Residence Time Calculation for Chemotactic Bacteria within Porous Media

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ABSTRACT Local chemical gradients can have a significant impact on bacterial population distributions within subsurface environments by evoking chemotactic responses. These local gradients may be created by consumption of a slowly diffusing nutrient, generation of a local food source from cell lysis, or dissolution of nonaqueous phase liquids trapped within the interstices of a soil matrix. We used a random walk simulation algorithm to study the effect of a local microscopic gradient on the swimming behavior of bacteria in a porous medium. The model porous medium was constructed using molecular dynamics simulations applied to a fluid of equal-sized spheres. The chemoattractant gradient was approximated with spherical symmetry, and the parameters for the swimming behavior of soil bacterium *Pseudomonas putida* were based on literature values. Two different mechanisms for bacterial chemotaxis, one in which the bacteria responded to both positive and negative gradients, and the other in which they responded only to positive gradients, were compared. The results of the computer simulations showed that chemotaxis can increase migration through a porous medium in response to microscopic-scale gradients. The simulation results also suggested that a more significant role of chemotaxis may be to increase the residence time of the bacteria in the vicinity of an attractant source.

INTRODUCTION

In the presence of a concentration gradient of attractant (often carbon sources such as sugars or amino acids), flagellated bacteria have been shown to bias their migration toward the source (Macnab and Koshland, 1972). In aqueous environments situations can exist that produce a local concentration gradient of an attractant chemical species. For example, consumption of a local food source by the bacterial population or generation from a carbon source such as decaying diseased cells can create an attractant gradient. Understanding the behavior of bacteria in the vicinity of local gradients of chemoattractants is important for predicting bacterial population dynamics.

Another important example of bacterial interaction with local gradients can occur with in situ bioremediation, a method for restoring contaminated soils by using bacteria to degrade pollutants. A chemical contaminant in ground water with a low solubility can create local concentration gradients (Mercer and Cohen, 1990; Powers et al., 1994; Wilson, 1994). For a range of droplet diameters (0.01–0.1 cm), the contaminant trichloroethylene diffused into the aqueous phase for up to 50 days (Wilson, 1994). For bioremediation the residence time of bacteria within the contaminated zone will be important for its removal. This residence time will depend on a combination of the random motility and chemotactic behavior of the bacteria. In porous media, where migration is complicated by the presence of obstacles in the medium, this chemotactic response may play a greater role.

In this paper we model a local gradient as a sphere of attractant with a steady-state gradient and use computer simulation to study the migration behavior of the soil bacteria *Pseudomonas putida* in a porous medium. The sphere of attractant is representative of three-dimensional diffusion of a chemical from a micropipette into an aqueous solution. In this way we examine the effect of chemotaxis on the residence time of *P. putida* within a local area represented by a sphere of known diameter.

P. putida propel themselves through their surrounding media by rotating flagella that form a tuft at one end of their body (Harwood et al., 1989). A single cell traces a path that consists of a series of runs interrupted by changes in direction. In the absence of a chemical gradient, this swimming pattern resembles a three-dimensional random walk similar to Brownian motion in molecular diffusion, except that changes in direction are due to the reversal of flagella rotation and not molecular collisions. This behavior is referred to as random motility.

In the presence of an increasing gradient of attractant, bacteria bias their migration toward the source by decreasing their frequency of changing direction (Berg and Brown, 1972). The response of bacteria when moving in the direction of a decreasing attractant gradient is not completely understood (Berg and Brown, 1972, 1974). Rivero et al. (1989) described an equation for the effect of an attractant gradient on the frequency of changing direction. They considered two scenarios for the situation when the bacteria move down an attractant gradient (i.e., away from an at-

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tractant source). The first assumed that the response to the negative gradient was generated by the same mechanism as the positive gradient, thus resulting in higher tumbling frequencies for negative gradients. The second assumed that the bacteria were insensitive to negative gradients and reverted back to the same tumbling frequency exhibited in the absence of an attractant gradient. Both possibilities were considered here for comparison.

Duffy et al. (1995) used a random walk algorithm to simulate the random motility of *P. putida* in a model porous medium. This method models the bacterial motion as a series of straight-line segments (runs) interrupted by turns (changes in direction). Comparison of the simulation to experimental results for *P. putida* migrating in porous media showed that the simulations are qualitatively equivalent and quantitatively similar (Duffy et al., 1995). A modification of this method to include chemotaxis is used here to study the residence time for bacteria in the presence of an attractant diffusing in three dimensions from a point source.

METHODS

Simulation methodology

The simulation methodology employed in this paper is a straightforward modification of that used by Duffy et al. (1995) to simulate the random motility of *P. putida* in a model porous medium, which in turn was a generalization of the cellular dynamics algorithm developed by Frymier et al. (1993) for simulating random motility and chemotactic response in bulk aqueous systems. We will briefly review the algorithm of Duffy et al. (1995), indicating the changes necessary to take chemotactic responses into account. The interested reader is directed to the original papers of Duffy et al. (1995) and Frymier et al. (1993) for further details.

A model porous medium is generated in three dimensions by using a simulation "box" filled with spheres of equal size, which act as obstacles. A random configuration of the spheres is created by using a molecular dynamics simulation of spheres (Allen and Tildesley, 1987). The free volume (or porosity, ϕ) of the medium is adjusted by increasing all sphere diameters by a fixed amount, and the porosity is calculated by using a Monte Carlo algorithm. Although overlap of the spheres is allowed, the amount of overlap is minimal for the fixed porosity of 0.37, at which all of the simulations are run. Each of 2048 spheres is assigned an equal diameter of 100 μm.

Initially, 10,000 cells are placed at the center of the simulation space. Each cell is given a random direction vector. After initialization, motility of a bacterial population is simulated by performing several steps on each bacterium separately. First, it is determined whether the cell changes direction. This depends on a frequency of turning β, which is specific to the species of bacteria under consideration. Second, if a new direction is required, then it is chosen by the method of Frymier et al. (1993) for the given turn angle distribution; otherwise the original direction is maintained. Third, the new position is calculated from the direction vector, the swimming speed, and the time step. For each pass of the algorithm, the time is incremented by the time step and the steps are repeated. The time is incremented by $\Delta t = 0.005$ s (equivalent to a step length of 0.22 μm at the swimming speed of 44 μm/s), which is equal to 0.0025 of the mean free run time of the simulated bacteria in the bulk (2 s, corresponding to a mean free path in the bulk of 88 μm). Thus the position r_i of bacterium i at time $t + \Delta t$ is given by

$$\mathbf{r}_i(t + \Delta t) = \mathbf{r}_i(t) + v\mathbf{s}_i\Delta t \tag{1}$$

where v is the three-dimensional swimming speed and $\mathbf{s}_i$ is the unit direction vector, which evolves in time according to

$$\mathbf{s}_i(t + \Delta t) = \lambda_i\mathbf{s}_i(t) + (1 - \lambda_i)\mathbf{s}_{i,\text{new}}(t) \tag{2}$$

where

$$\lambda_i = \begin{cases} 1 & \text{if } \rho_i \geq \beta\Delta t \\ 0 & \text{if } \rho_i < \beta\Delta t \end{cases} \tag{3}$$

Here ρ_i is a random number from the interval [0,1], and the new direction $\mathbf{s}_{i,\text{new}}(t)$ is found by randomly choosing θ_i, the angle between $\mathbf{s}_i(t)$ and $\mathbf{s}_{i,\text{new}}(t)$, from a distribution of turn angles measured by Duffy and Ford (1997). Thus each bacterium has a probability $\beta\Delta t$ of changing its direction and a probability of $(1 - \beta\Delta t)$ of continuing its run. Details of the method of choosing θ_i can be found in Frymier et al. (1993). The azimuthal angle is chosen from a uniform distribution. After the new position is computed, the fourth step is to check whether it falls in the void fraction of the porous medium. If it does, then the change in position is made; if not, the original position is maintained. In either case, the time is incremented by the time step and the steps are repeated. We believe this representation is consistent with observations by Frymier et al. (1995) of bacteria approaching an impenetrable boundary where bacteria swam in circles parallel to the surface for several seconds before returning to the bulk. The time of interaction with the surface calculated from the simulation algorithm is 3.5 s (Duffy, 1995) which is consistent with the qualitative observations reported by Frymier et al. (1995). Our modified algorithm successfully simulates the macroscopic migration properties of *P. putida* migration in porous media in the absence of an attractant, as was shown by comparison to experimental observations (Duffy et al., 1995).

Model equation for chemical gradient

Consider a source of attractant that is injected at a rate α within an aqueous medium. This is analogous to setting up an attractant gradient using a micropipette. The attractant source is simulated as originating from the center of the simulation box (Fig. 1). In the absence of convective flow and assuming spherical symmetry about the source, the diffusion equation for this situation is given by

$$\frac{\partial C}{\partial t} = D\frac{1}{r^2}\frac{\partial}{\partial r}\left[r^2\frac{\partial C}{\partial r}\right] \tag{4}$$

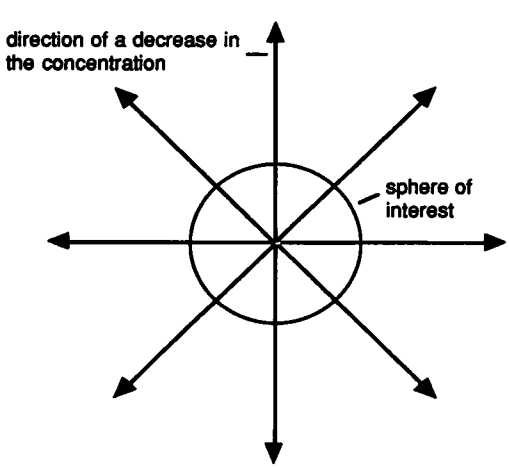

FIGURE 1 Two-dimensional representation of the three-dimensional medium with a point source of attractant at the center. The area of interest is a sphere with 0.2 cm diameter, shown by the circle.

where $C(r, t)$ is the concentration of attractant and D is the diffusion coefficient for the attractant. When the source is injected over a long period, the solution approaches its steady state ($\partial C/\partial t = 0$) limit, given by (Berg, 1993)

$$C(r) = \frac{\alpha}{4\pi D r} \tag{5}$$

Note that Eq. 5 is only an approximation to the attractant concentration in the presence of a porous medium. Its derivative represents an angular average of the local concentration gradients that result from the presence of the obstacles. We can argue that this approximation is reasonable and eliminates the need for laborious calculations accounting for the local gradients, if we note that diffusion times within the pores (~1 s) are much smaller than the overall diffusion time (~2200 s) within the sphere of interest.

Model equation for chemotactic response

Duffy and Ford (1997) showed that for *P. putida* the turn frequency β follows a Poisson process and is consequently equal to the inverse of the mean run time $\langle\tau\rangle$,

$$\beta = \frac{1}{\langle\tau\rangle} \tag{6}$$

Rivero et al. (1989) derived an equation relating the mean run time $\langle\tau\rangle$ to the attractant concentration C for bacterial migration in one dimension. This was extended to three dimensions by Frymier et al. (1993) to give

$$\ln\frac{\langle\tau\rangle}{\langle\tau_0\rangle} = \frac{\chi_0}{\nu}\frac{K_d}{(K_d + C)^2}\hat{\mathbf{s}}\cdot\nabla C \tag{7}$$

where K_d is the dissociation constant for bacterial receptor-attractant binding, χ_0 is the three-dimensional chemotactic sensitivity parameter, and $\langle\tau_0\rangle$ is the mean run time in the absence of an attractant gradient. Thus an expression relating attractant concentration C to the frequency of changing direction is

$$\beta = \beta_0\exp\left(-\frac{\chi_0}{\nu}\frac{K_d}{(K_d + C)^2}\hat{\mathbf{s}}\cdot\nabla C\right) \tag{8}$$

where β_0 is the basal frequency of changing direction in the absence of a chemical gradient. The chemotactic sensitivity parameter χ_0 is always positive, so that Eq. 8 holds for both positive and negative gradients.

The distribution of β given by Eq. 8 for a range of concentration gradients (based on Eq. 5) is given in Fig. 2 *A*. The solid lines in Fig. 2 are the values of β for the specific case of bacteria moving along the vector normal to the diffusion front. The values of β for a positive gradient cover a range that is realistic for *P. putida* in an attractant gradient. They correspond to experimental values found for *P. putida* in temporal gradients of 3-chlorobenzoate (3CB) (Harwood et al., 1990). These turn frequencies ranged from 0.1 to 0.5 s^{-1} for temporal gradients created by concentrations of 3CB in the range 2 mM to 0.02 mM (Harwood et al., 1990).

Equation 8 assumes that the bacteria use the same mechanism for positive and negative attractant gradients, except that the sensitivity is reversed. Another possibility observed for some bacteria is that they respond to swimming down an attractant gradient by reverting to the turning frequency in the absence of an attractant gradient, β_0 (Macnab and Koshland, 1972). Thus, in this situation, the frequency of changing direction returns to the basal rate when the bacteria face down-gradient. For this latter case Eq. 8 is replaced by

$$\beta = \begin{cases}\beta_0\exp\left(-\dfrac{2\chi_0}{\nu}\dfrac{K_d}{(K_d + C)^2}\hat{\mathbf{s}}\cdot\nabla C\right) & \text{for } \hat{\mathbf{s}}\cdot\nabla C > 0\\ \beta_0 & \text{for } \hat{\mathbf{s}}\cdot\nabla C \le 0\end{cases} \tag{9}$$

The necessity of including the approximate factor of 2 in Eq. 9 was shown by Frymier et al. (1994) and is related to the fact that the value of χ_0 that we will use was evaluated from experimental data, using a mathematical model that incorporated Eq. 8 rather than Eq. 9. Although this approximate factor of 2 was found empirically, it yields physically consistent results: both Eqs. 8 and 9 have the same ratio of β_{up}/β_{down}, where up and down refer to facing up and down the gradient. The distribution of β for a range of concentration gradients according to Eq. 9 is given in Fig. 2 *B*.

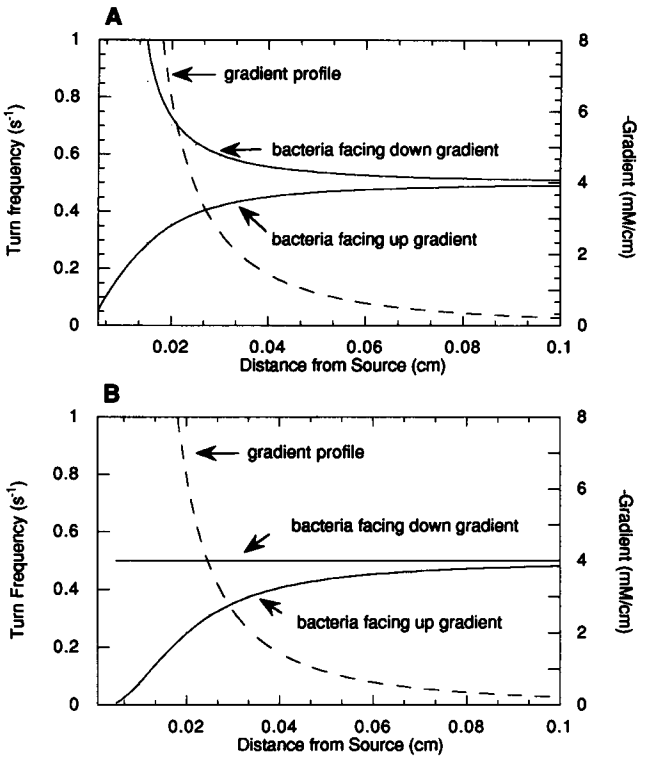

FIGURE 2 The turn frequency plotted as a function of distance from the source at the center of the simulation box along a line normal to the diffusion front (*solid lines and left axis*), using (*A*) Eq. 8 and (*B*) Eq. 9. The concentration gradient for the attractant is shown by the dashed line and corresponds to the right axis. Turn frequencies for bacteria moving up the gradient and down the gradient are distinguished.

Simulation parameters

We applied the cellular dynamics algorithm to an experimental system with which we have some familiarity—the response of *P. putida* to 3CB. The cell swimming speed (ν = 44 μm/s) and the turn frequency (β_0 = 0.5/s) are average values for *P. putida* reported by Harwood et al. (1989). The duration of a turn (t_0 = 0.025 s) is an average of the values reported by Harwood et al. (1989).

The model attractant is 3CB, which is a structural analog of benzoate and thus uses the same receptor system as benzoate to elicit a chemotactic response in *P. putida*. However, 3CB is not metabolized by *P. putida*. This simplifies the mathematical description by avoiding the creation of sec-

ondary gradients due to consumption. For 3CB the diffusion coefficient D_{3CB} is 1.24×10^{-5} cm^2/s (Barton and Ford, 1995). In the porous medium $D_{eff} = \phi D_{3CB}/T$, where T is the tortuosity. The porosity ϕ is 0.37 and the tortuosity T is approximately unity for small molecules such as 3CB. This yields an effective diffusion coefficient in the porous medium of $D_{eff} = 4.59 \times 10^{-6}$ cm^2/s. The source of 3CB was produced at a rate $\alpha = 3.5 \times 10^{-7}$ μmol/s.

The chemotactic sensitivity coefficient for *P. putida* responding to 3CB was reported by Barton and Ford (1995) to be $1.9 \pm 0.7 \times 10^{-4}$ cm^2/s. They determined this value by fitting experimental bacterial density profiles obtained in a stopped-flow diffusion chamber assay. The dissociation constant K_d for 3CB binding to *P. putida* receptors is 0.5 mM (Harwood et al., 1990).

RESULTS

Simulations of bacteria migration were performed for two different media: in bulk liquid media and in a porous medium consisting of a random assemblage of spheres. For each medium, residence times were determined with and without a chemotactic response. Results from simulations within the porous medium were used to interpret data from experiments by Barton and Ford (1995) on bacterial migration in a column packed with sand.

Simulations in the bulk phase

Simulations of bacteria in the bulk phase (i.e., no obstacles present) were performed first. The first passage time (FPT) was defined as the time required by each bacterium starting from the center of the simulation box to reach a distance 0.2 cm away (Tobochnik, 1990). The number of FPT cells in the 0.2-cm-diameter sphere were recorded over time to monitor the migration of the bacterial population. The residence time ψ was defined as the average FPT for all 10,000 bacteria in any given simulation. Thus

$$\psi = \int_0^\infty P(t)t \, dt \qquad (10)$$

where $P(t)$ is the probability of a bacterium reaching the FPT in a time t.

As a reference case, the chemotactic sensitivity coefficient χ_0 was set equal to zero to simulate motility without chemotaxis. The results are presented in Fig. 3. Most of the bacteria reached the edge of the sphere after 400 s. On average, each bacterium had a FPT of 134 s in the sphere, as reported in Table 1. This average was determined by the asymptote when all of the cells had reached the sphere boundary (zero on the right-hand axis of Fig. 3). As the number of cells remaining within the sphere approached zero, one can see that the residence time approached its asymptotic value. This was always used as the criterion for determining the residence time.

Consider the situation with chemotactic bacteria only responding to positive gradients. For this case the rate of changing direction returns to the basal rate when bacteria move in the direction of a negative gradient (Eq. 9). Using

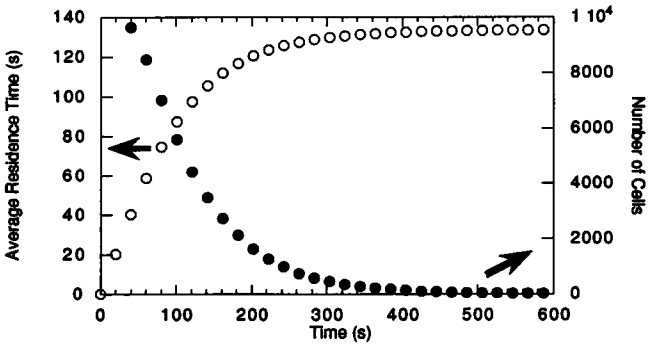

FIGURE 3 Average residence time for bacteria that have left the 2-cm-diameter sphere (*left axis*) and number of cells remaining in the 2-cm-diameter sphere (*right axis*), plotted as a function of simulation time. The results are for a reference case with no attractant.

Eq. 9 and the chemotactic sensitivity coefficient $\chi_0 = 2 \times 10^{-4}$ cm^2/s, the average residence time was 146 s (Table 1). This was greater than the situation without a chemotactic response, as expected. However, this increase was less than 10%. One might have expected the effect to be more dramatic. The small effect is due in part to the three-dimensional space available to the migrating bacteria. The chemotactic response is at its maximum for bacteria moving toward the source along the normal to the spherical surfaces of constant concentration surrounding the contaminant source. The response decreases to zero for bacteria moving tangential to these surfaces and is zero for all bacteria moving away from the source. Thus, for half of the possible directions of motion, there is no chemotactic effect on the bacterial swimming behavior.

We also simulated the case for which bacteria respond to negative gradients with the same mechanism that they use for positive gradients, but with the sensitivity reversed (Eq. 8). Using a chemotactic sensitivity coefficient of $\chi_0 = 2 \times 10^{-4}$ cm^2/s with Eq. 8, the average residence time approaches 167 s, representing a 25% increase over the reference case (Table 1). One reason for this larger increase is that bacteria described by Eq. 8 will spend a greater fraction of the time facing up the gradient (and extending their run lengths in that direction) than will bacteria described by Eq. 9.

TABLE 1 Residence times for bacterial simulations in a bulk liquid and a porous medium

Simulation conditions			
Medium simulated	Turning frequency calculation	χ_0 (cm^2/s)	Residence time (s)
Bulk liquid	Not applicable	0	134
Bulk liquid	Eq. 9	2×10^{-4}	146
Bulk liquid	Eq. 8	2×10^{-4}	167
Porous	Not applicable	0	4128
Porous	Eq. 8	2×10^{-4}	5939

Simulations in a porous medium

As a reference case, simulations were performed in a model porous medium with the chemotactic sensitivity coefficient set to zero. We used the same parameters and initial conditions that were used for the simulations in bulk liquid. Note that even without a chemotactic response, the overall residence time in the porous medium (4128 s) increased dramatically over that in the bulk (Table 1). This can be attributed to the path tortuosity caused by obstacles in the medium. For the obstacle size used (100 μm), the tortuosity parameter for the bacteria is $\sim$10 (Duffy et al., 1995).

We next simulated bacteria moving in a porous medium with the attractant concentration given by Eq. 5, as was done for the bulk simulations above. For $\chi_0 = 2 \times 10^{-4}$ cm^2/s, there is a more than 40% increase in the residence time over the situation where there was no attractant gradient (Table 2). This is considerably more than the increase of 25% for the bulk liquid medium (Table 1). However, this is likely due to using different diffusion coefficients D_{eff} for the porous medium versus $D_{3\text{CB}}$ for the bulk liquid (see Table 1). The important result is that even in the presence of a porous medium (which would tend to limit the extent to which bacteria could increase their mean run length), the chemotactic response provides a mechanism for increasing the residence time.

Application of simulation to interpret experimental observations

Barton and Ford (1995) investigated the effect of chemotaxis on bacterial migration in an experimental system with a porous medium. The porous medium consisted of commercially obtained silica sand sieved into a narrow distribution of particle sizes and placed in a column 2.5 cm in diameter and 6 cm in height. A dilute suspension of *P. putida* in an aqueous buffer saturated the bottom half of the column, and a solution of 5 mM 3CB saturated the top half of the column. An initial step change in the 3CB concentration gradually relaxed over the course of the experiment, and a well-defined macroscopic gradient was established by diffusion. Migration of bacteria into the upper half of the column was measured after 24 h by removing sections of the sand and performing plate counts to determine the bacterial population density profile within the sand column. Measurements of the overall migration of *P. putida* in experiments performed with and without a gradient of 3CB were indistinguishable (Barton and Ford, 1995). This result was unexpected.

TABLE 2 Residence times for bacterial simulations in bulk and porous media, according to Eq. 8, with the attractant gradient reversed

Simulation conditions		
Type of media simulated	χ_0 (cm^2/s)	Residence time (s)
Bulk liquid	2×10^{-4}	118
Bulk liquid	2×10^{-3}	62
Porous	2×10^{-4}	3421
Porous	2×10^{-3}	>100,000

P. putida have been shown to alter their run lengths by as much as fivefold in the presence of a temporal attractant gradient (Harwood et al., 1989). Within a porous medium such an increase in run length can result in an increase of as much as tenfold in the perceived path tortuosity of the medium (Duffy et al., 1995). Such an increase in path tortuosity might explain the similar penetration rates measured by Barton and Ford (1995) for the cases with and without the attractant gradient. To test this hypothesis, we performed simulations in porous media with and without an attractant, using Eq. 8, but with the direction of the gradient reversed. With the direction of the gradient reversed, the bacteria must migrate against the tortuosity imposed by the medium. If tortuosity alone can offset the chemotactic response, there should be no difference in residence time between the porous medium and the bulk liquid.

First, simulations for the bulk liquid were run with the attractant concentration given in Eq. 5, but with the sign of the gradient reversed. The chemotactic sensitivity coefficient measured by Barton and Ford (1995) ($\chi_0 = 2 \times 10^{-4}$ cm^2/s) and a value larger by an order of magnitude were used. The larger value produced a range of turn frequencies, for steep gradients with bacteria facing down a gradient, that were unrealistically high. Thus, for this case, an upper limit of 5 turns/s was set, corresponding to a number slightly higher than the maximum reported by Berg and Brown (1972) for *Escherichia coli*.

As expected, for both the low and high chemotactic sensitivity coefficients, the simulated bacteria left the sphere more quickly than in cases where the chemical gradient was directed inward (Table 2). Thus, in bulk solution, bacteria migrate through a greater volume when an attractant gradient is present than when it is not. The extent of penetration through the fluid is also dependent on the chemotactic sensitivity for the particular attractant. The higher χ_0 resulted in a lower overall residence time.

Within a porous medium and with an attractant gradient that is directed outward, the bacteria leave the sphere more quickly than for the reference case, in which bacteria do not respond to a chemical gradient (Table 2). These simulation results indicate that it is not necessarily true that path tortuosity will offset the effect of the chemoattractant. For this case an increase in path tortuosity alone would not account for the observations reported by Barton and Ford (1995).

If the chemotactic sensitivity coefficient χ_0 is increased by an order of magnitude to 2×10^{-3} cm^2/s, the residence time far exceeds that for the reference case (Table 2). For this case the average path length will be, on average, at least five times greater (Harwood et al., 1990). This means that on average, the path tortuosity will be an order of magnitude greater (Duffy et al., 1995), which may explain the longer residence time.

As a first approximation one might expect the effective overall residence time ψ_{eff} in the porous medium to be a product of the overall residence time ψ in the bulk and the path tortuosity T divided by the porosity ϕ of the medium, because either decreasing porosity or increasing the tortuosity will increase the residence time:

$$\psi_{eff} = \frac{T\psi}{\phi} \tag{11}$$

Using Eq. 11 and the results in Table 2, this product gives an estimate of 11.5 for the path tortuosity T in the porous medium without any attractant. This is in the range expected for an obstacle diameter of 100 μm (Duffy et al., 1995). For the case with an attractant in the reversed gradient and $\chi_0 = 2 \times 10^{-4}$ cm^2/s, the tortuosity $T = 11.1$. This is not significantly different and is, in fact, slightly less. For the simulated bacteria to have left the sphere more quickly, their run lengths must, on average, have been longer, and this should result in a higher tortuosity (Duffy et al., 1995). Because this is not the case, it supports the idea that the chemotactic response can overcome the effect of path tortuosity.

However, for the chemotactic sensitivity coefficient $\chi_0 = 2 \times 10^{-3}$ cm^2/s, the tortuosity is at least an order of magnitude greater, which corresponds to our earlier findings (Duffy et al., 1995). Thus path tortuosity can have a significant effect on the migratory behavior of bacteria moving in a chemotactic gradient. It is not beneficial for bacteria to increase their run lengths indiscriminately to migrate faster toward a source of food. If the increase in path length is too great, the path tortuosity due to the obstacles of the porous medium will work against their progress. These results suggest a physiological basis for the magnitude of the chemotactic sensitivity coefficient χ_0 measured by Barton and Ford (1995). For chemotactic sensitivity coefficients an order of magnitude greater, the increase in tortuosity becomes large and therefore is not beneficial for chemotaxis.

When Eq. 10 is used, the path tortuosity in the presence of an attractant is 12.3. Although not very different from the case with no attractant, it is larger. As *P. putida* are soil-inhabiting bacteria, it is likely that they have adapted to utilize the soil geometry and thus the tortuosity to their advantage. Thus they may use their chemotactic mechanism to increase their residence time in the vicinity of a nutrient source.

CONCLUSIONS

Experimental data have suggested that bacterial chemotaxis may not enhance bulk migration in porous media (Reynolds et al., 1989; Barton and Ford, 1995). The tortuosity of the porous medium experienced by a bacterium increases with an increase in the average bacterial run length (Duffy et al., 1995). Thus one hypothesis is that because chemotaxis increases the bacterial run length in the direction of an increasing gradient, the resulting increase in tortuosity may counteract the chemotactic response. However, the simulations reported here suggest that, depending on the conditions of the system, chemotaxis can still impact the migration of bacteria in porous media.

Another possible explanation for the result reported by Barton and Ford (1995) is that small-scale gradients reflecting heterogeneity within the porous matrix retard the bulk progress of the bacteria. The simulations reported here show that chemotaxis can nearly double the overall residence time of bacteria in a porous medium in the vicinity of an attractant source and thus support this explanation. For a bacterial species being used to remediate a contaminated site, an increase in residence time corresponds to an increase in contaminant consumption. Thus quantitative characterization of the chemotactic response of bacteria is important for assessing its impact on the efficiency of bioremediation.

When using bacteria for bioremediation, two factors, the speed with which they can get to a contaminant source and the amount of time they remain in its vicinity, are likely to be important. The simulations reported here show that these are dependent on the chemotactic sensitivity of the bacteria. In particular, it appears that the bacteria must maintain this sensitivity within certain bounds, depending on their needs. If it is more advantageous for a particular species of soil bacteria to be able to migrate to food sources quickly, then it must keep the chemotactic sensitivity parameter within a restricted range of values. However, if for another species of soil bacteria it is more important to remain for long periods in the vicinity of a food source, then larger values of the chemotactic sensitivity parameter will be beneficial.

The response of *P. putida* to positive and negative chemoattractant gradients is not fully understood. It is shown in this chapter that these responses can have an impact on the overall macroscopic behavior of the bacteria. Furthermore, as explained above, the degree of sensitivity to a chemoattractant and its gradient will affect their behavior near a food source. This behavior can have an impact on the distribution of bacteria in the natural environment as well as environmental technologies such as bioremediation.

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