NEURO-ONCOLOGY ADVANCES

ABSTRACT CODES/CATEGORIES:
- BSCI - Basic Science
- LPTO - Leptomeningeal Disease
- TRX - Clinical Trials
- THER - Medical Therapy (Chemotherapy, Targeted Therapy/Immuno-therapy)
- MLTI - Multimorbidity
- OTHER - Other
- RADI - Radiation
- SURG - Surgery

BASIC SCIENCE

BSCI-01. ACTIVATION OF C-MET/β1-INTEGRIN COMPLEX RESULTS IN INCREASE OF MENSENCHYMAL GENE EXPRESSION AND STEM CELL POPULATION IN METASTATIC BREAST CANCER TO THE BRAIN AND SPINE
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INTRODUCTION: C-met and β1-integrins play a central role in nearly all stages of cancer metastasis. They bind at the cell surface, driving ligand independent co-activation of downstream pathways. Greater complexity is seen in metastatic tumors vs. its primary tumor counterparts in patients. The molecular, cellular, and clinical effects of complex formation in metastatic breast cancer are investigated. METHODS: Utilizing variations of the MDA-231 breast cancer cell lines (standard MDA-231, inducible complex formation MDA-231, brain seeking MDA 231, lung seeking MDA 231, and bone seeking MDA-231), in vitro and in vivo studies were performed. Clinical correlates from patient samples were studied.

RESULTS: Induction of c-Met/β1 complex promotes breast cancer invasion (p<0.001), migration (p<0.05), circulation intravasation (p<0.01), and adhesion (p<0.01). These effects may be driven by the increased mesenchymal character (p<0.05) and larger stem cell population (p<0.001) caused by inducing c-Met/β1 complex formation. OS2966 (a therapeutic β1 integrin blocking antibody) decreases invasion (p<0.05), intravasation (p<0.05), and mesenchymal form factor (p<0.001) and gene expression (p<0.001) in MDA-MB-231 cells. Brain- and bone-seeking breast cancer cells have higher c-Met/β1 complex than parental controls and preferentially adhere to tissue-specific matrix (p<0.01). In intracardiac metastasis models, complex formation resulted in significantly higher metastatic burden and shorter survival (p<0.001). qPCR data suggests that complex formation may drive tumor colonization of cancer cells (micrometastasis) rather than tumor growth. Patient brain and bone metastases demonstrated high β1/C-Met levels. CONCLUSIONS: The c-Met/β1 complex drives intravasation and extravasation of breast cancer cells from the circulation. Preferential affinity for tissue-specific matrix enables the c-Met/β1 complex to drive formation of breast cancer metastases to the brain and bone. Pharmacological and genetic targeting of the complex with agents may provide therapeutic approaches to prevent metastasis, particularly to the brain and bone.

BSCI-02. T GLI1 IS A NOVEL, ACTIONABLE TARGET FOR THE TREATMENT OF BREAST CANCER BRAIN METASTASES
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Despite improvements in early detection and intervention, breast cancer remains the second leading cause of cancer-related death in women and the second most common cancer to metastasize to the brain. Current standard of care options for breast cancer brain metastases (BCBM) include stereotactic radiosurgery, whole-brain radiotherapy, and surgical resection. Local of care options for breast cancer brain metastases (BCBM) include stereo...
seeking 231-BR subclone of MDA-MB-231 TNBC cells, which harbors a loss of Pten compared to parental cells. Breast metastases were generated in nude mice by intracardiac injection of 1.75×10⁶ 231-BR cells engineered for expression of β-galactosidase confirmed by IVIS. Mice were killed weekly after injection. Metastatic brain metastases were treated by tail vein injection of control (PBS, n=7) or DX1 (20 mg/kg, n=7) 3x/week for 4 weeks. Mice were observed for behavior and weights, and brain radiance efficiency was monitored by weekly IVIS to track tumor growth. PAT-DX1 significantly suppressed growth of brain metastases based on absolute and relative radiance efficiencies in the brain, increased the median survival of the mice from 38 to 52 days (P<0.02), and was well tolerated. These results provide proof of concept for use of a re-engineered autoantibody against breast metastases.

**BSCI-05. HOW MICROGLIA, BRAIN RESIDENT MYELOID CELLS, RESPOND TO BREAST CANCER METASTASIS INTO THE BRAIN?**

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Brain metastasis from different cancers, including lung, breast, melanoma, colorectal or renal cell carcinoma is relatively common and its frequency increases with a prolonged survival of cancer patients. New anti-cancer therapies frequently fail to reduce metastatic burden. While the important role of tumor-associated macrophages as pro-tumorigenic cells facilitating tissue remodeling, invasion and metastasis is well documented, much less is known about the immune microenvironment of brain metastases and potential mechanisms that drive interactions of immune cells with brain microvascular and meningeal microglia. Triple-negative breast cancer metastases to the brain were discovered in 46% of patients. We evaluated the abundance and morphology of microglia on sections from breast cancer metastases using immunohistochemistry. We found that microglia cells are activated, surround the breast cancer cells, and do not infiltrate the solid tumor. Searching for a potential attractant of microglia, we discovered osteopontin levels in six human breast cancer cell lines and found upregulation of osteopontin in transformed cells, with the highest level in the triple-negative MDA-MB-231 cells. MDA-MB-231 cells activated primary murine microglia cultures when co-cultured. Invasion of MDA-MB-231 cells in co-cultures with murine immortalized BV2 microglial cells and human SV40 immortalized microglia was increased, as demonstrated by Matrigel Invasion Assay. Using immunofluorescence we found osteopontin in breast cancer cells in human breast cancer metastases. Moreover, we found that minocycline, a clinically used antibiotic, reduces the osteopontin production in human breast cancer cells and the most sensitive cells were MDA-MB-231 cells. Our study shows that metastatic cancer cells may employ osteopontin to facilitate extravasation and colonization of brain parenchyma. We postulate that osteopontin mediates interactions between microglia and metastatic cancer cells and minocycline may interfere with those interactions.

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**BSCI-06. FREQUENCY OF BRAIN METASTASIS FROM BREAST AND LUNG CANCER IN THE UNITED STATES – A POPULATION-BASED ASSESSMENT**

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**BACKGROUND:** Brain metastases (BM) are the most common central nervous system tumor in the United States and occur with increasing frequency due to improved screening and therapies leading to improved survival. Current estimates of frequency of BM vary significantly by cancer site and are typically not population-based. Population-based estimates of incidence have recently become possible due to collection of data on BM identified at diagnosis (“synchronous” BM, SBM). BM may occur at any point after cancer diagnosis. We report our recent population-based estimates of BM incidence and period BM (PBMT) from breast (BC) and lung cancer (LC). METHODS: Data from Surveillance, Epidemiology, and End Results (SEER) 2010–2016 diagnosed cases from SEER-Medicare (2008–2012 diagnoses for individuals 65+) were used to estimate SBM and linked data from SEER-Medicare (2008–2012 diagnoses for individuals 65+, with 2007–2014 claims) were used to estimate PBMT, for BM and LC overall and by BC and LC subtypes. RESULTS: Within the SEER data, 10.9% of LC cases present small cell LC (SCLC); 15.8% in non-small cell LC [NSCLC]); 0.4% of BC cases presented with SBM, 0.7% in triple negative (TNBC), 0.8% for HER2+, and 0.2% for ER+PR–/HER2–. Within the SEER-Medicare data, 13.3% of LC overall had LBMC weight 4.1% for SCLC, and 15.3% for NSCLC, with 4.2% in triple negative (TNBC), 3.1% for HER2+, and 1.1% for ER+PR+HER2. CONCLUSION: Frequency of synchronous and period BM varies by originating site as well as subtype. The new BM variable in SEER allows for estimation of this important statistic, while the SEER-Medicare linked data allows for estimation of PBMT, both on a population-level for the US population. These estimates are useful to clinical practice and critical for estimating morbidity and mortality due to BM.

**BSCI-07. BONE MARROW T CELL SEQUESTRATION IN THE SETTING OF BRAIN METASTASES**

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**INTRODUCTION:** Brain metastases remain one of the most dreaded consequences of late stage cancer, yet their incidence has risen as survival from primary cancers has improved. We have recently reported that tumors harbored within the brain, specifically, sequester T-cells within the bone marrow and subcutaneous fat; immune evasion. Sequestration of tumor-imposed loss of S1P1 receptor from the T-cell surface. Stabilization of the receptor on T-cells frees T-cells from sequestration and licenses T-cell activating therapies for intracranial tumors. While this phenomenon was initially observed in glioblastoma, its role in promoting immune evasion in brain metastases remains less clear. METHODS: Blood, bone marrow, and tumors were collected from mice bearing intracranial tumors commonly metastatic to the brain, including lung carcinoma (LLC), melanoma (B16F10), or breast carcinoma (E0771) and analyzed by flow cytometry. T-cell S1P1 levels, as well as total T-cell counts were assessed in each compartment. Correlation analyses were conducted between T-cell counts and S1P1 levels on T-cells in the bone marrow across intracranial and subcutaneous tumor models. RESULTS: T-cell lymphopenia and accompanying accumulation of T-cells in the bone marrow were observed in the murine models of lung carcinoma, melanoma, and breast carcinoma but only when these tumor lines were implanted intracranially. Sequestered T-cells in tumor-bearing mice showed decreased surface S1P1 levels in a manner correlating with their sequestration. CONCLUSION: S1P1-mediated bone marrow T-cell sequestration is a novel mode of cancer-induced T-cell dysfunction in intracranial tumors. Preventing receptor internalization abrogates T-cell sequestration and licenses T-cell activating therapies in glioblastoma. Sequestration is now observed in models of brain metastases. Pharmacologic strategies to stabilize S1P1, reverse sequestration, and restore circulating T-cell numbers are anticipated to improve immunotherapeutic efficacy for brain metastases.

**BSCI-09. MECHANISMS OF ENHANCED DRUG DELIVERY IN BRAIN METASTASES WITH FOCUSED ULTRASOUND-INDUCED BLOOD-TUMOR BARRIER DISRUPTION**

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Blood-brain-tumor barriers (BBB and BTB) and interstitial transport may constitute major obstacles to the transport of therapeutics in brain tumors. In this study, we examined the impact of focused ultrasound (FUS) in combination with microbubbles on the transport of two relevant chemotherapy-based anticancer agents in HER2-positive breast cancer brain metastases at cellular resolution: the non-targeted chemotherapeutic doxorubicin and the antibody-drug conjugate ado-trastuzumab emtansine (T-DM1). Using an orthotopic xenograft model of HER2-positive breast cancer brain metastasis and quantitative microscopy we demonstrate multiple increases in the extravasation of both agents (7-fold and 2-fold for doxorubicin and T-DM1, respectively) and we provide evidence of increased drug penetration (>100μm vs. < 20μm and 42±7μm vs. 12±4μm for doxorubicin and T-DM1, respectively) after application of FUS as compared to control (PBS). Integration of experimental data with physiologically based pharmacokinetic (PBPK) modeling of drug transport reveals that FUS in combination with microbubbles alleviates vascular barriers and enhances interstitial convective transport via increase in hydraulic conductivity. Combination of experimental data and PBPK modeling suggests that FUS in combination with microbubbles increases the endothelial cell transmembrane transport and uptake. PBPK modelling indicates selective increase in transvascular transport of the non-targeted small chemotherapeutic doxorubicin through small vessel-wall pores size with a narrow range (Diameter: 10-50nm). Our work provides a quantitative framework for the optimization of FUS-drug combinations to maximize intratumoral drug delivery and facilitate the development of novel therapeutic strategies against brain metastases.

**BSCI-10. NEUROLOGICAL DYSFUNCTION CAUSED BY BRAIN TUMOR-GENERATED SOLID STRESS IS REVERSED BY LITHIUM**

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