Hematopoietic Stem Cell Transplantation for Childhood Acute Lymphoblastic Leukemia and the Role of MRD: A Single Centre Experience from India

Divya Subburaj¹ · Lakshman Vaidyanathan² · Ramya Uppuluri¹ · Dhaarani Jayaraman¹ · Revathi Raj¹

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Abstract Hematopoietic stem cell transplantation (HSCT) is an effective curative option for children with relapsed and high risk acute lymphoblastic leukemia (ALL). The effect of minimal residual disease (MRD) prior to transplantation has a significant impact on the overall outcome. We performed a retrospective analysis of children with ALL who underwent HSCT at our centre from 2002 to 2016. From 2002 to 2008 disease status was determined by morphology and karyotyping and from 2008 onwards by flow cytometry. A total of 46 children were transplanted for ALL at our centre. Of the 19 children who were MRD positive prior to HSCT 5 had a relapse after the transplant. Among the remaining 26 MRD negative children, only one child relapsed post HSCT. The EFS was 66.6% in the MRD negative group and 63.1% in positive group with no significant survival advantage of the first group over the second, (p 0.37). GVHD was the major cause of mortality overall at 56.7% as well as in the MRD negative group at 77.7%(7/9). On the other hand, relapse was the major mortality factor at 71.4%(5/7) in the MRD positive group. Molecular remission prior to HSCT shows a trend towards lesser chance of relapse. We should strive to achieve MRD negative status prior to transplant to improve EFS. However, GVHD is also emerging as a crucial factor and its impact on survival outcome in children undergoing HSCT for ALL needs to be followed up.

Keywords ALL · HSCT · MRD

Introduction

Hematopoietic stem cell transplantation (HSCT) is an effective curative option for children with relapsed and high risk acute lymphoblastic leukemia (ALL). The curative effect of HSCT is hampered by relapse and treatment related mortality and morbidity. Minimal residual disease (MRD) status prior to transplant is an independent prognostic marker of relapse and overall survival outcome in children undergoing HSCT for ALL [1–5]. There is limited data on childhood cancer survival post transplant from India. Our study aims to assess the outcomes in children undergoing HSCT for ALL and analyse the effect of MRD status prior to HSCT on the relapse and event free survival (EFS) rates.

Patients and Methods

Between Aug. 2002 and Feb. 2016, 46 children aged less than 18 years underwent HSCT for ALL at our centre. The demographic data, disease characteristics, transplant variables and outcome were obtained from our medical record data base that contained prospectively collected data on pediatric patients transplanted for ALL. Informed written consent was obtained prior to transplant.

The disease status was assessed prior to transplant and was repeated on day 100 post HSCT. From 2002 to 2008 disease status was determined by morphology and karyotyping, however from 2008, MRD was determined by flow-cytometry. Leukemia associated aberrant immunophenotype (LAIP) were identified by multi-

✉ Divya Subburaj
divyanls@yahoo.co.in

¹ Department of Pediatric Hematology, Oncology and Blood and Marrow Transplantation, Apollo Speciality Hospital, 320, Padma Complex, Anna Salai, Chennai 600035, India

² Division of Hematology, Apollo Speciality Hospital, 320, Padma Complex, Anna Salai, Chennai 600035, India

parametric flow-cytometry and all patients had at least one phenotype for follow up. HLA compatibility was determined by low resolution typing in matched sibling donor (MSD) transplants and high resolution molecular typing in unrelated donor (URD) and haplo identical transplants. Related grafts and cord blood units were matched at HLA loci-A, B and DRB1. Unrelated donor grafts were matched at HLA-A, B, C, DRB1 and HLA-DQB1.

Definition

MRD positive-status was defined as any patient specific LAIP detected on flow-cytometry or by karyotyping findings.

Event was defined as either relapse or death.

Treatment Plan

The conditioning regimen consisted of cyclophosphamide and total body radiotherapy in patients with a MSD transplant. Anti-thymocyte globulin was given to all recipients of unrelated donor graft and post transplant cyclophosphamide was used in haplo identical transplant. Prophylactic antifungal and antiviral agents were started prior to conditioning. Transfusion support was provided with irradiated, leucodepleted blood products. Graft versus host disease (GVHD) prophylaxis consisted of tacrolimus in unrelated cord transplant and a combination of tacrolimus and methotrexate in the other graft sources.

Statistics

Event related data was measured from date of transplant to date of relapse or death. On the date of last follow up in patients without the respective event, data was censored. The data was analysed using Chi square test, log rank test and the probability of EFS was estimated by Kaplan–Meier method.

Results

A total of 46 children with ALL were transplanted at our centre from 2002 to Feb. 2016. The children were aged between 1 year and 18 years (median age 11 years). Fifty-eight percent of them were male. The most common indication for transplant was relapsed ALL (CR2) in 26 children (56.5%).

MSD graft was used in 21 (45.6%) children and URD graft in 20 (43.4%). Haplo identical transplant was performed in 3 children and mismatched family donor was used in remaining two. Peripheral blood stem cells were

used in 33 (71.75%) children followed by cord blood unit in 7 and bone marrow in 6.

MRD was documented negative in 27 (58.6%) children prior to HSCT. One child relapsed post transplant in this cohort when compared to 5 children who had a relapse in the MRD positive cohort.

Eighty percent of the children who experienced a relapse had no features of graft versus host disease (GVHD) in the post transplant period. The absence of GVHD showed a higher chance of relapse with a relative risk ratio of 8.33 (p 0.03).

The overall mortality rate in our study was 32.6% and EFS at end of 1 year was 64.8%. GVHD was the major cause of mortality in children who underwent HSCT for ALL at 56.7%. It was the principal factor contributing to mortality in MRD negative group at 77.7%(7/9). On the other hand, relapse was the major mortality factor at 71.4%(5/7) in the MRD positive group. However the EFS was 66.6% in the MRD negative group and 63.1% in positive group with no significant survival advantage seen in the first group over the second, (p 0.37)

Discussion

Around 80–85% of children with acute lymphoblastic leukemia can be cured with risk stratification based chemotherapy protocols [6–10]. A review article analysing the published literature on childhood ALL outcomes in India has put the overall survival rates from 45 to 81% and EFS rates from 41 to 70%. The relapse rates also had a wide range from 18 to 41% [11]. Allogenic HSCT provides a survival advantage over continuing chemotherapy in children with relapse or very high risk features like Ph+ve chromosome, MLL gene mutation, hypodiploidy, high presenting WBC count >100,000/ul [12] (Table 1).

Detection of MRD prior to transplant has been considered a predictive marker for relapse following HSCT. The study by Knetchli et al. [13] showed that high MRD burden prior to HSCT had a significant poor outcome. Studies by Vander Velden et al. [14] and Bader et al. [15] had confirmed the above finding. All the above studies determined MRD by polymerase chain reaction amplification (PCR) of antigen receptor region. In our study MRD was measured by flow cytometry which also showed a high relapse rate 31.5% in the MRD positive group versus 7.4% in the negative group which concurred with the findings of other studies that used PCR based technique. Achieving molecular remission prior to transplant with intense chemotherapy offers a survival outcome [13, 14] (Fig. 1).

Patients in the MRD negative group from our study (Fig. 2) did not have a survival advantage over the patients with MRD positive status as shown by the above studies.

Table 1 Demographic data

Median age	11 years
Sex	
Male	26/46 (58%)
Female	20/46 (42%)
Indications for HSCT	
Relapsed ALL—CR2	26/46 (56.5%)
High risk All—CR1-HSCT	20/46 (43.4%)
Failure to achieve molecular remission	10/20
Ph+veALL	3/20
Hypodiploidy	2/20
ETp ALL	1/20
MLL, ALL + SCID, Biphenotypic, complex karyotype	Each 1/20
MRD status	
MRD negative	27/46 (58.6%)
MRD positive	19/46 (41.3%)
Donor graft	
MMRD + URD	22/46 (47.8%)
MSD	21/46 (45.6%)
Haplo identical	3/46 (6.5%)
Source of stemcell	
PBSC	33/46 (71.7%)
Unrelated cord	7/46 (15.2%)
Bone marrow	6/46 (13%)

Fig. 1 Algorithmic representation of outcome of children with ALL who underwent HSCT

Higher incidence of GVHD (grade 3 and 4) in the MRD negative cohort has emerged as a factor that needs to be followed up to assess its impact on mortality. Another interesting observation made during the study was that children with unrelated or mismatch family donors (61.7%) grafts had higher incidence of GVHD (grade 3 and 4) when compared to MSD (32.2%). However studies by Harvey et al. have concluded that survival has improved in the

URD group with introduction of high resolution HLA typing and bone marrow as the source of graft rather than peripheral blood stem cells [16, 17]. Though high resolution HLA typing has been used in our URD transplants, PBSC still remained the major source of stem cells in this group.

GVHD provides a strong graft versus leukemia effect and reduces the chance of relapse [16, 17]. Our data also

Fig. 2 Kaplan–Meier survival curve for MRD positive and MRD negative group

confirms that children with no GVHD carried a higher relapse risk making GVHD a necessary evil. To strike the right balance between GVHD and relapse is crucial. It is also challenging as the degree of GVHD needed to provide graft versus leukemia effect is not uniform and needs to be individualised [18, 19].

The limitation of our study is the small sample size which restricted our ability to consider other confounding factors like cytogenetics, graft choice, underlying infections prior to transplant on the EFS. There is a lack of published data on HSCT for pediatric ALL from India and further follow up of this cohort will provide us a better understanding of the outcome variables.

Conclusions

Molecular remission prior to HSCT shows a trend towards lesser chance of relapse. We should strive to achieve MRD negative status prior to transplant to improve the EFS. However, GVHD is also emerging as a crucial factor and its impact on survival outcome in children undergoing HSCT for ALL needs to be followed up.

Compliance with ethical standards

Conflict of interest The authors declares that they have no conflict of interest.

Informed consent For this type of study formal consent is not required.

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