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ORIGINAL INVESTIGATION

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FISH analysis of hematological neoplasias with 1p36 rearrangements allows the definition of a cluster of 2.5 Mb included in the minimal region deleted in 1p36 deletion syndrome

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Abstract Rearrangements in the distal region of the short arm of chromosome 1 are recurrent aberrations in a broad spectrum of human neoplasias. However, neither the location of the breakpoints (BP) on 1p36 nor the candidate genes have been fully determined. We have characterized, by fluorescence in situ hybridization (FISH), the BP in 26 patients with hematological neoplasias and 1p36 rearrangements in the G-banding karyotype. FISH allowed a better characterization of all samples analyzed. Nine cases (35%) showed reciprocal translocations, 15 (58%) unbalanced rearrangements, and two (7%) deletions. We describe two new recurrent aberrations. In 18 of the 26 cases analyzed the BP were located in band 1p36, which is 25.5 Mb long. In 14 of these 18 cases (78%) and without distinction between myeloid and lymphoid neoplasias, the BP clustered in a 2.5 Mb region located between 1p36.32 and the telomere. Interestingly, this region is contained in the 10.5 Mb cluster on 1p36.22-1pter defined in cases with 1p36 deletion syndrome. The 2.5 Mb region, located on 1p36.32-1pter, has a higher frequency of occurrence of tandem repeats and segmental duplications larger than 1 kb, when compared with the 25.5 Mb of the complete 1p36 band. This could explain its proneness for involvement in chromosomal rearrangements in hematological neoplasias.

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Introduction

Cytogenetic analysis of various types of human malignancies, ranging from virtually all types of solid tumors, to leukemias and myeloproliferative disorders, have shown the non-random involvement of abnormalities of chromosome 1p36 (Atkin 1986; Dave et al. 1995; Weith et al. 1996; Mitelman et al. 1997). However, the location of the breakpoints (BP) on 1p36 and the candidate genes involved have not been fully determined (Johansson et al. 1999; Rowley 1999; Mitelman 2000). The affected regions frequently span many megabases of DNA, hampering the identification of critical genes (Boei et al. 1998; Schutte et al. 2001). Moreover, a decreased disease-free survival and a shorter overall survival have been correlated with breaks in the 1p32–p36 region (Levine et al. 1988; Offit et al. 1991).

Deletions of the short arm of chromosome 1 (1p36) are present in approximately 30% of neuroblastoma (NB) cases, the most common malignant solid childhood tumor derived from the neural crest (Brodeur et al. 1977; Gilbert et al. 1982; Weith et al. 1989). The biological significance of 1p loss has been indicated by an interesting experiment: Bader et al. (1991) transferred normal

1p material into a NB cell line carrying a 1p deletion and found suppression of tumorigenicity with induction of cell differentiation (Bader et al. 1991). Deletions of 1p are poor prognosis factors in NB (Caron et al. 1996; Maris et al. 2000).

In hematological neoplasias, the most frequent abnormalities involving 1p36 are unbalanced translocations, presented as add(1)(p36) at the karyotype level, in both myeloid and lymphoid neoplasias (Mitelman et al. 2003). Nine recurrent balanced rearrangements have been reported involving band 1p36 in hematological neoplasias, the most frequent being t(1;3)(p36;q21), which shares, with 3q21q26 syndrome, its association with myeloid malignancies and, in some cases, similar clinicopathological features (Shimizu et al. 2000; Mitelman et al. 2003; Lahortiga et al. 2004). Moreover, loss of heterozygosity (LOH) of the 1p36 region has been reported in 47% of the patients with chronic myelocytic leukemia (CML) in transformation, in 30% of the patients with progression of myelodysplastic syndrome (MDS) to acute myeloid leukemia (AML), and in 13% of patients with MDS (Mori et al. 1998, 2003; Hofmann et al. 2001). All these studies have contributed to the definition of the smallest region of overlap (SRO) located on 1p36, suggesting the location, in this region, of candidate genes involved in the malignant proliferation of myeloid precursor cells (Hofmann et al. 2001). Region 1p36 contains several genes that are involved in the regulation of cell proliferation and differentiation, including several genes coding for hematopoietic growth factor receptors, and that could be considered candidate genes (Hofmann et al. 2001). Characterization of the BP could help to identify genes with important roles in cancer development and contribute to the development of tailored treatment (Varga et al. 2001).

We report the characterization, by fluorescence in situ hybridization (FISH), of the BP in 26 patients with hematological neoplasias and 1p36 rearrangements. This analysis has allowed us to define a region of 2.5 Mb that is located between 1p36.32 and the telomere, a region in which the BP of 78% of cases with a BP located in band 1p36 are clustered. This region shows characteristics, such as the presence of genomic repeats, that could explain its proneness for involvement in chromosomal rearrangements.

Materials and methods

Patients

Twenty-six patients with hematological neoplasias and 1p36 rearrangements at the G-band level analyzed at the University of Navarra and at the University of Salamanca (Spain) were included. Of these, 16 patients had myeloid neoplasias, and 10 had lymphoid neoplasias: five AML, eight MDS, three myeloproliferative disorders (MPD), four ALL, two multiple myeloma (MM) and four NHL. Genetic characterization is included in Table 1. All samples were obtained with informed consent.

G-banding karyotype

Cytogenetic studies were performed on unstimulated short-term bone marrow cultures. G-banded karyotypes are shown in Table 1 (described according to ISCN 1995, see Mitelman 1995).

Fluorescence in situ hybridization

Fluorescence in situ hybridization (FISH) studies were performed with 51 bacterial artificial chromosomes (BACs) and P1 artificial chromosomes (PACs) located between 1p32.3 and 1p36.33 (Fig. 1). Sixty-five BAC and PAC clones were also used to characterize the BP in the partner chromosomes and in other rearrangements found in the karyotypes. All the clones were designed according to the current mapping data and were obtained from libraries from the Roswell Park Cancer Institute (Buffalo, N.Y.). The DNA from clones was extracted by using the Qiaprep Spin Miniprep kit (Qiagen, Hilden, Germany). Probes were labeled with Spectrum Green and Spectrum Orange (Vysis, Downers Grove, Ill., USA) by nick translation and were used pair-wise. Commercial probes employed were: TelVysion 1p, LSI TEL/AML1 ES, LSI CBFB dual colour rearrangement probe, LSI BCR/ABL ES, LSI IgH/CCDN1, LSI IgH/BCL2, LSI MLL dual colour rearrangement probe, and WCP for chromosomes 1, 4, 6, 8, and 13 (Vysis). FISH analysis was performed on bone marrow samples as previously described (Odero et al. 2001).

Spectral karyotyping analysis

The SkyPaint DNA kit for spectral karyotyping (SKY) and the hybridization reagents were obtained from Applied Spectral Imaging (Mannheim, Germany). Slides for spectral karyotyping were hybridized with the probe cocktail following the manufacture's protocol.

Bioinformatic analysis

The DNA sequences and their annotations were retrieved from the EnsEMBL Genome Browser Databases, making use of the EnsEMBL Perl Application Programming Interface (Hubbard et al. 2002). Pattern finding was carried out by using the fuzznuc program from the European Molecular Biology Open Software Suite (Rice et al. 2000). Segmental Duplications were computed from tables that are available at the University of Santa Cruz Genome Browser (http://genome.ucsc.edu/, version

Table 1 Genetic characterization of 26 patients with 1p36 rearrangements (*Dx* diagnosis, *R* relapse)

Case	Sex/age	Diagnosis	Status*	G-banded karyotype	Revised karyotype after FISH analysis
1	F/87	AML (M4)	Dx	t(1;21)(p36;q22)	t(1;21)(p36.33;q22).
2	F/66	AML (M3)	Dx	der(1)t(1;?)(p36;?)	der(1)ins(1;1)(p36.33;?).ish ins(1)(p36.33)(wcp1+)
3	M/39	AML (M3)	Dx	der(1)t(1;?)(p36;?)	der(1)ins(1;1)(p36.33;?).ish ins(1)(p36.33)(wcp1+)
4	M/91	AML	Dx	der(1)t(1;?)(p36;?),der(6)t(6;?)(p2?1;?), der(12)t(12;?)(p13;?)	der(1)dup(1)(p32.3p34.2)t(1;6)(p34.1;p21),der(12)t(1;12)(p32;p13),der(6)dup(6)(p21.1p24)t(1;6)(p34.1;p21).
5	F/67	AML (M6)	R	t(1;6)(p36;q24)	t(1;6)(p36.32;q23.2)
6	F/66	MDS (RAEB-2)	R	del(1)(p36), der(12)t(12;?)(p13;?)	t(1;12)(p36.11;p13.2)
7	F/56	MDS (RA)	R	t(1;5)(p32;q35)	t(1;5)(p34.1;q35)
8	M/49	MDS	Dx	del(1)(p36)	del(1)(p34.3)
9	F/62	MDS (RAEB-2)	Dx	der(1)t(1;?)(p36;?)	der(1)amp(1) p36.23p36.33).ish der(1)amp(1)(p36.23p36.33)(RP1-202O8amp,RP5-890O3amp)
10	F/63	MDS (RARS)	Dx	der(1)t(1;?)(p36;?)	der(1)dup(1)(p32.3p36.32).ish der(1)dup(1)(p32.3p36.32)(RP11-116M11++,RP1-163G9++)
11	M/74	MDS	Dx	der(1)t(1;?)(p36;?)	der(1)dup(1)(p32.3p34.1).ish der(1)dup(1)(p32.3p34.1)(RP11116M11++,RP51034F7++)
12	M/77	MDS	Dx	der(1)t(1;?)(p36;?)	der(1)amp(1)(p32.3p34.1).ish der(1)amp(1)(p32.3p34.1)(RP11116M11amp,RP51034F7amp)
13	M/77	MDS (RAEB-2)	Dx	t(1;3)(p36;q21)	t(1;3)(p36.32;q21.3)
14	F/78	MPD (ET)	R	t(1;3)(p36;q21)	t(1;3)(p36.3?2;q21)
15	M/59	MPD (CML)	R	t(1;6)(p36;p21)	t(1;6)(p34.2;p2?2)
16	F/64	MPD (CML)	Dx	t(1;9;22)(p36;q34;q11)	t(1;9;22)(p36.23;q34;q11)
17	F/17	ALL (L2)	Dx	del(1)(p36)	del(1)(p34.3)
18	M/75	ALL	Dx	der(1)t(1;?)(p36;?)	der(1)t(1;5)(p36.33;?q)der(1)del(1)(p1?)(q1?)
19	M/64	ALL(L3)	Dx	der(1)t(1;?)(p36;?)	der(1)t(1;1)(p36.33;q21)
20	F/34	ALL (T-ALL)	Dx	der(1)t(1;?)(p36;?)	t(1;2)(p34.3;p1?2)
21	F/60	LPD (MM)	Dx	+der(1)t(1;?)(p36;?)	+t(1;6)(p3?6;p?21)
22	M/55	LPD (MM)	Dx	der(1)t(1;18)(p36;?q21)	der(1)t(1;18)(p36.33;q?21)
23	F/49	NHL	Dx	t(1;7)(p36;q22)	der(1)t(1;1)(p36.33;q21)
24	M/88	NHL	Dx	der(1)t(1;?)(p32;?)	der(1)t(1;1)(p36.33;q21
25	M/70	NHL	Dx	der(1)t(1;5)(p36;q13)	der(1)t(1;5)(p36.32;q13)
26	F/12	NHL	Dx	der(1)t(1;?)(p36;?)	dup(1)(?)

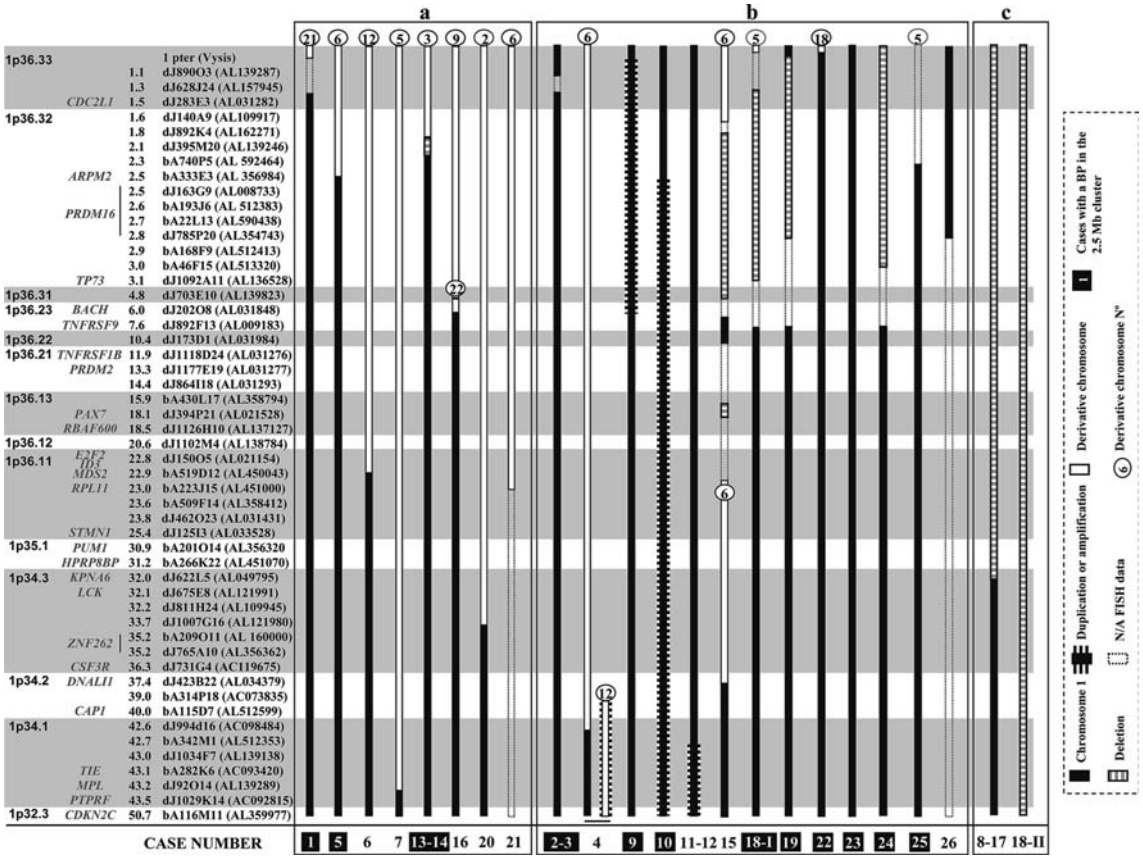


Fig. 1 Rearrangements with FISH results. *Left* An integrated map of the 1pter-1p32.3 region including the physical positions (in Mb) and locations of the clones used in this study (*brackets* GenBank accession number) and the candidate genes in cancer reported in the literature. *Vertical bars* Rearrangements found for each case distributed in balanced translocations (*a*), unbalanced rearrangements (*b*), and terminal deletions (*c*), *black bars* chromosome 1 regions, *striped gray bars* chromosome 1 deletions, *striped black bars* regions found to be duplicated or amplified, *dotted bars* regions where no FISH is data available, *white bars* regions translocated to a derivative chromosome listed within the *circle* at the telomeric end, *black boxes* cases with a BP located in the 2.5-Mb cluster

hg15, April 2003) and that make use of data in Bailey et al. (2001). Statistical analysis was performed with R (R Foundation for Statistical Computing, Austria).

Results

We have characterized, by FISH, the BP in bone marrow samples from 26 patients with hematological neoplasias and 1p36 rearrangements at the G-banding level. Nineteen cases had a complex karyotype, with more than three unrelated aberrations. Nine cases showed reciprocal translocations, and 15 had unbalanced translocations, three of them being recurrent and with complex rearrangements. One of the cases with an unbalanced rearrangement (case 18) had another clone with a terminal deletion. In addition, two cases showed a terminal deletion involving the same region. Revised karyotypes of the patients after FISH analysis are given in Table 1. A diagram of the rearrangements found in the 1pter-1p32.3 region is shown in Fig. 1. All cases were further characterized. During the study, the relative location for each clone used was adjusted on the basis of new sequencing information becoming available. The characterization of each case allowed the classification of the rearrangements into reciprocal translocations, unbalanced rearrangements, or terminal deletions.

Reciprocal translocations

Nine cases had reciprocal translocations (Table 1, Figs. 1a, 2a, b). The chromosome regions involved in the translocations with 1p were 2p, 3q (two cases), 5q, 6p, 6q, 9q, 12p, and 21q. Two of these cases (6 and 13) have been previously reported (Odero et al. 2002; Lahortiga et al. 2004).

Unbalanced translocations

Fifteen cases had unbalanced translocations, and in this group, three recurrences could be determined (Table 1, Figs. 1b, 2c–h). Cases 2 and 3, both with a diagnosis of AML M3, showed (at the G-banding level) a chromosome 1 with unidentified genetic material that was inserted at 1p36, and that FISH analysis with a chromosome 1 paint probe identified as a duplicated

region of chromosome 1 inserted between the two clones RP1-283E3 and PR5-890O3. However, no more material was available for further analysis.

In six patients, FISH detected either duplications (cases 4, 10, 11, and 26) or amplifications (cases 9 and 12) of 1p probes (Fig. 2c–e). Interestingly, in cases 11 and 12, both with a diagnosis of MDS, the same region located between 1p34.1 (43.0 Mb) and 1p32.3 (50.7 Mb) was duplicated in case 11 and amplified in case 12. Case 4 showed a highly complex translocation involving chromosomes 1, 6, and 12, with some regions being duplicated. There was a duplication in the der(1), from 1p32.3 to 1p34.2, and another duplication in chromosome 6, from 6p21.1 to 6p24.1. Therefore, a three-way translocation could have taken place among chromosomes 1p34.1, 6p21.1, and 12p13 (Fig. 2e). In case 26, the G-banding karyotype suggested that the 1p region was duplicated, but this could not be confirmed by FISH because of lack of material. Case 15 showed a complex translocation between chromosomes 1 and 6, with the presence of interstitial deleted regions (Fig. 2f).

Another recurrence was found in cases 19, 23, and 24: the duplication of 1q21 and the formation of an unbalanced translocation der(1)t(1;1)(p36;q21). However, the three cases showed differences regarding the presence or absence of a deletion in the 1p36 region and in its length: only case 24 had a terminal deletion, whereas case 19 did not have a terminal deletion, confirmed by the presence of the subtelomeric 1p region on the 1qter, and case 23 had the unbalanced translocation with no deletion of the 1p36 region (Fig. 2g).

Terminal deletions

Cases 8 and 17 had the same terminal deletion del(1)(p34.3) (Fig. 1c). Moreover, case 18 had a second clone with a chromosome der(1) that carried a terminal deletion on both the p and q arms (Fig. 2h).

Bioinformatic analysis

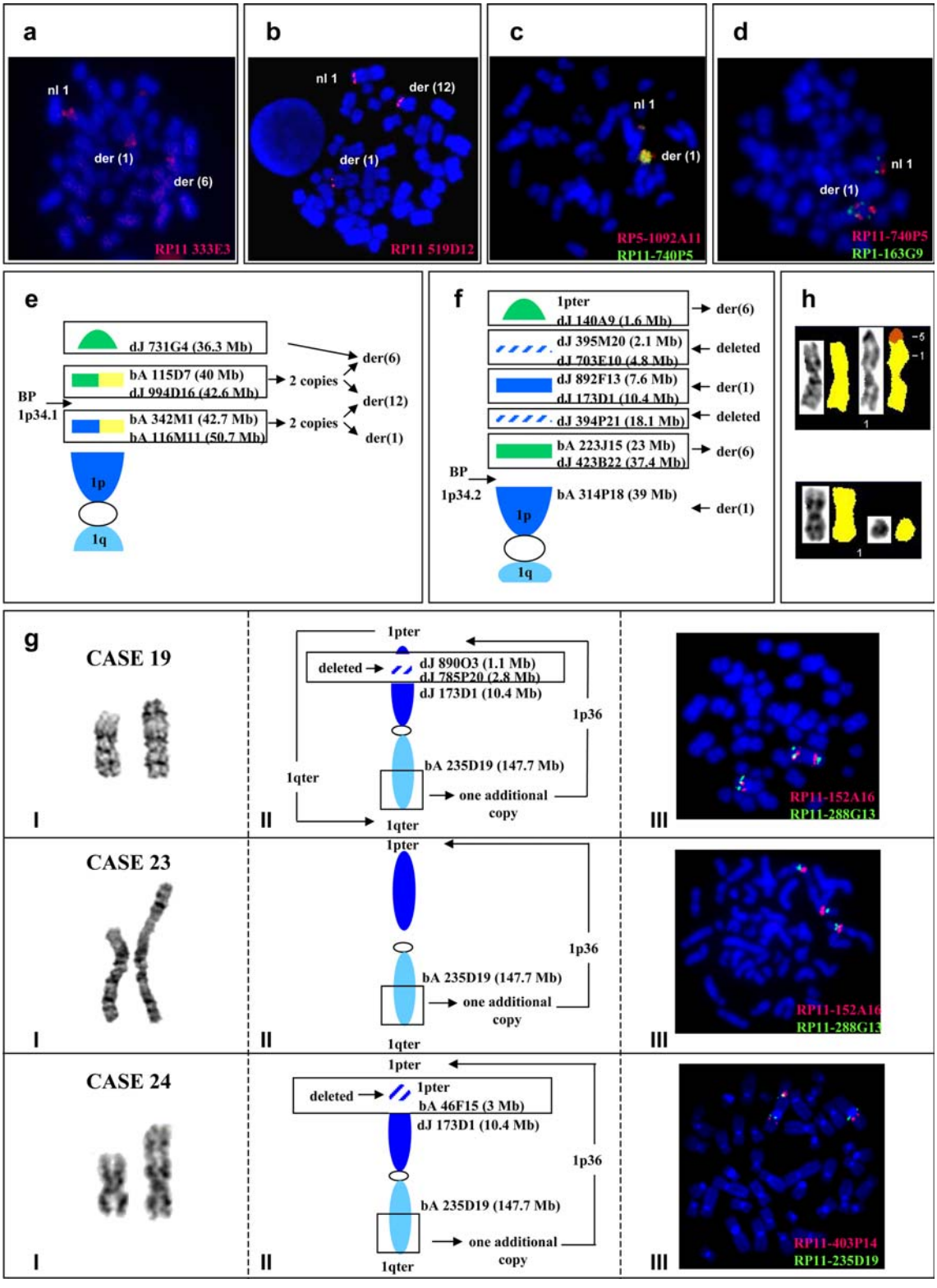
In 18 of the 26 cases analyzed, BP were located in band 1p36, which extends for 25.5 Mb. In 14 of these cases (77.8%, 14/18), eight with myeloid and six with lymphoid neoplasias, the BP clustered in a 2.5-Mb region located between 1p36.32 and the telomere (Fig. 1). Comparison by bioinformatics analysis of the most telomeric 2.5 Mb region, in which the BP of our patients cluster, and of the complete 1p36 band showed that the 2.5 Mb region has distinctive characteristics that could account for the large number of chromosomal rearrangements found. This 2.5 Mb region was richer in tandem and inverted repeats detected by the program TRF, as annotated in Ensemble (Benson 1999). The local percentage of sequence covered by these repeats, considering contiguous windows of 100 kb of genomic DNA, reached values as high as 20%, whereas analogous measurements for nearby sequences located in the rest of the 1p36 band were around 5%. We also observed a statistically significant larger number of segmental duplications in this 2.5 Mb region. As shown in Table 2, we computed the total number of duplicons larger than 1 kb in the most telomeric cytogenetic band of the short arm of each chromosome. Region 1p36 did not show differences in comparison with the rest of the bands analyzed. However, the 2.5 Mb region, in which the BP in our patients clustered, spanned the most telomeric sub-band within 1p36 (i.e. 1p36.33). Therefore, we computed the number of segmental duplications present at the most telomeric sub-band of each chromosome. As shown in Table 2, the percentage of segmental duplications per megabase is significantly higher in 1p36.33 than in the rest of the short arms.

Fig. 2 FISH analysis of some of the cases studied with probes for chromosome 1. **a** FISH analysis of case 5 with BAC 333E3. **b** FISH analysis of case 6 with BAC 519D12. **c** FISH analysis of case 9 with BAC 740P5 and PAC 1092A11. **d** FISH analysis of case 10 with BAC 740P5 and PAC 163G9. **e, f** Representation of FISH results from cases 4 and 15, respectively. **g** Analysis of cases 19, 23 and 24: partial G-banding of chromosomes 1 (*I*), representation of FISH results (*II*), and FISH analysis with probes from 1q (*III*). **h** SKY analysis of the two clones detected in case 18

Discussion

FISH analysis of 26 samples of patients with hematological neoplasias and 1p36 rearrangements in the G-banding karyotype has allowed us to define a 2.5Mb region located between 1p36.32 and the telomere, a region in which the BP of 78% of cases with a BP located in the band 1p36 cluster. There was no distinction between myeloid and lymphoid neoplasias. In our search for sequence elements that might account for the increased frequency of BP in the most telomeric 2.5 Mb of 1p36, we have found that this region is richer in tandem repeats and, most notably, is particularly rich in segmental duplications larger than 1 kb. Segmental duplications are known to be involved in chromosomal rearrangements responsible for human disease and have been recently shown to mediate the genesis of recurrent chromosomal translocations, such as t(9;22)(q34;q11) involving *BCR* and *ABL* or t(12;15)(p13;q25) involving *ETV6* and *NTRK3* (Ji et al. 2000; Osborne et al. 2001; Pujana et al. 2001; Saglio et al. 2002). In this regard, it is interesting to note that this feature is circumscribed to 1p36.33, which constitutes most of the 2.5 Mb region frequently rearranged in our samples, whereas the whole of 1p36 is not different from other telomeric bands. Thus, the particular enrichment of this region in segmental duplications might explain why so many BP are clustered here.

Interestingly, this 2.5 Mb region is contained in the 10.5 Mb cluster on 1p36.22-1pter defined in cases with 1p36 deletion syndrome. Monosomy 1p36 is the most



common congenital terminal deletion syndrome. Patients present multiple anomalies and mental retardation (Shapira et al. 1997; Shaffer et al. 2001). Heilstedt et al. (2003) have constructed a contig of overlapping large insert clones from the most distal 10.5 Mb of 1p36 (Fig. 3a) to evaluate the deletion sizes in various patients with 1p36 syndrome and have narrowed the BP region to 0.5 Mb bins (Heilstedt et al. 2003). Deletion sizes range widely, but most BP (12.5%) cluster 4.0–4.5 Mb from the telomere. Our BP clusters in the first 2.5 Mb and is included in the 10.5 Mb region defined for 1p36 deletion syndrome, although none of our BP are located

Table 2 Total number of segmental duplication (*Dups*) per megabase (*Mb*) in the most telomeric cytogenetic bands and sub-bands of the short arm of all chromosomes

Band	Size (Mb)	Dups	Dups/Mb	Sub-band	Size (Mb)	Dups	Dups/Mb
1p36	27.5	953.0	**34.7**	1p36.33	2.5	516.0	**206.4**
2p25	12.0	79.0	6.6	2p25.3	4.0	42.0	10.5
3p26	8.2	20.0	2.4	3p26.3	2.8	15.0	5.4
4p16	11.0	631.0	57.4	4p16.3	4.5	284.0	63.1
5p15	18.4	251.0	13.6	5p15.33	4.5	104.0	23.1
6p25	7.0	145.0	20.7	6p25.3	2.3	76.0	33.0
7p22	6.9	471.0	68.3	7p22.3	2.6	113.0	43.5
8p23	12.5	891.0	71.3	8p23.3	2.3	130.0	56.5
9p24	9.0	125.0	13.9	9p24.3	2.2	42.0	19.1
10p15	6.8	62.0	9.1	10p15.3	3.1	48.0	15.5
11p15	21.7	475.0	21.9	11p15.5	2.8	123.0	43.9
12p13	14.7	374.0	25.4	12p13.33	3.1	15.0	4.8
16p13	16.7	658.0	39.4	16p13.3	6.4	199.0	31.1
17p13	10.7	124.0	11.6	17p13.3	3.1	78.0	25.2
18p11	15.3	477.0	31.2	18p11.32	2.9	71.0	24.5
19p13	23.3	475.0	20.4	19p13.3	6.9	184.0	26.7
20p13	5.0	12.0	2.4	20p13	5.0	12.0	2.4
		Mean	26.5			mean	37.3
		Mean + 3 SD	**91.2**			Mean + 3 SD	**178.1**

in the most frequent 0.5 Mb (4.0–4.5 Mb) region defined by Heilstedt et al. (2003). The mechanism involved in deletions is frequently associated with duplications affecting the region. Therefore, the high frequency of duplications detected in our series by bioinformatic analysis of the 1p36 region might explain the high incidence of deletions in 1p36 deletion syndrome.

Several studies have focused on the definition of the SRO in diverse neoplasias in order to determine candidate oncogenes and/or tumor suppressor genes (TSGs) that could explain the proneness of the 1p36 region for involvement in chromosomal rearrangements. Our defined region of 2.5 Mb partially overlaps with the SRO on 1p36 reported for disorders such as NB or MDS in transformation (Fig. 3).

LOH has been widely studied in NB. The 2.5 Mb region defined in our patients partially overlaps with the consensus SRO for NB defined by Cheng et al. (1995) and Martinsson et al. (1995) (Fig. 3b, I). This region was further refined first by White et al. (1995; Fig. 3b, II) and later by Bauer et al. (2001; Fig. 3b, III). These refined regions show no overlap with our 2.5 Mb region; however, a study with an larger number of patients with NB (205 patients) has reported two SRO in which two suppressor loci involved in the pathogenesis of NB may be located; both regions include the 2.5 Mb region defined in our patients (Fig. 3b, IV-1, IV-2; Caron et al. 2001).

The 2.5 Mb region is contained in the most distal SRO of the two defined by Mori et al.(2003) for patients with MDS in transformation (Fig. 3 c-I, c-II). Mori et al. (1998, 2003) have suggested that putative TSGs that frequently play an important role in the evolution to blast crisis and in the progression of MDS to AML may reside in this region. Our region shows no overlapping with the SRO defined by LOH analysis in CML in transformation (Fig. 3 d; Mori et al. 1998), or in patients with MDS (Fig. 3 e; Hofmann et al. 2001). Differences from our series could be attributable to the different techniques used in the analysis. Moreover, the most telomeric microsatellite sequence included in both studies was D1S243 located at 2.1 Mb and was deleted in 63% and 20% of the patients with an informative loci, in CML and MDS, respectively (Mori et al. 1998; Hofmann et al. 2001); therefore, many of the patients included in these studies could have deletions in the 2.5 Mb region defined in our study.

This delimited region could be helpful for the search for putative candidate genes involved in hematological neoplasias. Some of these genes are located in the integrated 1p36 map in Fig. 1 (Mori et al. 1998; Spieker et al. 2001; Mori et al. 2003). Among these, *CDC2L1*, which codes a protein kinase implicated in apoptotic signaling, is located extremely distally and is hence most likely to be disrupted (Dave et al. 1999). A deletion of this gene locus has been observed in 88.5% of NHL cases containing 1p36 abnormalities, although the functional role of deletions of *CDC2L1* in NHL is still under consideration (Dave et al. 1999). *TP73* has also been considered a strong candidate TSG. TP73 protein has a structural and functional homology to TP53, including the ability to promote apoptosis when overexpressed in vitro and to up-regulate *TP53*-responsive genes involved in cell-cycle control, such as *CDKN1A* (Jost et al. 1997; Inokuchi et al. 2001).

Furthermore, three recurrences could be established after FISH analysis. To our knowledge, two of them have never been reported before: a der(1)ins(1;1)(p36.33;?) in two cases with AML-M3, and the duplication/amplification of the 1p32.3–p34.1 region in two cases with MDS. Moreover, two cases with NHL (cases 23 and 24) and one with ALL-L3 (case 19) showed a

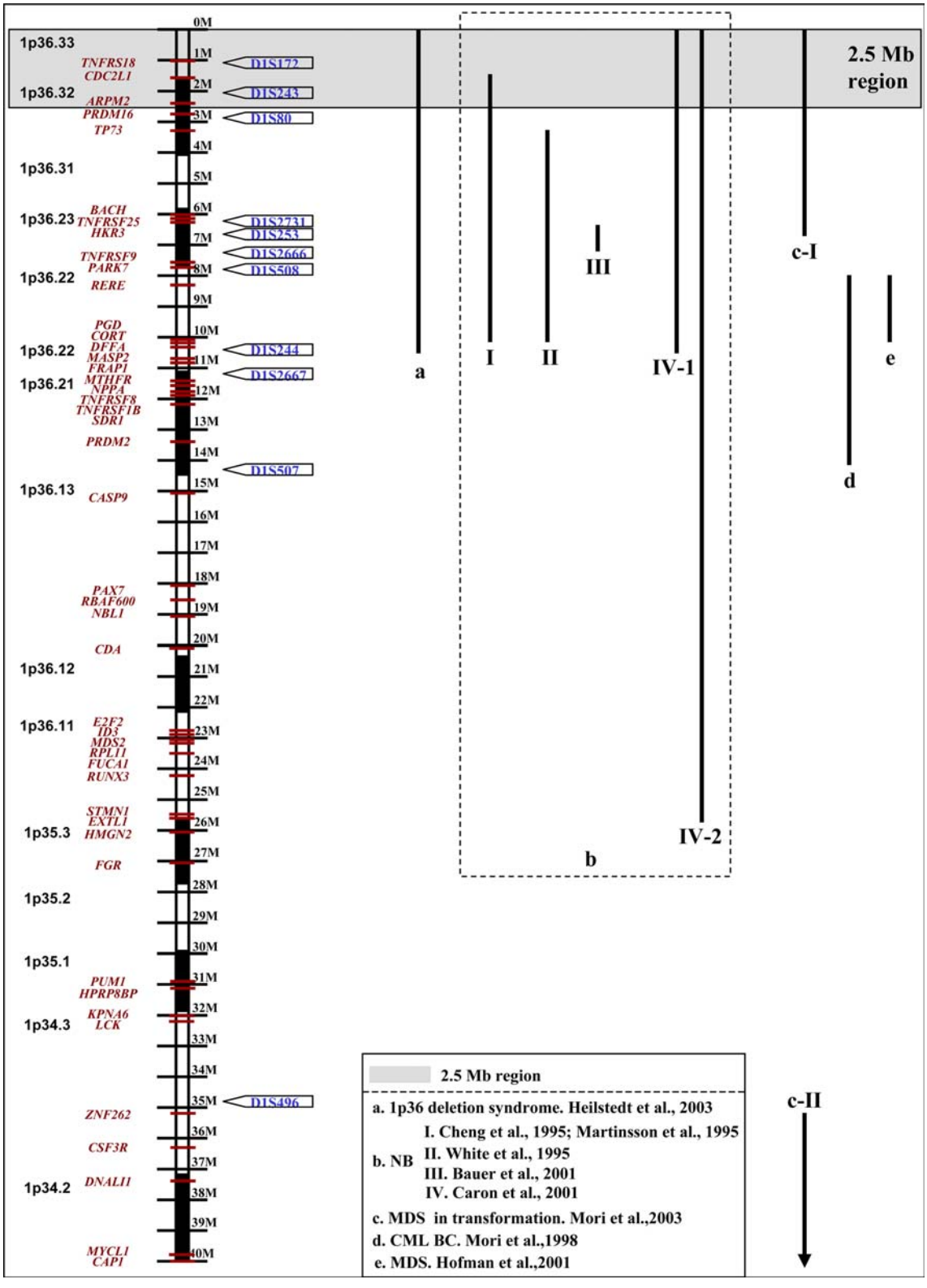


Fig. 3 Comparison of 1p SRO among various human cancers and in 1p36 deletion syndrome. *Left* An integrated map of the 1pter-1p34.2 region, including the physical position (in Mb), the location of candidate cancer genes reported in the literature, and the locations of the markers used in several studies (*vertical bars right* extent of SRO)

der(1)t(1;1)(p36.33;q21). Lestou et al.(2003) have reported the characterization, by FISH, of this recurrent translocation in NHL. The formation of this translocation includes the deletion of 1p36 and the duplication of 1q21 and is considered to be a non-random event in

NHL, suggesting that a deletion-duplication mechanism is involved in lymphoma progression (Lestou et al. 2003). Therefore, the high frequency of duplications detected in our series by bioinformatic analysis in the 2.5 Mb region might explain this mechanism. However, only case 24 has a true terminal deletion similar to those described by Lestou et al. (2003). All three cases showed duplication of the 1q21 region. Although the probes that we have used for the 1q region are different from those used by Lestou et al. (2003), the duplicated region seems to be the same (Fig. 2g; Lestou et al. 2003).

In conclusion, the characterization by FISH of 26 patients with hematological neoplasias and 1p36 rearrangements has allowed us to define a 2.5 Mb cluster region on 1p36.32, a region in which the BP of 78% of the cases analyzed (both myeloid and lymphoid) are located. This region, included in the 10.5 Mb minimal region deleted in 1p36 deletion syndrome, shows characteristics, such as the presence of genomic repeats, that could explain its proneness for involvement in chromosomal rearrangements.

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