Cancer Chemother Pharmacol (1983) 10:115–119

Cancer Chemotherapy and Pharmacology

Early-Phase Pharmacokinetics of Doxorubicin in Non-Hodgkin Lymphoma Patients

Dose-Dependent and Time-Dependent Pharmacokinetic Parameters

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Summary. *The early-phase (20 min) pharmacokinetics of doxorubicin was studied in 18 patients suffering from non-Hodgkin lymphoma and receiving various schedules and/or dosages of the drug. This pharmacokinetics was time-dependent in most patients over a 2-week interval: repeating similar doses in patients leads to a decrease of the drug exposure due to a decrease of the half-life and/or to a decrease of the extrapolation to 0 time (intercept parameter). The pharmacokinetics was generally time-independent within a 6-h interval in most patients. During this time interval, the kinetics was not linear: increasing the dose by large proportions does not lead to a proportional increase of drug exposure. This time- and dose-dependence of doxorubicin pharmacokinetics makes it very difficult to monitor the treatments according to the individual pharmacokinetic patterns of patients.*

Introduction

It has been demonstrated by several authors [1, 2, 7, 10] that there are wide individual variations in doxorubicin pharmacokinetic parameters. In similar patients, injection of similar doses of the drug leads to very different drug exposures, expressed for instance as C × t evaluations. Therefore, it can be of interest to try to monitor the therapeutic doses in individual patients according to their characteristic pharmacokinetic parameters determined after a test dose of the drug. The time-dependence and linearity of the kinetics of doxorubicin must be determined first, to allow the plasma levels after a therapeutic injection to be predicted.

We have already demonstrated by single-point measurements at 40 min or 3 h that a prior therapy with doxorubicin induces decreased doxorubicin plasma levels during subsequent therapy [4]. A preliminary paper by Gil et al. [5] showed an increase in the total plasma clearance of doxorubicin occurring in a patient who received successive courses of treatment. An early-phase study (0–4 h) by Piazza et al. [7] also showed important changes in the pharmacokinetic parameters during successive courses of the treatment. It can therefore be strongly suspected that the pharmacokinetics of this drug is time-dependent. Much less is known about dose-dependence of the pharmacokinetics of doxorubicin, which has been suggested by Powis et al. [8]. We have therefore developed a study of the very early phase pharmacokinetics of this drug in 18 patients with malignant non-Hodgkin lymphoma and receiving various dosages of doxorubicin either 6 h or 2 weeks apart. Our attention was focused on the first 20 min after the injection, because we had already demonstrated that the extrapolation of the first part of the plasma decay curve could be related to the clinical response to treatment in patients with locally advanced breast cancer [10]. Moreover, it is impossible to follow up the plasma levels of doxorubicin a long time after the injection of a test dose representing $1/10$ of the therapeutic dose.

Our results show that the early-phase kinetics of doxorubicin was time-dependent within the interval between two courses of chemotherapy (2 weeks). Within a time interval of 6 h the pharmacokinetic parameters were generally constant (but not every time). Moreover, the areas under the curves for the first phase of the kinetics were not proportional to the injected doses, which leads to the conclusion that the first-phase kinetics of this drug is not linear.

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Materials and Methods

1. Patients. Eighteen patients entered the study. They were suffering from non-Hodgkin lymphoma, the diagnosis of which was assessed by histological examination. Their ages ranged from 16 to 67; they had no signs of hepatic or renal dysfunction. The main clinical features of the patients are presented in Table 1. They received combination chemotherapy with doxorubicin (35 mg/m^2), cyclophosphamide (400 mg/m^2), and vincristine (0.7 mg/m^2). The doxorubicin dose was fractionated into two parts, the first given in the morning and the second in the afternoon, 6 h after the first one. The ratio between the morning dose and the afternoon one ranged between 1 and 10 in the various patients studied. The remainder of the chemotherapy was injected after the blood sampling for doxorubicin measurements.

Injections of doxorubicin were given by IV bolus over 3 min, whatever the dose injected. Blood samples were taken from the opposite arm in to tubes containing EDTA, at precise selected times generally close to 3, 6, 9, 12, 15, and 20 min after the end of the injection. The blood was centrifuged and the plasma was immediately frozen until analysis.

A first group of patients (patients 1–4) received a test dose in the morning amounting to $1/10$ of the therapeutic dose given in the afternoon. Patients 2 and 3 were similarly studied during a subsequent course of chemotherapy occurring 2 or 4 weeks later. Patients 5 and 6 received a test dose of $1/8$ of the therapeutic dose. The ratio between the therapeutic dose and

Table 1. Clinical features of the patients entering the study

Pt	Age	Sex	Lymphoma grade[a]	Clinical stage of the disease	Response after the first course of chemotherapy[b]
1	59	F	High	I	CR
2	57	M	Low	II	CR
3	16	M	High	IV	CR, then relapse
4	40	M	Low	IV	PR
5	61	M	High	I	CR
6	38	M	High	III	CR
7	61	M	Low	IV	PR
8	68	M	High	II	PR
9	47	M	High	III	CR, then relapse
10	57	M	Low	III	CR
11	65	F	Low	II	CR
12	58	M	Low	I	CR
13	55	M	High	I	CR
14	20	M	High	IV	CR, then relapse
15	40	M	High	II	CR
16	50	M	High	I	CR
17	40	M	Low	III	PR
18	66	F	Low	III	CR

[a] According to the Kiel classification [3]
[b] CR, complete response; PR, partial response

the test dose was 5 for patients 7, 8, and 9, 4 for patients 10 and 11, and 2.5 for patients 12 and 13. Patients 10, 11, and 13 were studied during two subsequent courses of treatment 2 weeks apart. Finally, patients 14–18 received the same amount in the morning as in the afternoon.

2. Analysis of the Drug. The extraction and analysis of the drug and its metabolites were performed as already described [9]. Briefly, plasma was run on Sep-Pak C 18 cartridges, from which the anthracyclines were eluted with an organic solvent mixture. This extract was injected in a Waters liquid chromatograph, using a microbondapak-phenyl column and an isocratic eluting mixture of acetonitrile/formate buffer 32/68, approximately as described by Israël et al. [6]. Detection was performed by flow spectrofluorometry with a Schoeffel SF 970 instrument. Daunorubicin was added in known amounts to the plasma before extraction and was used as an internal standard.

3. Mathematical Processing of the Data. The experimental data were plotted on semi-logarithmic graph paper. The first five points (3–15 min) were always found to be in alignment, sometimes also with the last one. A linear regression analysis was performed and provided parameter A as the intercept and parameter α as the slope of the straight line, with a coefficient of correlation always higher than 0.99. The drug exposure during the first part of the pharmacokinetics can be approximated by the calculation of the early-phase clearance (E.C.) according to the equation:

$$\text{E. C.} = \frac{\text{dose injected}}{\frac{A}{\alpha}}.$$

The half-life ($t_{1/2}$) of the drug during the first 15 or 20 min after the injection was estimated as usual according to the equation:

$$t_{1/2} = \frac{\text{Ln } 2}{\alpha}.$$

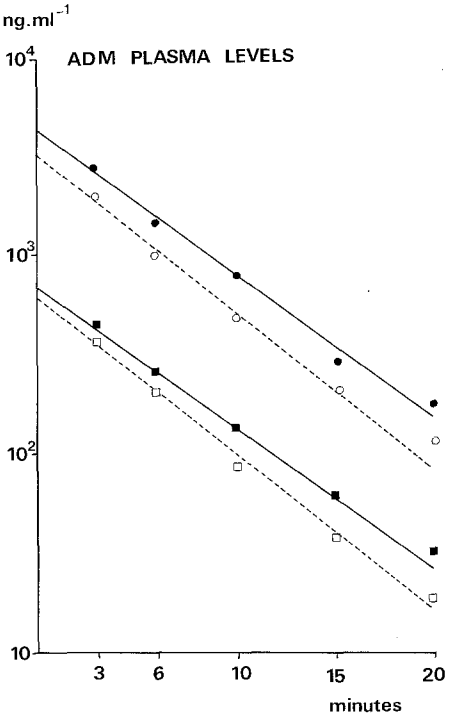

Fig. 1. Plasma levels of doxorubicin as a function of time in patient 2 during two courses of treatment 4 weeks apart. Two doses of 5 and 50 mg were performed on the same day for each course. (●——●, ◆——◆) First course of treatment; (○– – –○, ◇– – –◇) second course of treatment; (◇——◆) doses of 5 mg administered in the morning; (●——○) doses of 50 mg administered in the afternoon

In a process that is not time-dependent, the clearance of the drug is constant whatever the timing schedule of the injections; the linearity is the constancy of the clearance whatever the dose injected.

Results

When comparing the early clearances obtained during two successive courses of treatment 2 or 4 weeks apart (patients 2, 3, 10, 11, and 13) we observed an increase in this parameter in four cases, whether calculating from the test dose or from the therapeutic dose (Table 2). This increase ranged from 10% to 49% for the therapeutic doses and from 21% to 64% for the test doses. In one patient (patient 8), no change in the early clearance was observed for the therapeutic dose and only a small increase for the test dose. The first part of the kinetics of doxorubicin is therefore time-dependent in most patients over a 2-week interval; repeating similar doses in patients leads to a decrease of the drug exposure.

When two similar doses were administered on the same day (patients 14–18), in two cases we observed no change at all in the early clearance; in two other cases we observed a small difference (an increase of 13% and a decrease of 16%), and in one case an increase of 25% (Table 2). We cannot therefore assert that the pharmacokinetics of doxorubicin is not time-dependent within a 6-h period; however, since the differences are of lower magnitude and can be in either direction, we have assumed that the early-phase kinetics can be considered not to be time-dependent within a 6-h interval. Some of the results presented below corroborate this point of

Table 2. Values of the early clearances of doxorubicin in patients receiving two different doses at a 6 h interval during one or two courses of treatment

Patient	Course of treatment	1st dose (mg)	1st early clearance (ml/min)	2nd dose (mg)	2nd early clearance (ml/min)	Difference (%)
1	1st	5	1,251	50	1,750	+ 40
2	1st	5	1,180	50	2,028	+ 72
	3rd	5	1,542	50	2,893	+ 88
	Difference		+31%		+43%	
3	1st	4	1,332	40	2,210	+ 66
	2nd	4	2,186	40	3,094	+ 42
	Difference		+64%		+40%	
4	1st	5	1,153	50	1,829	+ 59
5	1st	5	782	40	2,153	+175
6	1st	10	2,690	80	2,901	+ 8
7	1st	10	2,405	50	2,489	+ 4
8	1st	10	1,981	50	2,532	+ 28
9	1st	10	1,304	50	1,598	+ 23
10	1st	10	1,574	40	1,962	+ 25
	2nd	10	1,806	40	1,900	+ 5
	Difference		+15%		−3%	
11	1st	10	1,257	40	1,549	+ 23
	2nd	10	1,517	40	1,698	+ 12
	Difference		+21%		+10%	
12	1st	20	2,687	50	2,616	− 3
13	1st	20	2,440	50	2,381	− 2
	3rd	20	3,634	50	3,459	− 5
	Difference		+49%		+45%	
14	1st	25	2,473	25	2,788	+ 13
15	1st	30	2,309	30	2,264	− 2
16	1st	25	1,545	25	1,296	− 16
17	1st	30	2,229	30	2,216	− 1
18	2nd	30	1,756	30	2,191	+ 25

Table 3. Values of the half-lives of the early-phase pharmacokinetics of doxorubicin in patients receiving two different doses 6 h apart during one or two courses of treatment

Patient	Course of treatment	1st dose (mg)	1st half-life (min)	2nd dose (mg)	2nd half-life (min)	Difference (%)
1	1st	5	3.81	50	4.28	+12
2	1st	5	4.28	50	4.15	− 3
	3rd	5	3.72	50	3.75	+ 1
	Difference		−13%		−10%	
3	1st	4	4.08	40	3.93	− 4
	2nd	4	3.92	40	4.08	+ 4
	Difference		−4%		+4%	
4	1st	5	3.97	50	4.27	+ 8
5	1st	5	4.92	40	4.12	−16
6	1st	10	4.07	80	4.70	+15
7	1st	10	3.57	50	3.63	+ 2
8	1st	10	3.27	50	3.55	+ 9
9	1st	10	4.17	50	5.43	+30
10	1st	10	4.93	40	5.05	+ 2
	2nd	10	3.95	40	4.35	+10
	Difference		−20%		−14%	
11	1st	10	5.05	40	5.39	+ 7
	2nd	10	4.60	40	4.25	− 8
	Difference		−10%		−21%	
12	1st	20	2.83	50	3.93	+39
13	1st	20	4.43	50	4.55	+ 3
	2nd	20	4.63	50	4.93	+ 6
	Difference		+5%		+8%	
14	1st	25	3.87	25	3.88	0
15	1st	30	4.90	30	5.32	+ 9
16	1st	25	4.42	25	4.25	− 4
17	1st	30	4.25	30	4.13	− 3
18	2nd	30	4.13	30	4.22	+ 2

view. We have therefore tried to determine the linearity of the kinetics within this 6-h interval.

In patients 1–6, receiving highly different dosages of doxorubicin in the morning and in the afternoon (the ratio therapeutic dose/test dose being 8 or 10), a large increase of the early clearance was noted, ranging from 40% to 175% (Table 2), except in one case (patient 6) for whom no significant change occurred. This increase was observed whatever the course of treatment studied. Increasing the dose by large proportions does not therefore lead to a similar increase in drug exposure. In patients 7–11, receiving two dosages at a ratio of 4 or 5, a much lower increase of the early clearance was observed (it ranged between 4% and 28%), which was not even significant in two cases. Finally, in patients receiving two doses at a ratio of 2.5, no difference was observed between the early clearances measured after the two injections (Table 2). The kinetics was linear for this dose ratio.

The half-lives of doxorubicin during this early-phase pharmacokinetics are presented in Table 3. In all cases but two, the half-life was constant for the injections 6 h apart. The half-lives were decreased by 10%–20% between the first course of chemotherapy and the following one in three cases (patients 2, 10, and 11) and were unchanged in the two other patients (patients 3 and 13).

The individual variation in the half-life of the early-phase kinetics is small, the ratio between extreme values being 1.9, while it reached 4.5 for the ultimate clearance values.

Discussion

The time-dependence of the pharmacokinetics of doxorubicin during the successive courses of treatment had been suggested by the work of several authors. Piazza et al. [7] showed a decrease of about 50% in the A parameter in two patients, with no significant change in the first half-life. In a third patient, they observed a two-fold increase of A concomitant with a significant decrease in the half-life, so that the calculated drug exposure during the first phase of the kinetics was unchanged at the fifth course of treatment. It should be pointed out, however, that in this third patient all the plasma levels measured at the fifth course of treatment were half the plasma levels measured during the first course of treatment, which in fact strongly suggests a decrease of the real drug exposure in this patient too.

The recent short communication of Gil et al. [5] does not give details about the first kinetic phase in the patient studied during three courses of treatment. An increase in the total plasma clearance of the drug between the first and the second courses is consistent with our observation. This clearance, however, returns to the initial value during the third course of treatment. In a previous study [4], we had observed a tendency for lower plasma levels of doxorubicin to be reached during successive courses of treatment with doxorubicin, suggesting that prior treatment with doxorubicin could have a prolonged effect on the fate of subsequent administration of the drug. From the results we present in this paper, and from other studies, it can be assumed that the pharmacokinetics of doxorubicin is time-dependent within the usual time intervals between two courses of treatment and that the clearance is generally increased after a first administration of the drug. This time-dependence may concern the intercept parameters or the half-lives, or both. We observed a decrease in the first half-life of the kinetics in three cases of five (patients 2, 10, and 11). P. Gil et al. (1982, personal communication) observed a decrease in this first half-life in the patient they studied [5], and Piazza et al. [7] observed such a decrease in one patient of three. The decrease in the A parameter was observed in our study in three patients of five (patients 2, 3, and 13); it was also observed in two of the three patients studied by Piazza et al. [7], but not in the patient studied by Gil et al. [5] (P. Gil, 1982 personal communication). Ehninger et al. [2] presented pharmacokinetic data recorded in a patient receiving two successive courses of chemotherapy consisting in three daily injections of doxorubicin. The half-lives and clearances were nearly constant from day to day during both courses, but a three-fold increase of the clearance was noticeable between the two courses of chemotherapy, the half-lives being decreased by 30%. However, in another patient, a decrease in the clearance between the first and the second courses was evident. The mechanisms by which the increase in clearance occurs in most cases remains unknown. Several mechanisms may occur, separately or together. For example, Gil et al. [5] have observed that an increased cumulative urinary excretion of the drug occurred in their patient from the first to the third course of treatment.

From our results, it is clear that the first part of the kinetics of doxorubicin in humans is dependent upon the injected dose. This dose-dependence only appears when a very small dose (5 or 10 mg) is compared with a therapeutic dose. Doses of 20 mg behaved similarly to doses of 50 mg. For doses of 5 mg, the early clearance of the drug was much less than expected from the behavior of doses of 50 mg. For doses of 10 mg, the clearance was closer to that observed for higher doses, but was significantly reduced in four cases of eight. This change in early clearance according to the dose is entirely due to the intercept parameter, since the half-lives changed in only two patients between the test dose and the therapeutic dose. The variations in doxorubicin pharmacokinetics according to the dose may be due to a saturation phenomenon, which might occur only from a dose of about 10 mg.

The individual variation in the pharmacokinetic parameters has been observed by several authors. The range of the early clearances we observed is significant, but much less so than that observed by Piazza et al. [7] and Ehninger et al. [2], who studied various types of cancer patients while our group of patients was homogenous and well-defined, none of them having received a prior chemotherapy. It must be emphasized that the early half-lives are much less scattered than the intercept parameters (after their correction for dose). It has not been possible to correlate any clinical parameter with the pharmacokinetic data as we did for locally advanced breast cancers [10]. This may be due to the absence of early objective assessment of tumor response to treatment. Moreover, the distribution and the size of the tumoral lymph nodes were not similar in the patients, and nor was the histology of the tumors, some of them being considered highly malignant and others less malignant [3].

The purpose of our study was to define a protocol for selecting individual doses of doxorubicin for patients according to their own pharmacokinetic pattern. The postulate of such a study is that the higher the drug exposure is, the better the patients will be treated. At least, such a study could make it possible to standardize drug exposures to allow comparison of the responses to treatments without the occurrence of individual variations of doxorubicin kinetics. Using a test dose of 5 mg leads to the introduction of a significant correction of the calculated theoretical dose so that the absence of linearity

of the kinetics can be taken into account. Using a test dose of 10 mg does not seem to require such a significant correction, the mean increase in the early clearance being 16%. This type of study has been undertaken in our Institute, the main difficulty being the definition of an optimal drug exposure.

Acknowledgements. This work has been supported by grants from INSERM (PRC Recherches Cliniques en Cancérologie, contract no. 119 009) and from the Fédération Nationale des Centres de Lutte contre le Cancer (Opération Vaincre le Cancer). We are grateful to the nurses of Fondation Bergonié for their cooperation in this study. Professor Cano's expert advice and his numerous suggestions and comments are gratefully acknowledged.

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Received May 17, 1982/Accepted November 5, 1982