Society of Hematopathology 2017 Workshop
Session 1 summary
Germline Predisposition Syndromes

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Myeloid Neoplasms with Germline Predispositions

new recognition in the revised WHO classification

• Without a pre-existing disorder or organ dysfunction
  – AML with germline CEBPA mutation (2 cases)
  – Myeloid neoplasms with germline DDX41 mutation (1 case)

• Pre-existing platelet disorder
  – Germline RUNX1 (10 cases)
  – Germline ANKRD26 (1 case)
  – Germline ETV6 mutation

• Other organ dysfunctions
  – Germline GATA2 (16 cases)
  – Bone marrow failure syndromes
  – JMML associated with NF, Noonan’s or Noonan-like disorders (3 cases)
  – Down syndrome
AML with germline *CEBPA* mutation

- Biallelic *CEBPA* mutations
  - Encodes a granulocyte differentiation factor on chromosome 19
  - Germline mutation at 5’ end of gene
  - Somatic mutation at 3’ end of the other allele
    - Acquired at the time of progression to AML
- Morphologic, immunophenotypic and cytogenetic features similar to sporadic AML with *CEBPA* mutations

**Case 230** P. Khattar: 39 yo with strong family
Myeloid neoplasms with germline *DDX41* mutations

- Inherited mutations in the gene on chromosome 5 encoding the DEAD box RNA helicase DDX41
  - Major subset DDX41 mutation is biallelic (one mutation is germline)
- Prevalence is unclear – *DDX41* mutations found in 1.5% of myeloid neoplasms
- Long latency – presentation in 60s

- **CASE 318 H. Kurt:**
  - 67 year-old man who had been having slowly decreasing white blood cell and platelet counts for the last 7 years
    - Presented with **AML**, normal karyotype, no dysplasia
    - Received transplant from his brother
  - After 4 months from stem cell transplant, the patient accepted skin biopsy for further genetic testing.
    - DDX41 NM 016222.2(DDX41): c.3G>A p.M1?
Myeloid Neoplasms with Germline Predispositions AND pre-existing platelet disorders

- Germline mutations in *RUNX1* gene
  - gene on chromosome band 21q22
  - encodes one subunit of the core binding transcription factor that regulates expression of several genes essential for hematopoiesis.
  - Somatic RUNX1 mutations are associated with poor prognosis in AML/MDS

- Mild to moderate thrombocytopenia
- Functional platelet defects -> prolonged bleeding
- Increased risk of developing MDS, AML or T-ALL
### Cases with germline RUNX1 mutations

**Familial platelet disorder with predisposition to AML**

<table>
<thead>
<tr>
<th>Case Number</th>
<th>Submitter</th>
<th>Age</th>
<th>Diagnosis</th>
<th>Interesting aspects</th>
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</thead>
<tbody>
<tr>
<td>219</td>
<td>Geyer</td>
<td>37</td>
<td>Thrombocytopenia</td>
<td>Variant of unknown signficance</td>
</tr>
<tr>
<td>271</td>
<td>Mosse</td>
<td>3</td>
<td>Thrombocytopenia</td>
<td>Extensive family h/o AML</td>
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<tr>
<td>309</td>
<td>Kanagal-Shamanna</td>
<td>13</td>
<td>Thrombocytopenia</td>
<td>46,XX,inv(9)(p12q13)[20]</td>
</tr>
<tr>
<td>364</td>
<td>Reddy</td>
<td>25</td>
<td>Thrombocytopenia</td>
<td>Incidental presentation</td>
</tr>
</tbody>
</table>
Case 271- extensive family history of AML

*Diagnostic criteria for myeloid neoplasm in this setting is the same as for sporadic cases

- Presence of germline $RUNX1$ mutations does not place case into category of myeloid neoplasm with germline predisposition syndrome category

- Thrombocytopenia with germline $RUNX1$ mutation
3 y/o male with no known family history presented with fever, generalized lymphadenopathy, hepatosplenomegaly, pleural effusions, ascites, thrombocytopenia and anemia.

- WBC 17.2, Hgb 7.8, platelets 14, BUN 85, Cr 0.7, Cystatin C 3.45, ALP 108, CRP 15, ESR 118, Albumin 2, and LDH 481. He also had elevated IL-6
- RUNX1 exon4 p.G87C (c.259G>T) (VAF 35%)
  - Germline analysis confirmed that the RUNX1 mutation is present in DNA from nail, lymph node and bone marrow at 50% VAF.
MDS/AML with germline *RUNX1* mutations

- Risk of transformation to MDS/AML is estimated to be ~30-40%
- Progression to MDS/AML likely requires additional mutations
  - may account for some of the variation in penetrance of MDS/AML as well as the variable neoplasm phenotypes that develop

<table>
<thead>
<tr>
<th>Case #</th>
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<th>Age</th>
<th>Diagnosis</th>
<th>Other mutations</th>
<th>Cytogenetic</th>
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<tbody>
<tr>
<td>38</td>
<td>Chisholm</td>
<td>12</td>
<td>AML</td>
<td></td>
<td>46,XX,t(2;11)(q31;p15)[20]</td>
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<tr>
<td>339</td>
<td>Kanagal-Shamanna</td>
<td>7</td>
<td>MDS-MLD</td>
<td></td>
<td>del(5)(q31q34)</td>
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<tr>
<td>284</td>
<td>Bailey</td>
<td>32</td>
<td>AML</td>
<td>NRAS, BCOR</td>
<td>t(2;21)(q23;q22)</td>
</tr>
</tbody>
</table>
Germline mutations in ANKRD26 gene
- located on chromosome band 10p12.1
- Mutations occur within the 5' untranslated region of the gene
  - disrupt the assembly of RUNX1 and FLI1 on the ANKRD26 promoter

Case 268 Neppalli
- 43 yo with long standing history of thrombocytopenia (and family history of thrombocytopenia)
  - Normocellular marrow but with decreased megakaryocytes and frequent hypolobated forms
  - Thrombocytopenia with germline ANKRD26 mutation
Myeloid neoplasms with germline predispositions AND other organ dysfunctions

• Germline $GATA2$ mutations
• Bone marrow failure syndromes
• JMML associated with NF, Noonan’s or Noonan-like disorders
• Down syndromes
Myeloid neoplasms with germline GATA2

- Four separate syndromes
  - MonoMAC syndrome
    - monocytopenia and non-tuberculous mycobacterial infection
  - Dedritic cell, monocyte B- and NK lymphoid (DCML) deficiency with vulnerability to viral infections
  - Familial MDS/AML
  - Emberger syndrome
    - Primary lymphadema, warts, predisposition to MDS/AML

AML with germline *GATA2* mutations

<table>
<thead>
<tr>
<th>Case Number</th>
<th>Age</th>
<th>Name</th>
<th>Diagnosis</th>
<th>Other mutations</th>
<th>Cytogenetics</th>
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<tbody>
<tr>
<td>20</td>
<td>18</td>
<td>Scordino</td>
<td>AML-MRC</td>
<td>-</td>
<td>Complex karyotype</td>
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<tr>
<td>48</td>
<td>6</td>
<td>Siegele</td>
<td>AML-MRC</td>
<td>WT1, JAK2, CSF3R, KRAS</td>
<td>Monosomy 7</td>
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<td>236</td>
<td>30</td>
<td>Boyer</td>
<td>AML-MRC</td>
<td>-</td>
<td>Complex karyotype</td>
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<tr>
<td>266</td>
<td>16</td>
<td>Batdorf</td>
<td>AML-MRC</td>
<td>-</td>
<td>Normal</td>
</tr>
</tbody>
</table>
# MDS with germline \textit{GATA2} mutation

<table>
<thead>
<tr>
<th>Case Number</th>
<th>Name</th>
<th>Age</th>
<th>Diagnosis</th>
<th>Other Mutations</th>
<th>Cytogenetics</th>
</tr>
</thead>
<tbody>
<tr>
<td>105</td>
<td>Williams</td>
<td>57</td>
<td>MDS-MLD</td>
<td>-</td>
<td>Normal</td>
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<tr>
<td>138</td>
<td>Malek</td>
<td>13</td>
<td>MDS-MLD</td>
<td>-</td>
<td>Normal</td>
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<tr>
<td>157</td>
<td>Crane</td>
<td>45</td>
<td>MDS-MLD</td>
<td>-</td>
<td>Trisomy 21 &amp; 8</td>
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<tr>
<td>337</td>
<td>Balakrishna</td>
<td>31</td>
<td>MDS-MLD</td>
<td>-</td>
<td>Trisomy 8</td>
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<tr>
<td>176</td>
<td>Koo</td>
<td>10</td>
<td>MDS-EB1</td>
<td>NRAS, PTN11, SETBP1, ASXL1</td>
<td>Monosomy 7</td>
</tr>
<tr>
<td>52</td>
<td>Moore</td>
<td>22</td>
<td>MDS-EB2</td>
<td>-</td>
<td>Monosomy 7</td>
</tr>
<tr>
<td>87</td>
<td>Chiu</td>
<td>5</td>
<td>RCC</td>
<td>ASXL1</td>
<td>Deletion 7q, trisomy 8</td>
</tr>
<tr>
<td>381</td>
<td>Wang</td>
<td>17</td>
<td>RCC</td>
<td>-</td>
<td>Trisomy 8</td>
</tr>
<tr>
<td>40</td>
<td>Chisholm</td>
<td>17</td>
<td>CMML-1</td>
<td>KRAS, NF1, SETBP1, STAT3, WT1</td>
<td>Monosomy 7</td>
</tr>
</tbody>
</table>
Other disorders with germline *GATA2* mutations

- Case 258 Hussein: 62 yo with FUO and 10 year history of lymphopenia and monocytopenia of unclear etiology

- HLH in a patient with bone marrow deficiency (MonoMAC) with germline *GATA2*
Myeloid Neoplasms with Germline Predispositions and organ dysfunction

Noonan’s syndrome

- relatively common (1/2000 births) developmental disorder
  - Characteristic appearance and congenital heart defects
  - Associated with mutations in genes that are part of the RAS/RAF/MEK/ERK signal transduction pathway
    - Variants in PTPN11 (50%), SOS1, RAF1, KRAS, NRAS, BRAF, MAPK1
- Increased risk of malignancy
  - JMML, ALL, rhabdomyosarcoma, neuroblastoma, glioma

Case 99 Knez – B lymphoblastic leukemia in 19 months old with Noonan’s syndrome and SHOC2 gene mutation
JMML

**Mandatory**
- Monocyte count > 1x10⁹
- Blast % in PB and BM <20%
- Splenomegaly
- Absence of BCR-ABL

**Oncogenics**
- Somatic mutation in PTPN11, KRAS or NRAS
- Clinical diagnosis of NF-1 or germline NF1
- Germline CBL mutation and loss of heterozygosity of CBL

If negative for oncogenics, 2 need to be met

**Case 292 Nguyen**: 49 day old with dysmorphic features c/w Noonan’s presents with splenomegaly and leukocytosis (50-97k/uL)
- PTPN11 c.218C>T (p.Thr73Ile) missense mutation

- Monosomy 7
- HbF increased for age
- Myeloid precursors in PB
- Spontaneous growth or GM-CSF hypersensitivity
- Hyperphosphorylation of STAT5
**Case 320 Curry**: Newborn with prenatal diagnosis of Noonan’s
- Persistent thrombocytopenia, leukocytosis, and borderline high Hgb F, but no hepatosplenomegaly
- De novo heterozygous pathogenic variant in the PTPN11 gene (p.S502L).
- Resolved at one year follow up
  - Transient Myeloproliferative Disorder in a patient with germline *PTPN11* (Noonan’s syndrome)

**Case 55 Bayerl**: 4 month-old asymptomatic girl was found to have splenomegaly, hepatomegaly, leukocytosis and anemia
- PB 9.5% blasts, *BM* 29% blast/blast equivalents
- Normal karyotype, *NF1* c.1139T>G (90%)
- After diagnosis of her leukemia, she was found to have >6 café au lait macules.
  - AML-NOS with germline *NF1* mutation
AML with other inherited conditions

<table>
<thead>
<tr>
<th>Case #</th>
<th>Submitter</th>
<th>Age</th>
<th>Diagnosis</th>
<th>Mutation</th>
<th>Cytogenetic</th>
<th>Syndrome</th>
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<tbody>
<tr>
<td>253</td>
<td>Leeman-Neil</td>
<td>23</td>
<td>AML-MRC</td>
<td>WT1 &amp; NF1 (somatic) BLM (germ)</td>
<td>Complex Karyotype</td>
<td>Bloom</td>
</tr>
<tr>
<td>225</td>
<td>Batdorf</td>
<td>18 months</td>
<td>Therapy-related AML</td>
<td>Germline PTCH TGRB1 microdeletion of unknown significance</td>
<td>t(8;16)(p11:2;p13.3);KAT6A-CREBBP</td>
<td>-</td>
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<tr>
<td>264</td>
<td>Leeman-Neil</td>
<td>18</td>
<td>AML-MRC</td>
<td>IDH1, NRAS, WT1</td>
<td>Complex</td>
<td>Mafucci</td>
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<td>234</td>
<td>Meyerson</td>
<td>53</td>
<td>AML</td>
<td>RUNX1, STAG2</td>
<td>Inv(3) and germline t(8;21)</td>
<td>-</td>
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MDS with other inherited syndromes

<table>
<thead>
<tr>
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<th>Diagnosis</th>
<th>Mutation</th>
<th>Cytogenetic</th>
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</thead>
<tbody>
<tr>
<td>170</td>
<td>Gong</td>
<td>11</td>
<td>MDS-EB2 &amp; LCH</td>
<td>RBM8A</td>
<td>Normal</td>
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<tr>
<td>196</td>
<td>Malek</td>
<td>11</td>
<td>MDS-MLD</td>
<td>G6PC3</td>
<td>Normal</td>
</tr>
<tr>
<td>80</td>
<td>Klco</td>
<td>3-4 and 14 months</td>
<td>RCC MIRAGE* , MDS/MPN, unclassifiable MIRAGE*</td>
<td>SAMD9 x3</td>
<td>Monosomy 7x3</td>
</tr>
<tr>
<td>273</td>
<td>Judd</td>
<td>5 months</td>
<td></td>
<td>SAMD9</td>
<td>Monosomy 7</td>
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</tbody>
</table>

*MIRAGE myelodysplasia, infections, restriction of growth, adrenal hypoplasia, enteropathy

TAR syndrome
## Lymphoid neoplasms

<table>
<thead>
<tr>
<th>Case</th>
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<th>Age</th>
<th>Diagnosis</th>
<th>Cytogenetics</th>
<th>Mutation</th>
</tr>
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<tbody>
<tr>
<td>101</td>
<td>Raciti</td>
<td>18 months</td>
<td>B lymphoblastic leukemia</td>
<td>46,XX,i(9)(q10)[2]/46,XX[21]</td>
<td>Heterozygous PAX5 mutation</td>
</tr>
<tr>
<td>194</td>
<td>Baker</td>
<td>1</td>
<td>B lymphoblastic leukemia</td>
<td>Monosomy 7</td>
<td>ELANE mutation</td>
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<tr>
<td>346</td>
<td>Pullarkat</td>
<td>19</td>
<td>Classical Hodgkin lymphoma</td>
<td>Normal</td>
<td>CSF3R (variant of undetermined significance)</td>
</tr>
<tr>
<td>342</td>
<td>Wake</td>
<td>40</td>
<td>T-LGL and PRCA</td>
<td>Normal</td>
<td>CTLA4</td>
</tr>
</tbody>
</table>
T-cell LGL and pure red cell aplasia  
case 342

- 40 yr old with PMH of recurrent respiratory and GI infections presented lymphocytic colitis
- Hypocellular bone marrow with diffuse, interstitial pattern and multiple non-paratrabecular lymphoid aggregates
- Erythroid precursors were absent
- Flow identified an abnormal expanded gamma delta T-cell population expressing CD3, CD8, CD57, CD2, CD7, TCR \( \gamma \delta \), and heterogeneous CD5
- Tcell clonality was positive
- Mutation in the gene \textbf{CTLA4 (151C>T; R51X)}, confirmed by Sanger sequencing
  - Also present in daughter

CD8 stain
### Incidental findings

<table>
<thead>
<tr>
<th>Case</th>
<th>Submitter</th>
<th>Age</th>
<th>Diagnosis</th>
<th>Genetic findings</th>
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<tr>
<td>333</td>
<td>Coberly</td>
<td>56</td>
<td>Histiocytic sarcoma</td>
<td>46,XY, t(12;18)(q15;q21)[20] Constitutional</td>
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<td>97</td>
<td>Raciti</td>
<td>6</td>
<td></td>
<td>MPL mutation MPNs have not developed</td>
</tr>
<tr>
<td>209</td>
<td>Velu</td>
<td>38</td>
<td>Follicular lymphoma</td>
<td>Li-Fraumeni syndrome Not reported before</td>
</tr>
</tbody>
</table>
Summary from Session 1

- 51 cases submitted in this category
- Most common cases submitted included germline mutations in \textit{GATA2} \((n=16)\) and \textit{RUNX1} \((n=10)\) genes
- Mostly myeloid neoplasms (MDS/AML) \((n=42)\) but lymphoid neoplasms (including B-ALL, FL, T-cell LGL) were also submitted
- No specific clinical features
  - germline mutations are associated with non-neoplastic hematological disorders, organ dysfunction, or inherited syndromic disorders
Myeloid neoplasms with germline predisposition

• Morphology of the neoplasm depends on its subtype
• Presence of a genetic predisposition does not in itself place a case into the category of a myeloid neoplasm
• Diagnostic criteria for the germline predisposition disorders are the same as those for sporadic cases
  – diagnosis of MDS may be challenging in some cases
    • Early dysplastic features may not progress to MDS or AML for decades
    • Increased blasts, increasing marrow cellularity, increasing cytopenias, and/or the presence of additional cytogenetic or molecular genetic abnormalities
Rise of NGS testing in clinical setting

• Somatic or germline mutations?
• Standard sequencing cannot distinguish, but could give clues...
  – Near heterozygous (40-60%) or near homozygous (>90%) allelic frequency
  – ‘Threshold’ allelic frequency to warrant germline testing is not standarized
  – Multiple mutations in CEBPA, RUNX1 or GATA2 genes
• Counseling and germline testing are next steps
Germline testing

• Cultured skin fibroblasts are preferred tissue
  – Can take 3-6 weeks for cultures to yield sufficient DNA
  – DNA from epithelial cells of hair follicles is more readily available

• Buccal swabs or saliva are frequently contaminated with hematopoietic cells and should be avoided

• Other sources include nail clippings or mesenchymal cells from bone marrow aspirate smears
Scenarios when genetic testing is advised in newly diagnosed patients with MDS/AML

• Somatic testing identified a mutation associated with germline predisposition syndrome (CEBPA, GATA2, RUNX1)
• Hematologic or cytogenetic characteristic of MDS/AML suggestive of germline predisposition
• Genetic syndrome known to predispose to cancer
• Previous malignancy, family history cancer
• Cytopenias, immune deficiency, atypical infections, lymphadema or organ-system manifestation
Thank you!