

REVIEW

Randomized Controlled Trials of Autologous Hematopoietic Stem Cell Transplantation for Autoimmune Diseases

The Evolution From Myeloablative to Lymphoablative Transplant Regimens

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Introduction

Hematologists/oncologists initiated the use of autologous hematopoietic stem cell transplantation

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(HSCT) as a treatment for severe autoimmune diseases beginning in the early 1990s. According to multicenter reports, this therapy has been plagued by a high rate of transplant-related mortality. However, transplant-related mortality from different individual centers varies widely. We will review and discuss how the differences in philosophy, rationale, and design of these protocols affect treatment-related mortality, and we will compare how these differences have influenced ongoing or soon-to-be-opened randomized controlled trials of autologous HSCT for patients with autoimmune diseases. In particular, we will emphasize the distinction between myeloablative and lymphoablative trials.

Autologous HSCT is a 3-step procedure involving collection of HSCs, treating the patient with a conditioning regimen to eliminate self-reactive lymphocytes within the body, and, finally, reinfusion of the previously frozen autologous HSCs (Figure 1). A patient's HSCs are collected by leukapheresis from the peripheral blood and may or may not undergo further purification or enrichment in the laboratory. The conditioning regimen used to eliminate self-reactive lymphocytes within

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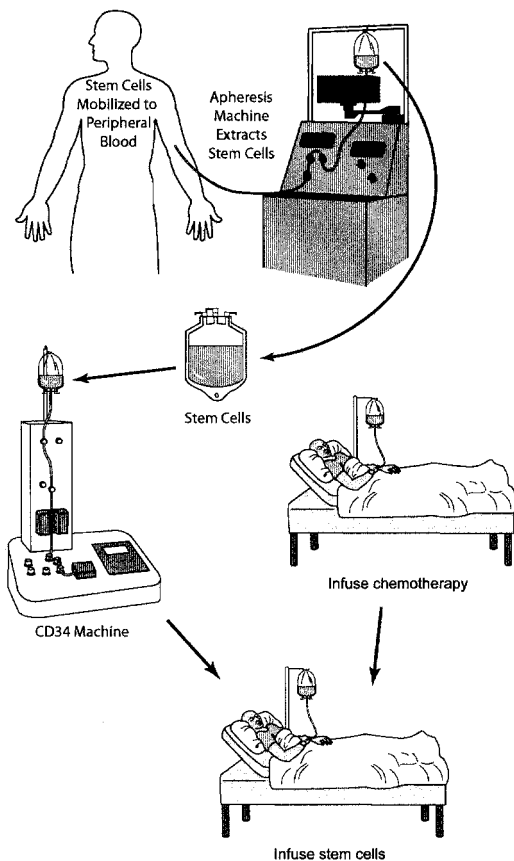


Figure 1. Three-step procedure of autologous hematopoietic stem cell (HSC) transplantation for autoimmune diseases. First, HSCs are harvested from the blood with or without further purification in the laboratory. Next, the patient undergoes chemotherapy to eliminate the immune system. Finally, autologous HSCs are infused to facilitate immune reconstitution.

the patient has been designed (depending on the investigator) either to specifically target lymphocytes (lymphoablative regimen) or to destroy the entire hematopoietic bone marrow compartment (myeloablative regimen). Nevertheless, the goal of autologous HSCT for autoimmune diseases is to generate new self-tolerant lymphocytes after elimination of self or autoreactive lymphocytes (i.e., lymphoablation), rather than to ablate and reconstitute the entire hematopoietic compartment (myeloablation).

Mortality following autologous HSCT

In 1999 and again in 2005, the European Bone Marrow Transplant/European League Against Rheumatism (EBMT/EULAR) registry reported on mortality

following autologous HSCT for autoimmune disorders in multiple centers (1,2). In 1999, when 74 patients had been treated, overall mortality was 12.2% (9 patients) and treatment-related mortality was 9.5% (7 patients) (1). In a followup EBMT/EULAR registry report in 2005 on 473 patients, overall mortality was 11% (53 patients) and mean treatment-related mortality was 7% (95% confidence interval 4–10%) (2). Infection was the most common cause of transplant-related mortality, but other causes included hemorrhage, cardiac failure, pneumonitis, and veno-occlusive disease of the liver. This rate of mortality is well above the <2% (3) or <3% to 5% (4) initially used to justify the risk of autologous HSCT for autoimmune diseases.

Autoimmune diseases are a highly heterogeneous group of disorders with variable organ system involvement, different prognoses, diverse etiologies and pathologies, involvement of different age groups, and variable prior immunomodulating and immunosuppressive treatment intensity. For these reasons, comparison of treatment-related mortality across diseases and between different centers using different transplant regimens has limitations. In contrast to the composite survival data quoted above, single-center treatment-related mortality may vary considerably. For example, Northwestern University reported a single-center treatment-related mortality rate of 0% and an overall mortality rate of 3.8% in its first 79 patients (5).

There are numerous possible explanations for apparent differences in treatment-related mortality, including 1) factors related to the organization and delivery of health care, a phenomenon known as a “center effect”; 2) biologic paradigms such as patient selection, the type of autoimmune disease, and disease severity; and/or 3) the type of treatment procedure. Examples of variables involved in a center effect include patient care team experience derived from procedure volume, treatment using only institutional review board–approved protocols, data safety monitoring board oversight, and extent of dedicated attending physician versus house staff management of patient care. Although a center effect influence on mortality has been reported for HSCT in patients with hematologic malignancies (6,7), it is currently not possible to attribute differences in treatment-related mortality to the local practice of health care delivery for autoimmune diseases, since there are no reports evaluating a center effect on HSCT mortality in autoimmune disorders. In terms of a biologic paradigm, the different types of diseases and selection criteria for transplant were developed jointly through international conferences and meetings, and

disease-specific selection criteria were shared both across the Atlantic and within the US. Therefore, without further information, the type of disease, patient selection bias, or lesser disease severity cannot currently explain single-center variability in treatment-related mortality.

For the rest of this discussion, we will focus on the role of the treatment procedure itself in causing treatment-related mortality. For an autologous HSCT procedure, the toxicity and efficacy arise from the treatment regimen (also known as the conditioning regimen). The autologous stem cells are used to achieve expedited recovery of hematopoiesis. In general, there are 2 different treatment approaches (myeloablative and nonmyeloablative) being utilized as conditioning regimens for patients with autoimmune disorders. Following a myeloablative treatment regimen, HSCs must be infused to reconstitute hematopoiesis in order to prevent irreversible and lethal pancytopenia. Following a nonmyeloablative conditioning regimen, HSCs are not necessary for hematopoietic recovery but are given as a safety measure to shorten the duration of posttreatment neutropenia by ~4–5 days. Since the term “not myeloablative” (i.e., nonmyeloablative) conveys what the treatment is not capable of doing, we will, in general, use the term lymphoablative instead of nonmyeloablative to convey the function for which a nonmyeloablative conditioning regimen is designed. The rationale behind myeloablative or lymphoablative regimens and their proposed use in randomized trials hinges on divergent but virtually uncontested and unproven scientific hypotheses.

Mobilization of HSCs

The most common method of collecting HSCs (HSCs are also immune stem cells) is by mobilization from the bone marrow into the peripheral blood (Figure 1). Since negligible numbers of HSCs, if any, are detectable in the peripheral blood during steady state, either a hematopoietic growth factor, such as granulocyte colony-stimulating factor (G-CSF), or cyclophosphamide along with G-CSF, is necessary to mobilize HSCs from the bone marrow into the peripheral blood for collection by leukapheresis (Figure 1). G-CSF is given daily by subcutaneous injection, and HSCs are collected by leukapheresis on the fourth day of G-CSF injection. The disadvantage of mobilization with G-CSF alone is, depending on the autoimmune disease, the potential for disease flare (8). Cyclophosphamide mobilization is performed by a 1–2-hour intravenous infusion

of cyclophosphamide (2.0–4.0 gm/m²) and, beginning 3–5 days later, addition of daily subcutaneous G-CSF. Mobilization with combined cyclophosphamide and G-CSF is associated with 1 day of neutropenia (usually 8–9 days after cyclophosphamide). HSCs are collected by leukapheresis upon white blood cell rebound, usually 10 days after cyclophosphamide administration. Advantages of cyclophosphamide/G-CSF mobilization are higher stem cell yields, an in vivo purge effect by selectively killing lymphocytes in the cell cycle, and a cyclophosphamide-mediated disease-ameliorating effect.

Philosophy behind transplant conditioning regimen: myeloablative versus lymphoablative

Myeloablative regimen. In a malignancy, such as leukemia or lymphoma, primitive hematopoietic progenitor cells usually harbor the mutated oncogene(s). Logically, the conditioning regimens were designed to eliminate these malignant HSCs. Cancer relapse generally occurs from residual malignant cells within the person's body that survived the conditioning regimen, rather than from tumor cells contaminating the reinfused autograft (9). Since malignant cells that survive the conditioning regimen often are the source of relapse, any surviving malignant cell is inherently “against us” and must be destroyed. Because malignancies do not spontaneously remit, plateau, or “burn out” and often markedly shorten life expectancy, aggressive intervention, sometimes with lethal consequences, is justifiable. This has resulted in the concept of maximizing treatment to eliminate minimal residual disease (10). As a result, autologous HSCT regimens for malignancies are designed for myeloablative intent. These regimens deliver radiation and/or chemotherapy beyond the ability of hematopoiesis to spontaneously recover and close to the limit of what other organs (kidneys, liver, and lungs) with normal function can tolerate. There is no such entity as nonmyeloablative autologous HSCT for patients with cancer. Following myeloablative HSCT for cancer, lymphoablation with increased risk of opportunistic infections would be an unwanted side effect.

Since HSCT for autoimmune disorders was initially proposed by medical oncologists, myeloablative regimens with which oncologists had experience in treating cancer were often chosen as transplant conditioning regimens for autoimmune diseases. In addition, anecdotal observations of autoimmune diseases remitting after myeloablative therapy for cancer supported such an initial approach. Following myeloablation, HSCs

must be reinfused because the bone marrow's ability to safely recover has been exceeded. Since myeloablative regimens are also lymphoablative, a myeloablative regimen will be extremely immunosuppressive. Regeneration of the immune system after severe lymphopenia would theoretically result in an "immune reset" without subsequent autopathology (11).

Lymphoablative regimen. In performing autologous HSCT as therapy for an autoimmune disease, it is assumed that while the patient may have a genetic predisposition, the disease is induced by environmental stimuli (12,13) and is not exclusively a genetically pre-ordained stem cell defect. Thus, the goal of the conditioning regimen is lymphoablation and, in contrast to treatment for malignancies, myeloablation would be the unwanted side effect. Indeed, it was first demonstrated by Slavin that lymphoablative conditioning with cytotoxic agents but without autologous stem cell transplantation resulted in complete restoration of self-tolerance and elimination of advanced autoimmunity in a patient with mixed cryoglobulinemia who has remained disease free for more than 25 years (14). This suggests that lymphoablative rather than myeloablative conditioning will be sufficient and possibly safer for eliminating autoimmunity and reinducing self-tolerance via new T lymphocyte development in the thymus.

Lymphoablative doses of cyclophosphamide (200 mg/kg) without stem cell support have been used to treat autoimmune diseases (15). Potential advantages in favor of lymphoablative HSCT, compared with dose-intense cyclophosphamide without stem cells, are as follows: 1) the ability to safely deliver a higher cyclophosphamide dose (~250 mg/kg), since cyclophosphamide is used both for mobilization of stem cells and for lymphoablation; 2) the ability to add other immunosuppressive agents such as antithymocyte globulin (ATG) to the regimen to further increase immunosuppression; and 3) the ability to decrease the duration of post-conditioning regimen neutropenia, the hospitalization interval, and the risk of infection by infusion of stem cells previously primed with G-CSF.

Myeloablative versus lymphoablative agents. Some agents used traditionally at maximal doses in HSCT for cancer are nonmyeloablative. Although they cause neutropenia and lymphopenia, hematopoiesis recovers without stem cell reinfusion. Examples of drugs that are lymphoablative but nonmyeloablative when given at maximal doses include fludarabine, pentostatin, cladribine, and cyclophosphamide. Although these agents induce transient neutropenia that recovers without stem cell support, HSC infusion shortens the neu-

tropenic interval. Other agents such as antilymphocyte antibodies cause lymphopenia without any neutropenic side effects. Common available antilymphocyte antibodies include rituximab (anti-CD20 antibody against B cells), equine ATG (polyclonal antibodies against T cells), rabbit ATG (polyclonal antibodies against T and B cells), and alemtuzumab (anti-CD52 antibody against T and B cells and macrophages).

Finally, there are numerous myeloablative agents used in the HSCT conditioning regimen for cancer that are not myeloablative when given at lower doses, including numerous chemotherapeutic drugs such as busulfan, etoposide, mitoxantrone, cytarabine, and the like. Myeloablative regimens utilize total body irradiation (TBI) and/or employ potentially myeloablative chemotherapeutic drugs at doses above the ability of hematopoiesis to recover. On the other hand, lymphoablative but nonmyeloablative protocols utilize antilymphocyte antibodies, nonmyeloablative chemotherapy (cyclophosphamide, fludarabine) at maximal doses, and/or potentially myeloablative agents but at reduced, nonmyeloablative doses.

Independent of using a myeloablative or nonmyeloablative regimen, some guiding principles should be considered in selection of the conditioning agents. Conditioning drugs should be selected to avoid further treatment-related injury to already-damaged organ systems that the treatment is designed to salvage. For example, treatments such as busulfan (16) and TBI (17) cause pulmonary fibrosis and should be avoided whenever possible in immune-mediated diseases complicated by pulmonary fibrosis, such as systemic sclerosis (SSc) (18). Similarly, since vascular injury from TBI is predominantly confined to the microvascular compartment (19,20), TBI should be used with caution in immune-mediated diseases involving microvasculature damage, such as SSc, a disease marked by microvascular damage and capillary dropout. The pharmacokinetics of most conditioning regimen agents in patients with renal and/or liver dysfunction are unknown, since dose-escalation pharmacokinetic studies in cancer were performed in patients with normal organ function. Using these agents in patients with liver and/or renal insufficiency in which drug clearance is unknown may alter drug half-life and consequently increase toxicity. For example, fludarabine is known to cause irreversible and potentially disabling cerebellar damage in cancer patients with renal insufficiency (21). Irreversible ataxia, movement disorders, and/or persistent nausea and vomiting may occur in systemic lupus erythematosus (SLE)

patients with nephritis receiving conditioning regimens containing high doses of fludarabine.

Some patients undergoing autologous HSCT receive grafts that have been enriched or purified *ex vivo* for HSCs, usually by selection for CD34, a hematopoietic progenitor cell marker. The rationale for CD34+ selection is to prevent reinfusion of lymphocytes that may precipitate disease relapse. The only study evaluating infusion of a CD34+-selected versus an unmanipulated graft compared 2 cohorts of patients with rheumatoid arthritis (RA) undergoing HSCT in Australia (22). The group receiving unselected HSCs (i.e., contaminated with lymphocytes) had a trend toward longer duration of clinical improvement (22). This was contrary to the expected outcome, because CD34+ selection is designed to increase duration of remission by removing disease-causing lymphocytes. One potential explanation is that CD34+ selection also removed disease-controlling Treg. Except for the Australian RA study, there are no reported comparisons of infusing a CD34+-selected versus an unmanipulated autologous graft. Therefore, whether CD34+ selection decreases or increases the relapse rate remains unclear. However, combining CD34+ selection with very intensive immunosuppressive (myeloablative or lymphoablative) regimens will increase the risk of posttransplant opportunistic infections.

A direct comparison of toxicity and efficacy between myeloablative and lymphoablative transplant regimens in autoimmune diseases has never been performed. A multicenter retrospective EBMT/EULAR registry analysis of conditioning regimen intensity comparing mild regimens such as cyclophosphamide and ATG, moderately intensive regimens such as BEAM (carmustine, etoposide, cytarabine, and melphalan), and intensive conditioning such as TBI-containing regimens or dose-intense myeloablative chemotherapy showed increased treatment-related mortality with more intensive regimens and, conversely, a tendency toward higher relapse rates with less intensive regimens (2). However, dose intensity does not necessarily separate myeloablative regimens from lymphoablative regimens, since lymphoablative regimens may be designed to be as intensive as or even more immunosuppressive than myeloablative regimens. Finally, even if relapse occurs, it has been reported that the disease is much easier to control with standard therapy to which the autoimmune disorder had been previously refractory (23).

Reports on regimen dose intensity are from uncontrolled multicenter retrospective registry data in which disease-specific eligibility, treatment approaches,

standard-of-care guidelines, and disease-specific definitions of remission were not standardized across centers. Thus, to date, questions remain unanswered regarding issues such as treatment-related mortality, overall survival, disease-free survival, severity of relapsed disease, duration of second remission, and quality of life after lymphoablative compared with myeloablative regimens. Given this lack of knowledge, randomized trials have been developed based principally on the investigator's bias toward either myeloablative or lymphoablative regimens.

Mechanism of remission with HSCT

Tolerance occurs both centrally in the thymus during new T cell development and after T cells have matured in the periphery. Developing T cells need positive signals to expand and survive. If the T cell receptor (TCR) does not recognize self major histocompatibility complex (MHC) in the thymus, the developing T cell dies by apoptosis through "neglect." Survival signals are provided by low-affinity interactions of the TCR with MHC displaying self peptides. Thus, T cells are in a sense selected for recognition of self. However, it is important that the developing T cells are not autoreactive and are not activated by the interaction with self peptides.

Thymic education that selects for repertoires with mild-to-moderate self recognition results in an autoimmunity paradox. Autoimmune cells are physiologic while autoimmune disease is pathologic. Autoreactive cells are normal while autoimmune disease is abnormal. Unlike a malignancy, in which any surviving tumor cell is pathologic, in autoimmune disease, autoimmune cells, which the treatment is designed to destroy, must be regenerated following HSCT. It may not be the autoimmune cells *per se* but rather the interaction between these cells or an abnormal precursor frequency of certain autoimmune cells that causes autoimmune disease. The analogy would be a dysfunctional work environment with self-destructive interpersonal relationships. Each person may be intrinsically good, yet that combination of individual social interactions is pathologic. Similarly, the immune system functions as a team of cells communicating with each other. It is this communication, not an individual cell *per se*, which may be pathologic in environmentally induced autoimmune disorders (12,13).

Evaluation of immune reconstitution following HSCT has been performed on a limited number of patients with SSc, SLE, multiple sclerosis (MS), and RA. In patients with SSc, 4 patients with a favorable response

to HSCT were compared with 3 patients with either no response or with relapse (24). The T cell repertoire remained skewed, with similar expansions before and after HSCT in both groups for 1 year. In SSc, persistent skewing of the T cell repertoire pre- and post-HSCT independent of disease response may suggest that antigen-driven expansion of both surviving peripheral and thymic regenerating T cells may be unrelated to disease manifestations (24). Assessment of immune reconstitution in MS following HSCT revealed different results from 2 different studies, both of which used myeloablative regimens. Muraro et al demonstrated an immune reset following HSCT for MS. Post-HSCT immune reconstitution was accompanied by an increase in thymic-derived naive T cells, decreased central memory T cells, increased output of recent thymic emigrants, and recovery of a diverse TCR repertoire (25). In another study, using an even more intensive myeloablative regimen, Sun et al failed to find evidence for immune reset. The functional and phenotypic properties and clonal composition of the reconstituted T cells did not seem to differ significantly from the original pre-HSCT immune system (26).

In autoimmune disorders, peripheral blood and/or tissue cytokine profiles may be altered. Following HSCT in patients with SLE, peripheral blood Th2 skewing normalized, with return of appropriate Th1:Th2 ratios (27,28). Following HSCT, interleukin-7, a glycoprotein involved in both thymic and extrathymic T cell stimulation, appeared to be deficient in patients with RA compared with patients undergoing HSCT for cancer (29). This suggests that immune reconstitution could be prolonged or delayed following HSCT in patients with RA compared with patients with cancer or other autoimmune disorders. Finally, patients with either Crohn's disease or juvenile idiopathic arthritis had a significant increase in thymic-derived Treg (CD4+, CD25+, foxp3+) following HSCT (30,31). Treg down-regulate autoimmune disease activity in several animal models (32).

In summary, immune reset following HSCT may occur through 1) loss or decreased frequency of specific autoreactive clones with reconstitution of a normal diverse repertoire, 2) normalization of imbalanced cellular and/or soluble networks (antibodies, cytokines), and/or 3) increase in immunosuppressive regulatory cells and networks. It is likely that equally intensive immunosuppressive lymphoablative and myeloablative regimens will result in a similar post-HSCT immune reconstitution, but this has yet to be tested. It remains to be clarified whether post-HSCT immune reconstitution

varies with factors such as type of autoimmune disease, patient age, or continued use of posttransplant immunosuppressive drugs. Similarly, the effect of delayed thymic reconstitution on duration of autoimmune disease remission following HSCT is unknown.

Randomized trials

Table 1 summarizes the reported nonrandomized HSCT trials (33–41) that form the basis of randomized controlled trials in SLE and SSc. Consistent with EBMT/EULAR registry reports, results of these trials imply lower treatment-related mortality with lymphoablative regimens.

SLE randomized trial. There is currently 1 randomized trial of autologous HSCT for SLE, termed the Lupus Immunosuppressive/Immunomodulatory Therapy or Stem Cell Transplant (LIST) trial. It is a study of a nonmyeloablative regimen designed for lymphoablation using cyclophosphamide and rabbit ATG conditioning with infusion of CD34+-enriched autologous HSCs. The trial is based on experience using nonmyeloablative regimens in Genoa, Italy by Marmont et al (42), in Berlin, Germany by Rosen et al (43), and in 50 patients with SLE treated over an 8-year interval at Northwestern University (34,44). HSCT was used as a salvage therapy for patients with active visceral disease that was refractory to standard therapy. In the Northwestern University SLE HSCT trial, the probability of disease-free remission (as defined by the Responder Index for Lupus Erythematosus criteria [45]) at 5 years is 50% (34).

The LIST regimen was selected because cyclophosphamide is a standard and effective drug for lupus, and dosage can be escalated to maximum tolerated doses without myeloablation. Due to uncertain cyclophosphamide metabolism in patients with renal insufficiency, for patients with significant renal impairment, dialysis is performed each morning following cyclophosphamide administration. Patients will be randomly assigned either to autologous HSCT or to a control arm of continued standard of care. A crossover to HSCT for continued active disease with the control arm is not included in the study. Eligibility criteria for entry are corticosteroid dependency and 1 of the following: 1) continued active nephritis after 6 months of intravenous cyclophosphamide, 2) other continued active visceral organ involvement after 3 months of oral or intravenous cyclophosphamide, or 3) continued active cytopenias or mucocutaneous disease or arthritis/myositis after at least 3 months of cyclophosphamide, azathioprine, mycophe-

Table 1. Reported mortality in patients undergoing autologous hematopoietic stem cell transplantation for SLE and SSc*

Author, year (ref.), type of trial	Transplant regimen: treatment (no. or % of patients)	No. of patients/no. of deaths
SLE		
Burt et al, 2006 (34), single center	Lymphoablative: CYC/ATG	50/8 (1 [2%] treatment related, 7 [14%] disease related)
Jayne et al, 2004 (33), EBMT/EULAR registry multicenter	Mixture of myeloablative and lymphoablative: CYC/ATG (48%), CYC/ATG/TLI (21%), CYC/TT (6%), CYC/melphalan (4%), CYC (4%), BEAM (4%), melphalan/etoposide (4%), BEAM/ATG (4%), CYC/fludarabine/ATG (2%), ATG (2%), busulfan/fludarabine/melphalan (2%)	53/12 (7 [13%] treatment related, 5 [11%] disease related)
SSc		
Tsukamoto et al, 2006 (41), single center	Lymphoablative: CYC	6/0
Oyama et al, 2005 (40), single center	Lymphoablative: CYC/ATG	10/1 (1 [10%] disease related)
Farge et al, 2004 (38), EBMT/EULAR registry multicenter	Mixture of myeloablative and lymphoablative: CYC (35), CYC/ATG (12), CYC/alemtuzumab (3), CYC/ATG/TLI (1), CYC/TLI (1), other (5)	57/13 (5 [9%] treatment related, 8 [14%] disease related)
McSweeney et al, 2004 (36), multicenter	Myeloablative: TBI/CYC/ATG	33/10 (5 [15%] treatment related, 5 [15%] disease related)
Farge et al, 2002 (39), single center	Mixture of myeloablative and lymphoablative: CYC (9), ALG/melphalan (1), ALG/CYC (1)	11/4 (1 [9%] treatment related, 3 [27%] disease related)†
McSweeney et al, 2002 (35), multicenter	Myeloablative: TBI/CYC/ATG	19/4 (3 [16%] treatment related, 1 [5%] disease related)
Binks et al, 2001 (37), multicenter	Mixture of myeloablative and lymphoablative: CYC (19), CYC/ATG/TBI (9), CYC/ATG (4), CYC/alemtuzumab (2), CYC/TLI (1), other (2), not transplanted (4)	41/11 (7 [17%] treatment related, 4 [10%] disease progression related)

* SLE = systemic lupus erythematosus; SSc = systemic sclerosis; CYC = cyclophosphamide; ATG = antithymocyte globulin; EBMT/EULAR = European Bone Marrow Transplant/European League Against Rheumatism; TLI = total lymphoid irradiation; TT = thiotepea; BEAM = carmustine, etoposide, cytarabine, and melphalan; TBI = total body irradiation; ALG = antilymphocyte antibody.

† The 1 treatment-related death occurred with the myeloablative regimen.

nolate mofetil, or cyclosporine. Continued active disease is equivalent to group A disease according to the British Isles Lupus Assessment Group (46).

SSc randomized trials. Three randomized trials of autologous HSCT for the treatment of SSc are under way. These are the ASTIS (Autologous Stemcell Transplantation International Scleroderma) (47), ASSIST (American Scleroderma Stem Cell versus Immune Suppression Trial), and SCOT (Scleroderma: Cyclophosphamide or Transplantation) trials. The ASTIS and ASSIST trials are designed to be lymphoablative, whereas the SCOT trial is myeloablative. The European ASTIS trial uses a conditioning regimen of cyclophosphamide and rabbit ATG, along with CD34+ selection. The protocol is based on experience with similar regimens at Hospital Saint Louis, Paris, France (39) and at Leiden University Medical Center, Leiden, The Netherlands (38). The American ASSIST trial is similar but utilizes cyclophosphamide and rabbit ATG without manipulation or CD34+ selection of the graft. Both of these protocols incorporate cyclophosphamide and rab-

bit ATG, which have been used and are well tolerated in SSc (48,49).

Because of the occurrence of cardiopulmonary deaths among SSc patients with pulmonary arterial hypertension (PAH) treated with HSCT in European studies, PAH is considered to be a contraindication for HSCT. To date, the ASTIS trial has enrolled more than 60 patients, and no treatment-related deaths have occurred (Farge D: personal communication, January 2006). The ASSIST trial is based on phase I experience with 10 SSc patients treated at Northwestern University (40). In that study and in a similar Japanese study (41), no transplant-related deaths occurred, and improvement in skin involvement measured by the Rodnan skin score (50) was observed along with stabilization or improvement in pulmonary function.

The SCOT trial uses a myeloablative regimen of cyclophosphamide, TBI, ATG, and CD34+ cell selection. The protocol is based on a phase I study of 33 patients, mostly performed at the Fred Hutchinson Cancer Center (Seattle, WA) (35,36). The preliminary

Table 2. Randomized controlled trials of autologous HSCT for autoimmune diseases*

Regimen type, trial	Disease	Conditioning regimen	Control	Crossover to HSCT if failure on control arm
Lymphoablative				
LIST	SLE	CYC, rabbit ATG, CD34 selected	Continued standard of care	No
ASTIS	SSc	CYC, rabbit ATG, CD34 selected	Monthly CYC	No
ASSIST	SSc	CYC, rabbit ATG	Monthly CYC	Yes
KISS	Crohn's disease	CYC, rabbit ATG	Continued standard of care	Yes
MIST	MS	CYC, rabbit ATG	Any FDA-approved therapy†	Yes
Myeloablative				
SCOT	SSc	TBI, CYC, rabbit ATG, CD34 selected	Monthly CYC	No
ASTIMS	MS	BEAM, rabbit ATG	Mitoxantrone	No

* A European randomized controlled lymphoablative trial for rheumatoid arthritis termed ASTIRA (Autologous Stem-cell Transplantation International Rheumatoid Arthritis) was closed due to competing therapies. A European randomized controlled lymphoablative trial for Crohn's disease termed ASTICS (Autologous Stem-cell Transplantation International Crohn's Syndrome) is in development. HSCT = hematopoietic stem cell transplantation; LIST = Lupus Immunosuppressive/Immunomodulatory Therapy or Stem Cell Transplant (NCT00230035 at www.clinicaltrials.gov); ASTIS = Autologous Stemcell Transplantation International Scleroderma (www.astistrial.com); ASSIST = American Scleroderma Stem Cell versus Immune Suppression Trial (NCT00278525 at www.clinicaltrials.gov); KISS = Crohn's (K) Immune Suppression versus Stem Cell (NCT00271947 at www.clinicaltrials.gov); MIST = Multiple Sclerosis International Stem Cell Transplant (NCT00273364 at www.clinicaltrials.gov); MS = multiple sclerosis; FDA = Food and Drug Administration; SCOT = Scleroderma: Cyclophosphamide or Transplantation (NCT00114530 at www.clinicaltrials.gov); ASTIMS = Autologous Stem Cell Transplant International Multiple Sclerosis (www.astims.org). See Table 1 for other definitions.

† Examples include interferon (IFN) beta-1a, IFN beta-1b, mitoxantrone.

results of this study showed that the Rodnan skin score improved in the majority of patients. However, 10 of 33 patients died (5 from the myeloablative regimen and 5 from disease progression). An additional 4 patients showed disease progression by skin score. Although not defined as disease progression, an additional 5 patients developed renal insufficiency, including 2 with end-stage renal disease, and the majority of the surviving patients, despite lung shielding, had deterioration in diffusing capacity for carbon monoxide (36).

A major difference between the myeloablative and lymphoablative HSCT regimens used in SSc is the use of TBI. Radiation raises several concerns in patients with SSc. First, radiation may have deleterious effects on the microvasculature. Diffuse small-vessel vasculopathy is a hallmark of SSc, as demonstrated by loss of small capillaries on nailfold microscopy. The primary target of radiation-related vascular toxicity is the microvascular compartment, and postradiation capillary dropout has been reported to occur months to years after radiation exposure (51). Even at very low doses, radiation can cause microvascular damage. A recent study on the effect of occupational radiation exposure compared capillary integrity in a cohort of healthy physicians (e.g., interventional cardiologists) exposed to radiation at doses considered safe in the workplace with that in a control cohort of physicians (e.g., internists) without occupational exposure to radiation (20). Physicians with workplace exposure had evidence of significant damage to nailbed capillaries compared with physicians without

exposure to low-level radiation. The difference was statistically significant when corrected for other possible confounding variables including age, sex, glucose level, hypertension, and smoking (20).

Radiation exposure has been repeatedly linked to progression or worsening in patients with SSc (52–55). High doses (4,500–5,400 rads to the breast and a 1,000-rad boost to the tumor bed) used to treat breast cancer have been linked to severe cutaneous and pulmonary fibrosis both within the radiation port and within the contralateral nonirradiated lung. For this reason, many experts consider conservative therapy (i.e., lumpectomy and breast irradiation) to be contraindicated in SSc patients with localized breast cancer (56,57). One study evaluated the efficacy of total nodal irradiation at lower doses (1,800 rads) for SSc. After enrolling only 6 patients (3 treated with total nodal irradiation and 3 controls), the study was terminated because all 3 total nodal irradiation-treated patients showed deterioration in pulmonary function (despite lung shielding), and 1 died from worsening gastrointestinal involvement (58).

In summary, there is widespread concern over use of an intervention in the conditioning regimen (radiation) that is known to damage the predominant organ system (microvascular compartment) affected in SSc (19,59). Disagreement over the use of TBI in SSc and over the rationale for HSCT (myeloablation versus lymphoablation) has led to the development of 2 distinct randomized autologous HSCT trials in the US: the SCOT trial, which includes TBI and uses a myeloablative

regimen, and the ASSIST trial, which uses a nonmyeloablative regimen and is similar to the European ASTIS trial; neither the ASSIST trial nor the ASTIS trial uses radiation. The 3 trials (ASTIS, ASSIST, and SCOT) have comparable eligibility criteria (high Rodnan skin score and visceral organ involvement) and control arms (monthly intravenous cyclophosphamide). Only 1 trial (ASSIST) allows crossover to HSCT if there is continued disease progression in the control arm.

Summary of HSCT randomized trials. In order to be complete, all current HSCT randomized controlled trials for autoimmune disorders including SLE, SSc, Crohn's disease, and MS are listed in Table 2. Only lymphoablative regimens are being offered to treat Crohn's disease, and concerns over the use of a myeloablative versus a lymphoablative regimen for MS, a disease with mortality similar to that in the general population, have already been reported in the neurology literature (60).

Summary

While autologous HSCT appears to induce remissions and improve quality of life in the majority of patients, it remains unclear whether either myeloablative or lymphoablative autologous HSCT can cure autoimmune disorders, and randomized controlled trials are needed to confirm the benefit and cost-effectiveness of this therapy. There are currently 2 disparate philosophical approaches to the design of these trials. One approach is to apply aggressive malignancy-specific myeloablative regimens. Although more intensive lymphoablative regimens could be designed, since cancer-specific myeloablative regimens are more intensive than currently utilized lymphoablative regimens, myeloablative regimens may result in longer duration of disease remission. The other philosophical approach is to use lymphoablative regimens that are disease specific and nonmyeloablative (i.e., do not irreversibly damage the hematopoietic stem cell compartment), maximize drugs already used to treat each disease, and avoid agents in the regimen that could cause further damage to already disease-injured organs. It appears that lymphoablative regimens are safer than myeloablative regimens, and it appears that even if relapse occurs, durable long-term remission may be achieved with conventional treatment to which the patient's disease was previously refractory.

If considered on a disease-specific basis, the high disease-related mortality of SSc could justify the higher treatment-related mortality associated with a myeloablative regimen. However, since radiation is already

considered a relative contraindication for SSc and radiation-induced and SSc-related microvascular compartment injuries are similar, and because TBI is associated with late myelodysplasia, leukemia, and lymphomas, some investigators have voiced concern over use of a TBI-based myeloablative regimen to treat SSc (19,59).

The controversy that surrounds the competing visions of lymphoablative versus myeloablative autologous HSCT regimens for autoimmune disease has yet to be resolved. Further experience will likely indicate the superiority of one of these approaches when both safety and efficacy are considered. Since both lymphoablative and myeloablative protocols are being offered for a select group of severely ill patients with autoimmune disorders refractory to conventional therapy, subspecialists who refer patients for such therapy should be aware of the emerging body of literature that at the very least suggests that lymphoablative regimens will be associated with less morbidity and mortality. Sharing of such information is crucial if patients are to provide consent that is truly informed.

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