Preview

Brain tumors on slice: A novel platform for personalized therapeutic screening

Yusha Sun,[1] Xin Wang,[2] Guo-li Ming,[2] and Hongjun Song[2,*]

[1]Neuroscience Graduate Group, Perelman School of Medicine, University of Pennsylvania, Philadelphia, PA 19104, USA
[2]Department of Neuroscience and Mahoney Institute for Neurosciences, Perelman School of Medicine, University of Pennsylvania, Philadelphia, PA 19104, USA
*Correspondence: shongjun@pennmedicine.upenn.edu
https://doi.org/10.1016/j.xcrm.2023.101059

Mann and Zhang et al. developed a robust *ex vivo* slice culture platform consisting of resected patient high- and low-grade glioma tissue engrafted onto rat organotypic brain slices, and interrogated tumor responses to clinically relevant therapeutics with a novel treatment-response algorithm.

Primary brain tumors remain some of the most difficult-to-treat cancers with little improvement in patient outcomes over the last few decades, despite preclinical development of numerous targeted small-molecules, biologic agents, and immunotherapies.[1] This limited success may be partially attributed to patient heterogeneity in treatment responses. A precision medicine approach is needed to determine the ideal course of treatment given properties of the patient's own tumor.

To model treatment responses in a more personalized manner, systems that more faithfully recapitulate patient intra-tumoral heterogeneity have been developed, including patient-derived organoids[2] or explants,[3] as opposed to tumor cell lines with *in vitro* clonal selection and genetic drift over long-term culture. These patient tissue-derived models can retain genetic, transcriptomic, and histopathological features of their parent tumors.[2,3] However, these platforms tend to gradually lose elements of the tumor microenvironment over time. One simple option is to incorporate a traditional neural substrate, such as organotypic brain slice culture (OBSC), which allows for the engraftment of cancer cells onto murine brain sections *ex vivo*.[4–6] Previous studies with OBSC have largely employed cell line or glioblastoma stem cell culture, but not primary tumor cells.

Mann and Zhang et al.[4] seeded primary patient glioma tissue from a spectrum of patient ages and tumor types onto an OBSC platform of coronal brain slices generated from 8-day-old rats. The method was first validated with a panel of tumor cell lines, including glioblastoma (GBM) and diffuse intrinsic pontine glioma, and invasion patterns on OBSCs correlated well with the patterns observed for these lines in orthotopic xenografts. Interestingly, low-passage cell lines that did not grow well in xenografts could engraft on OBSC. In all, 11 tumors ranging from adult, pediatric, and primary to metastatic were cultured in this manner, and, notably, low-grade tumors displayed consistent survival on OBSC, but not with standard *in vitro* culture methods. Surgical tissue could also be cryopreserved and thawed before engrafting onto OBSC without compromising tumor survival or response to therapeutics, adding an additional layer of flexibility to the system.

The authors measured treatment responses of either cell lines or patient tissue on OBSCs to a set of drug therapeutics, radiation therapy, and standard-of-care treatment (Figure 1). Prior to seeding, the authors employed a dissociation protocol to generate a near single-cell suspension and infected them with a lentivirus carrying a fluorescent marker. By bioluminescent imaging of fluorescently labeled tumor cells, dose-response curves were generated for each cell line and patient tissue. The effect of the treatment on normal rat brain tissue was also determined via a propidium iodide-based assay. The authors developed a novel algorithm that includes 11 distinct weighted parameters, such as area under the dose-response curve, EC_{50} slope, and concentrations at various points along the dose-response curve, to provide a normalized drug-sensitivity score (DSS) for each combination of tumor and treatment. This approach allows for comparison of the relative efficacy of different treatments side-by-side for each patient sample. Although preliminary, the authors provided an example that the DSS confirmed a lack of response to standard of care treatment (temozolomide and radiation) observed in a GBM patient despite MGMT methylation, suggesting the possibility of prediction of clinical responses.

Overall, this study provides an important advancement in the development of a patient-specific hybrid tumor-brain slice platform to enhance survival and integration of otherwise difficult-to-grow primary tissues. Lower-grade glioma can be more difficult to maintain in organoids compared to the more aggressive high-grade glioma,[2] and this study adds to other protocols that support lower-grade glioma *ex vivo* establishment, such as reduced oxygen tension.[7] Given the ability to engraft multiple types of brain tumors and the presence of neurons and diverse glia, this model opens the door for future studies of tumor biology, such as bidirectional signaling between central nervous system (CNS) tumors of various grades and neurons.[8] An additional strength of this platform is the ability to model how microenvironmental context modifies tumor response to treatment. For example, the authors found that caseinolytic protease inhibitors induced more complete tumor killing on OBSCs compared to standard culture. The DDS approach is also novel, going beyond the traditional EC_{50} on the dose-response curve.
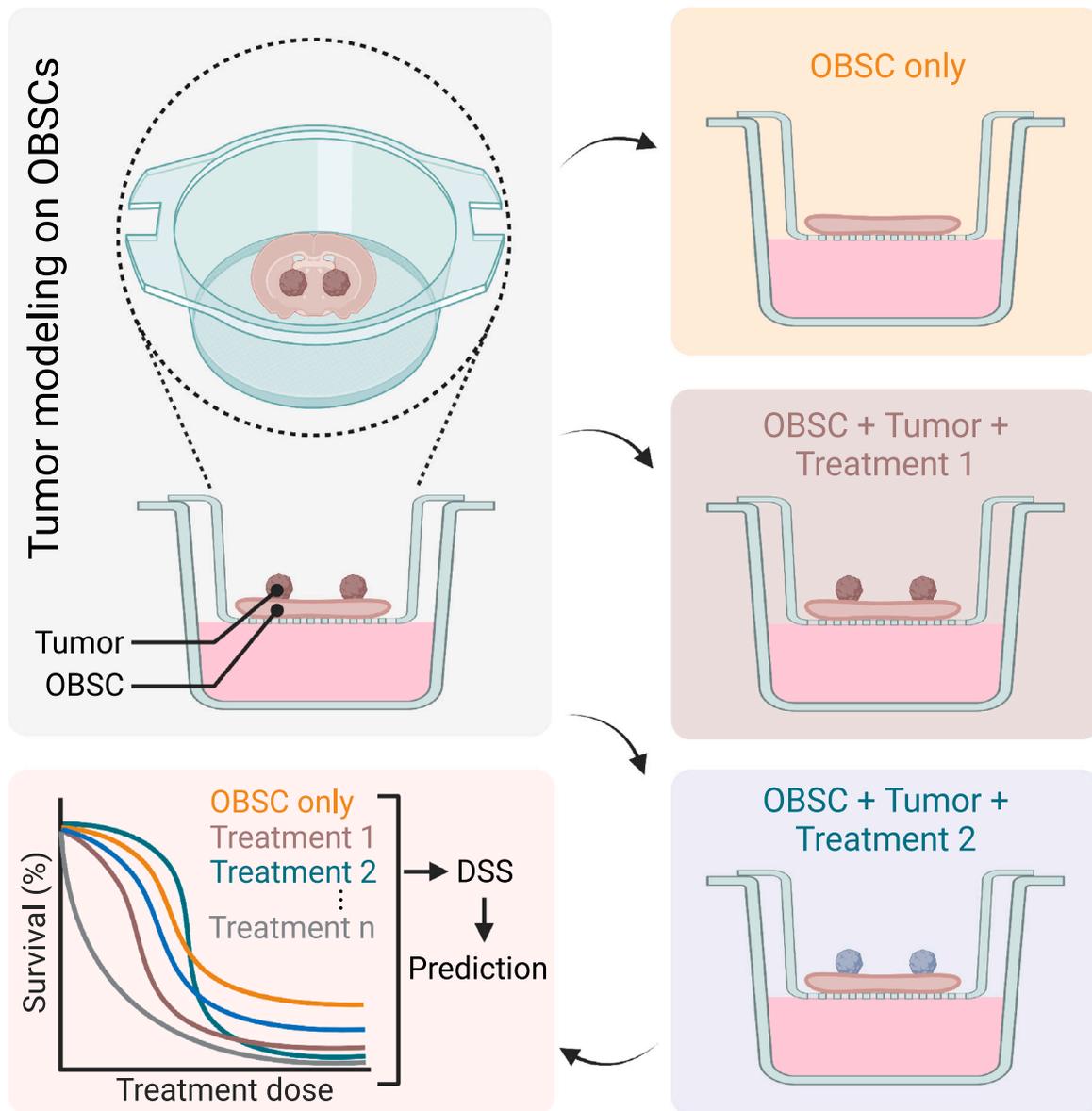
Figure 1. Tumor modeling on organotypic rat brain slice culture (OBSC) allows for the exploration of individualized treatment sensitivities
Responses of primary tumor tissue growth to several treatments (1, 2, …., n) at various doses are integrated with each treatment's impact on OBSC alone to generate the drug-sensitivity score (DSS). DSS across various treatments may predict relative patient clinical response, with the eventual goal of personalized therapeutic regimens.

One limitation of the current OBSC platform for tumor modeling is its non-human substrate that utilizes rat brain slices. Recently developed human stem cell-derived sliced neocortical organoids[9] could be harnessed to study tumor responses to treatments in an all-human system. The platform also does not yet model several known modulators of patient response to targeted therapeutics, including blood-brain barrier penetration or functional vasculature. Finally, more

experimentation will be required to validate the DSS in predicting patient responses. Future studies could focus on evaluating DSS with a larger cohort of brain tumor patients across a diverse array of treatments, particularly since the predictive ability of CNS patient-derived organoids still lags behind that of other cancers, such as colorectal cancer.[10]

All in all, the development of this glioma-on-slice platform not only retains intra-tumor heterogeneity but also pre-

sents opportunities to translate CNS therapeutics to the clinic in a more personalized and representative manner.

DECLARATION OF INTERESTS

The authors declare no competing interests.

REFERENCES

1. Aldape, K., Brindle, K.M., Chesler, L., Chopra, R., Gajjar, A., Gilbert, M.R., Gottardo, N., Gutmann, D.H., Hargrave, D., Holland, E.C., et al.

(2019). Challenges to curing primary brain tumours. Nat. Rev. Clin. Oncol. *16*, 509–520.

2. Jacob, F., Salinas, R.D., Zhang, D.Y., Nguyen, P.T.T., Schnoll, J.G., Wong, S.Z.H., Thokala, R., Sheikh, S., Saxena, D., Prokop, S., et al. (2020). A patient-derived glioblastoma organoid model and Biobank recapitulates inter- and intra-tumoral heterogeneity. Cell *180*, 188–204.

3. LeBlanc, V.G., Trinh, D.L., Aslanpour, S., Hughes, M., Livingstone, D., Jin, D., Ahn, B.Y., Blough, M.D., Cairncross, J.G., Chan, J.A., et al. (2022). Single-cell landscapes of primary glioblastomas and matched explants and cell lines show variable retention of inter- and intratumor heterogeneity. Cancer Cell *40*, 379–392.

4. Mann, B., Zhang, X., Bell, N., Adefolaju, A., Thang, M., Dasari, R., Kanchi, K., Valdivia, A., Yang, Y., Buckley, A., et al. (2023). A Living ex vivo platform for functional, personalized brain cancer Diagnosis. Cell Rep. Med. *4* https://doi.org/10.1016/j.xcrm.2023.101042.

5. Satterlee, A.B., Dunn, D.E., Lo, D.C., Khagi, S., and Hingtgen, S. (2019). Tumoricidal stem cell therapy enables killing in novel hybrid models of heterogeneous glioblastoma. Neuro Oncol. *21*, 1552–1564.

6. Marques-Torrejon, M.A., Gangoso, E., and Pollard, S.M. (2018). Modelling glioblastoma tumour-host cell interactions using adult brain organotypic slice co-culture. Dis. Model. Mech. *11*, dmm031435.

7. Abdullah, K.G., Bird, C.E., Buehler, J.D., Gattie, L.C., Savani, M.R., Sternisha, A.C., Xiao, Y., Levitt, M.M., Hicks, W.H., Li, W., et al. (2022). Establishment of patient-derived orga-noid models of lower-grade glioma. Neuro Oncol. *24*, 612–623.

8. Keough, M.B., and Monje, M. (2022). Neural signaling in cancer. Annu. Rev. Neurosci. *45*, 199–221.

9. Qian, X., Su, Y., Adam, C.D., Deutschmann, A.U., Pather, S.R., Goldberg, E.M., Su, K., Li, S., Lu, L., Jacob, F., et al. (2020). Sliced human Cortical organoids for modeling distinct Cortical layer formation. Cell Stem Cell *26*, 766–781.

10. Wensink, G.E., Elias, S.G., Mullenders, J., Koopman, M., Boj, S.F., Kranenburg, O.W., and Roodhart, J.M.L. (2021). Patient-derived organoids as a predictive biomarker for treatment response in cancer patients. Npj Precis. Oncol. *5*, 30.