_{20}\text{H}_{19}\text{N}_5\text{O}_6\text{S} \cdot 0.41 \text{ CH}_2\text{Cl}_2$	49.80	4.06	14.23	6.51	50.02	4.21	13.83	6.41