

Peripheral Blood Stem Cell Transplantation for Rheumatoid Arthritis — Australian Experience

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ABSTRACT. Trials of high dose immunosuppression and peripheral blood stem cell transplantation (PBSCT) in patients with severe rheumatoid arthritis (RA) have now commenced based on encouraging data from case reports of patients with coexistent malignancy and animal transplant models. Early case reports in Australia documented the potential for cure of RA in most patients receiving allogeneic or syngeneic transplants. However, the relatively high morbidity and mortality of these procedures has necessitated the use of autologous PBSCT, in accordance with international guidelines released by the EBMT/EULAR working party. Phase 1 trials in autologous PBSCT have seen substantial remissions of RA in the majority of patients who had previously failed all available therapies. Recurrence of disease occurs in most patients usually within 2 years; however, the use of disease modifying agents after recurrence results in substantial amelioration of the disease, again suggesting a form of “immunomodulation.” This observation raises the possibility of maintenance therapy associated with procedure to prolong responses. Other modifications of the procedure are discussed, including T cell depletion of the graft, currently the subject of a randomized trial. (*J Rheumatol* 2001;28 Suppl 64:8–12)

Key Indexing Terms:

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INTRODUCTION

Trials of peripheral blood stem cell transplantation (PBSCT) in rheumatoid arthritis (RA) have been initiated on the basis of 3 clinical observations. First, it was noted that patients with RA and bone marrow failure could achieve remission of their RA following allogeneic bone marrow transplants¹. In addition, RA patients with coexistent malignancy had their disease greatly attenuated after autologous grafts². These observations coincided with the use of peripheral blood stem cells (PBSC) for transplant instead of marrow, resulting in significantly lower transplant related mortality predominantly on the basis of more rapid engraftment³. Third, epidemiological studies showed that RA was not just a benign disability but a disease with a significantly increased mortality and morbidity compared to the general population⁴.

Once these clinical facts were appreciated it was a logical progression to assess the feasibility of PBSCT in a severe, relatively common autoimmune disease such as RA. In Australia, the assessment of PBSCT as a therapy of RA has progressed in a step-wise process with initial successful and informative case studies. This was followed by phase 1 dose

escalation studies and now finally to a randomized trial. The ultimate goals of the studies are primarily safety followed by feasibility and efficacy.

We recount this step-wise process with particular reference to longterm followup of early patients and the role of CD34 selection of stem cell grafts in this disorder.

CASE REPORTS

Three patients with RA and gold induced aplastic anemia were reported to be in complete remission of their arthritis up to 13 years after allogeneic bone marrow transplants from HLA matched siblings⁵. One patient had a recurrence of disease at 2 years, and recommenced disease modifying antirheumatic drugs (DMARD) for a subsequent 2 years, after which the disease had an attenuated course. Complete donor chimerism was never established in this patient, in contrast to a case report by McKendry, *et al*⁶ that showed complete donor chimerism; however, the disease also recurred at 2 years. Animal studies in adjuvant arthritis have suggested that allogeneic transplants would be curative⁷ and this would seem logical in the human setting due to a combination of factors such as more intense immunosuppression pre and post-transplant and the lack of autoreactive cells (such as lymphocytes) in the donor graft. These two cases of recurrence post-allogeneic transplant^{5,6}, however, suggest that other factors must be involved, for instance the presence of a small degree of residual recipient “disease causing cells” not detectable by current methodology. Conversely, some donor cells may attain the ability to recognize a recipient antigen that is responsible for the disease process — until a putative “auto-antigen” is found this hypothesis would be difficult to test.

McColl, *et al*⁸ reported a syngeneic (identical twin)

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PBSCT following cyclophosphamide/antithymocyte globulin (ATG) conditioning in a 42-year-old man with severe seronegative RA. Post-transplant T cell receptor V-beta gene usage of circulating lymphocytes yielded evidence of full donor T cells that corresponded to complete remission of the disease, now at 36 months' followup (personal communication, J. Szer). This case appears to demonstrate the importance of full donor chimerism at the T cell level — a situation that may be a necessity for allogeneic PBSCT to be totally successful.

Given that allogeneic grafting is a procedure with significant morbidity and mortality, it was the initial consensus of all investigators in this field to perform autologous PBSCT for autoimmune diseases⁹. In the setting of RA, it was not unreasonable to commence pilot studies using autologous stem cells, considering animal work had shown that autologous grafts were as successful as allogeneic in suppressing adjuvant arthritis¹⁰.

Joske, *et al*¹¹ reported the first autologous PBSCT performed solely for RA in Australia. A wheelchair bound patient who had received all available therapies for RA underwent stem cell collection with cyclophosphamide 4 g/m² and granulocyte colony stimulating factor (G-CSF). He then received 200 mg/kg cyclophosphamide as conditioning, followed by infusion of an unmanipulated PBSC graft. Transplant related morbidity was minimal and he attained an ACR70 (American College of Rheumatology) remission for 25 months. Reintroduction of only 10 mg of methotrexate (previously unsuccessful) has maintained his disease under substantial control for a further 12 months at last followup (personal communication, D. Joske). This important case illustrates that, although autologous PBSCT was not curative, it has resulted in a form of immunomodulation of the disease process that is long lasting in a previously resistant patient. The mechanism of this immunomodulation is unclear, but at 3 years' followup it presumably represents some form of "lymphohemopoietic re-setting" rather than simple immunosuppression due to the cyclophosphamide itself.

DOSE ESCALATION STUDIES

Following the publication of individual case reports, it was important to formalize the assessment of PBSCT in RA. Snowden, *et al*¹² performed a dose escalation study in 2 cohorts of 4 RA patients each — one receiving 100 mg/kg and the other 200 mg/kg cyclophosphamide. Analysis of the safety of the procedure was the primary objective of this study, but by comparing 2 dose schedules of cyclophosphamide it was possible to gain preliminary data upon which to base future randomized trials. All patients had stem cells collected with G-CSF alone and received unmanipulated PBSC. Cohort 1 (100 mg/kg cyclophosphamide) attained transient moderate responses — 20–50% reduction in joint counts for 2 to 3 months. Of interest, 3 of the 4 patients in cohort 1 have expressed an interest in having the procedure performed again, with one patient proceeding to a second autograft — an

indicator of the willingness of RA patients to take risks in their quest for a pain-free existence. That these patients had only transient responses suggests that pulse cyclophosphamide therapy (up to a total of 4–5 g/m² in this cohort) as used in systemic lupus erythematosus (SLE) may not be efficacious in patients with RA.

The second cohort has now been followed extensively for over 2 years. The degree of response in this group of patients has been longer and more substantial. Three of the 4 patients attained ACR70 status and one attained ACR50. However, there has not been complete eradication of disease, with all patients now having returned to some form of DMARD (Figure 1), albeit with an overall reduced dose of prednisone in all patients compared to pre-PBSCT. Two patients (2.2 and 2.3) had successful reintroduction of DMARD at 4 and 6 months, respectively — both patients had been unresponsive to these agents. The remaining 2 patients (2.1 and 2.4) had disease recurrence just prior to 2 years — one achieved rapid disease control within one month using leflunomide. The second patient has recommenced prednisone (at a smaller dose than pre-PBSCT); some joint discomfort persists despite DMARD. Overall, 3 of the 4 patients have expressed willingness to undergo the procedure again if required. There have been no longterm sequelae from the stem cell rescue. As a disease modulator and steroid sparing procedure one could argue it has been a success for these patients with severe resistant RA.

The second cohort has shown that (1) the procedure is relatively safe with no longterm sequelae at the 2 year time point; (2) the procedure is unlikely to be curative but can provide significant remissions for up to 2 years; and (3) autologous PBSCT appears to provide a form of immunomodulation to RA allowing disease to be held under control in previously resistant cases.

RATIONALE FOR A RANDOMIZED TRIAL

To improve the duration and degree of response in these patients a randomized trial was required to address questions based on our knowledge at the time. As shown in Figure 2, issues to be resolved included: (1) patient selection, (2) the PBSC product, (3) the conditioning regimen, and (4) the possibility of post-PBSCT maintenance.

Patient selection. Patients chosen for most PBSCT trials have had severe and resistant disease, which is not unreasonable given the experimental nature of the procedure. We chose to include patients who had failed at least 2 DMARD, one of which had to be methotrexate, the gold standard of therapy for RA. The recent advent of new biological agents including tumor necrosis factor- α (TNF- α) inhibitors¹³ may in the future necessitate either direct comparison with PBSCT or failure of response as an inclusion criterion in new PBSCT trials. It is of interest that anti-TNF- α therapy in conjunction with methotrexate resulted in 39% of patients achieving ACR50¹³ — leaving a significant number of patients eligible for therapies such as PBSCT, if failed anti-TNF- α therapy was

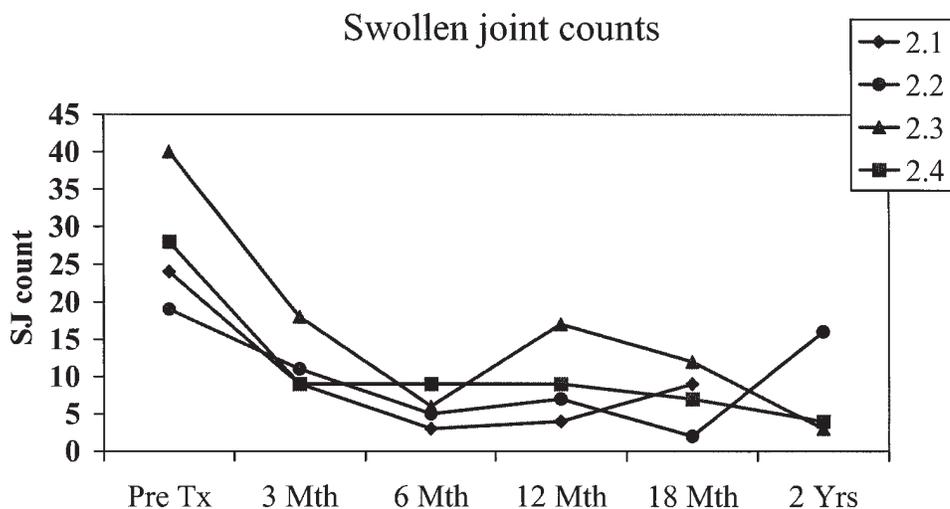


Figure 1. Swollen joint counts of 4 patients receiving 200 mg/kg cyclophosphamide and unmanipulated PBSC.

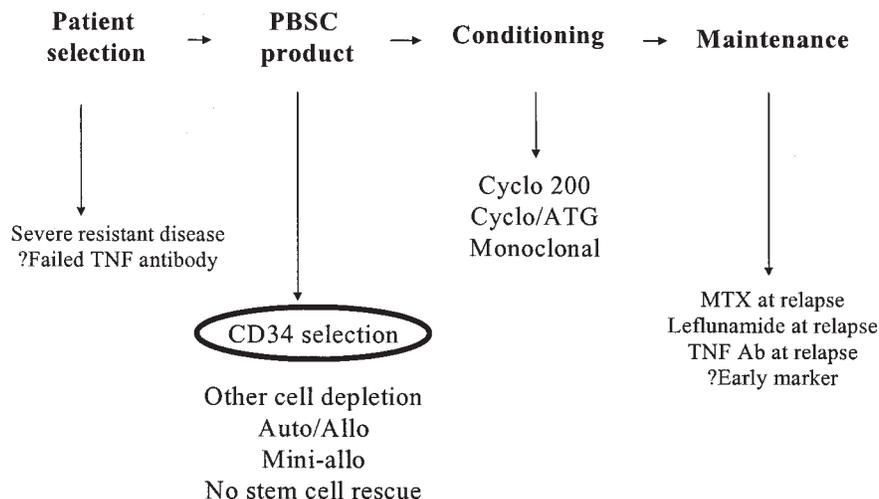


Figure 2. Future issues when considering PBSC in patients with severe RA.

made an inclusion criterion. Other considerations for patient selection would include adequate end organ function — fortunately this issue has less bearing in RA compared to other autoimmune diseases such as systemic sclerosis.

PBSC product. A contentious issue in PBSC for autoimmune diseases is the choice and/or manipulation of the PBSC graft. The increased morbidity and mortality of allografting and the subsequent consensus guidelines of investigators¹⁰ ensured that at present the use of autologous grafts is the preferred option. The EBMT/EULAR database established in Basel now has over 300 cases registered, of which the vast majority are CD34 selected (A. Tyndall, personal communication), indicative of the widely held belief that graft manipulation is

imperative to remove autoreactive cells (presumed to be T lymphocytes)¹⁴. CD34 selection entails exposing a PBSC product to an immunomagnetic column conjugated to the CD34 antigen¹⁵ — the marker believed to represent the pure stem cell capable of both self-renewal and differentiation. A secondary result of this selection process is T cell depletion of PBSC. In general, commercially available cell selection devices will deplete a PBSC product by 2.5–4 log. This still leaves around 10⁵⁻⁶ lymphocytes in the product; this level of T cell depletion is generally considered enough to prevent graft versus host disease in the allogeneic setting. However, the relevance of this degree of T cell depletion in the autologous setting remains unclear.

The depletion of T cells by CD34 selection in auto-PBSCT for autoimmune disease makes a number of assumptions. The first is that the autoreactive cell to be removed is known and that a 3 log depletion is enough to eradicate it. In RA, the pathogenic cell is difficult to define. The T cell is often considered the most likely culprit because of the association of RA with certain HLA-DR subtypes and because of the abundance of T cells in the synovium of RA sufferers. However, these T cells secrete very few proinflammatory cytokines and are relatively inactive, with little evidence of oligoclonality¹⁶. The number of T cells in the joint has little correlation with subsequent joint damage¹⁷, and anti-T cell therapies such as anti-CD4 monoclonal antibodies have been relatively ineffective¹⁸.

Another assumption associated with CD34 selection is that it is relatively safe. This is generally true — engraftment is usually similar to unmanipulated PBSCT; however, immune recovery of CD4 and in particular CD4+, 45RA+ cells is delayed in recipients of these grafts¹⁹. This has led to concern about infection with opportunistic organisms. Recent reports of a death in a CD34 selected patient from Epstein-Barr virus associated lymphoma²⁰ and an increased incidence of CMV viremia²¹ in recipients of autologous CD34 selected grafts add to this concern. Given the conflicting data outlined above it was decided at our institution that a randomized trial of CD34 selected versus unmanipulated autologous PBSCT would be an important step in trying to evaluate the role of PBSCT in RA (Figure 3).

Given the initial successes with allografting⁵ and the likelihood of relapse in the autologous setting, sustained remissions may be achieved with allografts using the “mini-allografting”²² technique with fludarabine based regimens. Previously it was thought that the use of high dose chemotherapy in the form of conditioning was needed to ablate residual disease in the recipient — this is now considered less imperative as it has become clear that the allogeneic donor immune system can eradicate disease by the “graft versus tumor effect.” In mini-allografting the conditioning is kept to a minimum but is highly immunosuppressive to allow adequate

engraftment of donor hematopoietic tissue — a similar rationale to transplantation in aplastic anemia. The main advantage of this radical new way of performing HSCT is that patients engraft rapidly, with less toxicity because of the reduced conditioning. Graft versus host disease, however, still remains a major problem. Thus mini-allografting has many potential advantages for autoimmune disease, and it may not be unreasonable to commence small pilot studies using this technique in severely affected patients.

Conditioning. From the dose escalation study it was clear that 200 mg/kg cyclophosphamide was safe, with acceptable toxicity levels. An argument could be made for modifying this regimen to cyclophosphamide 200 mg/kg with ATG 90 mg/kg, which has been successful in allografting for aplastic anemia²³, the prototype autoimmune disease in hematology. In the allogeneic setting ATG is used to overcome immunosuppression in the host, thus preventing rejection of the donor marrow; this is not an issue in the autologous setting. However, if immunosuppression of the disease process is the main objective of PBSCT, this agent may contribute significantly. If used, it would result in significant immunosuppression of the patient but will have little effect on the possibility of reinfusing autoreactive cells (presumably lymphocytes) in the PBSC product. In addition the use of ATG would probably increase the incidence of opportunistic infections — an unwanted complication in patients who are already taking immunosuppressive medications.

One group has reported high dose therapy without PBSC rescue²⁴ using 200 mg/kg cyclophosphamide — this form of therapy allows immunosuppression without the reinfusion of autoreactive cells. Although this report shows encouraging initial results, findings would need to be assessed in a randomized trial with PBSC rescue using the same conditioning. Hematological recovery would be longer in patients without PBSC rescue, which raises the possibility of increased morbidity or even death from neutropenic sepsis and bleeding.

Maintenance. The case reports and our second cohort of patients in the dose escalation study have shown that commencement of DMARD post-PBSCT may be an important

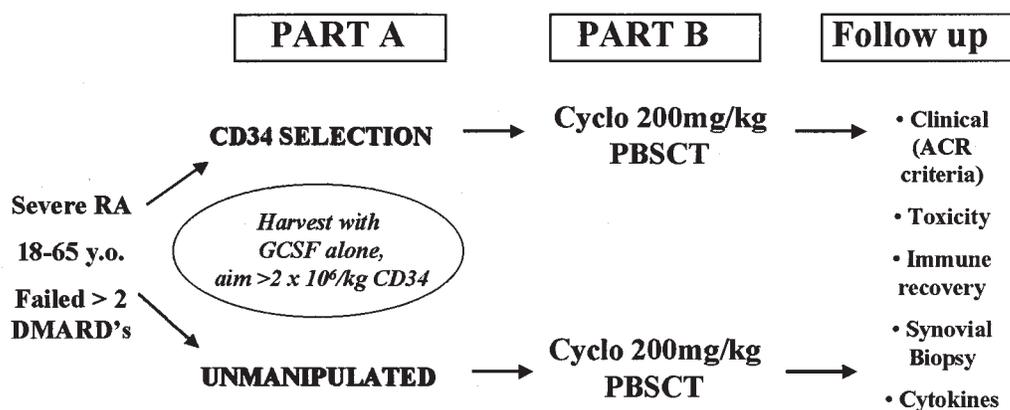


Figure 3. Design of the current randomized trial in Australia to investigate T cell depletion of the PBSC graft.

option to prolong remissions in these patients. The exact timing of recommencement of therapy and the agents to be used should be the subject of future studies. A prospective trial may allow the recognition of potential markers of disease recurrence before clinically evident, thus allowing the design of future protocols. In the current Australian randomized trial we see patients every month for a year and then 3 monthly to assess a number of disease variables, including inflammatory markers, cytokine analysis, immune reconstitution, and in some patients synovial biopsies.

CONCLUSION

The design of the current randomized trial of CD34 selection versus unmanipulated PBSCT in RA is illustrated in Figure 3. Using a conditioning regimen we know is effective, we have now transplanted 31 patients in this trial; this unique opportunity to assess the role of CD34 selection in this procedure will help us to gain a valuable insight into the pathophysiology of the disease. There have now been 41 PBSCT performed for severe RA in Australia and the mortality remains zero. Information from this trial may help optimize the response of RA to PBSCT, thus enabling it to be compared to more conventional therapies in the future.

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