

## Data Sheet

# Anti-Glycophorin A,B ( $\alpha,\delta$ ) Antibody, Mouse Monoclonal

Clone E3, purified from hybridoma cell culture

**G7650**

## Product Description

Monoclonal Anti-Glycophorin A, B ( $\alpha,\delta$ ) (mouse IgG1 isotype) is derived from the E3 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from BALB/c mice immunized with human thymus.<sup>1,2</sup> The isotype is determined by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents, Cat. No. ISO2.

Monoclonal Anti-Glycophorin A, B ( $\alpha,\delta$ ) recognizes an epitope located within amino acid residues 1-26, for example, in the fragment which is MN-unrelated and identical in glycophorins A and B. This epitope does not require sialylation.<sup>3,4</sup> The antibody localizes specifically the  $\alpha$ ,  $\delta$ ,  $\alpha\delta$ ,  $\alpha_2$ ,  $\delta_2$ ,  $\alpha_2\delta$ , and  $\alpha_3$  bands in extracts of human red blood cell ghosts, applying the immunoblotting technique. The product stains human erythrocytes in smears and tissue preparations using immunofluorescence assays and flow cytometry. It binds to a mean of 12% bone marrow nucleated cells. The E3 antibody was submitted to two Workshops on "Monoclonal Antibodies Against Human Red Blood Cells and Related Antigens". To the 1st Workshop (1987)<sup>5</sup>, the product was submitted under the code 32W3, clone 6H5C1 (E3). To the 2nd Workshop (1990)<sup>3,4</sup>, the product was submitted under the code 151, clone 6H5 clone 1. The antibody has been described by its developer as clone E3.<sup>1,2</sup>

Monoclonal Anti-Glycophorin A, B ( $\alpha,\delta$ ) may be used for the localization of glycophorins A and B using immunoblot, immunoprecipitation, immunocytochemistry, agglutination or flow cytometry.

The erythrocyte membrane contains four main sialic acid-rich polypeptides (sialoglycoproteins) known as glycophorins (GP)<sup>6</sup> they are denoted  $\alpha,\beta,\gamma,\delta$ , in order of decreasing apparent molecular weight. Other nomenclatures are also used.<sup>7</sup> GPA and GPB, the major constituents of the red cells, appear as single polypeptides ( $\alpha$  and  $\delta$ ) but form also stable dimeric complexes ( $\alpha_2$  and  $\delta_2$ ) and heterodimers ( $\alpha\delta$ ) under electro-phoretic conditions. GPA carries blood group M or N activity depending upon the amino acid residues at positions 1 and 5. GPB carries blood group N activity as well as blood group S or s antigen activity associated with amino acid substitution at residue 29. GPA and GPB have related protein sequences. GPA has been considered to be associated exclusively with erythroid cells, for example, to be expressed in pronormoblasts and later erythroid cells but not on the surface of normal committed erythroid cells and proerythroblasts or other hemopoietic cells. Antibodies specific for GPA have been widely used in the diagnosis of leukemias and erythroid differentiation.

## Reagents

Supplied in 0.01 M phosphate buffered saline, pH 7.4, containing 1% BSA and 15 mM sodium azide as a preservative.

## Precautions and Disclaimer

For R&D use only. Not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

## Product Profile

Antibody concentration is determined by absorbance at 280 nm ( $E_{1\%}^{1\text{cm}} = 14$ ).

**Immunoblotting:** A working dilution of 0.5-1  $\mu\text{g/mL}$  was determined using extracts of human red blood cell ghosts.

**Agglutination:** A working dilution of 1:400 was determined by agglutination of human erythrocytes.

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**Note:** In order to obtain best results in different techniques and preparations, it is recommended that each individual user determine their optimum working dilutions by titration assay.

## Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For extended storage freeze in working aliquots. Repeated freezing and thawing, or storage in "frost-free" freezers, is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use.

## References

1. Telen, M.J., and Bolk, T.A., Transfusion, 27, 309 314 (1982).
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3. Anstee, D.J., and Lisowska, E., J. Immunogenet., 17, 301 (1990).
4. Messeter, L., and Johnson, V. (eds), Proc. 2nd Int. Workshop and Symp. on Monoclonal Antibodies against Human RBC and Related Antigens, J. Immunol., 17, Nos. 4/5 (1990).
5. Rouger, P., and Anstee, D., Vox. Sang., 55, 57 (1988).
6. Anstee, D.J., Vox. Sang., 58, 1 (1990).
7. Anstee, D.J., and Tanner, M.J.A., Br. J. Haematol., 64, 211 (1986).

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