Electronic Supplementary Material (ESI) for Nanoscale.


## Supporting Information

# A switchable DNA origami nanochannel for regulating molecular transport at nanometer scale

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## Experimental Section

## METHODS

***Preparation of DNA origami nanochannel.*** Detailed structures of DNA origami nanochannel devices used in this study are shown in Supplementary Fig. S1-S5, and the sequences of the staple strands are shown in supplementary material. All short oligo-DNA strands were purchased from Invitrogen. DNA origami assembly was done by mixing scaffold and staples to a final concentration of 6.25 nM and 15.625 nM, respectively, in a 1×TAE-$Mg^{2+}$ buffer (20 mM Tris, pH 8, 2 mM EDTA, 12.5 mM $MgCl_2$). This mixture was cooled from 90 to 25 °C at a rate of − 1.0 °C /min using a PCR thermal cycler.

The assembled structures were purified from the excess staple strands by centrifugation with Millipore's 100kD molecule-cutoff Centricon spin-filter in three cycles at a speed of 3000 g for 10 min at 4 °C in the same 1×TAE-$Mg^{2+}$ buffer. The assembled origami structures were then collected at the end of the third cycle of filtration.

***Preparation of DNA-enzyme conjugates.*** DNA-enzyme conjugates were prepared using sulfo-EMCS as a bi-functional crosslinker. In a typical synthesis, glucose oxidase (GOx) or horseradish peroxidase (HRP) (0.5 mL, 12.5 mM in 20mM phosphate buffer pH 7.4 containing 0.15M NaCl) reacted with 100-fold excess of sulfo-EMCS at 25 °C for 6h. The excess of sulfo-EMCS was removed with Millipore's 30kD molecule-cutoff Centricon spin-filter in the same phosphate buffer. The product was then mixed with 5-fold excess of thiol-modified DNA at 25 °C

overnight. The final DNA-enzyme conjugates were purified with 30kD Centricon spin-filter to delete the excess DNA.

***Construction of enzyme cascade in DNA origami nanochannel.*** For preparing enzyme cascade in DNA nanochannel, the DNA-enzyme conjugates were assembled with DNA origami nanostructures (with DNA-enzyme conjugates' complementary strands) in stoichiometric ratio at 37 °C for 30 min. for preparing the closed nanochannel, a 10-fold excess of 15nt lock strands was added to the tube, meanwhile, for the open condition, the same volume of 1×TAE-$Mg^{2+}$ buffer was added. After a period of incubation (37 °C, 30 min), the assembled enzyme cascade on DNA origami (0.5 nM) was then mixed with glucose (10 mM) and indicator $ABTS^{2-}$ (0.5 mM) The enzyme cascade activity was measured by monitoring absorption value at 418 nm at 25 °C. For the data in figure 2b and S8, the three results obtained from independent experiments: firstly, 1×TAE-$Mg^{2+}$ buffer was added to the open sample, the same volume of 10-fold excess of lock strands were added to the closed and reopen sample; After a period of incubation (37 °C, 30 min), 1×TAE-$Mg^{2+}$ buffer was added to the open and closed sample, 10-fold excess of key strands were added to the reopen sample at 37 °C for 30 min. The remaining operations were same as before, these three samples were measured simultaneously.

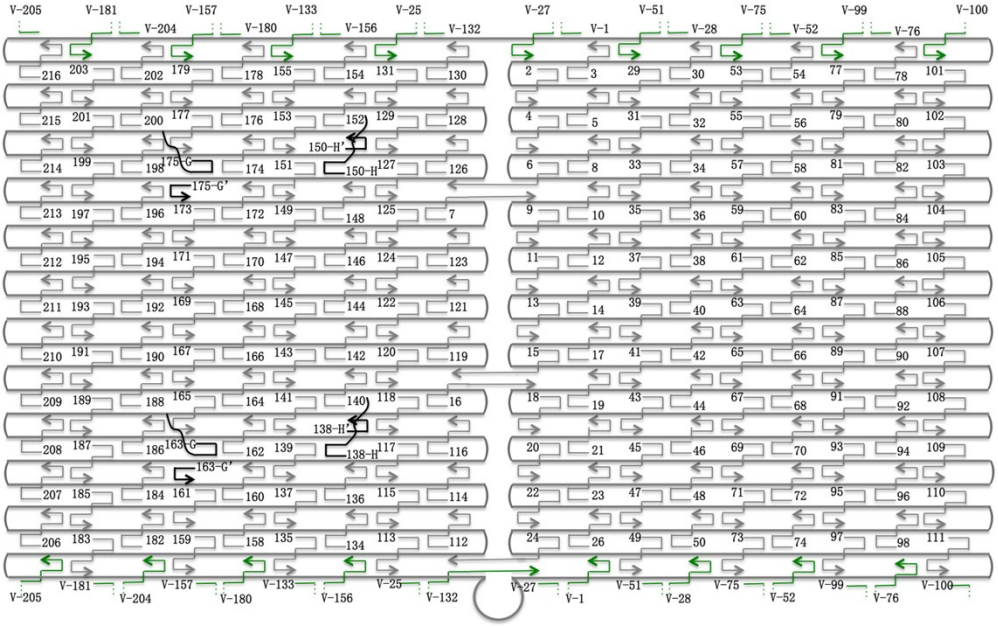
***AFM measurements.*** For each measurement, 5 uL of the sample was deposited onto a freshly cleaved mica surface and left to adsorb for 3 min. 30 uL of 1×TAE-$Mg^{2+}$ buffer was added to the liquid cell and the sample was scanned under ScanAsyst

model using a E scanner of AFM (Bruker Multimode 8). The proble used here was ScanAsyst Fluid+ (Olympus). The AFM analysis program without other treatment only flattened all the images.

**Materials**

All chemicals were purchased from Sigma-Aldrich or Alfa Aesar (Tianjing, China) and used without further purification. GOx and HRP were purchased from Sigma-Aldrich. All short oligo-DNA strands were purchased from Invitrogen. M13mp18 viral DNA was purchased from New England Biolabs. Crosslinker sulfo-EMCS was purchased from Pierce. Water used in all experiments was Milli-Q deionized (18.2 MΩ.cm).

**a**




**b**

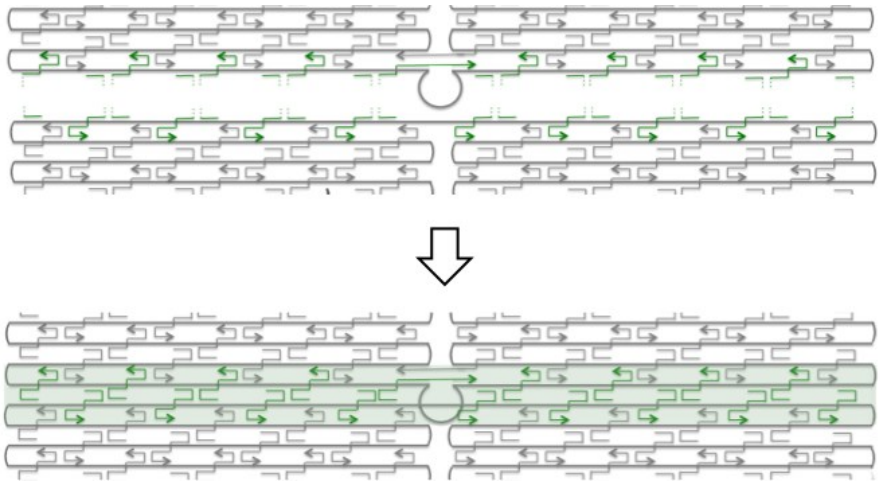


**Fig. S1** a) Design details of the DNA origami nanochannel with sticky ends between the top and bottom edges. The strand numbers are labeled at the 5' terminal. The green lines were the sticky ends，The black lines were designed to hybridize with DNA-enzyme complex. b) Design principle of the edge connections during the formation of DNA origami nanochannel.

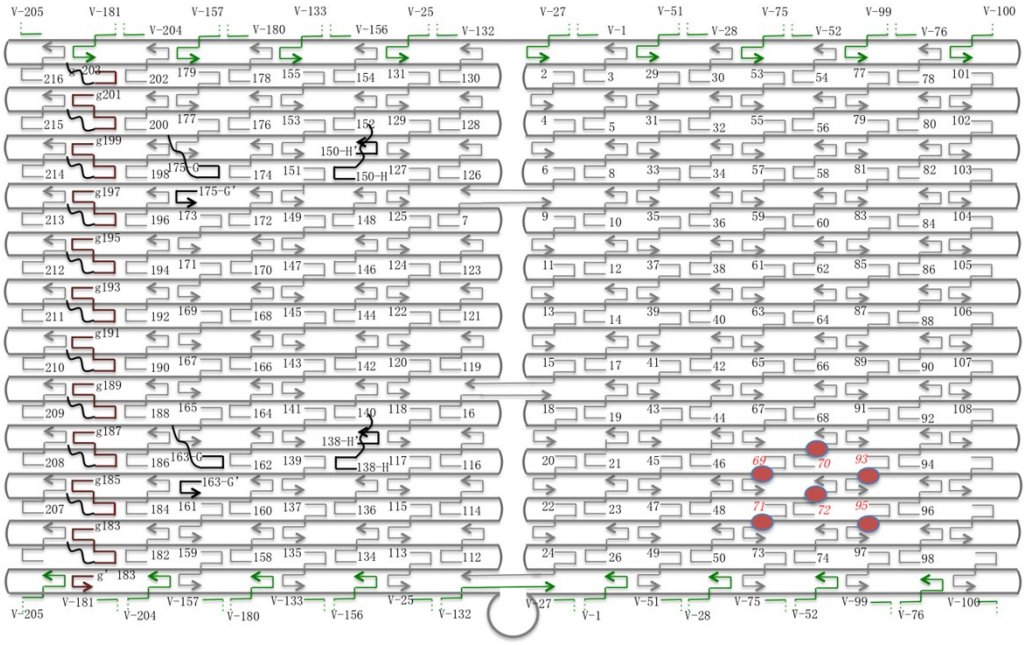



**Fig. S2** Design details of the 22 nm diameter DNA origami nanochannel with a shutter on the side of enzymes. For the construction of this kind of DNA origami nanochannel, Sequence g203, g201, g199, g197, g195, g193, g191, g189, g187, g185, g183 and g'183 were used to replace the sequences：203, 201, 199, 197, 195, 193, 191, 189, 187, 185 and 183. They were represented by the red lines with a black tail, which indicated the shutter strands. The red dots strands represented the index sequences.

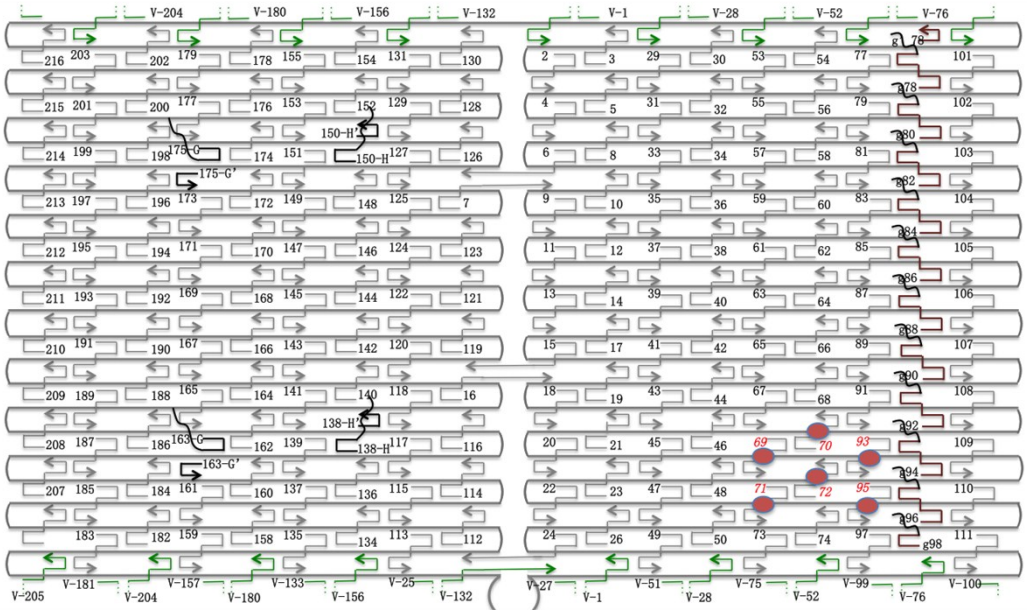


**Fig. S3** Design details of the 22 nm diameter DNA origami nanochannel with a shutter on the opposite side of enzymes. For the construction of this kind of DNA origami nanochannel, Sequence g98, g96, g94, g92, g90, g88, g86, g84, g82, g80, g78 and g'78 were used to replace the sequences：98, 96, 94, 92, 90, 88, 86, 84, 82, 80 and 78. They were represented by the red lines with a black tail, which indicated the shutter strands. The red dots strands represented the index sequences.

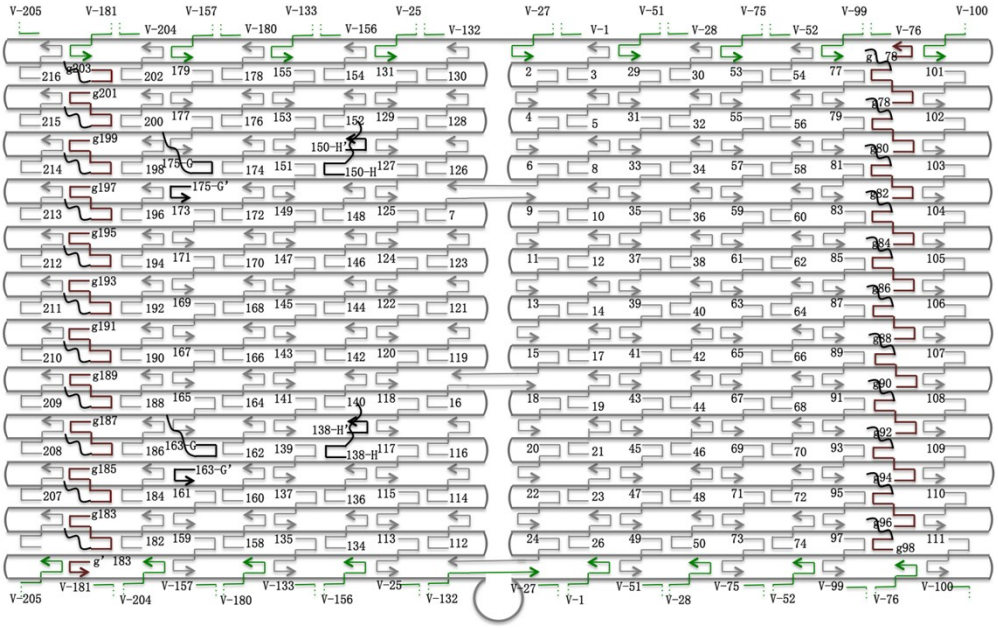


**Fig. S4** Design details of the 22 nm diameter DNA origami nanochannel with two shutters. For the construction of this kind of DNA origami nanochannel, Sequence g203, g201, g199, g197, g195, g193, g191, g189, g187, g185, g183, g'183, g98, g96, g94, g92, g90, g88, g86, g84, g82, g80, g78 and g'78 were used to replace the sequences: 203, 201, 199, 197, 195, 193, 191, 189, 187, 185, 183, 98, 96, 94, 92, 90, 88, 86, 84, 82, 80 and 78. They were represented by the red lines with a black tail, which indicated the shutter strands.

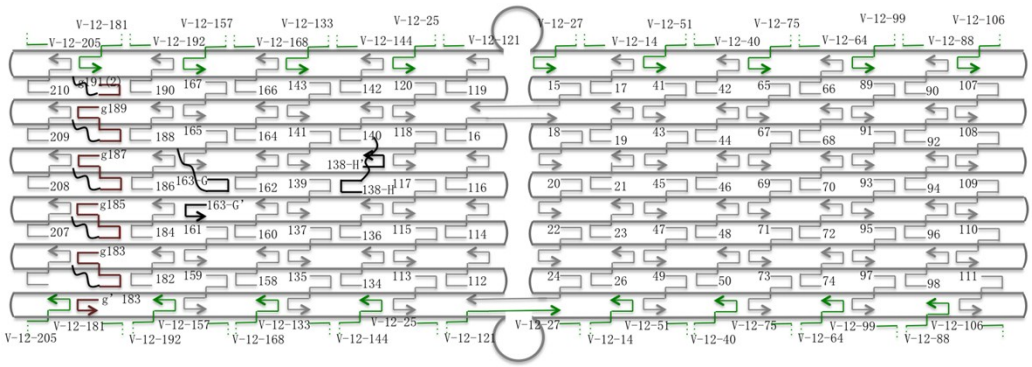



**Fig. S5** Design details of the 12 nm diameter DNA origami nanochannel with one shutter on the side of enzymes. For the construction of this kind of DNA origami nanochannel, Sequence 204, 180, 156, 132, 1, 28, 52, 76, 100, 216, 203, 202, 179, 178, 155, 154, 131, 130, 2, 3, 29, 30, 53, 54, 77, 78, 101, 215, 201, 200, 177, 176, 153, 152, 129, 128, 4, 5, 31, 32, 55, 56, 79, 80, 102, 214, 199, 198, 175-G, 175-G', 174, 51, 150-H, 150-H', 127, 126, 6, 8, 33, 34, 57, 58, 81, 82, 103, 213, 197, 196, 173, 172, 149, 148, 125, 7, 9, 10, 35, 36, 59, 60, 83, 84, 104, 212, 195, 194, 171, 170, 147, 146, 124, 123, 11, 12, 37, 38, 61, 62, 85, 86, 105, 211, 193, 169, 145, 122, 13, 39, 63, 87 were omitted. Sequences V-12-205, V-12-181, V-12-192, V-12-157, V-12-168, V-12-133, V-12-144, V-12-25, V-12-121, V-12-27, V-12-14, V-12-51, V-12-40, V-12-75, V-12-64, V-12-99, V-12-88 and V-12-106 were used to replace the staple strands: Sequence V-205, 181, V-192, 157, V-168, 133, V-144, 25, V-121, 27, V-14, 51, V-40, 75, V-64, 99, V-88, 106.

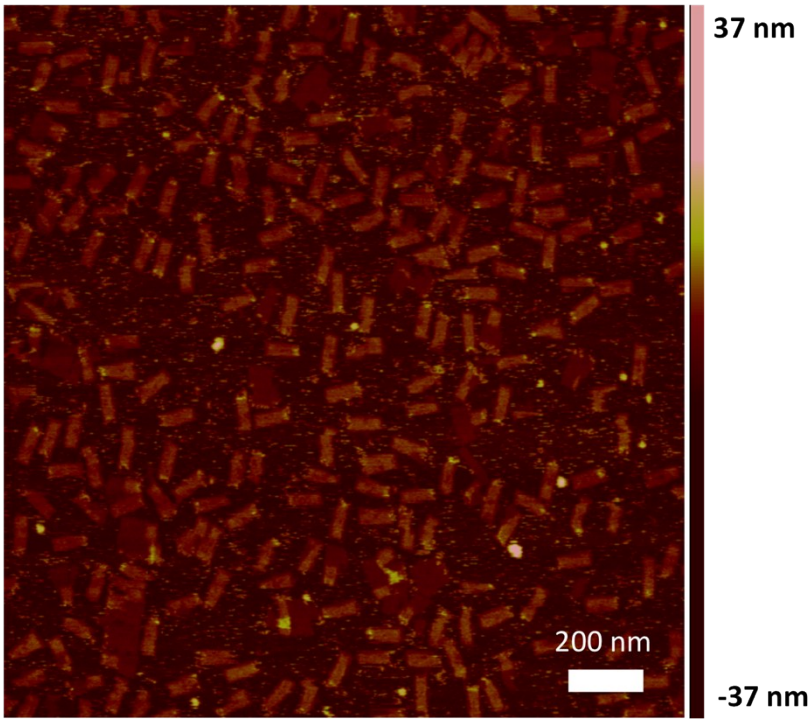



**Fig. S6** A large scale of AFM image for closed state of DNA nanochannel

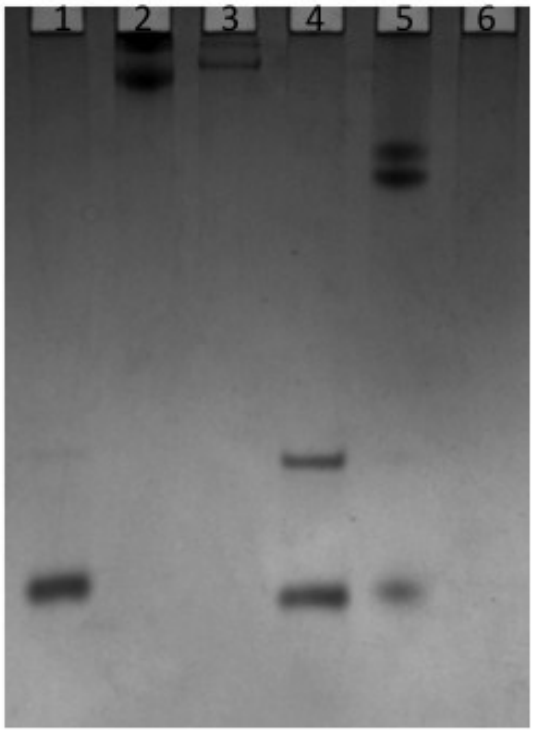



**Fig. S7** Enzyme-functionalized oligonucleotides were detected by 10% native PAGE 1) oligonucleotide; 2) GOx-functionalized oligonucleotide; 3) GOx; 4) oligonucleotide, the slow band was the dimer of oligonucleotides; 5) HRP-functionalized oligonucleotide. From bottom to top: the residual oligonucleotide; one oligonucleotide modified HRP and two oligonucleotides modified HRP; 6) HRP

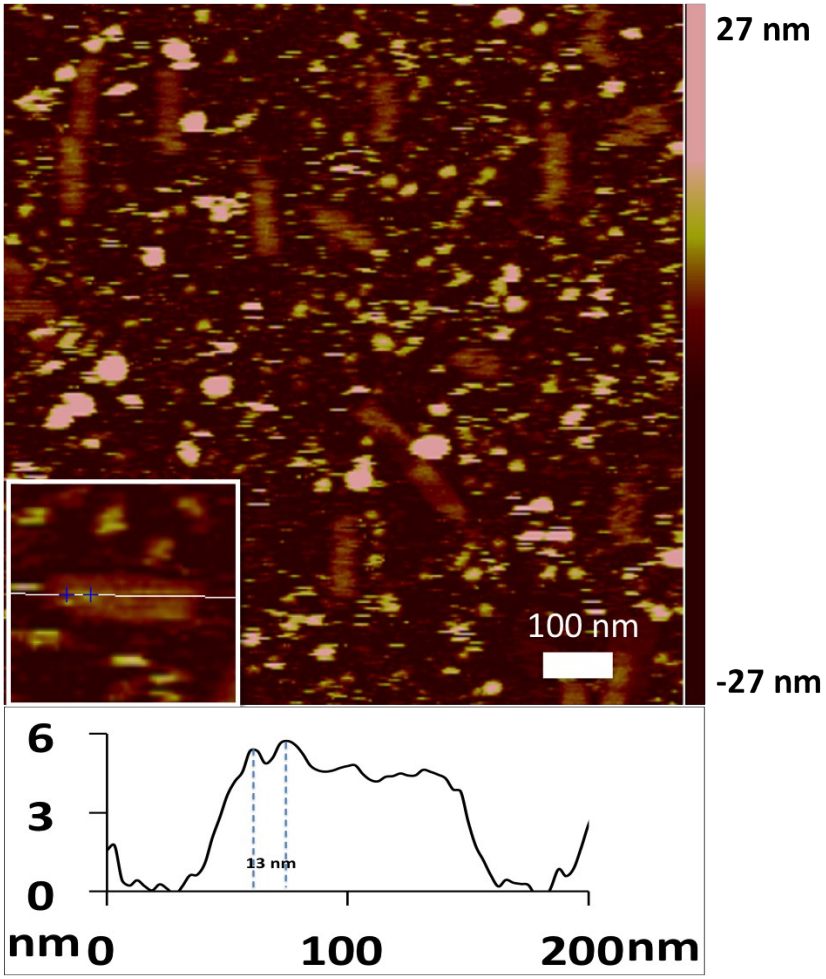



**Fig. S8** AFM images of the enzyme cascade on DNA nanochannels.

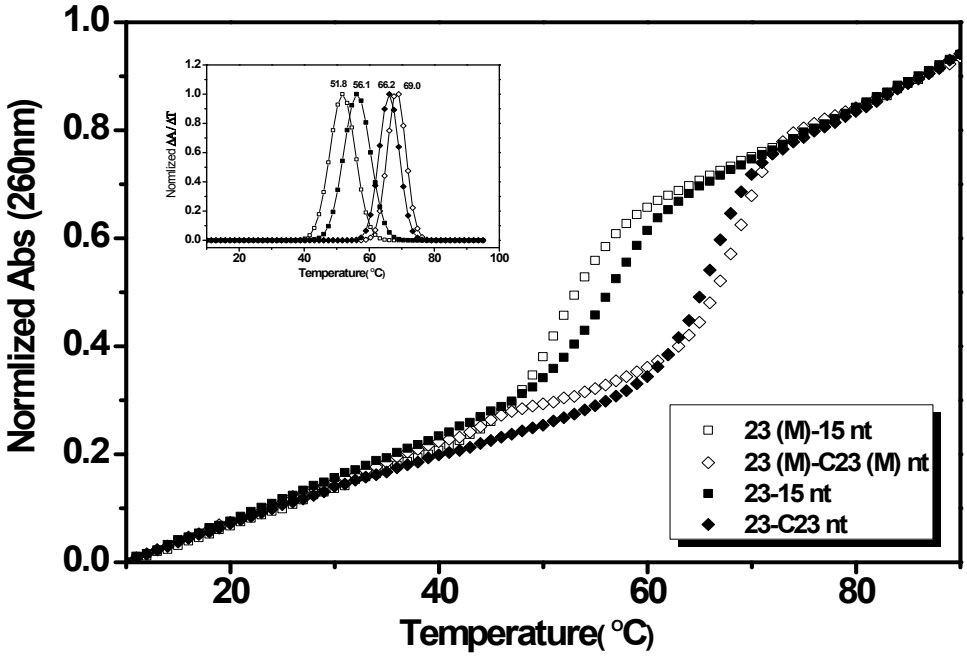



**Fig. S9** $T_m$ measurements of 15nt shutter strands and 23nt lock strands. The hollow shape represents 23bp lock strands with a mismatch; the solid shape represents 23bp lock strands without mismatch. UV melting experiments for absorption at 260 nm were carried out from 10 ℃ to 95 ℃ at a rate of 1 ℃/min. The concentration of each strand was 1 μM, Tm values can be obtained by the derivative of the corresponding UV absorption curves (see the Inset).

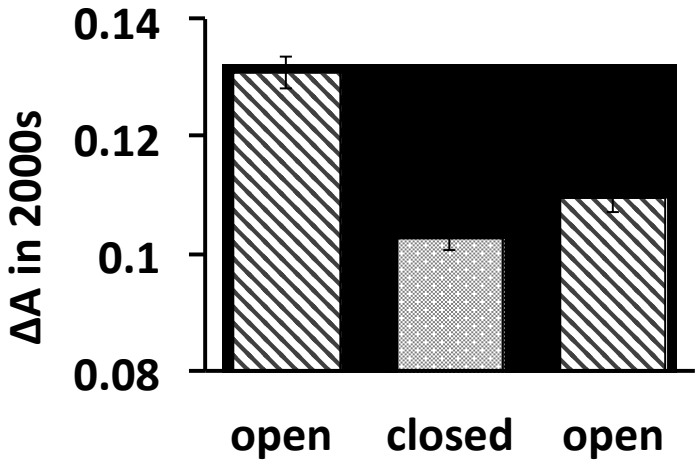



**Fig. S10** Absorbance increment in 2000 s of reaction controlled by the shutter state in one cycle. The lock strands complementary to the shutter strands without mismatch. The charts show results obtained from three independent experiments.

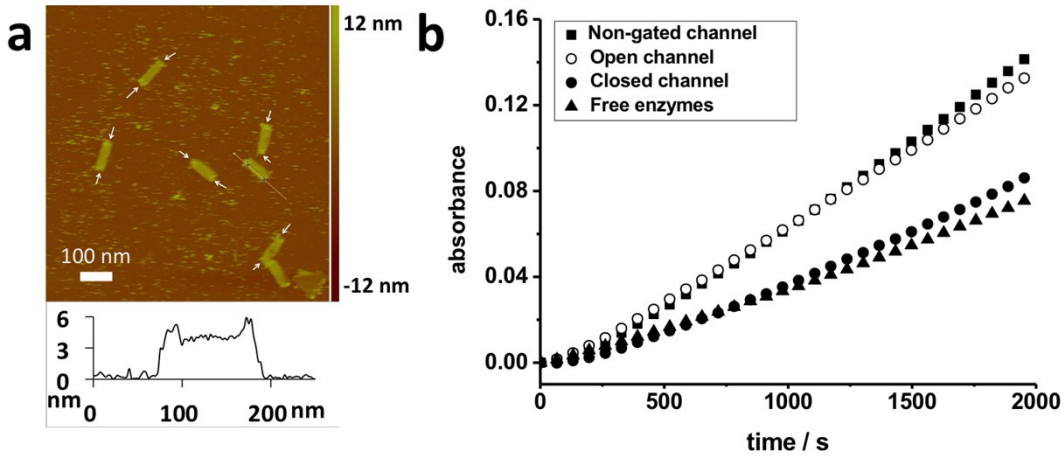



**Fig. S11** The nanochannel with two shutters on both ends. a) AFM image and height profile for closed DNA nanochannel, the height increment at both ends of nanochannel in height profile was due to the bond between biotin (modified at the 5' end of lock strands) and streptavidin; b) Plots of product concentration vs time for different state nanochannels and free enzymes. GOx: HRP: DNA nanochannel = 1 nM: 1 nM: 0.5 nM.

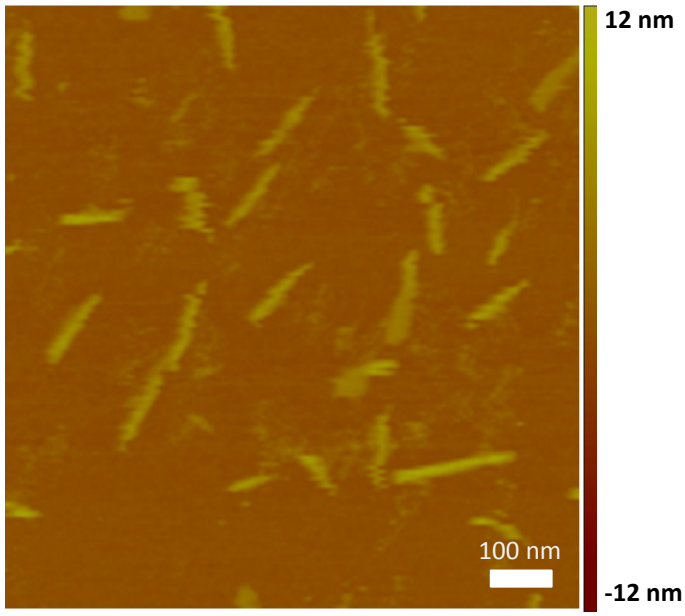



**Fig. S12** AFM image for open 12nm diameter nanochannel. There is no height increase at the end of nanochannel.

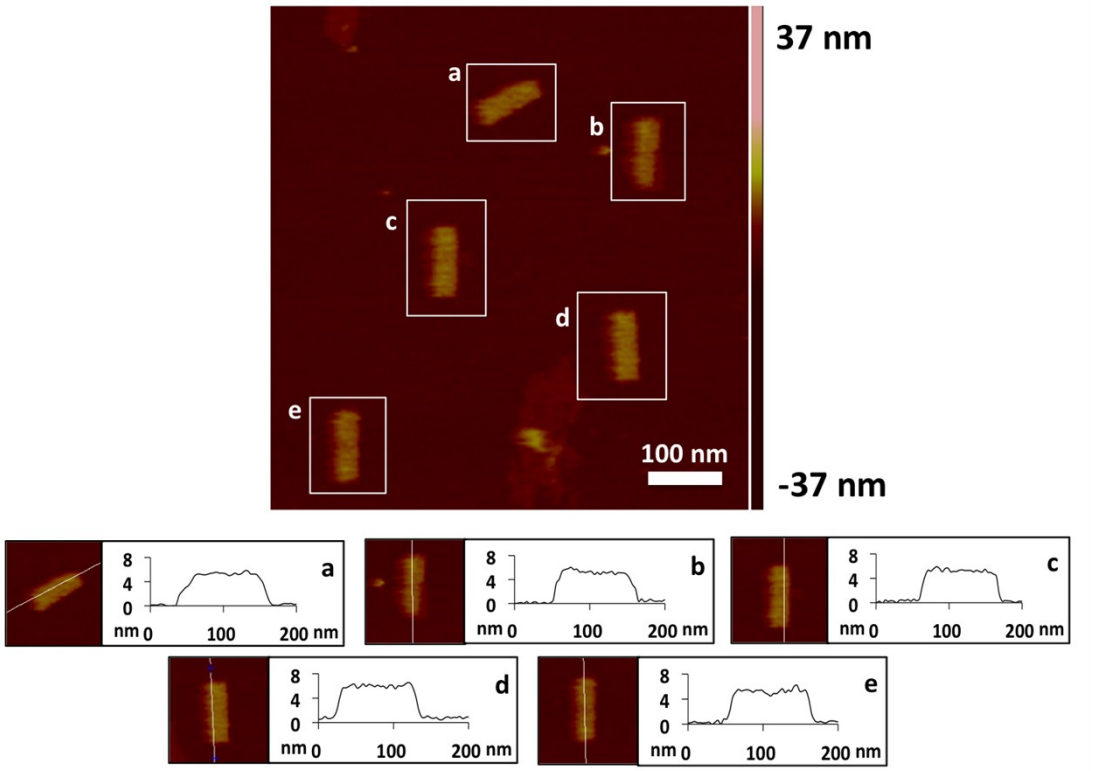



**Fig. S13** AFM images and height profiles for each structure in Figure 2c.

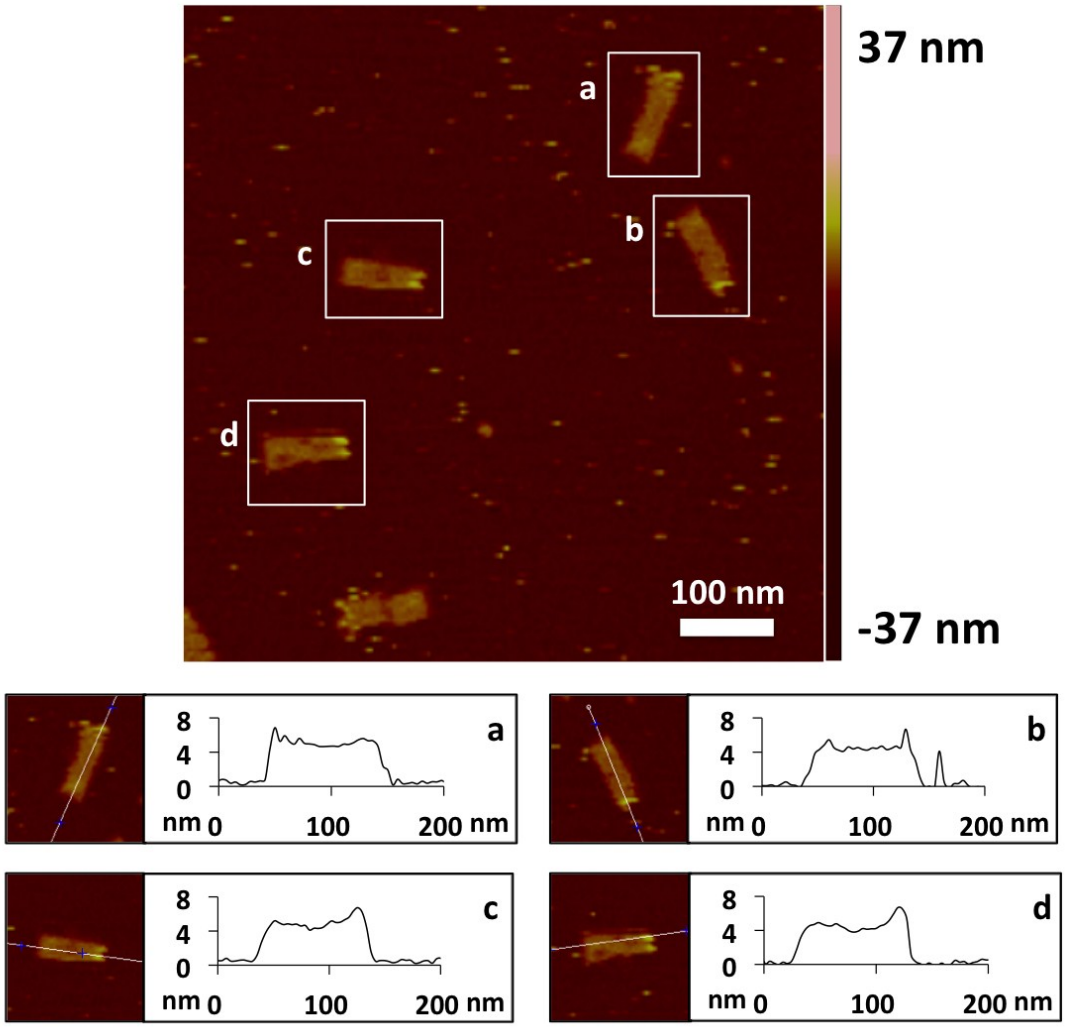



**Fig. S14** AFM images and height profiles for each structure in Figure 2d.

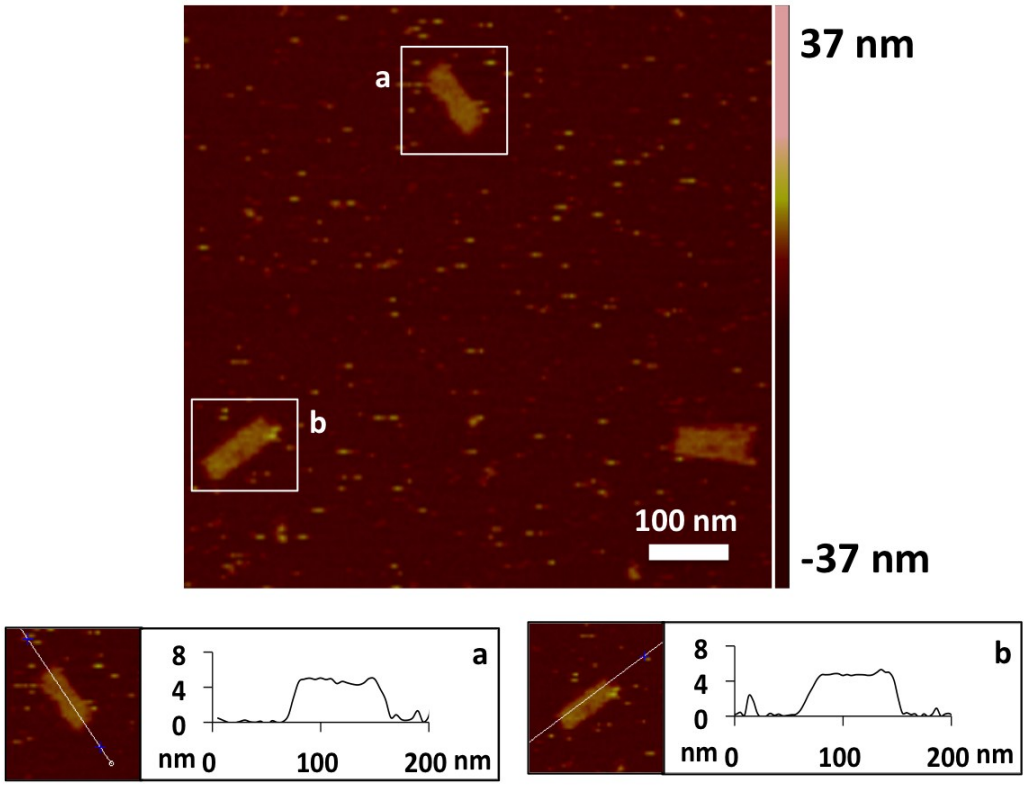



**Fig. S15** AFM images and height profiles for each structure in Figure 2e.

## Table S1 The sequences for the structure in Fig. S1

| | |
|---|---|
| V-1 | CGGCCTTGATAGGAACCCATGTACAAACAGTT |
| 2 | AATGCCCCGTAACAGTGCCCGTATCTCCCTCA |
| 3 | TGCCTTGACTGCCTATTTCGGAACAGGGATAG |
| 4 | GAGCCGCCCCACCACCGGAACCGCGACGGAAA |
| 5 | AACCAGAGACCCTCAGAACCGCCAGGGGTCAG |
| 6 | TTATTCATAGGGAAGGTAAATATTCATTCAGT |
| 7 | CATAACCCGAGGCATAGTAAGAGCTTTTTAAG |
| 8 | ATTGAGGGTAAAGGTGAATTATCAATCACCGG |
| 9 | AAAAGTAATATCTTACCGAAGCCCTTCCAGAG |
| 10 | GCAATAGCGCAGATAGCCGAACAATTCAACCG |
| 11 | CCTAATTTACGCTAACGAGCGTCTAATCAATA |
| 12 | TCTTACCAGCCAGTTACAAAATAAATGAAATA |
| 13 | ATCGGCTGCGAGCATGTAGAAACCTATCATAT |
| 14 | CTAATTTATCTTTCCTTATCATTCATCCTGAA |
| 15 | GCGTTATAGAAAAAGCCTGTTTAGAAGGCCGG |
| 16 | GCTCATTTTCGCATTAAATTTTTGAGCTTAGA |
| 17 | AATTACTACAAATTCTTACCAGTAATCCCATC |
| 18 | TTAAGACGTTGAAAACATAGCGATAACAGTAC |
| 19 | TAGAATCCCTGAGAAGAGTCAATAGGAATCAT |
| 20 | CTTTTACACAGATGAATATACAGTAAACAATT |
| 21 | TTTAACGTTCGGGAGAAACAATAATTTTCCCT |
| 22 | CGACAACTAAGTATTAGACTTTACAATACCGA |
| 23 | GGATTTAGCGTATTAAATCCTTTGTTTTCAGG |
| 24 | ACGAACCAAAACATCGCCATTAAA |
| V-25 | TGAGTTTCCGAGAAAGGAAGGGAACAAACTAT |
| 26 | TAGCCCTACCAGCAGAAGATAAAAACATTTGA |
| V-27 | CAAGCCCACTGGTAATATCCAGAACGAACTGA |
| V-28 | CCGCCAGCCACCACCCTCATTTTCCTATTATT |
| 29 | CTGAAACAGGTAATAAGTTTTAACCCCTCAGA |
| 30 | AGTGTACTTGAAAGTATTAAGAGGCCGCCACC |
| 31 | GCCACCACTCTTTTCATAATCAAACCGTCACC |
| 32 | GTTTGCCACCTCAGAGCCGCCACCGATACAGG |
| 33 | GACTTGAGAGACAAAAGGGCGACAAGTTACCA |
| 34 | AGCGCCAACCATTTGGGAATTAGATTATTAGC |
| 35 | GAAGGAAAATAAGAGCAAGAAACAACAGCCAT |
| 36 | GCCCAATACCGAGGAAACGCAATAGGTTTACC |
| 37 | ATTATTTAACCCAGCTACAATTTTCAAGAACG |
| 38 | TATTTTGCTCCCAATCCAAATAAGTGAGTTAA |
| 39 | GGTATTAAGAACAAGAAAAATAATTAAAGCCA |
| 40 | TAAGTCCTACCAAGTACCGCACTCTTAGTTGC |
| 41 | ACGCTCAAAATAAGAATAAACACCGTGAATTT |
| 42 | AGGCGTTACAGTAGGGCTTAATTGACAATAGA |
| 43 | ATCAAAATCGTCGCTATTAATTAACGGATTCG |
| 44 | CTGTAAATCATAGGTCTGAGAGACGATAAATA |
| 45 | CCTGATTGAAAGAAATTGCGTAGACCCGAACG |
| 46 | ACAGAAATCTTTGAATACCAAGTTCCTTGCTT |
| 47 | TTATTAATGCCGTCAATAGATAATCAGAGGTG |
| 48 | AGATTAGATTTAAAAGTTTGAGTACACGTAAA |
| 49 | AGGCGGTCATTAGTCTTTAATGCGCAATATTA |
| 50 | GAATGGCTAGTATTAACACCGCCTCAACTAAT |
| V-51 | CTCAGAGCCATTGCAACAGGAAAAATATTTTT |
| V-52 | GGAAATACACCGCCACCCTCAGAACTGAGACT |
| 53 | CCTCAAGAATACATGGCTTTTGATAGAACCAC |
| 54 | TAAGCGTCGAAGGATTAGGATTAGTACCGCCA |
| 55 | CACCAGAGTTCGGTCATAGCCCCCGCCAGCAA |
| 56 | TCGGCATTCCGCCGCCAGCATTGACGTTCCAG |
| 57 | AATCACCAAATAGAAAATTCATATATAACGGA |
| 58 | TCACAATCGTAGCACCATTACCATCGTTTTCA |
| 59 | ATACCCAAGATAACCCACAAGAATAAACGATT |
| 60 | ATCAGAGAAAGAACTGGCATGATTTTATTTTG |
| 61 | TTTTGTTTAAGCCTTAAATCAAGAATCGAGAA |
| 62 | AGGTTTTGAACGTCAAAAATGAAAGCGCTAAT |
| 63 | CAAGCAAGACGCGCCTGTTTATCAAGAATCGC |
| 64 | AATGCAGACCGTTTTTATTTTCATCTTGCGGG |
| 65 | CATATTTAGAAATACCGACCGTGTTACCTTTT |
| 66 | AATGGTTTACAACGCCAACATGTAGTTCAGCT |
| 67 | TAACCTCCATATGTGAGTGAATAAACAAAATC |
| 68 | AAATCAATGGCTTAGGTTGGGTTACTAAATTT |
| 69 | GCGCAGAGATATCAAAATTATTTGACATTATC |
| 70 | AACCTACCGCGAATTATTCATTTCCAGTACAT |
| 71 | ATTTTGCGTCTTTAGGAGCACTAAGCAACAGT |
| 72 | CTAAAATAGAACAAAGAAACCACCAGGGTTAG |
| 73 | GCCACGCTATACGTGGCACAGACAACGCTCAT |
| 74 | GCGTAAGAGAGAGCCAGCAGCAAAAAGGTTAT |
| V-75 | CCCTCAGACTACATTTTGACGCTCACCTGAAA |
| V-76 | GAAATGGATACTCAGGAGGTTTAGCGGGGTTT |
| 77 | TGCTCAGTCAGTCTCTGAATTTACCAGGAGGT |
| 78 | GGAAAGCGACCAGGCGGATAAGTGAATAGGTG |
| 79 | TGAGGCAGGCGTCAGACTGTAGCGTAGCAAGG |
| 80 | TGCCTTTAGTCAGACGATTGGCCTGCCAGAAT |
| 81 | CCGGAAACACACCACGGAATAAGTAAGACTCC |
| 82 | ACGCAAAGGTCACCAATGAAACCAATCAAGTT |

| | |
|---|---|
| 83 | TTATTACGGTCAGAGGGTAATTGAATAGCAGC |
| 84 | TGAACAAACAGTATGTTAGCAAACTAAAAGAA |
| 85 | CTTTACAGTTAGCGAACCTCCCGACGTAGGAA |
| 86 | GAGGCGTTAGAGAATAACATAAAAGAACACCC |
| 87 | TCATTACCCGACAATAAACAACATATTTAGGC |
| 88 | CCAGACGAGCGCCCAATAGCAAGCAAGAACGC |
| 89 | AGAGGCATAATTTCATCTTCTGACTATAACTA |
| 90 | TTTTAGTTTTTCGAGCCAGTAATAAATTCTGT |
| 91 | TATGTAAACCTTTTTTAATGGAAAAATTACCT |
| 92 | TTGAATTATGCTGATGCAAATCCACAAATATA |
| 93 | GAGCAAAAACTTCTGAATAATGGAAGAAGGAG |
| 94 | TGGATTATGAAGATGATGAAACAAAATTTCAT |
| 95 | CGGAATTATTGAAAGGAATTGAGGTGAAAAAT |
| 96 | ATCAACAGTCATCATATTCCTGATTGATTGTT |
| 97 | CTAAAGCAAGATAGAACCCTTCTGAATCGTCT |
| 98 | GCCAACAGTCACCTTGCTGAACCTGTTGGCAA |
| V-99 | TATCACCGTTATTTACATTGGCAGACATTCTG |
| V-100 | GTCACACGTTTTTATAAGTATAGCCCGGCCGTC GAG |
| 101 | AGGGTTGATTTTATAAATCCTCATTAAATGAT ATTC |
| 102 | ACAAACAATTTTAATCAGTAGCGACAGATCGAT AGC |
| 103 | AGCACCGTTTTTTAAAGGTGGCAACATAGTAGA AAA |
| 104 | TACATACATTTTGACGGGAGAATTAACTACAGG GAA |
| 105 | GCGCATTATTTTGCTTATCCGGTATTCTAAATC AGA |
| 106 | TATAGAAGTTTTCGACAAAAGGTAAAGTAGAG AATA |
| 107 | TAAAGTACTTTTCGCGAGAAAACTTTTTATCGC AAG |
| 108 | ACAAAGAATTTTATTAATTACATTTAACACATC AAG |
| 109 | AAAACAAATTTTTTCATCAATATAATCCTATCA GAT |
| 110 | GATGGCAATTTTAATCAATATCTGGTCACAAAT ATC |
| 111 | AAACCCTCTTTTACCAGTAATAAAAGGGATTCA CCAGTCACACGTTTT |
| 112 | CCGAAATCCGAAAATCCTGTTTGAAGCCGGAA |
| 113 | CCAGCAGGGGCAAAATCCCTTATAAAGCCGGC |
| 114 | GCATAAAGTTCCACACAACATACGAAGCGCCA |
| 115 | GCTCACAATGTAAAGCCTGGGGTGGGTTTGCC |
| 116 | TTCGCCATTGCCGGAAACCAGGCATTAAATCA |
| 117 | GCTTCTGGTCAGGCTGCGCAACTGTGTTATCC |
| 118 | GTTAAAATTTTAACCAATAGGAACCCGGCACC |
| 119 | AGACAGTCATTCAAAAGGGTGAGAAGCTATAT |
| 120 | AGGTAAAGAAATCACCATCAATATAATATTTT |
| 121 | TTTCATTTGGTCAATAACCTGTTTATATCGCG |
| 122 | TCGCAAATGGGGCGCGAGCTGAAATAATGTGT |
| 123 | TTTTAATTGCCCGAAAGACTTCAAAACACTAT |
| 124 | AAGAGGAACGAGCTTCAAAGCGAAGATACATT |
| 125 | GGAATTACTCGTTTACCAGACGACAAAAGATT |
| 126 | GAATAAGGACGTAACAAAGCTGCTCTAAAACA |
| 127 | CCAAATCACTTGCCCTGACGAGAACGCCAAAA |
| 128 | CTCATCTTGAGGCAAAAGAATACAGTGAATTT |
| 129 | AAACGAAATGACCCCCAGCGATTATTCATTAC |
| 130 | CTTAAACATCAGCTTGCTTTCGAGCGTAACAC |
| 131 | TCGGTTTAGCTTGATACCGATAGTCCAACCTA |
| V-132 | GAACGTGGGTCACCAGTACAAACTTAATTGTA |
| V-133 | TGTAGCATTAGAGCTTGACGGGGAAATCAAAA |
| 134 | GAATAGCCGCAAGCGGTCCACGCTCCTAATGA |
| 135 | GAGTTGCACGAGATAGGGTTGAGTAAGGGAGC |
| 136 | GTGAGCTAGTTTCCTGTGTGAAATTTGGGAAG |
| 137 | TCATAGCTACTCACATTAATTGCGCCCTGAGA |
| 138-H | GGCGATCGCACTCCAGTTTGACTACTGACGCGG ACATTC |
| 138-H’ | CCAGCTTTGCCATCAA |
| 139 | GAAGATCGGTGCGGGCCTCTTCGCAATCATGG |
| 140 | AAATAATTTTAAATTGTAAACGTTGATATTCA |
| 141 | GCAAATATCGCGTCTGGCCTTCCTGGCCTCAG |
| 142 | ACCGTTCTAAATGCAATGCCTGAGAGGTGGCA |
| 143 | TATATTTTAGCTGATAAATTAATGTTGTATAA |
| 144 | TCAATTCTTTTAGTTTGACCATTACCAGACCG |
| 145 | CGAGTAGAACTAATAGTAGTAGCAAACCCTCA |
| 146 | GAAGCAAAAAAGCGGATTGCATCAGATAAAAA |
| 147 | TCAGAAGCCTCCAACAGGTCAGGATCTGCGAA |
| 148 | CCAAAATATAATGCAGATACATAAACACCAGA |
| 149 | CATTCAACGCGAGAGGCTTTTGCATATTATAG |
| 150-H | ACGAGTAGTGACAAGATTTGACTACTGACGCGG ACATTC |
| 150-H’ | ACCGGATATACCAAGC |
| 151 | AGTAATCTTAAATTGGGCTTGAGAGAATACCA |
| 152 | GCGAAACATGCCACTACGAAGGCATGCGCCGA |

| | |
|---|---|
| 153 | ATACGTAAAAGTACAACGGAGATTTCATCAAG |
| 154 | CAATGACACTCCAAAAGGAGCCTTACAACGCC |
| 155 | AAAAAAGGACAACCATCGCCCACGCGGGTAAA |
| V-156 | CCCCGATTTCCACAGACAGCCCTCATCTCCAA |
| V-157 | CGTAACGACTAAATCGGAACCCTAGTTGTTCC |
| 158 | AGTTTGGAGCCCTTCACCGCCTGGTTGCGCTC |
| 159 | AGCTGATTACAAGAGTCCACTATTGAGGTGCC |
| 160 | ACTGCCCGCCGAGCTCGAATTCGTTATTACGC |
| 161 | CCCGGGTACTTTCCAGTCGGGAAACGGGCAAC |
| 162 | CAGCTGGCGGACGACGACAGTATCGTAGCCAG |
| 163-G | GTTTGAGGGAAAGGGGTTTATTCTACTTGAGA GAGCGA |
| 163-G’ | GATGTGCTAGAGGATC |
| 164 | CTTTCATCCCCAAAAACAGGAAGACCGGAGAG |
| 165 | AGAAAAGCAACATTAAATGTGAGCATCTGCCA |
| 166 | GGTAGCTAGGATAAAAATTTTTAGTTAACATC |
| 167 | CAACGCAATTTTTGAGAGATCTACTGATAATC |
| 168 | CAATAAATACAGTTGATTCCCAATTTAGAGAG |
| 169 | TCCATATACATACAGGCAAGGCAACTTTATTT |
| 170 | TACCTTTAAGGTCTTTACCCTGACAAAGAAGT |
| 171 | CAAAAATCATTGCTCCTTTTGATAAGTTTCAT |
| 172 | TTTGCCAGATCAGTTGAGATTTAGTGGTTTAA |
| 173 | AAAGATTCAGGGGGTAATAGTAAACCATAAAT |
| 174 | TTTCAACTATAGGCTGGCTGACCTTGTATCAT |
| 175-G | CCAGGCGCTTAATCATTTTATTCTACTTGAGAG AGCGAC |
| 175-G’ | TGTGAATTACAGGTAG |
| 176 | CGCCTGATGGAAGTTTCCATTAAACATAACCG |
| 177 | TTTCATGAAAATTGTGTCGAAATCTGTACAGA |
| 178 | ATATATTCTTTTTTCACGTTGAAAATAGTTAG |
| 179 | AATAATAAGGTCGCTGAGGCTTGCAAAGACTT |
| V-180 | GTAAAGCATCTAAAGTTTTGTCGTGAATTGCG |
| V-181 | ACGTTAGTCAAGTTTTTTGGGGTCAAAGAACG |
| 182 | TGGACTCCCTTTTCACCAGTGAGACCTGTCGT |
| 183 | TGGTTTTTAACGTCAAAGGGCGAAGAACCATC |
| 184 | GCCAGCTGCCTGCAGGTCGACTCTGCAAGGCG |
| 185 | CTTGCATGCATTAATGAATCGGCCCGCCAGGG |
| 186 | ATTAAGTTCGCATCGTAACCGTGCGAGTAACA |
| 187 | TAGATGGGGGGTAACGCCAGGGTTGTGCCAAG |
| 188 | ACCCGTCGTCATATGTACCCCGGTAAAGGCTA |
| 189 | CATGTCAAGATTCTCCGTGGGAACCGTTGGTG |
| 190 | TCAGGTCACTTTTGCGGGAGAAGCAGAATTAG |
| 191 | CTGTAATATTGCCTGAGAGTCTGGAAAACTAG |
| 192 | CAAAATTAAAGTACGGTGTCTGGAAGAGGTCA |
| 193 | TGCAACTAAGCAATAAAGCCTCAGTTATGACC |
| 194 | TTTTTGCGCAGAAAACGAGAATGAATGTTTAG |
| 195 | AAACAGTTGATGGCTTAGAGCTTATTTAAATA |

## Table S2 The sequences for the structures in Fig. S2-S5

| | |
|---|---|
| V-12-181 | ACCCAAATAGCAATAAAGCCTCAGTTATGACC |
| V-12-192 | CAAAATTACAAGTTTTTTGGGGTCAAAGAACG |
| V-12-157 | GTAAAGCACATACAGGCAAGGCAACTTTATTT |
| V-12-168 | CAATAAATCTAAATCGGAACCCTAGTTGTTCC |
| V-12-133 | CCCCGATTACTAATAGTAGTAGCAAACCCTCA |
| V-12-144 | TCAATTCTTAGAGCTTGACGGGGAAATCAAAA |
| V-12-25 | GAACGTGGGGGGCGCGAGCTGAAATAATGTGT |
| V-12-121 | TTTCATTTCGAGAAAGGAAGGGAACAAACTAT |
| V-12-27 | CGGCCTTGCGAGCATGTAGAAACCTATCATAT |
| V-12-14 | CTAATTTACTGGTAATATCCAGAACGAACTGA |
| V-12-51 | CCGCCAGCGAACAAGAAAAATAATTAAAGCCA |
| V-12-40 | TAAGTCCTCATTGCAACAGGAAAAATATTTTT |
| V-12-75 | GGAAATACACGCGCCTGTTTATCAAGAATCGC |
| V-12-64 | AATGCAGACTACATTTTGACGCTCACCTGAAA |
| V-12-99 | GAAATGGACGACAATAAACAACATATTTAGGC |
| V-12-88 | CCAGACGATTATTTACATTGGCAGACATTCTG |
| V-12-106 | GTCACACGTTTTCGACAAAAGGTAAAGTAGAGAATA |
| V-12-181 | ACCCAAATAGCAATAAAGCCTCAGTTATGACC |
| V-12-192 | CAAAATTACAAGTTTTTTGGGGTCAAAGAACG |
| V-12-157 | GTAAAGCACATACAGGCAAGGCAACTTTATTT |
| V-12-168 | CAATAAATCTAAATCGGAACCCTAGTTGTTCC |

| | |
|---|---|
| V-12-133 | CCCCGATTACTAATAGTAGTAGCAAACCCTCA |
| V-12-144 | TCAATTCTTAGAGCTTGACGGGGAAATCAAAA |
| V-12-25 | GAACGTGGGGGGCGCGAGCTGAAATAATGTGT |
| V-12-121 | TTTCATTTCGAGAAAGGAAGGGAACAAACTAT |
| V-12-27 | CGGCCTTGCGAGCATGTAGAAACCTATCATAT |
| V-12-14 | CTAATTTACTGGTAATATCCAGAACGAACTGA |
| V-12-51 | CCGCCAGCGAACAAGAAAAATAATTAAAGCCA |
| V-12-40 | TAAGTCCTCATTGCAACAGGAAAAATATTTTT |
| V-12-75 | GGAAATACACGCGCCTGTTTATCAAGAATCGC |
| V-12-64 | AATGCAGACTACATTTTGACGCTCACCTGAAA |
| V-12-99 | GAAATGGACGACAATAAACAACATATTTAGGC |
| V-12-88 | CCAGACGATTATTTACATTGGCAGACATTCTG |
| V-12-106 | GTCACACGTTTTCGACAAAAGGTAAAGTAGAGAATA |
| 8bp lock strand | GAGGATAG |
| 15bp lock strand | GTGATGAGAGGATAG |
| 23bp lock strand without mismatch | GTTAGTGAGTGATGAGAGGATAG |
| Key strand without mismatch | CTATCCTCTCATCACTCACTAAC |
| 23bp lock strand | GTTAGTGAGTGATGGGAGGATAG |
| Key strand | CTATCCTCCCATCACTCACTAAC |
| Strand conjugated to GOx | HS-GTCGCTCTCTCAAGTAGAAT |
| Strand conjugated to HRP | HS-GAATGTCCGCGTCAGTAGTC |
| Index-93 | GAGCAAAAACTTCTGATCCTCTTTGAGGAACAAGTTTCTTGTATAATGGAAGAAGGAG |

| | |
|---|---|
| Index-95 | CGGAATTATTGAAAGGTCC<br>TCTTTGAGGAACAAGTTTCT<br>TGTAATTGAGGTGAAAAAT |
| Index-72 | CTAAAATAGAACAAAGTCCT<br>CTTTGAGGAACAAGTTTCTT<br>GTAAACCACCAGGGTTAG |
| Index-70 | AACCTACCGCGAATTATCCT<br>CTTTGAGGAACAAGTTTCTT<br>GTTTCATTTCCAGTACAT |
| Index-69 | GCGCAGAGATATCAAATCCT<br>CTTTGAGGAACAAGTTTCTT<br>GT ATTATTTGACATTATC |
| Index-71 | ATTTTGCGTCTTTAGGTCCT<br>CTTTGAGGAACAAGTTTCTT<br>GTAGCACTAAGCAACAGT |
| | |