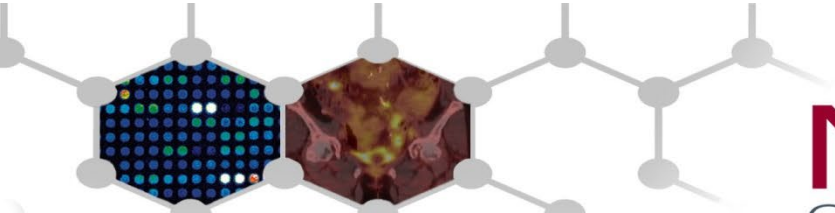

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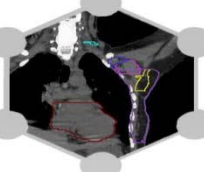

NRG Gyn Translational Committee Ovarian

July 2025

Translational Research Co-Chairs:

Rebecca C. Arend, MD, MSPH

Maggie Mullen, MD, MSCI

Disclosures

- **Arend:**
  - Merck, Abbvie, GSK, Excelixis – IITs
  - Natera, Tempus, CARIS, LifeNet, Cerflux, Artera - collaborative grants
  - AZ - consulting

- **Mullen:**
  None

Some of the presentations and discussions may include the off-label use of products/devices.

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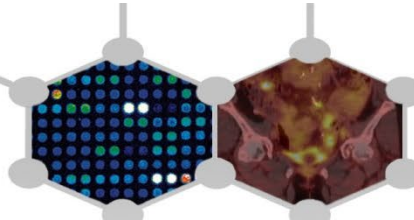

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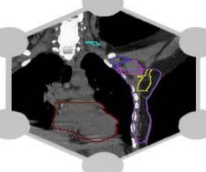


ACTIVE STUDIES

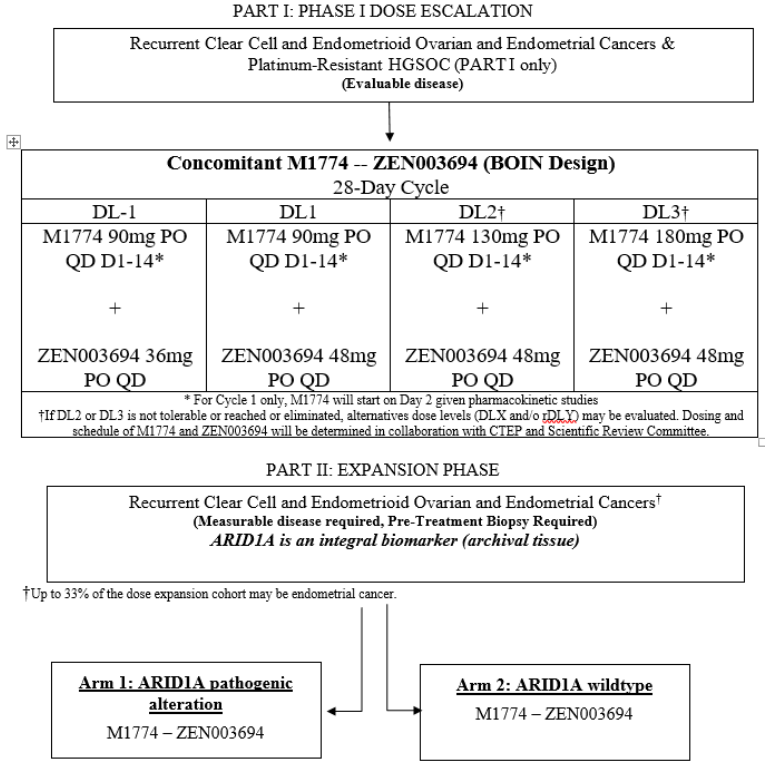
GY031 A Phase IB study of combination ATR (M1774) and BET inhibition (ZEN-3694) to exploit ARID1A loss in recurrent ovarian and endometrial cancer (Simpkins).

Integral Biomarkers: *ARID1A* mutation (local; Part II), γH2AX (BRC approved)

Integrated Biomarkers: ARID1A protein (Part II), *ARID1A* mutation (central; Part II), proteomics (Part II)

Exploratory Biomarkers: *ARID1A* mutation (local; Part I), ARID1A protein (Part I), PK (2/16/24 SC notified required by FDA for Part I analysis), *HEXIM1* (Part II), *ARID1A* mutation in cfDNA (Part II)

***Approved and funded by R01.**



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GY036 One vs. Two Years of Maintenance Olaparib, with or without Bevacizumab, in Patients with BRCA1/2 Mutated or Homologous Recombination Deficient (HRD+) Ovarian Cancer Following Response to First Line Platinum Based Chemotherapy (Ying Liu; TR Swisher)

Integrated Biomarkers: Methylation (pending approval)

Exploratory Biomarkers: Clonal hematopoiesis (pending approval)

*** Will require additional funds for TR work.**

Further work necessary to define translational biomarkers

Collecting: Blood samples- baseline, 1m, 2m, progression or EOT.
Tissue- archival

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COMBOMATCH

EAY191-A3, Palbociclib and Binimetinib in RAS-Mutant Cancers: A ComboMATCH Treatment Trial (Alliance: Geoffrey Shapiro, MD, PhD, Joyce Liu, MD, Ardaman Shergill, MD) Accrual: 43/199

- Cohort 1: Low Grade Serous Ovarian Cancer (LGSOC), MEK inhibitor naïve (KRAS/NRAS/HRAS, nonBRAF V600E actionable marker of interest or rare RAF fusions) Closed to Accrual as of 04/01/2025
- Cohort 2: Low Grade Serous Ovarian Cancer (LGSOC), progressed on prior MEK inhibitor (not genomically selected) Open
- Cohort 3: Pancreatic Cancer harboring any KRAS/NRAS/HRAS, non-BRAF V600E actionable marker of interest or rare RAF fusions. Temporarily Closed to Accrual as of 07/03/2024
- Cohort 4: Tumor agnostic harboring any KRAS/NRAS/HRAS, nonBRAF V600E actionable marker of interest or rare RAF fusions, excluding LGSOC, NSCLC, colorectal cancer, pancreatic, and melanoma. Temporarily Closed to Accrual as of 09/05/2024

EAY191-S3, Phase 2 Study of Paclitaxel (NSC #673089) + Ipatasertib (NSC #781451) in Taxane-Refractory Participants with AKT-Altered Advanced Non-Breast Solid Tumors: A ComboMATCH Treatment Trial (SWOG: Reva Basho, MD, Haider Mahdi, MD, MPH) Accrual: 22/33

-Excludes mutations in KRAS, NRAS, HRAS, BRAF

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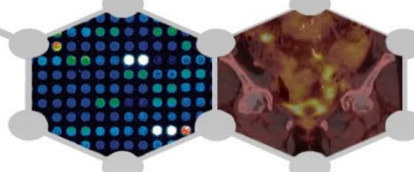

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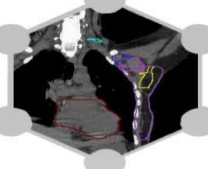


CLOSED STUDIES

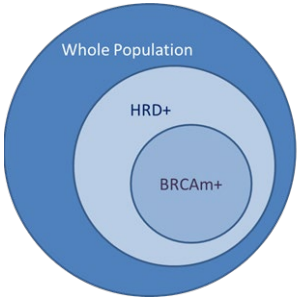
Closed Trial

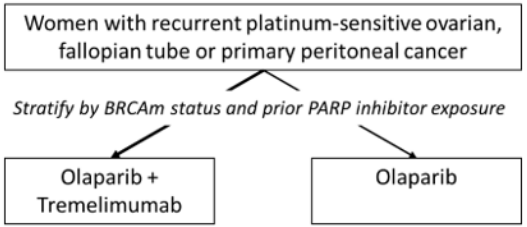
NRG GY021 (NCT04034927)

RPh2: Olaparib-Tremelimumab vs Olaparib (TR Adams)

(Group-wide)

- Recurrent HGSC with PFI > 6 months (following most recent platinum)
- No more than 3 prior regimens (including primary therapy)
- RECIST measurable disease
- No prior PARPi therapy (except primary maintenance). No prior immune checkpoint inhibitors
- Primary endpoint: PFS (pre-specified sequential analysis by whole population and subgroups)





- **TME** (leukocyte infiltrate or gene expression signature in archived tumor samples).
- **Blood based** (the impact of olaparib and tremelimumab versus olaparib monotherapy on circulating leukocyte subsets and cytokine/chemokine levels; cfDNA *BRCA* mutation status)
- Summary:
  - TMB does not appear to correlate with response to O+T
  - Unexpected association of macrophage infiltration with outcomes
  - TCR data suggest a polyclonal response in subjects receiving O+T
  - Marked differences in blood-based leukocyte subsets and inflammatory markers by treatment arm
  - Potential circulating biomarkers need further study

Adams S for NRG Oncology

NRG-GY009 A Randomized, Phase II/III Study of Pegylated Liposomal Doxorubicin and CTEP-Supplied Atezolizumab (IND #134427) Versus Pegylated Liposomal Doxorubicin, CTEP-Supplied Bevacizumab and CTEP-Supplied Atezolizumab Versus Pegylated Liposomal Doxorubicin and CTEP-Supplied Bevacizumab in Platinum Resistant Ovarian Cancer (Roisin O'Cearbhaill; TR Zamarin).

Presented at IGCS 2024

International Journal of Gynecological Cancer
Volume 34, Supplement 3, October 2024, Page A10

Late-Breaking Abstracts

LB005/#1605 Randomized, phase II/III study of pegylated liposomal doxorubicin, bevacizumab, and atezolizumab in platinum-resistant ovarian cancer (NRG-GY009): clinical outcomes by PD-L1 and T cell infiltration status

Roisin O'Cearbhaill [1], Dmitriy Zamarin [2], Michael Sill [3], Hoa Duong [4], Steven Waggoner [5], Rachel Grisham [1], Floor Backes [6], Robert Mannel [7], Janos Tanyi [8], Matthew Powell [9], Cara Mathews [10], Sharad Ghamande [11], Leah McNally [12], Alexander Olawaiye [13], David Bender [14], Linda Duska [15], Heather Lankes [16], Russell Schilder [17], Michael Bookman [18], Carol Aghajanian [1]

Conclusion/Implications

In this exploratory analysis, patients with PD-L1<1% tumors demonstrated improved PFS/OS when treated with PLD/BEV/ATEZO when compared to PLD/BEV, highlighting the unreliability of PD-L1 as a biomarker in PROC. Identification of immune biomarkers relevant to PROC remains an unmet need.

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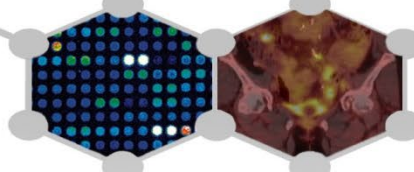

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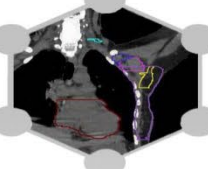


NAVIGATOR STUDIES

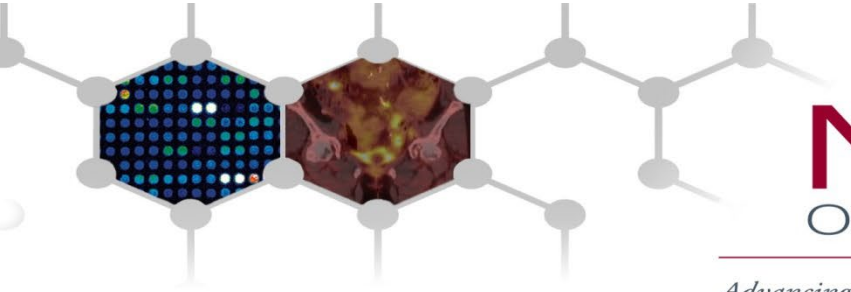
Navigator Ovarian Cancer Efforts

- AI-Based Markers of Response in Ovarian Cancer using clinical specimens from GOG-0213, GOG-0252 and NRG-GY004 (*Bazzara University of Minnesota*).
    - *LOI Approved. Full proposal pending.*
- Validation of a protein signature predictive of high grade serous ovarian which persists or progresses during initial therapy using NCTN specimens GOG218,182,252,262,GY003 *(Birrer University of Arkansas Medical School)*
    - *Revisions requested by CCSC*
- Evaluation of SERPINE1 as a biomarker for MEK inhibitor response in GOG-0281 *(Wong MD Anderson Cancer Center)*
    - *LOI Approved. Full proposal pending.*

Navigator – Ovarian Cancer Opportunities

Collection timepoints for the trials that do have plasma –

GOG 213: Optional prior to cytoreductive surgery
GOG 262: Prior to initiating front-line chemotherapy
GOG 273: For PK (C1D1 pre-Rx → 1, 6, 24hr post-paclitaxel)
GOG 281: Pre-Rx, C3D1
NRG-GY004: For PK (C1D8 pre-Rx → 0.5-1, 1-3, 4-6 hr post-study drug)
GOG 218: Optional prior to initiating front-line chemotherapy



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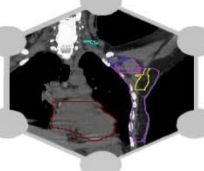

NRG Gyn Translational Committee
Uterine Corpus

July 25, 2025

Translational Research Co-Chairs:

Victoria Bae-Jump, MD, PhD

Katherine Fuh, MD, PhD

Disclosures

- **Bae-Jump:**
  - Merck – collaborative grants
  - Chimerix, Genentech – drugs for pre-clinical studies, collaborative grants
  - Gilead – DSMB member

- **Fuh:**
  - Merck – collaborative grants
  - Aravive, Genentech – drugs for pre-clinical studies, collaborative grants
  - Immunogen, GSK, AstraZeneca – advisory boards

GY012 A Randomized, Phase II Study Comparing Single-Agent Olaparib, Single Agent Cediranib, and the Combination of Cediranib/Olaparib in Women with Recurrent, Persistent or Metastatic Endometrial Cancer. PI: Helen Mackay

<u>Integrated Biomarkers:</u> plasma angiome (Nixon) and HRD (Swisher) include HRD (LOH/BRCA data) and p53, Abstract presented at ASCO 2023 – manuscript in prep.

- To assess if markers of angiogenesis in serial plasma samples are associated with response to cediranib alone or in combination with olaparib. (Integrated Biomarker) – pre-tx, after cycle #1 and recurrence
- To assess if mutations in DNA Homologous Repair Genes (assayed prior to all treatment and prior to the study treatment) are predictive of response to olaparib alone or in combination with cediranib. (Integrated Biomarker)
- ****Paper published: Rimel BJ, Enserro D, Bender D, Gunderson C, Tan A, Alluri N, Borowsky M, Moroney J, Hendrickson A, Backes F, Swisher E, Powell M, MacKay H. NRG-GY012: Randomized phase 2 study comparing olaparib, cediranib, and the combination of cediranib/olaparib in women with recurrent, persistent, or metastatic endometrial cancer. Cancer 2023 Dec 21.*

GY012 A Randomized, Phase II Study Comparing Single-Agent Olaparib, Single Agent Cediranib, and the Combination of Cediranib/Olaparib in Women with Recurrent, Persistent or Metastatic Endometrial Cancer. PI: Helen Mackay

Neoplastic and Blood-Based Biomarkers of Response in Patients with Advanced Endometrial Cancer: Results from NRG GY012 (Abstract #5525)

HJ MacKay, AB Nixon, D Enserro, D Bender, BJ Rimel, MA Powell, DL Richardson, LL Holman, N Alluri, CA Mathews, R Garg, S Gill, D Matei, CA Leath III, FJ Backes, A Fleury, EM Swisher

Odette Cancer Centre, Sunnybrook Health Sciences Centre, Toronto, ON; Duke University Medical Center, Durham, NC; NRG Oncology, Buffalo, NY; University of Iowa Hospitals and Clinics, Iowa City, IA; Cedars-Sinai Medical Center, Los Angeles, CA; Washington University, St Louis, MO; Stephenson Cancer Center/University of Oklahoma Health Sciences Center and Sarah Cannon Research Institute, Oklahoma City, OK; University of Oklahoma Health Sciences Center, Oklahoma City, OK; Pacific Cancer Research Consortium NCORP/St. Luke's Cancer Institute, Boise, ID; Women and Infants Hospital of Rhode Island, Providence, RI; City of Hope, Atlanta, GA; Nancy N and J C Lewis Cancer and Research Pavilion, Savannah, GA; Northwestern University, Chicago, IL; University of Alabama at Birmingham, Birmingham, AL; Ohio State University James Cancer Hospital Department of Radiation Oncology, Columbus, OH; Women's Cancer Centre of Nevada, Las Vegas, NV; University of Washington School of Medicine, Seattle, WA

Presented at ASCO 2023.

Conclusions

- LOH high status only occurred in cancers with TP53 mutation and was not associated with response to Olaparib.
- The number of HRR pathogenic variants was too low to determine their relationship to PARPi response.
- IL6 was not predictive for PFS or OS using the pre-established cut-point from GOG-218. Although a trend was seen with the median level established in GY018.
- PIGF is a putative predictor of benefit from cediranib.

Manuscript in prep from this ASCO presentation.
Second cohort submitted to ESMO.

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GY018 A PHASE III RANDOMIZED, PLACEBO-CONTROLLED STUDY OF PEMBROLIZUMAB (MK-3475, NSC #776864) IN ADDITION TO PACLITAXEL AND CARBOPLATIN FOR MEASURABLE STAGE III OR IVA, STAGE IVB OR RECURRENT ENDOMETRIAL CANCER. PI: Ramez Eskander

- TR work has been approved by CTEP/NCI - specimen acquisition is underway – TR work to be done at MSKCC with Carol Aghajanian as PI.
    - MSK Impact and IHC and the report the PFS/OS by TCGA group – data in early 2026.
- Current TR has shown that MMR loss was not a significant predictor of PFS (85% versus 75% progression free at 12 months in tumors with MMR mutation vs methylation).
- FDA approval – June 2024

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GY020 A Phase III Randomized Trial of Radiation +/- Pembrolizumab for Newly Diagnosed, Early-Stage HIR Mismatch Repair Deficient (dMMR) Endometrioid Endometrial Cancer. PI: Backes, TR PI: Zamarin

Last patient completed treatment on 10/2023 **Awaiting for primary endpoint to mature

Integral Biomarkers: MMR by IHC and/or microsatellite instability (MSI) testing

Integrated Biomarkers:

- T cell receptor (TCR) sequencing (Zamarin): FFPE tumor (to determine which TCR clones are present within the tumor), as well as pre-cycles 1 and 2 whole blood biospecimens, will be used for TCR sequencing.
- CyTOF (Cytometry of Time Of Flight) Fuh: Analysis of Immune Cell Populations: Pre-cycles 1 and 2 whole blood biospecimens will be used.

Exploratory Biomarkers: Awaiting for primary endpoint to mature to request specimens

- **ctDNA** Collection of baseline and on-treatment ctDNA. ctDNA at time of progression added - Winter 2024. (Signatera – working on contract with Natera).
- ****Working through a contract for ctDNA analysis and data sharing with Natera. Plan to initiate the studies before the clinical outcomes data become available. Will not be able to link the results to the outcome.**

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GY025 A RANDOMIZED PHASE II TRIAL OF NIVOLUMAB AND IPILIMUMAB COMPARED TO NIVOLUMAB MONOTHERAPY IN PATIENTS WITH DEFICIENT MISMATCH REPAIR SYSTEM RECURRENT ENDOMETRIAL CARCINOMA

PI: Haider Mahdi

Tissue Based:

- PDL1 expression (tumor and immune cells)
- RNA sequencing including IFN-g signature and other immune signature
- DNA sequencing and TMB and MSI
- MHC/HLA typing and B2M mutation/expression

Blood Based:

- PBMC: immunophenotypic characterization (C1D1, C2D1)
- Cytokines like TGF-B and other cytokines (C1D1, C2D1)
- TGF-beta signature and cytokine level
- ctDNA: bTMB, bTM (C1D1, C2D1, EOT/PD)
- Blood (PBMC and plasma) for immune monitoring based and CtDNA based aims as well as archival tissue for tissue based aims

NRG ONCOLOGY™

GY025 A RANDOMIZED PHASE II TRIAL OF NIVOLUMAB AND IPILIMUMAB COMPARED TO NIVOLUMAB MONOTHERAPY IN PATIENTS WITH DEFICIENT MISMATCH REPAIR SYSTEM RECURRENT ENDOMETRIAL CARCINOMA - PI: Haider Mahdi

- Integral – local MMR +/- MSI
- Close to 30 patients enrolled, safety lead in for combination arm.
- Funding from DOD to cover translational aim and preclinical aim in MSI-H endometrial cancer
- Working on TR amendment
- CTEP approved collecting and banking all samples (tissue and blood) needed for the TR plan – trial enrolling and samples being collected.

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GY026 (UC1972) A randomized phase III trial of paclitaxel/carboplatin alone or combined with either trastuzumab or trastuzumab/pertuzumab in HER2 Positive Stage I-IV uterine carcinoma (PIs: Erickson, Fader, Santin, Havrilesky QOL)

- Accrual is X pts of 326 (as of X) - redesign as now a phase III study, PFS as primary endpoint.
- No open to ALL STAGES (myoinvasive stage 1-4)
- Now open to ALL HISTOLOGIC SUBTYPES (not limited to USC and Carcinosarcoma)
- Integral biomarker is HER2 positive by local testing (or NGS).
- FFPE tumor blocks are required for each subject – planned central HER2 tsting; interested in the development of translational objectives regarding race/ethnicity and association with outcomes and tumor testing.
- Additional optional biospecimen collection including whole blood in Streck tubes and urine – ctDNA studies planned, pending NCI approval.

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NRG-GY028 (PI2129) Phase IB and Randomized Phase II trial of medroxyprogesterone acetate +/- ipatasertib in recurrent/metastatic endometrioid endometrial cancer (G1/2). PIs: Onstad, Westin

- Activated on 1/13/23 – Enrollment complete!!
- Biospecimens: tumor blocks, whole blood

Integrated Biomarkers:

- PTEN loss (IHC, Biomarker Review Committee approved)
- ER and PR status (IHC)
- Alterations in the PI3K/Akt/mTOR pathway (Whole Genome Sequencing/RNA sequencing, National Clinical Laboratory Network (NCLN) approved)
- Ipatasertib/Active Metabolite Pharmacokinetics (Liquid chromatography-tandem mass spectrometry)

Exploratory Biomarkers:

- pS6/total S6, pPRAS40/total PRAS40 (IHC)

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GOG210/Navigator EC Profiling Efforts

- GOG-210 NCI Cancer Moonshot MP2PRT Submission (Cosgrove)–ongoing, WGS/RNAseq, racial disparities focus
    - Sequencing completed. Bioinformatic analysis ongoing.
    - Data analysis underway – submit manuscript in 9/2025.
- GOG286B (Bae-Jump/Maxwell/Conrads): – proteomics of tumor tissue (sent to Inova and ongoing), metabolomics of blood (underwent analysis at Metabolon), awaiting clinical data after manuscript accepted (DSA submitted).
- GOG258/Molecular subtyping (modified ProMisE algorithm): OS data analysis presented at SGO. PFS data presented at IGCS 2023. Manuscript accepted at Gynecologic Oncology (Clements, Alvarez Secord).
- GOG258: NGS testing pending (LabCorp). Funding complete. MTA/DTA completed (Kuroki, Alvarez Secord, Powell, Bae-Jump)

Navigator EC Profiling Efforts

- GOG249: MMR and p53 analysis completed and abstract presented at SGO Annual Meeting 2025, manuscript development in progress, Dr. Angeles Secord working with Dr. Lindsay Kuroki.

- GOG258: POLE analysis completed, awaiting statistical analysis.
-
- GOG258 and GOG249: ER approved. Funding from WashU and Duke Institutional grant obtained. Specimen submission in progress. (Kuroki, Alvarez Secord, Powell)

- GOG258 and GOG249: Navigator proposal to evaluate HER2 protein expression approved – R21 re-submitted. Dr. Angeles Secord working with Dr. Maggie Mullen.

MP2PRT: GOG210

Casey Cosgrove, MD
The Ohio State University

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Clinical Data and Samples

- Excluded Cases Vs Included Cases
- No significant differences

- Supplement with failed cases
- Separate report re: opportunities examining failed TS samples – site, location, best practices, histology misclassification etc.

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We computed the Euclidean distance matrix and performed principal components analysis (PCA), which is equivalent to multi-dimensional scaling (MDS). The largest signal in the data, driving the first principal component, was a difference in transcriptional profiles caused by sample handling; specifically, whether the sample was stored as FFPE or as fresh-frozen tissue (Figure 02).

We compute the principal components of the data set. The results are shown in **Figure 2**. In the left panel, we color the samples by the tissue type (primary, P, or metastasis, M; and FFPE, 1, or fresh-frozen, 2). It is clear that the first component is driven by the difference between FFPE and fresh-frozen samples. The right panel highlights the paired samples from the same patients but with different preservation methods, and it shows that there is a very consistent shift between the two sample types.

- It is also worth noting that none of the metastatic samples are extreme outliers, which suggests that there is not much reason to worry about them arising from secondary concurrent neoplasms.

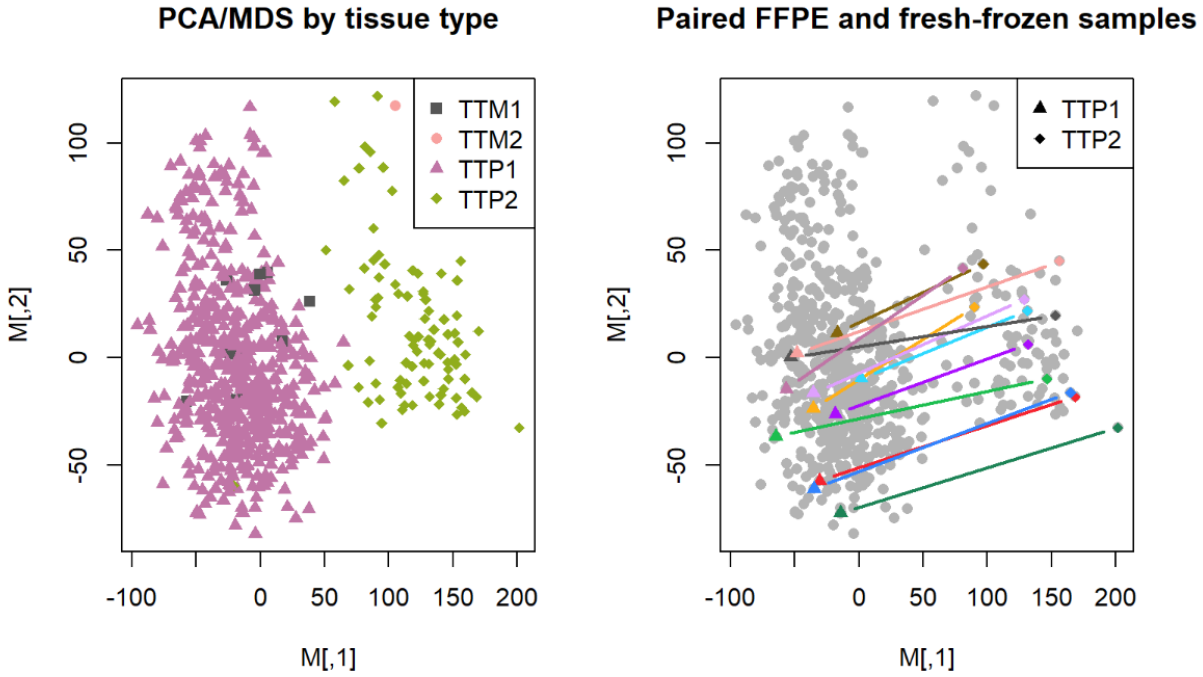

Figure 2 : Sample tissue types (FFPE or fresh-frozen) exhibit a consistent difference in expression patterns.

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Both batch correction methods successfully united the FFPE and fresh-frozen samples, and brought the twelve pairs of samples relatively close together (Figures 04 & 05)

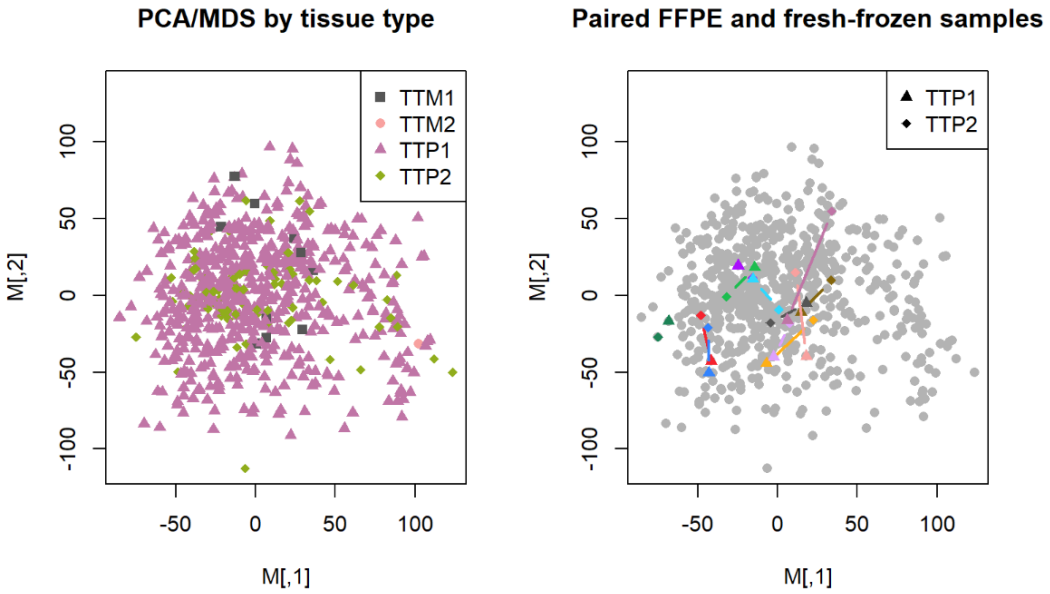

Figure 4 : PCA/MDS after batch correction using all samples.

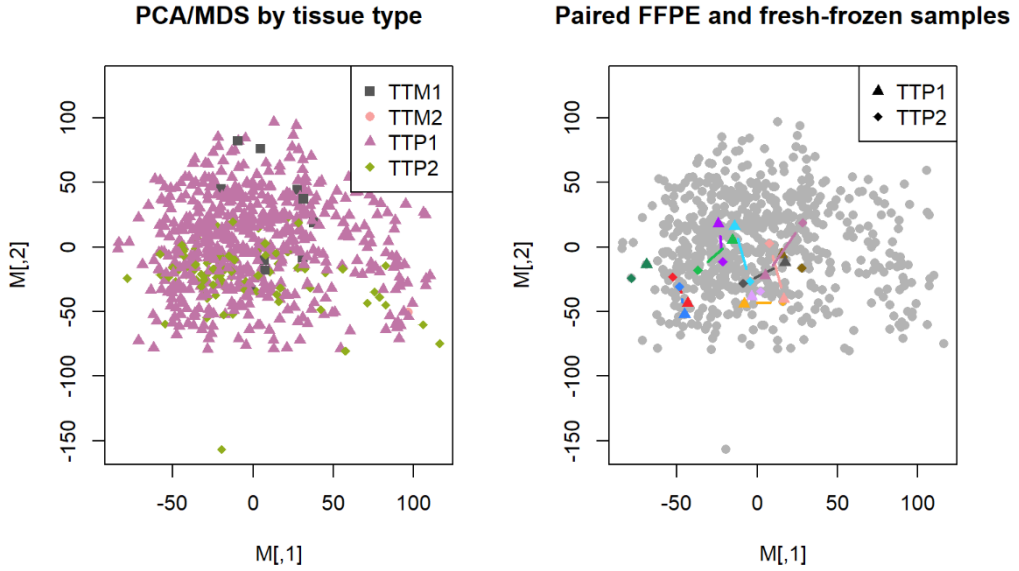

Figure 5 : PCA/MDS after batch correction using paired samples.

In this section, we perform batch correction using mean expressions computed from all samples of each type. **Figure 4** shows that this process is reasonably successful. The FFPE and fresh-frozen samples have been merged, and the paired samples are relatively close together.

Now we repeat the batch correction, using the paired samples instead of all samples. **Figure 5** again shows that this looks reasonable.

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Next, we performed hierarchical clustering starting from both sets of batch-corrected data (Figure 6), indicating that the choice of batch-correction method potentially has a big effect on downstream cluster analysis.

After cutting the dendrograms to produce five clusters, we found that only 63% of the samples were given consistent labels across the two analyses, with 37% of the data being reassigned to other clusters. In both analyses, eleven of the twelve pairs of samples were assigned the same labels, so this observation did not help to decide which batch-correction method was better.

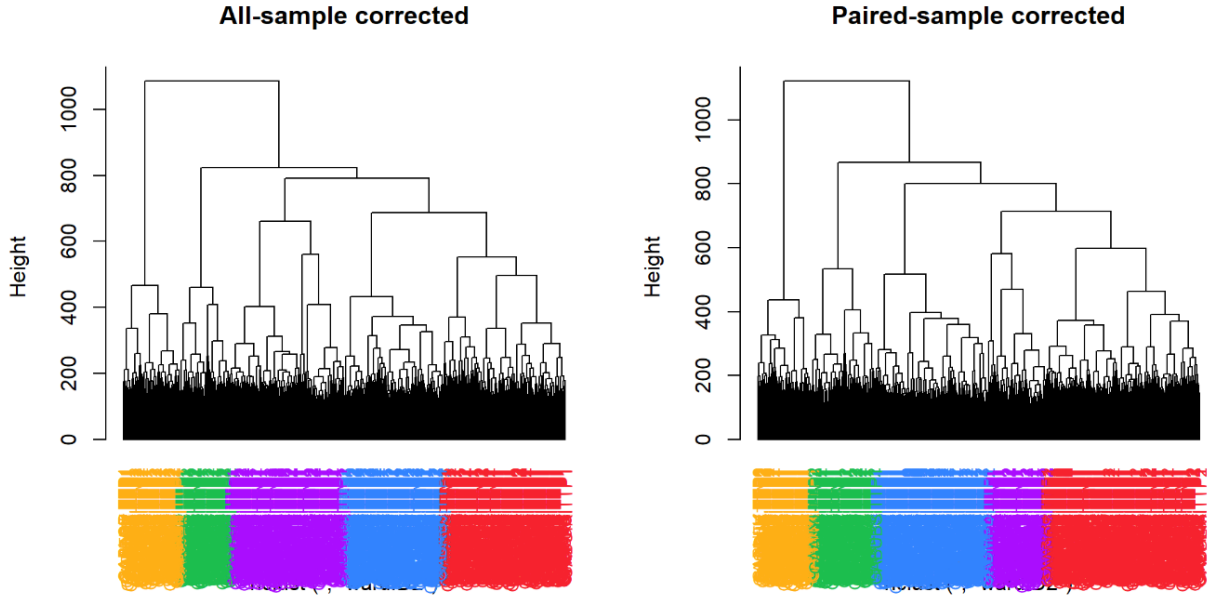

Figure 6 : Hierarchical clustering after two methods for batch correction.

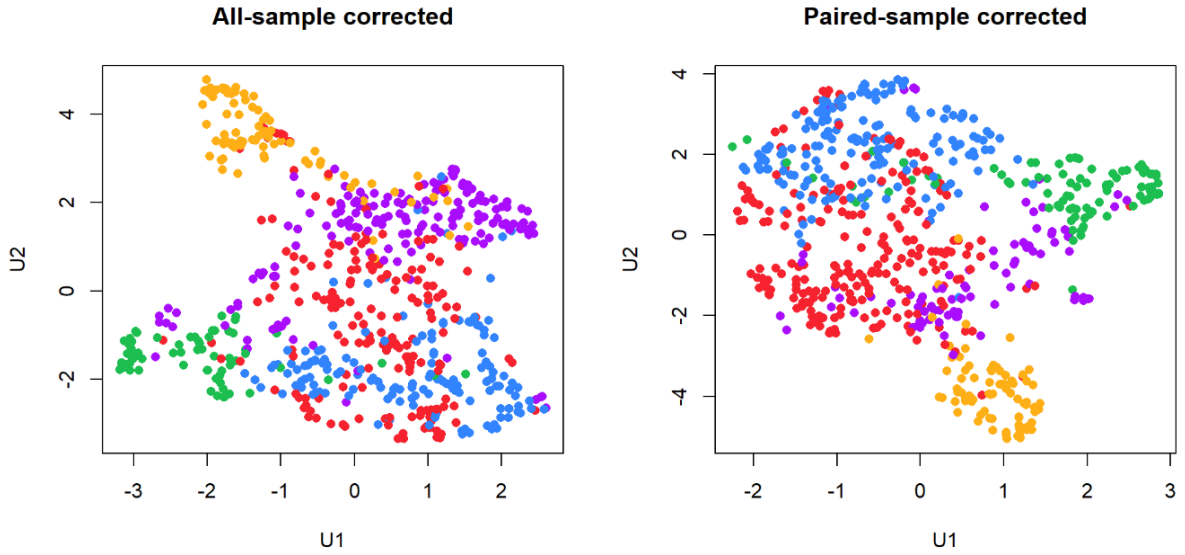

Figure 7 : UMAP plots after two methods for batch correction.

We would now like to assess the extent to which the difference in batch correction percolates downstream to affect the clustering results. Toward this end, we have performed hierarchical clustering after both methods of correction (**Figure 6**).

It is clear that the structure of the dendrograms is somewhat different. We can get a quantitative view of the magnitude of the differences by looking at the "confusion matrix" comparing the cluster assignments. About 63% of the samples are given consistent labels, and the remainng 37% get different labels.

We view this as saying that the clusters are very different. One way to decide which of the clustering methods is "better" is to see how often the paired samples are given the same labels:Well, that didn't help. In both methods, eleven of the twelve pairs of samples are given the same labels. (The same pair, GAAGKU, was mismatched in both methods.)

We can try taking another look at the hierarchical clusters; in this case, we use UMAP (**Figure 7**). The general layouts are similar, although they have been rotated by 180 degrees (which is the same as changing the sign of both components).

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Table 1

- Age
- BMI
- Race (will need to reconcile non-Black and White plan) Black = 342 White = 314
  - American Indian/Alaskan (8), Asian (27), Native Hawaiian/Pacific Islander (1)
- Grade will be for endometrioid only and can go with histology section
- Stage I/II and III/IV vs stages I/IV individually
- Depth of myometrial invasion – non-invasive, <50%, >50%
- LVSI (present / absent)
- Adjuvant therapy
- Don't think we need to have site/primary/recurrence since most are primary, same with FFPE vs Frozen here…
- Molecular class, TMB, other molecular features here?
  - POLE
  - MSI
  - TP53 mut vs wt vs copy number status

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Histology

Total Included			692
Endometrioid			
	Grade 1 or 2	Low grade	53
	Grade 3	High grade	171
Serous			191
Carcinosarcoma			121
Clear Cell			50
Mixed Histology		x27 combos	81
Un/DeDifferentiated			17
Carcinoma, other			8
	Adenocarcinoma	3	
	AC with Sq	4	
	Transitional	1	

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Stage

Stage IA	36
Stage IB	168
Stage IC	66
Stage IIA	20
Stage IIB	42
Stage IIIA	81
Stage IIIB	10
Stage IIIC	192
Stage IVA	1
Stage IVB	76

I/II	332
III/IV	360

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Differential Genes – Somatic Mutations at Different Rates in Women of Different Race

We display the top 20 genes in **Table 4**.

Table 4: Differentially mutated genes.

	Chi.X-squared	P	Black	White	Count	Rate
PTEN	38.76	0.0000000	26.32	50.83	256	38.79
ARID1A	29.09	0.0000001	28.79	50.17	267	40.45
OR2T4	27.75	0.0000001	23.22	7.59	109	16.52
TP53	20.91	0.0000048	65.63	47.19	367	55.61
RPL22	17.80	0.0000246	8.36	20.46	100	15.15
ELK3	17.73	0.0000255	2.48	11.22	47	7.12
PIK3CA	16.74	0.0000428	35.91	52.48	295	44.70
KRAS	14.82	0.0001181	8.05	18.81	90	13.64
RB1	14.82	0.0001181	8.05	18.81	87	13.18
LIPK	12.77	0.0003520	1.24	7.26	29	4.39
C9orf85	11.55	0.0006759	16.41	7.26	77	11.67
HEBP1	10.62	0.0011212	0.93	5.94	22	3.33
TXNL1	10.62	0.0011212	0.93	5.94	22	3.33
TRBV6-5	10.53	0.0011753	6.19	0.99	24	3.64
MYO1E	10.25	0.0013638	7.12	15.51	75	11.36
ACADM	10.19	0.0014125	3.72	10.56	47	7.12
BFAR	10.18	0.0014167	2.48	8.58	38	5.76
BRPF3	10.03	0.0015362	8.67	17.49	83	12.58
RDM1	9.93	0.0016301	0.00	3.63	12	1.82
MINDY2	9.81	0.0017383	2.17	7.92	33	5.00
TENM4	9.65	0.0018932	14.86	25.08	132	20.00
ZNF106	9.31	0.0022847	13.93	23.76	123	18.64
GCNT7	9.30	0.0022865	3.10	9.24	42	6.36
ANKRD44	9.23	0.0023794	4.64	11.55	52	7.88
UGT1A1	9.13	0.0025093	1.55	6.60	30	4.55
MTOR	9.00	0.0026972	15.17	25.08	129	19.55
LRRC8B	8.93	0.0028032	3.41	9.57	42	6.36
PTBP3	8.93	0.0028032	3.41	9.57	43	6.52
COBLL1	8.89	0.0028671	13.31	22.77	119	18.03
CHD7	8.87	0.0029022	15.79	25.74	142	21.52

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Differential Genes – Somatic Mutations at Different Rates in Women of Different Race

- We construct a table of "samples by mutations" for the set of 47 mutations that we have identified as recurrent. For each such mutation, we perform a chi-squared test to find out if it occurs at different rates in cohorts defined by self-reported race. The significant mutations (defined at the moment by $p < 0.05$, though we might eventually want to take a more stringent cutoff) are listed in **Table 3**.

Table 3: Reproducible mutations with different rates by race.

	Chi.X-squared	P	Black	White
NBPF26	45.91	0.000000	19.8	2.3
PDS5B	21.65	0.000003	14.2	3.3
NBPF1	17.79	0.000025	5.9	16.8
RNF43	15.39	0.000087	3.7	12.5
MSH3	15.19	0.000097	1.9	9.2
FRMPD3	11.46	0.000711	2.8	9.6
ANKRD20A8P	7.41	0.006498	6.2	12.9
SETD1B	7.26	0.007062	2.8	7.9
PTEN	6.59	0.010277	4.3	9.9
RPTN	6.05	0.013922	2.5	6.9
USP8	5.91	0.015072	4.3	9.6
RPL22	5.08	0.024167	9.6	4.6
OR10G9	4.55	0.032961	3.4	7.6

Three of these mutations (in *NBPF26*, *PDS5B*, and *RPL22*) are more frequent in black women; the others are more frequent in white women.

The problem with this analysis, however, is that t-tests are *too* sensitive as the number of samples increases. To understand why, consider the theory-based plot in **Figure 2**. This plot shows how the p-value decreases for a fixed difference in means as the sample size increases. Here we took a difference that was only one-tenth of the standard deviation, which is fairly small (and would usually represent a tiny fold change in expression). With 600 samples (as we have here), we would still expect to get a statistically (but not biologically) significant difference in expression.

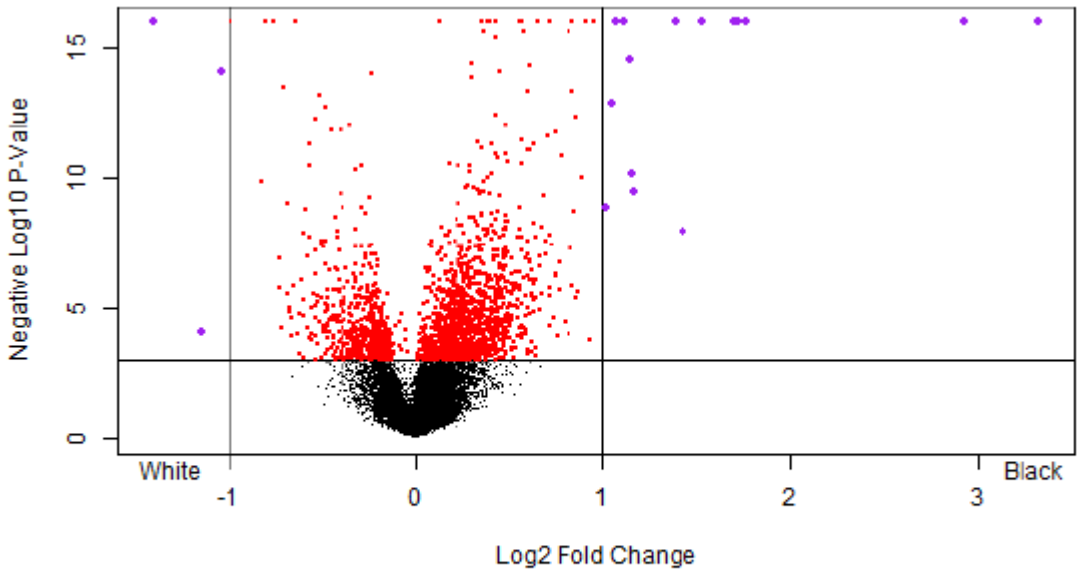

Figure 2: Volcano plot. Red points are statistically significant at FDr < 15. Purple points also have fold change at least two.

There are roughly 20 gens that satisfy the fold change criterion (FC > 2). Note that only three of these genes are more highly expressed in white patients than in black patients (TBC1D3, PIGR, and CCDC144A). (these are identified by having negative t-statistics, or "negative" fold change.)

Table 1: Top 20 differentially expressed genes by fold change

	ENTREZ	Tstats	NLog10PV	BlackMean	WhiteMean	FC
MTND1P23	100887749	16.73	17.000	4.43	1.10	10.1
MTCO1P12	107075141	18.01	17.000	7.74	4.81	7.6
RPL10P9	389342	20.11	17.000	2.93	1.17	3.4
MTCO1P40	107075172	13.76	17.000	2.42	0.70	3.3
F8A3	474384	17.16	17.000	3.17	1.47	3.2
MTND4P12	100293090	11.54	17.000	4.33	2.79	2.9
IGF2	3481	5.80	7.973	8.60	7.17	2.7
TBC1D3	729873	-14.03	17.000	2.41	3.81	-2.6
PWP2	5822	14.10	17.000	3.08	1.69	2.6
CTCFL	140690	6.39	9.473	2.53	1.36	2.2
LOC102724594	102724594	6.63	10.131	5.33	4.17	2.2
PIGR	5284	-3.97	4.087	4.57	5.72	-2.2
GSTM1	2944	8.11	14.574	2.54	1.40	2.2
TAS2R64P	338412	9.60	17.000	3.31	2.19	2.2
RPL10P6	285176	15.95	17.000	1.26	0.19	2.1
CLCN3P1	100419056	7.58	12.877	3.65	2.61	2.1
CCDC144A	9720	-7.98	14.122	4.68	5.72	-2.1
FBN3	84467	6.16	8.883	3.23	2.21	2.0
LOC102724159	102724159	-10.62	17.000	2.28	3.26	-2.0
TBC1D3D	101060389	9.35	17.000	1.66	0.71	1.9

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Thank You

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