Haematopoietic stem cell transplantation in systemic sclerosis

Ulrich A Walker,1 Lesley Ann Saketkoo,2 Oliver Distler3

ABSTRACT
Three randomised controlled trials of haematopoietic stem cell transplantation (HSCT) in systemic sclerosis (SSc) demonstrated long-term survival benefits, induction of clinically meaningful, sustained improvement of forced vital capacity with improvements in skin thickening, vasculopathy and health-related quality of life, in contrast to a clinical decline in standard of care control groups. These benefits, however, must be weighed against the increased risk of transplant-related mortality. Further, with disease progression, severe extensive internal organ involvement and damage ensues, constituting an exclusion criterion for safety reasons, leaving a limited window whereby patients with SSc are eligible for HSCT. Although autologous HSCT offers the possibility of drug-free remission, relapse can occur, requiring re-initiation of disease modifying antirheumatic drugs. HSCT is also associated with secondary autoimmune diseases and gonadal failure. HSCT should be proposed for carefully selected patients with early rapidly progressive diffuse SSc whose clinical picture portends a poor prognosis for survival, but yet lacks advanced organ involvement.

INTRODUCTION
Systemic sclerosis (SSc) is a rare, clinically heterogeneous multisystem autoimmune disorder driven by inflammation, fibrosis and a microangiopathic vasculopathy. Internal organ involvement greatly impacts physical and psychological functioning, impairing one’s ability to work and participate in social activities. SSc disease progression is the leading cause of patient all-cause mortality, largely related to end-stage organ involvement. Pulmonary and cardiac complications are the leading drivers of SSc-specific mortality.

Current SSc treatment recommendations have, until recently, focused on the management of individual organ manifestations. High grade evidence from randomised placebo-controlled clinical trials (RCTs) for potential disease modifying agents currently exists only for methotrexate and cyclophosphamide. The clinical benefit of these drugs is, however, limited by moderate or short-term efficacy. Great hope anticipates several ongoing prospective phase II/III RCTs of targeted therapies in SSc, with results expected in 2018/2019.

Haematopoietic stem cell transplantation (HSCT) has been used in the treatment of autoimmune disorders refractory to conventional immunosuppression for over 2 decades. Since 2013, the yearly frequency of HSCT has steadily increased, with the European Society for Blood and Marrow Transplantation (EBMT) registering approximately 2300 HSCT procedures in 2016 for a variety of autoimmune diseases. Three RCTs have now demonstrated clinically meaningful improvement of organ involvement and survival in patients with SSc undergoing HSCT, further increasing awareness and interest in this complex procedure. These results have sparked the European League against Rheumatism (EULAR) to recommend that HSCT be considered for patients with rapidly progressive SSc who are at risk for organ failure. The aim of this manuscript is to examine the rationale, benefits and risks of HSCT in SSc, predicated on the high-level findings produced from RCTs.

Rationale
HSCT aims to non-specifically immunoablate aberrant self-reactive T-cells and B-cells
via high-dose immunosuppression, with subsequent reconstitution of a renewed and tolerant immune system by means of infusing a patient’s previously collected bone marrow using substances like granulocyte colony-stimulating factor (G-CSF) and cyclophosphamide which causes the release of proteases with cleavage of adhesion molecules, facilitating the release of haematopoietic stem cells into the peripheral blood; however, G-CSF alone has been shown to be efficacious in mobilisation. Harvesting subsequently these stem cells are collected from peripheral circulation and stored in liquid nitrogen. At this point, either simple apheresis or stem cell manipulation with selection for CD34+ can occur (or if the patient’s umbilical cord blood was banked it can be used to supplement quantity). Though selection for and reinfusion of high concentration CD34+ cells may result in extended periods of severe immunodeficiency, such manipulation may prevent reinfusion of autoreactive cells. Conditioning: A few weeks following stem cell harvest, the majority of resident autoreactive T-lymphocyte and B-lymphocyte subsets are eliminated using high doses of cyclophosphamide, antithymocyte globulin (ATG) with/without total body irradiation (TBI). Conditioning regimens without TBI are predominantly lymphoablative, profoundly depleting lymphocytes, while preserving cyclophosphamide-resistant myelogenous stem cells, whereas regimens employing TBI are also myeloablative. Transplantation: shortly after conditioning, stem cells are thawed and reinfused. Endogenous haematopoiesis could reconstitute even without transplantation; however, stem cell grafting shortens the period of pancytopenia, allowing adaptive immunity to be rebuilt through clonal expansion of the remaining immunocompetent cells, formation of new non-autoreactive cells (thymopoiesis) and graft-derived regulatory T-cells. Complete elimination of autoreactive T-cells is impossible, but through clonal expansion they become outnumbered by the newer tolerant clones, with relatively few patients with autoimmune diseases relapsing despite persistent post-transplant autoreactive clones.

Benefits of autologous HSCT
Multiple cohorts and a case control study suggested beneficial effects of autologous HSCT in patients with rapidly progressive SSc. The majority of patients treated with HSCT had the early rapidly progressive diffuse cutaneous form of SSc, without yet having progressed to severe internal organ involvement.
Table 1  Comparison of patient selection, treatment modality and outcomes among three randomised trials investigating HSCT in SSc.

|                  | ASSIST<sup>13</sup> | ASTIS<sup>14</sup> | SCOT<sup>15</sup> |
|------------------|---------------------|-------------------|------------------|
| Patient number   | 19                  | 156               | 75               |
| Inclusion criteria | <60 years of age | 18–65 years of age | 18–69 years of age |
| Diffuse SSc      | mRSS≥15              | mRSS≥15            | mRSS≥16          |
| Disease duration ≤4 years | Disease duration ≤4 years | Disease duration ≤4 years |
| Internal organ involvement | Internal organ involvement | Internal organ involvement |
| Exclusion criteria | Mean PAP>25 mm Hg or PAPsys>40 mm Hg | Mean PAP>50 mm Hg | Mean PAP>30 mm Hg |
| -                | LVEF<40%             | LVEF<45%           | LVEF<50%         |
| -                | Creatinine >177 umol/L | Creatinine clearance <40 mL/min | Creatinine clearance <40 mL/min |
| Mobilisation     | Cyclophosphamide 2 g/m², G-CSF | Cyclophosphamide 4 g/m², G-CSF | G-CSF only |
| Conditioning     | Cyclophosphamide (200 mg/kg), rabbit ATG | Cyclophosphamide (200 mg/kg), rabbit ATG | Cyclophosphamide (120 mg/kg), equine ATG |
| Total body irradiation | No | No | Yes (800 cGy, lung and kidney shielding) |
| Stem cell manipulation | None | CD34+ selection | CD34+ selection |
| Comparator arm   | Cyclophosphamide 6 monthly intravenous courses (1000 mg/m²) | Cyclophosphamide 12 monthly intravenous courses (750 mg/m²). | Cyclophosphamide 12 monthly intravenous courses (750 mg/m²). |
| Primary outcome measure | >25% decrease in mRSS, or >10% increase in FVC at 12 months | Survival without new onset heart, lung or kidney failure | Global Rank Composite Score at month 54 |
| Follow-up        | 2.6 years (mean) | 5.8 years (median) | Up to 4.5 years |
| 12-month treatment-related mortality in comparator arm | 0 (0%) | 0 (0%) | 0 (0%) |
| 12-month transplant-related mortality | 0 (0%) | 8 (10.1%) | 1 (3%) |

ASSIST, American Scleroderma Stem cell versus Immune Suppression Trial; ASTIS, Autologous Stem cell Transplantation International Scleroderma Trial; ATG, antithymocyte globulin; FVC, forced vital capacity; GAVE, gastric antral vascular ectasia; G-CSF, granulocyte-colony stimulating factor; LVEF, left ventricular ejection fraction; mRSS, modified Rodnan skin score; PAP, pulmonary arterial pressure; SCOT, The Scleroderma Cyclophosphamide Or Transplantation; SSc, systemic sclerosis.

half in the transplant group, but increased in the control group. Similarly, significant and clinically meaningful improvement of HRQoL occurred in the HSCT group with a change in the total 36-item Short Form Health Survey (SF-36) score from 39 to 56 during the 12 months after transplantation, but deteriorated significantly from 50 to 40 in the control arm. After a mean follow-up of 2.6 years, 15 of the 17 patients undergoing HSCT maintained persistent improvement in mRSS and FVC. There were no deaths in either arm of the ASSIST trial. The study had limitations: due to the small sample size, baseline data were not matched, for example, higher baseline mRSS was reported in the treatment group favouring spontaneous regression to the mean. It was also a one centre trial not necessarily reflecting real-world circumstances.

These limitations were addressed in the second trial, the Autologous Stem cell Transplantation International Scleroderma Trial (ASTIS) trial<sup>14</sup>. This first phase III trial in SSc randomised a total of 156 patients (mean age 44 years) to receive autologous HSCT or 12-monthly pulses...
of intravenous cyclophosphamide. In the transplant arm of ASTIS, stem cells were mobilised with a total of 4 g/m² of intravenous cyclophosphamide and G-CSF. After conditioning with 200 mg/kg of intravenous cyclophosphamide, administered with hyperhydration and rabbit ATG, CD34-selected stem cells were reinfused (Table 1). The majority of the patients in ASTIS (87%) had lung involvement; the mean mRSS at entry was 25, and 10% had severe skin involvement without internal organ involvement. The primary endpoint was event-free survival (EFS), defined as the time in days from randomisation until the occurrence of death or major organ failure. In the group undergoing HSCT, there were better event-free and overall survival rates. During a median follow-up of 5.8 years, 19 deaths and 3 irreversible organ failures occurred in the HSCT group, while in the control group 23 deaths and 8 irreversible organ failures were recorded. In the control arm, an additional seven patients died subsequent to irreversible organ failure. Secondary endpoints of ASTIS, defined as the change in mRSS in the first 2 years, Health Assessment Questionnaire (HAQ), EuroQoL or SF-36 scores were also significantly better in the HSCT group. No significant changes were seen for left ventricular ejection fraction or diffusing capacity of the lung for carbon monoxide (DLCO). However, a 6.3% increase in FVC and a modest but statistically significant decrease in creatinine clearance was seen in the HSCT group. In ASTIS, HSCT was associated with a relatively high treatment-related mortality in the first year after transplantation, higher than that in the other RCTs. In the first year following randomisation, there were 11 deaths (13.9%) in the HSCT group vs 7 (9.1%) in the cyclophosphamide group. Eight of the 11 deaths (10%) in the HSCT group were treatment-related, and no death was attributed to treatment in the cyclophosphamide control arm. In subsequent years, the high treatment-related mortality observed in the first year post-HSCT was outweighed by a significant long-term all-cause mortality benefit observed in the HSCT arm. Between 12 and 24 months following HSCT, SSc relapsed in 22.4% of patients, but significantly fewer patients in the group undergoing HSCT had lung involvement; the mean mRSS at entry was 25, and 10% had severe skin involvement without internal organ involvement. The primary endpoint was event-free survival (EFS), defined as the time in days from randomisation until the occurrence of death or major organ failure. In the group undergoing HSCT, there were better event-free and overall survival rates. During a median follow-up of 5.8 years, 19 deaths and 3 irreversible organ failures occurred in the HSCT group, while in the control group 23 deaths and 8 irreversible organ failures were recorded. In the control arm, an additional seven patients died subsequent to irreversible organ failure. Secondary endpoints of ASTIS, defined as the change in mRSS in the first 2 years, Health Assessment Questionnaire (HAQ), EuroQoL or SF-36 scores were also significantly better in the HSCT group. No significant changes were seen for left ventricular ejection fraction or diffusing capacity of the lung for carbon monoxide (DLCO). However, a 6.3% increase in FVC and a modest but statistically significant decrease in creatinine clearance was seen in the HSCT group. In ASTIS, HSCT was associated with a relatively high treatment-related mortality in the first year after transplantation, higher than that in the other RCTs. In the first year following randomisation, there were 11 deaths (13.9%) in the HSCT group vs 7 (9.1%) in the cyclophosphamide group. Eight of the 11 deaths (10%) in the HSCT group were treatment-related, and no death was attributed to treatment in the cyclophosphamide control arm. In subsequent years, the high treatment-related mortality observed in the first year post-HSCT was outweighed by a significant long-term all-cause mortality benefit observed in the HSCT arm. Between 12 and 24 months following HSCT, SSc relapsed in 22.4% of patients, but significantly fewer patients in the HSCT group as compared with the control group required immunosuppressive medication (22% vs 44%).

The Scleroderma Cyclophosphamide Or Transplantation (SCOT) trial is the third and most recent RCT examining the effects of HSCT in SSc. Like the ASSIST and ASTIS trials, SCOT compared autologous HSCT with monthly cyclophosphamide pulses; the conditioning regimen, however, differed in that it added TBI. The rationale for irradiation is based in part on the observed higher effectiveness of the transplantation procedure with addition of irradiation in animal models. The SCOT trial also differed in that it did not employ cyclophosphamide for mobilisation and used less cyclophosphamide during conditioning compared with the previous trials (Table 1). Seventy-five patients with SSc (mean age 46 years) were randomised, with 97% having pulmonary involvement, characterised by a mean baseline FVC and DLCO of 74% and 53% predicted, respectively and a baseline mRSS of 30. Fifty-nine patients had received disease modifying antirheumatic medication prior to randomisation. The primary endpoint of the SCOT trial was a Global Rank Composite Score (GRCS), a tool that simultaneously accounts for multiple disease manifestations based on the following hierarchy of outcomes: death, EFS, FVC, HAQ-Disability Index (HAQ-DI) and mRSS. The GRCS does not measure disease activity or severity but compares patients by means of hierarchical ordered outcomes. The GRCS score at 54 months showed superiority of HSCT over cyclophosphamide. Superiority of HSCT was also demonstrated for all-cause mortality; of the 36 patients randomised to receive HSCT, 3 patients had died by month 54 and 1 death was considered treatment-related, while of the 39 patients in the cyclophosphamide arm, 11 died and no death was considered treatment-related. HSCT was also superior to cyclophosphamide in terms of mRSS evolution, change in FVC and HRQoL measures (HAQ-DI and SF-36). No pulmonary arterial hypertension or congestive heart failure was observed in the HSCT arm 54 months after HSCT, significantly less than the 15% and 12% of cases, respectively, observed in the cyclophosphamide arm. In the HSCT arm, 9% of participants required disease modifying antirheumatic drugs (DMARDs) post-transplantation, significantly less than the 44% of patients in the cyclophosphamide arm. There were similar rates of infections (of any grade) in both groups; however, the rate of grade 3 infections or higher per person-year was nominally higher in the transplantation group than in the cyclophosphamide group (0.21 vs 0.13 p=0.09). One patient in the HSCT arm and five patients in the cyclophosphamide arm experienced renal crisis during the 54 months follow-up period. The 54 months post-treatment EFS was 72.2% in HSCT group and 48.7% in control group. The overall survival of autologous HSCT-treated patients was 91% and 77% in the control patients.

In summary, all three RCTs reported significant improvement in organ specific manifestations, such as skin and pulmonary function, and HRQoL. Despite an elevated risk of treatment-related mortality early after the intervention, there were significant long-term survival advantages following HSCT in both of the two RCTs investigating long term survival.

**Risks of HSCT**

The benefits of any treatment procedure need to be weighed against its risks. The mortality associated with HSCT appears to be significant in the RCTs, especially in the ASTIS trial. There was no centre effect of mortality in ASTIS. The small sample size of the ASSIST trial and the ASTIS trial. There was no centre effect of mortality in ASTIS. The small sample size of the ASSIST trial and the selection of patients may explain the survival of all patients in this trial; similarly, the smaller sample size in SCOT than in ASTIS may contribute to differences in mortality. Further, in ASTIS, the solitary variable shown in a posthoc analysis to impact differences in EFS and overall survival was smoking status, as in former/current
smokers HSCT lacked the benefit demonstrated in the never-smoking subjects. Similarly, SCOT data also suggest that in former and current smokers, transplantation had no advantage over cyclophosphamide.15

The EBMT analysed mortality after autologous HSCT for severe autoimmune disease from 1996 to December 2007 and reported a 5% mortality by day 100 following transplantation.22 In 175 patients with SSc transplanted from 1996 until 2007, the mortality was slightly higher (6%) than in other autoimmune diseases, possibly due to the severity of SSc and the presence of major organ dysfunction in transplanted patients with SSc.23 25 The causes of death included SSc recurrence (23 patients), transplant-related mortality (12 patients), cardiotoxicity (1 patient), haemorrhage and secondary malignancies (2 patients each) as well as infections (4 patients).22 Careful patient selection is crucial in order to reduce treatment-related mortality. SSc has unique and complex cardiac manifestations and cyclophosphamide is associated with cardiotoxicity.26 A comprehensive pretransplant cardiac assessment is therefore recommended even in patients without cardiac symptoms and typically includes transthoracic echocardiography, cardiac MRI with gadolinium contrast, a Holter ECG and right heart catheterisation (RHC).34 A 6 min walk test is also recommended.26 Some authors and the EBMT suggest an intravascular fluid challenge during RHC (10 mL saline/kg body weight, given intravenously over 10 min).26 34 A mean pulmonary arterial pressure (PAP)>30 mm Hg or a systolic PAP>40 mm Hg after fluid challenge could detect patients with subclinical heart involvement related to cardiomyopathic restriction seen in SSc, who might fluid-sensitive and develop complications during hypervolemic interventions often necessary during the conditioning regimen or should systemic infection develop.26 34

HSCT can induce gonadal failure in both sexes. In men, azoospermia occurs in the majority of patients but in most cases testosterone levels remain within the normal limits.35 In premenopausal women, the conditioning regimen results in transitory or permanent amenorrhoea with concomitant infertility and menopausal symptoms. Before mobilisation and HSCT, consideration should therefore be given to infertility in both sexes (semen, oocyte or embryo cryopreservation as appropriate). Hormone replacement therapy should be started with gonadal failure.36 TBI used as part of the conditioning regimen plays a central role in post-transplantation infertility,38 but in regimen without TBI a substantial percentage of women have been described to remain fertile, give birth to healthy babies and without increased occurrence of miscarriages.37 The decline of post-HSCT rates of sexual activity and diminished interest/libido and adequate function is possibly related to preparation regimen using alkylating agents and TBI which impacts on the function of the hypothalamic-pituitary-gonadal axis function. Sexual dysfunction may persist long term in both sexes despite some recovery after the 6 months post-transplant nadir.38

Regarding renal protection and outcome, life-long post-transplant therapy with angiotensin-converting enzyme inhibitors or angiotensin receptor blockers is recommended to prevent renal crisis.26 36 However, it is unclear whether prophylactic ACE inhibition leads to more severe outcomes of renal crisis.40

In addition to better understanding frequency of and predisposition to relapse/graft failure, long-term follow-up of transplanted patients with SSc will be important to better characterise and quantify known late sequelae of HSCT, such as secondary autoimmune diseases and secondary malignancies.41 Five years after autologous HSCT for autoimmune diseases, the cumulative incidence of secondary autoimmune diseases was as high as 9.8%.42 Intriguingly, the ‘new’ autoimmunity developing after transplantation appeared to be mainly antibody-associated and organ-specific.43 44 Lupus erythematosus as primary autoimmune disease and ATG use were risk factors for the occurrence of secondary autoimmune diseases, whereas the presumed beneficial role of CD34+ graft selection is controversial.32 44 The attenuation of immunological memory in the B cell compartment after autologous HSCT also implies that patients must be reimmunised.45

Because of the high risk of treatment related side effects and of early treatment related mortality, the new EULAR treatment recommendations advice for careful selection of patients with SSc for HSCT. It is also highlighted that the experience of the medical team are of key importance.4

Research agenda

Head-to-head studies have not been carried out to determine the optimal stem cell collection and conditioning procedure in the HSCT regimen. It is unclear, if the ex vivo CD34+ selection of stem cells truly confers a benefit over non-selected cells.16 The potential risk of reinfusing autoreactive T-cells with non-selected grafts may be outweighed by reduced numbers of transplanted cells and increased immunosuppression in patients receiving CD34+ grafts.46 Reducing cyclophosphamide doses may on the one hand decrease toxicity in terms of infections, but on the other hand decrease the efficacy of HSCT. Cyclophosphamide may also induce haemorrhagic cystitis. Patients should receive urometixan.36 It is however unclear if the hyperhydration recommended for the prophylaxis of haemorrhagic cystitis may contribute to transplant related mortality in patients with decreased cardiac compliance.

Animal studies indicate that a conditioning regimen that adds irradiation to cyclophosphamide may provide a better control of autoimmune diseases.33 As the SCOT trial has inclusion criteria and a control group similar to those in the ASTIS trial, a comparative analysis with regard to the use and not-use of irradiation during conditioning may be possible. In the SCOT trial, only 9% of transplant recipients had initiated DMARDs by 24 months, as compared with 22% in the ASTIS trial.
suggesting better SSc control with myeloablative HSCT. Adding TBI, however, also implies an excess risk of cancers over a lifetime.

With regard to the known cardiotoxicity of high-dose cyclophosphamide used during the conditioning step, it is unclear which patients are at particular risk and which conditioning is best. In order to diminish cyclophosphamide requirements, conditioning regimens have been used that employ thiotepa, a non-cardiotoxic alkylating agent, but comparative studies are lacking.

It is also currently vague, if other agents used during the HSCT procedure put specific patients with SSc at risk. G-CSF could induce disease flares if administered alone, although the combination with cyclophosphamide in the mobilisation step effect may prevent flares and improve stem cell yields. ATG is associated with a risk of allergic reactions and its profound immunosuppression increases the risk of acquired and reactivated infections.

To address the issue of disease relapse after successful transplantation, the Scleroderma Treatment with Autologous Transplant (STAT) trial (ClinicalTrials.gov identifier: NCT01413100) is currently recruiting. The conditioning regimen of STAT uses no CD34+ selection after mobilisation of stem cells and apheresis and the conditioning regimen consists of cyclophosphamide and ATG, but without TBI. In the STAT trial, open label mycophenolate mofetil is added approximately 2–3 months post-transplant after the stem cells have ‘engrafted’ and mycophenolate mofetil is maintained for 2 years. The primary outcome of this single group trial is defined as EFS 5 years after transplantation.

Last, developing more specific and validated criteria for patient selection is ongoing. It is currently unclear which cardiac screening is optimal and which cardiac parameters are predictive of an increased transplant related cardiac mortality. Hopefully, analysis of past and future studies will help clarify risk stratifications incorporating factors of age, functional performance status, rate of disease progression, parameters of SSc damage, smoking and cardiac compliance.

**SUMMARY AND CONCLUSION**

Autologous HSCT is the first therapeutic intervention with proven survival advantages in a subgroup of patients with SSc. In addition to improving survival, three RCTs demonstrated significant improvement of organ function, vasculopathy, skin involvement and HRQoL. HSCT has induced clinically meaningful and sustained improvements of lung function despite decline in control groups. Autologous HSCT is currently the only disease modifying strategy with high-level evidence for improving SSc survival, prevention of organ worsening and improvement of pulmonary function. Although autologous HSCT offers the possibility of drug-free remission, patients may experience a relapse requiring disease modifying antirheumatic drugs. HSCT is also associated with secondary autoimmune diseases and gonadal failure. The benefits of HSCT must be balanced against the increased risk of transplant-related mortality in the first year. A patient with early SSc, internal organ involvement and concomitant poor prognostic factors has a limited window within which they are eligible for HSCT. As the disease progresses, severe internal organ damage ensues and constitutes an exclusion criterion for the procedure. The EULAR treatment recommendations indicate HSCT for carefully selected patients with rapidly progressive SSc at risk of organ failure. HSCT should be performed in centres experienced in HSCT. More specifically, we recommend considering HSCT in patients with early, progressive diffuse SSc with strongly reduced survival as predicted by validated tools, yet without severe organ involvement.

**REFERENCES**

1. Jaeger VK, Distler O, Maurer B, et al. Functional disability and its predictors in systemic sclerosis: a study from the DeSScipher project within the EUSTAR group. *Rheumatology* 2018;57:441–50.
2. Tyndall AJ, Bannett B, Vonk M, et al. Causes and risk factors for death in systemic sclerosis: a study from the EULAR Scleroderma Trials and Research (EUSTAR) database. *Ann Rheum Dis* 2010;69:1809–15.
3. Elhai M, Meune C, Boubaya M, et al. Mapping and predicting mortality from systemic sclerosis. *Ann Rheum Dis* 2017;76:1897–905.
4. Kowal-Bielecka O, Fransen J, Avouac J, et al. Update of EULAR recommendations for the treatment of systemic sclerosis. *Ann Rheum Dis* 2017;76:1327–39.
5. Khanna D, Furst D, Clements PJ, et al. Oral cyclophosphamide for active sclerodema lung disease: a decision analysis. *Med Decis Making* 2008;28:926–37.
6. van den Hoogen FH, Boerboom AS, Swaak AJ, et al. Comparison of methotrexate with placebo in the treatment of systemic sclerosis: a 24 week randomized double-blind trial, followed by a 24 week observational trial. *Br J Rheumatol* 1996;35:364–72.
7. Pope JE, Bellamy N, Seibold JR, et al. A randomized, controlled trial of methotrexate versus placebo in early diffuse scleroderma. *Arthritis Rheum* 2001;44:1351–8.
8. Nannini C, West CP, Erwin PJ, et al. Effects of cyclophosphamide on pulmonary function in patients with scleroderma and interstitial lung disease: a systematic review and meta-analysis of randomized controlled trials and observational prospective cohort studies. *Arthritis Res Ther* 2008;10:R124.
9. Khanna D, Denton CP, Jahreis A, et al. Safety and efficacy of subcutaneous tocilizumab in adults with systemic sclerosis (faSScinate): a phase 2, randomised, controlled trial. *Lancet* 2016;387:2630–40.
10. Khanna D, Distler JHW, Sandner P. Emerging treatment strategies for systemic sclerosis. *J Scleroderma Relat Disord* 2016;1:186–93.
11. Binks M, Passweg JR, Forst D, et al. Phase II/II trial of autologous stem cell transplantation in systemic sclerosis: procedure related mortality and impact on skin disease. Ann Rheum Dis 2001;60:577–84.

12. Snowden J. Autoimmune diseases working party. European society of blood and marrow transplantation - annual report. 2016.

13. Burt RK, Shah SJ, Dill K, et al. Autologous non-myeloablative haemopoietic stem-cell transplantation compared with pulse cyclophosphamide once per month for systemic sclerosis (ASSIST): an open-label, randomised phase 2 trial. Lancet 2011;378:498–506.

14. van Laar JM, Farge D, Sont JK, et al. Autologous haematopoietic stem cell transplantation vs intravenous pulse cyclophosphamide in diffuse cutaneous systemic sclerosis: a randomized clinical trial. JAMA 2014;311:2490–8.

15. Sullivan KM, Golantunz EA, Keys-Elstein L, et al. Myeloablative Autologous Stem-Cell Transplantation for Severe Scleroderma. N Engl J Med 2018;378:35–47.

16. Swart JF, Delemarre EM, van Wijk F, et al. Haemopoietic stem cell transplantation for autoimmune diseases. Nat Rev Rheumatol 2017;13:244–56.

17. Nash RA, McSweeney PA, Nelson JL, et al. Allogeneic marrow transplantation in patients with severe systemic sclerosis: resolution of dermal fibrosis. Arthritis Rheum 2006;54:1982–6.

18. Loh Y, Oyama Y, Statkute L, et al. Autologous hematopoietic stem cell transplantation for severe systemic sclerosis: graft-versus-autoimmunity without graft-versus-host disease? Bone Marrow Transplant 2007;39:435–7.

19. Sykes M, Nikolic B. Treatment of severe autoimmune disease by stem-cell transplantation. Nature 2005;435:620–7.

20. Nash RA, McSweeney PA, Crofford LJ, et al. High-dose immunosuppressive therapy and autologous hematopoietic cell transplantation for severe systemic sclerosis: long-term follow-up of the US multicenter pilot study. Blood 2007;110:1386–96.

21. Henricsson J, Mogel W, et al. Autologous stem cell transplantation for systemic sclerosis - a single-center longitudinal experience in 26 patients with severe organ manifestations. J Rheumatol 2012;39:269–75.

22. Farge D, Labopin M, Tyndall A, et al. Autologous hematopoietic stem cell transplantation for autoimmune diseases: an observational study on 12 years’ experience from the European group for blood and marrow transplantation working party on autoimmune diseases. Haematologica 2010;95:284–92.

23. Vonk MC, Marjanovic Z, van den Hoogen FH, et al. Long-term follow-up results after autologous haematopoietic stem cell transplantation for severe systemic sclerosis. Ann Rheum Dis 2008;67:98–104.

24. Oyama Y, Barr WG, Statkute L, et al. Autologous non-myeloablative hematopoietic stem cell transplantation in patients with systemic sclerosis. Bone Marrow Transplant 2007;40:549–55.

25. Del Papa N, Onida F, Zaccara E, et al. Autologous hematopoietic stem cell transplantation has better outcomes than conventional therapies in patients with rapidly progressive systemic sclerosis. Bone Marrow Transplant 2017;52:53–8.

26. Burt RK, Oliveira MC, Shah SJ, et al. Cardiac involvement and treatment-related mortality after non-myeloablative haemopoietic stem-cell transplantation with unselected autologous peripheral blood for patients with systemic sclerosis: a retrospective analysis. Lancet 2013;381:1116–24.

27. McSweeney PA, Nash RA, Sullivan KM, et al. High-dose immunosuppressive therapy for severe systemic sclerosis: initial outcomes. Blood 2002;100:1602–10.

28. Launay D, Marjanovic Z, de Bazelaire C, et al. Autologous hematopoietic stem cell transplantation in systemic sclerosis: quantitative high resolution computed tomography of the chest scoring. J Rheumatol 2009;36:1460–3.

29. Mininati I, Guiducci S, Conforti ML, et al. Autologous stem cell transplantation improves microcirculation in systemic sclerosis. Ann Rheum Dis 2009;68:94–8.

30. Aschwanden M, Daikeler T, Jaeger KA, et al. Rapid improvement of nailfold capillaroscopy after intense immunosuppression for systemic sclerosis and mixed connective tissue disease. Ann Rheum Dis 2008;67:1057–9.

31. Flemling JN, Nash RA, McLeod DO, et al. Capillary regeneration in scleroderma: stem cell therapy reverses phenotype? PLoS One 2010;5:e9412.

32. Farge D, Passweg J, van Laar JM, et al. Autologous stem cell transplantation in the treatment of systemic sclerosis: report from the EBMT/EULAR Registry. Ann Rheum Dis 2004;63:974–81.

33. van Bekkum DW. Effectiveness and risks of total body irradiation for conditioning in the treatment of autoimmune disease with autologous bone marrow transplantation. Rheumatology 1999;38:757–61.

34. Farge D, Burt RK, Oliveira MC, et al. Cardiopulmonary assessment of patients with systemic sclerosis for hematopoietic stem cell transplantation: recommendations from the European society for blood and marrow transplantation autoimmune diseases working party and collaborating par society for blood and marrow transplantation autoimmune diseases working party and collaborating partners. Bone Marrow Transplant 2017;52:1495–503.

35. Rovó A, Tichelli A, Passweg JR, et al. Spermatogenesis in long-term survivors after autologous hematopoietic stem cell transplantation is associated with age, time interval since transplantation, and apparently absence of chronic GvHD. Blood 2006;108:1100–5.

36. Snowden JA, Saccardi R, Allez M, et al. Haematopoietic SCT in severe autoimmune diseases: updated guidelines of the European group for blood and marrow transplantation. Bone Marrow Transplant 2012;47:770–90.

37. Massenkeil G, Alexander T, Rosen O, et al. Long-term follow-up of fertility and pregnancy in autoimmune diseases after autologous haematopoietic stem cell transplantation. Rheumatol Int 2016;36:1563–8.

38. Syrjavu K, Kurland BF, Abrams JR, et al. Sexual function changes during the 5 years after high-dose treatment and hematopoietic cell transplantation for malignancy, with case-matched controls at 5 years. Blood 2008;111:989–96.

39. van Laar JM, Naraghi K, Tyndall A. Haematopoietic stem cell transplantation for autoimmune diseases after haematopoietic stem cell transplantation. Br J Haematol 2015;160:219–33.

40. Hudson M, Baron M, Taitbout S, et al. Exposure to ACE inhibitors prior to the onset of scleroderma renal crisis-results from the International scleroderma renal crisis survey. Semin Arthritis Rheum 2014;43:666–72.

41. Curtis RE, Rowlands PA, Deeg HJ, et al. Solid cancers after bone marrow transplantation. N Engl J Med 1999;336:897–904.

42. Daikeler T, Labopin M, Di Gioia M, et al. Secondary autoimmune diseases occurring after HSCT for an autoimmune disease: a retrospective study of the EBMT autoimmune disease working party, and collaborating par society for blood and marrow transplantation autoimmune diseases working party. Blood 2011;118:1693–8.

43. Holbro A, Abinun M, Daikeler T. Management of autoimmune diseases after haematopoietic stem cell transplantation. Br J Haematol 2012;157:281–90.

44. Loh Y, Oyama Y, Statkute L, et al. Development of a secondary autoimmune disorder after hematopoietic stem cell transplantation for autoimmune diseases: role of conditioning regimen used. Blood 2007;109:2643–2548.

45. Singhal S, Mehta J. Reimmunization after blood or marrow stem cell transplantation. Bone Marrow Transplant 1999:23:637–46.

46. Oliveira MC, Labopin M, Henes J, et al. Does ex vivo CD34+ positive selection influence outcome after autologous hematopoietic stem cell transplantation in systemic sclerosis patients? Bone Marrow Transplant 2016;51:501–5.

47. Hennes JC, Koeter I, Horger M, et al. Autologous stem cell transplantation with thiotepa-based conditioning in patients with systemic sclerosis and cardiac manifestations. Rheumatology 2014;53:919–22.

48. Fransen J, Pope-Diaconou D, Hesselstrand R, et al. Clinical prediction of 5-year survival in systemic sclerosis: validation of a simple prognostic model in EUSTAR centres. Ann Rheum Dis 2011;70:1788–92.