Mechanochemical Theory for the ATP-fuelled Biomolecular Motors

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Abstract—Biomolecular motors are normally single or complex biomolecules exerting mechanical forces across molecular and cellular scales. The ATP-fuelled biomolecular motors can transduce the chemical energy from ATP hydrolysis into forces and motions in cells. In biomolecular motors, transport reactions are both stoichiometric and enzymatic. We set up a mechnochemical theory for biomolecular motors to understand its enzymatic kinetics and continuous dynamics. This theory is validated by modeling the operating mechanism and dynamics of several ATP-fuelled molecular motors driving various loads.

Keywords—biomolecular motors; mechnochemical properties; ATPase; enzymatics; Langevin dynamics.

I. INTRODUCTION

Biomolecular motors are natural proteins or hybrid molecular systems that exert forces and motions at both molecular and cellular scales while fulfilling unique cellular functions [1]. Biomolecular motors are normally powered by the hydrolysis or synthesis of nucleotides, such as adenosine triphosphate (ATP), and/or by photon or proton-motivated forces. For the ATP-fuelled biomolecular motors, mechnochemistry involves the conversion of ATP catalysis energy into mechanical forces and motions with exceptional thermodynamic efficiency. Experiments down to the single molecular level have revealed the operating dynamics of some biomolecular motors, such as, of the linear kinesin and myosin motors, and the rotary F$_1$-ATPase motor [1,3-7]. The dynamics are found to be dependent on the physiological and external load conditions. While in biomolecular motors the time scale of ATP reactions and conformational changes are at sub nano-second or shorter, the directional motions with loading are at micro-second or longer. Currently for the ATP-fuelled molecular motors, there are gaps in understanding of the molecular structures and pathways versus the operational forces and driving capabilities. Determining a valid mechnochemical theory is fundamentally important for a full understanding of biomolecular motors.

We have proposed a mechnochemical theory for the F$_1$-ATPase motor [2]. The theory was based on enzyme kinetics and Langevin dynamics. The introduction of an effective ‘ratchet’ drag coefficient and a chemomechanical coefficient has correlated the molecular catalysis and micro operating dynamics. The theory successfully reproduces results from motor experiments. We now extend it to the general cases of steady operations of the ATP-fuelled molecular motors.

II. MECHANOCHEMISTRY IN THE ATP-FUELLED BIOMOLECULAR MOTORS

A. Energy Transduction in the ATP-fuelled Biomolecular Motors

The ATP-fuelled molecular motors are intrinsically ATP synthase (ATPase) proteins. Their power source comes from the ATP hydrolysis (or synthesis) reaction,

\[ \text{ATP}^+ + \text{H}_2\text{O} \rightarrow \text{ADP}^3+ + \text{H}_2\text{PO}_4^- \] (1)

A core question is how these biomolecular motors harness the free energy from this simple reaction to generate forces and drive the entire motor in directional motions [1]. The tight coupling between ATP hydrolysis and the mechanical steps is the foundation of the mechnochemical mechanisms. This is important also because of the essential roles of biomolecular motors in energy transduction, regulation of metabolism and other cellular functions.

B. Mechnochemical Mechanism of Biomolecular Motors

In the ATP-fueled molecular motors, a conformational change of the structural elements caused by ATP hydrolysis at the ATP-binding site is communicated to either the next corresponding track-binding site (for kinesin and myosin) or to the alternative binding site (for F$_1$-ATPase). It is then relayed at much large spatial scales via homotropic structural elements to a mechanical amplifier of the active subunits [1,3-5]. The mechnochemical coupling ratio — defined as the number of ATP molecules hydrolysed per mechanical step — remains 1:1 over a wide range of loads [1,3,5-7]. The basics concerning the mechnochemistry can be broken down into the hydrolysis of nucleotides (mainly ATP and its analogues), inducing the conformational changes, and tightly coupled transferal of conformational changes into forces and other mechanisms of the active subunits.

III. MASTER EQUATIONS FOR THE MECHANOCHEMISTRY

To solve the mechnochemical processes from ATP reactions to the micro stepping/rotating motions, we set up the
following theory based on enzyme kinetics and modified Langevin dynamics.

A. Pathway of Conformational Changes

Under cellular physiological conditions (either *in vivo* or *in vitro*), the pathway of conformational changes [3-5] of the active subunits of the ATP-fuelled molecular motors is found to be,

\[
\begin{align*}
\text{MM}^0 & \xrightarrow{k_{\text{ATP}}} \text{MM}^\text{ATP} \xrightarrow{k_{\text{off}}} \text{MM}^\text{ADP} \xrightarrow{k_{\text{Pi}}} \text{MM}^\text{Pi} \\
\text{ATP} & \xrightarrow{k_{\text{off}}} \text{ADP} \\
\text{Pi} & \xrightarrow{k_{\text{Pi}}} \text{ADP}
\end{align*}
\]

where MM refers to the enzymatic molecular motor. The binding conformational states of the multiple catalysis subunits are identified as tight binding (TB, bound with ATP), loose binding (LB, bound with ADP and/or Pi), and Open (O), respectively. The Open (O) site has a very low affinity for substrates and is catalytically inactive. \(k_{\text{ATP}}, k_{\text{off}}, k_{\text{Pi}}, k_{\text{ADP}}\) refer to the rate constants of association and dissociation of ATP (or ADP and Pi) molecules to the motor, i.e., they are the rate of ATP (or ADP and Pi) binding to or unbinding from active subunits. The terms \(k_{\text{on}}, k_{\text{off}}\) are the hydrolysis and synthesis rate constants of ATP. In this catalysis pathway, the direction of hydrolysis resulting in binding/unbinding to the motor of ATP, ADP and Pi is given by the solid arrow heads. In each hydrolysis cycle, the different subunits interact in a cooperative way as the motion domains sequentially contact and interact with each other. The binding and conformational changes are highly coupled to the ATP hydrolysis cycle at different catalysis sites. In steady cycles of ATP hydrolysis reactions, the hydrolysis reactions at the active sites could be regarded as an equilibrium process with the same reaction sequence and dynamics.

B. Enzymatic Kinetics

To interpret Eq. (2) in terms of kinetics under a steady equilibrium, where the steps of forces and motions are coupled with the chemical reaction cycles, we derived the kinetics equations [2],

\[
\begin{align*}
p_{\text{O}} + P_{\text{ATP}} + P_{\text{ADP}} + P_{\text{Pi}} &= 1 \\
k_{\text{ATP}} \cdot [\text{ATP}] \cdot p_{\text{O}} + k_{\text{off}} \cdot P_{\text{off}} - (k_{\text{on}} + k_{\text{off}}) \cdot P_{\text{ATP}} &= 0 \\
k_{\text{on}} \cdot P_{\text{ATP}} + k_{\text{on}} \cdot [P] \cdot P_{\text{off}} - (k_{\text{syn}} + k_{\text{off}}) \cdot P_{\text{ADP}} &= 0 \\
k_{\text{off}} \cdot P_{\text{ADP}} + k_{\text{off}} \cdot [\text{ADP}] \\
k_{\text{ADP}} \cdot P_{\text{ADP}} + k_{\text{off}} \cdot [\text{ADP}] \cdot P_{\text{syn}} - (k_{\text{on}} + k_{\text{off}}) \cdot P_{\text{ATP}} &= 0 \\
k_{\text{ATP}} \cdot P_{\text{ATP}} + k_{\text{ATP}} \cdot [\text{ATP}] \cdot P_{\text{syn}} - (k_{\text{off}} + k_{\text{ATP}}) \cdot P_{\text{ADP}} &= 0
\end{align*}
\]

where [ATP], [ADP] and [Pi] are the concentrations of ATP, ADP and Pi respectively. \(p_{\text{O}}, P_{\text{ATP}}, P_{\text{ADP}}\) and \(P_{\text{Pi}}\) are the probability of states when different active subunits of the motor are either empty or occupied by ATP, ADP, Pi or ADP molecules, respectively. From Eq. (3), the overall kinetic rate of catalytic hydrolysis, \(R\), can be determined by

\[
R = k_{\text{on}} \cdot P_{\text{ATP}} - k_{\text{syn}} \cdot P_{\text{ADP}}.
\]

C. Langevin Dynamics with a ‘Ratchet’ Mechanism

Langevin dynamics can be used to implicitly simulate the effect of a solvent on biomolecules, which in our case are the motor proteins. In setting up the Langevin equations for biomolecular motors, the solvent molecules are excluded and their effect on the dynamics of the solute macromolecule is incorporated by an electrostatics potential-of-mean-force term added to the potential \(U(x)\), by random collisions (the random force vector below), and by a frictional drag on its motion through the solvent (the friction term \(\zeta\) or \(\zeta x\) below).

We define the master equations of Langevin dynamics for a (a) linear or (b) rotating molecular motor respectively as [2, 8],

\[
\begin{align*}
\dot{v} &= \frac{dx}{dt} = f(t), \\
\dot{p} &= \zeta v + f(t)
\end{align*}
\]

(b) for a rotating molecular motor

\[
\begin{align*}
\dot{\omega} &= \frac{dH}{dt} = \tau_{\text{ext}} - \zeta \omega + \tau_{\text{stat}} \quad (4b)
\end{align*}
\]

where \(v\) (or \(\omega\)) is the velocity (or angular velocity) of the molecular motor;

\(p\) (\(L\)) is the momentum (angular momentum) of the motor system. It is conserved for the steady operation of molecular motors;

\(f_{\text{stat}}\) (\(\tau_{\text{ext}}\)) is the force (torque) generated directly from the hydrolysis reactions at corresponding functional subunits of the motor, \(f_{\text{stat}} = -\frac{\partial U(x)}{\partial x}\) (\(\tau_{\text{ext}} = -\frac{\partial U(x)}{\partial \theta}\)) and \(U\) is the corresponding potential energy at position \(x\) (or \(\theta\)) of the molecular motor. It is expected that the molecular motor will switch between a series of ‘U’ when it steps or rotates and undergoes continuous conformational changes;

\(f_{\text{ext}}\) (\(\tau_{\text{ext}}\)) is the force (torque) on a load applied by an external force (if any);

\(\zeta v\) (\(\zeta \omega\)) is the frictional drag term in a viscous medium, with \(\zeta\) the frictional factor of the load against the media;

\(f_{\text{bd}}\) (\(\tau_{\text{bd}}\)) is the Brownian motion term due to thermal fluctuation-dissipation of the acting subunits of the motor and its loading.

Given the very fast Langevin relaxation time, e.g. approximately \(2 \times 10^{-12}\) sec for the case of \(F_1\) driving an actin filament, a steady state approximation implies that the moment of inertia is constant and the momentum (or angular momentum) should be conserved. But we have to take into account the ‘ratchet’ mechanism in the ATP-fuelled biomolecular motors. The ‘ratchet’ mechanism is a critical
feature of biomolecular motors. It is either a structural or thermodynamic mechanism that is capable of utilizing random thermal fluctuations to generate a unidirectional drive to the motor [2]. We define a conserved Langevin with the 'ratchet' restrictions for Eq. (4a) & (4b), as following,

(a) for a linear molecular motor
\[
\left\langle \frac{d \mathbf{r}}{dt} \right\rangle = \zeta \mathbf{v}/v
\]
\[
< f_g(t) >= 0, \quad < f_g(t) f_g(t') >= 2 k_B T \zeta \delta(t-t')
\]

(b) for a rotating molecular motor
\[
\left\langle \frac{d \mathbf{l}}{dt} \right\rangle = 0, \quad \left\langle \frac{d \mathbf{l}}{dt} \right\rangle_o = \zeta \mathbf{o}/\mathbf{r}
\]
\[
< \mathbf{r}_g(t) >= 0, \quad < \mathbf{r}_g(t) \mathbf{r}_g(t') >= 2 k_B T \zeta \delta(t-t')
\]

Note that \( \delta(t-t') \) is the Dirac \( \delta \)-function and the Brownian fluctuation is represented by Gaussian white noise. To account for the 'ratchet' mechanism occurring in biomolecular motors, we assume that the average momentum at any given time is in effect a constant drag (torque), \( \zeta \mathbf{v} \) (\( \zeta \mathbf{o} \)). Here a fitting constant, \( \zeta \), is introduced as the effective 'ratchet' drag coefficient. This will ensure the motor stepping or spinning unidirectionally. Physically in the distribution space, the value of 'ratchet' can be converted (via the Smoluchowski equation) to the diffusion constant of the motors' active subunits.

D. When Molecular Motors Drive a Load

When a molecular motor works steadily, e.g. when it drives an external load under the saturation [ATP] conditions, it was demonstrated that a stepwise stepping/rotation is accomplished with the hydrolysis of an ATP molecule per step [1,3,5-7]. Therefore we have, on average, \( \Delta U(x) = R \Delta G_{hod} \Delta t \) or \( \Delta U(\mathbf{0}) = R \Delta G_{hod} \Delta t \), which leads to,

(a) for a linear molecular motor
\[
\left\langle \frac{\partial U(x)}{\partial x} \right\rangle = \frac{\eta R \Delta G_{hod}}{v}
\]

(b) for a rotating molecular motor
\[
\left\langle \frac{\partial U(\mathbf{0})}{\partial \theta} \right\rangle = \frac{\eta R \Delta G_{hod}}{\mathbf{r}}
\]

where \( \eta \) as the chemomechanical coefficient, which indicates the efficiency of biomolecular motor in converting ATP hydrolysis energy into forces or torques. The perfect motor would possess \( \eta = 1.0 \). For a practical biomolecular motor, \( \eta \) is between 0~1.0 due to the fact that the motor is not a steady mechanochemical equilibrium system and it will have some energy dissipation, such as heat dissipation. As discussed previously, \( R \) is the overall ATP hydrolysis rate of the motor. In Eq. (6), the chemical energy released from ATP hydrolysis reactions is given by [9],

\[
\Delta G_{hod} = G_e + k_B T \ln \left[ \frac{[ADP][Pi]}{[ATP]} \right]
\]

with \( G_e = -50.74 \) pN.nm for the free energy released from the hydrolysis of a single ATP molecule at \( \rho H = 7 \) at \( 25^\circ C \).

In the single molecular and nano motor technologies [1], biomolecular motors normally drive two kinds of loads, (a) spherical beads or (b) polymeric filaments. We determine that the drag frictional factor, \( \zeta \), of these loads in the viscous media as,

(a) for linear-drag spherical beads [8],
\[
\zeta = 6 \pi r \zeta_o
\]

where \( \zeta_o \) (\( \sim 1 \times 10^3 \) Nm/\( s \)) the viscosity of the media, and \( r \) is the radius of the bead.

(b) for rotary-drag polymeric filaments [10],
\[
\zeta = \frac{4 \pi}{3} \frac{\zeta_o r^3}{\ln \left( \frac{l}{2r} \right)}
\]

where \( l \) and \( r \) are the length and the radius of the filament.

Substituting Eqs. (3), (5-8) into Eq. (4) allows us to determine the mechanochemical properties of biomolecular motors driving loads of beads or actin filaments. For example, the stepping speed of a motor, as a function of nucleotide concentration and the load strength or the actin length, can be determined.

IV. LINEAR KINESIN AND ROTARY F1-ATPASE

We now demonstrate the application of our mechanochemical theory to the ATP-fuelled motors of kinesin and F1-ATPase. In both cases, the Brownian term is treated as two parts: one is the effective drag force at the active subunits where the 'ratchet' mechanism takes place, and the remaining contribution is a disturbing term at the load. In Eqs. (4) and (5), the parameter \( \zeta_o \) accounts for the former component and \( \mathbf{r}_g \) (or \( \mathbf{r}_g \)) refers to the Brownian term from the loading and external perturbation. This treatment is justified as we attribute the internal Brownian fluctuation into the fitting parameter \( \zeta_o \) without knowing the exact potential landscape (i.e., \( U(\mathbf{0}) \)) in motors. This process averages the stochastic nature of internal fluctuations and accumulates the overall force from Brownian motions at the motor proteins.

A. Linear Kinesin Driving a Bead

Kinesin is a motor walking hand-over-hand along the microtubules (as shown in the inset of Figure 1) [4, 5, 7]. It was found that kinesin walks at the processive step of 8 nm/step while dragging different loads of beads [7]. The maximum velocity of stepping was estimated up to 800 nm/sec.

Figure 1 shows how the kinesin motor drives a bead load of different pN forces. The theoretical simulation ([ADP] and [Pi]) are set at cellular physiological conditions of 10 \( \mu \)M and 1.0 mM gives good agreement with the experimental data. The
chemomechanical coefficient of 0.6 ± 0.2 and an effective ‘ratchet’ drag coefficient of 0.03 ± 0.02 pN.nm.s are determined for this kinesin motor.

![Graph showing velocity vs ATP concentration](image)

Figure 1. Kinesin drags a bead of different load with an effective ‘ratchet’ drag coefficient of 0.03±0.02 pN nm s. $k_{\text{ATP}} = 1.1 \times 10^4$ M$^{-1}$s$^{-1}$, $k_{\text{ADP}} = \text{n.d.}$, $k_{\text{P}} = 1.5 \times 10^5$ s$^{-1}$, $k_{\text{PP}} = 2.0 \times 10^5$ s$^{-1}$, and $k_{\text{pp}} = 1.0 \times 10^5$ s$^{-1}$. $k_{\text{m}} = \text{n.d.}$ The symbols are from experimental measurements [7] and the solid lines are the simulation results.

B. Rotary $F_{1}$-ATPase Driving an Actin Filament

$F_{1}$-ATPase is a rotary motor while transducing energy from hydrolysis/synthesis of ATP molecules (as shown in the inset of Figure 2) [2, 3, 6].

![Graph showing actin length vs rotational rate](image)

Figure 2. The theoretical solid lines indicate that the chemomechanical coefficients of $F_{1}$-ATPase fall between 0.165 - 0.452 with an effective ‘ratchet’ drag coefficient of 1.07 pN nm s [2]. $k_{\text{m}} = 2.0 \times 10^5$ M$^{-1}$s$^{-1}$, $k_{\text{ATP}} = 8.90 \times 10^5$ M$^{-1}$s$^{-1}$, $k_{\text{ADP}} = 8.10 \times 10^5$ M$^{-1}$s$^{-1}$, $k_{\text{PP}} = 2.70 \times 10^5$ s$^{-1}$, $k_{\text{ADP}} = 4.90 \times 10^5$ s$^{-1}$, $k_{\text{P}} = 2.0 \times 10^5$ s$^{-1}$, and $k_{\text{pp}} = 4.5 \times 10^5$ s$^{-1}$, $k_{\text{m}} = 1.1 \times 10^5$ s$^{-1}$. The crosses are from experimental measurements [6]. [ADP] and [Pi] are set at cellular physiological condition of 10 μM and 1.0 mM, respectively.

Using the above chemomechanical theory, we could also determine the chemomechanical properties of the $F_{1}$-ATPase molecular motor when it drives an actin filament. The dependence of rotation rate upon the length of loading actin is plotted in Figure 2 (solid lines). The rotation rate is in units of radians per second (rad.s$^{-1}$). To compare our results with experiments, we used [ATP], [ADP] and [Pi] of 2 mM, 10 μM and 10 mM, respectively. The crosses in Figure 2 are from experimental measurements [6], and the two solid lines indicate that the $F_{1}$-ATPase motor works at chemomechanical coefficients within the range 0.165 ≤ η ≤ 0.452. The effective ‘ratchet’ drag coefficient, which is 1.07 pN nm s, in this case, is calculated to ensure the rotational rates’ convergence at zero length and to comprise the top and bottom range of the scattered experimental data. This value is found to be close to the drag coefficient of driving a 1-μm actin filament at one end. Yasuda et al. [6] claimed that the $F_{1}$-ATPase-actin motor system would have a thermodynamic coefficient of about 1.0, or may at least have a coefficient of ~ 0.8. However, our theoretical investigation indicates that this may not be the case. The theoretical rotational rates show that the motor system works with chemomechanical coefficients in the range of η = 0.165 to η = 0.452.

V. CONCLUSION

Our mechanochemical theory is able to predict the operation and dynamics of the ATP-fuelled biomolecular motors, in particular the coupling of ATP hydrolysis reactions to the motions and activities in the presence of spherical or polymeric loads. With more complexity and functions of these bio motors explored [11, 12], the molecular understanding of the ATP-fuelled biomolecular motors are expected to advance dramatically. This will help to control, manipulate and fabricate biomolecular motors, and harness their potential into useful nano/bio-technological applications.

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REFERENCES


