Clinical and biological features of B-cell neoplasms with CDK6 translocations: an association with a subgroup of splenic marginal zone lymphomas displaying frequent CD5 expression, prolymphocytic cells, and TP53 abnormalities

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Clinical and biological features of B-cell neoplasms with *CDK6* translocations: an association with a subgroup of splenic marginal zone lymphomas displaying frequent CD5 expression, prolymphocytic cells, and *TP53* abnormalities.

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Short-title: Mature B-cell neoplasms with *CDK6* translocations

SUMMARY

A translocation involving the *CDK6* gene [t(*CDK6*)] is a rare but recurrent abnormality in B-cell neoplasms. To further characterize this aberration, we studied 57 cases – the largest series reported to date. FISH analysis confirmed the involvement of *CDK6* in all cases, including t(2;7)(p11;q21) *IGK/CDK6* (n=51), t(7;14)(q21;q32) *CDK6/IGH* (n=2) and the previously undescribed t(7;14)(q21;q11) *CDK6*/*TRA*-*TRD* (n=4). Ten patients were diagnosed with chronic lymphocytic leukemia, monoclonal B-cell lymphocytosis or small lymphocytic lymphoma, and 47 had small B-cell lymphoma (SmBL) including 36 cases of marginal zone lymphoma (MZL: 34 splenic MZLs, 1 nodal MZL and 1 bronchus-associated lymphoid tissue lymphoma). Eighteen of the 26 cytologically reviewed cases of MZL (69%) had an atypical aspect with prolymphocytic cells. Among the 47 MZL/SmBL patients, CD5 expression was found in 26 (55%) and the *TP53* deletion in 22 (47%). The *TP53* gene was mutated in 10/30 (33%); the 7q deletion was detected in only one case, and no *NOTCH2* mutations were found. *IGHV* locus sequencing revealed that none harbored an *IGHV1-02*04* gene. Overall survival was 82% at 10 years and not influenced by *TP53* aberration. Our findings suggest that most t(*CDK6*)+ neoplasms correspond to a particular subgroup of indolent marginal zone B-cell lymphomas with distinctive features.

Key-words: CDK6, marginal zone lymphoma, *TP53*, prolymphocytic cell, CD5

INTRODUCTION

A translocation involving the *CDK6* gene is a rare but recurrent abnormality in mature B-cell neoplasms. Three different translocations have been described: t(2;7)(p11;q21) (the most frequent), t(7;14)(q21;q32) and t(7;22)(q21;q11), leading to juxtaposition of the *CDK6* gene with the *IGK*, *IGH* or *IGL* locus, respectively, and thus *CDK6* overexpression.

The t(2;7) was first reported by Vahdati et al. in 1983 in a patient with chronic lymphocytic leukemia (CLL) (Vahdati*, et al* 1983) and by Oscier et al. in 1993 in two cases of splenic lymphoma with villous lymphocytes (Oscier*, et al* 1993). Since then, about 40 cases of t(2;7) and its variants have been documented in patients with different diagnoses: splenic marginal zone lymphoma (SMZL) (Baro*, et al* 2008, Corcoran*, et al* 1999, Douet-Guilbert*, et al* 2014, Remstein*, et al* 2008, Salido*, et al* 2010, Xochelli*, et al* 2014), CLL (Cavazzini*, et al* 2008, Douet-Guilbert*, et al* 2014, Fink*, et al* 2006, Hayette*, et al* 2003), high-grade and low-grade B-cell lymphoma not otherwise specified (Chen*, et al* 2009), and monoclonal B-cell lymphocytosis (MBL) (Parker*, et al* 2011).

Given the rarity of *CDK6* translocations [t(*CDK6*)], the few studies published to date featured small numbers of patients. Hence, the associated clinical and biological features, genomic aberrations, and prognostic implications of this rare abnormality have not been fully characterized. Here, we describe the clinical, cytogenetic, and molecular features of 57 cases of B-cell neoplasms harboring a t(*CDK6*)– the largest such series reported to date.

METHODS

Patient selection

Databases from 18 French, Belgian and Canadian institutions (covering the period from 2000 and 2017) were retrospectively screened for patients with a mature B-cell neoplasm harboring a t(2;7)(p11;q21) or variants involving the *CDK6* gene. The patients' characteristics at diagnosis were recorded when available. If possible, the immunophenotype was scored according to Matutes et al. (Matutes and Catovsky 1994) or Moreau et al. (Moreau*, et al* 1997). Cytogenetic and molecular analyses were performed at diagnosis for 38 patients, during follow-up and before treatment for 16 (median time between diagnosis and sampling: 28.5 months), and at relapse for 3. The study was performed in accordance with the Declaration of Helsinki, and was approved by the local investigational review board (*CPP Ile-de-France VI*, Paris, France) on May 21th, 2014.

Karyotyping and FISH analysis

Standard chromosome banding analyses were used to obtain R-or G-banded chromosomes from peripheral blood (n=44), bone marrow (BM) (n=12) or lung biopsy (n=1) samples. Karyotypes (K) were reviewed by the members of the French-speaking Group for Cytogenetic Hematology (*Groupe Francophone de Cytogénétique Hématologique*) and classified according to the International System for Human Cytogenetic Nomenclature 2016. Complex K (CK) and high-complex K (HCK) were defined as the presence of at least three or at least five numerical or structural chromosomal abnormalities, respectively. FISH was performed according to standard procedures. The specific probes used were as follows: *IGK* (2p11) break-apart (Dako, Santa Clara, CA, and/or Cytocell, Cambridge, UK, or home-grown bacterial artificial chromosome (BAC) clones RP11-316G9, RP11-526L16 and RP11-1021F11 (Martin-Subero*, et al* 2002)), *IGH* (14q32) break-apart (Abbott Molecular, Des Plaines, IL), *TRA/TRD* (14q11) break-apart (Cytocell), *ATM* (11q22), *TP53* (17p13), D13S319 (13q14), centromere of chromosome 12 (MetaSystems Probes, Altlussheim, Germany), *BCL2* (18q21) break-apart and *BCL6* (3q27) break-apart (Dako). A break-apart probe for *CDK6* rearrangements was built using home-grown BAC clones RP11-246N14 and RP11-90H9. Extraction, labeling, and hybridization were performed as described previously (Cosson*, et al* 2014).

Diagnoses and cytological assessments

Of the 57 patients included in the study, 10 were initially diagnosed as having CLL (n=6), high-count CLL-like MBL (n=2) and SLL (n=2), according to the World Health Organization criteria (Swerdlow*, et al* 2017). A cytology review was performed in six of the 10 cases. Among the 47 other patients, 3 were classified as B-cell prolymphocytic leukemia (B-PLL) and 44 were considered as low grade lymphomas, excluding mantle cell lymphoma and follicular lymphoma. Most of these 44 cases were classified as MZL, including 4 with a histological confirmation on a splenectomy sample (n=3) or BM biopsy (n=1). A cytology review of blood smears and/or BM aspirate smears was performed in 28 of the 47 cases. The cytological criteria described by Matutes et al. were used to identify a typical MZL aspect (Matutes*, et al* 2008). For comparative purposes, blood smears of 10 SMZL cases (with a confirmed diagnosis after the histological assessment of a splenectomy sample) known to lack t(2;7)(p11;q21) or a variant were reviewed at the same time. The cytology review was performed by three expert cytologists (KM, MP, and CLL).

IGHV and mutational analyses

Molecular analyses were performed on DNA extracted from whole blood (n=28) or from cytogenetic pellets (n=9). *IGH* sequences were analyzed as described previously (Ghia*, et al* 2007). For cases of CLL/MBL/SLL, the *IGHV* genes were considered to be unmutated if the percentage identity with the germline counterparts was ≥98% (Ghia*, et al* 2007). For the other cases, sequences were considered to be unmutated (100% identity), minimally or borderline mutated (99.9%–97%) or significantly mutated (<97% identity), according to the criteria published by Bikos et al. (Bikos*, et al* 2012) for MZL.

Mutation hotspots in the *TP53* gene (exons 4-9) and *NOTCH2* gene (exon 34) were analyzed by Sanger sequencing, as described previously (Kiel*, et al* 2012, Pospisilova*, et al* 2012). The *MYD88 L625P* mutation was detected using real-time allele-specific PCR, as described previously (Xu*, et al* 2013).

Statistical analysis

Overall survival (OS) was defined as the time interval between diagnosis and death or last follow-up. Categorical variables were compared using a Fisher's exact test, whereas continuous variables were compared using the Mann-Whitney test. OS was estimated according to the Kaplan-Meier (KM) method. A log-rank test was used to compare the survival curves. A Cox proportional hazards model was used to determine hazard ratios and 95% confidence intervals. Median follow-up was calculated with the reverse KM method. All tests were two-sided, and the threshold for statistical significance was set to $p<0.05$. All statistical analyses were performed using IBM SPSS Statistics software for Windows (version 25.0, IBM, Armonk, NY).

RESULTS

Characteristics of the study population, and cytology reviews

Among 17752 patients with mature B-cell neoplasms, 57 (0.32%) carried a t(*CDK6*). The characteristics of the study population are summarized in Table I. There was a predominance of males (n=36, 63%), and the median (range) age at diagnosis was 69 (35-90). Thirteen of the 48 (27%) patients with data presented with splenomegaly, and 9 out of 49 (18%) had lymphadenopathy. The median (range) lymphocyte count was 7.2 G/L (1-112.4). All 10 cases (100%) of CLL/MBL/SLL and 26 of the 47 (55%) cases of MZL/unclassified small B-cell lymphoma (SmBL) patients expressed CD5 (Supplemental Table S1).

The initial diagnoses were CLL (n=6), high-count CLL-like MBL (n=2), small lymphocytic lymphoma (SLL, n=2), B-PLL (n=3), SMZL (n=25), nodular MZL (NMZL) (n=1), bronchus-associated lymphoid tissue (BALT) lymphoma (n=1), and unclassified SmBL (n=17). Six out of 10 CLL/MBL/SLL cases were reviewed, and were found to have a high proportion of mature lymphocytes and no prolymphocytic cells. The three cases diagnosed as B-PLL were reclassified as SMZL, since the proportion of prolymphocytic cells was in fact below 55% (ranging from 12% to 48% of the lymphoid blood cells), and the prolymphocytic cells were mixed with MZL cells (Supplementary Fig S1). Fifteen of the 25 SMZL were reviewed: they were found to have an SMZL aspect: six displayed a typical morphological aspect (according to Matutes' criteria) and 9 exhibited some atypical features consisting in the presence of prolymphocytic cells (median (range) proportion of lymphoid cells: 7% [3-22]). Among the 17 unclassified cases of SmBL, 6 had a BM biopsy whose aspect did not enable us to classify the type of lymphoma more precisely. Six of the 17 cases of SmBL were cytologically reviewed: one had a typical SMZL aspect, and 5 had an aspect compatible with SMZL but with some atypia: prolymphocytic cells (median (range) proportion of lymphoid cells: 8% [6-30]) were associated with classical marginal zone cells. In all, 47 cases were classified as SmBL, including 36 cases of MZL (34 SMZL, 1 NMZL and 1 BALT) and 10 cases of CLL/MBL/SLL (Table I and Supplementary Table S1).

In the 10 reviewed cases of SMZL lacking t(2;7) or a variant, the blood smears showed a typical SMZL aspect according to Matutes' criteria, with very few or no prolymphocytic cells (median (range): 1% [0-5]) (Supplemental Table S2).

Cytogenetic analysis

The karyotyping and FISH results are summarized in Supplemental Table S3. The FISH analysis confirmed the involvement of *CDK6* in all 57 cases. A t(2;7)(p11;q21) was found in 51 of the 57 (89%) patients, with an additional derivative chromosome 7 from t(2;7) in 4 of the 51. Forty-seven cases were analyzed with an IGK break-apart FISH probe. In the majority of the patients (n=33 out of 47, 70%), the breakpoint in *IGK* was located in the proximal region. In 14 patients (30%), the breakpoint was more centromeric (in the *IGKV* distal region)(Supplemental Fig S2).

Two cases (one CLL and one SLL) had a t(7;14)(q21;q32) *IGH/CDK6* and four (two SMZL and two CLL) had a t(7;14)(q21;q11) – a previously undescribed translocation involving the *CDK6* and *TRA/TRD* loci (*TRAD*) (as proven by a FISH analysis; Fig S3).

The t(*CDK6*) was the sole abnormality detected by karyotyping in 19 of the 57 (33%) patients, including 3 of the 4 t(7;14) *TRAD/CDK6* cases and 1 of the 2 t(7;14) *IGH/CDK6* cases. When the

translocation was not isolated, it was never a secondary event: it was present in the primary clone in 22 cases, found concomitantly with other chromosomal abnormalities in 12 cases, and found in an independent clone in 4 cases.

Twenty-eight (49%) patients exhibited a CK, and 18 of these (32%) exhibited a HCK. The median (range) number of chromosomal abnormalities was 2 (1-14). The most frequent chromosomal abnormality associated with t(*CDK6*) was a 17p deletion involving the *TP53* gene [del(17p)], which was detected in 23 of the 57 (40%) patients. The other frequent aberrations were as follows: 13q14 deletion [del(13q)] in 22/54 (39%), 8p deletion [del(8p)] in 14/57 (25%), trisomy 3 in 10/57 (18%), 8q gain in 7/57 (12%), trisomy 12 in 6/57 (11%), and trisomy 18 in 4/57 (7%) (Table I and Fig 1). Deletion of 7q [del(7q)] was observed in only one patient (patient #17 in Supplemental Table S3): a case of SMZL in which t(2;7) and del(7q) were found in independent clones.

The 17p deletion was significantly associated with the presence of prolymphocytic cells (93% in the del(17p) subgroup vs. 24% in the subgroup lacking del(17p), p<0.001), *TP53* mutation (60% vs. 10.5%; p=0.002), CK (78% vs. 29%; p<0.001), HCK (61% vs. 12%; p<0.001), a higher number of chromosomal abnormalities (median(range) 5 (2-13) vs. 1 (1-6); p<0.001), del(13q) (59% vs. 28%; p=0.028), del(8p) (48% vs. 9%; p=0.001), and 8q gain (26% vs. 3%; p=0.014). The correlations between del(17p) and these parameters were also statistically significant when considering only the 47 cases of MZL/SmBL, except for del(13q) (Supplemental Table S4).

Molecular analysis

TP53 gene sequencing revealed the presence of a mutation in 10 of the 36 (27%) tested patients. All the mutations affected the DNA-binding domain (Supplemental Fig S4). The *TP53* mutation was associated with a del(17) in 9 of the 10 patients. Overall, the *TP53* gene was disrupted (i.e. mutated and/or deleted) in 24 of the 44 (55%) tested patients.

A *MYD88* L265P mutation was found in 5 of the 37 patients, all in the MZL/SmBL subgroup. Sequencing of exon 34 of the *NOTCH2* gene did not reveal any mutations in the 27 tested patients.

IGHV genes were sequenced in 37 patients. The *IGHV* genes were mutated in 5 of 8 cases of CLL/MBL/SLL. The great majority of MZL/SmBL cases (25/29, 86%) carried mutations and 20 of these were significantly mutated (<97% homology). Eighteen different *IGHV* genes were identified among the 29 cases; the most frequent were *V3-23* (4/29, 14%) and *V4-59* (3/29, 10%) (Fig 1).

Survival analysis

The median follow-up time after diagnosis was 54 months. Twenty-eight of the 57 patients (49%) had been treated. The characteristics of these treatments are summarized in Supplemental Table S5. We assessed survival in the cohort as a whole (n=57) and in the MZL/SmBL subgroup (n=47) but not (given the small sample size) in the CLL/SLL/MBL subgroup. The median OS time was not attained in the cohort as a whole or in the MZL/SmBL subgroup. The calculated OS was 81% at 5 years and 73% at 10 years for the entire cohort, and 82% at both 5 and 10 years for the 47 patients with MZL/SmBL (Table I). The presence of del(17p) or a *TP53* mutation did not appear to influence survival in the cohort as a whole (p=0.197 and p=0.627, respectively) (Table II and Fig 2) or in the MZL/SmBL subgroup (p=0.426 and p=0.333, respectively) (Supplemental Table S6 and Fig S5).

DISCUSSION

Here, we report a large cohort of patients with mature B-cell neoplasm and a t(2;7)(p11;q21) translocation or variants involving *CDK6*.

The t(*CDK6*) was the sole anomaly in 33% of the patients. When accompanied by other abnormalities, t(*CDK6*) was never found in a subclone – suggesting that t(*CDK6*) is possibly a primary pathogenic event. The majority of the patients (89%) displayed a t(2;7) involving the *IGK* locus. Only two cases harbored a variant translocation with rearrangement of the *IGH* locus. In four cases, we identified the *TRAD* locus as a t(*CDK6*) partner for the first time.

We found this translocation in two cases of CLL and two cases of SMZL. The fact that the translocation was isolated in three of the four cases suggests that it is a “driver” genetic event in pathogenesis. Moreover, given the involvement of the *TRAD* locus, the translocation might have occurred at an early step in lymphoid differentiation. In mature B-cell neoplasms, 14q11 translocations are very rare. Eighteen cases are listed in the Mitelman database (https://mitelmandatabase.isb-cgc.org), 13 of which are CLL. The involvement of the *TRAD* locus has been proven by FISH analysis in only a few cases with *MYC* or *NKX2-5* locus rearrangement (Lau*, et al* 2008, Put*, et al* 2012, Su*, et al* 2008). Since the TCR enhancers are known to be lymphoid specific but not T-cell restricted, the functional consequence of these rearrangements is transcriptional activation of the partner genes. Hence, as seen for *IGK/CDK6* (Brito-Babapulle*, et al* 2002, Corcoran*, et al* 1999), t(7;14) *TRAD/CDK6* results in overexpression of *CDK6*. However, a lack of material prevented us from evaluating *CDK6* expression. Dual CDK4/6 inhibitors are currently under

development, with promising preclinical and clinical results in mature B-cell malignancies (Martin*, et al* 2019, Tanaka*, et al* 2020). They may represent a therapeutic option in patients with t(*CDK6*).

CLL/MBL/SLL cases. Of the 57 cases of t(*CDK6*), 10 had a diagnosis of CLL, CLL-like MBL or SLL (confirmed by clinical findings, cytology, and immunophenotyping, with a Matutes score of 4 or 5), including two cases with t(7;14)(q21;q32) *CDK6/IGH* and two cases with t(7;14)(q21;q11) *CDK6/TRAD*. Our results confirmed that (i) t(*CDK6*) is very rare in CLL and related diseases, (ii) variant translocations are frequent in this subgroup (4/10 cases), and (iii) *CDK6/IGH* is restricted to this subgroup (Hayette*, et al* 2003).

MZL/SmBL. In the present study, t(*CDK6*) was mostly associated with MZL (36/47, 76%). SMZL is difficult to diagnose. According to Matutes et al., the minimum criteria are either spleen histology results and an immunophenotype or (if a histological assessment of the spleen is unavailable) typical blood and BM morphologies, an immunophenotype, and intrasinusoidal infiltration by CD20-positive cells (Matutes*, et al* 2008). About 20-25% of cases of SMZL are CD5+ (Matutes*, et al* 2008, Salido*, et al* 2010). However, a histological analysis is not always performed when there is no indication for treatment, and SMZL is often only diagnosed after other types of low-grade B-cell lymphoma have been ruled out. In the present study, a histological analysis was not available for the majority of patients. The strong suspicion of SMZL was based on cytological and immunophenotyping data. In our series, the Matutes score was 3 or less for all 27 patients with available data, and more than a half of these were CD5+. In the absence of lymphoid cells with cleaved nuclei and a translocation involving BCL2, the diagnosis of follicular lymphoma was very unlikely. All CD5+ MZL/SmBL cases in our cohort (n=26/47, 55%) lacked t(11;14)(q13;q32), which rules out a diagnosis of leukemic non-nodal mantle cell lymphoma (MCL, in which this aberration is always present). We cannot formally rule out the possibility that CD5+ cases were t(11;14)/cyclin-D1-negative MCLs. However, the absence of generalized lymphadenopathy at diagnosis and the indolent clinical course argued against a diagnosis of MCL (Swerdlow*, et al* 2017). A differential diagnosis of lymphoplasmacytic lymphoma could be considered in two patients exhibited a *MYD88* L265P mutation and IgM monoclonal paraprotein (patients #23 and #20). Neither patient had plasma cells, and one presented with villous lymphocytes in the initial cytological assessment. In 11 patients, a cytology review could not be performed and so the initial diagnosis of SmBL could not be refined. However, by a process of elimination, we hypothesized that these 11 cases were also related to MZL, as t(2;7)(p11;q21) translocation is very strongly biased to B-cell lymphoproliferation of marginal-zone origin (Xochelli*, et al* 2014).

In our series, cases of MZL t(*CDK6*)+ frequently featured prolymphocytic cells in a cytological assessment (18/26, 69%). These prolymphocytes were mixed with a lymphoid population having the

features of MZL cells, and so we considered these to be “atypical” cases of MZL. Prolymphocytic cells (defining B-PLL if they account for more than 55% of lymphoid cells in a *de novo* context (Swerdlow*, et al* 2017) can typically be observed as CLL progresses (Kjeldsberg and Marty 1981) and in a leukemic form of MCL (Ruchlemer*, et al* 2004). In contrast, the presence of prolymphocytic cells in MZL is unusual and has been reported in a few cases with transformed splenic B-cell lymphoma only (Algashaamy*, et al* 2018, Hoehn*, et al* 2012)**.** The association between prolymphocytic cells and non-transformed t(*CDK6*)+ MZL is intriguing and seems to be a particular feature of these cases of MZL, as few or no prolymphocytic cells were observed in cases of SMZL without t(*CDK6*). The presence of prolymphocytes was correlated with the presence of del(17p) but not with OS.

We found that del(17p) and the *TP53* mutation were respectively present in 47% and 33% of cases of MZL/SmBL t(*CDK6*)+. These frequencies are higher than those observed in classical SMZL, in which del(17p) is detected in 18-33% of patients and a *TP53* mutation is detected in 15-16% (Jaramillo Oquendo*, et al* 2019, Parry*, et al* 2015, Salido*, et al* 2010) (Supplemental Table S7). Interestingly, del(7q) is the most frequent abnormality in SMZL (19-39% (Parry*, et al* 2015, Salido*, et al* 2010)) but we detected it only in one case. The deletion was present in a distinct clone that lacked t(2;7) –, suggesting that these two aberrations are mutually exclusive.

We found *IGHV* gene mutations in 86% of the patients with MZL/SmBL t(*CDK6*)+; this frequency is similar to that found in cases of SMZL (Bikos*, et al* 2012, Salido*, et al* 2010). However, the *IGHV* usage profile observed in our t(*CDK6*)+ patients differed: we did not find any cases harboring an *IGHV1-02*04* gene, while the latter is the most frequent allele (25%) in classical SMZL (Bikos*, et al* 2012).. Lastly, we did not find any patients with a mutation in the *NOTCH2* gene – the most frequently mutated gene in SMZL, and which accounts for about 20% of cases (Jaramillo Oquendo*, et al* 2019, Rossi*, et al* 2012).

Overall survival was good in our series of MZL/SmBL t(*CDK6*)+ cases: the 5-year OS rate was above 80%. This was in line with the favorable prognosis usually observed in SMZL (Arcaini*, et al* 2016). In SMZL, the presence of a *TP53* deletion or *TP53* mutations is reportedly an independent marker of poor OS (Parry*, et al* 2015, Salido*, et al* 2010). In our series of t(*CDK6*)+ lymphomas, *TP53* abnormalities were not significantly associated with poor OS. However, the median follow-up time was only 45 months, which prevents us from drawing definitive conclusions.

In summary, our present results demonstrated that *CDK6* translocations mainly involved the *IGK* locus and very rarely the *IGH* or *TRAD* loci. The latter partner was reported for the first time in the present study. We confirmed that t(*CDK6*) are rare events in CLL/MBL and SLL, and that they frequently involve alternative partners (*IGH* or *TRAD*). This large series extended our biological and molecular knowledge of a rare aberration in non-CLL B-cell lymphoproliferative disease. Our results

suggest that most cases of t(*CDK6*+) constitute a particular subgroup of B-cell marginal zone lymphomas with distinctive features: prolymphocytic cells, frequent CD5 expression, a high frequency of *TP53* deletion and mutation, a low frequency of 7q deletion, and the absence of *IGHV1-2*04* and *NOTCH2* mutation. The t(*CDK6*)+ lymphomas are indolent. Patients have a good OS, which is apparently not influenced by the presence of a *TP53* aberration. In case of disease progression, CDK6 may represent a therapeutic target with new CDK4/6 inhibitors.

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Table 1. Characteristics of the study population.

Parameter		Whole cohort (n=57)		MZL/SmBL cases (n=47)		CLL/MBL/SLL cases (n=10)	
Sex (n, %)	M	36	63	28	60	8	80
	F	21	47	19	40	2	20
Age at diagnosis	median (range)	69 (35-90)		69 (35-90)		68 (51-87)	
Lymphadenopathy (n, %)	no	40	82	37	88	3	43
	yes	9	18	5	12	4	57
Splenomegaly (n, %)	no	35	73	32	78	3	43
	yes	13	27	9	22	4	57
Treated (n, %)	no	29	51	27	57	2	20
	yes	28	49	20	43	8	80
Time to first treatment (months)	median (range)	27 (0-114)		27 (0-73)		7 (1-114)	
Number of lines of treatment (n, %)	0	29	51	27	55	2	20
	1	17	30	14	36	3	30
	2	4	7	3	3	1	10
	3	3	5	1	3	2	20
	4	3	5	1	0	2	20
	5	1	2	1	3	0	0
Follow-up (months)	median (95% CI)	54 (33.5-74.4)		45 (23.2-66.8)		75 (0-169.1)	
Overall survival (% [95%CI])	at 1 year	90.1 [82.1-98.8]		90.3 [81.6-99.9]		Not calculated*	
	at 5 years	81.2 [69.9-94.4]		82.3 [69.9-97.7]			
	at 10 years	73.1 [56.6-94.4]		82.3 [69.9-97.7]			
Paraprotein (n, %)	no	26	63	24	63	2	67
	yes	15	37	14	37	1	33
Lymphocytosis at diagnosis (G/L)	median (range)	7.2 (1-112.4)		7.3 (1-55.5)		5.4 (1.96-112.4)	
Hemoglobin level at diagnosis (g/dL)	median (range)	13.3 (5.6-16.9)		13.6 (5.6-16.9)		13 (8.1-14.3)	
Platelet count at diagnosis (G/L)	median (range)	205 (27-363)		208 (27-363)		183 (86-344)	
Prolymphocytic cells (n, %)	no	14	44	8	31	6	100
	yes	18	56	18	69	0	0
CD5 expression (n, %)	no	21	37	21	45	0	0
	yes	36	63	26	55	10	100
Light chain expression (n, %)	Kappa	33	59	29	63	4	40
	Lambda	23	41	17	37	6	60
Matutes score (n, %)	0	8	16	8	20	0	0
	1	19	38	19	47.5	0	0

	2	10	25	10	25	0	0
	3	3	6	3	7.5	0	0
	4 or 5	10	25	0	0	10	100
Number of chromosomal abnormalities (karyotype)	median (range)	2 (1-14)		2 (1-14)		2.5 (1-5)	
Isolated t(*CDK6*) (by karyotype) (n, %)	no	38	67	31	66	7	70
	yes	19	33	16	33	3	30
Complex karyotype (n, %)	no	29	51	24	51	5	50
	yes	28	49	23	49	5	50
Highly complex karyotype (n, %)	no	39	68	32	68	7	70
	yes	18	32	15	32	3	30
Partners of *CDK6* (n, %)	t(2;7) *IGK*	51	89	45	96	6	60
	t(7;14) *IGH*	2	4	0	0	2	20
	t(7;14) *TRAD*	4	7	2	4	2	20
Trisomy 12 (K + FISH) (n, %)	no	51	89	42	89	9	90
	yes	6	11	5	11	1	10
13q14 deletion (FISH) (n, %)	no	32	61	26	59	6	60
	yes	22	39	18	41	4	40
11q/ATM deletion (K + FISH) (n, %)	no	55	96	46	98	9	90
	yes	2	3,5	1	2	1	10
17p/TP53 deletion (K + FISH) (n, %)	no	34	60	25	53	9	90
	yes	23	40	22	47	1	10
Trisomy 18/18q (K + FISH) (n, %)	no	53	93	44	94	9	90
	yes	4	7	3	6	1	10
Trisomy 3/3q (K + FISH) (n, %)	no	47	82	38	81	9	90
	yes	10	18	9	19	1	10
8q gain (K + FISH) (n, %)	no	50	88	41	87	9	90
	yes	7	12	6	13	1	10
8p deletion (K) (n, %)	no	43	75	34	72	9	90
	yes	14	25	13	28	1	10
***IGHV* genes (n, %)**	unmutated	7	19	4	14	3	38
	mutated	30	81	25	86	5	62
***MYD88* L265P (n, %)**	absence	32	86	26	84	6	100
	presence	5	14	5	16	0	0
***TP53* mutation (n, %)**	absence	26	72	20	67	6	100
	presence	10	28	10	33	0	0
***TP53* disruption (mutated and/or deleted) (n, %)**	no	20	45	14	38	6	86
	yes	24	55	23	62	1	14
***NOTCH2* mutation (n, %)**	absence	27	100	23	100	4	100

MZL: marginal zone lymphoma; SmBL: small B-cell lymphoma; SLL: small lymphocytic lymphoma; CLL: chronic lymphocytic leukemia; MBL: monoclonal B-cell lymphocytosis; K: karyotyping; CI: confidence interval; *: the sample was too small.

Table II. Univariate analysis of overall survival, using the Kaplan Meier method.

		Whole cohort (n=57)				MZL/SmBL (n=47)			
Parameter		**OS at 5 years (%)**	**HR**	**95% CI**	***P*-value**	**OS at 5 years (%)**	**HR**	**95% CI**	***P*-value**
CD5 expression	negative	90.5	1.51	0.37-6.13	0.566	90.5	1.51	0.28-8.25	0.633
	positive	76.9				78.4			
Prolymphocytic cells	no	81.3	1.33	0.24-7.37	0.743	88.9	1.08	0.11-10.52	0.829
	yes	78.0				75.7			
Del(17p)	no	86.6	2.26	0.63-8.05	0.197	87.7	2.59	0.50-13.51	0.243
	yes	75.2				79.3			
***TP53* mutated**	no	86.7	1.97	0.24-16.01	0.627	92.3	3.33	0.26-41.97	0.333
	yes	90.9				90.0			
***TP53* disrupted**	no	83.3	2.70	0.54-13.53	0.465	90.0	3.43	0.40-29.70	0.426
(mut and/or del)	yes	78.2				80.9			

HR: Hazard ratio; CI: confidence interval

Figure 1. Distribution of recurrent cytogenetic and molecular abnormalities among the 57 patients. In the heat map, rows represent individual patients and columns correspond to aberrations. SLL: small lymphocytic lymphoma; CLL: chronic lymphocytic leukemia; MBL: monoclonal B-cell lymphocytosis; SMZL: splenic marginal zone lymphoma; USmBL: unclassified small B-cell lymphoma; NMZL: nodular marginal zone lymphoma; BALT: bronchus-associated lymphoid tissue lymphoma; PL cells: prolymphocytic cells; CK: complex karyotype; HCK: highly complex karyotype.

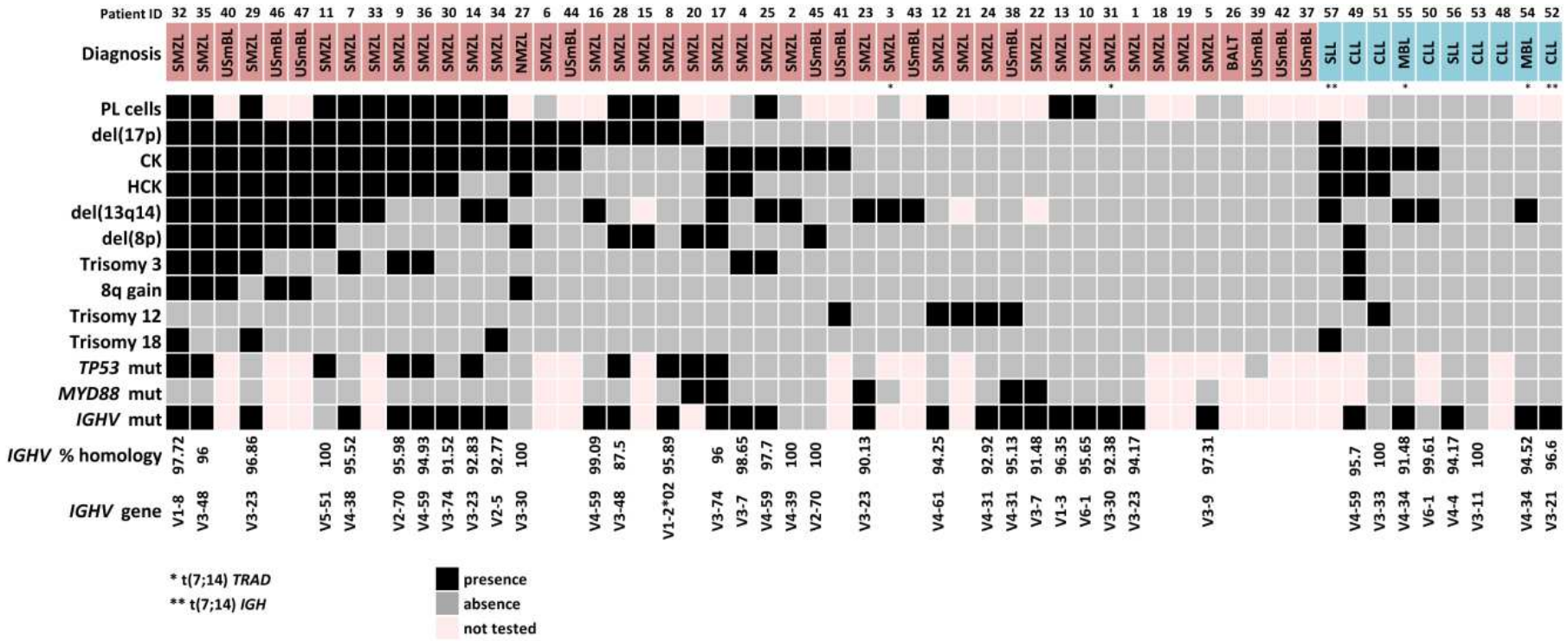
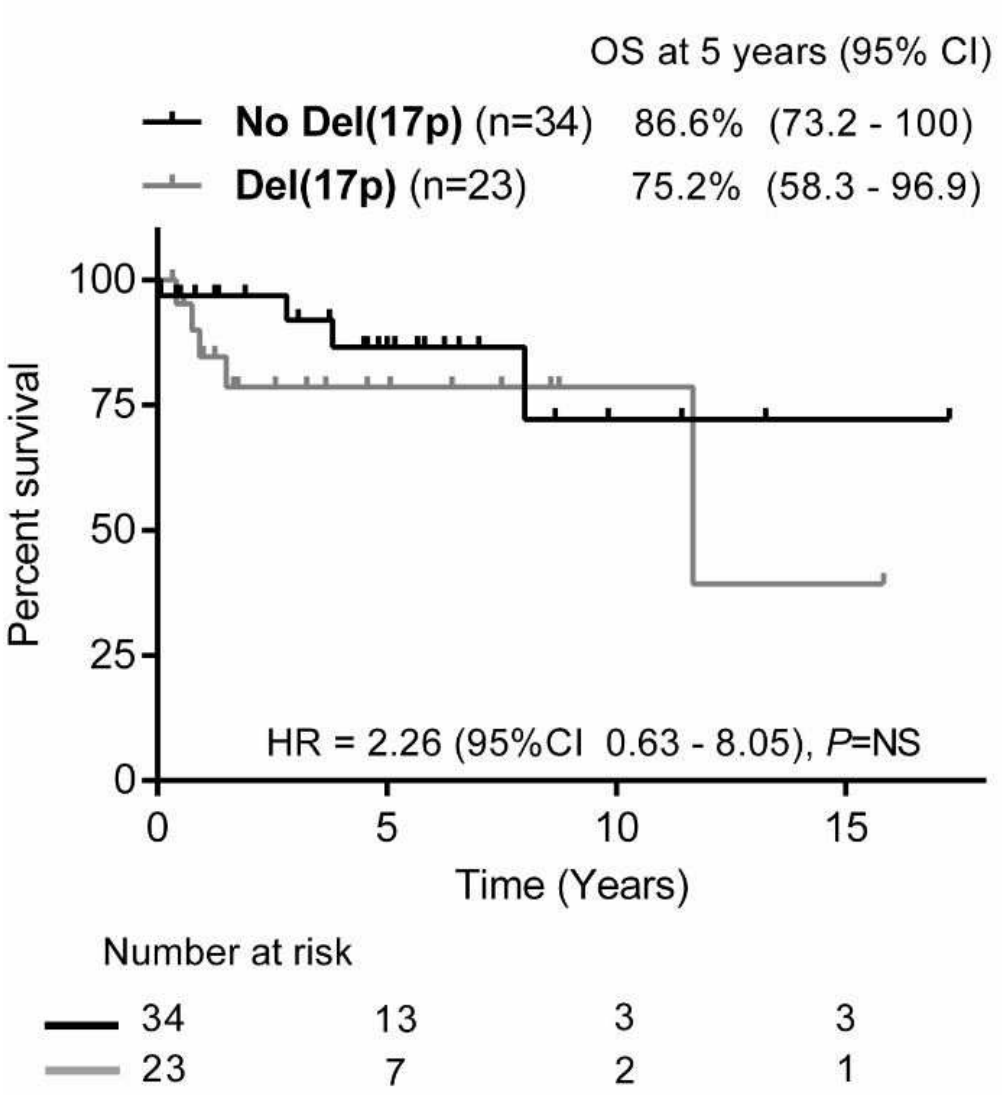
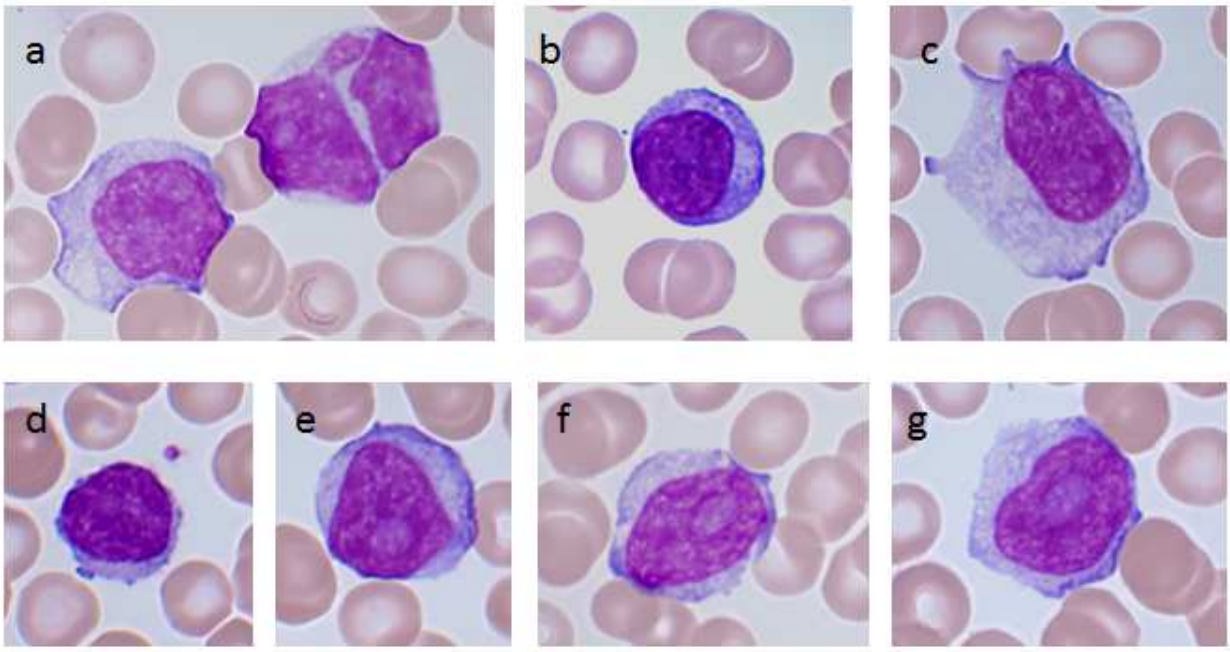
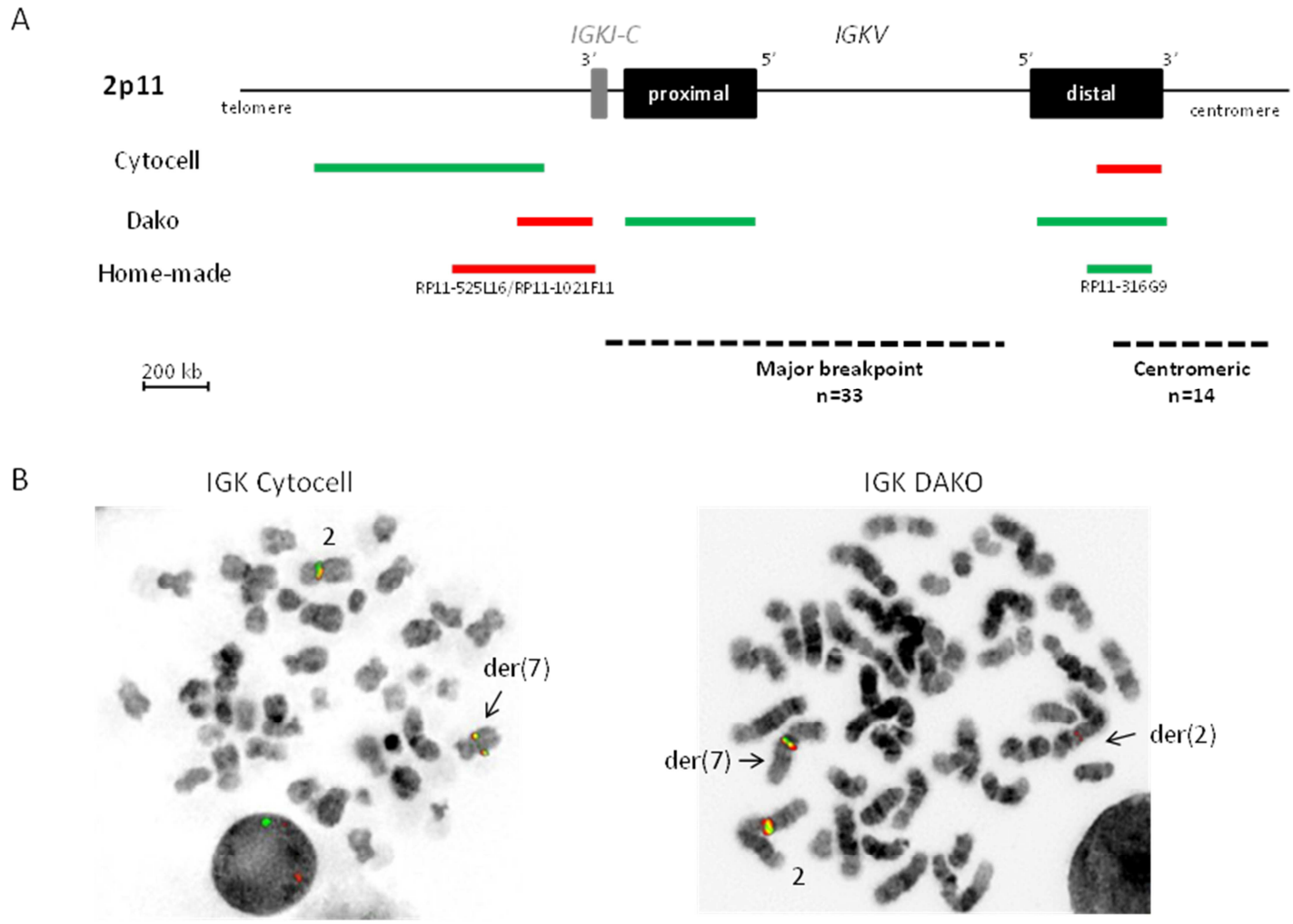


Figure 2. Kaplan-Meier estimates of OS, as a function of the 17p deletion. A. For the cohort as a whole (n=57). B. For cases of SMZL/SmBL (n=47).

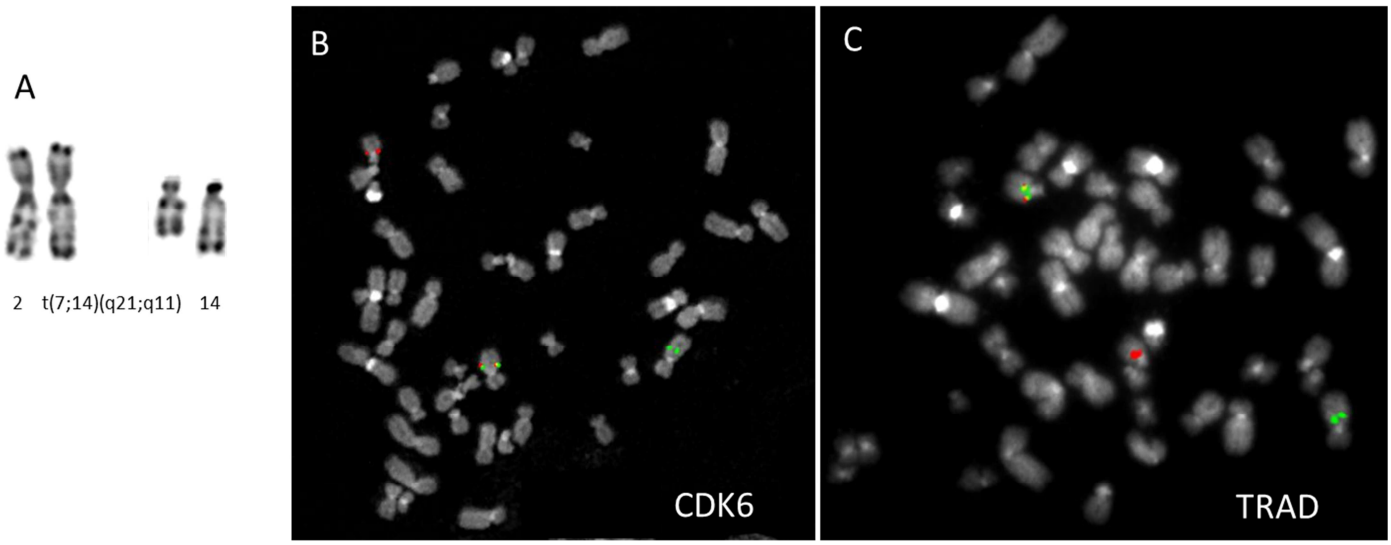




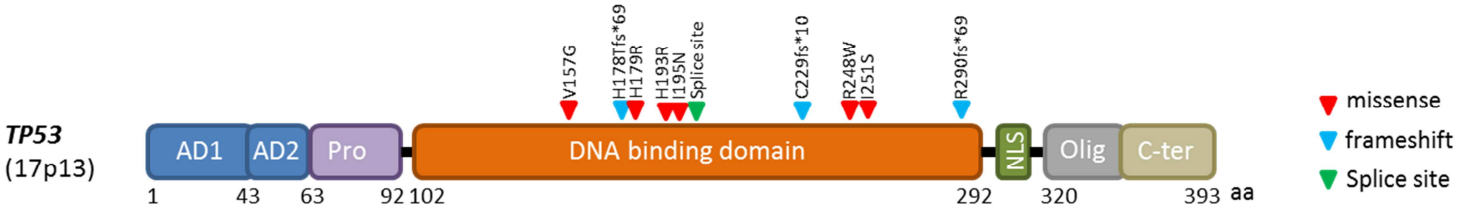
Supplemental Figure S1. Cytological aspect of an SMZL sample with prolymphocytic cells (case #14). The sample features a pleomorphic lymphocytic population with MZL cells (a, b and c), small lymphoid cells without specific features (d), and prolymphocytic cells (e, f and g).



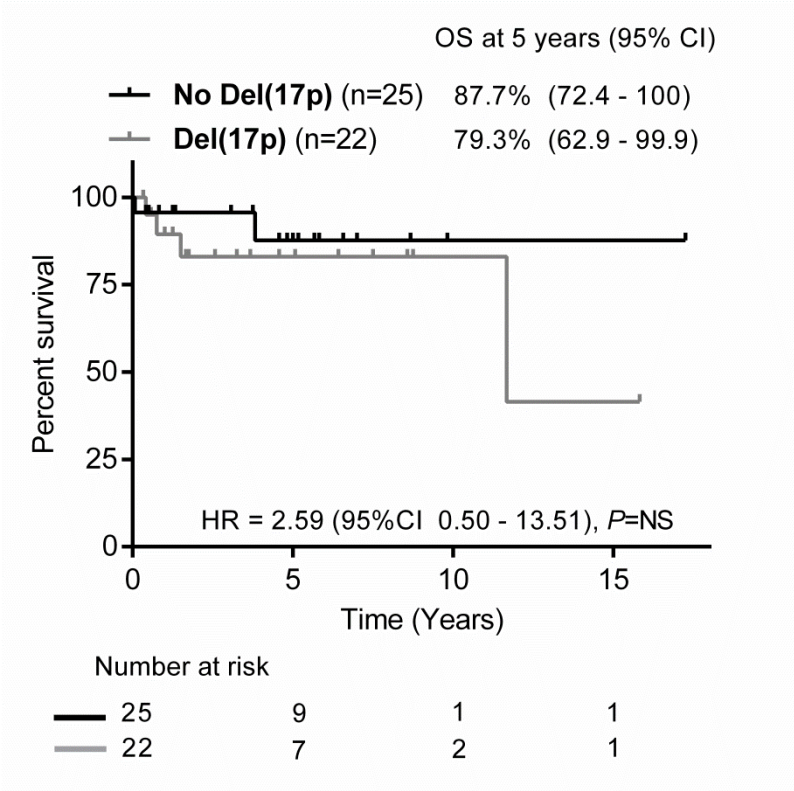
Supplemental Figure S2. **Translocation t(2;7) breakpoints in the *IGK* gene locus**. A. Schematic representation of the region covered by the different IGK probes used in the study (approximate probe sizes and gaps between red and green probes are provided), and the distribution of 2p11 breakpoints. B. FISH images for patient #8: no splitting signal was seen with the Cytocell IGK probe, which was entirely translocated to the derivative chromosome 7 (left panel), indicating a centromeric 2p11 breakpoint. With the Dako probe, a fusion signal was seen on the derivative chromosome 7, and a telomeric IGK (red) signal was still seen on the derivative chromosome 2; this suggest that the rearrangement mechanism was complex, with a probable inversion of the *IGK* locus prior to t(2;7) (right panel). A similar profile was observed in six other patients.



Supplemental Figure S3. Translocation t(7;14)(q21;q11). A. partial R-banded karyotype. B and C. FISH images. B. A representative metaphase from patient #3, hybridized with the home-made CDK6 break-apart probe (RP11-246N14 labeled with FITC + RP11-90H9 labeled with rhodamine). A red-green signal is seen on the normal chromosome 7, a green signal is present on the derivative chromosome 7, and a red signal is observed on the derivative chromosome 14. B. A representative metaphase from patient #3, hybridized with the *TRA/TRD* (14q11) break-apart probe (Cytocell). A red-green signal is seen on the normal chromosome 14, a red signal is present on the derivative chromosome 14, and a green signal is observed on the derivative chromosome 7.



Supplemental Figure S4. ***TP53*** **mutations identified in patients with t(*CDK6*).** AD1: transactivation domain 1; AD2: transactivation domain 2; Pro: proline-rich domain; NLS: nuclear localization signal; Olig: oligomerization domain; C-ter: carboxy terminus domain.



Supplemental Figure S5. Kaplan-Meier estimates of OS among patients with MZL/SmBL (n=47), as a function of the presence or absence of the 17p deletion.

Supplemental Table S1. Clinical, laboratory, cytological and immunophenotyping data for the 57 patients with a t(*CDK6*) translocation

Patient ID	Sex	Splenomegaly (0: no, 1: yes)	Lymphadenopathy (0: no, 1: yes)	Paraprotein	Initial diagnosis	Histology	Tissue examined for morphological review	Morphological review conclusion	Detailed morphology if atypical (*or not reviewed)	% prolymphocytic cells	Lymphocytosis at diagnosis (G/L)	CD5 expression	CD23 expression	Matutes' score
1	M	1	0	-	SMZL	BM : SMZL Spleen : SMZL	PB	SMZL		0	3.7	+	+	2
2	M	0	0	-	SMZL	BM : low-grade B-cell lymphoma	PB	SMZL		0	15.9	-	-	0
3	M	0	0	-	SMZL	-	PB	SMZL		0	5.28	-	-	2
4	M	1	0	IgM K	SMZL	-	PB	SMZL		0	11.3	-	+	1
5	M	0	0	-	SMZL	-	BM	SMZL		0	11	-	NA	NA
6	F	0	0	-	SMZL	-	BM	atypical SMZL	numerous lymphoplasmocytes	0	6.3	+	+	3
7	M	0	0	-	SMZL	-	PB	atypical SMZL	presence of prolymphocytes	3	7.2	+	+	2
8	M	0	0	IgM K	SMZL	-	PB	atypical SMZL	presence of prolymphocytes	3	2.15	-	-	1
9	F	0	1	IgM L	SMZL	-	PB	atypical SMZL	presence of prolymphocytes	3	43.09	+	-	1
10	M	0	0	-	SMZL	-	PB	atypical SMZL	presence of prolymphocytes	5	10.27	-	-	0
11	F	1	0	-	SMZL	-	BM	atypical SMZL	presence of prolymphocytes	7	29.37	+	-	1
12	F	1	0	-	SMZL	-	PB	atypical SMZL	nuclear irregularity, numerous naked nuclei, presence of prolymphocytes	9	5.8	+	+	2
13	M	0	0	IgM L	SMZL	-	PB	atypical SMZL	presence of prolymphocytes	11	3	-	-	0
14	F	0	0	IgM L	SMZL	-	PB	atypical SMZL	presence of prolymphocytes	16	4.25	-	-	0
15	M	1	0	IgM L	SMZL	-	PB	atypical SMZL	presence of prolymphocytes	22	7.25	+	-	1
16	F	NA	NA	NA	SZML	-	-	-			2.32	-	-	1
17	F	1	0	-	SMZL	Spleen : SMZL	-	-			4.4	-	+	1
18	M	0	0	-	SMZL	-	-	-			6.35	-	-	0
19	M	1	0	-	SMZL	-	-	-			8.08	-	NA	NA
20	M	0	0	IgM K and IgM L	SMZL	BM : low-grade B-cell lymphoma	-	-	* some villous lymphocytes		9	-	-	1
21	F	NA	NA	NA	SMZL	-	-	-			8.37	-	-	1

Patient ID	Sex	Splenomegaly (0: no, 1: yes)	Lymphadenopathy (0: no, 1: yes)	Paraprotein	Initial diagnosis	Histology	Tissue examined for morphological review	Morphological review conclusion	Detailed morphology if atypical (*or not reviewed)	% prolymphocytic cells	Lymphocytosis at diagnosis (G/L)	CD5 expression	CD23 expression	Matutes' score
22	M	1	1	NA	SMZL	Spleen : SMZL	-	-			4.488	+	-	1
23	F	0	0	IgM K and IgM L	SMZL	-	-	-			2.283	+	-	1
24	F	NA	NA	NA	SMZL	BM : SMZL	-	-			8.15	+	+	3
25	F	0	0	IgM K	SMZL	-	-	-	* presence of prolymphocytes	not evaluated	7.26	+	-	1
26	M	0	0	-	BALT	Lung: BALT	PB	typical MZL cells		0	3.9	-	+	1
27	M	NA	1	NA	NMZL	Lymph node: NMZL	-	-			1	+	-	2
28	F	0	0	NA	B-PLL	-	PB	atypical SMZL	presence of prolymphocytes	48	19.75	-	-	0
29	F	NA	NA	NA	B-PLL	-	PB	atypical SMZL	presence of prolymphocytes	18	NA	+	-	NA
30	M	0	0	-	B-PLL	-	PB	atypical SMZL	numerous lymphoplasmocytes and presence of prolymphocytes	12	2.26	-	-	0
31	M	0	0	-	USBCL	-	PB	SMZL		0	20.8	+	-	1
32	M	0	0	IgG K	USBCL	-	PB	atypical SMZL	a majority of lymphocytes with a nucleolus, some of them being "true" prolymphocytes	11	6.55	-	-	0
33	M	0	0	-	USBCL	-	PB	atypical SMZL	presence of prolymphocytes	6	8.48	+	+	2
34	M	0	0	-	USBCL	BM : low-grade B-cell lymphoma	PB	atypical SMZL	numerous lymphoplasmocytes and presence of prolymphocytes	8	5.69	+	+	2
35	M	0	1	-	USBCL	-	PB	atypical SMZL	presence of prolymphocytes	30	55.5	-	-	1
36	F	NA	NA	NA	USBCL	-	PB	atypical SMZL	polymorphic population with cells of unequal size, heterogeneous chromatin, often with a nucleolus, presence of prolymphocytes	8	46.4	-	-	1
37	M	0	0	-	USBCL	-	-	-			2.41	+	-	NA

Patient ID	Sex	Splenomegaly (0: no, 1: yes)	Lymphadenopathy (0: no, 1: yes)	Paraprotein	Initial diagnosis	Histology	Tissue examined for morphological review	Morphological review conclusion	Detailed morphology if atypical (*or not reviewed)	% prolymphocytic cells	Lymphocytosis at diagnosis (G/L)	CD5 expression	CD23 expression	Matutes' score
38	M	0	0	IgM K	USBCL	BM : not informative	-	-			2.2	+	NA	NA
39	M	0	0	-	USBCL	-	-	-			18.96	+	-	1
40	M	1	0	-	USBCL	-	-	-			5.5	+	+	2
41	F	0	0	IgG K	USBCL	BM : nodular lymphocytic infiltration	-	-			4.83	-	-	NA
42	M	0	0	IgM K	USBCL	BM : low-grade B-cell lymphoma	-	-			7.8	+	-	NA
43	F	0	1	yes (not typed)	USBCL	BM : low-grade B-cell lymphoma	-	-			14.39	+	-	NA
44	F	0	0	-	USBCL	BM : low-grade B-cell lymphoma	-	-			7.7	+	-	2
45	F	0	0	-	USBCL	-	-	-			35.85	+	-	1
46	M	0	0	NA	USBCL	-	-	-			12.99	+	-	1
47	F	0	0	-	USBCL	-	-	-			19.9	+	-	2
48	M	NA	NA	NA	CLL	-	BM	CLL		0	5.74	+	+	5
49	M	0	0	NA	CLL	BM: CLL	-	-			11	+	+	4
50	M	1	0	NA	CLL	-	PB	CLL		0	112.4	+	+	5
51	F	NA	NA	NA	CLL	-	BM	CLL		0	5.05	+	+	5
52	M	1	1	-	CLL	-	-	-			33.76	+	+	5
53	M	0	1	-	CLL	-	PB	CLL		0	7.25	+	+	4
54	F	NA	NA	NA	MBL	-	-	-			4.35	+	+	4
55	M	0	0	IgM K	MBL	-	BM	MBL		0	2.93	+	+	5
56	M	1	1	NA	SLL	Lymph node : SLL	BM	SLL		0	2.24	+	+	5
57	M	1	1	NA	SLL	Lymph node: SLL	-	-			1.96	+	+	5

Abbreviations: SMZL: splenic marginal zone lymphoma; NMZL: nodal marginal zone lymphoma; BALT: bronchus-associated lymphoid tissue lymphoma; B-PLL: B-cell prolymphocytic leukemia; USBCL: unclassified small B-cell lymphoma; SLL: small lymphocytic lymphoma; CLL: chronic lymphocytic leukemia; MBL: monoclonal B-cell lymphocytosis; NA: not available

Supplemental Table S2. Cytological findings and karyotypes for 10 cases of SMZL without the t(*CDK6*) translocation. The number of prolymphocytes is quoted as a percentage of total lymphoid cells.

ID	Prolymphocytes (%)	Karyotype
SMZL_1	0	46,XX,del(7)(q31q35)[6]/46,sl,add(3)(p26),del(8)(p12)[12]/93,sdl1x2,+?del(7)(q31q35),del(17)(p11)x2[1]/46,XX[1]
SMZL_2	0	48,XY,+3,+18[13]/46,XY[3]
SMZL_3	1	46,XY[20]
SMZL_4	1	46,XX,del(7)(q2?1q3?1)[2]/46,XX[18]
SMZL_5	1	46,XX[20]
SMZL_6	2	46,XX,del(7)(q21q35)[17]/46,XX,t(1;6)(p3?3;q2?5)[2]/46,XX[1]
SMZL_7	2	46,XX,t(2;8;12;15;17)(?;p1?2;p1?2;q2?5;q1?2)[18]/46,XX[2]
SMZL_8	2	46,XX[15]
SMZL_9	4	46,XY,t(6;19)(q26;p1?2),del(7)(q3?3q3?5)[1]/47,sl,+add(11)(q?13)[13]/48,sdl1,+12[3]/46,XY[3]
SMZL_10	5	46,XX,i(17)(q10)[16]

Supplemental Table S3. Karyotyping and FISH results in the 57 patients with t(*CDK6*).

Patient ID	Diagnosis	Tissue	Karyotype (ISCN 2016)	FISH results										
				translocation t(*CDK6*)	CDK6 rearrangement	IGK rearrangement	IGH rearrangement	TRAD rearrangement	Trisomy 12 (Centromere 12 probe)	13q14 deletion	ATM deletion	TP53 deletion	Trisomy 18q (BCL2 probe)	Trisomy 3q (BCL6 probe)
1	SMZL	Blood	46,XY,t(2;7)(p11;q21)[17]/46,XY[1]	t(2;7)	+	+	ND	ND	-	-	-	-	-	-
2	SMZL	Blood	46,XY,t(2;7)(p11;q21)[9]/46,sl,t(3;11;16)(p21;q13;q22)[7]/46,sl,del(20)(q11.2q13.1)[5]	t(2;7)	+	+	ND	ND	-	+	-	-	-	-
3	SMZL	Blood	46,XY,t(7;14)(q21;q11)[2]/46,XY[18]	**t(7;14) TCRA**	+	ND	ND	+	-	+	-	-	-	-
4	SMZL	Blood	46,XY,t(2;7)(p11;q21),der(3)?(3q29->3q?::?::3p21->3q29),add(16)(p12),add(19)(p11),der(20)t(6;20)(q21;p?11.2)[21]/46,sl,der(12)?t(6;12)(p21;q24)[3]	t(2;7)	+	+	ND	ND	-	-	-	-	-	+
5	SMZL	Bone Marrow	46,XY,t(2;7)(p11;q21)[28]/46,XY[1]	t(2;7)	+	+	ND	ND	-	-	-	-	-	-
6	SMZL	Blood	46,XX,t(2;7)(p11;q21),del(5)(q12q2?2)[10]/46,sl,del(17)(p11)[3]/46,XX[3]	t(2;7)	+	+	ND	ND	-	-	-	+	-	-
7	SMZL	Blood	46,XY,t(2;7)(p11;q21)[5]/46,XY,sl,t(5;13)(q1?4;q?14),del(11)(q23)[4]/46,XY,sl,der(22)t(3;22)(q2?2;q1?3)[3]/46,XY,sl,t(8;13)(q11;q13-14)[3]/46,XY[6]	t(2;7)	+	+	ND	ND	-	+	-	+	-	+
8	SMZL	Blood	46,XY,t(2;7)(p11;q21)[2]/46,sl,add(17)(p12)[3]/46,XY[15]	t(2;7)	+	+	ND	ND	-	-	-	+	-	-
9	SMZL	Blood	46,XX,t(2;7)(p11;q21),i(17)(q10),der(20)t(15;20)(q12;q13)[1]/46,sl,dup(3)(q13q29)[10]/46,sl,der(16)t(3;16)(q13;p13)[8]/46,sl,der(10)t(3;10)(q11;p11)[2]/46,XX[1]	t(2;7)	+	+	ND	ND	-	-	-	+	-	+
10	SMZL	Blood	46,XY,t(2;7)(p11;q21)[3]/46,XY[24]	t(2;7)	+	+	ND	ND	-	-	-	-	-	-
11	SMZL	Bone Marrow	44,XX,t(2;7)(p11;q21),der(8;17)(q10;q10),-13,der(19)t(13;19)(q14;p13)[4]/44,sl,add(1)(q32)[10]/46,XX[6]	t(2;7)	+	+	ND	ND	-	+	-	+	-	-
12	SMZL	Blood	46,XX,t(2;7)(p11;q21)[18]/46,XX[2]	t(2;7)	+	+	ND	ND	+	-	-	-	-	-
13	SMZL	Blood	46,XY,t(2;7)(p11;q21)[19]/46,XY[1]	t(2;7)	+	+	ND	ND	-	-	-	-	-	-
14	SMZL	Blood	46,XX,t(2;7)(p11;q21),del(17)(p11)[15]/46,sl,t(4;19)(q2?2;?p13)[2]/46,XX[3]	t(2;7)	+	+	ND	ND	-	+	-	+	-	-
15	SMZL	Blood	45,XY,t(2;7)(p11;q21),dic(8;17)(p11;p11)[29]/46,XY[8]	t(2;7)	+	+	ND	ND	ND	ND	-	+	ND	ND
16	SZML	Blood	46,XX,t(2;7)(p11;q21)[10]/46,XX[10]	t(2;7)	+	+	ND	ND	-	+	-	+	-	-
17*	SMZL	Blood	46,XX,t(2;7)(p11;q21)[2]/46,sl,t(X;2)(q27;q12),del(7)(q22q34)[1]/46,sdl1,del(6)(q15q27)[6]/45,sdl2,der(8;13)(p12;q13)[3]/46,sl,add(3)(p21),del(7)(q22q34)[2]/46,XX[1]	t(2;7)	+	+	ND	ND	-	+	-	-	-	-
18	SMZL	Blood	46,XY,t(2;7)(p11;q21)[11]/46,XY[3]	t(2;7)	+	+	ND	ND	-	-	-	-	-	-
19	SMZL	Blood	46,XY,t(2;7)(p11;q21)[7]/46,XY[3]	t(2;7)	+	+	ND	ND	-	-	-	-	-	-
20	SMZL	Bone Marrow	45,XY,t(2;7)(p11;q21),der(8;17)(q10;q10)[8]/46,XY[2]	t(2;7)	+	+	ND	ND	-	-	-	+	-	-
21	SMZL	Blood	47,XX,t(2;7)(p11;q21),+12[11]/46,XX[9]	t(2;7)	+	ND	ND	ND	+	ND	-	-	-	-
22	SMZL	Blood	46,XY,t(2;7)(p11;q21)[7]/46,XY[13]	t(2;7)	+	ND	ND	ND	ND	ND	-	ND	ND	ND
23	SMZL	Blood	46,XX,t(2;7)(p11;q21)[6]/46,XX[18]	t(2;7)	+	+	ND	ND	-	+	-	-	-	-
24	SMZL	Bone Marrow	47,XX,t(2;7)(p11;q21),+12[17]/46,XX[3]	t(2;7)	+	+	ND	ND	+	-	-	-	-	-
25	SMZL	Blood	47,XX,t(2;7)(p11;q21),+3[3]/46,sl,t(1;3)(p36;p14)[7]	t(2;7)	+	+	ND	ND	-	+	-	-	-	+
26	BALT	Lung	46,XY,t(2;7)(p11;q21)[20]	t(2;7)	+	+	ND	ND	-	-	-	-	-	-
27	NMZL	Bone Marrow	44,XY,del(1)(p32p21),t(2;7)(p11;q21),der(4)t(4;8)(q35;q12),t(5;6)(q31;q25),der(8;21)(q10;q10),ins(12;2)(q15;q32q35),der(13)del(13)(q13q21)dic(13;22)(p11;p11)[18]/46,XY[2]	t(2;7)	+	+	ND	ND	-	-	-	+	-	-
28	SMZL	Blood	45,XX,t(2;7)(p11;q21),der(8;17)(q10;q10)[20]	t(2;7)	+	+	ND	ND	-	-	-	+	-	-
29	SMZL	Blood	44~45,XX,del(1)(q21q34),t(2;7)(p15;q21),-6,+der(7)t(2;7),-8,der(10)(10pter->q11::8q?::->8q?::1q?->1qter),der(11)(11pter->11q23::10q?->10q?::6q16?->6qter),-13,der(15)t(10;15)(?q;p11),-15,+17,der(17)(15q?::17p11->17q?::6p or q->6?::17q?->17qter)x2,der(20)t(3;20)(?q;q11)[20]	t(2;7)	+	+	ND	ND	-	+	-	+	+	+
30	SMZL	Blood	46,XY,t(2;7)(p12;q21),add(13)(p11),dic(12;15)(p11;q26),r(17),add(18)(q21)[4]/46,sl,add(17)(p11)[2]/46,XY[14]	t(2;7)	+	+	ND	ND	-	-	-	+	-	-
31	SMZL	Blood	46,XY,t(7;14)(q21;q11)[19]/46,XY[1]	**t(7;14) TCRA**	+	ND	-	+	-	-	-	-	-	-
32	SMZL	Blood	46,XY,t(2;7)(p11;q21),der(17)t(17;22)(p11;q12),del(22)(q12)[12]/46,sl,der(21)t(3;21)(q23;q22)[4]/46,sl,-8,der(18)t(8;18)(q2?1;q2?1)x2,-del(22)(q12),+idel(22)(q12)[2]/46,XY[2]	t(2;7)	+	+	ND	ND	-	+	-	+	+	+
33	SMZL	Blood	46,XY,t(2;7)(p11;q21)[3]/46,sl,der(6)(6pter->6q14::12p13 or q24),der(8)(8p1?->8q?13::12q22),der(12)(8qter->8q22::12p12->12q?13 or q14::?),der(17)t(8;17)(p12;p12)[14]/46,XY[4]	t(2;7)	+	+	ND	ND	-	+	-	+	-	-
34	SMZL	Bone Marrow	46,XY,del(14)(q23q32)[4]/46,XY,t(2;7)(p11;q21)[3]/45,XY,add(17)(p11),-21[2]/46,XY[23]	t(2;7)	+	+	ND	ND	-	+	-	+	+	-
35	SMZL	Blood	46,XY,t(2;7)(p11;q21),i(8)(q10),add(17)(p11),+mar1,+mar2,+mar3[cp10]/46,XY[1]	t(2;7)	+	+	ND	ND	-	+	-	+	-	+
36	SMZL	Blood	46,XX,t(2;7)(p11;q21),der(11)t(3;11)(q2?2;q13),del(17)(p11)[8]/46,sl,der(9)t(9;14)(p21;q23),-14[10]/46,sl,?t(3;18)(p21;q12)[2]	t(2;7)	+	+	-	ND	-	-	+	+	-	+
37	SmBL	Blood	46,XY,t(2;7)(p11;q21)[8]/46,XY[2]	t(2;7)	+	+	ND	ND	-	-	-	-	-	-
38	SmBL	Bone Marrow	46,XY,t(2;7)(p11;q21)[7]/47,XY,+12[1]/46,XY[2]	t(2;7)	+	ND	ND	ND	-	-	-	-	-	-
39	SmBL	Blood	46,XY,t(2;7)(p11;q21)[10]	t(2;7)	+	+	ND	ND	-	-	-	-	-	-
40	SmBL	Blood	43,X,-Y,add(1)(q32),t(2;7)(p11;q21),i(8)(q10),add(16)(q12),-17,-18,+mar[cp5]/46,XY[26]	t(2;7)	+	+	ND	ND	-	+	-	+	-	+
41	SmBL	Bone Marrow	49,XX,t(2;7)(p11;q21),+der(7)t(2;7),+12,+15[10]	t(2;7)	+	+	ND	ND	+	-	-	-	-	-
42	SmBL	Bone Marrow	46,XY,t(2;7)(p11;q21)[9]/46,XY[1]	t(2;7)	+	+	ND	ND	-	-	-	-	-	-
43	SmBL	Bone Marrow	46,XX,t(2;7)(p11;q21)[6]/46,XX,del(13)(q13q21)[4]	t(2;7)	+	+	ND	ND	-	+	-	-	-	-
44	SmBL	Bone Marrow	44,X,-X,t(2;7)(p11;q21),der(12)t(12;17)(p12;q11),-17[7]/46,XX[3]	t(2;7)	+	+	ND	ND	-	-	-	+	-	-
45	SmBL	Blood	46,XX,der(1)t(1;8)(q32;q11),t(2;7)(p11;q21),der(8)del(8)(p11)del(8)(q11)[13]/45,sl,-der(8)[7]	t(2;7)	+	-	-	ND	-	-	-	-	-	-
46	SmBL	Blood	45-46,XY,t(2;7)(p11;q21),add(6)(p22),i(8)(q10)[2],r(?17),add(20)(q13)[cp4]/46,XY,t(2;7),del(3)(p14p25),t(5;8)(q14;p22),add(6),r(?17),del(18)(q21),add(20)[3]/45-47,t(2;7),del(7)(q21q35),+der(7)t(2;7),der(8)t(8;15)(p12;q21),+der(8),-15,r(?17),add(20)[3]/46,XY[1]	t(2;7)	+	+	ND	ND	-	+	-	+	-	-
47	SmBL	Blood	45,X,ins(X;?)(q21;?),t(2;7)(p11;q21),del(3)(p11p21),i(8)(q10),der(17;18)(q10;q10)[6]/45,sl,del(18)(q22)[4]	t(2;7)	+	+	ND	ND	-	+	-	+	-	-
48	CLL	Blood	47,XY,t(2;7)(p11.2;q22.1),+der(7)t(2;7)[20]/46,XY[1]	t(2;7)	+	ND	ND	ND	-	-	-	-	ND	ND
49	CLL	Blood	46,XY,t(2;7)(p11;q21),der(14)t(3;14)(q11;p13)[2]/46,sl,i(8)(q10)[2]/46,sdl1,-der(14),+der(14)del(14)(q23q31)t(3;14)(q11;p13)[4]/46,sdl2,-i(8),+add(8)(p12)[2]	t(2;7)	+	+	-	ND	-	-	-	-	-	+
50	CLL	Blood	46,XY,t(2;7)(p11;q21)[2]/46,sl,del(13)(q14q34)[12]/46,sl,der(3)t(3;13)(p23;q21),der(13)t(3;13)del(13)(q13q21)[6]	t(2;7)	+	+	ND	ND	-	+	-	-	-	-
51	CLL	Blood	48,XX,t(2;7)(p11;q21),+12,+21[12]/48,sl,-der(2)t(2;7),+der(2)(7qter->7q21::2p11->2q22::?),add(4)(p14)[7]/46,XX[1]	t(2;7)	+	+	ND	ND	+	-	-	-	-	-
52	CLL	Blood	46,XY,t(7;14)(q21;q32)[10]	**t(7;14) IGH**	+	ND	+	ND	-	-	-	-	-	-
53	CLL	Blood	46,XY,t(2;7)(p11;q21),t(14;19)(q32;q13)[8]/46,XY[6]	t(2;7)	+	+	+	ND	-	-	-	-	-	-
54	MBL	Blood	46,XX,t(7;14)(q21;q11)[13]/46,XX[7]	**t(7;14) TCRA**	+	ND	ND	+	-	+	-	-	-	-
55	MBL	Bone Marrow	46,XY,t(7;14)(q21;q11),del(13)(q13q22)[5]/46,XY,add(19)(q13)[2]/46,XY[14]	**t(7;14) TCRA**	+	ND	ND	+	-	+	+	-	-	-
56	SLL	Blood	46,XY,t(2;7)(p11;q21)[8]/46,XY[12]	t(2;7)	+	+	ND	ND	-	-	-	-	-	-
57	SLL	Blood	46,XY,t(7;14)(q21;q32),del(13)(q13q21),add(17)(p11),add(17)(q25)[5]/46,sl,del(8)(q22q24)[4]/46,XY[6]	**t(7;14) IGH**	+	ND	+	ND	ND	+	-	+	+	-

* t(2;7)(p11;q21) and del(7q) are in two distinct clones (#17); ND: not done

Supplemental Table S4. Associations between 17p deletion and the other biological parameters in the whole cohort of patients with t(*CDK6*) or in marginal zone lymphomas (MZL) and small B-cell lymphomas (SmBL) cases. Categorical variables were compared using a Fisher's exact test and continuous variables were compared using the Mann-Whitney test (§).

Parameter	Whole cohort (n=57)			MZL/SmBL (n=47)		
	No del(17p) n=34	del(17p) n=23	*P*-value	No del(17p) n=25	del(17p) n=22	*P*-value
Prolymphocytic cells	4/17 (24%)	14/15 (93%)	<0.001	4/11 (36%)	14/15 (93%)	0.003
***TP53* mutation**	1/21 (5%)	9/15 (60%)	<0.001	1/15 (7%)	9/15 (60%)	0.005
Complex Karyotype	10/34 (29%)	18/23 (78%)	<0.001	6/25 (24%)	17/22 (77%)	<0.001
Highly Complex Karyotype	4/34 (12%)	14/23 (61%)	<0.001	2/25 (8%)	13/22 (59%)	<0.001
Number of chromosomal abnormalities. median(range)	1 (1-7)	5 (2-14)	<0.001§	2.5 (1-6)	4 (1-14)	<0.001§
13q14 deletion	9/32 (28%)	13/22 (59%)	0.028	6/23 (26%)	12/21 (57%)	0.065
8p deletion	3/34 (9%)	11/23 (48%)	0.001	2/25 (8%)	11/22 (50%)	0.002
8q gain	1/34 (3%)	6/23 (26%)	0.014	0/25 (0%)	6/22 (27%)	0.007

Supplemental Table S5. Treatment data for the 57 patients with a t(*CDK6*) translocation.

Patient ID	Age at diag (years)	Disease	Number of lines of treatment	Time to first treatment (months)	Treatments	Response	Follow-up (months)	Death
1	65	SMZL	1	41	splenectomy	CR	118	No
2	66	SMZL	0				15	No
3	69	SMZL	0				10	No
4	69	SMZL	0				5	No
5	69	SMZL	0				10	No
6	73	SMZL	0				105	No
7	72	SMZL	0				103	No
8	48	SMZL	1	6	chlorambucil	CR	190	No
9	74	SMZL	1	10	R-idelalisib	CR	31	No
10	67	SMZL	0				207	No
11	78	SMZL	3	1	R-CVP R-bendamustine idelalisib	Failure Failure PR	21	No
12	61	SMZL	1	26	R-bendamustine	Not available	46	Yes
13	65	SMZL	1	41	RDC	Treatment in progress	79	No
14	62	SMZL	0				90	No
15	86	SMZL	0				18	Yes
16	90	SZML	0				55	No
17	78	SMZL	1	48	splenectomy	CR	84	No
18	70	SMZL	1	39	R-chlorambucil	PR	45	No
19	35	SMZL	0				1	No
20	66	SMZL	0				12	No
21	85	SMZL	0				16	No
22	50	SMZL	1	53	splenectomy	CR	60	No
23	77	SMZL	0				0	No
24	76	SMZL	1	42	R-chlorambucil	Not available	55	No
25	51	SMZL	1	56	RFC	Treatment in progress	58	No
26	71	BALT	0				1	Yes
27	66	NMZL	1	0	R-CHOP	CR	15	No
28	78	SMZL	1	7	R-idelalisib	CR	44	No
29	65	SMZL	1	0	R-CHOP	Not available	4	No
30	85	SMZL	0				7	No
31	60	SMZL	0				62	No
32	63	SMZL	5	73	chlorambucil splenectomy R-bendamustine R ESHA carbo R-CHOP	PR CR Progression Failure Failure	140	Yes
33	76	SMZL	0				7	No
34	74	SMZL	0				61	No
35	79	SMZL	1	27	R-bendamustine	CR	39	No
36	86	SMZL	0				0	Yes

Patient ID	Age at diag (years)	Disease	Number of lines of treatment	Time to first treatment (months)	Treatments	Response	Follow-up (months)	Death
37	59	SmBL	0				10	No
38	65	SmBL	1	58	R-bendamustine	PR	68	No
39	46	SmBL	0				0	No
40	54	SmBL	2	3	R-CHOP R-COPADEM	Failure Failure	5	Yes
41	79	SmBL	0				70	No
42	55	SmBL	0				6	No
43	64	SmBL	0				37	No
44	76	SmBL	0				20	No
45	72	SmBL	4	50	R-fludarabine chlorambucil chlorambucil fludarabine	CR Progression Progression PR	104	No
46	81	SmBL	0				9	Yes
47	68	SmBL	2	18	Mabcampath ibrutinib	PR Treatment in progress	77	No No
48	58	CLL	4	44	R-CHOP RFC R-bendamustine ibrutinib	CR CR CR CR	159	No No No No
49	73	CLL	2	58	bendamustine chlorambucil	Progression Not evaluated (toxicity)	96	Yes
50	66	CLL	2	11	RFC R-idelalisib	CR CR	75	No
51	55	CLL	1	114	RFC	CR	137	No
52	51	CLL	0				10	No
53	86	CLL	3	2	R-chlorambucil R-CHOP GEMOX	CR Failure Failure	34	Yes
54	76	MBL	0				1	No
55	87	MBL	1	3	chlorambucil	Not available	23	No
56	51	SLL	1	3	RFC	CR	54	No
57	70	SLL	4	1	R-idelalisib R-CHOP R-GEMOX ibrutinib	Failure Progression PR Progression	11	Yes

Abbreviations: R: rituximab; F: fludarabine; C: cyclophosphamide; CHOP: cyclophosphamide + hydroxydoxorubicin + vincristine + prednisone; CVP: cyclophosphamide + vincristine + prednisone: GEMOX: gemcitabine + oxaliplatine ; COPADEM: cyclophosphamide + vincristine + doxorubicin + prednisone + methotrexate
CR: complete remission; PR: partial remission

Supplemental Table S6. A univariate analysis of overall survival in the MZL/SmBL subgroup, according to the Kaplan-Meier method.

		MZL/SmBL (n=47)			
Parameter		**OS at 5 years (%)**	**HR**	**95%CI**	***P*-value**
CD5 expression	negative	90.5	1.51	0.28-8.25	0.633
	positive	78.4			
Prolymphocytic cells	no	88.9	1.08	0.11-10.52	0.829
	yes	75.7			
del(17p)	no	87.7	2.59	0.50-13.51	0.243
	yes	79.3			
***TP53* mutated**	no	92.3	3.33	0.26-41.97	0.333
	yes	90.0			
***TP53* disrupted**	no	90.0	3.43	0.40-29.70	0.426
(mut and/or del)	yes	80.9			

HR: hazard ratio; CI: confidence interval

Supplemental Table S7. Comparison of the frequencies of genetic aberrations and CD5 expression in cases of t(*CDK6*)+ marginal zone lymphoma (MZL) vs. small B-cell lymphoma (SmBL) in the current study, and literature data on splenic MZL (SMZL).

	t(*CDK6*)+ MZL/SmBL	SMZL				
Genetic abnormality	**Current study, (n=47) frequence (%)**	**Salido et al. [1] (n=330) frequence (%)**	**Rossi et al. [2] (n=117) frequence (%)**	**Parry, et al. [3] (n=175) frequence (%)**	**Bikos et al. [4] (n=337) frequence (%)**	**Oquendo et al. [5] (n=475) frequence (%)**
Complex karyotype	49	53				
del(17p)	47	18		33		
7q deletion	2	39		19		
Trisomy 3/3q	19	25				
Trisomy 18/18q	6	10				
Trisomy 12/12q	11	8				
del(8p)	28	4				
8q gain	13	2				
14q deletion	6	3				
14q32 translocation*	0	12				
***TP53* mutation**	33		14.5	16		15
***NOTCH2* ex34 mutation**	0		21.3	10		20
***MYD88* L265P mutation**	16		5.1	4		5
IGHV* mutated*	86	86			86.7	
IGHV1-02*04	0			13	23	
CD5 expression	55	25		27		

*except t(7;14)(q21;q32) *IGH/CDK6* ** <100% homology

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