MOLECULAR MOTORS AND FLUCTUATION THEOREM

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ABSTRACT
Biological motors and pumps operate far from global equilibrium and capable of coupling chemical, electrical and mechanical processes. The molecular walker such as biomolecular motor kinesin uses chemical energy provided by the hydrolysis of anedonine triphosphate to step in one direction along a macromolecular track The fluctuation theorem (FT) can quantify the hysteresis observed in the amount of the irreversible work of forward and backward movements of a macromolecule in nonequilibrium regimes. Modeling of motor proteins must take into account the collective behavior that is the energy coupling between the internal biochemical cycle of a macromolecule and its external load, such as random walk. These molecular motors are mechanochemical and stochastic systems, and take part in the cellular metabolism. FT describes how irreversible macroscopic behavior evolves from time-reversible miscroscopic dynamics and how the entropy production can be related to the forward and backward dynamical randomness of the trajectories paths of molecular motors. The thermodynamics of such motors is constrained by the FT, which is valid far from equilibrium and can provide a mathematical expression for the probability that entropy will flow in a direction opposite to that dictated by the second law of thermodynamics for a finite nonequilibrium system in a finite time.

INTRODUCTION

Thermodynamics can assess the behavior of systems at or near thermodynamic equilibrium, systems that are some distance from equilibrium and can return to equilibrium, and systems that are far from equilibrium and constrained by gradients (thermodynamic forces). The systems that are far from global equilibrium are stochastic in nature with varying spatial and time scales. The thermodynamic branch (Fig. 1) designates these three different situations as linear and nonlinear regions. Dissipative structures may emerge in order to reduce or degrade the gradient(s) and if dynamics and/or kinetic conditions are favorable for energy coupling [1,2].

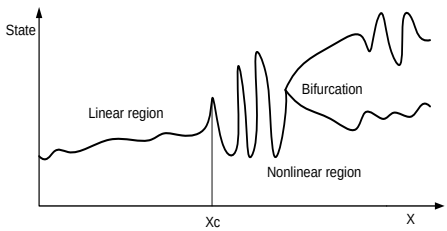

Figure 1. Thermodynamic branch and distance from global equilibrium [8].

In eukaryotes, oxidative phosphorylation occurs in mitochondria, while photophosphorylation occurs in chloroplasts to produce adenosine triphosphate (ATP). Oxidative phosphorylation involves the reduction of O_2 to H_2O with electrons donated by nicotinamide adenine dinucleotide (NADH) and flavin adenine dinucleotide ($FADH_2$) in all aerobic organisms. After carbon fuels are oxidized in the citric acid cycle, electrons with electron-motive force is converted into a proton-motive force. A protein structure called ATP synthase, or the F_oF_1, couples the energy of the proton electrochemical potential gradient to ATP synthesis, or the energy of ATP hydrolysis in F_1 to the proton translocation through the subunit F_o rotation (Fig. 2) [3].

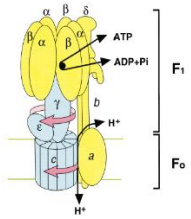

Figure 2. ATP-synthase (ATPase) couples ATP production to proton electrochemical potential gradient created by the respiration. ATP-synthase can either produce ATP driven by a proton gradient or hydrolyze ATP to pump protons, depending on conditions [2,3].

Energy production, storage, and conversion to maintain the nonequilibrium state form the basis for bioenergetics [2]. The hydrolysis of ATP is coupled [1] to synthesizing protein molecules, transporting ions and substrates, producing mechanical work, and other metabolic activity. ATP hydrolysis depends on the ratio of [ATP]/[ADP][P_i]. The need for energy by the cell regulates the tricarboxylic acid cycle, which acts in concert with the electron transfer chain and the ATPase to produce ATP in the inner mitochondrial membrane. The cell has limited amounts of ATP, adenosine diphosphate (ADP), and adenosine monophosphate (AMP). When ADP levels are higher than the levels of ATP, the cell needs energy, and hence NADH is oxidized rapidly and the tricarboxylic acid cycle is accelerated. When the ATP level is higher than the levels of ADP, the cell has the energy needed, hence, the electron transport chain slows down [2,3].

Molecular motors and pumps operate far from global equilibrium and convert the chemical energy released from the hydrolysis of ATP into mechanical work by coupling to a heat reservoir, a work reservoir, and particle reservoirs for ATP, adenosine diphosphate ADP, and inorganic phosphate (Pi). The ATPase usually synthesises ATP but can also work as a motor by hydrolyzing ATP [4]. Each motor state represents an ensemble of molecular conformations of the

enzyme that are thermally equilibrated. The states of enzyme together with the possible transitions between neighboring states create a dynamic network, which may be described by a continuous-time Markov process (or master equation) on this network [5]. Molecular motors, in nonequilibrium steady states, have net flows and require a continuous input of material, energy, and information to maintain their self-organized steady state as they continuously dissipate net energy [2,6].

Many motor proteins can generate directional net movement, including the muscle motor protein myosin and the kinesin proteins of microtubules [4,5]. Muscle fibers lengthen and contract in small volume changes to perform mechanical work as they utilize chemical energy released by the hydrolysis of ATP.

The fluctuation theorem (FT) relates the probability $p(\sigma_\tau)$ of observing a phase-space trajectory with entropy production rate of σ_τ over time interval τ, to that of observing a trajectory with entropy production rate of $-\sigma_\tau$

$$\frac{p(\sigma_\tau)}{p(-\sigma_\tau)} = \exp(\tau\sigma_\tau / k_B) \qquad (1)$$

where k_B is the Boltzmann constant. This result describes how the probability of violations of the second law of thermodynamics becomes exponentially small as τ or the system size increases. FT relates the work along nonequilibrium trajectories to the thermodynamic free energy differences, and applicable to single molecule force measurements [3,7,8].

This review briefly discusses the latest state of art developments on molecular motors and their operations between back and forth movements with the FT, which may provide a new insight of describing them.

MOLECULAR MOTORS

The molecular walker such as biomolecular motor kinesin uses chemical energy provided by the hydrolysis of (ATP) to step in one direction along a macromolecular track (Fig. 3) with the power stroke causing of dissociation of one head from the binding site and moving to the next available binding site. Even at chemical equilibrium (affinity = 0), due to the thermal noise the molecular motor continues to step forth and back along a macromolecular track as well as keeps catalyzing back and forth conversions of ATP and ADP plus P_i. Equilibrium of molecular motor is a dynamic state in which every forward motion is cancelled with the microscopic reverse of that motion [9].

$$\mathrm{ATP}+\mathrm{E_f} \underset{k_{-Tf}}{\overset{k_{+Tf}}{\rightleftarrows}} \mathrm{E(ATP \leftrightarrow ADP{\bullet}P_i)} \underset{k_{+Df}}{\overset{k_{-Df}}{\rightleftarrows}} \mathrm{E_f}+\mathrm{ADP}+\mathrm{P_i} \quad \text{front}$$

$$\mathrm{ATP}+\mathrm{E_b} \underset{k_{-Tb}}{\overset{k_{+Tb}}{\rightleftarrows}} \mathrm{E(ATP \leftrightarrow ADP{\bullet}P_i)} \underset{k_{+Db}}{\overset{k_{-Db}}{\rightleftarrows}} \mathrm{E_b}+\mathrm{ADP}+\mathrm{P_i} \quad \text{back}$$

Figure 3. Molecular walker with conversions of ATP and ADP plus P_i during back and fort actions [9].

Each head acts as an enzyme to catalyze the reaction between ATP and ADP + P_i. Using Michaelis-Menten kinetics overall equilibrium constant for (ATP = ADP + P_i) becomes $K_e = (k_{+ATP}k_{-ADP})/(k_{-ATP}k_{+ADP})$ where k_{ATP} and k_{ADP} are the forward and backward dissociation constants. Also the enzyme bound form E[ATP=ADP P_i] is a common intermediate for back and front heads. The dissociation constants given by k_{+ATP}/k_{-ATP} and $k_{+ADP}k_{-ADP}$ must be the same for front and back heads. These relationships between the rate constants are the thermodynamic constraints of the principle of microscopic reversibility. However, the ratios of the on and off rate constants for ATP and ADP $s_i = k_{-ATP}/k_{-ATP}$ at frond and back would not be the same; for example if $s_b/s_f >>1$ the binding/release of ATP would be much faster than binding/release of ADP at the back head and vice versa. This change would be possibly triggered by allosteric feedback initiated by a coupling mechanism represented by strain between the neck linker and the ATPase active site. Therefore, if excess levels of ATP exists then the reaction favors by mass action the hydrolysis of ATP and forward movement. If $s_b/s_f < 1$ then movement is backward where ATP is synthesized by the existence of strong force (load) [9]. A single molecule turnover time that is the time for one enzyme molecule to complete a reaction cycle fluctuates randomly; their effects average to zero over a long period of time or for a large number of molecules [10]. Their motion is unidirectional on average and stops at the thermodynamic equilibrium.

An individual molecular motor (i.e. Brownian motor) may be mechanically equilibrated and serves between chemical and physical reservoirs that may be far from equilibrium with one another. This understanding may help design molecular machines [9]. Motor proteins are enzyme catalysts that dramatically accelerate the rate of hydrolysis to repeat a cyclical sequence capable of carrying out useful functions or a load. Recent advancements in single-molecule experimental techniques and the results of structural genomic projects may be very helpful in understanding protein functions and enzyme kinetics on the molecular scale [11].

Motor molecules play a key role in muscular contraction, cell division, and cell transport. For example, for the transient response of muscles, the fastest characteristic times of the motors are in the range of miliseconds. An enzyme or molecular motor stochastically undergoes transitions from one state to another creating either rotary or directional action. In such a transition, a chemical reaction may be involved like hydrolysis which transforms one molecule ATP to ADP and a phosphate [12]. These three molecular species are externally maintained at nonequilibrium conditions thereby providing a source of chemical energy (work) to the system. In each transition, this work will be transformed into mechanical work, dissipated heat, or changes in the internal energy [4,5].

Molecular motors, over the course of their enzymatic cycle, perform work, as they move along a track distance Δx against a constant force F. In some motor models, enzymatic mechanisms explicitly are different from the work related mechanisms; for example, in the Huxley-Hill model motor force is generated within the biochemical step and work is subsequently performed when a motor relaxes within the potential well of a biochemical state [8]. According to fluctuating thermal ratchet model, motor force is generated when a ratchet potential is switched on and work is subsequently performed when a motor relaxes [11,13]. On the other hand, some recent studies support a chemical motor model in which reaction and space coordinates are intimately linked. Force is generated and/or work is performed with a thermally activated biochemical transition. For example, a motor structural change induced by ligand binding or by other

effects might directly perform work. Most chemical motor models assume that it is the external work ($W_{ext} = F\Delta x$), i.e. in moving the track, that is coupled to the free energy for that step [8]. Internal work, on the other hand may involve pulling out compliant elements in the motor, and is performed in stretching these internal elastic elements that are coupled to free energy ΔG. Motor enzymes, like myosin and kinesin, move along a track while catalyzing a hydrolysis reaction of ATP are self-consistent mechanochemical systems, in which the reaction mechanisms start and end with free enzyme, while the free enzyme is binded with the substrates and unbinded with products in some random order. The myosin protein uses the chemical energy released by the hydrolysis of ATP to create directed mechanical motion. All the myosin motor proteins share the same biochemical reaction pathway when hydrolyzing ATP. They operate far from equilibrium, dissipate energy continuously, and make transitions between steady states. The thermodynamic driving force of an enzymatic cycle $\Delta\mu$, can be extracted by the nonequilibrium turnover time traces of single enzyme molecules in living cells that might be measurable experimentally [3,8,11].

Modeling of motor proteins, such as kinesin and myosin-5, [13-15] must take into account the collective behavior that is the energy coupling between the internal biochemical cycle of a macromolecule and its external load such as random walk. These molecular motors are mechanochemical and stochastic systems, and take part in the cellular metabolism under far from equilibrium conditions. Molecular motors, over the course of their enzymatic cycle, perform work, as they move along a track a distance Δx against a constant force F. There are several models for explaining the relationship between a motor's enzymatic mechanisms and its mechanisms for work production [6,8,13]. Most chemical motor models assume that it is the external work ($W_{ext} = F\Delta x$), i.e. in moving the track is coupled to free energy ΔG; like myosin and kinesin, move along a track while catalyzing a hydrolysis reaction of ATP are self-consistent mechanochemical systems [8,13,15]. The driving force for a motor protein comes from the hydrolysis of ATP characterized by a two-state Markov process: $\mathrm{ATP+H_2O} \underset{k_b}{\overset{k_f}{\rightleftharpoons}} \mathrm{ADP+P_i}$, $J_{rf} / J_{rb} = \exp\left(A/RT\right)$, $A = -\sum_i \nu_i \mu_i$, $J_{rf} = k_f[\mathrm{ATP}]$, $J_{rb} = k_b[\mathrm{ADP}][Pi]$. Here A is the thermodynamic driving force called the affinity, J_{rf} and J_{rb} are the forward and backward reaction rates, respectively, J_r is the net reaction rate, and ν_i is the stoichiometric coefficient, which is positive for product and negative for reactants. For a reaction at isobaric and isothermal conditions, the affinity characterizes the distance from equilibrium [8].

In some systems, experiments have verified an overall type of reversibility, such as ATP-synthase which can either produce ATP driven by a proton gradient or hydrolyze ATP to pump protons, depending on conditions. Symmetry could be applied to a system which couples binding and catalysis, when the entire system is analyzed. The chemical potential difference $\Delta\mu$ is a generalized force and measures the free energy change per consumed fuel molecule by the hydrolysis of ATP: $\mathrm{ATP} \rightleftharpoons \mathrm{ADP+Pi}$ $\Delta\mu = \mu_{ATP} - \mu_{ADP} - \mu_P$. The dissipation Ψ for representative motor action is

$$\Psi = f_{ext} v + J\Delta\mu \geq 0 \qquad (2)$$

Eq. (2) identifies the independent fluxes and forces [3]. These forces cause motion and ATP consumption characterized by fluxes (currents) that are average velocity $v(f_{ext},\Delta\mu)$ and average rate of ATP hydrolysis $J(f_{ext},\Delta\mu)$. Molecular motors mostly operate far from equilibrium ($\Delta\mu \sim 10k_BT$) and the fluxes are not linearly dependent on the forces. However, if a linear flux force relationships hold due to multi inflection points [16]: $v = L_{11} f_{ext} + L_{12}\Delta\mu$; $J = L_{21} f_{ext} + L_{22}\Delta\mu$. Here L_{11} and L_{22} are the mobility coefficients, while L_{12} and L_{21} (Onsgaer's relation holds $L_{12} = L_{21}$) are the mechano-chemical coupling coefficients for polar filaments. Inequality in Eq. (2) will be satisfied if $L_{ii} > 0$ and $L_{22}\, L_{11} - L_{12}\, L_{21} > 0$. Thermal equilibrium ($\Delta\mu = 0, f_{ext} = 0$) represents a singular point. When $f_{ext} v < 0$, work is performed by the motor and the chemical work is the driving process, while $J\Delta\mu < 0$ requires that chemical energy is generated and the mechanical work is the driving process. When $f_{ext} v > 0$ and $J\Delta\mu > 0$, there is no single driving process nor driven process and dissipation is in the form of heat in the thermal bath. This may be passive system [8].

The efficiency of energy coupling η is the ratio of output and input powers in the representatrive dissipation equation $\Psi = f_{ext} v + J\Delta\mu \geq 0 = \text{output power} + \text{input power} \geq 0$, and the efficiency becomes: $\eta = (f_{ext} X_p)/(J\Delta\mu)$. In terms of the normalized flow ratio (j) and the normalized force ratio (x), the energy coupling efficiency becomes

$$\eta = jx = -(x+q)/(q+1/x) \qquad (3)$$

where $q = L_{12}/(L_{11}L_{22})^{1/2}$ $j = -J_p/(J_o Z)$, $x = X_p Z / X_o$, and $Z = \sqrt{L_p / L_o}$ [2,3]. Thus, the efficiency depends on the force ratio x and the degree of coupling q. The energy coupling efficiency is zero when either flow or force is zero. Therefore, at intermediate values of them, the efficiency passes through an optimum (maximum) $\eta_{opt} = \left(q/\left(1+\sqrt{1-q^2}\right)\right)^2$. Here, q represents a lump sum quantity for the various individual degrees of coupling of different processes [3,8].

FLUCTUATION THEORY

The thermodynamic driving force of an enzymatic cycle $\Delta\mu$, can be extracted by the nonequilibrium turnover time traces of single enzyme molecules that might be measurable experimentally [17]. From chemical master equations under nonequilibrium steady state, the ratio between the probability of M forward turnovers $P(dn_t = M)$ and that of M backward turnovers $P(dn_t = -M)$ is

$$\frac{P(dn_t = M)}{P(dn_t = -M)} = \exp\left(\frac{\Delta\mu}{k_B T} M\right), \qquad (4)$$

where M is a positive integer [8,17]. Eq. (4) is the consequence of microscopic reversibility and general as long as the enzyme completes a full cycle, even when the enzyme molecules exhibit more complex kinetic pathways [17]. By introducing internal conformational states to the Brownian particle and to coupling the hydrolysis of ATP with the motor protein movement leads to the following reaction-diffusion

system for the movement of a Brownian particle with internal structures and dynamics

$$\frac{\partial P(x,n,t)}{\partial t} = D\frac{\partial^2 P(x,n,t)}{\partial x^2} - \frac{\partial}{\partial x}\left(\frac{F(x)}{\beta}P(x,n,t)\right) \quad (5)$$
$$-k_{fnk}(x)P(x,n,t) + k_{bkn}(x)P(x,k,t)$$

where $P(x,n,t)$ is the probability of a motor protein with internal state n and external position x, and k_{fnk} is the transition rate constant from internal state n to state k when the protein is located at x. The states n and k, such as attached and detached states, driven by the ATP hydrolysis leads to a biased motion of the motor protein, in which the chemical energy of the hydrolysis of ATP is converted to the mechanical motion of the motor protein [8,17-19]. In general, the stationary solution of Eq. (5) will be far from equilibrium steady state with positive entropy production and heat generation. For an arbitrarily large ensemble of experiments from some initial time $t = 0$, consequence of the fluctuation theorem is that an ensemble average of the entropy production cannot be negative for any value of the averaging time t: $\langle \sigma_t \rangle \geq 0$. This inequality is called the second law inequality. It can be proved for systems with time dependent fields of arbitrary magnitude and time dependence. However, it does not imply that the ensemble averaged entropy production is nonnegative at all times. The fluctuation theorems are related to the entropy production and valid for systems far away from thermodynamic equilibrium

Assume that a finite system is in contact with a heat bath at constant temperature and driven away from thermodynamic equilibrium by some external time-dependent force. The FT relates to the probability distributions of the time-averaged irreversible entropy production $\bar{\sigma}$. The theorem states that, in systems away from equilibrium over a finite time t, the ratio between the probability that $\bar{\sigma}$ takes on a value A and the probability that it takes the opposite value, $-A$, will be exponential in At. Mathematically, the fluctuation theorem is expressed as

$$\frac{P(\bar{\sigma}_t = A)}{P(\bar{\sigma}_t = -A)} = e^{At} \quad (6)$$

The system is finite and coupled to a set of baths, each characterized by a constant intensive parameter. The dynamics are required to be stochastic, Markovian, and microscopically reversible. The probabilities of the time-reversed paths decay faster than the probabilities of the paths themselves and the thermodynamic entropy production arises from the breaking of the time-reversal symmetry of the dynamical randomness.

The transient FT describes how irreversible macroscopic behavior evolves from time-reversible miscroscopic dynamics as either the observation time or the system size increases. The transient FT also shows how the entropy production can be related to the forward and backward dynamical randomness of the trajectories or paths of systems as characterized by the entropies per unit time [8]. For example, the Crooks fluctuation theorem was used to estimate the free energy difference associated to the unfolding of a RNA molecule [7]. The thermal bath allows macromolecules to exchange energy with the molecules of the solvent through the breakage of weak molecular bonds that trigger the relevant conformational changes. The amount of energies involved in single macromolecules is small enough for thermal fluctuations over timescales to be relevant in many molecular processes [9]. In thermodynamics of small systems, a control parameter may define the system's state [13,15]; for example, a motor molecule can be described by an internal configuration $\{x_i\}$ and a control parameter x (there can be finite number of control parameters), then $u(\{x_i\},x)$ is the internal energy of the system. Upon variation of the control parameter x, energy conservation yields

$$du = \sum_i \left[\left(\frac{\partial u}{\partial x_i}\right)_x + \left(\frac{\partial u}{\partial x}\right)_{\{x_i\}} dx\right] = \delta Q + \delta W \quad (7)$$

The total work done on the system is $W = \int_0^{x_f} F(\{xi\},x)dx$, where x_f is the perturbation for a time t_f, and $F(\{x_i\},x)$ is the fluctuating force acting on the molecule $F(\{x_i\},x) = (\partial u/\partial x)_{\{x_i\}}$. A quantity that characterizes the stochastic nonequilibrium process is the probability distribution of work values $P(W)$ obtained along different trajectories. The average work over all trajectories $\langle W \rangle = \int WP(W)dW$ is larger than the reversible work and equal to the free-energy difference ΔG between the equilibrium states defined at $x = x_f$ and $x = 0$. If we define the dissipated work along a given trajectory as $W_{dis} = W - \Delta G$, second law can be written as, $W_{dis} \geq 0$. Under the assumption of microscopic reversibility (detailed balance), fluctuation theorems assert relations between the entropy production along a given forward and backward processes by [20,21]

$$\frac{P_f(W)}{P_b(-W)} = \exp\left(\frac{W_{dis}}{k_B T}\right) \quad (8)$$

where $P_f(W)$ and $P_b(-W)$ are the work distributions along the forward and backward processes, respectively. Eq. (8) indicates that a steady-state system is more likely to deliver heat to the bath (W is positive) than it is to absorb an equal quantity of heat from the bath (W is negative) and hold for any finite time [20]. Nonequilibrium steady state systems always dissipate heat on average.

The Crooks fluctuation theorem is used to estimate the free energy difference associated with the unfolding of a RNA molecule. In the experiment, a single molecule was repeatedly folded and unfolded. The periodic folding of a single molecule is analogous to the cycles of the kinesin displacement [18]. The Crooks theorem is also applied to the linear motor kinesin cycle [18]. Distinct forward and backward trajectories may have different probability weights if the system is out of equilibrium. For example, the probability for a driven Brownian particle having a trajectory from point 'a to b' is different from that having the same reverse trajectory from 'b to a.' The entropy production arises from the breaking of the time-reversal symmetry in the probability distribution of the statistical description of the nonequilibrium steady state [17].

The reaction rate of a single enzyme molecule fluctuates, which is a general feature of enzymes. A single molecule turnover time, which is the time for one enzyme molecule to complete a reaction cycle, also fluctuates. Since these fluctuations are random, their effects average to zero over a long period of time or for a large number of molecules. Kinesin is a large protein which can attach to a load on one

end and has two heads on the other end. It performs an asymmetric hand on hand walk along a microtubule dragging the load against an external force F and the viscous drag from the environment. Each step in this walk corresponds to a cycle, in which kinesin converts chemical energy released by the hydrolysis of one ATP molecule into useful work. The amount of energy of the hydrolysis of one ATP molecule is around $25k_BT$, where k_B is the Boltzmann constant and T is the bath (environment) temperature. The probability of a successful forward step over that of backward step is [18,21].

$$\frac{p_f}{p_r} = \exp\left(\frac{\Delta l}{2k_BT}[F_{st} - F]\right) \tag{9}$$

where $F_{st} \approx 7$ pN is the stalling force and F is the external force. Eq. (9) shows that the maximum work kinesin can do against the external force is $F_{st}\Delta l \approx 13.3k_BT$, which is close to half of the input energy of $25k_BT$.

To obtain the free energy change associated with the one step from observable data, the Crooks FT can be used [18]. Assume that the macroscopic initial and final states 1 and 2 are the initial and final states, respectively in the kinesin cycle, and the external parameter λ measures the progress of the molecule from one pair of docking sites to the next. A backward step implies that the forward work ($W = F\Delta l$) is reversed. For the free energy G, the Clausius inequality implies $W \leq -\Delta G$. Initially the system is at state 1. If the P_f is the probability that the system ends up in state 2, giving out work W and the P_b is the probability that the system, now starting from state 2, ends up in state 1 giving out work $-W$ when the evolution of λ is reversed. The Crooks FT states that

$$\frac{p_f}{p_b} = \exp\left(\frac{-1}{k_BT}[\Delta G + W]\right). \tag{10}$$

The probability ratio given in Eq. (10) implies that $\Delta G = (-\Delta l/2)[F_{st} + F]$ where $-\Delta G$ is the maximum work kinesin performs at constant temperature [8,18]. Ideally, all the energy available to the kinesin at the start of the cycle is about ($2F_{st}\,\Delta l = 26.6k_BT$) and is dissipated or goes to into the reversible work ($-\Delta G$). For isothermal docking we have $S_{\text{free}} - S_{\text{dock}} = q_{\text{dock}}/T$ where S_{free} and S_{dock} are the entropies of the free head and docked states, respectively. Part of the available work is left to be dissipated as heat by opposing the viscous drag or as excess kinetic energy to be absorbed by the docking site. Besides that, the cycle may fail, with the kinesin stepping backwards rather than forward; the actual average work is $\langle W\rangle = F\langle\Delta l\rangle$, where $\langle\Delta l\rangle = \delta\tanh\left(\frac{\Delta l}{4k_BT}[F_{st} - F]\right)$ is the average displacement [3,8].

CONCLUSIONS

FT applies to fluctuations far from equilibrium and requires knowledge of the initial distribution of molecular states, all time evolved final states at time t, and assumption of time reversal symmetry (all the equations of motion for either classical or quantum dynamics are in fact time reversible). One important implication from the FT is that molecular motors or even mitochondria in a cell, will spend part of their time actually running in 'reverse' and that these motors are able to generate work by taking heat from the environment. This is possible because there exists a symmetry relation in the work fluctuations associated with the forward and reverse changes as it is driven away from thermal equilibrium by the environment. FT may help understanding the molecular machines and hence design and manufacture of nanomotors and nanopumps by estimating rate constants or alternative reaction schemes.

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