

#### DESCRIPTION

<b>Species Reactivity</b>	Human
<b>Specificity</b>	Detects recombinant human CD69 protein in Direct ELISA.
<b>Source</b>	Recombinant Monoclonal Rabbit IgG Clone # 3190A
<b>Purification</b>	Protein A or G purified from hybridoma culture supernatant
<b>Immunogen</b>	Mouse myeloma cell line, NS0-derived human CD69 Gly64-Lys199 Accession # Q07108
<b>Formulation</b>	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose.

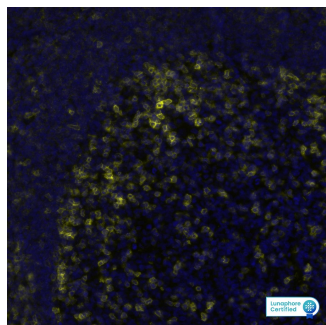
#### APPLICATIONS

**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.

	<b>Recommended Concentration</b>	<b>Sample</b>
<b>Western Blot</b>	2 µg/mL	Human tonsil, human thymus and human CD4+ T cells
<b>Multiplex Immunofluorescence</b>	25 µg/mL	Immersion fixed paraffin-embedded sections of human Tonsil
<b>Immunohistochemistry</b>	1-10 µg/mL	Immersion fixed paraffin-embedded sections of human lymph node and human spleen

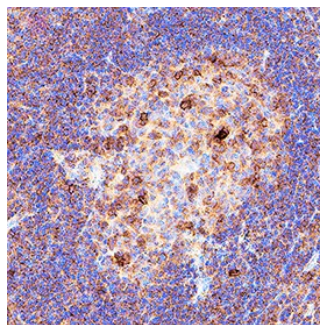
DATA

**Multiplex Immunofluorescence**



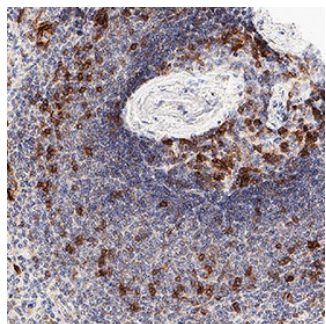
**CD69 in Human Tonsil via seqIF™ staining on COMET™**  
CD69 was detected in immersion fixed paraffin-embedded sections of human Tonsil using Rabbit Anti-Human CD69, Monoclonal Antibody (Catalog # MAB11691) at 25µg/mL at 37° Celsius for 4 minutes. Before incubation with the primary antibody, tissue 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™ Plus 647 Goat anti-Rabbit IgG Secondary Antibody at 1:200 at 37 ° Celsius for 2 minutes. (Yellow; Lunaphore Catalog # DR647RB) and counterstained with DAPI (blue; Lunaphore Catalog # DR100). Specific staining was localized to the membrane. Protocol available in [COMET™ Panel Builder](#).

**Immunohistochemistry**



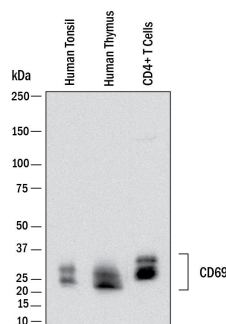
**Detection of CD69 in Human Lymph Node.** CD69 was detected in immersion fixed paraffin-embedded sections of human lymph node using Rabbit Anti-Human CD69 Monoclonal Antibody (Catalog # MAB11691) at 3 µg/ml for 1 hour at room temperature followed by incubation with the Anti-Rabbit IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC003) or the HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # HAF008). Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using VisUCyte Antigen Retrieval Reagent-Basic (Catalog # VCTS021). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to the cell surface. View our protocol for [IHC Staining with VisUCyte HRP Polymer Detection Reagents](#).

**Immunohistochemistry**



**Detection of CD69 in Human Spleen.** CD69 was detected in immersion fixed paraffin-embedded sections of human spleen using Rabbit Anti-Human CD69 Monoclonal Antibody (Catalog # MAB11691) at 3 µg/ml for 1 hour at room temperature followed by incubation with the Anti-Rabbit IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC003) or the HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # HAF008). Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using VisUCyte Antigen Retrieval Reagent-Basic (Catalog # VCTS021). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to the cell surface. View our protocol for [IHC Staining with VisUCyte HRP Polymer Detection Reagents](#).

**Western Blot**



**Detection of Human CD69 by Western Blot.** Western Blot shows lysates of human tonsil tissue, human thymus tissue and human CD4+ T cells. PVDF membrane was probed with 2 µg/ml of Rabbit Anti-Human CD69 Monoclonal Antibody (Catalog # MAB11691) followed by HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # HAF008). A specific band was detected for CD69 at approximately 20-35 kDa (as indicated). This experiment was conducted under reducing conditions and using [Western Blot Buffer Group 1](#).

**PREPARATION AND STORAGE**

<b>Reconstitution</b>	Reconstitute lyophilized material at 0.2 mg/ml in sterile PBS. For liquid material, refer to CoA for concentration.
<b>Shipping</b>	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.
<b>Stability &amp; Storage</b>	<p>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</p> <ul style="list-style-type: none"> <li>• 12 months from date of receipt, -20 to -70 °C as supplied.</li> <li>• 1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>• 6 months, -20 to -70 °C under sterile conditions after reconstitution.</li> </ul>

**BACKGROUND**

CD69, also known as Leu 23, AIM, EA-1, and MLR-3, is a type II transmembrane glycoprotein and is expressed on activated T cells, B cells, NK cells, neutrophils and eosinophils, Langerhans cells, and platelets.