A smarter route to the brain: Preserving the BBB in preclinical brain tumor models

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Introduction

Brain tumors and brain metastases remain among the most difficult cancers to treat, with many promising therapies failing in clinical trials, often due to inadequate modeling of the blood-brain barrier (BBB) in preclinical studies. Standard intra-cranial orthotopic models, while widely used, disrupt BBB integrity, limiting their ability to predict drug delivery and therapeutic response. Alternative strategies, such as intra-venous and intra-cardiac tumor cell inoculation, preserve the BBB but frequently result in widespread systemic metastases and premature study termination.

To address these limitations, we developed an intra-carotid implantation strategy that directs tumor cells specifically to the brain, achieving localized growth while maintaining BBB integrity. Tumor progression and growth kinetics were evaluated longitudinally using magnetic resonance imaging (MRI) and bioluminescence imaging (BLI), which confirmed consistent brain-targeted colonization and allowed precise monitoring of tumor burden over time.

This approach provides a more physiologically relevant model for both primary brain tumors, such as glioblastoma, as well as brain metastases from systemic cancers. By preserving BBB function and enabling reliable, longitudinal assessment of tumor growth, the intra-carotid model offers a robust platform for preclinical evaluation of BBB-penetrant therapeutics and may improve the predictive power of drug development in neuroscience and oncology.

Materials & Methods

MDA-MB-231_luc is a highly aggressive, invasive and poorly differentiated triple-negative breast cancer (TNBC) cell line as it lacks estrogen receptor (ER) and progesterone receptor (PR) expression, as well as HER2 (human epidermal growth factor receptor 2) amplification.

Mice were anaesthetized with IP injection of ketamine/xylazine cocktail. After surgical site prep, a small incision was made in the skin, and dissection of the surrounding tissue was performed to reach the CCA. After preparation of the CCA, the MDA-MB-231_luc cells were then injected into the CCA. The mouse was sutured, placed into a heating pad until fully conscious, and then returned to its cage.

Starting on Day 5 post inoculation, mice were assessed at multiple timepoints for the presence of tumor cells in the brain via IVIS bioluminescent imaging (BLI).

Starting on Day 7 post inoculation, mice were assessed at multiple timepoints for the integrity of the BBB and presence of tumor in the brain via magnetic resonance imaging (MRI) with a 7 tesla Bruker Biospec 70/20as small animal imaging system.

Mice were terminated based on signs of clinical disease and were on study for median of 30 days.

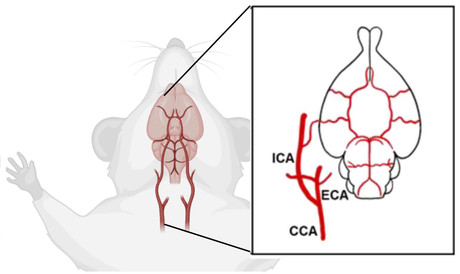
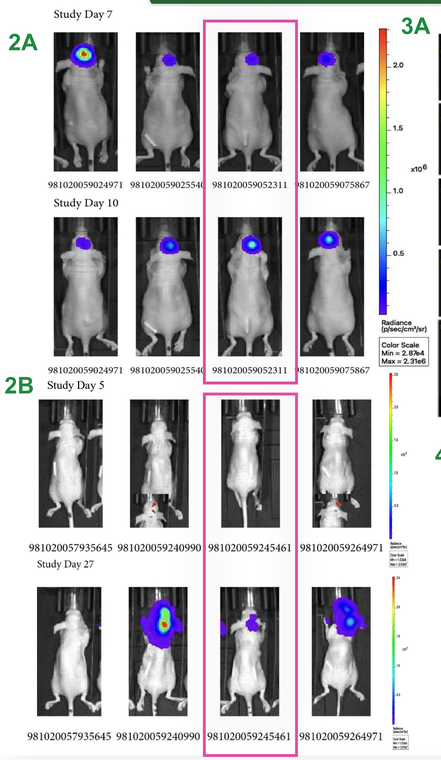
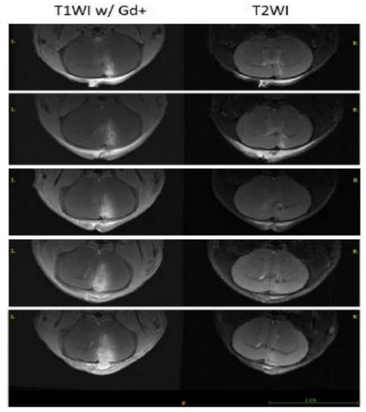
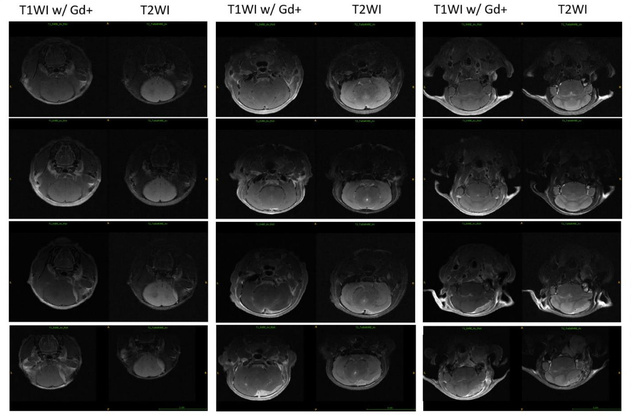


Figure 1: Representation of the common carotid artery (CCA), located laterally to the trachea, and ramified into the external carotid artery (ECA) and the internal carotid artery (ICA).

In Vivo Performance of Intra-Carotid Brain Tumor Models







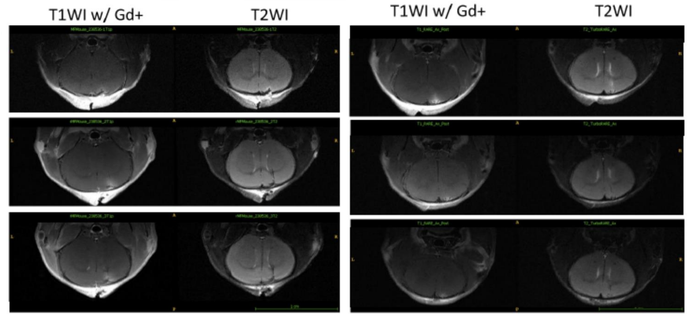


Figure 2A: Individual Animal In vivo Bioluminescence (Total Flux [p/s]) for intra-cranial MDA-MB-231_luc

Figure 2B: Individual Animal In vivo Bioluminescence (Total Flux [p/s]) for intra-carotid MDA-MB-231_luc

Figure 3A: MRI images* for Mouse #052311 (MDA-MB-231 intra-cranial) taken on Day 0 (4hr post inoculation), 2, 4, 8, 11.

Figure 3B: MRI images* for Mouse #2454561 (MDA-MB-231 intra-carotid) taken on Day 7, 14, 21, and 23.

Figure 4: Sham intra-cranial inoculation mice MRI images* taken on Day 2, 4, and 7 post inoculation. **NOTE**: The injection site's long T2 signal was significantly reduced on Day 4 and not visible on Day 7, which indicates the recovery of damaged tissue. However, the injection site and surrounding area was significantly enhanced in post-contrast T1WI on all three scan days, which indicates the damage of brain blood barrier related to the injection.

*MRI images were acquired axially in 32 0.5 mm-thick axial slices covering the whole brain with an in-plane spatial resolution of 0.078 mm × 0.078 mm. Contrast enhancement was conducted with a bolus injection of 0.01 mL gadobutrol IV.

Conclusion

The intra-carotid implantation model enables localized brain tumor growth while preserving BBB integrity, overcoming limitations of standard intracranial, intravenous, and intracardiac models. Longitudinal MRI and BLI imaging confirmed consistent brain-targeted tumor colonization and reproducible growth kinetics. This approach provides a translationally relevant platform for evaluating BBB-penetrant therapeutics in both primary brain tumors and metastatic lesions, improving the predictive power of preclinical neuro-oncology studies.

References

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