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Last updated by author(s): Dec 9, 2020

Reporting Summary

Nature Research wishes to improve the reproducibility of the work that we publish. This form provides structure for consistency and transparency in reporting. For further information on Nature Research policies, see our Editorial Policies and the Editorial Policy Checklist.

Statistics

For all statistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.

n/a	Confirmed	
☐	☒	The exact sample size (*n*) for each experimental group/condition, given as a discrete number and unit of measurement
☐	☒	A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
☐	☒	The statistical test(s) used AND whether they are one- or two-sided *Only common tests should be described solely by name; describe more complex techniques in the Methods section.*
☒	☐	A description of all covariates tested
☒	☐	A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
☐	☒	A full description of the statistical parameters including central tendency (e.g. means) or other basic estimates (e.g. regression coefficient) AND variation (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals)
☐	☒	For null hypothesis testing, the test statistic (e.g. *F*, *t*, *r*) with confidence intervals, effect sizes, degrees of freedom and *P* value noted *Give P values as exact values whenever suitable.*
☒	☐	For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
☒	☐	For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
☒	☐	Estimates of effect sizes (e.g. Cohen's *d*, Pearson's *r*), indicating how they were calculated

Our web collection on statistics for biologists contains articles on many of the points above.

Software and code

Policy information about availability of computer code

Data collection	FlexiWare v 8.1.3, Samtools v 1.9., Fastp v0.12, Nanozoomer, SWISS-Model, MacPyMol v 1.3.
Data analysis	Data analysis conducted using PRISM 7.0 or Microsoft Excel.

For manuscripts utilizing custom algorithms or software that are central to the research but not yet described in published literature, software must be made available to editors and reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Research guidelines for submitting code & software for further information.

Data

Policy information about availability of data

All manuscripts must include a data availability statement. This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A list of figures that have associated raw data
- A description of any restrictions on data availability

The raw data that support the findings of this study are available from the corresponding author upon reasonable request. Spike modeling based on PDB 6ACD.

Field-specific reporting

Please select the one below that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection.

☒ Life sciences ☐ Behavioural & social sciences ☐ Ecological, evolutionary & environmental sciences

For a reference copy of the document with all sections, see nature.com/documents/nr-reporting-summary-flat.pdf

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size	Sample sizes for experiments were determined by prior studies with coronaviruses that had power to detect changes in titer with a significance level of 0.05 (two-tailed). Gralinski, L.E., et al.mBio 4(2013).
Data exclusions	No data was excluded from the cacluation
Replication	In vitro experiments representative of at least 2 experiments with multiple samples per time point. In vivo experiment utilized multiple animal per group per time point and were from a single experiment (hamster) and 3 independent experiments (mice).
Randomization	Animals were assigned to groups at random. Cell for in vitro experiments were assigned at random.
Blinding	Investigators were not blinded during data collection or due to the limited capacity of workers able to complete high containment work. Data analysis was conducted by researcher who was not blinded.

Reporting for specific materials, systems and methods

We require information from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, system or method listed is relevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.

Materials & experimental systems

n/a	Involved in the study
☐	☒ Antibodies
☐	☒ Eukaryotic cell lines
☒	☐ Palaeontology and archaeology
☐	☒ Animals and other organisms
☒	☐ Human research participants
☒	☐ Clinical data
☒	☐ Dual use research of concern

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☒	☐ Flow cytometry
☒	☐ MRI-based neuroimaging

Antibodies

Antibodies used	RabbitSARS-CoV Spike (S) specific antibodies (Novus Biologicals #NB100-56578) Horseradish peroxidase (HRP)-conjugated anti-rabbit antibody (Cell Signaling Technology #7074S) SARS-CoV Nucleocapsid (N) specific antibodies were generated in house by Dr. Shinji Makino provided as gift.
Validation	Spike antibody- Orthogonal Validation- The target protein is examined with an antibody independent strategy and compared with results from an antibody-dependent strategy. A correlation between these two strategies indicates specificity between the antibody and its target protein. Examples of antibody independent techniques may include in situ hybridization, quantitative PCR, RNA-seq or mass spectrometry. Full length Spike protein transfected into UM92 cells was used as a positive control and an approximate 139 kDa band was observed. Dot Blot results using recombinant proteins for cross-reactivity testing revealed high reactivity to SARS-CoV-2 Spike Protein, 1000-1200 a.a. (NBP2-90973) and low/no reactivity towards MERS Spike 2 or H1N1 (NBP1-99041). HPR anti-rabit- This product is thoroughly validated with CST primary antibodies and will work optimally with the CST western immunoblotting protocol, ensuring accurate and reproducible results. SARS-CoV Nucleocapsid (N) specific antibodies (provided as a kind gift from Dr. Shinji Makino) - Previously published Harcourt et ao. Emerg Infect Dis. 2020 Jun;26(6):1266-1273. doi: 10.3201/eid2606.200516. Epub 2020 Jun 17. The plasmid, pBM302, (Das D. and Suresh MR, J Virol Methods. 2006 Nov;137(2):343-6), was used to express SARS-CoV N protein, with a C-terminal His6 tag, to high levels within the inclusion bodies of E.coli and the recombinant protein was purified from the inclusion bodies by nickel-affinity column chromatography under denaturing conditions. The recombinant SARS-CoV N protein was refolded by stepwise dialysis against Tris/phosphate buffer with decreasing concentrations of urea to renature the protein. The renatured, full-length, SARS-CoV N protein was used to immunize rabbits to generate an affinity-purified rabbit anti-SARS-CoV N polyclonal antibody.

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)	VeroE6 (gift from Ralph S. Baric, lab derivative of VERO C1008 ATCC CRL-1586); Calu3 2B4 (gift from Ralph Baric, lab derivative of ATCC HTB-55).
Authentication	None of the cells were authenticated
Mycoplasma contamination	VeroE6 and Calu3 2B4 cells are tested for mycoplasma contamination by RT-PCR periodically. Calu3 cells tested positive for mycoplasma contamination in July. Calu3 and Vero rederived from frozen stocks tested negative in August.
Commonly misidentified lines (See ICLAC register)	None

Animals and other organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research

Laboratory animals	Male Syrian hamsters (7-8 weeks old, 86–127g). Male and female K18-hACE C57BL/6J mice (strain: 2B6.Cg-Tg(K18-ACE2)2Prlmn/J, 5-9 wks old) were obtained from The Jackson Laboratory. The ABSL-3 room was kept between 68-74 degrees Farenheit with 30-60% and 12h-12h light cycles (6AM-6PM)
Wild animals	Study did not involve wild animals
Field-collected samples	Study did not involve field collected samples
Ethics oversight	All procedures were conducted under an animal protocol approved by the UTMB Institutional Animal Care and Use Committee and complied with USDA guidelines in an AAALAC-accredited lab. Animal Care and Use Committee at the Washington University School of Medicine (assurance number A3381–01)

Note that full information on the approval of the study protocol must also be provided in the manuscript.