

Anti-Influenza B Virus HA antibody, mouse monoclonal (10B8)

65-165 100 µg

Background: Hemagglutinin (HA) binds to sialic acid-containing receptors on the cell surface, bringing about the attachment of the virus particle to the cell. Plays a major role in the determination of host range restriction and virulence. Class I viral fusion protein. Responsible for penetration of the virus into the cell cytoplasm by mediating the fusion of the membrane of the endocytosed virus particle with the endosomal membrane. Low pH in endosomes induce an irreversible conformational change in HA2, releasing the fusion hydrophobic peptide. Several trimers are required to form a competent fusion pore.

Post-translational modificationⁱ

HAo is consists of 584 amino acids with molecular mass of 63,275. In natural infection, inactive HA is matured into HA1 and HA2. By the sequence similarity it is indicated to be palmitoylated.

Applications

- 1) Western blotting (1/500~1/1,000 dilution)
- 2) Immunofluorescent and Immunocytochemical staining (1/100~1/200 dilution)
- 3) Immunoprecipitation (1/200 dilution)
- 4) Neutralization of infectivity (NT) (assay dependent)
- 5) Hemagglutination Inhibition (HI) (assay dependent)
- 6) ELISA (assay dependent)

Immunogen: Human Influenza B Virus strain Nagasaki/1/87, one of the strains of B/Victoria group.

Specificity: According to Ref.1 during epidemic in Osaka 1996-97, 10B8 antibody reacted with HA protein of all Influenza B virus isolates belonging to Victoria group tested (73 strains) and none of clinical 27 isolates belonging to Yamagata group as examined by PAP staining. It also reacts with Victoria group vaccine strains; Shangdong/7/1997, Malasia/2506/2004. However, note that HA changes during passages and may change reactivity to this antibody. By western blotting, reactivity with B/Malasia/2506/2004 and B/Massachusetts/2/2012 was tested positive.

No cross reactivity with any strains of influenza A viruse.

Isotype: mouse IgG2a, kappa

Purity: Produced in serum-free medium and purified by proprietary

chromatography procedure under mild conditions.



Form: 1 mg/ml in PBS, 50% glycerol, filter sterilized
Storage: Shipped at 4°C or -20°C, and upon arrival, store at -20°C.
Data Link: UniProtKB <u>P03460</u> Influenza B/Lee/1940 HA protein.

References: This antibody was described and used in the following reference.

- 1.Nakagawa N. et al. Rapid detection and identification of two lineages of influenza B strains with monoclonal antibodies. <u>J Virol Methods</u>, 1999;79:113-2 **ICC, IP**
- Nakagawa N. et al. Heterogeneity of Influenza B Virus Strains in One Epidemic Season Differentiated by Monoclonal Antibodies and Nucleotide Sequences. <u>J Clin</u> <u>Microbiol.</u> 2000;38:3467-9. **HI, NT**
- 3. Nakagawa N. et al. Variation of the Conserved Neutralizing Epitope in Influenza B Virus Victoria Group Isolates in Japan. <u>J Clin Microbiol.</u> 2005;43:4212-4. **HI**
- 4. Nakagawa N. et al. Discovery of the neutralizing epitope common to influenza B virus victoria group isolates in Japan. <u>J Clin Microbiol.</u> 2006;44:1564-6. **HI**

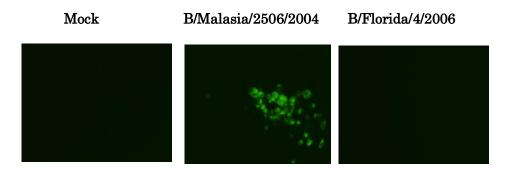


Fig.1 Immunofluorescence assay of MDCK (canine kidney) cells infected with Influenza B virus, using anti-Influenza B virus HA antibody (clone 10B8). Samples were taken at 24 hours post-infection. Anti-Influenza B Virus HA antibody (clone 10B8)) efficiently detected HA in the B/Malasia/2506/2004 virus (Victorial group) but not in B/Florida/4/2006 virus (Yamagata group) infected MDCK cells. The cells were fixed with 4% paraformaldehyde in phosphate-buffered saline (PBS) and permeabilized with 0.1% Triton X-100 in PBS. The bound antibody was visualized by a further reaction with an Alexa Fluor 488-conjugated secondary antibody.

Images on the left are mock-infected MDCK cells as negative control.



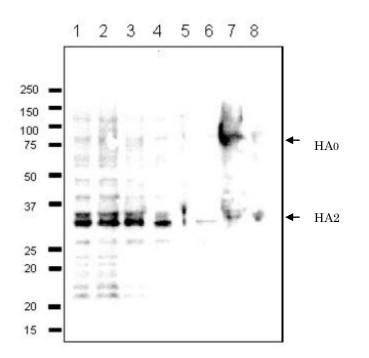


Fig. 2 Detection of HA protein in the crude extracts of MDCK cells infected with various Influenza B virus strains by western blotting using 10B8 monoclonal antibody.

- 1. B/Mie/1/1993
- 2. B/Johannes Burg/5/1999
- 3. B/Florida/4/2006
- 4. B/Lee/1940
- 5. B/Florida/4/2006
- 6. Shandong/7/97
- 7. B/Malasia/2506/2004
- 8. B/Massachusetts/2/2012

First antibody was used at 1/500 dilution and as 2nd antibody, HRP-conjugated goat anti-mouse IgG antibody was used at 1/10,000 dilution. Positions of marker proteins are indicated in kDa on the left. Clone 10B8 recognizes an epitope on HA2 region..