

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human CD99 in direct ELISAs and Western blots. In direct ELISAs, less than 5% cross-reactivity with recombinant mouse CD99 is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	Mouse myeloma cell line NS0-derived recombinant human CD99 Asp23-Asp122 Accession # P14209
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

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	0.1 µg/mL	Recombinant Human CD99 Fc Chimera (Catalog # 3968-CD)
Flow Cytometry	0.25 µg/10 ⁶ cells	See Below
Immunocytochemistry	5-15 µg/mL	See Below
CyTOF-ready	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	

DATA

<p>Flow Cytometry</p>	<p>Detection of CD99 in Human CD3⁺ Lymphocytes by Flow Cytometry. Human whole blood CD3⁺ lymphocytes were stained with Goat Anti-Human CD99 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3968, filled histogram) or control antibody (Catalog # AB-108-C, open histogram), followed by Phycoerythrin-conjugated Anti-Goat IgG Secondary Antibody (Catalog # F0107).</p>	<p>Immunocytochemistry</p>	<p>CD99 in HepG2 Human Cell Line. CD99 was detected in immersion fixed HepG2 human hepatocellular carcinoma cell line using Goat Anti-Human CD99 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3968) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (red; Catalog # NL001) and counterstained with DAPI (blue). Specific staining was localized to cell membranes. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.</p>
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PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> ● 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.

BACKGROUND

CD99 (also named MIC2, E2 and thymic leukemia antigen) is the founding member of the CD99 family of molecules. The CD99 family contains four members; CD99, CD99L2, XG and the pseudogene CD99L1 (1, 2, 3). Native human CD99 is 32 kDa in size and exists as a type I transmembrane glycoprotein. This is referred to as the long, or type I isoform. It is synthesized as a 185 amino acid (aa) precursor that contains a 22 aa signal sequence, a 100 aa extracellular domain (ECD), a 25 aa transmembrane segment, and a 38 aa cytoplasmic region (4). The ECD contains no identifiable motifs, N-linked glycosylation sites, or cysteine residues; it does possess sites for O-linked glycosylation. The cytoplasmic region, albeit short, does have signal transduction capability (5). There are apparently multiple isoforms for human CD99. One shows a 16 aa deletion in the ECD (aa 34-49), a second shows a 38 aa deletion in the cytoplasmic region (aa 122-159), and a third exhibits a three aa truncation at the C-terminus (6, 7, 8). The best studied isoform shows an Asp-Gly substitution for the C-terminal 27 amino acids. This is referred to as the 28 kDa type II isoform (9). The type I and II isoforms have distinctive signal transduction pathways (FAK-src for type I; PI3K plus src-ERK1/2 for type II), and mediate clearly different biological outcomes (5, 9, 10). The two numbered isoforms may or may not co-exist on the same cells. Peripheral T cells have only the long isoform, while double-positive thymocytes express both isotypes. What is unclear is the monomeric vs. dimeric status of CD99. In mouse, CD99 reportedly forms disulfide-linked homodimers (11). In human, however, CD99 is reportedly monomeric if only a type I isoform, and a covalent heterodimer if co-expressing type I and II isoforms (12, 13). Cells known to express CD99 include fibroblasts, neutrophils, T cells, double-positive thymocytes, CD34⁺ stem cells, monocytes and endothelial cells (2, 12, 14, 15). Homophilic interaction between CD99 on the neutrophil and CD99 on the endothelial cell regulates the transendothelial migration of neutrophils during inflammation (16). Human CD99 is only 48% aa identical to mouse CD99 (17).

References:

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