OncoTargets and Therapy

Dovepress
open access to scientific and medical research

Open Access Full Text Article

REVIEW

MDM2 antagonists as a novel treatment option for acute myeloid leukemia: perspectives on the therapeutic potential of idasanutlin (RG7388)

This article was published in the following Dove Medical Press journal:
OncoTargets and Therapy

Arushi Khurana
Danielle A Shafer

Department of Internal Medicine, Division of Hematology, Oncology & Palliative Care, Virginia Commonwealth University, Richmond, VA 23298, USA

Abstract: Acute myeloid leukemia (AML) is a clonal heterogenous malignancy of the myeloid cells with a poor prognosis lending itself to novel treatment strategies. TP53 is a critical tumor suppressor and plays an essential role in leukemogenesis. Although TP53 is relatively unusual in de novo AML, inactivation of wild-type p53 (WT-p53) is a common event. Murine double minute 2 (MDM2) is a key negative regulator of p53 and its expression; inhibition of MDM2 is postulated to reactivate WT-p53 and its tumor suppressor functions. Nutlins were the first small molecule inhibitors that bind to MDM2 and target its interaction with p53. RG7388 (idasanutlin), a second-generation nutlin, was developed to improve upon the potency and toxicity profile of earlier nutlins. Preliminary data from early phase trials and ongoing studies suggest clinical response with RG7388 (idasanutlin) both in monotherapy and combination strategies in AML. We herein briefly discuss currently approved therapies in AML and review the clinical data for RG7388 (idasanutlin) and MDM2 inhibition as novel treatment strategies in AML. We further describe efficacy and toxicity profile data from completed and ongoing trials of RG7388 (idasanutlin) and other MDM2-p53 inhibitors in development. Many targeted therapies have been approved recently in AML, with a focus on the older and unfit population for intensive induction therapy and in relapsed/refractory disease. The "nutlins", including RG7388 (idasanutlin), merit continued investigation in such settings.

Keywords: AML, myeloid leukemia, nutlins, MDM2, idasanutlin, RG7388, p53 inhibitor

Introduction

Acute myeloid leukemia (AML) is the most common acute leukemia in adults. It is much more prevalent in the elderly with a median age at diagnosis of 68 years.[1] Surveillance, Epidemiology and End Results Program (SEER) 2018 analysis estimates approximately 19,520 new cases of AML (1.1% of all cancers) and 10,670 deaths (1.8% of all cancers) in the US.[2] AML is a clonal disorder with heterogeneous molecular, cytogenetic, biological and clinical features.[3] Significant progress has been made in the last 4 decades understanding the genomic landscape of AML.[4,5] This has translated into remarkable growth in drug development and approval. After a very long drought in AML therapeutics, we have witnessed recent FDA approval for multiple new drugs. These new agents target molecular drivers of AML such as Fms-like tyrosine kinase 3 (*FLT3*), epigenetic regulators such as isocitrate dehydrogenase (*IDH1/2*) and monoclonal antibodies against surface markers on leukemia cells such as CD33.[6–10] Many other similar agents are currently in clinical trials.[11–13] One such target in AML is exploring the inhibition of the interaction between tumor suppressor gene *p53* and murine double minute 2 (*MDM2*). We herein briefly review

Correspondence: Danielle A Shafer
Department of Internal Medicine, Division of Hematology, Oncology & Palliative Care, Virginia Commonwealth University, 1300 East Marshall St, PO Box 980292, Richmond, VA 23298, USA
Email danielle.shafer@vcuhealth.org

submit your manuscript | www.dovepress.com
Dovepress
http://dx.doi.org/10.2147/OTT.S172315

the current AML treatment paradigm and explore in detail the pharmacology, mechanism of action and efficacy of the MDM2 inhibitor RG7388 (idasanutlin).

Despite recent advances, the 5-year survival rate for AML remains at 25%–30%. Standard of care for younger and fit patients eligible to undergo aggressive induction therapy remains the "7+3" regimen, 7-day continuous cytarabine infusion and 3 daily doses of an anthracycline agent, followed by consolidative strategies with high-dose cytarabine or allogeneic stem cell transplantation (Allo-SCT).[14,15] Older and unfit patients not suitable for aggressive treatments have dismal outcomes with median overall survival (OS) less than 1 year.[16,17] There are limited options for older and unfit patients including best supportive care or hypomethylating agents. The FDA has recently granted accelerated approval to venetoclax in combination with azacitidine, decitabine or low-dose cytarabine in newly diagnosed AML patients older than 75 years or with comorbidities precluding the use of intensive induction chemotherapy.[18] Although this novel approach is potentially paradigm shifting, the majority of patients will relapse. There is a great need for more effective therapies.

For patients with relapsed/refractory (R/R) disease, there are often fewer options. Given the inadequacy of current treatments, enrollment in clinical trials is always recommended whenever possible. Selected patients are able to undergo more intensive chemotherapy with a goal of transplant if eligible. However, for many patients, cytotoxic chemotherapy is not appropriate or has a low likelihood of success. In a Phase III trial evaluating elacytarabine, novel cytarabine, versus the investigator's choice of 1 of the 7 commonly used AML salvage regimens, the median survival was approximately 3 months in both arms.[19] That sobering statistic is an impetus for new drug development. Many of the recent FDA approvals in AML are targeted agents in the R/R setting. FLT3 is a tyrosine kinase receptor involved in normal hematopoiesis and cell proliferation. Activating *FLT3* mutations have been reported in up to 30% of the AML patients including 20% with internal tandem duplication (*FLT3*-ITD) mutations and 5%–10% with point mutations in the activating loop of the tyrosine kinase domain (*FLT3*-TKD). Gilteritinib, an oral FLT3 inhibitor, was recently approved for adults with *FLT3*-positive R/R AML.[20] In the ADMIRAL study that led to FDA approval, the rate of complete remission (CR) or CR with partial hematologic recovery (CRh) was 21%. Other highly selective FLT3 inhibitors such as quizartinib and crenolanib are in clinical trials both in the upfront and R/R settings. Disordered epigenetic regulation also has therapeutic potential. Both IDH1 (ivosidenib) and IDH2 (enasidenib) inhibitors are now approved in the R/R setting, in which they have clearly shown a survival benefit and improvement in the quality of life with transfusion independence and less febrile neutropenia requiring hospitalizations. Response rates for both agents approximate 30%–40%.[7,8] Multiple other targets are under investigation at this time including p53-MDM2 inhibitors-nutlins.

p53 and MDM2

TP53 mutations occur in about 7%–8% of the de novo AML cases, whereas inactivation of wild-type p53 (WT-p53) occurs in almost all AML subsets.[3] p53 transcription factor plays a crucial role in tumor suppression by various mechanisms including apoptosis, DNA repair, maintenance of normal stem cell pool and regulating self-renewal, thereby preventing leukemogenesis in AML.[21–24] *TP53*, the gene encoding p53, is known to be mutated in up to 50% of all human cancers.[25] Kemp et al[26] demonstrated that a lack of p53 led to the increased predisposition of various tumors in murine models. Multiple mechanisms have been described for inactivation of WT-p53. The best studied of those is through MDM2 overexpression and p14 (ARF) inactivation. The ARF-MDM2/4-p53 axis is involved in most AML cases, with ARF inactivation or MDM2 overexpression leading to non-functional p53.[27]

MDM2 serves as a negative regulator of p53. Wu et al[28] first described the mutual regulation between p53 and MDM2 through a feedback loop. Activation of p53 through any stimuli or DNA damage increases transcription of MDM2 mRNA and protein which in turn binds to p53 and directly inhibits its function via 3 primary mechanisms. First, the E3 ligase activity of MDM2 directly ubiquitinates p53 leading to its degradation through proteasomes. Second, MDM2 and p53 binding blocks p53 from binding to its target DNA causing lack of transcription. Third, MDM2 increases the export of p53 from the cell nucleus that makes it inaccessible to the target DNA for transcription.[29–31] In vivo studies have also confirmed this interaction and the oncogenic potential of MDM2 overexpression, with increased expression leading to increased tumor formation.[32–35] Oliner et al[36] first described MDM2 amplification in one-third of the human sarcoma samples. Momand et al[37] showed similar amplification in multiple other tumor types. Besides gene amplification, another mechanism for MDM2 overexpression was described by Bond et al[38,39] as single nucleotide polymorphism (SNP) in the MDM2 promoter region. *MDM2* gene amplification

remains the most essential implicated mechanism in MDM2 overexpression.[40] More importantly, *MDM2* gene amplification and *TP53* mutations are mutually exclusive in human cancers.[36,41,42] Of note, preclinical data suggest that about two-thirds of AML cell lines and patient-derived samples are sensitive to MDM2 inhibition, and as expected, the *TP53* mutated cells show resistance.[43,44]

Based on the MDM2-p53 interaction, inhibition of MDM2 was postulated to reactivate WT-p53 and its tumor suppressor functions, making it a potential therapeutic target. Momand et al[45] mapped the MDM2-p53 protein–protein interaction to the first 120 amino-terminal amino acid residues of MDM2 and the first 30 amino-terminal residues of p53. In 2004, Vassilev et al[46] first discovered "nutlins", the small molecule inhibitors that bind to MDM2 and target its interaction with p53. In vivo studies of nutlin-3 showed extensive reduction in tumor mass in the MDM2-amplified xenograft osteosarcoma model.[47] Pishas et al showed significant apoptotic responses on immunohistochemical analysis of nutlin-3 treated human sarcoma tissue samples.[48] These preclinical data led to the development of several potent and selective non-peptide small-molecule MDM2 inhibitors. The first MDM2 inhibitor to be advanced into human clinical trials was RG7112 (Hoffmann La Roche RO5045337).[49] RG7112 is several times more potent and selective for WT-p53 than nutlin-3; furthermore, it demonstrated efficacy in both in vitro and in vivo studies and had a dose-dependent effect on tumor regression. In several Phase I trials with both solid and hematological malignancies, RG7112 showed evidence of on-target activity resulting in p53 activation. After treatment with RG7112, there was an increased expression of downstream pro-apoptotic proteins.[50–52] In AML, RG7112 was studied both as monotherapy and in combination with low-dose cytarabine.[53] Some patients even achieved CR and were subsequently transplanted. Dose-limiting toxicities (DLTs) noted in the combination trials were rash, thrombocytopenia, and diarrhea (>20% of the adverse events [AEs] were gastrointestinal [GI] or infectious). The hematological toxicity with this drug was prolonged, as MDM2 plays a crucial role in hematopoiesis.[54] The higher dose to attain satisfactory p53 activation caused significant toxicities (cytopenias, diarrhea, sepsis, and deaths), and so the need for a more potent and less toxic agent was identified.

RG7388 (idasanutlin)

RG7388 (idasanutlin) is a second-generation MDM2 inhibitor. It was developed to improve upon the stereochemical and conformational properties of the spirooxindole series by the introduction of the cyanopyrrolidine core, which was thought to be more flexible.[55,56] It was found to be more potent, more selective, and had a better pharmacokinetic (PK) profile as compared to RG7112.[56] It also showed dose-dependent p53 stabilization, apoptosis, and cell cycle arrest. In SJSA1 osteosarcoma xenografts in nude mice, RG7388 (idasanutlin) was more effective than RG7112 at much lower doses.[56,57] RG7388 (idasanutlin) has also been studied in both solid and hematological malignancies. Here, we limit our discussion for its use and implications in AML.

In a multicenter Phase 1/1b study, RG7388 (idasanutlin) was evaluated in AML patients as monotherapy (daily for 5 days every 28 days) and in combination with cytarabine (ara-C 1 gm/m^2 IV ×5 days every 28 days) in a dose escalation study.[58] An extension cohort was initiated in both groups at the recommended Phase II dose (RP2D). The monotherapy extension arm included patients older than 70 years and patients older than 60 years with comorbidities. The combination extension arm included R/R patients with not more than 2 prior regimens. Patients with antecedent hematologic disorders or transplant were not eligible for the combination arm. The RP2D for RG7388 (idasanutlin) as monotherapy or in combination was determined to be 1,200 mg/day (600 mg bid for 5 days every 28 days). Only 1 DLT of prolonged myelosuppression was reported. The most common AEs were diarrhea and infection. Diarrhea was reported by greater than 85% of the patients and did not appear to be dose-dependent.

In the monotherapy arm, 9 patients were treated at the RP2D. The median age was 75 years (range 66–83); 8 of the 9 patients were reported to have an antecedent hematologic disorder. There were 3 responses including 1 complete response with incomplete recovery (CRi), 1 partial response (PR) and 1 hematological improvement (HI). There were 3 patient deaths in the first 30 days. Enrollment onto the monotherapy expansion phase was discontinued for prolonged myelosuppression which increased the risk of infection and early death. In the reported PK data, the $t_{1/2}$ was noted to be ~1 day and was irrespective of age, concomitant cytarabine, or azoles.[58]

Seventy-six patients were treated on the combination arm in the dose escalation (n=23) and dose expansion cohorts (n=21) with an additional 32 patients in a bridging cohort.[59] The bridging arm was added to characterize the safety and PK profile of a spray-dried powder formulation of RG7388 (idasanutlin). The CR rate was 25% (n=19); the composite CR rate (cCR, CR + CRp + CRi) was 29%. The cCR patients were followed until relapse or 1 year from

the start of therapy; the median duration of response was ~6.4 months (1.1–11.9 months). Five patients remained in CR at 1 year follow-up. Patients with cCR also had minimal residual disease (MRD) assessment with multiparametric flow cytometry on day 28 and for those experiencing cCR at subsequent assessments.[60] Median progression-free survival (PFS) for cCR patients was 315 days compared to 43.5 days in the non-responders. When MRD thresholds of $<1\%$ and $>1\%$ were applied, there was a statistically significant association with median PFS. Patients with MRD $<1\%$ had a median PFS of 367 days versus 84 days (p-value=0.001) in the MRD $>1\%$ group. The MRD findings provide further support of its utility in providing prognostic information in AML. To identify possible biomarkers of response, MDM2 protein expression was evaluated by intracellular flow cytometry on peripheral blood leukemic blasts and stem cells. Higher MDM2 expression in both leukemic blasts and stem cells was associated with CR; *TP53* mutational status alone was not. These results raise the potential of MDM2 expression in leukemic cells to serve as a predictive biomarker for response. Interestingly, responses were identified in patients with *TP53* mutations including 1 CR, suggesting *TP53* mutation as an inadequate companion diagnostic for AML patients. Zhong et al[61] previously reported similar results in in vitro AML cell lines but were based on whole blood samples instead of expression on leukemic cells only. Given the efficacy in Phase I/Ib AML study, RG7388 (idasanutlin) is currently undergoing evaluation in a Phase III trial in combination with cytarabine versus cytarabine alone for R/R AML patients (NCT02545283).

RG7388 (idasanutlin) is also being extensively explored in combination with other apoptotic agents such as the BCL2 inhibitor venetoclax. In preclinical studies in p53 wild-type AML tumor models, the combination of RG7388 (idasanutlin) and venetoclax was synergistic.[62] Similar results were seen in WT-p53 AML cell lines treated with the MDM2 inhibitor SAR405838 and bcl-2 inhibitor ABT-263 (navitoclax).[63] Cell viability and annexin binding assays showed not only synergism but also potent efficacy. Interestingly, RG7388 (idasanutlin) induced G1 arrest and caused nuclear fragmentation in the G1 phase of the second cycle while the bcl-2 inhibitor caused apoptosis in G1 compartments. Cells that were transiently missed from apoptosis by RG7388 (idasanutlin) were hit by the bcl-2 inhibitor. Further studies with the combination suggest that each agent can reciprocally overcome the apoptotic resistance to either agent alone.[64,65] The RG7388 (idasanutlin) and venetoclax combination is being evaluated in Phase I/Ib trial for patients 60 years and older with R/R AML who are not candidates for cytotoxic therapy (NCT02670044). A recent Phase I study of the pegylated intravenous prodrug of idasanutlin (RO6839921) suggested a similar PK profile to RG7388 (idasanutlin). Antileukemic activity was noted to be around 42% in the overall population (26 patients).[66]

Toxicity profile

Overall, RG7388 (idasanutlin) appears to be well tolerated. The most common AEs in the reported studies were limited to diarrhea, nausea and vomiting and myelosuppression causing febrile neutropenia and thrombocytopenia.[58,67,68] These are thought to be the result of on-target effects of the drug on the normal cells.[69] This toxicity profile is similar among all the MDM2 inhibitors. Long-term and off-target toxicities of these agents are currently unknown and will become evident with time.

Other MDM2 inhibitors

Other small molecule MDM2 inhibitors are currently undergoing investigation in AML. MK-8242 (SCH-900242) is an orally bioavailable and potent small molecule inhibitor of the MDM2-p53 interaction. In a Phase I study with 24 evaluable patients in R/R AML, MK-8242 was evaluated in 2 different schedules.[70] Two DLTs were identified, bone marrow failure and prolonged cytopenias; no MTD was identified. The most common AEs of any grade were GI and hematologic which was similar to other MDM2 inhibitors. Efficacy was modest with 1 PR, 1 CRi and 1 MLFS. AMG232, a potent oral MDM2 inhibitor, has also been evaluated in adults with R/R AML. In a Phase Ib study, AMG232 with or without trametinib (MEK inhibitor) was administered to 35 patients.[71] There was some early evidence of activity but no reported CRs in the monotherapy arm; there was, however, 1 CR in the combination arm. An MTD was identified; toxicities were again similar to those seen with other MDM2 inhibitors. AMG-232 is currently being evaluated in combination with decitabine in newly diagnosed AML patients with WT-p53. DS-3032b (milademetan) is another oral p53-MDM2 inhibitor currently under evaluation in patients with AML and other hematological malignancies. In a Phase I dose escalation study in 38 patients with R/R AML and high-risk myelodysplatic syndrome (MDS), the single-agent MTD was determined to be 160 mg daily 21/28 days.[72] The toxicity profile was similar to other MDM2 inhibitors with GI and hematological toxicity; 5 subjects had DLTs including grade 3 hypokalemia, grade 3 diarrhea, grade 3 nausea and vomiting, grade 2 renal insufficiency

Table 1 Selected list of the ongoing clinical trials with nutlins/MDM2 inhibitors in AML and MDS

Study ID	Disease	Drugs	Phase	Estimated completion date
NCT02670044	R/R AML patients >60 years, not candidates for cytotoxic therapy	Venetoclax + idasanutlin or venetoclax + cobimetinib	Phase Ib/II	January 15, 2020
NCT02545283	R/R AML, 18 and older	Idasanutlin + cytarabine vs cytarabine alone	Phase III	May 26, 2021
NCT03041688	R/R AML, newly diagnosed AML	AMG-232 and decitabine	Phase Ib	October 31, 2019
NCT03634228	R/R AML	DS-3032b and cytarabine	Phase I/II	May 1, 2020
NCT02319369	R/R AML, newly diagnosed AML, high-risk MDS	DS-3032b ± azacitidine	Phase I	July 2021
NCT03552029	R/R AML with FLT3 mutation	DS-3032b + quizartinib	Phase I	October 15, 2021
NCT02909972	R/R AML and high-risk MDS with WT-TP53	ALRN-6924 ± cytarabine	Phase I/Ib	April 2018 (Still listed as recruiting)

Abbreviations: AML, Acute myeloid leukemia; MDS, myelodysplatic syndrome; R/R, relapsed/refractory.

and grade 3 anorexia/fatigue. There were CRs in 2 patients with AML and 1 patient with MDS achieved a marrow CR. It is notable that each patient developed a TP53 mutation while on treatment. Further evaluation with azacitidine is ongoing (NCT02319369). DS-3032b is also being evaluated in combination with cytarabine and quizartinib in AML patients (NCT03552029).

More recently, dual inhibition of MDM2/MDMX as a therapeutic strategy is undergoing clinical evaluation. MDMX similar to MDM2 also represses TP53 transcriptional activity. It is hypothesized that targeting TP53 interactions with MDM2 and MDMx will have a more significant impact than MDM2 inhibition alone. ALRN-6924 is a novel "stapled peptide", which has been structurally stabilized in an α-helical configuration, to mimic the inhibitor binding region of TP53. By mimicking this region, ALRN-6924 can bind the 2 most important endogenous inhibitors of p53, MDM2 and MDMX. In Phase I/Ib study, ALRN-6924 was evaluated alone and in combination with cytarabine in AML patients.[73] In the preliminary results for 13 monotherapy and 19 combination arm patients, there were no DLTs and MTD was not identified. The most common AEs were notably GI toxicity and thrombocytopenia. In the 27 efficacy evaluable patients, there were 2 marrow CRs in 4 MDS patients; 1 AML patient had a 50% reduction in marrow blasts. Ongoing studies of MDM2 small molecule inhibitors and MDM2/MDMX stapled peptide drugs are listed in Table 1.

Challenges and future directions

MDM2 inhibition is a promising therapeutic target in AML. Long-term data are needed to further elucidate the potential toxicities, mechanisms of resistance and efficacy. Challenges that have been identified are related to its on-target effects to normal cells especially GI and hematological toxicities and the emergence of resistance. Data from the clinical studies to date suggest late hematological toxicity due to on-target effects on the bone marrow. Identifying the optimal dose for each MDM2 inhibitor during monotherapy as well as in combination especially with agents such as venetoclax which is known to cause myelosuppression is necessary.

Development of resistance to MDM2 inhibitors seems to occur due to the emergence of p53 mutations through a selection of p53-mutated clones or the emergence of p53 mutation.[74] Other mechanisms described are through point mutations in the p53-binding pockets of MDM2 and high MDMX (positive regulator of MDM2) levels.[75–78] In vitro, the AML cell lines that are treated with MDM2 inhibitors and develop resistance still retain sensitivity to BCL-2 inhibitors.[63] Hopefully, the combination with bcl-2 inhibitors will overcome this issue, although clinically such resistance is yet to be described. This again demonstrates the efficacy of combination strategy over monotherapy by targeting different apoptosis mechanisms simultaneously or sequentially.

The clinical relevance and applicability of targeting apoptotic mechanisms in AML have been confirmed with the recent approval of venetoclax (bcl-2 inhibitor) in combination with azacitidine, decitabine or low-dose cytarabine for newly diagnosed AML patients aged 75 or older who are not candidates for intensive induction. In Phase I/II studies, the combination showed overall response rates of 60%–70%.[18] Data are awaited on most of the combination studies for idasanutlin as the trials are ongoing. The challenge remains how best to incorporate it with currently approved therapies and which population to target for maximal benefits. To date, it seems to be well tolerated as monotherapy in older individuals not candidates for intensive chemotherapy, but efficacy results have been modest. It could potentially be incorporated in combination with induction therapy for younger/fit adults to improve response

especially in those with poor risk or chemotherapy refractory disease.[67]

Precision medicine is the future of oncology. AML patients are beginning to benefit from this approach with the recent approval of FLT3, IDH1/2 inhibitors. However, not all patients harbor a targetable mutation; for those patients, targeting the apoptosis pathway may prove to be an effective alternative. Further studies are required to further understand the mechanisms of resistance, toxicity and biomarkers for the prediction of response and prognosis.

Disclosure

The authors report no conflicts of interest in this work.

References

1. DeSantis CE, Lin CC, Mariotto AB, et al. Cancer treatment and survivorship statistics. *CA Cancer J Clin*. 2014;64:252–271. doi:10.3322/caac.v64.4
2. Acute myeloid leukemia – cancer stat facts. Available from: https://seer.cancer.gov/statfacts/html/amyl.html. Accessed October 2, 2018.
3. Marcucci G, Haferlach T, Döhner H. Molecular genetics of adult acute myeloid leukemia: prognostic and therapeutic implications. *J Clin Oncol*. 2011;29:475–486. doi:10.1200/JCO.2010.30.2554
4. Papaemmanuil E, Gerstung M, Bullinger L, et al. Genomic classification and prognosis in acute myeloid leukemia. *N Engl J Med*. 2016;374:2209–2221. doi:10.1056/NEJMoa1516192
5. Cancer Genome Atlas Research Network. Genomic and epigenomic landscapes of adult de novo acute myeloid leukemia. *N Engl J Med*. 2013;368:2059–2074. doi:10.1056/NEJMoa1301689
6. Stone RM, Mandrekar SJ, Sanford BL, et al. Midostaurin plus chemotherapy for acute myeloid leukemia with a FLT3 mutation. *N Engl J Med*. 2017;377:454–464. doi:10.1056/NEJMoa1614359
7. DiNardo CD, Stein EM, de Botton S, et al. Durable remissions with ivosidenib in IDH1-mutated relapsed or refractory AML. *N Engl J Med*. 2018;378:2386–2398. doi:10.1056/NEJMc1711583
8. Stein EM, DiNardo CD, Pollyea DA, et al. Enasidenib in mutant *IDH2* relapsed or refractory acute myeloid leukemia. *Blood*. 2017;130:722–731. doi:10.1182/blood-2017-04-779405
9. 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;15:986–996. doi:10.1016/S1470-2045(13)70510-2
10. Gamis AS, Alonzo TA, Meshinchi S, et al. Gemtuzumab ozogamicin in children and adolescents with de novo acute myeloid leukemia improves event-free survival by reducing relapse risk: results from the randomized phase III Children's Oncology Group trial AAML0531. *J Clin Oncol*. 2014;32:3021–3032. doi:10.1200/JCO.2013.54.6911
11. Saygin C, Carraway HE. Emerging therapies for acute myeloid leukemia. *J Hematol Amp Oncol*. 2017;10(1):93. doi:10.1186/s13045-017-0463-6.
12. Shafer D, Grant S. Update on rational targeted therapy in AML. *Blood Rev*. 2016;30:275–283. doi:10.1016/j.blre.2016.02.001
13. Yang X, Wang J. Precision therapy for acute myeloid leukemia. *J Hematol Amp Oncol*. 2018;11(1):5. doi:10.1186/s13045-017-0543-7.
14. Döhner H, Estey E, Grimwade D, et al. Diagnosis and management of AML in adults: 2017 ELN recommendations from an international expert panel. *Blood*. 2017;129:424–447. doi:10.1182/blood-2016-08-733196
15. Yates JW, Wallace HJ, Ellison RR, Holland JF. Cytosine arabinoside (NSC-63878) and daunorubicin (NSC-83142) therapy in acute nonlymphocytic leukemia. *Cancer Chemother Rep*. 1973;57:485–488.
16. Podoltsev NA, Stahl M, Zeidan AM, Gore SD. Selecting initial treatment of acute myeloid leukaemia in older adults. *Blood Rev*. 2017;31:43–62. doi:10.1016/j.blre.2016.09.005
17. Pleyer L, Burgstaller S, Stauder R, et al. Azacitidine front-line in 339 patients with myelodysplastic syndromes and acute myeloid leukaemia: comparison of French-American-British and World Health Organization classifications. *J Hematol Oncol*. 2016;9:39. doi:10.1186/s13045-016-0263-4
18. DiNardo CD, Pollyea DA, Jonas BA, et al. Updated safety and efficacy of venetoclax with decitabine or azacitidine in treatment-naive, elderly patients with acute myeloid leukemia. *Blood*. 2017;130:2628.
19. 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;32:1919–1926. doi:10.1200/JCO.2013.52.8562
20. Perl AE, Altman JK, Cortes J, et al. Selective inhibition of FLT3 by gilteritinib in relapsed or refractory acute myeloid leukaemia: a multicentre, first-in-human, open-label, phase 1–2 study. *Lancet Oncol*. 2017;18:1061–1075. doi:10.1016/S1470-2045(17)30416-3
21. Toledo F, Wahl GM. Regulating the p53 pathway: in vitro hypotheses, in vivo veritas. *Nat Rev Cancer*. 2006;6:909–923. doi:10.1038/nrc2012
22. Vousden KH, Lu X. Live or let die: the cell's response to p53. *Nat Rev Cancer*. 2002;2:594–604. doi:10.1038/nrc864
23. Wade M, Li Y-C, Wahl GM. MDM2, MDMX and p53 in oncogenesis and cancer therapy. *Nat Rev Cancer*. 2013;13:83–96. doi:10.1038/nrc3430
24. Stiewe T. The p53 family in differentiation and tumorigenesis. *Nat Rev Cancer*. 2007;7:165–167. doi:10.1038/nrc2072
25. Feki A, Irminger-Finger I. Mutational spectrum of p53 mutations in primary breast and ovarian tumors. *Crit Rev Oncol Hematol*. 2004;52:103–116. doi:10.1016/j.critrevonc.2004.07.002
26. Kemp CJ, Donehower LA, Bradley A, Balmain A. Reduction of p53 gene dosage does not increase initiation or promotion but enhances malignant progression of chemically induced skin tumors. *Cell*. 1993;74:813–822. doi:10.1016/0092-8674(93)90461-X
27. Prokocimer M, Molchadsky A, Rotter V. Dysfunctional diversity of p53 proteins in adult acute myeloid leukemia: projections on diagnostic workup and therapy. *Blood*. 2017;130:699–712. doi:10.1182/blood-2017-02-763086
28. Wu X, Bayle JH, Olson D, Levine AJ. The p53-mdm-2 autoregulatory feedback loop. *Genes Dev*. 1993;7:1126–1132. doi:10.1101/gad.7.7a.1126
29. Juven-Gershon T, Oren M. Mdm2: the ups and downs. *Mol Med*. 1999;5:71–83.
30. Freedman DA, Wu L, Levine AJ. Functions of the MDM2 oncoprotein. *Cell Mol Life Sci C*. 1999;55:96–107. doi:10.1007/s000180050273
31. Oren M. Regulation of the p53 tumor suppressor protein. *J Biol Chem*. 1999;274:36031–36034. doi:10.1074/jbc.274.51.36031
32. de Oca Luna RM, Wagner DS, Lozano G. Rescue of early embryonic lethality in mdm2-deficient mice by deletion of p53. *Nature*. 1995;378:203–206. doi:10.1038/378203a0
33. Jones SN, Roe AE, Donehower LA, Bradley A. Rescue of embryonic lethality in Mdm2-deficient mice by absence of p53. *Nature*. 1995;378:206–208. doi:10.1038/378206a0
34. Lundgren K, de Oca Luna RM, McNeill YB, et al. Targeted expression of MDM2 uncouples S phase from mitosis and inhibits mammary gland development independent of p53. *Genes Dev*. 1997;11:714–725.
35. Jones SN, Hancock AR, Vogel H, Donehower LA, Bradley A. Overexpression of Mdm2 in mice reveals a p53-independent role for Mdm2 in tumorigenesis. *Proc Natl Acad Sci U S A*. 1998;95:15608–15612. doi:10.1073/pnas.95.26.15608
36. Oliner JD, Kinzler KW, Meltzer PS, George DL, Vogelstein B. Amplification of a gene encoding a p53-associated protein in human sarcomas. *Nature*. 1992;358:80–83. doi:10.1038/358080a0
37. Momand J, Jung D, Wilczynski S, Niland J. The MDM2 gene amplification database. *Nucleic Acids Res*. 1998;26:3453–3459. doi:10.1093/nar/26.15.3453

38. Bond GL, Hu W, Bond EE, et al. A single nucleotide polymorphism in the MDM2 promoter attenuates the p53 tumor suppressor pathway and accelerates tumor formation in humans. *Cell*. 2004;119:591–602. doi:10.1016/j.cell.2004.11.022
39. Bond GL, Hu W, Levine AJ. MDM2 is a central node in the p53 pathway: 12 years and counting. *Curr Cancer Drug Targets*. 2005;5:3–8. doi:10.2174/1568009053332627
40. Oliner JD, Saiki AY, Caenepeel S. The role of MDM2 amplification and overexpression in tumorigenesis. *Cold Spring Harb Perspect Med*. 2016;6:a026336. doi:10.1101/cshperspect.a026336
41. Wasylishen AR, Lozano G. Attenuating the p53 pathway in human cancers: many means to the same end. *Cold Spring Harb Perspect Med*. 2016;6:a026211. doi:10.1101/cshperspect.a026211
42. Shvarts A, Steegenga WT, Riteco N, et al. MDMX: a novel p53-binding protein with some functional properties of MDM2. *Embo J*. 1996;15:5349–5357. doi:10.1002/embj.1996.15.issue-19
43. Long J, Parkin B, Ouillette P, et al. Multiple distinct molecular mechanisms influence sensitivity and resistance to MDM2 inhibitors in adult acute myelogenous leukemia. *Blood*. 2010;116:71–80. doi:10.1182/blood-2010-01-261628
44. Weisberg E, Halilovic E, Cooke VG, et al. Inhibition of wild-type p53-Expressing AML by the novel small molecule HDM2 inhibitor CGM097. *Mol Cancer Ther*. 2015;14:2249–2259. doi:10.1158/1535-7163.MCT-15-0429
45. Momand J, Wu HH, Dasgupta G. MDM2–master regulator of the p53 tumor suppressor protein. *Gene*. 2000;242:15–29. doi:10.1016/S0378-1119(99)00487-4
46. Vassilev LT, Vu BT, Graves B, et al. In vivo activation of the p53 pathway by small-molecule antagonists of MDM2. *Science*. 2004;303:844–848. doi:10.1126/science.1092472
47. Tovar C, Rosinski J, Filipovic Z, et al. Small-molecule MDM2 antagonists reveal aberrant p53 signaling in cancer: implications for therapy. *Proc Natl Acad Sci*. 2006;103:1888–1893. doi:10.1073/pnas.0507493103
48. Pishas KI, Al-Ejeh F, Zinonos I, et al. Nutlin-3a Is a Potential Therapeutic for Ewing Sarcoma. Clin Cancer Res. 2011;17(3):494–504. DOI: 10.1158/1078-0432.CCR-10-1587
49. Vu B, Wovkulich P, Pizzolato G, et al. Discovery of RG7112: a small-molecule MDM2 inhibitor in clinical development. *ACS Med Chem Lett*. 2013;4:466–469. doi:10.1021/ml4003138
50. Andreeff M, Kelly KR, Yee K, et al. Results of the phase I trial of RG7112, a small-molecule MDM2 antagonist in leukemia. *Clin Cancer Res*. 2016;22:868–876. doi:10.1158/1078-0432.CCR-16-0190
51. Kurzrock R, Blay J-Y, Nguyen BB, et al. A phase I study of MDM2 antagonist RG7112 in patients (pts) with relapsed/refractory solid tumors. *J Clin Oncol*. 2012;30:e13600.
52. Ray-Coquard I, Blay JY, Italiano A, et al. Effect of the MDM2 antagonist RG7112 on the P53 pathway in patients with MDM2-amplified, well-differentiated or dedifferentiated liposarcoma: an exploratory proof-of-mechanism study. *Lancet Oncol*. 2012;13:1133–1140. doi:10.1016/S1470-2045(12)70140-7
53. Martinelli G, Assouline S, Kasner M, et al. Phase 1b study of the MDM2 antagonist RG7112 in combination with 2 doses/schedules of cytarabine. *Blood*. 2013;122:498. doi:10.1182/blood-2012-12-471029
54. Mahfoudhi E, Lordier L, Marty C, et al. P53 activation inhibits all types of hematopoietic progenitors and all stages of megakaryopoiesis. *Oncotarget*. 2016;7:31980–31992. doi:10.18632/oncotarget.v7i22
55. Zhang B, Golding BT, Hardcastle IR. Small-molecule MDM2-p53 inhibitors: recent advances. *Future Med Chem*. 2015;7:631–645. doi:10.4155/fmc.15.13
56. Ding Q, Zhang Z, Liu J-J, et al. Discovery of RG7388, a potent and selective p53–MDM2 inhibitor in clinical development. *J Med Chem*. 2013;56:5979–5983. doi:10.1021/jm400487c
57. Tovar C, Graves B, Packman K, et al. MDM2 small-molecule antagonist RG7112 activates p53 signaling and regresses human tumors in preclinical cancer models. *Cancer Res*. 2013;73:2587–2597. doi:10.1158/0008-5472.CAN-12-2807
58. Yee K, Martinelli G, Vey N, et al. Phase 1/1b study of RG7388, a potent MDM2 antagonist, in Acute Myelogenous Leukemia (AML) patients (Pts). *Blood*. 2014;124:116.
59. Martinelli G, Pappayannidis C, Yee K, et al. Phase 1b results of idasanutlin + cytarabine (ARA-C) in acute…. EHA Learning Center. Pappayannidis C. [udated June 11, 2016]. 135260. Available from: https://learningcenter.ehaweb.org/eha/2016/21st/135260/cristina.pappayannidis.phase.1b.results.of.idasanutlin.2B.cytarabine.28ara-c29.in.html. Accessed October 20, 2018.
60. Lanza B, Martinelli G, Yee KW, et al. Minimal Residual Disease (MRD) assessment by multiparametric flow cytometry is prognostic for progression-free survival in phase 1/1b relapsed/refractory Acute Myeloid Leukemia (AML) patients treated with idasanutlin MDM2 antagonist. *Blood*. 2016;128:2843. doi:10.1182/blood-2016-06-724161
61. Zhong H, Chen G, Jukofsky L, et al. MDM2 antagonist clinical response association with a gene expression signature in acute myeloid leukaemia. *Br J Haematol*. 2015;171:432–435. doi:10.1111/bjh.2015.171.issue-3
62. Dangl M, Chien Y, Lehmann C, Friess T. Abstract 5505: synergistic anticancer activity of clinical stage, non-genotoxic apoptosis inducing agents RG7388 (MDM2 antagonist) and ABT-199 (GDC-0199, BCL2 inhibitor) in p53 wild-type AML tumor models. *Cancer Res*. 2014;74:5505. doi:10.1158/0008-5472.CAN-13-3514
63. Hoffman-Luca CG, Ziazadeh D, McEachern D, et al. Elucidation of acquired resistance to Bcl-2 and MDM2 inhibitors in acute leukemia in vitro and in vivo. *Clin Cancer Res*. 2015;21:2558–2568. doi:10.1158/1078-0432.CCR-14-2506
64. Pan R, Kojima K, Zheng Z, et al. Activation of p53 by novel MDM2 antagonist RG7388 overcomes AML inherent and acquired resistance to Bcl-2 inhibitor ABT-199 (GDC-0199). *Blood*. 2014;124:2162.
65. Pan R, Ruvolo V, Mu H, et al. BCL-2 inhibition by ABT-199 (venetoclax/GDC-0199) and p53 activation by RG7388 (idasanutlin) reciprocally overcome leukemia apoptosis resistance to either strategy alone: efficacy and mechanisms. *Blood*. 2015;126:673.
66. Yee K, Uy G, Assouline S, et al. Abstract A082: a phase I study of the MDM2 antagonist RO6839921, a pegylated intravenous prodrug of idasanutlin, in patients with AML. *Clin Trials*. 2018;17:A082–A082.
67. Cassier PA, Castets M, Belhabri A, Vey N. Targeting apoptosis in acute myeloid leukaemia. *Br J Cancer*. 2017;117:1089–1098. doi:10.1038/bjc.2017.281
68. Siu LL, Italiano A, Miller WH, et al. Phase 1 dose escalation, food effect, and biomarker study of RG7388, a more potent second-generation MDM2 antagonist, in patients (pts) with solid tumors. *J Clin Oncol*. 2014;32:2535. doi:10.1200/JCO.2013.54.6911
69. Kojima K, Konopleva M, Samudio IJ, et al. MDM2 antagonists induce p53-dependent apoptosis in AML: implications for leukemia therapy. *Blood*. 2005;106:3150–3159. doi:10.1182/blood-2004-04-1622
70. Ravandi F, Gojo I, Patnaik MM, et al. A phase I trial of the human double minute 2 inhibitor (MK-8242) in patients with refractory/recurrent acute myelogenous leukemia (AML). *Leuk Res*. 2016;48:92–100. doi:10.1016/j.leukres.2016.07.004
71. Erba HP, Becker PS, Shami PJ, et al. Dose escalation results of a phase 1b study of the MDM2 inhibitor AMG 232 with or without trametinib in patients (Pts) with relapsed/refractory (r/r) acute myeloid leukemia (AML). *J Clin Oncol*. 2017;35:7027. doi:10.1200/JCO.2017.35.15_suppl.7027
72. DiNardo CD, Rosenthal J, Andreeff M, et al. Phase 1 dose escalation study of MDM2 inhibitor DS-3032b in patients with hematological malignancies – preliminary results. *Blood*. 2016;128:593. doi:10.1182/blood-2016-06-724161
73. Sallman DA, Borate U, Cull EH, et al. Phase 1/1b study of the stapled peptide ALRN-6924, a dual inhibitor of MDMX and MDM2, as monotherapy or in combination with cytarabine for the treatment of relapsed/refractory AML and advanced MDS with TP53 wild-type. *Blood*. 2018;132:4066.
74. Michaelis M, Rothweiler F, Barth S, et al. Adaptation of cancer cells from different entities to the MDM2 inhibitor nutlin-3 results in the emergence of p53-mutated multi-drug-resistant cancer cells. *Cell Death Dis*. 2011;2:e243. doi:10.1038/cddis.2011.82

75. Jones RJ, Bjorklund CC, Baladandayuthapani V, Kuhn DJ, Orlowski RZ. Drug resistance to inhibitors of the human double minute-2 E3 ligase is mediated by point mutations of p53, but can be overcome with the p53 targeting agent RITA. *Mol Cancer Ther*. 2012;11:2243–2253. doi:10.1158/1535-7163.MCT-11-0824-T
76. Wei SJ, Joseph T, Sim AYL, et al. In vitro selection of mutant HDM2 resistant to nutlin inhibition. *PLoS One*. 2013;8:e62564. doi:10.1371/journal.pone.0062564
77. Wade M, Wong ET, Tang M, Vassilev LT, Wahl GM. Hdmx modulates the outcome of p53 activation in human tumor cells. *J Bio Chem*. 2006;281:33036–33044. doi:10.1074/jbc.M605405200
78. Garcia D, Warr MR, Martins CP, Brown Swigart L, Passegue E, Evan GI. Validation of MdmX as a therapeutic target for reactivating p53 in tumors. *Genes Dev*. 2011;25:1746–1757. doi:10.1101/gad.16722111