Autologous haematopoietic stem cell transplantation for Crohn's disease: a retrospective survey of long-term outcomes from the European Society for Blood and Marrow Transplantation

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Short title

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Abbreviations

AHSCT Autologous haematopoietic stem cell transplant

ASTIC Autologous stem cell transplant in Crohn's disease (clinical trial)

CD Crohn's disease

CDAI Crohn's disease activity index

CR Complete remission

ECCO European Crohn's and Colitis Organization

EBMT European Society for Blood & Marrow Transplantation

HR Hazard ratio

JACIE Joint Accreditation Committee-ISCT & EBMT

MTHS months

TNF tumour necrosis factor

YRS years

ABSTRACT

Background/Aims: Autologous haematopoietic stem cell transplantation (AHSCT) is a therapeutic option for patients with severe, treatment-refractory Crohn's disease (CD). The evidence base for AHSCT for CD is limited, with one randomised trial (ASTIC) suggesting benefit. The aim of this study was to evaluate safety and efficacy outcomes for patients undergoing AHSCT for CD in Europe outside the ASTIC trial.

Methods: We identified 99 patients in the European Society for Blood and Marrow Transplantation (EBMT) registry who were eligible for inclusion. Transplant and clinical outcomes were obtained for 82 pts from 19 centres in 7 countries.

Results: Median patient age was 30 years (range 20-65). Patients had failed or been intolerant to a median of 6 previous lines of drug therapy, including anti-TNF therapy in all cases. 61/82 (74%) had had surgery. Following AHSCT, 53/78 (68%) experienced complete remission or significant improvement in symptoms at a median follow-up of 41 months (range 6-174). 22/82 (27%) required no medical therapy at any point post-AHSCT. In patients who had re-started medical therapy at last follow-up, 57% (24/42) achieved remission or significant symptomatic improvement with therapies to which they had previously lost response or been non-responsive. Treatment-free survival at one year was 54%. On multivariate analysis, the presence of perianal disease was associated with adverse treatment-free survival (hazard ratio 2.34, 95% CI 1.14-4.83, p=0.02). One patient died due to infectious complications (CMV disease) at day +56. There was one death from infection at 8 years post-transplant.

Conclusions: In this multicentre retrospective analysis of European centres, AHSCT was relatively safe and appeared to be effective in controlling otherwise treatment-resistant

Crohn's disease. Further prospective randomised controlled trials against standard of care are warranted.

Key words: Autoimmune disease, Autologous haematopoietic stem cell transplant, Crohn's disease

INTRODUCTION

Crohn's disease (CD) is an immunologically mediated chronic disease characterised by episodic intestinal inflammation and dysregulation of the mucosa-associated immune system.(1) Anti-inflammatory and immunosuppressive agents are the mainstay of therapy but up to 25% of patients remain refractory to optimal medical therapy and a further 50% experience loss of response.(2, 3) Treatment-refractory CD is associated with adverse quality of life, recurrent hospitalization and increased mortality.(4, 5)

Autologous haematopoietic stem cell transplantation (AHSCT) is a potential therapeutic option for treatment-refractory CD.(6) AHSCT may lead to remission in CD by chemotherapy-mediated ablation of inflammatory cells followed by marrow reconstitution and restoration of immune tolerance.(7) Whilst the mechanisms underlying this process are incompletely defined, thymic re-activation, broadening of the total T, B, NK cell and plasma cell repertoire and resetting of regulatory T cell function have been suggested to play a role.(8)

Clinical experience of AHSCT for CD is limited, with several small series suggesting clinical benefits.(9-18) In a Phase 1/2 study of 24 patients with severe treatment-refractory CD, AHSCT resulted in clinical relapse-free survival of 91% at 1 year and 19% at 5 years, with a rapid and sustained improvement in Crohn's disease activity index (CDAI) post AHSCT.(10) Only one randomised trial of AHSCT for CD (ASTIC) has been reported to date.(19) This study enrolled patients with active CD not amenable to surgery and unresponsive to treatment with ≥3 immunosuppressive/biologic agents to AHSCT (n=23) or control (mobilization and AHSCT deferred for one year, n=22). One patient died of sepsis and hepatic veno-occlusive disease, and the trial failed to meet its primary endpoint of clinical and endoscopic 'cure' at 1 year, a composite of freedom from disease on imaging and endoscopy, CDAI < 150 and no active treatment for 3 months. This has been criticised for

being overly stringent, and patients demonstrated sustained improvement on pre-specified secondary endpoints.(19, 20)

EBMT guidelines, published in 2012, have included recommendations for AHSCT in CD: i) active and unresponsive disease despite multiple lines of therapy ii) extensive disease where surgical resection would expose the patient to small bowel syndrome risk, and iii) refractory colonic disease where a stoma is not acceptable to the patient.(6) As the number of patients undergoing AHSCT in any centre to date are limited, multi-centre studies are required. EBMT maintains a registry of all patients undergoing AHSCT for any indication and provides a means to identify the total European cohort. We therefore designed this retrospective study to evaluate the clinical use and outcomes of all AHSCT in CD performed in EBMT transplant centres outside the ASTIC trial.

METHODS

EBMT Registry

EBMT is a not-for-profit medical and scientific organization that represents over 500 HSCT centres from over 50 countries. The EBMT registry now contains details on over 500,000 allogeneic and autologous transplants performed since 1986. All patients included in the registry give written consent before transplant for the collection and analysis of anonymised data. The data are maintained in the central EBMT registry in line with legal and regulatory requirements for data protection, confidentiality and accuracy. EBMT implements regular quality assurance measures including ensuring centre accreditation, regular cross-checks with national registries, annual surveys and regular audit processes. This study was performed in line with EBMT guidelines and approved by the Autoimmune Diseases Working Party (ADWP).

Patient population:

Patients who underwent AHSCT for Crohn's disease were identified from the EBMT registry. All adult patients (aged ≥18 at time of AHSCT) undergoing AHSCT for a primary diagnosis of CD from 1997 to 2015 were eligible for inclusion. Patients who had participated in the ASTIC trial were excluded. From a total of 99 patients across 27 centres, data were obtained for 82 patients transplanted in 19 centres in 8 countries from 1996 to 2015 (listed in supplementary information). Data were unavailable for 17 patients due to lack of response to repeated requests.

Study endpoints:

Transplant and clinical outcomes for each patient were obtained directly from the EBMT registry supplemented by a standardised questionnaire completed by the treating clinicians in each centre. The primary study endpoint was clinical disease response (defined below) assessed by the patient's gastroenterologist one year following AHSCT, as compared to pre-mobilisation clinical status. Secondary endpoints included overall survival (OS), transplant-

related mortality (TRM), treatment-free survival and clinical disease response to mobilisation, at 100 days and at last clinical assessment. Variables considered for descriptive analyses were medical/surgical therapy pre- and post-AHSCT, disease extent and behaviour pre- and post-AHSCT, neutrophil and platelet engraftment dates. Data on complications post-AHSCT were recorded, including infectious complications requiring hospitalisation (bacterial, viral or fungal) up until 12 months post-AHSCT, and incidence of malignancy and secondary autoimmune disease post-AHSCT.

Definitions:

Clinical disease response was categorised as:

Remission: No abdominal pain and normal stool frequency

Improved: Improvement in abdominal pain and/or stool frequency.

Stable/no change: No appreciable improvement in abdominal pain and/or stool frequency.

Worse: Deterioration in abdominal pain and/or stool frequency,

The introduction, reduction or withdrawal of steroids, immunomodulators, or biological therapy and need for further surgical therapy were also recorded.

Disease behaviour was assessed as stricturing, penetrating, both or neither pre- and post-AHSCT (Appendix Table 2). Neutrophil engraftment was defined as time from day of transplant until day 1 of 3 consecutive days with an absolute neutrophil count ≥0.5 x 10^9/L, whereas platelet engraftment was defined as time from day of transplant until day 1 of 3 consecutive days with a platelet count ≥20 x 10^9/L. Treatment related mortality (TRM) was defined as any death after AHSCT within the first 100 days post-AHSCT. Treatment-free survival was defined as survival from transplantation without major surgery or medical therapy.

Statistics:

Qualitative variables were described as percentage, continuous variables using median and range. Overall survival and treatment-free survival were calculated according to the method of Kaplan and Meier. Variables considered in univariate and multivariate analyses of disease response and treatment-free survival were recipient age at AHSCT (>/≤median), time from diagnosis to AHSCT (>/≤median), patient sex (male vs female), disease classification (limited vs extensive without perianal disease vs extensive with perianal disease), disease behaviour (non-stricturing/non-penetrating vs stricturing vs penetrating), and pre-transplant smoking status. For treatment-free survival, a Cox proportional hazards model was used to evaluate the independent effect of co-variates on outcome. A p value of less than 0.05 was considered significant. Statistical analyses were performed with SPSS 24 (SPSS Inc./IBM, Armonk, NY, USA) and R 3.4.0 (R Development Core Team, Vienna, Austria) software packages.

RESULTS

Patient & disease characteristics

Patient and disease characteristics are summarised in Table 1. Median patient age was 30 years (range 20-65) and 52/82 (63%) were female. Median age at first diagnosis of CD was 17 years (range 2-53). Details of previous therapies are outlined in Table 2. Patients were heavily pre-treated, having failed or been intolerant to a median of 6 previous lines of therapy (range 3-10). 44/82 (54%) had received experimental therapy prior to AHSCT. This included participation in clinical trials of experimental immunosuppressants, faecal transplant, leukocytapheresis or mesenchymal stem cell therapy. Previous surgical treatment was common, with 61/82 (74%) of patients having undergone at least one operation. The median time from first diagnosis of CD to AHSCT was 12 years (range 1-26). Median length of follow-up following AHSCT was 41 months (range 6-174).

AHSCT characteristics & haematological outcomes

AHSCT details are summarised in Table 3. All patients underwent mobilisation with cyclophosphamide and peripheral blood stem cells were re-infused a median of 2 months (range 1-16) later. Patients received conditioning with cyclophosphamide 200mg/kg and 69/82 (86%) underwent in vivo T cell depletion with anti-thymocyte globulin (ATG). The median dose of ATG was 7.5mg/kg (range 2.0-10.0). The median CD34+ dose infused was 5.4 (range 2.4-40.6) x 10^6/kg. CD34+ selection of the autologous graft was performed in 11% of patients, and the remained were unmanipulated. All patients engrafted successfully. Neutrophil and platelet engraftment both occurred at a median of day 10 (range 6-22 and 6-44 respectively). 62% received post-transplant G-CSF.

CD outcomes

One year follow-up data were available for 76 patients (93%), as 1 patient died at 56 days

and data unavailable for 4 patients. 33/76 (43%) were in CR, 15/76 (20%) were reported as improved, 13/76 (17%) were unchanged and 15/76 (20%) had worsened.

At 100 days post AHSCT, data was available for 80 patients. 51/80 patients (64%) were in clinical remission (CR). A further 22/80 (28%) reported improvement. For 4/80 (5%) there was no change in disease and in 3/80 (4%) the disease worsened compared to baseline. At last follow-up, data were available for 78 patients. 34/78 (44%) were in CR, 19/78 (24%) were improved, 13/78 (17%) were unchanged and 12/78 (15%) had worsened. (Figure 1)

Predictors of achieving clinical disease remission or disease response (either remission or improvement) at one year were evaluated. There was no statistically significant impact of age at diagnosis, age at AHSCT, pre-transplant smoking status, time from diagnosis to AHSCT, patient sex, previous surgery, disease classification, and disease extent on the likelihood of achieving remission or disease response at one year.

Treatment-free survival was 54.6% (95% CI 43.8 - 65.5%) at one year, and 27% (95% CI 17-38%) and 22% (95% CI 11-33%) at three and five years respectively. There were no significant predictors of treatment-free survival identified on univariate analysis. On multivariate analysis extensive disease with perianal disease was found to be an independent predictor for adverse treatment-free survival with a hazard ratio of 2.34 (95% CI 1.14-4.83, p-value 0.02) (See **Table 4** for results of multivariate analysis).

Mortality & Complications

One patient died at day +56 post-AHSCT due to CMV infection, sepsis and multiorgan failure, i.e. a transplant-related mortality of 1.2%. Another patient died at 7.99 years post AHSCT from sepsis and multi-organ failure.

In the year post-AHSCT, 22/82 (27%) developed an infection requiring treatment post

AHSCT (9/82 (%) bacterial, 11/82 (12%) viral). EBV and CMV reactivation occurred in 5/82 (6%) and 3/82 (4%) respectively. There were no cases of fungal infection.

During follow-up post-AHSCT, a secondary autoimmune disease was reported in 9/82 (13%). These included thyroid disease (5/82; 6%), rheumatoid arthritis (2/82; 2%) and inflammatory disorders (enthesopathy, neuritis, myelitis).

New malignancy developed in 5/82 (6%, three cases of skin malignancy, one each of testicular and prostate cancer). The median time to diagnosis was 40 months (range 38-105) after AHSCT. 18/82 (23%) had other complications reported, which included drug effects (adrenal insufficiency secondary to corticosteroids; marrow toxicity presumed secondary to mercaptopurine) and late effects with uncertain links to AHSCT (hypertension, fibromyalgia, type 2 diabetes mellitus).

Five patients successfully conceived leading to the births of healthy infants post-AHSCT.

Post-AHSCT treatment of Crohn's disease

Seventy-three percent (60/82 patients followed up) resumed medical therapy for Crohn's disease at a median of 10 months (range 1-79) after AHSCT. Thirty-seven percent (30/82) required some form of surgery post-AHSCT, of which 21/82 (26%) underwent major GI surgery (laparotomy, resection, or formation of a stoma) at a median of 26 months (range 6-87). Stoma reversal was performed in 4/82 (5%) patients post-HSCT due to disease regression.

At last follow-up, 42/78 patients (54%) were on treatment. In patients who had re-initiated medical therapy at last follow-up, 24/42 (57%) achieved remission or significant symptomatic improvement with therapies (including anti-TNF therapy in 19/24) to which they had previously lost response or been non-responsive.

DISCUSSION

The principal finding of this retrospective survey using the EBMT registry is that AHSCT in patients with severe, treatment-refractory CD can induce complete remission or significant improvement in around two-thirds (68%) at long-term follow-up. 55% were alive and off all treatment at one year. In a multivariate analysis, extensive disease with perianal disease was associated with adverse treatment-free survival. This is in keeping with the results of ASTIC, which demonstrated that patients with perianal disease or current smokers had a higher incidence of complications following AHSCT.(20) As such, patients with perianal disease should be considered to be at higher risk of complications and relapse requiring re-initiation of treatment. An appreciable minority of 27% remained off all therapy until last follow-up, and 57% of patients who recommenced medical therapy following AHSCT were re-sensitised to therapies to which they had previously been refractory.

Although AHSCT alone does not frequently result in cure or long-term remission, it appears to have profound benefit in this highly refractory and difficult to treat patient population, where disease control and associated quality of life is poor, and life expectancy is reduced. Of note, chronic active CD treated with intense immunosuppressive regimens in the absence of AHSCT is also associated with significant morbidity and increased mortality.(21) This is the largest cohort of patients undergoing AHSCT for CD reported to date, and adds significantly to the evidence supporting its efficacy.

A further important finding of this study is that the safety of AHSCT in this population is similar to AHSCT for other common indications, such as myeloma and lymphoma, reflected by a transplant-related mortality of 1.2%.(7) There was one transplant-related death in our cohort and a second patient died at 7.99 years. 28% developed an infection post-transplant, in keeping with transplant-associated infective complications in other diseases. The three

cases of skin cancer observed may be linked to the longstanding multi-agent immunosuppression experienced by this patient cohort. Optimising supportive care and restricting AHSCT to experienced centres has been shown to help mitigate AHSCT risk.(22, 23)

In accordance with the Joint Accreditation Committee-ISCT & EBMT (JACIE) requirements, all AHSCT procedures in Europe are reported to the EBMT registry. The majority of patients in this study were treated following the 2012 EBMT Guidelines, which formed the basis for patient selection and transplant technique.(6) Through pan-European multi-centre collaboration, we were able to obtain patient-level data, including long-term follow-up. As a retrospective evaluation, however, our study has intrinsic limitations. First, evaluation of clinical response was performed retrospectively. However, to reduce the risk of recall bias, contemporaneous notes were reviewed in all cases. To ensure accurate information, data collection was performed by the patient's treating gastroenterologist. Second, the categorization of clinical response was necessarily broad, which is unlikely to fully reflect the spectrum of clinical disease response. We elected not to collect imaging, endoscopic or biomarker outcomes, as these investigations were not performed in a systematic manner for all patients. Finally, data were not available on quality of life outcomes. Outcomes from a subset of 19 patients in this cohort have been previously reported in a single-centre study.(17)

ASTIC is the only randomised controlled trial of AHSCT for CD to date.(19) The one-year follow-up data of 40 transplant recipients in ASTIC provide further evidence of efficacy, with complete endoscopic healing occurred in 50% of patients, and 47% were judged free of disease on endoscopy and imaging at one year.(20) There was also a significant improvement from baseline to one year post transplant across multiple clinical, quality of life and endoscopic endpoints. Those who did relapse were re-sensitised to TNF therapy to

which they had previously been refractory, as in our study.(20) Single-centre studies with longer-term follow-up have reported that AHSCT does not offer indefinite remission and, as in our study, high rates of restarting medical therapy are observed.(10, 17) However, CD appears to be more responsive to therapy after AHSCT even where a clinical relapse occurs. On this background, our findings lend support to a strategy of AHSCT with re-introduction of drug therapy to enable longer-term remissions in this complex patient cohort.

Recently, ECCO and EBMT have produced a collaborative update and review of the field, offering specific guidance on the clinical role of AHSCT and how it should be delivered.(24) We propose that future CD patients undergoing AHSCT outside of clinical trials are enrolled in a European registry study to ensure harmonisation of outcome assessment. Although our data suggest a complication rate similar to other indications for AHSCT, it must be recognised that AHSCT represents an intensive therapy with significantly higher short-term risks than conventional treatments for CD. Late effects are a risk both for AHSCT and more conventional immunosuppressive therapies due to the cumulative burden of many intense lines of treatment in these complex patients, which even in the absence of AHSCT is associated with significant morbidity and increased mortality.(21) Such late effects are broad in spectrum, affect many organ systems and require systematic evaluation. Our current study highlights some of the issues, for example the skin cancers and secondary autoimmune disease.(25) Long-term follow up of patients combined with prospective data collection should help to evaluate these risks post-AHSCT.

The mechanism of action of AHSCT in CD remains ill-defined. AHSCT has been shown to drive profound changes to the innate and adaptive immune system.(7, 26) First, cytotoxic chemotherapy in combination with T cell depletion ablates autoreactive effector cells that may have been refractory to previous immunosuppressive and biological therapies. Next, the immune system regenerates with thymic reactivation and diversification of the T cell receptor repertoire. New, tolerant regulatory T cells traffic and suppress re-emergent autoreactive T

effector cells. A small pilot study provides some evidence that the immunomodulatory effects of AHSCT apply in CD, with an increase in Foxp3+ T regulatory cells and a reduction in cytokine-secreting effector cells.(27) In CD, there may be additional effects from mobilisation and induction chemotherapy on the gastrointestinal mucosa, changes to the microbiome, and effects from G-CSF and antibiotics. It is likely that a combination of these factors underlies the disease response and regain of responsiveness to agents to which patients were previously refractory.

In conclusion, this study supports the safety and efficacy of AHSCT in patients with severe CD, yielding long-term clinical remissions in a patient cohort refractory to existing medical therapy. Important questions remain. These include defining parameters for selection of the patient subgroup most likely to respond to AHSCT, whether reduced intensity conditioning regimens could reduce safety and improve toxicity, and whether the effect of AHSCT can be optimised with early introduction of post-AHSCT maintenance therapy. Optimizing supportive care and restricting AHSCT to experienced centres is likely to help mitigate AHSCT risk.(22) Additionally, greater insight to the mechanisms by which AHSCT induces self-tolerance may open the door to novel targeted therapies. Further randomised clinical studies are warranted to assess the role of AHSCT in this challenging disease.

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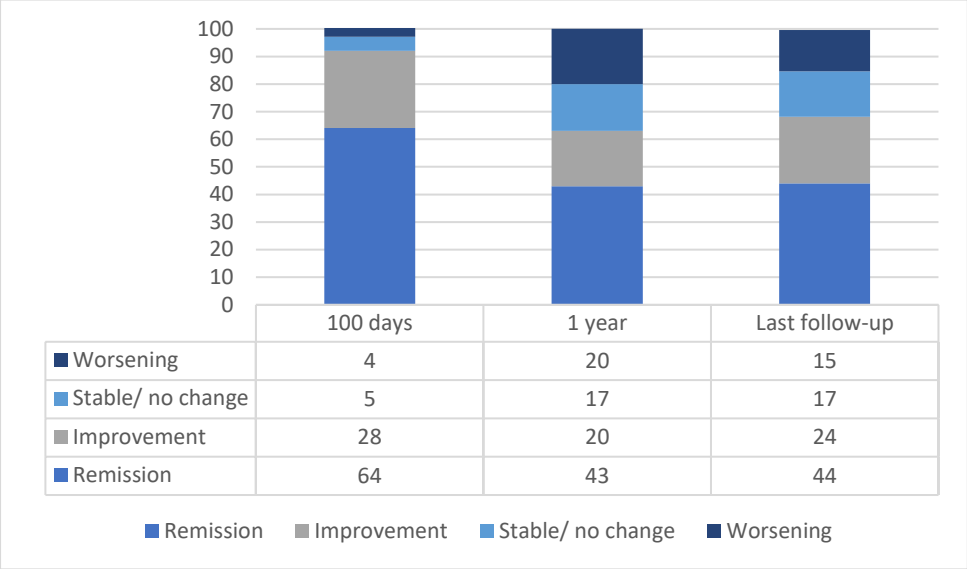
Fig 1: Clinical disease response



Percentage of patients in each clinical disease response category (remission, improvement, stable disease and worsening) at 100 days, 1 year & last follow-up (median 3.4 yrs).

Table 1: Patient & Disease characteristics

Characteristic	n (%)
Patient sex (Female/Male)	52 (63%) F/30 (37%) M
Median age at AHSCT (yrs)	30 (20-65)
Median age at diagnosis (yrs)	17 (2-53)
Extra-intestinal involvement at diagnosis	
None	54 (67%)
Joints+/-skin	15 (18%)
Skin	5 (6%)
PSC	2 (3%)
Other	4 (5%)
Median time from diagnosis to AHSCT (yrs)	12 (1-26)
Disease classification at mobilisation	
Limited	35 (46%)
Extensive without perianal disease	20 (26%)
Extensive with perianal disease	21 (28%)
Disease behaviour at mobilisation	
Stricturing	17 (21%)
Penetrating	8 (10%)
Stricturing+penetrating	14 (17%)
Non-stricturing/non-penetrating	42 (52%)
Perianal (p)	23 (28%)
Median follow-up (mths)	41 (6-174)

569 **Table 2: Previous therapies**

Details	**N (%)**
Previous surgery	61 (74%)
Ileostomy	18 (22%)
Colostomy	5 (6%)
Small bowel resection	24 (29%)
Ileocaecal resection	27 (33%)
Partial colectomy	14 (17%)
Total colectomy	11 (13%)
Proctectomy	6 (7%)
Strictureplasty	11 (13%)
Seton insertion	13 (16%)
Other	17 (18%)
Previous lines of drug therapy	6 (3-10)
Corticosteroids	82 (100%)
Thiopurine	78 (98%)
Methotrexate	66 (82%)
Anti-TNF	81 (99%)
Anti-integrin	16 (20%)
Primary enteral nutrition	23 (28%)

Experimental or other drugs	44 (54%)
Experimental biological therapy (IL6/IL2/IL10/IL17/CCR9/gamma IFN/HDAC inhibition)	10 (12%)
Ustekinumab	8 (10%)
Thalidomide	4 (5%)
Anti-MAd-CAM	3 (4%)
Faecal transplant	3 (4%)
Cyclophosphamide	3 (4%)
Mesenchymal stem cells	2 (2%)
Leukocytapheresis	2 (2%)
Other	9 (11%)

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572 **Table 3: AHSCT details**

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Mobilisation regimen:	
Cyclophosphamide/G-CSF	72 (91%)
G-CSF alone	2 (3%)
Conditioning regimen:	
Cyclophosphamide/ATG	69 (86%)
Cyclophosphamide/CD34+ selection	9 (11%)
Median dose CD34+ (x 10^6/kg)	5.4 (2.4-40.6)
Median time to neutrophil engraftment /days	10 (6-22)
Median time to platelet engraftment /days	10 (1-44)
Engraftment	82 (100%)

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Table 4:

Results from multivariate analysis for treatment-free survival

Variables		**HR**	**CI**	**p**
Age at HSCT > median (30y)		0.81	0.41 - 1.57	0.53
Time from diagnosis to HSCT > median (141 m)		1.20	0.65-2.23	0.56
Female vs male		1.39	0.80-2.48	0.26
Disease classification	Limited	1		
	Extensive with perianal disease	1.61	0.77-3.37	0.20
	Extensive without perianal disease	2.34	1.14-4.83	0.02
Smoker pre-transplant		1.64	0.85-3.15	0.14
Disease behaviour (3 classes)	Non-stricturing/non-penetrating (ref)	1		
	Stricturing	1.11	0.56 - 2.21	0.76
	Penetrating	0.61	0.28 - 1.32	0.21

583 **Supplementary Table 1:**

584 **Contributing EBMT Centres**

Country	Centre	CIC	PI	GI
Belgium	Leuven [Univ H]	209	Daan Dierickx	Severine Vermeire
Germany	Freiburg [University]	810	Jürgen Finke	Peter Hasselblatt/Wolfgang Kreisel
Germany	Jena [Friedrich-Schiller]	533	Inken Hilgendorf	Carsten Schmidt
Germany	Dresden [Universitaets Kl]	808	Martin Bornhäuser	Renate Schmelz
Greece	Thessaloniki [G Papanicolaou G H]	561	Achilles Anagnostopoulos	Jannis Kountouras
Italy	Milano [Osp Maggiore]	265	Francesco Onida	Andrea Cassinotti
Italy	Aviano [CRO]	162	Mariagrazia Michieli	
Italy	Palermo [Osp V Cervello]	392	Rosanna Scimè	Mario Cottone
Norway	Oslo [Rikshospitalet]	235	Tobias Gedde-Dahl	Knut Lundin
Portugal	Lisboa [Inst Oncologia]	300	Manuel Abecasis	João Pereira da Silva
Spain	Barcelona [H Clinic]	214	Montserrat Rovira	Elena Ricart
Spain	Madrid [H Ramón y Cajal]	615	Javier López-Jiménez	Antonio Lopez-Sanroman

Spain	Girona [Oncologia]	433	David Gallardo	David Busquets
Spain	Madrid [Puerta de Hierro]	728	Rafael Duarte	Yago González-Lama
United Kingdom	Edinburgh [Western General]	228	Peter Johnson	Jack Satsangi
United Kingdom	London [Guy`s]	721	Majid Kazmi	Jeremy Sanderson
United Kingdom	London [Kings College H]	763	Majid Kazmi	Ian Forgacs
United Kingdom	Newcastle-Upon-Tyne [Royal Victoria]	276	Matthew Collin	Nick Thompson
United Kingdom	Sheffield [Royal Hallamshire]	778	John Snowden	Alan Lobo

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588 **Supplementary Table 2:** Definitions of disease behaviour:(28)

Stricturing disease	The occurrence of constant luminal narrowing demonstrated by radiologic, endoscopic, or surgical examination combined with pre-stenotic dilation and/or obstructive signs or symptoms but without evidence of penetrating disease.
Penetrating disease	The occurrence of bowel perforation, intra-abdominal fistulas, inflammatory masses and/or abscesses at any time in the course of the disease, and not secondary to postoperative intra-abdominal complication (excludes isolated perianal or rectovaginal fistulae).
Stricturing and penetrating disease	The presence of both stricturing or penetrating phenotypes in the same patient, either at the same moment in time, or separately over a period of time
Non-stricturing, non-penetrating disease	Uncomplicated inflammatory disease without evidence of stricturing or penetrating disease.

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