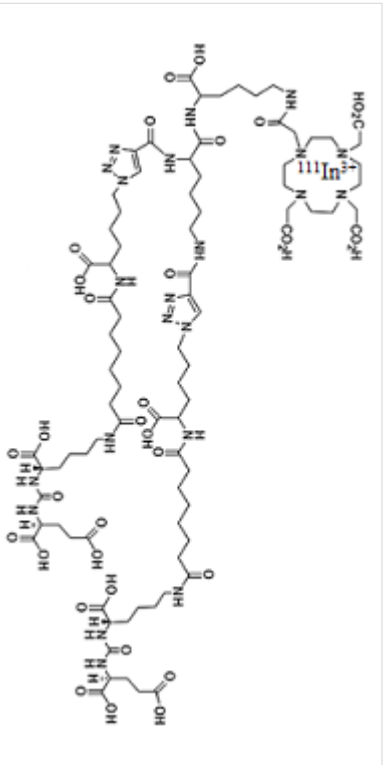
^{111}In-Labeled (7*S*)-26-(4-((1-((1-carboxy-5-(2-(4,7,10-tris(carboxymethyl)-1,4,7,10-tetraazacyclododecan-1-yl)acetamido)pentyl)amino)-1-oxo-6-(1-((7*S*)-1,3,7,22-tetracarboxy-5,13,20-trioxo-4,6,12,21-tetraazahexacosan-26-yl)-1*H*-1,2,3-triazole-4-carboxamido)hexan-2-yl)carbamoyl)-1*H*-1,2,3-triazol-5-yl)-5,13,20-trioxo-4,6,12,21-tetraazahexacosane-1,3,7,22-tetracarboxylic acid

[^{111}In]3

Liang Shan, PhD[1]

Created: October 31, 2012; Updated: December 19, 2012.

Chemical name:	^{111}In-Labeled (7S)-26-(4-((1-((1-carboxy-5-(2-(4,7,10-tris(carboxymethyl)-1,4,7,10-tetraazacyclododecan-1-yl)acetamido)pentyl)amino)-1-oxo-6-(1-((7*S*)-1,3,7,22-tetracarboxy-5,13,20-trioxo-4,6,12,21-tetraazahexacosan-26-yl)-1*H*-1,2,3-triazole-4-carboxamido)hexan-2-yl)carbamoyl)-1*H*-1,2,3-triazol-5-yl)-5,13,20-trioxo-4,6,12,21-tetraazahexacosane-1,3,7,22-tetracarboxylic acid
Abbreviated name:	[^{111}In]**3**
Synonym:	
Agent Category:	Compounds
Target:	Prostate-specific membrane antigen (PSMA)
Target Category:	Antigens
Method of detection:	Single-photon emission computed tomography (SPECT); gamma planar imaging
Source of signal / contrast:	^{111}In



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Activation:	No	
Studies:	• *In vitro* • Rodents	Structure of [^{111}In]**3** (1).

Background

[PubMed]

^{111}In-Labeled (7*S*)-26-(4-((1-((1-carboxy-5-(2-(4,7,10-tris(carboxymethyl)-1,4,7,10-tetraazacyclododecan-1-yl)acetamido)pentyl)amino)-1-oxo-6-(1-((7*S*)-1,3,7,22-tetracarboxy-5,13,20-trioxo-4,6,12,21-tetraazahexacosan-26-yl)-1*H*-1,2,3-triazole-4-carboxamido)hexan-2-yl)carbamoyl)-1*H*-1,2,3-triazol-5-yl)-5,13,20-trioxo-4,6,12,21-tetraazahexacosane-1,3,7,22-tetracarboxylic acid, abbreviated as [^{111}In]**3**, was synthesized by Banerjee et al. for use in single-photon emission computed tomography (SPECT) of tumors expressing prostate-specific membrane antigen (PSMA) (1).

PSMA is a type II transmembrane glycoprotein with three structural domains, including a 19-amino-acid intracellular domain, a 24-amino-acid transmembrane domain, and a large 707-amino-acid extracellular domain (2, 3). Two site-specific carboxypeptidase activities have been assigned to PSMA: *N*-acetylated α-linked acidic dipeptidase, which hydrolyzes the neuropeptide *N*-acetyl-aspartyl-glutamate (NAAG) in the brain to regulate release of neurotransmitters, and folate hydrolase activity, which is characterized by the cleavage of terminal glutamates from poly- and gamma-glutamated folates, which play a role in the cellular uptake of dietary folate (2). PSMA has been found to be expressed in the prostate at a level 1,000-fold greater than that in other tissues, and many folds higher in prostate cancer than in normal and benign prostate tissues (4, 5). High-grade and hormone-insensitive tumors have the greatest PSMA expression. PSMA is also consistently and abundantly expressed on the neovascular endothelium in a wide variety of human solid tumors, but not in blood vessels in normal tissues. These features of PSMA

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make it an optimal target for developing imaging and therapy strategies for prostate cancer (6).

Besides antibodies and antibody fragments, low molecular weight compounds have also been intensively tested as PSMA inhibitors and as PSMA-targeted imaging agents (6, 7). These compounds can be largely classified into two types: ureas and phosphoramidates (7). Both types of compounds possess a terminal glutamate at the P1' position, which enables productive binding with PSMA. They are also amenable to modification with bulky substituents that interact with the arginine patch or tunnel region on PSMA. Investigators have synthesized a series of ureas, including [^{18}F]DCFBC, [^{18}F]DCFPyL, and [^{111}In]**3** (1, 7, 8). [^{111}In]**3** is a bivalent compound that was synthesized by incorporating a PSMA-binding Lys-Glu urea motif for exploiting click chemistry and a second lysine residue for subsequent modification with an imaging or therapeutic moiety (1). Studies have shown that these radiolabeled PSMA inhibitors have desirable properties as imaging agents for prostate cancer imaging in animal models (1, 7, 8). This chapter summarizes the data obtained with [^{111}In]**3**. In another chapter, the data obtained with [^{18}F]DCFPyL are summarized. The data obtained with [^{18}F]DCFBC can be reviewed in the chapter on [^{18}F]DCFBC in MICAD.

Related Resource Links:

- PSMA-targeted imaging agents in MICAD
- Gene information in NCBI (PSMA)
- Articles in Online Mendelian Inheritance in Man (OMIM) (PSMA)
- PSMA-related clinical trials in ClinicalTrials.gov

Synthesis

[PubMed]

Banerjee et al. described the synthesis of precursor compounds in detail, including monovalent compound **1** ((3S,7S)-26-azido-5,13,20-trioxo-4,6,12,21-tetraazahexacosane-1,3,7,22-tetracarboxylic acid), bivalent compound **2** ((7S)-26-(4-((1-((5-amino-1-carboxypenty l)amino)-1-oxo-6-(1-((7S)-1,3,7,22-tetracarboxy-5,13,20-trioxo-4,6,12,21-tetraazahexacosan-26-yl)-1*H*-1,2,3-triazole-4-carboxamido)hexan-2-yl)carbamoyl)-1*H*-1,2,3-triazol-5-yl)-5,13,20-trioxo-4,6,12,21-tetraazahexacosane-1,3,7,22-tetracarboxylic acid), and DOTA-chelated bivalent urea compound **3** (1). Compound **3** (50–70 μg) was labeled with ^{111}In (111–148 MBq, 3–4 mCi) *via* incubation for 20 h at 95°C in 0.3 M NaOAc buffer. The radiochemical yield of [^{111}In]**3** was 70%–90% yield, and its specific radioactivity was 8.4–204.4 GBq/μmol (0.23–5.52 mCi/μmol). The radiochemical purity was >98%.

In Vitro Studies: Testing in Cells and Tissues

[PubMed]

The PSMA enzyme inhibition potency of compounds **1**–**3** was determined with a modified Amplex Red glutamic acid assay after incubation with the cell lysates of LNCaP cell extracts in the presence of NAAG for 2 h at 37°C (1). As shown by the enzyme inhibitory constant (K_i), the binding affinity was found to increase 5-fold from monovalent compound **1** to bivalent compound **2**. There was an 11-fold increase in the affinity for the DOTA-chelated bivalent compound **3** compared to compound **1**. Under the same experimental conditions in this study, the K_i value of ZJ-43 was reported to be 1.16 nM, indicating the high inhibitory potency of compound **3**. ZJ-43 is a urea-based potent inhibitor of NAAG and is used as an internal reference in the assay.

Table 1 PSMA inhibitory activity of compounds 1-3

Compound	Inhibitory constant	95 percent CI
1	0.91	0.58–1.45
2	0.10	0.07–0.16
3	0.08	0.05–0.12
ZJ-43	1.16	0.92–1.46

Inhibitory constant: nM

Animal Studies

Rodents

[PubMed]

Biodistribution of [^{111}In]**3** was determined at 2 h and 24 h after tail vein injection of 74 MBq (20 μCi) [^{111}In]**3** into nonobese diabetic severe-combined immunodeficient (NOD-SCID) mice that carried both PSMA-positive PC3-PIP and PSMA-negative PC3-flu xenografts (n = 4 mice/group) (1). The percentage of injected dose per gram tissue (% ID/g) was measured *ex vivo* in selected organs (Table 2). [^{111}In]**3** displayed PSMA-dependent binding in PC3-PIP xenografts, with continuous accumulation at the tumor site up to 24 h. The PC3-PIP tumor uptake values were 31.93 ± 5.87% ID/g and 34.03 ± 7.53% ID/g at 2 h and 24 h, respectively. The blood, spleen, gastrointestinal tract, kidney, and bladder displayed the highest uptake at 2 h. Steady clearance from the kidneys was observed, from 168.67 ± 14.12% ID/g at 2 h after injection to 66.86 ± 14.22%ID/g at 24 h after injection.

Table 2 Biodistribution data in selected organs in tumor-bearing mice

Organ	2 h	24 h
PC3-PIP to PC3-Luc ratio	203	379
Blood	0.12 ± 0.04	0.02 ± 0.01
Liver	0.19 ± 0.03	0.16 ± 0.03

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Kidney	168.67 ± 14.18	66.86 ±14.22
Bladder (empty)	2.61 ± 1.36	0.52 ± 0.27
PC3-PIP	31.93 ± 5.87	34.03 ± 7.53
PC3-Luc	0.16 ± 0.03	0.09 ± 0.03

SPECT was performed after tail vein injection of 44.4 MBq (1.2 mCi) [^{111}In]**3** to one NOD-SCID mouse that carried both PSMA-positive PC3-PIP and PSMA-negative PC3-flu xenografts (1). Radiochemical uptake was followed for up to 192 h after injection. A CT scan was performed before scintigraphy for both anatomical co-registration and attenuation correction. Intense radiotracer uptake was seen only in the PC3-PIP tumors and in the kidneys. Clearance of radioactivity from the kidneys and non-target tissues was more rapid than from the target tumor, such that by 48 h after injection a high tumor/background ratio was observed. The PC3-PIP tumor could still be imaged up to 8 d after injection.

Other Non-Primate Mammals

[PubMed]

No references are currently available.

Non-Human Primates

[PubMed]

No references are currently available.

Human Studies

[PubMed]

No references are currently available.

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