Chapter 1

Spatially inhomogeneous search strategies

Anne Hafner and Heiko Rieger
Department of Theoretical Physics and Center for Biophysics,
Saarland University, 66123 Saarbrücken, Germany
h.rieger@mx.uni-saarland.de

The efficiency of intracellular reactions which are driven by motor-assisted transport strongly depends on the spatial organization of the cytoskeleton. The cytoskeleton is a highly complex filament network which is generally neither homogeneous nor isotropic. In cells with a centrosome, microtubules emanate radially from the center, whereas actin filaments populate the cortex underneath the plasma membrane in a random manner. While intermittent search strategies with stochastic transitions between a slow reactive phase and a fast non-reactive phase have been shown to be advantageous in homogeneous, isotropic environments, the effect of a realistic global cytoskeleton topology has only very recently gained scientific interest. In this chapter we review the progress in analyzing the efficiency of spatially inhomogeneous search strategies.

1. Introduction

Random search processes are ubiquitous in nature and are fundamental to chemical kinetics: two reaction partners performing a random motion in space first have to find each other before they eventually can bind to each other. In particular at low concentrations of the reaction partner the random search process becomes the rate limiting factor. In conventional reaction-diffusion systems the reaction-partners are subject to thermal Brownian motion, and the efficiency of the search process depends solely on the diffusion constant D, the initial distance of the reaction partners R, the reaction range a and the size of the search domain V. For instance in three space dimensions the mean first passage time (MFPT) for a purely diffusive search process is $T^{3d}_{\mathrm{MFPT}} = V/(4\pi D)(a^{-1}-R^{-1})$ and in two dimensions $T^{3d}_{\mathrm{MFPT}} = A/(2\pi D)\ln R/a$.[1]

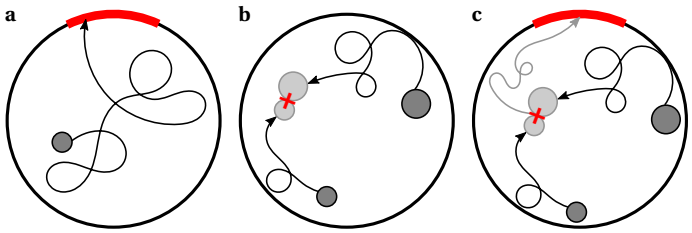

Fig. 1. Three standard search problems are encountered in cellular chemical kinetics: (**a**) the narrow escape problem which emerges during transport to a specific region on the domain boundary, (**b**) the reaction kinetics problem which considers the arrival of a searcher at a motile or immotile target in the bulk of the domain, and (**c**) the combined reaction-escape problem which occurs when cargo must be delivered to a narrow area on the boundary only after tethering to another particle beforehand.

In biological systems, in particular in living cells, the reaction kinetics is frequently enhanced by various mechanisms: One example are reactions involved in genomic transcription, where facilitated diffusion enhances the efficiency of the search of DNA-binding proteins for their specific binding site on a DNA molecule by alternating between linear 1-dimensional diffusion along the DNA molecule and 3-dimensional volume excursion events between successive dissociation from and rebinding to DNA.[2,3] Another prominent example is active intracellular transport of proteins and also larger objects like vesicles, endosomes and mitochondria, which are equipped with molecular motors that can randomly bind and unbind to the actin or microtubule filaments of the cell's cytoskeleton.[4] The resulting motion of these particles alternates stochastically between two modes: a diffusive mode and a ballistic mode along the direction of the filament when a molecular motor is bound. Both examples are also representative for intermittent search, which means that during the fast motility mode the searcher cannot find the target (i.e. particles cannot bind).[5]

It has been shown that switching between the two motility modes with certain rates can dramatically increase the search efficiency defined by the mean first passage time to find the target,[6,7] implying an enhanced reaction kinetics for molecular motor assisted search processes in cells.[8] The specific values of the transition rates between the two motility modes is commonly denoted as a "search strategy" to remind one of the fact that parameters could be varied to optimize the search efficiency. If these parameters are constant in space, we denote this as a "spatially homogeneous" strategy, and when they can vary in space we denote this as a "spatially inhomogeneous" strategy. In this sense the cellular cytoskeleton represents, with respect to

motor assisted random search, a spatially inhomogeneous search strategy, since cytoskeletal filaments are not homogeneously distributed in space but have a characteristic spatial organization as detailed below.

In this chapter we will review the recent progress in analyzing the efficiency of spatially inhomogeneous search strategies. Their advantage is obvious whenever additional information about the target location is available, like a preferential location in a particular sub-volume of the search domain or at its boundary. But we will see that inhomogeneous search strategies can even be superior to homogeneous strategies in cases when the random target location is homogeneously distributed. In the following we will specifically address three standard search problems encountered in cellular chemical kinetics, as illustrated in Fig. 1: the narrow escape problem (the target is a specific area on the domain boundary), the reaction kinetics problem (the target is randomly distributed in the search domain), and the reaction-escape problem, a combination of the two former. Then we will discuss the generality of the results and give an outlook to future applications.

2. The cytoskeleton - a specific spatially inhomogeneous search strategy

The efficiency of motor assisted transport strongly depends on the spatial organization of the cytoskeleton. The cytoskeleton is a highly complex filament network which is generally neither homogeneous nor isotropic. In cells with a centrosome, microtubules emanate radially from the center, whereas actin filaments are accumulated with random orientations in the cortex underneath the plasma membrane.[4]

While intermittent search strategies with stochastic transitions between a slow reactive phase and a fast non-reactive phase have been shown to be advantageous in homogeneous, isotropic environments,[5–11] the effect of a realistic global cytoskeleton topology has only very recently gained scientific interest.

A spatially inhomogeneous diffusion constant is included in models of surface-mediated diffusion.[12–19] In Ref. 12, Bénichou *et al.* investigated the narrow escape problem in two- and three-dimensional spheres S of radius R. A particle performs alternating phases of bulk diffusion with diffusion constant D_2 and surface-mediated diffusion along the surface of the sphere ∂S with diffusion constant D_1. When detaching from the surface after an exponentially distributed timescale with rate λ, the particle is radially delo-

cated at a distance $a \ll R$ from the surface into the bulk, where it exhibits bulk diffusion until it reaches the surface again and eventually the small target on ∂S is detected. Remarkably, the MFPT of a searcher, which is initially uniformly distributed on ∂S, can be minimized as a function of the desorption rate λ in dependence of D_1/D_2. In Ref. 17, Calandre *et al.* further showed that also the MFPT to a target in the bulk, reminiscent of the reaction problem, can be minimized as a function of the desorption rate λ, if the surface diffusion constant D_1 is sufficiently large (in particular larger than the bulk diffusion constant D_2). Consequently, a spatial inhomogeneity of the diffusion constant can substantially increase the search efficiency.

Cherstvy *et al.* investigated the transport of particles from the center to the surface of a circular disk.[20] The particle performs Brownian motion with a diffusion constant $D(r) = D_0 \frac{A}{A+r^2}$, $A > 0$, which is a function of the radial position r. The diffusion constant is thus the highest close to the center and gradually decreases with increasing distance. For $r \gg A$, the diffusion constant scales like a power-law $D(r) \sim 1/r^2$, whereas for $r \ll A$ diffusion is almost Brownian. With the aid of computer simulations, Cherstvy *et al.* found that the timescale $t_{1/2}$, at which the fastest half of the population arrives at the membrane, is defined by two asymptotes. Namely, the one with the slowest diffusivity $D(r = R)$ and the one with average diffusivity $\langle D \rangle = \int_a^R D(r)\mathrm{d}r/(2(R^2 - a^2))$, such that $t_{1/2}$ scales like R^4 in the leading order.[20]

Ando *et al.* investigated the influence of the topology of the cytoskeleton on the transport efficiency of particles which travel from the nucleus to an arbitrary position alongside the membrane.[21] Their model system is a two dimensional circular disk of radius R=10 μm, which possesses a nucleus of radius R_n. Tracer particles are initially positioned on the surface of the nucleus. In the cytoplasm they perform Brownian motion with diffusion constant D=0.011 $\mu\mathrm{m}^2/s$. The cytoskeleton is modeled as a shell of width w whose inner radius is positioned at R_a. In order to account for active transport, the diffusion constant is increased within this shell to $D_a = 100D$. They found that the MFPT can be minimized for shells positioned close to the nucleus if $R_n \gtrsim R/4$. Ando *et al.* further explicitly simulated filaments with fixed length which are randomly distributed in the cytoplasm. Tracer particles experience alternations of ballistic motion alongside the filaments and diffusion in the bulk. They found that the transport efficiency from the nucleus to the membrane is increased if the filament polarities collectively point towards the membrane.

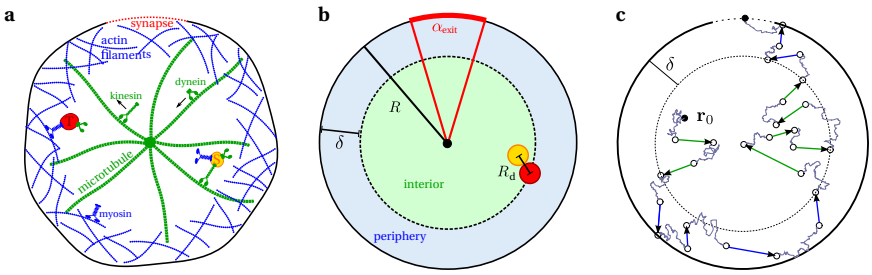

Fig. 2. **a** Targeted intracellular transport by molecular motors is a stochastic process which strongly depends on the spatially inhomogeneous organization of the cytoskeleton. **b** The cytoskeleton structure in a spherical cell of radius R is idealized by introducing a well-defined actin cortex of width δ. An exit zone on the cell boundary is characterized by the angle $\alpha_{\rm exit}$ and the detection distance of two particles is determined by $R_{\rm d}$. **c** In the narrow escape problem, particles which are initially located at $\boldsymbol{r}_0$ perform stochastic motion with alternating phases of ballistic and diffusive motion until they reach the exit. Parts a and b are reprinted from Ref. 25: Biophys. J., 114, A. Hafner and H. Rieger, 1420-1432, Copyright (2018), with permission from Elsevier. Part c is a reprinted figure with permission from Ref. 22: K. Schwarz et al., Phys. Rev. Lett., 117, 068101, 2016, (http://dx.doi.org/10.1103/PhysRevLett.117.068101). Copyright (2016) by the American Physical Society.

3. Spatially inhomogeneous cytoskeleton enhances intracellular reaction kinetics

In essence, the specific spatial organization of the cytoskeleton represents, in conjunction with motor-assisted transport, a search strategy in spatially inhomogeneous environments, which is intermittent if the searcher cannot find the target (i.e. bind to the reaction partner) in the ballistic mode. In order to study the efficiency of spatially inhomogeneous search strategies, a random walk model with two alternating motility modes was formulated in Refs. 22–25: (i) a ballistic motion state at velocity $\boldsymbol{v}$, which is associated to directed transport by molecular motors between binding and unbinding events, and (ii) a diffusive state with diffusivity D in which motors are unbound. Note that the case D=0 corresponds to a model with arrest states studied in Refs. 24,25. The limit of a vanishing diffusion constant is biologically relevant for intracellular cargo, such as vesicles, mitochondria, or macromolecules, which experience size-dependent subdiffusion in the crowded cytoplasm and thus undergo effectively stationary states.[26–29] But more importantly, since a single cargo is typically attached to several motor proteins concurrently, a full dissociation of the filament is rather unlikely.[30] Instead, arrest states at filament crossings are observed.[31–37] The

speed v is assumed to be constant. Transitions between the motility modes are determined by a constant attachment rate k and detachment rate k'. Generally, the rates can also be space-dependent.

The cytoskeleton structure in a spherical cell of radius R is idealized by the probability density $\rho_\Omega(\boldsymbol{r})$ to choose a direction Ω conditionally on the switch from the diffusive to the ballistic mode at position $\boldsymbol{r}$ and can, for simplicity, be parameterized as follows:

$$\rho_\Omega(\boldsymbol{r}) = \begin{cases} p\,\delta(\Omega-\Omega'(\boldsymbol{r})) + (1-p)\,\delta(\Omega-\Omega'(-\boldsymbol{r})), & \text{for } 0 < r < R-\delta, \\ 1/2\pi \text{ (in 2D)}; 1/4\pi \text{ (in 3D)}, & \text{for } R-\delta < r < R, \end{cases} \tag{1}$$

where $\Omega'(\boldsymbol{r})$ denotes the direction defined by the position vector $\boldsymbol{r}$ and p denotes the probability to move radially outwards. The probability p represents the contribution of kinesins and dyneins on the apparent motion of cargo. For p=1 the transport is solely managed by kinesins in the cell interior, and for p=0 only dyneins are active. In contrast p=0.5 is associated to an equal distribution of active kinesins and dyneins on the cargo. Instead of isotropic distributions in the periphery, the framework allows to study arbitrary distributions of actin filaments in the cortex, see Refs. 24,25. The exact distribution of orientations is objective of ongoing research, but it is reported that actin filaments align to microtubules[38] and are tangentially oriented to the membrane in cellular blebs.[39]

The distribution $\rho_\Omega(\boldsymbol{r})$ together with the state transition rates k and k' defines a search strategy which is generally inhomogeneous and anisotropic. However, δ=R_m leads to a spatially homogeneous and isotropic search strategy.

The proposed model allows the study of diverse search tasks. Here, the focus is on the three different, biologically relevant search problems mentioned in the introduction: the narrow escape, the reaction and the reaction-escape problem. At time $t = 0$ the particle starts at position $\boldsymbol{r_0}$, which may either be the cell center or a uniformly distributed position within the cell. Apart from the stochastic detachment events with rate k', a ballistically moving particle switches automatically to the diffusive mode at the MTOC (r=0), at the inner border of the actin cortex (r=R−δ), and at the cell membrane (r=R). The particle is propagated until termination of the respective search problem. For the narrow escape problem, the search is terminated when the particle hits the plasma membrane at the exit zone of opening angle α_exit, as illustrated in Fig. 2. In the case of the reaction problem, the search is terminated by encounter of searcher and target particle $|\boldsymbol{r}^\mathrm{S}-\boldsymbol{r}^\mathrm{T}| \leq R_\mathrm{d}$, see Fig. 2, if both particles are in the diffusive

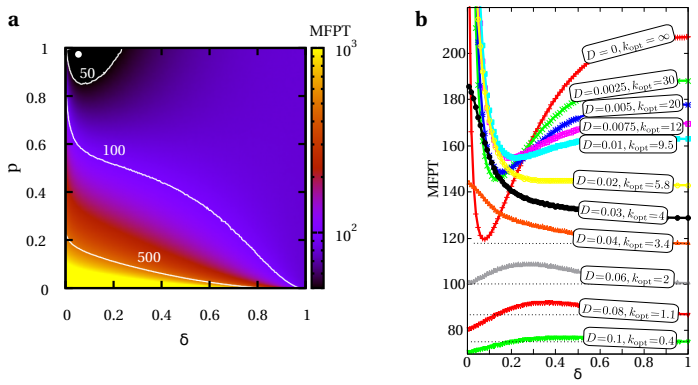

Fig. 3. Narrow escape problem for inhomogeneous spatial organizations of the cytoskeleton. **a** The MFPT versus δ and p for D=0, $k'_{\rm opt}$=0, and $k_{\rm opt}$=∞, which constitutes the optimal choice of transition rates for a spatially homogeneous cytoskeleton, i.e. δ=1, in the case of $\alpha_{\rm exit}$=0.1 and 2D domains. **b** MFPT as a function of δ for different diffusivities D and p=1 (i.e. exclusively radial outward transport in the ballistic mode) using the optimal rates $k'_{\rm opt}$=0 and $k_{\rm opt}(D)$ for δ=1 in the case of $\alpha_{\rm exit} \approx 0.1433$ and 3D domains. Part a is reprinted from Ref. 25: Biophys. J., 114, A. Hafner and H. Rieger, 1420-1432, Copyright (2018), with permission from Elsevier. Part b is a reprinted figure with permission from Ref. 22: K. Schwarz et al., Phys. Rev. Lett., 117, 068101, 2016, (http://dx.doi.org/10.1103/PhysRevLett.117.068101). Copyright (2016) by the American Physical Society.

state. In the reaction-escape problem searcher and target particle first have to react before the product particle can be transported to a specific zone on the membrane of the cell. The efficiency of a search strategy, defined by a specific cytoskeleton organization and transition rates, is measured in terms of the MFPT to target detection with the aid of an event-driven Monte Carlo algorithm.[40] In the following dimensionless spatial and temporal coordinates $\boldsymbol{r} \mapsto \boldsymbol{r}/R$, $t \mapsto vt/R$ and parameters $D \mapsto D/vR$, $k \mapsto Rk/v$, $k' \mapsto Rk'/v$, $\delta \mapsto \delta/R_{\rm m}$ are used.

3.1. *Narrow escape problem*

First, the search for a specific area on the domain boundary is considered, which is the so-called narrow escape problem.[41,42] A prominent example is directed secretion by immune cells which requires active transport of toxic vesicles towards the immunological synapse in order to kill tumorigenic or virus infected cells.[43–46]

In order to demonstrate the gain in search efficiency by a spatially inhomogeneous search strategy, corresponding to $0 < \delta < 1$, a homogeneous

cytoskeleton with δ=1 is considered to determine the optimal transition rates $k_{\rm opt}(D)$ and $k'_{\rm opt}(D)$. It turns out that the optimal detachment rate $k'_{\rm opt}$=0 is zero for all D, whereas the optimal attachment rate $k_{\rm opt}(D)$ increases with decreasing diffusivity D, such that $k_{\rm opt}$=∞ for D=0. Consequently, a motion pattern without directional changes in the bulk of the cell constitutes an optimal search strategy for a homogeneous cytoskeleton.

In Refs. 22–25, it is investigated whether an inhomogeneous filament structure ($\delta<1$) has the potential to solve the narrow escape problem more efficiently than its homogeneous counterpart. To answer this question, the influence of the actin cortex width δ and the probability for radially outward transport p on the MFPT is evaluated for the optimal parameters $k_{\rm opt}(D)$ and $k'_{\rm opt}$. Remarkably, for large probabilities of anterograde transport p the MFPT exhibits a minimum at small widths of the actin cortex $0<\delta<1$. The minimum is most pronounced for arrest states, i.e. D=0, but it is also found for $D>0$, as shown in Fig. 3. Moreover exclusive radial outward transport (p=1) in the ballistic mode represents the best strategy for the escape problem, which is plausible since the target location is on the boundary. This phenomenon is largely robust against changes in the transition rates k and k'. A small cortex width δ significantly reduces the MFPT, which emphasizes the general enhancement of the search efficiency by a spatially inhomogeneous filament structure. Note that for the narrow escape problem it is irrelevant whether the search is intermittent or not since the searching particle switches always to the diffusive mode when reaching the boundary, in particular when reaching the escape area.

3.2. *Reaction problem*

Next, the efficiency of spatially inhomogeneous search strategies for reaction with an immobile particle is addressed. When the searcher is in the diffusive mode and its position $\boldsymbol{r}$ comes closer to the target than $|\mathbf{r}^{\rm S}-\mathbf{r}^{\rm T}|\leq R_{\rm d}$ the search is successfully finished and for the moment is is assume that the search is intermittent, i.e. the searcher cannot find the target in the ballistic mode. The target position $\mathbf{r}^{\rm T}$ is either homogeneously distributed within the search domain or it is preferentially located in a specific sub-volume close to the center $r^{\rm T}\leq 0.5$ with probability w.

In contrast to the narrow escape problem, non-trivial optimal transition rates arise for the homogeneous reaction problem. The optimal attachment rate $k_{\rm opt}$ and detachment rate $k'_{\rm opt}$ depend on the diffusivity D and the detection radius $R_{\rm d}$. But for D=0, $k_{\rm opt}$=∞ (i.e. the absence of arrests) is

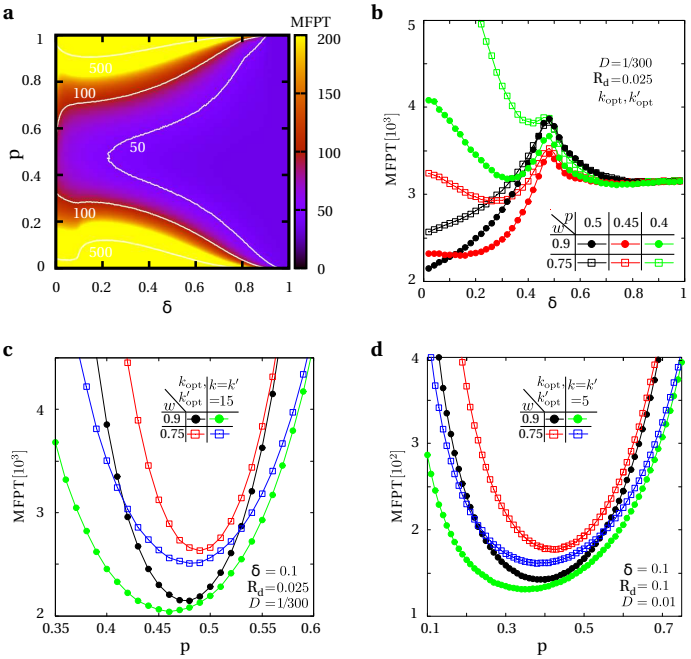

Fig. 4. Reaction problem for inhomogeneous spatial organizations of the cytoskeleton. **a** The MFPT versus δ and p for D=0 and R_d=0.1 in 2D spheres. The optimal transition rates for the homogeneous counterpart are applied, i.e. k'_opt=7, k_opt=∞. **b** MFPT as a function of δ for different values of p and w in a 3D search process with intermittent diffusion with D=1/300 and R_d=0.025. The optimal rates $k_\mathrm{opt}(D, R_\mathrm{d})$ from the homogeneous case δ=1 are applied. **c** MFPT as in (b) but as a function of p for a fixed cortex width δ=0.1 and different rates k and k' and probabilities w. **d** The same as in (c) but for D=0.01 and R_d=0.1. Part a is reprinted from Ref. 25: Biophys. J., 114, A. Hafner and H. Rieger, 1420-1432, Copyright (2018), with permission from Elsevier. Parts b-d are reprinted figures with permission from Ref. 22: K. Schwarz et al., Phys. Rev. Lett., 117, 068101, 2016, (http://dx.doi.org/10.1103/PhysRevLett.117.068101). Copyright (2016) by the American Physical Society.

optimal for all values of R_d.

In order to investigate the impact of inhomogeneous cytoskeleton organizations, the MFPT to an immobile bulk target is evaluated in Refs. 22–25 as shown in Fig. 4 in dependence of p_antero and δ, where the optimal transition rates for the homogeneous counterparts are applied. For all widths δ of the actin cortex, an optimal strategy to detect an immobile target within the cell is defined by p=0.5 even if the target is preferentially located close to the center. For a fixed cortex width δ=0.1 the MFPT is minimized for p close to 0.5 even for large w and also for fixed non-optimal

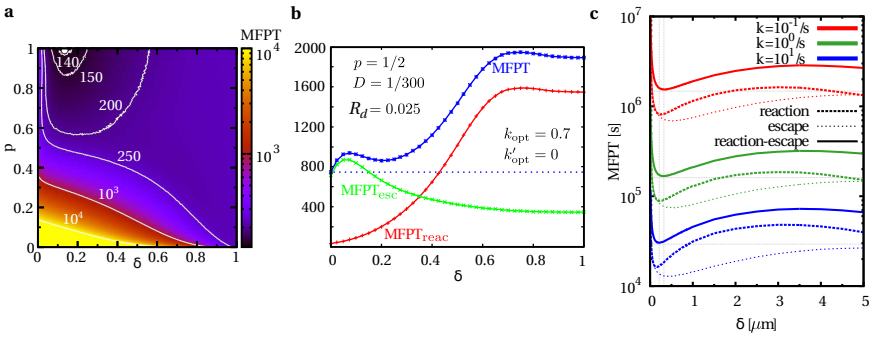

Fig. 5. Reaction-escape problem for inhomogeneous spatial organizations of the cytoskeleton. **a** The MFPT in dependence of the cortex width δ and the probability for radially outward transport p for 2D search processes with D=0. Transition rates $k'_{\rm opt}$=5, $k_{\rm opt}$=22, and parameters $R_{\rm d}$=0.1, $\alpha_{\rm exit}$=0.1 are applied. **b** MFPT for the reaction, the escape, and the combined reaction-escape problem as a function of δ in a 3D search process with intermittent diffusion with D=1/300. The optimal rates $k_{\rm opt}$, $k'_{\rm opt}$, and parameters p=0.5, $R_{\rm d}$=0.025, $\alpha_{\rm exit} \approx 0.1433$ are applied. **c** The MFPT of the escape, the reaction and the reaction-escape problem in dependence of δ for a three-dimensional cell of radius R=5 μm with $\alpha_{\rm exit}$=0.2, $R_{\rm d}$=0.1 μm, v=1 μm/s, $p_{\rm antero}$=1, k'=10/s, $k \in \{10^{-1}/{\rm s}; 10^{0}/{\rm s}; 10^{1}/{\rm s}\}$, and D=0. Parts a and c are reprinted from Ref. 25: Biophys. J., 114, A. Hafner and H. Rieger, 1420-1432, Copyright (2018), with permission from Elsevier. Part b is a reprinted figure with permission from Ref. 22: K. Schwarz et al., Phys. Rev. Lett., 117, 068101, 2016, (http://dx.doi.org/10.1103/PhysRevLett.117.068101). Copyright (2016) by the American Physical Society.

rates k, k', as shown in Fig. 4 c. While for D=0 a homogeneous isotropic cytoskeletal network with δ=1 is most efficient, as shown in Fig. 4 a, for $D \neq 0$ again a thin cortex $\delta \ll 1$ may yield a much smaller search time for p=0.5, as displayed in Fig. 4 b.

Note that similar results are obtained when the searcher can find the target also during the ballistic motion phase (i.e. non-intermittent search) and an optimal strategy for motile targets is given by p=0 and δ=0, as studied in Ref. 25 for D=0.

3.3. *Reaction-escape problem*

Finally, the efficiency of inhomogeneous search strategies for the combination of reaction and escape problem is discussed. Cargo first has to bind to a reaction partner before the product can be delivered to a specific area on the cell boundary. A prominent example is the docking of lytic granules at the immunological synapse of cytotoxic T-lymphocytes that requires the pairing with CD3 endosome beforehand.[47] The total MFPT of the reaction-

escape problem is composed of the $\mathrm{MFPT}_{\mathrm{react}}$ for the reaction problem and the $\mathrm{MFPT}_{\mathrm{escape}}$ for the following escape problem of the product particle to the exit zone on the membrane, i.e. $\mathrm{MFPT}=\mathrm{MFPT}_{\mathrm{reac}}+\mathrm{MFPT}_{\mathrm{esc}}$.

In order to explore the influence of the spatial inhomogeneity of the cytoskeleton, in Refs. 22–25 the MFPT for the reaction-escape problem is measured as a function of the cortex width δ for the transition rates k_{opt} and k'_{opt} which are optimal in the homogeneous case δ=1. Figure 5 a shows that in the case of D=0 a superior search strategy for the reaction-escape problem is defined by a high probability p of radially outward motion and a thin actin cortex δ. But a small cortex δ also reduces the total MFPT in comparison to the homogeneous strategy δ=1 for intermittent diffusion with $D \neq 0$. In general, inhomogeneous search strategies with $0 < \delta < 1$ which are more efficient than the homogeneous counterpart also exist for non-optimal transition rates k and k'. Remarkably, Fig. 5 c indicates that the optimal width of the actin cortex δ^{opt}=0.3 μm, predicted by our model under biologically reasonable conditions, is in good agreement to experimental data.[48,49]

4. Discussion and outlook

We reviewed the efficiency of spatially inhomogeneous intermittent search strategies and pointed out the importance of the spatial organization of the cytoskeleton for targeted intracellular transport, which occurs when cargo particles have to find reaction partners or specific target areas inside a cell. Remarkably, the confinement of randomly oriented filaments to a thin cortex is not a handicap for the cell, but can substantially increase the efficiency of diverse transport tasks. The best strategy for the narrow escape problem is to allow only radially outward transport from the center towards a thin cortex underneath the boundary, where multi-directional transport is possible. This thin cortex allows an accelerated random motion along the boundary to find the escape region. A similar result holds for the reaction problem, in which the target is located in the bulk of the domain: here again superior inhomogeneous strategies exist, but the optimal probability for radially outward transport is now around p=0.5. The reaction-escape problem combines both scenarios and the optimal forward/backward radial transport probability p depends on the size ratio of target and escape region. The basic mechanism underlying a higher search efficiency is actually reminiscent of an acceleration of purely diffusive search kinetics by following boundaries with an increased diffusivity.[12–19] For intracellular reaction

kinetics cells are able to economically realize efficient search strategies by intermittent transport on a cytoskeleton with specific spatial structure. Instead of supporting a resource demanding isotropic homogeneous filament network it is sufficient, and often even more efficient, to establish just a thin actin cortex underneath the cell membrane.

It is shown that first passage times of reactions in biological cells actually are broadly distributed, such that the most likely value may deviate significantly from the mean.[51–53] Consequently, it is worth studying the full distribution of first passage times for spatially inhomogeneous search strategies in more detail. And, in particular, considering multiple searcher and target particles within a cell is promising and opens a new range of questions. Spatially inhomogeneous search strategies potentially also reduce the cover time to several targets.[50] The study of extreme statistics (i.e. when does the first x particles arrive at a given target) is certainly also relevant for various biochemical reactions.

Acknowledgments

This work was financially supported by the German Research Foundation (DFG) within the Collaborative Research Center SFB 1027. We thank Karsten Schwarz for fruitful collaboration and Olivier Bénichou, Gleb Oshanin, Ralf Metzler, and Raphaël Voituriez for stimulating discussions.

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