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Following the Binding and Activity of Helicases with DNA Devices DIMITHREE KAHANDA, JASON SLINKER, Univ of Texas - Dallas, KEVIN DU PREZ, LI FAN, University of California, Riverside, UNIVERSITY OF DALLAS TEAM, UNIVERSIY OF CALIFORNIA, RIVERSIDE TEAM — Helicases are motor proteins that have the capability to separate two annealed strands of DNA or RNA. They are involved in many critical cellular processes such as DNA replication, transcription, translation, recombination and DNA repair. Much remains to be understood about the comparative roles of the 31 non-redundant DNA helicases. Here, we utilize probe-modified DNA monolayers on multiplexed gold electrodes as a sensitive recognition element of helicase binding. The electrochemical signals from these devices are highly sensitive to structural distortion of the DNA produced by the helicases. In this study we distinguished the details of three XPB helicases that belong to the Superfamily-2 of helicases. Clear changes in DNA melting temperature and duplex stability were observed upon helicase binding, shifts that could not be observed with the conventional UV-Visible absorption measurements. Binding dissociation constants were found in the range from 10-50 nM and correlated with observations of activity. ATP-stimulated DNA unwinding activity was also followed, showing exponential timescales and distinct time constants associated with conventional and molecular wrench modes of operation. These devices thus provide a sensitive measure of the structural thermodynamics and kinetics of helicase-DNA interactions.

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