

Anti-HEV (Hepatitis E Virus) Capsid antibody, mouse monoclonal (161)

65-090 100 µg

Shipping and Storage: Ship at 4°C and store at -20°C.

Immunogen: Recombinant truncated capsid protein (amino acids 112–608) of HEV (genotype 3)

Specific Reactivity: Reacts with the capsid protein of HEV

Epitope: P domain (amino acids 457 to 608) of HEV capsid protein.

Applications:

1. Western blot (1/500~1/1,000)
2. Immunofluorescence staining (1/500)
3. ELISA (assay dependent)

Other applications have not been tested.

Background: Hepatitis E virus (HEV) is a single-strand positive-sense RNA virus in the family Hepeviridae. The disease caused by HEV is an important public health problem in developing countries. A molecular phylogenetic analysis classifies HEV into four major genotypes (genotype 1-4). The genome HEV consists of about 7200 bases and contains three discontinuous and partially overlapping open reading frames (ORFs). ORF1 encodes a methyltransferase, protease, helicase and replicase; ORF2 encodes the capsid protein and ORF3 encodes a protein of undefined function. The viral capsid protein induces neutralizing antibodies, and contains three subdomains, S (aa112-319), M (aa 320-456) and P (aa 457-608). Recombinant HEV-VLP is composed of approximately 53 kDa, smaller capsid protein subunit.

Isotype: mouse IgG1

Product: 1.0 mg/ml in PBS, 50% glycerol, filter sterilized.

Purity: IgG, affinity-purified with Protein A

Data Link: UniProKB [Q6J8F7](https://www.uniprot.org/entry/Q6J8F7) (CAPSD_HEVMG)

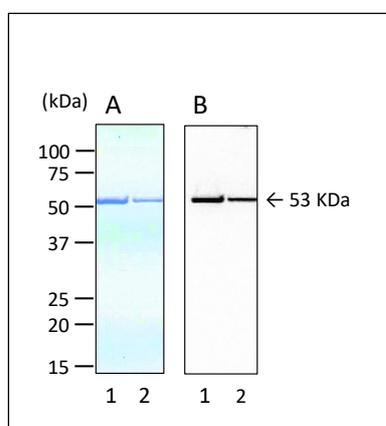


Fig.1. Identification of protein in HEV by SDS-PAGE (A) and Western blotting (B) using monoclonal antibody

Lane 1: Recombinant HEV- VLP (2.0mg/ml).

Lane 2: Recombinant HEV-VLP (0.5mg/ml)

The proteins were applied to SDS-PAGE and stained with Coomassie Brilliant Blue (CBB). In Western blotting, the monoclonal antibody was used at 1/500 dilution. A 53 kDa band was identified as HEV-VLP capsid protein.

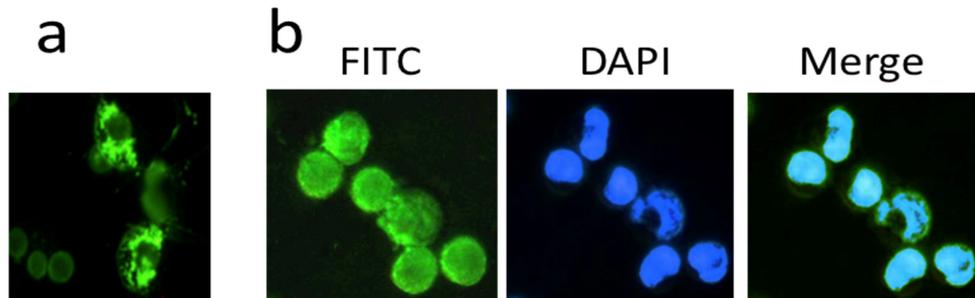


Fig.2. Detection of HEV protein infected in PLC/PRF/5 cells by immunofluorescence staining

(a) Infected and cultured cells on a slide glass. (b) Smear preparation after treatment with trypsin. The infected cells were fixed in cold acetone. The MAb was used at 1/500 dilution. As the second antibody, FITC-conjugated rabbit anti-mouse IgG was used at 1/4,000 dilution. The nucleus (DNA) was stained with DAPI.

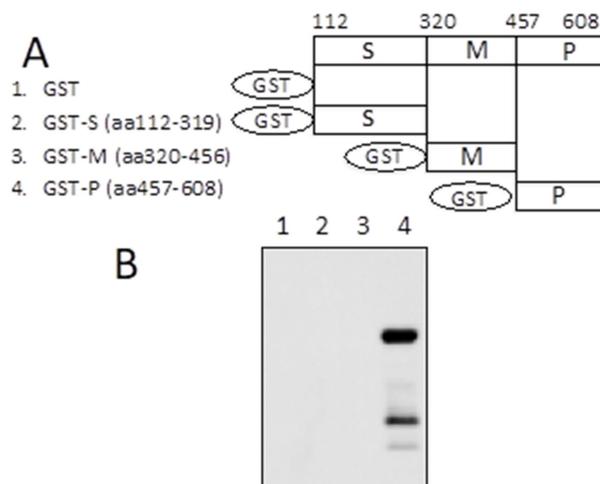


Fig.3. Determination of HEV-VLP subdomain recognized by monoclonal antibody 161

(A): Schematic representation of Glutathione-S-transferase (GST) -fused HEV-VLP subdomains used for epitope mapping. The cDNAs encoding the indicated amino acid residues were induced into a pGEX-4T3 vector and were expressed as GST-fused proteins in bacteria. (B): The lysates of cells expressing GST-fused proteins were analyzed by western blot, using anti-HEV monoclonal antibody clone 161. The lane numbers correspond to these in A. MAb recognized a region between aa 457 and 608 in the P subdomain. The main band (47kDa) is the GST fusion protein and the other bands are the degradation products.

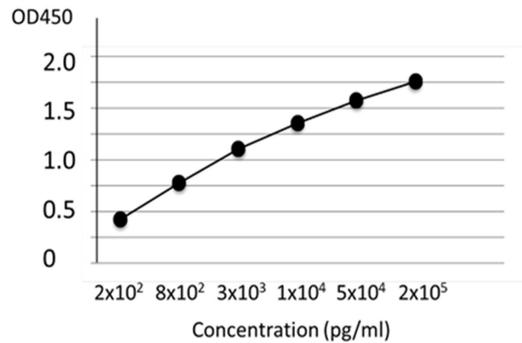


Fig.4. Titration of protein of HEV by indirect ELISA using monoclonal antibody

The indicated amounts of recombinant HEV-VLP was coated onto the wells of the ELISA plate. After blocking with 5% skim milk, monoclonal antibody at the 1/5000 dilution was added to the each well. HRP-conjugate goat anti-mouse IgG (100 μ l, x4000 dilution) was added. As substrate, orthophenylenediamine was used. Optical density (OD) measured at 490nm.

Reference: This antibody was described and used in the following publication.

Yamashita T et al. Biological and immunological characteristics of hepatitis E virus-like particles based on the crystal structure. [Proc Natl Acad Sci U S A](#). 2009 Aug 4;106(31):12986-91 PMID: [19620712](#) **WB, IP, ELISA**