Differential Expression of Adenosine Deaminase Isozymes in Acute Leukemia

By Howard Ratech, Frank Martiniuk, William Z. Borer, and Henry Rappaport

Total adenosine deaminase (ADA) and purine nucleoside phosphorylase (PNP) activities were measured in cell samples from 13 cases of de novo acute leukemia and from three cases of chronic myeloid leukemia in blast crisis (CMLBC). These cases could be separated into lymphoid and nonlymphoid types on the basis of enzyme activity, with two misclassifications. However, PNP activity added little or no discriminatory information. Analysis for expression of the various molecular weight (mol wt) ADA isozymes, ADA1 (40 Kd) and ADA2 (110 Kd), revealed that ADA2 was expressed exclusively in nonlymphoid cells whereas ADA1 was found in both lymphoid and nonlymphoid cell types. Identification of ADA1 divided these leukemia cases into lymphoid and nonlymphoid types with no misclassifications (P = .0002; Fisher's exact test). Acute nonlymphoblastic leukemia (ANLL) with a monocytic component tended to have a greater percentage of ADA2 than ANLL without a monocytic component. These studies suggest that ADA2 may be a novel biochemical marker for an immature nonlymphoid cell.

© 1988 by Grune & Stratton, Inc.
10 x 10^6 cells (−70°C). Immediately before use, the frozen samples were thawed in 1 mL 0.001 mol/L sodium phosphate, pH 6.5, and sonicated on ice (0°C) for three to five-second bursts, alternating with 30-second intervals of cooling, with a Branson Sonifier Cell Disruptor model 185 (Danbury, CT) with a microtip at setting 3. After centrifugation for two minutes in a microfuge, the clear supernatant was assayed for ADA and PNP activities using a linked-enzyme spectrophotometric assay, as described previously.13

**ADA isozyme analysis using Sephadex G-200 chromatography.** A frozen cell pellet (−70°C) from patient 14 of 5.0 x 10^6 cells was thawed and suspended in 3 mL 0.01 mol/L sodium phosphate, pH 6.5. The sample was sonicated as described above and centrifuged at 27,000 g for 15 minutes. The clear supernatant containing a total ADA activity of 420 nmol/min was applied to a Sephadex G-200 (2.5 x 90 cm) column equilibrated with 0.1 mol/L sodium phosphate/0.15 mol/L NaCl, pH 6.5, at a flow rate of 16 mL/h, and 2.5 mL fractions were collected. The various steps were maintained at 4°C. ADA activity was determined with adenosine as substrate on collected fractions by a linked enzyme spectrophotometric assay.15 Two hundred microliters of each column fraction plus 50 μL of a mixture of purine nucleoside phosphorylase and xanthine oxidase were added to 800 μL substrate to yield a final concentration of 3.5 mmol/L adenosine in 0.1 mol/L sodium phosphate, pH 6.5. The increase in absorbance at 293 nm owing to formation of uric acid was measured in a recording spectrophotometer during incubation at 37°C. The ADA1 or 110-Kd enzyme peak was collected and concentrated with a YM10 membrane under nitrogen pressure for further kinetic analysis.

**Micromethod for ADA isozyme analysis using size-exclusion high-performance liquid chromatography (HPLC).** Frozen cell pellets of 5 to 10 x 10^6 leukemia cells from patients no 1 through 13, 15, and 16 were thawed, sonicated, and centrifuged as described above. Two hundred microliters of each column fraction plus 50 μL of a mixture of purine nucleoside phosphorylase and xanthine oxidase were added to 800 μL substrate to yield a final concentration of 3.5 mmol/L adenosine in 0.1 mol/L sodium phosphate, pH 6.5. The increase in absorbance at 293 nm owing to formation of uric acid was measured in a recording spectrophotometer during incubation at 37°C. The ADA1 or 110-Kd enzyme peak was collected and concentrated with a YM10 membrane under nitrogen pressure for further kinetic analysis.

**Results**

**Leukemia cell samples.** The results of immunologic and cytochemical analyses performed on leukemia cell samples are listed in Table 1. Bone marrow (patients no 1, 6, and 9) or peripheral blood (patients no. 2 through 5, 8, and 10 through 16) was obtained from 13 patients with de novo acute leukemia and from three patients with chronic myeloid leukemia in blast crisis. The ages ranged from 12 to 82 years. There were nine men and seven women. The cases were morphologically classified according to the French-American-British (FAB) classification.16,17

The blasts of patient no. 2 contained Sudan black B-positive granules; however, cytochemical stains for myeloperoxidase and nonspecific esterase were negative. Although biphenotypic lineage cannot be completely excluded, the findings of positive Tdt and lymphoid markers and the lack of myeloperoxidase and nonspecific esterase suggest that this is an example of granular ALL.18

Five cases were classified as lymphoid type, including 3 non-B, non-T ALL; 1 T ALL and 1 CMLBC-lymphoid type (CMLBC-LY). Eleven cases were classified as nonlymphoid type including five acute myeloblastic leukemia (AML), two acute myelomonocytic leukemia (AMMOL), two acute monoblastic leukemia (AMOL), and two chronic myeloid leukemia in blast crisis-nonlymphoid type (CMLBC-MY).

**Total ADA and PNP activities in leukemia.** Total ADA and PNP activities were measured in patient de novo acute leukemia and CMLBC cell samples by a linked-enzyme spectrophotometric assay. All cell samples were circulating blasts in the peripheral blood, except for the samples from patients 1, 6, and 9, which were bone marrow. Figure 1 and Table 2 show that these cases could be separated into lymphoid and nonlymphoid types with two misclassifications. Additon of simultaneous measurement of PNP activity to the analysis contributed little or no discriminatory information. A single case of T ALL had very high ADA activity and a single case of CMLBC-LY had high ADA and PNP activities.

**ADA isozymes in leukemia.** To determine the contribution of each ADA isozyme to the total ADA activity in the leukemic cells, we used size-exclusion HPLC (patients no. 1 through 13, 15, and 16) (Fig 2) or Sephadex G200 column chromatography (patient no. 14) (Fig 3) to separate the isozymes. ADA activity in column fractions was determined by a spectrophotometric assay. Analysis of ALL cell samples using size-exclusion HPLC showed only one peak of enzyme activity at 40 Kd or ADA1 (Fig 2). In contrast, ANLL cell samples showed two peaks of ADA activity: a peak at 110 Kd or ADA2 and a peak at 40 Kd or ADA1 (Fig 2). The contribution of ADA2 varied from 1% to 54% of total ADA activity in the ANLL cell samples (Fig 2 and Table 2). ANLL with a monocytic component tended to have a higher percentage of ADA2. ADA2 at the 1% level could be detected by size-exclusion HPLC combined with spectrophotometric assay of the column fractions in AML but not in ALL (Fig 2 and Table 2). Thus, analysis for expression of ADA isozymes, ADA1 (40 Kd) and ADA2 (110 Kd), permitted assignment of these leukemia cases into lymphoid and nonlymphoid cell types with no misclassifications (P = .0002; Fisher’s exact test).

**Further characterization of ADA2.** To confirm that the 110-Kd ADA activity was indeed the ADA2 isozyme, the ADA isozymes from patient 14 (AMOL cells) were partially purified by 40% to 60% ammonium sulfate precipitation followed by Sephadex G200 column chromatography. The collected fractions were assayed for ADA activity. Two separate enzyme peaks eluted with approximate mol wt of 110 Kd (ADA2) and 35 to 40 Kd (ADA1) (Fig 3). Column fractions 91 to 110 (110-Kd ADA activity) were then concentrated, and the Km using adenosine as substrate was determined. The apparent Km was approximately 2 x 10^-3 mol/L. This corresponds well with reported values for ADA2 in spleen (Km = 2 x 10^-3 mol/L)19 and in serum (Km = 2.8 x 10^-3 mol/L).11

Sensitivity of the AMOL cell lysate to the ADA inhibitor erythro (9)-(2-hydroxy-3-nonyl) adenine (EHNA) was also tested. The AMOL cell lysate and a lysate of ADA-positive
### Table 1. Immunologic and Cytochemical Characterization of Leukemia Cells

<table>
<thead>
<tr>
<th>Patient No.</th>
<th>Age (yr)</th>
<th>Sex</th>
<th>Cell Source</th>
<th>Blast (%)</th>
<th>Diagnosis</th>
<th>TdT</th>
<th>HLA-DR</th>
<th>J5</th>
<th>B1</th>
<th>BA-1</th>
<th>BA-2</th>
<th>Leu1</th>
<th>T11</th>
<th>My4</th>
<th>My7</th>
<th>My9</th>
<th>MPX</th>
<th>SBB</th>
<th>ANAE</th>
<th>ANAE + Naf</th>
<th>NCA</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>12</td>
<td>M</td>
<td>BM</td>
<td>92</td>
<td>Non-B, non-T ALL</td>
<td>80</td>
<td>77</td>
<td>78</td>
<td>1</td>
<td>65</td>
<td>28</td>
<td>5</td>
<td>3</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>Neg</td>
<td>Neg</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
</tr>
<tr>
<td>2</td>
<td>20</td>
<td>F</td>
<td>PB</td>
<td>90</td>
<td>Non-B, non-T ALL</td>
<td>80</td>
<td>77</td>
<td>78</td>
<td>1</td>
<td>65</td>
<td>28</td>
<td>5</td>
<td>3</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>Neg</td>
<td>Neg</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
</tr>
<tr>
<td>3</td>
<td>75</td>
<td>M</td>
<td>PB</td>
<td>15</td>
<td>Non-B, non-T ALL</td>
<td>NT</td>
<td>NT</td>
<td>17</td>
<td>80</td>
<td>51</td>
<td>11</td>
<td>20</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>Neg</td>
<td>Neg</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
</tr>
<tr>
<td>4</td>
<td>13</td>
<td>M</td>
<td>PB</td>
<td>96</td>
<td>T-ALL</td>
<td>50</td>
<td>1</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>1</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>Neg</td>
<td>Neg</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
</tr>
<tr>
<td>5</td>
<td>59</td>
<td>M</td>
<td>PB</td>
<td>61</td>
<td>CMLBC-LY</td>
<td>80</td>
<td>89</td>
<td>62</td>
<td>14</td>
<td>11</td>
<td>&lt;1</td>
<td>&lt;1</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>Pos</td>
<td>Pos</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
</tr>
<tr>
<td>6</td>
<td>29</td>
<td>F</td>
<td>BM</td>
<td>69</td>
<td>AML</td>
<td>27</td>
<td>32</td>
<td>6</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>29</td>
<td>41</td>
<td>30</td>
<td>26</td>
<td>17</td>
<td>Pos</td>
<td>Pos</td>
<td>Pos</td>
<td>NT</td>
</tr>
<tr>
<td>7</td>
<td>78</td>
<td>F</td>
<td>PB</td>
<td>94</td>
<td>AML</td>
<td>3</td>
<td>6</td>
<td>1</td>
<td>3</td>
<td>8</td>
<td>82</td>
<td>NT</td>
<td>(Leu 4 - 1)</td>
<td>12</td>
<td>90</td>
<td>12</td>
<td>Pos</td>
<td>Pos</td>
<td>Pos</td>
<td>NT</td>
<td>NT</td>
</tr>
<tr>
<td>8</td>
<td>65</td>
<td>M</td>
<td>PB</td>
<td>90</td>
<td>AML</td>
<td>9</td>
<td>9</td>
<td>1</td>
<td>1</td>
<td>7</td>
<td>12</td>
<td>NT</td>
<td>(Leu 4 - 1)</td>
<td>12</td>
<td>90</td>
<td>12</td>
<td>Pos</td>
<td>Pos</td>
<td>Pos</td>
<td>NT</td>
<td>NT</td>
</tr>
<tr>
<td>9</td>
<td>82</td>
<td>M</td>
<td>BM</td>
<td>78</td>
<td>AML</td>
<td>25</td>
<td>69</td>
<td>38</td>
<td>4</td>
<td>NT</td>
<td>46</td>
<td>NT</td>
<td>(Leu 4 - 3)</td>
<td>11</td>
<td>58</td>
<td>35</td>
<td>Pos</td>
<td>Pos</td>
<td>Pos</td>
<td>NT</td>
<td>NT</td>
</tr>
<tr>
<td>10</td>
<td>27</td>
<td>F</td>
<td>PB</td>
<td>97</td>
<td>AML</td>
<td>10</td>
<td>64</td>
<td>0</td>
<td>3</td>
<td>64</td>
<td>8</td>
<td>4</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>Pos</td>
<td>Pos</td>
<td>Pos</td>
<td>Pos</td>
<td>Pos</td>
<td></td>
</tr>
<tr>
<td>11</td>
<td>48</td>
<td>F</td>
<td>PB</td>
<td>70</td>
<td>AMML</td>
<td>72</td>
<td>1</td>
<td>3</td>
<td>2</td>
<td>3</td>
<td>3</td>
<td>4</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>Pos</td>
<td>Pos</td>
<td>Pos</td>
<td>Neg</td>
<td>NT</td>
<td></td>
</tr>
<tr>
<td>12</td>
<td>67</td>
<td>F</td>
<td>PB</td>
<td>90</td>
<td>AMML</td>
<td>18</td>
<td>0</td>
<td>2</td>
<td>3</td>
<td>24</td>
<td>1</td>
<td>4</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>Pos</td>
<td>Pos</td>
<td>Pos</td>
<td>Neg</td>
<td>NT</td>
<td></td>
</tr>
<tr>
<td>13</td>
<td>41</td>
<td>M</td>
<td>PB</td>
<td>87</td>
<td>AMOL</td>
<td>NT</td>
<td>NT</td>
<td>28</td>
<td>6</td>
<td>NT</td>
<td>14</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>Pos</td>
<td>Pos</td>
<td>Pos</td>
<td>Neg</td>
<td>NT</td>
<td></td>
</tr>
<tr>
<td>14</td>
<td>33</td>
<td>M</td>
<td>PB</td>
<td>95</td>
<td>AMOL</td>
<td>NT</td>
<td>NT</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>4</td>
<td>2</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>Pos</td>
<td>Pos</td>
<td>Pos</td>
<td>Neg</td>
<td>NT</td>
<td></td>
</tr>
<tr>
<td>15</td>
<td>58</td>
<td>F</td>
<td>PB</td>
<td>45</td>
<td>CMLBC-MY</td>
<td>NT</td>
<td>NT</td>
<td>0</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>5</td>
<td>NT</td>
<td>NT</td>
<td>NT</td>
<td>Neg</td>
<td>Neg</td>
<td>Neg</td>
<td>Pos</td>
<td>NT</td>
<td></td>
</tr>
</tbody>
</table>

PB, peripheral blood; BM, bone marrow; Pos, positive; Neg, negative; NT, not tested; TdT, terminal deoxynucleotidyl transferase; J5, common acute lymphoblastic leukemia antigen; B1, B-cell-restricted antigen; BA-1 and BA-2, B-cell-associated antigens; Leu1, Leu4, Leu8, and T11, T-cell antigens; My4, My7, and My9, monocyctic and myeloid antigens; MPX, myeloperoxidase; SBB, Sudan black B; ANAE, α-naphthyl acetate esterase; NaF, sodium fluoride; NCA, naphthol chloroacetate esterase; ALL, acute lymphoblastic leukemia; AML, acute myeloid leukemia; AMML, acute myelomonocytic leukemia; AMOL, acute monoblastic leukemia; CMLBC, chronic myeloblast leukemia in blast crisis; LY, lymphoid type; MY, myeloid or monoblastic type.
TABLE 2. ADA Isozymes in Patient Leukemia Cell Samples

<table>
<thead>
<tr>
<th>Patient No.</th>
<th>Diagnosis</th>
<th>Total PNP Activity*</th>
<th>Total ADA Activity*</th>
<th>ADA Activity (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Non-B, non-T ALL</td>
<td>29</td>
<td>52</td>
<td>ND</td>
</tr>
<tr>
<td>2</td>
<td>Non-B, non-T ALL</td>
<td>52</td>
<td>97</td>
<td>ND</td>
</tr>
<tr>
<td>3</td>
<td>Non-B, non-T ALL</td>
<td>15</td>
<td>73</td>
<td>ND</td>
</tr>
<tr>
<td>4</td>
<td>T ALL</td>
<td>24</td>
<td>216</td>
<td>ND</td>
</tr>
<tr>
<td>5</td>
<td>CMLBC-LY</td>
<td>122</td>
<td>222</td>
<td>ND</td>
</tr>
<tr>
<td>6</td>
<td>AML</td>
<td>22</td>
<td>88</td>
<td>ND</td>
</tr>
<tr>
<td>7</td>
<td>AML</td>
<td>20</td>
<td>55</td>
<td>ND</td>
</tr>
<tr>
<td>8</td>
<td>AML</td>
<td>55</td>
<td>19</td>
<td>ND</td>
</tr>
<tr>
<td>9</td>
<td>AML</td>
<td>16</td>
<td>23</td>
<td>ND</td>
</tr>
<tr>
<td>10</td>
<td>AML</td>
<td>24</td>
<td>55</td>
<td>ND</td>
</tr>
<tr>
<td>11</td>
<td>AMMOL</td>
<td>37</td>
<td>49</td>
<td>ND</td>
</tr>
<tr>
<td>12</td>
<td>AMMOL</td>
<td>15</td>
<td>38</td>
<td>ND</td>
</tr>
<tr>
<td>13</td>
<td>AMOL</td>
<td>74</td>
<td>28</td>
<td>ND</td>
</tr>
<tr>
<td>14</td>
<td>AMOL</td>
<td>59</td>
<td>34</td>
<td>ND</td>
</tr>
<tr>
<td>15</td>
<td>CMLBC-MY</td>
<td>54</td>
<td>37</td>
<td>ND</td>
</tr>
<tr>
<td>16</td>
<td>CMLBC-MY</td>
<td>66</td>
<td>69</td>
<td>ND</td>
</tr>
</tbody>
</table>

Vo, ADA activity in the void volume of the size-exclusion column.

Patient leukemia cell samples (nos. 1–13, 15, 16) were analyzed for different mol-wt ADA isozymes by size-exclusion HPLC followed by a spectrophotometric assay for ADA activity in the collected fractions (details given in Materials and Methods section). Patient 14 was analyzed by Sephadex G200 column chromatography.

ND, not detectable (<0.3 nmols/min/mL).

*In nmol/min/mg protein.
Fig 3. Sephadex G200 column chromatography of ADA isozymes. Determination of the mol wt of AMOL ADA isozymes by gel-filtration column chromatography. A 4-mL homogenate of AMOL cells was applied to a Sephadex G200 column (2.5 x 96 cm) in 0.1 mol/L sodium phosphate/0.15 mol/L NaCl, pH 6.5, at a flow rate of 16 mL/h; 2.5-mL fractions were collected, and the column fractions were assayed for ADA activity spectrophotometrically (details given in the Materials and Methods section). Bovine serum albumin (68 Kd) (A), Ovalbumin (45 Kd) (B), and chymotrypsinogen (25 Kd) (C) were used for column calibration, and their elution volumes are indicated by arrows.

Fig 2. Representative ADA isozyme analyses of leukemia cell samples by a micro method: ALL, patient 1; AML, patient 6; AMOL, patient 13. HPLC size-exclusion chromatography used a 7.5 x 600-mm TSK G3000 SW Ultropak column, isocratic gradient 0.1 mL/min, 0.1 mol/L sodium phosphate/sodium sulfate, pH 6.5. ADA activity in each 1-mL collected fraction was measured by a linked spectrophotometric assay that converts adenosine to uric acid. The maximum absorbance of uric acid is at 293 nm. Mol-wt standards: thyroglobulin (670 Kd); β-amylose (200 Kd); purine nucleoside phosphorylase (PNP) (81 Kd) and carbonic acid anhydrase (31 Kd). Arrow indicates endogenous PNP activity, which was measured in each sample as an internal mol-wt marker.

Table 3. ADA Isozymes in Cultured Leukemia Cell Lines

<table>
<thead>
<tr>
<th>Cell Line</th>
<th>Type</th>
<th>Total ADA Activity</th>
<th>ADA Activity (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Vo</td>
<td>ADA1</td>
</tr>
<tr>
<td>KG1</td>
<td>Myeloblastic</td>
<td>226</td>
<td>ND</td>
</tr>
<tr>
<td>ML2</td>
<td>Myeloblastic</td>
<td>228</td>
<td>13</td>
</tr>
<tr>
<td>U937</td>
<td>Histiocytic</td>
<td>669</td>
<td>7</td>
</tr>
<tr>
<td>HL60</td>
<td>Promyelocytic</td>
<td>35</td>
<td>ND</td>
</tr>
<tr>
<td>K562</td>
<td>Erythroleukemia</td>
<td>501</td>
<td>ND</td>
</tr>
<tr>
<td>CCRF-CEM</td>
<td>T-Lymphoblastic</td>
<td>313</td>
<td>ND</td>
</tr>
</tbody>
</table>

Vo, ADA activity in the void volume of the size-exclusion column. This may represent either particulate or high-mol-wt ADA or, alternatively, aggregation of low-mol-wt ADA.

ADA isozymes were identified in the cultured leukemia cell lines by size-exclusion HPLC followed by a spectrophotometric assay for ADA activity in the collected fractions (details given in the Materials and Methods section).

ND, not detectable (<0.3 nmol/min/mL).

* nmols/min/mg protein.

Finding of ADA2 in myeloblastic and histiocytic cultured leukemia cell lines but not in promyelocytic, erythroid, or lymphoblastic cell lines.

Although the possibility of endocytosis of serum ADA1 by the patients' leukemia cells cannot be formally excluded, this seems unlikely for two reasons: the percentage of ADA2 activity in the ANLL cells was very high as compared with other cell types and the human leukemia cell lines KG1, ML2, and U937 also had high ADA2 activity, even when grown in vitro in horse serum, which has very low ADA activity.

ADA1 has been extensively studied, but relatively little is known about ADA2. In humans, ADA2 is a minor component of liver and spleen, but a major component of serum ADA activity. The cellular source for serum ADA2 has not yet been determined. ADA2 appears to be a major component of liver ADA activity in lower vertebrates.

By means of a sensitive radioactive ADA assay, an
aminohydrolase corresponding to ADA₂ has been detected by other researchers at very low levels (0.4% to 2.7% of total ADA activity of normal subjects) in ADA-positive and ADA-deficient B-lymphoblast cell lines. We may not have detected ADA₂ in the ALL cell samples studied either because we used a less sensitive spectrophotometric technique or because ADA₂ expression is different at different stages of lymphomatous maturation and differentiation.

To our knowledge, this is the first report identifying ADA₂ on the basis of mol wt, kinetic properties, and insensitivity to EHNA with a specific hematopoietic cell type, eg, ANLL and CMLBC-MY. ANLL with a monocytic component had relatively high ADA₂ activity. Bloom, in 1972,²² using agarose gel electrophoresis, detected a slow-moving, abnormal ADA band in all six of six AMML, in one of five AML but not in any of eight ALL patient cell samples, or in erythrocytes. This is consistent with our observations of an ADA isozyme, eg, ADA₂, which is differentially expressed at high levels in ANLL, especially in cases with a monocytic component.

Although the physiologic role of ADA₂ is unknown, nonlymphoid cells that express ADA₂ also coexpress ADA₁. The presence of ADA₂ in ANLL and CMLBC-MY may have important implications for the use of 2′-deoxycoformycin (DCF), an ADA inhibitor, in the chemotherapy of hematologic malignancies (reviewed in ref 12). Because ADA₂ is more resistant than ADA₁ to DCF inhibition,²⁰ standard doses of DCF, or DCF plus adenine nucleoside analog combinations, are likely to be relatively ineffective against neoplastic cell types expressing both ADA₁ and ADA₂ isozymes.

REFERENCES

Differential expression of adenosine deaminase isozymes in acute leukemia

H Ratech, F Martiniuk, WZ Borer and H Rappaport