

**International Blood Group
Reference Laboratory**500 North Bristol Park
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Antigen	CD233 / (Band 3) extracellular domain
Clone	BRIC 6
Product Code	9439
Immunoglobulin Class	Mouse IgG3, kappa light chain

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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-q22¹.

Clone

BRIC 6 was made in response to intact human erythrocytes². BRIC 6 binds to an exofacial epitope on CD233 and agglutinates normal erythrocytes directly. BRIC 6 fails to agglutinate pronase-treated normal erythrocytes but agglutinates cells that have been treated with trypsin, sialidase, or 6% aminoethylisothiuronium bromide. The exact binding site of BRIC 6 is unknown but, because it fails to agglutinate pronase treated erythrocytes, it probably binds somewhere on the extracellular loop between amino acids 545 and 567². BRIC 6 gives markedly reduced titres with chymotrypsin treated cells. Chymotrypsin treatment of intact erythrocytes cleaves the membrane domain of CD233 after Try-553³ and try-558⁴. BRIC 6 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. BRIC 6 reacts weakly by immune fluorescence with the myeloid cell line U937. BRIC 6 impedes the binding of anti-Wr^b. The Wr^b epitope is defined by interaction between CD233 and GPA and is dependent on GLU 658 in the fourth extracellular loop of CD233⁵. BRIC 6 reacts with normal CD233 but fails to react with South East Asian ovalocytosis CD233 when it is expressed in *Xenopus oocytes*⁶. BRIC 6 has been used to study the coexpression of band 3 mutants and Rh polypeptides⁷. BRIC 6 has also been used to study the band 3 macrocomplex in the erythrocyte membrane⁸. BRIC 6 has been used to investigate the key membrane protein changes during *in vitro* erythropoiesis of Protein 4.2 cells¹¹.

References

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