

Introduction

Most cases of aplastic anemia result from immune-mediated depletion of hematopoietic stem cells. It may also occur as a result of inborn errors of metabolism (for example, Fanconi anemia and Schwachman-Diamond syndrome) or exposure to chemicals or radiation. It is characterized by peripheral pancytopenia and a hypocellular bone marrow. The clinical presentation is typically due to symptoms associated with thrombocytopenia, anemia and neutropenia.

Aplastic anemia is classified on the basis of marrow cellularity and the degree of pancytopenia. Diagnostic criteria for severe aplastic anemia include a bone marrow biopsy showing an overall cellularity of less than 25% of the age-appropriate normal cellularity or less than 50% normal cellularity in which fewer than 30% of the cells are hematopoietic. At least two of the following should also be present: absolute reticulocyte count $<25,000/u/L$, absolute neutrophil count $<500/u/L$ or platelet count $<20,000/u/L$. Those who meet the bone marrow criteria for severe aplastic anemia, but have an absolute neutrophil count $<200/u/L$ are diagnosed with very severe aplastic anemia. Patients with pancytopenia who do not meet the criteria for severe aplastic anemia are classified as having non-severe aplastic anemia. Acquired aplastic anemia should also be distinguished from inherited bone marrow failure syndromes in young adults prior to initiating therapy.

Treatment strategies for aplastic anemia include immunosuppressive therapy and hematopoietic stem cell transplantation. The choice of therapy depends on the severity of the disease, patient age and availability of a human leukocyte antigen (HLA) matched sibling donor. Few clinical trials have addressed optimal treatment for non-severe aplastic anemia. However, a prospective randomized control trial has shown that combination immunosuppressive therapy, with cyclosporine and antithymocyte globulin, can reverse moderate pancytopenia and reduce transfusion requirements, suggesting a role for immunosuppressive therapy in patients with non-severe aplastic anemia.^{1,2} Patients with very severe aplastic anemia are treated in the same manner as those with severe aplastic anemia. This review will focus on the role of hematopoietic stem cell transplantation (HSCT) in the treatment of severe and very severe aplastic anemia.

Allogeneic Hematopoietic Stem Cell Transplantation

Allogeneic hematopoietic stem cell transplantation (HSCT) from an HLA matched sibling donor is the first-line treatment of choice for newly diagnosed patients with severe acquired aplastic anemia if they are younger than 30-40 years.^{3,4} It may also be considered second or third-line therapy in those who have relapsed or failed immunosuppressive therapy. Most of the data describing outcomes associated with HSCT in patients with aplastic anemia come from retrospective European (EBMT) or International (IBMTR) registries as well as single or combined center studies.

HLA Matched Sibling HSCT

Allogeneic stem cell transplantation from a matched sibling donor is a potentially curative treatment with long-term overall survival reported in 80% to 90% of children and minimally-transfused young adults.⁵ Transplant-related mortality has been shown to increase with age, reaching 50% in patients older than 40 years.² The 10-year overall survival for HLA-identical sibling transplantations is 83%, 73%, 68% and 51% for patients aged 1–20, 21–30, 31–40 and ≥40 years, respectively.⁶ In 2007, the EBMT-SAAWP published data from a retrospective cohort study of 1567 patients who received a stem cell transplant as first-line therapy for aplastic anemia.⁷ Favourable predictors for survival in this cohort included younger age, transplant after 1996, a matched sibling donor, a short diagnosis-to-transplant interval and no irradiation in the conditioning regimen. In this cohort, 10-year overall survival for patients transplanted from HLA-matched siblings after 1996 was 80%. This was superior to the outcome (73%) of similar patients treated over the same time period with immunosuppression alone.

Potential complications associated with a matched sibling HSCT include graft failure and graft-versus-host disease (GVHD). Graft rejection occurs in 5-15% of patients with SAA transplanted from matched sibling donors.⁸ Particularly at risk are patients who have been heavily transfused or alloimmunized.⁸ A GITMO/EBMT randomized trial comparing cyclosporine and methotrexate (CsA/MTX) versus cyclosporine (CsA) for GVHD prophylaxis after a matched sibling transplant for aplastic anemia showed a slight, non-statistically significant, difference in the development of grade 2 to 3 acute GVHD, 38% for CsA versus 30% for CsA/MTX.⁹ None of the patients developed grade 4 GVHD and there was no difference in transplant-related death prior to day 100 post-HSCT (2% versus 0%). However, all patients who developed acute GVHD went on to develop chronic GVHD.

The development of chronic GVHD is associated with growth and endocrine system dysfunction, pulmonary disease, cataracts, neurologic dysfunction, and secondary malignancy. It has been consistently associated with reduced quality of life, poorer general health and a reduced ability to function in the workplace.¹⁰ Risk factors for chronic GVHD include TBI-based conditioning, stem cell source, acute GVHD, and older age at the time of transplant.^{8,11} According to the IBMTR, rates of severe GVHD in adults are twice those reported in children (15%-20% in those ≤ 20 years versus 40%-45% in those > 20 years)¹², making the complications associated with GVHD of particular concern when considering HSCT for adult patients with severe aplastic anemia.

The current most widely used conditioning regimen for HLA-matched sibling HSCT is a nonmyeloablative approach, often using high-dose cyclophosphamide and ATG.⁸ Radiation-based conditioning has largely been abandoned for this indication due to its adverse impact on GVHD and secondary malignancy rates. A prospective clinical trial of 134 patients who were randomly assigned to receive cyclophosphamide alone or in combination with ATG prior to receiving a bone marrow graft from a HLA-matched sibling donor showed no difference in rates of graft failure and GVHD.¹³ There was a small, non-statistically significant difference in overall survival (74% for cyclophosphamide alone vs. 80% for cyclophosphamide with ATG, $p=0.44$) but the study was under-powered to detect a survival difference.

A recent report describing a novel conditioning regimen consisting of low-dose cyclophosphamide, ATG and fludarabine, shows low rates of GVHD and TRM, even in adults with severe aplastic anemia.¹⁴ The combination of cyclophosphamide plus fludarabine, with or without ATG, has also been shown to be associated with high rates of engraftment and survival, even in heavily transfused patients.^{15,16} A study of 26 patients with bone marrow failure syndromes, including 13 with refractory severe aplastic anemia, who received conditioning consisting of cyclophosphamide (120 mg/kg) and fludarabine (125 mg/m²) with or without ATG.¹⁷ All patients engrafted by day 30 post-transplant, resulting in transfusion independence. During the follow-up period (median of 21 months), 24 (92%) patients had survived disease-free with only one patient dying from transplant-related causes. However, a high incidence of acute (grades 2 to 4 in 65%) and chronic GVHD (56%) was observed. This is consistent with an NIH trial which also showed that the addition of fludarabine was associated with high rates of acute and chronic GVHD¹³, indicating that more investigation is necessary to determine whether the use of fludarabine will actually translate into improved survival. More recently, a small study has been published describing the outcomes associated with using a combination of

fludarabine, cyclophosphamide and alemtuzumab.¹⁸ In this study, peripheral stem cell grafts, obtained from HLA-matched sibling donors, were incubated with alemtuzumab ex vivo as a means of depleting T-cells within the graft in an attempt to maximize engraftment and minimize GVHD. Fifteen patients were enrolled with engraftment seen in all by day 18 post-HSCT as well as 100% survival during the study period (median duration of 1107 days). None of the patients developed acute or chronic GVHD. Although many of the proposed regimens show promise, further investigation is required to determine whether one particular conditioning regimen shows superiority over currently used regimens.

The use of growth factor-mobilized peripheral blood rather than bone marrow as a source of stem cells has resulted in reduced time to engraftment and improved overall survival in patients with hematological malignancy.¹⁹ In SAA, however, chronic GVHD is associated with significantly reduced overall survival, as described in a joint report from the EBMT and IBMTR.¹¹ In this report, the outcomes in 134 patients who received peripheral blood stem cell grafts and 558 patients who received bone marrow grafts were compared.¹¹ The rates of hematopoietic recovery, graft failure and acute GVHD were comparable between groups, regardless of age. Rates of chronic GVHD were higher after peripheral blood stem cell transplantation than after bone marrow transplantation in patients less than 20 years old (RR 2.82). The use of peripheral blood as a stem cell source was associated with reduced overall survival when compared with bone marrow in patients ≤ 20 years (85% vs 73%, $p=0.024$) and in patients >20 years (64% vs 52%, $p=0.119$). Chronic GVHD was the major cause of mortality in the peripheral blood group. This report highlights the impact that chronic GVHD has on overall survival, especially in patients with aplastic anemia who do not benefit from the graft versus tumor effect associated with GVHD.

An important factor for patients when making decisions about their own care is treatment-associated quality-of-life. A retrospective study, published in 2005, looked at quality-of-life outcomes in 52 patients who received HSCT and 155 patients who received IST between 1976 and 1999.²⁰ Overall and event-free survival was similar between the two groups. However, IST treated patients had longer periods of time with symptoms from drug toxicity, transfusion dependency, partial remission and secondary clonal disorders than those who underwent HSCT. In comparison, transplanted patients spent more time in complete remission without drugs and had longer periods free from symptoms, with the exception of those that developed chronic GVHD. Therefore, HSCT has been shown to be associated with better quality-of-life than IST, especially in those who do not develop GVHD.²⁰

Matched Unrelated Donor HSCT

A matched sibling donor is available in 20–30% of cases being considered for an allogeneic HSCT.²¹ For patients who fail a trial of immunosuppressive therapy and do not have an HLA-matched sibling donor, consideration of a matched unrelated donor (MUD) HSCT is an important treatment option. However, the mortality rate is about twice that observed in matched sibling transplants.⁵ An IBMTR retrospective study of outcomes in 318 MUD transplants performed for severe aplastic anemia from 1988 to 1998 indicated a rejection rate of 15%, grades 2 to 4 GVHD of 48% and 5-year survival was estimated at 39%.²² Since this report was published, the use of high-resolution, DNA-based techniques for HLA typing has led to improved outcomes for patients undergoing stem cell transplantation from unrelated donors. A report by Kojima et al, described a 60% overall survival in MUD HSCT using high resolution DNA-based testing for HLA-A, B, C and DR.²³ This study also described older age (>20 years), conditioning without ATG and a long (>3 years) interval between diagnosis and transplant as being unfavourable factors for survival for patients receiving MUD HSCT for severe aplastic anemia.

As with matched sibling HSCT, there has been interest in identifying the optimal conditioning regimen for MUD HSCT in patients with severe aplastic anemia to minimize morbidity and mortality. A 2005 study from EBMT-SAAWG using fludarabine, low-dose CY (1200 mg/m²) and ATG, without irradiation, reported a lower rate of acute and chronic GVHD.²⁴ In this study, patients under the age of 15 years had an 84% 2-year survival and only 5% graft rejection. However, in older patients, the results were less favourable with 61% survival and 32% rejection. Many transplant centers use the same conditioning regimens for MUD HSCT as they do for transplants from matched siblings in severe aplastic anemia.

Less toxic conditioning regimens and improved donor-recipient matching have shown improvements in survival and engraftment associated with MUD HSCT over the last decade.^{7,25} Despite these improvements, outcomes in adult patients (>20 years) remain inferior to those seen within the pediatric population.

Cord Transplant

The proportion of patients who lack either a matched sibling donor or suitable unrelated donor varies depending on the ethnic origin of the patient, but ranges between 5% and 40%.²⁶ HLA mismatch is better tolerated using umbilical cord blood (UCB) for HSCT, making it a potential treatment option for severe aplastic anemia when there are no other suitable donors. However, published data describing the use of UCB for transplantation in this setting is limited, especially in the adult population. Of particular concern with this approach is the small

number of stem cells available in a single UCB donation which may increase the risk of graft failure in a population typically associated with high rates of graft failure.

A study using the Japan Cord Blood Bank Network database, published in 2008, described the outcomes in 31 patients who were recipients of UCBT for severe aplastic anemia between 1998 and 2006.²⁷ Cord blood units were assessed using serology for 0 to 2 HLA locus mismatches in HLA-A, B and DRB1 and the unit with the largest cell dose (minimum of 2.0×10^7 /kg mononuclear cells) was selected for transplantation. The rate of grade 2 to 4 acute GVHD and chronic GVHD was 17.1% and 19.7%, respectively, with an estimated 2 year overall survival of 41.1%. The conditioning regimens used varied between institutions within this study. The use of low-dose total body irradiation, fludarabine and cyclophosphamide seemed to be associated with improved 2 year overall survival (80%), but must be considered cautiously due to a small sample size.

To date, there is insufficient evidence to define the role of UCB transplantation in adult patients with severe aplastic anemia and remains an area of intense interest. Researchers continue to explore issues associated with UCB transplantation, such as ideal stem cell dosing, multiple UCB transplants and novel conditioning regimens, which may clarify its place within this patient population.

Conclusion

The development of chronic GVHD has been shown in many studies to be a large contributing factor to transplant-related mortality, especially in adult patients. The use of an allogeneic HSCT protocol which minimizes GVHD while still promoting reliable, sustained engraftment is the key to achieving desired patient outcomes. Improvements in survival have been shown in association with both matched sibling and MUD HSCT through the use of more sophisticated techniques for HLA-matching and the development of less toxic conditioning regimens. Despite these improvements, adult patients (>20 years) have not achieved the outcomes routinely obtainable in the pediatric population with severe aplastic anemia. Treatment decisions should, ultimately, be based on patient age, functional status, comorbidities and individual institution practices for conditioning and GVHD prophylaxis. Since HSCT results in more rapid and more frequent restoration of hematopoiesis than immunosuppressive treatment, it remains the treatment of choice for young patients with severe and very severe aplastic anemia. The morbidity and high mortality rates observed in older adults (>40 years) suggests caution and a trial of immunosuppressive therapy in most situations. At the present time, there is a lack of evidence to support the

use of UCB grafts for transplant in patients with severe aplastic anemia outside of a clinical trial.

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