TWIST BIOPHARMA

Advancing Novel SARS-CoV-2 Therapeutic Antibodies

November 2020

@TwistBioscience #WeMakeDNA

Making Progress on a Global Need

Novel Therapeutic Antibody Leads	Well Characterized	Well Validated
IgG (TB181-8, 28, 36) and VHH (TB201-202)	High affinity and unbiased, leveraging Twist's synthetic DNA platform	Extensive pseudovirus and live virus data

Our DNA Platform Has Enabled a Rapid Discovery Process

Dual Focus

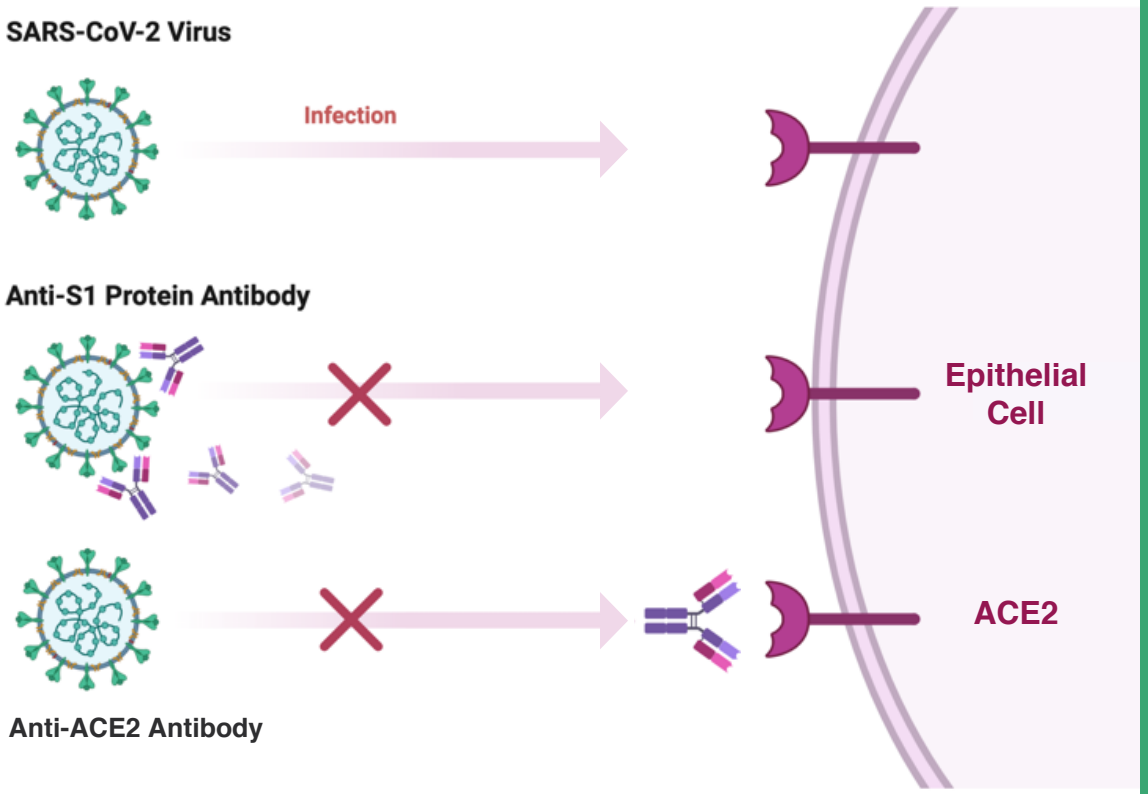
TWIST

#1

Spike Protein Neutralizing Ab

#2

Anti-ACE2 Blocking Ab



Synthetic Libraries

VANDERBILT UNIVERSITY MEDICAL CENTER

Based on Sequence of COVID-19 Survivor

Human Fab and VHH Libraries

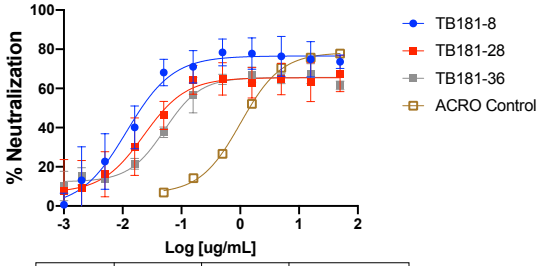
Fast Results

200+ antibody leads in 6 weeks

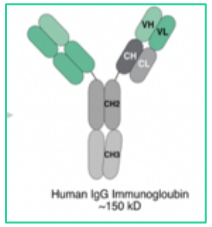
TB181 – High Affinity Anti-S1 IgG Antibodies

Affinity & Pseudovirus

	S1 K_D	S Trimer K_D
TB181-8	6.1 nM	11 nM
TB181-28	364 nM	11 nM
TB181-36	10 nM	18 nM

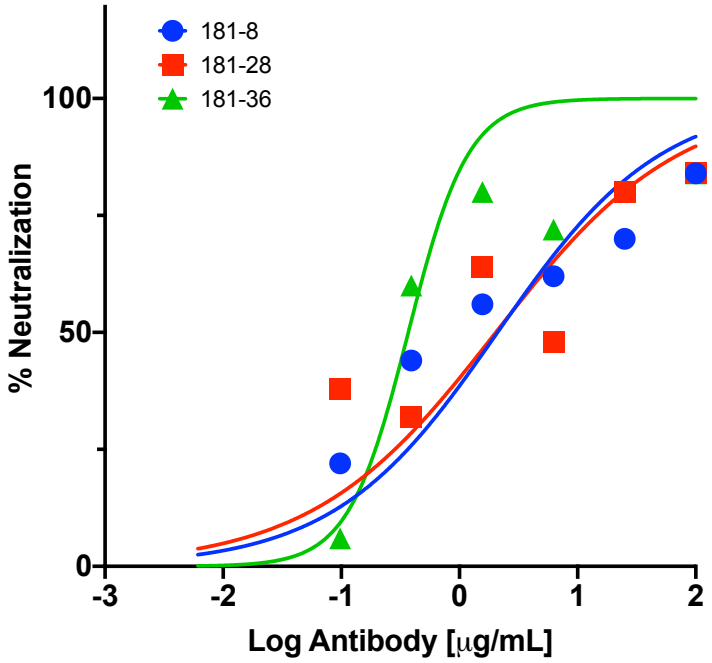


TB181-8	TB181-28	TB181-36	ACRO Control
0.01170	0.02293	0.05196	1.011



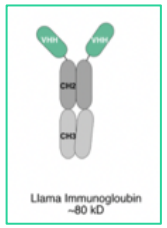
Live Virus FRNT

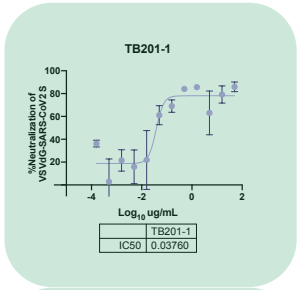
Conducted by Dr. James Brien, Saint Louis University

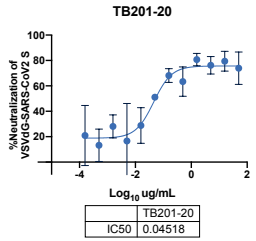


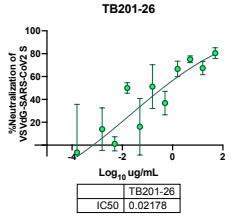
Antibody	EC50 (ug/mL)
181-36	0.37
181-28	2.03
181-8	2.10

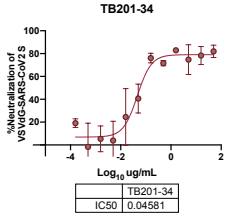
TB201-202 – High Affinity Anti-S1 VHH Single Domain Antibodies
VSV-pseudotype SARS-CoV2 Neutralization Analysis

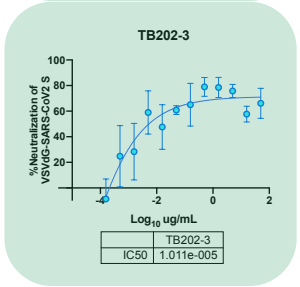


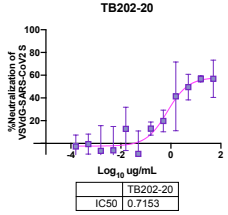


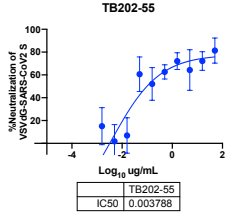


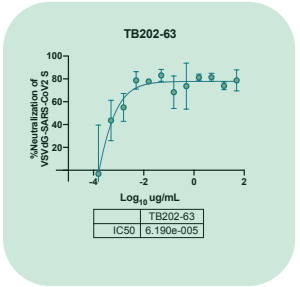


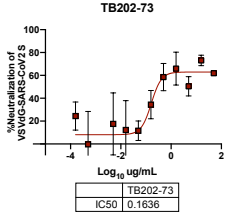


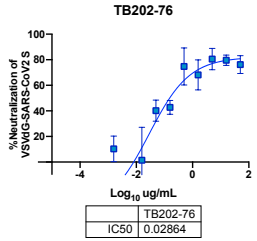


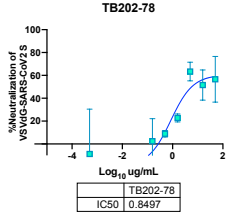


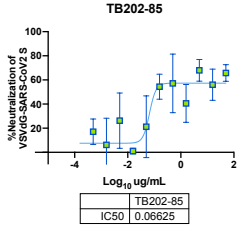












TB201-1, TB202-3 and TB202-63 are top candidates from pseudovirus testing

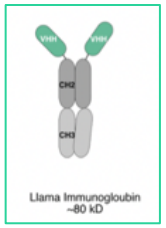
VHH Single Domain Leads (TB201-202) Show Higher Neutralization vs. IgG in Live SARS-CoV-2 Virus FRNT

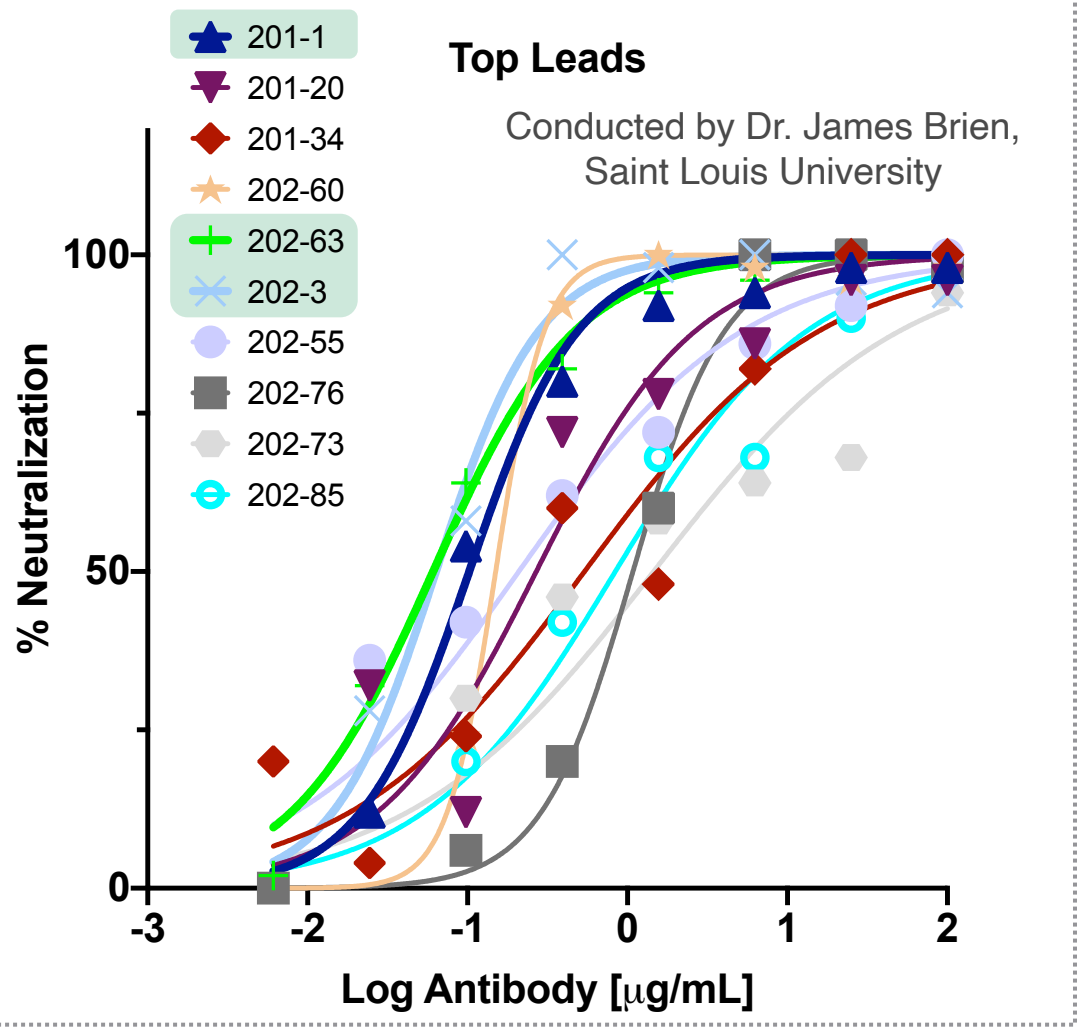
TB202-63, TB202-3, & TB201-1 show potent neutralization in live virus FRNT

IgG

VHH

Antibody	NC50 (ug/mL)
202-63	0.06
202-3	0.06
201-1	0.10
202-60	0.15
202-55	0.21
201-20	0.27
181-36	0.37
201-34	0.54
202-85	0.84
202-76	1.08
202-73	1.46
181-28	2.03
181-8	2.10
202-26	2.97
202-20	5.03
202-78	8.26
182-7	11.77
182-3	18.31
182-4	67.57
181-63	106.90



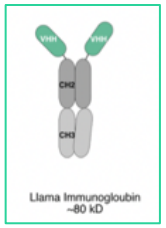


VHH Single Domain Leads (TB201-202) Show Potent Neutralization in Live SARS-CoV-2 Virus PRNT

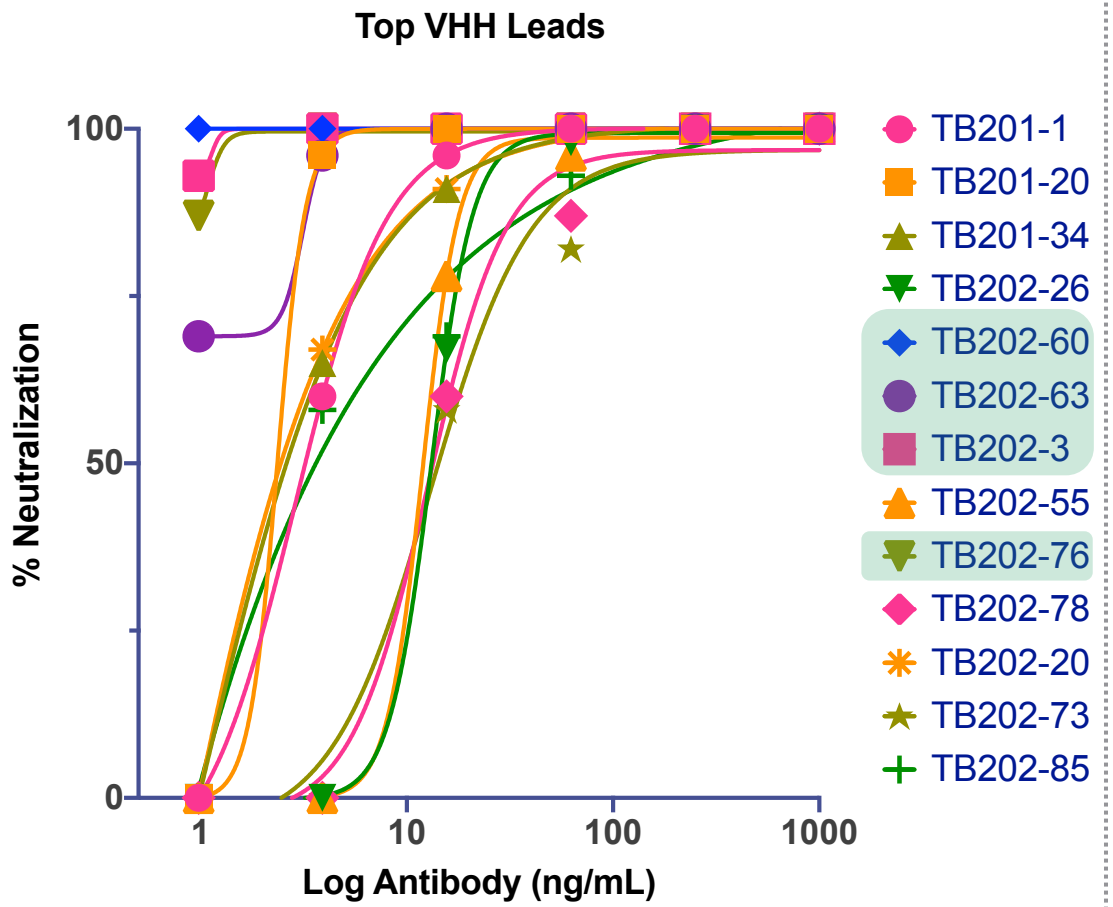
TB202-3, TB202-60, TB202-63, & TB202-76 show potent neutralization in live virus PRNT

Antibody	PRNT90 (ng/mL)*
TB201-1	15.6
TB201-20	3.9
TB201-34	15.6
TB202-26	62.5
TB202-60	<0.98
TB202-63	3.9
TB202-3	<0.98
TB202-55	62.5
TB202-76	3.9
TB202-78	250
TB202-20	15.6
TB202-73	250
TB202-85	62.5

* The antibody concentration required to reduce the number of plaques by 90% compared to free virus



Conducted by Dr. Richard Bowen, Colorado State University

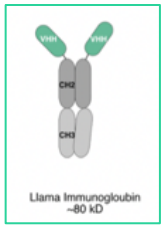


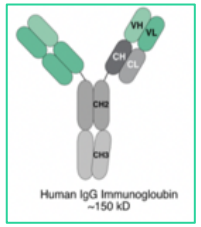
VHH Single Domain Leads (TB201-202) and TB181-36 Show Potent Neutralization in Live SARS-CoV-2 Virus PRNT

TB202-3, TB202-63, & TB181-36 show potent neutralization in live virus PRNT

Antibody	EC80 (ug/mL)
TB202-63 11429	0.057861
TB202-3 11435	0.115234
TB181-36 10953	0.171875
TB202-60 11428	0.236816
TB201-20 11433	0.376
TB202-76 11443	0.425781
TB202-55 11441	0.445313
TB201-34 11416	0.570313
TB202-42 11426	0.734375
TB201-1 11431	0.810547
TB181-8 10717	0.855469
TB202-85 11445	1.0625
TB201-38 11418	1.5
TB201-67 11434	1.566406
TB181-28 11237	1.78125
TB202-73 11442	2.015625
TB202-20 11437	2.3125
TB201-47 11419	2.457031
TB181-40 10955	2.78125
TB202-44 11440	3.15625

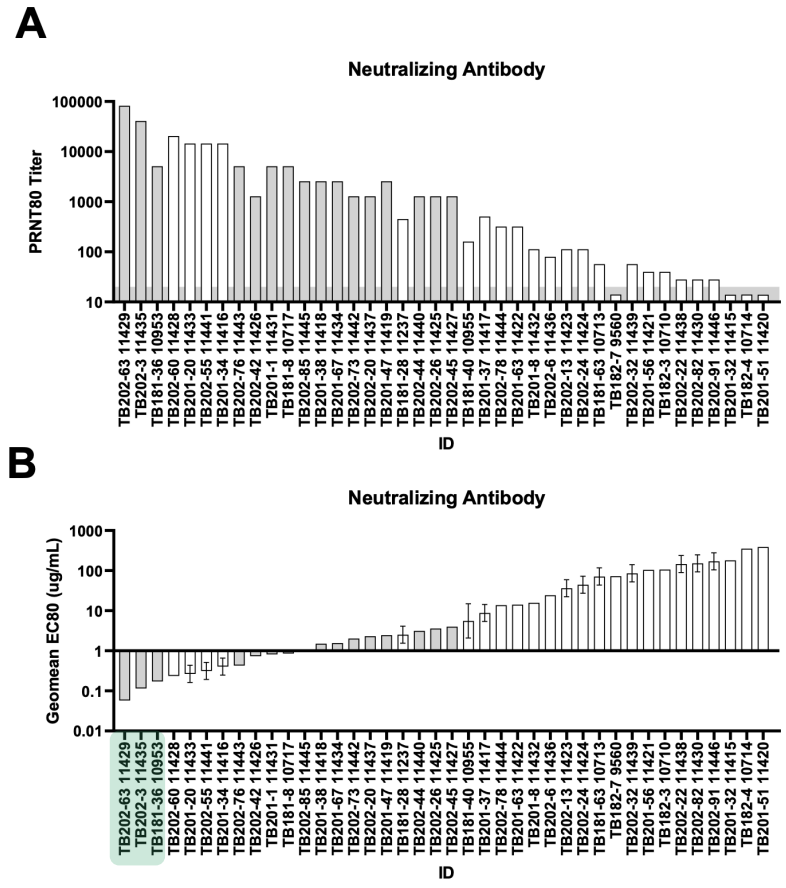
Antibody	EC80 (ug/mL)
TB202-26 11425	3.609375
TB202-45 11427	4.015625
TB201-37 11417	12.4375
TB202-78 11444	13.75
TB201-63 11422	14.1875
TB201-8 11432	15.8125
TB202-6 11436	24.375
TB202-13 11423	25.6875
TB202-24 11424	31.375
TB181-63 10713	50.5
TB202-32 11439	60.625
TB182-7 9560	72.34043
TB202-22 11438	103.25
TB201-56 11421	104.75
TB182-3 10710	106.5
TB202-82 11430	107.75
TB201-32 11415	180.1418
TB202-91 11446	240.5
TB182-4 10714	354.6099
TB201-51 11420	387.9433





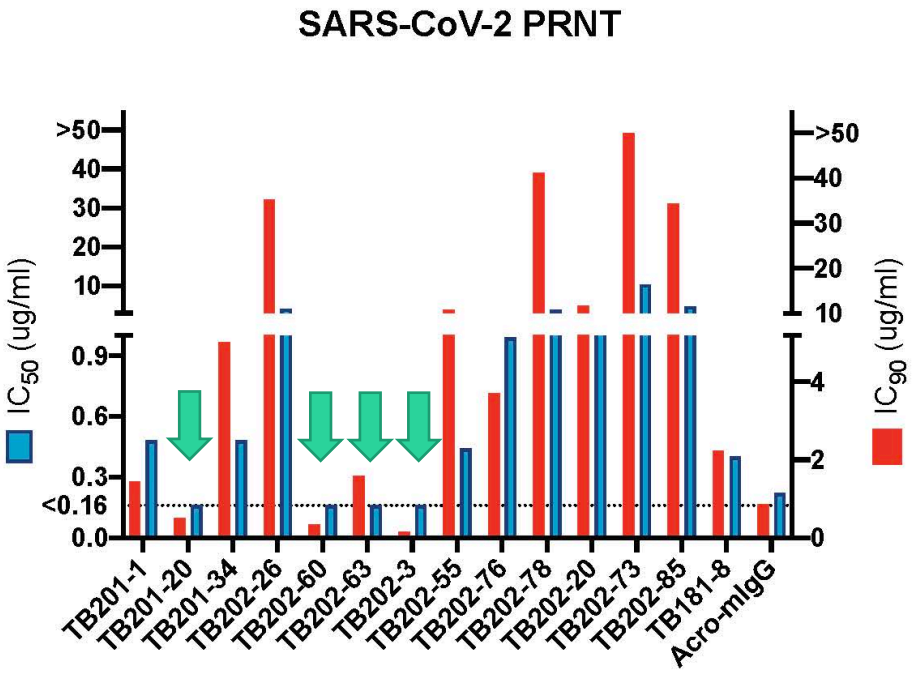
* The antibody concentration required to reduce the number of plaques by 80% compared to free virus

Conducted by Dr. Jay Hooper, USAMRIID

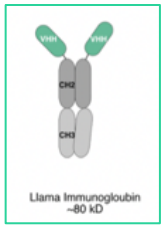


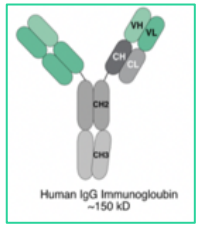
VHH Single Domain Leads (TB201-202) Show Potent Neutralization in Live SARS-CoV-2 Virus PRNT

TB201-20, TB202-60, TB202-63 and TB202-3 show potent neutralization in live virus PRNT



* The antibody concentration required to reduce the number of plaques by 50% or 90% compared to free virus





Pseudovirus and Live Virus
Conducted by IBT Bioservices

	Pseudovirus Neutralization Testing		SARS-CoV-2 PRNT	
mAB	NT50 (ug/mL)	%Neut recorded at highest concentration tested (i.e. 50 ug/mL)	IC50 (ug/mL)	IC90 (ug/mL)
TB201-1	0.0376	85.92% ±4.25	0.48	1.44
TB201-20	0.04518	73.88% ±12.82	<0.16	0.50
TB201-34	0.04581	82.02% ±5.43	0.48	5.01
TB202-26	0.02178	80.53% ±4.64	4.15	35.19
TB202-60	0.01467	77.58% ±7.25	<0.16	0.34
TB202-63	0.00104	78.71% ±9.23	<0.16	1.58
TB202-3	0.007	66.14% ±11.79	<0.16	0.16
TB202-55	0.158	81.38% ±11.04	0.44	10.85
TB202-76	0.02864	76.21% ±6.94	0.99	3.70
TB202-78	0.8497	56.6% ±19.95	3.79	41.11
TB202-20	0.7153	56.83% ±16.48	1.49	11.68
TB202-73	0.1636	61.89% ±1.03	10.42	>50
TB202-85	0.06625	65.8% ±6.9	4.72	34.28
TB181-8	0.01941	73.6% ±3.4	0.40	2.23

Cyclophosphamide-Syrian Hamster Model (Intranasal Virus Delivery)

Disruption of Adaptive Immunity Enhances Disease in SARS-CoV-2-Infected Syrian Hamsters

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ABSTRACT Animal models recapitulating human COVID-19 disease, especially severe disease, are urgently needed to understand pathogenesis and to evaluate candidate vaccines and therapeutics. Here, we develop novel severe-disease animal models for COVID-19 involving disruption of adaptive immunity in Syrian hamsters. Cyclophosphamide (CyP) immunosuppressed or *RAG2* knockout (KO) hamsters were exposed to severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) by the respiratory route. Both the CyP-treated and *RAG2* KO hamsters developed clinical signs of disease that were more severe than those in immunocompetent hamsters, notably weight loss, viral loads, and fatality (*RAG2* KO only). Disease was prolonged in transiently immunosuppressed hamsters and was uniformly lethal in *RAG2* KO hamsters. We evaluated the protective efficacy of a neutralizing monoclonal antibody and found that pretreatment, even in immunosuppressed animals, limited infection. Our results suggest that functional B and/or T cells are not only important for the clearance of SARS-CoV-2 but also play an early role in protection from acute disease.

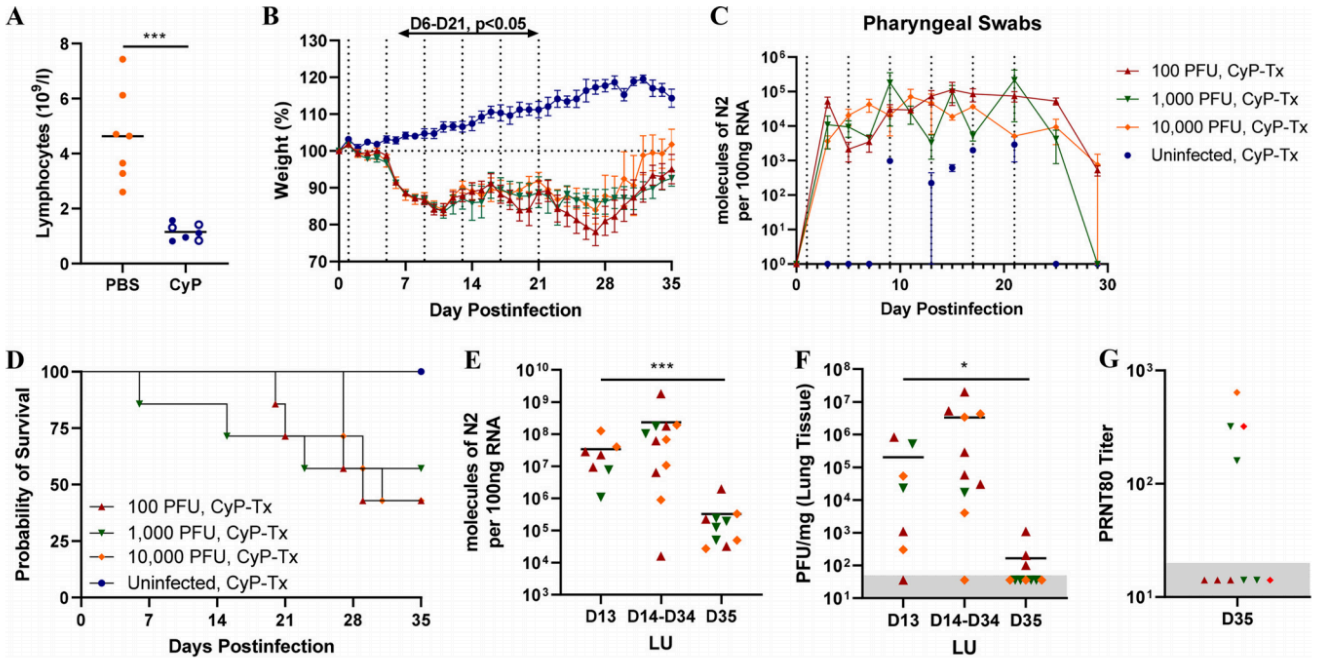
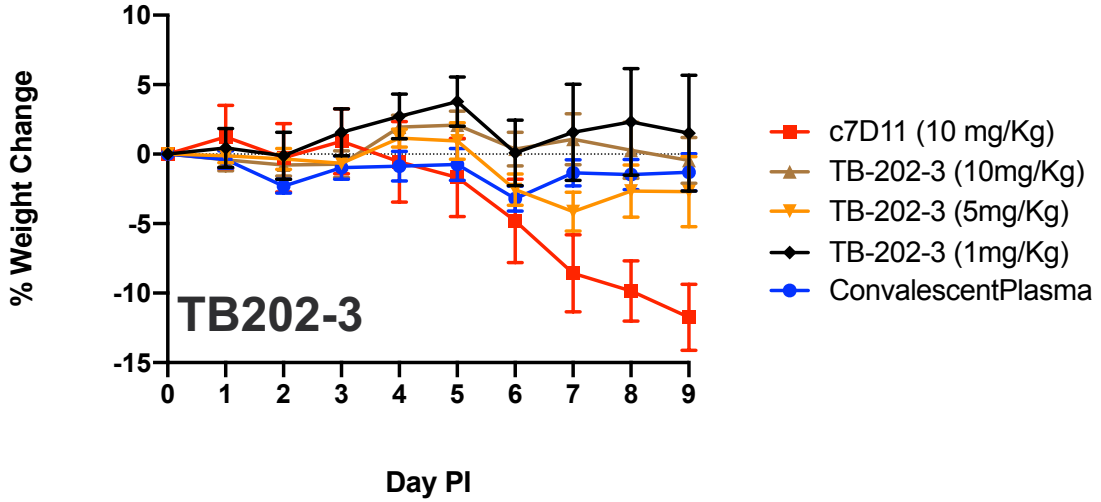


FIG 1 CyP-treated, SARS-CoV-2-infected hamsters. Groups of 10 Syrian hamsters each were immunosuppressed with CyP. (A) Lymphocyte counts were determined from whole blood 3 days (closed symbols) or 4 days (open symbols) following the CyP loading dose. Hamsters were exposed to increasing doses of SARS-CoV-2 intranasally on day 0. (B) Weights were monitored for 35 days. (C) Viral RNA copies per pharyngeal swab were assayed at the indicated times postinfection. CyP administration is depicted by vertical dotted lines in panels B and C. Hamsters were monitored for (D) survival, and lung tissue collected either at the time of death or scheduled euthanasia 13 or 35 dpi was assessed for viral load by (E) Reverse transcription-PCR (RT-PCR) and (F) plaque assay. (G) Blood was collected from surviving hamsters at 35 dpi and assessed by plaque reduction or neutralization test. LU, lung.

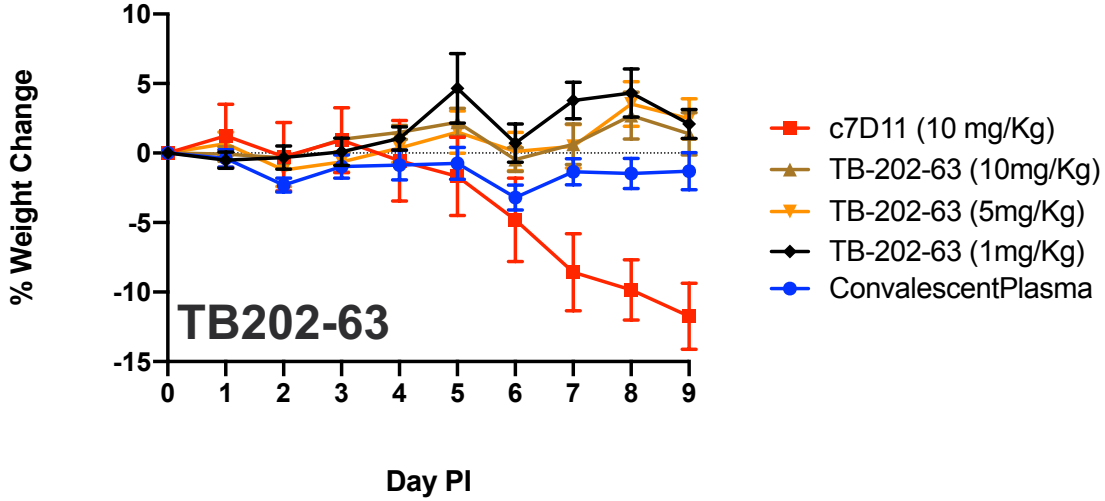
Transiently immunosuppressed (CyP every 4 days until Day 9) hamsters are more susceptible to SARS-CoV-2: greater weight loss, some mortality, nasal turbinate and lung pathology.

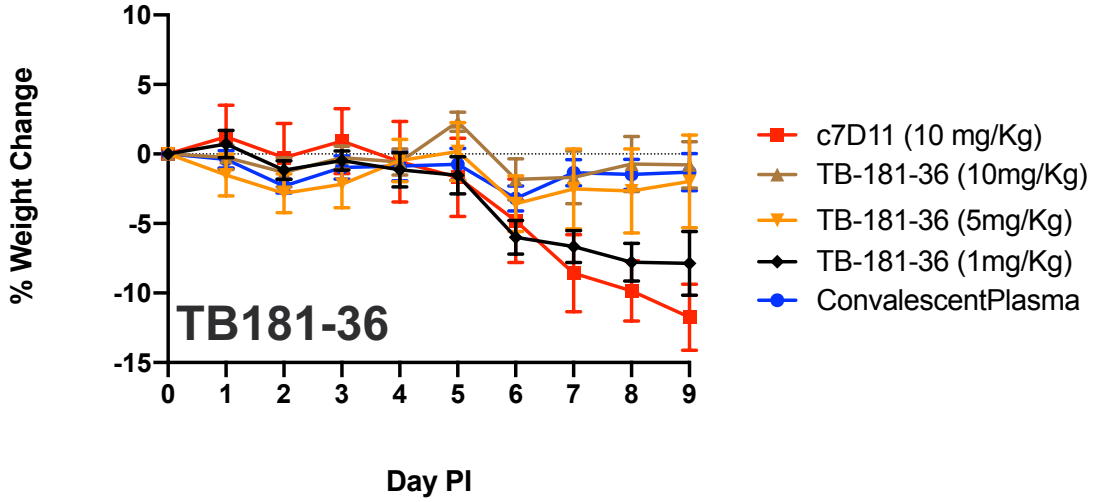
VHH Single Domain Leads (TB202-3, 63) and IgG Lead (TB181-36) Show Potent *In Vivo* Activity in Cyclophosphamide-Syrian Hamster Model

TB202-3 and TB202-63 protect against weight loss at the lowest dose of 1 mg/kg!



c7D11 = negative control mAb
ConvalescentPlasma = positive control





Promising Development Potential

Path to Development

- Hamster *in vivo* Protection Studies
- Multivalent homo- and heterotrimeric VHH Fc Designs to increase potency
- Identify partner for rapid antibody scale-up of clinical supply

Next Steps