Bone Marrow Transplantation (2000) 26, 1143–1147

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Retrospective appraisal of busulfan dose adjustment in children

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Summary:

This report describes and critically appraises our experience with busulfan dose adjustment in children undergoing bone marrow transplant between April 1997 and March 1999. All children received an initial busulfan dose of 40 mg/m^2 p.o. or by nasogastric tube. Whole blood samples were obtained 1, 1.5 and 6 h later and analyzed for busulfan content by gas chromatography with electron capture detection. The area under the whole blood busulfan concentration *vs* time curve (AUC) and an individualized dose which would achieve an AUC of 1300 μM/min were calculated. Mean and median busulfan doses were calculated using actual, ideal and effective body weight and stratified according to age. The relationship between the busulfan concentration at hour 6 and AUC was determined using linear regression. Thirty-nine courses of busulfan were evaluated in 38 patients. A change from the initial busulfan dose was required to achieve the target AUC in 34 courses (87%). Most children >1 to 5 years old required dose increments while most children >5 years old required dose reductions. Obesity did not significantly affect busulfan dose requirements. Busulfan concentrations at 6 h only weakly predicted the AUC achieved (r^2 = 0.496; P = 0.001). Based on these findings, we recommend that the initial busulfan dose be assigned according to patient age and actual body weight. We also recommend that busulfan AUC be calculated for children using a four-sample (1, 1.5, 4 and 6 h) limited sampling technique. *Bone Marrow Transplantation* (2000) **26,** 1143–1147.

Keywords: busulfan; pharmacokinetics; pediatrics; bone marrow transplant

Individualization of busulfan doses has been recommended in order to obtain sufficient dose intensity to achieve engraftment while avoiding relapse, and toxicity.[1–5] Busulfan dose adjustment to achieve a target area under the whole blood busulfan concentration *vs* time curve (AUC) was adopted by the Bone Marrow Transplant (BMT) Program at The Hospital for Sick Children (HSC) in September 1996. The dose adjustment methodology was based on previous work done at HSC.[6]

This report describes our experience with the previously described dose adjustment methodology and was undertaken to refine our technique.

Methods

Patients who underwent BMT at HSC between April 1997 and March 1999 and who received busulfan as part of their conditioning regimens were reviewed. The following data were obtained from each patient's health record and/or pharmacy profile: diagnosis, BMT date, age at transplant, height, weight, busulfan doses, busulfan serum concentrations and calculated busulfan AUC.

An initial busulfan dose of 40 mg/m^2 was given p.o. or through a nasogastric tube at 0500 hours. Patients received nothing by mouth 1 h before and 2 h after each busulfan dose. Whole blood samples were obtained at 1, 1.5, and 6 h following the dose. Busulfan concentrations were measured by gas chromatography with electron capture detection.[6]

The area under the busulfan concentration *vs* time curve (AUC) was calculated using the following equation: AUC = $45C_{1h} + 15C_{1.5h} + 270C_{1.5h}/(\ln C_{1.5h} - \ln C_{6h})$ where C_{1h}, $C_{1.5h}$ and C_{6h} represent the busulfan concentrations at 1, 1.5 and 6 h after the dose was given, respectively.[6] A dose was calculated for each patient based on a linear relationship between busulfan dose and AUC and a target AUC of 1300 μM/min using the following equation: adjusted dose = test dose × (target AUC/test dose AUC).[6] The intended acceptable AUC range was 900 to 1400 μM/min. The calculated busulfan dose was administered 24 to 48 h after administration of the initial dose and repeated every 6 h for a further 15 doses.

Busulfan was administered p.o. or n.g. as a combination of 2 mg (Glaxo Wellcome, Mississauga, Canada) or 25 mg (Glaxo, Wellcome, Greenford, UK) tablets. Use of the 25 mg tablets was stopped in February 1998 due to the lack of information regarding the relative bioavailability of the 2 and 25 mg tablets. Doses were rounded to the nearest whole tablet size; tablets were not halved or quartered. Patients received either whole tablets, whole tablets enclosed in a gelatin capsule, or tablets disintegrated in water.[7]

Serum busulfan concentrations were repeated if the initial set was uninterpretable (eg increasing busulfan concentrations throughout the 6 h sampling time) or if there was a large difference between the adjusted dose and the

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Received 20 January 2000; accepted 19 July 2000

test dose, as judged by the staff physician and/or pharmacist. At the discretion of the staff physician or pharmacist in the second and subsequent AUC determinations, more than three whole blood concentrations may have been obtained. If more than three busulfan concentrations were available, AUC was calculated using the trapezoidal rule.[8]

All patients received a loading dose of phenytoin (6.8 mg/kg/dose every 3 h × 3 p.o./i.v.) 24 h before receiving the busulfan test dose. Maintenance phenytoin dosing was started 8 h after the last of the loading doses was given and ended 24 h after the last busulfan dose.

Body surface area (BSA),[9] ideal body weight (IBW)[10] and effective body weight[10] (EBW) were calculated using standard equations.

Mean and median initial and final busulfan doses were calculated for the entire patient group and stratified according to age. The proportion of patients who required final busulfan doses higher or lower than the test dose was determined. The relationship between the busulfan concentration at hour 6 and AUC was described using linear regression. Dosage recommendations for the initial busulfan dose were made based on the median final busulfan dose requirement for each age group.

Results

Forty-one patients received busulfan conditioning for BMT with doses adjusted based on AUC during the evaluation period. Three patients were excluded since their health records were incomplete. One patient received busulfan conditioning on two separate occasions. Thirty-nine courses of busulfan were therefore evaluated in 38 patients. In all, 212 busulfan concentrations were obtained.

Patient demographic data are presented in Table 1. The mean AUCs achieved after administration of the initial 40 mg/m^2 busulfan dose are presented in Table 2. No dose change was required in five courses (13%), a decrease in the test dose was required in 23 courses (59%) and an increase in the test dose was required in 11 courses (28%) (Table 2). Most (82%) dose increases were required by children $\leqslant$5 years of age while most dose decreases (78%) were required by children $>$5 years of age (Table 3). The mean and median final doses required stratified by age are presented in Tables 3 and 4, respectively.

Busulfan concentrations were repeated in seven courses (18%) due to uninterpretable results after the initial dose. It was impossible to calculate an elimination rate constant after administration of the initial dose in six courses since the busulfan concentration increased throughout the sampling period. The concentration at 1 h after administration of the initial dose was extremely high in another course. The busulfan test dose was repeated in all these courses and busulfan AUC subsequently could be calculated. However, more than three busulfan concentrations were obtained after the second dose in four courses.

Table 1 Demographic data

Median age (years)	5.7
Age range (years)	0.17–17.5
Gender (M:F)	20:18
Diagnosis (No.)	
Acute myelogenous leukemia	22
Metabolic storage diseases	4
Chronic myelogenous leukemia	3
Immune deficiency syndromes	3
Histiocytosis	2
Soft tissue sarcoma	2
Congenital neutropenia	1
Hodgkin's lymphoma	1

Busulfan concentrations were repeated in 11 courses (28%) to verify that the target AUC was achieved after a mean dose adjustment of 51 ± 26% (range: 0–100%). Ten of these courses consisted of two AUC determinations while one consisted of three AUC determinations. The final busulfan AUC after dose adjustment was higher than expected (mean: 77 ± 66%; range: 4–59%) in five courses and lower than expected (mean: 26 ± 20%; range: 17–150%) in six courses. The mean final dose adjustment was 18 ± 12% (range: 0–40%) of the initial adjusted dose.

The relationship between the busulfan concentration at 6 h and AUC was evaluated in 51 6-h concentration–AUC pairs. Busulfan concentration at 6 h weakly predicted the AUC achieved ($r^2 = 0.496$; $P = 0.001$).

Discussion

Our initial three-sample method of AUC calculation was based on observations in nine children who received a slurry of crushed 2 mg busulfan tablets in water by nasogastric tube.[6] Half of these children were less than 5 years old; none of these children were older than 12 years of age. In these patients, AUC calculation using four samples (1, 1.5, 4 and 6 h) yielded similar results and variability to those calculated using three samples.

It was impossible, in practice, to standardize the route of administration for all patients. Many patients refused to have a nasogastric tube. Most patients found the crushed busulfan tablet slurry to be unpalatable when taken orally. In fact, busulfan tablets are placed in empty gelatin capsules for many patients in order to facilitate the administration of a large number of tablets and/or to mask their salty taste. Each administration method would be expected to display different absorption parameters (lag time, time to maximum concentration, maximum concentration), although the significance of the differences is unknown. The small sample size of this evaluation does not permit stratification of the data by route and method of administration. Our sample times (1, 1.5 and 6 h) may therefore not be appropriate or valid in every circumstance.

The sample times selected for the three-sample technique emphasize the absorptive phase of the busulfan disposition curve. Hassan *et al*[11] estimates the lag time after oral busulfan administration or the time until the first appearance of busulfan in the blood to be 1.5 min and the absorption rate constant to be 0.00525 min^{-1}. However, the variability around these parameters is 380% and 98%, respectively. Busulfan AUC may be more accurately described by a limited sampling model which better delineates the elimination phase of the busulfan disposition curve. The routine use of

Table 2 AUC achieved after initial busulfan dose of 40 mg/m^2 and proportion of busulfan courses requiring dose adjustment

Age (years)	*No. courses*	*Mean initial AUC (μM/min)*	*Dose increase (No. courses)*	*Dose decrease (No. courses)*	*No change (No. courses)*
0.17 to 1	5	1523 ± 874	1 (20%)	3 (60%)	1 (20%)
>1 to 5	13	1223 ± 546	8 (62%)	2 (15%)	3 (23%)
>5 to 12	13	2015 ± 587	1 (8%)	11 (85%)	1 (8%)
>12	8	2387 ± 1147	1 (13%)	7 (88%)	0
Total	39	1764 ± 858	11 (28%)	23 (59%)	5 (13%)

Table 3 Mean busulfan dose requirements to achieve an AUC of approximately 1300 μM/min

Age (years)	*No. courses*	*Mean final busulfan dose (± s.d.)*					
		mg/kg actual weight	*mg/kg ideal weight*	*mg/kg effective weight*	*mg/m^2 actual weight*	*mg/m^2 ideal weight*	*mg/m^2 effective weight*
0.17 to 1	5	2.12 ± 1.09	1.96 ± 0.79	1.97 (n = 1)	43.0 ± 18.3	41.4 ± 16.7	46.1 (n = 1)
>1 to 5	13	2.22 ± 0.73	2.21 ± 0.70	21.4 ± 0.38 (n = 6)	49.2 ± 14.9	49.0 ± 14.9	48.4 ± 7.8 (n = 6)
>5 to 12	13	1.13 ± 0.33	1.18 ± 0.30	1.16 ± 0.35 (n = 8)	30.9 ± 7.2	31.8 ± 7.4	32.9 ± 8.2 (n = 8)
>12	8	0.80 ± 0.32	0.79 ± 0.23	0.65 ± 0.20 (n = 3)	27.1 ± 9.8	26.8 ± 8.4	22.9 ± 7.0 (n = 3)

Table 4 Median busulfan dose requirements to achieve an AUC of approximately 1300 μM/min

Age (years)	*No. courses*	*Median final busulfan dose (range)*					
		mg/kg actual weight	*mg/kg ideal weight*	*mg/kg effective weight*	*mg/m^2 actual weight*	*mg/m^2 ideal weight*	*mg/m^2 effective weight*
0.17 to 1	5	1.71 (1.52–4.07)	1.53 (1.26–3.16)	1.97 (n = 1)	34.3 (30.8–75.0)	32.3 (26.2–66.3)	46.1 (n = 1)
>1 to 5	13	2.0 (1.4–3.7)	1.95 (1.3–3.7)	2.1 (1.8–2.8) (n = 6)	43.2 (32.7–82)	45.8 (28–82)	47.6 (41–61.6)
>5 to 12	13	1.05 (0.78–1.56)	1.11 (0.84–1.48)	1.05 (0.89–1.91) (n = 8)	27.6 (23.8–40.2)	28.4 (22.2–48.9)	27.7 (26.5–48.7)
>12	8	0.73 (0.38–1.44)	0.79 (0.47–1.19)	0.67 (0.44–0.84) (n = 3)	25.8 (20.0–46.8)	26.7 (16.0–42.5)	24.1 (15.4–29.2)

a four-sample calculation method (1, 1.5, 4 and 6 h) may therefore increase the validity of busulfan AUC calculation and dose adjustment. Ideally, routine use of the four-sample method would reduce the need to repeat busulfan concentration determinations with subsequent doses.

We were occasionally unable to interpret the busulfan levels observed. This occurred most commonly in patients with a delayed or prolonged absorption phase. Other investigators have also made this observation.[1,12,13] In the cohort reported by Dix *et al*,[1] 25% of all data sets were not evaluable due to lack of elimination phase data. This occurred in a higher proportion of night-time doses than morning doses. In our experience, elimination phase data could be interpreted when sampling was repeated after another morning busulfan dose. The high intra-individual variation in busulfan disposition is exemplified by this situation and by the fact that the AUCs achieved in our patients after dose adjustment, when verified, rarely (1/11) met the target exactly. Most of these patients, however, achieved an AUC after dose adjustment within the acceptable range (900–1400 μM). Other investigators have remarked on the poor predictability of the AUC determined after the first busulfan dose.[14] Dix *et al*[1] reported that repeat busulfan AUCs in five patients of unspecified age were 1.4 to 2.3 times lower

than expected. This finding was attributed to induction of busulfan metabolism by phenytoin and, possibly, to a systematic overestimation of the initial AUC. When adjusting busulfan doses to achieve a target steady-state concentration, other centers have reported that 'frequent' dose adjustments are required on dose 5 while dose adjustments are 'less often' required after dose 9.[15] A single AUC determination may not therefore be sufficient to predict the overall busulfan dose intensity experienced by an individual patient. Intravenous busulfan administration may significantly improve the predictability of dose adjustment.

Intra-individual variation in busulfan disposition may be caused by enzyme induction by phenytoin or phenobarbital given for seizure prophylaxis, auto-induction, circadian variation and underlying disease. Our finding of a higher than expected final busulfan AUC in approximately half of our patients with repeated AUC determinations does not support phenytoin induction as the sole cause of the observed variability. In addition, variations in steady-state busulfan concentrations of more than 500% (dose 1 *vs* dose 13) have been reported in children, most of whom did not receive phenytoin.[5] It is interesting that minimal intra-individual variation has been reported in assessments of morning busulfan doses given to a homogeneous population, none of whom received phenytoin or phenobarbital.[16] In this group of patients, the second AUC determination was within 20% of the first 90% of the time, although dose adjustments were not being made.

The inter-individual variability in the relationship between busulfan dose and AUC seen in our patients is consistent with the work of other investigators.[12,17,18] Sources of this variability may be related to product (2 mg *vs* 25 mg tablet), administration technique (n.g. *vs* p.o., administration of crushed *vs* intact tablets, administration in gelatin capsules), or the patient (gut and hepatic function, age, underlying disease).

Routine AUC calculation would be unnecessary if a single level reliably and accurately predicted AUC. Other investigators have observed that the busulfan concentration at 6 h after the dose (trough concentration) was predictive of the AUC achieved.[6,18] This observation was not confirmed in our larger and more varied group of children.

The effect of age on busulfan pharmacokinetic disposition is well known.[12,19,20] Children between the ages of 1 and 5 years have substantially higher apparent busulfan oral clearance than infants <1 year, older children or adults. This effect is likely due to higher hepatic and first pass intestinal clearance.[21] Bioavailability of busulfan does not appear to be influenced by age.[11] The apparent oral busulfan clearance in adolescents is similar to that of adults.[12] Our data reflect this relationship insofar as children aged 1 to 5 years required more drug to achieve the target AUC than did younger or older children. Vassal *et al*[18] have suggested that children below the age of 3 years should receive a total busulfan dose of 749 mg/m^2 or 47 mg/m^2/dose for 16 doses. We also observed a higher dose requirement in young children.

The final busulfan dose (mg/kg or mg/m^2) did not differ in patients whose actual weight was greater than their ideal body weight. This may not be true in infants <1 year of age but the small number of infants transplanted at our center during the review period precludes conclusions. Others have made similar observations in adults and adolescents.[14,22] However, a higher incidence of veno-occlusive disease (VOD) has been noted in obese adults who were not dosed according to ideal body weight although dose intensity was not standardized by means of concentration monitoring.[23]

Table 5 Recommended dosage guidelines for the initial busulfan dose

Patient age (years)	*Initial busulfan dose (mg/m^2/dose[a])*
0.17 to 1	35
>1 to 5	45
>5 to 12	30
>12	25

[a]Based on actual weight.

The relationship between busulfan dose intensity and either toxicity or efficacy is complicated. Due to the multiple diagnoses, donor types and variable extent of pre-existing hepatic toxicity within our study group, we are unable to comment on the contribution of busulfan dose adjustment to either engraftment, VOD, or survival in our patients. These relationships may depend on the underlying diagnosis and/or the conditioning regimen used. Higher busulfan clearance and lower AUC have been observed in a mixed group of 12 children with leukemia or lymphoma relative to children with inherited disorders.[17] The fact that the majority of patients examined in this report had leukemia may have skewed our results. However, there are no published data regarding the influence of underlying disease on busulfan disposition in children which would allow disease-specific refinement of our dosing recommendations or our target AUC. Some centers will not administer busulfan doses less than 1 mg/kg/dose at least for some diagnoses, regardless of the individual patient's calculated AUC.[24]

Based on this retrospective analysis of busulfan dose adjustment at our hospital, we recommend that the initial busulfan dose be adjusted for patient age (Table 5) and that it be calculated using body surface area determined using the actual patient's weight with no adjustment for obesity. Given the many factors which influence busulfan disposition, dose adjustment for age does not obviate the need for busulfan AUC calculation and dose individualization. We recommend that the busulfan AUC be calculated using four data points (1, 1.5, 4 and 6 h). The ability of these interventions to reliably produce acceptable busulfan AUCs must be prospectively evaluated.

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