Targeted Inhibition of Mutant IDH2 in Leukemia Cells Induces Cellular Differentiation

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A number of human cancers harbor somatic point mutations in the genes encoding isocitrate dehydrogenases 1 and 2 (IDH1 and IDH2). These mutations alter residues in the enzyme active sites and confer a gain-of-function in cancer cells, resulting in the accumulation and secretion of the oncometabolite (R)-2-hydroxyglutarate (2HG). We developed a small molecule, AGI-6780, that potently and selectively inhibits the tumor-associated mutant IDH2/R140Q. A crystal structure of AGI-6780 complexed with IDH2/R140Q revealed that the inhibitor binds in an allosteric manner at the dimer interface. The results of steady-state enzymology analysis were consistent with allostery and slow-tight binding by AGI-6780. Treatment with AGI-6780 induced differentiation of TF-1 erythroleukemia and primary human acute myelogenous leukemia cells in vitro. These data provide proof-of-concept that inhibitors targeting mutant IDH2/R140Q could have potential applications as a differentiation therapy for cancer.

Some point mutations affecting one of three active-site arginine residues of isocitrate dehydrogenase (IDH1/R132, IDH2/R140, and IDH2/R172) define distinct subsets of low-grade glioma and secondary glioblastoma, chondrosarcoma, cholangiocarcinomas, and acute myelogenous leukemia (AML) (1). IDH1 is a metabolic enzyme that interconverts isocitrate and α-ketoglutarate (αKG), but cancer-associated point mutations in IDH1 and IDH2 confer a neomorphic activity that allows reduction of αKG to the oncometabolite (R)-2-hydroxyglutarate (2HG) (2). High concentrations of 2HG have been shown to inhibit αKG-dependent dioxygenases, including histone and DNA demethylases, which play a key role in regulating the epigenetic state of cells (3–5). Patients harboring IDH mutations display a CpG island methylator phenotype (CIMP), and overexpression of IDH mutant enzymes can induce histone and DNA hypermethylation, as well as block cellular differentiation (6–8). In addition, mice engineered to express IDH1/R132H in hematopoietic tissue display an increased number of early hematopoietic progenitors, as well as splenomegaly, anemia, hypermethylated histones, and altered DNA methylation patterns similar to those found in AML patients harboring IDH1/2 mutations (9). Together, these data suggest that cancer-associated IDH mutations may induce a block in cellular differentiation to promote tumorigenesis.

To elucidate the relationship between mutant enzyme activity, 2HG levels, and oncogenic state, we developed a small molecule that selectively inhibits the IDH2/R140Q mutant expressed in:

Fig. 1. Chemical structure of AGI-6780 (A) and co-complex of AGI-6780 and NADP+ with IDH2/R140Q (B). Individual chains are colored in light blue and green. A single molecule of AGI-6780 binds at the dimer interface. NADP+ molecules are shown with carbons in black, AGI-6780 is shown with carbons in magenta, and Glu116 is shown with carbon chains matching their respective A and B chains. (C) Detailed view of the environment around AGI-6780. The loops (residues 151 to 168) that fold over the open end of the binding pocket are depicted in ribbon form. Hydrogen bonds are depicted as black dashes. (D) A top-down view of the binding pocket showing the 151–168 loops folded over the top. The green helices and the loop immediately above are from one monomer and the blue helices with the loop immediately above are from the opposite monomer. The 151–168 loops are shown in yellow and fold over the open end of the binding pocket. The Glu116 residues that hydrogen bond to AGI-6780 are shown in stick form with H-bonds depicted as dashed lines. The kinetically inert calcium ions that occupy the site of the catalytic divalent metal ions are shown as gray spheres. The metal ion is ligated by Asp114 and Asp118 from one monomer and Asp118 from the opposite monomer, with the rest of the coordination sphere being completed by water molecules when in the open conformation. The equivalent Asp114/Asp118-containing helix in cytosolic IDH1 was observed to unfold in the open conformation. Abbreviations for the amino acid residues are as follows: D, Asp; Lys; L, Leu; Q, Gln; V, Val; W, Trp; and Y, Tyr.

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The binding mode of AGI-6780 is consistent with the observation that similar drug concentrations can inhibit mutant IDH2 comparably in enzymatic- and cellular-based 2HG assays.

To confirm that the AGI-6780 binding mode was similar to that observed with the initial active molecules, we determined a high-resolution crystal structure of homodimeric IDH2/R140Q complexed with NADP+, Ca2+, and AGI-6780 (Fig. 1B and table S1). As expected, a single inhibitor molecule was observed at the central IDH2 dimer interface created by four helices, residues 290 to 299 and 310 to 322 from each of the A and B chains. The AGI-6780 binding site is opposite to the side of the helices that project aspartate residues necessary for the chelation of a catalytic divalent ion such as Mg2+. The highly symmetric AGI-6780 binding pocket extends up into the protein interface and is closed by loops composed of residues 152 to 167 (Fig. 1, C and D). These loops have a slightly higher temperature factor than the surrounding residues (Fig. 1D), suggesting increased mobility. They also fold over the pocket and would need to move to allow access into or out of the allosteric site, providing an explanation for the ratio of catalytic activity. They also fold over the pocket and would need to move to allow access into or out of the allosteric site, providing an explanation for the catalytic activity.

Table 1. Biochemical and cellular potency for 2HG inhibition by AGI-6780.

<table>
<thead>
<tr>
<th>AGI-6780 biochemical properties, IC50 for α-KG reduction</th>
<th>Incubation time (hours)</th>
<th>IC50 (nM)</th>
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<tr>
<td>Enzymes assayed</td>
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<tr>
<td>IDH2-R140Q</td>
<td>1</td>
<td>170 ± 47</td>
</tr>
<tr>
<td>IDH2-R140Q/WT</td>
<td>16</td>
<td>23 ± 1.7</td>
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<tr>
<td>IDH2-R140Q/WT</td>
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<td>120 ± 42</td>
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<tr>
<td>IDH1-R132H</td>
<td>16</td>
<td>4.0 ± 1.2</td>
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<tr>
<td>IDH1-R132H</td>
<td>16</td>
<td>11000 ± 84</td>
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<tr>
<td>Enzymes assayed</td>
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<tr>
<td>IDH2-WT</td>
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<tr>
<td>IDH2-WT</td>
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<td>IDH1-WT</td>
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<th>AGI-6780 cellular properties, IC50 for 2-HG formation</th>
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<tr>
<td>Cell line</td>
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<tr>
<td>TF-1 (IDH2-R140Q)</td>
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<td>18 ± 0.51</td>
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<tr>
<td>U87 (IDH2-R140Q)</td>
<td>48</td>
<td>11 ± 2.6</td>
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<tr>
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<td>48</td>
<td>&gt;100000</td>
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<tr>
<th>AGI-6780 dehydrogenase selectivity panel</th>
<th>Incubation time (hours)</th>
<th>IC50 (nM)</th>
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</thead>
<tbody>
<tr>
<td>Enzymes assayed</td>
<td></td>
<td></td>
</tr>
<tr>
<td>LDHA (pyruvate to lactate)</td>
<td>1</td>
<td>&gt;100000</td>
</tr>
<tr>
<td>LDHA (lactate to pyruvate)</td>
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</tr>
<tr>
<td>3PGDH</td>
<td>1</td>
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<tr>
<td>GDH</td>
<td>1</td>
<td>&gt;100000</td>
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<tr>
<td>G6PDH</td>
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The time-dependent inhibition against homodimeric or heterodimeric IDH2/WT or IDH2/R140Q mutant by AGI-6780 was investigated after either 1 or 16 hours of preincubation. AGI-6780 was tested in both human glioblastoma U87 and TF-1 cells expressing IDH2-R140Q, as well as against IDH1/R132H. Selectivity against other dehydrogenases, including the closely related IDH1-WT or R132H mutant enzymes (Table 1). The tight-binding, allosteric mode of inhibition of AGI-6780 is consistent with the observation that similar drug concentrations can inhibit mutant IDH2 comparably in enzymatic- and cellular-based 2HG assays.

From van der Wals contacts between the binding site on the protein and the more hydrophobic regions of AGI-6780. An open conformation for the protein was observed whereby the large cofactor binding domain (residues 41 to 158 and 324 to 456) is separated from the small catalytic domain (residues 159 to 218 and 224 to 323) and is nearly identical (<1 Å root mean square deviation) to that observed for the yeast mitochondrial IDH determined in the absence of substrate molecules (fig. S2A) (11). This open conformation is analogous to that observed for cytosolic IDH1 (12) with the major exception that the catalytic metal-coordinating helices remain intact in IDH2 (Fig. 1D) whereas they are unfolded in the IDH1 open conformation. Because IDH1 and IDH2 have different allosteric sites, highly selective targeting of the mitochondrial IDH2 isoform was achieved with AGI-6780.

Our approach to designing inhibitors against IDH2 with slow-tight binding and noncompetitive inhibitory properties arose from the hypothesis that antitumor efficacy would require 2HG to be fully suppressed to background levels under conditions where cellular cofactor concentrations may far exceed IDH2 mutant intrinsic affinity for NADPH (Km, NADPH = 200 nM). The discovery of heterocyclic urea sulfonamides that bind in a unique allosteric pocket of IDH2/R140Q confirmed this hypothesis. The binding of AGI-6780 at the dimer interface suggests that its role as an allosteric inhibitor arises from its ability to hold the IDH2/R140Q dimer in an open conformation that is incompatible with catalysis. The allosteric site where AGI-6780 is bound does not directly involve Gln140. Nonetheless, AGI-6780 exhibits excellent potency against the neomorphic activity of both the mutant homodimer and IDH2/WT:IDH2/R140Q heterodimer enzymes, although exhibiting less potency against the normal oxidative decarboxylation activity of IDH2/WT (Table 1).

The inhibitory activity against both mutant and wild-type subunits could offer an advantage in the context of heterozygous IDH mutations because studies of the IDH1/WT: IDH1/R132H heterodimer suggested that the wild-type and mutant subunits contribute to the production of 2HG either in concert or independently, depending on whether the carbon source is derived from isocitrate or from αKG, respectively (13). Genetic knockdown of IDH1/WT alone decreased 2HG production (14), which further underscores the contribution of the wild-type IDH allele to 2HG production in some cancer cells. Overall, we believe that both the allosteric nature observed by enzyme kinetics and crystallography and the slow-tight binding characteristics measured with enzyme kinetics are essential features of AGI-6780 for optimal suppression of 2HG production arising from the IDH2 mutation in cancer cells.

To study the biological effect of AGI-6780, we clonedly selected mutant IDH2/R140Q protein...
in granulocyte-macrophage colony-stimulating factor (GM-CSF)–dependent human TF-1 erythroleukemia cells (15). These cells (TF1/R140Q) produce large amounts of 2HG similar to that observed in human tumors (5 to 30 mM), which correlates with mutant IDH2 expression and growth in the absence of GM-CSF (Fig. 2A and fig. S3) (16, 17). Expression of IDH2/R140Q also induced a morphological change in TF-1 cells: They became more spindle in shape and attached to the tissue culture plate, two features often found in undifferentiated mesenchymal or “stem” cells (Fig. 2B) (18). Similarly, immunoblot analysis showed an increase in vimentin expression (Fig. 2C), a marker found in human hematopoietic stem cells (HSCs), but not in cells differentiating along the erythrocyte lineage (19). Together, these data suggest that IDH2/R140Q expression can induce a more immature phenotype. To test this possibility, we analyzed cell surface expression of CD34 and CD38 by flow cytometry. CD34 expression is found on human hematopoietic stem and progenitor cells, and high CD38 expression is only seen at later stages of hematopoietic differentiation. Although CD34 and CD38 are both found on the cell surface of TF-1/pLVX vector cells, CD38 expression was decreased in the IDH2/R140Q cells, suggesting a more primitive state of differentiation (Fig. 2D). These data demonstrate that IDH2/R140Q can either shift TF-1 cells toward an earlier stage and/or block hematopoietic cell differentiation and is similar to the results found with the IDH1/R132H mutation (20).

Because IDH mutations are reported to induce a block in hematopoietic cell differentiation (6, 9), we tested whether IDH2/R140Q expression could block erythropoietin (EPO)–induced differentiation of TF-1 cells. Unlike TF-1/pLVX cells, EPO treatment of TF-1/R140Q cells failed to induce differentiation, as evidenced by the lack of color change associated with hemoglobin expression (Fig. 2E). Next, we looked at the expression of hemoglobin gamma (HBG) and Kruppel-like factor 1 (KLF1), as both genes are known to be up-regulated during erythropoiesis (21). We found that EPO induced the expression of both genes in vector cells but not in IDH2/R140Q cells with high concentrations of 2HG (Fig. 2, F and G). However, treatment of TF-1/R140Q cells with AGI-6780, at concentrations that lowered 2HG to near-normal physiologic levels, restored expression of both HBG and KLF1 genes and the color change associated with differentiation (Fig. 2, E to G).

To investigate the effect of AGI-6780 on primary human AML cells ex vivo, we treated patient samples containing either the IDH2/R140Q mutant (n = 4) or IDH2/WT (n = 5) (table S2). Patient’s blood or bone marrow samples were sorted (Sytox staining) and cultured in condi-
tioned medium either in the presence or absence of a single dose of AGI-6780, and samples from each culture were collected over a 2-week time period (days 1, 3, 8, 10, and 13). IDH2 mutant samples treated with AGI-6780 showed a dose-dependent reduction in the amounts of extracellular (Fig. 3A and fig. S4A) and intracellular (Fig. 3B) 2HG. In the presence AGI-6780, a burst of cell proliferation was also observed only in the IDH2/R140Q mutant samples starting at day 4 of culture (Fig. 3C). Furthermore, AGI-6780 did not exert a cytotoxic effect as the number of viable, more mature CD45-positive cells increased in the presence of AGI-6780 only in the IDH2/R140Q mutant samples whereas the IDH2/WT samples remained unchanged (fig. S4B). In these studies, blasts were identified by flow cytometry analysis as CD45low cells with low side scatter (SSClow) using a SSC versus CD45 plot. After 6 days of inhibitor treatment, a dose-dependent decrease in the percentage of CD45low blast cells was observed in the IDH2/R140Q samples but not in IDH2/WT blasts (fig. S5A).

In parallel, maturation of AML blasts was also evaluated by flow cytometry analysis for changes in cell surface markers associated with monocytic and granulocytic differentiation (CD14, CD15, and CD11b) along with intracellular myeloperoxidase (MPO) as an indicator of maturation into the neutrophilic pathway. Although all samples (wild-type and IDH2/R140Q) underwent some level of spontaneous differentiation as a result of the cell-culturing conditions (i.e., cytokine addition), treatment with AGI-6780 induced an increase in the number of differentiated cells only in IDH2/R140Q patient samples. In the presence of AGI-6780, an increase in the mean fluorescence intensity for all cell surface markers and MPO could be seen in the IDH2 mutant samples, but not in wild-type IDH2 samples (Fig. 3D). Similarly, cytology revealed that by day 8 to 9 of treatment, the percentage of blasts or myeloblasts had decreased, and early signs of maturation were observed by the dose-dependent increase in the number of myelocytes and metamyelocytes (fig. S5, B to D). These effects on differentiation were only seen in mutant samples.

To establish that patient samples carrying the IDH2/R140Q allele were not counterselected during the in vitro differentiation assays, we cultured unfractionated nucleated bone marrow cells from patient #3 (IDH2/R140Q) for 13 days in methylcellulose-containing growth factors (stem cell factor, GM-CSF, interleukin-3, and erythropoietin) in the presence or absence of 5 μM AGI-6780. In the presence of AGI-6780, cells showed a threefold increase in the number of colony-forming units when compared to the number formed in vehicle-treated cells (fig. S5E). We determined the proportion of the IDH2/R140Q mutant among the colonies to eliminate the participation of residual normal progenitors in this

**Fig. 3. AGI-6780 can induce blast differentiation in primary human IDH2/R140Q AML patient samples.** A dose-dependent decrease in extracellular (A) and intracellular (B) 2HG was observed from IDH2/R140Q patient samples treated with AGI-6780. NT: no treatment. (C) The absolute number of cells increases upon treatment with AGI-6780 (5 μM) in IDH2/R140Q patient samples but not in IDH2/WT patient samples. Values were normalized to vehicle-treated cells and data are presented as mean ± SEM IDH2/R140Q (n = 3; closed circles, patients #1 to 3) and IDH2/WT (n = 3; open circles, patients #5 to 7) samples. (D) Flow cytometry analysis shows an increase in CD11b-, CD14-, CD15-, and MPO-positive cells in IDH2/R140Q patient samples but not in IDH2/WT samples after treatment with AGI-6780. The association between AGI-6780 treatment and cell surface maturation markers expression was tested with the Chi-square test with Yates’ continuity correction; P values are indicated.
process. The heterozygous IDH2/R140Q mutation was present in all of the colonies cultured with or without AGI-6780, confirming that the inhibitor promotes outgrowth and differentiation of cells harboring the mutated allele (fig. 5SF). Similar results were obtained with patient #3’s circulating leukemic cells cultured in methylcellulose and in liquid media. Taken together, these data demonstrate that targeted inhibition of IDH2/R140Q by AGI-6780 can specifically induce differentiation of AML blasts derived from patients with the IDH2/R140Q mutation.

In this study, we have used GM-CSF–dependent erythroleukemia TF-1 cells expressing IDH2/R140Q and a tool compound, AGI-6780, as a model system for evaluating the potential utility of IDH2 mutant inhibitors in the treatment of leukemias expressing the IDH2/R140Q mutant. Expression of mutant IDH2 in these cells conferred growth factor–independent proliferation and induced a block in erythroid differentiation that was reversed with AGI-6780 treatment. Furthermore, ex vivo treatment of primary human IDH2/R140Q mutant AML cells with AGI-6780 resulted in differentiation of the AML blasts down the macrophage and granulocytic lineages. These data are reminiscent of the leukemic cell differentiation response observed upon exposure to all-trans retinoic acid (ATRA) in acute promyelocytic leukemia (22) and demonstrate that inhibition of mutant IDH2 can relieve a block in differentiation present in this leukemic subset. Finally, these data support the clinical evaluation of IDH2 mutant–targeted agents in AML and other malignancies.

References and Notes
5. W. Xu et al., Cancer Cell 19, 17 (2011).
8. M. E. Figueroa et al., Cancer Cell 18, 553 (2010).
14. G. Jin et al., Cancer Res. 73, 496 (2013).

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Supplementary Materials
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Materials and Methods
Figs. S1 to S5
Tables S1 and S2
References (23–33)
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An Inhibitor of Mutant IDH1 Delays Growth and Promotes Differentiation of Glioma Cells

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The recent discovery of mutations in metabolic enzymes has rekindled interest in harnessing the altered metabolism of cancer cells for cancer therapy. One potential drug target is isocitrate dehydrogenase 1 (IDH1), which is mutated in multiple human cancers. Here, we examine the role of mutant IDH1 in fully transformed cells with endogenous IDH1 mutations. A selective R132H-IDH1 inhibitor (AGI-5198) identified through a high-throughput screen blocked, in a dose-dependent manner, the ability of the mutant enzyme (mIDH1) to produce -2-hydroxyglutarate (2-HG). Under conditions of near-complete -2HG inhibition, the mIDH1 inhibitor induced demethylation of histone H3K9me3 and expression of genes associated with gliogenic differentiation. Blockade of mIDH1 impaired the growth of IDH1-mutant—but not IDH1–wild-type–glioma cells without appreciable changes in genome-wide DNA methylation. These data suggest that mIDH1 may promote glioma growth through mechanisms beyond its well-characterized epigenetic effects.

Structural mutations in the metabolic enzyme isocitrate dehydrogenase (IDH1) have recently been identified in multiple human cancers, including glioma (1, 2), sarcoma (3, 4), acute myeloid leukemia (5, 6), and others. All mutations map to arginine residues in the catalytic pockets of IDH1 (R132) or IDH2 (R140 and R172) and confer on the enzymes a new activity: catalysis of alpha-ketoglutarate (2-OG) to the (R)-enantiomer of 2-hydroxyglutarate (R-2HG) (7, 8). R-2HG is structurally similar to 2-OG and, due to its accumulation to millimolar concentrations in IDH1-mutant tumors, competitively inhibits 2-OG–dependent dioxygenases (9).

The mechanism by which mutant IDH1 contributes to the pathogenesis of human glioma remains incompletely understood. Mutations in IDH1 are found in 50 to 80% of human low-grade (WHO grade II) glioma, a disease that progresses to fatal WHO grade III (anaplastic glioma) and WHO grade IV (glioblastoma) tumors over the course of 3 to 15 years. IDH1 mutations appear to precede the occurrence of other mutations (10) and are associated with a distinctive gene-expression profile (“proneural” signature), DNA hypermethylation [CpG island methylator phenotype (CIMP)], and certain clinicopathological features (11–13). When ectopically expressed in immortalized human astrocytes, R132H-IDH1 promotes the growth of these cells in soft agar (14) and induces epigenetic alterations found in IDH1-mutant human gliomas (15, 16). However, no tumor formation was observed when R132H-IDH1 was expressed from the endogenous IDH1 locus in several cell types of the murine central nervous system (17).

To explore the role of mutant IDH1 in tumor maintenance, we used a compound that was identified in a high-throughput screen for compounds that inhibit the IDH1-R132H mutant homodimer (fig. S1 and supplementary materials) (18).

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