Studies on the Use of “Prophylactic” Intrathecal Amethopterin in Childhood Leukemia

By DAVID K. MELHORN, SAMUEL GROSS, BARRY J. FISHER and ARTHUR J. NEWMAN

CENTRAL NERVOUS SYSTEM INFILTRATION in acute leukemia is especially common in children.¹ With the advent of a greater number of antileukemic agents and increasing variation in their use, the resultant prolongation in survival has been paralleled by a coincident increase in intracranial complications.² Precise data on the frequency of leukemic involvement of the central nervous system (CNS) are not available because the symptoms are often overlooked or, less commonly, because involvement produces no immediate symptoms.³⁻⁴ Consequently, the estimations in frequency of CNS involvement vary from 20 to 80 per cent.³⁻⁵ Other poorly understood aspects of CNS infiltration include the frequency of meningeal involvement at the time of the onset of the leukemic process and the relationship between the presenting peripheral white blood cell (WBC) count and CNS involvement.

The effective measures currently in use in the treatment of CNS leukemia include intrathecal amethopterin and/or ionizing irradiation to the head and spine.⁶⁻⁷ These agents are usually continued until the cerebrospinal fluid (CSF) is essentially free of blast cells.⁸ However, continuous maintenance with intrathecal agents following the initial appearance of CNS involvement has not attained general success in preventing its recurrence.⁹ Accordingly, a study was undertaken to determine the frequency of CNS infiltration at the time of initial diagnosis of leukemia, the relationship between the presenting WBC counts and the time to onset of CNS involvement, and the effectiveness of intrathecal amethopterin given prior to the onset of CNS involvement in preventing or delaying the occurrence of CNS leukemia.

METHODS

All children in whom the initial diagnosis of acute leukemia was made on the Pediatric Service at University Hospitals, Cleveland, between July, 1966, and January, 1969, were included in the study. The diagnosis was established on the basis of the history and physical examination as well as the evaluation of peripheral blood and bone marrow films.

From the Department of Pediatrics, Case Western Reserve University School of Medicine at Babies and Childrens Hospital, Cleveland, Ohio.

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DAVID K. MELHORN, M.D.: Advanced Clinical Fellow of the American Cancer Society, Instructor in Pediatrics, Case Western Reserve University School of Medicine. SAMUEL GROSS, M.A., M.D.: Associate Professor of Pediatrics, Case Western Reserve University School of Medicine. BARRY J. FISHER, M.D.: Clinical Fellow of the American Cancer Society. ARTHUR J. NEWMAN, M.D.: Assistant Clinical Professor of Pediatrics, Case Western Reserve University School of Medicine, Cleveland, Ohio.
During the study period, a total of 47 children presented with acute leukemia, of whom 46 had acute lymphocytic leukemia and one had acute myelogenous leukemia. Following diagnosis, the patients were assigned to one of two study groups at random. The treatment ("T") groups received a single intrathecal dose of 0.9 mg./kg. of amethopterin after removal of spinal fluid for complete CSF studies. The amethopterin was given within 24 hours after the diagnosis of leukemia was made. The nontreatment ("NT") group underwent lumbar puncture for CSF studies, but received no intrathecal medication.

Both groups were treated in an identical manner relative to specific antileukemic therapy, which included induction with prednisone and vincristine followed by continuous and sequential therapy with amethopterin, 6-mercaptopurine and cyclophosphamide. Supportive measures consisted of transfusions, antibiotics as needed, and local irradiation (except to the nervous system) when indicated.

After discharge from their initial hospitalization, the patients were seen at weekly intervals in the Pediatric Hematology Outpatient Clinic and were closely followed for symptoms and signs suggestive of CNS involvement. Neurologic and funduscopic examinations were routinely conducted at each visit, and lumbar punctures were performed when indicated.

Criteria for the diagnosis of CNS leukemia were based upon the identification of leukemic blast cells in the spinal fluid. In addition to CSF white cell counts performed in the usual manner, the cells were pre-incubated with albumin, and Wright's stained smears were prepared and examined. Additional criteria included elevated spinal fluid protein and pressure. In no situation was the diagnosis equivocal. In only one instance were there as few as 10 leukemic blast cells/mm.3 Blast cell counts in all other patients ranged from 200 to 1200/mm.3 In addition, no patients were found to have an elevated CSF pressure or protein without a coincident cellular response. For purposes of the study, the time to onset of CNS leukemia was defined as the length of the interval between the date of the marrow diagnosis and the date of the lumbar puncture indicating CNS infiltration.

All of the CSF specimens were cultured for bacterial growth, and none was positive. Viral cultures were not obtained. Further, no patient received antibiotic therapy during treatment of the initial occurrence of CNS infiltration; only intrathecal amethopterin and/or irradiation were used. No deaths related to CNS involvement were recorded. Twenty-eight patients died during the study period, and postmortem examinations were carried out in 27 cases.

RESULTS

The procedure of lumbar puncture performed on the patients at the time of the initial diagnosis of acute leukemia did not result either in mortality or morbidity. Of the 47 children admitted to the study, four patients (8.5 per cent) had CNS leukemic infiltration at the time of the original diagnosis. These patients had no symptoms or signs of CNS involvement. They were removed from the subsequent study groups and treated with intrathecal amethopterin. All four of these patients presented with peripheral white blood cell (WBC) counts greater than 10,000/mm.3

Of the remaining 43 patients, 24 were placed in the "T" group and 19 in the "NT" group. The two groups were similar in mean age of onset of leukemia and in mean length of survival. Thirty-three lumbar punctures were performed during the followup of study patients when clinical symptoms or signs suggested CNS involvement. Twenty-six CSF specimens (78.9 per cent) were found to contain leukemic blast cells. Thirteen of the 24 children in the "T" group (54.2 per cent) developed CNS infiltration, as compared to 13 of 19 patients (68.4 per cent) in the "NT" group (Table 1). This difference was not considered significant. Of the 43 patients in both "T" and "NT" groups, 26 (60.5 per cent) developed CNS involvement. In all the children in whom the
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Table 1.—Occurrence and Time to Onset of CNS Infiltration in “T” and “NT” Groups

<table>
<thead>
<tr>
<th></th>
<th>Number</th>
<th>Sex</th>
<th>Average Age and Age Range at Onset of Leukemia (yrs.)</th>
<th>Average Age When Developing CNS Infiltration</th>
<th>p* Value</th>
<th>Average Time to Onset of CNS Infiltration (mo.)</th>
<th>p† Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>“T” Group</td>
<td>24</td>
<td>16</td>
<td>8</td>
<td>5.6</td>
<td>13</td>
<td>54.2</td>
<td>11.9</td>
</tr>
<tr>
<td>“NT” Group</td>
<td>19</td>
<td>8</td>
<td>11</td>
<td>4.5</td>
<td>13</td>
<td>68.4</td>
<td>7.6</td>
</tr>
<tr>
<td>Total</td>
<td>43</td>
<td>24</td>
<td>19</td>
<td>5.05</td>
<td>26</td>
<td>60.5</td>
<td>9.75</td>
</tr>
</tbody>
</table>

* p—Comparison of number developing CNS infiltration in “T” and “NT” groups.
† p—Comparison of average time to onset of CNS infiltration in “T” and “NT” groups.

Table 2.—Relationship of Peripheral White Blood Cell Counts to Occurrence of CNS Infiltration

<table>
<thead>
<tr>
<th>Number and Percentage Developing CNS Infiltration</th>
<th>Number</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients with initial WBC counts &lt; 10,000/mm³</td>
<td>23</td>
<td>10</td>
</tr>
<tr>
<td>Patients with initial WBC counts &gt; 10,000/mm³</td>
<td>20</td>
<td>16</td>
</tr>
</tbody>
</table>

The children in both “T” and “NT” groups were then divided according to their peripheral white blood cell counts at the time of the original diagnosis of leukemia to determine whether a relationship existed between the WBC count at diagnosis and the frequency and time to onset of CNS infiltration (Table 2). Among the 23 patients with initial WBC counts less than 10,000/mm³ in both the “T” and “NT” groups, 10 (43.5 per cent) developed CNS infiltration. In the 20 patients in the “T” and “NT” groups with WBC counts greater than 10,000/mm³, 16 (80.0 per cent) showed CNS involvement. A comparison of the frequency of CNS infiltration in high and low WBC count patients indicates a significantly increased frequency of CNS involvement in patients who presented initially with elevated WBC counts (p < 0.005).

If the four patients with CNS infiltration at the time of initial diagnosis are added to the high WBC group who developed CNS involvement during the study period, 20 of 24 patients (83.3 per cent) with initially elevated WBC counts manifested CNS infiltration at some time during the course of the disease. Comparison of the frequency of CNS involvement between this group and those patients who presented with WBC counts less than 10,000/mm³ shows a difference of even greater significance, with a p value of 0.001.
Table 3.—Relationship of Peripheral White Blood Cell Counts to Occurrence and Time to Onset of CNS Infiltration in “T” and “NT” Groups

<table>
<thead>
<tr>
<th></th>
<th>Number</th>
<th>Number and Percentage Developing CNS Infiltration</th>
<th>Average Time to Onset of CNS Infiltration (mo.)</th>
<th>p * Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>“T” Group Patients with Initial WBC &lt; 10,000/mm³</td>
<td>15</td>
<td>6</td>
<td>40.0</td>
<td>11.1</td>
</tr>
<tr>
<td>“NT” Group Patients with Initial WBC &lt; 10,000/mm³</td>
<td>8</td>
<td>4</td>
<td>50.0</td>
<td>12.7</td>
</tr>
<tr>
<td>“T” Group Patients with Initial WBC &gt; 10,000/mm³</td>
<td>9</td>
<td>7</td>
<td>77.8</td>
<td>12.4 &lt; 0.005</td>
</tr>
<tr>
<td>“NT” Group Patients with Initial WBC &gt; 10,000/mm³</td>
<td>11</td>
<td>9</td>
<td>81.8</td>
<td>5.1</td>
</tr>
</tbody>
</table>

* p—Comparison of average time to onset of CNS infiltration.

The average time to onset of CNS infiltration in “T” patients with initial WBC counts less than 10,000/mm³ was 11.9 months, as compared to 12.7 months in the low WBC count “NT” patients (Table 3). The difference between these groups is not significant (p > 0.1). In the “T” patients with WBC counts greater than 10,000/mm³ who developed CNS involvement, the mean time to onset was 12.4 months, as compared to 5.1 months in the high WBC “NT” patients. A significant difference between these two groups is noted, with a p value of < 0.005.

DISCUSSION

The frequent occurrence of CNS infiltration in the study patients with acute leukemia confirms previous observations.1,2 Careful evaluation of the patients’ symptoms and detailed weekly neurologic examinations were selected in order to determine when lumbar punctures should be performed, rather than the performance of serial lumbar punctures at regular arbitrary intervals. It was recognized that CNS infiltration might not produce symptoms for a period of time after leukemic involvement of the central nervous system had taken place. It was also felt, however, that the infiltration would soon become manifest if patients were closely observed in regard to symptoms and signs of CNS involvement. This impression was borne out by the results of the 27 postmortem examinations on patients who died during the study period. Only one was found to have CNS infiltration which was not identified clinically prior to death. Careful attention to sometimes vague and “insignificant” symptoms, such as infrequent vomiting, occasional transient headache, and unexpected weight gain (suggesting hypothalamic involvement), resulted in a relatively high percentage of “positive” lumbar punctures. Approximately 80 per cent of all CSF examinations performed when indicated by the patients’ symptoms and signs revealed the presence of leukemic blast cells.

Lumbar puncture performed routinely in study patients during their initial hospitalization indicated that a significant minority (8.2 per cent in this study) of the leukemic children had CNS infiltration at the time of initial diagnosis.
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without clinical evidence of CNS involvement. It is noteworthy that the four patients in whom CNS infiltration was recorded at the time of the original diagnosis of leukemia presented with WBC counts above 10,000/mm.$^3$ No clinical evidence of CNS involvement was noted in these patients. The presence of such "silent" infiltration in children at the time of their original diagnosis suggests that CSF examination should be a part of the initial evaluation of children with acute leukemia. It is particularly indicated in patients with elevated peripheral white blood cell counts.

In contrast to the observations of Hardisty and Norman, who found no relationship between the initial peripheral white blood cell count and either the occurrence or time to onset of CNS infiltration,$^2$ the present data show that the occurrence of CNS involvement is significantly greater in patients who presented with WBC counts greater than 10,000/mm.$^3$ Patients not receiving intrathecal amethopterin who had initial WBC counts above 10,000/mm.$^3$ also developed infiltration more rapidly than "NT" patients with low WBC counts (5.1 mo. vs 12.7 mo; p < 0.005). The cause of the increased frequency of CNS infiltration in those patients initially presenting with elevated peripheral WBC counts is not clear. A reasonable speculation, however, is that CNS infiltration is the result of the migration of leukemic blast cells into the CNS from the peripheral circulation, and that the frequency of such migration is directly proportional to the number of blast cells in circulation.

The administration of "prophylactic" intrathecal amethopterin prior to onset of CNS involvement did not prevent or reduce the occurrence of CNS infiltration in study patients. However, the average time to onset of CNS involvement in "T" group patients was longer than that of "NT" patients. This difference between "T" and "NT" groups is a reflection of the more rapid time to onset of CNS infiltration in "NT" patients with elevated WBC counts. The time to onset of CNS infiltration was significantly longer in high WBC count patients in the "T" group than in similar patients in the "NT" group. Accordingly, the use of intrathecal amethopterin before the onset of obvious CNS infiltration appears to be a significant adjunct in delaying morbidity in those children who present with elevated peripheral WBC counts. It remains to be seen whether the administration of "prophylactic" intrathecal amethopterin at regular intervals after the initial dose might further delay the onset of CNS involvement in patients who present with elevated WBC counts, and even in those children with low initial WBC counts.

**Summary**

Central nervous system infiltration was studied in a group of 47 children with acute leukemia. CNS involvement was found to occur more frequently and appear more rapidly in patients who presented with elevated peripheral WBC counts. CNS infiltration not suspected clinically was identified by spinal fluid examination in a significant minority of children at the time of the initial diagnosis of leukemia, indicating that lumbar puncture should be a routine part of the initial evaluation of patients with acute leukemia. Intrathecal amethopterin administered "prophylactically" at the time of initial diagnosis of leukemia did not prevent or decrease the frequency of occurrence
of CNS infiltration. However, it did delay the onset of CNS involvement in patients with elevated WBC counts. Intrathecal amethopterin administered before the onset of CNS infiltration appears to be useful in delaying morbidity resulting from CNS involvement in children who present with elevated peripheral WBC counts.

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REFERENCES

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