Narrow Escape to a structured target and application to viral entry in the nucleus

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I. ABSTRACT

In cellular biology, reaching a target before being degraded or trapped is ubiquitous. An interesting example is given by the virus journey inside the cell cytoplasm: in order to replicate, most viruses have to reach the nucleus before being trapped or degraded. We present here a general approach to estimate the probability and the conditional mean first passage time for such a viral particle to attain safely the nucleus, covered with many (around two thousands) small absorbing pores. Due to this large number of small holes, which defines the limiting scale, any Brownian simulations are very unstable. Our new asymptotic formulas precisely account for this phenomena and allow to quantify the cytoplasmic stage of viral infection. We confirm our analysis with Brownian simulations.

II. INTRODUCTION.

Cellular trafficking describes how particles (molecules, proteins, DNA, RNA, viruses) are moving inside the complex and crowded cellular organization [18]. For example, vesicles or RNA granules [8] have to reach specific targets to deliver their payload or trigger protein synthesis. However, large DNA or plasmid are too large and cannot passes the cytoplasm [7]. In some cases, large particles are intermittently transported on the cytoskeleton by molecular motors such as dyneins that travel along microtubules (MTs) toward the nucleus. Many DNA viruses have the ability to hijack the cellular transport machinery to reach a nuclear pore and deliver their genetic material inside the nucleus [9, 25]. Although viral trajectories can be monitored in vivo for some viruses such as HIV or the Adeno-Associated Virus [1, 22], these trajectories are made of epochs that can be classified as effective diffusion and other as directed motion. The precise nature of these trajectories is still unclear. In addition, on their way to the nucleus, viruses can be trapped in the crowded cytoplasm or degraded through several pathways, including the ubiquitin-proteasome. To quantify the success of the early step of viral infection, all these phenomena should be taken into account and we recently used a modeling approach at the single particle level [10, 13]. Due to small parameters such the size of nuclear pore, Brownian simulations are always unstable and can only cover a specific fraction of the parameter space, thus we developed asymptotic formula that give precise dependencies. For generic viruses that can intermittently travel along MTs, using a stochastic approach, we derived expressions for the conditional mean first passage time (MFPT) $\tau_n$ and the probability $P_n$ to arrive to a target [10, 15]. However, our previous formula are limited to a target that does not contain many absorbing hole. We propose here to extend our analysis to that case. As revealed recently [19], this analysis relies on the explicit expansion of the Neumann-Green’s function to order three. We will need to further develop the method for many interacting small holes [4, 12, 21].

The paper is organized as follows. We will now recall our modeling of viral particles and the stochastic description of the associated trajectories. We will then extend the small hole interaction method to the case of a stochastic particle with a drift and derive the interaction matrix. Using the precise expansion of the Neumann-Green’s function for the sphere, we will derive new asymptotic formula for the probability and the mean conditioning time to reach one of the many nuclear pore. We will confirm our analysis with some Brownian simulations and show how our new formula improve the previous ones.

III. FROM SINGLE PARTICLE TRAJECTORIES TO THE MEAN TIME TO A NUCLEAR PORE.

The complex intermittent trajectories of a particle $x(t)$ can be described using the switching stochastic rule

$$dx = \begin{cases} \sqrt{2D}dw & \text{when } x(t) \text{ is free} \\ V dt & x(t) \text{ bound} \end{cases}$$

(1)

where $w$ is a standard 3d-Brownian motion, $D$ the diffusion constant and $V$ the velocity of the directed motion along MTs (randomly distributed). The switching dynamics depends on the attachment and detachment rates [14]. To be able to obtain an explicit analysis, we coarse grain this complex behavior into by a steady state stochastic equation

$$dx = b(x)dt + \sqrt{2D}dw,$$

(2)

where the effective drift $b(x)$ can be calibrated using the criteria that inside the cytoplasm, the mean first passage time of the two stochastic processes 1 and 2 is the same [14, 15]. $b$ depends on the cell geometry, the number and distribution of MTs and the rates of binding and unbinding of the particle to MTs.
FIG. 1. **Schematic representation of the cell cytoplasm as 3-dimensional domain** $\Omega$. (Right-hand side): Langevin trajectories, solutions of (2), can be annihilated or absorbed at the small windows located on $\partial S_a$. Right-hand side: a simplified spherical cell (radius $R$) with a spherical centered nucleus (radius $a$).

For most viruses, they have to reach one of the small circular absorbing windows (of radius $\epsilon \ll 1$) located on the boundary $\partial S_a$ of the nucleus, which is usually approximated as a small sphere $S_a$. The cell membrane defines a boundary $\partial \Omega_{ext}$. The cell cytoplasm is represented as a three-dimensional bounded domain $\Omega$, whose boundary $\partial \Omega = \partial \Omega_{ext} \cup \partial S_a$ is reflecting except for the $n$ small absorbing windows $\partial N_a$ located on the nucleus (FIG.1 left).

We model the degradation activity in the cytoplasm by a steady state killing rate $k(x)$ and a trajectory described by equation (2) can thus disappear before reaching the absorbing boundary. Thus the survival probability density function (SPDF) is solution of the Fokker-Planck equation [11]

$$\frac{\partial p}{\partial t} = \Delta p - \nabla \cdot b p - kp$$

with the boundary conditions:

$$p(x, t) = 0 \text{ on } \partial N_a \text{ and } J(x, t)n_x = 0 \text{ on } \partial \Omega - \partial N_a$$

where the flux density vector is

$$J(x, t) = -D\nabla p(x, t) + b(x)p(x, t).$$

and $n_x$ is the normal derivative at a boundary point $x$.

We recall that the mean probability $\langle P \rangle$ and the conditional MFPT $\langle \tau \rangle$ (averaged over the initial distribution) for a particle following equation (2) to reach the boundary $\partial N_a$ before degradation can be expressed using $\tilde{p}(x) = \int_0^\infty p(x, t)dt$ and $q(x) = \int_0^\infty tp(x, t)dt$ [10] as

$$\langle P \rangle = 1 - \int_\Omega k(x)\tilde{p}(x)dx,$$

and

$$\langle \tau \rangle = \frac{\int_\Omega \tilde{p}(x)dx - \int_\Omega k(x)q(x)dx}{1 - \int_\Omega k(x)\tilde{p}(x)dx}.$$
For a potential drift \( \mathbf{b}(\mathbf{x}) = -\nabla \Phi(\mathbf{x}) \), using an asymptotic expansion in \( \epsilon \),

\[
\begin{align*}
\langle P \rangle &= \frac{1}{4Dn\epsilon} \int_{\Omega} \frac{e^{-\frac{\Phi(\mathbf{x})}{\epsilon}} k(\mathbf{x})d\mathbf{x} + e^{-\frac{\Phi_{0}}{\epsilon}}}{\partial S} \\
\langle \tau \rangle &= \frac{1}{4Dn\epsilon} \int_{\Omega} \frac{e^{-\frac{\Phi(\mathbf{x})}{\epsilon}} k(\mathbf{x})d\mathbf{x} + e^{-\frac{\Phi_{0}}{\epsilon}}}{\partial S}
\end{align*}
\]

(8)

where \( \Phi_{0} \) is the constant value of the radial potential \( \Phi(\mathbf{x}) \) on the centered nucleus where the nuclear pores are uniformly distributed. The range of validity of these asymptotic expressions has been explored with Brownian simulations for a single hole \[13\].

However, these formulas do not account for the possible interactions between the small absorbing pores, and for a large number of nuclear pores \( n \gg 1 \), \( \lim_{n \to \infty, n\epsilon^2 < 1} \langle \tau \rangle = 0 \), which shows the limitation of the previous formula. We thus propose to find the correction term that will account for the nuclear geometry.

Interactions between absorbing windows can drastically affect the MFPT \[11, 12\], and we propose here to account for these interactions and to extend the narrow escape time for a stochastic particle (with a drift) in the presence of a killing field \( k(\mathbf{x}) \) to reach one of the interacting absorbing windows located on the nucleus. We obtain new estimate for the probability \( \langle P \rangle \) and the associated conditional MFPT \( \langle \tau \rangle \). Both quantities are solutions of a linear system of partial differential equations, and when the number of holes is not too large \( (n \ll \log \left( \frac{a}{\epsilon} \right) \) ), we first solve the linear system of equation in the limit \( \epsilon \ll 1 \). Then, for a very large number of holes covering homogeneously the nucleus, we extend our analysis by using mean field approximations and obtain formulas for \( \langle P \rangle \) and \( \langle \tau \rangle \) that are valid for a large range of \( \epsilon \) and \( n \) values, and that generalize formula \[3\]. Finally we test our asymptotical results against Brownian simulations, and we apply our formula to model the trafficking of virus \( \mathbf{b} \neq 0 \) and non-viral gene vectors (Brownian diffusion \( \mathbf{b} = 0 \) ) that have to reach one of the \( n \approx 2,000 \gg 1 \) \[17\] nuclear pores covering the nucleus in order to deliver their genetic payload.

IV. ASYMPTOTIC DERIVATIONS FOR \( \langle \tau \rangle \) AND \( \langle P \rangle \)

We assume that the \( n \) absorbing windows \( \partial \Omega_{i} \) have the same radius \( \epsilon \), centered at positions \( (\mathbf{x}_{i})_{i=1}^{n} \). We shall present the method to compute the steady state SPDF \( p \) solution of \( \mathbf{b} \).

We shall use the Neumann-Green function \( \mathcal{N}(\mathbf{x}, \mathbf{x}_{0}) \) solution of the differential equation \[10\]

\[
\Delta \mathcal{N}(\mathbf{x}, \mathbf{x}_{0}) = -\delta_{\mathbf{x}_{0}}(\mathbf{x}), \: \mathbf{x} \in \Omega,
\]

\[
D \frac{\partial \mathcal{N}}{\partial n}(\mathbf{x}, \mathbf{x}_{0}) = -\frac{1}{|\partial \Omega|} \mathbf{x} \in \partial \Omega.
\]

Computing

\[
I = \int_{\Omega} \left( \Delta \tilde{p}(\mathbf{x}) - \nabla \cdot \mathbf{b}\tilde{p}(\mathbf{x}) - k\tilde{p}(\mathbf{x}) \right) \mathcal{N}(\mathbf{x}, \mathbf{x}_{0})d\mathbf{x}
\]

\[
- \int_{\Omega} \Delta \mathcal{N}(\mathbf{x}, \mathbf{x}_{0}) \tilde{p}(\mathbf{x})d\mathbf{x}
\]

where \( \tilde{p}(\mathbf{x}) = \int_{0}^{\infty} p(\mathbf{x}, t)dt \) is solution of the differential equation

\[
\Delta \tilde{p}(\mathbf{x}) - \nabla \cdot \mathbf{b}\tilde{p}(\mathbf{x}) - k(\mathbf{x})\tilde{p}(\mathbf{x}) = -p_{\tau}(\mathbf{x})
\]

(9)

with the boundary conditions

\[
\tilde{p}(\mathbf{x}) = 0 \text{ on } \partial N_{a} = \bigcup_{i=1}^{n} \partial \Omega_{i} \text{ and } \mathbf{J}(\mathbf{x}).n_{\mathbf{x}} = 0 \text{ on } \partial N_{ext} \bigcup (\partial S_{a} - \partial N_{a})
\]

(10)

where \( \mathbf{J}(\mathbf{x}) = -D\nabla \tilde{p}(\mathbf{x}) + \mathbf{b}(\mathbf{x})\tilde{p}(\mathbf{x}) \). Consequently, we have

\[
I = -\int_{\Omega} p_{\tau}(\mathbf{x}) \mathcal{N}(\mathbf{x}, \mathbf{x}_{0}) + \tilde{p}(\mathbf{x}_{0})
\]

(11)
and from Green’s identity
\[ I = -\int_{\partial N_a} \tilde{J}(x) \cdot n(x) N(x, x_0) dx + \int_{\Omega} b(x) \cdot \nabla N(x, x_0) \tilde{p}(x) dx. \]

Thus, we have
\[ \int_{\Omega} (k(x) \tilde{p}(x) - p_i(x)) N(x, x_0) dx = -\int_{\partial N_a} \tilde{J}(x) \cdot n(x) N(x, x_0) dx + \int_{\Omega} b(x) \cdot \nabla N(x, x_0) \tilde{p}(x) dx + \frac{1}{|\partial\Omega|} \int_{\partial\Omega} \tilde{p}(x) dx - \tilde{p}(x_0) \]

using a system of normalized orthogonal eigenfunctions \( \Phi_i(x, \epsilon) \), \( i = 0, 1, \ldots \), we expand \( p(x) \) as
\[ p(x, t) = \sum_{i=0}^{\infty} a_i(\epsilon) \Phi_i(x, \epsilon) e^{-\frac{t}{\lambda_i(\epsilon)}} \]

where \( \lambda_i(\epsilon) \) are the eigenvalues of the operator \( L_\epsilon = \Delta - \nabla \cdot b - k \) and \( a_i(\epsilon) \) the coefficients of the initial distribution \( p_i(x) \). In the limit of small absorbing boundary condition \( \epsilon \to 0 \), the Dirichlet boundary condition drops and for a small degradation rate \( k(x) \ll 1 \), the first eigenvalue \( \lambda_0 \) tends to 0, which is the first eigenvalue of \( L_\epsilon \) for \( k = 0 \) associated to the normalized eigenfunction
\[ \Phi_0(x, 0) = \frac{e^{-\frac{\phi(x)}{\epsilon}}}{\int_{\Omega} e^{-\frac{\phi(x)}{\epsilon}} dx}, \]

with a reflecting boundary condition boundary \( (\partial\Omega_\epsilon = \partial\Omega) \) and \( a_0(\epsilon) \to 1 \). Consequently, for a point \( x \in \Omega \) outside the boundary layer of the absorbing boundary, we obtain the leading order expansion
\[ \tilde{p}(x) = \int_0^{+\infty} p(x, t) dt \approx C_\epsilon e^{-\frac{\phi(x)}{\epsilon}} + O(1). \]

Because, \( \lambda_0(\epsilon) \) tends to zero and \( a_0(\epsilon) \) tends to one, as \( \epsilon \) goes to zero, we obtain that \( C_\epsilon = \frac{a_0(\epsilon)}{\lambda_0(\epsilon)} \int_{\Omega} e^{-\frac{\phi(x)}{\epsilon}} dx \) tends to \( +\infty \). Furthermore,
\[ q(x) = \int_0^{+\infty} tp(x, t) dt = \left( C_\epsilon^2 \int_{\Omega} e^{-\frac{\phi(x)}{\epsilon}} dx \right) e^{-\frac{\phi(x)}{\epsilon}} + O(1), \]

For smooth initial distributions \( p_i \), the integral
\[ \int_{\Omega} p_i(x) N(x, x_i) dx \]

is uniformly bounded as \( \epsilon \to 0 \) (and is an integrable singularity for non smooth distributions (see [23] for details)) while all other terms in \[12\] are unbounded. Consequently, in the small degradation rate \( k \ll 1 \) limit, the integral equation \[12\] is to leading order:
\[ \tilde{p}(x_0) + O(1) = -\int_{\partial N_a} \tilde{J}(x) \cdot n(x) N(x, x_0) dx + \int_{\Omega} b(x) \cdot \nabla N(x, x_0) \tilde{p}(x) dx + \frac{1}{|\partial\Omega|} \int_{\partial\Omega} \tilde{p}(x) dx. \]

For \( x_0 \) at a distance \( O(1) \) away from absorbing windows, \( N(x, x_0) \) is uniformly bounded for \( x \in \partial\Omega_\epsilon \). In addition, integrating \[9\] over \( \Omega \) we obtain:
\[ \int_{\partial N_a} \tilde{J}(x) \cdot n(x) dx = 1 - \int_{\Omega} k(x) \tilde{p}(x) dx = \langle P \rangle \in [0, 1]. \]
Consequently, for \( x_0 \) at a distance \( O(1) \) away from absorbing windows, \( \int_{\partial S_a} \tilde{J}(x) \cdot n_x N(x, x_0) dx \) is uniformly bounded, and

\[
\frac{1}{|\partial \Omega|} \int_{\partial \Omega} \tilde{p}(x) dx + \int_{\Omega} b(x) \cdot \nabla N(x, x_0) \tilde{p}(x) dx = C e^{-\frac{\phi(x_0)}{\epsilon}} + O(1). \tag{18}
\]

Consequently, (17) reduces to

\[
\tilde{p}(x_0) + O(1) = -\int_{\partial S_a} \tilde{J}(x) \cdot n_x N(x, x_0) dx + C e^{-\frac{\phi(x_0)}{\epsilon}}.
\]

To compute \( \int_{\partial S_a} \tilde{J}(x) \cdot n_x N(x, x_0) dx = \sum_{i=1}^{n} \int_{\partial \Omega_i} \tilde{J}(x) \cdot n_x N(x, x_0) dx \), we decompose the flux

\[
\left( \tilde{J}(x) \cdot n_x \right)_{x \in \partial \Omega_i} = g_i(x) + f_i(x),
\]

where the leading order \( g_i(s = |x - x_i|) \) is given by

\[
g_i(s) = \frac{g_0^i}{\sqrt{\epsilon^2 - s^2}}, \tag{20}
\]

and \( g_0^i \) a constant. \( f_i \) is a regular function such that

\[
\int_0^\epsilon f_i(s) ds = O(\epsilon g_0^i). \tag{21}
\]

Choosing \( x_0 = x_i \) at the absorbing boundary condition, we get that \( \tilde{p}(x_i) = 0 \). For \( i \neq j \), and \( |x_i - x_j| \gg \epsilon \) and that for \( x \in \partial \Omega_j \),

\[
N(x, x_i) = N(x_j, x_i) + O(\epsilon). \tag{22}
\]

Consequently, considering the flux expansion (19), we have:

\[
\int_{\partial S_a} \tilde{J}(x) \cdot n_x N(x, x_i) dx = \int_{\partial \Omega_i} (g_i(x) + f_i(x)) N(x, x_i) dx
\]

\[
+ \sum_{j=1,j \neq i}^{n} \left( N(x_j, x_i) + O(\epsilon) \right) \int_{\partial \Omega_j} (g_j(x) + f_j(x)) dx.
\]

For \( x_i \) on the domain boundary \( \partial S_a \), the Neumann-Green’s function \( N(x, x_i) \) can be written as (24):

\[
N(x, x_i) = \frac{1}{2\pi D|x - x_i|} + \frac{L(x_i) + N(x_i)}{8\pi D} \log \left( \frac{1}{|x - x_i|} \right) + \omega_{x_i}(x), \tag{23}
\]

where \( L(x_i) \) and \( N(x_i) \) are the principal curvatures of \( \partial S_a \) at \( x_i \), and \( \omega_{x_i}(x) \) is a bounded function of \( x \) in \( \Omega \).

When the absorbing patches colocalize on a small sphere of radius \( a \), the expansion (23) of the Neumann function \( N(x_i, x_j) \) does not stand any more because the second term \( \frac{-1}{4\pi a D} \log \left( \frac{1}{|x_i - x_j|} \right) \) can be much larger than the first term \( \frac{1}{2\pi D|x_i - x_j|} \) when \( |x_i - x_j| \approx a \), for \( a \ll |\Omega|^\frac{1}{2} \). For \( x \) and \( x_0 \) in the neighborhood of the sphere \( S_a \), we develop the Neumann function \( N(x, x_0) \) as

\[
\hat{N}(x, x_0) = \hat{N}(x, x_0) + O(1), \tag{24}
\]

where \( \hat{N}(x, x_0) \) is the Neumann function solution of:

\[
D \Delta \hat{N}(x, x_0) = -\delta(x - x_0), \text{ for } x \in \mathbb{R}^3
\]

\[
D \frac{\partial \hat{N}}{\partial n}(x, x_0) = 0, \text{ for } x \in S_a.
\]

We analytically solve the equation above in the appendix and we obtain that for \( x \) and \( x_0 \) located on the sphere \( S_a \), that is for \( |x_0| = |x| = a \), \( \hat{N}(x, x_0) \) is given by:

\[
\hat{N}(x, x_0) = \frac{1}{2\pi D|x - x_0|} + \frac{1}{4\pi a D} \log \left( \frac{|x - x_0|}{2a + |x - x_0|} \right). \tag{25}
\]
Consequently, if \( s = d(P, x_i) \) is the geodesic distance, the flux term \( \int_{\partial N_a} \tilde{J}(x) \cdot n_x N(x, x_i) dx \) is given by:

\[
\int_{\partial N_a} \tilde{J}(x) \cdot n_x N(x, x_i) dx = \int_0^\epsilon \left( \frac{g_0^i}{\sqrt{\epsilon^2 - s^2}} + f_i(s) \right) \frac{1}{2\pi Ds} + \frac{1}{\pi a} \log \left( \frac{s}{2a + s} \right) + O(1) 2\pi ds + \sum_{j=1, j \neq i}^n (N(x_j, x_i) + O(\epsilon)) \int_0^\epsilon \left( \frac{g_0^j}{\sqrt{\epsilon^2 - s^2}} + f_j(s) \right) 2\pi ds.
\]

Using condition (21), we obtain:

\[
\int_{\partial N_a} \tilde{J}(x) \cdot n_x N(x, x_0) dx = \frac{g_0^i}{D} \left( \frac{\pi}{2} + \frac{\epsilon}{2a} \log \left( \frac{\epsilon}{a} \right) + O(\epsilon) \right) + 2\pi \epsilon \sum_{j=1, j \neq i}^n N(x_j, x_i) g_0^j (1 + O(\epsilon)) .
\]

Given that the constant flux \( g_0^i \) is of the order \( g_0^i = O \left( \frac{1}{\epsilon a} \right) \), and that \( N(x_j, x_i) = O \left( \frac{1}{|x_i - x_j|} \right) = O \left( \frac{1}{\epsilon} \right) \), we rewrite the expression above,

\[
\int_{\partial N_a} \tilde{J}(x) \cdot n_x N(x, x_0) dx = \frac{g_0^i}{D} \left( \frac{\pi}{2} + \frac{\epsilon}{2a} \log \left( \frac{\epsilon}{a} \right) \right) + 2\pi \epsilon \sum_{j=1, j \neq i}^n N(x_j, x_i) g_0^j + O \left( \frac{\epsilon}{a} \right) + O \left( \frac{1}{\epsilon} \right) .
\]

Injecting (26) in (19), for \( x_0 = x_i \), we obtain the relation for the coefficients:

\[
\left( \frac{\pi}{2D} + \frac{\epsilon}{2aD} \log \left( \frac{\epsilon}{a} \right) \right) g_0^i + 2\pi \epsilon \sum_{j=1, j \neq i}^n N(x_j, x_i) g_0^j = C e^{-\frac{\phi(x_i)}{D}} + O(1) .
\]  

Finally, using the flux expansion (19) and approximation (14) for the function \( \tilde{p} \) in the conservation equation (17) lead to

\[
2\pi \epsilon \sum_{i=1}^n g_0^i = 1 - C \epsilon \int_{\Omega} k(x)e^{-\frac{2\phi(x)}{D}} dx + O(1) .
\]  

Finally we obtain a linear system of \( n + 1 \) equations (26) and (27) for the flux constant \( g_0^i \) \((i \leq i \leq n)\), and for the parameter \( C_n \), summarized as

\[
\begin{cases}
\left( \frac{\pi}{2D} + \frac{\epsilon}{2aD} \log \left( \frac{\epsilon}{a} \right) \right) g_0^i + 2\pi \epsilon \sum_{j=1, j \neq i}^n N(x_j, x_i) g_0^j = C e^{-\frac{\phi(x)}{D}} + O(1) , \text{ for } 1 \leq i \leq n
\end{cases}
\]

\[
2\pi \epsilon \sum_{i=1}^n g_0^i = 1 - C \epsilon \int_{\Omega} k(x)e^{-\frac{2\phi(x)}{D}} dx + O(1) ,
\]

To obtain \( C_n \), \( \langle P \rangle \) and \( \langle \tau \rangle \), we shall solve the linear system of equations (28) in the asymptotic limit \( \epsilon \) small. Indeed, injecting the asymptotic expressions (14) and (15) for \( \tilde{p}(x) \) and \( q(x) \) in expressions (6) and (7), we obtain

\[
\langle P \rangle = 1 - C \epsilon \int_{\Omega} k(x)e^{-\frac{\phi(x)}{D}} dx \text{ and } \langle \tau \rangle = C \int_{\Omega} e^{-\frac{\phi(x)}{D}} dx .
\]

V. MEAN FIELD APPROXIMATION: ASYMPTOTICS OF \( \langle \tau \rangle \) AND \( \langle P \rangle \) FOR \( n \gg \frac{1}{\epsilon} \)

We obtain here approximation for \( \langle P \rangle \) and \( \langle \tau \rangle \) by considering that \( n \gg 1 \), and that holes are distributed with spatial density \( \rho(x) \) over the spherical nucleus \( S_a \).

Summing the equations Eq. (26) for \( 1 \leq i \leq n \) and re-ordering terms we obtain that

\[
\left( \frac{\pi}{2D} + \frac{\epsilon}{2aD} \log \left( \frac{\epsilon}{a} \right) \right) \sum_{i=1}^n g_0^i + 2\pi \epsilon \sum_{i=1}^n g_0^i \sum_{j=1, j \neq i}^n N(x_j, x_i) = C \epsilon \sum_{i=1}^n e^{-\frac{\phi(x_i)}{D}} + O(n) .
\]
We suppose that the charge \( x_i \) is located at the north pole. The distance \(|x_i - x_j|\) with the \( j^{th} \) charge located at \( x_j(\theta, \phi) \) is then equal to \(|x_i - x_j| = 2a \sin \left( \frac{\phi}{2} \right)\) and the Neumann function \( N(x_j, x_i) \) is then equal to

\[
\hat{N}(x_j(\theta, \phi), x_i) = \frac{1}{4\pi a D} \left( \frac{1}{\sin \left( \frac{\phi}{2} \right)} + \log \left( \frac{\sin \left( \frac{\phi}{2} \right)}{1 + \sin \left( \frac{\phi}{2} \right)} \right) \right).
\]

(31)

Then, given that the probability density function of the \( j \neq i \) holes (i being at the north pole) is given by \( \rho_i(\phi), \) with \( 2\pi a^2 \int_0^{\pi} \rho_i(\phi) \sin(\phi) d\phi = 1, \) we have

\[
\lim_{n \to \infty} \sum_{j=1, j \neq i}^n N(x_j, x_i) = 2n \pi a^2 \int_0^{\pi} \frac{1}{4\pi a D} \left( \frac{1}{\sin \left( \frac{\phi}{2} \right)} + \log \left( \frac{\sin \left( \frac{\phi}{2} \right)}{1 + \sin \left( \frac{\phi}{2} \right)} \right) \right) \rho_i(\phi) \sin(\phi) d\phi
\]

(32)

that is

\[
\lim_{n \to \infty} \sum_{j=1, j \neq i}^n N(x_j, x_i) = \frac{na}{2D} \int_0^{\pi} \left( \frac{1}{\sin \left( \frac{\phi}{2} \right)} + \log \left( \frac{\sin \left( \frac{\phi}{2} \right)}{1 + \sin \left( \frac{\phi}{2} \right)} \right) \right) \rho_i(\phi) \sin(\phi) d\phi.
\]

(33)

In addition, we can also approximate

\[
\lim_{n \to \infty} \sum_{i=1}^n e^{-\frac{g_i(x_i)}{D}} = na^2 \int_0^{2\pi} \int_0^{\pi} e^{-\frac{g(\phi, \theta)}{D}} \rho(\phi, \theta) d\phi d\theta,
\]

(34)

where \( \rho(\phi, \theta) \) is the probability density function of all holes among the spherical nucleus \( S_a. \)

Denoting the integrals

\[
I_i = \int_0^{\pi} \left( \frac{1}{\sin \left( \frac{\phi}{2} \right)} + \log \left( \frac{\sin \left( \frac{\phi}{2} \right)}{1 + \sin \left( \frac{\phi}{2} \right)} \right) \right) \rho_i(\phi) \sin(\phi) d\phi,
\]

(35)

and

\[
I_2 = \int_0^{2\pi} \int_0^{\pi} e^{-\frac{g(x, \theta)}{D}} \rho(\phi, \theta) \sin(\phi) d\phi d\theta,
\]

(36)

we obtain that

\[
\left( \frac{\pi}{2D} + \frac{\epsilon}{2aD} \log \left( \frac{\epsilon}{a} \right) \right) \sum_{i=1}^n g_i + \frac{na \pi \epsilon}{D} \sum_{i=1}^n g_i I_i^1 = C \cdot na^2 I_2 + O(n).
\]

(37)

When holes are identically distributed among the nucleus, then \( I_i^1 = I_1 \) for all \( 1 \leq i \leq n, \) and using the conservation equation\[^{[27]}\] in equation\[^{[37]}\] we obtain

\[
\left( \frac{1}{4D\epsilon} + \frac{1}{4\pi a D} \log \left( \frac{\epsilon}{a} \right) + \frac{na}{2D} \right) \left( 1 - C \int_\Omega k(x) e^{-\frac{g(x)}{D}} dx \right) = C \cdot na^2 I_2 + O(n).
\]

(38)

leading to

\[
C \cdot e = \frac{\pi a + \epsilon \log \left( \frac{\epsilon}{a} \right) + 2n \pi a^2 \epsilon I_1}{\left( \pi a + \epsilon \log \left( \frac{\epsilon}{a} \right) + 2n \pi a^2 \epsilon I_1 \right) \int_\Omega k(x) e^{-\frac{g(x)}{D}} dx + 4\pi n a D \epsilon I_2}
\]

(39)

To simplify expression above and obtain concise expressions for the probability \( \langle P \rangle \) and the MFPT \( \langle \tau \rangle \) of a virus to a single nuclear pore, we are now considering two typical distributions of holes: 1- randomly distributed and 2- homogeneously distributed (regular organization).

1- Random distribution of holes among the sphere
We consider that the \( n \gg 1 \) holes are randomly distributed among the sphere and that they are non-overlapping. Thus, the probability distribution of holes on the sphere is given by
\[
\rho(\phi, \theta) = \rho_i(\phi) = \frac{1}{4\pi a^2} \mathbf{1} \left\{ \phi > 2 \arcsin \left( \frac{\epsilon}{a} \right) \right\}
\]
for all \( 1 \leq i \leq n \), the condition \( \mathbf{1} \{ \phi > 2 \arcsin \left( \frac{\epsilon}{a} \right) \} \) ensuring that holes are non-overlapping. Making the change of variable \( y = \sin \left( \frac{\phi}{2} \right) \), we re-write \( I_1 \) as
\[
I_1 = \frac{1}{\pi a^2} \int_{\frac{\pi}{2}}^{\frac{\pi}{2}} \left( \frac{1}{y} + \log \left( \frac{y}{1+y} \right) \right) y \, dy,
\]
that is
\[
I_1 = \frac{1}{2\pi a^2} \left[ x + \log (1+x) + x^2 \log \left( \frac{x}{1+x} \right) \right]_{\frac{\pi}{2}}^{1},
\]
leading to
\[
I_1 = \frac{1}{2\pi a^2} \left( 1 - 2 \frac{\epsilon}{a} - \frac{\epsilon^2}{a^2} \log \left( \frac{\epsilon}{a} \right) \right).
\]
In addition, we have
\[
I_2 = \frac{1}{4\pi a^2} \int_0^{2\pi} \int_{\arcsin \left( \frac{\epsilon}{a} \right)}^{\pi} e^{-\frac{\Phi(\phi, \theta)}{\beta}} \sin(\phi) \, d\phi \, d\theta.
\]
Replacing \( I_1 \) and \( I_2 \) by their expressions (43 and 44) in equation 39, we obtain that the leading order of \( C_\epsilon \) for randomly distributed holes is given by
\[
C_{\epsilon, \text{random}} = \frac{\pi a + \epsilon \left( 1 - \frac{n \epsilon^2}{a^2} \right) \log \left( \frac{\epsilon}{a} \right) + n \epsilon \left( 1 - 2 \frac{\epsilon}{a} \right)}{\left( \pi a + \epsilon \log \left( \frac{\epsilon}{a} \right) + n \epsilon \left( 1 - 2 \frac{\epsilon}{a} \right) \right) \int_{\Omega} k(x) e^{-\frac{\Phi(x)}{\beta}} \, dx + n a D \epsilon \int_0^{2\pi} \int_{\arcsin \left( \frac{\epsilon}{a} \right)}^{\pi} e^{-\frac{\Phi(\phi, \theta)}{\beta}} \sin(\phi) \, d\phi \, d\theta}.
\]

2- Homogeneous distribution of holes among the nucleus \( S_a \)

When holes are homogeneously distributed among the sphere, we have that \( \rho(\phi) = \mathbf{1} \{ \phi > \arccos \left( 1 - \frac{2}{n} \right) \} \frac{1}{4\pi a^2} \), leading to
\[
I_1 = \frac{1}{2\pi a^2} \left[ x + \log (1+x) + x^2 \log \left( \frac{x}{1+x} \right) \right]_{\arccos \left( 1 - \frac{2}{n} \right)}^{1},
\]
that is, for \( n \gg 1 \)
\[
I_1 = \frac{1}{2\pi a^2} \left( 1 - 2 \frac{\epsilon}{\sqrt{n}} + \frac{n}{2} \right).
\]
Moreover, we have
\[
I_2 = \frac{1}{4\pi a^2} \int_0^{2\pi} \int_{\arcsin \left( \frac{\epsilon}{a} \right)}^{\pi} e^{-\frac{\Phi(x)}{\beta}} \, dx \sin(\phi) \, d\phi \, d\theta.
\]
Replacing \( I_1 \) and \( I_2 \) by their expressions (47 and 48) in equation 39, we obtain that the leading order of \( C_\epsilon \) for homogeneously distributed holes is given by
\[
C_{\epsilon, \text{homogen}} = \frac{\pi a + \epsilon \log \left( \frac{\sqrt{n} \epsilon}{a} \right) + n \epsilon \left( 1 - \frac{2}{\sqrt{n}} \right)}{\left( \pi a + \epsilon \log \left( \frac{n \epsilon}{a} \right) + n \epsilon \left( 1 - 2 \frac{\epsilon}{a} \right) \right) \int_{\Omega} k(x) e^{-\frac{\Phi(x)}{\beta}} \, dx + n a D \epsilon \int_0^{2\pi} \int_{\arcsin \left( \frac{\epsilon}{a} \right)}^{\pi} e^{-\frac{\Phi(\phi, \theta)}{\beta}} \sin(\phi) \, d\phi \, d\theta}.
\]
Reinjecting the formula [45] or [49] in the expressions [29] of the probability and the MFPT that a single virus reaches a nuclear pore, we obtain that

\[
\langle P \rangle = \frac{\int_{0}^{2\pi} \int_{0}^{\pi} e^{-\frac{\Phi(x,\theta)}{2D}} \sin(\phi)d\phi d\theta}{\left(\pi + \log \left(\frac{\pi}{a}\right) + \nu \left(1 - 2\alpha_0 - \alpha_0^2 \log(\alpha_0)\right)\right) \int_{\Omega} k(x) e^{-\frac{\Phi(x)}{2D}} dx + naDe \int_{0}^{2\pi} \int_{0}^{\pi} e^{-\frac{\Phi(x,\theta)}{2D}} \sin(\phi)d\phi d\theta},
\]

(50)

and

\[
\langle \tau \rangle = \frac{\int_{0}^{2\pi} \int_{0}^{\pi} e^{-\frac{\Phi(x,\theta)}{2D}} \sin(\phi)d\phi d\theta}{\left(\pi + \log \left(\frac{\pi}{a}\right) + \nu \left(1 - 2\alpha_0 - \alpha_0^2 \log(\alpha_0)\right)\right) \int_{\Omega} k(x) e^{-\frac{\Phi(x)}{2D}} dx + naDe \int_{0}^{2\pi} \int_{0}^{\pi} e^{-\frac{\Phi(x,\theta)}{2D}} \sin(\phi)d\phi d\theta},
\]

(51)

where \(\alpha_0 = \frac{\nu}{\pi}\) when holes are randomly distributed among the spherical nucleus, and \(\alpha_0 = \frac{1}{\nu}\) when holes are homogeneously distributed.

When the drift is pointing towards the nucleus center and is then constant \(\Phi(x) = \Phi_0\) over the nucleus surface, then probability and MFPT formulas reduce to

\[
\langle P \rangle = \frac{4\pi naDe e^{-\frac{\Phi_0}{D}}}{\left(\pi + \log \left(\frac{\pi}{a}\right) + \nu \left(1 - 2\alpha_0 - \alpha_0^2 \log(\alpha_0)\right)\right) \int_{\Omega} k(x) e^{-\frac{\Phi(x)}{2D}} dx + 4\pi naDe e^{-\frac{\Phi_0}{D}}},
\]

(52)

and

\[
\langle \tau \rangle = \frac{\int_{0}^{2\pi} \int_{0}^{\pi} e^{-\frac{\Phi(x,\theta)}{2D}} \sin(\phi)d\phi d\theta}{\left(\pi + \log \left(\frac{\pi}{a}\right) + \nu \left(1 - 2\alpha_0 - \alpha_0^2 \log(\alpha_0)\right)\right) \int_{\Omega} k(x) e^{-\frac{\Phi(x)}{2D}} dx + 4\pi naDe e^{-\frac{\Phi_0}{D}}},
\]

(53)

When the drift is not directed towards the nucleus center but reaches its minimum \(\Phi(x) = \Phi_0\), we approximate \(I_2\) using the Laplace’s method in the small diffusion limit \(D \ll \Phi(x)\)

\[
I_2 = \frac{D}{2a^2 \det[-H_{\Phi}(x_0)]} e^{-\frac{\Phi_0}{2D}},
\]

(54)

where \(\det[-H_{\Phi}(x_0)]\) is the determinant of the Hessian matrix of potential \(\Phi\) at \(x_0\). The probability and MFPT of a virus to a nuclear pore are then given by

\[
\langle P \rangle = \frac{2\pi naD^2 \epsilon \det^{-1}[-H_{\Phi}(x_0)] e^{-\frac{\Phi_0}{2D}}}{\left(\pi + \log \left(\frac{\pi}{a}\right) + \nu \left(1 - 2\alpha_0 - \alpha_0^2 \log(\alpha_0)\right)\right) \int_{\Omega} k(x) e^{-\frac{\Phi(x)}{2D}} dx + 2\pi naD^2 \epsilon \det^{-1}[-H_{\Phi}(x_0)] e^{-\frac{\Phi_0}{2D}}},
\]

(55)

and

\[
\langle \tau \rangle = \frac{\int_{0}^{2\pi} \int_{0}^{\pi} e^{-\frac{\Phi(x,\theta)}{2D}} \sin(\phi)d\phi d\theta}{\left(\pi + \log \left(\frac{\pi}{a}\right) + \nu \left(1 - 2\alpha_0 - \alpha_0^2 \log(\alpha_0)\right)\right) \int_{\Omega} k(x) e^{-\frac{\Phi(x)}{2D}} dx + 2\pi naD^2 \epsilon \det^{-1}[-H_{\Phi}(x_0)] e^{-\frac{\Phi_0}{2D}}},
\]

(56)

**Effect of holes coverage**

When the number of windows \(n \gg 1\) is large and distributed over a small surface \(S_a\), electrostatic arguments have led to the following estimation for the leading order term of the narrow escape time for a Brownian particle [20]

\[
\langle \tau \rangle_{ES} = \frac{|\Omega|}{D} \left(\frac{1}{C_{S_a}} + \frac{f(\sigma)}{4n\epsilon}\right),
\]

(57)

where \(|\Omega|\) is the volume of the domain where particles are freely diffusing, \(C_{S_a}\) is the capacity of the conducting surface \(\partial S_a\) where absorbing holes are distributed, and \(\sigma = \frac{N\pi e^2}{|\partial S_a|}\) is the fraction of the structure covered by the absorbing holes. When the conducting surface is a sphere of radius \(a\), then \(C_{S_a} = 4\pi a\), and the MFPT is given by

\[
\langle \tau \rangle_{ES} = \frac{|\Omega|}{D} \left(\frac{1}{4\pi a} + \frac{f(\sigma)}{4n\epsilon}\right).
\]

(58)

The function \(f(\sigma)\) is unknown and is given at leading order by \(f(\sigma) = 1\) [3]. Here, for a pure diffusing particle, without drift nor killing, our formula for the MFPT (eq. 51) reduces to

\[
\langle \tau \rangle_{\Phi=0,k=0} \approx \frac{|\Omega|}{D} \left(\frac{1}{4\pi a} + \frac{1}{4n\epsilon} \left(1 - \frac{n\epsilon}{\pi a} \left(2\alpha_0 - \alpha_0^2 \log(\alpha_0) + \frac{1}{n} \log \left(\frac{\epsilon}{a}\right)\right)\right)\right),
\]

(59)
where \( \alpha_0 = \frac{\sigma}{\pi} \) when holes are randomly distributed among the spherical nucleus, and \( \alpha_0 = \frac{1}{\sqrt{\pi}} \) when holes are homogeneously distributed. Thus, the function \( f(\sigma) \) is given by

\[
f(\sigma) = 1 - 8 \frac{\sigma}{\pi} + \frac{\epsilon}{a\pi} \log \left( \frac{\epsilon}{a} \right)^{1-4\sigma} + O \left( \frac{\epsilon}{a} \right)
\]

(60)

when non-overlapping absorbing holes are randomly distributed among the spherical nucleus, and

\[
f(\sigma) = 1 - 4 \frac{\sqrt{\sigma}}{\pi} + \frac{\epsilon}{a\pi} \log (\sqrt{\sigma}) + O \left( \frac{\epsilon}{a} \right)
\]

(61)

when absorbing holes are distributed homogeneously.

At this point, we make two remarks: First, for \( \sigma << 1 \), \( 8 \frac{\sigma}{\pi} < 4 \frac{\sqrt{\sigma}}{\pi} \), and the MFPT of a single particle to an absorbing hole is higher for randomly distributed holes compared to homogeneously distributed holes. Second, the two formulas (60) and (61) that we obtained for accounting for holes’ coverage in the MFPT formula are quite different for formulas that have been either intuitively with an effective medium treatment \( f(\sigma) = 1 - \sigma \) (27) or computed with Brownian simulations \( f(\sigma) = \frac{1 - \sigma}{1 + 3.8\sigma^{1/2}} \) (2). The discrepancy with previous Brownian simulations may come from the arrangement of holes among the sphere that has been simulated.

VI. TESTS AGAINST BROWNIAN SIMULATIONS

We consider a spherical cell (radius \( R \)) with a centered spherical nucleus \( S_a \) (radius \( a \)) uniformly covered by \( n \) small pure absorbing pores (radius \( \epsilon \)) (see Fig. 1(right)). We impose reflecting boundaries at the external membrane \( r = R \) and at the nuclear surface, excepting at nuclear pores \( \partial N_a = \bigcup_{i=1}^n \partial \Omega_i \), centered at randomly distributed locations \( (x_i)_{i=1}^n \). We consider a constant radial drift \( B \) directed toward the nucleus (potential \( \Phi(r) = -Br \)). To have concise expressions, we assume the killing rate is constant \( k(\mathbf{x}) = k_0 \) and consequently (62) and (63) lead to

\[
\langle P \rangle = \frac{e^{-\frac{n\sigma}{R}}}{\langle \tau \rangle_{\Phi=0, k=0}} \left( e^{-\frac{n\sigma}{R}} \left( \frac{D}{B} \right)^2 a^2 + D \right) a + 2 \left( \frac{D}{B} \right)^3 \right) - e^{-\frac{n\sigma}{R}} \left( \frac{D}{B} R^2 + 2 \left( \frac{D}{B} \right)^2 R + 2 \left( \frac{D}{B} \right)^3 \right) k + e^{-\frac{n\sigma}{R}},
\]

(62)

and

\[
\langle \tau \rangle = \frac{\langle \tau \rangle_{\Phi=0, k=0}}{\langle \tau \rangle_{\Phi=0, k=0}} \left( e^{-\frac{n\sigma}{R}} \left( \frac{D}{B} \right)^2 a^2 + D \right) a + 2 \left( \frac{D}{B} \right)^3 \right) - e^{-\frac{n\sigma}{R}} \left( \frac{D}{B} R^2 + 2 \left( \frac{D}{B} \right)^2 R + 2 \left( \frac{D}{B} \right)^3 \right) k + e^{-\frac{n\sigma}{R}}.
\]

(63)

In Fig. 2 we test these theoretical asymptotics as well as (6) against Brownian simulations for an increasing number of holes. The ratio \( \sigma = \frac{a^2}{\frac{\pi}{a}} \) of the nucleus surface covered by the absorbing windows is constant \( \sigma = 2\% \) (surface covered by 2,000 pores of 25nm diameter on the nucleus of a Chinese hamster ovary cell (17)). Numerical parameters are summarized in table I.

The nice agreement between the Brownian simulations and the new asymptotics (62) and (63) of \( P_n \) and \( \tau_n \) respectively is the central result of this article. We point out that the additive term that accounts for the interactions between the windows is crucial: for 100 windows, the new asymptotic for the conditioned MFPT \( \tau_n \approx 2\text{min.} \), which is very closed to simulations, is twice as large as the one derived in (10) \( \tau_n \approx 1\text{min.} \). In addition, \( \frac{|S_a|}{R^2} = \frac{a^2}{R^2} \) is not that small in our simulations, which confirms a large range of validity for the computed asymptotics. Finally, even if the surface covered by the absorbing windows is conserved, the conditioned MFPT to a window drastically decreases with \( n \): for \( n = 100 \) windows, the MFPT \( \tau_n \approx 2\text{min.} \) is divided by 2 compared to the single window case \( \tau_n \approx 4\text{min.} \).

VII. CONCLUSION

Interruption dynamics with alternative periods of free diffusion and directed motion along MTs characterizes many cellular transports. When the intermittent particle can be degraded through the ubiquitin-proteasome machinery or trapped by the crowded cytoplasm, we derived here new asymptotics for the probability \( P_n \) and the mean time \( \tau_n \) the particle reaches a small absorbing target among \( n \). These new asymptotics account for the geometrical interactions
FIG. 2. New theoretical asymptotics (62) and (63) (dashed line) for the probability $P_n$ (left) and the conditioned MFPT $\tau_n$ (right) are compared against Brownian simulations (solid line) for an increasing number of absorbing windows (the ratio $\sigma = \frac{n\pi\epsilon}{2\pi a^2}$ of the nuclear surface covered by nanopores is constant $\sigma = 2\%$ [17]). Asymptotics (6) that do not account for the interactions between the windows are also drawn (dotted line). 1000 random trajectories are simulated. The parameters are summarized in table I.

| Parameters | Description                               | Value                                                                 |
|------------|-------------------------------------------|-----------------------------------------------------------------------|
| $D$        | Diffusion constant of the virus           | $D = 1.3\mu m^2s^{-1}$ (as observed for the Associated-Adeno-Virus [22]) |
| $B$        | Drift                                    | $B = 0.2\mu ms^{-1}$                                                 |
| $\sigma$  | Percentage of the nuclear surface covered by the $n$ nuclear pores | $\sigma = 2\%$ [17]                                                    |
| $k$        | Degradation rate                          | $k = 1/360s^{-1}$ (10 times the rate observed for synthetic gene vectors [16]) |
| $R$        | Radius of the cell                        | $R = 15\mu m$ (for a Chinese hamster ovary cell)                       |
| $a$        | Radius of the nucleus                     | $a = 5\mu m$ [17]                                                     |

between the windows. When the targets colocalize on a small structure $S_a$, asymptotics of $P_n$ and $\tau_n$ are obtained in the limit $\frac{|S_a|}{|\Omega|} \ll 1$. In particular these formulas apply for DNA viruses that have to reach a small nuclear pore among the 2,000 that are distributed on the nucleus to deliver their genetic payload. These theoretical results are tested against Brownian simulations and we observe a very nice agreement between curves. In a future work, it would be very interesting to explore deeper the interactions between the small holes and get the dependency of both $P_n$ and $\tau_n$ to the coverage $\sigma$ intuited [27] or observed with simulations [2]. Quantifying viral movement in the cell cytoplasm would be very helpful for understanding the key limiting steps of infection and design optimal drugs and viral gene vectors [26].

VIII. APPENDIX

When the absorbing patches colocalize on a small sphere of radius $a$, the expansion (23) of the Neumann function $N(x_i, x_j)$ does not stand any more because the second term $\frac{-1}{4\pi a D} \log \left( \frac{1}{|x_i - x_j|} \right)$ can be much larger than the first term $\frac{1}{2\pi D |x_i - x_j|^2}$ when $|x_i - x_j| \approx a$, for $a \ll |\Omega|^3$. Consequently, the second log-term in the expansion of the Neumann function expression has to be carefully computed. For $x$ and $x_0$ in the neighborhood of the sphere $S_a$, we developed the Neumann function $N(x, x_0)$ as

$$N(x, x_0) = \tilde{N}(x, x_0) + O(1),$$

(64)
To compute that we rewrite

\[ \frac{\partial \tilde{\mathcal{N}}}{\partial n}(x, x_0) = 0, \text{ for } x \in S_a. \]

Consequently, to compute the log-term in the Neumann function expansion, we hereafter compute \( \tilde{\mathcal{N}}(x, x_0) \). In a first time, we decompose \( \tilde{\mathcal{N}}(x, x_0) = \frac{4\pi}{\rho} \delta(x - x_0) + \Phi(x, x_0) \) where \( \Phi \) is solution of the differential system:

\[ \Delta \Phi(x, x_0) = 0, \text{ for } x \in \mathbb{R}^3 \]

\[ \frac{\partial \Phi}{\partial n}(x, x_0) = -\frac{\partial}{\partial n} \left( \frac{1}{4\pi D} \right) \delta(x - x_0) \text{, for } x \in S_a. \]

To solve (65) we choose a coordinate system so that the source point \( x = x_0 \) is on the positive \( z \) axis. Then, since \( \rho \Phi = 0 \) and \( \Phi \) is axisymmetric, then \( \Phi \) admits the series expansion:

\[ \Phi(x, x_0) = \sum_{n=0}^{\infty} b_n(|x_0|) \frac{P_n(\cos(\theta))}{|x|^{n+1}}, \tag{65} \]

where \( P_n \) are the Legendre polynomials of integer \( n \), \( \theta \) is the angle between \( x \) and the north pole. \( b_n(|x_0|) \) are some coefficients that are determined upon satisfying the boundary condition in (65). We let \( \rho = |x| \) and calculate that for \( x \in S_a \):

\[ \frac{\partial \Phi}{\partial n}(x, x_0) = \frac{\partial \Phi}{\partial \rho}(\rho = a) = -\sum_{n=0}^{\infty} \frac{(n+1) b_n(|x_0|)}{a^{n+2}} P_n(\cos(\theta)). \tag{66} \]

On the other hand, for \( |x| < |x_0| \) we can develop:

\[ \frac{1}{4\pi D|x - x_0|} = \frac{1}{4\pi D} \sum_{n=0}^{\infty} \frac{|x|^n}{|x_0|^{n+1}} P_n(\cos(\theta)), \tag{67} \]

which leads to the boundary condition:

\[ -\frac{\partial}{\partial \rho} \left( \frac{1}{4\pi D|x - x_0|} \right) (\rho = a) = -\frac{1}{4\pi D} \sum_{n=0}^{\infty} \frac{na^{n-1}}{|x_0|^{n+1}} P_n(\cos(\theta)). \tag{68} \]

Reinjecting (66) and (68) in the boundary condition of (65), we obtain that for all \( n \geq 0 \):

\[ b_n(|x_0|) = \frac{1}{4\pi D} \frac{na^{2n+1}}{(n+1)|x_0|^{n+1}}. \tag{69} \]

The Neumann function \( \tilde{\mathcal{N}}(x, x_0) \) is then given by:

\[ \tilde{\mathcal{N}}(x, x_0) = \frac{1}{4\pi D|x - x_0|} + \frac{1}{4\pi D} \sum_{n=0}^{\infty} \frac{na^{2n+1}}{(n+1)|x_0|^{n+1}} P_n(\cos(\theta)), \tag{70} \]

that we rewrite

\[ \tilde{\mathcal{N}}(x, x_0) = \frac{1}{4\pi D|x - x_0|} + \frac{1}{4\pi D} \sum_{n=0}^{\infty} \left( \frac{a^{2n+1}}{|x|^{n+1}|x_0|^{n+1}} - \frac{a^{2n+1}}{(n+1)|x|^{n+1}|x_0|^{n+1}} \right) P_n(\cos(\theta)). \]

using the development (67), we have:

\[ \frac{1}{4\pi D} \sum_{n=0}^{\infty} \frac{a^{2n+1}}{|x|^{n+1}|x_0|^{n+1}} P_n(\cos(\theta)) = \frac{a}{4\pi D|x_0||x - a^2 x_0|} \]

To compute \( I(\rho) = -\sum_{n=0}^{\infty} \frac{a^{2n+1}}{(n+1)|x_0|^{n+1}} P_n(\cos(\theta)) \), we notice that

\[ I'(\rho) = \sum_{n=0}^{\infty} \frac{a^{2n+1}}{\rho^{n+2}|x_0|^{n+1}} P_n(\cos(\theta)) = \frac{a}{\rho|x_0||x - a^2 x_0|}. \]
that is

\[ I'(r) = \frac{1}{\rho a \left( 1 + \frac{|x_0|^2 r^2}{a^4} - 2 \frac{|x_0| r}{a^2} \cos(\theta) \right)^\frac{3}{2}}. \]  

(73)

Because \( \lim_{r \to \infty} l(r) = 0 \), we have:

\[ l(r) = - \int_r^\infty I'(s) ds = - \int_r^\infty \frac{ds}{sa \left( 1 + \frac{|x_0|^2 s^2}{a^4} - 2 \frac{|x_0| s}{a^2} \cos(\theta) \right)^\frac{3}{2}}. \]  

(74)

Some computations lead to:

\[ l(r) = \frac{1}{a} \log \left( \frac{|x_0| r}{a^2} (1 - \cos(\theta)) \right) \left( 1 - \frac{|x_0| r}{a^2} \cos(\theta) + \left( 1 + \frac{|x_0|^2 r^2}{a^4} - 2 \frac{|x_0| r}{a^2} \cos(\theta) \right)^\frac{3}{2} \right). \]  

(75)

Finally, the Neumann function \( \tilde{N}(x, x_0) \) solution of the differential system [65] is given by:

\[ \tilde{N}(x, x_0) = \frac{1}{4\pi D|x - x_0|} - \frac{1}{4\pi D|x - x_0| \cdot |x - \frac{x_0}{|x_0|^2}|} \]

\[ + \frac{1}{4\pi a D} \log \left( \frac{|x_0| |x|}{a^2} (1 - \cos(\theta)) \right) \left( 1 - \frac{|x_0| |x|}{a^2} \cos(\theta) + \left( 1 + \left( \frac{|x_0|^2 |x|^2}{a^4} \right)^\frac{3}{2} - 2 \frac{|x_0| |x|}{a^2} \cos(\theta) \right)^\frac{3}{2} \right). \]

We point out that when \( x \) and \( x_0 \) are on the sphere \( S_a \), that is for \( |x_0| = |x| = a \), \( \tilde{N}(x, x_0) \) is simply given by:

\[ \tilde{N}(x, x_0) = \frac{2\pi D|x - x_0|}{2a} + \frac{1}{4\pi a D} \log \left( \frac{|x - x_0|}{2a + |x - x_0|} \right). \]

In addition, for \( a \ll |\Omega|^{1/3} \), we can show that the MFPT \( \tau_a \) of a Brownian particle (diffusion coefficient \( D \)) to the small sphere \( S_a \) is asymptotically equal to:

\[ \tau_a = \frac{|\Omega|}{4\pi a^2} \int_{S_a} \tilde{N}(x, x_0), \]

(76)

that is

\[ \tau_a = \frac{|\Omega|}{4\pi D} \int_0^{\pi} \left( \frac{1}{2a \sin(\theta/2)} + \frac{1}{2a} \log \left( \frac{\sin(\theta/2)}{1 + \sin(\theta/2)} \right) \right) \sin(\theta) d\theta = \frac{\Omega}{4\pi a D}. \]

(77)

We thus recover the well-known formula:

\[ \tau_a = \frac{\Omega}{C_a D}, \]

where \( C_a = 4\pi a \) is the capacitance of the sphere \( S_a \).

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