

ORDERING INFORMATION

Catalog Number: AF3699

Lot Number: XXV01

Size: 100 µg

Formulation: 0.2 µm filtered solution in PBS with 5% trehalose

Storage: -20° C

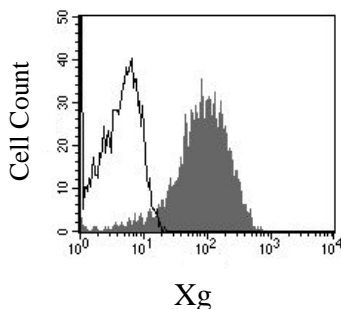
Reconstitution: sterile PBS

Specificity: human Xg extracellular domain

Immunogen: NS0-derived rhXg extracellular domain

Ig Type: goat IgG

Applications: Western blot
Flow Cytometry
Direct ELISA



Human erythrocytes were stained with anti-Xg (R&D Systems, Cat. # AF3699, filled histogram) or control antibody (R&D Systems, Cat. # AB-108-C, open histogram) followed by PE-conjugated anti-goat antibody (R&D Systems, Cat. # F0107).

Preparation

Produced in goats immunized with purified, NS0-derived, recombinant human Xg blood group (rhXg; aa 22 - 142; Accession # P55808) extracellular domain. Human Xg specific IgG was purified by human Xg affinity chromatography. Xg, also known as PBDX (pseudoautosomal boundary divided on the X chromosome), is a type I transmembrane glycoprotein that belongs to the CD99 family. It is an erythrocyte membrane protein that is used as the basis for the Xg blood group system. Mature Xg is a 26 kDa glycoprotein that consists of a 121 aa extracellular domain, a transmembrane segment, and a 17 aa cytoplasmic tail. Xg isoforms with either a 15 aa insertion or a deletion of nearly the entire ECD have been reported. Within the extracellular domain, human Xg shares 100% aa sequence identity with mouse Xg.

Formulation

Lyophilized from a 0.2 µm filtered solution in phosphate-buffered saline (PBS) with 5% trehalose.

Reconstitution

Reconstitute with sterile PBS. If 0.5 mL of PBS is used, the antibody concentration will be 0.2 mg/mL.

Storage

Lyophilized samples are stable for twelve months from date of receipt when stored at -20° C to -70° C. Upon reconstitution, the antibody can be stored at 2° - 8° C for 1 month without detectable loss of activity. Reconstituted antibody can also be aliquotted and stored frozen at -20° C to -70° C in a manual defrost freezer for six months without detectable loss of activity. **Avoid repeated freeze-thaw cycles.**

Specificity

This antibody has been selected for its ability to recognize human Xg in the applications listed below.

Applications

Western blot - This antibody can be used at 0.1 - 0.2 µg/mL with the appropriate secondary reagents to detect human Xg. The detection limit for rhXg is approximately 5 ng/lane under non-reducing and reducing conditions.

Flow Cytometry - This antibody was tested in flow cytometry using erythrocytes. Dilute this antibody to 50 µg/mL and add 10 µL of the diluted solution to 1-2.5 x10⁵ cells in a total reaction volume not exceeding 200 µL. The binding of unlabeled polyclonal antibodies may be visualized by adding a secondary developing reagent such as anti-goat IgG conjugated to a fluorochrome.

Direct ELISA - This antibody can be used at 0.5 - 1.0 µg/mL with the appropriate secondary reagents to detect human Xg. The detection limit for rhXg is approximately 0.2 ng/well.

Optimal dilutions should be determined by each laboratory for each application.