

Label-Free Enzyme Therapeutic Effectiveness Quantification with Optimized Silicon Transistors

Son T. Le^{1,2}, Michelle A. Morris^{3,^}, Antonio Cardone^{4,5}, Nicholas B. Guros^{3,6,#}, Jeffery B. Klauda⁶, Brent A. Sperling⁷, Curt A. Richter¹, Harish C. Pant⁸, and Arvind Balijepalli^{3,*}

¹Nanoscale Device Characterization Division, National Institute of Standards and Technology, Gaithersburg, MD 20899, USA ; ²Theiss Research, La Jolla, CA 92037; ³Microsystems and Nanotechnology Division, National Institute of Standards and Technology, Gaithersburg, MD 20899, USA; ⁴Software and Systems Division, National Institute of Standards and Technology, Gaithersburg, MD 20899, USA; ⁵University of Maryland Institute for Advanced Computer Studies, University of Maryland, College Park, MD 20742, USA; ⁶Department of Chemical and Biomolecular Engineering, University of Maryland, College Park, MD 20742, USA; ⁷Chemical Sciences Division, National Institute of Standards and Technology, Gaithersburg, MD 20899, USA; ⁸National Institute of Neurological Disorders and Stroke, National Institutes of Health, Bethesda, MD 20892, USA

*e-mail: arvind.balijepalli@nist.gov

[^]Present address: Virginia Polytechnic Institute and State University, Blacksburg, VA 24061

[#]Present address: Astra Zeneca, 950 Wind River Lane, Gaithersburg, MD 20878

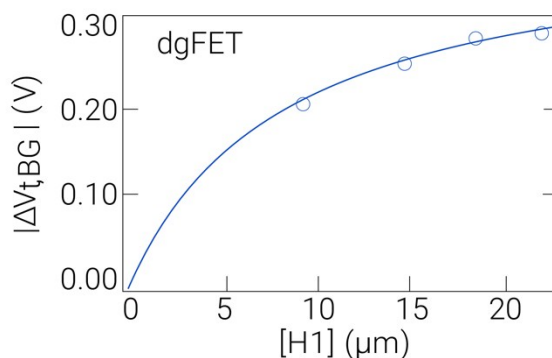


Figure S1: Steady-state measurements of Cdk5 activity using dual-gated field-effect transistors (dgFETs).¹ The change in the solution pH was used as a reporter to detect and quantify enzyme-mediated phosphorylation of the protein histone H1. The change in the back-gate threshold ($V_{T,BG}$) as a function of histone H1 concentration ($[H1]$) showed a monotonic increase. A simple model was used to estimate the activity coefficient, $k_a=(9.1\pm 0.9)$ μM .²

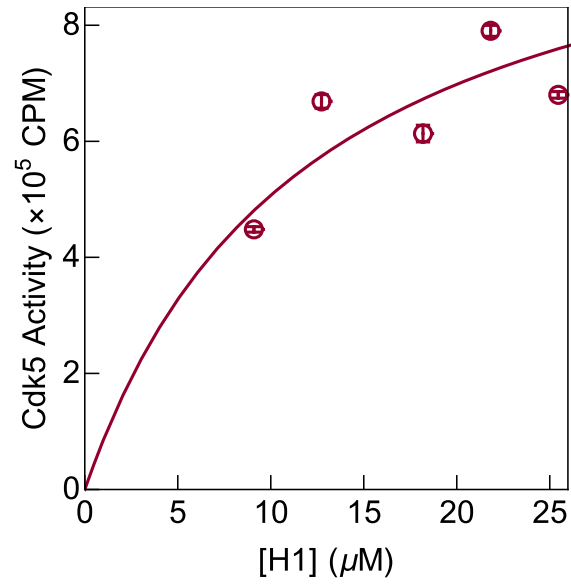


Figure S2: The activity of Cdk5/p25 was determined using a radioactively labeled adenosine triphosphate (γ - ^{32}P -ATP) assay as a function of the concentration of the protein histone ([H1]).³ A simple model was used to determine the activity coefficient, $k_a=(12.1\pm 2.3) \mu\text{M}$.² Adapted with permission from Ref. 2. The error bars represent the standard deviation in Cdk5 activity.

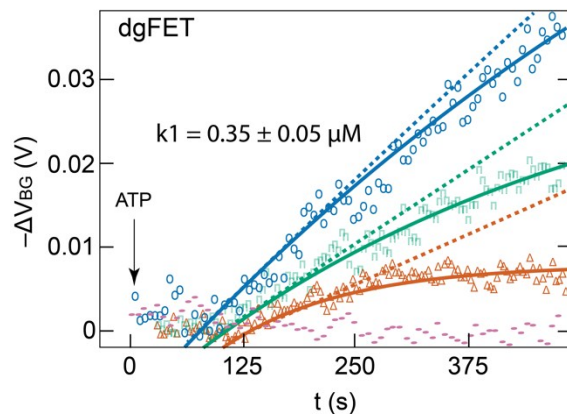


Figure S3: Time-series measurements of enzyme catalyzed phosphorylation of histone H1 were measured with a dual-gated field-effect transistor (dgFET). The histone concentrations ($[H1]$) were $9.1 \mu\text{M}$ (*orange*), $12.7 \mu\text{M}$ (*green*), $18.2 \mu\text{M}$ (*blue*) and a control sample with no histone (*pink*). The solid lines depict a first order kinetics model that describes the time course of phosphorylation, while the dashed lines represent an estimate of the reaction velocity during the first 100 s after a change in the signal was detected.

References

- 1 N. B. Guros, S. T. Le, S. Zhang, B. A. Sperling, J. B. Klauda, C. A. Richter and A. Balijepalli, *ACS Appl. Mater. Interfaces*, 2019, **11**, 16683–16692.
- 2 S. T. Le, N. B. Guros, R. C. Bruce, A. Cardone, N. D. Amin, S. Zhang, J. B. Klauda, H. C. Pant, C. A. Richter and A. Balijepalli, *Nanoscale*, 2019, **11**, 15622–15632.
- 3 B. K. Binukumar, Y.-L. Zheng, V. Shukla, N. D. Amin, P. Grant and H. C. Pant, *J. Alzheimers Dis.*, 2014, **39**, 899–909.