Supercoiling as a regulator: the role of biophysical feedback in transcription and chromatin dynamics

by

Christopher P. Johnstone

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ABSTRACT

Transcription induces a wave of DNA supercoiling, altering the binding affinity of RNA polymerases and reshaping the biochemical landscape of gene regulation. Instead of transcription being a simply biochemical progress acting over a static energy landscape, this supercoiling diffuses outward, dynamically reshaping the regulation of proximal genes and forming a complex feedback loop. While supercoiling is well studied as a biophysics problem, a theoretical framework is needed to integrate biophysical regulation with biochemical transcriptional regulation. To investigate the role of supercoiling-mediated feedback within multi-gene systems, we model transcriptional regulation—and especially transcriptional initiation—under the influence of supercoiling-mediated polymerase dynamics, allowing us to identify patterns of expression that result from physical inter-gene coupling. Gene syntax—–the relative ordering and orientation of genes—–defines the expression profiles, variance, burst dynamics, and inter-gene correlation of two-gene systems. In turn, this can enhance or weaken biochemical regulation. Together, the modeling results suggest that supercoiling couples behavior between neighboring genes, providing a regulatory mechanism that tunes transcriptional variance in engineered gene networks and explains the behavior of co-localized native circuits.

With these predictions in hand, we use integrated reporter circuits in human cells and demonstrate the reciprocal effects of transcription and DNA supercoiling: supercoiling-mediated biophysical feedback. This feedback effect regulates expression of adjacent genes in a syntax-specific manner, as predicted. Using Region Capture Micro-C and GapRUN, two recently published genomics techniques, we measure induction-dependent formation of supercoiled plectonemes and syntax-specific chromatin structures in human induced pluripotent stem cells. This first demonstration of supercoiling measurements in synthetic, integrated circuits in mammalian cells gives us unprecedented detail into the regulation of two-gene circuits. Applying syntax as a design parameter, we built compact gene circuits, tuning the mean, variance, and stoichiometries of expression across diverse delivery methods and cell types and improve both the titer of antibody production circuits and the efficacy of all-in-one inducible lentiviral circuits. Integrating supercoiling-mediated feedback into models of gene regulation will expand our understanding of native systems and enhance the design of synthetic gene circuits.

Thesis supervisor: Kate E. Galloway, PhD
Title: W. M. Keck Career Development Professor in Biomedical Engineering
Assistant Professor of Chemical Engineering