Future Strategies in Hematopoietic Stem Cell Transplantation for Rheumatoid Arthritis

RICHARD K. BURT, WALTER BARR, YU OYAMA, ANN TRAYNOR, and SHIMON SLAVIN

ABSTRACT. Patients with coincidental rheumatoid arthritis (RA) treated by allogeneic hematopoietic stem cell transplantation (HSCT) for drug induced aplastic anemia have been fortuitously cured of RA. Other than these examples with allogeneic HSCT, there is no known curative therapy for RA. Despite its potential to cure, allogeneic transplantation is not being offered to patients with RA due to transplant related mortality. Advances in HSCT conditioning regimens and better prevention of graft-versus-host disease should allow consideration of allogeneic HSCT as therapy for severe RA. We propose a new, well tolerated, nonmyeloablative allogeneic stem cell transplant regimen as treatment for RA. (J Rheumatol 2001;28 Suppl 64:42–8)

Key Indexing Terms:
RHEUMATOID ARTHRITIS HEMATOPOIETIC STEM CELL TRANSPLANTATION

IMMUNOLOGY OF RHEUMATOID ARTHRITIS

Patients with coincidental rheumatoid arthritis (RA) treated by allogeneic hematopoietic stem cell transplantation (HSCT) for drug induced aplastic anemia have been fortuitously cured of RA[1-4].

RA is an inflammatory disease of joints but may have extraarticular manifestations including rheumatoid nodules, interstitial pneumonitis, and vasculitis[5,6]. The etiology of RA remains elusive. It is an immune mediated disease but whether the response is directed against an infectious agent or against an unknown self-epitope (i.e., autoimmune) or both is unknown. If there is a predominate disease mediating cell (T or B lymphocyte, macrophage or synoviocyte), its identity also remains obscure.

Like many autoimmune diseases, RA is associated with particular HLA genotypes[7]. Severe RA has been associated with MHC class II DR4. Five RA prone alleles (DRB1*0401, DRB1*0404, DRB1*0405, and DRB1*0101), whose frequencies vary for different ethnic groups, all share a similar amino acid epitope sequence (LLEQKRAA or LLEQRRAA) encoded by codons 67–74[8,9]. While the prevalence of RA in the general population is 1 in 100 people, a heterozygote with any one of these alleles has an increased risk for RA varying from 1 in 20 to 1 in 80[10]. In an individual who has 2 HLA alleles associated with RA (DRB1*0401 and DRB1*0404) the risk of developing RA is 1 in 7[10].

The HLA sequence 67–74 on RA associated alleles is a HLA contact site for both peptide and T cell receptor binding. This suggests HLA presentation of a common infectious or self-antigen to T cells is involved in the pathogenesis of RA. An infection may lead to RA through molecular mimicry, bystander effects, or epitope spreading[11-15]. Molecular mimicry arises from cross reactive T cell responses to peptides that have similar homology[11]. The immune response initiated against an infectious agent may cross react with a self-peptide, leading to autoimmunity. Alternatively, inflammatory reactions against an infection may lead to bystander damage of normal tissues. Presentation of self-peptides by activated antigen presenting cells then precipitates the autoimmune disease. Once an immune process starts, T cell responsiveness can spread to other epitopes on the same or different peptides, a phenomenon termed determinant or epitope spreading[12-15].

While MHC genes predispose to RA, the majority of patients with RA associated MHC genes remain disease-free. Environment and/or non-MHC genes must, therefore, contribute towards development of disease. Adjuvant arthritis (AA) and collagen induced arthritis (CIA) in rats are models for RA and are induced by injection of adjuvant or collagen and adjuvant, respectively[16,17]. At least 14 genomic intervals or loci (Cia1 to Cia14) are associated with CIA[18,19]. The MHC loci correlate with Cia1. Putative genes associative with non-MHC Cia regions include cytokines such as interleukin 11 and transforming growth factor and growth regulating oncogenes such as B cell leukemia/lymphoma3 (BCL3) and Bcl-2 associated X protein (BAX)[20,21]. Mice with either AA or CIA, in which environmental stimuli are necessary to induce arthritis, may be cured by either autologous, syngeneic, or allogeneic hematopoietic stem cell transplantation[22-25]. The lowest

From the Division of Immunotherapy and Autoimmune Disease, and the Division of Rheumatology, Northwestern University, Chicago, Illinois, USA; and Department of Bone Marrow Transplantation, Hadassah University Hospital, Jerusalem, Israel.

R.K. Burt, MD, Division of Immunotherapy and Autoimmune Disease; W. Barr, MD, Division of Rheumatology, Northwestern University; Y. Oyama, MD; A. Traynor, MD, Division of Immunotherapy and Autoimmune Disease, Northwestern University; S. Slavin, MD, Department of Bone Marrow Transplantation, Hadassah University Hospital.

Address reprint requests to Dr. R.K. Burt, Division of Immunotherapy and Autoimmune Disease, 250 East Superior, Room 162, Chicago, IL 60611; E-mail:rburt@nwu.edu

relapse rate occurs in animals receiving an allogeneic transplant. Relapse after an autologous HSCT correlates inversely with intensity of the immune suppression conditioning regimen in various models of autoimmune diseases.

While RA may be an autoimmune disease, in some ways, it may be viewed as similar to a malignant lymphoproliferative disorder. Hallmarks of malignancy are loss of growth inhibition, oncogene mutations, and clonality. Fibroblast-like synoviocytes from patients with RA show autonomous *ex vivo* proliferation[26,27], a behavior that may contribute to *in vivo* cartilage destruction. One possible mechanism by which cells escape growth inhibition is oncogene mutation. Somatic mutations of the p53 oncogene occur *in vivo* within involved joints and *ex viv*o in RA synoviocyte cultures[28-30]. Skewing of B lymphocyte immunoglobulins and T cell receptors suggests clonal expansion of both B and T lymphocytes within inflamed RA synovium[31-33]. Similarly to a malignancy, RA can cause severe disability and pain, and can shorten life expectancy[34-44]. In summary, the immunology of RA intersects traditional aspects of rheumatology, autoimmunity, malignancy, and possibly infectious diseases.

RESULTS OF AUTOLOGOUS HSCT TRIALS FOR RA

If RA is caused by environmental exposure, brief but dose intense immune suppression and autologous HSCT may ablate the autoreactive immune cells and allow regeneration of a new immune system. Several patients have undergone autologous transplantation for lymphoma who had coincidental RA[45-47]. The autografts were not purged of lymphocytes, and the transplants were not tailored as therapy for RA. Nevertheless, short term complete responses were observed for 4 months, 20 months, and at last followup greater than 19 months[45-47].

Since the rationale for autologous HSCT is to use high dose chemotherapy to attempt ablation of the immune system, autologous HSCT allows maximization of immune suppression beyond otherwise marrow-limiting toxicity. Autologous stem cells are infused to reinitiate hematopoiesis and regenerate immunity. Based on testing the premise of dose intense immune suppression, several centers have reported early posttransplant outcomes when RA is the only indication for autologous transplantation (Table 1)[48-54]. In general, the procedure has been well tolerated without mortality. HSCT offers an almost immediate relief of symptoms. Patients become pain-free, sometimes for the first time in years. Activities required for daily living, such as buttoning a shirt or combing hair, rapidly return to normal. Morning stiffness resolves, rheumatoid nodules disappear, sedimentation rate normalizes, and rheumatoid factor may disappear.

Most of the protocols did not, however, increase immune suppression to the point of myeloablation. In the most common regimen used (200 mg/kg cyclophosphamide with or without antithymocyte globulin), hematopoiesis would recover even without stem cell infusion. While these studies showed that relatively high dose chemotherapy was well tolerated with marked American College of Rheumatology (ACR) improvements (ACR 50 or ACR 70), a complete remission was unusual and relapse within 2 years is common. There are suggestions of a dose-response effect. A dose escalation study of cyclophosphamide at 100 mg/kg revealed transient 1–2 month responses, but at 200 mg/kg response duration increased to 18–20 months[50]. The few regimens that were myeloablative (busulfan and cyclophosphamide) seem to indicate more durable remissions[51].

For an intense and expensive treatment such as HSCT to be considered for RA, sustained complete remissions or 70% improvement as defined by the ACR must be achieved[55]. Several modifications are being considered, including: (1) using the current easily tolerated nonablative yet highly immunosuppressive regimen with posttransplant immune modulation, e.g., a tumor necrosis factor (TNF) inhibitor, cyclosporine A, and/or methotrexate, (2) using a more intense myeloablative regimen such as busulfan and cytoxan, or (3) performing allogeneic HSCT.

One approach is to advance the current nonmyeloablative stem cell transplants (Table 2) into phase III studies, but to decrease the high relapse rate, add chronic posttransplant immune suppression. This approach assumes that the posttransplant disease is easier to control with immune suppression than prior to transplant. The risk of infections, which are one of the major causes of death for patients with RA, may increase with posttransplant immune suppression. Implicit in this philosophy is acceptance of RA as an incurable but chronic and controllable disease.

A second approach is to view RA as a disease that is potentially curable by reinduction of self-tolerance through autologous HSCT. Since nonmyeloablative regimens induced remission of refractory disease and a dose-response effect may exist, phase I/II pilot studies using more intense myeloablative regimens will be initiated in the hope of inducing more durable remissions. A proposed regimen includes fludarabine 25 mg/kg/day × 5 days plus oral busulfan (4 mg/kg/day × 3 days) or intravenous Busulfex (intravenous busulfan) (3.2 mg/kg/day × 3 days) versus a regimen of cyclophosphamide (120–200 mg/kg) and Busulfex (3.2 mg/kg/day × 3 days) (Table 3). If a high percentage of patients continue to relapse following a myeloablative regimen, an autologous transplant will probably be unlikely to cure and the regimen related toxicity of more intense conditioning regimens would probably be unacceptable.

ALLOGENEIC HSCT OF RA

Anecdotal case reports of patients transplanted for aplastic anemia who had coincidental RA suggest that an allogeneic

Table 1. Reports of autologous or syngeneic hematopoietic stem cell transplantation for RA.

Disease	Conditioning	Comment
RA[49,52]	CY, ATG (autograft 2-3 log lymphocyte depleted)	Marked improvement up to 18 mo, but 2 relapsed
RA[48]	CY (autograft not lymphocyte depleted)	Marked improvement 6 mo followup
RA[50]	CY (autograft not lymphocyte depleted)	*Cohort I — CY 100 mg/kg, response for 1–2 mo *Cohort II — CY 200 mg/kg — improved for 17–19 mo
RA[54]	CY, ATG (autograft not lymphocyte depleted)	Relapsed at 5 and 7 mo
RA[51]	Bu, CY (autograft lymphocyte depleted)	Remission > 10 mo
RA[53]	CY, ATG (identical twin)	Remission > 24 mo

RA: rheumatoid arthritis; CY: cyclophosphamide; Bu: busulfan; ATG: antithymocyte globulin.

Table 2. Possible nonmyeloablative autologous stem cell transplantation.

Day –7	–6	–5	–4	–3	–2	–1	0*
Fludara	Fludara CY	Fludara CY	Fludara ATG	ATG	ATG	ATG	CD34
Day –7	–6	–5	–4	–3	–2	–1	0*
Fludara	Fludara CY	Fludara CY	Fludara C-1H	C-1H	C-1H	C-1H	CD34 or PBSC

* Some posttransplant immune modulation such as a TNF inhibitor, cyclosporine, or methotrexate will be used as maintenance.
CY: cyclophosphamide, Fludara: fludarabine, C-1H: CAMPATH.

Table 3. Possible myeloablative autologous stem cell transplantation.

Day –8	–7	–6	–5	–4	–3	–2	–1	0*
Bu	Bu	Bu	CY	CY	CY	CY		CD34

* No posttransplant maintenance therapy. Bu: Busulfex (0.8 mg/kg q 6 h), CY: cyclophosphamide (50 mg/kg/day).

transplant may induce durable remissions (Table 4)[1-4]. Four were reported in the 1970s when transplant related mortality was higher than current standards[1]. Three died from transplant complications[1]. Four patients are in complete remission for 4–8 years after transplant despite discontinuation of all immune suppression[1-3]. In one patient, RA relapsed[4]. Hematopoietic chimerism at a molecular level using polymerase chain reaction for VNTR (variable number of tandem repeats) revealed 100% donor chimerism[4]. This does not necessarily suggest that relapse arose from donor immune cells. Evaluation of the involved organ system (i.e., joints) was not performed to determine if infiltrating lymphocytes, macrophages, and reactive synoviocytes were of donor or host origin. In addition, although clinically normal, the serologic status of the donor (e.g., rheumatoid factor) was not reported.

The rationale for allogeneic HSCT is to change the genetic susceptibility to disease and also provide an allogeneic graft versus autoimmunity (GVA) effect[56]. Hence, replacement of genetically susceptible host stem cells with resistant donor hematopoietic cells may prevent recurrence of the disease following transplantation even if the cause of RA is still present in the host. Perhaps more important is that it is very unlikely that the "last" self-reactive lymphocytes can be eliminated by intensive chemoradiotherapy alone, whereas after allogeneic bone marrow or blood stem cell

Table 4. Results of allogeneic hematopoietic stem cell transplantation in patients with aplastic anemia (AA) and RA.

Transplant Diagnosis	Autoimmune Disease	Type of Transplant	Outcome for Autoimmune Disease
AA[1]	RA	Allogeneic	Remission, died 93 days after transplant
AA[1]	RA	Allogeneic	Remission, died 75 days after transplant
AA[1]	RA	Allogeneic	Remission, died 58 days after transplant
AA[1]	RA	Allogeneic	Remission for > 2 yrs
AA[4]	RA	Allogeneic	Relapsed after 2 yrs
AA[2]	RA	Allogeneic	Remission for > 6 yrs
AA[3]	RA	Allogeneic	Remission for > 8 yrs

Table 5. Possible allogeneic transplant regimens for patients with RA.

Day –7	–6	–5	–4	–3	–2	–1	0	+1
Fludara	Fludara B/C	Fludara B/C	Fludara	Fludara				
							PBSC	
			CSA	→	→	→	→	→

Day –7	–6	–5	–4	–3	–2	–1	0	+1
Fludara C-1H	Fludara C-1H B/C	Fludara C-1H B/C	Fludara C-1H	Fludara				
							PBSC	
			CSA	→	→	→	→	→

T cell depleted (CD34+ enriched) graft								
Day –8	–7	–6	–5	–4	–3	–2	–1	0/+1
Fludara C-1H	Fludara C-1H B/C	Fludara C-1H B/C	Fludara C-1H	Fludara	Fludara			
								CD34+
				CSA	→	→	→	→

Day –6	–5	–4	–3	–2	–1	0
Fludara C-1H	Fludara B/C C-1H	Fludara B/C C-1H	Fludara B/C C-1H	Fludara C-1H		
						CD34+
		FK506/MMF	→	→	→	→

Fludara: fludarabine, C-1H: CAMPATH, CSA: cyclosporine, FK506/MMF: tacrolimus/mycophenolate mofetil, CD34+: infusion of lymphocyte depleted, i.e., CD34+ enriched, allogeneic stem cells, B/C: use of either busulfex or cyclophosphamide, PBSC: peripheral blood stem cells.

transplant all host hematopoietic cells can be replaced with donor hematopoietic cells, resulting in elimination by displacement of all self-reactive lymphocytes. Active cell mediated GVA effects may arise from mature donor lymphocytes eliminating, regulating, or inducing apoptosis of recipient autoreactive cells. A graft versus disease effect has already been established as the mechanism of remission for several hematologic malignancies, first discovered in 1981 and termed graft versus leukemia (GVL)[57,58]. The feasibility of alloreactive donor lymphocytes to eliminate all hematopoietic cells of host origin, including malignant lymphocytes that are fully resistant to all available chemoradiotherapy, was documented in early 1987[59-61]. In analogy, clinical evidence for GVA effects has recently been

reported[56]. While in theory a GVA effect may be beneficial, the most significant toxicity of allogeneic HSCT is an immunologic reaction of donor cells against normal host tissues, a complication known as graft-versus-host disease (GVHD). As a consequence, compared to autologous HSCT, allogeneic transplants may be complicated by higher morbidity and mortality, predominately due to GVHD. On the other hand, it was recently documented that hazardous myeloablative conditioning can be replaced with a much safer nonmyeloablative stem cell transplant, also known as mini-allograft[62-64]. Hence, it was documented that full and rapid engraftment of donor hematopoietic cells and parallel elimination of all host hematopoiesis can be accomplished by immunosuppressive rather than myeloablative conditioning, resulting in reduced procedure related toxicity and mortality[62-64]. Unfortunately, despite major improvement of the immediate outcome following HSCT, GVHD remains a major obstacle. It remains to be seen if the potential benefit of allogeneic HSCT justifies the risk of GVHD.

Based on the above, to be qualified for an allogeneic nonmyeloablative HSCT, candidates should have failed standard therapies and be in a high risk subset. There are individuals with RA who have a 5 year mortality of 30–70%, a rate higher than for triple vessel coronary artery disease or metastatic Hodgkin's lymphoma[65]. These patients may be identified by the number of involved joints or functional status as assessed by a questionnaire on activities of daily living (Health Assessment Questionnaire, HAQ). Therapies for RA include corticosteroids and disease modifying antirheumatic drugs such as hydroxychloroquine, azathioprine, gold, and methotrexate. TNF inhibitors (infliximab or etanercept) are promising new agents for RA[66-68]. The failure rate for TNF inhibitors is 25–40% and all patients relapse if therapy is stopped[67,68]. TNF inhibitors have been associated with demyelinating disease, a fair amount of serious and fatal infections, and a number of cases of myelosuppression. Candidates for HSCT should probably have failed combined TNF inhibitor and methotrexate — failure being defined as patients with active high risk disease such as 4–6 swollen joints, more than 20 involved joints, and being unable to answer more than 70% of the HAQ "with no difficulty." In summary, to justify the risk-benefit of allogeneic HSCT, candidates should be selected for refractory disease (despite TNF inhibitor and methotrexate) that are at high risk for RA related mortality (determined by number of involved joints and HAQ).

Transplant regimen related morbidity and mortality may be markedly diminished by safer conditioning of patients with nonmyeoloablative regimens designed to suppress the immune system enough to allow donor engraftment. Residual host hematopoiesis is eliminated by allogeneic donor immune cells. Of about 80 patients undergoing autologous HSCT, mortality is roughly 2% (Snowden J, personal communication). Regimen related mortality from a less intense nonmyeloablative HSCT regimen should, therefore, be under 2%. GVHD remains the major hurdle for safely performing allogeneic transplantation.

Allogeneic transplant regimens being considered include: (1) well tolerated conditioning involving the use of nonmyeloablative regimens with unmanipulated marrow cells, (2) nonmyeloablative regimens with unmanipulated peripheral blood stem cells (PBSC), or (3) nonmyeloablative regimens with $CD34^+$ enriched PBSC. In an unmanipulated nonmyeloablative HSCT, the incidence of extensive chronic GVHD remains controversial, but is probably 30–50%. Newer GVHD prophylactic agents currently under investigation, such as humanized anti-CD52 (CAMPATH-1H) given during conditioning, and newer posttransplant agents such as mycophenolate mofetil or TNF inhibitors are likely to diminish the incidence and severity of GVHD. This may be particularly important in RA, which is a disease of older individuals, especially considering that the risk of GVHD increases with age.

Lymphocyte depletion ($CD34^+$ enrichment) of an allograft will markedly decrease the risk of GVHD but increase the risk of graft failure. Engraftment is affected by intensity of the conditioning regimen, the number of donor stem cells infused, and the number of T cells facilitating engraftment. The risk of graft failure may be partially offset by maximizing donor stem cell numbers (target dose $\geq 10^7$ $CD34^+$ cells/kg recipient weight)[69]. To minimize the morbidity of allogeneic HSCT, nonmyeloablative, yet strongly immunoablative conditioning could be combined with $CD34^+$ enrichment of donor stem cells (Table 2). The regimen of CAMPATH-1H, cyclophosphamide, and fludarabine would be intensely immune suppressive but not myeloablative. In case the allograft is rejected, autologous hematopoietic reconstitution would be anticipated within 2 weeks, with at least transient improvement or resolution of RA. Indeed, a regimen currently being used to treat autoimmune diseases employs a nonmyeloablative transplant conditioning regimen (cyclophosphamide 200 mg/kg) without stem cell infusion[70]. If engraftment occurs with low level (microchimerism) or mixed chimerism, indicative of host-versus-graft tolerance, and the patient's RA returns, donor lymphocyte infusions could be given at escalating doses until full donor chimerism is accomplished or RA resolves. To minimize the risk of GVHD, donor lymphocyte infusions would have to be carefully titrated with gradual dose escalation over extended intervals to allow monitoring for GVHD onset[71]. Temporal separation of donor progenitor cell infusion from donor lymphocyte infusion may allow clarification of the contribution of stem cell genetics versus GVA in remission of RA.

Designing transplant protocols that minimize procedure related morbidity and mortality may offer potentially curative therapy to patients with RA. Future studies should focus on a curative procedure for patients with intractable RA.

Allogeneic stem cell transplantation should be considered for patients with a fully matched donor, focusing on adoptive allogeneic cell therapy for total elimination of self-reactive lymphocytes while using safe, reduced intensity conditioning.

REFERENCES

1. Baldwin JL, Storb R, Thomas ED, Mannik M. Bone marrow transplantation in patients with gold-induced marrow aplasia. Arthritis Rheum 1977;20:1043-8.
2. Jacobs P, Vincent MD, Martell RW. Prolonged remission of severe refractory rheumatoid arthritis following allogeneic bone marrow transplantation for drug-induced aplastic anaemia. Bone Marrow Transplant 1986;1:237-9.
3. Lowenthal RM, Cohen ML, Atkinson K, Biggs JC. Apparent cure of rheumatoid arthritis by bone marrow transplantation. J Rheumatol 1993;20:137-40.
4. McKendry RJ, Huebsch L, Leclair B. Progression of rheumatoid arthritis following bone marrow transplantation. A case report with a 13-year followup. Arthritis Rheum 1996;39:1246-53.
5. Hollingsworth JW. Non-articular complications of rheumatoid arthritis. Medical Times 1970;98:121-30.
6. Hart FD. Rheumatoid arthritis: extra-articular manifestations. II. BMJ 1970;2:747-52.
7. Nepom GT, Byers P, Seyfried C, et al. HLA genes associated with rheumatoid arthritis. Identification of susceptibility alleles using specific oligonucleotide probes. Arthritis Rheum 1989;32:15-21.
8. Gregersen PK, Silver J, Winchester RJ. The shared epitope hypothesis. An approach to understanding the molecular genetics of susceptibility to rheumatoid arthritis. Arthritis Rheum 1987;30:1205-13.
9. Wordsworth BP, Lanchbury JS, Sakkas LI, Welsh KI, Panayi GS, Bell JI. HLA-DR4 subtype frequencies in rheumatoid arthritis indicate that DRB1 is the major susceptibility locus within the HLA class II region. Proc Natl Acad Sci USA 1989;86:10049-53.
10. Nepom GT. HLA and rheumatoid arthritis. In: Lechler R, Warrens A, editors. HLA in health and disease. 2nd ed. London: Academic Press; 2000:181-5.
11. Albert LJ, Inman RD. Molecular mimicry and autoimmunity. N Engl J Med 1999;341:2068-74.
12. Tuohy VK, Yu M, Yin L, Kawczak JA, Kinkel PR. Regression and spreading of self-recognition during the development of autoimmune demyelinating disease. J Autoimmunity 1999; 13:11-20.
13. Vanderlugt CL, Begolka WS, Neville KL, et al. The functional significance of epitope spreading and its regulation by co-stimulatory molecules. Immunol Rev 1998;164:63-72.
14. Vanderlugt CL, Miller SD. Epitope spreading. Curr Opin Immunol 1996;8:831-6.
15. Lehmann PV, Forsthuber T, Miller A, Sercarz EE. Spreading of T cell autoimmunity to cryptic determinants of an autoantigen. Nature 1992;358:155-7.
16. Wooley PH. Animal models of rheumatoid arthritis. Curr Opin Rheumatol 1991;3:407-20.
17. Myers LK, Rosloniec EF, Cremer MA, Kang AH. Collagen-induced arthritis, an animal model of autoimmunity. Life Sci 1997;61:1861–78.
18. Remmers EF, Longman RE, Du Y, et al. A genome scan localizes five non-MHC loci controlling collagen-induced arthritis in rats. Nature Genetics 1996;14:82-5.
19. Griffiths MM, Wang J, Joe B, et al. Identification of four new quantitative trait loci regulating arthritis severity and one new quantitative trait locus regulating autoantibody production in rats with collagen-induced arthritis. Arthritis Rheum 2000;43:1278-89.
20. Wilder R, Remmers E, Kawahito Y, Gulko P, Cannon G, Griffiths M. Genetic factors regulating experimental arthritis in mice and rats. In: Theofilopoulos AN, editor. Genes and genetics of autoimmunity. Vol. 1. Basel: Karger; 1999:121–65.
21. Rapoport M, Mor A, Bistritzer T, et al. Protooncogenes and autoimmunity. Israel J Med Sci 1997;33:262-6.
22. Slavin S. Treatment of life threatening autoimmune diseases with myeloablative doses of immunosuppressive agents and autologous bone marrow transplantation — rationale and experimental background. Bone Marrow Transplant 1993;12:85-8.
23. van Bekkum DW. Stem cell transplantation in experimental models of autoimmune disease. J Clin Immunol 2000;20:10-6.
24. van Bekkum DW. Conditioning regimens for the treatment of experimental arthritis with autologous bone marrow transplantation. Bone Marrow Transplant 2000;25:357-64.
25. Kamiya M, Sohen S, Yamane T, Tanaka S. Effective treatment of mice with type II collagen induced arthritis with lethal irradiation and bone marrow transplantation. J Rheumatol 1993;20:225-30.
26. Gay S, Gay RE, Koopman WJ. Molecular and cellular mechanisms of joint destruction in rheumatoid arthritis: two cellular mechanisms explain joint destruction? Ann Rheum Dis 1993;52 Suppl 1:S39-47.
27. Firestein GS. Invasive fibroblast-like synoviocytes in rheumatoid arthritis: passive responders or transformed aggressors? Arthritis Rheum 1996;39:1781-90.
28. Han Z, Boyle DL, Shi Y, Green DR, Firestein GS. Dominant-negative p53 mutations in rheumatoid arthritis. Arthritis Rheum 1999;42:1088-92.
29. Firestein GS, Echeverri F, Yeo M, Zvaifler NJ, Green DR. Somatic mutations in the p53 tumor suppressor gene in rheumatoid arthritis synovium. Proc Natl Acad Sci USA 1997;94:10895-900.
30. Reme T, Travaglio A, Gueydon E, Adla L, Jorgensen C, Sany J. Mutations of the p53 tumour suppressor gene in erosive rheumatoid synovial tissue. Clin Exp Immunol 1998;111:353-8.
31. Williams DG, Moyes SP, Mageed RA. Rheumatoid factor isotype switch and somatic mutation variants within rheumatoid arthritis synovium. Immunology 1999;98:123-36.
32. Goronzy JJ, Zettl A, Weyand CM. T cell receptor repertoire in rheumatoid arthritis. Int Rev Immunol 1998;17:339-63.
33. Hall FC, Thomson K, Procter J, McMichael AJ, Wordsworth BP. TCR beta spectratyping in RA: evidence of clonal expansions in peripheral blood lymphocytes. Ann Rheum Dis 1998;57:319-22.
34. Callahan LF, Pincus T. Mortality in the rheumatic diseases. Arthritis Care Res 1995;8:229-41.
35. Pincus T, Brooks RH, Callahan LF. Prediction of long-term mortality in patients with rheumatoid arthritis according to simple questionnaire and joint count measures. Ann Intern Med 1994;120:26-34.
36. Mitchell DM, Spitz PW, Young DY, Bloch DA, McShane DJ, Fries JF. Survival, prognosis, and causes of death in rheumatoid arthritis. Arthritis Rheum 1986;29:706-14.
37. Leigh JP, Fries JF. Mortality predictors among 263 patients with rheumatoid arthritis. J Rheumatol 1991;18:1307-12.
38. Pincus T, Callahan LF, Vaughn WK. Questionnaire, walking time and button test measures of functional capacity as predictive markers for mortality in rheumatoid arthritis. J Rheumatol 1987;14:240-51.
39. Cobb S, Anderson F, Bauer W. Length of life and cause of death in rheumatoid arthritis. N Engl J Med 1953;249:553-6.
40. Myllykangas-Luosujarvi RA, Aho K, Isomaki HA. Mortality in rheumatoid arthritis. Semin Arthritis Rheum 1995;25:193-202.
41. Myllykangas-Luosujarvi RA, Aho K, Kautiainen H, Isomaki H. Shortening of life span and causes of excess mortality in a population-based series of subjects with rheumatoid arthritis. Clin Exp Rheumatol 1995;13:149-53.

42. Mitchell DM, Spitz PW, Young DY, Bloch DA, McShane DJ, Fries JF. Survival, prognosis, and causes of death in rheumatoid arthritis. Arthritis Rheum 1986;29:706-14.
43. Vandenbroucke JP, Hazevoet HM, Cats A. Survival and cause of death in rheumatoid arthritis: a 25-year prospective followup. J Rheumatol 1984;11:158-61.
44. Uddin J, Kraus AS, Kelly HG. Survivorship and death in rheumatoid arthritis. Arthritis Rheum 1970;13:125-30.
45. Euler HH, Marmont AM, Bacigalupo A, et al. Early recurrence or persistence of autoimmune diseases after unmanipulated autologous stem cell transplantation. Blood 1996;88:3621-5.
46. Cooley HM, Snowden JA, Grigg AP, Wicks IP. Outcome of rheumatoid arthritis and psoriasis following autologous stem cell transplantation for hematologic malignancy. Arthritis Rheum 1997;40:1712-5.
47. Jondeau K, Job-Deslandre C, Bouscary D, Khanlou N, Menkes CJ, Dreyfus F. Remission of nonerosive polyarthritis associated with Sjogren's syndrome after autologous hematopoietic stem cell transplantation for lymphoma. J Rheumatol 1997;24:2466-8.
48. Joske DJ, Ma DT, Langlands DR, Owen ET. Autologous bone-marrow transplantation for rheumatoid arthritis [letter]. Lancet 1997;350:337.
49. Burt RK, Georganas C, Schroeder J, et al. Autologous hematopoietic stem cell transplantation in refractory rheumatoid arthritis: sustained response in two of four patients. Arthritis Rheum 1999;42:2281-5.
50. Snowden JA, Biggs JC, Milliken ST, Fuller A, Brooks PM. A phase I/II dose escalation study of intensified cyclophosphamide and autologous blood stem cell rescue in severe, active rheumatoid arthritis. Arthritis Rheum 1999;42:2286-92.
51. Durez P, Toungouz M, Schandene L, Lambermont M, Goldman M. Remission and immune reconstitution after T-cell-depleted stem-cell transplantation for rheumatoid arthritis [letter]. Lancet 1998;352:881.
52. Burt RK, Traynor AE, Pope R, et al. Treatment of autoimmune disease by intense immunosuppressive conditioning and autologous hematopoietic stem cell transplantation. Blood 1998;92:3505-14.
53. McColl G, Kohsaka H, Szer J, Wicks I. High-dose chemotherapy and syngeneic hemopoietic stem-cell transplantation for severe, seronegative rheumatoid arthritis. Ann Intern Med 1999;131:507-9.
54. Pavletic S, O'Dell J, Ursick M, et al. Autologous blood stem cell transplantation can overcome and modulate therapeutic resistance in severe rheumatoid arthritis [abstract]. Blood 1999;94:404b.
55. Arnett F, Edworthy S, Bloch D, et al. The American Rheumatism Association 1987 revised criteria for the classification of rheumatoid arthritis. Arthritis Rheum 1988;31:315-24.
56. Slavin S, Nagler A, Varadi G, Or R. Graft vs autoimmunity following allogeneic non-myeloablative blood stem cell transplantation in a patient with chronic myelogenous leukemia and severe systemic psoriasis and psoriatic polyarthritis. Exp Hematol 2000;28:853-7.
57. Weiden PL, Sullivan KM, Fluornoy N, et al. Antileukemic effect of chronic graft-versus-host disease: Contribution to improved survival after allogeneic marrow transplantation. N Engl J Med 1981;304:1529-33.
58. Horowitz MM, Gale RP, Sondel PM, et al. Graft-versus-leukemia reactions after bone marrow transplantation. Blood 1990;75:555-62.
59. Slavin S, Or R, Naparstek E, Ackerstein A, Weiss L. Cellular-mediated immunotherapy of leukemia in conjunction with autologous and allogeneic bone marrow transplantation in experimental animals and man [abstract]. Blood 1988;72 Suppl 1:407a.
60. Slavin S, Naparstek E, Nagler A, et al. Allogeneic cell therapy with donor peripheral blood cells and recombinant human interleukin-2 to treat leukemia relapse post allogeneic bone marrow transplantation. Blood 1996;87:2195-204.
61. Slavin S, Naparstek E, Nagler A, Ackerstein A, Kapelushnik Y, Or R. Allogeneic cell therapy for relapsed leukemia following bone marrow transplantation with donor peripheral blood lymphocytes. Exp Hematol 1995;23:1553-62.
62. Slavin S, Nagler A, Naparstek E, et al. Immunotherapy of leukemia in conjunction with non-myeloablative conditioning: engraftment of blood stem cells and eradication of host leukemia with nonmyeloablative conditioning based on Fludarabine and anti-thymocyte globulin [abstract]. Blood 1996;88:614a.
63. Giralt S, Estey E, Albitar M, et al. Engraftment of allogeneic hematopoietic progenitor cells with purine analog-containing chemotherapy: Harnessing graft-vs-leukemia without myeloablative therapy. Blood 1997:89;4531-6.
64. Slavin S, Nagler A, Naparstek E, et al. Nonmyeloablative stem cell transplantation and cell therapy as an alternative to conventional bone marrow transplantation with lethal cytoreduction for the treatment of malignant and non-malignant hematologic diseases. Blood 1998;91:756-63.
65. Pincus T, Callahan LF. Remodeling the pyramid or remodeling the paradigms concerning rheumatoid arthritis — lessons from Hodgkin's disease and coronary artery disease. J Rheumatol 1990;17:1582-5.
66. O'Dell JR. Anticytokine therapy — a new era in the treatment of rheumatoid arthritis? N Engl J Med 1999;340:310-2.
67. Weinblatt ME, Kremer JM, Bankhurst AD, et al. A trial of etanercept, a recombinant tumor necrosis factor receptor:Fc fusion protein, in patients with rheumatoid arthritis receiving methotrexate. N Engl J Med 1999;340:253-9.
68. Elliott MJ, Maini RN, Feldmann M, et al. Randomised double-blind comparison of chimeric monoclonal antibody to tumor necrosis factor versus placebo in rheumatoid arthritis. Lancet 1994;344:1105-10.
69. Aversa F, Tabilio A, Velardi A, et al. Treatment of high-risk acute leukemia with T-cell-depleted stem cells from related donors with one fully mismatched HLA haplotype. N Engl J Med 1998;339:1186-93.
70. Brodsky RA, Petri M, Smith BD, et al. Immunoablative high-dose cyclophosphamide without stem-cell rescue for refractory, severe autoimmune disease. Ann Intern Med 1998;129:1031-5.
71. van Bekkum DW, Kinwel-Bohre EP. Risk of donor lymphocyte infusions following allogeneic bone marrow transplantation. Exp Hematol 1997;25:478-80.