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LEVERAGING PARENCHYMAL ANTIGEN PRESENTATION BY MICROGLIA FOR THE CONTROL OF BREAST CANCER BRAIN METASTASES

Aaron J Longworth*, Timothy P McMullen, Isam Adam, Tatyana Lev, Hannah Savage, Paige Halas, Devon A Lawson. *University of California, Irvine School of Medicine, Irvine, CA, USA*

Background During their lifetime, 1 in 8 women are expected to be diagnosed with breast cancer.¹ 10 to 15% of metastatic breast cancer patients will subsequently develop brain metastasis (BrM), which lowers their prognosis to a dismal 7.9 months.² Contrary to prevailing literature,^{3–5} we have observed that microglia shift to a pro-inflammatory phenotype and upregulate antigen presentation (AP) machinery in response to the breast cancer BrM, however this activity is often insufficient to control BrM. In these studies, we evaluate the potential of brain parenchymal CD40 agonism to drive a more robust antimetastatic immune response and evaluate peripheral delivery methods to control intracranial lesions.

Methods Employing single-cell RNA sequencing on CD45^{hi-int} from WT and T cell deficient (RAG1^{-/-}) mice at 4- and 10-days post-transplantation of E0771 brain metastases, we identified a marked upregulation of pro-inflammatory programs and antigen presentation machinery amongst the microglia population in the presence of T cell infiltration. We performed intracranial administration of a CD40 agonistic antibody against an isotype control to albino C57BL/6J mice harboring intracranial E0771 breast cancer tumors expressing enhanced GFP and firefly luciferase to monitor tumor progression. To demonstrate the AP activity is microglia-specific, we repeated the previous experiment following liposomal clodronate depletion of peripheral phagocytes, and confirmed results in B2M deficient mice that lack AP by class I major histocompatibility complexes (MHC). Lastly, we are assessing peripheral methods for delivery of CD40 agonists to the central nervous system (CNS) via injections into the primary mammary tumor to evaluate the effect on tumor growth and host mortality.

Results Despite reports in literature that microglia are tumor-promoting, we demonstrate that microglia undergo an appropriate shift to an anti-metastatic, pro-inflammatory phenotype upon initiation of breast cancer BrM. We observe that local administration of agonistic aCD40 upregulates AP machinery in microglia, including class I and II MHC complexes and costimulatory molecules such as CD80 and CD86, and that this results in reduced mortality of BrM-bearing mice. We confirmed that microglia are sufficient to recruit and locally activate T cell infiltrates, and that this activity is enhanced by CD40 agonism in the CNS.

Conclusions Intracranial administration of agonistic aCD40 enhances local antigen presentation in the CNS and drives a more robust anti-BrM T cell response. This activity can extend viability of murine hosts up to 40%. Investigation into peripheral delivery methods for translational application may provide treatment avenues to provide for a critical clinical unmet need.

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Ethics Approval All animal studies were performed ethically and with approval from the institutional animal care and use committee (IACUC) of the University of California, Irvine under animal use protocol AUP-22–033.

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