SUPPLEMENTARY MATERIAL

A compendium of *DIS3* mutations and associated transcriptional signatures in plasma cell dyscrasias

SUPPLEMENTARY MATERIALS AND METHODS

Mutation analyses

Genomic DNA was extracted using Wizard genomic purification DNA kit (Promega Corporation, Madison, WI, USA) according to manufacturer's instructions, spectrophotometrically quantified and amplified using FastStart High Fidelity Polymerase (Roche) and fusion primers (Roche) containing M13 adapter sequences and the sequence-specific primers (Supplementary Table S6) spanning *DIS3* exons 1–4 for the PIN domain and exons 10–18 for the RNB domain (RefSeq NM_014953.4, representing the longer transcript encoding the longest protein isoform).

Amplicon library A and B sequencing adapters and multiplex identifier (MID) tags were then added to both tails of amplicons by a second amplification step. PCR conditions were as follows: in the first amplification step, denaturation step at 94°C for 5 min. followed by 25 cycles at 94°C (30 sec. per cycle), annealing step at 53°C (for exon 1), 57°C (for exons 10, 11 and 17), or 58°C (for the remaining exons) (30 sec. per cycle), and extension at 72°C (45 sec. per cycle), followed by a final extension at 72°C for 7 min.; in the second amplification step, denaturation step at 94°C for 5 min. followed by 25 cycles at 94°C (20 sec. per cycle), annealing step at 55°C (20 sec. per cycle), and extension at 72°C (45 sec. per cycle), followed by a final extension at 72°C for 10 min.. PCR products were visualized on agarose gel, purified using AMPure XP DNA-binding paramagnetic beads (Agencourt Bioscience Corp., Beckman Coulter S.p.A, Milan, Italy), and quantified using picogreen dye (Life Technologies, Carlsbad, California) and the Victor X2 (Perkin Elmer, Waltham, Massachusetts) fluorometer. Samples were then pooled together at equimolar ratios to prepare for Roche/454 pyrosequencing. The obtained amplicon library was added to the emulsion PCR at a ratio of 0.8 molecules per bead and subjected to deep sequencing on the Genome Sequencer Junior instrument (Roche-454 Life Sciences). The obtained sequencing reads were mapped to the *DIS3* human reference sequence (RefSeq NC_000013.10) and analyzed by the Amplicon Variant Analyzer (AVA) software version 3.0 (Roche-454 Life Sciences) to establish the mutant allele frequency.

To verify the occurrence of *DIS3* variants at transcriptional level, total RNA of mutated samples was converted to cDNA using M-MLV reverse transcriptase (Invitrogen Life Technologies, Carlsbad, California) and random hexamers, and subjected to deep sequencing of the exon harboring the variant/s detected on genomic DNA. Sequence-specific exonic PCR primers were designed in the Primer 3 program (http://frodo.wi.mit.edu/primer3/) and are reported in Supplementary Table S7.

Gene expression profiling and data analysis

For gene expression profiling (GEP), samples were profiled on the GeneChip Human Gene 1.0 ST array (Affymetrix, Santa Clara, CA, USA), as previously described [1]. Briefly, the raw intensity expression values were processed by Robust Multi-array Average procedure (RMA) [2], with the re-annotated Chip Definition Files from BrainArray libraries version 18.0.0 [3], available at http://brainarray.mbni.med.umich.edu. Supervised analyses were performed using Significant Analysis of Microarrays software (SAM version 4.00; Excel front-end publicly available at http://www-stat.stanford.edu/tibs/SAM/index.html) [4] as previously described [5]. The cutoff point for statistical significance (q-value < 10%) was determined by tuning the Δ parameter on the false discovery rate (FDR) and controlling the q-value of the selected probes. Differentially expressed genes were also evaluated at higher stringency level (median FDR 0%). The functional annotation analysis on the selected lists was performed by means of DAVID 6.7 tool (http://david.abcc.ncifcrf.gov/) and the ToppFun option of ToppGene Suite (https://toppgene.cchmc.org/), using the default parameters.

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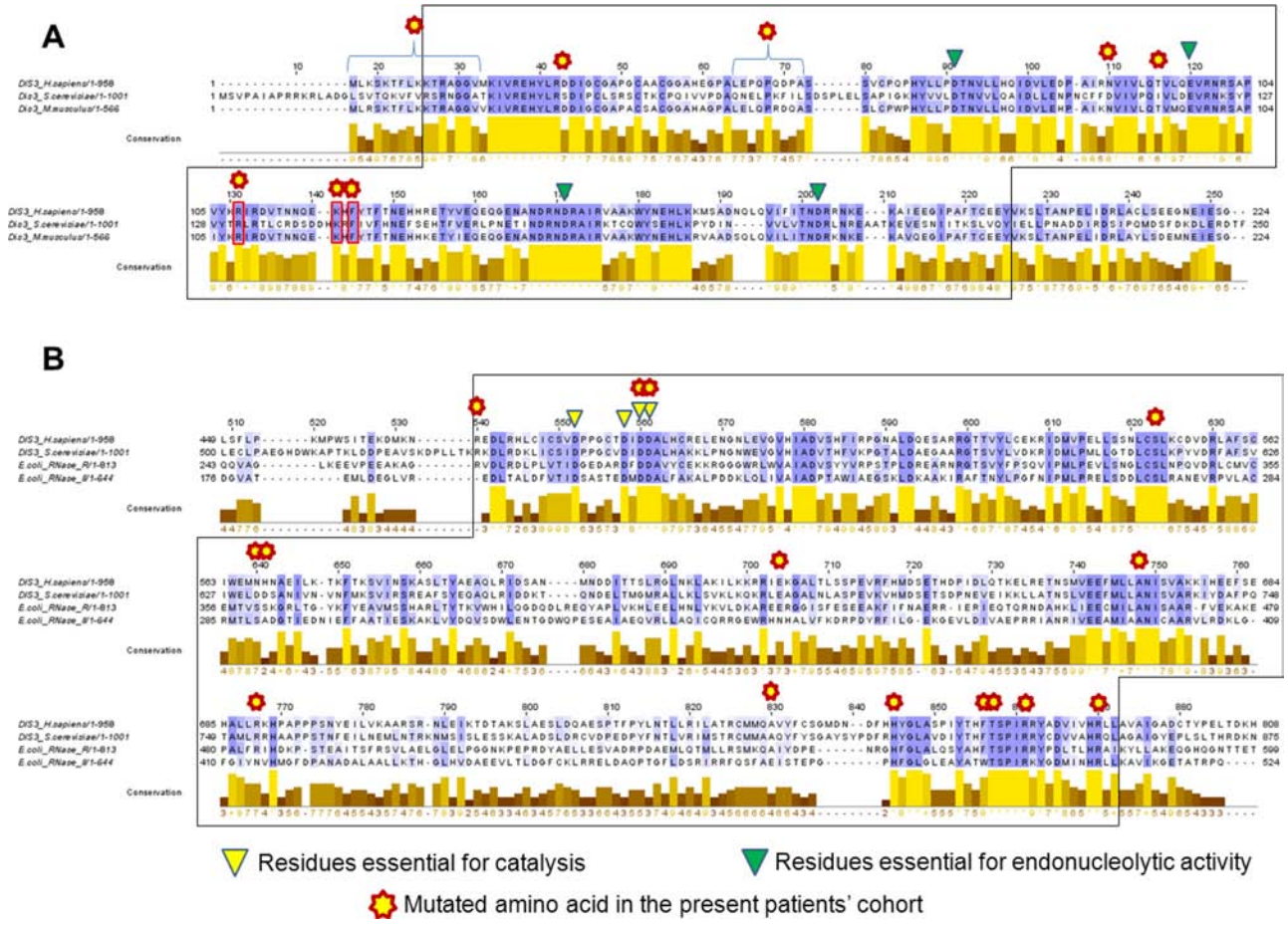
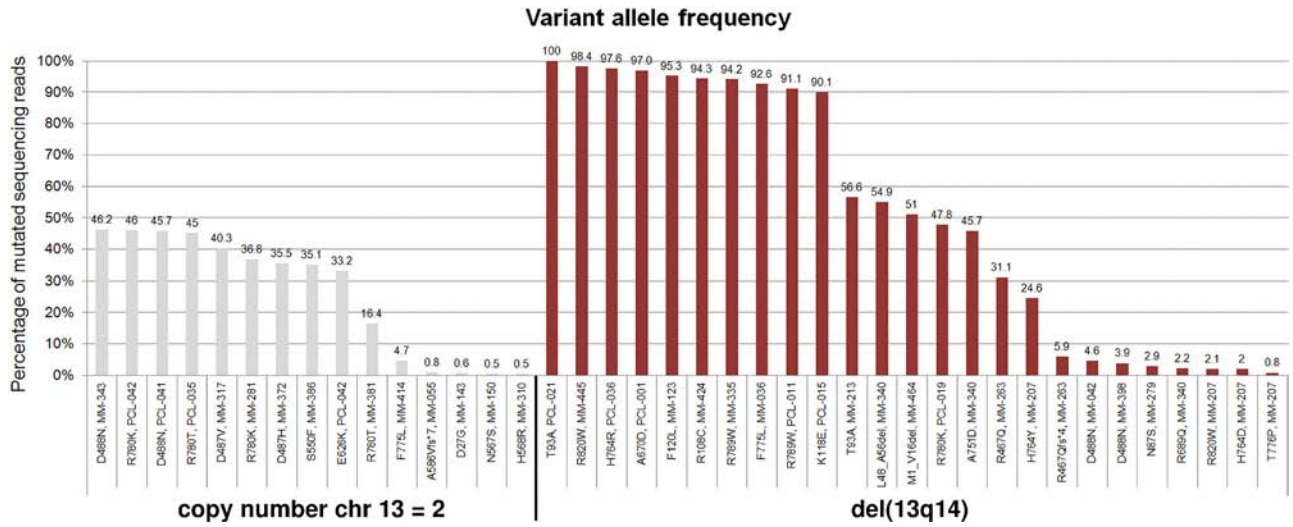
SUPPLEMENTARY FIGURES AND TABLES



Supplementary Figure S1: Detailed amino acid sequence alignment of DIS3 proteins of various species. In particular, human, yeast and murine DIS3 PIN domains were compared **(A),** and human and yeast RNB domains were aligned with *E.coli* RNAses II and R **(B).** Each domain is bordered by a black frame. Stars indicate genic positions targeted by missense mutations or in-frame deletions in the present primary patients' series. Green triangles indicate acidic residues predicted to coordinate a divalent cation and essential for endonucleolytic activity. Yellow triangles indicate residues involved in binding the magnesium ion at the active site. Sequence alignment was performed by Jalview software v2.7.



Supplementary Figure S2: Percentage of variant *DIS3* sequencing reads on total sequencing reads for the non-synonymous somatic mutations identified by NGS analysis in primary patients. Horizontal axis: sample id and carried amino acid variant are reported; patients are ordered according to copy number of chromosome 13 and decreasing mutation load.

Supplementary Table S1. Prediction of the functional relevance of the 26 missense *DIS3* mutations found in the present primary patients' cohort

AA change	Polyphen2	Mutation Taster	Mutation Assessor [FI score]	SIFT PREDICTION (SIFT score)	LRT (Likelihood Ratio test)
D27G	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (MEDIUM) [3.165]	DAMAGING (0.01)	DELETERIOUS
N87S	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (MEDIUM) [2.98]	DAMAGING (0.03)	DELETERIOUS
T93A	POSSIBLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (MEDIUM) [3.34]	DAMAGING (0)	DELETERIOUS
R108C	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (MEDIUM) [3.45]	DAMAGING (0)	NEUTRAL
K118E	POSSIBLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (MEDIUM) [3.24]	DAMAGING (0.01)	NEUTRAL
F120L	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (MEDIUM) [3.065]	DAMAGING (0.05)	DELETERIOUS
R467Q	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.675]	DAMAGING (0)	DELETERIOUS
D487H	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.74]	DAMAGING (0)	DELETERIOUS
D487V	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.74]	DAMAGING (0)	DELETERIOUS

(*Continued*)

AA change	Polyphen2	Mutation Taster	Mutation Assessor [FI score]	SIFT PREDICTION (SIFT score)	LRT (Likelihood Ratio test)
D488N	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.74]	DAMAGING (0)	DELETERIOUS
S550F	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.775]	DAMAGING (0)	DELETERIOUS
N567S	BENIGN	DISEASE CAUSING	PREDICTED NON-FUNCTIONAL (NEUTRAL) [-0.345]	TOLERATED (0.19)	DELETERIOUS
H568R	BENIGN	DISEASE CAUSING	PREDICTED NON-FUNCTIONAL (NEUTRAL) [-0.455]	TOLERATED (0.26)	NEUTRAL
E626K	BENIGN	DISEASE CAUSING	PREDICTED NON-FUNCTIONAL (LOW) [0.835]	TOLERATED (0.63)	DELETERIOUS
A670D	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.74]	DAMAGING (0)	DELETERIOUS
R689Q	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.77]	DAMAGING (0)	DELETERIOUS
A751D	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.77]	DAMAGING (0)	DELETERIOUS
H764D	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.77]	DAMAGING (0)	DELETERIOUS

(*Continued*)

AA change	Polyphen2	Mutation Taster	Mutation Assessor [FI score]	SIFT PREDICTION (SIFT score)	LRT (Likelihood Ratio test)
H764R	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.77]	DAMAGING (0)	DELETERIOUS
H764Y	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.77]	DAMAGING (0)	DELETERIOUS
F775L	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.77]	DAMAGING (0)	DELETERIOUS
T776P	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.77]	DAMAGING (0)	DELETERIOUS
R780K	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.77]	DAMAGING (0)	DELETERIOUS
R780T	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.77]	DAMAGING (0)	DELETERIOUS
R789W	POSSIBLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (HIGH) [4.77]	DAMAGING (0)	DELETERIOUS
R820W	PROBABLY DAMAGING	DISEASE CAUSING	PREDICTED FUNCTIONAL (MEDIUM) [3.275]	DAMAGING (0)	DELETERIOUS

Supplementary Table S2. Clinical and molecular characteristics of the 33 MM/PCL patients carrying *DIS3* non-synonymous mutations or indels

Sample name	Disease stage†	Variant*	Mutated reads (%)	AA change	del (13)	del (17p)	1q gain	1p loss	t(4;14)	t(11;14)	t(14;16)	t(14;20)	t(6;14)	HD‡
MM-036	MM	73336078A>C	92.57%	F775L	+	−	+	nd	−	−	−	−	−	−
MM-042	MM	73346338C>T	4.57%	D488N	+	−	+	−	+	−	−	−	−	+
MM-055	MM	73343049_ 73343050insA	0.83%	A586Vfs*7	−	−	−	nd	−	+	−	−	−	−
MM-123	MM	73355010G>C	95.30%	F120L	+	−	+	−	+	−	−	−	−	−
MM-143	MM	73355891T>C	0.62%	D27G	−	−	+	−	−	−	−	−	−	+
MM-150	MM	73345097T>C	0.51%	N567S	−	−	+	−	−	−	−	−	−	+
MM-207	MM	73336113G>A 73335837G>A 73336113G>C 73336077T>G	24.58% 2.05% 1.95% 0.79%	H764Y R820W H764D T776P	+	−	+	−	−	−	−	−	+	−
MM-213	MM	73355093T>C	56.57%	T93A	+	−	−	−	−	+	−	−	−	−
MM-263	MM	73346400C>T 73346400del	31.14% 5.93%	R467Q R467Qfs*4	+	−	+	−	+	−	−	−	−	−
MM-279	MM	73355110T>C	2.93%	N87S	+	−	+	−	−	−	−	nd	nd	nd
MM-281	MM	73336064C>T	36.80%	R780K	−	−	−	−	−	−	−	−	−	−
MM-310	MM	73345094T>C	0.50%	H568R	−	−	−	−	−	+	−	−	−	−
MM-335	MM	73335930G>A	94.20%	R789W	+	−	−	−	−	−	+	−	−	−

(*Continued*)

Sample name	Disease stage†	Variant*	Mutated reads (%)	AA change	del (13)	del (17p)	1q gain	1p loss	t(4;14)	t(11;14)	t(14;16)	t(14;20)	t(6;14)	HD‡
MM-340	MM	73355804_73355830del 73336151G>T 73337650C>T	54.86% 45.71% 2.2%	L48_A56del A751D R689Q	+	−	−	−	−	+	−	−	−	−
MM-343	MM	73346338C>T	46.18%	D488N	−	−	−	−	−	+	−	−	−	−
MM-372	MM	73346341C>G	35.48%	D487H	−	−	+	−	−	+	−	−	−	−
MM-381	MM	73336064C>G	16.38%	R780T	−	−	−	−	−	−	−	nd	nd	nd
MM-386	MM	73345240G>A	35.11%	S550F	−	−	nd	nd	−	nd	nd	nd	nd	nd
MM-398	MM	73346338C>T	3.85%	D488N	+	−	+	−	−	+	−	−	−	−
MM-414	MM	73336078A>C	4.71%	F775L	−	−	−	−	−	+	−	−	−	−
MM-424	MM	73355048G>A	94.33%	R108C	+	−	−	+	−	−	−	−	−	+
MM-445	MM	73335837G>A	98.36%	R820W	+	−	+	−	−	+	−	−	nd	−
MM-464	MM	73355968C>T	50.99%	M1_V16del	+	−	+	−	−	−	−	−	−	−
MM-317	MM	73346340T>A	40.3%	D487V	−	−	+	−	−	+	−	−	−	−
PCL-001	pPCL	73337707G>T	97%	A670D	+	−	nd	nd	−	−	−	+	−	nd
PCL-015	pPCL	73355018T>C	90.05%	K118E	+	−	+	−	−	−	+	−	−	−
PCL-019	pPCL	73336064C>T	47.75%	R780K	+	+	+	+	−	−	+	−	−	−
PCL-021	pPCL	73355093T>C	100%	T93A	+	−	+	−	+	−	−	−	−	−

(*Continued*)

Sample name	Disease stage†	Variant*	Mutated reads (%)	AA change	del (13)	del (17p)	1q gain	1p loss	t(4;14)	t(11;14)	t(14;16)	t(14;20)	t(6;14)	HD‡
PCL-035	pPCL	73336064C>G	45.04%	R780T	–	–	–	–	–	+	–	–	–	–
PCL-036	pPCL	73336112T>C	97.59%	H764R	+	–	–	–	–	+	–	–	–	–
PCL-011	sPCL	73335930G>A	91.1%	R789W	+	+	+	–	–	–	+	–	–	–
PCL-041	sPCL	73346338C>T	45.68%	D488N	–	–	nd	nd	–	nd	nd	nd	nd	nd
PCL-042	sPCL	73336064C>T 73342930C>T	45.95% 33.15%	R780K E626K	–	–	–	–	–	+	–	–	–	–

†MM: multiple myeloma; pPCL: primary plasma cell leukemia; sPCL: secondary plasma cell leukemia.
*Genomic positions based on hg19. ‡HD: presence of the hyperdiploid status on the basis of FISH evaluation criteria

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Supplementary Table S3. Mutational status of *DIS3* gene in 19 longitudinally analyzed patients

Sample name	Disease stage	*DIS3* status
MM-004	MM onset	wild type
	MM relapse	A827P, 97.1%
MM-146	MM onset	wild type
	MM relapse	wild type
MM-151	MM onset	wild type
	MM relapse	wild type
MM-200	MM onset	wild type
	MM relapse	wild type
MM-239	MM onset	wild type
	MM relapse	wild type
MM-263	MM onset	R467Q, 31.1%; R467Qfs*4, 5.9%
	MM relapse	R467Q, 81.9%
MM-271	MM onset	wild type
	MM relapse	wild type
MM-280	MM onset	wild type
	MM relapse	wild type
MM-282	MM onset	wild type
	MM relapse	wild type
MM-286	MM onset	wild type
	MM relapse	wild type
MM-327	MM onset	wild type
	MM relapse	wild type
MM-334	MM onset	wild type
	MM relapse	wild type
MM-340	MM onset	L48_A56del, 54.9%; A751D, 45.7%; R689Q, 2.2%
	MM relapse	L48_A56del, 61%; A751D, 42.3%
MM-429	MM onset	wild type
	MM relapse	wild type
MM-295	MM onset	wild type
	MM leukemic transformation	wild type
MM-281	MM onset	R780K, 36.8%
	MM leukemic transformation	R780K, 47.6%
PCL-026	pPCL onset	wild type
	pPCL relapse	wild type

(*Continued*)

Sample name	Disease stage	*DIS3* status
PCL-038	pPCL onset	wild type
	pPCL relapse	wild type
MM-442	MM leukemic transformation	wild type
	sPCL relapse	wild type

Supplementary Table S4. Modulated genes between *DIS3*- wild type and mutated patients

Gene	SAM score
LOC100132356	−7.27998
DKFZP434I0714	−7.24885
ILF3-AS1	−5.8891
FLJ30403	−5.73335
ARHGAP5-AS1	−5.63017
HEXA-AS1	−5.52727
C5orf54	−5.41888
C11orf71	−5.0731
ZNF594	−5.01186
FLJ38717	−4.90146
LOC100049716	−4.82276
CXorf21	−4.79455
LOC648987	−4.75759
TAPT1-AS1	−4.65361
C10orf111	−4.55991
APOBEC4	−4.54461
PRORSD1P	−4.35225
LINC00167	−4.34833
DDX60	−4.29767
CATSPER2P1	−4.21904
SPRYD4	−4.19583
POP1	−4.13861
PFN4	−4.08844
LINC00173	−3.94784
LOC153684	−3.93605
APOLD1	−3.92105
LOC100128398	−3.87278
NUDT9P1	−3.83567
C1orf220	−3.6607
RNU12	−3.64519
HTATSF1P2	−3.62834
DHX58	−3.61677
RNU11	−3.61058
CENPM	−3.58134
STPG2	−3.54494
FAM218A	−3.50969

(*Continued*)

Gene	SAM score
FREM3	−3.49656
FIGN	−3.47013
IFIT3	−3.46587
ZNF268	−3.46146
NS3BP	−3.40966
ANXA2R	−3.36509
LDOC1L	−3.3559
C21orf119	−3.34986
C11orf65	−3.33332
FLJ37201	−3.32968
LOC148696	−3.32702
DPM3	−3.30907
SLC4A5	−3.2872
LOC100506571	−3.28623
FLJ38576	−3.26721
PLA2G6	−3.24009
ZNRD1-AS1	−3.23349
PCDHGA1	−3.22484
PATL2	−3.21516
C9orf43	−3.21034
RNU5D-1	−3.20768
IL22RA1	−3.20672
C16orf54	−3.20079
SCIN	−3.1948
C5orf56	−3.18962
LINC00528	−3.17856
RBM45	−3.1685
GVINP1	−3.16364
KHDRBS2	−3.14396
MTG1	−3.1248
FAM154B	−3.12346
ST3GAL1	−3.12014
LOC100128288	−3.10885
C7orf13	−3.10235
TTC30B	−3.10072
CXXC4	−3.09534

(*Continued*)

Gene	SAM score
IFIT1	−3.08992
MIR320A	−3.08972
RNU4ATAC	−3.08157
PAPSS2	−3.07687
HSD17B7	−3.07454
MATN1-AS1	−3.0743
LRRC23	−3.05173
ZNF141	−3.04218
MAP2	−3.02161
MED31	−3.01969
SFR1	−3.01663
APOBEC3F	−3.01397
GDF9	−3.01336
MPZ	−2.99457
LOC645212	−2.98619
CDNF	−2.98353
C1orf74	−2.97908
TP53TG1	−2.97753
RASL11B	−2.97566
LOC100129726	−2.97513
GNRHR2	−2.97302
GPR135	−2.96435
OASL	−2.96341
CCDC87	−2.94704
HCG27	−2.93779
RNU5F-1	−2.92498
FAM227B	−2.91766
MIR23A	−2.91476
MIRLET7BHG	−2.91089
RIBC2	−2.8987
ZBTB26	−2.89665
PAN2	−2.89352
FZD7	−2.87535
CCDC85C	−2.87272
DLGAP1-AS1	−2.86802
HIST4H4	−2.85887

(*Continued*)

Gene	SAM score
KANSL1L	−2.85357
IFI6	−2.84554
EID2B	−2.83955
CCDC53	−2.83361
DDX60L	−2.82877
TTC23	−2.81725
ZNF70	−2.81337
COQ2	−2.81171
ODF3B	−2.80322
AGBL2	−2.79022
TMEM234	−2.77988

Supplementary Table S5. Clinical details of the 164 patients analyzed by NGS

Sample	*Sex*	*Age*	*Disease*	*Stage*[Δ]	*Phase*[‡]	*PP*[†]	*t(4;14)*	*t(11;14)*	*t(14;16)*	*t(14;20)*	*del(13)**	*del(17p)**	*1q gain**	*1p loss**	*HD°*
MM-004	F	58	MM	IA	D	Gκ	−	−	+	−	−	na	na	na	na
MM-015	M	71	MM	IIA	D	Gκ	−	+	−	−	−	−	−	−	−
MM-016	M	66	MM	IIIB	D	Gκ	−	−	−	−	−	−	+	−	+
MM-026	F	72	MM	IIIB	D	κ	−	+	−	−	−	−	−	na	na
MM-027	M	60	MM	IA	D	Gκ	−	−	−	−	+	−	na	na	na
MM-030	M	69	MM	IIIA	D	Gλ	−	−	−	−	−	−	−	+	+
MM-031	M	58	MM	IIIA	D	Aκ	−	+	−	−	−	−	+	+	−
MM-034	M	71	MM	IA	D	Gκ	−	−	−	−	−	−	−	−	+
MM-036	M	65	MM	IIA	D	Gκ	−	−	−	−	+	−	+	na	−
MM-037	F	50	MM	IIA	D	Gκ+Aκ	−	+	−	−	+	−	−	−	−
MM-038	F	67	MM	IIA	D	κ	−	−	−	−	+	−	−	−	+
MM-039	M	50	MM	IIA	D	Gλ	−	−	−	−	−	−	−	na	+
MM-042	M	54	MM	IIIA	D	Aλ	+	−	−	−	+	−	+	−	+
MM-043	F	73	MM	IA	D	Gκ+Gλ	−	−	−	−	−	−	+	−	−
MM-049	M	62	MM	IIIB	D	κ	−	−	−	−	−	−	−	−	+
MM-055	F	69	MM	IIIA	D	Gκ	−	+	−	−	−	−	−	na	−
MM-066	F	77	MM	IIIA	D	Aκ	+	−	−	−	−	−	−	na	−
MM-069	M	63	MM	IIA	D	Gκ	−	−	−	+	+	−	+	na	−
MM-078	F	59	MM	IIIA	D	κ	−	−	−	−	+	−	−	na	+
MM-079	F	74	MM	IIA	D	Gκ+Gλ	−	−	−	−	−	−	−	−	+
MM-087	F	84	MM	IIIA	D	Gλ	+	−	−	−	+	−	+	na	−
MM-115	F	53	MM	IIIA	D	Gλ	−	+	−	−	+	−	na	na	−
MM-123	M	55	MM	IIIA	D	Gκ	+	−	−	−	+	−	+	−	−
MM-131	M	73	MM	IA	D	Gκ	−	−	−	−	−	−	−	−	−
MM-143	M	61	MM	IIA	D	Gκ	−	−	−	−	−	−	+	−	+
MM-146	M	68	MM	IIA	D	Gλ	−	−	−	−	−	−	+	na	+

(*Continued*)

Sample	*Sex*	*Age*	*Disease*	*Stage*[Δ]	*Phase*[‡]	*PP*[†]	*t(4;14)*	*t(11;14)*	*t(14;16)*	*t(14;20)*	*del(13)**	*del(17p)**	*1q gain**	*1p loss**	*HD°*
MM-148	F	55	MM	IA	D	Aκ	–	–	–	–	–	+	+	na	+
MM-149	F	52	MM	IA	D	Gλ	–	–	–	–	–	–	–	–	+
MM-150	F	68	MM	IIA	D	Gλ	–	–	–	–	–	–	+	–	+
MM-151	F	71	MM	IA	D	Gλ	–	–	–	–	–	–	–	–	+
MM-152	M	66	MM	IA	D	Gκ	–	–	–	–	–	–	–	na	+
MM-154	F	71	MM	IIA	D	Gκ	–	–	+	–	+	–	+	–	–
MM-159	M	56	MM	IIA	D	κ	–	+	–	–	+	–	–	–	–
MM-174	M	85	MM	IIA	D	Aκ	–	–	–	–	–	–	–	–	+
MM-177	M	73	MM	IIIA	D	Gκ	–	–	–	–	+	–	+	–	–
MM-179	M	50	MM	IIIA	D	Gλ	–	+	–	–	–	+	–	–	+
MM-195	M	62	MM	IIA	D	Gκ	+	–	–	–	+	–	–	–	–
MM-200	F	63	MM	IA	D	Aλ	–	–	–	–	–	–	–	–	+
MM-202	M	64	MM	IIIA	D	Gκ	–	–	–	–	–	–	+	na	+
MM-206	F	73	MM	IIA	D	Gκ	+	–	–	–	+	–	–	+	–
MM-207	F	68	MM	IA	D	Aκ	–	–	–	–	+	–	+	–	–
MM-208	M	74	MM	IIA	D	Aλ	+	–	–	–	–	–	–	–	–
MM-209	F	65	MM	IIA	D	Gλ	–	–	–	–	–	–	–	–	+
MM-210	M	65	MM	IIB	D	Aλ	–	–	–	–	–	–	–	–	–
MM-212	F	55	MM	IIIA	D	Gκ	–	+	–	–	–	–	–	–	–
MM-213	M	66	MM	IIIA	D	λ	–	+	–	–	+	–	–	–	–
MM-219	M	73	MM	IIIA	D	Aλ	–	–	–	–	+	–	–	–	+
MM-224	F	52	MM	IIIA	D	Gκ	–	–	+	–	–	–	+	–	–
MM-229	M	75	MM	IIA	D	Gκ	–	–	–	–	–	–	–	–	–
MM-238	M	58	MM	IIB	D	Gκ	–	–	–	–	+	–	+	–	–
MM-239	F	72	MM	IIA	D	Aκ	–	–	–	–	+	–	–	–	+

(*Continued*)

Sample	*Sex*	*Age*	*Disease*	*Stage*[Δ]	*Phase*[‡]	*PP*[†]	*t(4;14)*	*t(11;14)*	*t(14;16)*	*t(14;20)*	*del(13)**	*del(17p)**	*1q gain**	*1p loss**	*HD°*
MM-240	M	70	MM	IA	D	Gλ	−	−	−	−	−	−	−	−	+
MM-241	M	54	MM	IIA	D	Gκ	−	−	−	−	−	−	−	−	+
MM-242	M	69	MM	IIIA	D	Aκ	−	−	−	−	+	−	−	−	+
MM-243	F	68	MM	IIA	D	Gκ	−	−	−	−	−	−	+	−	+
MM-246	M	71	MM	IIIA	D	Aλ	−	+	−	−	−	−	−	−	−
MM-252	F	77	MM	IIA	D	Aλ	−	+	−	−	−	−	+	−	−
MM-256	M	58	MM	IIIA	D	Aκ	−	−	+	−	+	−	−	−	−
MM-261	F	66	MM	IIB	D	Gκ	−	−	−	−	+	−	+	−	+
MM-262	M	74	MM	IA	D	λ	−	−	−	−	−	−	+	−	−
MM-263	F	65	MM	IIB	D	κ	−	−	−	−	+	−	+	−	−
MM-267	M	74	MM	IIIA	D	Gλ	−	−	−	−	+	−	−	−	+
MM-268	M	77	MM	IIIB	D	Gκ	−	−	−	−	−	−	−	−	−
MM-269	F	67	MM	IIA	D	Gκ	−	−	−	−	+	−	−	−	+
MM-271	M	76	MM	IA	D	Gκ	−	−	−	−	+	−	−	−	−
MM-274	M	59	MM	IIA	D	Gκ	+	−	−	−	−	−	−	−	−
MM-276	F	70	MM	IIB	D	Gκ	+	−	−	−	+	−	−	−	−
MM-278	M	73	MM	IIA	D	Gλ	−	−	−	−	−	−	−	+	+
MM-279	F	71	MM	IIIA	D	Gλ	−	−	−	na	+	−	+	−	na
MM-280	M	62	MM	IIIA	D	Gλ	−	+	−	−	−	−	−	−	−
MM-281	F	77	MM	IIA	D	Gκ	−	−	−	−	−	−	−	−	−
MM-282	M	66	MM	IIIA	D	Gκ	−	−	−	−	−	−	+	−	+
MM-284	M	49	MM	IIIA	D	κ	−	+	−	−	na	−	−	−	−
MM-286	F	71	MM	IIA	D	Gκ	−	−	−	−	+	−	−	−	−
MM-295	F	74	MM	IIIA	D	λ	−	−	−	−	+	−	na	na	na
MM-300	M	65	MM	IIB	D	Gκ	−	−	−	−	−	−	na	na	na
MM-301	F	72	MM	IIA	D	Aλ	−	−	−	−	−	−	+	−	+
MM-302	M	65	MM	IA	D	Aκ	−	−	−	−	−	−	+	−	+

(*Continued*)

Sample	*Sex*	*Age*	*Disease*	*Stage*[Δ]	*Phase*[‡]	*PP*[†]	*t(4;14)*	*t(11;14)*	*t(14;16)*	*t(14;20)*	*del(13)**	*del(17p)**	*1q gain**	*1p loss**	*HD°*
MM-308	M	58	MM	IA	D	Aκ	−	−	−	−	−	−	−	−	−
MM-313	M	66	MM	IIIA	D	Gλ	−	+	−	−	−	−	−	−	−
MM-310	M	67	MM	IIIA	D	Gλ	−	+	−	−	−	−	−	−	−
MM-314	F	70	MM	IIIA	D	Aλ	−	+	−	−	−	−	+	−	−
MM-317	M	56	MM	IIA	D	Gλ	−	+	−	−	−	−	+	−	−
MM-321	M	63	MM	IIIA	D	Gκ	−	−	−	−	+	−	−	−	+
MM-327	F	62	MM	IIA	D	absent	−	−	−	−	+	−	+	−	+
MM-330	F	61	MM	IIIA	D	Aκ	+	−	−	−	+	−	+	−	−
MM-334	F	45	MM	IA	D	Gλ	−	−	−	−	+	+	+	+	+
MM-335	F	68	MM	IIA	D	Aκ	−	−	+	−	+	−	−	−	−
MM-340	M	46	MM	IIIA	D	Gλ	−	+	−	−	+	−	−	−	−
MM-341	M	65	MM	IIIA	D	Gκ	−	−	−	−	−	−	+	−	−
MM-343	M	74	MM	IIIA	D	Aλ	−	+	−	−	−	−	−	−	−
MM-351	M	na	MM	IIA	D	Aκ	−	−	−	−	−	−	−	−	−
MM-362	F	80	MM	IIA	D	Gλ	−	−	−	−	+	+	+	−	−
MM-372	M	54	MM	IA	D	Gκ	−	+	−	−	−	−	+	−	−
MM-375	F	78	MM	IA	D	Gκ	+	−	−	−	−	+	−	−	−
MM-381	F	69	MM	IA	D	Gκ	−	−	−	na	−	−	−	−	na
MM-382	M	85	MM	IIA	D	Gκ	−	−	−	−	+	+	na	na	na
MM-385	F	76	MM	IIIA	D	Gλ	−	−	−	−	−	−	−	−	+
MM-386	F	76	MM	IIA	D	Aλ	−	na	na	na	−	−	na	na	na
MM-387	F	44	MM	IIIA	D	Gκ	−	−	−	−	−	−	+	−	+
MM-392	F	53	MM	IA	D	Gλ	−	−	−	−	+	−	+	−	+
MM-398	F	65	MM	IIA	D	Gκ	−	+	−	−	+	−	+	−	−
MM-402	M	67	MM	IIA	D	Gκ	−	+	−	−	+	−	−	−	−
MM-405	M	69	MM	IA	D	Gκ	−	+	−	−	−	−	−	−	−
MM-406	M	62	MM	IIB	D	Aκ	−	+	−	−	−	−	+	−	na

(*Continued*)

Sample	*Sex*	*Age*	*Disease*	*Stage*[Δ]	*Phase*[‡]	*PP*[†]	*t(4;14)*	*t(11;14)*	*t(14;16)*	*t(14;20)*	*del(13)**	*del(17p)**	*1q gain**	*1p loss**	*HD°*
MM-407	F	79	MM	IIIA	D	λ	−	−	−	−	+	−	−	−	−
MM-410	F	79	MM	IIIA	D	λ	−	−	−	−	−	−	+	−	−
MM-411	M	67	MM	IIIA	D	Gλ	−	−	−	−	−	−	−	−	+
MM-413	F	51	MM	IIA	D	Gκ	−	−	−	−	+	−	+	−	+
MM-414	F	67	MM	IIIA	D	Gλ	−	+	−	−	−	−	−	−	−
MM-422	F	74	MM	IIB	D	κ	−	−	−	−	+	−	−	−	−
MM-423	F	53	MM	IIA	D	λ	+	−	−	−	+	−	−	−	−
MM-424	F	84	MM	IA	D	Gκ	−	−	−	−	+	−	−	+	+
MM-425	F	65	MM	IIA	D	Aλ	−	−	−	−	+	−	+	−	−
MM-428	F	65	MM	IIA	D	Aλ	−	−	−	−	−	−	+	+	−
MM-429	F	63	MM	IIIA	D	Gκ	−	−	−	−	−	−	+	−	+
MM-430	na	62	MM	IIIA	D	Gλ	−	−	−	−	−	−	−	−	−
MM-431	F	70	MM	IA	D	Gλ	+	−	−	−	+	−	+	−	−
MM-433	M	72	MM	IIB	D	κ	−	+	−	−	−	−	−	−	−
MM-434	F	53	MM	IIA	D	κ	−	−	−	−	+	−	−	+	−
MM-435	na	42	MM	IIIA	D	Gλ	−	−	−	−	−	−	−	−	+
MM-437	F	72	MM	IIB	D	λ	−	−	−	−	+	−	+	−	−
MM-440	na	61	MM	IIA	D	Aκ	−	−	−	−	−	−	−	+	+
MM-441	na	na	MM	IA	D	Gλ	−	−	−	−	−	−	−	−	−
MM-442	F	65	sPCL	/	D	Gκ	−	−	−	−	+	−	+	+	−
MM-445	na	65	MM	IIA	D	κ	−	+	−	−	+	−	+	−	−
MM-446	na	52	MM	IIIA	D	Gλ	−	−	−	−	−	−	+	−	+
MM-447	na	57	MM	IIA	D	Gκ	−	+	−	−	−	−	+	−	−
MM-448	na	59	MM	IIA	D	Aκ	−	−	−	−	+	−	+	−	−
MM-449	na	54	MM	IIA	D	Gκ	−	−	−	−	+	−	−	+	−
MM-464	M	75	MM	IIIA	D	Gλ	−	−	−	−	+	−	+	−	−
PCL-001	F	51	pPCL	/	D	κ	−	−	−	+	+	−	na	na	na

(*Continued*)

Sample	*Sex*	*Age*	*Disease*	*Stage*[Δ]	*Phase*[‡]	*PP*[†]	*t(4;14)*	*t(11;14)*	*t(14;16)*	*t(14;20)*	*del(13)**	*del(17p)**	*1q gain**	*1p loss**	*HD°*
PCL-002	F	69	sPCL	/	R	λ	−	+	−	−	+	na	−	na	na
PCL-004	M	72	pPCL	/	D	Gκ	−	−	+	−	−	−	−	−	−
PCL-005	M	76	sPCL	/	R	Aκ	−	+	−	−	−	−	−	−	−
PCL-008	M	57	pPCL	/	D	Gλ	−	−	+	−	+	−	−	na	na
PCL-009	F	77	sPCL	/	R	Gκ	+	−	−	−	+	−	+	−	−
PCL-011	M	76	sPCL	/	R	Gκ	−	−	+	−	+	+	+	−	−
PCL-012	F	62	sPCL	/	R	Aκ	−	−	+	−	−	+	−	−	−
PCL-014	M	72	pPCL	/	D	λ	−	+	−	−	+	+	−	+	−
PCL-015	M	78	pPCL	/	D	κ	−	−	+	−	+	−	+	−	−
PCL-016	F	57	pPCL	/	D	Gκ	−	−	+	−	+	−	+	+	−
PCL-017	F	68	pPCL	/	D	Gκ	−	−	+	−	+	+	+	+	−
PCL-018	F	59	pPCL	/	D	κ	−	+	−	−	+	+	−	−	−
PCL-019	F	67	pPCL	/	D	Mκ	−	−	+	−	+	+	+	+	−
PCL-020	F	79	pPCL	/	D	Gλ	−	−	−	−	−	−	+	−	−
PCL-021	M	48	pPCL	/	D	Gλ	+	−	−	−	+	−	+	−	−
PCL-023	M	60	pPCL	/	D	Gκ	−	−	+	−	+	+	+	+	−
PCL-026	F	59	pPCL	/	D	Gκ	−	−	+	−	+	−	+	−	−
PCL-027	M	65	pPCL	/	D	λ	−	+	−	−	−	+	−	−	−
PCL-028	F	57	pPCL	/	D	κ	−	+	−	−	+	−	−	−	−
PCL-029	M	51	pPCL	/	D	Aλ	−	−	+	−	−	−	−	−	−
PCL-030	F	52	pPCL	/	D	κ	−	−	−	+	+	+	−	−	−
PCL-031	F	59	sPCL	/	D	Gλ	−	−	+	−	−	+	+	−	−
PCL-032	M	51	pPCL	/	D	Gκ	+	−	−	−	+	−	+	−	−
PCL-035	F	76	pPCL	/	D	κ	−	+	−	−	−	−	−	−	−
PCL-036	M	72	pPCL	/	D	Gκ	−	+	−	−	+	−	−	−	−
PCL-037	M	72	pPCL	/	D	Aλ	−	+	−	−	+	−	−	−	−
PCL-038	M	57	pPCL	/	D	Gκ	−	−	−	−	+	−	+	+	+

(*Continued*)

Sample	*Sex*	*Age*	*Disease*	*Stage*[Δ]	*Phase*[‡]	*PP*[†]	*t(4;14)*	*t(11;14)*	*t(14;16)*	*t(14;20)*	*del(13)**	*del(17p)**	*1q gain**	*1p loss**	*HD*[°]
PCL-039	M	54	sPCL	/	D	Aκ	−	−	−	−	−	+	−	+	−
PCL-041	M	na	sPCL	/	D	Gκ	−	na	na	na	−	−	na	na	na
PCL-042	F	69	sPCL	/	D	Gλ	−	+	−	−	−	−	−	−	−
PCL-043	F	68	pPCL	/	D	Gλ	−	+	−	−	−	−	−	−	−
PCL-046	F	50	pPCL	/	D	κ	−	−	−	−	+	−	+	−	+

[Δ]The Durie clinical staging system was adopted; [‡]D: Diagnosis; R: relapse;
[†]Paraprotein; *del(13), del(17), 1p loss and 1q gain were determined by FISH.
[°]HD: presence of the hyperdiploid status on the basis of FISH evaluation criteria. *na*: not available.

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Supplementary Table S6. Genome-specific primers for amplicon library preparation

Exon	Amplicon localization (hg19, chromosome 13)	Primer FWD	Primer REV
1	73355681–73356065	5′- GAAAGGGAAGAACCTCCGGG-3′	5′- TCCCTGTCATACCCCCTTGT-3′
2	73354916–73355271	5′- GCTTCTTGGCTTAACTTATTCAGTG-3′	5′- ACAGGAACCCTCTCCCGAA-3′
3	73352200–73352586	5′- TGCTAAGAGTTTTCACATATCCTTG-3′	5′- TCACATGAAGTTATATAGGACTACGA-3′
4	73351452–73351809	5′- AGGCTGTAGTGATGTGTAATTGC-3′	5′- GCTTACCCACCGACATTCC-3′
10	73346062–73346470	5′- TATGTTGTAGTTGTGCTTTGGAAAT-3′	5′- CAATATGCTTGACTGGGTAAATGTA-3′
11	73345868–73346221	5′- TTTGCTTGTTAATTACTCTTGTGAAG-3′	5′- TTCATGGCCGTTTAAGAATC-3′
12–13	73345012–73345325	5′- TTATGGCTAAGTAATCTGTGGTTCTA-3′	5′-AAATTAGAGATTAATAGCCATGAAACG-3′
14	73342807–73343130	5′- GTAGTGAAAGTAGGAGGACATATTG-3′	5′- GAAGCTAGAAGAAACAGGAGTCT-3′
15	73339975–73340297	5′- ACACTTGCTGTAGTCATTGTCTT-3′	5′- GCAAGCCAAATAAAGTAGAAATCAT-3′
16	73337539–73337917	5′- GCGGAGTAACTGAGAGATGAAAG-3′	5′- CAGGTAGATCAAAACACAAATAGATG-3′
17	73335921–73336221	5′- GCCGAATCTCCTACTTTTCCA-3′	5′- CCAAAAGCCGATGAACAATGA-3′
18	73335757–73336105	5′- CTTAGCGTCTCCAATATACACACA-3′	5′- CTAGCAGTATCGACAAAAGGCA-3′

Supplementary Table S7. Sequence-specific exonic PCR primers

Primer FWD	Primer REV
5′- CCGGGGTTAGGCGTATTCTA -3′	5′- AAGTGAGAAATCGCAGTGCC -3′
5′- TACTTGCTGCCCGACACTAA -3′	5′- GGAATACCAGCTTTCACTTGTG -3′
5′- ATGTTCCCCATCAGCCTTTT -3′	5′- AAAATGTGACGTGGACAGGC -3′
5′- GGGAAATGAATCACAATGCTGA -3′	5′- TCTGAACATGCTCTGCTTCG -3′
5′- AGTCTTTGGATCAGGCCGAA -3′	5′- GGCTATTGGGGCTGACTGTA -3′
5′- ACTATGGCTTAGCGTCTCCA -3′	5′- AACGTGCATCAGTGGCTTTT-3′