SIDNEY KIMMEL COMPREHENSIVE CANCER CENTER

BLOOMBERG~KIMMEL INSTITUTE
FOR CANCER IMMUNOTHERAPY

JOHNS HOPKINS
MEDICINE

Pancreatic Cancer is PRIMED to Become an Immunologic Disease

SITC 2020 Annual Meeting

Keynote Address

Elizabeth M. Jaffee, M.D.

Dana and Albert Broccoli Professor of Oncology
The Skip Viragh Pancreatic Cancer Center

November 12th, 2020

Disclosure Information

I will be discussing the investigational use of:

- GVAX
- Listeria Monocytogenes – mesothelin

Both licensed to Aduro Biotech; Dr. Jaffee and the Johns Hopkins University have the potential to receive royalties

Chief Medical Advisor for the Lustgarten Foundation

Co-Founder Abmeta Therapeutics

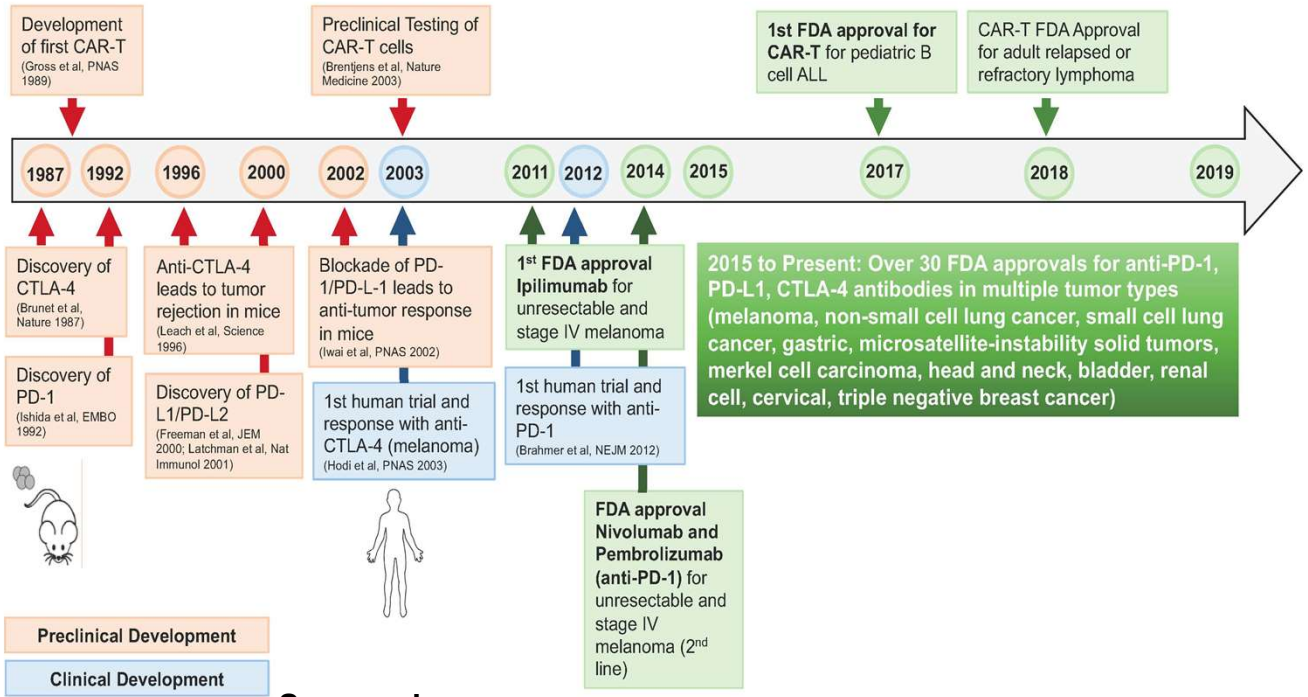
Scientific Adivisory Board activity:

- Genocea
- Adaptive Biotech
- DragonFly
- CSTONE
- Achilles
- Parker Institute

Grants: Aduro Biotech, Bristol Myer Squibb

30 Years of Scientific Discoveries Created this Historic Time of Accelerated Approvals of Durable and Curable Immunotherapies

T Cell Therapy: From Development to Approval



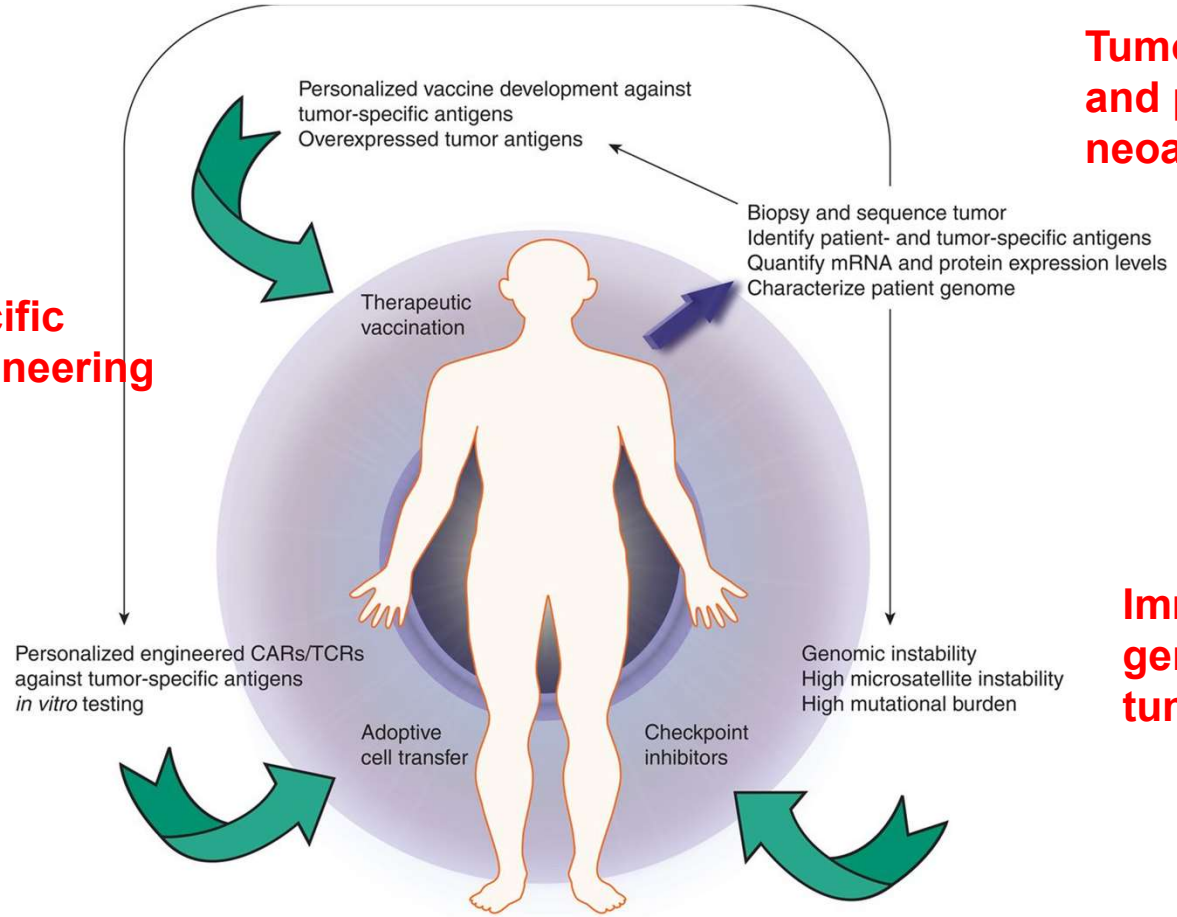
PCR 1985 → **Sequencing of human genome 2000** → **Next Generation WES/RNAseq/multiplex proteomics/metobolomics/single cell technologies 2010**

Dr. Neeha Zaidi

The Field is now in a new era of precision immunotherapy

Tumor subtype specific and patient specific neoantigen vaccines

Patient specific cellular engineering



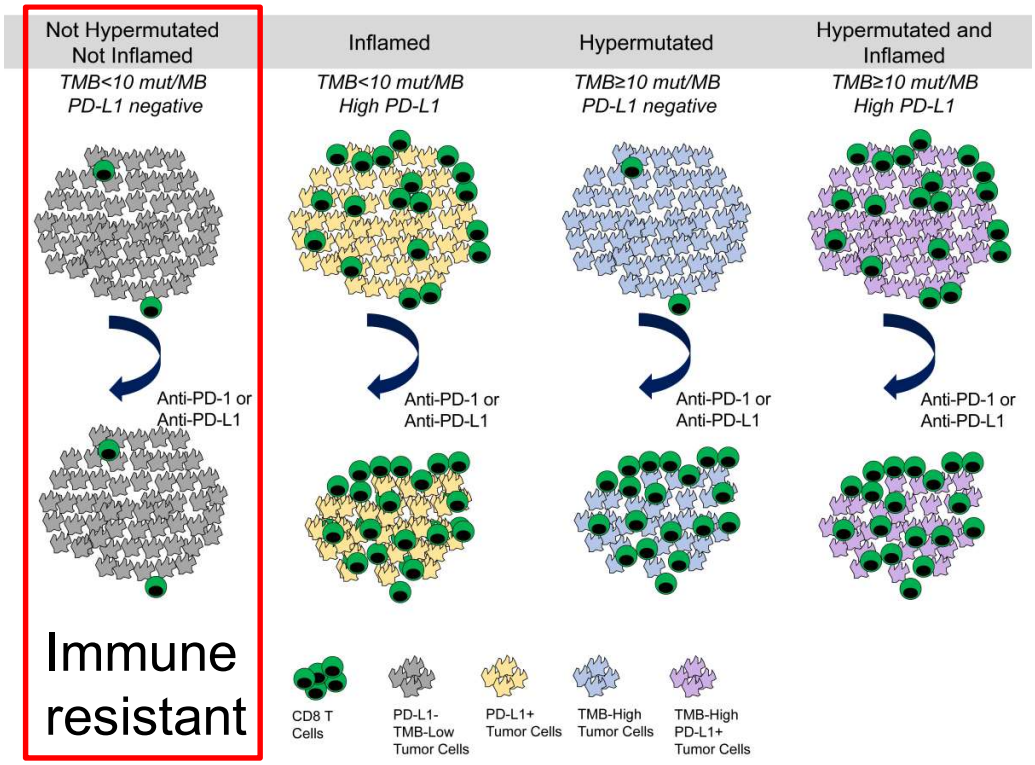
Mandal and Chan, Cancer Discovery 2016

Immunotherapy for genetically defined tumor subtypes

Immune checkpoint inhibitors have shown unprecedented responses against a number of advanced cancers

- High tumor mutational burden (TMB)
  - Single nucleotide variants
- Expression of other tumor antigens
  - Viral antigens (Merkel Cell)
  - Insertion and deletion (Indels) - derived neoantigens (RCC)
  - Fusion proteins
- High expression of PD-L1 in TME
- Available tumor-recognizing T cells in tumor or tumor draining lymph nodes

TMB/Neoantigen and PD-L1 Status Identify Immunologic Subtypes of Cancer



Mark Yarchoan

Yarchoan et al. JCI Insight 2019

Challenges to Progress in Immune Resistant Tumors

- The **tumor microenvironment** of immunotherapy **resistant** cancers have multiple immune suppressive signals that need to be bypassed to achieve clinical responses
- **Heterogeneity** within tumors from the same patient has become an area that needs more understanding
- **Quality T cells** need to be induced for immune checkpoints to work!

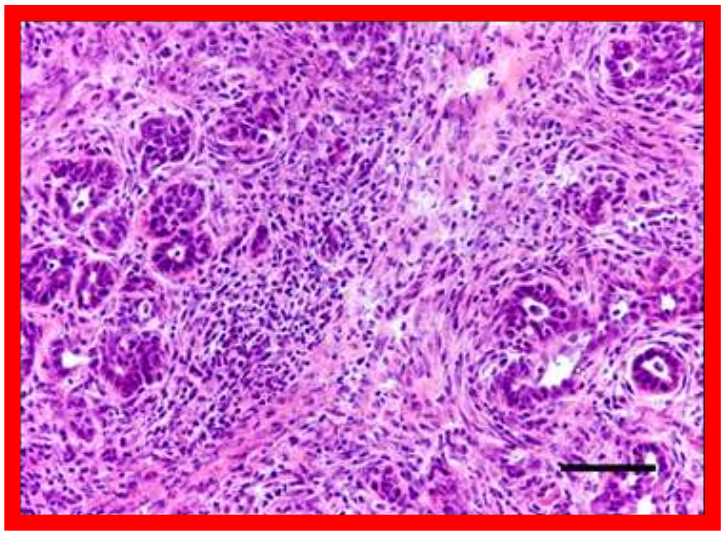
What we know from pancreatic cancer – Hostile environment to T cells – Example of an Immune Resistant tumor

Fibroblasts

Extracellular matrix

Macrophages

Immature myeloid cells



B cells

Regulatory T cells

Low TMB

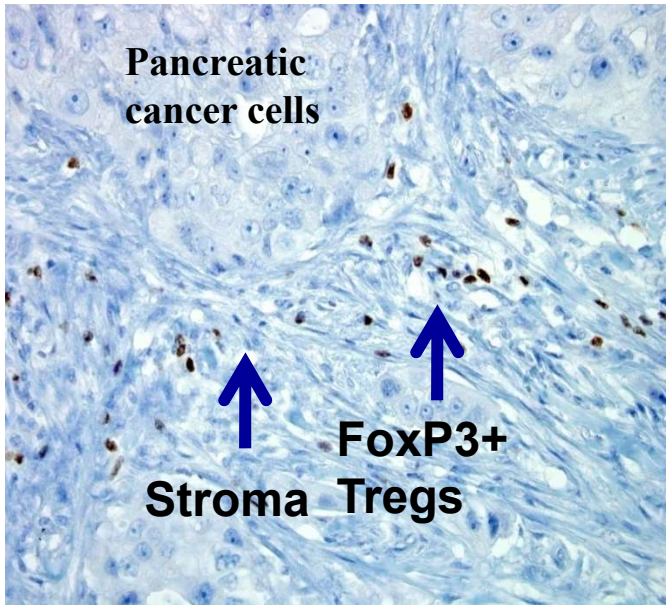
Low/No PD-L1

Desmoplastic stroma

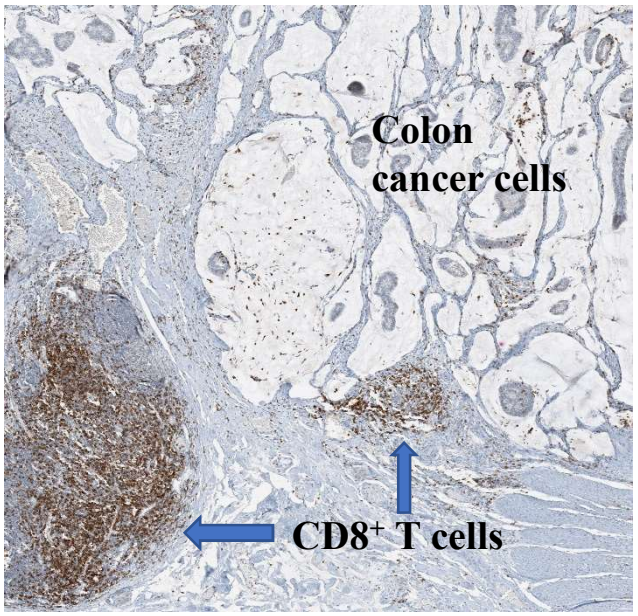
Emerging technologies and analysis platforms are providing the opportunity to understand the complex signaling networks

Invasive pancreatic tumors lack infiltration of effector T cells

Stroma supports both pro-carcinogenic and anti-cancer inflammatory cells



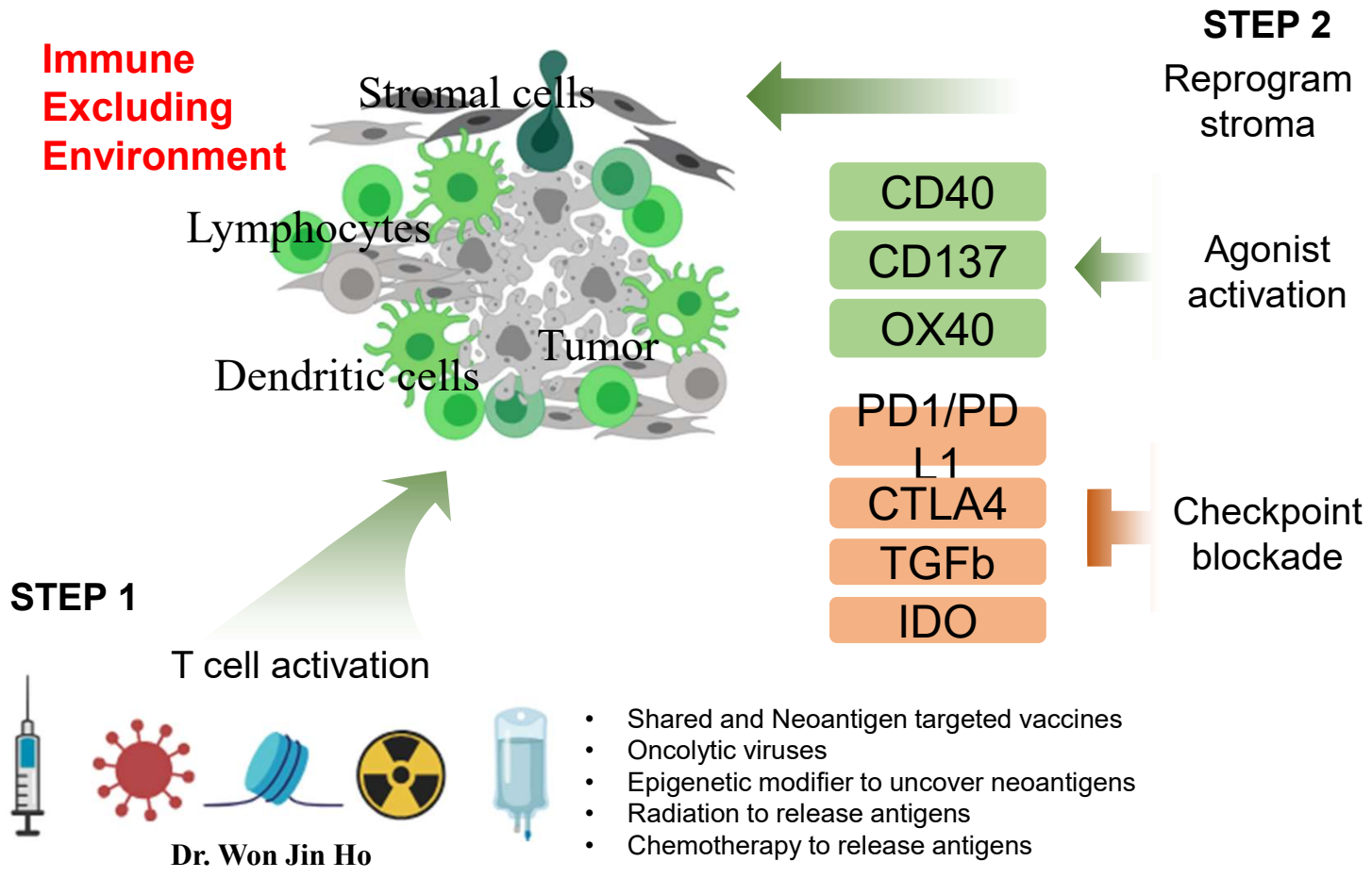
Pancreatic cancers are infiltrated with immune suppressive regulatory T cells (Tregs) and MDSCS (not shown)



Microsatellite instability high tumors are naturally infiltrated with effector T cells

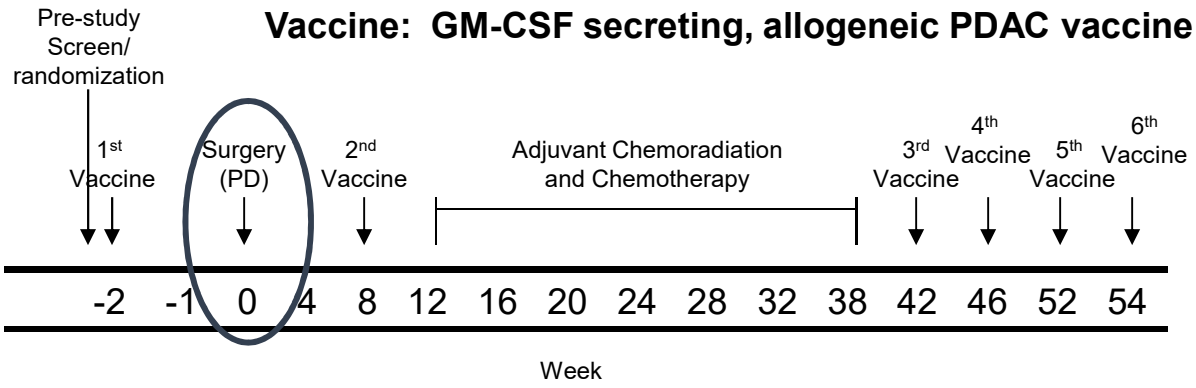
How can we convert an immunologically unresponsive tumor into one that responds to immune checkpoint therapy?

Naturally Non-Immunogenic Cancers Require at least a 2-Step Process to Reprogram the TME and Optimize Immunotherapy



(Neo)adjuvant Pancreatic Cancer Vaccine Study Provides Evidence Supporting T Cell Induction/Infiltration into Tumors

Cancer Immunology Research, 2014

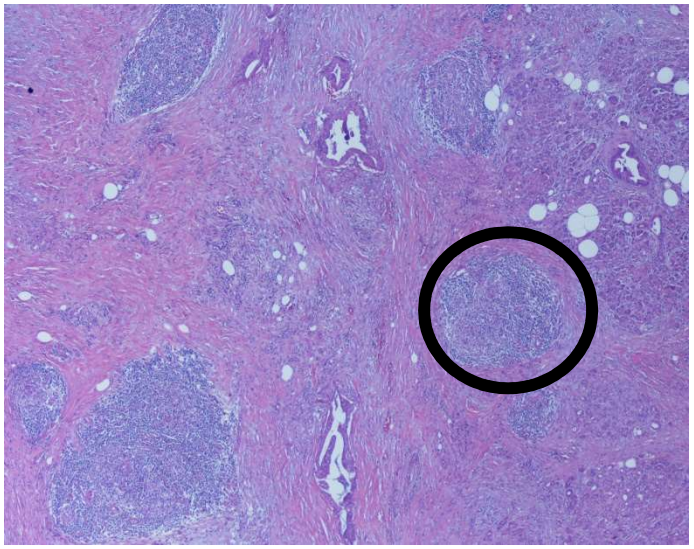


- **Significant improvement in disease-free and overall survival**
- **Associated with expanded mesothelin-specific $CD8^+$ T cell repertoire**
- **Increased T cell avidity associated with improved disease-free survival**

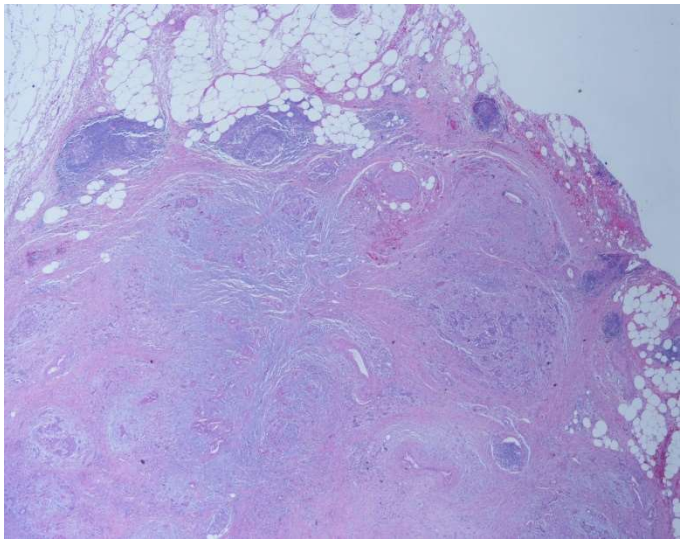



Lei Zheng, M.D./Ph.D. **Chris Wolfgang M.D./Ph.D.** **Dan Laheru, M.D.** **Eric Lutz, Ph.D.**

Lymphoid Aggregates found in 2 location patterns in vaccinated patients 2 weeks after a single vaccine

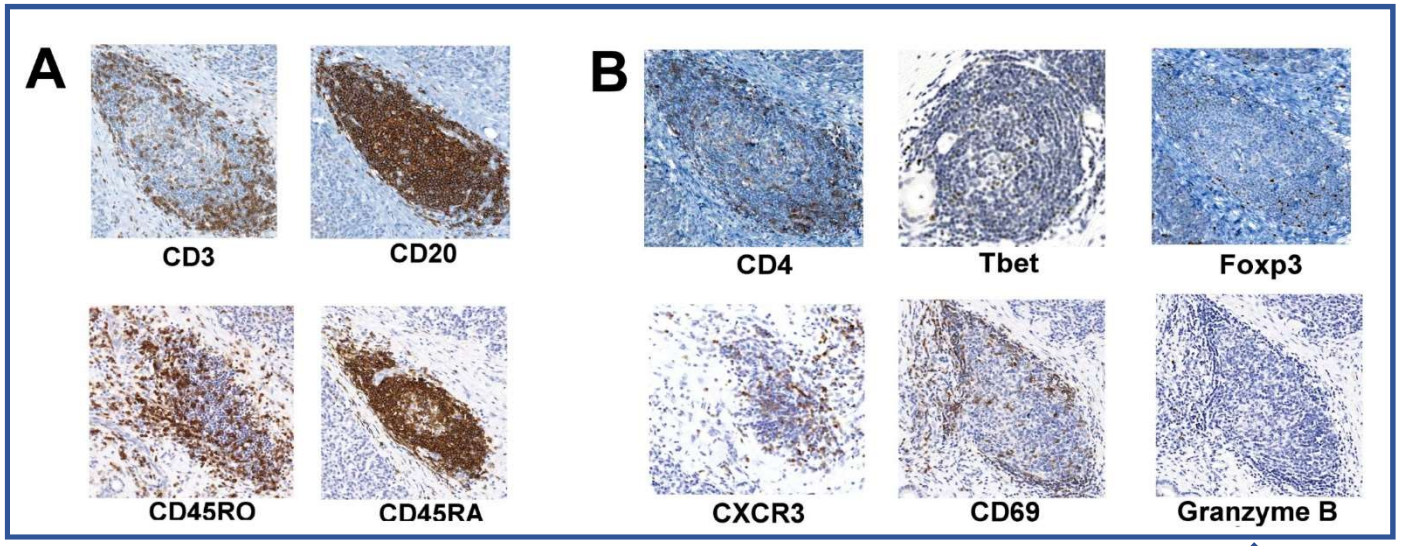


Intratumoral



Peri-tumoral

Lymphoid Aggregates Are Sites of Immune Activation and Regulation – Not Cytolysis



Antigen experienced T cells

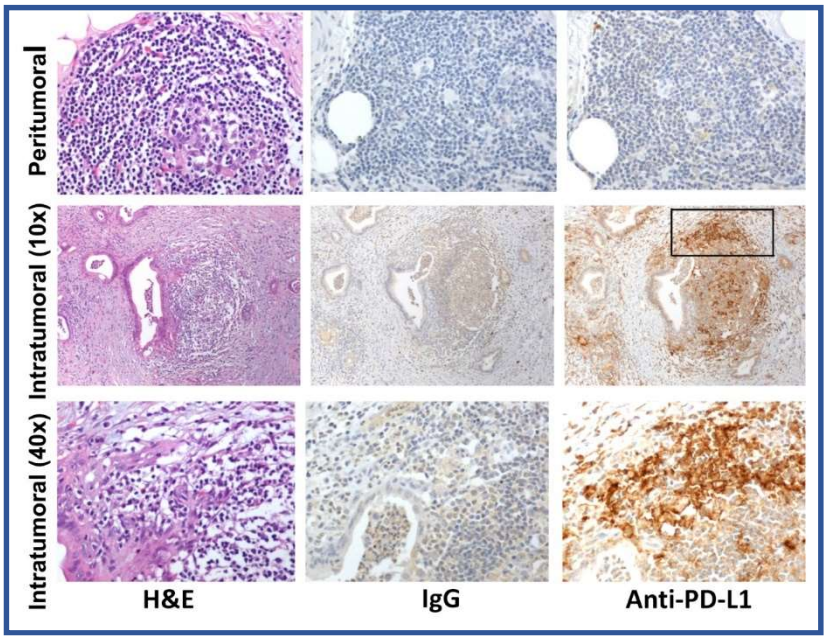
Antigen naive T cells

T cell trafficking marker

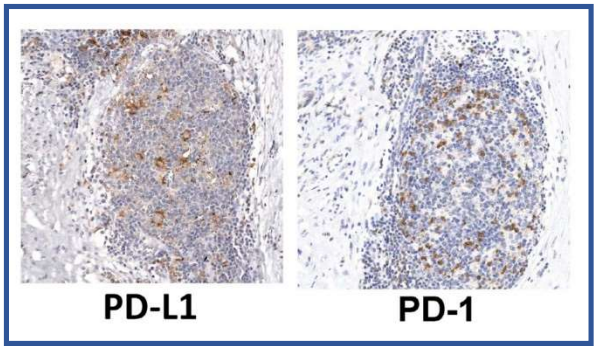
Early T cell activation marker

T cell function marker

CD8 T cell activation in lymphoid aggregates produce IFNγ which upregulates T cell inhibitory signals like PD-L1



Co-localization



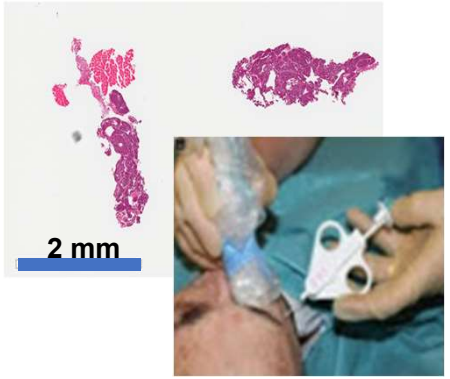
Robert Anders

Activated T cells → IFNγ production → PD-L1/PD-1

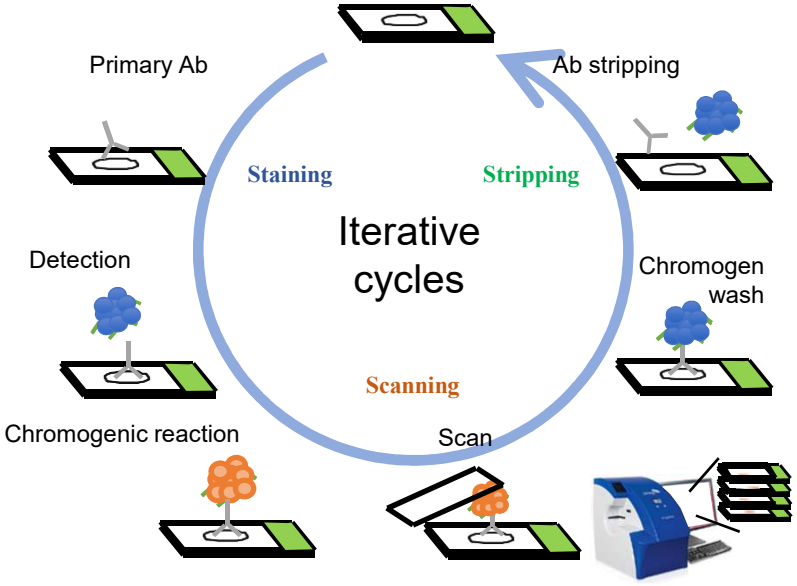
Multiplex Immunohistochemistry Approach To Interrogate The TME

collaboration with Lisa Coussen's group

20 ga core biopsy, x2



Tsujikawa T, et al. Cell Reports, 2017



Modified from

Tramu G, et al. J Histochem Cytochem 1978
Glass G, et al. J Histochem Cytochem, 2009
Stack EC, et al. Methods, 2014

Takahiro Tsujikawa

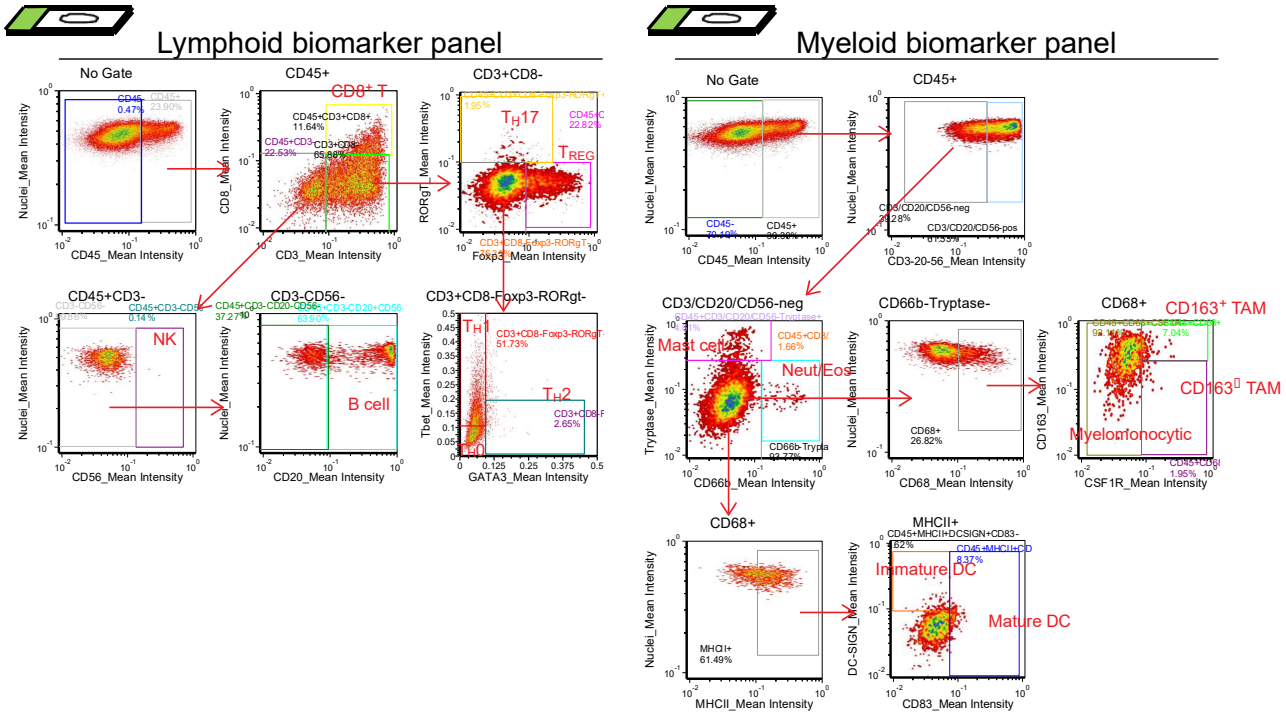
Lisa Coussens

CURE PANCREATIC CANCER
LUSTGARTEN FOUNDATION

ADMINISTERED BY AACR American Association for Cancer Research
FINDING CURES TOGETHER
SCIENTIFIC PARTNER OF STAND UP TO CANCER

STAND UP TO CANCER

Conversion of multiplex IHC data into image cytometry allows for quantification of cell types



Tsujikawa T, et al. Cell Reports, 2017.

Multiplex IHC enables detection of 12-different epitopes in a single FFPE section

Sequential IHC

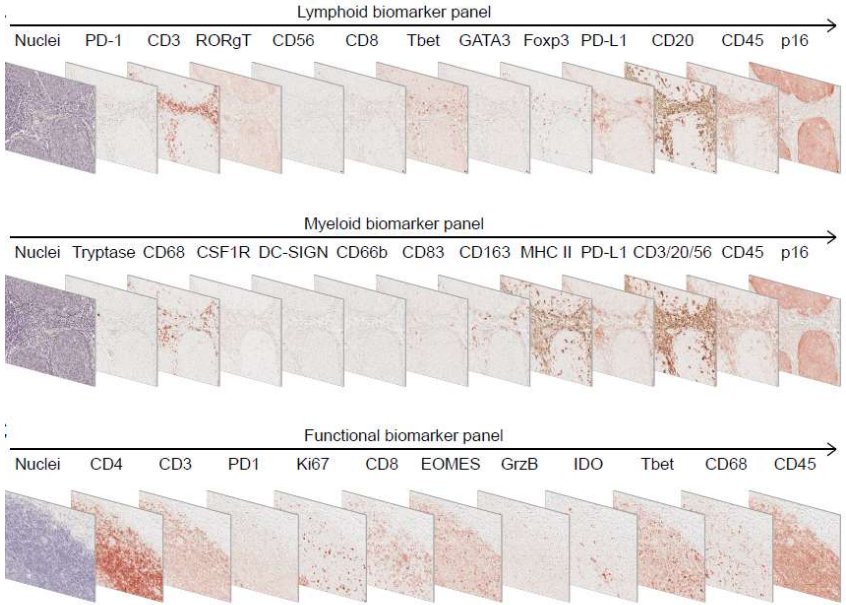
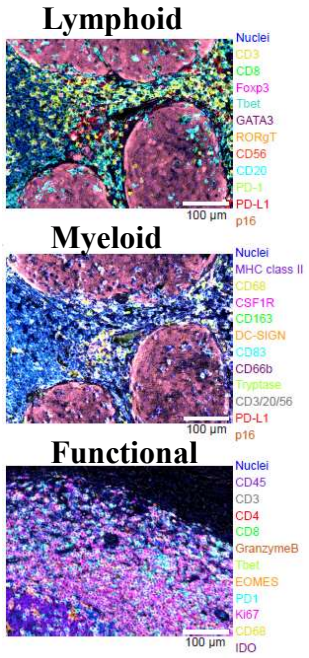


Image Co-registration

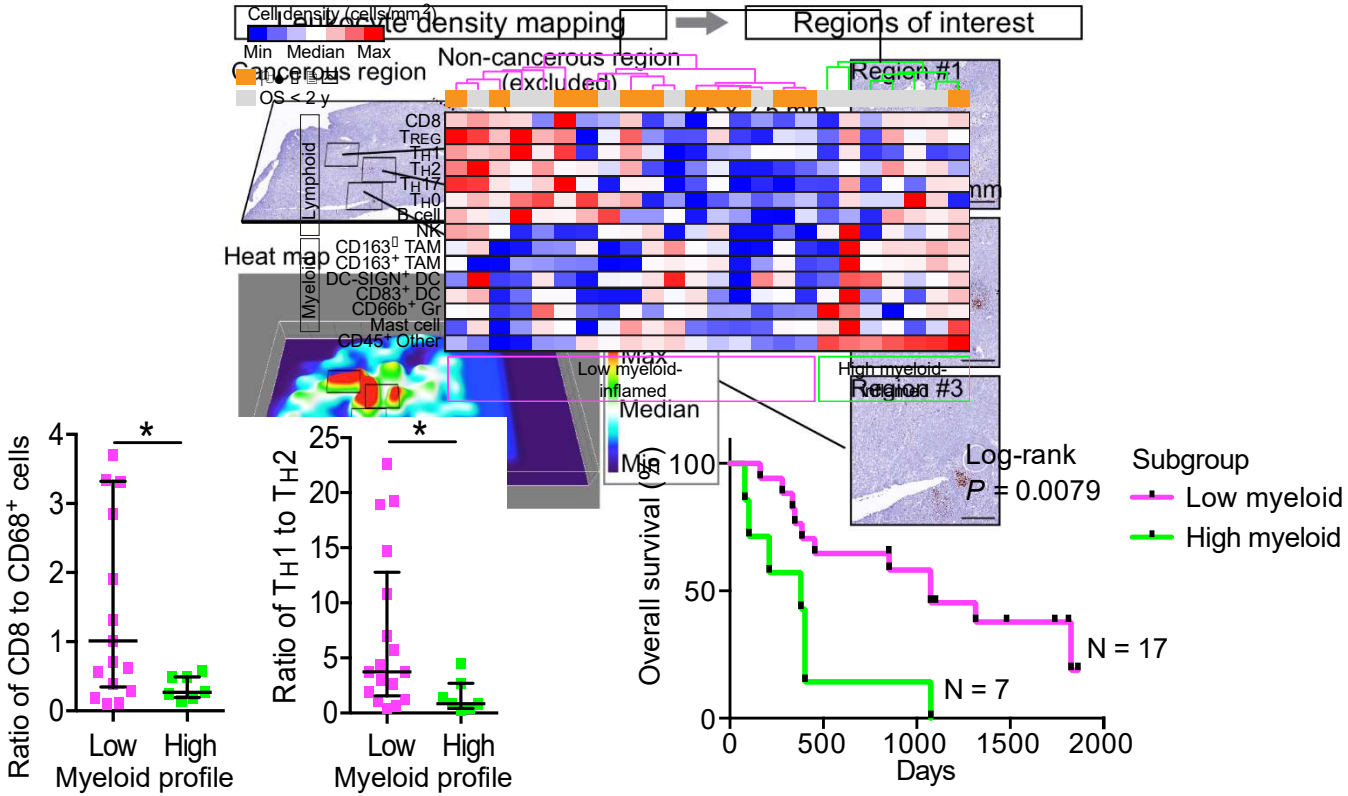
Color Deconvolution

Visualization



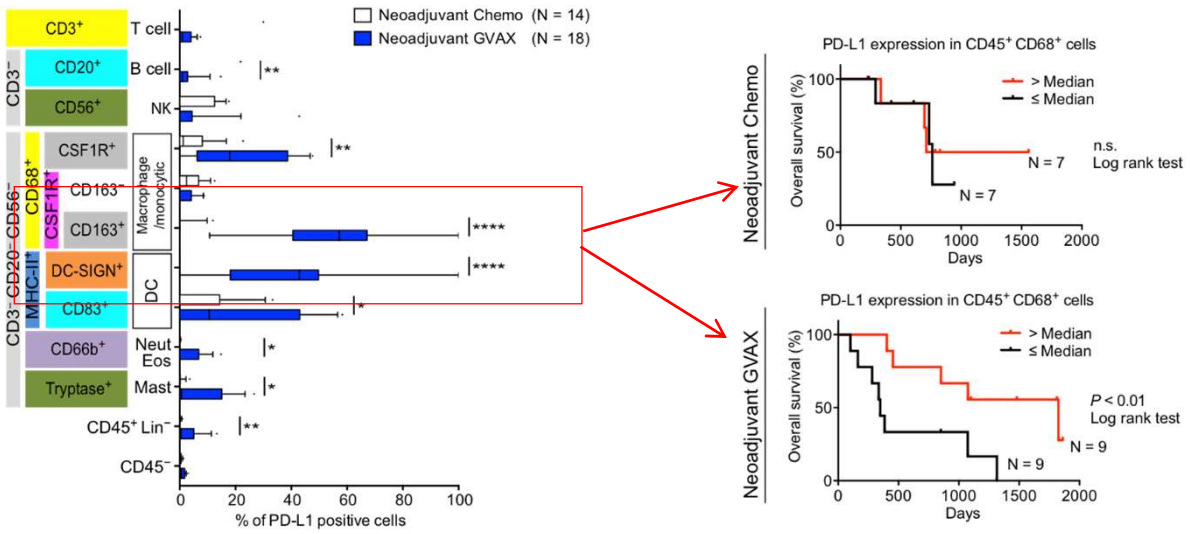
Tsujikawa T, et al. Cell Reports, 2017

Low myeloid content in CD45$^+$ inflammed" areas is associated with improved survival



Tsujikawa, et al. *Cell Reports*, 2017.

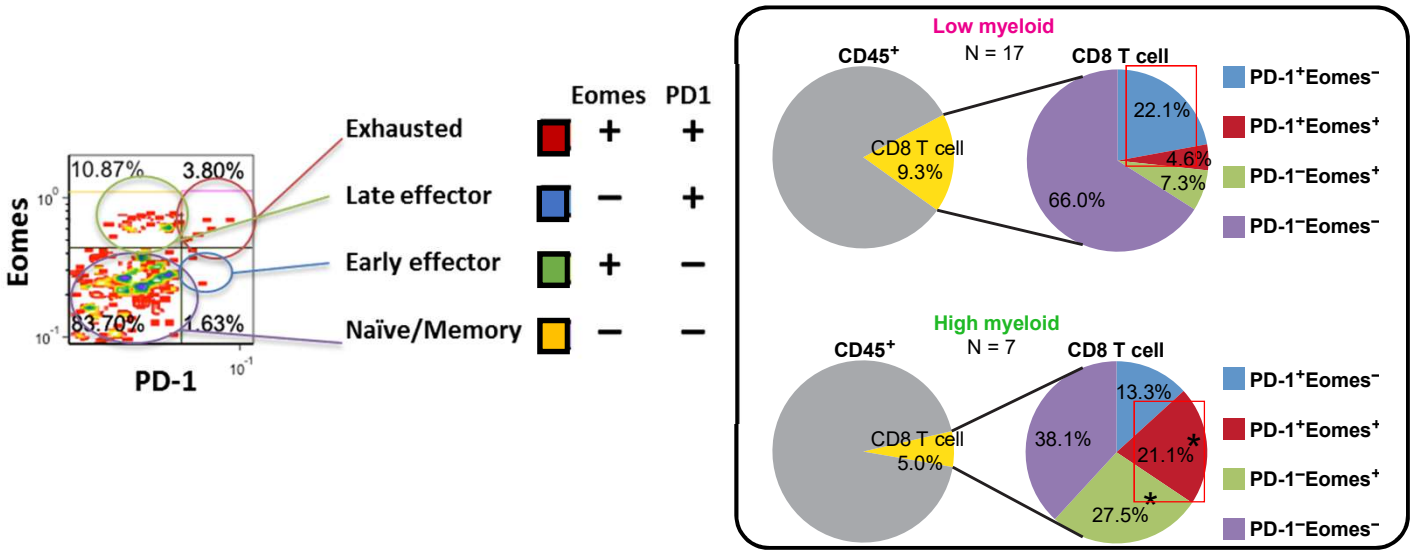
Neoadjuvant GVAX therapy is associated with PD-L1 upregulation in myeloid cell lineages correlating with prognosis



Tsujikawa T, et al. Cell Reports, 2017

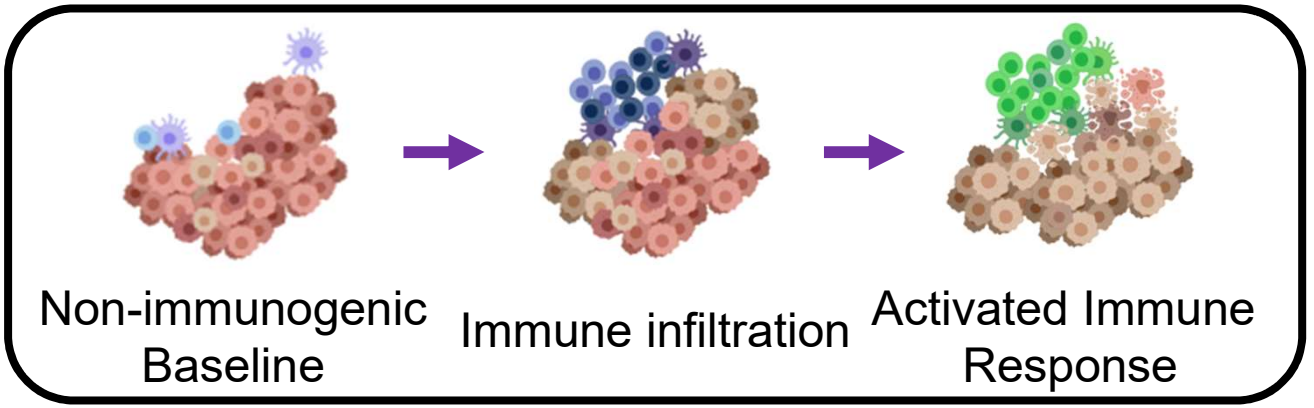
Low myeloid infiltration is associated with increased late effector and fewer exhausted T cells in lymphoid aggregates

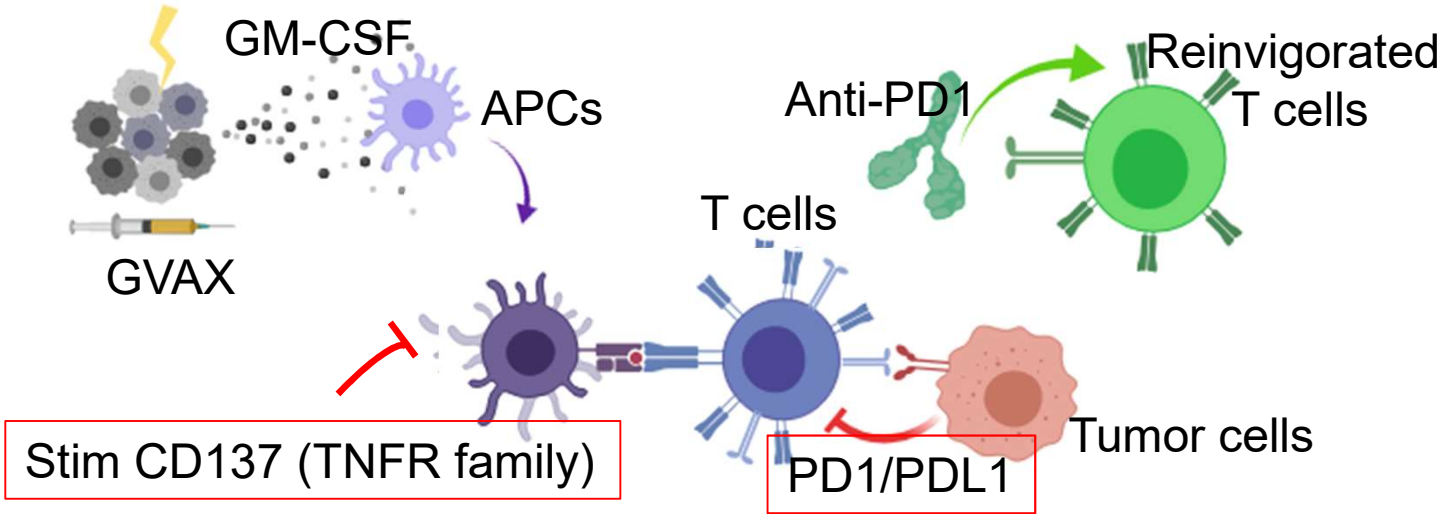
High myeloid infiltration is associated with increased exhausted and early effector T cells in lymphoid aggregates



Tsujikawa T, et al. Cell Reports, 2017

Neo-Adjuvant Study of Vaccine +/- PD-1 Blockade





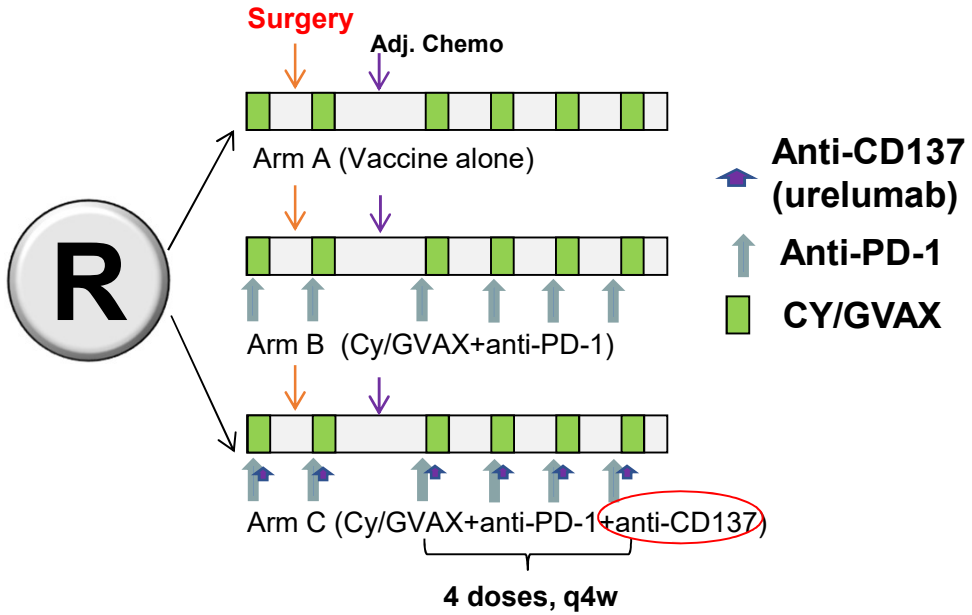
Dr. Won Ji Ho

A Platform phase II clinical trial of neoadjuvant and adjuvant CY/GVAX vaccine with or without anti-PD-1 antibody and/or anti-CD137 agonist antibody for resectable pancreatic cancer

Added 3rd arm after Arm A and Arm B completed enrolling 25 patients

50 Patients with resectable PDA

1:1:2 randomization

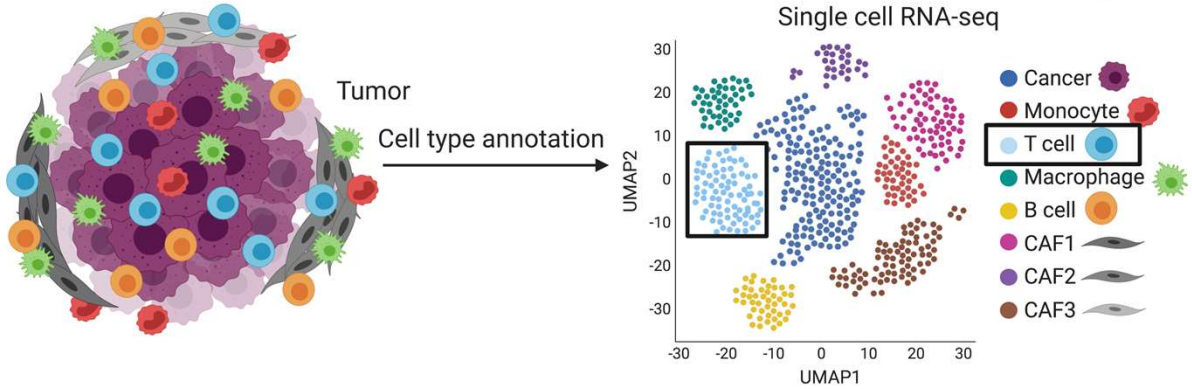


Three of 10 patients demonstrated partial pathologic response to one dose of the GVAX/aPD1/aCD137 treatment

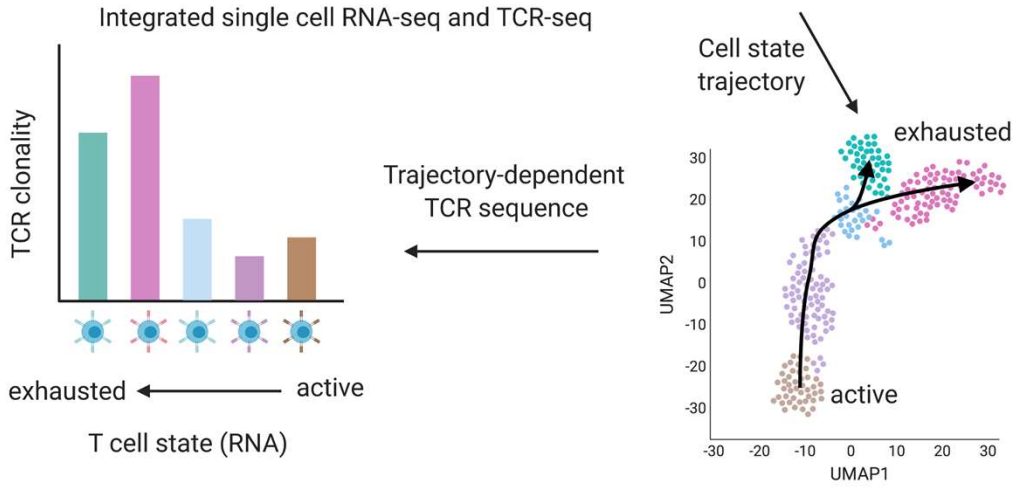
Newer technologies provide new mechanistic insights into prior and ongoing clinical trials

- Mass Cytometry
- Multiplex immunohistochemistry
- T cell receptor (TCR) sequencing
- Single cell RNA sequencing
- Computational biology

Single cell analyses classifies immune functional states in association with immunotherapies



Luciana Kagohara

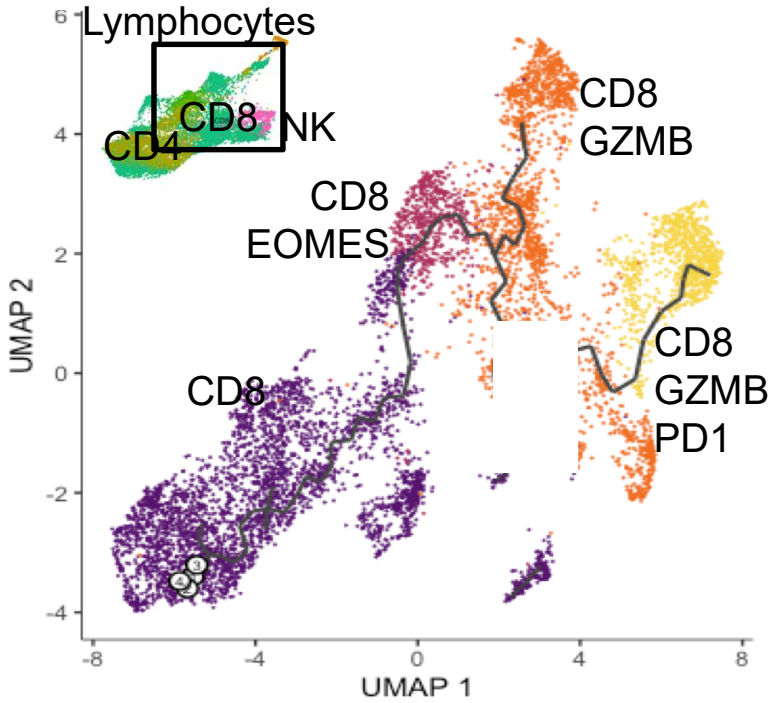


Elana Fertig

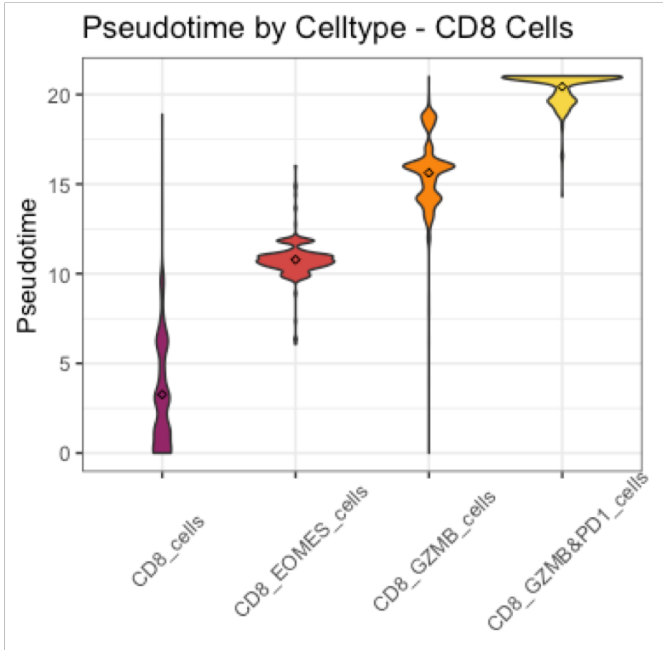
10X Genomics Visium Clinical Translational Research Network

Pseudotime quantifies variation in functional phenotype for CD8⁺ T cell populations - one snapshot quantifying different stages shown with Violin Plots

UMAP plot of CD8+ cells



CD8+ subtypes by pseudotime



Unpublished data

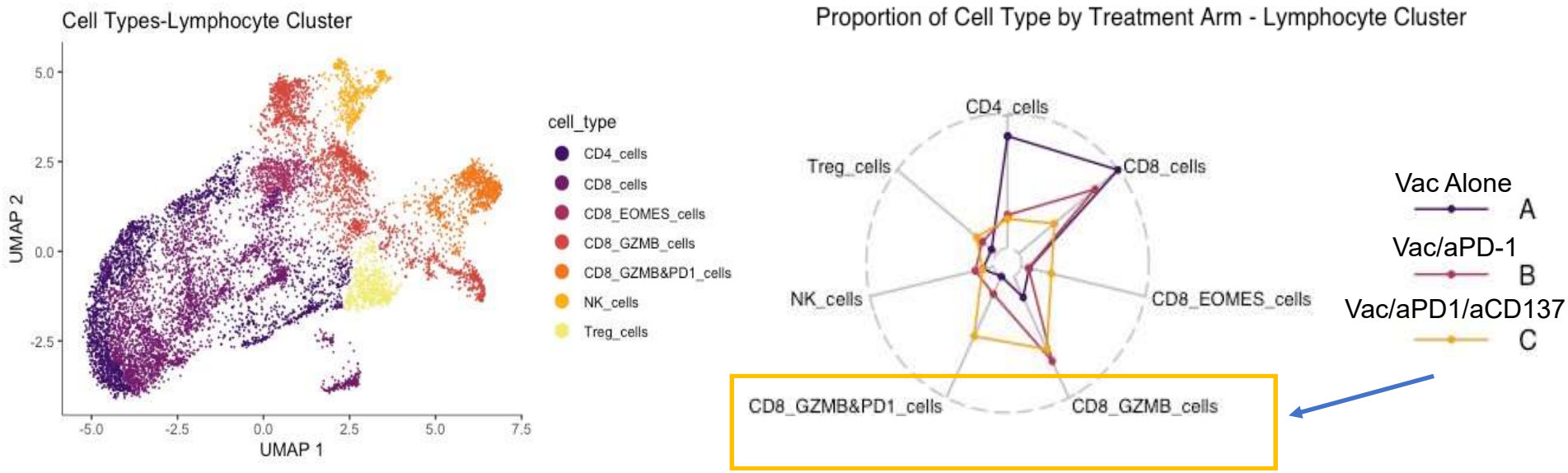
Melanie Loth

Luciane Kagohara

Elana Fertig

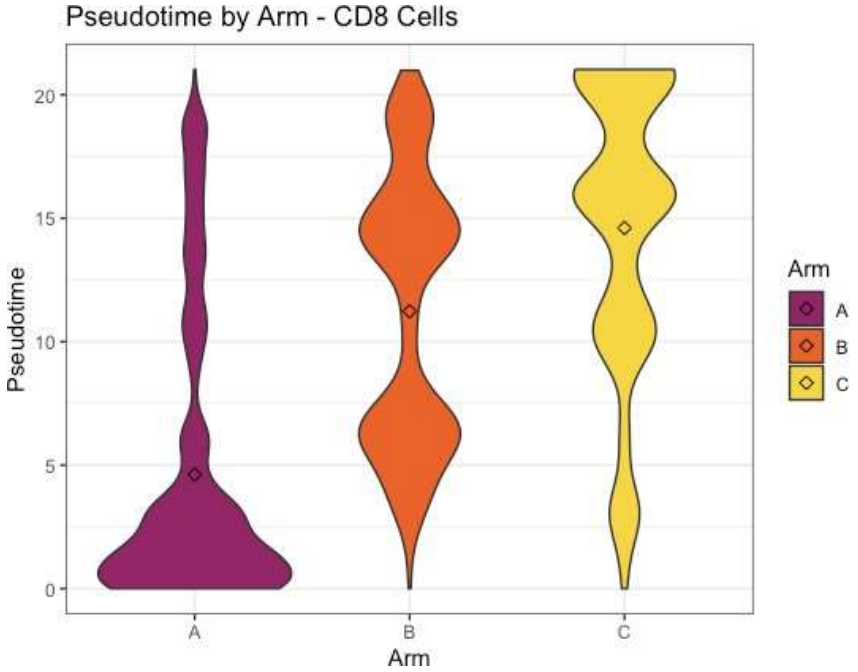
Lei Zheng

Cellular state changes distinguish therapeutic regimens



Lei Zheng, Melanie Loth, Luciane Kagohara, Elana Fertig
Unpublished data

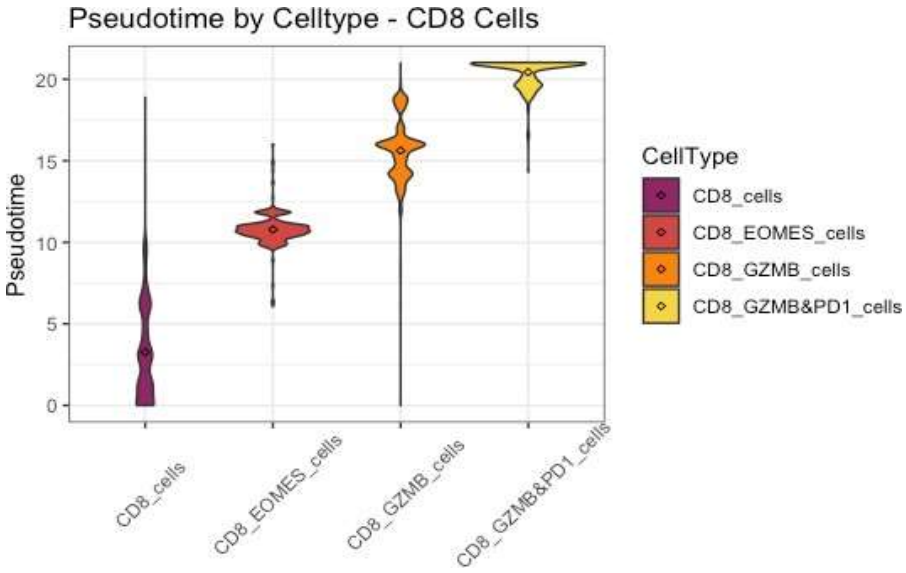
Violin plots showing cellular state changes by therapeutic regimens and cell phenotypes



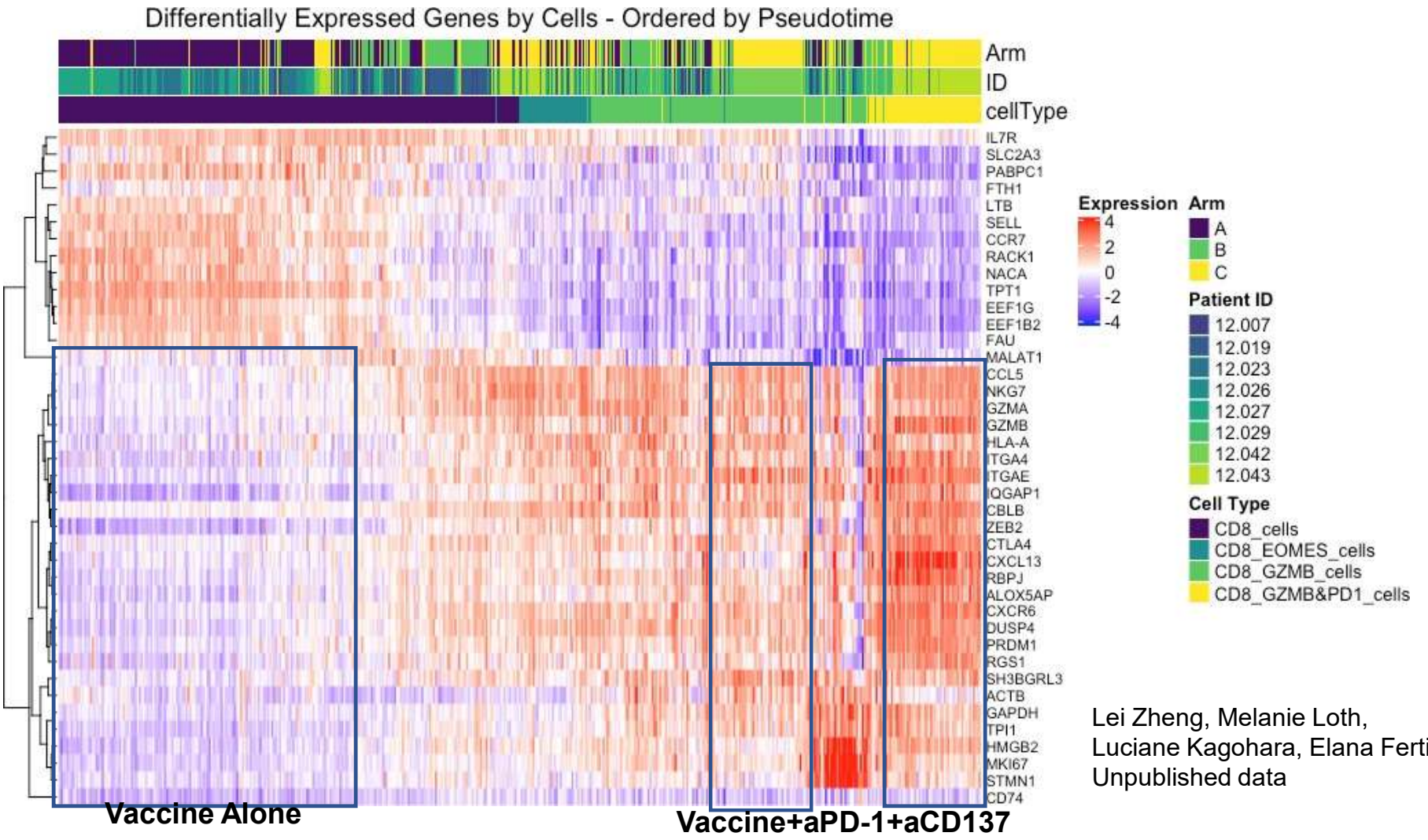
Vaccine Alone

Vaccine + anti-PD-1

Vaccine +anti-PD-1 + anti-CD137

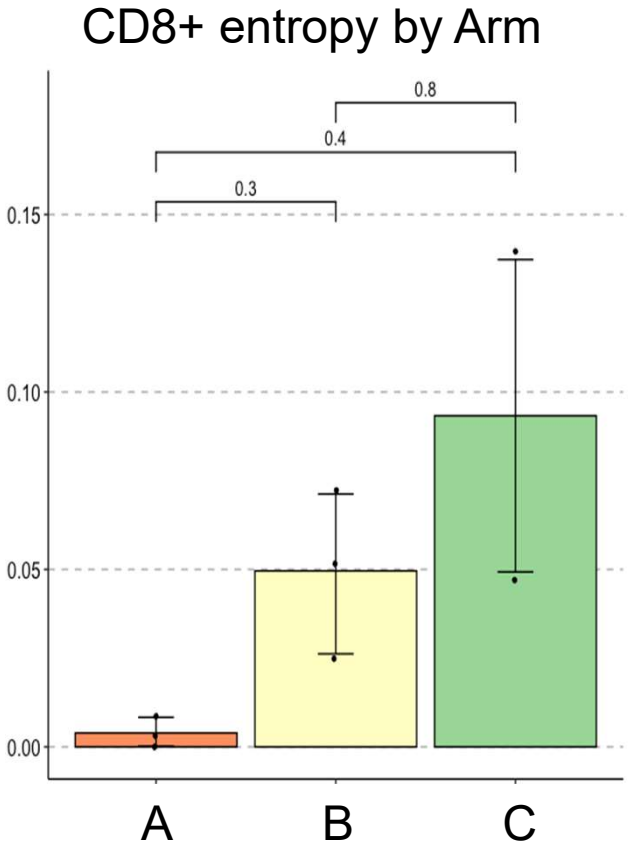


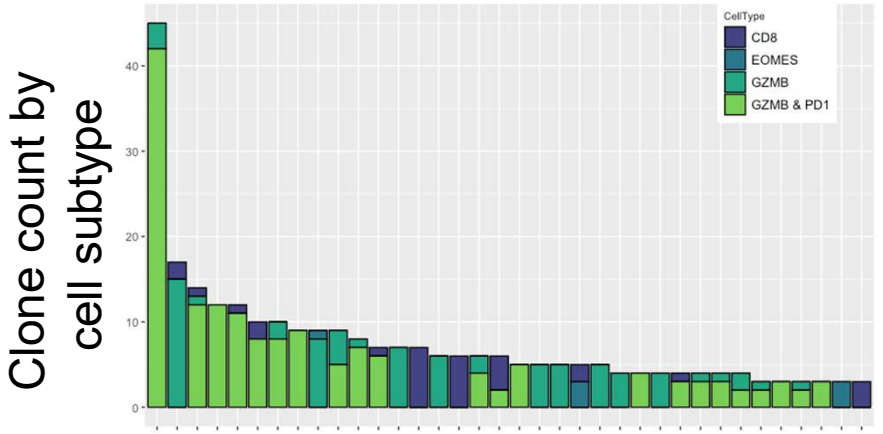
Lei Zheng, Melanie Loth, Luciane Kagohara, Elana Fertig
Unpublished data

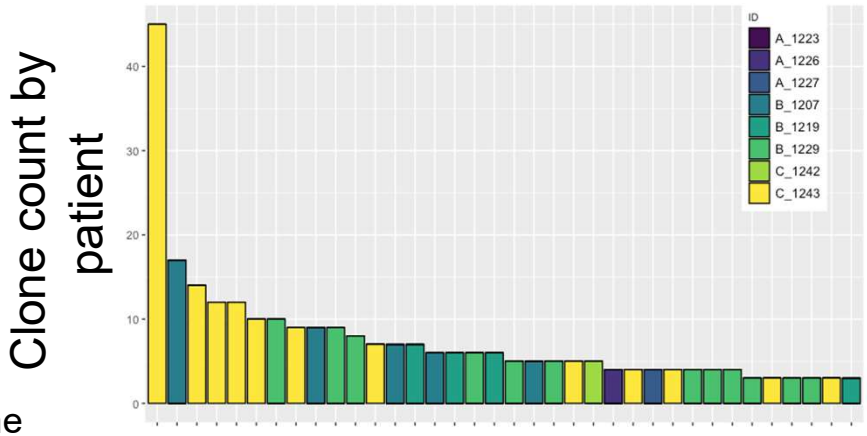


Lei Zheng, Melanie Loth,
Luciane Kagohara, Elana Fertig
Unpublished data

scTCR-seq integration shows enhanced numbers of activated T cell clones with vaccine+anti-PD-1 and vaccine + anti-PD-1 + anti-CD137

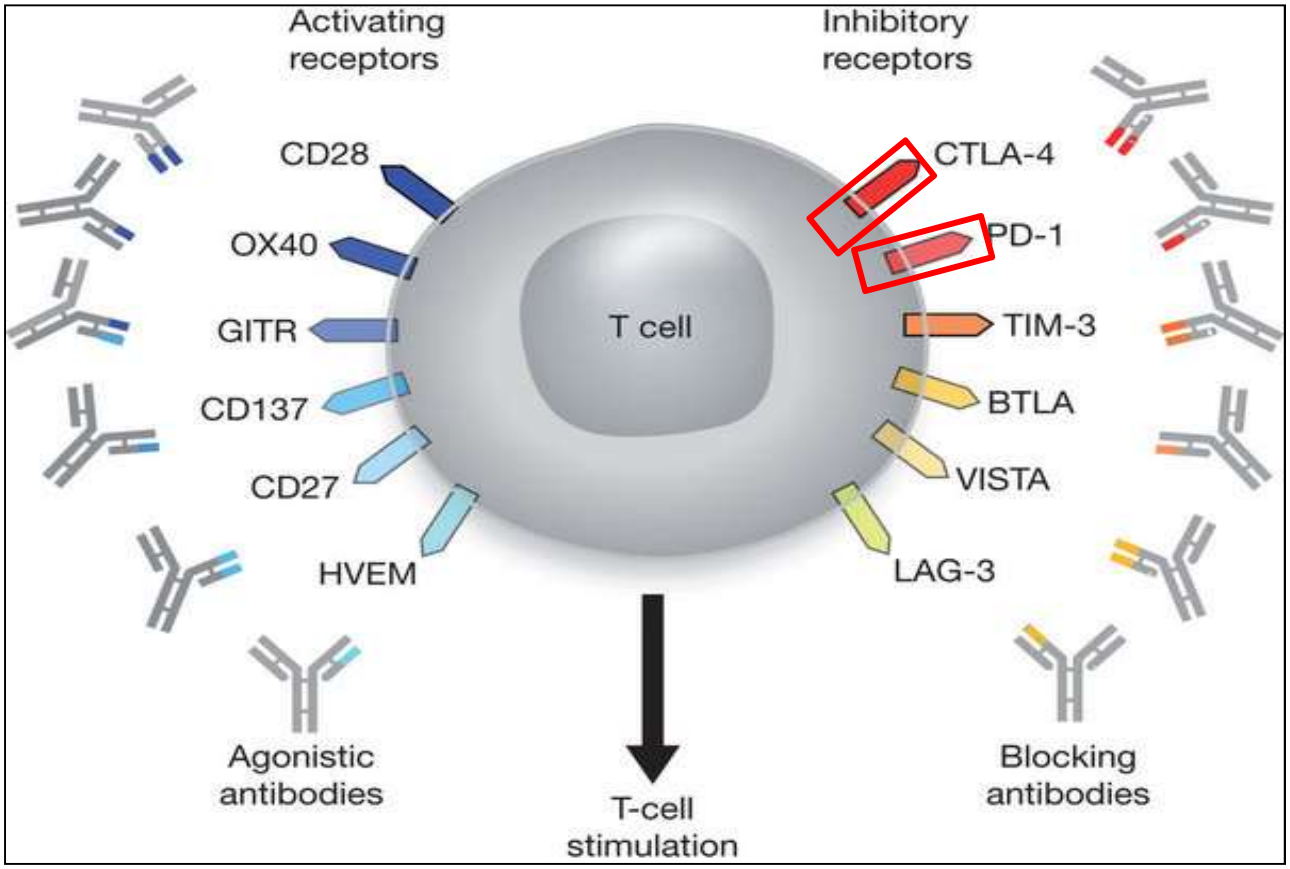






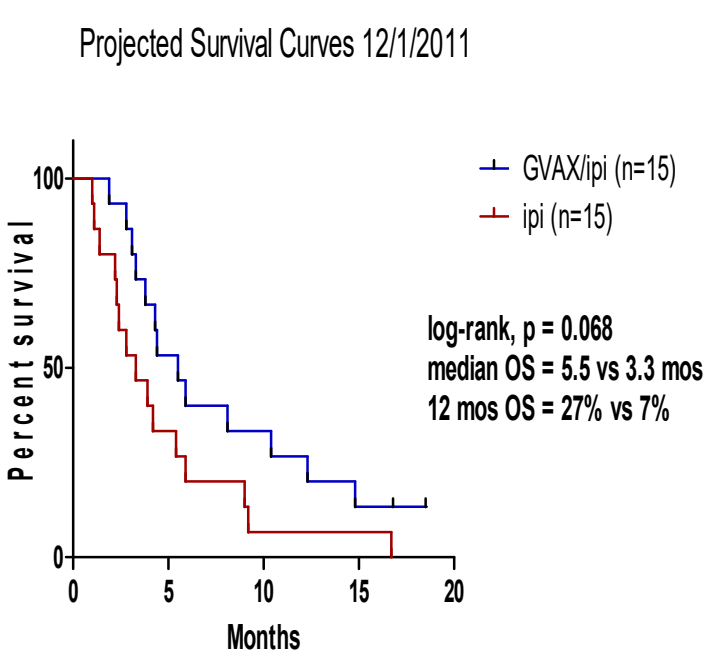
Melanie Loth, Ludmila Danilova, Janelle Montagne

NEXT STEPS: Combine Vaccines with Immune Modulators



Ipilimumab + Vaccine Improves Survival In Advanced Pancreatic Cancer Patients

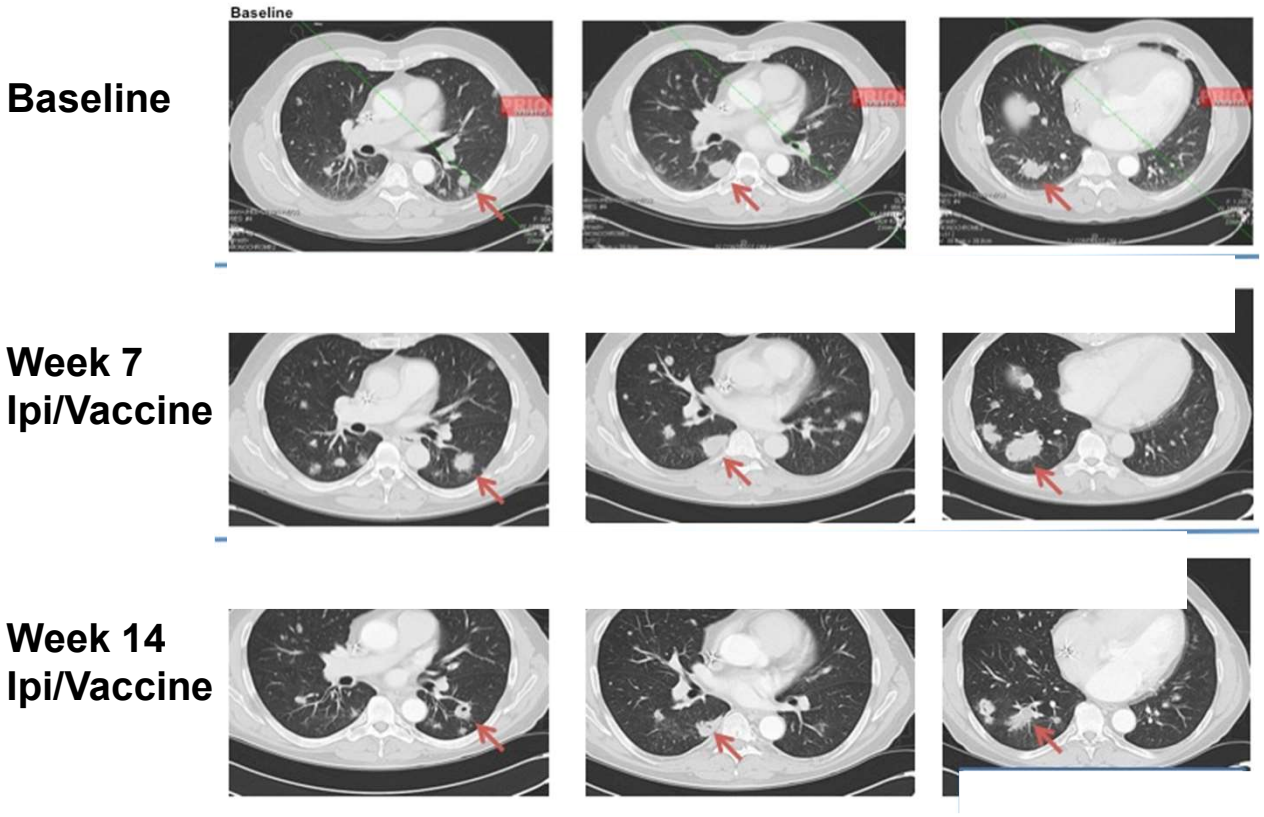
Le, et al., J Immunother 2013



Dr. Dung Le

- Metastatic patients failed >2 chemotherapies
- 7/15 patients in combo arm with clinical and/or biomarker response
- 0/15 in single Ipi arm with clinical and/or biomarker response

Radiographic Regressions After 14 Weeks Of Treatment with Ipilimumab (Ipi) + Vaccine



Mass Cytometry (CyTOF) Analysis

- Flow cytometry variant using heavy metal ion tags for antibody labeling
  - 31 unique markers with T cell focus
- Analyzed 20 patients with paired week 0 and week 7 PBL samples
  - Analyzed ~>**10 million cells**
- Analysis evaluated parameters in the context of patient clinical benefit
  - CA19.9 and CT scan data (9/20 benefited)
- Definition of Cohorts
  - **Clinical Benefit** = stable disease (did not meet recist for PR) or partial remission (met recist criteria)
  - **No Benefit** = progressive disease on therapy

Wu et al., Clin Ca Res 2020

Won Jin Ho

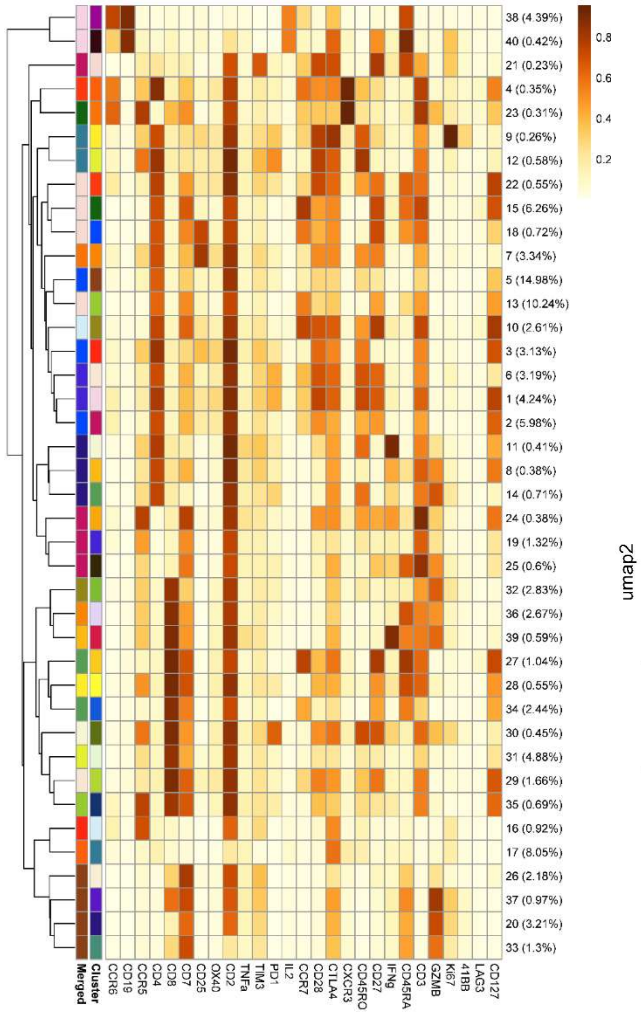
Annie Wu

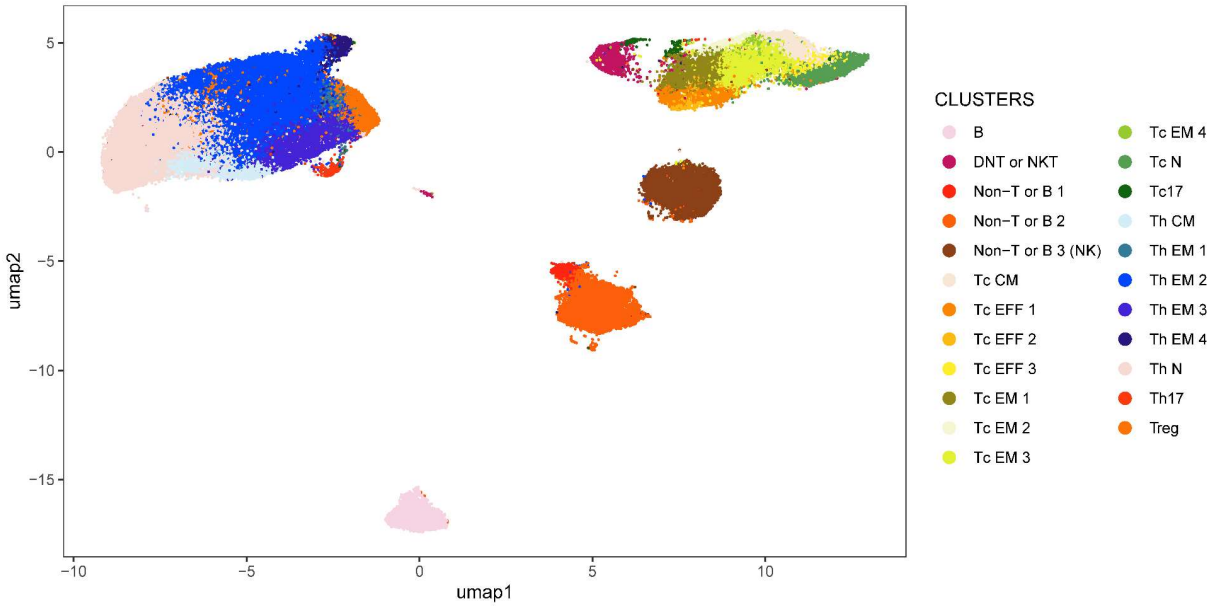
Elana Fertig

Single cell profiling identifies ipilimumab + vaccine induced signaling changes on multiple cell types

Changes demonstrate the heterogeneity of T cell populations and their functional responses

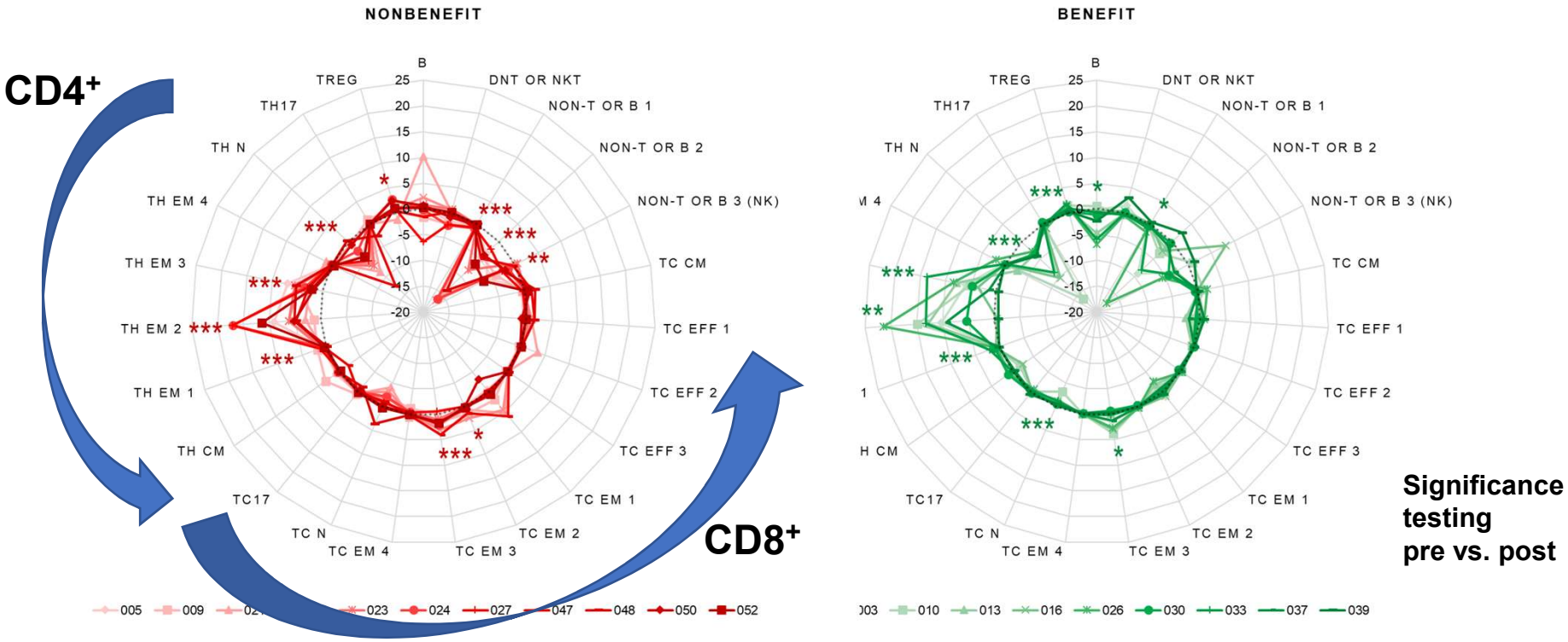
Phenograph of T cell clusters identified by FlowSOM algorithm: UMAP visualization confirms appropriate clustering





Wu et al., Clin Ca Res 2020

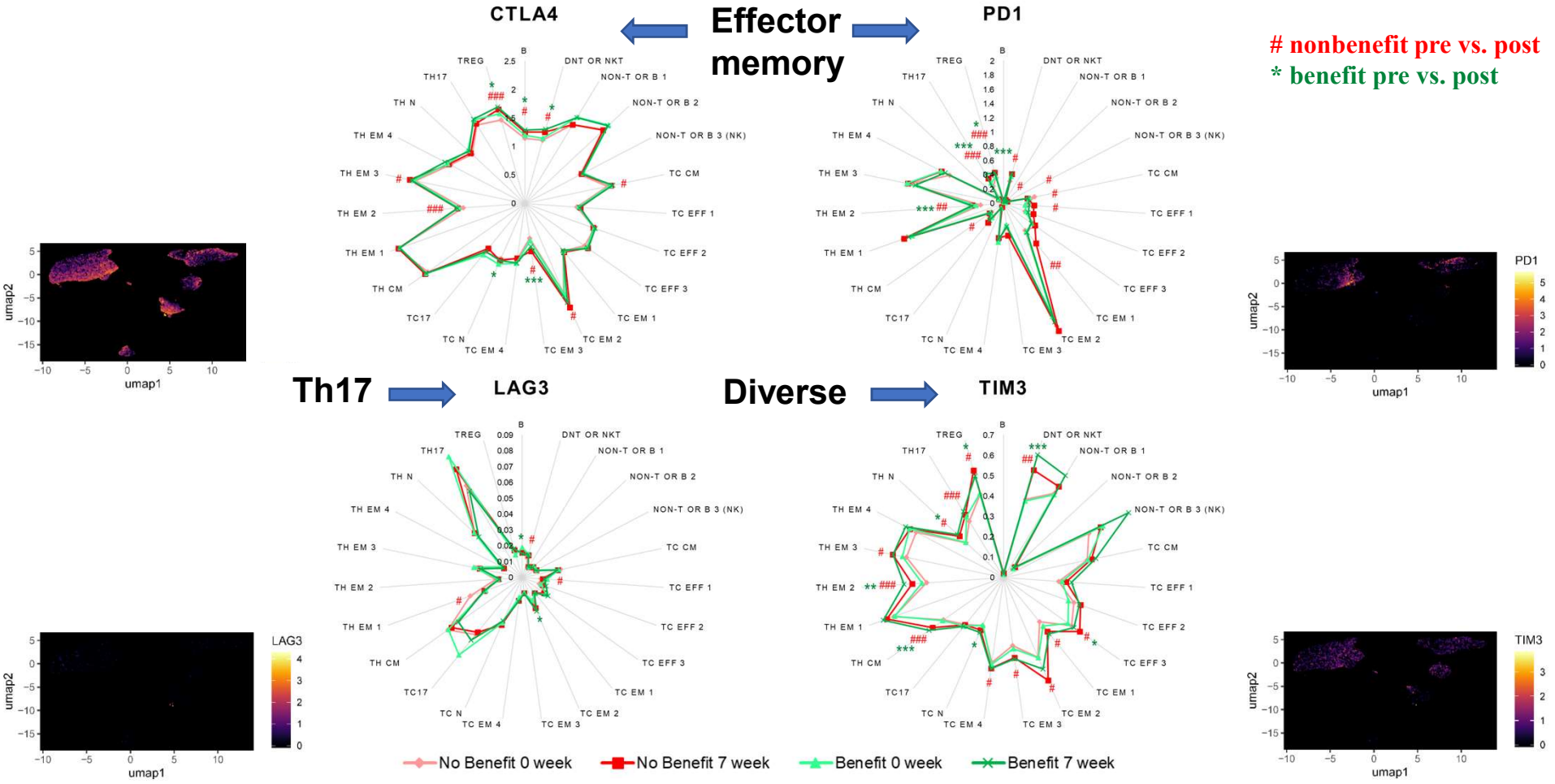
Ipilimumab + GVAX significantly promotes differentiation toward memory away from naïve in T cells regardless of clinical benefit status



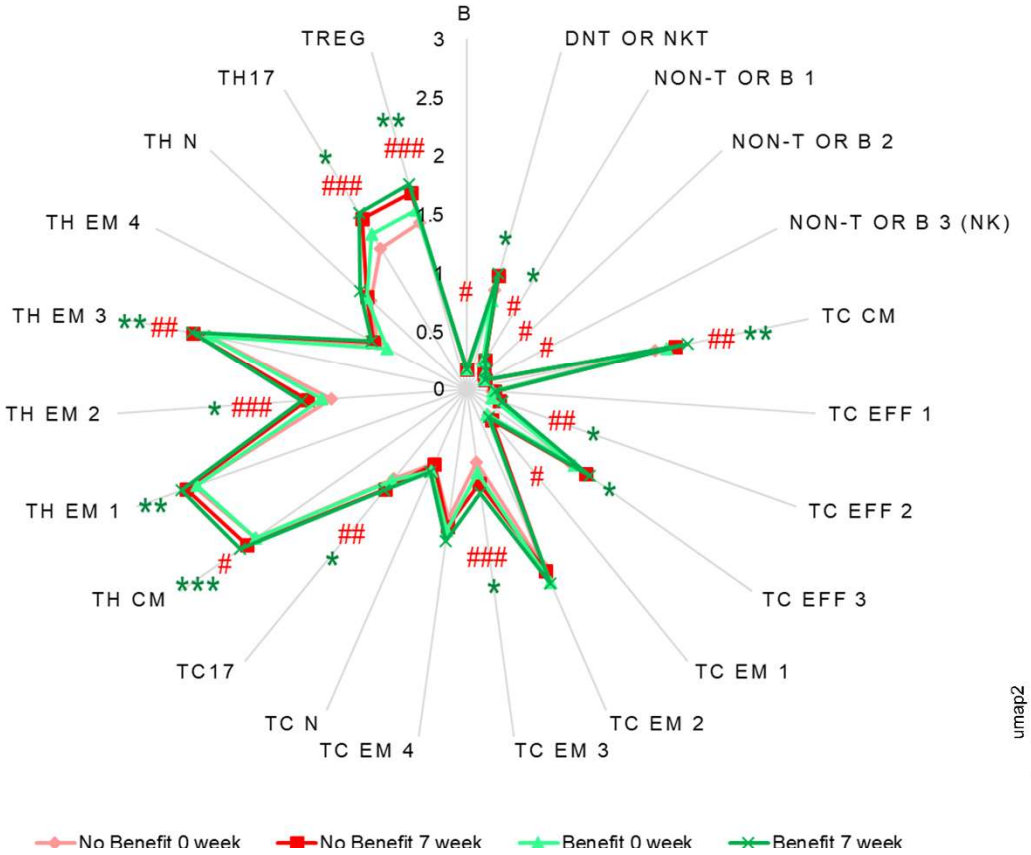
Significance testing pre vs. post

Radar plots showing differences in proportions (CD45%) of immune cell types between week 7 and baseline for individual patients

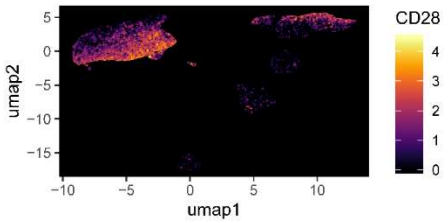
Upregulation of checkpoint expression with Ipi+GVAX is similar regardless of clinical benefit



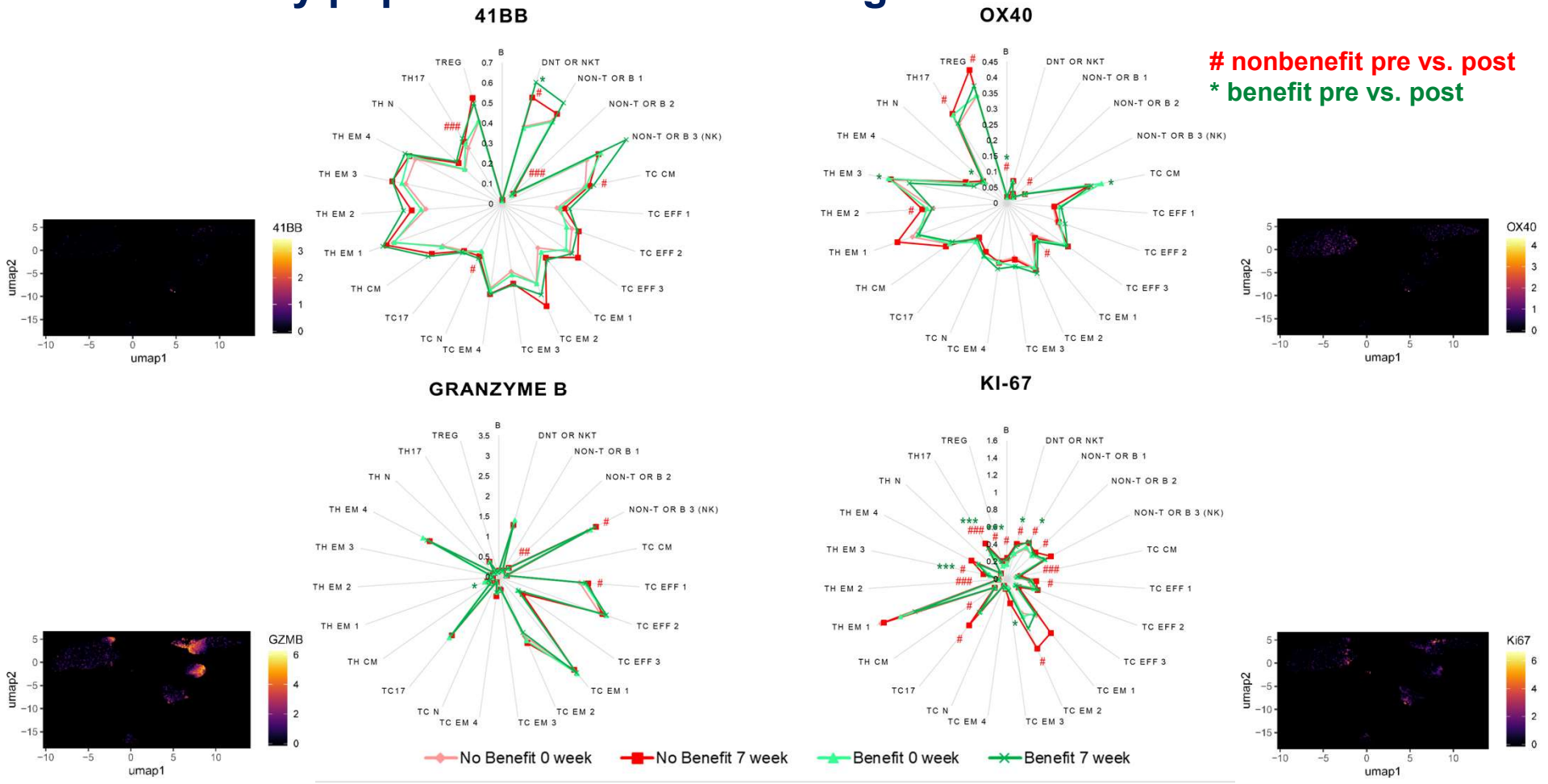
Upregulation of CD28 expression by Effector T cells, EM and CM T cells, and regulatory T cell populations regardless of clinical benefit



 # nonbenefit pre vs. post
*** benefit pre vs. post**

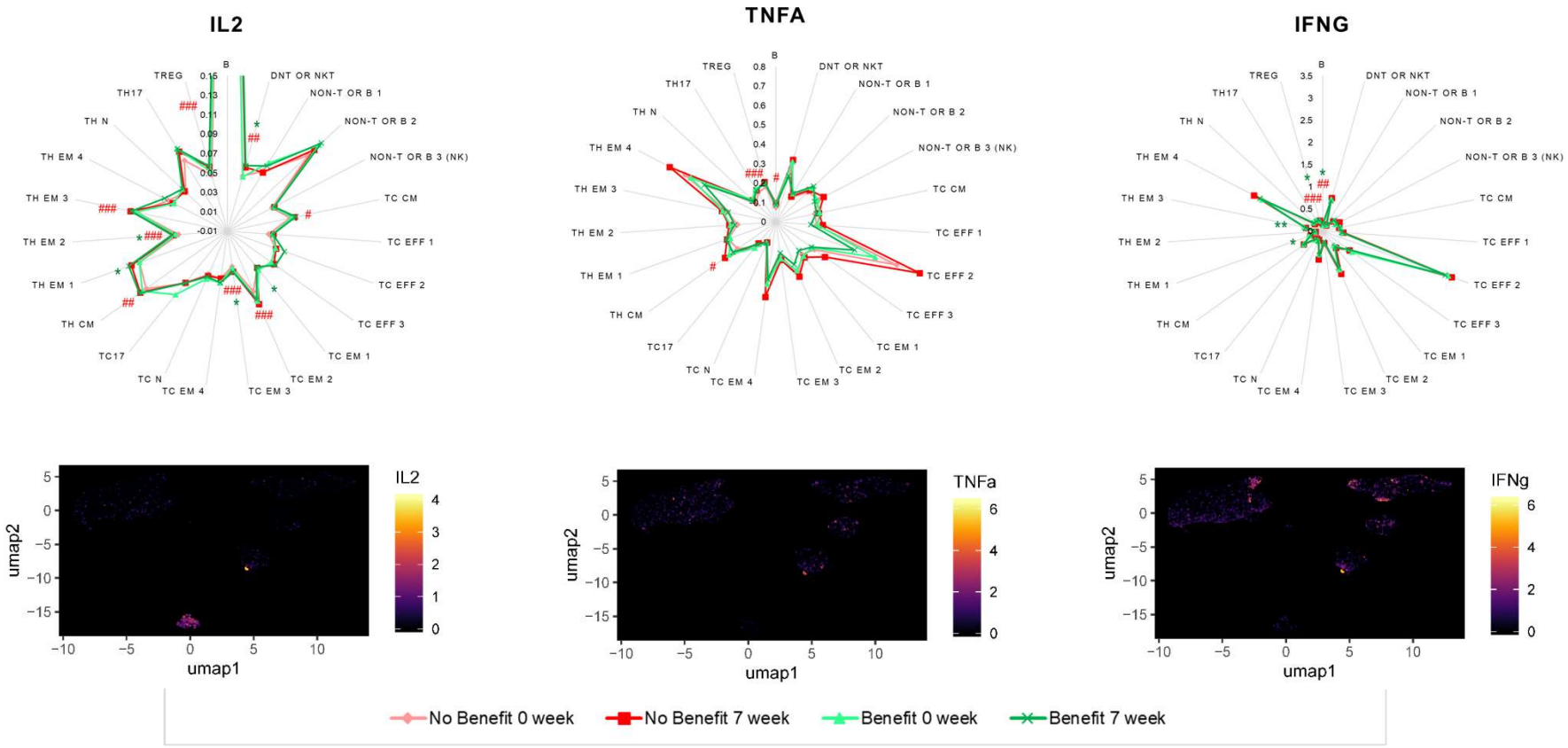


Upregulation of costimulatory and activation markers in effector memory populations are similar regardless of clinical benefit



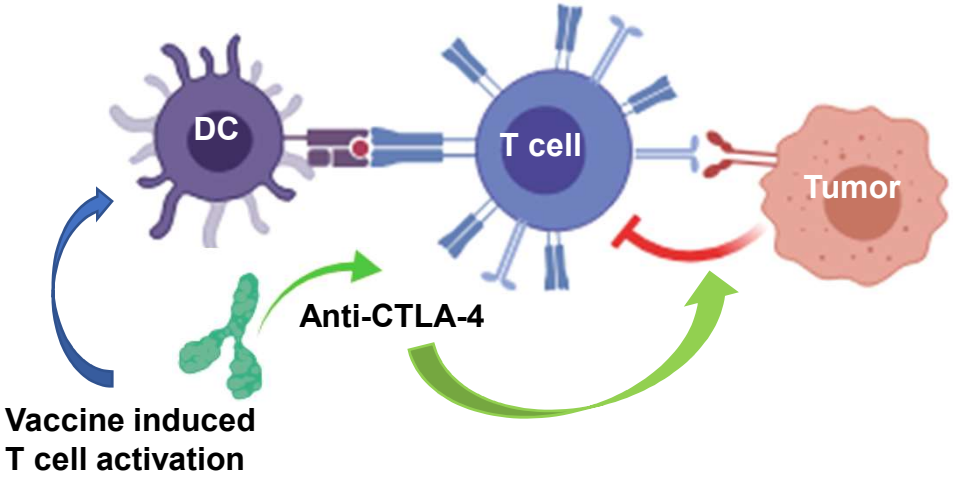
Changes in effector cytokine production in effector and effector memory T cells are similar regardless of the clinical benefit

 # nonbenefit pre vs. post
*** benefit pre vs. post**

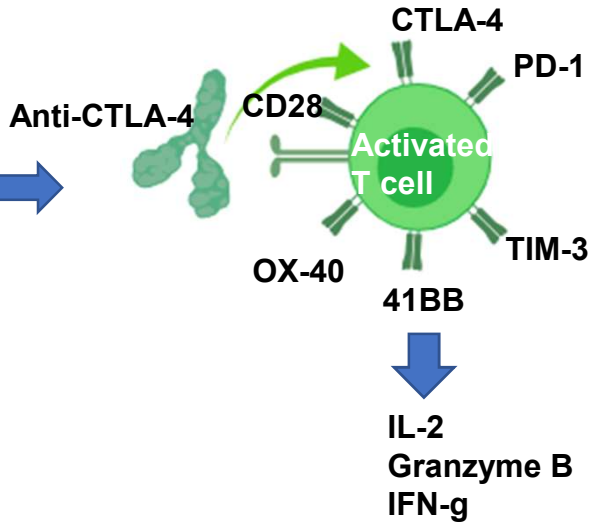


Proposed mechanism of CTLA-4 T cell activation

Naïve T cell converts to antigen experienced T cells



Upregulation of T cell activation and regulatory markers



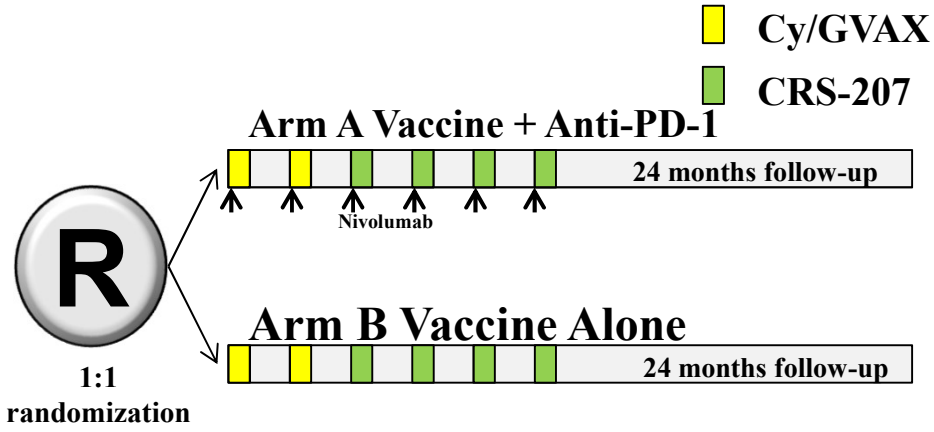
- T cell changes similar regardless of clinical benefit
- These changes suggest simultaneous activation of effector and regulatory T cell populations

Summary:

- Blocking CTLA-4 on vaccine-inducing cancer specific T cells results in upregulation of both activating and regulatory checkpoint signals
- This is an expected and compensatory mechanism to prevent normal tissue destruction
- Additional signal modifications are needed to maintain active T cells until all tumor cells are gone

GVAX + CRS-207 Heterologous Prime Boost Vaccination with Programmed Death-1 (PD-1) Blockade

Patients with metastatic pancreatic cancer; progresssing after 1 prior chemotherapy for metastatic disease



Dung Le

88 patients with previously treated metastatic pancreatic cancer randomized 1:1 to 2 treatment arms

GVAX + CRS-207 Heterologous Prime Boost Vaccination with Programmed Death-1 (PD-1) Blockade

Tsujikawa T et al, Clin Ca Res 2020

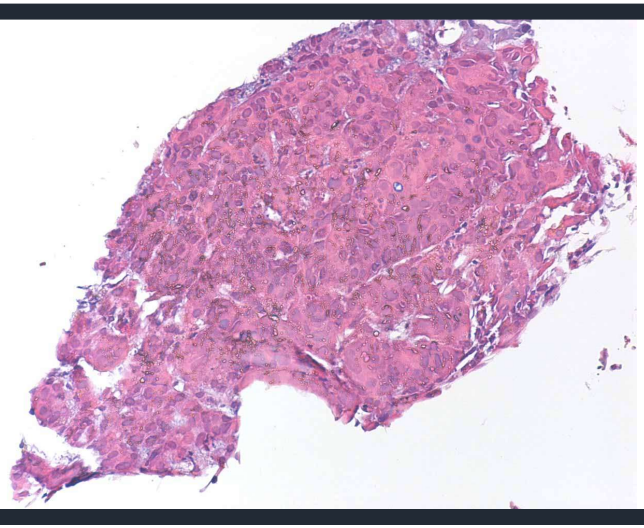
Dung Le

Baseline | Week 10 | Week 30

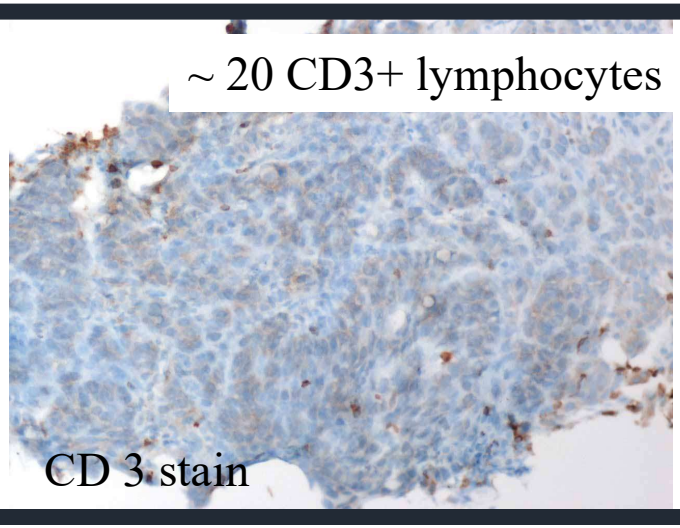
Pre-treatment Biopsy

During treatment Biopsy

Pre-treatment

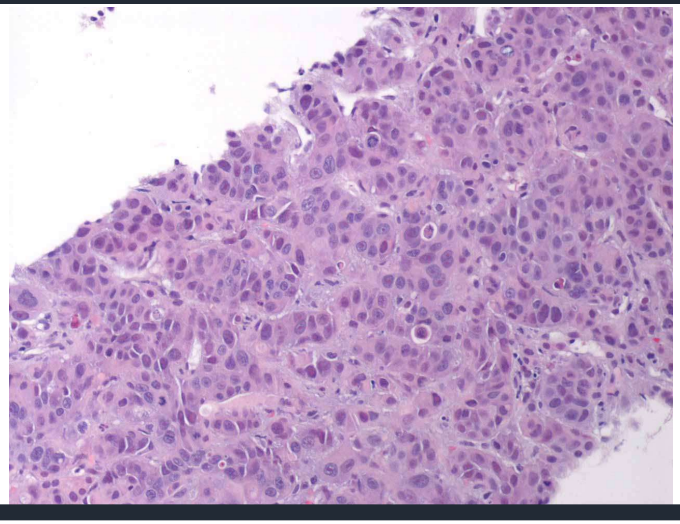


~ 20 CD3+ lymphocytes

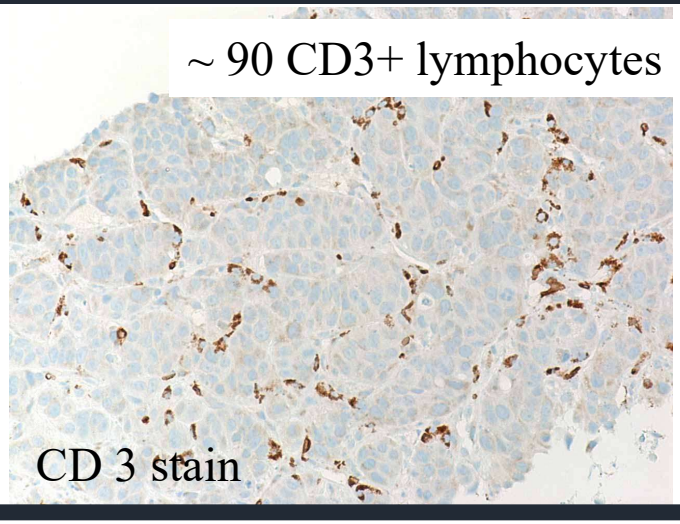


CD 3 stain

Post-treatment



~ 90 CD3+ lymphocytes



CD 3 stain

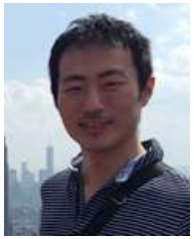
Robert Anders

Multiplex Immunohistochemistry

- Analyzed 35 patients with paired week 0 and week 10 biopsy samples
  - 3 panels, 8 markers per slide

- Analysis evaluated parameters in the context of patient clinical benefit
  - CA19.9 and CT scan data and overall survival

Lisa Coussens

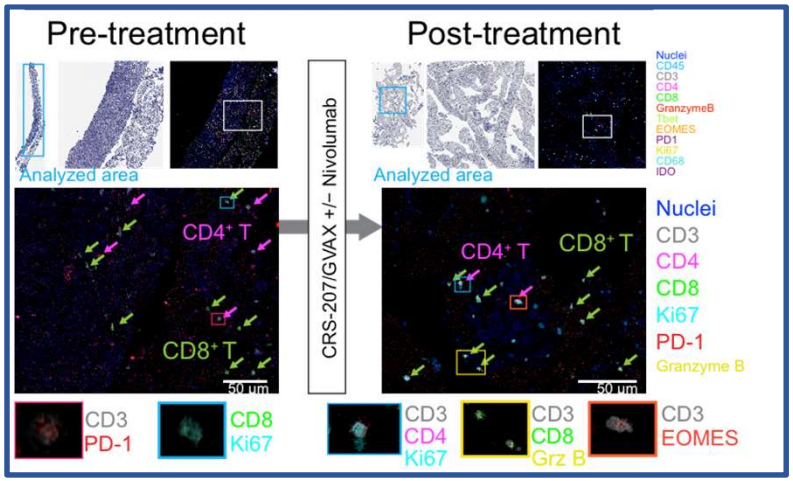


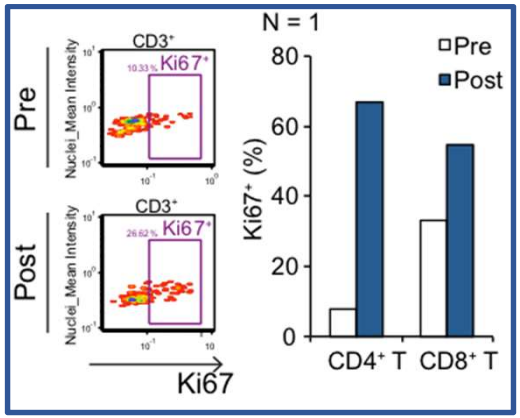
Takahiro Tsujikawa

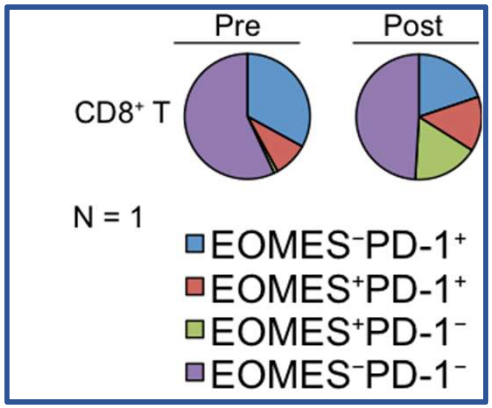
Le, Tsujikawa T, et al. Clin Ca Res, 2020

Multiplex IHC depicts evidence of T cell reinvigoration with GVAX/CRS207 + nivolumab in a responder

Biopsy specimen



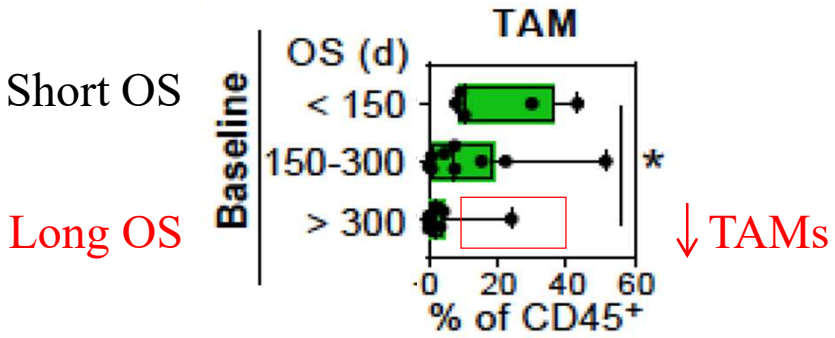


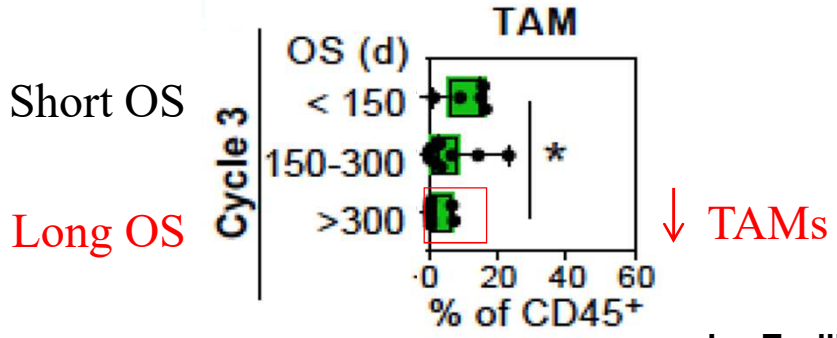


Post-vaccine increased EOMES expression which enhances T cell infiltration and is associated with a less exhaution phenotype for $CD8^+$ T cells

Le, Tsujikawa T, et al. Clin Ca Res, 2020

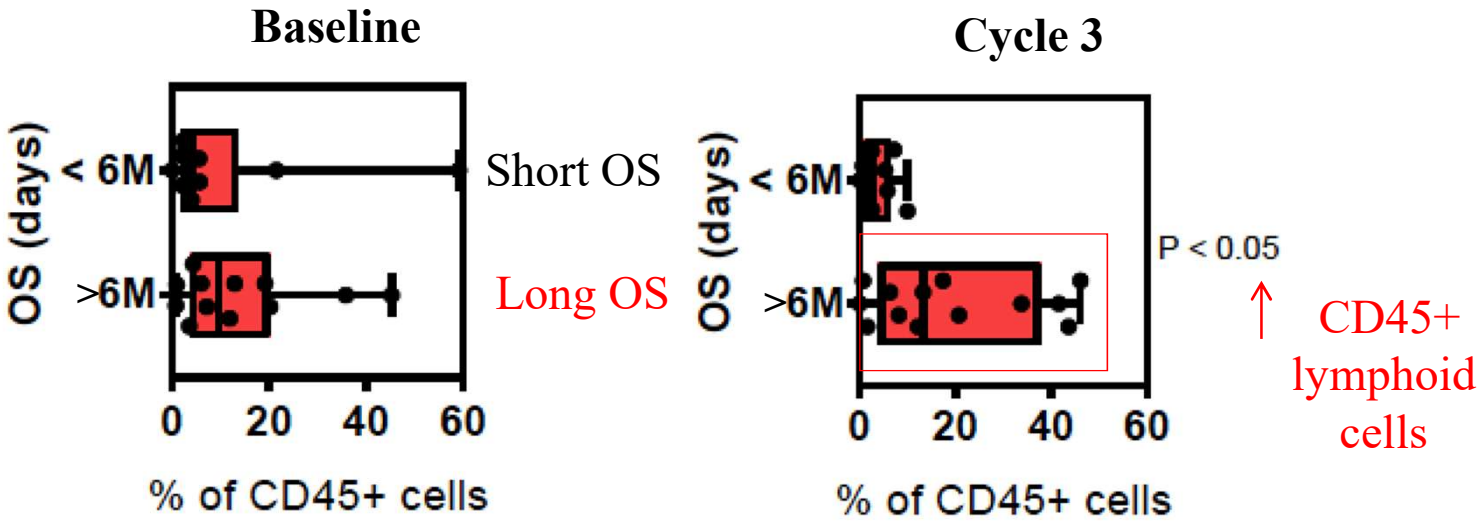
Fewer tumor associated macrophages (TAMs) at baseline and post-treatment correlates with longer overall survival





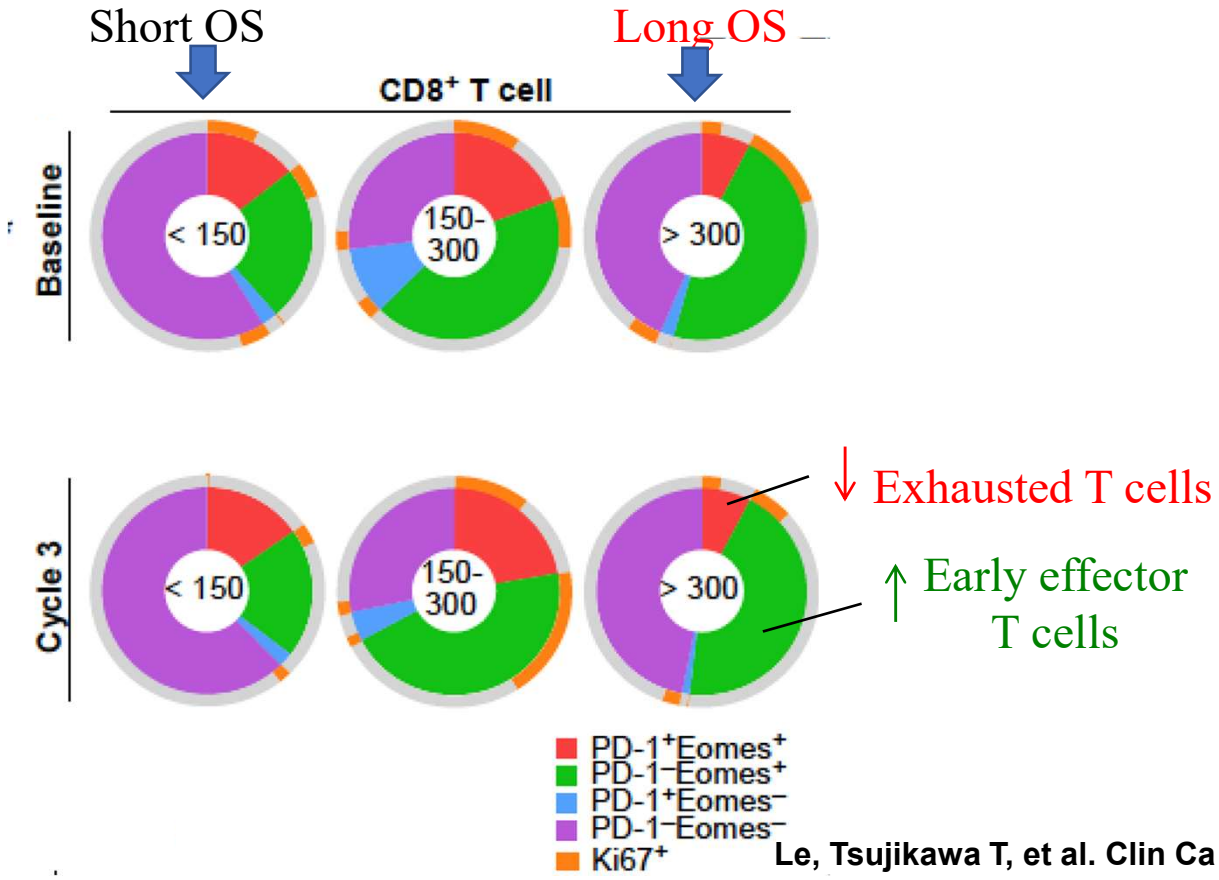
Le, Tsujikawa T, et al. Clin Ca Res, 2020

Longer overall survival correlates with high CD45+ lymphoid cell numbers detected after treatment



Le, Tsujikawa T, et al. Clin Ca Res, 2020

Less exhausted and more effector memory CD8⁺ T cells during treatment is associated with longer overall survival



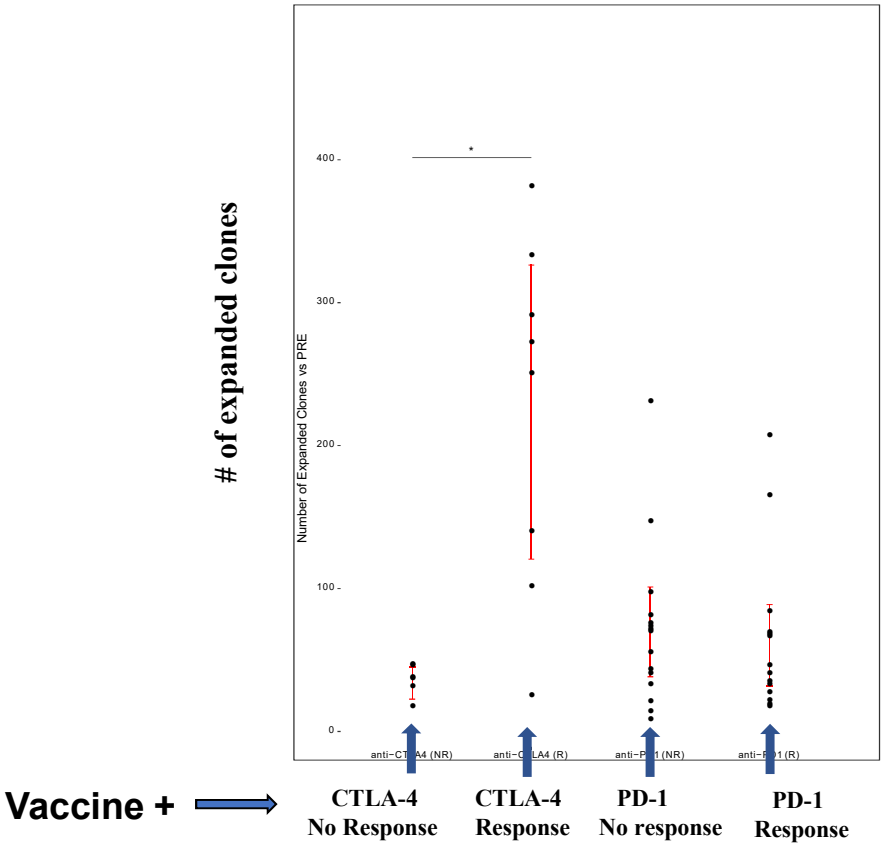
Le, Tsujikawa T, et al. Clin Ca Res, 2020

Next Steps: Can We Increase the Response Rate and Time to Response?

Sequential T cell receptor sequencing of PBL can predict immunotherapy responders

- PBL evaluated pre- and during treatment from 2 studies
- Patients treated with ipilimumab alone or with ipilimumab + vaccine
- Patients treated with vaccine alone or with vaccine + nivolumab

αCTLA-4 responders had significantly more expanded clones than non-responders (pre- vs post-treatment)

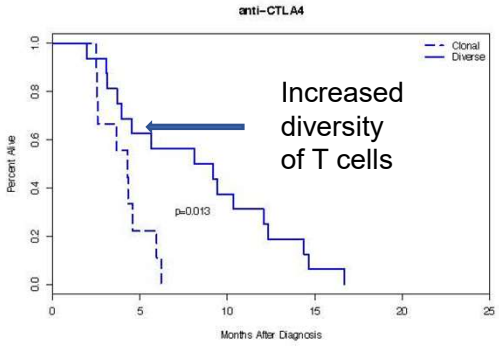


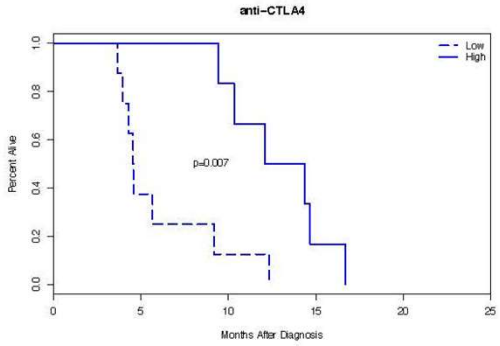
Alex Hopkins
JCI Insights, 2018

Kaplan Meier survival curves based on TCR clonality status or number of expanded clones

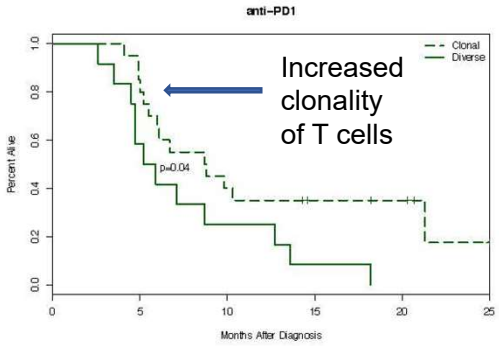
Clonality | **Expanded Clone #**

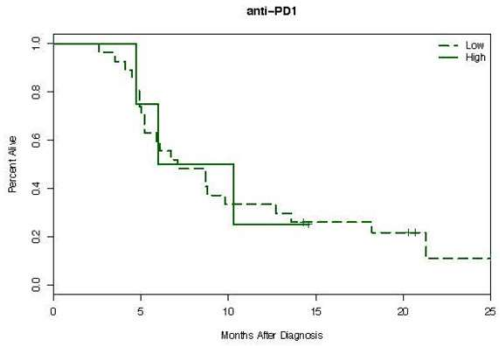
Anti-CTLA4





Anti-PD-1



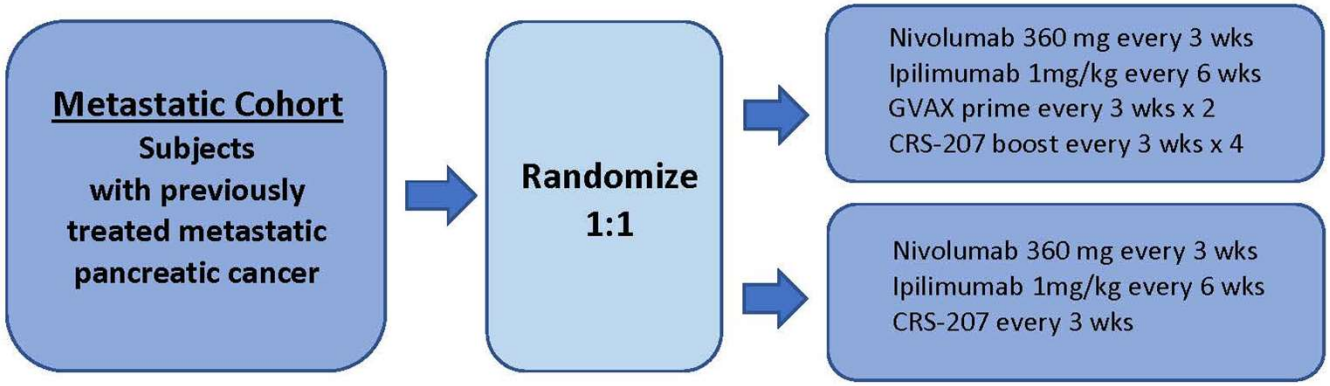


>100 expanded clones

<100 expanded clones

Study designed based on TCRseq Data
Currently Enrolling

Nivolumab (anti-PD-1) plus ipilimumab (anti-CTLA-4) and Listeria-mesothelin (CRS-207) with or without GVAX pancreatic cancer vaccine in patients with pancreatic adenocarcinoma



PANCREATIC CANCER ACTION NETWORK

ADURO BIOTECH

Bristol-Myers Squibb

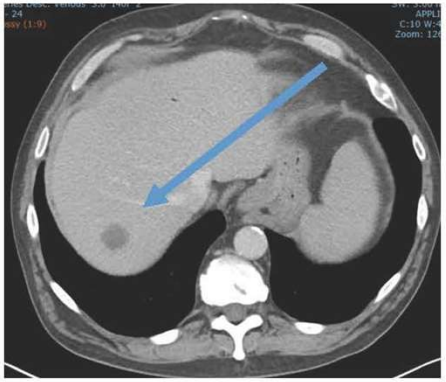
Addition of ipilimumab has increased rate and number of early responders

Responses seen in liver and peritoneal implants with combination Ipi and Nivo

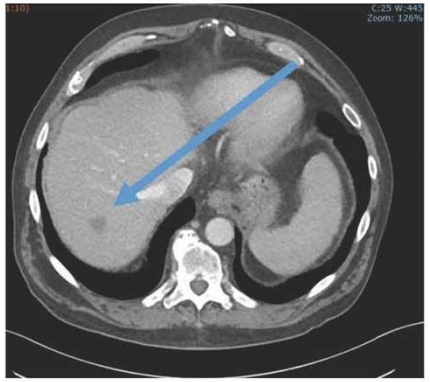
Nivo alone did not affect liver mets in pancreatic cancer patients

Dr. Dung Le

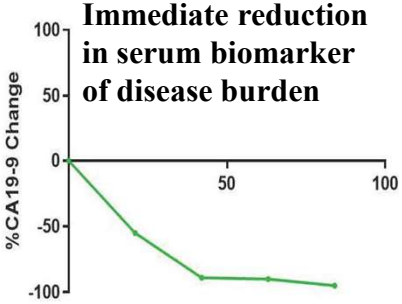
Pre-treatment



Post-2 treatment cycles
6 weeks after starting therapy



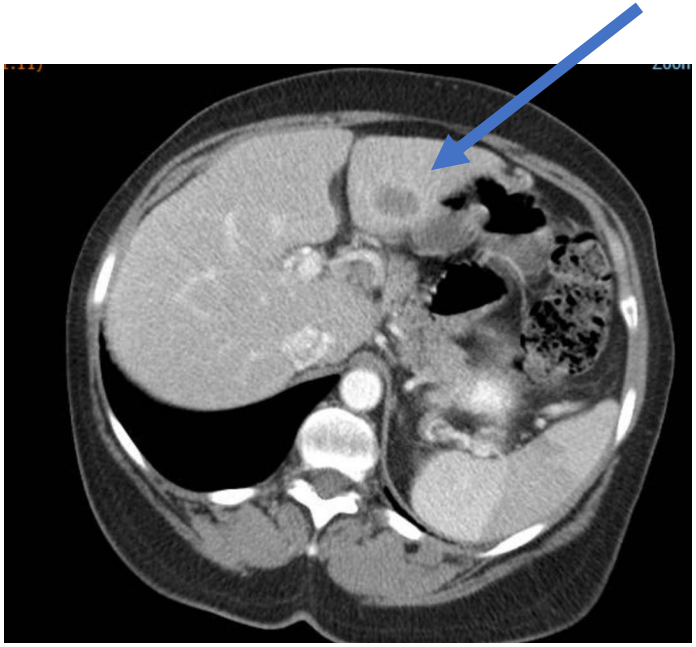
Immediate reduction in serum biomarker of disease burden

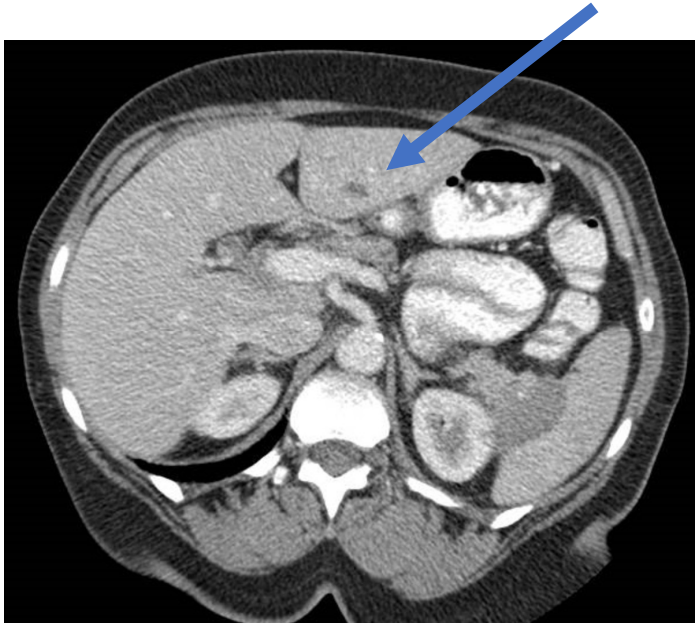


8 of 20 evidence of response so far

DO NOT POST

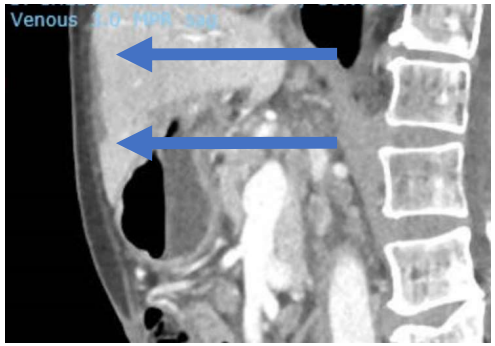
Vaccine + Ipilimumab + Nivolumab

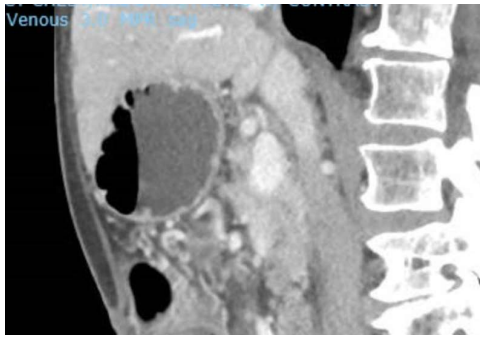


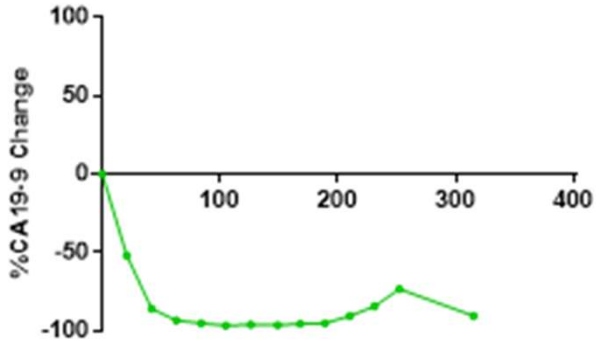


DO NOT POST

Vaccine + Ipilimumab + Nivolumab



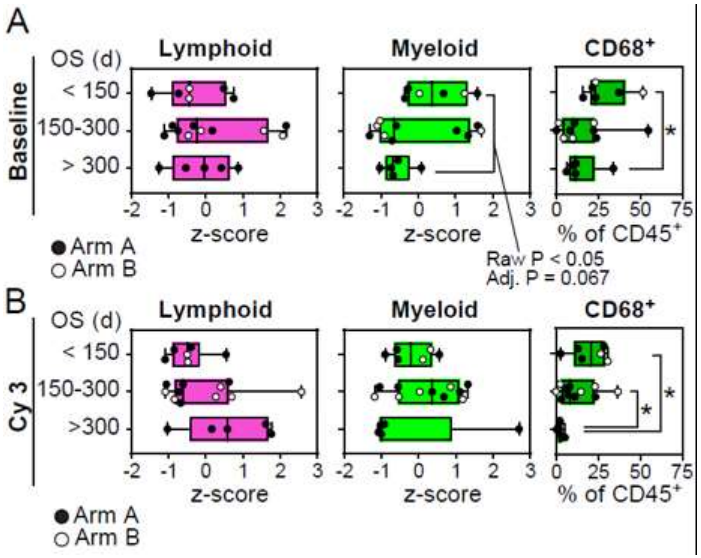


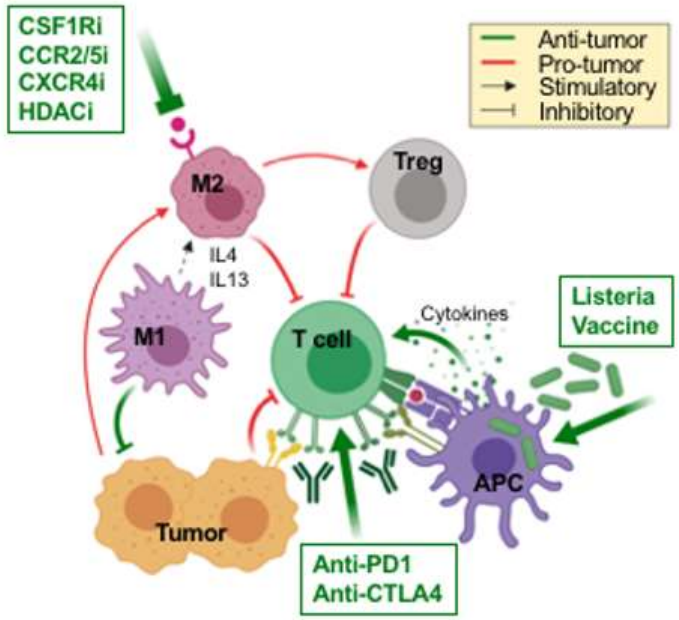


Biopsy proven implants

DO NOT POST

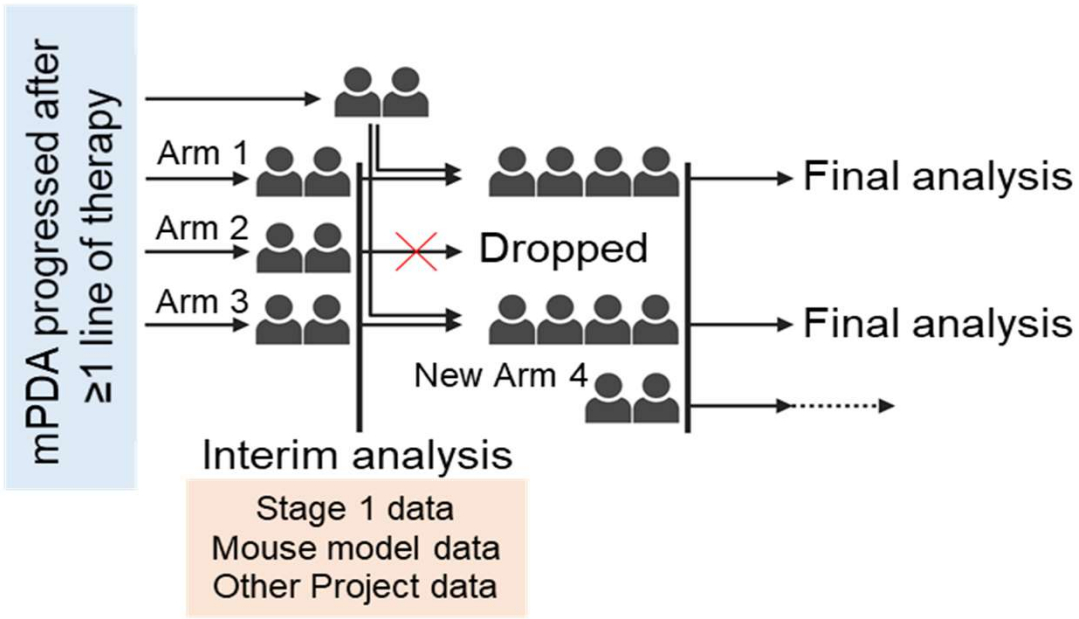
Next Steps: Myeloid Reprogramming

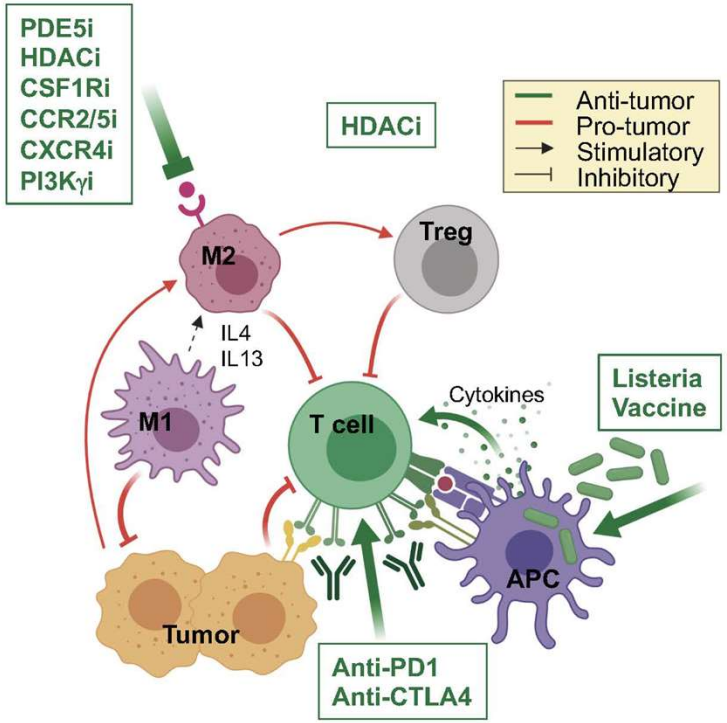




Won Ho & Katie Bever

ONGOING PLATFORM CLINICAL TRIAL DESIGN





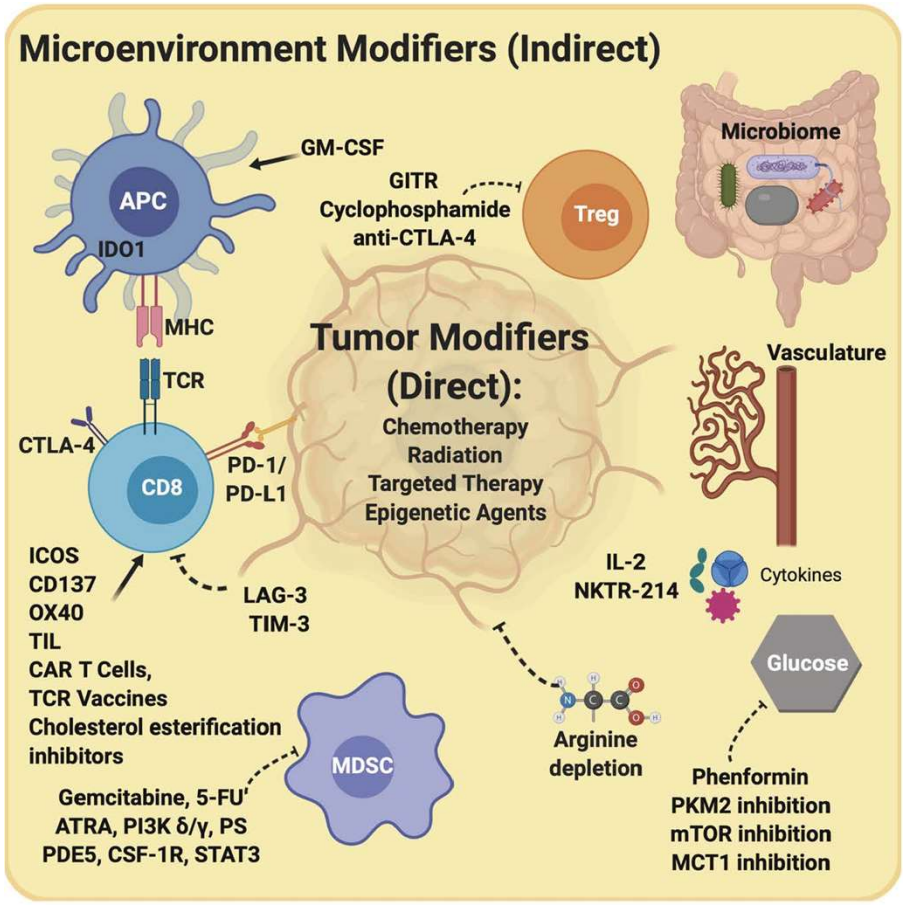
The Future of Cancer Immunotherapy: Micro-environment targeting combinations

Antigen processing and presentation

Optimizing T cell signaling

Adoptive T cell Therapy/CAR T

Reprogram Monocytes And fibroblasts



Murciano-Goroff, et al, Cell Research, 2020, 30:507-519

Reconstitute the microbiome

Modify the vasculature

Optimize metabolic imbalances

Assess heterogeneity

Scientific Partners

Dan Laheru
Dung Le
Lei Zheng
Elana Fertig
Luciane Kagohara
Todd Armstrong
Bob Anders
Melanie Loth
Guanlan Mo
Mark Yarchoan
Alex Hopkins
Nilo Azad
Katie Bever

Chris Wolfgang
Ralph Hruban
Rick Burkhardt
Carol Judkins
Jin He
Katrina Alino
Beth Onners
Jennifer Durham
Rose Parkinson

Lisa Coussens
Takahiro Tsujikawa

NCI GI Spore

NCI RO1

Skip Viragh Pancreatic Cancer Center

Aduro Biotech

PANCAN AACR

BMS and Merck

Bloomberg-Kimmel Institute

CURE PANCREATIC CANCER
• LUSTGARTEN FOUNDATION •

ADMINISTERED BY
AACR American Association for Cancer Research
FINDING CURES TOGETHER
SCIENTIFIC PARTNER OF STAND UP TO CANCER

STAND UP TO CANCER