Aetiology of Childhood Leukaemia

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What is Leukaemia?

- Leukaemia is a clonal disease originating in a single cell.
- It evolves by the accrual of mutations within a clone resulting in genetic diversification.
- Dominant mutant subclones are then naturally selected.
- The nature of the clone and how far it has evolved determines the clinical outcome.
- Delay in diagnosing increases the likelihood that the clone will have progressed to the point that additional mutations will have been acquired.
Age distribution of childhood leukaemia
Childhood cancers other than leukaemia

<table>
<thead>
<tr>
<th>Age in years</th>
<th>Female</th>
<th>Male</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>4.5%</td>
<td>-</td>
</tr>
<tr>
<td>1</td>
<td>3.0%</td>
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<tr>
<td>2</td>
<td>2.5%</td>
<td>-</td>
</tr>
<tr>
<td>3</td>
<td>2.0%</td>
<td>-</td>
</tr>
<tr>
<td>4</td>
<td>1.5%</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>1.0%</td>
<td>-</td>
</tr>
<tr>
<td>6</td>
<td>0.5%</td>
<td>-</td>
</tr>
<tr>
<td>7</td>
<td>0.0%</td>
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<tr>
<td>9</td>
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<tr>
<td>10</td>
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</tr>
<tr>
<td>12</td>
<td>0.0%</td>
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</tr>
<tr>
<td>13</td>
<td>0.0%</td>
<td>-</td>
</tr>
<tr>
<td>14</td>
<td>0.0%</td>
<td>-</td>
</tr>
</tbody>
</table>
Childhood Leukaemia

- Biological heterogeneity is well documented
- Two major morphological subtypes:
  - Acute lymphoblastic leukaemia (ALL)
  - Acute myeloblastic leukaemia (AML)
- Characterised by molecular alterations, over 200 of which have been identified
- Chromosomal translocations are common along with simple gains/losses of chromosomes
- In more advanced disease, gene deletions and mutations are also relatively common
## Subtypes of childhood ALL

<table>
<thead>
<tr>
<th>Cell type involved</th>
<th>Chromosome abnormality</th>
<th>Molecular Lesion</th>
</tr>
</thead>
<tbody>
<tr>
<td>B-cell (infants)</td>
<td>11q23 translocations</td>
<td><em>MLL-AF4, MLL-ENL,</em> and other fusions</td>
</tr>
<tr>
<td>B-cell precursor</td>
<td>hyperdiploidy</td>
<td>Increased gene dosage</td>
</tr>
<tr>
<td></td>
<td>t(12;21)(p13;q22)</td>
<td><em>TEL-AML1</em> fusion</td>
</tr>
<tr>
<td></td>
<td>t(1;19)(q23;p13)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>t(9;22)(q34;q11)</td>
<td><em>E2A-PBX1</em> fusion</td>
</tr>
<tr>
<td>T-cell precursor</td>
<td>1q deletion;</td>
<td><em>BCR-ABL</em> fusion</td>
</tr>
<tr>
<td></td>
<td>t(1;14)(p32;q11)</td>
<td></td>
</tr>
</tbody>
</table>
Molecular subsets of ALL in infants, children and adults

Taken from Greaves, BMJ 2002
### Subtypes of childhood AML

<table>
<thead>
<tr>
<th>Cell type involved</th>
<th>Chromosome abnormality</th>
<th>Molecular Lesion</th>
</tr>
</thead>
<tbody>
<tr>
<td>In infants</td>
<td>11q23 translocations</td>
<td><em>MLL-AF6,-AF9, -AF10</em> or other fusions</td>
</tr>
<tr>
<td></td>
<td>t(8;21)(q22;q22)</td>
<td><em>AML-ETO</em> fusion</td>
</tr>
</tbody>
</table>
Genes of interest

- **MLL**
  - found in infant ALL and secondary t-AML
  - over 50 partner genes
  - regulates homeotic gene expression and chromatin stability

- **AML1**
  - frequently involved in leukaemogenesis
  - combines with $TEL \rightarrow TEL-AML1$
  - activated tyrosine kinase or novel protein

- Knockout studies have demonstrated that $AML1/TEL/MLL$ are all essential for haemopoiesis
Chromosomal translocation to form the TEL-AML1 fusion gene

Taken from Greaves, BMJ 2002
TEL-AML1 gene fusion

Chromosome 12, TEL gene

Chromosome 21, AML-1 gene

Translocation (12:21)
TEL-AML1 fusion gene

Taken from Greaves, BMJ 2002
Origin of childhood leukaemia

- Neonatal blood spots and cord bloods from newborns support the hypothesis that chromosomal translocations can initiate leukaemia \textit{in utero}.
- Wiemels \textit{et al} demonstrated \textit{TEL-AML1} mutations in 6 out of 9 patients using Guthrie cards.
- Animal modelling studies and twin concordance rates support the theory that initiation can occur \textit{in utero}.
“Two hit” model for childhood leukaemia

Initiation (common) → Chromosomal Translocation → Transition to acute leukaemia (rare)

Covert preleukaemia

Birth  →  15 years old
• What is the second hit?

• Is there a single cause for all types of childhood leukaemia?

• Is there a role for a gene-exposure interaction?
Mechanisms of exposure

- Prenatal and early life exposures are believed to be important determinants of leukaemia
- Several possible mechanisms by which exogenous agents may be involved in the aetiology of childhood leukaemia
  - Early maternal contact
  - Paternal
  - *In utero*
  - Postnatally
Gene Pool

Parental gametes

Parental

Grandmother

Parental preconceptional

Environmental Exposures

Gestational

Maternal

Transplacental

Direct

Parental

Postnatal

Parental

e.g. retinoblastoma

e.g. trisomy & leukaemia

e.g. DES & vaginal adenocarcinoma

e.g. X-rays and leukaemia

e.g. hepatitis B virus & heptacellular carcinoma
Candidate exposures

• In adults and children, there is epidemiological evidence to suggest that certain exposures may play a role in the development of some subtypes of leukaemia and lymphoma
  – Ionising radiation
  – Chemical (e.g. benzene)
  – Viruses
  – bacteria

• However, whether any of these exposures have a major role in childhood cancer is unclear
In utero exposures

- DNA topoisomerase II inhibitors
- Folate
- Viruses
- Chemicals
- Infections
DNA topoisomerase II inhibitors

- *MLL* rearrangements seen in adult cases of leukaemia after chemotherapy using DNA topoisomerase II inhibitors
- Greaves suggested that *in utero* exposure may be important in infant leukaemia risk
- Extensive list of inhibitors
  - Benzene metabolites, bioflavanoids, herbal medicines, anthraquinone laxatives, pesticides
  - Present in tea, coffee, wine, fruits
- Some studies have identified increase risk of ALL following exposure
- Not clear whether due to inhibition of DNA topoisomerase II or affects on other pathways
- Single case where mother of child with leukaemia had been exposed to premethrin with evidence of MLL gene fusions
Folate

- Folate deficiency influences DNA methylation, impairs DNA synthesis and repair
- Major enzyme studied to date is MTHFR
- Polymorphisms have been reported at positions 677 and 1298
- Studies have reported that these polymorphisms are associated with reduced risk of ALL
Folate Pathway

DNA methylation

SAM

Homocysteine

Methionine

MS

B12

5,10-methyl THF

MTHFR

Folic acid

DHF

DHFR

THF

SHMT

5,10-methylene THF

10-formyl THF

Purines

DNA synthesis

dTMP

dUMP

TS
Other areas of interest

- Breast feeding

- Reproductive technologies

- Infections
  - Two hypotheses that abnormal response to infections may play a role in the development of ALL in children
    - “delayed infection”
    - “hygiene hypothesis”
Summary

• Genetic and environmental factors have been implicated in the aetiology of many human diseases including childhood cancer
• There is increasing genetic evidence to suggest that gene arrangements can originate \textit{in utero}
• However, whilst many environmental agents have been suggested as risk factors for childhood leukaemia the data are conflicting and often contradictory and the search for the candidate exposure – assuming they exist-continues
• Predominantly research has focused on the index child but we may be able to gain further insight by examining other family members