Recurring cytogenetic abnormalities in Leukemia and their prognostic significance

Jovita Reyes Memorial Pediatric Hematology/Oncology Nursing Conference

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Disclosures

• None
Objectives

• Identify the normal Chromosome number
• Understand the major different classifications of mutations
• Identify the recurring abnormalities in ALL
• Identify the recurring abnormalities in AML
• Know the prognostic significance of the above abnormalities
Normal Chromosome number

Concepts of Genetics
Can’t really see chromosomes
Genomic Instability

• Gains and losses of whole chromosomes is a hallmark of cancer cells
• Chromosomal rearrangements are one of the most common cytogenetic findings associated with cancer cells
• Important step in converting normal cells into Cancer cells
• Thought to confer further malignant potential and drug resistance
• Just because you have instability doesn’t mean you’ll get cancer
Types of Changes

• Large Changes
  – Translocations
  – Aneuploidy
  – Amplifications

• Small Change
  – Missense/nonsense mutations
  – Insertions/deletions
Translocations
Classic translocation

46,XX,t(9;22)(q34;q11.2)
Aneuploidy

• Gains and loss of chromosomes
Amplifications

• A segment amplified
Missense/nonsense mutations

Central Dogma

mutations

• Missense
  – V617F

• Nonsense
  – Early stop

• Insertions/deletions
  – Inframe
  – Out of frame

http://www.iusd.org
Diagnostic Tests Large Changes

• Karyotype
  – Grow the cells
  – Harvest chromosomes
  – Stain them
  – Count 20 metaphase
Karyotype

• Ploidy
• Identifiable Translocations
Fluorescent *in situ* Hybridization (FISH)
Example

- **CHROMOSOME REPORT:**
- **SPECIMEN TYPE AND TYPE OF STUDY:** A Bone Marrow: Full Study/FISH
- **FISH Results:** Abnormal  
  Chromosome Results: Abnormal
- **KARYOTYPE RESULTS:**
  46,XY,add(11)(q?23),t(13;15)(q14;q2?6),i(21)(q10)[8]/46,XY[5]

- **IMPRESSIONS AND RECOMMENDATIONS:** Eight of thirteen metaphase cells analyzed comprised a single abnormal clone with abnormalities involving chromosomes 11, 13, 15 and 21. Five cells appeared normal male. Seven further cells, which were not completely analyzable were found but showed one or more of the abnormalities found in the clone.

- Fluorescent in situ hybridization (FISH) was performed using the Vysis TEL/AML1 dual color probes. Of 200 interphase cells scored, 37 had the normal 2 red/2 green signals; 163 cells (81.5%) had abnormal signals: 2 red, 1 green/1 yellow/1 small red pattern indicating a rearrangement had taken place. The large double red signals most likely reflect the apparent isochromosome 21 seen in metaphase cells.
Another example

- CHROMOSOME REPORT: Bone Marrow: Full Study/FISH Supplemental
  indications: Acute Lymphocytic Leukemia
- FISH Results: Abnormal   Chromosome Results: None

- KARYOTYPE RESULTS: See below

- IMPRESSIONS AND RECOMMENDATIONS: This specimen was set up, cultured and
  harvested according to standard laboratory protocol. No metaphase cells were
  found in all available material. A repeat specimen is requested to further delineate
  potential clonal abnormalities, including amplification of AML1, suggested by FISH
  results, below. Fluorescent in situ hybridization (FISH) was performed with an ALL
  probe panel. Probe sets and number of cells scored are listed below.

- ABNORMAL FISH RESULTS: TEL/AML1: 7/200 cells (3.5%) had 6 or 7 signals for
  AML1. The chromosomal context of the extra signals remains unknown in the
  absence of metaphase chromosomes. Amplification of AML1 has been described
  in children and young adults with ALL. There was no evidence for TEL/AML fusion.
iAMP21

Kristine Harrison
Diagnostic Test Small Changes

- Molecular markers
- Sequence specific
- Insertions and deletions
- Mostly PCR based assays
FLT3-ITD
Molecular markers

- Specific DNA test
- sequenom
ALL

- Favorable Cytogenetics
  - t(12;21)
  - Hyperdiploid with trisomy 4, trisomy10
- Unfavorable Cytogenetics
  - t(9;22)
  - 11q23 rearrangement
    - t(4;11), t(10;11), etc
  - Hypodiploid (<44chr)
Prognosis of children with acute lymphoblastic leukemia (ALL) and intrachromosomal amplification of chromosome 21 (iAMP21)

Anthony V. Moorman,1 Susan M. Richards,2 Hazel M. Robinson,1 Jon C. Strefford,1 Brenda E. S. Gibson,3 Sally E. Kinsey,4 Tim O. B. Eden,5 Ajay J. Vora,6 Christopher D. Mitchell,7 and Christine J. Harrison,1 on behalf of the UK Medical Research Council (MRC)/National Cancer Research Institute (NCRI) Childhood Leukaemia Working Party (CLWP)
### Table 2. Overview of the AALL08B1 Classification System for B-precursor ALL

<table>
<thead>
<tr>
<th>Risk Group</th>
<th>Low</th>
<th>Average&lt;sup&gt;2&lt;/sup&gt;</th>
<th>High&lt;sup&gt;3&lt;/sup&gt;</th>
<th>Very High&lt;sup&gt;4&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Projected 5-yr EFS</td>
<td>&gt; 95%</td>
<td>90-95%</td>
<td>75-90%</td>
<td>&lt; 75%</td>
</tr>
<tr>
<td>NCI Risk-Group</td>
<td>SR</td>
<td>SR</td>
<td>SR</td>
<td>SR HR</td>
</tr>
<tr>
<td>Favorable Genetics&lt;sup&gt;1&lt;/sup&gt;</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>Any</td>
</tr>
<tr>
<td>Day 8 PB MRD</td>
<td>&lt; 0.01%</td>
<td>&gt;0.01%</td>
<td>&lt;1%</td>
<td>Any ≥ 1%</td>
</tr>
<tr>
<td>Day 29 BM MRD</td>
<td>&lt; 0.01%</td>
<td>&lt; 0.01%</td>
<td>&lt;0.01%</td>
<td>≥ 0.01%</td>
</tr>
<tr>
<td>Patient Accrual/Year</td>
<td>250</td>
<td>360</td>
<td>275</td>
<td>70 91 435 86 74 61</td>
</tr>
<tr>
<td>Total Patient Accrual/Year</td>
<td>250</td>
<td>635</td>
<td>596</td>
<td>221</td>
</tr>
<tr>
<td>Fraction of Patients</td>
<td>14.7%</td>
<td>37.3%</td>
<td>35%</td>
<td>13%</td>
</tr>
</tbody>
</table>

1 “Yes” is defined as the presence of double trisomy 4 and 10 OR *ETV6-RUNXI* fusion
2 NCI SR patients who are CNS2 will be included in Average Risk and will not be eligible for the LR Arm.
3 All patients with CNS3 or testicular involvement will be assigned to the HR study, but may change to VHR study if Day 29 MRD ≥ 0.01%.
4 Includes patients with hypodiploidy (< 44 chromosomes and/or DNA index < 0.81), Induction failure, and *MLL* rearrangement (not *MLL* deletion). *BCR-ABL1* positive patients are eligible for a separate, dedicated Ph+ ALL study, AALL0622 or successor protocol.
AML

• Favorable cytogenetics (>50%)
  – t(8;21)
  – INV16
  – t(15;17)
• Favorable molecular markers
  – Npm mutation
  – CEBP mutation
• Unfavorable cytogenetics (<50%)
  – Monosomy 7
  – Del 5q
• Unfavorable molecular markers
  – FLT3-ITD
**Risk Classification:** Based on cytogenetics, molecular markers and MRD post-Induction I.

Low Risk (LR): Inv(16), (t;8:21), nucleophosmin (NPM), CEBPa with any MRD status or standard risk cytogenetics with negative MRD at end of Induction I.

High Risk (HR): High allelic ratio FLT3/ITD+, monosomy 7, del(5q) with any MRD status or standard risk cytogenetics with positive MRD at end of Induction I.
Future directions

• New molecular markers
  – ALL
    • CRLF2
    • JAK mutations
    • Ikaros
    • ???
  – AML
    • JAK mutations
    • WT1, RUNX1, KIT, n-ras, etc
More to come...

• Thanks!!