Supporting Information

Simultaneous detection of multiple proteases using a non-array nanopore platform

Xiaohan Chen, Youwen Zhang, and Xiyun Guan*

Department of Chemistry, Illinois Institute of Technology, 3101 S Dearborn St, Chicago, IL 60616, USA

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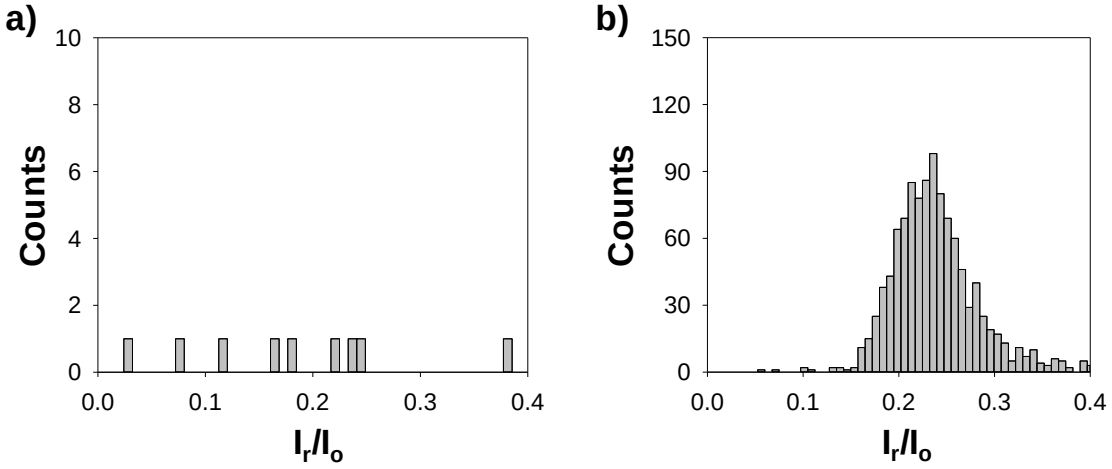

Figure S1. Amplitude histograms of peptides (a) ARL and (b) RLAQAVRSSSARLVFFKPLGL. The experiments were performed at + 120 mV in 3 M (*cis*) / 0.5 M (*trans*) NaCl with a wild type α-hemolysin protein nanopore. Both the concentrations of peptides ARL and RLAQAVRSSSARLVFFKPLGL used were 2.5 µM.

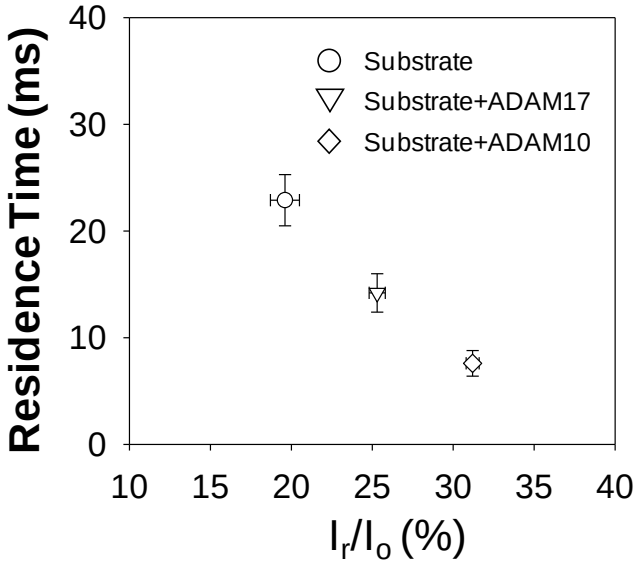

Figure S2. 2D plots of event residence time and residual current, showing the difference among the substrate, substrate digestion product by ADAM17, and substrate cleavage product by ADAM10. The experiment was performed at +120 mV in 3 M (*cis*) / 0.5 M (*trans*) NaCl with a wild type α-hemolysin protein nanopore.

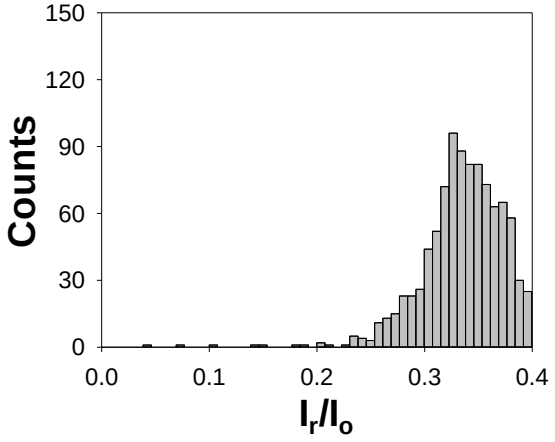

Figure S3. Amplitude histogram of peptide RSSSARLVFFKPLGL. The experiment was performed at +120 mV in 3 M (*cis*) / 0.5 M (*trans*) NaCl with a wild type α-hemolysin protein nanopore. The concentration of peptide RSSSARLVFFKPLGL used was 2.5 µM.

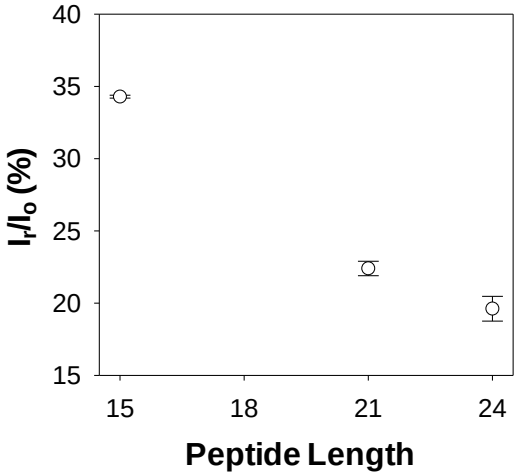

Figure S4. Effect of the peptide length on the mean residual current of peptide translocation events. The experiments were performed at +120 mV in 3 M (*cis*) /0.5 M (*trans*) NaCl with the wild type α-hemolysin protein nanopore. The concentrations of the peptides used were 2.5 μM each.

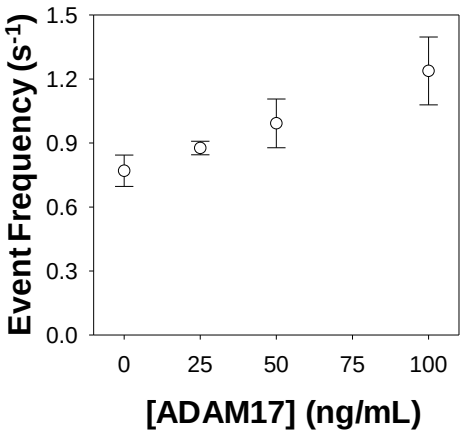

Figure S5. Plot of event frequency as a function of ADAM17 concentration. The experiments were performed at +120 mV in a salt gradient of 3 M (*cis*) / 0.5 M (*trans*) NaCl and in the presence of 2.5 μM peptide substrate and 175 ng/mL of ADAM10.

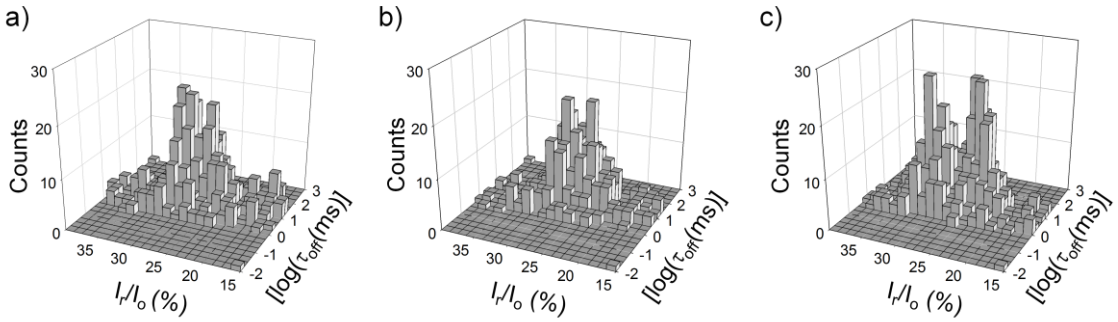

Figure S6. 3D plots of event counts vs. blockage amplitude vs. residence time, showing the simultaneous detection of ADAM10 and ADAM17. a) substrate (2.5 μM) + ADAM10 (175 ng/mL) + ADAM17 (25 ng/mL); b) substrate (2.5 μM) + ADAM10 (175 ng/mL) + ADAM17 (50 ng/mL); and c) substrate (2.5 μM) + ADAM10 (175 ng/mL) + ADAM17 (100 ng/mL). The experiments were performed at +120 mV in a salt gradient of 3 M (*cis*) / 0.5 M (*trans*) NaCl.