PET Imaging of Cancer Biomarkers

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Disclosure

No relevant financial relationships with commercial interests.

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Evolution of Radiology

Medicine

late 19th century

Radiology

early 20th century, Columbia Univ

Molecular Medicine

today
Radiosynthesis of $^{14}$C compounds ($T_{1/2} = 20.4$ min)

- 1975: $^{13}$C-glucose was prepared by photosynthesis using Swiss chard leaves. Mashed up, extracted and a "green solution" injected into humans.
- 2012: $^{13}$C-glucose is prepared by a "black box" automated versatile synthesizer producing drugs ready for human use.

Targeted Imaging – Frontiers of Diagnosis

- $^{99m}$Tc – Bone Scan
- $^{18}$F-FDG PET/CT Glycolysis
- $^{18}$F-FDHT PET/CT Androgen Receptor

Prostate Cancer: Revealing Heterogeneous Biology of Tumor Metastasis

- $^{18}$F-FDG PET/CT
- $^{18}$F-FDHT PET/CT

S. Larson, J. Fox, M. Morris et al.,
18F-FDHT PET/CT: Predictive and Targeted Response Biomarker

- Baseline
- 4 wks after MDV3100

18F-FES PET/CT: Predictive and Targeted Response Biomarker ER+ breast cancer patients?

- Imaging of Androgen Receptors
- N N N CH3
- N
- SHN NH
- CH3 CH3
- H3C 64Cu
- 64Cu-ATSM
- Imaging of Altered Amino Acid Metabolism

- 15O-Water
- Imaging of Blood Flow

Confirmed full target occupancy ~20 hours post dose

Courtesy of Drs. Dickler, Ulaner et al.
**18F-FDHT PET/CT: Predictive and Targeted Response Biomarker**

![Image of PET/CT scans showing baseline and 4 weeks after MDV3100 administration.](image)

**Rationale for Developing a Biomarker of AR Pathway Activity**

Changes in FDHT binding post MDV3100 are not uniformly associated with clinical response or other response indicators.

FDHT is a radioligand for AR that measures receptor occupancy by PET.

Each bar represents % change in FDHT uptake post MDV3100 for one pt.

Scher, Morris, Fox, Larson et al.

**Intra/Inter Tumoral Heterogeneity - Challenge to Precision Medicine: Can we/should we biopsy each and every lesion?**

![Image showing intra/inter tumoral heterogeneity.](image)
Radiopharmaceuticals

- Agents
  - Small molecules
  - Peptides
  - Monoclonal antibodies

### Radiopharmaceutical Imaging Target

<table>
<thead>
<tr>
<th>Radiopharmaceutical</th>
<th>Imaging Target</th>
<th>Cancer Site</th>
<th>Human studies</th>
</tr>
</thead>
<tbody>
<tr>
<td>[18F]-FLT</td>
<td>Tumor cell proliferation</td>
<td>Lymphoma, prostate, H&amp;N, NSCLC</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[18F]-FES</td>
<td>Estrogen receptor status</td>
<td>Breast</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[18F]-FDHT</td>
<td>Androgen receptor</td>
<td>Prostate</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[18F]-FMISO</td>
<td>Tumor oxygenation</td>
<td>Head &amp; Neck, Rectal</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[18F]-FACBC</td>
<td>Amino acid metabolism</td>
<td>Breast, Prostate, Brain</td>
<td>RDRC/GEMS IND</td>
</tr>
<tr>
<td>[18F]-FIAU</td>
<td>Gene expression</td>
<td>Prostate</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[18F]-ML10</td>
<td>Imaging apoptosis</td>
<td>Brain, NSCLC, H&amp;N, Non</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[18F]-dasatinib</td>
<td>Tyrosine kinases</td>
<td>Prostate, Breast</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[18F]-glutamine</td>
<td>Tumor metabolism</td>
<td>All solid malignancies</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[64Cu]-ATSM</td>
<td>Tumor oxygenation</td>
<td>Uterine Cervix, Rectal</td>
<td>ACRIN</td>
</tr>
<tr>
<td>[124I]-IAZGP</td>
<td>Tumor oxygenation</td>
<td>Rectal</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[124I]-FIAU</td>
<td>Gene expression</td>
<td>Prostate</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[124I]-Na</td>
<td>Na Iodide Symporter</td>
<td>Thyroid</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[124I]-PUH71</td>
<td>HSP-90</td>
<td>All solid malignancies and lymphoma</td>
<td>MSKCC IND</td>
</tr>
</tbody>
</table>

### Antibodies and Fragments (Imaging)

<table>
<thead>
<tr>
<th>Radiopharmaceutical</th>
<th>Imaging Target</th>
<th>Cancer Site</th>
<th>Human studies</th>
</tr>
</thead>
<tbody>
<tr>
<td>[68Ga]-Her2 F(ab’2)</td>
<td>HER2</td>
<td>Breast</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[64Cu]-DOTA-trastuzumab</td>
<td>HER2</td>
<td>Breast</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[124I]-A33</td>
<td>A33 antigen</td>
<td>Colon</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[124I]-3F8</td>
<td>GD2 antigen</td>
<td>Neuroblastoma (pediatrics)</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[124I]-8H9</td>
<td>8H9 antigen</td>
<td>Multiple tumors e.g. Leptomeninges (pediatrics)</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[124I]-G250</td>
<td>CA9 antigen</td>
<td>Renal</td>
<td>MSKCC IND</td>
</tr>
</tbody>
</table>

### Antibodies and Fragments (Therapy)

<table>
<thead>
<tr>
<th>Radiopharmaceutical</th>
<th>Therapy Target</th>
<th>Cancer Site</th>
<th>Human studies</th>
</tr>
</thead>
<tbody>
<tr>
<td>90Y-DOTA-cG250</td>
<td>CA9 antigen</td>
<td>Renal</td>
<td>LICR IND</td>
</tr>
<tr>
<td>[131I]-8H9</td>
<td>8H9 antigen</td>
<td>Multiple tumors e.g. Leptomeninges (pediatrics)</td>
<td>MSKCC IND</td>
</tr>
<tr>
<td>[131I]-3F8</td>
<td>GD2 antigen</td>
<td>Neuroblastoma (pediatrics)</td>
<td>MSKCC IND</td>
</tr>
</tbody>
</table>

### Nanoparticles (Imaging)

<table>
<thead>
<tr>
<th>Radiopharmaceutical</th>
<th>Imaging Target</th>
<th>Cancer Site</th>
<th>Human studies</th>
</tr>
</thead>
<tbody>
<tr>
<td>[124I]-Cdot nanoparticles</td>
<td>αvβ3</td>
<td>Melanoma</td>
<td>MSKCC IND</td>
</tr>
</tbody>
</table>

### Prostate Specific Membrane Antigen

- Folate hydrolase (FOLH1), 100 kDa
- Type 2 transmembrane glycoprotein, 720 amino acids
- Present in salivary glands and small intestines
- Prostate cancer and non-prostatic solid tumor neovascuature (i.e. bladder, pancreas, lung, kidney)
- FDA-approved [111In]-7E11 (intracellular epitope, low sensitivity for viable tumor sites)
Evaluating J591 as a Biomarker of Response to AR Pathway Directed Inhibitors in PCa

Pharmacologically Triggered Elevations in PSMA Expression can be Measured by PET with J591

The white arrows indicates the positions of the LNCaP-AR tumors

89Zr-DFO-J591 for ImmunoPET

PET imaging using a PSMA -ve control

ImmunoPET images recorded in a male athymic, nude mouse with sub-cutaneous PC-3 tumors (20 – 30 mm)

ImmunoPET images recorded in a male athymic, nu/nu mouse with sub-cutaneous LNCaP tumors (50–250 mm³).


PET Imaging of Prostate Cancer

FDG - 1d  [F-18] NaF - 4d  Zr-89 DFO-J591

Zr-89 J591 @ 7 days pi
Castrate Resistant Prostate Cancer

Coronal WB  Transaxial WB  Transaxial CT  Transaxial Fused
Imaging Androgen Receptor Signaling with a Radiotracer Targeting Free Prostate Specific Antigen

David Ulmert, Michael J. Evans, Jason P. Holland, Samuel L. Rice, John Wongvipat, Kim Pettersson, Per-Anders Abrahamsson, Peter T. Scardino, Steven M. Larson, Hans Lilja, Jason S. Lewis, and Charles L. Sawyers

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Evaluating 5A10 as a Biomarker of Response to AR Pathway Directed Inhibitors in PCA

Rationale for the Development of $^{89}$Zr-5A10

Our central hypothesis is that targeting a form of PSA more closely related to AR activity may enhance the diagnostic value of PSA expression changes.


Evaluating 5A10 as a Biomarker of Response to AR Pathway Directed Inhibitors in PCa

- Intact male mice bearing the indicated tumor were treated with $^{89}$Zr-5A10
- Tumors were analyzed ex vivo 24 h post injection of radiotracer

\*P<0.01; **P<0.05


$^{89}$Zr-5A10 Quantitatively Measures AR Inhibition by an Antianдрrogen in PCa xenografts

- Castrate male mice with LNCaP-AR tumors were treated for 7 d with MDV3100
- Tumors were harvested 24 h post injection of $^{89}$Zr-5A10

\*P<0.05 compared to vehicle

hK2 is an AR-Governed Kallikrein that is Abundantly Expressed in Healthy and Malignantly Transformed Prostatic Tissues

Can $^{99}$Zr-11B6 localize and demarcate normal and cancerous prostatic tissue in GEM models?

David Ulmert, Daniel Thorek et al.,

Cy5.5-11B6 Fluorescence Guided Surgery

David Ulmert, Daniel Thorek et al.,
Applying PET to Broaden the Diagnostic Utility of the Clinically Validated CA19.9 Serum Biomarker for Oncology

Nerissa Therese Viola-Villegas, Samuel L. Rice, Sean Carlin, Xiaohong Wu, Michael J. Evans, Kuntal Sevak, Marija Drobjuk, Govind Ragupathi, Ritsuko Sawada, Wolfgang W. Scholz, Philip D. Livingston, Jason S. Lewis

Memorial Sloan-Kettering Cancer Center
Mabvax Therapeutics, San Diego, California

Villegas et al., JNucl Med 2013 54:1876-1882
What is CA19.9?

- Supports selectin-dependent adhesion
- Carbohydrate antigen 19.9 (aka: sialyl Lewis-a)
  - Up to 200 copies/cell
  - Attached to as many as 50 proteins
- Elevated in several types of cancer, including PDAC (~90%)

\[ \text{SLe}^a \text{ or CA19.9} \]
- Serum tumor marker
- Present in pancreas, lung and colorectal lesions
- Aids in metastasis through cell adhesion

\[ \text{Zr-89 5B1} \]
- Immunoreactivity: ~ 84 %
- Specific activity: 12 mCi/mg
- RCY: > 80 %; RCP: > 99 %

Tumor models:
1. DMS79 small lung cancer
2. Colo205-luc colorectal cancer
3. BxPC3 pancreas cancer

Villegas et al., J Nucl Med 2013 54:1876-1882

\[ {^{18}}\text{F-FDG vs. Zr-5B1 PET} \]

\[ \text{Zr-5B1 shows better specificity and tumor localization compared to} \ {^{18}}\text{F-FDG}. \]

Villegas et al., J Nucl Med 2013 54:1876-1882
**Ex vivo CA19.9 Serum Values**

<table>
<thead>
<tr>
<th>Tumor type</th>
<th>Animal #</th>
<th>Tumor volume, mm³</th>
<th>CA19.9, U/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Colo205-luc</td>
<td>M1</td>
<td>200.5</td>
<td>322.7</td>
</tr>
<tr>
<td>Colorectal Cancer</td>
<td>M2</td>
<td>207.3</td>
<td>201.9</td>
</tr>
<tr>
<td></td>
<td>M3</td>
<td>201.3</td>
<td>1318.9</td>
</tr>
<tr>
<td>BaP3</td>
<td>M1</td>
<td>222.3</td>
<td>N.D.*</td>
</tr>
<tr>
<td>Pancreatic Cancer</td>
<td>M2</td>
<td>220.0</td>
<td>N.D.</td>
</tr>
<tr>
<td></td>
<td>M3</td>
<td>230.0</td>
<td>N.D.</td>
</tr>
<tr>
<td>DMS79</td>
<td>M1</td>
<td>288.0</td>
<td>N.D.</td>
</tr>
<tr>
<td>Small Cell Lung</td>
<td>M2</td>
<td>245.0</td>
<td>N.D.</td>
</tr>
<tr>
<td></td>
<td>M3</td>
<td>232.4</td>
<td>N.D.</td>
</tr>
<tr>
<td>Control</td>
<td>M1</td>
<td>-</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>M3</td>
<td>-</td>
<td>3</td>
</tr>
</tbody>
</table>

* Not detected.

$^{89}$Zr-$^5$Bm can detect CA19.9 at the tumor site even when the antigen is undetectable in serum.

Villegas et al., J Nucl Med 2013 54:1876-1882

**Issues with PDAC xenograft models**

- Does not accurately mimic tumor microenvironment
  - PDAC is highly stromal and has low vascular density
    - One of the reasons for poor drug (PET agent?) delivery
  - BxPC3 may not be from primary pancreas tumor
    - Lack of K-ras mutation = almost certainly not
  - Not located in or near pancreas
    - Liver uptake cause for concern with mAB PET agents

**New cell line and methods for CA19.9 imaging**

- CA19.9 is a human antigen not made naturally by mice
  - Mice lack the enzyme necessary for CA19.9 production
- Collaborating with Dr. David Tuveson (CSHL)
  - Development of mouse model expressing CA19.9
  - First step: develop mouse cell line with CA19.9
    - Dr. Dannielle Engle (Tuveson Lab)
    - Based on KPC mouse
  - Second step: imaging with novel cell line as proof of concept
    - 2.1: xenograft
    - 2.2: orthotopic pancreas model
Imaging PDAC (mouse CA19.9 cell line)

Orthotopic Pancreas Model (14 days)

PET/CT of the same mouse

Site-Specific Labeling of Antibodies
Multi-Modality Construction?

1. Clouse up sugars
2. Attach GeNA
3. Labletag

\( ^{119}\text{Zr-DFO-AF680-A33} \)

Imaging PDAC – bilateral SubQ nude

Site-click \(^{89}\text{Zr-DUAL-5Bs} \) (DFO + IRDye800CW)

BxPC3 – Right Flank
MiaPaCa2 – Left Flank
How Hard are We Hitting the Target?

- **Non-targeted scan**: Increase dose or alternate therapy
- **% decline**: No, then alter therapy.

**"F-FDHT PET** | **"Zr-J591 or "Zr-5A10 PET**

Current Lewis Lab

- Danya Askie Ali
- Dr. Nicole R. Carlin
- Kathleen Carstairs
- Dr. D. Huy Chai
- Dr. Donel Dencin
- Melissa Deri
- Dr. Jacob Houghton
- Michael Doran
- Dr. Alex Poot
- Yvette Mingo
- Brandon Narayanan
- Shashikanth Ponnala
- Dr. Dustin Demoin
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- Dr. Mirkka Sarparanta
- Dr. Hanwen Zhang
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- Many fabulous visiting scientists....

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    - Res CA119693
    - Res CA091256
    - Res CA175166
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A bottle of wine contains more philosophy than all the books in the world
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