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# Comparison of Outcomes of Allogeneic Transplantation for CML with Cyclophosphamide in Combination with Intravenous Busulfan, Oral Busulfan or TBI

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## Abstract

Cyclophosphamide in combination with busulfan (Bu) or total body irradiation (TBI) are the most commonly used myeloablative conditioning regimens in patients with Chronic Myeloid Leukemia (CML). We used data from the Center for International Bone Marrow Transplantation Research to compare outcomes in adults who underwent hematopoietic cell transplantation for CML in first chronic phase following myeloablative conditioning with cyclophosphamide (Cy) in combination with TBI, oral Bu or intravenous (IV) Bu. Four hundred thirty-eight adults received human leukocyte antigen (HLA)-matched sibling grafts and 235 received well-matched grafts from unrelated donors from 2000 through 2006. Important differences existed between the groups in distribution of donor relation, exposure to tyrosine kinase inhibitors and year of transplantation. In multivariate analysis, relapse occurred less frequently among patients receiving IV Bu compared to TBI (RR=0.36; P=0.022) or oral Bu (RR=0.39; P=0.028), but non-relapse mortality and survival were similar. A significant interaction was detected between donor relation and the main effect in leukemia-free survival (LFS). Among recipients of HLA-identical sibling grafts, but not URD grafts, LFS was better in patients receiving IV (RR=0.53; P=0.025) or oral Bu (RR=0.64; P=0.017) compared to TBI. In CML in first chronic phase, Cy in combination with IV Bu was associated with less relapse than TBI or oral Bu. LFS was better following IV or oral Bu compared to TBI.

## Introduction

Tyrosine kinase inhibitors (TKIs) have replaced allogeneic hematopoietic cell transplantation (HCT) as initial therapy of patients with chronic myeloid leukemia (CML). Nevertheless, many patients with CML eventually receive an allotransplant. Determining the best pretransplant conditioning regimen is important.

Cyclophosphamide combined with total body irradiation (Cy/TBI) has historically been the standard pretransplant conditioning regimen. [1-4] The combination of Cy with a fixed dose of oral busulfan (BuCy) has also proven effective in CML.[5] A randomized comparison of Cy/TBI to BuCy in patients with CML undergoing human leukocyte antigen (HLA)-identical sibling transplantation reported comparable relapse, leukemia-free survival (LFS) and overall survival (OS). BuCy was better tolerated, however, with shorter hospitalization

and less acute graft-versus-host disease (GvHD).[6] A second randomized study reported similar outcomes but with fewer relapses in the BuCy cohort. [7]

The development of an assay for plasma Bu was initially reported in 1983, [8] but an assay was not commercially available until 1996. [9] Studies of Bu kinetics revealed that oral Bu is erratically absorbed and that oral administration of a fixed-dose results in wide variations in plasma Bu levels.[10,11,12,13] Low plasma levels are associated with increased risks of graft-failure and relapse and high levels with increased toxicity. [10,11,12] Dose adjustment of oral Bu, based on plasma levels following the initial dose, decreases the variability and may improve outcomes.[14] An intravenous (IV) formulation of Bu was developed and its use in patients was first reported in 2002. [15,16] It provides complete bioavailability, much more consistent plasma levels and less acute toxicity and 100-day mortality than an oral fixed-dose.[15,16] Although a retrospective study in Acute Myeloid Leukemia (AML) from the European Group for Blood and Marrow Transplantation failed to show significant differences in outcome, [17] a recent large retrospective study in patients with AML in first remission from the Center for International Bone Marrow Transplant Research (CIBMTR) reported significantly less non-relapse mortality (NRM) and late relapse, and better LFS and OS with Cy in combination with IV, but not oral, Bu compared with TBI. [18] A recent prospective cohort analysis in persons with MDS, AML and CML reported better survival following IV Bu than with TBI.[19]

No prospective or retrospective study has compared Cy in combination with IV Bu, oral Bu or TBI in patients with CML in chronic phase. We used data from the CIBMTR to compare outcomes following these regimens.

## Patients and methods

### Data sources

The CIBMTR is a working group of more than 500 transplant centers worldwide that voluntarily contribute data on allogeneic and autologous transplants. Detailed demographic, disease, and transplant characteristics and outcome data are collected on a sample of registered patients including all unrelated donor (URD) transplants facilitated by the National Marrow Donor Program in the United States. Observational studies conducted by the CIBMTR are carried out with a waiver of informed consent and in compliance with HIPAA regulations as determined by the Institutional Review Board and the Privacy Officer of the Medical College of Wisconsin.

### Patients

The study population consisted of all patients ≥ 18 years of age reported to the CIBMTR who received a first HCT with an HLA-identical sibling or well-matched URD[20] from 2000-2006 for CML in first chronic phase after pretransplant conditioning with Cy/TBI (single-dose ≥5.5 Gy, fractionated ≥9 Gy) or Bu (≥9mg/kg) combined with Cy and no other anti-cancer drugs. The data set was derived from CIBMTR comprehensive report forms. Patients with a genetically-identical twin or cord blood donor, an ex vivo T cell depleted

graft, a less than well-matched URD, or receiving Cy post-transplant were excluded. Data regarding Bu pharmacokinetics (PK) were not collected.

### Study end points and definitions

The primary outcome studied was overall survival. Patients were considered to have an event at the time of death from any cause; survivors were censored at last contact. NRM was defined as death without evidence of leukemia recurrence; relapse, defined by hematologic, cytogenetic, or molecular criteria, was considered a competing event. LFS was defined as time to treatment failure (death or relapse). For relapse, NRM and LFS, patients alive in continuous complete remission were censored at last follow-up. Times to neutrophil and platelet recovery were calculated as the time from transplantation to achieving the first of three consecutive days with neutrophils $>0.5 \times 10^9$/L and platelets $> 20 \times 10^9$/L, 7 days from the last platelet transfusion. Acute GvHD was graded according to consensus criteria based on the pattern of severity of abnormalities in skin, gastrointestinal tract and liver. [21] Chronic GvHD was diagnosed by standard criteria. [22] For hematopoietic recovery and GvHD, death without the event was considered a competing event.

### Statistical Methods

In univariate analysis, probabilities of LFS and OS were calculated using the Kaplan-Meier method, with the variance estimated by Greenwood's formula. Hematopoietic recovery, GvHD, NRM, and relapse were estimated using the cumulative incidence method to account for competing risks.

In multivariate analysis, a forward stepwise selection procedure was performed using the proportional hazards Cox model for OS, LFS, NRM, GvHD, and relapse to adjust for the following variables considered for inclusion in each model - subject: age, gender, and Karnofsky performance score at transplant; disease: interval from diagnosis to transplant and TKI use prior to HCT, and transplant-related: donor-recipient gender and Cytomegalovirus (CMV) serological status, donor relation and graft source, year of transplant, ATG or alemtuzumab use, GvHD prophylaxis, and planned use of growth factors post-transplant. $P<0.05$ was used to select variables to enter and to retain as covariates in the model. The proportional hazards assumption was assessed for each variable by testing its time dependency. Interactions were checked between each selected variable and the main effect.

Adjusted LFS and survival probabilities were estimated through the direct adjusted survival curves estimation method.[23] SAS software, version 9.3 (SAS Institute, Cary, NC) was used in all analyses.

## Results

### Demographics and univariate analysis

Six hundred seventy-three adults received a first HCT from an HLA-matched sibling (N=438) or well-matched unrelated donor (URD; N=235) from January 1, 2000 through December 31, 2006 for CML in first chronic phase following myeloablative preparation with Cy combined with TBI, oral Bu or IV Bu. The median follow up of surviving patients

is five years. Characteristics of patients categorized according to pretransplant conditioning regimen are described in Table 1. Patients who received IV Bu were older (median age 39 years, 42% > 40 years) than those receiving TBI (median age 35 years, 31% > 40 years) or oral Bu (median age 34 years, 29% > 40 years). Eighty-three percent of patients receiving oral Bu and 67% receiving IV Bu received a transplant from an HLA-identical sibling compared to 36% of those receiving TBI. Sixty-seven percent of IV Bu patients, compared to 27% of oral Bu and 36% of TBI patients, received at least one TKI before transplant. Nine percent of oral Bu and 13% of IV Bu patients received anti-thymocyte globulin (ATG) or alemtuzumab compared to 18% of those receiving TBI. Sixty-eight percent of IV Bu patients, compared to 37% oral Bu and 23% of TBI patients, underwent HCT from 2004-2006, the last 3 years of study. Median and interquartile range (IQR) of radiation dose was 12 Gy (IQR: 12-13.2 Gy) and Cy dose was 119.5 mg/kg (IQR: 98 – 120 mg/kg) for patients receiving TBI. Median Cy doses were 119 mg/kg (IQR: 105 – 120 mg/kg) and 109 mg/kg (IQR: 98 – 120 mg/kg) for those receiving oral and IV Bu. Median Cy does were identical (119mg/kg) for patients receiving BuCy regardless of whether the donor was an HLA-identical sibling or unrelated. The median and IQR Bu dose was 15.7 mg/kg (IQR: 14 – 16 mg/kg) for patients receiving oral Bu and 12 mg/kg (IQR: 10 – 13 mg/kg) for those receiving IV Bu.

Neutrophil recovery at 28 days was similar among the groups, but platelet recovery at 28 days occurred in a higher proportion of oral (75%, 95% CI: 71-80%) or IV Bu (77%, 95% CI: 68-85%) patients than those receiving TBI (64%, 95% CI: 58-70%, P=0.009; Table 2). The incidences of hepatic veno-occlusive disease and interstitial pneumonia at 100 days, and NRM, LFS and OS at 5 years did not differ significantly among the three groups. (Table 2)

The incidence of relapse (hematologic, cytogenetic or molecular) at 5 years was 17% (95% CI: 12-23%) for TBI, 17% (95% CI: 12-22%) for oral Bu and 7% (95% CI: 2-14%) for IV Bu (P=0.014). Univariate analyses of specific clinical outcomes and covariates are summarized in supplemental Table S1. Thirty eight patients (4 for no hematopoietic recovery, 5 for graft failure, 7 for relapse, and 22 for whom the indication was missing) underwent either a second HCT (n=11) or donor lymphocyte infusion (n=27). Of these 38 patients, 26 are still alive.

## Multivariate Analysis

Relapse occurred significantly less frequently among patients receiving IV Bu compared to TBI (RR=0.36, 95% CI: 0.15 – 0.86; P=0.022) or oral Bu (RR=0.39, 0.17 – 0.90; P=0.028). NRM and OS were similar among the groups (Table 3).

The interaction term between donor relation (HLA-identical sibling, URD) and the main effect variable (oral Bu vs. IV Bu vs. TBI) was significant for acute GVHD ≥ Grade 3, chronic GVHD and LFS. The results for each donor relation are presented separately based on the multivariate models that included donor relation as a covariate as well as the significant interaction term. Among patients receiving grafts from HLA-identical siblings, the incidences of acute GvHD ≥ Grade 3 and chronic GvHD were similar for all three groups. For patients with URD, however, compared to TBI the incidence of acute GvHD ≥ Grade 3 was higher in those receiving oral (RR=1.76, 95% CI: 1.02 – 3.04; P=0.043) or IV

Bu (RR=2.62, 95% CI: 1.34-5.12; P=0.005). The incidence of chronic GvHD among patients with unrelated donors was higher for those receiving oral (RR=2.73, 95% CI: 1.82-4.10; P<0.0001), but not IV, Bu compared to TBI.

LFS was significantly better among recipients of HLA-identical sibling (Figure 1), but not URD, grafts receiving oral (RR=0.64, 95% CI: 0.44-0.92; P=0.017) or IV Bu (RR=0.53, 95% CI: 0.31-0.92; P=0.025) compared to TBI. In order to determine whether administration of higher radiation doses might contribute to inferior outcomes with TBI, the TBI cohort was divided into those receiving standard (<12.5 Gy) and high dose TBI in a separate multivariate analysis (Table 4). IV Bu remained associated with lower relapse (RR=0.38, 95% CI: 0.16-0.94; P=0.037) and, among recipients of related grafts, better LFS (RR=0.55, 95% CI: 0.31-0.98; P=0.044) than standard dose TBI.

LFS was worse for recipients of URD grafts who received oral, but not IV, Bu compared to TBI (RR=1.69, 95% CI: 1.11-2.56; P=0.014).The use of a TKI prior to HCT was not adversely associated with any of the reported outcomes and was associated with better LFS. (RR=0.69, 95% CI: 0.53-0.92; P=0.01)

## Discussion

Although transplantation is no longer first-line treatment for CML in first chronic phase many patients who are resistant to, or intolerant of, tyrosine kinase inhibitors continue to undergo the procedure.[24,25,26] CIBMTR registration retrieval for the United States alone identified 120 allogeneic transplants, of whom 113 recipients were ≥18 years of age, for CML in first chronic phase in 2012 and 2013 (Wael Saber, personal communication). Our retrospective analysis shows that, in patients with chronic phase CML, relapse occurred significantly less often among those who received IV Bu compared to oral Bu or TBI, regardless of whether patients received standard or high doses of TBI. LFS was better among those receiving HLA-identical sibling grafts who received IV or oral Bu compared to TBI including when the analysis was limited to those patients who received standard TBI doses.

OS, however, did not differ among the groups. The effectiveness of TKIs and donor lymphocyte infusions in extending survival following relapse in CML[27], and the limited (5 years) follow up of the present study probably account for similar OS despite the differences in relapse and LFS. Nevertheless, cure is the ultimate goal of HCT in CML. The lower incidence of relapse and better LFS with IV Bu (compared to TBI) support its use in patients receiving HLA-identical sibling grafts. LFS following HLA-identical sibling grafts was also superior to TBI in patients receiving oral Bu. It is likely that PK dosing was widely utilized among these patients. [18] The oral formulation has been largely displaced by the IV formulation,[28] but PK-based oral dosing might yield similar results. This was not specifically addressed in the present study. Important differences in LFS between IV Bu and TBI were not identified for recipients of URD grafts with the available sample size in this study.

In AML, studies of Cy combined with fixed dose oral Bu have reported some disadvantages, including higher relapse rates, compared to TBI. [29,30] In contrast, compared to TBI, IV Bu was associated with lower relapse rates beyond 1 year, less NRM and better LFS and survival in patients with AML in CR.[18] It would seem that the advantage of IV Bu might be magnified in CML where oral fixed-dose Bu showed advantages over TBI, including less toxicity[6] and relapse[7], and favorable results were reported with dose-adjusted oral Bu. [14] Notably, NRM was comparatively low in the present study in the IV Bu group relative to the TBI cohort at 1 and 3 years, but similar at 5 years. A higher proportion of patients receiving IV Bu who were alive at 3 years subsequently died from NRM compared to those receiving TBI, but no clear pattern in the cause of death emerged. In particular, only one death attributed to chronic GvHD occurred beyond three years in the IV Bu cohort (data not shown). We also have no precise explanation for the higher incidence of acute GvHD ≥ Grade III with Bu compared with TBI in recipients of URD grafts, however, TBI patients were more likely to have received marrow and ATG or alemtuzumab. These data contrast with reports of less GvHD with Bu in CML [6] and AML, [17] although those studies were performed with HLA identical sibling donors. Importantly, the use of a TKI before HCT did not adversely influence the outcomes reported and was associated with better LFS. These results support a previous report that imatinib use before HCT did not adversely influence transplant outcomes.[31] The significantly lower incidence of relapse with IV Bu and better LFS with IV (or oral) Bu compared to TBI in recipients of HLA-identical related grafts were present regardless of whether TKI use was considered in the multivariate model.

There are, of course, limitations to this retrospective analysis. First, it is not known why individual patients received specific preparative regimens. Second, there were important differences between the groups, especially in the distribution of related and unrelated donors, TKI exposure, and year when transplantation was performed. Multivariate analyses were performed to account for these differences, but the relatively few patients in the IV Bu arm, particularly those receiving grafts from URD, limits the effectiveness of that approach. In addition, over the course of the study, the use of molecular detection of relapse became more widespread. The application of more sensitive techniques to detect relapse, however, would likely result in earlier detection in patients undergoing transplantation in the later years of study, potentially leading to an underestimation of the difference in relapse with IV Bu. Also, data were not collected for PK studies and dose adjustment, which is reported to affect outcomes with IV [32] as well as oral Bu. [33] We were therefore unable to analyze the potential benefit of PK-directed dosing in patients receiving oral or IV Bu.

Absent results of a randomized trial, the association of IV Bu with lower relapses rates in first chronic phase CML patients, and better LFS compared to TBI among recipients of HLA-identical sibling grafts favors its use in that setting.

## Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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## References

1. Clift RA, Buckner CD, Thomas ED, et al. Treatment of chronic granulocytic leukemia in chronic phase by allogeneic marrow transplantation. Lancet. Sep; 1982 2(8299):621–623. [PubMed: 6125772]
2. Goldman JM, Baughan AS, McCarthy DM, et al. Marrow transplantation for patients in the chronic phase of chronic granulocytic leukemia. Lancet. Sep; 1982 2(8299):623–625. [PubMed: 6125773]
3. Goldman JM, Apperley JF, Jones L, et al. Bone marrow transplantation for patients with chronic myeloid leukemia. N Engl J Med. Jan; 1986 314(4):202–207. [PubMed: 3510388]
4. Thomas ED, Clift RA, Fefer A, et al. Marrow transplantation for the treatment of chronic myelogenous leukemia. Ann Intern Med. Feb; 1986 104(2):155–163. [PubMed: 3511810]
5. Biggs JC, Szer J, Crilley P, et al. Treatment of chronic myeloid leukemia with allogeneic bone marrow transplantation after preparation with BuCy 2. Blood. Sep; 1992 80(5):1352–1357. [PubMed: 1515648]
6. Clift RA, Buckner CD, Thomas ED, et al. Marrow transplantation for chronic myeloid leukemia: a randomized study comparing cyclophosphamide and total body irradiation with busulfan and cyclophosphamide. Blood. Sep; 1994 84(6):2036–2043. [PubMed: 8081005]
7. Devergie A, Blaise D, Attal M, et al. Allogeneic bone marrow transplantation for chronic myeloid leukemia in first chronic phase: a randomized trial of busulfan-cytoxan versus cytoxan-total body irradiation as preparative regimen: a report from the French Society of Bone Marrow Graft (SFGM). Blood. Apr; 1995 85(8):2263–2268. [PubMed: 7718899]
8. Ehrsson H, Hassan M, Ehrnebo M, et al. Busulfan kinetics. Clin Pharmacol Ther. Jul; 1983 34(1): 86–9. [PubMed: 6574831]
9. McCune JS, Baker KS, Blough DK, et al. Variation in prescribing patterns and therapeutic drug monitoring of intravenous busulfan in pediatric hematopoietic cell transplant recipients. J Clin Pharmacol. Mar; 2013 53(3):264–75. [PubMed: 23444282]
10. Grochow LB, Jones RJ, Brundrett RB, et al. Pharmacokinetics of busulfan: correlation with veno-occlusive disease in patients undergoing bone marrow transplantation. Cancer Chemother Pharmacol. 1989; 25(1):55–61. [PubMed: 2591002]
11. Slattery JT, Sanders JE, Buckner CD, et al. Graft-rejection and toxicity following bone marrow transplantation in relation to busulfan pharmacokinetics. Bone Marrow Transplant. Jul; 1995 16(1):31–42. [PubMed: 7581127]
12. Slattery JT, Clift RA, Buckner CD, et al. Marrow transplantation for chronic myeloid leukemia: the influence of plasma busulfan levels on the outcome of transplantation. Blood. Apr; 1997 89(8): 3055–3060. [PubMed: 9108427]
13. Buffery PJ, Allen KM, Chin PK, et al. Thirteen years' experience of pharmacokinetic monitoring and dosing of busulfan: can the strategy be improved? Ther Drug Monit. Feb; 2014 36(1):86–92. [PubMed: 24299921]

14. Radich JP, Gooley T, Bensinger W, et al. HLA-matched related hematopoietic cell transplantation for chronic-phase CML using a targeted busulfan and cyclophosphamide preparative regimen. Blood. Jul; 2003 102(1):31–35. [PubMed: 12595317]
15. Andersson BS, Kashyap A, Gian V, et al. Conditioning therapy with intravenous busulfan and cyclophosphamide (IV BuCy2) for hematologic malignancies prior to allogeneic stem cell transplantation: a phase II study. Biol Blood Marrow Transplant. 2002; 8(3):145–154. [PubMed: 11939604]
16. Kashyap A, Wingard J, Cagnoni P, et al. Intravenous versus oral busulfan as part of a busulfan/cyclophosphamide preparative regimen for allogeneic hematopoietic stem cell transplantation: decreased incidence of hepatic venoocclusive disease (HVOD), HVOD-related mortality, and overall 100-day mortality. Biol Blood Marrow Transplant. 2002; 8(9):493–500. [PubMed: 12374454]
17. Nagler A, Rocha V, Labopin M, et al. Allogeneic hematopoietic stem-cell transplantation for acute myeloid leukemia in remission: comparison of intravenous busulfan plus cyclophosphamide (Cy) versus total-body irradiation plus Cy as conditioning regimen--a report from the acute leukemia working party of the European group for blood and marrow transplantation. J Clin Oncol. Oct; 2013 31(28):3549–3556. [PubMed: 23980086]
18. Copelan EA, Hamilton BK, Avalos B, et al. Better leukemia-free and overall survival in AML in first remission following cyclophosphamide in combination with busulfan compared to TBI. Blood. Dec; 2013 122(24):3863–3870. [PubMed: 24065243]
19. Bredeson C, Leradematcher J, Kato K, et al. Prospective cohort study comparing intravenous busulfan to total body irradiation in hematopoietic cell transplantation. Blood. Dec; 2013 122(24): 3871–3878. [PubMed: 24081656]
20. Weisdorf D, Spellman S, Haagenson M, et al. Classification of HLA-matching for retrospective analysis of unrelated donor transplantation: revised definitions to predict survival. Biol Blood Marrow Transplant. Jul; 2008 14(7):748–58. [PubMed: 18541193]
21. Przepiorka D, Weisdorf D, Martin P, et al. 1994 Consensus Conference on Acute GVHD Grading. Bone Marrow Transplant. Jun; 1995 15(6):825–828. [PubMed: 7581076]
22. Shulman HM, Sullivan KM, Weiden PL, et al. Chronic graft-versus-host syndrome in man. A long-term clinicopathologic study of 20 Seattle patients. Am J Med. Aug; 1980 69(2):204–217. [PubMed: 6996481]
23. Zhang X, Loberiza FR, Klein JP, et al. A SAS macro for estimation of direct adjusted survival curves based on a stratified Cox regression model. Comput Methods Programs Biomed. Nov; 2007 88(2):95–101. [PubMed: 17850917]
24. Passweg JR, Baldomero H, Peters C, et al. Hematopoietic SCT in Europe: data and trends in 2012 with special consideration of pediatric transplantation. Bone Marrow Transplant. Jun; 2014 49(6): 744–50. [PubMed: 24637898]
25. Gratwohl A, Baldomero H, Gratwohl M, et al. Quantitative and qualitative differences in use and trends of hematopoietic stem cell transplantation: a Global Observational Study. Haematologica. Aug; 2013 98(8):1282–90. [PubMed: 23508009]
26. Baccarani M, Deininger MW, Rosti G, et al. European LeukemiaNet recommendations for the management of chronic myeloid leukemia: 2013. Blood. Aug; 2013 122(6):872–84. [PubMed: 23803709]
27. Martínez C, Gomez V, Tomás JF, et al. Relapse of chronic myeloid leukemia after allogeneic stem cell transplantation: outcome and prognostic factors: the Chronic Myeloid Leukemia Subcommittee of the GETH (Grupo Español de Trasplante Hemopoyético). Bone Marrow Transplant. Aug; 2005 36(4):301–306. [PubMed: 15968278]
28. Champlin RE. Busulfan or TBI: answer to an age-old question. Blood. Dec; 2013 122(24):3856–3857. [PubMed: 24311716]
29. Blaise D, Maraninchi D, Archimbaud E, et al. Allogeneic bone marrow transplantation for acute myeloid leukemia in first remission: a randomized trial of a busulfan-Cytoxan versus Cytoxan-total body irradiation as preparative regimen: a report from the Group d'Etudes de la Greffe de Moelle Osseuse. Blood. May; 1992 79(10):2578–2582. [PubMed: 1586710]

30. Gupta V, Lazarus HM, Keating A. Myeloablative conditioning regimens for AML allografts: 30 years later. Bone Marrow Transplant. Nov; 2003 32(10):969–978. [PubMed: 14595384]

31. Lee SJ, Kukreja M, Wang T, et al. Impact of prior imatinib mesylate on the outcome of hematopoietic cell transplantation for chronic myeloid leukemia. Blood. Oct; 2008 112(8):3500–3507. [PubMed: 18664621]

32. Pidala J, Kim J, Anasetti C, et al. Pharmacokinetic targeting of intravenous busulfan reduces conditioning regimen related toxicity following allogeneic hematopoietic cell transplantation for acute myelogenous leukemia. J Hematol Oncol.

33. McCune JS, Gibbs JP, Slattery JT. Plasma concentration monitoring of busulfan: does it improve clinical outcome? Clin Pharmacokinet. Aug; 2000 39(2):155–165. [PubMed: 10976660]

## Highlights

- IV busulfan is associated with lower relapse rates than oral busulfan or TBI following myeloablative HCT for CML.
- IV and oral busulfan are associated with better LFS than TBI in patients receiving grafts from HLA-identical siblings.
- Use of a TKI prior to transplant was associated with better LFS.

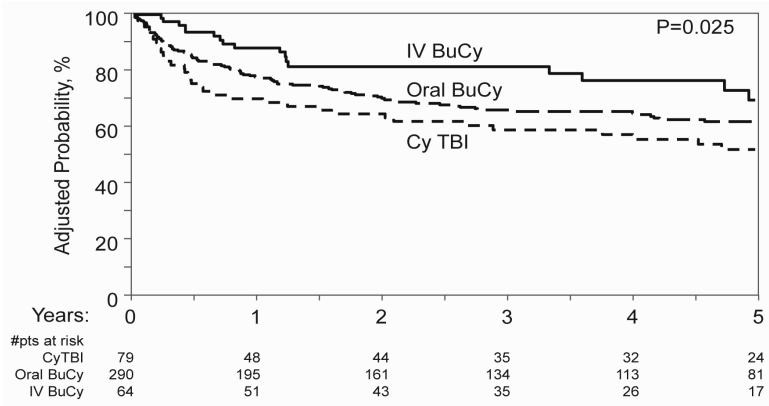


**Figure 1.**

Adjusted probabilities of LFS according to preparative regimen for recipients of grafts from HLA-identical sibling (adjusted covariates: TKI use before HCT, recipient age).

## Table 1

Characteristics of patients

| Characteristics of patients | Cy TBI | Oral BuCy | IV BuCy | Overall p-value |
|---|---|---|---|---|
| Number of patients | 222 | 354 | 97 | |
| Number of centers | 75 | 68 | 47 | |
| Patient-Related | | | | |
| Age, median (range), years | 35 (18-59) | 34 (18-59) | 39 (18-61) | 0.018 |
| 18-30 | 73 (33) | 136 (38) | 28 (29) | 0.167 |
| 31-40 | 80 (36) | 116 (33) | 28 (29) | |
| 41-50 | 47 (21) | 75 (21) | 27 (28) | |
| >50 | 22 (10) | 27 ( 8) | 14 (14) | |
| Sex | | | | 0.293 |
| Male | 145 (65) | 211 (60) | 56 (58) | |
| Karnofsky performance score at transplant | | | | 0.142 |
| <90% | 19 ( 9) | 21 ( 6) | 11 (11) | |
| >=90% | 193 (87) | 326 (92) | 83 (86) | |
| Missing | 10 ( 5) | 7 (2) | 3 (3) | |
| Disease-Related | | | | |
| Time from diagnosis to transplant, median (range), months | 10 (3 - 79) | 9 (2 - 96) | 10 (2 - 149) | 0.813 |
| TKI use pre transplant | | | | <0.001 |
| No | 143 (64) | 258 (73) | 32 (33) | |
| Yes | 79 (36) | 96 (27) | 65 (67) | |
| Transplant-Related | | | | |
| Donor-recipient sex match | | | | 0.503 |
| M-M | 102 (46) | 139 (39) | 37 (38) | |
| F-M | 43 (19) | 72 (20) | 19 (20) | |
| M-F | 44 (20) | 70 (20) | 24 (25) | |
| F-F | 33 (15) | 73 (21) | 17 (18) | |
| Donor Relation | | | | <0.001 |
| HLA-identical sibling | 80 (36) | 293 (83) | 65 (67) | |
| Well-matched URD | 142 (64) | 61 (17) | 32 (33) | |
| Donor-recipient CMV serological status | | | | <0.001 |
| +/+ | 75 (34) | 74 (21) | 26 (27) | |
| +/− | 56 (25) | 197 (56) | 35 (36) | |
| −/+ | 36 (16) | 26 ( 7) | 13 (13) | |
| −/− | 49 (22) | 45 (13) | 17 (18) | |
| Missing | 6 (3) | 12 ( 3) | 6 (6) | |
| HLA-iden sibling donor age, median (range), years | 37 (13 - 70) | 34 (3 - 65) | 40 (20 - 61) | <0.001 |
| Unrelated donor age, median (range), years | 35 (19 - 61) | 33 (20 - 46) | 38 (21 - 51) | 0.063 |

| Characteristics of patients | Cy TBI | Oral BuCy | IV BuCy | Overall p-value |
|---|---|---|---|---|
| Graft type | | | | <0.001 |
| Bone Marrow | 146 (66) | 168 (47) | 49 (51) | |
| Peripheral blood | 76 (34) | 186 (53) | 48 (49) | |
| Conditioning regimen | | | | |
| Bu dose, median (range), mg/kg | -- | 16 (10 - 25) | 12 (9-17) | -- |
| Cy/TBI (nonfrac 550-750) | 9 ( 4) | 0 | 0 | |
| Cy/TBI (nonfrac 800-1200) | 4 ( 2) | 0 | 0 | |
| Cy/TBI (frac 900-1170) | 13 ( 6) | 0 | 0 | |
| Cy/TBI (frac 1200-1300) | 123 (55) | 0 | 0 | |
| Cy/TBI (frac 1320-1395) | 40 (18) | 0 | 0 | |
| Cy/TBI (frac 1400-1500) | 33 (15) | 0 | 0 | |
| ATG or alemtuzumab use | | | | 0.004 |
| Yes | 41 (18) | 32 (9) | 13 (13) | |
| No | 181 (82) | 322 (91) | 84 (87) | |
| GVHD prophylaxis * | | | | <0.001 |
| TAC + MMF +− others | 6 (3) | 1 (<1) | 4 (4) | |
| TAC + MTX +− others (except MMF) | 49 (22) | 23 (6) | 44 (45) | |
| TAC + others (except MTX, MMF) | 6 (3) | 2 (<1) | 0 | |
| TAC alone | 2 (<1) | 0 | 1 (1) | |
| CSA + MMF +− others (except TAC) | 6 (3) | 4 ( 1) | 2 (2) | |
| CSA + MTX +− others (except TAC, MMF) | 140 (63) | 307 (87) | 40 (41) | |
| CSA + others (except TAC, MTX, MMF) | 5 (2) | 3 (<1) | 1 (1) | |
| CSA alone | 6 (3) | 4 (1) | 1 (1) | |
| Other | 0 | 2 (<1) | 3 (3) | |
| Missing | 2 (<1) | 8 (2) | 1 (1) | |
| Growth factors given post transplant | | | | 0.299 |
| No | 172 (77) | 253 (71) | 72 (74) | |
| Yes | 49 (22) | 101 (29) | 25 (26) | |
| Missing | 1 (<1) | 0 | 0 | |
| Year of transplant | | | | <0.001 |
| 2000 | 81 (36) | 93 (26) | 9 (9) | |
| 2001 | 45 (20) | 44 (12) | 8 (8) | |
| 2002 | 28 (13) | 59 (17) | 5 (5) | |
| 2003 | 15 (7) | 26 (7) | 10 (10) | |
| 2004 | 14 ( 6) | 43 (12) | 28 (29) | |
| 2005 | 21 (9) | 54 (15) | 19 (20) | |
| 2006 | 18 (8) | 35 (10) | 18 (19) | |
| Median follow-up of survivors, range, months | 72 (2-127) | 56 (2-129) | 59 (3-119) | |

*Cy indicates Cyclophosphamide; TBI, total body irradiation; Bu, busulfan; ATG, antithymocyte globulin; GvHD, graft versus host disease; TAG, tacrolimus; MMF, mycophenolate mofetil; MTX, methotrexate; GSA, cyclosporine, TKI, tyrosine kinase inhibitor

**Table 2**

Univariate analysis

| Outcomes | Cy TBI Probability (95% CI) | Oral BuCy Probability (95% CI) | IV BuCy Probability (95% CI) | Overall p-values |
|---|---|---|---|---|
| Neutrophil recovery | | | | |
| NEval | 222 | 354 | 97 | |
| @ 28 days | 92 (88-95) | 91 (88-94) | 95 (90-98) | 0.391 |
| Platelet recovery | | | | |
| NEval | 221 | 346 | 96 | |
| @ 28 days | 64 (58-70) | 75 (71-80) | 77 (68-85) | 0.009 |
| @ 100 days | 90 (85-93) | 93 (90-95) | 96 (91-99) | 0.103 |
| Acute GVHD (II-IV) | | | | |
| NEval | 222 | 352 | 97 | |
| @ 100 days | 56 (50-63) | 43 (39-49) | 46 (37-56) | 0.014 |
| Acute GVHD (III-IV) | | | | |
| NEval | 222 | 354 | 97 | |
| @ 100 days | 24 (19-30) | 20 (16-24) | 26 (18-35) | 0.290 |
| Hepatic Veno-occlusive Disease | | | | |
| NEval | 222 | 354 | 97 | |
| @ 100 days | 5 (2-9) | 9 (7-13) | 6 (2-12) | 0.17 |
| Interstitial Pneumonia | | | | |
| NEval | 222 | 354 | 97 | |
| @ 100 days | 9 (5-13) | 5 (3-8) | 4 (1-9) | 0.22 |
| Chronic GVHD | | | | |
| NEval | 216 | 348 | 95 | |
| @ 5 years | 55 (48-61) | 62 (57-68) | 67 (57-76) | 0.082 |
| Non-relapse mortality | | | | |
| NEval | 216 | 350 | 95 | |
| @ 1 year | 25 (20-31) | 20 (16-24) | 16 (9-24) | 0.142 |
| @ 3 years | 31 (25-38) | 24 (20-29) | 22 (14-31) | 0.139 |
| @ 5 years | 31 (25-38) | 25 (21-30) | 36 (25-48) | 0.119 |
| Relapse | | | | |
| NEval | 216 | 350 | 95 | |
| @ 5 years | 17 (12-23) | 17 (12-22) | 7 (2-14) | 0.014 |
| Leukemia free survival | | | | |
| NEval | 216 | 350 | 95 | 0.102 |
| @ 1 year | 67 (60-73) | 74 (69-78) | 80 (72-88) | 0.031 |
| @ 3 years | 55 (48-62) | 62 (57-68) | 74 (64-82) | 0.006 |
| @ 5 years | 52 (45-59) | 58 (52-64) | 57 (45-69) | 0.384 |
| Overall survival | | | | |
| NEval | 222 | 354 | 97 | 0.196 |
| @ 1 year | 74 (68-80) | 80 (75-84) | 84 (76-91) | 0.118 |

| Outcomes | Cy TBI Probability (95% CI) | Oral BuCy Probability (95% CI) | IV BuCy Probability (95% CI) | Overall p-values |
|---|---|---|---|---|
| @ 3 years | 67 (60-73) | 74 (69-78) | 77 (68-85) | 0.097 |
| @ 5 years | 66 (59-72) | 72 (67-77) | 61 (50-73) | 0.152 |

N Eval indicates number evaluable; Cy, cyclophosphamide; TBI, total body irradiation; IV, intravenous; Bu, busulfan, GvHD, graft versus host disease

## Table 3

Relative risks and 95% confidence intervals of a multivariate analysis

| | Oral BuCy vs. TBI RR (95% CI) | IV BuCy vs. TBI RR (95% CI) | Oral Bucy vs. IV BuCy RR (95% CI) | Overall *p* |
|---|---|---|---|---|
| aGVHD II-IV[a] | 0.87 (0.66, 1.13) | 0.81 (0.57, 1.14) | 1.07 (0.77, 1.49) | 0.405 |
| aGVHD III-IV for HLA sibs[b] | 0.71 (0.42, 1.18) | 0.89 (0.44, 1.81) | 0.79 (0.43, 1.47) | 0.37 |
| aGVHD III-IV for URD[b] | **1.76 (1.02, 3.04)** | **2.62 (1.34, 5.12)** | 0.67 (0.33, 1.36) | 0.01 |
| cGVHD for HLA sibs[c] | 1.22 (0.85, 1.74) | 1.31 (0.83, 2.05) | 0.93 (0.65, 1.33) | 0.47 |
| cGVHD for URD[c] | **2.73 (1.82, 4.10)** | 1.52 (0.94, 2.47) | **1.79 (1.04, 3.08)** | <.0001 |
| LFS for HLA sibs[d] | **0.64 (0.44, 0.92)** | **0.53 (0.31, 0.92)** | 1.19 (0.72, 1.98) | 0.025 |
| LFS for URD[d] | **1.69 (1.11, 2.56)** | 1.32 (0.75, 2.32) | 1.28 (0.71, 2.32) | 0.046 |
| Relapse[e] | 0.94 (0.61, 1.45) | **0.36 (0.15, 0.86)** | **2.58 (1.11, 6.03)** | 0.067 |
| NRM[f] | 1.17 (0.81, 1.68) | 1.26 (0.78, 2.03) | 0.93 (0.60, 1.45) | 0.576 |
| OS[g] | 1.15 (0.81, 1.62) | 1.19 (0.75, 1.88) | 0.97 (0.63, 1.48) | 0.679 |

* Bolded indicates p-value<0.05; aGVHD, acute GVHD; cGVHD, chronic GVHD; NRM, non-relapse mortality; LFS, leukemia-free survival; OS, overall survival

Other Significant factors in the multivariate model include:

[a] Graft type, Donor relation

[b] Donor relation, year of transplant

[c] Donor relation, Sex match, graft type, ATG or alemtuzumab use

[d] Donor relation, recipient age, TKI

[e] None

[f] Donor relation, recipient age, graft type, year of transplant

[g] Year of transplant, recipient age, donor relation

## Table 4

Relative risks and 95% confidence intervals of a multivariate analysis (TBI dose divided into standard and high dose)

| | **Oral BuCy vs. TBI (≤ 1250) RR (95% CI)** | **IV BuCy vs. TBI (≤ 1250) RR (95% CI)** | **TBI (>1250) vs. TBI(<1250) RR (95% CI)** | **Oral Bucy vs. IV BuCy RR (95% CI)** | **Oral BuCy vs. TBI (> 1250) RR (95% CI)** | **IV BuCy vs. TBI (> 1250) RR (95% CI)** | **Overall *p*** |
|---|---|---|---|---|---|---|---|
| aGVHD II-IV[a] | 0.82(0.61, 1.09) | 0.76 (0.53, 1.1) | 0.84 (0.58, 1.22) | 1.07 (0.77, 1.5) | 0.98 (0.67, 1.42) | 0.91 (0.59, 1.41) | 0.434 |
| aGVHD III-IV for HLA sibs[b] | 0.87 (0.47, 1.6) | 1.10 (0.5, 2.41) | 1.96 (0.81, 4.72) | 0.79 (0.43, 1.46) | 0.44 (0.21, 0.94) | 0.56 (0.23, 1.37) | 0.193 |
| aGVHD III-IV for URD[b] | **2.04 (1.09, 3.8)** | **3.05 (1.46, 6.35)** | 1.44 (0.73, 2.83) | 0.67 (0.33, 1.36) | 1.42 (0.73, 2.75) | 2.12 (0.98, 4.57) | 0.018 |
| cGVHD for HLA sibs[c] | 1.17 (0.78, 1.76) | 1.26 (0.77, 2.06) | 0.87 (0.41, 1.84) | 0.93 (0.65, 1.32) | 1.36 (0.69, 2.66) | 1.46 (0.71, 3.02) | 0.643 |
| cGVHD for URD[c] | **2.07 (1.35, 3.18)** | 1.14 (0.69, 1.89) | **0.46 (0.28, 0.76)** | **1.81 (1.06, 3.12)** | **4.52** (2.6, **7.83**) | **2.49 (1.36, 4.55)** | <.0001 |
| LFS for HLA sibs[d] | **0.66 (0.43, 0.99)** | **0.55 (0.31, 0.98)** | 1.1 (0.56, 2.16) | 1.19 (0.72, 1.97) | 0.6 (0.32, 1.09) | 0.5 (0.24, 1.03) | 0.059 |
| LFS for URD[d] | **1.96 (1.23, 3.13)** | 1.53 (0.84, 2.8) | 1.48 (0.89, 2.45) | 1.28 (0.71, 2.31) | 1.33 (0.8, 2.21) | 1.04 (0.55, 1.96) | 0.046 |
| Relapse[e] | 0.99 (0.6, 1.63) | **0.38 (0.16, 0.94)** | 1.17 (0.58, 2.36) | **2.58 (1.11, 6.03)** | 0.84 (0.45, 1.58) | **0.33 (0.12, 0.87)** | 0.131 |
| NRM[f] | 1.27 (0.84, 1.9) | 1.36 (0.82, 2.26) | 1.26 (0.77, 2.06) | 0.93 (0.6, 1.45) | 1 (0.62, 1.62) | 1.08 (0.61, 1.91) | 0.595 |
| OS[g] | 1.26 (0.86, 1.85) | 1.3 (0.8, 2.12) | 1.31 (0.83, 2.09) | 0.97 (0.63, 1.48) | 0.96 (0.61, 1.51) | 0.99 (0.57, 1.71) | 0.568 |

*Bolded indicates p-value<0.05; aGVHD, acute GVHD; cGVHD, chronic GVHD; NRM, non-relapse mortality; LFS, leukemia-free survival; OS, overall survival

Other Significant factors in the multivariate model include:

[a] Graft type, Donor relation

[b] Donor relation, year of transplant

[c] Donor relation, Sex match, graft type, ATG or alemtuzumab use

[d] Donor relation, recipient age, TKI

[e] None

[f] Donor relation, recipient age, graft type, year of transplant

[g] Year of transplant, recipient age, donor relation