D-Cys-D-Asp-Gly-HCit-Gly-Pro-Gln-D-Cys-Ebes-Ebes-Lys-Cy5.5

OA02-Cy5.5

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Created: September 6, 2007; Updated: October 18, 2007.

Chemical name:	D-Cys-D-Asp-Gly-HCit-Gly-Pro-Gln-D-Cys-Ebes-Ebes-Lys-Cy5.5	
Abbreviated name:	OA02-Cy5.5	
Synonym:		
Agent Category:	Peptide	
Target:	Integrin $\alpha_3\beta_1$	
Target Category:	Receptor binding	
Method of detection:	Optical, Near-Infrared	
Source of signal:	Cy5.5	
Activation:	No	
Studies:	• *In vitro* • Rodents	Click on protein, nucleotide (RefSeq), and gene for more information about integrin $\alpha_3\beta_1$.

Background

[PubMed]

Optical fluorescence imaging is increasingly used to obtain biological functions of specific targets (1, 2). However, the intrinsic fluorescence of biomolecules poses a problem when fluorophores that absorb visible light (350–700 nm) are used. Near-infrared (NIR) fluorescence (700–1,000 nm) detection avoids the background fluorescence interference of natural biomolecules, providing a high contrast between target and background tissues. NIR fluorophores have a wider dynamic range and minimal background interference as a result of reduced scattering compared with visible fluorescence detection. They also have high sensitivity as a result of low infrared background interference, and high extinction

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NLM Citation: Leung K. D-Cys-D-Asp-Gly-HCit-Gly-Pro-Gln-D-Cys-Ebes-Ebes-Lys-Cy5.5 . 2007 Sep 6 [Updated 2007 Oct 18]. In: Molecular Imaging and Contrast Agent Database (MICAD) [Internet]. Bethesda (MD): National Center for Biotechnology Information (US); 2004-2013.

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 becoming a non-invasive alternative to radionuclide imaging.

Integrins are a family of cell-surface heterodimeric glycoproteins that mediate diverse biological events involving cell–cell and cell–matrix interactions (3). They consist of an α and a β subunit. They are important for cell adhesion and signal transduction. The $\alpha_3\beta_1$ integrin plays an important role in normal lung, kidney, cerebral cortical, and epithelial development (4). On the other hand, it affects tumor growth, tumor invasiveness, and metastasis as the $\alpha_3\beta_1$ integrin is strongly expressed on tumor cells (5, 6). D-Cys-D-Asp-Gly-HCit-Gly-Pro-Gln-D-Cys (OA02) was identified to bind to the α_3 integrin on human ovarian cancer cells using one-bead-one-compound combinatorial libraries (7, 8). OA02 was conjugated with Cy5.5 to study *in vivo* biodistribution of the tracer in tumor-bearing mice (9). Cy5.5 is a NIR fluorescent dye with an absorbance maximum at 675 nm and emission maximum at 694 nm with a high extinction coefficient of 250,000 $(mol/L)^{-1}cm^{-1}$. OA02-Cy5.5 was found to have a high specific accumulation in $\alpha_3\beta_1$-positve ES-2 human ovarian tumor cells in nude mice.

Synthesis

[PubMed]

Cy5.5 monofunctional *N*-hydroxysuccinimide (NHS) ester was used to conjugate D-Cys-D-Asp-Gly-HCit-Gly-Pro-Gln-D-Cys-Ebes-Ebes-Lys using solid-phase synthesis to form OA02-Cy5.5 (9). The NHS ester of Cy5.5 reacted with the ε-amino group of the lysine. The peak containing the OA02-Cy5.5 conjugate was analyzed by matrix-assisted laser desorption/ionization-time-of-flight mass spectrometry. The measured mass was *m/z* 2,457.2, which was ~1 Cy5.5/OA02. The chemical purity was >90%.

In Vitro Studies: Testing in Cells and Tissues

[PubMed]

Receptor-mediated endocytosis of OA02-biotin in SKOV-3 human ovarian tumor cells (α_3-positive) was observed by fluorescent microscopy (9). The staining was confined to the cell membrane and cytoplasm. On the other hand, Raji B-cell lymphomas (α_3-negative) did not show any staining.

Animal Studies

Rodents

[PubMed]

Biodistribution studies of OA02-Cy5.5 were evaluated in nude mice bearing an ES-2 subcutaneous xenograft in the left flank and a Raji subcutaneous xenograft in the right

flank. Images were obtained after injection of 20 μg (8.14 nmol) OA02-Cy5.5 (9). The background fluorescent intensity was 225 ± 15 arbitrary units (AU) for both tumors. The ES-2 tumor uptake of OA02-Cy5.5 was 6,032 ± 4,640 AU at 15 min and 5,526 ± 3,696 AU at 70 min, whereas the Raji tumor uptake was 2,685 ± 1,103 AU at 15 min and 2,786 ± 1,583 AU at 70 min. There was a gradual clearance of the signal from the system *via* the kidneys. The fluorescent intensity decreased to ~2,000 AU at 1,440 min for both tumors. The tracer uptake in both tumors could be reduced to background level (~700 AU) at 70 min by pre-administration of anti-α_3 monoclonal antibody 30 min before OA02-Cy5.5 injection. *Ex vivo* imaging showed that most of the fluorescent signal intensity was from the ES-2 tumor, urinary bladder, and kidneys.

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

R33 CA89706, 5-T32-RR07038

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