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Effective Dose of L-Asparaginase for Induction of Remission in Previously Treated Children with Acute Lymphocytic Leukemia: A Report from Childrens Cancer Study Group[1]

Inta J. Ertel, Mark E. Nesbit, Denman Hammond, John Weiner, and Harland Sather

University of Michigan, Ann Arbor, Michigan 48109 [I. J. E.]; University of Minnesota Hospital, Minneapolis, Minnesota 55455 [M. E. N.]; University of Southern California, Los Angeles, California 90033 [D. H.]; and Childrens Cancer Study Group, Operations Office, Los Angeles, California 90031 [J. W., H. N. S.]

ABSTRACT

L-Asparaginase, in the dose of ≥6000 IU/sq m three times weekly, was demonstrated to be an effective agent in reinduction of remissions in childhood leukemia.

Four hundred thirteen children with acute lymphocytic leukemia were treated with L-asparaginase. Doses i.m. ranged from 300 to 12,000 IU/sq m. None of the patients had received prior asparaginase therapy. 6-Mercaptopurine was given p.o. concurrently. All of the patients had experienced several previous relapses, and their disease was not responsive to 6-mercaptopurine. L-Asparaginase was found to be effective in reinducing remissions at the following rates: 9.5% for 300 IU/sq m; 35.1% for 3,000 IU/sq m; 53.5% for 6,000 IU/sq m; and 62.5% for 12,000 IU/sq m. The drug was given three times weekly for four weeks. Hypersensitivity reactions occurred in 6.5% of patients.

INTRODUCTION

L-Asparaginase has been highly successful for inducing remissions in acute leukemia since 1967 (11). The doses used ranged from 300 IU (13, 15) to 425,000 IU (7) per sq m i.v. or i.m. (1, 4, 7, 13–15). The interval of drug administration has ranged from daily to once weekly.

A major toxicity of L-asparaginase has been hypersensitivity; one-fourth to one-third of the patients have shown significant reactions during the first course of i.v. therapy (12). Previously, we have shown that, by giving the enzyme i.m. and also by adding 6-mercaptopurine to therapy, the incidence of hypersensitivity reactions to L-asparaginase can be decreased (9, 10).

This paper summarizes the clinical results of a study to determine the most effective, but lowest, dosage of L-asparaginase when combined with 6-mercaptopurine for the reinduction of remission in children with acute lymphocytic leukemia in relapse.

MATERIALS AND METHODS

A cooperative study was initiated in 1971 by the Childrens Cancer Study Group to determine the most effective and least toxic dose schedule of L-asparaginase to be used for the reinduction of remission in children with previously relapsed acute lymphocytic, stem cell, or undifferentiated leukemia.

L-Asparaginase[2] was used after dilution with 0.9% NaCl solution (2 ml) without preservative. The enzyme was immediately injected i.m.

Patients who developed CNS[3] leukemia during induction therapy were treated with either i.t. methotrexate or radiation therapy. Patients were removed from the study if they failed to enter remission or if they had uncontrollable toxicity.

Patients were eligible for the protocol if they had not received L-asparaginase before and if their disease was resistant to 6-mercaptopurine as evidenced by either failing induction with 6-mercaptopurine or relapsing while on 6-mercaptopurine maintenance. A review of evidence for resistance to 6-mercaptopurine was performed retrospectively, and the patients with some question of 6-mercaptopurine resistance were not eligible for the induction evaluation. These patients were included in the evaluation for toxicity and other complications.

The initial randomization was between L-asparaginase doses of 300 and 12,000 IU/sq m i.m. 3 times weekly (Chart 1) combined with 6-mercaptopurine (75 mg/sq m p.o. daily). The randomization was changed to 3000 IU *versus* 6000 IU when significant differences in the induction rate of the original regimens were detected. The L-asparaginase was to be started on Day 3. The selection of these doses for L-asparaginase represented narrowing the spectrum of the doses reported to have some activity. Bone marrow examination was performed after 14 and 28 days of therapy, and if the patient had not achieved a complete remission (6) within that time it was permissible to extend the therapy for 2 more weeks.

RESULTS

Between January 1971 and August 1975, 413 children were admitted to both randomizations of this study. Of these, 297 were documented to be resistant to 6-mercaptopurine (evaluable for induction). In the first randomization, the lowest dose of L-asparaginase (300 IU/sq m) used in combination with 6-mercaptopurine was shown to have only minimal activity; 9.3% achieved an M-1 (bone marrow cell differential count with less than 5% blast forms) remission, while 55.3% reached an M-1 marrow on the higher dose of 12,000 IU/sq m ($p = 0.0002$). Following completion of this section of the study, patients were randomized between L-asparaginase doses of 3000 and 6000 IU/sq m. When the dose of 3000 IU/sq m was shown to be less effective (30.4% remissions, $p = 0.04$) than the higher dose of 6000 IU/sq m with 45.3% remissions, randomization

[1] Childrens Cancer Study Group investigators, institutions, and grant numbers are given in "Appendix." Address requests for reprints to Childrens Cancer Study Group Operations Office, 1721 Griffin Avenue, Los Angeles, Calif. 90031.

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[2] Crasnitin was provided in 2,000- and 10,000-unit vials by Bayer Co., whose representative in the United States is Delbay Pharmaceuticals, Inc., Bloomfield, N. J.

[3] The abbreviations used are: CNS, central nervous system; i.t., intrathecal.

was discontinued.

In the 413 patients originally entered into the study, the remission rates were directly related to the doses of L-asparaginase received. The results were analyzed 3 ways: Method 1, all patients based on assigned drug regimen; Method 2, excluding patients who were not documented to have relapsed while receiving 6-MP prior to this study; and Method 3, based on the actual amount of drug received by the patient.

Method 1. The results on all of the patients showed that L-asparaginase in the doses of 300, 3,000, 6,000, or 12,000 IU/sq m when given i.m. 3 times weekly for 4 to 6 weeks led to M-1 remissions in 9.3, 30.4, 45.3, or 55.3% of the patients, respectively. There was no significant difference between the remission rates at 6,000 and 12,000 IU [$p = 0.54$ (see Table 3)]. The duration of therapy is shown in Table 1. The majority of the patients received less than 5 weeks of therapy.

Method 2. The records were reviewed to identify the patients who could be documented to have relapsed while receiving 6-mercaptopurine prior to this study. Chart 2 shows the remission rates (9.5, 35.1, 53.5, and 62.5% for 300, 3,000, 6,000, and 12,000 IU, respectively). When only patients with documented prior relapse on 6-mercaptopurine are considered, the dose-response relationship is similar whether the patients had a previously documented relapse on 6-mercaptopurine or not.

Method 3. Results were based on doses received. Some patients did not get the complete assigned dose because of intercurrent illness or toxicity. Therefore, the patient data were further analyzed based upon the actual average amount of drug received per patient. Patients who received L-asparaginase (see Chart 3) doses of <3000 IU, within the range of 3000 to 6000 IU, and ≥ 6000 IU/sq m achieved an M-1 marrow at the rates of 24.2, 40.2, and 50.9%.

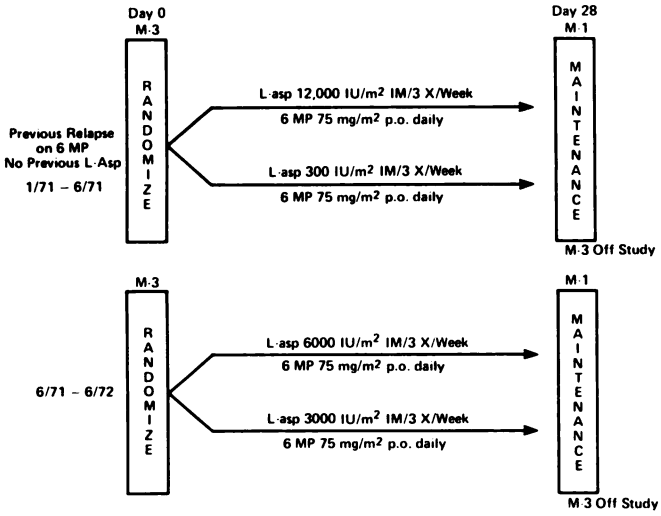

Chart 1. Schematic diagrams of reinduction therapy plans. *L-asp*, L-asparagine; *6 MP*, 6-mercaptopurine; *x*, times.

Table 1

Induction response by days on induction (patients with documented prior relapse on 6-mercaptopurine

	Days on induction		
	<35 days	>35 days	Total
M-1	98	22	120
Not M-1	152	25	177

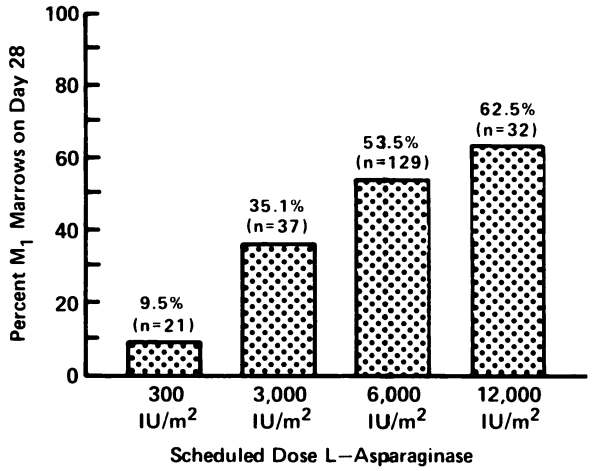

Chart 2. Dose-response relationship of L-asparaginase to Day 28 M-1 bone marrow (only patients with documented prior relapse on 6-mercaptopurine.

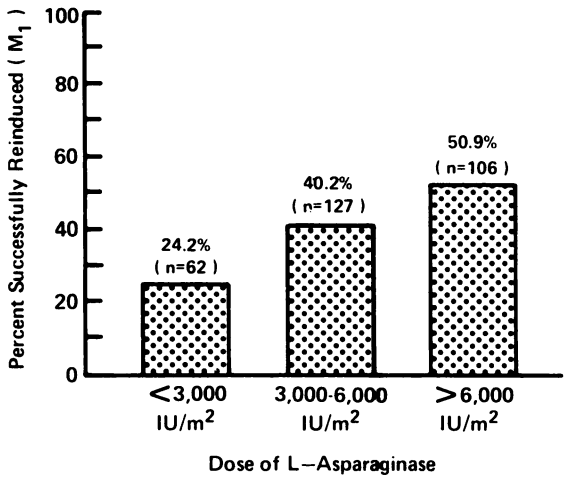

Chart 3. Dose-response relationship of L-asparaginase dose to successful reinduction (only patients with documented prior relapse on 6-mercaptopurine.

In an attempt to evaluate the early effect of L-asparaginase, an examination of the 14-day marrow was found to be quite helpful in predicting a favorable response. In the majority (71.7%) of the children who achieved a complete remission, a decreased number of blasts in the peripheral blood and the bone marrow were noted by 2 weeks of therapy (Chart 4). Most of the patients (53.5 to 62.5%) who achieved a remission did so in 4 weeks. Table 1 shows that 98 of 120 (81%) patients who achieved an M-1 marrow did so in less than 35 days.

Incidence of symptomatic CNS leukemia was documented in 12 to 15% of patients during the study period of 1 to 42 days. No correlations are apparent between the different dosage regimens and the appearance of this complication. (At 300 IU of L-asparaginase, 15% developed CNS disease, while 12% did so at 3,000 and 6,000 IU, and 13% became diseased at 12,000 IU.) Of those patients who developed CNS disease, the majority (70%) did so within the first 7 days of therapy, and 18% developed it after 14 days of the therapy. CNS leukemia evaluation was not a requirement in this study, and the data cannot be considered meaningful.

Although the patients between 2 and 8 years old (8) did minimally better than those <2 years or >8 years old, no

significant differences in remission rates could be demonstrated. The WBC at entry was less than 5000 in the majority of the patients. There appears to be no significant correlation between the WBC at entry and the achievement of remission.

Adequate data were available to evaluate complications and drug toxicity on 381 patients. Significant neutropenia was documented in 148 (39%) of the 381 patients, but it was difficult to differentiate drug-induced neutropenia from disease effects. The incidence of toxicity other than perhaps neutropenia was approximately 10%. No significant differences were seen between the different dosage regimens. One patient died of sepsis associated with marked neutropenia, possibly related to the therapy. No other deaths could be related to the drug therapy. Vomiting occurred in 28 (7%) while the L-asparaginase was being given. L-Asparaginase was discontinued in 10 patients. The reasons for stopping therapy were liver toxicity in 4 patients, pancreatitis in 3 patients, and allergic reactions in 3 patients. Diabetes was not recorded in any of the patients.

Hypersensitivity reactions were documented in 25 of 381 patients (6.5%) (Table 2). No reactions were fatal. One patient had anaphylaxis on Day 14, and another patient had laryngospasm on Day 21. In addition, either a generalized or a localized rash was observed in 23 patients. Urticaria and nausea was associated with the rash in 2 patients. Itching and swelling of hands and feet developed on Day 36 in one patient. All of the significant reactions, either allergic or due to toxicity, occurred after the 13th day of therapy. Most of the reactions occurred after the 19th day of therapy. It is worthwhile to note that no serious allergic reactions were reported with the lower doses in the present study. All of the serious reactions were seen with ≥6000 IU doses. The numbers are not statistically significant, since only 3 patients developed allergic reactions requiring cessation of therapy.

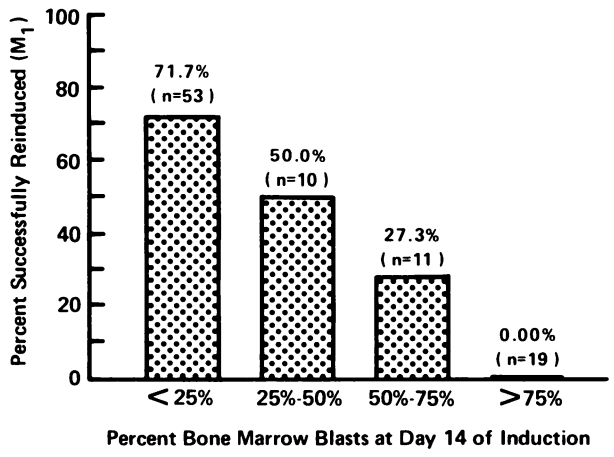

Chart 4. Relationship of Day 14 blast count to reinduction success (only patients with documented prior relapse on 6-mercaptopurine and without blasts in the peripheral blood at 14 days).

DISCUSSION

L-Asparaginase has been shown to be an effective agent in reinducing remissions in late-stage childhood leukemia. Some clinical effect (induction of remission) was demonstrable at all dosage levels from 300 to 12,000 IU/sq m; however, the proportion of complete remissions increased with increasing doses of the drug. At 2 weeks, a significant observable drug effect (decreased leukemic blasts) could be demonstrated in most of the patients who eventually went on to achieve a remission. The continuation of the drug beyond 4 weeks led to 11% more remissions in this patient population.

In view of the wide range of doses reported to have been used for L-asparaginase ranging from 300 to 425,000 IU/sq m, there was a definite relationship between the dose and the response in our study patients of up to 6000 IU (see Table 3). Previous reports show that in patients treated with the doses ranging from 300 to 150,000 units/sq m/day, M-1 or M-2 (bone marrow cell differential count with 5 to 25% blast forms) remissions were seen in 45 of 73 patients (62%). One-third of these patients developed allergic reactions (2, 15). In another report, 10 of 32 patients treated with L-asparaginase in the dose range of 6,000 to 30,000 IU achieved a complete remission (5). Additional reports show remissions in 9 of 21 (13), 33 of 79 (42%), and 40 of 81 (50%) patients (10).

When L-asparaginase was one of the drugs in multidrug therapy, remissions were reported in 70% of patients. In 5% of these patients, hypersensitivity reactions have been reported (3, 5). The above data are very similar to our results with 6.5% of patients developing hypersensitivity reactions if 6-mercaptopurine was used as an immunosuppressant. Many authors have consistently reported high levels of allergic reactions with the enzyme when it is used alone (3, 5, 12, 14, 15). The low levels of allergic reactions at all dosage regimens in this study may be related to the 6-mercaptopurine therapy (9). The overall complications and toxicity from L-asparaginase were low at all 4 dosage levels tested.

There were no reports of bleeding in the area of injection in these patients. L-Asparaginase i.m. appears safe even in patients with few or no circulating platelets. The i.m. route of administration may have contributed to the decreased rate of allergic reactions.

In conclusion, L-asparaginase is an effective agent in reinduction of remissions in childhood leukemia when given in the dose of ≥6000 IU/sq m 3 times weekly for 4 weeks. If given with 6-mercaptopurine, few hypersensitivity reactions occur. Antileukemic activity may be documented early in the course of therapy by a two-week examination of the bone marrow.

Table 2

Hypersensitivity reactions

No. of patients	Reaction
1	Anaphylaxis
1	Laryngospasm
2	Urticaria, nausea, swelling
15	Rash (generalized)
5	Rash (localized)
1	Itching and swelling of hands and feet
25	Total (25/381 or 6.5%)

Table 3

Dose-related differences in responses (based on 2-sided test of proportions)

	p
300 IU *versus* 12,000 IU	0.0002
300 IU *versus* 3,000 IU	0.08
300 IU *versus* 6,000 IU	0.0002
3,000 IU *versus* 6,000 IU	0.04
3,000 IU *versus* 12,000 IU	0.03
6,000 IU *versus* 12,000 IU	0.54

APPENDIX

Principal Investigators in Childrens Cancer Study Group

Institution	Investigator	Grant
University of Southern California at Los Angeles	Denman Hammond, M.D. John Weiner, Dr. P.H. Richard Honour, Ph.D. Harland Sather, Ph.D.	Ca 13539
University of Michigan, Ann Arbor	Ruth Heyn, M.D.	CA 02971
Childrens Hospital of the District of Columbia	Sanford Leikin, M.D.	CA 03888
Cornell Medical Center, New York	Denis R. Miller, M.D.	CA 14557
Childrens Hospital of Los Angeles	Gussie Higgins, M.D.	CA 02649
Babies Hospital, New York	James Wolff, M.D.	CA 03526
Childrens Hospital of Pittsburgh	Vincent Albo, M.D.	CA 07439
Childrens Hospital of Columbus	William Newton, M.D.	CA 03750
Childrens Orthopedic Hospital, Seattle	Ronald Chard, M.D.	CA 10382
University of Wisconsin, Madison	N. Shahidi, M.D.	CA 05436
University of Minnesota, Minneapolis	Mark Nesbit, M.D.	CA 07306
Childrens Memorial Hospital, Chicago	George Honig, M.D.	CA 07431
University of Utah Medical Center, Salt Lake City	Eugene Lahey, M.D.	CA 10198
Princess Margaret Hospital, Toronto	Marilyn Sonley, M.D.	Ontario Cancer Treatment and Research Foundation Grant 41-17
University of Rochester, New York	Martin Klemperer, M.D.	CA 11174
Childrens Hospital of Louisville	Donald R. Kmetz, M.D.	
University of British Columbia, Vancouver	Mavis Teasdale, M.D.	Vancouver Foundation
Childrens Hospital of Philadelphia	Milton Donaldson, M.D.	CA 11796
Indiana University, Indianapolis	Robert Baehner, M.D.	CA 13809
New Jersey College of Medicine, Newark	Leonard Vitale, M.D.	CA 12637
Harbor General Hospital, Torrance, Calif.	Jerry Finklestein, M.D.	CA 14560
University of California, San Francisco	Arthur Ablin, M.D.	CA 17829
Rainbow Babies and Childrens Hospital, Cleveland	Samuel Gross, M.D.	

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