

Supporting Information

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SI Text

The Fuel Needed to Drive the *Escherichia coli* Chemotaxis Network Is Comparable to That to Make New Components After Cell Division

Below, we address the question: How does the ATP cost of driving the *E. coli* chemotaxis sensing system compare with the energetic cost of making the components of this system?

Estimating Energetic Cost of Protein Production. The number of ATP molecules required to make a protein is approximately five per amino acid [BioNumbers ID 106158 (1)]. Table S1 reports the copy number ranges for key chemotaxis proteins and their molecular weight. These proteins need to be synthesized during the cell cycle to balance the loss upon cell division. Assembling these numbers, about $2\text{--}8 \times 10^7$ ATP are required to produce chemotaxis proteins over the cell cycle.

Estimating Energetic Cost of Driving the Chemotaxis Network. The chemotaxis network of *E. coli* is modeled as follows (2):



where A and Ap denote the unphosphorylated and phosphorylated forms of CheA, and Y and Yp the unphosphorylated and phosphorylated forms of CheY.

The reactions imply that in steady state:

$$k_A([A_T] - [Ap]) = k_t[Ap]([Y_T] - [Yp]) = k_r[Yp] = \dot{n}. \quad [\text{S4}]$$

The second and third terms in this equation (and thus the first) are expressions for the flux of CheY, \dot{n} , in steady state. The lifetime of CheYp is $\tau_l \approx 100$ ms (2), which is the inverse of k_r in our model: $k_r \approx 10/s$. A similar estimate for k_t can be obtained by noting that the deactivation is mediated by CheZ and calculating k_r from Michaelis–Menten kinetics as $k_r = (k_{\text{cat}}[Z][Yp]) / (K_m + [Z])$ using parameters in ref. 2. The total turnover rate \dot{n} is thus 10/s times the number of CheYp molecules. About a sixth of the 1,000–8,000 CheY molecules are phosphorylated in the steady state (2). Then the number of CheYp is roughly 200–1,500. The total turnover rate \dot{n} is thus 2,000–15,000 molecules per second, which is also the ATP turnover rate, because the phosphorylation reaction is essentially irreversible. If the cell cycle time is about an hour (3,600 s), the turnover of ATP is about $1\text{--}5 \times 10^7$ ATP per cell cycle.

From these calculations, we see that the ATP cost of driving the chemotaxis sensing machinery ($\approx 1\text{--}5 \times 10^7$ ATP per cell cycle) is comparable to the energetic cost of making the components of the sensing machinery ($\approx 2\text{--}8 \times 10^7$). This comparison depends critically on the growth rate of the cell and protein expression levels, which can vary in different media. Furthermore, it does not account for the energy to drive or build other components of the chemotaxis pathway, such as the flagella or the adaptation machinery. However, it provides an order-of-magnitude comparison, which suggests that the energy to drive a signaling pathway can

be comparable to the cost of making the components of that pathway.

Calculating the Sensing Error for a Biochemical Network

From Eq. 1 in the main text, the sensing error for a biochemical network depends on the gain and the variance of the readout. We have calculated the gains using a mean-field approximation for the steady-state level of the readout. We have calculated all variances using a linear-noise approximation (3). For nonlinear networks, the quality of the approximation improves with system size; it can already be quite good for systems with only 10 copies of each molecule (4).

The linear-noise approximation gives the covariance matrix Σ for stationary fluctuations in species' levels as the solution to the Lyapunov equation:

$$A\Sigma + \Sigma A^T + B = 0, \quad [\text{S5}]$$

where $A = S^T \nabla \nu$ and $B = S^T \text{Diag}(\nu) S$ in terms of the stoichiometric matrix S and the reaction propensity vector ν . The stoichiometric matrix describes how many molecules of each species are consumed or produced in each reaction, and the propensity vector describes the propensity (rate) of each reaction. For a network out of steady state (as in *Base Model: Taking Samples via Readout Activation* below), a nonstationary version must be used (3).

Langevin Approximation to the Dynamics of a Biochemical Network

The Langevin approximation to the dynamics of a biochemical network draws on the same framework as the linear-noise approximation (3). It expresses the fluctuations in species copy numbers N as follows:

$$\frac{dN}{dt} = AN + \eta(t), \quad [\text{S6}]$$

where N is a vector containing the copy numbers of all species and $\eta(t)$ are Gaussian noises, uncorrelated in time, with covariance B . A and B are the matrices defined in *Calculating the Sensing Error for a Biochemical Network*. The equation can be solved [e.g., by integrating factors (3)], yielding the result in the main text, Eq. 3, for the biochemical network considered there.

Derivation of Eqs. 2 and 3 of the Main Text

The principle is that the cell infers the concentration c from the instantaneous output x^* , by inverting the input–output relation $\bar{x}^*(c)$. Linearizing the input–output relation, and using error propagation, the fractional error of the concentration estimate is given by the following (5–7):

$$\left(\frac{\delta c}{c}\right)^2 = \frac{1}{c^2} \frac{\sigma_{x^*}^2}{\left(\frac{dx^*}{dc}\right)^2}. \quad [\text{S7}]$$

We can compute the variance of the output x^* , $\sigma_{x^*}^2$, using the linear-noise approximation to the master equation (see above, *Calculating the Sensing Error for a Biochemical Network*). Using the above expression, this yields the following:

$$\begin{aligned} \left(\frac{\delta c}{c}\right)^2 &= \left(\left((k_1 + k_2)^4 \left(k_{-r} + k_r + \frac{k_1(k_f + k_{-f})R_T}{k_1 + k_2} \right) \right. \right. \\ &\times \left(\left(k_{-r} + \frac{k_1 k_f R_T}{k_1 + k_2} \right) \left(k_r + \frac{k_1 k_{-f} R_T}{k_1 + k_2} \right) \left(k_{-r} + k_r + \frac{k_1(k_f + k_{-f})R_T}{k_1 + k_2} \right) \right. \\ &\left. \left. + \frac{k_1 k_2 (k_{-f} k_{-r} - k_f k_r)^2 R_T X_T}{(k_1 + k_2) \left((k_1 + k_2) (k_1 + k_2 + k_{-r} + k_r) + k_1 (k_f + k_{-f}) R_T \right) \right) \right) / \\ &\left(k_1^2 k_2^2 (k_{-f} k_{-r} - k_f k_r)^2 R_T^2 X_T \right). \end{aligned} \quad [S8]$$

Here, $k_1 = \tilde{k}_1 L$ (with L the ligand concentration) and k_2 are the rate constants for ligand binding and unbinding from the receptor, respectively, k_f is the rate of receptor-mediated phosphorylation, k_{-f} is the microscopically reverse reaction, k_r is the rate of dephosphorylation, k_{-r} is the microscopically reverse reaction, and R_T and X_T are the number of receptor and readout molecules, respectively. Clearly, the above expression is not very illuminating.

Eqs. 2 and 3 of the main text are much more illuminating. They describe the sensing error not in terms of the rate constants, but in terms of the collective variables $p, \tau_c, \tau_r, \dot{n}, \Delta\mu_1, \Delta\mu_2, \Delta$ that describe the resource limitations of the cell. The inspiration for the form of these expressions came from an analysis of a simpler network. We describe this analysis in the subsection below. Here, we first repeat Eqs. 2 and 3 for completeness, and then describe how it can be verified that they indeed correspond to Eq. S8.

Eqs. 2 and 3 of the main text are as follows:

$$\left(\frac{\delta c}{c}\right)^2 = \frac{1}{p(1-p)} \frac{1}{\bar{N}_I} + \frac{1}{(1-p)^2} \frac{1}{\bar{N}} \quad [S9]$$

and

$$\bar{N}_I = \frac{1}{\underbrace{(1 + 2\tau_c/\Delta)}_{f_I}} \frac{\overbrace{(e^{\Delta\mu_1} - 1)(e^{\Delta\mu_2} - 1)}^q}{\underbrace{e^{\Delta\mu} - 1}_{\bar{N}_{\text{eff}}}} \frac{\overbrace{\dot{n}\tau_r}^{\bar{N}}}{p}. \quad [S10]$$

To see the correspondence between Eqs. S9 and S10 (i.e., Eqs. 2 and 3 of the main text) and Eq. S8, note that variables in Eqs. S9 and S10 can be expressed in terms of the rate constants as follows:

$$p = \frac{\tilde{k}_1 L}{\tilde{k}_1 L + k_2} = \frac{k_1}{k_1 + k_2}, \quad [S11]$$

$$\tau_c = \frac{1}{\tilde{k}_1 L + k_2} = \frac{1}{k_1 + k_2}, \quad [S12]$$

$$\tau_r = \frac{1}{k_{-r} + k_r + k_f p R_T + k_{-f} p R_T}, \quad [S13]$$

$$\dot{n} = k_f \overline{RL\bar{x}} - k_{-f} \overline{RLx^*} = \frac{(k_f k_r - k_{-f} k_{-r}) p R_T X_T}{k_{-r} + k_r + (k_f + k_{-f}) p R_T}, \quad [S14]$$

$$\Delta\mu_1 = \log \frac{k_f \bar{x}}{k_{-f} x^*} = \log \frac{k_f (k_r + k_{-f} p R_T)}{k_{-f} (k_{-r} + k_f p R_T)}, \quad [S15]$$

$$\Delta\mu_2 = \log \frac{k_r \bar{x}^*}{k_{-r} x} = \log \frac{k_r (k_{-r} + k_f p R_T)}{k_{-r} (k_r + k_{-f} p R_T)}, \quad [S16]$$

$$\begin{aligned} \Delta &= 2\tau_r / (\bar{N}_{\text{eff}} / R_T) = \frac{2(k_{-f} k_{-r} - k_f k_r) \bar{x}^*}{(k_f \bar{x} - k_{-f} x^*)^2 (k_{-r} \bar{x} - k_f x^*)} \\ &= \frac{2(k_{-r} + k_f p R_T) (k_r + k_{-f} p R_T) (k_{-r} + k_r + (k_f + k_{-f}) p R_T)}{(k_{-f} k_{-r} - k_f k_r)^2 p R_T X_T}. \end{aligned} \quad [S17]$$

The relaxation time τ_r is calculated from the eigenvalues of the A matrix defined in *Calculating the Sensing Error for a Biochemical Network*. The free-energy drops are calculated following ref. 8. Substituting the expressions for these variables into Eqs. S9 and S10 (i.e., Eqs. 2 and 3 of the main text) yields Eq. S8 in terms of the rate constants.

The Inspiration for Eqs. 2 and 3 of the Main Text: The Signaling Network Viewed as a Sampling Device. The inspiration for Eq. 2 of the main text (Eq. S9) came from an analysis of a simpler system. For this simpler system, we can analytically obtain the sensing error by viewing the signaling network as a device that samples the receptor state. As we will show below, the expression for the sensing error in this simpler model (Eq. S18) has exactly the same form as Eq. 2 of the main text (Eq. S9), corresponding to the full system. Indeed, Eq. S18 provided the motivation to cast the expression for the sensing error of the full system (Eq. S8) in the form of Eq. 2 of the main text (Eq. S9).

We introduce this simpler model step by step: we first describe how samples are taken, and then how samples are erased. The derivation of the sensing error using the perspective of sampling is relegated to the appendices; below, we give the results. We then extend the model to discuss the effect of the finite pool of readout molecules and the reliability of the samples. In doing so, we will arrive at the full model of the main text.

Base Model: Taking Samples via Readout Activation. We consider a cell that responds at time T to a change in ligand concentration at an earlier time $t=0$, based on the output x^* of the simple reaction network $x + RL \xrightarrow{k_f} x^* + RL$ (Fig. 2B of the main text). We assume that the cell starts with a large pool of inactive readout molecules x and that activated molecules x^* are never deactivated. For descriptive ease, we assume the reaction is diffusion-limited, so that each collision between an inactive molecule x and a ligand-bound receptor leads to activation of x .

Readout molecules that collide with the receptor over time are modified depending on the ligand-occupation state of the receptor. The total rate at which inactive molecules collide with receptor molecules in any state (be they ligand bound or not) is $r = k_f x R_T \approx k_f X_T R_T$ for a large readout pool, and the total number of such collisions after time T is N , with $\bar{N} = rT$ on average. If a receptor molecule is bound to ligand at the time of a collision, the readout molecule is converted to its active form, whereas if it is not the readout remains unchanged. In this way, the state of the receptor at the time of a collision is encoded in the state of the readout molecule that collided with it, and the history of the receptor states is encoded in the states of the readout molecules at the time T (Fig. 2C). The readout molecules that collided with the receptor thus constitute samples of the receptor state. The average number of samples after time T is $\bar{N} = rT = k_f X_T R_T T$ —the product of the total number of receptors R_T and the number of samples per receptor $k_f X_T T$ during the integration time T .

The resulting sensing error can be derived via Eq. 1 of the main text (i.e., Eq. S7) from the chemical master equation, which describes fluctuations in the network (see *Calculating the Sensing Error for a Biochemical Network*, noting that the linear-noise approximation is exact for linear networks). The same expression can also be derived independently by viewing the signaling network as a system that samples the receptor state and using tools from discrete stochastic processes. This supports the idea that the signaling network truly samples the ligand-binding state of the receptor. This analysis is given in *Appendix A*. Here, we give the result. Interestingly, the sensing error for the base model has exactly the same form as Eq. 2 of the main text (Eq. S9) for the full model:

$$\left(\frac{\delta c}{c}\right)^2 = \frac{1}{p(1-p)} \frac{1}{\bar{N}_I} + \frac{1}{(1-p)^2} \frac{1}{\bar{N}}. \quad [\text{S18}]$$

Here, as in the main text, \bar{N} is the total number of samples and \bar{N}_I is the number of samples that are independent. The latter is given by the following:

$$\bar{N}_I = f_I \bar{N} = \frac{1}{1 + \frac{2\tau_c}{\Delta}} \bar{N}, \quad [\text{S19}]$$

when $T \gg \tau_c$. Here, $f_I = 1/(1 + (2\tau_c/\Delta))$ is the fraction of samples that is independent, which depends on the receptor–ligand correlation time τ_c and the time interval $\Delta \equiv T/(\bar{N}/R_T) = 1/(k_f X_T)$ between samples of the same receptor. This expression shows that the finite sampling rate r reduces the number of independent samples below the Berg–Purcell factor $R_T T/(2\tau_c)$, the maximum number of independent samples that can be taken during T . The latter is reached only when the sampling rate is infinite (e.g., the number of downstream molecules $X_T \rightarrow \infty$), so that $\bar{N} \rightarrow \infty$ and $\Delta \rightarrow 0$.

Erasing Samples via Readout Deactivation. The error in Eq. S18 decreases with the time T , suggesting that the cell can sense perfectly if it waits long enough before responding to a change in its environment. However, modification states of molecules decay, and their finite lifetime, τ_l , limits sensing, regardless of how long the cell waits. To explore this at the molecular level, we consider the network in the previous paragraph augmented with the deactivation reaction, $x^* \xrightarrow{k_d} x$, $k_r = 1/\tau_l$ (Fig. 2D of the main text). We consider the sensing error after long times ($T \gg \tau_l$), in steady state, again for a large pool of inactive readout molecules. For pedagogical clarity, we imagine the deactivation is mediated by a phosphatase and that the reaction is diffusion-limited.

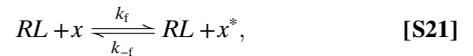
We can calculate the sensing error by solving the master equation or by viewing the system as one that discretely samples the receptor state, as described in *Appendix A*. It shows that the sensing error is again given by Eq. S18. However, the number of samples is lower than that in the case without deactivation, $\bar{N} = r\tau_l < rT$, and they are spaced effectively farther apart, $\Delta = 2\tau_l/(\bar{N}/R_T) = 2/(k_f X_T) > 1/(k_f X_T)$. The molecular picture of sampling provides a clear interpretation. As before, the readout molecules encode the state of a receptor and serve as samples of the receptor state. With deactivation, however, only those readout molecules that have collided with the receptor more recently than with the phosphatase reflect the receptor state. At any given time, the average number of such readout molecules, and hence samples, is $\bar{N} = r\tau_l$; the lifetime τ_l thus sets an effective integration time. As without deactivation, the fraction f_I of samples that are independent is determined by the effective spacing Δ between them. Although the time between the creation of samples is still $1/(k_f X_T)$, i.e., the spacing without readout deactivation, some of the samples are erased via collision with the phosphatase. We therefore expect that the spacing between remaining samples

is larger. Indeed, calculating the effective spacing between samples taking this effect into account yields $\Delta = 2/(k_f X_T)$, which is twice that without decay (*Appendix B*).

Beyond the Simple Model: Finite Pool of Readout Molecules. The copy numbers of signaling molecules are often small. To take this into account, we compute the sensing error from Eq. 1 of the main text for a finite number of readout molecules X_T using the linear-noise approximation to the master equation describing the biochemical fluctuations (see *Calculating the Sensing Error for a Biochemical Network*). Again, the sensing error can be written in the form given by Eqs. S18 and S19. This defines an effective number of samples, $\bar{N} = r\tau_r$, where r is, as before, the receptor sampling rate, and τ_r is the relaxation time of the network. The sampling rate $r = k_f \bar{x} R_T$, whereas $\tau_r = 1/(k_f p R_T + k_r)$. In essence, cells count only those samples created less than a relaxation time in the past; nothing that happened earlier can influence the current state, including its ability to sense. The fraction of samples that is independent is given by Eq. S19 with $\Delta = 2\tau_r/(\bar{N}/R_T) = 2/(k_f \bar{x})$, analogously to the previous paragraph.

Reliability of the Samples: The Full Model. All reactions are in principle microscopically reversible. Taking this into account, we recognize that active molecules that collide with the bound receptor sometimes become inactive, $x^* + RL \xrightarrow{k_{-f}} x + RL$, and that inactive molecules that collide with the phosphatase are sometimes activated, $x \xrightarrow{k_{-r}} x^*$ (Fig. 2E of the main text). These reverse reactions compromise the encoding of the receptor state into the readout, as described in the main text: an active x^* molecule no longer encodes the ligand-bound state of the receptor at a previous time with 100% reliability, because it can also result from a collision with the phosphatase; similarly, x , rather than x^* , may reflect a collision with the ligand-bound receptor.

Taking the reverse reactions into account, the full model of the main text is given by the following reactions:



From Eqs. 2 and 3 of the main text (Eqs. S9 and S10), it is seen that the error in the full model with reversible reactions has the same form as Eq. S18 above, but with the effective number of independent samples \bar{N}_I given by Eq. S10: $\bar{N}_I = f_I q \bar{N}$, where q is the quality factor of the sample. The quality factor q is zero when the network is not driven out of equilibrium and reaches unity when the network is driven far out of equilibrium. In the latter case, when $q \rightarrow 1$, this model reduces to that of the previous paragraph. Indeed, in this irreversible limit, the effective number of samples \bar{N}_{eff} as defined by Eq. 3 of the main text is $\bar{N}_{\text{eff}} = \bar{N} = \bar{n} \tau_r/p = r\tau_r = k_f \bar{x} R_T \tau_r$, with the flux \bar{n} in Eq. 3 of the main text reducing to p , with p , the probability that a receptor is bound to ligand, and $r = k_f \bar{x} R_T$, the rate at which the receptor is sampled in the irreversible model of the previous paragraph.

Derivation of Eq. 4 of the Main Text

Resources or combinations of resources that fundamentally limit sensing, are those (collective) variables Q_i that, when fixed, set a lower nonzero bound on the sensing error, no matter how the other variables in the system are varied. If we denote all of the eight variables in the system by

$k = \{k_1, k_2, k_f, k_{-f}, k_r, k_{-r}, R_T, X_T\}$, then the fundamental variables Q_i are defined by the following:

$$\text{MIN}_{k; Q_i = \text{const}} (\delta c / c)^2 = f(\text{const}) > 0. \quad [\text{S23}]$$

To find these fundamental collective variables, we numerically or analytically minimized the sensing error, constraining combinations of variables yet allowing the other variables not to be limiting. This procedure identified three fundamental resource classes: $Q_1 = R_T \tau_r / \tau_c$, $Q_2 = X_T$, and $Q_3 = \dot{w} \tau_r$. Each class puts a fundamental lower bound on the sensing error, which means that the sensing error is limited by the value of Q_i and cannot be reduced to zero by increasing another variable. We now derive the sensing limits for the respective resource classes.

Receptors and Their Integration Time, $R_T \tau_r / \tau_c$, Limiting. When only $R_T \tau_r / \tau_c$ is constrained, the other resources can be optimized. The sensing error is minimized when the system is strongly driven out of equilibrium, $\Delta \mu_1, \Delta \mu_2 \gg 0$. The receptor samples are then stored in the readout x with 100% fidelity and the quality factor reaches unity, $q \rightarrow 1$ (see Eq. 3 of the main text or Eq. S10 above). In this limit, we can ignore the microscopically reverse reactions, and the readout flux $\dot{n} = k_f \bar{x} R_T p - k_{-f} \bar{x}^* R_T p$ reduces to $\dot{n} = k_f \bar{x} R_T p$. This is indeed the rate at which the readout x is activated by ligand-bound receptor; it is thus the rate at which samples of the ligand-bound receptor are taken. The total number of samples of the receptor $\bar{N} = \dot{n} \tau_r / p$ obtained during the integration time τ_r therefore reduces to $\bar{N} = k_f \bar{x} R_T \tau_r$. Because $q = 1$, this is also the effective number of samples: $\bar{N}_{\text{eff}} = \bar{N} = k_f \bar{x} R_T \tau_r$. The sensing error is minimized when X_T and \bar{x} become infinite. The number of samples \bar{N}_{eff} then becomes infinite, but the spacing between the samples $\Delta = 2 \tau_r / (\bar{N}_{\text{eff}} / R_T) = 2 / (k_f \bar{x})$ becomes zero. Consequently, the number of independent samples becomes $\bar{N}_I = R_T \tau_r / \tau_c$ (see Eq. 3 or Eq. S10). Indeed, in the limit that the receptors and their integration time are limiting, the number of independent measurements \bar{N}_I reaches the Berg–Purcell factor, i.e., the number of receptors R_T times the number of independent measurements per receptor τ_r / τ_c . The bound on the sensing error is given by the first term of Eq. 2 of the main text or Eq. S9 above, which with $\bar{N}_I = R_T \tau_r / \tau_c$ yields the following:

$$\left(\frac{\delta c}{c}\right)_{R_T \tau_r / \tau_c}^2 \geq \frac{1}{p(1-p)R_T \tau_r / \tau_c} \geq \frac{4}{R_T \tau_r / \tau_c}, \quad [\text{S24}]$$

because the maximum of $p(1-p)$ is 0.25, obtained when $p = 0.5$.

Number of Readout Molecules X_T Limiting. The bound associated with the second resource class X_T can be derived from Eqs. 2 and 3 of the main text (Eqs. S9 and S10 above), but perhaps the most straightforward derivation is directly from Eq. 1 of the main text (i.e., Eq. S7 above). When X_T becomes limiting, the variance of x is dominated by the intrinsic binomial switching noise—the extrinsic contribution from the receptor becomes vanishingly small. This gives $\sigma_x^2 = f(1-f)X_T$, where $f = pk_f R_T / (pk_f R_T + k_r)$ is the fraction of X_T that is phosphorylated. The gain $\partial \bar{x}^* / \partial c = X_T f(1-f)(1-p)/c$. Together, this yields from Eq. 1 of the main text (Eq. S7 above):

$$\left(\frac{\delta c}{c}\right)_{X_T}^2 = \frac{1}{f(1-f)(1-p)^2 X_T} \geq \frac{4}{X_T}. \quad [\text{S25}]$$

Energy w Is Limiting. In the scenario that $w = \dot{w} \tau_r = \dot{n} \Delta \mu \tau_r$ is limiting and the other resources are in abundance, $f_I = 1$ and $\bar{N}_I = \bar{N}_{\text{eff}} = q \dot{n} \tau_r / p = q w / (p \Delta \mu)$ (see Eq. 3 of the main text and Eq. S10 above). The number of samples of ligand-bound receptor per amount of work w is $p \bar{N}_{\text{eff}} / w = q / \Delta \mu$. It is maximized when

$\Delta \mu_1 = \Delta \mu_2 = \Delta \mu / 2$ and $\Delta \mu \rightarrow 0$. In this limit, the work to take one effective sample of a ligand-bound receptor reaches its lower bound of $4k_B T$: $w / (p \bar{N}_{\text{eff}}) = 4$, as described in the main text. The number of independent measurements in this limit is $\bar{N}_I = w / (4p)$. Together with Eq. 2 of the main text (Eq. S9 above), this yields the following:

$$\left(\frac{\delta c}{c}\right)_w^2 \geq \frac{4}{(1-p)w} \geq \frac{4}{w}. \quad [\text{S26}]$$

When all resources are present in finite amounts, the minimum sensing error is given by the highest of the above lower bounds. This yields Eq. 4 of the main text:

$$\left(\frac{\delta c}{c}\right)^2 \geq \text{MAX} \left(\frac{4}{R_T \tau_r / \tau_c}, \frac{4}{X_T}, \frac{4}{\dot{w} \tau_r} \right). \quad [\text{S27}]$$

The resource classes $R_T \tau_r / \tau_c$, X_T , and $\dot{w} \tau_r$ are indeed fundamental. They each set a fundamental lower bound on the sensing error, which cannot be beaten by increasing another resource. For example, when $R_T \tau_r / \tau_c$ is constrained, the minimum sensing error can never be lower than the bound given by Eq. S24, no matter how much X_T is increased or no matter how much the power \dot{w} is raised. In contrast, R_T is not fundamental: for a given R_T , the sensing error can be reduced to zero by increasing τ_r . Similarly, the work w is fundamental: for a given w , the sensing error can never be lower than the bound set by Eq. S26. The power \dot{w} , however, is not fundamental: for a given \dot{w} , the error can be reduced to zero by increasing the integration time. Finally, we note that the factors 4 in Eqs. S24–S26 have a common origin: for each resource class, the optimal consumption of the resource occurs when it is equally partitioned into two pools: optimal allocation of energy occurs when the total energy drop is equally partitioned into the energy drops of activation and deactivation; the receptors are optimally allocated when one-half of them is ligand bound; and the readout pool is optimally allocated when it is equally partitioned between active and inactive molecules.

Connection with Optimal Resource Allocation Principle of Eq. 5 of the Main Text

Eq. 4 of the main text (Eq. S27) identifies the fundamental resource classes in cellular sensing systems. These resource classes cannot compensate each other in achieving a desired sensing precision, which is illustrated in Fig. 3 of the main text (how this figure is generated is described in the section below). Fig. 3 shows that the minimum sensing error tracks the highest of the lower bounds of Eqs. S24–S26: the minimum sensing error is set by the limiting resource class. This observation leads to the design principle of optimal resource allocation embedded in Eq. 5 of the main text, which states that, in an optimally designed system, all fundamental resource classes are equally limiting so that none is wasted. Importantly, this design principle does not depend on the relative costs of the resources. The reason is that fundamental resources that are in excess, cannot improve sensing and are thus wasted, no matter how cheap they are. Close to the optimum where all resources are equally limiting, the precise costs of the resources may become important, because, as Fig. 3 shows, close to the optimum there is, e.g., via the receptor, cross talk between the fundamental resource classes in setting the sensing precision: at the intersection of the dashed gray lines corresponding to the lower bounds of the respective resource classes, the minimum error (red line) is higher than the lower bound given by Eq. S27—as expected, the minimum sensing error crosses over smoothly from one bound to another, and not discontinuously. However, as Fig. 3 shows, even in the crossover regime the minimum error (red line) is never much higher than the lower bound of Eq. S27 (the dashed-gray lines). Even close to the optimum, the mutual dependence between the resource classes is

thus small. It is probably for this reason that the optimal resource allocation principle of Eq. 5, which does not rely on resource costs, gives such a surprisingly accurate prediction of the quantitative relationship between the number of receptors, readout molecules, integration time, receptor correlation time, and energy in the *E. coli* chemotaxis system.

Fig. 3 of the Main Text: No Trade-Offs Among Resource Classes

In Fig. 3B of the main text, we show how the sensing error depends on the pair of resources (readout copy number X_T , energy w). These results were obtained via numerical minimization of Eqs. 2 and 3 of the main text subject to constraints on X_T and w .

In Fig. 3C of the main text, we show how the sensing error depends the pair of resources (time/receptor copy number, energy). The plot for (time/receptor copy number, readout copy number) is the same. In this section, we describe the derivation of the results shown in this figure. To consider τ_r/τ_c not necessarily large, we need to use a form of the Berg–Purcell bound that is valid for short integration times (9):

$$\left(\frac{\delta c}{c}\right)_{\min}^2 > \frac{1}{p(1-p)} \frac{1}{R_T \left(1 + \frac{\tau_r}{\tau_c}\right)}, \quad [\text{S28}]$$

which identifies $R_T(1 + \tau_r/\tau_c)$ as a limiting resource, rather than the result of the main text, $R_T\tau_r/\tau_c$, which only holds in the limit $\tau_r \gg \tau_c$.

To elucidate how the sensing error depends on (time/receptor copy number, energy) and (time/receptor copy number, readout copy number), we calculate the minimum sensing error by optimizing over all parameters while fixing $R_T(1 + \tau_r/\tau_c)$ and either w or X_T , respectively. For a fixed $R_T(1 + \tau_r/\tau_c)$ and a fixed work w , the minimum sensing error is as follows:

$$\begin{aligned} \left(\frac{\delta c}{c}\right)_{\min}^2 = & \frac{w}{32} \left(-\frac{1}{\left(R_T \left(1 + \frac{\tau_r}{\tau_c}\right)\right)^2} \right. \\ & + \sqrt{\frac{1}{R_T \left(1 + \frac{\tau_r}{\tau_c}\right)} \left(\frac{1}{R_T \left(1 + \frac{\tau_r}{\tau_c}\right)} + \frac{32}{w} \right)^{3/2}} \\ & \left. + \frac{128}{w^2} + \frac{80}{w \left(R_T \left(1 + \frac{\tau_r}{\tau_c}\right)\right)} \right). \quad [\text{S29}] \end{aligned}$$

The equation for the dependence of the sensing error on (time/receptor copy number, readout copy number) is the same, with w replaced by X_T . The minimum is plotted in Fig. 3C. The minimum tracks the worst bound, again showing that the resources do not compensate each other.

Additional constraints on the values of rate constants will generally prevent the network from achieving these bounds. In particular, it is common to consider that the binding of ligand to receptor is diffusion-limited, so that the bound $4/(R_T(1 + \tau_r/\tau_c))$ is never achieved. Of course, additional constraints cannot improve the performance of the network beyond the bounds required here, nor can they alter the fact that all of the resources are needed for sensing.

Additional Networks

Networks are often more complicated than a simple one-level push–pull cascade. We investigate some common motifs to

understand whether they relax the trade-offs faced by sensory networks.

Multilevel Cascades. Often the signaling molecule activated by the receptor is not taken as the final readout; rather that molecule catalyzes the activation of another molecule, and so on in a signaling cascade. All of the molecules are reversibly degraded. Using the same approach as for the one-level cascade, we find that the sensing error is bounded by the work done driving just the last step of the cascade: $\bar{N}_{\text{eff}} \leq \dot{w}_i \tau_r / (4p)$, where $\dot{w}_i = \dot{n}_i \Delta \mu_i$ is the product of the flux of the last molecule through its cycle and the free-energy drop across that cycle, and τ_r is the slowest relaxation time in the cascade (i.e., the reciprocal of the largest eigenvalue of the relaxation matrix). Even more work is done at other levels of the cascade. The results suggest that cascades do not enable more energy efficient sensing. Additionally, each sample of an active state (bound receptor or active molecule upstream) still requires a molecule to store it.

Positive and Negative Feedback. A simple model of positive feedback is autocatalysis, in which the receptor-catalyzed activation of the readout is enhanced by the activated form of the readout, $x^*: x + x^* + RL \rightleftharpoons 2x^* + RL$. A simple model of negative feedback can be implemented by requiring inactive x for the activation: $2x + RL \rightleftharpoons x + x^* + RL$. In both cases, x^* degrades according to $x^* \rightleftharpoons x$. Neither positive feedback nor negative feedback changes the energetic requirements for sensing: $\bar{N}_{\text{eff}} = (\dot{n}\tau_r/p)(e^{\Delta\mu_1} - 1)(e^{\Delta\mu_2} - 1)/(e^{\Delta\mu} - 1)$. As before, the free-energy drops across the reactions were calculated as the ratio of mass-action propensities.

Cooperative Activation of the Readout. If the catalytic activation of the readout is mediated cooperatively by the receptors (i.e., $x + nRL \rightleftharpoons x^* + nRL$), then the error is reduced by a factor n^2 for the same amount of energy. One way to interpret the result is that each sample requires the same amount of energy as before, but the samples are individually more informative because they reflect n ligand bindings, instead of one—indeed, the instantaneous error is lower.

Appendix A: Discretely Sampling the Receptor State

In this appendix, we show for the simple model consisting of $x + RL \xrightarrow{k_t} x^* + RL$ and $x^* \xrightarrow{k_r} x$, how the sensing error can be calculated by viewing the network as a discrete sampling device. The important quantities in a sampling protocol are the number of samples taken and the spacing between them, in addition to the properties of the sampled signal. By viewing the biochemical process as a sampling process, we mean that the underlying parameters of the biochemical network affect the sensing error only insofar as they affect these quantities, or the stochasticity in these quantities. The benefit of viewing the network as a sampling process is that the number of samples and the spacing between them have intuitive, and well-known, effects on the sensing error: the more samples, the lower the error; the further apart the samples are, the more independent they are. Perhaps less well known are the effects on the sensing error of stochasticity in the number of samples or the spacing between samples; these effects emerge in the process of determining the error for a discrete sampling protocol, which we do below.

We consider the base model of readout activation, $x + RL \xrightarrow{k_t} x^* + RL$, and the base model of readout activation plus readout deactivation, $x + RL \xrightarrow{k_t} x^* + RL$ and $x^* \xrightarrow{k_r} x$. For the base model, we identify the molecules that have collided with the receptors as samples, because these molecules' states reflect the receptor states at the times of their collisions with the receptor. For the model with deactivation, we identify the molecules that

collided with the receptor more recently than with the phosphatase as samples. When we refer to the number of samples, we mean the number of these molecules; when we refer to the times of the samples, we mean the times at which these molecules collided with the receptor.

We begin by rewriting the equation for the sensing error in a form that makes the connection to discrete sampling explicit, Eq. S33 below. The cell senses its environment through the level of its readout x^* . However, this is no different from estimating the ligand concentration from $\hat{p} = x^*/\bar{N}$:

$$\left(\frac{\delta c}{c}\right)^2 = \frac{\sigma_{\hat{p}}^2}{\left(\frac{d\bar{p}}{d\mu_L}\right)^2} = \frac{\sigma_{x^*}^2}{\left(\frac{dx^*}{d\mu_L}\right)^2}, \quad [\text{S30}]$$

because \bar{N} is a constant, independent of μ_L . Note that the gain $d\bar{p}/d\mu_L$ is $d\bar{p}/d\mu_L = p(1-p)$.

We first consider the effect of the stochasticity in the total number of samples, N . The law of total variance allows us to decompose the variance in the estimate \hat{p} into terms arising from different sources:

$$\sigma_{\hat{p}}^2 = E[\text{var}(\hat{p}|N)] + \text{var}[E(\hat{p}|N)]. \quad [\text{S31}]$$

The first term of Eq. S31 reflects the mean of the variance in \hat{p} given the number of samples N ; the second term reflects the variance of the mean of \hat{p} given the number of samples N .

The mean and variance of \hat{p} given the number of samples N are more familiar quantities than their unconditioned counterparts, as we see below. Because, by definition, the samples reflect the state of the receptor at the times t_i of their collisions with the receptor, we can write the number of x^* at the final time as follows:

$$x^* = \sum_{i=1}^N n_i(t_i), \quad [\text{S32}]$$

where $n_i(t_i)$ denotes the value of the i th sample—the state of the receptor involved in the i th collision at the time t_i of that collision, 1, if bound to ligand, and 0, otherwise. In the following, we consider a single receptor, $R_T = 1$ and $n = n_i$. The results generalize to multiple receptors.

We can then rewrite Eq. S31 as follows:

$$\sigma_{\hat{p}}^2 = E\left[\frac{N^2}{N^2} \text{var}\left(\frac{\sum_{i=1}^N n(t_i)}{N} \middle| N\right)\right] + \text{var}\left[\frac{N}{N} E\left(\frac{\sum_{i=1}^N n(t_i)}{N} \middle| N\right)\right]. \quad [\text{S33}]$$

The equation is a bit complicated, but what is important is that it fully specifies the sensing error in terms of the number of samples, the spacings between them, and the stochasticity in these quantities. That is, this equation shows that the sensing error is the error of a sampling process. We can use it to calculate the sensing error independently from, for example, the master equation or the linear-noise approximation.

The first term describes the error of a very standard sampling process, one with a fixed number of samples. We recognize the variance,

$$\text{var}\left(\frac{\sum_{i=1}^N n(t_i)}{N} \middle| N\right), \quad [\text{S34}]$$

as the error of a statistical sampling protocol in which exactly N samples are taken at random times t_i . This is shown explicitly in Appendix B. In that appendix, it is shown that the error for such a sampling protocol is as follows:

$$\text{var}\left(\frac{\sum_{i=1}^N n(t_i)}{N} \middle| N\right) = p(1-p) \frac{1}{f_I N}, \quad [\text{S35}]$$

where f_I is the fraction of the samples that are independent, as given by Eq. S19. Then the first term in Eq. S33 is just the following:

$$E\left[\frac{N^2}{N^2} \text{var}\left(\frac{\sum_{i=1}^N n(t_i)}{N} \middle| N\right)\right] = E\left[\frac{N^2}{N^2} p(1-p) \frac{1}{f_I N}\right] = p(1-p) \frac{1}{f_I N}. \quad [\text{S36}]$$

That is, the first term in Eq. S33 is the error of a discrete sampling protocol with exactly \bar{N} samples, as stated in the main text. The only effect of the expectation in the first term is to swap \bar{N} for N . Dividing by the squared gain (Eq. S30), $d\bar{p}/d\mu_L = p(1-p)$, gives the first term in Eq. 2 in the main text and Eq. S18 above.

We now turn to the second term in Eq. S33. From the law of total variance, this term describes how stochasticity in the number of samples, N , contributes to the sensing error. Because the number of samples N is Poisson with mean and variance equal to \bar{N} :

$$\text{var}\left[\frac{N}{N} E\left(\frac{\sum_{i=1}^N n(t_i)}{N} \middle| N\right)\right] = \text{var}\left[\frac{N}{N} p\right], \quad [\text{S37}]$$

$$= \frac{p^2}{N^2} \text{var}[N], \quad [\text{S38}]$$

$$= \frac{p^2}{N}, \quad [\text{S39}]$$

where the probability a receptor is bound is $E[n(t_i)] = p$. Dividing by the squared gain gives the second term in Eq. 2 in the main text and Eq. S9 above. Thus, we have derived Eq. 2 in the main text as the result of a discrete sampling protocol.

The second term in Eq. 2 in the main text (i.e., Eq. S9) emerges in the derivations above as a consequence of the stochasticity in the number of samples N . However, it is more fundamentally a consequence of the fact that the cell does not distinguish between samples of the unbound receptor from blank samples that do not represent a receptor state—i.e., it does not distinguish x molecules that collided with the unbound receptor from those that never collided with the receptor in any state. A more standard sampling procedure would distinguish between these, and so would estimate \hat{p} as $\hat{p} = x^*/N$, not $\hat{p} = x^*/\bar{N}$, as above. As we show in Appendix C, this procedure gives rise to only the first term of Eq. 2 in the main text, allowing us to interpret the second term as the price the cell pays for not distinguishing readout molecules that collide with the unbound receptor from those that have never collided with the receptor in any state.

The derivations leading to Eq. S33 show that the sampling error for the sampling protocol must be the same as the sensing error for the biochemical network. To check this, we can calculate the sensing error for the biochemical network, Eq. 2 in the main text, in a more standard way, determining the gain and the variance of the output x^* and using Eq. 1 in the main text. This indeed gives exactly the same result.

Appendix B: Error of Discrete Sampling Protocols with a Fixed Number of Samples

In this section, we derive the first term of Eqs. S31 and S33, corresponding to the first term of Eq. 2 in the main text (Eq. S18), as the error of a discrete sampling protocol with a fixed number of samples N taken of receptor states over time. The average receptor occupancy is estimated as the following:

$$\hat{p} = \frac{1}{N} \sum n_i(t_i), \quad [\text{S40}]$$

where $n_i(t_i)$ is the state of the receptor involved in the i th sample at the time of that sample, 1, if the receptor was bound at time t_i , and 0, otherwise. In what follows, we consider a single receptor, $R_T = 1$ and $n(t_i) = n_i(t_i)$. The results generalize to multiple receptors. The times t_i of the samples represent the times at which the molecules that store the samples of the receptor collided with the receptor. Therefore, we choose the distribution of times between the samples to match the distribution of times between those collisions, which depends on the particular network under consideration, described below. We count time backward from the present time, $t = 0$. The number of samples N and the distribution of times at which they were taken specify a sampling protocol, independent of the chemical implementation.

The variance in the estimate of receptor occupancy is as follows:

$$\sigma_{\hat{p}}^2 = \text{var} \left(\frac{\sum_{i=1}^N n(t_i)}{N} \right), \quad [\text{S41}]$$

$$= \frac{\text{var} \left(\sum_{i=1}^N n(t_i) \right)}{N^2}, \quad [\text{S42}]$$

$$= \frac{\sigma^2}{N} + \frac{N(N-1)}{N^2} E[\text{cov}(n(t_i), n(t_j))], \quad [\text{S43}]$$

because N is fixed, where $\sigma^2 = p(1-p)$ is the variance of the instantaneous occupancy of a single receptor.

Base Model with Activation Only, $x + RL \xrightarrow{k_1} x^* + RL$. We first consider a statistical sampling protocol that matches the distribution of receptor-collision times of samples in the base model. The collisions occur at random times in the interval $[0, T]$, so we model N randomly placed samples. The time $\tilde{\Delta}$ between a randomly chosen pair of uniformly distributed samples, not necessarily consecutive, is distributed as follows:

$$p(\tilde{\Delta}) = \frac{2}{T} - \frac{2\tilde{\Delta}}{T^2}. \quad [\text{S44}]$$

Changing variables from t_i and t_j to $\tilde{\Delta} = |t_j - t_i|$, we have $\text{cov}(n(t_i), n(t_j)) = \sigma^2 e^{-\tilde{\Delta}/\tau_c}$. The expectation of the covariance is then the following:

$$E[\text{cov}(n(t_i), n(t_j))] = \sigma^2 \int e^{-\tilde{\Delta}/\tau_c} p(\tilde{\Delta}) d\tilde{\Delta}. \quad [\text{S45}]$$

Assembling the equations above yields the first term in Eq. 2 in the main text (Eq. S18) with $\Delta = T/N$ ($R_T = 1$), where we have simplified the result with the standard assumption that $T \gg \tau_c$ and $N \gg 1$ (it does not make sense to discuss the spacing between a single sample).

Base Model with Activation Plus Deactivation, $x + RL \xrightarrow{k_1} x^* + RL$ and $x^* \xrightarrow{k_2} x$. To take into account deactivation, we consider sampling times which match the distribution of the receptor collisions of only those N molecules storing samples. We thus have to take into account that some of the samples that have been taken are thrown away due to the deactivation process. We begin with an alternative expression for the expected covariance:

$$E[\text{cov}(n(t_i), n(t_j))] = \sigma^2 \int \int e^{-|t_j - t_i|/\tau_c} p(t_i, t_j) dt_i dt_j. \quad [\text{S46}]$$

To match the biochemical network, the sample times t_i, t_j of two samples must be independent from each other, because the collisions of different molecules with the receptor and phosphatase are uncoupled. Therefore, $p(t_i, t_j) = p(t_i)p(t_j)$. The marginal probability $p(t_i)$ is the probability that the collision time with the receptor of a given molecule storing a sample was t_i , i.e., $p(t_i|\text{sample})$. This can be written in terms of $p(\text{sample}|t_i)$, the probability that there was a collision with the receptor at the time t_i times the probability that, given a collision at that time, the associated molecule did not subsequently collide with the phosphatase:

$$p(\text{sample}|t_i) dt = r dt e^{-t_i/\tau_c}. \quad [\text{S47}]$$

Then:

$$p(t_i|\text{sample}) = \frac{p(\text{sample}|t_i)p(t_i)}{\int p(\text{sample}|t_i)p(t_i) dt_i} = \frac{1}{\tau_c} e^{-t_i/\tau_c}, \quad [\text{S48}]$$

because $p(t_i)$ is uniform.

Assembling results:

$$E[\text{cov}(n(t_i), n(t_j))] = \sigma^2 \int \int e^{-|t_j - t_i|/\tau_c} \frac{e^{-t_i/\tau_c}}{\tau_c} \frac{e^{-t_j/\tau_c}}{\tau_c} dt_i dt_j. \quad [\text{S49}]$$

It is instructive to change variables, defining $\tilde{\Delta} = |t_j - t_i|$, as before. Then:

$$E[\text{cov}(n(t_j), n(t_i))] = \sigma^2 \int_0^\infty e^{-\tilde{\Delta}/\tau_c} \frac{e^{-\tilde{\Delta}/\tau_c}}{\tau_c} d\tilde{\Delta}. \quad [\text{S50}]$$

From this expression we can identify $p(\tilde{\Delta}) = e^{-\tilde{\Delta}/\tau_c}/\tau_c$ as the distribution of times between two randomly chosen (not necessarily consecutive) samples, when molecules can decay. Simulations confirm this distribution.

Completing the integral and using it in the expression for the sensing error gives the first term in Eq. 2 in the main text (Eq. S18) for the effective spacing $\Delta = 2\tau_c/N$ (here, $R_T = 1$). We have made the simplifying assumptions that $N \gg 1$ (it does not make sense to talk of the spacing between just one sample) and $\tau_c \gg \tau_c$, a standard assumption. The effective spacing is not the mean nearest-neighbor spacing, but it is qualitatively similar and serves to summarize the fact that samples taken further apart in time are more independent. Clearly, from Eq. S50, the error depends on the distribution of all-pairs spacings, not necessarily nearest-neighbor spacings, and it depends on the full distribution, not just the mean.

Finally, we iterate that we can perform an independent check on the derivation in this section by computing the sensing error using the linear-noise approximation, which is exact for this linear network. As mentioned, this gives exactly the same result.

Appendix C: The Origin of the Second Term in Eq. 2 in the Main Text

The origin of the second term in Eq. 2 of the main text (i.e., Eq. S18) is that the cell cannot distinguish between those readout molecules that have collided with ligand-unbound receptors and readout molecules that have not collided with the receptors at all. One way to arrive at this conclusion is to imagine that all collisions with the receptor lead to modifications of x . However, although the ligand-bound receptor modifies x into state x^* , the unbound receptor modifies x into another state x^\dagger . Hence, in addition to the reaction $x + RL \rightarrow x^* + RL$, we consider the reaction

$x + R \rightarrow x^\dagger + R$. Then, $N = x^* + x^\dagger$. Analogously to Eq. 1 in the main text, we can then estimate the variance of $\hat{p} = x^*/N = x^*/(x^* + x^\dagger)$ by expanding to first order:

$$\delta\hat{p} \approx g_{\hat{p},x^*} \delta x^* + g_{\hat{p},x^\dagger} \delta x^\dagger, \quad [\text{S51}]$$

where the gains are the following:

$$g_{\hat{p},x^*} = \frac{d\hat{p}}{dx^*} = \frac{x^*}{(x^* + x^\dagger)^2}, \quad [\text{S52}]$$

$$g_{\hat{p},x^\dagger} = \frac{d\hat{p}}{dx^\dagger} = -\frac{x^\dagger}{(x^* + x^\dagger)^2}. \quad [\text{S53}]$$

The variance is then the following:

$$\sigma_{\hat{p}}^2 = g_{\hat{p},x^*}^2 \sigma_{x^*}^2 + g_{\hat{p},x^\dagger}^2 \sigma_{x^\dagger}^2 + 2g_{\hat{p},x^*} g_{\hat{p},x^\dagger} \sigma_{x^*,x^\dagger}, \quad [\text{S54}]$$

where the last term accounts for the covariance. The variances can be calculated in many ways because the system is linear. For example, they can be calculated exactly via the linear-noise approximation. The result is the first term of Eq. 2 in the main text (Eq. S18), as claimed. Indeed, there is no second term for the model described here. This is precisely because with this scheme the number of samples N is known. Although in the scheme of the main text (Fig. 2), the system cannot discriminate between the molecules that have collided with an unbound receptor and the molecules that have not collided with the receptor at all, in this scheme the system knows exactly how many collisions there have been with the receptor: $x^* + x^\dagger$.

- Milo R, Jorgensen P, Moran U, Weber G, Springer M (2010) BioNumbers—the database of key numbers in molecular and cell biology. *Nucleic Acids Res* 38(Database issue): D750–D753.
- Sourjik V, Berg HC (2002) Binding of the *Escherichia coli* response regulator CheY to its target measured in vivo by fluorescence resonance energy transfer. *Proc Natl Acad Sci USA* 99(20):12669–12674.
- Gardiner CW (1985) *Handbook of Stochastic Methods* (Springer, Berlin).
- Ziv E, Nemenman I, Wiggins CH (2007) Effect of feedback on the fidelity of information transmission of time-varying signals. *PLoS One* 2(10):1077.
- Berg HC, Purcell EM (1977) Physics of chemoreception. *Biophys J* 20(2):193–219.
- Bialek W, Setayeshgar S (2005) Physical limits to biochemical signaling. *Proc Natl Acad Sci USA* 102(29):10040–10045.
- Kaizu K, et al. (2014) The Berg-Purcell limit revisited. *Biophys J* 106(4):976–985.
- Hill T (1977) *Free Energy Transduction in Biology: The Steady-State Kinetic and Thermodynamic Formalism* (Academic Press, New York).
- Govern CC, ten Wolde PR (2012) Fundamental limits on sensing chemical concentrations with linear biochemical networks. *Phys Rev Lett* 109(21):218103.

Table S1. Molecular weights in amino acids are from ref. 1

Protein	Molecular weight, aa	Copy number
Tsr/Tar/Trg	~550	3,600–15,000
CheW	167	2,000–6,700
CheY	129	1,400–8,200
CheZ	214	560–3,200
CheA	654	1,100–6,700

Copy numbers are from ref. 2.

- UniProt Consortium (2014) Activities at the universal protein resource (UniProt). *Nucleic Acids Res* 42(Database issue):D191–D198.
- Li M, Hazelbauer GL (2004) Cellular stoichiometry of the components of the chemotaxis signaling complex. *J Bacteriol* 186(12):3687–3694.