

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human PSGL-1 in direct ELISAs.
Source	Monoclonal Mouse IgG ₁ Clone # 688102
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	Chinese hamster ovary cell line CHO-derived recombinant human PSGL-1 Gln42-Gly295 Accession # Q14242
Endotoxin Level	<0.10 EU per 1 µg of the antibody by the LAL method.
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.

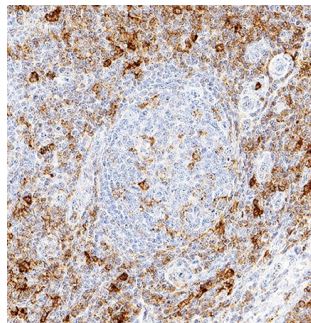
APPLICATIONS

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
Immunohistochemistry	5-25 µg/mL	See Below
Neutralization	The adhesion of U937 human histiocytic lymphoma cells (5×10^4 cells/well, 30 minute preincubation with the antibody) to immobilized Recombinant Human P-Selectin/CD62P (Catalog # ADP3 , 10 µg/mL, 100 µL/well) was inhibited by 10 µg/mL of the antibody.	

DATA

Immunohistochemistry



PSGL-1/CD162 in Human Tonsil. PSGL-1/CD162 was detected in immersion fixed paraffin-embedded sections of human tonsil using Mouse Anti-Human PSGL-1/CD162 Monoclonal Antibody (Catalog # MAB3345) at 5 µg/mL for 1 hour at room temperature followed by incubation with the Anti-Mouse IgG VisUCyte™ HRP Polymer Antibody (Catalog # [VC001](#)). Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # [CTS013](#)). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to cytoplasm in lymphocytes. View our protocol for [IHC Staining with VisUCyte HRP Polymer Detection Reagents](#).

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 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. • 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

Human PSGL-1 (P-Selectin Glycoprotein Ligand-1; also CD162), is a 120 kDa mucin-type glycoprotein that plays a key role in leukocyte adhesion (1-3). It is synthesized as a 412 amino acid (aa) preproprecursor that contains a 17 aa signal sequence, a 24 aa propeptide, a 279 aa extracellular domain (ECD), a 21 aa transmembrane segment and a 71 aa cytoplasmic region (4, 5). Following cleavage of the pre- and prosegments, it is expressed as a 240 kDa disulfide-linked homodimer. The extreme N-terminus (aa 1-16 of the mature molecule) contains one threonine (aa 16) and three tyrosines (aa 5, 7, and 10) that are involved in ligand binding. The Thr residue allows for O-linked glycosylation in the form of a core-2 structure (GalNAc-Gal) linked in a β 1,6 bond to a sialylated Lewis X motif (GlcNAc linked to both Fuc and Gal with a terminal sialic acid residue) (1, 2, 5, 6, 7). The three tyrosine residues allow for sulfation (8, 9). When binding to P-selectin, Tyr sulfation and glycosylation are essential. Tyr7 provides the most efficient sulfate moiety, while Fuc and sialic acid are essentially mandatory (7). When binding to E-selectin, only carbohydrate is needed, while both carbohydrate and Tyr10 are used for L-selectin binding (6, 8). There are 16 decameric aa repeats in the ECD of the longform of PSGL-1. This form is referred to as the A allele, and represents 65 - 80% of the population. Alleles B and C show deletions of decameric repeats #2 (aa 132-141) plus #9 and 10 (aa 222-241), respectively. Shorter forms may show weaker binding to P-selectin (9, 10). Soluble forms of PSGL-1 are also known. Neutrophil elastase will cleave somewhere within repeats #5-9, while cathepsin G cleaves after Tyr7 (11). The loss of Tyr5 and 7 should impact binding affinity. PSGL-1 is found on virtually all leukocytes and macrophages/DC's (1). Although there is similarity in the organization of the ECD between species, there is little aa identity. Human PSGL-1 ECD shares 51%, 52% and 43% aa sequence identity with equine, canine and mouse ECD, respectively.

References:

1. Yang, J. *et al.* (1999) *Thromb. Haemost.* **81**:1.
2. Cummings, R.D. (1999) *Braz. J. Med. Biol. Res.* **32**:519.
3. McEver, R.P. and R.D. Cummings (1997) *J. Clin. Invest.* **100**:485.
4. Sako, D. *et al.* (1993) *Cell* **75**:1179.
5. Veldman, G.M. *et al.* (1995) *J. Biol. Chem.* **270**:16470.
6. Bernimoulin, M.P. *et al.* (2003) *J. Biol. Chem.* **278**:37.
7. Leppanen, A. *et al.* (2000) *J. Biol. Chem.* **275**:39569.
8. Sako, D. *et al.* (1995) *Cell* **83**:323.
9. Afshar-Kharghan, V. *et al.* (2001) *Blood* **97**:3306.
10. Lozano, M.L. *et al.* (2001) *Br. J. Haematol.* **115**:969.
11. Gardiner, E.E. *et al.* (2001) *Blood* **98**:1440.