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On the Pharmacokinetics of 2-Chloro-2′-deoxyadenosine in Humans[1]

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ABSTRACT

The antitumoral effect of 2-chloro-2′-deoxyadenosine (CdA) in the treatment of lymphoproliferative diseases in general and of hairy cell leukemia in particular has recently been demonstrated. Detailed information on the pharmacokinetics of CdA, however, is lacking. The pharmacokinetics of CdA after 2- and 24-h infusions of 0.14 mg/kg was described in 12 patients with lymphoproliferative diseases using a newly developed high-performance liquid chromatography method. The plasma concentration data from individual patients were fitted to a two-compartment model with α- and β-half-lives of 35 ± 12 (mean ± SD) min and 6.7 ± 2.5 h, respectively. The volume of distribution was 9.2 ± 5.4 liters/kg. The steady-state concentration of CdA during the 24-h infusion was 22.5 ± 11.1 nM. The areas under the time *versus* concentration curves were 552 ± 258 and 588 ± 185 nM × h, respectively, for the 24- and 2-h infusions. The interindividual variability of the determinants of the plasma pharmacokinetics of CdA was small (the coefficients of variation were between 0.22 and 0.58). At 6.3 ± 1.5 h after the start of the 2-h infusion, the concentration of CdA was the same as the steady-state concentration during the 24-h infusion. When the mean plasma concentrations of the 12 patients were fitted to a 3-compartment model, the half-lives of the α-, β-, and τ-phases were 8 min, 1 h 6 min, and 6.3 h, respectively. The long terminal half-life of CdA after 2-h infusion supports the use of intermittent infusions.

INTRODUCTION

The relationship between the severe combined immunodeficiency syndrome with severe lymphopenia and the inherited deficiency of the enzyme adenosine deaminase triggered the interest in a number of previously synthesized purine analogues resistant to adenosine deaminase (1). The two most useful of these agents are F-ara-AMP[3] and CdA. It has been shown that CdA is clinically active against lymphoproliferative diseases in general and HCL in particular (2–4). Although its clinical role in most lymphoproliferative disorders is not yet defined, the use of CdA for front-line treatment of HCL seems fully justified. CdA is a prodrug, and intracellular phosphorylation to the triphosphate is needed for its cytotoxic effects. The first step of this metabolism is mediated via deoxycytidine kinase (1).

Phase I trials have shown that the maximum tolerated dose is 0.7 mg/kg/course given as a 7-day continuous infusion or as 2-h infusions during 5 consecutive days (2). Plasma concentration data have been presented using a radioimmunoassay method for detection. This method has a detection limit of 5 nM, which, however, is not enough for the determination of the pharmacokinetic characteristics of CdA (5). The aim of the present study was to delineate the pharmacokinetics of CdA. A newly developed high-performance liquid chromatography method was used for this purpose (6).

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[3] The abbreviations used are: F-ara-AMP, 9-β-D-arabinofuranosyl-2-fluoroadenine 5′-monophosphate; CdA, 2-chloro-2′-deoxyadenosine; HCL, hairy cell leukemia; AUC, area under the concentration *versus* time curve; CLL, chronic lymphocytic leukemia.

PATIENTS AND METHODS

Twelve patients were treated with CdA at a dose of 0.14 mg/kg during 5 consecutive days. Three patients had CLL of the B-cell (M. O., K. G., H. A.) or the T-cell type (M. H.). The tumor cell count in the peripheral blood was 36, 35, 7, and 55 × 10^9/liter in these patients, respectively. Three patients had low-grade malignant non-Hodgkin's lymphoma (O. M., M. A., E. G.), two patients had HCL (P. H., L. H.), and one patient had cutaneous T-cell lymphoma (G. M.); none of these patients had any significant tumor cell count. One patient had prolymphocytic leukemia of T-cell type (O. H.), and one patient had chronic myelogenous leukemia in lymphoblastic phase (T. G.), with 600 and 61 × 10^9 tumor cells/liter, respectively, in the peripheral blood. All patients had normal renal and hepatic function, defined as a deviation of serum creatinine, bilirubin, alkaline phosphatase, and aminotransferases of less than 50% from the normal (defined as the 95% confidence interval in healthy adults). On days 1, 3, 4, and 5, CdA was administered as a 2-h infusion. During day 2, CdA was given as a 24-h continuous infusion. Samples were taken before, 1 h after the start, and at 0 min, 10 min, 20 min, 30 min, 45 min, 1 h, 2 h, 4 h, 7 h, 10 h, 14 h, and 22 h after the end the infusion on day 1. Two or three samples were also taken 20 h after the start of the 24-h infusion. The study was approved by the local ethics committee, and all patients had given their informed consent to participate in the study. One patient (T. G.) chose to participate in the 2-h infusion study only.

Venous blood samples were taken in heparinized glass tubes, put in ice-water, and centrifuged 550 × *g* for 5 min, and the plasma was stored at −20°C until analysis. CdA was determined with high-performance liquid chromatography as previously described (6). Briefly, 125 pmol of Guaneran (a generous gift from Dr. Gertrude Elion, Wellcome Foundation, Research Triangle Park, NC) and 5 ml of ethyl acetate were added to 1 ml of plasma and vortexed. After centrifuging, the organic phase was evaporated and reconstituted in 40 μl of the mobile phase (sodium phosphate, 50 mM/methanol/acetonitrile, 85/10/5). CdA was separated on a Perkin-Elmer 80- x 4.6-mm, 3-μm C_{18} column (Perkin-Elmer, Norwalk, CT) at a flow rate of 1 ml/min. The recovery of CdA was 55%, and the detection limit was 1 nM.

The plasma concentration data for the elimination phase of the 2-h infusion were fitted to two- or three-compartment pharmacokinetic models using the standard equations (7) which were used to calculate the AUC of the elimination phase. The AUC during the 2-h infusion was calculated according to the trapezoid rule. The V_d was calculated as dose/(AUC × λ_2), where λ_2 is the slope of the terminal elimination phase (7).

The mean ± SD plasma concentration data from the 12 patients were fitted into a three-compartment model. To allow the calculation of mean ± SD concentrations when time points were missing or samples were taken at a slightly different time point, the concentration at a given time point in an individual patient was adjusted or calculated using the two-compartment model.

RESULTS

The pharmacokinetic data are displayed in Table 1. During the 2-h infusion, a CdA concentration of 198 ± 87 nM (range, 70–381 nM) was reached. The decline of CdA concentrations after the infusion in the individual patients could be fitted into a two-compartment model with α- and β-half-lives of 35 ± 12 min (r^2 = 0.86 ± 0.15) and 6.7 ± 2.5 h (r^2 = 0.92 ± 0.07), respectively (Fig. 1). The steady-state concentration during continuous infusion was 22.5 ± 11.1 nM. The AUCs for the 2-

Table 1 *Determinants of CdA plasma pharmacokinetics*

Patient	Peak (nM)	$t_{1/2}\alpha$ (min)	$t_{1/2}\beta$ (h)	$AUC_{2\,h}$ (nM × h)	AUC_{ci}[a] (nM × h)	$AUC_{2\,h}/AUC_{ci}$	Css[b] (nM)	V_d[c] (liters/kg)	Equiconcentration time[d] (h)
M. A.	140	27	5.6	422	407	1.04	17.0	9.3	5.7
M. O.	70	22	6.7	326	358	0.91	10.6	14.5	9.0
O. M.	381	52	5.6	838	481	1.74	19.0	4.7	8.1
E. G.	215	30	5.9	685	654	1.05	27.2	6.0	5.7
H. A.	236	19	2.8	665	612	1.09	25.5	3.0	7.1
M. H.	102	41	6.3	349	292	1.20	12.2	12.7	5.7
K. G.	296	25	3.7	707	776	0.91	32.2	3.7	6.0
P. H.	211	30	6.2	777	892	0.87	37.2	5.6	4.4
G. M.	195	58	9.6	793	1018	1.28	42.4	8.9	4.7
O. H.	224	33	8.5	470	273	1.72	11.4	12.8	7.6
L. H.	106	41	12.1	398	310	1.28	12.9	21.5	5.2
T. G.	205	38	7.0	629				7.8	
Mean	198	35	6.7	588	552	1.19	22.5	9.2	6.3
SD	87	12	2.5	185	258	0.30	11.1	5.4	1.5
C.V.[e]	0.44	0.34	0.37	0.31	0.47	0.25	0.50	0.58	0.23

[a] ci, continuous infusion.
[b] Css, the steady-state concentration during continuous infusion.
[c] V_d, the volume of distribution.
[d] The time point when the CdA concentration after a 2-h infusion equals the Css during continuous infusion in the same patient.
[e] C.V., coefficient of variation.

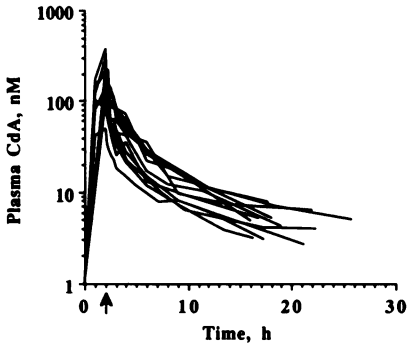

Fig. 1. Concentration of CdA in plasma after a 2-h (*arrow*) infusion of 0.14 mg/kg in 12 patients.

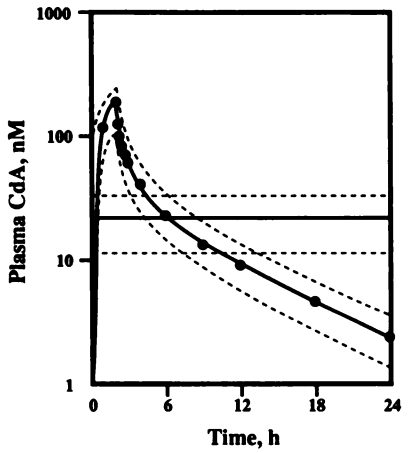

Fig. 2. Mean ± 1 SD of the plasma concentration of CdA after a 2-h and a 24-h infusion of 0.14 mg CdA in 12 patients. The concentration during the 24-h continuous infusion is shown as a concentration level assuming a steady-state situation.

h and the 24-h infusions were 588 ± 185 and 552 ± 258 nM × h, respectively, and the ratio between the AUCs was 1.19 ± 0.30. This indicates that the disposition of CdA in plasma is linear over the dose range, 6–70 μg/kg/h. The volume of distribution was 9.2 ± 5.4 liters/kg. After a mean of 6.3 h after the start of the 2-h infusion, the concentration of CdA had decreased to the steady-state concentration during the 24-h infusion (Fig. 2). The mean plasma concentration of all 12 patients fitted well into a three-compartment model ($r^2 > 0.99$ for all three phases). The α-, β-, and τ-half-lives were 8 min, 1 h 6 min, and 6.3 h, respectively (Fig. 2). The interindividual variation in the determinants of the pharmacokinetics of CdA was small, with coefficients of variation between 0.22 and 0.58 (Table 1). This is also shown by the narrow range between ±1 SD displayed in Fig. 2.

DISCUSSION

The present report represents the first study of CdA which characterizes in detail the pharmacokinetic properties of this clinically very promising drug. Although pharmacokinetic data concerning anticancer drugs have to be interpreted with great care (8), the long τ-phase after the 2-h infusions suggests that CdA can be administered intermittently with retained antitumoral activity when compared to continuous infusion. The comparison of plasma concentration levels and AUCs after intermittent and continuous infusion also supports this conclusion. Although no strict comparison of clinical treatment results between 7-day continuous infusion and 5-day intermittent infusion has been done, our treatment results with the latter mode of administration show that it has a definite effect in lymphoproliferative diseases (9). However, it is not yet clear whether the high plasma CdA concentration during the 2-h infusion has any bearing upon the therapeutic or toxic effects of the treatment.

The patients in this study were selected for normal hepatic and renal function, and thus the interindividual variability of plasma concentrations in this population was relatively small (10). This is probably also due to the resistance of CdA to deamination by adenosine deaminase and the tentative route of elimination, renal excretion. The importance of interindividual variations of plasma concentration for the clinical response is therefore probably limited. It has, however, been shown that the interindividual variability of plasma concentrations of other antimetabolites is much less than that of the corresponding intracellular active nucleotides (11, 12). It is also possible that the variability of the plasma pharmacokinetics between the patients in this study is due to differences in tumor burden. Such a relationship, however, is not apparent in the patients in this study if the tumor cell count is considered to reflect the tumor burden. Until data on the relationship between plasma

CdA and intracellular active metabolite concentrations are available, the significance of these plasma pharmacokinetic data must be interpreted with caution.

The treatment options for CLL improved recently when the impressive results with F-ara-AMP were presented (13). In heavily pretreated CLL, the activity of CdA is also striking (3). The comparison of these new treatment modalities with standard chlorambucil-prednisolone treatment would be greatly facilitated if oral preparations of the drugs were available. It has recently been shown that the bioavailability of F-ara-AMP after oral administration is good (14). The bioavailability of oral CdA is as yet unknown, but with the detection method now available (6) and the knowledge of the pharmacokinetic data presented here, such studies are feasible and on-going.

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