

Fig S1. Sequence alignment of the variable regions of antibodies. The CDR regions of each antibody were aligned using the IgBLAST website, and it was confirmed that the five cell lines secrete distinct monoclonal antibodies.

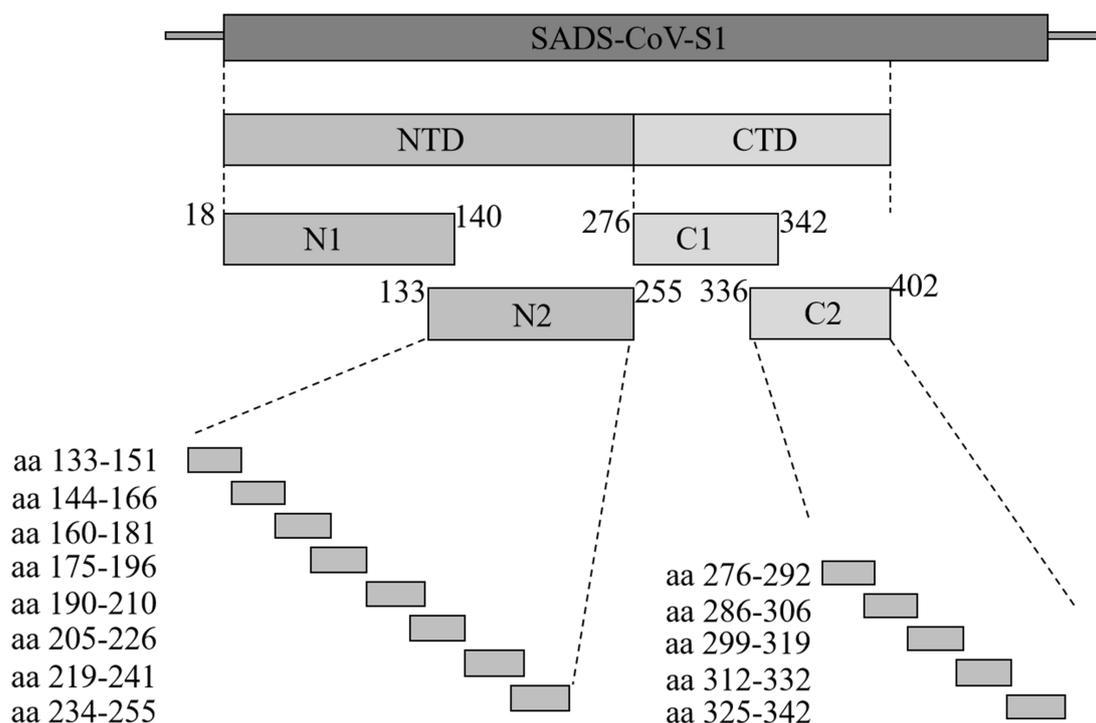


Fig S2. Truncation strategy of the S1 protein. In the first truncation, it was divided into four segments, namely N1, N2, C1 and C2. According to the results of the first epitope identification, N2 was truncated into eight segments, and C1 was truncated into five segments.

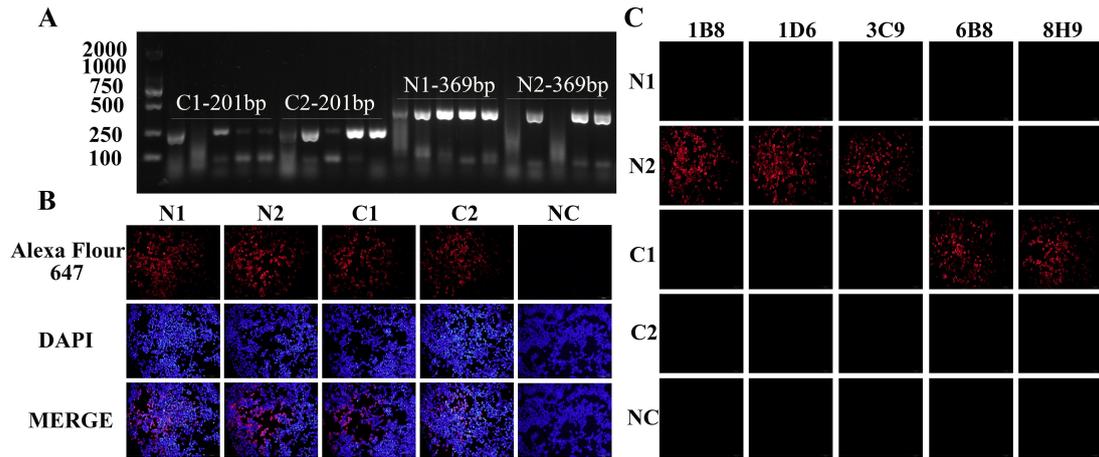


Fig S3. Expression of the initially truncated proteins and identification of B-cell epitopes. (A) PCR verification of the bacterial liquid containing truncated fragments. The bands of N1 and N2 are approximately at 369bp, and the bands of C1 and C2 are approximately at 201bp. (B) Verification of the expression of truncated proteins. Verification results indicated that all four truncated proteins were successfully expressed. NC indicates cells transfected with the pcDNA3.1(+) empty vector as a negative control. (C) Identification of the antibody recognition region by IFA. mAbs 1B8, 1D6, and 3C9 exhibited specific reactivity to the N2 fragment, while mAbs 6B8 and 8H9 demonstrated binding to the C1 segment. NC indicates cells transfected with the pcDNA3.1(+) empty vector as a negative control.

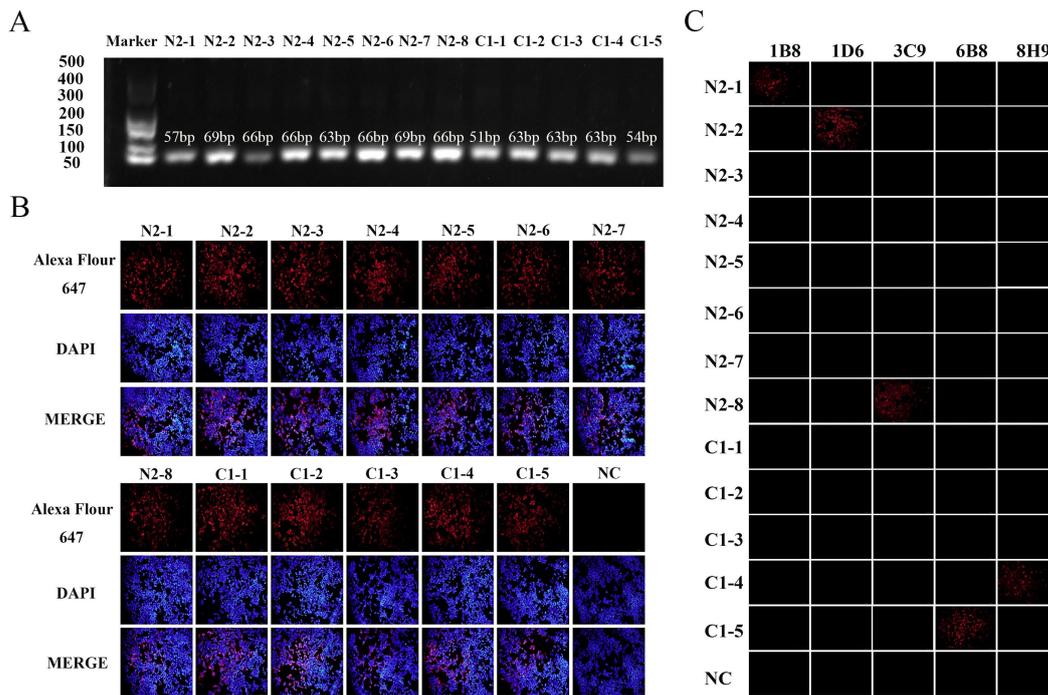


Fig S4. Expression of second-round truncated proteins and B-cell epitope mapping. (A) Bacterial colony PCR validation of truncated fragments. Thirteen truncated fragments exhibited bands approximately 60bp in size. (B) Expression verification of truncated proteins. All 13 truncated proteins were successfully expressed. NC indicates cells transfected with the pcDNA3.1(+) empty vector as a negative control. (C) Antibody recognition domains identified by indirect immunofluorescence assay. Antibodies 1B8, 1D6, and 3C9 recognized the N2-1, N2-2, and N2-8 fragments, respectively, while antibodies 8H9 and 6B8 recognized the C1-4 and C1-5 fragments, respectively. NC denotes cells transfected with the pcDNA3.1(+) empty vector as a negative control.

	N2-1	N2-2
SADS-CoV/HNNY/2023	QFM	144G
SADS-CoV/Guangxi/2021	QFM	144G
SADS-CoV/GDLX/2019	QFM	144G
SADS-CoV/CH/FJWT/2018	QFM	144G
SADS-CoV/GDZJ02/2018	QFM	144G
SADS-CoV/CH/JX1/2018	QFM	144G
SADS-CoV/CH/JX2/2018	QFM	144G
SADS-CoV/CH/JX/2018	QFM	144G
SADS-CoV/CN/GDST/2017	QFM	144G
SADS-CoV/CN/GDDE/2017	QFM	144G
SADS-CoV/CN/GDDCD/2017	QFM	144G
SADS-CoV/CN/GDGL/2017	QFM	144G
SADS-CoV/CN/GDWT/2017	QFM	144G
SADS-CoV/CN/GDLS/2017	QFM	144G
SADS-CoV/GDGL01/2016	QFM	144G
		N2-8
SADS-CoV/HNNY/2023	TVL	234S
SADS-CoV/Guangxi/2021	TVL	234S
SADS-CoV/GDLX/2019	TVL	234S
SADS-CoV/CH/FJWT/2018	TVL	234S
SADS-CoV/GDZJ02/2018	TVL	234S
SADS-CoV/CH/JX1/2018	TVL	234S
SADS-CoV/CH/JX2/2018	TVL	234S
SADS-CoV/CH/JX/2018	TVL	234S
SADS-CoV/CN/GDST/2017	TVL	234S
SADS-CoV/CN/GDDE/2017	TVL	234S
SADS-CoV/CN/GDDCD/2017	TVL	234S
SADS-CoV/CN/GDGL/2017	TVL	234S
SADS-CoV/CN/GDWT/2017	TVL	234S
SADS-CoV/CN/GDLS/2017	TVL	234S
SADS-CoV/GDGL01/2016	TVL	234S
	C1-4	C1-5
SADS-CoV/HNNY/2023	VNP	325G
SADS-CoV/Guangxi/2021	VNP	325G
SADS-CoV/GDLX/2019	VNP	325G
SADS-CoV/CH/FJWT/2018	VNP	325G
SADS-CoV/GDZJ02/2018	VNP	325G
SADS-CoV/CH/JX1/2018	VNP	325G
SADS-CoV/CH/JX2/2018	VNP	325G
SADS-CoV/CH/JX/2018	VNP	325G
SADS-CoV/CN/GDST/2017	VNP	325G
SADS-CoV/CN/GDDE/2017	VNP	325G
SADS-CoV/CN/GDDCD/2017	VNP	325G
SADS-CoV/CN/GDGL/2017	VNP	325G
SADS-CoV/CN/GDWT/2017	VNP	325G
SADS-CoV-CN-GDLS-2017	VNP	325G
SADS-CoV-GDGL01-2016	VNP	325G

Fig S5. Conservation analysis of antibodies. Sequence alignment of the S protein was performed using DNAMAN software. In the alignment of 15 SADS-CoV strains, the amino acids in N2-1: ¹³³SFNSTSHAADAGPTNAFEC¹⁵¹ and N2-8: ²³⁴SYCADGFVNGLQCKLRLFDIPP²⁵⁵ were completely conserved with no mutations observed. N2-2: ¹⁴⁴GPTNAFECLIEGSPYTHRNTGYM¹⁶⁶ and C1-5: ³²⁵GKFNPFTECNGLNRIVDG³⁴² each contained a single amino acid mutation, while C1-4: ³¹²PDQRDFQHLILSNGKFNPFTE³³² contained two amino acid mutations.

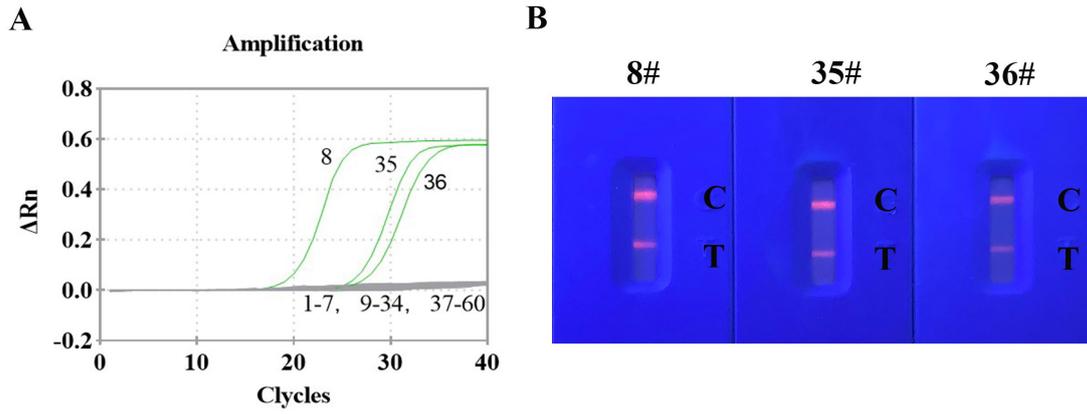


Fig S6. Detection results of clinical samples. (A) Detection results of quantitative fluorescence PCR. (B) Detection results of the fluorescence immunochromatographic strip. Both methods showed that among the 60 pig fecal samples tested, the number of positive results was three, and the detection results were highly consistent.