Positron Emission Tomography: Tool to Study Pharmacokinetics and to Facilitate Drug Development

Robert B. Innis, MD, PhD
Molecular Imaging Branch
National Institute Mental Health
Outline of Talk

1. PET has high sensitivity and specificity
2. PET used in therapeutic drug development
3. Pharmacokinetic modeling of plasma concentration and tissue uptake can measure receptor density
4. Study drug distribution: block distribution to periphery and increase distribution to brain
5. Study drug metabolism: inhibit defluorination
Imaging Receptors with PET
Positron Emission Tomography
### PET vs. MRI

<table>
<thead>
<tr>
<th></th>
<th>PET</th>
<th>MRI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Spatial Resolution</td>
<td>2 – 6 mm</td>
<td>&lt;&lt; 1 mm</td>
</tr>
<tr>
<td>Sensitivity</td>
<td>$10^{-12}$ M</td>
<td>$10^{-4}$ M</td>
</tr>
<tr>
<td>Temporal Resolution</td>
<td>minutes</td>
<td>&lt;1 sec</td>
</tr>
</tbody>
</table>

Radionuclide ($^{11}$C): high sensitivity  
Ligand (raclopride): high selectivity  
Radioligand [$^{11}$C]raclopride: high sensitivity & selectivity
Radioligand = Drug + Radioactivity

1. Drug administered at tracer doses
   a) No pharm effects
   b) Labels <1% receptors
   c) Labeled subset reflects entire population

2. Radioligand disposed like all drugs
   a) Metabolism & distribution

3. Radiation exposure
NIH Rodent PET Camera

$^{18}$F bone uptake rat

Developed By: Mike Green & Jurgen Seidel
PET: Tool in Therapeutic Drug Development

- Determine dose and dosing interval
- Identify homogeneous group
- Biomarker for drug efficacy
- Monitor gene or stem cell therapy
Lazabemide blocks $[^{11}\text{C}]$deprenyl binding to monoamine-oxidase-B (MAO-B)

Selegilene is more potent and longer acting than lazabemide
PET: Tool in Therapeutic Drug Development

- Determine dose and dosing interval
- Identify homogeneous group
- Biomarker for drug efficacy
- Monitor gene or stem cell therapy
Dopamine Transporter: Located on DA Terminals
Removes DA from Synapse
SPECT Imaging of Dopamine Transporter in Caudate and Putamen of Human Brain
$^{123}$I-β-CIT Dopamine Transporter SPECT: Decreased in Parkinson’s Disease

Healthy

Parkinson Stage 1
PET: Tool in Therapeutic Drug Development

- Determine dose and dosing interval
- Identify homogeneous group
- Biomarker for drug efficacy
- Monitor gene or stem cell therapy
Serial Dopamine Transporter Imaging in a Parkinson Patient

Institute for Neurodegenerative Disorders
PET Imaging of Amyloid: Biomarker for Alzheimer’s Disease
PET: Tool in Therapeutic Drug Development

- Determine dose and dosing interval
- Identify homogeneous group
- Biomarker for drug efficacy
- Monitor gene or stem cell therapy
Gene Therapy Using Viral Vectors

Viral vectors deliver gene that synthesizes dopamine (DA)
Infuse virus into striatum (target cells)

Target cells express the DA gene
PET Dopamine Imaging in Hemi-Parkinson Monkey:
Monitors gene for DA synthesis in right striatum

pre | post
--- | ---
Control Gene: Lac-Z | DA Synthesis Gene: AADC
**PET Imaging to Monitor Embryonic Stem Cell Treatment of “Parkinson Disease” in Rats**

<table>
<thead>
<tr>
<th>Normal</th>
<th>Unilateral Lesion</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="image1" alt="Embryonic Stem Cells" /></td>
<td><img src="image2" alt="PET &amp; MRI" /></td>
</tr>
</tbody>
</table>

The images above illustrate the differences in PET and MRI scans before and after treatment with Embryonic Stem Cells to monitor the progression of Parkinson Disease in rats.
Outline of Talk

1. PET has high sensitivity and specificity
2. PET used in therapeutic drug development
3. Pharmacokinetic modeling: plasma concentration and tissue uptake
4. Study drug distribution: “peripheral” benzodiazepine receptor
5. Study drug metabolism: inhibit defluorination
Brain Uptake of $[^{18}F]$Fluoxetine: Measures Density of Serotonin Transporters & Affinity of Fluoxetine

Patient

Healthy

Brain Drug

AUC=32

AUC=16

Time

Time
### Brain Uptake of $[^{18}\text{F}]$Fluoxetine:
- Measures Density of Serotonin Transporters & Affinity of Fluoxetine

<table>
<thead>
<tr>
<th></th>
<th>Patient</th>
<th>Healthy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inject Activity</td>
<td>20 mCi</td>
<td>10 mCi</td>
</tr>
</tbody>
</table>

AUC = 32

AUC = 16
Brain Uptake of $[^{18}\text{F}]$ Fluoxetine: Measures Density of Serotonin Transporters & Affinity of Fluoxetine

<table>
<thead>
<tr>
<th></th>
<th>Patient</th>
<th>Healthy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inject Activity</td>
<td>20 mCi</td>
<td>20 mCi</td>
</tr>
<tr>
<td>AUC</td>
<td>AUC=32</td>
<td>AUC=16</td>
</tr>
</tbody>
</table>
Brain Uptake of $[^{18}\text{F}]$Fluoxetine: Measures Density of Serotonin Transporters & Affinity of Fluoxetine

<table>
<thead>
<tr>
<th></th>
<th>Patient</th>
<th>Healthy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inject Activity</td>
<td>20 mCi</td>
<td>20 mCi</td>
</tr>
<tr>
<td>Weight</td>
<td>50 kg</td>
<td>100 kg</td>
</tr>
</tbody>
</table>
Brain Uptake of \(^{18}F\)Fluoxetine: Measures Density of Serotonin Transporters & Affinity of Fluoxetine

<table>
<thead>
<tr>
<th></th>
<th>Patient</th>
<th>Healthy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inject Activity</td>
<td>20 mCi</td>
<td>20 mCi</td>
</tr>
<tr>
<td>Weight</td>
<td>100 kg</td>
<td>100 kg</td>
</tr>
</tbody>
</table>
Brain Uptake of $[^{18}\text{F}]$Fluoxetine: Measures Density of Serotonin Transporters

<table>
<thead>
<tr>
<th>Brain Drug</th>
<th>Patient</th>
<th>Healthy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Time</td>
<td>AUC=32</td>
<td>AUC=16</td>
</tr>
<tr>
<td>Inject Activity</td>
<td>20 mCi</td>
<td>20 mCi</td>
</tr>
<tr>
<td>Weight</td>
<td>100 kg</td>
<td>100 kg</td>
</tr>
<tr>
<td>Liver disease</td>
<td>Yes</td>
<td>No</td>
</tr>
</tbody>
</table>

Patient: Yes
Healthy: No
Binding Potential (BP): Receptor Density * Affinity

BP equals uptake in brain relative to how much drug is delivered via arterial plasma.

\[
BP = \frac{\text{Area Brain Curve}}{\text{Area Plasma Curve}}
\]

\[
BP = \frac{16}{2} = 8
\]
**Binding Potential: Independent of Injected Dose**

Double Plasma Input => Double Brain Response

*If ligand does not saturate receptors - i.e., if tracer doses used

\[
\text{BP 1st Time } = \frac{16}{2} = 8
\]

\[
\text{BP 2nd Time } = \frac{32}{4} = 8
\]
BP can be calculated from the Area Under Curve (math integral) as well as rate constants (math differential).

From curves of plasma and brain radioactivity over time, estimate rate constants of entry and removal to/from tissue.

\[
BP = \frac{K_1}{K_2}
\]
Tissue uptake is proportional to density of receptors and the affinity of the drug

**Binding Potential**

\[ BP = \frac{B_{\text{max}}}{K_D} = B_{\text{max}} \times \frac{1}{K_D} = B_{\text{max}} \times \text{affinity} \]

- \( B_{\text{max}} \) = receptor density
- \( K_D \) = dissociation binding constant
- \( \frac{1}{K_D} \) = binding affinity drug
SUMMARY PET KINETICS

- Organ uptake is proportional to receptor density and affinity of drug
- Binding Potential (BP) = density X affinity
- “Drug Exposure” to tissue is AUC of: plasma concentration vs. time
- “Response” (uptake) of tissue is AUC of: tissue concentration vs. time

\[
BP = \frac{\text{Response}}{\text{Exposure}} = \frac{AUC_{\text{tissue}}}{AUC_{\text{plasma}}}
\]

- BP also equals ratio of rate constants of entry and removal to/from tissue

\[
BP = \frac{K_1}{k_2}
\]
Major Point of PET Pharmacokinetics
(in words)

• Plasma pharmacokinetics provides a limited view of what’s happening to drug in plasma.

• PET provides a limited view of what’s happening to drug in tissue.

• Concurrent measurement of drug in plasma and of drug in tissue allows quantitation of the target of drug action – i.e., receptor.
Outline of Talk

1. PET has high sensitivity and specificity
2. PET used in therapeutic drug development
3. Pharmacokinetic modeling: plasma concentration and tissue uptake
4. Study drug distribution: “peripheral” benzodiazepine receptor
5. Study drug metabolism: inhibit defluorination
Translocator Protein (18 kDa)
a.k.a. “peripheral benzodiazepine receptor”

1. Mitochondrial protein highly expressed in macrophages and activated microglia
2. Exists in periphery and brain
3. Multiple potential functions: steroid synthesis, nucleotide transport
4. Distinct from typical benzodiazepine $\text{GABA}_A$ receptor in brain
5. Marker for cellular inflammation
Receptor Blockade $[^{11}C]PBR28$ in Monkey Brain: more radioligand in plasma and brain

**BASELINE**

$BP = 130 \text{ mL/cm}^3$

**RECEPTORS BLOCKED**

$BP = 1.7 \text{ mL/cm}^3$

**BRAIN**

**PLASMA**

Conc radioactivity in putamin (%SUV)

Conc $[^{11}C]PBR28$ in plasma (%SUV)
Receptor blockade displaces from lung & kidney. Drives more to brain but doesn’t bind there.
Incidental Stroke

Original MRI (T1)

PET 6 weeks after MRI

Repeat MRI 8 weeks after PET

Repeat MRI (FLAIR, edema)
TSPO identifies epileptogenic focus in 15 of 16 patients.

Hirvonen et al., *JNM*, 2012
Human with low uptake is similar to monkey with receptor blockade

A) regular healthy subject

B) odd healthy subject

C) normal monkey

D) pre-blocked monkey
No Binding to $[^{11}C]PBR28$ in Brain and Periphery

**Normal Binding**
- Lungs
- Kidneys
- Spleen
- Heart

2 min 26 min 103 min

**No Binding**
 (~10% subjects)
TSPO rs6971 polymorphism causes differential affinity for PBR28

- Ala to Thr substitution
- Allelic frequency ~ 30%.
  - Prevalence of homozygotes ~ 9%
- Codominant expression
  - HAB - high affinity binding
  - LAB - low affinity binding
  - MAB - reduced binding (mixed affinity states)

Owen, JCBFM 2012
Brain Uptake of $[^{18}\text{F}]\text{Fluoxetine}$: Measures Density of Serotonin Transporters

<table>
<thead>
<tr>
<th>Patient</th>
<th>Healthy</th>
</tr>
</thead>
<tbody>
<tr>
<td>AUC=32</td>
<td>AUC=16</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Brain Drug</th>
<th>Time</th>
</tr>
</thead>
<tbody>
<tr>
<td>AUC=32</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Inject Activity</th>
<th>Patient</th>
<th>Healthy</th>
</tr>
</thead>
<tbody>
<tr>
<td>20 mCi</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Weight</th>
<th>Patient</th>
<th>Healthy</th>
</tr>
</thead>
<tbody>
<tr>
<td>100 kg</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Liver disease</th>
<th>Patient</th>
<th>Healthy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yes</td>
<td></td>
<td>No</td>
</tr>
</tbody>
</table>
Binding Potential (BP): Receptor Density * Affinity

\[
BP = \frac{\text{Area Brain Curve}}{\text{Area Plasma Curve}}
\]

\[
BP = \frac{16}{2} = 8
\]
Experimental Design: Effect of TSPO genotype on PBR28 binding

• PET study
  – 27 healthy volunteers
  – In vitro binding: Leukocyte displacement assay
  – In vivo binding: $^{11}$C]PBR28 PET imaging

• Post-mortem study
  – 47 healthy controls, 45 schizophrenia patients
  – Specific $^{3}$H]PBR28 binding in prefrontal cortex
  – Comparison with and without genotype correction

Kreisl, JCBFM 2013
PET Study: Both TSPO genotype and leukocyte binding assay determine affinity status

100% agreement between binding assay results and genotype

One-site fit = HAB
Two-site fit = MAB
PET Study: $[^{11}\text{C}]$PBR28 binding is 1.4-fold higher in high affinity binders than mixed affinity binders

Expect less than 2-fold difference because $[^{11}\text{C}]$PBR28 uptake represents specific and nonspecific binding

Mean HAB = 4.5 mL • cm$^{-3}$
Mean MAB = 3.2 mL • cm$^{-3}$
PET Study: Greater brain uptake in HH subjects with similar plasma concentration as HL subjects
Post-mortem study: High and mixed affinity binders also seen in schizophrenia patients

Kreisl, JCBFM, 2013
Correcting for TSPO genotype increases sensitivity to detect difference between schizophrenia and controls

Without genotype as covariate $p = 0.085$

With genotype as covariate $p = 0.011$
Summary

• PBR28 PET study:
  – Leukocyte binding assay predicts TSPO genotype
  – TSPO genotype influences $[^{11}\text{C}]$PBR28 total binding

• PBR28 Post-mortem study:
  – TSPO genotype influences specific binding
  – Genotype correction increases ability to measure difference in schizophrenia and controls

• Correcting for TSPO genotype expected to improve clinical use of $[^{11}\text{C}]$PBR28
Outline of Talk

1. PET has high sensitivity and specificity
2. PET used in therapeutic drug development
3. Pharmacokinetic modeling: plasma concentration and tissue uptake
4. Study drug distribution: “peripheral” benzodiazepine receptor
5. Study drug metabolism: inhibit defluorination
$[^{18}\text{F}]\text{FCWAY}$: Defluorination
Bone uptake: human skull at 2 h
$[^{18}F]FCWAY$: Defluorination

$^{18}$F-fluoride ion accumulates in bone
Miconazole Inhibits Defluorination & Bone Uptake

[$^{18}$F]Fluoride

Skull

Brain

[$^{18}$F]FCWAY: Miconazole

Baseline 15 mg/kg 30 mg/kg 60 mg/kg
Disulfiram: Decreases Skull Activity & Increases Brain Uptake

Baseline

Images at 2 h in same subject. Disulfiram 500 mg PO prior night

Disulfiram
Disulfiram: Decreases skull uptake of fluoride & Increases brain uptake of $[^{18}\text{F}]$FCWAY

**Skull**

**Temporal Cortex**
Disulfiram: Decreases plasma fluoride & Increases plasma radiotracer $[^{18}\text{F}]{\text{FCWAY}}$

$[^{18}\text{F}]{\text{fluoride}}$

$[^{18}\text{F}]{\text{FCWAY}}$ (parent tracer)
Summary

1. PET has high sensitivity and specificity
2. PET used in therapeutic drug development
3. Pharmacokinetic modeling of plasma concentration and tissue uptake can measure receptor density
4. Study drug distribution: block distribution to periphery and increase distribution to brain
5. Study drug metabolism: inhibit defluorination
Self-Assessment Quiz: True or False?

- Imaging with positron emission tomography (PET) involves the injection of a radioactively labeled drug that emits a particle called a positron.
- PET shows the location of radioactivity in a cross section (or tomograph) of the body.
- PET can be used to quantify the density of specific proteins in the body.
- Compartmental modeling of PET data typically uses measurements over time of 1) PET images of the target tissue and 2) concentrations of unchanged parent radioligand in plasma.