AWARD NUMBER: W81XWH-22-1-0852

TITLE: The DOD Omics Consortium to Study the Origins of Ovarian Cancer (DOCSOOC)

PRINCIPAL INVESTIGATOR: Ronny Drapkin, MD, PhD

CONTRACTING ORGANIZATION: University of Pennsylvania, Philadelphia, PA

REPORT DATE: September 2023

TYPE OF REPORT: Annual

PREPARED FOR: U.S. Army Medical Research and Development Command
Fort Detrick, Maryland 21702-5012

REPORT DOCUMENTATION PAGE

Form Approved
OMB No. 0704-0188

1. REPORT DATE	2. REPORT TYPE	3. DATES COVERED
September 2023	Annual	15Aug2022- 14Aug2023

4. TITLE AND SUBTITLE

The DOD Omics Consortium to Study the Origins of Ovarian Cancer (DOCSOOC)

5a. CONTRACT NUMBER
W81XWH-22-1-0852

5b. GRANT NUMBER

5c. PROGRAM ELEMENT NUMBER

6. AUTHOR(S)

Ronny Drapkin, MD, Ph.D.

E-Mail: rdrapkin@pennmedicine.upenn.edu

5d. PROJECT NUMBER

5e. TASK NUMBER

5f. WORK UNIT NUMBER

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)

University of Pennsylvania
421 Curie Blvd
Philadelphia PA, 19104

8. PERFORMING ORGANIZATION REPORT NUMBER

9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES)

U.S. Army Medical Research and Development Command
Fort Detrick, Maryland 21702-5012

10. SPONSOR/MONITOR'S ACRONYM(S)

11. SPONSOR/MONITOR'S REPORT NUMBER(S)

12. DISTRIBUTION / AVAILABILITY STATEMENT

Approved for Public Release; Distribution Unlimited

13. SUPPLEMENTARY NOTES

14. ABSTRACT

In the first year of this award, the DoD Consortium has successfully achieved most of the objectives outlined in aim 1. These include: i) launching monthly leadership calls and quarterly virtual meetings, ii) establishing a consortium website, iii) creating joint MTAs, iv) establishing research specific IRB protocols, v) submission of IRB exempt status for DoD HRPO approval. The DoD Consortium continues work to further progress on the remaining aims. This progress report highlights the Consortium's achievements and continued efforts to comprehensively evaluate cancer precursors for improved early detection.

15. SUBJECT TERMS

None listed.

16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
a. REPORT	b. ABSTRACT	c. THIS PAGE			USAMRDC
Unclassified	Unclassified	Unclassified	Unclassified	17	19b. TELEPHONE NUMBER *(include area code)*

TABLE OF CONTENTS

1. INTRODUCTION:

Ovarian cancer (OvCa) is the fifth leading cause of death for women in the USA with similar rates of occurrence and mortality observed worldwide. Of the various OvCas, high grade serous ovarian carcinoma (HGSC) is the most common. Due to difficulty of detection and nonspecific symptoms it is often caught at later stages, resulting in less effective treatments and little change in survival rates over the past thirty years. Early detection is crucial for successful treatment, yet effective diagnostic tests are lacking, and prevention usually involves surgical measures with significant consequences. Our collaborative effort aims to create a shared biorepository of high-quality STIC specimens, employ advanced tissue characterization technologies, and develop blood or Pap-based tests for early STIC detection. By using genomics to explore the molecular landscapes present in precursor lesions and their surrounding microenvironment, this consortium seeks to develop new and effective screening for early detection strategies.

2. KEYWORDS:

Genomics, ovarian cancer, high grade serous ovarian cancer, microenvironment, early detection, centralized resource, consortium, STIC lesions, stroma, laser capture microscopy, REAL-FAST

3. ACCOMPLISHMENTS:

What were the major goals of the project?

Specific Aim 1: To develop a pathology resource for pan-omic studies of tubal precursors.

Major Task 1: Facilitate communication and collaboration among the DOCSOOC. Create a Consortium website and make it available in the public domain.

- Subtask 1. Initiate monthly leadership calls and quarterly virtual meetings
- Subtask 2. Maintain monthly leadership calls and quarterly meetings.
- Subtask 3. Create a Consortium website
  - Milestone #1: Monthly meetings running successfully and DOCSOOC website available in the public domain soon.

Major Task 2: Obtain consortium regulatory approvals

- Subtask 1: Establish joint MTA among consortium sites
- Subtask 2: Establish Research Specific IRB protocols
- Subtask 3: Submission of IRB exempt status for DoD HRPO Approval
  - Milestone #2: Consortium MTA and IRB protocols are active. IRB/HRPO approval achieved.

Major Task 3: Identity, review, and centralize biospecimens

- Subtask 1: Retrieve incidental STICS and STICs with HGSC 1-6 months
  - All samples (FFPE) will come from active biorepositories at all 3 sites (40 incidental STICs, 40 STICs with HGSC and 60 benign FT controls).
- Subtask 2: Central pathology review of all cases for omics studies 1-36 months
- Subtask 3: Perform LCM to capture epithelial and stroma compartments of incidental STICs, STICs with HGSC, and benign FT epithelium 6-12 months

Subtask 4: Quality control of biomolecules (DNA, RNA, protein) obtained after LCM of all cases for omic studies. 12-48 months

Milestone #3: Biospecimins ready for spatial and omics studies.

We are in the process of completing this milestone, as some biospecimens have undergone spatial and omics studies.

Specific Aim 2. To comprehensively characterize the 'omics landscapes of fallopian tube precursor lesions and associated microenvironment and identify a combined, multi-analyte panel of epithelium and stromal biomarkers for sensitive and specific detection of STIC lesions

Major Task 4: Perform multi-spectral imaging to quantify the presence and spatial distribution of specific stromal and immune populations to STIC lesions

Subtask 1: Stain slides from Aim 1 used the Vectra Automated Quantitative pathology imaging system to identify hrMSCs, mutant p53 containing epithelium (p53), and T cell, B cell, and macrophage populations 6-18 months

Subtask 2: Perform quantitative special transcriptomic and proteomic profiling of epithelium and stroma within and surrounding STIC lesions using the Nanostring GeoMx DSP 12-36 months

Milestone #4: Complete digital library of spatially resolved cell types in and around STIC lesions. Examples to be shared on DOCSOOC website. Submit manuscript 1.

Manuscript 1 is in press. Complete digital library of samples is still in process,

Major Task 5: Perform multi-omic analysis of STIC lesions

Subtask 1: Perform genetic, epigenetic, and miRNA profiling of LCM-captured epithelial and stromal compartments of STIC lesions 6-48 months

Milestone #5: Identification of molecular changes in and around STIC lesions.

Characterization in and around STIC lesions is ongoing. As yet, this milestone has not been reached but we are on schedule.

Major Task 6: Bioinformatic processing and biomarker selection

Subtask 1: Bioinformatic processing and compilation of omics data 12-48 months

Subtask 2: Selection of a combined, multi-analyte (DNA methylation and mutation) panel of epithelium and stromal, STIC-specific biomarker for Aim 3.

Milestone #6: Identification of molecular changes in the epithelial and stromal compartments of STIC lesions. Identification of compartment-specific biomarkers for Aim 3.

Identification of molecular changes in STIC lesions is ongoing. As yet, this milestone has not been reached but we are on schedule.

Specific Aim 3. To detect the STIC-associated markers in tissue proximal fluids and blood.

Major Task 7: Develop multi-analyte test for early detection of STICs

Subtask 1. Prospectively collect incidental STICs with matching pap cytology, and plasma from women at high risk (BRCA1/2 mutation carriers).

This cohort will include FFPE tissues, plasma and Pap specimens obtained from 80 women, including: 20 with incidental STIC, 20 with STIC associated with HGSC patients and 40 normal controls (no detected lesions)

Subtask 2. Develop multi-analyte test by combining Safer-Seq for *TP53* mutation detection and dMSP for the top 10-15 differentially methylated loci identified in STIC-associated epithelium and stroma performed in AIM2.

Subtask 3. Apply test from subtask 2 in a pilot study of pap smear samples and plasma.

Milestone #7: Development of multi-analyte assay based on multi-omic characterization of STIC lesions.

What was accomplished under these goals?

AIM 1/ TASK 1/ Subtask 1: Initiate monthly leadership calls and quarterly virtual meetings.

Calls with the leadership (Drs Drapkin, Coffman, and Wang) and virtual meetings (all team members) have been initiated. During these calls, leadership have the opportunity to discuss project progress, address any emerging challenges, and make strategic decisions. These frequent interactions will ensure the project remains on track, and any issues are promptly identified and resolved. Consortium members will provide updates on their respective tasks and progress, fostering transparency and ensuring that all stakeholders are well-informed about the project's status. These meetings will encourage collaboration and the exchange of ideas among team members. The virtual format will make it easier for this national effort.

AIM 1/ TASK 1/ Subtask 2: Maintain monthly leadership calls and quarterly meetings.

Monthly and quarterly calls are on the calendar. The full team has already met four times to discuss progress and future steps. The format of the virtual meetings usually entail a PowerPoint presentation. By implementing these regular communication channels, we aim to foster a collaborative and informed environment within the consortium, ensuring that all members are aligned with our mission to advance ovarian cancer research. Our commitment to sharing progress with the public domain will contribute to raising awareness and support for our efforts. They will provide a forum for researchers to share insights, address challenges, and explore potential synergies in their work.

AIM 1/ TASK 1/ Subtask 3: Create a Consortium website

A DoD Consortium website is currently accessible at: https://cmsdev1.pmacs.upenn.edu/DOCSOOC/

AIM 1/ TASK 2: Obtain consortium regulatory approvals

Regulatory approvals have been obtained.

AIM 1/ TASK 2/ Subtask 1: Establish Research Specific IRB protocols

IRB approval for collection of human samples has been obtained.

AIM 1/ TASK 2/ Subtask 2: Obtain DoD HRPO approval.

The UPenn site (OHRO Log Number E03195.1a) was approved in September 2022. The University of Pittsburgh site (OHRO Log Number E03195.1b) and the Johns Hopkins site (OHRO Log Number E03195.1c) were approved by OHRO in July and August 2022.

AIM 1/ TASK 2/ Subtask 3: Collect and section FFPE tissues from women with normal FT, STIC containing FT and invasive carcinoma. Timeline 1-18 months.

The Drapkin Lab (site 1) has collected and reviewed 90 FFPE tissues total, 60 from women who underwent risk reducing surgery (30 BRCA1 and 30 BRCA2 mutation carriers) and 30 FFPE tissues from women who underwent surgery for noncancer indications. In addition, 50 cases from women undergoing cytoreductive surgery for ovarian cancer were also obtained. Confirmed incidental STICs and STICs associated with HGSC were sent to sites 2 and 3 for molecular studies. In addition, site 1 has conducted spatial proteomics and transcriptomics to supplement molecular studies.

The Coffman Lab (site 2) has also collected FFPE tissues in addition to those obtained from site 1. They have 5 normal FT, 18 STIC-containing FT, 10 invasive carcinoma, and are continuing to increase their collection.
Wang Lab, (site 3) has collected 5 incidental STICs and 10 STIC-like lesions with HGSC and 15 benign FT controls. Central pathology review is being coordinated with all three sites and quality control is ongoing.

AIM 1/ TASK 3: Identity, review, and centralize biospecimens 1-6 months

Digital images from all the H&Es have been generated and reviewed by the central pathology panel (Drs Drapkin, Shih, Vang, and Schwartz).

AIM 1/ TASK 3/ Subtask 1: Retrieve incidental STICS and STICs with HGSC

The Drapkin Lab (site 1), has collected 8 incidental STICs and 28 STICs with HGSC. Site 2 has collected 7 incidental STICs and 11 STICs with HGSC. Site 3 has collected 5 incidental STICs and 10 STIC-like lesions with HGSC and 15 benign FT controls.

AIM 1/ TASK 3/ Subtask 2: Central pathology review of all cases for omics studies

The central pathology panel (Drs Drapkin, Shih, Vang, and Schwartz) meets virtually on a quarterly basis to review all digital images generated from the H&Es collected for our study.

AIM 1/ Task 3/ Subtask 3: Perform LCM to capture epithelial and stroma compartments of incidental STICs, STICs with HGSC, and benign FT epithelium 6-12 months

Coffman Lab (site 2) has sectioned serial FFPE sections from the samples in subtask 3 onto LCM specific membrane slides. They have performed LCM on 4 normal FT samples thus far and will continue with LCM over the next year.

Wang Lab, (site 3) has collected 5 incidental STICs and 10 STIC-like lesions with HGSC and 15 benign FT controls. Central pathology review is being coordinated with all three sites and quality control is ongoing.

AIM 2/ TASK 4/ Subtask 1: Stain slides from Aim 1 used the Vectra Automated Quantitative pathology imaging system to identify hrMSCs, mutant p53 containing epithelium, and T cell, B cell, and macrophage populations
AIM 2/ TASK 4/ Subtask 2: Perform quantitative special transcriptomic and proteomic profiling of epithelium and stroma within and surrounding STIC lesions using the Nanostring GeoMx DSP 12-36 months

The Coffman Lab (site 2) has performed some multi-spectral imaging and nanostring DSP on all the fallopian tube stroma samples collected in subtask 3. They have quantified the proportion of high risk MSCs and normal MSCs within the stroma of these fallopian tubes, demonstrating STIC contain significantly higher numbers of hrMSCs compared to normal stroma (Appendix A; ***Fig. 1***). The nanostring DSP data is still undergoing analysis. As per the SoW, we do not expect to complete this until 12-36 months so we are currently ahead of schedule.

AIM 2/ TASK 5/ Subtask 1: Perform genetic, epigenetic, and miRNA profiling of LCM-captured epithelial and stromal compartments of STIC lesions 6-48 months

Drapkin Lab (site 1) has sent site 2: 13 STIC samples and 40 benign fallopian tube samples for LCM omic analyses and multi-spectral imaging. Site 1 will continue to send both sites samples as they are obtained. In addition, site 1 has performed cyclic immunofluorescence (CyCIF) on 28 samples (14 STICs and 14 STICs with HGSC).

Coffman Lab (site 2) have sectioned serial FFPE sections from the samples in subtask 3 onto LCM specific membrane slides. They have performed LCM on 4 normal FT samples thus far and will continue with LCM over the course of the next year. DNA methylation analysis will begin once all samples have been processed for LCM. However, they have already piloted this analysis pipeline to ensure appropriate quality control. The Coffman lab, Site 2, has made significant progress in both obtaining samples and performing multi-spectral imaging and DSP on these samples.

Wang Lab (site 3) applied Real-SeqS to assess aneuploidy in laser capture-microdissected fallopian tubal regions comprising p53 signatures, STICs, and normal-appearing fallopian tube epithelium (Appendix B ***Fig. 2A***) (3). REAL-FAST algorithm classifies various types of fallopian tube epithelium into five different Paths on a basis of aneuploidy found in different chromosomal arms. Based on the number of chromosomal arms showing gain or loss, they observed that proliferatively active STICs, as compared to other groups, had the highest numbers of chromosomal arm number changes ($p < 0.05$) (Appendix B ***Fig. 2B***).

Since the purpose of this study is to implement a molecular assay to detect STIC/HGSC in brushed samples that could provide a more sensitive way to detect occult STIC/HGSC, they generated an algorithm called "REAL-FAST" (**Real-SeqS**-based algorithm for **F**allopian tube **A**neuploidy pattern in **ST**IC). REAL-FAST applies an iterative binary decision tree (yes or no) to separate samples into two categories according to their unique aneuploidy patterns. In each category, the algorithm uses another decision tree to further separate cases according to their aneuploidy patterns (high-level gain or amplification vs. deletion). REAL-FAST allows an unsupervised profiling agnostic of pathology diagnoses. Five distinct "Paths" (clusters or groups) of fallopian tube epithelium and lesions,

designated Path-1 to Path-5 (Appendix C ***Fig. 3A***). Matching the 5 Paths to pathology diagnoses revealed that Path-2, Path-3, and Path-4 mainly corresponded to STICs, Path-5 to p53 signatures, and Path-1 to normal fallopian tube epithelium regions (Appendix C ***Fig. 3A***). They found that HGSCs grouped within either Path-2 or Path-3. They then compared molecular diagnosis (from Path-1 to Path-5) to their proliferative activity and growth pattern. Regarding the growth pattern on tissues, they assessed the presence or absence of the loosely adherent or detached STIC on the mucosal surface because they are most pertinent to early dissemination of STIC lesions to peritoneal cavity. The highest Ki-67 labeling index was in Path-2, followed by Path-3 and Path-4; Path-5 and Path-1 were similar to background level (Appendix C ***Fig. 3B***). Their new data shows both Path-2 and Path-3, as compared to other groups, had significantly more cases with a dis-cohesive pattern (Appendix C ***Fig. 3C, D***). These findings were recently accepted for publication in *Clinical Cancer Research*.

AIM 2/ TASK 6/ Subtask 1: Bioinformatic processing and compilation of omics data 12-48 months

Data is actively being compiled and as per the statement of work we expect to complete the bioinformatic processing in 12-48 months and we are on schedule.

AIM 3/ TASK 7/ Subtask 1: Prospectively collect incidental STICs with matching pap cytology, and plasma from women at high risk (*BRCA1/2* mutation carriers).

Drapkin Lab (site 1) and Coffman Lab (site 2) have an IRB protocol pending to allow for prospective collection of matched PAP cytology and plasma from women at high risk (*BRCA1/2* mutation carriers).

Wang lab (site 3) has an active protocol and is already collecting prospective samples.

AIM 3/ TASK 7/ Subtask 2: Develop multi-analyte test by combining Safer-Seq for *TP53* mutation detection and dMSP for the top 10-15 differentially methylated loci identified in STIC-associated epithelium and stroma performed in AIM2.

Work on this will begin once samples are collected.

AIM 3/ TASK 7/ Subtask 3: Apply test from subtask 2 in a pilot study of pap smear samples and plasma.

Work on this will begin once samples are collected.

What opportunities for training and professional development has the project provided?

Nothing to report.

How were the results disseminated to communities of interest?
As per DoD policy, our results will be disseminated via publications, our website, and open-source software. In addition, our patient advocate, Sachia Powell, meets with us regularly to provide prospective on impact. Moreover, we have partnered with the Powell-Drescher foundation to promote patient advocacy at national meetings and share the work of the DoD omics consortium with a broader public and patient advocacy community. For instance, the Powell-Drescher foundation recently sent

2 patient advocate trainees to the DoD Ovarian Cancer Research Academy annual retreat Boston (Oct 3-Oct 4, 2023). At this meeting, the advocates not only engaged with the early career investigators but spoke to them about the importance of early detection, collaboration, and the need to develop a deep understanding of cancer precursors, the focus of the DoD omics consortium.

What do you plan to do during the next reporting period to accomplish the goals?

Collection is going according to schedule, and we will continue to collect, process, and characterize these samples with the goals of developing a comprehensive understanding of precursors and developing a multi-analyte test for early detection of STICs.

4. IMPACT:

What was the impact on the development of the principal discipline(s) of the project?

While still in the early days, researchers involved in this initiative have expressed a sense of significant progress and forward momentum during the virtual meetings. The collaborative environment has proven to be instrumental in fostering innovative thinking and problem-solving. In Major Task 1, we have established ongoing communication with everyone on the DOCSOOC team and are developing a Consortium website to be made accessible to the public. In Major Task 2 we have obtained regulatory approvals for all sites to ensure the rights and privacy of patient specimens are preserved. In Major Task 3 we have begun work on centralizing biospecimens and readying them for identification and review between all sides. The remaining Tasks, 4-7, are in process with some spectral imaging and multi-omic analysis already completed for some specimens. "Aneuploidy landscape in precursors of ovarian cancer" is in press at *Clinical Cancer Research* and "Ultrasensitive detection of circulating LINE-1 ORF1p as a specific multi-cancer biomarker" is available on epub ahead of print at *Cancer Discovery* (see PRODUCTS below).

What was the impact on other disciplines?

While it may be too early to assess the direct impact on other disciplines, our initiative has the potential to serve as a model for addressing similar challenges in cancer research more broadly. There is growing interest for the establishment of consortiums dedicated to creating a pre-cancer atlas specifically tailored to ovarian cancer with groups like the Break Through Cancer Foundation, the Gray Foundation, and the Canary Foundation.

What was the impact on technology transfer?

Nothing to report.

What was the impact on society beyond science and technology?

Through initiatives like our consortium website, we aim to bridge the gap between scientific research and the broader community, ultimately paving the way for a deeper societal understanding of ovarian

cancer and its complexities. Though the impact is still in its infancy, we anticipate that our work will have far-reaching implications, leading to improved awareness, early detection, and treatment strategies. By including a patient advocate through all steps of our consortium, our collaboration extends not only to advancing science but also to building trust with patient advocates and the wider public, ultimately benefiting ovarian cancer patients and paving the way forward for a more inclusive, cooperative, and impactful community of practitioners and researchers.

5. CHANGES/PROBLEMS:

Changes in approach and reasons for change

No changes to report.

Actual or anticipated problems or delays and actions or plans to resolve them

Nothing to report.

Changes that had a significant impact on expenditures

Nothing to report.

Significant changes in use or care of human subjects, vertebrate animals, biohazards, and/or select agents

Nothing to report.

Significant changes in use or care of human subjects

Nothing to report.

Significant changes in use or care of vertebrate animals

Nothing to report.

Significant changes in use of biohazards and/or select agents

Nothing to report.

6. PRODUCTS:

- **Publications, conference papers, and presentations**

  Wang Y, Douville C, Chien YW, Wang BG, Chen CL, Pinto A, Smith, SA, Murdock T, **Drapkin R**, Vang R, Chui MH, Numan T, Vang R, Papadopoulos N, **Wang TL**, **Shih IM**. Aneuploidy landscape in precursors of ovarian cancer. Clin Cancer Res, in press, 2023.

Taylor MS, Wu C, Fridy PC, Zhang SJ, Senussi Y, Wolters JC, Cajuso T, Cheng WC, Heaps JD, Miller BD, Mori K, Cohen L, Jiang H, Molloy KR, Chait BT, Goggins MG, Bhan I, Franses JW, Yang X, Taplin ME, Wang X, Christiani DC, Johnson BE, Meyerson M, Uppaluri R, Egloff AM, Denault EN, Spring LM, **Wang TL**, **Shih IM**, Fairman JE, Jung E, Arora KS, Yilmaz OH, Cohen S, Sharova T, Chi G, Norden BL, Song Y, Nieman LT, Pappas L, Parikh AR, Strickland MR, Corcoran RB, Mustelin T, Eng G, Yilmaz OH, Matulonis UA, Skates SJ, Rueda BR, **Drapkin R**, Klempner SJ, Deshpande V, Ting DT, Rout MP, LaCava J, Walt DR, Burns KH. Ultrasensitive detection of circulating LINE-1 ORF1p as a specific multi-cancer biomarker. Cancer Discov. 2023 Sep 12. doi: 10.1158/2159-8290.CD-23-0313. Epub ahead of print. PMID: 37698949.

All the PIs have presented at national and international conferences on the subject of their consortium work.

- **Website(s) or other Internet site(s)**

The DoD Consortium website is currently in development and can be found at: https://cmsdev1.pmacs.upenn.edu/DOCSOOC/team.html. This website was updated during the past month and continues to be monitored on a regular basis. Once it goes live it will serve the following purposes: 1) To spread information about the consortium to the public and 2) to be used as a source of communication for members of the consortium itself. The intention of this website is to contain general information about the consortium's goals and progress.

- **Technologies or techniques**

Nothing to report.

- **Inventions, patent applications, and/or licenses**

Not currently.

- **Other Products**

Nothing to report.

7. PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS

What individuals have worked on the project?

Name	Project Role	Contribution to Project	University
Sachia Powell	Consumer Advocate	Helps guide communications between scientists, consumer groups and the larger ovarian cancer research community	-
Ronny Drapkin	Director, Site 1 Principal Investigator	In charge of overall operations and oversees the completion of the proposed project tasks. He manages the administrative aspects of DOCSOOC, including finalization of all study-related documents, submission and tracking of regulatory approvals, and budget management. Coordinates efforts with Dr Schwartz and the BioTrust collection (Euihye Jung and Natalie Shih).	UPenn
Tian-Li Wang	Site 3 Principal Investigator	Serves as site PI at Hopkins and coordinates the efforts of Drs. Shih, Cope, and Pisanic with Dr. Drapkin and the Pitt investigators	Hopkins
Len Coffman	Site 2 Principal Investigator	Responsible for experimental design and data interpretation pertaining to evaluation of the stromal TME. Oversees the proposed experiments and prepares manuscripts and presentations generated from this work.	UPitt
Thomas Pisanic	Research Scientist	Leads and provides oversight in the selection of STIC-specific genetic and epigenetic biomarkers, as well as the development of targeted assays for their detection	UPitt
Ie-Ming Singh	Co-investigator	Performs the clinic pathological characterization of clinical specimens, and takes an active role in the interpretation, characterization, and prioritization of the discovery pipelines. He serves as the site Co-PI and meets frequently with other leaders of this proposal to ensure successful collaboration and timely report of the research accomplishments.	Hopkins
Lauren Schwartz	Co-investigator	Coordinates procurement of specimens and serves on the central pathology review panel with Drs. Drapkin and Shih.	UPenn
Ronald J. Buckanovich	Co-investigator	Works closely with Dr. Coffman to assist in the experiments pertaining to evaluating the stromal TME.	UPitt
Francesmary Modugna	Co-investigator	Provides expertise on the public health aspects of the project and to support the tissue needs of the program. Also coordinates specimen procurement at Pitt and works with Drs. Drapkin, Schwartz, and Shih to meet consortium specimen needs	UPitt
Leslie Cope	Bioinformatics Core co-investigator	Supervises analysis of 'omics datasets generated by DOCSOOC and directs the Bioinformatics Core.	Hopkins

Has there been a change in the active other support of the PD/PI(s) or senior/key personnel since the last reporting period?

Nothing to report.

What other organizations were involved as partners?

Nothing to report.

8. SPECIAL REPORTING REQUIREMENTS

COLLABORATIVE AWARDS: Nothing to report.

QUAD CHARTS: Nothing to report.

Nothing to report.

9. APPENDICES:

The following items are contained in the Appendix, in support of this Progress Report:

A. Figure from Coffman Lab (site 2) Characterizing hrMSC Distribution in Fallopian Tubes with STIC Lesions

B. Figure from Wang Lab (site 3) Lesion Characterization through H&E, IHC, and Aneuploidy Profiling

C. Figure from Wang Lab (site 3) REAL-FAST Algorithm Identifies Key Pathways for STIC Lesions and HGSC Detection

Appendix A: Figure 1 from Coffman Lab (Site 2) Characterizing hrMSC Distribution in Fallopian Tubes with STIC Lesions

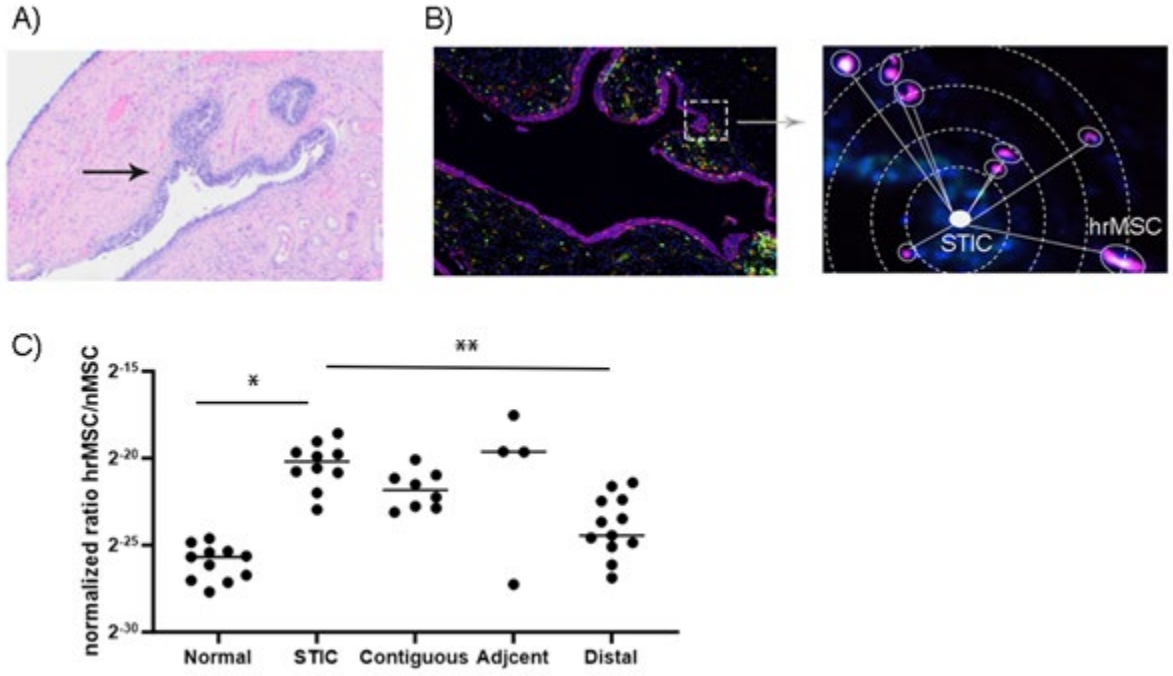


Figure 1. Analysis of hrMSCs within fallopian tubes. A) H&E of fallopian tube demonstrating a STIC (black arrow). B) Vectra imaging of a fallopian tube with a STIC lesion and spatial quantification of hrMSCs surrounding the STIC (second panel). C) Quantification of the ratio of hrMSC to normal MSCs in regions surrounding STIC or normal fallopian tube epithelium (normal: stroma in normal fallopian tube. STIC: stroma underlying STIC regions. Contiguous: stroma underlying 20 cells surrounding STIC regions. Adjacent: stroma underlying cells <2mm from STIC lesion. Distal: stroma underlying cells <2cm from STIC region.

Appendix B Figure 2 from Wang Lab (Site 3) Lesion Characterization through H&E, IHC, and Aneuploidy Profiling

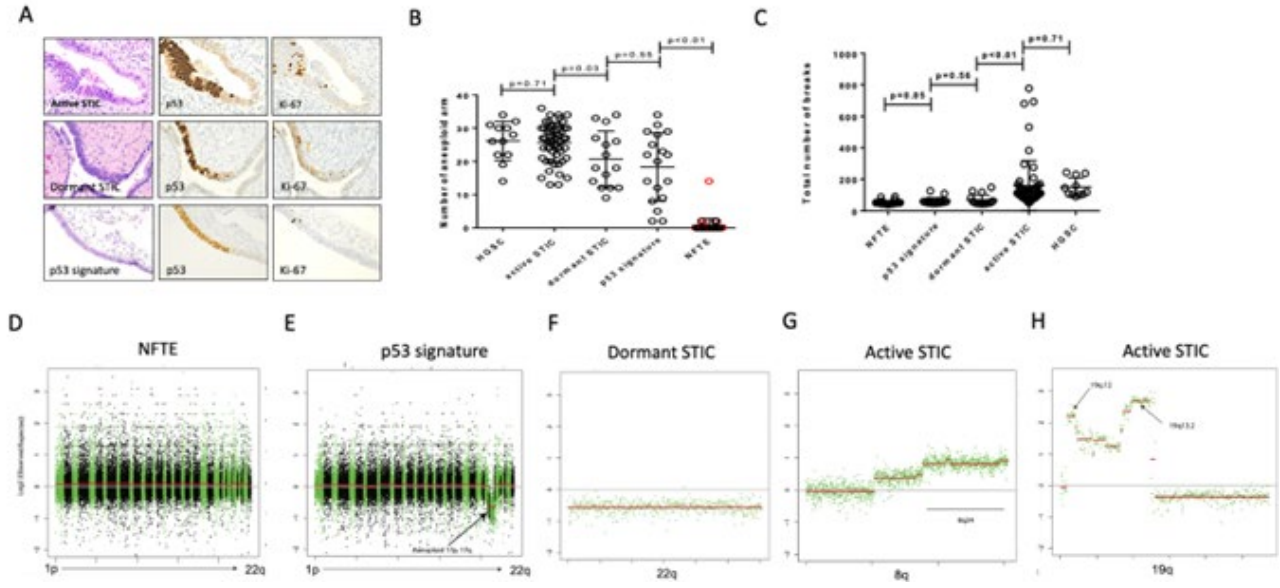
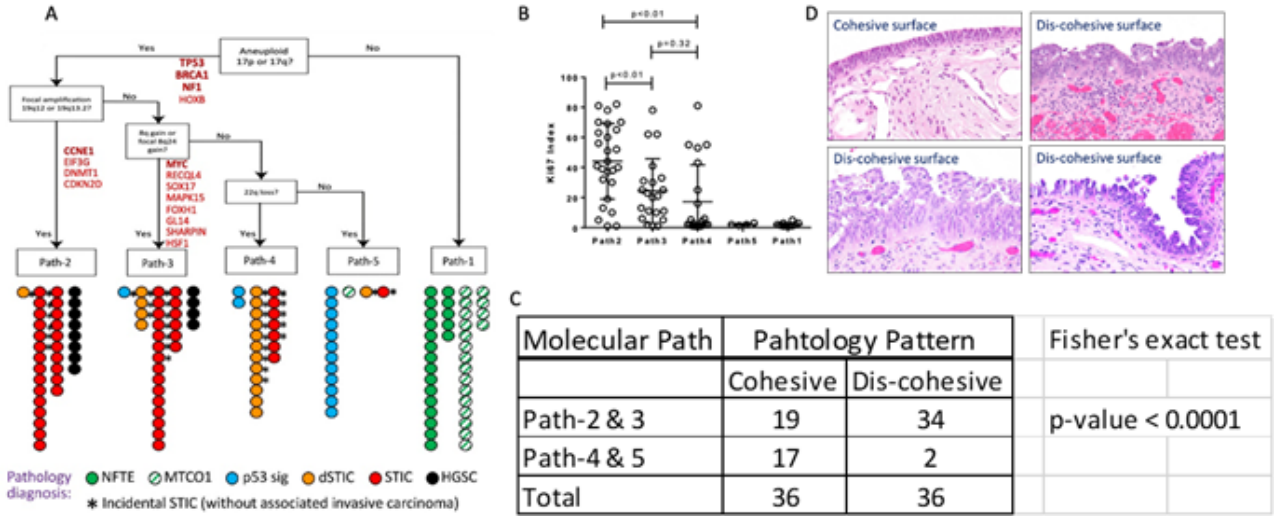


Fig. 2. Representative H&E and immunohistochemistry (IHC) images and aneuploidy profiles of the lesions studied. (A) Images of H&E, p53, and ki-67 IHC of active STIC (top), dormant STIC (middle), and p53 signature (bottom). (B) Scatter plot of the number of aneuploid arms in different lesions. (C) Scatter plot showing total number of chromosomal break points. (D) Representative image of Real-SeqS data on normal fallopian tube epithelium (NFTE). (E) Real-SeqS data on a p53 signature. The arrow indicates loss of both chr17 p-arm and q-arm in p53 signatures. (F) Real-SeqS data showing additional chr22q loss in dormant STIC compared to p53 signature. (G, H) Representative Real-SeqS showing additional chr8q24, chr19q12, and chr19q13.2 gains in active STIC compared to dormant STIC.

Appendix C: Figure 3 from Wang Lab (Site 3) REAL-FAST Algorithm Identifies Key Pathways for STIC Lesions and HGSC Detection



C

Molecular Path	Pahtology Pattern		Fisher's exact test
	Cohesive	Dis-cohesive	
Path-2 & 3	19	34	p-value < 0.0001
Path-4 & 5	17	2	
Total	36	36	

Fig.3. Aneuploidy-based algorithm for the diagnosis of STIC lesions, high-grade serous carcinoma (HGSC) and normal-appearing fallopian tube epithelium (NFTE). (A) REAL-FAST algorithm classifies various types of fallopian tube epithelium into five different Paths on a basis of aneuploidy found in different chromosomal arms. The individual specimens (filled circles) are colored according to their pathology diagnoses. Cancer-associated genes with copy number changes at each decision point are indicated. Bolding indicates the most relevant to ovarian cancer. (B) Proliferative activity (Ki-67 labeling index) among different molecular groups. (C) Dis-cohesive growth characterized by detached STIC cells is more common in Path-2 (*CCNE1* amplified) or Path-2/Path-3 STICs than Path-4. (D) Representative photomicrographs (H&E) from four different STICs showing either dis-cohesive or cohesive growth.