Theory of protein induced DNA looping and its role in the control of transcription

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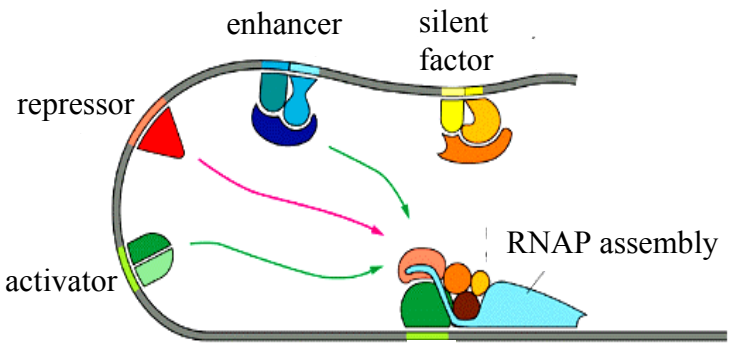
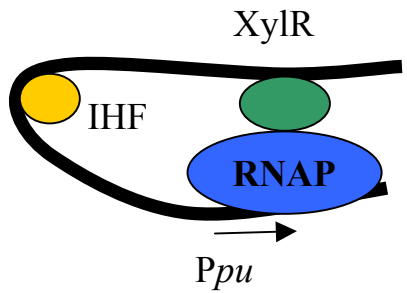
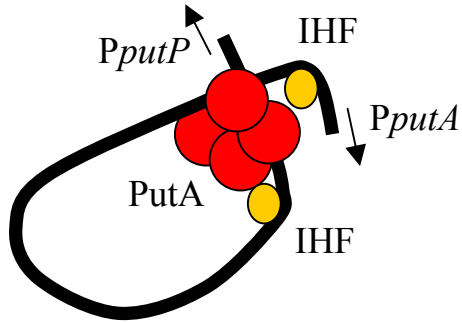
The functionality of molecular biological processes is determined the mechanics and dynamics of complex protein-DNA assemblies.

Transcription



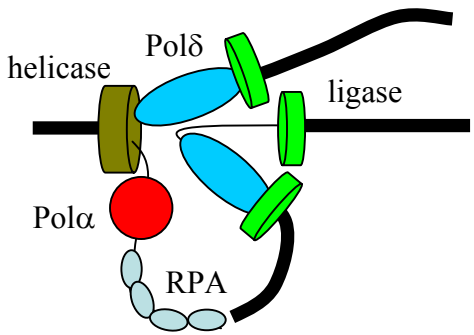


Activation

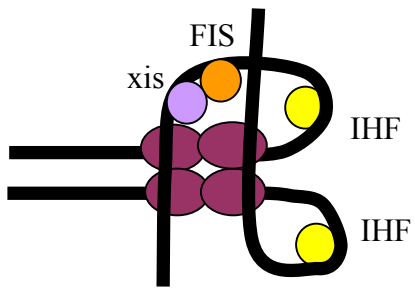


Repression

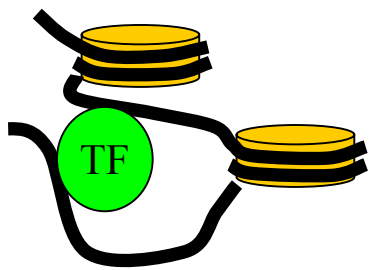
Replication



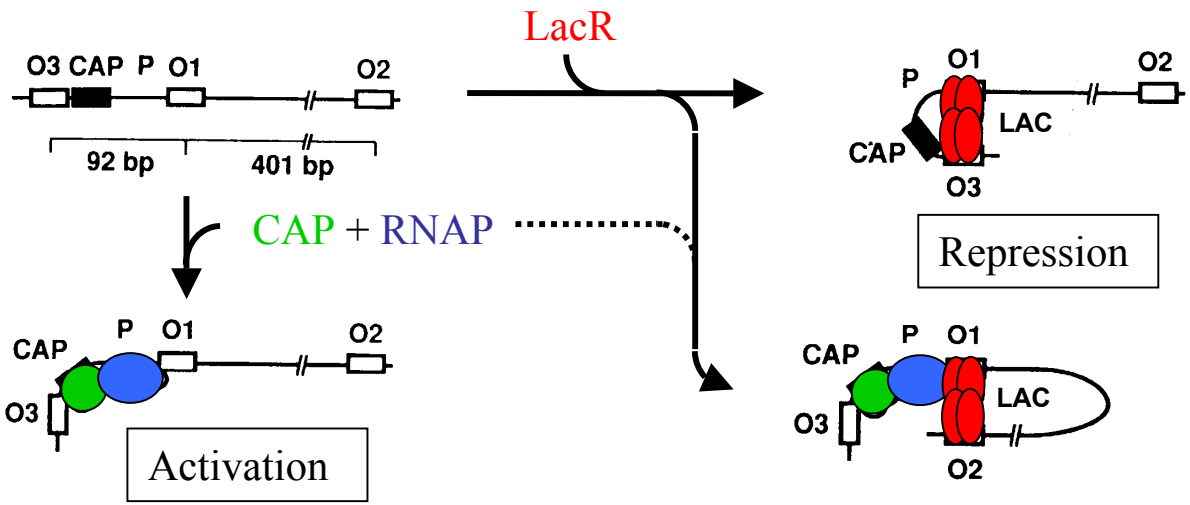
Recombination



Chromatin remodeling



Example: Regulation of the *Lac* operon

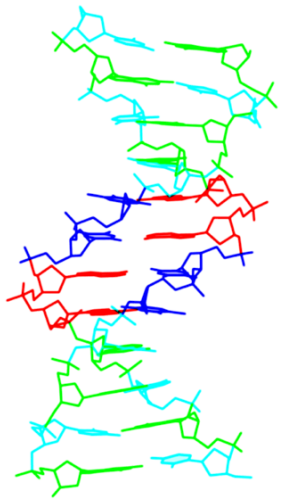


Lac repressor protein (LacR) binds to O1 and one of O2 and O3.
Absence of O2 and O3 decreases repression by 40 fold. [Oehler et al., *EMBO J.*, 1994]
Distance between O3/O2 and O1 affects repression 1-30 fold. [Müller et al., *J. Mol. Biol.*, 1996]

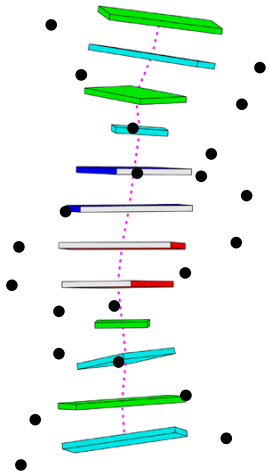
The presence of multiple LacR binding sites combined with mechanical deformation of DNA and LacR gives rise to a highly efficient and controllable repressive mechanism.

Modeling of the *Lac* operon

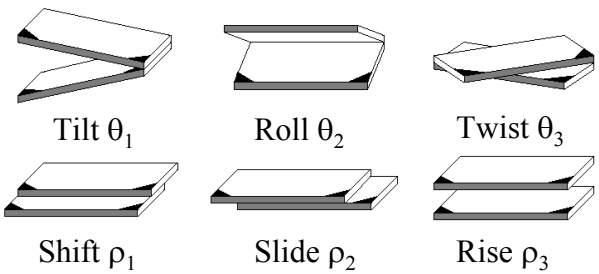
DNA



Atoms

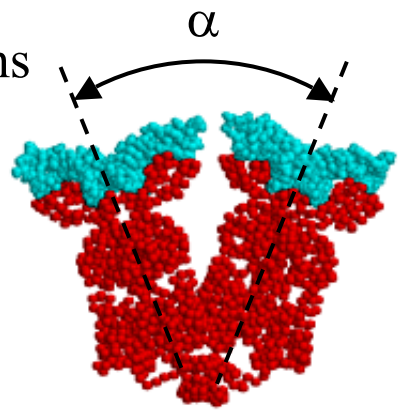


Base pairs

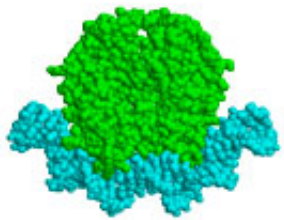


$$\Psi = \sum_{n=1}^{N} \psi^{n}(\theta_i^n, \rho_j^n) + \sum_{i<j} \frac{\delta_i \delta_j \, e^{-\kappa r_{ij}}}{4\pi\varepsilon \, r_{ij}}$$

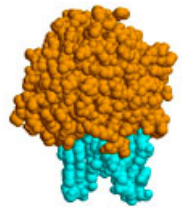
Proteins



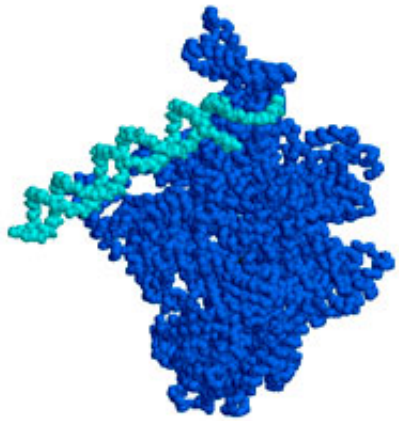
LacR (pdr026)



CAP (pdr023)



DNaseI (pde005)



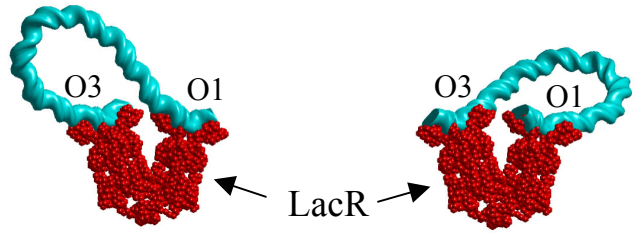
RNAP (pd0305)

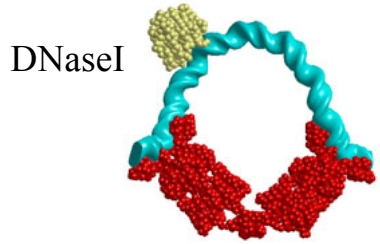
Computational techniques developed to date:

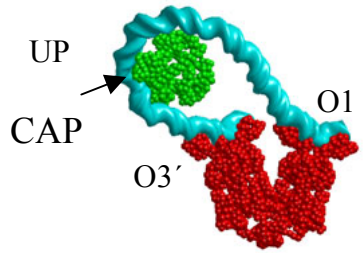
- calculation of DNA equilibrium configurations
  [Coleman, Olson, & Swigon, *J. Chem. Phys.*, to appear April 2003]
- MC algorithm for closure probability & free energy
- large-scale protein deformability, normal modes

New findings about the structure and dynamics of LacR-CAP-DNA assembly:

- DNase I footprinting reveals LacR extended in short loops
- Loop enhances repression by inhibiting CAP binding
- CAP binding allowed if O3 site is shifted upstream to O3´
- Cooperative effect between RNAP and LacR binding

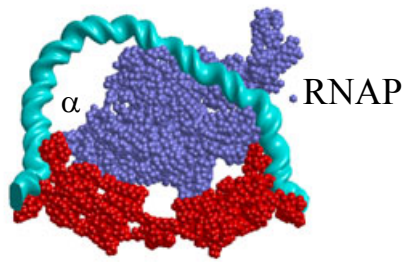






Work in progress:

- Dynamics of loop formation
- Kinetic model of Lac repression and CAP activation
- Modeling of prokaryotic operons Ara, NtrC, Put
- Experimental verification of structural results and mechanisms
- Design of transcription control elements



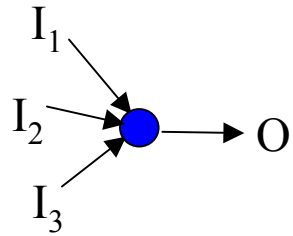
Future Directions

Use mechanical principles to bridge the gap between structural biology, macromolecular assembly, and biological function.

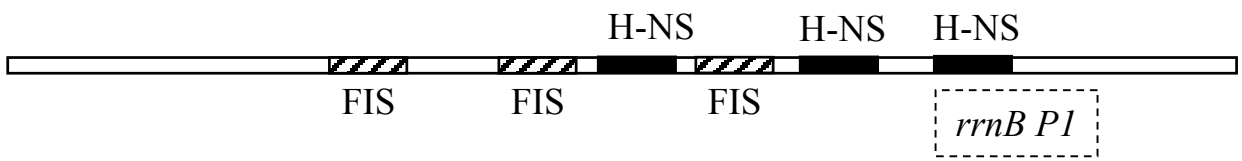
Molecular biomechanics: investigate the mechanics of transcription, replication, recombination; study the action of enzymes and molecular motors

Genetic networks: incorporate transcription-assembly mechanics in modeling the dynamics of genetic networks

$$[\mathrm{O}] = f\left([\mathrm{I}_1], [\mathrm{I}_2], [\mathrm{I}_3]\right)$$



Bioinformatics: apply conformational principles to the identification of promoters and transcription factor binding sites



Pharmaceutics: design drugs that alter DNA mechanics; apply to transcription regulation in gene therapy and development of antimicrobial agents