

Clinical and diagnostic relevance of NOTCH2 and KLF2 mutations in splenic marginal zone lymphoma

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Table 1. Supplementary

Primers used for PCR and Sanger sequencing of the *NOTCH2* and *KLF2* genes

Gene	Exon	Primer name	Sequence (5'-3')	Amplicon size (bp)	PCR Condition
<i>NOTCH2</i>	34	NOTCH2_AF	CAGTGCCAAGAGTACCATGC	185	65°C
		NOTCH2_AR	GAGAGGATGTGGTGTCCGAA		
<i>NOTCH2</i>	34	NOTCH2_BF	TTGCTATCCCACCACCACAT	289	65°C
		NOTCH2_BR	CCGCTGGTTGGGCAATACT		
<i>NOTCH2</i>	34	NOTCH2_CF	CAGCCTCAGTCCACCTGC	341	65°C
		NOTCH2_CR	CAGTCAGAAGCAGAGTGGGG		
<i>KLF2</i>	1	KLF2.1_1F	GCCACTCACCGGTGTCC	196	60°C
		KLF2.1_1R	GCAGCCCATGTTCTACTACC		
<i>KLF2</i>	2	KLF2.2_1 F	TAGGGTGGTAAAAGGCAAGC	287	55°C
		KLF2.2_2 R	GCAGCAGCTCAGACACCAG		
<i>KLF2</i>	2	KLF2.2_4Fnew	CCTGGTGTCTGAGCTGCTG	218	60°C
		KLF2.2_4Rnew	GACCTCGCATGCACGAAG		
<i>KLF2</i>	2	KLF2.2_5Fnew	CTTCGTGCATGCGAGGTC	201	55°C
		KLF2.2_5Rnew	CGTCGAAGAGACCGAAGG		
<i>KLF2</i>	2	KLF2.2_6F	ACACACCGCCGCTCAG	349	55°C
		KLF2.2_7R	TGCGAACTCTTGGTGTAGGT		
<i>KLF2</i>	2	KLF2.2_7F	CCCAGCCTTCGGTCTCTTC	331	55°C
		KLF2.2_8R	AAATTCATTTTCTGGCGCGC		
<i>KLF2</i>	3	KLF2.3_1F	GGGGAAGTACGCTTACTCT	340	55°C
		KLF2.3_2R	CTCTGTAGCCACGCTGTGC		

Table 2. Supplementary

ID	NOTCH2			Second variation NOTCH2			KLF2			Somatic status
	Nucleotide Change	aa Change	Mutation type	Nucleotide Change	aa Change	Mutation type	Nucleotide Change	aa Change	Mutation type	
SMZL20	c.7075C>G	p.P2359A	Missense				WT			N/A
SMZL46	c.7075C>G	p.P2359A	Missense				WT			N/A
SMZL70	c.7075C>G	p.P2359A	Missense				--			N/A
SMZL71	c.6449delCT	p.P2150fs*1	Frameshift deletion	c.7075C>G	p.P2359A	Missense	--			Yes
SMZL77	c.7075C>G	p.P2359A	Missense				WT			N/A
SMZL18	c.7198C>T	p.R2400*	Non sense				c.823-825 del 3pb	p.S275delS	In Frame deletion	N/A
SMZL62	c.7198C>T	p.R2400*	Non sense				--			N/A
SMZL25	c.7198C>T	p.R2400*	Non sense				WT			Yes
SMZL32	c.7198C>T	p.R2400*	Non sense				WT			Yes
SMZL50	c.7223T>A	p.L2408H	Missense				--			N/A
SMZL83	c.6909delC	p.I2304fs*2	Frameshift deletion	c.7223T>A	p.L2408H	Missense	--			N/A
SMZL34	c.6449delCT	p.P2150fs*1	Frameshift deletion				WT			Yes
SMZL76	c.7165C>T	p.Q2389*	Non sense	c.7064C>A	p.T2355N	Missense	c.81 G>A	p.W27*	Non sense	N/A
SMZL10	c.6344_6358del 14pb	p.S2115fs*9	Frameshift deletion				WT			Yes
SMZL6	WT						c.988-997 del 10pb	p.R330fs*19	Frameshift deletion	N/A
SMZL3	WT						c.76 del 1pb	p.R26fs*115	Frameshift deletion	N/A
SMZL38	WT						c.815-877 del 63pb	p.H272del21AA	In Frame deletion	Yes
SMZL59	WT						c.560_570 del 11pb	p.P187fs*109	Frameshift deletion	N/A
SMZL28	WT						c.798 del 1pb	p.R266fs*24	Frameshift deletion	N/A
SMZL37	WT						c.554-564 del 11pb	p.P185fs*111	Frameshift deletion	N/A
SMZL4	WT						c.976 C>T	p.H326Y	Missense	N/A
SMZL1	WT						c.862 C>T	p.H288Y	Missense	N/A

Table 3. Supplementary

Clinical findings	N°
Patients SMZL	84
Age at diagnosis, mean (range)	64 (32-98)
Sex (male/female)	25/51
Hemoglobin mean (g/dL)	10,9 (range 7-16.7)
White blood cell median (x10 ⁹ /L)	9 (range 0.12-84)
Lymphocyte count (x10 ⁹ /L)	8 (range 0.3-75)
Platelet median (x 10 ⁹ /L)	171 (range 0.5-761)
Lymph node involvement	19/47 (40%)
Treatment at diagnosis (splenectomy and/or chemotherapy)	46/76 (60%)
Splenectomy	43/46
Chemotherapy	3/46
Treatment at any moment	76/80 (95%)
Splenectomy	70/78
Chemotherapy	30/60
Large B cell transformation	6/78 (8.3%)
Patients assessable for OS	77
Dead	25/77 (33,3%)
Patients assessable for EFS	67
Event free survival (clinical progression, death, LBCL)	29/67 (43.2%)
Patients assessable for TFS	75
Treatment-free survival (months)	16,8 (range 1-163)