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AUTHOR'S VIEW

Metabolic reprogramming of pyruvate dehydrogenase is essential for the proliferation of glioma cells expressing mutant IDH1

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ABSTRACT

Mutations in isocitrate dehydrogenase 1 (*IDH1*) characterize most adult low-grade gliomas. Mutant IDH1 catalyzes production of the oncometabolite 2-hydroxyglutarate (2-HG). We recently discovered that the *IDH1* mutation also reprograms pyruvate metabolism in a 2-HG-dependent manner, and that reprogramming of pyruvate metabolism is essential for cell proliferation in glioma cells with mutant IDH1.

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Glioma; IDH1; metabolic reprogramming; pyruvate dehydrogenase; 2-HG

Metabolic reprogramming is increasingly viewed as a hallmark of cancer, with several common metabolic alterations such as increased glycolysis reported in a range of tumor types;[1] however, the metabolic alterations associated with recently discovered oncogenic mutations in isocitrate dehydrogenase 1 (*IDH1*) remain to be fully elucidated.

Mutations in *IDH1* characterize over 80% of adult low-grade gliomas. Cytosolic IDH1 normally catalyzes the production of α-ketoglutarate (α-KG) from isocitrate and plays an important role in the regulation of redox status, lipogenesis, and glucose and glutamine metabolism. Mutations in IDH1, most commonly at the R132 residue in the active site of the enzyme, instead lead to the conversion of α-KG to 2-hydroxyglutarate (2-HG). By inhibiting the activity of a variety of cellular α-KG-dependent enzymes, 2-HG induces epigenetic changes that block cellular differentiation and induce tumorigenesis.[2] We and others have shown that, in addition to an altered epigenetic profile and elevated 2-HG levels, IDH1 mutant cells also undergo broader metabolic reprogramming compared to their wild-type IDH1 counterparts.[3,4] Most notably, we observed a significant reduction in ^{1}H magnetic resonance spectroscopy (MRS)-detectable steady-state levels of lactate, phosphocholine, and glutamate in 2 genetically engineered cell models expressing mutant IDH1—a U87 glioblastoma-based model and a normal human astrocyte (NHA) model.[4] In a separate study we discovered that pyruvate dehydrogenase (PDH) activity was reduced in IDH1 mutant NHA cells.[5] Given that PDH is an important regulatory point for glucose oxidation via the tricarboxylic acid (TCA) cycle and, as a result, for glutamate production, we questioned the role of PDH in IDH1 mutant glioma cells.[6]

In a recently published study[6] we confirmed a significant reduction in PDH activity in both our U87 and NHA mutant IDH1 cells compared to wild-type.[6] ^{13}C MRS probing of the fate of 1-^{13}C-glucose to 4-^{13}C-glutamate, and hyperpolarized ^{13}C MRS probing of the fate of 2-^{13}C-pyruvate to 5–^{13}C-glutamate, showed that reduced PDH activity also resulted in a reduction in glucose flux to glutamate in IDH1 mutant cells relative to wild-type. This was consistent with, and mostly explained, the decrease in steady-state glutamate levels. We further found that IDH1 mutant cells showed increased expression, at both mRNA and protein levels, of pyruvate dehydrogenase kinase 3 (PDK3), a well-known regulator of PDH activity. The increase in PDK3 expression correlated with increased inhibitory phosphorylation of PDH, thereby explaining the reduction in PDH activity in IDH1 mutant cells. This effect was associated with increased levels of hypoxia inducible factor-1α (HIF-1α) in IDH1 mutant cells, consistent with previous observations and the stabilization of HIF-1α *via* inhibition of α-KG-dependent-prolyl hydroxylases.[7,8] Finally, we also showed that treatment of IDH1 wild-type cells with 2-HG recapitulated the effects of the IDH1 mutation, with increased levels of PDK3 and HIF-1α and decreased PDH activity in 2-HG treated cells. Collectively, our results therefore suggested that *IDH1* mutation results in increased PDK3 expression thereby reducing PDH activity and explaining the reduction in glutamate levels. Fig. 1 summarizes our findings and the mechanism by which mutant IDH1 leads to inhibition of PDH activity. Our finding was important because it indicated that the reprogramming of pyruvate metabolism is not linked to 2-HG-induced hypermethylation, but occurs on a timescale that is faster and possibly more amenable to intervention.

To test the value of PDH as a therapeutic target, we went on to address the functional consequences of reduced PDH activity in IDH1 mutant cells. Our study showed, to our knowledge for the first time, that reversing the metabolic reprogramming of PDH in mutant IDH1 cells was detrimental to the proliferation and clonogenic potential of these cells. Specifically, treatment with dichloroacetate (DCA), a PDK inhibitor,[9] not only increased PDH activity and glutamate production in IDH1

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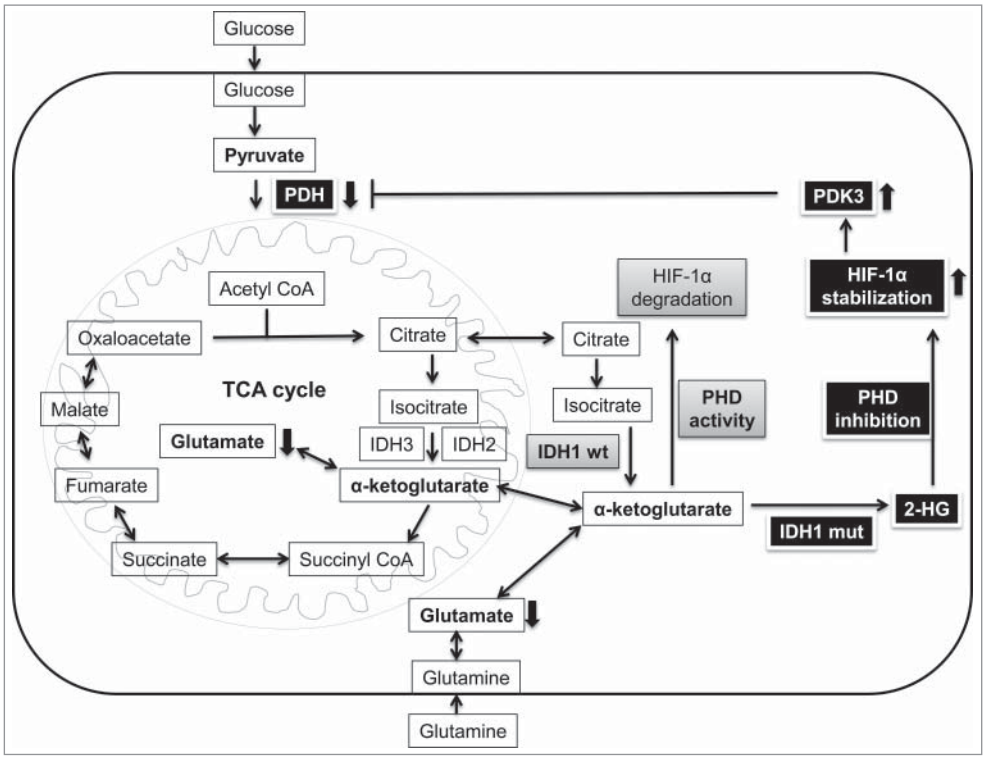

Figure 1. Isocitrate dehydrogenase 1 (IDH1) mutations lead to the downregulation of PDH activity. IDH1 wild-type enzyme (IDH1 wt) converts isocitrate to α-ketoglutarate (α-KG) in the cytosol. α-KG can then re-enter the tricarboxylic acid (TCA) cycle within mitochondria or remain in the cytosol and function as a co-factor for various enzymes, one of which is the family of prolyl hydroxylases (PHD). PHDs hydroxylate proline residues on hypoxia inducible factor-1α (HIF-1α), thereby targeting HIF-1α for proteasomal degradation. Mutant IDH1 enzyme (IDH1 mut), on the other hand, converts α-KG to 2-hydroxyglutarate (2-HG). 2-HG inhibits PHD activity, thereby leading to stabilization of HIF-1α levels. HIF-1α upregulates expression of pyruvate dehydrogenase kinase 3 (PDK3), which then phosphorylates and inhibits PDH activity.

mutant cells, but also completely abrogated the increased clonogenicity observed in cells expressing mutant IDH1. Moreover, DCA also inhibited proliferation of patient-derived mutant IDH1 neurosphere cultures, thereby also validating our findings in clinically relevant models. DCA treatment also reversed the metabolic alterations detected by ^{1}H and ^{13}C MRS.

Our results thus suggest that the reduction in PDH activity induced by the *IDH1* mutation is essential for proliferation and clonogenicity in IDH1 mutant glioma cells, and identify PDH as a possible therapeutic target for the treatment of mutant IDH1 cells. Our findings also highlight the value of MRS in elucidating the mechanisms of metabolic reprogramming in IDH1 mutant glioma cells. Importantly, ^{1}H MRS has been used as a non-invasive method of evaluating brain tumors in human patients, and, more recently, the value of hyperpolarized ^{13}C MRS was also demonstrated in patients.[10]

In summary, our recent study[6] identifies a potential therapeutic target for mutant IDH1 low-grade gliomas, as well as an associated companion MRS biomarker for agents that would modulate that target.

Disclosure of potential conflicts of interest

No potential conflicts of interest were disclosed.

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References

1. Phan LM, Yeung SCJ, Lee MH. Cancer metabolic reprogramming: importance, main features, and potentials for precise targeted anti-cancer therapies. Cancer Biol Med 2014; 11(1):1-19; PMID:24738035; http://dx.doi.org/10.7497/j.issn.2095-3941.2014.01.001
2. Yang H, Ye D, Guan KL, Xiong Y. IDH1 and IDH2 mutations in tumorigenesis: mechanistic insights and clinical perspectives. Clin Cancer Res 2012; 18(20):5562-71; PMID:23071358; http://dx.doi.org/10.1158/1078-0432.CCR-12-1773
3. Reitman ZJ, Jin G, Karoly ED, Spasojevic I, Yang J, Kinzler KW, He Y, Bigner DD, Vogelstein B, Yan H. Profiling the effects of isocitrate dehydrogenase 1 and 2 mutations on the cellular metabolome. Proc Natl Acad Sci U S A 2011; 108(8):3270-5; PMID:21289278; http://dx.doi.org/10.1073/pnas.1019393108
4. Izquierdo-Garcia JL, Viswanath P, Eriksson P, Chaumeil MM, Pieper RO, Phillips JJ, Ronen SM. Metabolic Reprogramming in Mutant IDH1 Glioma Cells. PLoS One 2015; 10(2):e0118781; PMID:25706986; http://dx.doi.org/10.1371/journal.pone.0118781
5. Izquierdo-Garcia JL, Cai LM, Chaumeil MM, Eriksson P, Robinson AE, Pieper RO, Phillips JJ, Ronen SM. Glioma cells with the IDH1 mutation modulate metabolic fractional flux through pyruvate carboxylase. PLoS One 2014; 9(9):e108289; PMID:25243911; http://dx.doi.org/10.1371/journal.pone.0108289
6. Izquierdo-Garcia JL, Viswanath P, Eriksson P, Cai L, Radoul M, Chaumeil MM, Blough M, Luchman HA, Weiss S, Cairncross JG, et al. IDH1 mutation induces reprogramming of pyruvate metabolism. Cancer Res 2015; 75(15):2999-3009; PMID:26045167
7. Xu W, Yang H, Liu Y, Yang Y, Wang P, Kim SH, Ito S, Yang C, Wang P, Xiao MT, et al. Oncometabolite 2-hydroxyglutarate is a competitive inhibitor of alpha-ketoglutarate-dependent dioxygenases. Cancer Cell 2011; 19(1):17-30; PMID:21251613; http://dx.doi.org/10.1016/j.ccr.2010.12.014
8. Sasaki M, Knobbe CB, Itsumi M, Elia AJ, Harris IS, Chio, II, Cairns RA, McCracken S, Wakeham A, Haight J, et al. D-2-hydroxyglutarate produced by mutant IDH1 perturbs collagen maturation and

basement membrane function. Genes Dev 2012; 26(18):2038-49; PMID:22925884; http://dx.doi.org/10.1101/gad.198200.112

9. Sutendra G, Michelakis ED. Pyruvate dehydrogenase kinase as a novel therapeutic target in oncology. Front Oncol 2013; 3:38; PMID:23471124; http://dx.doi.org/10.3389/fonc.2013.00038

10. Nelson SJ, Kurhanewicz J, Vigneron DB, Larson PE, Harzstark AL, Ferrone M, van Criekinge M, Chang JW, Bok R, Park I, et al. Metabolic imaging of patients with prostate cancer using hyperpolarized ; 1-(1)(3)Cpyruvate. Sci Transl Med 2013; 5(198):198ra08; http://dx.doi.org/10.1126/scitranslmed.3006070