Noise in Bacterial Chemotaxis: Sources, Analysis, and Control

PRATAP R. PATNAIK

Bacteria navigating through a chemotactic gradient in their natural habitats or in large bioreactors are under the influence of noise (or fluctuations) inside the cells, at the interfaces of the chemical ligands with the chemoreceptors, and in the external environment that contains the chemoattractant. These sources of noise interact with one another and may strongly affect the chemotactic motility of the cells. Although bacteria have evolved filtering mechanisms, mainly through feedback loops, for intracellular noise and receptor–ligand binding noise, external filters are required for environmental noise. With Escherichia coli *as a model system, these aspects are reviewed in terms of their effects on the chemosensory network, models for the filters, and optimization of chemotaxis under noise-affected conditions. It is suggested that stochastic resonance may be a key feature determining the design of an optimal filtering strategy that encompasses all sources of noise.*

Keywords: bacterial chemotaxis, intracellular noise, ligand binding noise, external fluctuations, filtering methods, stochastic resonance

Bacteria such as *Escherichia coli* and *Bacillus subtilis* sense and respond to the presence of chemical stimulants in their vicinity. Depending on the nature of the stimulant, the cells move either away from or toward the chemical. These guided movements, called *chemotaxis*, have important applications, such as in wound healing (Agyingi et al. 2010), the operations of microfluidic systems for biochemical reactions (Ahmed et al. 2010), and the degradation of undesirable chemicals in the environment (Singh and Olson 2008). These examples and many others indicate that most occurrences of chemotaxis involve chemical attractants (or *chemoattractants*), and therefore, the cells move toward the stimulus.

The mechanisms of chemotaxis in *E. coli* and *B. subtilis* are well established—more so in the case of *E. coli*. The chemosensory system of *E. coli* also contains fewer chemotaxis proteins than that of *B. subtilis*, and it has a simpler signaling network. Therefore, *E. coli* has served as a canonical model to enhance the understanding and regulation of the mechanisms and consequences of chemotaxis. However, the differences between *E. coli* and *B. subtilis* (Rao et al. 2004) and between prokaryotes and eukaryotes (Shibata and Ueda 2008) provide important information about how chemotaxis helps the evolution and survival of cells and how it may be used advantageously.

The chemotaxis machinery of *E. coli* comprises *chemoreceptors*, which detect and bind to chemoattractant molecules; a *signal transduction system*, which processes and transmits information from the receptors; and rotary motors, with flagella that rotate counterclockwise or clockwise in response to the outputs from the signal transduction network. Figure 1 is a schematic representation of the chemosensory system. The response of the system is encoded by six essential *che* genes—*cheA*, *cheB*, *cheR*, *cheW*, *cheY*, and *cheZ*—and five partially redundant chemoreceptor genes—*aer*, *tap*, *tar*, *trg*, and *tsr* (Zhulin 2001).

The CheA protein is a histidine protein kinase that catalyzes the transfer of phosphoryl groups from ATP (adenosine triphosphate) to one of its histidine imidazole side chains, from which it is transferred to an aspartyl side chain of the CheY protein (Baker et al. 2006). The phosphorylated CheY (CheY~P) then dissociates from CheA, diffuses through the cytoplasm and binds to the flagellar motor switch. The bound CheY~P functions as an allosteric regulator that governs the equilibrium between clockwise and counterclockwise rotations of the motor. Fluorescence resonance energy transfer data indicate that nonphosphorylated CheY does not bind to the motors (Sourjik and Berg 2002). This being the case, the phosphatase CheZ plays a key role in mediating the dephosphorylation of CheY~P.

The concentration of CheY~P is modulated through five chemoreceptors—Aer, Tap, Tar, Trg, and Tsr—which are present as large multimeric complexes with the CheA and CheW proteins (Francis et al. 2004). Briegel and colleagues (2012) and Liu and colleagues (2012) recently showed that these complexes comprise arrays of receptor dimers that are

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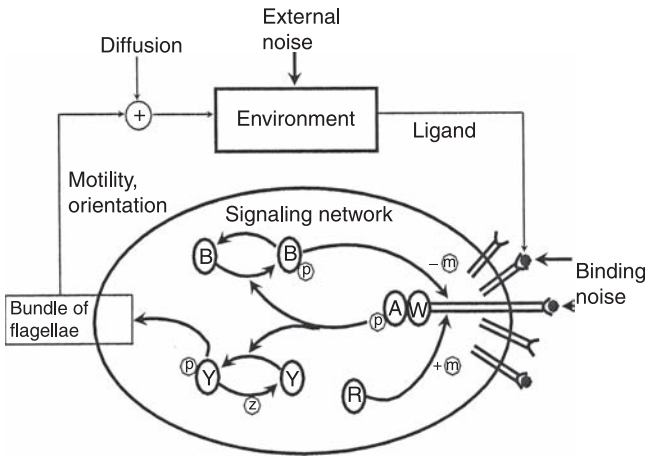

Figure 1. Schematic representation of the chemosensory network of* Escherichia coli. *In the figure, P is phosphate; m is methyl; A is a receptor protein kinase; W is a receptor complex component; and B and R are demethylating and methylating enzymes, respectively. Source: Adapted from Andrews and colleagues (2006).

hexagonally packed into trimers. As can be seen in figure 1, other chemotaxis proteins interact with the core assembly constituting the chemoreceptor, CheW, and CheA. The ligand binding domains of the receptors are outside the cytoplasmic membrane, whereas the rest of the chemosensory network is in the cytosol. The receptors therefore function as antennae to receive chemical signals and transmit them to the chemosensory network, which in turn generates signals through CheY~P that control the rotary motors.

The binding of a chemical ligand to its corresponding receptor complex causes reversible methylation of the receptor (Baker et al. 2006). Methylation is a critical component of chemosensing and chemotaxis in both *E. coli* and *B. subtilis* (Rao et al. 2004, Baker et al. 2006), because it contributes to their robust adaptation. This means that the chemotactic behavior of a population of cells reverts to its prestimulus state when the perturbing signal persists. Both robust adaptation and receptor–ligand binding have a direct relationship with binding noise, as is discussed below, and they have led to attempts to engineer microorganisms that are robust to a variety of disturbances, including those not yet experienced.

The final effect of the propagation of a chemoattractant signal through the chemosensory network is visible in the movements of the cells toward the attractant, executed with the help of the rotary motions of the cell's filamentous flagella. As a result of the alternate clockwise and counterclockwise rotations of the flagella, cellular motility comprises short alternate phases of straight-line motions (*runs*) and changes of direction (*tumbles*). In the absence of a stimulus, the runs and tumbles occur randomly, and a population of cells has no preferred direction of travel. The presence of a chemoattractant generates extended movement in the favorable direction, such that the population as whole moves toward the attractant.

The flagellar filaments are attached at their base to the rotary motors described above. The counterclockwise rotation of the flagella causes them to move in a coordinated bundle according to their left-handed helicity, and the result is a run (Wang et al. 2008). Clockwise rotations destabilize the bundle, causing the individual filaments to move independently; this creates random movements and results in a tumble. A typical run duration for *E. coli* is about 1 second, whereas a tumble takes just a tenth of this interval (Phillips et al. 1994, Berg 2000). Fluctuations in the chemoattractant concentration are therefore likely to interfere with the chemosensory response and thus to affect chemotaxis. Such fluctuations (or *noise*) occur both outside and inside the cells, as is described later, and therefore, it is important to understand their effects on cell motility in order to quantify and model bacterial chemotaxis under realistic conditions.

Sources of noise

Bacterial cells swimming though an environment containing a chemoattractant experience noise both from within themselves and from the environment. Much of the attention on the effects of noise has been concentrated on intracellular processes, presumably because they are present even in a noise-free environment and because they often have a more significant effect on cell motility than would noise from outside.

Inside the cells, noise arises mainly because molecules such as DNA, messenger RNA, and synthesized proteins are usually present at low (numerical or gravimetric) concentrations. Therefore, intracellular processes inevitably involve probabilistic collisions between these molecules, and these events are statistically interdependent (Paulsson 2005). This would suggest an inverse relation between system size and the magnitude of systemic noise (van Kampen 1992). In general, for a molecular abundance of N, the scale of the noise varies as $\sqrt{N}$ (Berg and Purcell 1977). Because the Che proteins of *E. coli* are expressed by specific genes, noise in gene expression has a significant influence on the chemotactic motility of the cells.

Noise associated with gene expression is often classified as intrinsic or extrinsic. *Intrinsic noise* itself may be gene intrinsic, network intrinsic, or cell intrinsic, as is depicted in figure 2. *Gene-intrinsic noise* refers to molecular-level noise in the reaction steps associated with gene expression. *Network-intrinsic noise* is generated by fluctuations in signal transduction. *Cell-intrinsic noise* arises from both of these sources and from fluctuations in cell-specific factors, such as metabolite concentrations, cell size, and cell age (Kaern et al. 2005).

Contrasted with intrinsic noise sources, *extrinsic noise* is associated with fluctuations and population variability. This should suggest that intrinsic noise creates differences between two reporters within a given cell, whereas extrinsic noise creates differences between two or more cells but affects both reporters equally inside each cell (Raser and O'Shea 2005). Elowitz and colleagues (2002) developed an elegant two-reporter assay to distinguish between the two types of noise, and Swain and colleagues (2002) proposed

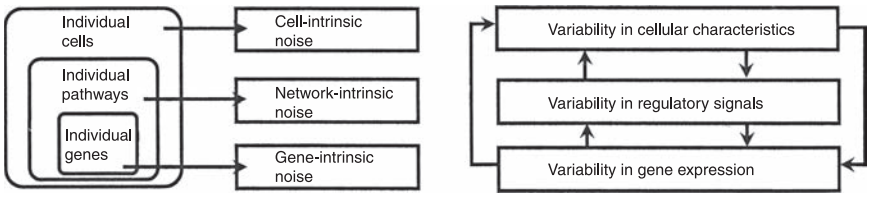

Figure 2. Sources of noise inside a cell and their interactions among themselves and with the external environment. Source: Adapted from Kaern and colleagues (2005) with permission from MacMillan Publishers Ltd. © 2005.

quantitative expressions for their contributions to the overall genetic noise.

Kaern and colleagues (2005) and Paulsson (2005) emphasized that the distinction between intrinsic and extrinsic noise may be context dependent. This perspective is also evident from two recent analyses of bacterial chemotaxis. Shibata and Fujimoto (2005) and Shibata and Ueda (2008) both explained that intrinsic noise is associated with (intracellular) reactions per se, whereas extrinsic noise represents how the noise of the inputs is propagated into noise in the outputs. The latter definition of *extrinsic noise* conforms with the concepts introduced by other theoretical (Raser and O'Shea 2005) and experimental (Elowitz et al. 2002) investigations, because propagation of internally driven fluctuations from one cell to another are a prime cause of differences between cells in a population.

Not all sources of noise reside inside the cells. Recall that receptors embedded in the cytoplasm serve as antennae that detect and bind to appropriate chemoattractant molecules (Baker et al. 2006). The binding process is also stochastic in nature, and bacterial cells have evolved methods to filter the associated noise. Modeling of this filter is described later. The aim of filtering is to detect the true signal (i.e., ligand concentration) from a noise-distorted signal. Efficient identification of the true signal requires optimal filtering, as is explained below. The length of time over which the signal is averaged in order to enable a cell to decide whether to tumble or run is inversely proportional to the filter cutoff frequency and determines the adaptation time (Andrews et al. 2006). If this time is too short, the noise is not filtered out sufficiently; if the time is too long, the bacteria cannot detect real changes in the chemoattractant gradient (Berg and Purcell 1977, Strong et al. 1998). An optimal filter provides the best balance between these two limits. Previous studies (Kaern et al. 2005, Patnaik 2007, Hornung and Barkai 2008) have indicated that a noise filter at its optimum settings allows noise of just that bandwidth that resonates with the relevant biochemical processes, thereby promoting the final outcome.

In many real situations, the chemoattractant in the extracellular environment is not static; it is also subject to fluctuations (Patnaik 2006a, 2007, Xu and Tao 2006). This is true not just of chemotaxis but of other microbial processes, as well. There is a remarkable similarity between control strategies for environmental noise and those employed by the cells for gene-intrinsic noise. For example, in continuous cultures of *Saccharomyces cerevisiae*, optimal filtering of feed stream noise with partial recycling of the filtered outputs helps to restore stable performance from chaotic behavior (Patnaik 2006b). Similarly, the galactose-use network in *S. cerevisiae* has two positive and one negative feedback loops. The positive feedback loops contribute to the establishment of two stable expression states, whereas the negative feedback controls the switching of the cells between these states so that the more favorable state is always chosen in a changing environment (Kaern et al. 2005, Hornung and Barkai 2008). In *E. coli*, both negative (Hooshangi and Weiss 2006) and positive (Hornung and Barkai 2008) feedback loops contribute to noise abatement while maintaining high sensitivity to genuine changes in the environment, without the corruption of noise. For example, changes may occur in the substration concentration distribution or in the viscosity as a result of bacterial metabolism, and the feedback loops help the cells to detect these changes by reducing the influence of noise.

It is impractical to remove noise entirely from the extracellular environment. Indeed, as is discussed later, this is not even desirable; controlled noise may favor chemotaxis more than the complete absence of noise. Because ligands with fluctuating concentrations participate in a noise-affected receptor-binding process, thereby sending noisy signals through a chemosensory network with its own noise sources, it is important, even though it is difficult, to model and control these interacting noise sources such that chemotactic performance is maximized. Some methods to do this are discussed below.

Analysis and control of noise

Since the chemosensory processes leading eventually to cell motility involve interactions between receptor molecules, Che proteins expressed by corresponding genes, and signal transfer to the rotary motor proteins FliM and FliY, it is expected that noise at the genetic and molecular level will be reflected in fluctuations in signal transduction and the resulting cellular movements (Shibata and Fujimoto 2005, Miyanga et al. 2007, Shibata and Ueda 2008). Since the maintenance of proper chemotaxis in real (noise-affected) environments is of practical importance (Singh and Olson 2008, Agyingi et al. 2010, Ahmed et al. 2010), many researchers have tried to describe, analyze, and regulate the effects of noise.

Most researchers have addressed noise inside bacterial cells and noise present in the binding of ligand molecules to their chemoreceptors. In fewer studies has environmental noise been considered; however, its significance is now being recognized, because fluctuations in ambient chemoattractant

concentrations inevitably affect binding kinetics (Sourjik and Berg 2002, Patnaik 2007). Intracellular noise in prokaryotic cells may be present at the translational level, at the transcription level, or at both levels. However, Ozbudak and colleagues' (2002) experiments showed that gene expression is influenced largely by translational noise. They incorporated the green fluorescence protein (GFP) under the control of an inducible promoter into the chromosome of *B. subtilis* and determined by flow cytometry the variation in expression of GFP from cell to cell. Their results revealed not only that translational noise was dominant but also that the spread in expression levels of GFP followed a greater-than-Poisson variation, which implies that the ratio of the variance to the mean was greater than one. This inference has been corroborated by later studies (Korobkova et al. 2004, Shibata and Fujimoto 2005, Shibata and Ueda 2008).

Whereas intracellular noise is present even without a chemical stimulus, the presence of a chemoattractant introduces an additional source of noise through the binding of the chemical ligand to the chemoreceptors. This noise, too, has its origin in the small number of molecules participating in the binding process. This was recognized more than 30 years ago by Berg and Purcell (1977), who postulated that a sensor of characteristic length a could count an average of $\bar{N} \approx \bar{c}a^3$ molecules when the mean concentration is $\bar{c}$. Each such measurement has a noise component $\Delta\bar{N} \approx \sqrt{\bar{N}}$. This noise inevitably affects the accuracy of measurement; for chemotaxis, this limits the sensitivity of detection of the "true" ligand concentration from a noisy signal by the receptor molecules.

Because genetic noise and receptor–ligand binding noise are germane to the cellular machinery, chemotactic bacteria have evolved methods to control these sources of noise. A common method of noise reduction in gene networks is the use of negative feedback. Whereas this idea is well established in engineering, Becskei and Serrano's (2000) study demonstrated it for bacterial cells. As for the greater-than-Poisson variation of translational noise mentioned above, the prevalence and stabilizing effect of negative feedback has been confirmed by other investigators (Simpson et al. 2003, Hooshangi and Weiss 2006).

Nevertheless, negative feedback can also have a destabilizing effect and can thus give rise to either dampened or sustained oscillations if it includes a time delay. This possibility has led to investigations into and arguments for positive feedback. One argument is that positive feedback promotes phenotypic diversity. The presence of phenotypically different cells in a population confers the benefit that, in a changing environment, there will usually be some phenotypes that will thrive, whereas others may not survive (Booth 2002). A corollary inference is that a heterogeneous population of cells may switch continuously from one preferred phenotype to another to maximize its survival in a fluctuating environment. Although it is beneficial to the cells, rapid switching is also accompanied by chatter (i.e., rapid fluctuations in the rotation of the flagellar motors), especially in a noisy environment. The robustness of chemotaxis to temporal changes was initially attributed to the flagellar motor's possessing a mechanism to prevent chatter (Morton-Firth and Bray 1998). However, the lack of correlation between switching behavior and the fluctuations in CheY concentration negated this theory, and it was shown later (Bren and Eisenbach 2001) that the flagellar motor switch may possess hysteresis, which is known to reduce chatter.

In spite of the destabilizing tendency of stochastic fluctuations inside the cells, it is remarkable that even "simple" unicellular organisms such as *E. coli* possess such robust chemotactic pathways. In fact, *E. coli* has the smallest sufficiently robust chemotactic signaling network among all bacteria; the topological organization of this network (Rao et al. 2004, Kollman et al. 2005, Steuer et al. 2011) illustrates the intricate design principles that nature has employed to enable the cells to function effectively even under noisy conditions. This robustness is not restricted to intrinsic fluctuations of intracellular components but extends to fluctuations in external conditions, such as in chemoattractant concentration (Patnaik 2007) and temperature (Oleksink et al. 2011). It has been shown that the effect of temperature on adaptation kinetics is counterbalanced in a preprogrammed way, such that optimal performance is achieved at the growth temperature.

Positive feedback also enhances the buffering of noise while maintaining high sensitivity, thereby reducing propagation of the noise downstream of its source (Hornung and Barkai 2008, Shibata and Ueda 2008). This is an important consideration, because biomolecular events operate at thermal noise energy levels, at which thermal fluctuations cause spontaneous fluctuations in the rate constants in the chemosensory network (Paulsson 2004, Bialek and Setayeshgar 2005). Negative feedback amplifies these fluctuations (Hornung and Barkai 2008), thereby creating large behavioral variability in a population (Emonet and Cluzel 2008). Korobkova and colleagues (2004) reached a similar conclusion from a different perspective. By analyzing the power spectra of individual cells of wild-type *E. coli* and their ensemble averages, they showed that temporal variations of the bias obtained from a population could be produced by uncorrelated, exponentially distributed intervals of clockwise and counterclockwise rotations of the flagellar motors.

The exponential distribution of the clockwise and counterclockwise rotation intervals has, however, been questioned by some investigators. Korobkova and colleagues (2006) stated that the switchings between clockwise and counterclockwise rotations are not rigidly separable but are stochastic in nature and follow a gamma distribution. More recently, Matthäus and colleagues (2009) also negated the exponential distribution in the presence of internal fluctuations but proposed a power-law distribution, which would suggest that bacteria might perform superdiffusive Levy-walk motion. They further argued that the cells perform Levy walks in response to fluctuations in the level of CheR

even when no chemoattractant is present, which indicates that intracellular noise confers a survival advantage when there is a shortage of nutrients.

Modeling noise-affected chemotaxis

The many experimental observations (Sako et al. 2000, Elowitz et al. 2002, Ozbudak et al. 2002, Sourjik and Berg 2002, Korobkova et al. 2004, Hooshangi and Weiss 2006) of the effects of noise (from different sources) have understandably led to attempts to model and quantify these effects. Shibata and Fujimoto (2005) addressed the basic issue of quantifying noise and its effects. They focused on intracellular noise and, as Swain and colleagues (2002) did, differentiated between intrinsic and extrinsic noise. Their interest was in relating noise to signal amplification in biochemical reaction networks. One of the reactions studied was the Monod–Wyman–Changeux model of chemotaxis (Monod et al. 1965), and the Michaelis–Menten equation was used as the reference point for the reason explained below.

Shibata and Fujimoto (2005) considered an arbitrary signal of intensity S and its corresponding response X. For chemotaxis, S is likely to be the concentration of a chemoattractant or of a cellular compound, such as the CheR protein, that is sensitive to the chemoattractant, and X is the movement of the cell. They then defined the amplification of the signal as

$$g = \frac{d\log X}{d\log S}. \tag{1}$$

Here, g is the gain (see box 1; equation 1 is repeated as equation B2) of the chemosensory system. Because the maximum possible gain, g_{max}, for such a reaction is unity, Shibata and Fujimoto (2005) used this as a reference value and defined an ultrasensitive reaction as one in which $g_{max} > 1$. On the basis of equation 2 below, they derived separate gain expressions for intrinsic noise and extrinsic noise (box 1). When X is the number of proteins resulting from a gene expression, $\theta = 1/(b + 1)$, where b is the translation efficiency.

Equation B6 in box 1 tells us that the gain, g, depends quadratically on the signal intensity, S, and the total variance, σ_{total}, of the noise. This is favorable to robustness, because the intensity of the noise is usually much smaller than that of the sensory signal triggered by the chemoattractant (Kollmann et al. 2005, Raser and O'Shea 2005, Andrews et al. 2006), and therefore, a quadratic dependence further attenuates the effect of noise on cell motility.

Many biochemical reactions may be modeled by the Michaelis–Menten rate equation, which has the form

$$r = \frac{kS}{K_M + S}, \tag{2}$$

where r is the rate of reaction, S is the signal concentration, k is a rate constant, and K_M is an equilibrium constant

Previous studies (Alon et al. 1998, Cluzel et al. 2000, Sourjik and Berg 2004) have shown that the chemosensory signal tranduction system (figure 1) of *E. coli* and other bacteria are ultrasensitive, and therefore, they have underlying stochasticity. It is therefore important to determine the relative intensities of intrinsic and extrinsic noise. This may be done through spectral analysis. Korobkova and colleagues' (2004) analysis showed that the intensity of the power spectrum varied inversely with the concentration of CheR in the low-frequency

Box 1. The model of Shibata and Fujimoto (2005) for the effect of noise.

Let a change ΔS in a signal S produce a change ΔX in X from its stationary value $\bar{X}$. Then the amplification is defined as the fractional change in $\bar{X}$ generated by the fractional change in S. This is called the *gain*:

$$g = \frac{\Delta X/\bar{X}}{\Delta S/S} \tag{B1}$$

When ΔS represents a noise in S, both ΔS and ΔX are usually small enough to replace equation (B1) by the following differential expression:

$$g = \frac{d\log X}{d\log S} \tag{B2}$$

Shibata and Fujimoto (2005) showed that

$$g \propto \sigma_{in}^2, \tag{B3}$$

where σ_{in}^2 is the variance of the intrinsic noise. Equation B3 leads to the gain–noise relation

$$g = \theta\frac{\sigma_{in}^2}{\bar{X}}, \tag{B4}$$

where θ is a reaction-dependent parameter.

Similar to equation B4, a gain–noise relation was also derived for extrinsic noise:

$$\frac{\sigma_{ex}}{\bar{X}} = g\frac{\sigma_s}{S}\sqrt{\frac{\tau_s}{\tau+\tau_s}}. \tag{B5}$$

Here, σ_{ex} and σ_s are square roots of the variances of the extrinsic noise and the input signal S, τ_s is the time constant of the noise in S, and τ the time constant of the signal transduction reaction. Summing up equations B4 and B5, one obtains the relative noise intensity of the total noise, $\sigma_{total}/\bar{X}$, in the stationary state, which is called the *gain–fluctuation relation*:

$$\frac{\sigma_{total}^2}{\bar{X}^2} = \frac{g}{\theta\bar{X}} + \frac{g^2\sigma_s^2}{S^2}\left(\frac{\tau_s}{\tau+\tau_s}\right) \tag{B6}$$

region and was nearly constant at high frequencies. From Shibata and Fujimoto's (2005) analysis revealing that extrinsic noise dominates at low frequencies, whereas intrinsic noise remains unchanged, Korobkova and colleagues' (2004) observations lead to the inference that, with increasing concentration of CheR, extrinsic noise gets suppressed, and there is reduced behavioral variation in a population of cells. Interpreted in the context of phenotypic diversity imparting survival advantages to the cells (Booth 2002, Kaern et al. 2005), the foregoing argument leads to the important—and possibly paradoxical—inference that some extrinsic noise is required to promote population variability.

Shibata and Ueda (2008) used Shibata and Fujimoto's (2005) theory to explain the effect of CheR on flagellar motor rotation. The main difference between this explanation and the one proposed earlier by Bren and Eisenbach (2001) seems to be that the latter study was for a deterministic system, whereas Shibata and Ueda (2008) employed stochasticity concepts. Both explanations, however, converge to the conclusion that variations in CheR concentration regulate the motor-switch protein FliM and thereby control motor rotation. The role of (extrinsic) noise is indirectly through its effect on CheR; this situation is plausible because chemoreceptors and flagellar motors are known to exhibit high gain responses (Cluzel et al. 2000, Sourjik and Berg 2002).

A question that has generated considerable thought is how a bacterial population possesses robust adaptation in spite of individual cells being ultrasensitive. From their analysis of the power spectra of individual *E. coli* cells and of the ensemble, Korobkova and colleagues (2004), using the thesis of Viswanathan and colleagues (1999), surmised that a power-law distribution of run lengths (under the influence of extrinsic noise) may provide the cells with an optimal strategy to adapt to an unfavorable and variable environment. This possibility also has the support of Matthäus and colleagues (2009), who attributed the robustness to the superdiffusive Levy motions that are characterized by a power-law distribution. Approaching the problem from a different perspective, Emonet and Cluzel (2008) used the fluctuation–dissipation theorem (van Kampen 1992) to relate cellular response to the impact of noise. They showed that the behavioral variability of an ensemble of cells was controlled by the slope of the histidine kinase activation curve.

It is remarkable that a bacterial population exhibits robust perfect adaptation in spite of individual cells' being ultrasensitive and under the influence of a number of sources of noise. However, this robustness is tempered by the observation (Carson and Doyle 2002, Kitano 2004) that complex systems, such as chemosensory networks, are also fragile, which implies that an unanticipated disturbance may drive the system into unexpected—and possibly detrimental—modes of functioning. For example, uncontrolled stochasticity generates a large number of cells that respond to high stress (and are therefore robust) but even more cells that cannot respond to weak stress and that may therefore be injured easily (Shahrezaei and Swain 2008).

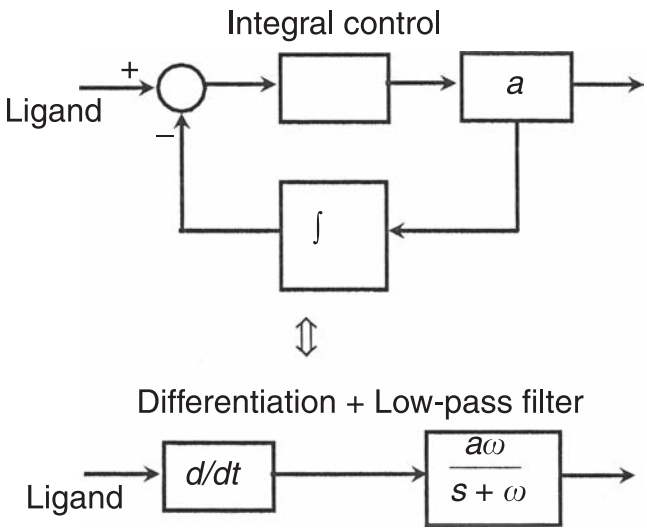

Figure 3. Integral feedback control scheme for the filtering of chemoreceptor–ligand binding noise in* Escherichia coli*, where* ω *is the cut-off frequency of the noise filter,* t *is the time,* s *is the Laplace domain equivalent of* t*. Source: Adapted from Andrews and colleagues (2006).

Given that molecular noise, both inside and outside the cells, is an ever-present feature, how do bacterial cells cope with this? The roles of positive and negative feedback in attenuating intracellular noise while maintaining sensitivity have been discussed above. Cells also have internal mechanisms to filter out receptor–ligand binding noise. This was analyzed by Andrews and colleagues (2006) on the basis of a model proposed earlier (Yi et al. 2000). Building on the concept that integral feedback loops provide stable behavior with zero offset in many engineering systems, Yi and colleagues (2000) proposed a similar mechanism to explain the perfect adaptation (equivalent to zero steady-state error) of bacterial cells. Their model (figure 3) is functionally equivalent to a differentiator in series with a first-order low-pass filter; the equivalence is useful in representing an analog simulation of bacterial chemotaxis with noise filtering. A low-pass filter has general validity for different kinds of time-varying chemical stimuli; demonstrating this, Tu and colleagues (2008) provided the important clarification that the signal transduction pathway is a low-pass filter for the time derivative of the signal and not of the signal itself, as was thought earlier (Block et al. 1983).

Andrews and colleagues (2006) incorporated Yi and colleagues' (2000) model into a simulation model (figure 4) to determine, for *E. coli* cells, an optimal cutoff frequency that would separate the low-frequency signal from the high-frequency noise of ligand binding. This frequency depended inversely on the level of binding noise and directly on the level of rotation diffusion of the chemoattractant molecules. They also noted that, because the variance of measurement uncertainty due to fluctuations in the diffusion

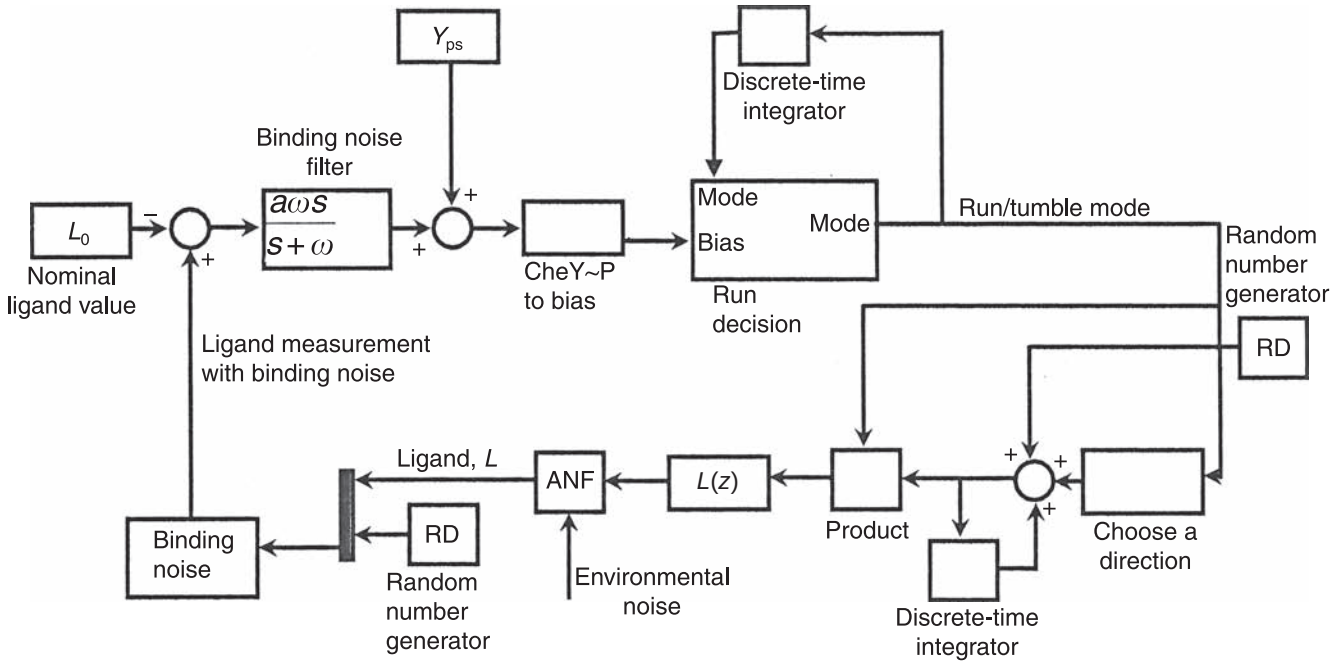

Figure 4. Simulation flow diagram of the* Escherichia coli *chemotaxis system with filtering mechanisms for ligand binding and environmental noise, where ω is the cut-off frequency of the noise filter, s is the Laplace domain equivalent of time. Abbreviations: ANF, autoassociative neural filter; CheY~P, phosphorylated CheY protein. Source: Adapted from Andrews and colleages (2006).

measurements of chemoattractant ligands (Berg and Purcell 1977, Bialek and Setyeshgar 2005) is four orders of magnitude smaller than that due to fluctuations in ligand binding dynamics (Orrell and Bolouri 2004), the former may often be ignored. However, there are two caveats to this inference. First is that "ligand diffusion provides a (perfect sensor) limit to the degree to which the optimum filter cut-off frequency can increase in response to decreasing binding-dynamics noise" (Andrews et al. 2006, p. 1413). Second, for chemotaxis through a gradient in ligand concentration, sensitivity to fluctuations may vary along the gradient and may depend on the relative diffusion coefficients of the bacteria, the chemoattractant, and the primary nutrient (Patnaik 2008). Then the role of diffusion may be significant in some segment or segments of the gradient and negligible elsewhere.

Bacteria in their natural habitats and when cultivated in large bioreactors are also subject to noise from their environment. Since the chemoattractant is present in this environment, external (or environmental) noise inevitably has an impact on ligand binding dynamics. However, recognition and quantitative analyses of this feature are just emerging (Patnaik 2006a, 2007, Xu and Tao 2006, Tu et al. 2008). Since cells do not have internal mechanisms for dealing with environmental noise, external filters have been provided. Previous studies of noise-affected microbial cultures (Tian and Burrage 2003, Patnaik 2006b) have shown that neural networks function effectively as filters for time-varying noise. On this basis, in Patnaik (2012), I incorporated a neural filter into the simulation model of Andrews and colleagues (2006) (see figure 4) and studied the effectiveness of different neural configurations in filtering Gaussian noise in the environment. All configurations improved the chemotactic performance of *E. coli*, with an autoassociative filter being the best. Seen in the background of previous studies, the improvement of chemotaxis through the optimal (but not complete) filtering of noise seems to be a consequence of stochastic resonance between the external filtered noise and intracellular noise. Broadly, stochastic resonance may be defined as a phenomenon in which the output variable of a nonlinear process is amplified and strengthened by the presence of controlled noise (McDonnell and Abbott 2009). In this definition, the process has to be nonlinear, and the noise should be controlled. Both requirements are satisfied in the noise-filtering analyses of chemotaxis reported here.

Stochastic resonance has been advanced as an explanation for diverse phenomena that include electronic circuits, finance models, and biological processes. Many biological processes have been discussed in two recent reviews (Hänggi 2002, McDonnell and Abbott 2009), and they cover widely different systems, such as physiological neural populations, human visual perception and blood pressure regulatory systems, the behavior of feeding paddle fish, and—most relevant to the present work—biochemical reactions. Although the driving forces and the output signals may differ from one example to another, noise is a common factor that enhances

system performance in all cases. Therefore, stochastic resonance will be a likely reason for the improvement of chemotaxis through optimal filtering of feed stream noise.

Conclusions

Under realistic conditions, bacteria navigating chemical gradients are subject to noise inside the cells, at the binding domains for the chemical ligands, and in the environment that contains the chemoattractant. Through evolution, cells have developed their own mechanisms to filter out intracellular noise and the noise associated with the binding of chemical ligands to their chemoreceptors.

Stochastic fluctuations in the level of gene expression, the rates of metabolic reactions, and the concentrations of the participating reactants and products give rise to intrinsic noise (Kaern et al. 2005, Raser and O'Shea 2005). A primary reason for stochasticity is the small number of the participating entities, which makes intracellular processes probabilistic in nature. A fundamental method by which this noise is attenuated is feedback. Both negative and positive feedback are present, each has its advantages and disadvantages, and cells may be engineered to promote either method of feedback.

Like intracellular noise, the modulation of ligand–receptor binding noise may also be represented by a low-pass filter with integral feedback. The purpose of the integral function is to prevent undesirable oscillations in the filtered signal and to ensure perfect adaptation. It has been shown for *E. coli* that chemotaxis is most favored by an optimum cutoff frequency of the filtered noise (Andrews et al. 2006). A similar optimum was recently shown to exist for environmental noise (Patnaik 2012), for which an autoassociative neural filter was superior to other configurations.

The results from different methods of filtering the noise at different levels reveal the interesting inference that optimum levels of noise generate more efficient chemotaxis than does the complete removal of noise. Maintaining a certain degree of stochasticity has benefits such as imparting a survival advantage to the cells (through increased phenotypic diversity) and robustness to parameter changes (Booth 2002). High stochasticity, however, may undermine chemotactic performance by upsetting the relative rates of metabolic reactions and flux balances. Optimum filtering of intracellular, ligand binding, and environmental noise has been hypothesized to create a stochastic resonance that favors chemotaxis (Orrell and Bolouri 2004, Patnaik 2006a, 2012). Realization of such an integrated optimum filtering system is a challenging but rewarding task.

Traditionally, the improvement of system performance through the inflow of noise is measured by the signal-to-noise ratio (SNR). However, as McDonnell and Abbott (2009) pointed out, the SNR may not be an appropriate metric for biological systems. They argued that if the aim is to evaluate the effect of noise on one or more biological functions, it is more meaningful to measure variations in the function or functions rather than the SNR. The results reported by Andrews and colleagues (2006) for bacterial chemotaxis illustrate precisely this point.

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Pratap R. Patnaik (pratap.r.patnaik@gmail.com) is a professor of chemical engineering and is the dean of research at the C. V. Raman College of Engineering, in Bhubaneswar, India.