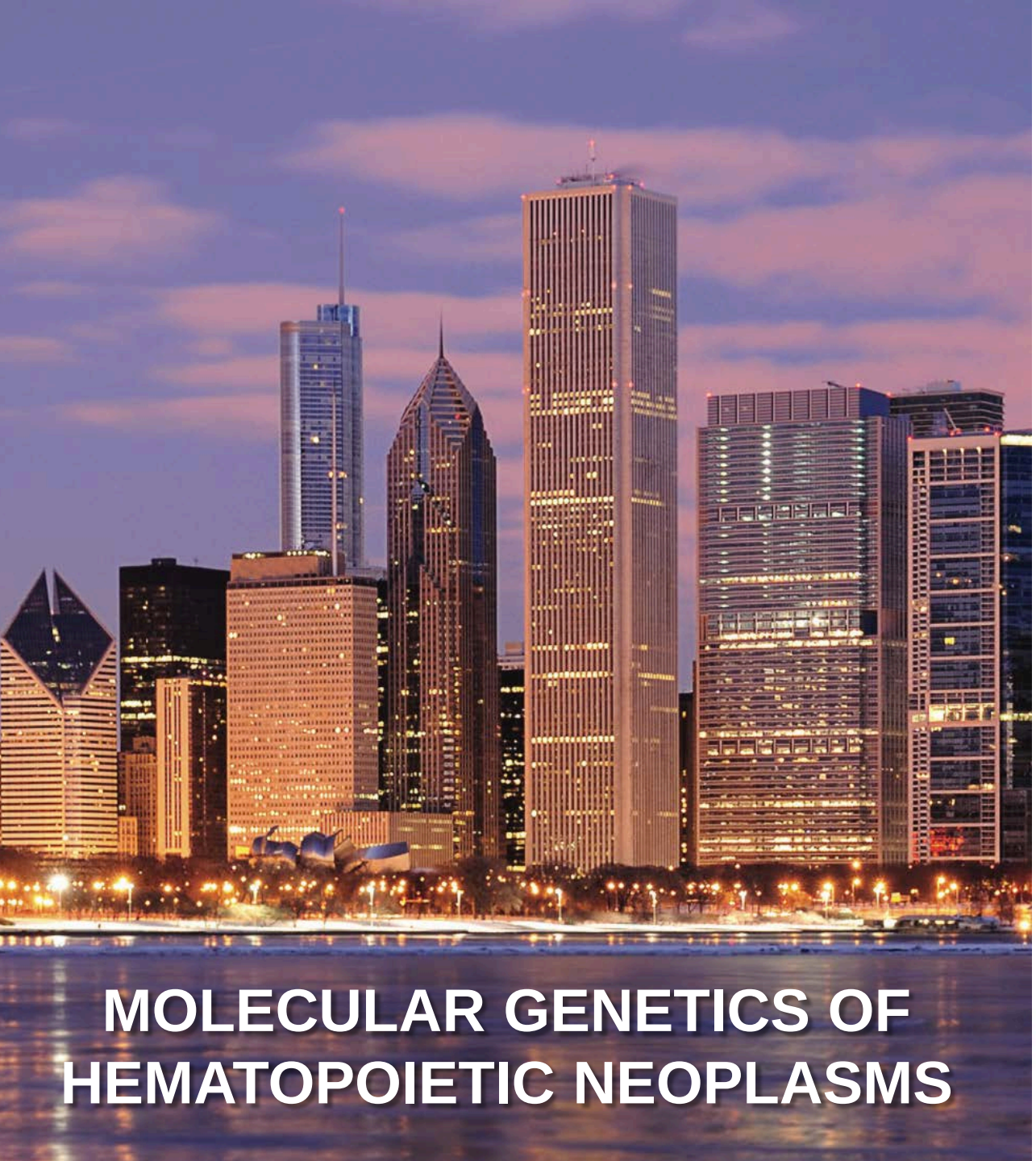

Society for Hematopathology
www.socforheme.org

Session 7 Summary

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European Association for Haematopathology
www.haematopathology.org

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

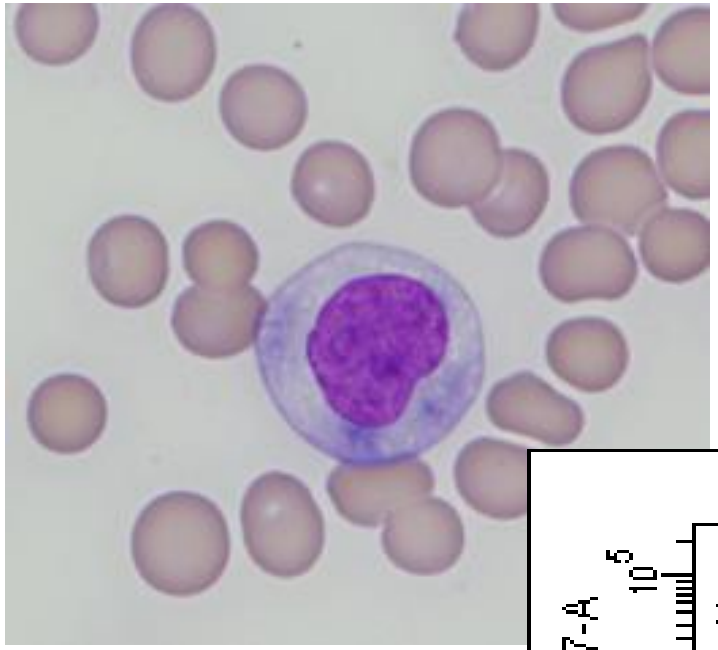
1. De novo acute leukemias and therapy-related myeloid/lymphoid neoplasms with unusual genetic features
2. Genetic abnormalities indicating residual disease or underlying hematopoietic neoplasm
3. Clonal relationship, clonal evolution and disease heterogeneity
4. Treatment: therapeutic targets and response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

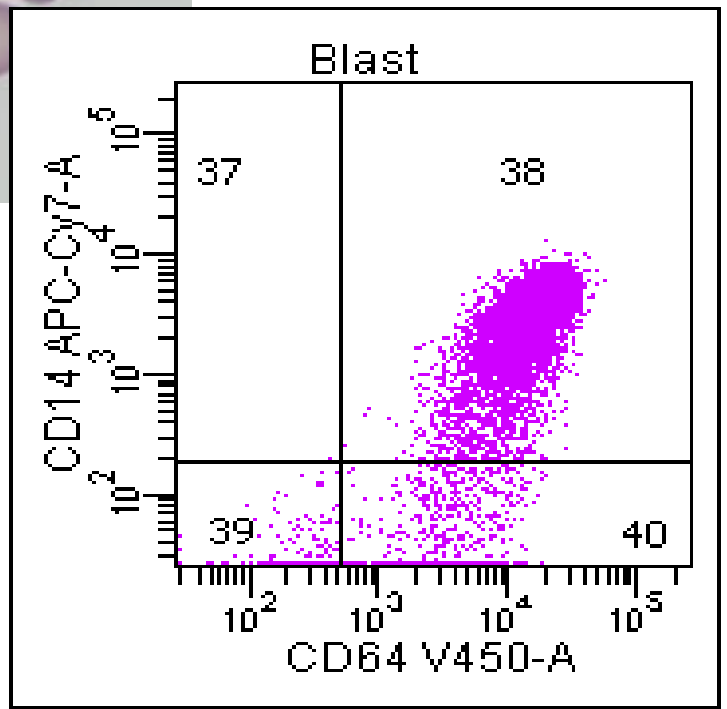
De novo AML and therapy-related lymphoid neoplasms with variant or novel *KMT2A* rearrangements

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
Metzler M et al. Leukemia 2008;22:1807

De novo AML with *JAK2* V617F mutations

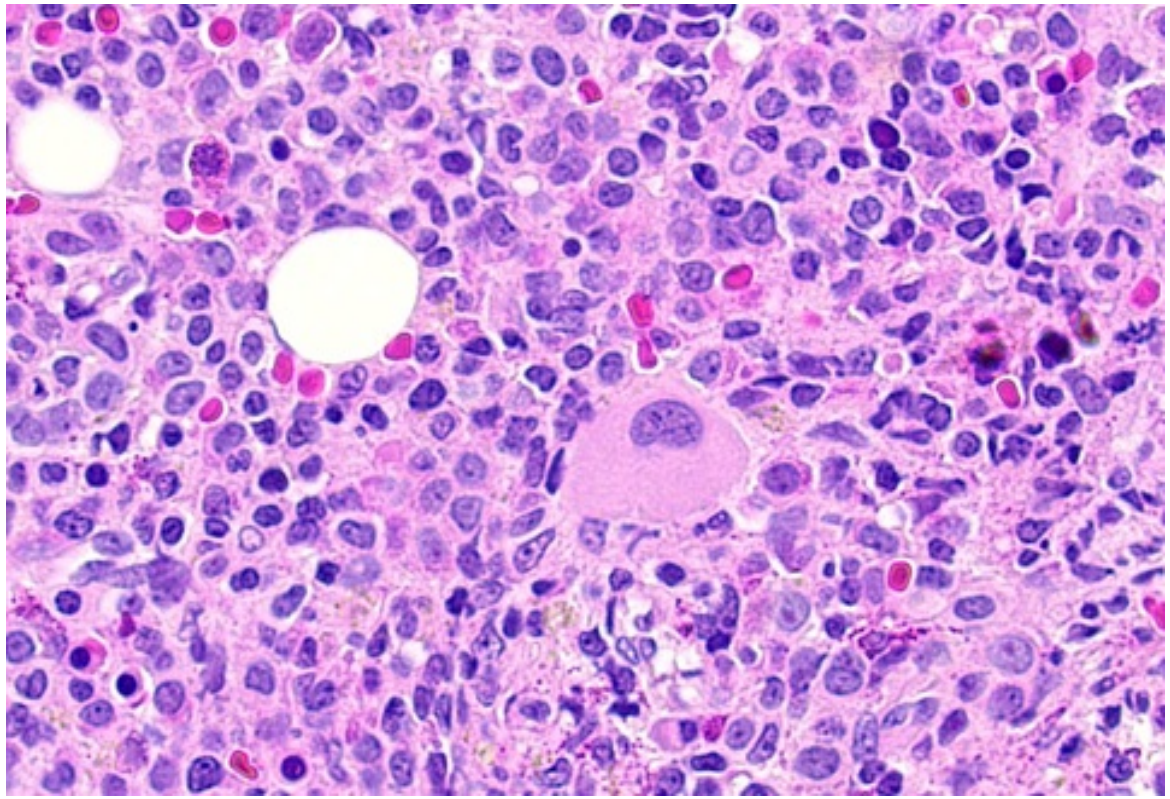
Case 96 Gridley

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

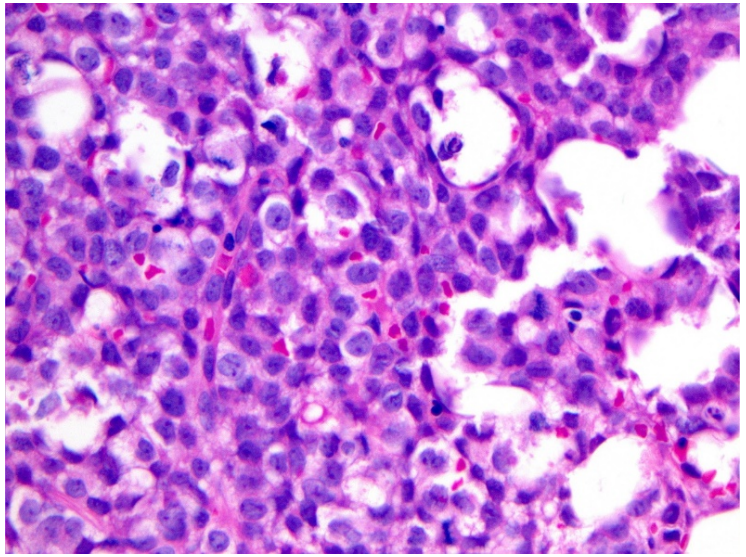
Case 37 Xu

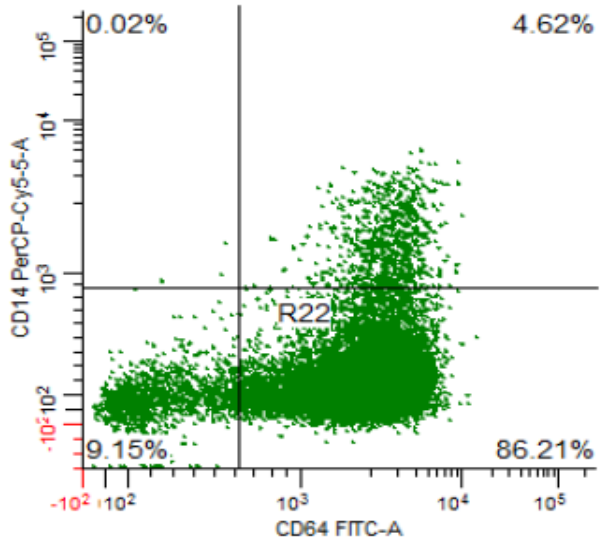
AML-MRC

49F, pancytopenia, blasts in PB

49,XX,+1,
der(1;12)(q10;q10),+8,+8,+mar[18]

BRAF p.V600E, *NPM1* W288fs



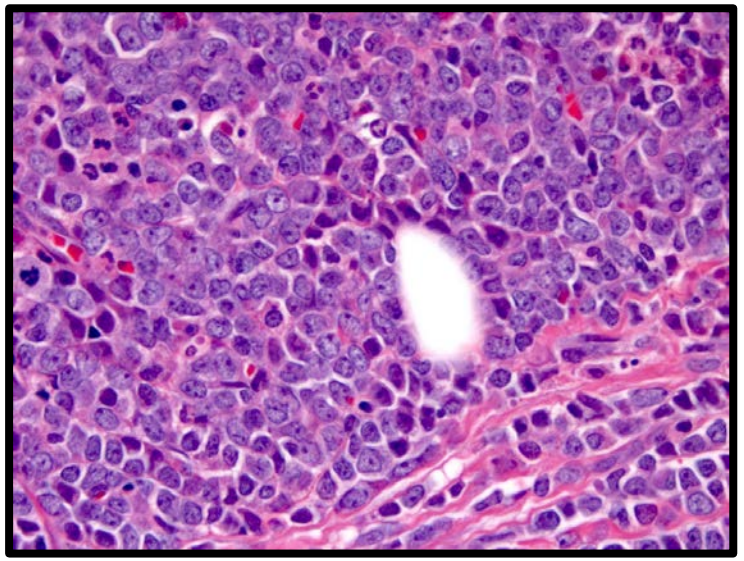


Case 116 Sadigh

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

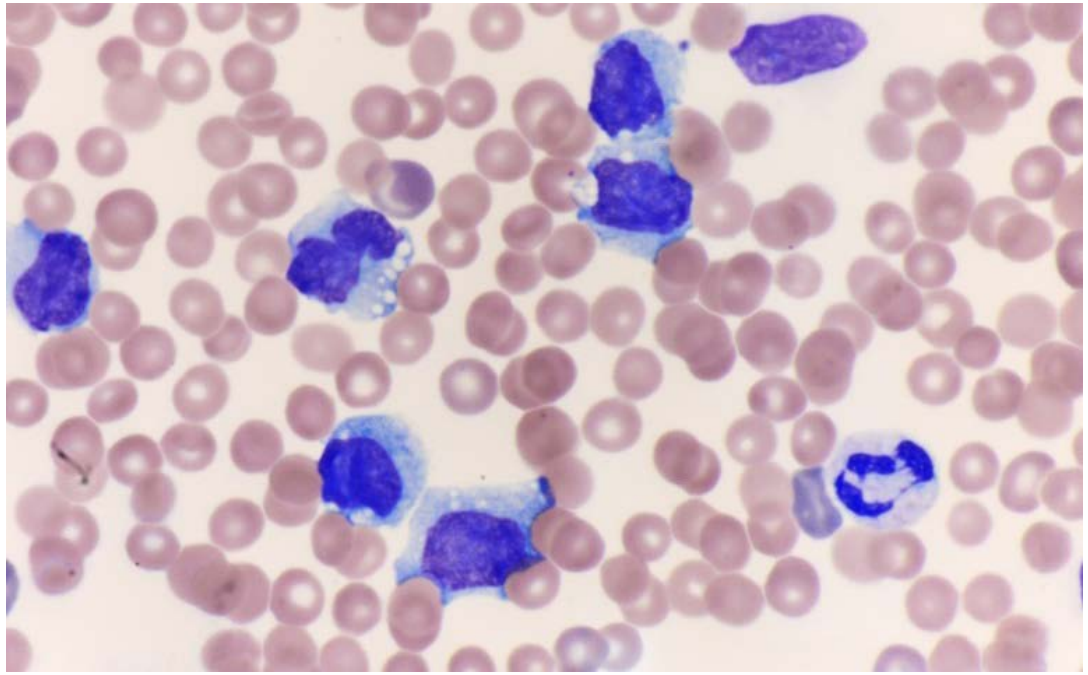


Acute myeloid leukemias with genetic abnormalities typically seen in lymphoid neoplasms

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

Hayashi A et al. Blood Cancer J 2016;6:e456
Takeoka K et al. Cancer Genet. 2015;208:85
Lim JH et al. Cancer Genet. 2014;207:40

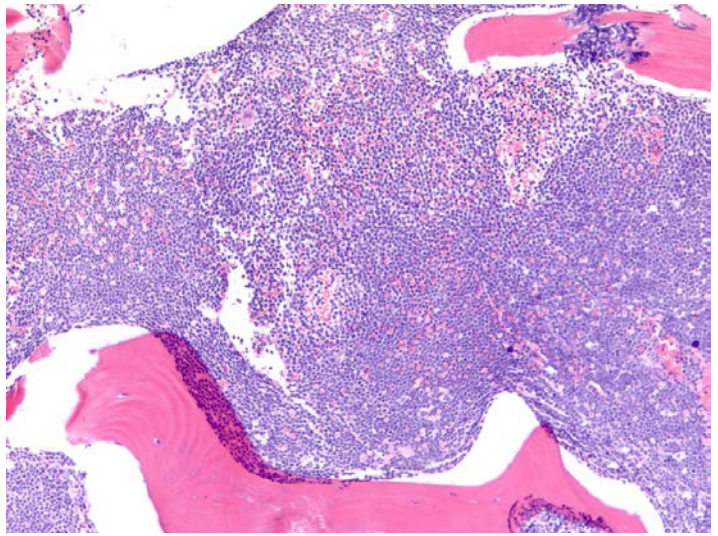
Lymphoblastic leukemias/lymphomas with genetic lesions typically seen in myeloid neoplasms

Case 66 Devins

B-ALL, NOS (with *U2AF1* mutation)

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

Normal karyotype; *U2AF1* c.101C>T

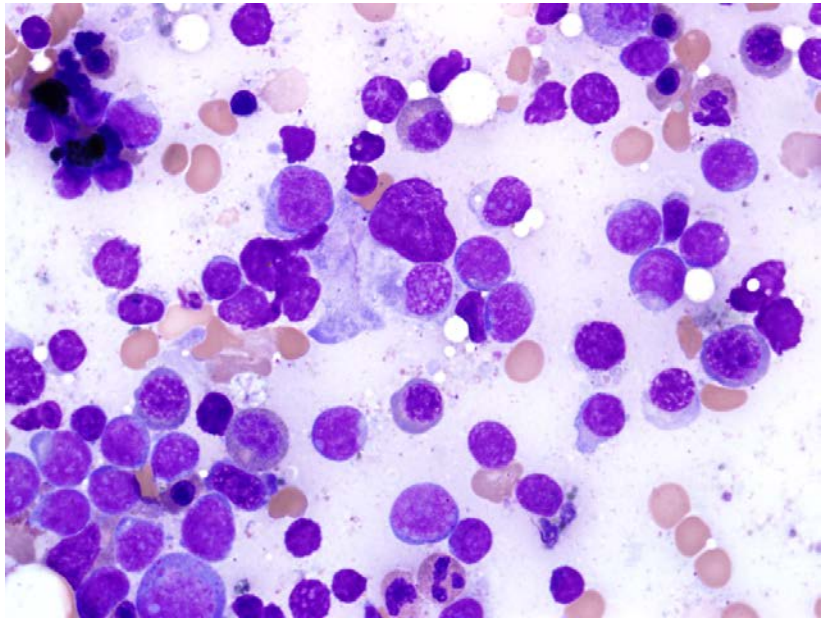


Case 367 Zhang

Recurrent B-ALL/LBL, NOS (with mutated *ATRX*)

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

gain of 9q34 (*ABL1*), loss of 9p21 (*CDKN2A*)
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

Miscellanea

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]

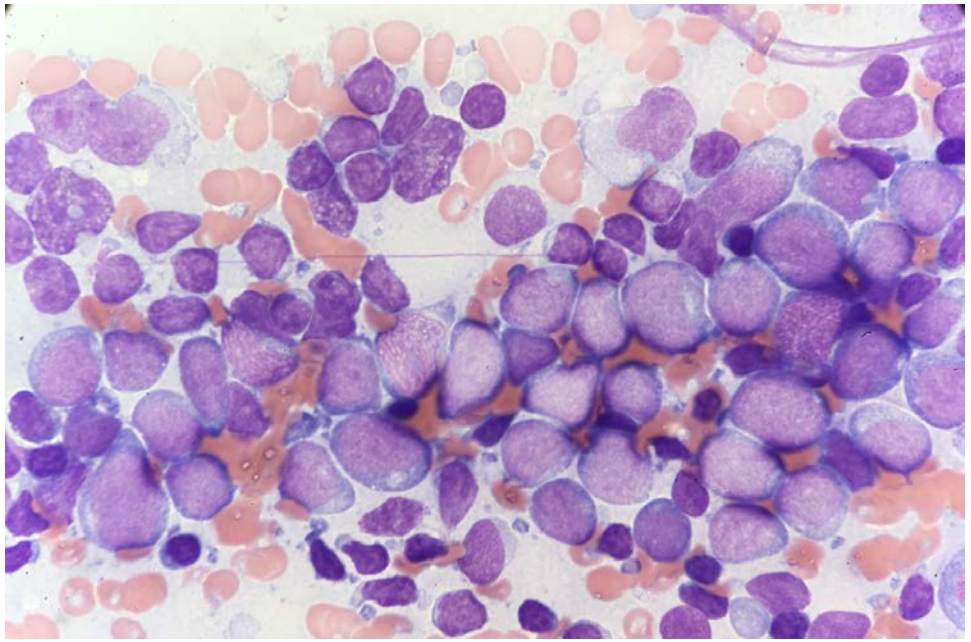
RUNX1-CBFA2T3; rare, seen primarily in t-AML

Case 232 Kuzu

T lymphoblastic leukemia/lymphoma (with BCR-ABL1 rearrangement)

57M, lymphadenopathy

Cytogenetics and FISH NA; RT-PCR positive for BCR-ABL1 p210



Ottone T et al. Genes Chromosomes Cancer 2009;48:213
Park IJ et al. Cancer Genetics Cytogenetics 2010;196:105
Raanani P et al. Acta Haematol 2005;113:181
Kamoda Y et al. Acta Haematol 2016;136:157-166

Miscellanea

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]

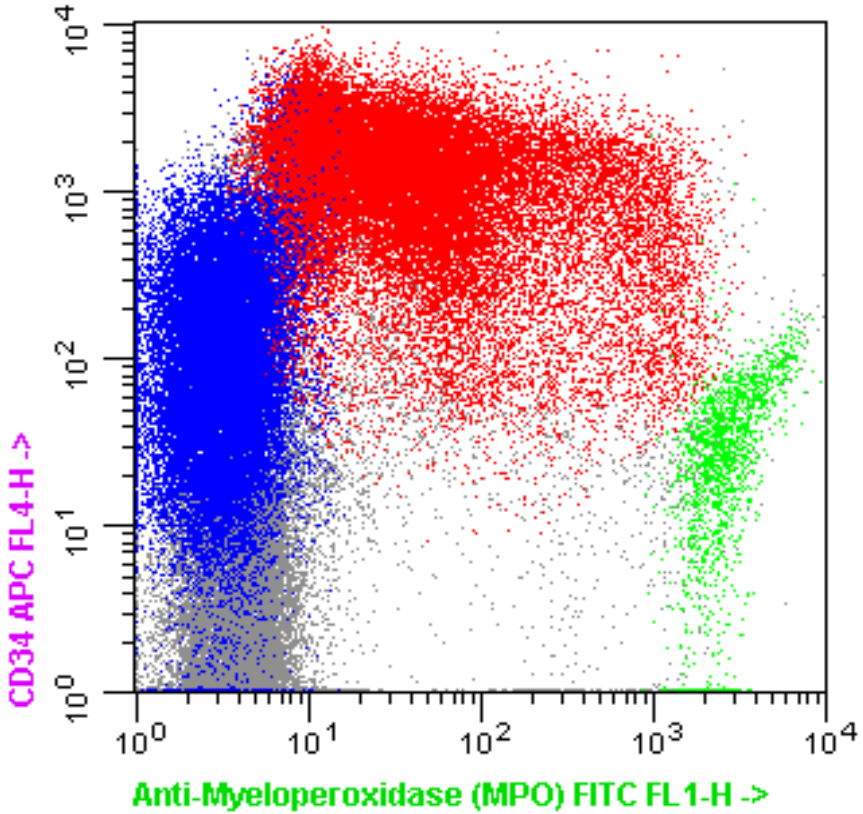
Case 348 Chen

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



Endo A et al. Cancer Sci 2016;107:1745
Jakovljevic G et al. Pediatr Blood Cancer 2010;54:606
Lanocha AA et al. Blood 2017;129: 393

Genetic abnormalities indicating residual disease or prior underlying neoplasm

Case 69 Devins

AML with mutated *NPM1*

68M, circulating blasts, anemia and thrombocytopenia

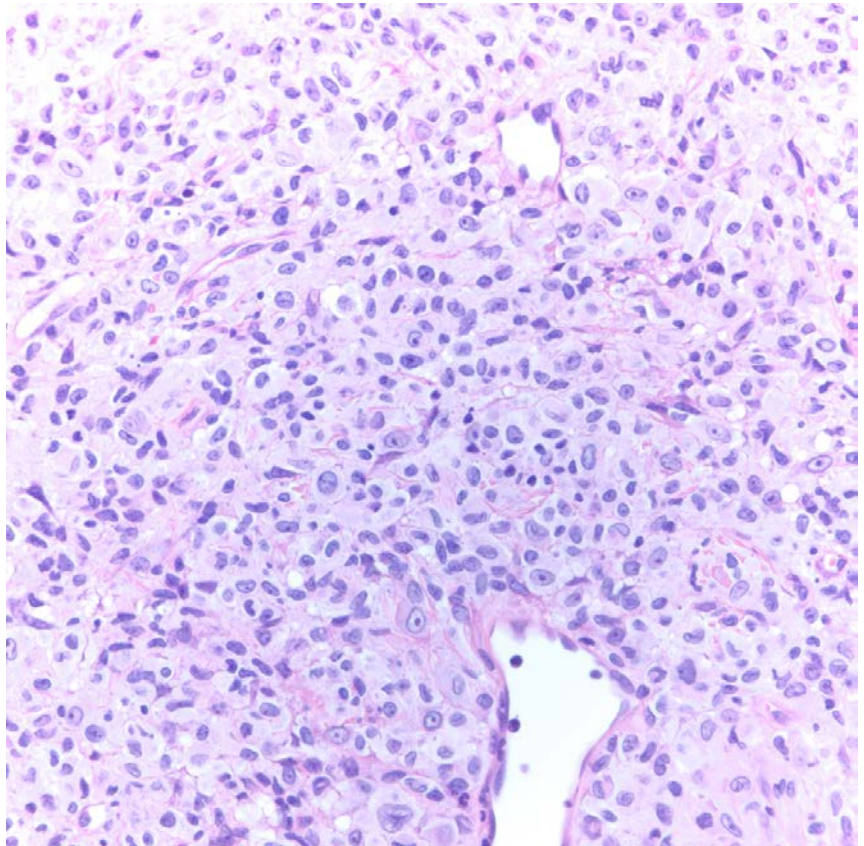
NPM1, KIT, DNMT3A and *TET2* at diagnosis; *DNMT3A* and *TET2* persistent on day 31 (blasts 0%) in unchanged allele frequency; subsequent relapse with the same clone

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

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

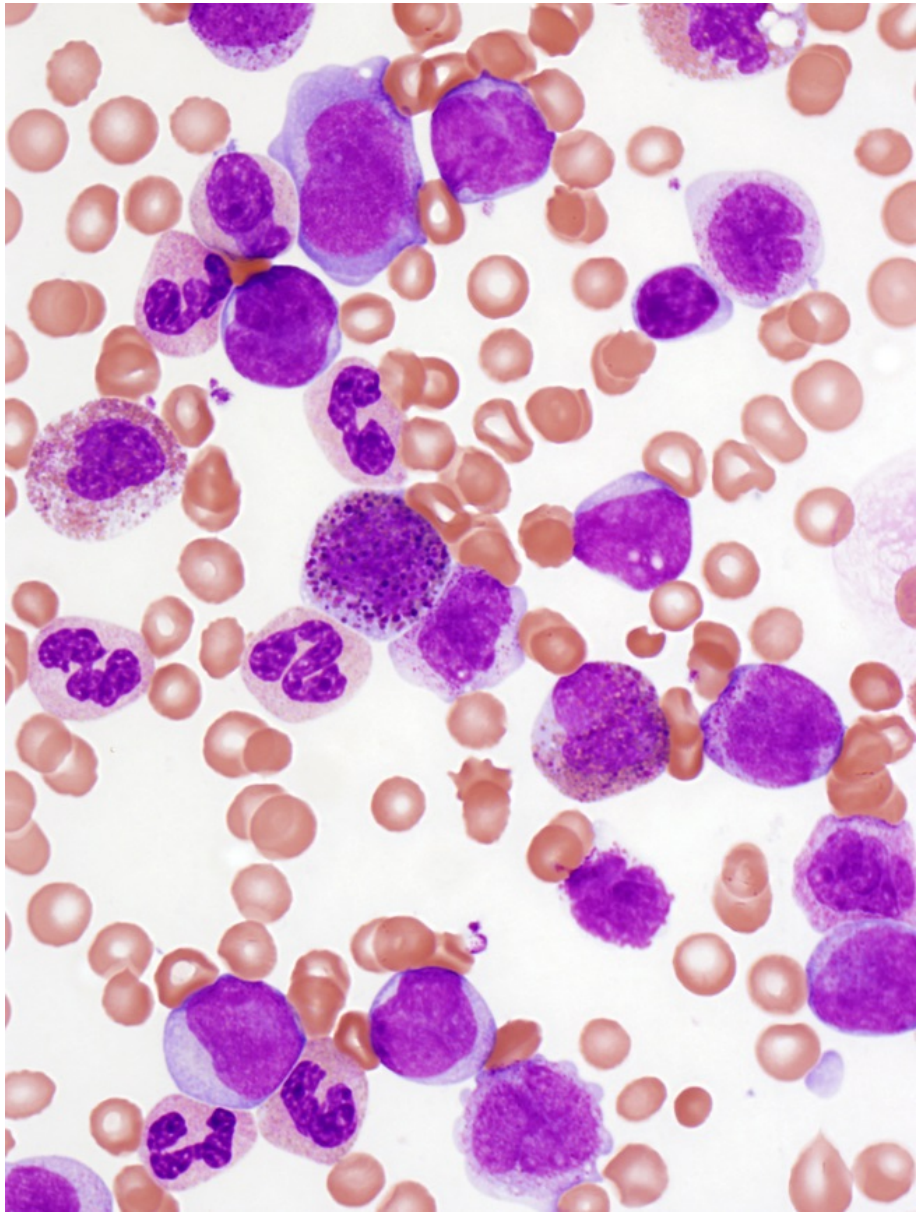
CML, *BCR-ABL1*+, in blast phase [with inv(16)(p13.1q22)]

24F, marked leukocytosis with numerous blasts, eosinophilia, basophilia and anemia

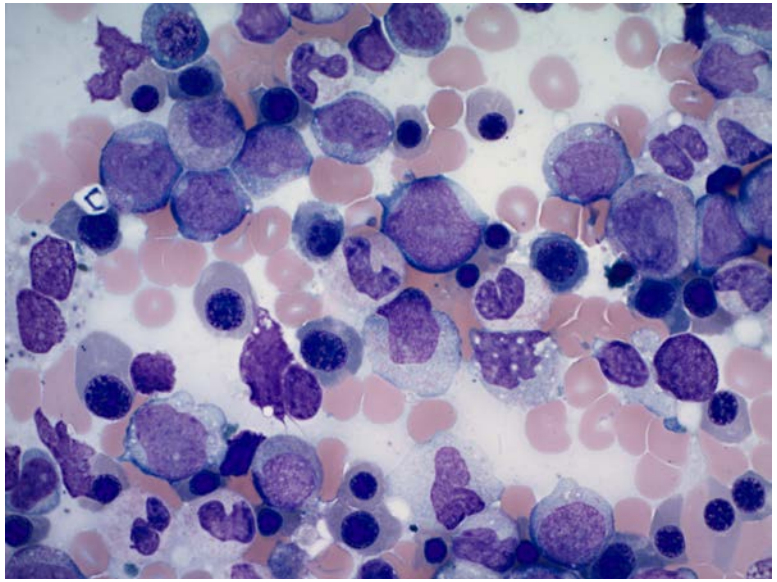
46,XX,t(9;22)(q24;q11.2),inv(16)(p13.1q22)[20]

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

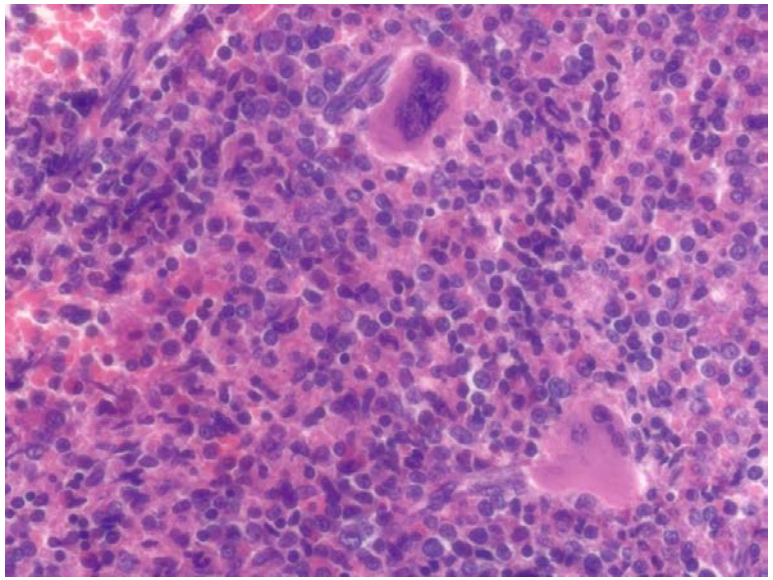


Case 56 Xu

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

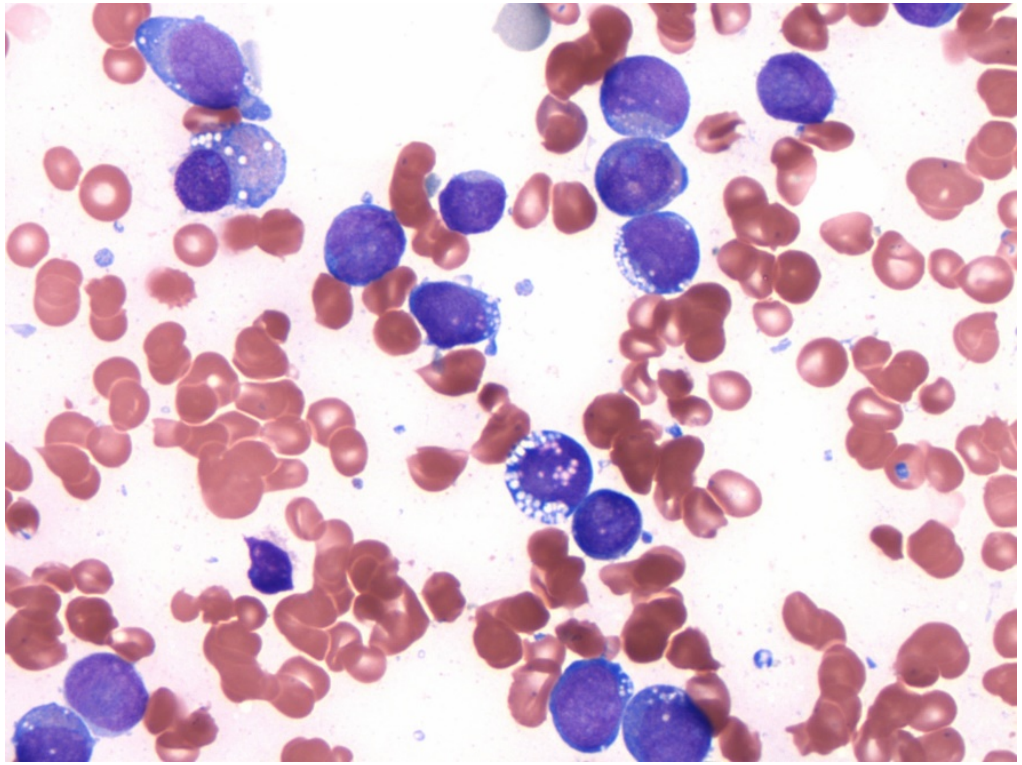
Fluctuating *FLT3, STAG2* and *CSF3R* (VUS) mutations. *CSF3R* variant confirmed to be a germline mutation of donor origin

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



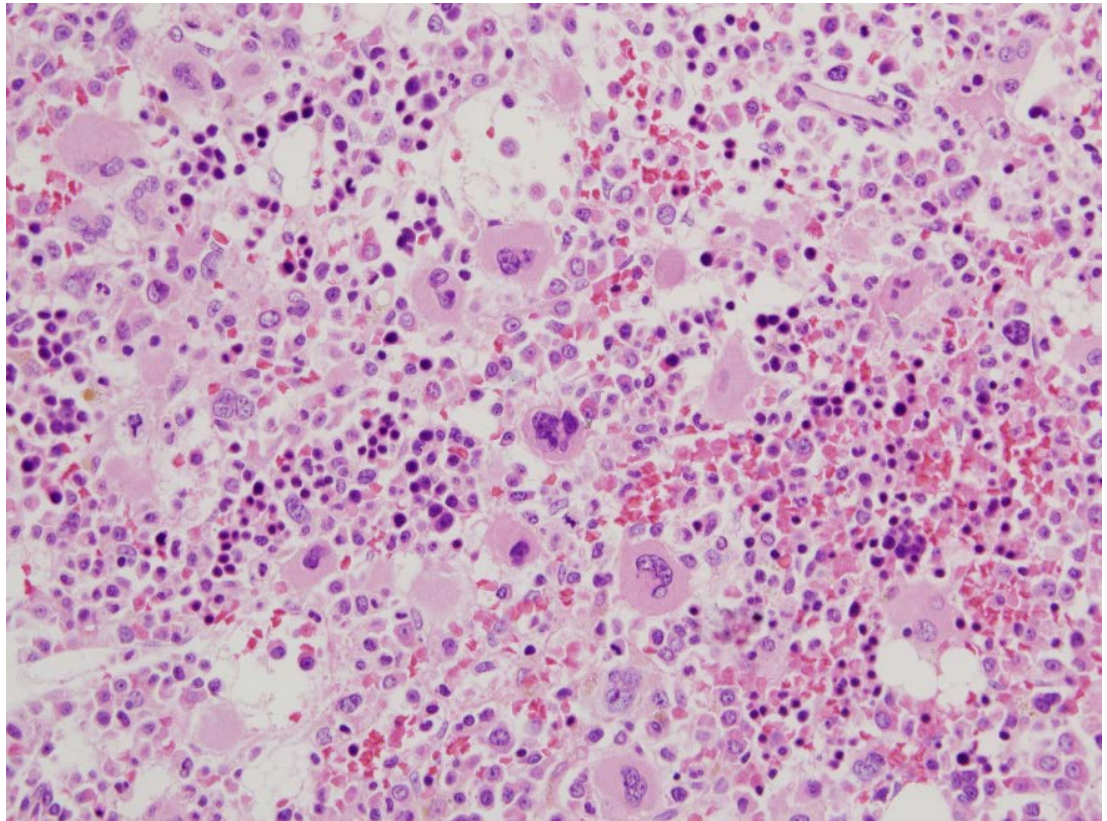
Clonal relationship, clonal evolution and disease heterogeneity

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

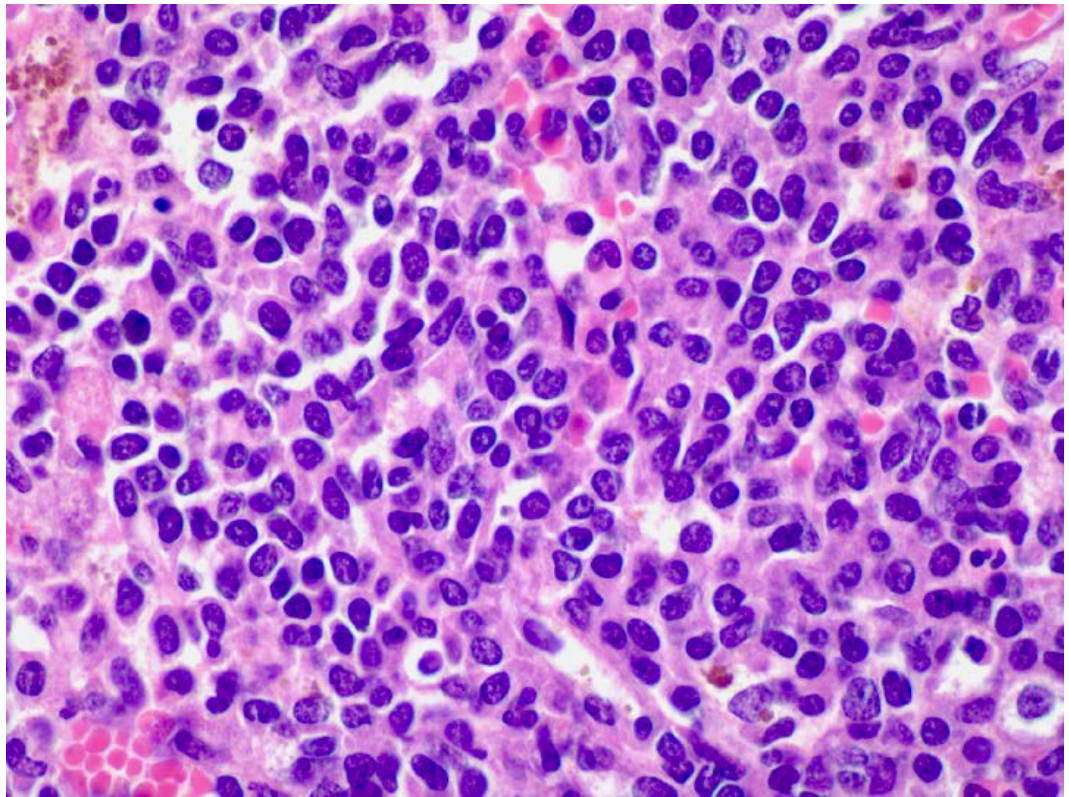
Case 297 Zhang

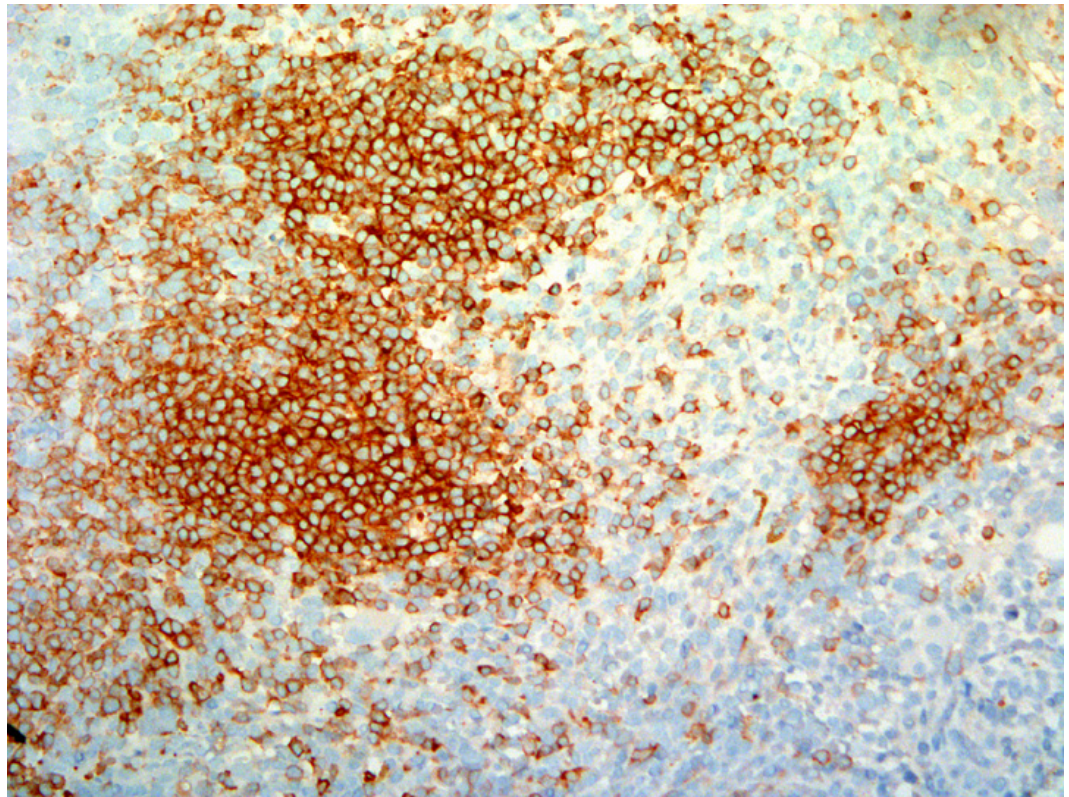
Therapy-related AML and BPDCN

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*

Therapeutic targets

Case 144 Bhattacharyya

Acute myeloid leukemia with mutated *NPM1*

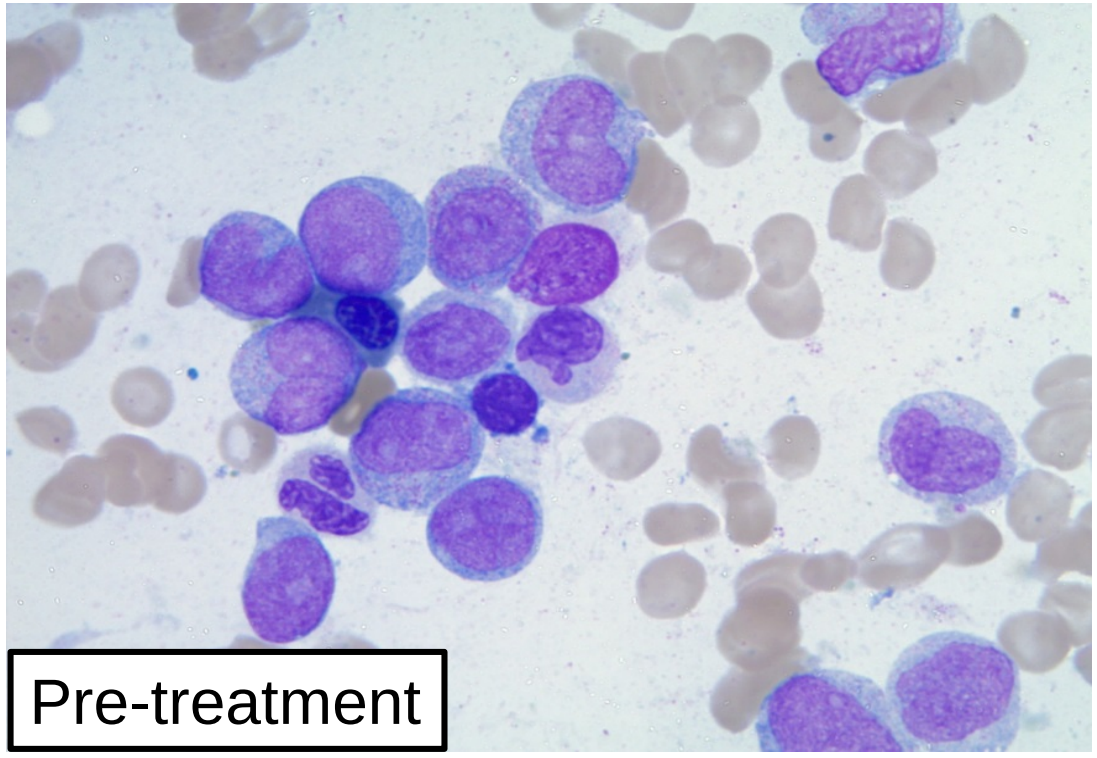
Case 217 Goyal

AML, NOS (AML with maturation) with differentiation

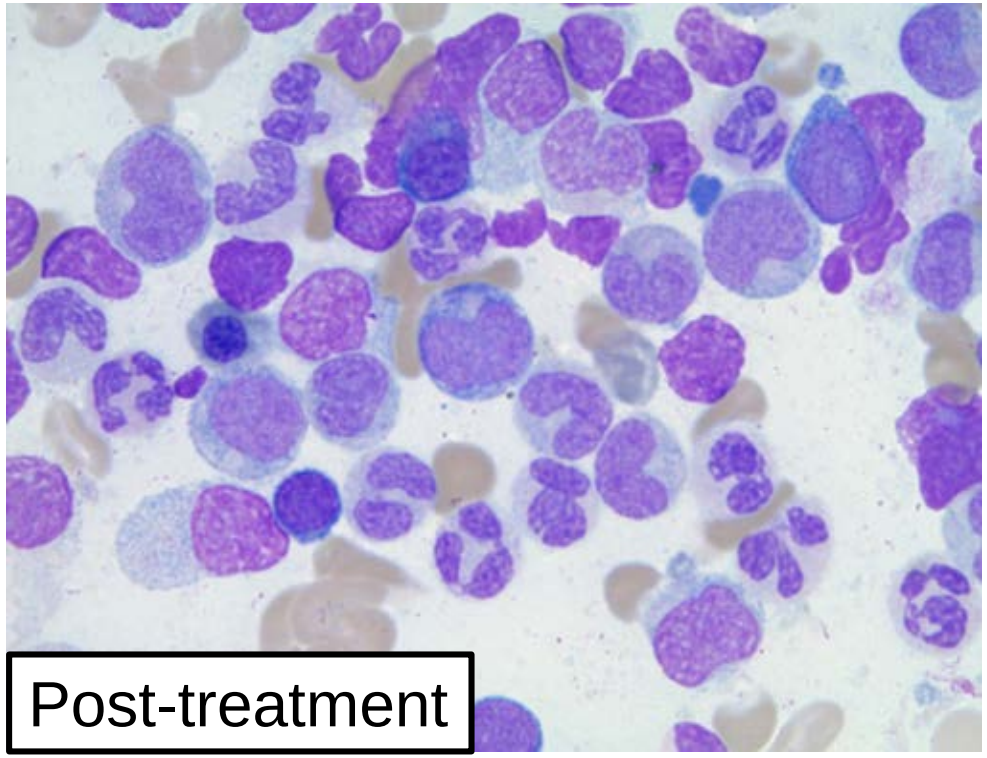
78M, h/o AML, M6 with mutated IDH2

Karyotype pre- and post-treatment: 47,XY,+10[20]

Post-treatment: *IDH2* c.515G>A, VAF 39%, *DNMT3A* c.1227G>A, VAF 41%



Pre-treatment



Post-treatment

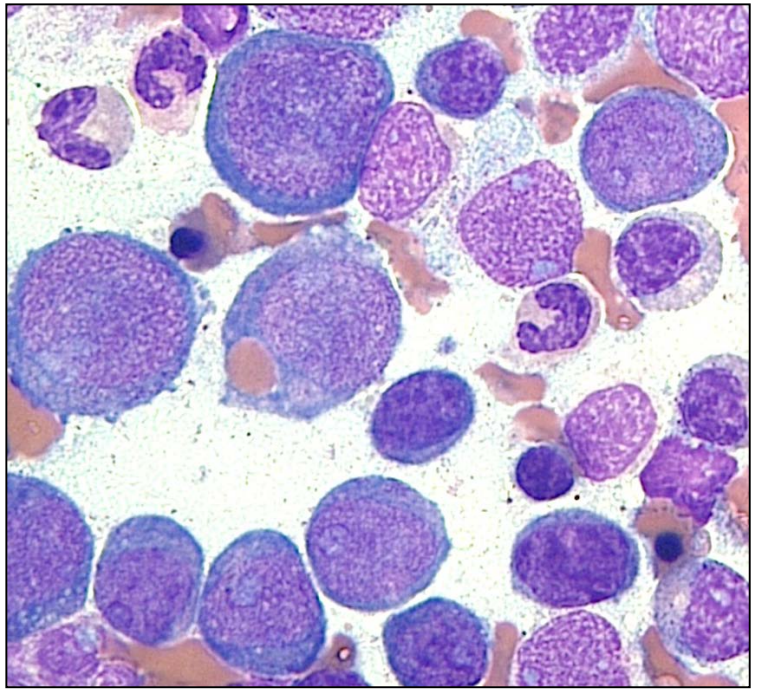
Prognostic implications

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

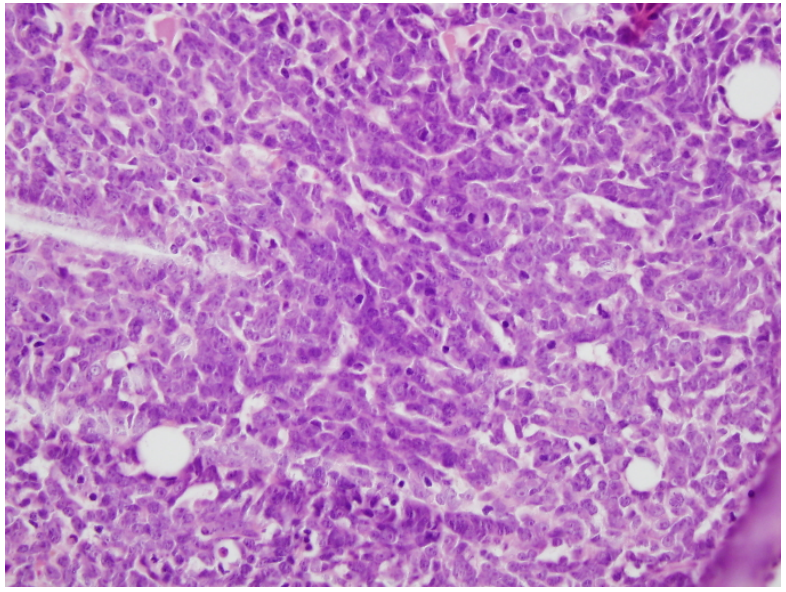
Diagnostic dilemma

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)

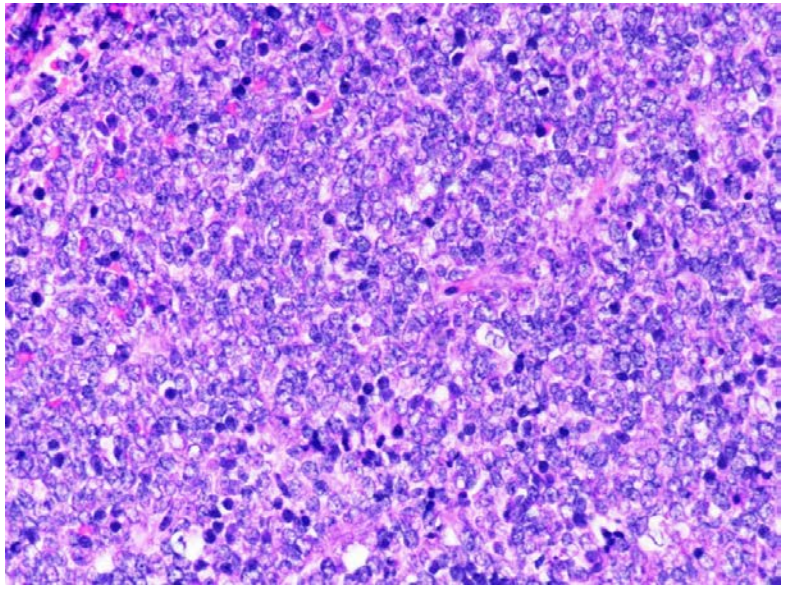


Case 165 Teruya-Feldstein

First biopsy: T-ALL/LBL

Second biopsy: Blastic undifferentiated neoplasm, not definitively classifiable

23M, HIV+, developed new tender lymphadenopathy



Diagnostic dilemma

Case 243 Yuksel

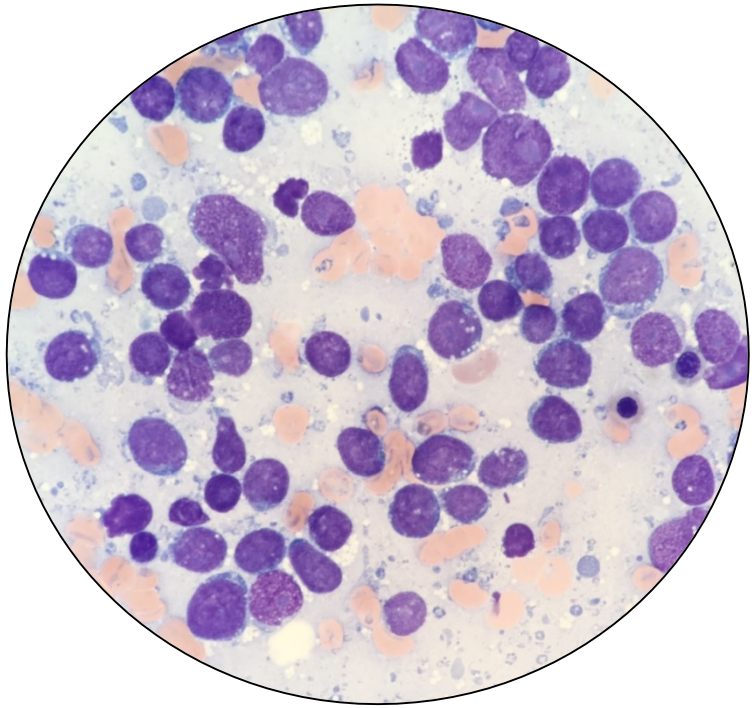
MPAL, B/myeloid, NOS

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?

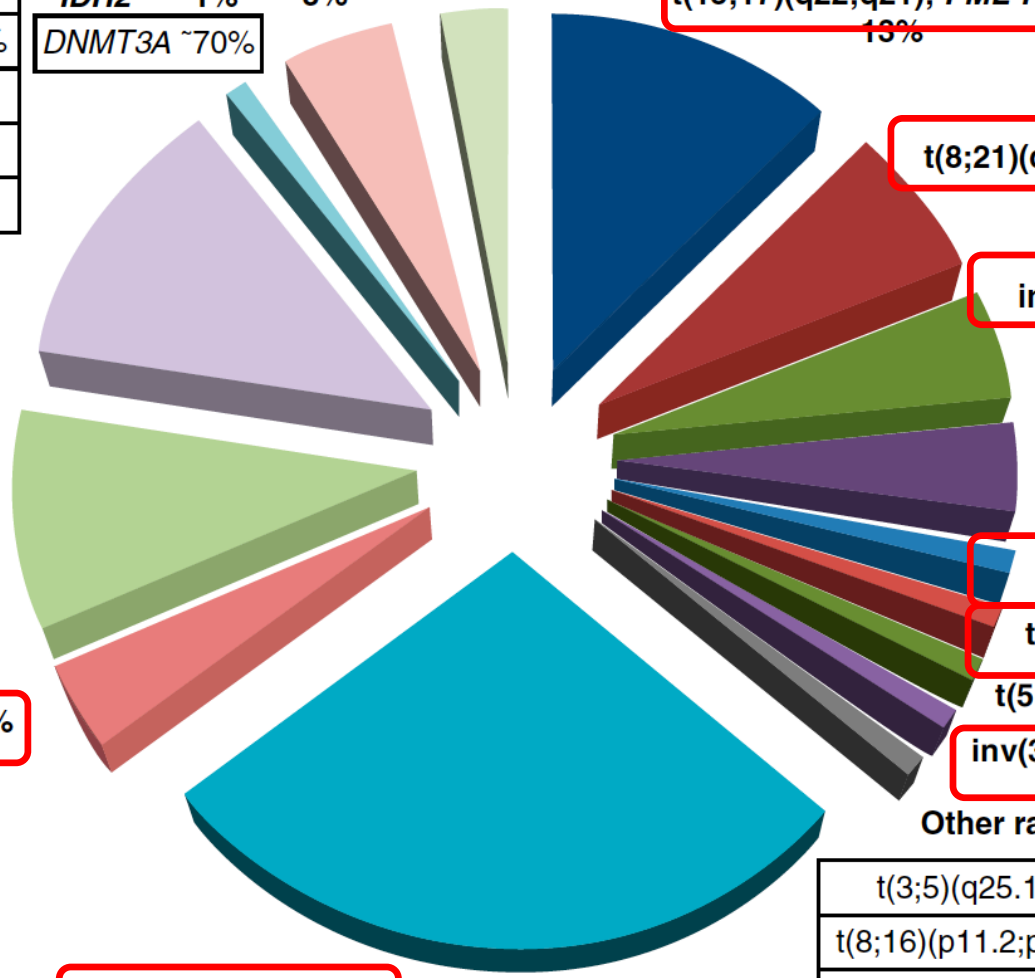
- Patterns of response to targeted therapy:
  Reconciliation of morphologic findings and results of cytogenetic/molecular studies

Dohner et al Blood 2017;129:424

Table 10. Novel therapies in clinical development in AML

Novel therapies in clinical development	
Protein kinase inhibitors	• FLT3 inhibitors (midostaurin, quizartinib, gilteritinib, crenolanib) • KIT inhibitors • PI3K/AKT/mTOR inhibitors • Aurora and polo-like kinase inhibitors, CDK4/6 inhibitors, CHK1, WEE1, and MPS1 inhibitors • SRC and HCK inhibitors
Epigenetic modulators	• New DNA methyltransferase inhibitors (SGI-110) • HDAC inhibitors • IDH1 and IDH2 inhibitors • DOT1L inhibitors • BET-bromodomain inhibitors
Chemotherapeutic agents	• CPX-351 • Vosaroxin • Nucleoside analogs
Mitochondrial inhibitors	• Bcl-2, Bcl-xL, and Mcl-1 inhibitors • Caseinolytic protease inhibitors
Therapies targeting oncogenic proteins	• Fusion transcripts targeting • EVI1 targeting • NPM1 targeting • Hedgehog inhibitors
Antibodies and immunotherapies	• Monoclonal antibodies against CD33, CD44, CD47, CD123, CLEC12A • Immunoconjugates (eg, GO, SGN33A) • BiTEs and DARTs • CAR T cells or genetically engineered TCR T cells • Immune checkpoint inhibitors (PD-1/PD-L1, CTLA-4) • Anti-KIR antibody • Vaccines (eg, WT1)
Therapies targeting AML environment	• CXCR4 and CXCL12 antagonists • Antiangiogenic therapies

Prognosis



RUNX1 ˜40%	*MLL*-PTD ˜25%
ASXL1 ˜20%	*DNMT3A* ˜20%
SRSF2 ˜20%	*STAG2* ˜15%
NRAS ˜15%	*FLT3*-ITD ˜15%
TET2 ˜15%	*BCOR* ˜10%
U2AF1 ˜10%	*PHF6* ˜10%
ZRSR2 ˜5%	*SF3B1* ˜10%
EZH2 ˜5%	

Chromatin-spliceosome 13%

No class 5%

No drivers 3%

IDH2^{R172} 1%
DNMT3A ˜70%

t(15;17)(q22;q21); *PML-RARA* 13%

FLT3-ITD ˜35%
FLT3-TKD ˜15%
WT1 ˜15%

t(8;21)(q22;q22.1); *RUNX1-RUNX1T1* 7%

KIT ˜25%	*NRAS* ˜20%
Cohesin[a] ˜20%	*ASXL2* ˜20%
ZBTB7A ˜20%	*ASXL1* ˜10%
EZH2 ˜5%	*KDM6A* ˜5%
MGA ˜5%	*DHX15* ˜5%

inv(16)(p13.1q22);[b] *CBFB-MYH11* 5%

NRAS ˜40%
KIT ˜35%
FLT3-TKD ˜20%
KRAS ˜15%

t(v;11q23.3); *X-KMT2A* 4%

KRAS ˜20%
NRAS ˜20%

t(9;22)(q34.1;q11.2); *BCR-ABL1* 1%

FLT3-ITD ˜70%
KRAS ˜20%

t(6;9)(p23;q34.1); *DEK-NUP214* 1%

t(5;11)(q35.2;p15.4); *NUP98-NSD1* 1%

FLT3-ITD ˜85%

inv(3)(q21.3q26.2);[c] *GATA2,MECOM* 1%

NRAS ˜30%	*KRAS* ˜15%
PTPN11 ˜20%	*SF3B1* ˜20%
GATA2 ˜15%	*ETV6* ˜15%
PHF6 ˜15%	*RUNX1* ˜10%
BCOR ˜10%	*ASXL1* ˜10%
NF1 ˜10%	

Other rare fusions 1%

t(3;5)(q25.1;q35.1); *NPM1-MLF1*
t(8;16)(p11.2;p13.3); *KAT6A-CREBBP*
t(16;21)(p11.2;q22.2); *FUS-ERG*
t(10;11)(p12.3;q14.2); *PICALM-MLLT10*
t(7;11)(p15.4;p15.2); *NUP98-HOXA9*
t(3;21)(q26.2;q22); *RUNX1-MECOM*

bi*CEBPA* mutant 4%

GATA2 ˜30%
NRAS ˜30%
WT1 ˜20%
CSF3R ˜20%

***NPM1* mutant 30%**

DNMT3A ˜50%	*FLT3*-ITD ˜40%	*Cohesin*[a] ˜20%	*NRAS* ˜20%
IDH1 ˜15%	*IDH2*^{R140} ˜15%	*PTPN11* ˜15%	*TET2* ˜15%

TP53 mutant - chromosomal aneuploidy[d] 10%

FLT3 TP53 AXL1 TET2, WT1, DNMT3A

Dohner et al Blood 2017:129:424 Arber et al Arch Pathol Lab Med 2016