BONE MARROW TRANSPLANTATION VERSUS IMMUNOSUPPRESSIVE THERAPY IN SEVERE APLASTIC ANEMIA, 1990 – 2001

Ardeshir Ghavamzadeh MD, Masood Iravani MD, Farahnaz Vafaiezadeh MD, Mohammad Jahani MD, Asadollah Mousavi MD

Background — Bone marrow transplantation (BMT) and immunosuppressive therapy are two choices of therapy for aplastic anemia. In BMT, abnormal cells are replaced by normal donor’s hematopoetic stem cells in those patients who have an HLA-identical match donor and are aged < 45 years old. In immunosuppressive therapy, antilymphocyte globulin (ALG) and cyclosporin are used in patients who do not have an HLA-identical match donor and are aged > 45 years old.

Methods — In this study we compared these two modalities of treatment in acquired severe aplastic anemia. We had 70 patients in two groups. Twenty-nine patients had completed BMT and 41 patients had completed non-BMT treatment. The conditioning regimen in BMT group was cyclophosphamide plus ALG. Patients with severe aplastic anemia who had been referred to the Hematology Clinic of Shariati Hospital from 1990 through 2001, were selected according to age (< 45 or > 45 years) and presence of HLA match donor. Ethical considerations were strictly followed. Data were analyzed by SPSS version 10. Survival probabilities were estimated using Kaplan-Meier method.

Results — The 5 years overall survival in BMT group was 67% and in cyclosporin group was 36.6% and we found that after the day 200 posttherapy overall survival in BMT group was higher in comparison with non-BMT treatment (p = 0.02).

Conclusion — BMT has the best results and long-term survival in severe aplastic anemia patients.

Keywords: Antilymphocyte globulin (ALG) • aplastic anemia • bone marrow transplantation (BMT) • cyclosporin

Introduction

Severe aplastic anemia (SAA) is a rare disease and probably an immune mediated disorder. It is manifested by pancytopenia in peripheral circulation. Patient’s hematopoetic activity in bone marrow histologically decreases, the number of CD34 cells is low, and colony formation in bone marrow is not significant. In most cases the etiology of SAA is unknown. Its pathophysiology is not completely understood and it is possible that there are various etiologies ultimately presenting as marrow failure.

Bone marrow transplantation (BMT) is the treatment of choice for SAA. The source of stem cells can be from bone marrow or peripheral blood and is generally performed for the patients who are younger and have HLA-identical sibling donor. During the 1970s the survival rate among patients receiving transplants from HLA-genotypically identical donor was commonly 40 – 50% compared to 60 – 90% in recent years. Successful allogeneic BMT is
dependent on a stem cell product of sufficient quality to guarantee stable engraftment.\textsuperscript{8} Intensive conditioning regimens are used for preventing graft rejection of allogeneic BMT from HLA-identical sibling donor.\textsuperscript{6 – 8} Infection and graft-versus-host disease (GVHD) are the major complications of BMT.\textsuperscript{9 – 10}

Another treatment for aplastic anemia is immunosuppressive therapy. This treatment is employed in the patients who are older and in cases without an HLA match donor.\textsuperscript{1, 5, 11, 12} The goal of treatment in aplastic anemia is an improvement in peripheral blood cell counts so that, these patients no longer require transfusions and are not at risk of opportunistic infections.

In contrast to BMT where the impaired organ is replaced by healthy stem cells, immunosuppressive therapy is not curative. The goal here is the elimination of the autoaggressive cells responsible for aplastic bone marrow that result in pancytopenia.

Today’s combination of antilymphocyte globulin (ALG) plus cyclosporin is the treatment of choice for aplastic anemia patients who are not candidates for BMT.\textsuperscript{2, 3, 7, 13 – 17} The rate of response to the combination of ALG and cyclosporin has been 60 – 70%.\textsuperscript{18} In contrast to BMT, response to immunosuppressive therapy is often slow and incomplete.

It has been shown that androgens could induce remission in congenital and acquired aplastic anemia but this does not seem to improve survival in aplastic anemia. It is therefore used as minor treatment in some cases of moderate disease where immunosuppression has failed.\textsuperscript{12, 19 – 21} Corticosteroids were used long before ALG to treat aplastic anemia. Today, most ALG protocols include a median dose of corticosteroids for a short duration in an effort to reduce the ALG toxicity and symptoms of serum sickness disease.\textsuperscript{12} Splenectomy should be considered for selected non-BMT patients who despite of maximum therapy need long-term transfusion.\textsuperscript{22}

Since BMT is curative for SAA, in our study we compared outcomes of BMT versus immunosuppressive therapy in patients with severe aplastic anemia.

Materials and Methods

All patients with SAA who had been referred to the Hematology Clinic of Shariati Hospital from 1990 through 2001, were selected according to age (< 45 or > 45 years) and presence of HLA match donor. All ethical considerations were strictly followed.

In patients who had an HLA-identical match sibling donor and were younger than 45 years old, BMT was performed. The conditioning regimen for immunosuppression was cyclophosphamide 50 mg/kg for 4 days and in high-risk patients ALG 10 mg/kg for 3 days.\textsuperscript{6}

The stem cells were infused after conditioning the bone marrow or peripheral blood. GVHD prophylaxis regimen was intravenous cyclosporin 3 mg/kg (days -3 to +5) followed by 12.5 mg/kg orally continuously for one year.\textsuperscript{23, 24} Acute complications of BMT such as infection and GVHD were treated if these occurred.

Immunosuppressive therapy was performed in patients older than 45 years and/or without an HLA-identical match donor. In this method oral cyclosporin 3 mg/kg for 6 months to 2 years was used. In ALG-containing group, ALG was infused 10 mg/kg for 4 days. Blood cyclosporin level was also maintained between 100 – 200 ng/mL.

In addition, in some cases androgens plus prednisolone were used. Danazol with an average dose of 100 mg (50 – 200 mg) or oxymetholone with an average dose of 150 mg (100 – 300 mg) was used and the average dose of prednisolone was 15 mg/day.\textsuperscript{19}

Statistical methods

Using the SPSS Version 10 software package, survival probabilities were estimated by the Kaplan-Meier method and comparisons were based on log-rank statistics and frequencies were calculated.

Results

In this study, we had a total of 70 patients of whom 29 patients were in the BMT group and 41 patients in the non-BMT group. Patient’s data have been summarized in Table 1. Possible etiologies in the different groups are presented in Table 2. The mean number of transfused packed cells before
BMT versus immunosuppressive therapy in SAA between years 1990 – 2001

Table 1. Data of the patients.

<table>
<thead>
<tr>
<th>Group</th>
<th>Number of patients</th>
<th>Mean age (yr)</th>
<th>Sex</th>
<th>Male</th>
<th>Female</th>
<th>Duration between diagnosis to treatment (D)</th>
<th>Follow-up (D)</th>
<th>Complete (%)</th>
<th>Partial (%)</th>
<th>5 years survival (%)</th>
<th>Mortality (%)</th>
<th>Splenectomy (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>BMT</td>
<td>29</td>
<td>19</td>
<td>19</td>
<td>10</td>
<td>377</td>
<td>878</td>
<td>65.5</td>
<td>10.3</td>
<td>67.5</td>
<td>31</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>Cyclosporin alone</td>
<td>24</td>
<td>25</td>
<td>18</td>
<td>6</td>
<td>110</td>
<td>403</td>
<td>20.8</td>
<td>20.8</td>
<td>36.6</td>
<td>58.3</td>
<td>41.6</td>
<td>—</td>
</tr>
<tr>
<td>Androgen</td>
<td>15</td>
<td>24</td>
<td>9</td>
<td>6</td>
<td>71</td>
<td>570</td>
<td>20</td>
<td>0</td>
<td>16</td>
<td>80</td>
<td>20</td>
<td>—</td>
</tr>
</tbody>
</table>

*D = day; BMT = bone marrow transplantation.

BMT was 11 units (0 – 100) and the mean number of transfused platelets before BMT was 16 units (0 – 100). In this group, the mean number of mononuclear cells (MNC) for transplantation was \(5.66 \times 10^8/\text{kg} \ (2.2 – 11.25)\). Of 29 patients of BMT group, 17 patients had received bone marrow stem cells and 12 patients had received peripheral blood stem cells transplantations. The patients were followed-up for between 24 to 2,750 days (mean 878 days). At the time of study 20 patients (69%) were still alive but 9 patients (31%) had died. The cause of death in 2 cases (22.2%) was GVHD, in 6 cases (66.6%) infection, and in one case due to no response. The overall response rate in this group was 75.9% (65.5% complete response, 10.3% partial response, and 24.1% no response).

As a complication of BMT, acute GVHD developed in 21 patients (72.4%). Data about acute GVHD are presented in Table 3. The mean time of GVHD development was day +10.5 (+2 to +46). Five years overall survival in BMT group was 67% and 5-year disease free survival was 67%. Eight patients (27.6%) experienced recurrence.

Table 2. Probable etiology of severe aplastic anemia.

<table>
<thead>
<tr>
<th>Group</th>
<th>Idiopathic</th>
<th>Chemicals</th>
<th>Hepatitis</th>
<th>Postpartum</th>
</tr>
</thead>
<tbody>
<tr>
<td>BMT</td>
<td>79.3</td>
<td>13.7</td>
<td>3.4</td>
<td>3.4</td>
</tr>
<tr>
<td>Cyclosporin alone</td>
<td>79.1</td>
<td>16.6</td>
<td>4.1</td>
<td>—</td>
</tr>
<tr>
<td>Androgen</td>
<td>93.3</td>
<td>6.7</td>
<td>—</td>
<td>—</td>
</tr>
</tbody>
</table>

BMT = bone marrow transplantation.

In the cyclosporin alone group, 24 patients had been treated with cyclosporin alone and 2 patients with cyclosporin plus ALG. The mean time of cyclosporin consumption was 10 months (2 – 28). The median time of response was 11.9 months (3 – 18). Complications of cyclosporin are presented in Table 4. Five years overall survival in cyclosporin alone group was 36.6%. There were 2 patients who had received cyclosporin plus ALG and at the time of study both of them were alive and both had partial responses with good general conditions and no more need for transfusion.

Among cases patients with moderate disease who had received androgens (oxymetholone or danazol) plus prednisolone. Complication of androgen therapy was cholestasis due to oxymetholone seen in one case. This led to drug interruption.

The comparison between 5 years overall survival in BMT group (67.5%) and cyclosporin alone group (36.6%) shows that despite a prominent difference, this is not statistically significant \((p = 0.30)\).

Five years overall survival in BMT group was 67.5% and in non-BMT group was 31% \((p = 0.1)\). This difference is not significant but when this is viewed according to the follow-up time after each treatment—groups divide to first 200 days and after +200 days—we can see that survival in non-BMT group between the days 0 to +200 is higher than BMT.

Table 3. Incidence of acute GVHD (grading and sites).

<table>
<thead>
<tr>
<th>Grade</th>
<th>Percentage</th>
<th>Site</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>24.1</td>
<td>Skin</td>
<td>65.5</td>
</tr>
<tr>
<td>II</td>
<td>27.6</td>
<td>GI</td>
<td>51.7</td>
</tr>
<tr>
<td>III</td>
<td>20.7</td>
<td>Liver</td>
<td>17.2</td>
</tr>
<tr>
<td>Total</td>
<td>72.4</td>
<td>—</td>
<td>—</td>
</tr>
</tbody>
</table>

GI = gastrointestinal.
group \((p = 0.0007)\) (Figure 1). After the day +200 survival is higher in BMT group than non-BMT group \((p = 0.02)\) (Figure 2).

Since the number of infused mononuclear cells (MNC) in stem cell transplantation may have a result on response rate, BMT patients were divided into two groups according to the number of infused MNC \(\times 10^8/\text{kg}\). The survival rate in the group of \(2 - 5 \times 10^8/\text{kg}\) was 62\% and in the group of \(> 5 \times 10^8/\text{kg}\) was 75\%. This difference was not statistically significant \((p = 0.4)\).

Considering age as a prognostic factor, patients in BMT group and non-BMT group were divided into three age groups: 0 – 19 years, 20 – 49 years, and > 50 years. In 0 – 19 years of non-BMT group (16 patients) survival was 18\% and in 20 – 49 years (22 patients) survival was 42\% \((p = 0.2)\). In BMT group in 0 – 19 years (14 patients) the overall survival was 62\% and in 20 – 49 years (15 patients) it was 71\%. This difference was also not significant \((p = 0.3)\). In non-BMT group 5 years survival in cyclosporin alone group was 36.6\% and in androgen group was 16\%, showing a significant difference \((p = 0.03)\) and highlighting a preference for immunosuppressive therapy rather than androgen therapy in the treatment of severe aplastic anemia (Figure 3).

Thirty-five percent of patients within the non-BMT group had undergone splenectomy and 65\% had not had this procedure. Splenectomy in non-BMT group with \(p = 0.38\) had no effect on survival rate in this group.

**Table 4.** Complications in cyclosporine alone group.

<table>
<thead>
<tr>
<th>Complication</th>
<th>Number</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Rise of creatinine</td>
<td>1</td>
<td>4.2</td>
</tr>
<tr>
<td>Gingival hypertrophy, skin hyperpigmentation</td>
<td>1</td>
<td>4.2</td>
</tr>
<tr>
<td>Gingival hypertrophy</td>
<td>1</td>
<td>4.2</td>
</tr>
<tr>
<td>Gynecomastia gingival hypertrophy</td>
<td>1</td>
<td>4.2</td>
</tr>
<tr>
<td>Headache, vomiting</td>
<td>1</td>
<td>4.2</td>
</tr>
<tr>
<td>Seizure</td>
<td>1</td>
<td>4.2</td>
</tr>
<tr>
<td>Skin-gingival hyperpigmentation</td>
<td>1</td>
<td>4.2</td>
</tr>
<tr>
<td>Skin hyperpigmentation</td>
<td>1</td>
<td>4.2</td>
</tr>
<tr>
<td>Total</td>
<td>8</td>
<td>33.6</td>
</tr>
</tbody>
</table>
BMT versus immunosuppressive therapy in SAA between years 1990 – 2001

The overall survival in BMT group who presented with GVHD was 70% and without GVHD was 60%. This difference was not significantly different ($p = 0.63$).

**Discussion**

SAA is a rare disease that, when treated with supportive care including blood transfusions and antibiotics, only 25 – 30% of patients survive for 2 years. More than 50% are expected to die within 6 months of diagnosis without specific therapy. Effective treatment for SAA consists of either BMT or immunosuppressive therapy with agents such as ALG and cyclosporin. For almost three decades, patients with SAA have been treated successfully by HLA-identical BMT. Outcome has improved from about 45% survival in early studies to 65 – 90% in most recent trials. Improvement is due in part to a decreased incidence of graft rejection, resulting from the judicious use of transfusions before BMT, the removal of sensitizing leukocytes from transfusion products, and a decrease in the incidence and severity of acute GVHD prophylaxis e.g. use of cyclosporine. Comparison

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**Figure 2.** Survival in BMT and non-BMT after the day +200 ($p = 0.02$).

**Figure 3.** Survival in cyclosporin alone versus androgen therapy groups ($p = 0.03$)
of the overall survival between our BMT group and non-BMT group in first 200 days and post 200 days after each treatment. This comparison shows that BMT has the best results and patients in BMT group have better long-term survival (p = 0.02).

Bone marrow and more recently stem cell transplantation from a histocompatible sibling usually cure the underlying bone marrow failure. Mortality rates for first 200 days after transplantation have decreased probably as a result of less graft rejection and improved control of infections.

Cyclophosphamide and ALG have been used as a conditioning regimen for patients receiving transplants since 1988.6, 8, 28 Since our BMT group patients had also received cyclophosphamide and ALG, according to our 75.9% response rate and 5 years overall survival of 67%, we can conclude that our conditioning regimen was appropriate in comparison with reports from other countries.

Despite the use of prophylaxis, GVHD continues to be a problem and has a strong adverse effect on survival.10, 29 Reduction of acute GVHD has resulted in improved survival.9, 10, 23 According to our study, despite the difference between survival in GVHD-developed (70%) and non-developed patients (60%), it was not significant (p = 0.63).

GVHD, the frequency and severity of which correlate with patient’s age, continues to limit the success of transplantation. In most analysis, GVHD also contributed to earlier death.2

The dose of mononuclear cells (MNC) infused is very important in SAA. In contrast to our results about survival according to number of MNC infused at the time of BMT, efforts should still be directed at maximizing the number of marrow cells infused by harvesting ≥ 3.5 × 10⁸ cell/kg.30, 31 There is an increased risk of graft failure seen in patients who receive low doses of marrow cells.

The comparison between cyclosporine therapy and androgen therapy in treatment of SAA showed preference of immunosuppression as a better treatment for SAA (p = 0.03) that increases the survival of these patients. Regarding age as a factor that may interfere in overall survival of patients, we did not find that age could have an impact on survival (p = 0.3). This finding may be due to our limited number of patients.

Despite the vast studies 4, 13 – 17 conducted on immunosuppressive therapy with ALG and cyclosporine, it is difficult to conclude firm results due to the small number of our patients who received this treatment (2 patients). Prior blood transfusion is the major risk factor for survival following BMT for SAA. Evidence indicates that dendritic cells in blood products are responsible for sensitization of patient and graft failure.32, 30 Un-transfused patients have a lower risk of graft failure than transfused recipients and the risk increases with larger numbers of transfusions. It seems that, regarding the mean number of blood products transfused to our patients, nearly all of our patients were high risk for transplantation, therefore our results were satisfactory in comparison with other places of the world.

Finally, bone marrow transplantation is curative for SAA patients who have histocompatible donor and are aged younger, and for those SAA patients who do not have a match donor, immunosuppressive therapy is preferred.

Acknowledgment

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References

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