PRODUCT INFORMATION

Contents: 10 μg Anti-hCD19-CD3, purified antibody, provided azide-free and lyophilized

Target: Human CD19 (hCD19) and human CD3 (hCD3)

Specificity: Cells expressing hCD19 and hCD3

Clonality: Monoclonal antibody

Source: CHO (Chinese hamster ovary) cells

Formulation: Anti-hCD19-CD3 is lyophilized from a 0.2 μm filtered phosphate buffer solution (pH 7.4) containing 5% saccharose.

Purity: > 90%. Purified by affinity chromatography

Antibody resuspension
Add 100 µl of sterile water to obtain a concentration of 0.1 mg/ml. Invert vial several times to ensure the product is fully dissolved.

Storage and stability
- Product is shipped at room temperature. Store lyophilized antibody at -20 °C. Lyophilized product is stable for at least 1 year.
- Reconstituted antibody is stable for 1 month when stored at 4 °C and for 1 year when aliquoted and stored at -20 °C. Avoid repeated freeze-thaw cycles.

Quality control
- Binding to hCD19 and to hCD3 has been confirmed by flow cytometry.
- Biological activity has been confirmed using cellular assays.
- The complete sequence of this antibody has been verified.
- The absence of bacterial contamination (e.g. lipoproteins and endotoxins) has been confirmed using HEK-Blue™ TLR2 and HEK-Blue™ TLR4 cells.

DESCRIPTION

Anti-hCD19-CD3 is a bispecific antibody that binds to two sites: hCD19 expressed on the surface of B cells, and hCD3, part of the T cell receptor. It features Blinatumomab single-chain variable fragments (scFv) joined by a glycine-serine linker. These two scFvs have been cloned from the anti-hCD19 (clone HD37) and anti-hCD3 (clone L2K-07) monoclonal antibodies. Blinatumomab is a bispecific antibody used for the treatment of refractory acute lymphocytic leukemia (ALL). By binding to hCD3 and hCD19, Blinatumomab engages unstimulated T cells to proliferate and exert cytotoxic activity on CD19-positive lymphoma cells (Figure 1). Of note, Blinatumomab does not cross-react with CD3 and CD19 from mice, rats, or dogs.

APPLICATION

Anti-hCD19-CD3 bispecific antibody can be used for fine-tuning studies of B cell contact-dependent killing and T cell activation/proliferation.

PROCEDURE

InvivoGen has developed a cellular assay to determine the ability of Anti-hCD19-CD3 to activate T cells in the presence of CD19-positive B cells. This assay utilizes the human B-cell lymphoma cell line Raji and InvivoGen’s Jurkat-Lucia™ NFAT cells, an immortalized T lymphocyte cell line that stably expresses an NFAT-inducible Lucia luciferase reporter gene (Figure 2, next page).

For more information visit http://www.invivogen.com/jurkat-lucia-nfat-cells.

Determination of T cell activation using Jurkat-Lucia™ NFAT cells
1. Dispense 20 µl of Anti-hCD19-CD3 (0.1-100 ng/ml final concentration) diluted in IMDM (Isocove’s Modified Dulbecco’s Medium) containing 10% heat-inactivated fetal bovine serum per well of a 96-well plate.
2. Into each well, distribute 90 µl of Raji cell suspension (100,000 cells/well).
3. Incubate 30 minutes at 37°C.
4. Into each well, distribute 90 µl of Jurkat-Lucia™ NFAT cell suspension (300,000 cells/well).
5. Incubate for 6, 8 and 24 hours at 37°C.
6. Levels of Lucia luciferase can be determined by measuring the luminescence at each time point using coelenterazine-based reagents such as QUANTI-Luc™ and QUANTI-Luc™ Gold.

FIGURE 1: Anti-hCD19-CD3 binds to hCD3 on T cells and to hCD19 on B cells leading to T cell proliferation and B cell lysis.

TECHNICAL SUPPORT
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https://www.invivogen.com/anti-hcd19-cd3
Figure 2: Jurkat-Lucia™ NFAT cell activation upon incubation with Raji B cells and Anti-hCD19-CD3.

*Note:* Jurkat T cells are CD4⁺CD8⁻. To assess B-cell lysis, we recommend to use primary CD8⁺ T cells.

**RESTRICTION USE**
This antibody is distributed for research purposes only. It is not intended for diagnosis or therapeutic use.

**RELATED PRODUCTS**

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<thead>
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<th>Product</th>
<th>Catalog Code</th>
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<tr>
<td>Anti-βGal-hCD3 (negative control)</td>
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<tr>
<td>Anti-hCD19-βGal (negative control)</td>
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<td>Jurkat-Lucia™ NFAT Cells</td>
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