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Acute myeloid leukemia - Section 2

# *Targeting mutated FLT3 in acute myeloid leukemia*

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## Take-home messages

- FLT3-mutated AML evolves during therapy, with FLT3-addicted clones emerging at relapse.
- FLT3 inhibition is likely to be incorporated into the management of FLT3-mutated AML in the very near future, but the roles of selective versus non-selective inhibitors remains to be determined.

## Introduction

Acute myeloid leukemia (AML) develops from mutations arising within the genomes of hematopoietic stem/progenitor cells. Our current understanding of this process is that founding mutations confer a proliferative advantage to stem cell clones, and the progressive occurrence of cooperating mutations leads to eventual transformation and overt clinical disease.[1] FLT3, which codes for a receptor tyrosine kinase, is one of the most commonly mutated genes in AML,[2] and because these mutations are associated with constitutive activation of that receptor, inhibiting FLT3 has been a goal of numerous clinical studies for several years now. This short review will summarize the current state of this field, and will offer some perspectives on the future of targeting FLT3 in AML.

## Current state of the art

There are two types of FLT3 mutations. The first type, occurring in ~23% of newly-diagnosed AML cases, consists of internal tandem duplications (ITDs), in which duplicated sequence (3-200+ base pairs) is inserted in tandem, and in-frame, into the region coding for the juxtamembrane domain (and sometimes extending into the kinase domain).[3,4] The resulting additional amino acid sequence disrupts the auto-inhibitory function of this domain and leads to constitutive receptor activation. It is important to note, however, that FLT3 receptors with an ITD mutation are still dependent on the cognate ligand, FLT3 ligand (FL), for full activation, and that the addition of FL to FLT3-ITD AML blasts *in vitro* has proliferative and anti-apoptotic effects.[5] The second type of FLT3 mutations occur in 7% of patients at diagnosis and consists of point mutations in the tyrosine kinase domain (TKD).[3] While FLT3-TKD mutations also cause constitutive activation, the signaling of FLT3-TKD receptors is less aberrant as compared to FLT3-ITD receptors. FLT3-TKD mutations probably have a mild negative effect on prognosis (although this remains somewhat controversial), while FLT3-ITD mutations confer a clear negative impact on survival. AML patients with FLT3-ITD mutations typically present with pronounced leukocytosis, and while cytarabine-based induction therapy will often result in remission, these patients relapse more frequently, and relapse earlier, than AML patients with wild type FLT3.

FLT3 mutations were first identified in 1996, and their prognostic impact was established after 2002, following the publication of a series of retrospective studies.[3] At that time, the only small molecule tyrosine kinase inhibitors (TKIs) available were compounds that had been developed for other molecular targets. Midostaurin was first introduced as an inhibitor of protein kinase C (PKC) almost 30 years ago,[6] while lestaurtinib had been initially studied as an inhibitor of TrkA.[7] In reality, both compounds are highly non-selective, inhibiting many other kinases (and likely other ATP-binding enzymes) as well. Neither drug was particularly effective as monotherapy for FLT3-mutated AML.[8,9] Trials of lestaurtinib combined with chemotherapy for newly-diagnosed or relapsed patients yielded no benefit.[10,11] On the other hand, the recent results of CALGB 10603 ("RATIFY") indicate that midostaurin administered immediately after a standard 3 + 7 induction improves response rates and overall survival for newly-diagnosed patients with either FLT3-ITD or FLT3-TKD mutations.[12] Sorafenib, a drug initially developed as a RAF kinase and VEGF-R inhibitor, can induce compete responses as monotherapy in relapsed FLT3-ITD AML,[13] and has shown remarkable activity in the post-allogeneic transplant setting or in combination with azacitidine.[14,15] While all of these studies were proceeding, quizartinib emerged as the first TKI specifically designed to target FLT3,[16] and it displayed a high level of activity as a single agent in the relapsed setting.[17] Patients

responding to quizartinib quickly developed resistance-conferring FLT3-TKD mutations,[18] and so the next-generation FLT3 TKIs, crenolanib and gilteritinib (both of which were also effective against TKD mutations) were introduced.[19,20]

The pre-clinical and clinical studies of these drugs have provided us with considerable insight into the biology of FLT3-ITD AML. When we merge these insights with the data provided from next generation sequencing studies of AML mutations, we have a clearer understanding of the problem and how best to approach it. At diagnosis, like all AML, FLT3-ITD AML is polyclonal, and the majority of the clones are not dependent on mutant FLT3 signaling for survival. Next generation sequencing (NGS) studies on AML samples indicate that several sub-clones are present at diagnosis,[21] and *in vitro* studies of FLT3-ITD AML blasts suggest that only a subset of these clones are dependent on mutant FLT3 signaling for survival.[22] At relapse, the disease is more oligoclonal, and the blasts display far greater sensitivity *in vitro* to FLT3 inhibition, highlighting the need for potent selective inhibitors such as quizartinib or gilteritinib in this setting. The underlying mechanism for this selection is not known, but it is conceivable that FL plays a role. Plasma levels of FL increase exponentially during repeated rounds of aplasia-inducing chemotherapy,[23] and it is possible that this constant 'bath' of the very cytokine which the blasts depend on for survival selects for the outgrowth of FLT3-addicted clones.

## Future perspectives

There are 9 completed or actively accruing phase 3/pivotal trials of FLT3 TKIs for the treatment of FLT3-mutated AML (Table 1). CALGB10603 met its primary endpoint of improved survival, and so FLT3 inhibition with a first generation inhibitor may well be a part of standard AML therapy in the near future. Assuming eventual approval of the later-generation drugs, how should we incorporate FLT3 inhibition into the current treatment paradigms? In keeping with the conventional approach used in BCR-ABL-driven acute leukemia, it seems likely that FLT3 inhibitors will be used early and continuously into induction, consolidation, and maintenance therapy (with or without allogeneic transplant) for FLT3-mutated AML. The uncertainty lies in the decision as to which drug to use, and when to use it. Data from CALGB 10603 suggests that midostaurin following induction chemotherapy produces more frequent and deeper responses, but its benefits in the maintenance setting seem much less clear, and it probably has no role in relapse.[12] In relapsed FLT3-ITD patients, potent selective like quizartinib and gilteritinib produce gratifyingly rapid responses, but these responses are incomplete and often short-lived. Interestingly, the combination of FLT3 inhibitors with azacitidine may be the most effective approach in the salvage setting,[15,24] although combinations with conventional intensive chemotherapy regimens is currently being studied. Finally, allogeneic transplant has emerged as a preferred consolidation for FLT3-ITD AML, and FLT3 inhibition appears to synergize with the allogeneic effect. Post-transplant maintenance with a selective, well-tolerated FLT3 TKI is a logical approach, and the benefit of this will be determined by an international trial just getting underway (BMT-CTN 1506).

**Table 1. Phase 3 trials of FLT3 inhibitors for the treatment of FLT3-mutated AML. UK MRC = United Kingdom Medical Research Council. CALGB = Cancer and Leukemia Group B. AMLSG = AML Study Group. BMT-CTN = Bone Marrow Transplant Clinical Trials Network.**

| Trial Name | Sponsor | Treatment | Population | Clinical trial number | Accrual start date |
|---|---|---|---|---|---|
| 204 | Cephalon | Chemotherapy +/- lestaurtinib | Relapsed FLT3-mutated AML | NCT00079482 | Completed |
| AML15/17 I | UK MRC | Chemotherapy +/- lestaurtinib | Untreated FLT3-mutated AML | ISRCTN17161961 SRCTN55675535 | Completed |
| CALGB10603 (RATIFY) | CALGB/Novartis | Chemotherapy +/- midostaurin | Untreated FLT3-mutated AML | NCT00651261 | Completed |
| QUANTUM-R | Daiichi- Sankyo | Chemotherapy vs. quizartinib | Relapsed FLT3-ITD AML | NCT02039726 | April 2014 |
| 'Admiral' | Astellas | Chemotherapy vs. gilteritinib | Relapsed FLT3-mutated AML | NCT02421939 | October 2015 |
| 'Lacewing' | Astellas | Azacitidine +/-gilteritinib | Untreated FLT3-mutated AML, less fit | NCT02752035 | June 2016 |
| 'Gossamer' | Astellas | Giltertinib maintenance | Untreated FLT3-mutated AML | NCT02927262 | October 2016 |
| AMLSG 19-13/ARO-007 | AMLSG/Arog | Chemotherapy +/- crenolanib | Relapsed FLT3-mutated AML | NCT02298166 | January 2017 |
| BMT-CTN1506/'Morpho' | BMT-CTN, Astellas | Giltertinib maintenance | FLT3-mutated AML s/p allo transplant | NCT02997202 | May 2017 |

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