A precision medicine umbrella trial to nominate promising therapies for glioblastoma

Jann N. Sarkaria

William H. Donner Professor

Department of Radiation Oncology

Mayo Clinic

Rochester, MN 55902

Sarkaria.jann@mayo.edu

The Neuro-oncology program of the German Cancer Consortium is congratulated for the completion of the paradigm-shifting Phase I/II NCT Neuro Master Match (N^2M^2) clinical trial[1]. In this precision medicine umbrella trial, patients with newly diagnosed glioblastoma (GBM) and an unmethylated MGMT promoter had comprehensive tumor molecular profiling including whole methylome array, whole exome sequncing, whole genome sequencing, RNA sequencing, and immunohistochemistry. Those patients with 'actionable' alterations, were assigned by a molecular tumor board to substudies evaluating radiation with concurrent and adjuvant palbociclib (CDK4/6 inhibitor), temsirolimus (mTORC1 inhibitor), idasanutlin (MDM2 inhibitor), alectinib (Alk inhibitor), or vismodegib (Smoothened inhibitor). The remaining patients without actionable alterations were randomized to therapy with atezolizumab (anti-PD-L1 antibody), asunercept (FAS-ligand inhibitor) or temozolomide (standard chemotherapy) during and after radiation. Salient results include i) limited to no accrual on the vismodegib, alectinib and idasanutlin substudies; ii) no therapeutic benefit for palbociclib, atezolizumab or asunercept; iii) promising progression-free and overall survival for temsirolimus. These results demonstrate the potential for umbrella trials to efficiently evaluate multiple clinical regimens and identify those regimens that are not suitable for further clinical study. While traditional Phase 2 testing of a single treatment regimen versus standard of care would have required at least 150 patients, the

N^2M^2 trial evaluated seven novel agents and nominated temsirolimus as the only promising regimen enrolling a total of 301 patients. The efficiency of this umbrella trial strategy cannot be overstated and will change how many novel agents for GBM are evaluated.

The success of the N^2M^2 umbrella trial design is highly dependent on use of accurate predictive biomarkers. Developing these biomarkers is resource intensive, as exemplified by the development of phospho-Ser2448-mTOR for temsirolimus. This immunohistochemistry assay first was validated through extensive laboratory testing and then tested in a multiplex analysis of samples from a trial randomizing MGMT unmethylated patients to treatment with RT/temsirolimus vs. RT/temozolomide. A multivariate analysis of this trial identified phospho-mTOR as a predictor of temsirolimus efficacy[2]. This rigorous approach to developing a predictive biomarker is commendable. However, other biomarkers were defined based on current understanding of complex signal transduction networks. For example, CDK4 amplification or CDKN2A/B deletion are anticipated to increase CDK4 signaling and was used as a predictor for response to palbociclib. While conceptually this makes sense, preclinical and clinical data supporting this biomarker signature are equivocal. Ultimately, the use of a signature that is not predictive runs the risk of discounting a truly effective therapy because the wrong cohort of patients were accrued to the substudy.

Testing of drugs specific for uncommon biomarkers is especially challenging in GBM. With a few thousand cases of GBM diagnosed annually in Germany, no patients were identified with Alk or hedgehog activation, which resulted in closure of the alectinib and vismodegib substudies, respectively. Similar challenges in identifying recurrent GBM patients in the United States with high mutational burden resulted in premature closure of the Alliance A071702 trial evaluating immune checkpoint inhibitor therapy. Thus, beyond identifying accurate predictive biomarkers, the absolute incidence of uncommon biomarkers in rare diseases like GBM must be carefully considered when developing specific substudies.

The selection of individual drugs for clinical testing is another major challenge. Significant investment is required to identify relevant molecular therapeutic targets, develop corresponding predictive biomarkers, and identify specific drugs. For each drug, pharma partners are continually assessing the available resources being dedicated for development in a rare tumor indication vs. more common indications that more closely align with their business development plan. Highlighting this dynamic, when the Phase 3 MIRROS trial of idasanutlin combined with cytarabine in relapsed and refractory AML failed to demonstrate a survival benefit[3], Roche discontinued development of the drug, despite promising pre-clinical activity in MDM2-ampilfied GBM. As a result, the N^2M^2 idasanutlin substudy was closed after accruing only 9 patients, which precludes any meaningful assessment of activity. This example highlights how competing interests can affect the ability to evaluate specific drugs in an umbrella trial.

Drug distribution into the brain is another key consideration for selecting individual drugs for testing in any GBM trial. The blood brain barrier (BBB) excludes the vast majority of drugs from the normal brain[4], and all GBM have significant tumor burden that is protected by the BBB[5]. Thus, selecting drugs that can effectively penetrate into all regions of a tumor is an important criterion for an umbrella trial. The N^2M^2 study team is commended for selecting several agents with excellent brain penetration (alectinib, vismodegib, idasanutlin). However, temsirolimus and palbociclib brain distribution is limited by efflux at the BBB[6,7], and everolimus and abemaciclib, respectively, may have been better choices for these substudies. Again, reflecting competing interests, most likely the latter two drugs were pursued by the study team but ultimately could not be secured from pharma partners. Large biomolecules like atezolizumab and asunercept are highly excluded by the intact BBB[4]. While there are anecdotal reports of immune checkpoint inhibitor activity in GBM, definitive randomized clinical trials testing these high molecular weight agents have been consistently negative. To move the field forward, we need to design clinical studies that interrogate the mechanisms of these failures and to consider whether limited, heterogenous delivery of these large biomolecules is a contributing factor.

The ultimate goal of N^2M^2 and other ongoing umbrella trials in neuro-oncology is to nominate only the most promising therapeutic strategies to move forward into definitive randomized Phase 3 testing. In this context, N^2M^2 was successful in validating the mTORC1 inhibitor temsirolimus as a promising therapeutic for phospho-mTOR activated GBM. However, the mTORC1 inhibitor everolimus may have better brain distribution, which presents a conundrum as to which drug to move forward into definitive testing. Since both drugs have some limitation in brain distribution, a surgical window of opportunity trial could be employed to evaluate if either drug adequately suppresses mTORC1 activity throughout the tumor in order to nominate the best drug to move into Phase 3 testing. While the team wrestles with this question, we look forward to a follow-on N^2M^2 trial testing the next generation of promising agents for this deadly disease.

1. Wick W, Lanz L-M, Wick A, et al. Molecularly matched targeted therapies plus radiotherapy in glioblastoma: the phase 1/2a N2M2 umbrella trial. *Nat Med . 2025 Sep 5. doi: 10.1038/s41591-025-03928-9. Online ahead of print.*

2. Wick W, Gorlia T, Bady P, et al. Phase II Study of Radiotherapy and Temsirolimus versus Radiochemotherapy with Temozolomide in Patients with Newly Diagnosed Glioblastoma without MGMT Promoter Hypermethylation (EORTC 26082). *Clin. Cancer Res.* 2016; 22(19):4797-4806.

3. Konopleva MY, Röllig C, Cavenagh J, et al. Idasanutlin plus cytarabine in relapsed or refractory acute myeloid leukemia: results of the MIRROS trial. *Blood advances.* 2022; 6(14):4147-4156.

4. Alavijeh MS, Chishty M, Qaiser MZ, Palmer AM. Drug metabolism and pharmacokinetics, the blood-brain barrier, and central nervous system drug discovery. *NeuroRX.* 2005; 2(4):554-571.

5. Sarkaria JN, Hu LS, Parney IF, et al. Is the blood-brain barrier really disrupted in all glioblastomas? A critical assessment of existing clinical data. *Neuro Oncol.* 2018; 20(2):184-191.

6. Lin F, de Gooijer MC, Hanekamp D, et al. PI3K-mTOR Pathway Inhibition Exhibits Efficacy Against High-grade Glioma in Clinically Relevant Mouse Models. *Clin. Cancer Res.* 2017; 23(5):1286-1298.

7. de Gooijer MC, Zhang P, Thota N, et al. P-glycoprotein and breast cancer resistance protein restrict the brain penetration of the CDK4/6 inhibitor palbociclib. *Invest. New Drugs.* 2015; 33(5):1012-1019.