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Evaluation of a Test Dose Strategy for Pharmacokinetically-Guided Busulfan Dosing for Hematopoietic Stem Cell Transplantation

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Targeted busulfan dosing helps limit chemotherapy-related toxicity and optimize disease outcomes in hematopoietic stem cell transplantation (HCT). The objective of this study was to evaluate busulfan exposure from a pharmacokinetic (PK)-guided dosing strategy using a test dose. This retrospective evaluation included adult patients who underwent HCT at our institution with busulfan-based myeloablative (>9 mg/kg) conditioning between January 2014 and October 2015. A weight-based test dose of 0.8 mg/kg was used with PK assessments to predict area under the curve (AUC_{pred}) achieved with weight-based dosing, with a target AUC of 4800 μM*minute (AUC_{target}). PK from the test dose was then used to calculate a PK-guided first myeloablative busulfan dose. PK assessments were also done after the first dose to assess if the goal area under the curve (AUC) had been achieved (AUC_{first}). A PK-guided first dose resulted in achievement of target AUC with target ranges of ±10% in 50% of patients, ±15% in 75%, and ±20% in 94%. This was an improved rate of target achievement compared with the 33%, 44%, and 63% of patients who achieved the desired AUC for these respective target ranges when using weight-based dosing ($P = .12$, .004, and <.001, respectively). The PK-guided strategy also decreased the variability of AUC from 3.6-fold in AUC_{pred} from the weight-based test doses (2700.8 to 9631 μM*minute; SD, 1211.6 μM*minute) to 1.8-fold in AUC_{first} from the PK-guided first doses (3672.1 to 6609.8 μM*minute; SD, 574.7 μM*minute). This reflects a 2-fold improvement in AUC variability with a PK-guided dosing strategy. This is also improved from the 3-fold variability in AUC reported in other studies. Weight and body surface area were significantly associated with the likelihood of AUC_{first} being within the ±10% target range ($P = .04$ for both associations). There was no significant association between AUC_{first} and death, relapse, or a composite of the two. These results demonstrate a significant improvement in target AUC attainment and less interpatient variability with PK-guided dosing using a test dose strategy compared with weight-based dosing.

INTRODUCTION

Busulfan has been used as a component of conditioning regimens in hematopoietic stem cell transplantation (HCT) since the 1950s. Formulations and dosing regimens have evolved since then [1]. Limitations to the use of oral busulfan include interpatient and intrapatient variability in exposure, the need for frequent dosing, and increased first-pass metabolism, potentially increasing the rate of sinusoidal obstruction syndrome (SOS) and other toxicities [2–4]. Intravenous busulfan improves pharmacokinetic (PK) variability, can be administered once daily, and reduces the toxicity profile [5–10]. Despite the improvements with i.v. administration, therapeutic drug monitoring (TDM) with busulfan is still warranted to optimize outcomes, particularly in the myeloablative setting, because i.v. busulfan has been associated with a 3-fold variability in area under the curve (AUC) [11–14]. In addition, subtherapeutic exposure has been associated with increased risk of

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relapse and lower disease-free and overall survival, whereas supratherapeutic concentrations have been linked to increased toxicity and decreased overall survival [15–18]. Thus, targeted busulfan exposure can help reduce toxicity and improve survival [12,19].

The use of busulfan test doses before conditioning chemotherapy has been evaluated by our group and others as a strategy for PK-guided dosing to achieve target AUC and optimize busulfan dosing [20–22]. In the present retrospective analysis, we sought to demonstrate the effectiveness of a test-dose PK strategy to predict daily exposure and guide accurate dosing of full myeloablative i.v. busulfan doses. This study will help characterize the utility of a test-dose PK-guided dosing strategy with confirmatory PK in clinical practice.

PATIENTS AND METHODS

Study Design and Population

All adult patients who received myeloablative conditioning containing once-daily i.v. busulfan (>9 mg/kg total dose over 4 days) followed by HCT at our institution between January 2014 and October 2015 were included [11,23]. Patients who were age <18 years, had received a pediatric protocol, had received reduced-intensity conditioning if PK analysis was not performed, or had missing or incorrectly documented data were excluded. For patients for whom first-dose PK data were missing, data were included in the analysis of toxicity and survival but excluded from PK analysis comparing test and first-dose parameters.

Methods of PK Analysis

All patients with full myeloablative busulfan-based conditioning received a test dose of 0.8 mg/kg in the outpatient HCT clinic a minimum of 2 days (average, 8 days; range, 3 to 29 days) before admission for conditioning chemotherapy. Total body weight was used for dose calculations unless the patient was at ≥120% of ideal body weight, in which case a 25% adjusted ideal body weight was used. All doses were infused at a rate of 80 mg/hour [20]. Busulfan serum concentrations were drawn at the end of the infusion and 30 minutes and 1, 2, 4, and 6 hours after the end of infusion and sent to the special chemistries referral laboratory at Emory University Hospital for analysis. Phoenix WinNonlin noncompartmental analysis (Certara, Princeton, NJ) was used to calculate the AUC and clearance from the test dose (CL_{TD}). Given the linear nature of busulfan PK and considering that the test dose is 25% of a 3.2 mg/kg myeloablative busulfan dose, the test dose AUC is proportional to, and thus predictive of, the AUC expected from a weight-based 3.2 mg/kg dose (predicted AUC [AUC_{pred}]). Thus, AUC_{pred} was considered the AUC achieved with weight-based dosing. PK analysis of the test dose was used to calculate the myeloablative busulfan dose required to achieve a target AUC (AUC_{target}) of 4800 μM*minute using the following equation: Dose = $AUC_{target} \times CL_{TD}$.

Busulfan concentrations were similarly drawn after the first myeloablative busulfan dose, and PK analysis was performed to compute the first-dose AUC (AUC_{first}) and CL (CL_{first}) to assess achievement of AUC_{target}. If the AUC_{first} was outside of ±10% of the AUC_{target}, then the remaining busulfan doses were adjusted using the following equation: Dose = $AUC_{target} \times CL_{first}$. Results were obtained the day after collection, permitting adjustments as necessary for doses 3 and 4.

Endpoints and Definitions

We determined the percentage of patents who achieved AUC_{target} ranges of 4800 μM*minute ±10%, ±15%, and ±20 for both the AUC_{pred} and the AUC_{first}. Endpoints assessed include the difference between AUC_{pred} and AUC_{target}, between AUC_{first} and AUC_{target}, and between AUC_{pred} and AUC_{first}. Also analyzed in this evaluation was a comparison of PK-guided myeloablative busulfan doses with other dosing strategies (ie, 3.2 mg/kg and 130 mg/m^2), the number of patients requiring PK-guided dose adjustments, toxicity assessments (ie, liver function test [LFT] elevations and incidence of SOS), and relapse and survival outcomes. LFT results were evaluated up to post-transplantation day 60 using Common Terminology Criteria for Adverse Events version 4.03 [24]. SOS was evaluated up to post-transplantation day 60 based on documentation of diagnosis or treatment for SOS in the electronic medical record.

Statistical Analysis

Continuous and binary variables were compared using the t test and χ^2 test, respectively. If normal assumptions were violated or the sample size was small, the Wilcoxon rank-sum or Fisher exact test was used instead for continuous or binary variables, respectively. Patients were divided into 3 groups according to the AUC (below, within, and above AUC_{target}). To evaluate which factors affected achievement of the target AUC, the F test was used for continuous outcomes and the Cochran-Mantel-Haenszel test was used for binary outcomes.

RESULTS

Patients

Fifty-seven patients were identified as receiving a myeloablative busulfan-containing regimen during the specified period. Five patients were excluded owing to age (n = 2), use of a pediatric protocol (n = 1), change in treatment plan to a reduced-intensity regimen (n = 1), or inaccurate PK documentation (n = 1). A total of 52 patients were included in the final analysis, 4 of whom had test dose PK sampling performed but did not undergo first dose PK analysis. The patients' baseline characteristics are summarized in Table 1.

AUC Analysis

The average AUC_{pred} from weight-based dosing was 5202.7 μM*minute (range, 2700.8 to 9631 μM*minute; SD, 1211.6 μM*minute). The AUC_{pred} was within ±10%, ±15%, and ±20% of the AUC_{target} in 17 (33%), 23 (44%), and 33 (63%) patients, respectively (Table 2). The mean percent difference between AUC_{pred} and AUC_{target} was 8% (range, -44% to 101%). This mean percent difference was calculated using both negative and positive values, with negative values representing an AUC_{pred} lower than the AUC_{target} in an effort to show directionality in the overall mean (ie, if on average, AUC_{pred} was higher or lower than AUC_{target}). The mean percent difference when not considering directionality in relation to the target was 20% (range, 0 to 101%). The average AUC_{first} from PK-guided dosing was 4585.8 μM*minute (range, 3672.1 to 6609.8 μM*minute; SD, 574.7 μM*minute), which was significantly lower than the average AUC_{pred} ($P < .01$). The AUC_{first} was within ±10%, ±15%, and ±20% of the AUC_{target} for 24 (50%), 36 (75%), and 45 (94%) patients, respectively. There was a nonsignificant improvement in the rate of AUC target attainment after receiving a PK-guided myeloablative dose compared with a weight-based dose with the ±10% range (50% vs 33%; $P = .12$) and significant improvements with the ±15% (75% versus 44%; $P = .004$) and ±20% (94% versus 63%; $P < .001$) ranges (Table 2). The directional mean percent difference between AUC_{first} and AUC_{target} was -4% (range, -23 to 38%). The mean percent difference without directionality was 10% (range, 0 to 38%). The AUC_{first} achieved from PK-guided dosing had an improved directional and nondirectional mean percent difference from AUC_{target}, as

Table 1
Patient Characteristics (n =52)

Covariate	Value
Age, yr, mean ± SD	47 ± 11
Female sex, n (%)	23 (44)
Weight, kg, mean ± SD	72 ± 12
Height, cm, mean ± SD	171 ± 10
BSA, m^2, mean ± SD	1.84 ± 0.21
BMI, kg/m^2, mean ± SD	30 ± 7
Obesity (BMI ≥30 kg/m^2), n (%)	25 (48)
Diagnosis, n (%)	
Acute myelogenous leukemia	31 (60)
Myelodysplastic syndrome	6 (12)
Chronic myelogenous leukemia	2 (4)
Non-Hodgkin lymphoma	5 (10)
Acute lymphoblastic leukemia	4 (8)
Other	4 (8)
Transplant type/donor status, n (%)	
Matched related donor	12 (23)
Matched unrelated donor	29 (56)
Mismatched unrelated donor	9 (17)
Other	2 (4)

Table 2
PK Analysis of AUC_{pred} and AUC_{first}

	AUC_{pred} (n = 52) (Weight-Based Dosing)			AUC_{first} (n = 48) (PK-Guided Dosing)			*P* Value
Mean ± SD AUC, μM*min (range)	5202.7 ± 1211.6 (2700.8-9631)			4585.8 ± 574.7 (3672.1-6609.8)			< .01
	Below	Within	Above	Below	Within	Above	For comparison of % within range
Target AUC ±10%, n (%)	12 (23)	17 (33)	23 (44)	18 (38)	24 (50)	6 (13)	.12
Target AUC ±15%, n (%)	8 (15)	23 (44)	21 (40)	8 (17)	36 (75)	4 (8)	.004
Target AUC ±20%, n (%)	5 (10)	33 (63)	14 (27)	2 (4)	45 (94)	1 (2)	<.001

Table 2. Pharmacokinetic analysis of AUC_{pred} and AUC_{first} in all patients. Mean AUC_{pred} and AUC_{first} was 5202.7 μM*min and 4585.8 μM*min, respectively.

well as improved variability compared with AUC_{pred} from weight-based dosing.

Evaluating the relationship between AUC_{pred} and AUC_{first} revealed that 15%, 31%, and 65% of patients had both an AUC_{pred} and AUC_{first} within ±10%, ±15%, and ±20% of the AUC_{target}, respectively (Table 3). For the 48 patients with available first-dose PK data, 53%, 80%, and 88% of patients with an AUC_{pred} outside of range from weight-based dosing achieved an AUC_{first} within range from PK-guided dosing for the ±10%, ±15%, and ±20% ranges, respectively. AUC analysis for the percentage of patients with an AUC_{pred} and AUC_{first} below, within, or above the ±10%, ±15%, and ±20% ranges for the overall population are displayed in Table 3. When the AUC_{pred} was outside of the ±10% range, it was most commonly supratherapeutic (66%). When the AUC_{first} was outside of this range, it was most commonly subtherapeutic (75%), resulting in subsequent dose increases (Table 2). Figure 1 displays the paired AUC_{pred} and AUC_{first} data for each patient.

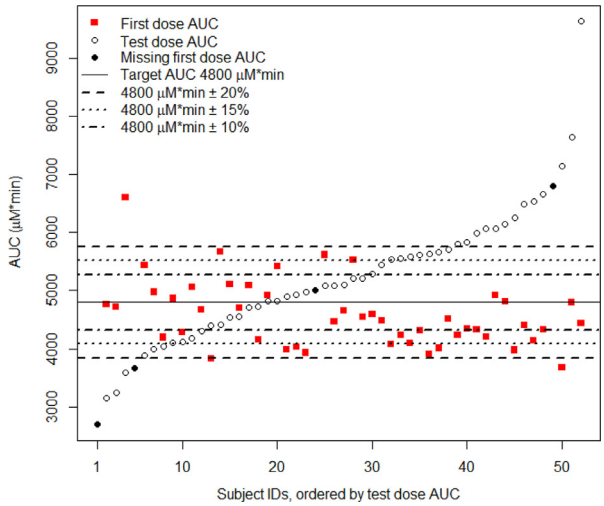

Figure 1. Comparison of AUC_{pred} and AUC_{first} with target AUC (AUC targets of 4800 μM*minute ± 10%, ± 15%, and ± 20%). The mean AUC_{pred} value was 5202.7 μM*minute, and the mean AUC_{first} value was 4585.8 μM*minute ($P < .01$).

Comparison of Dosing Strategies

A comparison of PK-guided busulfan doses with other dosing strategies was also performed. The average doses based on 3.2 mg/kg, PK-guided dosing, and 130 mg/m^2 were 229 mg, 218 mg, and 239 mg, respectively (Figure 2). The 3.2 mg/kg and 130 mg/kg doses were on average 11 mg (5%) and 21 mg (10%) higher than the PK-guided first doses ($P = .051$ and <.01, respectively). For patients with an AUC_{pred} below, within, and above the ±10% target range, 3.2 mg/kg dosing would have resulted in doses that were on average 52.2 mg (19%) lower, 2.6 mg (2%) higher, and 49.8 mg (28%) higher than PK-guided doses, respectively.

Twenty-three patients (48%) needed a dose adjustment after the first myeloablative busulfan dose because AUC_{first} was outside of the ±10% target range. Two patients with an AUC_{first} within ±15% of the target but outside ±10% of the target did not have a dose adjustment because ±15% was the cutoff for dose adjustments at that time. The mean adjusted dose was 257.6 mg, 39 mg (18%) higher than the mean first dose ($P = .01$) (Figure 2). Among the patients needing a dose adjustment after first-dose PK analysis, 74% had a dose increase and 26% had a dose decrease. For those patients with an AUC_{first} below the target, the mean dose increase was 42.4 mg (19%). The mean dose decrease was 37.5 mg (17%) for patients with an AUC_{first} higher than the target.

Factors Associated with AUC Targeting

To evaluate which factors affected the achievement of the target AUC using the ±10% range, we compared the mean values of the factors among the 3 AUC-based groups (below, within, and above) (Table 4). Weight and body surface area (BSA) were the only factors significantly associated with AUC_{first}. Patients with lower weight and BSA were significantly more likely to have an AUC_{first} exceeding the target compared with patients with higher weight and BSA ($P = .04$ for both variables). No significant association was found with body mass index (BMI) or obesity (as defined by World Health Organization criteria of BMI ≥30) [25]. Higher

Table 3
Relationship of AUC_{pred} and AUC_{first} (AUC Targets of 4800 μM*minute ± 10%, ±15%, and ±20%)

		AUC_{first}, % (n = 48)								
		Below			Within			Above		
	Target Range, %	±10	±15	±20	±10	±15	±20	±10	±15	±20
AUC_{pred}, % (n = 48)	Below	4	0	0	13	10	4	4	2	2
	Within	10	8	2	15	31	65	8	6	0
	Above	23	8	2	23	33	25	0	0	0

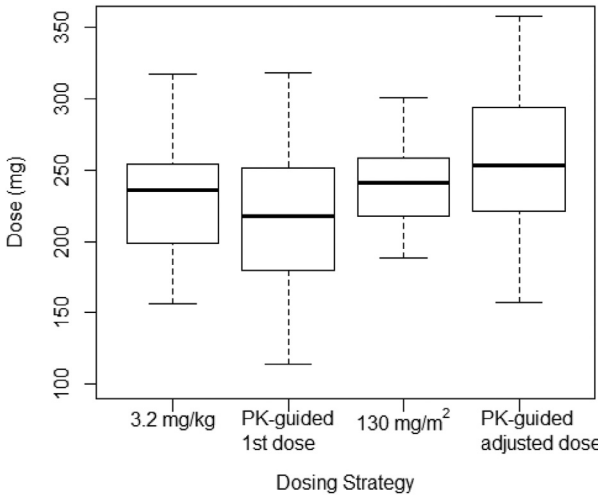

Figure 2. Mean myeloablative busulfan doses calculated by the following dosing strategies: 3.2 mg/kg, PK-guided dose, 130 mg/m^2, and the adjusted dose based on first dose confirmatory PK analysis.

AUC_{pred} values were associated with lower AUC_{first} values, but this association was not significant ($P = .17$).

Clinically significant drug interactions were documented in 15 patients (29%) during the test dose and in 17 patients (33%) during the myeloablative dose. Drugs included voriconazole, posaconazole, fluconazole, and acetaminophen. In 71% of these interactions, the interaction was only identified for either the test dose or the first dose, but not for both. Of these, only 1 involved posaconazole, whereas the others involved either fluconazole or acetaminophen. The majority of interactions with voriconazole and posaconazole were identified for both the test dose and the first dose. The variability of drug interaction documentation made it difficult to evaluate the impact of drug interactions on PK data; thus, drug interactions were not evaluated in the multivariate analysis.

Toxicity and Survival

The incidences of transaminitis, SOS, relapse, and death were assessed (Table 5). LFT elevations were assessed up to post-transplantation day 60. Forty-eight patients (92%) experienced LFT elevations of grade 1 or higher, including 36 (69%) with grade 1-2 elevations and 12 (23%) with grade 3-4 elevations. There was no statistically significant association between AUC_{first} and changes in LFT results ($P = .96$) (Table 4).

The development of SOS was also assessed up to post-transplantation day 60. Three patients (6%) developed SOS, including 1 patient from each of the AUC_{first} groups using the ±10% range (below, within, and above target range; $P = .50$) (Table 4). Two patients developed SOS within 60 days post-transplantation (1 diagnosed at day 40 and the other at day 41). The third patient had a total bilirubin rise that started on day 57 and was ultimately given a clinical diagnosis of SOS on day 63.

Table 5
Safety, Relapse, and Mortality in All Patients (n = 52)

Toxicity	n (%)
LFTs*	
Grade 0	4 (8)
Grade 1	25 (48)
Grade 2	11 (21)
Grade 3	10 (19)
Grade 4	2 (4)
SOS*	3 (6)
Death†	19 (37)
Relapse†	11 (21)
Death or relapse†	22 (42)

* Evaluated up to post-transplantation day 60.
† Median follow-up of 475 days (range, 100 to 744 days).

The median follow-up for assessment of relapse and death was 475 days post-transplantation (range, 100 to 744 days). Eleven patients (21%) experienced relapsed disease, and 19 patients (37%) died. The rate of death from relapse was 12% (n = 6), and that of nonrelapse mortality was 25% (n = 13). Causes of nonrelapse mortality included organ failure (n = 5), graft-versus-host disease (n = 2), infection (n = 5), and hemorrhage (n = 1). The average times to relapse and death post-transplantation were 131 and 128 days, respectively. There were no significant associations between AUC_{first} and death ($P = .50$), relapse ($P = .70$), or a combination of death and relapse ($P = .91$).

DISCUSSION

This retrospective study demonstrated improvement in AUC target attainment for myeloablative once-daily i.v. busulfan with the use of a PK-guided dose generated from a test dose compared with a weight-based dosing strategy. Improvement in target attainment with PK-guided doses was found for target ranges of ±10%, ±15%, and ±20%, with those for the ±15% and ±20% ranges being statistically significant. Here we used the AUC achieved from a test dose of 0.8 mg/kg to predict the AUC achieved from a weight-based dosing strategy of 3.2 mg/kg. Such an estimate was derived based on the understanding that busulfan PK is linear and that exposure from a 0.8 mg/kg test dose should be directly proportional to the AUC achieved from a 3.2 mg/kg myeloablative dose [10].

Table 4
Factors Associated with AUC Target* Attainment after the First Dose in Patients with AUC_{first} Data Available (n = 48)

Covariate	AUC_{first} below Target Range (n = 18)	AUC_{first} within Target Range (n = 24)	AUC_{first} above Target Range (n = 6)	*P* value†
Age, yr, mean ± SD	46 ± 11	47 ± 12	52 ± 7	0.33
Female sex, n (%)	5 (28)	13 (54)	3 (50)	0.16
Weight, kg, mean ± SD	78 ± 11	69 ± 11	71 ± 10	0.04
Height, cm, mean ± SD	176 ± 9	168 ± 10	172 ± 6	0.07
BSA, mean ± SD	1.96 ± 0.19	1.79 ± 0.20	1.83 ± 0.16	0.04
BMI, , mean ± SD	32 ± 6	30 ± 7	28 ± 10	0.28
Obesity (BMI ≥ 30), n (%)	11 (61)	11 (46)	2 (33)	0.20
AUC_{pred}, mean ± SD	5415 ± 823	5330 ± 1411	4497 ± 659	0.17
SOS, n (%)	1 (6)	1 (4)	1 (17)	0.50
LFT, mean ± SD	1.61 ± 1.09	1.58 ± 0.88	1.67 ± 1.51	0.96
Death, n (%)	7 (39)	7 (29)	4 (67)	0.50
Relapse, n (%)	5 (28)	3 (13)	3 (50)	0.70
Death or relapse, n (%)	9 (50)	8 (33)	4 (67)	0.91

* An AUC target of 4800 μM*minute ± 10% was used for these analyses.
† ANOVA was used for continuous outcomes, and the Cochran-Mantel-Haenszel test was used for binary variables.

We found that a weight-based dose would achieve an AUC_{target} of 4800 μM*minute ± 10%, ±15%, and ±20% in 33%, 44%, and 63% of patients, respectively. These percentages improved to 50%, 75%, and 94% of patients with the use of a PK-guided dose ($P = .12$, .004, <.001, respectively). In addition, 53%, 80%, and 88% of patients with an AUC_{pred} outside of the ±10%, ±15%, and ±20% ranges, respectively, from weight-based dosing were able to achieve an AUC_{first} within range with a PK-guided dose. These data support the continued use of a PK-guided dosing strategy using a test dose over weight-based dosing in our population.

Our results also support the use of confirmatory first-dose PK analysis. Of the 16 patients with an AUC_{pred} within the ±10% range who had first-dose PK data available, 56% had an AUC_{first} outside of the ±10% target range, necessitating a dose adjustment. If one were willing to accept a target range of ±20%, then one could potentially challenge the utility of confirmatory first-dose PK following a PK-guided initial dose, given that 94% of patients achieved a target of 4800 μM*minute ± 20%. This would reduce the cost and logistical challenges associated with doing both test dose and first-dose PK analyses at a center that is dependent on an outside referral laboratory to process busulfan samples.

A recent guideline published by the American Society of Blood and Marrow Transplantation states that the initial i.v. busulfan dose should be a weight-based dose of 0.8 mg/kg i.v. every 6 hours or 3.2 mg/kg i.v. daily and suggests that test dose strategies cannot be routinely recommended, because they do not predict clearance well enough to replace busulfan TDM. Our data suggest that the use of a test dose strategy along with, rather than in place of, confirmatory busulfan PK monitoring merits further review until data for test doses alone are more robust [11]. The use of a test dose to predict busulfan conditioning doses has been evaluated previously [21,26,27]. Beri et al [21] evaluated a PK-guided dosing strategy using a 0.8 mg/kg i.v. test dose in 23 adults receiving myeloablative conditioning with once-daily i.v. busulfan and fludarabine. Based on the predicted AUC from a weight-based test dose, they found that 65% of patients had an AUC within the target range of 4000 to 6000 μM*minute after weight-based dosing. This range translates to a target of 5000 μM*minute ± 20%. In our evaluation, 33% and 46% of patients achieved an AUC_{pred} within the target ranges of 4800 μM*minute ±10% and ±15%, respectively, after a weight-based dose. However, similar to the findings of Beri et al, 63% of patients were within range of a ±20% window. In the study of Beri et al, after receiving a PK-guided first dose, 88% of patients had an AUC_{first} within the target range. In our analysis, 94% of patients were within the 4800 μM*minute ± 20% target range after receiving a PK-guided first dose. Both evaluations demonstrate that a test dose strategy to calculate a PK-guided first dose increases the percentage of patients able to achieve these AUC target ranges with their first dose compared with patients who receive a weight-based initial dose.

Weil et al [27] also evaluated the efficacy of a busulfan test dose in adult patients undergoing myeloablative conditioning. In their observational study, they compared the percentage of patients who were able to achieve the desired busulfan steady-state goal between 30 patients who received a busulfan test dose and 30 patients who did not. There were a few notable differences between their evaluation and our present study. First, Weil et al used a test dose of 0.9 mg/kg in their evaluation, compared with our 0.8 mg/kg test dose. Second, the busulfan used in their myeloablative regimens was administered every 6 hours, whereas we used once-daily busulfan regimens. Finally, the PK parameter they used for busulfan TDM was a concentration at steady state (C_{ss}) target of 900 ng/mL ±10%, whereas we used AUC. Nonetheless, consistent with our findings, Weil et al reported that implementation of a test dose strategy significantly increased the percentage of patients who were able to achieve a therapeutic C_{ss} with the first myeloablative busulfan dose ($P < .001$) [27]. Their data and ours support the use of a test dose strategy even among heterogeneity in practice with respect to the dose used for the test dose, the busulfan dosing schedule used in myeloablative regimens, and the PK parameter used for busulfan TDM.

Other busulfan dosing strategies have been evaluated in the literature. For example, Madden et al [10] conducted a PK analysis of busulfan 130 mg/m^2 i.v. daily. Although that study did not evaluate patients attaining a specific target AUC range, 22 of 60 patients (37%) had an AUC within the range of 4500 to 5499 μM*minute. Perkins et al [13] also evaluated a protocol that used 130 mg/m^2 as the initial busulfan dose, followed by PK analysis after the first myeloablative busulfan dose in a once-daily dosing schedule. Dose adjustments were made to target an AUC of 5300 μM*minute ±10%, and 37% of patients were within that target AUC range. These results are similar to the approximate one-third of patients falling within the ±10% range in our study when weight-based dosing strategies were used. We showed that this rate of target attainment can be improved to between 50% and 94% depending on goal range (±10% to ±20%) with PK-guided dosing from a test dose.

Of note, the variability in AUC_{first} in our evaluation was numerically smaller than both our AUC_{pred} and the AUC ranges reported in the literature. The AUC_{pred} from weight-based dosing ranged from 2700.8 to 9631 μM*minute in our study (a 3.6-fold difference). Similarly, the AUC ranged from 2900 to 8200 μM*min (2.8-fold difference) and 2632 to 8219 μM*min (3.1-fold difference) in evaluations using weight-based doses by Madden et al [10] and Perkins et al, respectively [13]. Our AUC_{first} after PK-guided dosing had a much narrower range, from 3672.1 to 6609.8 μM*minute (a 1.8-fold difference), indicating that PK-guided initial dosing resulted in fewer patients with extremely high or low AUCs. This is critical, considering that reducing such a large degree of variability can limit the potential for toxicity or relapse associated with these extremes [16,28].

Although the mean AUC_{pred} and AUC_{first} were both within the target range of 4800 μM*minute ±10%, when AUC_{pred} was outside of this range, it was more commonly higher than the target (66%). Conversely, among the patients with an AUC_{first} outside of the target range, a greater proportion was below the target range (75%). Among the 22 patients with an AUC_{pred} above the target range and with first-dose PK data, 11 (50%) had a subsequent AUC_{first} below the target (Figure 1, Table 3). This suggests a tendency for the dosing equation to overcorrect the first dose, resulting in a subtherapeutic AUC_{first}. Considering the calculation used to compute the first dose ($Dose_{first} = AUC_{target} \times CL_{test}$), one could conclude that the PK analysis of the test dose may have potentially underestimated patient clearance; however, there was no significant difference between the average CL_{test} and CL_{first} (11.3 L/hour versus 11.9 L/hour; $P = .22$). It is notable that Beri et al [21] used a different equation to calculate the first dose [$Dose_{first} = (AUC_{target} \times Dose_{test})/AUC_{pred}$] and had no patients with a subtherapeutic AUC_{first}, albeit with a wider therapeutic index (4000 to 6000

μM*minute). Thus, one must consider the potential need for optimization of the equation used to compute PK-guided doses. However, the equation used in our practice is a fundamental PK calculation that should result in accurate prediction of these PK parameters, assuming a linear clearance across the range of doses studied [11]. Weight and BSA, but not BMI or obesity, were significantly associated with an AUC_{first} within the ±10% target range (P = .04). On average, weight- and BSA-based dosing strategies, even when using adjusted weights, resulted in higher myeloablative busulfan doses compared with the PK-guided dosing. The higher mean dose and AUC_{pred} from weight-based dosing introduce concerns for increased busulfan exposure, leading to potential toxicity with this dosing strategy.

Our evaluation did not identify any associations between AUC and toxicity, death, or relapse. This may be because we found less interpatient variability. With <2-fold interpatient variability, the high and low AUCs potentially associated with toxicity, death, and/or relapse were minimal. Furthermore, it is our practice to adjust myeloablative busulfan doses if the first-dose PK is not within 10% of the target range. Consequently, although we report the percentages of patients who were also within 15% and 20% of the target range based on their first-dose PK, subsequent dosing for busulfan doses 3 and 4 were adjusted to be within 10% of the range. Thus, we are unable to draw conclusions regarding the possible impacts on toxicity, relapse, and death of being within 10% versus 15% versus 20% of the target.

Our analysis was not without limitations. The inherent nature of a retrospective review makes it difficult to account for other variables that might have influenced our results. We evaluated an institution-specific busulfan PK dosing strategy involving a fixed infusion rate of 80 mg/hour, 6 postinfusion busulfan concentrations, and a target AUC of 4800 μM*minute [20]. Alternative strategies may differ in terms of infusion rate, number of levels drawn at different times postinfusion, PK parameter and target range used, dosing equation, or assays used to quantify plasma concentrations, limiting the applicability of these results to other centers [21,29–31]. The achievement of AUC_{target} for weight-based dosing strategies in this report versus our PK-guided strategy is dependent on the assumption that AUC_{pred} after the test dose is reflective of the AUC that would be achieved from a 3.2 mg/kg weight-based dose. Although this is a reasonable prediction given the linear PK and dose proportionality of busulfan, one must consider that none of our patients actually received a weight-based dose as their first dose with subsequent AUC analysis, because our practice is to administer a PK-guided first dose to all patients. In addition, we identified drug interactions that existed for the test dose, the first dose, or both. Medication documentation made it challenging to confirm that these medications were actually active during the time of busulfan administration, and thus we were unable to determine the effect of drug interactions on busulfan PK. Nonetheless, it is important to consider medication profiles to ensure that interacting medications are not initiated, discontinued, or modified during test doses or the conditioning regimen, to limit their impact on PK assessments across doses [11]. Finally, follow-up PK analysis was not completed after dose adjustments to myeloablative busulfan doses based on AUC_{first}, and thus we were unable to confirm whether dose adjustments based on AUC_{first} values were actually successful in attaining the target AUC for the entire 4-day course, or whether the AUC and CL_{ss} values over the 4 days of administration remained constant regardless of dose adjustments.

CONCLUSION

Our present evaluation suggests a significant improvement in target busulfan AUC attainment of both a 4800 μM*minute ±15% and ±20% AUC_{target} range and a nonsignificant but clinically meaningful effect at the ±10% range when comparing the AUC and CL_{ss} values obtained after PK-guided myeloablative busulfan dosing and weight-based dosing. This strategy also led to attainment of the AUC_{target} after the first myeloablative dose in the majority of patients even with an AUC_{pred} outside the target range after the test dose. Notably, there was less interpatient variability in AUC achieved from the PK-guided dose than was estimated to occur from a weight-based strategy reported in the literature. Confirmatory first-dose PK is necessary to adjust subsequent myeloablative busulfan doses with the aim of further improving busulfan exposure. In this retrospective review, we did not identify any associations between busulfan AUC and toxicity, death, or relapse, although this evaluation was limited by a small patient population with variations in demographic and disease-related characteristics. Further studies are needed to determine the effects of a test dose strategy with confirmatory TDM for PK-guided dosing on relapse, toxicity, or survival, and the approach described here outlines a potential methodology and provides justification for doing so.

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