Lenalidomide as salvage treatment for multiple myeloma relapsing after allogeneic hematopoietic stem cell transplantation: a report from the French Society of Bone Marrow and Cellular Therapy

Tereza Coman,[1] Emmanuel Bachy,[2] Mauricette Michallet,[2] Gérard Socié,[3] Madalina Uzunov,[4] Jean Henri Bourhis,[5] Simona Lapusan,[6] Alain Brebion,[7] Stéphane Vigouroux,[8] Sébastien Maury,[9] Sylvie François,[10] Anne Huynh,[11] Bruno Lioure,[12] Ibrahim Yakoub-Agha,[13] Olivier Hermine,[1,14] Noël Milpied,[8] Mohamad Mohty,[6, 15, 16] and Marie Thérèse Rubio[6,15,16]

[1]CNRS UMR, 8147, Hôpital Necker-Enfants Malades, Paris, France; [2]CHU Lyon Sud, Pierre Bénite; [3]Hôpital St Louis, Paris, France; [4]Hôpital Pitié Salpétrière, Paris Cedex 13, France; [5]Institut Gustave Roussy, Villejuif, France; [6]Hematologie Clinique et Therapie Cellulaire, Hopital Saint-Antoine, APHP, Paris, France; [7]CHU Fort de France, Fort-de-France Cedex; [8]CHU de Bx-Haut Lévêque University Bordeaux 2, Bordeaux, France; [9]Centre Hospitalier Henri Mondor, Créteil, France; [10]CHU Angers, Angers, France; [11]Hôpital Purpan, Toulouse cedex 9, France; [12]Hôpital Hautepierre, Strasbourg, France; [13]CHRU Lille, Lille, France; [14]Service d' Hématologie Adultes, Hôpital Necker-Enfants Malades, Paris, France; [15]Universite Pierre et Marie Curie, Paris, France; and [16]INSERM, UMRs 938, Paris, France

ABSTRACT

Optimal salvage treatment for multiple myeloma relapsing after allogeneic stem cell transplantation remains to be determined. Usually, such patients have been heavily pre-treated and present at relapse with a relatively refractory disease. Immunomodulatory properties of lenalidomide may be beneficial by facilitating a graft-*versus*-myeloma effect after allogeneic stem cell transplantation. However, the safety of such treatment is still under debate. We conducted a multicenter retrospective study and included 52 myeloma patients receiving lenalidomide alone or in combination with dexamethasone as salvage therapy after allogeneic stem cell transplantation. The first aim was to assess the efficacy and tolerance of this drug. The second aim was to evaluate its potential immunomodulatory effects evaluated on the occurrence of acute graft-*versus*-host disease under treatment. In this cohort, we show that lenalidomide can induce a high response rate of 83% (including 29% complete response). On lenalidomide therapy, 16 patients (31%) developed or exacerbated an acute graft-*versus*-host disease, which was the only factor significantly associated with an improved anti-myeloma response. Side effects were mostly reversible, whereas 2 deaths (4%) could be attributed to treatment toxicity and to graft-*versus*-host disease, respectively. With a median follow up of 16.3 months, the median overall and progression free survival were 30.5 and 18 months, respectively, independently of the occurrence of acute graft-*versus*-host disease under lenalidomide. Lenalidomide can induce high response rates in myeloma relapsing after allogeneic stem cell transplantation at least in part by triggering an allogeneic anti-myeloma response. Induced graft-*versus*-host disease has to be balanced against the potential benefit in terms of disease control. Further immunological studies would help us understand lenalidomide immunomodulatory activity *in vivo*.

Introduction

The use of new drugs such as proteasome inhibitors and immunomodulatory drugs (IMiDs) has dramatically improved the natural history of multiple myeloma (MM). Although new combination therapies can allow high complete remission rates, relapse still occurs in the majority of patients. Therefore, more intensive alternative therapies are discussed particularly for younger patients and those with poor risk factors.[1] In this context, allogeneic stem cell transplantation (allo-SCT) is an attractive treatment option, especially with the development of the so-called reduced toxicity conditioning regimens. However, despite achievement of a potent allogeneic graft-*versus*-myeloma (GVM) effect, definitive control of the disease remains rare because of frequent relapses.[2] Salvage treatment in this context has to deal with a refractory disease in patients who have usually received several prior treatment lines with cumulated toxicities, and who are receiving immunosuppressive drugs.[3] As the curative potential of allo-SCT relies on the GVM effect, donor lymphocyte infusion (DLI) has been commonly used as salvage therapy but has induced poor response rates and high morbidity due to concomitant development of graft-*versus*-host-disease (GVHD).[4] More recently, the use of thalidomide in combination with DLI as consolidation therapy proved to be able to increase the anti-MM response.[5] As salvage treatment after allo-SCT, thalidomide induced an overall response rate (ORR) of 29% without achieving complete responses (CR) in 31 patients.[6] The 2nd generation IMiD, lenalidomide represents another interesting candidate in this setting due to its well established efficacy in myeloma[7,8] and pharmacological properties including both tumoricidal and immunomodulatory activities.[9,10] At present, only few data are available on the safety and efficacy of lenalidomide given after allo-SCT for MM. Previous studies on small cohorts[5,11,12] showed the drug to have high efficacy with concerns about the risk of acute GVHD induction under lenalidomide treatment.[13]

We report here the results of a retrospective multicenter

 doi:10.3324/haematol.2012.069328
The online version of this article has a Supplementary Appendix. Manuscript received on May 6, 2012. Manuscript accepted on October 23, 2012.
Correspondence: marie-therese.rubio@sat.aphp.fr

series of 52 allografted MM patients who received lenalidomide for post-transplant relapse. We show that, in this setting, the use of lenalidomide is feasible and induces high response rates, at least in part due to its immunomodulatory effects.

Design and Methods

Study design, inclusion criteria, data collection

This was a retrospective study conducted in 13 different French centers collaborating with the French Society of Bone Marrow and Cellular Therapy (SFGM-TC). This study was approved by the scientific board of the SFGM-TC and performed according to institutional guidelines in accordance with the principles of the Declaration of Helsinki. Centers were requested to report: i) adult MM patients; (ii) relapsing after allo-SCT; iii) treated by lenalidomide at relapse; iv) lenalidomide alone or in association with dexamethasone; (v) regardless of the number of prior salvage treatment lines. We excluded patients who had received lenalidomide in maintenance or consolidation after allo-SCT or in combination with other molecules other than dexamethasone. Fifty-two patients treated with lenalidomide between 2006 and 2009 met these eligibility criteria. All clinical events occurring before and after allo-SCT were carefully assessed through direct review of report forms and medical charts by the first author (TC). For each patient, we collected detailed demographic data, diagnostic criteria, allo-SCT characteristics, and treatment lines administered before and after allo-SCT. In order to use an uniform definition of the number of lines of therapy, we adapted the IMWG criteria to the retrospective setting and considered that all planned treatment sequences constituted a unique treatment line.[14] Abnormal cytogenetics, mostly explored by fluorescence *in situ* hybridization (FISH), were defined by the presence of del(13), del (17p) and t (4; 14). Assessment of lenalidomide treatment modalities included dosage, administration schedule, duration of treatment and its association with dexamethasone.

Efficacy and safety assessments

Criteria used to evaluate response to treatment were adapted from those of the IMWG consensus,[14] especially for CR that could be considered in some rare cases without available immunofixation on the basis of two consecutive normal electrophoreses. Therefore, partial response (PR) was defined by a reduction of more than 50% of serum myeloma protein, and CR required a negative serum immunofixation or at least the disappearance of serum myeloma protein in two consecutives serum electrophoreses. In cases of no measurable serum protein at diagnosis, PR required a reduction of over 50% of Bence-Jones protein or urine proteins or free light chain and CR a disappearance of urine protein, a negative urine immunofixation or a normal serum ratio of kappa/lambda light chains. Very good partial response (VGPR) was defined as a reduction of more than 90% of the measurable parameter. Stable disease (SD) was defined as a reduction of less than 50% and progressive disease (PD) as an increase of at least 25% of the measurable parameter. Overall response rate (ORR) included CR, VGPR and PR. Evaluation of the response was based on the measurement of the abnormal protein before each lenalidomide cycle. All adverse events were assessed according to available clinical and biological data and were graded according to the CTCAE criteria (version 3.0). Lenalidomide dose was generally adjusted according to the practice of each center based on patient's clinical and biological tolerance, and GVHD symptoms. Thromboembolic prevention administered during lenalidomide treatment was captured and correlation with thromboembolic events was assessed.

Immunomodulatory effects

We focused especially on clinical and pathological evidence of acute and chronic GVHD occurring before, after and during lenalidomide treatment. We assessed the timing of immunosuppressive (IS) therapy withdrawal before lenalidomide initiation and considered a 'close' withdrawal when IS was discontinued three months or less prior to lenalidomide initiation. Acute and chronic GVHD were evaluated according to standard criteria.[15] *De novo* acute GVHD on lenalidomide treatment was defined as appearance of new symptoms and/or pathological evidence of acute GVHD while on treatment. Acute GVHD on lenalidomide included *de novo* acute GVHD as defined above and cases of rapidly exacerbated GVHD symptoms during early phase of lenalidomide treatment for patients still presenting acute GVHD symptoms at lenalidomide introduction. *De novo* chronic GVHD after lenalidomide treatment was defined as symptoms of chronic GVHD at the date of last follow up for patients who had not experienced chronic GVHD before lenalidomide introduction.

Study end points and statistical analyses

The primary end point of this analysis was to assess the efficacy and tolerance of lenalidomide used for myeloma relapsing after allo-SCT. Secondary end points included incidence and features of GVHD in order to highlight a possible immunomodulatory effect of the molecule. Correlations between patients' or disease characteristics and outcomes were assessed using χ^2 or Fisher's exact test when appropriate. When continuous parameters were analyzed, a non-parametric Mann-Whitney's test was used. To evaluate the impact of acute GVHD on the response to treatment, a Cox's model with GVHD considered as a time-dependent variable was used. Progression free survival (PFS) was estimated from lenalidomide introduction to the date of the first assessment showing disease progression, relapse or death during treatment. Patients who were alive or discontinued lenalidomide without evidence of disease progression were censored at time of last evaluation for PFS. Overall survival (OS) was calculated from time of lenalidomide introduction until death from any cause or censored at last follow up. Time to event analysis was assessed using the Kaplan-Meyer method and statistical difference between survival distributions was evaluated with the log rank test. All tests were two-sided and $P<0.05$ was considered statistically significant. Analyses were performed using SAS (SAS Institute Inc., Cary, NC, USA) versions 9.0 and 9.2.

Results

Patients' characteristics and lenalidomide treatment modalities

Patients' and allo-SCT characteristics are summarized in Table 1. The majority of MM (57%) presented with abnormal cytogenetics at diagnosis. Patients had received a median of two treatment lines (range 1-5) before allo-SCT, including thalidomide and lenalidomide in 50% and 10% of cases, respectively. Almost all patients (94%) had received an autologous SCT (auto-SCT) before allo-SCT and 13% a double auto-SCT. Allo-SCT was performed after a median of 32 months from MM diagnosis (range 8-93) mainly using a so-called reduced intensity conditioning regimen (85%) and G-CSF mobilized peripheral blood stem cells (PBSC) (86%) from an HLA-matched sibling donor (79%). Patients relapsed within a median of 11 months after allo-SCT (range 1.3-79).

Lenalidomide treatment was initiated at a median of 24

Table 1. Patients' characteristics at baseline and at allo-SCT.

	N. (%) or median (range)
Characteristics at diagnosis	**52**
Age, years	48 (32-61)
Sex	
Male	34 (65)
Female	18 (35)
Type of monoclonal component	
IgG	27 (52)
IgA	11 (21)
IgM	1 (2)
Light chain	13 (25)
Abnormal cytogenetics*	21 (57)
del (13)	19 (51)
t(4 ;14)	6 (16)
del (17p)	1 (3)
Auto-transplant before allo-SCT	
None	3 (6)
Auto SCT	49 (94)
Double auto SCT	13 (25)
Other treatments before allo SCT	
Thalidomide before allo-SCT	26 (50)
Lenalidomide before allo-SCT	5 (10)
Allograft features	
Disease status at allograft†	
CR	12 (23)
VGPR	11 (22)
PR	22 (43)
SD	1 (2)
PD	5 (10)
Conditioning regimen	
RIC	44 (85)
Myeloablative	8 (15)
Tandem auto/allograft	21 (40)
In vivo T-cell depletion	
No	29 (56)
Yes	23 (44)
Donor	
Matched related	40 (77)
Matched unrelated	9 (17)
Mismatched	3 (6)
Graft type	
PBSC	45 (87)
BM	7 (13)
GVHD	
Acute	30 (58)
Late acute	11 (21)
Chronic GVHD	20 (38)
Time to relapse after allo-SCT (months)	11 (1-79)
Salvage therapies after allo-SCT before lenalinomide initiation	
N. of salvage lines after allo-SCT and before lenalidomide	1 (0-6)
0	23 (44)
1	20 (38)
2	6
3	2
≥4	1
Type of salvage therapy before lenalidomide	
Thalidomide	11 (21)
Bortezomib	18 (35)
DLI	10 (19)

*ISS: International Staging system; PBSC: peripheral blood stem cell; BM: bone marrow; CR: complete remission; VGPR: very good partial remission; RIC: riduced intensity conditioning; PR: partial remission; SD: stable disease; allo-SCT: allogeneic stem cell transplantation; GVHD: graft-*versus*-host disease; DLI: donor lymphocyte infusion; *Data were missing for 15 patients; †Data was missing for 1 patient.*

months (range 1-97) after allo-SCT and was used as first salvage treatment line in 23 patients (44%) (Table 2). Lenalidomide was mainly started at the classical dose of 25 mg/day during 21 consecutive days of a 28-day cycle (79%). Dexamethasone was administered in 40 patients (77%) using various schedules (40 mg, n=23; or 20 mg, n=5), weekly. Of note, lenalidomide was introduced earlier after allo-SCT in patients who did not receive dexamethasone (median 7.4 months, range 2.4-84.6) as compared to those who received dexamethasone (median 28.1 months, range1.3-96.5) P=0.022). Other salvage treatments after allo-SCT included thalidomide (21%), bortezomib (35%), while 19% of patients received DLI. Altogether, 73% of the patients included in this study had received at least one IMiD before beginning or reintroducing lenalidomide and 45% of them were refractory (less than PR) to IMiDs. Response rate to salvage treatment with thalidomide and bortezomib after allo-SCT were 35% and 72%, respectively. Of note, only 10% of patients responded (≥ PR) to DLI. Venous thromboembolism prophylaxis was given in 22 patients (42%) and was based mostly on aspirin.

Patients received a median of 6 cycles of lenalidomide (range 0.2-23) and 11 patients (21%) were still under treatment at last follow up.

Response to lenalidomide and factors influencing response

In this heavily pre-treated cohort of patients, the use of lenalidomide in relapsing patients after allo-SCT was associated with an ORR of 83% including 29% of CR, 23% of

Table 2. Features of lenalidomide salvage therapy.

	N. (%) or median (range)
Age at lenalidomide initiation, years	52 (37-68)
Lenalidomide administration	
25 mg/day	41 (79)
others	11 (21)
Corticosteroid association	
No	12 (23)
Yes	40 (77)
Thromboembolic prophylaxis	
None	30 (58)
Aspirin	16 (31)
LMWH/ VKA	6 (11)
Median of lenalidomide duration, months	6.3 (0.2-39.6)
Number of cycles	6 (0.2*-23)
<1	1 (2)
1	3 (6)
2	4 (8)
3	8 (15)
4	1 (2)
5	5 (10)
6	8 (15)
≥7	22 (42)
Cause of lenalidomide withdrawal	
Planned	14 (34)
Progression	12 (29)
Toxicity	9 (22)
GVHD on lenalidomide	6 (15)

**7 days. GVHD: graft-*versus*-host disease; LMWH: low molecular weight heparin; VKA: vitamin K antagonist.*

VGPR and 31% of PR (Table 3). The optimal response was obtained after a median of 3 cycles (range 0.2-11). Factors influencing response to lenalidomide were analyzed (Table 3). Previous refractoriness to an IMiD, abnormal cytogenetics, *in vivo* T-cell depletion, the use of lenalidomide as first or further salvage line post-allo-SCT and its association with dexamethasone did not influence the ORR. The only factor significantly associated with ORR was the occurrence of acute GVHD symptoms while under lenalidomide treatment (Hazard ratio (HR) = 2.33, 95%CI: 1.09-4.95, *P*=0.03) considering GVHD occurrence as a time-dependent variable in a Cox's proportional hazard model. Moreover, the response was also faster for the patients developing acute GVHD under lenalidomide since they reached the best response in a median of 62 days (range 7-316) after the introduction of lenalidomide as compared to 138 days (range 9-356) for those who did not develop acute GVHD (*P*=0.018).

With a median follow up of 16.3 months (range 3.7-49.6), the median PFS and OS were 18.0 and 30.5 months, respectively (Figure 1A and B). We analyzed factors influencing OS and PFS (Table 5). Cytogenetics (including del(13), del(17p) and t(4;14)) was the only factor significantly influencing PFS (Hazard ratio (HR) = 2.5, 95%CI: 1.0-6.6, *P*=0.04) (Figure 1C) and marginally OS (HR = 3.6, 95%CI: 0.7-17.6, *P*=0.09) (Figure 1D). Because of limited number of patients presenting del(17p) or t(4;14), we could not analyze the individual prognostic value of those cytogenetic abnormalities. However, we observed a trend toward a shorter PFS (median PFS of 9 months *vs.* not achieved, *P*=0.053) and a shorter OS (24-month OS of 56% *vs.* 69%, *P*=0.069) for patients with isolated del(13) (n=14) as compared to those without any of the analyzed cytogenetic abnormalities (n=16) (*data not shown*). Lenalidomide as first salvage treatment, its combination with dexamethasone, refractoriness to a previous IMiD, did not have any influence on PFS and OS. The majority of deaths were directly attributed to disease progression (n=9.7%, i.e. 75% of all deaths). Despite a beneficial impact of the occurrence of acute GVHD on the ORR, this did not translate into a benefit in terms of PFS (*P*=0.94) or OS (*P*=0.45).

Tolerance

Most patients (82%) experienced at least one adverse event and 47% of them at least one grade 3-4 toxicity

Table 3. Response rates to lenalidomide after allo-SCT and assessment of influencing factors.

	Pts (n)	Response (%)				Non-response (%)		*P**
		ORR (%) (%)	CR (%)	VGPR (%)	PR (%)	SD (%)	PD	
Corticosteroids associated to lenalidomide								ns
Without corticosteroids	12	75	33	17	25	8	17	
With corticosteroids	40	85	28	25	32	8	7	
Salvage line								ns
Lenalidomide at first salvage line	23	83	35	22	26	13	4	
Lenalidomide at a subsequent salvage line	29	83	24	24	35	3	14	
Cytogenetics at diagnosis								ns
Abnormal cytogenetics	21	81	33	24	24	5	14	
Normal cytogenetics	16	82	12	25	44	13	6	
Stop of IS within100 days before lenalidomide initiation**								ns
No	35	86	26	23	37	6	8	
Yes	15	73	33	27	13	14	13	
Prior exposure to IMiDs								ns
No	14	86	43	14	29	14	0	
Yes	38	82	24	26	32	5	13	
Prior refractoriness to IMIDs†								ns
No	19	84	26	26	32	5	11	
Yes	16	81	19	25	37	6	13	
Exacerbation or *de novo* acute GVHD on lenalidomide^								**0.027**
No	35	80	26	23	31	9	11	
Yes	16	94	38	25	31	6	0	
***In vivo* T-cell depletion**								**ns**
No	**29**	**86**	**28**	**34**	**24**	**4**	**10**	
Yes	**23**	**78**	**30**	**9**	**39**	**13**	**9**	
All patients	**52**	**83**	**29**	**23**	**31**	**8**	**10**	

*ORR: overall response rate; IMiD: immunomodulatory drug (includes thalidomide and lenalidomide); GVHD: graft-*versus*-host disease; CR: complete remission; VGPR: very good partial remission; PR: partial remission; SD: stable disease; IS: immunosuppressive therapy.* *P *values for response* vs. *non-response according to parameters were calculated using Fisher's exact test for all comparisons except for the effect of acute GVHD incidence where a Cox's model with time-dependent covariate was performed. °Data were missing for 2 patients (1 in CR and 1 in PR); †Data were missing for 4 patients among the 35 patients who received a prior IMiD. ^Data were missing for 1 patient with PD; ** includes t(4;14), del 17p, del 13. Data were missing for 15 patients.*

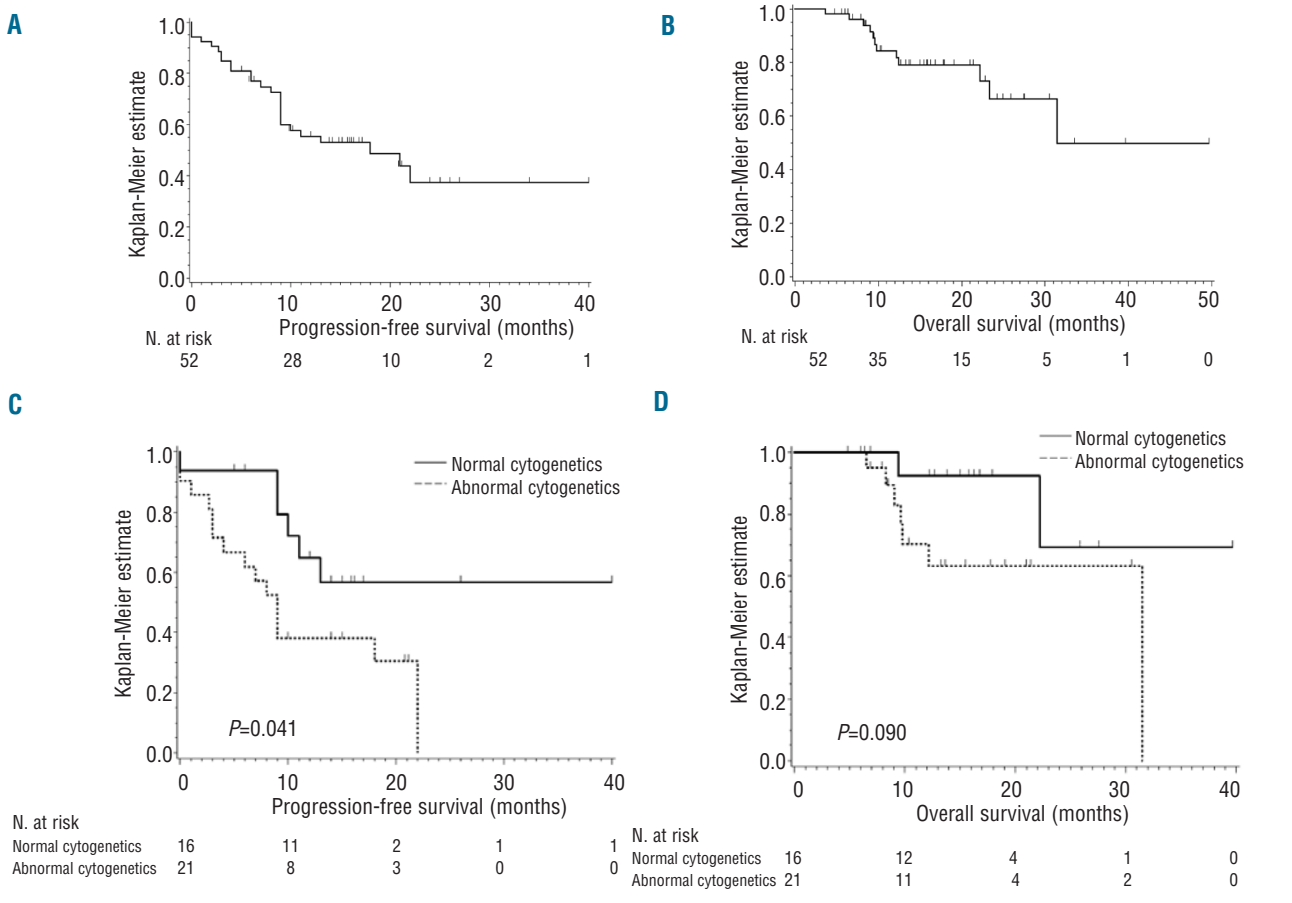

Figure 1. Evolution of patients after lenalidomide treatment. (A and B) Progression-free survival (PFS) and Overall survival (OS) of the whole cohort. (C and D) PFS and OS according to cytogenetics status in lenalidomide salvaged patients after allo-SCT failure.

(Online Supplementary Figure S1). Adverse hematologic events were recorded in 21 (41%) patients. Severe neutropenia was observed in 35% of patients (83% grade 3-4) and severe thrombopenia in 21% of patients (63% grade 3-4). Non-hematologic adverse events occurred in 70% of patients. Infectious episodes were the most common extra-hematologic adverse events (43% of patients; grade 3-4 in 55% of cases). However, only 4 cases (18%) of infectious complications occurred during neutropenic periods. We did not find any correlation between the occurrence of bacterial and viral infections and the administration of dexamethasone (P=0.99). Furthermore, the development of acute GVHD while under lenalidomide treatment was not associated with an increased risk of infections using a time-dependent model (P=0.61). Thromboembolic events were recorded in 7 (14%) patients and were correlated with the absence of prophylaxis (P=0.015) and with the association to dexamethasone (P=0.02). Other commonly described extra-hematologic toxicities involved peripheral neuropathy (18%), gastrointestinal tract disturbances (16%) and neuropsychiatric disorders (10%). Overall, 16 patients (44%) required dose adaptation and 9 (17%) discontinued lenalidomide because of toxicity. However, most adverse events were reversible (94%).

Two deaths (4%) were attributed to treatment toxicity. One patient with controlled MM presented an acute pulmonary infection in a non-neutropenic period while still on lenalidomide treatment (10 mg/day and dexamethasone 40 mg/week). The second patient died of septic shock following the introduction of immunosuppressive drugs for *de novo* acute GVHD occurring under lenalidomide therapy. No secondary malignancies have been reported with a median follow up of 16.3 months.

GVHD occurrence under lenalidomide treatment

In this cohort, the development of GVHD after allo-SCT was assessable in all patients (Table 4). At initiation of lenalidomide, 3 patients (6%) presented with symptoms of acute GVHD and 13 patients (26%) of chronic GVHD. During lenalidomide therapy, 13 patients (25%) experienced *de novo* acute GVHD, including 3 grade 3-4, after a median delay of 31 days (range 5-300) from the start of treatment. Additionally, the 3 patients with acute GVHD signs at lenalidomide introduction presented an exacerbation to grade 3-4 acute GVHD after a median of 19 days of lenalidomide treatment. The probability of acute GVHD development under lenalidomide was significantly higher for patients who had received lenalidomide as first salvage treatment after allo-SCT (P<0.001), those who discontinued immunosuppressive drugs within the 100 days before lenalidomide initiation (P=0.009), and patients presenting

Table 4. Occurrence of acute GVHD on lenalidomide treatment and assessment of influencing factors.

	Patients	Acute GVHD on lenalidomide (%)			No acute GVHD on lenalidomide	Univariate analysis	Multivariate analysis	
	N.	Grade 1/2	Grade 3/4	All	(%)	P*	OR (95% CI)	P§
Acute GVHD before lenalidomide initiation						ns		
No	22	27	14	41	59			
Yes	29	14	10	24	76			
GVHD at time of lenalidomide initiation						0.027		0.010
No	34	12	9	21	79		1	
Yes (acute or chronic)∞	17	35	18	53	47		13.5 (1.8-99.0)	
Stop of IS within100 days before lenalidomide initiation†						0.009		ns
No	34	14	6	20	77		1	
Yes	15	33	27	60	40		5.7 (0.9-35.8)	
Salvage line						<0.001		0.012
Lenalidomide at first salvage line	23	30	26	57	43		11.5 (1.7-78.2)	
Lenalidomide at a subsequent salvage line	28	11	0	11	89		1	
Corticosteroids associated to lenalidomide						ns		
Without corticosteroids	12	33	17	50	50			
With corticosteroids	39	15	10	26	74			
***In vivo* T-cell depletion**						**ns**		
No	**28**	**11**	**18**	**29**	**71**			
Yes	**23**	**31**	**4**	**35**	**65**			
All patients^Δ	**51**	**20**	**12**	**31**	**69**			

*P *values are associated with the risk of* de novo *GVHD occurrence on lenalidomide (whatever the type or stage of GVHD) and were calculated using Fisher's exact test; ∞3 patients had acute active and 13 chronic GVHD at time of lenalidomide initiation; †Data were missing for 2 patients without sign of aGVHD. §Data on* de novo *GVHD were missing for 1 patient. §Using a multivariate logistical regression model (only significant covariates were incorporated into the model).*

any signs of GVHD at lenalidomide introduction (*P*=0.027). By contrast, occurrence of acute GVHD was not influenced by the use of in *vivo* T-cell depletion within the conditioning regimen of the allograft performed a median two years earlier (*P*=0.634). In multivariate analysis, there was a significant correlation between the occurrence of acute GVHD on lenalidomide and the presence of any signs of GVHD at time of lenalidomide introduction or with lenalidomide introduction at first salvage line after allo-SCT. Although, the patients who received dexamethasone in association with lenalidomide developed relatively less acute GVHD than those who did not (26% *vs.* 50%, respectively), the difference was not statistically significant (*P*=0.157) (Table 5). As patients without dexamethasone also had an earlier lenalidomide introduction, we further assessed the role of these two parameters on the occurrence of acute GVHD. First, we analyzed the effect of dexamethasone association separately in patients treated with lenalidomide in a first-line setting after allo-SCT or in subsequent lines of treatment. We found that in both situations there was no significant benefit of the addition of dexamethasone to lenalidomide in terms of the risk of further development of acute GVHD (*P*=0.202 and *P*=0.964, respectively). Furthermore, to address the same question in a different way, we used a multivariate logistical regression model in which the effect of dexamethasone was adjusted for the line of treatment, either as a dichotomous parameter (1st *vs.* subsequent) or as an ordinal parameter. In none of these two different models was the use of dexamethasone associated with a reduction in the risk of acute GVHD development (*P*=0.410, OR=0.52, 95%CI: 0.11-2.42 and *P*=0.198, OR=0.37, 95%CI: 0.018-1.66, respectively). On the contrary, the line of treatment significantly influenced acute GVHD occurrence (*P*=0.002, OR=9.97, 95%CI: 2.24-42.6 and *P*=0.006, OR=0.26, 95%CI: 0.10-0.69, respectively). Altogether, these analyses demonstrate that the trend toward a lower risk of GVHD (50% *vs.* 26%) is due to the line of treatment that is, therefore, a confounding factor.

Management of acute GVHD required modification of lenalidomide administration in 13 patients by either dose adaptation (n=3) or transient (n=2) or definitive early (within the first month, n=6) or later (n=2) withdrawal. In addition, 9 of these patients received a standard systemic immunosuppressive therapy. The other 3 patients could continue on the same dose of lenalidomide, 2 of them were then treated by systemic immunosupression, while detailed GVHD treatment was missing in the third. None of them developed steroid-refractory GVHD and all cases were controlled at last follow up; but one patient died of sepsis shortly after GVHD onset. Overall, patients who experienced acute GVHD received a median of 4.5 cycles (range 0.2-10) of lenalidomide as compared to 6 cycles (range 2-23) for those who did not develop acute GVHD (*P*=0.013).

At last follow up, 11 patients presented chronic GVHD symptoms. Among those, 6 cases were *de novo* chronic GVHD, and all were localized to the skin (2 extensive and 4 limited forms) and controlled by adapted immunosuppressive treatment without adaptation of lenalidomide treatment. Actually, chronic GVHD never justified stopping lenalidomide.

Discussion

This series assessing the efficacy and tolerance of lenalidomide as salvage treatment in 52 MM patients

relapsing after allo-SCT is remarkable by the proportion of heavily pre-treated patients and the presence of abnormal and/or unfavorable cytogenetics. In this setting we could show an ORR to lenalidomide of 83% including 29% of CR, 23% of VGPR, and 31% of PR. Our results confirm those reported by previous reports on smaller series of patients (12-24 patients) that described a high efficacy of the drug as salvage therapy after allo-SCT in association or not with dexamethasone[5,11] or DLI.[12] These data also suggest a higher efficacy of lenalidomide in comparison to thalidomide (ORR 29%)[6] and at least a comparable efficacy to that of bortezomib (ORR 61-73%).[16-18] With a median follow up of 16.3 months, the median PFS was 18 months and the median OS was 30.5 months; results that appear superior to those reported with thalidomide (median OS of 12 months)[6] and bortezomib (median PFS of 6 months)[16,17] within comparable settings.

In our cohort, the only significant factor influencing PFS and OS was abnormal cytogenetics. However, when analyzed apart, isolated del(13) had only a tendency toward a lower PFS and OS, suggesting that the worse prognostic of abnormal cytogenetics was mainly carried out by the few cases of t(4;14) or del(17p) in our series, as previously published.[19,20]

As allo-SCT patients are prone to high morbidity, salvage treatment tolerance is of particular importance. In this series, we observed the commonly described adverse hematologic and infectious side effects of lenalidomide[5,7,8] leading to dose adaptation and withdrawal of the treatment in 44% and 17% of the patients, respectively. Thromboembolic events were slightly more frequent than previously reported (14%), probably because of frequent lack of prophylaxis (42%) and the association to dexamethasone.[21] Thus, according to recent publications,[22] we would recommend preventing thromboembolic events by using aspirin or low molecular weight heparin in association with lenalidomide treatment in the post allo-SCT setting.

The risk of developing acute GVHD after lenalidomide treatment after allo-SCT has been recently pointed out by the HOVON group.[13] While investigating lenalidomide as maintenance therapy with an introduction in the first three months post allo-SCT, they reported 43% of premature exit from the study because of occurrence of *de novo* grade 2 and above acute GVHD. In our cohort, *de novo* or exacerbation of pre-existing acute GVHD occurred in 31% of cases and was correlated to the introduction of lenalidomide early on after allo-SCT. On the contrary, the addition of dexamethasone to lenalidomide did not seem to influence significantly the occurrence of acute GVHD as demonstrated by two different statistical analyses. We also did not observe any impact of the use of *in vivo* T-cell depletion at transplant on acute GVHD occurrence under lenalidomide. This might be explained by the achievement of T-cell recovery by the time of lenalidomide introduction since the median time for starting lenalidomide was 24 months after allo-SCT. Importantly, all acute GVHD cases occurring under lenalidomide could be controlled, although one patient died of sepsis early after GVHD onset.

In univariate analysis, the occurrence of acute GVHD on treatment was the only factor influencing the response to lenalidomide, which reflects the potential of lenalidomide to enhance the immunological GVM effect. This is in line with the hypothesis of Kneppers *et al.* who found a beneficial impact on the time to progression after MM salvage therapy by lenalidomide in the group of patients who had been previously allografted as compared to those who did not.[23] In our cohort, however, the benefit in terms of response after developing acute GVHD under lenalidomide did not translate into an improved PFS or OS and thus raises the question of the relative benefit of the GVM *versus* cytotoxic anti-myeloma effects of the drug in allografted patients. A shorter duration of lenalidomide therapy in case of GVHD occurrence might explain in part the absence of improved PFS.

Another explanation would be that the GVHD-related GVM effect has a limited impact on the long-term disease control. Indeed, on the one hand, patients who stopped lenalidomide in the first month after the onset of acute GVHD and who most likely benefited essentially from the GVM effect (n=6) had a similar PFS as those who did not experience acute GVHD but had received a prolonged treatment, who mainly benefited from the cytotoxic anti-myeloma effect of lenalidomide (n=35), arguing in favor of a strong immunomodulatory effect. On the other hand, these later also had a similar PFS as patients who could continue lenalidomide despite the occurrence of acute GVHD and who may have benefited from both effects of the drug (n=10), suggesting that the duration of treatment may compensate the absence of acute GVHD immunomodulatory benefit (*data not shown*). Moreover, among the entire cohort, a prolonged treatment seemed protective since only 26% of the patients who had maintained lenalidomide after achieving the best response relapsed as compared to 56% of those who stopped

Table 5. Survival rates after lenalidomide introduction and assessment of influencing factors.

	OS HR (95% CI)	PFS *P**	HR (95% CI)	*P**
Corticosteroids associated to lenalidomide		ns		ns
Without corticosteroids	1		1	
With corticosteroids	0.6 (0.1-2.6)		1.2 (0.4-3.2)	
Salvage line		ns		ns
Lenalidomide at first salvage line	1.6 (0.5-5.0)		1.0 (0.4-2.3)	
Lenalidomide at a subsequent salvage line	1		1	
Cytogenetics at diagnosis		ns		0.04
Abnormal cytogenetics **	3.6 (0.7-17.6)		2.5 (1.0-6.6)	
Normal cytogenetics	1		1	
Stop of IS within100 days before lenalidomide initiation		ns		ns
No	1		1	
Yes	1.9 (0.5-6.8)		1.5 (0.6-3.4)	
Prior refractoriness to IMiDs		ns		ns
No	1		1	
Yes	0.5 (0.1-2.2)		0.9 (0.3-2.1)	
Exacerbation or *de novo* acute GVHD on lenalidomide		ns		ns
No	1		1	
Yes	2.4 (0.7-7.6)		1.8 (0.7-4.2)	

*OS: overall survival; PFS: progression-free survival; IMiD: immunomodulatory drug (includes thalidomide and lenalidomide); ns: not significant. *P values to compare survival distributions according to parameters were calculated using log rank test for all comparisons except for the effect of acute GVHD incidence where a Cox's model with time-dependent covariate was performed. **Includes t(4;14), del 17p, del 13.*

immediately after achieving the best response (*data not shown*). Far from any firm conclusions, these observations argue in favor of a prolonged treatment to maintain the response either by maintaining the GVM effect and/or inducing a prolonged cytotoxic anti-myeloma effect.

It has been previously reported in small series of patients that lenalidomide can modulate the activation and reconstitution of NK and T conventional and regulatory cells.[5,11] Analyzed on 12 patients, we observed stable $CD4^+$ T lymphocytes (P=0.157 in the delta of increase) but a relative increase in the $CD8^+$ T-cell counts (P=0.03) after the administration of lenalidomide (*data not shown*).

These results invite a further investigation into the *in vivo* immunomudulatory mechanism of action of lenalidomide.

In conclusion, lenalidomide is an effective salvage treatment after allo-SCT for MM patients and provides prolonged PFS and OS. The efficacy of lenalidomide in the post allo-SCT setting is likely related both to its immunomudulatory effects and to the intrinsic anti-MM activity of the molecule. The risk of acute GVHD induction should be taken into account, particularly when lenalidomide is introduced without steroids in the first months after allo-SCT and/or closely to the withdrawal of immunosuppression. However, by contrast with the maintenance setting in which the risk of acute GVHD may outweigh the theoretical benefit, we believe that in a salvage setting, the toxicity of the drug may be outweighed by the potential benefit in terms of PFS and OS. These promising results provide a rationale for a prospective phase I-II dose escalating study introducing lenalidomide at lower doses (from 5 mg/day) with or without dexamethasone and in association with aspirin in uncontrolled or progressive myeloma by at least four months post-allo-SCT with an adapted schedule of immunosupression withdrawal. By incorporating detailed immunomonitoring, such a trial could help determine whether the mechanisms underlying the improved anti-myeloma response in this setting are immune and/or drug-driven.

Authorship and Disclosures

Information on authorship, contributions, and financial & other disclosures was provided by the authors and is available with the online version of this article at www.haematologica.org.

References

1. Gahrton G. Progress in allogeneic transplantation for multiple myeloma. Eur J Haematol. 2010;85(4):279-89.
2. Bensinger WI. The current status of reduced-intensity allogeneic hematopoietic stem cell transplantation for multiple myeloma. Leukemia. 2006;20(10):1683-9.
3. Maloney DG, Molina AJ, Sahebi F, Stockerl-Goldstein KE, Sandmaier BM, Bensinger W, et al. Allografting with nonmyeloablative conditioning following cytoreductive autografts for the treatment of patients with multiple myeloma. Blood. 2003;102(9): 3447-54.
4. van de Donk NW, Kroger N, Hegenbart U, Corradini P, San Miguel JF, Goldschmidt H, et al. Remarkable activity of novel agents bortezomib and thalidomide in patients not responding to donor lymphocyte infusions following nonmyeloablative allogeneic stem cell transplantation in multiple myeloma. Blood. 2006;107(8):3415-6.
5. Kroger N, Badbaran A, Lioznov M, Schwarz S, Zeschke S, Hildebrand Y, et al. Post-transplant immunotherapy with donor-lymphocyte infusion and novel agents to upgrade partial into complete and molecular remission in allografted patients with multiple myeloma. Exp Hematol. 2009;37(7):791-8.
6. Mohty M, Attal M, Marit G, Bulabois CE, Garban F, Gratecos N, et al. Thalidomide salvage therapy following allogeneic stem cell transplantation for multiple myeloma: a retrospective study from the Intergroupe Francophone du Myelome (IFM) and the Societe Francaise de Greffe de Moelle et Therapie Cellulaire (SFGM-TC). Bone Marrow Transplant. 2005;35(2):165-9.
7. Weber DM, Chen C, Niesvizky R, Wang M, Belch A, Stadtmauer EA, et al. Lenalidomide plus dexamethasone for relapsed multiple myeloma in North America. N Engl J Med. 2007;357(21):2133-42.
8. Dimopoulos M, Spencer A, Attal M, Prince HM, Harousseau JL, Dmoszynska A, et al. Lenalidomide plus dexamethasone for relapsed or refractory multiple myeloma. N Engl J Med. 2007;357(21):2123-32.
9. Davies F, Baz R. Lenalidomide mode of action: linking bench and clinical findings. Blood Rev. 2010;24 (Suppl 1):S13-9.
10. Hayashi T, Hideshima T, Akiyama M, Podar K, Yasui H, Raje N, et al. Molecular mechanisms whereby immunomodulatory drugs activate natural killer cells: clinical application. Br J Haematol. 2005; 128(2):192-203.
11. Minnema MC, van der Veer MS, Aarts T, Emmelot M, Mutis T, Lokhorst HM. Lenalidomide alone or in combination with dexamethasone is highly effective in patients with relapsed multiple myeloma following allogeneic stem cell transplantation and increases the frequency of CD4+Foxp3+ T cells. Leukemia. 2009; 23(3):605-7.
12. El-Cheikh J, Crocchiolo R, Furst S, Ladaique P, Castagna L, Faucher C, et al. Lenalidomide plus donor-lymphocytes infusion after allogeneic stem-cell transplantation with reduced-intensity conditioning in patients with high-risk multiple myeloma. Exp Hematol. 2012;40(7):521-7.
13. Kneppers E, van der Holt B, Kersten MJ, Zweegman S, Meijer E, Huls G, et al. Lenalidomide maintenance after nonmyeloablative allogeneic stem cell transplantation in multiple myeloma is not feasible: results of the HOVON 76 Trial. Blood. 2011;118(9):2413-9.
14. Rajkumar SV, Harousseau JL, Durie B, Anderson KC, Dimopoulos M, Kyle R, et al. Consensus recommendations for the uniform reporting of clinical trials: report of the International Myeloma Workshop Consensus Panel 1. Blood. 2011; 117(18):4691-5.
15. Filipovich AH, Weisdorf D, Pavletic S, Socie G, Wingard JR, Lee SJ, et al. National Institutes of Health consensus development project on criteria for clinical trials in chronic graft-versus-host disease: I. Diagnosis and staging working group report. Biol Blood Marrow Transplant. 2005;11(12):945-56.
16. Bruno B, Patriarca F, Sorasio R, Mattei D, Montefusco V, Peccatori J, et al. Bortezomib with or without dexamethasone in relapsed multiple myeloma following allogeneic hematopoietic cell transplantation. Haematologica. 2006;91(6):837-9.
17. El-Cheikh J, Michallet M, Nagler A, de Lavallade H, Nicolini FE, Shimoni A, et al. High response rate and improved graft-versus-host disease following bortezomib as salvage therapy after reduced intensity conditioning allogeneic stem cell transplantation for multiple myeloma. Haematologica. 2008;93(3):455-8.
18. Mateos-Mazon J, Perez-Simon JA, Lopez O, Hernandez E, Etxebarria J, San Miguel JF. Use of bortezomib in the management of chronic graft-versus-host disease among multiple myeloma patients relapsing after allogeneic transplantation. Haematologica. 2007;92(9):1295-6.
19. Avet-Loiseau H, Attal M, Moreau P, Charbonnel C, Garban F, Hulin C, et al. Genetic abnormalities and survival in multiple myeloma: the experience of the Intergroupe Francophone du Myelome. Blood. 2007;109(8):3489-95.
20. Munshi NC, Anderson KC, Bergsagel PL, Shaughnessy J, Palumbo A, Durie B, et al. Consensus recommendations for risk stratification in multiple myeloma: report of the International Myeloma Workshop Consensus Panel 2. Blood. 2011;117(18): 4696-700.
21. Palumbo A, Rajkumar SV, Dimopoulos MA, Richardson PG, San Miguel J, Barlogie B, et al. Prevention of thalidomide- and lenalidomide-associated thrombosis in myeloma. Leukemia. 2008;22(2):414-23.
22. Dimopoulos MA, Palumbo A, Attal M, Beksac M, Davies FE, Delforge M, et al. Optimizing the use of lenalidomide in relapsed or refractory multiple myeloma: consensus statement. Leukemia. 2011; 25(5):749-60.
23. Kneppers E, Lokhorst HM, Eeltink CM, Huls G, Kersten MJ, Koedam J, et al. Analysis of efficacy and prognostic factors of lenalidomide treatment as part of a Dutch compassionate use program. Clin Lymphoma Myeloma Leuk. 2010;10(2): 138-43.