Preclinical and Clinical Imaging of the Immune System

Anna M. Wu, Ph.D.
Professor and Chair, Department of Immunology and Theranostics, DMRI
Co-Director, Center for Theranostics
Beckman Research Institute, City of Hope Medical Center, Duarte CA, USA

Biomarkers of Response for Radiation and Immuno-Oncology
AAPM Annual Meeting July 12, 2022

Disclosures

Founder, Board Member, and Consultant to ImaginAb, Inc.
Honorarium, Roche
Consultant, AstraZeneca
Consultant, Novartis Institute for Biomedical Research
Unlocking the immune system for cancer therapy

Adapted from Mellman, Coukos and Dranoff, Nature 480, 2011

LYMPH NODE

TUMOR

Metastatic Melanoma Response to ipilimumab (anti-CTLA-4)
Before Ipilimumab
04/22/11
After Ipilimumab
08/05/11

Courtesy of Antoni Ribas, UCLA

Conventional Imaging and Cancer Immunotherapy: Challenges

Anatomical imaging: CT or MRI

Tumor progression and/or immune responses can look the same using conventional imaging – Conventional imaging is non-specific

Pseudoprogression in melanoma patient treated with pembrolizumab (anti-PD-1)

FDG flare in melanoma patient treated with ipilimumab (anti-CTLA-4)
Guldbrandson et al. Diagnostics 2017
Beyond conventional imaging: Non-invasive molecular imaging of immune responses

1. Imaging **altered cellular metabolism** (e.g., $[^{18}\text{F}]$-fluorodeoxyglucose, FDG; $[^{18}\text{F}]$-fluoro-thymidine FLT; $[^{18}\text{F}]$-Clofarabine, $[^{18}\text{F}]$-AraG and analogs)

2. **Ex vivo** labeling of cells, followed by re-infusion ($[^{111}\text{In}]$-oxine; $[^{89}\text{Zr}]$-oxine; magnetic nanoparticles, $[^{18}\text{F}]$ nanoparticles)

3. Engineering cells with **reporter genes/tags** (optical, SPECT, PET; e.g., SSTR2, EGFRt, HSV-tk)

4. Imaging **activation markers** (Granzyme B, IFN-γ)

5. Direct imaging of **cell surface biomarkers** using antibodies, nanobodies, etc. Includes lineage as well as functional markers of activation/exhaustion

---

Imaging immune cells *in vivo*: Examples

1. Imaging nucleoside metabolism

   - $[^{18}\text{F}]$F-AraG is selectively taken up by activated CD8 T cells by phosphorylation by mitochondrial dGK
   - MC38 tumors treated with α-PD-1
   - PET imaging using $[^{18}\text{F}]$F-AraG shows increased uptake in tumor and draining lymph nodes in responders as early as 48 h
   - **Currently in clinical evaluation**

2. **Ex vivo** cell labeling

   - $[^{89}\text{Zr}]$-oxine labeling optimized for DC, naïve and activated CTL, OT-1
   - Viability and functionality assessed
   - DCs track to spleen and liver (upper panels)
   - Naïve CTL home to spleen and LN (lower panels)
   - **First-in-human $[^{89}\text{Zr}]$ PET imaging of WBC (Lapi et al.)**

---


Sato et al. Radiology 2015
3. Reporter gene imaging

- IL-13 zetakine cytotoxic T cells infused intratumorally in patients with GBM
- HSV1-tk reporter gene detected using $[^{18}\text{F}]$FHBG
- 7 patients imaged (6 pre- and post-treatment)

4. Immune cell activation

- Release of Granzyme B is a major mechanism of cellular cytotoxicity
- Tetrapeptide substrate analog of Granzyme B
- Treated with α-PD-1/α-CTLA-4
- Responding tumor show high GZP signal, indicating presence and activity of cytotoxic T cells
- Currently in clinical evaluation

5. Imaging cell surface biomarkers: ImmunoPET

- Antibodies: Specificity
- PET: Sensitivity, resolution, quantitation

**ImmunoPET research is accelerating due to advances in:**
- Availability of approved therapeutic antibodies; manufacturing infrastructure
- Positron-emitting radionuclides with longer physical half-lives (Cu-64, Zr-89, I-124) to match antibody circulating half-lives (days)
- Antibody engineering to optimize for clinical imaging – accelerated clearance
5. Imaging Checkpoint Biomarkers

PD-1 tracer: [\(^{89}\)Zr]-nivolumab

- 13 patients with NSCLC
- Heterogenous uptake of both tracers
- Correlated with IHC
- Some accumulation in brain metastases

Niemeijer et al. Nat Comm 2018

5. Imaging CD8 cytotoxic T Cells:
An early marker of response to immunotherapy

- Many cancer immunotherapies converge on the CD8 cytotoxic T lymphocyte as the key effector cell
- Several on-treatment biopsy studies have shown that infiltration of CD8+ cells soon after initiation of checkpoint inhibitor therapy correlates with outcome

PD-1 blockade; Tumeh, Ribas et al. Nature 2014
Preclinical applications – CD8 immunoPET

Murine-specific

$^{89}$Zr-DFO-169 anti-mouse CD8
cys-diabody
- Pan-specific (all strains)
- Rapid renal clearance

Human-specific

$^{89}$Zr-crefmirlimab berdoxam (IAB22M2C)
Fully humanized anti-human CD8 minibody
- High affinity (0.4 nM)
- Non-immunogenic, biologically inert
- Rapid hepatic clearance

Cancer immunotherapy:
- Checkpoint inhibitor therapy and combinations
- Adoptive cell therapy
- Oncolytic viruses
- Graft vs host disease
- Infectious disease (COVID-19)

Wild-type mouse

Humanized anti-human
CD8 minibody
- High affinity (0.4 nM)
- Non-immunogenic
- Biologically inert
- Rapid clearing

Phase I Clinical imaging of CD8 T lymphocytes using $^{89}$Zr-IAB22M2C (crefmirlimab berdoxam)

Objectives:
- Safety, tolerability & whole body distribution (including tumor sites)
- Determine recommended protein dose & scanning parameters for future studies

Design:
- Open-label, non-randomized, 2 stage:
  - Protein dose escalation (6 patients: 3 mCi $^{89}$Zr; 0.2-10 mg protein)
  - Expansion (9 patients: 1.0 – 1.5 mg)
- Serial imaging at 1-2 h; 6-8 h; 24 h; 48 h; 96-144 h
- Serial blood draws for pharmacokinetics, cytokines, anti-drug antibody

Tavaré, R. et al., J. Nucl. Med. 2015
T. Olafsen et al. abstract AACR 2016

Phase I $^{89}$Zr-crefmirlimab PET - CD8 T cells Visualized in Tumor

- 37 year old female with metastatic melanoma
- Ipi/Nivo (2 yr); Pembro
- 3 mCi/0.2 mg

Day 1: 6 h
FDG PET/CT

SUV = 0.7

Day 1: 6h
CD8 immunoPET/CT

6 days

1 day

Phase I Summary

Imaging conclusions:
- Rapid clearance; excretion primarily hepatobiliary
- Uptake in T-cell rich tissues (spleen, BM, LN)
- No/low uptake in normal organs (muscle, heart, brain, lungs)
- Tumor uptake variable and seen in 2/3 of patients
- Protein dose range with favorable biodistribution: 0.5-1.5 mg
- Most favorable imaging time: 24 hrs, although tumors seen as early as 1-2 hrs

Phase II Pre-treatment/On-treatment study (NCT03802123, NCT05013099)

- Patients with metastatic solid tumors, initiating checkpoint inhibitor therapy
  - Pre-treatment (baseline) CD8 PET scan and biopsy (3 mCi or 1 mCi/1.5 mg; 24 h)
  - Initiate immunotherapy (ipi/nivo/pembro standard of care)
  - On-treatment CD8 PET scan and biopsy (4-5 weeks after therapy initiation)
- Goals
  - Safety of repeat dosing and imaging
  - Correlation of CD8 PET with CD8 IHC
  - Correlation with RECIST and outcome
- Multi-center Phase IIa trial complete; Phase IIb iPREDICT in progress

Future: Non-invasive Imaging in Immuno-oncology

- Molecular imaging of immune responses: powerful, specific, whole-body approaches for monitoring immune responses
- Current approaches include metabolic imaging, ex vivo immune cell labeling, reporter gene imaging, and activation marker detection
- ImmunoPET for detection of cell surface biomarkers (PD-1/PD-L1 checkpoints, CD8 cytotoxic T cells)
- Many of these approaches show promise in clinical trials
- Potential role in cancer immunotherapy
  - Patient selection
  - Early on-treatment response, ongoing response
  - Optimization of combination therapy
  - Management of toxicities
- Potential role in other immune-mediated conditions and diseases
Acknowledgments

Research

Past Wu lab at UCLA:
- Deirdre LaPlaca
- Jennifer Chean
- Felix Salazar
- Bao Ying Chen

UCLA collaborators:
- Antoni Ribas
- Owen Witte
- Arion Chatziioannou

Wu Lab:
- Deirdre LaPlaca
- Jennifer Chean
- Felix Salazar
- Bao Ying Chen

Past Wu lab at UCLA:
- Wenting Tsai
- Richard Tavaré
- Scott Knowles
- Amanda Freise

UCLA collaborators:
- Antoni Ribas
- Owen Witte
- Arion Chatziioannou

Clinical

Phase I sites:
- MSKCC:
  - Neeta Pandit-Taskar
  - Wolfgang Weber
  - Jaxt Wistock
  - Michael Postow
  - Jason Lewis
  - Sergei Lyashchenko
  - Joseph O’Donoghue

- MSKCC:
  - Neeta Pandit-Taskar
  - Wolfgang Weber
  - Jaxt Wistock
  - Michael Postow
  - Jason Lewis
  - Sergei Lyashchenko
  - Joseph O’Donoghue

- UPenn:
  - Mike Farwell
  - David Markoff

- Honor Health
  - Mike Gordon
  - Ron Korn

Phase II sites:
...and our patients!

Industry

ImaginAb
- Ian Wilson
- William Le
- Alessandro Mascioni
- Jean Gudas
- Michael Torgov
- Tobe Olafsen

Research

Clinical

Industry

17