Effect of stochastic gating on channel-facilitated transport of non-interacting and strongly repelling solutes

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Ligand- or voltage-driven stochastic gating—the structural rearrangements by which the channel switches between its open and closed states—is a fundamental property of biological membrane channels. Gating underlies the channel's ability to respond to different stimuli and, therefore, to be functionally regulated by the changing environment. The accepted understanding of the gating effect on the solute flux through the channel is that the mean flux is the product of the flux through the open channel and the probability of finding the channel in the open state. Here, using a diffusion model of channel-facilitated transport, we show that this is true only when the gating is much slower than the dynamics of solute translocation through the channel. If this condition breaks, the mean flux could differ from this simple estimate by orders of magnitude. [<http://dx.doi.org/10.1063/1.4986902>]

I. INTRODUCTION

This paper focuses on diffusive transport of solutes through channels in biological membranes in the presence of stochastic gating, i.e., when the channel gate randomly opens and closes, and, as a result, the channels jumps between open and closed states, as schematically shown in Fig. 1. Although the phenomenon of stochastic gating is known for many decades, there is no quantitative theory of how the gating affects the flux through the channel, especially when the rates of the gating and solute translocation are comparable. A common assumption is that the gating simply reduces the flux by a factor equal to the probability of finding the channel in the open state.¹ The aim of this paper is to fill the gap and to develop an analytical theory of the mean flux through a stochastically gated channel. Our theory shows that the above-mentioned assumption holds only if the gating is much slower than the dynamics of the solute translocation through the channel. In the opposite limiting case of fast gating, the mean flux can be much higher than just the product of the channel open probability and the flux through the open channel.

We study the transport of both non-interacting solutes and solutes that strongly repel each other by assuming that the channel can be either multiply or singly occupied, respectively. Our main results are analytical expressions for the fluxes through multiply and singly occupied channels, given in Eqs. (3.8) and (3.17), which show how these fluxes depend on the channel geometry, the rate constants determining the gate opening and closing, and the solute diffusivities in the channel and in the bulk.

It is worth noting that stochastic gating plays an important role not only in channel-facilitated membrane transport but also in other biological processes, such as gated ligand binding to proteins^{2–10} and migration of small molecules in

proteins, which is controlled by passage through fluctuating bottlenecks.^{11–22} A distinctive feature of stochastic gating is that the gate opens and closes as a result of thermal fluctuations. There is a seemingly similar problem of resonant activation in escape of a diffusing particle from a potential well over a fluctuating barrier.^{23–33} The fundamental difference between the two phenomena is in the nature of the noise that drives the fluctuations: thermal noise in stochastic gating versus non-thermal noise in resonant activation. Escape over a fluctuating barrier when the fluctuations are produced by thermal noise is discussed in Ref. 34. The effects of slow variation in time of parameters (such as concentrations) on the kinetics of chemical reactions have also been analyzed.³⁵

The outline of this paper is as follows. In Sec. II, we discuss a diffusion model of transport through stochastically gated channels as well as the expressions for the fluxes of non-interacting and strongly repelling solutes in the absence of gating. Our main results, the expressions for the fluxes in the presence of stochastic gating, are presented and discussed in Sec. III. Their derivation is given in Secs. IV–VI. The final Sec. VII contains some concluding remarks.

II. MODEL

Consider transport of solutes through a cylindrical channel in a membrane of thickness L , which is equal to the channel length (see Fig. 1). It is assumed that solute molecules are of spherical shape. Then the effective channel radius a is the difference between the channel and molecule radii. It is also assumed that the solute concentration on the left side of the membrane is c , whereas its concentration on the right side is so low that the back flux can be neglected. We study diffusive transport, where the solute molecules may have different diffusivities in the channel (ch) and in the bulk (b), denoted

FIG. 1. Cartoon of a stochastically gated channel in the presence of the passively transported solute molecules (circles).

by D_{ch} and D_b , respectively. Our focus is on how the transport is affected by stochastic gating, assuming that the gate is located at the left end of the channel (see Fig. 1), and the transitions between the open and closed states are Markovian and described by the kinetic scheme,

where α and β are the rate constants. We consider transport through both multiply and singly occupied channels that model transport of non-interacting and strongly repelling solute molecules, respectively. In the rest of this section, we discuss transport through such channels in the absence of gating.

We begin with non-interacting solute molecules. The flux of solute molecules entering the channel from the bulk is given by the product of the bulk solute concentration c and the Hill rate constant, k_H , which describes trapping of diffusing molecules by a perfectly absorbing disk (channel entrance) on the otherwise reflecting flat wall (membrane). When a molecule enters the channel, it either traverses the channel or returns to the bulk through the same end it entered. Let P_{tr}^{ng} be the particle translocation (tr) probability in the case of no gating indicated by the superscript “ ng .” Assuming that the molecules do not interact with each other, even with the hard-core repulsion, one can write the flux through a multiply occupied (mo) channel, denoted by J_{mo}^{ng} , as the product of the influx, $k_H c$, and the translocation probability, P_{tr}^{ng} ,

$$J_{mo}^{ng} = k_H c P_{tr}^{ng}. \quad (2.2)$$

The Hill rate constant³⁶ and the translocation probability³⁷ are given by

$$k_H = 4aD_b \quad (2.3)$$

and

$$P_{tr}^{ng} = \frac{1}{2 + \nu} = \frac{1}{2 + \kappa L / D_{ch}} = \frac{1}{2 + 4LD_b / (\pi a D_{ch})}, \quad (2.4)$$

where the dimensionless parameter ν and the dimensional parameter κ are^{38,39}

$$\nu = \kappa L / D_{ch} = 4LD_b / (\pi a D_{ch}), \quad \kappa = 4D_b / (\pi a). \quad (2.5)$$

Thus, the flux through a multiply occupied non-gated cylindrical channel is

$$J_{mo}^{ng} = \frac{k_H c}{2 + \nu} = \frac{2aD_b c}{1 + 2LD_b / (\pi a D_{ch})}. \quad (2.6)$$

The flux of solute molecules with strong repulsive interaction, modeled by the requirement of channel single occupancy (so), is denoted by J_{so}^{ng} . It can be written as

$$J_{so}^{ng} = P_{tr}^{ng} / \tau^{ng}, \quad (2.7)$$

where τ^{ng} is the mean duration of the empty-occupied cycle of the channel. Denoting the mean lifetimes of the channel in the empty (emp) and occupied (oc) states by τ_{emp}^{ng} and τ_{oc}^{ng} , we can write time τ^{ng} as

$$\tau^{ng} = \tau_{emp}^{ng} + \tau_{oc}^{ng}. \quad (2.8)$$

The two mean lifetimes on the right-hand side of the above equation are

$$\tau_{emp}^{ng} = 1 / (k_H c), \quad \tau_{oc}^{ng} = L / (2\kappa). \quad (2.9)$$

The former expression follows from the single-exponential approximation of the channel survival probability in the empty state, which is the survival probability of a perfectly absorbing disk on the otherwise reflecting wall, $S(t) = e^{-k_H c t} = e^{-t / \tau_{emp}^{ng}}$. The second expression is derived in Ref. 40. Using Eqs. (2.4) and (2.7)–(2.9), we can write the flux through a singly occupied non-gated cylindrical channel as

$$J_{so}^{ng} = \frac{k_H c}{(2 + \nu) [1 + k_H c L / (2\kappa)]} = \frac{J_{mo}^{ng}}{1 + k_H c L / (2\kappa)}. \quad (2.10)$$

This flux, as might be expected, is smaller than its counterpart in the case of non-interacting solute molecules, i.e., multiply occupied channel, J_{mo}^{ng} in Eq. (2.6), characterized by the same parameters.

The aim of this work is to study how stochastic gating affects the fluxes through multiply and singly occupied channels given by Eqs. (2.6) and (2.10).

III. RESULTS

In this section, we present our analytical results that show how the fluxes through multiply and singly occupied channels are affected by stochastic gating. It is convenient to discuss the results in terms of the inverse flux which is the mean time, $T = 1/J$, between successive escapes of the molecules from the channel to the bulk on the right side of the membrane. In the absence of gating, according to Eqs. (2.6) and (2.10), the mean inter-transition times for multiply and singly occupied channels are

$$T_{mo}^{ng} = \frac{1}{J_{mo}^{ng}} = \frac{2 + \nu}{k_H c} \quad (3.1)$$

and

$$T_{so}^{ng} = \frac{1}{J_{so}^{ng}} = (2 + \nu) \left(\frac{1}{k_H c} + \frac{L}{2\kappa} \right). \quad (3.2)$$

As might be expected, the latter time is larger than the former one. As follows from the above equations, the two mean times are related as

$$T_{so}^{ng} = T_{mo}^{ng} + \frac{L}{\kappa} + \frac{L^2}{2D_{ch}}, \quad (3.3)$$

where we have used the fact that $\nu L / \kappa = L^2 / D_{ch}$.

A. Multiply occupied channel

We start with the case of non-interacting solute molecules. The flux through a stochastically gated (sg) multiply occupied channel, denoted by J_{mo}^{sg} , is given by Eq. (2.2) modified in two ways. First, the flux J_{mo}^{sg} should be multiplied by the probability of finding the channel open (op), P_{op} , because solutes can enter the channel only when the gate is open. Second, the transition probability P_{tr}^{ng} should be replaced by its counterpart in the presence of gating, P_{tr}^{sg} . As a result, we have

$$J_{mo}^{sg} = k_{Hc} P_{op} P_{tr}^{sg}, \quad P_{op} = \beta / (\alpha + \beta). \quad (3.4)$$

Note that while the first modification is widely accepted in the literature,¹ the second, to the best of our knowledge, has never been considered before. The only exception is a recent paper on stochastic gating in insect respiration, where a different version of the problem is analyzed.⁴¹

As shown in Sec. IV, the translocation probability P_{tr}^{sg} is

$$P_{tr}^{sg} = \frac{\alpha + \beta}{\alpha F_1(\lambda|\nu) + \beta(2 + \nu)}, \quad (3.5)$$

where function $F_1(\lambda|\nu)$ is given by

$$F_1(\lambda|\nu) = \frac{2\lambda\nu + (\lambda^2 + \nu^2) \tanh \lambda}{\lambda(\nu + \lambda \tanh \lambda)}. \quad (3.6)$$

Here, the dimensionless argument λ of function $F_1(\lambda|\nu)$ is the square root of the ratio of the characteristic time, L^2 / D_{ch} , of the particle passage through the channel and the gate equilibration time, $1/(\alpha + \beta)$,

$$\lambda = L \sqrt{(\alpha + \beta) / D_{ch}}. \quad (3.7)$$

As λ increases from zero to infinity, function $F_1(\lambda|\nu)$ monotonically decreases from $2 + \nu$ to unity. Its plot is shown in Fig. 2.

FIG. 2. Function $F_1(\lambda|\nu)$ at $\nu = 1, 10, 10^2$, and 10^3 , as indicated in the plot.

The flux J_{mo}^{sg} is used to find the inter-transition time T_{mo}^{sg} ,

$$T_{mo}^{sg} = \frac{1}{J_{mo}^{sg}} = \frac{1}{k_{Hc}} \left(2 + \nu + \frac{\alpha}{\beta} F_1(\lambda|\nu) \right). \quad (3.8)$$

This time is larger than its counterpart in the absence of gating, T_{mo}^{ng} , given in Eq. (3.1). The relation between the two times is

$$T_{mo}^{sg} = T_{mo}^{ng} + \frac{\alpha}{k_{Hc}\beta} F_1(\lambda|\nu). \quad (3.9)$$

To illustrate the gating effect on the flux, consider the ratio of the flux in the presence of gating, J_{mo}^{sg} , to its counterpart in the no-gating case, J_{mo}^{ng} . As follows from the above equations, this ratio is given by

$$\frac{J_{mo}^{sg}}{J_{mo}^{ng}} = \frac{\beta(2 + \nu)}{\alpha F_1(\lambda|\nu) + \beta(2 + \nu)}. \quad (3.10)$$

In the limiting cases of slow ($\lambda \rightarrow 0$) and fast ($\lambda \rightarrow \infty$) gating, this ratio reduces to

$$\frac{J_{mo}^{sg}}{J_{mo}^{ng}} = \begin{cases} \beta / (\alpha + \beta) = P_{op}, & \lambda \rightarrow 0, \\ \beta(2 + \nu) / [\alpha + \beta(2 + \nu)], & \lambda \rightarrow \infty. \end{cases} \quad (3.11)$$

Thus, the conventional assumption, $J_{mo}^{sg} = J_{mo}^{ng} P_{op}$, is applicable only at slow gating. The flux J_{mo}^{sg} can be much higher than its conventional estimate. This happens because the gating increases the translocation probability, P_{tr}^{sg} , Eq. (3.5), compared to P_{tr}^{ng} , Eq. (2.4). To illustrate the deviation of the flux J_{mo}^{sg} from its conventional estimate, consider the ratio

$$\frac{J_{mo}^{sg}}{P_{op} J_{mo}^{ng}} = \frac{P_{tr}^{sg}}{P_{tr}^{ng}} = \frac{(\alpha + \beta)(2 + \nu)}{\alpha F_1(\lambda|\nu) + \beta(2 + \nu)}. \quad (3.12)$$

FIG. 3. Ratio of the flux through the stochastically gated channel of multiple occupancy to that given by the conventional estimate, $J_{mo}^{sg} / (P_{op} J_{mo}^{ng})$ in Eq. (3.12), as a function of λ at the different values of open channel probabilities, $P_{op} = 10^{-3}, 10^{-2}, 3 \times 10^{-2}, 10^{-1}$, from top to bottom. Other parameters are channel length, $L = 5$ nm, effective channel radius, $a = 1$ nm, diffusivity in the bulk, $D_b = 10^{-10}$ m²/s, and diffusivity in the channel, $D_{ch} = 10^{-12}$ m²/s. It is seen that channel gating, when it is fast enough, is able to increase the flux compared to the simple conventional estimate, namely, the product of the flux through the non-gated channel and the open channel probability, by orders of magnitude.

As λ increases from zero to infinity, $F_1(\lambda|\nu)$ decreases from $(2 + \nu)$ to unity, and the ratio increases from unity to $(2 + \nu) / [1 + P_{op}(1 + \nu)]$. The deviation from the conventional estimate is most pronounced when $P_{op} \ll 1$, and the channel is predominantly closed. Figure 3 illustrates the λ -dependence of the ratio in Eq. (3.12), at different values of the open channel probability, P_{op} .

B. Singly occupied channel

The flux through a stochastically gated singly occupied channel, denoted by J_{so}^{sg} , is given by a formula analogous to that in Eq. (2.7), with τ_{tr}^{ng} in Eq. (2.8), in which not only the translocation probability, P_{tr}^{ng} , but also the channel mean lifetimes in the empty and occupied states, τ_{emp}^{ng} and τ_{oc}^{ng} , should be replaced by their counterparts in the presence of gating, denoted by τ_{emp}^{sg} and τ_{oc}^{sg} . Thus we have

$$J_{so}^{sg} = \frac{P_{tr}^{sg}}{\tau_{emp}^{sg} + \tau_{oc}^{sg}}. \quad (3.13)$$

The mean lifetimes τ_{emp}^{sg} and τ_{oc}^{sg} , found in Secs. V and VI, are given by

$$\tau_{emp}^{sg} = \frac{1}{\beta} \left[\frac{\alpha + \beta}{k_H c} + \frac{\alpha F_2(\lambda|\nu)}{\alpha F_1(\lambda|\nu) + \beta(2 + \nu)} \right] \quad (3.14)$$

and

$$\tau_{oc}^{sg} = \tau_{oc}^{ng} \frac{(\alpha + \beta)(2 + \nu)}{\alpha F_1(\lambda|\nu) + \beta(2 + \nu)}, \quad (3.15)$$

where function $F_2(\lambda|\nu)$ is

$$F_2(\lambda|\nu) = 1 - \frac{\nu}{\nu \cosh \lambda + \lambda \sinh \lambda}. \quad (3.16)$$

This function monotonically increases with λ from zero at $\lambda = 0$ to unity, as $\lambda \rightarrow \infty$. Its plot is shown in Fig. 4.

The flux J_{so}^{sg} is used to find the mean inter-transition time T_{so}^{sg} ,

$$T_{so}^{sg} = \frac{1}{J_{so}^{sg}} = T_{so}^{ng} + \frac{\alpha}{\beta} \left(\frac{1}{k_H c} F_1(\lambda|\nu) + \frac{1}{\alpha + \beta} F_2(\lambda|\nu) \right), \quad (3.17)$$

which is longer than its counterpart in the absence of gating, T_{so}^{ng} , as it must be. Using Eqs. (3.3), (3.4), and (3.7), one can check that this time is also longer than its counterpart for multiply occupied channels, T_{mo}^{sg} . The relation between these two mean inter-transition times is

$$T_{so}^{sg} = T_{mo}^{sg} + \frac{L}{\kappa} + \frac{L^2}{2D_{ch}} + \frac{\alpha}{\beta(\alpha + \beta)} F_2(\lambda|\nu). \quad (3.18)$$

The ratio of the fluxes through stochastically gated and non-gated singly occupied channels is given by

$$\frac{J_{so}^{sg}}{J_{so}^{ng}} = \frac{\beta(2 + \nu)(1 + V_{ch}c/2)}{\alpha[F_1(\lambda|\nu) + k_H c F_2(\lambda|\nu)/(\alpha + \beta)] + \beta(2 + \nu)(1 + V_{ch}c/2)}, \quad (3.19)$$

where $V_{ch} = \pi a^2 L$ is the channel volume related to the parameters k_H , L , and κ by the relation $k_H L / \kappa = V_{ch}$. This flux ratio reduces to that in Eq. (3.10) at low concentrations, $c \rightarrow 0$, where the particle repulsion can be neglected. In the limiting cases of slow ($\lambda \rightarrow 0$) and fast ($\lambda \rightarrow \infty$) gating, functions $F_1(\lambda|\nu)$ and $F_2(\lambda|\nu)$ simplify and Eq. (3.19) reduces to

$$\frac{J_{so}^{sg}}{J_{so}^{ng}} = \begin{cases} \frac{\beta}{\alpha + \beta} = P_{op}, & \lambda \rightarrow 0, \\ \frac{\beta(2 + \nu)(\alpha + \beta)(1 + V_{ch}c/2)}{\alpha(\alpha + \beta + k_H c) + \beta(2 + \nu)(\alpha + \beta)(1 + V_{ch}c/2)}, & \lambda \rightarrow \infty \end{cases}. \quad (3.20)$$

This shows that the conventional estimate of the gating effect on transport works at slow gating and fails when the gating is fast. To characterize the deviation from the conventional estimate, we again consider the ratio of the flux in the presence of gating to its conventional counterpart, which in the case of a singly occupied channel takes the form

$$\frac{J_{so}^{sg}}{P_{op} J_{so}^{ng}} = \frac{(\alpha + \beta)(2 + \nu)(1 + V_{ch}c/2)}{\alpha[F_1(\lambda|\nu) + k_H c F_2(\lambda|\nu)/(\alpha + \beta)] + \beta(2 + \nu)(1 + V_{ch}c/2)}. \quad (3.21)$$

As λ increases from zero to infinity, this ratio increases from unity to its plateau value $(2 + \nu) / [P_{op}(2 + \nu) + (1 - P_{op}) / (1 + V_{ch}c/2)]$. This plateau value is greater than its counterpart for the multiply occupied channel, to which it reduces as $c \rightarrow 0$. The λ -dependence of the ratio in Eq. (3.21) is similar to its counterpart in the case of multiply occupied channel shown in Fig. 3.

Finally, we use Eqs. (3.10) and (3.19) to establish the relation between the fluxes through singly and multiply occupied stochastically gated channels,

$$\frac{J_{mo}^{sg}}{J_{so}^{sg}} = \frac{J_{mo}^{ng}}{J_{so}^{ng}} Q. \quad (3.22)$$

Here Q is a function of the problem parameters given by

$$Q = \frac{\alpha[F_1(\lambda|\nu) + k_H c F_2(\lambda|\nu)/(\alpha + \beta)] + \beta(2 + \nu)(1 + V_{ch}c/2)}{[\alpha F_1(\lambda|\nu) + \beta(2 + \nu)](1 + V_{ch}c/2)}. \quad (3.23)$$

FIG. 4. Function $F_2(\lambda|\nu)$ at $\nu = 10^{-2}$, 10^{-1} , 1, and 10 for the solid curves, and $\nu = 10^2$ for the dashed one, as indicated near the curves.

As λ increases from zero (slow gating) to infinity (fast gating), function Q decreases from unity to its fast-gating limiting value,

$$Q|_{\lambda \rightarrow \infty} = 1 - \frac{V_{ch}c(1 - P_{op})}{(2 + V_{ch}c)[1 + P_{op}(1 + \nu)]}. \quad (3.24)$$

In the absence of gating, according to Eqs. (2.2) and (2.7)–(2.9), the ratio of the fluxes through multiply and singly occupied channels is

$$\frac{J_{mo}^{ng}}{J_{so}^{ng}} = 1 + \frac{k_H c L}{2\kappa} = 1 + \frac{V_{ch}c}{2}. \quad (3.25)$$

This allows us to write the flux ratio in the presence of stochastic gating, Eq. (3.22), as

$$\frac{J_{mo}^{sg}}{J_{so}^{sg}} = 1 + \frac{V_{ch}c}{2} \frac{2\kappa\alpha F_2(\lambda|\nu) + \beta(2 + \nu)(\alpha + \beta)L}{[\alpha F_1(\lambda|\nu) + \beta(2 + \nu)](\alpha + \beta)L}. \quad (3.26)$$

FIG. 5. The λ -dependences of the flux ratio in Eq. (3.26) at different values of open channel probabilities, $P_{op} = 10^{-1}$, 10^{-2} , 10^{-3} , and 10^{-4} , from top to bottom. Solute concentration is chosen to be $c = 6.02 \times 10^{26} \text{ m}^{-3}$; other parameters are given in the caption of Fig. 3.

As λ increases from zero to infinity, this flux ratio decreases from its value in the absence of gating, Eq. (3.25), to $1 + (V_{ch}c/2)\beta(2 + \nu)/[\alpha + \beta(2 + \nu)]$, which is its fast gating limiting value. The latter, at low channel open probability, can be significantly smaller than its no-gating counterpart.

One can see that the fast gating limiting value of the flux ratio J_{mo}^{sg}/J_{so}^{sg} approached unity as the ratio β/α and, therefore, the open channel probability tends to zero. In such regimes, fluxes through stochastically gated multiply and singly occupied channels become equal because the predominantly closed channel is open for such a short time that only one particle has a chance to enter the channel. The λ -dependences of the flux ratio at fixed open channel probabilities presented in Fig. 5 show a nonmonotonic behavior.

IV. TRANSLOCATION PROBABILITY

Consider a molecule entering the channel through the open gate (see Fig. 1) at $t = 0$. Let $G_i(x, t)$ be the molecule propagator in the channel, which is the probability density of finding the molecule at distance x , $0 < x < L$, from the gated channel end, located at $x = 0$, and the gate in state i , $i = op$ (open), cl (close), at time $t > 0$, on condition that the molecule did not escape from the channel during time t . The two components of the propagator satisfy

$$\frac{\partial G_{op}}{\partial t} = D_{ch} \frac{\partial^2 G_{op}}{\partial x^2} - \alpha G_{op} + \beta G_{cl}, \quad (4.1)$$

$$\frac{\partial G_{cl}}{\partial t} = D_{ch} \frac{\partial^2 G_{cl}}{\partial x^2} + \alpha G_{op} - \beta G_{cl}, \quad (4.2)$$

subject to the initial and boundary conditions,

$$G_{op}(x, 0) = \delta(x), \quad G_{cl}(x, 0) = 0, \quad (4.3)$$

$$D_{ch} \frac{\partial G_{op}}{\partial x} \Big|_{x=0} = \kappa G_{op}(0, t) = f_r(t), \quad \frac{\partial G_{cl}}{\partial x} \Big|_{x=0} = 0, \quad (4.4)$$

$$-D_{ch} \frac{\partial G_{op}}{\partial x} \Big|_{x=L} = \kappa G_{op}(L, t) = f_{tr}^{op}(t), \quad (4.5)$$

$$-D_{ch} \frac{\partial G_{cl}}{\partial x} \Big|_{x=L} = \kappa G_{cl}(L, t) = f_{tr}^{cl}(t).$$

Equations (4.1) and (4.2) describe diffusion of the molecule inside the channel and random opening and closing of the gate determined by the rate constants α and β [see Fig. 1 and Eq. (2.1)]. The initial conditions in Eq. (4.3) account for the fact that the molecule enters the channel through the open gate. The boundary conditions at the channel ends, Eqs. (4.4) and (4.5), depend on whether the gate is open or closed. When the gate is open, the two components of the propagator satisfy the radiation (partially absorbing) boundary conditions that involve the effective trapping rate κ of the molecule at the channel end, given by Eq. (2.5). This trapping rate (having the dimensions of length per time) determines the fate of the molecule coming to the channel end, namely, whether the molecule escapes from the channel into the bulk or continues its diffusion inside the channel. When the gate is closed, the propagator $G_{cl}(x, t)$ satisfies the reflecting boundary conditions at $x = 0$.

The radiation boundary conditions describe escape of the molecule from the channel to the bulk on the left and right from the membrane. The probability fluxes escaping from the channel at time t are denoted by $f_r(t)$, $f_{tr}^{op}(t)$, and $f_{tr}^{cl}(t)$, Eqs. (4.4) and (4.5). The returning (r) flux, $f_r(t)$, is due to those realizations of the stochastic trajectory of the molecule, which escape through the left channel end at $x = 0$. The fluxes $f_{tr}^{op}(t)$ and $f_{tr}^{cl}(t)$ are due to such realizations which escape through the right channel end at $x = L$. These realizations are responsible for the translocations of the molecule from one side of the membrane to the other. The superscripts “ op ” and “ cl ” indicate whether the gate is open or closed when the translocating molecule escapes from the channel.

We use fluxes $f_r(t)$, $f_{tr}^{op}(t)$, and $f_{tr}^{cl}(t)$ to find the return and translocation probabilities, denoted by P_r^{sg} , P_{tr}^{op} , and P_{tr}^{cl} ,

$$P_r^{sg} = \int_0^\infty f_r(t) dt = \kappa \hat{G}_{op}(0), \quad (4.6)$$

$$P_{tr}^{op,cl} = \int_0^\infty f_{tr}^{op,cl}(t) dt = \kappa \hat{G}_{op,cl}(L), \quad (4.7)$$

where $\hat{G}_i(x)$ denotes the integral of $G_i(x, t)$ with respect to time from zero to infinity, $i = op, cl$,

$$\hat{G}_i(x) = \int_0^\infty G_i(x, t) dt. \quad (4.8)$$

The translocation probability of interest, P_{tr}^{sg} , is the sum of the translocation probabilities P_{tr}^{op} and P_{tr}^{cl} ,

$$P_{tr}^{sg} = P_{tr}^{op} + P_{tr}^{cl}. \quad (4.9)$$

Because of the probability conservation, we have

$$P_r^{sg} + P_{tr}^{sg} = P_r^{sg} + P_{tr}^{op} + P_{tr}^{cl} = 1. \quad (4.10)$$

Integrating Eqs. (4.1), (4.2), (4.4), and (4.5) with respect to time from zero to infinity and using the initial conditions in Eq. (4.3), we find that functions $\hat{G}_i(x)$ satisfy

$$D_{ch} \frac{d^2 \hat{G}_{op}}{dx^2} - \alpha \hat{G}_{op} + \beta \hat{G}_{cl} = -\delta(x), \quad (4.11)$$

$$D_{ch} \frac{d^2 \hat{G}_{cl}}{dx^2} + \alpha \hat{G}_{op} - \beta \hat{G}_{cl} = 0, \quad (4.12)$$

subject to the boundary conditions

$$D_{ch} \frac{d\hat{G}_{op}}{dx} \Big|_{x=0} = \kappa \hat{G}_{op}(0) = P_r^{sg}, \quad \frac{d\hat{G}_{cl}}{dx} \Big|_{x=0} = 0, \quad (4.13)$$

$$\begin{aligned} -D_{ch} \frac{d\hat{G}_{op}}{dx} \Big|_{x=L} &= \kappa \hat{G}_{op}(L) = P_{tr}^{op}, \\ -D_{ch} \frac{d\hat{G}_{cl}}{dx} \Big|_{x=L} &= \kappa \hat{G}_{cl}(L) = P_{tr}^{cl}, \end{aligned} \quad (4.14)$$

where we have used the definitions of the return and translocation probabilities in Eqs. (4.6) and (4.7).

The solutions for $\hat{G}_{op,cl}(x)$ are obtained in the [Appendix](#). We use these solutions and the definitions in Eq. (4.14) to find

the translocation probabilities P_{tr}^{op} and P_{tr}^{cl} , as well as their sum, the total translocation probability, P_{tr}^{sg} , Eq. (4.9),

$$P_{tr}^{op} = \frac{\alpha(1 - F_2(\lambda|\nu)) + \beta}{\alpha F_1(\lambda|\nu) + \beta(2 + \nu)}, \quad (4.15)$$

$$P_{tr}^{cl} = \frac{\alpha F_2(\lambda|\nu)}{\alpha F_1(\lambda|\nu) + \beta(2 + \nu)}, \quad (4.16)$$

$$P_{tr}^{sg} = \frac{\alpha + \beta}{\alpha F_1(\lambda|\nu) + \beta(2 + \nu)}. \quad (4.17)$$

These formulas are the main results of this section. The expression in Eq. (4.17) is just the expression for the translocation probability in Eq. (3.5). The ratio of the translocation probability P_{tr}^{sg} in the presence of stochastic gating to its counterpart in the absence of gating, P_{tr}^{ng} in Eq. (2.4), is given by

$$\frac{P_{tr}^{sg}}{P_{tr}^{ng}} = \frac{(\alpha + \beta)(2 + \nu)}{\alpha F_1(\lambda|\nu) + \beta(2 + \nu)}. \quad (4.18)$$

Since function $F_1(\lambda|\nu)$ is smaller than $\nu + 2$, when $\lambda > 0$ (see Fig. 2), this ratio is larger than unity, and hence $P_{tr}^{sg} > P_{tr}^{ng}$, as might be expected. The ratio is equal to unity, and $P_{tr}^{sg} = P_{tr}^{ng}$, when $\lambda = 0$, because stochastic gating does not affect the translocation probability under such conditions.

V. MEAN CHANNEL LIFETIME IN THE OCCUPIED STATE

The mean lifetime of a singly occupied stochastically gated channel in the occupied state is the mean lifetime of the molecule in such a channel. For a molecule entering the channel at $t = 0$, its survival probability in the channel for time t , $S_{oc}^{sg}(t)$, is related to the two components of the molecule propagator by the relation

$$S_{oc}^{sg}(t) = \int_0^L (G_{op}(x, t) + G_{cl}(x, t)) dx. \quad (5.1)$$

The probability density of the molecule lifetime in the channel is $-dS_{oc}^{sg}(t)/dt$. We use this probability density to find the mean lifetime of the molecule in the channel, which is the mean channel lifetime in the occupied state, τ_{oc}^{sg} ,

$$\begin{aligned} \tau_{oc}^{sg} &= \int_0^\infty t (-dS_{oc}^{sg}(t)/dt) dt = \int_0^\infty S_{oc}^{sg}(t) dt \\ &= \int_0^L (\hat{G}_{op}(x) + \hat{G}_{cl}(x)) dx. \end{aligned} \quad (5.2)$$

The two components of the propagator are found in the [Appendix](#). Performing the integration over x , we arrive at the formula for τ_{oc}^{sg} , given in Eq. (3.15),

$$\tau_{oc}^{sg} = \tau_{oc}^{ng} \frac{(\alpha + \beta)(2 + \nu)}{\alpha F_1(\lambda) + \beta(2 + \nu)}. \quad (5.3)$$

Here $\tau_{oc}^{ng} = L/(2\kappa)$ is the mean lifetime of the singly occupied channel in the occupied state in the absence of gating, which is the mean lifetime of the molecule in the channel under such conditions, given in Eq. (2.9).

The formula for τ_{oc}^{sg} in Eq. (5.3) is the main result of this section. As follow from Eq. (5.3), the ratio of τ_{oc}^{sg} to

τ_{oc}^{ng} is identical to the ratio of the translocation probabilities P_{tr}^{sg} and P_{tr}^{ng} in the presence and absence of gating, given in Eq. (4.18). As a consequence, the relation between the two mean lifetimes is the same as the relation between the two translocation probabilities: $\tau_{oc}^{sg} > \tau_{oc}^{ng}$, when $\lambda > 0$, and $\tau_{oc}^{sg} = \tau_{oc}^{ng}$, when $\lambda = 0$.

VI. MEAN CHANNEL LIFETIME IN THE EMPTY STATE

The lifetime of a singly occupied channel in the empty state depends on whether the gate is open or closed when the molecule escapes from the channel. Let τ_{emp}^{op} and τ_{emp}^{cl} be the mean lifetimes of the empty channel, on condition that the gate is open and closed when the molecule leaves the channel, respectively. The realization probabilities of the two conditions, respectively, are $P_r^{sg} + P_{tr}^{op}$ and P_{tr}^{cl} , where the return and translocation probabilities, P_r^{sg} , P_{tr}^{op} , and P_{tr}^{cl} , are defined in Eqs. (4.6) and (4.7). The mean lifetime of a singly occupied stochastically gated channel in the empty state, τ_{emp}^{sg} , is defined as

$$\tau_{emp}^{sg} = \tau_{emp}^{op} (P_r^{sg} + P_{tr}^{op}) + \tau_{emp}^{cl} P_{tr}^{cl}. \quad (6.1)$$

The mean lifetime τ_{emp}^{cl} is longer than its counterpart τ_{emp}^{op} for the open state of the gate because a molecule cannot enter the channel when the gate is closed. The difference between these two mean lifetimes is equal to the mean lifetime β^{-1} of the gate in the closed state,

$$\tau_{emp}^{cl} = \tau_{emp}^{op} + \beta^{-1}. \quad (6.2)$$

Substituting this into Eq. (6.1) and using Eqs. (4.9) and (4.10), we obtain

$$\tau_{emp}^{sg} = \tau_{emp}^{op} + \beta^{-1} P_{tr}^{cl}, \quad (6.3)$$

where we have used the fact that $P_r^{sg} + P_{tr}^{op} + P_{tr}^{cl} = 1$, Eq. (4.10).

To find the mean lifetime τ_{emp}^{op} , we introduce the probabilities $W_i(t)$, $i = op, cl$, of finding the singly occupied channel empty and the gate open and closed, respectively, at time t , on condition that the gate is open when a molecule escapes from the channel at $t = 0$. These probabilities satisfy the rate equations, which describe the gate opening and closing, as well as the entry of a molecule into the channel when the gate is open,

$$\frac{dW_{op}(t)}{dt} = -(\alpha + k_{HC}) W_{op}(t) + \beta W_{cl}(t), \quad (6.4)$$

$$\frac{dW_{cl}(t)}{dt} = \alpha W_{op}(t) - \beta W_{cl}(t), \quad (6.5)$$

subject to the initial conditions

$$W_{op}(0) = 1, \quad W_{cl}(0) = 0. \quad (6.6)$$

The channel survival probability in the empty state at time t , denoted by $S_{emp}(t)$, is the sum of the probabilities $W_{op}(t)$ and $W_{cl}(t)$,

$$S_{emp}(t) = W_{op}(t) + W_{cl}(t). \quad (6.7)$$

The probability density of the channel lifetime in the empty state is $-dS_{emp}(t)/dt$. We use this probability density to find the mean lifetime τ_{emp}^{op} ,

$$\tau_{emp}^{op} = \int_0^\infty t \left(-\frac{dS_{emp}(t)}{dt} \right) dt = \int_0^\infty S_{emp}(t) dt = \hat{W}_{op} + \hat{W}_{cl}. \quad (6.8)$$

Here $\hat{W}_i$ denotes the integral of $W_i(t)$ with respect to time from zero to infinity, $i = op, cl$,

$$\hat{W}_i = \int_0^\infty W_i(t) dt. \quad (6.9)$$

Integrating Eqs. (6.4) and (6.5) with respect to time from zero to infinity, we find that $\hat{W}_{op}$ and $\hat{W}_{cl}$ satisfy

$$(\alpha + k_{HC}) \hat{W}_{op} - \beta \hat{W}_{cl} = 1, \quad (6.10)$$

$$\alpha \hat{W}_{op} - \beta \hat{W}_{cl} = 0. \quad (6.11)$$

Solving these equations, we obtain

$$\hat{W}_{op} = \frac{1}{k_{HC}}, \quad \hat{W}_{cl} = \frac{\alpha}{\beta k_{HC}}. \quad (6.12)$$

Substituting $\hat{W}_{op}$ and $\hat{W}_{cl}$ above into Eq. (6.8), we arrive at

$$\tau_{emp}^{op} = \frac{\alpha + \beta}{\beta k_{HC}} = \tau_{emp}^{ng} \left(1 + \frac{\alpha}{\beta} \right), \quad (6.13)$$

where $\tau_{emp}^{ng} = 1/(k_{HC})$ is the mean lifetime of this singly occupied channel in the absence of gating, given in Eq. (2.9). The above equation shows that the gating increases the channel lifetime in the empty state, $\tau_{emp}^{sg} > \tau_{emp}^{ng}$, as might be expected.

Finally, we find the mean lifetime τ_{emp}^{sg} by substituting into Eq. (6.3) the expressions for τ_{emp}^{op} and P_{tr}^{cl} given, respectively, in Eqs. (6.13) and (4.16). This leads to the formula for τ_{emp}^{sg} in Eq. (3.14), which can also be written as

$$\tau_{emp}^{sg} = \tau_{emp}^{ng} \left\{ 1 + \frac{\alpha}{\beta} \left[1 + \frac{k_{HC} F_2(\lambda|\nu)}{\alpha F_1(\lambda|\nu) + \beta(2 + \nu)} \right] \right\}, \quad (6.14)$$

where function $F_2(\lambda|\nu)$ is defined in Eq. (3.16). This establishes the relation between the mean lifetimes of the singly occupied channel in the empty state in the presence and absence of stochastic gating. The former, as might be expected, is longer than the latter.

VII. CONCLUDING REMARKS

In this paper, we develop a theory of solute transport through multiply and singly occupied cylindrical channels in the presence of stochastic gating. This is done in the framework of the diffusion model of the solute dynamics in the channel,^{37,38,40,42} assuming that the channel transitions between the open and closed states are Markovian. The theory shows that the widely accepted approximation, which assumes that the flux decrease due to stochastic gating can be described by multiplying the flux through the open channel by the probability of finding the channel in the open state, holds only when the gating dynamics are sufficiently slow. When the characteristic gating time is comparable with or even smaller than the characteristic time of the solute passage through the channel, the proportionality between the flux and the channel open probability breaks. There is a range of parameters where the flux differs from the conventional estimate by orders of magnitude.

A simple intuitive interpretation of the obtained results is as follows. The fate of a molecule entering the channel depends

on the state of the gate: when the gate is open, the molecule translocation probability, given by Eq. (2.4), is smaller than unity. However, if the gate closes just after the molecule enters and stays closed, the translocation probability is unity. At slow gating, the fraction of molecules entering the channel, which feel the change in the gate state, is negligibly small. The overwhelming majority of the entering molecules is unaware about gating. This is why the conventional approximation works well in this case. In contrast, when gating is fast, all molecules feel the gate opening and closing which leads to the increase in the translocation probability, Eq. (3.5). As a consequence, the flux under the fast gating conditions is higher than its conventional estimate.

The flux increase, being dependent on the relative rates of the gating and diffusional translocation, is pronounced only if the gating is fast. For this reason, the effect may be especially important in the case of metabolite-specific channels. Indeed, while the characteristic passage time of small ions through such channels is in the range of nanoseconds,⁴³ metabolite translocation occurs much slower⁴⁴ and is characterized by microseconds, as in the case of ATP molecules traversing the voltage-dependent channels of the outer mitochondrial membrane,⁴⁵ or even milliseconds, as in the case of sugar molecules diffusing through sugar-specific bacterial channels.^{46,47} It should be noted that stochastic gating of the channel can occur on a much shorter time scale. It is well-known that the characteristic time of the protein conformational transitions spans a huge range which extends, depending on the transition nature, from hundreds of seconds down to picoseconds.⁴⁸ In view of these facts, metabolite-specific channels would appear to be good candidates for experimental studies of the new fast-gating effects discussed in the present paper.

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APPENDIX: FUNCTIONS $\hat{G}_{op}(x)$ AND $\hat{G}_{cl}(x)$

Functions $\hat{G}_{op}(x)$ and $\hat{G}_{cl}(x)$ are solutions to Eqs. (4.11) and (4.12) subject to the boundary conditions in Eqs. (4.13) and (4.14). To find these functions, we first eliminate the delta-function on the right-hand side of Eq. (4.11). By integrating both sides of this equation with respect to x from $x = 0$ to $x = \varepsilon$, $\varepsilon \rightarrow 0$, we arrive at the boundary condition for $\hat{G}_{op}(x)$ shifted into the channel,

$$-D_{ch} \frac{d\hat{G}_{op}}{dx} \Big|_{x \rightarrow 0} = 1 - \kappa \hat{G}_{op}(0). \quad (\text{A1})$$

The solutions to the resulting homogeneous equations for $\hat{G}_{op}(x)$ and $\hat{G}_{cl}(x)$ are given by

$$\hat{G}_{op}(x) = \beta (A - Bx) + M e^{-\lambda x/L} + N e^{\lambda x/L}, \quad (\text{A2})$$

$$\hat{G}_{cl}(x) = \alpha (A - Bx) - M e^{-\lambda x/L} - N e^{\lambda x/L}, \quad (\text{A3})$$

where the dimensionless parameter λ is defined in Eq. (3.7). Factors A , B , M , and N in the above equations are

$$A = \frac{1 + \nu}{\kappa [\alpha F_1(\lambda|\nu) + \beta (2 + \nu)]}, \quad (\text{A4})$$

$$B = \frac{1}{D_{ch} [\alpha F_1(\lambda|\nu) + \beta (2 + \nu)]}, \quad (\text{A5})$$

$$M = \frac{\alpha L (\nu + \lambda)}{\lambda [\nu + \lambda + (\nu - \lambda) e^{-2\lambda}]} B, \quad (\text{A6})$$

$$N = \frac{\alpha L (\lambda - \nu) e^{-2\lambda}}{\lambda [\nu + \lambda + (\nu - \lambda) e^{-2\lambda}]} B, \quad (\text{A7})$$

with function $F_1(\lambda|\nu)$ defined in Eq. (3.6).

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