Multiple expressed endogenous glioma epitopes as novel vaccines for gliomas

Pedro R. Lowenstein and Maria G. Castro

Department of Neurosurgery, and Department of Cell and Developmental Biology, The Cancer Center, The Cancer Biology and The Immunology Graduate Programs, The University of Michigan School of Medicine, Ann Arbor, Michigan

Please address all correspondence to:
P.R. Lowenstein, MD, PhD
Richard Schneider Collegiate Professor
Professor, Dept. of Neurosurgery,
Professor, Dept. of Cell and Developmental Biology
MSRB II, Room 4570
Department of Neurosurgery
University of Michigan School of Medicine
1150 West Medical Center Drive
Ann Arbor, MI 48109-5689

Phone (office) 734-764-0851
e-mail: pedrol@umich.edu

The authors have no conflicts to declare.
Summary
A novel approach to immunization against glioma tumors is described. Immunization against eleven antigens expressed in malignant brain tumors elicits responses to one or more antigens in a large percentage of patients. This novel approach suggests that it could be extended to a Phase III trial.

In this issue of CLINICAL CANCER RESEARCH, Rampling and colleagues show the data from a first in man immunization of glioma patients with a vaccine containing 10 antigenic epitopes known to be expressed in glioma. Immunization to treat solid tumors has a long history. Non-specific immunization led to some individual responses, and these early results were then adapted by novel contemporary understandings of immune system function. More recent successes in the treatment of melanoma, small cell lung carcinoma and kidney cancer, as well as others being evaluated, with immunotherapy has reignited hope that similar approaches will work in patients with malignant brain tumors. Inhibitors of co-stimulating molecule pairs CTLA4-CD28 and PD1-PDL1 have shown strong anti-tumoral activities across disease stages, even after failure of standard of care, and various stages of disease progression. This comes after many years of various approaches to systemic immunization providing partially effective results without any definite breakthroughs. Vaccination in cancer has employed potential tumor antigens deduced from tumor proteins, in vitro activation of tumor infiltrating leucocytes, alone, or in combination with suppression of endogenous immune system followed by bone marrow transplantation to potentiate leukocyte engraftment. Results were marginally effective, with only occasional complete responses and long term patient survival. Checkpoint inhibitors have transformed the landscape of solid cancer immunotherapy achieving, in a clinically significant percentage of patients, complete remission, and partial remission in significant numbers.

Highly malignant gliomas (WHO grade IV; glioblastoma multiforme; GBM) remain universally fatal with median survival ~16-18 months. These are some of the most frequently targeted tumors as long term survival has improved marginally during the last 80 years. Surgery, radiation and chemotherapy constitute the standard of care; yet long term survival beyond 2-3 years is rare. Immunotherapy has been proposed and tested in patients with GBM. Potential tumor antigens derived from potentially immunogenic proteins have been used to stimulate the immune response to recognize these antigens in the patients’ GBM. Alternatively, groups have utilized patients’ own tumor lysates to stimulate the patients’ dendritic cells to be used as vaccines to elicit immunity against their endogenous tumor antigens. In spite of measurable T cell responses, therapeutic results have been marginal with occasional clinically significant responses.

Several of these approaches have entered phase III clinical trials. In spite of positive earlier phase clinical trials results, the recent failure of a vaccine against EGFRvIII to extend survival of patients in a phase III trial continues to challenge to approaches that target single antigenic epitopes. Analogously, trials utilizing some of the other vaccination approaches have shown increases in PFS, but limited extension of OS. Why has immunization not shown stronger clinical efficiency in extending life of patient with GBM?

Possible explanations are that immunizations and innate immune adjuvants are not strong enough, that exogenous antigens used are not the same ones as the endogenous ones presented by tumors on HLA,
that not enough antigens are retrieved from tumors in approaches attempting to load dendritic cells with tumor antigens, or that tumor downregulation of HLA would sequester antigens from T-cell recognition. Studies of tumor antigen repertoire are cumbersome, and thus the recent description of the neoantigen repertoire in many different tumors is of great importance to tumor immunotherapy. Tumor neoantigen expression is highest in melanoma and lung cancer and lowest in AML, ALL, and in pilocytic astrocytoma. Interest in expression of tumor neoantigens reflects the theory that such antigens would not have been negatively selected during T-cell development. They therefore represent novel targets for the immune system. The correlation between the neoantigen expression repertoire and the clinical efficacy of the immune checkpoint inhibitors supports this hypothesis. It also predicts why vaccines utilizing a low number of antigens would be less likely to succeed.

Taking these practical and theoretical elements into consideration Rampling et al. (1) present data on a novel vaccination clinical trial for patients with glioblastoma in which vaccines are loaded with endogenous epitopes known to be presented on HLA molecules in GBM. Specifically, Dutoit et al. (7) characterized 46000 HLA-bound peptides in GBM, of which 3000 were selectively bound to the most common HLA in humans, i.e., HLA-A*02. They then concentrated on studying 10 GBM-associated antigens based on levels of expression in GBM, and low expression in normal organs. Importantly, and as expected of neoantigens, actual patients did not display any T-cell tolerance to the selected peptides. Equally, patients’ CD8 + T cells induced specific lysis of tumour cells in vitro which expressed the novel antigens. Further, the identification of T-cells specific for the novel antigens in the GBM microenvironment supports the idea that such T-cells could be functionally lytic in vivo as well.

Rampling et al. in this issue (1), report the translation of these exciting basic science findings into a first in human phase I trial of a novel multi-peptide therapeutic vaccine in patients recently diagnosed with GBM [FIGURE 1]. The study was performed in two-cohorts of patients treated with standard of care and vaccine immunotherapy. Patients were all HLA-A*02, and were treated with 11 intradermal immunizations. GM-CSF was used as adjuvant and vaccines were administered over 6 months. Patients responded to either single antigens (90%), or to multiple antigens (50%). Importantly pre-treatment levels of Tregs, administration of chemo-radiotherapy, or treatment with steroids did not affect the capacity of the vaccine to stimulate immune responses to GBM antigens. These data together with the previous finding of T-cells specific to the GBM antigens within the GBM microenvironment strongly support the assertion of future combination of the novel type of immunization with checkpoint inhibitors.

Overall survival was not increased when patients responding to multiple antigens were compared to those only responding to one antigen. Median survival was comparable to median survival of the general patient population. However, striking difference in survival was detected when patients were evaluated in relation to the presence or absence of an injection site response. Under this comparison, patients in whom an injection site response was evident, displayed twice the survival of those without injection site response. Though statistical analysis provides a significant p value, the authors are careful not to overinterpret these data given that this was a phase I, rather than a double blind randomized phase III trial. The data from this trial thus provide strong support for the continued development of this vaccination approach. Though follow up larger trials of this approach would be predictable, rapid phase I
trials combining this vaccination strategy and checkpoint inhibitors could provide stronger responses, and increase the availability of a potentially useful treatment to desperately ill patients.

In context there are currently a number of trials with varying approaches ongoing for GBM. Amongst others, oncolytic viruses are being tested in early phase trials \(^5\), as are small molecule inhibitors of enzymes thought to be essential for the survival of glioma cells \(^9\), gene and immunotherapy to reengineer the brain immune system (https://clinicaltrials.gov/ct2/show/NCT01811992) \(^10\), and novel radiation approaches are all currently ongoing. In view of the profound and urgent need of novel treatments for patients suffering from GBM we hope that all our efforts will significantly contribute to achieving clinical breakthrough results soon.

References

FIGURE LEGEND.

The figure shows schematically the translational aspects of this work. Specifically the top part of the figure describes the work of Dutoit et al. (7) who isolated glioma antigenic peptides presented specifically by HLA.A2 molecules (in red) on the membrane of glioma tumor cells. The middle part of the figure summarizes how Rampling et al. () utilized these antigenic peptides to immunize patients. The lower part of the figure illustrates the mechanism of action, whereby the antigenic peptides used as immunogens are taken up by dendritic cells (DC) and transported to the draining lymph nodes where they are presented to naïve T-cells (TCN). The resulting T-effector cells (TCE) then migrate to the tumor in the brain where they recognize the antigenic epitopes presented by glioma cells on HLA.A2, and kill such target glioma cells. The progress made by this approach is the simultaneous immunization with eleven antigens to reduce the capacity of glioma cells to escape immune attack by reducing expression of such targeted antigens.
Figure 1:

HLA.A2

Antigens isolated from HLA.A2-positive gliomas

Antigens used for immunization

Skin

Draining lymph nodes

Circulation

Glioma

DC

DC

TC

N

TC

N

TC

E

TC

E

Glioma cell death

HLA.A2

© 2016 American Association for Cancer Research

Author manuscripts have been peer reviewed and accepted for publication but have not yet been edited.

Downloaded from clincancerres.aacrjournals.org on June 11, 2021. © 2016 American Association for Cancer Research.
Multiple expressed endogenous glioma epitopes as novel vaccines for gliomas

Pedro R. Lowenstein and Maria G Castro

Clin Cancer Res Published OnlineFirst August 12, 2016.

Updated version

Access the most recent version of this article at:
doi:10.1158/1078-0432.CCR-16-1477

Author Manuscript

Author manuscripts have been peer reviewed and accepted for publication but have not yet been edited.

E-mail alerts

Sign up to receive free email-alerts related to this article or journal.

Reprints and Subscriptions

To order reprints of this article or to subscribe to the journal, contact the AACR Publications Department at pubs@aacr.org.

Permissions

To request permission to re-use all or part of this article, use this link http://clincancerres.aacrjournals.org/content/early/2016/08/18/1078-0432.CCR-16-1477. Click on "Request Permissions" which will take you to the Copyright Clearance Center's (CCC) Rightslink site.