Communication distance in the intercellular signaling by cyto/chemokine or autacoid

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Abstract: Characteristic properties of signaling processes are studied by a single cell model in the intercellular transport. The diffusion equation is solved for molecules secreted from a spherical cell under the boundary conditions of prescribed fluxes at the cell surface. The solution is physicochemically compared with another one corresponding to constant source at the cell surface by Francis and Palsson.¹⁾ Discussion is given on communication distances ($\sim 50 \mu\text{m}$) estimated for human cytokines.

Key words: Intercellular signaling; secretion; diffusion equation; flux; communication distance.

Introduction. Physicochemical transport processes of cyto- and chemokines are important for signal transduction.¹⁾⁻⁴⁾ Francis and Palsson¹⁾ have adopted a solitary cell model to study fundamentals of cellular signaling processes in the intercellular communication. In the model a cell is assumed to secrete soluble chemical substances, which diffuse in the surrounding medium. The signal is transferred by its binding to cell's receptors. They have found that the process is governed by a dimensionless key parameter, α , which represents the ratio of a physicochemical time constant and a biological time constant. They have estimated an effective communication distance and the characteristic time associated with it as about $250 \mu\text{m}$ and 10-30 minutes, respectively, for human cytokines.

Let us consider a secreting spherical cell of radius a with its center fixed at the origin. Chemical substances (molecules) released from the cell surface make a random walk in the surrounding medium.⁵⁾ The concentration of molecules $c(r, t)$ is described by the diffusion equation^{6),7)} in the spherically symmetric case as

$$\left(\frac{D}{r^2} \frac{\partial}{\partial r} r^2 \frac{\partial}{\partial r} - \frac{\partial}{\partial t} \right) c(r, t) = 0 \quad [1]$$

with D being the diffusion coefficient of molecules in the surrounding medium, r the distance from the origin. At time $t = 0$ the cell starts to release molecules with a constant rate j_0 , which is the number of molecules

uniformly secreted at the cell surface ($r = a$) per unit time per unit area. The flux or current associated with $c(r, t)$ is given by

$$j(r, t) = -D \frac{\partial c}{\partial r}. \quad [2]$$

Initially there are no molecules and the concentration is completely diluted very far from the cell. These amount to the following boundary conditions

$$\begin{aligned} c(r, 0) &= 0 \text{ for } r > a, \\ \text{and } c(\infty, t) &= 0 \text{ and } j(a, t) = j_0 \text{ for } t > 0. \end{aligned} \quad [3]$$

The discussion of Francis and Palsson¹⁾ is based on the above equations, but they have used a different solution to introduce the concept of communication distances and characteristic times; actually their function satisfies eq. [1], but it corresponds to the boundary condition of constant source at the cell surface. The confusion comes from the difference in the meanings of 'source' in the secretion of a cell and 'source' in the heat diffusion in solids.⁶⁾

In this paper we investigate characteristic properties of the physicochemical intercellular signaling on the basis of a single cell model. We first show the appropriate solution of eq. [1] with eq. [3], and its generalization corresponding to the boundary condition of a prescribed time-dependent flux at the cell surface. We then make clear physicochemical meanings of the boundary conditions in the secretion of molecules, by discussing differences between our results and those of ref. 1. It is found by the numerical evaluation for human cytokines

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that the value of α mentioned above is found to be smaller by one order of magnitude than their value, and the same tendency for the communication distance as a consequence.

Theoretical analysis. We show the solution for eq. [1] combined with the boundary condition given by eq. [3]. For this purpose it is useful to consider $f(r, t) \equiv rc(r, t)$, which satisfies the one-dimensional diffusion equation

$$\left(D \frac{\partial^2}{\partial r^2} - \frac{\partial}{\partial t}\right) f(r, t) = 0 \quad [4]$$

in the semi-infinite region. It is linear and thus the function $g = g(r, t)$ defined by

$$g = -D \frac{\partial f}{\partial r} + \left(\frac{D}{a}\right) f \quad [5]$$

also satisfies the same diffusion equation of eq. [4]. On the other hand $g(r, t)$ can be expressed in terms of $c(r, t)$ as $g = -Dr \partial c / \partial r - D \{1 - (r/a)\}c$. Then, from eq. [4] the boundary conditions for $g(r, t)$ are

$$\begin{aligned} g(r, 0) &= 0 \text{ for } r > a, \\ \text{and } g(a, t) &= aj_0 \text{ for } t > 0. \end{aligned} \quad [6]$$

The solution of eq. [4] for $g(r, t)$ under the above boundary conditions is straightforward⁶⁾

$$\begin{aligned} g(r, t) &= aj_0 \operatorname{erfc} \left\{ \frac{(r-a)}{2\sqrt{Dt}} \right\} \\ &= aj_0 \left[1 + \frac{1}{\pi} \int_{-\infty}^{\infty} dk \left(\frac{i}{k} \right) \exp \left\{ ik(r-a) - Dt k^2 \right\} \right] \end{aligned} \quad [7]$$

for $r \geq a$, where $\operatorname{erfc}(x) \equiv \frac{2}{\sqrt{\pi}} \int_x^{\infty} dt \exp(-t^2)$ is the complementary error function.⁸⁾ Since the k -integral of $p(k) \exp(ikr - Dt k^2)$ for arbitrary $p(k)$ satisfies eq. [3], the second equality shows that eq. [7] is its particular choice in the Fourier transformation.⁹⁾ Substituting eq. [7] into eq. [4], we find the equation for $f(r, t)$ as

$$-D \frac{\partial f}{\partial r} + \frac{D}{a} f = aj_0 \operatorname{erfc} \left\{ \frac{(r-a)}{2\sqrt{Dt}} \right\}. \quad [8]$$

It is solved as

$$f(r, t) = \frac{aj_0}{D} \int_r^{\infty} dr' \exp \left\{ \frac{(r-r')}{a} \right\} \operatorname{erfc} \left\{ \frac{(r'-a)}{2\sqrt{Dt}} \right\} \quad [9]$$

by which $c(r, t) = f(r, t)/r$ is explicitly written as

$$\begin{aligned} c(r, t) &= \frac{aj_0}{Dr} \left[\operatorname{erfc} \left\{ \frac{(r-a)}{2\sqrt{Dt}} \right\} \right. \\ &\quad \left. - \exp \left(\frac{r}{a} - 1 + \frac{Dt}{a^2} \right) \operatorname{erfc} \left\{ \frac{(r-a)}{2\sqrt{Dt}} + \frac{\sqrt{Dt}}{a} \right\} \right] \end{aligned} \quad [10]$$

after some manipulations. The asymptotic behavior of $c(r, t)$ is $c(r, \infty) = aj_0/(Dr)$ and $c(\infty, t) = 0$. The former means that in the large t limit $c(r, t)$ converges to the stationary solution of eq. [1], which is inversely proportional to the distance r .

The above solution is easily extended to more general cases of a time-dependent flux at the cell surface as the boundary condition. We consider a prescribed time-dependent flux $p(t)$ at $r = a$

$$p(t) = -D \frac{\partial c}{\partial r} \text{ at } r = a \text{ for } t > 0 \quad [11]$$

instead of $j(r, t) = j_0$ in eq. [3]. It is equivalent to the boundary condition for $g(r, t)$ given by eq. [5],

$$\begin{aligned} g(r, 0) &= 0 \text{ for } r > a, \\ \text{and } g(a, t) &= ap(t) \text{ for } t > 0. \end{aligned} \quad [12]$$

The solution of eq. [4], satisfying these conditions, are given by using $q(x, t) \equiv \operatorname{erfc} \{ x / (2\sqrt{Dt}) \}$ as

$$\begin{aligned} g(r, t) &= a \int_0^t ds \frac{dp(s)}{ds} q(r-a, t-s) \\ &\quad + ap(0) q(r-a, t) \\ &= a \int_0^t ds p(s) \frac{\partial q(r-a, t-s)}{\partial t}. \end{aligned} \quad [13]$$

Following the same procedures as taken from eq. [7] to eq. [10], $c(r, t)$ is obtained as

$$\begin{aligned} c(r, t) &= \frac{a}{Dr} \int_r^{\infty} dr' \exp \left\{ \frac{(r-r')}{a} \right\} \\ &\quad \times \int_0^t ds p(s) \frac{\partial q(r'-a, t-s)}{\partial t}. \end{aligned} \quad [14]$$

Physicochemical considerations. We compare our result $c(r, t)$ given by eq. [10] with the function adopted by Francis and Palsson¹⁾

$$c_s(r, t) = \frac{S_0}{ar} \sqrt{\frac{t}{\pi D}} \left[\exp \left\{ -\frac{(r-a)^2}{4Dt} \right\} - \exp \left\{ -\frac{(r+a)^2}{4Dt} \right\} \right] - \sqrt{\frac{\pi}{4Dt}} |r-a| \operatorname{erfc} \left\{ \frac{|r-a|}{2\sqrt{Dt}} \right\} + \sqrt{\frac{\pi}{4Dt}} (r+a) \operatorname{erfc} \left\{ \frac{(r+a)}{2\sqrt{Dt}} \right\} \quad [15]$$

and discuss the difference in the physicochemical content. For convenience, the quantities ρ and F_0 in eq. [2] of ref. 1 are here expressed by a and $S_0 = F_0 a^2$, respectively.

It is easy to see that $rc_s(r, t)$ satisfies the diffusion equation given by eq. [4]. It assumes the existence of constant source with an intensity of S_0 at the cell surface, which is the number of molecules released per unit time divided by 4π . This fact may be understandable, for instance, from the equation satisfied by $c_s(r, t)$

$$\int_0^\infty dr r^2 \frac{\partial c_s}{\partial t} = S_0 \quad [16a]$$

which is directly examined by using eq. [15]. It is in contrast to the similar one

$$\int_a^\infty dr r^2 \frac{\partial c}{\partial t} = a^2 j_0 \quad [16b]$$

for $c(r, t)$ given by eq. [10] in the present case. Equation [16a] means that the secreting rate of molecules in the whole region is $4\pi S_0$, but eq. [16b] shows that the number of molecules in the outside region of the sphere increases at the constant rate of $4\pi a^2 j_0$. These two solutions would effectively look similar if $S_0 = a^2 j_0$. The difference is, however that $c_s(r, t)$ is meaningful in the whole semi-infinite region, but $c(r, t)$ is only for $r \geq a$ since the property of the secreting cell is solely represented by the boundary condition at the cell surface. Whereas the flux j_0 plays a role of source to the outside region (or surrounding medium), the source S_0 gives effects both to the inside region of the cell and the surrounding medium. Since the cell releases molecules only toward the outside region, it is clear that eq. [10] is physicochemically appropriate.

These two functions of eqs. [10] and [15] are different in the dependence on r and t . For example, Fig. 1 shows their behaviours near $r = a$ in the steady state ($t = \infty$). In the region of $r \leq a$, $c_s(r, \infty)$ remains constant, but $c(r, \infty)$ decreases as $1/r$; of course eq. [10] is plotted as a function of r only for guidance and the curve in this region has no physicochemical meanings. On the other

Fig. 1. The r -dependence of $c(r, t)$ given by eq. [10] and $c_s(r, t)$ given by eq. [15] with $S_0 = a^2 j_0$ in the steady state ($t = \infty$). In the horizontal axis $r = a$ shows the position of the cell surface.

hand they coincide for $r \geq a$. The derivative of $c_s(r, \infty)$ with respect to r is discontinuous at $r = a$ because of the existence of the source on the surface ($r = a$). It is worthwhile in this connection to note the asymptotic behaviour of the dimensionless (reduced) quantity $\varphi(\xi, \tau) \equiv \{D/(a j_0)\} c(r, t)$ in the region of large ξ and τ , where $\xi \equiv r/a$ and $\tau \equiv 4Dt/a^2$ are reduced variables of length and time. It is useful for this to note that $\varphi(\xi, \tau)$ corresponding to eq. [10] can be rewritten as

$$\varphi(\xi, \tau) = \frac{1}{\xi} \left\{ \operatorname{erfc}(x) - \exp(y^2 - x^2) \operatorname{erfc}(y) \right\} \quad [17]$$

with $x = (\xi - 1)/\sqrt{\tau}$ and $y = \{\xi - 1 + (\tau/2)\}/\sqrt{\tau}$. Making use of the asymptotic behaviour of the complementary error function as $\operatorname{erfc}(x) \approx (1/\sqrt{\pi}) \exp(-x^2)/x$ in the large x region,⁸⁾ we find

$$\varphi(\xi, \tau) \approx \frac{1}{2\xi\sqrt{\pi}} \exp\left(-\frac{\xi^2}{\tau}\right) \frac{\tau\sqrt{\tau}}{\xi\left(\xi + \frac{\tau}{2}\right)} \quad [18]$$

for large x . Thus, when $\xi \ll \tau$, $\varphi(\xi, \tau)$ agrees with the asymptotic behaviour of the reduced quantity $\varphi_s(\xi, \tau) \equiv \{D/(a j_0)\} c_s(r, t)$, but it is different from $\varphi_s(\xi, \tau)$ by the factor $\tau/2\xi$ for $\xi \gg \tau$.

Numerical results. It is necessary for the signal from the secreting cell to be received that $c(r, t)$ increases to the level of a critical K value for its receptor, where K represents the dissociation constant of the signaling ligand for the receptor. If $c(r, t)$ is less than K after an elapse of infinite time at some distance $r = r'$, the signal can not be effectively transferred to the dis-

tance r' . The concept of the communication distance, r_c , represents a measure of such distance associated with this process. Francis and Palsson¹⁾ have introduced it or its reduced quantity $\xi_c = r_c/a$ such that r_c is just half as large as r' for which $\varphi_s(r', \infty) = K$. Since $\varphi_s(r, t) = (\sigma F_0/D) \varphi_s(\xi, \tau)$ in their notation and $\varphi_s(\xi, \infty) = 1/\xi$, this means that $\varphi_s(\xi_c, \infty) = 2/\alpha$, or $\xi_c = \alpha/2$ with $\alpha = aF_0/(DK)$. They have also shown the existence of a simplified relation $\tau_c = (a_f \xi_c)^2$ for a reduced characteristic time constant τ_c and ξ_c because $\varphi_s(\xi, \tau)/\varphi_s(\xi, \infty)$ is the scaled function of ξ^2/τ for large ξ and τ .

In the present case the same definition is adoptable for ξ_c since $\varphi(\xi, \infty) = \varphi_s(\xi, \infty)$, but τ_c might be defined alternatively since the above mentioned scaling doesn't hold. It is not impossible to introduce these concepts differently: One way would be to use moments, *i.e.*, $\langle \xi^2 \rangle = \int d\xi \varphi(\xi, \tau) \xi^2 / \int d\xi \varphi(\xi, \tau)$, as $\varphi(\xi, \tau)$ has a meaning of probability. The reduced average distance ξ_{av} is calculated as $\xi_{av} = \sqrt{\langle \xi^2 \rangle}$ as a function of τ . The quantity τ_c is defined from $\alpha = \xi_{av}(\tau)$, and then ξ_c by $\varphi(\xi_c, \tau_c) = 1/\alpha$. It seems that the difference may not be so significant when ξ and τ are large. Quantitative discussion on this point and the appropriateness about definitions are, however, out of our concern in this paper.

We now focus on ξ_c and make some remarks on realistic estimations. First, we point out the inconsistency for F_0 in Francis and Palsson¹⁾: F_0 in their eq. [2] is the number of secreted molecules per unit time divided by 4π , but F_0 in the equation $-D\partial c/\partial r = F_0$ below their eq. [1] has the meaning of its number per unit time and unit area. If their eq. [2] or $c_s(r, t)$ is adopted from the beginning, $-D\partial c/\partial r$ should be equal to $F_0/(2\rho^2)$ at $r = \rho$ in their notations. It has not been clearly shown what numerical values are used for these quantities in the calculations.

For human cytokines the secretion rate (F_0) and the critical concentration of molecules (K) are experimentally about 2300–8000 molecules/cell/s by Savinell *et al.*,¹⁰⁾ 10–50 pM by Aggarwal and Gutterman,¹¹⁾ respectively. We adopt the same definition for ξ_c as Francis and Palsson,¹⁾ for a direct comparison of consequences resulting from $c(r, t)$ and $c_s(r, t)$, or $\xi_c = \alpha/2$ with $\alpha \equiv aj_0/(DK)$ in our notation. We have calculated the value of α based on the data given above; taking $D = 5 \times 10^{-6}$ cm²/s, $a = 10$ μ m, $F_0 = 3000$ molecules/cell/s and $K = 10$ pM as representative input values, we have found that $j_0 = F_0/(4\pi a^2) = 2.4 \times 10^{12}$ molecules/m²/s, and thus $\alpha = aj_0/(DK) = 100/4\pi \cong 10$. It is actually smaller by one order of magnitude than the estimated value of $\alpha = 100$ of ref. 1, and as a consequence the communication dis-

tance $\xi_c = \alpha/2$ turns out smaller by one order of magnitude than their value. It seems to us that the difference probably comes from the factor of 4π mentioned above. There is some uncertainty in the value of α depending on input values, but we could say that there is a systematic overestimation in their results.

Discussion. We have adopted the single cell model to study characteristic properties in the intercellular signaling process. The solution to the spherical diffusion equation for secreted molecules is obtained as eq. [10] under the boundary condition of constant flux at the cell surface. We have discussed the physicochemical difference of our present result given by eq. [10] from eq. [15] of Francis and Palsson,¹⁾ and shown our result as the appropriate solution corresponding to constant secretion rate in the intercellular signaling. It is found numerically for human cytokines that the parameter α , and thus the communication distance ξ_c are significantly smaller than their estimations.

In the single cell model, chemical substances released solely make a random walk with no interaction between them. Moreover, it is tacitly assumed that secreting cells themselves are independent of each other. These are apparent in the expression of the communication distance $\xi_c = aj_0/(2DK)$; a and j_0 represent properties of a secreting cell, K the biochemical property of receptors, and D a single-particle property of molecules. If cooperative properties have any significance in biological functions, the concept of the communication distance should include factors reflecting cooperative or feedback effects. The above expression is then regarded as its non-interaction limit. Francis and Palsson¹⁾ have argued that their estimated ξ value of about 250 μ m is realistic. Our result of $\xi = \alpha/2 \cong 5$ or $r_c \cong 50$ μ m is much smaller than their value. It seems to us that this rather implies a non-negligible contribution from correlational effects or others.

We have shown the solution of eq. [14] corresponding to the situation that the flux has a prescribed time-dependence at the cell surface. It is well known that for homeostasis the secretion rate is changing with time. A time-dependent model appropriately taking into account this feature will give us more detailed information on signaling processes occurring in realistic situations. It will be reported elsewhere in the future.

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