Allosterically Regulated Structural Fluctuation and Microtubule-Binding Affinity of KIF1A

– A Simulation Study of Coarse-Grained Model –

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Motivation

Kinesin switches its microtuble (MT) binding affinity between strong binding mode and weak binding mode depending on nucleotide states. For understanding this allosteric regulation mechanism, we studied the structural fluctuations of the motor head of the single-headed kinesin KIF1A in different nucleotide states using computer simulations of coarse-grained Go-like model.

0′		I′		II′		III′		IV′		V′		0′
MK	⇌	MKT	⇌	MK*T	→	MKDP	→	MK*DP		MKD	⇌	MK
⇅		⇅		⇅		⇅		⇅		⇅		⇅
K	⇌	KT	⇌	K*T	→	KDP	→	K*DP	→	KD	⇌	K
0		**I**		**II**		**III**		**IV**		**V**		**0**

Figure: KIF1A-MT hydrolysis cycle
K:KIF1A, M:MT, T:ATP, D:ADP, DP:ADP·Pi
0', I', II',III':Strong-binding mode, IV',V':Weak-binding mode

Method

1. A coarse-grained C_α Go-like model for *each* nucleotide state.
2. Nucleotides are also represented by a few beads
3. Langevin dynamics simulations for investigating the structural fluctuations when MT is absent.

Reference states

	I	II	III	IV	V
binding mode	strong			weak	
nucleotide state	ATP-1	ATP-2	ADP·Pi-1	ADP·Pi-2	ADP
analog	AMP-PCP	AMP-PNP	ADP-AlFx	ADP-Vi	ADP
PDB ID	1I6I	1VFV	1VFX	1VFZ	1I5S

Table: Nucleotide states and the corresponding analog states. PDB IDs indicate of X-ray structures used as the reference structures of Gō-like models.

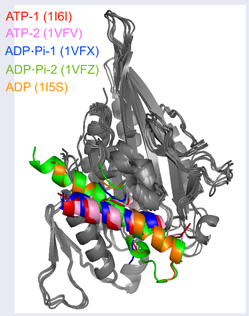

Figure: Superposition of the five reference structures. Highlighted are the switch II cluster regions.

Model

1. Total energy

$V_{\rm tot} = V_{\rm p}(\text{protein}) + V_{\rm n}(\text{nucleotide}) + V_{\rm pn}(\text{interaction}) + V_{\rm nn}(\text{for ADP·Pi})$

2. Intra-molecule energy

$$V_{\rm p,n}(\Gamma,\Gamma^{(0)}) = \sum_{\rm bonds} K_r\left(b_i - b_i^{(0)}\right)^2 + \sum_{\rm angles} K_\theta\left(\theta_i - \theta_i^{(0)}\right)^2 + \sum_{\rm dihedrals} K_\phi\left[\left(1-\cos(\phi_i-\phi_i^{(0)})\right) + \frac{1}{2}\left(1-\cos 3(\phi_i-\phi_i^{(0)})\right)\right] + \sum_{i<j-3}^{\substack{\text{native}\\ \text{contacts}}} k_{\rm nc}\left[5\left(\frac{r_{ij}^{(0)}}{r_{ij}}\right)^{12} - 6\left(\frac{r_{ij}^{(0)}}{r_{ij}}\right)^{10}\right] + \sum_{i<j-3}^{\substack{\text{non-native}\\ \text{contacts}}} k_{\rm nnc}\left(\frac{C}{r_{ij}}\right)^{12}$$

$\Gamma^{(0)}$: *referencestructure*,r_{ij}: distance between the ith and jth C_α,
$b_i = r_{i\ i+1}$

3. Interaction energy

$$V_{\rm pn}(\{r_{i,k}\}) = \sum_{i,k}^{\substack{\text{native}\\ \text{contacts}}} k_{\rm pn}\left[5\left(\frac{r_{ik}^{(0)}}{r_{ik}}\right)^{12} - 6\left(\frac{r_{ik}^{(0)}}{r_{ik}}\right)^{10}\right] + \sum_{i,k}^{\substack{\text{non-native}\\ \text{contacts}}} k_{\rm pn}^{\rm nnc}\left(\frac{C}{r_{ik}}\right)^{12}, \quad (1)$$

Similar for V_{nn}

Results without nucleotide

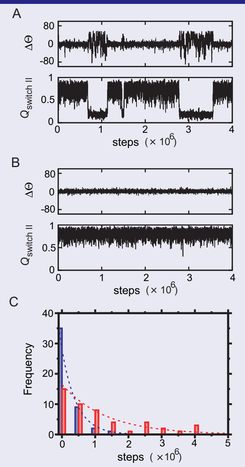

Figure: Dynamics of helix α4 for (A) ATP-1 (B) ADP·Pi-2. ΔΘ:Relative angle of α4 to the main body. $Q_{\rm switchII}$:Contact fraction of the native contacts between the switch II and the main body. (C) The histogram of the duration time steps for burst (blue: on-burst, and red: off-burst) in the ATP-1 state.

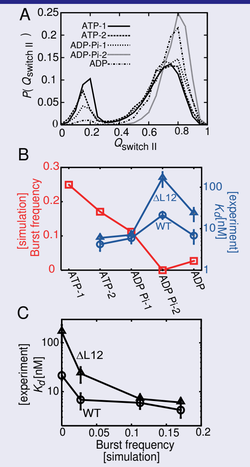

Figure: (A) Probability distribution $Q_{\rm switchII}$ (B) Nucleotide state dependence of the burst frequency at $T = 1.3$ and the equilibrium dissociation constants K_d obtained experimentally(R.Nitta et al., Science 305(2004) 678) (C) Burst frequency and the equilibrium dissociation constants.

Bust: A large structural fluctuation

1. The helix α4 at the MT binding site intermittently exhibits abrupt large structural fluctuations (Burst).
2. Frequency of the Burst varies systematically according to the nucleotide states and correlates strongly with the experimentally observed binding affinity to MT.

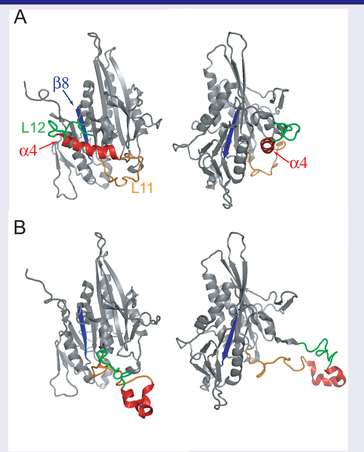

Figure: Snapshots of ATP-1 state. (A) off burst: $Q_{\rm switchII} \sim 0.75$. (B) on burst: $|\Delta\Theta_{\alpha 4}| \sim 80°$ and $Q_{\rm switchII} \sim 0.15$. blue: β8, res: α4, green: L12, orange: L11

Effect of temperature and nucleotide

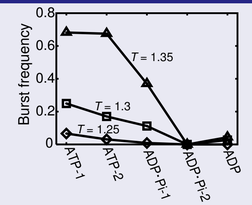

Figure: Temperature dependence of the burst frequency without nucleotide

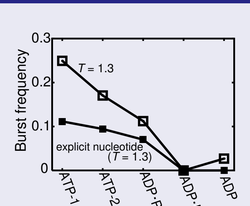

Figure: Burst frequency with and without nucleotide

Conclusion

We propose the following mechanism for the MT affinity regulation:

1. The flexibility of the MT binding site (helix α4 in particular) is allosterically controlled according to the nucleotide states during the ATP hydrolysis process.
2. The affinity to MT is regulated through the flexibility of the MT binding site.
3. The more flexible the MT biniding site is, the stronger the binding affinity becomes.

R.Kanada, F.Takagi and M.Kikuchi, Proteins **83** (2015) 809.