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A non-Markov ratchet model of molecular motors: processive movement of single-headed kinesin KIF1A*

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A fluctuating ratchet model of non-Markov process is presented to describe the processive movement of molecular motors of single-headed kinesin KIF1A, where the fluctuation perturbation to the local potential is introduced and the detailed ATPase pathway of the motor is included. The theoretical results show good quantitative agreement with the previous experimental ones.

Keywords: kinesin, model, molecular motor, kinetics
PACC: 3620, 8715B, 8715H, 8715K

1. Introduction

Kinesin KIF1A is a naturally single-headed motor protein that can move processively along microtubule (MT) by hydrolysing ATP (adenosine-triphosphate).[1,2] Recently, its dynamic behaviours have been experimentally studied with an optical trapping system.[3] Obviously, the microscopic mechanism for the processive movement of single-headed kinesins must be different from that of conventional two-headed kinesins.[4,5] For the latter, it was experimentally determined that they moved processively in a hand-over-hand manner.[6,7] The prevailing models for the dimeric kinesin require that the two heads move in a tightly coordinated manner.[8−13] Recently we proposed a new hand-over-hand model in which the ATPase cycle of the two heads was partially coordinated.[14] The model gives results showing good quantitative agreement with the experimental ones on both wild-type homodimeric kinesins and various mutant homodimeric and heterodimeric kinesins.

The unidirectional movement mechanism for single-headed kinesin motors such as KIF1A is usually described by the Brownian ratchet model of a Markov process.[15−17] In that model, the monomeric kinesin is treated as a Brownian particle that moves in an asymmetric periodic potential. ATP binding changes something about the motor, which then sees a new, different, but still periodic potential landscape. This flashing of the periodic potential breaks the balance and thus unidirectional movement becomes possible. In Refs.[1–3], Okada and co-workers proposed a modified ratchet model in order to explain their experimental results such as the step-size distribution and the load dependences of mean step size and velocity.

The Markov ratchet model predicts that the probability of futile hydrolysis is high, which is inconsistent with the experimental conclusion that a single ATP hydrolysis produces a single mechanical step of KIF1A.[3] In this work we present a non-Markov ratchet model to describe the unidirectional movement of single-headed kinesins. In this model, ATP hydrolysis induces a change in the single MT subunit to which the motor is bound at that time. Consequently, the potential is no longer instantaneously periodic. The model includes the detailed ATPase pathway of the motor and can give a quantitative comparison with the experimental results on KIF1A.[3] The experimental conclusion that a single ATP hydrolysis triggers a single stepping movement of the motor can be well explained by using the model.

2. Model

It has been determined that the single-headed

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kinesin KIF1A has +16 net elementary charges and each tubulin heterodimer of an MT has −27 net elementary charges (the α-tubulin monomer having −12 net charges and the β-tubulin monomer having −15 net charges) (PDB ID code 1IAO).[18] Thus the interaction between a kinesin head and a tubulin heterodimer should be mainly electrostatic. In addition, it is known that the affinity of a kinesin head for MT depends on its nucleotide state.[19,20] Generally, in nucleotide-free, ATP or ADP.Pi state, the kinesin head has a strong and stereospecific interaction with MT. In ADP (adenosine diphosphate) state, the kinesin head has a weak interaction with MT. The transition from strong to weak interactions occurs just at or after the Pi release.[20] We describe in detail the evolution of the interaction force between kinesin and MT during one ATPase cycle as follows. We start with the kinesin bound strongly to MT in nucleotide-free state, with the interaction potential schematically shown in Fig.1(a). The asymmetric potential is due to the asymmetric charge distribution on tubulin heterodimer surface. Then ATP binding and ATP hydrolysis occur while kinesin remains its strong binding to MT. As has been done in the model for two-headed kinesins[14] the interaction potential, immediately after Pi release, is assumed to be the same as that shown in Fig.1(b). After a period of time, $t_{\rm r}$, the potential changes to that shown in Fig.1(c). The origin for this evolution of the potential, as shown in Fig.1(b) and Fig.1(c), will be discussed later.

According to the above descriptions, the kinesin movement during one ATPase cycle can be divided into the following two periods:

(i) Stepping period—from Pi release to ADP release [Figs.1(b) and 1(c)]. In this period, kinesin has a weak interaction with MT in ADP state. This period corresponds to the ADP-release time (or inverse of ADP-release rate). It also corresponds to the stepping time of kinesin in its stepwise movement and is independent of ATP concentration, which is in agreement with the experimental observation.[3]

In the over-damped environment, the movement of the motor can be described by the following Langevin equation:

$$\Gamma\frac{{\rm d}x}{{\rm d}t} = -\frac{\partial}{\partial x}U(x,t) + \xi(t), \qquad (1)$$

where $U(x,t)$ is the interaction potential between the ADP-state kinesin and MT. Its shape corresponds to that shown in Fig.1(b) when $t \le t_r$, and to that shown in Fig.1(c) when $t > t_{\rm r}$. $\xi(t)$ is the fluctuating Langevin force, with $\langle\xi(t)\rangle = 0$ and $\langle\xi(t)\xi(t')\rangle = 2k_BT\Gamma\delta(t-t')$.

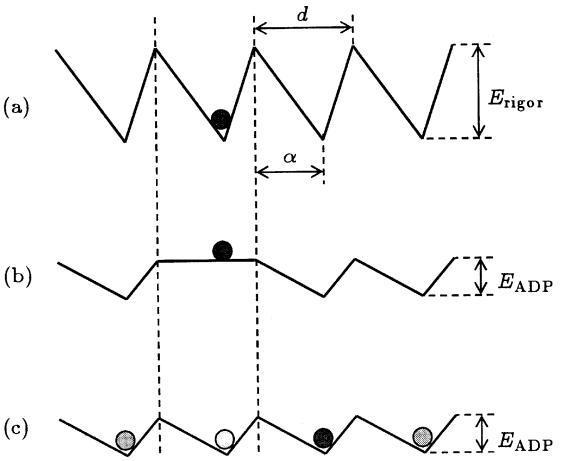


Fig.1. The asymmetric sawtooth potential for the single-headed kinesin in (a) rigor state (including nucleotide-free, ATP, and ADP.Pi states), (b) ADP state immediately after Pi release, (c) ADP state in equilibrium. The potential depth in (a) should be slightly different for the different nucleotide states. The grey scale of the sphere from dark to white in (c) represents the probability from high to low of the motor at different binding sites along MT.

In our calculation we assume that the kinesin releases ADP with a probability $P_{\rm release}$ near the potential minima. Its value is related to the ADP-release rate.

(ii) Dwell period—from ADP release to Pi release [Fig.1(a)]. In this period, ATP binding, ATP hydrolysis and Pi release occur sequentially, and kinesin remains its strong binding to MT at the position of the potential minimum where it has just released ADP. This period corresponds to the dwell time in the stepwise movement of kinesin, which depends on the ATP concentration.[3]

Here we discuss two possible origins for the evolution of the potential shown in Figs.1(b) and 1(c). (i) The release of Pi induces the conformational change in the binding site of kinesin head, which results in its weak binding to MT, with the depth, $E_{\rm ADP}$, of the interaction potential being much shallower than that in rigor state, $E_{\rm rigor}$. At the same time, the conformational change in kinesin head can also induce some conformational change in the binding site of the MT tubulin heterodimer where the kinesin head is bound. This conformational change in the binding site of the tubulin results in its weaker interaction with the kinesin head. The interaction potential is shown in

Fig.1(b). After a period of time, t_r, the conformation of the binding site of the tubulin where kinesin has just been detached relaxes to its normal conformation. Thus the local potential relaxes to the normal depth E_{ADP} as shown in Fig.1(c). It is argued here that the MT tubulin heterodimers also exhibit some elasticity rather than rigidity as previously believed. (ii) When there is a conformational change following Pi release, there appears a gap between the kinesin head and the tubulin heterodimer. Therefore, the solution will flow into the gap, and certain multivalent counterions in the solution will be bound to (or adsorbed onto) the negatively-charged tubulin heterodimer surface, resulting in the shielding effect on the negative charges of the tubulin heterodimer.[21] Thus the local electrostatic potential depth becomes much shallower immediately after Pi release. For simplicity, the local potential is treated as flat, as shown in Fig.1(b). Note that the charge density of counter-ions at onset should be higher than that at equilibrium, since, due to counteractive forces, the counter-ions approach the tubulin surface earlier than the like-charge ions. After a period of time, t_r, the charge density of the counterions adsorbed onto the tubulin surface will relax to equilibrium. The local potential relaxes to the normal depth E_{ADP} as shown in Fig.1(c).

3. Results and discussion

In order to compare with the experimental results,[3] we solve Eq.(1) numerically. From Stokes formula we obtain $\Gamma = 6\pi\eta r_k = 1.883 \times 10^{-9}\mathrm{kg \cdot s^{-1}}$, where the viscosity η of the aqueous medium around kinesin is approximately $0.01\mathrm{g \cdot cm^{-1} \cdot s^{-1}}$ and $r_k = 0.1\mu\mathrm{m}$, which is the radius of the beads attached to KIF1A in the experiment[3] To be consistent with the experimentally measured ADP-release rate of kinesin,[10,12,22] we take the ADP-release probability as $P_{release} = 300\mathrm{s^{-1}}$. Thus the mean stepping time is 3.3ms, with the stepping-time distribution shown in Fig.2. This value is consistent with the measured mean stepping time ($\approx 4\pm1$ms) for KIF1A[3] and the stepping-time distribution is very similar to the measured one (Fig.2(c) in Ref.[3]). The parameters for the potential, i.e., the potential barrier E_{ADP} and asymmetry α, are unavailable experimentally. For calculation, we take $E_{ADP} = 7.8k_BT$ and $\alpha = \frac{7}{10}d$, where $d = 8$nm is the period of MT. This value of α corresponds to the asymmetrical charge distribution on one tubulin heterodimer of MT.

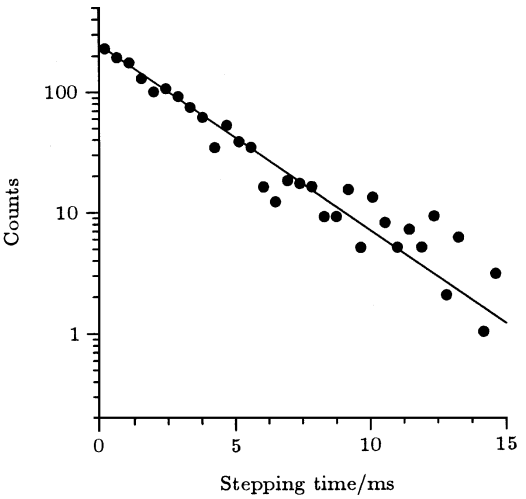


Fig.2. Distribution of the stepping time.

In Fig.3 we give the distributions of step size under no load for various t_r. First, we note that the distributions are of Gaussian form with a half width of about 24 nm. This is consistent with the experimental result (Fig.2(h) in Ref.[3]). Second, as t_r increases, the probability of futile ATP hydrolysis (i.e., the

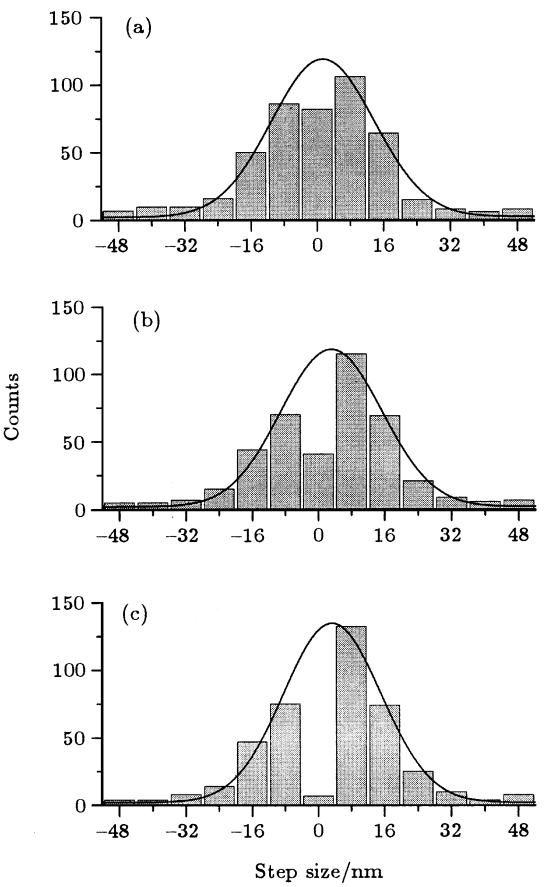


Fig.3. Distribution of the step size under no load with (a) $t_r = 0.2$ ms, (b) $t_r = 1$ ms, and (c) $t_r = 5$ ms. Solid curves are for Gaussian fittings.

probability at $-4\,\mathrm{nm} < x \leq 4\,\mathrm{nm}$) decreases. This result shows good agreement with the conclusion from Ref.[3] that a single mechanical step of KIF1A is produced by a single ATP hydrolysis.

To see the dependence of mean step size <step> on load, we plot in Fig.4 the force–<step> curve, together with the experimentally measured values. It is seen that the theoretical results are in good agreement with the experimental ones. As mentioned before, the total time period for one step (or one ATPase cycle) is $t_{\mathrm{total}} = t_{\mathrm{dwell}} + t_{\mathrm{stepping}}$, where $t_{\mathrm{dwell}} = t_{\mathrm{b}}/[\mathrm{ATP}] + t_{\mathrm{hydrolysis}} + t_{\mathrm{Pi\text{-}release}}$ is the dwell period and $t_{\mathrm{stepping}} = t_{\mathrm{ADP\text{-}release}}$ the stepping period of the motor movement. Thus the mean velocity of the kinesin is

$$V = <\mathrm{step}>/t_{\mathrm{total}} = <\mathrm{step}> \frac{k_{\mathrm{c}}[\mathrm{ATP}]}{[\mathrm{ATP}] + k_{\mathrm{c}}/k_{\mathrm{b}}}, \qquad (2)$$

where $k_{\mathrm{b}} = 1/t_{\mathrm{b}}$ is the ATP binding rate and $k_{\mathrm{c}} = 1/t_{\mathrm{c}} = (t_{\mathrm{hydrolysis}} + t_{\mathrm{Pi\text{-}release}} + t_{\mathrm{ADP\text{-}release}})^{-1}$ the ATP turnover rate. Note that Eq.(2) is the well-known Michaelis–Menten relation. Using Eq.(2) and the theoretical result (solid line) in Fig.4 we fit the experimental load–velocity curves (Fig.1(d) in Ref.[3]) and the results are given in Fig.5 (where $1\mathrm{M}=10^{-3}\mathrm{mol/cm}^3$). It can be seen that the theoretical results agree well with the experimental ones. The fitted value of $k_{\mathrm{c}} \approx 56\,\mathrm{s}^{-1}$ is close to the measured value of $\sim 110\ \mathrm{s}^{-1}$.[1,2]

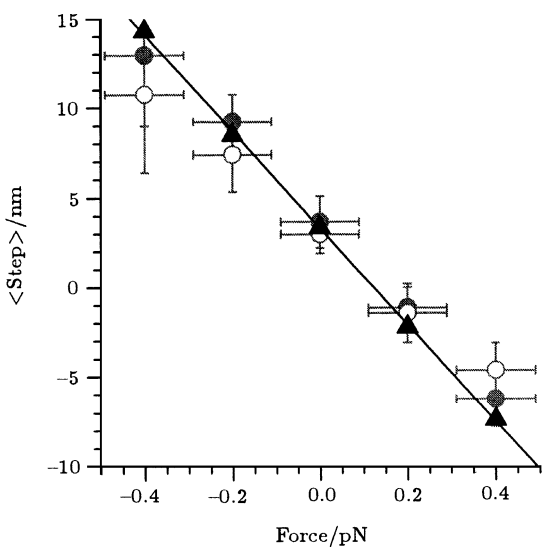


Fig.4. Dependence of the mean step size on load. The triangles are for theoretical results. The straight line is a linear fit to the theoretical points. The circles are for experimental results cited from Ref.[3].

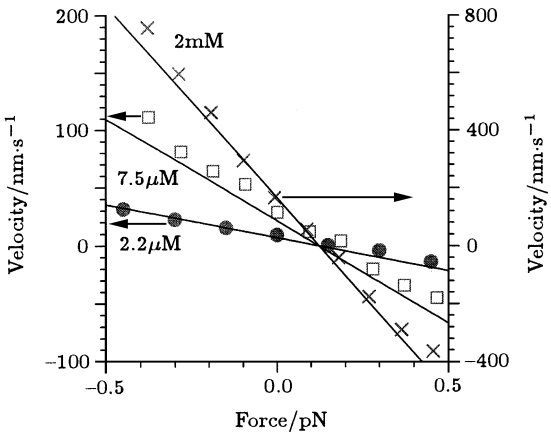


Fig.5. Force–velocity curves at different ATP concentrations. The symbols are for experimental results cited from Ref.[3]. The solid lines are for theoretical results with $k_{\mathrm{b}} = 1\mu\mathrm{M}^{-1}\cdot\mathrm{s}^{-1}$ and $k_{\mathrm{c}} = 56\,\mathrm{s}^{-1}$.

Now we give the explanation to the experimentally observed rapid plus-end-directed displacement of KIF1A on binding to the microtubule.[3] Before binding to MT the position of KIF1A should be distributed evenly along MT though the potential is asymmetric along MT. On binding to MT the kinesin will be driven rapidly to the potential minima. Thus the ensemble average binding-induced rapid displacement should be $\delta_{\mathrm{bind}} = \frac{1}{2}\alpha\left(\frac{\alpha}{d}\right) - \frac{1}{2}(d-\alpha)\left(1-\frac{\alpha}{d}\right) = 1.6\mathrm{nm}$, which is approximately consistent with the measured value $2.8 \pm 0.8\mathrm{nm}$.[3]

An interesting result from experiments is that the mean velocity of KIF1A motor under no load is insensitive to whether the motor is attached to a latex bead or not.[1–3] To explain this result, we have also calculated the step-size distribution using $r_{\mathrm{k}} = 10\,\mathrm{nm}$ for Γ under no load. We have noted that, except that the distribution is wider than that with $r_{\mathrm{k}} = 0.1\mu\mathrm{m}$, the mean step size <step>, $\sim 3.7\,\mathrm{nm}$, is close to $\sim 3.3\,\mathrm{nm}$ obtained with $r_{\mathrm{k}} = 0.1\mu\mathrm{m}$. This can be understood as follows. From our model, it is approximated that the mean step size and thus the mean velocity depend mainly on the free diffusion of Brownian particle in the range between the two dashed lines in Fig.1(b) and are insensitive to the diffusion outside this range.

Since both Astumian's model.[15] and the present model are based on the fluctuating ratchet, it is interesting to make a comparison between the two. In order to obtain a net unidirectional movement, both

models have the two prerequisites: an asymmetric periodic potential and a dichotomously fluctuating perturbation to it. However, in Astumian's model.[15] the perturbation extends to the global potential, which is induced by changing something of the motor following a change in its nucleotide state; whereas in the present model the fluctuating perturbation extends both to the global potential and to the local potential where the kinesin has just released Pi, which is induced by changing something of the MT subunit to which the motor is bound at that time. That means that, in the former model the Pi release induces only the conformational change in kinesin head and has no effect on MT subunit (i.e. MT subunit is presumed to be rigid); whereas in the present model the Pi release induces the conformational change in kinesin head, which in turn induces some conformational change in binding site of MT where the kinesin head is bound (i.e., MT subunit is presumed to be elastic). We interpret the interaction potential change to be due to counter-ion effect, which means that, in the former model the Pi release induces shielding effect on the positive charges of kinesin; whereas in the present model the Pi release induces shielding effect on the negative charges of MT at the site where kinesin is bound. This difference in perturbation leads to the fact that the present model gives a much more efficient mechanochemical coupling than the former model. For example, in the present model about 2 ATPs are consumed for moving 8 nm forward with $\alpha = \frac{7}{10}d$ (see Fig.4) and in the former model about 56 ATPs are consumed for moving 8 nm forward even with $\alpha = \frac{7}{8}d$ (see the discussion in Ref.[15]). Furthermore, as seen before, the results from the present model are in good quantitative agreement with the experimental results on KIF1A. However, as will be seen below, the results from the former model are not in agreement with the experimental ones.

First, from Astumian's model the probability distribution of the motor's position is described by $P(x) = \exp[-(x - t_{\rm off}F_{\rm load}/\Gamma)^2/4Dt_{\rm off}]/\sqrt{4\pi Dt_{\rm off}}$,[16] where $D = k_{\rm B}T/\Gamma$ is the diffusion coefficient. $t_{\rm off}$ is the time period when the potential is off, which is equal to the stepping time. It can be seen that, at $F_{\rm load} = 0$, $P(x)$ always has a maximum at x=0. Using this equation and $t_{\rm off}$= 3.3 ms it can be obtained that the half-width of the step-size distribution at $F_{\rm load} = 0$ is about 170 nm. This deviates significantly from the experimental result of about 24 nm (Fig.2(h) in Ref.[3]). The mean step size at, for example, $F_{\rm load} = -0.4$pN is approximately $-t_{\rm off}F_{\rm load}/\Gamma \approx$ 600 nm, which also deviates significantly from the experimental result of about 13 nm (Fig.2(i) in Ref.[3]). Second, in order to be consistent with the above experimental results for KIF1A, $t_{\rm off}$ has to be taken as $\sim$10 μs, which is about 3 orders of magnitude shorter than the measured mean stepping time ($\approx 4\pm1$ ms) for KIF1A.[3] Taking this value as $t_{\rm off}$ and using the model the estimated probability of futile hydrolysis should be larger than 20%. This is inconsistent with the conclusion from Ref.[3] that a single mechanical step of KIF1A is produced by a single ATP hydrolysis.

4. Conclusion

We present a new fluctuating ratchet model for unidirectional movement of single-headed kinesins. The model is different from the conventional ratchet model: in our model the fluctuating perturbation extends to both the whole potential and the local potential where the kinesin head is bound, whereas in the conventional model the perturbation extends only to the whole potential. The perturbation to the local potential leads to a non-Markov process and a more efficient mechanochemical coupling for the kinesin movement. In particular, using this model, the calculations for the unconventional single-headed kinesin KIF1A are in good quantitative agreement with previous measurements. Further research on the origin of this non-Markov interaction of kinesin with MT would be very important. This interaction could also be relevant to other motor proteins such as myosins.[23]

References

[1] Okada Y and Hirokawa N 1999 *Science* **283** 1152

[2] Okada Y and Hirokawa N 2000 *Proc. Natl. Acad. Sci.* USA **97** 640

[3] Okada Y, Higuchi H and Hirokawa N 2003 *Nature* **424** 574

[4] Vale R D and Fletterick R J 1997 *Annu. Rev. Cell Dev. Biol.* **13** 745

[5] Block S M 1998 *Cell* **93** 5

[6] Asbury C L, Fehr A N and Block S M 2003 *Science* **302** 2130

[7] Yildiz A, Tomishige M, Vale R D and Selvin P R 2004 *Science* **303** 676

[8] Peskin C S and Oster G 1995 *Biophys. J.* **68** 202s
[9] Duke T and Leibler S 1996 *Biophys. J.* **71** 1235
[10] Mandelkow E and Johnson K A 1998 *Trends Biochem. Sci.* **23** 429
[11] Vale R D and Milligan R A 2000 *Science* **288** 88
[12] Thomas N, Imafuku Y, Kamiya T and Tawada K 2002 *Proc. R. Soc. Lond. B. Biol. Sci.* **269** 2363
[13] Asenjo A, Krohn N and Sosa H 2003 *Nat. Struct. Biol.* **10** 836
[14] Xie P, Dou S X and Wang P Y 2005 *Chin. Phys.* **14** 734
[15] Astumian R D and Bier M 1994 *Phys. Rev. Lett.* **72** 1766
[16] Astumian R D 1997 *Science* **276** 917
[17] Jülicher F, Ajdari A and Prost J 1997 *Rev. Mod. Phys.* **69** 1269
[18] Kikkawa M, Sablin E P, Okada Y, Yajima H, Fletterick R J and Hirokawa N 2001 *Nature* **411** 439
[19] Uemura S, Kawaguchi K, Yajima J, Edamatsu M, Toyoshima Y Y and Ishiwata S 2002 *Proc. Natl. Acad. Sci.* USA **99** 5977
[20] Nitta R, Kikkawa M, Okada Y and Hirokawa N 2004 *Science* **305** 678
[21] For the effect of multivalent counter-ions in aqueous media on interaction between large biological polyelectrolytes, see, for example, Butler J C. Angelini T, Tang J X and Wong G C L 2003 *Phys. Rev. Lett.* **91** 028301 and references therein
[22] Gilbert S P, Moyer M L and Johnson K A 1998 *Biochemistry* **37** 792
[23] Xie P, Dou S X and Wang P Y 2005 *Chin. Phys.* **14** 744