**MS09 Structural Biology combining methods/High resolution**

MS9-02

Mechanistic basis for environment-controlled gene silencing by the histone-like nucleoid-structuring (H-NS) protein

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Abstract

As an environment-dependent pleiotropic gene regulator in Gram-negative bacteria, the H-NS protein is crucial for adaptation and toxicity control of human pathogens such as *Salmonella*, *Vibrio cholerae* or enterohaemorrhagic *Escherichia coli.* Changes in temperature affect the capacity of H-NS to form multimers that condense DNA and restrict gene expression. However, the molecular mechanism through which H-NS senses temperature and other physiochemical parameters remains unclear and controversial. Combining structural (X-ray crystallography, NMR and SAXS), biophysical and computational analyses, we show that H-NS forms superhelical filaments in crystals and in solution through the joint action of N-terminal and central dimerization domains. The handedness and pitch of these filaments are in agreement with plectonemic DNA supercoils. The human body temperature promotes the unfolding of the central dimerization domain, breaking up H-NS multimers. This unfolding event enables an autoinhibitory compact H-NS conformation that blocks DNA binding. High-resolution proton-less NMR analyses reveal fuzzy electrostatic interactions as the driving force for self-inhibition. We use computational multiscale modelling and simulations to obtain detailed insights into the mechanism of H-NS filament's sensitivity to environmental changes. We then combined molecular simulations and biophysical and structural analysis to reveal, at an atomistic level, how the same general mechanism was adapted through specific amino acid substitutions to suit bacteria that target plants and insects. Collectively, our results reveal the molecular basis of a mechanism that allows bacteria to sense their environment and adapt to it.

References

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Composite figure showing H-NS states