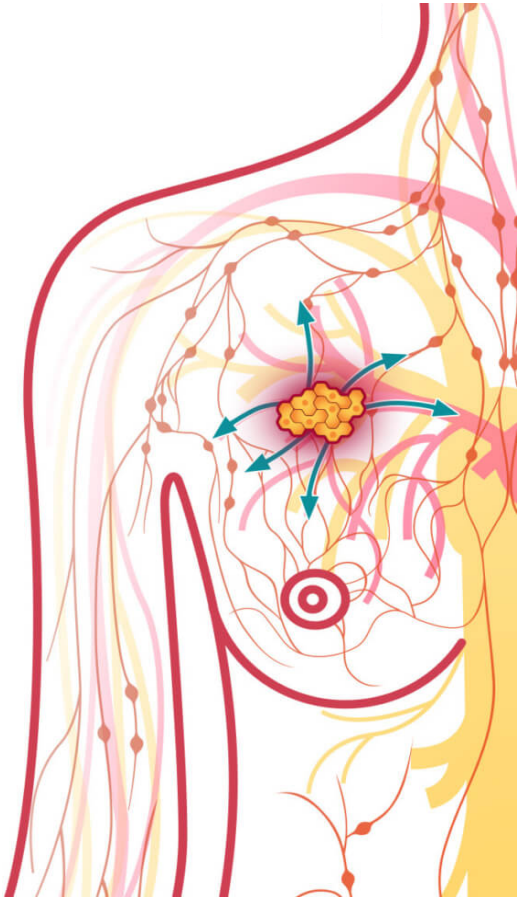
Tumor-Immune Interactions in Triple Negative Breast Cancer Brain Metastases

Work in Progress: Building Our Patient Cohort



- **Triple Negative Breast Cancer**
- **Breast-to-Brain Metastases**
  - TNBC
  - Leptomeningeal Disease (LMD)
- **High-level Overview of the Project**
  - Rationale
  - Patient Cohort
- **Next Steps**
- **Future Directions**

Maxine Umeh-Garcia, PhD, MSc.
SCIT T32 Seminar
Hayden Gephart and Plevritis Labs
April 7th 2021

Breast Cancer

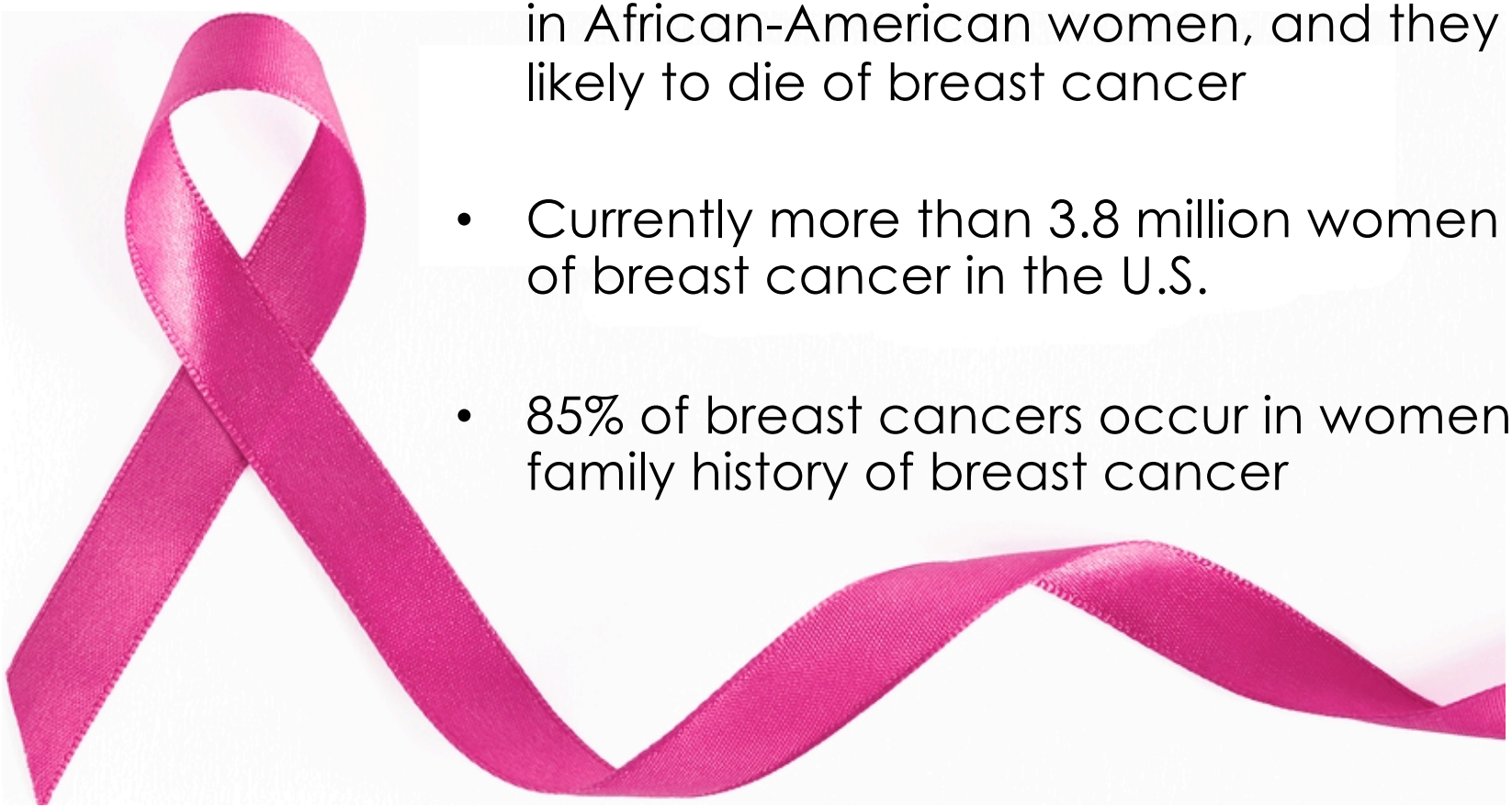
- 1 in 8 women in the U.S. will develop invasive breast cancer

- In 2021, an estimated 281,550 new cases (invasive) and 42,290 (non-invasive) breast cancer are expected to be diagnosed in women in the U.S., of which about 43,600 women are expected to die

- In women under 45, breast cancer is most common in African-American women, and they are more likely to die of breast cancer

- Currently more than 3.8 million women with a history of breast cancer in the U.S.

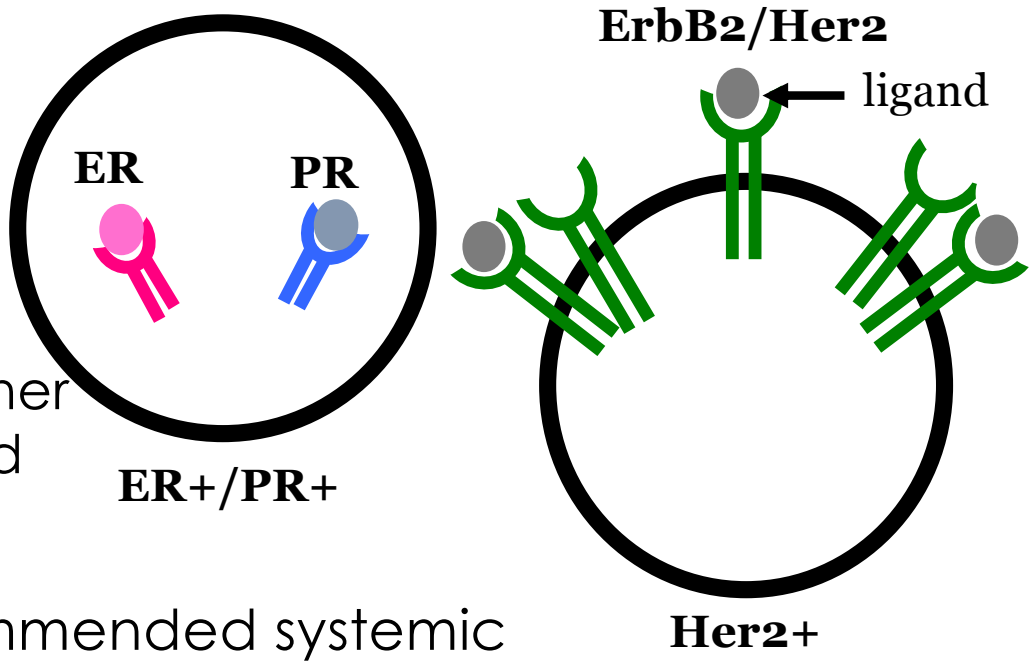
- 85% of breast cancers occur in women who have no family history of breast cancer



SEER.CANCER.GOV

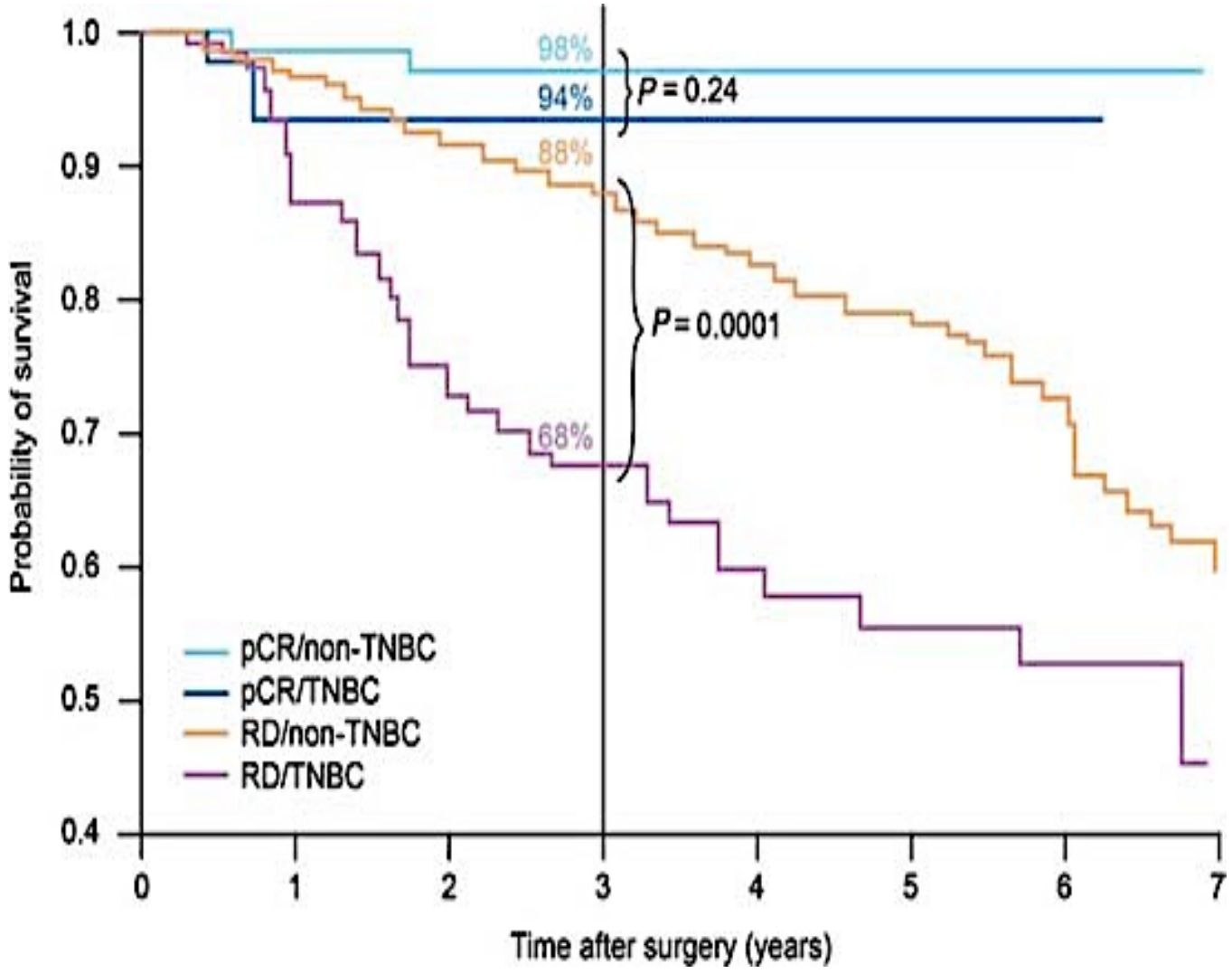
Triple Negative Breast Cancer (TNBC)

- TNBC is a heterogeneous group of tumors simply defined by the absence of estrogen **(ER)** and progesterone **(PR)** hormone receptors, and lack of overexpression of epidermal growth factor receptor 2 **(ErbB2/Her2)** gene



- TNBC account for **10-20%** of all invasive breast cancers
- TNBC is associated with African-American race, younger age, higher tumor grade, and more advanced tumor stage at diagnosis
- Chemotherapy is the **only** recommended systemic treatment, however ***only 30%*** of TNBC patients achieve pCR. Patients who do not have ***6-fold*** higher risk of relapse, and ***12-fold*** higher risk of death
- Survival at 3 yrs is lower (68%) for metastatic TNBC patients compared to other metastatic breast cancer types (88%)

Triple Negative Breast Cancer (TNBC)



Liedtke C, Mazouni C, Hess KR et al.

TNBC in African-American Women

- Women of African ancestry have a disproportionately **higher frequency** (up to 79%) of TNBC, compared to women of European ancestry
  - TNBC frequency is consistently higher in women of African ancestry than **any other racial/ethnic group**

- *But what about socioeconomic factors?*
  - Incidence and patient outcomes have historically been ascribed to socioeconomic factors, particularly lack of access to healthcare and screenings, and distrust of medical professionals.

  - 5-yr distant relapse-free survival is 62.8% for young black women, vs. 77% for young white women with equal access to health care

- Black women exhibit a significantly higher incidence of (and mortality from) metastatic breast cancer including breast-to-brain metastasis

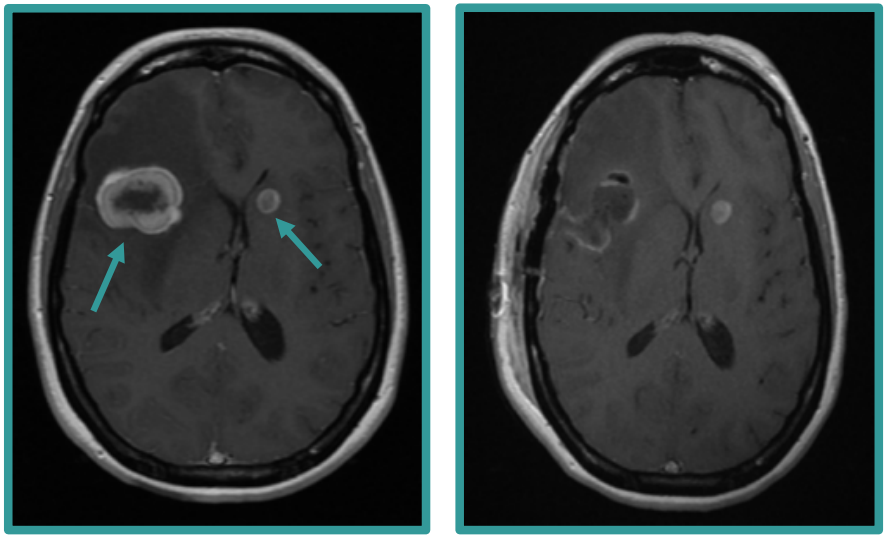
- Emerging evidence suggest there may be poorly defined race/ethnicity-related factors (in the tumor microenvironment) that contribute to this disproportion

Breast Cancer Brain Metastasis

- Breast cancer brain metastasis (BCBM) occurs in **10-30% of metastatic breast cancer patients**
  - Second leading cause of brain metastases following lung cancer

- Incidence of BCBM continues to increase
  - Prolonged patient survival
  - Improved imaging techniques

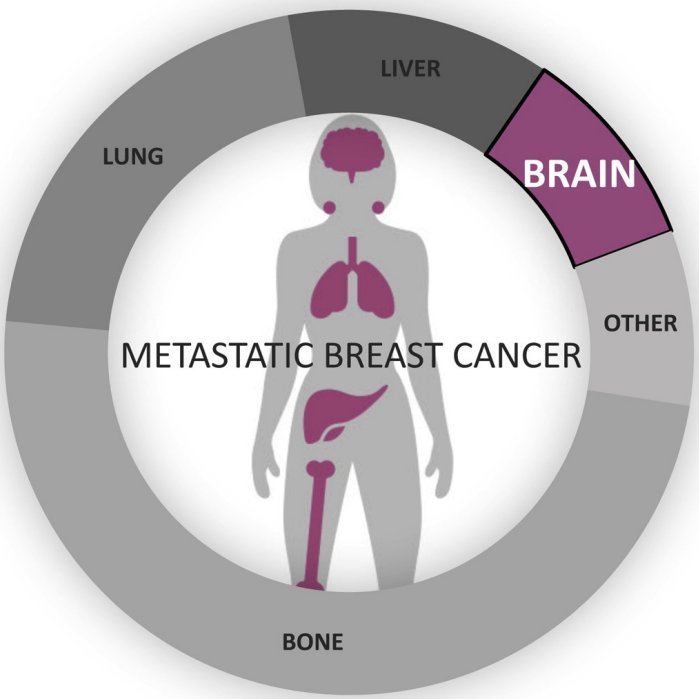
- Median survival ranges from 2 – 25.3 months
  - **Few patients survive past 1 year**
  - Associated with serve neurological decline

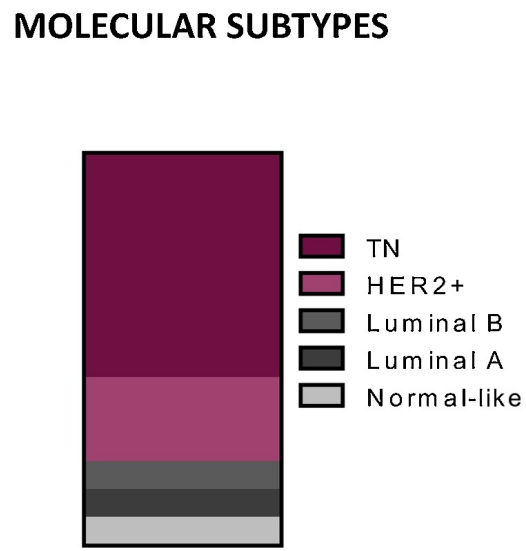


Before and After Surgical Resection

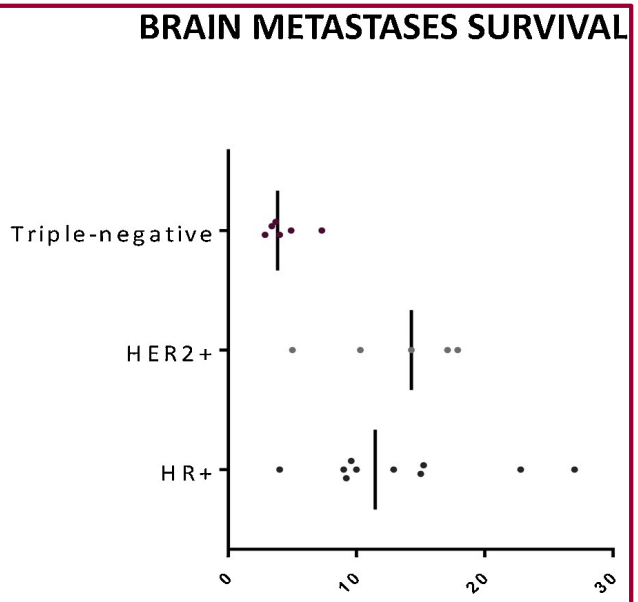
Breast Cancer Brain Metastasis

- **BCBM Incidence and Survival is breast cancer subtype dependent**





Breast Cancer Brain Metastases Incidence



Months of survival after diagnosis of Brain Metastases

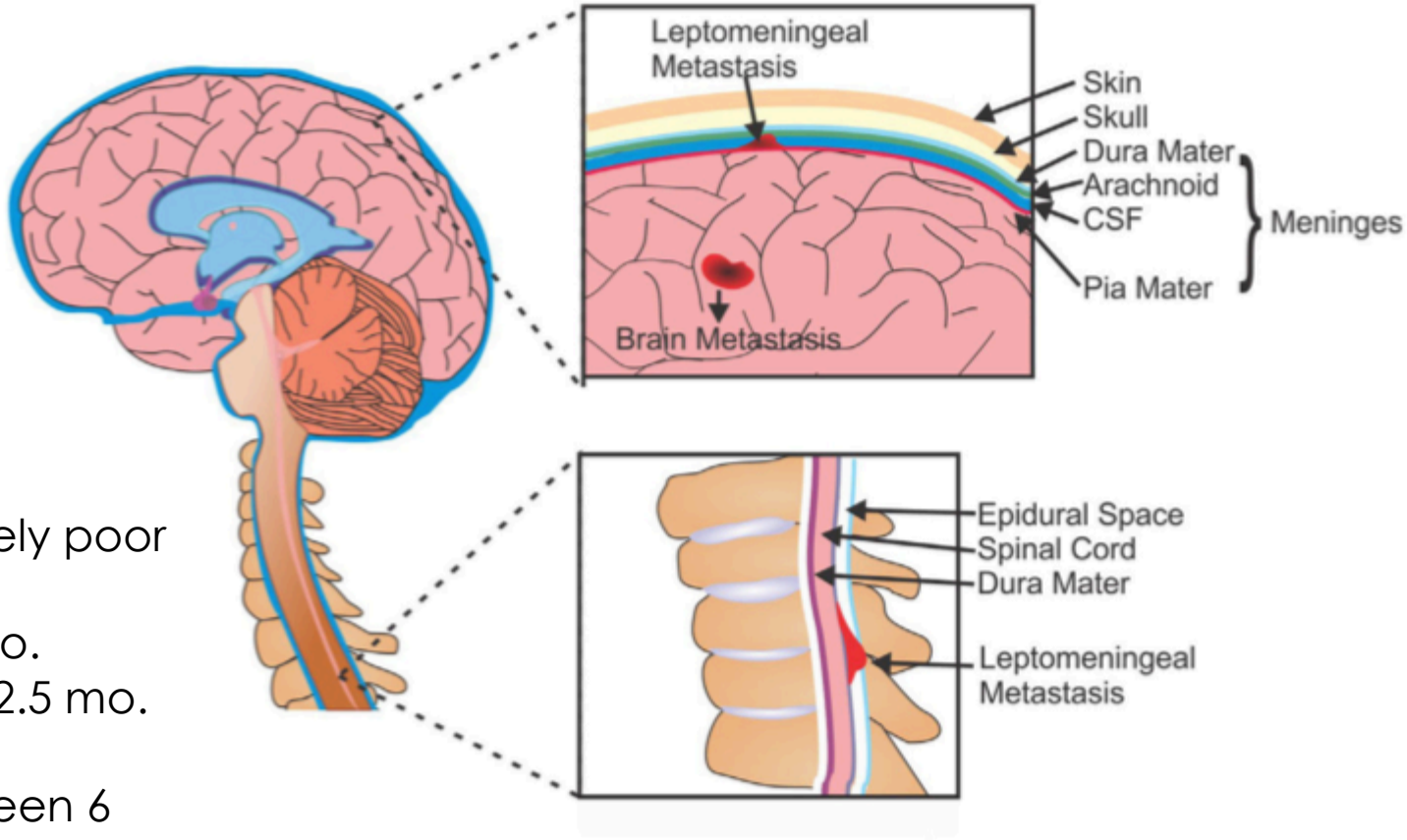
- **Current treatment strategies:**
  - Surgical resection
  - Whole brain radiation therapy (WBRT)
  - Stereotactic Radiosurgery
  - Chemotherapy
  - Targeted therapies (HR+: Tamoxifen, HER2+: Trastuzamab)

- **Major challenge in treating BCBMs is the Blood-Brain-Barrier**

- **Ongoing clinical trails…but no FDA-approved systemic treatments for BCBM**

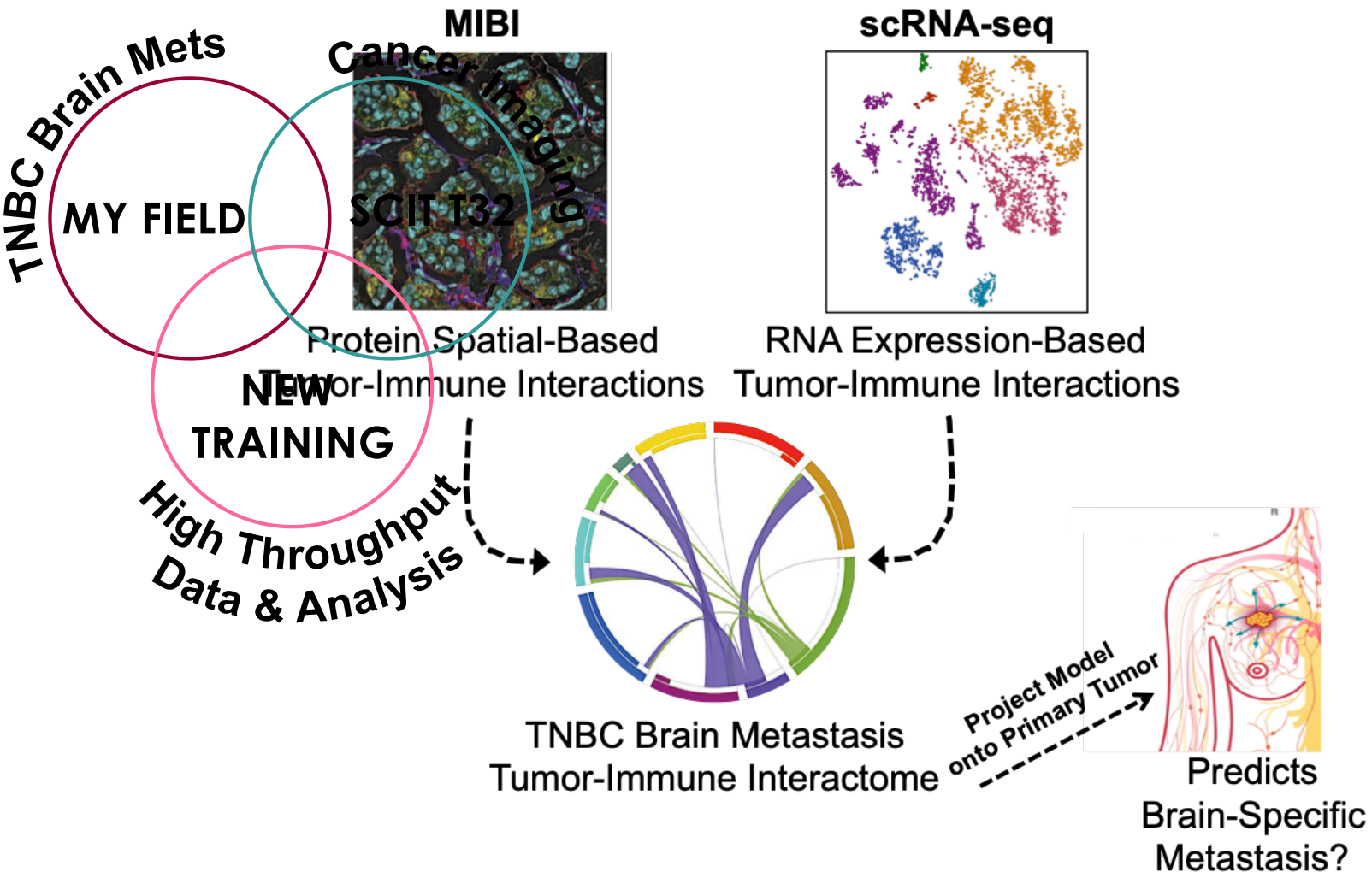
Leptomeningeal Disease (LMD)

- LMD is defined as **tumor spread within the leptomeninges** and subarachnoid space
- **10% of patients** with solid cancers present with LMD
- **Breast (TNBC), lung, and melanoma** are most common primary sites in LMD patients



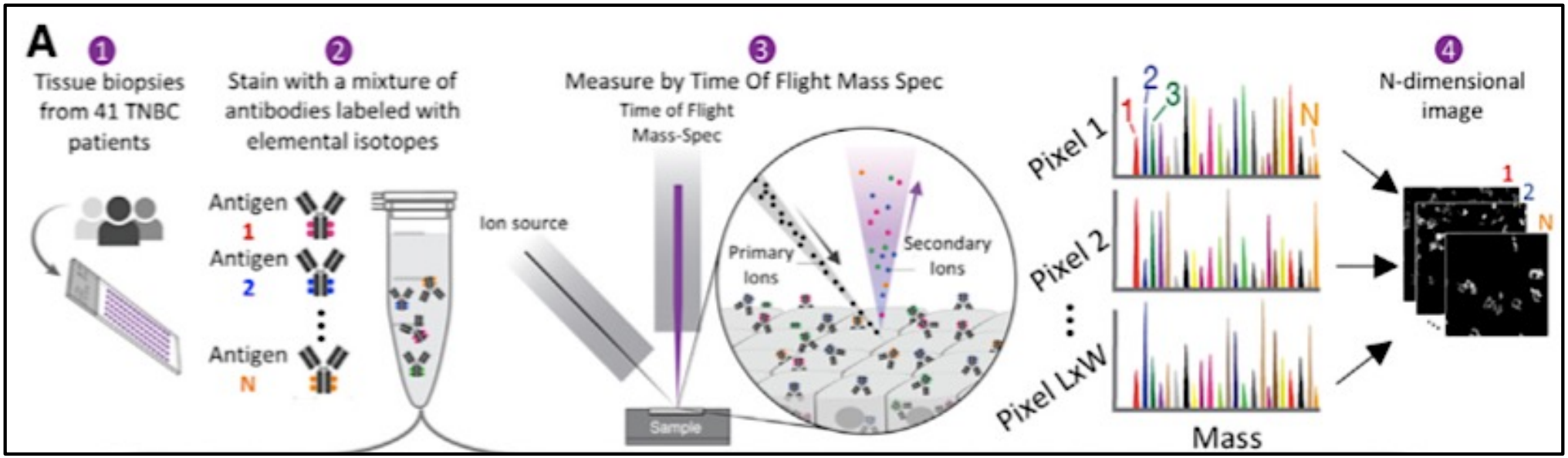
- LMD **survival** is extremely poor
  - Lung: 3 - 6 mo.
  - Breast: 3.5 – 4.4 mo.
  - Melanoma: 1.7 – 2.5 mo.
- To date, there have been 6 randomized clinical trails specifically on LMD treatment

High Level Project Overview

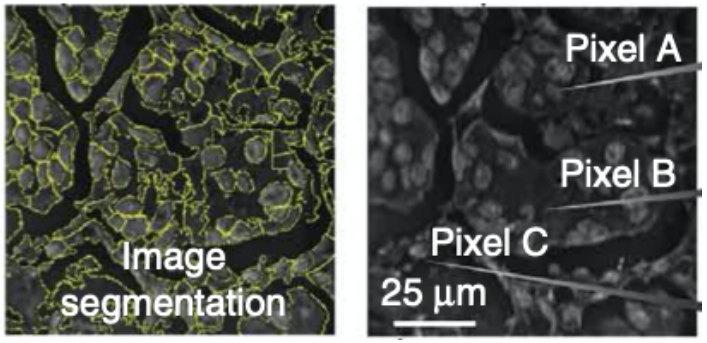


Hypothesis: The **spatial architecture** of the tumor microenvironment reflects distinct **tumor-immune interactions**; these interactions **prime** systemic immune **tolerance** of disseminated tumor cells, enabling brain-specific **metastases**.

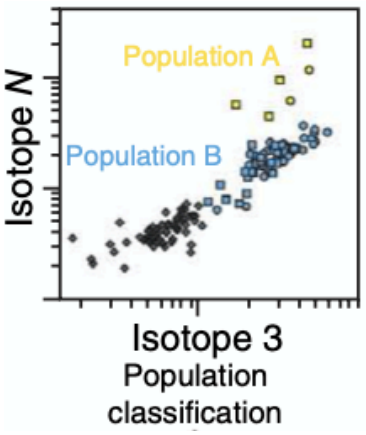
Multiplexed Ion Beam Imaging by Time-of-Flight Mass Spectrometry (MIBI-TOF)

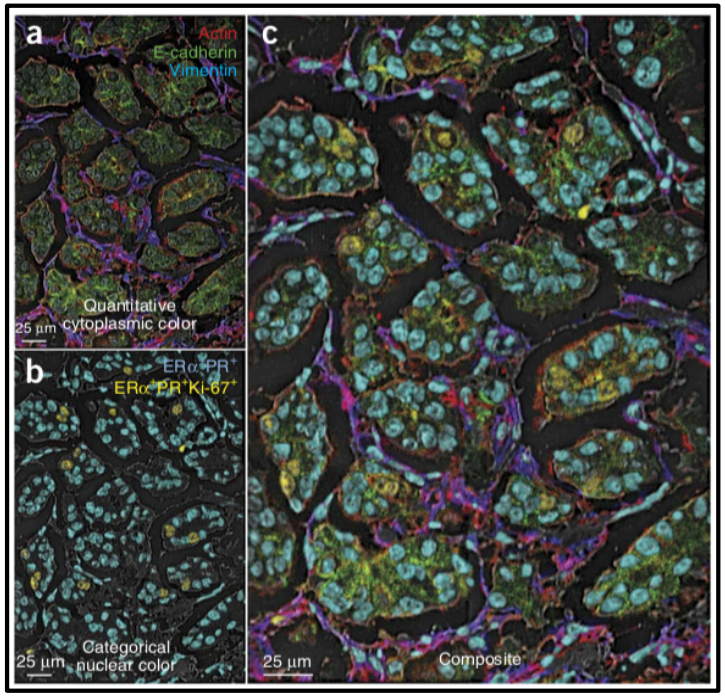


Tabulate and analyze resultant data using image segmentation with quantitative and categorical classifiers



	Isotope 1	2	3	....N
Cell 1	3,	8,	9	8
Cell 2	1,	8,	6	4
Cell 3	9,	9,	4	7





Angelo et al, 2014, Nature Medicine

Project Rationale

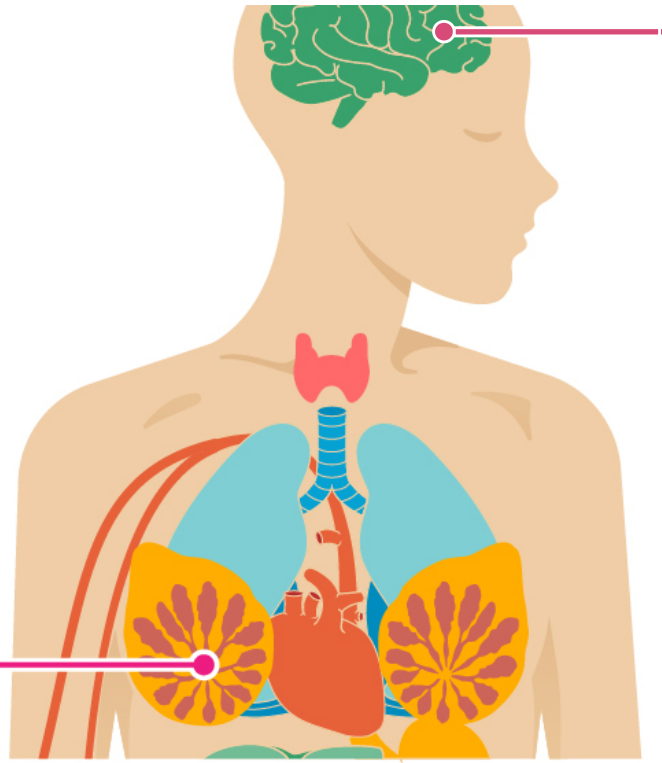
1. Immune infiltration is associated with patient survival in **specifically in TNBC subtype**

2. Angelo Lab – Immune landscape of 41 primary TNBCs using MIBI
    - Spatial organization associated with patient overall survival

3. Presence of immune cells in a mouse model of human TNBC brain metastases

4. Compartmentalized tumors (MIBI data) were less likely to be associated with recurrence than mixed tumors

Project Approach



"brain-focused"

Question: What features of the breast cancer brain metastases TME correlate with disease progression and patient outcomes?

Goal: Analyze TME (spatial and composition) of all breast cancer brain mets, correlate to patient clinical features (DFS, OS)

"breast-focused"

Question: What features of TNBC breast tumor prime immune tolerance of brain metastases?

Goal: Analyze TME (spatial and composition) of TNBC primary tumors in patients that develop brain mets vs. those who do not.

Build TWO distinct tissue microarrays (TMAs)

Next Steps & Future Directions

Next Steps

- Request "**breast-focused**" FFPE blocks from Pathology (Dr. West)
  - Slide annotation
- Identify normal brain controls (epileptic patients)
- Begin construction of "**breast-focused**" TMA
- MIBI panel construction and optimization

Tumor-Immune Panel Keren et al, 2018, Cell

Tumor

β-Catenin
EGFR
Keratin 6
Keratin 17
Pan-keratin
p53

Immune Cell Types

Lymphocytes

CD3
CD4
CD8
CD56
FoxP3
CD45RO
CD20
CD56
CD16
CD138

Monocytes

CD11b
CD11c
CD63
CD68

Neutrophils

MPO

Antigen Presentation

HLA1
HLA-DR
CD209

Immune Regulation

Lag3
PD1
PD-L1
IDO

Stroma

CD31
SMA
Vimentin

Cell Status

dsDNA
H3K27me3
Ki-67
H3K9ac
pS6

Next Steps & Future Directions

Next Steps

- Request "**breast-focused**" FFPE blocks from Pathology (Dr. West)
  - Slide annotation
- Identify normal brain controls (epileptic patients)
- Begin construction of "**breast-focused**" TMA
- MIBI panel construction and optimization

Next Steps & Future Directions

Next Steps

- Request "**breast-focused**" FFPE blocks from Pathology (Dr. West)
  - Slide annotation
- Identify normal brain controls (epileptic patients)
- Begin construction of "**breast-focused**" TMA
- MIBI panel construction and optimization
- Preliminary IHCs

But what about African-American women?

- Dr. Victoria Seewaldt – City of Hope
  - Early molecular changes predict aggressive biology
  - TNBC in African-American women
- How do spatial and temporal changes in the TME correlate with disease onset/progression?

Acknowledgments

- Mentors: Drs. Melanie Hayden Gephart & Sylvia K. Plevritis
- Cohort Building: Bryanna Godfrey & Monica Granucci
- TMA Team: Drs. Rob West, Hannes Vogel, Saman Ahmadian
- Gephart Lab Members
- Plevritis Lab Members
- Funding: NIH SCIT T32

Thank you for your attention!
Questions?