«Adverse Prognosis»
Acute Myeloid Leukemia

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AML

• Update on current definition of prognostic groups

• New biologic understanding (leading possibly to new therapeutic approaches) in some poor prognosis AML

• Summary on Myeloid Sarcoma
AML: Therapy Outcome

Overall prognosis in AML: OS improvement during 40 years

Improvements largely achieved in younger individuals by:

- Same (similar) cytotoxic/DNA damaging (anti-replicative) therapy
- Addition of allogeneic hematopoietic stem cell transplantation (Immunotherapy)
- Improved control of side-effects (early death, infection control)

Burnett et al. JCO 2011
Prognostic factors

Pretreatment factors

• **Patient associated factors** (Comorbidities): Predict *treatment-related early death* under current standard therapy (estimated to account for 1/3)

• **Leukemia associated factors** (leukemia biology, genetics): Predict *resistance* to current standard therapy (estimated to account for at least 2/3)

Posttreatment factors

• Response to therapy (CR, MRD; i.e. mostly leukemia associated factors)
Reduction in TRM

**Leukemia (2014) 28, 289–292**

**Original Article**
Declining rates of treatment-related mortality in patients with newly diagnosed AML given ‘intense’ induction regimens: a report from SWOG and MD Anderson

M Othus¹, H Kantarjian², S Petersdorf¹, F Ravandi¹, J Godwin³, J Cortes³, S Pierce⁴, H Erba⁵, S Faderl⁶, FR Appelbaum⁷, and E Estey⁸

Table 1. Distribution of patient characteristics over time

<table>
<thead>
<tr>
<th></th>
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</tr>
</thead>
<tbody>
<tr>
<td>N</td>
<td>SWOG</td>
<td>392</td>
<td>406</td>
<td>113</td>
<td>498</td>
<td></td>
</tr>
<tr>
<td></td>
<td>MDA</td>
<td>102</td>
<td>638</td>
<td>732</td>
<td>470</td>
<td></td>
</tr>
<tr>
<td>Age</td>
<td>SWOG</td>
<td>64 (17, 89)</td>
<td>65 (19, 85)</td>
<td>58 (20, 83)</td>
<td>49 (19, 60)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>MDA</td>
<td>58 (18, 83)</td>
<td>60 (17, 87)</td>
<td>59 (14, 85)</td>
<td>56 (18, 88)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>WBC (×10⁹)</td>
<td>SWOG</td>
<td>18 (0, 416)</td>
<td>13 (1, 244)</td>
<td>10 (1, 370)</td>
<td>13 (0, 545)</td>
<td>0.16</td>
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<tr>
<td></td>
<td>MDA</td>
<td>14 (0, 262)</td>
<td>8 (0, 394)</td>
<td>7 (0, 433)</td>
<td>5 (0, 204)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Platelets (×10³)</td>
<td>SWOG</td>
<td>56 (3, 537)</td>
<td>53 (2, 1200)</td>
<td>49 (5, 1200)</td>
<td>55 (2, 9300)</td>
<td>0.39</td>
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<tr>
<td></td>
<td>MDA</td>
<td>59 (5, 1355)</td>
<td>46 (3, 2292)</td>
<td>51 (4, 676)</td>
<td>45 (4, 535)</td>
<td>0.02</td>
</tr>
<tr>
<td>Blood blast %</td>
<td>SWOG</td>
<td>32 (0, 99)</td>
<td>30 (0, 99)</td>
<td>22 (0, 97)</td>
<td>31 (0, 99)</td>
<td>0.044</td>
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<tr>
<td></td>
<td>MDA</td>
<td>24 (0, 94)</td>
<td>20 (0, 99)</td>
<td>18 (0, 99)</td>
<td>17 (0, 98)</td>
<td>0.28</td>
</tr>
<tr>
<td>PS 0/1%</td>
<td>SWOG</td>
<td>69</td>
<td>84</td>
<td>81</td>
<td>87</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>MDA</td>
<td>60</td>
<td>65</td>
<td>80</td>
<td>86</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>De novo AML %</td>
<td>SWOG</td>
<td>80</td>
<td>78</td>
<td>85</td>
<td>100</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>MDA</td>
<td>80</td>
<td>78</td>
<td>85</td>
<td>100</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>TRM %</td>
<td>SWOG</td>
<td>18</td>
<td>13</td>
<td>12</td>
<td>3</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>MDA</td>
<td>16</td>
<td>14</td>
<td>9</td>
<td>4</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Abbreviations: AML, acute myeloid leukemia; MDA, MD Anderson; PS, performance status; SWOG, Southwest Oncology Group; TRM, treatment-related mortality; WBC, white blood cell. Median (min, max) or percentage reported. Summary of patient characteristics over four time periods between 1991 and 2009. In both the SWOG and MDA cohorts, median age decreased over time. Correspondingly, the proportion of patients with PS of 0 or 1 increased with 60–70% of patients in 1991–1995 and having PS of 0 or 1 increasing to >85% during 2006–2009. In the SWOG cohort, no patients treated between 2006 and 2009 had secondary AML, such patients being ineligible for the trial open during this period. The MDA cohort included more patients with secondary AML than did the SWOG cohort, reflecting the differences in criteria for antecedent hematological disorder noted above. Of most interest, in both SWOG and MDA TRM rates declined over time (P<0.001), from 18% and 16% during 1991–1995 to 3% and 4% during 2006–2009.

- About **4x reduction in d28 TRM** within 10-15 years
- Likely due to **better supportive care** (and patient selection?)
- same factors likely underlie the decrease in non-relapse mortality following allo-HSCT
Integration of patient prognostic (and some AML) factors in elderly for 8 wk mortality

- age > 80 years; poor performance (2-4 ECOG); elevated creatinine > 1.3 mg/dL. complex karyotypes (> 3 abnormalities)
- Distribution of Patients >65y with none (28%), 1 (40%), 2 (23%), or > 3 factors (9%)
- Estimated 8-week mortality rates of 16% (1), 31% (2), 55% (3), and 71% (3)

One of alternatives: **AML Risk Score** ([http://www.aml-score.org](http://www.aml-score.org)); estimates for

- CR and early death rate in patients ≥60 years and
- probabilities of OS and RFS in patients with AML with normal karyotype.
# AML: Prognostic Factors

## European LeukemiaNet Consensus 2010: Genetics

### Table 4. Standardized reporting for correlation of cytogenetic and molecular genetic data in AML with clinical data

<table>
<thead>
<tr>
<th>Genetic group</th>
<th>Subsets</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Favorable</strong></td>
<td>t(8;21)(q22;q22); RUNX1-RUNX1T1</td>
</tr>
<tr>
<td></td>
<td>inv(16)(p13.1q22) or t(16;16)(p13.1;q22); CBFB-MYH11</td>
</tr>
<tr>
<td></td>
<td>Mutated NPM1 without FLT3-ITD (normal karyotype)</td>
</tr>
<tr>
<td></td>
<td>Mutated CEBPA (normal karyotype)</td>
</tr>
<tr>
<td><strong>Intermediate-I</strong></td>
<td>Mutated NPM1 and FLT3-ITD (normal karyotype)</td>
</tr>
<tr>
<td></td>
<td>Wild-type NPM1 and FLT3-ITD (normal karyotype)</td>
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<td></td>
<td>Wild-type NPM1 without FLT3-ITD (normal karyotype)</td>
</tr>
<tr>
<td><strong>Intermediate-II</strong></td>
<td>t(9;11)(q22;q23); MLL3-MLL</td>
</tr>
<tr>
<td></td>
<td>Cytogenetic abnormalities not classified as favorable or adverse‡</td>
</tr>
<tr>
<td><strong>Adverse</strong></td>
<td>inv(3)(q21q26.2) or t(3;3)(q21q26.2); RPN1-EVI1</td>
</tr>
<tr>
<td></td>
<td>t(6;9)(p23;q34); DEK-NUP214</td>
</tr>
<tr>
<td></td>
<td>t(v;11)(v;q23); MLL rearranged</td>
</tr>
<tr>
<td></td>
<td>–5 or del(5q); –7; abnl(17p); complex karyotype‡</td>
</tr>
</tbody>
</table>

*Frequencies, response rates, and outcome measures should be reported by genetic group, and, if sufficient numbers are available, by specific subsets indicated; excluding cases of acute promyelocytic leukemia.

*Includes all AMLs with normal karyotype except for those included in the favorable subgroup; most of these cases are associated with poor prognosis, but they should be reported separately because of the potential different response to treatment.

‡For most abnormalities, adequate numbers have not been studied to draw firm conclusions regarding their prognostic significance.

§Three or more chromosome abnormalities in the absence of one of the WHO designated recurring translocations or inversions; that is, t(15;17), t(8;21), inv(16) or t(16;16), t(9;11), t(11;v)(v;q23), t(6;9), inv(3) or t(3;3); indicate how many complex karyotype cases have involvement of chromosome arms 5q, 7q, and 17p.
European LeukemiaNet Consensus 2010: Genetics

- Distribution of Risk Groups

N=818 (younger than 60 years)  N = 732 (60 years or older)

→ In older Patients more adverse/intermediate II, less favorable risk distribution

Mrozek et al.; JCO 2012
AML: Prognostic Factors

European LeukemiaNet Consensus 2010: Genetics

• Outcome according to Risk Groups and age

Fig 1. Outcome of patients with primary acute myeloid leukemia classified into the four European LeukemiaNet genetic groups according to the European LeukemiaNet recommendations. (A) Disease-free survival and (B) overall survival of patients younger than age 60 years; (C) disease-free survival and (D) overall survival of patients age 60 years or older.

⇒ In older Patients (clinical majority) no difference between IR I and II
AML: Prognostic Factors – Additional Genetic „Drivers“

Integrating Mutational Profiling

**Prognostic Relevance of Integrated Genetic Profiling in Acute Myeloid Leukemia**

Jay P. Patel, Mithat Gönen, Ph.D., Maria E. Figueroa, M.D., Hugo Fernandez, M.D., Zhuxin Sun, Ph.D., Janis Racevskis, Ph.D., Pieter Van Vlierberghe, Ph.D., Igor Dolgalev, B.S., Sabrena Thomas, B.S., Olga Aminova, B.S., Kety Huberman, B.S., Janice Cheng, B.S., Agnes Viale, Ph.D., Nicholas D. Soci, Ph.D., Adriana Hegay, Ph.D., Athena Cherry, Ph.D., Cail Vance, M.D., Rodney R. Higgins, Ph.D., Rhett P. Ketterling, M.D., Robert E. Gallagher, M.D., Mark Lizotte, M.D., Marcel R. van den Brink, M.D., Ph.D., Hilary M. Lazarus, M.D., Jacob M. Rose, M.D., Selina Luget, M.D., Adolfo Fernando, M.D., Ph.D., Elisabeth Pajetta, Ph.D., Martin S. Tallman, M.D., Ari Melnick, M.D., Omar Abdel-Wahab, M.D., and Ross L. Levine, M.D.

**Genomic Classification and Prognosis in Acute Myeloid Leukemia**

Elli Papaemmanuil, Ph.D., Moritz Gerstung, Ph.D., Lars Bullinger, M.D., Verena I. Gaidzik, M.D.,

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- Analysis of **18 genes** in **398 patients** younger than 60 years of age
- Analysis of **111 genes** in **1540 patients** in three prospective trials of intensive therapy
Adapted European LeukemiaNet Consensus 2017:
- 3 groups: adaptation of genetics (refined) and to relevant age groups

### Table 5. 2017 ELN risk stratification by genetics

<table>
<thead>
<tr>
<th>Risk category*</th>
<th>Genetic abnormality</th>
</tr>
</thead>
<tbody>
<tr>
<td>Favorable</td>
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<tr>
<td></td>
<td>Biallelic mutated CEBPA</td>
</tr>
<tr>
<td>Intermediate</td>
<td>Mutated NPM1 and FLT3-ITD&lt;sub&gt;high&lt;/sub&gt;‡</td>
</tr>
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<td></td>
<td>Wild-type NPM1 without FLT3-ITD or with FLT3-ITD&lt;sub&gt;low&lt;/sub&gt;† (without adverse-risk genetic lesions)</td>
</tr>
<tr>
<td></td>
<td>t(9;11)(p21.3;q23.3); MLLT3-KMT2A‡</td>
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<td>Complex karyotype,§ monosomal karyotypetl</td>
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<td>Wild-type NPM1 and FLT3-ITD&lt;sub&gt;high&lt;/sub&gt;‡</td>
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<td></td>
<td>Mutated RUNXI§</td>
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<tr>
<td></td>
<td>Mutated ASXL1¶</td>
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<tr>
<td></td>
<td>Mutated TP53#</td>
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### 2010 ELN risk groups

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→ 2017 Risk stratification meaningful for majority of AML patients
AML: Prognostic Factors

Beyond ELN: Integrating Combinations of Cytogenetics and Mutational Profiling

A Revised Risk Stratification

<table>
<thead>
<tr>
<th>Cytogenetic Classification</th>
<th>Mutations</th>
<th>Overall Risk Profile</th>
</tr>
</thead>
<tbody>
<tr>
<td>Favorable</td>
<td>Any</td>
<td>Favorable</td>
</tr>
<tr>
<td>FLT3-ITD-negative</td>
<td>Mutant NPM1 and IDH1 or IDH2</td>
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<tr>
<td>FLT3-ITD-negative</td>
<td>Wild-type ASXL1, MLL-PTD, PHF6, and TET2</td>
<td>Intermediate</td>
</tr>
<tr>
<td>FLT3-ITD-negative or positive</td>
<td>Mutant CEBPA</td>
<td></td>
</tr>
<tr>
<td>FLT3-ITD-positive</td>
<td>Wild-type MLL-PTD, TET2, and DNMT3A and trisomy 8-negative</td>
<td></td>
</tr>
<tr>
<td>FLT3-ITD-negative</td>
<td>Mutant TET2, MLL-PTD, ASXL1, or PHF6</td>
<td></td>
</tr>
<tr>
<td>FLT3-ITD-positive</td>
<td>Mutant TET2, MLL-PTD, DNMT3A, or trisomy 8, without mutant CEBPA</td>
<td>Unfavorable</td>
</tr>
<tr>
<td>Unfavorable</td>
<td>Any</td>
<td></td>
</tr>
</tbody>
</table>

A Effect of Mutational Profiling

<table>
<thead>
<tr>
<th>Cytogenetic Classification</th>
<th>Mutational Analysis</th>
<th>Integrated Classification</th>
</tr>
</thead>
<tbody>
<tr>
<td>Favorable</td>
<td>3-yr OS: 85%</td>
<td>Favorable: 26% of cohort (3-yr OS: 64%)</td>
</tr>
<tr>
<td>Intermediate: 63% of cohort (3-yr OS: 36%)</td>
<td>3-yr OS: 42%</td>
<td>Intermediate: 35% of cohort (3-yr OS: 42%)</td>
</tr>
<tr>
<td>Unfavorable: 18% of cohort (3-yr OS: 11%)</td>
<td>3-yr OS: 13%</td>
<td>Unfavorable: 39% of cohort (3-yr OS: 12%)</td>
</tr>
</tbody>
</table>

→ Integrated Classification: Revision of Risk Groups

Analysis of 18 genes in 398 patients younger than 60 years of age

N ENGL J MED 366;12 NEJM.ORG MARCH 22, 2012
AML: Prognostic Factors – Additional Genetic „Drivers“

Beyond ELN: Integrating Combinations of Cytogenetics and Mutational Profiling

- TF Fusions
- NPM1
- Tumor Suppressors
- DNA Methylation
- Activated Signaling
- Myeloid TFs
- Chromatin Modifiers
- Cohesin
- Spliceosome
- Cytogenetic Risk

TF Fusions

NPM1

Tumor Suppressors

DNA Methylation

Activated Signaling

Myeloid TFs

Chromatin Modifiers

Cohesin

Spliceosome

Cytogenetic Risk

Beyond ELN: Integrating Combinations of Cytogenetics and Mutational Profiling

- Exclusive across categories
- Co-occurring across categories
- Co-occurring within category
- 2-Hit mutation

Cytogenetic Risk

- Unfavorable
- Intermediate
- Favorable
- Unknown

Dendrix ++ Group

- A
- B
- C

200 AML Samples

N ENGL J MED 368;22 NEJM.ORG MAY 30, 2013
Beyond ELN: Risk Classification Using Knowledge Bank Approach

Multistage outcome predictions for 1,024 patients.

• Personally tailored management decisions could reduce the number of hematopoietic cell transplants in patients with AML by 20–25% while maintaining overall survival rates.
European LeukemiaNet Consensus 2017
• Additional info on combinations (w/o integration in prognostic model)

AML: Prognostic Factors
AML: Prognostic Factors

Post-treatment Prognostic Factors: minimal residual (measurable) disease

Ding et al, Nature 2012
Post-treatment Prognostic Factors: minimal residual (measurable) disease

- MRD positivity is likely a bad thing
- MRD negativity might be a good thing (cure) or a sign of insufficiently sensitive measurement
- Time to achievement of MRD negativity from start of therapy could be predictive of tumor mass development below detection level under continued treatment

*Ding et al, Nature 2012*
AML: Prognostic Factors

*NPM1*mut-based MRD monitoring during and after therapy

Monitoring of Minimal Residual Disease in *NPM1*-Mutated Acute Myeloid Leukemia: A Study From the German-Austrian Acute Myeloid Leukemia Study Group


(A) Cumulative incidence of relapse (CIR) and
(B) OS for patients in CR according to MRD status after induction therapy in bone marrow

→ *NPM1*mut-based MRD-monitoring during and after therapy allows for the discrimination of patients at high and low risk of relapse

*J Clin Oncol* 29:2709-2716. © 2011 by American Society of Clinical Oncology
AML: Prognostic Factors

Post-treatment Prognostic Factors: minimal residual (measurable) disease

Decision Point MRD – to be clarified

- Will MRD positive patients profit from more therapy and/or different therapies?
- Should MRD negative patients receive less treatment?

disease
clinical detection
facilitated detection
sensitive facilitated detection
most sensitive facilitated detection
CURE?
### AML: Prognostic Factors

HOVON/SAKK 132 Risk Classification and therapy adaptation in small group: 

a) at Diagnosis; b) upon Cycle II based on MRD

<table>
<thead>
<tr>
<th>Risk Group</th>
<th>Criteria at diagnosis and early/late CR</th>
<th>Plus criteria after cycle II based on MRD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Good (autoHSCT consolidation*)</td>
<td>t(8;21) or AML1-ETO, WBC≤20 inv<a href="16;16">16</a> or CBFB-MYH11 CEBPA-biallelic mutant+ FLT3ITD/-NPM1+,</td>
<td>irrespective of MRD- or MRD+ irrespective of MRD- or MRD+ irrespective of MRD- or MRD+ irrespective of MRD- or MRD+</td>
</tr>
<tr>
<td>Intermediate (autoHSCT consolidation&amp;**)</td>
<td>CN − X − Y, WBC≤100, Cre t(8;21) or AML1-ETO, plus WBC&gt;20 or mutant KIT</td>
<td>Intermediate risk features at baseline (see category in left section) and also MRD:- also MRD- also MRD- (if no MRD information available; see legend below)</td>
</tr>
<tr>
<td>Poor (alloHSCT consolidation***)</td>
<td>CN − X − Y, WBC≤100, Cre t(8;21) or AML1-ETO, WBC&gt;20 and/or mutant KIT CN − X − Y, WBC≤100, not Cre CN − X − Y, WBC&gt;100, CA, but non-CBF, MK-, no abn3q28</td>
<td>Intermediate risk features as above but MRD pos: but MRD+ but MRD+ irrespective of MRD- or MRD+ but MRD- but MRD-</td>
</tr>
<tr>
<td>Very Poor (alloHSCT consolidation***)</td>
<td>CN − X − Y, WBC&gt;100 CA, but non CBF, MK-, no abn3q28, EV11-neg</td>
<td>Poor risk features as above but MRD pos: but MRD+ but MRD+ Very poor risk features at diagnosis, independent of MRD assessment: irrespective of MRD- or MRD+ irrespective of MRD- or MRD+ irrespective of MRD- or MRD+ irrespective of MRD- or MRD++ irrespective of MRD- or MRD+</td>
</tr>
</tbody>
</table>

Flow-based MRD (LAP) and mol. based (spec. Mol marker) MRD: coverage of about 90% of AML
AML

• Update on current definition of prognostic groups
• New biologic understanding (leading possibly to new therapeutic approaches) in some poor prognosis AML
• Summary on Myeloid Sarcoma
Adverse Risk Group

- $t(6;9)(p23;q34.1)$; DEK-NUP214 (frequently with FLT3-ITD (70%) or NRAS (20%))
- $t(v;11q23.3)$; **KMT2A (MLL) rearranged**
- $t(9;22)(q34.1;q11.2)$; BCR-ABL1
- $\text{inv}(3) (q21.3q26.2)$ or $t(3;3)(q21.3;q26.2)$; **GATA2, MECOM (EVI1)**
- -5 or del(5q); -7; -17/abn(17p)
- Complex karyotype, monosom al karyotype
- Wild-type NPM1 and **FLT3-ITD high**
- Mutated RUNX1
- Mutated ASXL1
- **Mutated TP53 (10%)**
Mutated Flt3

- Activating Flt3 mutations (FLT3-ITD and TKD mutations) are present in about 30% of AML (predominantly in cytogenetically normal AML), thus being one of the most frequently affected genes in AML.

- ITD mutations result in amino acid sequence changes with intact coding frames. Leads to constitutive activation of the receptor tyrosine kinase.

- Results in constitutive activation of downstream signaling through JAK-STAT/RAS/RAF/MEK/mTOR growth and proliferation pathways and PI3K/AKT prosurvival pathways.

- Associated with highly proliferative AML.
**Mutated Flt3: Relevance of Allelic Ratio (AR, Flt3mut\(^{\text{high}}\) vs \(\text{low}\))**

- Mutant to wild-type (WT) allelic ratio (AR) and the size of the ITD consistently show association between high allelic burden with unfavorable outcome.

- High allelic ratio associated with at least in part loss of heterozygosity (LOH), and presumably provides a survival advantage for the leukemic blast cells.

- Loss of WT allele in mouse models → more aggressive AML.

- Mutational burden (VAF) is of relevance (bigger or smaller than 50%).

- <50% (and NPM1-mutation) better outcome.

- Major benefit of alloHSCT in first CR in patients with an AR of >0.51 with respect to RFS and OS.

Richard F. Schlenk et al., Blood 2014
Mutated Flt3 represents «target» for therapy

Quizartinib
AC220

Lestaurnib
CEP-701

Tandutinib
MLN-518

Crenolanib

Midostaurin
PKC-412

Midostaurin
Metabolite
CGP52421

Sorafenib

Sunitinib
t(v;11q23.3); KMT2A (MLL) rearranged

Histone-lysine N-methyltransferase 2A also known as acute lymphoblastic leukemia 1 (ALL-1), myeloid/lymphoid or mixed-lineage leukemia 1 (MLL1), is encoded by the KMT2A gene

- histone-modifying enzyme, positive global regulator of gene transcription / epigen. transcript memory

- MLL translocations are diverse and include the loss of the C-terminal H3K4 methyltransferase domain of MLL, followed by silencing of tumor suppressors

- Fusion partners of MLL interact with several proteins, including the H3K79 methyltransferase DOT1L

- Oncogenic interactions possibly therapeutic targets, and small molecule inhibitors targeting interactions with, e.g. DOT1L, are in development

inv(3)(q21.3q26.2) or t(3;3)(q21.3;q26.2); GATA2,MECOM(EVI1)

• EVI1 is a proto-oncogenic transcription factor involved in the regulation of hematopoietic stem cells
• Overexpression has been induced in retrovial SCID gene therapy trials (preferred integration site) and led to AML and MDS
• EVI1 deregulation is often accompanied by a nearby inversion inv(3) or translocation t(3;3) of largely non-genic sequence, but mechanistic links between these rearrangements and EVI1 expression changes have remained poorly understood
• Recently demonstrated: **GATA2 enhancer translocation** resulting in the simultaneous upregulation of a proto-oncogene and downregulation of a tumor suppressor gene in a spatio-temporal specific manner
Mutated TP53 (TP53 inactivation/dysfunction/loss)

- TP53 is the «guardian of the genome»: Most potent tumor suppressor, regulation of senescence, apoptosis, metabolism, DNA repair
- Impaired function or loss of function in about 8% of de novo AML and 15-20% of therapy related AML or AML with MDS-related changes
- Associated with complex genetic changes
- Poor outcome, chemoresistance, irrespective of age (and cancers)
Mutated TP53 (TP53 inactivation/dysfunction/loss)

Dysfunctional diversity of p53 proteins in adult acute myeloid leukemia: projections on diagnostic workup and therapy

Miron Prokocimer,1, Alina Molchadsky,2 and Varda Rotter2

1Sackler Faculty of Medicine, Tel-Aviv University, Tel-Aviv, Israel; and 2Department of Molecular Cell Biology, The Weizmann Institute of Science, Rehovot, Israel

• Non-mutational p53 abnormalities and gain of function might bear a greater AML-related significance than is currently appreciated

WT p54 inactivation mechanisms in AML

Mutant p54 gain of function in AML
Mutated TP53 (TP53 inactivation/dysfunction): Therapeutic Target?

- Encouraging results of clinical trials with MDM2 inhibitors (addresses dysfunctional wt p53 protein)
- Encouraging results with hypomethylating decitabine (which decreases mutant p53 levels)

→ p53-based therapies might improve AML outcome
TP53: Therapeutic Target dependent on status?

**Indicators of p53 alterations**
- cld age, cancer history, prior chemo/irradiation, chemoresistance, cancer family history, unfavorable cytogenetics, chromosomal translocations

**p53 genotyping**
- Cytogenetics/FISH, SNP-As, NGS, Taq-Based methods

**p53 phenotyping**
- Protein levels: p53, p21, Puma.
- Function: cell cycle arrest, apoptosis

**Functional vs. Dysfunctional**
- wtp53
  - No further p53 evaluation needed
  - Add therapy for wtp53 stabilization & activation [MDM2 inhibitors]
  - Search for specific therapeutically relevant wtp53 inactivating mechanisms
  - MDM2/4 up-regulation
  - p53 deacetylation
  - p53 cytoplasmic sequestration
  - ARF down-regulation

- mutp53
  - LOF / GOF
  - Add therapy for mutp53 reactivation [e.g. APR-246]
  - Search for specific therapeutically relevant mutp53 GOF properties
  - mutp53 binding to p73
  - mutp53 binding to HSP90
  - mutp53-induced MDR1 up-regulation
  - mutp53-induced NF-kB activation
  - APR-246 & RETRA
  - APR-246 & HSP90 inhibitors
  - APR-246 & MDR reversal agents
  - APR-246 & NF-kB inhibitors

*Source: Blood, 10 August 2017 - Volume 130, Number 6*
Conclusions Adverse Risk Leukemia

- Risk stratification represents a result of post-hoc analysis of currently available pretreatment parameters and current standard therapy outcome.

- Expectations are that molecular precision analysis will be extended via «big data» integration and subsequent prediction.

- MRD-based (response) strategies will play an increasingly important role.

- Efficient selective targeting will (hopefully) lead to subsequent «shifts» in risk populations.
AML

• Update on current definition of prognostic groups

• New biologic understanding (leading possibly to new therapeutic approaches) in some poor prognosis AML

• Summary on Myeloid Sarcoma
Add On: Myeloid Sarcoma Summarized

• MS in 2-8% of patients with AML, either as a single or as a multifocal tumor (most likely under-diagnosed)

• MS can predate AML (ca. 25%), occurs concomitantly with AML (ca. 25%), or occurs after AML diagnosis (ca. 50%)

• Frequent sites: skin, lymph nodes, GI, bone, soft tissue, testes

• MS displays mostly myelomonocytic or pure monoblastic morphology. AML blasts might display unique set of chemokine receptors including CCR5, CXCR4, CXCR7 and CX3CR1 compared with BM and blood blasts

• Standard AML workup is required

• Systemic AML therapies are recommended, even in isolated MS

• Surgery not recommended (only for local symptom relief), Radiation therapy not recommended

• **Outcome as in non-MS AML;** occasional reports that isolated MS show better prognosis

Therapeutic Advances in Hematology, 2011  Personal communication HOVON/SAKK  Clinical Lymphoma, Myeloma & Leukemia, Vol. 17, No. 5, 263-7 Crown Copyright © 2017
Thank You for Your Attention
Potential Functional (Driver) Targets in (poor risk) AML

At least 8 functional mutational categories in AML (Cancer Genome Atlas Research Network): Gain of function – natural target

- Activated Signaling
- TF Fusions
- Tumor Suppressors
- DNA Methylation
- Chromatin Modifiers
- Spliceosome Complex

Figure 1: Eight Functional Categories of Genes That Are Commonly Mutated in Acute Myeloid Leukemia.
Adapted European LeukemiaNet Consensus 2017:
- 3 groups: adaptation of genetics (refined) and to relevant age groups

Table 5. 2017 ELN risk stratification by genetics

<table>
<thead>
<tr>
<th>Risk category*</th>
<th>Genetic abnormality</th>
</tr>
</thead>
<tbody>
<tr>
<td>Favorable</td>
<td>t(8;21)(q22;q22.1); RUNX1-RUNX1T1</td>
</tr>
<tr>
<td></td>
<td>inv(16)(p13.1q22) or t(16;16)(p13.1;q22); CBFB-MYH11</td>
</tr>
<tr>
<td></td>
<td>Mutated NPM1 without FLT3-ITD or with FLT3-ITD&lt;sub&gt;low&lt;/sub&gt;†</td>
</tr>
<tr>
<td></td>
<td>Biallelic mutated CEBPA</td>
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<tr>
<td>Intermediate</td>
<td>Mutated NPM1 and FLT3-ITD&lt;sub&gt;high&lt;/sub&gt;†</td>
</tr>
<tr>
<td></td>
<td>Wild-type NPM1 without FLT3-ITD or with FLT3-ITD&lt;sub&gt;low&lt;/sub&gt;† (without adverse-risk genetic lesions)</td>
</tr>
<tr>
<td></td>
<td>t(9;11)(p21.3;q23.3); MLLT3-KMT2A‡</td>
</tr>
<tr>
<td></td>
<td>Cytogenetic abnormalities not classified as favorable or adverse</td>
</tr>
<tr>
<td>Adverse</td>
<td>t(6;9)(p23;q34.1); DEK-NUP214</td>
</tr>
<tr>
<td></td>
<td>t(v;11q23.3); KMT2A rearranged</td>
</tr>
<tr>
<td></td>
<td>t(9;22)(q34.1;q11.2); BCR-ABL1</td>
</tr>
<tr>
<td></td>
<td>inv(3)(q21.3q26.2) or t(3;3)(q21.3; q26.2); GATA2,MECOM(EVI1),−5 or del(5q); −7; −17/abn(17p)</td>
</tr>
<tr>
<td></td>
<td>Complex karyotype.§ monosomal karyotype¶</td>
</tr>
<tr>
<td></td>
<td>Wild-type NPM1 and FLT3-ITD&lt;sub&gt;high&lt;/sub&gt;†</td>
</tr>
<tr>
<td></td>
<td>Mutated RUNX1¶</td>
</tr>
<tr>
<td></td>
<td>Mutated ASXL1¶</td>
</tr>
<tr>
<td></td>
<td>Mutated TP53#</td>
</tr>
</tbody>
</table>

Frequencies, response rates, and outcome measures should be reported by risk category, and, if sufficient numbers are available, by specific genetic lesions indicated.

*Prognostic impact of a marker is treatment-dependent and may change with new therapies.
†Low, low allelic ratio (<0.5); high, high allelic ratio (≥0.5); semiquantitative assessment of FLT3-ITD allelic ratio (using DNA fragment analysis) is determined as ratio of the area under the curve “FLT3-ITD” divided by area under the curve “FLT3-wild type”; recent studies indicate that AML with NPM1 mutation and FLT3-ITD low allelic ratio may also have a more favorable prognosis and patients should not routinely be assigned to allogeneic HCT.57-59,77
‡The presence of t(9;11)(p21.3;q23.3) takes precedence over rare, concurrent adverse-risk gene mutations.
§Three or more unrelated chromosome abnormalities in the absence of 1 of the WHO-designated recurring translocations or inversions, that is, t(8;21), inv(16) or t(16;16), t(9;11), t(v;11)(q23.3); t(6;9), inv(3) or t(3;3); AML with BCR-ABL1.
¶Defined by the presence of 1 single monosomy (excluding loss of X or Y) in association with at least 1 additional monosomy or structural chromosome abnormality (excluding core-binding factor AML).116
¶¶These markers should not be used as an adverse prognostic marker if they co-occur with favorable-risk AML subtypes.
#TP53 mutations are significantly associated with AML with complex and monosomal karyotype.37,68-69

→ 2017 Risk stratification meaningful for majority of AML patients
AML: Mutated IDH1 and IDH2 Inhibition

**Isocitrate DeHydrogenase (IDH):** conversion of *isocitrate to a-ketoglutarate*

**Mutated IDH:** conversion of *a-ketoglutarate to b/2-hydroxyglutarate*

**Result:** Hypermethylation, altered gene expression, block of differentiation, founder mutations(?), oncogene addiction(?)

### IDH1 mutations
*(found in mitochondria)*
- Prevalence: 5-10%
- Risk stratification:
  - IDH$^{R132}$: adverse

### IDH2 mutations
*(found in cytoplasm)*
- Prevalence: 10-15%
- Risk stratification:
  - IDH$^{R140}$: favorable
  - IDH$^{R172}$: adverse

**Cave:** mutations increased with age, 30% abnormal cytogenetics at diagnosis, mutational context matters

---

**New drugs**
- **AG-120** Phase II
  - ORR 31%
  - CRR 15%
- **IDH 305** Phase I

**New drugs**
- **AG-221** Phase II
  - ORR 41%
  - CRR 18%
• Results consistent across subgroups

AML: Midostaurin – Younger Patients (<60y)

Results Subgroup Analysis

Overall Survival, by FLT3 group, treatment arm

+ Censored

FLT3 Group, Treatment arm
- FLT3-ITD<0.7, Midostaurin
- FLT3-ITD<0.7, Placebo
- FLT3-ITD>=0.7, Midostaurin
- FLT3-ITD>=0.7, Placebo
- FLT3-TKD, Midostaurin
- FLT3-TKD, Placebo

ASH 2015
Survival according to ELN AML risk group 2017

**Mutated RUNX1**

RUNX1 is a critical transcription factor, essential during embryogenesis in HSC generation, and in adulthood during HSC homeostasis and differentiation.

Various missense and frameshift mutations in RUNX1 are identified in AML patients (10-15%), especially in pt with hem disorders or sec AML, advanced age, increased cytopenias ad diagnosis.

Mutations are thought to establish a compromised stem cell phenotype and HSC exhaustion.
Survival according to ELN AML risk group 2017

**Mutated ASXL1 (Additional sex comb-like 1)**

Loss-of-function mutations in the chromatin midifiere gene ASXL1 occur in 10-20% of AML, associated with advanced age, prior hem malignancy, and concurrent runt-related transcription factor-1 (RUNX1) and spliceosome mutations.

ASXL1 is a chromatin-binding protein that interacts with the polycomb repressor complex 2, including H3K27 methyltransferase EZH2.

Mutant ASXL1 decreases polycomb repressor complex recruitment and stabilization, and reduces repressive H3K27 trimethylation marks at various loci.
Table 5. Selected Newer Agents in Clinical Development for the Treatment of AML.

<table>
<thead>
<tr>
<th>Drug Class and Action</th>
<th>Agent</th>
<th>Trial-Registration Number</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Epigenetic modifiers</strong></td>
<td></td>
<td></td>
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<tr>
<td>Decitabine (Dacogen)</td>
<td>NCT01757535</td>
<td></td>
</tr>
<tr>
<td>Azacitidine (Vidaza)</td>
<td>NCT02348489</td>
<td></td>
</tr>
<tr>
<td>Oral azacitidine (CC-486)</td>
<td>NCT01757535</td>
<td></td>
</tr>
<tr>
<td>Guadecitabine (SGI-110)</td>
<td>NCT02348489</td>
<td></td>
</tr>
<tr>
<td>IDH1 inhibitor</td>
<td>AG-120</td>
<td>NCT02074839</td>
</tr>
<tr>
<td>IDH2 inhibitor</td>
<td>AG-221</td>
<td>NCT01915498</td>
</tr>
<tr>
<td>DOT1L inhibitor</td>
<td>EPZ-5676</td>
<td>NCT01684150</td>
</tr>
<tr>
<td>Bromodomain inhibitors</td>
<td>OTX015</td>
<td>NCT01713582</td>
</tr>
<tr>
<td>LSD1 (also called KDM1A inhibitor)</td>
<td>GSK2879552</td>
<td>NCT02177812</td>
</tr>
<tr>
<td>Histone deacetylase inhibitors</td>
<td>Vorinostat</td>
<td>NCT01802333</td>
</tr>
<tr>
<td></td>
<td>Panobinostat</td>
<td>NCT01242774</td>
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<tr>
<td></td>
<td>Pracinostat</td>
<td>NCT01912274</td>
</tr>
<tr>
<td></td>
<td>Valproic acid</td>
<td>NCT00151255</td>
</tr>
<tr>
<td><strong>Tyrosine kinase inhibitors</strong></td>
<td></td>
<td></td>
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<tr>
<td>FLT3 inhibitors</td>
<td></td>
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</tr>
<tr>
<td>First-generation</td>
<td>Midostaurin</td>
<td>NCT00651261; NCT01477606</td>
</tr>
<tr>
<td></td>
<td>Sunitinib</td>
<td>NCT00783653</td>
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<tr>
<td></td>
<td>Sorafenib</td>
<td>NCT00373373, NCT00893373</td>
</tr>
<tr>
<td>Second-generation</td>
<td>Quizartinib</td>
<td>NCT02039726</td>
</tr>
<tr>
<td></td>
<td>Crenolanib</td>
<td>NCT01657682, NCT02298166</td>
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<tr>
<td></td>
<td>ASP2215</td>
<td>NCT02014558</td>
</tr>
<tr>
<td>KIT inhibitors</td>
<td>Dasatinib</td>
<td>NCT02013648; NCT01238211</td>
</tr>
<tr>
<td></td>
<td>Midostaurin</td>
<td>NCT01830361</td>
</tr>
</tbody>
</table>

**Cell-cycle and signaling inhibitors**

|                          |                        |                           |
| MDM2 inhibitor           | Idasanutin (RG-7388)   | NCT01773408               |
| PLK inhibitor            | Volasertib             | NCT01721876               |
| Aurora kinase inhibitors | Barasertib             | NCT00952588               |
| Cyclin-dependent kinase inhibitors | Alvocidib | NCT01413880               |
| Phosphatidylinositol 3-kinase inhibitor | Palbociclib | NCT02310243               |
| PIM kinase inhibitor     | Rigosertib             | NCT01926587               |
| Hedgehog-pathway inhibitors | Vismodegib            | NCT01880437               |
| mTor inhibitors          | PF-04449913            | NCT01546038               |
|                        | Everolimus             | NCT01154439               |
|                        | Temsiroliimus          | NCT01611116               |

**Nuclear export inhibitor**

| XPO1 (also called CRM1) inhibitor | Selinexor (KPT-330) | NCT02088541 |

SGH/SSH Fortbildungskurs Lausanne, 11.11.2017

Drugs Against Potential Functional (Driver) Targets in AML

N ENGL J MED 373;12 NEJM.ORG SEPTEMBER 17, 2015
Analysis of 111 genes in 1540 patients in three prospective trials of intensive therapy