Optimizing the management of FLT3-mutated AML

Mark Levis MD PhD
Disclosures

• Astellas Global Pharma
  • Research funding

• Daiichi-Sankyo
  • Consulting/honoraria

• Novartis
  • Research funding
  • Consulting/honoraria
Outline

• Characterizing FLT3-mutant AML
• Treating FLT3-mutant AML with currently available therapies
• FLT3 inhibitors
FLT3-mutant AML
The mutational landscape of AML

FLT3-mutated AML is common!
FLT3 mutations confer a worse prognosis...

...but this is old data!

Frohling et al Blood 2002; 100:4372
2016 World Health Organization classification of AML

Acute myeloid leukemia (AML) and related neoplasms

<table>
<thead>
<tr>
<th>Acute myeloid leukemia (AML) and related neoplasms</th>
</tr>
</thead>
<tbody>
<tr>
<td>AML with recurrent genetic abnormalities</td>
</tr>
<tr>
<td>AML with t(8;21)(q22;q22.1); RUNX1-RUNX1T1</td>
</tr>
<tr>
<td>AML with inv(16)(p13.1;q22) or t(16;16)(p13.1;q22); CBFB-MYH11</td>
</tr>
<tr>
<td>APL with PML-RARA</td>
</tr>
<tr>
<td>AML with t(9;11)(p21.3;q23.3); MLLT3-KMT2A</td>
</tr>
<tr>
<td>AML with t(6;9)(p23;q34.1); DEK-NUP214</td>
</tr>
<tr>
<td>AML with inv(3)(q21.3;q26.2) or t(3;3)(q21.3;q26.2); GATA2, MECOM</td>
</tr>
<tr>
<td>AML (megakaryoblastic) with t(1;22)(p13.3;q13.3); RBM15-MKL1</td>
</tr>
<tr>
<td><strong>Provisional entity: AML with BCR-ABL1</strong></td>
</tr>
<tr>
<td>AML with mutated <em>NPM1</em></td>
</tr>
<tr>
<td>AML with biallelic mutations of <em>CEBPA</em></td>
</tr>
<tr>
<td><strong>Provisional entity: AML with mutated RUNX1</strong></td>
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<tr>
<td>AML with myelodysplasia-related changes</td>
</tr>
</tbody>
</table>

Therapy-related myeloid neoplasms

<table>
<thead>
<tr>
<th>Therapy-related myeloid neoplasms</th>
</tr>
</thead>
<tbody>
<tr>
<td>AML, NOS</td>
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<tr>
<td>AML with minimal differentiation</td>
</tr>
<tr>
<td>AML without maturation</td>
</tr>
<tr>
<td>AML with maturation</td>
</tr>
<tr>
<td>Acute myelomonocytic leukemia</td>
</tr>
<tr>
<td>Acute monoblastic/monocytic leukemia</td>
</tr>
<tr>
<td>Pure erythroid leukemia</td>
</tr>
<tr>
<td>Acute megakaryoblastic leukemia</td>
</tr>
<tr>
<td>Acute basophilic leukemia</td>
</tr>
<tr>
<td>Acute panmyelosis with myelofibrosis</td>
</tr>
<tr>
<td>Myeloid sarcoma</td>
</tr>
<tr>
<td>Myeloid proliferations related to Down syndrome</td>
</tr>
<tr>
<td>Transient abnormal myelopoiesis (TAM)</td>
</tr>
<tr>
<td>Myeloid leukemia associated with Down syndrome</td>
</tr>
</tbody>
</table>

How do FLT3 mutations fit into this?
FLT3 mutations make everything worse!

- Acute promyelocytic leukemia
  - FLT3 mutated in ~30%
  - Higher relapse rate, worse survival
    - *Ann hematol* 2014;93:2001-10

- NPM1
  - FLT3-mutated in ~30%
  - FLT3-ITD confers higher relapse rate, worse survival
    - *Blood.* 2008;111:2776-2784

- Core-binding factor AML
  - FLT3 mutated in ~20%
  - Higher relapse rate

- Bcl-2 inhibitors (venetoclax)
  - FLT3-ITD mutations confer resistance
    - ASH 2017 abstract #1348
The FLT3 receptor bound to FLT3 ligand (FL)

Juxtamembrane Domain
Autoinhibition of FLT3 kinase activity and location of ITD mutations

Activation Loop:
Activating mutations at D835
“FLT3-TKD mutations”
Analogous to similar mutations in PDGFR, c-Kit
FLT3-ITD Signaling

- Akt
  - PIP₃
  - PIP₂
- PI-3 kinase
- SOS
- Grb2
- Ras
- Raf
- Mek
- ERK
- Stat5
- P-FLT3
- FLT3
- Growth
- Anti-apoptosis
FLT3 mutations occur relatively late in leukemogenesis

Sci Transl Med. 2012 August 29; 4(149): 149ra118
Clonal Evolution of AML by Molecular Profiling

## Risk category

<table>
<thead>
<tr>
<th>Category</th>
<th>Genetic Abnormality</th>
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</thead>
</table>
| **Favorable** | t(8;21)(q22;q22.1); *RUNX1-RUNX1T1*<sup>†</sup>  
inv(16)(p13.1q22) or t(16;16)(p13.1;q22); *CBFB-MYH11*<sup>‡</sup>  
Mutated *NPM1* without *FLT3-ITD* or with *FLT3-ITD*<sup>low</sup>  
Biallelic mutated *CEBPA* |
| **Intermediate** | Mutated *NPM1* and *FLT3-ITD*<sup>high</sup>  
Wild-type *NPM1* without *FLT3-ITD* or with *FLT3-ITD*<sup>low</sup> (without adverse-risk genetic lesions)  
t(9;11)(p21.3;q23.3); *MLLT3-KMT2A*<sup>‡</sup> |
| **Adverse** | t(6;9)(p23;q34.1); *DEK-NUP214*  
t(v;11q23.3); *KMT2A* rearranged  
t(9;22)(q34.1;q11.2); *BCR-ABL1*  
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,<sup>§</sup> monosomal karyotypell  
Wild-type *NPM1* and *FLT3-ITD*<sup>high</sup>  
Mutated *RUNX1*<sup>¶</sup>  
Mutated *ASXL1*<sup>¶</sup>  
Mutated *TP53*<sup>‡</sup> |

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<sup>†</sup> Dohner et al Blood. 2017; 129(4): 424-447
FLT3 coding sequence

JM

C-lobe

N-lobe
An internal duplication, inserted in tandem = FLT3-ITD mutation
Current method of PCR for FLT3-ITD mutations

Wild type allele

45 bp

330 bp

Mutant allele

45 bp

45 bp

375 bp

35 cycles PCR

Murphy et al
J Mol Diagn. 2003;5:96

WT

ITD

Size (bp)→ 330 375
Wild type allele

45 bp

Mutant allele

45 bp

45 bp

330 bp

375 bp

Current method of PCR for FLT3-ITD mutations

28 cycles PCR

Thiede et al, Blood 2002 99:4326
Current method of PCR for FLT3-ITD mutations

Wild type allele

Mutant allele

Primer

330 bp

375 bp

50 cycles PCR

WT

ITD

PCR assay not helpful for MRD!
Conclusion: FLT3-ITD allelic ratio <0.51 ratio means patient will benefit from transplant…

Methods of determining FLT3-ITD allelic ratio are NOT STANDARDIZED!

Conclusion: FLT3-ITD allelic ratio <0.51 ratio means no transplant is needed…
Treatment of FLT3-mutant AML
Newly-diagnosed FLT3-mutated AML patient → Induction → Consolidation → Cure!
Induction

Newly-diagnosed FLT3-mutated AML patient

Which anthracycline? Which dose?

All patients
AraC + Ida (12 mg) vs. AraC + Daunorubicin (90 mg)

FLT3-ITD

Lee et al. *J Clin Oncol* 35:2754
Newly-diagnosed FLT3-mutated AML patient

Induction

Consolidation

Chemotherapy or allogeneic transplant?
The role of allogeneic transplant for FLT3/ITD AML: Retrospective studies

<table>
<thead>
<tr>
<th>Study</th>
<th>Location</th>
<th>Number of patients</th>
<th>Transplant Beneficial?</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gale et al&lt;sup&gt;1&lt;/sup&gt;</td>
<td>United Kingdom</td>
<td>283</td>
<td>No</td>
</tr>
<tr>
<td>Bornhauser et al&lt;sup&gt;2&lt;/sup&gt;</td>
<td>Germany</td>
<td>175</td>
<td>Yes</td>
</tr>
<tr>
<td>Schlenk et al&lt;sup&gt;3&lt;/sup&gt;</td>
<td>Germany/Austria</td>
<td>164</td>
<td>Yes</td>
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<tr>
<td>Dezern et al&lt;sup&gt;4&lt;/sup&gt;</td>
<td>USA</td>
<td>31</td>
<td>Yes</td>
</tr>
<tr>
<td>Labouret al&lt;sup&gt;5&lt;/sup&gt;</td>
<td>France</td>
<td>24</td>
<td>Yes</td>
</tr>
<tr>
<td>Lin et al&lt;sup&gt;6&lt;/sup&gt;</td>
<td>Taiwan</td>
<td>34</td>
<td>Yes</td>
</tr>
<tr>
<td>Brunet et al&lt;sup&gt;7&lt;/sup&gt;</td>
<td>International</td>
<td>120</td>
<td>Yes</td>
</tr>
</tbody>
</table>

4. *Biol Blood Marrow Transplant*. 2011;17(9):1404-1409

International Consensus
Allogeneic transplant is preferred, when feasible
Allogeneic transplant eliminates the negative prognostic effect of FLT3-ITD mutations

Adapted from: Frohling et al Blood 2002; 100:4372
Most patients not transplanted

66% CR rate
85% of patients underwent alloTx in CR1
Some use of FLT3 inhibitors
Survival after allogeneic transplant is worse for FLT3-ITD AML patients

Brunet et al *J Clin Oncol*. 2012. 30:735

Newly-diagnosed FLT3-mutated AML patient

Induction

Induction Failure

Allogeneic Tx

Relapse

• Survival after relapse is worse for FLT3-ITD AML patients.
  • How to prevent relapse?
  • How to treat relapsed/refractory disease?

Ravandi et al. Leuk Res 2010. 34:752
FLT3 inhibitors
TKIs that have been studied as FLT3 inhibitors:

- Sorafenib
- Lestaurtinib
- Midostaurin
- TAK-659
- Sunitinib
- Quizartinib
- Cabozantanzanib
- Gilteritinib
- PLX3397
- KW2449
- Crenolanib
- FF10101
TKIs that have been studied as FLT3 inhibitors:

- Sorafenib
- Midostaurin
- Quizartinib
- Gilteritinib
- Crenolanib
Kinase selectivity profiles of FLT3 inhibitors

Adapted from Davis et al; *Nat Biotechnology* 2011; 29(11):1046-1051
Randomized trials of FLT3 inhibitors

<table>
<thead>
<tr>
<th>Drug</th>
<th>Trial</th>
<th>Regimen</th>
<th>Population</th>
<th>Trial number</th>
<th>Start date</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sorafenib</td>
<td>Sorafenib in elderly AML</td>
<td>Chemo +/- sorafenib</td>
<td>Untreated AML &gt; 60</td>
<td>NCT00373373</td>
<td>Completed</td>
</tr>
<tr>
<td>Sorafenib</td>
<td>SORAML</td>
<td>Chemo +/- sorafenib</td>
<td>Untreated AML &lt; 60</td>
<td>NCT00893373</td>
<td>Completed</td>
</tr>
<tr>
<td>Midostaurin</td>
<td>RATIFY</td>
<td>Chemo +/- midostaurin</td>
<td>Untreated FLT3 mutant AML &lt;60</td>
<td>NCT00651261</td>
<td>Completed</td>
</tr>
<tr>
<td>Quizartinib</td>
<td>QuANTUM-R</td>
<td>Chemo +/- quizartinib</td>
<td>Relapsed FLT3-ITD AML</td>
<td>NCT02039726</td>
<td>Enrollment completed</td>
</tr>
<tr>
<td>Quizartinib</td>
<td>QuANTUM-First</td>
<td>Chemo +/- quizartinib</td>
<td>Untreated FLT3-ITD AML</td>
<td>NCT02668653</td>
<td>October 2016</td>
</tr>
<tr>
<td>Gilteritinib</td>
<td>Admiral</td>
<td>Chemo vs. gilteritinib</td>
<td>Relapsed FLT3 mutant AML</td>
<td>NCT02421939</td>
<td>October 2015</td>
</tr>
<tr>
<td>Gilteritinib</td>
<td>Lacewing</td>
<td>Azacitidine +/- gilteritinib</td>
<td>Untreated FLT3 mutant AML, unfit</td>
<td>NCT02752035</td>
<td>June 2016</td>
</tr>
<tr>
<td>Gilteritinib</td>
<td>Gossamer</td>
<td>Post-chemo maintenance</td>
<td>FLT3 mutant AML in CR1</td>
<td>NCT02927262</td>
<td>October 2016</td>
</tr>
<tr>
<td>Gilteritinib</td>
<td>Morpho</td>
<td>Post-transplant maintenance</td>
<td>FLT3-ITD AML post-allo Tx</td>
<td>NCT02997202</td>
<td>May 2017</td>
</tr>
<tr>
<td>Crenolanib</td>
<td>AMLSG-19-13</td>
<td>Chemo +/- crenolanib</td>
<td>Relapsed FLT3 mutant AML</td>
<td>NCT02298166</td>
<td>March 2017</td>
</tr>
</tbody>
</table>

Randomized trials of FLT3 inhibitors
Midostaurin

- A multi-targeted kinase inhibitor
  - In vitro activity against FLT3 in leukemia cells
  - Active against both ITD and TKD mutations

Partial FLT3 inhibition in vivo

Pharmacokinetics:
Highest levels during first month

Strati et al. Am J Hematol. 2015 90:276-81

RATIFY: Induction chemotherapy +/- midostaurin for newly diagnosed FLT3-mutated AML

Stratify* FLT3 ITD or TKD

FLT3 WILD TYPE not eligible for enrollment

Study drug is given on Days 8-21 after each course of chemotherapy, and Days 1-28 of each 28 day Maintenance cycle.

* Stratification: TKD; ITD with allelic ratio <0.7; ITD with allelic ratio >=0.7
Midostaurin prolongs survival for patients with newly-diagnosed FLT3-mutant AML

- 5 year survival rate:
  - Midostaurin 50.9% vs. Placebo 43.3%

Midostaurin effect on survival was similar across all FLT3 subtypes

Event-free survival

- Event: no CR within 60 days, relapse or death
- 5 year EFS rate: Midostaurin 27.6% vs. Placebo 19.3%

Best survival:
Treated with midostaurin and transplanted in first remission
Midostaurin

• Received regulatory approval from U.S. FDA and EMA
  • Newly diagnosed FLT3-mutated AML in combination with standard cytarabine and daunorubicin induction and cytarabine consolidation.
  • Not indicated as a single agent
  • Approved for use in systemic mastocytosis
  • EMA indication includes maintenance
• Prolongs survival when given day 8-21 of induction and consolidation
  • Effective in FLT3-ITD and FLT3-TKD AML
  • Most of the benefit probably occurs EARLY
  • Best survival in patients receiving midostaurin with induction, then allo transplant in CR1

Midostaurin

No activity as a single agent in relapsed FLT3 mutant AML:

CR rate: 0%
PR rate 3%

Fischer et al. J Clin Oncol. 28:4339-4345

No benefit in maintenance

Larson et al ASH 2017

Figure 1: Landmark analysis of DFS during the 12 cycles of maintenance, censoring pts at the time they completed the planned maintenance or discontinued study drug early.
Induction of a Newly-diagnosed FLT3-mutated AML patient with Midostaurin may lead to Allogeneic Tx. However, the effectiveness of Midostaurin in this context is uncertain.
TKIs that have entered phase 3 trials for FLT3-mutated AML:

- Sorafenib
- Midostaurin
- Quizartinib
- Gilteritinib
- Crenolanib
Midostaurin

IC$_{50}$ in plasma: 700 nM

Quizartinib

IC$_{50}$ in plasma: 18 nM


Patient treated with Midostaurin:
% Control: 100 47

Patient treated with Quizartinib:
% Control: 100 0

In vivo FLT3 inhibition

P-FLT3

FLT3

Pre Post

P-FLT3

FLT3

Pre Post
• Phase 2 results for quizartinib in relapsed and refractory FLT3-ITD AML:
  • Overall response rate (CR/CRi/CRp/PR): 74-77%
  • Composite CR rate (CR/CRi/CRp): 46-56%
  • Cortes et al. *Lancet Oncol.* 2018; (in press)
Monotherapy: terminal myeloid differentiation in the marrow

Pre-treatment  Day 15  Day 29

No change in cellularity or mutant allelic burden

Sexauer et al. Blood 2012; 120:4205

Quizartinib

Resistance-conferring point mutations emerge during quizartinib treatment:

-Mostly FLT3-TKD mutations…

<table>
<thead>
<tr>
<th>Subject number</th>
<th>Age</th>
<th>Gender</th>
<th>New Mutation at Relapse</th>
<th>Weeks on Study</th>
</tr>
</thead>
<tbody>
<tr>
<td>1005-004</td>
<td>60</td>
<td>F</td>
<td>F691L</td>
<td>19</td>
</tr>
<tr>
<td>1005-006</td>
<td>43</td>
<td>M</td>
<td>D835Y</td>
<td>6</td>
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<tr>
<td>1005-007</td>
<td>59</td>
<td>F</td>
<td>D835V</td>
<td>23</td>
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<tr>
<td>1005-009</td>
<td>68</td>
<td>M</td>
<td>D835Y</td>
<td>18</td>
</tr>
<tr>
<td>1005-010</td>
<td>52</td>
<td>M</td>
<td>F691L</td>
<td>19</td>
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<td>1009-003</td>
<td>75</td>
<td>F</td>
<td>D835Y</td>
<td>12</td>
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<tr>
<td>1009-007</td>
<td>64</td>
<td>F</td>
<td>F691L, D835V</td>
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<tr>
<td>1011-006</td>
<td>70</td>
<td>M</td>
<td>D835Y</td>
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<tr>
<td>1011-007</td>
<td>56</td>
<td>F</td>
<td>D835Y</td>
<td>8</td>
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</tbody>
</table>

Type 1 FLT3 inhibitors: Active against both ITD and TKD mutations

<table>
<thead>
<tr>
<th>Cell line</th>
<th>Crenolanib IC$_{50}$ pFLT3 (nM)</th>
<th>Quizartinib IC$_{50}$ pFLT3 (nM)</th>
<th>Fold difference</th>
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</thead>
<tbody>
<tr>
<td>Ba/F3 ITD</td>
<td>1.3</td>
<td>1.7</td>
<td>1.2</td>
</tr>
<tr>
<td>Ba/F3 ITD/D835Y</td>
<td>8.7</td>
<td>93.1</td>
<td>10.7</td>
</tr>
<tr>
<td>Ba/F3 WT D835Y</td>
<td>6.9</td>
<td>33.7</td>
<td>4.9</td>
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<tr>
<td>Ba/F3 WT D835F</td>
<td>6.5</td>
<td>72.7</td>
<td>11.2</td>
</tr>
<tr>
<td>Ba/F3 WT D835H</td>
<td>19.8</td>
<td>20.0</td>
<td>—</td>
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<tr>
<td>Ba/F3 WT D835N</td>
<td>4.3</td>
<td>2.3</td>
<td>0.5</td>
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<tr>
<td>Ba/F3 WT D835V</td>
<td>2.3</td>
<td>63.7</td>
<td>27.4</td>
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<tr>
<td>Ba/F3 ITD/F691L</td>
<td>67.8</td>
<td>36.4</td>
<td>0.5</td>
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</tbody>
</table>

<table>
<thead>
<tr>
<th>FLT3 receptor subtype</th>
<th>Gilteritinib IC$_{50}$ (nM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wild type</td>
<td>5 nM</td>
</tr>
<tr>
<td>Molm14 (ITD)</td>
<td>1.8 nM</td>
</tr>
<tr>
<td>TF/ITD</td>
<td>1.4 nM</td>
</tr>
<tr>
<td>Ba/F3 ITD</td>
<td>0.7 nM</td>
</tr>
<tr>
<td>Ba/F3 D835Y</td>
<td>0.5 nM</td>
</tr>
<tr>
<td>Ba/F3 D835H</td>
<td>1.9 nM</td>
</tr>
<tr>
<td>Ba/F3 D835V</td>
<td>0.7 nM</td>
</tr>
<tr>
<td>Ba/F3/ITD/F691L</td>
<td>17.6 nM</td>
</tr>
</tbody>
</table>

Smith et al. *PNAS.* 2014;111:5319
Lee et al. *Blood.* 2017;129:257
FLT3 phosphorylation

Marrow blast reduction

Clinical response

Gilteritinib

Newly-diagnosed FLT3-mutated AML patient

Induction

Induction Failure

Quizartinib

Gilteritinib

Crenolanib

Allogeneic Tx

Relapse
<table>
<thead>
<tr>
<th>Quizartinib</th>
<th>Gilteritinib</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>“QuANTUM-R”</strong></td>
<td><strong>“Admiral”</strong></td>
</tr>
<tr>
<td>- Quizartinib versus salvage chemotherapy for relapsed/refractory FLT3-ITD AML.</td>
<td>- Gilteritinib versus salvage chemotherapy for relapsed/refractory FLT3-mutant AML.</td>
</tr>
<tr>
<td>- Randomized, Phase 3</td>
<td>- Randomized, Phase 3</td>
</tr>
<tr>
<td>- Accrual complete, results expected 2018</td>
<td>- Accrual complete, results expected 2018</td>
</tr>
</tbody>
</table>
Relapsed FLT3-mutated AML:

Treated with salvage chemotherapy

Median survival ~5 months

Treated with gilteritinib monotherapy

Median survival ~7 months
AMLSG-19-13 (NCT02298166) currently accruing…

- Relapsed/refractory FLT3-mutated AML
- Patients 18-60 years of age
- Endpoints: Overall Survival, Event-free Survival
- Salvage chemotherapy:
  - cytarabine 1000 mg/m2/day days 1-6
  - mitoxantrone 10 mg/m2/day days 1-3
- Crenolanib or placebo starting day 7
- Maintenance crenolanib/placebo after allogeneic transplant
Paradigm for treating a patient with FLT3-mutated AML

Newly-diagnosed patient

Induction chemotherapy
- Initiate search for HLA-compatible donor

Complete Remission
- High dose AraC
  - ONLY until allogeneic transplant can be arranged

FLT3 inhibitor

Maintenance therapy?

Allogeneic transplant
QuANTUM-First
NCT02668653

- Newly-diagnosed FLT3-ITD+ AML
- Age 18-75
- Induction chemotherapy (7+3) + Quizartinib or placebo
- Primary endpoint: Event Free Survival
FLT3-ITD AML at Johns Hopkins

Age 18-59
FLT3-ITD only
May 2008–present

66% CR rate
85% of patients
underwent alloTx

2009–present
FLT3-ITD, age 18-69
65% CR rate
89% underwent alloTx in CR1

Metzelder et al. Leukemia. 2012; 26:2353
• Does maintenance therapy with a FLT3 inhibitor even work?
  • No randomized studies…

• Which patients need maintenance therapy?
  • Many patients are already cured after allogeneic transplant.

• Can we use minimal residual disease (MRD) as a guide?
  • Is FLT3 a useful marker for minimal residual disease?
FLT3 mutations occur relatively late in leukemogenesis

Sci Transl Med. 2012 August 29; 4(149): 149ra118
The FLT3-ITD is a fingerprint for the patient’s disease

Amino acid sequences of 30 patients with FLT3-ITD mutations
A rationale for the use of FLT3-ITD mutations as a marker of MRD:

- Patients with FLT3-ITD AML are prone to relapse
- The ITD mutation essentially represents the disease:
  - Occurs late in leukemogenesis
  - Unique to each patient
  - Specificity therefore is high
    - Sensitivity....?
A hybrid PCR/Next-Gen Sequencing assay for MRD for FLT3/ITD AML

Exons 14 and 15 of FLT3 are amplified by PCR. Products detected by a refined NGS technique.

Sensitivity equivalent to detection of at least one ITD-containing cell out of 10,000 with a minimum input of 100,000 cell equivalents of DNA.

Assay validated using marrow samples from patients with FLT3/ITD AML in CR

Levis et al 2018 Blood Advances (in press)
Results from cell lines

- MRD assay is linear to $10^{-4}$
- Sensitive down to $10^{-5}$ and lower

Clinical Examples of MRD Assay

- **44 yo woman**
  - 48 bp FLT3/ITD
  - Allelic ratio 0.75
  - Induction: AraC/Ida/VP16
  - Marrow aspirate: Morphologic CR
  - FLT3/ITD negative
  - MRD assay: 48 bp ITD mutation, 1.7 x 10^{-4}

- **68 yo woman**
  - 24 bp FLT3/ITD
  - Allelic ratio 0.37
  - Induction: AraC/Ida (7+3)
  - Marrow aspirate: Morphologic CR
  - FLT3/ITD negative
  - MRD assay: 24 bp ITD mutation, 1.1 x 10^{-4}

- **47 yo man**
  - 72 bp FLT3/ITD
  - Allelic ratio 0.05
  - Induction: AraC/Ida/sorafenib (7+3)
  - Marrow aspirate: Morphologic CR
  - FLT3/ITD negative
  - MRD assay: 72 bp ITD mutation, 2.8 x 10^{-5}

- **60 yo man**
  - 15, 39 bp FLT3/ITD
  - Allelic ratios 0.05, 0.54
  - Induction: AraC/Ida (7+3)
  - Marrow aspirate: Morphologic CR
  - FLT3/ITD negative
  - MRD assay: 15, 39 bp ITD mutations, 1.35 x 10^{-5}, 3.33 x 10^{-4}

**All 4 have undergone HCT**
BMT CTN 1506
A Multi-Center, Randomized, Double-Blind, Placebo-Controlled Phase III trial of the FLT3 Inhibitor Gilteritinib Administered as Maintenance Therapy Following Allogeneic Transplant for Patients with FLT3-ITD AML

Study Chairs: Yi-Bin Chen, MD, Mark Levis, MD, PhD
BMT-CTN 1506/Morpho:
346 post-transplant FLT3-ITD AML patients

173 patients
Placebo

173 patients
Gilteritinib

Is there a benefit to FLT3 inhibition post-transplant?

Does the detection of a FLT3/ITD mutation by a validated, sensitive MRD assay predict relapse?

Does a potent FLT3 inhibitor prevent relapse when the MRD assay detects a FLT3/ITD mutation?
The management of FLT3-mutated AML…

• …is changing!

• Midostaurin administered with induction and consolidation chemotherapy prolongs survival.

• Early allogeneic transplant is an essential component of treatment.

• Results from two phase 3 trials for relapsed FLT3-mutated AML will be available this year.
  • Quizartinib
  • Gilteritinib

• FLT3 inhibitors are being tested in all phases of treatment.