



(R)-2-hydroxyglutarate drives immune quiescence in the tumor microenvironment of IDH-mutant gliomas

Leland G. Richardson, Bryan D. Choi, William T. Curry

Department of Neurosurgery, Massachusetts General Hospital, Harvard Medical School, Boston, MA, USA

Correspondence to: William T. Curry. Department of Neurosurgery, Massachusetts General Hospital, Harvard Medical School, 55 Fruit Street, Boston, MA 02114, USA. Email: wcurry@mgh.harvard.edu.

Comment on: Bunse L, Pusch S, Bunse T, *et al.* Suppression of antitumor T cell immunity by the oncometabolite (R)-2-hydroxyglutarate. *Nat Med* 2018;24:1192-203.

Submitted Dec 14, 2018. Accepted for publication Jan 02, 2019.

doi: 10.21037/tcr.2019.01.08

View this article at: <http://dx.doi.org/10.21037/tcr.2019.01.08>

Diffuse low-grade and anaplastic gliomas (grades II and III, respectively) are infiltrative primary brain tumors that are considered malignant because of their invasiveness, resistance to therapy, and likelihood of transforming to higher grades. Median survival varies from 3 to 15 years as a proportion relentlessly progress, while others may remain stable for years (1). Molecular classification plays an integral role in the diagnosis and prognostication of patients with gliomas, as evidenced by the recently revised 2016 World Health Organization (WHO) Classification of Central Nervous System Tumors (2). One genetic marker that is of critical importance is the isocitrate dehydrogenase 1 (*IDH*) gene, which is found to be mutated in 80–90% of all WHO grades II and III gliomas and in a minority of grade IV tumors (glioblastoma, or GBM) (3). The most striking clinical feature of IDH-mutant (IDH-1 or IDH-2) gliomas, irrespective of tumor grade and histology, is their survival advantage when compared to IDH-wildtype gliomas. Although these tumors exhibit profound alterations to their genetic, metabolic and immunologic make up, it is still unclear what drives this survival advantage. Since its discovery, the R132H IDH mutation has changed the way that glial tumors are conceptualized and consequently treated, and it is well appreciated that the underlying biology and clinical behavior of these tumors are distinct and respond differently to current treatment paradigms (4).

While patients bearing IDH-mutant tumors have improved prognoses compared to those with wildtype tumors, most eventually succumb to disease, exacting a significant toll on human lifespan and productivity. Given that IDH-mutant glioma patients maintain the mutant allele

throughout progression and even after recurrence to higher grade, this highly conserved genetic alteration, expressed solely in tumor cells, represents an attractive therapeutic target, including, potentially, for immunotherapy.

To date, most brain tumor immunotherapy clinical trials have engaged subjects with GBM, most of which is genotypically IDH-wildtype. Far fewer protocols have been available for patients with low-grade and anaplastic glioma (5). However, there has been an increasing effort to target mutant IDH through small molecule inhibitors, immune checkpoint inhibition, and vaccination, both preclinically and in human subjects (6,7). As we assess disease response, it is crucial that we comprehensively study and understand the interactions between host immunity and IDH-mutant tumors within the central nervous system (CNS).

A growing literature suggests that IDH-1 mutation confers an immunologically quiescent phenotype (*Figure 1*) onto glial tumors. IDH-mutant gliomas exhibit fewer tumor infiltrating lymphocytes (TILs) and reduced protein expression of programmed death ligand 1 (PD-L1) than wild-type counterparts (8). This relationship is recapitulated at the transcriptional level by data analyzed from The Cancer Genome Atlas (TCGA). Additional TCGA studies have also shown IDH-mutant mediated immune suppression through reduced expression of CD8⁺ T cell and interferon- γ (IFN- γ) associated genes, critical for the recruitment and activation of a vigorous anti-tumor T cell response (9,10). It is tempting to speculate that IDH-mutant gliomas would respond less well to checkpoint inhibition relative to IDH-wildtype tumors, but no clinical data confirms this hypothesis (11).

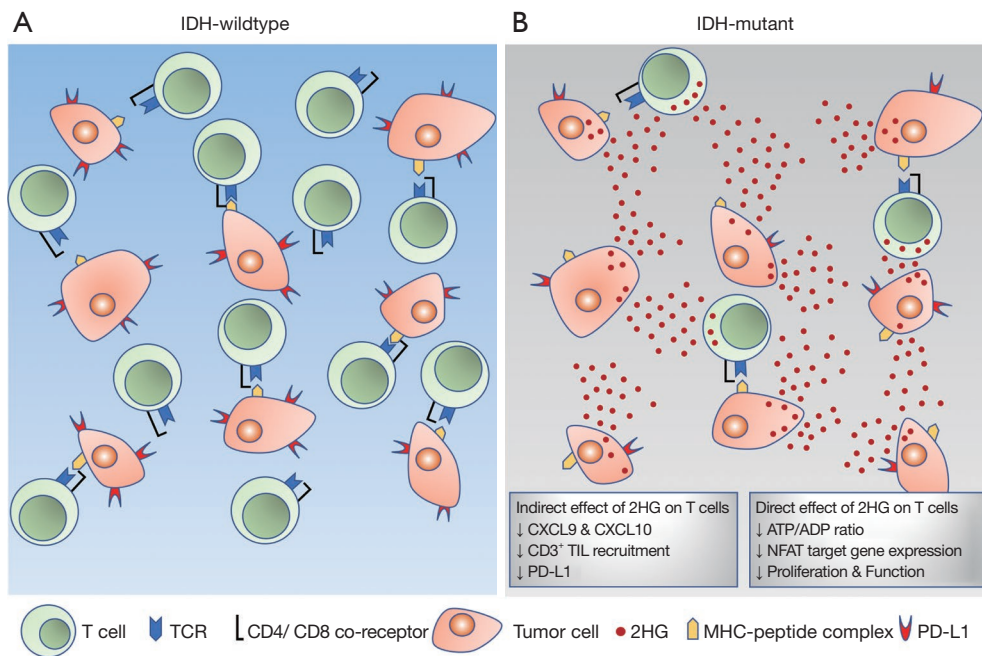


Figure 1 Overview of the distinct tumor immune microenvironment of IDH-mutant gliomas. (A) IDH-wildtype glioma contains greater number of TILs and PD-L1 expression, while (B) IDH-mutant glioma exhibit an immunologically quiescent tumor microenvironment, displaying fewer TILs and diminished expression of PD-L1. Here, 2HG-mediated T cell suppression occurs through two disparate mechanisms. (I) The indirect tumor intrinsic mechanism prevents the recruitment of T cells by lowering CXCL9 and CXCL10 levels. (II) 2HG accumulates outside of IDH-mutant tumor cells and enters T cells through a direct paracrine fashion, altering TCR signaling pathways, resulting in the impairment of T cell anti-tumor immunity. As such, combining IDH inhibition with immunotherapy is of interest for the management of IDH-mutant gliomas. IDH, isocitrate dehydrogenase; TILs, tumor infiltrating lymphocytes; TCR, T-cell receptor; 2HG, (R)-2-hydroxyglutarate; MHC, major histocompatibility complex; PD-L1, programmed death-ligand 1; NFAT, nuclear factor of activated T cells; CXCL9, C-X-C motif chemokine 9; CXCL10, C-X-C motif chemokine 10.

Nevertheless, mounting evidence suggests that the enzymatic product of mutant IDH plays a role in modulating anti-tumor immunity. (R)-2-hydroxyglutarate (2HG) is an oncometabolite produced by the neomorphic activity of the mutated IDH enzyme, which catalyzes the conversion of α -ketoglutarate to 2HG (12). The accumulation of 2HG leads to DNA hypermethylation through the inhibition of key histone demethylase enzymes (13). 2HG-mediated hypermethylation leads to epigenetic dysregulation, resulting in the repression of key immune related genes (10). 2HG appears to inhibit CD8⁺ T cell recruitment in IDH-mutant gliomas. In one preclinical model, the introduction of 2HG, either transgenically or through therapeutic delivery, led to the diminished leukocyte production of C-X-C motif chemokine 9 (CXCL9) and C-X-C motif chemokine 10 (CXCL10) through the suppression of signal transducer and activator of transcription 1 (STAT1). This in turn resulted in

reduced CD8⁺ T cell infiltration of tumors, consistent with observations in human immunohistochemistry (IHC) studies. Importantly, blocking the production of 2HG through an IDH inhibitor reversed this immunosuppressive phenotype (9). While tumor intrinsic epigenetic dysregulation is implicated as one source of immune impairment in IDH-mutant gliomas, a recent study published in *Nature Medicine* by an international multicenter group introduces an additional mechanism, centered around the direct effect of 2HG on tumor infiltrating T cells (14).

Bunse *et al.* (14) elegantly and thoroughly demonstrate that 2HG exported from tumor cells is imported by T cells in a paracrine fashion through a sodium-dependent dicarboxylate transport system, resulting in the direct suppression of T cell activation and proliferation (*Figure 1*). Moreover, metabolically, they show that 2HG inhibits an ATP dependent T cell receptor (TCR) signaling and polyamine biosynthesis pathway, which leads to diminished

T cell proliferation and function through the reduction of nuclear factor of activated T cells (NFAT) target gene expression. Oral administration of an IDH inhibitor in combination with R132H peptide vaccination, adoptive T-lymphocyte immunotherapy, or PD-1 blockade demonstrated synergistic effects resulting in increased overall survival in a syngeneic orthotopic IDH-mutant mouse model (14) in addition to measurably boosting the Th1 immune response within the tumors. Given that several IDH inhibitors are currently in clinical trials, these data offer support for human studies combining treatment with IDH inhibition with immunotherapy.

Central questions remain. For instance, why do patients with IDH-mutant glioma live longer despite such baseline intratumoral immune quiescence? What other aspects of the IDH-modulated tumor immune microenvironment may impact the natural course of disease in these tumors? Though touched upon by Bunse and colleagues, further study is needed to determine the effects of 2HG on infiltrating myeloid cells, that are often present at greater frequencies than T-cell populations within the glioma tumor microenvironment. Do myeloid lineage cells import 2HG as do the T cells and, if so, does intracellular accumulation of 2HG influence tumor associated macrophage polarization? As 2HG is exported from tumor cells into extracellular spaces and can be detected systemically (15), does it exert effects on tumor immunity beyond the immediate tumor milieu?

Clinical questions that remain unanswered include whether WHO grade is associated with the host immune response to IDH-mutant tumors, and what might be the impact of either tumor progression or contemporary treatment of these tumors, including ionizing radiation and alkylating chemotherapy. Does therapeutic induction of a hypermutated state render these tumors more immunotherapy responsive, mimicking the state of mismatch repair deficiency in colorectal carcinoma (16), or is persistence of the IDH mutation sufficient to maintain hyporesponsiveness over time?

Bunse *et al.* and others have provided ample mechanistic support for going forward with clinical studies combining latest or next generation IDH inhibitors with immunotherapy, whether it be vaccination, IDH-directed adoptive T lymphocyte transfer, or immune checkpoint blockade. While IDH mutations may be oncogenic and drive tumor-associated immunosuppression, they may also provide keys to eradicating these tumors, using rational combinations of molecularly targeted therapy

and immunotherapy. Given the relatively long survival of patients with IDH mutant low-grade gliomas, surrogate markers of immune and tumor response will be of great use in these future studies.

Acknowledgments

Funding: This work was supported by the Jenny Fund (www.thejennyfund.org).

Footnote

Provenance and Peer Review: This article was commissioned and reviewed by the Section Editor Xian-Xin Qiu (Shanghai Proton and Heavy Ion Center (SPHIC), a.k.a. the Proton and Heavy Ion Center of Fudan University Shanghai Cancer Center (FUSCC), Shanghai, China).

Conflicts of Interest: All authors have completed the ICMJE uniform disclosure form (available at <http://dx.doi.org/10.21037/tcr.2019.01.08>). The authors have no conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

Open Access Statement: This is an Open Access article distributed in accordance with the Creative Commons Attribution-NonCommercial-NoDerivs 4.0 International License (CC BY-NC-ND 4.0), which permits the non-commercial replication and distribution of the article with the strict proviso that no changes or edits are made and the original work is properly cited (including links to both the formal publication through the relevant DOI and the license). See: <https://creativecommons.org/licenses/by-nc-nd/4.0/>.

References

1. Cancer Genome Atlas Research Network, Brat DJ, Verhaak RG, et al. Comprehensive, Integrative Genomic Analysis of Diffuse Lower-Grade Gliomas. *N Engl J Med* 2015;372:2481-98.
2. Louis DN, Perry A, Reifenberger G, et al. The 2016 World Health Organization Classification of Tumors of the Central Nervous System: a summary. *Acta Neuropathol* 2016;131:803-20.

3. Yan H, Parsons DW, Jin G, et al. IDH1 and IDH2 mutations in gliomas. *N Engl J Med* 2009;360:765-73.
4. Miller JJ, Shih HA, Andronesi OC, Cahill DP. Isocitrate dehydrogenase-mutant glioma: Evolving clinical and therapeutic implications. *Cancer* 2017;123:4535-46.
5. Pollack IF, Jakacki RI, Butterfield LH, et al. Antigen-specific immunoreactivity and clinical outcome following vaccination with glioma-associated antigen peptides in children with recurrent high-grade gliomas: results of a pilot study. *J Neurooncol* 2016;130:517-27.
6. Schumacher T, Bunse L, Pusch S, et al. A vaccine targeting mutant IDH1 induces antitumour immunity. *Nature* 2014;512:324-7.
7. Han CH, Batchelor TT. Isocitrate dehydrogenase mutation as a therapeutic target in gliomas. *Chin Clin Oncol* 2017;6:33.
8. Berghoff AS, Kiesel B, Widhalm G, et al. Correlation of immune phenotype with IDH mutation in diffuse glioma. *Neuro Oncol* 2017;19:1460-8.
9. Kohanbash G, Carrera DA, Shrivastav S, et al. Isocitrate dehydrogenase mutations suppress STAT1 and CD8+ T cell accumulation in gliomas. *J Clin Invest* 2017;127:1425-37.
10. Amankulor NM, Kim Y, Arora S, et al. Mutant IDH1 regulates the tumor-associated immune system in gliomas. *Genes Dev* 2017;31:774-86.
11. Choi BD, Curry WT. IDH mutational status and the immune system in gliomas: a tale of two tumors? *Transl Cancer Res* 2017;6:S1253-6.
12. Dang L, White DW, Gross S, et al. Cancer-associated IDH1 mutations produce 2-hydroxyglutarate. *Nature* 2010;465:966.
13. Turcan S, Rohle D, Goenka A, et al. IDH1 mutation is sufficient to establish the glioma hypermethylator phenotype. *Nature* 2012;483:479-83.
14. Bunse L, Pusch S, Bunse T, et al. Suppression of antitumor T cell immunity by the oncometabolite (R)-2-hydroxyglutarate. *Nat Med* 2018;24:1192-203.
15. Kalinina J, Ahn J, Devi NS, et al. Selective Detection of the D-enantiomer of 2-Hydroxyglutarate in the CSF of Glioma Patients with Mutated Isocitrate Dehydrogenase. *Clin Cancer Res* 2016;22:6256-65.
16. Le DT, Uram JN, Wang H, et al. PD-1 Blockade in Tumors with Mismatch-Repair Deficiency. *N Engl J Med* 2015;372:2509-20.

Cite this article as: Richardson LG, Choi BD, Curry WT. (R)-2-hydroxyglutarate drives immune quiescence in the tumor microenvironment of IDH-mutant gliomas. *Transl Cancer Res* 2019;8(Suppl 2):S167-S170. doi: 10.21037/tcr.2019.01.08