



Product Information

MONOCLONAL ANTI-HUMAN CD62P (P-Selectin, LECAM-3), CLONE AK-4, FITC CONJUGATE

Product Number **F0799**

Product Description

Monoclonal Anti-Human CD62P (P-Selectin, LECAM-3) (mouse IgG1 isotype) is derived from the AK-4 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from BALB/c mice immunized with a purified human platelet glycoprotein membrane fraction (100-200 kD) immunodepleted of GP-lib/IIIa (CD41).¹ The immunoglobulin fraction of antibody to CD62P is purified (protein A) from ascites fluid and then conjugated to fluorescein isothiocyanate (FITC) isomer I. The conjugate is purified by gel filtration and contains no detectable free FITC.

Monoclonal Anti-Human CD62P (P-Selectin, LECAM-3) – FITC Conjugate reacts specifically with CD62P (P-Selectin) antigen expressed on the surface of activated endothelial cells and platelets

P-Selectin (CD62P, GMP-140, LECAM-3, PADGEM) is a member of the LEC-CAM family of adhesion receptors that recognize specific carbohydrate ligands, and mediates an early step in the interaction of leukocytes with endothelium, megakaryocytes and platelets. It is a cell surface 140-150 kD glycoprotein, of which the extracellular region contains a NH₂-terminal C-type lectin domain, followed by a homologous EFG-like domain and nine short consensus repeats (SCR).² P-Selectin is an α granule membrane glycoprotein that is rapidly translocated to the plasma membrane of platelets in response to various inflammatory and thrombogenic agents. It is located in the endothelial Weibel-Palade bodies and on the surface of activated endothelial cells. A circulating soluble form of P-selectin is found in the blood.³ Levels of soluble P-selectin in biological fluids may be elevated in patients with various pathological conditions.

P-selectin mediates transient neutrophil adhesion to thrombin-stimulated endothelial cells membrane ('rolling' at physiological shear stress) and the binding of activated platelets to myeloid cells. It binds to the carbohydrate sialyl Lewis^x (CD15s) on neutrophils and to sulphated galactosyl ceramides on neutrophils and tumor cells. P-Selectin glycoprotein ligand -1 mediates leucocyte - endothelial adhesion by binding to P-selectin on activated endothelia and platelets.

Binding of P-selectin to its ligand is Ca²⁺ dependent.⁴ Monoclonal Anti-Human CD62P (P-selectin) was shown to partially inhibit the P-selectin-mediated rosetting of HL-60 cells by thrombin-activated platelets.⁵

Reagents

The product is supplied in 0.01 M phosphate buffered saline, pH 7.4, containing 1% BSA and 15 mM sodium azide (see MSDS)* as a preservative.

The F/P molar ratio of the product is 3-8.

Precautions and Disclaimer

* Due to the sodium azide content a material safety sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazardous and safe handling practices.

Storage/Stability

For continuous use store at 2-8°C. **Protect from prolonged exposure to light.** If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use.

Product Profile

When assayed by flow cytometric analysis (FACScan), using 10 μ l of the antibody to stain 1 x 10⁶ cells (ADP activated human platelets), a fluorescence intensity is observed similar to that obtained with saturating monoclonal antibody levels. Also, the percent population positive is at the maximum percentage positive using saturating monoclonal antibody levels.

Notes:

1. In order to obtain best results in different techniques and preparations we recommend determining optimal working dilution by titration test.
2. It is advisable to run the appropriate negative controls. Negative controls establish background fluorescence and non-specific staining of the primary antibody. The ideal negative control reagent is an FITC conjugated mouse monoclonal antibody or myeloma protein. It should be isotype-matched, F/P

molar ratio-matched, not specific for the tested preparation, and of the same concentration as the tested antibody. The degree of autofluorescence or negative control reagent fluorescence will vary with the type of preparation under study and the sensitivity of the instrument used. For fluorescent analysis of preparation expressing Fc receptors, the use of isotype-matched negative controls is mandatory.

References

1. Skinner, M.P., et al., *Biochem. Biophys. Res. Commun.*, **164**, 1373 (1989).
2. Johnston, G.I., et al., *Cell*, **56**, 1033 (1989).
3. Dunlop, L.C., et al., *J. Exp. Med.* **175**, 1147 (1992).
4. Tedder, T.F., et al., *FASEB J.*, **9**, 866 (1995).
5. Skinner, M.P., et al., *J. Biol. Chem.*, **266**, 5371 (1991).

Lpg/pcs 11/98

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