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Ecole Polytechnique *Amphithéâtre Curie, Ecole Polytechnique*

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Deciphering Bacterial Chromosome Organization: Investigating the Activity of MukBEF Bacterial Condensin

Chromosomes are long polymers of DNA that, when unraveled, far exceed the size of the cells containing them. Cellular mechanisms are thus at play to facilitate their integration into the cell while keeping the DNA accessible to multiple cellular processes such as replication, segregation, transcription, and repair. One of the main actors in this chromosome biology is the SMC (Structural Maintenance of Chromosomes) complex. These highly conserved and universally present proteins play an essential role and are involved in various aspects of chromosome management.

In *Escherichia coli*, employing a combination of "chromosomal conformation capture" approaches enable us to directly observe the activity of the SMC complex on the chromosome. Specifically, the MukBEF SMC complex, crucial for *E. coli*'s rapid growth, precise chromosome segregation, and positioning, instigates an augmentation of long-range DNA contacts, a phenomenon we can systematically monitor. Through a diverse array of *in vivo* assays, we clarify the mechanisms by which MukBEF governs chromosome conformation, and discern how the MatP/*matS* system imposes constraints on MukBEF activity within specific chromosomal domains.

Our results indicate that the loading of MukBEF occurs preferentially on newly replicated DNA, at multiple loci on the chromosome where it can promote long-range contacts in cis, even though MukBEF can also promote long-range contacts in the absence of replication. Through Hi-C and ChIP-seq analyses in strains with rearranged chromosomes, the inhibition of MukBEF activity increases with the number of *matS* sites, and this effect is likely a result of the unloading of MukBEF by MatP. Altogether, our findings shed light on the operational principles governing MukBEF's role in chromosome folding and segregation in *E. coli*.