Intraindividual variability in busulfan pharmacokinetics in patients undergoing a bone marrow transplant: assessment of a test dose and first dose strategy

Celeste Lindley[a], Thomas Shea[b], Jeannine McCune[a], Stacy Shord[a], Jodi Decker[a], Donald Harvey[a], William P. Petros[c], Don Garbriel[b], Jonathan Serody[b], Suzanne Kirby[b] and Joseph Wiley[b]

Busulfan systemic exposure correlates with regimen-related toxicity, engraftment and relapse in select patients receiving high-dose oral busulfan (HD-BU) (1 mg/kg dose or 40 mg/m^2 dose every 6 h for 16 doses) as part of a preparative regimen for bone marrow transplantation. Therapeutic drug monitoring is frequently conducted after the first HD-BU dose in order to determine necessary dose adjustments. Limitations with this method include the need for rapid determination of busulfan plasma concentration and difficulties with estimating apparent oral clearance in patients who exhibit delayed absorption of HD-BU. This pharmacokinetic study was conducted to evaluate the ability of the apparent oral clearance obtained after administering a lower (0.25 mg/kg) test dose and the traditional (1 mg/kg) first dose to predict the dose required to achieve a desired area under the concentration–time curve (AUC) at steady-state (13th dose). In addition, the pharmacokinetic parameters of test, first and 13th dose were compared to assess intrasubject variability. Twenty-nine patients received a test dose of oral busulfan (0.25 mg/kg) the day immediately prior to initiation of HD-BU. Busulfan serum concentrations were measured following the test, first and 13th doses using gas chromatography with electron capture detection. The AUC and apparent oral clearance were calculated using non-compartmental analysis. Therapeutic drug monitoring following the first dose of HD-BU was conducted for clinical purposes in six patients, and dose adjustment between the first and 13th dose occurred in only two patients. The dose-corrected test dose and first dose AUC and apparent oral clearance were not bioequivalent (two one-sided *t*-tests, ±20%). The first dose and 13th dose AUC and apparent oral clearance were also not bioequivalent. Six of the 29 patients receiving HD-BU dose based on weight (1 mg/kg) would have achieved a steady-state AUC of 3600–5400 ng·h/ml, a frequently used target AUC, as compared to eight and 13 patients if their dose was based on the apparent oral clearance following the test dose and first dose HD-BU, respectively. Monitoring busulfan concentrations after a test dose or a first dose provides a better estimate of the dose needed to achieve the target steady-state AUC as compared to traditional weight-based dosing. However, significant intraindividual variability exists in the apparent oral clearance of busulfan and follow-up therapeutic drug monitoring is recommended particularly if the desired target AUC range is narrow. *Anti-Cancer Drugs* 15:453–459 © 2004 Lippincott Williams & Wilkins.

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Schools of [a]Pharmacy and [b]Medicine, University of North Carolina at Chapel Hill, Chapel Hill, NC, USA and [b]Duke University Medical Center, Durham, NC, USA.

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Correspondence to C. Lindley, School of Pharmacy, University of North Carolina, CB #7360, Beard Hall, Chapel Hill, NC 27599-7360. USA.
Tel: +1 919 962-0028; fax: +1 919 962-0644;
e-mail: celeste_lindley@unc.edu

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Introduction

High-dose busulfan (HD-BU) is commonly used in preparative regimens for patients undergoing allogeneic bone marrow transplantation. The standard HD-BU regimen is administered based on a fixed dose of 1 mg/kg orally every 6 h for 4 days (total dose of 16 mg/kg) [1]. Children less than 6 years old typically receive busulfan 40 mg/m^2 orally every 6 h for 4 days (total dose of 640 mg/m^2) [2]. The intersubject coefficient of variation in busulfan apparent oral clearance in adults is 23% [3] and the busulfan area under the concentration–time curve (AUC) varies 3- to 7-fold in patients receiving HD-BU dose on weight or body surface area [2,4,5]. The steady-state AUC for HD-BU has been correlated with regimen-related toxicity, engraftment and relapse in select patients receiving the busulfan/cyclophosphamide preparative regimen. These pharmacodynamic relationships, along with the substantial interpatient variability in the steady-state AUC after fixed dose administration, have led several centers to conduct therapeutic drug monitoring for HD-BU in bone marrow transplant patients [6,7]. A busulfan AUC of 3600–5400 ng·h/ml in partially

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matched or unrelated bone marrow transplant recipients and 1200–5400 ng·h/ml in allogeneic matched sibling bone marrow transplant patients receiving the busulfan/cyclophosphamide preparative regimen is desired to minimize toxicity and prevent rejection [4,8–12].

Therapeutic drug monitoring includes collection of serial blood samples over the 6 h between the first and second dose of HD-BU. Busulfan concentrations are quantitated to calculate AUC and guide dosage adjustment to achieve the target AUC. Dose adjustments are required in 25–50% of patients receiving HD-BU [8,13]. However, 12–26% of patients have delayed absorption, which hinders accurate estimation of AUC [8,13] and rapid dose adjustments (i.e. the by fourth to fifth HD-BU dose), which are related with improved outcomes [11,14].

This pharmacokinetic study was undertaken to (i) determine the ability of the test dose and first dose AUC to predict the dose required to achieve the steady-state AUC (i.e. 13th dose) in bone marrow transplant patients, and (ii) compare pharmacokinetic parameters of the test, first and 13th dose (i.e. steady state).

Methods

This was an open-labeled non-randomized pharmacokinetic study in patients undergoing a bone marrow transplantation using HD-BU (40 mg/m^2 dose in children less than 6 years or 1 mg/kg dose in older patients, administered orally, every 6 h for 16 doses) as part of their preparative regimen at UNC Hospitals. Busulfan plasma concentrations obtained from this investigation were neither available nor intended for use in clinical HD-BU therapeutic drug monitoring. Actual body weight was used to dose busulfan, except adjusted ideal body weight (ideal body weight plus 25% of the difference between actual and ideal body weight) was used in obese patients (body mass index above 27 kg/m^2) [3]. Patients who required lorazepam during the test dose or HD-BU were excluded due to assay interference. Approval of the Lineberger Cancer Center Protocol Review Committee and the UNC Committee on the Protection of the Rights of Human Subjects was obtained prior to study initiation, and written informed consent was obtained in all patients.

The test dose (0.25 mg/kg) was administered before 12:00 noon on the day immediately prior to starting HD-BU, and the first HD-BU dose and 13th HD-BU dose were each administered at 6:00 a.m. As per standard of care in patients receiving HD-BU, seizure prophylaxis with phenytoin was started 12 h after administering the test dose. Antiemetics were not required or administered during HD-BU. Blood samples were drawn at the following times: 0, 0.5, 1, 1.5, 2, 3, 4, 5, 6, 8 and 12 h following the test dose, and 0, 0.5, 1, 1.5, 2, 3, 4, 5 and 6 h following the first and 13th doses as necessitated by the need for repeat dosing at 6 h during HD-BU administration. Samples were centrifuged in a swinging bucket rotor to separate plasma from red blood cells and were stored at –70°C until further analysis. A Hewlett-Packard 6890 gas chromatograph with Ni electron capture detection was used to determine the busulfan concentrations. The internal standard [1,8-bis(methanesulfonyloxy)] was added to the plasma samples before they underwent liquid extraction and derivitization with 2,3,5,6-tetrafluorothiopenol. The lower limit of quantitation was 0.04 μg/ml, and the assay was linear between 0.05 and 2 μg/ml. Within day and between day variability, measured by coefficient of variation, were below 10% for all busulfan concentrations [15].

The AUC, apparent oral clearance (CL/F) and volume of distribution (V_d) were calculated using a non-compartmental model (WinNonlin software, Professional Version; Pharsight, Mountain View, CA). The apparent oral clearance was determined using the AUC from time 0 extrapolated to infinity for the test and first dose, and from time 0 to 6 h for the 13th dose (steady state). The dose range that would have achieved an AUC of 3600–5400 ng·h/ml was determined using the patient's apparent oral clearance following the 13th dose and the following equations derived from this established equation: dose = AUC × CL/F.

- 13th dose CL/F (ml/h) × 3600 ng·h/ml: this is the dose needed to achieve an AUC of 3600 ng·h/ml (i.e. the minimal therapeutic dose)
- 13th dose CL/F (ml/h) × 5400 ng·h/ml: this is the dose needed to achieve an AUC of 5400 ng·h/ml (i.e. maximal therapeutic dose)

The predicted dose to achieve the desired midpoint steady-state AUC (4500 ng·h/ml) was calculated from the apparent oral clearance for the test dose and first dose using the following equation:

- Test dose or first dose CL/F (ml/h) × 4500 ng·h/ml: this is the dose needed to achieve AUC of 4500 ng·h/ml (i.e. midpoint therapeutic dose)

The apparent oral clearance determined following the test and first doses were compared to the apparent oral clearance determined following the first and 13th doses, respectively, using two one-sided *t*-tests (TOST) using SAS software (version 6.12; SAS Institute, Cary, NC). TOST, the preferred statistical test of bioequivalence, determines the 90% confidence interval for the ratio between the test dose and first dose AUC and the 13th dose AUC, which by definition cannot exceed the defined pharmaceutical definition of bioequivalence (80–120%) [16]. An *a priori* level of significance ($\alpha = 0.05$) was used for analysis. With 30 patients, each one-sided *t*-test had 98% power to reject the null hypothesis with an overall power of 96%. This power analysis is based on an estimate

of 20% intrinsic variability that equals the standard deviation of 0.112 on a log scale.

The predictive performance of the test and first doses was determined using the steady-state AUC calculated from the 13th dose as the gold standard. The AUC calculated for the test and first doses were used to assess precision and bias. Precision was quantitated using absolute percent error [|(test dose AUC or first dose AUC–13th dose AUC)|/13th dose AUC × 100)] and bias was quantitated using the actual percent error [(test dose AUC or first dose AUC–13th dose AUC)/13th dose AUC × 100].

Results

Patient characteristics

Thirty patients were enrolled in this study between July 1998 and July 2001 (Table 1). Data from one patient was excluded due to analytical equipment failure and, therefore, this analysis includes 29 patients. Two patients were less than 6 years of age. Chronic and acute leukemia were the most common indications for bone marrow transplantation. Eleven of 29 patients (38%) received partially matched or unrelated donor allogenic transplants.

Comparison of test, first and 13th dose pharmacokinetic parameters

The mean (SD) AUC and apparent oral clearance following the test, first and 13th doses are found in Table 2. Figures 1 and 2 illustrate each subject's dose-adjusted test, first and 13th dose apparent oral clearance (ml/h) and AUC (ng·ml/h). Five patients were omitted from the TOST statistical analysis. Four due to delayed absorption after the first dose, and therefore lack of a clear elimination phase, and one due to assay difficulties with the 13th dose. Neither the dose-corrected AUC nor the apparent oral clearance were bioequivalent (TOST ± 20%, $p > 0.05$) for the test and first dose nor the test and 13th dose. Precision and bias (median and range) were 36% (1–12%) and 33% (−63 to 120%) for the test dose AUC (dose corrected) compared to steady-state AUC measured following the 13th dose, respectively. The test dose predicted the first dose AUC with a precision and bias of 10% (86–1%) and −1% (−24 to 86%), respectively. The TOST analysis similarly found that neither the apparent oral clearance or the AUC of the first dose and 13th dose were bioequivalent. Precision and bias were 35% (0–100%) and 4% (−100 to 68%) for first dose prediction of the 13th dose AUC, respectively.

Table 1 **Patient characteristics**

Characteristic	*N*
Total no. of patients	29
Gender: male/female	19/10
Race: Caucasian/African-American/other	19/8/2
Age (years)[a]	39 (4–56)
Age: pediatric[b]/adult	2/27
Diagnosis	
acute leukemia	8
chronic leukemia	15
lymphoma	3
other	3
Transplant	
allogeneic: partially matched or unrelated[c]	11
allogeneic: matched sibling	17
autologous	1

[a]Median (range).
[b]Pediatric patients were ≤ 6 years old.
[c]Therapeutic drug monitoring, $n = 9$; dose adjustments, $n = 2$.

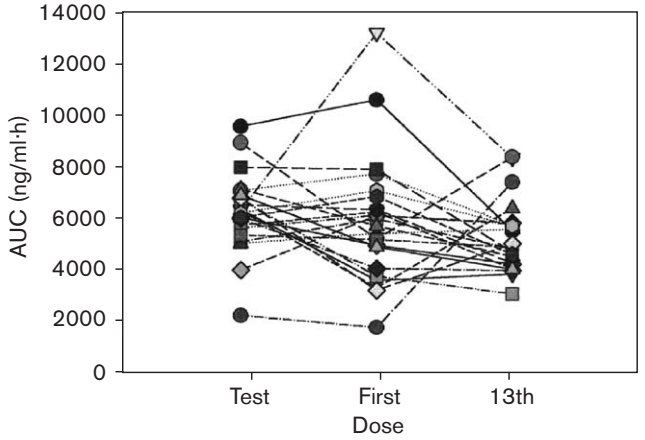
Table 2 **AUC and apparent oral clearance of busulfan test dose, and the first and 13th busulfan therapeutic dose [mean (SD)]**

Dose	AUC (ng·h/ml)	Apparent oral clearance (ml/h)
Test	2072 (1016)	9247 (2938)
1	6791 (2858)	10694 (4169)
13	6049 (1685)	11218 (4030)

Table 3 illustrates the number of patients who were within the desired target AUC following the first dose and 13th dose. The majority of patients exceeded the upper limit of the desired AUC following the first dose and 50% or less of patients were within the desired target range, although more patients were within the target AUC following the 13th dose. It is important to note that real-time analysis of busulfan concentration and attempts to target a desired AUC was not performed in this patient population. The majority of patients (23 of 29) included in this analysis did not have therapeutic drug monitoring performed, as 17 of the 29 patients received an allogeneic matched sibling bone marrow transplantation which allows for a lower desired busulfan AUC. Busulfan dose adjustments during the 4-day regimen occurred in only two patients.

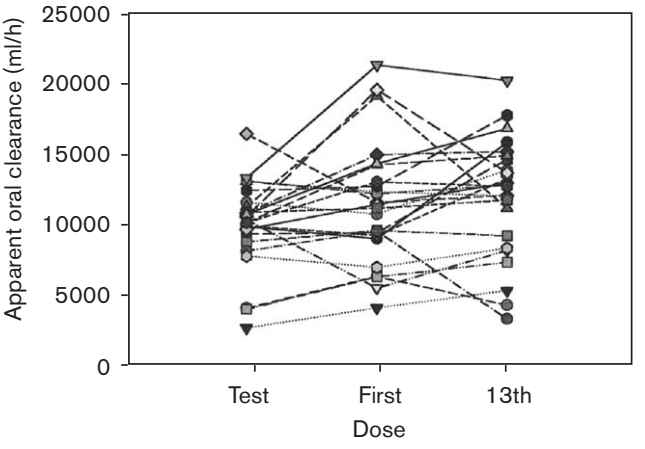
Further analysis was conducted to determine the ability of the apparent oral clearance of the test and first doses to predict the dose range required to achieve the target AUC in the range of 3600–5400 ng·h/ml at steady state (i.e. 13th dose). Table 4 shows the predicted dose calculated using the test dose and first dose apparent oral clearance that would have achieved the midpoint of the desired AUC range between 3600 and 5400 ng·h/ml compared to the predicted dose that would have achieved the lower and higher target AUC based on the apparent oral clearance at steady state (13th dose). The apparent oral clearance determined for the test dose successfully predicted the dose required to achieve the midpoint of the steady-state AUC in eight of 24 patients (33%). Similarly, the apparent oral clearance determined for the first dose successfully predicted the dose needed to achieve the target AUC in 13 of 24 patients (54%). However, four of the five patients omitted from this analysis were excluded due to delayed absorption and the inability to accurately estimate the AUC from the first dose. Thus, it is more accurate to use the estimate that

Fig. 1



Comparison of AUC calculated from test dose, first dose and 13th dose of HB-BU ($n=24$).

Fig. 2



Comparison of apparent oral clearance calculated from test dose, first dose and 13th dose of HB-BU ($n=24$).

Table 3 AUC determined for first dose and 13th dose

AUC (ng·h/ml)	First dose[a]	13th dose[b]
Matched related ($n=14$)		
<1200	0 (0%)	0 (0%)
1200–5400	3 (22%)	5 (36%)
>5400	11 (78%)	9 (64%)
Partially matched or unrelated ($n=10$)		
<3600	0 (0%)	0 (0%)
3600–5400	3 (30%)	6 (60%)
>5400	7 (70%)	4 (40%)

[a]Four patients were omitted from first dose analysis due to lack of elimination data.
[b]One patient was omitted from analysis for inadequate sample collection.

Table 4 Steady-state dose prediction with test dose and first dose apparent oral clearance

Predicted dose to achieve AUC 4500 ng·h/ml		Predicted dose to achieve target AUC using apparent oral clearance of the 13th dose	
Test dose (mg)	First dose (mg)	3600 ng·h/ml	5400 ng·h/ml
41	37	54	81
49[a]	45[b]	47	70
41[b]	38	45	67
43[b]	22	27	41
39	39	50	75
36[a]	40[a]	31	46
70	49[a]	41	61
40[b]	48[a]	44	66
55[a]	52[a]	41	61
55[a]	51[a]	43	65
52	54	61	91
34	40	10	15
16[a]	26	13	20
56	91[a]	69	104
10	16[b]	17	26
46[a]	47[a]	40	59
16	26[a]	24	36
44[b]	84	46	69
45[b]	63[a]	52	77
42	60[a]	51	76
45	61[a]	57	86
32[a]	29[a]	28	41
42[b]	55[a]	43	64
47[a]	82	37	56

[a]Denotes successful prediction of dose to achieve a steady-state target AUC between 3600 and 5400 ng·h/ml.
[b]Within 10%.

first dose apparent oral clearance predicted the dose needed to achieve the target AUC in 13 of 28 patients (46%).

Discussion

HD-BU is an important component of preparative regimens for patients undergoing hematopoietic stem cell transplant. The majority of studies have demonstrated a pharmacodynamic relationship between busulfan's AUC or C_{ss} and the outcome for patients receiving busulfan/cyclophosphamide preparative regimens [9,8,11–12]. Considerable intersubject variability exists in HD-BU pharmacokinetics, and age and obesity have been identified as independent predictors of busulfan clearance [3,9]. However, the relatively narrow therapeutic window supported by the current literature of an AUC of 3600–5400 ng·h/ml (C_{ss} = 600–900 μg/l) in partially matched or unrelated transplant recipients is not consistently achieved even when these factors are taken into account. Ideally, differences in intersubject variability could be reduced by either improving our understanding of the metabolic fate of busulfan, by *a priori* dose adjustments or by estimating apparent oral clearance after a test or first dose.

Although a host of factors could theoretically account for the interindividual differences in busulfan pharmacokinetics, with the exception of age and body weight, no factors have been conclusively shown to affect clearance.

In addition, the metabolic rate of busulfan has not been fully characterized. Currently, conjugation of busulfan by glutathione (GSA) to form γ-glutamyl-β-(*S*-tetrahydrothiopheniumion)alanyl-glycine (THT^+) is the only recognized biotransformation pathway for busulfan. Glutathione S-transferase (GST) catalyzes THT^+ formation. GSTA1-1, the major liver GST, is the predominant GST involved in busulfan's conjugation [17,18].

Age-related differences in GST expression have been implicated as a potential cause for the age-related differences in busulfan AUC [19]. However, clinically significant interpatient and intrapatient variability in glutathione content and GST induction are currently not recognized. Interestingly, the conjugation of busulfan in the intestinal epithelial cells from young children (1–3 years) was reported to be 77% higher relative to older children (9–17 years), suggesting that the oral bioavailability may be substantially reduced in young children [18]. Systemic exposure to busulfan has been anecdotally reported to decrease, increase and not change with concomitant administration of cytochrome P450 enzyme inducers and inhibitors, respectively [20,21]. However, studies in animals and human microsomes have failed to show that cytochrome P450 enzymes are involved in the biotransformation of busulfan [22].

Intraindividual variability in AUC presents far greater complexity than interindividual differences when one considers the desirability of therapeutic drug monitoring. The current study focused on the ability to use apparent oral clearance estimated from test dose and first dose to predict the 13th dose AUC. The potential advantages with the use of a test dose include convenience in terms of patient and personnel scheduling, and a potentially longer time during the 96-h HD-BU regimen when patients would be within the desired therapeutic range. The use of a test dose (0.5 mg/kg), administered 4 days prior to hematopoietic stem cell transplantation in 34 pediatric patients, resulted in 54% having a target AUC of 3600–5400 ng·h/ml following the first dose of HD-BU as reported by Bolinger *et al.* in abstract form [14]. In addition, Bleyzac *et al.* observed in 29 children undergoing hematopoietic stem cell transplantation that the expected AUC calculated from administration of a test dose (0.5 mg/kg) 2–124 days prior to hematopoietic stem cell transplantation correlated well ($r^2 = 0.7914$) with the average of the observed AUC over 4 days [23]. These investigators applied Bayesian pharmacokinetics monitoring over the 4 days of busulfan treatment with sparse sampling conducted after a test dose, as well as the first dose on each of the 4 days, with busulfan dose adjustments made prior to the third dose each of the 4 days. Although difficult to ascertain the precise number of dose adjustments required, the authors commented that the test dose allows initial dosage individualization only, and that further monitoring and dose adjustments are subsequently needed to minimize intrapatient pharmacokinetic variability. Of importance, both Bolinger *et al.* and Bleyzae *et al.* reported improvement in major clinical outcomes from HD-BU therapeutic drug monitoring, including superior engraftment and reduction in early mortality.

In our study, neither the dose corrected AUC or apparent oral clearance for the test dose were bioequivalent to the AUC and the apparent oral clearance for busulfan at steady state (13th dose) using the strict criteria of the two one-sided *t*-tests (TOST). Of greater practical significance, only eight of 24 patients would have fallen within the range of 3600–5400 ng·h/ml, if the apparent oral clearance from the test dose was used to target the midpoint of the steady-state therapeutic range for partially matched or unrelated donors. Of potentially greater clinical importance, the AUC and the apparent oral clearance for the first and 13th doses were also not bioequivalent (TOST ± 20%, $p > 0.05$). In four cases, the AUC following the first dose could not be calculated due delayed absorption. Previous studies similarly report prolonged absorption prevented using the first dose AUC for therapeutic drug monitoring [11,25–27]. In this study, the first dose apparent oral clearance and AUC successfully predicted the dose needed to achieve the midpoint of the target steady-state AUC (4500 ng·h/ml) range in only 13 of 24 patients for an overall success rate of 13 of 28 (46%) when the first dose AUC could be accurately estimated. This is particularly problematic in that it is common practice in many institutions to base the busulfan dose on the AUC achieved following the first dose.

A number of investigators have remarked on the poor ability of the AUC determined after the first dose to predict the dose needed to achieve the target steady-state systemic exposure [6,11,12,26,25]. An 18–32% reduction in the AUC from the first dose to the 13th dose to the 16th dose has been reported by a number of investigators due to increased busulfan clearance during the 4-day HD-BU course [9,21,28]. This finding was attributed to the induction of busulfan metabolism by phenytoin and, possibly, a systematic overestimation of the first dose AUC. Variations in steady-state HD-BU concentrations above 500% (first versus 13th dose) have also been reported in children, most of whom did not receive phenytoin [12]. Two recent reports similarly comment that intrapatient variability in busulfan apparent oral clearance limits the ability to use a single busulfan AUC to successfully achieve the target AUC [25,26]. In fact, one center reported that 'frequent' dose adjustments are required on the fifth dose, while dose adjustments are 'less often' required after the ninth dose, when adjusting the dose to achieve the target

steady-state concentration [6]. Similarly, another center that assessed the AUC after the first, fifth and ninth doses of oral busulfan reported sequential pharmacokinetic monitoring was preferable to the use of a single test result to establish the busulfan dosage for myeloablative therapy due to intrapatient variance [26]. An obvious disadvantage associated with sequential monitoring is the need for repeated blood sampling, sophisticated on-site analytic equipment and pharmacokinetic expertise.

Clearly, predictive ability of both test and first dose would have been superior if the target range of 1200–5400 ng·h/ml was used as the test criteria. This lack of predictive ability of the test dose may have been, in part, a function of the smaller size of the test dose (0.25 mg/kg or 25% of standard dose in HD-BU regimens). However, the oral bioavailability for busulfan is approximately 80–90% [7], providing no reason to suspect dose-dependent absorption based on the available literature. Since the lower limit of detection of the assay used in this study was adequate for quantitation of a smaller test dose, it was decided to use the least amount of busulfan possible to obtain the pharmacokinetic parameters of interest.

The contribution of absorption to variability in busulfan disposition was not able to be determined in the past due to the lack of an i.v. busulfan formulation. Recently, it has been rather conclusively demonstrated that unpredictable and erratic intestinal absorption of busulfan contributes to wide interpatient and intrapatient variability in busulfan systemic exposure. Five recent articles in the *Journal of Biology of Blood and Marrow Transplantation* describe experience with i.v. busulfan (Busulfex injection; Orphan Medical, Minnetonka, MN) in patients receiving preparative regimens with HD-BU and two doses of cyclophosphamide or 5 days of fludarabine [29–33]. An 0.8 mg/kg i.v. dose of busulfan was found to produce AUCs similar to that obtained with 1 mg/kg orally. However, intrasubject variability between dose 1 and dose 7 or 9 ranged from 1 to 9% in pharmacokinetic sampling of i.v. busulfan [30,31]. Intersubject variability also appeared to be reduced compared to previous results with oral HD-BU. Pharmacokinetic parameters, including clearance, half-life, maximum concentration and AUC, demonstrated that the first dose profile of i.v. busulfan was highly predictive of later dose pharmacokinetic profiles [30,31]. In addition, in both of the trials, i.v. busulfan was well tolerated, and demonstrated excellent antitumor efficacy and reduced toxicity compared to historical controls which the authors ascribed to busulfan dose assurance with predictable pharmacokinetics.

Conclusion

The data from this study and other studies suggest that busulfan elimination is altered, at least in some patients, with repeated dosing. These alterations resulted in both increases and decreases in busulfan AUC at dose 13 compared to dose 1. A mechanism that could account for this is unclear, since the metabolic disposition of busulfan suggests that drug interactions with busulfan would only occur by depleting glutathione content or inducing GSTs. Neither of these mechanisms have been investigated to date. Busulfan's metabolism following repeated doses, and the influence of comorbid diseases and concomitantly administered medications needs to be further investigated.

Busulfan is an important component of preparative regimens for bone marrow and stem cell support protocols. Until very recently, busulfan has been available only in an oral dosage formulation. The desired average steady-state busulfan concentration and AUC to decrease the risk of rejection and veno-occlusive disease has been derived from a number of patient series. In many of these reports, the pharmacokinetics following the first oral dose of busulfan is used to guide dosage adjustment to achieve the target AUC. This report highlights the inadequacy of this approach, particularly for partially matched family and matched unrelated donors, where the desired steady-state concentration or AUC is within a fairly narrow range. Intrapatient variability in busulfan pharmacokinetics following an oral test dose and the traditional oral HD-BU regimen negated the value of both the test dose and first dose approach to target busulfan systemic exposure at steady state. The recently marketed intravenous formulation of busulfan has demonstrated far less intrapatient variability in two recent reports [30,33]. A study that investigates the ability of a test dose of i.v. busulfan to predict dose required to achieve target systemic exposure at steady state is in process at this institution. The role that genotypic and phenotypic differences in intestinal and hepatic glutathione and GST play in busulfan absorption and metabolism requires further investigation.

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