

**International Blood Group  
Reference Laboratory**500 North Bristol Park  
Northway  
Filton  
Bristol  
BS34 7QH**Antigen** CD233 / Band 3 (extracellular domain)**Clone** BRAC 18**Product Code** 9452**Immunoglobulin Class** Rat IgG2b, kappa light chain**Protein Development  
and Production Unit****Tel:** +44 (0)117 921 7500**Fax:** +44 (0)117 912 5796**Website:** <http://ibgri.blood.co.uk>**Email:** [enquiries.IBGRL@nhsbt.nhs.uk](mailto:enquiries.IBGRL@nhsbt.nhs.uk)**Antigen Description and Distribution**

CD233 (also known as erythrocyte band 3, EPB3, anion exchange protein 1, AE1, solute carrier family 4, SLC4A1) is an integral membrane protein in human erythrocytes, present at approximately  $10^6$  copies per human erythrocyte. It comprises two domains that are structurally and functionally distinct. The cytoplasmic N-terminal 40kDa domain has binding sites for erythrocyte cytoskeletal proteins, namely ankyrin and protein 4.2, which help to maintain the mechanical properties and integrity of the erythrocyte. This domain also binds a number of other erythrocyte peripheral proteins. The 55kDa glycosylated C-terminal membrane-associated domain contains 12-14 membrane spanning segments which function as a chloride/bicarbonate anion exchanger involved in carbon dioxide transport. The cytoplasmic tail at the extreme C-terminus of the membrane domain binds carbonic anhydrase II. CD233 associates with the erythrocyte membrane protein glycophorin A (GPA) which promotes the correct folding and translocation during biosynthesis of CD233. Many CD233 mutations are known in man and these mutations can lead to two types of disease; destabilization of the erythrocyte membrane leading to hereditary spherocytosis, and defective kidney acid secretion leading to distal renal tubular acidosis. Other CD233 mutations that do not give rise to disease result in novel blood group antigens, which form the Diego blood group system. The CD233 gene is located on chromosome 17q21-q22<sup>1</sup>.

**Clone**

BRAC 18 was made in response to intact human erythrocytes<sup>2</sup>. BRAC 18 binds to an exofacial epitope on CD233 and agglutinates normal erythrocytes indirectly. BRAC 18 fails to agglutinate pronase-treated normal erythrocytes but agglutinates cells that have been treated with trypsin, sialidase, or 6% aminoethylisothiuronium bromide. BRAC 18 gives markedly reduced titres with chymotrypsin treated cells. Chymotrypsin treatment of intact erythrocytes cleaves the membrane domain of CD233 after Try-553<sup>3</sup> and try-558<sup>4</sup>. The exact binding site of BRAC 18 is unknown but, because it fails to agglutinate pronase treated erythrocytes, it probably binds somewhere on the extracellular loop between amino acids 545 and 567<sup>2</sup>. BRAC 18 does not react with electrophoretically separated components of human erythrocyte membranes by immunoblotting. Immunoprecipitation from radioiodinated untreated and trypsin treated erythrocytes show a diffuse labelled band of Mr 95, 000. BRAC 18 reacts by immune fluorescence with normal peripheral blood granulocytes and the myeloid cell lines HL60 and U937.

**References**

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3. Steck T.L. J Supramolec Struct 8:311 (1978).
4. Jennings M.L., Adams-Lackey M., Denney G.H. J Biol Chem 259: 4652 (1984).