

The Use of High Dose Immunoablative Therapy with Hematopoietic Stem Cell Support Therapy in the Treatment of Severe Autoimmune Diseases

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1. Introduction

The dose of an immunosuppressive agent in the treatment of an autoimmune disease (AD) has in the past been limited by its organ toxicity, mostly the bone marrow, even though some patients do not respond to such tolerated maximal doses. The use of autologous haematopoietic stem cells (HSCs) as support following myeloablative chemotherapy has allowed this threshold to be surpassed in the treatment of AD and is subject of an international project. Not all such high dose immunosuppression is myeloablative eg cyclophosphamide (Cy), which cannot kill HSCs, but the prolonged period of hematopoietic reconstitution may be shortened and therefore be less risky with a hematopoietic stem cell transplant (HSCT).

An international consensus statement on the issue was published in April 1995, before the first patient was treated [1], and the first publication of an AD patient receiving an HSCT as primary treatment of the AD was Oct 1996 [2]. Since then, over 500 patients have been so treated, most within the context of phase I and II studies following published guidelines [3]. The concept evolved from a combination of animal model support for the concept, coincidental malignancy and AD cases undergoing HSCT and improving safety and technological aspects of transplantation.

1.1. Autoimmune Disease

The mechanism of human AD is partly genetic, partly environmental and only partly understood. Several current reviews [4,5] have pointed out the over simplistic view of a clonal expansion of autoaggressive lymphocytes, and stress the complex matrix of physiological

events (continuous self antigen presentation is needed for maintenance of peripheral tolerance) which may deviate to an autoimmune reaction under certain conditions eg intercurrent infection, inflammation or overwhelmed apoptosis all leading to activation of nave autoreactive T cells, migration and finally location and destruction of self structures.

In addition, the possibly pivotal role of the innate immune system in initiating autoimmunity has been recently revalued. Whatever the final mechanism will prove to be, it will almost certainly be a dysregulation of multicellular/cytokine microenvironments, rather than a single clonal disease. In fact all attempts to "cure" AD with specific monoclonal antibodies or agents aimed at single cell types have failed. In addition, some broader based strategies, such as CAMPATH 1 treatment of MS have shown an emergence of a second AD, ie thyroid in 1/3 of the treated patients. More recently the concept of regulatory T cells (CD4+, CD25+ secreting IL10 and TGF β) has re-emerged as probable key players in this equilibrium between tolerance and autoimmunity [6]. These were previously called suppressor cells.

This dysregulation concept has been observed during the phase I and II AD HSCT trials eg following HSCT for RA, there was no apparent advantage or even a disadvantage of T cell purging of the graft product, perhaps suggesting the benefit in some cases of not removing regulator cells. In addition, some relapses were easily controlled with agents which had failed pre-HSCT eg methotrexate in SSc or RA.

In any case the oncological concept of removing all malignant cells for a cure is not applicable to AD, but rather a de-bulking followed by resetting of the normal physiological controls. It has been observed that many AD patients are clinically well 5 years post autologous

HSCT despite clear evidence ongoing autoantibody production such as antinuclear antibodies. In addition, some AD patients have relapsed following allogeneic HSCT with full chimerism and donor type lymphocytes found in the lesions.

For these reasons it is most likely that agents such as Cy which affect many cell lineages are still the mainstay of treating severe AD such as renal SLE, rather than more focussed agents such as mycophenolate mofetil or cyclosporin, so useful in solid organ transplantation. An additional factor is also the chronification and recruitment which has occurred in AD before the first symptom, as opposed to the exact date of onset of the immune disease in solid organ transplantation.

1.2. Coincidental Autoimmune Disease and HSCT

Since the onset of allogeneic BMT and HSCT, and more recently autologous HSCT it has been observed that some patients receiving an HSCT for a malignancy also showed improvement or long term remission of a coincidental AD. This has been extensively reviewed [7,8], but there are some important observations to be made.

1. The reporting has not been systematic and was mostly retrospective.
2. The conditioning has been mostly rather aggressive, given the primary indications.
3. The details concerning the AD severity and activity are mostly incomplete.

In addition transfer of AD has occurred with allo HSCT, as has emergence of a second AD following auto HSCT. However, these reports in addition to the animal model data gave sufficient grounds to move into phase I and II pilot studies in humans with severe AD.

2. Phase I and II Studies

There are currently 458 reports from 113 transplant centres in 21 countries of HSCT given for a severe AD in the EBMT/EULAR data base, 437 being autologous and 21 allogeneic. Around another 100 cases in the

IBMTR data base and reported in the literature.

The diseases transplanted are seen in Table 1. The preponderance of secondary progressive MS and SSc reflects the lack of other effective options in these disorders. Protocols were mostly in accordance with published consensus guidelines [3], and the overall actuarially adjusted transplant related mortality (TRM) was 7 (4-10)%. A striking difference between diseases was seen, with a 12.5% TRM in SSc and only one death in 63 RA patients, none in the Cy only conditioned patients. This probably reflects a less aggressive and a general better condition of the RA patients at the time of transplant.

2.1. Multiple Sclerosis

An analysis of the first 85 MS patients showed a 3 year progression free survival of 78% of the secondary progressive cases, with a risk of death from any cause at three years being 10 (± 7) months, 95% confidence interval [9]. The median follow-up was 16 (3-59) months. Fewer positive responses were seen in primary progressive MS (66% \pm 23%)

A more impressive effect was seen with the MRI changes, considered a surrogate marker of MS. In a ten patient subgroup analysis from one center, the gadolinium enhancing MRI lesions reduced to zero after a median 15 (4-30) month follow-up [10]. However more long term follow up is required in this chronic disease, including a randomised controlled trial (see ASTIMS Trial below).

The majority of MS cases received a peripheral stem cell transplant, being mobilised with Cy (mostly 4 g/m²) and G-CSF. There was a suggestion that G-CSF alone could induce a flare of the MS, being less when combined with Cy. Conditioning was mostly with BEAM (carmustine, etoposide, cytosine arabinoside and melphalan) alone (16%) or BEAM plus ATG (47%). Other regimens included Cy plus TBI and busulphan/Cy. No clear advantage of T cell purging was observed.

2.2. Systemic Sclerosis (SSc)

In the first 45 patients, an improvement of 25% or more was seen in 70% of the patients, with a TRM of 17% [11]. Several protocols were used, mostly either Cy based (4 g/m² Cy mobilisation and Cy 200 mg/kg body weight conditioning or radiation 8 Gy/Cy 120 mg/kg body weight. With further patient recruitment, the TRM fell to 12.5%, considered to be related to more careful patient selection. Lung function tended to stabilise and some factors were identified as potentially hazardous for HSCT eg pulmonary hypertension >50 mmHg mean pulmonary arterial pressure, severe cardiac involvement, severe pulmonary fibrosis and uncontrolled systemic hypertension. When such patients were excluded from the analysis, the TRM was 7%, suggesting that the proposed randomised controlled trial (see ASTIS Trial below) would be ethical, given a near 50% 5 year mortality of this subgroup of patients.

Table 1.

Disease.

Multiple sclerosis	131	MCTD	4
Myasthenia	1	Cryoglobulinemia	3
Neuropathy	1	Behcet	3
ALS	2	Wegener's	3
SSc	69	Polychondritis	1
SLE	53	ITP	12
Rheumatoid arthritis	72	AIHA	8
Psoriatic arthritis	2	PRCA	7
Juvenile chron. arthritis	47	Evans	1
Ankylosing spondylitis	2	TTP	2
Sjogren	1	Bowel disease	2
Vasculitis	1	Other	3
Dermatomyositis	6		

2.3. Rheumatoid Arthritis

Of the 72 patients transplanted, an analysis of the first 51 showed significant improvement, with 78% achieving an ACR 50 response. This is a composite score clinical and laboratory parameters which should improve by at least 50%. Most of the patients had failed at least 3 conventional disease modifying antirheumatic drugs (DMARDs) such as methotrexate or sulphasalazine before the transplant. Some degree of relapse was seen in 73% of patients post transplant, but was relatively easy to control with drugs which had proven ineffective pretransplant. The median follow-up was 18 (6-40) months, and the majority of patients received a conditioning regimen of Cy 200 mg/m² alone and received peripherally harvested stem cells after either G-CSF or Cy/G-CSF (equal numbers) mobilisation.

These phase I and II experiences have been integrated into a phase III randomised study (ASTIRA Trial, see below.)

2.4. Juvenile Idiopathic Arthritis

A total of 43 children with idiopathic juvenile arthritis, mostly the systemic form called Stills disease, have been registered. Most of these cases were treated in two Dutch centers using a bone marrow obtained stem cell source and a conditioning protocol of Cy 200 mg/kg body weight, TBI 4Gy and ATG (N Wulfraat, personal communication).

In the whole group there were 15 complete remissions and 3 partial remissions reported. In those attaining remission, the corticosteroid dose could be reduced and some patients experienced puberty and catch up growth. Three patients died from macrophage activation syndrome, thought to be related to intercurrent infection or uncontrolled systemic activity of the disease at the time of transplantation. Protocols were modified accordingly such that systemic activity is controlled before the transplant with methyl prednisolone intravenously. Since this modification, no further such deaths have occurred.

Further phase I and II pilot studies using a cyclophosphamide, non radiation based protocol showed similar results, and therefore a prospective randomised (ASTIJA) trial using a non radiation based regimen is being planned (Wulfraat, EBMT Meeting, Montreux 2002).

2.5. Systemic Lupus Erythematosus

Of the 53 registrations, most had either renal and/or CNS involvement, and 37 had failed conventional Cy treatment. A peripheral stem cell source after mobilisation with Cy and G-CSF was used in the majority. Fifteen patients received a conditioning with Cy and ATG, and five other regimens were employed. There were 4 deaths due to treatment and one from progressive disease, resulting in an actuarially adjusted TRM of 11 (2-20)%.

In those patients with sufficient data for analysis, 30 achieved a "remission", defined as a SLEDAI (Systemic Lupus Erythematosus Disease Activity Index) of ≤ 3 and steroid reduction to < 10 mg/day. Twelve others did not reach this end point. Half of the remissions relapsed to some degree, and were mostly easily controlled on standard agents which had previously been ineffective.

A more complete analysis of the data is underway with a view to proposing the most appropriate phase III protocol. This trial (ASTIL) will most likely include a post transplant maintenance agent, either azathioprine or mycophenolate mofetil

2.6. Refractory Autoimmune Cytopenias

A preliminary review of 16 cases has been reported [12] consisting of the following diseases: idiopathic thrombocytopenic purpura (n=9), pure red cell aplasia (n=4), autoimmune haemolytic anaemia (n=2) and Evans Syndrome (n=1). Median age was 31 (4-45) years and median time to transplantation was 93 months (12-236). Most received stem cells from blood mobilised with either growth factors (n=7) or Cy plus growth factors (n=6). There were two bone marrow harvests. Conditioning regimens included Cy alone (n=3), Cy with other drugs or ATG (n=9), melphalan (n=2) or were fludarabine based (n=2). Most (n=12) had purging of immune cells from the graft product.

Median follow up of surviving patients is 30 months (5-53). Three patients died within 100 days post transplantation, two of hemorrhage and one from progressive haemolysis. Eight patients showed a response to treatment, 4 complete remissions (2 ITP, 1 Evans and 1 PRCA), sustained in three patients.

Randomised trials are under discussion for ITP.

The numbers of cases with vasculitis, inflammatory bowel disease, Behets disease, relapsing polychondritis and other ADs are too small to draw meaningful conclusions, with further phase I and II standardised protocol pilot studies proceeding.

3. Randomised, Prospective Controlled Trials

3.1. The ASTIMS (Autologous Stemcell Transplantation International Multiple Sclerosis) Trial

Secondary progressive MS patients with an EDSS score between 3.5 and 6.0 will be randomised to either HSCT (BEAM and ATG) followed by an unmanipulated graft or mitoxantrone. The primary end point is progression free survival at 3 years, and each arm will recruit 80 patients. Further details are available on the EBMT website www.EBMT.org or E-mail: neurolab@cisi.unige.it

3.2. The ASTIS (Autologous Stemcell Transplantation International Scleroderma) Trial

Diffuse skin SSc (scleroderma) patients are selected who have less than 4 years of diffuse skin involvement and evidence of progressive and organ or life threa-

tening disease. The primary end point on which the trial is powered is event free survival at 2 years, events being arbitrarily but precisely defined to capture irreversible and severe end organ failure or death.

Exclusion criteria are based on the phase I and II data to avoid an unacceptably high TRM risk together with a minimal chance of clinically significant improvement.

The treatment arm is mobilisation with Cy 4 g/m² and G-CSF, followed by CY 200 mg/kg body weight conditioning plus ATG and a CD34 selected graft. The control arm is monthly IVI pulse CY 750 mg/m² for 12 months. Each arm will have 100 patients.

ASTIS is running, and further details are available on the website: www.astistrial.com, or E-mail astistrial-fps@unibas.ch. Currently (July 2002) there has been no transplant related mortality

3.3. The ASTIRA (Autologous Stemcell Transplantation International Rheumatoid Arthritis) Trial

Active RA patients who have failed at least 4 DMARDS including methotrexate and anti TNF alpha programs with a disease duration between 2-15 years will all receive stem cell mobilisation with Cy 4 g/m² and G-CSF. Randomisation then occurs to either continued conventional therapy with either methotrexate or leflunamide or conditioning with Cy 200 mg/m² and ATG. The graft is unmanipulated, and maintenance with methotrexate or leflunamide is given. The primary end point is the number of patients reaching a good or moderate EULAR response or and ACR 20 at six months. 16 patients in each arm are required, calculated on a >50% difference in the two groups. The trial is running.

Further details from www.EBMT.org or E-mail: 114077.1034@compuserve.com

4. Open Issues

4.1. Immune Reconstitution

A centralised international initiative has begun to gather and share the data on immune reconstitution in order to fine tune later protocols and/or better understand the immunopathology of the ADs being transplanted. So far, anecdotal data has not produced an immune cell phenotypic pattern which predicts either remission or relapse. As already known, the CD8 RO pos "memory cell" compartment expands post transplant, with later appearance of CD8 and CD4 RA pos "naive" T cells. CD19 and 20 B cells and NK cells reconstitute within weeks to months, but CD4 cells may take months to years, depending on the severity of the conditioning and T cell depletion.

4.2. Allogeneic HSCT Including Mini-transplantation

It has often been stated that to cure an AD one must

replace the "sick", autoaggressive immune system with a genetically "healthy" one through allotransplantation. This concept, which is probably true for malignant clonal disease in which only a graft versus malignancy effect can eradicate all cells, is over simplistic for AD. Apart from the theoretical issues of autoimmune reactions being networks of autoreactivity (cells, cytokines, growth factors), there are concrete clinical examples suggesting that the issue is far from clear.

There are well documented cases of autotransplanted patients who have attained a long (<5 years) clinical remission, despite evidence of ongoing autoantibody production. On the other hand, one well documented case of RA receiving an allograft from her brother to treat gold induced aplasia experienced a 4 year remission followed by clinical and serological relapse [13]. All examined immune competent cells were of donor origin, suggesting either in the new environment, the "healthy" immune system was deviated to autoimmunity, or more likely that siblings carry autoimmune genes waiting to be expressed. Other equally plausible explanations exist including the well known fact that the vast majority of immune effector cells are recruited into an immune reaction, and are not the antigen specific initiators. In other words, a tiny number of residual host autoreactive memory cells could re-initiate the process leading eventually to AD clinical expression.

The concept of "resetting" the immune system rather than ablating has grown in the last years. What is not in doubt is the increased risk of allo versus auto transplantation, and therefore the strategy of "safety first" with the auto transplant program which has already shown a higher than anticipated TRM.

4.3. Gene Marking

In allografting it is relatively easy to identify donor and recipient cells, but with auto HSCT gene marking would be needed to identify those cells eg lymphocytes which originate from the graft product and those which arise from cells remaining in the patient after conditioning.

The problem of gene marking is well known-if a gene product expressed on the surface of the cell is used, then it may change the function of the cell, and in any case a significant loss of gene expression and indeed persistence may and does occur during multiple cell divisions.

If one adds to this the phenomenon of massive non specific recruitment of immunocompetent cells during an immune reaction, the interpretation of finding gene marked cells in a lesion would be very difficult.

5. Conclusion

The phase I and II data suggest that for some highly selected AD patients HSCT could offer another treatment option with an ethically acceptable benefit/risk ratio. The most important next step is to show through prospective, randomised, multicentre trials that HSCT

with all its associated risks imparts an advantage concerning quality of life to such patients.

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