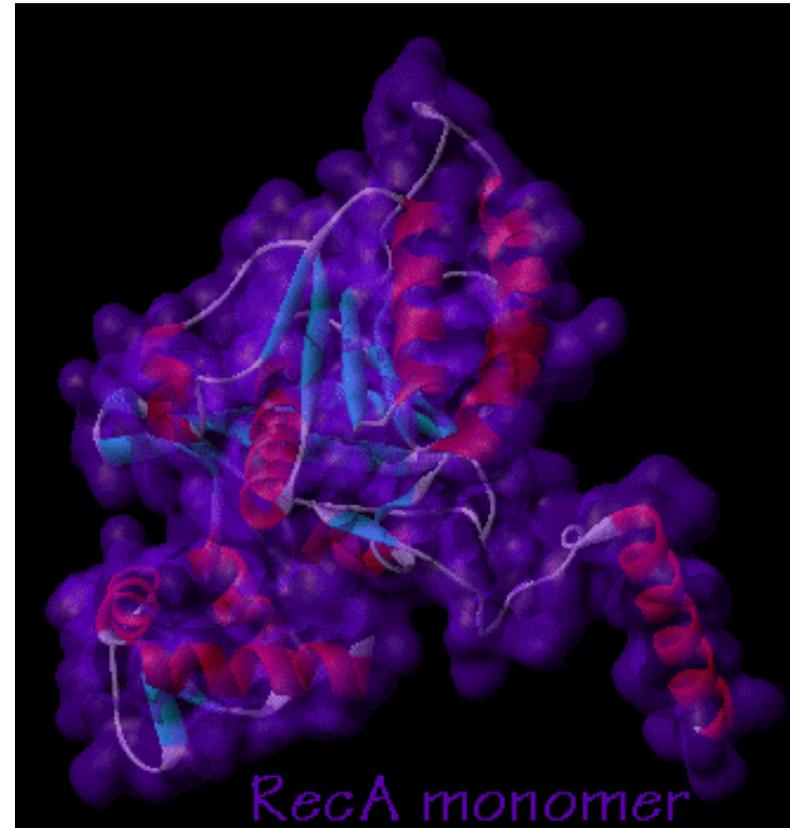


**Single-molecule FRET  
study  
on the filament  
dynamics of RecA**



<http://www.callutheran.edu/BioDev/omm/reca/recamast.htm>

## RecA is a protein

38 kDa

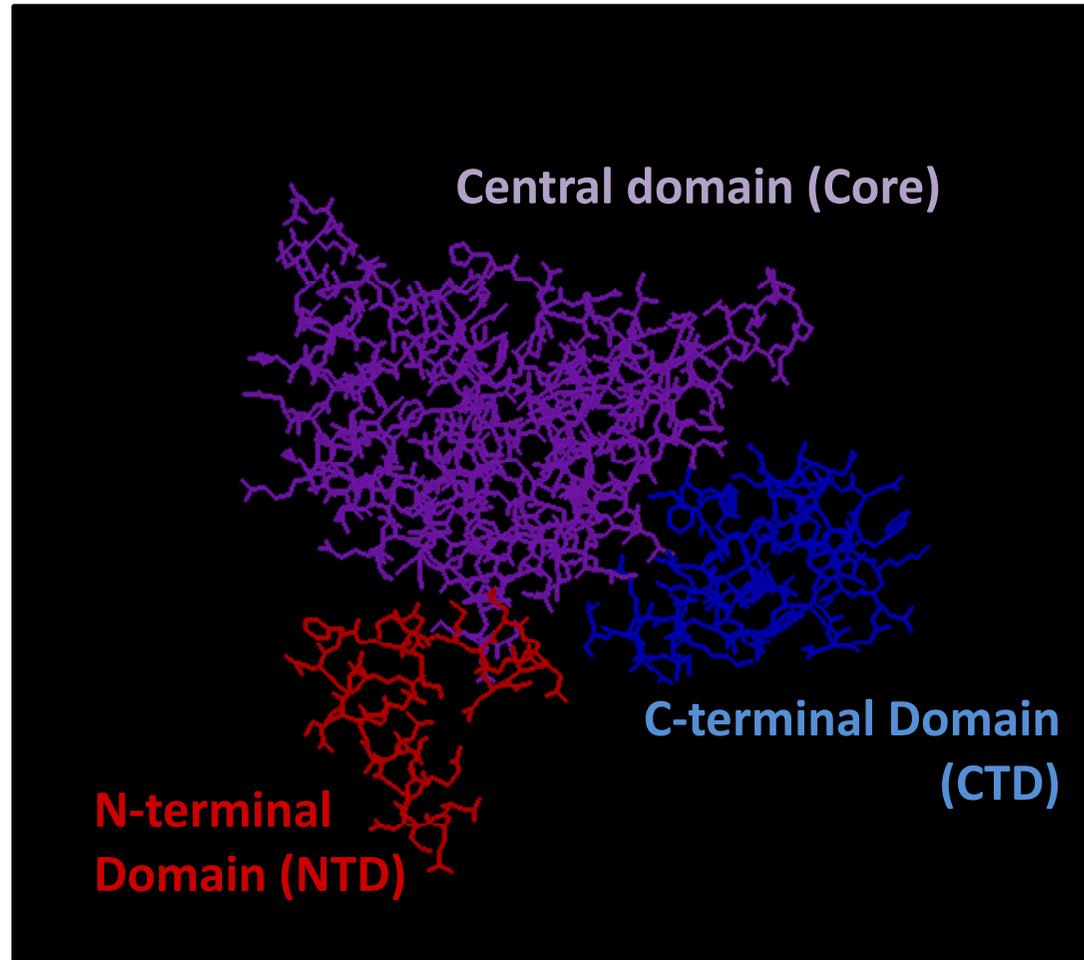
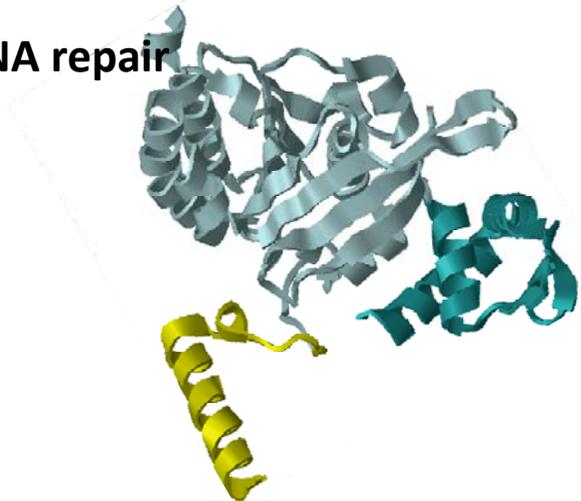
332 Residues

3 domains

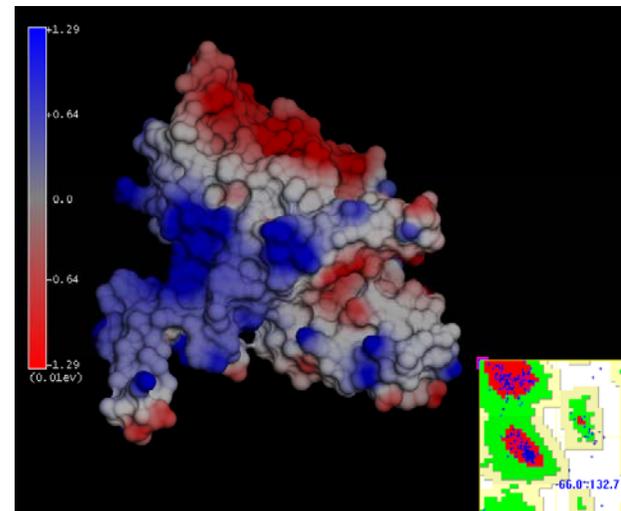
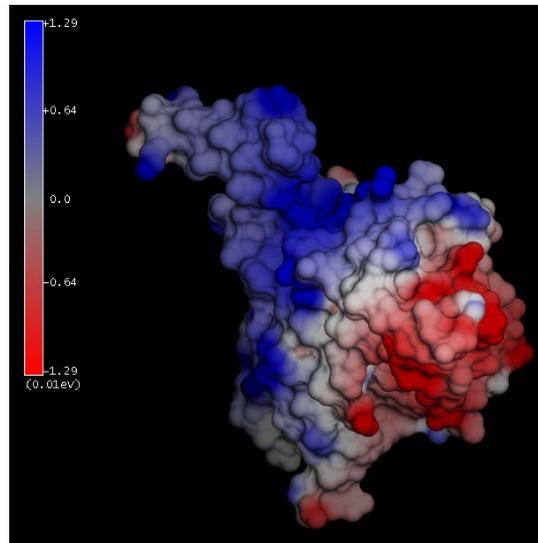
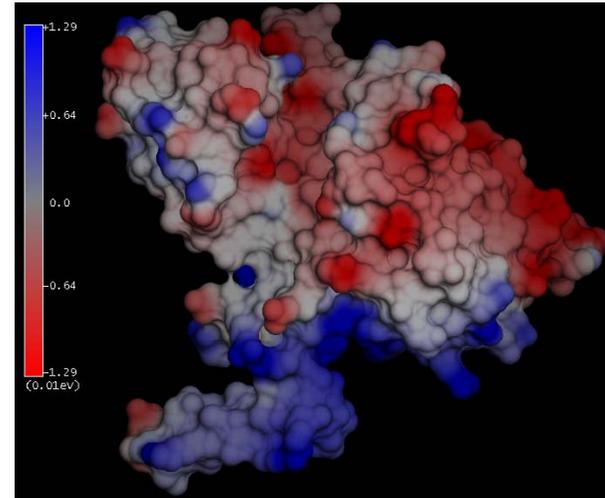
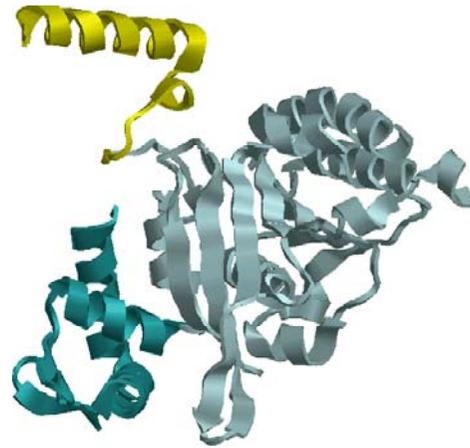
DNA binding activity

DNA dependent ATPase

DNA repair



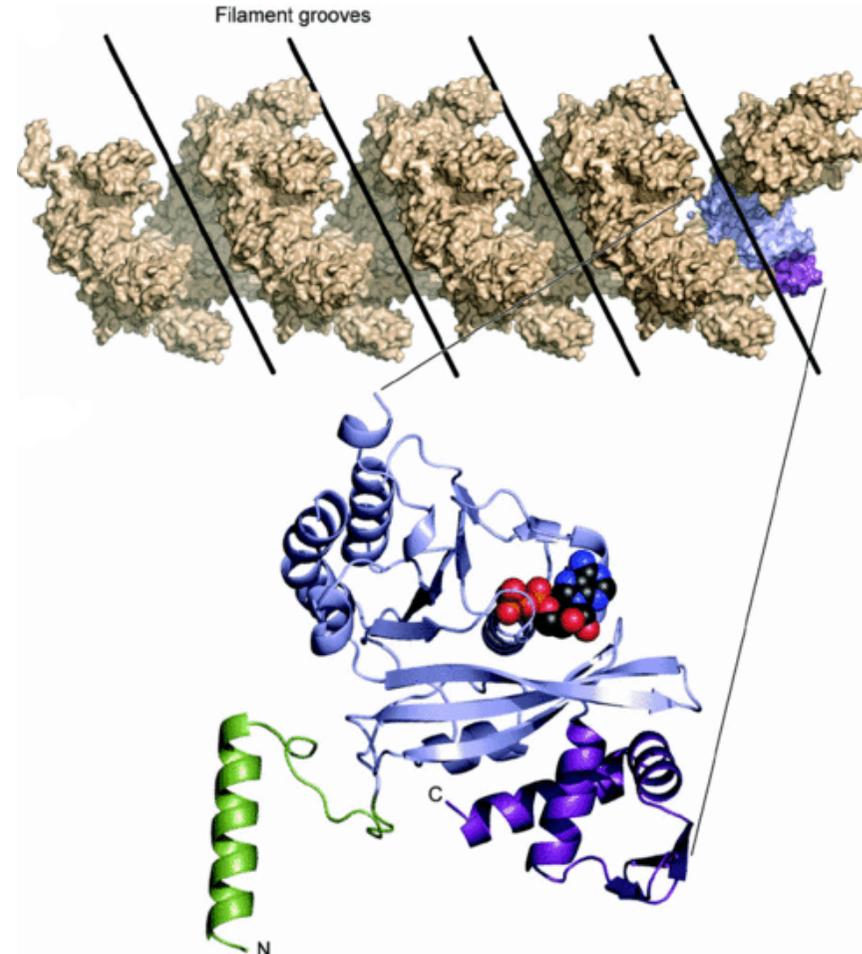
# Surface of RecA



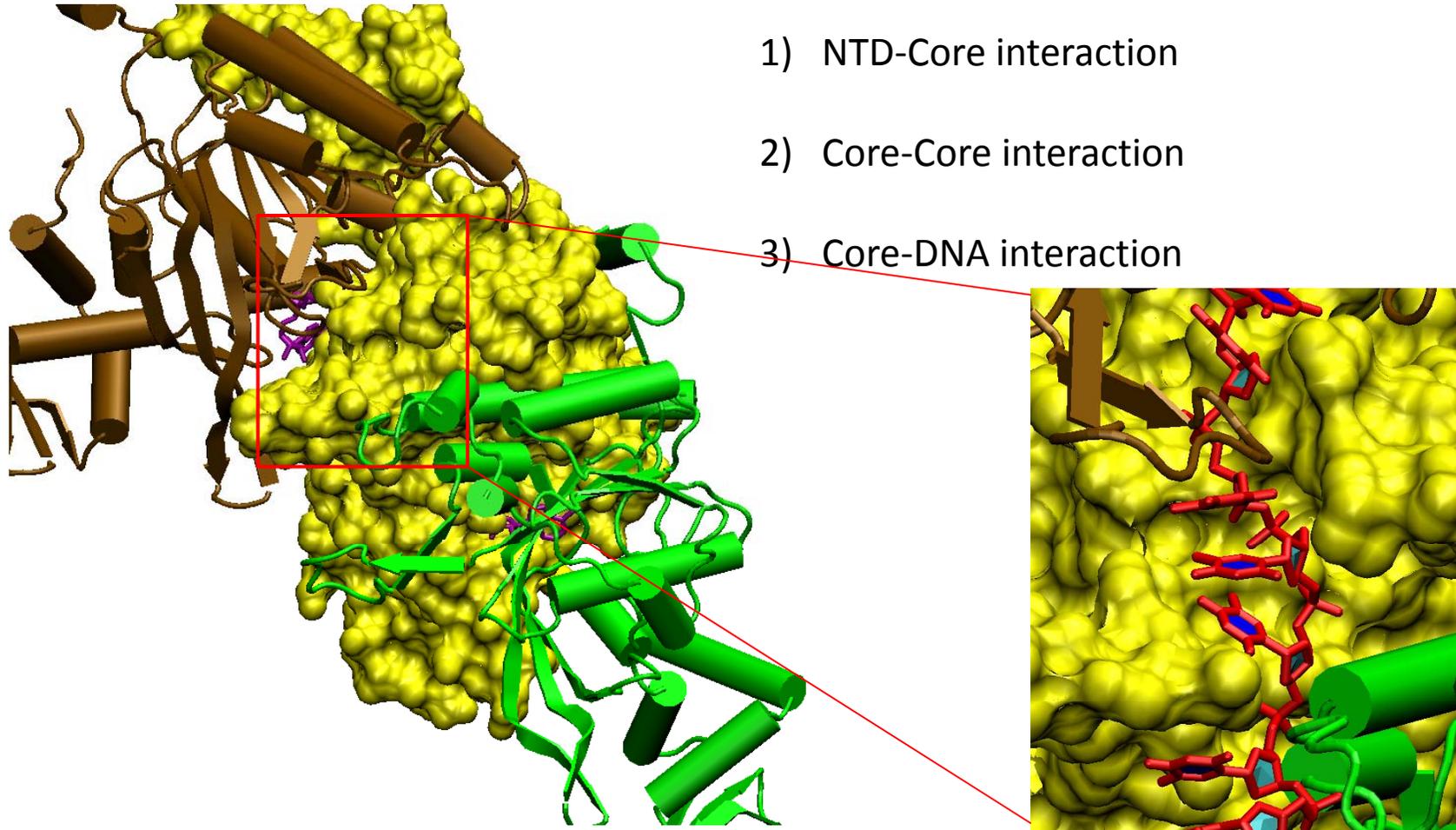
## RecA forms filament on a ssDNA

### RecA filaments

- Homologous Recombination
- Regulation of SOS response

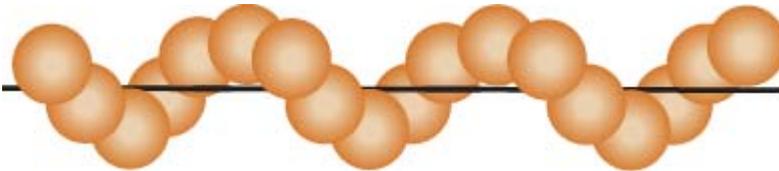
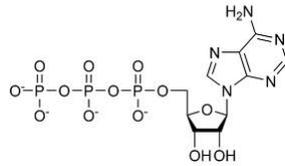


## RecA make a three major contacts with neighbors

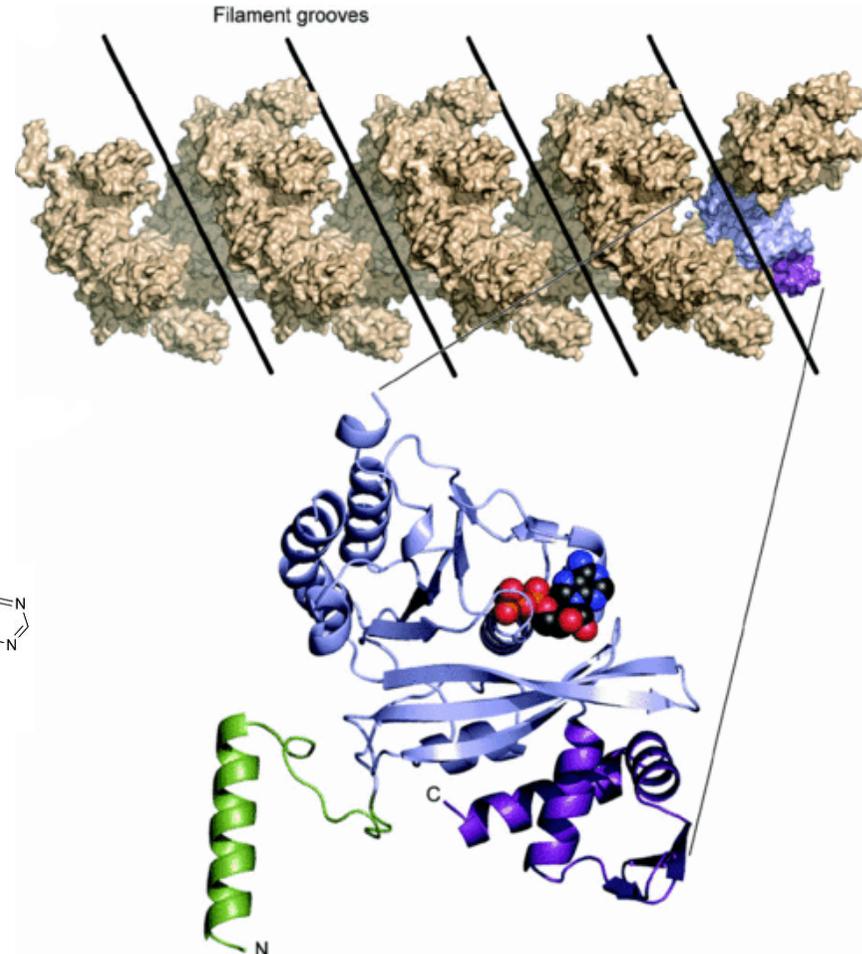
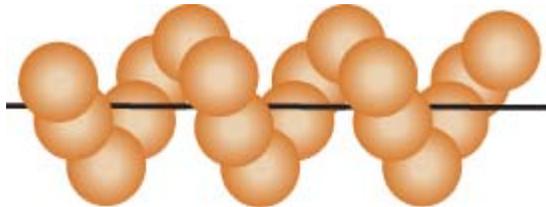
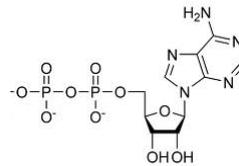


# Filament pitch length changes with the nucleotide cofactor

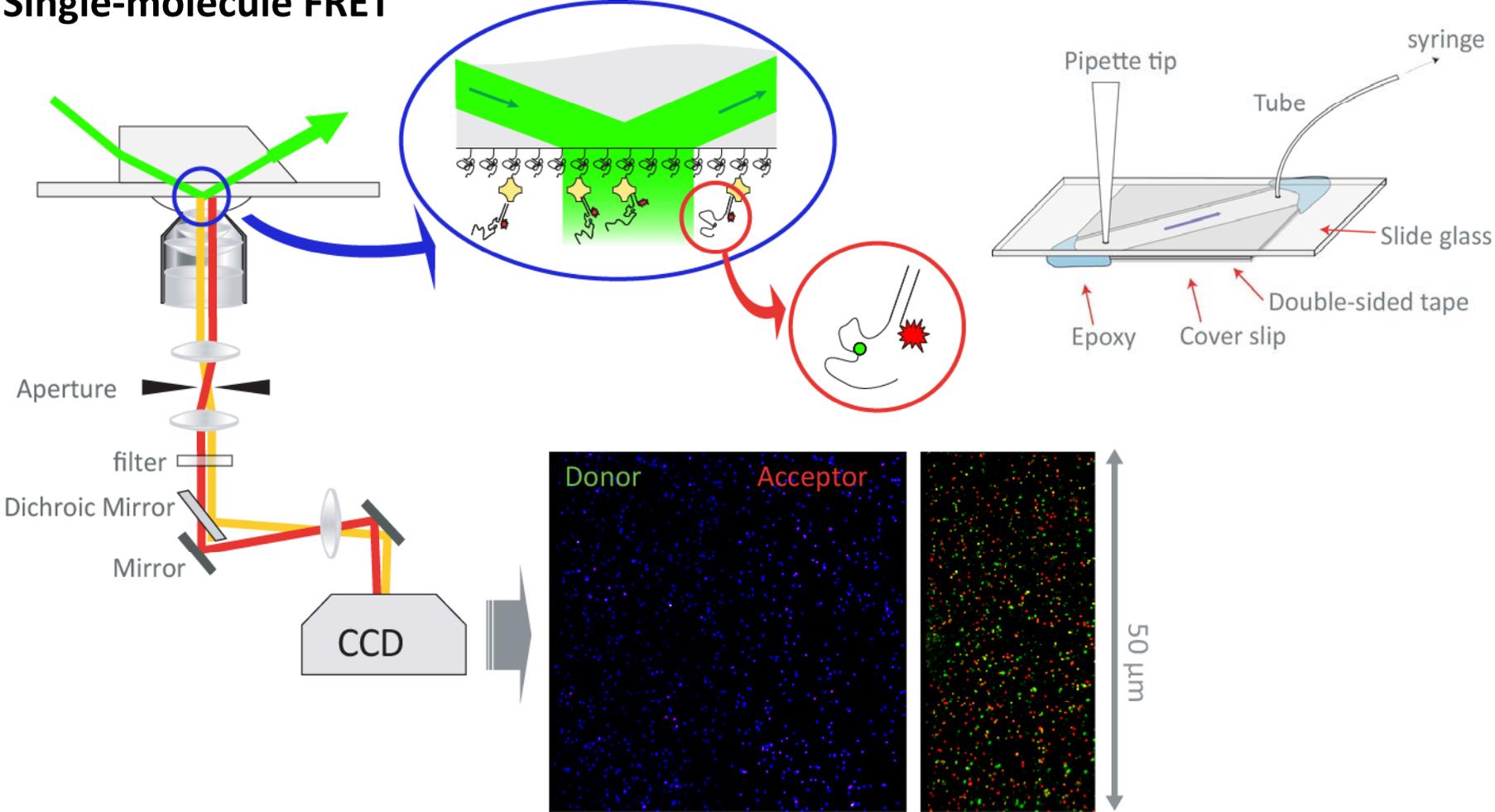
Extended (Active) form  
- ATP, ATP $\gamma$ S



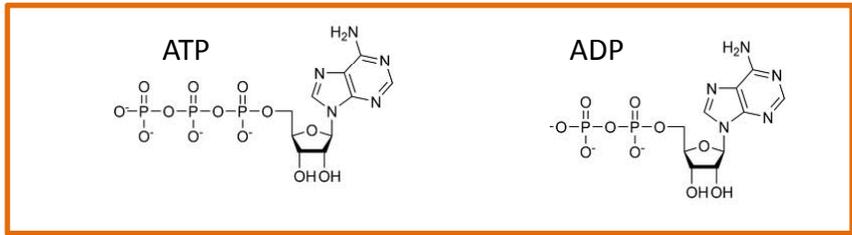
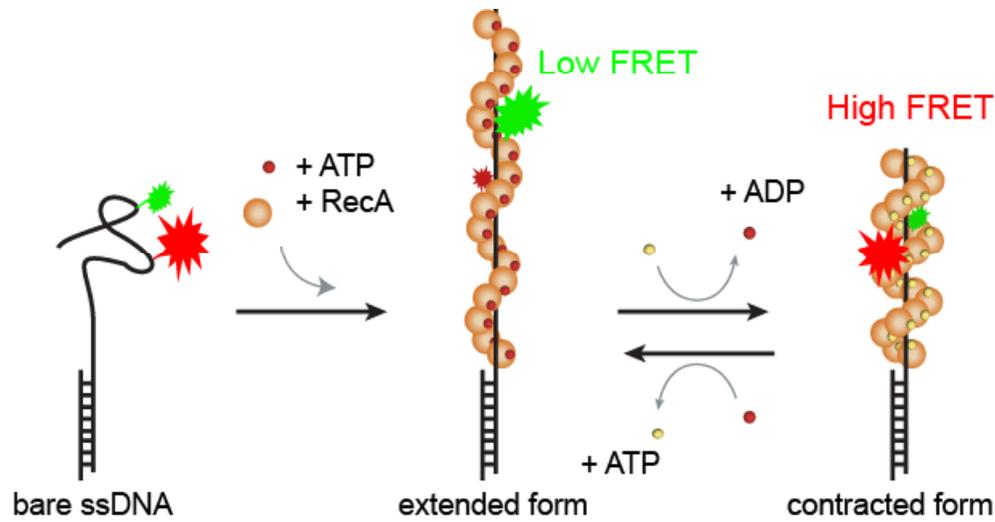
Contracted (Inactive) form  
- ADP, no cofactor



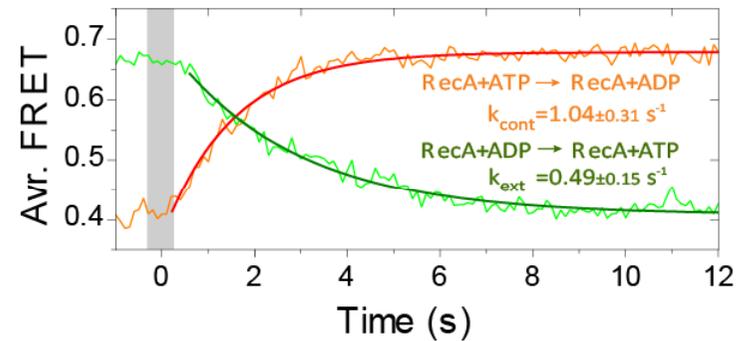
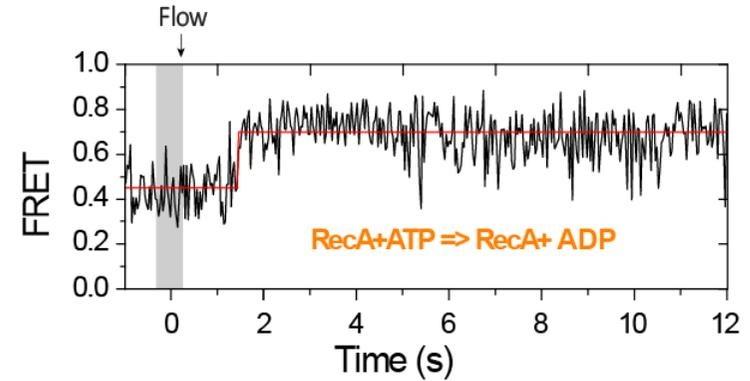
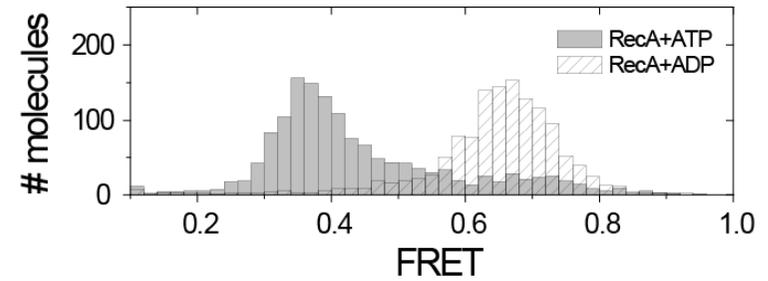
# Single-molecule FRET



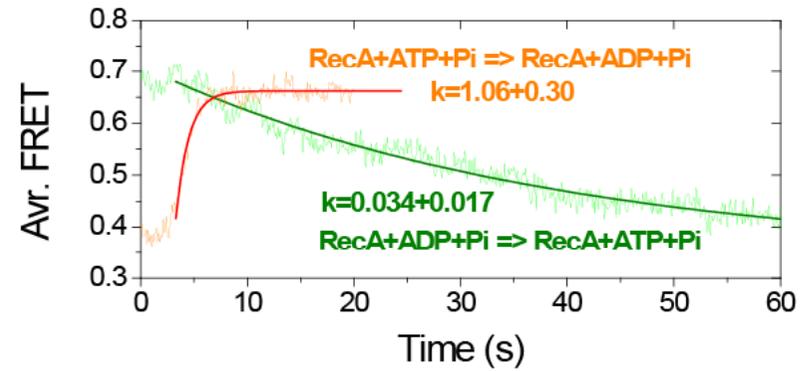
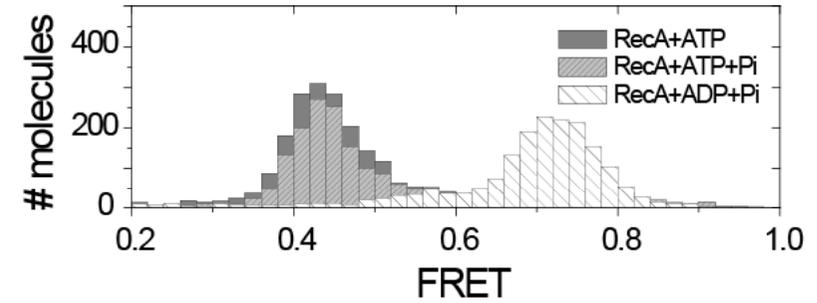
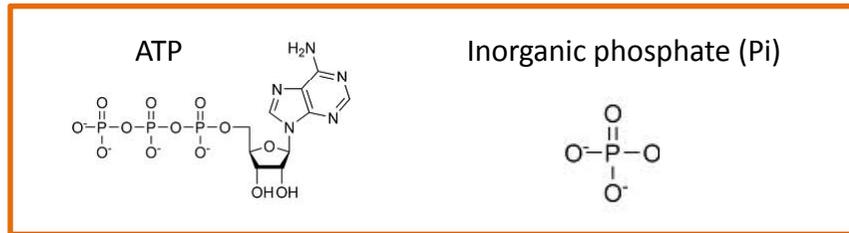
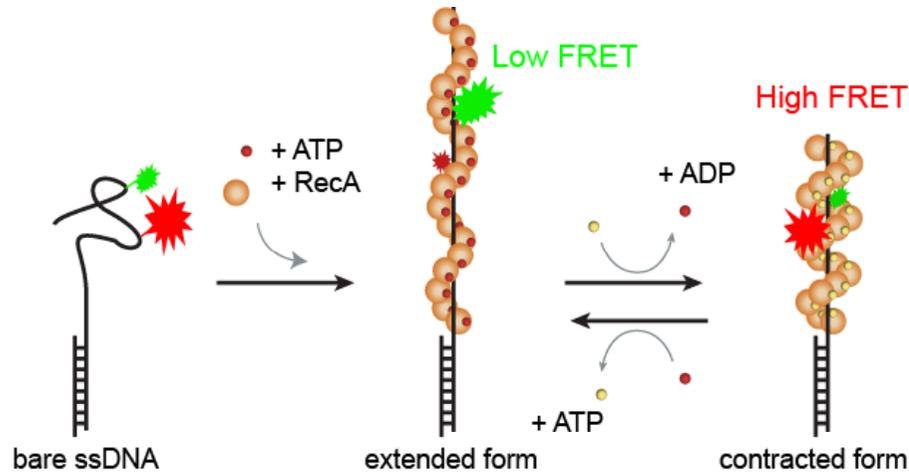
# Filament extension and contraction



- extension and contraction is cooperative
- energy barrier may exist



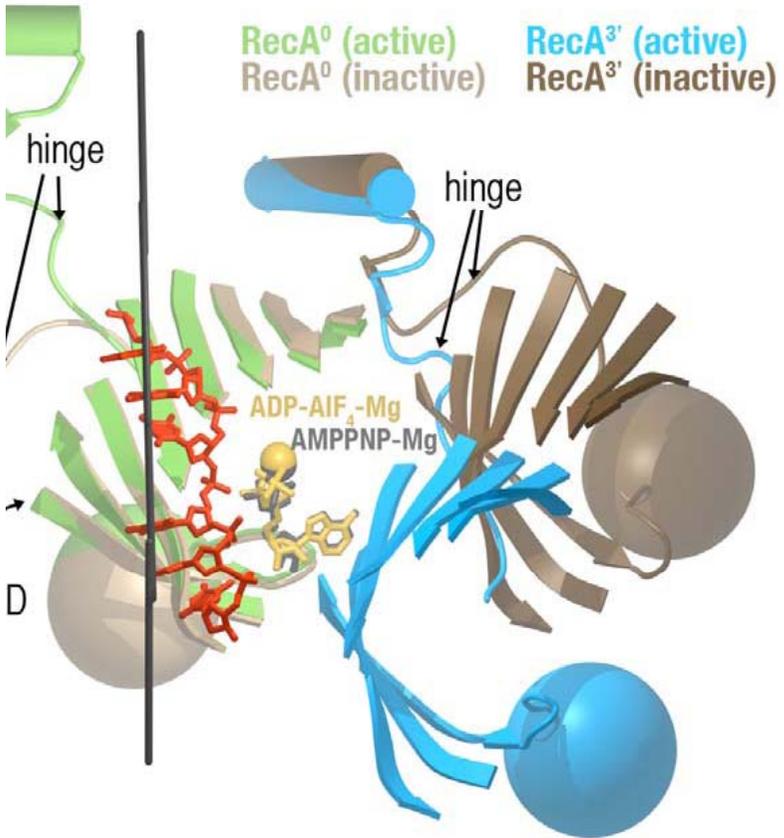
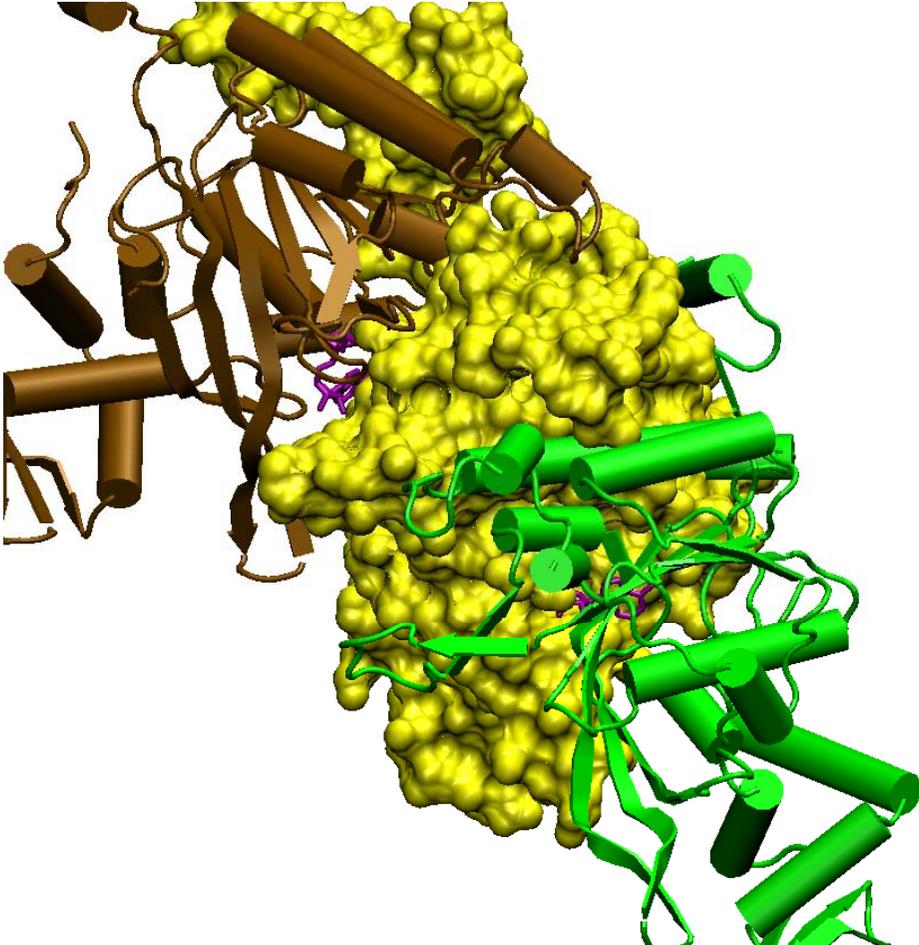
# The role of Pi



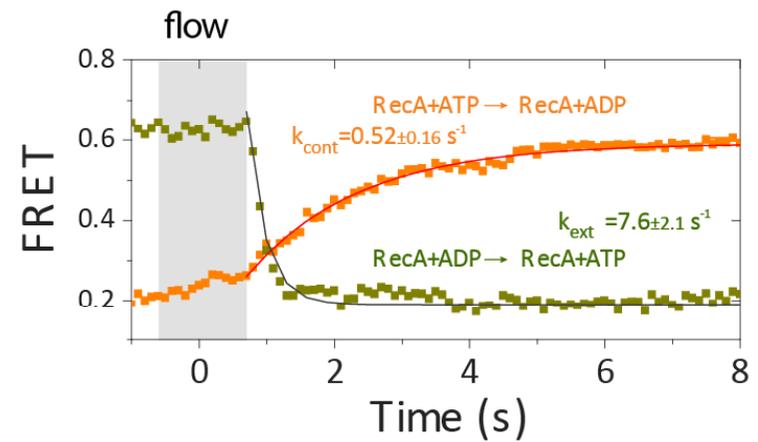
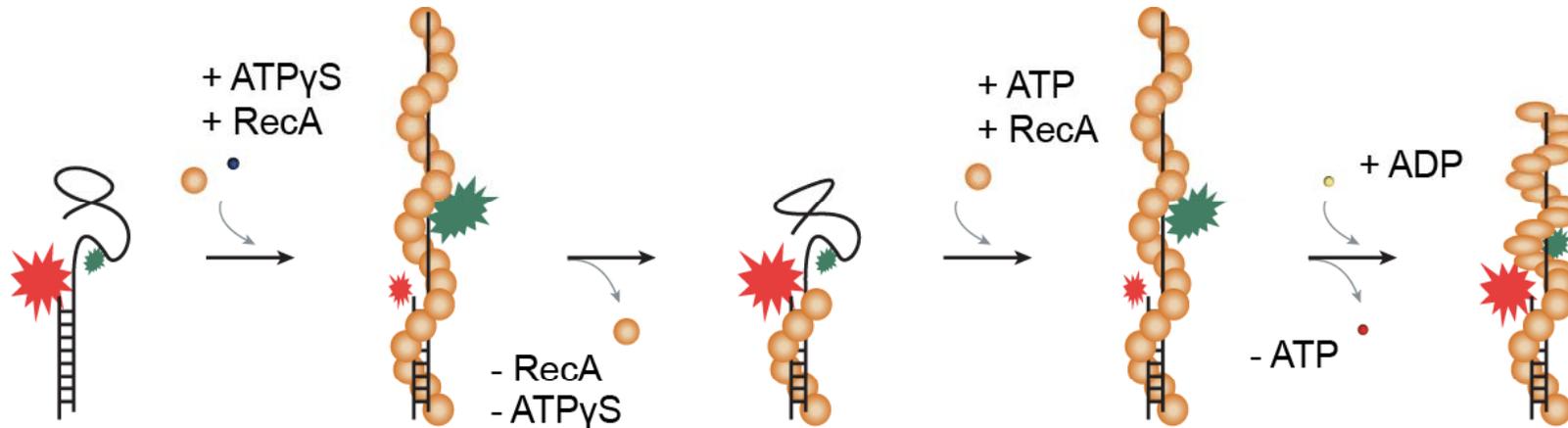
**Inorganic phosphate hinders ATP rebinding**

→ Pi may compete with ATP for the ATP binding pocket

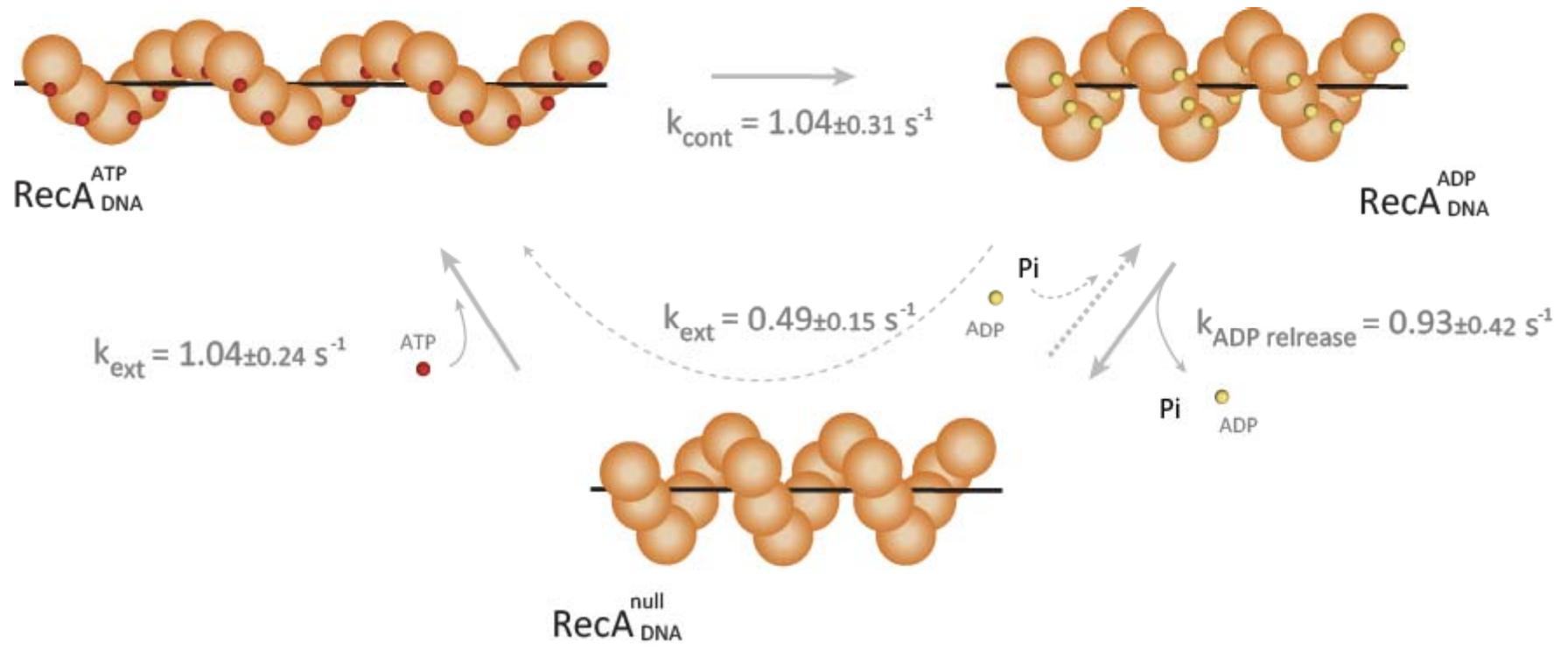
# Filament pitch length changes via subunit reorientation



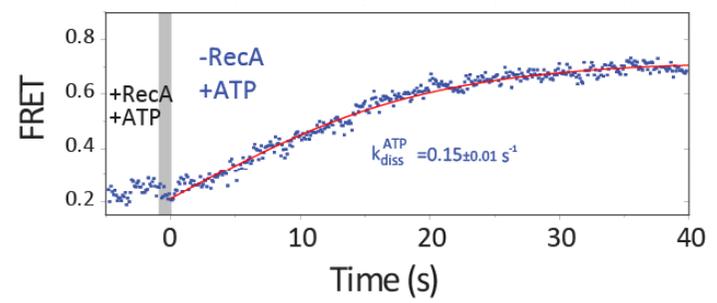
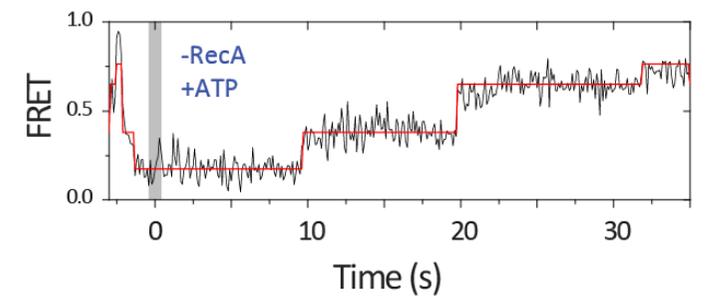
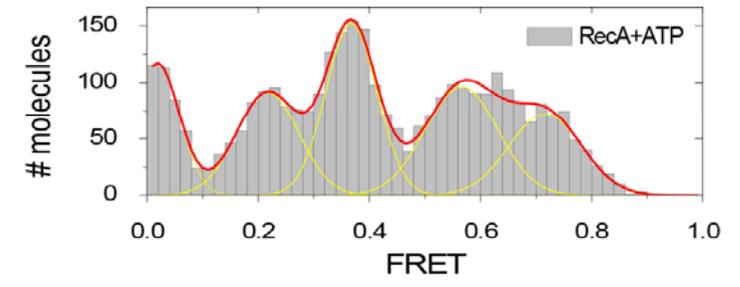
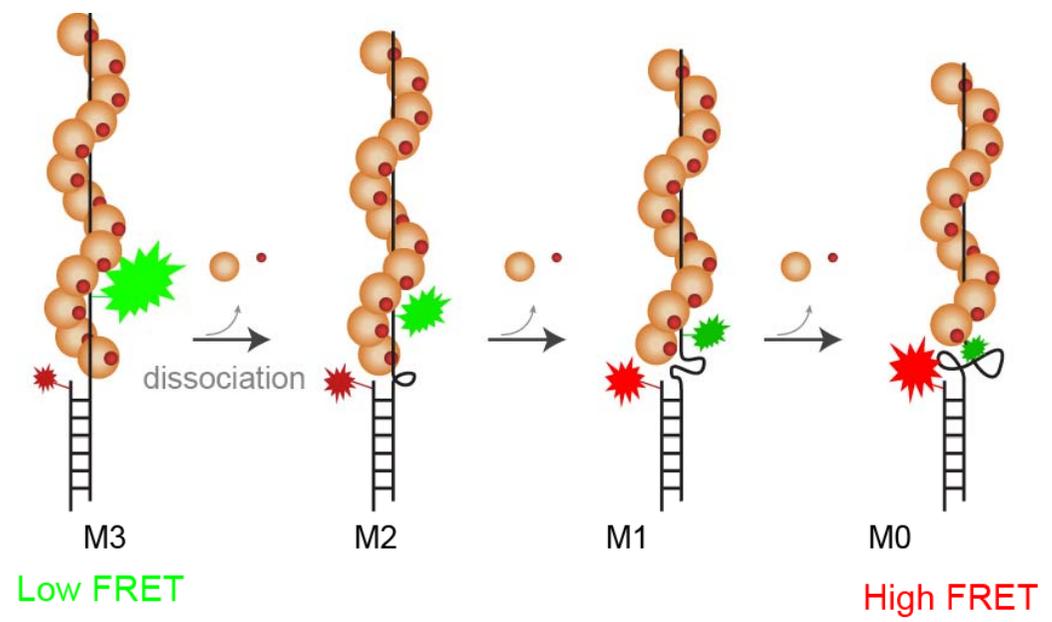
If neighbor is in extended form, extension process get fueled.



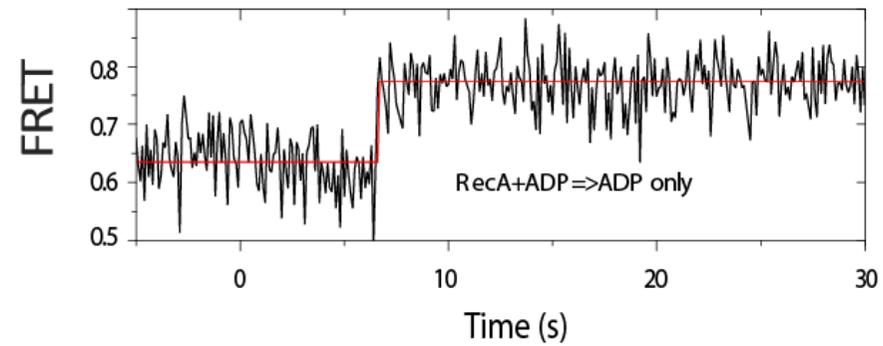
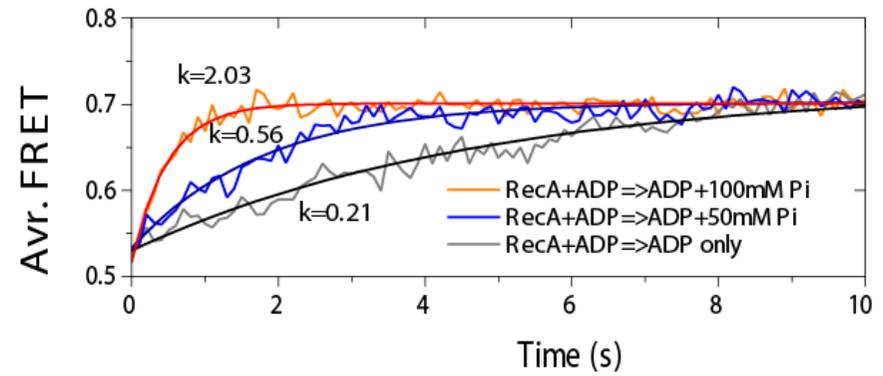
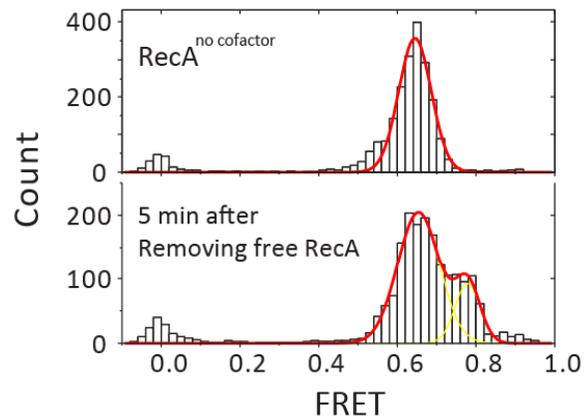
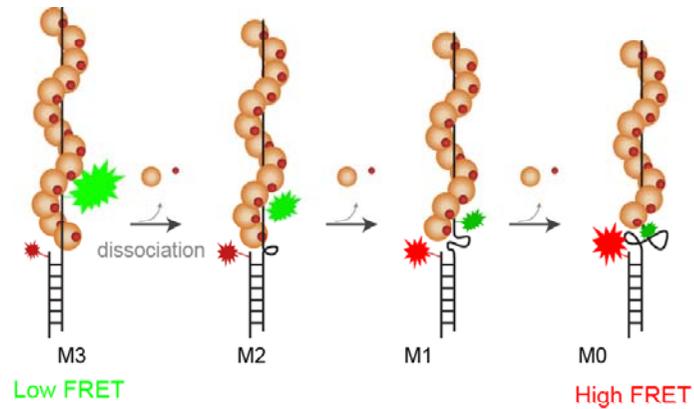
# Filament contraction and extension



# Dissociation of monomer

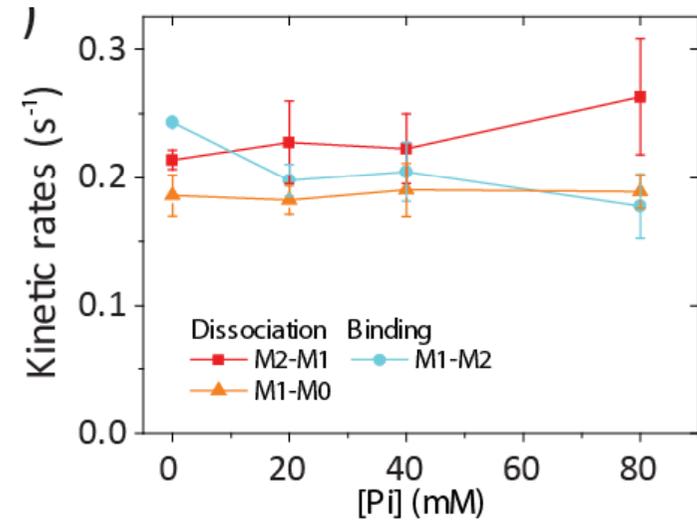
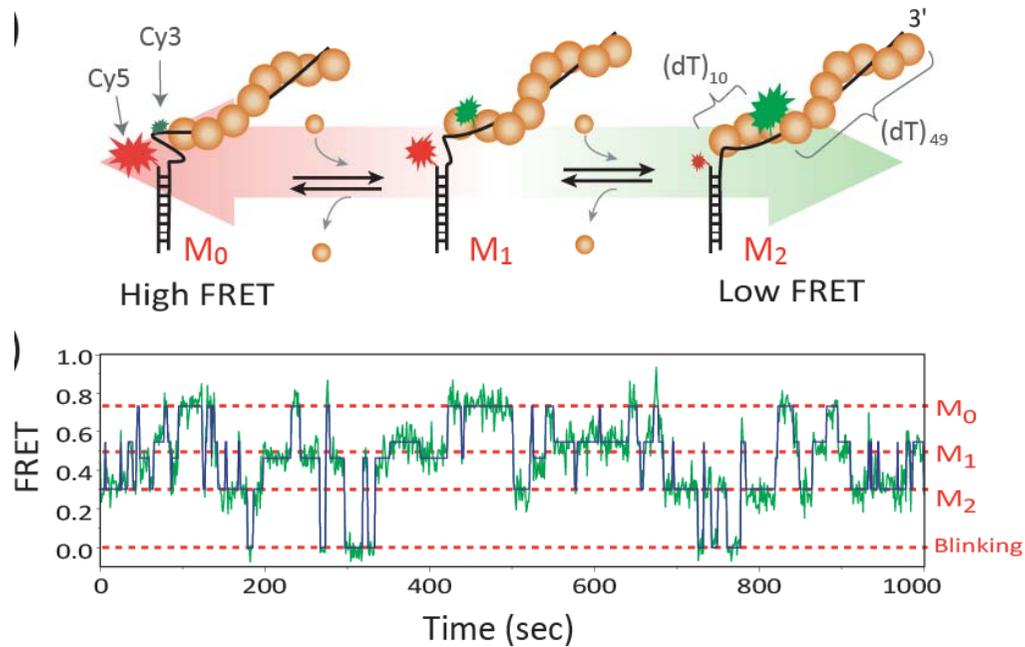


# Dissociation of inactive filaments



- ➔ ADP is required for dissociation
- ➔ Pi promotes dissociation

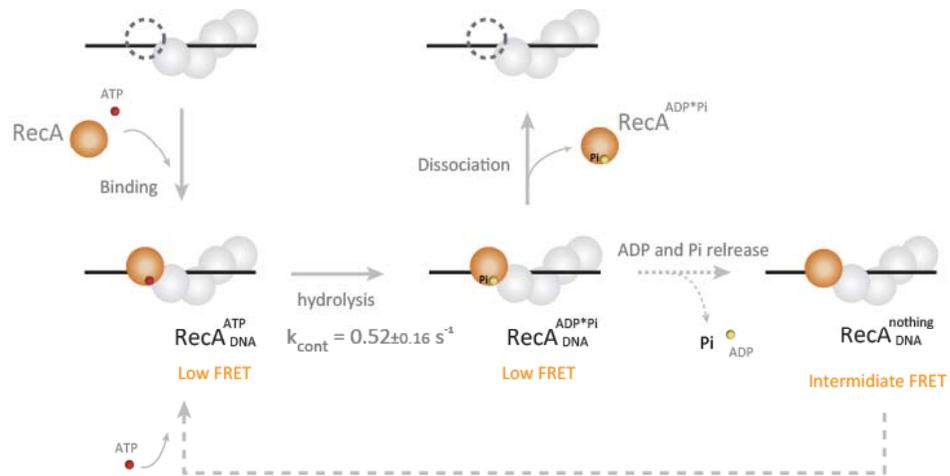
## Dissociation of active filaments



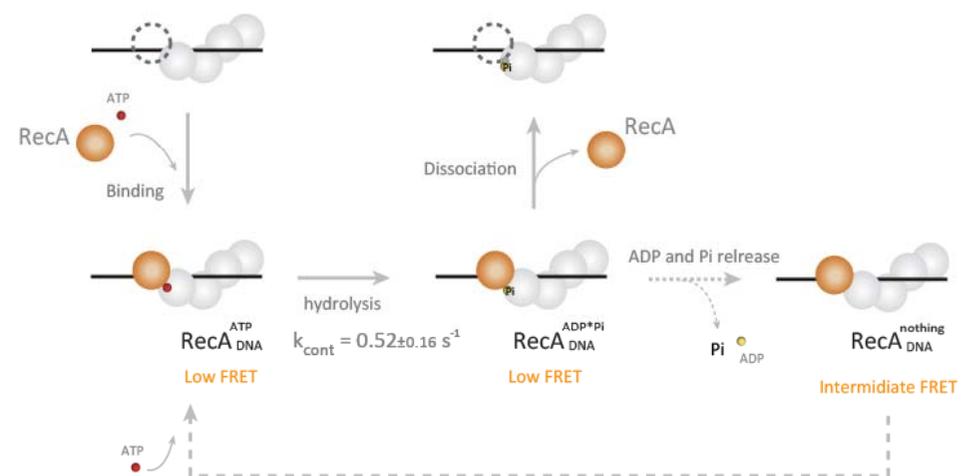
Dissociation of active filament is not affected by Pi

- 1) End monomer does have ADP and Pi when it dissociates
- 2) End monomer cannot have ADP and Pi

## Model 1



## Model 2



**Sorry for all these headaches.**

**And thank you for your attention.**

**Time to LUNCH!!!!**