

12-8491: Anti-Respiratory Syncytial Virus (Clone: RSV-14N4)-Biotin

Clonality :	Monoclonal
Clone Name :	RSV-14N4
Application :	ELISA
Alternative Name :	RSV, Orthopneumovirus
Isotype :	Human IgG1k

Description

Specificity: RSV-14N4 activity is directed against antigenic site II of the RSV fusion (F) protein. RSV-14N4 readily competes with clone RSV-12I1 on post-fusion F, but the competition is less pronounced on prefusion F. A plaque reduction neutralization assay showed RSV-14N4 is capable of neutralizing RSV strain A2. By ELISA RSV-14N4 binds to both prefusion and post-fusion F proteins with equal affinity. Competition-binding studies showed that RSV-14N4 targets antigenic site II, which is the target of palivizumab, an antiviral monoclonal antibody used as a prophylactic treatment. Saturation alanine scanning mutagenesis identified residues Asp263, Ile266, Asp269, and Lys271 as critical for 14N4 binding. Binding to antigenic site II was confirmed by x-ray crystallography and electron microscopy as well as by binding to scaffolded epitopes containing site II.

Antigen Distribution: F protein is a surface glycoprotein.

Background: Respiratory syncytial virus (RSV) is a common respiratory virus that infects the majority of children by two years old^{1, 2}. While usually mild, RSV can be serious in infants and older adults and is the leading cause of bronchiolitis and pneumonia in children less than one year of age in the United States¹. Antibodies have been described that bind and neutralize RSV fusion (F) protein²⁻⁴. RSV F protein is a type I integral membrane protein that is synthesized as a 574 amino acid inactive precursor, assembled into a trimer, post-translationally modified, then cleaved to produce F1, F2, and intervening peptide pep273. Functional F protein has both pre- and post-fusion conformations. RSV F protein is highly conserved among RSV isolates from both A and B subgroups³ and is the primary target for antiviral drug development³ with several antigenic regions capable of introducing neutralizing antibodies². RSV-14N4 was generated from human donors using human hybridoma technology⁴. Transformed B cells were screened by ELISA against recombinant post-fusion RSV A2 F protein and isolated by single-cell flow cytometric sorting. A plaque reduction neutralization assay showed RSV-14N4 is capable of neutralizing RSV strain A2. An ELISA binding assay showed that RSV-14N4 binds to both prefusion and post-fusion F proteins with equal affinity. Competition-binding studies showed that RSV-14N4 targets antigenic site II, which is the target of palivizumab, an antiviral monoclonal antibody licensed as a prophylactic treatment. Saturation alanine scanning mutagenesis identified residues Asp263, Ile266, Asp269, and Lys271 as critical for 14N4 binding. Binding to antigenic site II was confirmed by x-ray crystallography and electron microscopy as well as by binding to scaffolded epitopes containing site II. RSV-14N4 readily competes with RSV-12I1 on post-fusion F, but the competition is less pronounced on prefusion F. The RSV-14N4 isotype is IgG1.

Product Info

Amount :	100 µg / 250 µg Purity : ≥95% monomer by analytical SEC
Purification :	Preparation : Recombinant antibodies are manufactured in an animal free facility using only in vitro protein free cell culture techniques and are purified by a multi-step process including the use of protein A or G to assure extremely low levels of endotoxins, leachable protein A or aggregates.

Content :

Concentration: 0.5 mg/ml

Formulation: This recombinant monoclonal antibody is aseptically packaged and formulated in 0.01 M phosphate buffered saline (150 mM NaCl) PBS pH 7.2 - 7.4 with no carrier protein, potassium, calcium or preservatives added. Due to inherent biochemical properties of antibodies, certain products may be prone to precipitation over time. Precipitation may be removed by aseptic centrifugation and/or filtration.

Storage condition :

Functional grade preclinical antibodies may be stored sterile as received at 2-8°C for up to one year. For longer term storage, aseptically aliquot in working volumes without diluting and store at $\geq -70^{\circ}\text{C}$. Avoid Repeated Freeze Thaw Cycles.

Application Note

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