Busulfan with 400 Centigray of Total Body Irradiation and Higher dose Fludarabine. An alternative regimen for Hematopoietic Stem Cell Transplantation in Pediatric Acute Lymphoblastic Leukemia.

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September 4, 2023

Abstract

Background Hematopoietic stem cell transplantation can be curative for children with difficult to treat leukemia. The conditioning regimen utilised is known to influence outcomes. We report outcomes of the conditioning regimen used at the Alberta Children's Hospital, consisting of busulfan (with pharmacokinetic target of 3750µmol*min/day +/-10%) for 4 days, higher dose (250 mg/m2) fludarabine and 400 centigray of total body irradiation. **Procedure** This retrospective study involved children receiving transplant for acute lymphoblastic leukemia (ALL). It compared children who fell within the target range for busulfan with those who were either not measured or were measured and fell outside this range. All other treatment factors were identical. **Results** Twenty-nine children (17 within target) were evaluated. All subjects engrafted neutrophils with a median (IQR) time of 14 days (8-30 days). The cumulative incidence of acute graft versus host disease was 44.8% (95% CI 35.6 – 54.0%), while chronic graft versus host disease was noted in 16.0% (95% CI 8.7% - 23.3%). At two years, the overall survival was 78.1% (95% CI 70.8% - 86.4%) and event free survival was 74.7% (95% CI 66.4% - 83.0%). Cumulative incidence of relapse was 11.3% (95% CI 5.1% - 17.5%). There were no statistically significant differences in between the group that received targeted busulfan compared with the untargeted group. **Conclusion** The current regimen used in children with ALL results in outcomes comparable to standard treatment with acceptable toxicities and significant reduction in radiation dose. Targeting Busulfan dose in this cohort did not result in improved outcomes.

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Word count

Structured Abstract : 249 words

Main text (excluding title page, abstract, Conflicts of Interest, Acknowledgments, References, Tables, Figures, and Legends): 3475 words

Count of Figures and Tables

Figures: 5

Tables: 1

Short Running Title

Bu, Flu and 400cGy TBI for HSCT in Pediatric ALL.

Keywords

Pediatric, Acute Lymphoblastic Leukemia, allogeneic transplant, low-dose radiation, busulfan, fludarabine

List of Abbreviations (in alphabetical order)

ACH	Alberta Children's Hospital
aGvHD	Acute graft versus host disease
ALL	Acute lymphoblastic leukemia
ATG	Anti-thymocyte globulin
AUC	Area under the curve
BM	Bone marrow
Bu	Busulfan
CAR-T	Chimeric antigen receptor T-cell
cGvHD	Chronic graft versus host disease
CHC	Chemotherapy-based conditioning
CI	Confidence interval
CIR	Cumulative incidences of relapse
CMV	Cytomegalovirus
Cy	Cyclophosphamide
EBV	Epstein Barr Virus
EFS	Event free survival
Flu	Fludarabine
FORUM	For Omitting Radiation Under Majority age
GvHD	Graft versus host disease

ACH	Alberta Children's Hospital
HLA	Human leukocyte antigen
HSCT	Hematopoietic stem cell transplantation
IgG	Immunoglobulin G
IQR	Interquartile range
IV	Intravenous
MRD	Minimal Residual Disease
NRM	Non-relapse mortality
OS	Overall survival
PBSC	Peripheral blood stem cell
SOS	Sinusoidal obstructive syndrome
TBI	Total body irradiation
TDM	Targeted drug monitoring
Treo	Treosulfan

Abstract

Background

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Procedure

This retrospective study involved children receiving transplant for acute lymphoblastic leukemia (ALL). It compared children who fell within the target range for busulfan with those who were either not measured or were measured and fell outside this range. All other treatment factors were identical.

Results

Twenty-nine children (17 within target) were evaluated. All subjects engrafted neutrophils with a median (IQR) time of 14 days (8-30 days). The cumulative incidence of acute graft versus host disease was 44.8% (95% CI 35.6 – 54.0%), while chronic graft versus host disease was noted in 16.0% (95% CI 8.7% - 23.3%). At two years, the overall survival was 78.1% (95% CI 70.8% - 86.4%) and event free survival was 74.7% (95% CI 66.4% - 83.0%). Cumulative incidence of relapse was 11.3% (95% CI 5.1% - 17.5%). There were no statistically significant differences in between the group that received targeted busulfan compared with the untargeted group.

Conclusion

The current regimen used in children with ALL results in outcomes comparable to standard treatment with acceptable toxicities and significant reduction in radiation dose. Targeting Busulfan dose in this cohort did not result in improved outcomes.

Introduction

Treatment for acute lymphoblastic leukemia (ALL) is based on established risk stratification criteria that also include response to therapy. Such measures have increased the rates of survival from less than 10% in the 1960s to over 90% today.[1]

Hematopoietic stem cell transplantation (HSCT) has a role in the management of difficult-to-treat leukemia by inducing cures in these children.[2,3]. One factor influencing outcomes is the choice of the conditioning regimen.[2,4] HSCT regimens have utilised a combination of total body irradiation (TBI) and high dose cyclophosphamide (Cy) and/or other agents such as etoposide.[5,6,19] However TBI has been associated with significant adverse effects including neurocognitive decline, endocrine and metabolic concerns as well as apprehensions in regards to secondary malignancies.[7,19] Busulfan (Bu) is a bifunctional DNA alkylating agent that has been used to replace TBI. Initial studies that incorporated oral Bu in combination with Cy resulted in poor outcomes when compared with Cy/TBI.[7] This was mainly attributed to the toxicity of oral Bu, specifically the incidence of sinusoidal obstructive syndrome (SOS).[5,8] The erratic absorption of Bu following oral administration with resultant unpredictable exposure was another contributing factor. This was especially concerning in pediatric patients owing to their higher drug clearance and less predictable pharmacokinetic profiles. [9-11]

The advent of an intravenous formulation of Bu solved the problem of erratic oral absorption however, interpatient variability in clearance was a persisting concern. Targeted drug monitoring (TDM) of Bu in children, once introduced, allowed for better prediction of serum drug levels.[49] Although not consistently, subsequent studies have shown improvements in outcomes following TDM of Bu.[12]

At the Alberta Children's Hospital, Bu (with comparatively lower pharmacokinetic target of 3750µmol*min/day for 4 days, with a preceding test dose) is combined with higher dose (250 mg/m^2) fludarabine (Flu), 400 cGy of TBI and anti-thymocyte globulin (ATG) in a regimen that was initially used in adults and later adapted for HSCT in children.[13]

We report our experience with this conditioning regimen for children diagnosed with ALL requiring HSCT.

Methods

This is a retrospective analysis performed at the Alberta Children's Hospital (ACH) in Calgary, Canada. The inclusion criteria for the study were: age less than 18 years at time of transplant, a diagnosis of ALL requiring HSCT, first transplant, Bu/Flu/ATG + TBI conditioning regimen and transplant using bone marrow (BM) or peripheral blood stem cell (PBSC) products.

Institutional Review Board approval and waiver of consent was obtained prior to accessing the electronic records which spanned 17 years, from 2003 to 2020.

Conditioning

Prior to 2008 Bu was administered as an intravenous (IV) infusion on day -5 through -2 at a dose that was age determined (4mg/kg/dose in children less than four years of age and 3.2 mg/kg/dose if older) and without pharmacokinetic monitoring. Between 2008 and late 2010 pharmacokinetic monitoring was introduced and thereafter (In November 2010) a TDM dosing regimen was applied. In this regimen a test dose of Bu (25% of the actual dose) was administered on day -7. Serum drug levels were measured at end of infusion and 1, 3, 5 and 7 hours later. Based on the area under the curve (AUC) generated, the dosage of the Bu was adjusted. The first full dose of Bu was administered on day -5 and the drug levels were repeated to ensure adequate exposure to the drug. An AUC of 3750µmol*min was targeted across all Bu days including the test dose (total exposure 15000umol*min). Flu was administered on day -6 through -2 at a dose of 50 mg/m^2 infused IV over one hour once . Rabbit - ATG (Thymoglobulin ® Sanofi Aventis) was given to all patients as a three-day course starting with 0.5 mg/kg on the first day (day -3) and weight-based dosing for the remaining two days (children over 30 kg received 2 mg/kg/dose and those less than 30kg received 2.5 mg/kg/dose, on days -2 to -1). Serum levels for ATG were not performed. Finally, 400 cGy TBI was given to all children in two divided doses on day -1.

Donor Sources

Donor and recipient human leukocyte antigen (HLA) matching was performed using the sequence based typing method until 2019 when next generation sequencing was introduced. HLA matching was done at 10

alleles for both PBSC and BM sources. Donors who were identical on all 10 alleles were considered matched, while those who were nine of ten were labelled mismatched. A haploidentical transplant was defined as any donor matching at five to eight out of 10 alleles.

GVHD prophylaxis and treatment

Cyclosporine was used for graft versus host disease (GvHD) prophylaxis in all subjects starting day -1 with dosage adjusted based on serum levels to target a therapeutic concentration between 150ng/ml and 200ng/ml. In addition, methotrexate 15 mg/m^2 was administered on day 1 and 10mg/m^2 on days 3, 6 and 11 post-transplant. Cyclosporine was subsequently weaned over five weeks starting on day 42 post-transplant in the absence of any GvHD.

Grading of GvHD was based on the modified Glucksberg criteria for acute graft versus host disease (aGvHD) and the National Institute of Health (NIH) criteria for chronic graft versus host disease (cGvHD).[14,15] Once diagnosed, GvHD was managed as per institutional guidelines.

Supportive care

All patients received seizure prophylaxis during the administration of Bu and infectious prophylaxis with acyclovir, fluconazole, metronidazole and pentamidine (switched, after the first month, to sulfamethoxazole and trimethoprim). Cytomegalovirus (CMV) and Epstein Barr Virus (EBV) titres were monitored once weekly as were immunoglobulin G (IgG) levels. Chimerism testing was performed routinely on days 21 and 100 post-transplant and a level of donor cells more than 95% was considered complete donor chimerism.

Definitions and End points

In this study, the primary end point was Event Free Survival (EFS) at two years post-transplant. This was defined as the proportion of patients who remained alive and free from disease relapse and secondary malignancies. Overall survival (OS), defined as the proportion of subjects alive after the transplant as measured from the day of transplant to date of death or censored at day of last follow up, was also calculated.

Secondary end points included neutrophil engraftment which was defined as the first of three days with neutrophil count more than 500 cells/µl post-transplant, and platelet engraftment as the first day of platelets over 20,000 cells/ul for seven days without support with platelet transfusions.

The frequency and severity of GvHD (both and acute and chronic) were also calculated as were the rates of infections and SOS.

Statistical Analysis

For this analysis, we combined patients between 2008 and 2010 who met our assigned target AUC, with those transplanted after 2010 (who had Bu doses adjusted after pharmacokinetic monitoring). This targeted cohort was compared to an untargeted group. The untargeted group comprised of children who were transplanted before 2008 combined with those who had Bu measurements between 2008 and 2010 were outside the target range and were not corrected (Figure 1).

Patient characteristics and transplant data were described and proportions of the various characteristics between the Bu-targeted and untargeted groups. The two groups were compared using the Mann-Whitney U test for age at transplant and chi-square test for the nominal variables to detect the presence of any significant differences.

Neutrophil and platelet engraftment were described highlighting proportions of children successfully engrafting and the median time to engraftment. Cumulative rates of incidence of acute and chronic GvHD, viral infections and SOS were calculated using the Kaplan-Meier method. This analysis was also used to calculate the EFS and OS, cumulative incidences of relapse (CIR) and non-relapse mortality (NRM). The resultant values were compared using a log rank test between the Bu targeted and untargeted groups. Finally, a univariate analysis was performed to study the effect of variables on the EFS and OS. All statistical analysis was performed using SPSS version 26.

Results

Patient Demographics

Over the 18-year period from 2003 to 2020, 29 patients met eligibility criteria. They were split into the targeted group which comprised of 17 subjects and the untargeted group which was made up of the remaining 12 subjects.

Patient Demographic data is presented in Table 1. The recipients had a median age of 13 years (range 2-18 years) and 72% were male. All patients were in morphologic remission at the time of transplant. Fourteen patients (48%) were in CR1 and were transplanted due to high-risk disease: Eight had persistently positive Minimal Residual Disease (MRD), three were Philadelphia or Philadelphia like chromosome positive, two had hypodiploid ALL and one patient had bi-phenotypic leukemia. The remaining 15 (52%) were transplanted in CR2. Data on MRD status prior to transplant was available for 25 subjects. Three-quarters of this group (65% of the entire cohort) had a level below 0.01% by flow cytometric analysis and were considered negative.

Engraftment

Neutrophil engraftment was seen in all patients at a median of 14 days (range: 8-30 days). Platelet engraftment was successful in 28 of the 29 patients, the remaining one died prior to engraftment. The median time to platelet engraftment was 16 days (range 0-89 days). Two patients had engraftment beyond day 60, one of whom required continued platelet infusion due to persistent hematuria.

Graft versus host disease

Fourteen of the 29 subjects developed grade 2 or higher aGvHD, resulting in a cumulative incidence at day 100 of 44.8% (95% CI 35.6 – 54.0%). Severe aGVHD (grade three or higher) was seen in six patients which gave a cumulative incidence at day 100 of 20.7% (95% CI 9.9 – 40.3%).

Chronic graft versus host disease (cGvHD) was seen in four subjects. The cumulative incidence of cGvHD at one year was 16.0% (95% CI 8.7% - 23.3%). cGvHD was graded as severe in three of the four subjects. All three had pre-existing aGvHD.

Toxicity

CMV infection was documented in nine of 29 subjects in the cohort giving a cumulative incidence of 31% (95% CI 22.4% - 39.6%) at one year. The median time to reactivation was 24 days (IQR of 16 - 34 days). Two of these 9 children had a primary infection. Clinically significant EBV reactivations that required pre-emptive treatment were documented in five of 27 patients for whom data was available. Culture proven bacterial infections were documented in nine subjects within the first 100 days post-transplant. Gram-positive organisms were cultured in blood in two-thirds of positive cases. Finally fungal infections were seen in two subjects, both of whom developed oral thrush.

Grade three mucositis was seen in all 29 subjects in this analysis. Sinusoidal obstructive syndrome (SOS) was seen in three of 20 patients in the cohort for whom data was available. The cumulative incidence at 100 days post transplant was 15.0% (95% CI 7.0% - 23.0%). Only one participant had severe SOS resulting in death from multi-organ dysfunction.

Survival analysis

The median follow-up for the cohort was 3.25 years (1-10 years). There were six deaths recorded, all occurring within two years of transplant resulting in a 2-year overall survival for the entire cohort of 78.1% (95% CI 70.8% - 86.4%). The Bu targeted group had a similar 2-year overall survival calculated at 79.9% (95% CI 69.4% - 90.4%) compared to the untargeted group which was 75.0% (95% CI 62.5% - 87.5%). This difference did not reach statistical significance, p value = 0.6 (Figure 2).

In the subjects who succumbed, two deaths were due to progressive disease post relapse, one patient was lost to SOS and another to causes directly related to cGvHD. The remaining two children died due to progressive

organ dysfunction.

Overall, there were a total of seven events. No events occurred after 2 years of transplantation. This resulted in a 2-year EFS of 74.7% (95% CI 66.4% – 83.0%). In the Bu targeted group, the 2-year EFS was 79.9% (95% CI 69.4% - 90.4%) compared to the untargeted group: 66.7% (95% CI 53.1% - 80.3%), p = 0.4. (Figure 3).

Three children in the cohort relapsed post-transplant, giving a cumulative incidence of relapse at two years of 11.3% (95% CI 5.1% - 17.5%). One of the three children went on to receive a successful second transplant (Figure 4).

The non-relapse mortality calculated at two years was 15.4% (95% CI 8.3% - 22.5%) for the entire group and was 15.4% (95% CI 5.4% - 25.4%) and 16.7% (95% CI 5.9% - 27.5%) p = 0.7 in the Bu targeted and untargeted groups respectively (Figure 5).

Univariate analysis did not reveal any factors, including pre-transplant MRD status, that correlated significantly with the risk of death or relapse.

Discussion

The conditioning regimen used for our children with acute leukemia was initially developed for adult transplant recipients. It included Bu (targeted at an AUC of 3750µmol*min) and Flu (dosed at 250mg/m^2 total), to which ATG was added to reduce the incidence of GvHD.[13,16] TBI at 400cGy was included in the protocol to reduce the incidence of relapse and was noted to significantly improve outcomes.[17] Our protocol uses a higher Flu dose compared with other centers using similar regimens.

Survival outcomes, relapse incidence and non-relapse mortality in our cohort were similar between the subjects who had TDM and those who did not. The number of subjects in our cohort played an important role in the results not reaching significance levels. Previous studies that have compared Bu based conditioning to TBI regimens in children with malignancies have presented outcomes favouring TBI based conditioning.[8,18] This has mainly been due to higher rates of relapse and non-relapse mortality rates in the Bu groups. However, these earlier studies have not consistently reported measurements of Bu serum levels.[8,18,47]

Recently, the results of the international FORUM study (For Omitting Radiation Under Majority age), a multicenter randomised trial comparing a fractionated TBI/Etoposide regimen to various chemotherapy-based conditioning (CHC) regimens has shown conclusively that a TBI based conditioning regimen is superior for children above four years of age with ALL.[19] In that study individual institutions were allowed to choose between Bu and Treosulfan (Treo) as a backbone for their CHC regimens. A potential drawback to this flexibility is the introduction of heterogeneity.[51] Therefore, despite an overall equivalence between the Bu and Treo based regimens, data does not allow a direct comparison across individual CHCs. Furthermore, Bu serum level targeting was allowed but not mandated by the FORUM study and no analysis is done to compare outcomes between the centers that used serum targeting of Bu and those that did not.[51]Additionally, although cranial radiation was allowed for central nervous system positive disease, none of the CHC regimens in the FORUM study included TBI. For these reasons, a direct comparison of outcomes with the regimen used in the current study is not appropriate.

In 2007, Chaudhury et al published an abstract reporting their findings using a regimen incorporating both Bu and TBI.[20]Although the targets for Bu TDM were similar (4000 µmol *min/day), an Important difference in their preparation regimen was their use of lower cumulative doses of Flu (150 mg/m^2 over five days). They reported a one-year OS and PFS of 63% and 56% respectively. A recent article by Rosoff et al further expanding analysis of the same cohort, using Bu-Flu-ATG and 400cGy TBI regimen, reported a five-year OS of 37% in their ALL cohort consisting of only nine children (one of whom had positive MRD).[21] In both studies by Chaudhury and Rosoff the most important reason for failure was high rates of relapse, seen in 29% and 28% of subjects respectively.[20,21] We speculate that the lower cumulative incidence of relapse in our cohort may potentially be related to the higher doses of Flu. Evidence for this has recently been published in both transplant and Chimeric Antigen Receptor T-cell (CAR-T) settings with studies showing

that both lower and very high serum Flu levels result in inferior outcomes driven by increased relapse rates and toxicity respectively.[52,53]

Although the rates of aGvHD reported in this cohort are higher than those in other studies, which quote incidences closer to 30%,[18,21,23] they are similar to results reported by Chaudhury et al who report their severe aGvHD rate as being 32%.[20] A striking similarity between both studies is the use of PBSC products, utilised in close to 90% of transplants in the current study and in 75% of cases in the study by Chaudhury. PBSC products have been shown to be associated with increased aGvHD in multiple studies although the difference has not been consistently significant.[24,25,48] Six (20%) of the donor-recipient pairs in our study were female donors to male recipients. Pulsipher et al, have shown that the presence of aGvHD is related to a lower risk of relapse,[50] which perhaps contributed to the EFS in the current study.

The cumulative incidence of cGvHD in our study was comparable with most other studies. [18,20,29] It was lower than rates reported by Rosoff et al and those in a report by Modi et al (which used a similar conditioning regimen for adults with AML).[21,23] The reason for the low rates of cGvHD in our study despite a higher incidence of aGvHD may be explained by our method of giving ATG.[30,31] At our institution we follow a regimen similar to one recommended by Bacigalupo et al.[31] In their 2001 paper, they suggested all patients receive ATG at 7.5mg/kg as this dose was not associated with increased risk of infections and was shown to reduce the incidence of cGvHD.

The incidence and timing of viral infections seen in our cohort were similar to those reported in literature[32-34,36] with the exception of CMV which appeared to occur earlier than some studies have described (median of 24 days in our study compared with 33 to 41 days in some studies).[34,54] There was no effect on outcomes which is contrary to findings in other publications,[32,35] with the exception of delayed platelet engraftment dates in those subjects receiving platelet infusions for hematuria secondary to BK virus induced cystitis Bacterial blood stream infection incidence, timing and the organisms grown were also comparable to other reports.[37-41] The incidence of mucositis was 100% in this cohort which is in keeping with other reports using a similar regimen.[21]

The incidence of SOS in studies using conditioning regimens including Bu and Flu compared to other combinations have had mixed results with some studies reporting a reduction in SOS[42-44] and others showing no difference.[45] In our cohort the cumulative incidence of SOS at d100 was 15% (three patients). Although this rate appears higher compared with other studies, it may not be significant owing to the small number of patients developing SOS.[20,46] In the adult study by Kabriaei et al, six patients in their cohort of 107 patients developed SOS resulting in a prevalence of 6%.[46] Similarly, Chaudhury et al reported a SOS prevalence of 10% (three cases).[6] It is notable that SOS was cited as a direct cause of death in only one case in each of the two studies. The univariate analysis in our cohort revealed no significant correlation of the occurrence of SOS with mortality.

The limitations to our study include firstly, that this is a retrospective chart review and therefore dependant on the quality of data collected and stored. Data pertaining to SOS were missing for our earlier subjects. Secondly, the number of patients, once split into the two cohorts, was limited and this may have contributed to the lack of significance when comparing the outcomes following Bu pharmacokinetic targeting. Furthermore, the comparison with a historic cohort introduces some inconsistencies as there is a possibility that improvements in outcomes over the years were in fact due to an unidentified confounder and not the practice of targeting Bu serum levels. It should be noted however, that no significant changes to supportive care were introduced during the period of data collection. Related to this, the combination during analysis of those children who did not have any Bu monitoring with the group that were monitored and were out of range may have introduced a bias, as the earlier group is a heterogenous one and may contain some children who were within acceptable serum levels. Finally, we did not differentiate between T-cell and B-cell ALL which introduces heterogeneity in the cohort analysed. Despite these limitations, the paper documents outcomes following transplant in children with ALL using a unique and previously unpublished regimen of Bu-ATG and lower dose TBI regimen combined with a higher dose of Flu. Although previous studies have utilized lower dose radiation in this setting, those cohorts have been of limited size compared with the current report.

We have demonstrated similar outcomes to other conditioning regimens when serum Bu levels are targeted suggesting this may be a substitute to consider when lowering radiation dose is desirable.

Conclusion

The use of a conditioning regimen for allogeneic hematopoietic cell transplantation in children with ALL using Bu along with high dose Flu, ATG and 400 Centigray of TBI results in outcomes comparable to standard treatment with acceptable toxicities and significant reduction in the dose of irradiation. Targeting the Bu dose based on pharmacokinetic monitoring did not however improve outcomes in this cohort.

Conflict of Interest Statement

The authors have no conflicts of interest to declare.

Acknowledgements

The authors would like to acknowledge the contribution of the Alberta Health Services.

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Figure Legends:

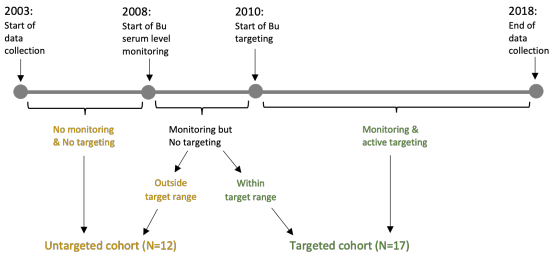
Figure 1: graphical representation of the cohorts for analysis

Figure 2: Overall survival stratified for busulfan pharmacokinetic targeting. The targeted group 3-year OS of 79.9% (95% CI 69.4% - 90.4%) compared with the untargeted group: 75.0% (95% CI 62.5% - 87.5%). P value = 0.6

Figure 3: Event Free survival stratified for busulfan pharmacokinetic targeting. The targeted group 2-year EFS of 79.9% (95% CI 69.4% - 90.4%) compared with the untargeted group: 66.7% (95% CI 53.1% - 80.3%). P value = 0.4

Figure 4: Cumulative incidence of relapse (CIR). 3 events, with CIR = 11.3% (95% CI 5.1% - 17.5%)

Figure 5: Non-Relapse mortality stratified for busulfan pharmacokinetic levels. The targeted group NRM of 15.4% (95% CI 5.4% - 25.4%) compared with the untargeted group: 16.7% (95% CI 5.9% - 27.5%). P value = 0.7



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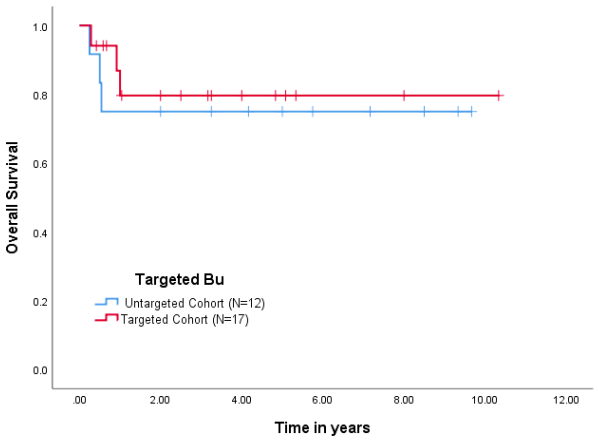

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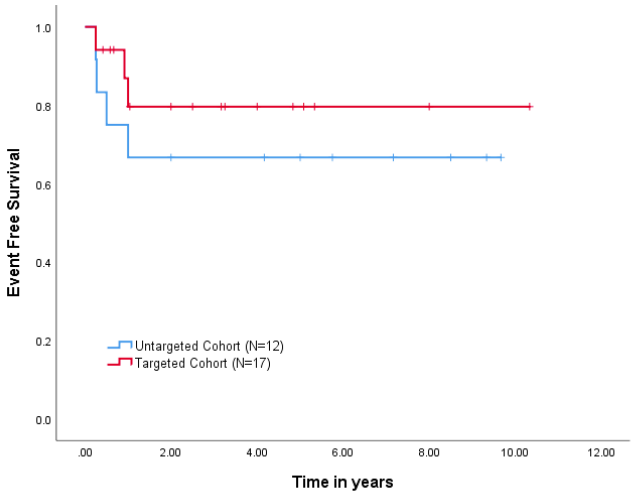

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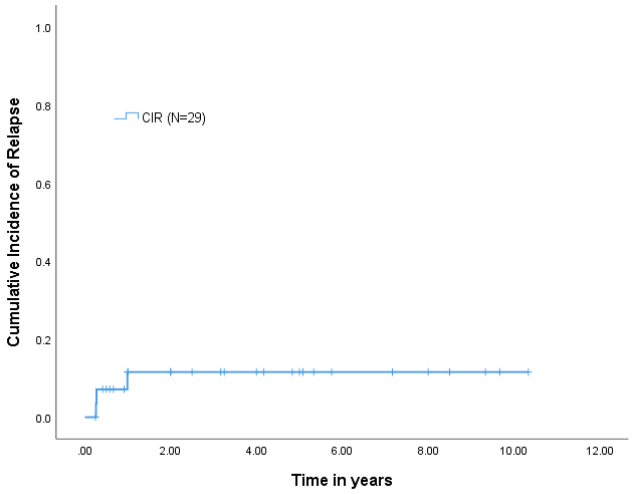

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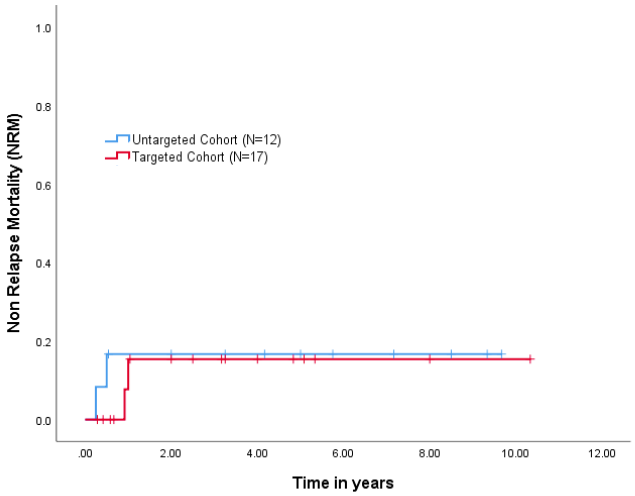

Figure 5: Non-Relapse mortality stratified for busulfan pharmacokinetic levels. The targeted group NRM of 15.4% (95% CI 5.4% - 25.4%) compared with the untargeted group: 16.7% (95% CI 5.9% - 27.5%). P value = 0.7

TABLE 1 Characteristics of children undergoing transplant.

VARIABLE	TOTAL =29 number (%)	TARGETED=17 number (%)	UNTARGETED=12 number (%)	P
Age: median (IQR)	13.0 (9.0 – 15.0)	13.0 (9.5– 14.5)	12.5 (7.5 – 15.0)	1.0
Patient sex:				
Male	21 (72)	11 (65)	10 (83)	0.27
Female	8 (28)	6 (35)	2 (17)	
Minimal Residual Disease (N=25):				
Positive ≥0.01%	6 (24)	5 (31)	1 (11)	0.26
Negative <0.01%	19 (76)	11 (69)	8 (89)	
Stem cell source:				
PBSC	27 (93)	16 (94)	11 (92)	0.80
BM	2 (7)	1 (6)	1 (8)	
Donor:				
Related	13 (45)	6 (35)	7 (58)	0.22
Unrelated	16 (55)	11 (65)	5 (42)	
HLA matching:				
Matched	27 (93)	15 (89)	12 (100)	0.22
Mismatched	2 (7)	2 (11)	0 (0)	
Recipient CMV Status:				
Negative	17 (59)	8 (47)	9 (75)	0.13
Positive	12 (41)	9 (53)	3 (25)	

BM – bone marrow; CMV – cytomegalovirus; CR – complete remission; HLA – human leukocyte antigen; IQR – interquartile range; PBSC – peripheral blood stem cells