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Autologous stem cell transplantation for progressive multiple sclerosis: Update of the European Group for Blood and Marrow Transplantation autoimmune diseases working party database

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Over the last decade, hematopoietic stem cells transplantation (HSCT) has been increasingly used in the treatment of severe progressive autoimmune diseases. We report a retrospective survey of 183 multiple sclerosis (MS) patients, recorded in the database of the European Blood and Marrow Transplantation Group (EBMT). Transplant data were available from 178 patients who received an autologous graft. Overall, transplant related mortality (TRM) was 5.3% and was restricted to the period 1995–2000, with no further TRM reported since then. Busulphan-based regimens were significantly associated with TRM. Clinical status at the time of transplant and transplant techniques showed some correlations with toxicity. No toxic deaths were reported among the 53 patients treated with the BEAM (carmustine, etoposide, cytosine-arabioside, melphalan)/antithymocyte globulin (ATG) regimen without graft manipulation, irrespective of their clinical condition at the time of the transplant. Improvement or stabilization of neurological conditions occurred in 63% of patients at a median follow-up of 41.7 months, and was not associated with the intensity of the conditioning regimen. In this large series, HSCT was shown as a promising procedure to slow down progression in a subset of patients affected by severe, progressive MS; the safety and feasibility of the procedure can be significantly improved by appropriate patient selection and choice of transplant regimen. *Multiple Sclerosis* 2006; **12**: 814–823. <http://msj.sagepub.com>

Key words: autoimmune diseases; immunosuppression; multiple sclerosis; stem cells transplantation; treatment safety

Introduction

In the last decade, intense immunosuppression followed by autologous transplantation of hematopoietic stem cells (AHSCT) has been proposed as a possible strategy for the treatment of severe autoimmune disorders (ADs) [1–4], including multiple sclerosis (MS). The graft is made up of hematopoietic stem cells (HSCs), which are usually collected from peripheral blood (PB). Under physiological conditions, HSCs are mostly located in the bone marrow (BM), but can be mobilized into the PB by the administration of a non-myeloablative chemotherapy, followed by haematopoietic growth factors, such as granulocytic colony stimulating factor (G-CSF). The most common mobilization regimen in ADs is cyclophosphamide (CY), ranging between 1.5 and 4 g/m², followed by G-CSF [5]. G-CSF alone can also be used as a unique mobilizing agent. In an autologous setting, PB-HSCs are preferred to BM-HSCs because the shorter engraftment of PB-HSCT results in a safer procedure. After the harvest of HSCs, the patient is treated with a conditioning or preparatory regimen, consisting of high doses of immunosuppressive drugs, which may also include total body irradiation (TBI), followed by the reinfusion of HSCs. The graft may be depleted of autoreactive cells by different methods of *ex vivo* manipulation (graft purging), ie,

CD34 positive cells selection or T-cell depletion. An *in vivo* depletion of autoreactive T cells, which could either have been reintroduced with the graft or which escaped the conditioning regimen, is usually carried out by either animal-derived antithymocyte antiserum (ATG) or monoclonal antibodies.

The rationale for AHSCT derives from the possibility of eradicating self-reactive immune cells by intensive immunosuppression, followed by full immune reconstitution on the engraftment of the autologous HSCs. Beyond its immunosuppressive potential, AHSCT may also be beneficial for the possible ‘resetting’ of the immune system due to a *de novo* regeneration of the T-cell compartment, with an extensive renewal of clonal specificities after therapy, thereby becoming tolerant against self-antigens [6], possibly for an extended period of time. This has never been shown previously with purely immunosuppressive treatments. Tolerance-inducing mechanisms of action of HSCT in ADs have been recently reviewed [7].

In MS, currently available therapies, such as interferons and glatiramer acetate, are of limited efficacy and many cases continue to deteriorate. Mitoxantrone, the only approved immunosuppressive treatment for MS, is now used when first line immunomodulating therapy fails, but because of its cumulative cardiotoxicity, the total dosage of

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100–120 mg/m² cannot be exceeded and, therefore, its use has to be limited [8–10].

In recent years, a number of studies have addressed the feasibility, toxicity, impact on MS progression and MRI findings of AHSCT, utilizing different mobilization and conditioning regimens [11–18]. Data published so far show that AHSCT is a promising approach to MS cases which fail the first line disease modifying drugs (DMDs) and continue to rapidly worsen. However, such an intensive therapy carries morbidity and mortality risks that can only be justified by disease severity and proven benefits.

In 1997, the Autoimmune Diseases Working Party (ADWP) of the EBMT set the guidelines for the application of AHSCT to ADs in phase I-2 clinical studies and advised that all cases treated should be registered in the EBMT database [19]. The aim was to discourage the proliferation of protocols resulting in anecdotal and uninterpretable results. The first retrospective report on 85 MS cases registered in the EBMT database was published by Fassas *et al.* in 2002 [20]. At that time, this retrospective study showed that the mortality of the procedure was 7.4%, while 74% of treated cases did not progress over the three years following HSCT. The EBMT has also promoted a prospective trial, named ASTIMS (Autologous Stem Cell Transplantation in Multiple Sclerosis), which compares AHSCT with mitoxantrone (www.astims.org). The ASTIMS trial started in April 2004.

In this study, we report an updated, retrospective, analysis carried out on the total number of MS cases now registered in the EBMT database, mainly focusing on transplantation safety and the evaluation of disease progression by the Kurtzke Expanded Disability Status Scale (EDSS).

Patients and methods

Patient characteristics

From June 1995, data on 183 patients with progressive MS, enrolled for the AHSCT procedure, have been reported to the EBMT ADWP registry from 45 centers in Europe, Russia, China and the US. In order to collect essential information on treatment schedule, toxicity and the clinical outcome of the treatment, a data collection form was created and sent to all the centers. Data on transplant characteristics and toxicity were obtained for all cases, whilst information on the neurological outcome was available in 145 cases. Due to a lack of standardized procedures and missing data, MRI information was not collected. Patient characteristics are shown in Table 1.

Table 1 Characteristics of the patients

No. of patients	183
Gender (F/M)	105/78
Age (years), median (range)	34 (15–58)
Type of MS	
Secondary progressive	99 (54%)
Primary progressive	32 (18%)
Relapsing progressive	19 (10%)
Relapsing remitting	22 (12%)
Unknown	11 (6%)
Disease duration (years), median (range)	6.7 (0.2–28.5)
Mobilized only	5
Transplanted	178
Evaluable for neurological outcome	145

Disease duration indicates the interval between the reported date of diagnosis and HSCT. Patients are considered evaluable for neurological outcome when the follow-up is greater than six months and adequate feedback was obtained from the consulting physicians.

Disability was severe, with a median EDSS before mobilization of 6.5 (range: 3.5–9). In 151 of 183 patients, data on previous treatments were available (Table 2): 27 cases had been treated with glucocorticosteroids only; 88 received, in addition to steroids, DMDs such as interferons, glatiramer acetate, and immunosuppressive drugs, such as azathioprine, methotrexate, CY, mitoxantrone, cladribine and others. Thirty-six cases were treated with DMDs, only 21 of which being IFNs.

After the mobilization procedure, five patients did not undergo AHSCT. In this retrospective analysis, we also evaluated, as a separate cohort, patients fulfilling the inclusion criteria used in the ASTIMS trial (ie, age ≤ 50 years, documented worsening of EDSS in the last year, failure of conventional therapies, EDSS 3.5–6.5 at the time of AHSCT, and secondary progressive (SP) or relapsing-remitting (RR) form of MS), and these cases are referred to as 'ASTIMS eligible' cases.

Transplant characteristics

A total of 169 evaluable patients underwent a mobilization procedure. Mobilization regimens

Table 2 Treatments prior to HSCT

Corticosteroids only	27 (18%)
Corticosteroids + up to 4 DMDs	88 (58%)
Corticosteroids + IFN only	20/88
DMDs alone	36 (24%)
IFN alone	21/36
Total	151

Disease modifying drugs (DMDs) are considered azathioprine, cyclophosphamide, glatiramer acetate, interferons, mitoxantrone, intravenous immunoglobulins, methotrexate, cladribine and others. IFN, interferon.

consisted of CY with G-CSF in the majority of patients (131/169; 78%); the CY dose varied from 1.5 to 4 g/m², with the most common dose of 4 g/m² given in 58 cases. The dose of G-CSF varied between 5 and 12 µg/kg per day. The other mobilization regimens used were G-CSF only ($n = 19$), CY with GM-CSF ($n = 6$) or alone ($n = 3$), and other chemotherapeutic agents with G-CSF ($n = 2$). In nine cases, data on mobilization regimens were not available. Patients receiving a bone marrow transplant ($n = 14$) were mostly not mobilized. The grafts were manipulated to remove immune cells in 97/178 transplanted cases (54%), in 77 of the cases, by positive selection of CD34+ cells; other purging methods were T cell-depletion by either negative immunomagnetic selection or by Campath-1H incubation; several grafts were purged by incubation with cytostatic drugs.

A total of 178 patients underwent AHSCT. The most common conditioning regimen was BEAM (carmustine, etoposide, cytosine-arabioside, melphalan), with or without antithymocyte globulin (ATG). Conditioning regimens are summarized in Table 3. ATG was utilized in 132 (74%) patients. In 62 (35%) cases, a particularly intensive immunosuppressive regimen was used with the association of ATG and *in vitro* manipulation (double purging).

Hematologic engraftment was defined as a polymorphonuclear cells (PMN) count $>0.5 \times 10^9/L$.

Outcome evaluation

Outcomes are reported based on the last follow-up of each patient. Neurological assessment consisted of measuring changes in the EDSS score for a minimum of six months of follow-up compared to baseline. The EDSS score was evaluated before mobilization (baseline) and six months after AHSCT, then every year until the last follow-up of each patient. Clinical improvement, confirmed at the subsequent evaluation, was defined as a decrease in the EDSS score of at least 0.5 points if the

score at baseline was >5 and by ≥ 1 EDSS points if EDSS score at baseline was ≤ 5 . Progression was defined as an increase of ≥ 1 EDSS points if EDSS score at baseline was ≤ 5 , and by 0.5 EDSS points if EDSS score at baseline was >5 , confirmed at the subsequent evaluation. The time of the first increase of EDSS was taken as the time of disease progression. The primary endpoint was the assessment of the safety of the procedure. Any adverse event related to the procedure was registered; such events were considered early or late when they occurred before or after 100 days from transplantation, respectively. Death was related to transplant procedure if it occurred within 100 days from transplant. If death occurred after this point, then the physician had to report in the collection form whether it could be related in any way to the procedure, to the progression of disease or another unrelated event. Confirmed progression-free survival (PFS) was the secondary endpoint of this study, consisting of the probability of being alive without clinical progression when compared to baseline.

Statistical analyses

SPSS ver. 12 (SPSS Inc. Chicago, IL, US) was utilized for all statistical analyses. Categorical data were compared using Fisher exact test and continuous variables were compared with Student's *t*-test. Regarding TRM, variables associated with outcome in univariate analysis at $P < 0.20$ were further tested in a multivariate binary logistic analysis with stepwise forward (or backward) selection.

Nonetheless, the limited number of outcomes ($n = 9$), in comparison with the amount of variables which would be logical to consider, weakens the statistical power of our multivariable analysis. The end points of survival analysis were either death or progression, respectively. Time variable was the interval between transplantation and either the event or the last examination, when no event occurred. The Kaplan–Meier method was used to compute the time to event curves and to estimate median values. Log rank test was used to compare curves in univariate models, significant variables were those included in Cox proportional hazard model.

Results

Mobilization

Mobilization data were available from 160 patients. Mobilization failure was registered in three patients, who were successfully re-mobilized with G-CSF, G-CSF and CY and CY plus vesepide, respectively.

Table 3 Conditioning regimens

Regimen	<i>n</i> (%)
BEAM + ATG	74 (41)
BEAM	30 (17)
BCNU + CY + ATG	20 (11)
TBI/CY + ATG	16 (9)
Bus + ATG	10 (6)
Others	19 (11)
Unknown	9 (5)

BEAM = BCNU (carmustine) = 300 mg/m²; ARAC (cytosine arabinoside) = 200 mg/m² per day; VP (etoposide) = 200 mg/m² per day; M (Melphalan) = 140 mg/m². ATG, anti-thymocyte globulin; Bus, busulphan; Cy, cyclophosphamide; Mel, melphalan.

Of 125 patients, evaluable for adverse events 25 (20%) patients were reported to have a mild toxicity during mobilization, mostly related to the immunosuppression. Six patients were reported to have transient neurological events.

Transplants and transplant toxicity

Five patients were mobilized only; the remaining 178 received a transplantation of PB-HSCs (163), BM-HSCs (14) or both (1). Such a distribution reflects the current trend to use PB-HSCs in an autologous setting. All 178 patients did engraft. Early non-neurological toxicity was reported in 80/144 (56%) evaluable patients, mostly associated with the immunosuppression and as expected for autologous HSCT in other settings. This early toxicity is detailed in Table 4. Neurological toxicity was reported in 26/149 (17%) evaluable patients; it occurred within 60 days from the transplant and was reported as transient. Late toxicity occurred in 9/160 evaluable patients (6%): mean time of onset from AHSCT was 20 months, ranging between four and 41. Three out of nine were Varicella Zoster Virus (VZV) infections; five patients (3.4%) showed non-neurological new ADs, such as autoimmune thyroiditis ($n=3$), and acquired anti-factor VIII inhibitors ($n=2$).

Mortality analysis

Kaplan–Meier analysis showed an overall survival of 91.2% at eight years (Figure 1). Out of 169 evaluable

Table 4 Early, non-neurological toxicity

Event	<i>n</i>	%
Immunosuppression related		
Neutropenic fever	23	
Sepsis	19	
CMV reactivation	8	
Urinary tract infection	5	
Gastroenteritis	5	
Pneumonia	2	
Generalized HSV infection	1	
Total	63	79
Others		
Engraftment syndrome	2	
Allergy to ATG	10	
Severe mucositis	3	
VOD	1	
TTP	1	
Total	17	21

Non-neurological toxicity recorded within 100 days from HSCT. It has been split into events related to the immunosuppression and to others. The last column reports the percentage of the two groups relative to the total number of reported events. VOD, veno-occlusive disease; TTP, thrombotic thrombocytopenic purpura; CMV, cytomegalovirus; HSV, herpes simplex-virus.

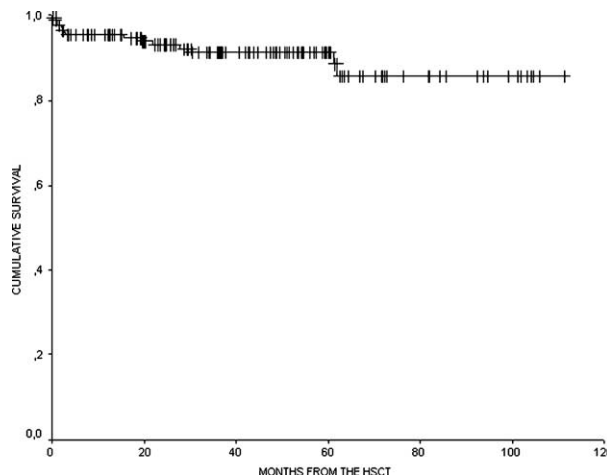


Figure 1 Overall survival curve. Kaplan–Meier curve of the cumulative overall survival. The thick marks represent censored events. The analysis was performed on the 178 patients who underwent the transplant.

patients, six died from progression of MS (4.7%). In these cases, the treating physician communicated that the neurological condition of the patients continued to deteriorate in spite of the therapy, and that the patients died after a period of 15–42 months due to the progression of the disease. Nine out of 169 evaluable patients died for reasons connected with the transplantation procedure, resulting in an overall 5.3% TRM (Table 5). All the deaths were recorded before the end of 2000, whilst no toxic deaths were reported among the 62 evaluable patients transplanted after this date.

Inclusion criteria for the ASTIMS trial were chosen according to a consensus amongst various experienced specialists. In particular, an age of ≤ 50 years, documented worsening of at least 1 point at EDSS in the last year, failure of conventional therapies, EDSS ≤ 6.5 and SP or RR form of MS, were the most relevant criteria in terms of clinical feasibility. We then, retrospectively, evaluated the population of 169 MS transplanted cases with and without the inclusion criteria for ASTIMS. TRM, among the 87 patients who did not fulfil ASTIMS inclusion criteria, was 6.9% compared to 3.6% in the patients who did fulfil the criteria. Moreover, if we consider the 53 patients who received the conditioning regimen used in the ASTIMS trial (BEAM followed by ATG without manipulation of the graft), no TRM was reported, irrespective of their clinical condition at the time of the transplantation.

Finally, a correlation analysis between TRM and items likely to be related to it was carried out: either busulphan or TBI including regimens, graft manipulation and the association of combined *in vivo*

Table 5 Transplant-related mortality

Patient	Days from HSCT	Conditioning	Purging	Cause of death
1	7	BEAM	Yes	Cardiac toxicity
2	26	BEAM+ATG	Yes	Bacterial infection, hemorrhage
3	586	BusCy+ATG	Yes	Pneumococcal septicemia, post-operative
4	20	BusCy+ATG	Yes	Influenza pneumonia
5	58	BEAM+ATG	Yes	Cerebral aspergillosis
6	63	Bus+Mel	Yes	Infection, neurologic deterioration
7	81	Fludarabine+ATG	Yes	Neurologic deterioration
8	841	Bus+ATG	No	Acquired Factor VIII inhibitor
9	30	BusCy+ATG	Yes	Infection, VOD

Descriptive report of deaths referred by the consulting specialists to the HSCT and statistical analysis of same (see Table 6). Statistical methodology is reported in the text. Abbreviations are reported in Table 3 legend.

and *in vitro* purging (double purging), age, MS subgroup, EDSS at the time of the transplant, disease duration more or less than 5 years before the HSCT were evaluated both in uni- and multivariate analysis. Results are reported in Table 6. Regimens including busulphan exerted a negative impact both in univariate ($P=0.001$) and multivariate analysis ($P=0.001$), respectively. It was noted that six of nine TRM events were associated with infections, and in five a double purging was performed (Table 5). However, this item did not result significantly associated with TRM either in univariate ($P=0.077$) or multivariate analysis ($P=0.2$). There was also a trend between age and TRM, which did not reach statistical significance. The low number of events might account for the low statistical power.

Neurological outcome

Neurological follow-up was available in 145 patients, but in two patients, the period of follow-up was <6 months, and thus were not considered. Manipulation of the graft (*ex vivo* purging) did not

show any advantages in this series of patients. The MS subgroup has been previously addressed as a major prognostic factor, with primary progressive (PP) forms more prone to progression [20]. In our analysis, only 19/142 (13%) evaluable patients were considered to have a PP form; no differences in the two groups were found, possibly due to the numerical imbalance.

Overall, the EDSS at follow up (median: 41.7 months) decreased or remained stable in 63% of cases, whilst in 37% of patients it worsened. Interestingly, we evaluated, as a separate cohort, the patients treated with the ASTIMS protocol (ie, BEAM followed by ATG without manipulation of the graft) and 75% of cases remained stable compared with 25% that worsened with time. If one considers only the 'ASTIMS eligible' cases treated with the ASTIMS protocol, the percentage of stable or improved patients remain almost the same (71 versus 29%). If 'ASTIMS eligible' cases are treated with a more intense regimen (double purging, *in vitro* and *in vivo*), this more aggressive treatment does not, apparently, add any advantage (62% better or stable versus 38% of worsened cases).

Age and interval prior to HSCT considered separately showed some impact on disease progression, although not statistically significant. We arbitrarily compared the cumulative PFS of patients <40 years and transplanted within five years from diagnosis versus older patients transplanted after five years (Figure 2). The difference between the two groups resulted statistically significant ($P=0.01$) and was independent, in multivariate analysis, from other relevant variables (MS subtype, conditioning intensity, double purging, EDSS at HSCT). While EDSS before and after AHSCT was available in 145 cases, the trend was also investigated, looking at the EDSS one year before transplantation compared with the years after. Data concerning this issue were available in 63 patients and showed a rapid worsening in the year before

Table 6 Transplant-related mortality analysis

Variables	Univariate (P)	Multivariate (P)
EDSS > 6.5	0.37	
BEAM+ATG	0.32	
Double purging	0.077	0.2
Primary progressive	0.37	
<i>Ex vivo</i> purging	0.08	
Busulphan	0.001	0.001
Age	0.068	0.082
Diagnosis-HSCT interval	0.23	

Descriptive report of deaths referred by the consulting specialists to the HSCT (Table 5) and statistical analysis of same (Table 6). Statistical methodology is reported in the text. Abbreviations are reported in Table 3 legend.

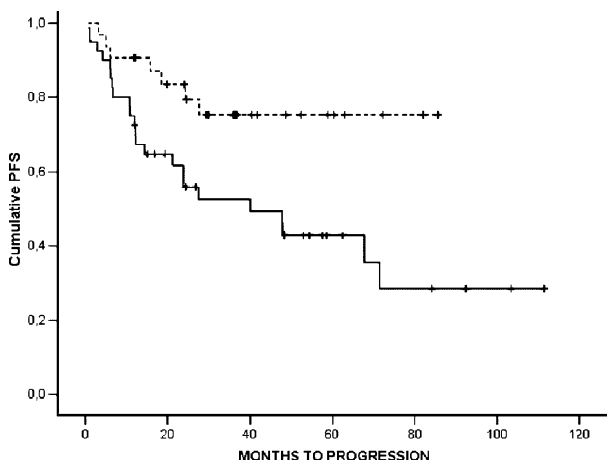


Figure 2 Progression-free survival according to the interval prior to HSCT and age. Cumulative progression-free survival plotted according to both the interval between diagnosis and HSCT and to the age of the patient at the time of HSCT. The dotted line represents the 32 patients <40 years of age, who were transplanted within five years from diagnosis. The solid line represents the 40 patients transplanted after such interval, and who were >40 years of age. The Kaplan–Meier method was used to plot the curves. The thick marks represent censored events. Statistical comparison of the two groups was significant ($P=0.01$).

transplantation and a substantial stabilization of EDSS progression in the subsequent time period (Figure 3).

Discussion

Since 1997, when Fassas *et al.* reported, for the first time, the use of therapeutic regimens aimed at

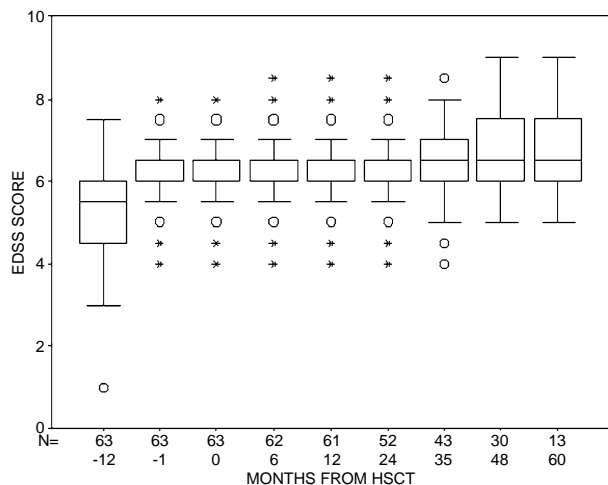


Figure 3 EDSS score before and after HSCT ($n=63$). Boxplot graphs of EDSS score one year before and at different time points after the transplant. Each box shows the median, quartiles, and extreme values within each time point. The two lines below the graph represent the number of cases and the months from the HSCT, respectively.

obtaining an intense immunosuppression, followed by marrow rescue by means of infusion of AHSCs, a number of severe MS cases have been treated with AHSCT in Europe, the US, Russia and China. The results of single center studies [12,13,21–23], and country multi-center co-ordinated studies [16,18], have since been published. From these studies, it appears that AHSCT is a promising therapeutic strategy in severe, rapidly worsening, MS cases, unresponsive to conventional therapies. AHSCT has an effect on inflammatory activity detected by MRI, with complete abrogation of gadolinium-enhancing areas [15,17]. It also has an impact on the clinical evolution of the disease, with a clinically relevant reduction of the frequency of relapses [22], and slowing down the progression of the disease, demonstrated in almost 70% of severe MS cases [18,20,24], at least in the first three years of follow-up. However, the treatment is associated with known toxicity, infections and side effects are common and, of major concern, there is an associated TRM. On the other hand, transplant-related toxicity is usually limited to the peri-transplant period in the vast majority of cases, and the patients may thereafter experience a prolonged period of neurological stability, free of treatment. Moreover, the toxicity can be controlled by a better selection of patients and also by the experience of the transplant center [25]. An approach to the use of allogeneic HSCT for MS is currently under investigation [26]. In 2002, the ADWP of EBMT published a retrospective analysis of the first 85 cases treated in Europe with AHSCT [20], showing that nearly 74% of MS-treated cases did not progress after three years, with a TRM of 7.4%. This mortality risk was considered high by many in the neurological community, even for rapidly worsening MS cases, who are at high risk of becoming wheelchair bound or bed-ridden in a short period of time. A significant reduction in the TRM, as well as the demonstration of durable clinical benefit, are considered important by neurologists for further consideration of AHSCT as a possible therapeutic option in severe MS cases. Transplant-related adverse events can be reduced by avoiding high intensity regimens, with their concomitant organ toxicity and profound, sustained immunosuppression. Selection of patients might also have an impact on both safety and clinical outcome, as more advanced patients and patients without signs of inflammation at MRI, are at risk of treatment failure.

A major aim of this work was to retrospectively evaluate these variables, specifically in MS patients. Despite the limitations of a retrospective analysis, such as, eg, missing data in some cases or lacking of information on the presence of relapses before and after therapy, the present study has had the unique

opportunity of examining the largest series of MS cases treated with AHSCT reported so far, with the aim of supplying evidence to provide an equipoise for designing prospective trials.

In this study, we confirmed that a more aggressive conditioning regimen, including busulphan, is statistically associated with a higher risk of mortality in the single, as well as the multivariate, analysis. In addition, an intense immunosuppression achieved through the association of graft manipulation and *in vivo* T-cell depletion was associated with a higher risk of TRM, although not statistically significant either in univariate ($P=0.077$) or in multivariate ($P=0.2$) analysis. Of great relevance is that the procedure utilized in the ASTIMS trial (BEAM followed by ATG, without any *in vitro* purging) was not associated with mortality: 53 MS cases have been treated with this regimen without any toxic deaths. These findings also confirm in MS a relationship between intensity of conditioning and TRM, as well as in other Ads [27]. While less intense, immunosuppressive-only regimens are possible, many such strategies over the past 30 years have failed to induce durable long-term remission once the immunosuppression is withdrawn. The recent work of Muraro *et al.* [6], suggests, for the first time, that an intense autologous HSCT (CY, ATG and TBI) regimen could induce durable long-term remission. The results presented here suggest that this may also be possible with a moderate regimen (and therefore less toxic, such as BEAM/ATG), with a durable benefit seen in some patients up to nine years post-HSCT.

Although there was a trend for an increased risk of mortality in more disabled patients, this item was not shown as statistically significant. However, in immobilized patients, the risk of infection is intuitively higher and, therefore, we strongly recommend that only ambulatory cases, ie, with an EDSS not higher than 6.5, should undergo this procedure. A possible exception to this statement are MS cases with a rapidly worsening clinical course, unresponsive to traditional therapies, or who have acquired a severe loss of strength in the lower limbs in a few months [28].

When the mortality was plotted against the year of treatment, it appeared that in the last three years (2001–2003) no deaths occurred among the 63 transplanted patients, compared to 1997–2000, when nine of 121 patients died for reasons related to the procedure. It might be speculated that, as well as in other hematological disorders, a better patient selection and transplant experience has led to such a finding. This 'learning curve' shows that, in selected MS cases, the *de novo* mortality risk has shown a trend that renders the procedure acceptable due to benefits that cannot be

obtained by currently available immunosuppressive therapies.

AHSCT was not associated with important neurological complications related to the procedure, despite the frequent occurrence of transient worsening during the neutropenic phase of the post-conditioning period, when fever occurred in most individuals.

Data on neurological outcome confirm the results of phase 1–2 studies, with a consistent number of patients showing an arrest of disability progression. When we examined all the evaluable patients (143 cases with a median follow-up of 41.7 months), the disease remained stable or improved in 63% of cases and worsened in 37%.

In principle, the inflammatory component of MS is the major target of such an intensive immunosuppressive treatment, considering also previously published data on the effect of AHSCT on MRI gadolinium-enhancing areas on cases registered in the present EBMT database, which demonstrate a complete suppression of the enhancing activity [15,17]. Taking into account that inflammation is mostly active in the initial phase of the disease, early treatment should be more effective. In this series, we showed that younger patients, transplanted within five years from diagnosis, show a significantly better progression-free survival, thus confirming the previous assumption and indicating that AHSCT could be considered if, after the failure of conventional treatments, the patients continue to progress rapidly. Progression in MS is most likely linked to inflammation in the early stages of disease, but can subsequently proceed independently due to degenerative mechanisms [29].

The place of autologous HSCT in the treatment of severe, treatment refractory MS needs to be established with prospective, randomized controlled trials, which are proceeding or in the planning stage.

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