Session 7
Summary

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MOLECULAR GENETICS OF HEMATOPOIETIC NEOPLASMS
Cases according to 2016 WHO classification

- Acute myeloid leukemia: 26
  - AML with recurrent genetic abnormalities: 9
  - AML-MRC: 4
  - AML, NOS: 7
- Acute leukemia of ambiguous lineage: 6 (MPAL, B/myeloid 4)
- Therapy-related myeloid and lymphoid neoplasms: 6
- B lymphoblastic leukemia/lymphoma: 4
- T lymphoblastic leukemia/lymphoma: 2
- Transformation (blast phase) of chronic myeloid neoplasms: 3
Session 7 categories

1. De novo acute leukemias and therapy-related myeloid/lymphoid neoplasms with unusual genetic features
2. Genetic abnormalities indicating residual disease or underlying hematopoietic neoplasm
3. Clonal relationship, clonal evolution and disease heterogeneity
4. Treatment: therapeutic targets and response patterns
5. Prognostic implications
6. Diagnostic dilemmas
De novo AML and therapy-related lymphoid neoplasms with variant or novel KMT2A rearrangements

Case 136  El Hussein
AML, NOS (acute monocytic leukemia, with variant KMT2A translocation)
11M; facial nerve palsy, periorbital bruising, testicular mass, anemia, thrombocytopenia

46, Y, t(X;11) (q26;q23)[17]/46, XY[3]
KMT2A FISH in 98% nuclei (BAP)
Postulated partner: CT45A2

Cerveira N et al. BMC Cancer 2010;10:518
De novo AML and therapy-related lymphoid neoplasms with variant or novel KMT2A rearrangements

Case 302 Paessler
Therapy-related B-ALL with KMT2A-MALM rearrangement
10F; numerous circulating blasts, previous history of Ewing sarcoma

46,XX,inv(11)(q21q23),der(18)t(11;18)(q14.2;q22.2)inv(11)[20].ish
inv(11)(5'MLL+,3'MLL+),der(18)(5'MLL+,3'MLL+)/46,XX[1]/Confirmed by FISH &
ArcherDx NRAS c.181C>A

SNP studies of Ewing sarcoma not suggestive of an underlying cancer predisposition (no loss of p53 or other tumor suppressors)

Case 0306 Mariani
Therapy-related T-ALL with KMT2A-MALM rearrangement
5M; B-LL, BCR-ABL1+ at 2 years of age, currently mediastinal mass and circulating blasts

46,XY, inv(11)(q21q23)[14]/46,XY,idem,+7,+18[4]/46,XY[2]

Menu E et al. BMC Cancer 2017;17:363
De novo AML with JAK2 V617F mutations

Case 96 Gridley
AML-MRC
68M, back pain, B-symptoms, hepatosplenomegaly, circulating blasts, anemia, mild thrombocytopenia; no prior hematologic history

43~46,XY,-4,add(5)(q13),add(7)(q22),add(10)(q22),-13,add(16)(q11.2),-17,-19,-20,+2~5mar[cp19] /46,XY[1]

JAK2 c.1849G>T, DNMT3A c.2644C>T

Case 57 Aynardi
AML, NOS (acute myelomonocytic leukemia, with JAK2 mutation)

Bullinger L et al. JCO 2017;35:934
Acute myeloid leukemias with genetic abnormalities typically seen in lymphoid neoplasms

**Case 37  Xu**
AML-MRC
49F, pancytopenia, blasts in PB
49,XX,+1,
der(1;12)(q10;q10),+8,+8,+mar[18]
BRAF p.V600E, NPM1 W288fs

**Case 116  Sadigh**
AML with t(8;21)(q22;q22.1);RUNX1-RUNX1T1 presenting as myeloid sarcoma (with FBXW7 mutation)
36M, left back pain, paraspinal mass
FISH: t(8;21)(q22;q21)/ RUNX1-RUNX1T1 and FBXW7 c.1394G>A
Acute myeloid leukemias with genetic abnormalities typically seen in lymphoid neoplasms

Case 224 Teruya-Feldstein
AML, NOS (acute monocytic leukemia, with ALK rearrangement)
58M, leukocytosis with blasts and monocytosis, anemia, thrombocytopenia

t(2;2)(p23;q12) [20] (confirmed by metaphase FISH with break-apart probe)

Negative for FLT3-ITD, NPM1, CEBPA, CKIT mutations

Takeoka K et al. Cancer Genet. 2015;208:85
Lim JH et al. Cancer Genet. 2014;207:40
Lymphoblastic leukemias/lymphomas with genetic lesions typically seen in myeloid neoplasms

**Case 66 Devins**

**B-ALL, NOS (with \textit{U2AF1} mutation)**

29M, dyspnea and headaches, blasts in PB, mild anemia and thrombocytopenia

Normal karyotype; \textit{U2AF1} c.101C>T

**Case 367 Zhang**

**Recurrent B-ALL/LBL, NOS (with mutated \textit{ATRX})**

20M, h/o B-LL with atypical BCR/ABL1 fusion with recent recurrence

gain of 9q34 (\textit{ABL1}), loss of 9p21 (\textit{CDKN2A})

\textit{ATRX} c.5579A>G

Spinella JF et al. Oncotarget 2016;7:65485
Lindqvist CM et al. Oncotarget 2016;7:64071
Schenkel et al. Epigenetics & Chromatin 2017;10:10
Case 83 Woodham
Therapy-related myeloid neoplasm with features of MPAL, B/myeloid
69M, h/o neuroendocrine carcinoma, s/p chemotherapy/radiation, circulating blasts
46,XY,t(16;21)(q24;q22)[5]/46,sl,del(2)(q24q32),del(7)(q31.2)[2]/46,XY[3]

RUNX1-CBFA2T3; rare, seen primarily in t-AML

Case 232 Kuzu
T lymphoblastic leukemia/lymphoma (with BCR-ABL1 rearrangement)
57M, lymphadenopathy
Cytogenetics and FISH NA; RT-PCR positive for BCR-ABL1 p210

Park IJ et al. Cancer Genetics Cytogenetics 2010;196:105
Case 265  Yuan
B-ALL/LBL, NOS (with MYC rearrangement)
56F, numerous blasts in PB, generalized lymphadenopathy, splenomegaly

46,XX,dup(1)(q12q42)x2,t(8;14)(q24.1;q32),inv(9)(p11q13)[17]/46,XX,inv(9)(p11q13)[3]

Case 348  Chen
MPAL, B/myeloid, NOS (with EWSR1 rearrangement)
10 month old F, pallor, bruising, pancytopenia

46,XX,t(2;22)(q34;q12),add(4)(p15.2)[20]

EWSR1 (22q12) rearrangement confirmed by FISH

Lanocha AA et al. Blood 2017;129: 393
Case 69  Devins
AML with mutated \textit{NPM1}
68M, circulating blasts, anemia and thrombocytopenia

\textit{NPM1, KIT, DNMT3A} and \textit{TET2} at diagnosis; \textit{DNMT3A} and \textit{TET2} persistent on day 31 (blasts 0\%) in unchanged allele frequency; subsequent relapse with the same clone

Case 73  Shanmugam
Leukemia cutis: cutaneous involvement by the patient's known myeloid neoplasm (possibly CMML), with Langerhans cell differentiation
56M, h/o AML, possible underlying CMML, presented with cutaneous papules

\textit{ASXL1, IDH1, KRAS, NRASx2, RUNX1, SRSF1}, seen previously in AML, post-therapy BM suspicious for CMML and in skin
Genetic abnormalities indicating residual disease or prior underlying neoplasm

Case 294 Chen
CML, BCR-ABL1+, in blast phase [with inv(16)(p13.1q22)]
24F, marked leukocytosis with numerous blasts, eosinophilia, basophilia and anemia

46,XX,t(9;22)(q24;q11.2),inv(16)(p13.1q22)[20]

FISH: Positive for BCR-ABL1 fusion and CBFB rearrangement

Interphase FISH confirmed BCR-ABL1 positive neutrophils, and the presence of BCR-ABL1 clone without inv(16)
Clonal relationship, clonal evolution and disease heterogeneity

**Case 56  Xu**  
**Therapy-related CMML-2**  
58F, h/o B-LL with normal karyotype and MLL deletion, developed pancytopenia with monocytosis  
Normal karyotype, similar deletion of *KMT2A* gene suggests common clonal origin

**Case 81  Al-Ghamdi**  
**ET in blast crisis (with *BCR-ABL1* rearrangement)**  
70M, 17 year h/o ET, JAK2+, current circulating blasts  
46,XY,t(9;22)(q34;q11.2)[20]

**Case 94  Snider**  
**AML with mutated *RUNX1* (with cryptic *NUP214-ABL1* rearrangement)**
Clonal relationship, clonal evolution and disease heterogeneity

**Case 155 Crane**

**Therapy related-AML**

38F, h/o breast carcinoma, treated with chemotherapy and radiation, *BRCA1*+, t-AML, s/p SCT, developed recurrent AML refractory to treatment

Fluctuating *FLT3*, *STAG2* and *CSF3R* (VUS) mutations. *CSF3R* variant confirmed to be a germline mutation of donor origin

**Case 184 Yin**

**AML, NOS (AML with maturation) with clonal evolution upon progression**

61M, pancytopenia; recurrent AML, underwent SCT

Stepwise acquisition of new mutations and clone expansion including *FLT3* and *P53*, both associated with inferior survival
Clonal relationship, clonal evolution and disease heterogeneity

**Case 187** Al-Ghamdi
Acute myeloid leukemia with t(8;21)(q22;q22.1);RUNX1-RUNX1T1 (and subclonal BCR-ABL1)
39M, flu-like symptoms for 2 weeks and circulating blasts
Late acquisition of BCR-ABL1 in a course of AML is rare and is associated with poor outcome

**Case 240** Kaygusuz
1. AML with mutated NPM1. 2. MPN-U
32M, diagnosed with AML and developed thrombocytosis on day 28 of treatment
Initially, NPM1 mutation, after therapy developed JAK2 V617F mutation at increasing VAF
Clonal relationship, clonal evolution and disease heterogeneity

**Case 279  Naeini**
AML with t(16;16)(p13.1;q22); CBFB-MYH11 (with JAK2 mutations at evolution)
30F, no prior hematologic history, presented with acute leukemia

At initial diagnosis FLT3-ITD and FLT3-TKD, subsequent: JAK2 V617F, JAK2 Exon 12 and WT1

**Case 285  Bogusz**
AML, NOS (acute monoblastic leukemia, with multiple mutations in RAS pathway and multiple WT1 mutations)
75F, presented with leukocytosis and concern for MPN; 2 weeks later diagnosed with AML

FLT3, KRAS, NRAS, 5 different WT1 mutations, fluctuating over disease course

**Case 317  Rangan**
B-ALL/LBL with t(9;22)(q34;q11.2); BCR-ABL1 (and BCL2 rearrangement)
59F, leukocytosis with circulating blasts, anemia, thrombocytopenia; prior h/o RA treated with etanercept and methotrexate
Clonal relationship, clonal evolution and disease heterogeneity

Case 297  Zhang
Therapy-related AML and BPDCN
54M, h/o seminoma and t-MDS with trisomy 8 and monosomy 7, progression to t-AML

FISH MDS deletion of 7q or -7 in 97.5% nuclei

*TET2*, c.2677G>A, VAF 50.78%; and *ZRSR2* c.827+1G>A, VAF 82.66%
Therapeutic targets

**Case 177 Mahon**
AML with *BCR-ABL1* (and *KMT2A* rearrangement)
47M, referred for treatment from an outside institution
FISH: positive *BCR-ABL1* rearrangement and MLL gene rearrangement

**Case 329 Zhou**
AML, NOS (with *CSF3R* mutation)
69F, anemia, neutropenia, frequent blasts in PB
*CSF3R* (T640N), *TET2* (C1193Y), *TET2* (Q622Rfs*17)

**Case 301 Jain**
AML with mutated NPM1
68F, shortness of breath, leukocytosis, macrocytic anemia, thrombocytopenia
Normal karyotype, mutations: *DNMT3A*, *IDH1*, *NPM1*, *PTPN11*, *RUNX1*
Case 144 Bhattacharyya
Acute myeloid leukemia with mutated \textit{NPM1}

Case 217 Goyal
AML, NOS (AML with maturation) with differentiation
78M, h/o AML, M6 with mutated IDH2

Karyotype pre- and post-treatment: 47,XY,+10[20]
Post-treatment: \textit{IDH2} c.515G>A, VAF 39%, \textit{DNMT3A} c.1227G>A, VAF 41%
Prognostic implications

**Case 357 Parilla**
AML-MRC [with t(8;16)(p11.2;p13.3); *KAT6A-CREBBP*, arising from prior CMML]
80M, MGUS with progression to MM, persistent monocytosis and dyspoiesis, progression to AML
*TET2, SRSF2, SETBP1, ASXL1*

**SH2017-0148**
AML-MRC [with t(1;16;8)(q21;p13;p11); *KAT6A-CREBBP*

**Case 252 El Hussein**
t-MDS/AML [with t(1;3)(p36;q21)]
88M, h/o NHL, chemotherapy, pancytopenia and abdominal pain
Diagnostic dilemma

**Case 30, O’Malley**
Acute leukemia of ambiguous lineage vs. BPDCN (with *MYC* rearrangement)
71M, colon cancer, chemotherapy in 1999, current leukemic presentation, no other lesions reported

Complex karyotype, MYC rearrangement (unknown partner)

**Case 165 Teruya-Feldstein**
First biopsy: T-ALL/LBL
Second biopsy: Blastic undifferentiated neoplasm, not definitively classifiable
23M, HIV+, developed new tender lymphadenopathy
Case 243 Yuksel
MPAL, B/myeloid, NOS
68M, cytopenias, hepatosplenomegaly

IHC: positive CD34, MPO, CD20, CD79a, PAX5, TDT, BOB1 and weak CD19

FC BM: positive HLA_DR, CD19, CD10, CD34, CD38, CD24, sCD22, cCD79a, TDT, CD20 and CD58, partial MPO and CD123

Complex karyotype

SH2017-0119 Frederiksen
B-ALL, BCR-ABL1-like vs. MPAL, B/myeloid
Conclusions
Classification and nomenclature: What to prioritize?

1. Therapy-related MDS/AML

2. AML with classic recurrent genetic abnormalities

3. AML-MRC (complex karyotype or originating from preexisting myeloid neoplasm)
   - if recurrent cytogenetic lesion-mention it
   - morphologic dysplasia does not supersede recurrent genetic lesions, but act as a category in itself in the absence of these lesions

4. AML with mutated NPM1, biallelic CEBPA, RUNX1

5. AML-MRC defined by morphologic dysplasia

6. AML, NOS
   - In myeloid sarcoma-include AML type and myeloid sarcoma as presentation in final diagnosis, e.g. AML with t(8;21)(q22;q22.1);RUNX1-RUNX1T1 presenting as myeloid sarcoma
   - Cases of myeloid sarcoma without marrow involvement should be worked-up as acute leukemia (karyotyping, FISH, molecular) and classified as such
Diagnosing MPAL may be challenging in select cases

• Diagnosis is the least challenging in cases with 2 separate populations, each fulfilling criteria for lymphoid or myeloid leukemia

• Most of the true mixed phenotype acute leukemias show heterogeneity in the expression of multiple markers (e.g. multiple myeloid and lymphoid markers are simultaneously positive)

• **Area of controversy:** typical B-ALL immunophenotype positive for only one myeloid marker-myeloperoxidase
Residual disease, underlying hematopoietic neoplasm, clonal relationship and clonal evolution

• Cannot underestimate patient history including prior CBCs and review of original diagnostic slides
  Recommendations of ASH/CAP, NCCN and ELN

• Looking beyond blast population: value of interphase FISH to identify unrecognized underlying CML in cases in blast crisis

• In AML with *BCR-ABL1*: review molecular panels for abnormalities of genes which can support a diagnosis of de novo Ph+ AML (deletion of *IGH, TCR, IKZ, CDNK2A*)

• Testing sequential samples with conventional karyotyping, FISH and molecular genetic studies may be valuable to confirm clonal relationships

• Repeating molecular studies may reveal clonal evolution and identify subclones with therapeutic targets
Therapeutic targets

• Targeted therapy: Which genetic abnormalities should be tested?

• Patterns of response to targeted therapy: Reconciliation of morphologic findings and results of cytogenetic/molecular studies

Dohner et al Blood 2017;129:424