More Tools for Speedy Antibody-Drug Conjugate Discovery

αHFc-CL-DX8951

Anti-human IgG Fc-DX8951 Antibody with Cleavable Linker

Catalog Number **AH-107-DX**

DESCRIPTION

αHFc-CL-DX8951 is anti-human IgG Fc specific antibody conjugated to exatecan mesylate (DX8951) with a cleavable linker. The antibody portion is a polyclonal antibody which is specific to the Fc region of human IgGs. Exatecan mesylate (DX8951) is a topoisomerase I inhibitor. The cleavable linker (GGFG) connecting DX8951 to the antibody is stable in extracellular fluid, but is cleaved by in endosome once the conjugate has entered a cell via endocytosis.

APPLICATIONS

Antibody-drug conjugates (ADCs), which have become a new targeted therapy against cancer, consist of an antibody linked to a cytotoxic drug. The ADCs bind selectively to the target cancer cells via the monoclonal antibody portion. Internalization of the ADCs releases the drug to do its damage. Prior to testing the function of ADCs in cell-based assays, each monoclonal antibody is typically conjugated directly with a cytotoxic drug. This step is time consuming and expensive, requiring milligram quantities of purified antibody, separate conjugation of each antibody, and further isolation of the ADC from the unconjugated drug. Using secondary antibody-drug conjugates (2°ADC) in a cell-based cytotoxic assay is a quick and economical alternative to pre-screening monoclonal antibodies as ADC candidates against tumor cells. Instead of conjugating the monoclonal antibody with a cytotoxic drug, the naked monoclonal antibody is added directly to the cells in the presence of the 2°ADC. Internalization of the monoclonal antibody/2°ADC complex can achieve a similar effect of targeted drug release within the cells as the monoclonal antibody-drug conjugate. Furthermore, the 2°ADC can also be applied to screen protein ligands for receptor-mediated cell targeting.

αHFc-CL-DX8951 is a 2°ADC for pre-screening antibodies with a human IgG Fc moiety or recombinant human IgG Fc fusion proteins to determine their cytotoxicity as exatecan mesylate (DX8951) bioconjugates. **αHFc-CL-DX8951** alone displays no obvious toxicity against multiple cell lines at 1.5 μg/ml (10 nM) or lower concentration. When applied in combination with tumor specific monoclonal antibodies or recombinant Fc fusion proteins, **αHFc-CL-DX8951** can help determine the cytotoxic potential for these antibodies or proteins against target cell lines.

EXAMPLE DATA

It has been demonstrated that Herceptin antibody-drug conjugate (T-DM1) displayed potent killing activity against Her2-overexpressing tumor cells but not normal Her2 expression or Her2 negative cells [1]. Here cytotoxicity of naked Herceptin was tested in four breast cancer tumor cell lines expressing different amount of Her2 marker in the presence and absence of **αHFc-CL-DX8951**. SKBR3 and HCC1954 are Her2 overexpressing cells, MCF7 has normal Her2 expression, and MDA-MB468 is Her2 negative. *In vitro* SKBR3 is slightly sensitive to unconjugated Herceptin treatment, while HCC1954, MCF7, and MDA-MB468 are resistant to unconjugated Herceptin [1]. The unconjugated anti-human IgG Fc antibody (αHFc) does not change the apparent effect of Herceptin against these cell lines (Fig A). In contrast, in the presence of 6.6 nM **αHFc-CL-DX8951**, Herceptin displays potent cytotoxicity against both Her2-overexpressing SKBR3 and HCC1954 cells, some killing of MCF7, and shows no killing of MDA-MB468 cells (Fig B).

Fig A. Cytotoxic Profile of Herceptin in the Presence of Unconjugated αHFc Secondary IgG

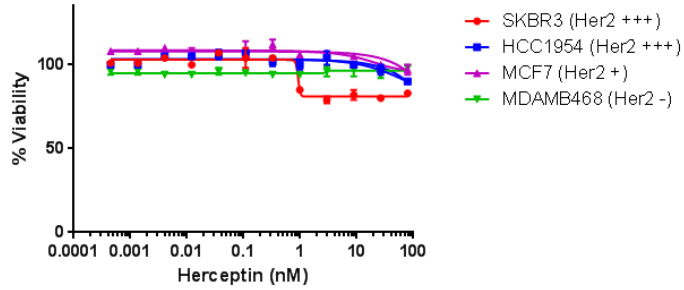
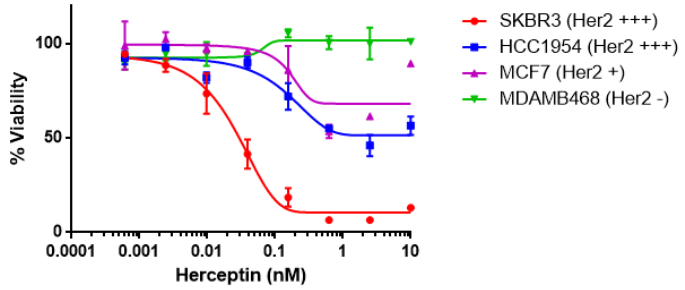


Fig B. Cytotoxic Profile of Herceptin in the Presence of 6.6 nM αHFc-CL-DX8951



STORAGE

Store in -20°C or -70°C manual defrosted freezer. Avoid repeated freeze-thaw cycle.

REFERENCE

1. Lewis Phillips GD, et al. Targeting Her2-positive breast cancer with trastuzumab-DM1, an antibody-cytotoxic drug conjugate. (2008), Cancer Res, **68(22)**, page 9280-90**.**