

Datasheet for 200-301-980

H5N1 Antibody VN04-16**Overview**

Description:	H5N1 Antibody (VN04-16) - 200-301-980
Item No.:	200-301-980
Size:	100 µg
Applications:	Neutralization, FC
Reactivity:	Virus
Host Species:	Mouse

Product Details

Background:	Antibody raised against the hemagglutinin (HA) surface glycoprotein of the A/Vietnam/1203/04 (H5N1) influenza virus. Generally referred to as "bird flu", the H5N1 influenza A virus has been documented in poultry and humans across ten Eurasian countries, from Japan in the north to Indonesia in the south. Without immunity, humans would have no protection against H5N1 influenza viruses, which could potentially cause a catastrophic pandemic influenza. This antibody, directed against the HA surface glycoprotein of the A/Vietnam/1203/04 (H5N1) influenza virus, is intended to further our understanding of the mechanisms underlying antigenic variation and evolution of novel variants. The major functions of HA include receptor-binding and fusion activities, but there may also be a structural role for HA in viral particle formation. Following attachment of HA to surface receptors on susceptible cells, the influenza virus enters the cell via endocytosis and membrane fusion.
Synonyms:	mouse anti-H5N1 Antibody, mouse anti-VN04-16 Antibody, H5HA antibody, Hemagglutinin 5 antibody, H5N1 antibody
Host Species:	Mouse
Clonality:	Monoclonal
Clone ID:	18E1
Format:	IgG2a

Target Details

Gene Name:	HA
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Reactivity:	Virus
Immunogen Type:	Native Protein
Immunogen:	This monoclonal antibody was produced by intraperitoneal immunization of BALB/c mice with concentrated purified virus preparation containing hemagglutinin (HA) protein of influenza A virus [strain A/Vietnam/1203/04 (H5N1)] using the modification of the method described by Kohler and Milstein. Each mouse received two immunizations of 15 µg HA with incomplete Freund's adjuvant, administered 3 week apart.
Purity/Specificity:	This product was purified from tissue culture supernatant fluid by Protein A chromatography and is specific for H5 hemagglutinin (HA) protein of influenza A virus [strain A/Vietnam/1203/04 (H5N1)]. VN04-16 monoclonal antibody did not cross-react with influenza viruses of other HA subtypes. This monoclonal antibody reacted with H5N1 influenza viruses representatives of different clades and subclades of the H5 HA subtype.
Relevant Links:	• NCBI - 58618437 • NCBI - 159144921 • UniProtKB - A8UDQ2

Application Details

Tested Applications:	Neutralization
Suggested Applications:	FC (Based on references)
Application Note:	This monoclonal antibody can be used for hemagglutination inhibition (HI) assays to provide antigenic characterization of the influenza A viruses of the H5 HA subtype. This monoclonal antibody is suitable for virus neutralization assays (in cell culture and in embryonated chicken eggs), ELISA, immunoprecipitation, immunohistochemistry and western blotting.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ELISA:	1:5,000
IHC:	User Optimized
IP:	User Optimized
Neutralization:	User Optimized
WB:	User Optimized

Formulation

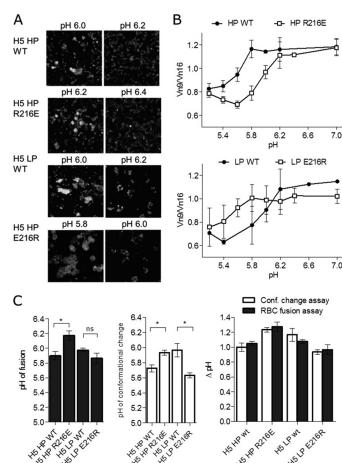
Physical State:	Liquid (sterile filtered)
Concentration:	0.974 mg/mL by UV absorbance at 280 nm

Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	0.01% (w/v) Sodium Azide
Stabilizer:	None

Shipping & Handling

Shipping Condition:	Dry Ice
Storage Condition:	Store vial at -20° C prior to opening. Aliquot contents and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.
Expiration:	Expiration date is one (1) year from date of receipt.

Images

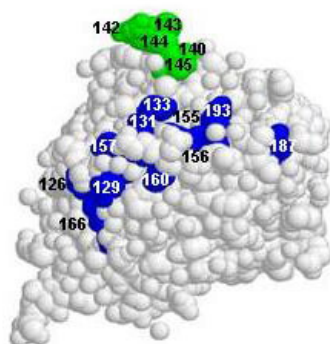


Flow Cytometry

Effect of a mutation of opposite charge at position 216 on the pH dependence of fusion and of conformational change of H5 HP and H5 LP HA proteins, respectively. (A) Representative images of the RBC fusion assay for H5 HP and LP wild-type HAs and respective mutant proteins after incubation at the indicated pH at 37°C (n ≥ 3). (B) pH-dependent conformational change of H5 HP and LP wild-type HAs and mutant proteins as detected by antibody binding and flow cytometry, Vn04-9 (p/n 200-301-977) / Vn04-16 (p/n 200-301-980). Vn04-9/Vn04-16 ratios of median fluorescent intensities were plotted as a function of pH. The average and standard errors are shown (n ≥ 3). (C) Summary of data obtained from the RBC fusion and the conformational change assays illustrated in panels A and B, respectively. The pH of fusion of the wild-type and mutant proteins as assessed by the RBC fusion assay at 37°C corresponds to the highest pH at which full fusion of membranes could still be detected (n ≥ 3). The pH of conformational change corresponds to the pH of 50% change of the antibody Vn04-9/Vn04-16 ratio obtained from median fluorescence intensities. In the right panel, the differences in the pH shift of fusion and conformational change in relation to the highly pathogenic wild type are shown. Error bars represent the standard errors of the means. Statistical difference was determined by an independent t test (*, P < 0.05; ns, nonsignificant). Fig 4. PMID: 25187542

Diagram

Schematic representation of the antigenic sites and the epitopes on the globular head of the HA H5 HA molecule. Images were created with RasMol 2.6, and the HA structure was obtained from the Protein Data Bank (PDB accession number 1JSM). Amino acid positions are designated in H3 numbering. Image provided courtesy of Elena Govorkova Ph D.



References

- Mair CM. et al. A histidine residue of the influenza virus hemagglutinin controls the pH dependence of the conformational change mediating membrane fusion. *J Virol.* (2014)
- Reed ML. et al. Amino acid residues in the fusion peptide pocket regulate the pH of activation of the H5N1 influenza virus hemagglutinin protein. *J Virol.* (2009)
- Kaverin NV. et al. Epitope Mapping of the Hemagglutinin Molecule of a Highly Pathogenic H5N1 Influenza Virus by Using Monoclonal Antibodies. *ASM Journals Journal of Virology* (2007)

Disclaimer

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