Acute lymphoblastic leukemia/lymphoma

BHS Training Course on Acute Leukemia

Pr Carlos Graux

Saturday December 14th, 2013
Hof Ter Musschen, Brussels
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**Multistep leukemogenesis**

**Cells and Progenitors**

- **STEM CELLS**
  - CLP
  - CMP
  - Self-renewal

- **COMMITTED PROGENITORS**
  - Pre-T cell
  - Pre-B cell
  - BFU-E
  - CFU-E
  - Meg-CFC
  - Mast-CFC
  - Eo-CFC
  - GM-CFC
  - M-CFC
  - Oc-CFC (?)

- **MATURE CELLS**
  - T-Lymphocyte
  - B-Lymphocyte/Plasma cell
  - Erythrocyte
  - Megakaryocyte/Platelets
  - Basophil/Mast cell
  - Eosinophil
  - Neutrophil
  - Monocyte/Macrophage/Kupffer cell
  - Dargenhan cell
  - Dendritic cell
  - Osteoclast
### Chromosomal rearrangements involving TCR → activation of transcription factors

- t(7;10)(q34;q24), t(10;14)(q24;q11) → **TLX1 (HOX11)** (7%/31%)
- * t(5;14)(q35;q32) (cryptic) → **TLX3 (HOX11L2)** (20%/13%), * **BCL11B /14q32**
- inv(7)(p15q34) (cryptic) → **HOXA** (3%)
- t(1;14)(p32;q11) → **TAL1** (3%)
- t(7;19)(q34;p13) → **LYL1** (<1%)
- t(11;14)(p15;q11) → **LMO1** (2%)
- t(11;14)(p13;q11) and t(7;11)(q35;p13) → **LMO2** (3%)
- t(7;9)(q34;q34.3) → **NOTCH1** (<1%)
- t(6;7)(q23;q24) → **MYB** (<1%)
- ...

### Formation of fusion genes

- 1p32 deletion → **SIL-TAL1** (9-30%)
- t(10;11)(p13;q14) (often cryptic) → **CALM-AF10** (10%)
- t(11;?) (q23;?) → **MLL-?** (4-8%)
- t(9;9)(q34;q34) (most often on amplified episomes) → **NUP214-ABL1** (6%)
- ...

### (Cryptic) deletions

- 9p21 → loss of **P16 (CDKN2A)** (65%)
- del(6q) → ?
- ...

### Duplications

- 6q23.3 → **MYB**
- 9q34 → **ABL1, VAV2, TRAF2, NOTCH1?**
- ...

### (Activating or inactivating) mutations

- **NOTCH1, PTEN, FBXW1, FLT3, N-RAS, JAK1**
- ...

### Aneuploidy
Deregulation of (pre)-TCR and other signaling components
- ABL1, LCK, RAS, FLT3, JAK1/2

Deregulation of cell cycle components
- CDKN2a, CCND2

Aberrant expression of oncogenes
- TAL1, TAL2, LYL1 +/- LMO1/2
- SET-NUP214
- CALM-AF10
- MLL-fusions
- TLX1, TLX3

Deregulation of thymopoiesis regulators
- HOXA
- MYB
- E2A
- NOTCH1
- FBXW7, PTEN

Response to growth signals
Cell cycle control
Regulation of hematopoiesis
Self-renewal capacity
Multistep leukemogenesis

T-ALL
Proliferation↑
Apoptosis↓
Differentiation arrest
Accumulation of cells

Multistep leukemogenesis
Age-specific incidence of ALL

SE Sallan. Hematology 2006
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**Precursor B or T-cell acute lymphoblastic leukemia (WHO)**

- L1 ALL (FAB)
- L2 ALL (FAB)
- L3 ALL (FAB)

**Burkitt’s lymphoma (WHO)**
Immunophenotyping

Pui C-H and Looks AT. Lancet 2008
### GEIL/EGIL Scoring system

<table>
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<tr>
<th>Points</th>
<th>B lineage</th>
<th>T lineage</th>
<th>Myeloid lineage</th>
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<tr>
<td>2</td>
<td>CD79</td>
<td>CD3</td>
<td>MPO (lysozyme)</td>
</tr>
<tr>
<td></td>
<td>cμ</td>
<td>TCR</td>
<td></td>
</tr>
<tr>
<td></td>
<td>cCD22</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>CD19</td>
<td>CD2</td>
<td>CD13 CD33 CD117</td>
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<td></td>
<td>CD10</td>
<td>CD5</td>
<td>CD65 CD117</td>
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<tr>
<td></td>
<td>CD20</td>
<td>CD8</td>
<td></td>
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<tr>
<td>0.5</td>
<td>TdT</td>
<td>TdT</td>
<td>CD14 CD15 CD64</td>
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<tr>
<td></td>
<td>CD24</td>
<td>CD7</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>CD1a</td>
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Biphenotypic AL: > 2 points for myeloid antigens and one of the lymphoid lineage
**GEIL/EGIL classification of B-cell ALL**

<table>
<thead>
<tr>
<th></th>
<th>cCD79/CD19/CD22 (s ou c)</th>
<th>CD10</th>
<th>C-μ</th>
<th>sIg</th>
</tr>
</thead>
<tbody>
<tr>
<td>B1</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>B2</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>B3</td>
<td>+</td>
<td>+/-</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>B4</td>
<td>+</td>
<td>+/-</td>
<td>+/-</td>
<td>+</td>
</tr>
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*B1 = pro-B-ALL, B2 = Common B-ALL, B3 = pre-B-ALL, B4 = mature B-ALL*
**GEIL/EGIL classification of T-cell ALL**

<table>
<thead>
<tr>
<th></th>
<th>cCD3</th>
<th>CD7</th>
<th>CD2/CD5/CD8</th>
<th>CD1a</th>
<th>sCD3/CD1a-</th>
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</thead>
<tbody>
<tr>
<td><strong>T1</strong></td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td><strong>T2</strong></td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td><strong>T3</strong></td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td><strong>T4</strong></td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
</tr>
</tbody>
</table>

*T1 = Pro-T-ALL, T2 = Pre-T-ALL, T3 = cortical T-ALL, T4 = mature T-ALL*
Relevance of immunophenotyping

- **Diagnosis** of B-ALL/T-ALL/bi-phenotypic AL

- **Prognosis** value of some sub-types
  - CD10+ B-ALL (common): favorable?
  - CD1a+ T-ALL (cortical): favorable?
  - CD34+ T-ALL: unfavorable?

- **Specific therapy**
  - Identifying mature B-cell ALL (Burkitt’s)
  - Some surface markers are potential targets for antibody therapy and for other innovative therapies (CD20, CD22, CD52, CD33, ERBB2, CD19 …)

- In most cases **minimal residual disease** can be conveniently assessed by flow cytometry
Risk assessment

Balance between risk of relapse and toxicity of the treatment

Takes into account:

- **patient (host) characteristics**
  - age (comorbidity), social situation (compliance), general condition,...
  - pharmacodynamics, pharmacogenetics

- **disease characteristics**
  - clinical prognostic features
  - genetics (chromosomal/gene abnormalities, MDR genes expression, gene expression profiling, ...)

→ selecting therapy that will avoid excessive toxicity but maintain a high cure rate
Age

**Patient (host) characteristics**
- Age
  - < 6 months: especially poor outcome
  - 1-9 y: do globally better
  - > 35 y: negatively impacts on transplantation success
  - > 65 y: generally worse prognosis
  - comorbidity: generally worse prognosis
  - specific trials: Pui CH. NEJM 2004

**Disease characteristics**
- High frequency of unfavourable genetic features and low rate of favourable genetic abnormalities in adults.
- Prognosis of certain genetic subtypes depends on age.
  - Ph+ ALL (1-9 y do better)
  - MLL-AF4 ALL (<1 y and adults do worse)

Pui CH. NEJM 2004
Pharmacodynamics/genetics

Polymorphisms in genes that encode drug-metabolizing enzymes, transporters, receptors, and drug targets

→ wide differences in terms of drug disposition and pharmacologic effects
→ influence toxicity and efficacy of chemotherapy

• Drug interactions!

Phenytoin, phenobarbital, carbamazepine
→ induce the production of cytochrome P-450 enzymes
→ increase the systemic clearance of antileukemic agents
→ adversely affect treatment outcome
**Definition**

**Epidemiology**

**Diagnosis**

**Risk assessment**

**Treatment**

**New drugs**

**Ccl**

**Patient (host) characteristics**

**Disease characteristics**

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**Homozygous or heterozygous deficiency of thiopurine methyltransferase**

**Cumulative Incidence of Mercaptopurine Dose Reductions**

- Mutant TPMT
- Heterozygous
- Wild type
- P < 0.001

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**C>T polymorphism** at position 677 in the methylenetetrahydrofolate reductase (MTHFR) gene

**According to MTHFR Genotype**

- CC
- CT
- TT

**P < 0.05**

---

**Tandem-repeat polymorphism** within the enhancer region of the thymidylate synthase gene - one of the major targets of methotrexate

**Probability of Event-free Survival**

- 2R/2R or 2R/3R
- 3R/3R
- P = 0.001

---

**Host Genotypes**

- Whites
- Blacks

- TPMT
- MTHFR
- TS
- GSTT1
- MDR1
- RFC
- CYP3A5
- NQO1
- CBS

---

**All Blast Genotypes**

- Hyperdiploidy
- TEL-AML1
- HOX11
- TAL1
- E2A-PBX1
- MLL-AF4
- BCR-ABL

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**Pui CH. NEJM 2004**
Clinical features

- Leukocyte count
  - > 30,000/µL (B-ALL)
  - > 100,000/µL (T-ALL)

- Extramedullary disease

- High LDH level,

- Low Hgb level, low platelet count

- CNS involvement

Negatively impact on prognostic
## Cytogenetics

### B-cell precursor ALL

**Favorable features:**
- hyperdiploidy (> 50 chromosomes)
- t(12;21) → **TEL-AML1**
- t(1;19) → **E2A-PBX1**  
  (CD34-, CD20-)
- trisomy 4, 10, 17
- t(12;21) → **TEL-AML1** in 30% of childhood cases
- t(1;19) → **E2A-PBX1** in 5% of adult cases
- t(1;19) → **E2A-PBX1** in 5% of adult cases
- trisomy 4, 10, 17

**Unfavorable features:**
- hypodiploidy (< 45 chromosomes)
- t(4;11) → **MLL-AF4**  
  (CD10-, CD19+, CD15+)
- t(9;22) → **BCR-ABL1** (p190 or p210)  
  (CD34+, myeloid antigens, CD25)

**Recommended treatment:**
- HD MTX
- Intensive Asparaginase
- Intensive
- HD Ara-C
- Glivec/ new TKI

### T-cell precursor ALL

**Favorable features:**
- t(7;10) and t(10;14) → **HOX11 (TLX1)**  
  (CD10+/-, CD1a+)
- t(11;19) → **MLL-ENL**

**Unfavorable features:**
- t(5;14) (cryptic) → **HOX11L2**

**Recommended treatment:**
- HD MTX, Ara-C, cyclophosphamide

**Controversial**
- Impact of NUP214-ABL1 expression?
Impact of cytogenetics on prognostic
Gene expression profiling/ CNA arrays

• Demonstrates specific gene-expression patterns in known genetic subtypes of leukemia

• Reveals new subtypes (ex: the BCR-ABL1 like subtype)

• Allows classification of samples according to outcome

• Identifies genes
  - whose expression may have prognostic significance (IKZF1 deleted, NOTCH1 mutated)
  - related to the intracellular disposition of antileukemic agents in vivo
  - associated with resistance to chemotherapy

• Reveals new pathogenic mechanisms to identify novel therapeutic targets
Definition
Epidemiology
Diagnosis
Risk assessment
Treatment
New drugs
Ccl

**Genetic abnormalities**

**Host genome variability**
- *TPMT* alleles
- *TPMT* alleles

**Clinical prognostic features**

**Gene expression**
- Poor response
- Good response

**Proteomics**

**New targets**
**New medications**
**Better treatment?**
**Disease prevention?**
Treatment

Chemotherapy
- non specific
- narrow therapeutic index

Optimal use of the same antileukemic agents
Steroid sensitivity (prednisone 60 mg daily for 7 days: blast cells should be less than 1000/µL in peripheral blood by day 8)

Remission induction

• **Goal**
  - to eradicate > 99 % of the initial burden of cells
  - to restore normal hematopoiesis
  - to restore a normal performance status

• **Always includes the administration of:**
  - a glucocorticoid (prednisone, prednisolone, or dexamethasone),
  - vincristine,
  - and at least one other agent (usually asparaginase, an anthracycline, or both). Interest of cyclophosphamide in T-ALL.

→ complete remission rates of 96-99 % for children and 78-93 % for adults
Evaluation of response

Response to treatment depends on interconnected variables:

- the ability of individual patients to metabolize anti-leukemic drugs (polymorphisms)
- clinico-biologic features of the disease
- chemotherapy dosages, schedule of administration & interactions

Leukemia cytoreduction = rate of clearance of leukemic cells during remission induction

- reflects the collective impact of these variables
- consistently useful prognostic indicator
- evaluated by morphology at day 15 (insensitive)

Measure of the minimal residual disease by molecular and flow cytometric methods is >100-fold more sensitivity

< 0.01 % (10^-4) during or on completion of initial remission-induction therapy
   → exceptionally good treatment outcome

> 1 % at the end of remission-induction therapy or > 0.1 % at later times
   → a very high risk of relapse
Minimal residual disease

**MRD at day 29 by immunophenotyping**

- **88±1%**
- **59±5%**
- **49±6%**

**MRD at day 30 by RQ-PCR for IgH/TCR rearrangement**

- **Undetectable**
- **10^-6 to less than 10^-5**
- **10^-5 to less than 10^-4**
- **10^-4 to less than 10^-3**
- **10^-3 to less than 0.01**
- **0.01 or more**

**Event-free survival probability**

- MRD negative (≤0.01%) \(n=1588\)
- 0.01% < MRD ≤ 0.1% \(n=175\)
- 0.1% < MRD ≤ 1.0% \(n=141\)
- MRD > 1.0% \(n=67\)

**Years**

- **P < 0.0001**

**Borowitz MJ et al. Blood 2008**


→ identify patients predicted to have superior outcome who might be candidates for trials testing less intensive therapies
Intensification (consolidation) / re-induction

• Goal
  - eradicate drug-resistant residual leukaemic cells
  - reduce risk of relapse

• No consensus on the best regimen and duration
  - Intensification:
    • high dose methotrexate (→ 5 gr/m2) + mercaptopurine
  - Reinduction treatment:
    • essentially a repetition of the initial induction therapy:
      - Frequent pulses of vincristine and corticosteroids
      - Prolonged high doses of asparaginase
      - Cytarabine, cyclophosphamide, anthracyclines (in adults)
Intensification - Allogeneic HSCT

- Ultimate form of treatment intensification

- Risk of relapse decreases with allogeneic HSCT but the concomitant TRMortality eliminates the potential survival benefit
- Also to long term TRMorbidity

- > 35 y, in Ph- ALL, improved outcome seen in patients who undergo a MUD allogeneic HST is progressively lost when using myeloablative regimen

- Allogeneic transplantation benefits certain very-high-risk pediatric and adult patients
  - Clearly
    - BCR-ABL+ ALL
    - poor initial response to treatment (CR > 28 d)
    - adults who have ALL with t(4;11)
    - Second remission
  - Less clear
    - WBC > 30,000? >100,000 in T-ALL?
    - Refractory ALL?
    - CNS ALL?

- Among adults with high risk ALL,
  - long-term DFS of 30 to 40 % have been obtained with chemotherapy,
  - as compared with 45 to 75 % with allogeneic HSCT
Patient characteristics:

- at least one of the following features
  - > 35 y or
  - B-ALL or
  - WBC > 30000 or
  - t(9;22) or t(4;11) or t(1;19) or
  - failure to achieve CR

if HLA identical sibling

⇒ Allo HSCT

If no HLA identical sibling or age > 50 Y

⇒ auto BMT

**Ph+ ALL**

**Before imatinib**
- Allogeneic HSCT conferred similar OS and relapse rates for Ph+ patients compared with those with normal cytogenetics supporting a graft-versus-leukemia (GVL) effect  
- but
  • Ph+ ALL increase with age
  • Availability of a donor
  • Low rate of remission
  • Relapse before transplantation

**With imatinib**
- Given during induction → CR rate increase from approximately 60% to 90% → more HSCT
- Given after transplantation → decrease relapse rate

→ Imatinib + conventional chemotherapy provided results comparable with allogeneic HSCT  
  » de Labarthe A. Blood 2007
  • but clinical resistance to imatinib develops
  • kinase domain mutations of BCR-ABL1 give rise to relapse (frequently precede imatinib-based therapy)  
  » Pfeifer H. Blood 2007

→ Still recommended to proceed to HSCT in Ph+ ALL whenever possible (at least in adults ph+ ALL)

**New TKI (dasatinib, nilotinib)**
**Ph+ ALL**

Dombret et al. Blood 100 p2357, 2002

Doney et al. Biol Blood Marrow Transplant. 2003
Patients with ALL generally require prolonged continuation therapy
- for two years or more
- 12 months would be enough for most pediatric cases (2/3) but no reliable markers available

The base of most continuation regimens is a combination of
- methotrexate administered weekly and
- mercaptopurine given daily

Accumulation of increased intracellular concentrations of the active metabolites of methotrexate and mercaptopurine, and administration of this combination to the limits of tolerance, have been associated with improved clinical outcome

The identification of inherited deficiency of thiopurine-S-methyltransferase among patients with hematopoietic toxic effects allows the clinician to lower the dose of mercaptopurine selectively without modifying the dose of methotrexate
CNS prevention treatment

- CNS relapses account for 30-40% of initial relapses

- Factors associated with an increased risk of CNS relapse include:
  - high risk genetic features,
  - T-cell immunophenotype,
  - a large leukemia-cell burden: hyperleukocytosis, extramedullary disease
  - presence of leukemia cells in the cerebrospinal fluid (even from iatrogenic introduction through a traumatic lumbar puncture)
  - certain polymorphisms in genes coding for drug metabolizing enzymes

- Based on:
  - cranial irradiation (second cancers, late neurocognitive deficits, and endocrinopathy, ...) ... now avoided in most pediatric protocols
  - largely been replaced by
    - intrathecal therapy: methotrexate, cytarabine (depocyte)
      - no traumatic lumbar punctures
    - systemic chemotherapy: HD methotrexate, dexamethasone
CNS-ALL

• At diagnosis
  - > 5 WBC/µL with typical morphology
  - Incidence: +/- 7%
  - Treatment (not standardized):
    • intrathecal drug(s) twice weekly until clearance of blast cells
    • +“intensive” systemic (allogeneic HSCT)
    • CNS irradiation

• 2-10% of relapses restricted to the CNS
  - outcome depends on the duration of remission,
  - T-cell ALL or prior cranial irradiation are bad factors
Patients aged 15-20 years.

Nicolas Boissel et al. JCO. 2003

Paediatric treatments are more effective
Better adherence by patients, parents, and doctors

→ need for protocols adapted to young adults
The elderly patient (> 55 y → > 65 y)

- Increased incidence of ALL

- Biological differences in the spectrum of ALL (more Ph+ ALL, less T-ALL, less favorable cytogenetic features)

- Coexisting medical disorders → decreased tolerance for chemotherapy
  High mortality rate during induction if treated according to young adult programs (corticoids- vincristine, l-asparaginase, ...)

- Since TKI therapy area → Ph+ ALL is “a good prognostic factor” in the elderly

- New less toxic formulations of old-drugs (PEG-asparaginase, liposomal cytarabine, vincristine, liposomal and PEGylated anthracyclines, ...)

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<td>Induction</td>
<td>Intensification</td>
<td>Continuation</td>
<td>CNS prevention</td>
<td>Specific situations</td>
<td></td>
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The elderly patient (> 55 y → > 65 y)

Delannoy et al. Leukemia 2006
The relapsing patient

The length from first CR (> vs < 2 years) has a major impact on outcome

No standard rescue therapy (Hyper-CVAD, …)

- CR rates with various regimens ± 50%
- CR duration ± 2-5 months

Autologous transplantation: possibly superior outcome

Allogeneic transplantation: whenever feasible (+ 20-30% long-term DFS)
New drugs

- Monoclonal antibodies
  - NOTCH1
  - Gamma-secretase inhibitors
  - CD20: rituximab
  - CD22: epratuzumab
  - CD33: gemtuzumab ozogamicin
  - CD52: alemtuzumab
  - CD19: blinatumomab

- Tyrosine kinase receptor
  - P
  - Midostaurin
  - Sinitinib
  - Imatinib
  - Dasatinib
  - Linotinib
  - ABL1
  - SRC

- RAS
  - RAF
  - PI3K
  - MYC
  - JAK
  - STAT

- CDK
  - AKT
  - mTOR
  - NFkB

- BCL2

- Protein
  - Oblimersen
  - Sirolimus
  - Bortezomib

- Nucleus
  - HDAC
  - CH3
  - DMT
  - Azacytidine
  - Decitabine
  - Vorinostat
  - Valproic acid

- Cell membrane
  - Cytoplasm
Blinatumomab (MT103) is a Bispecific T-cell Engager (BiTE®) antibody designed to direct cytotoxic T-cells to CD19 expressing cancer cell

Blinatumomab: mode of action
Blinatumomab: mode of action
Blinatumomab: mode of action
Conclusion

- Cure rate of **childhood** ALL > 80%
  - Still serious acute and late complications due to treatments (osteonecrosis, hyperglycemia, anthracycline-induced myocardial injury, neurologic defects, ...)
  - Shift toward the reduction of deleterious acute and **late** effects of treatment

- Cure rate of **adults** ALL remains low (< 40%)
  - The future resides in defining the molecular pathways underlying the pathogenesis of ALL in order to find proteins suitable for less toxic targeted therapy

- Further elucidating the underlying pharmacogenetic factors of the host

- When comparing different treatment trials, remember that slightly different median ages can translate into relatively large differences in outcome
References


