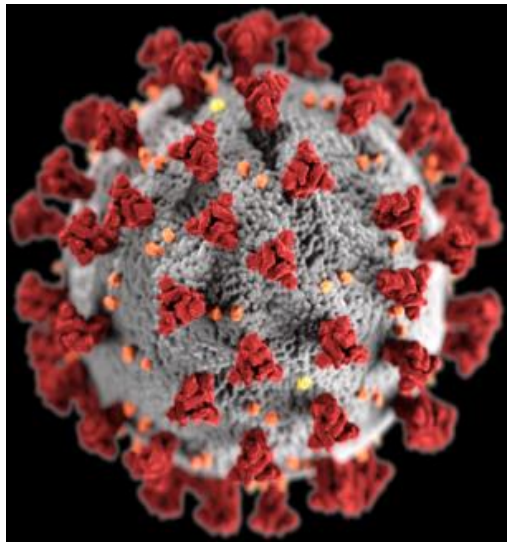
HKU Med LKS Faculty of Medicine The University of Hong Kong 香港大學李嘉誠醫學院

isirv: COVID-19
6th October 2020

Viral replication kinetics and antibody responses

Malik Peiris
The School of Public Health
The University of Hong Kong
malik@hku.hk

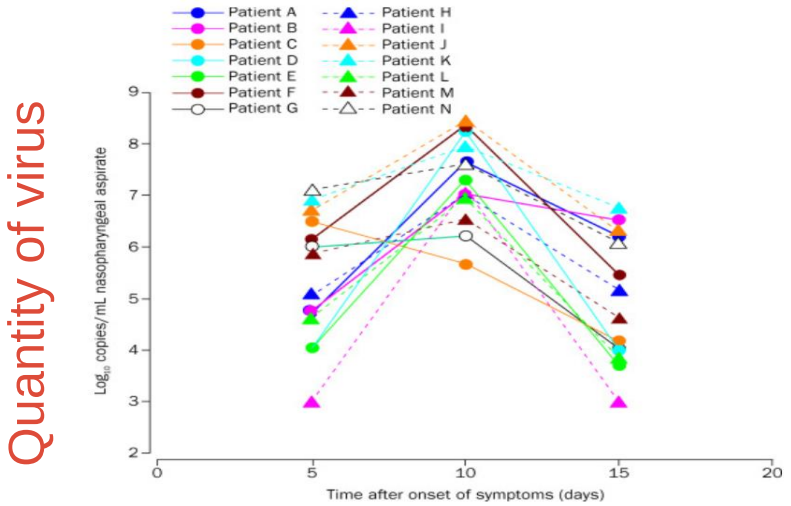


Viral dynamics and infectiousness of SARS-CoV-2

Lancet. 2003 May 24;361(9371):1767-72.

Clinical progression and viral load in a community outbreak of coronavirus-associated SARS pneumonia: a prospective study.

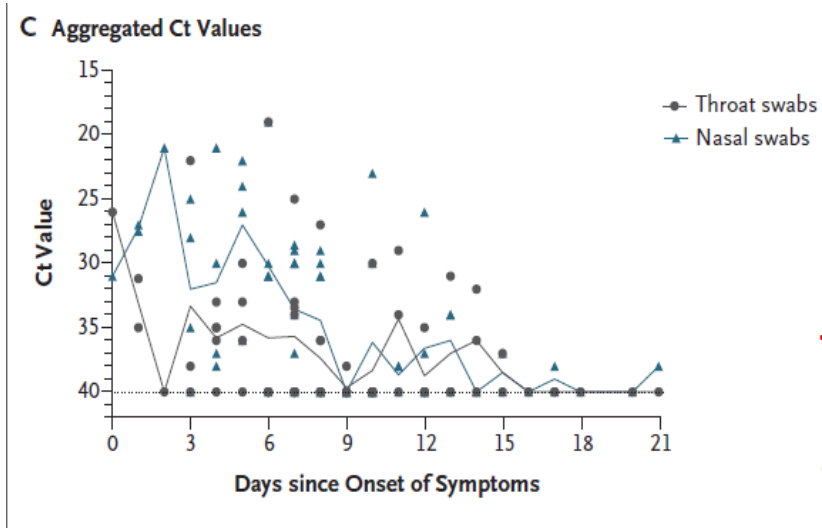
Peiris JS[1], Chu CM, Cheng VC, Chan KS, Hung IF, Poon LL, Law KI, Tang BS, Hon TY, Chan CS, Chan KH, Ng JS, Zheng BJ, Ng WL, Lai RW, Guan Y, Yuen KY; HKU/UCH SARS Study Group.



Quantity of virus

SARS

Peiris et al., Lancet 2003



Quantity of virus

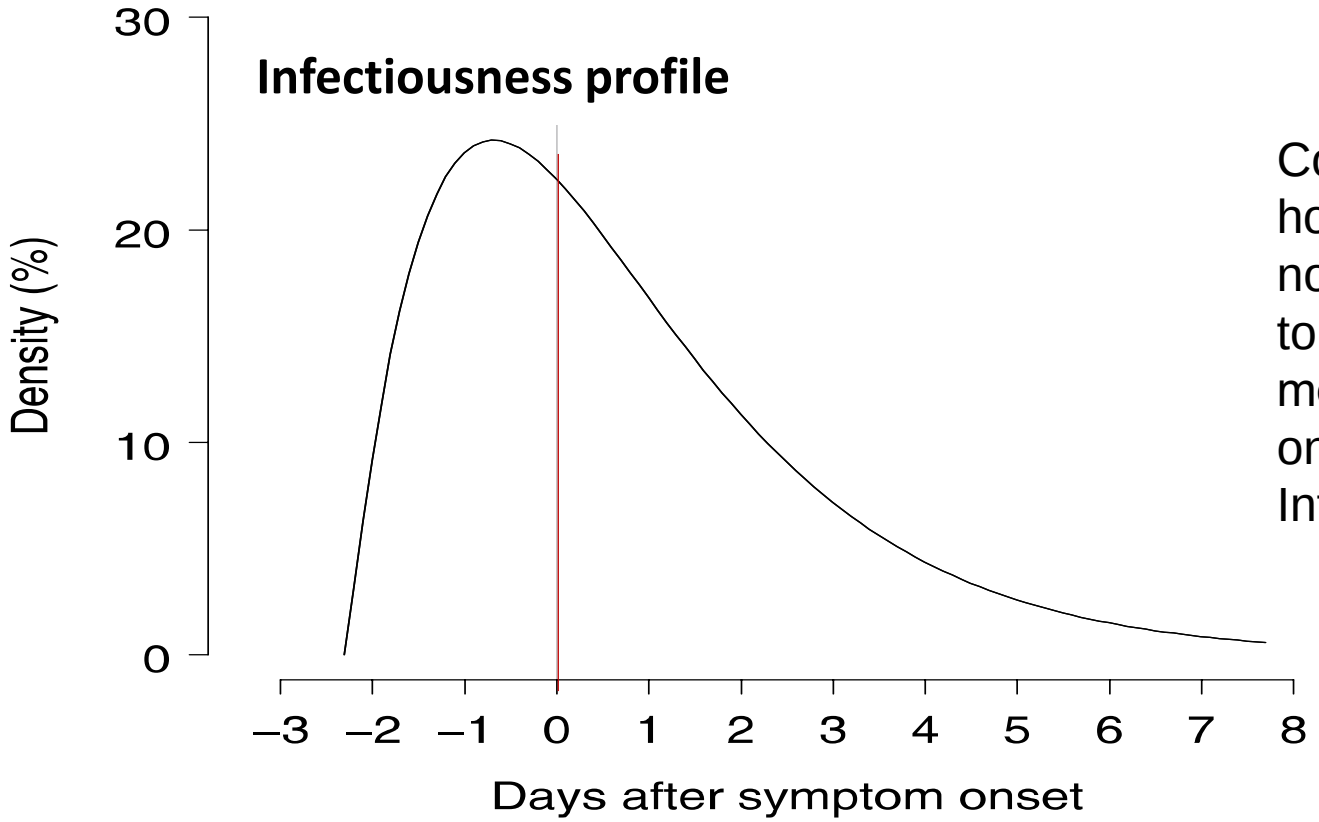
COVID -19; Flu

Zou et al NEJM Feb 18 2020

"- - pattern of patients infected with SARS-CoV-2 **resembles that of patients with influenza**"

"-- **require strategies different** from those required for the control of SARS-CoV. "

Infectiousness profile of SARS-CoV-2



Contact tracing → high-risk household and hospital contacts did not develop infection if their exposure to a case patient started 6 days or more after the case patient's illness onset (Cheng et al., 2020 JAMA Intern Med)

Estimated that about 44% of infectiousness occurred in pre-symptomatic stage

He et al Nature Medicine 2020

Duration of SARS-CoV-2 RNA detection

Omar S et al Eurosurveillance 2020; 25(30):2001292

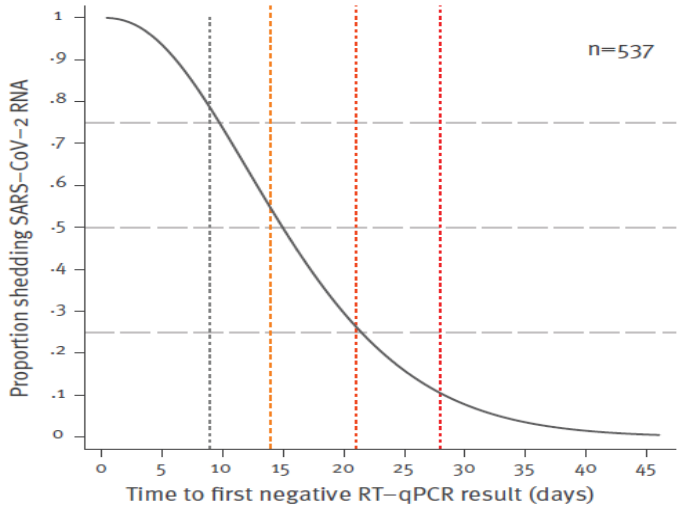
TABLE 1

Reports providing descriptive statistics on duration of SARS-CoV-2 RNA positivity by RT-qPCR with at least 30 subjects and entry in PubMed by 8 April 2020 (n = 9)

Ref	Country	Setting	Severity	Population size	Detection period	Sampling interval[a]	Distribution of outcome in population (days)			Time to event analysis	Antivirals, comments
							Median or mean	IQR or SD	Maximum		
[5]	China	Hospital	53% moderate, 39% severe, 8% critical	137	Symptom onset to two negative results	Regular; every other day	Median: 20.0	IQR: 17.0–24.0	37	None	n=41 lopinavir/ritonavir; analysis restricted to 137/191 survivors opens potential for selection bias
[7]	China	Hospital	Mild to moderate	56	Symptom onset to first negative result	Irregular	Median: 24.0	IQR: 18.0–31.0	42	None	None
[6]	China	Hospital	94% moderate, 6% severe	70	Symptom onset to first negative result	Irregular	Median: 22.0	IQR: 19.0–32.0	No data	None	Focus of study is on re-occurring positives
[8]	China	Hospital	74% moderate, 25% severe, 1% critical	120	Symptom onset to two negative results	Regular; every other day	$Median_{overall}$: 23.0	IQR: 18.0–32.0	No data	Kaplan–Meier, Cox-regression	n=78 lopinavir/ ritonavir and n=42 no treatment
							$Median_{antiviral}$: 22.0	IQR: 18.0–29.0			
							$Median_{non\text{-}antiviral}$: 28.5	IQR: 19.5–38.0			
[9]	China	Hospital	75% non-ICU 25% ICU	32	Symptom onset to first negative result	Irregular	$Mean_{non\text{-}ICU}$: 15.67	SD: 6.68	No data	None	None
							$Mean_{ICU}$: 22.25	SD: 3.62			
[10]	China	Hospital	28% severe	113	Symptom onset to first negative result	Regular, daily	Median: 17.0	IQR: 13.0–22.0	No data	Kaplan–Meier	n=55 umifenovir, n=19 ribavirin
[11]	China	Hospital	83% non-severe; 17% severe	59	From first positive to first negative result	Regular, daily	Median: 14.0	IQR: 10.0–18.0	25	Kaplan–Meier, Cox-regression	All patients ribavirin; variation in definition of detection period may explain lower estimates
[12]	China	Hospital	86% moderate; 14% severe	147	Symptom onset to first negative result	Regular, every other day	Median: 17.0	IQR: 12.0–21.0	47	Kaplan–Meier	All patients received antiviral treatment, 'most commonly ribavirin and interferon'
[13]	Korea	Outpatient	100% mild	199	Diagnosis to two negative results	Irregular; every other day to once a week	$Mean_{overall}$: 24.5	SD: 4.8	No data	none	53 asymptomatic and 146 symptomatic subjects

Median ranges 14 – 24.5 days
Maximum ranges from 25 to 47 days

Mild symptomatic cases managed at home, Germany



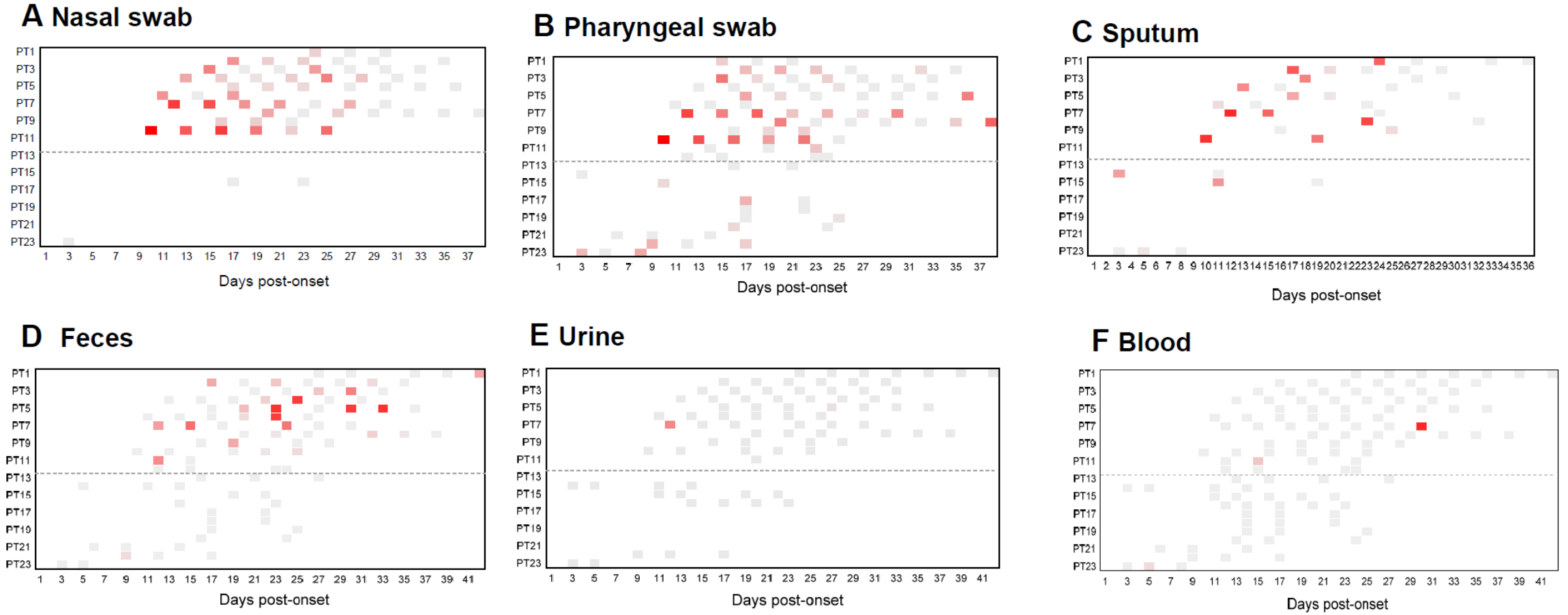
Relevant to differentiating persistence from "re-infection" - CDC 3 months
https://www.cdc.gov/coronavirus/2019-ncov/hcp/duration-isolation.html

Factors affecting duration of RNA detection:

- Type of specimen: sputum > NPS; Peak Temperature; hydrocortisone Rx
  Wang et al Chest 2020 June; Li TZ et al J Med Virol 2020 Jul
- Asymptomatic vs. matched mild cases: 19 (IQR 15-26) days vs. 14 (IQR 15-26) days (p=0.03)
  Long et al Nature Med 2020.

SARS-CoV-2 viral load in different clinical specimens of severe (n=12) and mild (n=11) cases over time

Wang Y et al J Clin Invest 2020

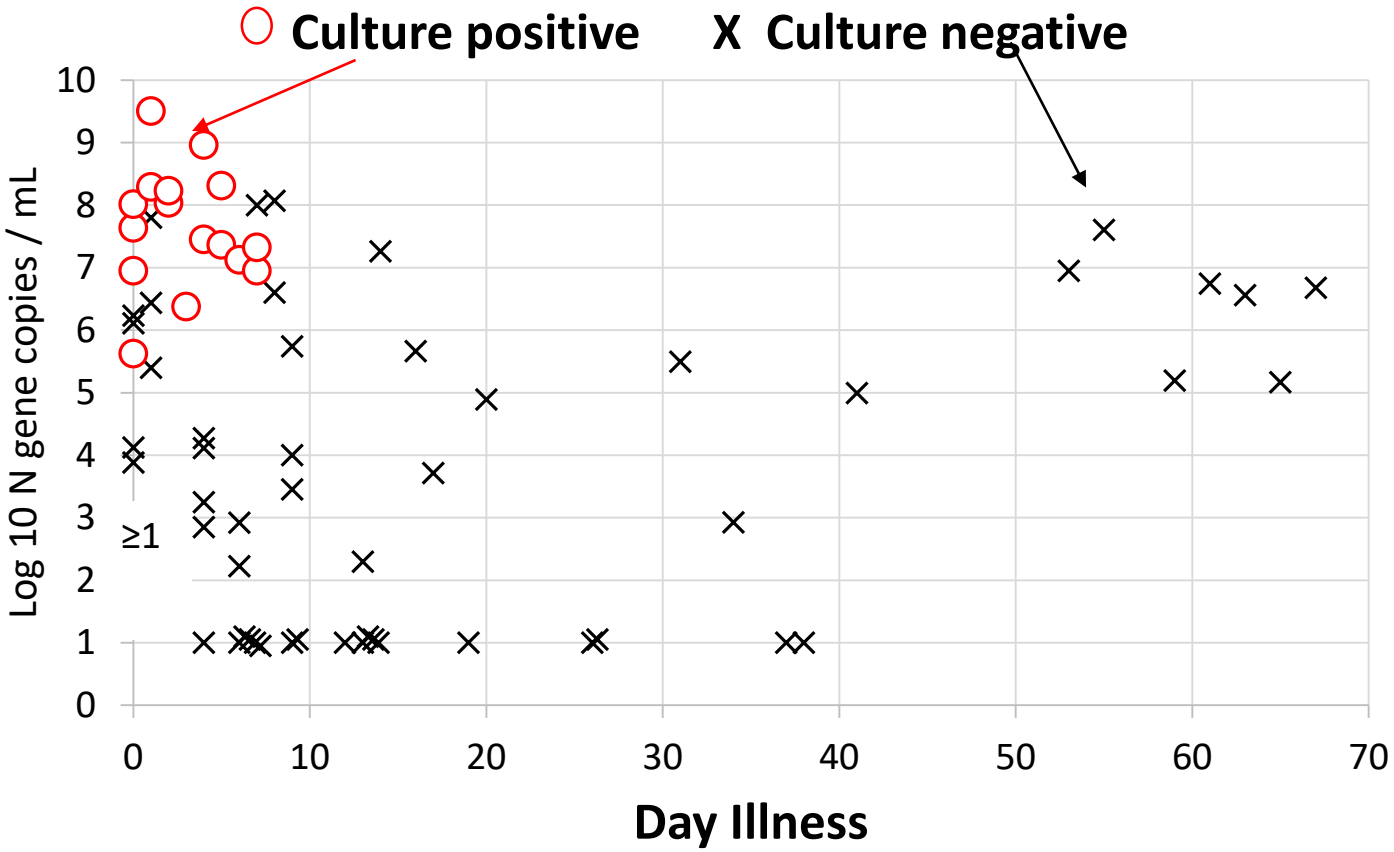


Median age 56 yrs (range 24-82 yrs)

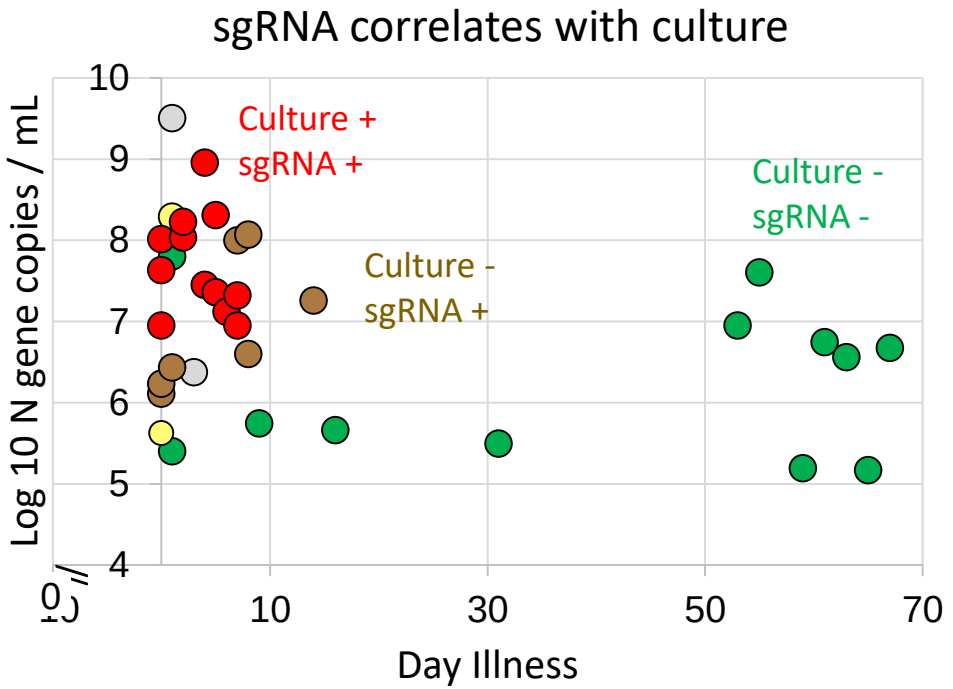
Severe cases: viral RNA up to 30-40 days; Mild cases: viral RNA ≤15 days

Duration of infectiousness? (Mild COVID-19)

Viral RNA load, virus culture vs. days illness.



Virus culture vs. subgenomic RNA vs. viral RNA load

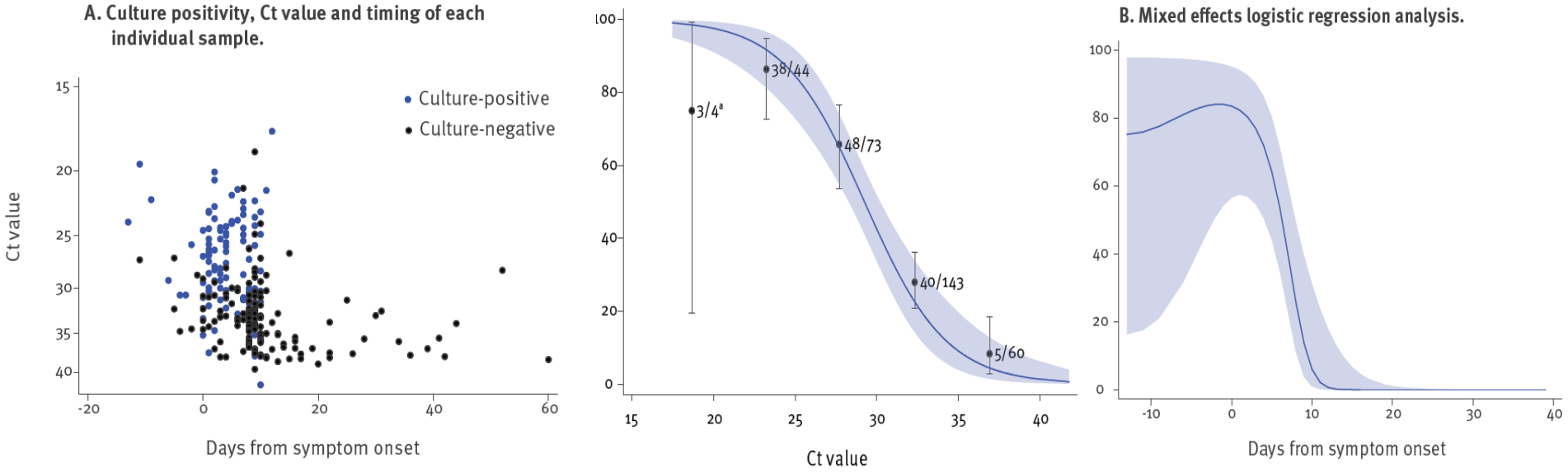


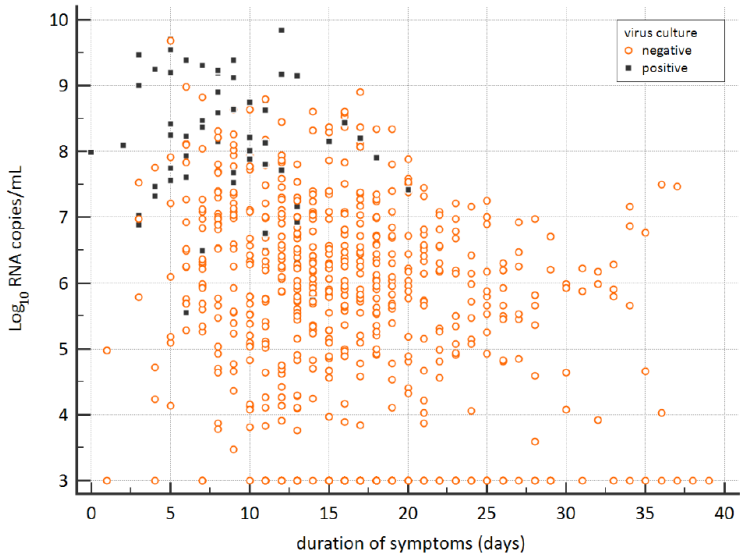
Perera et al Emerging Infectious Diseases 2020

Duration of infectiousness, England

Mild/moderate disease, N=324

A Singanayagam et al Eurosurveillance 2020; 25 (32): pii=2001483



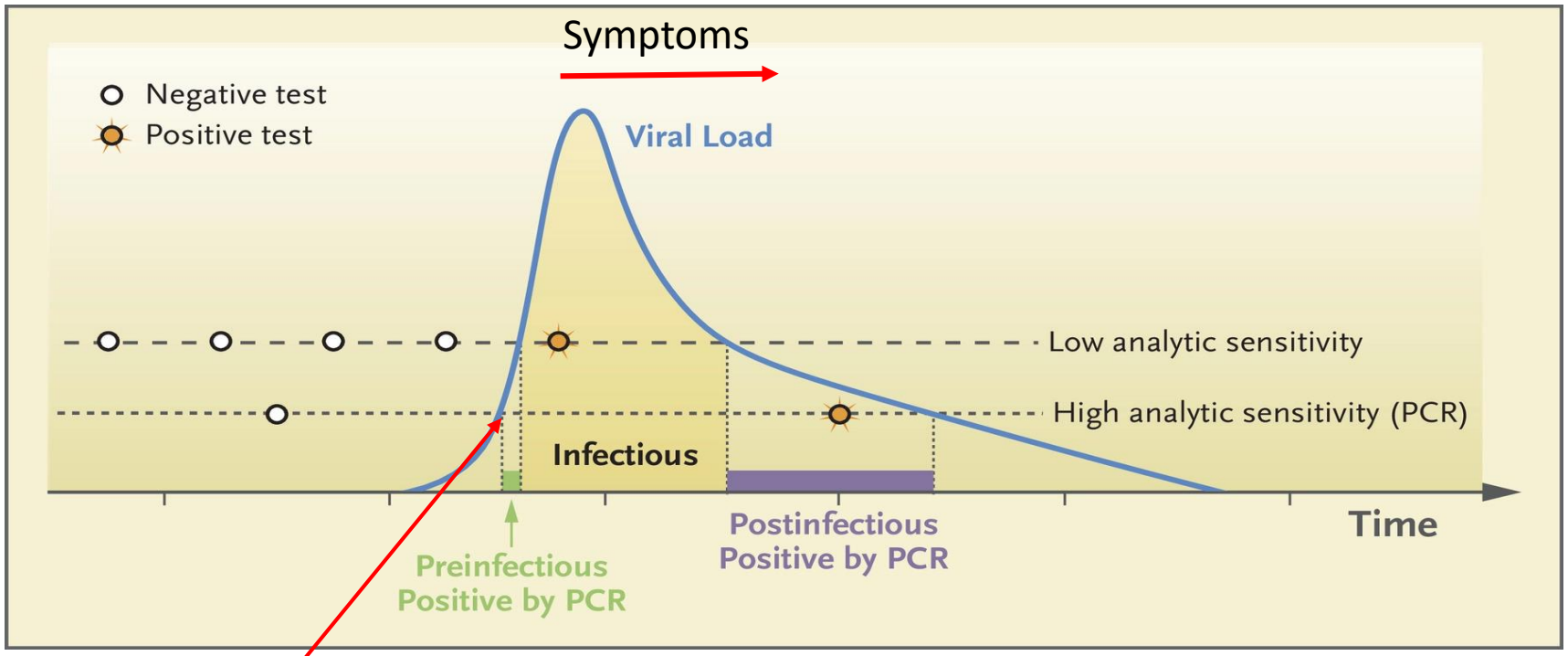


Severe cases / immunocompromised shed infectious virus for longer.

Even in severe cases, 88% culture negative by day 10, 95% negative by day 15

Van Kampen et al medRxiv

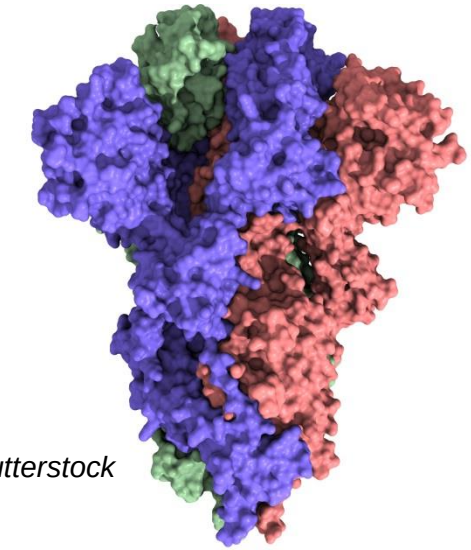
Infectiousness, test “sensitivity” and testing strategy



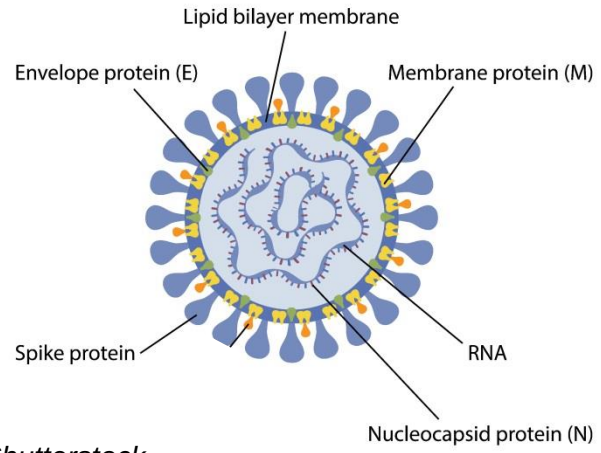
Modified from Mina et al NEJM Oct 2020

Viral load
Antibody

Serology



Shutterstock



Shutterstock

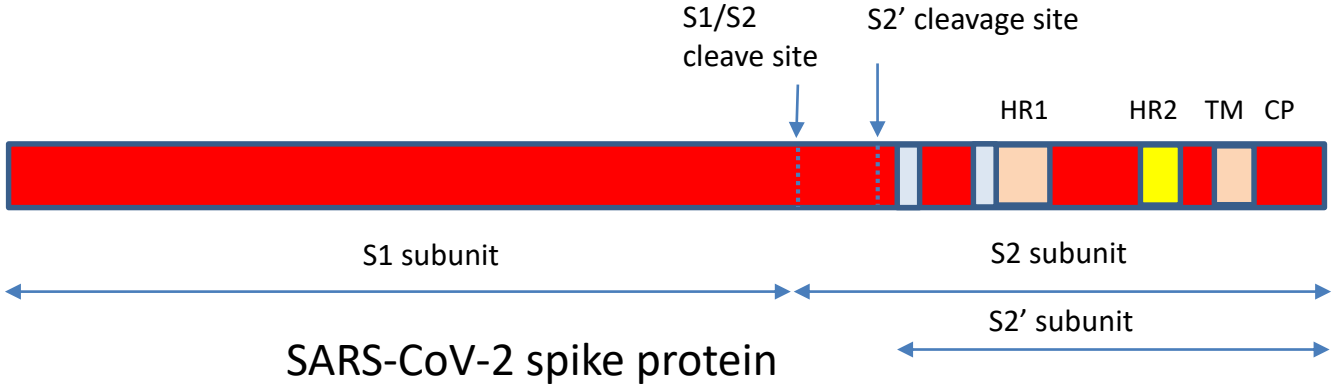


Figure independently created by GSK based on concepts from the references. Data first published in Amanat et al. Nat Med 2020. DOI: 10.1038/s41591-020-0913-5

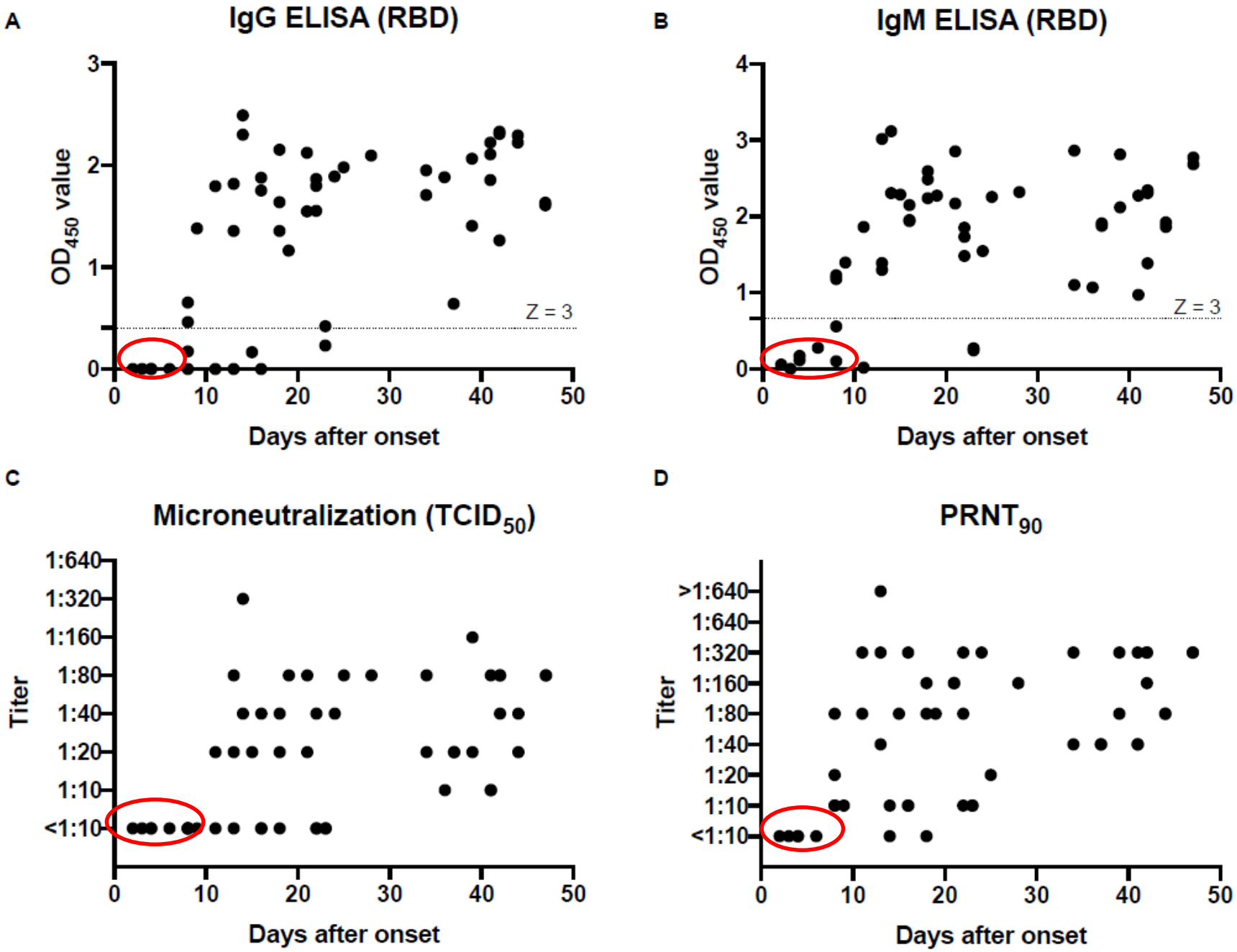
Genus	Virus	Spike protein				N
		RBD	NTD	S2	Whole S	
Beta CoV	SARS-CoV	73%	53%	90%	77%	90%
	BatCoV RaTG13	90%	99%	99%		
	Pangolin-CoV	97%	67%	98%		
	MERS-CoV	NS	NS	43%	33%	49%
	HCoV-HKU1	NS	NS	41%	32%	34%
	HCoV-OC43	NS	NS	42%	33%	34%
Alpha CoV	HCoV-NL63	NS	NS	34%	28%	29%
	HCoV-229E	NS	NS	35%	30%	28%
*NS: No statistically significant sequence alignment can be observed						

Adapted from Okba NMA et al. Emerg Infect Dis. 2020;26(7):1478-1488. doi:10.3201/eid2607.200841

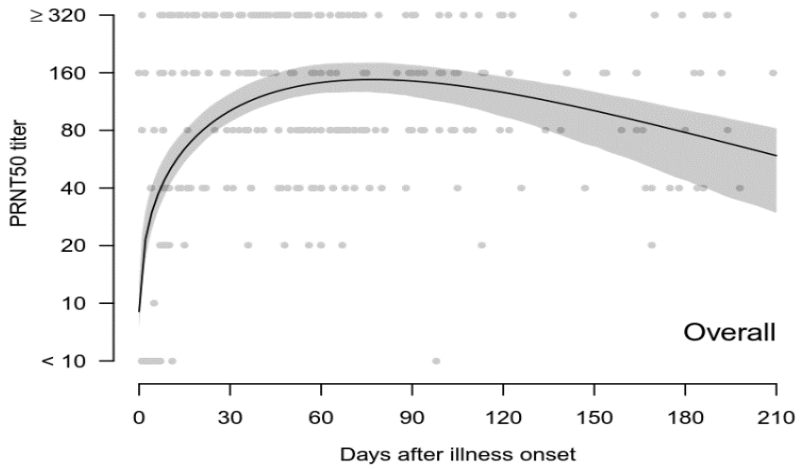
Amanat F et al. Nat Med 2020. DOI: 10.1038/s41591-020-0913-5; Okba NMA et al. Emerg Infect Dis. 2020;26(7):1478-1488. doi:10.3201/eid2607.200841

- Cohort of 24 patients followed up with multiple serum samples
- First 4 days → no detectable antibody
- >28 days 100% have antibody
- 200 blood donors: No cross reactions
- RBD IgG ELISA ideal screening assay for large scale population sero-epi

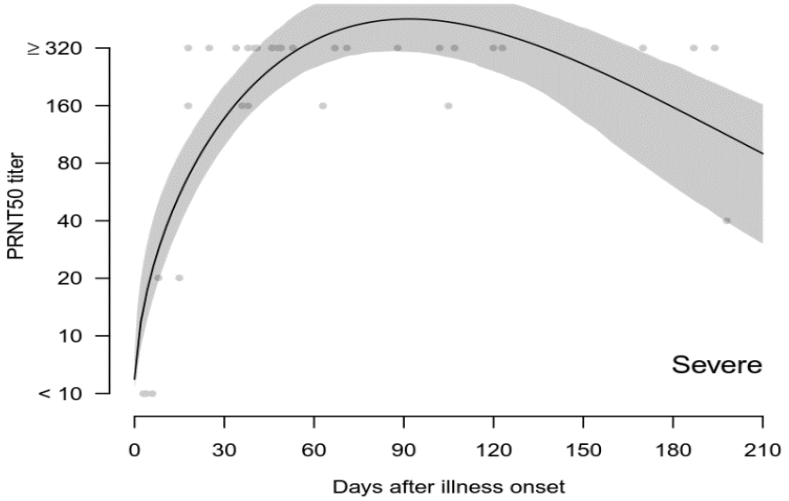
Perera et al Eurosurveillance 2020

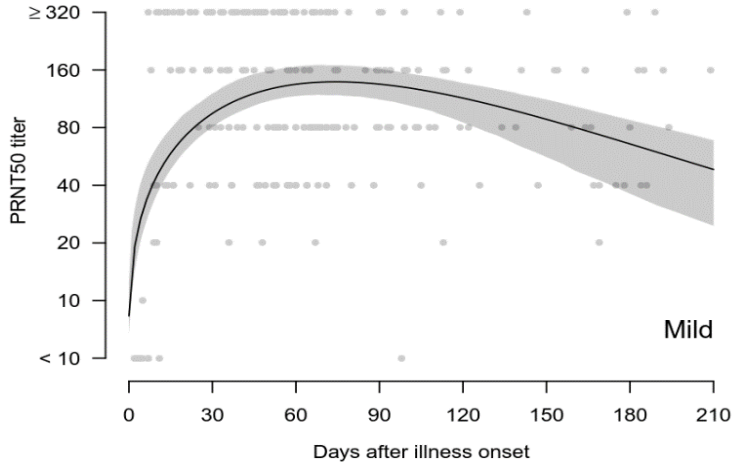


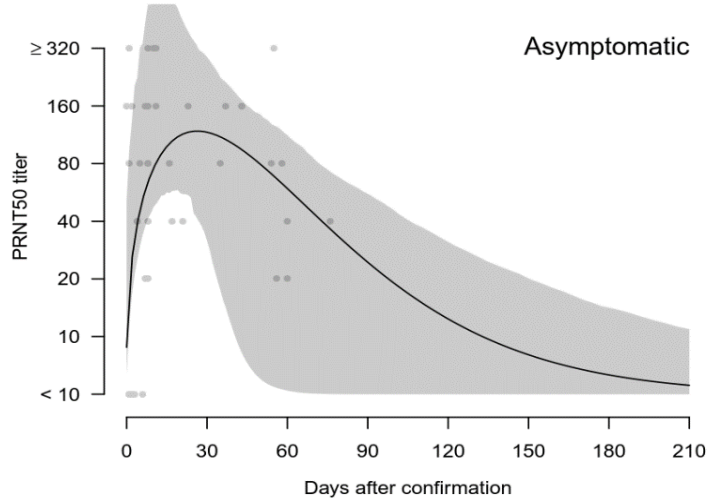
Duration of 50% plaque reduction neutralization test (PRNT) antibodies in COVID-19 convalescence (192 patients; 293 sera)



- Asymptomatic – 31; mild – 151; severe – 13
- Day >90 after onset (n=61); 98% remains PRNT50 pos
- Severe patients: Higher peak, peaks later
- Estimates 372, 416 and 133 days to fall to undetectable in severe, mild and asymptomatic infections
- Age >60 trend to higher antibody titres, adjusted for severity, corticosteroid use

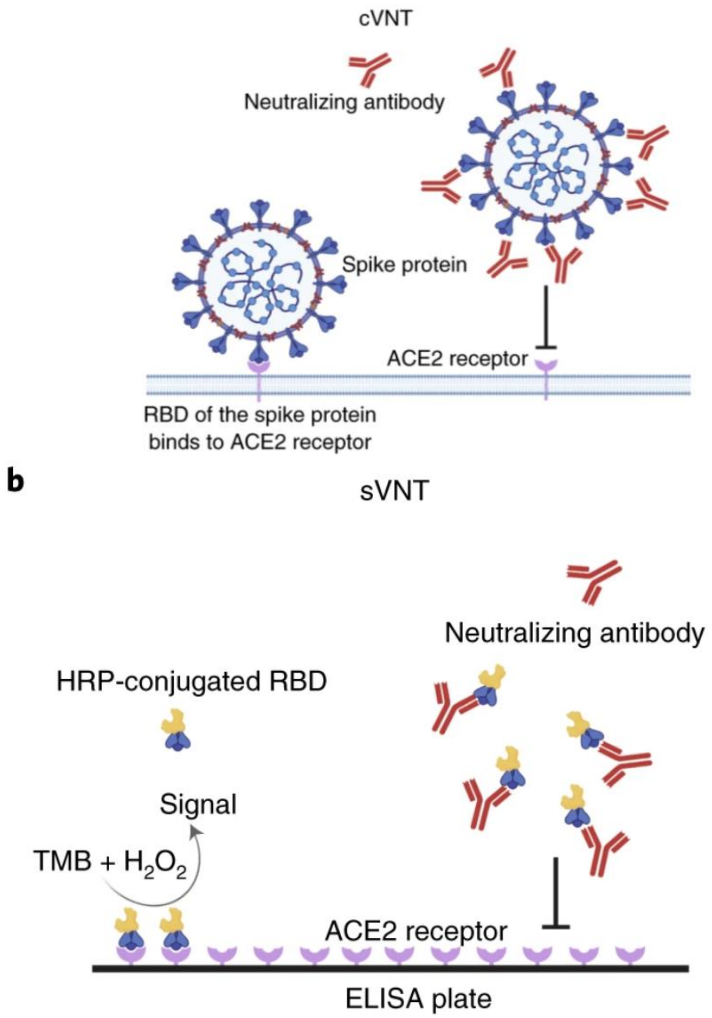




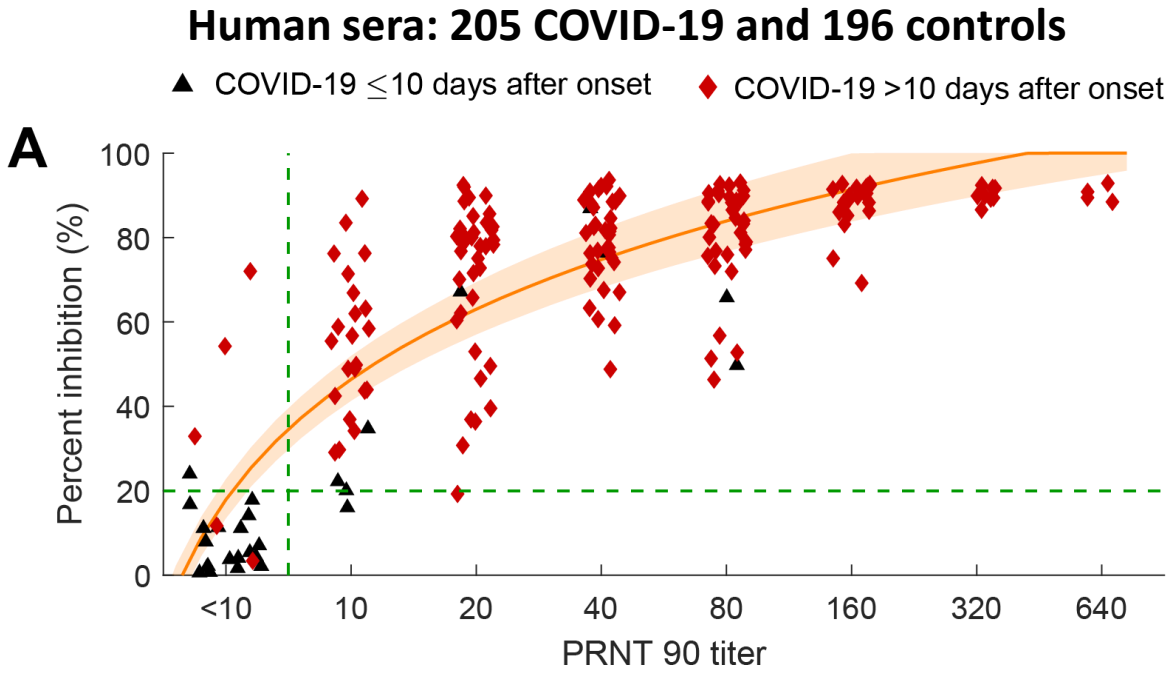


Peiris - Unpublished data

Surrogate virus neutralization test (sVNT)



Tan et al Nature Biotechnol 2020

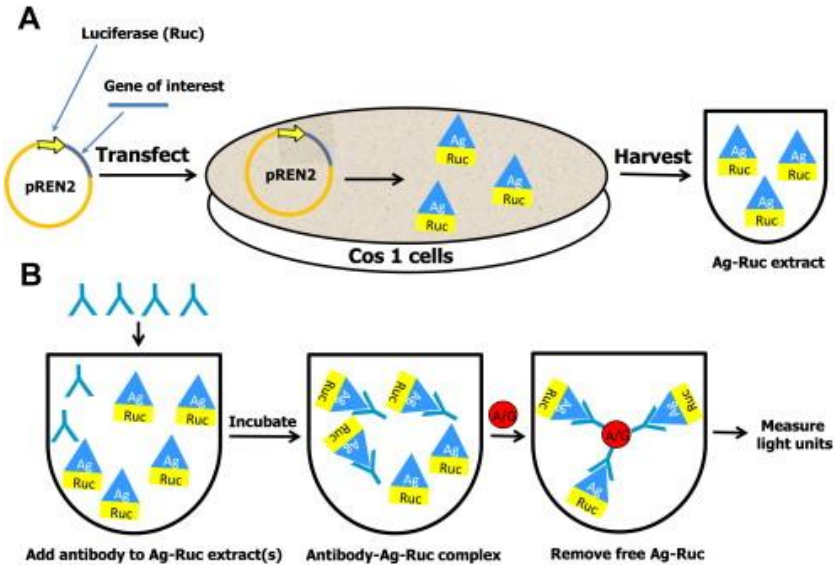


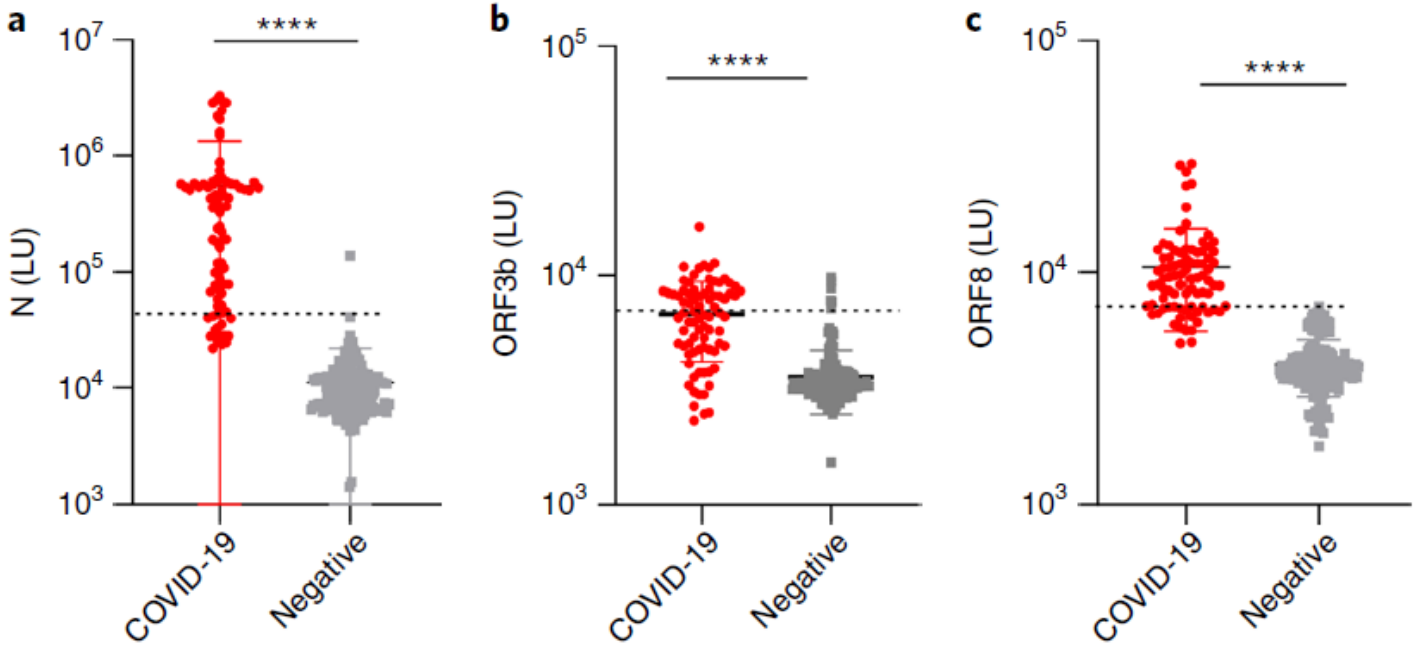
- With PRNT90 as gold standard, sVNT has sensitivity of 98.9%, specificity 98.7%, PPV 97.8%, NPV 99.4%
- Overall concordance 98.4%
- Species independent; validated on human, dog, cat, hamster sera.

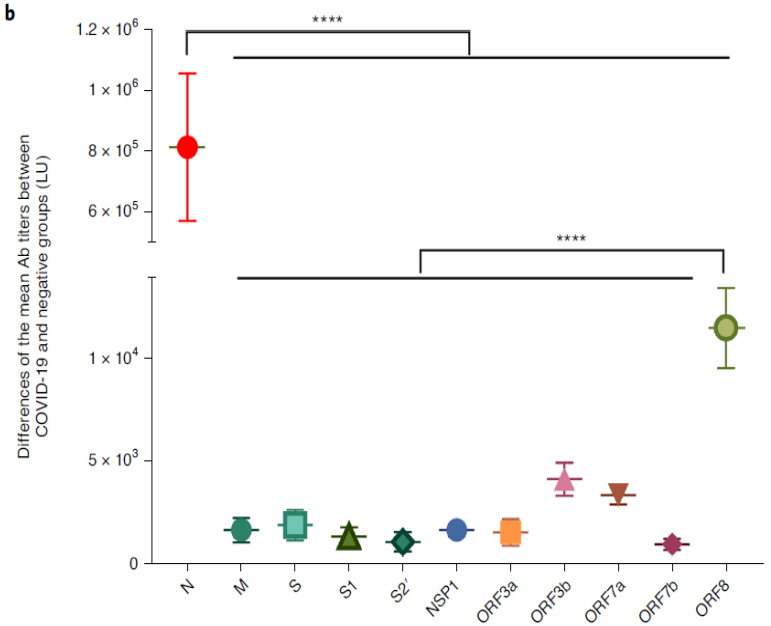
Peiris - unpublished

Landscape of antibody responses to 15 SARS-CoV-2 proteins using luciferase immuno-precipitation

- Immuno-dominance N>ORF8>ORF3b
- ORF8: Not found in seasonal HCoV

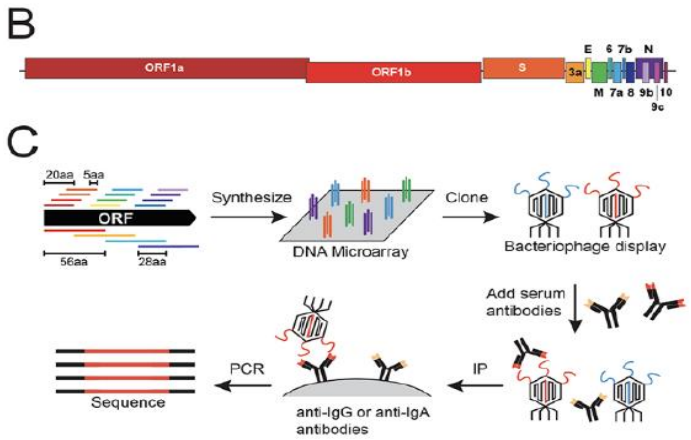




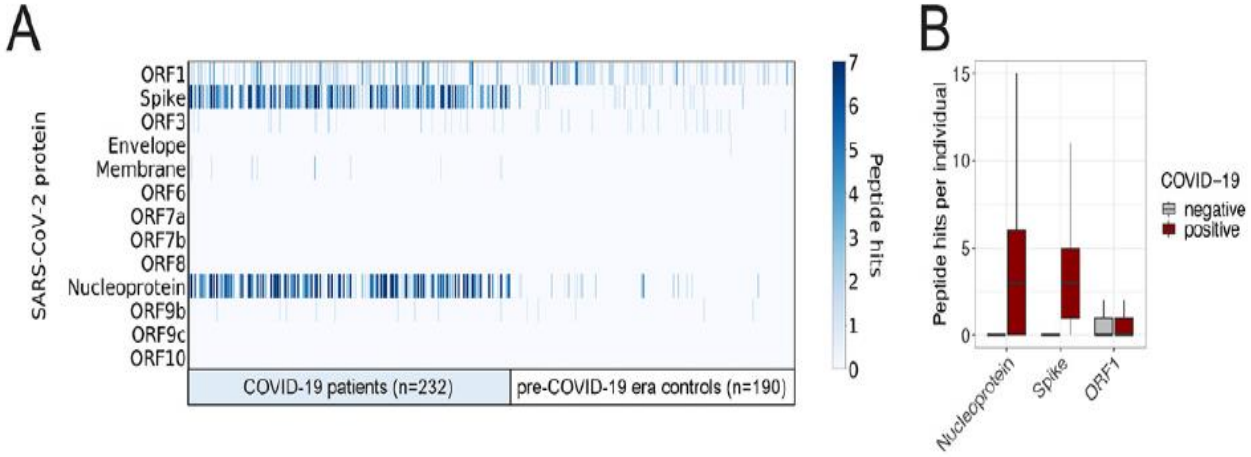


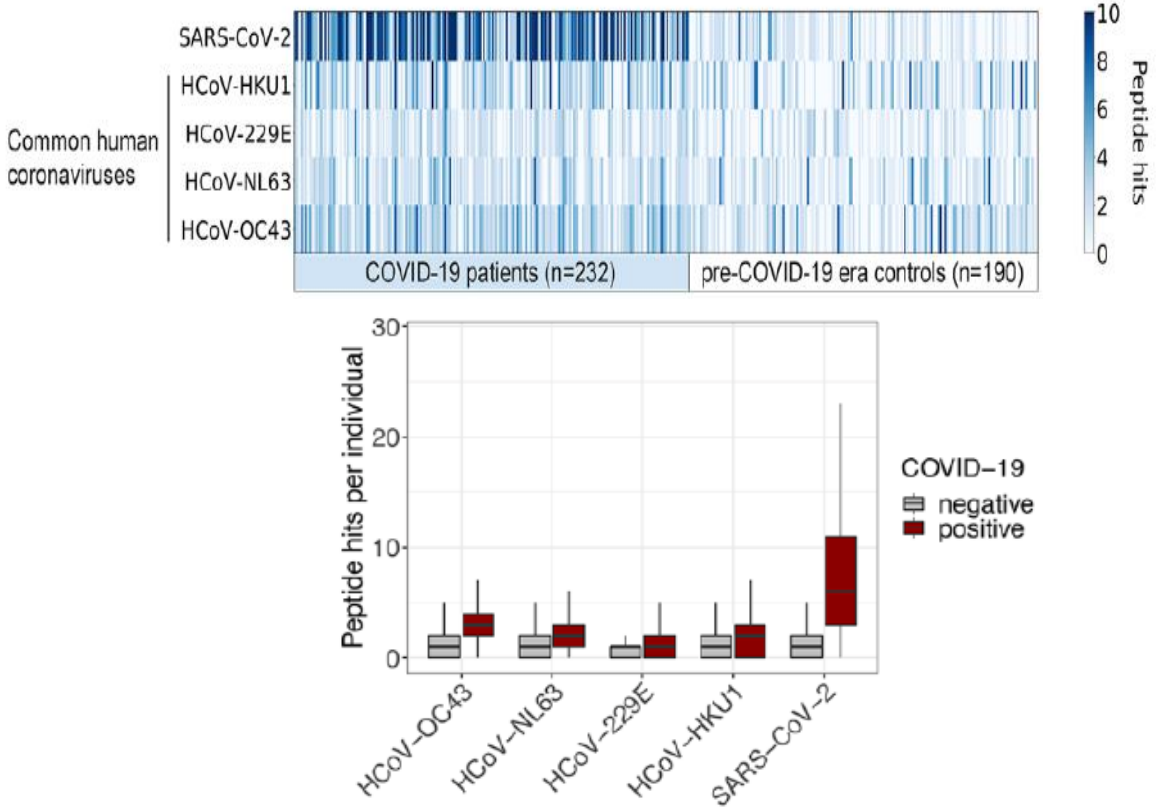
Hachem et al Nature Immunol 2020

Virus epitope profiling of COVID-19



Peptide scan of SARS-CoV-2 proteome
Immuno-dominance: N>S>>ORF1>>>>others
ORF1 is cross reactive with seasonal CoVs, non-discriminative





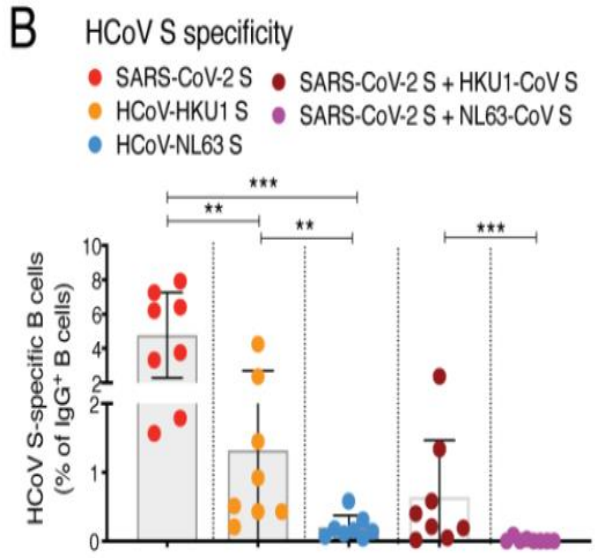
COVID-19 sera: Increased peptide hits to seasonal CoVs. Anamnestic booster responses to prior seasonal CoV memory

Shrock et al Science Sept 2020

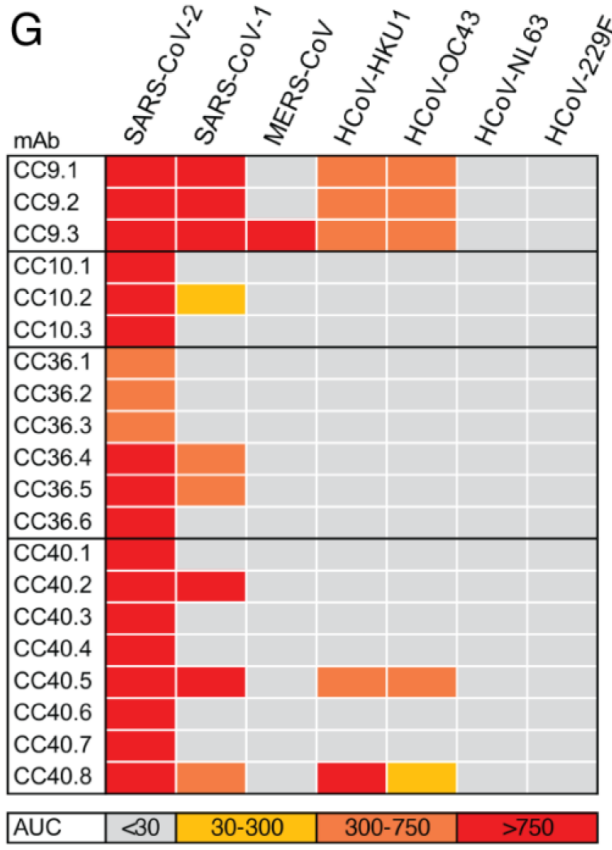
Cross-reactive antibody B cell responses to SARS-CoV-2 spike

- Weak evidence of pre-existing SARS-CoV-2 cross reactive antibody in pre-pandemic sera
- Stronger evidence of pre-existing cross-reactive memory B cells activated following SARS-CoV-2 infection
- Mabs with varying degree of cross reactivity with beta-CoV. These have higher binding constants (KDs) to HKU1 rather than SARS-CoV-2 → suggests initially elicited by HKU1

Memory B cells detected in COVID-19 donors



HCoV spike reactivity of MAbs from COVID-19 donors in cell-ELISA



Broad cross rn w betaCoV

Weak neut SARS-CoV-2

Song et al bioRExiv Sept 2020

Summary

- Viral load kinetics of SARS-CoV-2 more like flu than SARS-CoV-1 → explains early transmission from pre/asymptomatic infections → challenge for control
- Viral shedding may be prolonged but infectiousness does not correlate with positive RT-PCR. In mild infections, virus can be isolated in culture in first 8-9 days after onset of illness and correlates with viral load $>10^6$/mL and sub-genomic RNA rather that RNA detection per-se.
- Robust neutralizing antibody responses which are long lasted, likely similar to SARS-CoV-2. Older people make at least as good antibody responses as younger adults.
- SARS-CoV-2 infection boosts some memory cross-reactive B-cell epitopes acquired from prior infections with seasonal HCoV.

Acknowledgements

- Leo LM Poon, Sophie Valkenburg, Daniel Chu, Mahen Perera, Niloufar Kavian, Asmaa Hachem, Eric Lau, Ben Cowling, Kathy Leung, Joe Wu, School of Public Health / Pasteur Research Pole, University of Hong Kong
- Clinical collaboration: Dr Owen Tsang & Dr Mike Kwan Princess Margaret Hospital, Prof David Hui, Prince of Wales Hospital, Dr Susan Chiu, Queen Mary Hospital, Dr Wai-Hung Chan, Queen Elizabeth Hospital, Hong Kong
- Prof Jincun Zhao, State Key Laboratory for Respiratory Disease, National Clinical Research Center for Respiratory Disease, Guangzhou Institute of Respiratory Health, Guangdong
- Dr Jie Wu, Guangdong Provincial Centre for Disease Control and Prevention
- Research Funding: CEIRS Program, US NIAID Contract (contract no. HHSN272201400006C); Health and Medical Research Fund - Commissioned Research on the Novel Coronavirus Disease (COVID-19) (reference nos. COVID190126)