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Recurrent Chromosomal Abnormalities in Lymphomas in Fine Needle Aspirates of Lymph Node

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A B S T R A C T

The detection of specific chromosomal abnormalities is important in the diagnostic workup of aggressive lymphomas, giving its impact on the treatment strategies and prognosis. This has been accomplished by using the fluorescent in situ hybridisation method (FISH) performed on fine needle aspiration (FNA) specimens what is attractive in the diagnosis of lymphoma in the comparation with other methods for collecting samples. The cytogenetic analyses were performed in series of 80 patients with lymphoma (43 women and 37 men, median age 48, range 3–90 years). In our series 89.0% (71) of the specimens yield sufficient numbers of analysable metaphases, comprising 63 non-Hodgkin lymphomas (NHL) and 8 examples of Hodgkin disease (HD). Among 71 successful karyotyped specimens 58 (82.0%) showed clonal karyotypic abnormalities. Numerical changes in 4, structural changes in 20 and both and numerical with structural changes in 30 of 54 NHL cases. Trisomies 3, 7, 8, 12, 18, X and monosomies 1 were most common numerical abnormalities. The NHL cases were typically characterised by structural rather than numerical aberrations with chromosome arms 1p/q, 3p/q, 6q, 11q, 17p and 14q most frequently involved. The expected translocation (14;18)(q32;q21) in 8 and t(8;14)(q24;q34) in 6 cases, both translocations at the same time in three cases, complex rearrangement with chromosome 8, 14, and 18, namely t(8;14;18)(q24;q32;q21) in one case, t(11;14)(q13;q32) in three and one case with translocation 14q32 with chromosome 3q27, 6q and 14q32 were found. In 28 of 54 (52%) NHL cases t(14;v) was present. Four abnormal clones detected in Hodgkin disease were typically consisted of a small percentage of metaphases. The use of FISH method enable the detection of loss or gain of genetic material and reveal rearrangements unsuspected by conventional cytogenetics in 34 (48.0%) cases.

Key words: *fine needle aspirate, lymphoma, cytogenetics*

Introduction

Lymphomas are the group of clinically important neoplasms with a complex biology, which makes their classification and treatment difficult. They cause significant morbidity and mortality[1]. The World Health Organization (WHO) classification based on morphology, immunophenotype, genotype, the normal cell counterpart and clinical behaviour recognizes three main groups (with multiple subsets): non-Hodgkin lymphoma (NHL) of B cells and T/natural killer (NK) cells and Hodgkin disease (HD)[2]. The B- and T-cell neoplasms are stratified into precursor (lymphoblastic lymphomas), or lymphoblastic neoplasms, and mature (peripheral) B- and T-cell neoplasms[3,4]. The mature B- and T-cell neoplasms are grouped according to their major clinical features into the: predominantly disseminated (leukemic), primarily extranodal, and predominantly nodal NHL which result from transformation of B and T/NK cells[5]. From 75 to 85% of lymphomas are B-cell type[1,6]. Their hallmark is chromosomal translocation resulting from aberrant rearrangements of immunoglobulin (IG) and T-cell receptor

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(TCR) genes. That leads to an inappropriate expression of genes at reciprocal breakpoints that regulate a variety of cellular functions (gene transcription, cell cycle, apoptosis) and tumor progression. Cytogenetics and molecular genetic analysis of some of the recurring translocations gave new insights into the pathogenesis of lymphoma, which may play a role in their classifications, to provide lymphoma progression and clinical behaviour[1,7,8]. Certain lymphoid malignancies harbor a characteristic chromosomal translocation, a finding that may have significant implications for an individual's prognosis or response to therapy[9]. B-NHL is characterized by recurrent translocations involving the immunoglobulin heavy chain (IGH) gene in approximately 50% of cases[6]. The most consistent chromosomal abnormalities in B-NHL, reciprocal translocations t(11;14)(q13;q32) and t(14;18)(q32;q21) in mantle cell lymphoma (MCL) and follicular lymphoma (FL), respectively, are now viewed as highly specific markers, present in greater than 90% of the cases[10]. For example, Burkitt lymphoma (BL), a highly aggressive lymphoma, is associated with a translocation involving c-myc gene in more than 90% of cases. In over 95% of mantle cell lymphomas (MCL), a t(11;14)(q13;q32) is found involving the cyclin D1 and IGH genes. In addition, a translocation of the BCL2 gene to the IgH gene locus resulting in a t(14;18) is a hallmark of follicular lymphoma and is only seen in 20–30% of the diffuse large B cell lymphoma (DLBCL) IGH translocations[11]. Chromosomally aberrant clones are rarely detected in HD (20–50% of cases), with relatively simple chromosome aberrations[7]. In contrast, cytogenetically identical translocations may be found in several types of disease. Chromosomal translocations involving the immunoglobulin heavy-chain locus (IGH) are a feature of mature B-cell malignancies and result in deregulated expression of the translocated genes due to proximity of transcriptional enhancers with IGH[12]. The IGH locus is located on chromosome 14, at band q32.3 and spans 1,250-kilobase (kb) region[5]. IG translocations are usually detected by cytogenetics often supplemented with fluorescence in situ hybridisation (FISH) methods using probes that span the IG loci[13]. Previous reports have shown that FISH can be performed on FNA specimens[10,14]. Karyotypes obtained by conventional cytogenetics are usually complex, with numerous structural abnormalities. Therefore, banding techniques may fail to identify cryptic translocations or insertions and those partners located in the terminal bands of other chromosome. FISH is well-suited to determine the incidence of those specific translocations in complex karyotype[6].

However, there is only limited publishing data regarding the diagnostic utility of FISH-based detection of these translocations. Cytogenetic analysis, based on banding techniques, present an overview of all cytogenetic aberrations. However, lack of success in culturing tumour cells, low mitotic indices and the lack of fresh material often complicates the use of this technology for routine diagnosis[11]. Interphase FISH has an enhanced ability over conventional cytogenetics to detect specific chromosomal abnormalities[14].

We present the results of 80 cases of lymphoma diagnosed from the fine needle aspiration (FNA) of lymph node combining with cytomorphology and cytogenetic analysis for a definitive diagnosis.

Patients and Methods

Aspirates from 80 patients undergoing diagnostic FNA for the evaluation of clinically suspected lymphoma were collected. Of these, 43 were women and 37 were men, ages 3 to 90, with a median age of 48 years.

Cytomorphological methods

The specimens were obtained using a 23-gauge disposable needle and 10 ml disposable syringe. At each FNA 5–10 passes in the lymph node were completed. The aspirate from one or more FNA (depending on sufficient material) was used for cytomorphological diagnoses. The material from FNA was immediately smeared onto several slides, and air dried smears stained by the May-Grünwald-Giemsa method. Cytochemical analysis (myeloperoxidase, non-specific esterase, DNA staining) and imunocytochemical analysis were also performed[15–17].

Conventional cytogenetics

The material from one or more FNA was collected only as a needle and syringe washings in a 10-ml tube with 2ml RPMI 1640 tissue culture medium and two drops of heparin solution. This was accomplished by withdrawing the contents of the tube into the syringe barrel and gently expelling into the tube and repeating the procedure at least three times to ensure maximum recovery of the material until the medium was visibly cloudy. The lymph node cells were incubated at 37 °C and cultured for 24 and 48 hours with stimulation with FdU. After exposing the culture to Colcemid (0.05 μg/mL) for 20 minutes the suspensions were resusspended in 0.074 mol/L KCl for 10 minutes and fixed with a 3:1 mixture of methanol and glacial acetic acid. After repeating the fixation process three times, the slide preparation was made, and G-banded with Wright's stain[17,18]. All metaphase plates were analysed, recorded, and saved. Abnormal clone was defined as two or more cells with the same structural abnormality or the same extra chromosome, or the presence of three or more cells with the same missing chromosome. Case with only one mitotic cell with an abnormal karyotype, was considered a malignant clone if there was structural abnormality known to be associated with lymphoma. When it was possible, it was confirmed by fluorescent in situ hybridization (FISH)[19].

Molecular cytogenetics

FISH was performed using specific DNA probes (Kreatech, Qbiogene, Vysis) for aneuploidy centromere probes of chromosome 1–22 and XY and for detection of specific translocations t(8;14), t(11;14) and t(14;18), re-

spectively, according to the manufacturer's instructions on metaphase and interphase cells. The chromosome paints and telomeric probes were chosen according to the chromosomal rearrangment observed in conventional cytogenetics. FISH signals were analyzed using a AX70 fluorescent microscope (Olympus) equipped with appropriate filter sets for observing Spectrum -Green, -Orange, or -Aqua and DAPI signals. Abnormal results were defined when the percent of cells with any given chromosome abnormality exceeded the normal cut-off value of 200 analysed cells. FISH probes were validated using negative or positive controls. Appropriate positive controls were selected from archival material of patients with prior cytogenetic evidence of the reciprocal translocations. FISH permitted to detect loss or gain of genetic material and also reveal rearrangements unsuspected by conventional cytogenetics[6]. The karyotypes and FISH results were described according of the International system for Human Cytogenetic Nomenclature 2005 (ISCN 2005)[20].

Results

The 80 cases of lymphoma were cytomorphologically and cytogenetically identified in the FNA material. In 89% (71/80) specimens material was available for classification and showed a predominant B-cell lymphoma (B-NHL) in 75.0% (53), T-cell in 14.0% (10) and Hodgkin lymphoma (HD) in 11.0% (8) samples (Table 1). Of the 53 cytology specimens of B-NHL's, 6 were precursor B-cell lymphoma and 47 mature B-cell lymphoma, including 22 cases of Burkitt lymphoma (BL), 11 cases of diffuse large B-cell lymphoma (DLBCL), 8 cases of follicular lymphoma (FL), 3 cases of mantle cell lymphoma (MCL), 2 cases of extranodal marginal zone B-cell lymphoma of MALT type (MALT) and one case of lymphoplasmacytic lymphoma (LPL) (Table 2). Among the 71 successfully karyotyped specimens, 82.0% (58) showed clonal karyotypic abnormalities.

Abnormal karyotype of 53 B-cell NHL's specimens showed numerical changes in 7.5% (n=4) cases. Trisomies 3, 7, 8, 12, X and monosomy 1 were most common numerical changes. Structural changes were found in 37% (20/53) cases, while both numerical and structural changes were discovered in 56.6% (30/53) cases of B-NHL. Structural aberrations involving chromosome arms 1p/q, 3p/q, 6q, 11q, 17p and 14q were most frequent. An IGH rearrangement was found in 53% (28/53) cases of B-NHL, in 20% (2/10) cases of T-NHL and 12.5% (1/8) cases of HD. Translocation t(8;14)(q24;q34) was found in 28.6% (4/14) cases of BL and 1% (1/10) case of NHL-T and 12.5% (1/8) cases of HD. Translocation (14;18)(q32;q21) was found in 62.5% (5/8) cases of FL, 14.3% (2/14) cases of BL, and 25% (1/4) cases of B-cell NOS. Both translocations, t(8;14)(q24;q34) and (14;18)(q32;q21) were detected in three cases at the same time: 14.3%(2/14) in BL and 9% (1/11) in DLBCL. Complex rearrangement involving chromosome 8, 14, and 18, namely t(8;14;18)

TABLE 1
NUMBERS OF DIFFERENT TYPES OF MALIGNANT LYMPHOMA WITH CLONAL CHROMOSOMAL ABNORMALITIES

		Cytogenetic results	
		Cytogenetically abnormal No (%)	Cytogenetically normal No
B-cell NHL	53 (75%)	37 (69.8%)	7
T-cell / NKLL	10 (14%)	8 (80%)	2
Hodgkin lymphoma	8 (11%)	4 (50%)	4
Total	71	58 (82%)	13

TABLE 2
NUMBERS OF DIFFERENT SUBTYPES OF B-CELL NON-HODGKIN LYMPHOMA WITH CLONAL CHROMOSOMAL ABNORMALITIES

Cytomorphology	No (%) of samples	Cytogenetic results	
		Cytogenetically abnormal No (%)	Cytogenetically normal No
Precursor B-cell neoplasms	6 (11%)	6	–
LPL	1 (2%)	1	–
MALT	2 (4%)	2	–
FL	8 (15%)	6	2
MCL	3 (5.5%)	3	–
DLBCL	11 (21%)	9	2
BL	22 (41.5%)	19	3
Total	53	37 (69.8%)	7

LPL – lymphoplasmacytic lymphoma, MALT – mucosa-associated lymphoid tissue lymphoma (extranodal marginal zone B-cell lymphoma), FL – follicular lymphoma, MCL – mantle cell lymphoma, DLBCL – diffuse large B-cell lymphoma, BL – Burkitt lymphoma

TABLE 3
CYTOMORPHOLOGY AND KARYOTYPE IN SUBTYPES OF MALIGNANT LYMPHOMA

B-cell neoplasms			
Precursor B-cell neoplasms			
No	Age/sex	Cytomorphology	Cytogenetics
1.	68/M	B-cell NOS	46,XY,t(14;18)(q32;q21)[2]/46,XX[10]
2.	67/M	B-cell NOS	85–88,XY...inc[3]/46,XY[11]
3.	57/F	B-cell NOS	90,XXX,der(9)t(9p;17q) x 2,-17 x 2[8]/46,XX[3]
4.	72/F	B-cell	47~49,XX,+3,del(3q),t(9;14)(q21;q11),?add(20p),+mar[cp10]/46,XX[6]
5.	24/F	B-cell NOS	46,XX,i(17)(q10) [10]
6.	31/M	B-cell	47,XY,+8[15]
Mature B-cll neoplasms			
1.	69/F	LPL	47,XX,del(5q)+12[2]/46,XX[10]
2.	49/M	MALT	49,Y,del(Xp), der(14q32),der(14q32),+18,+mar[5]/46,XY[2]
3.	57/M	MALT	47,XY,+?X,der(1)t(1q;10p)del(3)(p21),add(4q),del(7)(q36),-10,?inv(11q), add(13p), der(19)t(1q;19p),-22,+2mar[cp12]
4.	41/F	FL I	46,XX,add(1)(p36)dup(1)(q21),t(14;18)(q32;q21),del(17)(q25)[8]
5.	50/F	FL I	nuc ish t(14;18)(q32;q21) 10%
6.	66/M	FL I	47,XY,t(14;18)(q32;q21),+18[15]
7.	55/M	FL I	46,XX,del(4)(q21q2?7),t(14;18)(q32;q21)[8]/46,XX[5]
8.	-/F	FL II	45,X,-X,-3,+12[12]
9.	-/M	FL II	47,XY,+2,ins(12;?)(q15;?),t (14;18)(q24;q21)[10]/46,XY[5]
10.	72/F	FL II	46,XX[15]
11.	-/F	FL II	46,XX[10]
12.	55/F	MCL	46,XX,t(11;14)(q13;q32)[2]
13.	60/F	MCL	47,XX,add(12q),i(17)(q10),+mar[9]/46,XX[5]
14.	-/F	MCL	45,XX,+7,-9,t(11;14)(q13;q32),-13,der(15)t(?;15)[4]/46,X X[30]
15.	33/F	DLBCL	48,XX,i(7q),add(14)(q32),+i(?),+mar[8]
16.	-/F	DLBCL	46,XX,der(7),t(7p;?)[10]
17.	-/M	DLBCL	46,XY[10]
18.	-/F	DLBCL	46,XX,del(5q),der(6)t(1;6)(q23;q27),der(10)t(10;11)(p13-14;q13) [7]/92,XX[3]/46,XX[3]
19.	-/M	DLBCL	50,XY,-1,+3,+12,add(14)(q32),add(15)(p11)i(18q),+mar x 2 [3]/46,XY[20]
20.	56/M	DLBCL	46,XY,del(6q) [2]/46,XY[30]
21.	-/M	DLBCL	46,XY,der(11)add(11)(p15)del(11q21)[3]
22.	56/M	DLBCL	46,XY[17]
23.	64/F	DLBCL	46,XX,t(1p;?),dup(3)(pter?q27qter),t(8;14)(q24;q32),t(14;18)(q32;q21), t(11q;12q)[15]/46,XX[2]
24.	65/M	DLBCL	70,XY,der(Xq),der(1p),-2,add(2q),-3,-6,-6,dic(6;?),+7,-8,-9,add(9p),add(10q), add(10q), +12,add(16p),add(17q),+19,-22,-22,add(22p),+5mar[1]
25.	55/M	DLBCL	46,XY,der(4)t(1;4)(q23;q35)[10]/46,XY[3]ncl:del(1p),i(q)
26.	-/M	BL like	48,XY,+3,t(11;14)(q13;q32),+12[15]/46,XY[5]
27.	-/M	BL like	86,XXYY…+3mar,1-2mar, dmin [6]
28.	44/F	BL like	46,XX,dup(1)(q21q31),i(7q),t(8;14)(q24;q32)[10]/46X,i(Xq),idem[8]
29.	3/F	BL like	46,XX [5]
30.	13/F	BL like	46,XX,dup(1q),t(2p;8q),add(7q),del(9q),add(21p)[cp20]
31.	70/F	BL like	46,XX,t(2;?;3),dup(5q) [15]/46,idem,del(9p) [2]/47,idem,+mar [3]/46,XX[4]
32.	20/F	BL like	49,XX,+7,t(8;14)(q21;32),+ 10,t(14;18)(q32;21),+der(18)t(14;18)(q32;q21) [14]
33.	57/M	BL like	48~52,XY,+X,del(1p),+5,del(6q),+7,+12,t(14;18)(q32;q21),+22 [cp9]/46,XY[3]
34.	75/F	BL	46,XX,t(2,12)(q21;q22),der(3q),t(6;14)(p21;q32),del(6q),der(8p),add(9p), del(11p), tas(1;12)(q21;q24),der(14) [cp12]
35.	59/M	BL	49,XY,+del(1)(p31),t(2;22)(p12;q11),+7,+8,t(8,14)(q24;q32),t(13;15)(p12;q14), t(14;18) (q32;q21)[12]/50,idem,+7[8]

B-cell neoplasms			
Precursor B-cell neoplasms			
No	Age/sex	Cytomorphology	Cytogenetics
36.	-/M	BL	46, XY[14]
37.	58/F	BL	48,XX,+X,add(11p),t(14;18)(q32;q21),+mar,0-2dmin[11]
38.	90/M	BL	46,XY,t(8;14))(q24;32),-21,+der(21)t(1; 21)(q14;p12)[14]
39.	-/M	BL	49,XY,+X,add(3q),+7,add(11q),+12[15]/ 46,XY[1]
40.	24/F	BL	46,XX,der(1),t(1pter;?),-8,+ 11[10]
41.	45/F	BL	46, XX,t(8;14)(q24;32) [10]/46,XY[7]
42.	8/M	BL	46,XY[9]
43.	-/F	BL	46,XX,t(3;14)(q27;q32)[2]/46,XX[3]
44.	38/F	BL	46,XX,t(8;14)[6]/46,XX[2]
45.	72/F	BL	48,X,-X,+3,+ i(6)(p10) x 2,del(6q),del(9p22),del(15q)?7?BM 47,XX,+12?1?
46.	51/F	BL	46,XX,add(10)(q26)[4]
47	51	BL	47,XX,del(2p),del(3q),t(8;14;18)(q21;q32;q21),-13+2mar?10?/46,XX ?2?
T-cell and NK-cell neoplasms			
1.	-/F	NHL-T NOS	51,XX,-1,add(4q),add(4q),+7,+8,+10,+2mar[8]/46,XX[5]
2.	54/M	NHL-T NOS	46,XY[10]
3.	32/F	NHL-T NOS	47,XX,+?17[15]
4.	29/M	T-LBL	46, XY,del(6q),-14,+mar [15]/46,XY[5]
5.	-/M	NHL(T-ANAPLASTIC)	46,XY,del(3p),der(3)add(3p)add(3q),del(6q),der(12)t(1;12)(q21;q24),-13,t(14;14), der(16); dup(16p),+mar[cp12]
6.	46/F	NHL –T NOS	46,XX[16]
7.	19/M	T-ALL/LBL	47,XY,+21[25]
8.	35/M	T-ALL/LBL	I 46,XY[17] II 46,XY,t(1;4)(p32;p12),t(8;14)(q24;q32)[20]
9.	43/M	NHL(T-ALCL)	50~52,Y,der(Xp),t(1,16)(p36;q24),t(2;5),del(4q21),t(4;12)(q31;q12),+der(5)t(2;5), del(6) (q23), +i(8)(q10), der(10)t(10;11)(q26;q13),der(17)t(2;17)(p13;p14),inv(?20),+21,+22, +1-2mar[cp12]/46,XY[2] PB 47,XY,+i(8)(q10) [4]/46,XY[13]
10.	75/F	AITL	54,XX,+X,+X,+2,+5,+7,+12,del(12p),+17,+21[2]/46,XX[20]
Hodgkin lymphoma			
1.	-/F	HL	51,X,del(Xq),dup(1)(q25q32), +3,+3, t(4;11)(q21;q23),+der(11)t(4;11),t(8;14)(q24;q32), +mar1,+mar2[10]/46,XX[8]
2.	40/M	HL	46,XY[6]
3.	72/F	HL	46,XX[10]
4.	72/F	HL	46,X,?-X,del(3)(q21),+12[2]/47,XX,+mar[1]/46,XX[15] BM 47,XX,-1,+5,+mar[2]/46,XX[20]
5.	-/M	HL	1995-46,XY[4] 2006-46,XY[10]
6.	43/M	HL-LP	45,XY,dic(13;15)[15]
7.	57/M	HL/ALCL	46,XY[20]
8.	72/F	HL	46,X,-?X,del(3)(q21),+12[2]/46,XX[5] BM 47,XX,+r(12)[1]

NHL – non-Hodgkin lymphoma, LPL – lymphoplasmacytic lymphoma, MALT – mucosa-associated lymphoid tissue lymphoma (extranodal marginal zone B-cell lymphoma), FL – follicular lymphoma, MCL – mantle cell lymphoma, DLBCL – diffuse large B-cell lymphoma, BL – Burkitt lymphoma, HL –Hodgkin lymphoma, LP – lymphocytic predominance, ALCL – anaplastic large cell lymphoma, AITL – angioimmunoblastic T-lymphoma, NOS – not otherwise specified, T-ALL/LBL – T-lymphoblastic leukemia/lymphoblastic lymphoma

(q24;q32:q21) was found in one case BL. Translocation (11;14)(q13;q32) was found in two cases MCL and one case BL. The translocation 14q32 with chromosome 3, 6 or 14 was found in 5 cases (MALT, DLBCL x 2 and BL x 2). Abnormal clones in 50.0% (n=4) cases of Hodgkin disease consisted of a small percentage of metaphases. The

various chromosomal translocations in lymphoma are reviewed below according to cytologic subtype of disease (Table 3).

FISH detected breakpoints, loss or gain of genetic material and revealed rearrangements unsuspected by conventional cytogenetics like case with t(8;14;18) and in cases with low mitotic index, namely in 34 (48%) of them.

Discussion and Conclusion

Recurrent chromosomal translocations are important diagnostic and prognostic markers contributing to the management of patients with a variety of hematologic malignancies[13]. While the acute leukemias are associated with only a single chromosomal translocation, malignancies of mature B cells often exhibit enormous cytogenetic complexity, with multiple and complex translocations, deletions, and amplifications in the 1 clone[21,22]. Functional consequence of gene dysregulation in lymphoid lineage development and lymphomagenesis may be caused by juxtaposition of homeobox (Hox) genes with imunoglobulin heavy chain gene IGH[5,23]. Specific chromosomal translocations are closely associated with distinctive subtypes of non-Hodgkin lymphoma. Activation of BCL2 (18q21) and MYC (8q24) oncogenes usually occurs in B-cell non-Hodgkin lymphoma by translocation to IGH (14q32) allele[4,22,23]. Cytogenetic findings in lymphomas might be helpful in understanding pathogenesis of those disorders[24–26].

The aim of the current study was to determine whether conventional and molecular cytogenetics studies improved the ability to diagnose and/or subclassify B-cell NHL, T-cell NHL and HD on FNA. By incorporating the results of conventional cytogenetic and FISH, a definitive classification was reached in 53 cases of B-cell NHL, 10 T-cell NHL and 8 cases of HD according to their cytomorphologic diagnosis and abnormal karyotypes. Types of chromosomal abnormalities are similar to those reported in the literature where material was either lymph-node specimens and/or bone marrow biopsy sections. In 82% (58/71) patients with diagnosed lymphoma, the karyotypes showed one or more abnormalities. In 54% (28/54) NHL cases t(14;v) was present. Major recurrent clonally chromosomal changes were structural rather than numerical aberrations, involving chromosome 14q32 and chromosome arms 1p/q, 3p/q, 6q, 11q, 17p. Trisomies 3, 7, 8, 12, X and monosomy 1 were also present. Characteristic translocations were t(8;14) found in six cases and t(14;18) found in eight cases, both translocations at the same time were found in three cases, t(11;14) in three and one case with translocation 14q32 with chromosome 3, 6 and 14. Complex rearrangement including chromosome 8, 14, and 18 in one case, with concurrent activation of BCL2 and MYC by translocation of both oncogenes to both IGH alleles. Abnormal clones containing discrete chromosome aberrations, with a small percentage of metaphases were found in four (50.0%) cases of HD. This yield of abnormal metaphases in NHL and HD is comparable to previous studies[5,14,15].

This study illustrate the utility of cytogenetic analysis as an additional ancillary technique to improve the diagnosis and classification of lymphoma. By using FNA, we have shown that 89% of specimens presented recurrent chromosomal abnormalities. With proper handling and management of specimens, FNA can routinely provide samples adequate for molecular genetic studies, in addition to cytomorphology, making it possible to consistently render accurate and primary diagnoses in a subset of NHL's. Moreover, some cytogenetic alterations define clinically relevant subgroups and are, therefore, crucial for therapy decisions.

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UČESTALE KROMOSOMSKE ABNORMALNOSTI U LIMFOMIMA, UZORCI DOBIVENI CITOLOŠKOM PUNKCIJOM LIMFNOG ČVORA TANKOM IGLOM

S A Ž E T A K

Otkrivanje kromosomskih abnormalnosti u limfomima važno je u postavljanju dijagnoze agresivnih limfoma te doprinosi strategiji liječenja i prognozi. U posljednjih nekoliko godina je fluorescentna in situ hibridizacijska metoda (FISH) neizostavna dopuna evaluaciji malignih hematoloških bolesti, a na uzorcima dobivenim citološkom punkcijom limfnog čvora (FNA) u usporedbi s drugim metodama i vrlo izazovna metoda u dijagnostici limfoma. Ovaj rad obrađuje citogenetsko istraživanje 80 pacijenata s limfomom (od kojih su 43 žene a 37 muškarci, prosječne dobi 48, u rasponu od 3 do 90 godina starosti). U našoj skupini u 89% (n=71) uzoraka je bio dovoljan broj metafaza podobnih za analizu, a činili su ga 63 primjera Non-Hodgkinovog sindroma (NHL) i 8 primjera Hodgkinove bolesti (HD). Od 71 uspješno kariotipiziranih uzoraka 82,0% (58) je slučajeva pokazalo klonalne kariotipske abnormalnosti. Numeričke promjene u 4 od 53 slučaja NHL-a i numeričke te strukturne promjene u 51 od 53 slučaja NHL-a. Od numeričkih promjena su trisomije 3, 7, 8, 12, 18, X i monosomija 1 bile najučestalije. Slučajeve NHL-a uglavnom obilježavaju učestalije strukturne nego numeričke aberacije s najčešćim kromosomom 1p/q, 3p/q, 6q, 11q, 17p i 14q. Očekivano, t(14;18)(q32;q21) u 8 i t(8;14)(q24;q34) u 6 slučajeva a obje su se translokacije pojavile u isto vrijeme u tri slučaja dok je kompleksni rearanžman s kromosomima 8, 14 i 18, poglavito t(8;14; 18)(q24;q32;q21) u jednom slučaju, t(11;14)(q13;q32) u tri slučaja i jedan slučaj s translokacijom 14q32 s kromosomima 3, 6 i 14. U 24 od 64 (37,5%) slučajeva NHL-a bila je prisutna t(14;v). Četiri abnormalna klona otkrivena su u HD i obično su sastojali od malog postotka metafaza. Primjena tehnike FISH omogućila je otkrivanje gubitka ili viška genetskog materijala i preuredbe gena koje se nisu očekivali klasičnom citogenetikom u 34 (48%) slučaja.