Haematologica 2000; 85:250-255
original paper

Malignant Lymphomas

Chromosomal aberrations evaluated by CGH, FISH and GTG-banding in a case of AIDS-related Burkitt's lymphoma

ANNALISA ZUNINO,* SILVIA VIAGGI,° LAURA OTTAGGIO,* GILBERTO FRONZA,* ANDREA SCHENONE,# SILVIO RONCELLA,@^ ANGELO ABBONDANDOLO*°

**Mutagenesis Laboratory, National Cancer Institute (IST), Genoa, Italy; °Department of Oncology, Biology and Genetics, University of Genova; #Biophysics Laboratory, National Cancer Institute (IST), Genoa; @Unit of Pathologic Anatomy, S. Andrea Hospital, La Spezia; ^Clinical Immunology Division, National Cancer Institute (IST), Genoa, Italy*

ABSTRACT

Background and Objectives. **We have previously reported on a complex chromosome rearrangement [der(17)] in a B-cell line, BRG A, established from an AIDS patient with Burkitt's lymphoma (BL). The aim of the present study was the definition of der(17) composition and the identification of complete or partial chromosome gains and losses in two cell clones (BRG A and BRG M) derived from this patient.**

Design and Methods. **We applied comparative genome hybridization (CGH) to detect the DNA misrepresentations in the genome of the two cell clones. Findings from CGH and banding analysis could then direct the choice of probes for chromosome painting experiments to elucidate der(17) composition.**

Results. **CGH analysis identified gains of chromosomes 1q, 7q, 12q, 13q, 15q, 17p, 20p,q and losses of chromosomes 3p and 5q in BRG A and gain of chromosome 1q and loss in chromosome 6q in BRG M. Some of the detected alterations had already been described in lymphomas, while others appeared to be new. The combination of these techniques allowed a precise definition of der(17), composed by translocated regions from chromosomes 12 and 15.**

Interpretation and Conclusions. **We demonstrated CGH to be a powerful tool in the identification of recurrent chromosome aberrations in an AIDS-related BL and in ascertaining the origin of marker chromosomes. We were also able to identify a different pattern of aberrations and assess an independent sequence of events leading to the 1p gain in the two subclones.**

Key words: CGH, FISH, Burkitt's lymphoma, AIDS, cytogenetics

Correspondence: Annalisa Zunino, M.D., Mutagenesis Laboratory, National Cancer Institute (IST), largo Rosanna Benzi 10, 16132 Genova, Italy. Phone: international +39-010-5600251 – Fax: international +39-010-5600992 – E-mail: zuninoa@hp380.ist.unige.it

Comparative genomic hybridization (CGH) is a molecular cytogenetic method that makes it possible to screen indirectly for copy number aberrations throughout the genome in a single hybridization.[1,2] CGH is essential in the cytogenetic study of solid tumors, but also has a potential clinical use in the diagnosis of recurrent chromosome aberrations in certain leukemias and lymphomas[3] and in ascertaining the origin of marker chromosomes.[4]

In Burkitt's lymphoma (BL), both sporadic and endemic, the most consistent cytogenetic abnormalities are the 8;14 translocation and its variants t(2;8) and t(8;22). In addition to these known translocations with pathologic significance, additional recurrent structural and numerical changes have been described which may have a role during the later stages of disease development. To this class of alterations belong some structural aberrations of chromosomes 1 and 6, and trisomy of 7, 12, 13, 17q, 18 and 21.[5,6] The most common of these abnormalities is a partial duplication of 1q.[6,7] Similar chromosome abnormalities have been reported in AIDS-related BLs.[7]

We previously reported on a complex chromosome rearrangement that could have a functional significance in an IgA-expressing B cell line (BRG A) established from an AIDS patient with BL.[8] The rearranged chromosome [der(17)] was apparently composed of 17q, of a partially deleted 17p and of other material of 17p origin that was interspersed with regions that we could not identify because of lack of a clear banding pattern. A functional chromosome 17 centromere was contained in der(17) together with two additional centromeres of unknown origin that were by all evidence inactive.

Given the complexity of the BRG A karyotype, we have re-examined this cell line by CGH, to identify complete or partial chromosome gains and losses. The results of CGH were compared with chromosome banding analysis and enriched by using chromosome painting with specific probes. The same analysis was also performed on BRG M, the IgM expressing cell clone from which BRG A derived *in vivo* by an isotype switch, as demonstrated by identity in *c-myc* and IgH chain gene rearrangement.[9,10]

Design and Methods

Cell lines

The case of patient BRG, a 30-year old female seropositive for HIV, has been reported in detail previously.[9,10] A B-cell line was derived spontaneously from the patient's peripheral blood mononucleated cells. At diagnosis the majority of cells expressed IgM and a minority were positive for surface IgA1. Sublines of cells expressing only IgM (BRG M) or IgA1 (BRG A) were obtained by cell sorting separation.[9]

Comparative genomic hybridization

Hybridization was performed as described elsewhere.[2] Briefly, normal human genomic DNA (control DNA) was labeled with digoxigenin-11-deoxyuridine triphosphate (dig-11-dUTP, Boehringer Mannheim, Germany), and B-cell DNA (test DNA) was labeled with biotin-16-deoxyuridine triphosphate (bio-16-dUTP, Boehringer Mannheim) by standard nick translation reactions. DNase I concentration was adjusted to result in an average fragment size of 200 to 1,500 bp. Then, 250 ng of labeled test DNA, 250 ng of labeled control DNA, and 30 µg of human Cot-1 DNA (GIBCO-BRL-Life Technologies, Italy) were co-hybridized to slides with metaphases prepared from blood lymphocytes of a healthy female donor. After hybridization for 3 days and post-hybridization washes, test and control DNA were detected with fluoresceinated (FITC)-avidin (Vector, USA) and anti-dig rhodamin (TRITC)-conjugated antibody (Boehringer Mannheim), respectively. Chromosomes were counterstained with 4,6-diamidino-2-phenylindole (DAPI), resulting in a Q-banding-like pattern that was used for chromosome identification.

Digital image analysis, image acquisition, processing, and evaluation were performed as described elsewhere.[2] Images were acquired using an epifluorescence microscope (Eclipse E800, Nikon, Japan) equipped with a CCD camera (Cohu, USA). DAPI, FITC and TRITC images of metaphase cells were acquired with selective single-bandpass filters. Ratio profiles of individual chromosomes were obtained with dedicated software (Cytovision, Applied Image, USA). For each experiment, the mean ratio profiles of ten metaphase cells were calculated. Thresholds for the identification of imbalances were defined as 0.75 (lower threshold) and 1.25 (upper threshold).[2] Chromosomes or chromosomal regions with a fluorescence ratio outside this interval were considered to be under- or over-represented, respectively.

Fluorescence in situ hybridization

Sample preparation. C-metaphases were obtained by standard procedures, using 0.1 µg/mL colcemid (GIBCO-BRL) for 2 hours. Samples for hybridizing procedures and GTG-banding were fixed in Carnoy's solution (methanol:glacial acetic acid 3:1), splashed, dried, and aged 3-7 days.

Molecular probes. The following probes were used for fluorescence *in situ* hybridization (FISH) on metaphase chromosomes: whole chromosome painting (WCP) for chromosome 15 (WCP #151) and partial chromosome painting (PCP) probes for chromosomes 17 (PCP #2 for 17p), 12 (PCP #424) and 13 (PCP #123). All probes were obtained from M. Rocchi, Resources for Human Molecular Cytogenetics Project by Italian Telethon and AIRC.[11] Alu PCR products from PCP #424, WCP #151 and PCP #123 were nick translated with bio-16-dUTP and those from PCP #2 and WCP #151 with dig-11-dUTP.

Hybridization and detection. Hybridization was performed as previously described[12] and according to reagent manufacturers' recommendations with minor modifications. The biotinylated probes were detected with Cy3-avidin (Amersham, Italy). Digoxigenated probes were detected using anti-dig-FITC-conjugated antibody (Boehringer Mannheim). WCP #151, labeled with both bio-16-dUTP and dig-11-dUTP, were painted with a mixture of Cy3 and FITC. Metaphases were counterstained with DAPI.

Image acquisition. Images were acquired using an epifluorescence microscope (Eclipse E800, Nikon, Japan) equipped with a CCD camera (Cohu, USA). DAPI, FITC and TRITC images of metaphase cells were acquired with selective single-bandpass filters. Single channel images were overlaid and processed with a FISH dedicated software (Cytovision, Applied Image, USA).

GTG banding technique

After fixation, air-dried metaphase spreads were banded by the standard trypsin and Giemsa staining technique.

Results

BRG A cell clone

CGH analysis on BRG A cells showed overrepresentation of DNA sequences as the main recurrent anomaly. Figure 1a shows the average ratio profile of ten metaphases. Gains were observed in seven chromosomes: 1q (interstitial region: 1q21-31) with a green to red ratio of about 1.5, 7q (the whole q arm), 12 (pericentromeric region: 12p11.2-q14), 13q (subtelomeric region: 13q22-32), 15q (the whole q arm), 17p (the whole p arm) and 20 (pericentromeric region: 20p12-q13.2). Losses involved chromosomes 3p (interstitial region: 3p13-21) and 5q (interstitial region: 5q14-21). All gains and losses were clearly visible in superimposed FITC, TRITC and DAPI images (not shown).

The comparison of the cytogenetic and CGH data is shown in Table 1. Aberrations of chromosomes 1, 3, 7 and 17 are clearly detected in GTG- and DAPI-banded metaphases (Figure 1b and Table 1), while the alterations in chromosomes 5, 12, 13, 15 and 20 were not detectable by GTG- and DAPI-banding because of the small size of the involved region and/or their complex rearrangements. The regions on chromosomes 12 and 17 appeared to be amplified to the same extent with a ratio of about 2.5, suggesting a possible involvement of chromosome 12 in the constitution of der(17). The findings from CGH analysis oriented the choice of chromosome painting probes for characterizing der(17) further towards probes for chromosomes 12, 13 and 15. The chromosome 17p probe was also used in order to define the position of 17p material (Figure 2a) with respect to the other regions of unknown origin.

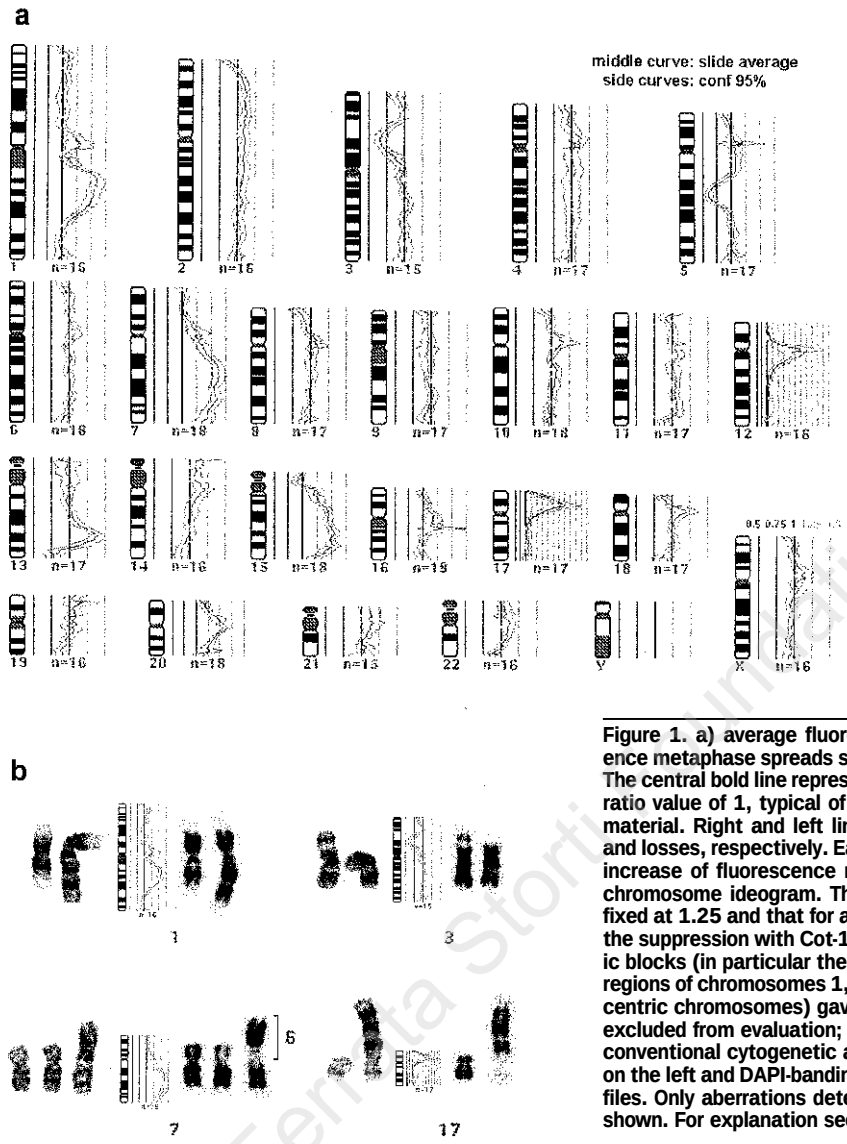

Figure 1. a) average fluorescence ratio profile of ten reference metaphase spreads subjected to CGH with BRG A DNA. The central bold line represents the green-to-red fluorescence ratio value of 1, typical of a balanced state of chromosome material. Right and left lines represent chromosomal gains and losses, respectively. Each line indicates a decrease or an increase of fluorescence ratio of 0.25, as shown on the X chromosome ideogram. The threshold to define a gain was fixed at 1.25 and that for a loss at 0.75, respectively. Due to the suppression with Cot-1 DNA fraction, the heterochromatic blocks (in particular the centromeric and paracentromeric regions of chromosomes 1, 9 and 16 and the p-arm of all acrocentric chromosomes) gave unreliable ratio values and were excluded from evaluation; b) comparison of CGH results and conventional cytogenetic analysis in BRG A cells: G-banding on the left and DAPI-banding on the right of average ratio profiles. Only aberrations detected by the three techniques are shown. For explanation see the text.

FISH analysis on metaphases from BRG A using a probe for chromosome 12 showed, beside the expected signals on the two 12 homologs (not shown), two pairs of distinct small hybridization regions on der(17) (Figure 2a, small arrows). The probe hybridized on both sides of the two small, unidentified centromeric regions previously shown.[5] Taking into account the composition of the probe used, spanning from 12p11 to 12qter, and the Cot-1 suppression hybridization at the centromeres, it is reasonable to assign the unidentified centromeric regions of the der(17) to chromosome 12. CGH showed a gain of chromosome 12 between p11.2 and q14: this finding, too, is in agreement with the results of the painting experiment. The probe for chromosome 15 gave three different signals: one on the telomeric zone of der(17) (Figure 2a, arrowheads), one on an apparently normal chromosome 15 (Figure 2b) and the last on a rearranged acrocentric der(13)t(13;15) (Figure 2b). The probe for chromosome 13 gave no signal on der(17), but four different signals. One of them was on a normal chromosome 13 (Figure 2c), and one on the rearranged acrocentric der(13)t(13;15) (Figure 2c). The remaining two signals were on two small chromosomes (not shown), as expected since the probe was not obtained from a monochromosomic hybrid (*for more information see http://bioserver.uniba.it/fish/rocchi/welcome.html*).

BRG M cell clone

CGH analysis of BRG M cells showed gain of 1q (interstitial region: 1q21-31) with a green to red ratio of about 2.5 and loss of 6q (terminal region: 6q16-qter) (Figure 3a). The ratio profile of chromosome 2 exhibited a shift toward underrepresentation without this being significant. This could indicate that the deletion involved is too small or that it is present in a minor fraction of the cell population, below the sensitivity threshold of the technique. The gain detected

Table 1. Results of CGH and banding analysis in a case of AIDS-related BL compared with literature data.

	BRG–A		BRG–M		BL (data from literature)
	aberrations detected by:		aberrations detected by:		
Chrom #	CGH	banding* (15 met. scored)	CGH	banding* (15 met. scored)	Described aberrations[refs] °
1	enh(1)(q21–31)	1q+	amp(1)(q21–31)	+1q‡	del, t, dup q–arm,[6,7,13]
3	dim(3)(p13–21)	3p–	–	–	t, part. del[7,14]
5	dim(5)(q14–21)	–	–		––
6	–	–	dim(6)(q16–qter)	6q–	dup p–arm, del[6]
7	enh(7)(q11–qter)	+7q	–	–	trisomy[6, 15]
11	–	–	–	–	trisomy, del q–arm[6]
12	amp(12)(p11.2–q14)	–	–	–	trisomy[6,13,15]
13	enh(13)(q22–32)	–	–	–	t, del[13,14,16]
15	enh(15)(q11–qter)	–	–	–	t,[17]
17	amp(17)(p11.2–pter)	der(17)§	–	–	del, i(17q),[6,17]
18	–	–	–	–	trisomy,[6]
19	–	–	–	–	trisomy,[13]
20	enh(20)(p12–q13.2)	–	–	–	trisomy,[5]
21	–	–	–	–	trisomy,[6]
X	–	–	–	–	trisomy,[6]

**Aberrations detected in >80% of scored metaphases. °Classical t(8;14) or variant t(2;8) and t(8;22) were omitted. ‡These aberrations have been characterized in GTG– and DAPI–banding only with the contribution of CGH data. §Described in ref. #8.*

in chromosome 9 fell in the centromeric zone conventionally excluded from the analysis.

GTG-banding karyotype of BRG M appeared to be monosomic for chromosomes 1q and 6 and showed three derivative chromosomes, der(2), der(3) and der(6) (Figure 3b). Banding techniques, supported by CGH results, allowed the definition of der(3) as derived from a translocation t(1;3)(q?;q?) and der(6) as derived from a translocation t(1;6)(q?;p?) (GTG-banding, Figure 3b; DAPI-banding, not shown). Thus the amplification of a region of 1q and the loss of 6q, as shown by CGH data, are also identifiable in DAPI- and GTG-banding (Table 1).

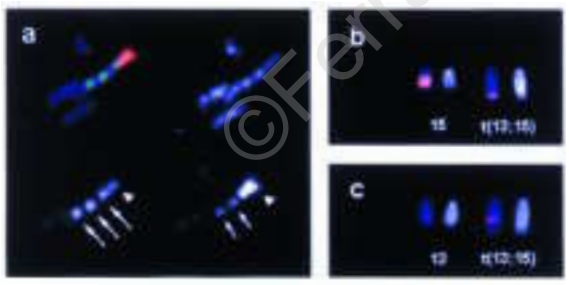

Figure 2. FISH analysis on BRG A cells with painting probes. a) First panel. Four color hybridization on der(17): blue signal, DAPI-staining; green signal, chromosome 17p probe; red-violet signal, chromosome 12 probe; orange signal, chromosome 15 probe; black and white panels show, clockwise, the blue, red-violet and green acquisition channels; small arrows, chromosome 12 probe; arrowheads, chromosome 15 probe; large arrows, chromosome 17p probe. b) On the left, results of hybridization; on the right, DAPI counterstaining. Chromosome 15 probe: signals on a normal chromosome 15 and on a rearranged acrocentric chromosome t(13;15). The third signal of chromosome 15 probe is on the telomeric zone of der(17) (arrowhead, panel a). c) Chromosome 13 probe: signals on a normal chromosome 13 and on the rearranged acrocentric chromosome t(13;15).

Discussion

CGH has been demonstrated to be a powerful tool for the study of solid tumors. Here we give an example of how CGH can add to the power of classical and molecular cytogenetic analysis in the study of hematopoietic tumors. We also show that the information provided by CGH can be used to target FISH analysis to specific chromosomes. Painting experiments on BRG A showed that chromosomes 12 and 15 contributed to the der(17) formation.

Chromosome 12 has been found to be trisomic in B-cell lymphomas,[6, 18] leukemias[19] and in AIDS-related BLs.[6] Chromosome 12 has also been demonstrated to be necessary for HIV gene expression,[20] suggesting a selective advantage for BL cells bearing an extra copy of chromosome 12. However, it has been shown that HIV-infected B cells invariably die and only not infected HIV-related BL cells survive.[21] Thus the relationship between trisomy 12 and HIV infection could be indirect.[6] Our results suggest that the region relevant for lymphomagenesis could be restricted to 12p11.2-q14, and the aberration involving chromosome 12 could be even more frequent than described, not being detectable by conventional chromosome banding.

We have previously shown that the formation of der(17) involved triplication of deleted 17p.[8] CGH analysis confirmed the gain of 17p. Loss of 17p, as well as presence of i(17q), has often been reported in lymphomas.[6] In contrast, gain of 17p is not a recurrent finding in lymphoma. One reason could be that the region involved is too small to be detected by conventional cytogenetic analysis. It is also possible that gain of 17p11.1-12 was present in our case since it was a consequence of amplification of chromosome 12 during the formation of der(17).

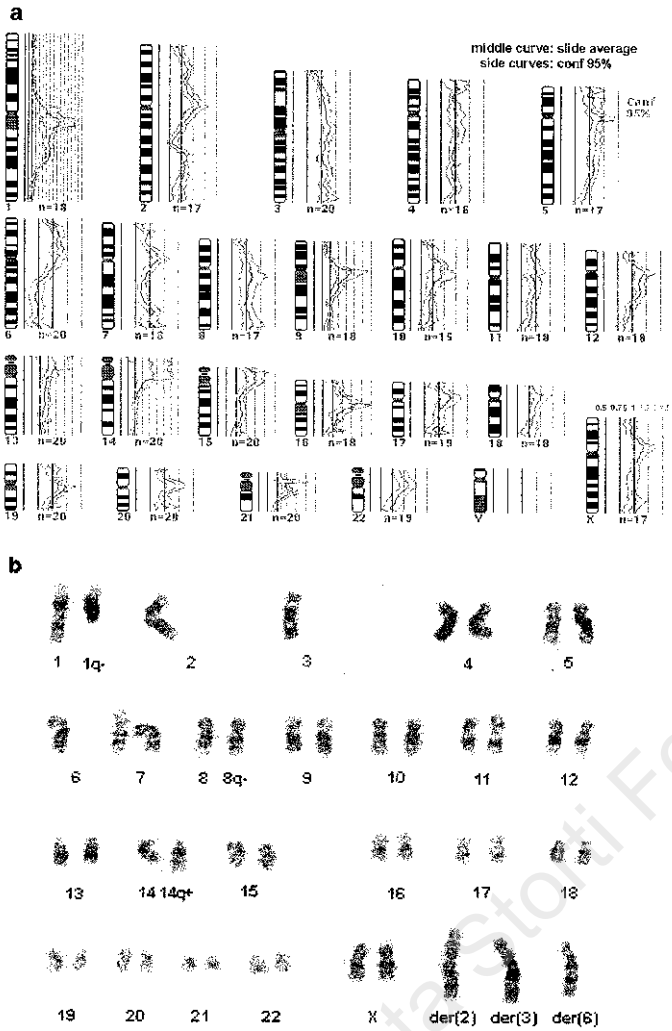

Figure 3. a) average fluorescence ratio profile of ten reference metaphase spreads subjected to CGH with BRG M DNA. For detailed explanations see legend to Figure 1a; b) G-banded karyotype of BRG M.

CGH analysis of BRG A clearly showed the presence of a gain of 1q21-32. Apart from the diagnostic translocation, rearrangements involving 1q21-32 are the most frequent rearrangements in BL,[6,22] particularly in AIDS-related cases,[7] and are associated with a poor prognosis.[6] Furthermore, CGH analysis demonstrated the gain of the whole 7q; GTG- and DAPI-banding confirmed the presence of an extra copy of 7q, translocated to the p-arm of chromosome 6. It has already been reported that 10-30% of BLs show total or partial trisomy of chromosomes 7 and 1[6,15,23] and that chromosome 7 is probably involved in the progression of this tumor.[15]

CGH analysis of BRG M showed only the gain of 1q21-32 and the loss of 6q16-ter; both these aberrations have already been described in lymphomas by using conventional cytogenetics.[6,18,23] Gain of 1q21-32 was the only aberration found in both BRG clones. This is not surprising considering that IgA expressing cells originated *in vivo* from IgM expressing cells by isotype switch.[10] However CGH showed a difference in the degree of 1q amplification in BRG A and BRG M (ratio 1.5 vs 2.5). Moreover, as shown by chromosome banding, the gained region resided on chromosome 1 in BRG A, while it was translocated on different chromosomes in BRG M. This could imply an independent sequence of events leading to 1q21-32 gain, supporting its relevance in lymphomagenesis.

The analysis of archival metaphases obtained from fresh peripheral blood at the time of diagnosis showed that two cell clones were already present in the cell population: one bearing the typical BRG A derivative chromosomes [1q+; der(6)t(6;7); 3p–; der(17)] and the other bearing those specific to BRG M [(1q–; der(6)(6p;1q), der(3)t(3;1q)] (not shown). Thus these aberrations did not arise *in vitro* and may have played a role during the development of BL.

Contributions and Acknowledgments

AZ, SV, LO were equally responsible for CGH, banding techniques, FISH analysis and drafting the paper. GF contributed to the analysis and interpretation of data. AS was responsible for image acquisition and processing. SR dealt with cell culturing and characterization of subclones. AA was responsible for the conception of the study. All the authors contributed to the critical revision and approved the final ver-

sion of the paper. We thank Dr. G. Tonini for hospitality in the Biology of Solid Tumors Unit at CBA, Genoa, where the CGH analysis was performed. We thank Dr. C. Lo Cunsolo and Dr. K. Mazzocco (CBA), Dr. M. Aubele and Dr. J. Bruch (GSF, Oberschleissheim) for their helpful advice.

Funding

This work was partially supported by grants from the Italian Association for Research on Cancer (AIRC) and MURST to AA, and the Istituto Superiore di Sanità (AIDS project), Italy.

Disclosures

Conflict of interest: none.

Redundant publications: our study begins where a previous study finished (Cancer Genet Cytogen 1999; 110:1-6).

Manuscript processing

Manuscript received June 14, 1999; accepted September 13, 1999.

Potential implications for clinical practice

- Here we gave an example of how CGH can add to the power of classical and molecular cytogenetic analisys in the study of hematopoietic tumors: complex karyotypes and small regions of amplification, not detectable by conventional banding, could be easily identified. This technique, applied to a larger number of cases, could allow the definition of a restricted chromosomal region relevant for the pathology, which might be used as marker for diagnosis, prognosis and therapy.

References

1. Kallioniemi A, Kallioniemi OP, Sudar D, et al. Comparative genomic hybridization for molecular cytogenetic analysis of solid tumors. Science 1994; 258:818-21.
2. Kallioniemi OP, Kallioniemi A, Piper J, et al. Optimizing comparative genomic hybridization for analysis of DNA sequence copy number changes in solid tumors. Gene Chromosome Canc 1994; 10:231-43.
3. Bentz M, Huck K, Du Manoir S, et al. Comparative genomic hybridization in chronic B-cell leukemias shows a high incidence of chromosomal gains and losses. Blood 1995; 85:3610-8.
4. Bryndorf T, Kirchhoff M, Rose H, et al. Comparative genomic hybridization in clinical cytogenetics. Am J Hum Genet 1995; 57:1211-20.
5. Xiao H, Volk MS, Tebbi CK, Kakati S. American Burkitt lymphoma stage II with 47,XY, +20, t(8;14)(q24;q32). Cancer Genet Cytogen 1990;48:275-7.
6. Kornblau SM, Goodacre A, Cabanillas F. Chromosomal abnormalities in adult non-endemic Burkitt's lymphoma and leukemia: 22 new reports and a review of 148 cases from the literature. Hematol Oncol 1991; 9:63-78.
7. Polito P, Cilia AM, Gloghini A, et al. High frequency of EBV association with non-random abnormalities of the chromosome region 1q21-25 in AIDS-related Burkitt's lymphoma-derived cell lines. Int J Cancer 1995; 61:370-4.
8. Ottaggio L, Moro F, Fronza G, et al. Derivative chromosome 17 in a case of Burkitt's lymphoma with 8;14 translocation. Cancer Genet Cytogen 1999; 110:1-6.
9. Roncella S, Cutrona G, Favre A, et al. Apoptosis of Burkitt's lymphoma cells induced by specific interaction of surface IgM with a self-antigen: implications for lymphomagenesis in acquired immune deficiency syndrome. Blood 1996; 88:599-608.
10. Jain R, Roncella S, Hashimoto S, et al. A potential role for antigen selection in the clonal evolution of Burkitt's lymphoma. J Immunol 1994; 153:45-52.
11. Antonacci R, Marzella R, Finelli P, et al. A panel of subchromosomal painting libraries representing over 300 regions of the human genome. Cytogenet Cell Genet 1995; 68:25-32.
12. Pinkel D, Straume T, Gray JW. Cytogenetic analysis using quantitative, high-sensitivity, fluorescence hybridisation. Proc Natl Acad Sci USA 1986; 83:2934-8.
13. Bernheim A, Berger R. Cytogenetic studies of Burkitt lymphoma-leukemia in patients with acquired immunodeficiency syndrome. Cancer Genet Cytogen 1988; 32:67-74.
14. Laï JL, Daudignon A, Kerckaert JP, et al. Translocation (3;13)(q27;q14): a nonrandom and probably secondary structural change in non-Hodgkin's lymphomas. Cancer Genet Cytogen 1998; 103:140-3.
15. Knutsen T. Cytogenetic changes in the progression of lymphoma. Leuk Lymphoma 1998; 31:1-19.
16. Barin C, Valtat C, Briault S, et al. Structural rearrangements of chromosome 13 as additional abnormalities in Burkitt's lymphoma and type 3 acute lymphoblastic leukemia. Cancer Genet Cytogen 1992; 60: 206-9.
17. Daly P, Brito-Babapulle V, Lawlor E, Blaney C, Parreira A, Catovsky D. Variant translocation t(8;22) and abnormalities of chromosome 15(q22) and 17(q12-21) in a Burkitt's lymphoma/leukaemia with disseminated intravascular coagulation. Br J Haematol 1986; 64:561-9.
18. Monni O, Joensuu H, Franssila K, Knuutila S. DNA copy number changes in diffuse large B-cell lymphoma. Comparative genomic hybridization study. Blood 1996; 87:5269-78.
19. Liso V, Capalbo S, Lapietra A, Pavone V, Guarini A, Specchia G. Evaluation of trisomy 12 by fluorescence in situ hybridization in peripheral blood, bone marrow and lymph nodes of patients with B-cell chronic lymphocytic leukemia. Haematologica 1999; 84:212-7.
20. Hart CE, Ou CY, Galphin JC, et al. Human chromosome 12 is required for elevated HIV-1 expression in human-hamster hybrid cells. Science 1989; 246:488-91.
21. De Rossi A, Ometto L, Roncella S, et al. HIV-1 induces down-regulation of bcl-2 expression and death by apoptosis of EBV-immortalized B cells: a model for a persistent "self-limiting" HIV-1 infection. Virology 1994; 198:234-44.
22. Heim S, Mitelman F. Malignant lymphomas. In: Alan R. Liss, ed. New York: Cancer Cytogenetics; 1987. p. 210-2 2585.
23. Lai JL, Fenaux P, Zandecki M, Nelken B, Huart JJ, Deminatti M. Cytogenetic studies in 30 patients with Burkitt's lymphoma or L3 acute lymphoblastic leukemia with special reference to additional chromosome abnormalities. Ann Genet-Paris 1989; 32:26-32.