

# PRODUCT DATA SHEET

## ANTI-HUMAN CEACAM20 MONOCLONAL ANTIBODY

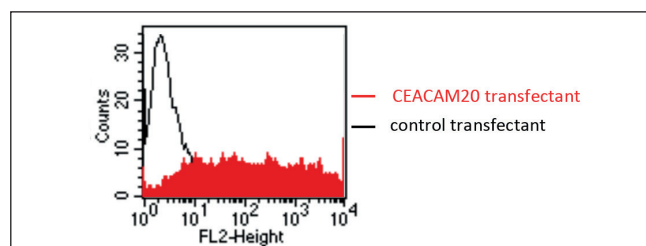
### PRODUCT INFORMATION

<b>Catalog Number:</b>	GM-0516	<b>Clone:</b>	HT-12D8
<b>Description:</b>	purified monoclonal mouse antibody	<b>Specificity:</b>	anti-human CEACAM20
<b>Isotype:</b>	IgG1/kappa	<b>Purification:</b>	Protein G
<b>Storage:</b>	short term: 2°C – 8°C; long term: –20°C (avoid repeated freezing and thawing)	<b>Buffer:</b>	phosphate buffered saline, pH 7.2
<b>Immunogen:</b>	genetic immunization with cDNA encoding human CEACAM20	<b>Selection:</b>	based on recognition of the complete native protein expressed on transfected mammalian cells

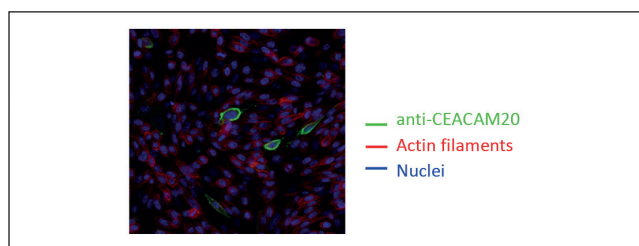
### WORKING DILUTIONS

<b>Flow cytometry:</b>	1.2 µg/10 <sup>6</sup> cells	<b>CELISA:</b>	1:200 – 1:400
<b>Immunofluorescence:</b>	1 µg/10 <sup>6</sup> cells	For each application a titration should be performed to determine the optimal concentration.	

### SPECIFICITY TESTING BY FLOW CYTOMETRY AND BY SPECTRAL CONFOCAL MICROSCOPY



**Fig. 1:** FACS analysis of BOSC23 cells using HT-12D8 Cat.# GM-0516. BOSC23 cells were transiently transfected with an expression vector encoding either CEACAM20 (red curve) or an irrelevant protein (control transfectant). Binding of HT-12D8 was detected with a PE- conjugated secondary antibody. A positive signal was obtained only with CEACAM20 transfected cells.



**Fig. 2:** Spectral Confocal Microscopy of CHO cells using HT-12D8 Cat.# GM-0516. CHO cells were transiently transfected with an expression vector encoding CEACAM20. Binding of HT-12D8 was visualized with a FITC- conjugated secondary antibody (green). Actin filaments are labeled with Alexa Fluor-555 Phalloidin (red). Cell nuclei are stained with DAPI (blue).

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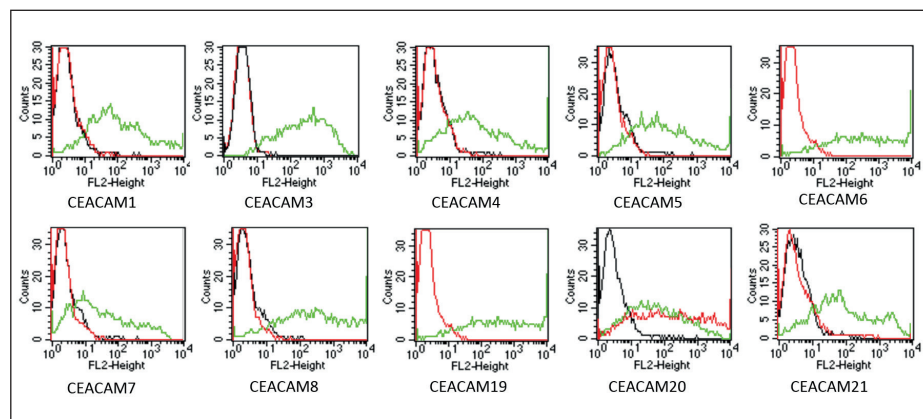
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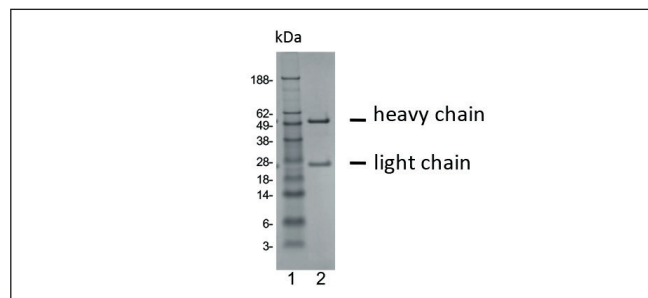
## ANTIBODY CROSS-REACTIVITY WITH MEMBERS OF THE CEA FAMILY



**Fig. 3:** BOSC23 cells were transiently transfected with expression vectors containing either the cDNA of CEACAM1, CEACAM3-8 or CEACAM19-21. Expression of the constructs was tested with monoclonal antibodies known to recognize the corresponding proteins (CEACAM1,3,4,5 and 6: D14HD11; CEACAM7: BAC2; CEACAM8: Tet2; CEACAM19,21:  $\alpha$ -myc; CEACAM20:  $\alpha$ -flag; green curves). An irrelevant monoclonal antibody served as a negative control (black curves). For specificity testing, protein G-purified HT-12D8 was tested on all CEACAM transfectants. A positive signal was obtained only with CEACAM20 transfected cells (red curve).

## SDS-PAGE ANALYSIS OF HT-12D8

The antibody was purified by protein G affinity chromatography from cell culture supernatants and verified by SDS-Page (Fig. 4).



**Fig. 4:** SDS-PAGE analysis of purified HT-12D8 monoclonal antibody. Lane 1: molecular weight marker, Lane 2: 2  $\mu$ g of purified HT-12D8 antibody. Proteins were separated by SDS-PAGE and stained with RAPID Stain™ Reagent.

## BACKGROUND

CEA-related cell adhesion molecule 20 (CEACAM20) belongs to the carcinoembryonic antigen (CEA) gene family (1). It encodes a putative glycoprotein which is membrane-bound via a transmembrane domain. The CEACAM20 protein contains a single N domain followed by 4 immunoglobulin-like A (A1, A2) and B (B1, B2) domains. Expression of CEACAM20 can be found in tissues of prostate, testis, duodenum and small intestine with highest expression in prostate. The function of CEACAM family members varies widely: they function as cell adhesion molecules, tumor suppressors, regulators of lymphocyte and dendritic cell activation, receptors of *Neisseria* species and other bacteria. High expression of CEACAM20 in tissue of prostate carcinoma and in prostate carcinoma cell lines suggest that CEACAM20 can be used as a tumor marker.

## 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.

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