Low levels of monoclonal small B cells in the bone marrow of patients with diffuse large B-cell lymphoma of activated B-cell type but not of germinal center B-cell type

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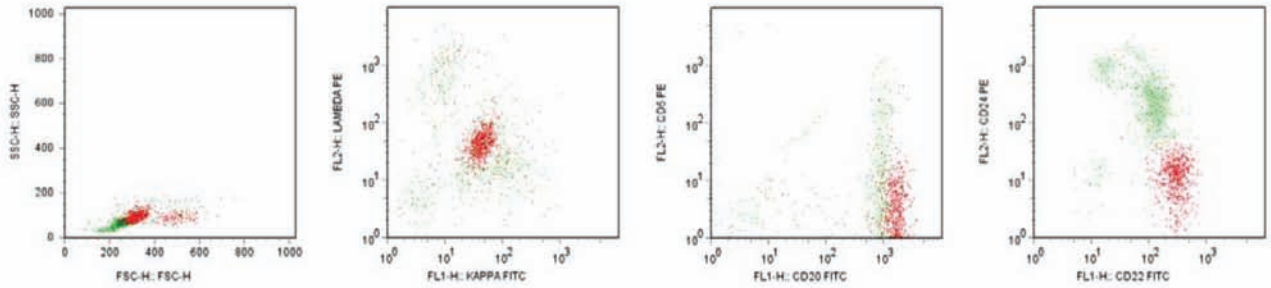
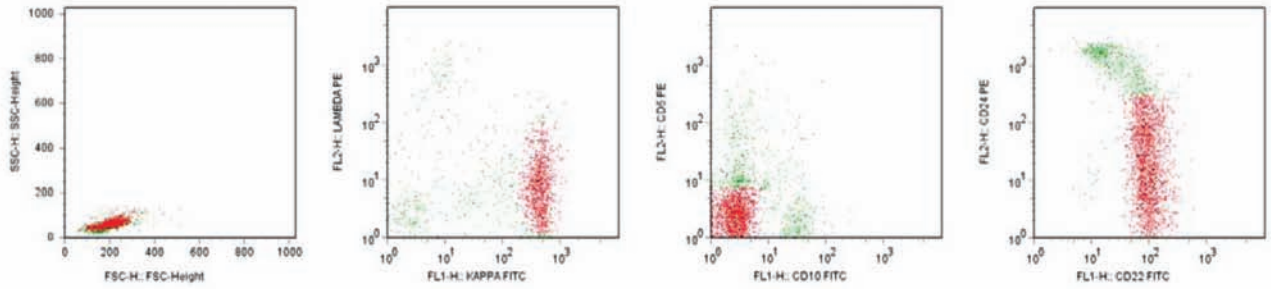
Online Supplementary Table S1. Clinical data of patients with marrow infiltration with DLBCL.

Diagnosis	Localization	% BM infiltration (biopsy)	% BM infiltration (flow cytometry)	Stage	IPI
T/HRBCL	Cervical LN	5%	negative	IVB	4
T/HRBCL	Spleen	90%	negative	IVB	2
DLBCL/NOS GCB	Bone	50%	negative	IVA	2
DHBCL	Abdomen	negative	0.5%	IVA	5
DHBCL	Axillar LN	80%	2%	IVB	5
IVBCL	Spleen/BM	10%	<0.1%	IVB	2
DLBCL/NOS ABC	Lung	30%	1%	IVB	5
POSTTR. DLBCL	Abdomen	10%	negative	IVB	5
DHBCL	Cervical LN	5%	< 1%	IVB	5
DLBCL/NOS ABC	Cervical LN	10-15%	0.1%	IVB	3
DHBCL	Abdomen	90%	30%	IVB	3

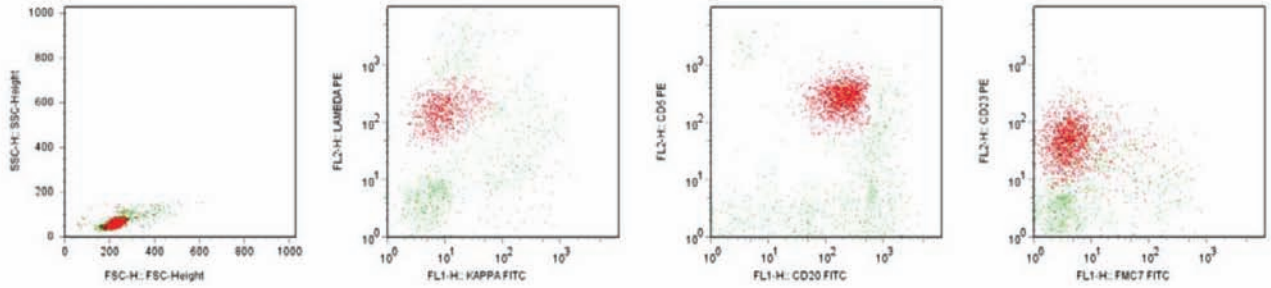
PATIENT 1



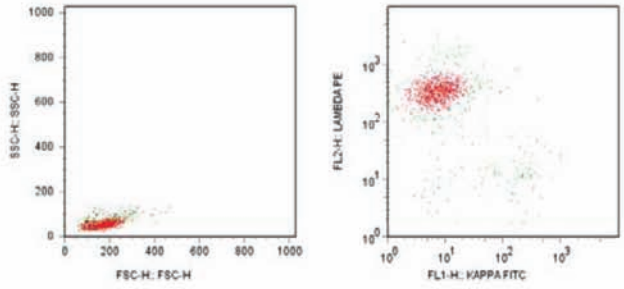
PATIENT 9



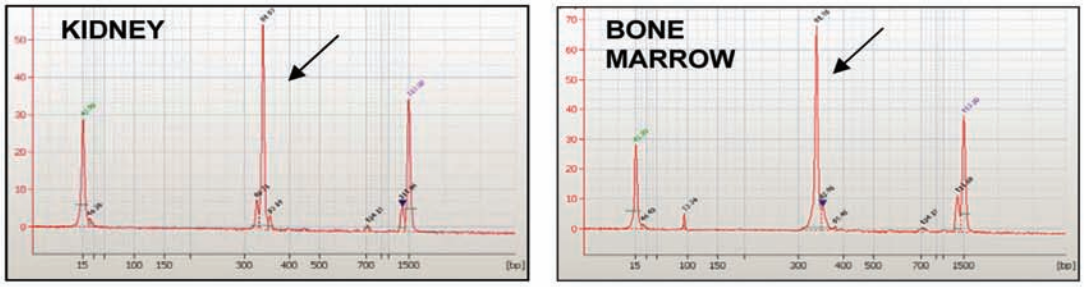
PATIENT 11



PATIENT 22



Online Supplementary Figure S1. Flow cytometry bivariate dot plots of patients 1, 9, 11 and 22, respectively. MSBC are represented by red dots. Immunophenotypic markers with their fluorochrome are indicated in the X and Y axes. Patient 1 shows MSBC without evident immunoglobulin light chain expression and without CD24 or CD5 expression. Patient 9 shows immunoglobulin κ light chain restriction and absent CD10 and CD5 expression. The MSBC of Patient 11 show immunoglobulin λ light chain restriction with expression of CD5 and CD23. Patient 22 shows MSBC with immunoglobulin λ light chain restriction. FSC: forward scatter; SSC: side scatter.

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                          <---------------------------------------- FR1 - IMGT
                         1                5                   10                  15

PT 11                    ... ... ... ... ... ... ... ... ... ... ... ... ... ... .ct
                          Q   V   Q   L   Q   E   S   G   P       G   L   V   K   P
IGHV4-30-4*01            cag gtg cag ctg cag gag tcg ggc cca ... gga ctg gtg aag c--

                         ----------------------------------------> _________________
                                        20                  25                  30
                          S   E   T   L   S   L   T   C   S   V   S   G   G   S   I
PT 11                    tcg gag acc ctg tcc ctc acc tgc agt gtc tct ggt ggc tcc atc
                              Q                           T
IGHV4-30-4*01            --a c-- --- --- --- --- --- --- -c- --- --- --- --- --- ---

                         __  CDR1 - IMGT _____________ <--------------------------
                                        35                  40                  45
                          S           S   G   D   Y   Y   W   S   W   I   R   Q   P
PT 11                    agc ... ... agt ggt gat tac tac tgg agt tgg atc cgc cag ccc

IGHV4-30-4*01            --- ... ... --- --- --- --- --- --- --- --- --- --- --- ---

                          FR2 - IMGT  ------------------------> _____________  CDR2
                                        50                  55                  60
                          P   G   K   G   L   E   W   I   G   H   I   Y   Y   S
PT 11                    cca ggg aag ggc ctg gag tgg att ggg cac atc tat tat agt ...
                                                            Y
IGHV4-30-4*01            --- --- --- --- --- --- --- --- --- t-- --- --- --c --- ...

                          - IMGT  __________ <---------------------------------------
                                        65                  70                  75
                                  G   S   T   Y   Y   N   P   S   L   K       S   R
PT 11                    ... ... ggg agc acc tac tac aac ccg tcc ctc aag ... agt cga

IGHV4-30-4*01            ... ... --- --- --- --- --- --- --- --- --- --- ... --- ---

                         -------------------------------  FR3 - IMGT  ---------------
                                        80                  85                  90
                          V   T   I   S   V   D   P   S   K   N   Q   F   S   L   K
PT 11                    gtt acc ata tca gta gac ccg tcc aag aac cag ttc tcc ctg aag
                                            T
IGHV4-30-4*01            --- --- --- --- --- --- a-- --- --- --- --- --- --- --- ---

                         ----------------------------------------------------------> ____
                                        95                  100                 104
                          L   S   S   V   T   D   A   D   T   A   V   Y   Y   C   A
PT 11                    ctg agc tct gtg act gac gca gac acg gcc gta tat tac tgt gcc
                                        A
IGHV4-30-4*01            --- --- --- --- --- -c- --- --- --- --- --g --- --- --- ---

                         ____________________  CDR3 -  IMGT ______________________

                          R   W   D   K   G   D   P   R   A   L   G   F   D   Y   W
PT 11                    agg tgg gac aag ggt gac cca aga gcc ttg ggc ttt gac tac tgg

IGHV4-30-4*01            --a ga


                          G   Q   G   T   L   V   T   V   S   S   G
CASE 11                  ggc cag gga acc ctg gtc acc gtc tcc tca ggt
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Online Supplementary Figure S2. PCR fragment analysis (top) of the amplified rearranged immunoglobulin heavy chain gene. The arrows indicate similar-sized fragments in both the DLBCL in the kidney and MSBC in the bone marrow of patient 11. The rearranged immunoglobulin heavy chain gene sequence of the MSBC and DLBCL (top sequence) is compared with the closest homologous germline gene sequence, IGHV-30-4.01 (bottom sequence). Only mutations with respect to the germline sequence are given. Eight mutations are seen, of which five are replacement mutations (replacement amino acids are given in capital letters above the nucleic acid mutations). The replacement mutations are all in the framework regions (FR) while silent mutations are seen in both FR and complementarity determining regions (CDR).