

Thermodynamics of Gradient Driven Transport: Application to Single-Particle Tracking

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Single-particle tracking techniques allow measurements of the trajectories of individual particles (and sometimes single molecules) as they move. Here, we show, in accord with an equality derived by Bier et al. (*Phys. Rev. E* 1999, 59, 6422–6432), that even in the presence of gradients of pressure, temperature, electric field strength, and so forth, the ratio of the probabilities for forward and backward trajectories between any two points is given by the exponential of the difference between the particle's free energy at the start and end points of the trajectory, that is, $P(\mathbf{x}_a \rightarrow \mathbf{x}_b, \Delta t) = P(\mathbf{x}_b \rightarrow \mathbf{x}_a, \Delta t) \exp(-\Delta G/k_B T)$, where $\Delta G = G(\mathbf{x}_b) - G(\mathbf{x}_a)$. Thus, experimental approaches based on single-particle tracking can be used to map the free-energy landscape for transport of a particle without reference to whether the overall system is, or is not, in thermodynamic equilibrium.

1. Introduction

The Gibbs free energy of a particle or macromolecule in solution is a well-defined equilibrium property that depends on the extensive parameters of the particle, its molecular volume, dipole moment, entropy, and so forth, and on the intensive parameters of the solution, the temperature, pressure, electric field intensity, and so forth. The differential of the free energy dG , the change due to a very small change in the intensive parameters, can be expressed as the sum of products¹

$$dG = \sum_i \theta_i dF_i \quad (1)$$

where the θ_i and F_i are the canonically conjugate extensive and intensive thermodynamic parameters of the system, respectively. Examples include the familiar pairs of thermodynamic variables SdT , where S is the particle entropy and T is the temperature, and Vdp , where V is the molecular volume and p is the pressure, as well as pairs of mechanical parameters $md(g h)$, where m is the effective mass, g is the acceleration due to gravity, and h is the height, and $l d\mathbf{f}$, where l is length in the direction of an external force \mathbf{f} , and pairs of electromagnetic parameters $\mu d\mathbf{E}$, where μ is the molecular polarization and \mathbf{E} is the electric field, and $q d\phi$ where q is the charge and ϕ is the electrical potential. The free-energy change of a particle $\Delta G_{1,2} = G(\mathbf{F}(2)) - G(\mathbf{F}(1))$ and $\Delta G_{2,3} = G(\mathbf{F}(3)) - G(\mathbf{F}(2))$ due to reversible but noninfinitesimal changes of the intensive parameters of the system (Figure 1a) $\mathbf{F}(1) \rightarrow \mathbf{F}(2) \rightarrow \mathbf{F}(3)$ (Figure 1a) is, in general, a nonlinear function of the intensive parameters, that is, $G(\mathbf{F}(2)) - G(\mathbf{F}(1)) \neq G(\mathbf{F}(3)) - G(\mathbf{F}(2))$ even if $\mathbf{F}(2) - \mathbf{F}(1) = \mathbf{F}(3) - \mathbf{F}(2)$.

In the presence of a spatial gradient ∇F of one of the intensive parameters (e.g., temperature, electric field strength, pressure, and so forth), a particle will move, tending to seek the minimum of its free energy. In Figure 1b, a diffusive trajectory for a particle is shown, where the particle is at some position \mathbf{x}_a at time t_i , and by Brownian motion, it arrives at the position \mathbf{x}_b at

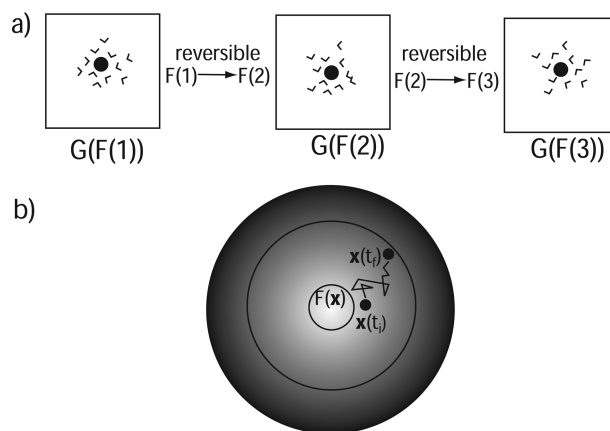


Figure 1. (a) Schematic illustration of a reversible (quasi-static) process in which an intensive parameter is changed, $F(1) \rightarrow F(2) \rightarrow F(3)$, leading to the change in the free energy of a particle $G[F(1)] \rightarrow G[F(2)] \rightarrow G[F(3)]$. For large changes of the intensive parameter, $G[F]$ is a nonlinear function of F , but the overall process is well described by equilibrium thermodynamics. (b) Illustration of a single-particle trajectory in a gradient ∇F . We prove theoretically and show experimentally that the ratio of the probabilities for forward and backward trajectories is the exponential of the free-energy difference at the end points, $P[\mathbf{x}_a \rightarrow \mathbf{x}_b, \Delta t] = P[\mathbf{x}_b \rightarrow \mathbf{x}_a, \Delta t] \exp(-\Delta G/k_B T)$.

some time t_f , where the vector $\mathbf{x} = (x, y)$ specifies the position in a two-dimensional Cartesian coordinate system. Onsager² postulated that the average current in an ensemble of particles is related to the gradient of the “thermodynamic potential” by a linear relation, $RJ = \nabla F$. Onsager and Machlup³ extended this picture to include fluctuations due to thermal noise in terms of the overdamped Langevin equation $R\dot{\alpha} = \nabla F + \varepsilon(t)$, where $\dot{\alpha}$ is the rate of change of some thermodynamic variable.

2. Results and Discussion

2.1. Onsager–Machlup Theory for Single-Particle Trajectories. Here, following Onsager and Machlup, we propose that the local velocity $\dot{\mathbf{x}}$ for a single particle can be related to the equilibrium free energy by a similar Langevin equation, reflecting a locally linear relation between the velocity and the thermodynamic force that caused it

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$$\dot{\mathbf{x}} = -\gamma^{-1} \frac{\partial G}{\partial F} \nabla F + \sqrt{2D} \varepsilon(t) \quad (2)$$

where γ is the coefficient of viscous friction and where the term $\varepsilon(t)$ models thermal noise due to collisions between the particle of interest and the solvent. The transport coefficients γ and D both may depend on F and hence on position. The statistical properties of each component of the vector $\varepsilon(t)$ are taken to be independent of one another and to be given by $\langle \varepsilon(t) \rangle = 0$ and $\langle \varepsilon(t) \varepsilon(t') \rangle = \delta(t - t')$, describing so-called “white” or uncorrelated noise.

For small gradients and displacements such that the average velocity (v) = $-\gamma^{-1}(\partial G/\partial F)\nabla F$ is approximately constant, the stochastic eq 2 is equivalent to the equation for diffusion with drift^{4,5} with constant transport and diffusion coefficients and is easily solved for the probability to find the particle at any \mathbf{x}_b at time t_f given that it started at position \mathbf{x}_a at time t_i , $P(\mathbf{x}_b, t_f | \mathbf{x}_a, t_i)$. A heuristic example for motion of a single particle in a gravitational potential has been given,⁶ where the conjugate variables are m and (gh) and where $\gamma^{-1}(\partial G/\partial(gh))\nabla(gh)$ is obviously constant over a very large range. Duhr and Braun⁷ have shown a similar approach to hold in a small temperature gradient, where the average velocity is linearly proportional to the gradient and where the individual particle trajectories obey the simple expression for diffusion with constant drift. There, the conjugate parameters were S and T .

In general, for large gradients or long trajectories, where the function $\gamma^{-1}(\partial G/\partial F)\nabla F$ is not constant, the diffusion with drift approximation is not valid. Breakdown of the diffusion with drift approach is reflected in a nonlinear dependence of the average velocity on the magnitude of the gradient⁸ and the failure of the probability $P(\mathbf{x}_b, t_f | \mathbf{x}_a, t_i)$ for single-particle trajectories to be correctly predicted by a Boltzmann-like equation.⁸ Using the path integral approach of Onsager and Machlup,³ however, Bier et al.⁹ showed that the ratio between the conditional probability to go to any position \mathbf{x}_b in a time t , given the starting position \mathbf{x}_a , to the probability to go to \mathbf{x}_a in time t , given the starting position \mathbf{x}_b , is both path- and time-independent, irrespective of the shape of the function $\gamma^{-1}(\partial G/\partial F)\nabla F$ in the interval $\Delta \mathbf{x} = \mathbf{x}_a - \mathbf{x}_b$. This result can be simply derived.

For discretized time $t_j = t_0 + j\delta t$, with $j = 1, 2, 3, \dots$, the $\varepsilon(t_j)$ are independent and Gaussian distributed with a zero average and a standard deviation of $1/\sqrt{t}$. The probability that a particular sequence of “Brownian kicks” $\varepsilon(t_1), \varepsilon(t_2), \dots, \varepsilon(t_n)$ is realized is proportional to $\prod_{j=1}^n \exp[-(1/2)\varepsilon^2(t_j)\delta t] = \exp[-(1/2)\sum_{j=1}^n \varepsilon^2(t_j)\delta t]$. Going to the limit of continuous time that runs from $t = t_i$ to t_f , the sum becomes an integral, and we find

$$P[\varepsilon(t)] \propto \exp\left(-\frac{1}{2} \int_{t_i}^{t_f} dt \varepsilon^2(t)\right) \quad (3)$$

The Langevin eq 2 maps between position (\mathbf{x}) space and noise (ε) space; therefore, substituting the algebraic expression for $\varepsilon^2(t)$ from eq 2, we find the probability for a particle starting at position \mathbf{x}_a at time t_i to end at position \mathbf{x}_b at time t_f having followed the specific trajectory $\mathbf{x}(t)$ to be

$$P[\mathbf{x}_b, t_f | \mathbf{x}(t), t | \mathbf{x}_a, t_i] \propto J(\mathbf{x}) \exp\left[-\frac{1}{2} \int_{t_i}^{t_f} dt \left(\frac{\dot{\mathbf{x}} + \gamma^{-1} \frac{\partial G}{\partial F} \nabla F}{\sqrt{2D}}\right)^2\right] \quad (4)$$

where $J(\mathbf{x}(t)) = d\varepsilon(t)/d\mathbf{x}(t)$ is the Jacobian matrix by which we transform from a trajectory in noise space $\varepsilon(t)$ to a trajectory in position space $\mathbf{x}(t)$. Clearly, $P[\mathbf{x}_b, t_f | \mathbf{x}(t), t | \mathbf{x}_a, t_i]$ is time- and path-dependent. Remarkably, however, even for large gradients and arbitrarily long trajectories, the ratio

$$\begin{aligned} \frac{P[\mathbf{x}_b, t_f | \mathbf{x}(t), t | \mathbf{x}_a, t_i]}{P[\mathbf{x}_a, t_f | \mathbf{x}(t_f - t), t | \mathbf{x}_b, t_i]} &= \exp\left(-\int_{t_i}^{t_f} dt \frac{\dot{\mathbf{x}} \frac{\partial G}{\partial F} \nabla F}{\gamma D}\right) \\ &= \exp\left(-\int_{\mathbf{x}_a}^{\mathbf{x}_b} d\mathbf{x} \frac{\frac{\partial G}{\partial F} \nabla F}{\gamma D}\right) \end{aligned} \quad (5)$$

is time-independent since the Jacobian does not depend on direction. Further, the ratio is also approximately path-independent if the local fluctuation dissipation relation $\gamma(\mathbf{x})D(\mathbf{x}) = k_B[T^0 + \delta T(\mathbf{x})]$ holds and if the deviation $\delta T(\mathbf{x})$ from the reference temperature T^0 is small $\delta T(\mathbf{x}) \ll T^0$ in the interval of \mathbf{x}_a to \mathbf{x}_b . The expression in eq 5 then evaluates to

$$\frac{P[\mathbf{x}_a \rightarrow \mathbf{x}_b, \Delta t]}{P[\mathbf{x}_b \rightarrow \mathbf{x}_a, \Delta t]} \approx \exp\left(\frac{-\Delta G}{k_B T^0}\right) \quad (6)$$

where $\Delta G = \{G[F(\mathbf{x}_b)] - G[F(\mathbf{x}_a)]\}$. Bier’s relation, eq 6, (which is an identity for an isothermal system)⁹ is closely related to the generalized fluctuation dissipation theorem of Bochkov and Kuzovlev,¹⁰ who showed that the ratio of the probabilities for a forward and a time reverse trajectory is the exponential of the internal energy change in the system due to the forward trajectory. As noted by Bier et al.,⁹ eq 6 can be derived for isothermal systems directly from microscopic reversibility and knowledge that at equilibrium, the densities are given by a Boltzmann distribution. The derivation given by Bier et al.⁹ from the Langevin equation of motion does not use the densities at all and thus makes clear that the relation between the conditional probabilities (eq 6) does not require that the system be in statistical equilibrium. A very nice illustration of some of the properties of stochastic trajectories has been given using analogue electrical circuits.¹¹

The Onsager–Machlup approach used here to derive eq 6 is often stated to be valid only near equilibrium and in the linear regime. Technically, this is true, but it is necessary to carefully understand these requirements since they are not nearly as restrictive as they may, at first thought, seem to be. Onsager and Machlup themselves stated it most clearly, “The essential physical assumption about the irreversible processes is that they are linear, that is, that the fluxes depend linearly on the forces that ‘cause’ them”.³ For the motion of individual particles or single molecules, this is, the requirement of mechanical equilibrium, the sum of the deterministic forces (including the viscous drag force) acting at any instant is approximately 0 and hence where there is no acceleration. This condition is reflected in the equation of motion, eq 2. The overall system need not be close to thermodynamic equilibrium, nor must the gradient be small such that the “force” $(\partial G/\partial F)\nabla F$ is an approximately linear function of the position \mathbf{x} ;^{5,6} therefore, the approach is valid even for “long” trajectories in “large” gradients.

Let us consider what exactly is meant by a “large gradient” in the context of a thermal gradient. Imagine a cube of water, 1 cm on a side, and let the left-hand side be maintained at a temperature of 100 °C (373 K) and the right-hand side be maintained at 0 °C (273 K). Intuitively, we doubtless imagine horrible things happening in the liquid. Most of these are due

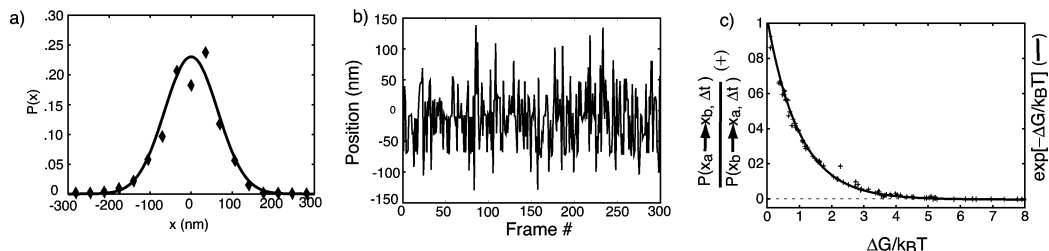


Figure 2. (a) Plot of the relative occupancy for a particle as a function of position averaged over several minutes. These data were used to obtain the trap stiffness ($\kappa = 5 \times 10^{-3}$ pN/nm) and were in good agreement with the value estimated by the laser intensity. (b) Raw data for the stochastic trajectory of a single latex sphere with a diameter of 970 nm (Interfacial Dynamics Corporation) in an optical trap with a trap stiffness determined to be $\kappa = 5 \times 10^{-3}$ pN/nm. The position reflects the center of optical density of the particle, and each frame is $\delta t_{\text{frame}} = 1/30$ s. (c) The ratio of the number of times the particle is in the bin $x_a \pm \delta x$ and then in the bin $x_b \pm \delta x$ j frames later or $\Delta t = j\delta t_{\text{frame}}$ seconds later to the number of times the particle is in the bin $x_b \pm \delta x$ and then in the bin $x_a \pm \delta x$ Δt seconds later, where $x_a < x_b$, plotted against $(\Delta G/k_B T)$, where $\Delta G = [G(x_b) - G(x_a)] = \kappa(x_b^2 - x_a^2)$. Similarly good agreement is obtained also by direct simulation of the Langevin eq 2 by the technique of Gillespie¹⁴ (not shown).

to the absolute magnitudes of the temperatures involved; after all, the left-hand side is boiling, and the right-hand side is freezing. Contrast this with the case of an optical trap where the energy deposition maintains the temperature at the center 1 K higher than the temperature 10 μm away from the center.¹² The maximum gradient, $\sim 10^5$ K/m, is 10 times larger than the gradient in our hypothetical cube of water. However, the temperature difference across a 1 μm particle from the side nearest the center to the side furthest from the center is only 0.1 K, suggesting that the approximation that the local fluctuation dissipation relation, $\gamma(\mathbf{x})D(\mathbf{x}) = k_B[T^0 + \delta T(\mathbf{x})]$, holds is probably very good. Further, for a $\Delta \mathbf{x} = 10 \mu\text{m}$ trajectory, the total change in temperature is at most 1 $^\circ\text{C}$, and therefore, when dropping the δT in comparison to $T^0 \approx 300$ K, the temperature at the center of the trap is appropriate. Thus, eq 6 is a very good approximation even for investigating the effects of “large” thermal gradients $\nabla T = 10^5$ K/m on diffusion of particles in optical traps.

2.2. Experimental Results for Single-Particle Tracking.

Experimental results obtained by single-particle tracking of a latex bead in an optical trap¹³ are shown in Figure 2. For this system, the relevant conjugate parameters include the electric field intensity (F) and the polarizability of the particle (θ). The center of optical density of the particle is determined and binned at each time frame to produce a string of positions and frame numbers (see raw data, Figure 2b). The ratio of the number of occurrences in which the particle is observed in the bin at x_a and at x_b j frames later (with $\Delta t = j(\text{frame interval})$) to the number of occurrences in which the particle is observed in the bin at x_b and at x_a j frames later is plotted versus the free-energy difference at x_a and x_b calculated from $G(x) = kx^2$ (see Figure 2c). The spring constant k is determined by the distribution of the light intensity in the optical trap and by fitting the long-time average occupancy of the spatial bins to the Boltzmann equation (Figure 2a). Both methods give the same result. As seen, the data fit almost perfectly to the exponential dependence predicted by eq 6.

3. Conclusions

The method described here gives a direct recipe for determining the equilibrium free energy $G(\mathbf{F}(r))$ from single-particle trajectories in gradients of the intensive parameters. The derivation is based on the Onsager–Machlup thermodynamic action theory in which we focus on the ratio between the probability for a forward trajectory and the probability for the microscopic reverse of the forward trajectory. The procedure provides a thermodynamic theory for the sign of the velocity of a particle based on the difference of the free energy of the

particle at the two end points of the trajectory. We do not have a thermodynamic theory for the magnitude of the velocity; for this, a model-specific theory incorporating hydrodynamic and kinetic effects will be necessary.

Our approach will be useful not only for in vitro experiments but also in vivo experiments, where the motion of proteins is measured in living cells or their membranes.¹⁵ A particularly interesting question that may be usefully addressed by the approach described here is that of anomalous diffusion of proteins in membranes.¹⁶ Apparent anomalous diffusion can occur either for kinetic reasons (i.e., because the friction and diffusion coefficients depend on position) or for thermodynamic reasons (there are gradients, e.g., of the surface tension in the membrane that influence the free energy of the diffusing protein). By looking at the ratios of forward and backward transitions, one can, by fitting to eq 6, immediately determine the presence of gradients or specific binding sites that are thermodynamically relevant for the protein. If the protein moves between locations with different stabilities (i.e., free energies), the ratio between probabilities for the forward and backward processes will be different than unity. Barriers or space-dependent transport coefficients, however, do not affect the stability of the protein but only the kinetics. In this case, the ratios of probabilities for forward and backward processes will be unity, even if the diffusion is anomalous, that is, where $\langle \mathbf{x}^2 \rangle \neq Dt$ with a constant diffusion coefficient.

Time- and path-independent relations that hold even in the presence of gradients offer great potential for the investigation of the mechanisms of thermodynamic control of transport processes in many systems. An immediate application may be toward analysis of thermophoresis, or the Soret effect,^{7,8,12,17,18} which is directed motion due to a temperature gradient. Also, it will be useful to consider such gradient-driven transport in the context of catalytic nanomotors,¹⁹ particles on which different catalytic processes on different sides of the particle lead to a local pH and/or reduction/oxidation potential gradients in the vicinity of the particle. The particle moves directionally in this local gradient, and therefore, if the orientation of the particle can be externally controlled, for example, by an applied magnetic field, macroscopic-directed motion arises. Both thermodiffusion in 10^5 K/m temperature gradients and directed motion by catalytic nanomotors are examples of processes that are normally described in the context of “far-from-equilibrium” thermodynamics. Consideration of the single-particle trajectories and the simple thermodynamic theory for the ratio of probabilities for forward and microscopic reverse processes provided by application of the Onsager–Machlup theory to eq 2 forces a reconsideration of what is meant by “equilibrium” for single

particles. On the basis of the equation of motion, we argue that the only sensible criterion for equilibrium is the classic criterion for mechanical equilibrium, whether the sum of the deterministic forces and hence the acceleration is zero or not. For molecules or particles in solution under almost any imaginable circumstance, acceleration is negligible on experimentally relevant time scales, and consequently, the particle or molecule can be viewed as a mechanically equilibrated system that nonetheless serves as a conduit for the flow of energy between macroscopic reservoirs that are not in thermodynamic equilibrium with one another. In combination with Brownian motion, and with spatial asymmetry, some of this energy flow can be used to do work on the surroundings, allowing these systems to function as Brownian machines.^{5,20}

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References and Notes

- (1) Alberty, R. A. *Pure Appl. Chem.* **2001**, 72, 1349–1380.
- (2) Onsager, L. *Phys. Rev.* **1931**, 38, 2265–2280.

- (3) Onsager, L.; Machlup, S. *Phys. Rev.* **1953**, 91, 1505–1512.
- (4) Berg, H. *Random Walks in Biology*; Princeton University Press: Princeton, NJ, 1983.
- (5) Astumian, R. D. *Phys. Chem. Chem. Phys.* **2007**, 9, 5067–5083.
- (6) Astumian, R. D. *Am. J. Phys.* **2006**, 74, 683–688.
- (7) Duhr, S.; Braun, D. *Phys. Rev. Lett.* **2006**, 96, 168301.
- (8) Duhr, S.; Braun, D. 2006; arXiv:cond-mat/0609554.
- (9) Bier, M.; Derenyi, I.; Kostur, M.; Astumian, R. D. *Phys. Rev. E* **1999**, 59, 6422–6432.
- (10) Bochkov, G. N.; Kuzovlev, Yu. E. *Physica* **1981**, 106A, 443–479.
- (11) Luchinsky, D. G.; McClintock, P. V. E. *Nature* **1997**, 389, 463–466.
- (12) Duhr, S.; Braun, D. *Proc. Natl. Acad. Sci. U.S.A.* **2006**, 103, 19678–19682.
- (13) Crocker, J. C.; Grier, D. G. *J. Colloid Interface Sci.* **1996**, 179, 298–310.
- (14) Gillespie, D. T. *Am. J. Phys.* **1996**, 64, 225–240.
- (15) Saxon, M. J.; Jacobson, K. *Annu. Rev. Biophys. Biomol. Struct.* **1997**, 25, 373–399.
- (16) Condamin, S.; Tejedor, V.; Voituriez, R.; Benichou, O.; Klafter, J. *Proc. Natl. Acad. Sci. U.S.A.* **2008**, 105, 5675–5680.
- (17) Rasuli, S. N.; Golestanian, R. *Phys. Rev. Lett.* **2008**, 101, 108301.
- (18) Wurger, A. *Phys. Rev. Lett.* **2009**, 102, 078302.
- (19) Wang, J. *ACS Nano* **2009**, 3, 4–9.
- (20) Astumian, R. D.; Hanggi, P. *Phys. Today* **2002**, 55 (11), 33–39.

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