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Role of allogeneic transplantation for FLT3/ITD acute myeloid leukemia: Outcomes from 133 consecutive newly-diagnosed patients from a single institution

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Abstract

AML patients with FLT3/ITD mutations have an inferior survival compared to AML patients with wild-type (WT) FLT3, primarily due to an increased relapse rate. Allogeneic transplant represents a post-remission therapy that is effective at reducing the risk of relapse for many cases of poor-risk AML. Whether or not allogeneic transplant in first complete remission (CR) can improve outcomes for patients with FLT3/ITD AML remains controversial. Our institution has adopted a policy of pursuing allogeneic transplant, including the use of alternate donors, for FLT3/ITD AML patients in remission. As part of an IRB-approved study, we performed a review of the clinical data from November 1, 2004 to October 31, 2008 on all adult patients under the age of 60 presenting in consecutive fashion to the Sidney Kimmel Comprehensive Cancer Center at Johns Hopkins with newly diagnosed non-M3 AML. We followed their outcomes through August 1, 2010. During the study period, 133 previously untreated AML patients between the ages of 20 and 59 were diagnosed and received induction and consolidation therapy at our institution. Of these 133 patients, 31 (23%) harbored a FLT3/ITD mutation at diagnosis. The median OS (overall survival) from the time of diagnosis for the FLT3/ITD AML patients was compared to the OS of the entire cohort and found to be comparable (19.3 months versus 15.5 months p=0.56.) Historically, OS for FLT3/ITD AML patients is significantly worse than for AML patients lacking this mutation. However, the OS for the 31 FLT3/ITD patients reported here was comparable to the 102 patients with WT FLT3 over the same 4 year time period. One difference that might have contributed to the surprising outcomes for the FLT3/ITD group is our aggressive pursuit of allogeneic BMT in CR1 within this group (60% of FLT3/ITD vs. 17% with WT). Our single institution study of consecutively treated AML patients supports the hypothesis that allogeneic transplant in early CR1 improves the long term outcomes for FLT3/ITD AML.

Keywords

Stem Cell transplantation; FLT3/ITD

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Introduction

In recent decades, survival in younger patients (age < 60 years) with acute myeloid leukemia (AML) has improved, largely due to intensification of post-remission therapies and advanced supportive care of critically ill patients. The rate of complete remission after initial induction therapy (CR1) now approaches 80% [1]. However, many of these patients will eventually relapse, and die from their AML.

Attention has recently focused on determining the post remission therapies most likely to decrease rates of relapse. Following successful allogeneic hematopoietic stem cell transplantation (HSCT)[1], the relapse rate is significantly reduced. However, the use of HSCT is limited by treatment-related morbidity and mortality. Continuing studies of HSCT are required to determine which patients will most benefit from this therapy with its associated morbidity and mortality.

The choice of HSCT as post remission therapy is guided by prognostic indicators [2]. Cytogenetic risk has been widely used to explore the efficacy of HSCT for patients with AML in CR1. In adults with AML, the karyotype at the time of diagnosis is the most widely used prognostic indicator [3]. For those patients with unfavorable-risk cytogenetics, the beneficial effect of allogeneic HSCT has been demonstrated in a large meta-analysis [4]. However, up to 50% of patients do not have clonal chromosomal aberrations [5] usable for this prognostication. Therefore there is a need to determine other prognostic markers beyond conventional cytogenetics. FLT3/ITD is such a prognostic marker.

In 1996 it was reported that internal tandem duplication (ITD) of base pairs of the FMS-Like-Tyrosine kinase-3 (FLT3) could result in the constitutive activation of the gene in AML patients. [6] These mutations in FLT3 are found in about 30% of cases of acute myeloid leukemia, and confer an increased relapse rate and reduced overall survival [7-10].

To further investigate if this prognostic marker could be used to guide the decision to move towards earlier HSCT, our institution has adopted a policy of pursuing allogeneic HSCT for FLT3/ITD AML patients in CR1. Here we present the data from 31 FLT3/ITD patients age 18-59.9 years,and compare the outcomes for patients receiving allogeneic HSCT in CR1 with that for patients who received chemotherapy alone.

Materials and Methods

Data sources

We reviewed the clinical databases at the Sidney Kimmel Comprehensive Cancer Center (SKCCC) at Johns Hopkins Hospital in Baltimore. Our observational study was carried out with a waiver of informed consent, in accordance with the Declaration of Helsinki, and in compliance with the Health Insurance Portability Accountability Act regulations as determined by the Institutional Review Board of the Johns Hopkins Hospital.

Study population

The study population consisted of all patients with non-M3 AML, ages 18-60, presenting consecutively to the SKCCC from November 1, 2004 to October 31, 2008. We followed the outcomes of these 133 patients through August 1, 2010. The cohorts consisted of patients separated by FLT3/ITD mutational status, cytogenetics, and treatments applied.

Diagnosis of FLT3/ITD mutants

All patients had the status of their FLT3 internal tandem duplication mutation determined by the Clinical Laboratory Improvement Amendments (CLIA) certified test at the SKCCC.

This assay identifies internal FLT3 tandem duplication mutations via a single multiplex DNA polymerase chain reaction. After amplification, the polymerase chain reaction products are analyzed by capillary electrophoresis for length mutations and resistance to *Eco*RV digestion.[11] Each patient in the cohort had this test performed at presentation, and the results were clinically available to guide therapies.

Cytogenetics

Unfavorable risk cytogenetics were defined according to the Southwest Oncology Group/ Eastern Cooperative Oncology Group (SWOG/ECOG) classification[12] and included: del(5q)/−5, −7/del(7q), abnormality 3q, 9q, 11q, 20q, 21q, 17p, t(6;9), t(9;22), and complex cytogenetics (≥3 unrelated abnormalities). Cytogenetics results were reviewed as provided by the genetics laboratory. Abnormalities were further classified as either complex cytogenetics (≥3 unrelated abnormalities), or normal (46 XX or 46 XY).

Treatment Schedule

All patients received their induction therapy at the SKCCC. Fitness for intensive induction therapy was determined using Eastern Cooperative Oncology Group (ECOG) performance status at presentation, or by complicated medical comorbidities.

Two intensive induction regimens were employed: 1) an institutional protocol of flavopiridol 50 mg/m^2 given by 1 h infusion daily × 3 days beginning on day 1, followed by 2 g/m^2/72 h cytarabine beginning day 6, and 40 mg/m^2 mitoxantrone on day 9 (FLAM)[13]; or 2) timed sequential therapy (TST)[14] consistent with our institutional standard. This consisted of cytarabine 667 mg/m^2 given by 24-hour continuous infusion daily × 3 and daunorubicin 45 mg/m^2 intravenous push daily × 3 both beginning day 1, followed by etoposide 200 mg/ m^2 intravenous infusion over 3 hours daily on days 8-10 (AcDVP16).[15]

For patients receiving fully matched allogeneic transplants from a sibling or unrelated donor, the preparative regimen for myeloablative HSCT consisted of busulfan at 4 mg/kg/d orally or 3.2 mg/kg/d intravenously given in 4 daily divided doses for 4 consecutive days, followed by cyclophosphamide (Cy) at 50 mg/kg intravenously for 2 consecutive days. The fifth and subsequent doses of busulfan were adjusted according to first-dose pharmacokinetic measurements to achieve a target area under the curve of 800 to 1400 mol/L* min.[16]

Graft versus host disease (GVHD) prophylaxis was the institutional standard of cyclophosphamide 50 mg/kg/d given intravenously on days 3 and 4 after transplantation. [17,18] Mesna (80% of cyclophosphamide dose) was administered in 4 divided doses on all days of cyclophosphamide administration. Tacrolimus was additional GVHD prophylaxis in patients post transplantation.

Patients undergoing non-myeloablative HSCT from a haploidentical donor received a preparative regimen on an institutional protocol. This conditioning consisted of fludarabine, 30 mg/m2 per day from days −6 to −2, and total body irradiation (TBI), 2 Gy on day −1. All patients received Cy, 50 mg/kg on day 3, mycophenolate mofetil from day 4 to day 35, and tacrolimus from day 4 to day ≥ 50.[17]

All dosing of chemotherapeutic agents was based on ideal body weight. Colony-stimulating factors were not given. All supportive care measures were administered according to institutional protocols and included prophylaxis against *Pneumocystis jirovecii, Candida albicans*, and herpes zoster/simplex infections. All blood products except for the allografts were irradiated before transfusion. Cytomegalovirus (CMV)–seronegative patients were given transfusions from CMV-seronegative donors or leukoreduced blood products if CMV products were unavailable. Supportive care measures were identical for all recipients of

allografts and conventional chemotherapy. Nine of the 11 FLT3/ITD patients transplanted in CR1 required an additional cycle of consolidative chemotherapy prior to transplantation due to the time necessary to prepare for the transplant.

Outcome measures

The primary outcome measure was overall survival (OS). Other outcomes analyzed were (1) event-free survival (EFS), defined as the time to relapse or death, (2) relapse rate, and (3) non-relapse mortality, defined as time to death censored at relapse. Kaplan-Meier curves were used, and all treatment comparisons were by intention to treat. These analyses were performed with GraphPad Prism 4.

Results

The cohort included 102 patients with wild type (WT) FLT3 and 31 with FLT3/ITD mutations. The demographics of each cohort are shown in Table 1. The median age at diagnosis was 49.5 years for the patients with WT FLT3 and 51.7 years for the patients with FLT3/ITD. The median white blood cell count at diagnosis was 37,000/μL for the FLT3/ITD patients and 11,700/μL for the WT patients. Most (24/31) of the FLT3/ITD patients had normal cytogenetics; 2 of these patients had unfavorable cytogenetics and 1 had favorable. Of the WT cohort, 47 had unfavorable cytogenetics, 44 had intermediate (36 normal) and 11 had favorable. The WT cohort also included 14 patients with treatment-related AML and 26 with antecedent hematologic disorders.

Figure 1 outlines the disposition of patients by therapy. There were a total of six induction deaths between the two groups. Fourteen patients were deemed unfit for intensive induction therapy by ECOG[12] performance status at presentation or complicated medical comorbidities. Two were in the induction death group. None of these patients had FLT3/ITD mutations. These patients were excluded from further analyses. All other patients received one of two intensive regimens as previously described.

The median OS from diagnosis for all 119 patients receiving intensive induction was 19.6 months. The median survival times for favorable, intermediate, and unfavorable cytogenetic groups were 57.3, 18.9, and 9.4 months, respectively. The median OS for the FLT3/ITD AML patients was 19.3 (range 0.0-69.9) months and was similar to the median OS of the WT patients of 15.5 (range 0.7 – 64.6) months (p =0.56). (Figure 2) Induction success was similar between the two groups with remissions obtained in 65% (20/31) of the FLT3/ITD patients and 61% (52/85) of WT patients.

Of the 20 FLT3/ITD patients in CR1, 11 (55%) underwent allogeneic HSCT in CR1 (4 myeloablative, HLA-matched sibling donors, 5 myeloablative, HLA-matched unrelated donors, and 2 nonmyeloablative haplo-identical related donors). The remaining 9 FLT/ITD patients in CR1 did not go to allogeneic HSCT due to lack of a suitable donor or precluding comorbidities following induction. The median relapse-free survival in the FLT3/ITD non-transplant group was 8.6 months (range 5.3-43.3 months), which was significantly shorter than the 54.1 months (range 6.4 – 69.9 months) in the FLT3/ITD transplant (p=0.03) (Figure 3).

In contrast, of the 52 WT patient in CR1, 17 (33%) of WT patients underwent HSCT in CR1 (5 myeloablative HLA-matched sibling donors, 9 myeloablative, HLA-matched unrelated donors, 1 syngeneic transplant, 1 autologous transplant, and 1 nonmyeloablative haplo-identical related donor.) (Table 1) The median OS in the WT, non-HSCT group was 57.3 (range 3.9- 64.4) months while the median OS in the WT transplant group is greater than 60

months (p=0.02). Figure 4 demonstrates the difference in the survival curves for all the patients in both cohorts based on any transplant during their course.

Discussion

As FLT3/ITD mutants fall into the category of unfavorable risk, it has been hypothesized that these patients will benefit from allogeneic HSCT in CR1 to improve the outcomes and survival. The date to prove or refute this hypothesis remains controversial.

In 2005, Gale and colleagues at the Medical Research Council of the United Kingdom investigated whether AML patients with a FLT3/ITD mutation have an improved outcome if they undergo HSCT, compared to similar patients receiving chemotherapy.[19] In a retrospective analysis of patients, comparisons were made between patients receiving autografts versus allografts, and between patients receiving autografts versus no transplant. The presence of the FLT3/ITD mutation was an accurate independent predictor of relapse, and it remained prognostic for increased relapse even in those patients who received a transplant.[19] The authors therefore concluded was that there was insufficient evidence that FLT3/ITD status should influence the decision to transplantation.[19] However, the data analysis in the 2005 study limited the application of the results, because there was no direct comparison between FLT3/ITD patients receiving allografts and those receiving chemotherapy alone.[20]

The impact of different consolidation therapies on overall survival (OS), and on the probability of relapse in patients with FLT3/ITD mutation versus wild-type (WT) was studied by the German study initiative leukemia group.[21] The study showed that after a median follow-up of 53 months, OS was not significantly different between FLT3/ITD mutants and WT. In contrast, chemotherapy alone as consolidation therapy had inferior OS, and increased rates of relapse the FLT3/ITD mutants. [21]

Given the conflicting results in these larger cohorts, we sought to evaluate our experience of 31 FLT3/ITD patients with a comparable OS outcome to that for the 102 patients with WT FLT3 over the same 4 year time period. Though the transplanted population of FLT3ITD patients is quite small (11 patients), this finding is of clinical interest and adds to the previous studies suggesting an advantage for transplantation in this group. Our analysis is derived from a consecutive series of newly-diagnosed AML patients and lacks the potential selection biases inherent in data derived from prospective trials. As such, our report represents a “real world” perspective on the challenging management of FLT3/ITD AML. Historically, the OS for AML patients with the FLT3/ITD mutation is significantly worse than for AML patients lacking this mutation. The outcomes presented here are comparable to other published results[18;22] and suggest that our patient set and the responses to treatment are representative of a typical adult AML population, including the influence of unfavorable cytogenetics in nearly half the WT cohort. Although two different induction therapies were used, these subgroups were fairly well balanced as evidenced by the percent of patient achieving a CR1 in each treatment group. Inclusion of the larger WT cohort is intended to contrast the FLT3ITD group and develop the background for the generalizability of the groups. It is possible that our institution’s use of post transplant cyclophosphamide to mitigate GVHD contributed to the apparent benefit of allogeneic transplant for FLT3/ITD patients. Certainly, this approach did allow us to consider all available donor transplants for both cohorts, especially the FLT3 cohort, including haplo-identical and unrelated. However, the rates of relapse in hematologic malignancies after post transplant cyclophosphamide are comparable to the rates of relapse after more traditional immunosuppression in other centers. [17] Furthermore, other groups have suggested that allogeneic transplant is the preferred consolidation therapy for FLT3/ITD AML.[21] This would suggest that this approach could be

considered at other institutions willing to proceed to alternative donor transplants. During this time period, our institution was not routinely testing NPM1 mutation status in all patients. The impact this additional information would have had on the outcomes of these patients is not known. We have also not generalized this approach to FLT3 TKD mutants given their controversial prognostic significance.[23-25]

The surprising outcome for the FLT3/ITD group may be partly attributed to our aggressive pursuit of allogeneic BMT in CR1 within this group (60% of FLT3/ITD vs. 17% with WT). Our single institution study of consecutively treated AML patients supports the hypothesis that allogeneic transplant in early CR1 may improve the long term outcomes for patients with FLT3/ITD AML.

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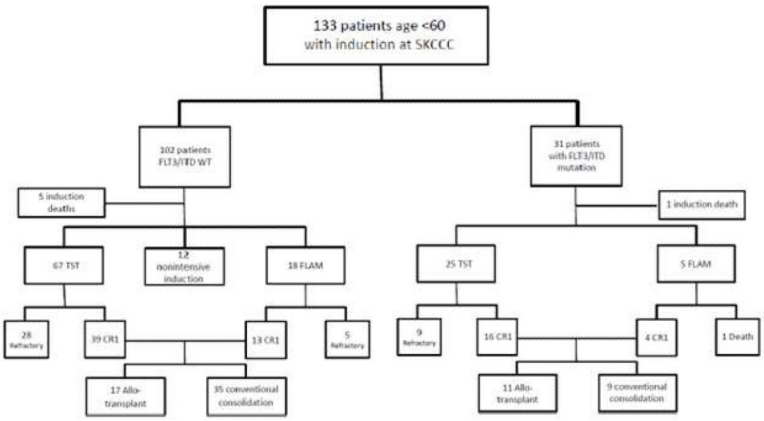
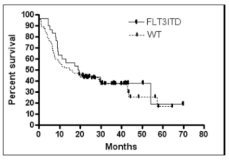
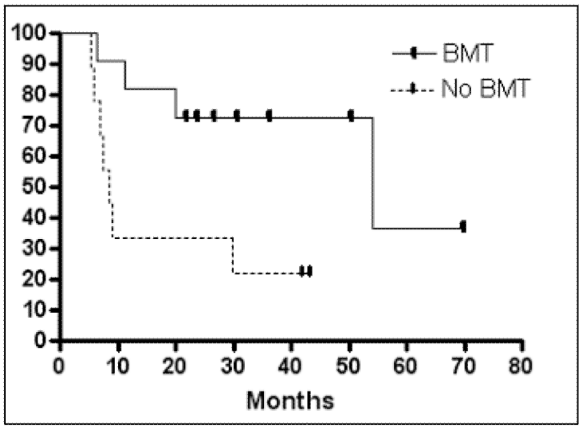


Figure 1.
Patient Disposition by FLT3/ITD Mutation Status and Treatment



	TOTAL	DEATHS	CENSORED	MEDIAN SURVIVAL
WT	102	66	36	15.5 mos
FLT3/ITD	31	19	11	19.3 mos

Figure 2.
Overall Survival of 133 Patients by FLT3/ITD Mutation Status



	TOTAL	EVENTS	CENSORED	MEDIAN SURVIVAL
BMT	11	4	7	54.1 mos
No BMT	9	7	2	8.6 mos

Figure 3.
Event Free Survival of FLT3/ITD Patients transplanted in CR1

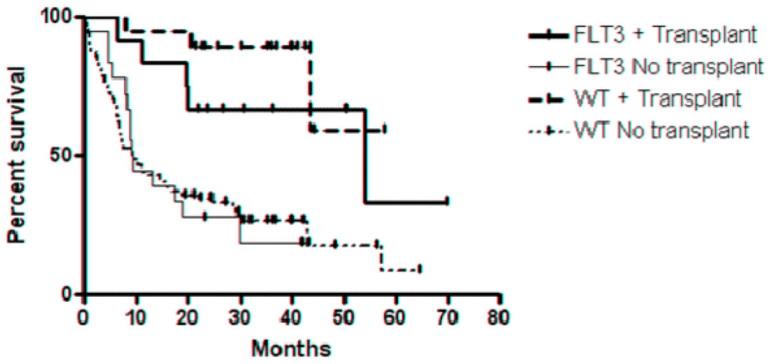


Figure 4.
Overall survival of FLT3/ITD and WT patients transplanted versus no transplant

Table 1

Demographics of the FLT3/ITD and Wildtype Cohorts

	Wild-Type	FLT3/ITD
Total patients	102	31
Age (years)	49.5	51.7
Sex (%Males)	51	52
WBC at Diagnosis (1000 /cu mm)	11.7	37.0
Type		
De Novo	62	26
Antecedent disorder	26	5
tAML	14	0
Cytogenetics		
Favorable	11	1
Intermediate	42	23
Unfavorable	47	7
% Normal Cytogenetics	35	74
% Transplanted	17	60
Transplant Types		
Matched sibling	6	4
Matched unrelated	9	5
Haploidentical	1	2
Syngeneic	1	0
Auto	1	0