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Investigation of TEL/AML1 and BCR/ABL genes fusion in Acute lymphoblastic leukemia (ALL) Patients and Follow-up Study in 25 Bone Marrow Transplanted (BMT) Patients Using Interphase Fluorescence In Situ Hybridization (FISH).

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Background:

Introduction: BCR/ABL fusions in hematopoeitic cells are known to induce resistance to apoptosis and cell changes in response to cell-cell and cell-matrix interactions, on the other hand, patients with TEL/AML1 gene fusions respond differently to treatment, depending on therapeutic protocols.
Aims: We conducted a prospective cohort study to investigate how these translocations affect a person's quality of life, and to evaluate their responses to bone marrow transplantation therapy.
Methods and Materials: TEL, AML1, ABL and BCR probes were applied to cells during interphase, using cytogenetic techniques and FISH analysis to obtain the karyotype of 100 patients, which included genes involved in fusion, signal distributions, age, sex, positive familial background, and responses to therapies. After BMT was performed in 25 patients, all of the above data was collected once again and the results were compared.
Results: In our study, 46% of child patients demonstrated an abnormal FISH pattern (23% with fused ABL/AML1, 3% with deletion, 7% with a gain in TEL gene, and 3% and 10% with deletion and a gain in AML1 genes, respectively. In adults, 27% had an abnormal FISH pattern, while 3% had fused TEL/AML1 genes and other abnormalities, as was evident in chil-

dren. A gain in gene copy occured twice as often as a loss in gene copy, except for child ALL patients with t(12;21), where in 58% of cases, lost TEL gene children with t(12;21) had longer survival periods, while adults with t(9;22) had shorter ones. Post BMT revealed that 65% of BM cells karyotyped normal, compared to 24% pre-BMT. WBC count increased positively with the onset of ALL, although an increase in WBC count decreased survival time. A relationship between positive familial background and ALL was also seen.
Conclusion: FISH is the better method for diagnosing genetic disorders in ALL patients compared to other methods.

Keywords: ALL, BMT ,FISH

Introduction:

Acute lymphoblastic leukemia (ALL) is a progressive, malignant colonal disease of the bone marrow (BM) in which early lymphoid precursors proliferate and replace the normal haematopoietic cells of the BM. ALL may be distinguished from other malignant lymphoid disorders by the immunophenotype of the cells, which is similar to B or T precursor cells. Immunochemistry, cytochemistry, and cytogenetic markers could also aid in categorizing the malignant lymphoid clone.[1] In ALL, lymphoid precursor cells are arrested in the early stages of development by the abnormal expression of genes, caused mostly by chromosomal translocations.[2] In acute lymphoblastic leukemia, 81% of cases appear to have a clonal chromosome abnormality that an be detected by cytogenetic or molecular cytogenetic analysis.[2,3] The most common genetic aberration in child cases of ALL are found to be t(12; 21). The translocation between the short arm of chromosome 12 and the long arm of chromosome 21 was discovered in 1994 by FISH [4], and is a structural chromosomal malformation that has the possibility of identification with routine cytogenetic procedures of less than 5%. It is also reported that TEL/AML1 fusion occurs in approximately 25% of the B-precursor cells of ALL patients. This aberration usually could not be detected by conventional cytogenetic techniques.[3,5,6,7,8] Researchers believe that this translocation may occur during fetal development in some patients.[9] But this fusion is not found in cases of T-cell lineage, acute myelogenous leukemia (AML), or non-Hodgkin lymphoma.[10] The fusion has also been detected in adult ALL patients, though with remarkably low frequency (0-4%) compared to those found in child cases of ALL.[11,12,13] TEL/AML1 fusion is associated with a favorable prognosis impact, exceeding age and leukocyte count, which are included in the present standards of risk assessment.[14,15,16] In child cases of ALL, TEL/AML1 expression identifies a large group of low-risk patients whose relapse rates were revealed to be less than 10% by modern treatment protocols.[17,18,19] The prognosis in patients with TEL/AML1 fusion seems to vary depending on the therapeutic protocol used, though recent studies have reported relapse in this group of patients.[20,21,22] Cytogenetic analysis in ALL patients is often hampered by poor chromosome morphology and few malignant metaphases, and sometimes only normal metaphases are found after cell culture.[23,24]

ALL is diagnosed by blood and bone marrow cell analysis by determining the type

of proliferating lymphocyte. Its treatment includes intensive combination chemotherapy, therapy for secondary infections, hyperurecimia, irradiations, and intrathecal methotrexate. Bone marrow therapy is one of the most effective therapies for ALL recovery time, though this depends on age, phenotype of disease, genetic rejection, and type of prophylaxy. In our study, we performed a follow up after bone marrow transplantation to determine how effective this therapy is for treatment. Up to 65% of leukaemic patients revealed these translocations.[25,26,27] Another abnormality observed was the Philadelphia (Ph) chromosome which is the most frequent cytogenetic abnormality known in human leukemia, and is found in more than 95% of patients with chronic myeloid leukemia (CML), 20% to 40% of ALL adult patients, 2% to 5% of ALL child patients, and in rare cases of acute myelogenous leukemia.[27,28,29,30,31,32] The t(9; 22) translocation leads to a head-to-tail fusion of the ABL proto-oncogene from chromosome 9 with the 5' half of the breakpoint cluster region (BCR) sequences to chromosome 22.[33,34] Transcription of BCR-ABL results either in an 8.5-kilobase (kb) messenger RNA (mRNA) that codes for a 210-kd protein or in a 7.5-kb mRNA encoding a 190-kd protein.[35,36] For the p210 protein, exon b2 or exon b3 of the BCR gene (M-bcr region) is coupled to ABL exon 2 (b2a2 or b3a2 junction), whereas the p190-kd protein results from a break within the first intron of BCR (M-bcr region), splicing the first exon of the BCR gene to the second exon of the ABL gene (e1a2).[27] Other fusion products are observed at much lower frequencies. The bcr-abl proteins exhibited an increased in tyrosine kinase activity compared to the normal 145-kd ABL gene product. Moreover, p190 shows a higher transforming potential than p210 in transfection assays and transgenic mouse models.[36,37] In brief, BCR-ABL expression in hematopoietic cells is thought to induce resistance to apoptosis, growth factor independence, as well as alterations in cell-cell and cell-matrix interactions.[38,39,40,41]

Subjects and Methods:

Bone marrow (BM) samples were obtained from 50 children and 50 adults with acute lymphoblastic leukemia (ALL) diagnosed at the "Hematology-Oncology and bone marrow transplantation research center", and "Medical Genetics laboratory" (P. Mehdipour), respectively, from June 1997 to December 2004 in Tehran city. ALL patients were morphologically classified according to the French-American-British (FAB) classification. In order to measure survival period, only patients who had been diagnosed with ALL in the past five years were allowed to participate.

Bone marrow samples were processed directly in short term culture according to the standard protocol.[42,43] Chromosomes were stained with the trypsin-Giemsa (GTG) banding technique. Slides of samples were pretreated with 2X standard saline citrate (SSC; 300 mmol/L sodium chloride and 30 mmol/L sodium citrate) for 2 minutes at 37°C and then dehydrated with cold 70%, 85%, and 100% ethanol for 2 minutes each. After drying, 10 microlitres of probe mixture

(probe + hybridization buffer) was added to each hybridization area, covered with 22x22 mm cover slips and sealed with rubber cement. The DNA and probe solutions were simultaneously denatured for 5 minutes at 75C. Slides were hybridized for 24 to 48 h at 37C 0.4x SSC ? After hybridization, the cover slips were removed and slides washed in 0.4 X SSC at 72°C for 2 min and in 2 X SSC at room temperature for 30 sec. The cells were counterstained with 10 μl of probes mixture 4'-6'- of diamine-2-phenylindole dihydrochloride (DAPI) and antifade in a ratio of 1:4. The image was analyzed using a Lieca m-FISH. One hundred (100) nuclei were scored for TEL/AML1 and 100 for BCR/ABL probe signals. Nuclei with ambiguous signals and cells with poor morphology were excluded from the scoring.

AML1 and TEL gene probes spanned the entire gene length, including the common breakpoint of t(12;21). This method directly labels TEL and AML 1 probes with different colors, (LSI TEL/AML1 ES Dual colors translocation probe Vysis Inc., IL, USA). TEL probe is labeled with the spectrum green fluorophore and the AML1 probe is labeled with the spectrum orange fluorophore. The TEL probe begins between exons 3 and 5 and extends approximately 350 Kb toward the telomere in chromosome 12. Thus, TEL probe covers the 12p13 region distal to the common breakpoints of t(12; 21).

The LS1 BCR/ABL dual color probe is a mixture of the LS1 ABL probe labeled with spectrum orange and the LS1 BCR probe labeled with spectrum green. The ABL probe begins between exons 4 and 5 and continues for about 300 kb toward the telomere of chromosome 9. The LS1 BCR probe begins between BCR exons 13 and 14 (major BCR exons being 2 and 3) and extends toward the centromere on chromosome 22 for approximately 300 kb, extending well beyond the M-bcr region. A nucleus lacking the t(9; 22) will exhibit two orange and two green signal patterns. In a nucleus containing a simple balanced t(9; 22), one orange, one green and one yellow (orange/green) fusion signal pattern will be observed. This simple probe design detects the 5' BCR /3' ABL gene fusion and is useful for detecting cells possessing this translocation in higher percentage.

Results:

In the present study, the ALL patients included 50 adults and 50 children As it is shown in table 1, from 50 adult ALL patients, 36 (72%) were male and 14 28%) were female. From 50 children ALL patients, 29 (58%) were male and 21 (42%) were female.

The average age of adult patients was 24.26 years with a median of 21. Average and median age of child patients were two and 15, respectively. The WBC count in adults was 200_96000/μl with average of 32136+/-26293/μl. The WBC count in children was 4000_55000/μl with average of 15416+/-13186/μl.

The lowest and highest survival times for adults were 14 months and 85 months, respectively, with an average of 33 months. This range was five months and 118 months for children with an average of 46 months (the survival time was con-

sidered only for those patients which it had past five years from their being diagnosed with ALL. Also, some of the patients were alive at the time of study, so their survival was considered from the time of diagnosis until study time). Average ages of patients, and survival periods are given in Table 1.

Table 1: Some general characteristics of the patients

Patients	Adults	Children
Total number of patients	50	50
Male	36(36%)	29(58%)
Female	14(28%)	21(42%)
Age(years)	24.26(average)	2-15
Lowest survival period	14	5
Highest survival period	85	118

As the age of affection increased, the WBC count raised. The survival time decreased with increase in WBC count.

In morphologic classification of FAB in adults with ALL, 22 patients (44%) were type L1 and 1 patient (2%) was of type L3. In pediatrics, 23 (46%) patients were type L1, 27 patients (54%) were type L2, and finally 45% of whole patients were in group type L1, 54% were type L2, and 1% were type L3. As the age of the patient increased, the WBC count also raised. In this study, with every one year increase in the age of the patient, the survival time was reduced by 0.6 months. The survival time also decreased with increases in WBC count. In this study, out of 10000 cells at the interphase stage (without considering 2500 cells related to 25 transplant patients), 8627 cells were normal regarding to TEL gene numbers containing two genes of TEL, while 1373 cells had an increase or decrease in the number of TEL genes. The increase in the number of all four genes under study was more common than their decrease. The ABL gene had the greatest number of normal genes, while the AML1 gene had the least. In our study, approximately 1.14% of adults cells had none of the two TEL genes, 2.68% lacked one of them, 3.98% had three copies of TEL genes, 3.90% had two excess copies, and 0.68% had more than four copies of the TEL gene (Table 2).

The cells which contained more than two copies of each gene were more than the cells containing less than two copies. On the other hand, the numerical chromosomal malformations in ALL patients were mostly of increased type. Of 100 patients, in 25 (15 adults and 10 children) who had BMT, the youngest was 16 years old and the oldest was 33 years old. In the cells of six patients with transplantation during metaphase, there were no cells with normal chromosomes before BMT, while after BMT more than 65% of their cells were chromosomally normal. In adults, the number of BCR/ABL fusions decreased after BMT. None of the patients who had TEL/AML1 went under BMT. The genetic variation of the metaphasic cells was very high but this variation decreased and normal chromosome containing cells increased after BMT. BMT caused a decrease in

both numerical and structural chromosomal malformations, though structural malformations were influenced more by BMT and significantly decreased after.

Table 2: The average number of copies of studied genes in bone marrow of adult and child ALL patients

	Average number of copies of the gene							
	Studied gene	**Average number of cells without gene (%)**	**Average number of cells containing one copy of gene (%)**	**Average number of cells containing two copies of gene (%)**	**Average number of cells containing three copies of gene (%)**	**Average number of cells containing four copies of gene (%)**	**Average number of cells containing more than four copies of gene (%)**	**Sum (%)**
adults	TEL	1.14	2.68	87.60	3.98	3.90	0.68	100
	AML1	1.58	3.14	86.64	6.26	2.10	0.50	100
	BCR	1.40	3.96	87.94	3.80	2.30	0.66	100
	ABL	1.16	2.72	89.28	3.94	2.24	0.74	100
children	TEL	1.00	3.82	84.74	5.96	3.34	1.12	100
	AML1	1.66	3.80	84.06	6.32	3.48	0.94	100
	BCR	1.24	4.16	86.18	5.02	2.60	0.76	100
	ABL	1.26	3.14	86.12	5.50	3.06	0.68	100

Regarding the FISH studies, in four adult patients which had BCR/ABL fusions before BMT, the number of BCR/ABL fusions decreased after BMT. None of adult patients who had TEL/AML1 underwent BMT.

In child bone marrow transplanted patients, no one had BCR/ABL fusion, while two of the patients who had t(12; 21) translocations went under BMT. Two child patients with TEL/AML1 fusion before BMT carried this fusion after BMT (Table 3).

Table3: The state of TEL, AML1, ABL and BCR genes in transplanted patients

Groups	**Adults**				**Children**			
	Gain		Loss		Gain		Loss	
Gene	before BMT	after BMT	before BMT	after BMT	before BMT	after BMT	before BMT	After BMT
TEL	4	3	1	0	5	4	1	0
AML1	6	2	2	1	5	4	1	1
BCR	3	1	2	0	6	6	1	0
ABL	5	2	1	0	5	5	0	0

Investigating the affection of close relatives of patients concerning ALL, breast, and prostate cancers revealed that 17 patients had positive familial history and 83 patients had negative familial history of these cancers.

The disccusion concerning the correlation between positive or negative familial history and clinical manifestations follows.

Discussion:

FISH has become a powerful tool in cytogenetic analysis. Recent reports have shown that t(12;21), (23% in children and 3.3% in adults), ,t(4;11), t(8;14), and t(1;19), which are the most common translocations, can not be detected by cytogenetic techniques. Structural chromosomal abnormalities called false diploidity mostly included the Philadelphia chromosome. In our study no significant correlation was found between the presence of Ph+ and survival period, both in adults, and children. The presence of a Ph chromosome in adults was associated with high WBC, L2 FAB- classification and with old age.

In this study, all patients with the t (12; 21) (p13; q22) had pre-B ALL and all of them remained at the first remission with no relapse Translocation of chromosomes 12 and 21 using FISH was found in 3.3% and 23% in adult and child patients, respectively

In children, out of 50 patients, 12 contained t(12; 21). Seven of these patients (58%) with t(12; 21) had a decrease in the number of TEL genes. In contrast, out of the 38 patients with no TEL/AML1 fusion, only one case (2.6%) had a decrease in the TEL gene. This genetic defect had not been explored by routine cytogenetic procedures. Other studies also admit the inability of routine cytogenetic techniques in diagnosing TEL/AML1fusions.[44,45,46,47,48] Deletion of the TEL gene has been frequently reported by many investigators. McLean and his colleagues suggested that a decrease in the TEL gene can be due to a renewal of probable gene fusion oncogenic potential which had been abolished because of a lack of the natural allele of TEL and consequently, growth advantage, compared to cells with t(12;21).[49] This suggestion can explain deletions of TEL allele. In adult patients, none of the two patients with TEL/AML1 fusion had a decrease in the number of TEL genes. Of 50 children, 15 patients had an increase in the number of AML1genes, and only two of these patients had translocations t(12;21). The shared resource of most of these increases is the polisomic increase in chromosome 21. This abnormality had been recognized in seven cases by cytogenetic techniques. Other investigators have also reported this abnormality [50,51,52], however, eight child cases demonstrated an increase in the AML1 gene without polysomy of chromosome 21, and all of these eight patients had structural malformations in cytogenetic studies. Average WBC count in these patients was more than other patients in this group and they a shorter survival period (with 300 cells more three months less survival), but this difference was not significant statistically. It seems as if interchromosomal proliferation has been the cause of extra transcripts of the AML1 gene in cases which lack polychromosome 21. In adults, eight patients had an increase of the AML1 gene. In four of them, cytogenetic studies had revealed this abnormality. None of them had TEL/ AML1 fusion. The average survival after disease onset was 27 months, which was six months less than other adult patients. Out of 12 children with TEL/ AML1fusion, 11 patients survived and one patient expired. Patients with t(12; 21) were dif-

ferent with respect to number of TEL/AML1 fusions that varied from 32 to 75 fusions. In pediatric patients, where 86% of patients had this kind of fusion, there was a significant relationship between number of fusions and number of WBC (P=0.025), so that those patients who had more fusions had less WBC count. No reports addressing the number of fusions and WBC count were found, suggesting an urgent need for further investigation. In pediatric patients there was a significant relationship seen between the survival time of patients with t(12; 21) and without t(12; 21), with a very high confidential coeficiency (P<0.0001). The high survival time of the patients with t (12; 21) is due to very good prognosis of this translocation and appropriate response to therapy and chemotherapy with low dose. This is also consistent with the results of other studies. In contrast to pediatric patients, presence of the TEL/AML1 fusion didn't increase survival in adults and there was no significant relationship between the patients with positive or negative fusions. In this study, 85% of children with translocation t(12;21) also had a significant decrease in the TEL gene. This is also consistent with other studies[46,49,53,54,55] and this decrease in the natural allele of the TEL gene highlights its importance in the prognosis of cancer with t(12;21). Overall, 14 patients (12 children, 2 adults) had TEL/AML1 fusion. Using the Spearman correlation test, there was no significant relationship between the number of TEL/AML1 fusions, age, WBC count, and survival. This study was performed separately for 12 children with the TEL/AML1 fusion. There was a significant relationship between the number of fusions and WBC count (P=0.02), while there was no significant correlation between the number of TEL/AML1 fusions and age or survival time. In adult patients, eight carried the BCR/ABL fusion, with the number of fusions varying from 10 to 81. The relationship between the number of fusions with age and WBC count was found to be insignificant. There was a significant relationship between survival time and being positive (more than six fusions) or negative (less than six fusions) for TEL/AML1 fusion, as detected by the Mann-Whitney U test, but an insignificant relationship for the BCR/ABL fusion was observed. In adult patients, there was no significant correlation between survival time and type of fusion (positive or negative), while in pediatric patients, those who were positive for the TEL/ABL fusion had significantly longer survival periods compared to others (P<0.0001), and patients positive for BCR/ABL had significantly less survival (P<0.066).

A direct relationship between the presence of fused TEL/AML1 genes and decreased WBC (P<0.05) was observed, but this was not significant in adults. The adults with more than 50000 WBC had a significantly lower survival period (P<0.05). One patient had the fused TEL/AML1, accompanied by additional materials, including TEL, ABL, and BCR genes, with a 43000 WBC, and with a survival period of 55 months. The incidences of the BCR/ABL fusion, and the signal distribution of ABL and BCR in the present Iranian ALL patients, regarding chromosomal gains and losses, are in concordance with previous reports. In

spite of this, the incidence is lower than that reported from Hong Kong and CALGB studies.[26] The number of TEL (P=0.174) and ABL genes (P=0.173) were not related to the gender of the patient, as determined by a Chi-Square test, though the number of AML1 genes (P=0.001) and the number of BCR genes (P=0.033) have been associated to gender in these patients,with men demonstrating a greater increase in these genes. Performing this study in adult patients, the number of ABL genes (P=0.962) was independent of gender while the number of TEL genes (P=0.002), AML1 genes (P=0.001), and BCR genes (P=0.009) was dependent on the gender of the patients. In children, the number of ABL genes (P=0.141) was independent of gender, while the number of TEL genes (P=0.001), AML1 genes (P=0.001) and BCR genes (P=0.079) was dependent on gender. Among 50 adult patients, eight (16%) had t(9; 22) detected by FISH. The frequency of this translocation was 16% which is compatible with findings in other studies.[56] In pediatric patients, those who had BCR/ABL had less survival compared to other ALL patients (P=0.06). Although the P-value was not less than 0.05, but 0.06, it used a high confidential coeficiency and up to 80% is compatible with other studies. ALL is more common in males. According to statistical analysis, the ABL gene had no relationship to gender in ALL patients, while TEL, AML1, and BCR genes were associated to gender in adults. TEL and AML1 genes were related to pediatrics and AML1 and BCR genes were dependent on gender in all patients. In all cases, the increase in these genes was greater in males compared to females. It is possible that the apoptosis of cells containing abnormal genetic matter in female patients is the cause for deletion and decrease of frequency of the aforementioned genes. The role of feminine hormones in adult female patients was also under observation. In two out of 25 patients no chromosomal abnormality was seen in any of their cells, two to three months after BMT and in seven of them, there were three metaphase cells having chromosomal malformation after BMT, indicating a success of 90%. In all patients between the ages of 5 to 15 years, normal karyotype was seen after BMT, while only nine patients between the age of 3 to 12 had shown normal karyotype before BMT. One patient had tetrasomy of chromosome 12 before BMT, but after BMT there was two cases of trisomy of chromosome 12 in two metaphase cells (Table 4).

Regarding the findings in other studies (57,58,59); the decrease in the number of numerical chromosomal abnormalities in our study after BMT was compatible while structural chromosomal abnormalities, especially in pediatric patients. The structural chromosomal abnormalities decreased significantly in our patients after BMT, while in other studies this change has not been significant in children and sometimes even new structural chromosomal abnormalities had been created. The reason for this difference can be the time of sampling after BMT which was 2.5 months for our patients and was five months in other studies.

Table 4: Comparison of karyotypic results before and after BMT

Line	Code	Age	Before transplantation	After transplantation
1	72- 920	16	47, XX, +mar., del. (5)(P12)/47,XX+mar./pseudo hyperperdiploididy[2] Mar.®small areocentric	46,XX[9]/47,XX,tmar.[3]
2	72-1038	20	,XX[10]/47,XX+Mar./45,XX-21/46,XX,del.(6)(q23)[1]	4] 46, XX [15]/47, XX, +mar. [
3	74-2478	16	45,XY,-21[12]/46,XY[2)	46,XY[12]/45,XY,-21[3]
4	75-3737	19	Pseudo diploidy/46,XY,del.(2)(Pter)[14]	46, XY [10]/46, XY, del. (2) (Pter.)[5)
5	75-3681	32	47,XY,+mar.[10]/Poly[3]	46,XY[5]/48,XY,+11,+12
6	77-4955	21	45,XY,+1, -21, -22[10)	46,XY[12]/47,XY, +3
7	77-4970	23	46, XY [14]/46, XY,-22+frag. [4]/47, xy, +del. (1) (qter.)	46,XY[2]/46,XY,9q+,15P+[12]
8	78-6278	17	47,XY,+8[7]/46,XY,inc.frag.[2]	46,XY[10]/47,XY, +8[2]
9	78-6457	16	46,XY[7]/47,XY, +21[11]	46,XY[13]/46,XY,9q+[3]/45,X[4]
10	79-7010	21	47,XX,+14[12]/47,XX,+4,7q+[3]	46,XX[9]/47,XX,+14[3]
11	77-5009	22	47,XX,+4[7]/46,XX[4]	46,XX[10]/47,XX,+4[1]
12	76-3952	20	46,XY[10]/47,XY,+6P[7]	46,XY[12]/45,XY,-13[1]
13	80-7315	17	47,XY,+4[8]/46,XY[5]	46,XY[13]/47,XY,+4[2]
14	75-3101	15	46,XX,-6,+mar.[14]/45,X[2]	46,XX[10]/45,X[1]/46,-6,+mar[5]
15	76-4518	3	46,XY,+14,-13[10]/46,XY[4]	46,XY[14]/45,XY-13[3] graft from umbilical cord)
16	78-5992	5	47,XY,+21[5]/45,X,-Y[3]	46,XY[12] graft from sister
17	78-6059	12	46,XY[9]/46,del.(X)(q2)Y[5]	46,XY[13]/45,X[1]/46,XY,6q- [1]
18	78-6139	14	47,XX,+3[10]/48,XX,+3,+13[3]	46, XX [14]/47, XX, +8[3]/47, XX, del. (1) (qter.)[1]
19	78-6340	12	46,XX[12]/45,XX,-1[5]	46,XX[15]/45,XX,-1[3]
20	80-7344	14	48,XY,+10+17[11]/46,XY[5]	46,XY[10]/47,XY,+17[3]
21	76-4307	14	47,XX,+2,del.(3)(Pter)[12]/46,XX,Inc.2frags.[10]	46,XX[8]/46,XX,Inc./frag.[4]
22	76-4687	12	47,XX,+17,del.(11)(qter)[12]/46,XX[3]	46,XX[11]/47,xx,+17[5]
23	79-7187	11	48,XY,+1q,+20[12]/49,XY,+1q,-1,+2,+20[3]	46,XY[11]/47,XY+20[5]
24	77-5472	30	48,XY,+12+12[10]/46,XY[5]/45,XY-15[3]	46,XY[7]/45,XY,-15[3]/47,XY,+12[2]
25	77-5579	33	46,XY[12]/48,XY,+4P,-1,+6,-13[7]	46,XY[13]/47,XY,+6[6]

In pediatric patients there was no significant change in the number of BCR/ABL and TEL/AML1 fusions after BMT, while in adults there was significant decrease in the number of these fusions after BMT. This shows that BMT is more successful in adult patients compared to pediatrics. In 17 patients, there was a positive familial history of ALL, breast cancer, and prostate cancer in their first degree relatives. But only one case demonstrated a significant relationship between familial history and clinical manifestations. Only adult patients with a positive FH (familial history) had more survival compared to others. The interviews with these four adult patients and their families showed no other cause for this difference except previous recollections and mental preparation for accepting the disease, knowing the effects of early therapy and acquaintance with therapy procedures, and accessing drugs. There was no significant correlation between the median of survival time and familial history in FH positive and negative patients (p=0.662). In

conclusion, FISH is an effective diagnosing technique that can help us make quick diagnoses and consequently, implement immediate interventions for ALL patients. We found BMT therapy to be more effective than other treatments, and offers the best quality of life. For these reasons, we believe these techniques should be used in clinics as standard treatment for these cases.

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