

#### DESCRIPTION

|                           |   |
|---------------------------|---|
| <b>Species Reactivity</b> | Human   |
| <b>Specificity</b>        | Detects human PSGL-1/CD162. Recognizes sLe <sup>x</sup> -bearing core 2 O-glycan structures. It does not recognize sLe <sup>x</sup> on an extended core 1 O-glycan. The sLe <sup>x</sup> -bearing, core 2 O-glycan structure decorates the P-Selectin ligand PSGL-1, and the presence of this glycan structure is required for high affinity P-Selectin binding (1). This antibody stains human and canine leukocytes but does not recognize monkey, mouse, rabbit, porcine, feline or bovine leukocytes. |
| <b>Source</b>             | Monoclonal Mouse IgM Clone # CHO131   |
| <b>Purification</b>       | IgM-specific Affinity-purified from hybridoma culture supernatant   |
| <b>Immunogen</b>          | CHO Chinese hamster ovary cell line transfected with human PSGL-1/CD162   |
| <b>Conjugate</b>          | Alexa Fluor 647<br>Excitation Wavelength: 650 nm<br>Emission Wavelength: 668 nm   |
| <b>Formulation</b>        | Supplied 0.2 mg/mL in a saline solution containing BSA and Sodium Azide. See Certificate of Analysis for details.<br><br>*Contains <0.1% Sodium Azide, which is not hazardous at this concentration according to GHS classifications. Refer to the Safety Data Sheet (SDS) for additional information and handling instructions.  |

#### 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>Flow Cytometry</b> | 0.25-1 µg/10 <sup>6</sup> cells  | Human whole blood monocytes and granulocytes |

#### PREPARATION AND STORAGE

|                                |   |
|--------------------------------|---|
| <b>Shipping</b>                | The product is shipped with polar packs. Upon receipt, store it immediately at the temperature recommended below. |
| <b>Stability &amp; Storage</b> | <b>Protect from light. Do not freeze.</b><br>● 12 months from date of receipt, 2 to 8 °C as supplied.             |

#### 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 β1,6 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.

**PRODUCT SPECIFIC NOTICES**

This product is provided under an agreement between Life Technologies Corporation and R&D Systems, Inc, and the manufacture, use, sale or import of this product is subject to one or more US patents and corresponding non-US equivalents, owned by Life Technologies Corporation and its affiliates. The purchase of this product conveys to the buyer the non-transferable right to use the purchased amount of the product and components of the product only in research conducted by the buyer (whether the buyer is an academic or for-profit entity). The sale of this product is expressly conditioned on the buyer not using the product or its components (1) in manufacturing; (2) to provide a service, information, or data to an unaffiliated third party for payment; (3) for therapeutic, diagnostic or prophylactic purposes; (4) to resell, sell, or otherwise transfer this product or its components to any third party, or for any other commercial purpose. Life Technologies Corporation will not assert a claim against the buyer of the infringement of the above patents based on the manufacture, use or sale of a commercial product developed in research by the buyer in which this product or its components was employed, provided that neither this product nor any of its components was used in the manufacture of such product. For information on purchasing a license to this product for purposes other than research, contact Life Technologies Corporation, Cell Analysis Business Unit, Business Development, 29851 Willow Creek Road, Eugene, OR 97402, Tel: (541) 465-8300. Fax: (541) 335-0354.