Human CD30/TNFRSF8 Antibody

Monoclonal Mouse IgG_1 Clone # 1115851

Catalog Number: MAB11768

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human recombinant CD30 protein in Direct ELISA.
Source	Monoclonal Mouse IgG_1 Clone # 1115851
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	Mouse myeloma cell line, NS0-derived human CD30/TNFRSF8 Phe19-Lys379 Accession # P28908
Formulation	Lyophilized from a 0.2 μm filtered solution in PBS with Trehalose.

APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Western Blot	2 µg/mL	HDLM-2 human Hodgkin's lymphoma cell line
Multiplex Immunofluorescence	25 µg/mL	Immersion fixed paraffin-embedded sections of Human Hodgkin's Lymphoma
Immunohistochemistry	3-25 µg/mL	Immersion fixed paraffin-embedded sections of human hodgkin's lymphoma

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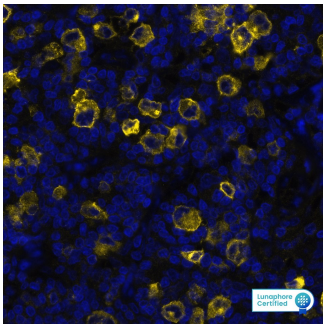
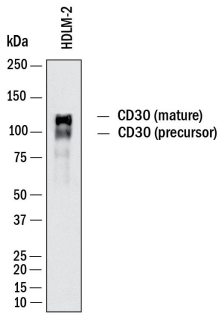
DATA

Multiplex Immunofluorescence



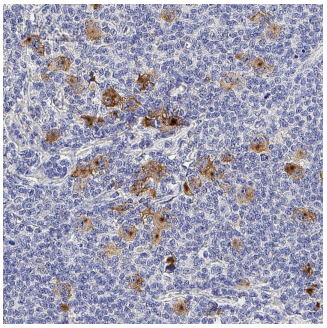
CD30/TNFRSF8 in Human Hodgkin's Lymphoma via seqIF™ staining on COMET™ CD30/TNFRSF8 was detected in immersion fixed paraffin-embedded sections of human Hodgkin's Lymphoma using Mouse Anti-Human CD30/TNFRSF8, Monoclonal Antibody (Catalog# MAB11768) at 25ug/mL at 37° Celsius for 16 minutes. Before incubation with the primary antibody, tissue underwent an all-in-one dewaxing and antigen retrieval preprocessing using PreTreatment Module (PT Module) and Dewax and HIER Buffer H (pH 9; Epredia Catalog #TA-999-DHBH). Tissue was stained using the Alexa Fluor™ 647 Goat anti-Mouse IgG Secondary Antibody at 1:200 at 37 ° Celsius for 2 minutes. (Yellow; Lunaphore Catalog # DR647MS) and counterstained with DAPI (blue; Lunaphore Catalog # DR100). Specific staining was localized to the membrane and cytoplasm. Protocol available in COMET™ Panel Builder.

Western Blot



Detection of Human CD30/TNFRSF8 by Western Blot. Western Blot shows lysates of HDLM-2 human Hodgkin's lymphoma cell line. PVDF membrane was probed with 2 µg/ml of Mouse Anti-Human CD30/TNFRSF8 Monoclonal Antibody (Catalog # MAB11768) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # Catalog # HAF018). Specific bands were detected for CD30/TNFRSF8 at approximately 90kDa and 120 kDa (as indicated). This experiment was conducted under reducing conditions and using Western Blot Buffer Group 1.

Immunohistochemistry



Detection of CD30/TNFRSF8 in Human Hodgkin's Lymphoma. CD30/TNFRSF8 was detected in immersion fixed paraffin-embedded sections of human hodgkin's lymphoma using Mouse Anti-Human CD30/TNFRSF8 Monoclonal Antibody (Catalog # MAB11768) at 5 µg/ml overnight at 4 °C. Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using VisUCyte Antigen Retrieval Reagent-Basic (Catalog # Catalog # VCTS021). Tissue was stained using the HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # Catalog # HAF007) and counterstained with hematoxylin (blue). Specific staining was localized to the cell membrane of Reed Sternberg cells. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

PREPARATION AND STORAGE

Reconstitution	Reconstitute lyophilized material at 0.2 mg/ml in sterile PBS. For liquid material, refer to CoA for concentration.
Shipping	Lyophilized product is shipped at ambient temperature. Liquid small pack size (-SP) is shipped with polar packs. Upon receipt, store immediately at the temperature recommended below.
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution.

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BACKGROUND

CD30, also known as Ki-1 antigen and TNFRSF8, is a 120 kDa type I transmembrane glycoprotein belonging to the TNF receptor superfamily (1, 2). Mature human CD30 consists of a 361 amino acid (aa) extracellular domain (ECD) with six cysteine-rich repeats, a 28 aa transmembrane segment, and a 188 aa cytoplasmic domain (3). In contrast, mouse and rat CD30 lack 90 aa of the ECD and contain only three cysteine-rich repeats. Within common regions of the ECD, human CD30 shares 53% and 49% aa sequence identity with mouse and rat CD30, respectively. Alternate splicing of human CD30 generates an isoform that includes only the C-terminal 132 aa of the cytoplasmic domain. CD30 is normally expressed on antigen-stimulated Th cells and B cells (4 - 6). However, it is upregulated in Hodgkin's disease (on Reed-Sternberg cells), other lymphomas, chronic inflammation, and autoimmunity (7). CD30 binds to CD30 Ligand/TNFSF8 which is expressed on activated Th cells, monocytes, granulocytes and medullary thymic epithelial cells (1, 5). CD30 signaling costimulates antigen-induced Th0 and Th2 proliferation and cytokine secretion but favors a Th2-biased immune response (8). In the absence of antigenic stimulation, it can still induce T cell expression of IL-13 (9). CD30 contributes to thymic negative selection by inducing the apoptotic cell death of CD4+CD8+ T cells (10, 11). In B cells, CD30 ligation promotes cellular proliferation and antibody production in addition to the expression of CXCR4, CCL3, and CCL5 (5, 12). An 85-90 kDa soluble form of CD30 is shed from the cell surface by TACE-mediated cleavage (13, 14). Soluble CD30 retains the ability to bind CD30 Ligand and functions as an inhibitor of normal CD30 signaling (15).

References:

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