

Electronic Supplementary Material

A Self-Biomaterialized Novel Adenovirus Vected COVID-19 Vaccine for Boosting Immunization of Mice

Shengxue Luo^{1,2}✉ • Panli Zhang² • Peng Zou² • Cong Wang² • Bochao Liu^{2,3} • Cuiling Wu⁴ • Tingting Li² • Ling Zhang² • Yuming Zhang¹✉ • Chengyao Li²✉

1. Department of Pediatrics, Shenzhen Hospital, Southern Medical University, Shenzhen 518101, China;
2. Department of Transfusion Medicine, School of Laboratory Medicine and Biotechnology, Southern Medical University, Guangzhou 510515, China;
3. Guangzhou Bai Rui Kang (BRK) Biological Science and Technology Limited Company, Guangzhou 510000, China;
4. Department of Pediatrics, Nanfang Hospital, Southern Medical University, Guangzhou 510515, China.

Supporting information to DOI: 10.1007/s12250-021-00434-3

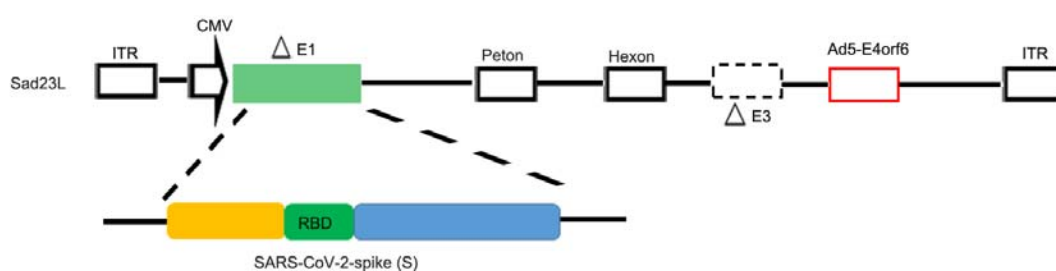


Fig. S1 Flow chart for construction of vaccine Sad23L-nCoV-S. Sad23L vector carries the full-length S gene of SARS-CoV-2 within the deleted E1 region.

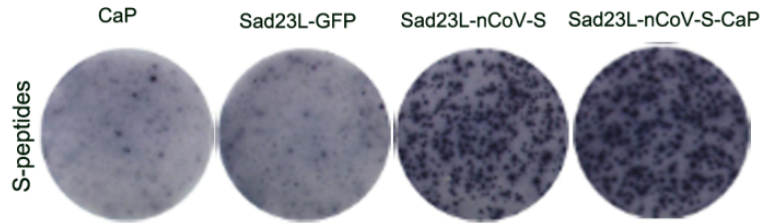


Fig. S2 The ELISpot images of IFN- γ . Mice were primed with CaP, Sad23L-GFP, Sad23L-nCoV-S or Sad23L-nCoV-S-CaP. The blue colored spots indicated positive cellular immune responses by stimulating with S peptides.

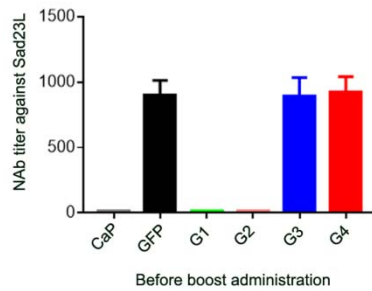


Fig. S3 Measurement of serum NAb titers to Sad23L vectors in Sad23L-GFP vaccinated mice. Mice of GFP, G3 and G4 groups were intramuscularly inoculated by priming immunization with 10^7 PFU Sad23L-GFP, and CaP, G1 and G2 groups were intramuscularly inoculated by priming immunization with PBS. After 4 weeks post-immunization, the NAb titers against Sad23L were detected. CaP: PBS (prime) and CaP (boost); GFP: Sad23L-GFP (prime) and Sad23L-GFP-CaP (boost); G1: PBS (prime) and Sad23L-nCoV-S (boost); G2: PBS (prime) and Sad23L-nCoV-S-CaP (boost); G3: Sad23L-GFP (prime) and Sad23L-nCoV-S (boost); G4: Sad23L-GFP (prime) and Sad23L-nCoV-S-CaP (boost).

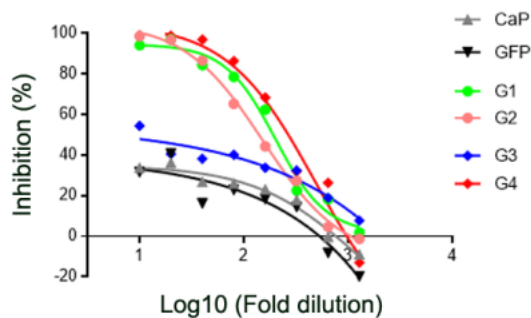


Fig. S4 Inhibition of SARS-CoV-2 pseudovirus. The heat-inactivated sera were acquired from mice of CaP, GFP, G1, G2, G3 and G4.

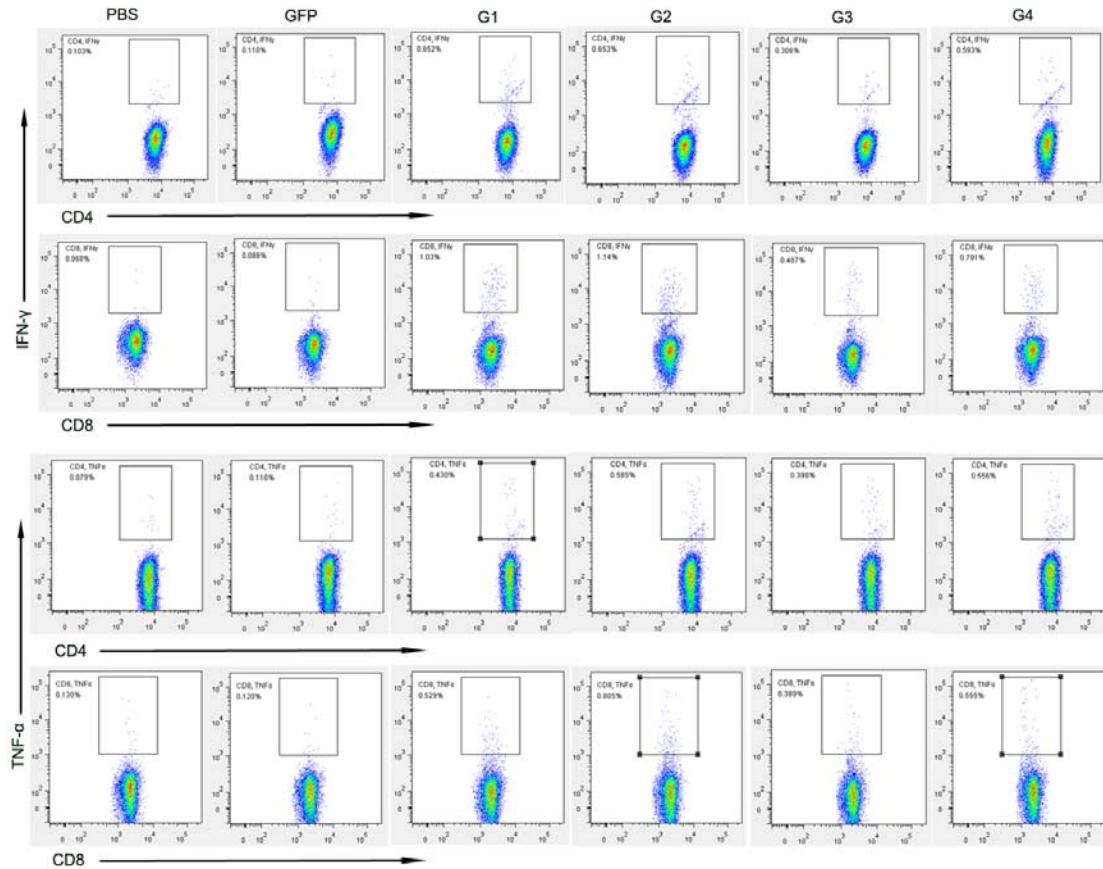


Fig. S5 Frequency of intracellular IFN- γ or TNF- α expressing CD4⁺ or CD8⁺ T cell responses of splenocytes from mice (CaP, GFP, G1, G2, G3 and G4) to S peptides was determined by ICS, respectively.

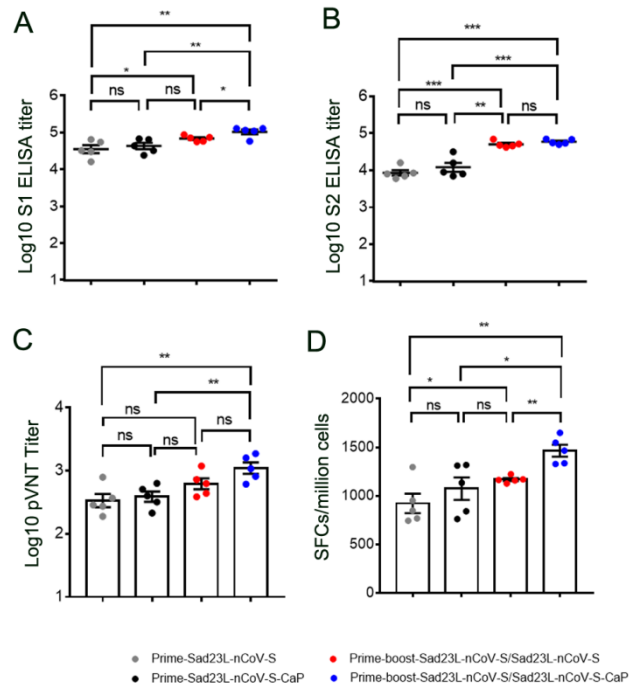


Fig. S6 The comparison of humoral and T cell response in prime-only (Fig. 3) and prime-boost (Fig. 5) groups. Anti-S1 (A) or S2 (B) binding antibodies titer was compared. C NAb against SARS-CoV-2 pseudovirus was showed. D ELISpot detected IFN- γ secreting level to S peptides. Data are shown as mean \pm SEM. *P* values are calculated with two-tailed t test. Statistically significant differences are shown with asterisks (*, *P*<0.05; **, *P*< 0.01; ***, *P*< 0.001 and ns, *P*>0.05 or no significant difference).

Table S1. Immunization regimen in naïve mice.

Group	Number	Vaccine	Titer (PFU)
Sham	5	CaP	100 μ L
	5	Sad23L-GFP	10 ⁷
Vaccination	5	Sad23L-nCoV-S	10 ⁷
	5	Sad23L-nCoV-S-CaP	10 ⁷

Table S2. Immunization regimen in pre-exposed mice.

Group	Number	Prime	Titer (PFU)	Boost	Titer (PFU)
CaP	5	PBS	100 μ L	CaP	100 μ L
GFP	5	Sad23L-GFP	10 ⁷	Sad23L-GFP-CaP	10 ⁷
G1	5	PBS	100 μ L	Sad23L-nCoV-S	10 ⁷
G2	5	PBS	100 μ L	Sad23L-nCoV-S-CaP	10 ⁷
G3	5	Sad23L-GFP	10 ⁷	Sad23L-nCoV-S	10 ⁷
G4	5	Sad23L-GFP	10 ⁷	Sad23L-nCoV-S-CaP	10 ⁷

Table S3. Immunization of mice by prime-boost vaccination regimen.

Group	Number	Prime	Titer (PFU)	Boost	Titer (PFU)
CaP	5	PBS	100 μ L	CaP	100 μ L
GFP	5	Sad23L-GFP	10 ⁷	Sad23L-GFP-CaP	10 ⁷
V1	5	Sad23L-nCoV-S	10 ⁷	Sad23L-nCoV-S	10 ⁷
V2	5	Sad23L-nCoV-S	10 ⁷	Sad23L-nCoV-S-CaP	10 ⁷

Table S4. Peptides derived from amino acid sequences of SARS-CoV-2 S protein used in ELISpot and ICS.

Peptides	Sequence	Peptides	Sequence
1	SSVLHSTQDLFLPF	41	TTRTQLPPAYTNSF
2	FLGVYYHKNNKSWM	42	HTPINLVRDLPQGF
3	FLPFFSNVTWFHAI	43	TTAPAICHDGKAHF
4	QGFSALEPLVDLPI	44	KTPPIKDFGGFNFS
5	KTQSLIVNNTATNV	45	TTDAVRDPQTLEIL
6	REFVFKNIDGYFKI	46	MSFPQSAPHGVVFL
7	AAYYVGYLQPRTEL	47	LTPTWRVYSTGSNV
8	FQFCNDPFLGVYYH	48	LTDEMIAQYTSALL
9	VSSQCVNLTTRTQL	49	YSNNSIAIPTNFTI
10	RVYSSANNCTFEYV	50	TITSGWTFGAGAAL
11	VSGTNGTKRFDNPV	51	LTESNKKFLPFQGF
12	VVIGIVNNTVYDPL	52	SALLAGTITSGWTF
13	QVAVLYQDVNCTEV	53	ILPDPSKPSKRSFI
14	SSVLNDILSRLDKV	54	FTRGVYYDPDKVFRS
15	FIAGLIAIVMVTIM	55	STPCNGVEGFNCYF
16	YQTSNFRVQPTESI	56	CVADYSVLYNSASF
17	AENSVAYSNNNSIAI	57	DLCTNVYADSFVI
18	RSFIEDLLFNKVTL	58	YQPYRVVLSFELL
19	AIPTNFTISVTTEI	59	FPNITNLCPFGEVF
20	SIVRFPNITNLCPF	60	NNLDSKVGGNYNLYL
21	SAPHGVVFLHVITYV	61	YLYRLFRRKSNLKP
22	VAYSNNNSIAIPTNF	62	DSKVGGNYNLYRL
23	FAMQMAYRFNGIGV	63	DYSVLYNSASFSTF
24	LIAIVMVTIMLCCM	64	CYGVSPKLNLDLFC
25	TGIAVEQDKNTQEV	65	YGFQPTNGVGYQPY
26	VFAQVKQIYKTPPI	66	RKRISNCVADYSVL
27	VTYVPAQEKNFTTA	67	TRFASVYAWNRKRI
28	NLTKQLSSNFGAI	68	DFTGCVIAWNSNNL
29	FQPTNGVGYQPYRV	69	SVLYNSASFSTFKC
30	QDVVNQNAQALNTL	70	VSPTKLNLDLCTNV
31	ARSVASQSIIAYTM	71	IAPGQTGKIADYNY
32	CAQKFNGLTVLPPL	72	FKCYGVSPKLNLDL
33	GVTQNVLYENQKLI	73	YSVLYNSASFSTFK
34	ENQKLIANQFNLSAI	74	SGINASVVNIQKEI
35	AEHVNNSYECDIPI	75	IAGLIAIVMVTIML
36	TFGAGAALQIPFAM	76	AKNLNESLIDLQEL
37	AISSVLNDILSRLD	77	QYIKWPWYIWLGF
38	AEIRASANLAATKM	78	VMVTIMLCCMTSCC
39	IRGWIFGTTLDSTQSL	79	LGKYEQYIKWPWYI
40	ITPGTNTSNQVAVL		