

## # M051-1

### Product Information

Catalog Number: M051-1  
Clone / Isotype: Xia.B4 / Rat (Wistar) IgG1  
Contents: FITC-labeled immunoglobulin in 20 mM Tris buffer with 137 mM NaCl, 0.5% BSA and 0.09% (w/v) sodium azide  
Size: 1.5 ml / 300 tests

**For research use only, not for diagnostic or therapeutic use. This product is no medical device.**

**Specificity:** The Xia.B4 antibody reacts with mouse GPIX (CD42a), a platelet/megakaryocyte-specific 17-22 kDa polypeptide that forms a non-covalent complex with GPIb $\alpha$ / $\beta$  (150/24 kDa) in the membrane. The GPIb-IX complex is the platelet receptor for von Willebrand factor (vWf)<sup>1,2</sup>. In contrast to GPIb $\alpha$ , GPIX is not sensitive to proteolytic cleavage during platelet activation. Therefore, Xia.B4 binds to resting and activated platelets with similar intensity<sup>3</sup>.

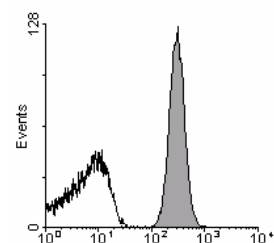
**Preparation and Storage:** The antibody was purified from hybridoma cell culture supernatant by Protein G-Sepharose chromatography. The antibody was conjugated with FITC under optimum conditions. The solution is free of unbound FITC. Store product undiluted at 4°C and avoid prolonged exposure to light. Stable for one year from date of shipment. Do not freeze.

**Usage:** The antibody preparation is optimized for flow cytometric applications: Use 5  $\mu$ l to stain  $\sim 10^6$  platelets or  $\sim 0.5 \times 10^6$  cells in a recommended volume of 25  $\mu$ l. Incubate for 15 minutes at room temperature, stop reaction by addition of 400  $\mu$ l PBS and analyze samples within 30 minutes. For immunofluorescent staining of acetone-fixed frozen sections, the appropriate dilution must be determined individually.

**Caution:** Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer.

#### Detection of GPIX on mouse platelets

Mouse blood was diluted 1:20 and 25  $\mu$ l of this dilution were stained with 5  $\mu$ l control IgG-FITC (emfret Analytics, P190-1, *black line*) or Xia.B4-FITC (*shaded area*) for 15 min at RT and analyzed directly. Platelets were gated by FSC/SSC characteristics.



Log fluorescence intensity. FL1  
Mouse platelets

#### References:

- Berndt MC, Shen Y, Dopheide SM, et al. (2001) The vascular biology of the glycoprotein Ib-IX-V complex. *Thromb Haemost.* 2001 Jul;86(1):178-88.
- Bergmeier W, Rackebrandt K, Schroder W, Zirngibl H, Nieswandt B. (2000) Structural and functional characterization of the mouse von Willebrand factor receptor GPIb-IX with novel monoclonal antibodies. *Blood.* 95:886-93.
- emfret Analytics. Unpublished results.