NLM Citation: Leung K. DiD-Labeled anti-EpCAM-directed NK-92-scFv(MOC31) zeta cells. 2009 Aug 28 [Updated 2009 Sep 30]. In: Molecular Imaging and Contrast Agent Database (MICAD) [Internet]. Bethesda (MD): National Center for Biotechnology Information (US); 2004-2013.
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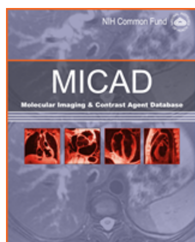

DiD-Labeled anti-EpCAM-directed NK-92-scFv(MOC31) zeta cells

DiD-NK-92-scFv(MOC31) zeta cells

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Created: August 28, 2009; Updated: September 30, 2009.

Chemical name:	DiD-Labeled anti-EpCAM-directed NK-92-scFv(MOC31) zeta cells	
Abbreviated name:	DiD-NK-92-scFv(MOC31) zeta cells	
Synonym:		
Agent category:	Antibody scFv fragment	
Target:	Epithelial cell adhesion molecule (EpCAM)	
Target category:	Adhesion molecule	
Method of detection:	Optical, near-infrared (NIR) fluorescence	
Source of signal/contrast:	1,1'-Dioctadecyl-3,3,3',3'-tetramethylindodicarbocyanine (DiD)	
Activation:	No	
Studies:	• *In vitro* • Rodents	Click on protein, nucleotide (RefSeq), and gene for more information about EpCAM.

Background

[PubMed]

Optical fluorescence imaging is increasingly being used to monitor biological functions of specific targets in small animals (1-3). 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 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 non-invasive alternative to radionuclide imaging in small animals (4, 5).

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Natural killer (NK) cells exert a high selective cytotoxicity against tumor cells without cytotoxicity against normal cells (6). Adoptive cancer immunotherapy has been performed with systemic infusion of *ex vivo* activated autologous or donor NK cells (7-9). Cellular immunotherapy with NK-92, a clonal NK cell line, has been initiated in a phase I clinical trial against acute myeloid leukemia. The epithelial cell adhesion molecule (EpCAM) is found on the cell surface of epithelial cells of many epithelial tissues such as in the pancreas, jejunum, colon, kidney, salivary gland, and prostate (10). EpCAM is responsible for intracellular signaling and polarity, as well as mediation of cell differentiation, proliferation, migration, and adhesion (11, 12). EpCAM is a pan-epithelial differentiation antigen that is expressed on almost all carcinomas (13). Approximately 70% of human prostate cancers show high levels of EpCAM expression (14, 15). Tavri et al. (16) have generated NK-92 cells transduced with a humanized single-chain Fv antibody fragment (scFv) of EpCAM antibody MOC31 fused with CD3 zeta chain and tagged with 1,1'-dioctadecyl-3,3,3',3'-tetramethylindodicarbocyanine (DiD) (DiD-NK-92-scFv(MOC31) zeta cells) for noninvasive NIR imaging of EpCAM expression in prostate tumor cells in rats. DiD is a NIR fluorescence dye with an absorbance maximum at 644 nm and an emission maximum at 665 nm.

Synthesis

[PubMed]

Tavri et al. (16) reported the labeling of NK-92-scFv(MOC31) zeta cells and NK-92 parental cells with DiD. The NK cells were incubated with 5 μg/ml DiD for 15 min and then washed three times with phosphate-buffered saline. The NK-92 parental cells showed one-fold higher labeling efficiency than the NK-92-scFv(MOC31) zeta cells. The labeling procedure had no effect on the NK cell viability.

In Vitro Studies: Testing in Cells and Tissues

[PubMed]

NK-92-scFv(MOC31) zeta cells (not labeled with DiD) lysed EpCAM-expressing DU145 prostate cells, whereas the parental NK-92 showed no cell lysis (16). MOC31 monoclonal antibody blocked cytotoxicity by >90%. Fluorescence microscopy analysis showed that the dye was integrated into the cell membrane of labeled NK cells. The dye remained in the NK cells for up to 24 h after labeling.

Animal Studies

Rodents

[PubMed]

Tavri et al. (16) performed whole-body NIR fluorescence imaging of rats (n = 6/group) bearing DU145 tumors after intravenous injection of 1.5×10^7 DiD-NK-92-scFv(MOC31) zeta cells or 1.5×10^7 DiD-NK-92 cells. There was an increase in fluorescence signal in the tumors at 1.5 and 8 h after injection of DiD-NK-92-scFv(MOC31) zeta cells, followed by a leveling off at 24 h. The fluorescence units were <1, 8, 12, and 11 at 0, 1.5, 8, and 24 h after injection, respectively. On the other hand, DiD-NK-92 parental cells showed <1 at all time points. *Ex vivo* NIR fluorescence imaging showed the tumor/liver, tumor/lung, tumor/spleen, and tumor/sternum ratios of 3, 5, 6, and 15 at 24 h after injection of DiD-NK-92-scFv(MOC31) zeta cells, respectively. In contrast, DiD-NK-92 parental cells exhibited tumor/nontargeted tissue ratios of <1. No blocking experiment was performed.

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.

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