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ABSTRACT

Histological transformation into diffuse large B-cell lymphoma is a rare complication in patients with Waldenström macroglobulinemia (WM) usually associated with a poor prognosis. The objective of this study was to develop and validate a prognostic index for survival in transformed WM patients. Through this multicenter, international collaborative effort, we developed a scoring system based on data from 133 patients with transformed WM who were evaluated between 1995 and 2016 (training cohort). Univariate and multivariate analyses were used to propose a prognostic index with 2-year survival after transformation as an end-point. For external validation, a data set of 67 patients was used to evaluate the performance of the model (validation cohort). By multivariate analysis, three adverse covariates were identified as independent predictors of 2-year survival after transformation: elevated serum LDH (2 points), platelet count < 100 x 10^9/L (1 point) and any previous treatment for WM (1 point). Three risk groups were defined: low-risk (0-1 point, 24% of patients), intermediate-risk (2-3 points, 59%, hazard ratio (HR) = 3.4) and high-risk (4 points, 17%, HR = 7.5). Two-year survival rates were 81%, 47%, and 21%, respectively (P < 0.0001). This model appeared to be a better discriminant than the International Prognostic Index (IPI) and the revised IPI (R-IPI). We validated this model in an independent cohort. This easy-to-compute scoring index is a robust tool that may allow identification of groups of transformed WM patients with different outcomes and could be used for improving the development of risk-adapted treatment strategies.

INTRODUCTION

Waldenström macroglobulinemia (WM) is a rare B-cell lymphoproliferative disorder characterized by lymphoplasmacytic bone marrow infiltration and production of an IgM monoclonal component.[1] Histological transformation (HT) to diffuse large B-cell lymphoma (DLBCL) has been reported to occur in 2 to 10% of WM patients.[2,3] Most transformed patients present with high-risk features such as extranodal disease, elevated serum lactate dehydrogenase (LDH) levels and high International Prognostic Index (IPI) scores.[3,4] Patients who experience HT have an inferior survival compared to patients who do not transform during their disease course.[3,5] Patients with transformed WM are mainly treated with strategies used in *de novo* DLBCL but exhibit low cure rates, with a median survival from the time of HT ranging from 16 to 32 months.[3-5] However, as some patients experience prolonged survival, identifying those with high-risk features in order to select them for therapeutic intensification and/or novel agents is important. In a previous study of 77 patients, we identified elevated LDH and time to HT above 5 years as possible predictors of shorter survival[4] but there is a need to develop an accurate predictive model for overall survival in a larger cohort of transformed WM patients.

Prognostic indices have been validated and are routinely used in aggressive non-Hodgkin lymphomas (NHL). The IPI was established in 1993 based on the clinical data of patients with *de novo* aggressive NHL treated with CHOP-like chemotherapy.[6] The revised IPI (R-IPI) was proposed in 2006 for a more accurate prediction of outcome in the era of R-CHOP treatment.[7] However, data pertaining to the prognostic value of these scores in the setting of transformed WM are sparse. Moreover, a majority of patients with transformed WM (65% to 76%) present with high IPI scores, probably limiting the accuracy of the IPI in this setting.

The objective of this large international collaborative study was to collect the data on characteristics at WM diagnosis and at HT of a large number of patients with transformed WM and to develop a prognostic index predicting survival following transformation, the

transformed Waldenström International Prognostic Index (tWIPI). The final model was validated in an independent cohort of patients with transformed WM.

METHODS

Patients and data collection for development of the prognostic model

Patients older than 18 years were included in the study if they had a diagnosis of WM and a concurrent or sequential histological diagnosis of DLBCL. The diagnosis of WM was based on criteria established in the Second International Workshop on WM.[8] Patients with a diagnosis of indolent lymphoma other than WM were excluded. Histological assessment of transformation was mandatory for being considered in this study. We retrospectively identified 133 patients diagnosed with HT between January 1995 and December 2016 from French FILO (French Innovative Leukemia Organization) centers, Dana-Farber Cancer Institute (Boston), UCLH (University College London Hospitals) and Nieuwegein (Netherlands) (detail of the centers in Supplementary Table 1). This retrospective study was conducted in accordance with the Declaration of Helsinki and was approved by the institutional review board at each participating institution.

The clinical and disease characteristics considered as candidate prognostic factors were analyzed after reviewing medical records at both time of WM diagnosis and HT. Variables considered as covariates for model building are detailed in the Supplementary methods. In addition, the IPI and the R-IPI were assessed.[5,6] The presence of *MYD88*^{L265P} mutation was tested by allele-specific polymerase chain reaction on bone marrow samples at diagnosis of WM[9].

Validation cohort

The data of 96 patients diagnosed between 1988 and 2018 and treated at Mayo Clinic (Rochester, MN, USA), Athens (Greece), Salamanca (Spain), Amsterdam (Netherlands) and

Toulouse (France) were analyzed (Supplementary Table 1). Information on the 3 parameters of the tWIPI was available for 67 patients.

Statistical methods

The main endpoint of statistical analyses was 2-year overall survival (OS) calculated from diagnosis of HT to the date of death or last follow-up. The survival curves were plotted using the Kaplan-Meier method and compared using the log-rank test for categorical variables. Univariate and multivariate analyses were performed using the Cox proportional hazards model. For the continuous variables, the cutoffs were defined on the basis of published thresholds, for ease of clinical use. The multivariate Cox proportional hazards model included all variables with a *P*-value < 0.10 by univariate analysis. A manual backward selection of covariates was used. The results were presented as hazard ratio (HR) and 95% confidence intervals (CI). A weighted risk score was assigned to each factor included in the final multivariable model. The prognostic score was then defined as the sum of single-risk parameters. Risk subgroups were pooled according to the number of patients within each category and the relative risk of death. The discriminatory value of the prognostic model and the score was assessed using the Concordance Probability Estimates (CPE) by Harrell's concordance index (C-index).[10] Calibration was assessed using the May and Hosmer test for goodness-of-fit. An internal validation of both model and score was performed using the bootstrap resampling method[11] (replication on 2000 different samples drawn with replacement). External validation was performed in a second dataset of subjects. All tests of statistical significance were two-sided, and a *P*-value < 0.05 was considered statistically significant. Statistical analyses were performed using SAS 9.4 (SAS Institute, Cary, NC, USA).

RESULTS

Patient characteristics

The patient characteristics are summarized in Table 1. Of the 133 patients, 17 (13%) were diagnosed at the time of initial diagnosis of WM. Fifty-six percent of patients were male and the median age at WM diagnosis was 64 years (range, 32-86 years). According to the International Prognostic Scoring System (IPSSWM)[12] when available, 31 patients (41%) were classified as low-risk, 30 (39%) as intermediate-risk and 15 (20%) as high-risk. After WM diagnosis, treatment was not initiated in 35 patients (26%) until the diagnosis of HT. The median number of lines of therapy for WM was 1 (range, 0-9). Half of the patients (n = 67) had received rituximab alone or in combination for WM before HT.

The median time from WM diagnosis to HT was 4.3 years (range, 0-25 years). The median age at HT was 68 years (range, 33-89 years). Extranodal involvement by the DLBCL component was noted in 86% of patients. Serum LDH was elevated in 85 patients (72%). The first-line regimens given at HT are listed in Table 1. The median number of lines of therapy given for HT was 1 (range, 0-5). The majority of patients (80%) were treated with first-line regimens used in de novo DLBCL (CHOP +/- rituximab). Rituximab was part of the first-line treatment for HT in 110 patients (87%).

The median follow-up for all patients was 6.4 years (range 0.1-33.7 years) after the diagnosis of WM and 2.3 years (range, 0-16.6 years) after HT. The median OS after HT was 19 months (95% CI 12-31 months) (Supplementary Figure 1A). When we divided the cohort into two groups on the basis of the HT diagnosis date (using three cut-offs: 2004, 2008 and 2012), we did not observe any significant differences in terms of survival (data not shown). At the date of last follow-up, 83 patients (62%) had died. The majority of deaths were related to progressive disease (75%) or infections (14%).

Prognostic factors

In univariate analysis, 6 variables that were associated with shorter 2-year survival after HT were identified for inclusion in multivariate analyses: previous treatment for WM (*P* = .02), prior rituximab exposure (*P* = .01), time to transformation more than or equal to 5 years (*p* = .006), elevated LDH (*P* = .001), B symptoms (*P* = .02) and platelet count less than 100 x 10^9/L (*P* = .006) (Supplemental Figure 2). Age and ECOG performance status at HT were of no significant prognostic value. Among other variables included in the IPI, Ann Arbor stage III and IV and extranodal involvement were not only very common (86% for both) but also not associated with worse outcome. Serum IgM level at transformation was of no significant prognostic value (*P* = .51). The prognostic values of the clinical and biological characteristics for survival at transformation are reported in Table 2.

Development of the prognostic model and the scoring system

In multivariate analysis, independent factors of 2-year survival after HT were elevated serum LDH (*P* = .003; HR = 3.6, 95% CI = [1.53-8.50]), platelet count less than 100 x 10^9/L (*P* = .03; HR = 1.8, 95% CI = [1.04-3.19]) and previous treatment for WM (*P* = .04; HR = 2, 95% CI = [1.04-3.94]) (Table 3). Bootstrapping of the multivariable model showed good internal validity. The May and Hosmer goodness-of-fit test did not identify any calibrations issues (*P* > .6 for each stratum) and the model's Harrell's C-index was 0.75 (CI 95% = [0.65-0.84]). The prognostic model comprised these 3 variables all available for 109 patients. Based on the relative HRs, platelet count < 100 x 10^9/L and previous treatment for WM were scored with 1 point and elevated serum LDH with 2 points. As a result, there were groups of patients with score ranking from 0 to 4. Patients with score 0 were combined with those with score 1 because they were too few to constitute a separate risk group. Patients with scores of 2 and 3 were combined because they correspond both to a group with an intermediate prognosis. The tWIPI was thus created and comprised 3 risk categories: low (0-1 point, 24% of patients), intermediate (2-3 points, 59%) and high (4 points, 17%). The 2-year survival rates

were 81%, 47% and 21%, respectively ($P < .0001$). The distribution of patients into these 3 groups and hazard ratios are shown in Table 4. The survival curves are shown in Figure 1A. The prognostic index displayed high model performances, as assessed by CPE. The Harrell's C-index was 0.75 (95% CI = [0.66-0.85]). The May and Hosmer goodness-of-fit test did not identify any calibrations issues ($P > .7$ for each stratum). Excluding patients with concurrent disease (WM and DLBCL), the model also identified three risk groups with significant different 2-year survivals and displayed good discrimination and calibration properties (Supplementary Figure 3A and Supplementary Table 2A).

Comparison with the International Prognostic Index (IPI) and the revised IPI (R-IPI)

The complete information for the parameters of the IPI (age, serum LDH level, performance status, Ann Arbor stage and number of extranodal sites of disease) was available in 99 of 109 patients used for building the tWIPI. The distribution of patients into the four IPI and the three R-IPI risk groups is shown in Supplementary Table 3. The IPI and the revised IPI were not able to discriminate subgroups of patients with significantly different survival outcome ($P = .33$ and .24 respectively) (Figure 2).

External validation

We applied the tWIPI to 67 other patients with transformed WM. The median follow-up from WM diagnosis and from HT was 8.8 (range, 0.2-20.8) and 3.1 years (range, 0-13.4) respectively. The main clinical characteristics are shown in Table 1. The median survival after HT was 18 months (95% CI 13-NR months) (Supplementary Figure 1B). The model successfully divided the cohort into three groups with 2-year survival rates of 71%, 39% and 0% for the low, intermediate and high-risk groups, respectively ($P = .0001$) (Figure 1B). The prognostic significance of the tWIPI in the external cohort demonstrated good performance for discrimination. The Harrell's C-index was 0.79 (95% CI = [0.64-0.92]). The May and Hosmer goodness-of-fit test did not identify any calibrations issues ($P > .8$ for each stratum).

In the same way as for the training cohort, the results were similar when patients with concurrent disease were excluded (Supplementary Figure 3B and Supplementary Table 2B).

Impact of MYD88 mutation status on survival after transformation

By combining the training and the validation cohorts, we were able to analyze 64 patients with available data on *MYD88* mutation status at time of WM. 43 patients (67%) carried $MYD88^{L265P}$ mutation and 21 (33%) were wild type (WT). The characteristics of the subset of patients for which *MYD88* mutation results were available and that for which the status was not known (n = 136) were comparable, except for a shorter time to transformation in the cohort in which the $MYD88^{L265P}$ mutation status was known. The 2-year survival rates after HT were 67% and 49% in patients with $MYD88^{WT}$ and $MYD88^{L265P}$, respectively (P = .018) (Supplementary Figure 4).

DISCUSSION

The prognosis of transformed indolent lymphomas is historically poor despite combination chemoimmunotherapy, especially in chronic lymphocytic leukemia (Richter syndrome) and Waldenström macroglobulinemia.[4,13] Characterization of adverse prognostic factors in this setting is important for identifying specific risk groups and comparing different therapeutic approaches. There is no specific prognostic score for transformed WM and the existing scores such as the IPI appear not to discern prognosis appropriately.

We developed an easy-to-use prognostic index relying on a model with three-risk groups defined by the presence, or not, of one or more of the following parameters: previous treatment for WM, serum LDH level and platelet count at the time of HT. Previous treatment for WM is typically associated with previous exposure to rituximab and a prolonged time to transformation. This parameter could reflect chemo-resistance and/or immunologic impairment related to the disease and its previous treatment. Serum LDH level is a well-

established prognostic factor both in hematological malignancies and solid tumors.[14-17] Its prognostic role has been validated in both WM and DLBCL, being one of the variables included in the revised IPSSWM and the IPI, respectively.[6,18] Low platelet count, also part of the IPSSWM, is usually associated with a poor prognosis in hematological malignancies[12,19] and could reflect a critical level of bone marrow involvement. For development of the prognostic score, only pretreatment characteristics were considered. Nevertheless, despite the retrospective nature of the study, first-line treatments at HT were quite uniform with a majority of patients being treated with R-CHOP chemoimmunotherapy, similarly to *de novo* DLBCL. This is unlikely to have influenced the analysis.

Using this index, we were able to separate patients with transformed WM into 3 risk groups. In patients with a good prognosis (score 0-1), the 2-year survival is 81%. This indicates that standard R-CHOP regimen could lead to a prolonged control of high grade component in a majority of these patients. In the intermediate group (score 2-3), less than half of the patients are alive after 2 years. The role of consolidative therapies such as high-dose therapy with stem cell transplantation in younger patients or association with targeted therapies would be interesting to investigate. For patients in the high-risk group (score 4), the outcome is very poor with a 2-year survival of 21%. Innovative therapies are required and these patients should be directed to clinical trials with new agents. Chimeric antigen receptor (CAR) T-cell therapies have shown to be effective and to lead to durable response in relapsed/refractory DLBCL including transformed follicular lymphomas.[20-22] The potential effectiveness of CAR T-cell therapy in transformed WM has recently been suggested based on one case report with complete response maintained at 1 year.[23] Clinical trials are needed to evaluate the place of CAR T-cell therapy in WM and transformed WM.

An important finding of our study is that the IPI and the R-IPI do not seem appropriate to identify patients with significantly different outcomes in the particular setting of transformed WM. The application of the IPI in our cohort showed a lack of separation of the 2 intermediate-risk and the high-risk groups, most patients with transformed WM falling in the

high-intermediate or the high-risk categories. In addition, of the IPI risk factors, only serum LDH level showed prognostic relevance in univariate analyses. The IPI and the R-IPI have been studied in other transformed lymphomas such as transformed follicular lymphoma and marginal zone lymphoma and could predict survival.[24,25] In Richter syndrome, the RS prognostic score has been proposed and is based on 5 adverse risk factors.[13] Interestingly, the 3 variables of our score are part of the RS score.

We performed internal validation by bootstrap[11] and confirmed a high stability of the developed model. Despite the rarity of the disease, we were able to validate the prognostic index in an independent cohort of patients with transformed WM. Our model displayed good discrimination properties in the validation cohort, identifying three risk groups with similar 2-year survival after transformation to the ones in the training set. This external validation confirms the robustness and the reproducibility of the tWIPI.

Advances in the biology of WM have demonstrated the role of mutation status for outcome prediction. *MYD88*^{L265P} mutation is found in 95% of WM patients and represents an important diagnostic marker.[26] *MYD88*^{WT} WM patients seem to have a worse outcome and a higher incidence of DLBCL events.[5,27] In our study, molecular parameters were available only in one third of patients and so could not be included in the initial analysis. By combining the 2 cohorts, we could analyze 64 patients and found that patients with *MYD88*^{L265P} mutation had a significantly shorter 2-year survival after transformation compared to patients with *MYD88*^{WT} disease. This finding should be confirmed in a larger cohort to perform multivariate analysis. However, this result is in line with previous studies showing that MYD88 mutations are associated with worse survival in *de novo* DLBCL.[28-30]

Our study has some limitations. First, a majority of patients were exposed to chlorambucil and/or fludarabine-based regimen as therapy for WM. Half of the patients received rituximab alone or in combination and very few patients were treated with Bruton tyrosine kinase (BTK) inhibitors such as ibrutinib. The tWIPI warrants further validation in a cohort of transformed

WM patients treated with more contemporary regimens at time of WM. Secondly, in the present study, we were not able to assess the clonal relationship between the original WM and DLBCL. It is known that the occurrence of DLBCL in WM can result either from HT or arise as a *de novo*, not clonally related lymphoma.[31] This phenomenon has been widely described in Richter syndrome where *de novo* DLBCL usually carries a better prognosis (median survival of 5 years vs 8-16 months for clonally related DLBCL transformation).[32] Nevertheless, one strength of our study was the strict and homogeneous definition of transformation by restricting inclusion to histologically documented transformation.

In conclusion, through this large multicenter study with the aim to identify prognostic factors of survival in transformed WM, we developed a prognostic model and validated it in an independent series of patients. Retrospective and prospective international multicenter studies are needed to define the optimal therapeutic strategies in transformed WM. Our prognostic score could help physicians individualize treatment strategy and improve the management of patients with transformed WM by selecting the most appropriate treatment.

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Table 1. Patient characteristics at diagnosis of WM and at transformation in the training and validation sets

Variable	Training set	Validation set
Entire cohort	**n = 133**	**n = 67**
Median age (years)	64 (range, 32-86)	63 (range, 27-82)
Sex male/female (ratio)	75/58 (1.3/1)	42/25 (1.7/1)
Prior MGUS history	20 (15%)	4 (17%)
WM characteristics (concurrent WM and HT excluded)	**n = 116**	**n = 64**
Serum IgM level, g/L	17.7 (range, 1.4-66.7)	26 (range, 0.9-106)
IPSS score	n = 76	n = 43
0-1	31 (41%)	15 (40%)
2	30 (39%)	7 (18%)
≥ 3	15 (20%)	16 (42%)
Median number of regimens prior to HT	1 (range, 0-9)	1 (range, 0-9)
Therapies before HT	n = 98	n = 54
Chlorambucil	43 (44%)	15 (28%)
Fludarabine-based regimens	41 (42%)	16 (30%)
Bendamustine +/- rituximab	19 (19%)	6 (11%)
CHOP +/- rituximab	17 (17%)	11 (20%)
Bortezomib-based regimens	15 (15%)	7 (13%)
RCD	14 (14%)	12 (22%)
Ibrutinib	5 (5%)	1 (2%)
Autologous stem-cell transplantation	4 (3%)	0 (0%)
Rituximab (alone or in combination)	67 (50%)	41 (76%)
HT characteristics	**n = 133**	**n = 67**
Median age (years)	68 (range, 33-89)	69 (range, 31-89)
PS (0-1/≥ 2)	59/48 (55%/45%)	30/18 (63%/37%)
B symptoms	56 (47%)	30 (49%)
Extranodal involvement	111 (86%)	46 (69%)
Serum IgM level, g/L	6.9 (range, 0-66.6)	6.3 (range, 0.3-43.9)
Absolute neutrophils, x 10^9/L	4.1 (range, 0.2-20.2)	3.6 (range, 0.4-12.3)

Absolute lymphocytes, x 10^9/L	0.9 (range, 0.1-56)	1.2 (range, 0.2-30)
Hemoglobin, g/L	104 (range, 46-155)	111 (range, 43-154)
Platelets, x 10^9/L	172 (range, 9-610)	186 (range, 8-576)
Elevated LDH	85 (72%)	37 (55%)
Albumin level < 3.5 g/dL	62 (56%)	30 (51%)
Stage III or IV	96 (86%)	43 (83%)
Median number of lines	1 (range, 0-5)	2 (range, 0-5)
First-line therapies after HT	n = 127	n = 63
CHOP-like regimen +/- rituximab	102 (80%)	42 (67%)
DHAP +/- rituximab	10 (8%)	3 (6%)
GEMOX +/- rituximab	3 (2%)	3 (6%)
Rituximab-containing regimen	110 (87%)	44 (70%)
Autologous stem-cell transplantation	20 (16%)	13 (21%)
Allogeneic stem-cell transplantation	6 (5%)	2 (3%)

MGUS, monoclonal gammopathy of undetermined significance; IPSS, International Prognostic Scoring System; WM, Waldenström macroglobulinemia; HT, histological transformation; CHOP, cyclophosphamide-doxorubicine-oncovin-prednisone; RCD, rituximab-cyclophosphamide-dexamethasone; PS, performance status; LDH, lactate dehydrogenase; IPI, International Prognostic Index; DHAP, dexamethasone-cytarabine-cisplatin; GEMOX, gemzar-oxaliplatin.

Table 2. Results of the univariate analysis of prognostic factors

Characteristic	**No. of patients (%)**	**2-year OS (%)**	***P***
Sex			
Male	75 (56)	54.7	
Female	58 (44)	48.3	.64
Previous treatment for WM			
No	35 (26)	65.7	
Yes	98 (74)	46.9	**.02**
Prior rituximab exposure			
No	66 (50)	57.6	
Yes	67 (50)	46.3	**.01**
Time to transformation			
Less than 5 years	77 (58)	59.7	
5 years or more	56 (42)	41.1	**.006**
Age at transformation			
65 y or less	44 (33)	45.5	
More than 65 y	89 (67)	55.1	.61
Performance status (ECOG)			
0-1	59 (55)	50.9	
More than 1	48 (45)	45.8	.22
B symptoms			
Absence	62 (53)	58.1	
Presence	56 (47)	41.1	**.02**
Extranodal involvement			
Absence	18 (14)	61.1	
Presence	111 (86)	51.4	.78
Ann-Arbor stage			
I-II	16 (14)	43.8	
III-IV	96 (86)	49	.88
Leukocyte count			
4 x 10^9/L or more	56 (48)	46.4	
Less than 4 x 10^9/L	61 (52)	55.6	.73

Hemoglobin level 100 g/L or more Less than 100 g/L	 68 (57) 52 (43)	 52.9 46.2	 .78
Platelet count 100 x 10^9/L or more Less than 100 x 10^9/L	 88 (75) 29 (25)	 56.8 27.6	 **.006**
Serum albumin 35 g/L or more Less than 35 g/L	 48 (44) 62 (56)	 54.2 50	 .80
Serum LDH Less than or equal to ULN Greater than ULN	 33 (28) 85 (72)	 78.8 42.4	 **.001**
Serum β2-microglobulin Less than 3 mg/L 3 mg/L or more	 16 (28) 41 (71)	 50 53.7	 .37

WM, Waldenström macroglobulinemia; ECOG, Eastern Cooperative Oncology Group; LDH, lactate dehydrogenase; OS, overall survival; ULN, upper limit of normal.

Table 3. Results of the Cox regression analysis: final prognostic model

Variable	Adverse factor	Hazard ratio	95% CI	*P*
Previous treatment for WM	≥ 1	2	1.04-3.94	.04
Platelet count at HT	< 100 x 10^9/L	1.8	1.04-3.19	.03
LDH at HT	> ULN	3.6	1.53-8.50	.003

WM, Waldenström macroglobulinemia; HT, histological transformation; LDH, lactate dehydrogenase; ULN, upper limit of normal; CI, confidence interval.

Table 4. The transformed Waldenström International Prognostic Index: outcome and relative risk of death according to risk group

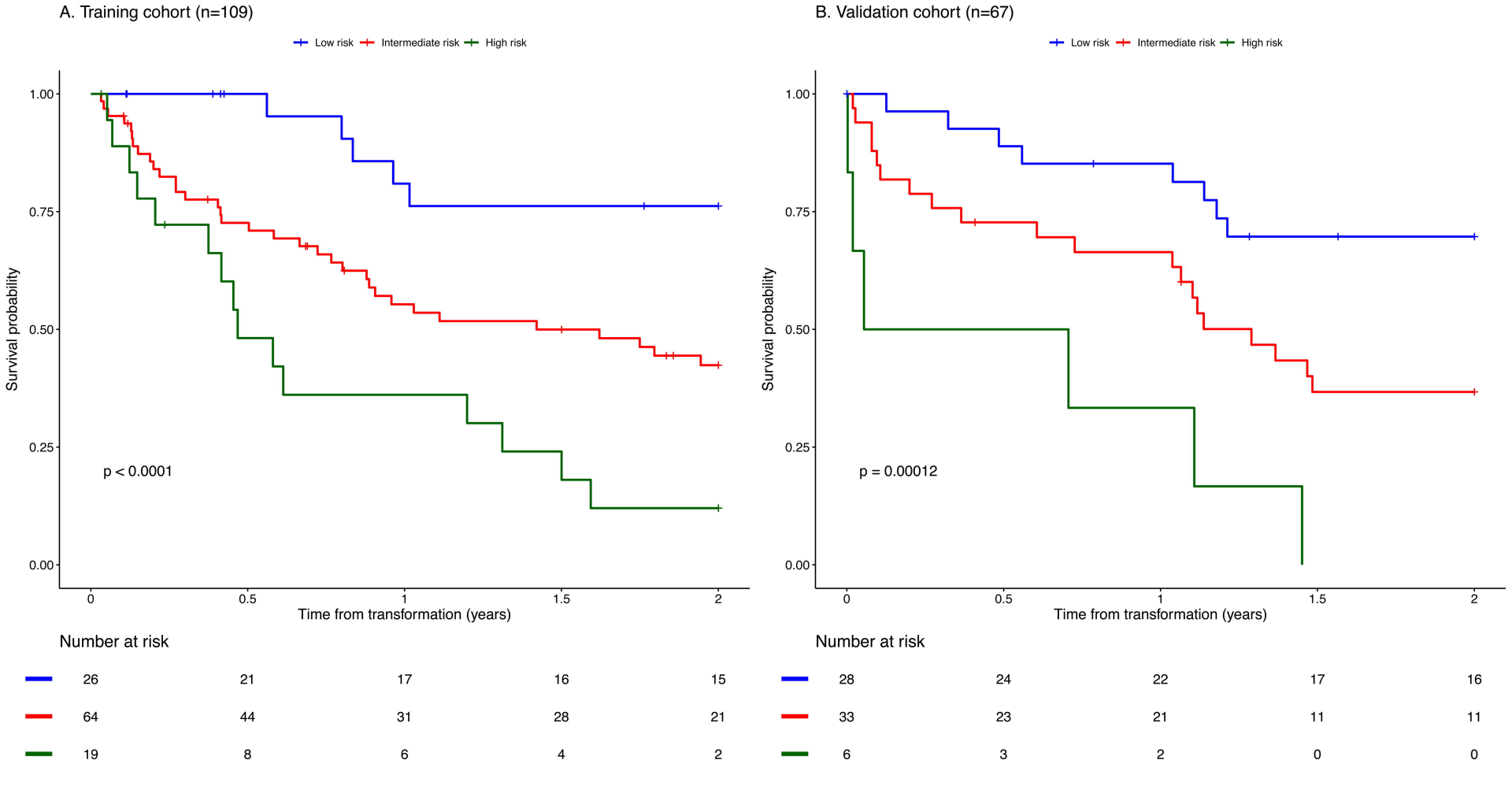
Risk group	Score	No. of patients (%)	2-year OS, %	Median survival, months	HR	95% CI
Low	0-1	26 (24)	80.8	NR	1.0	NA
Intermediate	2-3	64 (59)	46.9	16.8	3.4	1.3-8.7
High	4	19 (17)	21.1	4.8	7.5	2.7-20.7

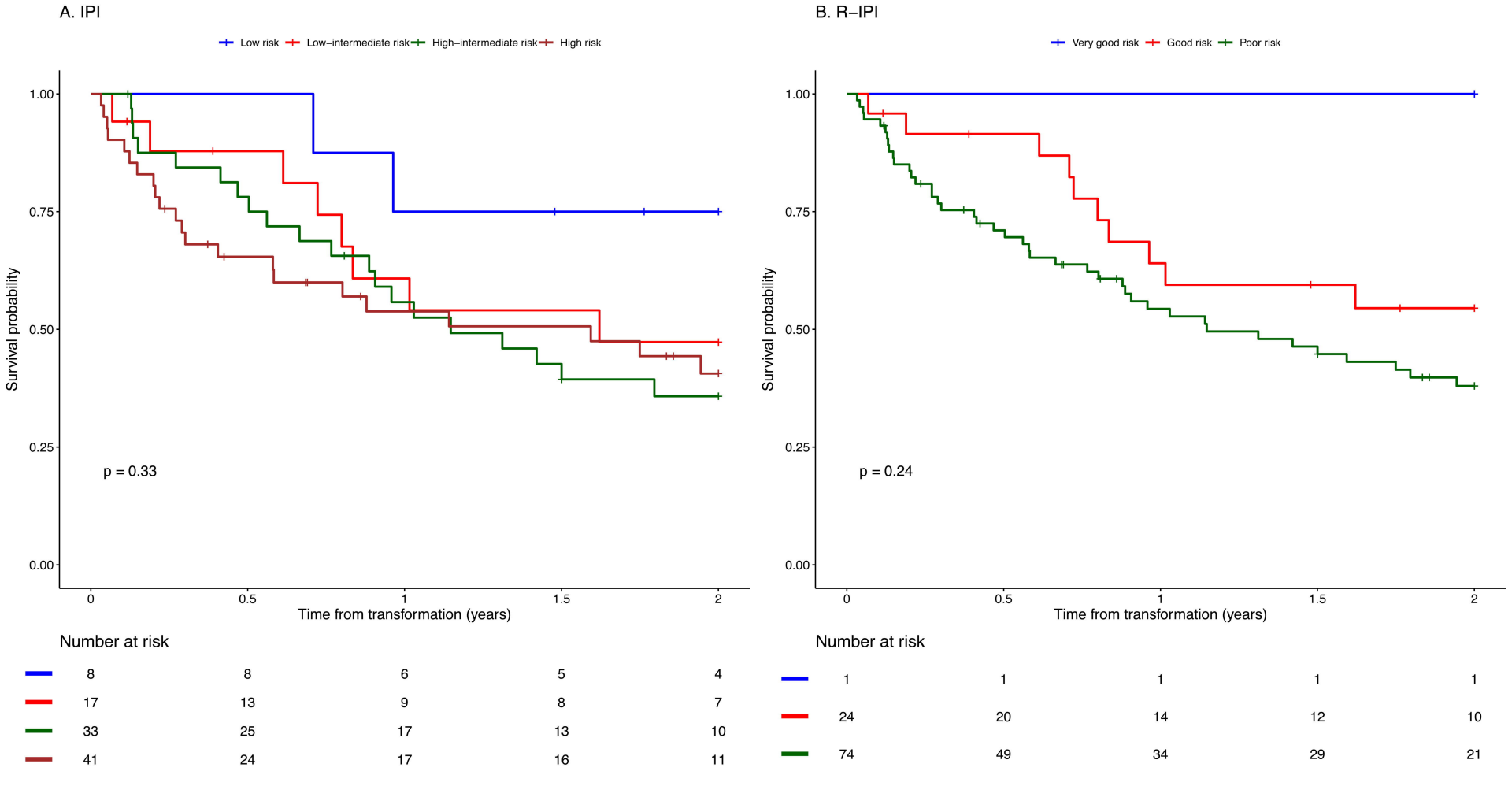
OS, overall survival; HR, hazard ratio; CI, confidence interval; NR, not reached; NA, not applicable

FIGURE LEGENDS

Figure 1. Kaplan-Meier curves for survival after transformation according to subgroups defined by the tWIPI. (A) The model was built using 3 variables: previous treatment for WM, LDH at transformation and platelet count at transformation. It divided the cohort into three risk groups: low-, intermediate-, and high-risk with a 2-year survival after transformation of 80.8, 46.9 and 21.1%, respectively. (B) Validation cohort: 2-year survival after transformation of 71.4, 39.4 and 0%, for the low-, intermediate-, and high-risk groups, respectively.

Figure 2. Kaplan-Meier curves for survival after transformation of 99 patients according to risk group as defined by the IPI (A) and the revised IPI (B).


A. Training cohort (n=109)
Low risk
Intermediate risk
High risk
1.00
0.75
0.50
0.25
0.00
Survival probability
p < 0.0001
0
0.5
1
1.5
2
Time from transformation (years)
Number at risk
26 21 17 16 15
64 44 31 28 21
19 8 6 4 2
B. Validation cohort (n=67)
Low risk
Intermediate risk
High risk
1.00
0.75
0.50
0.25
0.00
Survival probability
p = 0.00012
0
0.5
1
1.5
2
Time from transformation (years)
Number at risk
28 24 22 17 16
33 23 21 11 11
6 3 2 0 0


A. IPI
Low risk
Low−intermediate risk
High−intermediate risk
High risk
1.00
0.75
0.50
0.25
0.00
Survival probability
p = 0.33
0
0.5
1
1.5
2
Time from transformation (years)
Number at risk
8 8 6 5 4
17 13 9 8 7
33 25 17 13 10
41 24 17 16 11
B. R−IPI
Very good risk
Good risk
Poor risk
1.00
0.75
0.50
0.25
0.00
Survival probability
p = 0.24
0
0.5
1
1.5
2
Time from transformation (years)
Number at risk
1 1 1 1 1
24 20 14 12 10
74 49 34 29 21

Supplementary Appendix

Supplement to: Durot E, Kanagaratnam L, Zanwar S et al. A prognostic index predicting survival in transformed Waldenström macroglobulinemia.

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Supplementary Methods

Patients and data collection for development of the prognostic model

Fifteen variables were considered as covariates for model building. These covariates were: sex, number of lines of treatment for WM, rituximab exposure for the treatment of WM, time to transformation, and at time of HT: age, Eastern Cooperative Oncology Group (ECOG) performance status (PS, 0-4), presence of B symptoms (defined as recurrent fever, nights sweats, or > 10% weight loss), extranodal involvement, Ann Arbor stage (I-IV), leucocytes, hemoglobin and platelets counts, and serum albumin, LDH and β2-microglobulin levels. Extranodal involvement at HT was confirmed by the site of biopsy.

Training and validation cohorts

The training and the validation cohorts were formed consecutively. After development of the prognostic model and the scoring system, other centers were contacted to provide data from patients with transformed WM so that the validation cohort was independent. The inclusion criteria for the validation set were similar to those for the training set.

End point

Given the reported poor survival after HT in transformed WM ranging from 1.5 to 3 years[1-3] and the fact that the majority of events, mainly deaths, occur in the first 2 years following the diagnosis of DLBCL[4], 2-year survival after HT was chosen as the main end point for statistical analyses.

Statistical methods

Descriptive statistics included all clinical and demographic characteristics. Continuous variables were expressed as median and range and categorical variables as number and percentages.

Log-linearity and the proportional hazards assumptions were checked. When the log-linearity assumption was not verified, continuous variables were converted into categorical form according to thresholds used in clinical practice or literature data determined before analyzes.

The C-index estimates the proportion of all pairs of patients in whom prediction and outcome are concordant and takes values from 0.5 (no discrimination) to 1.0 (perfect discrimination).

The Harrell's C-index and the May and Hosmer test for goodness-of-fit were used to assess discrimination and calibration in the validation cohort, as was done in the training cohort.

References

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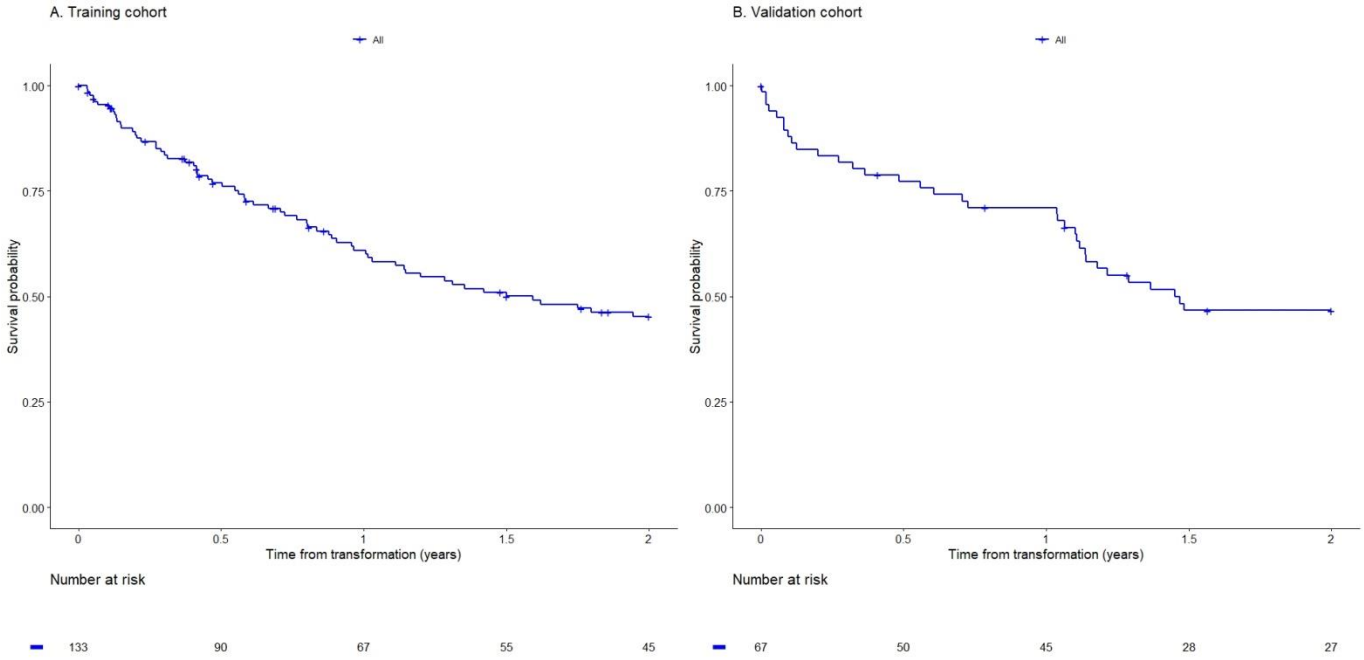
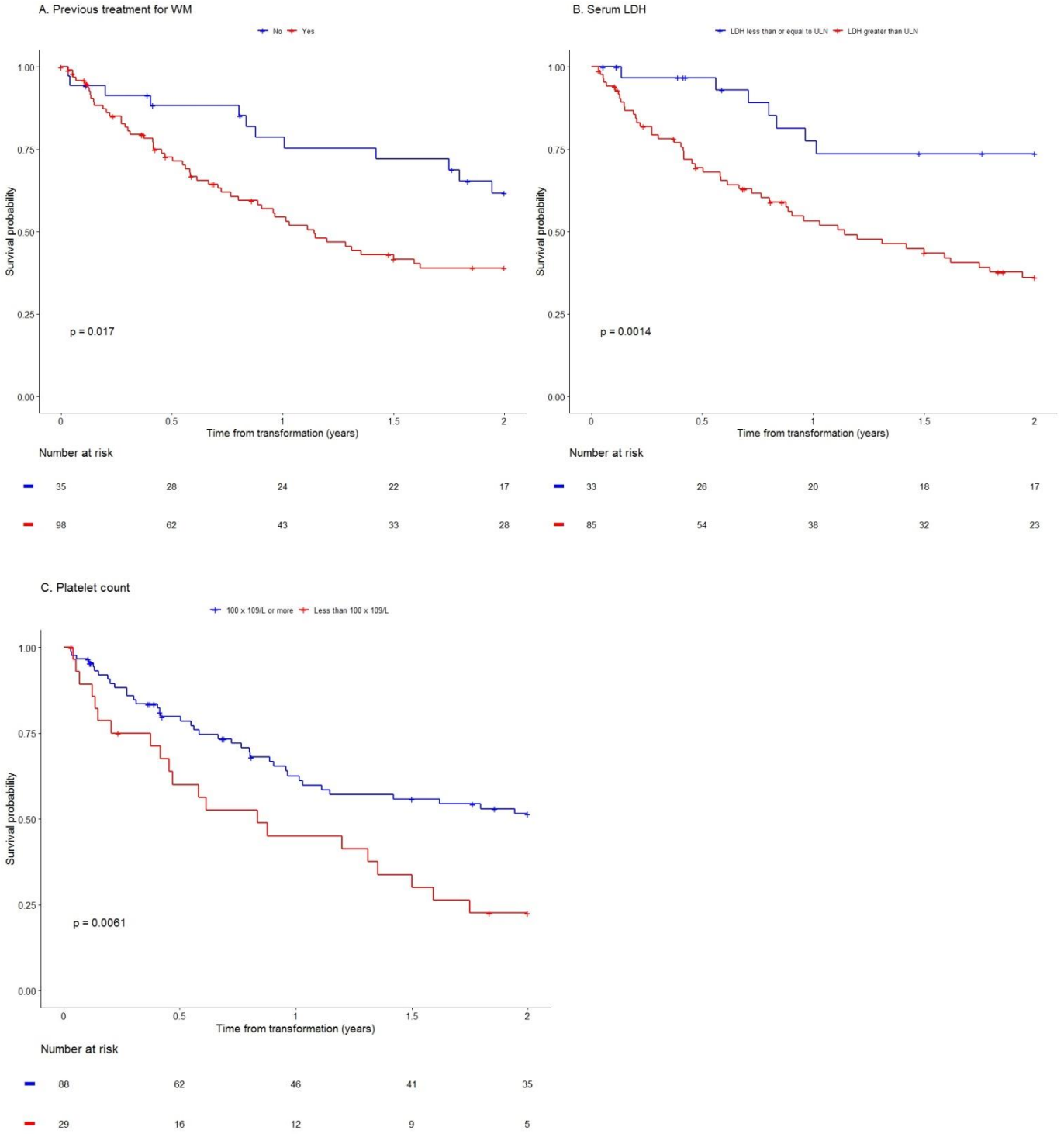
Supplementary Table 1. Accrual of patients

Center	Training set (n = 133)	Validation set (n = 67)
FILO centers (France and Belgium)	**80**	**8**
Lens	10	
Pitié Salpêtrière, Paris	9	
Poitiers	8	
Toulouse		8
Lyon (Léon Bérard)	7	
Strasbourg	7	
Henri Mondor, Créteil	5	
Reims	5	
Rouen	5	
Amiens	3	
Besançon	3	
Bruxelles	3	
Versailles	3	
Clermont-Ferrand	2	
Cochin, Paris	2	
Grenoble	2	
Saint-Louis, Paris	2	
Argenteuil	2	
Le Mans	1	
Nancy	1	
Dana Farber Cancer Institute, Boston, MA, USA	**36**	
University College London Hospitals, London, UK	**13**	
Nieuwegein, The Netherlands	**4**	
Mayo Clinic, Rochester, MN, USA		**27**
University Hospital and IBSAL, Salamanca, Spain		**15**
National and Kapodistrian University of Athens, Greece		**14**
Academical Medical Center, Amsterdam, The Netherlands		**3**

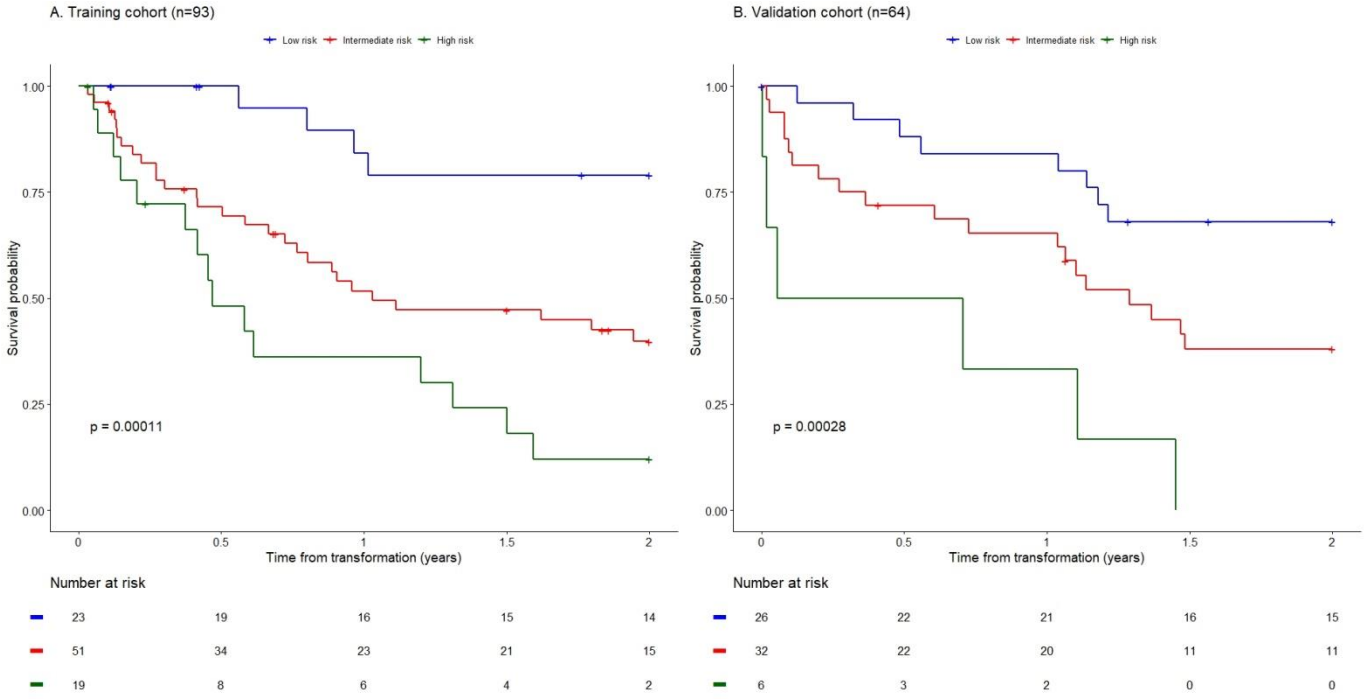
Supplementary Figure 1. Kaplan-Meier curves of survival after transformation. The median OS after HT was 19 months (95% CI 12-31 months) in the training cohort (A) and 18 months (95% CI 13-NR months) in the validation cohort (B).



Supplementary Figure 2. Kaplan-Meier curves of survival after transformation stratified by (A) previous treatment for WM, (B) LDH level at transformation, and (C) platelet count at transformation.



Supplementary Figure 3. Kaplan-Meier curves of survival after transformation according to subgroups defined by the tWIPI after exclusion of patients with concurrent disease. (A) Training cohort. (B) Validation cohort.



Supplementary Table 2. The transformed Waldenström International Prognostic Index: outcome and relative risk of death according to risk group after exclusion of patients with concurrent disease. (A) Training cohort. (B) Validation cohort.

(A) Training cohort (n = 93)

Risk group	Score	No. of patients (%)	Median survival, months	HR	95% CI
Low	0-1	23 (25)	NR	1.0	NA
Intermediate	2-3	51 (55)	12.3	4.2	1.5-11.9
High	4	19 (20)	5.6	8.4	2.8-25.5

HR, hazard ratio; CI, confidence interval; NR, not reached; NA, not applicable

Harrell's C-index: 0.75 (95% CI = [0.65-0.85])
The May and Hosmer goodness-of-fit test: P value > 0.8 for each stratum

(B) Validation cohort (n = 64)

Risk group	Score	No. of patients (%)	Median survival, months	HR	95% CI
Low	0-1	26 (41)	NR	1.0	NA
Intermediate	2-3	32 (50)	15.5	2.5	1.1-5.6
High	4	6 (9)	4.6	7.5	2.6-22.0

HR, hazard ratio; CI, confidence interval; NR, not reached; NA, not applicable

Harrell's C-index: 0.81 (95% CI = [0.65-0.93])
The May and Hosmer goodness-of-fit test: P value > 0.4 for each stratum

Supplementary Table 3. Outcome according to risk group as defined by the IPI and R-IPI in 99 patients with data available for tWIPI and IPI

Risk group	Number of IPI factors	Distribution of patients, %	2-year OS, %	HR	95% CI
Standard IPI					
Low risk	0-1	8	75	1.0	NA
Low-intermediate	2	17	52.9	2.5	0.5-11.7
High-intermediate	3	33	39.4	3.3	0.8-13.9
High	4-5	41	46.3	3.4	0.8-14.3
Revised IPI					
Very good	0	1	100	NA	NA
Good	1-2	24	58.3	NA	NA
Poor	3-4-5	74	43.2	NA	NA

IPI, International Prognostic Index; OS, overall survival; HR, hazard ratio; CI, confidence interval; NA, not applicable.

Supplementary Figure 4. Kaplan-Meier curve of survival after transformation according to MYD88 L265P mutation status.