

PRODUCT DATA SHEET

ANTI-HUMAN CEACAM1, 3, 4, 5, 6 MONOCLONAL ANTIBODY

PRODUCT INFORMATION

Catalog Number:	GM-0505	Clone:	D14HD11
Description:	purified monoclonal mouse antibody	Specificity:	anti-human CEACAM1,3,4,5,6 (CD66a,c,d,e,CGM7)
Isotype:	IgG1/kappa	Purification:	Protein G
Storage:	short term: 2°C – 8°C; long term: –20°C (avoid repeated freezing and thawing)	Buffer:	phosphate buffered saline, pH 7.2
Immunogen:	immunization with extracted protein of CEACAM5	Selection:	based on recognition of the complete native protein expressed on transfected mammalian cells

WORKING DILUTIONS

Flow cytometry:	1.2 µg/10 ⁶ cells		
ELISA:	1:200 – 1:400	CELISA:	1:200 – 1:400
Western Blot:	4µg/ml	Immunofluorescence:	1µg/10 ⁶ cells

For each application a titration should be performed to determine the optimal concentration.

SPECIFICITY TESTING BY FLOW CYTOMETRY AND BY IMMUNOFLUORESCENCE MICROSCOPY

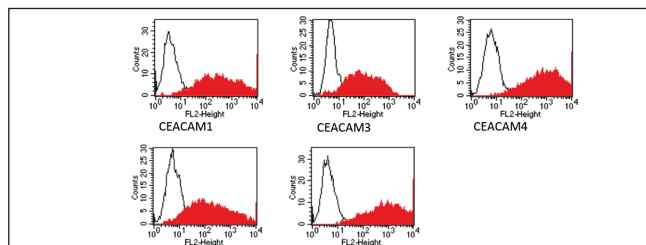


Fig.1: FACS analysis of BOSC23 cells using D14HD11 Cat.# GM-0505. BOSC23 cells were transiently transfected with an expression vector encoding either CEACAM1,3,4,5,6 (red curves) or an irrelevant protein (control transfectant: black curves). Binding of D14HD11 was detected with a PE- conjugated secondary antibody. A positive signal was obtained only with CEACAM1, CEACAM3, CEACAM4, CEACAM5 and CEACAM6 expressing cells.

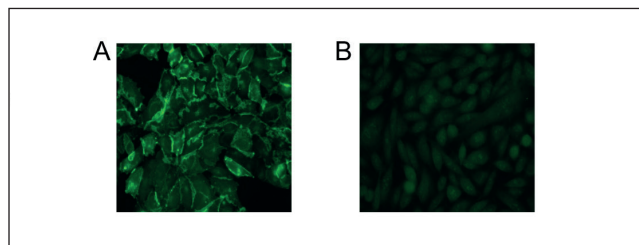


Fig.2: Immunofluorescence Microscopy of CHO cells using D14HD11 Cat.# GM-0505. CHO cells were transfected with an expression vector encoding CEACAM1 (A). Non-transfected CHO parental cells served as negative control (B). Binding of D14HD11 was visualized with a FITC-conjugated secondary antibody.

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ANTIBODY CROSS-REACTIVITY WITH MEMBERS OF THE CEA FAMILY

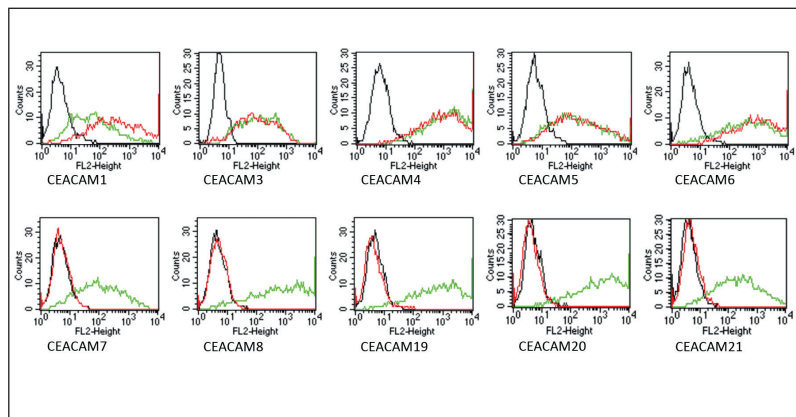


Fig. 3: BOSC cells were transiently transfected with expression vectors containing either the cDNA of CEACAM1, CEACAM3-CEACAM8 or CEACAM19-21. Recognition of CEACAM4 was tested on CHO cells stably transfected with a CEACAM4 expression vector. Expression of the constructs was confirmed with monoclonal antibodies known to recognise the corresponding proteins (CEACAM1: 4/3/17, CEACAM3,4: D14HD11, CEACAM5: 26/3/13, CEACAM6: 9A6, CEACAM7: BAC2, CEACAM8: GM-2H6, CEACAM19-21: anti-myc, green curves). An irrelevant monoclonal antibody served as a negative control (black curves). For specificity testing, protein G purified D14HD11 was tested on all CEA-CAM transfectants. A positive signal was obtained with CEACAM1, CEACAM3, CEACAM4, CEACAM5 and CEACAM6 expressing cells (red curves).

BACKGROUND

CEA-related cell adhesion molecules (CEACAM) belong to the carcinoembryonic antigen (CEA) family (1). The CEA family proteins belong to the immunoglobulin (Ig) superfamily and are composed of one Ig variable-like (IgV) and a varying number (0-6) of Ig constant-like (IgC) domains (1,2). CEACAM molecules are membrane-bound either via a transmembrane domain or a glycosylphosphatidyl inositol (GPI) anchor. CEACAM molecules are differentially expressed in epithelial cells or in leucocytes. Over-expression of CEA/CEACAM5 in tumours of epithelial origin is the basis of its wide-spread use as a tumour marker (2). The function of CEACAM family members varies widely: they function as cell adhesion molecules, tumour suppressors, regulators of lymphocyte and dendritic cell activation, receptors of *Neisseria* species and other bacteria (1).

REFERENCES

1. **Zimmermann W (2002).** Carcinoembryonic antigen. In Wiley Encyclopedia of Molecular Medicine (T. Creighton, ed.), John Wiley & Sons Inc., New York, USA, pp. 459-462.
2. **Hammarström S (1999).** The carcinoembryonic antigen (CEA) family: structures, suggested functions and expression in normal and malignant tissues. *Semin. Cancer Biol.* 9, 67-81.
3. **Grunert F, AbuHarfeil N, Schwarz K and von Kleist S (1985).** Two CEA and three NCA species, although distinguishable by monoclonal antibodies, have nearly identical peptide patterns. *Int. J. Cancer* 36, 357-362.
4. **Grunert F, Stocks SC, Nagel G., Zimmermann W, Thompson JA, Jantschkeff P and Kromer B. (1996).** CD66 family Workshop: Binding of myeloid blind panel antibodies and CD66 Subsection antibodies to HeLa transfectants expressing individual CD66 molecules. In *Leukocyte Typing VI: White cell Differentiation Antigens* (T. Kishimoto et al., eds.), Garland Publishing Inc., New York and London, pp. 1012-1025.

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