MYD88 Mutation Status Impacts Overall Survival and Risk of Histological Transformation in Waldenstrom's Macroglobulinemia.

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Introduction

Activating mutations in MYD88 and CXCR4 are present in 93-95% and 30-40% of patients with Waldenstrom's Macroglobulinemia (WM), respectively. Mutations in MYD88 trigger NF-KB dependent growth and survival of WM cells through BTK/IRAK, and AKT/ERK through HCK (Yang et al, Blood 2013; 2016). Patients with MYD88 mutations show high levels of response activity to ibrutinib, that targets BTK and HCK, while responses are poor in MYD88^{WT} patients denoting biological differences between these two subgroups (Treon et al, NEJM 2015). The mutational landscape of MYD88 wild-type (WT) patients is under investigation, though is likely to involve alternative signaling pathways based on transcriptome studies (Hunter et al, Blood 2016). In a previous study, we observed that patients with MYD88^{WT} were at higher risk of death, while CXCR4 mutation status had no impact on overall survival (Treon et al, Blood 2014). Given the uncommon nature of MYD88^{WT} WM disease, we sought in this study to investigate the impact of MYD88^{WT} status in a larger population of WM patients.

Patients and Methods

WHO and WM Consensus guidelines were utilized to establish WM diagnosis, and thorough pathological and laboratory testing was undertaken to exclude non-WM IgM secreting B-cell entities. We utilized sorted CD19^{+} lymphoplasmacytic cells (LPC) obtained from the bone marrow of WM patients, and determined MYD88 mutation status by highly sensitive and specific AS-PCR for MYD88^{L265P} as before (Xu et al, Blood 2013). Patients with wild-type MYD88 by AS-PCR underwent Sanger sequencing to exclude non-L265P MYD88 mutations as before (Treon et al, NEJM 2015). CXCR4 mutation status was determined in sorted LPC by AS-PCR and Sanger sequencing as before (Xu et al, BJH 2016). We identified 46 MYD88^{WT} WM patients by these efforts, and compared their findings at time of diagnosis, and survival outcome to 262 patients with MYD88 mutated (MYD88^{MUT}) disease who were diagnosed over the same time-period. Median follow-up for all patients was 74.7 (0.5-324.9 months), and was similar for MYD88^{WT} and MYD88^{MUT} WM patients (64.1 vs. 73.7 months, respectively; p=0.71).

Data sets were analyzed by analysis of variance and nonparametric comparisons made by the Fisher's exact probability test. The survival from WM diagnosis, defined as the time between WM diagnosis to last follow-up or death, and the time from WM diagnosis to transformation to an aggressive lymphoma were estimated using the Kaplan-Meier method. Survival curves were compared using the log-rank test. For the survival estimates in WM patients who did or did not transform to an aggressive lymphoma, we used the Simon-Makuch method, and the Mantel-Byar test for comparison (stsplit in STATA). Follow-up time was estimated by reverse censoring. Univariate and multivariate survival analyses were performed using Cox proportional-hazard regression models. The outcome of interest is reported as hazard ratio (HR) with 95% confidence interval (CI). A P value < .05 was deemed to be significant. All calculations and graphs were obtained using STATA/SE 13.1 (StataCorp, College Station, TX).

Results

Table 1. Baseline characteristics for MYD88^{WT} and MYD88^{MUT} patients

WM patients	N=	Age (yrs)	Gender Male/Female	IgM (mg/dL)	BM (%)	Hb (g/dL)	B_2M (mg/L)
MYD88^{WT}	46	59 (range 29-85)	47.8% /52.2%	2,980 (range 160-9,000)	37.5 (range 2.5-95)	11.0 (range 4.0-15.5)	3.4 (range 1.5-19.2)
MYD88^{MUT}	262	61 (range 31-91)	62.9% /37.1%	2,650 (range 94-12,400)	35 (range 5-95	12.0 (range 6.0-16.3)	2.8 (range 1.4-11.8)
p-value		0.29	0.07	0.77	0.88	0.002	0.09

Among 262 MYD88^{MUT} patients, 261 had L265P and 1 the S243N MYD88 mutations; 101 (38.5%) were CXCR4 mutated. During the follow-up period, there were 11 (23.9%) deaths for the MYD88^{WT} versus 15 (5.7%) deaths among MYD88^{MUT} patients (p=0.003). **Figure 1A** shows overall survival based on MYD88 and CXCR4 mutation status from time of diagnosis for all patients. The estimated 10-year survival was 73% (95% CI 52-86%) and 90% (95% CI 82-95%) for MYD88^{WT} and MYD88^{MUT} patients, respectively (Log-rank p<0.001), and did not differ within MYD88^{MUT} patients by CXCR4 mutation status (Log-rank p=0.69). Multivariate analyses that included age, gender, serum IgM, hemoglobin, BM disease involvement, serum B2M, MYD88 and CXCR4 mutation status showed that MYD88 mutation status alone was a significant determinant for overall survival. The results of the survival analyses are shown in **Table 2**.

Fig. 1A. Overall survival estimates for MYD88 and CXCR4 genotyped patients with WM.

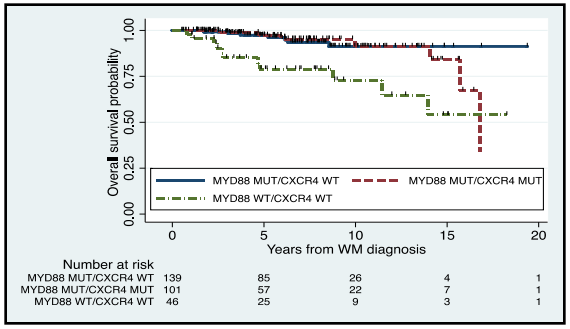


Table 2. Univariate and multivariate Cox proportional hazard regression models for overall survival in WM.

	Univariate analysis		Multivariate analysis	
	HR (95% CI)	p-value	HR (95% CI)	p-value
Age ≥65 years	2.51 (1.13-5.54)	0.02	1.79 (0.64-4.98)	0.27
Male sex	1.20 (0.53-2.69)	0.67	1.45 (0.49-4.29)	0.50
Hemoglobin <11.5 g/dL	0.66 (0.30-1.44)	0.30	2.92 (0.82-10.4)	0.10
Serum IgM ≥4,000 mg/dL	1.32 (0.59-2.96)	0.51	2.34 (0.82-6.67)	0.11
Bone marrow ≥50%	1.22 (0.54-2.76)	0.63	0.88 (0.31-2.45)	0.80
Serum B2M ≥3 mg/dL	1.99 (0.85-4.66)	0.11	2.24 (0.82-6.12)	0.12
MYD88^{MUT}	4.24 (1.94-9.24)	<0.001	7.47 (2.27-24.6)	<0.001
CXCR4^{MUT}	0.78 (0.33-1.81)	0.56	1.07 (0.36-3.12)	0.91

Transformation to diffuse large B-cell lymphoma (DLBCL) occurred during the follow-up period in 7 (15.2%) and 2 (0.76%) of *MYD88^{WT}* and *MYD88^{MUT}* patients, respectively. The incidence of transformation to DLBCL at 10 and 20 years was 1% (95% CI 0.1-5%) and 8% (1-39%) in *MYD88^{MUT}* patients versus 20% (95% CI 8-45%) and 29% (95% CI 12-58%) in *MYD88^{WT}* patients (HR for DLBCL transformation 19.8, 95% CI 4.08-95.8, p<0.001; **Figure 1B**). Overall survival estimates at 10 and 20 years for WM patients who did not transform to DLBCL were 88% (95% CI 81-93%) and 66% (95% CI 40-83%), respectively, and for WM patients who transformed to DLBCL were 63% (95% CI 23-86%) and 42% (7-75%), respectively (Mantel-Byar p=0.003; **Figure 1C**).

Fig. 1B. Incidence of DLBCL according to MYD88 mutational status.

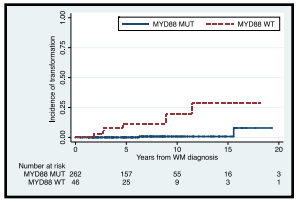
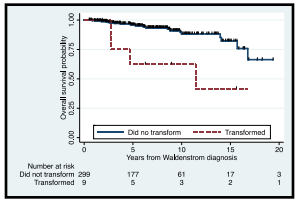


Fig. 1C. Overall survival for MYD88^{WT} patients with and without a DLBCL event.



Conclusions

WM patients with MYD88^{WT} disease had a high incidence of associated DLBCL events and significantly shorter survival versus those with MYD88^{MUT} disease. DLBCL events in MYD88^{WT} patients were associated with shortened survival.