Stem cell harvesting after bortezomib-based re-induction for myeloma relapsing after autologous transplant: results from the BSBMT/UKMF Myeloma X (Intensive) trial.

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Abstract

The phase III BSBMT/UKMF Myeloma X trial (MMX) demonstrated prospectively, for the first time, superiority of salvage autologous stem cell transplantation (ASCT2) over chemotherapy maintenance for multiple myeloma (MM) in first relapse after prior ASCT (ASCT1). However, many patients have insufficient stored stem cells (PBSC) for ASCT2 and robust evidence for remobilisation after ASCT1 is lacking. We therefore report on the feasibility, safety and efficacy of remobilisation after bortezomib-doxorubicin-dexamethasone reinduction in MMX, and outcomes of ASCT2 with these cells. 110 patients underwent ≥1 remobilisation with 32 and 4 respectively undergoing second and third attempts. Toxicities of remobilisation were similar to those seen in first line mobilisation. After all attempts, 52% of those with insufficient previously stored PBSC had harvested sufficient to proceed to ASCT2. Median PBSC doses infused, neutrophil engraftment and time to discharge after ASCT2 were similar irrespective of stem cell source, as were the toxicities of ASCT2. No significant differences between PBSC sources were noted in depth of response to ASCT or time to progression. Harvesting after bortezomib-doxorubicin-dexamethasone re-induction for MM at first relapse is safe and feasible, and yields a reliable cell product for second ASCT. The study is registered with ClinicalTrials.gov (NCT00747877) and EudraCT (2006-005890-24).

Introduction

The demonstration of the efficacy of high dose melphalan in 1983[1], and later autologous stem cell transplant (ASCT), for multiple myeloma (MM), ushered in a new era of MM therapy. Subsequent randomised controlled trials demonstrated improved response rates and in some studies improved progression-free (PFS) and overall survival (OS) compared to conventional chemotherapy[2-7]; as a result ASCT quickly became established front line therapy for patients deemed sufficiently fit. The incorporation of newer biological 'novel agents' (thalidomide, lenalidomide and bortezomib) into induction, consolidation and maintenance regimens has since allowed further improvements in the outcomes of ASCT for myeloma in first line therapy[8-15] (reviewed in[16]).

Unfortunately, despite advances in therapy, the vast majority of patients with MM relapse, and although widely adopted for front line treatment and often used in relapse, the role of ASCT in the management of relapsed disease (salvage ASCT) had until recently not been rigorously assessed. The randomised, open-label, phase III BSBMT/UKMF Myeloma X trial (MM X) compares high-dose melphalan plus salvage ASCT (HDT-ASCT) against weekly cyclophosphamide (C-weekly) after re-induction with PAD (bortezomib, doxorubicin and dexamethasone) for relapsed MM, and has shown a clear benefit in time to progression (TTP) for those patients receiving ASCT[17]. In light of this strong evidence, a salvage ASCT should be considered at first relapse for all transplant-eligible patients having an initial response to ASCT of >18 months. Crucially,

however, the modality is often considered only for those with sufficient stored stem cells from their initial harvest.

Peripheral blood stem cell (PBSC) collection after first induction therapy is a well-validated and widely employed practice in myeloma therapy worldwide[18]; it is convenient and safe and results in enhanced granulocyte and platelet engraftment compared to bone marrow-derived stem cells[19-21]. What is less clear, is whether stem cells can be successfully mobilised and harvested after re-induction therapy for patients relapsing after first line ASCT. The impact of previous treatment on the stem cells and their niche, and whether such cells constitute a safe, reliable and equivalent product for subsequent ASCT require clarification. As a secondary endpoint of the MM X trial we therefore evaluated the feasibility of PBSC mobilisation after re-induction therapy for first relapse after prior ASCT.

Methods

Study design and patients

The NCRI Myeloma X Relapse (Intensive) phase 3 trial is a randomised, multi-centre, open-label, parallel-group comparison between salvage ASCT and C-weekly as consolidation after PAD reinduction for multiple myeloma at first relapse or progressive disease (Figure 1). Patients were recruited from 51 NHS hospitals and were eligible if they were over 18 years of age, required treatment for first relapse or progressive disease at least 18 months after a previous ASCT (reduced to 12 months in 2011[22]) and

were deemed fit enough to undergo intensive treatment. Patients were excluded if they had received therapy for their relapsed disease, had an ECOG performance status of 3-4, grade 2 peripheral neuropathy, known resistance to PAD, or comorbidity that would preclude high-dose chemotherapy. Full details of inclusion and exclusion criteria for the trial, sample size, laboratory testing at trial entry, ethical review and composition of the trial management group are published elsewhere[17]. The study is registered with ClinicalTrials.gov (NCT00747877) and EudraCT (2006-005890-24).

Disease and response assessments

Response and disease progression were assessed according to the IMWG uniform response criteria[23] and were confirmed by a central laboratory and by an independent myeloma physician masked to treatment allocation at baseline, after re-induction, 100 days after ASCT (or 30 days after C-weekly), every year after randomisation and at disease progression. Full details of cytogenetic analysis are available in Cook *et al.*[17].

Trial procedures, randomisation and masking

All patients received re-induction chemotherapy with 2-4 cycles of PAD (intravenous (IV) twice weekly bortezomib and IV doxorubicin with oral dexamethasone; those achieving ≥VGPR after two cycles discontinued PAD, the remainder continued to four cycles unless precluded by toxicity); those with complete response (CR), partial response (PR) or stable disease (SD) were eligible to proceed to the randomisation if they had an

adequate stem cell dose stored (defined as ≥2x10^6 CD34$^+$ cells per kg, or ≥2x10^8 peripheral blood mononuclear cells per kg) and no clinical evidence of deterioration in cardiac function since registration. Patients without an adequate stem cell dose underwent PBSC mobilisation and harvesting, and could then proceed if an adequate dose was available (combining previously stored and re-harvested cells). Those with an adequate dose already stored prior to trial entry could also undergo mobilisation and harvesting at clinician and patient discretion. Mobilisation regimens were at the discretion of treating physicians (see results section and Figure 2). Patients eligible to proceed were then randomly assigned on a 1:1 basis to receive either high dose melphalan and ASCT or weekly cyclophosphamide; those receiving C-weekly will not be considered further in this report. Patients randomised to the HDT-ASCT arm received a single infusion of intravenous melphalan (200mg/m^2) followed by PBSC infusion after 24 to 48 hours.

Outcomes

The primary endpoint was TTP (published elsewhere[17]); secondary endpoints were objective response, progression-free survival, overall survival, pain and quality of life, and assessment of the feasibility, safety and efficacy of stem cell remobilisation and the outcomes when these PBSC are used for subsequent ASCT. Toxicity and safety were assessed using NCI CTCAE criteria (version 3.0).

Statistical methods

Full details of statistical methods are given in the supplementary material.

Results

Enrolment

Between April 2008 and November 2012, 297 patients were registered (Figure 1), 293 of whom went on to receive treatment with PAD induction[17]. 276 patients (92.9%) had at least stable disease and remained in the trial after re-induction therapy – these patients were eligible for randomisation to either high-dose melphalan and ASCT rescue or oral cyclophosphamide only if they had adequate stem cells available to allow ASCT (>$2x10^6$ $CD34^+$ /kg). 170 patients (57.2%) already had some stem cells stored at trial entry (median dose $3.3x10^6$ $CD34^+$ /kg, range 0.6-13) of whom 149 (50.2%) had a sufficient dose to proceed to ASCT (>$2x10^6$ $CD34^+$/kg): Figure 1. 26 patients with an adequate stored dose elected to undergo re-harvesting anyway, along with 84 of the 127 patients without sufficient stored cells. Mobilisation and peripheral blood stem cell (PBSC) collection was therefore undertaken within the trial for 110 patients, of whom 26 (23.6%) already had an adequate dose stored. Patient characteristics are illustrated in Table 1. Of patients with at least stable disease and without adequate stored cells, harvesting was not undertaken in 43 (33.9%), due to clinician decision (n=22), death (n=17; due to progressive disease in n=13), withdrawal from the trial (n=3), or progressive disease (n=1). After all harvesting attempts, 193 patients (65.0%) remained

in remission and had adequate stored cells to proceed; 174 were then randomised to either high-dose melphalan with ASCT rescue (n=89) or oral cyclophosphamide (n=85).

Harvesting procedures

Of the 110 patients who underwent PBSC harvesting, 78 (70.9%) underwent only one attempt at mobilisation, 28 (25.5%) tried twice, and 4 (3.6%) patients made three attempts. The first mobilisation was a median of 21 days after the last cycle of PAD (range 1-126). Mobilisation regimens were at the discretion of treating physicians, and are illustrated in Figure 2 (see also Supplementary Table 1). Of the 110 remobilised patients, 54 (49.1%) achieved a satisfactory cell dose after all attempts: 41 (37.3%) at the first mobilisation attempt (yield unknown, n=23), 10 (31.3%) at the second attempt (yield unknown, n=6) and 3 (75%) at the third attempt. There was no difference in yields between first, second and third attempts (p=0.9699, Kruskal-Wallis test). Two patients (1.8%) withdrew during PBSC mobilisation, 7 (6.4%) died during mobilisation (6 with progressive disease: 5.5%, 1 (0.9%) with infection), 1 (0.9%) patient had progressive disease and was ineligible to continue and 30 patients (27.3%) did not mobilise sufficient stem cells after all attempts. Taking into account previously stored as well as re-harvested cells, 70 patients (63.6%) of the 110 who underwent re-harvesting were therefore able to proceed to randomisation. Since 26 of these patients already had an adequate stored PBSC dose, 44 (52%) of the 84 patients without an adequate stored dose had achieved an adequate PBSC collection and were able to proceed. Adverse events occurring between completion of induction therapy and randomisation to

consolidation therapy are shown in Table 2. Although sensory neuropathy was reported during stem cell harvesting, when this was compared to adverse events reported during PAD treatment the prevalence and severity of sensory neuropathy was in general lower during PBSC harvesting (80% of patients experienced a lower grade of sensory neuropathy during stem cell harvesting than at the end of PAD, Supplementary Table 2), implying that as might be anticipated this toxicity is secondary to PAD therapy rather than PBSC mobilisation and harvesting.

ASCT procedures

89 patients were randomised to receive HDT and ASCT rescue, of whom 83 (93.3%) completed the procedure. Of these, 42 (50.6%) received stem cells stored prior to trial entry (PBSC1), 29 (34.9%) received cells harvested after PAD (PBSC2) and 11 (13.3%) received a combination of cells from the two sources (PBSCMix); the stem cell source is unknown for one patient. These groups were not matched within the trial protocol, but had broadly similar pre-ASCT characteristics (Table 1). Gender, age, paraprotein heavy and light chain isotypes, previous therapies, response to prior ASCT, and blood results (e.g. haemoglobin, platelet count, creatinine clearance, bilirubin, alanine aminotransferase) at randomisation were not significantly different between groups. Time from registration to randomisation was longer in PBSC2 and PBSCMix than PBSC1, reflecting the additional time taken for stem cell mobilisation and harvesting. Some differences between groups were noted in ISS stage at diagnosis, PFS after 1st ASCT,

time from diagnosis to randomisation and 13q deletion (Table 3); these are discussed further below.

Median stem cell doses infused, neutrophil engraftment and time to discharge were similar across groups ($p>0.05$ in each case; Table 3). Although the stem cell dose infused was not significantly different between groups, the slightly lower dose in the PBSCMix group may account for the non-significantly prolonged time to discharge in that group: PBSCMix: 18 days, PBSC2: 16 days, PBSC1: 16 days ($p=0.4980$). Platelet engraftment was also slightly slower in PBSC2 and PBSCMix than PBSC1, again possibly reflecting the slightly lower median stem cell dose. Adverse events reported during high dose melphalan and ASCT consolidation were in line with published reports and are shown in Supplementary Table 3. No significant differences were seen in toxicities according to stem cell source. Only 9 serious adverse events (SAEs) were reported in relation to ASCT (infection n=6, 7.3%; GI disturbance n=1, 1.2%; neoplasia n=2, 2.4%) thus no inferences are made about the frequency of SAEs or readmission in relation to PBSC source.

Outcomes

Maximal responses to ASCT are shown in Supplementary Table 4. An ordinal logistic regression showed no differences between stem cell sources in terms of response to ASCT ($X^2 = 1.53$ with 2 degrees of freedom, $p=0.4647$, adjusting for the trial stratification factors). Median TTP was as follows: PBSC1: 18 months (95% CI: 13-27), PBSC2: 24

months (19-28), PBSCMix: 33 months (11-∞); p=0.3553 (Figure 3). Hazard ratios for TTP as compared with PBSC1: PBSC2 0.85 (95% CI: 0.45-1.63), PBSCMix: 0.49 (0.19-1.3).

Discussion

The MM X trial has demonstrated for the first time in a prospective multi-centre randomised phase III study that following bortezomib-doxorubicin-dexamethasone re-induction for MM at first relapse, salvage ASCT results in significantly longer TTP than chemotherapy consolidation therapy. Clearly, this finding will be of crucial importance for patients and myeloma physicians when choosing therapy for transplant-eligible patients relapsing after a prior ASCT. However, this therapeutic option naturally depends on the availability of stored stem cells for ASCT – a resource no longer remaining for many patients after their first ASCT. Key questions, therefore, are whether it is feasible and safe to mobilise and harvest stem cells from such patients after re-induction therapy, and whether those stem cells are suitable for subsequent ASCT.

Of the 110 patients who underwent PBSC harvesting in the MM X trial, after all attempts, 64% were able to mobilise and harvest an adequate dose to allow salvage ASCT. Only 32 proceeded to a second attempt and of those only 4 to a third attempt. Interestingly the rate of successful harvesting remained high at these subsequent attempts suggesting more patients could have benefitted from repeated mobilisations, which is in keeping with the majority of previous data[24-26] . Nevertheless, only 30

patients left the trial due to inability to mobilise sufficient stem cells. Thus, the majority of patients are able to harvest sufficient stem cells after bortezomib salvage therapy for a second high dose procedure.

The mobilisation regimens used for PBSC harvesting within the trial were at the discretion of the treating physicians and accordingly a range of regimens was employed. The majority of patients received cyclophosphamide and G-CSF, and a smaller number G-CSF alone, in keeping with standard practice[18,27]. A number of mobilisations also employed the CXCR4 antagonist, Plerixafor, particularly for second and third mobilisation attempts, either as a planned treatment or to salvage a failed mobilisation with other agents. As suggested by previous retrospective studies[24,25,28,29], the drug was effective in this context, and improved yields were seen compared to G-CSF alone, as in the setting of harvesting during first line treatment[30]. There is ongoing debate about the relative costs and efficacies of Plerixafor *vs.* chemotherapy based regimens[31]. Since mobilisation regimen was not a controlled randomisation within our trial a direct comparison is therefore not possible, although similar efficacies were seen (Figure 2 and Supplementary Table 1). Since the inception of this study Plerixafor has gained a product license and is funded by the UK NHS for both mobilisation after a previous failed attempt and pre-emptive mobilisation in patients with inadequate $CD34^+$ mobilisation on the planned day of harvesting, and it is therefore likely that its use for remobilisation at relapse will increase. Given that the majority of patients who failed to harvest a

successful stem cell dose were not exposed to Plerixafor, it is likely that regular use of the drug may well further increase the proportion of successful harvests in the future.

Stem cell remobilisation was well tolerated, with neutropenia, thrombocytopenia and infection being the main toxicities encountered, at frequencies similar to published reports of harvesting after first line therapy[30,32-34]. Sensory neuropathy was frequently reported during mobilisation and harvesting, although this almost certainly relates to the preceding treatment with bortezomib. Reassuringly, we did not see a marked increase in toxicity with repeated mobilisation attempts, in keeping with the published data on toxicity of remobilisation[35]. Given the efficacy of salvage ASCT in this context and the high chance of success already discussed, further attempts after one failed mobilisation are therefore both rational and safe.

Patients undergoing ASCT with cells previously stored (PBSC1), harvested after re-induction within the trial (PBSC2) or a combination of the two (PBSCMix) had similar outcomes: there were no statistically significant differences in neutrophil engraftment or time to discharge between groups. Platelet engraftment was slightly slower in the PBSC2 and PSBCMix groups, possibly reflecting the lower mean stem cell dose in those groups, although other authors have also reported delayed platelet engraftment in this setting in a small cohort[36]. Responses by IMWG criteria were comparable. In comparing the outcomes of the PBSC2/PBSCMix and PBSC1 groups, it is important to acknowledge that since re-mobilisation incurs a further period of time prior to ASCT, a selection bias

may be introduced whereby only those with better-performing disease (i.e. those remaining in remission) can undergo re-harvesting. Since median time from PAD to remobilisation was 21 days, indicating the majority of patients were remobilised rapidly once recovered from induction, this is not expected to impact significantly upon results. Whilst it is tempting to speculate that bortezomib re-induction, or therapy preceding it, might alter the characteristics of the harvested stem cell product, no significant differences in outcome have thus far been seen between groups. Nonetheless, there is potential for genetic or epigenetic damage to stem cells by preceding therapy, which might plausibly increase the risk of sequelae such as myelodysplasia. Previous retrospective studies have suggested an increased risk of myelodysplasia in patients receiving stem cells harvested after prior therapy[24,37], particularly in more heavily pre-treated cohorts[24,37]; ongoing clinical follow-up of the groups and planned companion studies in this prospective study to evaluate key biological characteristics of stem cells stored previously and harvested after bortezomib-based re-induction will allow further characterisation of the stem cell products and differences in outcomes. Importantly, the toxicities of ASCT were similar to published reports[3,7], irrespective of stem cell source.

In light of the findings of the MM X trial, mobilisation and harvesting of sufficient PBSC for at least two ASCT procedures is a rational strategy after 1st induction treatment for all transplant eligible patients. Recent data suggest that with modern harvesting regimens, for example incorporating Plerixafor[38], the vast majority of patients can achieve this target. It should be emphasised that despite the encouraging results

presented here some patients were unable to proceed to ASCT2 due to inability to mobilise PBSC. Other researchers have reported variables which may predict poor remobilisation at relapse, such as thrombocytopenia, anaemia, bone marrow cellularity, hypoalbuminaemia etc.[37] and this is of interest, but such observations are unlikely to be helpful in clinical decision making since these variables can be assessed only once cells are needed and not available. We therefore consider that the goal should always be to harvest enough PBSC at first line to allow a second ASCT at relapse. Nevertheless, for many current and future patients, PBSC will not be stored for a second high dose procedure – there are many reasons for this including storage limitations, costs associated with long term stem cell storage and concerns about their viability, lack of evidence for second ASCT at the time of harvesting, inability to harvest sufficient cells with older, less effective regimens and the fact that in many healthcare economies a second HDT/ASCT may not be affordable or funded (for example Medicare in the United States will reimburse for only one ASCT in those patients achieving at least a PR in response to chemotherapy; tandem or multiple transplantation is not covered). Indeed, in our trial, of the 276 patients who completed re-induction therapy, only 149 had an adequate stored PBSC dose from their previous transplant to allow a second ASCT. For patients in this situation, mobilisation and harvesting after a bortezomib-doxorubicin-dexamethasone re-induction regimen is feasible and safe, and importantly it facilitates a superior therapeutic option for patients with myeloma at first relapse.

Contributors

Authorship was determined in accordance with a pre-existing Trial Management Group policy delineated in the protocol. GC designed the study and GC, JMB, DAC and TCMM analysed the data. CP provided independent review of response and wrote the manuscript with GC. CW, JMB, DAC, JC, JAS, JA, JiC, HH, JMB, AC, KY, SS, SC and TCMM collected data, revised the article and gave final approval.

Conflicts of interest

CP has received support to attend scientific meetings from Janssen; TCMM has received research support and support to attend scientific meetings from Janssen; CDW has received research funding, and advisory board and speaker bureau fees from Janssen; JC has received research funding and advisory board and speaker bureau fees from Janssen; JAS has received honoraria, research funding and speakers bureau fees from Janssen, honoraria for educational events from Celgene and has participated in advisory board for Genzyme/Sanofi; JMB has received honoraria and speakers bureau fees from Janssen; GC has received honoraria, research funding and speakers bureau fees from Janssen. DAC, JA, HH, AC, JMB, KY, SS and SC have no conflicts of interest to declare. The funders of the study had no role in study design, data collection, data analysis, data interpretation or preparation of the manuscript.

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Supplementary Material

Supplementary material is available at Biology of Blood and Marrow Transplantation's website.

Figures and Tables

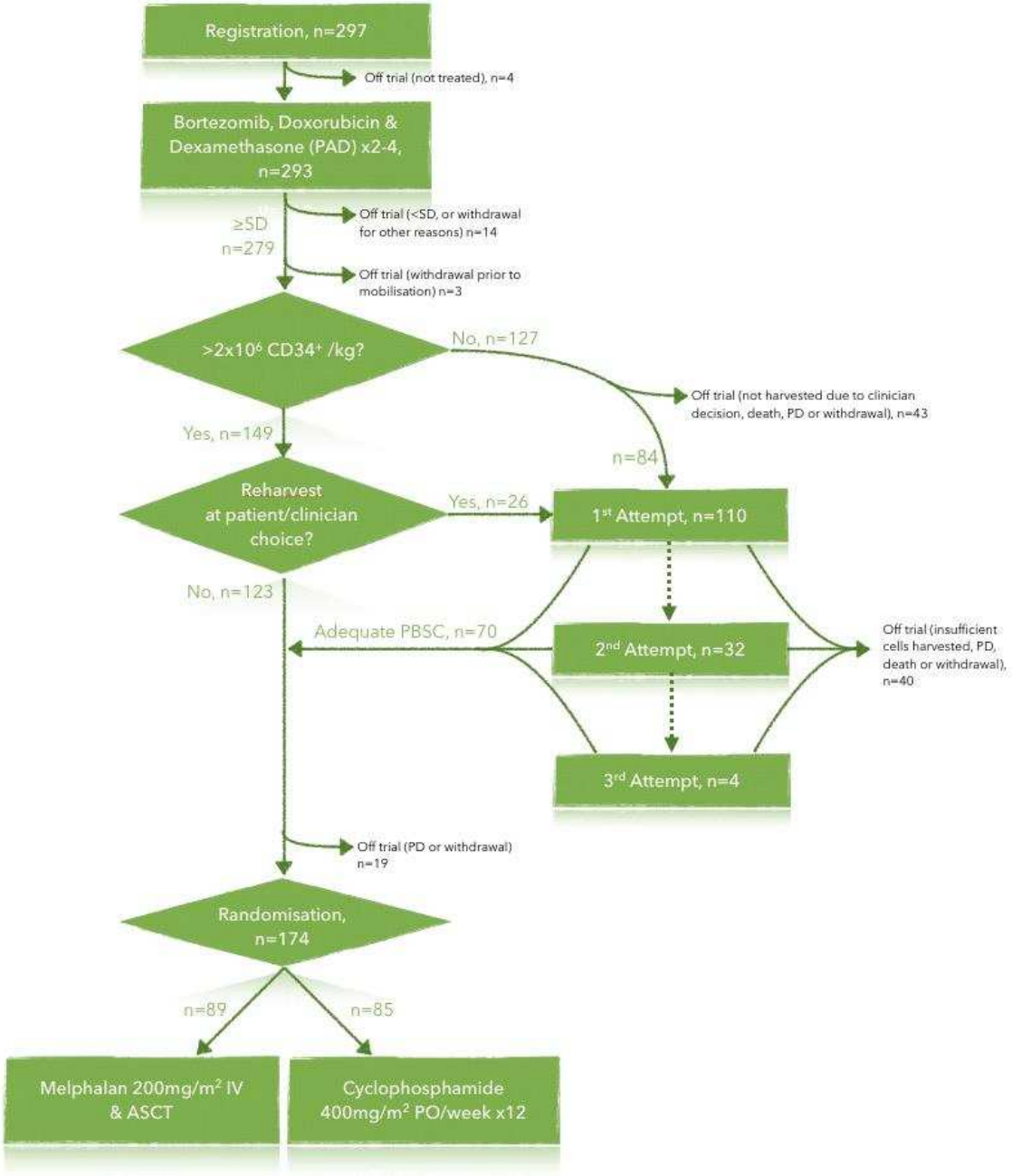

Figure 1: Trial CONSORT Diagram. SD=stable disease, PD=progressive disease, PBSC=peripheral blood stem cell, ASCT= autologous stem cell transplant. A full consort diagram for the trial is published (Cook *et al.*[17]).

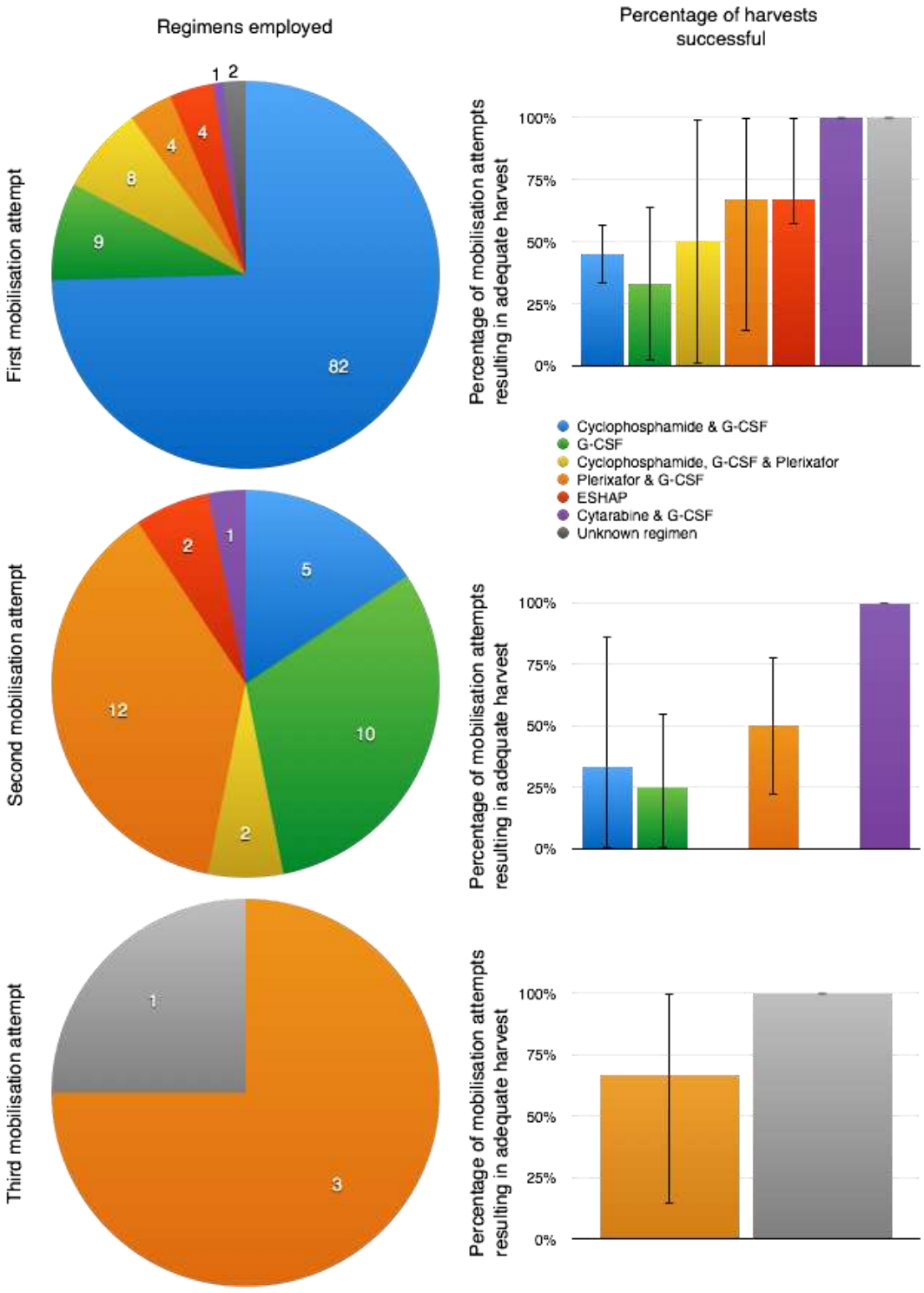

Figure 2: Mobilisation regimens employed and percentage of mobilisation attempts leading to successful PBSC harvest at first, second and third mobilisation attempts. Error bars show 95% confidence intervals.

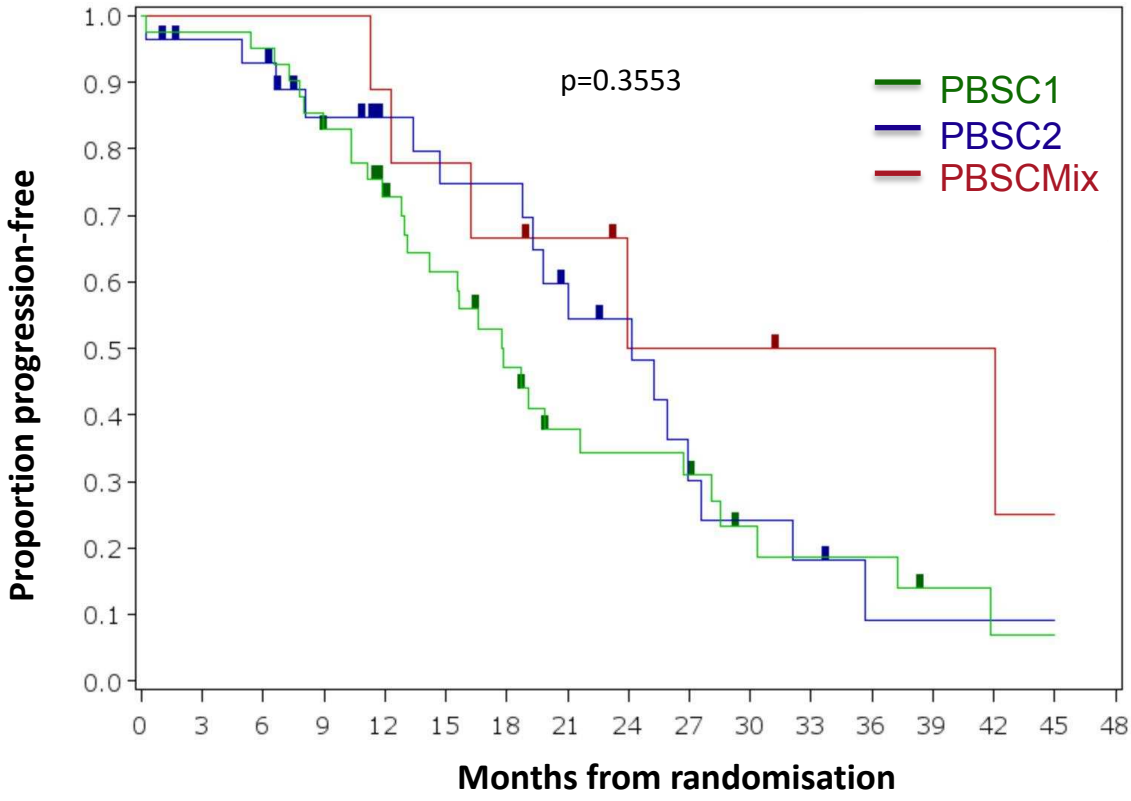

Figure 3: Kaplan-Meier curves after ASCT for patients receiving PBSC harvested prior to trial entry (PBSC1), re-harvested in the trial (PBSC2) and a combination of both sources (PBSCMix).

		Registered patients (n=297)	Remobilised patients (n=110)	Patients undergoing ASCT			
				PBSC1 (n=42)	PBSC2 (n=29)	PBSCMix (n=11)	p-value[#]
Gender	Male	208 (70.0%)	76 (69.1%)	31 (73.8%)	20 (69.0%)	10 (90.9%)	0.413
	Female	89 (30.0%)	34 (30.9%)	11 (26.2%)	9 (31.0%)	1 (9.1%)	
Age at randomisation (median)		61	62	61	63	59	0.370
Myeloma isotype	IgG	190 (64.0%)	69 (62.7%)	26 (61.9%)	19 (65.6%)	8 (72.7%)	0.955
	IgA	55 (18.5%)	22 (20%)	6 (14.3%)	5 (17.2%)	2 (18.2%)	
	IgM/IgD	3 (1.0%)	1 (0.9%)	1 (2.4%)	0	0	
	Light chain	26 (8.8%)	11 (10%)	5 (11.9%)	2 (6.9%)	0	
	Non-secretory	9 (3.0%)	4 (3.6%)	1 (2.4%)	2 (6.9%)	0	
	Missing	14 (4.7%)	3 (2.7%)	3 (7.1%)	1 (3.4%)	1 (9.1%)	
Light chain isotype	Lambda	82 (27.6%)	30 (27.3%)	15 (35.7%)	4 (13.8%)	3 (27.3%)	0.326
	Kappa	185 (62.3%	68 (61.8%)	21 (50.0%)	20 (69.0%)	6 (54.5%)	
	Missing	30 (10.1%	3 (2.7%)	6 (14.3%)	5 (17.2%	2 (18.2%)	
ISS stage at diagnosis	I	88 (29.6%)	34 (30.9%)	9 (21.4%)	12 (41.4%)	2 (18.2%)	0.049
	II	93 (31.3%)	28 (25.5%)	11 (26.2%)	8 (27.6%)	1 (9.1%)	
	III	38 (12.8%)	17 (15.5%)	10 (23.8%)	1 (3.4%)	5 (45.5%)	
	Missing	78 (26.3%)	31 (28.2%)	12 (28.6%)	8 (27.6%)	3 (27.3%)	
Previous therapies	Vincristine	84 (28.8%)	33 (30%)*	14 (33.3%)	10 (35.7%) *	4 (36.4%)	0.821
	Thalidomide	202 (68.9%	75 (68.2%)	28 (66.7%)	23 (79.3%)	8 (72.7%)	0.515
	Bortezomib	13 (4.4%)	6 (5.5%)*	2 (4.8%)	0 *	0	0.554
	Bisphosphonate	226 (77.1%)	85 (77.3%)	35 (83.3%)	21(72.4%)	7 (63.6%)	0.288
Response to prior ASCT	**s**CR/CR	157 (54.1%)*	61 (55.5%)	23 (54.8%)	17 (58.6%)	6 (54.5%)	0.282
	VGPR/PR	119 (41.0%)*	45 (40.9%)	17 (40.5%)	12 (41.4%)	3 (27.3%)	
	SD	14 (4.8%)*	4 (3.6%)	2 (4.8%)	0	2 (18.2%)	
PFS after 1st ASCT /years (median, range)*		2.5 (0.4-12.4)	2.6 (1-12.4)	2.3 (1.1-6.6)	3.0 (1.3-12.2)	3.4 (1.6-12.4)	0.031
Time from diagnosis to randomisation /years (median, range)*				3.6 (2.2-8.9)	4.8 (2.7-13.3)	4.8 (29-13.6)	0.019
Time from registration to randomisation /months (median, range)				3.5 (1.6-4.9)	4.9 (3-9.7)	4.6 (3.2-5.5)	<0.001
Cytogenetics at trial entry*							
Patients with available data		149	55	22	8	7	
t(4;14)		14 (9.4%)	6 (10.9%)	3 (13.6%)	2 (25%)	0	0.079
t(11;14)		15 (10.1%)	5 (9.1%)	1 (4.5%)	0	1 (14.3%)	0.065
t(14;16)		3 (2.0%)	1 (1.8%)	0	0	0	
17p del		11 (7.4%)	2 (3.6%)	2 (9.1%)	1 (12.5%)	0	0.102
13q del		58 (38.9%)	21 (38.2%)	15 (68.2%)	2 (25%)	3 (42.9%)	0.023
Hyperdiploidy		20 (13.4%)	11 (20%)	2 (9.1%)	1 (12.5%)	0	0.102
Adverse risk cytogenetics		24 (16.1%)	7 (12.7%)	4 (18.2%)	2 (25%)	0	0.085

Table 1: Pre-ASCT characteristics by stem cell source subsequently used.
* Data not available for all patients; figures and percentages given are for known patients. Cytogenetics are by interphase fluorescence *in-situ* hybridisation
#P-value compares distribution of characteristics by stem cell source (PBSC1 *vs.* PBSC2 *vs.* PBSCMix). Continuous variables are compared using the Kruskal-Wallis test and categorical variables using Fisher's exact test.

	All grade toxicity, n (%)			Grade 3-4 toxicity, n (%)			p
Total number of mobilisation attempts undertaken	1 (n=74)	2 (n=32)	3 (n=4)	1 (n=74)	2 (n=32)	3 (n=4)	
Neutropenia	23 (31.1)	13 (40.6)	2 (50)	15 (20.3)	8 (25)	1 (25)	0.588
Thrombocytopenia	30 (40.5)	14 (43.8)	2 (50)	11 (14.9)	8 (25)	0	0.604
Infection	8 (10.8)	6 (18.8)	1 (25)	4 (5.4)	4 (12.5)	1 (25)	0.471
Nausea	11 (14.9)	7 (21.9)	0	1 (1.4)	1 (3.1)	0	0.508
Vomiting	6 (8.1)	4 (12.5)	0	1 (1.4)	2 (6.3)	0	0.608
Skin toxicity	3 (4.1)	2 (6.25)	0	0	1 (3.1)	0	0.475
Pyrexia	1 (1.4)	4 (12.5)	0	0	1 (3.1)	0	0.127
Neuropathy (motor)	1 (1.4) *	0	0	0 *	0	0	-
Neuropathy (sensory)	21 (28.4)	10 (31.3)	1 (25)	4 (5.4) #	3 (9.4)	0	0.920
Lethargy	12 (16.2)	8 (25)	0	0	0	0	0.729
Anaemia	7 (9.5)	2 (6.3)	0	0	0	0	-
Diarrhoea	3 (4.1)	3 (9.4)	0	0	0	0	-
Thrombosis	1 (1.4)	1 (3.1)	0	1 (1.4)	1 (3.1)	0	-
All other toxicities	≤2 (≤2.7)	≤1 (≤3.1)	≤1 (≤25)	0	0	0	-
Safety data missing	1 (1.4)	0	0	1 (1.4)	0	0	-

Table 2: Frequency of toxicities reported between end of induction and randomisation, in patients undergoing 1, 2 and 3 attempts at PBSC mobilisation. * 1 patient experienced motor neuropathy of unknown grade. # 2 further patients experienced sensory neuropathy of unknown grade. P-values are for Fisher's exact test comparing all-grades of toxicity across groups defined by the number of attempts at PBSC mobilisation.

	PBSC1 (n=42)	PBSC2 (n=29)	PBSCMix (n=11)	p-value[#]
Stem cells infused /x10^6 $CD34^+$ /kg: median (range)	3.2 (1.6-13.6)	2.9 (1.4-7.0)	2.6 (2.1-6.6)	NS
Days to neutrophils >0.5x10^9/L: median (range)	12.0 (3-25)	12 .0 (4-25)	12 .0 (11-33)	NS
Days to platelets >20x10^9/L: median (range)	12.5 (7-105)	12 (6-82)	16 (9-33)	NS
Days to platelets >50x10^9/L: median (range)	18.0 (10-179)	21.0 (6-124)	24.0 (16-146)	0.033
Days to discharge after ASCT: median (range)	16.0 (9-45)	16.0 (12-33)	18.0 (12-50)	NS

Table 3: ASCT procedure characteristics by stem cell source.
#P-value compares distribution of characteristics by stem cell source. Continuous variables are compared using the Kruskal-Wallis test and categorical variables using Fisher's exact test.

Supplementary Table Legends

Supplementary Table 1: Mobilisation regimens and stem cell harvest outcomes. Abbreviations: G-CSF: granulocyte colony stimulating factor, ESHAP: etoposide, methylprednisolone, cytarabine and cisplatin. * Percentage of collections where the yield is known and is >2x10^6 $CD34^+$ /kg.

Supplementary Table 2: Sensory neuropathy (CTCAE grade) for patients at the end of PAD and then at the end of mobilisation.

Supplementary Table 3: Frequency of toxicities reported during high dose melphalan and ASCT. P values are for Fisher's exact test comparing all-grades of toxicity across groups defined by the source of stem cells (PBSC1, PBSC2, PBSCMix).

Supplementary Table 4: Maximal responses to stem cell transplant. 1 patient died within 100 days of ASCT – the stem cell source used for this patient is not known. 6 patients who were randomised to ASCT did not undergo the procedure.

Supplementary Table 5: Co-investigators contributing to this study.

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