Spatially patterned neutralizing icosahedral DNA nanocage
for efficient SARS-CoV-2 blocking

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Supplementary Figure 1. The distances between amino acid residues which Apt (CoV2-6C3 neutralizing aptamer) binds on receptor-binding domain (RBD) of spike trimer at two different conformational states (“closed” and “open”). The distance between amino acid residues of (a) Gln474 or (b) Phe486 on RBD at all closed states. The distance between amino acid residues of (c) Gln474 or (d) Phe486 on RBD at one open state. The molecular docking-based computation above utilized the PDB: 7DDN."}

Supplementary Figure 2. Overhang layout on the edges of the icosahedral DNA origami. The blue dots represent the assembly sites of the aptamer. ID+30toe and ID+3toe were ID with thirty toeholds and three toeholds, respectively. IDNA-30 was ID+30toe functioned with aptamer of molar equivalents of thirty; IDNA-R5 was ID+30toe functioned with neutralizing aptamer of molar equivalents of five; and IDNA-3 was ID+3toe functioned with neutralizing aptamer of three molar equivalents.
Supplementary Figure 3. Dispersion of ID and IDNA-30 after 7 days at 4 ℃. After 7 days, IDNA-30 were still well-composed and well-dispersed, indicating the superior stability of IDNA-30.

Supplementary Figure 4. Representative gels and plots indicating the integrity of IDNA after storing for several days. (a) 1.5 % Agarose gel electrophoresis of IDNA-30, stored at 4 °C for 7, 14, 21, 28, 35 days (corresponding to lanes 3-7) and lane 1: p7560, lane 2: ID. (b) Analysis of the band intensity of IDNA-30 by Image J.

Supplementary Figure 5. Representative gels and plots indicating the integrity of IDNA in a complex matrix. (a) 1.5 % Agarose gel electrophoresis analysis of 5 nM IDNA-30 incubated in 80 % DMEM (10 % FBS) at 37 ℃ for 0, 2, 4, 8, 24 and 48 h (corresponding to lanes 4-9) and lane 1: p7560, lane 2: ID, lane 3: IDNA-30. (b) Analysis of the band intensity of IDNA-30 by Image J. Although IDNA-30 displayed little aggregation or sporadic stripe tailing after incubating with 80 % DMEM (10 % FBS) at 37 ℃, it exhibited slower migration compared with ID. Thus, IDNA-30 showed integrity of aptamer assembly with excellent enzyme resistance.
Supplementary Figure 6. Cryo-EM images of SARS-CoV-2 pseudovirus bound with IDNA-30.

Supplementary Figure 7. TEM images of SARS-CoV-2 pseudovirus bound with IDNA-30.
Supplementary Figure 8. TEM images of SARS-CoV-2 pseudovirus bound with IDNA-30. The particle concentration ratio of SARS-CoV-2 pseudovirus and IDNA-30 were I: 100:1; II: 80:1; III: 50:1; IV: 20:1; V: 10:1, respectively. Scale bar=100 nm.

Supplementary Figure 9. Selected confocal frames at different time points of from top row: virus particles (red) incubated with IDNA-30 (green) and bottom row: virus particles (red) at room temperature. Scale bar=1 μm.
Supplementary Figure 10. Time-lapsed, separate fluorescence channels and merged confocal images of (A) SARS-CoV-2 pseudovirus or (B) SARS-CoV-2 pseudovirus bound with IDNA-30 show viral entry or inhibition on ACE2-transfected HEK293T at room temperature. Cell nuclei (blue), cell membrane (yellow) and virus (red) were stained with Hoechst, Dil and DiD, respectively. The white arrows point to the representative viral particles.
Supplementary Figure 11. Confocal images of 2 nM IDNA-30, 60 nM Apt or 60 nM random sequence (RS) incubated with ACE2-transfected HEK293T cells at 37 °C for 4 h. It is obvious that IDNA-30 (green) did not show cell internalization.

Supplementary Figure 12. Flow cytometric analysis of 2 nM IDNA-30, 60 nM Apt or 60 nM random sequence (RS) incubated with ACE2-transfected HEK293T at 37 °C for 4 h.
Supplementary Figure 13. Representative images of 293T-SARS-COV-2-Spike-Del18-HA-OE(GFP) cells after pretreatment without (A) or with (B) 5 nM IDNA-30 or with (C) 10 nM IDNA-30 before co-culturing with ACE2-transfected HEK293T cells (dyed with Hoechst) for 48 hours. The white arrows point to representative syncytium formation. 293T-SARS-COV-2-Spike-Del18-HA-OE-GFP and ACE2-transfected HEK283T cells were defined as effector cells and target cells, respectively. Three fields were randomly selected in each well. The white arrows point to the representative syncytium formation.
Supplementary Figure 14. Scheme of inhibition experiment of SARS-CoV-2 pseudovirus. Utilizing a lentivirus packaging system, SARS-CoV-2 pseudovirus was packaged with spike trimers protein of SARS-CoV-2 as a surface capsid glycoprotein, and the RNA genome comprising the gene of CMV-promoter, GFP, IRES and luciferase.
Supplementary Figure 15. Enhanced cck-8 assays of cell viability. Different concentrations DNA nanostructures (ID or IDNA-30) were incubated with ACE2-transfected HEK293T cells for 48 h.

Supplementary Figure 16. Flow cytometry analysis of 0.5 nM IDNA-30, Apt and random sequence (RS) incubated with white blood cells at 37 °C/30 min.
Supplementary Figure 17. Early immunogenic response of 5 nM IDNA-30 by measuring the IFN-γ, TNF-α, IL-10, IFN-α levels in mouse plasma samples. Plasma proinflammatory cytokine concentrations of C57BL/6J mice injected with IDNA-30 or PBS showing negligible change.
Supplementary Figure 18. Histological analysis of mice injected with IDNA-30. Representative H&E staining analysis of organs at the end of the experiment.
**Supplementary Table 1. DNA sequences of DNA origami.**

The sequences are marked with colors; replaceable staples with **bold font** and **yellow background**, bases with **green background** are spacers, the 18-nt nucleotides with **red** are toeholds of DNA origami.

| Staple Code | Length (bp) | DNA sequences (5’-3’) |
|-------------|-------------|----------------------|
| 1           | 49          | AGACATTTTTGTCAAATCACCAGTACCCCGGTGTTTTTTTGATAATCAGAAA |
| 2           | 49          | AATTCTTTTTGCGCTCTGGCCTAGTATCGGGCTTTTTTCAGGAAGATTCGCT |
| 3           | 22          | TTTTTTTTTTTAAAAATCGCA |
| 4           | 43          | AATTATTTTTACCGTTGTAGCGATAGGGTGAAAATTTTTTGTTTG |
| 5           | 49          | TAGGGTTTTTCGCTGGCAAGTGGAACGGTACGTTTTTCCAGAATCTGCA |
| 6           | 33          | TTCCACCCCGATTTAATTTTTGAGCTTGACGAG |
| 7           | 49          | ATTTGCTTTTTTCTATATAGCATATCTATGATACATTGTTTTTCCAGAATCTGCA |
| 8           | 49          | CCAATTTTTTACTCGGAATCTCATCGGCTTTTTTATTTTTATTCGAGCTTCA |
| 9           | 22          | TTCTATTTTTCTATAGTACAG |
| 10          | 43          | AGGCTTTTTTTTGCAAAGACCTCATATATTTTTTTTTTAAT |
| 11          | 49          | AAGTTTTTTTTGAGTAAACTATTTAGAAATTACTATTTTTTTGCTTTAGTAAGA |
| 12          | 33          | GCAATGCGACGTTTTTTTTTCCAGTCAGAC |
| 13          | 49          | TAGACCTTTTTTTAAACACGCGGCCTTGAGTTTTTTGACCTCATATGATA |
| 14          | 49          | ATAAATTTTTACAGAGGTAGCTAATATTCTATTTTTTTTTAGGAGACTA |
| 15          | 22          | GGCTTTTTTTGATTCGCTGCA |
| 16          | 43          | ATGCGTTTTTTCCAATGATATTTATCCGCTCATTTTTCAATCC |
| 17          | 49          | TCAAAATTTTTCTATCGGACTACGACTATCTATTTTTTTTTTCCATCTGACCT |
| 18          | 33          | CACACGCGTATTGGGCTTTTTTGCAGGCTGGT |
| 19          | 49          | TCACCTTTTTGTACTCAGGAGCGACTCTCATATTTTTGTTAGCGTAAAG |
| 20          | 22          | GGGTTTTTTTTGCTACGATAC |
| 21          | 49          | TTTGCTTTTTTTAAAAACATTTTGACTAGTTTTTTTTCGCGCGGCAAA |
| 22          | 33          | CATGAAGATTTTTCTATTTTTAAACGGGTAA |
| 23          | 43          | AATAAAATTTTTATCCTCATATAAACATTTTATTTTTTTATGAAA |
| 24          | 49          | ACTAATTTTTAAACACTCATCTAGGAGACGTGTATTATGAAACCGGTGTA |
| 25          | 49          | CGCCTTTTTGCCAGCATTGACATCAAATTTTTATTTTTTCATTTGTGAATTTA |
| 26          | 22          | TTGAATTTTTTCGCTAATATCAG |
| 27          | 49          | CATATTTTTTTTCTTGATAATTATCCGCTACCTTTTTTTTACATCGGAGAA |
| 28          | 33          | GCCGAAACGATACAAATTTTTTTATAATATG |
| 29          | 43          | ATTTTTTTTTCACAACCAGTAAACATAAAAACATTTTTTAGGAA |
| 30          | 49          | TCTTTTTTTTGAACAACTACGCTTTTTTCAATTTTTTTTTTATTAGTAA |
| 31          | 49          | GGCGTTTTTTTTTAGCGAACCCTTAATTGAGATTTTTATCGCCCATATT |
| 32          | 22          | TAGGATTTTTATCCGACGCG |
| 33          | 49          | TATAATTTTTAGTACCGCAATATCCTATTTTTTTTCAAGAACCGG |
| 34          | 43          | AAAAGGTATTTTTCACGCAAGAGCCATTTTGGGATTTTTATTAGA |
35 33 GCCAGTTAGCGTTTGTCTTTTTCATCTTTTCTATA
36 49 CCCCTCTTGTAGAAACGCGACAAACTGGCAGATATTTTTTAAGACTCCTT
37 32 ACTCACATTAATTGGGCGATGGCCCGGTTAATA
38 24 AACCGCTCATATCATCGTAAACT
39 31 GATGAACCGGTATCTGTGGGAGAGGAGGCCG
40 24 CAGGCTGCGCAATATCATCGCAGT
41 31 TCGTAACCGGTGTTGCGCTCCAAATAT
42 31 ATCGGCAAAATTTATCAAAATATCATCAACGCA
43 24 GAACCGCCTATCATATCGTAAAAC
44 31 GGAGCTAAAAAGATATGCAACTACGGGGCG
45 24 CAACATGTTTTTATTAGCAGCATAAAT
46 23 AGGTGCGGTAATCCTTATATAAT
47 31 TTGCAATTCTTATTTGAAATATCTCCACTTTA
48 24 ATTTTTTCAGTTAAAGGAAAGC
49 31 ATCAAAAAAGATTCTAATATCGGATGATAGCT
50 32 CAGAGCATAAAGTCTACAAAGGCTAGCATCAA
51 24 TTHTTGTAGGATTTTGCTGAAGAGTGA
52 31 AGCCTTTATTTTTTGTGCTGAAATTAGCGAG
53 24 TCGCCTGATAAAATCAACATGAATGCA
54 31 ATACACACATTTATGCGACAGACTATTTAA
55 24 ATCAAAAAAGATATGCAACTACGGGGCG
56 31 ATTCTATATATATGCAAGAGTGA
57 31 GGAGCTAAAAAGATATGCAACTACGGGGCG
58 24 CAACATGTTTTTATTAGCAGCATAAAT
59 23 ATCAAAAAAGATATGCAACTACGGGGCG
60 31 TTTTTGAGAGATTGCGAACGTAT
61 24 TCGCAATATCAGGCTCCAGTAA
62 31 TTGCAATTCTTATTTGAAATATCTCCACTTTA
63 24 ATCAAAAAAGATATGCAACTACGGGGCG
64 31 GACGGTGCAATCTCAAAAATCAGGTAGAATAC
65 24 GCAGCAAGCGGTGTTGCCAGAAGCG
66 23 ATCAAAAAAGATATGCAACTACGGGGCG
67 31 TTTTTGAGAGATTGCGAACGTAT
68 32 TTTTTGAGAGATTGCGAACGTAT
69 24 GGCCTGTGTTTTCGAGGTTGTA
70 31 GACGGTGCAATCTCAAAAATCAGGTAGAATAC
71 24 GGCCTGTGTTTTCGAGGTTGTA
72 23 ATCAAAAAAGATATGCAACTACGGGGCG
73 31 TTTTTGAGAGATTGCGAACGTAT
74 24 GGCCTGTGTTTTCGAGGTTGTA
75 31 GACGGTGCAATCTCAAAAATCAGGTAGAATAC
|   |   |   |
|---|---|---|
|   | 24 | ATGACCATAAATTTTTGAGGACTA |
| 77 | 31 | CAGAACGAGTATGAAGCCCTTTTTACCCAGAG |
| 78 | 32 | AGCTATCTTACCTCCTGAGCAAAAAGAGGTAA |
| 79 | 24 | TCATTTCATTATGTACCTAAGGTC |
| 80 | 31 | TAGATTTTCTGTCTAAAGCCATAATTATCAT |
| 81 | 24 | TAATAAAACGAATGTAAATTGGGC |
| 82 | 24 | AAAAAATCTAAGITCTATTTTGAATT |
| 83 | 23 | ATTTAACAATTGAAAATAGCAG |
| 84 | 31 | ACGTCAAAAAATTCCGGAATCATAAAAAGTTGCT |
| 85 | 24 | TAAGAATAAACATATCAGCACAGACA |
| 86 | 31 | ATGCAAAATCCCATCATCACCCTTCTCTTCTAGAA |
| 87 | 31 | AATTTCATCATAAAGGCGACATAACCGGA |
| 88 | 32 | GCGCCAAAGACATCCGGAATCATAAAGTTGCT |
| 89 | 24 | CCAACCTCATTTTAGAAACCCAATC |
| 90 | 31 | TACGAGCATTTCTCTTTTCTGCTAAAGAGAA |
| 91 | 24 | TGGAAATACCTATGTAATAAAGCCA |
| 92 | 23 | GCACCTTCTTTGCAGAATTTATC |
| 93 | 24 | GCTTTTGATGATTTCGCAAATTTTTC |
| 94 | 31 | AAACCGAGGAATAACAGAGATGTACGGCCAGCA |
| 95 | 24 | CCAGTTCAAAAAATAACGCAATAATA |
| 96 | 31 | TATTCATAAATTTAACAGCACATAACATAT |
| 97 | 32 | TATTTAAAAACAGGAAGCTCAAAGGCGAAA |
| 98 | 32 | GCGACGTGTGTATGTACCCGAGAGGCTAGCTA |
| 99 | 32 | TTCAAAAGCGCTTTTACGCGCATTAGGATT |
| 100 | 32 | TCGTAGTTTGCAATCAGATCAAACCCGCGGCTAG |
| 101 | 32 | CCAATAGGTTTTAAACCTATAGTGATAGCTA |
| 102 | 32 | AAGAAAGCCGGAAGCTATATATAATGCTGTAGCT |
| 103 | 32 | AAAATGTATCTAATAgCTAAAAAGCCT |
| 104 | 32 | TGGCCGAAACGACGCGGATGTCTTTCTCCAAAGT |
| 105 | 32 | ACCGAACGCATAAAATTAATGAGTAAACAGGG |
| 106 | 32 | ATCATTGCTACGAGTGCGGCTTGGAGGATTTT |
| 107 | 32 | TAAAGAGGTGTTAATAATATGCTAGCTAATGCA |
| 108 | 32 | TAAAATGAAATTGTTGCTATTAGGCACCAGTAC |
| 109 | 32 | GTCAGGATCAGACCAGGGAATTGCGAATAAT |
| 110 | 32 | AAACGAAATGCGCCATAAAGCTAAGGAACAGA |
| 111 | 32 | ACGATAAAATCTAACAGAGGAGTTTGTATCA |
| 112 | 32 | ACCAGAAGGATTTTGAAGAAAAATCTACGT |
| 113 | 32 | TACATTTGAGATTGTTAATCTCTAGCTAATA |
| 114 | 32 | TATTAATACCTTGCGACAGCAGCAGAAATG |
| 115 | 32 | TTTGAATAAGAATACTTTTATCAAAAATCATAG |
| 116 | 32 | TTTTCGAGCAACATGTACAGGAAAAAAGCCTCA |
| 117 | 32 | GCAAGCCGAAGTACCGGCCACCCTCAGAGCCA |
| 118 | 32 | CAAGAGAAACCATCGCCTTGCAGGGAGTTAA |
| 119 | 32 | ATTGGGCTGCGCATAGAAGAACCAGTATTCA |
| 120 | 32 | CCTGGAACGGATTGCCCGAGAGGCCGAATTAT |
| 121 | 32 | GCCTTAATCTGACCTTAAATAAGGCGTTAAA |
| 122 | 32 | AAGTATAGAAACTGCCGAAACGTCACAGCAC |
| 123 | 32 | AACCCTCTCCACCCGAAGGTAAAGCAGTCATAG |
| 124 | 32 | CGCCACCACCACAAAGACAATGAAATAGCAAT |
| 125 | 32 | CATACATAGTATGTTTACTCCAGAGCCATTTTG |
| 126 | 32 | TATCCCGGTCAAGCAAATTCATATGGTTTACCA |
| 127 | 56 | CCAAAATCGGACTCCAAGATTTGTACACCTATTAACAAGGAGGTTTGGAATTGGGGTCG |
| 128 | 56 | GAGTCTGGAATTAATGTATTTCATAGAGCATATATACAATATTAGAATC |
| 129 | 56 | TCTTCGCTCCAGGCAACGCGCACCACCTTCTGGTTTGAACCGCTGAGTAGCTGGCG |
| 130 | 56 | GTCACGGTTAGGAGGTGACCTCATCCTCAACAAACGAGCAGTCTACGTAGTGCGGCA |
| 131 | 56 | TGGGGAAAATGCGGGTTCTCAGCTCAAGTAAAGGGCGGTTTAACATACGGCAGCTG |
| 132 | 56 | AGTTTCATATTGCTAGGGCGGAAGATGGCCTTTTGACCATAGAGGTCAACAGTTGA |
| 133 | 56 | ACCCTGTATAGCAAAAAATCATACAGGCAAGGTTTAGAACAGTTTGCCGCGGAGA |
| 134 | 56 | TGAATTGTGCGAGGTCAGTCACATCAGAAAGGTGTTTTACGCTTCTGACAAATGCAATG |
| 135 | 56 | GATCCCCGGGTGGTGCAATTTCCTCCGAAACTCTTGAATTGCCCTAAAAGCTCAGAAT |
| 136 | 56 | ATCCTGTTCACACCAGACGGCGCAACGCTTGACTGCGCAATAATCTGCTCCGAAA |
| 137 | 56 | GAAAAATATAAACCAACGGTGAAGGTCCAGACGCTTCTAAGGTTAAAATCCTAATT |
| 138 | 56 | GCTTTCATCCGCGCTTCTTCTCAGCAGTACTTAACTGACAGTACGCGGGAAGAGCAGCG |
| 139 | 56 | AAGGAGCCACAATCAAAGCAACGCTGAGAATAACAGCTTCAACAGTTTATCGGTTT |
| 140 | 56 | ATAGTCAGCTTTAACCAGAAGGCCAGATTCCCTTACACTGCTATACAAAGGCAGGAG |
| 141 | 56 | GTTACTTAAAGGTACACCTCCTTCTTACATTAAGCAGCTTGGAGCCCCAGAGGCCGA |
| 142 | 56 | AAGATTCAGACCGTTGGGAACCTGGCTCATTATACAGCAGCCTAAAATCATTAGG |

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|   |   |   |
|---|---|---|
| 143 | 56 | TAAAACAGTGTATTGGAAGCCGTCATCAATATATACGTAAGAAGATGATG AAATTCGG |
| 144 | 56 | ACCCTCAACACGCTGATTCTGTAACCGCCTGCCAAGGTATGCGTCACTG GGTGCACTG |
| 145 | 56 | TTATATAAATAGTGAAGTGGCACAACGCTGAGGAGAAAACGATAGCTT AAATGCTG |
| 146 | 56 | ATATTATCTACATGCAAATTTTAAAGGAACAATATTCCTGGCCAAGCTGGTAAG ATTCACC |
| 147 | 56 | ATGTACCGTCAAGAACCACACTCATCCTCAGAACTCCACAGAGTTTAGTA AGTTCGTA |
| 148 | 56 | AGCATCGGCCTGAGGGCCACGATAAACCCATATTCATGGAAGATTTAA TAGCAACG |
| 149 | 56 | AAGGCTTTGATCTTTGACCGTGCTCCCTCATGTTTAATTCAGGAGGTA GAAACAC |
| 150 | 56 | CATCAAGACAAAAATCGCATTGATGCTTTGGAATATAATGGAATATTAGCCG TTAATAC |
| 151 | 56 | GTTUGTATCCGTGTGAAAAATTTAATGTTGACAGTAGGCTCAGACTG TTAATAC |
| 152 | 56 | GGGGACCTAGGGCCGATCAGGAGGACCATTACCCCCCTTACAAAAATCA AGACTGTA |
| 153 | 56 | CCGGGGTAGCCTCAGGCTAGGAGCCGAGTCCTCTATTTGCAAGCCAGGAA AGTACAG |
| 154 | 56 | TAGCCGAAAGGCAAGAAAATTTAGTATTAGGCTAGCACCACAAAAGCCTCAGA GCGAGAAGG |
| 155 | 56 | AAAATAAGGAGCGTCTTTGCAAACGTCTTTACCAAGAGAATCAATTTTA TTTGTATA |
| 156 | 56 | GGGAGGTATAGAAAAATCGATATTTTTTGTCACTGGACACCCACGGG ACGGAAAT |
| 157 | 32 | AGCCCCAATTGGAACACTACGTAAGGAAAGCATCA |
| 158 | 32 | ATACATCTGAAAAGGTGCATGCTATTGCTTCCCTGA |
| 159 | 32 | ACTCCAGCGGTGAGAAGCGATCGGTGCGGGCC |
| 160 | 32 | CCGACAGTGGGCAGAGCCGATACGGTGTCAGGAG |
| 161 | 32 | TTAAATTTAACGCTATCCTGCTTTCCAG |
| 162 | 32 | GAAAGCCCGAAAGGAGAAGTAGCAGGGTGCTCAGGA |
| 163 | 32 | GCAATTAATAGAATCTGGTACCCAAACATCATG |
| 164 | 32 | GTTGTTAAACGTTTATTATAAGTGTTCTTTAGTG |
| 165 | 32 | CGCATTTTAACCCACACTGCCCTGAAAGACGGAG |
| 166 | 32 | TTTCTTTTTCTCAAGGAGGCCCACGGAAAGG |
| 167 | 32 | GAAGTGTTTCTGTCCAGATAAGTGCCTGAAACAA |
| 168 | 32 | ATCTAAAGTTTTCTCTGAGGACAGGTATAAAGCT |
| 169 | 32 | AAGCGAAGTACGAGATAAAAAAGGCTCAGAA |
| 170 | 32 | ATACGTAAGAGGCAAACCTTTAACCCTGACTATT |
GCAACACTAACCACCAACCACGACCTGCTCCAT
CTTAATGCGAGCGGAACACATTATCAGGTAGA
ACAACTAAAGGATTACAAAATTATTTCGACG
GAGTGAATAATTTTCCCAACCTCTAACATCAA
GAAAAGCGTGCTATTATCCCGCTTAGGTGTTG
AACACGCCCCAGTAAATCAGTGATACAGTAGG
ATTAAACCTTTTTATTAGCCCAATAGGAACC
TGCAACACAGGATTGCTCAGCAGCGGAAAGAC
CAGACCGATGTATTCTGCTCATTCAGTGAAT
ACAATACACAAAAGTCAAGAGATGTGAAACAAA
TTTCATCTTCAGAGATTTTACTAGAAAAAGCCT
AGCGGATCCCGGAATGTAGCGACAGAATAAGAA
ATCAAAATCCCTCAGATGTAATAAGATTAA
AGAGATAAGAACACAGAAGAAAAGTAAAGCAGA
ATTACGCAAAAGGTGGCTTTATATTCCAATCC
CCCCATAGATTTCTAGTCAACCCGATGAGGGA
GAACCCTAAGAACGTAAGTTTTTCAAGAGTCTAA
AGCATGTCAGCTGATAAGCAAACAGATATTCATTTGGGGC
GATTAAGTGCGGAAAATTACGCAATGTGAGTGAAGAT
TTGAGGGGTTAAATGTGGTGTAAGCGAGCTTTAAGCAC
CGGGGAGATGTAAAGCCCTGTCGTAGCCGGAATTTTTTA
GATTTAGTAGAGCTTATCCCATATATTTTTGCAGGAGGAGG
TAAAAATTCAAAGAATATACTTTTCAGAGGGGTAATAG
CTATCTACAGGAAGAAACACCTACTCTAGTTAATCCCTT
TTCCCTGAGGCACTCCTCTGCTACCGGACATCGCCATTAAAAAT
CAAAGAAAATTGCGCCTTGTGATTGTTGATTAGTAATAAC
AATAATCGGACGCAAAATATCCCGTAGAATTCTCCACGAG
AAAGGGGATACCACACCGTTAGAACACACGCTCGGACGTAG
ATTCTAAAGAAAAGATTCTTAATT TGCAAGGATACCCAC
CCAAGACTCAAATGAAGCAAAGTATTCATTCACCCATACT
GAACGTACCGCCGAAACCGCCGAAACCACGATTACCAGACG
GATACATACCCATGCAGCTGAGCAGCGAAACAAAGAAACC
GATGAAATATGCAATATTTAAAGCAATTTCAATAGTAA
AATCAGAAACAGTGCTCAATATCTCAAAATGCTG
GGAACATTACCCGCGACCTTGTCAGCATCAGACAGAGCA
GGCGAATTACCGGTAAAAATTACGCGGAAAGACCCAC
AACACTCTCTCCCTTAGAACGAGATGAGGGATAC
TTCCCACAAGAGTCATATCTGCAGTGAGC
AAGACATTATTTCTCAGGAAGGAGAGAGCTGAGACTCCT
TTGAGATGCAAGAGTACCCCTAGCAGTGGAGCGGACAGTCAGC
ACCTTTTCCAAAGTTAAAAACAAAAAGAGAATATTAAACTGAAC
ACGCTCAAAATACCGATCATATGCTCGGGAAGGTTTTGA
| Sequence | Length | Description |
|----------|--------|-------------|
| GGTCATAGCATTAGCATTAGCGTCCCCAGGTAGCGTTGATAT | 40 | |
| CCCCTGCCCTGAGATTTAGTGCCCTTGGGAAAAGCACCCGG | 40 | |
| ACCGGAATAATAATAAGGAAGTTACCACCACCCTCAGAGC | 40 | |
| CTTTTACACGCTAACGAAACGATTTCCTGAAATAGAAAATA | 40 | |
| ACCGTCACCACAATCAAATATTGAATAAGTTAGAAGGCT | 40 | |
| GGAACCCCTAAGAAGTAAAGTTTTTCAAGAGTCTAAGCAAAG | 58 | GAGAGTTAGGAAATGT |
| AGCATGTCAGCTGATAAGCGAAAGCAGATATTCCATTGGGGC | 58 | GAGAGTTAGGAAATGT |
| GATTAAGTGCCCGAAAATTACGCAATGTGTTAGGAAAGGAAG | 58 | GAGAGTTAGGAAATGT |
| TTAGGGGTTAATAGTGTTTAGACAGCTTTTAAAGCAAC | 58 | GAGAGTTAGGAAATGT |
| GATTAAATGAGCTAGTCTATCCATATATTGGGACGAGAAAGG | 58 | GAGAGTTAGGAAATGT |
| TAAAAATCAAAGAATAATACCTTTTCAGAGGGTAAATTAGT | 58 | GAGAGTTAGGAAATGT |
| CTAATCTACAGGAGAACAACCTTACTCGTATTAAATCCTT | 58 | GAGAGTTAGGAAATGT |
| TTCCTGTGACCTCCTGGTACCGACATCGCCATTAAAAAT | 58 | GAGAGTTAGGAAATGT |
| CAAAGAAATTGCCCTTGATGGTGTTTGATTAGTAATAAC | 58 | GAGAGTTAGGAAATGT |
| AATAATCGGACGACAAATATCCCAGTAATTCTCCACCGAG | 58 | GAGAGTTAGGAAATGT |
| AAAGGGATACCACCCGTTAGAACACGCTGCGACGTTAG | 58 | GAGAGTTAGGAAATGT |
| ATTTCTTAAGAAGGATTATTTATTACGAGATCCAAACAG | 58 | GAGAGTTAGGAAATGT |
| CCGAAGACTCACAATGAAGCAAGATTATTCTACCAACCTA | 58 | GAGAGTTAGGAAATGT |
| GAACTGACCGACGACGACGACGACGACGACGATACCAGACG | 58 | GAGAGTTAGGAAATGT |
| GATACATACCGTACGTCAGTGGAGCGGAACAAAGGAAACCCCATCGT | 58 | GAGAGTTAGGAAATGT |
| GATGAAATATCCTGATAGAATAAAGGCAATATTCAATAGATAA | 58 | GAGAGTTAGGAAATGT |
| AATTGAGGAACGACGTGCATATATCTATTAACAACATCGTCG | 58 | GAGAGTTAGGAAATGT |
|    |    | AAGAACGCAAGAGTCACTATATGTAGATTAAGGACAATAT | TTTTAGAGAGTAGGAATGT |
|----|----|------------------------------------------|---------------------|
| 235| 58 | GGGACATTACGCCAGCATTTGGCATATCCAGACAGAGGCA | TTTTAGGAATGT |
| 236| 58 | TGTAGCATCGCCACCTAACACTGCCGCAACCGGACAAACAA | TTTTAGGAATGT |
| 237| 58 | AAGACTTTTTATTCGTAACGAGGGAGGAGCTGAGACTCCT | TTTTAGGAATGT |
| 238| 58 | TTAGGAATGT |
| 239| 58 | TTAGGAATGT |
| 240| 58 | ACGCTCAAATACCGATCATATGCCTGGAGGTTTTGAA | TTTTAGGAATGT |
| 241| 58 | GGTACATAGCATTTAGCGTCCAGTGGACAGTTGATAAT | TTTTAGGAATGT |
| 242| 58 | CCCCCTGCCTGAATTTAGTGCCTTGTGGAAAGCACCACCGG | TTTTAGGAATGT |
| 243| 58 | ACGGAATAATAATAAGCAAAGTTACCACCACCTCAGAGC | TTTTAGGAATGT |
| 244| 58 | CCTTTACACGCTAACGAACGATTTTCCTGAATGAATAAT | TTTTAGGAATGT |
| 245| 58 | ACCGTACACCACAATCAAATATTGAATAAGTGAAGGCT | TTTTAGGAATGT |
Supplementary Table 2. DNA sequences used in this work.

| Name   | Length (bp) | DNA sequences (5'-3')            |
|--------|-------------|----------------------------------|
| Apt    | 46          | CGCAGCACCACAAAGAACGGACTGCTTAGGATTGCGATAGG TTCGG |
| Apt-T  | 66          | CGCAGCACCACAAAGAACGGACTGCTTAGGATTGCGATAGG TTCGG ACATTCTAACACTCTAAA |

Supplementary Table 3. DNA strands corresponding to the DNA nanostructures.

| Name       | DNA strands involved                  |
|------------|---------------------------------------|
| ID         | Staple Code.1-216                     |
| ID+3toe    | Staple Code.1-186, 189, 190,191, 193-216, 217, 218 and 222 |
| IDA-3      | Staple Code.1-186, 189, 190,191, 193-216, 217, 218 and 222; Apt-T |
| ID+30toe   | Staple Code.1-186 and 217-246         |
| IDNA-30    | Staple Code.1-186 and 217-246; Apt-T  |

Supplementary Table 4. Molecular mass conversion

| Name   | Calculated formula | Relative molecular mass (g/mol) |
|--------|--------------------|---------------------------------|
| ID     | 7560*660           | 4989600                         |
| ID+3toe| 7560*660+3*18*330  | 5007420                         |
| ID+30toe| 7560*660+30*18*330 | 5167800                         |
| IDNA-30| 7560*660+30*18*330+30*66*330 | 5821200 |
**Supplementary Table 5.** Comparison of the previous reported neutralizing antibodies to IDNA-30.

| Name     | Wild-typed pseudovirus neutralization IC50 (μg/mL) | Wild-typed pseudovirus inhibition efficiency | Wild-typed authentic virus neutralization IC50 (μg/mL) | Authentic virus inhibition efficiency | Manufacture method | References |
|----------|-------------------------------------------------|---------------------------------------------|-----------------------------------------------------|--------------------------------------|-------------------|------------|
| IDNA-30  | 0.0048 (0.82 aM)                                 | ~92%                                        | NA                                                  | 85.5% (D614G)                       | DNA molecular self-assembly | This work  |
| VHH E    | 48 nM                                           | NA                                          | NA                                                  | NA                                  | Single-domain antibodies (VHHs) isolated from a llama immunized with coronavirus spikes | *Science* 2021, 371, eabe6230 |
| VHH EV   | 0.7 nM                                          | NA                                          | NA                                                  | NA                                  | Single-domain antibodies (VHHs) isolated from a llama immunized with prefusion stabilized coronavirus spikes, and subsequent mass expressed in *Pichia pastoris*. VHH-72-Fc included two SARS VHH-72 molecules connected by a (GGGGS)_3 linker and a genetic fusion of SARS VHH-72 to the Fc domain of human IgG1. | *Cell* 2020, 181, 1004–1015 |
| VHH VE   | 1.32 nM                                         | NA                                          | NA                                                  | NA                                  | Single-domain antibodies (VHHs) isolated from a llama immunized with prefusion stabilized coronavirus spikes, and subsequent mass expressed in *Pichia pastoris*. VHH-72-Fc included two SARS VHH-72 molecules connected by a (GGGGS)_3 linker and a genetic fusion of SARS VHH-72 to the Fc domain of human IgG1. | *Cell* 2020, 183, 1–11 |
| n3088    | 3.3                                             | 100%                                        | 2.6                                                 | ~ 80%                               | Fully human single-domain antibodies expressed in *Escherichia coli* | *Cell Host & Microbe* 2020, 27, 891–898 |
| n3130    | 3.7                                             | 100%                                        | 4.0                                                 | ~ 60%                               | Fully human single-domain antibodies expressed in *Escherichia coli* | *Cell Host & Microbe* 2020, 27, 891–898 |
| n3088+n3113 | 0.51                                         | 100%                                        | NA                                                  | NA                                  | Fully human single-domain antibodies expressed in *Escherichia coli* | *Cell Host & Microbe* 2020, 27, 891–898 |
| n3130+n3113 | 0.70                                         | 100%                                        | NA                                                  | NA                                  | Fully human single-domain antibodies expressed in *Escherichia coli* | *Cell Host & Microbe* 2020, 27, 891–898 |
| BD-368-2 | 0.0012                                          | 100%                                        | 0.015                                               | 100%                                | High-throughput single-cell RNA and VDJ sequencing of antigen-binding B cells before candidate genes were transfected into HEK293 cells. | *Cell* 2020, 182, 73–84 |
| BD-629   | 0.006                                           | 100%                                        | NA                                                  | NA                                  | High-throughput single-cell RNA and VDJ sequencing of antigen-binding B cells before candidate genes were transfected into HEK293 cells. | *Cell* 2020, 183, 1–11 |
|       |       |       |       |       |
|-------|-------|-------|-------|-------|
| 1-20  | 0.127 | 100%  | 0.008 | 100%  |
| 2-38  | 0.232 | ~95%  | 0.208 | 100%  |
| 2-4   | 0.394 | ~95%  | 0.057 | 100%  |
| 2-30  | 0.512 | ~95%  | 0.050 | 100%  |
| CV1   | 15    | 70%   | NA    | NA    |
| CV30  | 0.03  | 100%  | NA    | NA    |
| P2C-1F11 | 0.03 | 100%  | 0.03  | 100%  |
| P2C-2F6 | 0.05 | ~95%  | 0.41  | ~100% |
| P2C-1A3 | 0.62 | ~95%  | 0.28  | ~95%  |

|       |       |       |       |       |
|-------|-------|-------|-------|-------|
| CA1   |       |       |       |       |
| Huh7  | 1.276 | 100%  |       |       |
| Calu-3| 0.527 | 100%  |       |       |
| HEK2  | 4.659 | 100%  |       |       |
| 93T   |       |       |       |       |
| Vero E6 |     |       | 30h  | 0.382 | ~80%  |
|       |       |       | 72h  | 4.981 | 100%  |
| CB6   |       |       |       |       |
| Huh7  | 0.036 | 100%  |       |       |
| Calu-3| 0.023 | 100%  |       |       |
| HEK2  | 0.041 | ~100% |       |       |
| 93T   |       |       |       |       |
|       |       |       | 30h  | 0.036 | ~100% |
|       |       |       | 72h  | 0.835 | 100%  |
| S309  | 0.150 | 100%  | 0.079 | 90%   |
| CC6.29| 0.002 | NA    | NA    | NA    |
| CC6.30| 0.001 | NA    | NA    | NA    |
| COV2-2196 | 0.0007 | 100% | 0.015 | 100% |
| COV2-2130 | 0.0016 | 100% | 0.107 | 100% |

Single antigen-specific memory B cell antibody cloning

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