B cell lymphoproliferative disorders following hematopoietic stem cell transplantation: risk factors, treatment and outcome

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Summary:

Twenty-six cases of B cell lymphoproliferative disorder (BLPD) were identified among 2395 patients following hematopoietic stem cell transplants (HSCT) for which an overall incidence of BLPD was 1.2%. The true incidence was probably higher, since 9/26 of the diagnoses were made at autopsy. No BLPD was observed following autologous HSCT, so risk factor analyses were confined to the 1542 allogeneic HSCT. Factors assessed were HLA-mismatching (>1 antigen), T cell depletion (TCD), presence of acute GvHD (grades II–IV), donor type (related vs unrelated), age of recipient and donor, and underlying disease. Factors found to be statistically significant included patients transplanted for immune deficiency and CML, donor age >18 years, TCD, and HLA-mismatching, with recipients of combined TCD and HLA-mismatched grafts having the highest incidence. Factors found to be statistically significant in a multiple regression analysis were TCD, donor age and immune deficiency, although 7/8 of the patients with immunodeficiencies and BLPD received a TCD graft from a haploidentical parent. The overall mortality was 92% (24/26). One patient had a spontaneous remission, but subsequently died >1 year later of chronic GVHD. Thirteen patients received therapy for BLPD. Three patients received lymphocyte infusions without response. The only patients with responses and long-term survival received alpha interferon (αIFN). Of seven patients treated with αIFN there were four responses (one partial and three complete). These data demonstrate that αIFN can be an effective agent against BLPD following HSCT, if a timely diagnosis is made.

Keywords: EBV; lymphoproliferative disease; risk factors; outcome

B cell lymphoproliferative disorders (BLPD) represent a spectrum of clinically and morphologically heterogeneous lymphoid proliferations, that are almost invariably found associated with T cell dysfunction and the presence of Epstein–Barr virus (EBV).1–4 The increased risk of EBV-associated BLPD in patients with T cell dysfunction, both primary and secondary, is well-recognized, but the pathogenesis of BLPD in the immunocompromised host has not been fully characterized.1–4 EBV, a member of the herpesvirus family, infects and immortalizes human B lymphocytes in vitro and in vivo, establishing lifelong viral latency in peripheral blood lymphocytes in healthy EBV-seropositive individuals.5 Following primary infection in the immunocompetent host, proliferation of EBV-infected B cells is controlled by a multitude of immune mechanisms including virus-specific cytotoxic T cells, humoral responses, antibody-dependent cellular cytotoxicity, natural killer activity, and cytokine regulation.6–11 All of these functions may be temporarily compromised following lymphohematopoietic transplantation.

BLPD is a highly lethal complication of allogeneic bone marrow transplantation (BMT).4,11–16 In previous analyses, BLPD has been linked with recipient-donor HLA incompatibility, T cell depletion (TCD), and severe GVHD, especially when treated with anti-T cell immunotherapy (anti-thymocyte globulin or monoclonal antibodies).11–21 Surprisingly, despite intensive immunosuppression from cytoreductive regimens and the use of immunosuppressive agents to prevent/or treat GVHD, BLPD is relatively infrequent following HLA-identical related BMT.1,12,13 During the past 15 years, the use of alternative donors, i.e., partially mismatched related or unrelated donors, has been greatly expanded. Transplants with grafts from alternative donor sources are complicated by higher rates of graft rejection and GVHD compared with matched sibling BMT.14,19,20 Additionally, relative delays in immune reconstitution may contribute to an increased risk of fatal opportunistic infections following HSCT with alternative donor grafts when compared with transplants with grafts from an histocompatible sibling, especially in the setting of TCD of the donor graft.22–26 This delayed immune recovery may also partially account for the apparent recent increase in the diagnosis of EBV-associated BLPD following HSCT.

In this report, 26 cases of BLPD following hematopoietic stem cell transplantation (HSCT) were analyzed. The purpose of this communication is to: (1) highlight the most significant risk factors for development of post-transplant BLPD in a diverse population of HSCT recipients, and (2)
summarize the results using α-interferon (αIFN) to treat EBV-associated BLPD in a subset of patients post-HSCT.

Patients and methods

Patients

Between March 1974 and May 1996, 2395 hematopoietic stem cell transplants (HSCT) were performed at the University of Minnesota. A review of the clinical research database for all cases of new malignancy following HSCT identified 26 cases of BLPD. Characteristics and analyses of some of these cases have been previously reported.12,27 BLPD data were integrated with other clinical and demographic data from the Bone Marrow Transplant Database, which contains systematically and prospectively collected data on all patients receiving a bone marrow transplant at the University of Minnesota. All patients were advised of procedures and attendant risks in accordance with institutional guidelines and gave informed consent.

BLPD diagnosis

The diagnosis of BLPD was made in fulfillment of the histologic criteria developed for B cell lymphoproliferative processes following transplantation.2 Clonality of BLPD was determined by immunophenotyping of tumor cells for immunoglobulin light chain restriction and/or identification of clonal immunoglobulin heavy chain (IgH) gene rearrangement in DNA extracted from diagnostic material. The presence of EBV was determined either by in situ hybridization and/or Southern blot analysis as previously described.12,28

Statistical analysis

Estimation of the incidence of BLPD was calculated by Kaplan–Meier estimation. Time to BLPD was measured from the date of transplant and patients were censored at the time of death or at 6 months post-HCST. Three patients developed BLPD greater than 6 months post-transplant. Univariate comparisons were completed with 95% confidence intervals and the Mantel–Cox log-rank statistic. Multiple regression analysis was performed using the Cox proportional hazards regression model.

Since no cases of BLPD were pathologically confirmed following autologus HSCT, recipients of autografts were excluded from risk factor analysis. Factors considered as potential predictors for the development of BLPD among allogeneic patients were HLA mismatching (defined as 1 or more antigen disparity for HLA A, B, and/or DRB1 loci), T cell depletion (TCD), donor type (related vs unrelated), age of recipients and donor, underlying disease, and acute GVHD (grades II–IV). Acute GVHD was assessed as a time-confounded risk factor. The interaction between T cell depletion and HLA disparity was also explored.

Results

Hematopoietic stem cell transplantation

During the time period under study, 2395 HSCT were performed: 853 were autografts and 1542 were allografts. Of the 1542 allografts, 1112 received grafts from related donors and 430 from unrelated donors. Recipients were HLA identical (HLA-A, B and DRB1 loci) in 1206 of the allogeneic HSCT, and in 315 recipient:donor disparity for at least one HLA locus was recorded. Ex vivo depletion of T cells in donor graft was performed in 247 HSCT (170 were HLA identical and 76 HLA disparate). There were 1295 allografts transplanted without TCD (1036 were HLA identical and 239 HLA disparate).

BLPD cases

Characteristics of the 26 patients with BLPD compared to patients who did not develop BLPD following allogeneic HSCT are summarized in Table 1. There were no differences between the groups for year of transplant, age, gender or preparative regimen, although there was a trend toward more BLPD in patients receiving chemotherapy only as preparative regimen.

Table 2 is a more detailed summary of the 26 patients with BLPD and their HSCT. The median time from HSCT to diagnosis of BLPD was 86 days (range 30–1183 days). The diagnosis was made at autopsy in nine patients. Primary diagnosis which brought patients to HSCT included 14 with leukemia, eight with primary immunodeficiency, two with metabolic disorders, one with Hodgkin’s disease and one with severe aplastic anemia. Sixteen had related donors and 10 unrelated donors. Fourteen received grafts that were known to be mismatched at one or more HLA locus, 11 related and three unrelated donors. The remaining 12 patients received grafts that were phenotypically matched at HLA-A, B and DRB1 loci, five related and seven unrelated donors. Sixteen received TCD grafts, 11 also had HLA-mismatched grafts. Sixteen patients developed GVHD (grades II–IV), nine of whom experi-
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Table 2
Characteristics of BLPD patients and HSCT

| UPN | Age/Sex | Diagnosis | Donor | HLA match | Recipient:Donor | Preparative regimen | GVHD prophylaxis | GVHD |
|-----|---------|-----------|-------|-----------|-----------------|---------------------|------------------|------|
| 190 | 1/F     | SCID      | Parent | Mismatched | CY/TBI          | TCD (1)             | None             |      |
| 309 | 0.6/M   | SCID      | Parent | Mismatched | CY/TBI          | TCD (1)             | None             |      |
| 332 | 7/M     | WAS       | Parent | Mismatched | CY/TBI/ATG      | TCD (2)             | None             |      |
| 469 | 1/F     | AML       | Parent | Mismatched | AraC/TBI        | TCD (2)             | None             |      |
| 473 | 16/F    | AML       | Sibling| Matched   | CY/TBI          | MAP                | I                |      |
| 541 | 36/M    | CML       | Sibling| Matched   | CY/TBI          | TCD (2)             | II               |      |
| 588 | 12/M    | CML       | Parent | Mismatched | CY/TBI          | TCD (2)             | II               |      |
| 596 | 30/F    | CML       | Unrelated| Matched | CY/TBI          | MAP                | II               |      |
| 600 | 50/M    | CML       | Sibling| Mismatched | CY/TBI          | TCD (2)             | III              |      |
| 864a| 15/F    | SAA       | Parent | Mismatched | BU/CY           | TCD (2)             | None             |      |
| 953 | 1/M     | SCID      | Parent | Mismatched | BU/CY           | TCD (2)             | None             |      |
| 1297a| 22/F    | HD        | Sibling| Matched   | BU/CY           | MAP                | III              |      |
| 1333| 1/M     | SCID      | Unrelated| Mismatched | BU/CY/ATG      | TCD (2)             | IV               |      |
| 1362a| 5/F     | CHS       | Parent | Mismatched | CY/Vp/TBI      | TCD (2)             | II               |      |
| 1405| 5/F     | CML       | Unrelated| Matched   | CY/TBI          | MC                 | IV               |      |
| 1409a| 13/M    | CML       | Unrelated| Mismatched | CY/TBI          | MC                 | IV               |      |
| 1502a| 37/F    | CML       | Unrelated| Matched   | CY/TBI          | MC                 | III              |      |
| 1511| 51/M    | CML       | Sibling| Matched   | CY/TBI          | MAP                | II               |      |
| 1521| 1/M     | SCID      | Parent | Mismatched | BU/CY/ATG      | TCD (2)             | None             |      |
| 1529| 0.7/M   | SCID      | Parent | Mismatched | BU/CY/ATG      | TCD (2)             | None             |      |
| 1595| 44/M    | CML       | Sibling| Matched   | CY/TBI          | MAP                | III              |      |
| 1783a| 1/M     | Hurlers   | Unrelated| Mismatched | BU/CY           | TCD (3)             | None             |      |
| 2241a| 46/F    | ALL       | Unrelated| Matched   | CY/TBI          | TCD (3)             | III              |      |
| 2271a| 1/M     | Globoid   | Unrelated| Matched   | BU/CY/ATG      | TCD (3)             | None             |      |
| 2320a| 34/M    | CML       | Unrelated| Matched   | CY/TBI          | TCD (3)             | III              |      |
| 2370| 13/F    | CML       | Unrelated| Matched   | CY/TBI          | TCD (3)             | III              |      |

-Diagnosis made at autopsy.

SCID = severe combined immunodeficiency; WAS = Wiskott–Aldrich syndrome; CHS = Chediak–Higashi syndrome; SAA = severe aplastic anemia; CY = cyclophosphamide; BU = busulfan; TBI = total body irradiation; ATG = anti-thymocyte globulin; TCD (1) = soybean agglutination and erythrocyte rosetting; TCD (2) = immunotoxin (ricin)-conjugated monoclonal antibodies; TCD (3) = counterflow elutriation; MAP = methotrexate, ATG and prednisone; MC = methotrexate and cyclosporine.

enced severe GVHD (grades III–IV). Nine patients with BLPD had no acute GVHD.

The presence of EBV was confirmed in diagnostic material in all cases tested (20/20). Tumor cloning was studied in 17 cases. Fifteen cases were found to be monoclonal and in one case the immunohistochemical studies suggested polyclonal disease but IgH gene rearrangement studies were indicative of monoclonal disease (data not shown). Patient No. 1333 with spontaneous remission had documented monoclonal disease. Patient No. 309 with polyclonal disease died of progressive BLPD, in spite of lymphocyte infusions, corticosteroids and intravenous acyclovir.

Incidence of BLPD

The majority of BLPD occurs within 5–6 months post-BMT. In patients UPN 190, 332 and 1783, BLPD developed 484, 1183 and 280 days, respectively, after the grafting procedures. Two of these patients (UPN 190 and 332) were at a heightened risk of EBV-related BLPD independent of HSCT due to their diagnosis of primary immunodeficiency. UPN 332 rejected his graft outright, while UPN 190 showed declining donor engraftment at the time of BLPD diagnosis. Late onset BLPD, ie >6 months following HSCT, has been associated with patients treated for primary immunodeficiency and autologous hematopoietic recovery. In this study, risk factor analysis was restricted to 6 months post-HSCT, and the above mentioned three cases were not included in the analyses (Table 3).

The incidence of BLPD in the allogeneic setting was 2.0% (1.2–2.8%) (Table 3). Patients with unrelated donors had an incidence of 3.3% (1.3–5.3%) compared with 1.6% (0.8–2.4%) for related donors, but this difference was not significant. Recipient age (<18 years vs ≥18 years) had no effect on BLPD incidence; however, donor age (related donors only) had a significant effect, 0.3% incidence for donor age ≤18 years vs 2.7% for donors ≥18 years (P < 0.01). Donor age as a risk factor among URD recipients was not examined in univariate analysis due to the small number of events in this group and the fact that donors are adults only. Patients receiving HSCT for CML (3.5%, 1.5–5.5%) and primary immune deficiency (8.1%, 4.9–14.3%) had a statistically significant higher incidence of BLPD than other underlying diagnoses. The incidence of BLPD was higher for patients receiving grafts depleted of mature T cells (TCD) ex vivo compared to recipients of non-TCD marrow (7.2% vs 1.3%, P < 0.01). Recipients of HLA-mismatched grafts had a higher incidence than HLA-matched recipients (5.3% vs 1.3%, P < 0.01). When both TCD and HLA mismatching coincided the actuarial incidence of BLPD by 6 months rose to 15.7% (6.1–41.3%).
Table 3  Factors affecting actuarial risk of BLPD at 6 months following HSCT

| Factor                                      | No. of BLPD cases | Risk of BLPD (%) | 95% Confidence interval | P value |
|---------------------------------------------|-------------------|------------------|-------------------------|---------|
| Total                                       | 2395              | 23               | 1.2                     | 0.7–1.7            |
| Autologous                                  | 853               | 0                | 0.0                     | —           |
| Allogeneic                                  | 1542              | 23               | 2.0                     | 1.2–1.8            |
| Donor type                                  |                   |                  |                         |         |
| Related donor                               | 1112              | 14               | 1.6                     | 0.8–2.4            | 0.13    |
| Unrelated donor                             | 430               | 9                | 3.3                     | 1.3–5.3            |         |
| Recipient age                               |                   |                  |                         |         |
| <18 years                                   | 763               | 14               | 2.3                     | 1.1–2.5            | >0.20   |
| >18 years                                   | 779               | 9                | 1.8                     | 0.7–2.9            |         |
| Donor age (related donors only)             |                   |                  |                         |         |
| <18 years                                   | 454               | 1                | 0.3                     | 0.0–0.8            | <0.01   |
| >18 years                                   | 658               | 13               | 2.7                     | 1.2–4.2            |         |
| Underlying disease                          |                   |                  |                         |         |
| Severe aplastic anemia                      | 164               | 1                | 0.8                     | 0.3–1.7            | <0.01   |
| Immune deficiency                           | 104               | 6                | 8.1                     | 1.9–14.3           |         |
| Metabolic disease                           | 108               | 1                | 0.1                     | 0.0–2.5            |         |
| ALL                                         | 276               | 2                | 1.0                     | 0.8–2.2            |         |
| AML                                         | 337               | 2                | 0.8                     | 0.8–2.2            |         |
| CML                                         | 394               | 10               | 3.5                     | 1.5–5.5            |         |
| Other malignancies                          | 159               | 1                | 0.9                     | 0.0–2.7            |         |
| Acute GVHD (grades II–IV)                   |                   |                  |                         |         |
| No                                          | 834               | 8                | 1.3                     | 0.3–2.3            | 0.09    |
| Yes                                         | 708               | 15               | 2.8                     | 1.4–4.2            |         |
| Recipient:Donor disparity                   |                   |                  |                         |         |
| HLA matched                                 | 1206              | 12               | 1.3                     | 0.6–2.0            | <0.01   |
| HLA mismatched                              | 315               | 11               | 5.3                     | 2.3–8.3            |         |
| T cell depletion (TCD)                      |                   |                  |                         |         |
| No TCD                                      | 1295              | 10               | 1.1                     | 0.5–1.7            | <0.01   |
| TCD                                         | 247               | 13               | 7.2                     | 3.5–10.9           |         |
| TCD and HLA disparity                       |                   |                  |                         |         |
| HLA matched/No TCD                         | 1036              | 8                | 1.0                     | 0.3–1.7            | <0.01   |
| HLA matched/TCD                            | 170               | 4                | 3.4                     | 0.0–5.1            |         |
| HLA mismatched/No TCD                      | 239               | 2                | 1.5                     | 0.0–3.5            |         |
| HLA mismatched/TCD                         | 76                | 9                | 15.7                    | 6.1–41.3           |         |

Table 4  Multivariate analysis of risk factors for BLPD following HSCT

| Factor                      | Relative risk | 95% Confidence interval | P value |
|-----------------------------|---------------|--------------------------|---------|
| HLA mismatching             | 2.2           | 0.8–6.1                  | 0.13    |
| T cell depletion            | 5.4           | 2.3–12.7                 | <0.01   |
| Immune deficiency           | 3.8           | 1.3–11.4                 | 0.01    |
| Donor age (continuous)      | 1.04          | 1.01–1.08                | 0.01    |
| Unrelated donor             | 0.8           | 0.3–2.1                  | >0.20   |

Multivariate analysis was performed and identified TCD, relative risk (RR) = 5.4, immune deficiency (RR = 3.8) and donor age (RR = 1.04 per year of age) as the only statistically significant variables (Table 4). HLA mismatch had a RR of 2.2 but was not statistically significant (P = 0.13).

Treatment and outcome

Overall mortality was 92% (24/26). Twenty-one of the deaths were directly attributable to BLPD. Other infections present at time of death included: bacterial (seven), cytomegalovirus (three), adenovirus (two) and fungus (four). One child experienced spontaneous resolution of BLPD (UPN 1333), but eventually died of infections secondary to chronic GVHD more than 1 year later. Thirteen patients were not specifically treated for BLPD; nine of these cases were incidentally diagnosed at autopsy (Table 2).

Thirteen patients received a variety of therapies after diagnosis of BLPD. Nine patients received acyclovir intravenously as prophylaxis (prior to BLPD diagnosis) and/or as treatment. Seven of these nine patients also received weekly intravenous immunoglobulin (IVIG). Three patients were treated with intravenous corticosteroids, one of these patients was additionally treated with anti-lymphocyte globulin, and later chemotherapy, ie cyclophosphamide and vincristine. Three patients received lymphocyte...
infusions (two had donor lymphocytes, and one received lymphocytes from an EBV-seropositive parent who was not the original bone marrow donor). None of the above-mentioned treatments resulted in objective improvements of BLPD. The only responses to therapy were observed in patients treated with αIFN.

Seven patients were treated with αIFN (Table 5). Three (43%) of these individuals achieved complete clinical remission (CR) of their BLPD. Of these three, two are long-term survivors, and one patient died of aspergillosis without evidence of BLPD at autopsy. One additional patient (UPN 953), a boy with SCID who had rejected a maternal TCD graft, showed significant improvement of symptoms attributed to BLPD, including lysis of fever and remarkable decrease in diffuse lung nodules, but eventually experienced progression of BLPD coincident with recrudescence of adenoviral infection. One patient’s disease did not progress, but he eventually died with BLPD. Two patients died of hemorrhage within a few days after institution of αIFN therapy, and response to this treatment was not evaluable. Progression of GVHD was not observed in any patient treated with αIFN.

Discussion

In this report, 26 cases of BLPD following allogeneic HSCT were identified and analyzed. In all cases tested for the presence of EBV, the association was confirmed. The estimated overall incidence of BLPD following allogeneic HSCT was 2.0%. In nine cases the cause of death was felt to be multi-system organ failure attributed to overwhelming sepsis and/or GVHD, but determined at autopsy to be disseminated BLPD. Since over a third of BLPD cases in this series were diagnosed post-mortem and fewer than half of the patients who died underwent post-mortem examination, it is reasonable to assume that the true incidence of BLPD is higher than our estimate. The published incidence of BLPD following BMT varies between series, 0.6–10%.4,12–16 These differences may reflect differences in the proportions of ‘high-risk’ patient groups, and/or differences in criteria for diagnosis. The latter possibility is not trivial, since the wide spectrum of clinical and histologic presentations of these disorders can make diagnosis difficult. For example, a non-lethal ‘infectious mononucleosis-like’ syndrome, as in UPN 1333, may resolve spontaneously; while disseminated disease, as in the nine cases diagnosed at autopsy, may go unrecognized as BLPD. Therefore, a high index of suspicion along with an aggressive diagnostic process involving imaging studies and biopsies may increase the number of pre-mortem BLPD diagnoses.

Factors associated with increased risk of BLPD identified in other reports include recipient–donor HLA incompatibility and T cell depletion (TCD).4,12–18,21 The results of our analysis support those findings. Even in the absence of TCD, the use of intense immunosuppressive prophylaxis and/or therapy of GVHD, especially anti-T cell agents such as OKT3 or anti-thymocyte globulin have been associated with the development of BLPD.11–18,20 Sixteen patients developed GVHD (grades II–IV), nine of whom experienced severe GVHD (grades III–IV). However, the presence of acute GVHD in this mixed population of children and adults was not found to have significant influence on the development of BLPD in our analysis. The role of HLA mismatching in the pathogenesis of BLPD is not clear. However, it has been hypothesized that mismatched grafts may be a source of chronic antigenic stimulation,12 or delayed immune reconstitution. In our cohort, mismatching at ≥1 HLA-A, B, DRβ1 locus was found to be significant in univariate analysis, but was not found to have a significant effect separate from TCD (Tables 3 and 4). Since many of the HSCT in this report were undertaken before the era of precise molecular HLA typing, analyses were not performed to determine if the degree of mismatching influenced the risk of developing BLPD, i.e. haploidentical vs 2 or 1 antigen disparity. Clearly, antigenic disparity exists in all unrelated donor transplants. The National Marrow Donor Program (NMDP) reported the incidence of BLPD in unrelated donor BMT to be 2% overall, 5% with TCD and 1% with no TCD.19 In our cohort, the overall incidence of BLPD recipients of unrelated donor HSCT was 3.3%. Although this incidence is higher than observed in recipients of related donors, the difference was not found to be statistically significant (Tables 3 and 4). The incidence in recipients of TCD vs no TCD unrelated donor grafts was not evaluated due to small numbers of TCD unrelated donor grafts.

Older donor age emerged as a significant risk factor in both the univariate and multivariate analyses (Tables 3 and 4). The multivariate analysis demonstrated that the incidence of BLPD increases with donor age, i.e. approximately 4% per year (Table 4). This effect does not appear to be attributable to the incidence of GVHD with increased donor age, since this was found to be an independent risk factor.

### Table 5: Patients treated with αIFN for BLPD following HSCT

| UPN | Clonality | Therapy | Response | Outcome (days after treatment) |
|-----|-----------|---------|----------|--------------------------------|
| 541 | Monoclonal | ACV, DLI, αIFN | SD | Died w/BLPD (36 days) |
| 596 | Monoclonal | ACV, αIFN | CR | Alive w/o BLPD (3682+ days) |
| 953 | Monoclonal | αIFN, IVIG | PR/PD | Died w/BLPD (171 days) (Adenovirus) |
| 1405 | Monoclonal | αIFN | NE | Died w/BLPD (8 days) (hemorrhage) |
| 1511 | Monoclonal | αIFN, ACV, IVIG | CR | Died w/o BLPD (20 days) (Aspergillosis) |
| 1521 | Monoclonal | αIFN | NE | Died w/BLPD (8 days) (hemorrhage) |
| 1529 | Monoclonal | αIFN | CR | Alive w/o BLPD (1503+ days) |

ACV = acyclovir; DLI = infusion of donor lymphocytes; IVIG = intravenous immunoglobulin; αIFN = α-interferon; NR = no response; SD = stable disease; CR = complete response; PR = partial response; PD = progressive disease; NE = not evaluable for response.
Possible explanations include the probability of delayed or poorer immune reconstitution following HSCT from older donors, and may reflect a higher percentage of EBV transmission from adult donors.\(^4\)\(^,\)\(^12\)\(^,\)\(^13\)\(^,\)\(^30\)\(^,\)\(^32\)

The association between T cell depletion and BLPD has been repeatedly cited, often drawing attention to the methods and extent of TCD.\(^12\)\(^,\)\(^15\)\(^,\)\(^19\) TCD methods in this series varied. Two were pan T cell depletion methods, ie soybean agglutination and erythrocyte rosetting (SBAE\(^-\)) and ricin-conjugated monoclonal antibodies, which yield in excess of 2 logs of T cell depletion. In recent years, counterflow elutriation was used. Elutriation is a physical method of producing a cellular fraction that is enriched for mature and immature hematopoietic progenitor cells, but depleted of the majority of T, B, NK cells and monocytes yielding a product that is 1.5–2.0 log T cell depleted.\(^32\)\(^,\)\(^34\)\(^,\)\(^35\) Due to small numbers, an analysis comparing the effect of different TCD methods was not performed.

In the univariate analysis (Table 3), patients receiving HSCT for CML and immune deficiency had a higher incidence of BLPD, 3.5% and 8.1%, respectively, and immune deficiency was found to be a significant factor on multivariate analysis, RR = 3.8 (Table 4). It is well recognized that patients with immune deficiency, both acquired and inherited, are at increased risk of developing an EBV-associated B cell lymphoproliferative process outside the context of HSCT.\(^1\)\(^–\)\(^4\) In two immune deficient patients who developed BLPD post-HSCT (UPN 190 and 332), BLPD developed following autologous hematopoietic recovery. Therefore, it is difficult in such cases to delineate the influence of HSCT vs uncorrected immunodeficiency in the development of BLPD. It has been reported that the incidence of BLPD in HSCT for immunodeficiency is 32% for TCD, haploidentical HSCT, 8% with unrelated donors and 0% using a graft from a matched sibling.\(^36\) Since seven of the eight patients with immune deficiency and BLPD received a TCD graft from a haploidentical parent, association with TCD and mismatching becomes a significant confounding factor.

Overall mortality in this cohort was 92% (24/26), and 21 (88%) of the deaths were directly attributable to BLPD, similar to that found by Zutter et al;\(^11\) 93% deaths overall, 87% attributed to BLPD. Unlike other post-transplant secondary malignancies, such as solid tumors and myelodysplastic syndrome, BLPD does not arise as a late effect of chemotherapy and/or radiation.\(^36\)\(^,\)\(^20\)\(^,\)\(^27\) Rather, the majority of BLPD cases are diagnosed in the first few months post-transplant when endogenous immunity is very primitive, especially with TCD grafts, representing, in essence, an opportunist infectious complication. Thus, the use of immune therapy has appeal as a first-line therapy for BLPD.

Anti-B cell monoclonal antibody therapy has proven useful in some cases of polyclonal BLPD, but not monoclonal disease.\(^35\) Delayed recovery of lymphocytes and EBV-specific cytotoxic T cells have been associated with development of BLPD,\(^18\)\(^,\)\(^25\) and infusion of donor leukocytes (DLI) has been demonstrated to be successful in the treatment of BLPD post-BMT.\(^1\) However, severe GVHD has also been associated with donor leukocyte infusions,\(^36\)\(^,\)\(^37\) and death due to a 'shock-like syndrome' and ARDS have been reported.\(^11\) Interestingly, three patients in this series received leukocytes (two from the donor, one from non-donor parent EBV immune) as therapy without a clinical response. Successful treatment of BLPD using αIFN has been reported anecdotally.\(^12\)\(^,\)\(^38\) Of the 13 patients who received therapy for BLPD in this series, the only objective responses to therapy were observed in patients treated with αIFN.

Of the seven patients who received αIFN (Table 4), four (57%) had a clinical response – one partial response and three (43%) complete responses. The mechanism of action for the anti-BLPD effect observed using αIFN is not known. However, it has been shown that patients newly diagnosed with BLPD demonstrate an imbalance of cytokines in their serum: increased levels of IL-4 (B cell proliferative stimulus) and relatively decreased, or undetectable levels of αIFN compared to healthy EBV-seropositive controls. In contrast, allograft recipients receiving cyclosporine as chronic immunosuppression also have elevated IL-4 serum concentrations, but αIFN levels are comparable to normal controls.\(^10\) Interestingly, the patient who experienced a spontaneous remission (UPN 1333) was the only patient studied, to date, whose peripheral blood mononuclear cells at the time of diagnosis demonstrated significant spontaneous production of αIFN in vitro (data not shown). Thus, we reasoned that treatment of BLPD patients with pharmacologic doses of αIFN might restore cytokine balance, modifying the milieu that had favored proliferation of EBV-transformed B cells. Since αIFN appears not to increase the risk of GVHD, it may be beneficial against BLPD post-HSCT in settings where donor leukocytes are unavailable or not promptly available, or where there is the presence or risk of severe GVHD.

Using CMV disease post-transplant as a paradigm, prevention/prophylaxis against infection and/or early detection with pre-emptive therapy may improve the outcome of BLPD post-HSCT. Indeed, it has been observed that EBV-seronegative recipients of organ allografts are at increased risk of BLPD.\(^39\) Screening of donors and recipients for EBV serostatus is not routinely performed prior to HSCT, therefore the effect of EBV serostatus on the development of BLPD is unknown. It has been claimed that antiviral prophylaxis with acyclovir prevents BLPD,\(^1\) but our experience does not support this. The majority of patients reported were receiving acyclovir prophylaxis for HSV and/or CMV at the time the BLPD occurred.

Pre-emptive therapy prophylaxis for BLPD post-HSCT is an attractive option. To make this approach practical and successful, it is necessary to first identify ‘at risk’ patients. Secondly, sensitive and reliable methods of detecting early EBV infection, viral reactivation, or early BLPD must be available. Semi-quantitative determinations of EBV DNA in peripheral blood, ie viral load, appear to correlate with BLPD development,\(^40\)\(^,\)\(^41\) and may be useful in following ‘high-risk’ patients. Finally, effective, readily available, cost effective and relatively non-toxic therapeutic interventions are desirable. In this context, the use of αIFN, low dose donor leukocytes or even EBV-specific T cells,\(^40\)\(^,\)\(^42\) deserve comparative scrutiny in the prevention and early treatment of EBV-associated BLPD.
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