



Deutsche  
Gesellschaft  
für Nuklearmedizin  
e.V.



# **Translational Research in Molecular Imaging and Radionuclide Therapy**

September 4 – 6, 2014

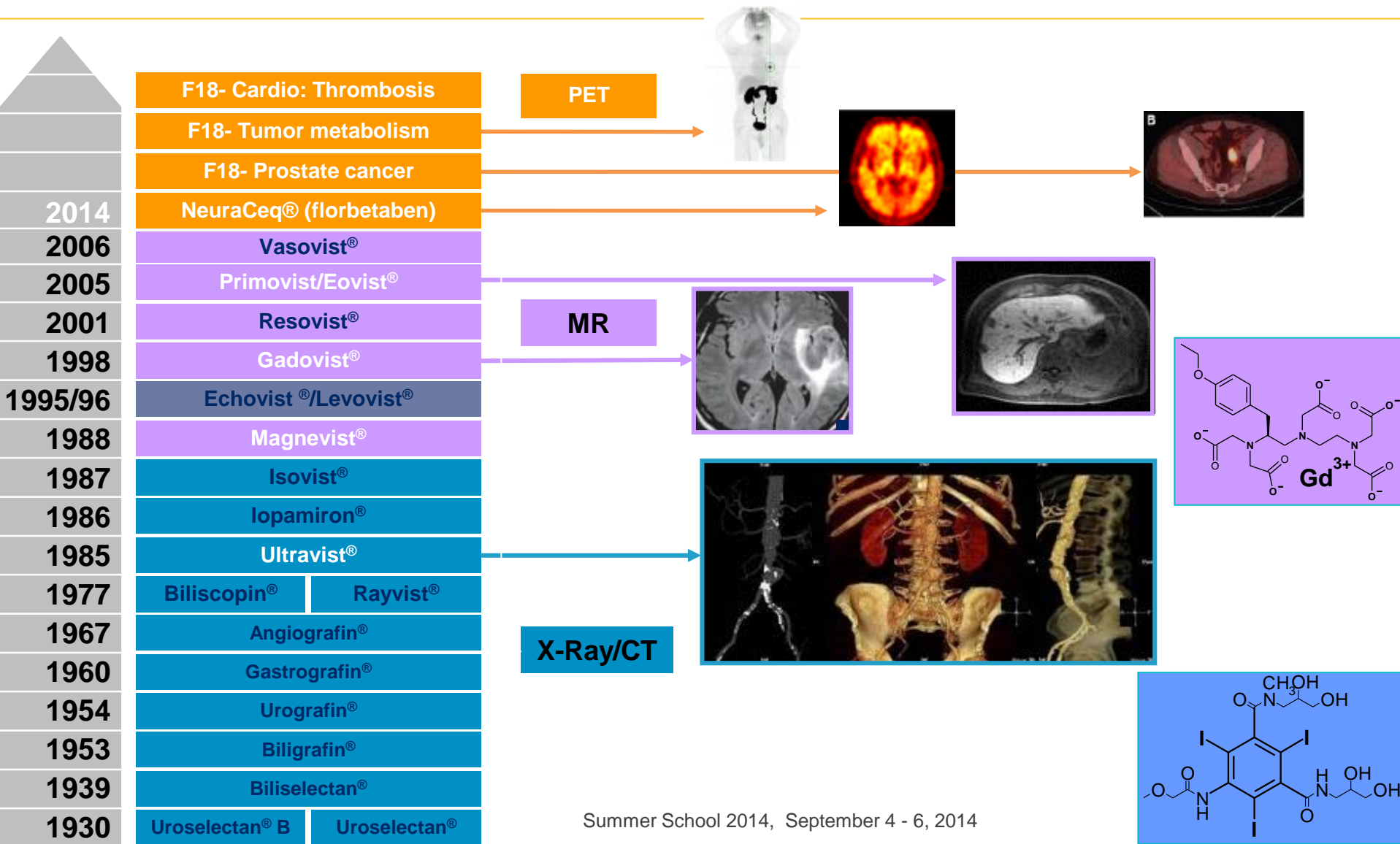
## **Tracer Development and Translation into Clinical Studies – Commercial Research**

Dr. Ludger Dinkelborg  
Piramal Imaging, Berlin

## Making the **Invisible Visible**

1. Introduction
2. Industrial Research Approach
3. Proof-of-Mechanism Concept
4. Examples from neuro-, onco- and cardiovascular imaging

# Diagnostic Imaging in Berlin – Innovation & Leadership Since 1930



# Piramal Imaging – at a Glance

## The Company

- Piramal Imaging was established in 2012 through Piramal's acquisition of Bayer AG's molecular imaging portfolio

## Locations

- R&D hub in Berlin, Germany and subsidiaries in Cambridge, UK and Boston, USA

## Our Focus

- Innovative radiopharmaceuticals for PET imaging for the diagnosis of life-threatening diseases in neurology, oncology and cardiology

## First Product in Market

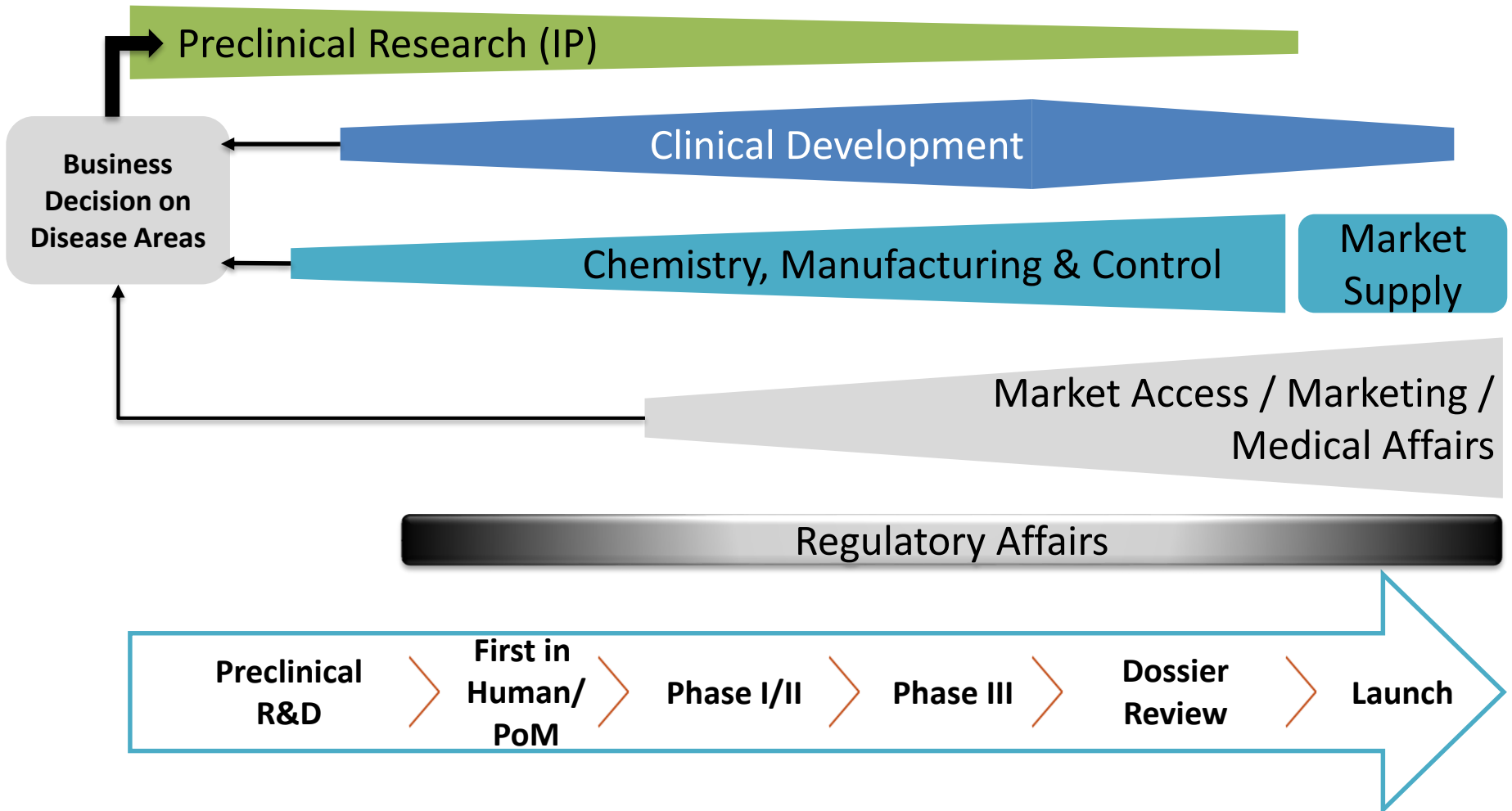
- NeuraCeq™ (Florbetaben F18) approved in Europe and USA

## Radiopharmaceuticals in Development

- Neurology: MAO-B for neuro-inflammation; Tau for neurodegeneration
- Oncology: FSPG for brain tumor and Bombesin for prostate cancer
- Cardiovascular: GP1 for thrombus imaging



# Stakeholders in commercial research



# Personalized Medicine and Molecular Imaging

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**Diagnosis Down to Genetic Defect**



**Increasingly Specific (Targeted) Therapies**



**Increasingly Costly Therapies**



**Increasing Need for Early Diagnosis,  
Disease Characterization, Targeted Therapy,  
Monitoring and Justification (Outcome)**

# Molecular Imaging versus Therapeutic Research

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- At the start of a molecular imaging research project, a targeting mechanism as well as lead structures are usually known from previous therapeutic studies
  - Only limited target identification and validation in animals
  - Low dose of intravenously injected “tracers” allows for clinical proof of mechanism (target validation) during research phase
  - Only intravenous application tracers
  - Additional intellectual property is generated by inventing methods to include the radioisotope (e.g. F-18) into the molecule of choice and optimizing its pharmacokinetics
- ⇒ This results in an earlier selection of successful development candidates, a lower attrition rate at late development stages leading to reduced R&D costs if compared to therapeutic drugs

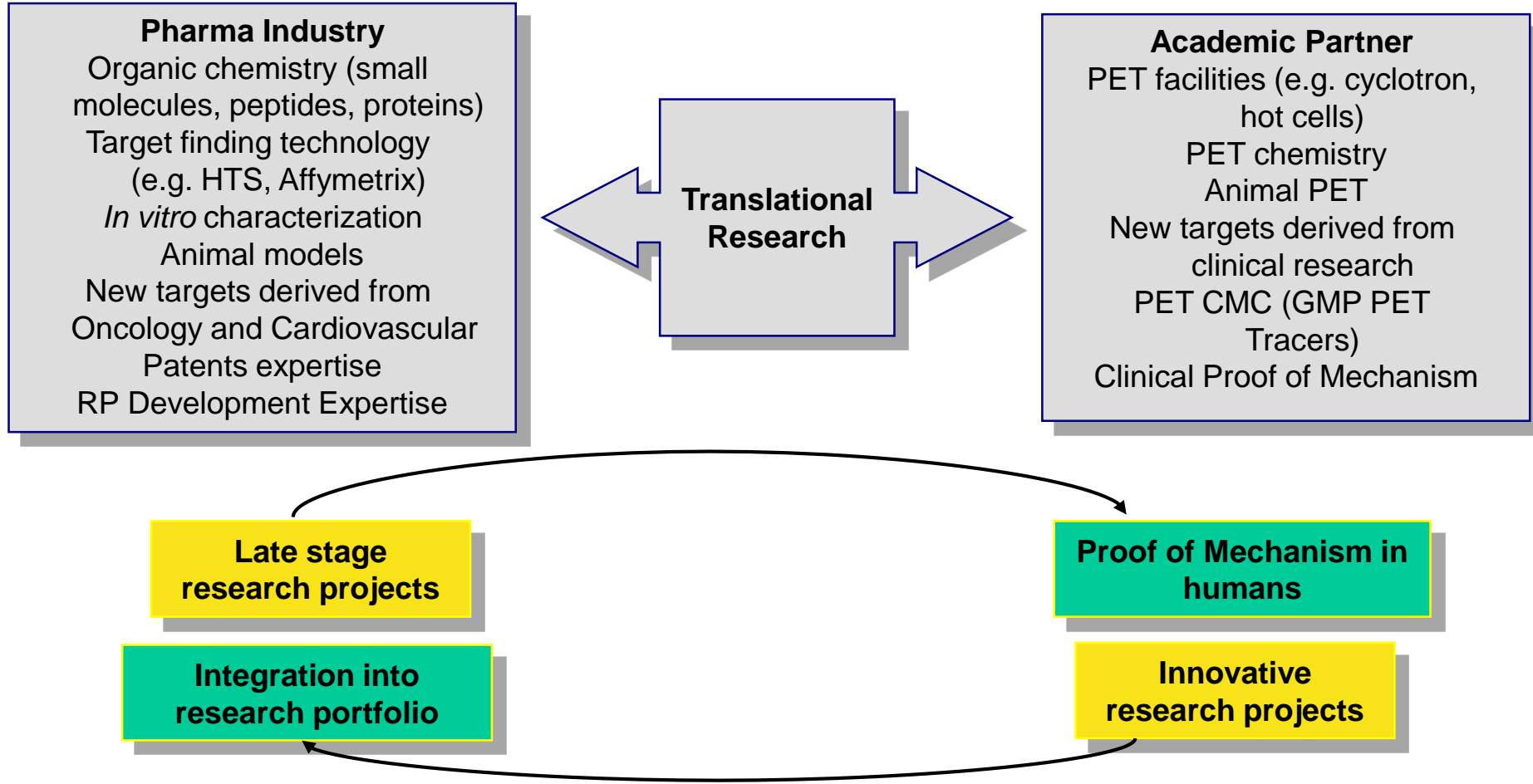
# Early selection of promising radiopharmaceuticals for development

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- Proof-of-mechanism (PoM) is the early evaluation of a pre- or clinically based targeting hypothesis (mechanism of action) of a radiopharmaceutical in patients making use of microdosing (or exploratory IND in the USA) procedure
- In addition, information on target accessibility (*in vivo* stability, pharmacokinetics) of the investigated tracer is obtained
- Microdosing trials (ICH guideline M3, R2, Dec 2009)
  - tracers injected < 100 µg
  - < 1/100th the dose calculated to yield a pharmacological effect
  - introduced in June 2004
- Limited safety requirements
- Labeling of tracers will be done on-site

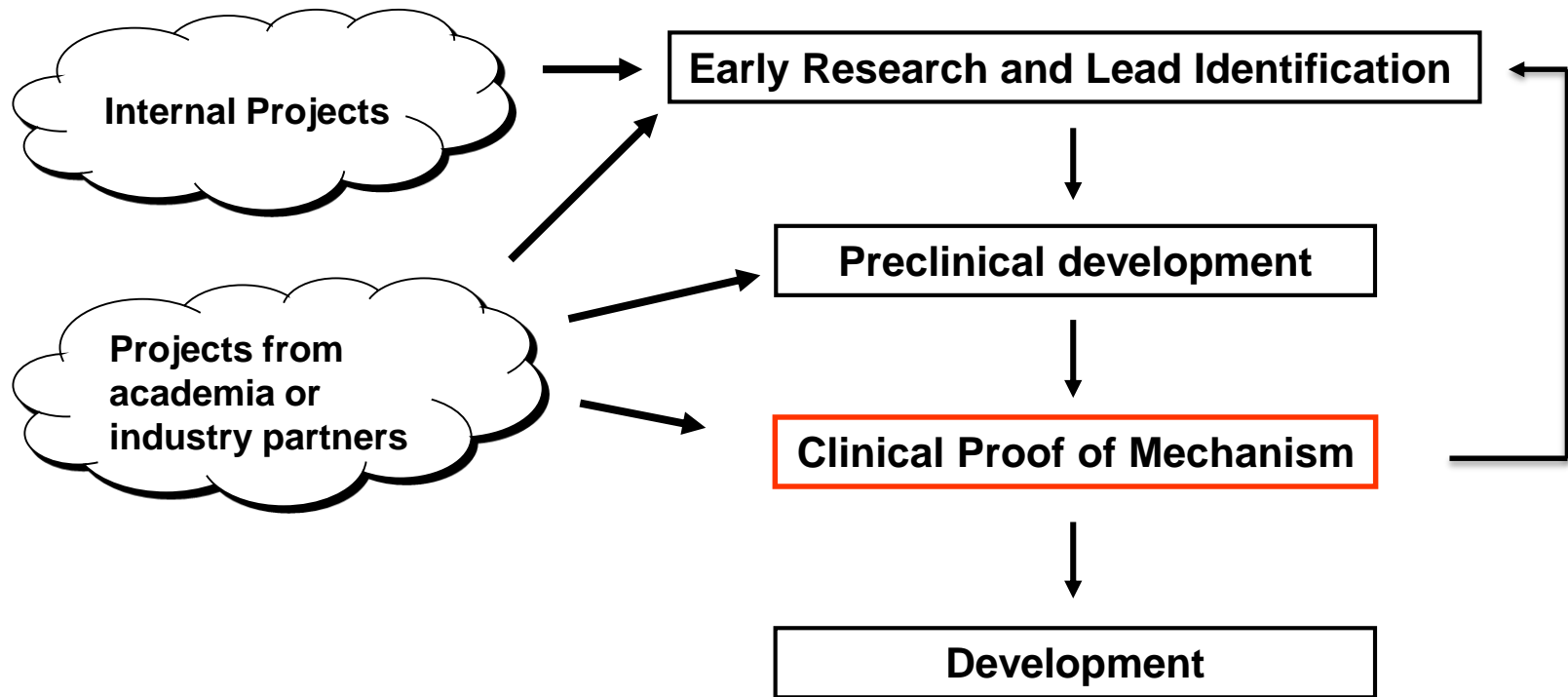


# Translational Research in PET



# Early Proof of Mechanism in PET

## Rapid Evaluation of Efficacy



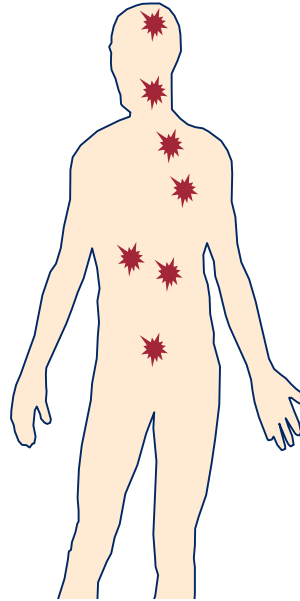
- Early selection of development candidates
- Higher success rates in later development

# Oncology Imaging - Medical Needs Beyond FDG

## Medical Needs From Top to Toe

### FDG 'Problem' Areas:

- Brain tumors & mets
- H&N, thyroid Ca
- Lung Ca (differentiation from inflammation)
- Breast tumors
- HCC & Upper GI tract
- Pancreatic Ca
- Colorectal Ca
- Prostate Ca
- Ovarian Ca
- Renal cell Ca



**FDG is not tumor-specific**

**FDG falls short in several indications & settings**

### Medical Need:

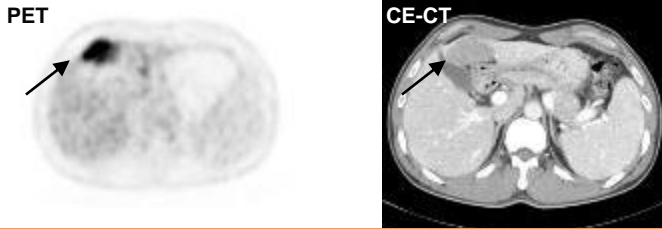
- Increased sensitivity (FDG non-avid tumors)
- Increased specificity (inflammation differentiation)
- Improved tumor to background ratio (no uptake in healthy organs, e. g. brain, muscle, pelvis)
- Early therapy monitoring

### Our approach:

- Targeting of intermediary tumor metabolism & specific surface receptors
- ↓
- Use of F-18-labeled amino acids as pre-cursors for anaplerotic reactions and detoxification processes & tumor-specific receptor ligands

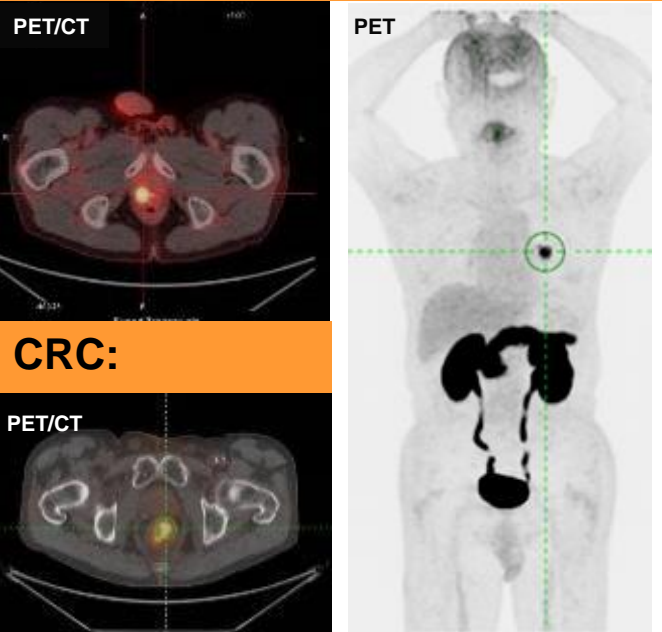
# FSPG PET imaging of system $x_C^-$ transporter activity reveals a dominant pathway in tumor metabolism

## HCC:

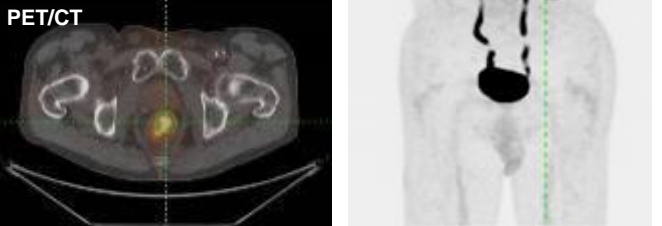


## Prostate Ca:

## NSCLC:



## CRC:



## Goals:

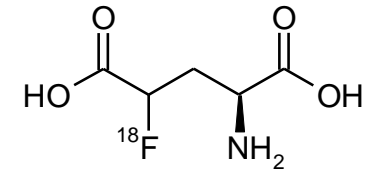
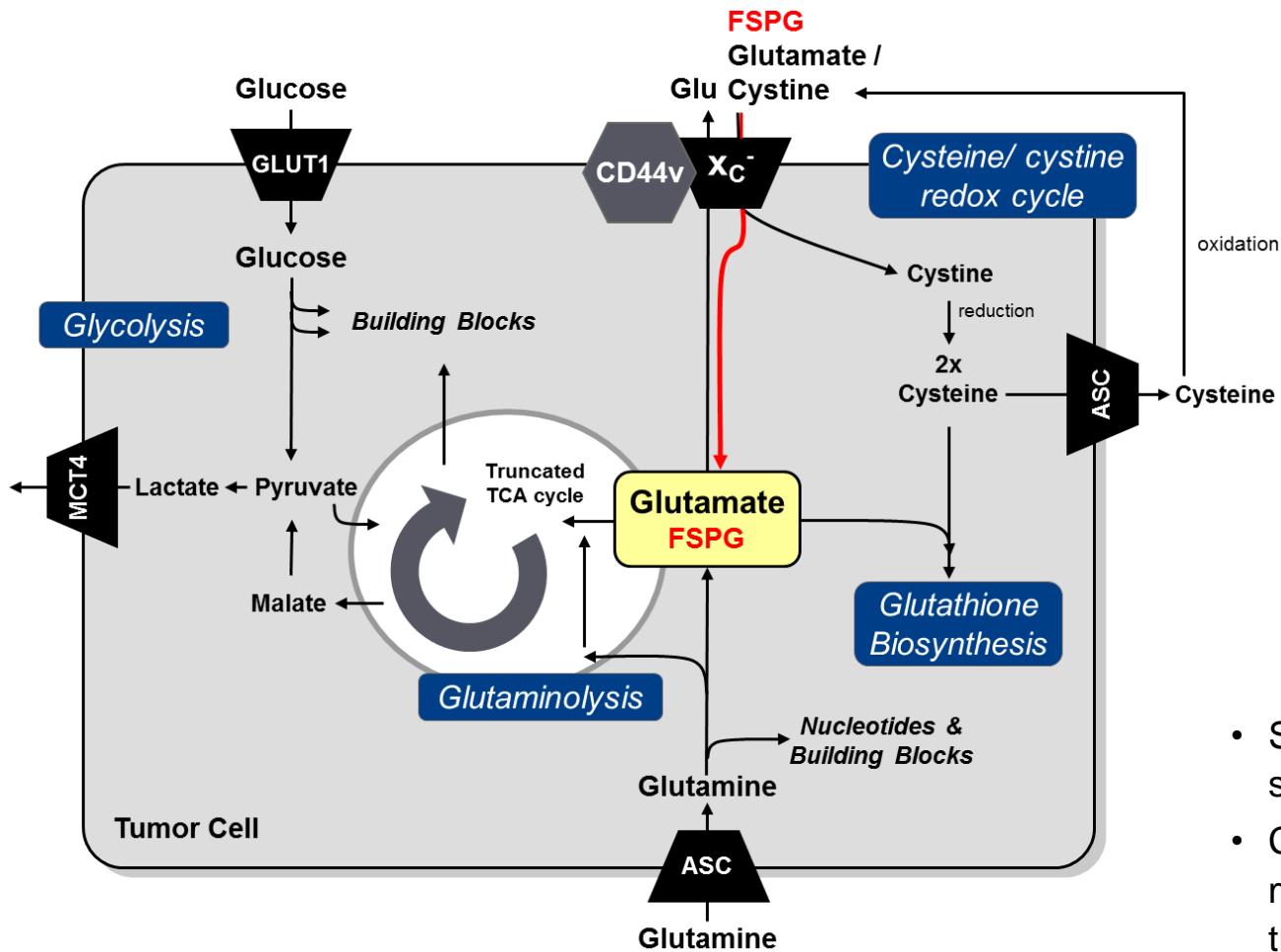
- Identification or exclusion of malignancies in patients where FDG has proven inadequate sensitivity or specificity
- Potentially guiding therapy decisions for improved outcome

## Tracer profile:

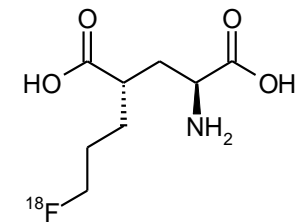
- F-18 labeled glutamate derivative, specifically transported by system  $x_C^-$ , which correlates with CD44 expression in tumors
- Rapid clearance, low background especially in brain enabling detection of brain metastases

Baek et. al Clin Cancer Res. 2012 Oct 1;18(19):5427

# FSPG visualizes a dominant pathway in tumor metabolism



4-[F-18]F-glutamate

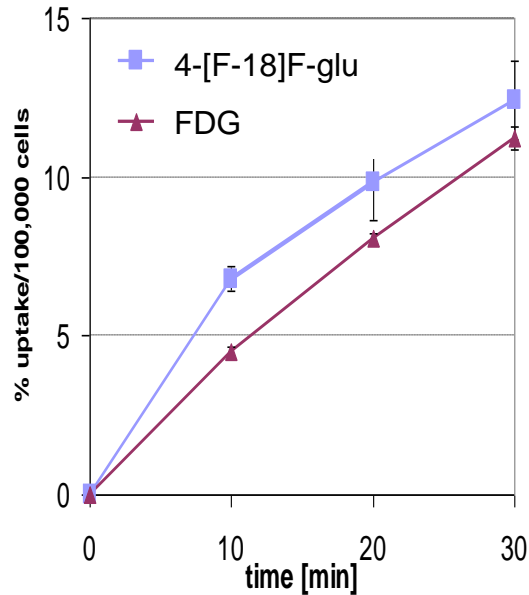


4-[F-18]F-propyl-glutamate (FSPG)

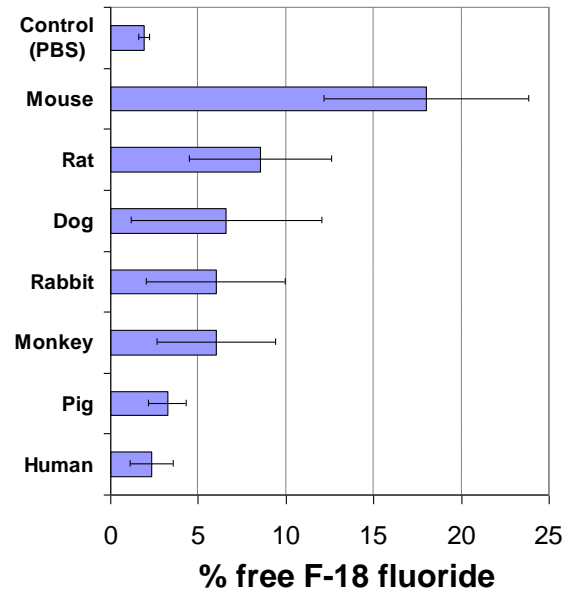
- Specifically transported by system  $x_C^-$
- Correlates with cancer stem cell marker CD44 (promotes transporter activity)

# 4-[F-18]F-glutamate: Preclinical characterization of a new probe to study tumor metabolism

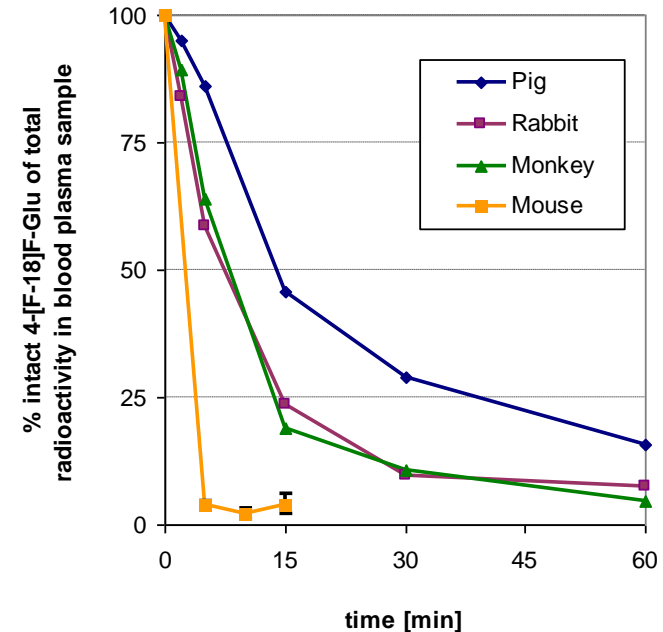
High tumor cell uptake of 4-[F-18]F-glu



Species dependent defluorination (in vitro)



Species dependent defluorination (in vivo)

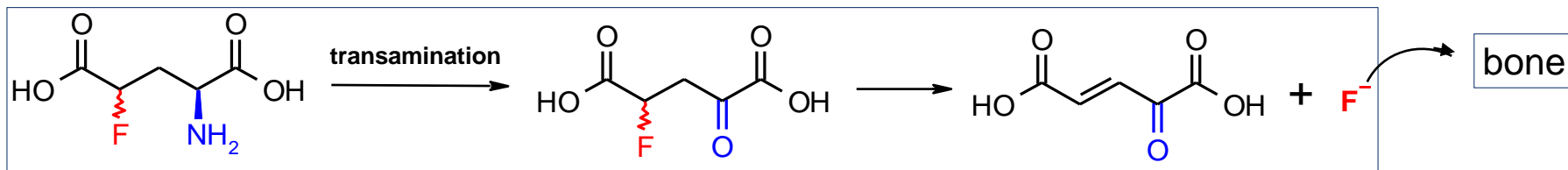
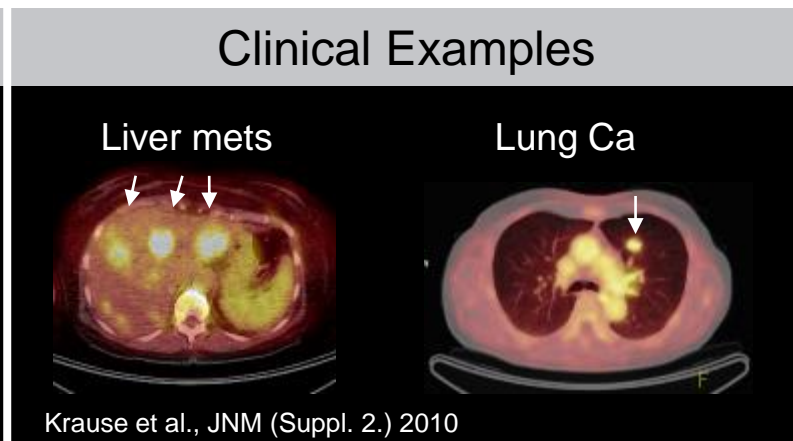
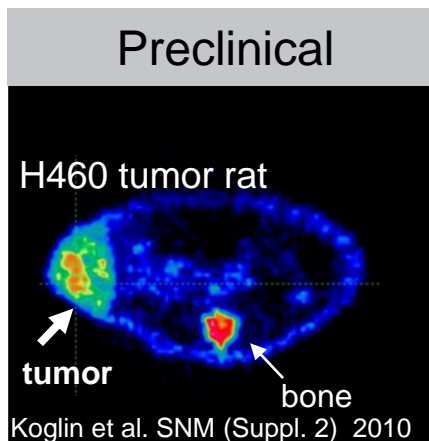
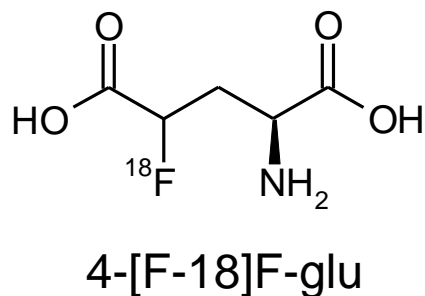


- Uptake in vitro comparable to FDG
- Tumor visualization in rodents

- Highest stability in human and pig blood samples

- Highest stability in pigs

# First generation PET-Tracer: 4-[F-18]F-glutamate



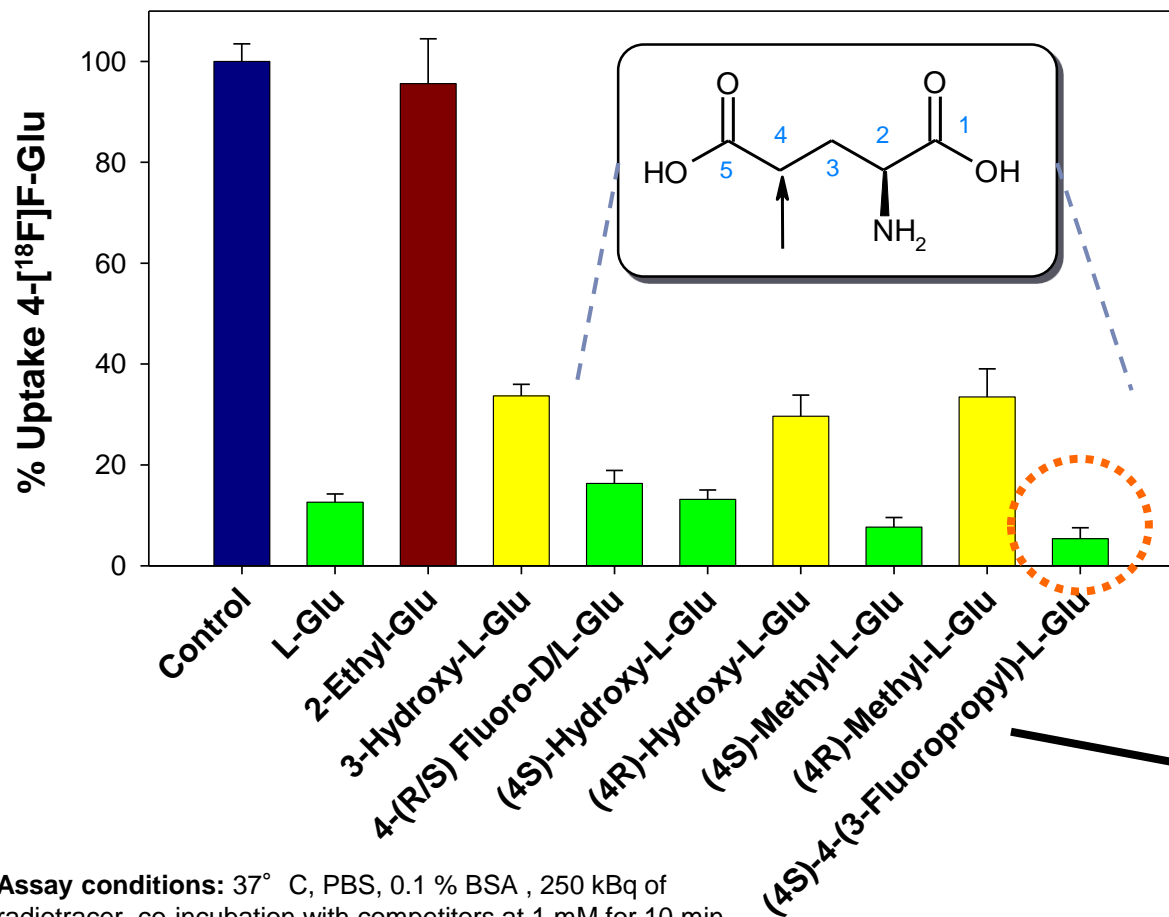
- Tumor visualization
- Favorable low background except bones at later time points
- Stability lower than anticipated

Concept proven  
*But* higher  
stability needed

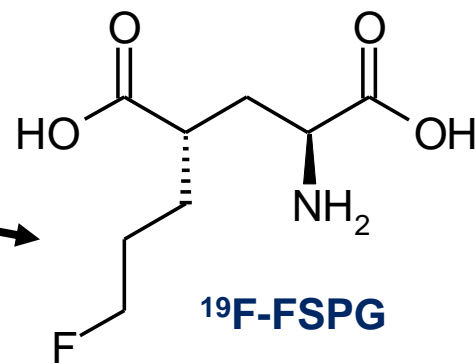
Find stable  
compound

# 4-Substituted glutamate derivatives still show inhibition in cell uptake competition assay

A549 cell competition assays using radiolabeled glutamate for screening:



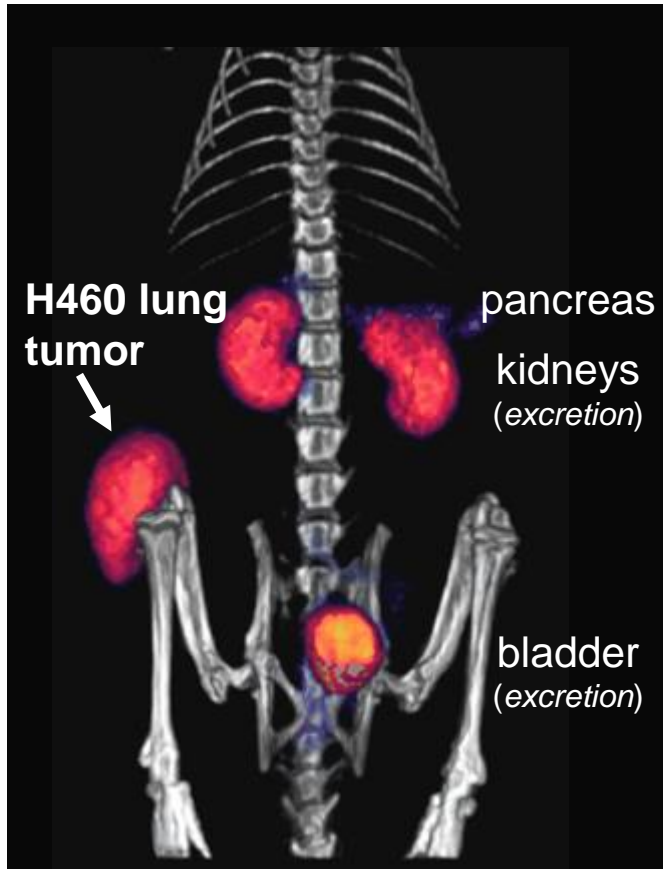
- Substitutions in 4-position tolerated
- Derivatives in 4S-configuration showed high competition
- (4S)-3-Fluoropropyl derivative promising



Assay conditions: 37° C, PBS, 0.1 % BSA , 250 kBq of radiotracer, co-incubation with competitors at 1 mM for 10 min



# FSPG visualizes tumors in rodent models with low background



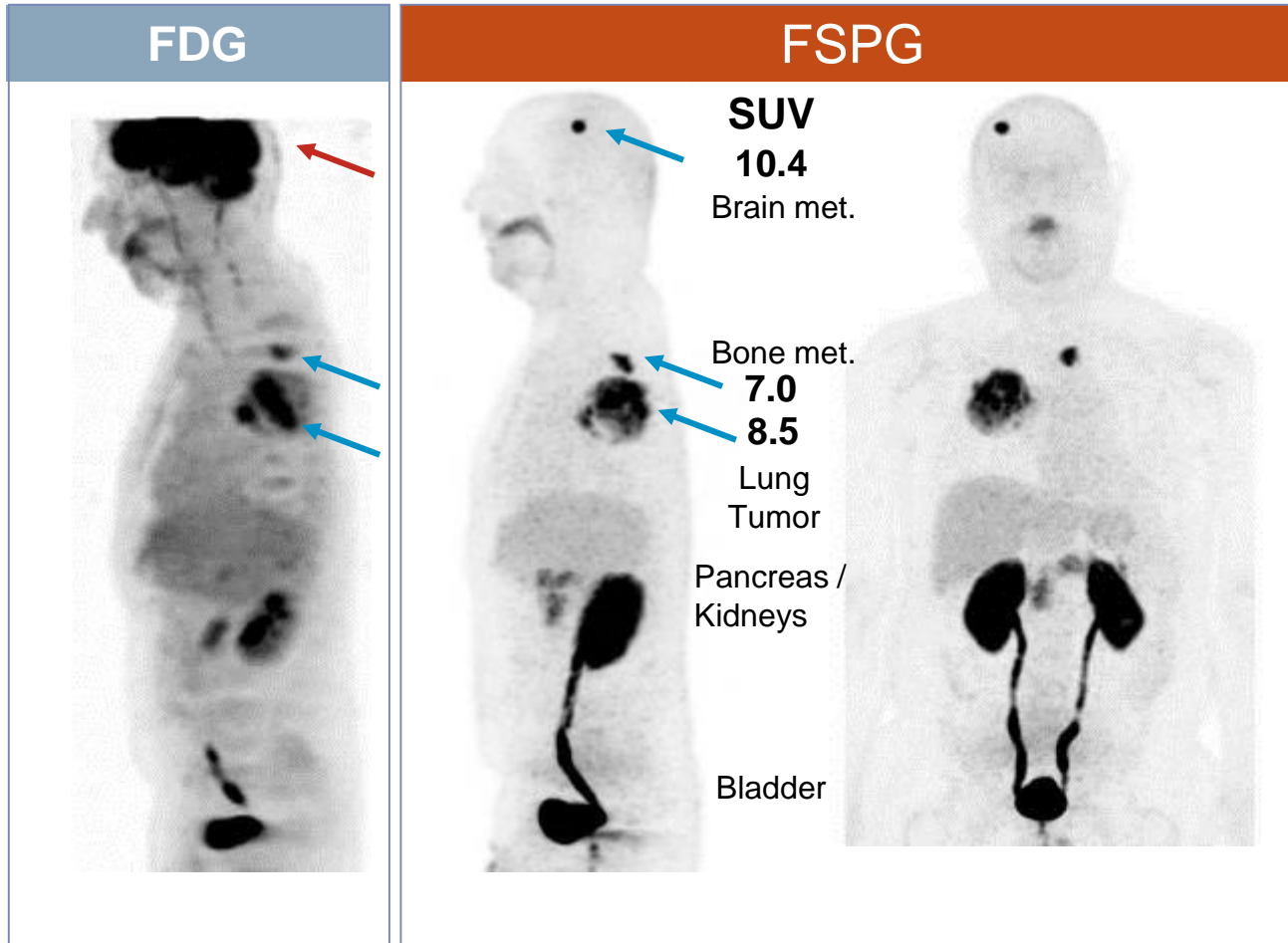
## FSPG PET imaging:

- High tumor uptake and retention
- No defluorination
- Rapid renal clearance, low background in healthy tissues
- Excellent tumor imaging in various models
  - Lung cancer (A549, H460, LL)
  - Prostate cancer (LNCaP, PC3, DU-145)
  - Breast cancer (MCF7, 4T1)
  - Liver cancer (Huh7, MH3924a)
  - Glioblastoma (GS9L)
  - Colon cancer (HT29, HCT116)
  - Melanoma (SK-Mel3, B16F1)

Preclinical PET / CT imaging of human H460 lung tumors in nude rats, 90 min p.i., Inveon PET/CT

Koglin et al., Clin Canc Res 2011

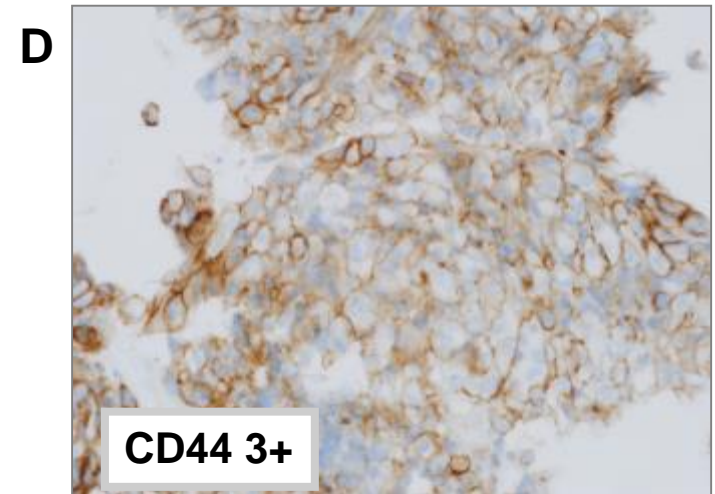
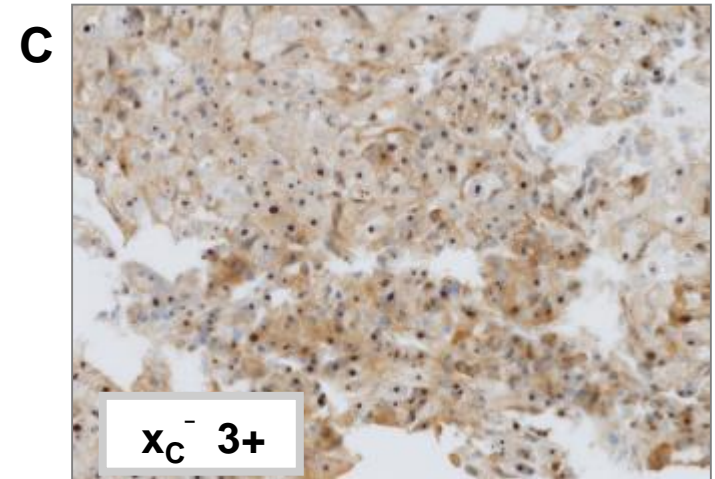
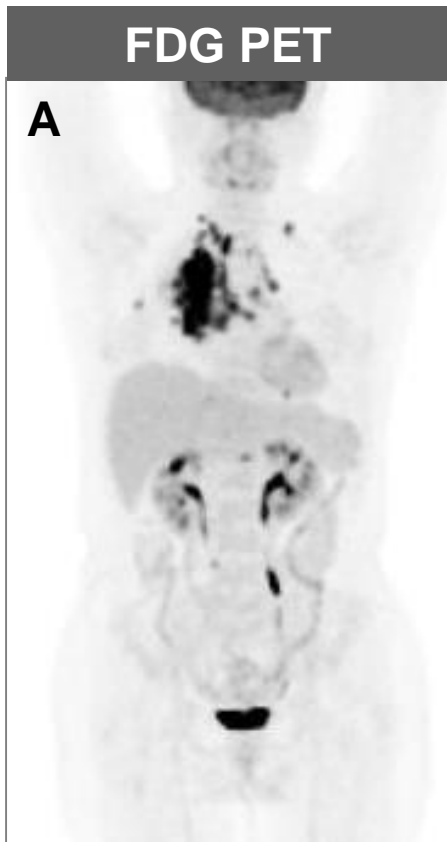
# FSPG accumulates in primary and metastatic NSCLC



## FSPG:

- High tumor uptake and retention
- Low background
- Rapid blood clearance
- Rapid urinary excretion

# FSPG Uptake correlates with staining of system $x_C^-$ & CD44



Courtesy of Prof. D.H. Moon & Prof. S.J. Oh (Asan MC)

High detection rate in NSCLC & additional lesions (arrows) found with FSPG compared to FDG

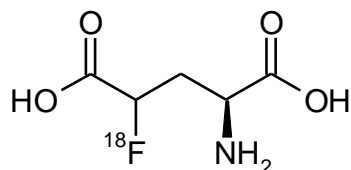
Baek S et al. Clin Cancer Res. 2012



# Iterative Radiotracer Development: The glutamate example

## 4-<sup>18</sup>F-fluoro glutamic acid

Preclinical & chemical development

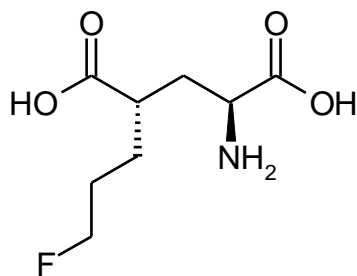


First in man, proof of mechanism

Stability lower than anticipated

## 18F-fluoropropyl glutamic acid (FSPG)

Preclinical & chemical development



First in man, proof of mechanism

Identify suitable tumor indication for development

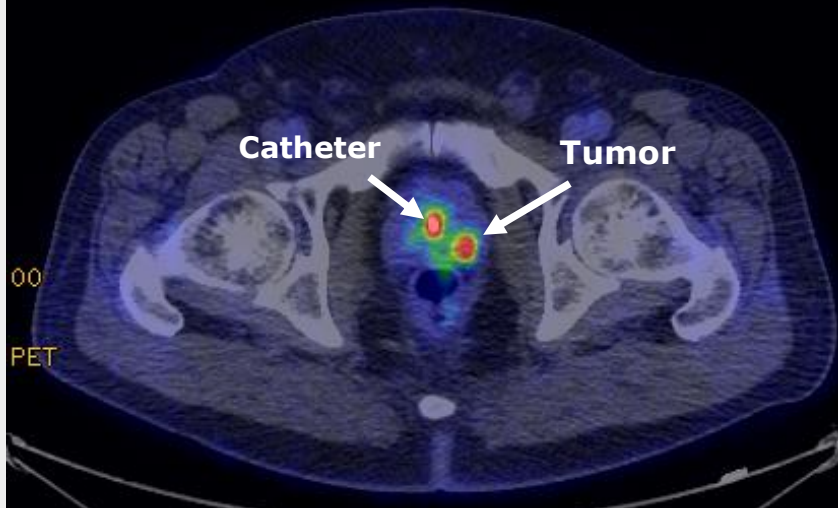
# Bombesin: Improve prostate cancer detection and location



Fire-bellied toad (*Bombina bombina*)



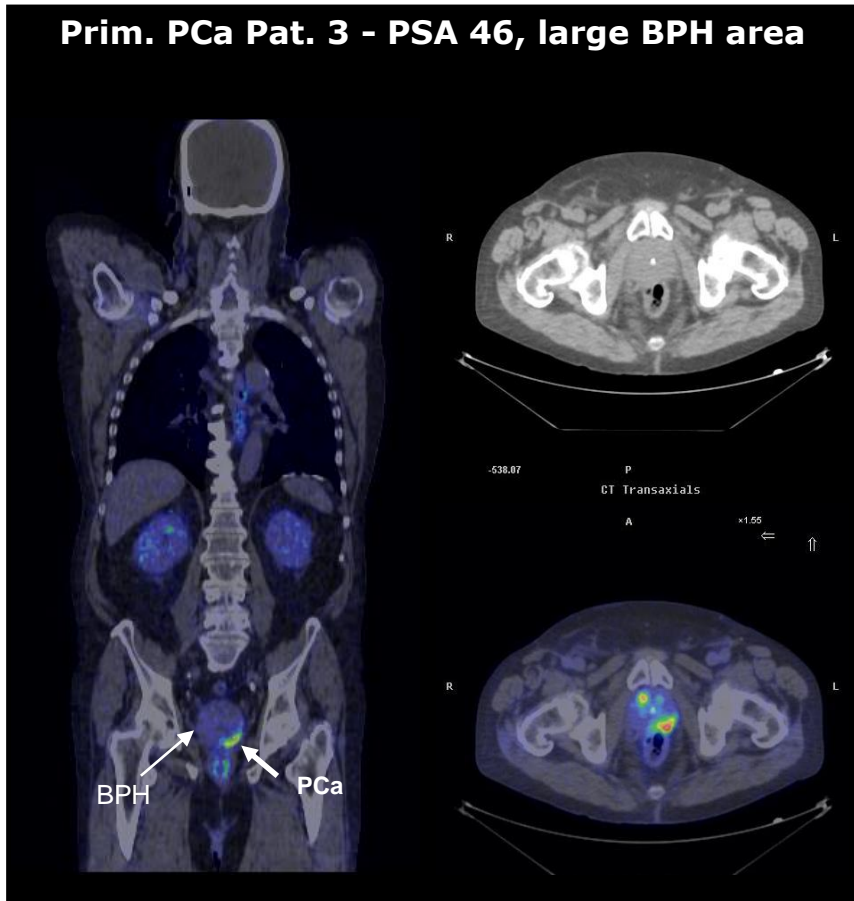
Primary PCa, PSA 24, Gleason 3+4



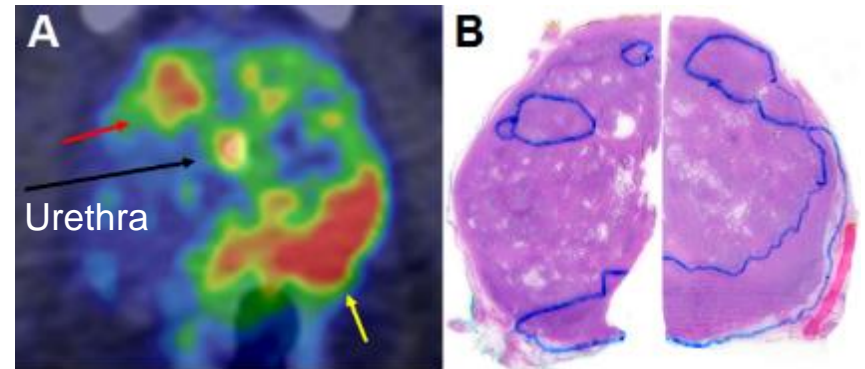
## Tracer profile:

- Ga-68 labeled Bombesin antagonist specifically binding to gastrin-releasing peptide receptors (GRPr) overexpressed in prostate cancer
- Histopathological analyses indicates significantly higher uptake in cancer compared to benign tissue
- High Sensitivity (89%) and Specificity (81%) for PCa detection
- Accurate detection of PCa in 10/11 subjects, with change in patient management in 36% (4/11) of subjects

# Ga-68-Bombesin PET correlates well with tumor histopathology



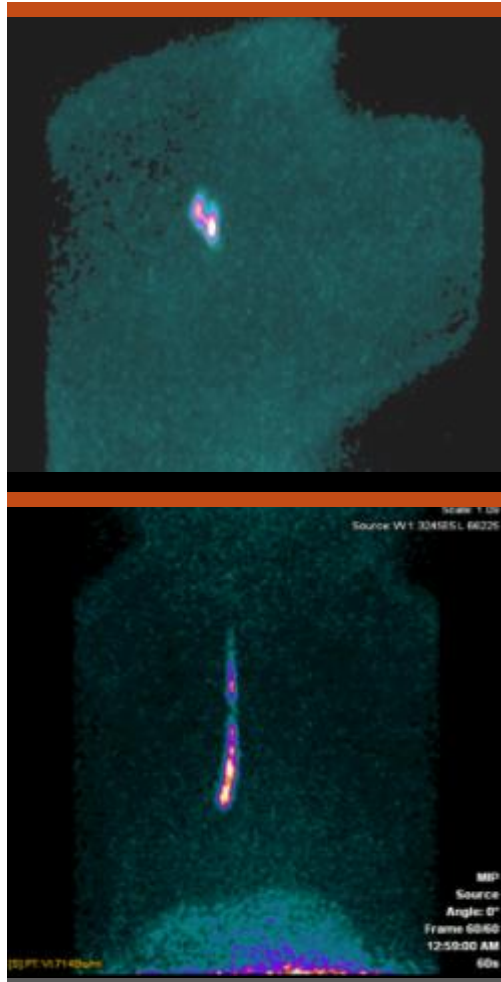
*Courtesy of PET Centre Turku, Finland (H. Minn)*



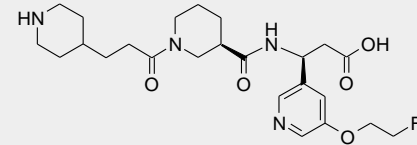
## Next steps:

- Initiate Phase I/II study in EU to evaluate potential of  $^{68}\text{Ga}$ -Bombesin for tumor detection in low, medium and high risk patients

# GP1: Identify patients at high risk for thrombo-embolic events



## Tracer profile:



- GP1 is a <sup>18</sup>F-labelled, specific high affinity binder to the GPIIb/IIIa receptor of human platelets
- Visualization of small thrombi should allow improved therapeutic decision making

## Potential indication:

- PET-Imaging of thrombi and sources of emboli that caused cerebral ischemic events or exclusion of thrombotic or embolic cause in patients with Transient Ischemic Attacks (TIA)

## Next steps:

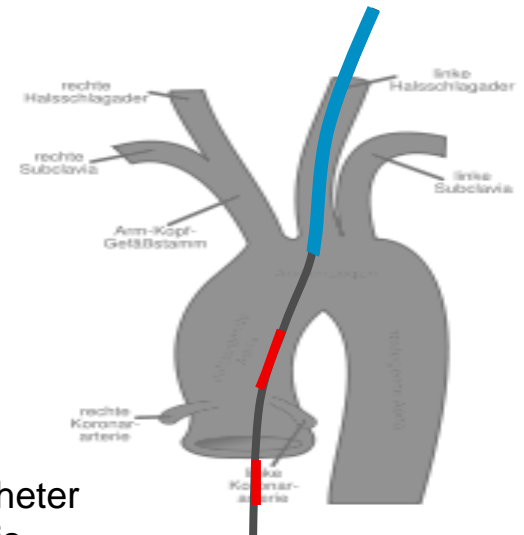
- First-in-man study ongoing

# GP1 images thrombi by binding to activated platelets

## Preclinical Thrombus PET-Imaging



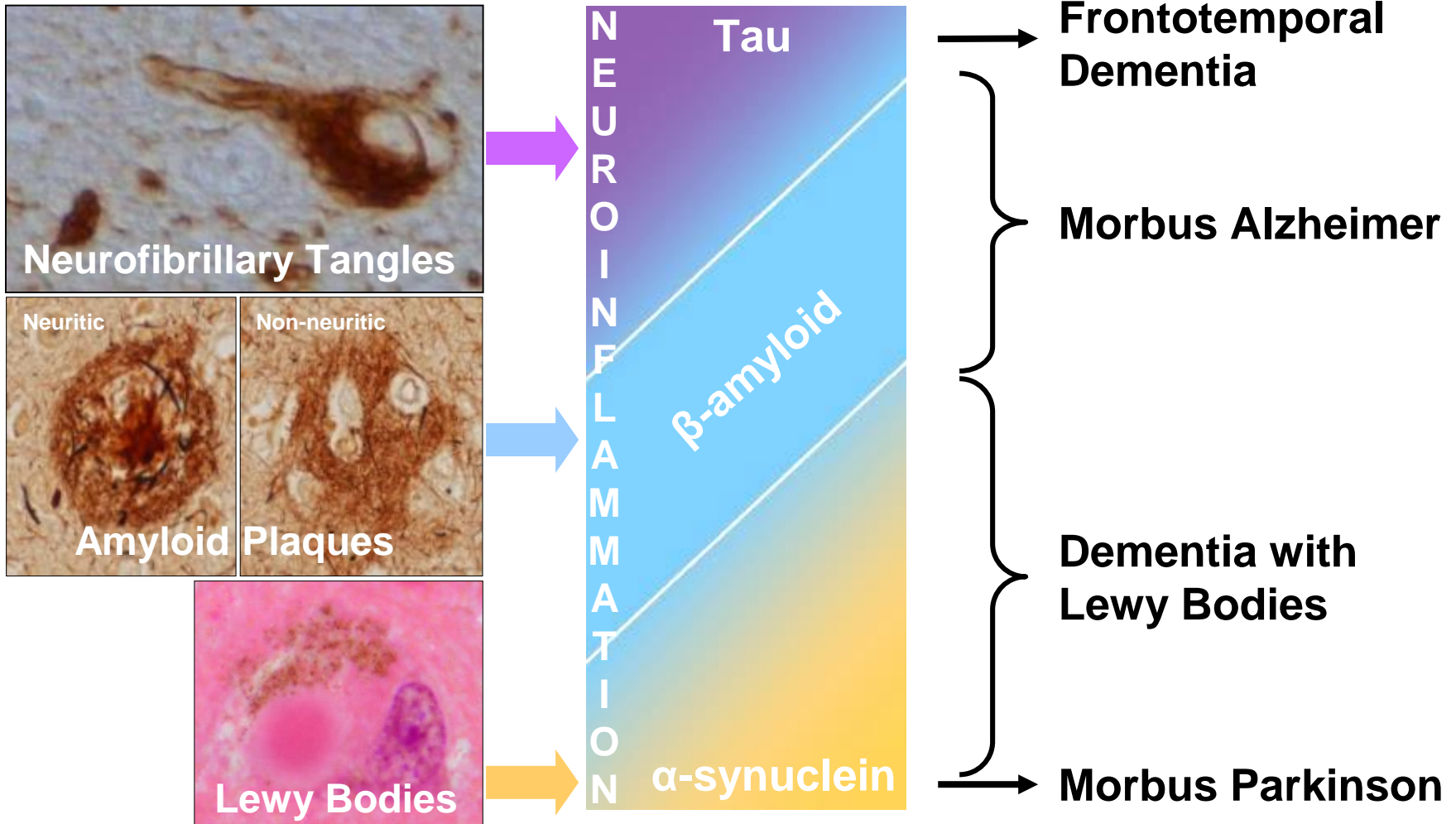
In monkeys, GP1 demonstrated sensitive and specific detection of platelet depositions on catheter and endothelium surfaces as well as on emboli



Introduction of a catheter through the A. carotis into the ascending aorta of cynomolgus monkeys



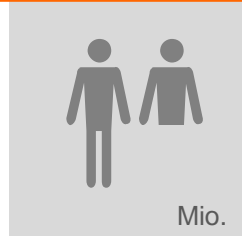
# Targeting pathologic proteins of neurodegenerative diseases



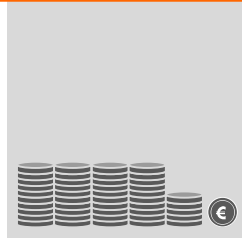
# Alzheimer's disease: The health care challenge

## Today

- >1.5 Mio. Germans are demented
- ~65 % of those (1 Mio) have AD

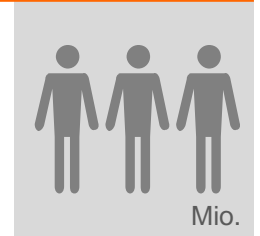


- **Costs Germany:**  
25-45 Mrd €/year

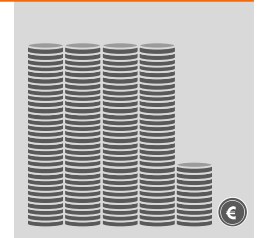


## 2050

- 3 Mio. Germans will be demented
- ~2 Mio will have AD



- **Costs Germany:**  
>130 Mrd €/year

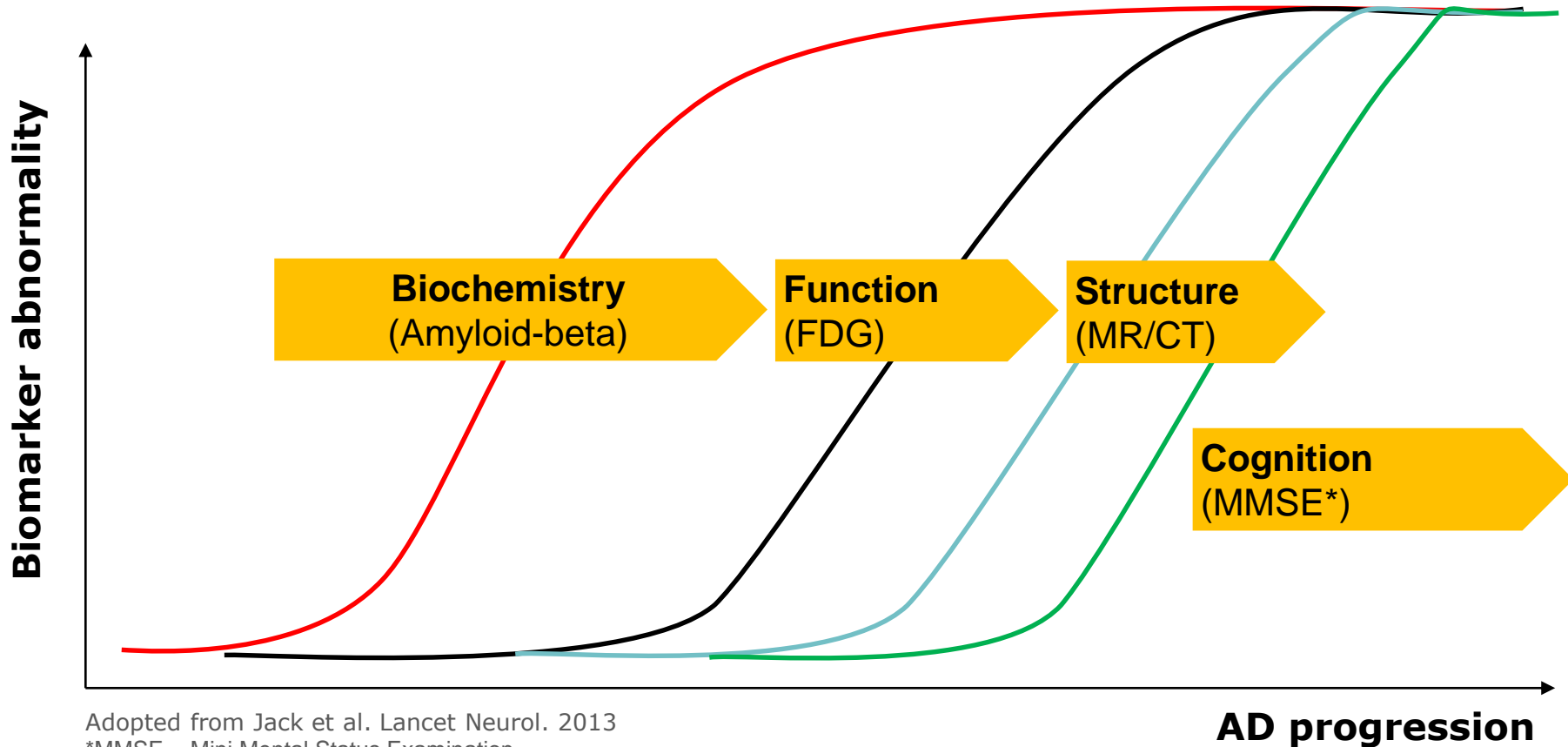


### Urgent need for:

- Early, accurate diagnosis of AD
- Disease Modifying Drugs
  - Delaying onset of Alzheimer's dementia by 5 years  
→ 50% fewer patients in 2050 → Savings in the Germany: 65 Mrd/year<sup>2</sup>

# Amyloid show earliest abnormality during progression of AD

- Amyloid shows abnormality 10-15 years before cognitive decline:

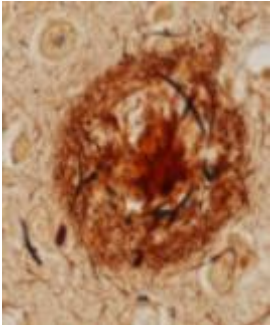


# Amyloid-beta and Tau play crucial roles in Alzheimer Disease pathology

## Amyloid-beta

### "Plaque"

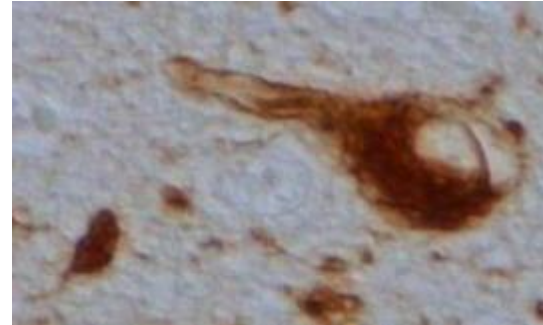
Deposition starts before cognitive decline occurs



## Tau

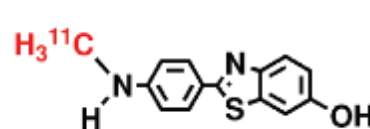
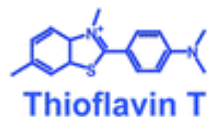
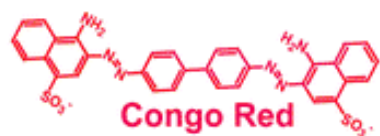
### "Neurofibrillary Tangle"

Correlates with cognitive decline

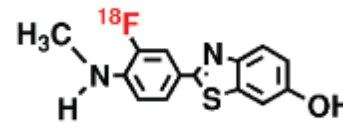


**So far, confirmation of AD during autopsy by detection of amyloid plaques & neurofibrillary tangles using silver staining and/or immunohistochemistry**

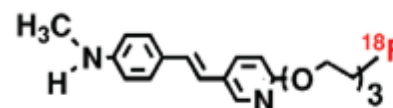
# From Dyes for Staining of Amyloid Plaques to Tracers for PET Imaging



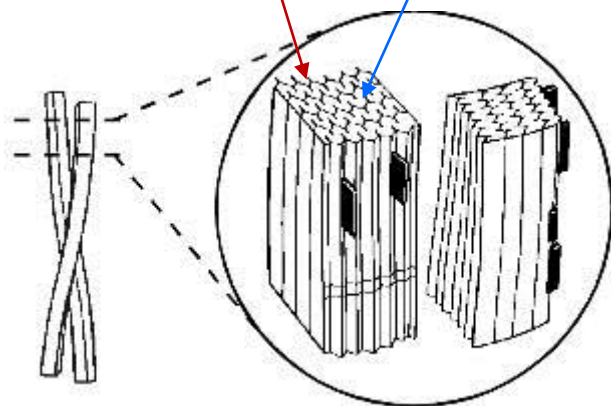
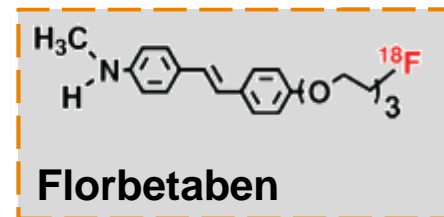
$^{11}C$ -PIB



Flutemetamol



Florbetapir



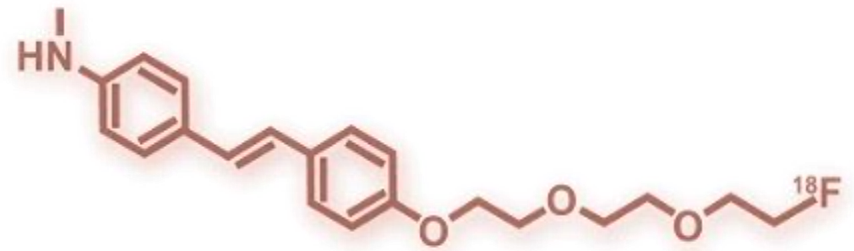
## $\beta$ -sheet structure of amyloid

Sites of binding of Congo red dye are indicated by the solid blocks  
(Glennner, NEJM 1980)

$^{11}C$ -PIB and  $^{18}F$ -stilbene derivatives were discovered by academic groups, patented and subsequently licensed to companies for development

# Florbetaben: A F-18 Labeled Stilbene with High Affinity to Amyloid-Beta

- Stilbene derivative
- Labeled with F-18 for PET detection (PEG linker)
- Trade name: NeuraCeq
- High affinity to amyloid-beta plaques
- High specificity: no binding to NFTs (Tau), Pick bodies, Lewy bodies
- Ability to cross the blood brain barrier and detecting amyloid-beta deposits in the brain



**Physical Half Life:**

110 min

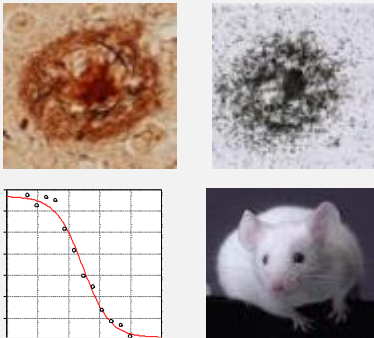
**Shelf Life:**

10 hours

# Florbetaben - Overview of non-clinical development

## Pharmacology

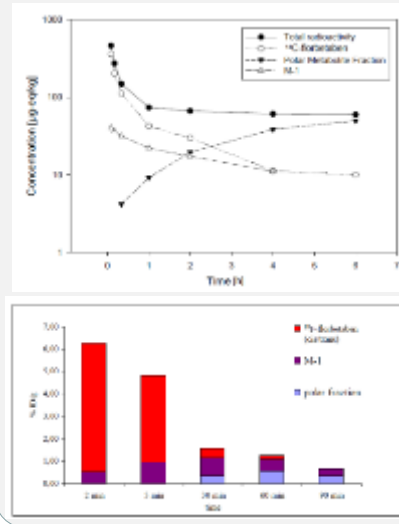
- High affinity and specific binding to A $\beta$  plaques



- **Favorable brain penetration and clearance**

## Pharmacokinetic

- PK profile
- Distribution
- Metabolism
- Excretion



## Safety

Studies in rats and dogs to show safety in:

- Central nervous system
- Cardiovascular system
- Respiratory system
- Renal function

➤ **No concerns**

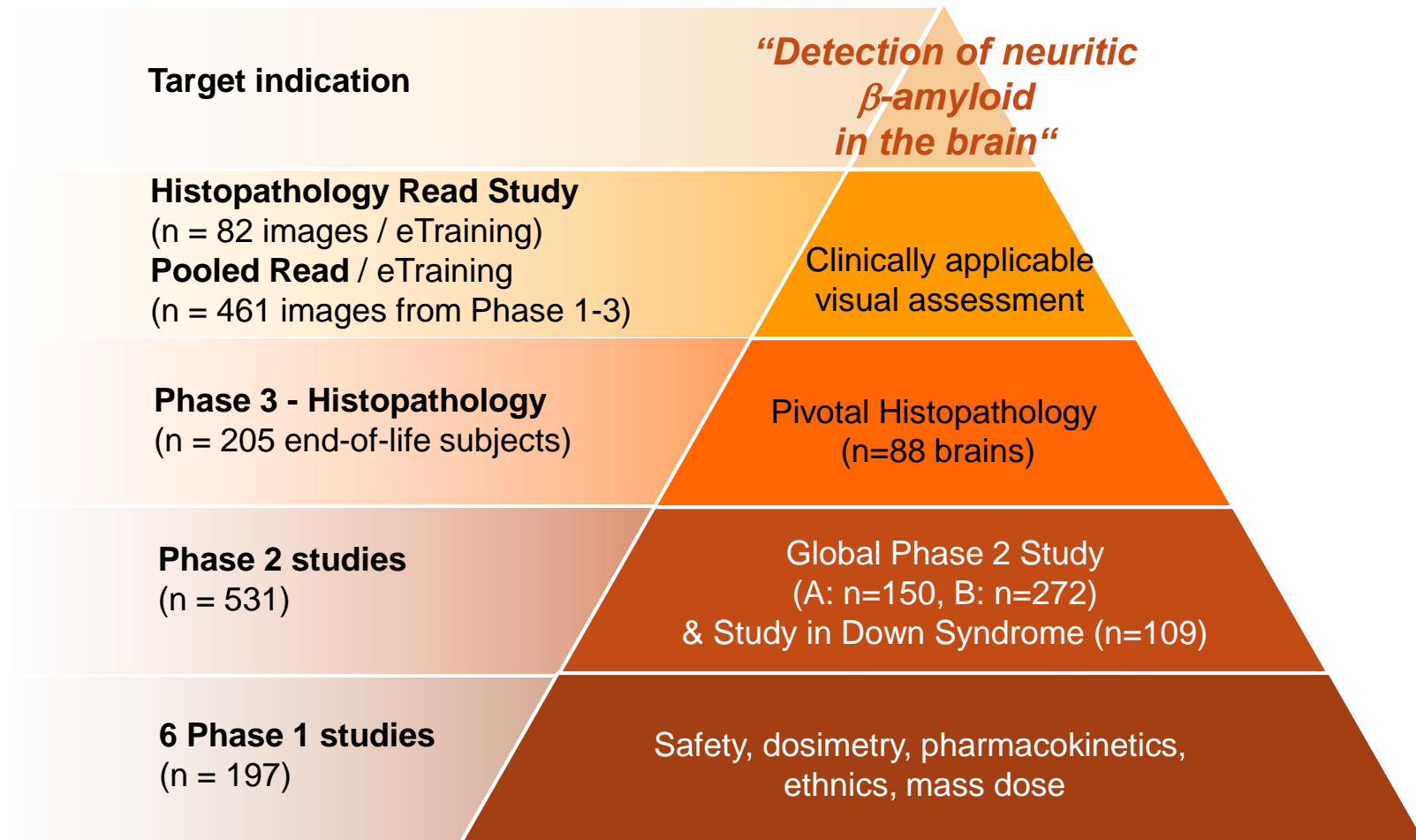
## Toxicology

- Single & repeated dose tox in rats and rabbits or dogs
- Genotoxicity
- Carcinogenicity
- Testing potential impurities and decayed clinical formulations

➤ **No concerns**

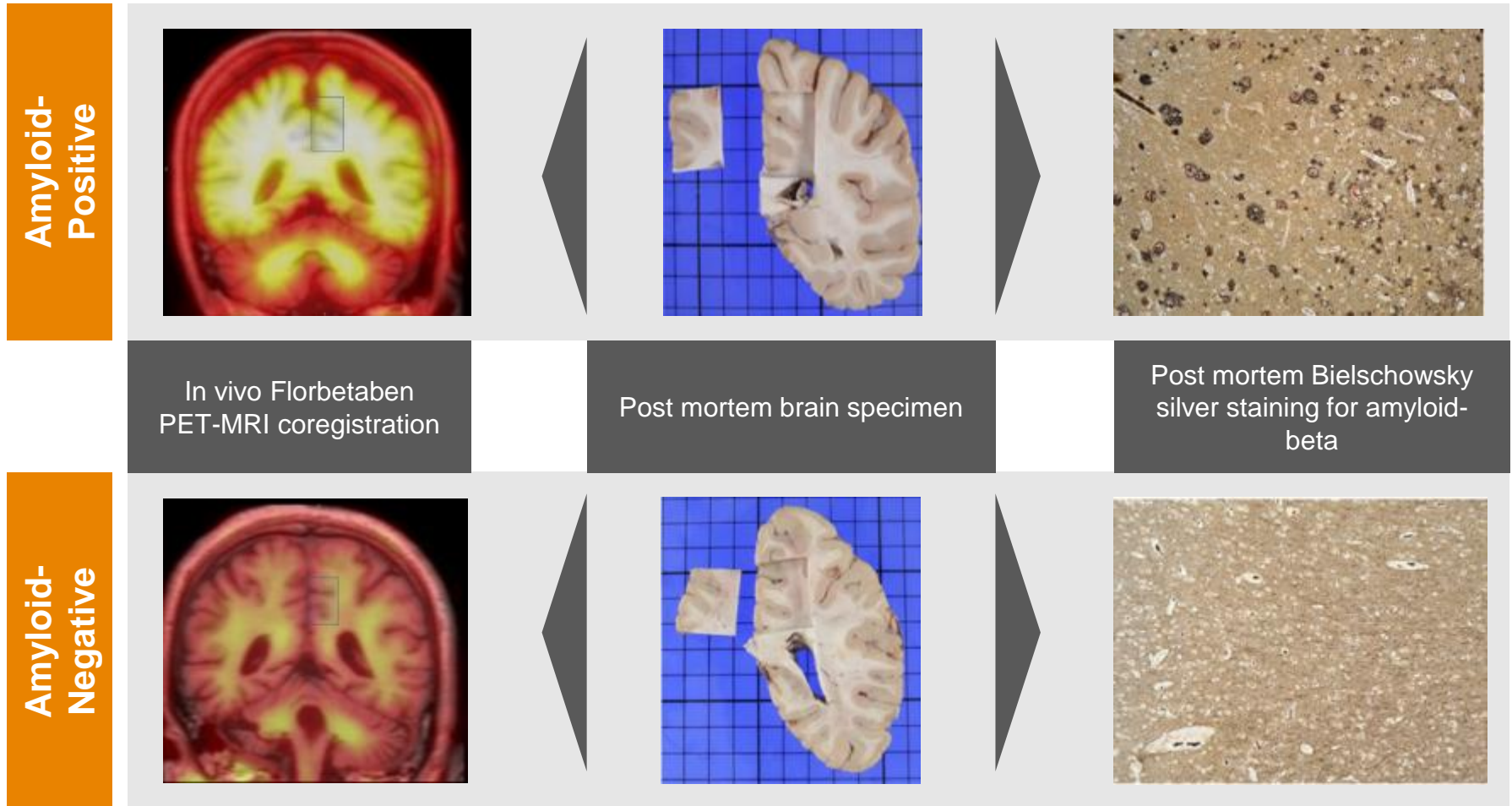
- **Favorable pharmacologic and pharmacokinetic profile**
- **No non-clinical safety and toxicological concerns**

# Florbetaben Clinical Development: >900 Subjects Studied



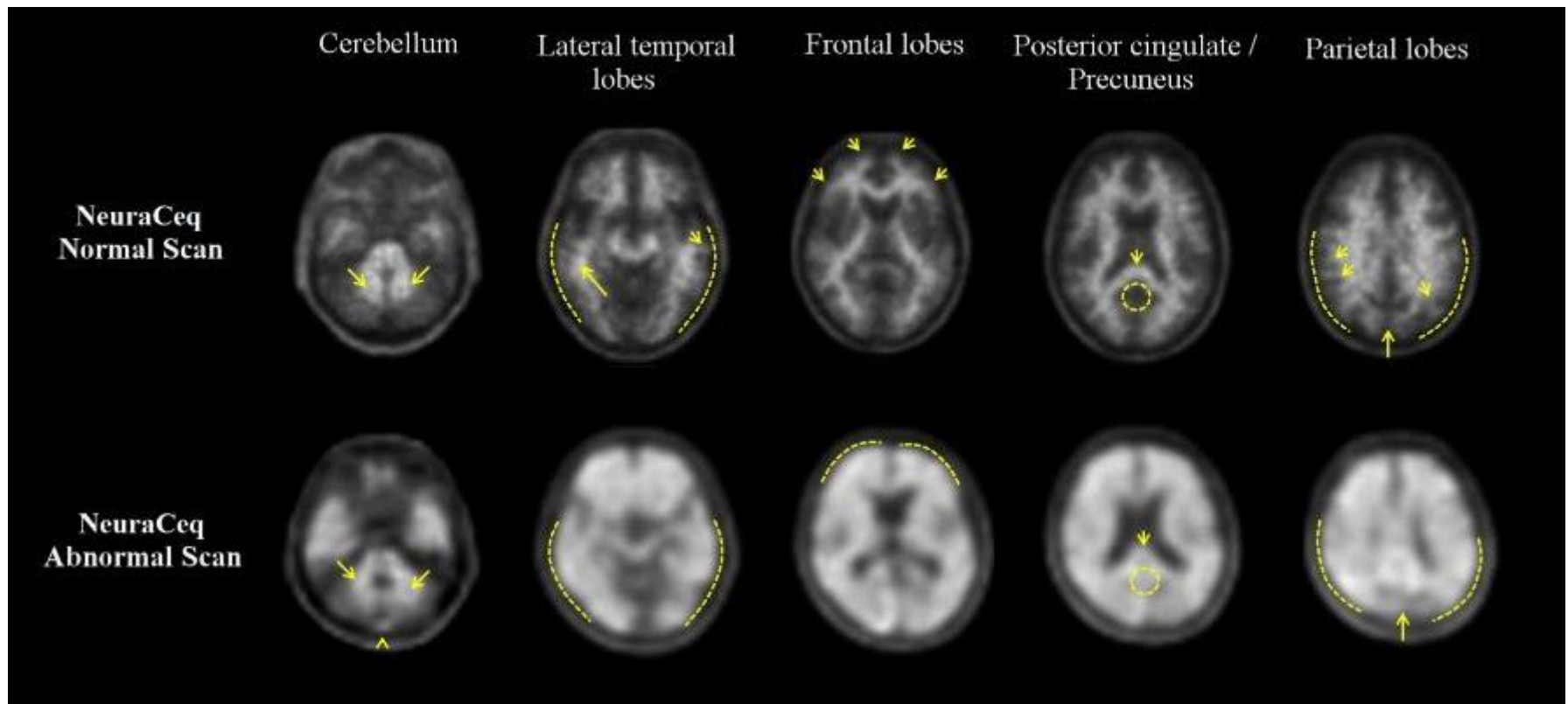


# Florbetaben uptake correlates with amyloid-plaque deposition



# Robust visual assessment method was validated for clinical practice

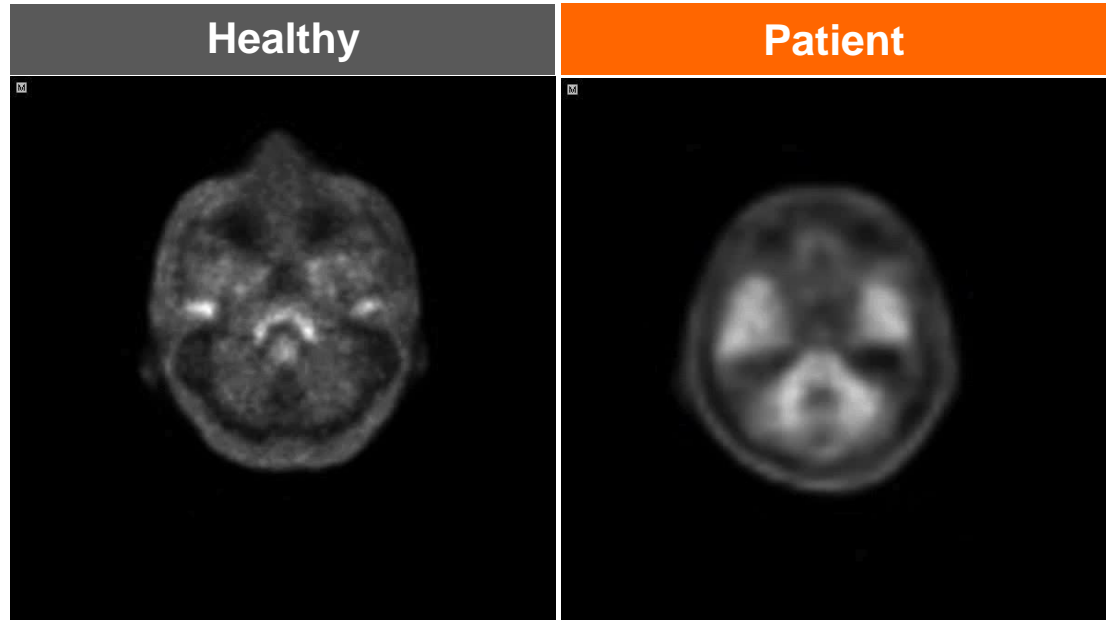
- Acquisition: for 20 min starting approx. 90 min p. i.
- Systematic visual Interpretation: gray scale, binary assessment



# Florbetaben detects amyloid plaques with high sensitivity and specificity

## Systematic visual analysis of the brain PET scans

- Scans from 46 of 47 subjects with beta-amyloid were correctly read as positive  
→ **Sensitivity: 98%**
- Scans from 24 of 27 subjects without beta-amyloid were correctly read as negative  
→ **Specificity: 89%**



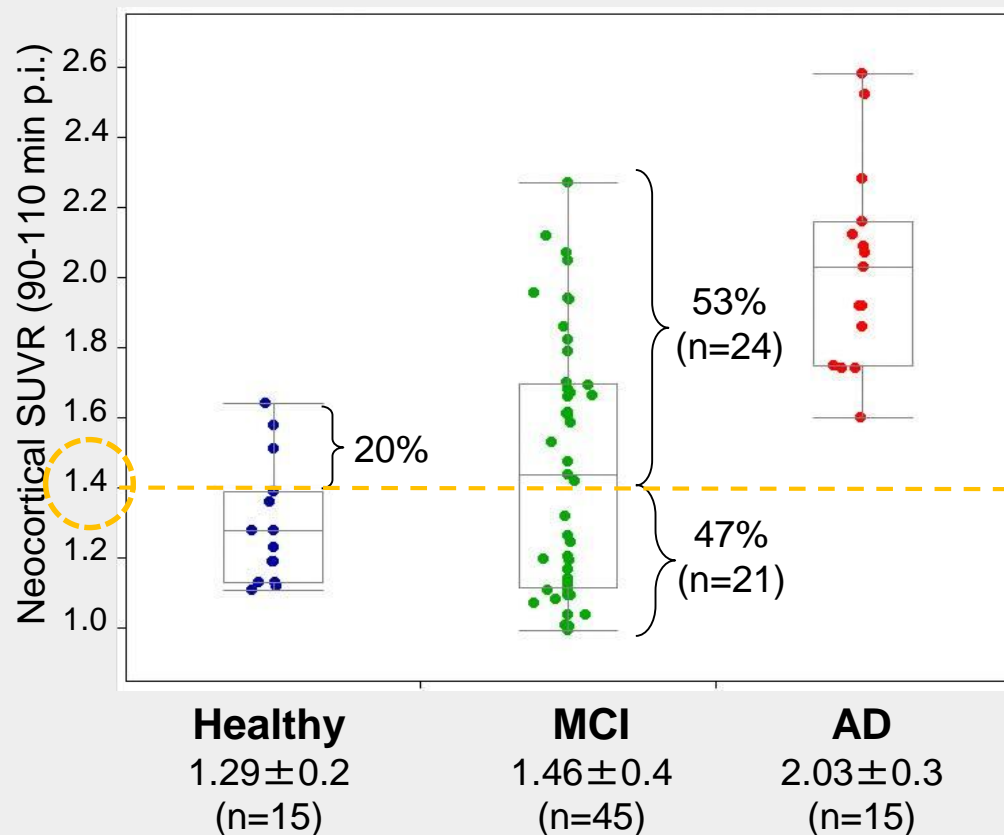
**Negative scans** allow physicians to consider alternative causes of cognitive impairment not associated with  $\beta$ -amyloid pathology

**Positive scans** along with other tests help determine if  $\beta$ -amyloid pathology is due to AD or some other cognitive disorder

# FBB MCI-Study 4 year follow-up data:

## No subject with negative scan converted to AD

### PET image analysis (baseline scan)



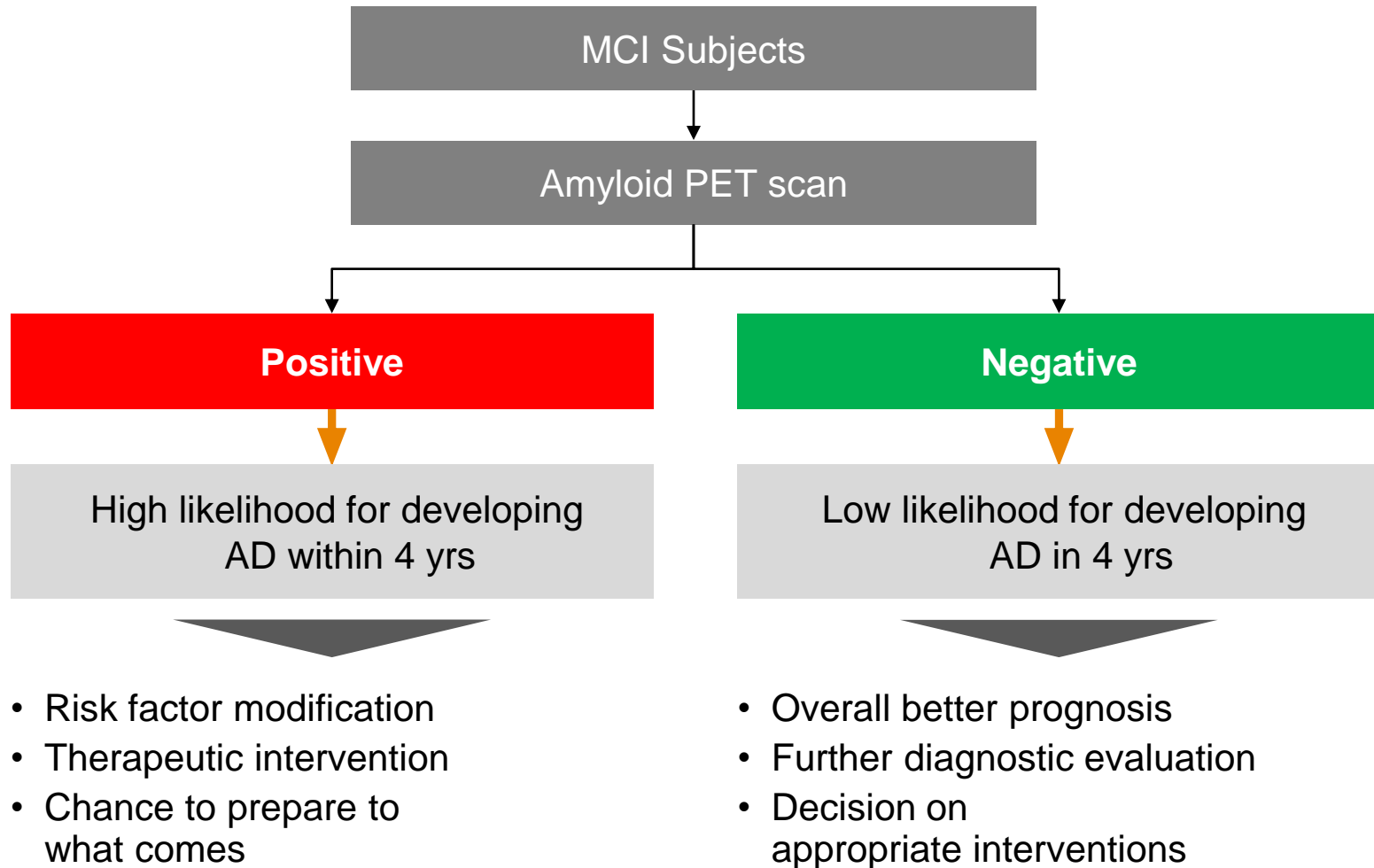
### MCI 4-year follow-up

- 88 % (21/24) of florbetaben positive MCI convert to clinical AD

**Predictive Accuracy: 94%**

- None (0%) of florbetaben negative MCI convert to AD
- 24 % (5/21) of florbetaben negative MCI convert to other forms of dementia

# Hints for high predictive accuracy of amyloid-PET in the FBB MCI-study



# Summary Neuroimaging

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1. Amyloid beta PET imaging increases the **diagnostic accuracy**, hence the **confidence** for patients and physicians
  - A negative scan in a symptomatic patient essentially rules out Alzheimer's disease and requires further diagnostic investigation
2. Amyloid beta PET may be important to predict **progression** / non-progression to AD in a population of MCI
3. Early and accurate detection of amyloid beta are a prerequisite to identify and develop **disease modifying drugs**

# Summary

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- Applied research selects candidates for indications with proven clinical relevance
- It does not replace basic research
- Molecular Imaging has great potential to personalize the therapy and improve the quality of life for patients with significant diseases
- Proof-of-mechanism studies allow for early selection of promising radiopharmaceuticals for development
- Further research and development for molecular tracers in neurodegenerative diseases, oncology and cardiovascular imaging is warranted

**Close collaboration of academia and industry is key**