

Cancer Research Day

2026 Abstract Book

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POSTER #1

ANTITUMOR EFFECT OF A NEW PYRUVATE KINASE M2 (PKM2) INHIBITOR ACYCLOVIR IN EXPERIMENTAL ESOPHAGEAL ADENOCARCINOMA

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Background: Esophageal adenocarcinoma (EAC) is one of the most aggressive human cancers with poor prognosis even with modern combination therapies due to high resistance to chemotherapy. Therefore, new therapeutic approaches are urgently needed. Pyruvate kinase M2 (PKM2) is not only a key enzyme regulating cancer glycolysis but also translocate into the nucleus to regulate transcription. Although PKM2 is overexpressed in various tumor tissues including EAC, its functional role in esophageal EAC chemotherapy remains unexplored. The objective of this study was to determine whether acyclovir can act as an inhibitor of PKM2 and EAC tumor growth.

Methods: Virtual in silico screening using molecular docking was conducted to screen an FDA-approved chemical library for identification of potential PKM2 inhibitors. HER-2 overexpressing OE19, LPR-OE19 and OE33 EAC cell lines were used. The lapatinib-resistant OE19 (LPR-OE19) cell line was generated from parent OE19 cells through intermittent exposure to increasing concentrations of lapatinib for over six months. Acyclovir and nanoparticle albumin-bound paclitaxel (nab-paclitaxel), alone or in combination, were tested for effects on cell proliferation, lactate production, apoptosis, signaling pathways and tumor growth. Antiproliferative activities were measured by WST-1 assay. Western blotting was performed to evaluate expression of PKM2, apoptotic markers and cell signaling proteins. In-vivo antitumor efficacy was measured in a patient-derived xenograft (PDX) model of human EAC.

Results: Molecular docking identified acyclovir as a potential PKM2 inhibitor that showed lowest docking and glide scores, indicating a strong binding interaction with PKM2. Acyclovir inhibited both PKM2 expression and lactate production. It inhibited cell proliferation in 2D, 3D and organoid cultures of EAC in a dose dependent manner. Interestingly, addition of nab-paclitaxel enhanced the antiproliferative effect of acyclovir. Acyclovir increased expression of proapoptotic proteins and decreased expression of phospho AKT in EAC cells. In a subcutaneous PDX model, acyclovir induced a tumor regression compared to control as monotherapy, and acyclovir in combination with nab-paclitaxel showed a significant enhancement effect of tumor regression. The net change in tumor size in the control, acyclovir, nab-paclitaxel, and combination groups was $551.19 \pm 99.69 \text{ mm}^3$, $347.33 \pm 71.41 \text{ mm}^3$, $221.55 \pm 43.73 \text{ mm}^3$, and $159.37 \pm 39.29 \text{ mm}^3$. Reduction in PDX tumor growth corroborated decreased tumor cell proliferation results.

Conclusions: These data suggest that acyclovir, acting as a PKM2 inhibitor, in combination with nab-paclitaxel should be further investigated as a potential therapeutic strategy for HER2-positive EAC patients and could be a novel treatment strategy for EAC.

Basic Science Faculty

POSTER #2

ACQUIRED RESISTANCE TO PARP1-SPECIFIC INHIBITOR SARUPARIB (AZD5305) REMAINS VULNERABLE TO DNA DAMAGE RESPONSE-TARGETED THERAPEUTICS

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The development of poly (ADP)-ribose (PARP) inhibitors (PARPi) for the synthetic lethal killing of BRCA1/2-deficient cancers revolutionized BRCA1/2-deficient patient outlook and survival. PARPi are now commonly used to treat BRCA1/2-deficient or homologous recombination (HR)-deficient breast, ovarian, prostate, and pancreatic cancers. Despite this clinical success, 30-40% of BRCA1/2-deficient patients exhibit intrinsic resistance and do not respond to PARPi, while the vast majority of those that do initially respond ultimately develop PARPi resistance. This represents a major challenge limiting the clinical impact of PARPi treatment. Understanding PARPi resistance mechanisms is essential for developing better treatment strategies for intrinsic and acquired PARPi-resistant cancers. Recent efforts to better treat BRCA1/2-deficient cancers and combat PARPi resistance led to the development of saruparib (AZD5305), a PARP-specific inhibitor with significantly improved safety profiles due to limited off-target inhibition of other PARP family members and hence reduced toxicity. Saruparib is currently undergoing phase 3 clinical trials and is anticipated to be the standard of care PARPi for BRCA1/2- and HR-deficient cancer patients. We have generated 5 saruparib-resistant (SR) cell lines from parental BRCA1-deficient MDA-MB-436 triple negative breast cancer (TNBC) cells that are >1,000-fold resistant to saruparib but exhibit differential sensitivity to other nonspecific clinical PARPi. SR cell lines exhibit altered inhibition of cellular PARP activity and PARP trapping in response to saruparib and the nonspecific PARPi talazoparib. Whole genome sequencing identified PARP1 active site mutations in each SR cell line, and *in vitro* reconstitution of these PARP1 mutants suggests that they are driving resistance to saruparib and altered sensitivity to other clinical PARPi. Importantly, despite acquired saruparib resistance, SR cell lines retain sensitivity to other DNA damage response (DDR)-targeted therapeutics. Collectively, this work characterizes BRCA1-deficient models of acquired saruparib resistance and uncovers vulnerabilities that may inform rational combination strategies and novel therapeutic approaches for patients who progress on saruparib in the clinic.

Basic Science Faculty

POSTER #3

POPULATION HETEROGENEITY IN IMMUNOTHERAPY TOXICITIES

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Population heterogeneity in treatment-related toxicities remains incompletely characterized in cancer immunotherapy. Using large-scale real-world data, we investigated population-associated differences in reported toxicities following cancer immunotherapy. Several toxicity categories exhibited higher reporting odds among patients across multiple organ systems. These findings highlight population heterogeneity in toxicity profiles associated with cancer immunotherapy and provide real-world evidence that may inform future mechanistic and clinical studies.

Basic Science Faculty

POSTER #4

METABOLOMIC ASSAY FOR ORAL CANCER EXPOSED TO ORAL BACTERIAL-DERIVED SPHINGOLIPID

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Objectives: Oral squamous cell carcinoma (OSCC) is the most common head and neck cancer arising from the oral cavity and lip. The accelerated accumulation level of ceramide/dihydroceramide sphingolipid metabolites in OSCC leads to the suppression of OSCC proliferation. *Porphyromonas gingivalis* (P.g) produces a structurally unique ceramide known as phosphoglycerol-isoC17:0-dihydroceramide-1 (PGDHC). Our knowledge about metabolomic changes in OSCC exposed to PGDHC remains limited. Therefore, we aimed to evaluate the effect of PGDHC on human OSCC cell lines using an untargeted metabolomic assay and a targeted LC-MS/MS assay in vitro.

Methods: Human OSCC cell line OECM-1 was exposed to PGDHC. We included human gingival epithelial cell line OBA-9 as an assay control. Total metabolites and lipids were isolated using Hydrophilic Interaction Liquid Chromatography/MS and Reversed-Phase Liquid Chromatography/MS, employing the developed semisupervised deep learning-based approach for peak curation. Machine learning bioinformatic pathway analysis was then performed to understand the impact of PGDHC on OSCC. We further performed ceramide/dihydroceramide sphingolipids screening by targeted LC-MS/MS assay on OECM-1.

Results: The metabolomic profiling detected a total of 1908 metabolites, and 1276 compounds were confidently identified. PGDHC exposure on OECM-1 showed a significant reduction of glucose metabolites. In contrast, there was no significant effect on glucose metabolites on OBA-9. Machine learning bioinformatic pathway analysis revealed that the exposure of PGDHC on OECM-1 significantly suppressed sphingolipid metabolism and showed the tendency of accelerated levels of ceramides. In addition, pathway analysis showed a significant suppression effect on the Warburg Effect in OECM-1 exposed to PGDHC. Furthermore, the targeted LC-MS/MS assay revealed that PGDHC significantly elevated the intracellular concentration of C20:0, C26:0, C26:1 ceramides, and dhC14:0, dhC16:0, dhC18:0, dhC20:0, dhC20:1, dhC22:1, dhC22:0, dhC24:1, dhC24:0, dhC26:1 dihydroceramides.

Conclusions: The present study demonstrated for the first time that P. g-derived sphingolipid metabolite PGDHC has a suppressive effect on OSCC through sphingolipid metabolism.

Basic Science Faculty

POSTER #5

A HUMAN PHAGE DISPLAY FAB ANTIBODY LIBRARY RESOURCE FOR RAPID ANTIBODY DISCOVERY

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A Human Phage Display Fab Antibody Library Resource for Rapid Antibody Discovery

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Phage display technology is a powerful platform for the rapid discovery of antigen-specific antibodies without the need for immunization, thereby avoiding the labor and time required for antibody humanization. We constructed a large-scale non-immunized human phage display Fab antibody library using peripheral blood mononuclear cells (PBMCs) from 55 healthy donors. The resulting Fab library reached a diversity of approximately 10^{10} variants, representing a broad repertoire of human antibody sequences suitable for screening against diverse targets.

Using this library, we have successfully screened antibodies targeting human CD45, the CD45RA isoform, and B-cell maturation antigen (BCMA). Typically, three to five rounds of biopanning are performed to enrich antigen-specific binders. Enrichment of target-binding antibodies in each round is confirmed by bulk phage ELISA. Individual colonies from the final round of enrichment are then screened using target-phage single-point ELISA and control phage ELISA. Positive colonies showing strong binding to the target antigen and minimal or no binding to control antigens are used to infect host bacteria and subsequently subjected to sequencing. The antibody variable region sequences are analyzed and validated using the IMGT immunogenetics database to confirm their human origin and structural integrity.

This platform enables efficient identification of antigen-specific antibody sequences that can be further developed for research or translational applications. Our workflow typically delivers validated antibody sequences to collaborating investigators. Upon request, we can also express recombinant antibodies and evaluate their antigen-binding specificity. In addition, identified antibody sequences can be adapted for advanced therapeutic engineering, such as incorporation into chimeric antigen receptor (CAR) constructs for CAR-T development.

Given the broad applications of human phage display antibody libraries in biomedical research, we welcome collaborations with investigators interested in screening customized antibodies against proteins, peptides, or other antigens of interest.

POSTER #6

THE CHIMERIC ANTIGEN RECEPTOR REDIRECTED MEMORY T CELLS SPECIFIC FOR CD45 ISOFORM ANTIGENS FOR HEMATOLOGIC MALIGNANCIES

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Background: The primary objective of this project is to engineer chimeric antigen receptor (CAR) T cells to target CD45. CD45 is a transmembrane protein uniformly expressed on hematopoietic cells and progenitors and is also highly expressed in various hematologic malignancies, including leukemias and lymphomas. This ubiquitous expression makes CD45 a compelling therapeutic target. However, its presence on normal hematopoietic cells poses a significant challenge due to on-target, off-tumor toxicity and, importantly, fratricide during CAR manufacturing. To mitigate those limitations, we aim to engineer CD45RO⁺ memory T cell (T_{mem}; defined by CD45RA⁺, B & C⁻ RO⁺) to target CD45 isoform antigens, starting with the dominant CD45RA isoform, to eliminate antigen-bearing hematologic malignancies as well as a strategy to myeloablate the marrow in conditioning to stem cell transplant.

Methods: Firstly, we built a third-generation CAR construct containing a CD45RA-specific single-chain variable fragment (scFv), CD28, and 4-1BB costimulatory domains, and produced lentivirus particles. Secondly, we sorted CD45RA⁻ RB⁻ RC⁻ RO⁺ T cells, generated the CD45RA-CAR- T_{mem}, and evaluated their memory T cell phenotype during cell expansion when supplied with different doses of IL-2, IL-7, and IL-15. Finally, we validated the cytotoxicity of CD45RA-CAR-T_{mem} against various CD45RA-bearing cancer cell lines.

Results: The antigen specificity of the CD45RA-CARs was validated by antigen-dependent killing of targeted cells. The CD45RA-CAR-T_{mem}s recognize CD45RA isoform-bearing cells without cross reactivity to CD45RO, thus mitigating fratricide. CD45RA-RO⁺ T_{mem}s retain their memory T cell features, expressing CD45RA-RO⁺ for six days and expanding when supplied with IL-7, IL-15, and IL-2. For a longer period of cell expansion, IL-21 is required. In a co-culture assay, the CD45RA-CAR-T_{mem}s kill CD45RA-positive Raji cells, KG-1 cells, and HL60 cells, as determined by flow cytometry analysis and LDH assay.

Conclusions: By selectively incorporating the CAR transgene into the CD45RA⁻RO⁺ T_{mem}s as a proof of principle, we showed the engineered T cells to target alternative CD45 RA containing isoforms—either individually or combined, and such CAR-T_{mem}s exert cytotoxic effect on the antigen-expressing cancer cells. Next, we will evaluate the potent cytotoxic activity of the CD45RA-CAR-T_{mem}s *in vivo* using xenograft models of acute myeloid leukemia and other hematologic malignancies. If successful, this work will serve as a foundation for a clinical trial aimed at eradicating minimal residual disease and establishing an effective pre-transplant conditioning regimen for patients with acute leukemias and lymphomas.

Basic Science Faculty

POSTER #7

UTILIZING SUPRAPHYSIOLOGICAL CONCENTRATIONS OF 5 β -DHT TO TARGET CRPC CELLS HARBORING ANDROGEN RECEPTOR MUTATIONS

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Prostate cancer is the most common malignancy and the second leading cause of cancer-related deaths in men in the United States. Due to its dependence on androgen signaling, initial treatment involves androgen deprivation through chemical or surgical castration. However, nearly all patients develop castration-resistant prostate cancer (CRPC), which is lethal and marked by the reactivation of the androgen receptor (AR). Multiple studies have shown that this reactivation can be driven by intratumoral androgenesis from androgen precursors or metabolites (APMs), or through direct activation of AR by the APMs themselves.

Paradoxically, Bipolar Androgen Therapy (BAT) has emerged as a promising strategy for CRPC patients. Unlike traditional ADT, BAT cycles between castrate and supraphysiological levels of testosterone. Clinical trials have demonstrated that BAT can achieve up to 30% efficacy in CRPC patients, leading to measurable growth suppression and PSA response through AR overactivation. However, current BAT protocols rely on systemic testosterone administration, which is limited by a short duration of response (average 6 months) and off-target effects potentially due to AR expression in healthy tissues. This underscores an unmet need to specifically target CRPC cells.

We hypothesize that supraphysiological levels of select APMs could be used to directly target CRPC cells containing mutant AR. Our data shows that 5-beta-dht, a testosterone metabolite thought to be inactive, could be an ideal candidate. Our data demonstrates that 5-beta-dht can induce cellular proliferation in hormone deficient conditions, and can inhibit the growth of CRPC cells with mutant AR when treated with supraphysiological conditions. Furthermore, we show that 5-Beta DHT is specific to AR mutants by demonstrating no metabolism to testosterone. We will further test our hypothesis by utilizing a GFP reporter to verify that no WT AR is being activated by 5-beta DHT and perform resistance studies to determine its effect on therapeutic windows.

Basic Science

Graduate Student

POSTER #8

THE ROLE OF ZNFX1 ANTISENSE RNA 1 (ZFAS1) IN MOLECULAR-IMMUNE INTERACTIONS AND PLATINUM RESISTANCE IN HIGH GRADE SEROUS OVARIAN CANCER

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High-grade serous tubo-ovarian carcinoma (HGSC) is the most common and prognostically the most unfavorable histological subtype of epithelial ovarian cancers. The standard of care for HGSC patients includes a primary debulking surgery followed by platinum (Pt) based chemotherapy. Despite initial response to standard of care treatment, around 80% of the patients eventually develop resistance, leading to recurrent, drug-resistant HGSC, which is uniformly fatal. Long non-coding RNAs (lncRNAs), a class of non-protein coding transcripts, have emerged as crucial regulators of tumor progression and therapy response in ovarian cancer (OC). We found that the lncRNA, ZNFX1 antisense RNA 1 (ZFAS1), is upregulated in HGSC tumors, and in HGSC cell lines, promotes proliferation, survival and invasion, as assessed by colony formation and migration assays. Additionally, we found that ZFAS1 is significantly upregulated in Pt-resistant patient tumors and Pt-resistant HGSC cell lines compared to their sensitive counterparts. However, the underlying mechanism and function of ZFAS1 in HGSC, overall and in Pt-resistance, is incompletely understood. Analysis of TCGA_OV dataset and RNA-seq data from Pt resistant HGSC patients revealed that high ZFAS1 expression is associated with suppression of immune-related pathways and reduced immune cell signature. Based on this association, we hypothesize that ZFAS1 could suppress immune response by regulating inflammatory cytokine signaling. Consistent with this hypothesis, ZFAS1 knockdown in HGSC cells resulted in upregulation of pro-inflammatory cytokines TNF- α , CCL5, CXCL5, and IL-6, indicating that ZFAS1 downregulates pro-inflammatory cytokine expression in HGSC. Notably, qPCR and western blot analysis showed that induction of pro-inflammatory cytokines was greater in Pt-resistant cells compared to their parental sensitive counterparts. These observations suggest that elevated expression of ZFAS1 in Pt resistant HGSC tumors suppresses pro-inflammatory cytokines and contributes to an immunosuppressive environment. To determine the functional role of ZFAS1 in mediating immune response, HGSC cells with ZFAS1 knockdown were co-cultured with natural killer (NK) cells. ZFAS1 depletion resulted in increased NK cell-mediated cytotoxicity towards HGSC cells, indicating that ZFAS1 downregulates immune response. Collectively, these findings identify a previously unrecognized oncogenic role of ZFAS1 in HGSC and further indicate that high ZFAS1 expression in Pt resistant HGSC promotes immune suppression by downregulating pro-inflammatory cytokines and NK cell activity.

Basic Science

Graduate Student

POSTER #9

ASSESSMENT OF SEX DIFFERENCES IN URETHANE-INDUCED LUNG TUMORIGENESIS IN THE FOUR CORE GENOTYPES MOUSE MODEL

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Background/Objectives:

Sex differences in lung cancer incidence and outcomes are well recognized, yet the biological mechanisms underlying these disparities remain incompletely defined. In particular, the relative contributions of sex chromosome complement and gonadal sex are difficult to distinguish because these factors are inherently linked in most experimental systems. We hypothesized that sex chromosome complement and gonadal sex independently influence disease progression in urethane-induced lung tumorigenesis. To test this hypothesis, we used the Four Core Genotypes (FCG) mouse model, which decouples chromosomal sex (XX vs. XY) from gonadal sex (female vs. male), enabling independent evaluation of these variables.

Methods: FCG mice (6–8 weeks old) received intraperitoneal urethane injections (1 g/kg body weight) weekly for 10 weeks and were evaluated after a 20-week latency period. Lung tumor multiplicity, tumor area ratios, normalized lung weight, and Ki-67 proliferation indices were quantified histologically. Bronchoalveolar lavage fluid (BALF) cellularity was analyzed to assess airway inflammation. Tumor mutation status was determined by Sanger sequencing, and mutant RAS (Q61R) and phosphorylated ERK (p-ERK) immunoreactivity were quantified by immunohistochemistry. Hepatic Cyp2e1 expression was measured by quantitative PCR to evaluate urethane bioactivation capacity. Survival differences were analyzed using Kaplan–Meier methods with log-rank testing.

Results: Survival differed significantly across genotypes. XYM mice exhibited reduced survival compared with XXF mice (log-rank $P = 0.0157$). Stratified analyses also revealed poorer survival in XY compared with XX mice ($P = 0.046$) and in gonadal males relative to gonadal females ($P = 0.0252$). Despite these survival differences, tumor burden metrics did not significantly differ among genotypes. Tumor multiplicity was comparable across groups (Poisson regression LR $\chi^2 = 6.97$, $df = 3$, $P = 0.073$), and genotype did not significantly influence tumor area ratios or Ki-67 proliferation indices. Urethane exposure significantly increased lung weight independent of genotype. BALF analyses demonstrated increased airway inflammation following urethane treatment, including elevated lymphocyte counts, with significant main effects of genotype ($P = 0.0449$) and treatment ($P = 0.0256$) but no genotype \times treatment interaction. Sanger sequencing confirmed canonical Kras Q61R mutations in urethane-induced tumors. Quantitative immunohistochemistry revealed a significant genotype effect on mutant RAS (Q61R) expression ($F_{3,23} = 3.48$, $P = 0.032$), with the highest H-scores observed in XYF tumors. In contrast, cytoplasmic and nuclear p-ERK levels did not differ significantly across genotypes. Hepatic Cyp2e1 expression also did not differ by genotype or treatment, indicating comparable urethane bioactivation capacity.

Conclusions:

Sex chromosome complement and gonadal sex influence survival following urethane exposure independently of tumor burden, proliferative activity, MAPK signaling, or carcinogen metabolism. These findings suggest that sex-associated differences in disease outcomes arise from systemic or tumor-microenvironmental mechanisms rather than intrinsic differences in tumor growth.

Basic Science *Graduate Student*

POSTER #10

MOLECULAR CHARACTERIZATION OF STOMACH METAPLASIA PHENOTYPES INDUCED BY CYTOKINE TREATMENT OR BACTERIAL INFECTION

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The primary cause of gastric cancer is *Helicobacter pylori* (Hp) infection, which is associated with chronic inflammation including production of the type 1 cytokine interferon-gamma (IFN γ). Inflammation elicits a series of tissue changes that culminate in metaplasia (pre-cancer), dysplasia, and finally tumor formation. Metaplasia is characterized by gastric expression of intestinal mucins like MUC2 and MUC4. However, there are still many open questions about the cell(s) of origin of metaplasia and the druggability of this cell state. Thus, we sought to recapitulate Hp-induced metaplastic phenotypes by treating human gastric cell lines and healthy mouse stomach organoids with recombinant IFN γ . Metaplasia phenotypes were analyzed by qRT-PCR, western blotting, and immunostaining.

Compared to control-treated organoids, organoids treated with IFN γ were larger and irregularly shaped, and had a significant induction of the metaplasia markers Muc4 and Tff2. Withdrawal of IFN γ reversed these phenotypes, suggesting that metaplasia may be a transient response to inflammation. The type 1 cytokine tumor necrosis factor- α and the type 2 cytokine IL-13 did not elicit these phenotypes, suggesting that these changes may be unique to IFN γ treatment. In AGS gastric cancer cells, 24 hours of IFN γ treatment did not induce metaplasia marker expression. However, AGS cells infected with Hp for 24 hours did have significant upregulation of MUC4 and TFF2. In contrast, GES-1 cells, an SV40-immortalized cell line arising from healthy stomach, did not exhibit MUC4 or TFF2 induction in response to either IFN γ treatment or Hp infection. Taken together, these results suggest that IFN γ treatment is sufficient to induce metaplasia phenotypes in healthy gastric organoid cells, but not immortalized gastric cell lines. Future work will use these model systems to define the molecular mechanisms underlying metaplasia marker induction in response to cytokine treatment and/or Hp infection, and to explore the druggability of metaplastic cell populations.

Basic Science

Graduate Student

POSTER #11

IDENTIFYING TRANSCRIPTIONALLY ACTIVE BIOMARKERS OF TRIPLE-NEGATIVE BREAST CANCER BY ANALYZING CDK-DEPENDENCE USING PHOSPHOPROTEOMICS

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Triple-negative breast cancer (TNBC) is an aggressive subtype of breast cancer with high morbidity and poor prognosis, accounting for 15% of all breast cancer patients. It is classified as triple-negative because it lacks expression of estrogen, progesterone, and Her2 receptors. As a result, there are limited therapeutic options available.¹ Because TNBC tumors are highly dependent—or “addicted”—to transcription factors such as MYC for continued proliferation and survival, one strategy for targeting this aggressive subtype is to disrupt the transcriptional machinery.² Transcription is regulated by RNA polymerase II (RNAPII), consisting of 12 distinct subunits with subunit RPB1 containing the c-terminal domain (CTD). The CTD is composed of a conserved heptad repeat (YSPTSPS), subject to many post-translational modifications, among which phosphorylation plays a vital role.³ Phosphorylation of the CTD is regulated by cyclin-dependent kinases (CDKs), which control transcriptional progression by altering phosphorylation levels of residues in the heptad repeat.⁴ Some scientists believe using the phosphorylation status of RNAPII-CTD could be a potential biomarker of cancer, but the method used, ChIP-seq (Chromatin Immunoprecipitation Sequencing), is technically challenging and not easily performed in clinical labs. ChIP-seq uses monoclonal antibodies directed against repetitive phosphorylation sites in the RNAPII CTD, the repeats can cause epitope overlap with neighboring residues, and avidity effects from multivalent antibody binding to heptad repeats, all of which lead to inaccurate quantification and localization of phosphorylation events.⁵ Therefore, the purpose of this study is to identify transcriptionally active biomarkers that can be quantitated with high selectivity and specificity that are specific to TNBC. In this work, this will be performed by evaluating CDK dose-dependent responses in protein phosphorylation using flavopiridol. Flavopiridol is a potent pan-CDK inhibitor, extremely effective in inhibiting transcription through targeting of CDK9, which forms part of the P-TEFb complex (Positive Transcription Elongation Factor b) required for transcriptional elongation.⁶ Two breast tissue cell lines will be used, MDA-MB-231 and MCF10A. MDA-MB-231 cell lines were used to model TNBC, categorized as triple-negative with high MYC expression and sensitivity to CDK inhibitors.² Completed work in HEK293T cell lines were treated with DMSO (dimethyl sulfoxide), 125 nM, 350 nM, and 1,000 nM of flavopiridol. We will describe potential targets along with our analysis strategy for our extensive datasets for HEK293T that we will extend to new studies in TNBC cell lines to discover TNBC specific targets.

Basic Science

Graduate Student

POSTER #12

DIVERGENT TRANSCRIPTOMIC AND PROTEOMIC CONSEQUENCES OF DISEASE-ASSOCIATED MUTATIONS IN SUBUNITS OF THE RNA EXOSOME

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The RNA exosome is a conserved, multi-subunit complex that is critical for RNA processing, quality control, and degradation in eukaryotes. Although these proteins function within the same complex, mutations in distinct RNA exosome subunits drive divergent diseases, including Multiple Myeloma (MM) and neurodegenerative diseases. Mutations in DIS3, the 3'-5' exonuclease of the complex, occur in approximately 10% of MM patients as recurrent heterozygous hotspot or homozygous non-hotspot mutations. Variants in EXOSC3, a critical cap subunit of the complex, are associated with a rare neurodegenerative disorder called Pontocerebellar Hypoplasia Type 1b (PCH1b). Although DIS3 and EXOSC3 are within the same ribonucleolytic complex, it remains unclear whether mutations in different subunits of the same complex produce distinct or overlapping molecular consequences. We hypothesize that mutations in DIS3 and EXOSC3 produce both shared and subunit-specific alterations in global RNA and protein abundance, reflecting a shared disruption of RNA exosome function alongside distinct consequences arising from each subunit's unique structural and catalytic role.

In collaboration with the Indiana University School of Medicine Gene Editing Core and Dr. Brian Walker, we generated human cell models of MM and PCH1b using non-hotspot and hotspot DIS3 mutations in KMS11 and EXOSC3 mutations in HEK293T cell lines, respectively. Transcriptomic changes were quantified by total RNA-sequencing (n=3), and global proteome changes were quantified using TMT-labeled mass spectrometry (n=4). Integrated multi-omics analysis was performed to identify shared and subunit-specific alterations in the transcriptome and proteome along with gene set enrichment analysis.

Transcriptomic analysis revealed a greater number of significantly decreasing transcripts compared to the control in both the PCH1b and MM cell models, suggesting impaired RNA exosome function in both disease models. Both disease models had significant enrichment of transcripts regulated by AU-rich elements, consistent with previous RNA exosome studies reported in yeast. Notably, lncRNAs were significantly increased only in the MM models, while PCH1b models exhibited more lncRNA downregulation. Proteomic analyses revealed broader alterations of the abundance of RNA exosome subunits and known accessory complex members in the PCH1b models than the DIS3 models, suggesting greater disruption of complex integrity and protein complex dynamics in PCH1b than MM. Gene enrichment analysis revealed shared and subunit-specific pathways between the disease models, defining distinct molecular signatures associated with each disease pathology.

These findings demonstrate that mutations within the same complex are not functionally equivalent and drive distinct disease pathologies. MM associated DIS3 mutations have a unique impact on the transcriptome and proteome that is mechanistically distinct from the impact observed from EXOSC3 mutations associated with PCH1b. Defining subunit-specific consequences of RNA exosome dysfunction provides mechanistic insight into MM pathogenesis and may inform precise therapeutic strategies targeting RNA metabolism vulnerabilities from DIS3 mutations.

Basic Science

Graduate Student

POSTER #13

DISRUPTION OF KU-ALU RNA BINDING BY SMALL MOLECULE KU-TARGETED INHIBITORS AND ITS IMPLICATION IN DEVELOPING NOVEL CANCER THERAPY

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DNA double strand breaks (DSBs) are cytotoxic lesions. In mammalian cells, DSBs are repaired primarily by non-homologous end joining (NHEJ), which functions throughout the cell cycle, or homologous recombination (HR), which occurs mainly during S and G2. The Ku70/80 (Ku) heterodimer is a DNA damage sensor protein that plays a role in detecting DNA DSBs. Ku's binding to DNA DSB ends is required for the recruitment and activation of the transducer kinase DNA dependent protein kinase catalytic subunit (DNA-PKcs), ultimately facilitating repair via the NHEJ pathway. Given its role in DSB repair, Ku is an attractive cancer therapeutic target. Inhibition of Ku can enhance sensitivity to DNA DSB inducing agents and induce synthetic lethality in homologous recombination (HR) deficient tumors. Ku is essential in human cells, but not in other organisms. Interestingly, Ku's essentiality in humans is independent of its role in DNA repair. This is evidenced by the fact that no human patients with biallelic loss of function mutations in *XRCC5* (Ku80) or *XRCC6* (Ku70) have been identified, whereas loss of function mutations in downstream NHEJ genes have been observed in humans. Thus, Ku's essentiality in humans must stem from functions beyond DNA repair, potentially linked to emerging evidence of its role in RNA regulation. Increased expression of Ku occurred concurrently with expansion of Alu repeats (primate-specific, repetitive, short interspersed nuclear elements (SINES)) in the genome at the prosimian-new world monkey evolutionary junction. Two models have been proposed for Ku's RNA-binding functions in cell survival: one suggests Ku binds Alu elements to prevent dsRNA-induced innate immune activation, while another proposes it binds antisense intronic Alu elements to block cryptic splice site usage. Although the precise mechanism remains unclear, we sought to determine whether our novel Ku-DNA binding small molecule inhibitors (Ku-DBis) also disrupt Ku-RNA interactions. We hypothesize that our novel Ku-DBis disrupt Ku-RNA interactions, contributing to observed toxicity and thereby informing the development of more selective inhibitors. We tested five different KuDBis utilizing fluorescent polarization with an antisense stem-loop 1 Alu (asSL1) substrate. Ku-DBi 3392 inhibited Ku-RNA binding when pre-incubated with Ku and when introduced after Ku-RNA complex formation. Electrophoretic mobility shift assays (EMSA) reveal compounds 3392 and 3393 inhibit binding when pre-incubated with Ku. The data reveal compound 3392 is more potent than 3393, with both exhibiting titratable effects. Data will also be presented describing asSL1 Alu substrate optimization, investigation of the relationship between Ku and the translation initiation factor *eIF3b*, and examination of the impact of post-translational modifications on Ku-RNA binding. Defining Ku-RNA interactions will clarify Ku's DNA repair independent roles and inform development of selective cancer therapeutics.

Basic Science

Graduate Student

POSTER #14

UNDERSTANDING AN INTERPLAY BETWEEN HSF1 AND ERR α IN TRIPLE NEGATIVE BREAST CANCER

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Breast cancer is the most common malignancy in women, with 1 in 8 at risk during their lifetime. Despite decades of research, incidence and mortality continue to rise. Oncogenesis often stems from genetic alterations that drive transcriptional dysregulation, enabling cancer cells to withstand oxidative, metabolic, and proteotoxic stresses and thereby promote survival. ERR α (Estrogen-Related Receptor α), an orphan nuclear receptor that regulates metabolic gene expression, has been implicated in ovarian and breast cancers. Although ERR α inhibition shows therapeutic promise, its mechanisms of action remain incompletely defined. HSF1 (Heat Shock Factor 1), a master regulator of the heat shock response and proteostasis, is critical for oncogenesis and promotes tumor progression through diverse pathways including proliferation, EMT, and immune evasion. However, potential cooperation between ERR α and HSF1 has not been investigated.

ChIP-seq analyses revealed extensive overlap in ERR α and HSF1 binding peaks and shared target genes enriched for cancer-relevant pathways such as proliferation, cell junction assembly, and Wnt signaling. Using a novel ERR α transcriptional activity signature, we observed a strong correlation between ERR α and HSF1 activity in breast cancer patient datasets, with high co-activity predicting poorer prognosis. Mechanistically, ERR α enhanced HSF1 transcriptional activity, while HSF1 expression increased ERR α activity. Pharmacologic inhibition of ERR α (XCT790) reduced HSF1 protein levels in HEK293FT and HCC1937 cells, whereas HSF1 inhibition (SISU102) reduced ERR α protein expression. Co-immunoprecipitation confirmed a physical interaction between ERR α and HSF1, supporting the formation of a protein complex. Functionally, co-overexpression of HSF1 and ERR α significantly increased cell proliferation and colony formation compared to control conditions, demonstrating an enhancement of tumorigenic potential.

Together, these findings support a model in which ERR α and HSF1 form a stabilizing protein complex that co-regulates pro-tumorigenic transcriptional programs, thereby promoting breast cancer growth and progression. Ongoing studies are evaluating the role of this complex in therapy resistance, with the goal of targeting this interaction to improve chemotherapeutic response.

Basic Science Graduate Student

POSTER #15

TO INVESTIGATE THE ROLE OF ENTEROENDOCRINE CELLS IN IMMUNE SUPPRESSION IN BRAF-MUTANT COLORECTAL CANCER

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BRAF is a serine/threonine kinase essential to MAPK signaling, regulating cell proliferation, growth, and survival. Activating BRAF mutations-primarily BRAF^{V600E}-are found in ~12% of metastatic colorectal cancer (CRC) cases and are associated with aggressive tumor behavior and resistance to chemotherapy. Although immune checkpoint inhibitors benefit microsatellite-*instable* CRCs, 70-80% of BRAF-mutant CRCs are microsatellite *stable* (MSS) and largely unresponsive, contributing to their poor prognosis. Recent studies reveal an enrichment of enteroendocrine cell (EEC) (**specialized neuroendocrine cells** of the intestine) progenitors in BRAF-mutant colorectal cancer compared to non-mutant cases. Neuroendocrine cells have been increasingly linked to lineage plasticity and are enriched in aggressive and therapy-resistant subtypes of certain cancers like neuroendocrine prostate cancer, Merkel cell carcinoma, and small-cell lung cancer. However, it is not well understood how neuroendocrine cells contribute to the aggression and therapy-resistance of these cancers. Therapy responses in cancer are strongly shaped by the density, composition, and spatial organization of tumor-infiltrating immune cells. Microsatellite-*stable* BRAF-mutant CRCs have an immune “cold” tumor microenvironment that contributes to their insensitivity to immune checkpoint inhibitors. My preliminary orthotopic mouse study using microsatellite-*stable* mouse BRAF-mutant CRC organoids engrafted into colons of wild-type mice showed that tumors arising from EEC-deficient CRC organoids exhibited increased immune-cell infiltration and necrosis compared to EEC-containing controls, consistent with enhanced immunogenic cell death. **Hence, I hypothesize that EECs play a key role in promoting and maintaining tumor growth by suppressing cytotoxic immune responses in BRAF-mutant CRC.** Aims of this study focus on determining the effect of EECs on immune suppression and to accurately capture the immune landscape during early stages of tumor development. The effect of the adaptive immune response on modulating tumor development in the absence of EECs will also be studied using an immunodeficient *Rag1*KO mouse model. Mechanistic insights into EEC-mediated immune suppression will be gained by tumor secretome analysis. Overall, this study will help understand how EECs affect the immune profile and activity in BRAF-mutant colorectal cancer. It will also highlight a new link between neuroendocrine cells and immune modulation in cancer. A better understanding of enhanced immune cell infiltration will improve existing anti-cancer immunotherapy outcomes and pave the way for developing novel therapeutic strategies.

Basic Science

Graduate Student

POSTER #16

DIRECT PHARMACOLOGICAL TARGETING OF ASPARAGINE SYNTHETASE TO OVERCOME RESISTANCE TO L-ASPARAGINASE IN ALL THERAPY

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Acute lymphoblastic leukemia (ALL) is the most common pediatric cancer, arising from both B- and T-cell lineages. Current therapy exploits leukemia cells' low expression of asparagine synthetase (ASNS) by using L-asparaginase, a bacterial enzyme that depletes circulating asparagine. Although L-asparaginase is generally effective, resistance can develop in some patients, leading to refractory or relapsed disease. The amino acid-sensing kinase GCN2 in the integrated stress response contributes to this resistance by promoting ASNS expression. Furthermore, additional mechanisms may contribute to elevated ASNS expression and resistance to L-asparaginase therapy. In this study, we addressed the efficacy of L-asparaginase in combination with genetic or pharmacological inhibition of GCN2 and a novel ASNS inhibitor designated ASX-173. Using a *Kras*^{G12D}-driven mouse model of T-ALL, we found that GCN2 is dispensable for leukemogenesis. However, genetic inactivation or pharmacologic inhibition of GCN2 sensitized ALL cells to asparagine depletion, correlating with impaired ASNS induction. While GCN2 targeting enhanced sensitivity to asparagine depletion, a subset of *Gcn2*⁺ T-ALL cells retained high ASNS expression and remained resistant to L-asparaginase. Likewise, some human T-ALL cells with elevated ASNS levels were refractory to GCN2 inhibition even under asparagine-depleted conditions. When combined with L-asparaginase, ASX-173 effectively eliminated ASNS-high leukemic cells *in vitro* and *in vivo*. These findings suggest that direct targeting of ASNS provides therapeutic benefit in leukemias that express high ASNS and are resistant to GCN2 inhibition under asparagine-depleted conditions.

POSTER #17

INHIBITION OF ASPARAGINE SYNTHETASE ACTIVATES THE INTEGRATED STRESS RESPONSE

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Asparagine synthetase (ASNS) catalyzes the ATP-dependent biosynthesis of L-asparagine (L-Asn) from L-aspartate and L-glutamine and plays a central role in therapies targeting amino acid metabolism, including L-asparaginase treatment for acute lymphoblastic leukemia. In response to L-Asn depletion, ASNS is strongly induced through the GCN2–ATF4 arm of the integrated stress response (ISR), enabling tumor cells to maintain amino acid homeostasis under nutrient stress. Despite the therapeutic potential of targeting this pathway, direct pharmacologic inhibition of ASNS has been limited by the poor cellular activity of existing inhibitors. Here, we characterize the molecular and cellular mechanism of ASX-173, a cell-penetrant ASNS inhibitor with potent biochemical and cellular activity that has the potential to deprive tumors of asparagine and enhance the efficacy of metabolic combination therapies.

In HEK-293A cells cultured without exogenous L-Asn, ASX-173 markedly reduced intracellular L-Asn levels, activated the ISR, and increased ATF4-dependent transcriptional activity. ISR activation was fully reversed by physiological levels of L-Asn, confirming on-target activity. Genetic deletion of GCN2 lowered basal ASNS expression, impaired ATF4 induction, and increased cellular sensitivity to ASX-173. Similarly, pharmacologic inhibition of GCN2 with GCN2iB suppressed ASX-173-induced ATF4 activity and synergistically inhibited proliferation across multiple cancer cell lines, including renal and prostate cancer models. Cell-based thermal protein profiling demonstrated direct target engagement, shifting the ASNS melting temperature from 45 °C to 54 °C.

Biochemical studies using recombinant ASNS showed that ASX-173 inhibits L-Asn synthesis without affecting L-glutamate production and displays reduced activity at the K_m for Mg^{2+} -ATP. Differential scanning fluorimetry revealed that ASX-173 binding requires Mg^{2+} -ATP. The cryo-EM structure further revealed that ASX-173 is bound within the C-terminal synthetase subdomain, alongside AMP, pyrophosphate, and two Mg^{2+} ions, forming a composite pocket stabilized by hydrophobic contacts, π - π interactions, and hydrogen bonding to AMP. Structural analysis suggests that ASX-173 promotes ATP hydrolysis while blocking ammonia transfer, consistent with an uncompetitive mechanism of inhibition.

Together, these findings establish ASX-173 as a potent ASNS inhibitor with strong cellular target engagement and ISR-activating activity. The enhanced sensitivity of GCN2-deficient cells and the observed synergy with pharmacologic GCN2 inhibition highlight the therapeutic potential of dual ISR targeting and support ASNS inhibition, alone or in combination with ISR-modulating strategies, as a promising metabolic vulnerability in cancer.

Basic Science

Graduate Student

POSTER #18

SPATIAL TRANSCRIPTOMICS UNVEILS TUMOR MICROENVIRONMENT CHANGES IN MURINE GLIOBLASTOMA UPON EGFRVIII SUPPRESSION

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EGFRvIII, a tumor-specific in-frame deletion of exons 2–7 that generates a constitutively active EGFR mutant, drives potent oncogenic signaling in a molecularly defined subset of glioblastoma (GBM) and is implicated in altered invasion, therapy resistance and immune modulation; because EGFRvIII occurs in a substantial fraction of GBMs we asked how suppression of this mutation in established tumors reshapes tumor cells and immune microenvironment in order to reveal treatment opportunities. We used an inducible EGFRvIII GBM mouse model to perform single-cell RNA sequencing and spatial transcriptomics on established with and without transcriptional suppression of EGFRvIII. Findings were validated in spatial transcriptomic profiles from human GBM specimens stratified by EGFR status.

EGFRvIII suppression increases infiltration of myeloid and lymphoid cells into core tumor regions, slowing most tumor growth and increasing mouse survival. However, it also promotes invasion and proliferation in EGFRvIII-independent tumor-subpopulations, due to loss of EGFRvIII-driven communication between EGFRvIII-positive and -negative subpopulations. These observations are consistent with prior literature knowledge that direct EGFR inhibition often leads to resistance by vIII-negative populations, and complements it with insights into its mechanistic bases. Overall, our data argue for approaches that exploit the immune microenvironment changes to enhance direct EGFRvIII-targeted strategies for vIII-positive GBM.

Basic Science

Graduate Student

POSTER #19

GCN2-EIF2 α SIGNALING AXIS DOWNREGULATES HSF1 AND INHIBITS CANCER CELL GROWTH

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Under states of proteotoxic stress, Heat Shock Factor 1 (HSF1) serves as the primary transcription factor responsible for sensing stress and transcribing proteins essential for restoring protein homeostasis. Also tasked with alleviating cellular stress is General Control Nonderepressible 2 (GCN2). Activated by a shortage in amino acids, GCN2 triggers the phosphorylation of eIF2 α (p-eIF2 α) and the subsequent downregulation of global protein synthesis while selectively translating proteins such as Activating Transcription Factor 4 (ATF4), to promote cell survival. Due to rapid proliferation, uncontrolled growth, and increased metabolic demands, cancer cells often exploit the HSF1 and GCN2 pathways to mitigate cellular damage. In Glioblastoma (GBM), HSF1 promotes tumor growth, migration, invasion, and apoptosis evasion, while GCN2 is critical in maintaining cancer cell homeostasis, proliferation, and survival. Recently, NXP800 (CCT361814) has emerged as both an HSF1 pathway inhibitor and a GCN2 pathway agonist, leading to antitumor activity both *in vitro* and *in vivo*. However, it remains unclear whether there is a mechanism by which HSF1 and GCN2 biologically integrate to modulate GBM fate. Using patient data, we first demonstrated a negative correlation between the GCN2 and HSF1 pathways in GBM. Subsequent *in vitro* studies showed pharmaceutical activation (NXP800 or Halofuginone) of the GCN2 pathway leads to a decrease in HSF1 protein levels and pathway activity while inhibition or knock-out of GCN2 leads to a partial rescue of the effects. Antagonizing GCN2-pathway activation using ISRIB rescued HSF1 protein levels and activity, confirming that the suppression of HSF1 is p-eIF2 α dependent. Additionally, activation of the GCN2 pathway leads to decreased cell viability which is mitigated by HSF1 knock-out. Together, these findings identify a novel GCN2-dependent mechanism of HSF1 regulation in GBM.

Basic Science Graduate Student

POSTER #20

METHYLATION OF A CONSERVED LYSINE REGULATES CLIENT BINDING BY 14-3-3 PROTEINS

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Lysine methylation (Kme) is an important post-translational modification (PTM) with roles in protein regulation beyond its classical function in chromatin biology. Kme has been detected on nearly a quarter of the human proteome, but detection of these sites has far outpaced their functional annotation. Importantly, lysine methyltransferases (KMTs) and lysine methylation sites are frequently dysregulated in cancer, where they contribute to altered gene expression and signaling states that promote tumorigenesis. Defining the functional consequences of methylation on individual protein substrates is therefore an important gap in knowledge requiring further study.

In this work, we investigate the functional significance of Kme on the 14-3-3 family of phospho-binding scaffold proteins. 14-3-3 proteins bind hundreds of phosphorylated client proteins and serve as central regulators of cellular signaling networks controlling processes including cell cycle progression, apoptosis, metabolism, and stress responses. Dysregulation of 14-3-3 signaling has been implicated in many cancer types, where altered expression or activity of specific paralogs contributes to tumor initiation, progression, and metastasis. Despite their central role in coordinating signaling networks, the molecular mechanisms that regulate 14-3-3 binding specificity remain incompletely understood. Previous proteomic studies by our group show that a conserved lysine residue within the amphipathic binding groove of 14-3-3 proteins is commonly methylated. Structural analysis suggested that methylation at this position could disrupt electrostatic interactions between 14-3-3 and phosphorylated client proteins. We therefore hypothesized that methylation of this residue acts as a post-translational regulator of 14-3-3 binding by weakening electrostatic interactions with its clients.

To investigate this, we employed methyl-lysine analog (MLA) chemistry, a synthetic chemical biology technique, to site-specifically and methyl-state-specifically install analogs of lysine and methyl-lysine on recombinant 14-3-3 proteins. This model system has been used successfully in studies of histones, but to our knowledge this is its first application on a non-histone protein. Intact and bottom-up mass spectrometry confirmed efficient installation of the expected modifications. Using fluorescence polarization assays, we found that a Kme2 analog decreases binding affinity between 14-3-3 and client phosphopeptides relative to unmodified lysine analogs and wild-type protein. These results suggest that Kme within the 14-3-3 binding groove weakens client binding. To evaluate the impact of this modification in cells, we are performing immunoprecipitation-mass spectrometry experiments using recombinant MLA-modified 14-3-3 proteins and exogenously expressed mutants to functionally define how Kme alters the 14-3-3 interactome. In ongoing work, we are also examining how this modification influences interactions with specific signaling partners, including the Hippo pathway effector YAP1, which is frequently dysregulated in cancer. In parallel, we are conducting a lysine methyltransferase (KMT) overexpression screen to identify the enzyme(s) responsible for depositing this mark. Together, these studies aim to define lysine methylation as a regulatory mechanism controlling 14-3-3 signaling networks in cancer.

POSTER #21

PP2A-B56 α ACTIVATION DRIVES ABERRANT MACROPINOCYTOSIS IN PANCREATIC CANCER

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The microenvironment of pancreatic ductal adenocarcinoma (PDAC) has increased interstitial fluid pressure and poor vascularization, leading the cancer cells to be nutrient deprived. In order to bypass this depletion, PDAC cells will often activate alternative pathways to acquire the nutrients needed to support their survival. One such pathway shown to be activated in PDAC is macropinocytosis, an actin-driven nutrient scavenging pathway. During macropinocytosis, extracellular fluid is internalized into intracellular vesicles called macropinosomes, which fuse with lysosomes to replenish nutrients. Because macropinocytosis is important for PDAC cell survival, the inhibition of the pathway is considered a potential therapeutic strategy. Previous work has shown that tumor suppressor protein phosphatase 2A (PP2A) is involved in the regulation of macropinocytosis. To better understand the role of PP2A in macropinocytosis, high molecular weight Oregon Green Dextran (70kDa), which is taken up through macropinocytosis, was used to analyze macropinocytosis. Upon treatment of DT-061, a pharmacological PP2A activator, as well as genetic overexpression of the specific PP2A subunit, B56 α , there is a significant increase in dextran signal compared to the control condition. To further assess the role of PP2A in the vesicle trafficking, endocytic (Rab5 and Rab7) and lysosomal markers (Lamp1) and their colocalization with dextran was analyzed. Upon DT-061 treatment, there was no significant difference between the colocalization of Rab5 or Rab7 with macropinosomes, while there was a significant decrease in colocalization of Lamp1 with macropinosomes compared to vehicle. Given this result, Dye Quenched Bovine Serum Albumin (DQ-BSA) was added to cells, which will only fluoresce upon cleavage in low pH environments such as lysosomes. With DT-061 treatment, the number of DQ-BSA puncta were significantly decreased compared to vehicle, suggesting that PP2A activation disrupts macropinosome-lysosome fusion. Through the use of co-immunoprecipitation and proximity ligation assay, we have identified that PP2A-B56 α interacts with PIKfyve, a lipid kinase that has been implicated in lysosomal fusion. Future studies will interrogate this impact of this interaction on macropinocytosis and determine the functional consequence of PP2A-regulated phospho-sites. Together, these findings identify novel posttranslational mechanisms that contribute to PDAC macropinocytosis and implicate PP2A as a novel therapeutic target to suppress macropinocytosis-driven nutrient scavenging.

Basic Science

Graduate Student

POSTER #22

TRUNCATED ETV1 UTILIZES THE BGN-TLR4-P38 AXIS TO DRIVE CELL MIGRATION AND ONCOGENIC PHENOTYPES IN PROSTATE CELLS

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In the majority of prostate cancer (PCa) cases, a gene encoding an ETS transcription factor is fused to an active promoter resulting in aberrant expression in the prostate. Of the ETS factors, ERG (50%), ETV1 (10%), and ETV4 (5%) have been implicated in prostate cancer. While much is known regarding how ERG functions to induce oncogenic changes in the prostate epithelium, the role of ETV1 in PCa remains poorly defined. In PCa, ETV1 is expressed as either a full-length or truncated protein (tETV1) with a 131-amino acid N-terminal deletion. While this results in the loss of the N-terminal transactivation domain of ETV1, it removes a binding site for the E3 ubiquitin ligase COP1, increasing protein stability. Given the incidence of tETV1 in prostate tumors, any key residues required for oncogenesis are likely retained in the truncated protein. Through our investigations, we have found that expression of tETV1 leads to increased expression of both toll-like receptor 4 (TLR4) and its noncanonical ligand biglycan (BGN). Previous studies have shown that oncogenic transcription factors, including ERG, can activate BGN-TLR4 signaling through a positive feedback loop via the Ras/MAPK pathway. We show that activation of TLR4 by tETV1 results in downstream activity of the MAPK p38, which phosphorylates and activates tETV1 to drive cellular migration and spheroid formation. Consistently, treatment of tETV1-expressing prostate cells with either a TLR4 or p38 inhibitor led to a reduction in transwell migration. We also found that a MAPKAPK (MAPK-associated protein kinase) phospho-null mutant of tETV1, which lacks target MAPKAPK phosphorylation sites, can neither increase BGN expression nor induce migration. Lastly, through RNA sequencing, we identified collagen/integrin signaling as a potential mechanism for MAPKAPK- dependent tETV1 migration. To conclude, phosphorylation by MAPKAPKs is required for tETV1 to drive oncogenic phenotypes via a BGN-TLR4-p38 positive feedback mechanism.

Basic Science

Graduate Student

POSTER #23

PHYSIOLOGICALLY RELEVANT OXYGEN EXPANSION OF HEMATOPOIETIC STEM AND PROGENITOR CELLS IMPROVES IMMUNE CELL RECOVERY FOLLOWING TRANSPLANTATION

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Hematopoietic cell recovery is a critical determinant of patient outcome post hematopoietic cell transplantation. Patients with rapid hematopoietic cell recovery exhibit improved overall and progression-free survival and decreased rates of post-transplant complications. Therapies to augment recovery remain a critical unmet need. As a proportion of total cells transplanted, we demonstrate that donor grafts expanded under physiologically relevant oxygen conditions (1-14% O₂) increase hematopoietic cell recovery compared to grafts expanded at ambient air (21% O₂). Grafts expanded at 1% O₂ in particular show enhanced recovery compared to other physiologically relevant expansion conditions. Mechanistically, human lymphoid progenitors expanded under 1% O₂ display an altered transcriptional profile associated with reduced cellular stress. Donor-derived T cells from 1% O₂ expanded grafts show no functional impairment upon stimulation and reduced activation status compared with donor-derived T cells from non-expanded grafts. In direct functional comparisons, ambient air expanded grafts have reduced capacity to support hematopoietic cell recovery and regenerate organs such as the thymus. Thus, expansion under physiologically relevant oxygen conditions yields a more functional donor graft for transplantation and represents a viable strategy for enhancing hematopoietic cell recovery following hematopoietic stem cell transplantation.

Basic Science

Graduate Student

POSTER #24

CHARACTERIZATION OF A CANCER ASSOCIATED FIBROBLAST POPULATION DERIVED FROM CANCER CELLS IN BRAFV600E COLORECTAL CANCER.

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BRAF mutations are found in ~10% of metastatic colorectal cancer (CRC) cases. *BRAF* is an oncogene in the RAS-MAPK pathway that promotes cell survival and growth. The most frequent mutation is BRAF^{V600E} and these mutations are associated with resistance to standard chemotherapy. The standard of care for patients with refractory metastatic BRAF^{V600E} CRC includes a BRAFV600E inhibitor, Encorafenib, plus an EGFR inhibitor, cetuximab. In a BRAF^{V600E} syngeneic mouse model, we observed an increase in the density of tumor stroma in mice treated with Encorafenib and an EGFR inhibitor as compared to vehicle. In the normal colon, fibroblasts help to maintain homeostasis via secretion of factors that support stem cells at the base of the crypt and induce differentiation as cells move up the crypts. They also play an important role in wound healing by secreting factors that induce Epithelial-to-Mesenchymal transition of nearby cells to reform the epithelium and remodeling the ECM after damage. In cancers, fibroblasts become highly activated and are known as cancer associated fibroblasts (CAFs). CAFs can provide advantages to cancer cells by creating a supportive tumor microenvironment via secretion of tumor promoting factors and remodeling the ECM. To begin to explore the role of CAFs in BRAF^{V600E} CRC, I isolated CAFs from syngeneic BRAF^{V600E} colon tumors that were established by engrafting mouse CRC organoids into the colons of wildtype mice. Unexpectedly, we identified two populations of isolated CAFs, a wildtype population and a population with the mutations found in our organoid model, suggesting that they were derived from the tumor cells. These mutant CAFs are morphologically similar to CAFs. We also confirmed the existence of wildtype and mutant CAFs in our colon tumors using copy number variation analysis of single cell RNA sequencing (scRNA-seq) data from the same model. It has been seen that fibroblasts can be derived from epithelial cells under some conditions, so **we hypothesize that there is a population of CAFs originating from tumor cells in our BRAF^{V600E} CRC model that promotes tumorigenesis.** Through my analysis of the scRNAseq data, I have determined that the mutant CAFs express neural gene signatures whereas the wildtype CAFs are predominantly matrix generating. I am currently working on ways to isolate the two populations of CAFs and aim to investigate the role both CAF populations play in BRAF^{V600E} CRC formation and therapy response.

Basic Science

Graduate Student

POSTER #25

PP2A-B56A AS A CRITICAL REGULATOR OF CELLULAR PLASTICITY IN NSCLC

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Lung cancer is the leading cause of cancer related deaths, with a 5-year survival rate of 23 percent. Non-small cell lung cancer is the most prominent type of lung cancer (85%) with approximately 30% of these patients harboring activating mutations in epidermal growth factor receptor, EGFR. EGFR is a main driver and therapeutic target in NSCLC; however, resistance to anti-EGFR therapies is a rapid and common event. Histological transformation has been identified as a unique mechanism of resistance that occurs in ~15% of NSCLC cases. This type of resistance includes epithelial-to-mesenchymal transition (EMT) and NSCLC to small-cell lung cancer transformation, both of which highlight the cell's ability to circumvent therapeutic intervention by utilizing cellular plasticity.

Protein phosphatases provide critical negative regulation of oncogenic pathways. Protein phosphatase 2A (PP2A) is serine/threonine phosphatase composed of an A (scaffolding), B (regulatory) and C (catalytic) subunit. The diverse family of B subunits determines the target specificity, with >90 complexes possible. In lung cancer, CIP2A, an endogenous inhibitor of the B56 family, is overexpressed and correlates with therapeutic resistance in patients, implicating the dysregulation of the B56 family in therapeutic resistance. PP2A-B56a, regulates many of the downstream effectors of EGFR and has been implicated in the regulation of cellular plasticity; however, its role in EMT and therapeutic resistance is poorly understood. Our data indicate that suppression of PP2A-B56a leads to a morphological shift of epithelial cancer cells to a mesenchymal cell state, with cells displaying increased vimentin and decreased e-cadherin expression, as well as increased migration and invasion, consistent with EMT. Furthermore, these phenotypes can be reversed with PP2A-B56a overexpression. These results implicate PP2A-B56a as a critical regulator of cellular plasticity in EGFR mutant NSCLC and support the use of PP2A activating compounds as a novel strategy to combat resistance mechanisms.

Basic Science

Graduate Student

POSTER #26

CT26 LIVER METASTASES POTENTIATE CACHEXIA IN COLORECTAL CANCER

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Colorectal cancer (CRC) remains a major global health challenge, ranking among the most commonly diagnosed malignancies worldwide and accounting for almost 9% of cancer-related mortality. While tumor progression is a primary determinant of outcome, systemic complications have increasingly shown to contribute to morbidity and reduced therapeutic efficacy. Among these is cancer cachexia, a multifactorial syndrome characterized by progressive skeletal muscle wasting with or without fat loss; representing a major clinical concern. Up to 60% of patients with advanced CRC develop cachexia, which profoundly impairs functional capacity, diminishes tolerance to chemotherapy, and contributes to poor survival outcomes. Despite its clinical importance, there are currently no effective therapeutic options capable of halting or reversing this progressive loss of muscle mass and function. Advanced CRC frequently metastasizes to the liver, and the presence of liver metastases (LMs) exacerbates cachexia. However, despite this association, therapeutic strategies targeting cachexia in the context of metastatic CRC remain largely unexplored, in part due to a lack of viable models. One potential model, CT26, has shown inconsistent cachexia phenotypes in the literature, being reported as both cachectic and non-cachectic when examined subcutaneously. However, whether CT26 LM exacerbates cachexia, as seen in other CRC models like C26 or MC38, remains unexplored. To address this gap, CT26 cells were injected either subcutaneously (s.c.; CT26) to model localized disease or via intrasplenic injection (mCT26) to mimic metastatic spread to the liver. This approach enabled direct comparison of cachectic progression between non-metastatic and metastatic CRC states. Our findings revealed that mice bearing CT26-derived liver metastases exhibited pronounced body weight loss, marked reduction in muscle mass, and strength compared to animals with subcutaneous tumors. Notably, animals with s.c. tumors failed to exhibit overt indices of cachexia. These phenotypic changes were accompanied by molecular signatures consistent with enhanced activation of catabolic pathways in mCT26 tumor hosts, including upregulation of muscle-specific E3 ubiquitin ligases. Systemically, mCT26-bearing mice demonstrated elevated circulating levels of Activin A, a known inflammatory and cachexia-associated mediator, whereas mice with s.c. tumors maintained levels comparable to controls. These findings further suggest that hepatic metastases intensify inflammatory and catabolic signaling, thereby accelerating muscle wasting. Collectively, our model recapitulates key features of metastatic CRC-associated cachexia and provides evidence that liver metastases are not merely a marker of advanced disease but an active contributor to skeletal muscle deterioration. These results highlight the crucial interplay between metastatic burden, systemic inflammation, and muscle metabolism, and underscore the importance of targeting metastasis-driven systemic signaling pathways to preserve muscle function and improve survival in CRC patients.

Basic Science Graduate Student

POSTER #27

THE ROLE OF SSDNA IN ALTERNATIVE DNA DSB REPAIR AND THE OPPORTUNITY FOR THERAPEUTIC INTERVENTION

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DNA double-stranded breaks (DSBs) create genomic instability, a driving factor in cancer development. Targeting DNA repair is a pivotal strategy in cancer therapy by exploiting synthetic lethal interactions with PARP inhibitors in patients deficient in homologous recombination (HR) DSB repair. Despite these advances, the clinical outcomes and recurrence rate for these patients have remained relatively stagnant. Recurrence is a direct result of treatment resistance mechanisms which can include reactivation of HR or the use of alternative end-joining (alt-EJ) DSB repair pathways. Theta-Mediated End Joining (TMEJ) and Single-Strand Annealing (SSA) can be employed to maintain genome stability and elevated expression of proteins involved in these pathways, including PolQ and XPF, have been reported in ovarian and lung cancer. SSA and TMEJ repair pathways involve common steps of resection, homology searching/annealing, and DNA synthesis. However, protection and processing of the ssDNA intermediates has not been addressed. Replication Protein A (RPA) is the major single-stranded DNA (ssDNA) binding protein involved in replication and repair and has been implicated in both TMEJ and SSA repair pathways though definitive involvement and the putative mechanisms have not been elucidated. We propose that RPA impacts TMEJ and SSA dependent DNA DSB repair via binding to ssDNA intermediates. The impact of RPA activity on TMEJ and SSA was assessed via genetic knockdown (KD) of RPA using siRNA in HEK293T and H1299 cell lines. Following siRNA KD, TMEJ and SSA repair activity were measured using a repair pathway specific dual luciferase extrachromosomal reporter assay. Results demonstrate that RPA stimulates both TMEJ and SSA activity as loss of RPA activity resulted in a $\geq 50\%$ reduction repair activity for both pathways. Furthermore, a qPCR based extrachromosomal reporter assay requiring an additional 25bp of synthesis for complete repair was also reliant on RPA. Interestingly, treatment of RPA KD cells with ART558, a PolQ inhibitor, had less of an impact compared the control cells suggesting that cells can use a PolQ-independent mechanism of TMEJ for repair under certain conditions. Additionally, the impact of XPF-ERCC1 activity on SSA repair was considered as XPF-ERCC1's major function is to cleave 3' ssDNA overhangs, a key intermediate that must be processed in SSA. The impact was assessed via SSA repair activity in H1299 XPF-ERCC1 genetic knockout cells compared to the H1299 Cas9 control cells. Loss of XPF-ERCC1 activity resulted in approximately a 6-fold decrease in SSA activity indicating that SSA repair heavily relies of XPF-ERCC1 for ssDNA processing. Collectively, these data establish that RPA and XPF have a stimulatory role in TMEJ and SSA repair. The insights gained from this research can be used to better understand how certain types of cancer modify their DNA repair mechanisms to enhance their chances of survival.

Basic Science

Graduate Student

POSTER #28

INVESTIGATING OXYGEN DEPENDENT EPIGENETIC REGULATION OF HEMATOPOIETIC STEM AND PROGENITOR CELL POTENCY

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Hematopoietic stem cell transplantation (HSCT) is one of the most effective treatments for many blood disorders. However, its success is limited by multiple factors such as graft failure, relapse, and poor survival. Hematopoietic stem and progenitor cells (HSPCs) normally reside in low-oxygen niches in vivo, yet they are often expanded ex vivo at atmospheric oxygen, a higher level than their hypoxic niche, creating a physiological mismatch that may alter their functional potency. Our preliminary data suggests that oxygen tension influences gene expression in human HSPCs and that oxygen-sensitive epigenetic mechanisms, particularly histone modifications, may regulate stem cell potency. This project will investigate how physiological oxygen (1-14%) versus ambient air (21%) shapes epigenetic regulation in both nonmalignant hematopoietic cells and leukemic cell lines. Because histone modifications control chromatin accessibility and gene programs that determine HSPC fate, we utilized epiproteomic profiling of histone modifications and found that HSPCs cultured at 1%, 5%, and 21% oxygen exhibit distinct histone methylation patterns, including changes in repressive marks (H3K9me3, H3K27me3) and an activating mark (H3K4me3). Histone extraction and Western blotting further validated oxygen dependence, with H3K9me3, H3K27me3, and H3K4me3 showing reduced accumulation at higher oxygen tensions. In addition, inhibition of the H3K27me3 demethylase using GSK-J4 produced oxygen-dependent effects on cell proliferation, supporting a functional link between oxygen-sensitive chromatin regulation and HSPC behavior. To extend these findings, we performed CHIP-qPCR to map oxygen-dependent histone modifications at promoters of oxygen-responsive genes and apply immunophenotyping with intracellular histone-mark staining to define histone enrichment across HSPC subpopulations. Together, these studies will clarify how oxygen-dependent epigenetic states regulate HSPC fate and may reveal new strategies to improve HSCT outcomes, with the potential to benefit patients with diverse blood disorders.

Basic Science *Graduate Student*

POSTER #29

HSF1 DOWNREGULATES CCL5 AND IMMUNE-MEDIATED TUMOR KILLING IN BREAST CANCER THROUGH REGULATION OF THE CGAS/STING PATHWAY

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Breast cancer is the second leading cause of cancer-related deaths in women, with a 1 in 8 probability of developing invasive disease over their lifetime. In this cancer, the stress-responsive transcription factor HSF1 plays a central role in driving tumor progression and poor prognosis. Recently, we found that HSF1 influences tumor-immune interactions by suppressing expression of the chemokine CCL5, which is essential for recruiting CD8⁺ T cells and enabling immune-mediated tumor killing. Beyond this, only a few studies have examined HSF1 in the context of immunity, highlighting how little is known about its role in shaping tumor-immune interactions. This project seeks to define the mechanism by which HSF1 regulates CCL5 and therefore, T-cell recruitment. ChIP-seq data indicate that HSF1 does not directly bind the CCL5 gene, suggesting an indirect mechanism. In parallel, my preliminary data show that HSF1 overexpression downregulates the cGAS/STING pathway, a central regulator of innate immune signaling and chemokine production. Based on these observations, our central hypothesis is that HSF1 suppresses CCL5 by inhibiting the cGAS/STING pathway, thereby limiting T-cell infiltration into tumors. Supporting this idea, I recently found that a STING agonist (diABZI) enhances CCL5 upregulation induced by HSF1 knockdown, whereas a STING inhibitor (H-151) attenuates this effect. The mechanism by which HSF1 suppresses cGAS/STING signaling remains undefined, and ChIP-seq data provide little evidence of direct transcriptional regulation of core pathway genes. Given that cGAS/STING activity is tightly controlled by inhibitory factors, I will test whether HSF1 promotes expression of a negative regulator that dampens pathway activation. To address this, I will identify and validate HSF1-dependent negative regulators and determine their effects on chemokine expression, T-cell migration, and tumor growth *in vitro* and *in vivo*. I will also evaluate whether combining HSF1 inhibition with STING agonists enhances T-cell recruitment and suppresses tumor growth in mouse models. By defining a novel link between HSF1 and cGAS/STING signaling, this work will establish a new mechanism of tumor immune evasion and provide a rationale for combination strategies that enhance anti-tumor immunity in breast cancer and other HSF1-driven cancers.

Basic Science

Graduate Student

POSTER #31

INVESTIGATING THE ANTAGONISTIC FUNCTIONS OF SETD8 AND PHF8 IN GLIOBLASTOMA CELL CYCLE REGULATION

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Glioblastoma multiforme (GBM) is the most common malignancy of the central nervous system, with a high recurrence rate of 90-100% and a short survival time of approximately 12-18 months. GBM is classified as a WHO grade IV glioma and consists of the most aggressive, invasive, and undifferentiated type of glioma. The main treatment remains surgical resection followed by concurrent radiotherapy and chemotherapy with temozolomide (TMZ), but options are limited. Thus, there is a critical need to identify new therapeutic targets to improve the survival of GBM patients. Lysine methylation is a reversible post-translational modification on histone and non-histone proteins shown to regulate a variety of cellular processes. Lysine methyltransferases (KMTs) and lysine demethylases (KDMs) add or remove this mark and are frequently dysregulated in many human cancers. Recent reports show the lysine methyltransferase SETD8 (KMT5A or PR-SET7) is overexpressed in a variety of solid and hematological cancers, including GBM, and is associated with poorer prognosis. SETD8 is the only known KMT that mono-methylates histone 4 on lysine 20 (H4K20me1), an important regulator of DNA repair and the cell cycle. PHF8 (KDM7B) is a Jumonji (JmjC) domain-containing lysine demethylase that negatively modulates H4K20me1, opposing the action of SETD8. SETD8 and PHF8 expression are reported to be positively correlated, and disruption of the balance between SETD8 and PHF8 can lead to different levels of H4K20me1, altering cell cycle regulation. The overall objective of this work is to understand the antagonistic functions of SETD8 methyltransferase activity and PHF8 demethylase activity on H4K20me1 in cell cycle regulation and GBM tumorigenesis. We report to our knowledge the first analysis of SETD8 and PHF8 expression during the cell cycle in two GBM cell lines. We also characterize the impact of SETD8 and PHF8 knockdown on GBM cell proliferation. To determine whether SETD8 and PHF8 share common substrates beyond H4K20me1, we performed in vitro methyltransferase assays on fractionated cell lysates, revealing several non-histone substrates of SETD8. Interestingly, in vitro demethylase assays with PHF8 on these substrates results in reduced methylation. Future work aims to identify these proteins, determine how methylation impacts their function, and the impact of these signaling events on GBM cell growth and cell cycle regulation.

Basic Science

Graduate Student

POSTER #32

COOPERATION OF YBX1 AND ETS TRANSCRIPTION FACTORS IN PROSTATE CANCER

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Prostate cancer is the most common cancer in men, accounting for nearly 30% of all cancer cases. Approximately half of prostate tumours are driven by erroneous expression of ERG, a transcription factor in the ETS family. ERG is normally absent in the prostate but can become expressed after a chromosomal rearrangement that fuses *ERG* to the androgen-driven promoter of *TMPRSS2*. In prostate cancer cells ERG functions in complex with the proteins EWS and PABPC1. My preliminary data suggest that YBX1 is also involved in this complex. YBX1 is an RNA- and DNA-binding protein that is involved in many DNA/RNA events, including transcriptional coactivation. YBX1 has been implicated in multiple cancers and is upregulated in ERG-driven prostate cancer, but its role in the ERG-EWS-PABPC1 complex is unknown. This research will improve our understanding of the molecular mechanism behind ETS-driven prostate cancer and will determine if YBX1 is a viable target for prostate cancer therapeutics.

Basic Science

Graduate Student

POSTER #33

DISSECTING SEX CHROMOSOME AND GONADAL INFLUENCES ON CANCER- AND CHEMOTHERAPY-INDUCED MUSCULOSKELETAL WASTING

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Cancer cachexia is a multifactorial syndrome marked by progressive skeletal muscle wasting and bone loss, significantly impacting morbidity and mortality in cancer patients. Individuals with colorectal cancer (CRC) are particularly vulnerable, with up to 60% developing cachexia during their disease and treatment. This condition often arises alongside systemic chemotherapy, which can independently compromise muscle and bone health and exacerbate tumor induced deterioration. Notable sex differences exist in cachexia progression, with male patients typically exhibiting more severe symptoms; however, preclinical studies frequently overlook sex as a biological variable and rarely account for treatment-related toxicity, creating a critical gap in understanding. A central unresolved question is whether these sex differences arise primarily from gonadal hormones, sex chromosomes, or interactions between the two. Traditional male-female comparisons cannot disentangle these factors, leaving the biological drivers of sex-biased cachexia unclear. To address this gap, we utilized the Four Core Genotypes (FCG) mouse model, which independently dissects chromosomal sex (XX vs XY) and gonadal sex (ovaries vs testes), generating four genotypes –XXO, XXT, XYO, and XYT– allowing mechanistic resolution of chromosomal versus hormonal influences on cancer- and chemotherapy-induced musculoskeletal decline. 4.5-month-old FCG (n=2-6/group) mice were subcutaneously injected with MC38 CRC cells and treated with chemotherapy (Folfiri) for 4 weeks. All animals were assessed for changes in body composition, skeletal muscle mass and function, bone microarchitecture, and mechanical strength to determine how chromosomes and gonads shape cachexia susceptibility. Preliminary findings reveal that XXO tumor-bearing mice exhibited reduced body weight compared to their control counterparts, while tumor weight was unchanged across genotypes, indicating no sex-biased tumor growth. As expected, lean mass was consistently lower in gonadal females (XXO, XYO) relative to gonadal males, independent of tumor status. Across gastrocnemius, quadriceps, and tibialis anterior muscles there was a main effect of cancer on muscle mass. Specifically, gastrocnemius mass was reduced in tumor-bearing XYT mice relative to controls. Functional assessments revealed that absolute torque and rate of contraction exhibit a main effect of genotype, whereas the rate of relaxation demonstrated a main effect of cancer. Notably, tumor-bearing XXO mice display reduced rate of relaxation when compared to controls, indicating impaired contractile dynamics. Bone outcomes similarly reflect gonadal influences: control and tumor gonadal females displayed lower total and femur BMD than gonadal males. Mechanical testing further shows decreased pre-yield displacement in tumor-bearing XXO mice, indicating compromised bone material properties. Together, these findings indicate that gonadal sex exerts a dominant influence on lean mass, bone density, and bone material properties, while chromosomal sex contributes to select systemic and functional responses. Although clear chromosome-gonad interactions have not yet emerged, the genotype-specific patterns observed here suggest that such interactions may become evident as additional animals and endpoints are incorporated into the dataset.

POSTER #34

RAN REGULATION OF KINESIN-14 DRIVES CENTROSOME CLUSTERING IN CANCER CELLS

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Centrosomes are the major microtubule organizing centers that facilitate bipolar spindle assembly for the equal segregation of chromosomes during mitosis. Extra centrosomes can result in the formation of multipolar spindles, leading to multipolar divisions and eventual cell death. While centrosome amplification (CA) is a hallmark of aggressive cancers, cancer cells evade multipolar divisions, by clustering their extra centrosomes into a bipolar spindle mediated by Kinesin-14, a microtubule motor protein that crosslinks and slides microtubules through its motor and tail domains. While Kinesin-14 is critical for the survival of cancer cells with CA, it is dispensable in normal cells with two intact centrosomes, making it a potential therapeutic target. In addition to high levels of CA, cancer cells also have elevated genomic content, which leads to increased Ran-GTP levels around chromosomes. In areas where Ran-GTP is high, importins are released from the Kinesin-14 tail allowing it to bind microtubules facilitating microtubule cross-linking and sliding. Thus Ran-GTP gradient spatially regulates Kinesin-14 activity. Analysis of cancer patient and cell line databases revealed point mutations in the microtubule binding domain of Kinesin-14, which also overlaps with its importin binding domain. Biochemical studies of these mutants demonstrated that they reduced importin binding affinity of Kinesin-14 without strongly affecting its microtubule binding. These mutants would be expected to facilitate Kinesin-14 binding to microtubules throughout the spindle, without any importin inhibition which could make them more efficient at centrosome clustering. To test whether these RIS mutants (RIS for Reduced Importin Sensitivity) could reduce multipolarity in cancer cells, we treated cells with paclitaxel to induce multipolarity and found that the RIS mutants were more efficient compared to the wild-type Kinesin-14. This data supports a model in which the Ran-GTP gradient promotes centrosome clustering in cancer cells by spatially enabling Kinesin-14 mediated microtubule crosslinking and/or sliding. To determine if Kinesin-14 mediated sliding is necessary for clustering the extra poles, we utilized a Kinesin-14 mutant (N593K) that has its motor-mediated microtubule sliding activity uncoupled from its crosslinking activity. Knockdown/rescue studies in cancer cells treated with paclitaxel revealed that the N593K mutant reduced multipolarity as efficiently as the wild-type protein, suggesting that Kinesin-14 mediated MT sliding is not necessary for clustering the extra poles. This data further suggests that the crosslinking function of Kinesin-14, rather than its sliding ability, is critical for clustering the extra poles, which contradicts existing models of Kinesin-14 mediated centrosome clustering. Together, these findings provide a foundation for understanding how the Ran-GTP gradient may regulate Kinesin-14 mediated centrosome clustering and the molecular mechanisms by which Kinesin-14 reduces spindle multipolarity. A mechanistic understanding of this process could enable the design of new therapeutic strategies for aggressive cancers with high levels of CA.

Basic Science

Graduate Student

POSTER #35

TRYPTOPHAN METABOLISM PLAYS A SIGNIFICANT ROLE IN MULTIPLE MYELOMA SURVIVAL

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Introduction

Multiple myeloma (MM) is a hematological malignancy caused by abnormally proliferating plasma cells in the bone marrow. It is considered incurable, as patients almost always go through cycles of treatment, remission and treatment-resistant relapse. Early-stage MM depends on the Bone Marrow Microenvironment (BMME) for survival. We have previously shown that MM interacts with BMME Dendritic Cells (DC) which induces DC-production of Indolamine Dioxygenase 1 (IDO1), which catabolizes tryptophan (TRP) to kynurenine (KYN). The production of KYN activates pro-survival pathways in MM through the activation of the transcription factor Aryl Hydrocarbon Receptor (AHR). We now show that MM cells can produce KYN independently of DCs through expression of the TRP metabolizing enzyme TDO.

Methods

Patient RNA expression data were taken from the CoMMpass database. We measured MM cell lines U266, 8226, MM1S and KMS11 for expression of TRP metabolizing enzymes with western blot and qPCR. TRP metabolizing enzyme Tryptophan 2,3-Dioxygenase (TDO) was knocked down with shRNA or inhibited with TDO-specific inhibitor 680C91. AHR activation was measured through qPCR of CYP1a1, a transcriptional downstream target. KYN production was measured by ELISA. KMS11 cells were treated with 680C91, then underwent bulk RNAseq and were analyzed using gene set enrichment analysis.

Results

CoMMpass patients with the highest quartile of TDO expression have significantly lower rates of progression-free and overall survival. Three of our cell lines 8226, MM1S and KMS11 express TDO, while U266 does not. Inhibiting TDO with the non-competitive inhibitor 680C91 or knocking down TDO significantly reduced MM cell survival in TDO+ MM cell lines. Inhibiting TDO reduced KYN production. Inhibiting TDO also reduced AHR activation as shown by expression of downstream target CYP1a1 in TDO+ MM cell lines, but did not affect a TDO- MM cell line. Treatment with AHR ligand TCDD rescued MM cell viability from TDO inhibitor-induced cell death, indicating that TRP metabolism to KYN is important to MM survival. RNAseq with KMS11 cells revealed that cells treated with TDO inhibitor 680C91 had significantly reduced expression of genes involved in MYC signaling, AHR signaling, cholesterol homeostasis and fatty acid metabolism; 680C91-treated KMS11 cells had upregulated expression of genes involved in TNF α signaling through NF κ B, hypoxia and apoptosis.

Conclusions

MM depends on TRP metabolism for survival both in the BMME and as it becomes independent of it. MM cells express TDO, which supports MM survival through the activation of AHR and could repress T effector activation through the depletion of TRP. We now show evidence that TRP metabolism in MM also supports survival through MYC signaling and through upregulation of metabolic pathways. TRP metabolism is a novel treatment target in MM and could lead to more effective cell killing and immunotherapy, especially in relapse/refractory disease.

POSTER #36

DISSECTING THE ROLE OF PRDM9 METHYLTRANSFERASE ACTIVITY ON DOUBLE-STRAND BREAK REPAIR

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DNA repair is a foundational biological process critical for maintaining genomic stability over iterations of DNA replication. Due to the centrality of DNA repair pathways in maintenance of the genome, their disruption is often implicated in cancer development through mutations in oncogenes and tumor suppressor genes. Additionally, DNA repair machinery can be exploited as a therapeutic target to increase the susceptibility of tumor cells to DNA-damaging agents. Despite the importance of understanding DNA repair pathways in preventing and treating cancer, key mechanistic aspects of DNA repair remain unresolved. One such aspect is how cells mediate which repair pathway is chosen when correcting DNA double strand breaks (DSBs). DSBs are primarily repaired by two pathways: the high-fidelity homologous recombination (HR) pathway and the error-prone nonhomologous end joining (NHEJ) pathway. PRDM9, a lysine methyltransferase enzyme that modifies histone tail residues and is frequently altered in cancers, has been demonstrated to perform a critical role in facilitating HR repair. Previous studies have shown that fusing PRDM9 to the Cas9 nuclease increases the ratio of HR repair to NHEJ repair observed after a Cas9-induced DSB. A potential explanation for this observation is that trimethylation of histone residue H3K36 biases DNA repair towards the HR pathway when repairing a DSB. To investigate this hypothesis, mutants of PRDM9 were created with substitutions to residues critical for mediating the binding of its two histone substrates, H3K4 and H3K36. Scintillation proximity assays confirmed that mutating specific PRDM9 residues alters the ratio of H3K4 trimethylation (H3K4me3) to H3K36 trimethylation (H3K36me3). Variants of PRDM9 that display altered histone substrate specificity will be fused to Cas9. We plan to observe alteration in the HR:NHEJ ratio following DSB induction via a GFP-to-BFP reporter assay in HEK293 cells. Briefly, a GFP reporter gene is targeted with the Cas9-PRDM9 construct and a single-stranded oligodeoxynucleotide (ssODN) repair template encoding a point mutation that shifts fluorescent expression in the reporter from GFP to BFP. If the DSB is repaired from the ssODN template by HR, cells will fluoresce blue. Repair from the NHEJ pathway frequently introduces indels, knocking out the fluorescent reporter. Levels of blue fluorescence to fluorescence knockout can be used to infer the occurrence of HR and NHEJ, respectively. We plan on selecting PRDM9 mutants with altered HR:NHEJ ratios and performing further characterization of histone trimethylation to connect repair pathway outcomes to histone methylation patterns. This work would enhance the current understanding of DSB repair and expand the utility of gene editing and cancer therapy applications.

Basic Science

Graduate Student

POSTER #37

DISSECTING THE ROLES OF PARP1 CATALYTIC INHIBITION AND DNA TRAPPING IN PARP INHIBITOR CYTOTOXICITY

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DNA replication is a dynamic process defined by several closely coordinated interactions between key enzymes and sufficient plasticity to respond to diverse cellular stressors. One enzyme responsible for the direct detection of damaged DNA and unprocessed replication intermediates is poly(ADP ribose) polymerase 1 (PARP1). PARP1 is an established chemotherapeutic target for the treatment of BRCA-null cancers, as its inhibition results in increased cellular death in this genetic background due to dysregulation of Okazaki fragment maturation (OFM). Despite strong initial treatment responses, most tumors develop resistance, limiting long term therapeutic benefit. PARP inhibitors (PARPi) exert their effects through both catalytic inhibition and stabilization of PARP1 on DNA, often referred to as PARP1 trapping; however, the relative contribution of these mechanisms to dysregulation of OFM remains unclear. Here, we aim to clarify how the biochemical activities of PARP1 regulate the minimally essential OFM enzymes (DNA polymerase δ , FEN1, and LIG1) and how pharmacological inhibition contributes to the initial cytotoxic response. Using recombinant human proteins and purified DNA substrates that mimic OFM intermediates, we examine how PARP1 activity affects the biochemical activities of individual OFM enzymes *in vitro*. Our reconstituted assays reveal that PARP1 activity influences the efficiency of OFM intermediate processing *in vitro* and that genetic perturbations designed to separate PARP1 catalytic activity from DNA trapping indicate that PARP1 trapping is a stronger contributor to this dysregulation. These findings provide insight into how PARP1 regulates OFM and may help clarify the molecular basis of PARP inhibitor cytotoxicity in BRCA-null cancers.

Basic Science

Graduate Student

POSTER #38

COMPLEMENTARY MECHANISMS OF ENZALUTAMIDE AND PARP INHIBITORS DRIVE COMBINATION EFFICACY IN PROSTATE CANCER

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Prostate cancer is the most common malignancy and the second leading cause of cancer-related deaths in men in the United States. Disease progression is driven by androgens and the androgen receptor (AR) signaling axis, making most cases initially sensitive to androgen deprivation therapy. However, despite initial effectiveness, tumors frequently recur as castration-resistant prostate cancer (CRPC), a highly lethal form that remains AR-dependent. Although patients with CRPC initially respond to AR antagonists such as enzalutamide (ENZ), nearly all develop treatment-resistant disease through mechanisms including the emergence of AR mutations, AR variants, and homologous recombination repair mutations (HRRm). Poly (ADP-ribose) polymerase inhibitors (PARPis) target HRRm tumors by trapping PARP at sites of DNA damage, leading to replication fork collapse and cell death. However, recent clinical trials combining ENZ with the PARPi talazoparib demonstrated improved outcomes in patients lacking HRRm, suggesting additional mechanisms that remain unclear. Utilizing HRRm-free prostate cancer cell models, we show that PARPis suppress basal AR transcriptional activity, whereas ENZ primarily inhibits androgen-induced AR signaling. Combined ENZ and PARPis treatment further suppress AR activity and decreases cell viability compared to either agent alone. Notably, knockout or overexpression of PARP1, PARP2, and PARP7 does not attenuate PARPi-mediated AR suppression, indicating that this effect occurs independently of PARP inhibition or trapping. While PARP1 loss shifts the IC₅₀ values for PARPis, the combination treatments remain effective. Collectively, these findings demonstrate that PARPis modulate AR signaling through PARP-independent mechanisms that complement AR inhibition by ENZ. These data further provide mechanistic insight into the clinical efficacy of ENZ and PARPis combination beyond patients harboring HRRm.

Basic Science

Graduate Student

POSTER #39

THE TRANSCRIPTION FACTOR ZEB1 CONTROLS DIFFERENTIATION PROPERTIES OF AFRICAN-ANCESTRY ENRICHED MULTIPOTENT STROMAL CELLS AND THE BREAST MICROENVIRONMENT.

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Disparity in breast cancer outcomes in women of African Ancestry (AA) as compared to European Ancestry (EA) has been thought to derive from socioeconomic factors. However, emerging evidence demonstrating genetic ancestry-dependent differences in normal and cancer genome, and immunosuppressed tumor microenvironment suggests the involvement of biological factors. To investigate breast cancer outcome-associated biologic factors, we previously analyzed breast tissues of healthy donors of distinct genetic ancestry. We found that PROC⁺/ZEB1⁺/PDGFR α ⁺ stromal cells (hence, called PZP) are more abundant in normal breast tissues of women of AA as compared to EA. PZP cells display the properties of multi-lineage fibroadipogenic mesenchymal stromal cells that can differentiate into adipogenic and osteogenic lineages.

In this study, we examined the role of PZP cells in the context of breast cancer initiation and progression. Analysis of single cell nucleus atlas data of normal and cancerous breast tissues demonstrated co-expression of *PROC*, *ZEB1* and *PDFRA* genes in a fraction of fibroblasts, which also overlapped with a myofibroblastic gene signature. Consistent with immunosuppressive role of myofibroblasts, secreted factors from PZP cells reduced the fraction of Granzyme B and TNF α expressing CD8⁺ T cells in an *in vitro* T cell activation assay. In our *in vivo* studies with an Estrogen Receptor positive TMCF7 breast cancer model, we found that injecting PZP cells with TMCF7 cancer cells significantly increased the tumor volume, and the percentage of tumor-infiltrating Arginase 1-positive M2-like immunosuppressive macrophages as compared to tumors generated with TMCF7 cells alone in nude mice.

To target PZP cells, we focused our efforts on Zinc finger E-box-binding homeobox 1 (ZEB1), which is a master transcription regulator of stemness, and epithelial-to-mesenchymal transition (EMT) expressed in PZP cells. We generated *ZEB1* gene knockout clones of PZP cells using CRISPR-Cas9 and performed RNA-sequencing of *ZEB1* knockout clones and parental cells. The expression of epithelial markers KRT 7, 8, 18 and adipogenic markers PPARG and PPARGC1B was upregulated upon *ZEB1* knockout. Additionally, *ZEB1*-deficiency increased the levels of inflammatory fibroblast markers CXCL1, IL6 and IL1B, while downregulating the expression of myofibroblast markers encoding extracellular matrix proteins Tenascin-C and matrix metalloproteinase 1. *ZEB1* knockout also downregulated osteogenic differentiation and upregulated adipogenic differentiation of PZP cells, as determined by differentiation assays. These results suggest the role of *ZEB1* in lineage specificity and cell fate conversion of PZP cells. Ongoing studies are investigating the potential inhibitors to reverse immunosuppressive effects of PZP cells on CD8 T cells and extracellular-matrix inhibitors to target PZP cells in the breast tumor microenvironment. Thus, we identified a novel role of *ZEB1* in stromal PZP cells and successful targeting PZP cells will provide a stroma-based therapies against breast cancer.

POSTER #40

TARGETING TRANSLATION INITIATION AND ELONGATION PROCESSES IN PROSTATE CANCER THERAPIES

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Prostate cancer (PCa) is the second leading cause of cancer-related deaths in men in the United States. The androgen receptor (AR) is a critical driver of PCa growth, and while anti-androgen therapies are the current standard of care, resistance occurs in advanced PCa, highlighting the need for novel treatment approaches. Our lab has shown that PCa cells rely on constitutive activation of GCN2 protein kinase and the integrated stress response (ISR) for the maintenance of amino acids (AA) and sustained proliferation (Cordova et al., *eLife*, 2022). In response to nutrient depletion, GCN2 phosphorylation of eIF2 regulates translation initiation, enhancing expression of AA transporters and biosynthetic genes that are critical for the maintenance of AAs that fuel PCa progression. Pharmacological or genetic inhibition of GCN2 reduces intracellular AAs and greatly decreases the growth of PCa cells in culture and xenograft models. These results suggest that targeting translation initiation processes through GCN2 and the ISR can be an effective therapeutic strategy for starving PCa of critical nutrients.

Recently, we showed that alternate-day fasting (ADF) can be an effective strategy for enhancing anti-androgen therapy in PCa (Cordova et al., *Cancer Research*, 2025). We found that xenograft PCa tumors from mice fed an ADF diet exhibited reduced tumor growth and AR protein levels compared to a control diet. Of importance, the effects of caloric restriction on cell growth and AR protein levels were recapitulated in both 2D and 3D PCa culture models. Although AR mRNA translation is reduced during nutrient starvation, the underlying mechanism does not appear to involve impaired translation initiation, but rather unproductive translation elongation. Our results suggest that intracellular AA depletion causes ribosomes to stall on long stretches of synonymous codons in the AR coding sequence, including contiguous polyglutamine, polyglycine, and polyproline stretches.

Ongoing research in the lab involves targeting metabolic processes that will impact ribosome stalls on these AR coding regions, and how GCN2 inhibition can augment this to reduce PCa viability. In particular, treatment of PCa cells with halofuginone, a compound that induces ribosome stalls on proline codons, reduces AR protein levels and function. In addition, halofuginone treatment in PCa cells enhances the growth defect of enzalutamide, a standard of care AR antagonist. Overall, our study highlights how nutrient availability can impact both translation initiation and elongation processes, which are crucial for effective therapies of both early- and late-stage PCa.

Basic Science

Graduate Student

POSTER #41

LOSS OF ARID1A IN GRANULOSA CELLS AS A DRIVING EVENT FOR OVARIAN CARCINOMA

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INTRODUCTION:

Endometriosis is a chronic inflammatory disease characterized by the ectopic growth of endometrial-like tissue, most often on the ovaries. Endometriosis shares multiple hallmarks with cancer, including recurrent mutations in cancer-driving genes such as *ARID1A*, *PTEN*, and *KRAS*. Importantly, women with ovarian endometriosis have a 19-fold increased risk of developing endometrioid and clear-cell ovarian cancer, malignancies in which *ARID1A* loss-of-function mutations are highly prevalent and considered an early driver of tumorigenesis.

ARID1A encodes a key subunit of the SWI/SNF chromatin-remodeling complex and functions as a tumor suppressor, regulating transcriptional programs involved in proliferation, differentiation, and hormone signaling. While *ARID1A* loss has been extensively studied in epithelial tumor cells, its role in ovarian granulosa cells, which are critical regulators of folliculogenesis and steroidogenesis, remains poorly defined. Given the central role of granulosa cells in shaping the ovarian hormonal microenvironment, *ARID1A* dysfunction in these cells may contribute to altered follicular signaling, including overproduction of estradiol, and a pro-tumorigenic milieu, associated with endometriosis-driven ovarian cancer.

This study aims to determine how *ARID1A* loss in granulosa cells affects folliculogenesis-associated pathways and cellular behaviors relevant to ovarian tumorigenesis.

METHODS:

To investigate the functional consequences of *ARID1A* loss, we used the human granulosa cell tumor cell line, KGN, and the immortalized human granulosa cell line, hGrC1. After viral packaging, cells were transduced with a non-targeting (shNT) or 2 independent *ARID1A* (sh*ARID1A1*, sh*ARID1A2*) short-hairpin RNA molecules. Cells underwent selection in puromycin. GFP-positive populations were sorted using flow cytometry. After confirmation of *ARID1A* knockdown, cellular phenotypes were assessed under conditions with and without follicle-stimulating hormone.

RESULTS:

Successful transduction of the lentiviral system in KGN and hGrC1 was confirmed by EVOS imaging, and GFP positive cells were sorted. Immunofluorescence studies conducted in KGN-sh*ARID1A1*-GFP cells showed decreased expression of *ARID1A* when compared to KGN cells. KGN-sh*ARID1A1*-GFP cells revealed a significantly higher cell proliferation rate at t=72 hours and t=96 hours when compared with KGN

cells (n=8, p<0.01). KGN-shARID1A1-GFP cells showed wound closure from 400µm to 0µm in 9 hours compared to 600µm to nearly 400µm in KGN cells after 9 hours (n=18, p<0.001).

CONCLUSIONS:

Loss of the putative tumor suppressor, ARID1A, in KGN cells may lead to more aggressive cellular behaviors. KGN cell represent a granulosa cell tumor cell line. Future studies will evaluate the role of loss of ARID1A in the non-luteinized immortalized ovarian granulosa cell line, a non-cancerous cell line.

Basic Science *Graduate Student*

POSTER #42

LEVERAGING P53 AND MCL-1 AS THERAPEUTIC TARGETS IN SMALL CELL LUNG CANCER

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Background:

Small cell lung cancer (SCLC) is a neuroendocrine carcinoma known for its acquired chemoresistance. While chemoresistance is common after platinum-based strategies, lurbinectedin, or bispecific T-cell engagers, a need remains for novel therapeutic strategies in relapsed SCLC. Importantly, SCLC harbors near ubiquitous alterations in TP53, a historically undruggable target. New approaches with p53 reactivators, APR-246 and rezatapopt, can stabilize the conformation of mutant p53 to induce apoptosis. Here, we seek to explore their use, alone, and in combinations, for SCLC.

Methods:

For drug response, 1,000 cells were incubated for 24 hours (h) at 37°C. APR-246 (0.39-50 µM), rezatapopt (0.078-10 µM), or DMSO was added for 120 h, prior to evaluation of cell viability (CellTiter-Glo®). H82 and H524 were seeded in 384-well plate using Bravo automated liquid handling platform. Following 24 h, a 500-drug library screen was performed with 4 doses (0.1, 0.5, 2.5, 10 µM) of compounds +/- 2 µM APR-246 using Echo650. After 5 days, cell viability was measured for luminescence. To evaluate APR-246 effects, 1 X 10⁶ cells were incubated for 24 h, exposed to drug (1 or 5 µM APR-246, or DMSO) and harvested at 24 or 48 h. For western blot, standard lysate preparation was used. Membranes were incubated in Every Blot Blocking Buffer with 1:500-1000 primary antibody (0.75 h) followed by 1:5000 secondary Ab (0.5 h). Four SCLC lines were chronically exposed to increasing doses (up to 75 µM) of APR-246 to generate “APR-R”. RNA sequencing (RNA-seq) was performed on extracted RNA with an Illumina array.

Results:

Within a panel of SCLC, most SCLC lines demonstrated sensitivity to APR-246 as a single agent (IC₅₀ range, 1.04-3.77 µM), compared to a p53 homozygous null line H1299 (IC₅₀ >50 µM). In the context of TP53 alterations, no difference in APR-246 IC₅₀ was observed (P = 0.23). Across neuroendocrine (NE) subtypes (SCLC-A, SCLC-N, and SCLC-P), no difference in APR-246 sensitivity was seen (P = 0.86). Similarly, H748, a p53 Y220C mutant SCLC line, is sensitive to both rezatapopt (IC₅₀, 0.39 µM) and APR-246 (IC₅₀, 1.52 µM). Proteomics of H82 and H524 APR-R (compared to parental lines) both demonstrated overexpression of midkine (MDK) and nephronectin (NPNT). RNA-seq confirmed transcriptional activation of MDK and NPNT in APR-R versus parental lines. Interestingly, an ASCL1+ SCLC H146 which is less sensitive to APR-246 (IC₅₀, 6.28 µM) was found to have overexpression of BCL-2 protein, when compared to other SCLC tested. A 500-drug screen in conjunction with APR-246 demonstrated a class effect with APR-246 and MCL-1 inhibitors (MCL-1i).

Conclusions:

MCL-1i with p53 reactivators is a promising therapeutic combination for SCLC, independent of NE subtype or TP53 alteration. Mechanistic understanding of how BAX/BAK homeostasis may regulate this response in SCLC is underway.

Basic Science

Graduate Student

POSTER #43

DELINEATING THE BIOCHEMICAL ROLE OF HUMAN PRIMASE AND POLYMERASE PRIMPOL IN DNA LESION BYPASS AND REPLICATION STRESS TOLERANCE

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DNA replication is a dynamic process requiring tight regulation of the proteins involved to ensure high fidelity. DNA damage can result in replication fork stalling and uncoupling of helicases and polymerases, that leads to the formation of long stretches of single-stranded DNA (ssDNA). These ssDNA intermediates are protected from degradation by eukaryotic ssDNA binding protein Replication Protein A (RPA). RPA acts as a critical platform to recruit DNA damage response (DDR) and repair proteins such as the ATR kinase. Prolonged replication stress can lead to fork collapse and formation of double stranded breaks, resulting in genomic instability. To overcome stalled forks, cells employ DNA damage tolerance pathways to restart the replication fork. One of these pathways involves PrimPol mediated repriming downstream of the lesion to restart replication fork. PrimPol is a primase polymerase enzyme possessing the ability to synthesize RNA and DNA primers. Additionally, it is a low-fidelity polymerase lacking 3'-5' exonuclease activity that can elongate de novo as well as pre-existing primers. It has been reported to function as a translesion DNA synthesis (TLS) polymerase bypassing lesions including 8-Oxo-2'-deoxyguanosine, O6-methylguanine and 1,2-intrastrand cisplatin cross-links. In vivo studies have shown PrimPol mediated repriming downstream of DNA lesions as an important mechanism to tolerate DNA damage under replication stress. However, this phenomenon is yet to be characterized in vitro. Existing studies have shown that RPA plays a role in recruiting PrimPol to stalled fork, the mechanistic details of which are yet to be determined.

The objective of this study is to delineate the role of PrimPol in DNA lesion bypass and to investigate the determinants governing its choice between TLS and repriming. We further aim to assess the impact of RPA on PrimPol activity and pathway preference. To address these questions, recombinant human PrimPol was purified using affinity chromatography. The effect of RPA on PrimPol activity is being examined through in vitro polymerase assays employing primer-template substrates with varying lengths of exposed single-stranded DNA template in the presence and absence of RPA. To evaluate the influence of DNA damage on pathway choice, substrates harboring distinct lesions, including abasic sites, PARP inhibitor-induced lesions, and cisplatin adducts are being utilized. The relative contributions of PrimPol-mediated repriming versus TLS are being assessed using stalled fork substrates, where primer extension reflects TLS activity and gap induction reflects repriming. Repriming activity is specifically interrogated using substrates with chain-terminating ddNTP-blocked primers to prevent extension from pre-existing termini. Together, these approaches are designed to dissect the mechanistic basis of PrimPol's dual functionality and the regulatory influence of RPA on its activity and pathway selection. This study aims to advance our understanding of replication stress management while also revealing mechanistic insights into how cells tolerate DNA damage during replication.

Basic Science Graduate Student

POSTER #44

NOISE-TOLERANCE SPATIAL MULTI-OMICS INTEGRATION IN SEMI-METRIC SPACE

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Emerging spatial multi-omics technologies, including spatial transcriptomics, proteomics, metabolomics, and imaging, are reshaping our understanding of biology and diseases. However, interpreting the biological and pathological mechanisms by integrating spatial-temporal pieces of spatial multi-omics across diverse resolutions, noises, and scales remains a prominent challenge.

Here, we present a fused partial Gromov-Wasserstein approach to map spatial multi-omics across biological scales within theoretically guaranteed semi-metric measurement. Different from existing Optimal Transport works using distance-like divergence, this metric-based approach aligns multi-scale and multi-level biological structures with accuracy and precision, tolerant to inherent technical and biological noises. In addition, the proposed approach shows magnitudes of computational efficiency over classical Optimal Transport tools, making it feasible for large-scale spatial omics studies.

In spatial transcriptomics studies on colorectal cancer and chronic kidney disease, the proposed approach faithfully enhances the dissecting of the tumor microenvironment and glomeruli structures by mapping between imaging-based 10X Visium and sequencing-based 10X Xenium samples, which mitigates resolution and scale discrepancies and corrects biases from classical works. Across multiple omics in the striatum of human brains, this approach accurately projects dopamine metabolites onto dopamine-enriched regions annotated by transcriptomics. In breast cancer studies with spatial proteomics, this approach outperforms existing prediction approaches in diagnosis tasks across multiple data sources by describing discrepancies among structural details and bypassing classical pooling steps, which shows potential in translational medicine.

Basic Science

Graduate Student

POSTER #45

PROTEOMIC CHARACTERIZATION OF DIS3 MUTATIONS IN MULTIPLE MYELOMA USING OLINK EXPLORE 3072 AND MASS SPECTROMETRY WITH TMT-BASED PROTEIN QUANTITATION

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Introduction: Multiple myeloma (MM) is a malignant plasma cell disorder characterized by abnormal monoclonal antibody production and dysregulated cell growth within the bone marrow. This malignancy is genomically complex as it has many genetic mutations. DIS3 is a secondary genetic mutation in myeloma arising during tumor progression, and it's one of the most frequently mutated genes with an occurrence of 10% in patients. Mutations in DIS3 recurrently occur at amino acid residues D479, D488, and R780 which are known as hotspot mutations located in the DIS3 protein catalytic active site. DIS3 serves as one of the catalytic ribonuclease subunits of the RNA exosome, which is responsible for RNA processing and degradation. Our lab has shown that DIS3 mutations cause widespread proteomic dysregulation; however, mass spectrometry is not able to detect low-abundance proteins. To account for this, we plan to use affinity-based proteomic platforms such as Olink Explore to supplement global mass spectrometry analysis to further explore low abundance secretory proteins and cell surface markers in DIS3-mutated cell lines.

Methods: Patient data from the Multiple Myeloma Research Foundation (MMRF) and IUSM (n=1,349) was used to determine the most frequently occurring hotspot mutations which were D478G, D488N, and R780K. Endogenous DIS3 was mutated in the KMS11 MM cell line using CRISPR-Cas9 homology directed repair, and cells were screened using Sanger sequencing for the correct missense mutations. Samples were then collected in triplicate and prepped for TMT-labeled mass spectrometry and Olink Explore 3072. Proteomic datasets from both platforms will be analyzed to determine significant differentially abundant proteins (p -value <0.05 , \log_2 fold change $>|0|$, NPX $>|0|$) compared to the DIS3 control.

Results: Olink detected upwards of 300 more significant proteins than mass spectrometry alone. Furthermore, Olink revealed enhanced sensitivity for low-abundance secretory proteins, such as TNF and IL15, and cell surface markers including LY9 and HLA-DRA. Other low abundance proteins like MPHOSPH8 were also detected in the Olink assay. Using all differentially abundant proteins found in Olink and mass spectrometry, we will perform downstream analysis including protein-protein interaction network and functional enrichment analysis. The secretory proteins demonstrating the largest significance ($p<0.05$) in abundance change will be validated using Western blotting, whereas cell surface markers will be validated using flow cytometry.

Conclusion: Integrating both Olink affinity-based and mass spectrometry proteomic approaches provides a higher detection of protein coverage and further guides future validation studies. Our findings support the utilization of combined proteomic platforms when characterizing mutation-associated molecular changes which allows for potential biomarker identification.

POSTER #46

HIGH-FAT DIET EXACERBATES HELICOBACTER PYLORI-ASSOCIATED METAPLASIA PHENOTYPES

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Gastric adenocarcinoma is a malignant tumor originating from epithelial cells in the stomach. This disease is one of the deadliest major cancers, in part because of the difficulty of early diagnosis. Stomach infection and obesity are two important risk factors for gastric cancer, both related to chronic inflammation. Stomach infection with the bacterium *Helicobacter pylori* (*Hp*) is widespread and is considered the primary risk factor for gastric cancer development, driving chronic inflammation, metaplasia (pre-cancer), and ultimately malignancy. While obesity is an established gastric cancer risk factor, the mechanistic basis for this association is not well understood. We previously established “*Hp*+KRAS+” transgenic mice as a clinically relevant model of *Hp*-associated metaplasia. Mice are infected with *Hp* or mock-infected, and tamoxifen is used to induce an oncogenic KRAS allele (G12D) in the stomach. Within six weeks, *Hp*+KRAS+ mice have severe inflammation, metaplasia, and mild dysplasia (abnormal cells). To test the impact of diet-induced obesity on these phenotypes, weanlings were fed a high-fat diet (HFD) or a standard diet. After three months, mice were administered tamoxifen and infected with *Hp* or mock-infected, then euthanized after 6 weeks. In mock-infected mice, diet-induced obesity did not impact inflammation or metaplasia marker expression. However, in *Hp*+KRAS+ mice, diet-induced obesity elicited greater inflammation, including upregulation of the cytotoxic T cell marker *Cd8a* and its effector cytokine *Ifng*. As well, these mice had increased metaplasia markers, including *Muc4* and *Areg*, and the stomach tissue was thicker in these mice, suggesting hyperplasia. Intriguingly, *Hp* bacterial loads were greater in the mice with diet-induced obesity despite the increased inflammation in this group. Together, these results suggest that high-fat diet-induced obesity itself does not cause significant immunopathology in the stomach, but may exacerbate *Hp*-driven disease phenotypes. Future work will include induction of weight loss via caloric restriction or medication and depletion of cytotoxic T cells to determine whether these interventions are protective.

Basic Science

Graduate Student

POSTER #47

INVESTIGATING THE CONSEQUENCES OF NF2 LOSS ON NEURAL PROGENITOR DIFFERENTIATION

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Background: NF2-related schwannomatosis (NF2) is a tumor predisposition syndrome caused by mutations of the NF2 gene and characterized by the development of central nervous system tumors. Half of patients with NF2 will develop spinal ependymomas throughout their lifetime, and this tumor can cause weakness, pain, and genitourinary symptoms due to its location. There are currently no medical therapies for spinal ependymoma, and surgery remains the standard of care for this tumor. Yet, surgical resection is also associated with high morbidity due to the complex location of the tumor. With previous studies suggesting that spinal ependymomas may arise from neural progenitor cells, we hypothesize that mutations in the NF2 gene may prevent normal neural differentiation.

Methods: We performed NF2 CRISPR knockouts in neuroepithelial stem cells isolated from the hindbrain of a human embryo. These cells were cloned out to select for true knockouts and validated with western blot and PCR.

Results: Our preliminary results show that these NF2-mutant cells do not undergo normal differentiation, as they retain neural progenitor morphology and gene expression. Furthermore, NF2-mutant cells form clusters in vitro which appear to expand over time. Bulk RNA sequencing shows several upregulated pathways in the NF2-mutant cells, including JAK-STAT and epithelial to mesenchymal pathways.

Discussion: Our findings suggest that NF2 may be required for normal neural progenitor differentiation, and that mutations may prevent these cells from giving rise to mature neurons and glia. Given the known role of NF2 loss in tumorigenesis, this model may serve as a platform to study spinal ependymoma development and identify targets for potential therapies.

Basic Science *MSTP Student*

POSTER #48

PD-1 AND VEGF PEPTIDE VACCINE COMBINATION DISPLAY SYNERGISTIC ANTI-TUMOR EFFECT IN CANCER IMMUNOTHERAPY

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Cancer immunotherapy with checkpoint inhibitors (PD-1, CTLA-4, etc.) have had promising effects on suppressing tumor growth in numerous cancers. Programmed death protein 1 (PD-1) is a receptor commonly expressed on T cells where it plays a key role in maintaining self-tolerance and preventing autoimmunity. In a normal functioning immune system, when PD1 ligand (PD-L1) binds PD-1 receptor, T cell activation is suppressed. Meanwhile, vascular endothelial growth factor (VEGF) is a growth factor secreted by tumor cells that promotes angiogenesis, a key requirement by which tumors grow and invade surrounding tissue. In this study, we aim to exploit the immunological roles of PD-1 and VEGF to promote further T cell activation and inhibit angiogenesis in colon tumors, respectively. By administering the combination of B cell epitopes PD-1 and VEGF peptide vaccines in mouse models, we hypothesize the host immune system will elicit a response by generating neutralizing antibodies to the B cell epitopes in a synergistic manner. To test this, BALB/c mice were immunized with the PD-1/VEGF combination three times and subsequently challenged with the CT26 colon cancer cell line. Antigenicity and immunogenicity were monitored by collecting serum and performing enzyme-linked immunosorbent assay (ELISA). Tumor growth was monitored for 2 weeks and mice survival rates were tracked as well. Results indicate increasing immunogenicity upon subsequent immunizations, along with slower tumor growth compared to controls. These data suggest the combination of PD-1 and VEGF peptide vaccines synergistically enhance survival outcomes by slowing tumor growth and hold a strong potential as an applicable treatment strategy for colon carcinomas. These findings support further investigation of vaccine efficacy in other cancer models.

Basic Science Medical Student

POSTER #49

GLOBAL PROTEOME AND KINOME ANALYSIS OF HUMAN ADAMANTINOMATOUS CRANIOPHARYNGIOMA REVEALS UPREGULATION OF HER2

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Adamantinomatous craniopharyngioma (ACP) is a highly morbid disease with no medical therapies. Although benign, ACP and subsequent skull base surgery and radiation can cause permanent vision loss, panhypopituitarism, and hypothalamic injury. There is an unmet need for novel therapeutics to minimize morbidity, prevent spread into sensitive areas, and shrink tumors away from fragile structures before surgical resection. Kinase inhibitors are widely used in cancer treatment, but their application in ACP is limited by a limited knowledge of upregulated kinases. No study had directly probed the kinome of ACP; however, past studies have revealed that ACP is defined by β -catenin mutations, overactivation of Wnt and MAPK pathways, and overall serine/threonine kinase upregulation.

SureQuant is a mass spectrometry-based technique that uses a heavy-isotope peptide library to quantify select biomolecules in a complex sample with high sensitivity from limited material. Our collaborators developed a SureQuant library capable of quantifying over 300 kinases in a sample. We hypothesized that this SureQuant kinome library could quantify significant kinases in ACP and that many kinases would be upregulated compared to healthy brain tissue.

To test our hypothesis, we completed a global proteomic and kinome analysis on 8 human ACP samples that revealed distinct differences between tumor and control brain tissue from temporal lobectomies. Gene ontology analysis of global proteomic data revealed upregulation of plasma membrane repair, actin filament remodeling, and mRNA splicing regulation. Using this data as a backdrop for our kinome analysis, we used our SureQuant library to quantify > 175 kinases. Of these, 29 were elevated in ACP compared to controls including known targets in the Wnt and MAPK pathway. We confirmed upregulation of HER2, EGFR, BTK, and CSF1R in tumor regions using immunofluorescent staining and noticed specific regional patterns of these kinases. HER2 was upregulated in sharply delineated whorl regions with increased β -catenin nuclear localization. This same pattern has been seen in breast cancer, where HER2 promotes β -catenin nuclear localization to drive tumorigenesis. Our findings suggest that many kinases including HER2 play a role in ACP pathogenesis. Future work will involve testing the HER2 inhibitor, tucatinib, in combination with MAPK inhibitors in patient derived cell lines.

Our results comprise the first proteomic analysis between ACP tumor and healthy brain tissue and the first kinome-specific analysis of this tumor. We showed that multiple kinases are upregulated in ACP and that their upregulation corresponds to distinct tumor architecture. Future studies will test HER2 inhibitors in patient derived ACP cell lines and an ACP mouse model. Our data have the potential to provide rational kinase targets for inhibitor therapy in this devastating tumor.

Basic Science

Medical Student

POSTER #50

BIPHASIC REMODELING OF CA²⁺ SIGNALING PROGRAMS DURING T HELPER CELL DIFFERENTIATION

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Within the CD4⁺ T cell compartment, T helper 1 (Th1), Th2, Th9, Th17, and regulatory T (Treg) cells represent distinct lineages that arise from a common naïve precursor and play central roles in immunity and cancer immunotherapy. Although Ca²⁺ signaling is essential for T cell activation and cytokine production, how Ca²⁺ circuits are dynamically regulated during lineage commitment remains poorly understood. Using human and mouse CD4⁺ T cell *in vitro* differentiation systems combined with functional analyses of Ca²⁺ signaling, we show that calcium signaling undergoes profound remodeling during T helper cell differentiation. Early during polarization, developing Th1, Th2, Th9, Th17, and Treg cells exhibit enhanced Ca²⁺ responses compared with activated naïve T cells cultured under neutral conditions. Longitudinal analyses further reveal progressive remodeling and divergence in Ca²⁺ signaling dynamics among lineages, suggesting the emergence of lineage-specific calcium set points. Notably, at late stages of differentiation, effector T helper cells display reduced basal Ca²⁺ levels, diminished constitutive Ca²⁺ entry, impaired store-operated Ca²⁺ entry (SOCE), and attenuated T cell receptor-dependent as well as pharmacologically induced Ca²⁺ responses relative to activated T cells under neutral conditions, indicating a restriction of calcium signaling competence once lineage commitment is established. These features are recapitulated in *in vivo*-differentiated human T helper cells. Moreover, the distinct Ca²⁺ signaling profiles of Th1, Th2, Th9, Th17, and Treg cells correlate with the expression of Ca²⁺ channels and cross-species comparisons revealed both conserved and divergent axes of Ca²⁺ regulation. Together, our findings uncover a biphasic calcium signaling program that accompanies CD4⁺ T cell differentiation, characterized by an early amplification of Ca²⁺ responses during polarization followed by a progressive restriction of calcium signaling competence in terminally differentiated effector cells.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #51

SINGLE-CELL ANALYSIS OF PLERIXAFOR-MOBILIZED HEMATOPOIETIC STEM CELLS REVEALS DISTINCT TRANSCRIPTIONAL LANDSCAPES OF DONOR CELLS

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Background: Current clinical assessments of hematopoietic stem cell (HSC) grafts rely heavily on coarse quantitative metrics, such as absolute CD34⁺ yield, which fail to capture the underlying biological heterogeneity of the mobilized graft. The molecular architecture and functional states of hematopoietic populations entering circulation remain poorly characterized across different donor and mobilization contexts. Specifically, it remains unclear how divergent mobilization regimens (standard G-CSF versus incorporation of the CXCR4 antagonist plerixafor) for donor cell collection alter the composition of primitive progenitors, lineage-primed intermediates, and immature myeloid compartments.

Methods: We performed high-resolution single-cell RNA-sequencing (scRNA-seq) analysis on mobilized peripheral blood from eight donors: healthy donors mobilized with G-CSF (G) ($n=4$) and autologous donors mobilized with G-CSF + plerixafor (GP) ($n=4$). To ensure comprehensive coverage hematopoietic hierarchy, we profiled both CD34⁺ enriched and lineage-negative (Lin^{-}) fractions for each donor. Post quality control, cell states were annotated and compared using donor-level pseudobulk analysis, pathway and transcription factor inference.

Results: GP-mobilized cells showed distinct transcriptional differences across a progenitor-to-immature-myeloid continuum. Primitive compartments, particularly hematopoietic stem cells/multipotent progenitors (HSC/MPP) and to lesser extent lympho-myeloid primed progenitors (LMPP), were enriched for hypoxia-, p53-, and TGF β -linked programs, suggesting a relatively quiescent progenitor state. In contrast, downstream progenitors and immature myeloid populations (e.g. cycling progenitors, megakaryocyte precursors, promonocytes, Eo/Baso/Mast precursors) showed recurrent enrichment of E2F, G2M checkpoint, mitotic spindle, and MAPK/EGFR/PI3K-associated programs, indicating an active proliferative state. G-mobilized cells instead demonstrated interferon-, TNF/NF-kB-, JAK-STAT-, complement-, and antigen-associated programs, particularly in monocyte and dendritic compartments. Transcription factor analysis supported this distinction, with GP-associated E2F-centered proliferative circuitry contrasting against G-associated STAT1/IRF1/RELA-NF-kB and RFX-complex activity.

Conclusion: Our findings suggest that G-mobilized cells are globally marked by an inflammatory and immune-activated hematopoietic landscape. Conversely, GP-mobilized primitive progenitors are defined by a more quiescent-like transcriptome, whereas more mature myeloid progenitors shift toward a proliferatively active state. These data provide novel insight into the impact of divergent donor backgrounds and mobilization regimens on the hematopoietic landscape, and support future investigation into molecular-driven differences in transplantation outcomes.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #52

GENOMIC DETERMINANTS OF PIK3CA MUTATION DRIVEN BREAST CANCER INITIATION

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PIK3CA is the second most mutated and/or amplified gene in breast cancer after *p53*. The *PIK3CA*-specific inhibitor Alpelisib is an FDA approved treatment for breast cancer. Toxicity and rapid development of resistance limit its clinical utility. Mutations in *PIK3CA* are found in many normal organs including the breast suggesting that additional genomic aberrations are needed for mutant *PIK3CA* to initiate breast cancer, and those aberrations can be exploited for therapy. Consistent with this possibility, our previous studies have demonstrated that immortalized breast epithelial cells derived from *BRCA1/2* mutation carriers are susceptible to transformation by the *PIK3CA* mutant, whereas transformation of immortalized breast epithelial cells from healthy donors required combinations of mutant *PIK3CA* and SV40-T/t antigens. These results suggest that transformation of breast epithelial cells by *PIK3CA* mutants requires additional genomic aberrations like those induced by SV40-T/t antigens or *BRCA1/2* mutations. To decipher genomic aberrations required for *PIK3CA* mutant driven breast epithelial transformation, we used model system of immortalized breast luminal epithelial cell lines from healthy donors. We show that mutations that activate MEK/ERK pathway such as NF-1 mutation in parallel with *PIK3CA* mutations initiate ductal carcinoma- *in situ* (DCIS)-like breast tumors with accompanying reduction in BRCA2 protein, whereas mutant *PIK3CA* plus SV40-T/t antigens generate invasive ductal adenocarcinoma (IDC). DCIS and IDC cells differed significantly in the expression of extracellular matrix components including reduced expression of LAMC2 and COL17A1 and upregulation of drivers of aneuploidy in IDC cells. Consistent with the possibility of mutant *PIK3CA* and MEK/ERK pathway collaboration in breast cancer initiation, Estrogen Receptor-positive/*PIK3CA* mutation-positive breast tumors contained higher levels of activated ERK compared to Estrogen Receptor-positive/*PIK3CA*-wild type breast cancers.

Significance: To our knowledge, this is the first study to generate DCIS like lesions from breast epithelial cells from healthy women using combinations of breast cancer-enriched genomic aberrations and without the use of viral proteins such as SV40-T/t antigens or HPV E6. In this process, we have developed an isogenic DCIS and IDC model to study breast cancer progression. These results also suggest that mutant *PIK3CA* is a weaker oncogene and the effective therapy for mutant *PIK3CA* driven breast cancers needs to include inhibitors of *PIK3CA* as well as drugs that target genomic aberrations that cooperate with mutant *PIK3CA*.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #53

CHROMOSOMAL AND GONADAL EFFECTS ON TUMOR GROWTH IN LUNG CANCER

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Background: Lung cancer (LC) is the first and second most diagnosed cancer in males and females, respectively. Recent discoveries have reopened the discussion about the potential influence of hormonal factors, particularly estrogens, in contributing to the sex differences in LC outcomes. To test the hypothesis that female gonadal hormones contribute to LC tumor progression, we used the orthotopic Lewis Lung Carcinoma (LL/2) LC model in the Four Core Genotypes (ArnoJ) mouse model, in order to determine the effects of the sex hormones and chromosomes in vivo.

Methods: We orthotopically injected LL/2-Luc2 murine cells into the left lung of 6-9 weeks old ArnoJ gonadal male (XYM and XXM) and female (XYF and XXF) mice. To track tumor progression, we measured the bioluminescence signal at different time points (day 7, 10, 13, 16, 19, and 21 post-injection). Mice were euthanized at 21 days post-injection or before if they reached humane endpoint.

Results: Our preliminary results showed a primarily gonad-derived influence on tumor growth. From the mice that reached the 21-day time endpoint (n=3-7), we showed an increased primary tumor volume and tumor weight in female mice compared to male mice (two-way ANOVA by sex, $p < 0.001$). However, male mice exhibited increased bioluminescence signal intensity and metastases compared to female mice (two-way ANOVA by sex, $p = 0.03$). To understand if these results were influenced by immune or hormone-related mechanisms, we performed gene expression analysis on the tumor samples (n=3-5) and identified that female mice had significantly increased Cd40lg expression (Limma, $\text{adj.}p=0.03$), while XX mice showed upregulation in Esr1 and Cyp1b1 (two-way ANOVA by chromosome, $p=0.03$).

Conclusion: This preliminary data and model suggest that hormonal and chromosomal factors may play a role in tumor control and progression via adaptive immune mechanisms, potentially involving Cd40lg signaling, and estrogen-specific pathways (Esr1 and Cyp1b1), respectively. Understanding the mechanisms by which hormones and chromosomes impact specific immune cell populations and tumor growth highlights the translational potential for targeting these pathways or their downstream effectors to develop sex-specific therapeutics.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #54

NANOPARTICLE PHYSICOCHEMICAL PROPERTIES SHAPE MYELOID CELL UPTAKE DURING RADIOTHERAPY

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Radiotherapy induces tumor cell death and releases tumor-derived antigens that can initiate antitumor immune responses. However, these antigens are often inefficiently captured by antigen-presenting cells because soluble proteins rapidly diffuse, are degraded by proteases, and lack particulate features that efficiently engage endocytic pathways. Within the tumor microenvironment, macrophages readily engulf cellular debris and antigens but typically process them through degradative pathways that limit cross-presentation to CD8⁺ T cells. In contrast, type 1 conventional dendritic cells (cDC1) possess specialized machinery that enables efficient antigen cross-presentation and priming of cytotoxic CD8⁺ T-cell responses. Strategies that capture and stabilize tumor-derived antigens in particulate forms accessible to cross-presenting dendritic cells may therefore enhance antitumor immunity.

Nanoparticles provide a means to capture extracellular proteins because they readily adsorb proteins onto their surfaces and present them in particulate form that promotes immune-cell uptake. In this study, we investigated whether gold nanoparticles (AuNPs) can capture tumor-derived proteins and influence their uptake by myeloid cells. AuNPs were synthesized and characterized by UV-Vis spectroscopy, dynamic light scattering (DLS), and scanning electron microscopy (SEM) to determine particle size, dispersity, and morphology. Model proteins, including bovine serum albumin (BSA), and proteins derived from murine tumor cell lysates, were used to evaluate protein corona formation on the nanoparticle surface. Protein-coated AuNPs were incubated *ex vivo* with murine splenocytes and lymph node cell suspensions, and nanoparticle uptake by myeloid cell populations was quantified by flow cytometry.

These studies define how nanoparticle physicochemical properties influence antigen capture and uptake across myeloid cell populations, including dendritic cells, macrophages, monocytes, and neutrophils. Establishing how nanoparticle-bound proteins are distributed among these immune cell subsets provides a foundation for designing nanoparticles that direct tumor antigens toward antigen-presenting cells and improve antigen delivery during radiotherapy.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #55

PRIMARY STROMAL FIBROBLAST ISOLATION FROM OVARIAN ENDOMETRIOMAS IN THE CONTEXT OF ENDOMETRIOSIS-ASSOCIATED OVARIAN CANCER

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Background: Endometriosis is a chronic inflammatory condition defined by the growth of endometrial-like tissue outside the uterus. Ovarian endometriomas (EOMA) are the most common presentation. Women with EOMA face a 19-fold increased risk of ovarian cancer. A key hallmark of EOMA is adhesive disease with fibrosis, likely due to contributions of stromal fibroblasts within the microenvironment. EOMA stromal fibroblasts, similar to cancer-associated fibroblasts, facilitate extracellular matrix remodeling and fibrosis. Primary stromal fibroblasts from EOMA remain poorly characterized due to limited surgical samples and difficulties in isolating viable cells.

Objectives: To characterize the clinical features associated with the successful isolation of primary stromal fibroblasts and the molecular features of isolated primary stromal fibroblasts.

Methods: With IRB approval, fresh tissues (EOMA and endometrium) were procured. Primary stromal fibroblasts were isolated using mechanical and enzymatic digestion, size filtration, and selective growth. Immunofluorescence was used to confirm positive expression of stromal fibroblast markers (*e.g.*, vimentin) and the absence of expression of epithelial (*e.g.*, cytokeratin) and endothelial cell markers (*e.g.*, CD31). Descriptive statistics summarized subject demographics. For *in vitro* decidualization, fibroblasts were treated with 10nm 17 β -estradiol, 1mm medroxyprogesterone acetate, 0.5mM 8-bromo-cAMP) for 96 hours. QPCR was used to evaluate the expression of prolactin (PRL), insulin-like growth factor binding protein-1 (IGFBP1), and bone morphogenetic protein-2 (BMP2).

Results: A total of 53 fresh tissue samples were procured from EOMA (n=41) and endometrium (n=12). There was no difference in median age between EOMA and control (EOMA: median 39 years; control: median 32.5 years; Mann-Whitney, p=0.08). There was no difference in the ability to successfully culture stromal fibroblasts between the two groups (EOMA: 51.2%; control 66.7%; Fisher's exact test, p=0.51). All EOMA and control stromal fibroblast cultures showed positive vimentin staining. None of the stromal fibroblasts from controls showed contamination with cytokeratin or CD31-positive cells. 18.2% of EOMA were cytokeratin-positive, and 9.1% were CD31-positive. Stromal fibroblasts from control endometrium treated with decidualization cocktail exhibited a strong induction of decidualization markers compared to vehicle [PRL: 319.4-fold increase; IGFBP1: 16.3-fold increase; BMP2: 2.84-fold increase (n=3)]. Stromal fibroblasts from EOMA treated with decidualization cocktail exhibited a statistically significant blunted induction of decidualization markers compared to control induction [PRL: 93.5-fold increase; IGFBP1: 2.98-fold increase; BMP2: 1.85-fold increase (n=3), t-test, p<0.05].

Conclusions: Primary stromal fibroblast cultures can be established from EOMAs. Careful evaluation of contaminating cellular populations is important and may require optimization of isolation protocols. Primary stromal fibroblasts from EOMAs exhibit a blunted decidualization response, consistent with progesterone-

resistant disease. Progesterone is considered anti-inflammatory and may exhibit anti-fibrotic properties. Future studies will characterize the pro-fibrotic properties of EOMA stromal fibroblasts as a feature of the ovarian cancer tumor microenvironment.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #56

DUAL-ACTION MITOCHONDRIAL-TARGETING PEPTIDE ENHANCES IMMUNOTHERAPY AND OVERCOMES TCE RESISTANCE IN MYELOMA

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Background: T cell engaging (TCE) therapies, including bispecific antibodies and CAR T cells, have improved outcomes in multiple myeloma (MM) treatment, yet resistance remains common due to tumor evasion and T-cell dysfunction. Clinically, resistance often occurs without antigen loss, limiting the benefit of teclistamab (a BCMA×CD3 bispecific antibody) dose escalation or checkpoint blockade. We developed PP-K, a dual-function peptide that binds PD-L1 to relieve PD-1 mediated T-cell suppression while delivering a mitochondrial-targeted proapoptotic peptide (KLAKLAKLAK)₂ to PD-L1-expressing tumor cells. Here, we tested whether PP-K can overcome teclistamab resistance in clinically relevant MM models.

Methods: Teclistamab-resistant OPM2-TCE^R were generated by prolonged exposure to the FDA-approved bispecific antibody teclistamab and analyzed alongside their parental lines. PP-K cytotoxicity was assessed by MTS assays. PP-K, teclistamab, and combination were tested in MM cells and healthy donor CD3⁺ T-cells co-cultures. MM cells were engineered to express GFP and secreted luciferase, tumor viability was quantified by luciferase activity. T-cell activation, differentiation, and effector function were assessed by flow cytometry and IFN- γ / granzyme B ELISA. PP-K induced cell death was analyzed by Annexin V/PI staining. Pan-caspase inhibitor (ZVAD-FMK) and necroptosis inhibitor (Necrostatin-1) dissected death mechanisms. For in-vivo studies, NSG mice were co-injected subcutaneously with OPM2-TCE^R cells and human T cells, followed by intraperitoneal administration of PP-K, teclistamab, or combination treatment. Tumor burden was monitored by tumor volume and circulating secreted luciferase.

Results: PP-K showed consistent single-agent cytotoxicity across both standard and teclistamab-resistant MM cell lines, with IC₅₀ of 5–8 μ M, independent of BCMA expression. OPM2-TCE^R cells retained BCMA and PD-L1 levels comparable to parental lines and remained sensitive to PP-K. In OPM2-TCE^R co-cultures, increasing concentration of teclistamab induced minimal T-cell activation, reflected by low CD8⁺CD25⁺ cells and increased PD-1 expression, indicating T-cell exhaustion and poor tumor killing compared to similarly treated OPM2-control co-cultures. Although OPM2-TCE^R co-cultures were unresponsive to teclistamab alone (0.031-0.062 nM), combining PP-K with teclistamab achieved >90% tumor killing, whereas single agents were ineffective. Cytotoxicity correlated with increased IFN- γ and granzyme B secretion and enrichment of cytotoxic CD8⁺ TEMRA (CD45RA⁺CCR7⁻) cells. Mechanistically, PP-K induced rapid tumor cell death within 1 hour, with >50% of cells entering late apoptosis/necrosis. However, neither pan-caspase nor necroptosis inhibition rescued PP-K-induced cell death, indicating a largely caspase- and necroptosis-independent mechanism. In vivo, NSG mice co-injected with GFP/luciferase OPM2-TCE^R cells and human T

cells showed significant tumor inhibition with combination therapy compared with single-agent therapy, as evidenced by reduced tumor volume and circulating luciferase.

Conclusion: PP-K improves responses in teclistamab-resistant MM models by directly killing PD-L1+ tumor cells and enhancing CD8⁺ T-cell activation. Future studies will use patient-derived xenografts and chronic stimulation to model T-cell dysfunction to better understand resistance and response mechanisms.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #57

GATA2^{T354M} HEMIZYGOSITY COOPERATES WITH BI-ALLELIC CEBPA DELETIONS TO INITIATE AML

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Inherited mutations in the transcription factor *GATA2* lead to *GATA2* deficiency syndrome, which is characterized by opportunistic infections and cytopenias of monocytes, B cells, and NK cells. These patients also have a ~70% chance of developing myelodysplastic syndromes (MDS) and/or acute myeloid leukemia (AML). The most common missense mutations in *GATA2* deficiency syndrome are T354M, R398W/Q, and R396Q/W. Among them, *GATA2*^{T354M} is the most strongly associated with AML progression. *GATA2* mutations are also found in 4-6% of *de novo* AML cases. These mutations are heterozygous in ~98% of cases. Interestingly, the remaining wild-type allele is epigenetically silenced in nearly all cases. Therefore, only the mutated allele is expressed in AML. In addition, hemizygous expression of a mutated *GATA2* allele has been shown to closely track with cytopenias in patients. Approximately half of *GATA2* mutated AML cases also have bi-allelic *CEBPA* deletions, suggesting that these mutations cooperate to initiate AML. However, it remains unknown at a molecular level how *GATA2* mutations promote AML and why *CEBPA* and *GATA2* mutations tend to co-occur.

To answer these questions, we developed a germline *Gata2*^{T354M} knock-in mouse model. Surprisingly, we found that *Gata2*^{T354M/T354M} homozygous mice were viable. This is first mouse model in which only a mutated *Gata2* allele is expressed, which is also observed clinically. Flow cytometric analysis demonstrated a significant reduction in long-term hematopoietic stem cells from *Gata2*^{T354M/T354M} and *Gata2*^{T354M/+} mice, with decreases of 87.0% and 54.1%, respectively, compared to WT mice. Additionally, *Gata2*^{T354M/T354M} mice exhibited a 66.9% reduction in B cells, which was accompanied by a marked depletion of upstream progenitor populations, including a 93.4% reduction in lymphoid-primed multipotent progenitor cells and an 88.4% reduction in common lymphoid progenitors, suggesting that the loss of *GATA2* function impairs early B cell lineage specification and commitment. We also observed a 46.0% and 58.5% reduction in monocytes and NK cells in *Gata2*^{T354M/T354M} mice. Therefore, *Gata2*^{T354M/T354M} mice display clinical features of *GATA2* deficiency syndrome, including cytopenias of B cells, monocytes, and NK cells.

Another important clinical feature is the development of AML, which the *Gata2*^{T354M/+} mice develop, albeit with long latency (532 days) and low penetrance (<10%). This suggests that secondary mutations are required for transformation. To model the co-association with *Cebpa* deletions, we utilized CRISPR/Cas9 genome editing to induce these deletions and *Gata2* hemizygosity in bone marrow from *Gata2*^{T354M/+} mice. Bone marrow transplantation of these cells showed that *Gata2*^{T354M/-} hemizygosity cooperates with *Cebpa*^{-/-} to induce 100% penetrance with a latency of ~200 days. This latency was 50 days shorter than mice that received *Gata2*^{T354M/+} and *Cebpa*^{-/-} cells. Therefore, we have developed the first mouse model that recapitulates AML initiation by an inherited *Gata2* mutation, and the first to model *Gata2* hemizygous mutations.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #58

FOLR1 SUSTAINS METABOLIC FITNESS TO DRIVE IMMUNE CHECKPOINT RESISTANCE IN KRAS^{G12D}-MUTANT COLORECTAL CANCER

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Introduction

Colorectal cancer (CRC) remains a leading cause of cancer-related mortality worldwide. Activating KRAS mutations occur in approximately 40–50% of CRCs, with KRAS^{G12D} representing one of the most prevalent subtypes. Emerging evidence indicates that KRAS-mutant CRC responds poorly to immune checkpoint blockade (ICB), reflecting strong tumor-intrinsic resistance to immunotherapy. To identify key regulators capable of overcoming this resistance, we performed an in vivo CRISPR/Cas9 genetic screen in a KRAS^{G12D}-driven CRC model and investigated the underlying mechanisms contributing to immune escape.

Methods

KRAS^{G12D} colorectal cancer cell lines (MC38K and Caco2K) were generated. An immune-related sgRNA library was constructed based on genes associated with PD-1/PD-L1 signaling, MHC-I antigen presentation pathways, and ESTIMATE-derived immune signatures, and MC38K cells carrying the library were screened in vivo under anti-PD-1 treatment. RNA sequencing, untargeted metabolomics, and Seahorse metabolic assays were performed to characterize tumor-intrinsic mechanisms. Multiple CRC models were used to evaluate Fcrl1 inhibition combined with anti-PD-1 therapy, and a Fcrl1-based peptide vaccine was developed to overcome immunotherapy resistance.

Results

KRAS^{G12D} overexpression accelerated tumor growth and conferred resistance to anti-PD-1 therapy in MC38 models. Through the in vivo CRISPR/Cas9 screen, Fcrl1 emerged as the top depleted gene, ranking first among candidates associated with immunotherapy resistance. Importantly, FOLR1 is a clinically actionable target with FDA-approved therapeutic antibodies, highlighting its translational relevance.

Integrated transcriptomic and metabolomic analyses of Fcrl1-deficient MC38K and Caco2K cells revealed global metabolic impairment, characterized by broad suppression of central metabolic pathways. In particular, pathways associated with glycolysis, hypoxia, and mTORC1 signaling were significantly downregulated, which was validated by qRT-PCR, OCR/ECAR assays, and lactate measurements.

Importantly, metabolic disruption triggered activation of the cGAS–STING innate immune signaling pathway, resulting in induction of interferon-stimulated genes and coordinated upregulation of MHC-I antigen-processing machinery (including NLRC5, TAP1/2, B2M, and HLA/H2 genes), thereby enhancing tumor cell visibility to cytotoxic T cells.

Functionally, Fcrl1 knockout significantly sensitized tumors to anti-PD-1 therapy, leading to delayed tumor growth and prolonged survival across multiple CRC models. Furthermore, a Fcrl1-based peptide vaccine effectively suppressed tumor progression.

Conclusions

Our study identifies Fcrl1 as a key tumor-intrinsic regulator of immunotherapy resistance in KRAS^{G12D}-mutant colorectal cancer. Targeting Fcrl1 induces metabolic impairment and activates cGAS–STING–mediated innate immune signaling, enhancing antigen presentation and restoring responsiveness to anti–PD-1 therapy. These findings establish FOLR1 as a promising therapeutic target to overcome immune resistance in KRAS-driven CRC.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #59

DYNAMIC STIFFENING OF PDAC TUMOR ENVIRONMENT INCREASES EARLY TUMOR BURDEN AND RESISTANCE TO CHEMOTHERAPY IN AN ORTHOTOPIC MODEL

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The desmoplastic stroma in the pancreatic tumor microenvironment (TME) is one characteristic of the disease that makes pancreatic ductal adenocarcinoma (PDAC) one of the deadliest malignancies and contributes to drug resistance and wasting (cachexia). This stroma is comprised mainly of extracellular matrix (ECM) and cancer-associated fibroblasts (CAFs), which contribute to clinical challenges through a myriad of complex mechanisms. To identify and develop novel therapeutics to effectively treat PDAC, it is critical to understand these mechanisms. As peripheral areas of primary tumors are stiffer than the core areas with higher fibrous and matrix protein concentrations, we hypothesized that pancreatic cancer cells (PCCs) grown in a stiff matrix would be more aggressive and resistant to treatment compared to cells grown in a softer matrix. To test this, we engineered biomimetic hydrogels crosslinked by decellularized small intestine submucosa-norbornene (dSIS-NB) and thiolated hyaluronic acid (THA) that can be stiffened or softened using a photoinitiation system. PCCs were grown in and subsequently recovered from hydrogels of varying stiffness and mixed with CAFs for orthotopic pancreatic implantation, and mice were treated with a standard chemotherapy regimen of Gemcitabine/Abiraterone (Gem/ABX) for three weeks. Bioluminescent imaging (BLI) showed similar growth rates between PCCs cultured in soft vs stiff gels. However, Gem/ABX-treated tumors generated from PCCs grown in stiff hydrogels (shear moduli ~ 2.2 kPa) were significantly larger and more metastatic compared to treated tumors generated from cells grown in soft hydrogels (shear moduli ~ 0.5 kPa), suggesting that higher matrix stiffness altered PDAC cell response in vivo, potentially through mechanical memory in the cells resulting in treatment-resistant tumors. To ascertain whether softening the stiff hydrogels could ameliorate treatment resistance, PCCs were grown in hydrogels that were either soft (soft) or stiff initially and then softened (stiff-to-soft) before recovery of PCCs. Three weeks post implant, vehicle-treated tumors derived from soft PCCs were significantly smaller than vehicle-treated tumors derived from stiff>soft PCCs. At 5 weeks post implant, mice bearing tumors from stiff-to-soft PCCs displayed greater reductions in quadriceps, gastrocnemius, and tibialis anterior muscles, suggesting exacerbated cachexia compared to soft PCCs tumor hosts. Additionally, weekly BLI revealed that soft PCC tumors were initially more sensitive to Gem/ABX compared to stiff-to-soft PCC tumors. These differences were lost at later time points, suggesting the mechanical memory driving PCC aggression and treatment resistance is limited and can potentially be reduced by dynamically softening the early TME. Our results suggest that dynamic softening of the environment in early carcinogenesis can slow tumor progression, reduce muscle wasting, and sensitize tumors to chemotherapy. Future studies will continue to investigate this paradigm to delineate the mechanisms by which tumor stiffness contributes to disease progression, identifying signaling axes that can be targeted therapeutically to treat the disease.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #60

PRSS2 REGULATES HUMAN HEMATOPOIETIC STEM CELL POTENCY AND TRANSPLANTATION EFFICIENCY

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Background: Hematopoietic stem and progenitor cells dynamically respond to physiologic and pathologic stressors, including transplantation and malignant transformation. Among the key microenvironmental regulators of this process is oxygen tension, yet the gene-specific pathways linking hypoxia to stem cell potency remain incompletely defined.

Methods: *PRSS2* (encoding Trypsin-2) was identified through bulk and single-cell RNA-sequencing analyses correlated with measured or predicted hematopoietic cell potency in transplantation models and publicly available patient datasets. To determine its functional role in human hematopoietic stem and progenitor cells, cord blood human cord blood CD34⁺ hematopoietic stem and progenitor cells underwent siRNA-mediated *PRSS2* knockdown or recombinant protein treatment, followed by immunophenotyping and colony-forming unit (CFU) assays under variable oxygen conditions. Engraftment potential was evaluated using humanized mouse transplantation models. Associations between *PRSS2* expression and repopulating capacity were assessed using cord blood transplantation datasets. Patient cohorts were analyzed to determine the prognostic relevance of *PRSS2* in pediatric leukemia and aplastic anemia. Knockdown efficiency was validated by qPCR in CD34⁺ and RPMI8226, multiple myeloma cell line cells, with protein confirmation by immunofluorescence.

Results: Transcriptomic analyses identified *PRSS2* as a candidate regulator of hematopoietic potency. *PRSS2* was detected in CD34⁺ hematopoietic stem and progenitor cells and enriched in primitive subsets, with expression decreasing as oxygen tension increased. Higher *PRSS2* expression was observed in cord blood units with superior repopulating capacity in transplantation assays and in cells predicted to yield improved clinical outcomes.

Mining of publicly available datasets revealed that *PRSS2* is enriched in pediatric acute leukemias with poor prognosis and in multiple myeloma, while expression is reduced in the bone marrow of patients with aplastic anemia, partially recovering following immunosuppressive therapy. These findings indicate a strong association between *PRSS2* expression and both normal and malignant hematopoietic function.

Functionally, siRNA-mediated knockdown of *PRSS2* in human cord blood CD34⁺ cells reduced immunophenotypically defined HSC frequencies and impaired colony-forming capacity, with the most pronounced loss in multi-lineage GEMM colonies under hypoxia. In freshly isolated cord blood, knockdown significantly reduced total, multilineage (GEMM), granulocyte-macrophage (GM), and erythroid (BFU-E) colonies. Engraftment of *PRSS2*-deficient cells in immunodeficient mice was significantly decreased at 4, 8, and 12 weeks post-hematopoietic cell transplantation. Conversely, ex vivo treatment with recombinant Trypsin-2 enhanced CD34⁺ cell expansion and multilineage colony output. In malignant models, *PRSS2* knockdown reduced proliferation of AML and multiple myeloma cell lines. Immunofluorescence in RPMI8226 cells confirmed that *PRSS2* protein expression is induced under hypoxia and effectively suppressed following siRNA knockdown.

Conclusions: These findings establish *PRSS2* as a regulator of human hematopoietic stem and progenitor cell potency that integrates microenvironmental oxygen cues with functional output. *PRSS2* supports stem cell maintenance, multilineage colony formation, and transplantation efficiency. Collectively, this work highlights

PRSS2 as a candidate biomarker of donor potency and a potential target to improve hematopoietic cell transplantation, with broader implications for both normal and malignant hematopoiesis.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #61

C-C CHEMOKINE RECEPTOR TYPE 2 (CCR2) PROMOTES HEMATOPOIETIC STEM AND PROGENITOR CELL MOBILIZATION

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Background

Numerous hematologic malignancies, including leukemia, originate from dysfunction of hematopoietic stem cells (HSCs). Hematopoietic stem cell transplantation (HSCT) provides a potentially curative therapy for these disorders by replacing diseased HSCs with healthy donor cells. Successful transplantation critically depends on efficient mobilization of hematopoietic stem and progenitor cells (HSPCs) from the bone marrow (BM) into the peripheral blood (PB) for collection. Clinically, granulocyte colony-stimulating factor (G) and the CXCR4 antagonist plerixafor (P) are widely used to disrupt BM retention signals and promote HSPC mobilization. However, a substantial proportion of patients undergoing autologous HSPC collection fail to mobilize sufficient numbers of cells for transplantation, particularly those previously treated with chemotherapy drugs.

Hypothesis

We previously identified a functional interaction between the chemokine receptors CXCR4 and CCR2 that regulates BM homing and niche occupancy. Mice transplanted with *Ccr2*^{-/-} BM cells showed reduced homing, engraftment, and blood cell reconstitution at week 2 compared with wild-type mice. However, higher engraftment and T-lymphocyte recovery were seen at later time points, supporting a possible role for CCR2 in HSPCs egress from BM at steady-state. Based on these observations, we hypothesized that CCR2/CXCR4 interactions regulate HSPC mobilization from BM to PB during donor cell collection.

Methods

To examine the role of CCR2 in vivo mobilization, the CCR2 ligand CCL7 (C) was incorporated into G±P mobilization regimens in C57BL/6 mice. Graft chimerism, white blood cell counts (WBCs), Lin⁻Sca-1⁺c-Kit⁺ (LSK) cells, peripheral blood colony-forming units (CFUs), and lymphocyte recovery were measured. To investigate CCR2 function in human HSPC trafficking, HSPCs were enriched from donor apheresis products mobilized with G or GP. Flow cytometry and high-resolution imaging flow cytometry were used to analyze CXCR4 and CCR2 expressions and receptor co-localization. Functional assays included intracellular calcium mobilization and transwell migration toward CCR2 ligand CCL7 and CXCR4 ligand CXCL12.

Results

In mice, GC administration increased WBC counts, mobilized LSK cells, and increased PB CFUs compared with G alone. Addition of CCL7 to the GP regimen (GPC) further increased circulating WBCs, LSK cells, and CFU output. Transplantation of GPC-mobilized cells resulted in faster lymphocyte recovery at four weeks post-transplantation with similar long-term engraftment potency as GP-mobilized cells. In human samples, GP-mobilized grafts (HSCs^{GP}) with higher mobilization yield contained increased frequencies of CXCR4⁺ and CCR2⁺ cells. In particular, CD49f⁺ HSCs^{GP}, granulocyte-monocyte progenitors^{GP}, and lymphoid-primed multipotent progenitors^{GP} exhibited higher proportions of CXCR4⁺CCR2⁺ cells. Imaging flow cytometry demonstrated co-localization of CCR2 and CXCR4 on the cell surface. CCL7 enhanced intracellular calcium

mobilization in synergy with CXCL12. Functionally, HSPC^{GP} showed greater baseline chemotaxis and increased migration toward CCL7 and CXCL12 gradients.

Conclusion

Collectively, these findings identify CCR2 as a key regulator of HSPC trafficking. Targeting CCR2 signaling may improve HSPC mobilization efficiency and ultimately optimize transplantation outcomes.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #62

EXPRESSION AND FUNCTIONAL ROLE OF ICAM-1 IN ORAL SQUAMOUS CELL CARCINOMA: FROM BIOMARKER TO MECHANISTIC TARGET

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Background:

Oral squamous cell carcinoma (OSCC) represents a significant global health burden due to its aggressive nature, late-stage diagnosis, and poor survival rates. Identifying molecular drivers of oral carcinogenesis is essential for developing early biomarkers and therapeutic targets. Intercellular adhesion molecule 1 (ICAM-1), a transmembrane glycoprotein involved in cell adhesion and signal transduction, plays a key role in various pathologic conditions, including inflammation, immune response, and cancer progression. Using a 4-NQO-induced oral carcinogenesis mouse model, we previously demonstrated that ICAM-1 expression is absent in intact oral epithelium but progressively increases across stages of carcinogenesis—present in 87.5% of hyperplastic tissues and 100% of dysplastic and papilloma tissues—with overall expression in 93.2% of OSCC tissues. These findings identified ICAM-1 as a promising biomarker for early OSCC detection; however, its functional role in oral epithelial biology and carcinogenesis remained undefined.

Methods:

To investigate the functional significance of ICAM-1 in oral epithelial homeostasis and tumorigenesis, we generated two novel mouse models: K5-Cre;ICAM-1^{flox/flox} mice for epithelial-specific ICAM-1 deletion, and ICAM-1-CreERT2;EGFP mice for inducible lineage tracing of ICAM-1-expressing cells. Lineage tracing was performed under homeostatic conditions, wound healing, and 4-NQO-induced carcinogenesis. Tumor formation was compared between K5-Cre;ICAM-1^{flox/flox} and control mice following 4-NQO treatment.

Results:

Lineage tracing using ICAM-1-CreERT2;EGFP mice revealed that under homeostatic conditions, EGFP+ cells transiently form clones that are rapidly turned over, indicating that ICAM-1 marks short-lived progenitor cells derived from basal stem cells rather than long-term stem cells themselves. During wound healing, the fate of ICAM-1+ progenitors was significantly skewed, demonstrating their previously unrecognized cellular plasticity. In the 4-NQO carcinogenesis model, lineage tracing revealed a striking parallel to wound healing: ICAM-1+ cells acquired a survival advantage and stem-like properties, contributing to tumor formation. Epithelial-specific deletion of ICAM-1 in K5-Cre;ICAM-1^{flox/flox} mice significantly reduced tumor formation following 4-NQO treatment, providing causal evidence that ICAM-1 expression promotes oral carcinogenesis.

Conclusions:

ICAM-1 marks a plastic progenitor cell population whose fate can be co-opted during both wound healing and malignant transformation. Its progressive upregulation during oral carcinogenesis, combined with the tumor-suppressive effect of its epithelial deletion, establishes ICAM-1 not only as a promising early biomarker for OSCC detection but as a functionally relevant driver of tumor initiation and progression—and a compelling therapeutic target.

POSTER #63

NQO1-BIOACTIVATABLE THERAPY SENSITIZES PARP INHIBITOR-RESISTANT PANCREATIC CANCER CELLS TO OXIDATIVE STRESS-INDUCED CELL DEATH

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Acquired resistance to PARP inhibitors (PARPi) remains a major barrier to durable therapeutic responses in pancreatic ductal adenocarcinoma (PDAC). To investigate mechanisms underlying PARPi resistance and identify strategies to overcome it, we generated Rucaparib-resistant (Ru-R) PDAC cell models using MiaPaCa2 and Capan2 cells through long-term stepwise drug selection. Drug sensitivity assays confirmed that these Ru-R clones exhibit robust resistance to Rucaparib and cross-resistance to additional clinically used PARP inhibitors, including Olaparib and Talazoparib, indicating a stable, class-wide resistance phenotype. Molecular characterization revealed adaptive transcriptional and protein-level changes associated with drug resistance, including upregulation of the drug efflux transporter ABCG2, NAD⁺ metabolic enzymes NAMPT and NMNAT2, and growth factor signaling protein IGFBP3, accompanied by reduced expression of PAR metabolism regulators such as PARG and PARP1. These alterations suggest that PARPi-resistant PDAC cells undergo coordinated metabolic and stress-adaptive reprogramming. We next investigated whether NQO1-bioactivatable therapy could target these resistant cells. The novel NQO1-bioactivatable drug, IB-DNQ, retained strong cytotoxic activity in Ru-R cells and induced robust accumulation of reactive oxygen species, DNA double-strand breaks, and apoptotic cell death in an NQO1-dependent manner. However, IB-DNQ treatment also triggered rapid phosphorylation of AKT, suggesting activation of a compensatory pro-survival signaling pathway. Pharmacological inhibition of AKT using MK2206, which has also been reported to inhibit the drug efflux transporter ABCG2, significantly enhanced IB-DNQ-induced cytotoxicity and demonstrated strong synergistic activity in PARPi-resistant cells. These findings indicate that PARPi-resistant PDAC cells develop multi-layered adaptive mechanisms involving drug efflux, metabolic rewiring, and survival signaling. Targeting redox vulnerabilities with NQO1-bioactivatable therapy, particularly in combination with AKT inhibition, represents a promising strategy to overcome PARPi resistance and improve therapeutic responses in pancreatic cancer.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #64

THE TYROSINE PHOSPHATASE PTPN2 IS A NEGATIVE REGULATOR OF TH9 CELL EFFECTOR FUNCTIONS

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IL-9-producing CD4⁺ T helper cells (Th9 cells) exhibit potent antitumor activity in preclinical models and outperform Th1 and Th17 cells in controlling solid tumors following adoptive transfer. However, the molecular mechanisms that regulate Th9 cell differentiation and effector functions remain incompletely understood, limiting the development of Th9-based immunotherapies. Here we identify the protein tyrosine phosphatase non-receptor type 2 (PTPN2) as a negative regulator of Th9 cell functions.

Pharmacological inhibition of PTPN2 using the small-molecule inhibitor ABBV-CLS-484 (AC484) markedly enhanced Th9 differentiation and effector activity, characterized by increased production of IL-9, IL-21 and granzyme B. These effects were confirmed using a second PTPN2 inhibitor, a PROTAC-mediated PTPN2 degrader and CRISPR-Cas9-mediated PTPN2 deletion. Mechanistically, PTPN2 inhibition amplified JAK-STAT signaling, resulting in increased phosphorylation of JAK1/3 and downstream STAT1, STAT3, STAT5 and STAT6. Using STAT1^{-/-} mice, we found that STAT1 deficiency prevented the induction of IL-9 from Th9 cells restimulated with AC484, suggesting that STAT1 is primarily responsible for the effects of PTPN2 inhibition on Th9 cells.

Adoptive transfer of AC484-treated Th9 cells significantly improved tumor control in both transplantable and spontaneous melanoma models, resulting in prolonged survival. Enhanced antitumor immunity was associated with increased infiltration of CD8⁺ T cells and elevated IFN- γ production within the tumor microenvironment. Neutralization studies further demonstrated that IL-9 and IL-21 are key mediators of the augmented antitumor activity induced by PTPN2 inhibition.

These findings overall identify PTPN2 as a central regulator of Th9 cell differentiation and effector function and establish pharmacological PTPN2 blockade as a strategy to enhance Th9-mediated antitumor immunity.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #65

SENESCENT TUMOR-ASSOCIATED MACROPHAGES PROMOTE PERITONEAL METASTATIC NICHE FORMATION IN GASTRIC CANCER

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Peritoneal metastasis represents a major clinical challenge in gastric cancer (GC), yet the mechanisms underlying metastatic niche formation remain poorly understood. In particular, the specific tumor-associated macrophage (TAM) subsets that drive peritoneal dissemination remain poorly characterized. Here, we identify a subset of senescent macrophages that promote metastatic niche propagation in GC. Using a newly established GC cell line, GAN-KPC (harboring a $Kras^{G12V}$ mutation with *Trp53* and *Cdh1* knockout), and a syngeneic gastric wall transplantation model in C57BL/6 mice, we investigated the cellular landscape of metastatic niche formation. High-resolution spatial transcriptomics (10x Visium HD) revealed macrophages within metastatic niches exhibiting transcriptional features of cellular senescence. Pharmacologic clearance of senescent cells with the senolytic agent ABT263 markedly reduced peritoneal tumor formation, and macrophage-specific *p16^{INK4a}* knockout further confirmed the essential role of senescent macrophages in metastatic niche formation. Complementarily, single-cell mass cytometry (CyTOF) of ascitic fluid from GC patients with peritoneal metastasis revealed a macrophage subset expressing p16 and SASP-associated factors. These findings uncover a previously unrecognized role of senescent macrophages in peritoneal metastatic niche formation and suggest that targeting senescent cells may represent a promising therapeutic strategy to limit peritoneal dissemination in GC.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #66

TARGETING MYC SIGNALING IN LUNG NEUROENDOCRINE CARCINOMAS WITH PIM2 AND AURKA INHIBITION

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Introduction: Small cell lung cancer (SCLC) and large cell neuroendocrine carcinoma of the lung (LCNEC) are aggressive neuroendocrine carcinomas (NECs) characterized by frequent chemoresistance and limited effective treatment options for relapsed disease. Recent studies have identified amplifications of c-MYC and its paralogs as key drivers of tumor plasticity across the neuroendocrine spectrum in both SCLC and LCNEC. JP11646 is a potent non-ATP competitive inhibitor of PIM family members, which phosphorylate c-MYC at Ser329, and has demonstrated preclinical efficacy in multiple malignancies, including triple negative breast cancer and multiple myeloma. Alisertib, an AURKA inhibitor currently under Phase II investigation for relapsed SCLC, blocks the Aurora A-mediated stabilization of c-MYC and N-MYC and has shown promising antitumor activity. Here, we investigate the functional effects of JP11646 in lung NECs and evaluate whether the combination therapy with JP11646 and alisertib produces synergistic antitumor effects.

Methods: JP11646 and alisertib sensitivity were evaluated separately with serial dilutions in a panel of NEC cell lines (H1155, H524, H82, H209), and cell viability was measured after 3-day drug exposure by CellTiter-Glo Luminescent Assay. H1155 and H82 cells were treated with JP11646, alisertib, or combination of both for 24 hour (h) prior to western blot analysis of target proteins. For CUT&RUN (Cleavage Under Targets & Release Using Nuclease) assays, H1155 cells were treated with JP11646 or control for 24 h in two biological replicates, and bound with three different antibodies targeting H3K4me3, N-MYC, and PIM2 in a total of 12 reactions. All 12 CUT&RUN libraries were prepared, sequenced, and analyzed following EpiChypher's instruction.

Results: The panel of MYC-paralog amplified cell lines differentially exhibit nanomolar potency for JP11646 (IC₅₀ range, 49 - 136 nM) and alisertib (IC₅₀ range, 3 - 189 nM). Following 24 h JP11646 treatment, both H1155 and H82 cells exhibited reduction of PIM2 and MYC-paralogs and extensive downregulation of AURKA. With 24 h alisertib treatment alone, both H1155 and H82 cells had decreased phosphorylation of AURKA and slightly increased WEE1 protein expression. With the combination treatment of JP11646 and alisertib, both H1155 and H82 cells had decreased WEE1 and nearly complete loss of AURKA phosphorylation. Processed CUT&RUN data were visualized using the UCSC Genome Browser, focusing on key target genes including PIM2, AURKA, and MYC paralogs, as well as subtype-specific transcription factors ASCL1 and NEUROD1. In brief, treatment with JP11646 showed decreased chromatin enrichment (lower peak intensity) at the PIM2, AURKA, N-MYC and NEUROD1 loci in H1155 cells compared to water-treated controls.

Conclusion: JP11646 reduces MYC-paralog expression in lung NECs and exerts its anticancer effects likely by blocking PIM2-MYC interactions and AURKA phosphorylation. Future studies are underway to further examine the mechanism and synergistic effects of JP11646 and alisertib, including combination screening and xenograft studies.

Basic Science

Post-Doctoral/Medical Fellow

POSTER #67

ALZHEIMER'S DISEASE ATTENUATES BREAST CANCER PATHOLOGY IN A 5XFAD MOUSE MODEL

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Alzheimer's disease (AD) is the leading cause of dementia worldwide, accounting for approximately 60–70% of cases. AD is most common in people age 65 and older, with women accounting for two-thirds of people living with Alzheimer's disease. Breast cancer (BC) is the second most prevalent cancer globally, yet it remains understudied in older populations. A growing number of research has reported an inverse relationship between Alzheimer's disease and cancer. Furthermore, clinical studies have also revealed an inverse association between AD and BC. These findings raise the possibility that biological mechanisms active during early AD progression may exert anti-tumorigenic effects. However, this relationship has not been investigated in preclinical models. In this study, syngeneic EO771 breast cancer cells were orthotopically implanted into the #4 mammary gland of female 5xFAD transgenic mice and age-matched C57BL/6 wild-type controls at 2 and 4 months of age. Tumor volume was measured for 28 days following injection, and tumor and brain tissues were subsequently collected for analysis. At 2 months, 5xFAD mice exhibited reduced tumor growth compared to wild-type controls. By 4 months of age—corresponding with increased AD pathology—tumor growth rates were similar between the two groups. These initial findings suggest a stage-dependent alteration in breast cancer tumorigenesis during AD progression. Ongoing studies will further evaluate common pathological markers, including amyloid- β , tau, microglia, and CD206 aggregation. Uncovering the underlying mechanisms behind this temporal shift may produce novel insights into disease interactions and inform therapeutic developments for aging BC populations.

Basic Science Postbaccalaureate Fellow

POSTER #68

INTRODUCTION OF MOLECULAR DYSFUNCTION TO HUMAN GRANULOSA CELLS BY LOSS OF ARID1A AND OXIDATIVE STRESS INDUCTION

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Granulosa cells are characterized as somatic cells of the ovary. Existing initially as primordial follicles, they undergo a series of temporally spaced differentiation to develop as mural or cumulus cells. This specialization allows them to act in both oocyte development and maturation to provide nutrients, hormones, and growth factors as well as in endometrial remodeling through hormone release to promote the continuous cycle of the thickening, decidualizing, shedding and repair of the endometrium. We believe that molecular dysfunction in these cells promotes malignancy in ovarian endometriosis-associated lesions by epigenetic destabilization of AT-rich interactive domain 1A (ARID1A) and gain of oncogenic stress destabilizing cell cycle properties and the tumor microenvironment (TME). Cellular senescence is unique type of cell cycle arrest defined by tumor suppressor gene activation, lack of apoptosis, but the presence of a secretory phenotype (SASPs) in aged or damaged cells. The generation and accumulation of pro-inflammatory cells introduces a new system of tissue remodeling, immune modulation, and resistance to therapies as it relies on its own chronically inflamed feedback loops. Our mouse model study of conditional knockout of ARID1A and knock-in of oncogenic Kras^{G12D} in the surface epithelium and granulosa cells by Amhr2 Cre (Arid1A^{Flox/Flox}, Lox-Stop-Lox Kras^{G12D}, Amhr2^{Cre/+}) led to the formation of spontaneous ovarian endometriomas and an increased inflammatory phenotype (Control, 4.3±0.56; AKA, 9.7±1.9, P≤0.05, n=11). Furthermore, transcriptomic overlay of the endometriomas with human data showed a significant overlap of dysregulated pathways relating to hormone release, TGFβ signaling, and senescence-associated genes.

To understand how loss of ARID1A and cellular stress impacts granulosa cell behavior, shARID1A clones of human ovarian granulosa-like tumor (KGN) cells of and non-luteinized human granulosa cells (HGrC1) have been generated to represent a malignant and benign model, respectively, and will be treated with 5uM H₂O₂ for 2 hours every 24 hours for 4 days before being evaluated for senescence-associated markers and gene expression. Preliminary immunofluorescence evaluation of ARID1A knockdown in shARID1A KGN cells show a decreased expression compared to parent and 12Z cells. Furthermore, shARID1A KGN show increased proliferation and migration rates compared to parent cells (n = 8, p < 0.01; n = 18, p < 0.001). Continuous H₂O₂ sub-lethal dosing of KGN cells show a 60% increase in β-galactosidase expression compared to untreated cells as evaluated colorimetrically (n = 5, p < 0.001) and by flow cytometry (n = 6, p < 0.0001). Further work is underway across separate biological replicates to confirm knockdown by western blot and qPCR analysis before cell proliferation capacity is continued. With this work, we will elucidate models for granulosa cell senescence, which will warrant further assessment of genomic instability driven senescence in both our human and genetically modified mouse models of endometriosis and endometriosis-associated ovarian cancers.

POSTER #69

UNCOVERING FKBP5'S ROLE IN THE DEVELOPMENT OF CANCER CACHEXIA

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Background: Cancer is frequently accompanied by cachexia, an uncured multi-organ wasting syndrome that leads to impaired musculoskeletal health, physical function, and overall survival. FK506-binding protein-51 (FKBP5) is a co-chaperone and mediator of the stress response, with recent findings suggesting a regulatory role of FKBP5 in metabolic disease. Here, we investigated FKBP5's role in the development of cancer cachexia. **Methods:** Expression of skeletal muscle FKBP5 was assessed in tumor-bearing and control mice. Cultured C2C12 myotubes were overexpressed with FKBP5, or FKBP5 was silenced in myotubes exposed to tumor conditioned media. 10-week-old male wildtype (WT) C57BL/6J or FKBP5^{-/-} (KO) mice (n=6-12/group) were intrasplenically injected with MC38 colorectal cancer cells (1.25×10^5) to induce liver metastases (LM), while controls received saline (Sham). Animals were assessed for indices of cachexia, including muscle mass, torque, and bone quality. **Results:** Expression of FKBP5 was significantly increased ($p < 0.05$) in skeletal muscle of mice bearing lung (LLC), pancreatic (KPC), and colorectal (C26, HCT116, MC38) cancers. *In vitro* studies demonstrated that FKBP5 overexpression caused myotube atrophy (-26%; $p < 0.0001$) and reductions in AKT/mTOR, while gene set enrichment analysis exposed drivers of autophagic mechanisms and proteasome-mediated-ubiquitin-dependent-protein-catabolic process. In contrast, genetic blockade of FKBP5 protected against cancer-induced myotube atrophy (+20%; $P < 0.0001$). WT-MC38 hosts displayed reductions in muscle mass (quadriceps: -25%; gastrocnemius: -24%; tibialis anterior: -20%; $p < 0.0001$) and plantarflexion torque ($p < 0.01$) compared to WT-Sham. Meanwhile, muscle mass and torque were unchanged in KO-MC38 compared to KO-Sham. Supporting the phenotype, we observed significant upregulation of *murfl* and *atrogen1* in the skeletal muscle of WT-MC38, which was counteracted in KO tumor hosts. Additionally, RNA sequencing revealed improvements in gene networks related to mitochondrial function and oxidative phosphorylation in skeletal muscle of KO-MC38. Like muscle, cortical bone volume fraction (Ct.BV/TV: -9%) and trabecular bone volume fraction (Tb.BV/TV: -33%) were reduced in WT-MC38 compared to WT-Sham, while differences were not observed between KO-MC38 and KO-Sham. **Conclusion:** Our data suggests that targeting FKBP5 protects against cancer cachexia, representing a new strategy to improve musculoskeletal health and quality of life in cancer patients.

POSTER #70

REGULATION OF ALLOGENEIC T CELL-DRIVEN ACUTE GRAFT-VERSUS-HOST DISEASE BY A LONG NONCODING RNA-ENCODED NONCANONICAL MICROPROTEIN

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Allogeneic hematopoietic stem cell transplantation (allo-HSCT) is a therapy used to treat high-risk hematological malignancies or immune disorders. Acute graft-versus-host disease (aGVHD) limits allo-HSCT efficacy and affects up to 50% of allo-HSCT patients. Mechanisms regulating allo-T cells are incompletely understood, thus identifying novel ways of targeting allo-T cells may lead to improved outcomes.

Long noncoding RNAs (lncRNAs) are key immune regulators, and their expression is context and cell-type specific. Some lncRNAs, particularly those localized in the cytoplasm and able to access translation machinery, harbor small open reading frames (sORFs) which code for noncanonical microproteins (<100 amino acids). These lncRNA-encoded microproteins can function independent of their parent transcripts, creating “bifunctional” genes whose RNA and protein products can act concordantly or separately. However, the role of microproteins in T cell function remains unknown.

We previously identified Regulatory Long coding RNA of T Cells (*ReLoT/LINC00402*) as a potential biomarker for aGVHD whose expression in T cells inversely correlates with aGVHD severity in multiple cohorts of allo-HSCT patients (Peltier et al., *Sci. Transl. Med.*, 2021). Since *ReLoT* localizes in the cytoplasm, we performed an in vitro transcription-translation assay revealing a 10 kDa microprotein. Translation initiation bioinformatic tools suggested an open reading frame in the 5' portion of the second exon was the most likely to be translated and encoded a 10 kDa microprotein. Deletion and frameshift overexpression constructs along with an anti-sORF antibody confirmed this.

To determine the *ReLoT* microprotein dependent versus RNA molecule dependent functions, we generated mutant overexpression constructs able to drive each individually or together. These were then used to generate Jurkat T cells lines expressing each construct. Jurkat cells were chosen for this because they do not express endogenous *ReLoT*. Results from these cells showed that both the lncRNA and microprotein independently suppressed IL-2 production after T cell receptor activation ($p < 0.01$), without affecting ERK phosphorylation. These data suggest *ReLoT* has both coding and noncoding (i.e. bifunctional) roles on T cell receptor responses.

The above overexpression constructs were then introduced into primary murine T cells via lentivirus transduction of hematopoietic stem cells, followed by adoptive transfer into congenic mice. Splenic T cell proliferation following anti-CD3/28 stimulation was inhibited by overexpression of the microprotein only ($p < 0.05$). Conversely, CRISPR/Cas9 disruption of the sORF in human T cells increased proliferation ($p < 0.01$).

In an allo-T cell driven murine aGVHD model (B6→BALB/c), recipients of microprotein overexpressing T cells but not T cells overexpressing a construct only capable of driving *ReLoT* RNA molecule-dependent effects, exhibited reduced mortality, clinical scores (mortality $p = 0.01$; score $p = 0.007$), and acute GVHD histopathology scores in the small intestines. Preliminary measurement of the accumulation of T cell subsets known to either augment or inhibit aGVHD suggested that the *ReLoT* microprotein inhibits accumulation of inflammatory Ror γ ⁺ Th17/Tc17-like cells in the spleen.

These results suggest *ReLoT* is a bifunctional regulator of T cells. However, in the setting of aGVHD, these data suggest the *ReLoT* microprotein but not the RNA molecule is the primary molecular product of this

bifunctional gene mitigating allo-T cell-driven aGVHD. Thus, augmenting or maintaining the expression or function of the *ReLoT* microprotein may be a novel approach for limiting aGVHD or other T cell-mediated disorders.

Basic Science

Research Technician

POSTER #71

HALOFUGINONE PRESERVES HUMAN HSC STEMNESS VIA A GCN2-DRIVEN INTEGRATED STRESS RESPONSE.

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Halofuginone preserves human HSC stemness via a GCN2-driven integrated stress response

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Hematopoietic stem and progenitor cells (HSPCs) sustain blood cell production throughout life, and their transplantation is an important therapy for various hematologic and non-hematologic diseases. However, successful transplantation outcomes, including durable hematopoietic reconstitution, depend on the infusion of HSPCs that retain quiescence and long-term functional potency. Ex vivo manipulation, post transplantation inflammatory stress, and subsequent chemotherapy exposure can result in a loss of potency. Our lab recently showed that the small molecule GW4869 activates the integrated stress response (ISR) and improves long-term HSPC function by likely training cells to better tolerate inflammation, though the mechanisms underlying this cellular memory remain unknown.

Here, we utilize halofuginone hydrobromide (HF), a pharmacologic activator of the ISR via the GCN2 kinase, to further investigate how ISR activation in HSPCs improves long-term transplantation outcomes after ex vivo culture. Human CD34⁺ cells were isolated from mobilized peripheral blood and cultured ex vivo with HF (20 nM) or vehicle control for up to 14 days. Cultured cells were assessed daily for growth and viability. Immunophenotypic HSPC composition was assessed by flow cytometry alongside measurements of proteostasis, mitochondrial reactive oxygen species (ROS), cell cycle status, and cellular metabolism. Functional potency was evaluated by colony-forming unit (CFU) assays and in vivo repopulation capacity was assessed by NSG xenotransplantation. HF significantly reduced HSPC proliferation, misfolded/aggregated proteins, and mitochondrial ROS relative to controls. HF also increased the frequency of immunophenotypic long-term HSCs (CD34⁺CD38⁻CD45RA⁻CD90⁺CD49f⁺) and short term-HSCs (CD34⁺CD38⁻CD45RA⁻CD90⁻CD49f⁻), consistent with preservation of stemness during prolonged culture. Preliminary data in NSG mice show modest improvements in human CD45⁺ cell chimerism of HF-treated grafts. Together, these data support HF as a clinically relevant ex vivo conditioning strategy to mitigate culture-associated stress and preserve stem-like HSPC features, with the potential to improve transplantation outcomes.

Basic Science Research Technician

POSTER #72

ESPLIT AND ISWAP: STREAMLINED STRATEGIES AND DESIGN PRINCIPLES FOR CONDITIONAL ALLELE ENGINEERING USING SHORT ARTIFICIAL INTRONS

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Deciphering gene function is central to cancer biology, yet generating conditional alleles for in vivo interrogation remains a major technical barrier to mechanistic and translational progress. Short artificial introns (SAIs) have emerged as powerful tools to simplify allele design and characterization, yet practical guidelines for their implementation remain limited. Here, we describe two streamlined strategies, eSPLIT (exon split) and iSWAP (intron swap), that enable efficient generation of conditional alleles using SAIs. In eSPLIT, a compact cassette containing essential intronic elements flanked by loxP sites is integrated within an exon, whereas in iSWAP, a native intron is replaced with an SAI. In the absence of Cre recombinase, the SAI is recognized as an intron and removed by the splicing machinery, allowing normal gene expression. Following Cre-mediated recombination, excision of critical intronic sequences disrupts splicing, leaving residual sequences, including stop codons in all reading frames, thereby causing premature translation termination and gene inactivation. We optimized these approaches by generating SCYL1 alleles in human cells and validating their general applicability in mouse models across multiple genes. We further extended the approach to the FLP-FRT system in vivo. By defining practical rules for SAI placement, we establish a robust and scalable framework for engineering conditional alleles, with broad utility in functional genomics and disease modeling.

Basic Science

Research Technician

POSTER #73

UNTARGETED METABOLOMICS REVEALS 1-METHYLNICOTINAMIDE AS A METABOLIC HALLMARK OF TUMOR-ASSOCIATED MACROPHAGE POLARIZATION.

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Macrophages are the most abundant immune cells in the solid tumor microenvironment. In cancer, macrophages adopt a maladaptive phenotype known as tumor-associated macrophages (TAMs). The phenotypic transition of macrophages into TAMs is coupled with metabolic reprogramming. While the pro-tumorigenic role of TAMs is clearly defined, there are specific metabolic changes that occur within these transitions that remain underexplored. In this study, we utilized untargeted liquid chromatography-mass spectrometry (LC-MS) metabolomics profiling on murine bone marrow-derived macrophages (BMDMs). We compared the metabolic signatures of unstimulated macrophages against those polarized toward a TAM-like state using tumor-conditioned media. Our analysis revealed a significant metabolic divergence between the two populations, most notably characterized by the consistent upregulation of 1-methylnicotinamide (1-MNA) in the TAM populations. This enrichment of 1-MNA may suggest a redirection of nicotinamide metabolism and a potentially altered nicotinamide N-methyltransferase (NNMT) activity within the tumor microenvironment (TME). These significant alterations may suggest a metabolic reprogramming within the TME, critical for understanding tumor progression and a potential novel therapeutic intervention.

Basic Science Research Technician

POSTER #74

NFX1-123 IS A NOVEL RNA-BINDING PROTEIN WITH AN ALTERED BINDING PROFILE AFTER HPV 16E6 INFECTION

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The high-risk Human Papillomavirus type 16 (HPV 16) is the most common oncogenic HPV with persistent infection leading to cervical and oropharyngeal squamous cell carcinomas. Its E6 protein (16E6) has been shown in complex with the E3 ubiquitin ligase, E6-associated protein (E6AP), to bind cellular proteins for post-transcriptional gene regulation. This gene regulation using processes such as alternative splicing is largely controlled by RNA-binding proteins (RBP). NFX1-123 is an isoform of the nuclear transcription factor, X-box binding 1 (NFX1) gene that is highly expressed in HPV-associated cancers and is a protein partner of 16E6. It contains a N-terminal PAM2 motif necessary for PABPC binding, a central domain with a PHD/RING domain and six zinc-like fingers required for DNA binding, and a unique C-terminus with two additional zinc-like fingers and an R3H domain with single-stranded nucleic acid binding capabilities. In this study, we used enhanced cross-linked immunoprecipitation (eCLIP) to identify NFX1-123 as a novel RBP that has its binding pattern influenced by 16E6. We found that fewer RNAs bound to NFX1-123 in the presence of 16E6, but there were more genes in RNA processing and DNA replication pathways. Using cells with deleted R3H domains or mutated PAM2 motifs, we also found that these are important for the RNA binding function of NFX1-123 regardless of 16E6 presence. This study expands the library of RBPs interacting with HPV 16 through validation of NFX1-123 as a novel RBP.

Basic Science

Research Technician

POSTER #75

PI3K SIGNALING DRIVES THE EXPANSION OF A NOVEL METAPLASTIC LINEAGE IN THE STOMACH

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Gastric cancer is the 5th most common type of cancer and is the 4th leading cause of global cancer deaths. With more than 80% of gastric cancer cases attributed to *Helicobacter pylori* (*Hp*), this infection is the largest risk factor for this disease. *Hp* infection causes gastric inflammation that, in certain individuals, leads to metaplasia (pre-cancer) and then cancer development. PI3K (phosphoinositide 3-kinase) signaling activates AKT to drive numerous cell functions, including growth, proliferation, and survival. Gastric cancers commonly harbor PI3K genetic alterations, but without a better understanding of the functional significance of PI3K pathway activation during gastric carcinogenesis, we have a limited ability to target this signaling pathway to prevent or treat gastric cancer. PTEN is a tumor suppressor that plays a critical role in regulating cell growth and survival by dephosphorylating proteins that interact with the PI3K/AKT signaling pathway. Transgenic mice lacking PTEN can be used as a tool to induce PI3K activity and assess its effects on metaplasia development. We developed a transgenic mouse model in which tamoxifen administration triggers loss of *PTEN* in two gastric epithelial cell types believed to be the cell of origin of metaplasia in the stomach. Mice were infected with *Hp* or mock-infected and tamoxifen was administered to delete *PTEN*. Mice were euthanized after 8-18 weeks and stomachs were assessed by immunostaining and *in situ* hybridization, qRT-PCR, and western blotting. We found that loss of *PTEN* was sufficient to elicit metaplastic phenotypes in the stomach, including expansion of metaplastic pit (foveolar) cells expressing the pit cell mucin MUC5Ac and the intestinal mucins MUC4 and MUC13. *Hp* infection exacerbated disease progression driven by *PTEN* loss, with an increase in proliferation and higher pathology scores compared to mock-infected mice. In a simplified model system, we found that *Hp* infection of AGS gastric cancer cells, which harbor two activating mutations in the PI3K catalytic subunit *PIK3CA*, induced expression of the metaplastic pit cell markers *MUC5Ac* and *MUC4*. Treatment with the PI3K inhibitor LY294002 prior to *Hp* infection prevented the induction of these mucins. Taken together, these results demonstrate that *Hp* infection synergizes with PI3K signaling in gastric epithelial cells, leading to metaplasia. In ongoing work, we are evaluating whether the pan-AKT inhibitor capivasertib can reverse metaplastic pit cell phenotypes *in vivo*.

Basic Science *Research Technician*

POSTER #76

INVESTIGATING NOVEL NUCLEAR RECEPTOR BINDING MOTIFS IN HEAT SHOCK FACTOR 1 AND ITS INTERACTION WITH ESTROGEN-RELATED RECEPTOR A IN CANCER

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Breast cancer is the most frequently diagnosed human cancer and is the second-leading cause of cancer deaths in women. Heat Shock Factor 1 (HSF1) is a transcription factor that plays a critical role in the heat shock response and the unfolded protein response. HSF1 activity has been shown to cause worse prognosis in breast cancer. Estrogen Related Receptor α (ERR α) is an orphan nuclear receptor that mitigates metabolic stress by altering mitochondrial biogenesis. It is unknown if these two pathways have any interaction. We have observed a cooperation and a physical interaction between HSF1 and ERR α . Co-expression of both HSF1 and ERR α led to changes in transcriptional activity of both transcription factors as assessed by luciferase reporter assays. Additionally, we observed a physical interaction between HSF1 and ERR α via co-immunoprecipitation. ERR α interacts with its coactivator PGC1 α via a Leucine-rich nuclear receptor binding motif. We identified several novel nuclear receptor binding motifs in the HSF1 protein sequence. To determine the function of this motif in HSF1, residues in these motifs were mutated by replacing the flanking leucines with alanines to render them non-functional. Sequencing data confirmed that mutagenesis of each of these nuclear receptor binding motifs was successful. These mutants can be used in subsequent studies to discover a deeper understanding of the interaction of HSF1 with nuclear receptors. To investigate the effect of these of these alanine substitutions, a co-immunoprecipitation was performed using HEK-293FT cells which have endogenous HSF1 knocked out, ERR α overexpressed, and transfected with the mutants. The results from this study highlight two distinct mutations which were able to uncouple the physical interaction between HSF1 and ERR α . These mutants will undergo further investigation to determine the molecular and phenotypic effects of the loss of this physical interaction. Immunoprecipitation-mass spectrometry will be conducted to identify other binding partners and unveil if these sites are involved with interacting with other nuclear receptors. We will express mutant HSF1 in the triple-negative breast cancer cells line MDA-MB-231 and repeat previous experiments. We will also examine changes to proliferation, viability, and colony formation to determine the biological consequences of this interaction. These studies identify a novel interaction between two significant transcription factors in breast cancer, and future studies will elucidate the molecular underpinnings of their interaction.

Basic Science

Undergraduate Student

POSTER #77

INVESTIGATING THE RELATIONSHIP BETWEEN ATF4 EXPRESSION AND HSF1 REGULATION.

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Glioblastoma is the deadliest form of adult brain cancer. HSF1 is a transcription factor responsible for implementing the heat shock response in cells under environmental stress. Additionally, GCN2 is a serine/threonine kinase in the integrated stress response (ISR) that detects low amino acid concentrations in cells and leads to activation of the ISR pathway. Upon activation, GCN2 phosphorylates eIF2 α , which decreases global translation but allows for increased translation of specific proteins such as the stress response transcription factor ATF4. NXP800 is a compound with antitumor effects that was first identified as an HSF1 pathway inhibitor as it caused the loss of HSF1 protein, but has more recently been shown to activate the GCN2 pathway. However, the question remains whether this dual effect was the result of an unknown overlap between HSF1 and the GCN2 pathway. Our data indicate that the loss of HSF1 protein caused by agonism of the GCN2 pathway is caused by the shutdown of HSF1 translation by eIF2 α . In addition to eIF2 α , we also determined whether ATF4 affects HSF1. Over-expression of ATF4 led to a decrease in both HSF1 protein levels and its transcriptional targets. Additionally, we found that knock-down of ATF4 leads to a partial rescue of the NXP800-induced decrease in HSF1 activity. Therefore, our results indicate that translational repression of HSF1 by eIF2 α and activity of ATF4 both contribute to the downregulation of HSF1. This study indicates a direct mechanistic link between the GCN2 and HSF1 stress response pathways that could be taken advantage of by therapeutic approaches for glioblastoma therapies.

Basic Science Undergraduate Student

POSTER #78

THE ROLE OF PP2A IN THE REGULATION OF AMPHIREGULIN IN PANCREATIC DUCTAL ADENOCARCINOMA

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Pancreatic ductal adenocarcinoma (PDAC) has the worst 5-year survival rate of all major cancers, therefore there is an urgent need for further investigation and research. Approximately 90% of PDAC tumors are driven by activating mutations in the small GTPase KRAS. However, direct therapeutic targeting of KRAS has proven challenging, as tumors often develop resistance to KRAS-directed inhibitors. Protein Phosphatase 2A (PP2A) is a serine/threonine phosphatase that can negatively regulate downstream effectors of KRAS. Because of this, PP2A is a point of interest when considering potential PDAC therapeutic strategies. The PP2A complex is made up of 3 subunits: scaffolding (A), regulatory (B), and catalytic (C). There are many different regulatory B subunits, and the target of the PP2A complex is determined by which particular B subunit is incorporated into the complex. The role of the various regulatory B subunits is not well understood in the context of PDAC. The B56a regulatory subunit was previously described as a tumor suppressor, but our lab found that in PDAC, B56a overexpression promotes tumorigenic phenotypes by inducing the EGFR signaling pathway. Amphiregulin, an EGFR ligand, plays a key role in mediating these effects through EGFR activation. Here we focus on elucidating the mechanism of regulation between PP2A-B56a and the EGFR-AREG signaling axis to understand how this pathway is modulated in PDAC.

Basic Science

Undergraduate Student

POSTER #79

PP2A-B56 α AND ITS ROLE IN EPITHELIAL-TO-MESENCHYMAL TRANSITION AND METASTASIS IN PANCREATIC CANCER

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Pancreatic cancer is the leading cause of cancer-related deaths, with a 5-year survival rate of 13.3%. Over 80% of pancreatic patients present with metastasis upon diagnosis, further lowering the survival rate to 3.2%. Understanding metastasis is pivotal to understanding how to treat pancreatic cancer. The initiating step of metastasis is epithelial-to-mesenchymal transition (EMT), where epithelial cells lose their cell-cell adhesion and start to increase their mobility, transforming them into a more migratory and invasive mesenchymal cell. Epidermal growth factor receptor (EGFR) and Kirsten Rat Sarcoma (KRAS) pathways drive cell growth, whose signaling mutations are commonly hyperactivated and drive pancreatic cancer. PP2A is a major ser/thr phosphatase in the cell and is a major tumor suppressor that negatively regulates many pathways, including the KRAS and EGFR pathways. Previous work in lung cancer has shown that when there is a decrease of PP2A-B56 α , the cancer cells enter EMT and begin the process of metastasis as shown by an increase in Vimentin, the mesenchymal cell-state marker, and a decrease in E-cadherin, the epithelial cell-state marker. Data from our lab suggests that PP2A-B56 α plays a role in maintaining an epithelial cell-state within lung cancer, however, it is unknown whether this is the case in pancreatic cancer as well. Assessing the role of PP2A-B56 α in regulating cell state in pancreatic cancer is crucial in understanding metastasis in one of the most fatal cancers. Early preliminary data from our lab suggests that when PP2A-B56 α is completely removed from pancreatic cancer cells, there is a change in morphology, characteristic of an EMT. In my project, I will be using HPAFII pancreatic cancer cells with CRISPR knockout of B56 α to analyze the changes that occur within the cell via qRT-PCR, western blot, and immunofluorescent analysis in addition to phenotypic assays.

Basic Science

Undergraduate Student

POSTER #80

KRAS LOSS OF HETEROZYGOSITY MODULATES RESPONSE TO HP INFECTION IN GASTRIC CANCER CELLS

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Introduction: Nearly 80% of stomach cancers are attributed to infection with the bacterium *Helicobacter pylori* (*Hp*), which elicits chronic stomach inflammation that induces mutations. Almost 10% of gastric cancers have a mutation in *KRAS*, a component of the RAS/MAP kinase pathway that promotes cell growth and proliferation. AGS cells, a commonly used gastric cancer cell line, have one wild-type (WT) and one constitutively active *KRAS* allele (G12D). *KRAS* loss of heterozygosity (LOH) is loss of the WT allele, leaving only the mutated allele. *KRAS* LOH is associated with an aggressive and invasive phenotype in pancreatic cancer but has not been thoroughly investigated in gastric cancer. We therefore sought to isolate the contribution of WT *KRAS* vs. *KRAS* G12D in driving gastric cancer phenotypes in AGS cells.

Methods: CRISPR/Cas9 was used to delete WT *KRAS* from AGS cells, as confirmed by long-read sequencing. Gene and protein expression in the clones (expressing *KRAS* G12D only) was compared to the parent line (expressing *KRAS* WT/G12D) at baseline and 24 hours post-infection with *Hp*. Xenograft studies in female NRG mice were used to investigate tumorigenic potential of the cell lines.

Results: Loss of WT *KRAS* did not impact *KRAS* protein levels, but total RAS protein levels were increased, suggesting increased HRAS and/or NRAS. In the parent line, *Hp* infection decreased *KRAS* expression, whereas clones with loss of WT *KRAS* had increased *KRAS* expression after *Hp* infection. We also investigated the epidermal growth factor receptor, EGFR, because EGFR expression can be modulated by *KRAS* activity. Loss of WT *KRAS* led to decreased EGFR gene and protein expression compared to the parent line. *Hp* infection caused increased total and phospho-EGFR in all cell lines, with greater increases in the clones than in the parent line. Xenograft studies are ongoing; thus far both the parental line and the clones have engrafted, and tumor size is being monitored. Bioinformatic analysis of the prevalence of *KRAS* LOH in human gastric cancer samples is ongoing.

Conclusion: Our findings from cell culture studies suggest that WT *KRAS* contributes to downregulating the RAS/MAPK pathway in gastric cancer cells, possibly through reducing upstream activation of EGFR, and that WT *KRAS* affects the response to *Hp* infection in these cells. Xenograft studies will reveal whether WT *KRAS* contributes to tumor formation.

Basic Science

Undergraduate Student

POSTER #81

INVESTIGATING PP2A-B56 α AS A TUMOR SUPPRESSOR IN KRAS-DRIVEN NSCLC

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Lung cancer is the third most common type of cancer, with Non-Small Cell Lung Cancer (NSCLC) making up 87% of cases. The two most common mutations in NSCLC are in KRAS and EGFR, however targeting KRAS has proven difficult, so we must find another part of the pathway to target for therapeutic intervention. One approach is targeting PP2A, a critical “off switch” phosphatase that negatively regulates oncogenic signaling pathways, that regulates downstream effectors of EGFR and KRAS. Previous studies found that the specific regulatory B subunit, B56 α , shows significant tumor suppressive abilities in multiple types of cancer. The PP2A complex is composed of 3 subunits: a scaffolding “A” unit, a catalytic “C” unit, and the regulatory “B” subunit. The differing regulatory “B” subunits determine the specificity of the complex by directing substrate binding and cellular localization of the complex. This project investigates the role of PP2A-B56 α in Human Bronchial Epithelial Cells (HBEC), with a KRAS G12V mutation and p53 knockdown (HBEC-KP). By adding this mutation and knockdown, the cells become partially transformed, yet not fully tumorigenic, thus provide a model for studying early tumorigenic progression. Since B56 α regulates c-Myc, a regulator of growth, proliferation, and survival and known oncogenic transcription factor, we hypothesize that the loss of B56 α will cause these partially transformed cells to decrease regulation of c-Myc and increase their tumorigenic qualities. Findings from our lab indicate that HBEC-KP with an shRNA-mediated knockdown of B56 α show a potential loss of cell identity through a 3D network formation assay, where they appear to lose normal structure and organized epithelial morphology. Phenotypic assays, including low-attachment growth, 3D network formation, transwell migration, proliferation, and clonogenic assays, as well as mRNA and protein expression analysis of relevant signaling and identity markers, will investigate if the loss of B56 α subunit increases tumorigenic qualities and leads to a loss of cell identity in this partially transformed lung epithelial model.

Basic Science

Undergraduate Student

POSTER #82

DEVELOPMENT OF PATIENT DERIVED ADAMANTINOMATOUS CRANIOPHARYNGIOMA CELL LINE

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Adamantinomatous Craniopharyngioma (ACP) is a rare, slow-growing epithelial tumor of the sellar and suprasellar region that predominantly affects children. ACP often causes significant morbidity due to its location near critical vascular structures, including the optic apparatus, hypothalamus, and major cerebral vessels. Tumor growth and recurrence can result in long-term complications such as visual impairment, endocrine dysfunction, and neurocognitive deficits. Current management primarily involves surgical resection and radiation therapy, both of which can lead to substantial long-term complications. Progress towards targeted therapies has in part been limited by a lack of reliable models in vitro that capture the biology of patient tumors. The development of patient-derived tumor models may therefore provide an important tool for advancing research in ACP.

In this study, our main goal was to establish patient-derived ACP cell lines using tumor specimens obtained during surgical resection. Fresh tumor tissue from multiple patients were collected and processed to generate primary cultures. Samples were mechanically and enzymatically dissociated and plated in two-dimensional culture conditions designed to support epithelial tumor cell growth. Cultures were maintained under controlled conditions and monitored over time to assess cell viability, proliferation, and stability in vitro. Serial passaging was performed to evaluate the ability of cultured cells to expand and maintain growth across multiple passages.

Further characterization was performed to assess whether these cultures retain molecular and phenotypic features consistent with ACP. Establishing patient-derived ACP cell lines provides a platform for studying tumor biology and exploring potential therapeutic strategies underlying tumor growth and recurrence.

Developing additional patient-derived models may help address current limitations in ACP research by providing platforms for investigating tumor biology and evaluating potential therapeutic approaches. These models may ultimately support translational studies aimed at identifying more effective and less morbid treatment strategies for patients with this challenging tumor.

Basic Science Undergraduate Student

POSTER #83

DETERMINING THE ROLE OF HDAC2 IN MEDIATING EMT AFTER PP2A-B56A SUPPRESSION IN NON-SMALL CELL LUNG CANCER

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Lung cancer is one of the most commonly diagnosed cancers and a leading cause of cancer deaths worldwide, accounting for an estimated 18% of all cancer deaths. Non-small cell lung cancer (NSCLC) makes up between 80-85% of all these lung cancer cases. NSCLC is extremely lethal as many patients already have metastasis, the spread of cancer from the primary site to additional sites, at the time of diagnosis, making it harder to treat. Metastasis is initiated via epithelial-to-mesenchymal transition (EMT), where stationary epithelial cells transition into mesenchymal cells by losing their cell-cell adhesion proteins, such as E-cadherin, and gaining the ability to metastasize. While the transcriptional regulation of EMT is well characterized – such as the knowledge of transcription factor ZEB1 driving EMT, its post-translational regulation remains understudied. Protein phosphatase 2A (PP2A) is a heterotrimeric serine/threonine phosphatase with scaffolding (A), regulatory (B), and catalytic (C) subunits. The B subunit determines the substrate specificity of PP2A and consists of four different families. Our lab studies PP2A-B56a, which acts as a tumor suppressor in cancer. Our lab has found that suppression of PP2A-B56a in EGFR-mutant NSCLC leads to EMT both in vitro and in vivo. This is associated with a persistent loss of E-cadherin that is not rescued even after acute reintroduction of PP2A-B56a, suggesting epigenetic regulation of E-cadherin in EMT. Histone Deacetylase 2, HDAC2, is a member of the NuRD chromatin remodeling complex. HDAC2 is recruited by the transcription factor ZEB1 to repress the gene coding for E-cadherin to drive EMT. Interestingly, our proteomic analyses showed HDAC2 to be significantly upregulated with PP2A-B56a knockdown. Our proteomic analyses show increased phosphorylation at S394, which is a site that is important for HDAC2 binding activity, and it resides close to a PP2A-B56a binding motif. Our hypothesis is that suppression of PP2A-B56a leads to increased HDAC2 phosphorylation, leading to it constitutively repressing E-cadherin expression in EMT in NSCLC. To determine how HDAC2 contributes to EMT after PP2A-B56a suppression, we will pharmacologically inhibit HDAC2 in HCC827 cell lines to assess effects on E-cadherin and other downstream EMT targets. We will also investigate its binding at the E-cadherin locus after PP2A-B56a suppression using techniques such as ChIP-qPCR.

Basic Science Undergraduate Student

POSTER #84

TRAINING ESTHETICIANS TO PROVIDE CANCER PREVENTION EDUCATION: PRELIMINARY EVALUATION FINDINGS

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Introduction: Skin and cervical cancers are significant, yet highly preventable, public health issues in the United States. Skin cancer is the most common cancer nationwide, particularly among adolescents and young adults (AYAs), with rates on the rise. Human papillomavirus (HPV), the most common sexually transmitted infection, is responsible for nearly all cervical cancer cases despite the availability of an FDA-approved vaccine. Young women aged 18-34 years are at a high risk for these cancers due to high rates of sunburn, indoor tanning use, and suboptimal HPV vaccination uptake. Identifying trusted messengers and novel settings for cancer prevention is critical to improving prevention behaviors in this priority population. Estheticians (i.e., licensed skincare professionals) have regular and recurring visits with their clients. Prior research indicates that estheticians frequently observe a variety of skin-related issues during their waxing services, including evidence of skin cancer, severe sunburns, and genital warts. Previous work also indicates estheticians have an interest in supporting their clients' health but report limited training, knowledge, and confidence in initiating health-related conversations.

Methods: To address this, we implemented an IUSCCC-funded pilot research study designed to increase estheticians' knowledge of skin cancer and cervical cancer and build confidence in discussing related topics with clients. Six, one-hour virtual training sessions were conducted with a team of estheticians (N=10) from across Indiana and included education on skin cancer awareness and detection and HPV and cervical cancer prevention. Participants completed brief online surveys before and after training completion. Surveys assessed HPV-related and skin cancer knowledge and confidence; perceived preparedness in addressing client health concerns and discussing cancer prevention; perceived ability to make a difference in client health outcomes; and perceived client trust in the esthetician.

Results: A total of six estheticians completed the training (100% women). Across most domains, post-session scores were higher than pre-session, highlighting the potential effectiveness of the educational sessions. The largest observed changes were in self-reported skin cancer confidence and perceived client trust. On scales from 1-20, mean skin cancer confidence scores increased from 10.9 to 18.2 (Cohen's $d=1.60$), while perceived client trust increased from 14.6 to 18.6 (Cohen's $d=1.25$). HPV-related confidence and preparedness to discuss cancer prevention topics with clients showed similar positive changes between the pre- and post-session tests.

Discussion: These preliminary findings suggest that tailored cancer education trainings may increase estheticians' perceived confidence and readiness to engage clients in conversations about cancer prevention. While this pilot study was not designed to test statistical significance, current findings suggest promise in engaging estheticians as community-based partners in cancer prevention initiatives. Future work will involve conducting focus groups with both estheticians and clients to better understand what kinds of intervention initiatives may be most useful in the salon setting.

Behavioral Graduate Student

POSTER #85

COMPLEMENTARY AND ALTERNATIVE MEDICINE USE AMONG BREAST CANCER SURVIVORS: INSIGHTS FROM SOCIAL MEDIA COMMUNITY DISCUSSIONS

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Title. Complementary and Alternative Medicine Use Among Breast Cancer Survivors: Insights from Social Media Community Discussions

Author. Xue-Jing Liu, Danny Valdez, Eric R Walsh-Buhi

Introduction. Breast cancer survivors frequently experience a range of physical and psychological challenges during and after treatment. Many patients turn to complementary and alternative medicine (CAM) to manage symptoms and improve quality of life. However, little is known about how breast cancer survivors discuss CAM use, motivations, and concerns in online peer-support communities, such as Reddit, one of the most popular social media.

Method. A qualitative content analysis was conducted on 100 posts randomly selected from Subreddit r/breastcancer. Posts were manually coded using a structured codebook to capture post characteristics, treatment experiences, symptoms, functional status, use, reasons for use, and concerns about CAM. Descriptive statistics were used to summarize coding results.

Results. A total of 100 posts were analyzed, contributed by 99 breast cancer survivors and one family member (age range 23-58 years). Posts primarily involved sharing personal experiences (n = 83), seeking medical information (n = 79), and seeking emotional support (n = 44). Conventional treatments frequently discussed included chemotherapy (n = 52), surgery (n = 35), and radiation therapy (n = 27), while targeted therapy (n = 18), hormone therapy (n = 22), immunotherapy (n = 8), and supportive medications (n = 28) were mentioned less often.

Commonly reported symptoms included anxiety or stress (n = 27), pain (n = 25), fatigue (n = 20), and musculoskeletal symptoms (n = 17). Emotional (n = 71) and physical (n = 51) functioning impairments were the most frequently described functional impacts. Informational needs were the most prominent unmet need (n = 84), followed by decision-making support (n = 47), and emotional needs (n = 42).

Attitudes toward CAM were largely neutral (n = 43) or mixed (n = 20), with fewer positive (n = 19) and negative (n = 9) views. Frequently mentioned types included dietary supplements (n = 28), mind-body practices (n = 27), and physical therapies (n = 27). Posts most often described using CAM to manage symptoms or treatment side effects (n = 69) and to improve quality of life (n = 49). Overall, 56 posts reported current use, with massage (n = 8), yoga (n = 7), and vitamin use (n = 6) most commonly mentioned. Concerns related to CAM centered on uncertainty about how to choose among options (n = 37), doubts about efficacy (n = 18), safety concerns (n = 11), and potential interactions with conventional treatments (n = 10).

Conclusion. Reddit reveals that breast cancer survivors frequently use CAM as a complementary strategy to manage symptoms and improve quality of life. However, many survivors also report uncertainty about selecting appropriate options and express concerns about effectiveness and safety. These findings highlight the importance of providing accessible, evidence-based information and improving patient-provider communication regarding CAM use in cancer care.

Behavioral Graduate Student

POSTER #86

CO-DESIGNING A MOBILE HEALTH PROTOTYPE TO SUPPORT ORAL ANTICANCER MEDICATION MANAGEMENT IN PATIENTS WITH BREAST CANCER

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Background

The growing number and use of oral anticancer medications (OAMs) is shifting an increasing amount of medication management responsibility from clinicians to patients. Historically, most cancer treatment occurred in institutional settings through intravenous infusions, with trained healthcare professionals overseeing much of the medication management tasks, including dosing, monitoring, and coordination. However, OAMs are self-administered at home, requiring patients to manage complex regimens, monitor side effects, and navigate access challenges at home. Existing interventions are often focused on education but inconsistent in their effectiveness. Building on findings from a prior pilot study that identified key barriers to medication management and informed the development of an initial intervention concept, this study aimed to further refine an early-stage prototype intervention to support OAM management among patients with breast cancer.

Methods

We applied a participatory co-design approach informed by prior pilot data to iteratively refine a mobile health intervention. Four structured co-design sessions were conducted with breast cancer patients recruited from an outpatient clinic in a federally qualified health center. Eligible participants were adults diagnosed with breast cancer and currently prescribed OAMs. Co-design sessions involved guided discussions to explore lived experiences with OAM use, including topics related to medication adherence, side effect management, and financial and insurance-related challenges. Participants reviewed and interacted with wireframes and draft feature concepts, provided feedback on content clarity and usability, and prioritized desired functions. Insights from each session were synthesized and integrated into subsequent prototype revisions. Session notes and session recordings were reviewed to identify recurring themes and inform iterative design modifications.

Results

Six patients participated in the four co-design sessions, along with two researchers who served as facilitators and contributors to the design panels. Reported medications included tamoxifen, anastrozole, and ribociclib. The iterative co-design process led to the development of a refined mobile app prototype with five core functions: (1) daily medication management to digitally organize and track medications; (2) a side effect tracker providing tailored self-management guidance; (3) insurance navigation support offering clear next steps for common coverage issues, including an embedded educational module explaining treatment-related and insurance terminology; (4) an online peer support community; and (5) resources function aligned with previously identified patient needs. Participants reported that the integrated functions addressed major gaps in their medication management experience and responded positively to the prototype's perceived usefulness and relevance.

Conclusions

Leveraging prior pilot data and participatory co-design methods, we developed patient-centered, multi-component mobile intervention prototypes targeting key challenges in OAM management. Future research will evaluate feasibility, alignment with clinical workflows, and preliminary efficacy in improving medication adherence and patient-reported outcomes.

Behavioral Graduate Student

POSTER #87

COMPARATIVE VALIDITY, RESPONSIVENESS, AND MINIMALLY IMPORTANT DIFFERENCE OF FEAR OF CANCER RECURRENCE SCALES IN BREAST CANCER SURVIVORS

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Objectives/purpose:

Fear of cancer recurrence (FCR) is a prevalent and distressing concern among breast cancer survivors. Despite widespread use of five FCR measures—the Fear of Cancer Recurrence Inventory (FCRI; total and severity short form), FCR-7, FCR-4, and Concerns About Recurrence Scale (CARS)—no gold standard has emerged, and few studies have evaluated them side by side in intervention trials. This study assessed reliability, validity, responsiveness, minimally important difference (MID), and discriminative accuracy of these measures.

Sample and setting:

Secondary data were used from a U.S.-based, three-arm randomized controlled trial (NCT05364450) comparing Acceptance and Commitment Therapy, Cognitive Behavioral Therapy, and enhanced usual care. Participants ($N = 384$; mean age = 55.8 years) were women with stage I–IIIA breast cancer (completed treatment ≤ 5 years prior to enrollment; screened positive for clinical FCR). Most were non-Hispanic White (80.5%) or Black (13.8%) and college-educated (61.2%).

Procedures:

Baseline and post-intervention data were analyzed. Internal consistency (α), convergent and construct validity (r), standardized response means (SRMs), between-group effect sizes, MID estimates (anchor- and distribution-based), and area under the curve (AUC) values were examined. A post-intervention participant-reported global FCR improvement item was used for anchor-based analyses to classify participants as “Worse,” “Same,” or “Better.”

Results:

All measures demonstrated acceptable internal consistency (α s = 0.70–0.93) and convergent validity (r s = 0.73–0.94). Construct validity was demonstrated by small to moderate correlations with related symptoms and quality of life. All measures differentiated participants reporting global FCR improvement at post-intervention from those unchanged (SRMs = 0.73–0.84). Between-group effect sizes were small (0.21–0.32) and similar for shorter and longer measures. MID estimates ranged from 1 (FCR-4) to 7 (FCRI-Total). AUCs showed comparable accuracy across measures in detecting any and significant global improvement.

Conclusion and clinical implications:

FCRI-Total, FCRI-Severity, FCR-7, FCR-4, and CARS are valid, reliable, and responsive tools for assessing meaningful FCR change in clinical trials and survivorship care.

Behavioral Graduate Student

POSTER #88

LEVERAGING MAMMOGRAPHY TO INCREASE LUNG CANCER SCREENING: EVALUATION OF A PILOT PRINT-TO-WEB OUTREACH INTERVENTION

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Background: Lung cancer (LC) remains the leading cause of cancer death among women in the United States, claiming more lives annually than breast and ovarian cancers combined. Annual lung cancer screening (LCS) by low-dose computed tomography (LDCT) detects LC at an earlier, more curable stage; yet uptake remains severely low. Wide mammography adoption offers an opportunity to promote LCS to a receptive population. Yet, practical implementation such as identifying and engaging eligible women requires scalable tools that overcome LCS barriers.

Method: In this single-center feasibility study (March–July 2024), LCS outreach materials were integrated into routine screening mammography workflows at a Midwestern hospital. Tailored flyers that promoted LCS with a “Lung Check” gain-framed message, addressed psychological and logistical barrier and adopted to a grade 3 reading level, were mailed with mammography appointment’s reminder and result’s letter and displayed in mammography’ setting waiting and changing rooms. Materials directed participants to a navigator phone line or via QR code and web link to a website offering education, eligibility assessment, and -if eligible- self-referral to a centralized LCS program. Feasibility was defined as successful workflow integration and participant engagement, measured by reach (web visits or calls) and efficacy (eligibility assessment completion, self-referral and LCS completion). Secondary outcomes were LCS eligibility rates among respondents and completed LCS results.

Result: Among 1,276 mammography recipients, 269 (21.1%) responded (19 phone, 150 QR, 100 direct web visits). Of those, 133 completed an LCS-eligibility assessment. Twelve individuals (12/133,9.0%) were identified as LCS-eligible, 9/12 (75.0%) self-referred, and all 9 completed the shared decision-making (SDM) visit. Of the 9 who completed SDM, 2 had a chest CT in the past year, and 7 completed a LDCT. Findings were negative/benign (Lung-RADS 1–2) in five and very suspicious (Lung-RADS 4B) in two, one of whom was diagnosed with stage IIB lung adenocarcinoma. Based on the observed eligibility rate (9.0%), we estimated that approximately 115 individuals within the full mammography cohort would have met LCS eligibility criteria. The completion of LDCT by seven individuals therefore represents an estimated absolute increase in LCS uptake of 6.1% (7/115) attributable to the intervention.

Conclusion: A print-to-web outreach strategy embedded in mammography workflows was feasible and showed preliminary effectiveness in identifying LCS-eligible women and activating self-referrals. Future

work should evaluate wider implementation and cost-effectiveness, especially when deployed with other outreach modalities.

Behavioral Post-Doctoral/Medical Fellow

POSTER #89

POLYUSE OF CANNABIS, OTHER SUBSTANCE, AND SYMPTOM MANAGEMENT MEDICATIONS IN A REPRESENTATIVE SAMPLE OF U.S. ADULTS WITH CANCER, 2021-2024

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PURPOSE: Adults with cancer frequently face persistent challenges including pain, sleep disturbance, and anxiety. While opioids and benzodiazepines are standard treatments, interest in cannabis as an alternative or adjunct therapy is increasing. This study examines polyuse patterns of cannabis and other symptom management medications in a nationally representative sample of adult cancer patients.

GAPS: While patient-reported surveys suggest a significant opioid-sparing effect—where cannabis allows patients to reduce opioid dosage while maintaining pain control—a recent meta-analysis of clinical trials found little to no effect on actual opioid dose reduction in oncology populations. Also, a population-based study of patients with early-stage cancer indicates that cannabis frequently serves as an adjunct therapy, rather than as a substitute, in a pattern of polyuse, particularly among patients with higher symptom burdens. Also, the patterns of substance and medication polyuse in recent samples of cancer patients have been understudied while some poly- or co-use poses clinical safety concern. In addition, there is a lack of research comparing how ‘oxy’ opioids (e.g., oxycotin, oxycodone, oxymorphone) versus ‘hydro’ opioids (e.g., hydrocodone, zohydro, hydromorphone) interact with cannabis use patterns in cancer patients.

METHODS: We analyzed four recent waves (2021-2024) of the National Survey on Drug Use and Health (NSDUH). The sample included 185,427 adults, with a sub-extraction of 1,593 individuals reporting a cancer diagnosis in the past year. Individuals with non-melanoma skin cancer (n=1,396) have been dropped from the cancer sub-sample due to noncomparability to other cancers in terms of survival rates, likelihood of metastasis, and rates of cannabis use. Categorical outcomes were compared using Rao & Scott adjusted chi-square tests, and logistic regressions examined adjusted odds ratios (aORs) of past-year cannabis use predictors in cancer patients. Benjamini-Hochberg adjustments were used to control for Type I error.

RESULTS: Cancer patients were significantly more likely than non-cancer counterparts to use 'oxy' opioids (19.9% vs. 7.9%), 'hydro' opioids (26.0% vs. 13.5%), benzodiazepines (16.2% vs. 9.4%), and pain relievers (51.7% vs. 26.6%) (all $p < 0.001$). Among cancer patients, 17.2% reported past-year cannabis use. Cannabis users had higher prevalence of using benzodiazepines, 'hydro' opioids, and sedatives. In fully adjusted models, past-year pain reliever use (aOR=3.22), sedative use (aOR=2.68), tobacco use (aOR=2.23), and binge drinking (aOR=2.41) remained significant predictors of cannabis use.

CONCLUSION: Cancer patients exhibit high levels of polyuse of cannabis and other substances, including opioids and sedatives. Further research is required to evaluate the safety and efficacy of polyuse of cannabis and other substances, and assess the use of cannabis as an adjunct (i.e., polyuse) versus a substitute to reduce opioid intake (i.e., opioid-sparing effect).

Behavioral Post-Doctoral/Medical Fellow

POSTER #90

WHAT DO RURAL CANCER CAREGIVERS NEED? A SCOPING REVIEW

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Introduction

Informal caregivers (caregivers) in rural areas face many stressors that affect their mental and physical health. This scoping review synthesizes the current needs of cancer caregivers in rural areas to inform future research and interventions.

Methods

Five databases (PsycInfo, CINAHL, EMBASE, PubMed, and Family & Society Studies Worldwide) were queried to identify studies examining mental health and quality-of-life outcomes in rural caregivers of cancer patients worldwide, using the JBI scoping review methodology. A priori criteria included rural caregivers of adult cancer patients in peer-reviewed studies from 2005 to 2025.

Results

Database searches yielded 337 records. After title and abstract screening, 134 articles were reviewed, and of these, 35 met a priori criteria. The most frequent problems identified among rural cancer caregivers were mental health issues (depression and anxiety), loneliness or isolation, fatigue and physical issues, provider issues, and financial issues, especially related to travel, balancing duties, and cultural differences between rural populations and urban providers. Most articles were from outside the U.S.; however, results from rural caregivers in the U.S. described needs reflected in the literature from other countries.

Discussion

Given the increased utilization of caregivers in the U.S. and the rural burden of cancer, it is crucial to identify caregivers' unmet needs. Home-based cancer care is increasing, necessitating additional studies to determine needs and develop effective interventions for rural caregivers. Rurally competent care from healthcare providers who interface with caregivers of their cancer patients is vital, as caregivers are well-positioned to partner with providers to improve the well-being of those patients.

Behavioral Post-Doctoral/Medical Fellow

POSTER #91

FACTORS ASSOCIATED WITH WILLINGNESS TO USE HPV SELF-COLLECTION

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Introduction/Background: Low-income women in the U.S. experience nearly double the mortality rate of cervical cancer (CC) and lower CC screening rates compared to high-income women. Self-collection for human papillomavirus [HPV] testing is a new CC screening method recently approved by the Food and Drug Administration for in-clinic use. However, little is known about the women who would be most likely to use self-collection. The current study had two objectives: (1) compare sociodemographic characteristics (age, education, race/ethnicity), healthcare utilization, and theoretical constructs (HPV knowledge, perceived importance, and Diffusion of Innovation [DOI] constructs [perceived advantages, complexity, and self-efficacy]) between women who were UTD with CC screening and those who were not UTD; (2) among women who were not UTD with screening, identify factors associated with willingness to complete vaginal HPV self-collection using bivariate analyses and a multivariable regression model.

Methods: Eligible women aged 25 to 65 years old from an online research panel were recruited via an email invitation to complete a cross-sectional survey. Participants were given a description and images of the self-collection process. Bivariate analyses compared characteristics of women who were UTD vs. women who were not UTD with CC screening. Bivariate analyses and multivariable linear regression models were used to identify the willingness of participants to use vaginal self-collection among women not UTD with CC screening.

Results: Participants (n = 1,997) had a mean age of 48.8 years (SD = 11.3). Women not UTD with CC screening reported significantly fewer healthcare visits, higher perceived advantages, higher self-efficacy, lower HPV knowledge, and lower perceived importance of CC screening compared to women who were UTD. Among women not UTD, healthcare utilization, race/ethnicity, and all theoretical constructs were significantly associated with willingness to use vaginal self-collection bivariate with self-efficacy, advantages, and importance showing the strongest associations. In the hierarchical regression, sociodemographics explained 3.3% of variance (p = .042), and addition of theoretical constructs significantly improved model fit (R² = .151, Δ R² = .118, p < .001). Perceived complexity (β = .29, p < .001), Hispanic ethnicity (β = .14, p = .003), and perceived importance of CC screening (β = .13, p = .009) emerged as significant independent predictors of willingness to use self-collection.

Conclusions: While multiple theoretical constructs were bivariate associated with willingness to use self-collection, complexity was the strongest factor, suggesting that reducing the difficulty of self-collection may be most impactful for increasing uptake among women not UTD with screening. Notably, Hispanic women reported greater willingness, indicating cultural receptivity that could be leveraged in targeted outreach. These findings support the development of multi-media educational materials to accompany self-collection kits.

Behavioral Undergraduate Student

POSTER #92

“EMPOWERHER: A COMMUNITY-ENGAGED BREAST CANCER PREVENTION MODEL FOR LATINO WOMEN”

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Background: Latino women face persistent inequities in breast cancer prevention driven by social determinants of health (SDoH), including limited access to culturally relevant information, healthcare services, preventive medications, and digital health resources. Community-level myths and fatalistic beliefs about breast cancer, often shaped by migration experiences, language barriers, and historical exclusion from healthcare systems, continue to hinder engagement in preventive behaviors linked to modifiable risk factors.

Methods: This study draws on qualitative findings from focus groups conducted with Latino women in underserved communities to examine perceptions, myths, and structural barriers related to breast cancer prevention. These insights informed the development of *EmpowerHER*, a culturally tailored breast cancer prevention educational project focused on behavior change related to modifiable risk factors, including physical activity, nutrition, alcohol use, and preventive health-seeking behaviors. A core implementation strategy is the partnership with trusted civil society organizations (CSOs), particularly La Casa de Amistad, which serves as a community anchor to facilitate engagement, reduce technology and health literacy gaps, and support navigation of healthcare and medication access.

Results: Focus group participants demonstrated limited awareness of evidence-based prevention strategies and frequently attributed breast cancer risk to emotional stress, physical injury, destiny, or heredity. Structural barriers, including lack of insurance, cost of care and medications, fear related to immigration status, language discordance, and low confidence in using digital health tools, were consistently identified as obstacles to adopting healthy lifestyles. CSOs emerged as critical intermediaries, offering trusted spaces for learning, culturally concordant communication, and hands-on support with technology use and healthcare navigation. These findings directly shaped *EmpowerHER*'s design, which integrates community-based education delivered through CSOs, practical skill-building for healthy lifestyle adoption, and guidance to access preventive services within constrained systems.

Conclusions: Breast cancer prevention for Latino women requires approaches that simultaneously address behavior change and the structural conditions shaping access to health and technology. By embedding prevention education within trusted CSOs and explicitly targeting health and digital literacy alongside SDoH, *EmpowerHER* represents an equity-centered, scalable model for breast cancer prevention. This approach leverages community trust to counter myths, empower women to adopt healthier lifestyles, and reduce persistent disparities in breast cancer risk and outcomes among Latino communities.

Community friendly research poster Faculty

POSTER #93

ECONOMIC IMPACT OF FLUOROPYRIMIDINE-BASED CHEMOTHERAPY LEADING TO HOSPITALIZATION: A RETROSPECTIVE REAL-WORLD CLAIMS DATA ANALYSIS

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BACKGROUND:

Pharmacogenetic testing for variants in *DPYD* can reduce fluoropyrimidine (FP)-toxicities, but additional real-world data regarding the clinical and economic impact of FP-induced toxicities is needed to inform *DPYD* testing policies.

METHODS:

We conducted a retrospective observational study using the de-identified commercial claims database of patients in the US with commercial insurance from the Merative™ MarketScan® Commercial Database. We identified patients who appeared to be hospitalized due to FP-induced toxicities using 3 different strategies of ICD-10 codes. They are patients who were hospitalized within 50 days of starting their first FP dose from 01/01/2016 to 12/31/2022 had enrollment records within 180 days before or one year after the first FP dose. Using 2022 data, we identified ICD-10 codes present in patients with adverse event (AE)-related diagnoses and used those to select patients in all years; patients were excluded if their admission codes indicated non-FP toxicity related causes. All groups of patients were filtered using same FP-induced toxicity codes with different data approaches: Group A and B with inpatient admissions data (summary for each admission) and group C with inpatient services data (details on individual services); two data are linked with the case IDs. For group A, patients with definite indicator for presenting toxicity-related ICD-10 codes on admission, but group B was sorted by the indicator with not 'No' (i.e., missing/unknown, unreported/not used, clinically undetermined, or yes). For group C, all diagnosis codes of individual services were considered to apply FP-induced toxicity codes. To evaluate the variability, we compared the number of patients identified using three selection strategies differing in the frequencies of hospitalizations, days in intensive care units, and emergency department usage.

RESULTS:

59,731 patients were prescribed FPs. We identified 1,027 patients (~1.7 % of new FP prescriptions) who were hospitalized most likely due to FP-related toxicities (see table). Costs associated with the hospitalizations are being calculated and will be reported as part of the presentation.

CONCLUSION:

These real-world data illustrate the substantial healthcare burden caused by the FP-related toxicities.

Community friendly research poster

Graduate Student

POSTER #94

DOES HOW WE MEASURE DNA CHANGES IN TUMORS AFFECT ACCESS TO IMMUNOTHERAPY?

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Tumor mutational burden (TMB) is a test used in cancer care to help doctors decide whether a patient may benefit from a certain immunotherapy. These drugs help the immune system recognize and attack cancer cells. TMB measures how many DNA changes (mutations) are found in a tumor. The idea is simple: tumors with more mutations may create more “flags” (called neoantigens) that help the immune system see the cancer more clearly. TMB is reported as the number of mutations per DNA length, known as a megabase (1 million units of DNA). The TMB number used to decide if a cancer patient is eligible for immunotherapy is 10 or more mutations per 1 million units of DNA.

The most accurate way to measure TMB compares a patient’s tumor DNA to their normal (non-cancer) DNA. This helps doctors tell which mutations are truly from the cancer. However, often in cancer care only the tumor sample is tested. In these cases, doctors use public DNA databases and reference genomes (example: GRCh38) to estimate the TMB of individual. There are many examples of DNA and the example to calculate TMB can change the number.

There is concern that not all reference DNA and public DNA databases represent people of patients with African Descent DNA. This means that some inherited DNA differences in patients of African Descent may not be properly recognized as normal. As a result, tumor-only testing could mistakenly count inherited changes as cancer mutations. This could make TMB appear higher than it truly is and possibly affect treatment decisions. Newer and more complete reference genomes, such as GRCh38 and the Human Pangenome, include more genetic diversity. These updated tools may improve how accurately TMB is measured across people from different backgrounds.

In our study, we are analyzing DNA from 366 cancer patients, including 72 self-reported Black participants and 294 self-reported White participants. We are comparing TMB results using older and newer reference genomes. Early findings show that TMB values change depending on which reference genome is used.

Our goal is to understand whether newer reference genomes provide more accurate and fair TMB results across patients with different ancestries. This work aims to improve how TMB is calculated so that immunotherapy decisions are based on the most accurate information possible for every patient.

Community friendly research poster

Graduate Student

POSTER #95

HEALTH COMMUNICATION AND CANCER SCREENING DISPARITIES AMONG HISPANIC WOMEN IN TIPPECANOE COUNTY: A DIFFUSION OF INNOVATIONS PERSPECTIVE

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Background: Hispanic women in the United States continue to experience significant disparities in breast and cervical cancer screening. While structural and individual barriers are well documented, less is known about how health communication influences screening behaviors within specific local contexts. Tippecanoe County, Indiana, a mid-sized Midwestern community with a growing Hispanic population, has documented persistent screening gaps, highlighting the need to understand how preventive care information is shared and experienced.

Objective: This qualitative study examined how health communication influences screening participation among Hispanic women in Tippecanoe County using Diffusion of Innovations (DOI) theory.

Methods: Four focus groups were conducted with Hispanic women ages 21–65, along with semi-structured interviews with healthcare providers, and community organization representatives. Inductive thematic analysis identified patterns in participants communication experiences, followed by a deductive analysis mapping findings onto DOI stages: knowledge, persuasion, decision, implementation, and confirmation.

Results: 16 women and 14 healthcare and community representatives participated. Screening participation was not a single event but a process that developed over time influenced by communication across interpersonal, community, and institutional settings. In the knowledge stage, women described confusion about eligibility and inconsistent provider messaging. During persuasion, screening was viewed as important but complicated by cost concerns, insurance navigation, language barriers, and immigration-related fears. Trusted social networks and community spaces reinforced participation, while providers emphasized clinical reminders and logistical access. This misalignment between institutional messaging and women lived realities often delayed decision and implementation. Sustained engagement was strengthened through culturally grounded, peer-based communication rather than formal healthcare messaging alone.

Conclusions: Overall, screening disparities in this community reflect not only access barriers but also gaps in how preventive information is communicated between healthcare systems and women lived experiences. Using DOI theory clarified where diffusion slowed and where communication strategies could better support preventive care.

Community friendly research poster

Graduate Student

POSTER #96

INCREASING LUNG CANCER SCREENING THROUGH OPPORTUNISTIC REFERRALS

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Lung cancer is the leading cause of cancer-related death in the United States and the second most diagnosed cancer. Although lung cancer screening can reduce lung cancer mortality by up to 20%, national screening rates remain around 16%—far lower than for other common cancer screenings. Our research group found that many patients who receive another type of screening for coronary artery calcium may be eligible for lung cancer screening but have not yet been screened, illustrating an opportunity to increase lung cancer screening rates within routine healthcare encounters. This article explores how screening referrals made outside of traditional lung cancer screening programs—which we refer to as 'opportunistic referrals'—can serve as a strategy to improve early detection. We aim to highlight practical strategies and resources that can improve awareness and access. We also discuss how policy changes and system-level strategies can support more equitable screening practices, particularly in underserved populations.

Community friendly research poster

Medical Student

POSTER #97

LEVERAGING MAMMOGRAPHY TO INCREASE LUNG CANCER SCREENING: PILOT EVALUATION OF A PRINT-TO-WEB OUTREACH INTERVENTION

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Background: Lung cancer (LC) remains the leading cause of cancer death among women in the United States, claiming more lives annually than breast and ovarian cancers combined. Annual lung cancer screening (LCS) by low-dose computed tomography (LDCT) detects LC at an earlier, more curable stage; yet uptake remains severely low. Wide mammography adoption offers an opportunity to promote LCS to a receptive population. Yet, practical implementation such as identifying and engaging eligible women requires scalable tools that overcome LCS barriers.

Method: In this single-center feasibility study (March–July 2024), LCS outreach materials were integrated into routine screening mammography workflows at a Midwestern hospital. Tailored flyers that promoted LCS with a “Lung Check” gain-framed message, addressed psychological and logistical barrier and adopted to a grade 3 reading level, were mailed with mammography appointment’s reminder and result’s letter and displayed in mammography’ setting waiting and changing rooms. Materials directed participants to a navigator phone line or via QR code and web link to a website offering education, eligibility assessment, and -if eligible- self-referral to a centralized LCS program. Feasibility was defined as successful workflow integration and participant engagement, measured by reach (web visits or calls) and efficacy (eligibility assessment completion, self-referral and LCS completion). Secondary outcomes were LCS eligibility rates among respondents and completed LCS results.

Result: Among 1,276 mammography recipients, 269 (21.1%) responded (19 phone, 150 QR, 100 direct web visits). Of those, 133 completed an LCS-eligibility assessment. Twelve individuals (12/133,9.0%) were identified as LCS-eligible, 9/12 (75.0%) self-referred, and all 9 completed the shared decision-making (SDM) visit. Of the 9 who completed SDM, 2 had a chest CT in the past year, and 7 completed a LDCT. Findings were negative/benign (Lung-RADS 1–2) in five and very suspicious (Lung-RADS 4B) in two, one of whom was diagnosed with stage IIB lung adenocarcinoma. Based on the observed eligibility rate (9.0%), we estimated that approximately 115 individuals within the full mammography cohort would have met LCS eligibility criteria. The completion of LDCT by seven individuals therefore represents an estimated absolute increase in LCS uptake of 6.1% (7/115) attributable to the intervention.

Conclusion: A print-to-web outreach strategy embedded in mammography workflows was feasible and showed preliminary effectiveness in identifying LCS-eligible women and activating self-referrals. Future work should evaluate wider implementation and cost-effectiveness, especially when deployed with other outreach modalities.

Community friendly research poster

Post-Doctoral/Medical Fellow

POSTER #98

IDENTIFYING BEST PRACTICES TO REMOVE BARRIERS TO QUALITY BREAST CANCER PATIENT CARE

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In 2026, approximately 2.1 million Americans are expected to be diagnosed with cancer, highlighting the growing need to address barriers that affect access to timely and high-quality care (Cancer Facts & Figures 2026, American Cancer Society). Despite advances in treatment, marginalized patients often encounter structural, cultural, and psychosocial barriers that reduce access to high-quality care and worsen treatment experiences. This study examines how unmet social needs, medical mistrust, and coping strategies relate to communication, shared decision-making, adherence, and engagement across the cancer care continuum. Although research has identified social and structural barriers in cancer care, it remains unclear whether unmet social needs predict lower engagement in cancer screening and treatment and whether medical mistrust predicts poorer communication, reduced shared decision-making, and lower adherence to recommended care. These relationships are rarely examined together, limiting the identification of actionable, trauma-informed targets to improve cancer care experiences. To address this gap, the present study tests the following hypotheses: (H1) Higher levels of unmet social needs will be associated with lower engagement in cancer screening and treatment; (H2) Greater medical mistrust will be associated with less shared decision-making, poorer communication quality, and lower adherence to recommended care. This study will conduct a literature review, qualitative interviews, and a mixed-methods survey of cancer patients, their support persons, and clinicians. Measures include unmet social needs (SDOH-related barriers), medical mistrust, coping strategies, communication quality, shared decision-making, adherence, and care engagement (screening participation and treatment follow-through). Qualitative data will contextualize barriers across stakeholder groups and identify modifiable clinical practices aligned with trauma-informed care. We expect that participants reporting higher unmet social needs will demonstrate lower screening and treatment engagement, while those reporting higher medical mistrust will report poorer communication, reduced shared decision-making, and lower adherence to recommended care. Differences are also expected across patients, support persons, and clinicians in how barriers to cancer care are perceived. Although prior literature suggests that medical mistrust and unmet social needs contribute to cancer care disparities, limited research integrates these factors with modifiable clinical interaction processes. By examining these domains within a single framework and comparing perspectives across patients, support persons, and clinicians, this study aims to identify trauma-informed and patient-centered clinical strategies, particularly in communication and shared decision-making, that can reduce engagement barriers, improve treatment adherence, and ultimately enhance cancer care experiences for marginalized patients.

Community friendly research poster

Post-bacc student

POSTER #99

USING THE INDIANA STATE CANCER REGISTRY (ISCR) TO EVALUATE A COMMUNITY CANCER CLUSTER NEAR AN EPA SUPERFUND SITE

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Background: The Indiana Department of Health (IDOH) updated its cancer cluster investigation methods in alignment with the CDC's revised Guidelines for Examining Unusual Patterns of Cancer and Environmental Concerns. Following community concern regarding an EPA Superfund site in Martinsville, Indiana, a cancer cluster investigation was conducted to evaluate whether cancer incidence in the community exceeded expected levels.

Methods: The IDOH Cancer Cluster Inquiry Team conducted a 20-year retrospective analysis. Utilizing data from the Indiana State Cancer Registry (ISCR), age-adjusted rates and standardized incidence ratios (SIRs) were calculated for 5-year intervals to assess temporal and geographic patterns. Martinsville was evaluated alongside Morgan County and statewide rates. Eight cancer types that are known to be associated with the environmental exposure of concern, as well as ten additional cancer types were included in the analysis.

Results: No consistent evidence of an unusual cancer pattern was identified in Martinsville. Morgan County historically has one of the highest overall cancer incidence rates in Indiana, complicating interpretation of elevated rates at sub-county levels. While there were significant results, both Martinsville and Morgan County shared similar results or results were inconsistent between timeframes. Among cancers known to be associated with environmental exposure, both Morgan County and Martinsville showed elevated rates of kidney cancer while Martinsville alone had elevated incidence of myeloma and leukemias during certain timeframes. For cancer types not known to be related to the EPA site, both areas showed elevated melanoma and oropharyngeal cancer incidence, while Morgan County showed elevated rates of thyroid, prostate, and breast cancers during some timeframes. Where a statistically significant higher rate was found, no conclusions regarding causation between the environmental site of concern and the results could be made.

Conclusion: Cancer cluster investigations are frequently conducted in response to community concern, yet they rarely show causal links between environmental exposures and cancer outcomes. With hundreds of different cancers all with varying risk factors and latency periods, defining a cancer cluster within a specific area, period, and population of similar cases is extremely difficult. Proving causality between a case of cancer and a specific exposure present substantial analytic challenge. The cancer registry is a valuable tool but has limitations in addressing community concerns. This investigation shows the importance and complexity of clear communication to the public regarding the interpretation constraints of cancer cluster analyses.

Population Science/Epidemiology

Epidemiologist

POSTER #100

ASSOCIATION OF AFLATOXIN WITH DETECTION/PERSISTENCE OF ONCOGENIC HUMAN PAPILLOMAVIRUS AND LACK OF HIV IMMUNE CONTROL AMONG KENYAN AND UGANDAN WOMEN

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BACKGROUND. Cervical cancer is caused by persistent infection with oncogenic (high-risk, or HR) HPV, and is common among women living in sub-Saharan Africa. Only a small percentage of HR-HPV-infected women will develop this malignancy, suggesting that cofactors are important. HIV is one of these cofactors. Dietary aflatoxin exposure may also be a co-factor, as these carcinogen and immunosuppressive agents contaminate corn in sub-Saharan African countries. We performed studies to assess the association of aflatoxin exposure on detection and persistence of HR-HPV, and on immune control of HIV.

METHODS. Demographics, behavioral data, plasma, and cervical swabs were collected from Ugandan and Kenyan women. Cervical HPV detection and persistence were compared between women with or without detectable plasma aflatoxin and in relation to plasma aflatoxin concentration. In a separate analysis in HIV-infected Kenyan women (all receiving ART), aflatoxin detection was compared between women with undetectable or detectable HIV viral load measurements. In a third analysis in HIV-infected and HIV-uninfected Ugandan women, HPV 16 and HPV 18 detections were compared in relation to plasma aflatoxin concentrations.

RESULTS. In HIV-uninfected Kenyan women, 49 women (57.0%) had aflatoxin detected. Aflatoxin detection and concentration were strongly associated with detection of HR-HPV types, as well as higher risk of persistent detection of any HPV type, HR-HPV types, and HR-HPV types not included in the 9-valent HPV vaccine. In a separate study, aflatoxin was detected in plasma from 100% of 114 Ugandan women. A higher plasma aflatoxin concentration was associated with persistent detection of HPV 16 and HPV 18 after controlling for HIV status and other factors. In a separate study of 56 HIV-infected Kenyan women (all receiving ART), 8 women (14.3%) had a detectable HIV viral load (mean 12,439 copies/mL). Aflatoxin was detected in 23 of these 56 women (41.7%); 17 of 48 women (35.4%) with an undetectable HIV viral load, and 6 of 8 women (75.0%) with a detectable HIV viral load ($p=0.053$). The mean plasma aflatoxin concentration for these women was 0.0403 pg/ μ L; 0.0341 pg/ μ L and 0.0771 pg/ μ L for women with undetectable or detectable HIV viral loads, respectively ($p=0.039$).

Conclusions: Aflatoxin was detected in a high percentage of Kenyan and Ugandan women. Aflatoxin detection was associated with detection and persistence of HR-HPV, and with a lack of immune control in HIV-infected Kenyan women. While it has not been proven that aflatoxin exposure increases the risk of cervical cancer, further studies are needed to define a mechanism for these associations.

POSTER #101

LUNG CANCER RISK AND SCREENING INTEREST AMONG PEOPLE EXPERIENCING HOMELESSNESS

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Background: People experiencing homelessness (PEH) have a long-standing higher prevalence of cigarette smoking compared to the general U.S. population, with rates ranging from 68–80% (versus 9.9–11.6% among U.S. adults overall)[1]. Existing interventions for this population have primarily focused on smoking cessation support, and their acute healthcare encounters such as emergency department visits [2]. In contrast, other lung cancer risk factors and preventive strategies, particularly lung cancer screening (LCS) remains poorly understood and under-explored among this population.

Methods: A cross-sectional survey was conducted at two large homeless shelters (Horizon House and Lafayette Transitional Housing Center) in Indiana. Surveys were administered in-person with questions read aloud by trained study researchers to accommodate varying literacy levels. Data collected included participants' smoking history, exposure to other lung cancer risk factors, perceptions of lung cancer risk, the interest in LCS and eligibility for LCS based on United States Preventive Services Task Force (USPSTF) guidelines.

Results: A total of 143 participants were enrolled between November 2025 and February 2026. Of those, 60.8% were male and 39.2% were female. 54.2% were White and 32.9% were Black, and 93.7% identified as non-Hispanic. The mean age was 51.8 years. Participants reported experiencing homelessness an average of three times, with a mean lifetime duration of 41.5 months. Regarding housing status, 53.1% were staying in shelters, 37.8% were unsheltered, and 9.1% reported others such as staying with family, friends, or in hotels. Most participants (82.5%) had ever smoked, of whom 88.1% were current smokers. Several additional lung cancer risk factors were reported at rates substantially higher than national averages, including secondhand smoke exposure (85.3% vs. 20.8%[3]), disability (46.2% vs. 12.2% [4]), e-cigarette use (26.6% vs. 6.5%[5]), cigar use (20.3% vs. 3.5%[6]), chronic obstructive pulmonary disease (COPD) (19.6% vs. 6.4%[7]), and first-degree family history of lung cancer (18.2% vs. 6.6%[8]). Among the 118 participants who had ever smoked, 63.6% met USPSTF guideline eligibility criteria for LCS. 83% indicated a preference to receive LCS, and 90% reported they would undergo screening if it were offered through a proposed mobile unit at a shelter setting.

Conclusion: Our findings demonstrate a significantly high smoking prevalence among PEH, consistent with prior reported data. Participants also reported substantially elevated rates of additional lung cancer risk factors compared with national averages. A significant proportion meet eligibility criteria for LCS under USPSTF guidelines and a majority expressed interest in undergoing LCS. Our data further suggests interest in screening through a proposed mobile unit in a shelter-based setting. These findings underscore the need for future research to evaluate the feasibility, acceptability, and appropriateness of implementing mobile LCS programs tailored to this population.

POSTER #102

BENEFITS AND HARMS OF LUNG CANCER SCREENING BY COMORBIDITY BURDEN: EVIDENCE FROM A MULTICENTER COHORT AND MICROSIMULATION MODELING

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Background: Lung cancer remains one of the leading causes of cancer-related mortality in the United States and worldwide. Low-dose computed tomography (LDCT) screening has been shown to reduce lung cancer mortality among high-risk populations and has been recommended by clinical guidelines. However, current approaches for evaluating lung cancer screening effectiveness largely rely on age and smoking history, with limited systematic consideration of comorbidity burden. In real-world populations, individuals eligible for lung cancer screening often have multiple chronic conditions due to heavy smoking histories, increasing their risks of lung cancer and competing mortality, ultimately affecting the long-term benefits from screening. However, participants in screening trials may be generally healthier than the general screen-eligible population, limiting our understanding of how varying levels of comorbidities may affect screening outcomes. Using data from the Personalized Lung Cancer Screening (PLuS) cohort, a contemporary real-world screening population, we evaluated the impact of comorbidities on lung cancer screening outcomes using a microsimulation modeling approach.

Methods: We adapted a previously established lung cancer microsimulation model, the Lung Cancer Interventions and Decision Simulation (LuCID-Sim) model, to simulate lung cancer-related events and other-cause mortality by comorbidity level. We incorporated 13 individual comorbidity conditions (e.g., COPD, hypertension, and cardiovascular disease) and categorized them into three strata (0–1, 2–3, and ≥ 4 conditions). We calibrated the model to reproduce key distributions across comorbidity strata observed in the PLuS study, including lung cancer histology and stage distribution, five-year lung cancer incidence, and five-year all-cause mortality. We then evaluated the long-term benefits from screening, including lung cancer deaths averted and life years gained compared to no screening, and harms, such as false positives and overdiagnosis rates, by comorbidity level under real-world and hypothetical screening adherence rates. Screening efficiency was assessed as benefits per number of screens, and tradeoffs between benefits and harms were measured using the ratio of benefits to overdiagnosis.

Results: The adapted LuCID-Sim model successfully captured the differences in lung cancer incidence, histology and stage distribution, and mortality outcomes across comorbidity strata observed in PLuS. We observed that lung cancer deaths averted per screen increased with the number of comorbidities, whereas life years gained per screen decreased with the number of comorbidities. After accounting for screening harms, the benefit-to-harm ratios, measured either as lung cancer deaths averted or life years gained per overdiagnosed case, decreased with the number of screens.

Conclusions: Screen-eligible individuals with high comorbidity burden may be at elevated risks of lung cancer but also have higher competing mortality, leading to reduced gains in screening benefits, especially in life years gained, as well as higher overdiagnosis rates. Our findings highlight the importance of considering comorbidity burden in lung cancer screening decisions to better balance the benefits and harms of screening.

Population Science/Epidemiology

Graduate Student

POSTER #103

TO KNOW OR TO NOT KNOW?: RETURN OF RESULTS (ROR) PREFERENCES IN GENOMIC TESTING WITHIN THE SUSAN G. KOMEN TISSUE BANK POPULATION

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Title:

To Know or To Not Know?: Return of Results (ROR) Preferences in Genomic Testing within the Susan G. Komen Tissue Bank Population

Background:

The Susan G. Komen Tissue Bank (KTB) is a biobank of healthy breast tissue and blood samples donated to advance breast cancer research. As genomic sequencing becomes increasingly integrated into biomedical research, incidental or secondary findings may reveal clinically actionable genetic variations. Ethical return of results (ROR) requires careful consideration of donor preferences and informed consent protocols.

Objective:

To identify sociodemographic and clinical subpopulations within the KTB cohort that are more or less likely to desire return of genomic results.

Methods:

This study included 1,876 KTB donors who completed IRB-approved consent forms between January and December 2025. ROR preference (“Yes” or “No”) was documented either at the time of tissue donation consent or during medical follow-up. Sociodemographic variables collected included age, race, education, donor history, and mammogram history. Bivariate analyses and multivariable logistic regression models were used to evaluate associations between participant characteristics and ROR preference.

Results:

Overall, 92.1% (n=1,728) of donors indicated they would like to receive genetic results, while 7.9% (n=148) declined ROR. Decreased desire for ROR was observed among women aged 50–59 (adjusted OR 0.51, 95% CI 0.31–0.85) and ≥ 70 years (adjusted OR 0.33, 95% CI 0.13–0.81), compared to women ≤ 39 years. Higher education was significantly associated with increased desire for ROR, including those with a bachelor’s degree (adjusted OR 1.98, 95% CI 1.19–3.31) and advanced degrees (adjusted OR 2.85, 95% CI 1.63–5.00). Participants who indicated ROR preference at initial consent were also significantly more likely to opt for results compared to follow-up respondents (adjusted OR 3.51, 95% CI 1.50–8.40).

Conclusions:

Although most KTB donors prefer return of genomic results, meaningful variation exists across age and educational subgroups. These findings highlight the importance of continued education regarding genomic testing and the need for structured, ethically grounded ROR processes that prioritize clinically actionable findings while respecting donor autonomy. As genomic research expands, systematic and preference-aligned communication strategies will be critical to maintaining donor trust and promoting informed decision-making.

Population Science/Epidemiology

Graduate Student

POSTER #104

LUNG CANCER INCIDENCE IN INDIANA VS. THE UNITED STATES: SEX-SPECIFIC TRENDS AND WIDENING DISPARITIES FROM 2000 TO 2022

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Background: Lung cancer is the second most common cancer and the leading cause of cancer-related deaths for both sexes in Indiana. Despite overall declines in incidence, Indiana continues to report higher rates than national averages, and sex-specific temporal trends within the state are not well characterized.

Objective: Sex-specific temporal trends in lung cancer incidence in Indiana from 2000 to 2022 were analyzed to assess whether the state's disparities compared with national trends have changed over time.

Methods: Indiana lung cancer incidence data were obtained from the Indiana Department of Health Cancer Registry, and population data were extracted from the U.S. Census Bureau. National incidence rates were obtained from the Surveillance, Epidemiology, and End Results (SEER) program. Rates were age-adjusted to the 2000 U.S. standard population. Temporal trends were analyzed using Joinpoint regression to estimate the average annual percent change (AAPC) and identify changes in trends over time. Pairwise comparisons were conducted to assess differences in trends between Indiana and national rates. Age-Period-Cohort modeling analysis is planned to further explore underlying age, period, and cohort effects by sex.

Results: From 2000 to 2022, lung cancer incidence declined significantly among males in both Indiana (AAPC = -2.09%, $p < 0.05$) and the United States (AAPC = -2.57%, $p < 0.05$). Among females, national incidence decreased significantly (AAPC = -0.86%, $p < 0.05$), whereas the trend in Indiana was not statistically significant (AAPC = -0.15%, $p > 0.05$). Joinpoint pairwise comparison indicated that trends differed significantly between Indiana and the U.S. among males ($p < 0.01$) and females ($p < 0.01$), with a steeper decline observed nationally.

In 2000, lung cancer incidence in the U.S. was 27% lower than in Indiana ($p < 0.001$). Sex-specific comparisons showed that U.S. males had a 31% lower incidence than Indiana males, whereas U.S. females had a 20% lower incidence than Indiana females. By 2022, this gap had widened, with the overall U.S. incidence being 35% lower than that in Indiana ($p < 0.001$). In that year, U.S. males had a 38.6% lower incidence than Indiana males, and U.S. females had about a 32.8% lower incidence than Indiana females.

Conclusions: Despite overall declines in lung cancer incidence, Indiana consistently exhibited higher rates than the national average across sexes, and its slower rate of decline led to a widening disparity by 2022. Planned Age-Period-Cohort modeling analysis will further inform the temporal and generational contributions underlying these state-national differences by sex. Future research is needed to better understand the factors driving Indiana's persistently higher incidence and to identify effective prevention strategies that could help reduce the state's lung cancer burden.

Population Science/Epidemiology

Graduate Student

POSTER #105

BREAST CANCER SURVIVORSHIP OUTCOMES BY OBESITY STATUS AND URBANICITY IN THE UNITED STATES FROM THE BEHAVIORAL RISK FACTOR SURVEILLANCE SYSTEM (BRFSS) STUDY (2019-2023)

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Background. Obesity has been associated with poorer health outcomes like increased pain among breast cancer survivors (BCS). Due to their increased risk of obesity, BCS living in rural communities may be at an increased risk of experiencing negative cancer survivorship care outcomes. This population also faces other unique challenges like limited access to medical care, lack of insurance coverage, and lower socioeconomic status. The purpose of this study was to determine differences in Cancer Survivorship Care (CSC) outcomes among BCS of different body weight status across urban and rural communities.

Methods. Breast Cancer Survivors ($N= 3,767$) from the 2019-2023 Behavioral Risk Factor Surveillance System (BRFSS) were analyzed for relationships between body mass index (BMI) and urban/rural status with CSC outcomes. Outcomes related to treatment summary and follow-up plans, insurance and financial navigation, and pain and symptom management. Logistic regressions analyzed the interaction of BMI and urban/rural status on survivorship outcomes while accounting for demographic covariates and complex survey sample weighting.

Results. Compared to normal weight urban BCS, overweight rural BCS were nearly twice as likely to not receive a summary of treatment (adjusted odds ratio = 1.90, 95% confidence interval = [1.00,3.60]). Overweight rural BCS were also nearly twice as likely to not receive follow-up care plans, though the significance of this finding attenuated when adjusting for covariates (unadjusted OR = 2.94 [1.12,7.75]; adjusted OR = 1.97, [0.93, 4.20]). Both urban and rural obese BCS were more likely to lack insurance coverage for cancer treatment, and this relationship persisted for rural obese BCS upon model adjustment (14.67, [3.58, 56.06]). Rural overweight and normal weight BCS were less likely to report pain compared to the normal weight urban BCS, and this relationship persisted for rural normal weight BCS after adjustment (0.44, [0.22, 0.90]).

Conclusions. Rurality uniquely impacts CSC outcomes among BCS of different body statuses. Continuity of care and insurance coverage for rural BCS of higher body weight status is uniquely impacted, which can lead to less equitable care. Rural BCS may need more self-advocacy with healthcare and coverage providers, and rural providers should be aware of inequities impacting BCS.

Population Science/Epidemiology

Graduate Student

POSTER #106

MODIFIABLE FACTORS STRONGLY ASSOCIATED WITH LOW UPTAKE OF LCS IN PUTNAM COUNTY, INDIANA

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Background:

Lung cancer screening with low-dose computed tomography (LDCT) is guideline-recommended for high-risk adults, yet uptake remains low, particularly in rural populations. Limited data exist on community-specific, modifiable factors associated with lung cancer screening non-receipt in rural counties. Putnam County, Indiana has an adult smoking rate of 22.9%, substantially exceeding both the Indiana state average (17.3%) and the U.S. average (14.4%). Lung cancer mortality in the county reaches 61.5 per 100,000 residents, and Putnam County has been identified among 34 Indiana counties with cancer death rates exceeding the state average in prior IU Simon Cancer Center research. Despite this elevated burden, the county received no local tobacco control grant funding between 2021 and 2023.

Objective:

To identify individual, knowledge-based, structural, and healthcare system factors associated with non-receipt of LDCT screening among eligible adults in Putnam County, Indiana. Preliminary survey data from a small cohort of county residents suggest that approximately 80% of screening-eligible adults had never heard of LDCT screening, motivating a more systematic investigation of awareness and related barriers.

Methods:

This study will use a cross-sectional survey design to collect data from adults meeting USPSTF lung cancer screening eligibility criteria. Participants will be recruited through community events and public venues in Putnam County to maximize reach among residents who may not have an active healthcare relationship. The survey will assess screening history, awareness and perceived eligibility, affective factors (including fear of screening or results), access barriers (transportation, cost, time), insurance coverage, and healthcare system factors such as provider recommendation. The primary outcome will be self-reported receipt of LDCT screening (ever vs never). Multivariable logistic regression will be used to identify factors independently associated with screening non-receipt. Sample size targets are based on achieving sufficient precision to estimate the prevalence of key barriers with 95% confidence intervals, enabling comparison with estimates from similar rural populations.

Anticipated Results:

Based on preliminary data from this community and published literature on rural lung cancer screening barriers, we anticipate that lack of awareness of screening eligibility will be the most prevalent barrier. We also anticipate identifying fear of cancer diagnosis and perceived hopelessness — factors documented in prior rural screening literature — as significant affective contributors to non-uptake.

Conclusions:

Findings from this study will inform targeted, community-based interventions to improve lung cancer screening uptake in rural populations. Results will also be used to optimize outreach strategies for lung cancer screening programs in Putnam County, including a mobile LDCT screening initiative through IU Health.

Population Science/Epidemiology

IUB Graduate and incoming medical student at IUSOM

POSTER #107

UNDERSTANDING PERSPECTIVES OF CERVICAL CANCER PREVENTION IN PATIENTS ATTENDING THE INDIANA UNIVERSITY STUDENT OUTREACH CLINIC

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Background: Nearly all cases of cervical cancer (CC) are caused by HPV. Although HPV vaccination and regular screenings serve as crucial interventions in preventing CC, significant health inequities persist. Indiana ranks 10th in the U.S. for CC incidence, over half of new CC cases occur in under screened population and children's HPV vaccination rate is 59.6% (ranks 36th). While programs like Medicaid and Vaccines for Children cover preventative cares, barriers to participation in underserved communities remain poorly understood. This study evaluates the knowledge, attitudes, and barriers regarding CC prevention among families at a free community clinic.

Methods: This is an interim analysis of a cross-sectional survey being conducted at the Indiana University Student Outreach Clinic (IUSOC), a free urban clinic in Indianapolis. Participants completed surveys assessing CC history, knowledge (15-item scale), and perceived barriers. Data were analyzed using descriptive statistics and compared by gender.

Results: The analysis included 84 participants (45 females, 39 males; mean age 52). The cohort was racially and socioeconomically diverse: 39.3% African American, 29.8% Non-Hispanic Caucasian, 20.2% Hispanic; 28.8% were uninsured, and 58.2% were unemployed. No significant demographic differences existed between genders. Among women, 52.3% were underscreened (never or >10 years). Self-reported HPV vaccination was 24.1%. Knowledge scores were low overall, though women scored significantly higher than men (8.7 vs. 5.5 correct answers; $p < 0.001$). The primary barriers to screening were cost (49.4%), lack of provider recommendation (40.8%), and preference for a female physician (39.5%). Regarding pediatric HPV vaccination, barriers included lack of vaccine knowledge (50.0%), cost (45.3%), and lack of provider recommendation (41.9%). For adult vaccination, the main barriers were lack of provider recommendation (51.3%), cost (46.8%), and lack of awareness (45.5%). No significant gender differences were found on these barriers. Notably, high willingness was reported for free IUSOC services, including screening (59.5% of women) and vaccination for children (66.2%) or self (66.7%).

Conclusions: Despite poor baseline knowledge and significant barriers, primarily cost and lack of provider recommendation, there is high patient interest in CC prevention. Free clinics like IUSOC are uniquely positioned to improve health equity by providing direct access to screening, vaccination, and targeted patient education.

Population Science/Epidemiology

Medical Student

POSTER #108

USE OF SUPPORTIVE CARE MEDICATIONS IS ASSOCIATED WITH PANCREATIC CANCER LOCATION AT DIAGNOSIS

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Background: Pancreatic cancer (PC) often presents with non-specific symptoms that vary by tumor location. Tumors in the body and tail often present later and are more commonly associated with pain, nausea, or cachexia. Supportive care medications (SCMs) such as psychotropics, pain relief medications, anti-nausea agents, and appetite stimulants are commonly used to manage these symptoms yet little is known about whether prescribing patterns differ by tumor location. We evaluated whether pancreatic tumor location at diagnosis is associated with subsequent use of SCMs among older adults with pancreatic cancer.

Methods: We conducted a retrospective cohort study using Surveillance, Epidemiology, and End Results (SEER)-Medicare data (2007-2019). The study included patients aged ≥ 65 years old with incident pancreatic cancer and continuous Medicare parts A, B, and D coverage for 3 months before diagnosis. Patients with another cancer diagnosis within 6 months or missing baseline data were excluded. Pre-diagnosis SCM use was ascertained from Medicare Part D prescription claims. Tumor location at diagnosis were categorized as pancreatic head (C250), body (C251), tail (C252), or other (C253, C254, C257, C258, C679). We used multivariable logistic regression to evaluate the association between SCM use and tumor location adjusting potential confounders.

Results: Compared to tumors in the pancreatic head, tumors in the body were associated with higher odds of receiving most SCM classes including psychotropic medications (OR for any psychotropic: 1.13; 95% CI: 1.08-1.19), pain medications (OR for any pain medication: 1.25; 95% CI: 1.19-1.31), anti-nausea agents (OR: 1.08; 95% CI: 1.03-1.13), appetite stimulants (OR: 1.16; 95% CI: 1.11-1.21). Tail tumors were associated with increased use of anxiolytics (OR: 1.07; 95% CI: 1.00 – 1.14) and non-opioid analgesics (OR: 1.25; 95% CI: 1.15-1.37), but lower odds of anti-nausea use (OR: 0.85; 95% CI: 0.81-0.89). Tumors in other locations generally had lower odds of SCM use across nearly all categories.

Conclusion: Pancreatic cancer location at diagnosis is associated with distinct patterns of supportive care medication use. Patients with tumors in the body of the pancreas were more likely to receive symptom-directed therapies than those with head tumors while those with tail or other locations had more variable patterns. These findings suggest that tumor location may influence supportive care needs and should be considered when tailoring symptom management strategies.

POSTER #109

PREDICTORS OF ANTIDEPRESSANT USE IN OLDER ADULTS AFTER PANCREATIC CANCER DIAGNOSIS: A SEER-MEDICARE ANALYSIS

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Background: Depression is highly prevalent among patients with pancreatic cancer (PC), driven by both the biological effects and the profound psychosocial burden of diagnosis. Untreated depression in this setting contributes to poorer quality of life, reduced treatment adherence, increased symptom burden, and worse survival. Prior work has suggested that sociodemographic inequities may shape access to supportive care services, yet little is known about how these factors operate specifically within older adults with PC. We aimed to characterize demographic, clinical, and socioeconomic predictors of antidepressant use following pancreatic cancer diagnosis, with the goal of identifying inequities and informing more patient-centered approaches to supportive care delivery in this high-risk population.

Methods: We conducted a retrospective cohort study using Surveillance, Epidemiology, and End Results (SEER)-Medicare data among patients aged ≥ 65 years diagnosed with primary PC between 2007 and 2019. Antidepressant use was identified from Medicare Part D claims up to 24 months post-diagnosis. We compared patient characteristics by antidepressant use status. Multivariable logistic regression models estimated adjusted odds ratios (ORs) and 95% confidence intervals (CIs).

Results: Among 81,535 patients with PC, 23.5% (N=19,181) received antidepressants post-diagnosis. Compared with those living in the highest-SES neighborhoods, patients in the lowest SES quintile had 19% lower odds of receiving antidepressants (aOR: 0.81; 95% CI 0.74–0.88). Increasing age was strongly associated with reduced use (≥ 80 vs. 65–69 years: aOR:0.60; 95% CI 0.57–0.63), while female sex was associated with higher odds (aOR:1.19; 95% CI 1.15–1.23). Marked racial/ethnic disparities were observed: non-Hispanic Black (aOR:0.63; 95% CI 0.59–0.67) and Asian (aOR:0.60; 95% CI 0.56–0.65) patients were substantially less likely to receive antidepressants compared with non-Hispanic White patients. Higher comorbidity burden modestly increased use (CCI per point: aOR:1.07; 95% CI 1.06–1.08). Strong associations were observed for prior supportive care medication use (aOR:2.02; 95% CI 1.94–2.09) and hospice enrollment (aOR:1.73; 95% CI 1.67–1.80). Tumor location and stage demonstrated statistically significant but clinically smaller effect sizes.

Conclusion: A quarter of older adults diagnosed with PC received antidepressants, with substantial variation by socioeconomic status, race/ethnicity, age. The markedly lower use among racial/ethnic minority and lower-SES patients indicates potential inequities in mental health treatment access. Increased use among those with higher comorbidity burden and hospice enrollment suggests that antidepressants are more often introduced in the context of advanced symptom burden or end-of-life care rather than early in the cancer course. These findings underscore the need for systematic depression screening across all demographic groups and proactive integration of mental health services into routine pancreatic cancer care.

POSTER #110

IDENTIFYING METABOLIC SYNERGIES WITH REF-1 TARGETING FOR COMBINATION TREATMENT OF PANCREATIC CANCER

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Pancreatic ductal adenocarcinoma (PDAC) is the third leading cancer-related mortality in human malignancies with a five-year survival rate of 13%. Poor clinical outcomes in PDAC are largely attributed to the dense, reactive stroma, robust antioxidant defenses, and metabolic plasticity associated with the disease. The tumor cells' ability to fuel growth with many different metabolic pathways along with this reactive tumor microenvironment (TME) is believed to contribute to the difficulty in treating this lethal disease. A key mediator of this adaptive capacity is Redox factor-1 (Ref-1), a multifunctional protein involved in DNA repair and activation of cancer-relevant transcription factors (TFs) including HIF-1 α , NF- κ B, and STAT3. The reduction of key cysteines in these TFs promotes their DNA binding, leading to activation of gene programs important for tumor cell survival, response to hypoxic conditions, and mitochondrial function, contributing to the progression of PDAC. Our group has developed APX2014, a potent and selective small-molecule inhibitor targeting the redox function of Ref-1. However, given the complexity of PDAC, single agent therapies are unlikely to achieve a complete response, therefore additional exploitable vulnerabilities must be identified. To do this, we performed a metabolism-centered CRISPR/Cas9 synthetic-lethal screen in MIA-PaCa2 cells treated with APX2014. This screen identified seven top metabolic and antioxidant genes (*NADK*, *G6PD*, *SEPHS2*, *TXNRD1*, *PRDX1*, *ALAS1*, and *ALAD*) required to sustain redox balance when Ref-1 activity is inhibited. These genes are integral to NADPH generation, selenoprotein and heme biosynthesis, and thioredoxin/peroxiredoxin redox cycling, supporting the Ref-1 redox cycle function and homeostasis. Currently, we are validating the candidate gene dependencies using a targeted siRNA-mediated knockdown approach. Subsequently, knockdown cells are treated with APX2014, and survival is assessed to determine if gene silencing enhances the sensitivity to Ref-1 inhibition. We hypothesized co-targeting Ref-1 with APX2014 plus siRNA knockdown of these identified genes will limit the metabolic flexibility in PDAC by restricting compensatory pathways resulting in tumor suppression, thereby overcoming therapeutic

resistance. Preliminary data demonstrates knocking down *TXNRD1* and *PRDX1* synergizes with Ref-1 inhibition, which further validates the data from our CRISPR screen. At present, we are validating Glucose-6-phosphate dehydrogenase knockdown (*G6PD^{KD}*) as another potential co-target with Ref-1 inhibition. G6PD contributes to NADPH production in the cell environment through the oxidative pentose phosphate pathway (PPP), which participates in the redox cycle of Ref-1 to maintain redox homeostasis and reduce oxidative stress to enable cell survival in PDAC. Current data showed a synergistic effect with Ref-1 inhibition in experiments conducted over a 144-hour period. This work will confirm therapeutic targets to enhance the efficacy of APX2014, elucidate promising combination strategies for modulating metastasis and potentially lead to mechanism-based approaches to improve the clinical outcomes of patients with PDAC.

Translational/Clinical Research *ACS-Postbaccalaureate student*

POSTER #111

BRATS-PATHOLOGY 2024: INSIGHTS AND FUTURE DIRECTIONS INFORMED BY THE AI-RANO & RANO-RGP EFFORT TO ASSESS GLIOBLASTOMA HETEROGENEITY

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INTRODUCTION: Glioblastoma is the most common malignant adult brain tumor with poor prognosis, largely due to its heterogeneous landscape. The AI-RANO and RANO-RGP groups organized the BraTS-Pathology 2024 challenge, providing a publicly-available dataset and a benchmarking environment for AI models to automatically identify six clinically-relevant histopathologic sub-regions in H&E-stained whole slide images (WSIs). **METHODS:** We reclassified the TCGA-GBM and TCGA-LGG data according to the WHO 2021 criteria and identified diagnostic WSIs of 188 glioblastoma (IDH-wt, CNS WHO Gr.4) patients. A well-defined annotation protocol was communicated to eight neuropathologists, across eight institutions, who created “ground truth” annotations for Cellular Tumor (CT), Geographic Necrosis, Cortical Infiltration, Pseudopalisading Necrosis (PN), Microvascular Proliferation, and Penetration into White Matter (PWM). Comprehensive WSI curation for elimination of artifactual content and follow up partitioning resulted in approximately 300,000 data samples/ patches. **FINDINGS:** Inter-observer variability assessment highlighted difficulties in establishing a “gold standard”. The least annotated sub-regions (PWM and PN) were the most challenging to the participating methods, whereas all methods achieved their highest accuracy in the most annotated sub-region (CT). The highest average performance across sub-regions was obtained by a foundation model-based method. However, this top-ranked solution achieved an F1 score of 0.51 (Accuracy=0.65, Sensitivity=0.56, Specificity=0.93). **CONCLUSIONS:** Our findings suggest that achieving state-of-the-art performance, in automatically identifying clinically-relevant glioblastoma sub-regions, remains an unsolved research problem. Analysis of the ground-truth annotations underscores the critical need for more precisely defined and neuropathologically standardized annotation protocol. Current AI algorithms are facing difficulties in addressing glioblastoma’s heterogeneous histologic landscape in the absence of large, comprehensively, and consistently annotated data. Building upon these findings, and towards developing more accurate AI algorithms, the 2025 iteration of the BraTSPath challenge is currently on-going by leveraging two million data samples, aiming to enhance our disease understanding and improve diagnostic accuracy, ultimately contributing to improved patient outcomes.

Translational/Clinical Research

Data Engineer

POSTER #112

REF-1 INHIBITION DISRUPTS MITOCHONDRIAL METABOLISM AND REVEALS CO-TARGETABLE METABOLIC VULNERABILITIES IN PANCREATIC CANCER

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Ref-1 Inhibition Disrupts Mitochondrial Metabolism and Reveals Co-targetable Metabolic Vulnerabilities in Pancreatic Cancer

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Objective: Treating pancreatic ductal adenocarcinoma (PDAC) remains exceptionally difficult, with limited progress in therapy over the last three decades. Dubbed the “graveyard of chemotherapy,” PDAC continues to pose formidable clinical challenges. Even the most effective first-line chemotherapies extend survival in metastatic disease only slightly beyond 12 months. The poorly vascularized and fibrotic tumor microenvironment drives hypoxia and nutrient deprivation. To survive, PDAC cells adapt metabolically via autophagy and macropinocytosis. Oncogenes such as HIF1A, KRAS, and MYC further reshape tumor metabolism toward glycolysis, fueling metastasis and therapy resistance. Targeting such metabolic dependencies offers promise to overcome chemoresistance. Ref-1 (APE1) regulates multiple transcription factors, including HIF-1 α , and plays a key role in PDAC metabolic reprogramming. Our prior work shows Ref-1 redox inhibition disrupts the TCA cycle, OXPHOS, and tumor growth. *Therefore, we hypothesize that combining Ref-1 inhibition with additional tumor-specific metabolic targets will yield synthetic lethality in PDAC.*

Methods: To assess the impact of Ref-1 inhibition on tumor metabolism, mitochondrial function, and growth patient-derived PDAC cells, cancer-associated fibroblasts, and PDX models were used. Mechanistic insights were gained using global metabolomics, RNA sequencing, and functional assays including mitochondrial substrate utilization, ATP measurement, and 3D tumor-CAF co-culture spheroids treated with Ref-1 inhibitors alone or in combination with metabolic disruptors.

Results: Treatment with Ref-1 inhibitors impaired TCA cycle activity, suppressed ATP production, and dramatically reduced mitochondrial volume and mitochondrial DNA content, indicating disruption of mitochondrial biogenesis. These effects were amplified under metabolic stress conditions (galactose media), where cells are forced to rely on OXPHOS. Global metabolomics and RNA sequencing revealed downregulation of glycolysis, glutaminolysis, and nucleotide biosynthesis, alongside activation of redox balancing pathways such as glutathione metabolism. Functionally, combining Ref-1 inhibition with metabolic disruptors (e.g., dmaKG, PFK158, STF-31) led to significant cytotoxicity in 3D PDAC co-culture spheroids composed of tumor cells and cancer-associated fibroblasts (CAFs), with combinatorial treatments outperforming single agents. In vivo validation using PDX models confirmed that Ref-1 inhibition alone significantly suppresses tumor growth.

Conclusion: Ref-1 is a key regulator of PDAC metabolic fitness, particularly mitochondrial integrity. Inhibiting Ref-1 reveals actionable metabolic weaknesses that can be co-targeted to induce synthetic lethality. These data support a novel strategy to exploit metabolic liabilities in PDAC using Ref-1-directed therapy in rational combinations.

Translational/Clinical Research Faculty

POSTER #113

HSP90 β INHIBITION ENHANCES THE EFFICACY OF NAB-PACLITAXEL/GEMCITABINE THERAPY IN PRECLINICAL PANCREATIC CANCER MODELS

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Objective:

Pancreatic ductal adenocarcinoma (PDAC) is a highly aggressive malignancy with an extremely poor prognosis. Standard treatment with nab-paclitaxel and gemcitabine (NPT+GEM) yields a median survival of only 8.5 months. Heat shock protein 90 (Hsp90) client proteins—including EGFR, IGF-1R, Raf, PI3K, AKT, CXCR4, and MMP2/9—are implicated in tumor proliferation, survival, angiogenesis, metastasis, and chemoresistance in multiple tumor types, including PDAC. Traditional Hsp90 inhibitors that bind to the N-terminal ATP-binding site exhibit pan-inhibitory activity against all four Hsp90 isoforms, leading to induction of the heat shock response, which can lead to chemoresistance and dose-limiting toxicities. Here, we evaluated the antitumor efficacy of novel Hsp90 β -selective inhibitors in preclinical PDAC models.

Methods:

In vitro cell proliferation assays were performed using the colorimetric WST-1 method. Protein expression levels were analyzed by immunoblotting. Tumor growth studies were conducted in NOD/SCID mice bearing subcutaneous xenografts derived from AsPC-1 and Panc-1 cells. Survival studies were performed using PDAC peritoneal dissemination xenograft models generated with AsPC-1 cells.

Results:

Overexpression of Hsp90 β and its client proteins—including EGFR, IGF-1R β , CXCR4, and AKT—was observed across PDAC-associated epithelial, endothelial, and stromal cells, whereas normal human pancreatic tissue showed negligible expression. The Hsp90 β -selective inhibitors NDNB-25 and NDNB-21, synthesized using a structure-based approach, demonstrated dose-dependent antiproliferative activity and synergistic effects when combined with standard chemotherapy in Hsp90 β -expressing PDAC epithelial and stromal cell lines. NDNB-25 reduced the expression of key Hsp90 client proteins (EGFR, IGF-1R, HER2, p-MEK, p-ERK, p-S6, and c-Myc) without inducing Hsp90 expression. Treatment also induced the expression of apoptosis markers (cleaved caspase-3 and cleaved PARP-1) and the epithelial differentiation marker E-cadherin. In subcutaneous xenograft models using Hsp90 β -overexpressing PDAC cell lines (AsPC-1 and Panc-1), NDNB-25 and NDNB-21 significantly inhibited tumor growth and showed additive effects when combined with chemotherapy. In AsPC-1 xenografts, tumor growth inhibition ranged from 47–61% with NPT+GEM, 58–72% with NDNB-25 or NDNB-21 monotherapy, and 79–85% with the combination treatment. In Panc-1 subcutaneous xenografts, both NDNB-25 and NDNB-21 produced similarly strong antitumor activity, with marked tumor growth inhibition. In Capan-2 PDAC xenografts with low Hsp90 β expression, the antitumor effects of NDNB-25 and NDNB-21 were less pronounced. In AsPC-1 peritoneal dissemination models, Hsp90 β inhibitors provided limited survival benefit.

Conclusion:

These preclinical findings support the continued development of Hsp90 β -selective inhibitors as next-generation therapeutic agents capable of improving treatment outcomes in PDAC, particularly in tumors with high Hsp90 β expression.

POSTER #114

TARGETING DNA DAMAGE SENSORS (DDS) TO ENHANCE OSIMERTINIB RESPONSE IN EGFR-DRIVEN CANCERS.

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Lung cancer remains the leading cause of cancer-related mortality worldwide. Approximately 85% of cases are classified as non-small cell lung cancer (NSCLC) and activating mutations in the epidermal growth factor receptor (EGFR) are present in about 16% of metastatic NSCLC tumors. The development of EGFR tyrosine kinase inhibitors (EGFR-TKIs), including the third-generation agent Osimertinib, has significantly improved patient outcomes. Nonetheless, its efficacy is limited by both on-target and off-target resistance mechanisms, including C797S, MET amplification, and histological transformation. Emerging evidence indicates that EGFR signaling influences DNA repair and that persistent proliferative signaling in EGFR-driven tumors elevates replication stress (RS), making them uniquely vulnerable to inhibition of the DNA damage response (DDR). While prior approaches have primarily targeted DDR kinases such as ATR, CHK1, and WEE1, the role of DNA damage sensors (DDS), including replication protein A (RPA) and the Ku70/80 complex, remains largely unexplored. In this study, we focus on this strategy by targeting RPA and Ku70/80, which recognize DNA structures generated by direct DNA damage or RS and initiate the DDR. Using small-molecule inhibitors that target RPA and Ku-DNA binding, in combination with clinically relevant EGFR-TKIs, we aimed to elucidate the mechanistic interactions between DDR pathways and EGFR signaling and how DDR modulation affects therapeutic response and resistance in EGFR-driven NSCLCs. Our results show that the DDS inhibitors NERx-329 (RPAi) and Ku-DBi 3392 (Ku70/80) in combination with Osimertinib further enhance antiproliferative and cytotoxic effects in TKI-sensitive, EGFR-mutant NSCLC cells. Our data demonstrates that DDR inhibition is effective in TKI-sensitive EGFR-driven models, thereby potentiating Osimertinib activity. Importantly, this is independent of a DNA-damaging agent, suggesting that inhibition of DDR alone is sufficient to increase the therapeutic effect of Osimertinib. This effect appears to be EGFR-specific, as no potentiation of Alectinib activity was observed in an EML4-ALK-driven NSCLC model. These preliminary findings support functional crosstalk between DDS-mediated DDR pathways and EGFR signaling, highlighting a potential therapeutic strategy to overcome, delay, or prevent the recurrence of resistance in EGFR-driven NSCLC.

Translational/Clinical Research Faculty

POSTER #115

THE IMPACT OF OXYGEN EXPOSURE ON CLINICAL BIOMARKERS – AN UNDERRECOGNIZED SOURCE OF PRE-ANALYTIC VARIABILITY

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When preclinical research fails to replicate human biology, scientific progress stalls, clinical trials falter, and patients continue to suffer. While many factors contribute to these failures, lack of attention to pre-analytic variability is a seminal issue. We recently reported that even short-term exposure to ambient air is sufficient to trigger signaling changes in tumor and non-malignant biospecimens. Those changes in turn alter their biology and responsiveness to targeted therapies. Thus, characterization of tumors collected and processed under physioxia (3% O₂) instead of current practice of collection and processing under ambient air (21% O₂) will help to identify clinically relevant biomarkers that are affected by O₂ tensions. This approach may help to reduce clinical trial failure rates and increase clinical translation of preclinical studies. Towards this goal, we collected human specimens (biopsies, ascites and pleural effusions from 94 donors) under physioxia, then divided the same specimen into two groups; one group maintained under physioxia, the other group exposed to ambient air. Both were for 45-60 minutes before fixing/processing. Samples were subjected to various biomarker analysis using IHC/IF, Western blotting to measure proteins, and nanopore sequencing for DNA methylation. We found the levels of pAKT, a clinically used biomarker of targeted therapy, were constantly higher in clinical samples under physioxia compared to ambient air. Similar effects of O₂ tension on pERK and MDM4 levels occurred in cells isolated from ascites or pleural effusion in a time-dependent manner. A significant decrease in pEGFR and p53 levels were observed in cells under physioxia. Moreover, O₂ tension-dependent differences extended to key epigenetic regulators including TET2. TET2 levels were lower under physioxia compared to ambient air. Consistently, nanopore sequencing revealed distinct differences in DNA methylation patterns under physioxia and ambient air. The observed differences in signaling pathways extended to cultured cells from ascites fluids and pleural effusions. However, there is a specificity in the effects of O₂ tensions on biomarkers as we did not observe significant differences in pPDGFR_β, ATE1 and many other biomarkers under two O₂ conditions. These results imply that the O₂ tension affects specific biomarkers and epigenome. Collectively, O₂ tension could result in dynamic and extensive changes in cell membrane, cytoplasmic and nuclear biomarkers. Observable changes of biomarkers could occur within an hour following exposure to ambient O₂, and some changes could last for prolonged period. Thus, our current study lays out a new physiologically relevant biomarker validation/discovery platform, which may accelerate evaluation of physiologically relevant signaling networks, new drug discovery, and enhance clinical translation of preclinical observations.

Translational/Clinical Research Faculty

POSTER #116

PRELIMINARY BRAIN NEUROINFLAMMATION EVIDENCE IN BREAST CANCER SURVIVORS EXPERIENCING AROMATASE INHIBITOR-ASSOCIATED MUSCULOSKELETAL SYNDROME

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Aromatase inhibitor-associated musculoskeletal syndrome (AIMSS), characterized by arthralgia, myalgia, and muscle stiffness, is a major side effect of breast cancer therapy, yet the contribution of central neuroinflammatory mechanisms remains unclear. This pilot study aimed to characterize translocator protein (TSPO) signal as a marker of neuroinflammation in women with AIMSS. Postmenopausal women with AIMSS (n=10), no-AIMSS breast cancer controls (n=10), and age- and sex-matched healthy controls (n=9) completed simultaneous TSPO positron emission tomography and magnetic resonance imaging along with selected pain assessments and quantitative sensory testing. Whole-brain voxel-wise analyses showed no group differences between AIMSS and no-AIMSS or between the combined breast cancer cohort and healthy controls. In hypothesis-driven analyses of primary and secondary somatosensory cortices, thalamus, insula, and medial frontal cortex (MFC), breast cancer participants demonstrated a trend toward elevated right secondary somatosensory cortex signal compared with healthy controls ($p=0.057$). Within the AIMSS group, exploratory partial correlations controlling for TSPO genotype revealed that MFC signal was positively associated with pressure pain thresholds at the trapezius ($r=0.638$, $p=0.060$) and wrist ($r=0.651$, $p=0.057$), wrist pain tolerance ($r=0.743$, $p=0.022$), and conditioned pain modulation threshold and tolerance ($r=0.758$, $p=0.018$), and negatively associated with temporal summation at the wrist ($r=-0.696$, $p=0.037$). These AIMSS-specific associations suggest that regional neuroinflammation in MFC may relate to reduced pain sensitivity, altered temporal summation, and impaired descending pain modulation. These preliminary findings highlight the potential role of cortical neuroinflammation in central pain processing in AIMSS and underscore the need for larger longitudinal studies to clarify mechanistic pathways.

Translational/Clinical Research Faculty

POSTER #117

INTEGRATING SINGLE-CELL AND BULK TRANSCRIPTOMICS TO IDENTIFY PROGNOSTIC GENE SIGNATURES AND THERAPEUTIC TARGETS IN LUNG ADENOCARCINOMA

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Background/Objectives: Despite advances in treatment, the prognosis for lung adenocarcinoma (LUAD) remains poor, and tools for predicting patients at high risk of adverse outcomes are lacking. Prognosis among lung adenocarcinoma patients is linked to tumor heterogeneity. We integrate scRNA-seq with bulk RNA-seq data to identify cell-type-specific prognostic gene signatures, develop predictive models, and explore therapeutic implications. **Methods:** We harmonized five scRNA-seq datasets from 30 lung adenocarcinoma patients to create a comprehensive tumor microenvironment atlas and cell-type-specific gene signatures. We applied these signatures to The Cancer Genome Atlas for lung adenocarcinoma bulk samples and assessed their prognostic significance using multivariate Cox models. We compared the predictive performance of models integrating gene signatures with clinical variables to those using clinical variables alone and validated it in an external cohort. Gene Ontology over-representation analysis and Open Targets queries identified functional pathways and therapeutic opportunities. **Results:** We identified six gene signature models that independently predicted overall survival. High signature scores for CD8+ T cells, B cells, dendritic cells, and AT2 cells indicated better survival, while high scores for macrophages and mast cells indicated poorer prognosis. Our integrated model, which combined gene signature and clinical variables, significantly outperformed the clinical-only model for predicting 3-year survival in validation dataset (AUC = 0.801 vs. 0.665). Functional analysis showed protective signatures enriched for MHC class II assembly, phagocytosis, and chemotaxis. Whereas the adverse mast cell signature was enriched for actin filament-based movement. No significant pathway identified for the macrophage signature. Furthermore, drug-target analyses revealed actionable pathways associated with adverse signatures. **Conclusions:** Cell-type-specific gene signatures provide independent and clinically relevant prognostic insight in lung adenocarcinoma supporting tumor microenvironment-informed biomarkers for precision oncology.

Translational/Clinical Research

Graduate Student

POSTER #118

ANCESTRY-ENRICHED ACKR1 GERMLINE VARIANT AND ITS FUNCTIONAL IMPACT ON NORMAL AND BREAST CANCER BIOLOGY

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Atypical chemokine receptor 1 (*ACKR1*) gene, harbors single nucleotide polymorphisms in its regulatory and coding regions. The regulatory region variant rs2814778 is enriched in African and Arab populations and it confers protection against malarial infection. However, it is responsible for the Duffy-Null (CC) /heterozygous (C/T) phenotype, characterized by a significant reduction in *ACKR1* expression in epithelial and non-epithelial cells. *ACKR1* functions as a decoy receptor for several chemokines including the breast cancer metastasis-associated CXCL12, to regulate immune/ inflammatory pathways. There is a growing interest to include Duffy genotyping in clinical trial design to account for normal biological differences underlying aggressive breast cancer outcomes in black women. We seek to investigate the potential influence of *ACKR1* variant on breast cancer outcomes, with a long-term goal of identifying therapeutic vulnerabilities. We established a model system by generating immortalized breast epithelial cell lines with functional *TT* (wild type that express *ACKR1* -African and European ancestry), heterozygous (*C/T*) and homozygous (*CC*), using breast tissues from the institutional resource of Komen Normal Tissue bank. Cell lines with *TT* expressed higher *ACKR1* mRNA levels relative to *C/T* or *CC* in the regulatory region. We hypothesized that reduced *ACKR1* expression alters chemokine signaling which may influence intrinsic and extrinsic signaling to activate downstream oncogenic pathways. To test this, we investigated WNT/GSK-3 β signaling and observed that *ACKR1* variant influences differential phosphorylation of β -catenin, facilitating nuclear translocation. We sought to explore further the effect of cytokines including inflammatory IL-6 secreted as a result of *ACKR1* variant on downstream oncogenic pathways. We observed increased phosphorylation of STAT3, a surrogate of IL-6 activity in breast epithelial cells of *ACKR1* *CC* and *C/T* cells compared to *TT*. Our analysis of the UALCAN database further revealed that reduced *ACKR1* expression in breast cancer correlates with progression to brain metastases. Consistent with this, our results demonstrate enrichment of CD271-high populations; indicative of cancer stem-like properties in *ACKR1* *CC* and *C/T* cells. These findings suggest that reduced *ACKR1* expression may promote acquisition of stem-like and metastatic properties. In vivo studies are underway to evaluate the effect of *ACKR1* status on distinct tumor characteristics including cancer stem cell properties, increased metastatic potential and response to targeted and conventional chemotherapy. A positive outcome from this study will have a transformative impact, establishing *ACKR1* germline variants as determinants of breast cancer biology and highlight the need to integrate ancestry-informed genotypes into clinical trial design and therapeutic decision making.

Translational/Clinical Research

Graduate Student

POSTER #119

INVESTIGATING THE THERAPEUTIC POTENTIAL OF A NOVEL PRMT5 INHIBITOR FORMULATED WITH NANOCRYSTAL TECHNOLOGY IN PANCREATIC DUCTAL ADENOCARCINOMA

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Persistent inflammation is a hallmark of pancreatic ductal adenocarcinoma (PDAC), largely driven by sustained NF- κ B activation. Protein arginine methyltransferase 5 (PRMT5), a promoter of tumorigenesis in several cancers, including PDAC, has emerged as a therapeutic target. Clinical data show that high PRMT5 expression correlates with shorter patient survival. Our lab developed small-molecule PRMT5 inhibitors, including the patented compound PR5-LL-CM01 (CM01), which showed superior anti-tumor efficacy and lower toxicity than the commercial inhibitor EPZ015666. However, CM01's poor water solubility limits its clinical potential. To address this, we formulated it as an albumin-coated nanocrystal (NC) (Abxtal) to improve dispersibility and bioavailability. CM01 NCs (~87 nm) remain stable for at least three months at -20°C. We hypothesize that CM01 NC more effectively inhibits PRMT5-mediated NF- κ B signaling and synergizes with gemcitabine (Gem) to suppress PDAC. Supporting this, CM01 NC inhibited PDAC cell growth, spheroid formation, and migration more effectively than CM01 and reduced NF- κ B transcriptional activity and target genes (TNF- α , IL-8). Chou-Talalay analysis confirmed synergy between CM01 NC and Gem. Ongoing in vivo studies will assess pharmacokinetics and therapeutic efficacy. This work may establish CM01 NC, alone or with Gem, as a promising treatment strategy for PDAC.

Translational/Clinical Research

Graduate Student

POSTER #120

PRECLINICAL MODELING OF DIFFERENTIAL TARGETING EFFECTS OF CDK INHIBITORS IN NF1-ASSOCIATED MPNST

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Purpose: The leading cause of premature death for patients with Neurofibromatosis type 1 (NF1) is the development of malignant peripheral nerve sheath tumors (MPNST). These tumors render current medical treatment strategies largely ineffective, prompting an investigation of rational targeted therapies. Cyclin-dependent kinase (CDK) inhibitors are favorable candidates to strategically oppose the chronic cell cycle deregulation driving MPNST growth. However, characterization of differential kinase targeting and associated tumor responses is needed to determine optimal combination therapies.

Methods: Multiplexed kinase inhibitor bead (MIB) affinity chromatography coupled with mass spectrometry (MIB/MS) was used to identify target spectra of CDK4/6 inhibitors (abemaciclib, palbociclib) and CDK2 inhibitors (tagtociclib, INX-315). Molecular and phenotypic responses to CDK inhibitors were evaluated in multiple MPNST cell line models. CRISPR/Cas9 knockout of *RBI* in an MPNST cell line was also used to evaluate inhibitor dependence on RB *in vitro* and in orthotopic implant mouse models.

Results: MPNST cell lines demonstrated sensitivity to single agent treatment by both CDK4/6 and CDK2 inhibition. In MIB/MS competition assays, previously reported off-target kinases exclusive to abemaciclib were identified, while palbociclib remained selective to CDK4/6. Similarly, tagtociclib demonstrated differential binding beyond CDK2 compared to INX-315. *RBI* knockout MPNST cells exhibited resistance to palbociclib treatment, but remained sensitive to abemaciclib and INX-315, in long-term exposure studies. RB-deficient cells were less responsive to CDK4/6 inhibitor monotherapy *in vivo*.

Conclusion: CDK inhibitors continue to show promise as targeted therapies against NF1-associated MPNST with dysregulated cell cycle signaling. Binding of abemaciclib to CDK2 reinforces cell growth suppression, and initial tests of pharmacologic CDK2 inhibitors in MPNST cell lines demonstrate efficacy. Informed by MIB/MS binding profiles, optimized multi-CDK inhibitor strategies could offer superior durability. Characterization of these combinations and their *in vivo* efficacy is therefore warranted.

Translational/Clinical Research

Graduate Student

POSTER #121

BCR PROGNOSTIC SIGNATURES FROM BULK RNA DATA THAT DIFFERENTIALLY EXPRESS ACROSS DIFFERENT SITES WITH RECURRENCE AND NO RECURRENCE

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BCR prognostic signatures from bulk RNA data that differentially express across different sites with recurrence and no-recurrence.

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Abstract

Background

Biochemical recurrence after radical prostatectomy remains clinically heterogeneous, with recurrence occurring in bone, lymph nodes, or locally in the prostatic fossa. Current risk stratification relies primarily on Gleason grade and pathologic stage, which do not fully capture the molecular programs within the primary tumor that may influence recurrence. Mesenchymal signaling and extracellular matrix remodeling have been associated with aggressive prostate cancer, yet the relationship between recurrence associated transcriptional changes and quantitative pathomic features remains incompletely defined.

Objective

To identify differentially expressed genes associated with biochemical recurrence and to determine whether these transcriptional changes are reflected in quantitative stromal pathomic features.

Hypothesis

We hypothesize that primary tumors from patients who develop biochemical recurrence exhibit distinct transcriptional programs consistent with mesenchymal and microenvironmental remodeling, and that these gene expression changes correlate with measurable pathomic features within the tumor tissue.

Methodology

Bulk RNA expression data from 48 primary prostate tumors, including 8 cases with biochemical recurrence and 40 without recurrence, were analyzed using linear modeling with empirical Bayes moderation in the limma framework. Genes were ranked by moderated t statistics with correction. Although no transcripts met predefined false discovery rate thresholds, nominally significant genes were evaluated at the pathway level using MSigDB Hallmark gene sets. A predefined mesenchymal gene set was used to compute a composite EMT signature score per sample. Pathomic features were extracted from segmented histologic regions and aligned for exploratory gene morphology correlation analysis.

Results

No genes met genome wide false discovery rate thresholds. Pathway level analysis indicated enrichment of mesenchymal and extracellular matrix-related signaling in recurrence-associated tumors. EMT and proliferation scores were significantly elevated in recurrence cases ($p = 0.0011$ and $p = 0.021$). Moderate correlations were observed between selected mesenchymal transcripts and stromal pathomic features ($\rho = 0.4$); however, these associations did not remain significant after multiple testing correction. Correlation strengths were comparable across stromal and gland pathomic compartments ($p = 0.437$), indicating no clear compartment specific enrichment in this cohort. Although the findings are exploratory, we would be validating them on 300 patient's cohort.

Translational/Clinical Research

Graduate Student

POSTER #122

A MICROENVIRONMENT-AWARE DEEP LEARNING FRAMEWORK LEVERAGING GLAND-STROMA MORPHOLOGICAL SIGNATURES FOR PROSTATE CANCER PROGNOSTICATION

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Background: Biochemical recurrence (BCR) remains a significant clinical challenge, affecting up to 40% of patients following radical prostatectomy. Traditional post-surgical risk stratification relies heavily on glandular epithelial patterns, such as the Gleason grading system. However, this focus often overlooks the prognostic potential of the tumor microenvironment. The stroma, which comprises the supporting connective tissue surrounding cancer glands, undergoes structural remodeling that reflects disease aggressiveness. While the prognostic role of stromal morphology has historically been understudied, capturing these complex epithelial-stromal interactions through high-dimensional feature embeddings offers a path toward more precise, individualized survival modeling.

Objective: We present a multi-stage computational pathology framework designed to explicitly model glandular and stromal-specific morphological signatures within cancerous regions of whole-mount histology images. Our goal is to evaluate whether a compartment-aware approach, separating gland and stroma before feature extraction, improves the prediction of time-to-BCR. Furthermore, we aim to validate the generalizability of these prognostic signatures across independent patient cohorts.

Methods: The framework was developed and evaluated using a primary dataset of 429 patients and a separate validation cohort of 45 patients. The pipeline consists of three distinct stages. First, a cancer segmentation model was applied to whole-mount histopathology slides to delineate tumor regions, which were subsequently refined via morphological post-processing. Second, a tissue compartment segmentation model used a U-Net architecture to subdivide these cancerous regions into glandular and stromal compartments. Third, feature embeddings were extracted from these compartments using a pathology-specific foundation model. These patient-level features were used to train Cox proportional hazards survival models to predict the risk of BCR over time.

Results: The whole-mount cancer segmentation achieved a Dice coefficient of 0.81, ensuring accurate tumor localization. In the tissue compartment stage, the U-Net achieved Dice scores of 0.87 for benign tissue and 0.74 for cancerous regions. The lower performance in cancer regions highlights the diagnostic challenge of morphologically complex tissue. Survival analysis on the separate 45-patient cohort revealed that glandular and stromal embeddings were independently prognostic, yielding C-indices of 0.830 and 0.796, respectively. The successful stratification of risk using signatures from both compartments demonstrates that the support tissue provides a significant prognostic signal that aligns with glandular findings.

Conclusion and Discussion: This study demonstrates that incorporating stromal microenvironmental features significantly enhances the accuracy of BCR prognostic modeling in prostate cancer. By leveraging foundation model embeddings within a compartment-aware pipeline, we successfully captured nuanced morphological signals that complement traditional glandular analysis. The independent prognostic significance of the stroma confirms that the tumor microenvironment holds vital information for predicting disease progression. This

work establishes a validated workflow for automated risk assessment. Future efforts are focused on the continued validation of these signatures using independent cohorts from multiple institutions.

Translational/Clinical Research

Graduate Student

POSTER #123

A DUAL EPIGENETIC INHIBITOR-STING AGONIST COMBINATION STRATEGY TO ENHANCE TUMOR IMMUNITY IN HIGH GRADE SEROUS OVARIAN CANCER

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Epithelial ovarian cancer (OC) remains one of the most lethal gynecological malignancies primarily due to its late-stage diagnosis and high recurrence rates. High-grade serous ovarian cancer (HGSC) is the most prevalent and aggressive subtype and is initially treated with cytoreductive surgery combined with platinum-based chemotherapy. However, most patients develop platinum-resistant disease with poor long-term survival. Additionally, immune checkpoint inhibitors (ICIs) have demonstrated limited clinical efficacy in HGSC. Recent studies in HGSC have focused on the role of immune pathways such as stimulator of interferon genes (STING) signaling, which may offer new therapeutic strategies. By controlling the transcription of host defense genes including pro-inflammatory cytokines, chemokines, and type I interferons, the STING pathway plays an essential role in initiating an immune response and may serve as a potential therapeutic target. We hypothesize that combination therapy with the STING agonist diABZI and the DNA methyltransferase inhibitor decitabine (DAC) will remodel the immunosuppressive tumor microenvironment in HGSC and enhance tumor-intrinsic and tumor-extrinsic immune responses. Time course analysis of HGSC cell lines treated with diABZI alone upregulated ($p < 0.05$; within 3-9 hrs) mRNA levels of chemokines (CXCL10, CCL5). Multiplex ELISA of conditioned media confirmed increased ($p < 0.05$) secretion of both chemokines at 6 hours, indicating functional activation of STING-dependent chemokine signaling. STING pathway activation was further validated using western blot analysis for phosphorylation of TBK1 and IRF3, downstream targets of STING, which showed significant upregulation with combination treatment. Treatment with diABZI with DAC reduced HGSC cell migration ($p < 0.05$), compared to single agents, indicating enhanced anti-tumor activity. In parallel, analyses of patient tumors revealed that tumors with high STING expression were associated with increased ($p < 0.05$) infiltration of natural killer (NK) cells, key effectors of innate immunity. To assess whether NK cells are cytotoxic against HGSC cells, co-culture assays were performed with NK-92-MI cells. The results demonstrated enhanced cytotoxicity against HGSC cells in an effector-to-target (E:T) ratio-dependent manner ($p < 0.05$), suggesting that STING activation increased interferon signaling and promoted chemokine-mediated NK cell recruitment via CXCL10 and CCL5 signaling. Taken together, these observations support a role for STING signaling as a key regulator of cytotoxic lymphocyte recruitment and underscore the therapeutic potential for STING agonists to enhance anti-tumor immunity in HGSC. We are using the combinatorial strategy to enhance STING signaling, increase NK cell recruitment, activation and infiltration, and remodel the immunosuppressive TME in HGSC. This work will provide mechanistic insight into overcoming the immunosuppressive tumor microenvironment in HGSC and ultimately inform the development of new immunotherapeutic strategies for this deadly disease.

Translational/Clinical Research

Graduate Student

POSTER #124

DISTINCT TRANSCRIPTIONAL AND IMMUNOMODULATORY EFFECTS OF THE RXR AGONIST V-125 IN MAMMARY TUMORS OF MMTV-NEU MICE COMPARED WITH BEXAROTENE.

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Background:

The retinoid X receptor (RXR) is a ligand-activated nuclear receptor that forms heterodimers with multiple nuclear receptor partners to regulate transcriptional programs governing cell differentiation, metabolism, immune modulation, and apoptosis. RXR agonists have demonstrated anti-tumor activity in preclinical and clinical settings; however, the clinical utility of first-generation agents such as the FDA-approved drug bexarotene has been limited by adverse metabolic effects, including hyperlipidemia. V-125 is a next-generation, selective RXR agonist designed to improve pharmacokinetic properties and reduce lipogenic side effects while maintaining anti-tumor efficacy. This study aimed to compare the molecular, transcriptional, and functional effects of V-125 and bexarotene in HER2⁺ breast cancer models.

Methods:

Female MMTV-Neu transgenic mice bearing spontaneous mammary tumors were treated for 10 days with control diet, V-125 (100 mg/kg diet), or bexarotene (100 mg/kg diet). RNA sequencing was performed to identify differentially expressed genes and pathway alterations induced by each compound. Key gene targets were validated by quantitative PCR (qPCR) and immunohistochemistry (IHC). Immune modulation within the tumor microenvironment was assessed by IHC staining for CD8⁺ cytotoxic T cells and CD206⁺ tumor-promoting macrophages. In parallel, in vitro studies were conducted using the HER2⁺ human breast cancer cell line JIMT-1 to evaluate RXR target gene activation and clonogenic potential following treatment.

Results:

V-125 induced broader and more distinct transcriptional reprogramming compared to bexarotene. Notably, V-125 selectively upregulated genes such as *Nrg1*, *Nfasc*, *Lrrc26*, and *Chi311*, which are associated with improved patient survival in breast cancer cohorts. Pathway enrichment analysis revealed significant modulation of immune activation pathways, cancer-associated signaling networks, and lipid metabolism pathways. Both V-125 and bexarotene reduced clonogenic survival in JIMT-1 cells, confirming RXR-dependent suppression of tumor cell growth. However, V-125 demonstrated unique immunomodulatory effects in vivo, including increased CD8⁺ T cell infiltration and reduced CD206⁺ macrophage abundance within tumors—effects not observed with bexarotene treatment. These findings indicate that V-125 more effectively reshapes the tumor immune microenvironment toward an anti-tumor phenotype.

Conclusions:

Collectively, these data demonstrate that V-125, unlike bexarotene, reprograms tumor transcriptional networks and the immune landscape in HER2⁺ breast cancer models. By enhancing anti-tumor immunity while suppressing tumor-promoting macrophage populations, V-125 exhibits both direct anti-proliferative and immune-modulatory activity. These findings support the therapeutic potential of selective RXR activation as a promising strategy for HER2⁺ breast cancer treatment and highlight V-125 as a next-generation RXR agonist with improved translational promise.

Translational/Clinical Research

Graduate Student

POSTER #125

TCR-ENGINEERED GAMMA-DELTA T CELLS SHOW SPECIFICITY AND ENHANCED KILLING EFFICACY AGAINST TRIPLE-NEGATIVE BREAST CANCER

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Triple-negative breast cancer (TNBC) is an aggressive breast cancer subtype defined by the lack of estrogen receptor, progesterone receptor, and HER2 expression, which limits the use of targeted endocrine or HER2-directed therapies. TNBC is associated with early relapse, visceral metastasis, and a disproportionate burden in younger patients and individuals of African ancestry, underscoring the need for more effective and accessible therapies. Immunotherapies have improved outcomes for some patients, but durable responses remain inconsistent, motivating new cellular approaches that can overcome tumor heterogeneity and immune evasion. Gamma-delta ($\gamma\delta$) T cells are an attractive platform for TNBC immunotherapy because $\gamma\delta$ T cells can integrate features of conventional T cells and innate immune cells, while recognizing stress-induced and non-classical antigens without strict dependence on peptide-HLA presentation, supporting broad anti-tumor activity. Using flow cytometry and T cell receptor (TCR) profiling, our laboratory identified a ~threefold activation/expansion of $\gamma\delta$ T cells in response to TNBC-derived exosomes, yielding distinct exosome-expanded clonotypes. Importantly, these exosome-expanded $\gamma\delta$ TCR clonotypes demonstrated increased TNBC-directed cytotoxicity *in vitro* compared with non-expanded $\gamma\delta$ T cells.

Building on these observations, we developed CRISPR-based lipid nanoparticle (LNP) engineering to generate TNBC-specific $\gamma\delta$ T cells expressing a recurrent TNBC exosome-expanded TCR clonotype (V γ 3V δ 1). CRISPR-LNP editing enables rapid installation of V γ 3V δ 1 TCR genes either into induced pluripotent stem cells (iPSCs) to produce $\gamma\delta$ T-iPSCs that can be differentiated into V γ 3V δ 1 effector T cells, or directly into peripheral blood-derived $\gamma\delta$ T cells to accelerate generation of engineered products. *In vitro*, V γ 3V δ 1 T cells derived from engineered $\gamma\delta$ T-iPSCs exhibited enhanced induction of TNBC apoptosis relative to controls. Similarly, direct CRISPR-LNP engineering of primary $\gamma\delta$ T cells with the V γ 3V δ 1 TCR produced rapid and enhanced killing of TNBC cells, with minimal cytotoxicity against non-malignant primary mammary and airway cells and no detectable activity against A549 (non-small cell lung cancer) cells. Notably, engineered V γ 3V δ 1 cells did not show improved killing versus unmodified $\gamma\delta$ T cells against MCF-7 (luminal A) breast cancer cells, supporting tumor-context specificity rather than generalized hyper-cytotoxicity. Together, these data suggest that TNBC exosome-informed $\gamma\delta$ TCR discovery combined with CRISPR-LNP engineering provides a practical path to generate TNBC-specific $\gamma\delta$ T cell products with enhanced anti-tumor activity and a favorable preliminary specificity profile. If successful, this framework could be extended to identify and engineer $\gamma\delta$ TCRs targeting other solid tumors, enabling scalable development of cancer-specific $\gamma\delta$ T cell immunotherapies.

Translational/Clinical Research

Graduate Student

POSTER #126

MECHANISTIC INSIGHTS INTO DIS3-DRIVEN PATHOGENESIS IN MULTIPLE MYELOMA: INTEGRATING PATIENT GENOMICS WITH NOVEL CELLULAR MODELS

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Multiple myeloma (MM), the second most common hematological cancer, is associated with a poor prognosis that varies based on the genetic alterations present at the time of diagnosis. Missense mutations in DIS3 are found in 10% of MM patients and can be either recurrent heterozygous hotspot mutations or homozygous non-hotspot mutations, which are associated with a worse prognosis. However, the molecular impact of these mutation groups has yet to be investigated. This knowledge is essential to understanding the biological role DIS3 mutations play in the progression of MM. To determine the function of these mutation groups, we analyzed the Multiple Myeloma Research Foundation CoMMpass Study of patient samples (n=1,245) which underwent DNA and RNA sequencing. We complemented this analysis by utilizing CRISPR-Cas9 homology directed repair to introduce 7 mutations (3 hotspot; 4 non-hotspot) into endogenous DIS3 in the MM cell line KMS11, which were analyzed by multi-omics (n≥3). DIS3 mutations did not affect DIS3 mRNA levels, although non-hotspot mutations resulted in a 25-fold decrease (p<0.0001) in DIS3 protein expression. In contrast, hotspot mutations exhibited no significant protein expression change, as confirmed by mass spectrometry. Non-hotspot DIS3-mutated protein loss was also observed in a Western blot of MM patient bone marrow samples, therefore confirming the reproducibility of our cell line results in patient samples. Total RNA-sequencing and differential gene abundance analysis in our cell models revealed DIS3 non-hotspot mutations result in a significant accumulation of DIS3-sensitive transcripts, including promoter upstream transcripts (PROMPTs), enhancer (e)RNAs, and lncRNAs. More specifically, 10 lncRNAs that have been previously found to be associated with a worse prognosis in MM patients showed greater dysregulation in the non-hotspot mutations. Certain lncRNAs were also predicted to interact with RNA-associated proteins such as DEAD-box RNA helicases. These results were also observed in the CoMMpass Study dataset, further supporting the validity of our DIS3-mutated cell line models. Further, DIS3 mutations caused dysregulation of immunoglobulin super-enhancers, likely driving widespread genetic alterations and contributing to disease progression. Analysis of mass spectrometry data indicated that non-hotspot mutations greatly impact the proteome by resulting in the significant (FDR<0.05) abundance change in an average of 200 proteins, whereas hotspot mutations resulted in an average of 12 proteins. Functional enrichment analysis showed that non-hotspot mutations led to a greater downregulation of RNA processing pathways compared to hotspot mutations at both the RNA and protein levels. Comparing the transcriptome and proteome changes displayed notable discordance in the abundance of some targets, particularly in RNA-associated proteins involved in RNA processing and ribosome biogenesis. Our findings provide insight into the molecular impact of DIS3 mutations, highlighting key differences between mutation groups which aid in deepening our understanding of MM pathogenesis and may contribute to the development of novel therapeutic strategies.

Translational/Clinical Research

Graduate Student

POSTER #127

INVESTIGATING THE FUNCTION OF PRMT1-MEDIATED ARGININE METHYLATION OF EWSR1 IN MULTIPLE MYELOMA

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Multiple myeloma (MM) is the second most common blood cancer and remains incurable despite significant advances in proteasome inhibitors, immunomodulatory drugs, monoclonal antibodies, and cellular immunotherapies. Frequent MM relapses and therapeutic resistance highlight the need to identify novel molecular vulnerabilities that sustain MM cell survival and progression. Post translational modifications such as arginine methylation have emerged as important regulators of transcription, RNA processing, and protein stability in MM. Protein arginine methyltransferase 1 (PRMT1) is the predominant type I PRMT and is highly expressed in MM. Studies in our lab has shown that PRMT1 is associated with relapse and poor clinical outcomes of MM. Preliminary proteomic studies suggest that PRMT1 interacts with the RNA-binding protein EWSR1, a member of the FET family involved in transcriptional regulation and RNA metabolism. Both PRMT1 and EWSR1 are involved in the survival of multiple myeloma cells. Given that both PRMT1 and EWSR1 are functionally important in MM, we hypothesize that PRMT1-mediated arginine methylation regulates EWSR1 function and contributes to MM cell survival.

The goal of this study is to define the functional relationship between PRMT1 and EWSR1 in MM and to develop a targeted nanoparticle-based strategy to deliver PRMT1 directed therapeutics. Firstly, PRMT1 inhibition experiments were performed in multiple myeloma cell lines, and EWSR1 expression was evaluated using RT-qPCR and western blot to distinguish transcriptional versus post translational regulation. These experiments showed that PRMT1 inhibition reduced EWSR1 protein levels without significantly altering EWSR1 mRNA expression, suggesting that PRMT1 regulates EWSR1 primarily through post-translational mechanisms. Future studies will determine whether PRMT1 regulates EWSR1 stability through ubiquitin-mediated degradation using cycloheximide chase and proteasome inhibition assays and will further examine how PRMT1 affects EWSR1 nuclear localization using immunocytochemistry. EWSR1 RNA interactions will be assessed by RIP-seq and its chromatin association will be examined by CUT and RUN to determine how PRMT1 regulates EWSR1-dependent transcriptional programs.

To translate these mechanistic insights into a targeted therapeutic strategy, lipid nanoparticles conjugated with varying densities of the BCMA-targeting antibody belantamab were engineered and loaded with Cy3 labeled siRNA. Flow cytometry analysis of Cy3 fluorescence demonstrated enhanced nanoparticle uptake in MM cells treated with BCMA-targeted LNP compared with non-targeted controls, with low to moderate antibody densities providing the most efficient delivery while maintaining cell viability. Low density BCMA-LNPs reduce B2M expression in cells. In vivo biodistribution studies indicated increased accumulation of antibody-conjugated LNP in bone associated tumor and liver sites compared to non-conjugated LNP. Future studies will evaluate BCMA-targeted nanoparticles delivering siPRMT1 in MM xenograft models to assess tumor-specific delivery, therapeutic efficacy, and systemic toxicity. Findings from this work will identify PRMT1-EWSR1 axis as a potential regulatory mechanism in MM and support the development of BCMA-targeted nanoparticles for selective therapeutic delivery in MM.

Translational/Clinical Research

Graduate Student

POSTER #128

DUAL INHIBITION OF CDK4/6 AND MEK IN ANAPLASTIC PLEOMORPHIC XANTHOASTROCYTOMA PRECLINICAL MODELS: INTERROGATION OF THERAPEUTIC RESPONSE MECHANISMS

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Typical survival for pediatric high-grade gliomas remains less than 18 months despite recent improved understanding of the molecular drivers of these tumors. Hyperactivating MAPK and CDK4/6 pathway mutations are common and targetable alterations implicated in tumorigenesis and malignant transformation in pediatric glioma. We have established and characterized a novel patient-derived xenograft (PDX) model, RHT128, from a pediatric patient diagnosed with the high-grade glioma anaplastic pleomorphic xanthoastrocytoma (APXA). The molecular landscape of PDX RHT128 exhibits molecular fidelity to the patient's tumor. Clinical precision genomics analysis of the tumor revealed a novel BRAF fusion protein and CDKN2A/B deletion. Based on this molecular signature, the patient was treated with MEK inhibitor trametinib as a monotherapy and, following progression of disease, with CDK4/6 inhibitor ribociclib. However, the tumor continued to progress. In this study our objective is to simultaneously target the CDK4/6 and MAPK pathways in RHT128 as well as two additional APXA PDX models, D2363 (BRAF wild-type) and D645 (BRAFFV600E mutant) that show CDK4/6 hyperactivation. We aim to determine to what extent this combination therapy minimizes emergence of therapeutic resistance. Single-agent efficacy assessments in a subcutaneous RHT128 PDX showed significant dose-dependent reduction in tumor volume after treatment with the blood-brain barrier-permeable abemaciclib ($p < 0.05$) and MEK inhibitor mirdametinib ($p < 0.0001$). Analysis of the global kinome in CDK4/6 inhibitor-treated PDX tissues compared to vehicle treatment using multiplexed-inhibitor bead chromatography–mass spectrometry demonstrated effective inhibition of CDK4/6 and dose-dependent increases in MAPK pathway kinases, supporting a combined MEK and CDK4/6 inhibitor therapeutic strategy. Indeed, RHT128 PDX and D645 models showed improved survival after treatment with combined mirdametinib and abemaciclib compared to either monotherapy or vehicle. An improvement in survival was not observed in the BRAF-WT D2363 PDX treated with this combination, suggesting that BRAF status may be a marker of response to this treatment. Importantly, long-term treatment with combined mirdametinib and abemaciclib in the RHT128 PDX is tolerable and trends toward a survival benefit compared to vehicle treatment. Examination of the molecular response in these treated tumors is ongoing and will investigate potential mechanisms of therapeutic resistance. Future studies include testing this combination in intracranial PDX models of RHT-128 and the pediatric high-grade glioma model SJGBM2, which will provide important insight into the blood-brain barrier penetrability of the combination and how the brain microenvironment may impact efficacy. The pervasiveness of alterations to the MAPK and CDK4/6 pathways in pediatric gliomas make this approach promising for further study in a broader range of these deadly tumors.

Translational/Clinical Research

Graduate Student

POSTER #129

PARP1-MEDIATED REGULATION OF PROSTATE CANCER CELL DEATH IN RESPONSE TO PARP INHIBITORS AND MMS-INDUCED DNA DAMAGE

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Prostate cancer remains a leading cause of cancer-related mortality among men worldwide, largely due to the emergence of therapeutic resistance in advanced disease. Targeting the DNA damage response (DDR) pathway represents a promising strategy to selectively eliminate cancer cells. Poly(ADP-ribose) polymerase 1 (PARP1) plays a critical role in the detection and repair of single-strand DNA breaks. Pharmacologic inhibition of PARP1 can convert these lesions into cytotoxic double-strand breaks during DNA replication, particularly in the presence of additional genotoxic stress. While PARP inhibitors have demonstrated clinical benefit in homologous recombination-deficient prostate cancers, the determinants of response and the mechanistic basis of cytotoxicity remain incompletely defined.

In this study, we systematically assessed the effects of clinically relevant PARP inhibitors alone and in combination with the alkylating agent methyl methanesulfonate (MMS) across multiple prostate cancer cell lines, including PARP1-proficient 22Rv1 cells and genetically engineered PARP1-knockout models. Our results show that MMS significantly potentiates PARP inhibitor-mediated cytotoxicity in parental 22Rv1 cells, even at low inhibitor concentrations. In contrast, PARP1-deficient cell lines exhibit markedly reduced sensitivity to combination treatment.

These findings demonstrate that functional PARP1 is essential for optimal PARP inhibitor-induced cell death and identify PARP1 status as a key determinant of therapeutic response. Moreover, the selective vulnerability observed in 22Rv1 cells highlights the potential of PARP1-dependent biomarkers to guide patient stratification and inform rational combination strategies in prostate cancer treatment.

Translational/Clinical Research

Graduate Student

POSTER #130

TARGETING REDOX VULNERABILITIES IN MALIGNANT PERIPHERAL NERVE SHEATH TUMORS THROUGH REF-1 INHIBITION AND DISRUPTION OF REDOX PATHWAYS

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Malignant Peripheral Nerve Sheath Tumors (MPNSTs) are aggressive soft tissue sarcomas representing 5-10% of all soft tissue sarcomas and frequently arise in patients with neurofibromatosis type 1 (NF1), a genetic disorder affecting approximately 1 in 3,000 individuals. NF1 patients have an 8-13% lifetime risk of developing MPNST, which is the leading cause of death in this population. Clinical outcomes remain poor, with 5-year survival rates of 20-50% and median survival of 12-18 months for metastatic disease. Standard chemotherapy regimens such as doxorubicin and ifosfamide yield response rates of only 17-25%, and no FDA-approved targeted therapies currently exist, highlighting a major unmet clinical need.

MPNSTs experience elevated oxidative stress due to rapid proliferation and loss of NF1-mediated RAS pathway regulation, making them highly dependent on antioxidant defense systems. The apurinic/apyrimidinic endonuclease 1/redox factor 1 (APE1/Ref-1) regulates redox activation of transcription factors including NF- κ B, HIF-1 α , STAT3, and AP-1 that support tumor growth and survival. Our laboratory has developed selective Ref-1 redox inhibitors, including APX2009 and APX2014, which block redox signaling without disrupting base excision repair. These inhibitors demonstrate strong preclinical activity, with IC50 values of ~0.05-0.3 μ M in pancreatic cancer models and ~0.1-2.5 μ M in MPNST models.

Preliminary data show that disrupting key antioxidant regulators significantly enhances MPNST sensitivity to Ref-1 inhibition. In MPNST cell lines (NF90-8, RHT163, and RHT172), PRDX1 knockdown reduced the IC50 of Ref-1 inhibitors with APX2014 and showed up to ~7-8-fold increased sensitivity following PRDX1 depletion. These findings identify the redox buffering network as a key vulnerability. Our study focuses on four major redox regulators: peroxiredoxin-1 (PRDX1), thioredoxin-1 (TRX1), thioredoxin reductase-1 (TXNRD1), and glucose-6-phosphate dehydrogenase (G6PD) that maintain NADPH-dependent ROS detoxification in tumor cells.

To investigate these dependencies, we are performing CRISPR/Cas9 genetic perturbation studies across multiple MPNST cell lines, beginning with NF90-8 and expanding to ST88-14 and S462, to evaluate effects on proliferation, clonogenic survival, intracellular ROS, and APX2014 sensitivity. Parallel immunohistochemical analyses of human tumor tissue microarrays and genetically engineered mouse models are examining PRDX1 and TRX1 expression across tumor progression from plexiform neurofibromas to MPNST. In addition, a targeted CRISPR screen of ~200-300 redox-associated genes is being conducted to identify further synthetic lethal interactions with Ref-1 inhibition.

Using these genetic insights, we are identifying clinically relevant drugs targeting these redox nodes for combination with APX2014, including auranofin (FDA-approved TXNRD1 inhibitor currently in oncology trials (NCT01419691)), adenanthin (PRDX1 inhibitor in preclinical development), PX-12 (TRX1 inhibitor previously tested in Phase I solid tumor trials), and 6-aminonicotinamide (G6PD inhibitor). These combinations will be evaluated for synergistic antitumor activity in vitro and in orthotopic in vivo MPNST models. Together, this integrated approach aims to define redox dependencies in MPNST and establish a foundation for future combination therapeutic strategies.

Translational/Clinical Research

Graduate Student

POSTER #131

MYC AND HSF1 CO-AMPLIFICATION AS A BIOMARKER FOR TREATMENT IN HIGH-GRADE SEROUS OVARIAN CANCER

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Ovarian cancer is a disease that affects approximately 1 in 91 women. The 5-year relative survival percentage for those with the disease is 51%. The most lethal and common form of ovarian cancer is high-grade serous ovarian cancer (HGSOC). Current precision-based medicine approaches to treating HGSOC are limited to PARP inhibitors for BRCA mutant and homologous combination deficient patients. There is a desperate need for more options for ovarian cancer patients. Preferably these new treatments would both work for a large population of those with high-grade serous ovarian cancer and be precise to a specific biomarker of the disease. We have shown that one possible biomarker for disease treatment is the *MYC* and *HSF1* (*Heat Shock Factor 1*) co-amplification which occurs in approximately 31% of ovarian cancer patients. We have shown in cell viability assays, colony formation, and spheroid assays that ovarian cancers with this co-amplification are more sensitive to the PLK1i volasertib compared to ovarian cancers that do not carry this co-amplification. Since volasertib is not being pursued clinically as a monotherapy due to its own adverse effect concerns, we aimed to find if there are other drugs that HGSOC with the *MYC* and *HSF1* co-amplification are sensitive to. Considering that HSF1 and MYC are both transcription factors, we hypothesized that an epigenetic inhibitor could be an effective disruptor of MYC and HSF1 activity in HGSOC. We found in a drug screen of 479 drugs that ovarian cancers with this co-amplification are sensitive to class I HDAC inhibitors. We have shown using one of these top-performing HDAC class I inhibitors, entinostat, that MYC and HSF1 protein levels are decreased after 24 hours of treatment with MYC protein stability being able to be rescued by blocking the proteasome with MG132. RNA-sequencing of cells treated with entinostat show a significant decrease in MYC activity. Further studies will aim to establish the mechanism between how HDAC class I inhibition leads to a decrease in MYC and HSF1 protein and if this mechanism is responsible for cell death seen with class I HDAC inhibition. This establishes a promising new therapeutic strategy for approximately one-third of HGSOC patients that harbor the MYC-HSF1 co-amplification.

Translational/Clinical Research

Graduate Student

POSTER #132

FIELD STRENGTH-DEPENDENT PERFORMANCE VARIABILITY IN DEEP LEARNING-BASED ANALYSIS OF MAGNETIC RESONANCE IMAGING FOR BREAST TUMOR SEGMENTATION

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Background: Magnetic resonance images (MRIs) show distribution shifts due to scanner and acquisition variability. This study evaluates how magnetic field strength (1.5T vs. 3.0T) influences the performance and generalizability of deep learning-based breast tumor segmentation.

Methods: 1,271 dynamic contrast enhanced MRI examinations from the MAMA-MIA breast tumor segmentation dataset were analyzed, comprising 851 acquired at 1.5T and 420 at 3.0T. Three nnU-Net-based segmentation models were developed: A model trained on 1.5T data only (model1.5T), a model trained on 3.0T data only (model3.0T), and a model trained on pooled 1.5T and 3.0T data (modelcombined). Each model was evaluated on both 1.5T and 3.0T validation sets using the Dice similarity coefficient (DSC) as the primary segmentation metric. Field-strength-dependent performance differences were further investigated via Uniform Manifold Approximation and Projection (UMAP)-based clustering and radiomic analysis, including 23 first-order and texture features extracted from whole volumes and segmented regions.

Results: Model3.0T demonstrated superior performance on all validation sets. On 1.5T validation set, model3.0T achieved a DSC of 0.4939 (95% CI: 0.4495–0.5383) and significantly outperformed model1.5T (DSC: 0.4105 [95% CI: 0.3690–0.452]) and modelcombined (DSC: 0.3725 [95% CI: 0.3313–0.4136]) ($p < 0.0001$). A similar trend was observed on the 3.0T validation set, where model3.0T achieved a DSC of 0.4326 (95% CI: 0.3948–0.4704) significantly outperforming model1.5T (DSC: 0.2889 [95% CI: 0.2559–0.3220]) and modelcombined (DSC: 0.2684 [95% CI: 0.2356–0.3012]) ($p < 0.0001$). Radiomic analysis revealed moderate field-strength-dependent clustering in soft tissues (silhouette scores 0.23–0.29).

Conclusion: Magnetic field strength of the training data substantially influences the performance of deep learning-based segmentation of breast tumor, important for sensitivity to small lesions, suggesting that higher field strength yields superior generalizability. This warrants consideration of magnetic field strength as a confounding factor in studies evaluating AI performance on MRI, particularly in multicenter trials and longitudinal studies.

Translational/Clinical Research

Graduate Student

POSTER #133

RADIATION-INDUCED TRANSIENT METABOLIC REWIRING UNCOVERS AN ASCT2 THERAPEUTIC VULNERABILITY IN TRIPLE-NEGATIVE BREAST CANCER

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Abstract

Background: Glutamine metabolism is a vulnerability in TNBC but is buffered by metabolic flexibility. We posited that ionizing radiation (IR) enforces a short-lived glutamine dependency that can be targeted.

Methods: TNBC cell lines and in vivo models were subjected to IR with or without pharmacologic inhibitors of glutamine metabolism. We quantified glutamine flux using stable-isotope tracing, DNA repair kinetics, cell-cycle progression, apoptosis, tumor growth/metastasis, and changes in the immune microenvironment.

Results: IR triggered a radiation-related anabolic shift (RAAS), increasing ASCT2-mediated glutamine uptake and temporarily rerouting external glutamine-derived nitrogen toward de novo nucleotide synthesis. Isotope tracing showed exogenous glutamine supports post-IR glutamine → glutamate → IMP synthesis, whereas the aspartate → IMP pathway does not rely on external glutamine. Inhibiting ASCT2 with V-9302 abolished RAAS, impaired DSB repair, caused G2/M arrest, and increased apoptosis. Compared to other pathway inhibitors, ASCT2 blockade provided the most consistent radiosensitization across TNBC lines. In vivo, low-dose V-9302 combined with fractionated radiotherapy slowed primary tumor growth and reduced lung metastasis without causing significant systemic toxicity. The combination therapy increased CD8⁺/CD4⁺ T-cell infiltration, M1 macrophage polarization, and TNF- α production. Data support a metabolic bottleneck mechanism: limiting tumor glutamine access constrains tumor adaptation.

Conclusions: IR temporarily induces glutamine dependence in TNBC. ASCT2 inhibition (V-9302) takes advantage of this window to block glutamine-driven DNA repair and boost local anti-tumor immune polarization, leading to strong radiosensitization and anti-metastatic effects. These results support clinical trials combining glutamine transport inhibition with radiotherapy in TNBC.

Keywords: triple-negative breast cancer; glutamine transport; ASCT2; V-9302; radiotherapy; radiosensitization; metabolic plasticity; de novo nucleotide biosynthesis; DNA repair; tumor immune microenvironment; metabolic competition.

Translational/Clinical Research

Graduate Student

POSTER #134

COMPREHENSIVE SPATIAL EXPRESSION ATLAS OF COLORECTAL CANCER ACROSS CLINICALLY RELEVANT MOLECULAR SUBTYPES

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Colorectal cancer (CRC) is a complex ecosystem in which tumor cells interact with diverse accessory cell populations, including cancer-associated fibroblasts (CAFs), B cells, T cells, myeloid cells, and endothelial cells (ECs). Despite advances in single-cell genomics and bulk transcriptomics, fundamental questions remain regarding how ecological interactions between diverse cell populations manifest spatially, whether consistent spatial patterns emerge despite CRC heterogeneity, and how oncogenic drivers influence the spatial organization of the tumor microenvironment (TME).

To address these questions, we employed 10x Visium Spatially Resolved Transcriptomics (SRT) to construct a spatially resolved atlas of treatment-naïve CRC, validated using single-cell protein profiling via multiplex immunofluorescence (COMET). Our study encompasses clinically relevant molecular subtypes, including mismatch repair proficient (pMMR), mismatch repair deficient (dMMR), BRAF V600E-mutant, KRAS-mutant, and wild-type CRC. In total, we analyzed 22 tumor samples and two normal controls, generating 264,255 high-quality spots after stringent quality control.

By integrating spatial transcriptomics, single-cell proteomics, and ecological modeling with advanced computational analyses, we identified ten spatially organized multicellular neighborhoods, termed spatial ecotypes (SEs), that define the CRC microenvironment. These SEs, mapped through hierarchical network analysis, are governed by distinct biological programs orchestrating tumor-immune-stromal dynamics across three spatial territories: tumor core, peritumoral, and stromal zones. Tumor-enriched ecotypes exhibited activation of oncogenic programs including MYC targets, mTOR, and PI3K/AKT signaling, whereas immune-rich ecotypes demonstrated coordinated inflammatory and interferon responses. Stromal-dominant ecotypes were characterized by epithelial-mesenchymal transition, angiogenesis, and TGF- β signaling.

Comparative ecosystem analysis revealed fundamental spatial and functional differences between pMMR and dMMR tumors. Mismatch repair-deficient tumors maintain distributed immune activation across all spatial zones, with persistent CD8⁺ T cell and cytotoxic immune enrichment. In contrast, pMMR tumors construct stratified immunosuppressive barriers through myCAF-enriched peritumoral boundaries, elevated regulatory T cells, M2 macrophages, and enhanced TGF- β -driven stromal remodeling, providing a mechanistic basis for their resistance to immune checkpoint blockade.

Notably, BRAF V600E-mutant CRC tumor cells exhibit primitive fetal-like transcriptional signatures, neuroendocrine features, and bivalent epigenetic reprogramming reminiscent of embryonic stem cell states. These developmental programs actively reshape the TME by driving oncofetal reprogramming, characterized by spatial colocalization of FOLR2⁺ tumor-associated macrophages, POSTN⁺ CAFs, and PLVAP⁺ ECs, establishing an immunosuppressive oncofetal niche that fosters immune evasion and tumor progression.

These findings provide a high-resolution map of the spatial architecture of CRC, redefine it as a dynamic ecosystem rather than a collection of discrete subtypes, and establish a framework for subtype-specific biomarker discovery and novel therapeutic strategies targeting spatial tumor organization.

POSTER #135

KINASE-INDEPENDENT PIM2 SIGNALING CONTROLS MYC-DRIVEN TRANSCRIPTIONAL NETWORKS IN MULTIPLE MYELOMA

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Multiple myeloma (MM) remains an incurable plasma cell malignancy due to persistent drug resistance and cancer cell plasticity. Our previous work established that PIM2, a pro-survival kinase highly expressed in MM, exerts both kinase-dependent and kinase-independent functions critical to tumor cell persistence. JP11646 (JP1), a novel non-ATP-competitive PIM2 inhibitor, reduces both PIM2 mRNA and protein expression, suggesting an autoregulatory feedback mechanism. To dissect these mechanisms, we developed analogs JP2 (kinase-independent selective) and JP3 (kinase-dependent selective). We hypothesize that PIM2 overexpression is sustained through a kinase-independent, MYC-mediated positive feedback loop that can be disrupted by compounds targeting this function (JP1, JP2). Using ChIP-seq, RNA-seq, and co-immunoprecipitation, we examined how these inhibitors alter transcriptional networks. In PIM2 knockdown MM cells and JP1-treated MM cells, ChIP-seq revealed a global reduction in MYC binding across regulatory loci. RNA-seq analysis showed that JP1 and JP2—but not JP3—downregulated MYC target genes and suppressed pathways related to mitochondrial gene expression and translation. We confirmed these transcriptional effects using MRPS10, a representative mitochondrial gene, which demonstrated expected decreases in mRNA and protein levels following JP1 and JP2 treatment but not JP3. ChIP-qPCR at the CDK4 and PIM2 promoters confirmed reduced MYC occupancy following JP1, JP2, and MYC inhibitor (10058-F4), but not with AZD1208 or JP3. Moreover, ChIP-seq using FLAG-tagged exogenous PIM2 demonstrated PIM2 promoter binding by both wild-type and kinase-dead PIM2, confirming a kinase-independent PIM2–MYC regulatory circuit and autoregulatory feedback loop sustaining PIM2 expression. These findings identify the kinase-independent PIM2–MYC axis as a promising therapeutic target in refractory multiple myeloma.

Translational/Clinical Research

Graduate Student

POSTER #136

IMPROVING CROSS-ANCESTRY ACCURACY OF TUMOR MUTATIONAL BURDEN THROUGH CONTEMPORARY REFERENCE GENOMES

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Introduction: Tumor mutational burden (TMB) is an established predictive biomarker for response to immune checkpoint inhibitors, particularly anti-PD-1/PD-L1 therapies. Tumors with higher numbers of somatic mutations generate more neoantigens and respond better to immune checkpoint inhibitors. Thus, the FDA-approved cut-off for TMB is ≥ 10 somatic mutations per megabase (mut/Mb) of coding sequence. The gold standard method for TMB calculation uses matched tumor-normal sequencing. However, in routine clinical practice, TMB is often derived from tumor-only sequencing and filtered using public germline variant databases aligned to reference genomes such as GRCh37. Since newer reference genomes better represent true germline variability, we hypothesize that the aligning the somatic genomes to the newer GRCh38 alignment will produce more accurate TMB estimations.

Methods: To test our hypothesis, we obtained matched germline and tumor whole-exome sequencing data from participants enrolled in the Indiana University Simon Comprehensive Cancer Center Total Cancer Care protocol via the Manifold data browser (n=366). Our cohort includes self-reported Black (n=72) and White (n=294) participants. Genomic ancestry was inferred using ADMIXTURE in supervised mode with the 1000 Genomes reference panel. Tumor-normal and tumor-only TMB was calculated using standardized somatic variant calling and filtering approaches across three reference genome builds: GRCh37, GRCh38. Tumor-only TMB was calculated using tumor sequencing files aligned to GRCh37 and GRCh38. After filtering germline variants in the gnomAD database, cancer driver mutations in the COSMIC Cancer Gene Census, and variants with a variant allele frequency < 0.05 , TMB was calculated as variants per megabase of callable bases.

Results: Our tumor-normal analysis shows that TMB estimations were lower using GRCh38 than GRCh37 by 3 muts/Mb and 5 muts/Mb for self-reported Black participants (n=15) and White participants (n=35), respectively. Based on this finding, we also now hypothesize that TMB calculated using the Pangenome as the reference will produce even more accurate TMB estimations, especially for non-white patients. Based on the FDA recommended TMB cutoff for immune checkpoint therapy, the observed discrepancies have the potential to reclassification many patients for immune checkpoint inhibitor therapy.

Conclusion: The TMB calculations depend on the reference genome build that is used to estimate the number of tumor mutations. Newer genome builds that more comprehensively include germline genetic variants improve the estimations of the tumor mutation burden and may impact patients' eligibility for immune checkpoint therapy.

Translational/Clinical Research

Graduate Student

POSTER #137

DEEP LEARNING DERIVED FEATURES FROM PRE-TREATMENT MRI PREDICT FAILURE OF HIGH INTENSITY FOCUSED ULTRASOUND (HIFU) THERAPY IN PROSTATE CANCER PATIENTS.

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INTRODUCTION AND OBJECTIVES

High-intensity focused ultrasound (HIFU) is an emerging focal therapy for localized prostate cancer (PCa) for low and intermediate risk patients without the side effects associated with radical therapies. However, previous studies have shown that nearly 37% of patients experience recurrence following HIFU, especially those presenting with unfavorable intermediate risk disease. Identifying quantitative imaging markers from pre-HIFU MRI that can predict treatment response remains a key unmet clinical need. Artificial intelligence (AI) based methods have demonstrated promise in identifying prognostic biomarkers from pre-treatment prostate MRI for radical therapies. In this study, we evaluated whether deep learning derived quantitative imaging features extracted from pre-treatment prostate MRI are associated with HIFU treatment failure and whether these imaging signatures can stratify patients according to risk of recurrence following focal therapy.

METHODS

A retrospective cohort of N = 82 patients who underwent 3-Tesla prostate MRI prior to HIFU treatment was analyzed. Recurrence status was determined primarily using follow-up biopsy results, with elevated prostate-specific antigen (PSA) levels serving as supportive evidence in select cases. Bi-parametric MRI sequences including T2-weighted imaging and apparent diffusion coefficient (ADC) maps were analyzed. A previously trained 3D nnU-Net deep learning model was used as a feature encoder to extract quantitative imaging features from the ablated hemisphere region of interest corresponding to the treated side of the prostate. A total of 1,120 deep learning-derived imaging features were extracted per patient. Feature selection was performed using an ANOVA F-test, and the top eight discriminative features were retained. These selected features were used to train a logistic regression classifier (C_{AI}) using nested 5×5 cross-validation to evaluate predictive performance. Model-derived risk scores were subsequently used to stratify patients into high- and low-risk groups. Kaplan Meier survival analysis and the log-rank test were used to evaluate differences in recurrence-free survival between these groups.

RESULTS

The classifier trained on the selected deep learning features (C_{AI}) achieved a mean AUC of 0.68 ± 0.08 across cross-validation folds. Patients stratified using the model-derived risk score demonstrated significant separation in recurrence-free survival on Kaplan Meier analysis (log-rank $p < 0.05$). In contrast, traditional clinical markers including median PI-RADS score and Gleason grade group (ISUP) did not demonstrate statistically significant separation in survival outcomes.

CONCLUSIONS

Deep learning derived signatures within the ablated hemisphere from pre-HIFU MRI were prognostic of HIFU failure. Incorporating imaging-based biomarkers into treatment planning may improve patient selection and risk stratification for HIFU therapy. Future work validating these features on larger multi-institutional datasets may enable pretreatment identification of candidates most likely to benefit from HIFU.

Translational/Clinical Research

Graduate Student

POSTER #138

CONVERGENT CDK4/6 AND PI3K/MTOR PATHWAY HYPERACTIVATION DEFINES A TARGETABLE AXIS IN OSTEOSARCOMA ACROSS RB1-PROFICIENT AND RB1-DEFICIENT CONTEXTS

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Osteosarcoma (OS) in pediatric, adolescent, and young adult (AYA) patients is an aggressive malignancy with limited therapeutic progress. About 40% of OS patients develop metastases over time with 15-20% of OS patients already exhibiting metastases at initial diagnosis. Thus, there is a critical need to develop better therapeutics. Genomic analyses from our institution and others highlight recurrent alterations in CDKN2A and CDK4/6, suggesting that CDK4/6 inhibition (CDK4/6i) is a rational therapeutic vulnerability. Although RB1 proficiency (RB1+) is considered essential for CDK4/6i response, over 70% of OS tumors are RB1-deficient (RB1-), raising questions about the utility of CDK4/6i in these patients. Emerging evidence in other solid tumors and in our preliminary OS studies, suggests that CDK4/6i may retain antitumor activity in RB1- contexts. Compounding this complexity, pharmacologic CDK4/6 blockade induces compensatory PI3K/mTOR signaling, which can restore cyclin D-CDK4/6 activity and drive resistance.

We hypothesized that co-targeting PI3K/mTOR would enhance CDK4/6i efficacy irrespective of RB1 status by suppressing adaptive signaling. We evaluated palbociclib, voxtalisib, and the combination across RB1+ and RB1- OS models, using cell lines, patient derived xenografts (PDXs), and an experimental metastasis model. In vitro drug interactions (Chou-Talalay, Bliss) and mechanistic assays (cell cycle, senescence, autophagy) were complemented by in vivo tumor growth kinetics and pharmacodynamic profiling through histopathology, kinome, and proteomic analyses.

In RB1+ OS cells in vitro, palbociclib induced G1 arrest and senescence, accompanied by increased PI3K/AKT phosphorylation consistent with adaptive feedback. Voxtalisib suppressed this response, reinforced autophagic signaling, and maintained pathway inhibition. In vivo, combination therapy was well tolerated and produced significant tumor growth suppression in treatment-naïve and metastatic PDXs. In the RB1+ lung-colonization model, CDK4/6i alone reduced metastatic burden, with combination therapy achieving comparable control.

To interrogate RB1 as a biomarker, CRISPR-engineered human and mouse RB1- OS clones (MG63.3, K7M2) were generated and characterized. As expected, voxtalisib response was RB1-independent, and palbociclib sensitivity was reduced in RB1- cells. However, RB1- cells still exhibited growth inhibition at higher palbociclib concentrations. Notably, combination therapy of palbociclib+voxtalisib produced additive-to-synergistic growth inhibition regardless of RB1 status. In vivo validation studies are ongoing. These findings

identify convergent CDK4/6–PI3K/mTOR hyperactivation as a targetable axis in OS and support further evaluation of CDK4/6i-based strategies, including in RB1-deficient disease.

Translational/Clinical Research

Graduate Student

POSTER #139

CLASSIFICATION OF SEMINOMA AND NON-SEMINOMA TESTICULAR GERM CELL TUMORS USING ARTIFICIAL INTELLIGENCE (AI) APPROACHES ON MRI

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Classification of Seminoma and Non-Seminoma Testicular Germ Cell Tumors using Artificial Intelligence (AI) approaches on MRI

Background: Preoperative differentiation of seminoma from non-seminoma testicular germ cell tumors (TGCTs) is essential for guiding treatment decisions, as management strategies differ significantly between subtypes. Conventional MRI interpretation is subjective, and quantitative approaches leveraging AI may offer more reproducible tumor characterization. This study compares different AI based approaches including handcrafted radiomics, deep learning, and a late-fusion model for T2-weighted MRI-based TGCT subtype classification.

Methods: Thirty-six patients with histopathological confirmed TGCTs (27 seminoma, 9 non-seminoma) on 3T T2-weighted (T2W) MRI were retrospectively analyzed. Tumor segmentation was performed using an nnU-Net framework trained on N=23 patient studies and validated on N=13. A set of N=107 radiomic features quantifying tumor morphology, intensity distribution, and texture heterogeneity were extracted from the tumor regions on T2W using the PyRadiomics library. Feature selection was conducted using repeated nested stratified 5-fold cross-validation (200 repetitions), retaining features selected in $\geq 70\%$ of inner folds, followed by L1-regularized logistic regression. Machine learning models were trained to distinguish seminoma from non-seminomas that included a radiomic feature based model (CR), a ResNet-18 deep learning (CDL) model fine-tuned on T2W and late-fusion model (Ccomb) combining radiomics (CR) and DL (CDL) predicted probabilities was evaluated in terms of AUC, sensitivity, and specificity.

Results: Tumor segmentation model achieved a Dice similarity coefficient of 0.97 on the test set. Top ranked radiomic features that include shape-based sphericity, GLSZM zone entropy, GLCM contrast, GLDM dependence non-uniformity, and first-order kurtosis, reflecting tumor compactness, intratumoral heterogeneity, and gray-level texture complexity. CR achieved AUC = 0.93, sensitivity = 1.00, specificity = 0.82 while CDL resulted in AUC = 0.85, sensitivity = 0.56, specificity = 0.82. The fusion model Ccomb achieved an AUC of 0.91, sensitivity (1.00) and specificity (0.85).

Conclusions: In this small-cohort study, we observed that AI based classification models were promising in distinguishing seminomas from non-seminoma TGCTs on MRI. Given the small cohort of studies, radiomics-based machine learning outperformed deep learning and fusion approaches for MRI-based TGCT subtype differentiation. Ongoing work includes prospective external validation on larger datasets.

Figure:

Translational/Clinical Research

Graduate Student

POSTER #140

DNA METHYLTRANSFERASE INHIBITION PREVENTS PLATINUM-INDUCED OVARIAN CANCER STEM CELL ENRICHMENT

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Platinum-based therapy is the standard first-line treatment for high-grade serous ovarian cancer (HGSC). However, most patients develop resistance and recurrence despite an initial response to therapy. Ovarian cancer stem cells (OCSCs) are enriched in recurrent tumors and contribute to platinum resistance. These tumors also show promoter DNA hypermethylation, and DNA methyltransferase inhibitors (DNMTis) have been shown to restore sensitivity in platinum-resistant ovarian cancer cells. Here, we demonstrated that combining DNMTi with platinum prevented the platinum-induced enrichment of OCSCs and identified NF- κ B and STAT3 signaling pathways as potential regulators of platinum-induced OCSC enrichment. STAT3 was active at baseline in OC cells and platinum treatment alone activated NF- κ B while maintaining STAT3 activity. Platinum combined with DNMTi decreased STAT3 activation, while still inducing NF- κ B activation. Knockdown experiments demonstrated that the presence of both NF- κ B and STAT3 was necessary for platinum-induced OCSC enrichment. Analysis of STAT3 and NF- κ B subunit p65 CUT&RUN data showed increased binding in introns and intergenic regions in response to platinum. Additionally, DNMTi enriched NF- κ B binding at endogenous retroviruses (ERVs), which correlated with changes in expression of nearby genes when DNMTi was combined with platinum. We conclude that combining DNMTi with platinum modulates STAT3 and NF- κ B activation and genomic binding, potentially influencing target gene expression and preventing platinum-induced enrichment of OCSCs.

Translational/Clinical Research

Graduate Student

POSTER #141

INHIBITORS OF ENDOCANNABINOID DEGRADATION DIFFERENTIALLY SUPPRESS CHEMOTHERAPY-INDUCED NEUROPATHIC PAIN BEHAVIORS AND TUMOR GROWTH IN A MOUSE MODEL OF BREAST CANCER

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Female breast cancer has been the most diagnosed breast cancer worldwide, with approximately 2.3m million cases diagnosed in 2022. Chemotherapeutic treatment for breast cancer holds anti-tumor efficacy but also holds dose-limiting side effects like chemotherapy-induced peripheral neuropathy (CIPN). Pharmacological manipulations of the endocannabinoid system have shown promise in reversing CIPN and hold anti-cancer potential. More specifically, upregulation of the major endogenous cannabinoid 2-Arachidonoylglycerol (2-AG) via inhibition of its primary hydrolyzing enzyme, monoacylglycerol lipase (MAGL), shows efficacy in the treatment of CIPN behaviors. 2-AG binds to both cannabinoid type 1 (CB1) and cannabinoid type 2 (CB2) receptors, which have both displayed analgesic efficacy for CIPN when upregulated. The activation of CB1 and CB2 receptors has also displayed anti-cancer efficacy preclinically. Inhibition of MAGL has also reduced breast cancer tumor progression preclinically. Heightened MAGL expression in tumor cells have also been correlated with more aggressive cancer progression. MAGL inhibitors hold immense therapeutic potential for cancer patients, but CNS-penetrant MAGL inhibitors display tolerance in analgesic efficacy and physical dependence symptoms. We have previously discovered a brain impermeant MAGL inhibitor, LEI-515, that reversed established CIPN without producing analgesic tolerance, CNS-mediated side effects, or CB1-mediated physical dependence symptoms in non-tumor-bearing mice. Our group has also discovered a more potent brain impermeant MAGL inhibitor, JR045, that has more specific targeting to MAGL and does not have an off target of hormone sensitive lipase (HSL) LEI-515. In our preliminary studies ran in separate cohorts, mechanical and cold hypersensitivity were evaluated in tumor-bearing female mice that had 4T1 cells inoculated into their mammary fat pads. Mechanical thresholds were assessed using the von Frey test, whereas cold stimulus response times were measured using the acetone test, respectively. Tumor volumes were assessed once daily using calipers. Both brain impermeant MAGL inhibitors LEI-515 and JR045 successfully suppressed mechanical and cold hypersensitivity induced by paclitaxel in female mammary tumor-bearing mice. Interestingly, LEI-515 significantly enhanced paclitaxel's ability to reduce tumor size, whereas JR045 did not significantly enhance or impede paclitaxel's ability to reduce tumor size. Recently, HSL has been implicated as a driver for breast cancer tumor progression and metastasis. This may explain LEI-515's anti-tumor synergism with chemotherapeutic treatment, as LEI-515 suppresses HSL as an off-target effect. In mechanism of action studies, LEI-515 was found to be CB2-mediated in reversing CIPN-induced mechanical hypersensitivity, whereas JR045 required both CB1 and CB2 receptors to suppress mechanical hypersensitivity induced by taxane agent paclitaxel. In addition to these compounds, a brain penetrant MAGL inhibitor JZL184 was assessed as a positive control in tumor and non-tumor bearing studies. JZL184 failed to suppress mechanical or cold hypersensitivity when administered prophylactically alongside paclitaxel. This indicates a mechanistic difference between brain permeant and brain impermeant MAGL inhibitors in their respective abilities to suppress CIPN behaviors in tumor and non-tumor bearing animals. In addition to this, JZL184 did enhance paclitaxel's ability to slow breast cancer tumor progression in vivo, indicating that brain impermeability of MAGL inhibitors may not play a role in the anti-cancer potential of MAGL inhibitors. Lastly, colonic contents were significantly enhanced by chemotherapeutic agent paclitaxel compared to a vehicle control group, and these effects were not exacerbated by MAGL inhibitors JZL184 or LEI-515 in tumor-bearing mice. Our findings indicate that MAGL inhibition may be a promising combinatorial treatment alongside chemotherapeutic treatment for cancer patients regardless of brain permeability. In the context of

treating chemotherapy-induced peripheral neuropathy, brain impermeant MAGL inhibition may be a promising therapeutic avenue to pursue due to no analgesic tolerance development, no physical dependence, and minimal side effects.

Translational/Clinical Research

Graduate Student

POSTER #143

A NOVEL POST-SURGICAL STRATEGY USING NK CELL-DERIVED EXTRACELLULAR VESICLES TO PREVENT GLIOBLASTOMA RECURRENCE

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Glioblastoma (GBM) is the most aggressive primary brain tumor, with a median survival of ~15 months and nearly universal recurrence. The current standard of care includes maximal surgical resection followed several weeks later by radiotherapy and concurrent chemotherapy with temozolomide. However, complete tumor removal is impossible due to the highly infiltrative nature of GBM, and ~90% of recurrences arise near the resection margin. Residual tumor cells, particularly glioblastoma stem-like cells (GSCs), persist and become enriched after surgery, driving tumor recurrence through their self-renewal capacity and multilineage differentiation potential. In addition, the post-surgical tumor microenvironment (TME) is characterized by inflammation, blood-brain barrier disruption, and infiltration of stromal and immune cells, creating an immunosuppressive niche that supports tumor regrowth. Therefore, targeted eradication of residual GSCs within the surgical margin, together with reversal of the immunosuppressive TME that supports GSC propagation and tumor regrowth, represents a promising strategy to prevent GBM recurrence. Natural killer (NK) cells are innate immune effectors capable of eliminating tumor cells without prior antigen priming and have been shown to target GSCs. Increased NK cell infiltration has also been correlated with improved patient survival, supporting NK-based therapies for eradicating residual tumor cells, including GSCs, following surgery. However, endogenous NK cells within GBM are limited in number and often functionally suppressed by the TME. To overcome these limitations, we developed a strategy using NK cell-derived extracellular vesicles (NKEVs) to deliver NK cytotoxic activity directly to residual tumor cells within the post-surgical cavity. NKEVs were isolated from the human NK-92 cell line, which has been widely used in clinical trials. Transmission electron microscopy confirmed the characteristic cup-shaped morphology of the NKEVs, and protein analysis demonstrated the presence of NK cytotoxic proteins, including perforin and granzyme B. Functional assays using multiple patient-derived GSC lines showed that NKEVs induce dose-dependent cytotoxicity in GSCs while sparing normal brain cells such as astrocytes. Confocal microscopy further demonstrated efficient uptake of NKEVs by GSCs. RNA sequencing analysis comparing NKEV-treated GSCs and control revealed widespread transcriptional changes associated with increased apoptosis, cell cycle arrest, inflammatory signaling, interferon responses, and antigen processing and presentation pathways. Gene set enrichment analysis further indicated activation of innate immune sensing and viral mimicry pathways, including RIG-I/MDA5/MAVS and cGAS-STING signaling, as well as pathways associated with antitumor T cell responses and M1 macrophage activation, suggesting that NKEVs may convert the immunologically “cold” TME into a more immune-active “hot” state. Ongoing studies are evaluating the therapeutic potential of NKEVs in GBM surgical resection mouse models to determine whether local delivery of NKEVs into the tumor cavity can eliminate residual GSCs and reshape the tumor microenvironment *in vivo*. Together, these findings support NKEVs as a promising postoperative immunotherapeutic strategy for preventing GBM recurrence.

Translational/Clinical Research

Graduate Student

POSTER #144

PREGNANCY, LACTATION, AND INVOLUTION-INDUCED REMODELING AND CELL STATE CHANGES IN BREAST TISSUES OF HEALTHY WOMEN.

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Pregnancy, lactation, and involution have opposing effects on breast cancer incidence and tumor aggressiveness. While breast feeding and subsequent involution protects against breast cancers, pregnancy-associated breast cancers (breast cancers diagnosed during pregnancy and within 2 years postpartum) are associated with poor outcome. Clear understanding of these opposing effects of reproductive cycle needs single cell resolution of breasts during pregnancy, lactation and involution. We recently reported a reference single nucleus chromatin accessibility and transcriptome atlas of breast tissues of women of diverse genetic ancestry and reported 10 major cell types in the breasts. These include three epithelial cell types [luminal hormone sensing (LHS), luminal adaptive secretory precursors (LASP), and basal-myoeptithelial cells (BM)], two adipocyte subtypes, two endothelial cell subtypes, T cells, macrophages, and fibroblasts. In this study, we generated single nucleus atlas of breast tissues of healthy women collected during pregnancy, lactation, and involution. Pregnancy, lactation, and involution are associated with dramatic changes in cell type proportions and cell state shifts within a cell type. LASP cell numbers increased from 24% in the normal reference breasts to 71% during pregnancy, 63% during lactation but returned to 25% during involution. There is also a dramatic shift in LASP cell states during pregnancy, lactation and involution. While the LASP cells in the normal reference breasts and breasts during pregnancy were in both LASP-basal-luminal (LASP-BL) and LASP-alveolar (LASP-AP) states, LASP-AP cells were dominant in lactating and involuting breasts. Increase in LASP cells during pregnancy and lactation is at the expense of BM cells, which reduced from 27% in the normal reference breasts to 7-9% during pregnancy and lactation. While T cell proportion was similar between normal reference and pregnant breasts (2-3%), it increased to 7-8% during lactation and involution, which a recent study has suggested to be responsible for breast feeding-associated protection against breast cancer. Increase in T cells during lactation and involution is likely due to expansion of tissue resident T (T_{RM}) cells as T cells of lactating and involuting breasts expressed higher levels of T_{RM} cell markers CD69 and CXCR6. We present pregnancy, lactation and involution-associated chromatin accessibility and transcriptome changes at individual gene and cell type levels. Several biomarkers associated with these changes have been identified, which will be a useful resource to mechanistically evaluate evolution of breasts during reproductive history and breast cancer development.

Translational/Clinical Research

Laboratory Research specialist

POSTER #145

INTEGRATED TRANSCRIPTOMIC ANALYSIS IDENTIFIES TNFR2 AND LGALS9/TGF β SIGNALING AXES ASSOCIATED WITH REGULATORY T CELL PROGRAMMING IN NF1-ASSOCIATED MALIGNANT PERIPHERAL NERVE SHEATH TUMORS

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Neurofibromatosis type 1 (NF1) is an inherited tumor predisposition syndrome caused by pathogenic variants in the *NF1* gene, resulting in dysregulated RAS signaling and a propensity to develop neurocutaneous tumors. Plexiform neurofibromas (pNF) are congenital peripheral nerve sheath tumors that arise from NF1-deficient Schwann cell precursors and are present in up to 50% of individuals with NF1. A subset of pNF can transform into malignant peripheral nerve sheath tumors (MPNST), which occurs in approximately 8–13% of individuals with NF1 and represents the leading cause of early NF1-related mortality. MPNST are highly aggressive sarcomas with poor prognosis, characterized by an immunosuppressive tumor microenvironment (TME) that limits anti-tumor immunity and lacks effective targeted immunotherapies. To dissect the cellular and molecular architecture of the NF1 TME, we integrated single-cell RNA sequencing data from 67 human NF1 tumor samples (45 pNF, 22 MPNST). Cell identities were annotated using canonical marker genes for stromal and immune populations, and tumor cells were identified through copy number variant inference. Ligand-receptor interactions were modeled separately in pNF and MPNST using CellChat, and sample-level associations were tested between inferred signaling programs and a defined Treg functional gene module. In MPNST, increased expression of *TNFR2* on regulatory T cells, within the TNF α /TNFR2 signaling axis, was positively correlated with enhanced Treg programming. Additionally, elevated *LGALS9* expression in macrophage-like cells, coupled with increased *TGFBRI* expression in non-Treg populations, was similarly associated with elevated expression of *FOXP3* and other core Treg effector genes. These findings elucidate TNFR2-mediated signaling and a macrophage-driven LGALS9/TGF β 1 axis as potential drivers of immunosuppressive Treg induction in NF1-associated MPNST. Our results reveal novel, testable therapeutic targets aimed at disrupting Treg-mediated immune evasion and restoring anti-tumor immunity in NF1-associated MPNST.

Translational/Clinical Research

MD/PhD Student

POSTER #146

PROGNOSTIC UTILITY OF LACTATE DEHYDROGENASE IN PATIENTS WITH METASTATIC SEMINOMA

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Background: Serum tumor markers play an important role in risk classification, prognostication, staging, and monitoring of treatment in germ-cell tumors (GCTs). While alpha-fetoprotein (AFP) and human chorionic gonadotropin (hCG) are validated prognostic markers, recent investigations have suggested a correlation with lactate dehydrogenase (LDH) and treatment response and survival. Here, we evaluated LDH levels and survival in patients with advanced seminoma.

Methods: The prospectively maintained Indiana University (IU) testicular cancer database was queried for patients with metastatic seminoma treated with first-line therapy. Pts were assigned to having a LDH <2.5 upper limit of normal (ULN) or ≥ 2.5 ULN based on pre-treatment LDH levels. Baseline characteristics were summarized. The Kaplan-Meier method was used to analyze progression free survival (PFS) and overall survival (OS).

Results:

113 pts were included in the study. Median age at diagnosis was 39.21 (22.80-68.72). Primary site was testis in 104 (92%), retroperitoneum in 6 (5.3%), and mediastinum in 3 (2.7%). Metastasis sites were retroperitoneal lymph nodes in 85%, pelvic lymph nodes in 12.4%, lung in 7.1%. IGCCCG risk was good in 92% and intermediate in 8%. 68.6% of pts were treated with BEPX3, 19.8% with EPX4, 1.2% with VIPX4, and 9.3% with other regimen. Median hCG was 3.50 (0.5-11,600). Median LDH was 349 (34-6550).

Median follow-up was 2.30 years (0.02-19.11). 61 pts had LDH <2.5 ULN, 52 pts had LDH ≥ 2.5 ULN. For those with LDH <2.5 ULN, 3yr PFS was 91.5% (78.7-96.8) vs. 62.8% (45.2-76.1) for those with LDH ≥ 2.5 ULN, $p = 0.0050$. However, 3yr OS for those with LDH <2.5 ULN was 100% (100-100) vs. 95.2% (82.0-98.8), $p = 0.6218$.

Conclusions: Patients with metastatic seminoma and LDH ≥ 2.5 ULN had inferior 3-yr PFS compared with normal LDH, but there was no difference in OS.

POSTER #147

EVALUATING THE RELATIONSHIP BETWEEN SOCIAL VULNERABILITY AND CANCER RECURRENCE IN HEAD AND NECK CANCER PATIENTS IN INDIANA

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Background: Head and neck cancer (HNC) outcomes are influenced by multiple clinical and sociodemographic factors, including access to timely diagnosis, treatment, and appropriate follow up care. The Centers for Disease Control and Prevention's Social Vulnerability Index (SVI) is a composite measure that may help identify populations at risk for barriers to healthcare access and delivery. This study examines the association between SVI and cancer recurrence in HNC patients in Indiana.

Methods: Patients with HPV- mucosal head and neck squamous cell carcinoma who underwent surgical treatment at a tertiary academic medical center between 2013-2023 were reviewed. Patient addresses were linked to census tracts to assign SVI values. The SVI consists of a number 0-1 for each county in the US, with values closer to 0 being less at risk, and values closer to 1 being more at risk. Additionally, the CDC provides a further breakdown of SVI into four sub-themes. These sub-themes are socioeconomic status, household characteristics, racial and ethnic minority status, and housing type/transportation. SVI and the sub-themes were analyzed both as raw values and broken up into quintiles. Univariate and multivariate logistic regression models were performed to evaluate associations between SVI and cancer recurrence.

Results: Across 112 eligible patients, SVI was found to be significantly associated with increased odds of recurrence for both raw value ($p=0.017$) and quintiles ($p=0.031$). Additionally, the sub-theme of socioeconomic status was significantly associated with increased odds of recurrence ($p=0.043$, $p=0.025$). When performing multivariate analysis to control for gender, age, geographic classification, marital status, medical comorbidities, BMI, tobacco use, alcohol use, substance abuse, tumor location, advanced cancer at diagnosis, and presence of adjuvant therapy, SVI became barely insignificant at $\alpha = 0.05$ ($p=0.063$).

Conclusion: Higher social vulnerability was associated with increased odds of cancer recurrence following surgical treatment for HNC patients in univariate analysis, but not multivariate. Socioeconomic status is a sub-theme of SVI that may contribute to disparities in oncologic outcomes. These findings suggest that social determinants of health may have an impact on overall cancer outcomes. These findings support further investigation into the relationship between SVI and oncologic outcomes, as well as the mechanisms by which subthemes may contribute to worse HNC outcomes. Targeted interventions addressing barriers related to socioeconomic status may help improve cancer surveillance.

Translational/Clinical Research

Medical Student

POSTER #148

EFFECT OF PARP INHIBITORS ON MITOCHONDRIAL DYNAMICS IN PROSTATE CANCER CELLS

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Introduction. Prostate Cancer is the second leading cause of cancer death in men in US. The primary treatment for prostate cancer is androgen deprivation therapy (ADT). If cancer progresses while being treated with ADT, the cancer is classified as metastatic castration-resistant prostate cancer. Poly (ADP-ribose) polymerase inhibitors (PARPi) have demonstrated effectiveness in treating castration-resistant (and castration-sensitive) prostate cancer and were recently approved by the FDA. While these agents prevent cancer cells from repairing their nuclear DNA leading to cell death, their impact on organelle such as mitochondria dynamics remain unknown. Therefore, the aim of this study is to investigate the effects of PARPi compared to Antimycin A, electron transport chain complex III inhibitor, on mitochondrial membrane dynamics, specifically mitochondrial interactions with the endoplasmic reticulum and plasma membrane.

Methods. LNCaP and MDA PCa 2b, human prostate cancer cell lines, were treated with 12.5 μ M of Olaparib and Rucaparib (FDA-approved PARPi), and with Antimycin A for 48 hours. A proximity ligation assay (PLA) was used to highlight mitochondrial interactions with the endoplasmic reticulum and with the plasma membrane. Confocal microscopy was used to image these interactions. A custom-made algorithm on CellProfiler was implemented to quantify the number of interactions between the organelles in the PLA. A pretrained machine learning model, Cellpose, was used to segment the nuclei and obtain the number of interactions between organelles per cell.

Results. When treated with Olaparib and with Rucaparib, LNCaP cell lines had more mitochondria-endoplasmic reticulum interactions compared to the control ($P < 0.05$). There were no significant changes in the number of mitochondria-plasma membrane interactions in LNCaP cell lines when treated with PARPi. There were no significant changes in the number of mitochondria-endoplasmic reticulum and mitochondria-plasma membrane interactions in MDA PCa 2b cell lines when treated with PARPi. Interestingly, preliminary results for treatment with Antimycin A demonstrate increased mitochondria-endoplasmic reticulum interactions compared to untreated control in both cell lines.

Conclusion. Findings suggest that Antimycin A and PARPi affect organelle crosstalk in prostate cancer cells.

Translational/Clinical Research

Medical Student

POSTER #149

ARTIFICIAL INTELLIGENCE APPROACHES FOR QUANTITATIVE ANALYSIS OF LYMPHATIC IMAGING IN TRANSLATIONAL DISEASE RESEARCH

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Background:

Artificial intelligence (AI) has had a growing impact on biomedical research by enabling objective and scalable analysis of complex imaging datasets. In lymphatic biology and lymphedema research, accurate quantification of histologic and near-infrared imaging data remains challenging due to tissue heterogeneity, staining variability, and artifacts that can obscure true biological signal. Traditional manual analysis is labor intensive and limited by human visual interpretation. AI-based image analysis offers a potential solution by enabling automated identification of biologically relevant spatial patterns across large imaging datasets.

Methods:

We implemented two complementary AI-based approaches to support translational lymphatic research. The first approach utilizes annotation-driven platforms that allow investigators to train algorithms through expert labeling of image features. These tools enable automated classification, segmentation, and phenotyping of structures in histologic and fluorescence microscopy datasets while allowing batch processing of large image collections. The second approach involves custom machine learning and deep learning workflows. Convolutional neural networks (CNNs) are trained on labeled datasets of normal and diseased lymphatic tissues to identify pixel-level features associated with lymphatic remodeling and inflammatory changes. These models iteratively learn spatial patterns through convolutional filtering and hierarchical feature extraction, enabling classification and prediction based on complex image characteristics.

Results:

Together, these approaches enable automated extraction of quantitative metrics from histologic and lymphatic imaging datasets, including cellular morphology, staining patterns, and structural organization of lymphatic networks. By integrating multiple spatial and morphological features simultaneously, AI-based analysis improves reproducibility and reduces observer bias compared with traditional manual methods.

Conclusion:

Artificial intelligence provides a scalable and generalizable framework for quantitative analysis of lymphatic imaging data. By combining annotation-driven tools with custom machine learning pipelines, investigators can perform standardized and reproducible analyses across large datasets. These approaches may enhance the study of lymphatic remodeling and inflammatory tissue changes in lymphedema and other disease processes, supporting more objective investigation of lymphatic biology in translational research.

Translational/Clinical Research

Medical Student

POSTER #150

MOTION MANAGEMENT IN PEDIATRIC RADIOTHERAPY: ASSESSMENT OF INTRAFRACTION MOTION USING AN AUDIO/VISUAL DISTRACTION METHOD

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Purpose/Objective(s): Immobilization for pediatric cancer patients often requires the use of general anesthesia (GA) to ensure safe setup and minimize the risk of intrafraction motion; however, daily anesthesia can incur significant medical risk and healthcare cost. We hypothesized that the use of the Audio-Visual Assisted Therapeutic Ambience in Radiotherapy (AVATAR) audio/visual distraction technique would produce acceptable intrafraction motion management compared to the use of general anesthesia thereby reducing anesthesia-related risks.

Materials/Methods: A retrospective analysis was conducted on 30 patients with local stage III Wilms tumor (20 AVATAR, 10 GA) treated at a single institution between 2019 and 2025 with anterior posterior (AP) and/or posterior anterior (PA) treatment fields. Intrafraction motion was quantified by measuring the displacement of the treatment isocenter relative to stable vertebral landmarks using paired daily kilovoltage setup (AP or PA image) and megavoltage field verification images (AP or PA) obtained on the first day of treatment. Measurements were independently verified by two reviewers. Differences in horizontal (lateral) and vertical (superior-inferior) shifts between the two cohorts were analyzed using independent t-tests. The time intervals between setup imaging and field verification imaging on the first day, and between setup imaging and the completion of treatment delivery for the third and fifth fractions, were captured from the record and verify system.

Results: No statistically significant difference in intrafraction motion was observed between the two modalities. Mean horizontal displacement was 1.1 +/- 1.7 mm for AVATAR vs. 0.6 +/- 0.7 mm for GA (p=0.20). Mean vertical displacement was 0.9 +/- 1.2 mm for AVATAR vs. 0.6 +/- 0.04 mm for GA (p=0.23). Inter-observer variability was minimal (average 0.8 mm). The average time between setup imaging and field verification imaging on the first day (intrafraction motion measured in this study) was greater than the average time from setup imaging to completion of treatment delivery at the 3rd and 5th fraction (Table).

Conclusion: The AVATAR system demonstrated effective intrafraction immobilization for patients with stage III Wilms tumor with average vertical and horizontal displacement well below acceptable PTV margins. In addition, we demonstrate that the intrafraction measurement points in this study effectively estimate the actual treatment delivery time for these patients. These findings support AVATAR as a safe, non-invasive standard of care for select pediatric Wilms tumor patients, offering a viable alternative that mitigates the physiological and logistical burdens of daily anesthesia.

	1st Fx Average KV to MV Time	3rd Fx Average KV to Tx Completion Time	5th Fx Average KV to Tx Completion Time
AVATAR	263.7s	180.2s	161.3s
GA	290.4s	154.5s	158.0s

POSTER #151

TRENDS IN AGGRESSIVE END-OF-LIFE CARE FOR METASTATIC CERVICAL CANCER

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Purpose/Objective(s): Aggressive medical intervention near the end of life often incurs high costs with minimal benefit to patient quality of life. While recent clinical trials have demonstrated a survival benefit to chemotherapy and immunotherapy in metastatic cervical cancer, participants in these trials generally had an excellent performance status and a life expectancy of at least several months. This study investigates trends in chemo/immunotherapy and radiation for patients with metastatic cervical cancer, and whether there has been a shift away from palliative radiation toward more aggressive use of immunotherapy in patients with a very poor prognosis.

Materials/Methods: A retrospective analysis was conducted using the National Cancer Database. The study population included all patients diagnosed with metastatic cervical cancer between 2015 and 2022. Outcomes were analyzed for the total cohort and specifically for a subset of patients who died within 90 days of diagnosis (DWND) – a proxy for poor prognosis/performance status and end of life care.

Results: The study identified 10,285 patients with metastatic cervical cancer, of whom 2,399 (23.3%) died within 90 days of diagnosis. Throughout the study period, chemotherapy use remained stable for both the overall population (~69%) and the DWND group (~24%). In contrast, immunotherapy use saw a significant increase, rising from 11% in 2014 to 44% in 2022 for all patients, and from 0% to 12% in the DWND group. Notably, the use of radiation therapy showed a downward trend in both groups. In the DWND cohort, any radiation use fell from 37% to 21%.

Conclusion: While treatment patterns for chemotherapy and immunotherapy generally align with current guideline-recommended therapy for otherwise healthy patients with metastatic disease, there is a concerning trend toward increased immunotherapy use in patients with a very poor prognosis – the group of patients least likely to derive a survival or quality of life benefit from this treatment. Conversely, the utilization of radiation therapy has decreased among patients who DWND, a patient population that is often profoundly symptomatic near the end of life. These findings highlight the importance of further discussion regarding the relative value of treatment options near the end of life for patients with a poor prognosis.

Metastatic Cervical Cancer 2014 2015 2016 2017 2018 2019 2020 2021 2022

All patients, n 1,089 1,119 1,100 1,215 1,171 1,108 1,083 1,195 1,205

Chemo 69% 69% 69% 69% 68% 69% 70% 66% 70%

Immuno 11% 15% 19% 19% 27% 30% 30% 38% 44%

Any rads 54% 53% 49% 50% 52% 47% 48% 41% 42%

>15 fxs rads	30%	26%	25%	26%	26%	22%	23%	18%	18%
Death within 90 days, n	261	264	275	252	269	265	233	298	282
Chemo	25%	23%	22%	25%	25%	22%	27%	25%	22%
Immuno	0%	1%	4%	4%	5%	8%	9%	10%	12%
Any rads	37%	34%	33%	31%	29%	28%	26%	26%	21%
>15 fxs rads	9%	5%	4%	3%	2%	2%	3%	3%	2%

Translational/Clinical Research

Medical Student

POSTER #152

EVALUATING GUIDELINE CONCORDANCE IN COLORECTAL CANCER USING A TWO-STAGE LLM-BASED ABSTRACTION PIPELINE

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Introduction: Evidence-based cancer guidelines, including molecular testing recommendations in colorectal cancer (CRC), improve outcomes, but concordance varies across settings and populations. Manual abstraction to assess concordance is labor-intensive and difficult to scale. Large language models (LLMs) can extract and synthesize information from unstructured text, enabling automation of research workflows and quality evaluation. We developed an LLM-enabled automated pipeline to assess guideline concordance from unstructured clinical notes.

Methods: An extraction pipeline was developed using gpt-oss-120b, an open-weight LLM, designed for high-reasoning, tool use, and agentic workflows. Clinical notes from Stage I-III CRC patients were split into two cohorts using keyword-based screening: notes containing mismatch repair (MMR-related; n=495) terms and those without MMR-related terms (non-MMR; n=1,000). The extraction pipeline then proceeded with broad entity extraction (e.g., test type, date, and results), followed by date reconciliation to determine the earliest date and categorize results as pMMR/MSS vs. dMMR/MSI-H.

Results: In the non-MMR cohort, the model achieved 99.9% specificity (999/1,000 classified as null). In the MMR cohort, a definitive status was extracted from 69.5% of notes (293 pMMR/MSS and 51 dMMR/MSI-H). At the patient level, the pipeline successfully identified definitive molecular profiles for 101 patients (59.1%) in the MMR cohort. Overall, these findings indicate high precision for automated abstraction, with moderate note-level sensitivity.

Conclusion: The multi-stage LLM pipeline effectively extracts and interprets longitudinal electronic health records of CRC patients. By automating the identification of guideline-concordant care, this approach can enable large-scale quality audits and support near-real-time feedback to surgical teams, enhancing quality improvement initiatives.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #153

TIMING AND SEQUENCE OF SYSTEMIC THERAPY AND CYTOREDUCTIVE SURGERY IN A NATIONAL STUDY OF STAGE IV COLORECTAL CANCER WITH ISOLATED PERITONEAL METASTASES

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INTRODUCTION

Stage IV colorectal cancer (CRC) with isolated peritoneal metastases (PM) has classically been treated with systemic therapy alone, but the addition of colorectal resection with cytoreductive surgery (CRS) has emerged as a potential treatment. However, the optimal sequencing of systemic therapy and surgery is unknown. This study examined (1) current practice patterns for sequencing of CRS and systemic therapy, (2) factors associated with treatment sequence selection, and (3) the association between timing of treatment sequence on overall survival (OS).

METHODS

Patients with stage IV CRC with isolated PM in the 2016-2021 National Cancer Database who underwent both chemotherapy and colorectal resection were analyzed. Treatment sequence timing was defined as the interval between induction chemotherapy or upfront surgery followed by surgery or adjuvant chemotherapy. A multivariable logistic regression model analyzed patient, tumor, and hospital factors associated with treatment sequence. Adjusted Cox proportional hazards modeling examined associations between treatment sequence and 5-year OS.

RESULTS

Of the 242,581 patients with stage IV colorectal adenocarcinoma, 27,072 (9.0%) had PM. Among these patients, 6,217 (4.1%) had definitive surgical resection and chemotherapy. 1,666 (26.8%) patients received induction chemotherapy and 4,551 (73.2%) had surgery followed by adjuvant therapy. Compared to induction chemotherapy followed by surgery, patients were more likely to have upfront surgery if female, (OR 1.26, 95% CI 1.11-1.44; $P < 0.001$), Hispanic, (OR 1.37, 95% CI 1.05-1.78; $P = 0.019$), uninsured (OR 2.22, 95% CI 1.59-3.10; $P < 0.001$) or covered by Medicare (OR 1.36, 95% CI 1.12- 1.65; $P = 0.002$). Upfront surgery was also less likely to be performed at academic centers (OR 0.33, 95% CI 0.25-0.44; $P < 0.001$). Compared to upfront surgery, induction chemotherapy was associated with improved survival when CRS was performed after 3-6 months (HR 0.79, 95% CI 0.70-0.89, $P < 0.001$) or >6 months (HR 0.75, 95% CI 0.66-0.86; $P < 0.001$). CRS within 0-3 months was associated with worse survival (HR 1.51, 95% CI 1.28-1.78, $P < 0.001$).

CONCLUSIONS

Only a quarter of patients received induction chemotherapy before surgery and those patients were less likely to be female or uninsured but more likely to be treated at academic centers. In this retrospective analysis, OS

was improved in patients who had induction chemotherapy ≥ 3 months prior to CRS suggesting that a longer interval between induction chemotherapy and CRS for colorectal PM may be beneficial.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #154

THE NRF2 INHIBITOR MSU38225 SYNERGIZES WITH KRAS INHIBITION TO ARREST TUMOR GROWTH IN LUNG CANCER

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Nuclear factor erythroid 2-related factor 2 (*NRF2*) is a transcription factor that coordinates the defense against toxic and oxidative stress. *NRF2* activity is commonly elevated in many types of cancer, conveying the cancer cells with growth benefits and promoting tumor progression and metastasis. Moreover, high levels of *NRF2* protect cancer cells against targeted immune and chemotherapy. *KRAS* is an important regulator of cell growth and survival and is frequently mutated in certain types of cancer, such as pancreatic, colon, and lung cancer. *KRAS*G12C mutations are present in 11-14% of non-small cell lung cancer (NSCLC) patients and constitute 45% of all *KRAS* mutations in this type of cancer. While *KRAS*G12C-specific inhibitors like sotorasib and adagrasib prove to be highly effective, patients quickly develop resistance and relapse. Amongst several mechanisms, *KRAS* mutant cells can acquire independence from *KRAS* by boosting *NRF2* signaling, which, in turn, activates metabolic pathways that allow for the continuous growth of the cancer cells.

We hypothesize that combining mutant *KRAS* inhibitors with *NRF2* inhibition will result in the more efficient killing of cancer cells and, consequently, a more durable therapy response. Previously, we identified a novel *NRF2* inhibitor, MSU38225, in an in vitro screening assay using human NSCLC cells. To further test its efficacy, we performed cell viability and proliferation assays in multiple cancer cell lines, treating either MSU38225 alone or in combination with the *KRAS* inhibitors, adagrasib or daraxonasib. Our data show that simultaneous targeting of *NRF2* and *KRAS* has a synergistic effect on inhibiting cancer cell proliferation. To test our hypothesis in vivo, we established a subcutaneous mouse model of lung cancer and treated these mice with either adagrasib, MSU38225, or a combination thereof. Preliminary results demonstrate the inhibitory effect of either drug or their combination on tumor growth. We are currently evaluating the efficacy of this combinatorial approach in cell lines exhibiting differential levels of *NRF2* activity. Furthermore, we will elucidate the effect of *NRF2* inhibition on the immune environment and how this translates to reduced tumor growth. Lastly, we aim to identify a subset of human patients that will be most likely to benefit from this approach. Together, we demonstrate that targeting *NRF2* signaling, either alone or in combination with *KRAS* inhibitors, constitutes a promising approach in treating lung cancer with *KRAS* mutations.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #155

ESTABLISHING HISTONE 3 GLUTAMINE 5 HISTAMINYLATION AS A NEW EPIGENETIC MARKER IN GASTRIC CARCINOGENESIS

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Gastric cancer (GC) is the 5th most diagnosed cancer worldwide. Despite advances in treatment, GC continues to be a leading cause of cancer-related mortality, because it is frequently diagnosed at advanced stages, where the five-year survival rate is approximately 7%. Histologically, GC is classified into intestinal and diffuse subtypes, each arising through distinct biological pathways. The intestinal type develops through a multistep cascade, beginning with chronic atrophic gastritis, commonly driven by stomach infection by *Helicobacter pylori*, progressing to intestinal metaplasia, then dysplasia, and ultimately invasive carcinoma. In contrast, the diffuse type lacks a clearly defined precursor sequence and is often more aggressive. A major dilemma in GC management is that gastric intestinal metaplasia, a highly plastic and potentially reversible epithelial state likely driven by epigenetic reprogramming, precedes most intestinal-type tumors. However, only a subset of patients with metaplasia progresses to malignancy and highlights the urgent need for reliable epigenetic biomarkers that can stratify patients according to progression risk at the early stage. A recently discovered histone modification is “histaminylation,” wherein the hormone histamine is installed on the glutamine 5 residue on histone 3. The enzyme transglutaminase 2 (TGM2) both “writes” and “erases” this H3Q5his epigenetic mark.

In gastric cancer, transglutaminase 2 (TGM2) is frequently upregulated, and histamine levels are particularly elevated in the gastric microenvironment, especially during infection with *Helicobacter pylori*. Based on these observations, we hypothesize that TGM2-mediated histone 3 glutamine 5 histaminylation could drive chromatin remodeling and epigenetic reprogramming that promotes the progression from metaplasia to malignancy. To test this hypothesis, we infected AGS gastric adenocarcinoma cells with *H. pylori* at varying multiplicities of infection and time points. Remarkably, infected cells exhibited significantly elevated H3Q5his levels compared to mock-treated controls, confirming that H3Q5his is induced following *Hp* exposure. Also, infection resulted in a 2.5-fold increase in TGM2 expression after 24 hours. To further investigate H3Q5his accumulation in the context of gastric disease, we utilized our *Hp*+KRAS+ transgenic mouse model, which develops gastric intestinal metaplasia and dysplasia within 12 weeks of *H. pylori* infection and tamoxifen-mediated induction of oncogenic KRAS (G12D). TGM2 and H3Q5his expression were quantified in gastric tissues from these mice by western blotting. We observed a significant enrichment of TGM2 and H3Q5his in *Hp*+KRAS+ stomachs compared to healthy stomachs. As well, immunostaining of fixed stomach tissues showed a significant enrichment of H3Q5his puncta in *Hp*+KRAS nuclei. In short, these findings identify H3Q5his as a novel epigenetic biomarker of *H. pylori*-associated metaplasia. Future studies will use pharmacological and genetic approaches to define the consequences of H3Q5his during gastric carcinogenesis. Ultimately, therapeutic targeting of TGM2 may offer a strategy to reverse premalignant epigenetic states and prevent gastric cancer progression at its earliest stages.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #156

LUNG-RADS 0: INTENT VERSUS EXECUTION

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Background

Lung cancer screening (LCS) with low-dose CT (LDCT) reduces lung cancer mortality but is associated with false-positive findings that may increase healthcare costs and patient anxiety. Infectious and inflammatory findings are a major contributor to false positives. Lung-RADS v2022 introduced the ability to classify indeterminant infectious/inflammatory findings as Lung-RADS 0 (LR0), requiring short-term follow-up. Further recommendations advise that tree-in-bud (TiB) nodules and small ground-glass (GG) nodules be categorized as Lung-RADS 2, Benign. This study evaluated institutional utilization of LR0, the application to TiB/GG nodules, and the impact of cardiothoracic (chest) subspecialty training on use of guidelines.

Methods

We performed a retrospective review of LCS LDCT exams that received a classification of LR0 between January 1, 2024 and December 31, 2024. Cases were excluded if the LR0 was a follow up to a prior LCS or the patient was lost to follow up. Radiology reports were reviewed and cases were reclassified based solely on wording provided in the report compared to Lung-RADS definitions. Cases of malignancy in the LR0 group were proven by pathology. Radiologist training status was classified as chest trained or other subspecialty trained. Misclassification rates were compared between chest trained and other specialty trained radiologists, with proportions reported with 95% confidence intervals and group comparisons performed using Fisher's exact test.

Results

Seventeen of 119 lung cancer screening examinations (14.3%) initially classified as Lung-RADS 0 met criteria for Lung-RADS 2 on report review. Most misclassifications involved TiB/GG nodules (14/17, 82.4%). No malignant cases occurred in the misclassified group, while the malignancy rate among correctly classified examinations was 7.8%. Misclassification was significantly more frequent among non-specialty trained radiologists compared with cardiothoracic-trained radiologists (27.9% vs 6.6%; relative risk 4.24, 95% CI: 1.63–11.0; $p = 0.002$). Among Tib/GG nodule misclassifications, 35.7% required two follow-up examinations to document resolution.

Conclusion

Misclassification as Lung-RADS 0 was uncommon but occurred significantly more often in examinations interpreted by radiologists trained in a subspecialty other than cardiothoracic imaging and was predominantly associated with TiB/GG nodules in both groups. No malignant cases were missed; however, over one-third of TiB/GG misclassifications required multiple follow-up examinations to document resolution, indicating

increased downstream imaging utilization. This demonstrates the need for further education and possibly updated guidance focused on TiB/GG nodules to reduce misclassification leading to unnecessary follow up imaging.

Translational/Clinical Research

Faculty

POSTER #157

GRN DEFICIENCY LOOSENS STROMAL BARRIERS AND ENHANCES MHC-II SIGNALING YET REDUCES MHC-I DRIVEN CYTOTOXIC IMMUNITY

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Progranulin (GRN) is a multifunctional regulator of tissue remodeling and myeloid biology. Although neuroprotective in the CNS, increasing evidence indicates that GRN supports a fibrotic and immune suppressive tumor microenvironment.

We investigated whether host GRN restrains anti-tumor immunity and whether its inhibition could be therapeutically leveraged. Using C57BL/6J Grn^{-/-} mice across three syngeneic models (KPC pancreatic ductal adenocarcinoma, B16-F10 melanoma, MC38 colorectal carcinoma), GRN loss consistently delayed tumor growth, with the most pronounced effect in KPC subcutaneous tumors, indicating a substantive but incomplete contribution of host GRN to tumor control.

Single-cell RNA-seq and cell-cell interaction analyses of KPC tumors revealed coordinated rewiring of antigen presentation and stromal programs in Grn^{-/-} hosts: MHC-II signaling predominantly B to T was increased, whereas MHC-I inputs decreased. Cancer-associated fibroblasts exhibited downregulation of chemokine/GPCR sensing, ER-phagosome and complement pathways. CAF composition shifted toward iCAF expansion with mCAF/vCAF reduction, and fibroblast-tumor ECM adhesion pairs (COL/FN1/LAM-CD44/SDC) were diminished, consistent with a loosened stromal matrix.

In T cells, CD4⁺/CD8⁺ central or resident memory compartments expanded, while cytotoxic and exhausted CD8⁺ populations declined; pseudotime analyses demonstrated attenuated progression from memory toward cytotoxic or exhausted endpoints.

Collectively, these data indicate that host GRN sustains a myeloid-CAF program that strengthens stromal tension and supports MHC-I driven T-cell terminal differentiation. GRN loss relaxes this circuit, reducing MHC-I availability while enhancing B-T MHC-II engagement, which shifts T cells toward memory states and limits cytotoxic output. These findings identify GRN blockade as a TME reprogramming strategy that should be paired with agents restoring CD8⁺ signal-1 (MHC-I) to convert enlarged memory pools into durable cytotoxic control in PDAC.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #158

CHRONIC PHYSIOLOGICAL OXYGEN TENSION AND DEXAMETHASONE COOPERATIVELY REPROGRAM GLIOBLASTOMA PLASTICITY, STEMNESS, AND THERAPEUTIC RESISTANCE

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Glioblastoma (GBM) is the most aggressive primary brain tumor and remains uniformly fatal despite surgery, radiation, and chemotherapy. Tumor progression is driven by cellular plasticity, invasion, and therapy resistance. One critical yet often overlooked factor influencing tumor biology is oxygen tension within the tumor microenvironment. While physiological oxygen levels in the brain range from ~3-8% O₂ (physioxia), most in vitro studies are conducted under atmospheric oxygen (~21% O₂; normoxia), which may not accurately model tumor cell behavior. In addition, dexamethasone (DEX), a glucocorticoid commonly administered to GBM patients to manage cerebral edema, may influence tumor cell signaling and phenotype.

In this study, we investigated how chronic physioxia and DEX exposure influence GBM cell behavior using established and patient-derived GBM models. Chronic physioxia (5% O₂) significantly increased migratory capacity in GBM001 and GBM10 cells while reducing proliferation and promoting accumulation of cells in the G0/G1 phase, consistent with a migratory “go-or-grow” phenotype. Physioxia also increased phosphorylation of PDGFR β , AKT, and ERK and elevated expression of the EMT regulator SNAI2 (Slug), suggesting activation of invasion-associated signaling pathways. Chemotherapy response to 5-fluorouracil (5-FU) varied by cell line, with GBM10 cells displaying limited physioxia-associated resistance at low drug concentrations. Patient-derived AR022 and AR023 GBM cells similarly demonstrated increased proliferation and migration under physioxia.

We further found that oxygen tension modulates glucocorticoid receptor signaling. DEX induced NOS2 expression under normoxia but increased ALDH1A1 and Slug expression under physioxia, indicating oxygen-dependent transcriptional rewiring. While DEX reduced proliferation, it enhanced migratory capacity in GBM10 cells.

Collectively, these findings demonstrate that physiological oxygen tension reshapes GBM signaling, plasticity, and therapeutic responses while altering the biological effects of dexamethasone. These results highlight the importance of modeling physiologic oxygen conditions to better understand GBM progression and treatment responses.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #159

ROLE OF NRP2 IN PROGRESSION OF ENDOMETRIOSIS TO TYPE I OVARIAN CANCER.

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Role of NRP2 in progression of endometriosis to type I ovarian cancer.

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Background:

Endometriosis affects approximately 10% of reproductive-aged women and confers a markedly increased risk of type I ovarian cancer, particularly in patients with ovarian endometriomas, where risk may rise up to 19-fold. Both endometriosis and type I ovarian cancer frequently harbor ARID1A loss and oncogenic KRAS activation, alterations that converge on pro-tumorigenic pathways such as PI3K/AKT signaling, driving survival, proliferation, angiogenesis, and malignant progression. Despite these shared molecular events, the critical mediators that promote transition from benign endometriosis to ovarian cancer remain poorly defined. Neuropilin-2 (NRP2), a regulator of angiogenesis, lymphangiogenesis, and cell migration, has emerged as a potential driver of tumor progression and may represent a key link in this malignant continuum.

Hypothesis/Objective:

We hypothesize that NRP2 upregulation drives malignant progression from endometriosis to type I ovarian cancer by activating pro-migratory and PI3K/AKT signaling pathways. The objective of this study was to define the expression pattern of NRP2 in endometriosis-associated ovarian cancer progression using cross-species transcriptomic and experimental validation approaches.

Methods:

To investigate molecular alterations during disease progression, we utilized the genetically engineered AKA mouse model (Amhr2^{Cre/+}; Arid1a^{flox/flox}; Kras^{flox-STOP-flox-G12D}), which recapitulates spontaneous development of ovarian endometriomas and progression to carcinoma. RNA sequencing was performed on 6-month AKA ovaries and compared with age-matched controls to identify differentially expressed genes. Cross-species comparison using Venn diagram analysis identified 158 genes consistently upregulated and 163 genes were downregulated in both 6-month AKA mouse ovaries and the human endometrioma dataset (GSE7305). Differential expression and pathway enrichment analyses were performed, with statistical significance defined as adjusted $P \leq 0.05$. Finally, we check NRP2 expressions in human immortalized endometriotic epithelial cells (12Z) Adenomyosis (tAEC21) and type I ovarian cancer cell lines (TOV21G and ES2) by q-PCR.

Results: RNA-sequencing data from human endometrioma samples and the AKA mouse model demonstrated commonly elevated NRP2 expression. Among these shared genes, NRP2 expression was significantly

increased in human endometrioma compared with normal endometrium (log₂ fold change = 1.184; adjusted p = 0.0005) and further elevated in type I ovarian cancer samples relative to endometrioma (p < 0.01). Ingenuity Pathway Analysis (IPA) further revealed that NRP2 expression is markedly increased in ovarian cancer tissues, particularly in ovarian clear cell carcinoma (OCCC). Additionally, IPA analysis of relevant cell line datasets confirmed NRP2 overexpression. We selected several benign and OCCC cell lines in Hawkins' laboratory and experimentally evaluated NRP2 expression, demonstrating that NRP2 is significantly ($n=3$; $P<0.0001$) upregulated in OCCC models.

Conclusion:

Elevated NRP2 expression across endometriosis and type I ovarian cancer supports a mechanistic model in which ARID1A loss and KRAS activation enhance PI3K/AKT signaling, with NRP2 functioning as a downstream effector that promotes angiogenic and invasive processes. These findings identify NRP2 as a potential mediator of malignant transformation in endometriosis-associated ovarian cancer and highlight its promise as a therapeutic target. Future studies will define the functional role of NRP2 using in vitro knock-in and knockdown approaches.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #160

PREDICTING PATIENT CHOICE OF HOSPITAL FOR PANCREATICOUDENECTOMY: GOING BEYOND TRAVEL DISTANCE ALONE

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Background:

Oncologic and surgical outcomes after pancreaticoduodenectomy (PD) are known to be superior when the operation is performed at a high-volume center. There are ongoing policy efforts to regionalize complex surgeries, including PD, as well as determine ideal volume thresholds and quality metrics to stratify centers. Prior studies simulating how regionalization of PD would impact outcomes and access to care assumed that patients will travel to their closest hospital. The validity of this assumption has not been evaluated. The objectives of this study were to (1) assess the accuracy of a distance-only model in predicting where a patient received PD, (2) determine whether a gravity model incorporating hospital volume improved accuracy, and (3) identify characteristics of patients for whom hospital assignment accuracy improved with gravity modeling.

Methods:

Data on patients undergoing PD from 2016 – 2017 in New York or Florida were obtained from the Statewide Inpatient Databases of the Healthcare Cost and Utilization Project (HCUP). Hospitals were assigned a “gravity” coefficient for attracting patients based on volume and distance. Model accuracy was determined by comparing the predicted hospital to the actual hospital where the patient received care.

Results:

Among the 2,949 included patients, distance-alone models were accurate for only 16.3% overall; accuracy significantly improved to 32.9% when hospital volume was added to the model ($p < 0.001$). Gravity models more accurately predicted observed travel burden (average misestimation of 8.3 miles) compared to distance-only models (15.1 miles). Patients who were more accurately classified with the gravity model (vs. distance-only) were more likely to live in major metropolitan areas (76% vs. 57%, $p < 0.001$) and less likely to have Medicaid (6% vs. 12%, $p = 0.002$). Patients who were not accurately classified by any model (vs. those classified accurately by at least one model) were more likely to live in major metropolitan areas (75% vs 70%, $p < 0.001$) or be insured by Medicaid (12% vs 9%, $p < 0.001$) and were less likely to be Non-Hispanic White (61% vs 70%).

Conclusions:

Distance-only models were largely inaccurate for predicting where patients received PD. Model performance varies by geography and patient characteristics, with disadvantaged patients particularly vulnerable to

misclassification. Policy simulation relying on distance-alone models risks underestimating the burden of regionalization. To avoid potentially exacerbating disparities in access to high-quality surgical care, more accurate models that incorporate a broader set of hospital, patient, and market-level factors are needed to better estimate the population-level impact of centralization.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #161

CONTEMPORARY TIME TO UPTAKE OF CLINICAL TRIALS: DEBUNKING THE 17 YEAR TO IMPLEMENTATION PARADIGM

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Introduction

Implementation of new medical evidence into clinical practice is often cited to lag by 17 years based on a landmark analysis that predates modern advancements in scientific publishing, digital dissemination, and clinical data infrastructure. This estimate may no longer reflect current implementation speeds, especially in oncology where trial volume and data access have increased. The objectives of this study were to (1) recalculate the time from landmark trial publication to clinical adoption using real-world oncology registry data, and (2) compare these observed uptake patterns to historic linear models estimating time to implementation.

Methods

A retrospective cohort study was conducted using National Cancer Database (NCDB) data to evaluate uptake of four landmark trials: ACOSOG Z0011 (breast cancer), CONKO-001 (pancreatic cancer), MAGIC (gastric cancer), and DeCOG-STL/MSLT-II (melanoma). Uptake was defined as the annual proportion of eligible patients receiving the trial-endorsed treatment. Two modeling approaches were used to estimate time to 50% uptake: (1) the original linear model published by Balas and Boren in 2000, and (2) a modified model accounting for non-zero baseline use prior to publication, and results of both models were compared to real-world uptake.

Results

A total of 161,963 patients across the four disease sites were included (74,264 breast, 28,169 pancreas, 37,550 gastric, and 21,980 melanoma). Across all trials, the average time to 50% uptake was consistently below 17 years in real world NCDB data and both modeling approaches (**Table 1**). For ACOSOG Z0011, the proportion of patients undergoing axillary lymph node dissection declined from 41.0% in 2012 to 10.9% in 2021. For CONKO-001, adjuvant chemotherapy after pancreatic resection increased from 37.6% in 2004 to 56.6% in 2021, with 50% uptake reached by 2010. For the MAGIC trial, perioperative chemotherapy use increased from 25.5% at publication and reached 54.2% by 2011. For DeCOG-STL/MSLT-II, omission of completion lymph node dissection increased rapidly, with 50% uptake reached within 1 year of publication. The predicted mean time to 50% uptake across trials was 7.8 years (range: 1.8–12.4 years) using the original linear model and 5.9 years (range: 3.4–10.7 years) in a modified model accounting for baseline practice patterns.

Conclusions

Real-world adoption of practice-changing oncologic trials occurs substantially faster than the historically cited 17-year gap. Updated models that incorporate baseline use and nonlinear uptake may better reflect modern implementation dynamics. These findings support more optimistic timelines for evidence integration into cancer care and may influence the speed of development of oncology quality measures.

POSTER #162

TARGETING NQO1 WITH THE BIOACTIVATABLE AGENT IP-DNQ SENSITIZES GEMCITABINE-RESISTANT PANCREATIC CANCER TO GEMCITABINE

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Gemcitabine is a standard chemotherapy for pancreatic ductal adenocarcinoma (PDAC), yet the rapid development of resistance greatly limits its clinical efficacy. NAD(P)H:quinone oxidoreductase 1 (NQO1), an enzyme frequently overexpressed in pancreatic cancer while low expressed in adjacent tissues, provides an opportunity for tumor-selective bioactivatable therapy. Here, we evaluated whether the novel NQO1-bioactivatable compound, IP-DNQ, could overcome gemcitabine resistance. We found that IP-DNQ selectively induced cytotoxicity in gemcitabine-resistant pancreatic cancer cells. Mechanistically, IP-DNQ suppressed the AKT/GSK3 signaling pathway, leading to apoptosis in resistant cells. Notably, the combination of IP-DNQ and gemcitabine produced enhanced antitumor effects compared with either agent alone. This therapeutic synergy was further confirmed in an orthotopic pancreatic cancer mouse model, where the combination treatment significantly inhibited tumor growth. These findings suggest that targeting NQO1 with IP-DNQ represents a promising strategy to overcome gemcitabine resistance in pancreatic cancer.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #163

MULTI-FRUGAL: MULTIMODAL FLEXIBLE REDUNDANCY-AWARE UNIFIED GATED LEARNING FOR HEAD AND NECK CANCER PROGNOSIS

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Introduction: Head and neck cancer is the seventh most common malignancy worldwide associated with poor prognosis. Accurate overall survival (OS) and progression-free survival (PFS) prediction, critical for risk stratification and treatment planning, remains challenging due to patient heterogeneity. Multimodal healthcare data provide information of complementary diagnostic and prognostic value, yet most computational analysis has relied on unimodal analysis partly due to inconsistently missing data across modalities.

Materials and Methods: We leverage the HANCOCK challenge's retrospective cohort of 763 patients, comprising 701 primary tumor whole slide images (WSIs), 396 lymph node WSIs, 368 TMAs, 720 free-text reports, and 763 sets of structured clinical information, with 48% modality-patient pairs missing. We present Multi-FRUGAL, an intermediate-fusion information-budgeted framework with built-in data missingness handling. Modality-specific encoders project heterogeneous inputs into a shared latent space, including Virchow2 for WSI, VGG16 for TMA, Qwen-2 for reports, and multilayer perceptrons for structured data. Embeddings are first processed through an input-conditioned stochastic gating mechanism prioritizing complementary information, while suppressing cross-modal redundancy, and subsequently fused via masked self-attention pooling.

Results: Multi-FRUGAL's performance in test dataset ($AUC_{OS}/AUC_{PFS}=0.833/0.801$) was consistently superior to that of the alternative multimodal approaches ($AUC_{OS}/AUC_{PFS}=0.601/0.672$) and to the best unimodal approach ($AUC_{OS}/AUC_{PFS}=0.82/0.80$, yielded by the structured data).

Conclusions: Structured data, including summarized pathologist assessment, provide the strongest prognostic signals. Computationally-captured subtle WSI cues contribute complementary information in multimodal analyses. Multi-FRUGAL's information-budgeted gated fusion preserves distinct complementarity, while mitigating redundancy and enabling robust multimodal prediction under missing data.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #164

DUAL EPIGENETIC CONSEQUENCES OF DECITABINE IN AML: VIRAL MIMICRY-DRIVEN IMMUNE ACTIVATION AND CONCURRENT ONCOGENIC REPROGRAMMING

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Background: Acute myeloid leukemia (AML) is characterized by widespread epigenetic dysregulation, including aberrant DNA methylation that contributes to transcriptional silencing of tumor suppressor and immune regulatory genes. Decitabine (DCB), a hypomethylating agent, widely used in AML to reverse DNA methylation mediated repression; however, global epigenetic reprogramming may also derepress repetitive genomic elements and oncogenic networks. The extent to which DCB induced hypomethylation promotes tumor immunogenicity versus oncogenic activation remains incompletely defined.

Methods: AML cells were treated with 0.5, 1.5, and 2.5 μ g DCB doses for 72 and 96 hours. Transcriptomic profiling was performed using RNA sequencing with isoform level transposable element analysis, differential gene expression, and gene set enrichment analysis. CpG hypomethylation at repetitive element loci was validated using bisulfite PCR. Cytokine secretion and interferon signaling were assessed by flow cytometry. Selected oncogenic and tumor suppressor genes were validated in primary AML samples using quantitative PCR.

Results: Decitabine induced dose- and time-dependent CpG hypomethylation and preferential derepression of endogenous retroviral elements (ERVs), particularly ERVL, ERVL-MaLR, ERV1, and ERVK families. ERV activation correlated with induction of interferon-stimulated genes and innate immune transcriptional programs. GSEA demonstrated enrichment of TNF α /NF- κ B, IFN- α / β , interferon- γ , cytokine signaling, and antigen-processing pathways. Mechanistically, ERV derepression was associated with cytosolic dsRNA/dsDNA accumulation and activation of RIG-I/MDA5-MAVS and cGAS STING pathways, leading to increased IRF transcription factor expression and secretion of IFN- β , CCL2, and CXCL9/10/11. Decitabine increased IFN- γ production in co-cultured T cells and upregulated cancer-testis antigens (MAGEA1, MAGEB1, MAGEB2, DAZL, HORMAD1) and IFN receptors, supporting enhanced tumor immunogenicity.

Decitabine suppressed proliferative and cell cycle programs, downregulated histone cluster genes, and activated DNA damage and replication-stress pathways, consistent with growth inhibition and apoptosis. Transcriptomic analyses revealed induction of oncogenic and proto oncogenic programs, including MYC/E2F targets, PI3K-AKT, MAPK, mTOR, and TGF- β signaling, as well as regulators such as USP15. Validation in primary AML samples demonstrated partial normalization of epigenetic and differentiation associated genes (CEBPA, DNMT3A, TET2, TP53), alongside persistent dysregulation of adverse prognostic and oncogenic signaling genes, including WT1, ASXL1, PHF6, KIT, FLT3, NRAS, IDH1, and IDH2. Integrated pathway analysis revealed reciprocal modulation between ERV silencing machinery and viral mimicry programs, supporting hypomethylation-induced chromatin destabilization coupled to innate immune activation.

Conclusions: Decitabine induces coordinated epigenetic reprogramming in AML characterized by ERV mediated viral mimicry, robust innate immune activation, enhanced tumor immunogenicity, and suppression

of proliferative signaling. Simultaneously, it activates oncogenic transcriptional networks and germline associated programs. These findings define a dual biological effect of hypomethylating therapy, providing mechanistic insight into both its immunomodulatory potential and its transcriptional risks in AML.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #165

THE FUNCTIONAL CROSSTALK BETWEEN TISSUE TRANSGLUTAMINASE (TG2) AND CD24 ENHANCES CANCER STEM CELL TRAITS AND THERAPY RESISTANCE IN HIGH GRADE SEROUS OVARIAN CANCER

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High-grade serous ovarian cancer (HGSOC) remains one of the most lethal gynecologic malignancies, largely due to persistent cancer stem cells (CSCs) that drive disease progression. Our data revealed that tissue transglutaminase 2 (TG2) is a central mediator of OCSC biology through its interaction with fibronectin (FN) and integrins, activating oncogenic pathways, including Wnt signaling, that promote stemness and platinum resistance. We further identified that TG2 cooperates with the immune-regulatory receptor CD24 to anchor OCSCs to peritoneal organs, facilitating early metastatic implantation and ascites formation.

To therapeutically disrupt this axis, we synthesized a selective peptide inhibitor (BP) that blocks the TG2–FN interaction and developed a CD24-neutralizing antibody (CD24-Ab), each of which suppresses CSC function in preclinical models. We then combined these modalities into a single first-in-class antibody–peptide conjugate, APC (CD24-Ab–TG2-BP), designed to simultaneously disrupt CD24 signaling and TG2-mediated CSC–tumor microenvironment (TME) interactions.

TCGA analysis of HGSOC revealed TG2 and CD24 mRNA amplification in 3% and 2% of cases, respectively. Flow cytometry confirmed robust TG2 (58%, 51%) and CD24 (98%, 100%) expression, with substantial co-expression (55%, 45%) in OVCAR4 and OVCAR5 cells, respectively. APC treatment significantly downregulated CSC-associated markers (NANOG, OCT4, SOX2, ALDH1A1) and disrupted spheroid architecture. Co-immunoprecipitation demonstrated a specific TG2–CD24 interaction in SKOV3 and OVCAR4 cells, while biochemical fractionation localized the complex to lipid raft microdomains, signaling platforms that integrate receptors and downstream oncogenic effectors. In OC xenograft models, treatment with APC significantly reduced tumor volume ($0.22 \pm 0.11 \text{ mm}^3$ vs. $1.7 \pm 1.1 \text{ mm}^3$; N =6, P < 0.001), tumor weight ($0.64 \pm 0.21 \text{ gm}$ vs. $1.53 \pm 0.46 \text{ gm}$; N =6, P < 0.01), and metastasis ($5.3 + 3.6$ vs. $10 + 3.6$; N =6, P < 0.001) when compared with IgG controls, thus demonstrating potent anti-tumor efficacy.

Collectively, these findings identify a lipid raft–restricted TG2-CD24 complex as a novel driver of OCSC maintenance and chemoresistance in HGSOC. Targeting this interaction with APC effectively disrupts CSC–TME crosstalk and suppresses tumor progression, establishing TG2/CD24 axis as a tractable therapeutic vulnerability for overcoming resistance and recurrence in HGSOC.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #166

GLIOBLASTOMA BEYOND THE ENHANCING TUMOR MARGINS: A LONGITUDINAL CASE STUDY COMPARING FET PET WITH MRI-BASED AI INFILTRATION MAPS

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Background: Glioblastoma is the most common malignant brain tumor, with rapid proliferation, diffuse infiltration, and poor prognosis. Imaging plays a central role in its diagnosis, treatment planning, and disease monitoring. Positron Emission Tomography with O-(2-[¹⁸F]fluoroethyl)-L-tyrosine (FET PET) depicts metabolically active tumor irrespective of the blood-brain barrier integrity. Here, we compare FET PET with AI-generated infiltration maps (AI-InfM) derived from multiparametric MRI (mpMRI - T1, T1Gd, T2, T2-FLAIR, DSC) to identify viable tumor beyond the enhancing tumor, potentially corresponding to early recurrence sites.

Methods: We identified a patient with a newly-diagnosed glioblastoma in a non resectable eloquent area. Pre-operative scans were acquired at baseline and two follow-up timepoints, during and post-treatment, at six-month intervals. Each scanning session comprised a 50-minute dynamic FET PET scan and mpMRI performed at Research Center Juelich, Germany. AI-InfM, based on Support Vector Machines, were trained following within-patient, self-normalized measures of heterogeneity across pre-operative mpMRI of treatment-naïve glioblastoma in a subset of the public UPENN-GBM dataset. AI-InfM were then generated for the identified patient and compared with FET PET visually and quantitatively using clinically-established measures, e.g., tumor-to-brainratios (TBR).

Results: Although FET PET showed no increased tracer uptake at baseline, the AI-InfM identified a region with elevated signal. This region exhibited increased uptake of FET in the first follow-up scan (TBR_{max} from 1.9 at baseline to 2.2 at follow-up), supporting the AI-InfM's early prediction. After completion of adjuvant temozolomide therapy, the third scan denoted the predicted high-risk area with reduced tumor activity across FET PET, AI-InfM, and mpMRI.

Conclusions: Our findings indicate potential complementary value of FET PET and mpMRI-derived AI-InfM, for early recurrence identification. This case study underscores the potential to enhance early detection of tumor progression, by virtue of the infiltrative spread in glioblastoma, and warrants continued development and evaluation in a larger multi-institutional cohort.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #167

IMAGING SURROGATE SIGNATURE OF THE GLIOMA EPIGENETIC LIQUID BIOPSY (GELB) SCORE FOR NON-INVASIVE DETECTION OF TUMOR PROGRESSION

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Background: Gliomas are characterized by clinical and molecular heterogeneity and typically harbor a poor prognosis, particularly in high-grade forms. While multi-parametric Magnetic Resonance Imaging (mpMRI) assessment remains the clinical standard for monitoring disease progression, it is subjective and prone to misidentifying treatment-related changes as true progression. The Glioma epigenetic Liquid Biopsy (GeLB) score is a non-invasive blood-based biomarker capable of detecting tumor progression earlier than MRI. Here, we seek an objective radiomics-based surrogate signature of the GeLB score, towards democratizing GeLB's detecting capability without the need for epigenetic analysis.

Methods: We retrospectively analyzed mpMRI data (T1, T1ce, T2, T2-FLAIR) of 65 longitudinal paired GeLB-mpMRI samples (72.31% glioma vs 27.69% non-glioma), from 18 patients. GeLB over 50% defined glioma, and lower values non-glioma. 521 radiomic descriptors were extracted from i) the whole tumor (WT) defined by the abnormal T2-FLAIR envelope, and ii) the peritumoral edematous/infiltrated subregion (WT excluding enhancement and necrosis). A Support Vector Machine classifier was trained to distinguish GeLB-defined glioma/non-glioma, using sequential feature selection to eliminate redundant descriptors over 5-fold cross-validation. To mitigate overfitting and reduce selection bias associated with the limited sample size, performance metrics were averaged across five independent runs.

Results: For edema-based descriptors, our signature yielded mean accuracy=67.69% (± 0.102), sensitivity=80% (± 0.163), and specificity=40% (± 0.255). WT-based descriptors, slightly improved accuracy=73.85% (± 0.078), and sensitivity=91.11% (± 0.130), with comparable specificity=35% (± 0.122).

Conclusions: Our findings support radiomic descriptors derived from clinical routine mpMRI sequences as promising imaging surrogates for the GeLB score, offering a potential non-invasive approach to improved progression detection, while obviating the need for epigenetic analysis. Although the specificity remains modest and the dataset limited in size, this proof-of-concept study highlights the potential of radiogenomics for improved response assessment. Future work will focus on validating these findings in larger, multi-institutional cohorts and leveraging more sophisticated descriptors.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #168

CLINICAL OUTCOMES OF PEMBROLIZUMAB AND ENFORTUMAB VEDOTIN IN VARIANT HISTOLOGY UROTHELIAL CARCINOMA

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Background:

Enfortumab vedotin (EV) in combination with pembrolizumab is an FDA-approved regimen for patients with locally advanced or metastatic urothelial carcinoma. However, its clinical benefit in the setting of variant histology remains uncertain.

Methods:

The Indiana University bladder cancer database was queried for patients diagnosed with variant histology treated with EV plus pembrolizumab between 2023 and 2025. Demographics, treatment line, metastatic sites, and histological variants were collected. We sought to evaluate clinical outcomes in patients with urothelial carcinoma harboring variant histology who received EV and pembrolizumab. Outcomes were stratified by degree of variant histology: $\geq 50\%$ versus $< 50\%$ variant component on pathology. Kaplan-Meier estimates were used to evaluate progression-free survival (PFS) and overall survival (OS). The log rank test was used to compare groups.

Results:

A total of 32 patients were identified. Median age was 72 (range 49–89). EV plus pembrolizumab were administered as first-line therapy in 18 patients, second-line in 9, third-line in 4, and fourth-line in 1. Sites of metastasis included lymph nodes (n=19), bone (n=9), lung (n=4), liver (n=4), and adrenal glands (n=2). Variant percentages were available for 26 patients; 10 demonstrated $\geq 50\%$ variant histology. Common variants included squamous differentiation (n=14), micropapillary (n=6), plasmacytoid (n=3), sarcomatoid (n=1), pure squamous (n=1), and dedifferentiated urothelial carcinoma (n=1). Six patients exhibited multiple variant patterns. Median follow-up was 10.1 months (range 1.6–20.3). Patients with $\geq 50\%$ variant histology had a median PFS of 3.9 months (95% CI, 1.0–5.4), whereas those with $< 50\%$ variant component achieved a median PFS of 8.2 months (95% CI, 4.2–19.6) (p=0.0355). For the entire cohort, median PFS was 5.8 months (95% CI, 4.3–15.5), median OS was 20.1 months (9.9–Not evaluable).

Conclusion:

EV combined with pembrolizumab demonstrated clinical activity in patients with variant histology urothelial carcinoma. Outcomes appeared more favorable in patients with a lower proportion of variant differentiation, suggesting that variant burden may influence response to therapy.

Translational/Clinical Research

Faculty

POSTER #169

PROGRESSION AFTER FIRST-LINE CHEMOTHERAPY WITH NORMAL SERUM TUMOR MARKERS (STM) IN NON-SEMINOMATOUS GERM-CELL TUMORS (NSGCT)

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Background

Surveillance of patients with NSGCT following first-line chemotherapy typically relies on a combination of STM and imaging. While STM are sensitive, a proportion of relapses occur without STM elevation. We aimed to characterize the incidence of progression with normal STM in patients with NSGCT who had initial elevated STM prior to first-line chemotherapy.

Methods

The Indiana University (IU) germ cell tumor database was reviewed for patients with metastatic NSGCT who had elevated tumor markers prior to first-line chemotherapy (AFP >25 ng/mL or β -hCG >5 mIU/mL) and subsequently had disease progression. Patients with radiographic relapse in the absence of STM were classified as “normal STM relapse.” Clinicopathologic variables, sites of relapse, and histology at progression were compared between elevated and normal STM groups at progression using chi-square or Fisher’s exact test, as appropriate.

Results

A total of 725 patients were eligible. Primary tumor site was testis/retroperitoneal in 88.4% and mediastinal in 11.6%. A total of 82 (11.3%) progressed/relapsed with normal STM. Among the 82 patients who progressed with normal STM, sites of progression were lymph nodes in 60 (73.2%), lungs in 44 (53.7%), brain in 11 (13.4%), liver in 6 (7.3%), and bones in 3 (3.7%). Histologic evaluation at relapse revealed similar rates of viable germ cell tumor between patients with normal STM and elevated STM at progression (20.7% vs. 16.5%, $p = .349$). However, teratoma (40.2% vs. 11.8%, $p < .001$) and malignant transformation of teratoma (14.6% vs. 2.6%, $p < .001$) were significantly more common among patients with normal STM relapse.

Conclusions

Among patients with metastatic NSGCT who have elevated STM at diagnosis and progress after front-line chemotherapy, a clinically meaningful subset will have radiographic progression with normal STM distinguished by increased prevalence of teratoma and malignant transformation. These findings underscore the role of routine imaging in addition to STM in post-therapy follow-up of NSGCT.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #170

SURVEILLANCE AFTER COMPLETE RESPONSE (CR) TO FIRST-LINE CHEMOTHERAPY IN NON-SEMINOMATOUS GERM-CELL TUMOR (NSGCT)

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Background

Management of NSGCT following CR after first-line chemotherapy remains variable across institutions. Recent data indicates patients with teratoma and/or yolk sac tumor in the orchiectomy specimen was associated with finding teratoma or viable non-teratomatous GCT at post-chemotherapy retroperitoneal lymph node dissection (PCRPLND) specimen. It is not clear whether this finding impacts long-term survival outcomes. We sought to evaluate long-term outcomes of surveillance after CR at Indiana University (IU) specifically in this patient population.

Methods

We retrospectively reviewed the IU GCT database (January 1990-June 2024). Eligible patients were diagnosed with metastatic testicular/retroperitoneal NSGCT who achieved CR after first-line chemotherapy defined by no residual mass >1cm in longitudinal axis. Patients who were treated with chemotherapy outside IU were excluded to reduce referral bias. Patients were stratified according to orchiectomy specimen into two groups: Those with teratoma and/or YST and those without either component. Primary outcomes were to estimate progression-free survival (PFS) and overall survival (OS) in these subgroups using Kaplan-Meier methodology.

Results

A total of 261 patients met the inclusion criteria. Of these, 21 (8.0%) had teratoma, 50 had YST (19.2%), and 65 had both (24.9%). IGCCCG risk was good in 85.1%, intermediate in 6.1%, and poor in 8.8%. Median follow-up was 4.97 years (range, 0.01-28.80). Pre-chemo retroperitoneal LN size was <1 cm in 19 (7.3%), 1-3cm in 102 (39.1%), >3cm in 67 (25.7%), and unknown in 73 (28.0%). Four-year PFS for patients with teratoma or YST in orchiectomy specimen was 92.2% vs 95.3% in pts without teratoma/YST (P=.37). Four-year OS for patients with teratoma/YST in orchiectomy specimen was 98.0% vs 98.1% in pts without teratoma/YST (P=.59). Late relapse occurred in 3.7% (n=5) of those with teratoma/YST and 2.4% (n=3) for those with neither (P=.72). Of the 16 patients who relapsed, 8 underwent delayed RPLND, 3 received salvage chemo, 2 were treated with salvage chemo and RPLND, and 3 underwent resection of other metastatic sites. At last follow-up, 12 patients who relapsed were without evidence of disease, 2 were alive with disease, and 2 died of NSGCT.

Conclusions

In this large cohort, most patients with NSGCT who achieved CR after first-line chemotherapy remained disease-free on surveillance, regardless of the presence of teratoma or YST in the orchiectomy specimen. Relapses were uncommon and most patients who relapsed were salvaged successfully with surgery and/or chemotherapy.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #171

IMPROVING RECOGNITION AND MANAGEMENT OF HEPARIN RESISTANCE: A TWO-PHASE AGILE QUALITY IMPROVEMENT STUDY USING RETROSPECTIVE EPIDEMIOLOGY AND EPIC-BASED CLINICAL DECISION SUPPORT

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Background: Unfractionated heparin (UFH) is a standard anticoagulant, yet many patients develop heparin resistance (HR), where escalating doses fail to reach therapeutic levels. Currently, Eskenazi Health relies on paper-based protocols and inconsistent clinician judgment to identify HR, leading to delayed management and increased risks of thrombosis or bleeding.

Objective: To improve the timely recognition and standardized evaluation of suspected heparin resistance among adult inpatients receiving therapeutic UFH at Eskenazi Health.

Aim: By June 30, 2027, increase the proportion of adult inpatients meeting HR criteria who receive a complete diagnostic evaluation from an estimated baseline of <10% to $\geq 90\%$, while maintaining stable rates of major bleeding.

Methods:

Phase 1 will perform a five-year retrospective analysis of adult inpatients treated with therapeutic UFH using Epic data extracted from the Epic Data Warehouse. The analysis will quantify the institutional burden of HR, compare operational definitions of HR (including fixed-dose and weight-based criteria), evaluate patterns of diagnostic evaluation and specialty consultation, and examine associations between HR definitions and clinical outcomes.

Phase 2 will develop and deploy an Epic-based clinical decision support (CDS) pathway embedded within the UFH workflow. A best practice advisory integrated with the Epic Willow heparin titration tool will identify patients meeting predefined criteria for suspected HR and prompt standardized evaluation, including anti-factor Xa testing, antithrombin activity measurement, and hematology consultation. The CDS pathway will facilitate ordering and consultation while preserving clinician discretion. Implementation will occur iteratively with stakeholder input from hematology, pharmacy, nursing, and Epic analysts.

Preliminary feasibility analysis identified ~500 patients meeting weight-based, cumulative, or combined definitions of heparin resistance over the past five years, confirming adequate case volume for the project.

Measures: Primary process measures include the proportion of patients meeting HR criteria who receive anti-Xa testing, antithrombin activity measurement, and hematology consultation, as well as time from HR identification to the first diagnostic or management action. Outcome measures include time to therapeutic

anticoagulation, new or progressive thrombosis within 30 days, and hospital or ICU length of stay. Balancing measures include International Society on Thrombosis and Haemostasis (ISTH)-defined major bleeding and CDS alert burden.

Conclusion: Embedding evidence-informed CDS within the UFH workflow may improve recognition, diagnostic evaluation, and management of heparin resistance while maintaining clinician autonomy and patient safety. This approach may provide a scalable framework for anticoagulation stewardship across Epic-based health systems.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #172

DEFINING RESISTANCE: CLINICAL OUTCOMES BASED ON VARYING DEFINITIONS OF HEPARIN RESISTANCE

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Introduction: Heparin resistance is the need for higher doses of unfractionated heparin for therapeutic anticoagulation and lacks a universal definition. Common criteria include a daily dose >35,000 units or a weight-based dose >25 units/kg/hr. While anti-factor Xa assays are more accurate, many institutions still rely on PTT, potentially leading to misdiagnosis. We conducted a retrospective study to assess clinical outcomes associated with various definitions of heparin resistance.

Methods: We reviewed patients receiving heparin drips at our institution (2017–2025). Patients were classified into three groups:

- **Definition 1:** >35,000 units/day
- **Definition 2:** >25 units/kg/hr (excluding loading dose)
- **Definition 3:** Met both criteria

Data on demographics, survival (30-day and 1-year), length of stay (LOS), and bleeding complications were analyzed using Cox proportional hazards models and Wilcoxon rank-sum tests in R (v4.5.0).

Results: Among 5,414 patients, 5,041 met Definition 1, 60 met Definition 2, and 313 met both. The 30-day mortality was highest in Definition 2 (24.6%) compared to Definition 1 (13.4%) and Definition 3 (21.4%) ($p < 0.001$). One-year mortality followed a similar trend, peaking at 47.2% for Definition 2 ($p < 0.001$). While Definition 3 had a significantly longer median LOS (15 days), Definition 1 was associated with the highest rate of major bleeding (17% vs. 7.8% in Definition 2; $p = 0.002$).

Hazard ratios indicated that patients meeting Definition 2 alone had an 85% increased risk of mortality (HR 1.85; 95% CI: 1.34–2.56; $p < 0.001$), while those meeting both definitions had a 38% increased risk (HR 1.38; 95% CI: 1.17–1.63; $p < 0.001$).

Conclusion: Weight-adjusted definitions of heparin resistance (Definition 2) are associated with significantly worse long-term survival and provide greater prognostic utility than fixed-dose thresholds. Standardizing these definitions is essential for early identification and intervention to improve clinical outcomes.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #173

AN EVALUATION OF OUT-OF-POCKET COSTS FOR LONG COURSE CHEMORADIATION AND SHORT COURSE RADIATION THERAPY FOR RECTAL CANCER

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Introduction: Neoadjuvant short course radiation and long course chemoradiation are standard treatments for locally advanced rectal cancer. Regimens balance local control rates, pathologic complete response rates, toxicity, and patient preference. In patients eligible for both, cost may be a neglected consideration. We aimed to (1) compare out-of-pocket costs (OOPC) between short course radiation and long course chemoradiation; and (2) determine if specific insurance plan types were associated with regimen selection.

Methods: Locally advanced rectal cancer patients were identified from Merative MarketScan (2016-2023). Encounters and billing codes were used to categorize neoadjuvant regimens into long course chemoradiation (≥ 25 episodes), short course radiation (≤ 5), or other (6-24). OOPC were the sum of patient deductibles, copayments, and coinsurance for radiation and chemosensitizers. Patient and insurance plan characteristics were compared. OOPC were estimated with a two-part model: logistic regression for any OOPC, then a log-link gamma model for positive OOPC; estimates adjusted for patient factors. Plans were categorized as: high deductible (HD), preferred provider organization (PPO), comprehensive, exclusive provider/health maintenance organization (EPO/HMO), point of service (PS). A separate logistic regression evaluated the association between plan type and receipt of long course therapy.

Results: Between 2016-2023, 7,542 patients were identified with rectal cancer: 3,426 (45%) underwent radiation therapy with 2,285 (67%) completing long course chemoradiation, 861 (25%) short course radiation, and 280 (8%) other. After adjustment, the estimated mean OOPC were \$1,136 for long course (95% CI \$1,070-\$1,212) and \$896 for short course (95% CI \$774-\$1,021). When compared to HD insurance plans, patients with comprehensive plans were more likely to undergo long course chemoradiation (RR 1.17, 95% CI 1.06-1.29, $p=0.001$). Patients were less likely to undergo long course chemoradiation if they had a PPO plan (RR 0.88, 95% CI 0.83-0.93, $p<0.001$) or an EPO/HMO (RR 0.9, 95% CI 0.83-0.98, $p=0.017$) plan. No difference was found for PS plans.

Conclusion: Long course chemoradiation had higher costs (26% more) for patients when compared to short course radiation. Radiation course type was significantly associated with patient insurance type, with comprehensive plans more likely to undergo long course therapy. In patients who are candidates for both neoadjuvant regimens, costs may be an additional consideration guiding treatment selection.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #174

WHY A PATIENT-POWERED REGISTRY IS IMPORTANT FOR PEOPLE WITH RARE CONDITIONS: “WE NEED AS MUCH RESEARCH AS WE CAN GET!”

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Background: Rotationplasty (RP) is a surgical option for bone tumors involving the knee. In this procedure, the ankle joint is used to create a knee joint and the foot serves as support for the prosthesis. The result is a backwards facing foot on a shortened leg. Benefits to RP include no activity limitations and fewer subsequent surgeries than other options. However, there are barriers to RP. There is no central repository for RP data; this limits the ability of patients/caregivers, clinicians, and researchers to access data on this rare but increasingly requested surgery. Additionally, there are issues with insurance authorization for prosthetics because there is no billing code for RP. Limited research affects clinical bias, which limits the inclusion of RP as an option in shared decision-making.

Patient-powered registries serve as a database of health information, managed by patients rather than institutions. Both qualitative and quantitative patient-provided data may be collected within a registry, with the purpose of advancing patient-specific needs and research priorities. We aimed to create the Rotationplasty Patient-Powered Registry to provide a cumulative repository of patient-reported data.

Methods: Parents of children and adults who have or had RP were invited to complete quantitative assessments including age-appropriate versions of the Patient-Reported Outcomes Measurement Information System (PROMIS), the Toronto Extremity Salvage Score (TESS-LE) for the lower extremity, and the Decision Regret Scale to measure limb function and quality of life. Patients were also asked to answer qualitative questions on appearance, function, prosthetics, and rehabilitation.

Results: Following its release in January 2026, 8 participants between 22-53 years completed the RP registry. Participants were 8 months-40 years post-op. Reasons for RP include osteosarcoma, congenital limb difference, Ewing sarcoma, and synovial sarcoma. Two participants with osteosarcoma had a failed limb salvage surgery and later chose RP. While most reported high levels of satisfaction and limb function with RP, less satisfaction and function were noted at 8 months post-op. Participants also noted the importance of self-advocating for RP and the need for a patient-powered registry to inform clinicians and people considering RP. Vignettes for each participant were created using ChatGPT EDU 5.3 and revised by qualitative analysts to provide succinct patient stories to aid future researchers in understanding the patient experience in their own words.

Conclusions: This patient-powered registry provides a voice to a rare and understudied population. Understanding life after surgery in participants' own words may aid future patients and families in information gathering on RP as a surgical option. The creation of this registry can help clinicians engage in shared decision-making and improve access to data for research. Finally, we hope that this registry will provide the evidence needed for a standardized RP billing code.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #175

PROFUSEME: PROSTATE CANCER BIOCHEMICAL RECURRENCE PREDICTION VIA FUSED MULTI-MODAL EMBEDDINGS

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Almost 30% of prostate cancer (PCa) patients undergoing radical prostatectomy (RP) experience biochemical recurrence (BCR), characterized by increased prostate-specific antigen (PSA) and associated with increased mortality. Accurate early prediction of BCR at the time of RP would support prompt, adaptive clinical decision-making and improved patient outcomes. In this work, we propose prostate cancer BCR prediction via fused multi-modal embeddings (PROFUSEme), which learns cross-modal interactions of clinical, radiology, and pathology data, following an intermediate fusion configuration in combination with Cox Proportional Hazard regressors. Quantitative evaluation of our proposed approach reveals superior performance when compared with late fusion configurations, yielding a mean C-index of 0.861 (std=0.112) on the internal 5-fold nested cross-validation framework, a C-index of 0.7107 and 0.7197 on the hold-out data of the CHIMERA 2025 challenge validation leaderboard and test leaderboard, and top-ranked as the best multi-modal method.

Translational/Clinical Research

Post-Doctoral/Medical Fellow

POSTER #176

DIFFERENTIAL SENSITIVITY OF PAIRED FUS-TFCP2+ RMS PDX MODELS DEVELOPED FROM TUMOR SPECIMENS OBTAINED PRIOR TO AND AFTER ALK INHIBITOR THERAPY

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FUS-TFCP2-positive rhabdomyosarcoma (FUS-TFCP2+ RMS) is a rare subtype of spindle cell/sclerosing RMS with a craniofacial predilection. These tumors are highly aggressive, quick to metastasize and evade conventional chemotherapies. FUS-TFCP2 fusion results in a gain of function transcription factor that promotes proliferation and activates survival pathways while blocking myogenic differentiation and inhibiting DNA repair. Currently, there is no effective standard of care, surgery can be difficult due to location and patients typically succumb to the disease within 15 months of diagnosis. Overexpression of downstream target, Anaplastic Lymphoma Kinase (ALK), is characteristic of FUS-TFCP2+ RMS. However, while case reports of targeted treatment with ALK inhibitors (ALKi) have shown modest results, resistance always emerges.

In collaboration with the Pediatric Cancer Precision Genomics Program at the Riley Hospital for Children, we developed a paired set of FUS-TFCP2+ RMS patient-derived xenografts (PDXs). ALKi-sensitive PDX174 was derived from a patient tumor sample obtained prior to an 11-month ALKi (lorlatinib) regimen. Subsequently, a second sample was acquired following disease progression from which ALKi-resistant PDX199 was established. RT-PCR validated the fusion site in both models. In vivo studies confirmed PDX174 sensitivity and PDX199 resistance to lorlatinib (0.1mg/kg and 1mg/kg). Concordant with FUS-TFCP2+ characterization, transcriptome analysis showed significantly increased expression of ALK in both PDX174 (14.9-fold) and PDX199 (11.1-fold). Western blot analysis revealed robust overexpression of ALK isoforms in PDX174, whereas ALK expression in the ALKi-resistant PDX199 was only barely detectable. Instead, PDX199 exhibited increased levels of TERT, CDK4/6, and BET proteins.

To further compare the models, we utilized two complementary approaches to evaluate the activated kinome of ALKi-resistant PDX199 compared to ALKi-sensitive PDX174. Kinase activity profiling using PamGene peptide microarrays revealed a statistically significant increase in kinase activity of components involved in

PI3K/AKT pathway and cell cycle CDKs (CDK1,2,4), with concomitant suppression of kinase activity of JNK/p38 MAPKs, indicating a shift toward PI3K/mTOR-driven pro-survival signaling and potential vulnerability to PI3K/AKT and CDK inhibition. Global kinome analysis using multiplexed inhibitor beads also showed an increase in CDK4/6 activation in ALKi-resistant PDX199 versus ALKi-sensitive PDX174.

Pre-clinical models such as these provide a platform to connect molecular signatures with targeted therapy, increase our mechanistic understanding of tumor adaptive responses and design therapies that will mitigate the emergence of therapeutic resistance.

Translational/Clinical Research Research Technician

POSTER #177

RAPID 'WARM' AUTOPSY CLINICAL STUDY FOR PATIENTS WITH SMALL CELL LUNG CANCER (SCLC)

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Small cell lung cancer (SCLC) is a highly aggressive malignancy that accounts for an estimated 270,000 deaths worldwide each year. Prognosis remains poor, with a 5-year survival rate of 10%. Although initial response rates to first-line platinum-based chemotherapy are high, relapse due to platinum resistance is nearly universal. Progress in SCLC research has been hindered by the lack of clinically relevant *in vitro* and *in vivo* models due to the limited availability of patient-derived tissue samples. Rapid autopsy programs offer a promising approach to overcome this limitation by enabling the collection of high-quality tumor tissue shortly after death. This pilot study assesses the feasibility of implementing a rapid autopsy protocol at the Indiana University Simon Comprehensive Cancer Center (IUSCCC) to obtain viable tumor tissue and blood samples from SCLC patients for basic and translational research. These samples will be used to characterize SCLC tumor biology and genetic alterations, investigate mechanisms driving tumor growth and metastasis, and identify pathways underlying resistance to conventional chemotherapy. Patients diagnosed with SCLC may enroll in the study at any point during their disease course. The patient cohort comprises male and female veterans recruited from oncology outpatient clinics at the IUSCCC, Eskenazi Health, and the Richard L. Roudebush VA Medical Center. This cohort was selected due to the high incidence of SCLC in this population and their historical underrepresentation in oncology trials. Since 2023, 17 patients have been enrolled, and recruitment remains ongoing. Following death, participants are transported to the designated facility, where a rapid autopsy and tissue collection are performed within 10 hours. Tumor samples from lungs, lymph nodes and liver, are collected and distributed for analysis. Tumor and blood specimens undergo protein and gene expression profiling, RNA sequencing, and whole-exome sequencing. The histological review of current samples collected from 12 patients confirms SCLC in primary and metastatic sites, with >50% of tumor tissue in most lung and liver samples. Overall, this study aims to advance understanding of the molecular and biological mechanisms underlying SCLC, while establishing infrastructure at the IUSCCC for clinically relevant rapid warm autopsy models that may facilitate the development of more effective therapies.

Translational/Clinical Research

Research Technician

POSTER #178

EFFECTS OF THE NRF2 ACTIVATOR BARDOXOLONE METHYL (CDDO-ME) ON BRCA1-MUTANT BREAST CANCER PROGRESSION

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Breast cancer is one of the most prevalent malignancies worldwide and a leading cause of cancer-related morbidity and mortality among women. Mutations in the breast cancer-associated gene 1 (BRCA1) are among the most common genetic alterations identified in familial breast cancer. BRCA1 functions as a critical tumor suppressor and plays essential roles in DNA damage response and repair, cell-cycle regulation, and apoptosis. Although poly (ADP-ribose) polymerase (PARP) inhibitors have improved outcomes in BRCA1-deficient tumors, resistance remains common, highlighting the need for alternative or complementary therapeutic strategies. In this context, targeting cellular redox regulation through modulation of the Kelch-like ECH-associated protein 1–nuclear factor erythroid 2-related factor 2 (KEAP1–NRF2) pathway represents a promising approach.

2-Cyano-3,12-dioxooleana-1,9(11)-dien-28-oic acid methyl ester (CDDO-Me or bardoxolone methyl) is a synthetic triterpenoid derivative of the naturally occurring oleanolic acid. CDDO-Me modifies cysteine residues in KEAP1, leading to the release of NRF2 and consequent translocation to the nucleolus, where it activates several downstream effectors. CDDO-Me exerts complex, context-dependent effects in cancer cells. Besides modulating redox signaling, it inhibits pro-survival pathways such as nuclear factor- κ B (NF- κ B) and signal transducer and activator of transcription 3 (STAT3), increases intracellular reactive oxygen species (ROS) under certain conditions, and promotes apoptosis via mitochondrial dysfunction.

We hypothesize that CDDO-Me induces redox reprogramming and influences inflammatory signaling, thereby impairing tumor cell proliferation and increasing susceptibility to DNA damage. To explore this, we evaluated CDDO-Me in both in vitro and in vivo models of BRCA1-deficiency. In vitro, 72 hours of treatment of breast BRCA1-mutated human and mouse cell lines with CDDO-Me reduced cell viability (IC₅₀<1 μ M). In addition, CDDO-Me (300 nM) increased the expression of cell cycle arrest markers (p27 and p21), decreased NF- κ B, and increased expression of the inflammatory markers p-STING and IRF3. In vivo, a BRCA1-mutated murine model of breast cancer (BRCA1^{Co/Co};p53^{+/-};MMTV-Cre) was fed a powdered control diet starting at 15 weeks. Once tumors were detected and reached $\sim 32\text{mm}^3$ mice were randomized to a control diet or a diet containing CDDO-Me (80 mg/kg diet - 20 mg/kg body weight). CDDO-Me delays tumor progression, with treated mice surviving an average of 40 days post-tumor detection (vs. 32 days for vehicle-treated mice).

Overall, these results highlight the potential of CDDO-Me as a therapeutic strategy in BRCA1-mutated breast cancer and provide a rationale for future combination studies with PARP inhibitors.

Translational/Clinical Research

Research Technician

POSTER #179

BET INHIBITION POTENTIATES SALVAGE CHEMOTHERAPY RESPONSE THROUGH TXNIP UPREGULATION IN OSTEOSARCOMA

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Osteosarcoma (OS) is an aggressive bone cancer in pediatric adolescent and young adult patients. Survival rate for metastatic and relapsed OS patients remains dismal at < 30%. Additionally, no effective standardized salvage therapy currently exists for these patients, in part due to genomic complexities arising from moderate levels of replication stress (RS). Bromodomain and extra-terminal domain protein inhibitors (BETi) are an underexplored option to target RS, as BETi creates an imbalance between transcription-replication kinetics thereby exacerbating oncogenic RS and cell death. BETs (BRD2,3,4) are epigenetic readers that play a role in regulating gene expression networks as well as DNA replication and repair. We hypothesized that BET inhibition with AZD5153 would suppress OS growth by inducing tumor suppressive mechanisms that exacerbate DNA damage and sensitize tumors to DNA-damaging chemotherapeutic agents.

AZD5153 monotherapy significantly suppressed OS tumor growth compared to vehicle ($p < 0.05$) in female and male patient-derived xenografts (PDXs) established from both treatment-naïve (PDX96, PDX115, PDX112) and metastatic OS (PDX77-TT2). Mechanistic evaluation showed increased γ -H2AX and p-RPA2 S8 following AZD5153 exposure in PDX96, indicative of enhanced RS. RNA-seq and protein analyses revealed dysregulation of DNA damage response genes, including upregulation of TXNIP, a tumor suppressor that promotes DNA damage and apoptosis, and downregulation of PDGFRA. Similar TXNIP upregulation and PDGFRA suppression were observed in AZD5153-treated PDX115. Increased TXNIP expression was also evident in vitro when BET/BRD4 activity was inhibited in OS cell lines by siRNA, PROTAC-mediated degradation, or disruption of BRD4-histone interactions via AZD5153. These effects were accompanied by elevated c-PARP levels, a marker of cell death. Ongoing studies suggest BRD4 inhibition elevates TXNIP via AKT pathway suppression, as indicated by reduced phospho-AKT in AZD5153-treated tumors.

In vitro combination studies showed additive-to-synergistic effects between AZD5153 and a variety of salvage agents (etoposide, SN38, and topotecan). In particular, AZD5153 in combination with topotecan resulted in enhanced apoptosis, increased TXNIP, decreased AKT, and elevated RS markers (γ -H2AX, comet assays). In vivo, dose finding studies indicated that PDX77-TT2, was resistant to salvage agents ifosfamide and SN38, but moderately sensitive to topotecan. Furthermore, AZD5153 alone or in combination with topotecan improved survival compared to single agents ($p < 0.05$) and was well tolerated.

Collectively, these data support BET inhibition alone or with salvage therapy as a promising therapeutic approach for aggressive OS, potentially mediated through TXNIP upregulation and AKT pathway suppression.

Translational/Clinical Research

Research Technician

POSTER #180

POSITIVE ASSOCIATION BETWEEN PERIPROSTATIC ADIPOSE TISSUE VOLUME AND PROSTATE VOLUME ON PREOPERATIVE MAGNETIC RESONANCE IMAGING.

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Introduction

Obesity is a risk factor for multiple malignancies, including prostate cancer, where it contributes to tumor invasion, metastasis, and therapy resistance. A key pathological feature of prostate cancer invasion is extraprostatic extension (EPE), characterized by tumor cell infiltration beyond the prostatic capsule into the surrounding tissue including periprostatic adipose tissue. However, the mechanisms by which adipose tissue influences prostate tumor invasion are understudied. This knowledge gap is being enforced by the historical reliance on body mass index (BMI) as a proxy for obesity in previous studies, which fails to capture the distinct metabolic profiles of different adipose depots, including periprostatic, visceral (VAT), and subcutaneous adipose tissue (SAT). Unlike SAT, VAT is metabolically active and secretes higher levels of inflammatory mediators and growth factors that promote tumor progression. VAT accumulation has been associated with prostate cancer aggressiveness, particularly in Black men.

Objective of the study

The objective of this study is to characterize and compare the metabolic landscapes of VAT and SAT in prostate cancer patients with high (BMI >35) and low (BMI <25) body mass indices.

Methods

Twenty patients undergoing radical prostatectomy were prospectively enrolled in this cohort study. Comprehensive clinical parameters were collected, including date of birth, age at surgery, body weight, preoperative prostate-specific antigen (PSA) levels, clinical stage, biopsy Gleason Grade (GG), pathological Gleason Grade, pathological stage, and surgical margin status. Preoperative magnetic resonance imaging was performed to quantify periprostatic fat volume and thickness. Intraoperatively, periprostatic adipose tissue samples were obtained from multiple visceral anatomical sites. Regional lymph nodes were harvested and will be examined for metastatic disease.

Results

Preliminary correlative analyses demonstrated a positive association between periprostatic adipose tissue volume and prostate volume on preoperative magnetic resonance imaging.

Conclusion

These observations underscore the need for comprehensive metabolic and cellular characterization of adipose depots to elucidate their role in prostate cancer progression and extraprostatic extension.

Future Directions

First, we will be increasing our sample size to determine association between periprostatic adipose tissue volume and pathological stage T3 using Logistic Regression. Second, collected adipose tissue samples will undergo comprehensive multi-omic profiling to define depot-specific metabolic and cellular landscapes. In parallel, single-cell and single-nucleus

RNA sequencing will be conducted to characterize cellular heterogeneity within adipose depots and to investigate cellular plasticity and population dynamics in the tumor microenvironment. These integrated approaches will provide mechanistic insights into how adipose tissue metabolism and cellular composition influence prostate cancer invasion and may identify novel therapeutic targets for obesity-associated aggressive disease.

Translational/Clinical Research Research Technician

POSTER #181

TUMOR SELECTIVE RETROVIRAL REPLICATING VECTORS FOR THE TREATMENT OF LEPTOMENINGEAL MEDULLOBLASTOMA

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Medulloblastoma is the most common malignant brain tumor in children, with Group 3 medulloblastomas, characterized by high Myc expression, bearing the poorest prognosis. These tumors often present with leptomeningeal dissemination and are associated with high recurrence rates and limited survival following relapse. Current treatment approaches, including surgery, chemotherapy, and craniospinal irradiation, result in significant long-term morbidity, underscoring the urgent need for novel therapies that improve outcomes while minimizing toxicity. Retroviral replicating vectors (RRVs) offer a promising, tumor-selective gene therapy strategy. Toca 511 (now DB107) is an RRV encoding a yeast cytosine deaminase (CD) gene that converts the antifungal prodrug 5-fluorocytosine (5-FC) into the chemotherapeutic agent 5-fluorouracil (5-FU) within infected tumor cells. Despite mixed results in Phase 3 trials for glioma, DB107 demonstrated survival benefits in specific subgroups, warranting further investigation in other CNS malignancies. This study aims to evaluate the efficacy and biodistribution of RRV-CD therapy in leptomeningeal medulloblastoma. Initially I am investigating RRV distribution following intrathecal delivery in immunodeficient murine models. RRV spread will be assessed via histology, flow cytometry, and qPCR. Preliminary results have shown viral spread in both the brain and spine of mice using flow cytometry. Further studies will include increasing the amount of viral spread for suicide gene therapy activation. This study will provide critical data on optimizing RRV-CD therapy for disseminated medulloblastoma, including assessing vector spread and efficacy. The goal is to translate this gene therapy approach into clinical trials for children with high-risk leptomeningeal medulloblastoma.

Translational/Clinical Research

Research Technician

POSTER #182

NON-VIRAL CRISPR–LNP NANOFORMULATIONS ENABLE EFFICIENT GENERATION OF CD19 AND BCMA CAR T CELLS

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CAR T-cell therapies have reshaped the treatment landscape for hematologic malignancies, particularly B-cell leukemias/lymphomas and multiple myeloma, by enabling potent, antigen-directed cytotoxicity and deep clinical responses in otherwise refractory disease. Despite these advances, major barriers continue to limit broader access and constrain next-generation product design. Most clinical manufacturing workflows rely on viral vectors, which increase cost and logistical complexity, introduce variability in vector copy number and transgene expression, and can hinder standardization across manufacturing sites. Integrating viral approaches also carry a safety consideration related to insertional mutagenesis, in which semi-random genomic integration can dysregulate nearby genes and, in rare settings, promote clonal outgrowth/leukemogenesis. In parallel, relapse remains a significant clinical challenge, often driven by antigen downregulation or lineage switching (e.g., CD19-negative escape in B-ALL) and by multi-antigen heterogeneity and variable antigen density in diseases such as myeloma. Together, these limitations motivate scalable, non-viral engineering strategies and more flexible CAR designs, including dual-target approaches that can maintain activity when a single antigen is lost and reduce selective pressure on any single epitope.

To address both manufacturing and relapse/escape challenges, we developed a scalable, non-viral platform using lipid nanoparticle (LNP)–delivered CRISPR nanoformulations to engineer primary human T cells with a single-target CD19 CAR, a single-target BCMA CAR, or a bicistronic CD19/BCMA CAR cassette. This modular approach is intended to support standardized production while enabling rapid iteration from single-antigen to dual-antigen CAR programs aimed at mitigating antigen escape. Primary human T cells were activated and edited using optimized CRISPR–LNP formulations designed to install CAR constructs via targeted genome engineering. Engineered products were assessed for immediate post-editing viability and subsequent expansion, surface CAR expression, and immunophenotype, including memory-associated features and activation/exhaustion marker profiles relevant to persistence and function. Functional potency was evaluated using antigen-specific activation and cytotoxicity assays against CD19⁺ and/or BCMA⁺ tumor targets, with antigen-negative controls to assess specificity. For the bicistronic program, dual-antigen activity was interrogated across single-positive and dual-positive target conditions to quantify breadth of response and to model clinically relevant heterogeneity.

Across multiple donors, CRISPR–LNP treatment reproducibly generated CAR T-cell products for each configuration while preserving robust post-engineering viability and expansion. Edited cells demonstrated antigen-dependent activation and cytotoxicity against CD19⁺ and/or BCMA⁺ targets, and bicistronic CD19/BCMA CAR T cells retained functional activity across relevant antigen contexts, supporting a strategy to mitigate antigen escape. Collectively, these findings establish CRISPR–LNP nanoformulations as a flexible, target-agnostic approach for producing single-input and dual-input CAR T cells, with potential to enable more standardized manufacturing and accelerate translation of next-generation CAR designs that require precise, non-viral genome engineering.

Translational/Clinical Research

Research Technician

POSTER #183

THE MACROPHAGE 27-HYDROXYCHOLESTEROL MEDIATES PROSTATE CANCER METABOLIC REWIRING.

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Introduction

The liver X receptors- α/β (LXR α/β) nuclear receptors regulate genes involved in cholesterol and lipid metabolism via sensing the levels of cholesterol metabolites, oxysterols. The most abundant oxysterol: 27-hydroxycholesterol (27HC), is synthesized by an enzymatic reaction involving the cytochrome P450 (CYP) enzyme CYP27A1 and is catabolized by CYP7B1 enzyme. Unlike the other main oxysterols: 5,6-epoxy cholesterol (5,6-EC) and 25-hydroxycholesterol (25HC), which can be produced by cholesterol autoxidation (no enzyme needed), 27HC is only produced by an enzymatic transformation.

Methods

We explored whether macrophages could be a source of LXR endogenous modulators in prostate cancer by mining cBioPortal. We then performed a series of multiomics experiments to examine the impact of 27HC on tumor cell metabolism, gene expression and DNA methylation.

Results

In TCGA, we first compared the expression level of CYP27A1 in tumor and macrophage and found that CYP27A1 is highly expressed in macrophages compared to tumor cells. Isotope labeling by adding cholesterol-[2,3,4-¹³C₃] to tumor (T) and macrophage (M) showed selective conversion of cholesterol-[2,3,4-¹³C₃] to 27HC-[2,3,4-¹³C₃] in macrophages. To examine the biological function of 27HC, we found that tumor/inflammation relevant concentration of 27HC (1 μ M) suppressed the expression of *Acaca*, *Hmgcr* and *Abcg1* in tumor cells. In addition, elevated 27-HC promoted aberrant hypermethylation, in part by amplifying flux through the one-carbon metabolic cycle.

Conclusion

These findings position the cholesterol -27HC axis as a novel upstream regulator of the cancer methylome and highlight methionine metabolism as a critical node linking lipid signaling to epigenetic reprogramming. Therapeutic strategies targeting cholesterol biosynthesis, 27-HC production via CYP27A1, or methionine cycle enzymes may therefore offer promising avenues for reversing pathological methylation states and restoring tumor suppressor function in cholesterol-rich tumor contexts.

Translational/Clinical Research

Research Technician

POSTER #184

CAF-DRIVEN INFLAMMATORY REPROGRAMMING OF CD34 HSPCs PROMOTES CARCINOGENIC STATES

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Background: Cancer-associated fibroblasts (CAFs) are key regulators of the tumor microenvironment, yet their direct impact on normal HSPCs remains poorly understood. In AML, stromal remodeling accompanies disease progression, but whether AML-derived CAFs can actively reprogram normal CD34 HSPCs into tumor-supportive states remains unclear. We hypothesized that AML bone marrow derived CAFs are sufficient to induce inflammatory, metabolic, and oncogenic reprogramming in normal CD34 cells, distinct from healthy fibroblasts.

Methods: Normal human CD34 cells were co-cultured with CAFs isolated from AML bone marrow or fibroblasts from healthy donors for 24, 48, 72, and 96 hours. Morphologic changes were assessed by microscopy. Bulk RNA sequencing was performed on conditioned CD34 cells, followed by differential gene expression and GSEA. Temporal pathway evolution was analyzed to distinguish early versus sustained transcriptional programs. scRNA-seq was performed on AML-derived CAFs to define functional subtypes and infer niche contributions.

Results: CAF co-culture induced marked morphologic alterations in CD34 cells, including cytoskeletal remodeling and activated phenotypes. Transcriptomic profiling revealed coordinated activation of innate immune and inflammatory pathways, including Toll-like, NOD-like, and RIG-I-like receptors, IL17, TNF α /NF- κ B, and JAK/STAT3 signaling. Complement components and antigen presentation machinery (MHC II genes, TAP2, PSMB8/9) were strongly upregulated, together with PD-L1 expression and enrichment of PD-1/PD-L1 checkpoint signaling, indicating immune activation with immunoregulatory priming.

CAF-exposed CD34 cells also activated stress-adaptive and survival pathways, including PI3K/Akt, MAPK, FoxO, p53, HIF-1, autophagy, and senescence programs, supporting persistence under inflammatory and metabolic stress. Metabolic rewiring included enrichment of central carbon metabolism, lipid and sphingolipid metabolism, and AGE-RAGE signaling. Genes regulating cytoskeletal dynamics, adhesion, and stromal integration were induced, supporting enhanced niche engagement.

Temporal analysis showed progression from early inflammatory priming (24 hours) to a stabilized chronic inflammatory and oncogenic core at 96 hours, marked by sustained NF- κ B, TNF, JAK/STAT, and checkpoint signaling. In contrast, normal fibroblasts preserved mitochondrial metabolism, DNA repair, oxidative phosphorylation, and maintained a homeostatic progenitor state.

scRNA-seq identified heterogeneous CAF subtypes, including myofibroblastic, inflammatory, immune-regulatory, antigen presenting like, metabolic/stress-adapted, hypoxia-responsive, proliferative, and signaling active CAFs. These subpopulations exhibited complementary functions driving inflammatory amplification, checkpoint ligand induction, metabolic conditioning, and progenitor fate modulation.

Conclusions: AML bone marrow derived CAFs are sufficient to reprogram normal CD34 cells toward an inflammation-primed, immune-interactive, metabolically adapted, tumor-supportive phenotype. Persistent exposure stabilizes a chronic inflammatory and oncogenic signaling core distinct from programs maintained by normal fibroblasts. These findings define a CAF-driven reprogramming axis that conditions hematopoietic progenitors toward carcinogenic states and highlight the stromal niche as a therapeutic target in AML.

Translational/Clinical Research

Research Technician

POSTER #185

PATIENT CHARACTERISTICS AND SURVIVAL OUTCOMES OF EARLY-STAGE RELAPSED, METASTATIC RENAL CELL CARCINOMA

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Background: The standard of care for clinically localized renal cell carcinoma (RCC) remains surgical resection followed by consideration of adjuvant pembrolizumab for patients (pts) with high-risk localized RCC. Many predictive models exist to help risk stratify patients and predict postoperative recurrences, though pts with early stage RCC are generally thought to have low rates of recurrence. Here, we describe patient characteristics and survival outcomes of pts with initially early stage RCC who relapsed with metastatic disease.

Methods: A prospectively maintained RCC database at Indiana University was queried for pts with localized, either stage I or II, RCC who were treated with surgery (partial or radical nephrectomy) and later had relapse of RCC with metastatic disease. Baseline characteristics, including disease characteristics and genomic alterations identified by NGS, were summarized. The Kaplan-Meier method was used to analyze progression free survival (PFS) and overall survival (OS).

Results: 38 pts were included. Median age at diagnosis was 56.1 years (range: 29-73). 27 (71.1%) of pts were male. Histology was 71.1% clear cell, 10.5% chromophobe, 7.8% papillary, 5.3% mixed, and 5.3% unclassified. Rhabdoid and/or sarcomatoid features were present in 10.6% of pts. Pathologic T staging was T1 in 65.8% and T2 in 34.2%. IMDC risk was good in 47.4% of pts, intermediate in 50%, and unknown in 2.6%. Fuhrman grade was 4 in 3.4%, 3 in 4.2%, 2 in 47.4%, 1 in 2.6%, and unknown in 42.4%. Metastasis sites included lung 52.6%, regional lymph nodes (LN) 36.8%, bone 36.8%, distant LN 23.7%, liver 15.8%, and brain 10.5%. Most common genomic alterations included VHL 50.0%, PBRM1 34.2%, TP53 21.1%, SETD2 18.4%, and PTEN 10.5%. 2-year PFS was 20.2% (7.5-37.2) and 2-year OS was 82.4% (64.7-91.7).

Conclusion: In this cohort of pts, almost two-thirds of pts had initially T1 resected disease. The majority of pts actually had Fuhrman grade 2 disease, and there was a surprisingly low rate of sarcomatoid/rhabdoid features. Over half of pts relapsed with lung metastasis. 2-yr OS was high, likely reflective of slower disease biology.

Translational/Clinical Research Resident

POSTER #186

AI-BASED PROGNOSTIC RISK STRATIFICATION OF ADULT-TYPE DIFFUSE GLIOMAS USING H&E-STAINED SLIDES

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Background: Adult-type diffuse gliomas (ADG) are aggressive brain tumors with highly variable clinical outcomes. Reliable prognostic assessment from clinical routine H&E-stained whole slide images (WSIs) contributes to clinical decision-making and patient management optimization, but remains challenging due to tumor heterogeneity. We present an artificial intelligence (AI) approach for overall survival risk stratification directly from H&E-stained WSIs offering a promising approach to uncover novel prognostic markers.

Data and Method: We utilized 1,533 WSIs (735 patients) from the multi-institutional TCGA-GBM and TCGA-LGG collections, under a 10-fold cross-validation configuration with patient-level partitioning. After comprehensive WSI curation, we systematically benchmarked eleven pathology-specific foundation models (FMs) and an AI model pretrained on ImageNet, along seven multiple instance learning (MIL) methods, within a weakly supervised framework to predict patient-level risk scores. These scores were used to further stratify patients in high- vs. low-risk groups, and evaluate performance according to subtype (oligodendroglioma, astrocytoma, glioblastoma).

Results: The UNI FM model with MambaMIL achieved the optimal C-index of 0.77 (95% confidence interval (CI): [0.734, 0.784]). Stratifying patients into high- vs. low-risk groups ($p < 0.005$, log-rank) yielded a highly favorable hazard ratio (HR) of 2.97 (95%CI:[0.41, 9.54]). Subtype-specific analysis in astrocytoma, IDH-mutant) and glioblastoma, IDH-wt showed HR of 1.82 (95% CI : [0.94, 1.76]) and 1.28 (95% CI : [0.94,1.76]), respectively, demonstrating subtype-dependent performance. Key insights include: (i) pathology-specific FMs substantially outperform ImageNet-based model; (ii) advanced spatially informed MILs contribute marginal performance improvements; (iii) no single MIL method consistently improves performance across all FMs, indicating that the optimal MIL choice may be FM-dependent. **Conclusion:** Our results, indicating strong concordance between predicted risk and actual outcomes, support the integration of FMs with MIL methods for WSIs-based risk stratification across ADG patients. Our findings underscore the potential of computational pathology complementing existing neuropathological diagnostic assessment towards enhancing patient prognostic stratification, improving clinical decision-making in glioma care, and advancing personalized neuro-oncology.

POSTER #187

SUBCLONAL GENOMIC HETEROGENEITY INFLUENCE IMMUNE MICROENVIRONMENT AND CLINICAL OUTCOMES IN MULTIPLE MYELOMA

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Introduction: Subclones—distinct populations of myeloma cells—play a critical role in disease progression, treatment response, and relapse in multiple myeloma (MM). Understanding subclone-level transcriptomic and genomic changes, along with immune interactions, is essential for identifying therapeutic targets. We analyzed MM samples - smoldering (SMM); newly diagnosed (NDMM); and relapsed/refractory (RRMM) - to explore the genomic landscape of subclones and their interactions with immune microenvironment.

Methods: We used CD138+ scRNA-seq and scATAC-seq multiomic data from myeloma patients (n=49; dbGAP phs003220) and matched CD138- scRNA-seq (immune cell) data (Sudha et al., IMS 2023). Subclones were identified using inferCNV and a custom integration pipeline. CellChat inferred subclone-immune cell interactions. Copy number abnormalities (CNAs) from single-cell and whole genome sequencing (WGS) were compared for concordance. Transcription factors (TFs) were identified from differentially expressed and accessible genes/regions for subclones, which were then mapped to the CNAs and ligand-receptor interaction pathways to identify the key therapeutic targets.

Results: Myeloma subclones were identified (23 in SMM; 79 in NDMM; and 53 in RRMM) and their CNAs were compared to WGS with a mean Jaccard index (JI) of 0.57 for SMM; 0.69 for NDMM; and 0.5 for RRMM. Samples with low JI tended to have more subclones and more complex CNAs as measured through ploidy. Gain1q and Del13q are consistently frequent across stages of MM. In NDMM patients with co-occurring 1q gain and 13q loss within the same clone had significantly shorter progression-free survival (P = 0.004) compared to those with at least one of these alterations. In NDMM, subclones with Gain1q had a lower average percentage of interactions with CD8T cells (P=0.005). As MM progress, we identified an increase in MIF-CD74/CXCR4 interactions. SMM subclones with Del13p tended to have a lower percentage of MHC-I interactions in CD8T, CD4T and NK cells. Gain6p subclones had a higher percentage of MK pathway interactions in SMM subclones. In NDMM, Gain1q and Del 13q is associated with significantly reduced MHC-I pathway interactions with CD8T cells and reduced MHC-II pathway interactions with CD4T cells. Del17p showed reduced MHC-I interaction in CD8T cells and MHC-II interaction in CD4T cells in Relapse samples. *ETV1*, *SMAD1*, *TEAD1*, *GLI3*, *KLF4* were identified as top frequently occurring TFs in MM.

Conclusions: The identification of subclone-specific immune interaction networks provides new opportunities for targeted therapeutic intervention. These results establish subclonal CNA profiling as both a prognostic tool and guide for precision immunotherapy in multiple myeloma, particularly for high-risk disease characterized by specific genomic alterations co-occurring in subclones. This study underscores the importance of single-

cell analysis to fully capture the clinically relevant genomic heterogeneity that drives disease progression and treatment resistance.

Translational/Clinical Research *Staff*

POSTER #188

STRESS, PHYSICAL FUNCTION, AND COPING DIFFERENCES AMONG BREAST CANCER SURVIVORS WITH AND WITHOUT CARDIOVASCULAR COMORBIDITIES

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Background: There are over 4 million breast cancer survivors (BCS) in the US; breast cancer treatment can impact cardiovascular health. In addition to treatment, BCS can face an increased risk of cardiovascular comorbidities (CC), such as coronary artery disease, heart failure, and hypertension, due to overlapping risk factors including stress, poor physical functioning, and negative coping. The purpose of this study was to examine treatment, stress, physical function, and substance use for coping differences among BCS with and without CC.

Methods: This is a cross-sectional, secondary data analysis of a larger parent study. Participants were recruited online through social media platforms and cancer-affiliated websites. Those interested in participating were sent a questionnaire to first assess eligibility. Eligible participants then completed a questionnaire which included demographic and treatment information, comorbidities, Perceived Stress Scale (PSS-10), Physical function-10 (PF-10), and Brief COPE, substance use coping item. The PSS-10 measures perceived stress over the past month on a 4-point scale with higher scores indicating greater perceived stress. The PF-10 assesses perceived limitations in physical functioning in daily activities on a 3-point scale, with higher scores indicating fewer physical limitations and better physical functioning. The Brief COPE measures the frequency of coping strategies related to diagnosis and treatment on a 4-point scale, we focused on the item related to substance use, where higher scores indicated the use of drugs and/or alcohol for coping. We analyzed two groups, BCS with and without CC. We used descriptive statistics, independent t- tests, and regression to analyze the data.

Results: Of 589 BCS, 41.6% (N=245) had at least one CC. The frequency of CC per participant was 1 CC 26.5% (n=156), 2 CC 13.1 % (n=77), 3 CC 1.9% (n=11), and 4 CC 0.2% (n=1). On average, BCS with CC were older than those without M=59.22 (SD=11.07) vs. M=53.94 (SD=11.99). Overall, BCS with CC had significantly poorer physical functioning (p<0.001) and were significantly older (p<0.001). Stress and coping via substance use were not significantly different between groups. In a regression analysis with age, education, and CC with physical functioning as the outcome, the model was statistically significant [F(3,556) =30.78, R²=0.142, adjusted r²=0.138, p<0.001]. The model explained 13.8% of the variance of physical functioning, with education (β =0.251, p<0.001) and CC (β =-0.238, p<0.001) related to physical functioning. These results indicated that more education and no CC, were related to better physical functioning.

Conclusions: Older BCS, especially those with CC, are more likely to suffer from poor physical functioning. Multidisciplinary teams contributing to bedside care, should monitor for CC among BCS as well as potential related factors such as impaired physical functioning. More research in this area is essential to fully understand the risk factors and downstream impact of CC among BCS.

Translational/Clinical Research

Undergraduate Student

POSTER #189

EVALUATING PI(4,5)P₂ SPATIAL ORGANIZATION AS A POTENTIAL BIOMARKER IN TRIPLE-NEGATIVE BREAST CANCER

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Triple-negative breast cancer (TNBC) is the most aggressive breast cancer subtype and lacks effective selective therapies that target cell surface proteins, highlighting the need for biomarkers of disease progression. To address this need, the Kimble-Hill lab has endeavored to find suitable cell surface lipid biomarkers. Recent work suggests the concentration of phosphatidylinositol lipid PI(4,5)P₂ increases under metabolic stress making it a suitable biomarker for aggressive TNBC, but it remained unclear whether this reflects spatial reorganization from junctions to the apical membrane during cancer progression. We hypothesized that PI(4,5)P₂ would lose spatial coordination with adhesion proteins in TNBC compared to non-cancerous cells. To test this, cells were immunolabeled for junctional proteins and PI(4,5)P₂, imaged using fluorescence microscopy, and analyzed using colocalization coefficients to evaluate cell-cell junction organization. Pearson correlation analysis showed that tight junction protein occludin colocalization with PI(4,5)P₂ remained strong across all cell lines, indicating retention of PI(4,5)P₂ at cell-cell junctions. However, more work is needed to understand potential functional changes at adherens junctions. These results narrow a key gap of knowledge in the role of these phosphatidylinositols in TNBC progression, further demonstrating the need for more spatial analysis in determining suitable lipid biomarkers.

Translational/Clinical Research

Undergraduate Student

POSTER #190

ESTROGEN (ER) AND PROGESTERONE (PR) IMMUNOHISTOCHEMISTRY IN BREAST CANCER IN TISSUE MICROARRAYS WITH ARTIFICIAL INTELLIGENCE QUANTITATIVE IMAGE ANALYSIS

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In 2024, an estimated 310,720 new cases of invasive breast cancer are expected in women in the U.S., along with 2,790 cases in men. Tissue Microarrays are a platform for analysis of tissue on multiple cases at once. This platform is constructed of a paraffin block that act as a recipient block when transferring tissue specimen. HR estrogen and progesterone refers to immunostaining hormone receptors in breast cancer cells, where "HR +" means the cancer is "hormone receptor-positive" and is fueled by hormones like estrogen (ER) and progesterone (PR). In this study, 6 tissue microarrays of patients from 1992-2012 were analyzed by immunohistochemistry (IHC) to determine expression and localization in breast carcinoma cases with the following well-recognized immunostains: ER and PR. Comparison of the two AI software methodologies were similar. ER contained 37 weak positive and 31 strong positive cores in Aperio, and 45 weak positive and 63 strong positives cores with QuPath. PR contained 25 weak positive and 15 strong positive cores with Aperio and 50 weak positive and 44 strong positive cores with Qupath. The differences in the number of positive cores between Aperio and QuPath is due to the difference in the way they analyze cores. Aperio analyses based on the pixels , which QuPathanalyses the cores based on number of stained cells. Overall, there is a consistent pattern with both algorithms see figures 3 and 4.

Translational/Clinical Research

Undergraduate Student

POSTER #191

MYC-HSF1 CO-AMPLIFIED OVARIAN CANCERS DISPLAY SENSITIVITY TO HDAC AND PLK1 INHIBITION

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High-grade serous ovarian cancer (HGSOC) remains one of the most lethal cancers, with approximately 20,000 women diagnosed annually in the United States and only 51% surviving five years after diagnosis. Current treatment options are limited, and unlike other cancers, where targeted therapies utilize specific molecular features, such as HER2- directed therapy in HER2-positive breast cancer or hormone-receptor inhibition in ER- or AR-driven tumors, HGSOC lacks widely used precision-based strategies other than platinum-based therapy regimens with PARP inhibitors. Even with these treatments, patients inevitably advance disease progression as tumor cells gain resistance. One underexamined therapeutic avenue is the targeting of gene-level amplifications. Notably, 31% of HGSOC tumors display co-amplification of the transcriptional regulators *MYC* and Heat Shock Factor 1 (*HSF1*), suggesting a new plan of action for treatments. *MYC* functions as a master oncogenic transcription factor that drives cell growth, metabolism, and proliferation by activating genes involved in ribosome biogenesis, nucleotide synthesis, and cell-cycle progression. *HSF1*, traditionally known for coordinating the heat-shock response, has been shown to support malignant transformation through a cancer-specific transcriptional program that promotes proteostasis, metabolic rewiring, stress tolerance, and survival under oncogenic pressure. Importantly, HSF1 can enhance *MYC*-driven transcriptional outputs, forming an oncogenic network that enhances tumor aggressiveness and treatment resistance. In our study, we investigated how *MYC* and *HSF1* co-amplification shapes therapeutic sensitivity in HGSOC. We found that co-amplified ovarian cancer cells exhibit a distinct drug-response profile compared to non-co-amplified cells. These tumors show marked sensitivity to the Polo-like Kinase 1 (PLK1) inhibitor volasertib, revealing a previously unidentified dependence on PLK1 signaling. Using in vitro assays, including Cell Titer Blue viability assays and colony-formation assays, we further demonstrate that *MYC*-*HSF1* co-amplified cells are sensitive to HDAC inhibitors such as quisinostat and entinostat, which we show suppress *MYC* and *HSF1* transcriptional activity. These findings identify *MYC* and *HSF1* co-amplification as a molecular signature that yields selective vulnerability to PLK1 inhibition and highlights a precision-medicine strategy for a large percentage of patients with HGSOC.

Translational/Clinical Research

Undergraduate Student

POSTER #193

UNDERSTANDING PERSPECTIVES OF CERVICAL CANCER PREVENTION IN PATIENTS ATTENDING THE INDIANA UNIVERSITY STUDENT OUTREACH CLINIC

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Background: Nearly all cases of cervical cancer (CC) are caused by HPV. Although HPV vaccination and regular screenings serve as crucial interventions in preventing CC, significant health inequities persist. Indiana ranks 10th in the U.S. for CC incidence, over half of new CC cases occur in under screened population and children's HPV vaccination rate is 59.6% (ranks 36th). While programs like Medicaid and Vaccines for Children cover preventative cares, barriers to participation in underserved communities remain poorly understood. This study evaluates the knowledge, attitudes, and barriers regarding CC prevention among families at a free community clinic.

Methods: This is an interim analysis of a cross-sectional survey being conducted at the Indiana University Student Outreach Clinic (IUSOC), a free urban clinic in Indianapolis. Participants completed surveys assessing CC history, knowledge (15-item scale), and perceived barriers. Data were analyzed using descriptive statistics and compared by gender.

Results: The analysis included 84 participants (45 females, 39 males; mean age 52). The cohort was racially and socioeconomically diverse: 39.3% African American, 29.8% Non-Hispanic Caucasian, 20.2% Hispanic; 28.8% were uninsured, and 58.2% were unemployed. No significant demographic differences existed between genders. Among women, 52.3% were underscreened (never or >10 years). Self-reported HPV vaccination was 24.1%. Knowledge scores were low overall, though women scored significantly higher than men (8.7 vs. 5.5 correct answers; $p < 0.001$). The primary barriers to screening were cost (49.4%), lack of provider recommendation (40.8%), and preference for a female physician (39.5%). Regarding pediatric HPV vaccination, barriers included lack of vaccine knowledge (50.0%), cost (45.3%), and lack of provider recommendation (41.9%). For adult vaccination, the main barriers were lack of provider recommendation (51.3%), cost (46.8%), and lack of awareness (45.5%). No significant gender differences were found on these barriers. Notably, high willingness was reported for free IUSOC services, including screening (59.5% of women) and vaccination for children (66.2%) or self (66.7%).

Conclusions: Despite poor baseline knowledge and significant barriers, primarily cost and lack of provider recommendation, there is high patient interest in CC prevention. Free clinics like IUSOC are uniquely positioned to improve health equity by providing direct access to screening, vaccination, and targeted patient education.

Community friendly research poster Medical Student

POSTER #194

ECONOMIC IMPACT OF FLUOROPYRIMIDINE-BASED CHEMOTHERAPY LEADING TO HOSPITALIZATION: A RETROSPECTIVE REAL-WORLD CLAIMS DATA ANALYSIS

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BACKGROUND:

Pharmacogenetic testing for variants in *DPYD* can reduce fluoropyrimidine (FP)-toxicities, but additional real-world data regarding the clinical and economic impact of FP-induced toxicities is needed to inform *DPYD* testing policies.

METHODS:

We conducted a retrospective observational study using the de-identified commercial claims database of patients in the US with commercial insurance from the Merative™ MarketScan® Commercial Database. We identified patients who appeared to be hospitalized due to FP-induced toxicities using 3 different strategies of ICD-10 codes. They are patients who were hospitalized within 50 days of starting their first FP dose from 01/01/2016 to 12/31/2022 had enrollment records within 180 days before or one year after the first FP dose. Using 2022 data, we identified ICD-10 codes present in patients with adverse event (AE)-related diagnoses and used those to select patients in all years; patients were excluded if their admission codes indicated non-FP toxicity related causes. All groups of patients were filtered using same FP-induced toxicity codes with different data approaches: Group A and B with inpatient admissions data (summary for each admission) and group C with inpatient services data (details on individual services); two data are linked with the case IDs. For group A, patients with definite indicator for presenting toxicity-related ICD-10 codes on admission, but group B was sorted by the indicator with not 'No' (i.e., missing/unknown, unreported/not used, clinically undetermined, or yes). For group C, all diagnosis codes of individual services were considered to apply FP-induced toxicity codes. To evaluate the variability, we compared the number of patients identified using three selection strategies differing in the frequencies of hospitalizations, days in intensive care units, and emergency department usage.

RESULTS:

59,731 patients were prescribed FPs. We identified 1,027 patients (~1.7 % of new FP prescriptions) who were hospitalized most likely due to FP-related toxicities (see table). Costs associated with the hospitalizations are being calculated and will be reported as part of the presentation.

CONCLUSION:

These real-world data illustrate the substantial healthcare burden caused by the FP-related toxicities.

Translational/Clinical Research

Graduate Student

POSTER #192

AI-ENABLED RESEARCH PLATFORM AT INDIANA UNIVERSITY SIMON COMPREHENSIVE CANCER CENTER

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The Biospecimen Collection and Banking Core (BC2) is a vital component of the Cancer Center, and provides comprehensive biospecimen management, supporting cancer research through collection, storage, distribution, and annotation of biospecimens and related data. BC2 includes over 28,000 participants with 31,000 cancer diagnoses representing 70 unique cancers and 557 unique histologies and over 100 TB of related data. BC2 has partnered with Manifold to build an AI-enabled trusted research environment. The system has ingested, transformed, and harmonized data from multiple sources. This integration provided immediate, full visibility into a previously inaccessible dataset. Complex specimen and data queries that used take weeks of research can be done in minutes. Cancer Center researchers now have access to the full scope of data and specimens available for research, enabling them to get answers to feasibility questions faster and accelerate research timelines.

Basic Science Core Facility