

## Anti-HB-EGF (human) antibody, mouse monoclonal (4G10)

71-501 100 ug

**Storage:** Shipped at 4°C and store at -20°C (Do not store below -20°C)

**Product:** Mouse monoclonal antibody produced in serum-free medium and purified by combination of chromatography

**Reactivity:** React with human, but not with mouse

**Immunogen:** Recombinant human HB-EGF ectodomain expressed in SF21 cell

**Epitope:** Amino acids 136-149 in the EGF domain

**Applications** The antibody can react with both soluble and cell-surface attached forms of HG-EGF.

- 1) Western blotting (0.2~1 ug/ml), non-denaturing condition.
- 2) Immunoprecipitation (2 ug/ml)
- 3) Immunofluorescence staining (5~10 ug/ml)
- 4) Immunohistochemistry (assay dependent)
- 5) Inhibition of HB-EGF ectoderm shedding (Ref 1)
- 6) Inhibition of Diphtheria Toxin binding to HB-EGFR (Ref 1)

**Isotype:** IgG1 (mouse)

**Form:** 1 mg/ml in PBS, 50% glycerol, filter-sterilized, azide-free

**Background:** Heparin-binding epidermal growth factor-like growth factor (HB-EGF) is synthesized as a membrane-anchored precursor that is proteolytically cleaved to release the soluble mature growth factor, HB-EGF (1, 2). The former functions as juxtacrine and the latter as paracrine growth factor. Soluble HB-EGF shows several forms in Western blotting with apparent molecular weights 19~27 kDa due to heterogeneous O-glycosylation and N-terminal truncation. HB-EGF activates EGFR and ErbB4 and promotes the development in many tissues. In human ProHB-EGF is the cellular receptor for diphtheria toxin (3). Non-toxic mutant of diphtheria toxin, CRM197, inhibits HB-EGF function. As HB-EGF level is elevated in most ovarian cancer, CRM197 is being tested as an anticancer drug (4). The hybridoma clone 4G10 was established and characterized in the laboratory of Prof. E. Mekada of Osaka University, who is a leading scientist in this field

**Data Link** UniProtKB/Swiss-Prot [Q99075](#) (HBEGF\_HUMAN)

**Figure Identification of human HB-EGF by using anti-HB-EGF 4G10**

**(a) Western blotting (Non-reducing condition)**

Samples 1: Vero cell extract

Sample 2: Vero cells carrying human HB-EGF expression vector

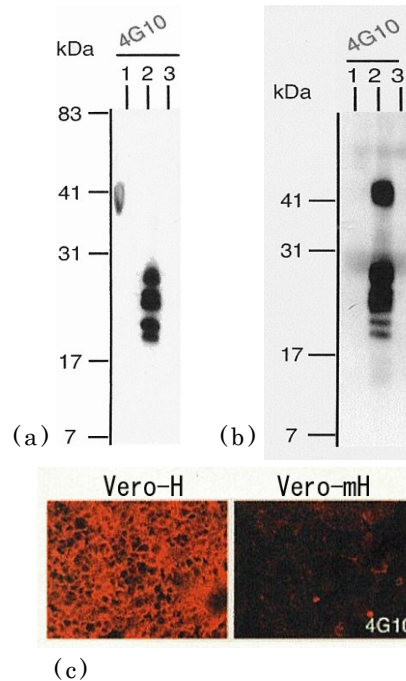
Sample 3: Vero cells carrying mouse HB-EGF expression vector

**(b) Immunoprecipitation**

Samples are the same as (a) except that the cell surface was biotinylated. After IP with the antibody, HRP-conjugated streptavidin was used to detect the HB-EGF by WB.

**(c) Immuno-cytochemistry**

Samples: (Vero-H) Vero cells carrying human HB-EGF expression vector, (Vero-mH) Vero cells carrying mouse HB-EGF expression vector. Cells treated with antibody 4G10 were fixed with 4% PFA and reacted with Cy3 conjugated 2<sup>nd</sup> antibody.



\*HB-EGF is glycosylated at multiple sites and processed, which reflects multiple-form in WB.

**Reference:** This antibody has been described and used in the following publications..

1. Hamaoka M. et al. Anti-human HB-EGF monoclonal antibodies inhibiting ectodomain shedding of HB-EGF and diphtheria toxin binding. J Biochem. 2010 Jul;148(1):55-69. PMID: [20332144](https://pubmed.ncbi.nlm.nih.gov/20332144/) WB, IP, IF/ICC (human)

**Related product:** # [71-503](#) Anti-HB-EGF antibody, biotinylated

# [01-515](#) Diphtheria toxin mutant CRM197

# [01-517](#) Diphtheria toxin