Description:	This purified rabbit monoclonal antibody recognizes recombinant wildtype SARS-CoV-2 Spike RBD and trimeric spike proteins. Additionally, this antibody cross-reacts with the Delta variant (B.1.617.2) Spike S1 and trimeric proteins and with the Delta Plus variant (B.1.617.2.1) trimer. This antibody also cross reacts with variant Alpha (B.1.1.7; U.K.), Beta (B.1.351; South African), and Gamma (P.1; Brazilian) Spike RBD and trimeric proteins. This neutralizing antibody impedes the interaction between SARS-CoV-2 Spike and human ACE2 receptor. ACE2 receptor is known to mediate COVID-19 infection through direct binding of the SARS-CoV2 Spike protein. This neutralizing antibody has been functionally tested using BPS Bioscience Spike RBD: ACE2 Inhibitor Screening Kits and using Spike pseudotyped lentiviruses in cell-based assays.
Concentration:	0.5 mg/ml
Species:	Rabbit
Clone:	C-A11
Isoform:	IgG
Immunogen:	SARS-CoV-2 Spike RBD
Formulated In:	PBS
Purification:	Protein A affinity chromatography
Format:	Aqueous buffer solution
Storage:	4°C
MW:	150 kDa
Assay Conditions:	*Experimental design and assay protocol for measuring the neutralizing activity of the antibody using Spike Trimer (S1+S2) (B.1.1.7 Variant) (SARS-CoV-2): ACE2 Inhibitor Screening Colorimetric Assay Kit, BPS Bioscience #78175.* 1. Coat a flat bottom clear 96-well plate with 1 µg/ml spike protein diluted in PBS overnight at 4°C. 2. The next day, dilute one volume of 3x Immuno Buffer 1 (BPS Bioscience #79311) with 2 volumes of distilled water to bring it to 1x Immuno Buffer 1. Wash three times with 100 ul of 1x Immuno Buffer 1. Then, block with 100 ul of Blocking Buffer 2 (BPS Bioscience #79728) for 1 hour at room temperature with gentle shaking. 3. Wash with 1x Immuno Buffer 1 and add serial dilutions of neutralizing antibody (suggested initial range: 100 nM to pM range) to the plate containing the coated spike protein, for 30 minutes at room temperature with gentle shaking. 4. Without removing the antibody, add 100 ul of ACE2-biotin (BPS Bioscience, #100665) diluted in Blocking Buffer 2 at a final concentration of 1.5 ng/ul. Incubate for 1 hour at room temperature with gentle shaking. 5. Wash three times with 1x Immuno Buffer 1 and add HRP-Streptavidin diluted in Blocking Buffer 2. 6. Wash three times with 1x Immuno Buffer 1 and add 100 µl of Colorimetric HRP substrate. Quench the reaction with 100 µl of 1N HCL after color development. 6. Read absorbance at 450 nm.

Figure 1A.

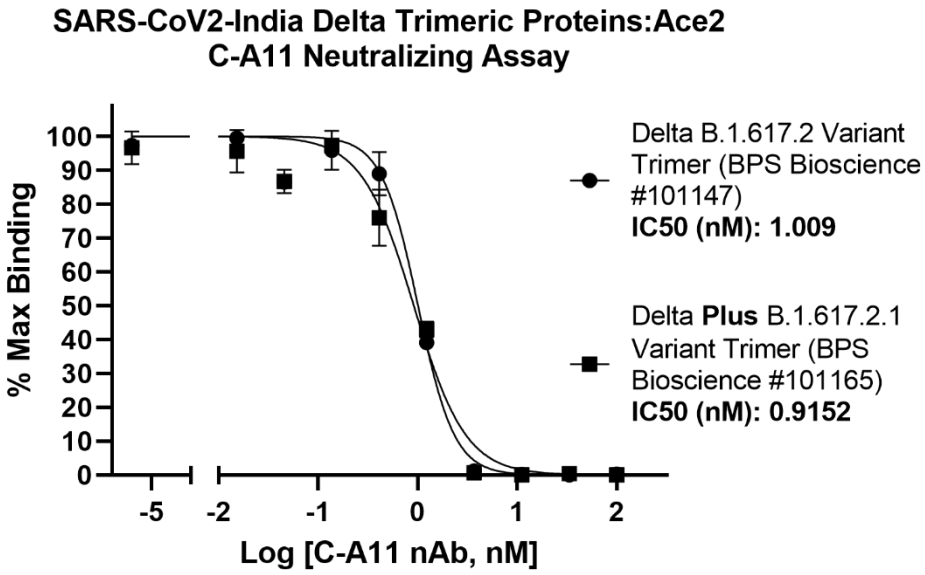

Figure 1B.

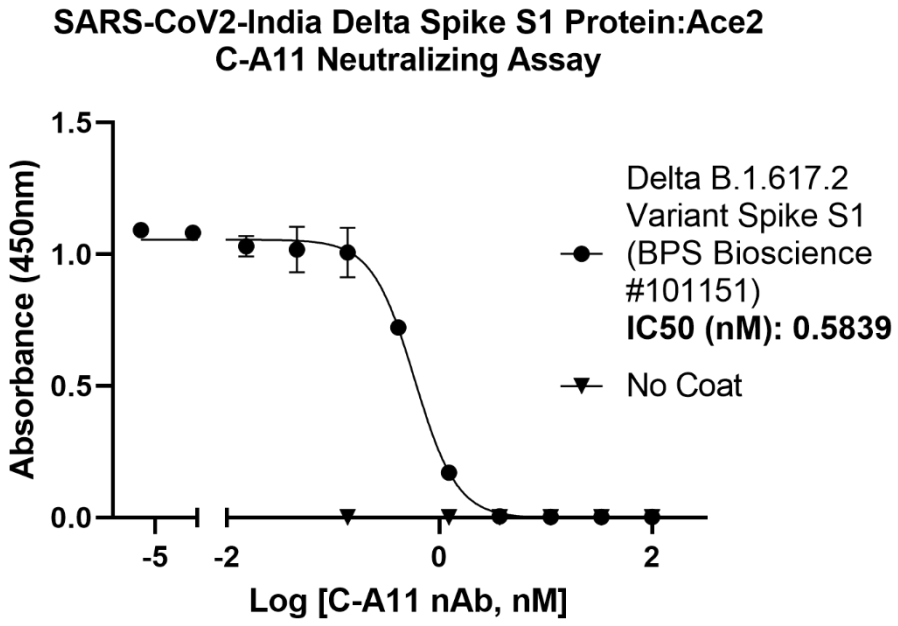

Figure 1C.

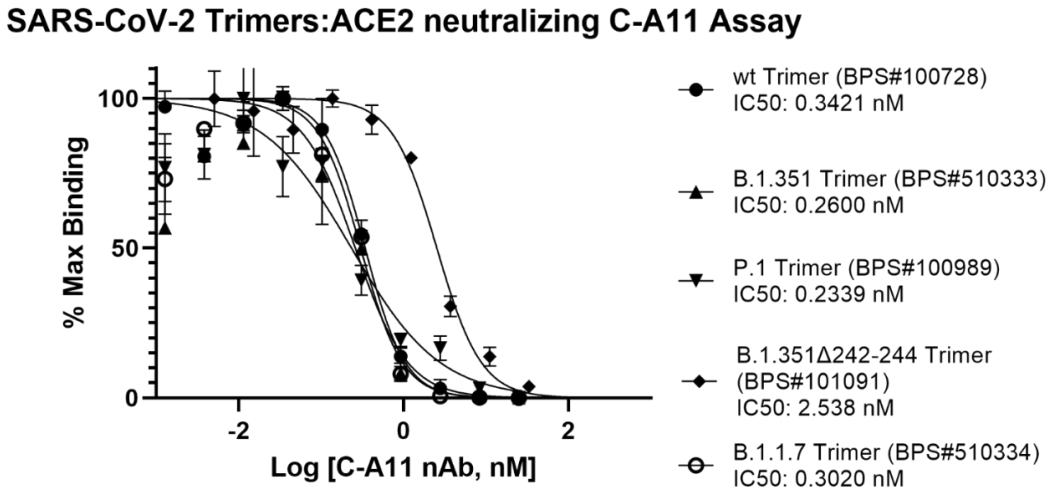

Figure 1D.

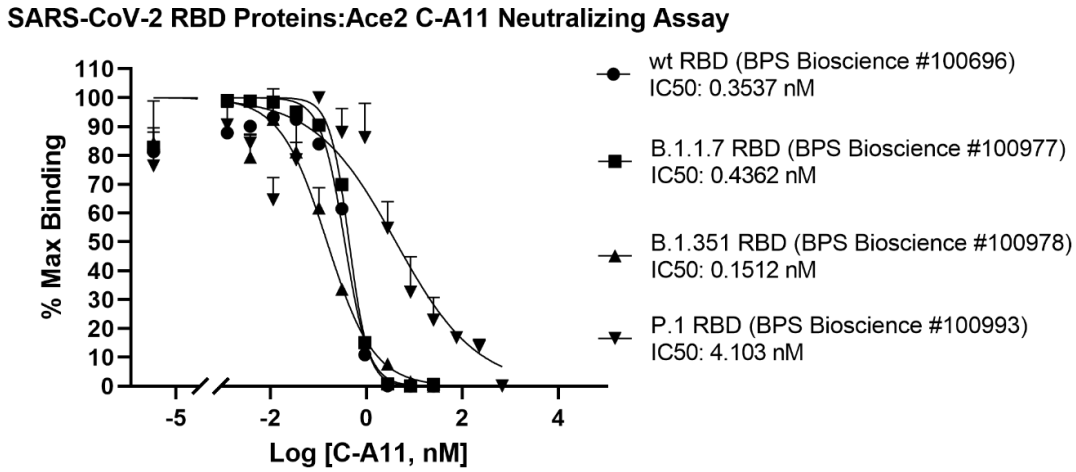

Figure 1. Neutralization of the interaction between spike protein variants and human ACE2

Clone C-A11 Neutralizing antibody competes with and blocks the binding of ACE2-biotin to immobilized recombinant SARS-CoV-2 Spike proteins (1 µg/ml) following the assay conditions described above. This neutralizing activity was empirically tested in a colorimetric ELISA assay with the following target SARS-CoV-2 variant proteins:

A) India Delta B.1.617.2 Trimer (BPS Bioscience #101147) and India Delta Plus B.1.617.2.1 Trimer (BPS Bioscience #101165)

B) Emerging India Delta B.1.617.2 Spike S1 Protein (BPS Bioscience #101151)

C) Spike trimers wild-type (BPS Bioscience #100728), B.1.351 (BPS Bioscience #510333), P.1 (BPS Bioscience #100989), B.1.351 Δ242-244 (BPS Bioscience #101091), and B.1.1.7 (BPS Bioscience #510334)

D) Wild-type spike RBD (BPS Bioscience #100696), B.1.1.7 RBD (BPS Bioscience #100977), B.1.351 RBD (BPS Bioscience #100978), and P.1 RBD (BPS Bioscience #100993)

*This data is representative, please inquire for lot-specific information

Figure 2

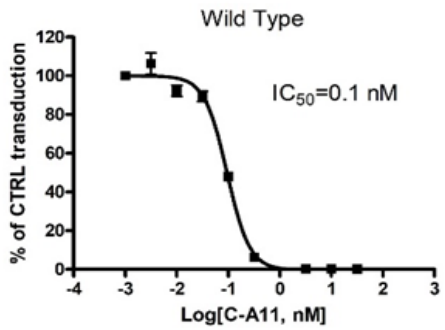

Figure 2A

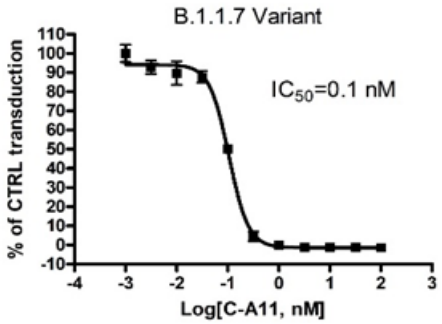

Figure 2B

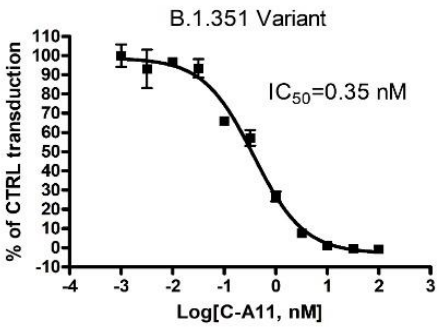

Figure 2C

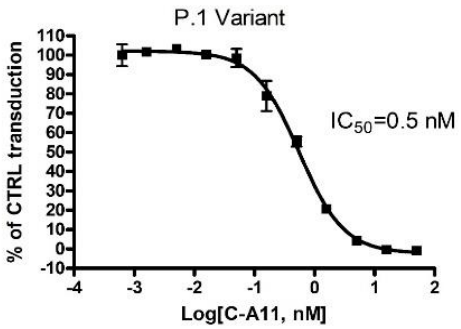

Figure 2D

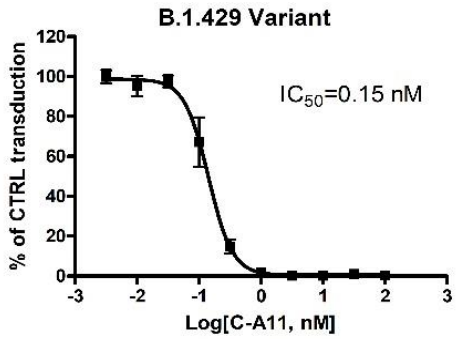

Figure 2E

Figure 2: Cell-based neutralization assay using anti-SARS-CoV-2 Spike antibody (clone# C-A11)
Approximately 8,000 ACE2-HEK293 cells/well were transduced with 10 µl/well of Spike (SARS-CoV-2) Pseudotyped Lentivirus (Luc reporter)/anti-Spike antibody mix. After 48 hours of transduction, ONE-Step Luciferase reagent (BPS Bioscience #60690) was added to the cells to measure the luciferase activity, indicative of the transduction efficiency. The transduction efficiency measured in the wells containing only the pseudovirus (no antibody treatment) was set as 100%, while the transduction efficiency measured in the wells without virus was set as 0%.
A. Spike (SARS-CoV-2) Pseudotyped Lentivirus (Luc reporter) (BPS Bioscience #79942);
B. Spike (B.1.1.7 Variant) (SARS-CoV-2) Pseudotyped Lentivirus (Luc reporter) (BPS Bioscience #78112);
C. Spike (B.1.351 Variant) (SARS-CoV-2) Pseudotyped Lentivirus (Luc reporter) (BPS Bioscience #78142);
D. Spike (P.1 Variant) (SARS-CoV-2) Pseudotyped Lentivirus (Luc reporter) (BPS Bioscience #78144);
E. Spike (B.1.429 Variant) (SARS-CoV-2) Pseudotyped Lentivirus (Luc reporter) (BPS Bioscience #78172)