

Supplementary Fig. 1: Sequencing data analysis. a) Schematic illustrating the general sequencing and bioinformatics workflow used to determine the virus ratios in Fig. 1e-g. For each virus mixture in the NEC, BEC, and lung explants samples, RNA was extracted from apical washes at 2 and/or 6 dpi and sequenced on the MinION sequencer using a modified version of the ARTIC protocol for SARS-CoV-2 sequencing (1200 bp amplicons with the midnight primer scheme, see methods for specific modifications)²⁶. Live GPU basecalling was performed using Guppy v.5.1.15 (Oxford Nanopore technologies) in high-accuracy mode and the downstream analysis was performed using a modified version of the ARTIC bioinformatics pipeline (https://artic.network/ncov-2019/ncov2019bioinformatics-sop.html). Briefly, input reads were filtered based on read length and then mapped to the Wuhan-Hu-1 reference genome (accession MN908947.3 [www.ncbi.nlm.nih.gov/bioproject/?term=MN908947.3]) using the 'artic minion' command. BAM alignment files from the ARTIC pipeline were then used for variant calling in longshot (v.0.4.4) with an input VCF file containing VOC Delta and Omicron-BA.1 mutations provided to call variants at specific nucleotide sites. The downstream analysis of VCF files was performed in R v.4.1.3 and involved filtering the VCF file for each sample to exclude mutations shared between both viruses in the mixture (shared, blue), non-lineage-defining mutations (non-LDM, grey), and any sites with overlapping mutations in both viruses that were difficult to call (other, green). Mutations called with a depth of coverage lower than 100 were also excluded from the downstream analysis (bottom left panel). Mutations in filtered VCF files (bottom middle panel) were then used to calculate the mean mutation frequency for each virus per sample. Finally, the mean \pm sd virus ratio was calculated for each time point (bottom right panel). b) Stacked bar plot showing the frequency of individual mutations called in the VCF file for the WT-614G and Omicron inoculum before (top) and after (bottom) filtering. c) Stacked bar plot showing the frequency of individual mutations in the filtered VCF files for all WT-614G and Omicron NEC samples (3 donors, 2 and 6 dpi). These values were used to calculate a mean mutation frequency for each virus per sample, which is shown in d).



Supplementary Fig. 2: Experimental outline for studies with Syrian hamsters and ferrets. Donor hamsters (n=6) were intranasally inoculated with either an Omicron-BA.1, -Delta or an Alpha-Delta mixture at iso-titer. A Competition studies in hamsters. Each donor was co-housed with one contact I hamster 1dpi. 4dpi after euthanasia of the donors, one contact II hamster was introduced to each contact I hamster. B Study outline for the ferret competition studies with Omicron-BA.1-Delta and Alpha-Delta mixtures. C Timeline for the ferret study with single-variant-inocula (either Omicron-BA.1 or Delta). 6dpi six donor animals, which were inoculated with Omicron-BA.1, were euthanized for determination of viral organ load in URT and LRT.



Supplementary Fig. 3: Body weight changes and survival rate of hamsters/ferrets in the competitive infection and transmission experiments. a) Survival of Syrian hamsters during competitive infection and transmission experiment between Alpha and Delta VOC. b) Survival of Syrian hamsters during competitive infection and transmission experiment between Delta and Omicron-BA.1 VOC. c) Percentages of body weight change in Syrian hamsters competitively inoculated with Alpha and Delta VOC. d) Percentages of body weight change in Syrian hamsters competitively inoculated with Delta and Omicron-BA.1 VOC. e) Percentages of body weight change in ferrets during single infection study with Delta VOC. f) Percentages of body weight change in ferrets during single infection study with Omicron-BA.1 VOC. Red star and arrow show timepoint of euthanasia (6 dpi) for six ferrets to analyze viral load distribution in organs. g) Percentages of body weight change in ferrets competitively inoculated with Delta and Omicron-BA.1 VOC. h) Percentages of body weight change in ferrets competitively inoculated with Delta and Omicron-BA.1 VOC. h) Percentages of body weight change in ferrets competitively inoculated with Delta and Omicron-BA.1 VOC. Alpha and Delta VOC. h) Percentages of body weight change in ferrets competitively inoculated with Delta and Omicron-BA.1 VOC. Note:

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Supplementary Fig. 4: Viral load in organs of Contact I and Contact II hamsters competitively inoculated with Delta and Omicron-BA.1 VOC at respective euthanasia timepoints Viral genome load in upper (URT) and lower (LRT) respiratory tract tissues of Syrian hamsters in the competitive transmission experiment between SARS-CoV-2 VOCs Delta and Omicron-BA.1. Syrian hamsters were inoculated with comparable genome equivalent mixture of Delta and Omicron-BA.1 VOC. Absolute quantification was performed by RT–qPCR analysis of tissue homogenates of Contact I and Contact II hamsters in relation to a set of defined standards. Tissue samples were collected at euthanasia (Euth.). Pie chart colors illustrate the ratio of variants detected in each sample at the indicated dpi or days post contact (dpc). Pie chart sizes are proportional to the total viral genome copies reported above. Grey pies indicate values below the LOD ($<10^3$ viral genome copies per mL).



Supplementary Fig. 5: Competitive infection of hamsters with SARS-CoV-2 Delta and Alpha Six donor hamsters were each inoculated intranasally with 10^{4.625} TCID₅₀ determined by back titration and composed of a mixture of SARS-CoV-2 Alpha (dark blue) and Delta (purple) at 1.95:1 ratio determined by back-titration of the original single virus amounts used in the experiment. Donor hamsters, contact I and II hamsters were co-housed sequentially as shown in Supplementary Data Fig.2. Nasal washings were performed daily from 1-9 dpi and afterwards every two days until 21 dpi. Each pie chart illustrates the ratio of the respective viruses in nasal washings for each sampling day. Total genome copies/mL are indicated above or below the respective pies. Hamster silhouettes are colored according to the dominant variant (>66%) detected in the latest sample of each animal. Black crosses indicate the respective animal was already dead.



Supplementary Fig. 6: Competitive infection of ferrets with SARS-CoV-2 Alpha and Delta Six donor ferrets were each inoculated with 10^5 TCID₅₀ determined by back titration and composed of a mixture of Alpha (dark blue) and SARS-CoV-2 Delta (purple) at a 1.33:1 ratio determined by back-titration of the original single virus amounts used in the experiment. Donor and Contact ferrets were co-housed sequentially as shown in Supplementary Data Fig.2. Pie charts illustrate the ratio of either SARS-CoV-2 Alpha or SARS-CoV-2 Delta detected in nasal washings of the donor or contact ferrets in the respective ferret groups at indicated dpi. Viral genome copies/mL are shown above or below respective pie charts; Grey pies indicate values below the LOD (< 10^3 viral genome copies per mL). Coloring of the ferret silhouettes refers to the predominant SARS-CoV-2 variant (>66%) detected in the latest sample of the respective animal.



Supplementary Fig. 7: ELISA and VNT100 of sera received from competitive infection experiments with Alpha and Delta in hamsters/ferrets. Blue dots represent neutralization of Alpha variant, purple dots represent neutralization of the Delta variant in the respective animal according to the highest dilution where virus neutralization was visible (left Y-Axis). Black dots show RBD-ELISA-reactivity of animal sera at respective euthanasia timepoint (right Y-Axis) (a) VNT100 and RBD-ELISA from animal sera of the Alpha vs Delta competitive infection and transmission experiment in hamsters (b) VNT100 and RBD-ELISA from animal sera of the competitive infection and transmission experiment with Delta and Alpha VOC in ferrets.



Supplementary Fig. 8: Viral load in organs of Donor, Contact I and Contact II hamsters competitively inoculated with Alpha and Delta VOC at respective euthanasia timepoints. Viral genome load in upper (URT) and lower (LRT) respiratory tract tissues of Syrian hamsters in the competitive transmission experiment between SARS-CoV-2 VOCs Alpha and Delta. Syrian hamsters were inoculated with comparable genome equivalent mixture of either Alpha or Delta VOC. Absolute quantification was performed by RT–qPCR analysis of tissue homogenates of donor, contact I and contact II hamsters in relation to a set of defined standards. Tissue samples were collected at euthanasia (Euth.). Pie chart colors illustrate the ratio of variants detected in each sample at the indicated dpi or days post contact (dpc). Pie chart sizes are proportional to the total viral genome copies reported above. Grey pies indicate values below the LOD (<10³ viral genome copies per mL).



b Competition experiments with Delta, Omicron, S-Delta and S-Omicron



Supplementary Fig. 9: Delta spike mutations drive enhanced fitness in humanized mice. a) hACE2-KI mice (7 to 16 week-old male) were intranasally inoculated with $10^{4.3}$ tissue culture infectious dose 50 (TCID₅₀) of Delta or Omicron isolates. The left graph reports the body weight loss for each of the hACE2-KI in Fig. 3a. The right graph depicts the viral copies in brain and olfactory bulb samples quantified using E-gene probe-specific RT–qPCR. Data are mean ± s.d. from the indicated number of biological replicates from a single experiment. Statistical significance was determined using an unpaired two-tailed Student t-test; **P < 0.01. **b**) hACE2-KI mice (7 to 19 week-old female, n=6/group) were intranasally inoculated with 10⁴ TCID50 of a 1:1 mix of Delta and Omicron or SARS-CoV-2^{S-Delta} and SARS-CoV-2^{S-Omicron}. The graph on the left shows the body weight loss for each of the inoculated animal. The graph on the right shows the histopathological score in these mice. Data are mean ± s.d. from the indicated number of biological replicates from a single experiment.



Supplementary Fig. 10: mRNA vaccine induced reduction in replication and pathogenesis of SARS-CoV-2 clones in K18-hACE2 transgenic mice. a) Female K18-hACE2 transgenic mice (7 to 15 weeks old, n=16 mice) were immunized intramuscularly with a single dose of 1 µg of mRNA-Vaccine Spikevax (Moderna). After two weeks the neutralizing antibody titers against SARS-CoV- 2^{D614G} were determined. Later, mice were (n=8 mice/group) intranasally inoculated with 10⁴ tissue culture infectious dose 50 (TCID₅₀) of SARS-CoV- 2^{D614G} , SARS-CoV- $2^{S-Delta}$ and SARS-CoV- $2^{S-Omicron}$. b) The body weight change and c) the clinical scores of the mice were monitored daily. d) Histopathological scores were given to evaluate the severity of the lung pathology. e) Brain and olfactory bulb samples of the infected mice were collected at 2 or 6 days post-infection (dpi) to determine the viral load (n=4 for each group). Viral RNA-dependent RNA polymerase (RdRp) gene copies of brain and olfactory bulb tissues were quantified using probe-specific RT–qPCR. f) Infectious virus titers from the brain samples were determined using TCID₅₀ assays in VeroE6/TMPRSS2 cells. g) Virus neutralization capacities of the serum collected from infected mice at 6 dpi are tested against SARS-CoV- $2^{S-Delta}$ and SARS-CoV- $2^{S-Omicron}$ clones. Each dot in the graphs represents one animal, and the bars show mean values +/- SD. The color key in b also applies to c, d, e and f. Statistical significance was determined using ordinary two-way ANOVA (a–d) and P values were adjusted using Tukey's multiple-comparison test; *P < 0.05, **P < 0.01, ***P < 0.001, ****P < 0.001. Data were obtained from one experiment. Each data point represents one biological replicate.

Supplementary Information Guide

- 1. Supplementary Information Table 1: List of the viruses used in the experiments.
- 2. **Supplementary Information Table 2:** Histopathological characterization of hACE-2-KI mice infected with Delta or Omicron-BA.1 isolates
- 3. **Supplementary Information Table 3:** Histopathological characterization of K18hACE2 mice infected with SARS-CoV-2^{S-Delta} or SARS-CoV-2^{S-Omicron}
- 4. Supplementary Information Table 4: Primer Pairs for Delta and Omicron.
- 5. **Supplementary Information Table 5:** Primers and probes for viral RNA detection via RT-qPCR.

SARS-CoV-2	Reference	Model	Inocula
		hACE2-K18 mice	SARS-CoV-2 ^{D614G} Single
			SARS-CoV-2 ^{D614G} Single
CARDO O-V OD614G	https://doi.org/10.1038/s		SARS-CoV-2 ^{D614G} vs Delta
SARS-Cov-2-	41586-021-03361-1	hNEC/hBEC	SARS-CoV-2 ^{D614G} vs Omicron-BA.1
			SARS-CoV-2 ^{D614G} vs SARS-CoV-2 ^{S-Delta}
			SARS-CoV-2 ^{D614G} vs SARS-CoV-2 ^{S-Omicron}
Alpha	EPI_ISL_751799	Hamster and ferret	Alpha vs Delta
		Ferret	Delta Single
		Hometor and ferrat	Alpha vs Delta
		Hamster and terret	Delta vs Omicron-BA.1
	EPI_ISL_1760647		Delta Single
Delta			SARS-CoV-2 ^{D614G} vs Delta
			Delta vs Omicron-BA.1
			Delta vs SARS-CoV-2 ^{S-Delta}
		LACE2 KI mice	Delta Single
	EFI_IOL_2000400		Delta vs Omicron-BA.1
		Ferret	Omicron-BA.1 Single
	EF1_13L_0909000	Hamster and ferret	Delta vs Omicron-BA.1
		hACE2-KI mice	Omicron-BA.1 Single
Omicron-RA 1			Delta vs Omicron-BA.1
	EDI 191 7062525		Omicron-BA.1 Single
	EFI_13L_1002323		SARS-CoV-2 ^{D614G} vs Omicron-BA.1
			Delta vs Omicron-BA.1
			Omicron-BA.1 vs SARS-CoV-2 ^{S-Omicron}
		hACE2-KI mice	SARS-CoV-2 ^{S-Delta} vs SARS-CoV-2 ^{S-Omicron}
	Backbone:		SARS-CoV-2 ^{S-Delta} Single
	Sequence: Wuhan-Hu-1 NC 045512.2		SARS-CoV-2 ^{D614G} vs SARS-CoV-2 ^{S-Delta}
SAKS-00V-2	Spike:		Delta vs SARS-CoV-2 ^{S-Delta}
	EPI_ISL_5769545		SARS-CoV-2 ^{S-Delta} vs SARS-CoV-2 ^{S-Omicron}
		PCLS	SARS-CoV-2 ^{S-Delta} vs SARS-CoV-2 ^{S-Omicron}
		hACE2-KI mice	SARS-CoV-2 ^{S-Delta} vs SARS-CoV-2 ^{S-Omicron}
	Backbone: NCBI Reference		SARS-CoV-2 ^{S-Omicron} Single
	Sequence: Wuhan-Hu-1 NC_045512.2		SARS-CoV-2 ^{D614G} vs SARS-CoV-2 ^{S-Omicron}
SARS-COV-2	Snike	TINEC/TIBEC	Omicron-BA.1 vs SARS-CoV-2 ^{S-Omicron}
	EPI_ISL_7062525		SARS-CoV-2 ^{S-Delta} vs SARS-CoV-2 ^{S-Omicron}
		PCLS	SARS-CoV-2 ^{S-Delta} vs SARS-CoV-2 ^{S-Omicron}

	Delta -Infection									Omicron Infection												
Days post-infection		dj	bi 2			dp	si 4			dş	i 2			dj	pi 4							
Mouse	K1 #13	KI #14	KI#15	KI#16	KI #17	KI #18	KI#19	KI #20	KI #21	KI#22	KI#23	KI #24	KI #25	KI#26	KI #27	KI #28						
Overview																						
Alveolar edema	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Emphysema	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Atelectasis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Higher magnification																						
Inflammation score																						
Alveolar infiltrates	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Grade	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Predominant inflammatory cell type	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Interstitial infiltrates	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0						
Grade	0	0	0	0	0	4	0	0	0	0	0	0	0	0	0	0						
Predominant inflammatory cell type	0	0	0	0	0	M + L	0	0	0	0	0	0	0	0	0	0						
Peribronchial infiltrates	0	1	1	0	3	1	1	0	0	1	0	0	0	0	0	0						
Grade	0	1	1	0	2	2	2	0	0	1	0	0	0	0	0	0						
Predominant inflammatory cell type	0	M + L	M + L	0	M + L	M + L	M + L	0	0	M + L + N	0	0	0	0	0	0						
Necrotizing bronchitis	0	3	1	0	3	0	1	0	0	0	0	0	0	0	0	0						
Grade	1	2	2	0	2	1	2	2	1	1	1	0	1	1	1	1						
Predominant inflammatory cell type	M	M	M + L	0	M	M	M	M	M	M	M	0	M	M	M	M						
Intrabronchial mucus increase	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Perivascular infiltrates	1	0	0	1	0	0	0	1	0	1	0	0	0		0	0						
Grade	1	0	0	1	0	0	0	1	0	1	0	0	0	1	0	0						
Predominant inflammatory cell type	M + L	0	0	M + L	0	0	0	M + L	0	M + L	0	0	0	L	0	0						
Vascular lesions																						
Intravascular rolling inflammatory cells	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0						
Endotheliitis	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0						
Vasculitis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Hyaline thrombi	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Hemosiderin	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Arterial media hypertrophy/ hyperplasia	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Vascular occlusion	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Diffuse alveolar damage (DAD)/Necrosis																						
Necrosis alveolar epithelial cells (AEC)	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
DAD	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Intraalveolar fibrin	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Varia																						
Hypertrophy/hyperplasia bronchiolar																						
epithelium	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Hyperplasia /hypertrophy type II pneumocytes	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Atypical cells/syncytia	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
Pleura	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
BALT	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0						
TOTAL SCORE	3	7	5	2	12	9	6	4	1	5	1	0	1	3	1	1						

		0,	82		
KI#1	KI#2	KI #3	KI #4	KI#S	KI#E
0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	1	0	0
0	0	0	2	0	0
0	0	0	м	0	0
0	1	1	0	0	0
0	1	1	0	0	0
0	M + L	M + L + N	0	0	0
0	1	3	1	0	0
0	1	2	1	0	1
0	M	M	M	0	M
0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	0	0	0
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0	4	7	5	0	1

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MI #7	N1 40	dg March	ni 2	101 101 1	10.013
KI #7	KI #8	KI \$9	KI#10	KI#11	KI #12
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0	0	0	0	0	0
0	0	0	0	0	0
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0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	0	0	1
0	0	0	0	0	Mal
0	0	0	0	0	0
1	1	1	1	1	1
M	M	M	M	M	M
0	0	0	0	0	0
0	1	1	0	0	1
0	1	2	0	0	1
0	M + L	M + L	0	0	M + L + N
			-		
0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	0	0	0
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0	0	0	0	0	0
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0	0	0	0	0	0
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0	0	0	0	0	0
0	0	0	0	0	0
0	0	0	0	0	0
1	3	4	1	1	5

									614G	49							504						Soluta								54								Janieron							
Vaccinated											Unvaco	inated							Vaci	nated				Unvacionated										Vac	inated				Unvaccinated							
Days post-infection	dpi 2 dpi 6					dqi 6			4	si 2			69	46			وك	12			dpi	6			69	2			dgi	6			dpi 2			dp	16			de	12			dpi 6		
Mouse	K18 #1	818 52	X18 f2	K18 P	1 818 65	X18 P	6 K18 F	7 818 68	K18 #12	818 614	818 #15	K18 #16	111.09	K18 #22	818 #11	K18 #12	K18 #17	818 818	K18 #19	K18 #20	K18 f21	K18 #22	K18 #22	K18 #24	K18 #25	818 625	818 #22	K18 #28	K18 #29	K18 #30	K18 #31 K1	3 #22 K18	#22 818 #2	K18 #29	K18 P40	K18 #22	K18 #34	K18 #35	K18 #26	K18 P41	K18 M2	818 662	18 644 8	8 645 8387	M6 8181	M7 818 M8
Querview																																														
Alveolar edema	0	0	0	0	0	0	0	0	0	0	0	0	2	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	2	2	2	0 0	0	0	0	0	1	0	0	0	0	0	0	0 1	0	0
Emphysema	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0 0		0	0	0	0	0	0	0	0	0	0	0 0	0	0
Atelectaria	0	0	0	0	Ó	0	0	0	0	0	Ó	0	2	1	2	2	0	0	0	0	0	0	0	0	1	1	1	1	2	2	0	2 0	0	0	0	1	Ó	0	1	0	0	0	1	1 1	0	1
Haber materification																																														
Inflammation score																																										_				
Alveolar infiltrates	0	0	0	0	0	0	0	0	0	0	0	0	2	1	2	2	0	0	0	0	0	0	0	0	1	0	0	0	1	2	2	2 0	0	0	0	1	0	0	0	0	0	0	1	0 0		0
Grade	0	0	0	0	0	0	0	0	0	0	0	0	2	2	2	2	0	0	0	0	0	0	0	0	2	0	0	0	2	2	2	2 0	0	0	0	1	0	0	0	0	0	0	4	0 0	0	0
Predominant inflammatory cell type	0	0	0	0	0	0	0	0	0	0	0	0	M + N	M+N	M+N	M+N+L	0	0	0	0	0	0	0	0	M+L	0	0	0	M+N+L	M+L+N	M+N+L 5	(-N 0	0	0	0	M	0	0	0	0	0	0	M+N	0 0	0	0
Interctical infitrates	0	0	0	0	0	0	0	0	0	0	0	0	2	1	2	2	0	0	0	0	0	0	0	0	1	1	1	1	2	2	2	2 0		0	0	2	0	0	1	0	0	0	1	1 1	0	1
Grade	0	0	0	0	0	0	0	0	0	0	0	0	2	2	3	4	0	0	0	0	0	0	0	0	2	2	ž	ž	4	4	4	4 0		0	0	2	0	0	3	0	0	0	3	3 1	0	2
Predominant inflammatory cell type	ô	Ó	0	0	Ó	0	0	ô	0	ŷ	Û.	0	M+1	M+L	1	M+L+N	0	ō	0	0	0	Û	0	0	M+L	M+1	M+L+N	M+1	M+1	M+L	M+L 2	d+1 1	. Ó	Ó	Ô	M+1	Û.	0	M+L	ò	ů	0	M+L 8	4+1 M+	ė	M + L
Peribranchial infitrates	0	0	0	- 2	1	0	1	1	0	0	Ó	0	1	1	0	2	1	1	1	0	1	2	2	2	0	1	1	1	2	2	2	2 3	1	0	0	2	2	1	1	0	0	0	1	1 1	0	1
Grade	0	0	0	- 2	2	0	1	1	0	0	Ó	0	2	3	0	2	2	1	1	0	1	2	4	2	0	1	1	1	2	2	2	4 M+1	+N 1	0	0	2	2	1	1	0	0	0	2	2 2	0	2
Predominant inflammatory cell type	0	0	0	M+L+	N M+L	0	M + L	M+L	0	0	Ó	0	M + L	M.+L	0	M+L	M+L	M = L	M+L	0	M+L	M+L+N	M+L	M + L + N	0	M+L	M+L	M + L	M + L	M + L	M+L 8	4+L C	M+1	0	0	M + L + N	M+L	M + L	M+L	0	0	0	M+L 8	4+L M+	-L 0	M + L
Negrotizing branchitis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0		0	0	0	0	0	0	0	0	0	0	0	0 0	0	0	0	0	0	0	0	0		0	0	0 0	0	0
Grade	0	1	1	1	1	1	1	1		1	1	1	1	1	0	1	- 0	2	1	1	1	1	0	1	1	1	1	1	1	2	2	1 0	0	0	1	0	0	1	0	1	1	1	1	2 2	0	1
Predominant inflammatory cell type	0	M	M	M	M	м	M	M	0	M	M	M	M	M	0	M	0	M	M	м	M	M	0	M	M	M	M	M	M	M	M	M 0	0	0	M	0	Ó	M	0	M	M	M	M	M M	0	- M
intrabranchial mucus increase	0	2	0	0	Ó	0	0	2	0	0	Ó	0	0	0	0	2	0	0	Ó	- 0	Ó	Ó	0	Ó	Ó	0	Ó	0	Ó	Ó	Ó	0 0	0	0	0	0	Ó	0	0	0	0	0	0	0 0	0	0
Perfeascular infiltrates	0	0	0	3	1	1	1	0	0	0	2	0	3	1	3	2	1	1	1	1	Ó	2	2	1	2	1	1	1	2	2	2	2 0	1	0	1	1	1	0	3	0	0	1	0	3 3	0	1
Grade	0	0	0	4	1	1	1	0	0	0	2	0	2	3	2	2	1	1	2	1	Ó	4	2	1	1	2	2	2	2	2	4	2 0	2	0	2	2	1	0	2	0	0	1	0	3 1	0	2
Predominant inflammatory cell type	0	0	0	M+L+	N M+L	M+1	. M+L	. 0	0	ů.	M.+L	0	M+L	M.+L	M = L	M+L	M+L	M = L	M+L	M = L + N	0	M+L+N	M+L	M+L	M+L	M+L	M+L	M+L	M+L	M+L	M+L 8	4+L C	M+L+	0	M+L	M + L + h	M+L	0	M+L	0	ů.	M+L	0 7	3+1 M+1	.+£ 0	- M + L
Vascular locioes																																										_	_	_		
intravacular rolling inflammatory cells	1	ů.	0	2	ů.	0	1	Ó	0	0	Ó	1	1	3	3	1	0	1	1	0	0	2	Ó	0	2	2	2	1	2	Ó	2	2 1	1	ů.	0	1	Ó	1	1	0	ů.	0	0	1 1	0	2
Endothelitis	1	0	0	0	Ó	1	0	0	0	0	Ó	0	0	1	1	2	0	0	Ó	0	Ó	Ó	0	Ó	1	1	Ó	1	2	1	1	0 0	0	0	0	1	Ó	0	Ó	0	0	0	0	0 0	0	0
Vascultis	0	0	0	1	Ó	0	0	0	0	0	Ó	0	0	0	0	Ó	0	0	Ó	0	Ó	Ó	0	Ó	2	0	Ó	1	Ó	Ó	Ó	0 0	0	0	0	0	Ó	0	Ó	0	0	0	0	0 0	0	0
Hyatine thromos	0	0	0	0	Ó	0	0	0	0	0	Ó	0	0	0	0	Ó	0	0	Ó	0	Ó	Ó	0	Ó	Ó	0	Ó	0	Ó	Ó	Ó	0 0	0	0	0	0	Ó	0	Ó	0	0	0	0	0 0	0	0
Hemosiderin	0	0	- 0	0	0	- 0	0	0	- 0	0	0	- 0	ů.	÷	ů.	0	- 0	ů.	0	÷	0	0	0	0	0	0	0	0	0	0	0	0 0	0	0	0	0	0	0	0	ů.			0	0 0		0
Arterial media hypertraahy/ hyperalasia	0	0	-	0	0		0	0		0	0	- 0	0		0	0	- 0	0	0		0	0	0	0	0	0	0	0	0	0	0	0 0		0	0	0	0	0	0	0		- 0	- 0	0 0		- 0
Vascular occlusion	0	0	•	0	0	•	0	0	•	0	0	•	0	0	0	0	•	0	0		0	0	0	0	0	D	0	0	0	0	0	0 0	0	0	D	D	0	0	0	0	0	0	0	0 0	0	- 0
Diffuse alveolar damage (DAD)/Necrock																																										_	_			_
Necrosis alvectar epithelial cells (ASC)	0	Ó	0	ů.	0	0	ů.	ů.	0	ů.	0	0	0	0	ů.	0	0	0	0	- 0	0	0	0	0	0	0	0	0	ů.	0	0	0 0	1 0	ů.	0	0	0	0	0	0	ů.	ů.	0	0 0	0	. 0
DAD	0	0	0	0	Ó	0	0	0	0	0	Ó	0	0	0	0	Ó	0	0	Ó	0	Ó	Ó	0	Ó	Ó	0	Ó	0	Ó	Ó	Ó	0 0	0	0	0	0	Ó	0	Ó	0	0	0	0	0 0	0	0
Intralveolar fibrin	0	0	0	0	0	0	0	0	ů.	0	0	0	٥	0	0	0	0	0	0	÷	0	0	0	0	0	0	0	0	0	0	0	0 0	1 0	0	0	0	0	0	0	0	0		0	0 0	0	0
Varia																																										_	_			_
Hypertrophy/hyperplasia bronchiolar epithelium	0	Ó	0	ů.	0	0	ů.	1	0	ů.	0	0	0	0	ů.	0	0	0	0	1	ů.	0	0	0	0	0	0	0	ů.	0	0	0 0	1 0	ů.	0	0	0	0	0	0	ů.	ů.	0	1 0	0	. 0
Hyperplacia /hypertrophy type II pneumocytes	0	Ó	0	ů.	0	0	ů.	ů.	0	ů.	0	0	2	2	2	2	0	0	0	- 0	ů.	0	0	0	1	1	1	1	2	2	2	2 0	1 0	ů.	0	1	0	0	1	0	ů.	ů.	1	1 1	0	. 1
Atypical cells/leyncytia	ů.	Ó	0	ů.	0	0	ů.	ů.	0	ů.	0	0	0	0	ů.	0	0	0	0	- 0	ů.	0	0	0	0	0	0	0	ů.	0	0	0 0	1 0	ů.	0	0	0	0	0	0	ů.	ů.	0	0 0	0	. 0
Philip	0	0	•	0	0		0		0	0	ů.		ů.	0	0	U		d	U		ů.	U	U	U	U	U	U	é	0	U	U	0 0		0			ů.	é	U	d	u	<u>u</u>	u	0 0	- 0	
EALT	0	0	0	0	0	0	0	0	0	0	Ú.	0	ů.	¢.	ů.	U	0	d d	U	- 0	Ó	U	U	U	U	U	U	Ó	0	U	U	0 0		0	0	0	Ú.	Ó	U	d d	u	U	u	0 0		0
TOTAL SCORE																							14		101	16	1.4	1.4				44 4				• 34							75	78 8 12		

Spike-Variant	Primer name	Primer sequence (5' to 3')	Amplification product (bp)	PCR Template (cDNA or YAC from previous clones)					
	WU-19-F	GGAGTCACATTAATTGGAGAAGC	4507	C-D0140					
	CoV2-Sp-T19R-R	GTATGCAGGGGGTAATTGAGTTCTGGTTcTAAGATTAACACACTGACTAG	1557	3000140					
	CoV2-Sp-T19R-F	CTAGTCAGTGTGTTAATCTTAgAACCAGAACTCAATTACCCCCTGCATAC	200	C-D0140					
	CoV2-Sp-T95I-R	TATTATGTTAGACTTCTCAaTGGAAGCAAAATAAACACCATCATTAAATG	209	3000140					
	CoV2-Sp-T95I-F	TTTAATGATGGTGTTTATTTTGCTTCCAtTGAGAAGTCTAACATAATAAG	004	0-04420					
	CoV2-Sp-E156d-E157d- R158G-R1+R2	GCAATTATTCGCACTAGAATAAACTCcACTTTCCATCCAACTTTTGTTG	234	SpG142D					
SARS-COV-2S-Della	CoV2-Sp-E156d-E157d- R158G-F1+F2	CAAAAACAACAAAAGTTGGATGGAAAGTgGAGTTTATTCTAGTGCG	1100	Sol 452D					
	CoV2-Sp-T478K-R	CCTTCAACACCATTACAAGGTtTGCTACCGGCCTGATAGATTTCAGTTG	1199	SpL452R					
	CoV2-Sp-T478K-F	CAACTGAAATCTATCAGGCCGGTAGCAaACCTTGTAATGGTGTTGAAGG	4400	SpD614G					
	CoV2-Sp-D950N-R	GCTTGTGCATTTTGGTTGACCACATtTTGAAGTTTTCCAAGTGCACTTGC	1400	3pD014G					
	CoV2-Sp-D950N-F	GCAAGTGCACTTGGAAAACTTCAAaATGTGGTCAACCAAAATGCACAAGC	4555	C-D0140					
	WU-22-R	TCATGTTCAGAAATAGGACTTGTTG	1000	3pD014G					
	WU-19-F	GGAGTCACATTAATTGGAGAAGC	4000	C-D0140					
	WU-60-R	TTGTTCGCGTGGTTTGCCAAG	1203	3pD014G					
	WU-61-F	CTTGGAATGCTGATCTTTATAAGC	1001	cDNA of Omicron for Omicron spike and OmNTD; SpD614G					
	WU-62-R	TAGAAAAGTCCTAGGTTGAAGATAAC	1221	for OmRBD and OmCS					
	WU-63-F	TTCGGCTTTAGAACCATTGGTAG	1200	cDNA of Omicron for Omicron spike and OmRBD; SpD614G					
Stars Superior	CoV2-D614G-R	GCAACAGGGACTTCTGTGCAGTTAACACCCTGATAAAGAACAGCAACCTG	1209	for OmNTD and OmCS					
SARS-Cov-2	CoV2-D614G-F	CAGGTTGCTGTTCTTTATCAGGGTGTTAACTGCACAGAAGTCCCTGTTGC	945	cDNA of Omicron for Omicron spike and OmCS ; SpD614G					
	WU-64-R	AACAGTGCAGAAGTGTATTGAGC	615	for OmNTD and OmRBD					
	WU-65-F	TGATTGCCTTGGTGATATTGCTG	1000	cDNA of Omicron for Omicron spike; SpD614G for					
	WU-66-R	CAACTGGTCATACAGCAAAGCAT	1206	OmNTD, OmRBD, OmCS					
	WU-67-F	GACATCTCTGGCATTAATGCTTC	077	S=D614C					
	WU-22-R	TCATGTTCAGAAATAGGACTTGTTG	0//	SpD614G					

Assay Name	Oligo name	Sequence (5´- 3´)	Conc.	Position
	Alpha-S-22011-F	AAA GTT GGA TGG AAA GTG AGT TCA	10 µM	22011
SARS-CoV-2-Alpha-S assay	SARS2-S-22132R	CCT AAG ATT TTT GAA ATT ACC CTG T	10 µM	22132
	SARS2-S-22074FAM	FAM- TCT CTC AGC CTT TTC TTA TGG ACC T -BHQ1	5 µM	22074
	Delta-S-22011-F	AAA GTT GGA TGG AAA GTG GAG	10 µM	22011
SARS-CoV-2-Delta-S assay	SARS2-S-22132R	CCT AAG ATT TTT GAA ATT ACC CTG T	10 µM	22132
	SARS2-S-22074FAM	FAM- TCT CTC AGC CTT TTC TTA TGG ACC T -BHQ1	5 µM	22074
	O-S22172-F	TATTCTAAGCACACGCCTATTATAG	10 µM	22172
SARS-CoV-2-Omicron-BA.1-S assay	O-S22280-R	TAGTGATGTTAATACCTATTGGCAAATC	10 µM	22280
	O-S22202-FAM	FAM-CGTGAGCCAGAAGATCTCCCTC-BHQ1	5 µM	22202