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Single molecule processivity and dynamics of cAMP-dependent protein kinase (PKA) PATRICK C. SIMS, YONGKI CHOI, CHENGJUN DONG, Department of Physics and Astronomy, University of California, Irvine, ISSA S. MOODY, Department of Molecular Biology and Biochemistry, University of California, Irvine, MARIAM IFTIKHAR, Department of Chemistry, University of California, Irvine, O. TOLGA GUL, Department of Physics and Astronomy, University of California, Irvine, GREGORY A. WEISS, Departments of Molecular Biology and Biochemistry, and Chemistry, University of California, Irvine, PHILIP G. COLLINS, Department of Physics and Astronomy, University of California, Irvine — Using single-walled carbon nanotube (SWNT) transistors, we monitored the processivity and dynamics of single molecules of cAMP-dependent protein kinase (PKA). As PKA enzymatically phosphorylates its peptide substrate, it generates an electronic signal in the transistor that can be monitored continuously and with 20 μs resolution. The electronic recording directly resolves substrate binding, ATP binding, and cooperative formation of PKA's catalytically functional, ternary complex. Statistical analysis of many events determines on- and off-rates for each of these events, as well as the full transistion probability matrix between them. Long duration monitoring further revealed minute-to-minute rate variability for a single molecule, and different mechanistic statistics for ATP binding than for substrate. The results depict a highly dynamic enzyme offering dramatic possibilities for regulated activity, an attribute that is useful for an enzyme that plays crucial roles in cell signaling.

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