

Supporting Information

Macromolecule Sensing and Tumor Biomarker Detection by Harnessing Viral DNA Packaging Motor Channels into Membranes and Flow Cells

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Table S1: Oligos for mutations construction

Mutations	Oligos for mutations construction
Phi29-1	5'-CAAG TGTATAACCA GTATGAAGGT ATTGCC TTA G TTATCTTCGC TCAC ATA GCT C TCTTCATTCTCTCTATAGAAGTG-3' 5'-CACTTCTATAGAGAGAATGAAGAGAGCTATGTGAGCGAAGATAACTAAGGCAATACCTTCATACTGGTTATACACTTG-3'
Phi29-2	5'-ATATAGGATCCATGATACAGCGTCAAAAACGGAATAG-3' 5'-ATATACTCGAGCTAATGGTGATGGTGATGGTGGCCACCCTCAITTTGTTTCACCGT-3'
Phi29-3	5'-CAAG TGTATAACCA GTATGAAGGT ATTGCC TTA G TTATCTTCGC TCAC ATA GCT C TCTTCATTCTCTCTATAGAAGTG-3' 5'-CACTTCTATAGAGAGAATGAAGAGAGCTATGTGAGCGAAGATAACTAAGGCAATACCTTCATACTGGTTATACACTTG-3'
Phi29-4	5'-GACACCCGCTTAAATTAGAGCAAATGACATTATCCAACCTGAGCTTAAACAAG-3' 5'-CTTGTTTAAAGCTCAGTTGGATAATGTCAITTTGCTCTAATTAAGACGGGTGTC-3' 5'-TGCCCTGTTATCTTCGCTCACATAGCTCTCTTCAGTGACTCTATAGAAGTGTTTAAG-3' 5'-CTTAAACACTTCTATAGAGTCACTGAAGAGAGCTATGTGAGCGAAGATAACAGGGGCA-3'
C-Δ25 Phi29	5' oilgo-ATATCGCTAGCATGCACCACCATCACCATCAGCA CGTAAACGCAGTAACACATAC 3' oilgo-CTTTAACAATTGAGACCATATAGTAGATTAACTTTTAGACATGCTTCTTCCCTCGAC 3'-oligo:TATATCTCGAGCTAGATTAGATATCTGAATTAACTTTAACATTGAGACCATATAGTA
T7	5'-CTTAGTGAATCCTGCTGGTATCACCAGCTAATCATACTGACCTTTGCTCAGACTGGTGACTTCG TTA CTGGTCG-3' 5'-CGACCAGTAACGAAGTCACCAGTCTGAGCAAAGGTCAGTATGATTAGCTGGGTGATACCAGCAGGATTCTACTAAG-3'

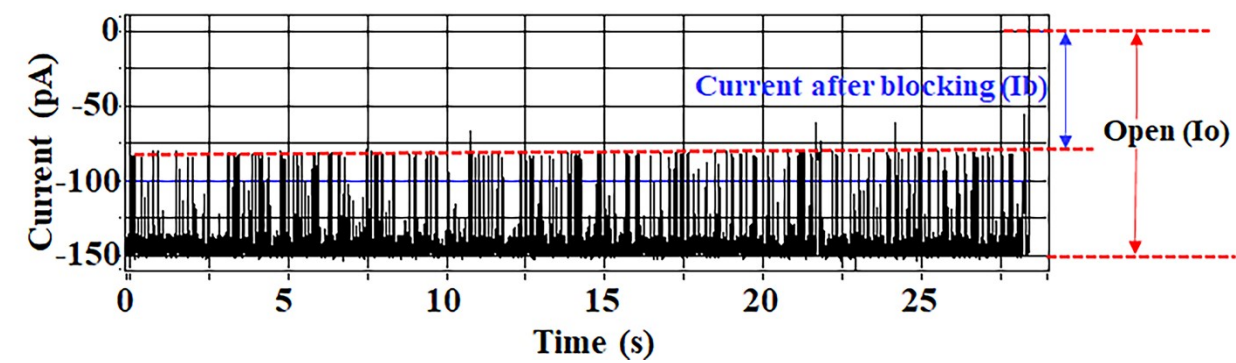


Figure S1: Illustration of the measurement of current blockage in this study.

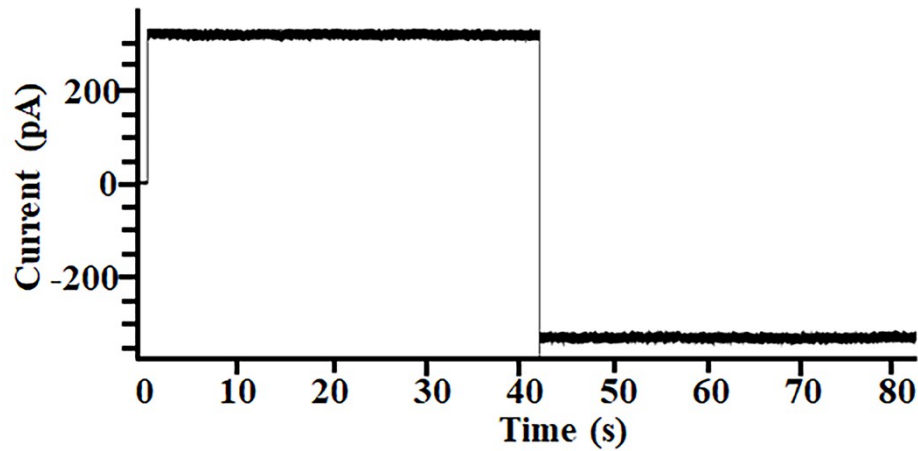


Figure S2: Current trace showing no R12 translocation was observed on the patch-clamp system using the Phi29 connector N-terminus insertion at ± 50 mV. Peptides (1mg/mL) were added to the *Cis* chamber in 1M KCl, 5mM HEPES, pH7.4 buffer.

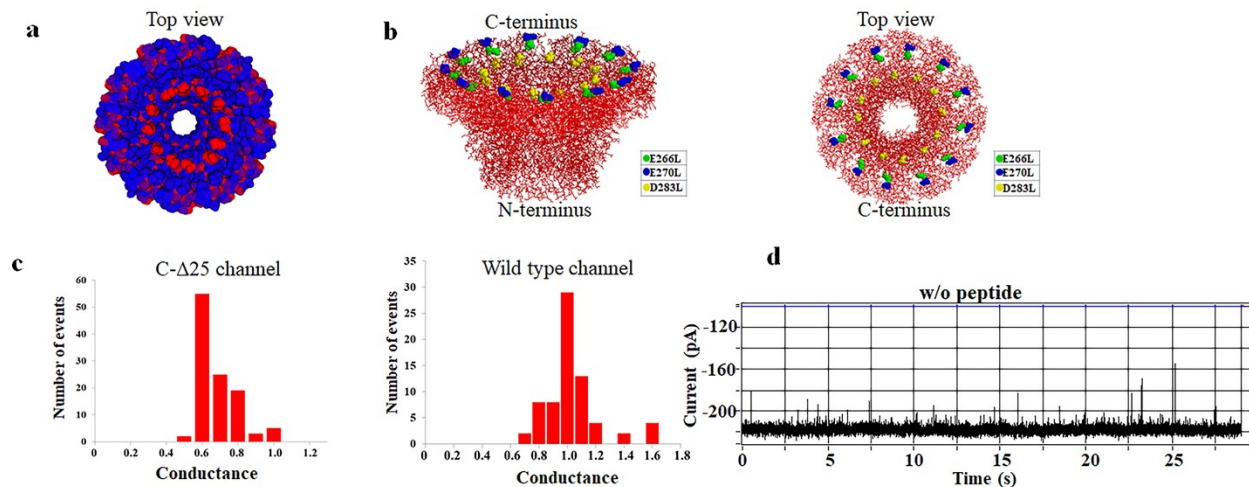


Figure S3: Construction of C-terminal mutants of Phi29 connector channel for direct membrane insertion and peptide translocation. (a) PDB structure shows the hydrophobic and hydrophilic amino acids distribution at the C-terminus of Phi29 connect channels. (Blue, hydrophilic amino acid; red, hydrophobic amino acids). (b) PDB structure derived images showing the sites of mutation of C-terminal mutant Phi29 connector. (c) Histogram of conductance of C-Δ25 and wild-type Phi29 connector. Buffer: 0.15 M KCl, 5 mM HEPES, pH 7.4. (d) Current trace before the addition of R12 to C-Δ25 channel.

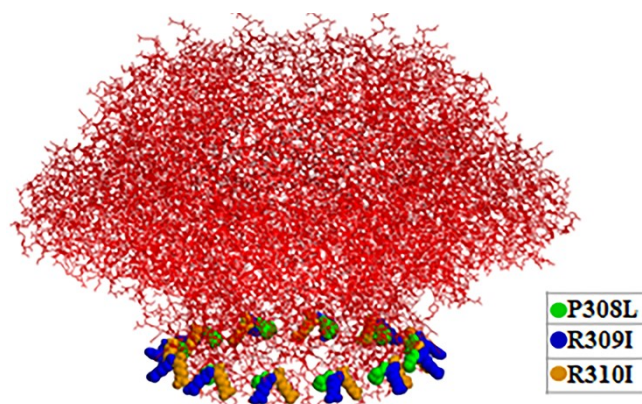


Figure S4: PDB structure derived images showing the mutation sites of the T7 connector.

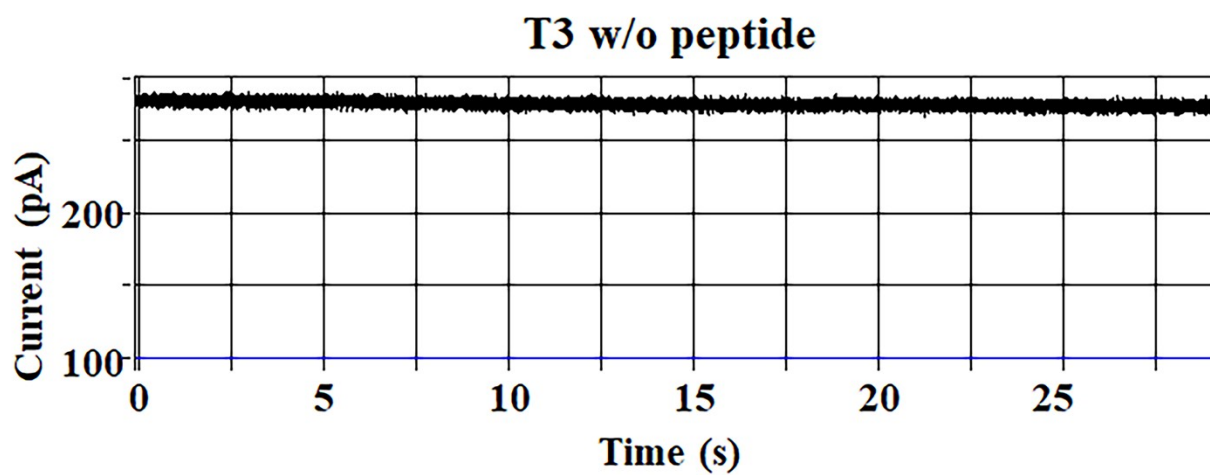


Figure S5: Current trace before the addition of R12 to the T3 channel.

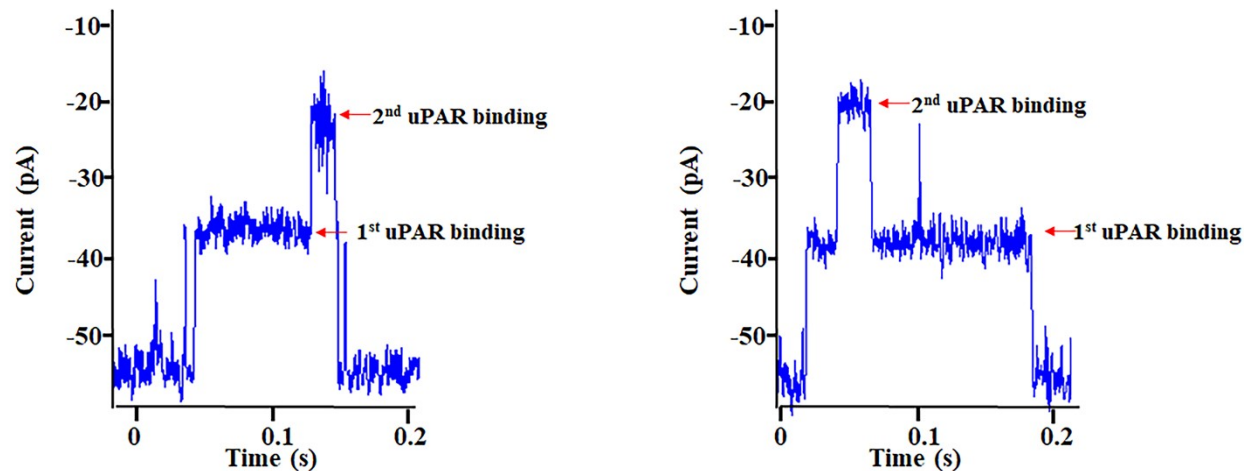


Figure S6: Current signature of two uPAR proteins binding to the C-terminal probe simultaneously.

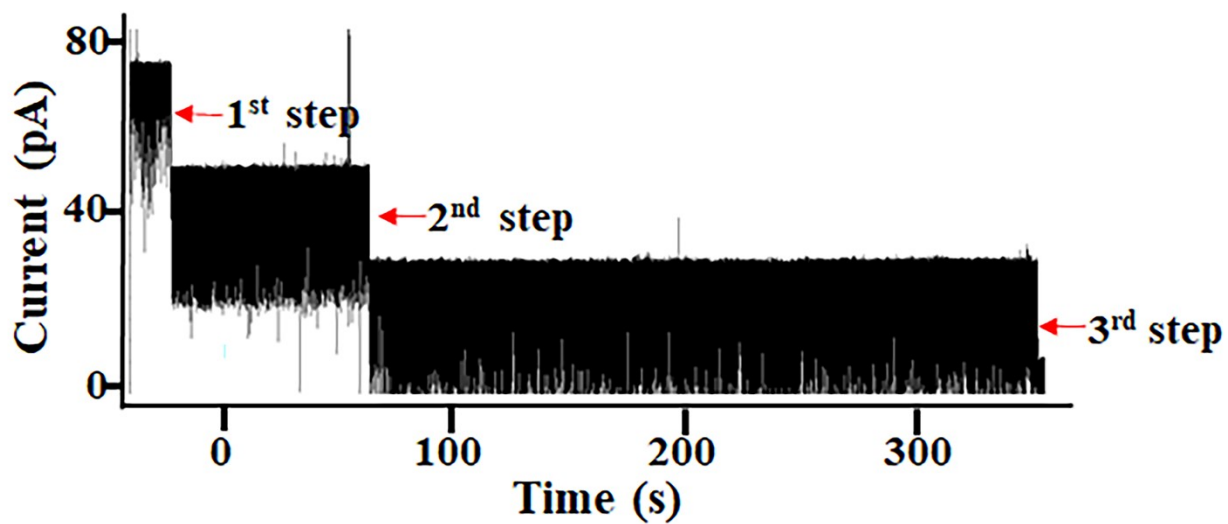


Figure S7: Three-step gating due to the addition of uPAR protein to N-HisGP10-C-uPAR channel.

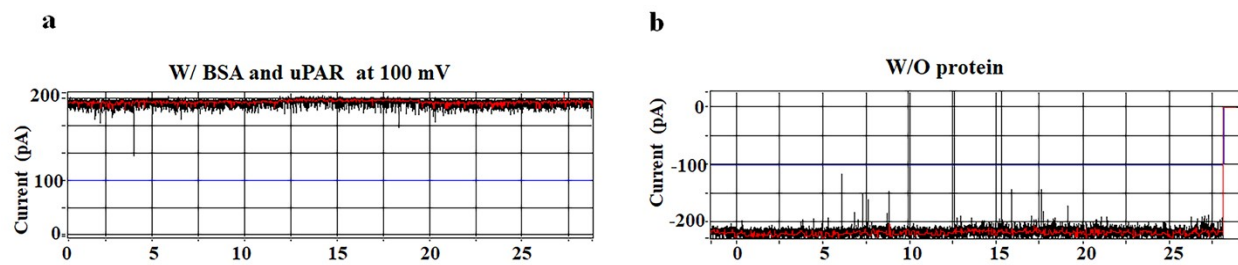


Figure S8: (a) The current trace result of the N-His GP10-C-uPAR channel with BSA and uPAR protein at 100 mV. (b) The current trace result of the N-His GP10-C-uPAR channel without protein at -100 mV.