Cy7-(3*S*,7*S*)-26-Amino-5,13,20-trioxo-4,6,12,21-tetraazahexacosane-1,3,7,22-tetracarboxylic acid

Cy7-3

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Chemical name:	Cy7-(3*S*,7*S*)-26-Amino-5,13,20-trioxo-4,6,12,21-tetraazahexacosane-1,3,7,22-tetracarboxylic acid	
Abbreviated name:	Cy7-3	
Synonym:		
Agent category:	Compound	
Target:	Prostate-specific membrane antigen (PSMA), or *N*-acetyl α-linked acidic dipeptidase (NAALADase)	
Target category:	Antigen	
Method of detection:	Optical, near-infrared fluorescence (NIR) imaging	
Source of signal:	Cy7	
Activation:	No	
Studies:	• *In vitro* • Rodents	Click on protein, nucleotide (RefSeq), and gene for more information about PSMA.

Background

[PubMed]

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Optical fluorescence imaging is increasingly used to monitor biological functions of specific targets in small animals (1-4). However, the intrinsic fluorescence of biomolecules poses a problem when fluorophores that absorb visible light (350–650 nm) are used. Near-infrared (NIR) fluorescence (650–1,000 nm) detection avoids the natural background fluorescence interference of biomolecules, providing a high contrast between target and background tissues in small animals. NIR fluorophores have a wider dynamic range and minimal background fluorescence as a result of reduced scattering compared with visible fluorescence detection. NIR fluorophores also have high sensitivity, attributable to low background fluorescence, and high extinction coefficients, which provide high quantum yields. The NIR region is also compatible with solid-state optical components, such as diode lasers and silicon detectors. NIR fluorescence imaging is a noninvasive alternative to radionuclide imaging in small animals (4, 5).

Prostate-specific membrane antigen (PSMA) is a cell-surface glycoprotein with a molecular weight of ~100 kDa. It is a unique, type II, transmembrane-bound glycoprotein that is overexpressed on prostate tumor cells and in the neovasculature of most solid prostate tumors, but not in the vasculature of normal tissues (6, 7). PSMA has also been detected in other tissues such as the kidneys, the proximal small intestine, and the salivary glands (7). PSMA was found to have *N*-acetyl α-linked acidic dipeptidase (NAALADase) or glutamate carboxypeptidase II activity (8). PSMA may play an important role in the progression of prostate cancer and glutamatergic neurotransmission, as well as in the absorption of folate (9). In the central nervous system, PSMA metabolizes *N*-acetyl-aspartyl-glutamate, and in the proximal small intestine it removes γ-linked glutamates from poly-γ-glutamate folate and folate hydrolase (7). PSMA can be used as a marker for the detection of metastatic cancers with imaging agents. Although a commercially available monoclonal antibody (^{111}In-labeled Capromomab pendetide (^{111}In-CYT-356)) is in clinical use for the detection of prostate cancer, the results obtained with this antibody are not entirely reliable (10). In addition, this IgG antibody has limited access to tumors and also may produce low signal/noise ratios because the target is the intracellular domain of PSMA (11, 12). IRDye800CW-(3*S*,7*S*)-26-Amino-5,13,20-trioxo-4,6,12,21-tetraazahexacosane-1,3,7,22-tetracarboxylic acid (YC-27, also known as 800CW-3) is a small molecule inhibitor of NAALADase (13). YC-27 was able to visualize PSMA-positive tumors in nude mice, but accumulation of signal was high in the kidney. Chen et al. (14) evaluated a series of dye-3 conjugates with Cy5.5, IGC, IRDye800RS, and Cy7 in nude mice bearing human PSMA-positive xenografts. Injected Cy7-3 exhibited good tumor accumulation with lower liver and kidney accumulation than YC-27.

Related Resource Links:

- Chapters in MICAD (PSMA)
- Gene information in NCBI (PSMA)
- Articles in OMIM Articles in Online Mendelian Inheritance in Man (OMIM) (PSMA)
- Clinical trials (PSMA)

Synthesis

[PubMed]

N,*N*-Diisopropylethylamine (0.0115 mM) was added to a solution of compound 3 (710 nmol) in dimethyl sulfoxide, followed by *N*-hydroxysuccininide ester of Cy7 (610 nM) (14). The mixture was incubated at room temperature for 2 h. Cy7-3 was purified with high-performance liquid chromatography with 64% yield. The molecular weight of Cy7-3 (1.27 kDa) with one Cy7 per targeting molecule was confirmed with mass spectroscopy. Cy7-3 exhibited an absorbance maximum at 743 nm and an emission maximum at 767 nm.

In Vitro Studies: Testing in Cells and Tissues

[PubMed]

The binding affinity of Cy7-3 for PSMA was determined with the use of a NAALADase assay (14). Cy7-3 exhibited an inhibition constant (K_i) value of 5.0 ± 0.2 nM. The K_i value of 800CW-3 was 20 ± 5 nM.

Animal Studies

Rodents

[PubMed]

Chen et al. (14) performed NIR fluorescence imaging studies of Cy7-3 or YC-27 (1 nmol) in nude mice (n = 3) bearing PSMA-positive PC3-PIP tumor xenografts and PSMA-negative PC3-flu tumor xenografts. The PC3-PIP tumors were clearly visualized at 24 h after injection of the tracers. On the other hand, the NIR signal from the PC3-flu tumors was weak. Kidney accumulation of fluorescence was more prominent for YC-27 than Cy7-3. The tumor/kidney ratio for Cy7-3 was higher than that for YC-27. *Ex vivo* NIR imaging showed that the PC3-PIP tumors exhibited higher fluorescence intensity than the kidneys, liver, spleen, intestines, and PC3-flu tumors for Cy7-3. Co-injection of 1 nmol Cy7-3 with 1,000 nmol DClBzL (a PSMA inhibitor) in mice bearing the two tumors inhibited the NIR fluorescence in the PC3-PIP tumors nearly to the background level of the PC3-flu tumors at 24 h after injection. The *ex vivo* NIR signals in the tumors were consistent with the *in vivo* imaging.

Other Non-Primate Mammals

[PubMed]

No publication is currently available.

Non-Human Primates

[PubMed]

No publication is currently available.

Human Studies

[PubMed]

No publication is currently available.

NIH Support

R01 CA134675

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