### BSCI-26. COMPARATIVE METHYLATION PROFILING OF EGFR MUTANT LUNG ADENOCARCINOMA AND PAIRED BRAIN METASTASIS

Yasin Mamatjan[1], Jeffrey Zuccato[1], Shirin Karimi[1], Michael Cabanero[1], Jessica Weiss[1], Hadas Sorotsky[1], Ming Tsao[1], Frances Shepherd[1], Kenneth Aldape[2] and Gelareh Zadeh[1]; [1]Princess Margaret Cancer Centre, Toronto, ON, Canada, [2]Laboratory of Pathology, National Cancer Institute, Bethesda, MD, USA

**BACKGROUND:** Adenocarcinomas (ADC) are the most common lung tumours and EGFR-mutant lesions have a higher risk of brain metastasis development, which confers poorer survival. Genetic and epigenetic signatures of brain metastasis have not been comprehensively identified. The aim of this study is to compare the methylome of EGFR-mutant primary lung tumor and matched brain metastasis to identify mechanisms of brain metastasis and new treatment targets. METHOD: Seven matched primary to brain metastasis tumours were profiled using the Illumina Infinium MethylationEPIC BeadChip array. Hierarchical clustering and principal component analyses (PCA) were performed using most variable CpG sites. Supervised analyses were performed between lung and brain tumour samples. Copy number variation (CNV) plots identified alterations between pairs along with Leukocytes unmethylation for purity (LUMP) score and prediction lymphocyte proportion analyses to measure immune infiltration. RESULTS: Unsupervised clustering showed that the fourteen tumours clustered according to patient with similar methylation profiles between each of seven matched pairs. On the supervised analysis using 83K significant CpG sites, the fourteen samples clustered into two groups based on tumour site being lung or brain. Of these 83K CpG sites, 2.4K were either hypermethylated or hypomethylated in all lung samples. One quarter of these 2.4K CpG sites were located in promoter regions. CNV analyses showed losses of FGFR1, C19MC, CDKN2A, PTCH1, and MYCN genes with higher deep deletions in brain versus lung primary samples. Immune infiltration measures were similar between lung and brain metastasis pairs (LUMP-score=0.64) consistent with high immune cell infiltration. CONCLUSION: In this EGFR-mutant lung adenocarcinomas and matched brain metastases, differentially methylated CpG sites and CNV alterations are identified that distinguish lung from brain samples. Further work with additional matched samples may further elucidate signatures specific to brain metastasis and aid in our understanding of the mechanisms of brain metastasis.

### BSCI-27. MELATONIN REDUCES MALIGNANCY OF BREAST CANCER BRAIN METASTATIC CELLS

Beatriz Fernandez Gil, Katherine Rodriguez, Paula Schiapparelli, Carla Vazquez Ramos, Germaine Escames, Alfredo Quinones-Hinojosa, and Rachel Sarabia-Estrada; Mayo Clinic, Jacksonville, FL, USA

Around fifteen to thirty percent of stage IV breast cancer metastasizes to the brain, severely decreasing the quality of life of these patients by causing neurological decline and eventually death. In metastatic cancers there is a small subset of cells in the primary tumor bulk called Metastatic Tumor Initiating Cells (MTICs) which are able to escape and produce a niche establishment at distal sites where they can quickly become resistant to surgery and radiation. Melatonin has shown an inhibitory role in the viability and invasiveness of breast cancer and in modulating the expression of proteins related to Breast Cancer Stem Cells (BCSCs). These findings suggest its potential anti-metastatic role in different breast cancer cell lines. In this study we aimed to evaluate the effects of melatonin treatment *in vitro* for breast cancer brain metastasis. The cell line MDA-BT was originally obtained from MDA-MB-231, passed through the rat's heart and then isolated once engrafted as a tumor in the brain. After a dose response assay, cells were treated with melatonin at doses of 1500 and 3000 µM for 48hrs. Clonogenic assay, MTT, as well as a stem cell signature through RT-qPCR, including CD44, CD24 and ALDH1 markers, were performed to evaluate the malignancy of the MTICs. The results showed that melatonin at high doses impacts morphology, declines viability, reduces colony formation ability, and decreases stemness in MDA-BT cells. Therefore, our findings highlight melatonin as a relevant therapeutic candidate to target breast cancer brain metastases.

## LEPTOMENINGEAL DISEASE

### LPTO-01. LEPTOMENINGEAL METASTASIS FROM OVARIAN CARCINOMA TREATED WITH SYSTEMIC HIGH-DOSE METHOTREXATE

Elizabeth Coffee, Mina Lobbous and L. Burt Nabors; University of Alabama at Birmingham, Birmingham, AL, USA

**BACKGROUND:** Central nervous system metastasis in ovarian cancer is reported in approximately 1% of cases and typically localizes to the parenchyma. Leptomeningeal disease (LMD) is exceedingly rare. Prognosis is extremely poor with median survival of 60 days. Given the rarity of LMD in ovarian cancers, there is no consensus on the optimal treatment approach. High-dose systemic methotrexate (HD-MTX) has been studied in LMD. CASE REPORT: A 41-year-old Caucasian woman with a history of BRCA-positive stage IIIC ovarian carcinoma who was treated with cytoreduction and multiple rounds of intravenous and intraperitoneal chemotherapy presented to the emergency room with progressive headaches, vomiting and neck pain. Contrasted MRI of the brain showed nodular enhancement along the cerebellar folia, concerning for leptomeningeal disease. Cerebrospinal fluid cytology analysis was positive for malignancy and she was diagnosed with leptomeningeal metastasis 2.5 years after her diagnosis with ovarian carcinoma. Induction therapy with systemic HD-MTX was given every other week and complete response was achieved after two induction cycles. Serum CA-125 level decreased from 565.7 U/mL (prior to treatment) to 50.4 U/ML after the sixth induction cycle and she was transitioned to maintenance HD-MTX. Approximately 6 months after initiation of HD-MTX, contrasted MRI demonstrated LMD recurrence with corresponding rise in CA-125 to 395 U/mL. Whole body CT imaging did not show local disease recurrence. Temozolomide was added to the HD-MTX regimen and craniospinal radiation therapy was planned but her neurologic condition continued to decline. She died 8 months after her diagnosis of LMD. CONCLUSION: LMD has a dismal prognosis. HD-MTX may prolong the overall survival in patients with LMD secondary to ovarian carcinoma. In these patients, serum CA-125 level may serve as a biomarker for monitoring CNS disease burden. Due to the rarity of LMD, multi-institutional prospective studies are needed to explore novel therapeutics in LMD.

### LPTO-02. INTRATHECAL (IT) TRASTUZUMAB (T) FOR THE TREATMENT OF LEPTOMENINGEAL DISEASE (LM) IN PATIENTS (PTS) WITH HUMAN EPIDERMAL RECEPTOR-2 POSITIVE (HER2+) CANCER: A MULTICENTER PHASE 1/2 STUDY

Priya Kumthekar[1], Andrew B Lassman[2], Nancy Lin[3], Sean Grimm[4], William Gradishar[4], Elena Pentsova[5], Suriya Jeyapalan[6], Morris Groves[7], Melissa Melisko[8] and Jeffrey Raizer[1]; [1]Northwestern University, Chicago, IL, USA, [2]Columbia University, New York, NY, USA, [3]Dana-Farber Cancer Institute, Boston, MA, USA, [4]Northwestern Medicine, Warrenville, IL, USA, [5]Memorial Sloan-Kettering Cancer Center, New York, NY, USA, [6]Tufts Medical Center, Boston, MA, USA, [7]US Oncology, Austin, TX, USA, [8]University of California, San Francisco, San Francisco, CA, USA

Patients with HER2+ breast cancer have frequent LM. A multi center phase 1/2 study assessing safety and efficacy of IT T in LM patients was conducted. The primary endpoint in Phase 2 was response rate (RR). Complete response (CR) required cytologic CR (CCR) + radiographic CR (RCR) + stable clinical function. Partial response (PR) required either CCR with stable/improved imaging or RCR with stable cytology + stable/improved clinical symptoms. Pts received IT T via intraventricular Ommaya reservoir. Phase 1 dosing started at 10 mg, then increasing by 20 mg up to 80 mg. Each cycle (C) was 4 weeks with 2x treatment/week in C1, weekly in C2, and every two weeks after C2. Pts were allowed to continue on hormonal agents if systemic disease was controlled at the time of LM development. Concurrent radiation therapy was not allowed unless exceptionally needed locally for pain control. 34 pts were enrolled with 26 its in the phase 2. The median age was 51 (25–69). IT T was well tolerated with no DLTs seen throughout; determined MTD was 80 mg for phase 2. All patients treated in the Phase 2 had HER2+ breast cancer, 2 patients in the Phase 1 had non-breast histologies. Median cycles completed was 2 (1–22). Median follow up was 9.1 months (0.4- 28.9). In Phase 2, 5 pts (19.2%) had PR, 13 (50%) had SD, and 8 (30.8%) had PD. For Phase 2 pts, median PFS was 2.4 months (CI 1.0–5.5) and median OS was 12.1 months (CI 4.3–19.6) IT T was well tolerated up to a dose of 80 mg. Primary endpoint of 25% RR was not met, however 69% had clinical benefit (stable disease or better). Median OS exceeded historical controls. Future studies are warranted to evaluate IT T in HER2+ LM.

### LPTO-03. IN-VITRO & IN-VIVO CULTURE OF PATIENT (PT) DERIVED CSF-CTCS IN LEPTOMENINGEAL DISEASE (LMDZ) FROM MELANOMA TO IDENTIFY NOVEL TREATMENT STRATEGIES

Vincent Law, Brittany Evernden, Rajappa Kenchappa, John Puskas, Gisela Caceres, Elena Ryzhova, Inna Smalley, Arnold Etame, Solmaz Sahebjam, Anthony Magliocco, Keiran Smalley and Peter Forsyth; Moffitt Cancer Center, Tampa, FL, USA

**BACKGROUND:** Approximately 5% of melanoma pts develop LMDz. There are essentially no models of LMDz available for therapeutic development. Here we report, the *in-vitro* & *in-vivo* culturing of CSF-CTCs. METHODS: CSF-CTCs were detected by the Veridex CellSearch® System. Cell-free DNA and cell-associated DNA were extracted, sequenced and profiled. Expanded *ex-vivo* CSF-CTCs were grown *in-vitro* and tested for drug sensitivity. CSF-CTCs were grown successfully *in-vivo* from 1 pt; labeled human Braf V600E WM164 cells were injected IT in as a control. RESULTS: *CSF-CTCs:* 12 LMDz