# OCMF 2023

November 13 - 14, 2023

Minneapolis, MN

# Welcome

## We are delighted to present the 5th annual Ovarian Cancer Midwest Focus (OCMF) Conference

The event will be hybrid, with both in-person and live streamed discussion rounds with national and international speakers taking place on November 13 & 14, 2023 in Minneapolis, MN.

| Program Chairs |
| --- |
| **Martina Bazzaro, Ph.D.** - University of Minnesota School of Medicine |
| **Ronny Drapkin, M.D., Ph.D.** - University of Pennsylvania - Perelman School of Medicine |
| **Andrew Godwin, Ph.D.** - University of Kansas Medical Center - Kansas Institute for Precision Medicine |
| **Gottfried E. Konecny, M.D.** - University of California - Los Angeles |
| **Boris Winterhoff, M.D., M.S.** - University of Minnesota School of Medicine |
| **Paola Vermeer, Ph.D.** - Sanford Research - Sioux Falls and University of South Dakota |

| Staff |
| --- |
| **Danielle Girtz** - Event Coordinator, University of Minnesota |
| **Kristina Cecka** - Program/Project Specialist, University of Minnesota |
| **Brenda Olson** - Program/Project Specialist, University of Minnesota |
| **Amy D. Cavanaugh -** Associate Administrator, University of Minnesota |

***OCMF 2023 would like to thank the following sponsors for their generous contributions. Your support and partnership played a key role in the success of our conference, and we appreciate your participation.***

CARIS LIFE SCIENCES

gsk

Early Bird Medical Inc.

KANSAS INSTITUTE FOR PRECISION MEDICINE
ADVANCING CARE ONE PATIENT AT A TIME
COBRE

immunogen

AstraZeneca

minerva SURGICAL

SANFORD HEALTH

MERCK

OCRC
OVARIAN CANCER RESEARCH CENTER

MASONIC CANCER CENTER
UNIVERSITY OF MINNESOTA

# DAY 1: NOVEMBER 13, 2023

| | |
|---|---|
| **Location:** | Coffman Memorial Union, Mississippi Room |
| **Breakfast:** | 7:15 AM - 8:00 AM |
| **Opening Remarks:** | 8:00 AM - 8:15 AM<br>**Martina Bazzaro, Ph.D.**, University of Minnesota |
| **Welcome Videos:** | 8:15 - 8:20 AM<br>**Tina Smith**, U.S. Senator from Minnesota |

**Session I: Targeting the Cell of Origin and the Microenvironment of Ovarian Cancer**
**Moderator:** Andrew Godwin, Ph.D., University of Kansas Medical Center

| Time | Title | Presenter |
|---|---|---|
| **8:25 - 8:45 am** | Cytomegalovirus (CMV) and Patient-Reported Outcomes in Ovarian Cancer | **Rachel Vogel, Ph.D.,** University of Minnesota |
| **8:45 - 9:05 am** | The Role of the Extracellular Matrix in Ovarian Cancer | **Sandra Orsulic, Ph.D.**, University of California, Los Angeles |
| **9:05 - 9:25 am** | Molecular Mechanisms Which Regulate Fitness Fingerprints Between Cancer and its Microenvironment | **Esha Madan, Ph.D.,** Virginia Commonwealth University |
| **9:25 - 9:45 am** | Housing temperature as a new model for studying the impact of chronic adrenergic stress on tumor growth and anti-tumor immunity | **Elizabeth Repasky, Ph.D.,** Roswell Park Comprehensive Cancer Center |
| **9:45 - 10:05 am** | Question and Answer Session | |
| **10:05 - 10:30 am** | Break | |

# DAY 1: NOVEMBER 13, 2023

| Time | Title | Presenter |
|---|---|---|
| **Abstract Flash Talks** | | |
| **10:30 - 10:50 am** | The Aged Microenvironment Worsens Disease Progression in a Syngeneic HGSOC Model | **Katherine Cummins, Ph.D.**, University of Pennsylvania |
| | Ovarian and Fallopian Tube Crosstalk via Extracellular Vesicles Promotes the Origin of Ovarian Cancer and the Precancerous Landscape | **Jared Sipes**, Kansas University Medical Center |
| | Circulating extracellular vesicles protein biomarkers for early detection of high-grade serous ovarian cancer | **Sagar Rayamajhi, Ph.D.,** University of Kansas Medical Center |
| | Armored CAR-NK cell Therapy for Ovarian Cancer | **Joshua Krueger**, University of Minnesota |
| **10:50 - 11:10 am** | Question and Answer Session | |
| **11:10 - 11:40 pm** | Lunch Break | |
| **Sponsor Presentations** | | |
| **11:40 - 11:50 am** | Caris Life Sciences | **Jeff Hirst, Ph.D.** |
| **11:50 - 12:00 pm** | Immunogen | **Darren Kress, B.S.** |
| **12:00 - 12:10 pm** | Sanford Research | **Ellie Gibbons** |
| **12:10 - 1:10 pm** | Poster Session | |

## Session II: Ovarian Cancer Stem Cells

**Moderator:** Martina Bazzaro, Ph.D. - University of Minnesota School of Medicine

| Time | Title | Presenter |
|---|---|---|
| **1:10 - 1:30 pm** | CDON: A potential therapeutic vulnerability in ovarian cancer | **Denise Connolly, Ph.D.**, Fox Chase Cancer Center |
| **1:30 - 1:50 pm** | Combining DNA methyltransferase inhibitors with platinum as a strategy to prevent recurrent ovarian cancer | **Heather O'Hagan, Ph.D.,** Indiana University School of Medicine – Bloomington |

# DAY 1: NOVEMBER 13, 2023

| Time | Title | Presenter |
|---|---|---|
| **1:50 - 2:10 pm** | Targeting mitochondrial vulnerabilities for cancer therapy | **Evripidis Gavathiotis, Ph.D.,** Albert Einstein College of Medicine |
| **2:10 - 2:30 pm** | Question and Answer Session | |
| **2:30 - 2:40 pm** | Break | |
| **2:40 - 3:10 pm** | Advancing Immunotherapy in Ovarian Cancer | ***Keynote Speaker*** **George Coukos, M.D., Ph.D.** University of Lausanne |
| **3:10 - 3:30 pm** | Intra-Tumoral Nerves in Immunosurveillance | **Sebastien Talbot, Ph.D.,** Queen's University, Ontario |
| **3:30 - 3:50 pm** | Susceptibility Genes in the Less Common Histotypes of Epithelial Ovarian Cancer | **Susan Ramus, Ph.D.,** School of Clinical Medicine, UNSW Sydney |
| **3:50 - 4:10 pm** | The DOD OCRP Ovarian Cancer Academy: Early Career Investigators Making a Difference in Ovarian Cancer Research | **Kenneth Nephew, Ph.D.,** Indiana University - School of Medicine |
| **4:10 - 4:25 pm** | Question and Answer Session | |
| **4:25 - 4:30 pm** | Closing Remarks | |

## *RECEPTION*

**Monday, November 13**

**Post-Conference - 9:00 PM**

*Join us for heavy apps, drinks, games, and bowling at Goldy's Gameroom. Located in the basement of Coffman Memorial Union!*

# DAY 2: NOVEMBER 14, 2023

| | |
|---|---|
| **Location:** | Coffman Memorial Union, Mississippi Room |
| **Breakfast:** | 7:15 AM - 8:00 AM |
| **Opening Remarks:** | 8:00 AM<br>**Martina Bazzaro**, **Ph.D.**, University of Minnesota School of Medicine |
| **Welcome Video:** | 8:00 - 8:05 AM<br>**Jakub Tolar**, **MD, PhD,** Dean, University of Minnesota Medical School |

## Session 3: Tumor Microenvironment 2 and Aging

**Moderators:** Ronny Drapkin, M.D., Ph.D. - University of Pennsylvania
Paola Vermeer, Ph.D. - Sanford Research and University of South Dakota

| Time | Title | Presenter |
|---|---|---|
| **8:05 - 8:30 am** | Developing Tools to Detect Millimeter and Sub-millimeter Ovarian and Fallopian Tumors-Towards Early Detection and Interception of Ovarian Cancer | ***Keynote Speaker***<br>**Angela Belcher, Ph.D.,** Koch Institute for Integrative Cancer Research |
| **8:30 - 8:50 am** | Replication Driven Vulnerabilities In Cyclin E Overexpressing Ovarian Cancers | **Priyanka Verma, Ph.D.,** Washington University School of Medicine |
| **8:50 - 9:10 am** | Progesterone: Ovarian and Breast Cancer in BRCA1 Carriers | **Jaeyeon Kim, DVM, Ph.D., M.S., M.S.,** Indiana University School of Medicine |
| **9:10 - 9:25 am** | Question and Answer session | |
| **Awards Session**<br>**Presenters:** Tim Schacker, MD, Aaron Schilz, MPA, Rahel Ghebre, M.D., MPH | | |
| **9:25 - 9:40 am** | Award for Outstanding Support for Ovarian Cancer Research | **Kathleen Gavin, MPH**, Minnesota Ovarian Cancer Alliance |
| | Inspiration and Philanthropy Award | **Randy & Roseann Shaver**, Randy Shaver Cancer Foundation |
| | Andrew Godwin Award for Excellence in Community Outreach | **Sarah DeFeo, MPA**, Ovarian Cancer Research Alliance |

# DAY 2: NOVEMBER 14, 2023

| Time | Title | Presenter |
|---|---|---|
| **9:40 - 9:50 am** | Break | |
| **9:50 - 10:10 am** | Engineered bacteria for cancer therapy | **Tal Danino, Ph.D.,** Columbia University |
| **10:10 - 10:30 am** | Role of ZNF217 in ovarian cancer metastasis | **Achuth Padmanabhan, Ph.D., B.S.**, University of Maryland |
| **10:30 - 10:40 am** | Question and Answer session | |
| **Abstract Flash Talks** | | |
| **10:40 - 11:00 am** | Exploiting JAK/STAT signaling to inhibit highly advanced and resistant forms of ovarian cancer | **Esther Rodman**, Mayo Clinic |
| | Tumor-Infiltrating Nociceptor Neurons in Ovarian Cancer Progression and Treatment Resistance | **Austin Walz, M.S.**, Sanford Research |
| | Novel Myc-dependent compound DL78 selectively induces mitotic catastrophe and cell death in high grade serous ovarian cancer | **Jessica Teitel,** University of Michigan |
| | CTPS1: an Unexplored Vulnerability in Ovarian Cancer | **Xiyin Wang**, Mayo Clinic |
| **11:00 - 11:20 am** | Question and Answer session | |
| **11:20 - 12:00 pm** | Lunch Break | |
| **Sponsor Presentations** | | |
| **12:00 - 12:10 pm** | Masonic Cancer Center | **Rahel Ghebre, M.D., MPH** |
| **12:10 - 12:20 pm** | Minerva Surgical | **Matt Flaig** |
| **12:20 - 1:00 pm** | Poster Session | |

# DAY 2: NOVEMBER 14, 2023

**Session 4: Translating Innovation to the Clinic**

**Moderators:** Gottfried E. Konecny, M.D., University of California - Los Angeles
Boris Winterhoff, M.D., M.S., University of Minnesota School of Medicine

| Time | Title | Presenter |
|---|---|---|
| **1:00 - 1:20 pm** | Beyond Darwin: understanding cancer persister cells | **Yaara Oren, Ph.D., BSc,** School of Medicine, Tel Aviv University |
| **1:20 - 1:40 pm** | Novel ADC's in ovarian cancer | **Stephanie Lheureux, M.D., Ph.D.,** UNH Toronto |
| **1:40 - 2:00 pm** | Combinatorial Therapies For Ovarian Cancer Treatment | **Joyce Liu, M.D., MPH,** Dana-Farber Cancer Institute |
| **2:00 - 2:20 pm** | Question and Answer Session | |
| **2:20 - 2:30 pm** | Break | |
| **2:30 - 2:50 pm** | Targeting immune vulnerabilities and heterogeneity in ovarian cancer | **Dmitriy Zamarin, M.D., Ph.D.,** Tisch Cancer Institute at Icahn School of Medicine at Mount Sinai |
| **2:50 - 3:10 pm** | Targeting the Damaged Transcriptome as an Emergent Vulnerability in Ovarian Cancer | **Robert Rottapel, M.D.,** University of Toronto |
| **3:10 - 3:20 pm** | Question and Answer Session | |
| **3:20 - 3:30 pm** | Closing Remarks | |

Thank You

*On behalf of the OCMF Program Chairs, we would like to express our sincere gratitude to the Ethel N. Ruvelson Memorial Lectureship in Ovarian Cancer for their generous support of this conference. The Ethel N. Ruvelson Memorial Lectureship in Ovarian Cancer fund helps us continue to fund vital research lectureships and provide educational programs for our faculty and researchers at the Univ. of Minnesota.*

## JOIN US NEXT YEAR!

OCMF Annual Conference 2024
November 18 - 19, 2024
Sioux Falls, South Dakota

# FLASH TALK ABSTRACTS
# TABLE OF CONTENTS

# The Aged Microenvironment Worsens Disease Progression in a Syngeneic HGSOC Model

**Katherine Cummins**, University of Pennsylvania; **Matthew Knarr**, University of Pennsylvania; **Ronny Drapkin**, University of Pennsylvania

It is noteworthy that over 70% of high-grade serous ovarian cancer (HGSOC) patients are diagnosed after age 55, and the average patient who loses their battle with the disease is 70 years old. Despite this strong correlation with advanced age, most ovarian cancer studies, and cancer research generally, utilize young mice and do not investigate age as a variable despite it being a top risk factor for cancer. An improved understanding of how an aged TME affects HGSOC progression could enable development of alternative treatment strategies to block the metastatic cascade.

We developed a secretory epithelial line from C57BL/6 oviducts and used CRISPR to create a tumorigenic line via Trp53, Pten, and Brca2 deletions. These cells were injected intraperitoneally into 6- and 56-week-old B6 mice, comparable to 10- and 50-year-old humans. A drastic increase in tumor burden was evident in older mice and overall survival was worse than in the young cohort. In a longitudinal kinetics study with bioluminescent imaging, aged mice demonstrated significantly higher tumor burden at initial readings, and tumor growth was faster in aged animals.

Through bulk RNA sequencing, only 5 genes were differentially expressed in samples obtained from aged mice compared to young (FDR < 0.1) indicating the tumor is not likely undergoing significant changes, but instead, the aged environment is more permissive. When stratifying the data by metastatic site, more genes were identified as significant; gene ontology for tumors lining the peritoneum indicated upregulation in humoral immune response and lipid metabolism in older animals.

Initial studies utilizing flow cytometry to assess the tumor-naïve microenvironment demonstrated a drastic increase in B-cell populations and a modest, yet still significant, increase in T cells. When probing for T-cell exhaustion, PD-1 was found to be expressed on more CD4+ T cells in aged mice, and while no increase was seen in CD8+ T cells, PD-1 expression was high across both groups. It is perhaps counterintuitive that an increase in total immune cells is associated with a more tumor-permissive environment in aged animals, however, immune cell state may vary widely between the age groups. Ongoing studies are aimed at further characterizing the functional phenotype of these lymphocytes, as well as profiling infiltrating immune cells in tumors and ascites.

# Ovarian and Fallopian Tube Crosstalk via Extracellular Vesicles Promotes the Origin of Ovarian Cancer and the Precancerous Landscape

**Jared Sipes[1], Didi Zha[2], Sagar Rayamajhi[1], Harsh Pathak[1], Mihaela Sardiu[3], Leonidas E. Bantis[3], Joanna E. Burdette[2], Andrew K. Godwin[1 4]**

[1]Department of Pathology and Laboratory Medicine, University of Kansas Medical Center. [2]Department of Pharmaceutical Sciences, College of Pharmacy, University of Illinois at Chicago. [3]Department of Biostatistics and Data Science, University of Kansas Medical Center. [4]Kansas Institute for Precision Medicine, University of Kansas Medical Center.

Small extracellular vesicles (sEVs) are 50–200 nm sized lipid bilayer enclosed particles containing protein, miRNA, and other cell components. Circulating sEV levels are elevated in many cancers and EVs contribute to cancer progression, metastasis, and immune escape via transfer of selective biomolecules. However, models of EV release show cancer EVs alone cannot explain these concentration changes, suggesting significant EV-mediated cross-communication between cancer and normal tissues. High grade serous ovarian cancer (HGSOC), the most common and lethal epithelial ovarian cancer (EOC) subtype, arises from fallopian tube (FT) precursor lesions. Oncogenic insults from follicular fluid, ovarian secretions, and the FT play a role in progression from early lesion to high grade cancer, but the role of EVs in this development is under studied. We hypothesized that EOC EVs may mimic early oncogenic insults to the FT which contribute to HGSOC initiation and can reveal biomarkers for early detection. To explore this question, we model EV-tissue interaction using a 3D dynamic culture system (PREDICT). Samples of hFTE from women undergoing non-cancer related surgeries (n=10) were exposed to physiological levels (~1.5x109) of EVs derived from cancerous (OVCAR3)- or immortalized non-tumorigenic FT (FT240)-derived cells. 24 hours post exposure, FT tissue samples were collected, processed, and profiled using the Cancer Transcriptome Atlas panel (~1,800 mRNA gene targets) and the GeoMx Digital Spatial Profiler. We show that OVCAR3-derived EVs upregulate chemokines(e.g., CXCL1, CCL2 , and CCL20), increase expression of SOX17 (an EOC immunomarker), and enhance expression of cell adhesion genes, VCAM and ICAM1, suggesting the development of preneoplastic landscape. We confirm changes in VCAM, CCL2, and SOX17 protein via immunohistochemistry and hypothesize a mechanism whereby B7-H3 (present in OVCAR3, but not FT240 EVs) may activate the STAT3 pathway to increase CCL2 transcript expression, an encoded chemokine which has been indicated in M2 macrophages polarization and reduction of IFNγ secretion by CD8+ T cells. Our study has shown for the first-time important implications of sEV mediated crosstalk in the earliest stages of HGSOC, which can have broader applications in the development of EV-based biomarkers and therapeutic targets for the management of ovarian cancer.

# Circulating extracellular vesicles protein biomarkers for early detection of high-grade serous ovarian cancer

**Sagar Rayamajhi[1], Jared Sipes[1], Mihaela Sardiu[2], Andrew K. Godwin[1 3 4]**

[1]Department of Pathology and Laboratory Medicine, University of Kansas Medical Center. [2]Department of Biostatistics and Data Science, University of Kansas Medical Center. [3]Kansas Institute for Precision Medicine, University of Kansas Medical Center. [4]University of Kansas Cancer Center, University of Kansas Medical Center.

Early detection is key to improving the survival rate of high-grade serous ovarian cancer (HGSOC) patients. Detecting biomarkers in blood, also known as liquid biopsy, for early detection is a long-sought goal. Extracellular vesicles (EVs) – tiny lipid-bilayer delimited particles released by cells – are considered a new class of liquid biopsy biomarkers. EVs are heterogeneous with various subtypes based on their size and biogenesis pathways. Here, we focus on small EVs (sEVs), with a size range of 50-200 nm. sEVs contain a cargo of selectively sorted biomolecules that can mirror the physiological status of the cells of origin. We hypothesize that sEVs play an active role in early pathogenesis and are secreted in circulation with cargo reflective of early malignancy, and hence could be used as biomarkers for early detection. To advance this idea, we established a case-control study cohort of archival plasma samples from 30 HGSOC patients (10 early-stage and 20 late-stage) and 40 healthy controls. sEVs were isolated using size exclusion chromatography with an optimized method aimed to enrich sEVs with minimal soluble proteins. sEV-enriched fractions (F7-F11) were pooled and concentrated for tandem mass-spectrometry-based proteomics following the data-independent acquisition method. 1,078 EV proteins (exo-proteins) were identified across all samples with 133 differentially expressed exo-proteins (DEP) (logFC>1, $p<0.05$) in HGSOC plasma vs. healthy controls. Upregulated DEPs were compared with the proteome dataset of serous tubal intraepithelial carcinoma (STIC) of fallopian tube tissue, a precursor for most HGSOC. 5 upregulated exo-proteins (MYL6, PRDX1, PRDX6, GSTP1, FLNA) in early-stage HGSOC were shared with FT-lineage specific STIC proteome, showing the potential of these exo-proteins to reflect the earliest stages of ovarian cancer pathogenesis. Furthermore, among 133 DEPs, upregulated membrane proteins (UMPs) were prioritized aiming for their application in our EV-capture and detection assays (PMID:31123323). Overall, 17 UMPs in early-stage HGSOC, 13 UMPs in late-stage HGSOC, and 3 UMPs (SLC25A5, SLC25A6, MUC1) in both early and late-stage HGSOC were identified. In summary, our exo-proteome analysis using clinical samples has uncovered a set of exo-proteins that are more abundant in early-stage HGSOC and could be used as biomarkers for its early detection.

# Armored CAR-NK cell Therapy for Ovarian Cancer

**Joshua B. Krueger, Jae-Woong Chang, Young Y. Vue, Alexandria Gilkey, Timothy D. Folsom, Ethan Niemeyer, Joseph G. Skeate, Beau R. Webber, and Branden S. Moriarity** (Department of Pediatrics, University of Minnesota, Minneapolis, MN, USA. Masonic Cancer Center, University of Minnesota. Center for Genome Engineering, University of Minnesota. Stem Cell Institute, University of Minnesota.)

Immunotherapy with chimeric antigen receptor (CAR) expressing NK cells engineered via viral vectors has shown exceptional efficacy against hematologic cancers in clinical trials. This method, however, is limited in cargo capacity and large-scale manufacturing for clinical use is cost-prohibitive. We have addressed these shortcomings by engineering CAR-NK cells using non-viral transposon engineering that can be paired with CRISPR/Cas9 editing simultaneously. We believe that this approach avails opportunities that will allow researchers to address some of the more difficult solid tumor cancer types.

Here, we developed a CAR-NK therapy with specific advantages to killing mesothelin positive ovarian cancers which tend to form solid tumors, a new area for CAR NK cells. We started by testing 3 different epitope targets of mesothelin named A, B, and C. Also built into the engineering scheme is the secretion of the cytokine IL15 for increased persistence of the CAR NK cells. The A, B, and C CAR NK cells with soluble IL15 (Armored-NK) were tested in vitro and in vivo. No single CAR target stood out during the in vitro testing so all three went into a pilot in vivo study. In this experiment NSG mice were engrafted with a mesothelin high tumor cell line, A1847. A1847 cells were injected 14 days prior to the Armored CAR NK injection. Both tumor cells and Armored CAR NK cells were injected Intraperitoneal.

At week 6, the control mice, B treated, and C treated mice had tumors growing exponentially. The A CAR seemed to be controlling the tumor. This trend continued for 3 more weeks as the SS1 treated mice did not see significant tumor growth while all other mice in the study had to be euthanized due to tumor burden. A CAR treated mice survived indefinitely with the only detectable tumor being a mass that grew at the injection site, outside of the IP space.

With those results we decided to continue with the A CAR and are planning to add more edits to these cells before attempting a phase 1 clinical trial. The two other modifications that were announced in our pre-IND is a knockdown or knockout of CISH, an immune checkpoint inhibitor as well as a TGFb dominant negative receptor.

# Exploiting JAK/STAT signaling to inhibit highly advanced and resistant forms of ovarian cancer

**Esther Rodman[1], Michael Emch, Ph.D.[1], Archit Bajaj[1], Xiaonan Hou, Ph.D.[2], Scott Kaufmann, M.D. Ph.D.[2], S. John Weroha, M.D. Ph.D.[2], John Hawse, Ph.D.[1]**
[1]Department of Biochemistry and Molecular Biology. [2]Department of Oncology, Mayo Clinic.

Ovarian cancer is the most fatal of all female reproductive cancers due to its late detection, aggressive nature, and propensity to develop resistance to current standards of care. Targeted therapies, including PARP-inhibitors (PARPi) and VEGF-inhibitors, are being included with platinum- and paclitaxel-based chemotherapies in the front-line setting; however, they have thus far failed to improve overall survival in patients without homologous recombination defects. Over 70% of ovarian cancers recur with increasingly resistant disease and there is an obvious need to elucidate the basis for chemotherapy- and PARPi-resistance and devise novel approaches to overcome resistance. Using a small molecule drug screen, we identified lestaurtinib, a known tyrosine kinase inhibitor as a potent inhibitor of treatment naive and highly advanced models of ovarian cancer. To identify lestaurtinib targets, we preformed RNAseq and discovered the JAK/STAT signally pathway as a top hit. We confirmed that essential components of JAK/STAT signaling are constitutively activated in chemotherapy- and PARPi-resistant cells, suggesting that induction of this pathway may drive resistance. Importantly, we found that lestaurtinib-sensitive patient-derived-xenografts (PDXs) exhibited higher expression of multiple JAK/STAT signaling pathway members compared to PDXs that did not respond to lestaurtinib. Genetic knockdown of STAT1 and STAT3 via siRNA, or knockout via CRISPR/Cas9, resulted in significant growth inhibition of ovarian cancer cell lines, confirming their importance in maintaining cell viability and progression. Combining lestuartinib with cisplatin or the PARPi, olaparib, was shown to be synergistic in multiple models, including a chemotherapy- and PARPi-resistant cell line. Finally, we assessed a panel of JAK/STAT inhibitors and found profound differences in their ability to block STAT-phosphorylation and inhibit cell proliferation. Thus, it is critical to further define the mechanisms by which specific STAT-phosphorylation may drive ovarian cancer progression. Ongoing studies are aimed at identifying the unique roles of STAT1 and STAT3, the specific contributions of their tyrosine and serine phosphorylation, and the relevance of up-/down-stream mediators of the JAK/STAT pathway on ovarian cancer cell growth with the goal of informing future clinical trials.

# Tumor-Infiltrating Nociceptor Neurons in Ovarian Cancer Progression and Treatment Resistance

**Austin Walz[1], Daniel W. Vermeer[1], Caitlin S. Williamson[1], Allison Jorgensen[1], Ronny Drapkin[2], Paola D. Vermeer[1]**

[1]Cancer Biology and Immunotherapies Group, Sanford Research. [2]Penn Ovarian Cancer Research Center, Department of Obstetrics and Gynecology, Division of Gynecologic Oncology, University of Pennsylvania, Perelman School of Medicine.

Ovarian cancer is a heterogeneous disease with multiple histologic subtypes that display unique biologic and clinical behaviors. To define the innervation status of the five histological subtypes of ovarian cancer, FFPE samples of: high-grade serous (HGSOC), low-grade serous, clear cell, mucinous and endometrioid carcinoma were collected and stained for β-III tubulin (a pan neuronal marker). Interestingly, only HGSOC emerged as densely innervated. Given the stark difference in tumor innervation evident between the histological subtypes of ovarian cancer, we wondered whether this reflected differences in the neurite outgrowth capabilities of their tumor-released small extracellular vesicles (sEVs). Consistent with the patient data, sEVs from the majority of HGSOC cell lines demonstrated robust neurite outgrowth activity, while very few clear cell lines harbored neurite outgrowth activity. Given that HGSOCs arise from fallopian tube (FT) secretory cells, we tested sEVs from immortalized FT cell lines and found them lacking in neurite outgrowth activity. However, stable expression of a single oncogene (e.g., Myc, Ras) endows these FT sEVs with robust neurite outgrowth. These findings indicate that acquisition of oncogenic changes may critically contribute to tumor innervation. We have previously demonstrated that TRPV1-expressing sensory nerves innervate HGSOC. Thus, to determine the impact of these tumor-infiltrating nerves on disease progression and survival, p53-/-Pten-/- cells (a model of HGSOC) were implanted into either nociceptor intact (C57Bl/6) or nociceptor neuron ablated (TRPV1cre::DTAfl/wt) female mice. The absence of nociceptor neurons significantly decreased tumor growth but did not improve survival. Given that chemotherapy remains the standard-of-care for ovarian cancer patients, we also tested whether nociceptor neurons influence treatment response by treating tumor-bearing animals with carboplatin. While there was no significant difference on growth and survival in nociceptor intact animals, there was a significant improvement in survival of nociceptor-ablated animals suggesting that tumor-infiltrating nociceptor neurons might contribute to treatment resistance in HGSOC.

# Novel Myc-dependent compound DL78 selectively induces mitotic catastrophe and cell death in high grade serous ovarian cancer

**Jessica Teitel**, Department of Pathology & Rogel Cancer Center, University of Michigan; **Michele Cusato**, Department of Pathology & Rogel Cancer Center, University of Michigan; **Margaret Farah**, Department of Pathology & Rogel Cancer Center, University of Michigan; **Pil Lee**, Ph.D., College of Pharmacy, University of Michigan; **Andy White**, Ph.D., College of Pharmacy, University of Michigan; **Agharnan Gandhi**, Department of Pathology & Rogel Cancer Center, University of Michigan; **John Takyi-Williams**, Ph.D., Pharmacokinetics Core, University of Michigan; **Bo Wen**, Ph.D., Pharmacokinetics Core, University of Michigan; **Andre Monterio Da Rocha**, DVM, Ph.D., Caridiovascular Regeneration Core, University of Michigan; **Analisa DiFeo**, Ph.D., Department of Pathology & Rogel Cancer Center, University of Michigan.

High grade serous cancer (HGSC) is the most common, and most lethal, form of ovarian cancer due to its molecular heterogeneity and lack of preventative screening. As patients succumb to chemoresistance, there is a need to uncover novel targeted drugs. We sought to identify a new therapeutic utilizing a computational screen, named DrugPredict, which led to the discovery that amiodarone, an antiarrhythmic, is a potent apoptosis-inducer and Myc-degrader in numerous patient-derived ovarian cancer cell lines. In search of a more selective, but less toxic compound, structure-activity relationship was applied to identify DL78. Due to its structural differences, DL78 is independent of amiodarone and acts through a unique mechanism. DL78 lacks hERG activity, retains Myc regulation, and gains selective anticancer properties. This compound produces significant cell death selectively in patient-derived HGSC cells, including isogenic cisplatin-resistant lines, but does not harm non-transformed fallopian tube cells. Cancer cells treated with DL78 arrest in prophase due to inhibition of cdc25c. The prolonged arrest leads to loss of Myc, mitotic catastrophe, and apoptosis selectively in the HGSC cells, while fallopian tube cells can cycle normally. DL78's activity is confirmed to be dependent on Myc expression because targeted inhibition of Myc decreases its potency and prevents the DL78-induced cell cycle arrest. A fluorescent-based cellular thermal shift assay confirmed DL78 acts through destabilization of Myc. Finally, pharmacokinetic analysis shows that DL78 specifically infiltrates the tumor within 3 hours, while maintaining low concentration and high clearance in the bloodstream of normal mice. Therefore, we are also investigating DL78 as a theranostic candidate, such that adding a radioactive carbon will allow for tumor diagnosis via PET imaging while simultaneously offering therapeutic benefit. Overall, DL78 is a promising candidate that selectively routes to and kills cancer cells via Myc-dependent mitotic catastrophe.

# CTPS1: an Unexplored Vulnerability in Ovarian Cancer

**Xiyin Wang[1 2], Michael Emch[2], Lauren Voll[2 3], Rebecca Russell[2 4], Esther Rodman[1 2], Nicole Pearson[2], Xiaonan Hou[5], Scott Kaufmann[5], John J. Weroha[5], Philip Beer[6], John Hawse[2]**

[1]Graduate School of Biomedical Sciences, Mayo Clinic, Rochester, MN; [2]Department of Biochemistry and Molecular Biology, Mayo Clinic, Rochester, MN; [3]College of Saint Benedict & Saint John's University, Collegeville, MN; [4]Hillsdale College, Hillsdale, MI; [5]Department of Oncology, Mayo Clinic, Rochester, MN, [6]Step Pharma, Saint-Genis-Pouilly, France

In 2023, an estimated 19,710 new cases of ovarian cancer will be diagnosed with an additional 13,270 deaths directly resulting from this disease. Despite the use of potent cytotoxic chemotherapies and multiple forms of targeted therapies, recurrence is frequent and survival rates for patients with advanced metastatic disease are dismal. These realities highlight the need to better understand drug-resistant forms of ovarian cancer, in order to identify novel biomarkers and therapeutic vulnerabilities and to develop innovative, effective and well-tolerated treatment strategies. We bioinformatically interrogated publically available RNA-seq and genome-wide CRISPR screen datasets and discovered that CTPS1 is highly expressed in many ovarian cancer cell lines and that deletion of this gene results in cancer cell lethality. CTPS1 is one of two enzymes (the other being CTPS2) responsible for the conversion of uridine triphosphate (UTP) to cytidine triphosphate (CTP) which is important for the biosynthesis of phospholipids and nucleic acids. Further interrogation of CTPS1 revealed that it is more highly expressed in tumors relative to respective normal control tissues. siRNA mediated knockdown of CTPS1 inhibited cancer cell proliferation and viability. Using a first-in-class, highly selective and orally bioavailable CTPS1 inhibitor (STP938) developed by Step-Pharma, we identified IC50s in the low nM range for ovarian cancer, including cell models that are resistant to existing standard-of-care therapies. Inhibition of CTPS1 was shown to arrest cells in S phase, and upon longer exposure induced apoptosis. To better understand the mechanisms by which STP938 elicits its potent anti-cancer effects, RNA-Seq was performed on ovarian cancer cell lines. Pathway analysis identified DNA replication and cell cycle as top-ranked biological processes. To extend the investigation of STP938's therapeutic potential from cell-based assays to a more clinically relevant setting, PDX tumors were used to evaluate the efficacy of STP938. We profiled CTPS1 expression in over 400 ovarian PDX models. Our PDX-derived organoids (PDXO) have shown that higher expression of CTPS1 confers sensitivity to STP938 while low expression does not. Based on our initial studies using this drug, we provide pre-clinical evidence of its efficacy in multiple models of primary and advanced ovarian cancer.