Chapter 1

Genetics of Acute Lymphoblastic Leukemia

Ruth Maribel Forero, María Hernández and
Jesús María Hernández-Rivas

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1. Introduction

Acute lymphoblastic leukemia (ALL) is mainly a disease of childhood that arises from recurrent genetic alterations that block precursor B- and T-cell differentiation and drive aberrant cell proliferation and survival [1]. Due to the advances in the cytogenetic and molecular characterization of the acute leukemias in the past two decades, genetic alterations can now be identified in more than 80% of cases of ALL [2]. These genetic lesions influence the prognosis and therapeutic approach used for treatment of ALLs [3]. This chapter describe genetic subtypes of ALL according to the hematological malignancies classification (WHO) 2008, risk groups, frequency of cytogenetic abnormalities, and their relationship with the prognosis of ALL, copy number alterations and somatic mutations in ALL.

2. Acute Lymphoblastic Leukemia (ALL) — Genetic subtypes

2.1. Definition and genetic subtypes according to the hematological malignancies classification (WHO) 2008

Acute lymphoblastic leukemia (ALL) is mainly a disease of childhood that arises from recurrent genetic alterations that block precursor B- and T-cell differentiation and drive aberrant cell proliferation and survival [1]. ALL is characterized by the accumulation of malignant, immature lymphoid cells in the bone marrow and, in most cases, also in peripheral blood. The disease is classified broadly as B- and T-lineage ALL [1].

ALL occurs with an incidence of approximately 1 to 1.5 per 100,000 persons. It has a bimodal distribution: an early peak at approximately age 4 to 5 years with an incidence as high as 4 to 5 per 100,000 persons, followed by a second gradual increase at about age 50 years with an

incidence of up to 2 per 100,000 persons. ALL, the most common childhood malignancy, represents about 80% of all childhood leukemias; but only about 20% of adult leukemias [4]. The rate of success in the treatment of ALL has increased steadily since the 1960s. The five-year event-free survival rate is nearly 80 percent for children with ALL and approximately 40 percent for adults [5].

Diagnosis of ALL relies on an assessment of morphology, flow cytometry immunophenotyping, and identification of cytogenetic-molecular abnormalities [4]. Conventional and molecular genetics allow the identification of numerical and structural chromosomal abnormalities and the definition of prognostically relevant ALL subgroups with unique clinical features [6, 7]. However, acute lymphoblastic leukemia subtypes show different responses to therapy and prognosis, which are only partially discriminated by current diagnostic tools, may be further determined by genomic and gene expression profiling [4]. More accurate delineation of genetic alterations can also provide information important for prognosis. Minimal residual disease (MRD) detection and quantification have proven important in risk-group stratification for both pediatric and adult ALL [7].

It seems likely that one or several changes in the genome are required for a blast cell to evolve into a leukemic clone, and that all cases probably harbor some form of genetic alteration [7]. Due to the advances in the cytogenetic and molecular characterization of the acute leukemias in the past two decades, genetic alterations can now be identified in greater than 80% of cases of ALL [2]. Improvement in recognizing abnormalities in the blast cells will help in understanding the mechanisms that underlie leukemogenesis.

The cloning and characterization of recurrent chromosomal translocations has allowed the identification of genes critical for understanding of the pathogenesis and prognosis of ALL [5, 8, 9]. These genes are implicated in cell proliferation and/or survival, self-renewal, cell differentiation and, and cell cycle control [10, 11]. The main causes of gene deregulation are: (i) oncogene activation with ensuing ectopic or over-expression, which is mainly due to juxtaposition with T-cell receptor loci; (ii) gain of function mutations; (iii) tumor suppressor gene haploinsufficiency or inactivation, which is usually the result of deletion and/or loss of function mutation; and (iv) chromosomal translocations producing fusion proteins which are associated with specific subgroups of ALL [10].

Efforts to define the genetic lesions that underlie ALL have identified a number of different subtypes of ALL based on their lineage (T- versus B-cell), chromosome number, or the presence or absence of chromosomal translocations. Collectively, these genetic lesions account for approximately 75% of cases, and their presence significantly influences the therapeutic approach used for treatment [3].

B-lineage ALL (B-ALL) shows considerable genetic heterogeneity. Within the category ''B lymphoblastic leukemia/lymphoma with recurrent genetic abnormalities'', the 2008 World Health Organization classification of hematopoietic neoplasms recognizes seven recurrent genetic abnormalities including t(9;22) (q34;q11.2) *BCR-ABL1*, t(v;11q23) *MLL* rearranged, t(12;21)(p13;q22) *TEL-AML1* (*ETV6-RUNX1*), t(5;14)(q31;q32) *IL3-IGH*, t(1;19) (q23;p13.3) *E2A-PBX1* (*TCF3-PBX1*), hypodiploidy and hyperdiploidy [12].

Burkitt lymphoma/mature B-ALL (BL) was included in the category of mature lymphatic neoplasms in the new WHO classification [12]. BL is characterized by translocation of *MYC* at 8q24.21 with an immunoglobulin gene locus, which in most cases is the immunoglobulin heavy chain locus (*IGH@*, 14q32.33) with rare translocations with the light chain genes for kappa (*IGK@*, 2p11.2) and lambda (*IGL@*, 22q11.2). These translocations result in constitutive expression of the *MYC* gene in peripheral germinal-center B cells, driven by the immunoglobulin gene enhancer [13]. The BL clone must acquire chromosomal aberrations secondary to the *IG-MYC* fusion. The most frequent secondary changes in BL detected by conventional cytogenetics are gains in 1q as well as in chromosomes 7 and 12 [14, 15].

In B-ALL, malignant cells often have additional specific genetic abnormalities, which have a significant impact on the clinical course of the disease. In contrast, although the spectrum of chromosomal abnormalities in T-lineage ALL (T-ALL) has been further widened by the finding of new recurrent but cryptic alterations, no cytogenetically defined prognostic subgroups have been identified [16, 17].

T-ALL is mainly associated with the deregulated expression of normal transcription factor proteins. This is often the result of chromosomal rearrangements juxtaposing promoter and enhancer elements of T-cell receptor genes *TRA@* (14q11), *TRB@* (T-cell receptor b, 7q34-35), *TRG@* (T-cell receptor g, 7p15) and *TRD@* (T-cell receptor d, 14q11) to important transcription factor genes [18]. In most cases, these rearrangements are reciprocal translocations, and lead to a deregulation of transcription of the partner gene by juxtaposition with the regulatory region of one of the TCR loci (e.g. *TCRB*/*HOXA*, *TCRA*/*D-HOX11*, *TCRA*/*D-LMO2*, *TCRA*/*DLMO1*). These chromosomal aberrations affect a subset of genes with oncogenic properties, such as 1p32(*TAL1*), 1p34(*LCK*), 8q24(*MYC*), 9q34(*TAL2*), 9q34(*TAN1*/*NOTCH1*), 10q24 (*HOX11*), 11p13 (*RBTN2*/*LMO2*), 11p15(*RBTN1*/*LMO1*), 14q32(*TCL1*), 19p13 (*LYL1*), 21q22(*BHLHB1*) and Xq28 (*MTCP1*) [10, 17].

Other type of rearrangement in T-ALL, mostly translocations, results in formation of 'fusion genes' that are associated with specific subgroups of T-ALL (*CALM-AF10*, *NUP98*-t, *MLL*-t and *ABL1*-fusions). In these translocations, parts of both genes located at the chromosomal breakpoints are fused 'in frame' and encode a new chimeric protein with oncogenic properties [10]. Chromosomal translocations producing fusion proteins also are associated with specific subgroups of T-ALL.

In addition, gain of function mutations (*NOTCH1* and *JAK1*) and tumor suppressor gene haploinsufficiency or inactivation, which is usually the result of deletion (*CDKN2A-B*) and/or loss of function mutation (*PTEN*); are frequent in T-ALL. These genetic alterations could be concomitant with other genomic changes [10, 19].

2.2. Risk groups in ALL

During the past three decades, the prognosis of has been improved and the treatment achieved cure rates exceeding 80%. ALL in adults has followed the same trend with long-term survival of about 40%. One main factor behind this improvement is the development of risk-adapted

therapy, that permit to stratify the patients in different clinical categories according to risk factors with prognostic influence and to define the intensity and duration of treatment [20].

The prognosis of patients with ALL is influenced by clinical, hematologic and genetic factors, including age, leukocyte count at diagnosis, percentage of blast in peripheral blood, immunophenotype, central nervous system (CNS) involvement, the presence or absence of mediastinal tumor, cytogenetic and molecular alterations and the presence of minimal residual disease (MRD) in different stages of treatment which is currently a defined risk of adapted therapy strategies [20-24].

With respect to age, children less than 24 months and adults more than 50 years old have a worse prognosis, while the better results are achieved for children between 1 and 10 years, followed by adolescents and young adults. The leukocytosis (>30X10^{9}L in B-ALL and >100X10^{9}L in T-ALL), the phenotype Pro-B ALL, and T-ALL, are related to a poor outcome and are used to stratify patients as high risk [23].

The study of these prognostic factors allows recognition of three subgroups with outcome clearly differentiated in children: standard risk (40% of cases - 90% survival), intermediate (45-50% - 70-80% survival) and high risk (10-15%-less than 50% survival) [23, 25], and two subgroups in adult, standard-risk (20-25% of cases, 60% survival) and high risk (75-80% - 30% survival) [23, 26].

3. Cytogenetic alterations in ALL

3.1. Cytogenetic alterations in B-cell precursor ALL (BCP-ALL)

A correlation between prognosis and the karyotype at diagnosis in ALL was firstly demonstrated by Secker-Walker (1978) [24]. Subsequently, during the third International Workshop on Chromosomes in Leukemia (IWCL, 1983), the first large series of newly diagnosed ALL were analyzed to establish cytogenetic and prognostic correlations. Sixty-six percent of the patients analyzed showed clonal aberrations, which were identified both high-risk and low-risk ALL patients [27]. Since then it has been considered that the cytogenetic alterations have prognostic value of first order in the ALL.

Development of methods in cytogenetics has contributed to the understanding that ALL is not a homogeneous disease. Chromosome abnormalities have been detected by conventional G-banding in approximately 60–70% of all cases [7, 28]. Abnormal karyotypes have been reported in up to 80% of children and 70% of adults with ALL [29, 30]. There had been considerable developments in fluorescence in situ hybridization (FISH) for the detection of significant chromosomal abnormalities in leukemia in the 1990s [31]. The development of 24-color fluorescence in situ hybridization (FISH), interphase FISH with specific probes, and polymerase chain reaction (PCR) methods has improved the ability to find smaller changes and decreased the proportion of apparently normal karyotype to less than 20% in ALL [7].

In cases with B-ALL (excluding mature B-ALL), the most important subgroups for modal number are hypodiploidy, pseudodiploidy, and hyperdiploidy with a chromosome number

greater than 50 [32]. The most structural rearrangements include translocations that generate fusion transcripts with oncogenic potential. The most important of the translocations are t(1;19)(q23;p13)(*TCF3-PBX1* fusion gene; alias *E2A-PBX1*), t(4;11)(q21;q23)(*MLL-AFF1* fusion gene; alias *MLL-AF4*), t(9;22)(q34;q11)(*BCR-ABL1*),and t(12;21)(p13;q22)(*ETV6-RUNX1* fusion gene; previously *TEL-AML1*) [32]. These cytogenetic subgroups have distinctive immunophenotypic characteristic as well as age and prognostic associations [24].

3.1.1. Ploidy alterations

The presence of hypodiploidy (less than 45 chromosomes) is found in only 2% of ALL, and is associated with a very poor outcome [33]. The high hyperdiploidy (with more than 50 chromosomes) is the most common cytogenetic subgroup in childhood BCP-ALL, and associated to a long survival. Hyperdiploidy is more frequent in children (15%) than in adults (6%) [34].

The gain of chromosomes is nonrandom, the eight chromosomes that account for 80% of all gains are: +4(78%), +6 (85%), +10 (63%), +14 (84%), +17 (68%), +18 (76%), +21 (99%), and +X (89%) [24]. Trisomy 4, 10, and 17 are associated to favorable outcome in children [33]. Unlike hypodiploidy ALL patients, hyperdiploid ALL cases have an extremely good prognosis with event-free survival rates near 90% [21]. These patients seem to particularly benefit from high dose methotrexate [33].

Approximately 20% of hyperdiploid ALL have activating mutations in the receptor tyrosine kinase *FLT3*. These mutations are interesting because not only they trigger the activation of the tyrosine kinases as potential oncogenes in hyperdiploid ALL, but also in that it suggests that tyrosine kinase inhibitors could be of benefit to patients with this leukemia. [9].

3.1.2. E2A-PBX1 fusion t(1;19) (q23;p13)

The t(1;19) (q23;p13) represents 5% of children ALL, and 3% in adults ALL, this translocation is frequently associated with the pre-B immunophenotype, in approximately 25% of cases [5, 34, 35]. The t(1;19) (q23;p13) forms a fusion gene that encodes a chimeric transcription factor, *E2A-PBX1* (*TCF3-PBX1* fusion). It disrupts the expression the expression of *HOX* genes and the targets of the *E2A* transcription factor [5]. The t(1;19) has good prognosis with high-dose methotrexate treatment; however this translocation is a risk factor for CNS relapse [1, 21]

3.1.3. BCR-ABL fusion t(9;22) (q34;q11)

As a result of the t(9;22) (q34;q11)/Philadelphia chromosome (Ph+), the *BCR* gene at 22q11.2 is joined to the *ABL* protooncogene at 9q34, giving rise to the *BCR-ABL* fusion gene. The fusion gene encodes an oncogenic fusion protein with enhanced tyrosine kinase activity that interacts with *RAS, AKT*, and *JAK/STAT* pathways [1]. This translocation is found in approximately 3% of children and 30% of adults, and is associated with unfavorable prognosis [34]. Imatinib mesylate plus intensive chemotherapy improve early treatment outcome against Philadelphia chromosome–positive (Ph+) in ALL, one of the highest risk pediatric ALL groups, however imatinib resistance develops rapidly [36].

3.1.4. 11q23-MLL rearrangements

Chromosomal rearrangements of the human *MLL* gene are the most common genetic abnormality in the first year of life, but it occurs in only 8% of children and 10% adults with ALL [34]. The 11q23-*MLL* rearrangements are associated with high-risk pediatric, adult and therapy-associated acute leukemias [37].

Some 104 different *MLL* rearrangements of which 64 fused translocation partner genes (TPGs) are now characterized at molecular level [37]. The five most common *MLL* rearrangements, present about 80% of *MLL*-translocated acute leukemia (*MLL*-t AL), are t(4;11)(q21;q23) encoding *MLL-AF4*, t(9;11)(p22;q23) encoding *MLL-AF9*, t(11;19) (q23;p13.3)-ncoding *MLL-ENL*, t(10;11)(p12;q23)encoding *MLL-AF10*, and t(6;11)(q27;q23)encoding *MLL-AF6* [38].

MLL rearrangements are associated with unfavorable prognosis. However, outcomes could be improved with high-dose cytarabine for some rearrangements [1]. *MLL* fusions with *AF4, AF9* and *ENL* recruit small serine/proline-rich proteins with nuclear localization signals, which may generate unique chimeric transcriptional transactivators [1]. The t(4;11)(q21;q23) have poor prognosis and predominance in infancy, especially those < 6 months of age. This rearrangement has been associated with overexpression of *FLT3* [21].

3.1.5. ETV6-RUNX1 t(12;21) (p13;q22)

The t(12;21) (p13;q22) leads to a fusion *ETV6-RUNX1* (*TEL-AML1*). It occurs in 22% of children and 2% adults with ALL [34]. This translocation t(12;21)(p13;q22) is the most common translocation in childhood BCP-ALL [39]. Moreover, is associated to an excellent prognosis with intensive chemotherapy, including asparaginase therapy [1, 21]. *TEL-AML1* is a leukemogenic, chimeric transcription factor encoding the amino-terminal basic helix-loop-helix (bHLH) domain of the ETS family member *TEL* fused to the *AML1* DNA-binding Runt and transactivation domains [40]. *TEL-AML1* may generate a pre-leukemia clone by repression of activated *AML1* target genes or by *TEL* inhibition of other ETS family proteins via binding through the TEL's pointed domain [1].

3.2. Cytogenetic alterations in Burkitt lymphoma/mature B-ALL (BL)

3.2.1. *MYC/IG* (t(8;14), t(2;8) and t(8;22))

The t(8;14)(q24;q32) and its variants t(2;8)(p11;q24) and t(8;22)(q24;q11) are associated with BL [13]. The t(8;14) is most common, found in 85%, whereas t(2;8) and t(8;22) are found in around 5 and 10%, respectively [24]. The crucial event in all three reciprocal translocations is the juxtaposing of *C-MYC* (from 8q24) under the control of immunoglobulin (Ig) gene enhancers of the heavy chain (*IGH*-14q32), kappa light chain (*IGK*-2p12), or lambda light chain (*IGL*-22q11), leading to deregulation and increased transcription of *MYC* [13]. The 2008 World Health Organization classification of hematopoietic neoplasms, established that *MYC* translocations are not specific for BL. Most *MYC/IG* breakpoints in endemic BL originate from

aberrant somatic hypermutation. On the other hand, in sporadic cases the translocation involves the *IG* change regions of the *IGH* locus at 14q32 [12].

The abnormalities of *C-MYC* are an important step in the development of BL. *MYC* is a transcription factor with both activating and repressing function and is involved in the regulation of roughly 10–15% of all human genes. *MYC* regulates a number of critical biologic processes such as cell cycle control, cell growth, protein synthesis, angiogenesis, and apoptosis [41]. The upregulation of *C-MYC* disrupts many aspects of cell function, such as cell cycle progression, differentiation, metabolism, apoptosis, telomerase activity, and cell adhesion. These effects of *C-MYC* are likely to be of pathogenetic relevance in human tumors [42].

3.2.2. Secondary chromosome changes in BL

Several cytogenetic reports have correlated the presence of cytogenetic abnormalities with the outcome of patients with non-Hodgkin lymphomas, showing that secondary chromosome changes may influence the clinical phenotype of lymphoid tumors [43].

Most of the secondary chromosome changes are unbalanced rearrangements, leading to DNA gains or losses. These changes have been studied in Burkitt's lymphoma-derived cell lines and primary tumors by cytogenetic techniques including karyotype analysis [44-48], fluorescence in situ hybridization (FISH) [49], multiplex FISH (M-FISH) [50], spectral karyotype analysis (SKY), comparative genomic hybridization (CGH)[43, 51-54], and microarray analysis [55].

Additional recurrent chromosomal abnormalities have involved chromosomes 1, 6, 7, 12, 13, 17, and 22. Gains of the long arm of chromosomes 1 (+1q) or 7 (+7q) or 12 (+12q), deletion (del) 17p13 and abnormalities of band 13q34 usually occur in adult BL, without or in the setting of an HIV infection [13, 44-46, 51, 56]. Some secondary abnormalities have been associated with tumor progression, such as abnormalities on 1q, + 7q and del(13q) which have been independently associated with a worse outcome [43-46, 49, 50].

3.3. Cytogenetic alterations in T-ALL

Conventional karyotyping identifies structural chromosomal aberrations in 50% of T-ALL. Numerical changes are rare, except for tetraploidy which is seen in approximately 5% of cases. The presence of chromosomal abnormalities is not associated to the prognosis [19]. Some nonrandom translocations that are specific to T-lineage malignancies have been identified. They involve genes coding for transcriptional regulators transcriptionally deregulated in malignancies [57].

Extensive characterization of specific chromosomal abnormalities for T-ALL led to the identification of several oncogenes whose expression was up-regulated under the influence of the transcriptional regulation elements of genes which are normally expressed during T-cell differentiation [58]. T-cell malignancies are often associated with unfavorable features compared with childhood precursor B-cell ALL. However, the use

of more intensive treatments and risk adapted therapy has significantly improved the outcome of patients with T-ALL. Event-free survival rates of 60% to 70% are now reported in children [57].

3.3.1. Rearrangements involving TCR genes

3.3.1.1. Deregulation of homeobox genes

The homeobox (*HOX*) family of transcription factors is divided into two classes. Class I *HOX* genes are organized in four distinct clusters (*HOXA@*, *HOXB@*, *HOXC@* and *HOXD@*) at four chromosomal loci (7p15, 17q21, 12q13, and 2q31 whereas class II *HOX* genes are dispersed throughout the whole genome. In the class I *HOX* genes, the *HOXA@* cluster is involved in T-ALL, while that in the class II *HOX* genes, *TLX1* (*HOX11*) and *TLX3* (*HOX11L2*) have been extensively studied in the context of T-ALL [18].

3.3.1.1.1. TLX1 (HOX11) (t(10;14)(q24;q11) and its variant t(7;10)(q35;q24))

The translocation t(10;14)(q24;q11) and its variant t(7;10)(q35;q24) are a nonrandom alteration identified in T-ALL. Either of these is present in 5% of pediatric to 30% of adult T-ALL [1]. Both of them lead to the transcriptional activation of an homeobox gene, *HOX11* gene (*TLX1*; *TCL3*), that is not expressed in healthy T-cells, by bringing the *HOX11* coding sequence under the transcriptional control of regulatory sequences of the T-cell receptor gene (*TRA@* or *TRB@* genes, respectively). However, the overexpression of *HOX11* in thymocytes has been also demonstrated in the absence of a 10q24 rearrangement, suggesting that other, trans-acting mechanisms could lead to this aberrant gene expression, probably by disrupting gene silencing mechanisms that operate during normal T-cell development [18, 57, 58].

There is some evidence that *HOX11* may play an important role in leukemogenesis. It has been particularly shown that constitutive expression of *HOX11* favors expansion and, in some instances, immortalization of murine hematopoietic progenitors in vitro [59, 60]. However, *HOX11* has better prognosis than other T-ALL molecular subtypes [1].

3.3.1.1.2. TLX3 (HOX11L2) (t(5;14)(q35;q32))

The cryptic translocation, t(5;14)(q35;q32), is restricted to T-ALL, is present in approximately 20% of childhood T-ALL and 13% of adult cases. This translocation is associated with strong ectopic expression of another homeobox gene called *HOX11L2* (*RNX*; *TLX3*) [17, 58, 61], because of possibly the influence of regulatory elements of *CTIP2* (*BCL11B*), a gene highly expressed during T-lymphoid differentiation [17, 57]. Other variant chromosomal aberrations, each targeting *TLX3*, have been observed as well, including a t(5;7)(q35;q21), in which the *CDK6* gene is involved on 7q21[18].

Although *TLX1* and *TLX3* themselves and the gene expression profiles of *TLX1* and *TLX3* expressing T-ALL samples are very similar [18], the t(5;14) and/or *HOX11L2* ectopic expression has been associated with a very poor outcome in children with T-ALL [57].

However, the exact prognostic meaning of *TLX3* expression alone or in combination with other markers is not clear [18].

3.3.1.1.3. HOXA@ cluster (inv(7)(p15q34))

Other rearrangement involving *TCR* genes that affecting *HOXA@* cluster (7p15) is associated with the inv(7) (p15q34), t(7;7)(p15;q34), and t(7;14)(p15;q11). The chromosomal inversion inv(7)(p15q34) has been observed in approximately 5% of T-ALL cases. This inversion juxtaposes part of the *TRB@* locus (7q34-35) to part of the *HOXA@* cluster (7p15), resulting in elevated *HOXA10* and *HOXA11* expression. In addition, 2% of the cases showed elevated *HOXA10* and *HOXA11* expression in the absence of inv(7), suggesting that other activating mechanisms may exist [18].

In contrast to *TLX1* and *TLX3*, which are normally not expressed in the hematopoietic system, *HOXA10* and *HOXA11* are expressed in developing thymocytes. While *HOXA11* is expressed at different stages of T-cell differentiation, *HOXA10* expression is only detected at the earliest stages of differentiation, suggesting that its downregulation is required for full maturation of T-cells to the CD4 and CD8 single positive stages [18].

3.3.1.2. Deregulation of TAL1-related genes

TAL1-related genes (*TAL1, TAL2* and *LYL1*), encode a distinct subgroup of basic helix-loop-helix (bHLH) proteins that share exceptional homology in their bHLH sequences [62]. The malignant potential of these proteins is likely to reside largely within their HLH domains that potentially mediate sequence-specific DNA recognition [63].

Although expression of *TAL1*, *TAL2* or *LYL1* has not been observed during normal T-cell development, the rearranged alleles of these genes are readily transcribed in T-ALL cells, and the ectopic expression of these genes in T-lineage cells may be a contributing factor in T-ALL pathogenesis [62].

3.3.1.2.1. TAL1 (SCL,TCL5) (t(1;14)(p32;q11), t(1;14)(p34;q11) and t(1;7)(p32;q34))

Alteration of the *TAL1* (*SCL, TCL5*), a gene located on chromosome 1p32, is considered as the most common nonrandom genetic defect in childhood T-ALL. *TAL1* disruption is associated with a t(1;14)(p32;q11), t(1;14)(p34;q11) and t(1;7)(p32;q34) (*TRA@/TRAD@-TAL1* respectively) in 1% to 3% of childhood T-ALL [1, 57]. In other 9% to 30% of childhood T-ALL, *TAL1* is overexpressed as a consequence of a nonrandom submicroscopic interstitial deletion between a locus called *SIL* and the 5′ untranslated region (UTR) of *TAL1* at 1p32, giving rise to an *SIL-TAL* fusion transcript [19].

As the translocation as interstitial deletion disrupt the coding potential of *TAL1* in a similar manner, leading to its ectopic overexpression in T-cells [57]. Nevertheless, high expression levels of *TAL1* in the absence of detectable *TAL1* rearrangement are observed in about 40% of T-ALL [19].

The deletions aberrantly triggers activated *TAL1* during thymocyte maturation, promoting transformation [1]. *TAL1* alteration leads to silencing of genes target encoding E47 and E12 variants of E2A transcription factors. Several studies have proposed that the reactivation of silenced genes by administering histone deacetylase (HDAC) inhibitors may prove efficacious in T-ALL patients expressing *TAL1* [18, 57].

3.3.1.2.2. TAL2 (t(7;9)(q34;q32))

As a consequence of t(7;9) (q34;q32), the *TAL2* gene is juxtaposed to the *TRB@* locus. The *TAL2* gene is activated as a result of this translocation. The activation of the *TAL2* or *LYL1* genes is less common, affecting <2% of T-ALL patients [18, 62]. The properties of *TAL2* broadly resemble those described previously for *TAL1*. Therefore, this support the idea that both proteins promote T-ALL by a common mechanism [64].

3.3.1.2.3. LYL1 (t(7;19)(q34;p13))

In the t(7;19)(q35;p13), the *LYL1* coding sequences are juxtaposed to the *TRB@* locus. This gene is constitutively expressed in T-ALL, whereas its expression is absent in normal T-cells. The ectopic *LYL1* expression is found in some human T-cell leukemias, suggesting that it may participate in T-cell leukemogenesis. Similar to *TLX1*, *TLX3*, and *TAL1*, the ectopic expression of *LYL1* is mutually exclusive, although rare exceptions to this rule have been described [18].

LYL1 encodes another class II basic helix-loop-helix (bHLH) transcription factor that forms heterodimers with class I bHLH proteins, including E2A (E47 and E12) and HEB. *LYL1*-transgenic mice developed CD4+CD8+ precursor T-cell ALL (pre-T-LBL), probably by dimerization with E2A, inhibition of CD4 promoter activity, and downregulation of a subset of *E2A/HEB* target genes, suggesting a block in cell differentiation [1, 65]

3.3.1.3. Deregulation of LIM-domain only genes LMO1 and LMO2

3.3.1.3.1. LMO1 (t(11;14)(p15;q11) and LMO2 (t(11;14)(p13;q11))

The genes encoding the LIM-domain only proteins *LMO1* (*RBTN1* or *TTG1*, 11p15) and *LMO2* (*RBTN2* or *TTG2*, 11p13) are frequently rearranged with the T-cell receptor loci in T-ALL, resulting in overexpression of *LMO1/LMO2* [1, 66]. The most common alterations are t(11;14)(p15;q11) and t(11;14)(p13;q11) juxtaposing *LMO1* or *LMO2* to the *TRA@* or *TRAD@* loci [1], nevertheless other genetic alterations have also been reported like t(7;11)(q34;p15) and t(7;11)(q34;p13) translocations, involving *TCRB* and *LMO1* or *LMO2* loci [17].

Generally the ectopic expression of *LMO1* and *LMO2* are not mutually exclusive, because abnormal expression of *LMO1/2* has been found in 45% of T-ALL, even in the absence of typical chromosomal changes, but often in association with deregulation of *LYL1* (*LMO2*) or *TAL1* (*LMO1* and *2*) [19]. Studies in transgenic mice have showed that *TAL1* expression in itself is not sufficient to induce T-cell malignancies and that co-expression of *LMO1* or *LMO2* is strictly required [18].

3.3.1.4. Deregulation of family of tyrosine kinases — LCK gene (t(1;7)(p34;q34))

The lymphocyte-specific protein tyrosine kinase (*LCK*), a member of the SRC family of tyrosine kinases, is highly expressed in T-cells and plays a critical role in proximal TCR-based signaling pathways [67]. The *LCK* gene is activated due to the t(1;7)(p34;q34) that juxtaposing *LCK* with *TRB@* loci [18]. *ABL1* is located downstream of *LCK* in the TCR signaling pathway. Based on these results, *SRC* kinase inhibitors and the dual *SRC*/*ABL* kinase inhibitors could be used for treating T-ALL patients with hyperactive LCK [18].

3.3.1.5. Deregulation of MYB gene — Duplication and t(6;7)(q23;q34)

MYB is the cellular homolog of the *V-MYB* oncogene of the avian myeloblastosis virus. A t(6;7)(q23;q34), juxtaposing *MYB* to *TCRβ* regulatory elements, and a submicroscopic amplification of the long arm of chromosome 6 at 6q23.3 caused by ALU-mediated homologous recombination, has been detected in 8–15% of T-ALL. It leads to upregulation of *MYB* expression and a blockade in T-cell differentiation that could be reversed with *MYB* knockdown [1, 68]. The upregulation of *MYB* has raised expectations that *MYB* may be used as a molecular target for therapy in these patients [66].

Finally, other rearrangements involving TCR genes affect genes like *BCL11B* (inv(14)(q11q32); and t(14;14)(q11;q32)), *TCL1* (inv(14)(q11q32), and t(14;14)(q11;q32)), *CCND2* (t(7;12)(q34;p13.3), and t(12;14)(p13;q11)), *NOTCH1* (t(7;9)(q34;q34.3)), and *OLIG2* gene (t(14;21)(q11;q22)) [19, 24].

3.3.2. Fusion genes rearrangements

3.3.2.1. PICALM-MLLT10 (CALM-AF10) — t(10;11)(p13;q14)

The *PICALM-MLLT10* (*CALM-AF10*) fusion gene is caused by a recurrent translocation, t(10;11)(p13;q14). It is detected in about 10% of childhood T-ALL. and it has been associated with a poor prognosis [11]. This translocation has also been observed in other leukemias, including acute myeloid leukemia [18].

The precise mechanism for *CALM-AF10* mediated transformation is not known, although *CALM-AF10* T-ALL are characterized by overexpression of *HOXA* cluster genes, including *HOXA5, HOXA9*, and *HOXA10*. *CALM-AF10*+ T-ALL has also showed overexpression of several *AF10* downstream genes (*DNAJC1, COMMD3, BMI1* and *SPAG6*) located close to the *AF10* gene breakpoint. From these four *AF10* downstream genes, *BMI1* is the only one known to be associated with an increase of self-renewal of hematopoietic stem cells and oncogenesis [69].

3.3.2.2. MLL-fusions

Translocations implicating *MLL* with various partners represent about 4% of T-ALL cases [18]. The t(11;19)(q23;p13) *MLL-MLLT1* (ENL) gene fusion is the most common *MLL* translocation partner in T-ALL. Nevertheless, other *MLL* translocations also occur in T-ALL [11].

CALM-AF10+ T-ALL and *MLL-t* AL share a specific *HOXA* overexpression, triggering activate common oncogenic pathways [69]. *MLL* fusion proteins enhance transcriptional activity, resulting in increased expression of *HOXA9, HOXA10, HOXC6*, and overexpression of the *MEIS1 HOX* coregulator [18].

MLL controls skeletal patterning, regulates the establishment of functional hematopoietic stem cells, and early hematopoietic progenitor cell development [1, 70]. T-ALL cells with *MLL* fusions are characterized by differentiation arrest at an early stage of thymocyte differentiation, after commitment to the TCR gammadelta lineage [11].

3.3.2.3. ABL1-fusions

Translocations of *ABL1* are rare, except for *NUP214-ABL1* fusion (t(9;9)(q34;q34)) identified in up to 6% of T-ALL as a result of episomal formation with amplification. Recurrent translocations involving *NUP98*, such as the t(4;11)(q21;p15) with the *NUP98/ RAP1GDS1* gene fusion), another protein of the nucleopore complex, are reported very rarely [19]. The t(9;12)(p24;p13) encoding *ETV6-JAK2* fusion gene, with an important leukemogenic role, results in constitutive tyrosine kinase activity in positive T-ALL patients [71].

4. Copy number alterations in acute lymphoblastic leukemia

In spite of continually improving event-free (EFS) and overall survival (OS) for ALL, particularly in children, a number of patients on current therapies will relapse. Therefore it is important to know the group of patients with high risk of relapse [72, 73]. As the risk-stratification of ALL is partly based on genetic analysis, different genomic technologies designed to detect poor-risk additional genetic changes are being expanded substantially. Analyses of somatic DNA copy number variations in ALL aided by advances in microarray technology (array comparative genomic hybridization and high density single nucleotide polymorphism arrays) have allowed the identification of copy gains, deletions, and losses of heterozygosity at ever-increasing resolution [74].

Several microarray platforms have been used for the analysis of DNA copy number abnormalities (CNAs) in ALL, such as array-based comparative genomic hybridization (array-CGH), bacterial artificial chromosome (array-BAC) array CGH and oligonucleotide array CGH (oaCGH), single nucleotide polymorphism array (aSNP) and single molecule sequencing [75]. These microarray platforms vary in resolution, technical performance, and the ability to detect DNA deletions, DNA gains, and copy neutral loss of heterozygosity. These techniques have improved the detection of novel genomic changes in ALL blast cells [76]. The aCGH also detects the majority of karyotypic findings other than balanced translocations, and may provide prognostic information in cases with uninformative cytogenetics [77, 78]. In addition, the use of these methods documented multiple regions of common genetic cryptic alterations. These analyses provide information about multiple submicroscopic recurring genetic alterations including target key cellular pathways. However, many aberrations are still undetected in most cases, and their associations with established cytogenetic subgroups remain unclear [28, 79].

4.1. CNA in BCP-ALL

Most of ALL (79-86%) showed alterations in the number of copies (CNA) by aCGH techniques. The CNA frequently involved chromosomes 1, 6, 8, 9, 12, 15, 17 and 21; and rarely chromosomes 2, 3, 14 and 19. The losses have been more frequent than gains [6, 7, 28, 35, 77, 78, 80-85].

In precursor B-cell ALLs, most of the abnormalities have been gains of 1q (multiple loci), 9q, 17q, amplification of chromosome 21 (predominantly tetrasomy 21), and loss of 1p and 12p. Other recurrent chromosomal rearrangements have been found in both B-and T ALLs, such as loss of 6q (heterogeneous in size), 9p (9p21.3), 11q, and 16q, as well as gain of 6q and 16p. Other recurrent findings have included dim (13q), dim (16q) and enh (17q) [6, 7, 28, 35, 77, 78, 80-85] (Figure 1).

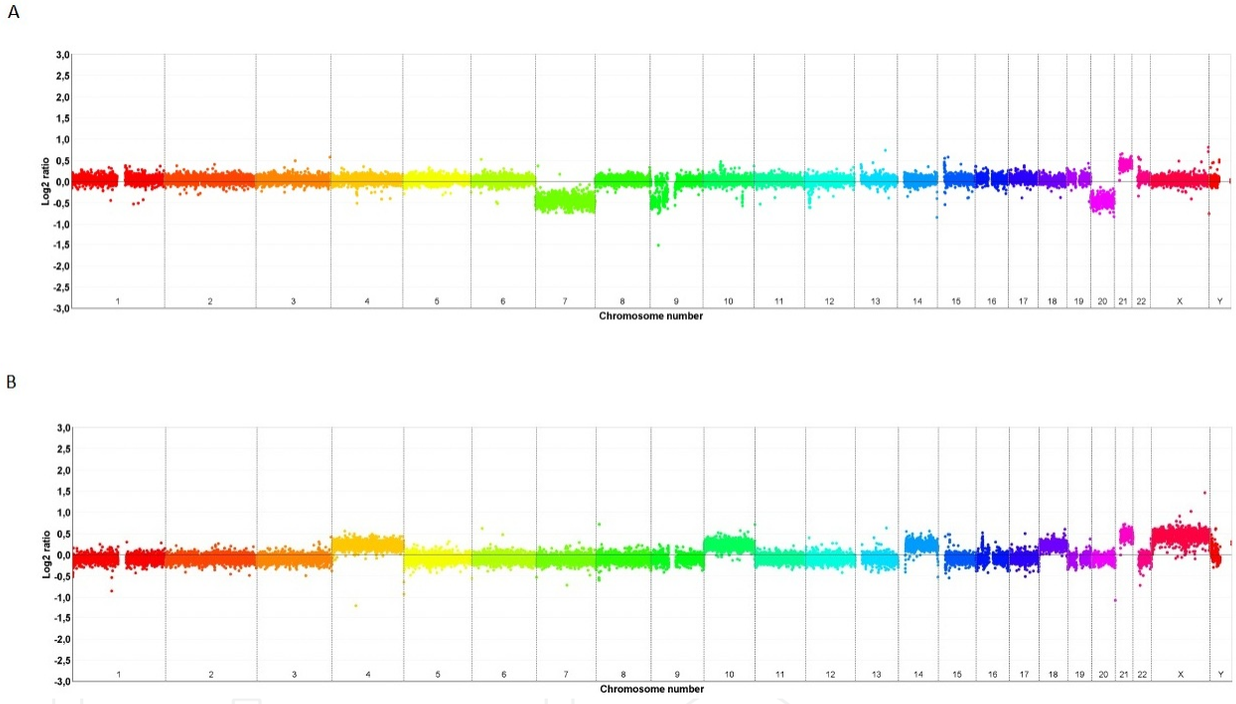

Figure 1. Copy number alterations in two newly diagnosed BCP-ALL patients. A. Male patient with losses of 7, 9p24-q21 and 20 chromosome and gain of 21q chromosome. B. Male patient with whole gains of 4, 10, 14,18, 21, and X chromosome.

Several observations suggest that the CNAs are biologically important. The identification of these recurrent chromosomal rearrangements in ALL has defined Minimal Critical Regions (MCR), which are target small regions of the genome, that are often small enough to pinpoint the few candidate genes that present in these chromosomal regions [75].

Many of these MCR contain genes with known roles in leukemogenesis of ALL. These lesions include deletions of lymphoid transcription factors and transcriptional coactivators (e.g. *PAX5, EBF1, E2-2, IKZF1*-Ikaros, *ETV6* (*TEL*), *ERG*, *TBL1XR1*, and *LEF1*), tumor suppressor and cell cycle regulatory genes (e.g. *CDKN2A/B, NF1, PTEN, RB1*, and *ATM*), as well as genes with other established roles in B-cell development, such as *RAG1* and *RAG2*, *FYN*, *PBEF1* or *CBP*/

PAG. Moreover putative regulators of apoptosis (e.g. *BTG1*), lymphoid signaling molecules (e.g. *BTLA/CD200, TOX*), micro-RNAs (e.g. mir-15a/16-1), steroid receptors (e.g. *NR3C1, NR3C2*), genes at fragile sites (e.g. *FHIT, DMD*), and genes with unknown roles in leukemogenesis of ALL (e.g. *C20orf94/MKKS, ADD3*, and *DMD*) have been located in these regions. It is notable that about 40% of B-progenitor ALL cases present genomic alterations in genes that regulate B-lymphocyte differentiation [6, 7, 28, 35, 77, 78, 80-86] (Table 1).

Loss/gain	Chromosome	Cytoband	Size (Mb)	Start position (Mb)*	End position (Mb)*	Candidate genes	Reference
loss	1	p33	0.039	47.728	47.767	*TAL1*	[75, 85]
loss	1	q44	1.74	245.113	246.853	*LOC440742 Adjacent to gen ZNF528*	[80, 110]
loss	2	p21	0.287	43.425	43.712	*THADA*	[75, 85]
loss	3	p22.3	0.306	35.364	35.670	*No annotated gene ARPP-21*	[85, 110]
loss	3	p14.2	0.254	60.089	60.343	*FHIT*	[75, 83, 85, 110]
loss	3	q13.2	0.148	112.055	112.203	*CD200, BTLA*	[75, 85, 110]
loss	3	q26.32		Various		*TBL1XR1*	[75, 83, 85, 110]
loss	4	q25	0.049	109.035	109.084	*LEF1*	[75, 85, 110]
loss	4	q31.23	0.145	149.697	149.842	*None; telomeric to NR3C2*	[75, 85, 110]
loss	5	q31.3	0.087	142.780	142.867	*NR3C1, LOC389335*	[75, 85]
loss	5	q33.3	0.553	157.946	158.499	*EBF1*	[75, 80, 83, 85, 110]
loss	6	p22.22	0.023	26.237	26.260	*Histone cluster, HIST1H4F, HIST1H4G, HIST1H3F, HIST1H2BH*	[75, 85, 110]
loss	6	q21	0.088	109.240	109.328	*ARMC2, SESN1*	[75, 85, 110]
loss	7	7p		Whole p-arm	Whole p-arm		[35, 85]
loss	7	q21.2	0.209	92.255	92.464	*LOC645862, GATAD1, ERVWE1, PEX1, DKFZP564O0523, LOC442710, MGC40405, CDK6*	[85, 110]
loss	7	p12.2	0.048	50.419	50.467	*IKZF1 (ZNFN1A1, Ikaros)* ,	[75, 83, 85]
loss	8	q12.1	0.094	60.032	60.126	*Immediately 5' (telomeric) of TOX*	[75, 85, 110]
loss	9	p21.3	0.237	21.894	22.131	*CDKN2A/CDKNA2B* , *MTAP* , *MLLT3 (AF9)*	[6, 35, 75, 80, 83, 85, 110]
loss	9	p13.2	0.088	36.932	37.020	*PAX5 CNA or sequence mutation*	[75, 80, 83, 85, 110]
loss	10	q23.31	0.062	89.676	89.738	*PTEN*	[75, 85, 110]
loss	10	q24.1	0.178	97.889	98.067	*BLNK*	[75, 85, 110]
loss	10	q25.1	0.078	111.782	111.860	*ADD3*	[75, 85, 110]
loss	11	p13	0.155	33.917	34.072	*No gene; immediately 5' of LMO2*	[75, 85]
loss	11	p12	0.008	36.618	36.626	*RAG1/2* , *LOC119710*	[75, 85, 110]
loss	11	q22.3	0.034	36.600	36.634	*ATM*	[80, 110]
loss	11	q23.3	0.274	118.369	118.643	*16 genes distal to MLL breakpoint, including 3' MLL*	[80, 85]
loss	12	p12.1	4.5	19.309	23.809	*KRAS*	[35, 110]

Loss/gain	Chromosome	Cytoband	Size (Mb)	Start position (Mb)*	End position (Mb)*	Candidate genes	Reference
loss	12	p13.2	0.086	11.813	11.899	*ETV6, KLRA-D family*	[6, 35, 75, 80, 83, 85, 110]
loss	12	q21.33	0.218	92.291	92.509	*BTG1*	[75, 80, 85, 110]
loss	13	q14.11	0.031	41.555	41.586	*ELF1 , C13orf21, LOC400128*	[75, 85, 110]
loss	13	q14.2	0.149	49.016	49.165	*RB1*	[6, 75, 80, 83, 85, 110]
loss	13	q14.2-3	0.889	50.573	51.462	*DLEU2, RFP2, KCNRG, MIRN16-1, MIRN15A, DLEU1, FAM10A4, LOC647154, LOC730194, DLEU7*	[75, 85]
loss	15	q12	0.038	26.036	26.074	*ATP10A*	[80, 110]
loss	15	q14	–	–	–	*SPRED1 (5')*	[75, 110]
loss	15	q15.1	0.792	41.258	42.050	*18 genes including LTK and MIRN626*	[85, 110]
loss	17	17p		Whole p-arm	Whole p-arm	*TP53*	[83]
loss	17	q11.2	0.169	29.066	29.235	*LOC729690, SUZ12P, CRLF3, LOC646013, C17orf41, C17orf42, [NF1]¶*	[75, 83, 85, 110]
loss	17	q21.1	0.045	37.931	37.976	*IKZF3 (ZNFN1A3, Aiolos)*	[75, 85, 110]
loss	19	p13.3	0.229	1.351	1.580	*63 genes telomeric to TCF3; region may include TCF3*	[75, 85, 110]
loss	20	20p12.1	0.035	10.422	10.457	*C20orf94*	[75, 85]
loss	20	q11.22	1.426	32.304	33.730	*Several genes , VPREB1*	[6, 110]
loss	21	q22.12	0.004	36.428	36.432	*No gene, but immediately distal to RUNX1*	[75, 85]
loss	21	q22.2	0.023	39.784	39.807	*ERG*	[75, 85, 110]
gain	1	q23.3-q44	81.326	164.759	qtel	*719 genes telomeric of PBX1, including 3' region of PBX1*	[75, 85]
Gain	6	q23.3	0.182	135.492	135.674	*MYB, MIRN548A2, AHI1*	[75, 80, 85]
Gain	9	9q		Whole q-arm	Whole q-arm	*ABL*	[83, 85]
Gain	9	q34.12-q34.3	7.676	133.657	qtel	*155 genes telomeric of ABL1, including 3' region of ABL1*	[75, 85, 110]
Gain	10	10p		Whole p-arm	Whole p-arm	–	[83, 85]
Gain	21	21	46.8	Whole chromosome	Whole chromosome	*Several genes*	[6, 83]
Gain	21	21q		Whole q-arm	Whole q-arm	*AML1, BACH, ERG*	[35, 83]
Ampl	21	iAMP21**	11.713	–	–	*Several genes*	[6]
Gain	21	q22.3	0.589	42.775	43.364	*7 genes*	[80]
Gain	21	q22.11-12	4.022	32.322	36.344	*34 genes*	[80]
Gain	21	q22.11-q22.12	2.303	33.974	36.277	*33 genes including RUNX1*	[75, 85]

Loss/gain	Chromosome	Cytoband	Size (Mb)	Start position (Mb)*	End position (Mb)*	Candidate genes	Reference
Gain	22	q11.1-q11.23	21.888	ptel	23.563	*277 genes telomeric (5') of BCR, including 5' region of BCR*	[75, 85]

* Assembly GRCh37/hg19 from Genome Reference Consortium

Table 1. Recurring regions of copy number alteration reported in ALL and involved genes with known or putative roles on leukemogenesis and cancer.

The average number of CNAs per ALL case is usually low, suggesting that this disease is not characterized by inherent genomic instability. This has been shown in a large SNP arrays study performed on pediatric ALL cases (B-progenitor and T-lineage). It allowed to identify a relatively low number of CNAs in ALL -a mean of 6.5 lesions per case- indicating that gross genomic instability is not a feature of most ALL cases [75, 85], although it is higher that the number of genomic changes in myeloid malignancies. Furthermore, similar studies have found 4.2 lesions per case in the precursor B-cell childhood ALLs (3.1 losses and 1.1 gains), and 2.6 lesions per case in the T-ALLs (1.7 losses and 0.9 gains) [80].

In spite of the large number of novel alterations, most of them have been focal deletions (less than a megabase) that involve only one or a few genes in the minimal region of genetic alteration. Apart from high hyperdiploid ALL, gains of DNA have been specifically uncommon and a few of them were focal gains [75, 85].

The pattern and number of CNAs is similar in the genetic ALL subtypes. Notably, less than one deletion per case was observed in *MLL*-rearranged ALL, typically presenting early in infancy. Therefore it has been suggested that a few additional genetic lesions are required for inducing leukemia. In contrast, other ALL subtypes such as *ETV6-RUNX1* and *BCR-ABL1*, typically presenting later than childhood, had over 6 copy number alterations per case, and some cases had over 20 lesions. These results are consistent with the concept that the initiating translocations are developed early in childhood, previous to clinically manifest leukemia (particularly for *ETV6-RUNX1* leukemia). Additional lesions are subsequently required for establishment of the leukemic clone. The deletion of *IKZF1* is also a lesion in *BCR-ABL1* ALL, but it is exceptionally uncommon in *ETV6-RUNX1* ALL [75, 76, 85-87].

High-resolution genomic profiling studies in childhood ALL also reveals recurrent genetic lesions, affecting genes with an established and critical role in leukemogenesis such as *CDKN2A*, *ETV6* (*TEL*), *RUNX1* (*AML1*) and other genes, such as *MLL*, that are used to stratify the patients [80]. Furthermore, many of these recurrent CNA were different between B-ALL and T-ALL subtypes. For example, deletions involving *ADD3*, *C20orf94*, *ERG*, *ETV6*, the fragile histidine triad gene *FHIT, TBL1XR1*, and a histone cluster at 6p22 were common in B-ALL but rare in T-ALL, whereas deletion of *CDKN2A/B* (9p21), are more frequent in T-ALL (72%-90%) than B-ALL (34%) [11, 76, 85].

4.2. CNA in T-ALL

Genome-wide profiling in T-ALL has been used to identify copy number alterations accompanying novel structural abnormalities, such as the *NUP214-ABL1* and *SET-NUP214* fusion genes. The amplification on extrachromosomal episomes of *ABL1* has been associated with the cryptic fusion of *NUP214* to *ABL1* gene, in around 6% of individuals with T-ALL. This fusion gene triggers the constitutive expression of the chimeric protein tyrosine kinase *NUP214-ABL1* and it is sensitive to the tyrosine kinase inhibitor imatinib. This amplification could improve outcome or decrease treatment-related morbidity of T-ALL cases, but large studies are needed to confirm these results [88]. Moreover, the cryptic and recurrent deletion, del(9)(q34.11q34.13), in pediatric T-ALL cases, results in a conserved *SET-NUP214* fusion product, that contribute to T-ALL pathogenesis by inhibition of T-cell maturation by the transcriptional activation of the *HOXA* genes [89].

Using SNP, BAC, or oligo-array CGH platforms, focal deletions have also identified in T-ALL, leading to deregulated expression of *TAL1* [85] and *LMO2* [90]; deletions of the *RB1* [85]; deletion and mutation of *PTEN* [85, 91]; deletion or mutation of the U3 ubiquitin ligase *FBXW7* [92]; and duplications of the protooncogene *MYB*, present in about 8% of T-ALL cases, that occur in combination with other genetic rearrangements contributing to T-cell differentiation arrest (*TAL/LMO, TLX1, TLX3, HOXA*) [68, 75, 93].

4.3. CNA in BL

High rates of CNAs have been reported in BL. CNAs have been observed in 65% [53] and 76% [43] of BL cases by conventional CGH. CNAs have been reported in 54% and 100% of BL patients by oaCGH and aSNP respectively [14, 55]. In addition, high-resolution molecular inversion probe (MIP) SNP assay have been reported 64% of CNAs in BL [94].

CGH and aCGH studies on cases of BL have shown that the increased number of gains and losses are significantly associated with shorter survival [43]. Gains are more frequent than losses in a range from 52% to 65% [14, 53, 94]. These studies have reported gains on chromosomes 1q, 7, 8q, 12, 13, 22 and Xq and losses in 6q, 13q, 14q, 17p, and Xp [14, 15, 43, 51, 53-55, 94, 95]. Some studies have also identified cases with gains on 2p [43, 55], 3q27.3 [14], 4p [43], 15q [51, 55], and 20p12-q13 [51].

It has been demonstrated that chromosomal gains or losses in the most frequently altered regions in BL, such as 1cen-q22, 1q31-q32, 7q22-qter, 8q24-qter, 13q31-q32, and 17p13-pter, influence changes in locus-specific gene expression levels of many genes that probably are associated with pathogenesis of BL. For example, the chromosomal region 1q showed increased gene expression levels in cases with gains, and correlates with the expression of germinal center-associated genes. By contrast, genetic losses in the chromosomal region 17p13 lead to a down regulation of genes located in this region, not only *TP53*, but also many other genes such as *AURKB,* that may influence the biological behavior as a consequence of deregulated expression [53].

4.4. CNA analysis of paired diagnostic and relapse ALL samples

Detailed comparative analysis of paired diagnostic and relapse ALL samples, using high resolution genomic profiling, have showed the next findings: i) frequent changes in DNA copy number abnormalities have been observed at relapse, ii) there are loss of copy number lesions present at diagnosis in ALL relapse samples, and acquisition of new additional (secondary) lesions in the relapse samples in nearly all analyzed patients, iii) deletions were more common than gains about newly acquired copy number abnormalities in the relapse samples. These data support the clonal evolution in ALL. The pattern of deletions on the antigen receptor loci was comparable between relapse and diagnosis, suggesting the emergence of a related leukemic clone, rather than the development of a distinct second leukemia. It should be noted that several cases were found in which the diagnosis and relapse samples carrying alternative lesions affecting the same gene(s), including *CDKN2A* and *PAX5*, suggesting that the inactivation of these genes were secondary but essential events required to develop a full-blown leukemia. Additionally, genomic abnormalities distinct from those presented at diagnosis has been identified lately, involved genes such as, *IKZF1, IKZF2, IKZF3, RAG, ADD3, ETV6, BTG1, DMD* and *IL3RA*/*CSF2RA,* suggesting that they confer a selective advantage and resistance to therapy in ALL [75, 96, 97].

These findings indicate that relapse is frequently the result of the emergence of a leukemic clone that shows significant genetic differences from the diagnostic clone. Whether these represent rare clones at the time of diagnosis or are the emergence of new clones should be further investigated [96].

5. Somatic mutations in acute lymphoblastic leukemia

Genome-wide profiling of DNA copy number alterations (CNA) coupled with focused candidate gene resequencing has identified novel genetic alterations in key signaling pathways in the pathogenesis of both B-progenitor and T-ALL. These findings are associated with leukemogenesis, treatment outcome in ALL, and are being exploited in the development of new therapeutic approaches and in the identification of markers of poor prognosis [72, 98].

5.1. Gene mutations in BCP-ALL

Somatic mutations in several genes are present in BCP-ALL. These mutations have identified in genes which are involved in RAS signaling (48%), B-cell differentiation and development (18%), JAK/STAT signaling (11%), TP53/RB1 tumor suppressor (6%) and noncanonical pathways and in other/unknown genes (17%) [72]. The incidence of the most recurrently mutated genes in ALL is described in the Table 2.

The frequency of alterations in the TP53/RB1, RAS, and JAK signaling pathways is much higher in High Risk B-Precursor Childhood Acute Lymphoblastic Leukemia (HR B-ALL)

BCP-ALL			
Pathway	**Gene**	**Frequency**	**Reference**
RAS signaling	*NRAS*	17%	[72]
	KRAS	16%	
	FLT3	7%	
	PTPN11	5%	
	NF1	3%	
B-cell differentiation and development pathway	*PAX5*	15%	
	IKZF1 (IKAROS)	3%	
JAK/STAT signaling	*JAK1*	2%	
	JAK2	9%	
TP53/RB1 pathway	*TP53*	4%	
	RB1	1%	
	CDKN2A/CDKN2B	1%	
Others	*TBL1XR1*	2%	
	ETV6	4%	
	CREBBP	2%	
	Unknown genes	9%	
T-ALL			
Pathway	**Gene**	**Frequency**	**Reference**
Cell cycle defects	*CDKN2A/CDKN2B*	96%	[18]
	TP53, RB, p27	4%	
Differentiation impairment	*TAL1 plus LMO1/2*	39%	
	LYL plus LMO2	20%	
	TLX1	7%	
	TLX3	20%	
	HOXA10/11	7%	
	PICALM-MLLT10	5-10%	
	MLL-fusions	4%	
	TAL2	<1%	
Proliferation and survival	*ABL1-fusions*	8%	
	NRAS	5%	
	FLT3	5%	
	LCK	<1%	
	ETV6-PBL2	<1%	
	ETV6-JAK2	<1%	
	PTEN	<1%	
	Unknown genes	"/>78%	
Self-renewal capacity	*NOTCH*	56%	
	Unknown genes	"/>44%	

Table 2. Frequency of the different mutations observed in ALL.

cohort than reported for unselected pediatric B-precursor ALL patients. In this subgroup of patients have been recently proposed new targeted therapeutics, such as the RAS/MAPK signaling pathway [98].

5.1.1. Ras signaling

Deregulation of the *RAS-RAF*-mitogen-activated protein kinase/extracellular signal-regulated kinase (*ERK*) kinase (*MEK*)-*ERK* signaling cascade is often caused by somatic mutations in genes encoding proteins that influence the activity of this pathway, such as *NRAS, KRAS2, FLT3, PTPN11,* and *BRAF* [99]. As observed in myeloid malignancies, up-regulated RAS signaling, due to mutations in RAS genes or in genes coding for proteins controlling *RAS* function, represent a major pathway driving the aberrant growth of malignant B-cell precursors [100].

In BCP-ALL, a number of associations with other genetic changes are already known, such as the link between mutations of genes within the RAS signaling pathway and high hyperdiploidy [79, 99, 101]. These mutations have been found in ~60% of high hyperdiploid childhood cases ALL. They are invariably mutually exclusive, and additional cooperative genetic events in this subgroup of patients [99, 101, 102].

5.1.1.1. NRAS and KRAS

RAS genes are part of the small GTPase family and consist of three separate genes, *NRAS, KRAS2*, and *HRAS. HRAS* is rarely mutated in hematologic tumors and is expressed at a low level compared to the other two isoforms in the hematopoietic cells in leukemia [102]. The *RAS* proteins activate several downstream pathways to promote proliferation, differentiation, survival, and apoptosis, depending on cellular conditions [102].

Mutations in *NRAS* and *KRAS* have been recognized as a recurring molecular event in childhood ALL, with a reported incidence of between 15% and 30% [98, 100, 102]. The incidence and spectrum of mutations at diagnosis and relapse are similar, although the presence is not a significant risk factor [99, 101]. Moreover, it has not been found any association of *RAS* mutation with an adverse clinical outcome [103]. The presence or number of mutations in the RAS signaling pathway have not been associated with relapse-free survival [98].

5.1.1.2. FLT3

Activating mutations in the receptor tyrosine kinase *FLT3* have been identified in approximately 20-25% of hyperdiploid and *MLL*-rearranged ALL samples [9, 104]. This observations supports the idea that the activation of tyrosine kinases as potential oncogenes in hyperdiploid ALL, as well as that leukemogenic fusion proteins such as MLL fusions cooperate with activated kinases to promote leukemogenesis [9]

Furthermore, small molecule tyrosine kinase inhibitors have activity against *MLL*-rearranged and hyperdiploid ALL with activating mutations in *FLT3*. Therefore *FLT3* inhibitors are

validated as a potential therapeutic target in this leukemia [9]. The presence of *FLT3* mutations in those patients with relapsed ALL harbored these alterations at diagnosis, suggested that *FLT3* inhibition could represent a therapeutic opportunity in at least a subset of patients with relapsed ALL [104].

5.1.1.3. PTPN11

PTPN11 encodes SHP2, a protein tyrosine phosphatase that positively controls *RAS* function. Somatic missense mutations in *PTPN11* cause SHP2 constitutive activation and enhance signaling through the mitogen-associated protein (MAP) kinase pathways [5].

PTPN11 mutations occur in approximately 6 to 7.3% of children with B-cell precursor ALL [5, 100]. Although *PTPN11* defects have been negatively associated with most of the gene rearrangements (*TEL-AML1, E2APBX1, BCR-ABL,* and *AF4-MLL*), and other gene lesions (*NRAS* and *KRAS2*), it has been observed higher prevalence of *PTPN11* mutations in children and adolescents with hyperdiploid DNA content [100].

PTPN11 mutations have been observed at disease presentation but are undetectable at remission, supporting the presence of the mutated gene in the leukemic clone and role of *PTPN11* lesions in leukemogenesis. Nevertheless, the prognostic significance of these mutations remains unknown [100].

5.1.1.4. BRAF

The *BRAF* gene, a member of RAF family, intermediates downstream in the *RAS/RAF/MAP* kinase pathway. This gene has been described mutated in most of hairy cell leukemias, but is less frequently mutated in acute leukemias, indicating that the *RAS-RAF* kinase pathway in some leukemias may be desregulated by somatic mutations of *BRAF* [105].

Mutations in *BRAF* have been reported with a frequency of 20% in B-cell ALLs cases [105, 106]. *BRAF* is expressed in hematopoietic cells, and the expression of activated *BRAF* could relieve the cytokine dependence and could result in the transformation of hematopoietic cells [105]. The functional significance of the most of the BRAF mutations is unknown, though all mutations are located within the kinase activation domain of *BRAF* [106]. Therapies that target *RAS-RAF-MEK-ERK-MAP* kinase pathway would be very valuable in treating tumors with activating mutations of BRAF [105].

5.1.2. B-cell differentiation and development pathway

5.1.2.1. PAX5

PAX5 (*paired box 5*) encodes a transcription factor which is known as B-cell specific activator protein. This protein plays a key role in B-cell commitment by activating essential components of B-cell receptor signaling and repressing the transcription of genes that are necessary for T-lymphopoiesis [107]. *PAX5* is the most common transcription factor which is altered in both children and adults B-ALL (32% of cases) [108]. Alterations of *PAX5,* including deletions, focal

amplifications, novel translocations, and sequence mutations, have not influence treatment outcome [107].

By SNP arrays, monoallelic deletion of *PAX5* has been observed in about 30% of children and adults with B-ALL, resulting in loss of PAX5 protein expression or in the production of a PAX5 isoform lacking the DNA binding domain and/or transcriptional regulatory domain [107, 109]. It has been demonstrated that the *PAX5* deletions are present in a dominant leukemic clone, consistent with a role in leukemogenesis during the establishing the leukemic clone [85, 110].

By sequencing, inactivating mutations of *PAX5* have been observed between 7–30 % of B-ALL cases [107]. These somatically acquired mutations have different patterns of alterations among some genetic subtypes of pediatric ALL [85]. The most point mutations of *PAX5* are hemizygous reducing or inhibiting normal *PAX5* functional activity [85].

Inactivating point mutations in *PAX5* have more effect on the intracellular transcriptional network within primary leukemic cells. These mutations are clustered in exons encoding the DNA-binding or transcriptional regulatory domains, which leads to lose or to alter DNA-binding or transcriptional regulatory function [85].

Chromosomal translocations *PAX5* are relatively rare, occurring in 2.5% of B-ALL cases; it has been reported at least 12 different fusion partners including transcription factors, structural proteins, and protein kinases (e.g. *ETV6, ENL, FOXP1, ZNF521, PML, C20ORF112, AUTS2, JAK2, POM121, HIPK1, DACH1, LOC392027, SLCO1B3, ASXL1*, and *KIF3B*) [107, 111].

In *PAX5* rearrangements, the DNA binding domain of *PAX5* and/or a variable amount of the C-terminal trans-activating domains are fused to functional domains of the partner genes, resulting in a loss of *PAX5* function rather than in a gain of functional elements [107, 110]. The fusion proteins may also influence the expression of genes which are normally regulated by the partner protein, each of which has been implicated in B-cell development or hematopoietic malignancies [85].

5.1.2.2. IKZF1 (IKAROS)

IKZF1 has been established as one of the most clinically relevant genes in pathogenesis of ALL, because it plays a key role in tumor suppression in pediatric B-cell ALL and in high-risk B-cell ALL [112]. Deletions or mutations of this gene have been described in 15% of all pediatric B-ALL. However the incidence in BCR-ABL ALL is higher (80%) and is associated with a poor outcome. In addition, recent genomic profiling studies (GEP) have produced strong evidence that *IKAROS* plays a key role in tumor suppression in pediatric B-cell ALL and in high-risk B-cell ALL. Thus the GEP of ALL cases with losses in IKZF1 is similar to the observed in BCR-ABL1 positive ALL [112]. Further studies, in larger series of patients, are needed to assess the clinical value of the deletion/mutations in *IKAROS* in the other subtypes of ALL.

5.1.3. JAK/STAT signaling

5.1.3.1. JAK

Activating mutations involving the pseudokinase and kinase domains of Janus kinases (primarily *JAK2*, but also *JAK1* and *JAK3*) have been reported in 10% of *BCR-ABL1*-negative high-risk pediatric ALL cases [86, 98, 113]. The childhood high-risk ALL cases, which harbor activating mutations *JAK*, have a gene-expression profile similar to *BCR-ABL1* pediatric ALL ("*BCR-ABL1*–like" -Ph-like), and are associated to a poor outcome [98].

These mutations are transforming in-vitro, and trigger constitutive *JAK-STAT* activation of the mouse Ba/F3 hematopoietic cell line expressing the erythropoietin receptor transduced with mutant *JAK* alleles [108]. This transformation is abrogated by pharmacologic *JAK1/2* inhibitors, suggesting that these agents may be a useful approach for treating patients harboring these mutations [108, 113].

The presence of *JAK* mutations have been associated with concomitant *IKZF1* and *CDKN2A/B* alterations, suggesting that genetic lesions target multiple cellular pathways, including lymphoid development (*IKZF1*), tumor suppression (*CDKN2A/B*), and activation of tyrosine kinase signaling (*BCR-ABL1, JAK*, or other kinase mutations) that cooperate to induce aggressive lymphoid leukemia in high-risk *BCR-ABL1*-ALL [113].

Particularly, gain-function mutations in *JAK2* are a common molecular event which is present about 18% of ALL Down's syndrome (DS-ALL) cases [114]. These findings suggest that *JAK2* inhibition might be a useful therapeutic approach in *JAK2*-mutated acute ALL associated with Down syndrome, because children with DS-ALL are especially sensitive to toxic effects of conventional chemotherapy [115].

5.1.3.2. Mutations in JAK regulators. CRLF2 and IL7R

CRLF2 encodes cytokine receptor–like factor 2 (also known as TSLPR-thymic stromal lymphopoietin receptor), a lymphoid signaling receptor molecule that forms a heterodimeric complex with interleukin-7 receptor alpha (*IL7R*) and binds TSLP [116]. *CRLF2*-mediated signaling promotes B lymphoid survival and proliferation [117].

Signaling from the TSLP receptor activates signal transducer and activator of transcription (*STAT5*) by phosphorylation of JAK1 and JAK2 through association with *IL-7R* and *CRLF2*, respectively [118]. Genetic alterations dysregulating *CRLF2* expression may contribute to the pathogenesis of ALL [117], by induced activation of STAT proteins, especially STAT5 and STAT1 [119].

CRLF2 rearrangements, such as *IGH@-CRLF2* or *P2RY8-CRLF2* fusion, are present in up to 60% of children with Down Syndrome ALL (DS-ALL) and about 10–15% of high-risk *BCR-ABL1* negative childhood and adult ALL [22]. In both DS-ALL and non-DS-ALL, approximately half of *CRLF2* rearranged cases have concomitant activating *JAK* mutations (the most common in *JAK2* but occasionally in *JAK1*), suggesting that the two alterations cooperate downstream in the signal transduction and transformation [108, 116].

Furthermore, in high-risk ALL, *IKZF1* alterations, *CRLF2* rearrangement and *JAK* mutations are frequently observed together. They are associated with very poor outcome, even with current maximal intensive therapy [108, 117]. These leukemias may be sensitive to *JAK* inhibitors, suggesting the potential for a targeted therapy. Thereby, detection of *IKZF1, CRLF2,* and *JAK* mutations should be considered at diagnosis in childhood ALL [117].

Moreover, somatic mutations of Interleukin-7 receptor (*IL7R*) (the heterodimeric partner of *CRLF2*) have been reported in pediatric B and T ALL. Some *IL7R* mutations have been observed in both diagnosis and relapse, but other mutations have been only present in relapse, whereas *CRLF2* expression have been already described at diagnosis, suggesting that the *IL7R* mutation may be a progression event [120]. Mutations of *IL7R* are gain-of-function mutations that cooperate with *CRLF2* to form a constitutively activated TSLP receptor. *IL7R* activating mutations trigger cytokine-independent growth of progenitor lymphoid cells, and constitutive activation of STAT and mTOR pathways [120].

5.1.4. TP53/RB1 pathway

Mutations of the tumor suppressor gene *TP53* have been associated with resistance to treatment and worse prognosis of patients in several tumors. Alterations of the *TP53* gene are important at relapse in childhood ALL, in which they independently predict high risk of treatment failure in a significant number of patients[121].

The presence of *TP53* mutations is associated with a reduced response rate to reinduction therapy. In addition, *TP53* mutations correlate with a shortened duration of survival (from time of relapse and from time of diagnosis), even after successful reinduction therapy [122].

The clinical significance of exclusive deletions might be explained by *TP53* haploinsufficiency. Moreover, an additional mutation appeared during or after relapse therapy in some relapse patients with exclusive deletion and nonresponse to treatment or second relapse, indicating outgrowth of fully *TP53* altered clones that might contribute to the poor outcome [121].

5.2. Gene mutations in T-ALL

T-ALL has been associated with four different classes of mutations: (i) Affecting the cell cycle (*CDKN2A*/*CDKN2B*); (ii) Impairing differentiation (*HOX* genes, *MLL, LYL1, TAL1/2* and *LMO1/2*); (iii) Providing a proliferative and survival advantage (*LCK* and *ABL1*); (iv) Providing self-renewal capacity (*NOTCH1*) [10, 11, 18, 123]. The genes most recurrently mutated in T-ALL are described in Table 2.

5.2.1. CDKN2A/CDKN2B

In up to 90% of ALL cases, the *CDKN2A/2B* genes, located in tandem at chromosome 9p21, are inactivated by cryptic deletions, promoter hypermethylation, inactivating mutations or (post)-transcriptional modifications. Homozygous or heterozygous inactivation of the genomic *CDKN2A* and *CDKN2B* loci are the most frequent genetic abnormalities in T-ALL [124].

Inactivation of *CDKN2A* and *CDKN2B* by homozygous deletion has been described in 65% and 23% of T-ALL samples, respectively. Hemizygous *CDKN2A* and *CDKN2B* deletions are observed in approximately 10% and 15% of the samples [18].

The haploinsufficiency or inactivation of these tumor suppressor genes are involved in the development of T-ALL, because they not only promote uncontrolled cell cycle entry, but also disable the p53-controlled cell cycle checkpoint and apoptosis machinery. Thus, *RB1* and *TP53* pathways have been identified as possible targets for therapy of T-ALL [10, 11, 18, 19, 123].

5.2.2. Tp53

The acquisition of mutations in *TP53* has been described in T-cell lines and T-ALL patients [123, 125]. The *TP53* mutations are infrequent at diagnosis (5% of T-ALL cases) and tend to be associated with poor clinical outcome [123]. Copy number and sequence alterations of *TP53* have been observed in 6.4%-24% of patients with T-cell ALL relapse, suggesting the importance of these alterations in the progression of the disease, in which they independently predict high risk of treatment failure in a significant number of patients [121, 123].

5.2.3. NOTCH1

Gain-of-function mutations in *NOTCH1* have been identified in more than 50% of T-ALL samples resulting in constitutive NOTCH signaling [126]. They have been associated with a favorable early treatment response [11, 127]. *NOTCH1* is a transmembrane receptor that plays a role in normal hematopoiesis as an early transcription factor and regulates self-renewal of stem cells and lineage commitment of lymphoid progenitor cells towards T-cell development [11, 128]. The intracellular NOTCH (ICN) released after proteolytic cleavage step of *NOTCH1* mediates in the nucleus the expression of various target genes including *HES1, HEY1, MYC, PTCRA, DTX1* and members of the NFkB pathway. At the protein level, activation of *NOTCH1* mutations could also cause phosphorylation of multiple signaling proteins in the mTOR pathway [11]. *NOTCH1* receptor is a promising target for drugs such as gamma-secretase inhibitors which block a proteolytic cleavage required for *NOTCH1* activation signaling pathway [91].

The presence of subclonal duplications of the chromosomal region 9q34 are present in about 33% of pediatric T-ALL patients; the critical region encloses many genes including *NOTCH1*. Although this duplication appears as an independent genetic event from both the episomal *NUP214-ABL1* amplification and the *NOTCH1* mutations, it could induce subtle changes in *NOTCH1* expression levels and contribute to global *NOTCH1* activation in T-ALL [11, 129].

5.2.4. FBXW7

F-box protein FBXW7 is an E3-ubiquitin ligase that regulates the half-life of other proteins including CyclinE, cMYC and cJUN [11]. Heterozygous *FBXW7* single mutations have been identified in 8–30% of T-ALL patients, and usually are combined with *NOTCH1* mutations affecting the heterodimerization (HD) domain. *FBXW7* mutations render *FBXW7* inactive to prime target proteins like *NOTCH1* for proteosomal degradation, therefore these mutations

represent an alternative mechanism for *NOTCH1* activation in T-ALL [11]. The presence of both *FBXW7* and *NOTCH1* mutations has been associated with good treatment response in T-ALL patients [130].

5.2.5. JAK1

Somatic activating *JAK1* mutations occur about 10-20% of adults with T-cell precursors ALL, and have low prevalence in children and adolescents T-ALL [131]. *JAK1* gene defects are associated with a poor response to therapy, frequent relapse, and reduced overall survival, identifying such mutations as a novel informative prognostic marker in adult T-ALL [132].

JAK1 gene encodes a cytoplasmic tyrosine kinase that it is noncovalently associates with a variety of cytokine receptors and plays a nonredundant role in lymphoid cell precursor proliferation, survival, and differentiation. T-cell origin with mutated *JAK1* share a gene expression signature which is characterized by transcriptional up-regulation of genes positively controlled by JAK signaling [132].

Gain-of-function mutations in *JAK1* may be concomitant with other genomic changes, such as *NOTCH1* defects. The activation of *JAK1* and *NOTCH1* transduction pathways might cooperate in T-ALL pathogenesis and/or progression [10, 19, 132].

5.2.6. PTEN

PTEN loss of function mutation and deletions occur in approximately 25% to 35% of cases of T-cell ALL [98]. *PTEN* mutations and loss of PTEN protein could be also found as a secondary event during disease progression, thereby it could represent a progression marker rather than an initiating event in T-ALL [11].

The PTEN phosphatase has been identified as an important regulator of downstream (pre)TCR signaling. It directly opposes the activity of the phosphor-inosital-3 kinase (PI3K) functioning as a negative regulator of the oncogenic PI3K-AKT signaling [11, 133]. Inactivation of *PTEN* has been associated with activation of the PI3K-AKT pathway resulting in enhanced cell size, glucose uptake and proliferation [91]. Furthermore, the detection of abnormalities in the *PTEN*, *PI3K*, and *AKT* genes in a large subset of primary T-ALL samples have demonstrated a prominent role for oncogenic PI3K-AKT signaling in the pathogenesis of T-ALL [133].

Independent from activation following (pre)TCR stimulation, *PTEN* is negatively regulated by *NOTCH1* [11]. There are some small molecule inhibitors of γ-secretase (GSIs) which block *NOTCH1* activation in T-ALL cell lines with prototypical activating mutations in *NOTCH1*, However some of them are GSI-resistant. This resistance to GSI action is mediated by molecular abnormalities in signaling pathways that promote cell growth downstream of *NOTCH1* [91]. It has been reported that mutational loss of *PTEN* is associated with human T-ALL resistance to pharmacological inhibition of *NOTCH1* performed by GSIs [91]. Therefore *PTEN* deletions appeared to impart a high risk of induction failure with contemporary chemotherapy in T-ALL patients [91]

5.2.7. RAS

In T-ALL, activating *RAS* mutations have been identified only in 4–10% of cases without a prognostic impact [98, 123, 134, 135]. Nevertheless, it has been identified an alternative *RAS* activation mechanism in T-ALL cases with *NF1* microdeletions on chromosome 17 without clinical evidence for neurofibromatosis with mutations on the remaining *NF1* allele. *NF1* is a negative regulator of the RAS signaling pathway. The presence of mutations on the remaining *NF1* allele, confirmed the potential *NF1* inactivation as an alternative *RAS* activation mechanism in these T-ALL cases. Therefore, T-ALL patients with activated *RAS* could potentially benefit from additional treatment with *RAS* inhibitors, such as farnesylthiosalicylic acid [11].

5.2.8. WT1

WT1 mutations is a recurrent genetic alteration in T-ALL. They are present in around 10% of T-ALL both in childhood and adults [136]. These mutations are highly associated with direct or indirect aberrant *HOX* genes expression in T-ALL cases with aberrant rearrangements of the oncogenic *TLX1*, *TLX3*, and *HOXA* transcription factor oncogenes [137]. Survival analysis have demonstrated that *WT1* mutations do not confer adverse prognosis in either pediatric and adult T-ALL cases [136].

5.2.9. Mutated genes in Early Thymic Progenitors (ETP)-ALL

A new T-ALL subgroup, which is defined by a specific gene expression profile and a characteristic immunophenotype (CD1a-, CD8-, CD5weak with expression of stem cell or myeloid markers), has been recently described in pediatric T-ALL patients with poor outcome. This subgroup likely originates from early thymic progenitors (ETP) and has been called ETP-ALL. Recently, it has been described the high presence of *FLT3* mutations in ETP-ALL [138] while in T-ALL patients with a non-ETP immunophenotype are rare (1-3%). In some patients, these mutations are only present in leukemic subclones [139, 140],, indicating that FLT3 mutations may represent a T-ALL progression marker rather than an initiating event [11].

Moreover a recent study of whole-genome sequencing in ETP-ALL cases, has identified activating mutations in genes regulating cytokine receptor and RAS signaling in 67% of cases (*NRAS, KRAS, FLT3, IL7R, JAK3, JAK1, SH2B3 and BRAF*), inactivating lesions disrupting hematopoietic development involving 58% of patients (*GATA3, ETV6, RUNX1, IKZF1* and *EP300*) and histone-modifying genes in 48% of patients (*EZH2, EED, SUZ12, SETD2* and *EP300*) [141]. The global transcriptional profile of ETP ALL was similar to normal and myeloid leukemia hematopoietic stem cells. These findings could be related to the prognosis of ETP ALL patients [141].

In summary, the recent development of the genome wide analysis has provided new and critical knowledge of genetic changes in ALL. These new chromosomal imbalances and mutations could provide new insights for the management of the disease that is still associated with a dismal prognosis in the adult patients.

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Author details

Ruth Maribel Forero[1,2], María Hernández[1] and Jesús María Hernández-Rivas[1,3*]

*Address all correspondence to: jmhr@usal.es

1 IBSAL,IBMCC, Centro de Investigación del Cáncer, Universidad de Salamanca-CSIC, Spain

2 Universidad Pedagógica Y Tecnológica de Colombia, Colombia

3 Servicio de Hematología, Hospital Clínico Universitario de Salamanca, Spain

References

[1] Teitell, M.A. and P.P. Pandolfi, *Molecular genetics of acute lymphoblastic leukemia.* Annu Rev Pathol, 2009. 4: p. 175-98.

[2] Bacher, U., A. Kohlmann, and T. Haferlach, *Gene expression profiling for diagnosis and therapy in acute leukaemia and other haematologic malignancies.* Cancer Treat Rev, 2010. 36(8): p. 637-46.

[3] Downing, J.R. and C.G. Mullighan, *Tumor-specific genetic lesions and their influence on therapy in pediatric acute lymphoblastic leukemia.* Hematology Am Soc Hematol Educ Program, 2006: p. 118-22, 508.

[4] Jabbour, E.J., S. Faderl, and H.M. Kantarjian, *Adult acute lymphoblastic leukemia.* Mayo Clin Proc, 2005. 80(11): p. 1517-27.

[5] Pui, C.H., M.V. Relling, and J.R. Downing, *Acute lymphoblastic leukemia.* N Engl J Med, 2004. 350(15): p. 1535-48.

[6] Bungaro, S., et al., *Integration of genomic and gene expression data of childhood ALL without known aberrations identifies subgroups with specific genetic hallmarks.* Genes Chromosomes Cancer, 2009. 48(1): p. 22-38.

[7] Usvasalo, A., et al., *Acute lymphoblastic leukemias with normal karyotypes are not without genomic aberrations.* Cancer Genet Cytogenet, 2009. 192(1): p. 10-7.

[8] Rowley, J.D., *The critical role of chromosome translocations in human leukemias.* Annu Rev Genet, 1998. 32: p. 495-519.

[9] Armstrong, S.A. and A.T. Look, *Molecular genetics of acute lymphoblastic leukemia.* J Clin Oncol, 2005. 23(26): p. 6306-15.

[10] Gorello, P., et al., *Combined interphase fluorescence in situ hybridization elucidates the genetic heterogeneity of T-cell acute lymphoblastic leukemia in adults.* Haematologica, 2010. 95(1): p. 79-86.

[11] Van Vlierberghe, P., et al., *Molecular-genetic insights in paediatric T-cell acute lymphoblastic leukaemia.* Br J Haematol, 2008. 143(2): p. 153-68.

[12] Swerdlow, S.H., et al., *WHO classification of tumours of haematopoietic and lymphoid tissues*. 4ª ed. 2008, IARC Lyon: World Health Organization.

[13] Miles, R.R., S. Arnold, and M.S. Cairo, *Risk factors and treatment of childhood and adolescent Burkitt lymphoma/leukaemia.* Br J Haematol, 2012. 156(6): p. 730-43.

[14] Scholtysik, R., et al., *Detection of genomic aberrations in molecularly defined Burkitt's lymphoma by array-based, high resolution, single nucleotide polymorphism analysis.* Haematologica, 2010. 95(12): p. 2047-55.

[15] Boerma, E.G., et al., *Translocations involving 8q24 in Burkitt lymphoma and other malignant lymphomas: a historical review of cytogenetics in the light of todays knowledge.* Leukemia, 2009. 23(2): p. 225-34.

[16] Chiaretti, S., et al., *Gene expression profile of adult T-cell acute lymphocytic leukemia identifies distinct subsets of patients with different response to therapy and survival.* Blood, 2004. 103(7): p. 2771-8.

[17] Cauwelier, B., et al., *Molecular cytogenetic study of 126 unselected T-ALL cases reveals high incidence of TCRbeta locus rearrangements and putative new T-cell oncogenes.* Leukemia, 2006. 20(7): p. 1238-44.

[18] De Keersmaecker, K., P. Marynen, and J. Cools, *Genetic insights in the pathogenesis of T-cell acute lymphoblastic leukemia.* Haematologica, 2005. 90(8): p. 1116-27.

[19] Graux, C., et al., *Cytogenetics and molecular genetics of T-cell acute lymphoblastic leukemia: from thymocyte to lymphoblast.* Leukemia, 2006. 20(9): p. 1496-510.

[20] Usvasalo, A., et al., *Prognostic classification of patients with acute lymphoblastic leukemia by using gene copy number profiles identified from array-based comparative genomic hybridization data.* Leuk Res, 2010. 34(11): p. 1476-82.

[21] Pui, C.H., et al., *Biology, risk stratification, and therapy of pediatric acute leukemias: an update.* J Clin Oncol, 2011. 29(5): p. 551-65.

[22] Izraeli, S., *Application of genomics for risk stratification of childhood acute lymphoblastic leukaemia: from bench to bedside?* Br J Haematol, 2010. 151(2): p. 119-31.

[23] Sancho, J.M.C., *Avances en el diagnóstico y tratamiento, y significado pronóstico de la infiltración neuromeníngea en leucemias agudas y linfomas agresivos*, in *Facultad de Medicina.* 2011, Universidad Autónoma de Barcelona. : Barcelona. p. 36.

[24] Heim, S. and F. Mitelman, *Cancer cytogenetics*. Third ed. Acute Lymphoblastic Leukemia. 2009, New Jersey: John Wiley & Sons, Inc. Hoboken.

[25] Pui, C.H., D. Campana, and W.E. Evans, *Childhood acute lymphoblastic leukaemia--current status and future perspectives.* Lancet Oncol, 2001. 2(10): p. 597-607.

[26] Ribera, J.J.O.y.J.M., *Leucemia Aguda Linfoblástica*. 16 ed. Farreras-Rozman, Medicina Interna., ed. F.C.e. En C Rozman. 2009, Barcelona: Elsevier.

[27] Mittelman, F., *The Third International Workshop on Chromosomes in Leukemia. Lund, Sweden, July 21-25, 1980. Introduction.* Cancer genetics and cytogenetics, 1981. 4(2): p. 96-98.

[28] Kuchinskaya, E., et al., *Array-CGH reveals hidden gene dose changes in children with acute lymphoblastic leukaemia and a normal or failed karyotype by G-banding.* Br J Haematol, 2008. 140(5): p. 572-7.

[29] Kim, J.E., et al., *A rare case of acute lymphoblastic leukemia with t(12;17)(p13;q21).* Korean J Lab Med, 2010. 30(3): p. 239-43.

[30] Harrison, C.J. and L. Foroni, *Cytogenetics and molecular genetics of acute lymphoblastic leukemia.* Rev Clin Exp Hematol, 2002. 6(2): p. 91-113; discussion 200-2.

[31] Harrison, C.J., et al., *Interphase molecular cytogenetic screening for chromosomal abnormalities of prognostic significance in childhood acute lymphoblastic leukaemia: a UK Cancer Cytogenetics Group Study.* Br J Haematol, 2005. 129(4): p. 520-30.

[32] De Braekeleer, E., et al., *Cytogenetics in pre-B and B-cell acute lymphoblastic leukemia: a study of 208 patients diagnosed between 1981 and 2008.* Cancer Genet Cytogenet, 2010. 200(1): p. 8-15.

[33] Graux, C., *Biology of acute lymphoblastic leukemia (ALL): clinical and therapeutic relevance.* Transfus Apher Sci, 2011. 44(2): p. 183-9.

[34] Downing, J.R. and K.M. Shannon, *Acute leukemia: a pediatric perspective.* Cancer Cell, 2002. 2(6): p. 437-45.

[35] Steinemann, D., et al., *Copy number alterations in childhood acute lymphoblastic leukemia and their association with minimal residual disease.* Genes Chromosomes Cancer, 2008. 47(6): p. 471-80.

[36] Schultz, K.R., et al., *Improved early event-free survival with imatinib in Philadelphia chromosome-positive acute lymphoblastic leukemia: a children's oncology group study.* J Clin Oncol, 2009. 27(31): p. 5175-81.

[37] Meyer, C., et al., *New insights to the MLL recombinome of acute leukemias.* Leukemia, 2009. 23(8): p. 1490-9.

[38] Meyer, C., et al., *The MLL recombinome of acute leukemias.* Leukemia, 2006. 20(5): p. 777-84.

[39] Forestier, E., et al., *Outcome of ETV6/RUNX1-positive childhood acute lymphoblastic leukaemia in the NOPHO-ALL-1992 protocol: frequent late relapses but good overall survival.* Br J Haematol, 2008. 140(6): p. 665-72.

[40] Golub, T.R., et al., *Fusion of the TEL gene on 12p13 to the AML1 gene on 21q22 in acute lymphoblastic leukemia.* Proc Natl Acad Sci U S A, 1995. 92(11): p. 4917-21.

[41] Frick, M., B. Dorken, and G. Lenz, *New insights into the biology of molecular subtypes of diffuse large B-cell lymphoma and Burkitt lymphoma.* Best Pract Res Clin Haematol, 2012. 25(1): p. 3-12.

[42] Hecht, J.L. and J.C. Aster, *Molecular biology of Burkitt's lymphoma.* J Clin Oncol, 2000. 18(21): p. 3707-21.

[43] Garcia, J.L., et al., *Abnormalities on 1q and 7q are associated with poor outcome in sporadic Burkitt's lymphoma. A cytogenetic and comparative genomic hybridization study.* Leukemia, 2003. 17(10): p. 2016-24.

[44] Lones, M.A., et al., *Chromosome abnormalities may correlate with prognosis in Burkitt/Burkitt-like lymphomas of children and adolescents: a report from Children's Cancer Group Study CCG-E08.* J Pediatr Hematol Oncol, 2004. 26(3): p. 169-78.

[45] Poirel, H.A., et al., *Specific cytogenetic abnormalities are associated with a significantly inferior outcome in children and adolescents with mature B-cell non-Hodgkin's lymphoma: results of the FAB/LMB 96 international study.* Leukemia, 2009. 23(2): p. 323-31.

[46] Onciu, M., et al., *Secondary chromosomal abnormalities predict outcome in pediatric and adult high-stage Burkitt lymphoma.* Cancer, 2006. 107(5): p. 1084-92.

[47] Xiao, H., et al., *American Burkitt lymphoma stage II with 47,XY,+20,t(8;14)(q24;q32).* Cancer Genet Cytogenet, 1990. 48(2): p. 275-7.

[48] Lai, J.L., et al., *Cytogenetic studies in 30 patients with Burkitt's lymphoma or L3 acute lymphoblastic leukemia with special reference to additional chromosome abnormalities.* Ann Genet, 1989. 32(1): p. 26-32.

[49] Nelson, M., et al., *An increased frequency of 13q deletions detected by fluorescence in situ hybridization and its impact on survival in children and adolescents with Burkitt lymphoma: results from the Children's Oncology Group study CCG-5961.* Br J Haematol, 2010. 148(4): p. 600-10.

[50] de Souza, M.T., et al., *Secondary abnormalities involving 1q or 13q and poor outcome in high stage Burkitt leukemia/lymphoma cases with 8q24 rearrangement at diagnosis.* Int J Hematol, 2011. 93(2): p. 232-6.

[51] Zunino, A., et al., *Chromosomal aberrations evaluated by CGH, FISH and GTG-banding in a case of AIDS-related Burkitt's lymphoma.* Haematologica, 2000. 85(3): p. 250-5.

[52] Zimonjic, D.B., C. Keck-Waggoner, and N.C. Popescu, *Novel genomic imbalances and chromosome translocations involving c-myc gene in Burkitt's lymphoma.* Leukemia, 2001. 15(10): p. 1582-8.

[53] Salaverria, I., et al., *Chromosomal alterations detected by comparative genomic hybridization in subgroups of gene expression-defined Burkitt's lymphoma.* Haematologica, 2008. 93(9): p. 1327-34.

[54] Barth, T.F., et al., *Homogeneous immunophenotype and paucity of secondary genomic aberrations are distinctive features of endemic but not of sporadic Burkitt's lymphoma and diffuse large B-cell lymphoma with MYC rearrangement.* J Pathol, 2004. 203(4): p. 940-5.

[55] Toujani, S., et al., *High resolution genome-wide analysis of chromosomal alterations in Burkitt's lymphoma.* PLoS One, 2009. 4(9): p. e7089.

[56] Berger, R. and A. Bernheim, *Cytogenetics of Burkitt's lymphoma-leukaemia: a review.* IARC Sci Publ, 1985(60): p. 65-80.

[57] Cave, H., et al., *Clinical significance of HOX11L2 expression linked to t(5;14)(q35;q32), of HOX11 expression, and of SIL-TAL fusion in childhood T-cell malignancies: results of EORTC studies 58881 and 58951.* Blood, 2004. 103(2): p. 442-50.

[58] Ballerini, P., et al., *HOX11L2 expression defines a clinical subtype of pediatric T-ALL associated with poor prognosis.* Blood, 2002. 100(3): p. 991-7.

[59] Hawley, R.G., et al., *Transforming function of the HOX11/TCL3 homeobox gene.* Cancer Res, 1997. 57(2): p. 337-45.

[60] Keller, G., et al., *Overexpression of HOX11 leads to the immortalization of embryonic precursors with both primitive and definitive hematopoietic potential.* Blood, 1998. 92(3): p. 877-87.

[61] Bernard, O.A., et al., *A new recurrent and specific cryptic translocation, t(5;14)(q35;q32), is associated with expression of the Hox11L2 gene in T acute lymphoblastic leukemia.* Leukemia, 2001. 15(10): p. 1495-504.

[62] Wadman, I., et al., *Specific in vivo association between the bHLH and LIM proteins implicated in human T cell leukemia.* EMBO J, 1994. 13(20): p. 4831-9.

[63] Xia, Y., et al., *TAL2, a helix-loop-helix gene activated by the (7;9)(q34;q32) translocation in human T-cell leukemia.* Proc Natl Acad Sci U S A, 1991. 88(24): p. 11416-20.

[64] Xia, Y., et al., *Products of the TAL2 oncogene in leukemic T cells: bHLH phosphoproteins with DNA-binding activity.* Oncogene, 1994. 9(5): p. 1437-46.

[65] Zhong, Y., et al., *Overexpression of a transcription factor LYL1 induces T- and B-cell lymphoma in mice.* Oncogene, 2007. 26(48): p. 6937-47.

[66] Harrison, C.J., *Cytogenetics of paediatric and adolescent acute lymphoblastic leukaemia.* Br J Haematol, 2009. 144(2): p. 147-56.

[67] Palacios, E.H. and A. Weiss, *Function of the Src-family kinases, Lck and Fyn, in T-cell development and activation.* Oncogene, 2004. 23(48): p. 7990-8000.

[68] Clappier, E., et al., *The C-MYB locus is involved in chromosomal translocation and genomic duplications in human T-cell acute leukemia (T-ALL), the translocation defining a new T-ALL subtype in very young children.* Blood, 2007. 110(4): p. 1251-61.

[69] Dik, W.A., et al., *CALM-AF10+ T-ALL expression profiles are characterized by overexpression of HOXA and BMI1 oncogenes.* Leukemia, 2005. 19(11): p. 1948-57.

[70] Krivtsov, A.V. and S.A. Armstrong, *MLL translocations, histone modifications and leukaemia stem-cell development.* Nat Rev Cancer, 2007. 7(11): p. 823-33.

[71] Lacronique, V., et al., *A TEL-JAK2 fusion protein with constitutive kinase activity in human leukemia.* Science, 1997. 278(5341): p. 1309-12.

[72] Harrison, C.J., *Key pathways as therapeutic targets.* Blood, 2011. 118(11): p. 2935-6.

[73] Harrison, C.J., et al., *Three distinct subgroups of hypodiploidy in acute lymphoblastic leukaemia.* Br J Haematol, 2004. 125(5): p. 552-9.

[74] Macconaill, L.E. and L.A. Garraway, *Clinical implications of the cancer genome.* J Clin Oncol, 2010. 28(35): p. 5219-28.

[75] Mullighan, C.G. and J.R. Downing, *Global genomic characterization of acute lymphoblastic leukemia.* Semin Hematol, 2009. 46(1): p. 3-15.

[76] Mullighan, C.G., *Genomic analysis of acute leukemia.* Int J Lab Hematol, 2009. 31(4): p. 384-97.

[77] Rabin, K.R., et al., *Clinical utility of array comparative genomic hybridization for detection of chromosomal abnormalities in pediatric acute lymphoblastic leukemia.* Pediatr Blood Cancer, 2008. 51(2): p. 171-7.

[78] Yasar, D., et al., *Array comparative genomic hybridization analysis of adult acute leukemia patients.* Cancer Genet Cytogenet, 2010. 197(2): p. 122-9.

[79] Harrison, C., *New genetics and diagnosis of childhood B-cell precursor acute lymphoblastic leukemia.* Pediatr Rep, 2011. 3 Suppl 2: p. e4.

[80] Kuiper, R.P., et al., *High-resolution genomic profiling of childhood ALL reveals novel recurrent genetic lesions affecting pathways involved in lymphocyte differentiation and cell cycle progression.* Leukemia, 2007. 21(6): p. 1258-66.

[81] Strefford, J.C., et al., *Genome complexity in acute lymphoblastic leukemia is revealed by array-based comparative genomic hybridization.* Oncogene, 2007. 26(29): p. 4306-18.

[82] Paulsson, K., et al., *Microdeletions are a general feature of adult and adolescent acute lymphoblastic leukemia: Unexpected similarities with pediatric disease.* Proc Natl Acad Sci U S A, 2008. 105(18): p. 6708-13.

[83] Okamoto, R., et al., *Genomic profiling of adult acute lymphoblastic leukemia by single nucleotide polymorphism oligonucleotide microarray and comparison to pediatric acute lymphoblastic leukemia.* Haematologica, 2010. 95(9): p. 1481-8.

[84] Dawson, A.J., et al., *Array comparative genomic hybridization and cytogenetic analysis in pediatric acute leukemias.* Curr Oncol, 2011. 18(5): p. e210-7.

[85] Mullighan, C.G., et al., *Genome-wide analysis of genetic alterations in acute lymphoblastic leukaemia.* Nature, 2007. 446(7137): p. 758-64.

[86] Mullighan, C.G., *Genomic profiling of B-progenitor acute lymphoblastic leukemia.* Best Pract Res Clin Haematol, 2011. 24(4): p. 489-503.

[87] Greaves, M.F. and J. Wiemels, *Origins of chromosome translocations in childhood leukaemia.* Nat Rev Cancer, 2003. 3(9): p. 639-49.

[88] Graux, C., et al., *Fusion of NUP214 to ABL1 on amplified episomes in T-cell acute lymphoblastic leukemia.* Nat Genet, 2004. 36(10): p. 1084-9.

[89] Van Vlierberghe, P., et al., *The recurrent SET-NUP214 fusion as a new HOXA activation mechanism in pediatric T-cell acute lymphoblastic leukemia.* Blood, 2008. 111(9): p. 4668-80.

[90] Van Vlierberghe, P., et al., *The cryptic chromosomal deletion del(11)(p12p13) as a new activation mechanism of LMO2 in pediatric T-cell acute lymphoblastic leukemia.* Blood, 2006. 108(10): p. 3520-9.

[91] Palomero, T., et al., *Mutational loss of PTEN induces resistance to NOTCH1 inhibition in T-cell leukemia.* Nat Med, 2007. 13(10): p. 1203-10.

[92] O'Neil, J., et al., *FBW7 mutations in leukemic cells mediate NOTCH pathway activation and resistance to gamma-secretase inhibitors.* J Exp Med, 2007. 204(8): p. 1813-24.

[93] Lahortiga, I., et al., *Duplication of the MYB oncogene in T cell acute lymphoblastic leukemia.* Nat Genet, 2007. 39(5): p. 593-5.

[94] Schiffman, J.D., et al., *Genome wide copy number analysis of paediatric Burkitt lymphoma using formalin-fixed tissues reveals a subset with gain of chromosome 13q and corresponding miRNA over expression.* Br J Haematol, 2011. 155(4): p. 477-86.

[95] Capello, D., et al., *Genome wide DNA-profiling of HIV-related B-cell lymphomas.* Br J Haematol, 2010. 148(2): p. 245-55.

[96] Mullighan, C.G., et al., *High-Resolution SNP Array Profiling of Relapsed Acute Leukemia Identifies Genomic Abnormalities Distinct from Those Present at Diagnosis.* ASH Annual Meeting Abstracts, 2007. 110(11): p. 234-.

[97] Kuiper, R.P., et al., *Detection of Genomic Lesions in Childhood Precursor-B Cell ALL in Diagnosis and Relapse Samples Using High Resolution Genomic Profiling.* ASH Annual Meeting Abstracts, 2007. 110(11): p. 995-.

[98] Zhang, J., et al., *Key pathways are frequently mutated in high-risk childhood acute lymphoblastic leukemia: a report from the Children's Oncology Group.* Blood, 2011. 118(11): p. 3080-7.

[99] Case, M., et al., *Mutation of genes affecting the RAS pathway is common in childhood acute lymphoblastic leukemia.* Cancer Res, 2008. 68(16): p. 6803-9.

[100] Tartaglia, M., et al., *Genetic evidence for lineage-related and differentiation stage-related contribution of somatic PTPN11 mutations to leukemogenesis in childhood acute leukemia.* Blood, 2004. 104(2): p. 307-13.

[101] Paulsson, K., et al., *Mutations of FLT3, NRAS, KRAS, and PTPN11 are frequent and possibly mutually exclusive in high hyperdiploid childhood acute lymphoblastic leukemia.* Genes Chromosomes Cancer, 2008. 47(1): p. 26-33.

[102] Wiemels, J.L., et al., *RAS mutation is associated with hyperdiploidy and parental characteristics in pediatric acute lymphoblastic leukemia.* Leukemia, 2005. 19(3): p. 415-419.

[103] Perentesis, J.P., et al., *RAS oncogene mutations and outcome of therapy for childhood acute lymphoblastic leukemia.* Leukemia, 2004. 18(4): p. 685-92.

[104] Armstrong, S.A., et al., *FLT3 mutations in childhood acute lymphoblastic leukemia.* Blood, 2004. 103(9): p. 3544-6.

[105] Lee, J.W., et al., *BRAF mutations in acute leukemias.* Leukemia, 2003. 18(1): p. 170-172.

[106] Gustafsson, B., et al., *Mutations in the BRAF and N-ras genes in childhood acute lymphoblastic leukaemia.* Leukemia, 2005. 19(2): p. 310-2.

[107] Iacobucci, I., et al., *Cytogenetic and molecular predictors of outcome in acute lymphocytic leukemia: recent developments.* Curr Hematol Malig Rep. 7(2): p. 133-43.

[108] Roberts, K.G. and C.G. Mullighan, *How new advances in genetic analysis are influencing the understanding and treatment of childhood acute leukemia.* Curr Opin Pediatr, 2011. 23(1): p. 34-40.

[109] Iacobucci, I., et al., *The PAX5 gene is frequently rearranged in BCR-ABL1-positive acute lymphoblastic leukemia but is not associated with outcome. A report on behalf of the GIMEMA Acute Leukemia Working Party.* Haematologica, 2010. 95(10): p. 1683-90.

[110] Mullighan, C.G., et al., *Deletion of IKZF1 and prognosis in acute lymphoblastic leukemia.* N Engl J Med, 2009. 360(5): p. 470-80.

[111] Zhou, Y., et al., *Advances in the molecular pathobiology of B-lymphoblastic leukemia.* Hum Pathol, 2012. 43(9): p. 1347-62.

[112] Payne, K.J. and S. Dovat, *Ikaros and tumor suppression in acute lymphoblastic leukemia.* Crit Rev Oncog, 2011. 16(1-2): p. 3-12.

[113] Mullighan, C.G., et al., *JAK mutations in high-risk childhood acute lymphoblastic leukemia.* Proc Natl Acad Sci U S A, 2009. 106(23): p. 9414-8.

[114] Bercovich, D., et al., *Mutations of JAK2 in acute lymphoblastic leukaemias associated with Down's syndrome.* Lancet, 2008. 372(9648): p. 1484-92.

[115] Mullighan, C.G., *JAK2--a new player in acute lymphoblastic leukaemia.* Lancet, 2008. 372(9648): p. 1448-50.

[116] Mullighan, C.G., et al., *Rearrangement of CRLF2 in B-progenitor- and Down syndrome-associated acute lymphoblastic leukemia.* Nat Genet, 2009. 41(11): p. 1243-6.

[117] Harvey, R.C., et al., *Rearrangement of CRLF2 is associated with mutation of JAK kinases, alteration of IKZF1, Hispanic/Latino ethnicity, and a poor outcome in pediatric B-progenitor acute lymphoblastic leukemia.* Blood, 2010. 115(26): p. 5312-21.

[118] Rochman, Y., et al., *Thymic stromal lymphopoietin-mediated STAT5 phosphorylation via kinases JAK1 and JAK2 reveals a key difference from IL-7-induced signaling.* Proc Natl Acad Sci U S A, 2010. 107(45): p. 19455-60.

[119] Buettner, R., L.B. Mora, and R. Jove, *Activated STAT signaling in human tumors provides novel molecular targets for therapeutic intervention.* Clin Cancer Res, 2002. 8(4): p. 945-54.

[120] Shochat, C., et al., *Gain-of-function mutations in interleukin-7 receptor-alpha (IL7R) in childhood acute lymphoblastic leukemias.* J Exp Med, 2011. 208(5): p. 901-8.

[121] Hof, J., et al., *Mutations and deletions of the TP53 gene predict nonresponse to treatment and poor outcome in first relapse of childhood acute lymphoblastic leukemia.* J Clin Oncol, 2011. 29(23): p. 3185-93.

[122] Diccianni, M.B., et al., *Clinical significance of p53 mutations in relapsed T-cell acute lymphoblastic leukemia.* Blood, 1994. 84(9): p. 3105-12.

[123] Kawamura, M., et al., *Alterations of the p53, p21, p16, p15 and RAS genes in childhood T-cell acute lymphoblastic leukemia.* Leuk Res, 1999. 23(2): p. 115-26.

[124] Cayuela, J.M., et al., *Multiple tumor-suppressor gene 1 inactivation is the most frequent genetic alteration in T-cell acute lymphoblastic leukemia.* Blood, 1996. 87(6): p. 2180-6.

[125] Cheng, J. and M. Haas, *Frequent mutations in the p53 tumor suppressor gene in human leukemia T-cell lines.* Mol Cell Biol, 1990. 10(10): p. 5502-9.

[126] Weng, A.P., et al., *Activating mutations of NOTCH1 in human T cell acute lymphoblastic leukemia.* Science, 2004. 306(5694): p. 269-71.

[127] Breit, S., et al., *Activating NOTCH1 mutations predict favorable early treatment response and long-term outcome in childhood precursor T-cell lymphoblastic leukemia.* Blood, 2006. 108(4): p. 1151-7.

[128] Sambandam, A., et al., *Notch signaling controls the generation and differentiation of early T lineage progenitors.* Nat Immunol, 2005. 6(7): p. 663-70.

[129] van Vlierberghe, P., et al., *A new recurrent 9q34 duplication in pediatric T-cell acute lymphoblastic leukemia.* Leukemia, 2006. 20(7): p. 1245-53.

[130] Malyukova, A., et al., *The tumor suppressor gene hCDC4 is frequently mutated in human T-cell acute lymphoblastic leukemia with functional consequences for Notch signaling.* Cancer Res, 2007. 67(12): p. 5611-6.

[131] Vainchenker, W. and S.N. Constantinescu, *JAK/STAT signaling in hematological malignancies.* Oncogene, 2012.

[132] Flex, E., et al., *Somatically acquired JAK1 mutations in adult acute lymphoblastic leukemia.* J Exp Med, 2008. 205(4): p. 751-8.

[133] Gutierrez, A., et al., *High frequency of PTEN, PI3K, and AKT abnormalities in T-cell acute lymphoblastic leukemia.* Blood, 2009. 114(3): p. 647-50.

[134] Yokota, S., et al., *Mutational analysis of the N-ras gene in acute lymphoblastic leukemia: a study of 125 Japanese pediatric cases.* Int J Hematol, 1998. 67(4): p. 379-87.

[135] von Lintig, F.C., et al., *Ras activation in normal white blood cells and childhood acute lymphoblastic leukemia.* Clin Cancer Res, 2000. 6(5): p. 1804-10.

[136] Tosello, V., et al., *WT1 mutations in T-ALL.* Blood, 2009. 114(5): p. 1038-45.

[137] Renneville, A., et al., *Wilms' Tumor 1 (WT1) Gene Mutations in Pediatric T-Acute Lymphoblastic Leukemia.* ASH Annual Meeting Abstracts, 2009. 114(22): p. 3075-.

[138] Neumann, M., et al., *High Rate of FLT3 Mutations In Adult ETP-ALL.* ASH Annual Meeting Abstracts, 2010. 116(21): p. 1031-.

[139] Paietta, E., et al., *Activating FLT3 mutations in CD117/KIT(+) T-cell acute lymphoblastic leukemias.* Blood, 2004. 104(2): p. 558-60.

[140] Van Vlierberghe, P., et al., *Activating FLT3 mutations in CD4+/CD8- pediatric T-cell acute lymphoblastic leukemias.* Blood, 2005. 106(13): p. 4414-5.

[141] Zhang, J., et al., *The genetic basis of early T-cell precursor acute lymphoblastic leukaemia.* Nature, 2012. 481(7380): p. 157-63.

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