Developing and Validating a Simplified Score to Predict Early Relapse in Newly Diagnosed Multiple Myeloma (S-ERMM): Analysis from a Pooled Dataset of 2190 Patients

SUPPLEMENTARY APPENDIX

This supplementary material has been provided by the authors to give readers additional information about their work.

Supplementary Methods

iFISH

Bone marrow plasma cells (BMPCs) for interphase fluorescence *in situ* hybridization (iFISH) analyses were enriched using anti-CD138-coated magnetic MicroBeads and AutoMACS Pro Separator (Miltenyi Biotech, San Diego, US-CA) following manufacturer's instructions. BMPCs were then fixed in Carnoy's solution and stored at -20 °C. Slides for iFISH were prepared using probes purchased from Cytocell (Cambridge, United Kingdom), according to manufacturer's instructions. The iFISH routine panel included baseline evaluation of del(13), del(17p) and immunoglobulin H translocations. Nuclei were analyzed using a fluorescent light microscope. Two hundred to 300 BMPCs nuclei from each sample were scored.

ER24 analysis

In the analysis of patients experiencing an early relapse within 24 months from diagnosis (ER24), patients who died for reasons other than progressive disease (PD) or who withdrew consent within 24 months were excluded from the analysis because they were not at risk of progression for the entire first 24 months. Patients experiencing PD within 24 months from diagnosis were included in the ER population; those not experiencing PD within 24 months were included in the reference population. The reference population was then divided into two groups: patients experiencing PD after 24 months at the time of their last follow-up (Late relapse group) and patients who were free from progression at the time of their last follow-up (No PD group).

Clinical endpoints

Overall survival (OS) from relapse was calculated from the first progressive disease (PD1) until the date of death or the date the patient was last known to be alive; second progression-free survival (2^{nd} PFS) was calculated from the start of the second line of treatment after PD1 until the date of PD after the second line of treatment (PD2) or until the date of death (regardless of the cause of death), whichever came first.

Statistical analysis

OS, PFS2, 2^{nd} PFS and OS from relapse were analyzed as time-to-event data using the Kaplan–Meier method. The Cox proportional hazards model was used to estimate the hazard ratio (HR) values and the 95% confidence intervals (CIs). A survival analysis according to early relapse (ER) was performed, including a landmark analysis of OS and PFS2, with the landmark point at the optimal cut-off defined for ER. OS and PFS2 were also analyzed stratifying patients according to the Simplified Early Relapse in Multiple Myeloma (S-ERMM) score.

Supplementary Results

ER24 analysis

55 patients died for reasons other than PD and 57 withdrew their consent within 24 months and were excluded. Patients eligible for the analyses were 2075. Patients with complete data (n=1212) were then split into training (n=838) and validation (n=374) sets. Patient characteristics are presented in Table S2.

ER24 analysis: based on univariate (UV) analysis, 8/14 features were included in the multivariate (MV) analysis: free light chain (FLC), BMPCs, del17p, t(4;14), albumin, β 2-microglobulin (β2m), lactate dehydrogenase >upper limit of normal (LDH>ULN), and Revised International Staging System (R-ISS) stage. In the MV analysis incorporating the R-ISS, both R-ISS II/III vs. I and increased BMPCs increased the risk of ER within 24 months (ER24). When the MV analysis was performed including single features defining the R-ISS, then β2m, BMPCs, del17p and t(4;14) increased the probability of ER24 (Table S3).
Each MV model was then tested on the validation set. *T*he area under the curve (AUC) was 0.59 (95% CI=0.51-0.65) for the ER24 model including the R-ISS and 0.64 (95% CI=0.57-0.71) for the model incorporating individual features.

DS-ERMM score - additional analyses
The Dynamic (D)S-ERMM logistic regression model integrated the achievement of at least a very good partial response (≥VGPR) at 9 months into the baseline S-ERMM score.
In the landmark population included in this analysis (landmark point at 9 months, n=673) 162 patients (24%) experienced ER18, and the baseline score (S-ERMM) retained a discrimination of patients with different ER rates that was comparable to that in the original training population (18%, 34%, and 59% in the S-ERMM Low, Int, and High sub-cohorts, respectively). In fact, in this landmark population, the S-ERMM score retained its prognostic impact, significantly discriminating three groups of patients with different risks of ER18: Int vs. Low (OR=2.41, 95% CI=1.61-3.60, p<0.001) and High vs. Low (OR=6.75, 95% CI=3.36-13.54, p<0.001).
In this same sub-cohort, the concordance (C)-index assessed in the DS-ERMM score (0.68) outperformed the C-index assessed in the S-ERMM score (0.63).
In the validation set, the landmark DS-ERMM population included 327 patients, of whom 30 experienced ER18 (9%), and 265 (81%) achieved ≥VGPR. The prognostic impact of the DS-ERMM score on the prediction of ER18 was not confirmed in the validation set: Int vs. Low (OR=0.97, 95% CI=0.42-2.29, p=0.95) and High vs. Low (OR=2.40, 95% CI=0.88-6.58, p=0.09). Still, in the validation set, the DS-ERMM score partially held a prognostic value in terms of PFS2 and OS. DS-ERMM Int vs. Low: PFS2, HR=2.05, 95% CI=0.76-5.54, p=0.16; OS, HR=1.38, 95% CI=0.37-5.15, p=0.63. DS-ERMM High vs. Low: PFS2, HR=10.15, 95% CI=3.85-26.79, p<0.001; OS, HR=8.95, 95% CI=2.69-29.79, p<0.001.

Outcome after relapse in the ER18 analysis
Patients who received second-line treatment were 261 (84%) in the group of patients who experienced an early relapse within 18 months from diagnosis (ER18) and 274 (82%) in the Late relapse (LR) group. In the ER18 group, 152 (58%) patients received a proteasome inhibitor (PI), 43 (16%) an immunomodulatory agent (IMiD), 35 (13%) autologous stem-cell transplantation (ASCT) and 31 (12%) other treatments. In the LR group, 158 (55%) patients received a PI, 28 (10%) an IMiD agent, 67 (23%) ASCT and 34 (12%) other treatments.
ER18 patients vs. LR patients had a shorter OS from relapse (median, 29.6 vs. 44.5 months, HR=1.48, 95% CI=1.19-1.86; p<0.001) and a shorter 2nd PFS (median, 11.0 vs. 22.0 months, HR=1.59, 95% CI=1.32-1.92; p<0.001; Figure S4A-B).

Table S1A. Treatment regimens in the source studies

Trial	Regimens and doses	N	Age (y)
	TRAINING SET		
IST-CAR-506 (1) NCT01346787 Phase II	K: carfilzomib IV 20 mg/m^2 d 1,2 of Cycle 1 followed by 36 mg/m^2 d 8,9,15,16 and then for all subsequent cycles Cy: cyclophosphamide PO 300 mg/m^2 d 1,8,15 D: dexamethasone PO 40 mg d 1,8,15,22 (nine 28 days cycles followed by maintenance with carfilzomib alone until PD)	58	≥65
IST-CAR-561 (2) NCT01857115 Phase I/II	K: carfilzomib given 20 mg/m^2 IV once daily on d 1 of Cycle 1 only followed by 36/45/56/70 mg/m^2 on days 8, 15 of Cycle 1, then for all subsequent doses 70 mg/m^2 IV once daily on days 1,8,15, followed by 14-day rest period (from d 16 to 28) Cy: cyclophosphamide PO 300 mg/m^2 on days 1,8,15 D: dexamethasone PO 40 mg d 1,8,15,22, or 20 mg d 1-2,8-9,15-16,22-23 (nine 28 days cycles followed by maintenance with carfilzomib alone until PD)	63	≥65
EMN01 (3,4) NCT01093196 Randomized Phase III	ARM A R: lenalidomide PO 25 mg/die for 21 days D: dexamethasone PO 40 mg d 1,8,15,22, or 20 mg in >75-year patients ARM B M: melphalan PO 0.18 mg/Kg or 0.13 mg/Kg in >75-year patients d 1-4 P: prednisone PO 1.5 mg/Kg d 1-4 R: lenalidomide PO 10 mg/die for 21 days ARM C Cy: cyclophosphamide PO 50 mg/die for 21 days or 50 mg every other day in >75-year patients P: prednisone PO 25 mg every other day R: lenalidomide PO 25 mg/d for 21 days (nine 28-day cycles followed by maintenance with lenalidomide or lenalidomide and prednisone)	217 217 220	≥65
RV-MM-EMN-441 (5) NCT01091831 Randomized Phase III	Induction R: lenalidomide PO 25 mg/d for 21 days D: dexamethasone PO 40 mg d 1,8,15,22 (four-28 days cycles, followed by cyclophosphamide 3 g/m^2 and stem-cell mobilization and harvest) Consolidation (ARMS A and B) ARM A Cy: cyclophosphamide PO 300 mg/m^2 d 1,8,15 R: lenalidomide PO 25 mg/d for 21 days D: dexamethasone PO 40 mg d 1,8,15,22 (six 28-day cycles followed by maintenance with lenalidomide or lenalidomide and prednisone) ARM B 2 melphalan IV 200 mg/m^2 followed by stem-cell support (followed by maintenance with lenalidomide or lenalidomide and prednisone)	194 193	≤65

RV-MM-PI-209 (6) NCT00551928 Randomized Phase III	Induction R: lenalidomide PO 25 mg/d for 21 days D: dexamethasone PO 40 mg d 1,8,15,22 (four 28-day cycles, followed by cyclophosphamide 4 g/m^2 and stem-cell mobilization and harvest) Consolidation (ARMS A and B) ARM A M: melphalan PO 0.18 mg/Kg d 1-4 P: prednisone PO 2 mg/Kg d 1-4 R: lenalidomide PO 10 mg/d for 21 days (six 28-day cycles followed by maintenance with lenalidomide or no maintenance) ARM B 2 melphalan IV 200 mg/m^2 followed by stem-cell support (followed by maintenance with lenalidomide or no maintenance)	200 199	≤65
26866138MMY2069 (7) NCT01190787 Randomized Phase II	GROUP1 V: bortezomib sc 1.3 mg/m^2 d 1,8,15,22 P: prednisone PO 50 mg every other day (nine 28-day cycles followed by maintenance with bortezomib until PD) GROUP 2 C: cyclophosphamide PO 50 mg every other day V: bortezomib sc 1.3 mg/m^2 d 1,8,15,22 P: prednisone PO 50 mg every other day (nine 28-day cycles followed by maintenance with bortezomib until PD) GROUP 3 V: bortezomib sc 1.3 mg d 1,8,15,22 M: melphalan PO 2 mg every other day P: prednisone PO 50 mg every other day (nine 28-day cycles followed by maintenance with bortezomib until PD)	51 51 50	≥65

VALIDATION SET			
FORTE (8,9) NCT02203643 Randomized Phase II	ARM A Induction C: carfilzomib 20 mg/m^2 IV d 1-2 Cycle 1 only, followed by 36 mg/m^2 IV once daily on days 8-9, 15-16 cycle 1, then for all subsequent doses 36 mg/m^2 IV once daily d 1-2,8-9,15-16. C: cyclophosphamide300 mg/m^2 PO d 1,8,15. D: dexamethasone 20 mg PO or IV d 1,2,8,9,15,16,22,23. Repeat for 4 28-day cycles. Intensification with high-dose melphalan followed by ASCT Consolidation (90-120 days from melphalan and ASCT): C: carfilzomib36 mg/m^2 IV once daily d 1-2,8-9,15-16. C: cyclophosphamide 300 mg/m^2 PO d 1,8,15. D: dexamethasone 20 mg PO or IV d 1,2,8,9,15,16,22,23. Repeat for 4 28-day cycles. (followed by maintenance with lenalidomide or lenalidomide and carfilzomib)	159	≤65
	ARM B Induction C: carfilzomib 20 mg/m^2 IV d 1-2 Cycle 1 only, followed by 36 mg/m^2 IV once daily d 8-9, 15-16 Cycle 1, then for all subsequent doses 36 mg/m^2 IV once daily on days 1-2,8-9,15-16. R: lenalidomide 25 mg PO daily d 1-21. D: dexamethasone 20 mg PO or IV d 1,2,8,9,15,16,22,23. Repeat for 4 28-day cycles. Intensification with high-dose melphalan followed by ASCT Consolidation (90-120 days from melphalan and ASCT): C: carfilzomib 36 mg/m^2 IV once daily d 1-2,8-9,15-16. R: lenalidomide 25 mg PO daily d 1-21. D: dexamethasone 20 mg PO or IV d 1,2,8,9,15,16,22,23. Repeat for 4 28-day cycles (followed by maintenance with lenalidomide or lenalidomide and carfilzomib)	158	
	ARM C C: carfilzomib 20 mg/m^2 IV d 1-2 Cycle 1 only, followed by 36 mg/m^2 IV once daily d 8-9, 15-16 Cycle 1, then for all subsequent doses 36 mg/m^2 IV once daily d 1-2, 8-9, 15-16. R: lenalidomide 25 mg PO daily d 1-21. D: dexamethasone 20 mg PO or IV d 1,2,8,9,15,16,22,23. Repeat for 12 28-days cycles (4 induction, 4 intensification and 4 consolidation) (followed by maintenance with lenalidomide or lenalidomide and carfilzomib)	157	

Abbreviations. IV, intravenous; PO, oral; sc, subcutaneous; d, day; PD, progressive disease; ASCT, autologous stem-cell transplantation.

Table S1B. Eligibility criteria for clinical trials

Trial	Inclusion criteria	Exclusion criteria
TRAINING SET		
IST-CAR-506 (1)	**Age** ≥ 65 years	**Previous treatment:** any previous treatment
	Disease: NDMM, not previously treated	**Infections**: active hepatitis type B or C, or HIV
	Stem-cell transplantation eligibility: not eligible	**Malignancies:** within the last 5 years (except for basal or squamous cell carcinoma, or in situ cancer of the cervix, or localized prostate cancer of Gleason score <7 with a stable PSA)
	Measurable disease definition: - serum monoclonal protein value (≥ 0.5 g/dL of M-protein) *and/or* - urine light-chain excretion of >200 mg/24 hours; - oligo or non-secretory MM: measurable plasmacytoma >2 cm as determined by clinical examination or applicable radiographs (i.e., MRI, CT-Scan) or an abnormal free light chain ratio (n.v.: 0.26-1.65).	**Other clinical conditions:** - myocardial infarction or unstable angina ≤4 months or other clinically significant heart disease (e.g., CHF NY Heart Association class III or IV, uncontrolled hypertension, history of labile hypertension, or history of poor compliance with an antihypertensive regimen); - peripheral neuropathy >CTCAE grade 2 and ≥grade 2 painful peripheral neuropathy; - pleural effusions requiring thoracentesis or ascites requiring paracentesis within 14 days prior to randomization.
	Life expectancy: >3 months	**Pregnancy:** Y
	Performance status: Karnofsky performance status ≥60%	**Other**: known history of allergy to Captisol® (a cyclodextrin derivative used to solubilize carfilzomib).
	Laboratory test: - platelet count ≥50 x 109/L (≥30 x 109 /L if MM involvement in the bone marrow >50%); - absolute neutrophil count (ANC) ≥1 x 109/L - corrected serum calcium ≤14 mg/dL (3.5 mmol/L).	
	Contraception required: Yes	
IST-CAR-561 (2)	**Age** ≥65 and ≤75 years	**Previous treatment:** does not include radiotherapy, bisphosphonates, or a single short course of steroid; ≤ the equivalent of dexamethasone 40 mg/day for 4 days.
	Disease: NDMM, not previously treated	**Infections:** active hepatitis type B or C, or HIV; acute active infection requiring treatment (systemic antibiotics, antivirals, or antifungals) within 14 days prior to randomization.
	Stem-cell transplantation eligibility: not eligible	**Malignancies:** non-hematologic malignancy within the past 3 years with the exception of (a) adequately treated basal cell carcinoma, squamous cell skin cancer, or thyroid cancer; (b) carcinoma in situ of the cervix or breast; (c) prostate cancer of Gleason Grade 6 or less with stable prostate-specific antigen levels; *or* (d) cancer considered cured by surgical resection or unlikely to impact survival during the duration of the study, such as localized transitional cell carcinoma of the bladder or benign tumors of the adrenal or pancreas.

	Measurable disease definition: - serum monoclonal protein value (generally, but not necessarily, ≥0.5 g/dL of M-protein); - urine light-chain excretion >200 mg/24 hours. Oligo- or non-secretory MM: measurable plasmacytoma >2 cm as determined by clinical examination or applicable radiographs (i.e., MRI, CT-Scan) or abnormal free light chain ratio (n.v.: 0.26-1.65).	**Other clinical conditions:** - Major surgery within 21 days prior to randomization; - pulmonary hypertension; - QTc Interval ≥450 msec; - uncontrolled atrial fibrillation/flutter; - history of torsade de pointe, ventricular tachycardia, ventricular fibrillation; - uncontrolled infection; - unstable angina or myocardial infarction within 4 months prior to randomization, NYHA Class III or IV heart failure, uncontrolled angina, history of severe coronary artery disease, or ECG evidence of acute ischemia or grade 3 conduction system abnormalities unless subject has a pacemaker; - uncontrolled hypertension, uncontrolled congestive heart failure (CHF) or uncontrolled diabetes within 14 days prior to randomization; - significant neuropathy (grades 3–4, or grade 2 with pain) within 14 days prior to randomization; - pleural effusions or ascites paracentesis within 14 days prior to randomization.
	Life expectancy: ≥3 months	**Pregnancy:** Y
	Performance status: Eastern Cooperative Oncology Group (ECOG) performance status 0–2	**Other:** Known history of allergy to Captisol® (a cyclodextrin derivative used to solubilize carfilzomib)
	Laboratory test: - adequate hepatic function; - absolute neutrophil count (ANC) ≥1.0 × 109/L; - corrected serum calcium ≤14 mg/dL (3.5 mmol/L); - hemoglobin ≥8 g/dL (80 g/L); - platelet count ≥50 × 109/L (≥30 × 109/L if MM involvement in the bone marrow >50%) within 14 days prior to randomization. Creatinine clearance (CrCl) ≥15 mL/minute	
	Other clinical data: LVEF ≥40%. 2D transthoracic ECHO is the preferred method of evaluation. Multigated Acquisition Scan is acceptable if ECHO is not available.	
	Contraception required: Yes	
EMN01 (3,4)	**Age** ≥65 years	**Previous treatment**: does not include radiotherapy bisphosphonates, or a single short course of steroid; < to the equivalent of dexamethasone 40 mg/day for 4 days).
	Disease: NDMM, not previously treated	**Infections**: NA
	Stem cell transplantation eligibility: not eligible	**Malignancies**: prior history of malignancies, other than multiple myeloma, unless the subject has been free of the disease for ≥3 years. Exceptions include the following: basal cell carcinoma of the skin, squamous cell carcinoma of the skin, carcinoma in situ of the cervix, carcinoma in situ of the breast, incidental histologic finding of prostate cancer (TNM stage of T1a or T1b).

	Measurable disease definition: - serum monoclonal protein value (generally, but not necessarily, greater than 1 g/dL of IgG M-Protein and greater than 0.5 g/dL of IgA M-Protein) *and*, where applicable, - urine light-chain excretion of >200 mg/24 hours; - non-secretory MM: >30% plasma cells in the bone marrow and at least 1 plasmacytoma >2 cm as determined by clinical examination or applicable radiographs (i.e., MRI or CT scan).	**Other clinical conditions: NA**
	Life expectancy: >6 months	**Pregnancy:** Y.
	Performance status: Karnofsky performance status ≥60%	**Other:** NA.
	Laboratory test: - Platelet count ≥75 x 109/L; - absolute neutrophil count (ANC) ≥1.0 x 109/L; - corrected serum calcium ≤14 mg/dL (3.5 mmol/L); - aspartate transaminase (AST) ≤2.5 x the upper limit of normal (ULN); - alanine transaminase (ALT): ≤2.5 x the ULN; - total bilirubin: ≤1.5 x the ULN; - calculated or measured creatinine clearance ≥30 mL/minute.	
	Other clinical data: adequate cardiac function and pulmonary function	
RV-MM-EMN-441 (5)	**Age** ≤65 years	**Previous treatment:** any anti-MM therapy does not include radiotherapy, bisphosphonates, or a single short course of steroid; < to the equivalent of dexamethasone 40 mg/day for 4 days.
	Disease: NDMM, not previously treated	**Infections:** HBV, HCV, and HIV positive test
	Stem-cell transplantation eligibility: eligible	**Malignancies:** prior history of malignancies, other than MM, unless the subject has been free of the disease for ≥3 years. Exceptions include the following: basal cell carcinoma of the skin, squamous cell carcinoma of the skin, carcinoma in situ of the cervix, carcinoma in situ of the breast, incidental histologic finding of prostate cancer (TNM stage of T1a or T1b).
	Measurable disease definition: - any quantifiable serum monoclonal protein value (generally, but not necessarily, greater than 1 g/dL of IgG M-Protein and greater than 0.5 g/dL of IgA M-Protein) *and*, where applicable, - urine light-chain excretion >200 mg/24 hours; - measurable plasmacytoma >2 cm as determined by clinical examination or applicable radiographs (i.e., MRI, CT-Scan); - bone marrow plasma cells >10%. - Measurable free light chain as only parameter of measurable disease can be considered eligible.	**Other clinical conditions:** Patients previously diagnosed as bearing deep venous thrombosis or arterial thromboembolic event within the latest 12 months.
	Life expectancy: >6 months	**Pregnancy: Y**

	Performance status: Karnofsky performance status ≥60	**Other:** NA.
	Laboratory test: - platelet count ≥75 x 109/L; - absolute neutrophil count (ANC) ≥1.5 x 109/L; - corrected serum calcium ≤14 mg/dL (3.5 mmol/L); - aspartate transaminase (AST): ≤2.5 x the upper limit of normal (ULN); - alanine transaminase (ALT): ≤2.5 x the ULN; - total bilirubin: ≤1.5 x the ULN; - calculated or measured creatinine clearance: ≥20 mL/minute.	
	Other clinical data: - normal ECG and NYHA ≤2; an evaluation of ejection fraction by ECHO or MUGA is optional; - normal chest X ray; an evaluation of pulmonary function studies on mechanical aspects (FEV1, FVC, etc.) and diffusion capacity (DLCO) is optional.	
	Contraception required: Y	
RV-MM-PI-209 (6)	**Age** ≤65 years	**Previous treatment:** previous treatment with anti-MM therapy (does not include radiotherapy, bisphosphonates, or a single short course of steroid; < to the equivalent of dexamethasone 40 mg/day for 4 days).
	Disease: NDMM, not previously treated	**Infections:** not active infectious hepatitis type B or C, and HIV negative test.
	Stem cell transplantation eligibility: eligible	**Malignancies:** prior history of malignancies, other than MM, unless the subject has been free of the disease for ≥ 3 years. Exceptions include the following: basal cell carcinoma of the skin, squamous cell carcinoma of the skin, carcinoma in situ of the cervix, carcinoma in situ of the breast, incidental histologic finding of prostate cancer (TNM stage of T1a or T1b).
	Measurable disease definition: - quantifiable serum monoclonal protein value (generally, but not necessarily, >1 g/dL of IgG MProtein and >0.5 g/dL of IgA M-Protein) *and*, where applicable, - urine light chain excretion >200 mg/24 hours; - measurable plasmacytoma >2 cm as determined by clinical examination or applicable radiographs (i.e., MRI, CT-Scan); bone marrow plasma cells>10%.	**Other clinical conditions:** - Neuropathy of grade ≥2 severity. - previously deep venous thrombosis or arterial thromboembolic event within the latest 12 months or bearing a clear indication for antiplatelet or anticoagulant therapy or bearing a high risk of bleeding complications are ineligible for the sub-study protocol.
	Life expectancy > 6 months	**Pregnancy:** Y.
	Performance status: Karnofsky performance status ≥60%	**Other:** NA.

	Laboratory test: - platelet count ≥75 x 109/L; - absolute neutrophil count (ANC) ≥1.5 x 109/L; - corrected serum calcium ≤14 mg/dL (3.5 mmol/L); - aspartate transaminase (AST): ≤2.5 x the upper limit of normal (ULN); - alanine transaminase (ALT) ≤2.5 x the ULN; - total bilirubin ≤1.5 x the ULN; - calculated or measured creatinine clearance ≥ 30 mL/minute.	
	Other clinical data: - ejection fraction by ECHO or MUGA >50% performed within 60 days prior to registration; - adequate pulmonary function studies >50% of predicted on mechanical aspects (FEV1, FVC, etc.) and diffusion capacity (DLCO) >50% of predicted. Patients unable to complete pulmonary function tests because of MM-related chest pain, must have a high-resolution CT scan of the chest and must also have acceptable arterial blood gases defined as P02 >70.	
	Contraception required: Y	
26866138MMY2069 (7)	**Age** ≥75 years or <75 years with abnormal cardiac, pulmonary, renal, or hepatic function (unsuitable for protocol with standard inclusion/exclusion criteria).	**Previous treatment:** previous treatment with anti-MM therapy (does not include radiotherapy, bisphosphonates, or a single short course of steroid; < to the equivalent of dexamethasone 40 mg/day for 4 days).
	Disease: NDMM, not previously treated.	**Infections:** known positive for HIV or active infectious hepatitis type A, B or C.
	Stem-cell transplantation eligibility: not eligible.	**Malignancies:** NA
	Measurable disease definition: - Secretory myeloma: o any quantifiable serum monoclonal protein value (generally, but not necessarily, greater than 1 g/dL of IgG M-Protein and greater than 0.5 g/dL of IgA M-Protein) *and*, where applicable, o urine light-chain excretion >200 mg/24 hours; - Non-secretory MM: >30% plasma cells in the bone marrow and at least 1 plasmacytoma >2 cm as determined by clinical examination or applicable radiographs (i.e., MRI or CT scan).	**Other clinical conditions:** peripheral neuropathy or neuropathic pain grade ≥2, as defined by National Cancer Institute Common Toxicity Criteria (NCI CTC) 3.0; infiltrative pulmonary disease.
	Life expectancy: >3 months	**Pregnancy:** Y.
	- **Performance status:** Karnofsky performance status >50%	**Other:** NA.
	Laboratory test: platelet count ≥80x109/L; hemoglobin ≥8 g/dL; absolute neutrophil count (ANC) ≥1.0x109/L; AST ≤2.5 times the upper limit of normal (ULN); ALT ≤2.5 times the ULN; total bilirubin ≤1.5 times the ULN; creatinine clearance ≥20 ml/min.	
	Other clinical data: NA	
	Contraception required: Y	

VALIDATION SET		
FORTE (8,9)	**Age** ≤65 years and ≥18 years and eligible for ASCT.	**Previous treatment:** previous treatment with anti-MM therapy (does not include radiotherapy, bisphosphonates, or a single short course of steroid ≤ to the equivalent of dexamethasone 40 mg/day for 4 days).
	Disease: NDMM, not previously treated	**Infections**: clinical active infectious hepatitis type A, B, C or HIV; acute active infection requiring antibiotics or infiltrative pulmonary disease.
	Stem-cell transplantation eligibility: eligible	**Invasive Malignancies:** within past 3 years.
	Measurable disease definition: measurable disease according to IMWG criteria	**Other clinical conditions:** - myocardial infarction or unstable angina ≤4 months or other clinically significant heart disease; - peripheral neuropathy or neuropathic pain grade ≥2, as defined by National Cancer Institute Common Toxicity Criteria (NCI CTC) 4.0; - known history of allergy to Captisol® (a cyclodextrin derivative used to solubilize carfilzomib.
	Life expectancy: ≥3 months	Pregnancy: Y
	Performance status: Karnofsky performance status ≥60%	**Other:** non-secretory MM unless serum free-light chains are present, and the ratio is abnormal or a plasmacytoma with minimum largest diameter >2 cm.
	Laboratory test: - platelet count ≥75 x 10^9/L (≥50 x 10^9 /L if MM involvement in the bone marrow >50%); - absolute neutrophil count (ANC) ≥1 x 10^9/L; - corrected serum calcium ≤14 mg/dL (3.5 mmol/L); - alanine transaminase (ALT) ≤3 x the ULN; - aspartate transaminase (AST) ≤3 x the ULN; - total bilirubin ≤2 x the ULN; - calculated or measured creatinine clearance ≥30 mL/minute.	
	Other clinical data: LVEF ≥40%. 2-D transthoracic echocardiogram (ECHO) is the preferred method of evaluation. Multigated Acquisition Scan (MUGA) is acceptable if ECHO is not available.	
	Contraception required: Y	

Abbreviations. MM, multiple myeloma; NDMM, newly diagnosed MM; ASCT, autologous stem-cell transplantation; HIV, human immunodeficiency virus; Y: yes; NA: not applicable; TNM: tumor nodes metastasis; PSA: prostate-specific antigen; CTCAE, Common Terminology Criteria for Adverse Events; MRI, magnetic resonance imaging; CT scan, computed tomography scan; NYHA, New York Heart Association; ECG, electrocardiogram; LVEF Left ventricular ejection fraction; HBV: hepatitis B Virus; HCV; hepatitis C Virus; ECHO echocardiogram; MUGA, Multigated acquisition scan; NCI CTC, National Cancer Institute Common Toxicity Criteria; ALT, alanine transaminase; AST, aspartate transaminase; IMWG, International Myeloma Working Group.

Table S2. Patient characteristics in the overall population and stratified according to the ER24 outcome as training set and validation set

	TRAINING SET*				VALIDATION SET*			
	Overall population	ER24 population	Reference population	P	Overall population	ER24 population	Reference population	P
N of pts, (%)	838	391 (47%)	447 (53%)		374	79 (21%)	295 (79%)	
Age y, Median, [IQR]	66.0 [57.0, 73.0]	67 [57.0, 75]	65.0 [57.0, 73.0]	0.162	57.0 [51.0, 62.0]	56.0 [50.0, 62]	58.0 [52.0, 62.0]	0.127
Albumin, g/dL: Median, [IQR]	3.8 [3.4, 4.2]	3.7 [3.2, 4.2]	3.8 [3.5, 4.2]	0.012	3.9 [3.5, 4.3]	3.7 [3.5, 4.2]	3.9 [3.5, 4.3]	0.078
β2m, mg/dL: Median, [IQR]	3.6 [2.5, 5.4]	4.0 [2.8, 6.1]	3.2 [2.4, 4.9]	<0.001	2.90 [2.0, 4.2]	3.6 [2.2, 6.0]	2.8 [2.0, 4.0]	0.012
LDH>ULN, N, (%)	83 (10)	54 (14)	29 (6)	0.001	54 (14)	19 (24)	35 (12)	0.011
del17p: N, (%)	121 (14)	73 (19)	48 (11)	0.002	52 (14)	13 (16)	39 (13)	0.579
t(4;14): N, (%)	117 (14)	77 (20)	40 (9)	<0.001	57 (15)	21 (27)	36 (12)	0.003
t(11;14): N, (%)	155 (18)	67 (17)	88 (20)	0.390	87 (23)	18 (23)	69 (23)	1
t(14;16): N, (%)	32 (4)	18 (5)	14 (3)	0.353	19 (5)	4 (5)	15 (5)	1
R-ISS, II/III: N, (%)	609 (73)	315 (81)	294 (66)	<0.001	250 (67)	63 (80)	187 (63)	0.009
Creatinine, mg/dL: Median, [IQR]	0.90 [0.8, 1.2]	0.9 [0.8, 1.2]	0.9 [0.8, 1.1]	0.244	0.8 [0.7, 1.0]	0.8 [0.7, 1.2]	0.8 [0.7, 1.0]	0.736
BMPCs, >60%	243 (29)	130 (33)	113 (25)	0.014	139 (37)	32 (41)	107 (36)	0.575
FLC, λ: N, (%)	300 (36)	151 (39)	149 (33)	0.129	143 (38)	27 (34)	116 (39)	0.481
M component, IgA: N, (%)	185 (22)	86 (22)	99 (22)	1	57 (15)	12 (15)	45 (15)	1
Plasmacytomas N, (%)	78 (9)	38 (10)	40 (9)	0.792	49 (13)	12 (15)	37 (13)	0.666

* *Patients with complete data only*.

Abbreviations. N, number; y, years; ER24, early relapse within 24 months from diagnosis; IQR, interquartile range; alb, albumin; β2m, β 2-microglobulin; LDH, lactate dehydrogenase; ULN, upper limit of normal; del17p, deletion 17p; t, translocation; R-ISS, Revised International Staging System; BMPCs, bone marrow plasma cells; FLC, free light chains; M, monoclonal.

Table S3. Univariate (UV) and multivariate (MV) analyses of the baseline features to predict ER24

	ER24*					
	Analysis including R-ISS			Analysis including individual features		
	UV Analysis	MV Analysis		UV Analysis	MV Analysis	
	p	Odds ratio (95% CI)	p	p	Odds ratio (95% CI)	p
Albumin, (increased by 1 mg/dL)				0.012	-	-
β2m, (increased by 1 mg/dL)				<0.001	1.04 (1.00 – 1.08)	0.045
LDH (> vs. ≤ULN)				0.001	2.12 (1.30 - 3.44)	0.003
del17p (presence vs. no)				0.002	1.84 (1.23 - 2.76)	0.003
t(4;14) (presence vs. no)				<0.001	2.52 (1.65 - 3.83)	<0.001
R-ISS (II/III vs. I)	<0.001	1.97 (1.42 - 2.72)	<0.001			
BMPCs % (increased by 5%)	<0.001	1.04 (1.01 - 1.07)	0.005	<0.001	1.04 (1.01 - 1.07)	0.004
FLC (λ vs. k)	0.129	-	-	0.129	-	-

**Only significant (p<0.1) features according to UV are shown.*
- Excluded before the MV analysis by the Akaike information criterion *(AIC).*

Abbreviations. ER24, early relapse within 24 months from diagnosis; y, years; UV, univariate; MV, multivariate; CI, confidence interval; β2m, β 2-microglobulin; LDH, lactate dehydrogenase; ULN, upper limit of normal; del17p, deletion 17p; t, translocation; R-ISS, revised international staging system; BMPCs, bone marrow plasma cells; FLC, free light chains; M, monoclonal.

List of supplementary figure files

- **Figure S1**. Spline function according to ER18 to dichotomize BMPCs (A) and albumin (B)
- **Figure S2**. Multivariate analysis to predict ER18 according to S-ERMM score at baseline and achievement of ≥VGPR at 9 months
- **Figure S3**. Univariate analysis to predict ER18 comparing DS-ERMM High and Int vs. Low
- **Figure S4**. Landmark analysis (landmark point at 18 months) of OS (A, C) and PFS2 (B, D) in patients with multiple myeloma stratified according to ER18
- **Figure S5.** OS from relapse (A) and 2nd PFS (B) in patients with ER18 vs. late relapse (LR)
- **Figure S6.** OS of ASCT-eligible (A) and ASCT-ineligible (B) patients according to S-ERMM score

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