Cancer Chemother Pharmacol (1998) 41: 347–352

ORIGINAL ARTICLE

**Leanne Embree · Karen Gelmon · Anthony Tolcher
Norma Hudon · Jean Heggie · Carole Dedhar
Patricia Logan · Marcel B. Bally · Lawrence D. Mayer**

Pharmacokinetic behavior of vincristine sulfate following administration of vincristine sulfate liposome injection

Received: 19 November 1996 / Accepted: 25 July 1997

Abstract The pharmacokinetic behavior of vincristine sulfate (VINC) following administration of vincristine sulfate liposome injection (VSLI), 0.16 mg/ml, as an intravenous infusion over 60 min in 24 of 25 patients enrolled in a phase I clinical study of this drug is described. Plasma samples for determination of the pharmacokinetic behavior of VINC were collected during the infusion at 15, 30 and 60 min as well as at 2, 4, 8, 12, 48 and 72 h postinfusion. Total VINC concentration was determined using a validated high-performance liquid chromatographic (HPLC) assay. Patients receiving doses of 0.5 to 1.5 mg/m^2 VSLI did not provide useful pharmacokinetic data at late time-points owing to the limit of quantitation of the HPLC assay (28.6 ng/ml). Sufficient concentration-time data were available for seven of the patients receiving doses of VSLI from 2.0 to 2.8 mg/m^2 for compartmental modelling. A two-compartment open model (PCNONLIN Model 10) was the best fit for the observed VINC plasma data for these patients. The mean maximum observed concentration values were significantly greater for patients receiving VSLI at 2.8 mg/m^2 (2260 ± 212 ng/ml, $n = 2$) than for those receiving 2.0 mg/m^2 and 2.4 mg/m^2 (891 ± 671 ng/ml, $n = 6$; 679 ± 634 ng/ml, $n = 6$, respectively). No significant differences were observed in maximum concentration values between patients at 2.0 mg/m^2 and those at 2.4 mg/m^2. A trend towards higher parametric AUC (0 to ∞) values with increasing dose (on a milligram per meter squared basis) was observed but statistical significance was not reached. Comparison of the pharmacokinetic behavior of VSLI observed in this study with nonencapsulated VINC demonstrated that (1) the variability observed for VSLI pharmacokinetic parameters was similar to nonencapsulated VINC, (2) although variability in absolute concentration was observed between patients, the behavior of VSLI in individual patients followed a two- rather than a three-compartment open model, and (3) VINC plasma concentrations were significantly greater following administration of VSLI than described for nonencapsulated VINC. Overall, the results for patients treated with VSLI from 2.0 to 2.8 mg/m^2 suggest that this formulation protects VINC from the early phase of rapid elimination seen with nonencapsulated drug, resulting in significantly elevated VINC plasma concentrations over extended periods of time.

Key words Vincristine sulfate · Liposome-encapsulated vincristine sulfate · Clinical pharmacokinetics

This work was supported in part by Inex Pharmaceuticals Corporation and the British Columbia Cancer Agency

L. Embree[1] · K. Gelmon · A. Tolcher · N. Hudon · J. Heggie
C. Dedhar · M.B. Bally · L.D. Mayer
Division of Medical Oncology, British Columbia Cancer Agency and Faculty of Medicine, University of British Columbia, Vancouver, British Columbia, Canada

L. Embree · L.D. Mayer
Faculty of Pharmaceutical Sciences, University of British Columbia, Vancouver, British Columbia, Canada

P. Logan
Inex Pharmaceuticals Corp., Vancouver, British Columbia, Canada

M.B. Bally
Faculty of Medicine, Department of Pathology, University of British Columbia, Vancouver, British Columbia, Canada

L.D. Mayer (✉)
Department of Advanced Therapeutics, BC Cancer Agency, 600 West 10th Ave., Vancouver, B.C., V5Z 4E6 Canada
Tel. +1-604-877-6000, ext. 3153; Fax: +1-604-877-6011;
E-mail: lmayer@bccancer.bc.ca

Present Address:
[1] Angiotech Pharmaceuticals, Inc. 303-2386 East Mall, Gerald McGavin Building, University of British Columbia, Vancouver, B.C., V6T 1Z3 Canada

Introduction

Vincristine (VINC) is a naturally occurring dimeric catharanthus alkaloid which has been used extensively as an antitumor agent since the 1960s. The accepted

mechanism by which VINC and other vinca alkaloids exert their antitumor activity is by interacting with tubulin which causes disruption of microtubules of the mitotic apparatus, thereby arresting cell division in metaphase [4]. Differences in the spectrum of activity, clinical efficacy and toxicity, as well as the pharmacokinetic behavior of vinca alkaloids, exist despite minor differences in the structure of these large alkaloid molecules. A considerable number of semisynthetic vinca alkaloid derivatives have been developed which demonstrate desirable pharmacological characteristics that have recently been evaluated [22].

A relationship between tumor responsiveness and the pharmacokinetic behavior of VINC has been found using human tumor xenograft models [10, 11]. These studies have demonstrated that the therapeutic activity of VINC increases significantly under conditions where extended drug circulation lifetimes are observed. This is consistent with in vitro observations indicating that increasing the exposure time of VINC to L1210 cells from 4 h to 72 h increases the antitumor potency of VINC by a factor of 10^5 [16]. Furthermore, the effect of potential resistance-modulating agents, for example verapamil, on VINC pharmacokinetics has received significant attention [11, 12]. Together with the availability of state-of-the-art analytical technology for antineoplastics, these issues have generated a resurgence of interest in the clinical pharmacology of these agents.

Early studies by Bender et al. [1] in humans involving the administration of radiolabelled VINC showed the importance of renal excretion and metabolism to the pharmacokinetics of VINC. Subsequent investigations to further characterize the metabolism of VINC and other vinca alkaloids [5, 13, 20, 25] have revealed modifications within the catharantine moiety with conservation of the dimeric structure. Postulated metabolites for VINC are the *N*-oxide, *N*-desformyl and desacetyl derivatives and their conjugates (glucuronide and/or sulfate).

A three-compartment open model appears to best describe the pharmacokinetic behavior of VINC [2, 18, 21]. However, some studies have not characterized the terminal elimination phase, as samples were not collected for sufficient time following administration [19, 23]. Using either administration of radiolabelled VINC or immunoassay methods for quantitation, the α, β, and γ half-lives reported range from 0.85 to 7.0, 7.4 to 164, and 164 to 5100 min, respectively. Recently, evaluation of VINC pharmacokinetics in children using a high-performance liquid chromatographic (HPLC) assay indicated that, in this population, VINC pharmacokinetic behavior can be described by a two-compartment model [6]. Differences between the specificity of the assays employed and patient populations evaluated may have contributed to the conflicting number of compartments required to fit the data. The majority of studies conducted to date have utilized three-compartment open models to describe the behavior of VINC, suggesting that the differences between two- and three-compartment modelling may reflect differences in the pharmacokinetic properties of VINC between children and adults.

Along with the development of VINC analogues and combinations of VINC with other agents to alter the observed pharmacokinetic behavior, liposomal formulations of VINC also offer potential to significantly improve therapy. One such liposomal formulation which has been tested clinically at the British Columbia Cancer Agency uses 120-nm diameter liposomes prepared from distearoylphosphatidylcholine (DSPC) and cholesterol at a molar ratio of 55:45 [16]. Previous preclinical studies with this and other liposomal formulations have demonstrated improvements in the toxicity and/or efficacy properties of VINC. These improvements arise from the ability of liposomes to alter the pharmacokinetic and tissue distribution properties of encapsulated VINC [7, 8]. Animal studies with small liposomes (less than 200 nm) that retain encapsulated VINC after intravenous (IV) injection indicate that there is a reduction in the accumulation and subsequent damage to healthy tissues, resulting in a modest reduction in drug toxicity [14, 15]. Of more significance is the dramatic increase in the antitumor potency of VINC when encapsulated in liposomes [3, 15–17, 24]. These increases correlate with plasma total VINC concentrations which are 100–1000-fold higher than those observed after administration of unencapsulated drug over 48 h postadministration [17, 24]. The elevated plasma concentrations also result in increased and extended exposure of tumors to VINC in vivo [17, 24]. These preclinical pharmacodynamic studies were possible only through the use of high specific activity preparations of tritiated VINC and combining the use of radiolabelled tracer with HPLC to assess the specificity of the radioactivity present in plasma for VINC [3, 17, 24]. Such approaches to evaluate the pharmacokinetic properties of liposome-encapsulated VINC in a clinical setting would be extremely difficult if not impossible to perform from a technical perspective, and would present serious ethical concerns.

Clinical evaluation of this VINC sulfate liposome injection (VSLI) formulation has been initiated based on its improved therapeutic activity compared to conventional VINC in several animal models [3, 15–17]. The phase I clinical study was designed to define the maximum tolerated dose and the spectrum of toxicity, as well as the pharmacokinetic behavior of VINC when adminstered IV in an encapsulated form. The preclinical observations for VSLI suggest that the use of a liposomal formulation would likely provide extended exposure to VINC in patients receiving VSLI. Owing to extensive metabolism of VINC [1, 13, 20, 25], the selectivity of an antibody or radiolabel-based assay for VINC may not be sufficient at extended plasma sampling times (24 h and beyond) to allow reliable determination of the pharmacokinetic parameters of the parent compound. In order to allow pharmacokinetic evaluation of VINC in human plasma with the necessary selectivity and sufficient sensitivity to quantitate VINC following

administration of VSLI, we have developed and validated an HPLC assay using ultraviolet detection for VINC [9]. This report describes the pharmacokinetic analysis of patients receiving VSLI as part of the phase I clinical study of this new agent. The pharmacokinetic behavior of VSLI is also compared with historical data reported for conventional, unencapsulated VINC.

Materials and methods

Patient selection

This study received approval from the Health Protection Branch, BCCA Medical Oncology Division, BCCA Clinical Investigations Committee, and the University of British Columbia Ethics Committee. All patients gave informed consent. Eligible patients had histologically confirmed malignant tumors, refractory to conventional forms of cancer therapy or for which no effective conventional therapy existed. All patients included in the trial had adequate bone marrow, liver, and renal function as defined by the following: absolute granulocyte count >1500 cells/μl, platelet count >100000 cells/μl, hemoglobin >8.0 g/l, total bilirubin <1.25 times normal, and serum creatinine of ≤ 1.25 times normal. The exclusion criteria were as follows: neurological disease, previous chemotherapy with vinca alkaloids, chemotherapy within 4 weeks of entering study (6 weeks for nitrosoureas, mitomycin C, phenylalanine mustard, or cisplatin), less than 18 years of age, sensitivity to VINC or vinca alkaloids, pregnancy or lactation, chronic severe liver disease. Patients who were deemed unable to comply with the protocol were also excluded.

Treatment plan and pharmacokinetic blood sampling

Patients received 0.5, 1.0, 1.5, 2.0, 2.4 or 2.8 mg/m^2 VSLI, 0.16 mg/ml, as an IV infusion over approximately 60 min. On the first dose of VSLI, blood samples were taken at the following times: pretreatment (two samples), 20, 40, 60 (or end of infusion) min, and 15, 30 and 60 min, and 2, 4, 8, 12, 24, 48 and 72 h postinfusion for a total of 15 samples per patient. Blood samples were collected in 7-ml EDTA Vacutainer tubes (Becton Dickinson Canada, Mississauga, Canada), plasma was prepared by centrifugation (10 min at $600 \times g$), and the plasma was stored at -20 °C until analysis.

Materials

Reagent grade diethylamine, potassium phosphate, monobasic (anhydrous), sodium phosphate, dibasic anhydrous *o*-phosphoric acid, and HPLC grade phosphoric acid, acetonitrile and methanol were obtained from Fisher Scientific Company (Vancouver, B.C.). HPLC grade water was prepared fresh daily with a Milli-Q system (Millipore Corporation, Bedford, Mass.). Ammonium hydroxide (97–99%) was purchased from Aldrich Chemical Company Inc. (Milwaukee, Wis.). Potassium chloride was obtained from Mallinckrodt Canada. (Pointe-Claire, Quebec). Undenatured ethanol (100%) was purchased from Stanchem (Vancouver). VINC sulfate (Oncovin) was purchased from Eli Lilly Canada (Scarborough, Ontario). Vinblastine sulfate USP was obtained from A + C American Chemicals (Toronto, Ontario) for use as the internal standard.

Preparation of vincristine sulfate liposome injection

VSLI, 0.16 mg/ml, is a three-part formulation consisting of DSPC/cholesterol liposomes for injection, 100 mg/ml, sodium phosphate for injection, 14.2 mg/ml, and Oncovin (vincristine sulfate for injection). Both the DSPC/cholesterol liposomes for injection, 100 mg/ml, and sodium phosphate for injection, 14.2 mg/ml, used in this study were prepared according to 'Good Manufacturing Practices' by the Medical Oncology Investigational Drug Section (IDS) at the British Columbia Cancer Agency (Vancouver). The Oncovin was used as purchased without modification. The combination of these three agents to form VSLI was used to characterize the recovery of VINC from plasma samples containing VSLI. The encapsulation procedure involved addition of 1 ml vincristine sulfate injection, 1 mg/ml, and 0.2 ml DSPC/cholesterol liposomes for injection, 100 mg/ml, into a sterile vial and mixing (by inverting the vial five times). Sodium phosphate for injection, 14.2 mg/ml, buffer solution (5 ml) was then added and the sample was mixed. The mixture was heated for 10 min at 63 °C (60–65 °C) in a waterbath and mixed. This encapsulation procedure was also used by the pharmacists for preparation of the VSLI administered to patients. This formulation was maintained under strict quality control which included testing both the liposomes and final VSLI. For the liposomes, the following parameters were evaluated (with typical results in parentheses) pH (3.98 to 4.00), osmolality (575 to 587 mOsm/kg), cholesterol content (27.0 to 27.4 mg/ml), phospholipid content (67.3 to 70.0 mg/ml), phospholipid purity (94.1 to 96.5%), particle size (0.127 to 0.131 μm). Similarly, VSLI was evaluated for the following: pH (7.21 to 7.25), osmolality (507 to 516 mOsm/kg), cholesterol content (0.96 to 0.99 mg/ml), phospholipid purity (88.7 to 94.1%), particle size (0.131 to 0.137 μm), VINC content (0.138 to 0.150 mg/ml), unencapsulated VINC (3.76 to 5.09%).

HPLC

All plasma samples were evaluated for VINC using a previously described validated HPLC assay [9]. Briefly, chromatography was carried out on a Symmetry C8 (4.6 mm ID $\times$ 250 mm) analytical column with a Delta-Pak C18 guard column (Waters Associates, Milford, Mass.). The mobile phase consisted of 34.9% HPLC grade water, 0.1% diethylamine adjusted to pH 7.0 with *o*-phosphoric acid, 40% acetonitrile and 25% methanol, and was used at a flow rate of 1.0 ml/min. The detection wavelength was 297 nm and the injection volume was 70 μl. Prior to solid phase extraction (87% recovery, see reference 9) chromatographic analysis, plasma samples were prepared by addition of the internal standard (400 ng vinblastine sulfate) to all patient and calibration standard samples with the exception of the pretreatment blank samples. A standard curve of peak area ratio values (VINC/internal standard) versus VINC concentration from the calibration standards was used for quantitation.

Pharmacokinetic calculations

For analysis of VSLI pharmacokinetics, the concentration of total VINC was determined in all plasma samples as described above. Initial pharmacokinetic parameter estimates were calculated and iterative least squares estimation was carried out with PCNONLIN V4.2 (SCI Software, Lexington, Ky.) for one-, two- and three-compartment open models with constant IV input, first-order output (PCNONLIN Models 2, 10 and 19) and no weighting of data. Data from each patient were evaluated with these three models. The model which most accurately described the data was determined by the AIC Criteria provided by PCNONLIN as well as an overlay of the PCNONLIN model and the individual observed concentration values versus time for each patient. Analysis of variance and one-tailed *t*-tests were used for statistical evaluation of the pharmacokinetic data.

Results

Total VINC plasma concentrations were evaluated in 24 patients receiving VSLI. Sensitivity limitations of the

assay prohibited pharmacokinetic modelling of the data from patients receiving low doses of VSLI, from 0.5 to 1.5 mg/m^2. The mean Cmax (observed) for patients in the 0.5, 1.0 and 1.5 mg/m^2 groups were 99.7, 303 and 374 ng/ml, respectively. After 1–3 h following initiation of VSLI administration, the VINC plasma concentration values were below the limit of quantitation for the HPLC procedure and consequently insufficient data were available to pharmacokinetically model the elimination behavior of this formulation. At higher doses, specifically 2.0, 2.4 and 2.8 mg/m^2 VSLI, sufficient concentration-versus-time data were available for 7 of the 15 patients to enable parametric evaluation of the behavior of VSLI at these doses.

For the remaining 8 of 15 patients, although chromatographic peaks corresponding to VINC were obtained for extended time periods, alterations in the chromatography of the internal standard (vinblastine) precluded accurate quantitation and subsequent pharmacokinetic analysis of VINC. This phenomenon, which was manifested as a shift of the internal standard peak to earlier elution times, occurred only at later time-points (8 h and beyond) and could not be attributed to endogenous patient plasma components as no changes in internal standard chromatography were observed in pretreatment samples. Interestingly, the addition of exogenous VINC to such samples indicated that such a shift in retention time did not occur for VINC. It is important to note that peak areas corresponding to VINC in these patients compared favorably with the seven patients for whom no internal standard complications were observed.

After administration of VSLI, an initial rapid decline in plasma VINC concentrations was observed which represents a distribution phase. The half-life for this phase of elimination ($t_{1/2\alpha}$) was rapid with respect to subsequent elimination of VINC from the body as represented by the half-life of the terminal elimination phase ($t_{1/2\beta}$). Thus, the plasma concentration of VINC followed a biexponential decline after the infusion of VSLI and concentration-time data were fitted to a two-compartment open model. The pharmacokinetic data for patients receiving 2.0 to 2.8 mg/m^2 VSLI are shown in Table 1 and Fig. 1. The Cmax values shown in Table 1 include all patients studied, while the remaining parameters relate only to patients in whom parametric estimates were possible. The limit of quantitation of the

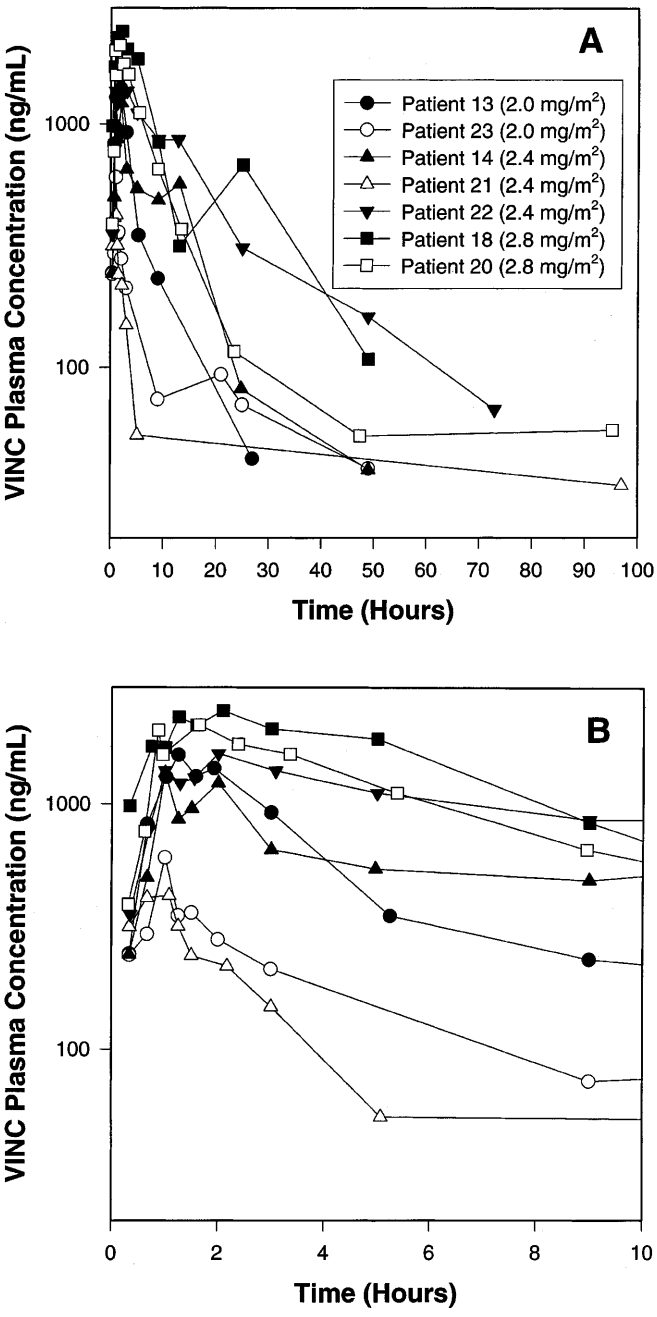

Fig. 1A,B Plasma concentrations versus time for patients receiving VSLI at 2.0, 2.4 and 2.8 mg/m^2. All data (**A**) and the initial distribution phase (**B**) are represented

Table 1 Summary of selected plasma pharmacokinetic parameters. Values are means (SD). Cmax(observed) values are nonparametric while the remaining parameters are based on the pharmacokinetic model

Parameter	2.0 mg/m^2 patients	2.4 mg/m^2 patients	2.8 mg/m^2 patients
$t_{1/2\alpha}$ (min)	117 (101, $n=2$)	317 (472, $n=3$)	434 (286, $n=2$)
$t_{1/2\beta}$ (min)	1350 (591, $n=2$)	555 (390, $n=3$)	5960 (453, $n=2$)
AUC(0 to ∞) (μgmin/ml)	389 (36.8, $n=2$)	890 (829, $n=3$)	2410 (1030, $n=2$)
Cmax[a] (Observed) (ng/ml)	891 (671, $n=6$)	679 (634, $n=6$)	2260 (212, $n=2$)
Vc (ml)	4350 (841, $n=2$)	3220 (938, $n=3$)	2260 (467, $n=2$)
K_{21} (h^{-1})	0.0428 (0.0344, $n=2$)	0.696 (0.870, $n=3$)	0.0159 (0.00356, $n=2$)

assay resulted in insufficient data for parametric estimates for all patients.

The mean values for AUC (0 to ∞), half-lives and Cmax (observed) were evaluated. Mean values for AUC(0 to ∞) ($P = 0.20$, 0.11 and 0.11) and $t_{1/2\alpha}$ ($P = 0.28$, 0.19 and 0.38) were not significantly different for the three dose groups (P-values from comparison of 2.0 and 2.4, 2.4 and 2.8, 2.0 and 2.8 mg/m^2, respectively). Significant differences in the mean $t_{1/2\beta}$ values were observed between patients receiving VSLI at 2.0 and those receiving 2.8 mg/m^2 ($P = 0.006$) as well as between the 2.4 and 2.8 mg/m^2 patient groups ($P = 0.003$). The difference between $t_{1/2\beta}$ values for the 2.0 and 2.4 mg/m^2 patient groups was not significant ($P = 0.12$). Similarly, the mean values for Cmax (observed) were significantly different between the 2.0 and 2.8 mg/m^2 patient groups ($P = 0.002$) as well as between the 2.4 and 2.8 mg/m^2 patient groups ($P = 0.0009$). In contrast, patients receiving 2.0 mg/m^2 and those receiving 2.4 mg/m^2 VSLI had Cmax values that were not significantly different ($P = 0.29$).

For all patients at the 2.0 mg/m^2 to 2.8 mg/m^2 VSLI dose levels, the mean values for α and β half-lives, AUC (0 to ∞) and Cmax (observed) were 293 $\pm$ 327 min. ($n = 7$), 2330 $\pm$ 2540 min. ($n = 7$), 1180 $\pm$ 1070 μgmin/ml ($n = 7$) and 996 $\pm$ 793 ng/ml ($n = 14$), respectively. This variability in the pharmacokinetic parameters describing the behavior of VSLI ranged from a relative standard deviation (RSD) of 80% for Cmax (observed) to 112% for $t_{1/2\alpha}$. There was no significant difference in the variability of the AUC (0 to ∞) or α half-life values for these three dose levels ($P = 0.12$ and 0.70, respectively). The variances for both the β half-life ($P = 0.0005$) and Cmax (observed) ($P = 0.0005$) were significantly different for the three dose groups. The difference observed in the variances of the β half-life was a result of the similarity between the two patients dosed at 2.8 mg/m^2. The statistical variance for the β half-life values for the patients receiving VSLI at 2.0 mg/m^2 and those receiving 2.4 mg/m^2 were not significantly different ($P = 0.16$), while significance was demonstrated for this parameter between patients receiving 2.4 mg/m^2 and those receiving 2.8 mg/m^2 ($P = 0.007$). For observed Cmax values, however, (see Table 1 and Fig. 1B) significant differences were found between the 2.0 and 2.4 mg/m^2 patient groups ($P = 0.0007$), between the 2.4 and 2.8 mg/m^2 patient groups ($P = 0.0007$) and between the 2.0 and 2.8 mg/m^2 patient groups ($P = 0.04$). Administration of VLSI produced a trend towards higher AUC (0 to ∞) values with increasing dose (on a milligram per meter squared, basis), but statistical significance was not reached.

Discussion

Total VINC plasma concentration values for patients treated with VSLI reported here are the sum of both encapsulated and nonencapsulated VINC. Owing to the rapid elimination behavior of nonencapsulated VINC, which has been previously reported [1, 2, 6, 18, 19, 21, 23], the major part of each total VINC concentration value is likely to have been encapsulated VINC. The sensitivity of the selective assay for VINC used for this work precluded pharmacokinetic evaluation of nonencapsulated VINC over the 72 h postinfusion time course evaluated here owing to the extremely low concentrations of drug released from the liposomes [17].

The observed variability in the pharmacokinetic behavior of VSLI (RSD ranging from 80% for Cmax (observed) to 112% for $t_{1/2\alpha}$ is comparable to that reported for nonencapsulated VINC. Specifically, both De Graaf et al. [6] (RSD 102% for Vc, $n = 17$) and Nelson [18] (RSD 88% for K_{20}, $n = 4$) observed a variability for nonencapsulated VINC similar to that reported here for VSLI. For VSLI, the observed variability in pharmacokinetic parameters is a combination of the variability for unencapsulated drug plus that for liposome clearance and release of encapsulated drug as previously observed with a liposomal formulation of doxorubicin [8]. It is important to note, however, that this variability was not related to the liposome formulation characteristics (see Materials and methods) or complications observed with the internal standard for some patients. With regard to the latter, absolute peak areas corresponding to VINC in these patients would predict pharmacokinetic properties of VSLI comparable to those reported in Table 1. Unfortunately, the nature of the internal standard chromatographic changes in these patients could not be resolved, and hence the accuracy of associated pharmacokinetic analyses could not be assured.

For a 2-mg dose of radiolabelled VINC administered over 1 min, Bender et al. [1] reported whole-blood pharmacokinetics based on evaluating the radioactivity of blood. At 210 min postadministration, a whole-blood VINC concentration of approximately 15 nmol/l (12 ng/ml) was observed. The patients, in the present study received VSLI (doses ranging from 3.0 to 6.2 mg for the 2.0, 2.4 and 2.8 mg/m^2 dose groups) as a 60-min infusion. At approximately 210 min postadministration (270 min) or later, the VSLI patients had a mean VINC concentration of 547 ng/ml with a range from below the limit of quantitation (28.6 ng/ml) to 1850 ng/ml (Fig. 1B). These VINC concentrations are 2.5- to 154-fold higher than those previously reported [1]. The VINC pharmacokinetics described by Bender et al. [1] were derived using an assay that is known to be nonselective for quantitation of VINC so that the increased concentration seen for VSLI reflect minimum values.

Differences in administered dose, infusion time or assay specificity do not account for the observed large difference in concentration of VINC following administration of the two formulations. Plasma VINC concentration values following administration of VSLI appear to be greater than conventional VINC because of a difference in elimination. Van den Berg et al. [23] and Owellen et al. [19] did not provide sufficient data for each patient to allow elimination characteristics for

non-encapsulated VINC to be compared with the pharmacokinetic parameters obtained for VSLI. However, comparison of the terminal elimination half-life observed in this study for VSLI with that for nonencapsulated VINC described by Nelson [18] in adults using an immunoassay for quantitation demonstrates that there is no significant difference ($t_{1/2\gamma}$ of 85.0 h, $P = 0.16$). This suggests that encapsulation of VINC reduces or eliminates the initial rapid decline in concentration while maintaining terminal elimination characteristics similar to nonencapsulated VINC. There was, however, a significant difference between the terminal elimination half-life observed here for VSLI and that reported by De Graaf et al. [6] for VINC in children ($P = 0.03$) as reflected by a VSLI/VINC half-life ratio of 2.83. The difference between the values reported by de Graaf et al. [6] and those reported by Nelson [18] may be explained by the fact that de Graaf et al. used a selective HPLC assay for VINC analysis and studied children while Nelson [18] reported pharmacokinetic data for adult patients derived using an immunoassay. Nevertheless, the results obtained in this study suggest that the primary effect of liposome encapsulation on VINC pharmacokinetics is the loss of the initial distribution/elimination phase(s). This accounts for the apparent shift from a three-compartment model to a two-compartment model and leads to significant increases (approximately 45-fold) in plasma VINC concentrations over extended times, consistent with results from preclinical studies [16, 17]. Such observations in conjunction with preclinical correlations between VINC circulation lifetimes and antitumor activity provide encouraging signs that the hoped-for improved therapeutic activity of VSLI relative to unencapsulated VINC may be attainable.

Acknowledgements Funding for this project was obtained from Inex Pharmaceuticals Corporation and the British Columbia Cancer Agency.

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