Blood Spotlight

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Midostaurin, enasidenib, CPX-351, gemtuzumab ozogamicin, and venetoclax bring new hope to AML

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In 2017, 4 drugs received US Food and Drug Administration marketing approval for acute myeloid leukemia (AML) treatment: targeted therapies for mutant *FLT3* and *IDH2*, a liposomal cytarabine-daunorubicin formulation for therapy-related AML and AML with myelodysplasia-related changes, and resurgence of an antibody-drug conjugate designed to target CD33. Promising results also emerged for the BCL-2 inhibitor venetoclax combined with low-intensity therapy in older patients unfit for intensive chemotherapy. This quintet of new drugs is likely to reshape the therapeutic landscape of AML. (*Blood*. 2017;130(23):2469-2474)

Introduction

Once touted as the "boulevard of broken dreams," acute myeloid leukemia (AML) has been a therapeutic graveyard for a litany of failed drug development programs attempting to reform the AML treatment landscape.[1] In this article, we highlight a selection of emerging drugs starting to make an impact in the care of patients with AML.

Midostaurin (Novartis Pharmaceuticals, Inc.)

Clinical impact in AML

Midostaurin, an *N*-benzoyl staurosporine analog derived from *Streptomyces staurosporeus*, was initially characterized as an inhibitor of protein kinase C. Found to inhibit C-FOS and MAPK, it was also shown to have multikinase inhibitory activity against platelet-derived growth factor receptor, CDK1, KIT, and vascular endothelial growth factor.[2] A drug screen identified midostaurin to have FLT3 inhibitory activity, which led to its repurposing as a drug for *FLT3* mutant AML.[3] A decade-long clinical development journey culminated in a pivotal trial (Randomized AML trial in FLT3 in patients less than 60 years [RATIFY]), which combined midostaurin or placebo with standard induction and consolidation chemotherapy, followed by 12 months of midostaurin or placebo maintenance for adults with *FLT3*-ITD and *FLT3*-TKD mutant AML.[4] This global effort, which involved 13 AML cooperative groups, demonstrated that midostaurin significantly improved 4-year overall survival (OS) from 44.3% to 51.4% (hazard ratio, 0.78; $P = .009$), compared with placebo.[4] This led to US Food and Drug Administration (FDA) approval of midostaurin, with the benefit of midostaurin observed in patients with *FLT3*-ITD low (0.05-0.7) and high allelic ratio (>0.7), as well as in patients with *FLT3*-TKD.[4] These mutations are collectively found in up to 40% of the AML population.[5-7] Although FLT3 testing in the RATIFY trial was performed by academic laboratories, LeukoStrat CDx *FLT3* Mutation Assay (Invivoscribe Technologies, Inc.) was approved in parallel as a companion assay for the detection of *FLT3* mutant AML in the United States by the FDA. Rapid screening for both *FLT3*-ITD and *FLT3*-TKD at diagnosis will now be routinely required to effectively incorporate midostaurin into the standard of care for AML. Furthermore, the FDA label does not restrict use to patients 18 to 59 years of age, as occurred in RATIFY. Therefore, routine screening and treatment of mutant FLT3 is likely to extend to older populations fit for intensive chemotherapy.

Future research questions and challenges

Several questions remain, however, regarding the optimal use of midostaurin in AML. The median exposure of patients to midostaurin was only 42 days, suggesting that the main benefit was derived early on in treatment.[4] The magnitude of benefit and optimal duration of midostaurin as maintenance therapy after completion of chemotherapy is contentious. Further study to demonstrate a significant survival outcome is likely to require a formidable number of patients and time. A more feasible objective may be demonstration that maintenance treatment can effectively eliminate minimal residual disease and prolong relapse-free survival.

Concurrent *NPM1* mutation partially mitigates the adverse prognostic impact of *FLT3*-ITD,[8] and future post hoc analyses to examine the magnitude of midostaurin benefit in various *FLT3*-ITD/*NPM1* subgroups is warranted. The RATIFY study also suggested greater benefit for midostaurin in males, but not females, with *FLT3*-ITD and conversely, for females, but not males, with *FLT3*-TKD.[4] The data also indicated that males had a worse baseline outcome than females with *FLT3*-ITD. Further work is needed to confirm and unravel these gender conundrums.

In the RATIFY study, posttransplant survival was marginally better in those receiving prior midostaurin ($P = .07$), with benefit limited to patients transplanted in first remission.[4] Future work should verify whether midostaurin delivers more patients to transplant in first remission without minimal residual disease and determine whether the addition of midostaurin or other FLT3 inhibitors to the postallogeneic stem cell transplant (SCT) setting will lead to further improvements in survival.

The recent FDA approval of midostaurin as frontline therapy for *FLT3*-mutant AML may create hurdles for new FLT3 inhibitors seeking first-line drug registration because the control arm will need to

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include midostaurin, thereby setting a higher clinical bar for new investigational agents to surpass. With midostaurin as the comparator, it remains to be seen whether the greater FLT3 potency associated with newer generation inhibitors, such as quizartinib, crenolanib, and gilteritinib, will have more clinical relevance than the multikinase effects of midostaurin in randomized head-to-head studies. To date, it is not clear why midostaurin succeeded, whereas other multikinase FLT3 inhibitors, such as lestaurtinib did not.[9] Midostaurin's multikinase mechanism of action is likely to see it explored in combination with standard chemotherapy in *FLT3* wild-type AML. This will follow the lead of sorafenib, another multikinase inhibitor, which improved event-free survival in unselected adults with AML when combined with intensive chemotherapy.[10]

Enasidenib (Celgene and Agios Pharmaceuticals, Inc.)

Clinical impact in AML

First identified from the whole genome sequence of an index patient with AML in 2009,[11] recurrent hotspot mutations affecting the catalytic domains of IDH1 (Arg132) and isocitrate dehydrogenase 2 (IDH2 [Arg140 and Arg172]) occur in ~8% and ~12% of cases, respectively.[7,12,13] Prognostic impact of mutant *IDH2* is contentious (reviewed by Medeiros et al),[14] and isolated *IDH2*-R172 has been linked to a favorable outcome in 1 study.[6] Studies linking mutant *IDH1/2* to the neometabolite 2-hydroxyglutarate and arrested myeloid differentiation,[15-17] propagated the development of targeted inhibitors to mutant IDH2 (AG-221; enasidenib)[18,19] and IDH1 (AG-120; ivosidenib and BAY1436032).[20,21] The allosteric IDH2 inhibitor enasidenib effectively suppressed production of 2-hydroxyglutarate, releasing myeloid blasts from differentiation block.[19] As an orally administered drug in relapsed/refractory *IDH2* mutant AML, enasidenib (100 mg daily) produced complete remission (CR) and CR with incomplete hematologic recovery (CRi) in 26.6% of patients.[18] An additional 12% achieved either partial remission or morphologic leukemia-free state, giving an overall response rate of 38.5%.[18] Median duration of response was 5.6 months (8.8 months if CR achieved) and OS 9.3 months (19.7 months if CR achieved), in contrast to ~3 months with standard therapies.[22] Interestingly, despite reduced affinity for *IDH2*-R172, the overall response rate to enasidenib was 53.3%, compared with 35.4% for *IDH2*-R140.[23] Clinical responses to enasidenib were observed without reduction in the *IDH* mutant allele burden, reflecting conversion from predominantly undifferentiated, to differentiated clonal hematopoiesis.[19] The median time to best response was 3.7 months, with 82% of responses observed by cycle 7.[18] Responses have also been observed in patients with very small IDH clone sizes, raising the possibility that the drug may have additional paracrine effects on non-*IDH* mutant blasts.[19,24] Enasidenib has a distinct toxicity profile, the most important being IDH inhibitor–associated differentiation syndrome (IDH-DS), which occurs in 14% (grade 3+ severity in 7%).[18] IDH-DS may develop with or without concurrent hyperleukocytosis, and as late as 5 months after therapy initiation. Rapid myeloid proliferation manifesting as noninfectious leukocytosis has also been observed, requiring hydroxyurea administration and occasionally measures to mitigate tumor lysis syndrome.[18,25] The terminal half-life of enasidenib is ~5.7 days[18]; thus, drug cessation alone may not ameliorate IDH-DS. A high degree of clinical vigilance is needed and empirical use of steroids frequently warranted in cases of suspected IDH-DS, emphasized by a black box warning in the product information.

Future research questions and challenges

A particular challenge for patients and doctors will be attempts to maintain patients with persistent AML on IDH inhibitors for prolonged periods while waiting for a clinical response. Identifying predictive biomarkers of IDH inhibitor response will be highly valuable in justifying the decision to maintain patients on therapy. Preliminary results suggest a low likelihood of response when mutant *NRAS* is present.[19] Further work is needed to confirm the predictive value of this and other molecular markers. Enhancing clinical outcomes by combining IDH inhibitors with other drugs, such as hypomethylating agents (HMAs) or standard chemotherapy is already being evaluated. Registration studies in the first-line setting, however, will be challenged by the need for real-time mutation screening, the relatively low frequency of mutant *IDH1* and *IDH2* in the AML population and increasing commercial competition from non-IDH targeted drug options currently in development for AML that are also active in this patient subgroup.

CPX-351 (Jazz Pharmaceuticals, plc)

Clinical impact in AML

Prior preclinical studies demonstrating that cytarabine and daunorubicin delivered at molar ratios between 1:1 and 10:1 were synergistic, whereas lower ratios (1:5-1:10) were antagonistic, led to the development of CPX-351, which encapsulates cytarabine and daunorubicin at a fixed 5:1 molar ratio.[26] This ratiometric liposomal delivery system enhanced drug concentration in bone marrow and drug uptake into AML blasts, promoting superior antileukemic efficacy in vivo.[27,28] In human trials, the mean elimination half-life for CPX-351 was 25 hours for daunorubicin and 37 hours for cytarabine, substantially longer than pharmacokinetic exposures to free drugs. This could explain the longer time to neutrophil (36 vs 32 days) and platelet (37 vs 28 days) recovery with CPX-351, compared with conventional 7+3 chemotherapy.[29] Despite greater marrow suppression, a randomized phase 2 trial in patients 60 to 75 years did not show a significant increase in 30-day treatment-related mortality with CPX-351 (3.5%), compared with 7+3 (7.3%) as first-line therapy.[30] Although CPX-351 failed to increase OS in the overall study population, a preplanned analysis identified a superior CR rate and OS for CPX-351 in patients with secondary AML.[30] A pivotal phase 3 study therefore recruited 309 patients aged 60 to 75 years with a history of prior cytotoxic treatment, antecedent myelodysplastic syndrome (MDS) or chronic myelomonocytic leukemia, or AML with World Health Organization–defined MDS-related cytogenetic abnormalities. This study confirmed the higher CR rate (47.7% vs 33.3%, $P = .016$) and OS (median, 9.56 vs 5.95 months; hazard ratio, 0.69; $P = .005$) for CPX-351 over 7+3 (60 mg/m^2 daunorubicin),[31] prompting the FDA to approve CPX-351 for therapy-relatedAML (*t*-AML) and AML with myelodysplasia-related changes (AML-MRC).

Future research questions and challenges

The road to CPX-351 approval highlights the importance of studying subgroup responses to investigational therapies within clinical trials. The virtue of identifying a responder population within the context of a randomized phase 2 study followed by validation with a targeted phase 3 study was successfully demonstrated. Perplexingly, the rationale for benefit in *t*-AML and AML-MRC remains an open question, with one hypothesis that liposomal drug delivery may overcome Pgp-mediated drug resistance.

Table 1. Regulatory status of midostaurin, enasidenib, CPX-351, gemtuzumab ozogamicin and venetoclax

Drug and indication	Regulatory status
Midostaurin (Rydapt)	
Adult patients with newly diagnosed AML who are *FLT3*$^+$, as detected by an FDA-approved test, in combination with standard cytarabine and daunorubicin induction and cytarabine consolidation.	FDA approval 28 April 2017
In combination with standard daunorubicin and cytarabine induction and high-dose cytarabine consolidation chemotherapy, and for patients in complete response followed by Rydapt single-agent maintenance therapy, for adult patients with newly diagnosed AML who are *FLT3*$^+$.	European Medicines Agency approval 20 September 2017
CPX-351 (Vyxeos)	
Treatment of adults with *t*-AML or AML with AML-MRC.	FDA approval 3 August 2017
Enasidenib (Idhifa)	
Treatment of patients with relapsed or refractory AML with an *IDH2* mutation detected with an FDA-approved assay.	FDA approval 1 August 2017
Gemtuzumab ozogamicin (Mylotarg)	
Adults with newly diagnosed AML whose tumors express the CD33 antigen (CD33$^+$ AML). Patients aged 2 y and older with CD33$^+$ AML who have experienced a relapse or who have not responded to initial treatment (refractory).	FDA approval 1 September 2017
Venetoclax (Venclexta)	
Venetoclax in combination with HMAs for the treatment of patients with untreated (treatment-naïve) AML who are ineligible to receive standard induction therapy (high-dose chemotherapy).	FDA breakthrough designation 28 January 2016
Venetoclax in combination with LDAC for elderly patients with previously untreated AML who are ineligible for intensive chemotherapy.	FDA breakthrough designation 28 July 2017

The FDA label includes patients with *t*-AML and AML-MRC, which therefore extends the eligible population to include (1) younger patients and (2) AML with multilineage dysplasia (MLD), both of which were not specifically examined in the pivotal study. The morphologic definition of MLD is subject to interobserver bias,[32] and the revised World Health Organization 2016 definition of MLD now excludes patients with *NPM1*^{MUT} and biallelic *CEBPA*^{MUT}.[33] Del(9q) has also been removed from the list of MDS-related cytogenetic abnormalities.[33] Cytogenetic information may not be readily available when treatment needs to commence; therefore, it remains to be seen how these nuances will affect prescribing and drug provision practices. AML with preceding myeloproliferative neoplasm was not included in the definition of AML-MRC.[34] In first relapse, a randomized phase 2 study failed to show benefit for CPX-351 in AML compared with other salvage regimens, except in patients with a high European Prognostic Index risk score.[35] Therefore, future research will be important to validate the role of CPX-351 in the salvage setting, as well as in defined cytogenetic and molecular AML subgroups not adequately powered in the primary study to define the magnitude and consistency of benefit. Safety in combination with other novel therapies will also be an area of increasing future interest.

Gemtuzumab ozogamicin (Pfizer, Inc.)

Clinical impact in AML

Gemtuzumab ozogamicin (GO) is a humanized immunoglobulin G_4 antibody (hP67.6) directed against CD33 and conjugated via a hydrolysable linker to the DNA toxin calicheamicin. GO/CD33 complexes are internalized into lysosomes, releasing calicheamicin and promoting single and double-strand breaks and cellular death. GO initially received accelerated FDA approval in 2000 for the treatment of CD33$^+$ AML aged ≥60 years in first relapse, with the requirement that the company undertake a confirmatory postmarketing study.[36,37] A phase 3 study (S0106) was conducted by SWOG in untreated de novo AML, comparing daunorubicin/cytarabine (DA, 45 mg/m^2 daunorubicin) plus GO 6 mg/m^2 on day 4 with DA alone (60 mg/m^2 daunorubicin). The GO arm had higher induction mortality (5.5% vs 1.4%), without improving CR or relapse-free survival.[38] Based on these negative results, Pfizer was forced to withdraw GO from the market on 21 June 2010. Over the next decade, 4 additional investigator-led randomized studies in Europe (GOELAMS AML2006IR,[39] MRC AML15,[40] ALFA-0701,[41] and NCRI AML16[42]) were completed. ALFA-0701 randomized 278 patients with untreated de novo AML aged 50 to 70 years to DA (60 mg/m^2 daunorubicin) alone or in combination with a fractionated GO induction schedule (3 mg/m^2 on days 1, 4, and 7).[41] A single dose of GO (3 mg/m^2) was also given on day 1 of each of 2 consolidation cycles. Although CR with or without platelet recovery and early deaths were similar, patients in the GO arm had significantly improved median event-free survival (19.6 vs 11.9 months; $P = .00018$) and OS (34 vs 19.2 months; $P = .046$), with a subanalysis revealing benefit limited to patients with favorable and intermediate-risk karyotype.[41] A meta-analysis of 3325 patients from 5 randomized studies in untreated AML (aged 18-84) concluded that GO improved OS in patients with favorable and intermediate-risk karyotype when combined with standard induction chemotherapy.[43] Rates of veno-occlusive disease (VOD) and 30- and 60-day mortality were lower with 3 mg/m^2 vs 6 mg/m^2 GO.[44] The MyloFrance-1 study also gave 3 mg/m^2 on days 1, 4, and 7 to patients with AML at first relapse.[45]

Future research questions and challenges

The first approved dose of GO (9 mg/m^2 repeated after 2 weeks) was associated with grade 3-4 hyperbilirubinemia (23%) and elevated transaminases (17%), as well as prolonged severe myelosuppression.[46] At 9 mg/m^2, GO supersaturated CD33 binding sites, even after 2 weeks, resulting in internalization of GO/CD33 complexes and reduced availability of unbound target antigen.[47] Because only one-half of the antibody pool is actually conjugated to calicheamicin, increased binding site competition from unconjugated and inactive drug was another concern.[36] Delivering GO using a fractionated dosing schedule substantially improved the safety profile without compromising clinical outcomes.[41,45] A major concern for patients receiving GO is the risk of VOD, especially among patients who received SCT within 3 months. Revised dosing schedules appear to have lowered rates of VOD (<5% in ALFA-0701[48] and none were reported in the MyloFrance-1 study[45]). Experience from these studies, however, remains limited, and

clinicians should remain alert to the risk of VOD by avoiding concurrent hepatotoxic medications, minimizing SCT within 3 months of GO and continuing to monitor rates of VOD in the postmarketing period.

Although the US label for GO is broad, many unanswered questions remain. The limited GO activity in adverse karyotype AML requires further investigation, but may relate to reduced CD33 expression in adverse karyotype and lower rates of GO response in patients with increased expression of Pgp or MDR1.[45] In contrast, GO has pronounced efficacy in acute promyelocytic leukemia, which strongly expresses CD33.[49,50] For consolidation therapy in the ALFA-0701 study, GO was combined with DA,[41] which differs from higher dose cytarabine-based consolidation regimens used extensively in other parts of the world. Further research to explore the safety and additional efficacy of fractionated-dose GO in combination with higher dose cytarabine in consolidation are therefore warranted. Although GO has been included in the FDA label for patients with relapsed and refractory disease, the MyloFrance-1 study was a phase 2 uncontrolled trial,[45] making it difficult to determine the superiority of GO over other conventional salvage options.

Much remains to be learnt regarding the potential for GO to be combined with other novel drugs currently in development for AML. For example, *NPM1* mutant AML exhibits high levels of CD33 and BCL-2 expression, which may make it attractive to combine GO with venetoclax if this combination is found to be tolerable.[51,52] The frequent association between *NPM1* and *FLT3* mutations may also stimulate studies combining GO with FLT3 inhibitors. Therefore, despite almost 2 decades since its initial approval, research into the full use and potential of GO is just beginning.

Venetoclax (AbbVie Inc., Genentech Inc.)

Clinical impact in AML

Approximately one-third of elderly patients (>75 years) with AML are palliated without active therapy.[53] Increasing medical comorbidities, a higher frequency of poor risk gene mutations, and prior HMA failure are some of the diverse challenges limiting progress in elderly patients with AML.[54,55] Clinical responses (CR/CRi) to standard AML therapies used in the elderly, such as azacitidine (28%), decitabine (26%), or low-dose cytarabine (LDAC, 11% to 18%) are modest.[23,56,57] Increased expression of the pro-survival protein BCL-2 relative to the pro-apoptotic protein BAX is associated with reduced CR rates, earlier relapse, and inferior OS in patients receiving intensive chemotherapy for AML.[58] The BCL-2 inhibitor venetoclax was only modestly effective as monotherapy in relapsed/refractory AML (19% CR/CRi).[59] Recent phase 2 studies in elderly patients unfit for intensive chemotherapy have combined venetoclax with either HMAs or LDAC, producing CR/CRi rates of 62% to 68% and 12-month survival outcomes of 50% to 70%.[60,61] Responses were achieved rapidly (median, 1 month) and early mortality was low (2%). Registration studies are currently under way to validate the benefit of venetoclax in combination with standard therapies in elderly patients with AML (NCT02993523 and NCT03069352).

Future research questions and challenges

The robust activity of venetoclax in combination with low-intensity therapies in elderly patients with AML provides a competitive alternative to other inhibitors, such as FLT3 and IDH inhibitors, without the need for preemptive mutation screening. This may increase the difficulty of patient recruitment into registration studies of elderly patients with AML targeting a specific subgroup in the frontline setting. The response rates and 12-month OS for venetoclax/HMA or LDAC also compare favorably with results achieved using intensive chemotherapy in the elderly AML population.[62] Therefore, the distinction between "fit" and "unfit" older patients when selecting therapy may lose relevance if a highly active treatment with relatively low toxicity becomes available. In younger adults, and in relapsed/refractory AML, future studies will likely determine if venetoclax can be safely combined with more intensive chemotherapy approaches. The best outcomes for venetoclax/HMA or LDAC appear to be in patients with *NPM1* mutant AML, which notably express high levels of BCL-2.[52] Future research should also seek to understand mechanisms of clonal resistance, and the potential for BH3-mimetics targeting other pro-survival proteins, such as MCL1 to be combined with BCL-2 targeting in AML.[63,64]

Conclusions

Despite a relatively unchanging therapeutic landscape for several decades in AML, the stage has now been revitalized by the debut of 4 new FDA approvals within the space of just 6 months in 2017 for patients with *FLT3* mutant AML, *IDH2* mutant AML, CD33 positive AML, *t*-AML, and AML-MRC (Table 1). The majority of the AML population may now have treatment outcomes augmented by the addition of a novel drug in the clinic. Additionally, venetoclax is also making solid strides toward a possible drug registration in elderly patients with AML. Although cytarabine and daunorubicin will continue to play an important role in AML, patients and physicians will now have the help of several new recruits in their fight against this lethal blood cancer.

Authorship

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References

1. Sekeres MA, Steensma DP. Boulevard of broken dreams: drug approval for older adults with acute myeloid leukemia. *J Clin Oncol*. 2012;30(33):4061-4063.
2. Fabbro D, Buchdunger E, Wood J, et al. Inhibitors of protein kinases: CGP 41251, a protein kinase inhibitor with potential as an anticancer agent. *Pharmacol Ther*. 1999;82(2-3):293-301.
3. Weisberg E, Boulton C, Kelly LM, et al. Inhibition of mutant FLT3 receptors in leukemia cells by the small molecule tyrosine kinase inhibitor PKC412. *Cancer Cell*. 2002;1(5):433-443.
4. Stone RM, Mandrekar SJ, Sanford BL, et al. Midostaurin plus chemotherapy for acute myeloid leukemia with a FLT3 mutation. *N Engl J Med*. 2017;377(5):454-464.
5. Levis M. FLT3 mutations in acute myeloid leukemia: what is the best approach in 2013?

Hematology Am Soc Hematol Educ Program. 2013;2013:220-226.

6. Papaemmanuil E, Gerstung M, Bullinger L, et al. Genomic classification and prognosis in acute myeloid leukemia. *N Engl J Med*. 2016;374(23): 2209-2221.
7. Metzeler KH, Herold T, Rothenberg-Thurley M, et al.; AMLCG Study Group. Spectrum and prognostic relevance of driver gene mutations in acute myeloid leukemia. *Blood*. 2016;128(5): 686-698.
8. Gale RE, Green C, Allen C, et al.; Medical Research Council Adult Leukaemia Working Party. The impact of FLT3 internal tandem duplication mutant level, number, size, and interaction with NPM1 mutations in a large cohort of young adult patients with acute myeloid leukemia. *Blood*. 2008;111(5):2776-2784.
9. Knapper S, Russell N, Gilkes A, et al. A randomized assessment of adding the kinase inhibitor lestaurtinib to first-line chemotherapy for FLT3-mutated AML. *Blood*. 2017;129(9): 1143-1154.
10. Röllig C, Serve H, Hüttmann A, et al.; Study Alliance Leukaemia. Addition of sorafenib versus placebo to standard therapy in patients aged 60 years or younger with newly diagnosed acute myeloid leukaemia (SORAML): a multicentre, phase 2, randomised controlled trial. *Lancet Oncol*. 2015;16(16):1691-1699.
11. Mardis ER, Ding L, Dooling DJ, et al. Recurring mutations found by sequencing an acute myeloid leukemia genome. *N Engl J Med*. 2009;361(11): 1058-1066.
12. Ley TJ, Miller C, Ding L, et al.; Cancer Genome Atlas Research Network. Genomic and epigenomic landscapes of adult de novo acute myeloid leukemia. *N Engl J Med*. 2013;368(22): 2059-2074.
13. Papaemmanuil E, Gerstung M, Malcovati L, et al.; Chronic Myeloid Disorders Working Group of the International Cancer Genome Consortium. Clinical and biological implications of driver mutations in myelodysplastic syndromes. *Blood*. 2013;122(22):3616-3627, quiz 3699.
14. Medeiros BC, Fathi AT, DiNardo CD, Pollyea DA, Chan SM, Swords R. Isocitrate dehydrogenase mutations in myeloid malignancies. *Leukemia*. 2017;31(2):272-281.
15. Dang L, White DW, Gross S, et al. Cancer-associated IDH1 mutations produce 2-hydroxyglutarate. *Nature*. 2009;462(7274): 739-744.
16. Gross S, Cairns RA, Minden MD, et al. Cancer-associated metabolite 2-hydroxyglutarate accumulates in acute myelogenous leukemia with isocitrate dehydrogenase 1 and 2 mutations. *J Exp Med*. 2010;207(2):339-344.
17. Ward PS, Patel J, Wise DR, et al. The common feature of leukemia-associated IDH1 and IDH2 mutations is a neomorphic enzyme activity converting alpha-ketoglutarate to 2-hydroxyglutarate. *Cancer Cell*. 2010;17(3): 225-234.
18. Stein EM, DiNardo CD, Pollyea DA, et al. Enasidenib in mutant IDH2 relapsed or refractory acute myeloid leukemia. *Blood*. 2017;130(6): 722-731.
19. Amatangelo MD, Quek L, Shih A, et al. Enasidenib induces acute myeloid leukemia cell differentiation to promote clinical response. *Blood*. 2017;130(6):732-741.
20. Chaturvedi A, Herbst L, Pusch S, et al. Pan-mutant-IDH1 inhibitor BAY1436032 is highly effective against human IDH1 mutant acute myeloid leukemia in vivo. *Leukemia*. 2017;31(10): 2020-2028.
21. DiNardo C, de Botton S, Pollyea DA, et al. Molecular profiling and relationship with clinical response in patients with IDH1 mutation-positive hematologic malignancies receiving AG-120, a first-in-class potent inhibitor of mutant IDH1, in addition to data from the completed dose escalation portion of the phase 1 study. *Blood*. 2015;126(23):1306.
22. Roboz GJ, Rosenblat T, Arellano M, et al. International randomized phase III study of elacytarabine versus investigator choice in patients with relapsed/refractory acute myeloid leukemia. *J Clin Oncol*. 2014;32(18):1919-1926.
23. Dombret H, Seymour JF, Butrym A, et al. International phase 3 study of azacitidine vs conventional care regimens in older patients with newly diagnosed AML with >30% blasts. *Blood*. 2015;126(3):291-299.
24. Chaturvedi A, Araujo Cruz MM, Jyotsana N, et al. Enantiomer-specific and paracrine leukemogenicity of mutant IDH metabolite 2-hydroxyglutarate. *Leukemia*. 2016;30(8): 1708-1715.
25. Birendra KC, DiNardo CD. Evidence for clinical differentiation and differentiation syndrome in patients with acute myeloid leukemia and IDH1 mutations treated with the targeted mutant IDH1 inhibitor, AG-120. *Clin Lymphoma Myeloma Leuk*. 2016;16(8):460-465.
26. Mayer LD, Harasym TO, Tardi PG, et al. Ratiometric dosing of anticancer drug combinations: controlling drug ratios after systemic administration regulates therapeutic activity in tumor-bearing mice. *Mol Cancer Ther*. 2006;5(7):1854-1863.
27. Kim HP, Gerhard B, Harasym TO, Mayer LD, Hogge DE. Liposomal encapsulation of a synergistic molar ratio of cytarabine and daunorubicin enhances selective toxicity for acute myeloid leukemia progenitors as compared to analogous normal hematopoietic cells. *Exp Hematol*. 2011;39(7):741-750.
28. Lim WS, Tardi PG, Xie X, et al. Schedule- and dose-dependency of CPX-351, a synergistic fixed ratio cytarabine:daunorubicin formulation, in consolidation treatment against human leukemia xenografts. *Leuk Lymphoma*. 2010;51(8): 1536-1542.
29. Feldman EJ, Lancet JE, Kolitz JE, et al. First-in-man study of CPX-351: a liposomal carrier containing cytarabine and daunorubicin in a fixed 5:1 molar ratio for the treatment of relapsed and refractory acute myeloid leukemia. *J Clin Oncol*. 2011;29(8):979-985.
30. Lancet JE, Cortes JE, Hogge DE, et al. Phase 2 trial of CPX-351, a fixed 5:1 molar ratio of cytarabine/daunorubicin, vs cytarabine/daunorubicin in older adults with untreated AML. *Blood*. 2014;123(21):3239-3246.
31. Lancet JE, Uy GL, Cortes JE, et al. Final results of a phase III randomized trial of CPX-351 versus 7+3 in older patients with newly diagnosed high risk (secondary) AML. *J Clin Oncol*. 2016;34 (15_suppl):7000.
32. Weinberg OK, Pozdnyakova O, Campigotto F, et al. Reproducibility and prognostic significance of morphologic dysplasia in de novo acute myeloid leukemia. *Mod Pathol*. 2015;28(7): 965-976.
33. Arber DA, Orazi A, Hasserjian R, et al. The 2016 revision to the World Health Organization classification of myeloid neoplasms and acute leukemia. *Blood*. 2016;127(20):2391-2405.
34. Swerdlow SH, Campo E, Harris NL, et al. WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues. Lyon: IARC Press; 2008.
35. Cortes JE, Goldberg SL, Feldman EJ, et al. Phase II, multicenter, randomized trial of CPX-351 (cytarabine:daunorubicin) liposome injection versus intensive salvage therapy in adults with first relapse AML. *Cancer*. 2015;121(2):234-242.
36. Bross PF, Beitz J, Chen G, et al. Approval summary: gemtuzumab ozogamicin in relapsed acute myeloid leukemia. *Clin Cancer Res*. 2001; 7(6):1490-1496.
37. Larson RA, Boogaerts M, Estey E, et al.; Mylotarg Study Group. Antibody-targeted chemotherapy of older patients with acute myeloid leukemia in first relapse using Mylotarg (gemtuzumab ozogamicin). *Leukemia*. 2002;16(9):1627-1636.
38. Petersdorf SH, Kopecky KJ, Slovak M, et al. A phase 3 study of gemtuzumab ozogamicin during induction and postconsolidation therapy in younger patients with acute myeloid leukemia. *Blood*. 2013;121(24):4854-4860.
39. Delaunay J, Recher C, Pigneux A, et al. Addition of gemtuzumab ozogamycin to chemotherapy improves event-free survival but not overall survival of AML patients with intermediate cytogenetics not eligible for allogeneic transplantation. Results of the GOELAMS AML 2006 IR Study. *Blood*. 2011;118(21):79.
40. Burnett AK, Hills RK, Milligan D, et al. Identification of patients with acute myeloblastic leukemia who benefit from the addition of gemtuzumab ozogamicin: results of the MRC AML15 trial. *J Clin Oncol*. 2011;29(4):369-377.
41. Castaigne S, Pautas C, Terré C, et al. Acute Leukemia French Association. Effect of gemtuzumab ozogamicin on survival of adult patients with de-novo acute myeloid leukaemia (ALFA-0701): a randomised, open-label, phase 3 study. *Lancet*. 2012;379(9825):1508-1516.
42. Burnett AK, Russell NH, Hills RK, et al. Addition of gemtuzumab ozogamicin to induction chemotherapy improves survival in older patients with acute myeloid leukemia. *J Clin Oncol*. 2012; 30(32):3924-3931.
43. Hills RK, Castaigne S, Appelbaum FR, et al. Addition of gemtuzumab ozogamicin to induction chemotherapy in adult patients with acute myeloid leukaemia: a meta-analysis of individual patient data from randomised controlled trials. *Lancet Oncol*. 2014;15(9):986-996.
44. Burnett A, Cavenagh J, Russell N, et al. UK NCRI AML Study Group. Defining the dose of gemtuzumab ozogamicin in combination with induction chemotherapy in acute myeloid leukemia: a comparison of 3 mg/m2 with 6 mg/m2 in the NCRI AML17 Trial. *Haematologica*. 2016; 101(6):724-731.
45. Taksin AL, Legrand O, Raffoux E, et al. High efficacy and safety profile of fractionated doses of Mylotarg as induction therapy in patients with relapsed acute myeloblastic leukemia: a prospective study of the alfa group. *Leukemia*. 2007;21(1):66-71.
46. Magwood-Golston JS, Kessler S, Bennett CL. Evaluation of gemtuzumab ozogamycin associated sinusoidal obstructive syndrome: Findings from an academic pharmacovigilance program review and a pharmaceutical sponsored registry. *Leuk Res*. 2016;44:61-64.
47. van Der Velden VH, te Marvelde JG, Hoogeveen PG, et al. Targeting of the CD33-calicheamicin immunoconjugate Mylotarg (CMA-676) in acute myeloid leukemia: in vivo and in vitro saturation and internalization by leukemic and normal myeloid cells. *Blood*. 2001;97(10):3197-3204.
48. FDA Oncologic Drugs Advisory Committee Briefing Document - Mylotarg. (gemtuzumab ozogamicin). July 11, 2017. https://www.fda.gov/downloads/AdvisoryCommittees/CommitteesMeetingMaterials/Drugs/OncologicDrugsAdvisoryCommittee/UCM566013.pdf. Accessed 8 October 2017.
49. Lancet JE, Moseley A, Komrokji RS, et al. ATRA, arsenic trioxide (ATO), and gemtuzumab ozogamicin (GO) is safe and highly effective in patients with previously untreated high-risk acute promyelocytic leukemia (APL): final results of the

SWOG/Alliance/ECOG S0535 Trial. *Blood*. 2016; 128(22):896.

50. Abaza Y, Kantarjian H, Garcia-Manero G, et al. Long-term outcome of acute promyelocytic leukemia treated with all-trans-retinoic acid, arsenic trioxide, and gemtuzumab. *Blood*. 2017; 129(10):1275-1283.
51. De Propris MS, Raponi S, Diverio D, et al. High CD33 expression levels in acute myeloid leukemia cells carrying the nucleophosmin (NPM1) mutation. *Haematologica*. 2011;96(10):1548-1551.
52. Chyla B, Popovic R, Potluri J, et al. Correlative biomarkers of response to venetoclax in combination with chemotherapy or hypomethylating agents in elderly untreated patients with acute myeloid leukemia. *Blood*. 2016;128(22):1709.
53. Medeiros BC, Satram-Hoang S, Hurst D, Hoang KQ, Momin F, Reyes C. Big data analysis of treatment patterns and outcomes among elderly acute myeloid leukemia patients in the United States. *Ann Hematol*. 2015;94(7):1127-1138.
54. Tsai CH, Hou HA, Tang JL, et al. Genetic alterations and their clinical implications in older patients with acute myeloid leukemia. *Leukemia*. 2016;30(7):1485-1492.
55. Creutzig U, Zimmermann M, Reinhardt D, et al. Changes in cytogenetics and molecular genetics in acute myeloid leukemia from childhood to adult age groups. *Cancer*. 2016;122(24):3821-3830.
56. Kantarjian HM, Thomas XG, Dmoszynska A, et al. Multicenter, randomized, open-label, phase III trial of decitabine versus patient choice, with physician advice, of either supportive care or low-dose cytarabine for the treatment of older patients with newly diagnosed acute myeloid leukemia. *J Clin Oncol*. 2012;30(21):2670-2677.
57. Burnett AK, Milligan D, Prentice AG, et al. A comparison of low-dose cytarabine and hydroxyurea with or without all-trans retinoic acid for acute myeloid leukemia and high-risk myelodysplastic syndrome in patients not considered fit for intensive treatment. *Cancer*. 2007;109(6):1114-1124.
58. Del Poeta G, Venditti A, Del Principe MI, et al. Amount of spontaneous apoptosis detected by Bax/Bcl-2 ratio predicts outcome in acute myeloid leukemia (AML). *Blood*. 2003;101(6):2125-2131.
59. Konopleva M, Pollyea DA, Potluri J, et al. Efficacy and biological correlates of response in a phase II study of venetoclax monotherapy in patients with acute myelogenous leukemia. *Cancer Discov*. 2016;6(10):1106-1117.
60. Pratz K, Pollyea DA, Jonas BA, et al. Safety and efficacy of venetoclax (Ven) in combination with decitabine or azacitidine in treatment-naive, elderly patients (≥65 years) with acute myeloid leukemia (AML). *Haematologica*. 2017;102(s2): S472.
61. Wei AH, Strickland SA, Roboz GJ, et al. Venetoclax plus low-dose cytarabine in treatment-naive acute myeloid leukemia patients aged ≥65 years and unfit for standard induction therapy. *Haematologica*. 2017;102(s2):S473.
62. Löwenberg B, Ossenkoppele GJ, van Putten W, et al.; Swiss Group for Clinical Cancer Research (SAKK) Collaborative Group. High-dose daunorubicin in older patients with acute myeloid leukemia [published correction appears in *N Engl J Med*. 2010;362(12):1155]. *N Engl J Med*. 2009; 361(13):1235-1248.
63. Kotschy A, Szlavik Z, Murray J, et al. The MCL1 inhibitor S63845 is tolerable and effective in diverse cancer models. *Nature*. 2016;538(7626): 477-482.
64. Moujalled D, Pomilio G, Ghiurau C, et al. A dual BH3-mimetic approach targeting both BCL-2 and MCL1 is highly efficacious and well-tolerated in acute myeloid leukemia. *Haematologica*. 2017; 102(s2):175.