Pharmacokinetics of Cladribine (2-Chlorodeoxyadenosine) in Children with Acute Leukemia

Christine M. Kearns, Raymond L. Blakley, Victor M. Santana, and William R. Crom

Departments of Pharmaceutical Sciences [C. M. K., W. R. C], Hematology-Oncology [V. M. S.], and Molecular Pharmacology [R. L. B.], St. Jude Children's Research Hospital, and Departments of Clinical Pharmacy [W. R. C.], Pediatrics [V. M. S.], and Pharmacology [R. L. B.], University of Tennessee, Memphis, Tennessee 38101-0318

ABSTRACT

Cladribine is a synthetic purine nucleoside with demonstrated activity in hairy cell leukemia and acute myeloid leukemia. We have studied the pharmacokinetics of this drug in 25 pediatric patients with acute leukemia treated with cladribine as a single agent, 8.9 mg/m²/24 h, for 5 days by continuous i.v. infusion. Twelve patients were in relapse, and acute myeloid leukemia was newly diagnosed in 13 patients. Plasma, urine, and cerebrospinal fluid cladribine concentrations were determined by a radioimmunoassay with a limit of detection of 1 nM. An open two-compartment model was fit to the plasma concentration data. The mean (SD) clearance was 39.4 (12.4) liters/h/m² and ranged from 14.4-55.4 liters/h/m². When clearance was normalized to body weight (liters/h/kg) it was negatively correlated with age, with older patients having slower clearances per unit of body weight. However, when clearance was normalized to body surface area, no significant correlation with age was observed. The mean (SD) steady-state plasma concentration (predicted 120-h concentration) was 37.7 (17.3) nM and ranged from 23.2-64.5 nM. The terminal phase half-life in 22 patients ranged from 14.3-25.8 h, with a mean (SD) of 19.7 (3.4) h. The volume of distribution at steady state was highly variable, with a mean (SD) of 356.6 (225.2) liters/m². None of these parameters was significantly different between patients in relapse and patients with newly diagnosed disease. Renal clearance was determined in 7 patients and ranged from 34.6-643.6 ml/min/m², with a mean (SD) of 317.9 (208.7) ml/min/m². Renal clearance as a percentage of total systemic clearance ranged from 11.0-85.1%, with a mean of 51.0%. In 11 patients, the mean (SD) cerebrospinal fluid concentration was 6.1 (3.97) nM, a mean of 18.2% of the steady-state plasma concentration. The CSF:plasma concentration ratio was significantly higher on day 5 (22.7% in 7 patients) than on day 4 (7.6% in 3 patients; P = 0.03). Additional studies are needed to further define the metabolic fate of cladribine. In this paper we provide the first comprehensive description of the pharmacokinetics of this drug in children and provide data which suggest that cladribine may be useful in the treatment of patients with meningeal leukemia or malignancies of the central nervous system.

INTRODUCTION

Cladribine (2-chloro-2'-deoxyadenosine, 2-CDA, Leustatin) is a synthetic purine nucleoside analogue that is not catabolized by adenosine deaminase (1). It is rapidly taken up and phosphorylated by lymphoid and myeloid cells (2, 3); the triphosphate form strongly inhibits ribonucleotide reductase (4, 5) and is a good substrate for human DNA polymerases (6). Incorporation into DNA results in rapid chain termination such that DNA replication in dividing cells is quickly arrested (4). In nondividing cells, cladribine causes accumulation of DNA strand breaks (7, 8), probably due to inhibition of DNA repair. This in turn results in the depletion of NAD and ATP, probably due to poly(ADP) ribosylation of nuclear proteins. The unrepairred DNA damage, like DNA damage caused by other oncolytic agents (9-11), triggers programed cell death (apoptosis) (12, 13).

Cladribine is highly effective in the treatment of hairy cell leukemia (14-16) but may also prove useful to treat lymphoma (17), chronic myeloid leukemia (18), and chronic lymphoid leukemia (19-21). In clinical trials at St. Jude Children's Research Hospital, we have demonstrated notable activity in children and young adults with relapsed acute leukemia (AML3 and acute lymphocytic leukemia) (2, 22) and in previously untreated AML.4

There have been few studies of the pharmacokinetics of cladribine. Lilieermal and Julisson (23) described the pharmacokinetics after 2- and 24-h infusions of 0.14 mg/kg in 12 adult patients with lymphoproliferative diseases. More recently, Liliemark et al. (24) compared cladribine pharmacokinetics in adult patients to whom the drug was administered as a 2-h infusion, s.c., or orally. We previously reported (22) preliminary pharmacokinetic data from 5 patients with acute myeloid or lymphoid leukemia in first or later relapse, who were given cladribine by continuous infusion for 5 days at 8.9 mg/m²/day. No other cladribine pharmacokinetic studies in pediatric patients have been reported. The purpose of the present study was to extend those initial observations to a larger pediatric population, including both relapsed leukemia patients and newly diagnosed patients with AML, and to determine the renal clearance and CSF concentrations of cladribine during a 120-h i.v. infusion.

MATERIALS AND METHODS

Patients and Drug Administration. Demographic characteristics of the patients are summarized in Table 1. Twenty-five patients, 12 males and 13 females, ranging in age from 8 months to 23 years (mean and median, 9.6 years) were studied. Twelve had relapsed hematological malignancies, 6 each with acute myeloid leukemia and acute lymphoid leukemia, and were treated in a phase II study of cladribine. The clinical results of this study have been published (22). For entry into the phase II study, all patients were required to have a performance status of 0-2 (Eastern Oncology Cooperative Group criteria) and a life expectancy of at least 6 weeks and be free from uncontrolled infection. Patients who had received prior chemotherapy were required to have recovered from any previous treatment toxicity. All patients were required to have adequate renal and hepatic function as defined by: blood urea nitrogen, <40 mg/dl; serum creatinine, <1.5 mg/dl; total bilirubin, <1.5 mg/dl; serum aspartate aminotransferase and serum alanine aminotransferase, both <200 IU/liter. Metabolic parameters were required to be within a normal range.

Thirteen study subjects were newly diagnosed patients with acute myeloid leukemia and were treated with one course of cladribine as a single agent in a phase II "therapeutic window" prior to treatment with a conventional combination of daunorubicin, cytarabine, and etoposide. Patients were not required to have normal biochemical measurements prior to entry into the study.

Cladribine was synthesized for clinical use by a previously published method (3). Purity of the final product (>99%) was confirmed by elemental

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2 To whom requests for reprints should be addressed, at Pharmaceutical Sciences Department, St. Jude Children's Research Hospital, 332 N. Lauderdale, Memphis, TN 38105.

3 The abbreviations used are: AML, acute myeloid leukemia; CSF, cerebrospinal fluid; Vc, volume of the central compartment; Kcs, elimination rate constant, Kcint, intercompartment rate constant; Kcint, intercompartment rate constant; Vdss, volume of distribution at steady state; f1u, initial apparent plasma half-life; f1u, terminal phase apparent plasma half-life.

Renal clearance was calculated by dividing the cladribine urinary excretion rate (amount excreted per unit of time) by the mean plasma cladribine concentration during the urine collection period. All the clearances calculated from the individual urine collection periods were averaged for each patient.

RESULTS

The mean (SD) clearance, normalized to body surface area, for these 25 patients was 39.4 (12.4) liters/h/m², and clearance ranged from 14.4–55.4 liters/h/m². When normalized to body weight, the mean (SD) clearance was 1.44 (0.62) liters/h/kg and ranged from 0.31–2.43 liters/h/kg. The steady-state plasma cladribine concentration was defined as the concentration at 120 h, predicted from each patient’s individual pharmacokinetic parameters. These values ranged from 23.2–84.5 nm, with a mean (SD) of 37.7 (17.3) nm and a median of 29.6 nm.

The $t_{1/2\beta}$ was determined for 22 patients who had plasma concentrations measured for at least 12 h after the end of the infusion. Plasma concentrations were determined for only 12 h postinfusion in 2 patients, for 28 h in one patient, and for 48–72 h postinfusion in the other 19 patients. In this group, the mean (SD) $t_{1/2\beta}$ was 19.7 (3.4) h and ranged from 14.3–25.8 h. Cladribine appears to distribute into a relatively large volume at steady state. In our population, $V_d$, ranged from 32.0–799 liters/m², with a mean (SD) of 356.6 (225.2) and median of 323.0 liters/m². Pharmacokinetic parameters are summarized in Table 2.

Clearance, $t_{1/2\beta}$, and $V_d$ each normalized to both body weight and body surface area were compared with all the demographic variables shown in Table 1 to determine whether any significant correlations were present. Only clearance normalized to body weight (liters/h/kg) was correlated with any of these variables. It was highly correlated with weight, body surface area, height, and age, all of which are highly correlated with each other. The predictive abilities ($R^2$) of the linear regression equation for each of these variables versus clearance was 58.4, 55.7, 42.6, and 41.8%, respectively ($P < 0.0005$ in each case). The relationship between clearance and age is shown in Fig. 1. Clearance, normalized to body weight, decreased as any of these measures of body size increased. However, when clearance was normalized to body surface area (liters/h/m²), it was only weakly correlated with body weight ($R^2=19.6\%, \ P < 0.027$).

Serum creatinine and total bilirubin were significant predictors of clearance (normalized to body weight) with $R^2$ values of 32.7% ($P = 0.003$) and 20.9% ($P = 0.021$), respectively. Clearance decreased as either total bilirubin or serum creatinine increased, as shown in Fig. 2 for serum creatinine. However, this relationship was not present for clearance normalized to body surface area. Total bilirubin was also a significant predictor of the steady-state plasma concentration, although the predictive ability was only 20.7% ($P = 0.022$). This is consistent with the relationship between total bilirubin and clearance, since clearance and steady-state plasma concentration are inversely related. It is noteworthy that all the serum creatinine values were in the normal range, and only one total bilirubin value was abnormal (2.3

### Table 2 Mean (SD) of cladribine pharmacokinetic parameters

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value (SD)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plasma clearance</td>
<td>1.44 (0.62)</td>
</tr>
<tr>
<td>$V_d$ (L/kg)</td>
<td>39.4 (12.4)</td>
</tr>
<tr>
<td>$V_d$ (L/m²)</td>
<td>356.6 (225.2)</td>
</tr>
<tr>
<td>Steady-state plasma concentration (µM)</td>
<td>32.7 (17.3)</td>
</tr>
<tr>
<td>Central distribution volume</td>
<td>1.11 (0.74)</td>
</tr>
<tr>
<td>Terminal apparent half-life (h)</td>
<td>19.7 (3.4)</td>
</tr>
</tbody>
</table>
mg/dl) in this study population. However, the lowest cladribine clearance (and highest steady-state plasma concentration) was observed in this patient. There was no significant difference for any pharmacokinetic parameters between the relapsed patients and those previously untreated. There were also no significant differences for any pharmacokinetic parameters among the cytological subtypes of acute leukemia.

Of the 24 patients evaluable for response to cladribine, there were 8 who achieved a complete response, 6 who had a partial response, and 10 who had no response. However, there was no difference for any pharmacokinetic parameter, including clearance, steady-state plasma concentration, and $t_{1/2}$, among these three response groups.

Renal cladribine clearance was determined in 7 patients, for whom both urinary excretion and plasma concentration data were available. The mean (SD) renal clearance was 317.9 (208.7) ml/min/m² and ranged from 34.6–643.6 ml/min/m², as shown in Fig. 3. The renal clearance was an average of 51.0% of the total systemic clearance, but the percentage of total clearance accounted for by renal clearance ranged from 11.0–85.1%. When renal clearance was plotted against serum creatinine, there was an apparent negative linear relationship between serum creatinine and renal cladribine clearance (renal clearance decreased with increasing serum creatinine). However, this correlation did not achieve statistical significance, due to the small sample size.

Cladribine CSF concentrations were determined for 10 patients on either day 4 (3 patients) or day 5 (7 patients) of the infusion. The mean (SD) CSF concentration was 6.1 (3.97) nM and ranged from 1.68–19.75 nM. The mean (SD) CSF:plasma concentration ratio (expressed as a percentage) was 18.2% (10.0). There appeared to be a relationship between plasma and CSF concentrations, although CSF concentrations were highly variable at any given steady-state plasma concentration. The highest CSF concentration, 14.75 nM, was achieved in the patient with the highest steady-state plasma concentration, 84.5 nM. However, 7 of the patients had plasma concentrations between 20 and 30 nM, with concurrent CSF concentrations in the range of 2.25–11.25 nM. In addition, there appeared to be a time-dependent effect on the CSF cladribine concentrations achieved. In 7 patients from whom CSF samples were obtained on day 5 (the last day of the infusion), the mean CSF:plasma ratio was 22.7% (range 12.4–38.0%). In contrast, for the 3 day 4 patients, the mean CSF:plasma ratio was 7.6% (range, 5.5–9.4%). This difference was statistically significant ($P = 0.03$, Mann-Whitney $U$ test), as shown in Fig. 4.

**DISCUSSION**

The present study represents the first detailed pharmacokinetic study of cladribine in a pediatric patient population, as well as the first pharmacokinetic study of cladribine administered as a prolonged 120-h infusion. Previously, investigators (23, 24) have examined the pharmacokinetics of cladribine in adults only. In those studies, Liliemark et al. (23, 24) described the disposition of cladribine following 2- and 24-h i.v. infusions, as well as after s.c. and oral administration. Plasma drug concentrations were determined by high-performance liquid chromatography (26). In addition, a 3-compartment model was fit to the data following the 2-h i.v. infusion. Despite these
methodological differences, very comparable results were obtained. The mean (SD) clearances following the 2-h infusion in the two studies by Liliemark et al. (23, 24) (calculated from data in the paper) were 0.74 (0.30) liters/h/kg and 0.93 (0.33) liters/h/kg, respectively, and following a 24-h infusion in the same patients in the second study, the mean (SD) clearance was 1.08 (0.47) liters/h/kg. These values compare well with our mean clearance of 1.44 liters/h/kg, especially when the higher clearances in younger children are considered. In 7 patients in our study who were 15 years of age or older, the mean (SD) clearance was 0.71 (0.42) liters/h/kg, and values ranged from 0.31–1.27 liters/h/kg, which are very similar to data from the studies of Liliemark et al. The mean plasma concentration for the 24-h infusion in the first study of Liliemark et al. (23) was 22.5 nm, compared to our mean plasma concentration of 37.7 nm. These differences in steady-state concentrations, despite similar plasma clearances, are accounted for by the lower drug dosage used by Liliemark et al. of 0.14 mg/kg/24 h, which was about one-half the dosage administered in our study.

In contrast, the mean (SD) terminal half-life following the 2-h infusion was 6.7 (2.5) and 9.9 (4.6) h in the studies of Liliemark et al., compared to our value of 19.7 (3.4) h. This difference is probably due to the differences in drug administration and blood sampling between the studies of Liliemark et al. and our work, as well as somewhat greater sensitivity of our radioimmunoassay (~0.2 nm), which permits measurement of plasma concentrations for a longer time, resulting in a longer terminal half-life.

Our observation that plasma clearance normalized to body weight is lower in older children and young adults than in young children is entirely consistent with observations for other anticancer drugs (27, 28). This finding suggests that differences in cladribine clearance related to body size and/or age are more completely accounted for when drug dosage is based on body surface. Therefore, drug dosages based on body surface area are more likely to achieve a consistent plasma concentration across the age range seen in this study than dosages based on body weight. This may be due to the relative size of clearing organs (liver and kidneys) relative to total body size. It has been suggested that for some drugs the “functional hepatic mass,” which approximates liver volume, is a major determinant of hepatic drug metabolism. Liver volume estimated by ultrasound studies is similar in children and adults when expressed as a percentage of body surface area but is greater in children when normalized to body weight, with the ratio of liver volume to weight decreasing with increasing age (39). Therefore, these results suggest that dosages based on body weight (in the pediatric population) will yield lower serum concentrations in younger patients, while dosages based on body surface area are more likely to result in similar concentrations in patients of different ages.

The lack of any predictive relationship between drug disposition and demographic or biochemical variables limits the ability to use these data to adjust dosages (based on body surface area) for differences in renal or hepatic function or on the basis of race or sex. However, the lack of significant correlations is not surprising since almost all of these patients had biochemical measurements in the normal range, which was a requirement for study enrollment. Similarly, the lack of a relationship between drug exposure and either effect or toxicity is not surprising. Regardless of systemic plasma exposure, all of the patients experienced grade 3 or grade 4 hematological toxicity at this dosage. Intrinsic sensitivity of the malignant cells to the drug is probably more important than systemic exposure in producing therapeutic responses. Decreased cellular sensitivity may be due, for example, to reduced kinase activity, resulting in lower intracellular conversion of cladribine to the active nucleotide form or to reduced activity of the various DNA polymerases that are the targets of cladribine.

This is the first clinical study which has measured the renal clearance of cladribine. On average, about half the plasma clearance of this drug is accounted for by excretion of unchanged drug in the urine. However, our data from 7 patients suggests that this may be highly variable, with as little as 10% or as much as 85% of the total drug clearance accounted for by renal clearance. The fate of the remainder is unknown, because no metabolites have been identified in urine or plasma. It is possible that the remainder of the drug is bound to tissues or intracellular sites. Also, our observation of the lowest cladribine clearance in our only patient with an elevated serum indirect bilirubin suggests that biotransformation by hepatic enzymes may play a role in drug elimination. However, additional work needs to be done to determine the role and importance of nonrenal clearance mechanisms.

One previous report by Saven et al. (30) described CSF concentrations of cladribine. In their study, 3 patients achieved CSF concentrations of 2.2–24.7 nm at dosages of 0.1, 0.15, and 0.2 mg/kg/day by continuous i.v. infusion. These values are similar to our mean CSF concentration of 6.54 nm observed in 10 patients (range, 2.25–14.75 nm). In addition, we observed an apparent time dependency on the penetration of cladribine into the CSF, with higher concentrations relative to plasma concentrations after 5 days of continuous infusion, compared to 4 days. These results must be interpreted with extreme caution, however, since so few observations (3 on day 4 and 7 on day 5) are available. Nevertheless, transfer of cladribine into the CSF may be a “membrane-limited” process rather than a “flow-limited” one. This suggests that the amount of drug reaching the CSF is directly related to the length of time a concentration gradient is maintained between the plasma and the CSF. In the paper by Saven et al. (30), it is not specified on which day of the 7-day infusion the CSF specimens were obtained. In combination, these results suggest a potential role for systemic cladribine therapy in the treatment of primary malignancies of the central nervous system, as well as for meningeal leukemia. However, much more work is needed to identify the factors (plasma concentration, length of exposure, etc.) which influence the amount of drug reaching the central nervous system.

In summary, this paper provides the first comprehensive description of cladribine pharmacokinetics in children and the first renal clearance data in patients. Clearance normalized to body weight is higher in children than adolescents or adults, but this difference is not seen when clearance is normalized to body surface area. The potentially cytotoxic cladribine concentrations achieved in the CSF provide a rationale for the use of this drug in treating malignancies of the central nervous system. Additional investigation is needed to determine the metabolic fate of cladribine as well as to define a relationship between systemic drug exposure and tumor response or toxicity.

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REFERENCES


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