

Isolation of potent SARS-CoV-2 neutralizing antibodies and protection from disease in a small animal model

Rogers TF, Zhao F, Huang D, Beutler N, Burns A, He WT, Limbo O, Smith C, Song G, Woehl J, Yang L, Abbott RK, Callaghan S, Garcia E, Hurtado J, Parren M, Peng L, Ramirez S, Ricketts J, Ricciardi MJ, Rawlings SA, Wu NC, Yuan M, Smith DM, Nemazee D, Teijaro JR, Voss JE, Wilson IA, Andrabi R, Briney B, Landais E, Sok D, Jardine JG, Burton DR. Isolation of potent SARS-CoV-2 neutralizing antibodies and protection from disease in a small animal model. *Science*. 2020 Aug 21;369(6506):956-963. doi: 10.1126/science.abc7520.

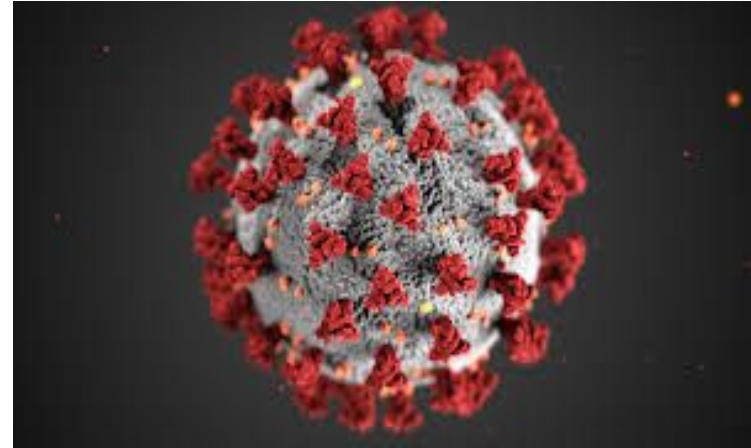
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Outline

- The state of the current troubles: SARS-CoV-2
- Neutralizing antibodies to the cause of SARS-CoV-2 can help guide vaccine design
- SARS-CoV-2 neutralizing antibody isolation strategy
- Cohort study using CC6, CC12, and CC25 showed varied responses to SARS-CoV RBD and S
- Binding antibodies show neutralization activity between RBD/S and S binders
- Functional activity of down-selected antibodies
- Outcome of passive transfer & challenge in Syrian hamsters
- Efficacy of RBD nAb in vivo in Syrian hamsters shows promise for human studies

The state of the current troubles: SARS-CoV-2

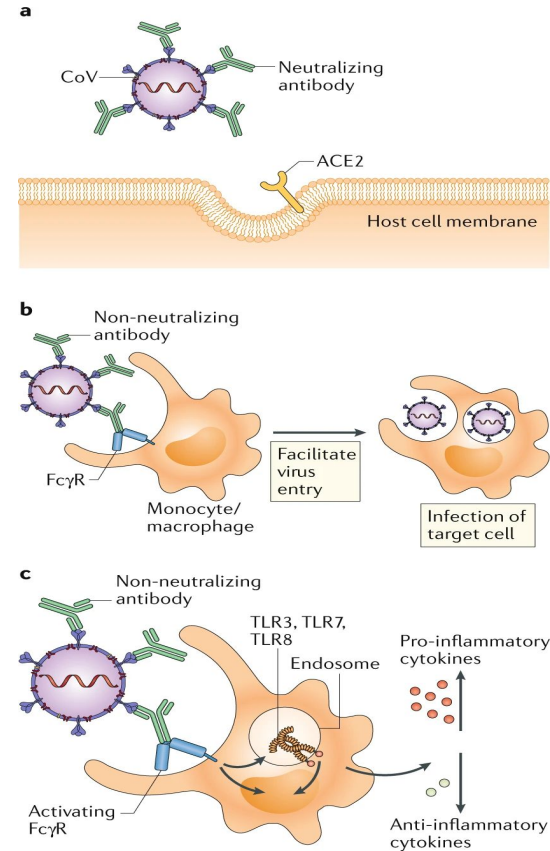
- SARS-CoV-2 emerged from Wuhan, China and poses a global threat to humans
 - Symptoms are similar to those caused by SARS-CoV
 - Susceptibility to SARS-CoV is largely determined by the affinity of viral RBD and ACE2 receptor of the host
- SARS-CoV-1 and SARS-CoV-2 share ACE2 receptor
 - ACE2 regulates cross-species transmission as well as human to human transmission



(CDC, 2020)

Neutralizing antibodies to the cause of SARS-CoV-2 can help guide vaccine design

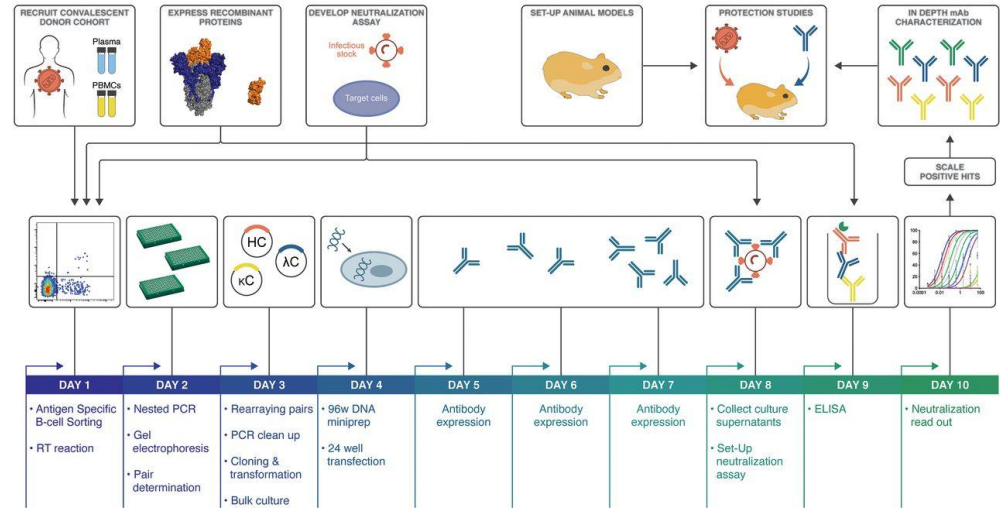
- Enrolled recovered SARS-CoV-2 patients
- Isolated potent neutralizing antibodies (nAbs)
 - Two epitopes on the RBD and non-RBD epitopes on the spike protein
- Found an nAb to SARS-CoV-2 and demonstrated its efficacy in small animals
 - Likely useful in medical interventions in humans
- nAbs they used has a potential role in prophylaxis
 - Could be a form of therapy for SARS-CoV-2



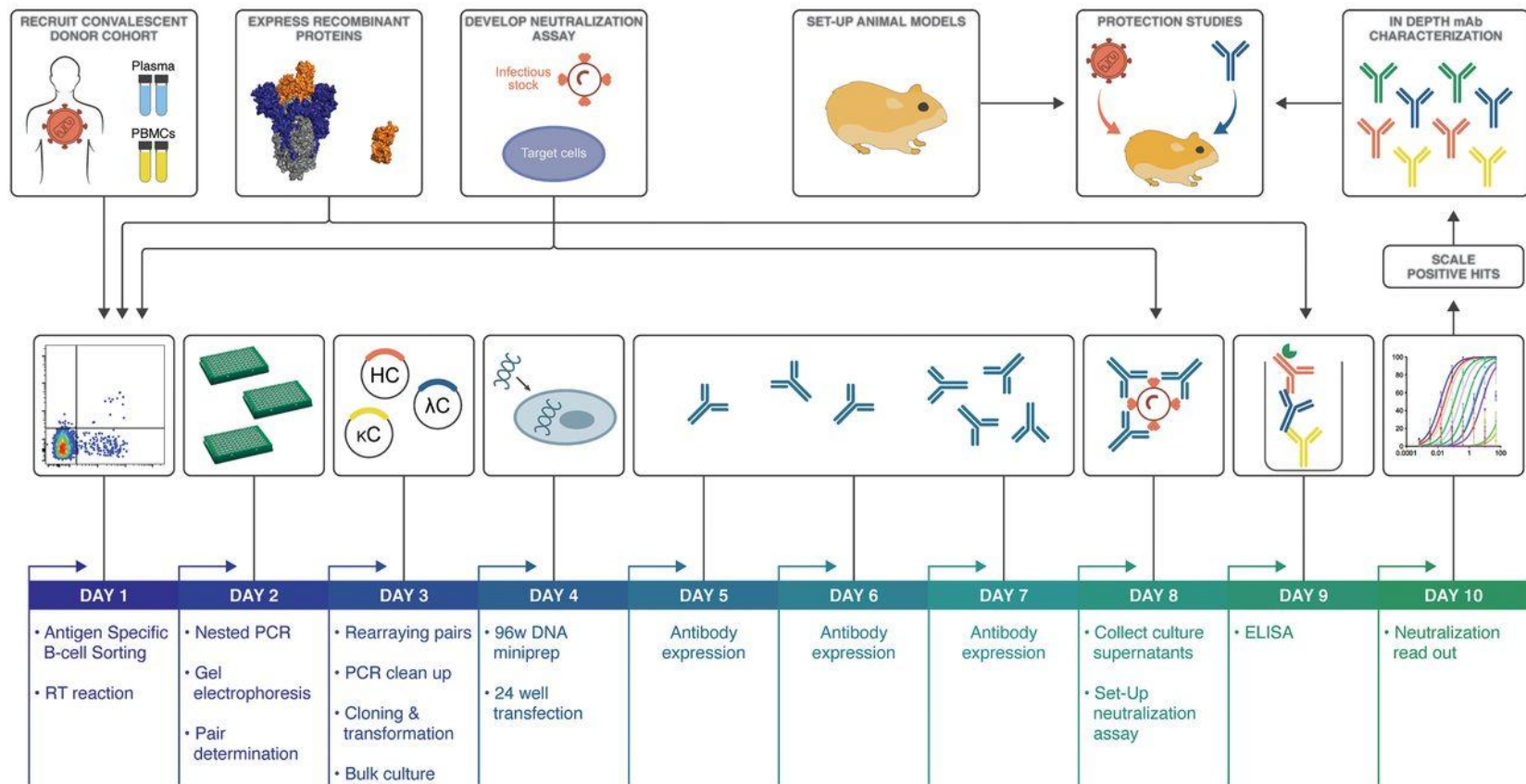
Mechanism of neutralizing antibodies (Iwasaki and Yang, 2020)

SARS-CoV-2 neutralizing antibody isolation strategy

- Figure shows the methods for isolating highly potent Neutralizing antibodies (nAbs) to SARS-CoV-2
- Article explains how their in vivo efficacy in a Syrian hamster model suggests their potential clinical use



(Rogers et al, 2020)



(Rogers et al, 2020)

Cohort study using CC6, CC12, and CC25 showed varied responses to SARS-CoV RBD and S

A

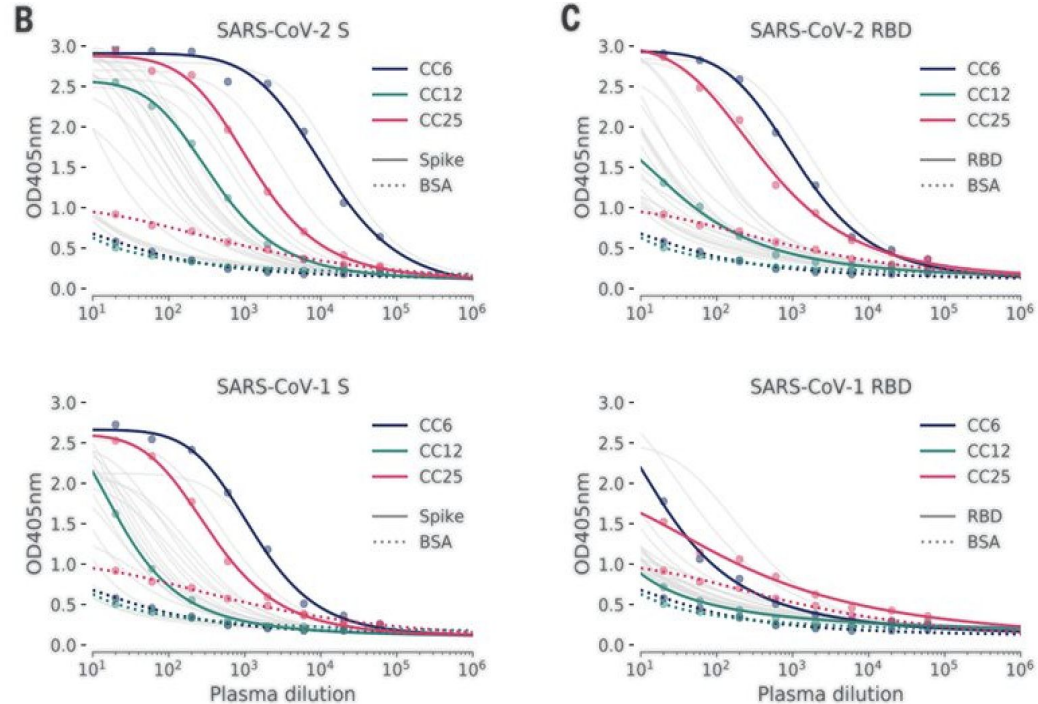
Patient Demographics	
Participants	17
Female	8 (47%)
Male	9 (53%)
Average Age	50
Days post-symptom onset	17
Disease severity	
Mild	10 (58%)
Moderate	3 (18%)
Severe	3 (18%)
Critical (intubated)	1 (6%)
Significant past medical history	
None	13 (76%)
Asthma	1
Hypertension	1
HIV	1
Chronic kidney disease	1
Coronary artery disease	1
Other	1

- Cohort of 17 individuals was established
- CC6, CC12, and CC25 were used for further study

(Rogers et al, 2020)

Cohort study using CC6, CC12, and CC25 showed varied responses to SARS-CoV RBD and S

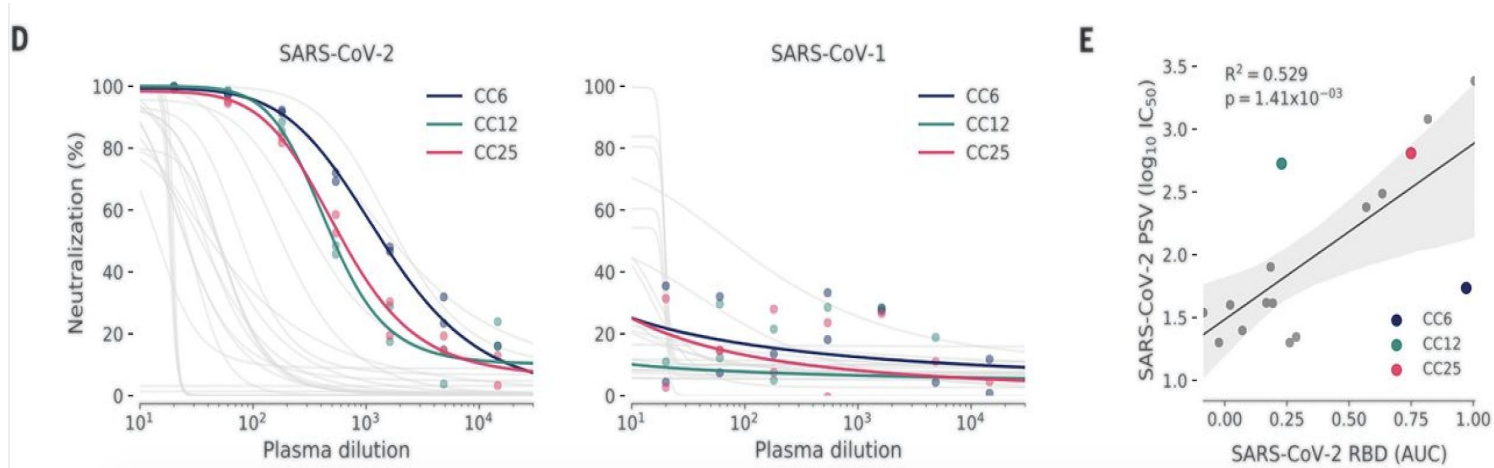
- Enzyme-Linked Immunosorbent Assay (ELISA) used
- SARS-CoV-2 showed higher OD405nm in both RBD and S
 - Three participants had different responses



(Rogers et al, 2020)

Cohort study using CC6, CC12, and CC25 showed varied responses to SARS-CoV RBD and S

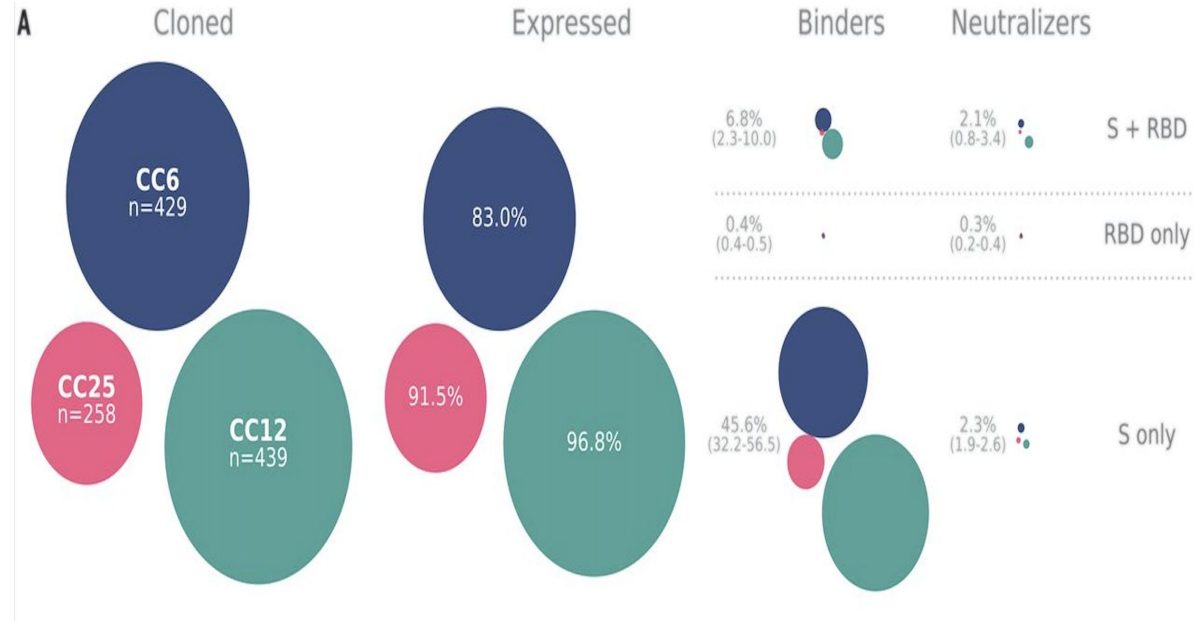
- Better percent neutralization of SARS-CoV-2 than SARS-CoV-1
- RBD binding and PSV neutralization were well correlated



(Rogers et al, 2020)

Binding antibodies show neutralization activity between RBD/S and S binders

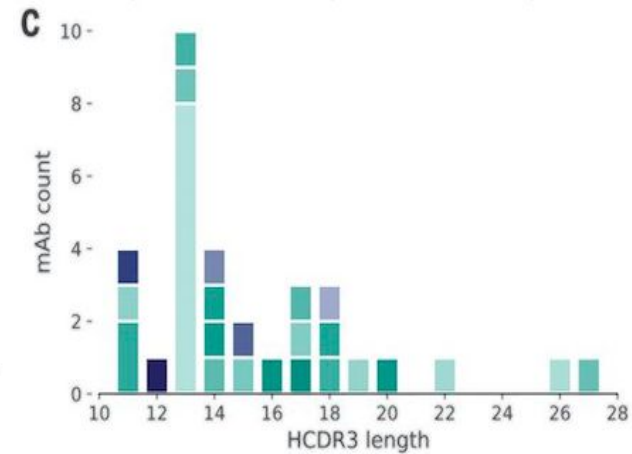
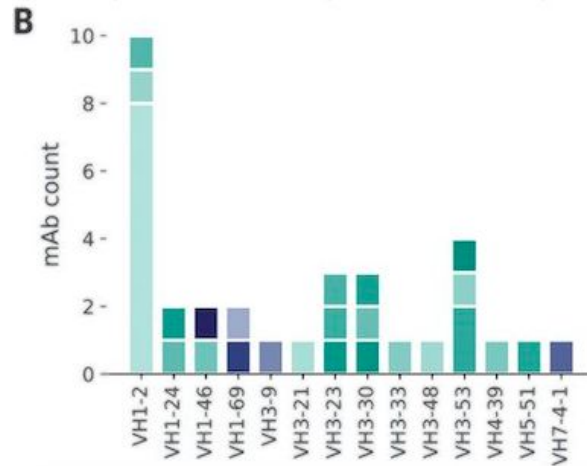
- RBD only shows little neutralization activity
- Strong response against S protein
 - Most are non-neutralizing



(Rogers et al, 2020)

Monoclonal antibody count by gene family and HCDR3 length

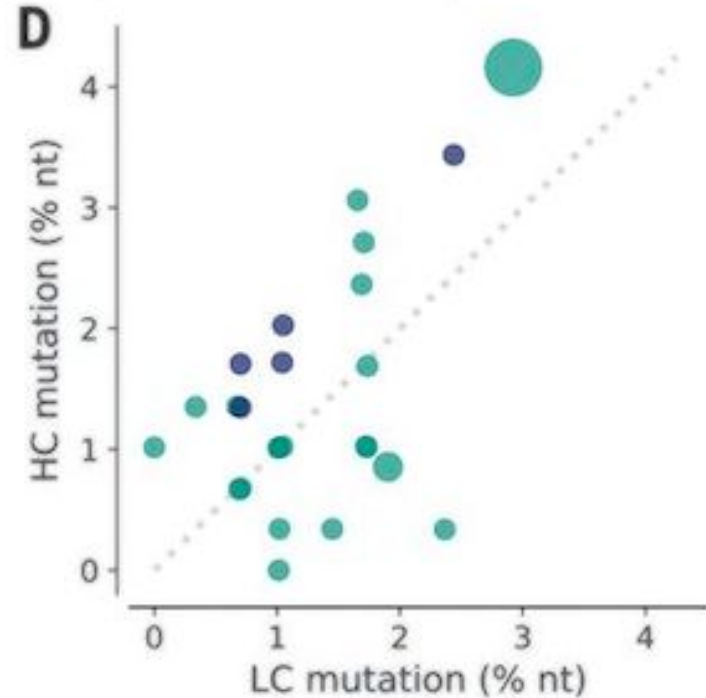
- VH1-2 gene family is most overrepresented in monoclonal antibodies
- Thirty-three antibodies were selected for in-depth characterization
 - There was diversity in CDR3 lengths



(Rogers et al, 2020)

Mutation Frequency of Down-Selected mAb Lineages

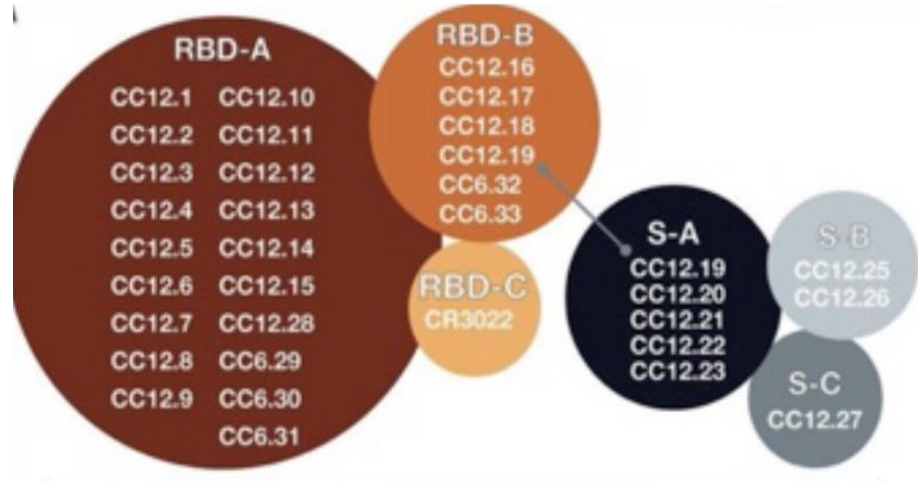
- Bubbles indicate mutation frequency for each lineage



(Rogers et al, 2020)

Identifying monoclonal antibody epitope bins

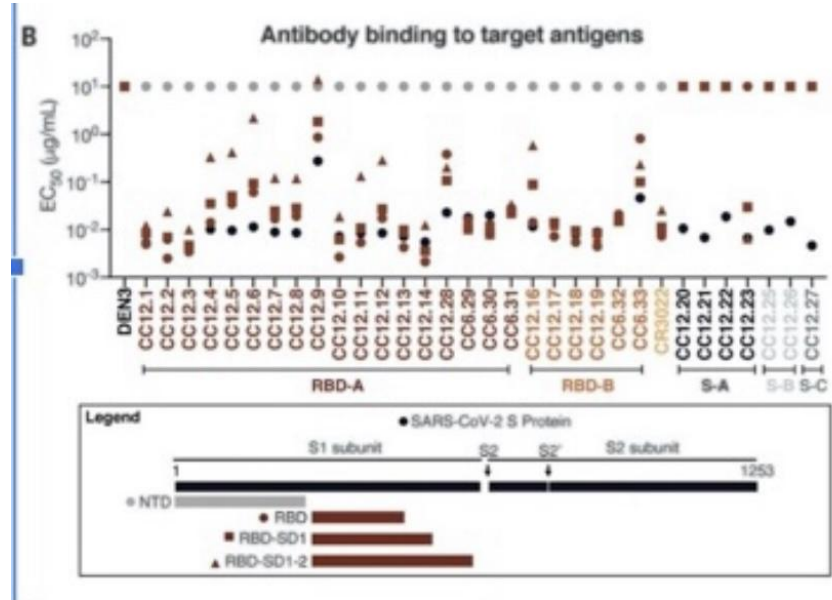
- 6 epitope bins
 - 3 S protein bins
 - 3 RBD bins
- CC12.19 might target epitopes RBD-B through S-A
 - Appears to compete with antibodies in these epitopes



(Rogers et al, 2020)

Evaluating epitope specificities

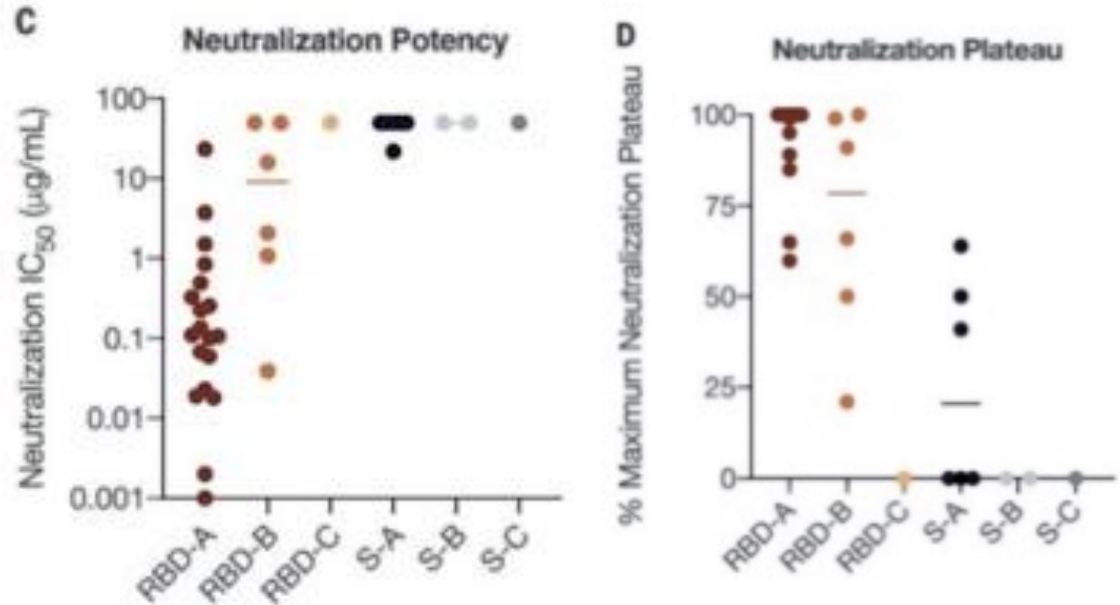
- No binding to NTD
- CC12.19 bound to all other constructs
 - All S-A bin antibodies did not bind in RBD or its subdomains



(Rogers et al, 2020)

Neutralization potency against SARS-CoV-2 and SARS-CoV-1 pseudoviruses

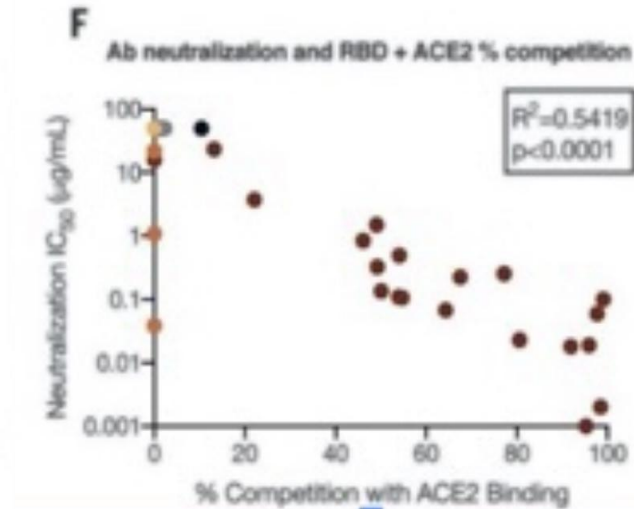
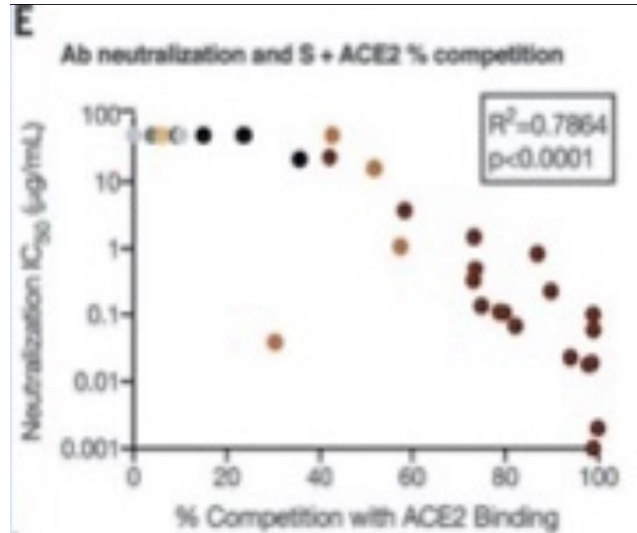
- Most potent neutralizing are in RBD-A
- CC6.29 and CC6.30 had high neutralization potency



(Rogers et al, 2020)

Evaluating RBD-A epitope on ACE2 binding site

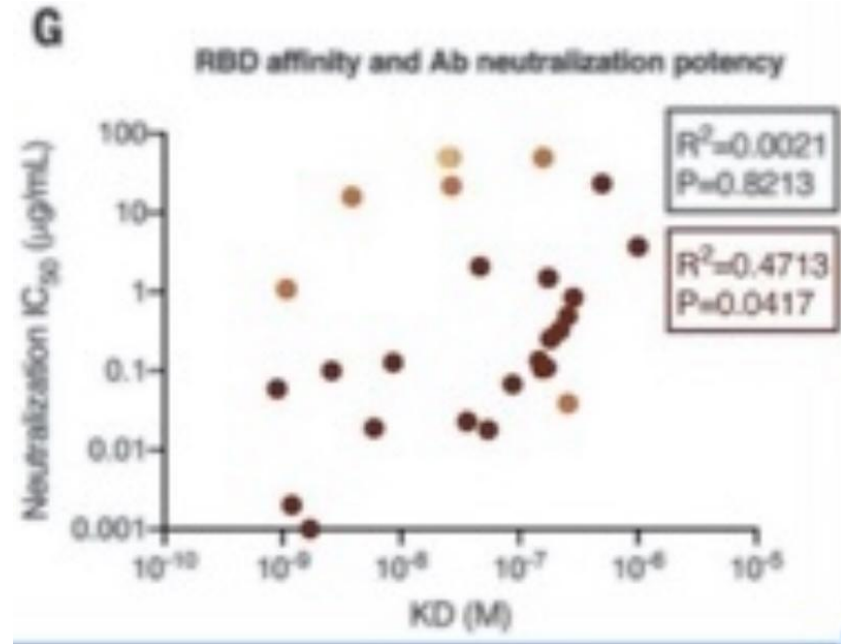
- Antibodies targeting RBD-A epitope competed ACE2 receptor



(Rogers et al, 2020)

Affinity of RBD-specific antibodies to soluble RBD

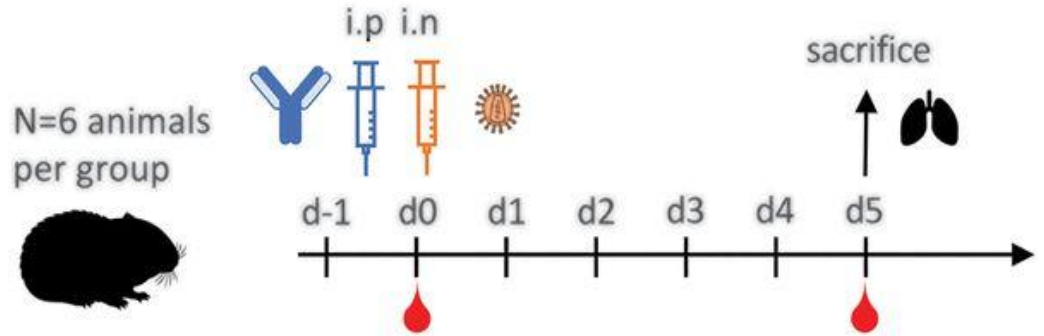
- Found low correlation between neutralization and affinity



(Rogers et al, 2020)

In vivo testing using Syrian hamsters

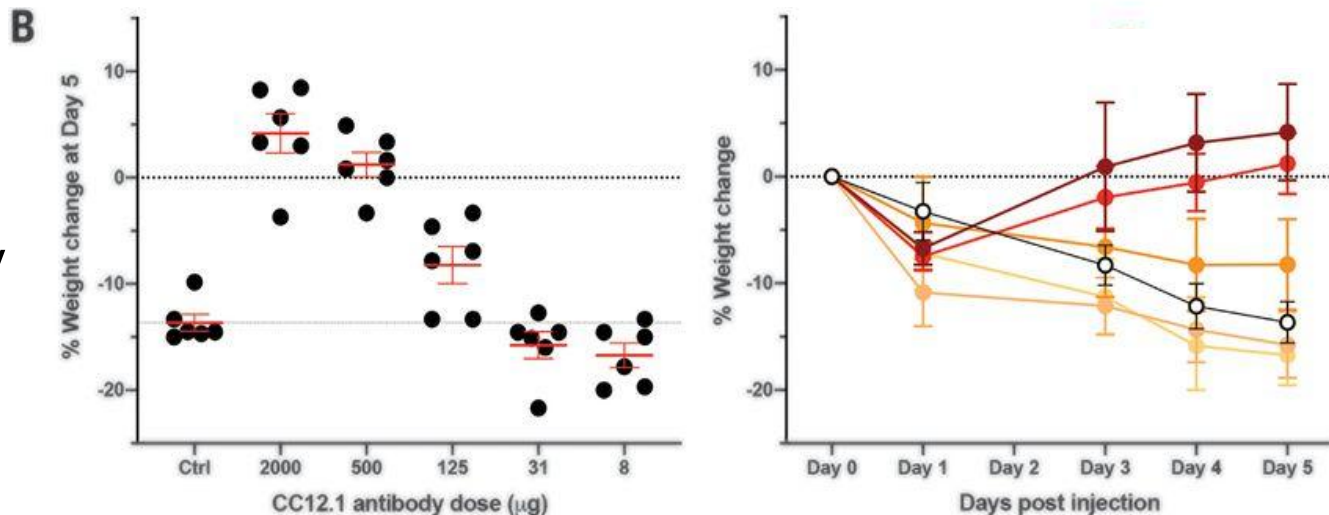
- Treated with an anti-SARS-CoV-2 antibody
- Anti-Dengue virus antibodies used as control
- Weight monitored every 24 hours after exposure to SARS-CoV-2



(Rogers et al, 2020)

Visualization of % weight change by antibody dose & days post injection

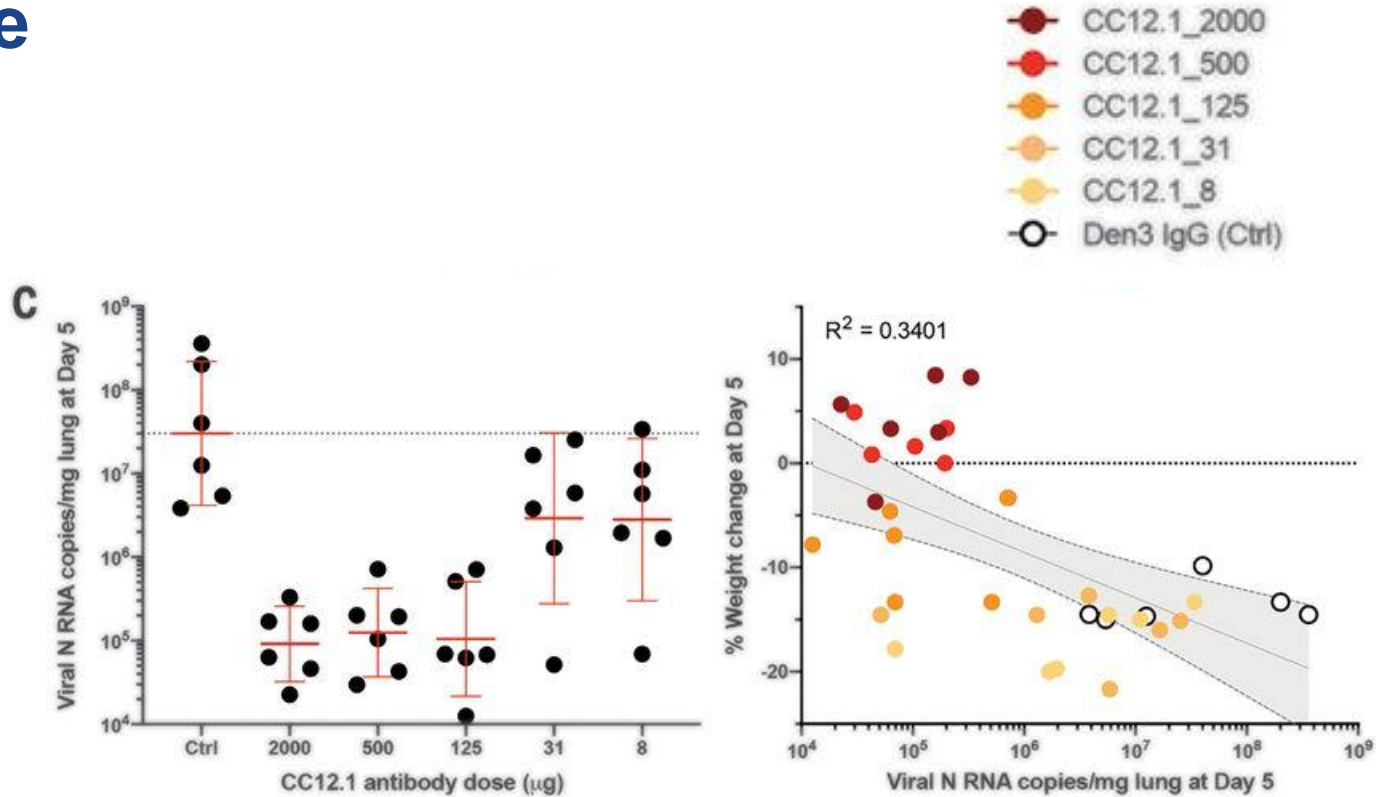
- 2 highest antibody doses prevented weight loss
- Difference between lowest dosage & control is statistically insignificant



(Rogers et al, 2020)

Visualization of viral load by antibody dose and % weight change

- 2 highest antibody doses resulted in reduced viral load
- Weight loss corresponded with higher viral load

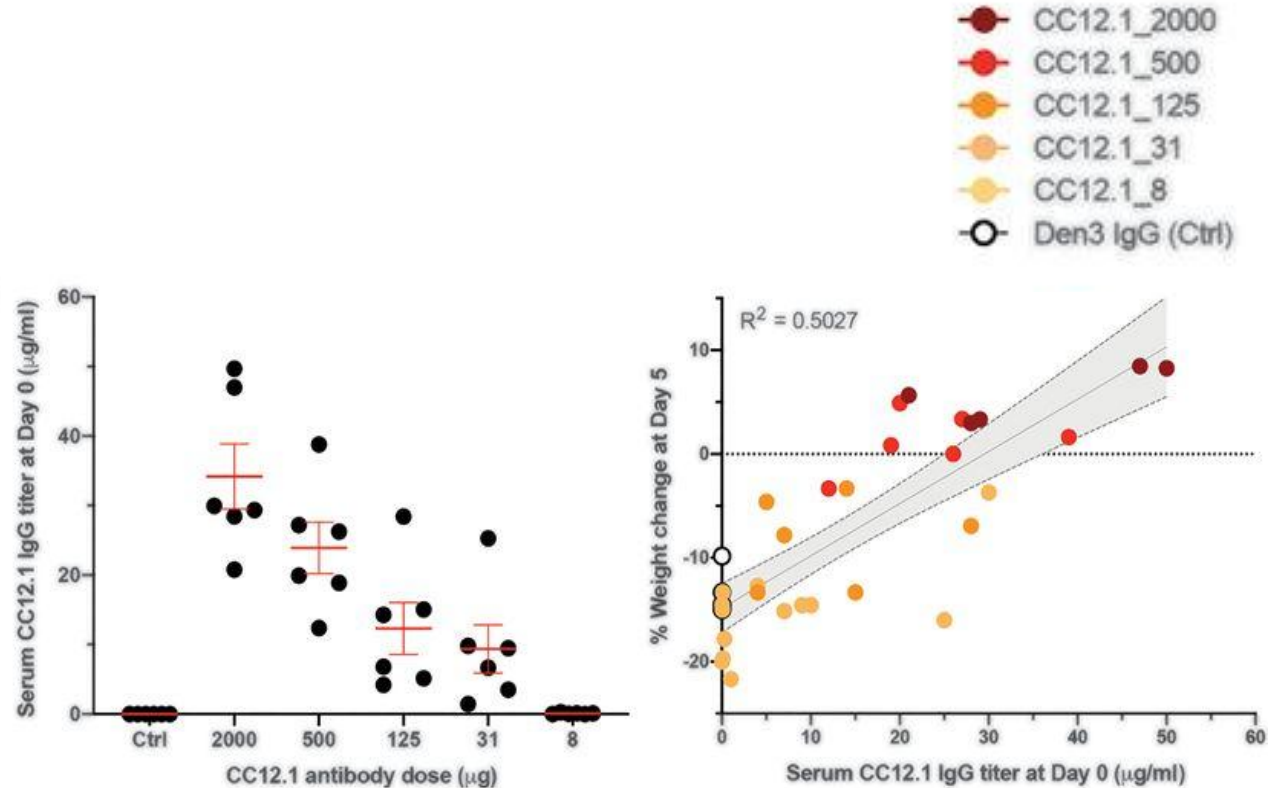


(Rogers et al, 2020)

Visualization of % weight change by antibody dose and serum titer

- Dose concentrations are reflected in serum titer
- Weight loss & active antibody concentration
 - Have a positive relationship

D



(Rogers et al, 2020)

Efficacy of RBD nAb in vivo in Syrian hamsters shows promise for human studies

- The most potent Abs target the RBD-A epitope, which overlaps with the ACE2 binding site
- Low potency and neutralization in Abs to the S protein
- RBD-A nAbs were able to compete with ACE2
 - Most preferred for prophylactic treatments
 - Could be a form of therapy for SARS-CoV-2 or lead to development of a vaccine
- Vaccine development should focus on the RBD, as there are strong nAb responses visible
- Future research should investigate possibility of antibody-mediated enhanced disease

Summary

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Acknowledgements

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LMU Biology Department

BIOL 368 Class
