Fusion Protein For Anti-Cd19 Chimeric Antigen Receptor Detection

Tech ID: 25876 / UC Case 2010-810-0

SUMMARY

Researchers at UCLA have developed a fusion protein that can detect immune cells expressing anti-CD19 chimeric antigen receptors with higher specificity and lower background than existing antibodies.

BACKGROUND

CD19, a glycoprotein specific to the B-cell lineage, is a target of a number of cancer immunotherapies. Clinical trials testing the effectiveness of T cells equipped with anti-CD19 chimeric antigen receptors (CARs) in B-cell malignancy treatments are currently underway. Improving the detection of T cells that carry anti-CD19 CARs would greatly improve the characterization of the cells used for these immunotherapies and increase therapeutic potential.

The detection of CD19 CAR expression by FACS is typically determined through the use of an antibody to either the hinge region or FC portion of the receptor. Detection using commercially available antibodies can result in high levels of background and non-specific binding, especially in blood samples where many IgG expressing cells are present.

INNOVATION

Dr. Donald Kohn and colleagues have developed a CD19 fusion protein that can be used to detect anti-CD19 CAR+ cells. This fusion protein can be secreted from producer cells (e.g. 293T) and purified from the cell culture supernatant. The purified protein molecule is conjugated with a fluorescent fluorophore and used directly to detect anti-CD19 CAR+ cells in fluorescence-based techniques, such as flow cytometry (also known as fluorescent activated cell sorting, or FACS) or confocal imaging. This fusion protein contains specific domains of CD19 that should allow the detection of all anti-CD19 CAR+ cells irrespective of the antibody binding domain used to construct the CAR. Immune cells with the anti-CD19 CAR will recognize and specifically bind the CD19 fusion protein, positively labelling the cells. The CD19 domain incorporated into the fusion protein binds anti-CD19 CAR in a highly specific manner and enhances the detection of anti-CD19 CAR+ cells.

APPLICATIONS

- Detection of anti-CD19 CAR+ cells using a fluorescence-based technique, such as FACS or confocal microscopy
- Quantitative measurement of blood cell phenotype and frequency

ADVANTAGES

- Increase specificity compared to available antibodies to anti-CD19 CAR
- Decrease non-specific binding compared to available anti-CD19 CAR detection antibodies
- Improve cell characterization for immunotherapies
- Increase therapeutic potential of anti-CD19 CAR cell therapies

STATE OF DEVELOPMENT

- The fusion protein has been constructed, expressed, purified and conjugated.
- The conjugated fusion protein has been shown to detect anti-CD19 CAR expression in T-cells, including immortalized cell lines (Jurkat) as well as in primary cells and in blood samples from mice engrafted with human cells.

RELATED MATERIALS

CONTACT

UCLA Technology Development Group
ncd@tdg.ucla.edu
tel: 310.794.0558.

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INVENTORS

- Kohn, Donald B.

OTHER INFORMATION

KEYWORDS

fusion protein, antibody, CD19, CAR, flow cytometry, confocal imaging, microscopy, cancer, immunotherapy, oncology, T cell, B cell

CATEGORIZED AS

- **Imaging**
  - Molecular
- **Medical**
  - Disease: Blood and Lymphatic System
  - Disease: Cancer
  - Gene Therapy
  - Research Tools
- **Research Tools**
  - Antibodies

RELATED CASES

2010-810-0

▶ De Oliveira SN … Kohn DB, Hollis RP. A CD19/Fc fusion protein for detection of anti-CD19 chimeric antigen receptors. J Transl Med. 2013;11:23.

ADDITIONAL TECHNOLOGIES BY THESE INVENTORS

- ▶ Gene Editing of Monogenic Disorders in Human Hematopoietic Stem Cells
- ▶ Augmentations to Lentiviral Vectors to Increase Expression
- ▶ Improvement To Retroviral Vectors Containing The Human Ubiquitin C Promoter
- ▶ Generation Of Minimal Enhancer Elements Using Massively Parallel Reporter Assays
- ▶ Optimized Lentiviral Vector for Stem Cell Gene Therapy of Hemoglobinopathies
- ▶ Transient Expression Of BCL-2 To Ameliorate Cytotoxicity Of Gene Modification Reagents In Stem Cells

UCLA Technology Development Group
10889 Wilshire Blvd., Suite 920,Los Angeles,CA 90095
https://tdg.ucla.edu
Tel: 310.794.0558 | Fax: 310.794.0638 | ncd@tdg.ucla.edu

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