J. Math. Biol. (1991) 29: 389–404

Journal of
Mathematical
Biology

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Mathematical analysis of a basic model for epidermal wound healing

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Received February 21, 1990; received in revised form July 26, 1990

Abstract. The stimuli for the increase in epidermal mitosis during wound healing are not fully known. We construct a mathematical model which suggests that biochemical regulation of mitosis is fundamental to the process, and that a single chemical with a simple regulatory effect can account for the healing of circular epidermal wounds. The numerical results of the model compare well with experimental data. We investigate the model analytically by making biologically relevant approximations. We then obtain travelling wave solutions which provide information about the accuracy of these approximations and clarify the roles of the various model parameters.

Key words: Wound healing – Mathematical models – Travelling waves

1. Biological background

Mammalian skin in composed of two parts. The outer part is the epidermis, which consists of several layers of cells, with the bottom layer undergoing frequent mitosis to renew the layers above. As cells are displaced upwards, keratin (a tough, fibrous protein) accumulates in their interior, gradually replacing cytoplasm until the cells die. Such dead "keratinised" cells constitute the external surface of the skin, and are gradually sloughed. Beneath the epidermis is the dermis, which contains, in addition to several cell types, blood vessels, nerves and protein fibres, all of which are absent from the epidermis.

When the epidermis is injured, healing occurs by "epidermal migration", in which epidermal cells spread across the wound. This also occurs in deeper wounds, at the same time as much more complicated healing processes in the dermis. Thus epidermal wounds provide a relatively simple context in which to study epidermal migration.

The mechanism of epidermal migration is only partially understood. Normal epidermal cells are non-motile. However, in the neighbourhood of the wound they undergo marked phenotype alteration ("mobilisation") that gives the cells the ability to move via lamellipodia (Clark 1989). The main factor controlling cell movement seems to be contact inhibition (Irvin 1984; Bereiter-Hahn 1986). However, chemotaxis and contact guidance may also be involved (Clark 1988).

Remnants of glands and hair follicles can act as sources of migrating cells (Rudolph 1980) but we consider only wounds in which all such appendages are removed at injury. Extension of the model would be possible by regarding the wound as a series of micro-wounds extending from one appendage to the next, a representation proposed by Winter (1972).

Two mechanisms have been proposed for the movement of the cell sheet. In the 'rolling mechanism', the leading cells are successively implanted as new basal cells, and other cells roll over these (Krawczyk 1971; Winstanley 1975; Ortonne et al. 1981). In the 'sliding mechanism', on the other hand, the cells in the interior of the sheet respond passively to the pull of the marginal cells. However, all of the migrating cells have the potential to be motile: for example, if a gap opens up in the migrating sheet, cells at the boundary of this develop lamellipodia and move inwards to close the gap (Trinkaus 1984). Though the morphological data of mammalian epidermal wound healing is convincingly explained by the rolling mechanism (Stenn and DePalma 1988), unequivocal evidence is lacking, whereas the sliding mechanism is well documented in simpler systems such as amphibian epidermal wound closure (Radice 1980).

Soon after the onset of epidermal migration, mitotic activity increases in a band (about 1 mm thick) of the new epidermis near the wound edge, providing an additional population of cells (Bereiter-Hahn 1986). The greatest mitotic activity is actually at the wound edge, where it can be as much as 15 times the rate in normal epidermis (Winter 1972); activity decreases rapidly across the band, going away from the wound. The stimulus for this increase in mitotic activity is uncertain. Two factors that are certainly involved are the absence of contact inhibition, which applies to mitosis as well as to cell motion (Clark 1988), and change in cell shape: as the cells spread out they become flatter, which tends to increase their rate of division (Folkman and Moscona 1978). There is also experimental evidence, which we now briefly review, for production by epidermal cells both of chemicals that inhibit mitosis and of chemicals that stimulate it.

The former are "chalones", a generic term for inhibitors of cell proliferation that are produced by the cell types on which they act. Although the term itself has acquired a somewhat bad reputation (Iversen 1985), the evidence for such inhibitory growth regulators is now considerable. There are two established epidermal chalones, which act at different points in the cell cycle. Their chemical properties are summarized in Fremuth (1984, pp. 37–38). Experimental work to investigate dose-response relationships has shown a general increase in inhibitory effect with dosage (e.g. Hondius-Boldingh and Laurence 1968; Iversen 1978; Marks 1973) although beyond this it is inconclusive, which Iversen (1981) attributes to the fact that one has to use skin extracts since the chalones are not yet available in pure form. There are few direct experimental studies of the role of chalones in wound healing, although Yamaguchi et al. (1974) investigated the variation of proliferation rate with time near the edge of wounds in mice, concluding that inhibition occurs at three distinct points in the cell cycle.

Turning to epidermal growth activators produced by epidermal cells themselves, evidence for these is provided in recent work by Eisinger et al. (1988a, an in vivo study; 1988b, an in vitro study). In the in vivo study, an extract derived from epidermal cell cultures was found to increase the rate of epidermal migration when applied, on a dressing, to wounds in pigs. In the in vitro study, the same extract was found to increase the growth rate of cultures of epidermal cells. It is unclear whether the chemical(s) causing activation of mitosis in these studies are growth factors that have already been characterised or 'new' growth factors.

There is also recent evidence that the production by epithelial cells of the chemical SPARC (secreted protein acidic and rich in cysteine) increases following wounding. Since SPARC affects the proliferation rate of cells, this is another possible activator (Mason et al. 1986; Engel et al. 1987; Sage et al. 1989).

2. Derivation of the model

Two simple models were proposed by Sherratt and Murray (1990) who, based on comparison with experiment, concluded that the following was the more biologically realistic. The model consists of two conservation equations, one for the cell density per unit area (n) and one for the concentration (c) of the mitosis-regulating chemical. We consider two cases, one in which the chemical activates mitosis and the other in which it inhibits it. The epidermis is sufficiently thin that we consider the wound to be two dimensional. This is a reasonable assumption since we consider wounds whose linear dimensions are O(cms) while the thickness of the epidermis is $O(10^{-2}\,\text{cm})$ (Odland 1983). The general form of the governing equations is:

Rate of increase of cell density	= Cell migration	+ Mitotic generation	– Natural loss	
Rate of increase of chemical concentration	= Diffusion	+ Production by cells	– Decay of active chemical.	

We use a diffusion term to model contact inhibition controlled cell migration. Following the representations of short-range cellular diffusion in models discussed by Murray (1989), this diffusion term has a constant diffusion coefficient, independent of n. Sherratt and Murray (1990) show that a density dependent diffusion term, in the absence of biochemical control, is unable to capture crucial aspects of the healing process. We consider the mathematical representation of each reaction term in turn.

Time decay of active chemical. Such decay is typically governed by first order kinetics, so we model this term by $-\lambda c$, where λ is a positive rate constant. In the absence of the other terms on the right hand side of the equation this gives an exponential decay with time.

Production of chemical by the cells. This is a function of n, which must equal zero when $n = 0$ (when there are no cells, nothing can be produced by them) and λc_0 when $n = n_0$, so that the unwounded state is a steady state. Here n_0 and c_0 are the unwounded cell density and chemical concentration: we assume a non-zero concentration of chemical in the unwounded state. Further, the chemical production function, $f(n)$ say, must reflect an appropriate cellular response to injury depending on whether the chemical activates or inhibits mitosis. The qualitative form of $f(n)$ in the two cases is as shown in Fig. 1. We take simple functional forms that conform to these requirements, namely:

$$f(n) = \lambda c_0 \cdot \frac{n}{n_0} \cdot \left(\frac{n_0^2 + \alpha^2}{n^2 + \alpha^2}\right) \quad \text{for the activator}$$

$$f(n) = \frac{\lambda c_0}{n_0} \cdot n \quad \text{for the inhibitor,}$$

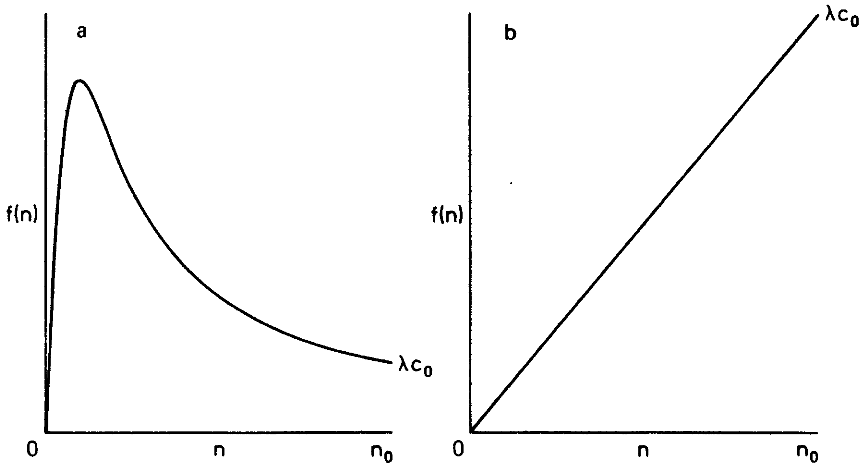

Fig. 1. The qualitative form of the function $f(n)$, which reflects the rate of chemical production by epidermal cells: **a** activator; **b** inhibitor

where α is a positive constant which relates to the maximum rate of chemical production.

Rate of natural cell loss. This is due to the sloughing of the outermost layer of epidermal cells, and we take it as proportional to n, say kn.

Chemically controlled cell division. We choose this term so that when $c = c_0$, the unwounded concentration, the sum of this term and the previous one is of logistic growth form, $kn(1 - n/n_0)$. This is a commonly used metaphor for simple growth in population biology models; k is the linear mitotic rate. Thus we model this term with $s(c) \cdot n \cdot (2 - n/n_0)$, where $s(c)$ reflects the chemical control of mitosis, and $s(c_0) = k$; $s(c)$ has the qualitative form shown in Fig. 2. In the case

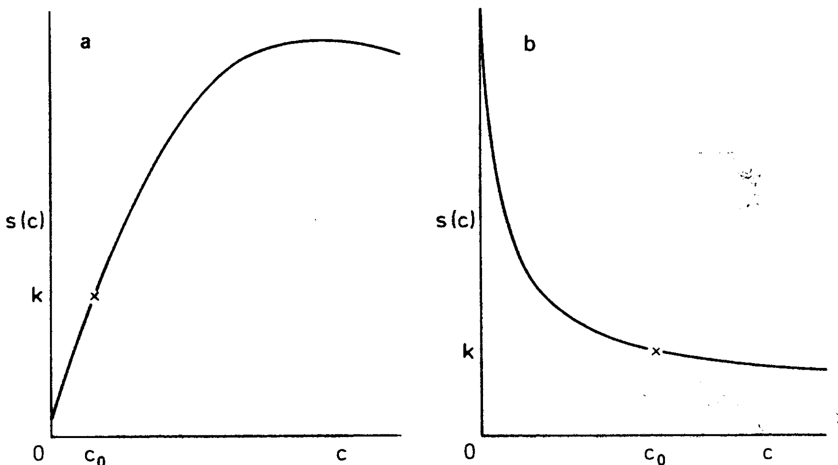

Fig. 2. The qualitative form of the function $s(c)$, which reflects the chemical control of mitosis: **a** activator; **b** inhibitor. Here c_0 represents the steady state chemical concentration in the unwounded state, and k is a parameter equal to the reciprocal of the cell cycle time

of a chemical activator, a decrease of $s(c)$ to $s(0)$ for large c is included because it is found experimentally (Eisinger et al. 1988a); we will show that it has little effect on the solution of the model equations. In both cases we require $0 < s_\infty < s_{\max} = hk$, say, where h is a constant, and we take $s_\infty = k/2$. Again we take simple functional forms satisfying these criteria, namely:

$$s(c) = k \cdot \left\{ \frac{2c_m(h-\beta)c}{c_m^2 + c^2} + \beta \right\} \quad \text{where } \beta = \frac{c_0^2 + c_m^2 - 2hc_0c_m}{(c_0 - c_m)^2}$$

for the activator, where c_m ($> c_0$) is a constant parameter which relates to the maximum level of chemical activation of mitosis, and

$$s(c) = \frac{(h-1)c + hc_0}{2(h-1)c + c_0} \cdot k \quad \text{for the inhibitor.}$$

Thus the model system is

$$\frac{\partial n}{\partial t} = D\nabla^2 n + s(c) \cdot n \cdot \left(2 - \frac{n}{n_0}\right) - kn,$$

$$\frac{\partial c}{\partial t} = D_c \nabla^2 c + f(n) - \lambda c,$$

with initial conditions

$$n = c = 0 \quad \text{at } t = 0 \quad \text{inside the wound domain,}$$

and boundary conditions

$$n = n_0 \quad \text{and} \quad c = c_0 \quad \text{on the wound boundary, for all } t.$$

There is a debate in the biological literature as to whether mitosis drives cell migration or vice versa (see, for example, Potten et al. 1984; Wright and Alison 1984, chap. 14). The biologically reasonable results given by our model and discussed below are based on the assumption that, in fact, both processes are dependent on the local cell density.

3. Nondimensional form and linear analysis

To clarify the roles of the various parameters, we nondimensionalise the model using a length scale L (a typical linear dimension of the wound) and time scale $1/k$ (the cell cycle time seems the most relevant time scale). We define the following dimensionless quantities (denoted by *):

$$n^* = n/n_0, \quad c^* = c/c_0, \quad \vec{r}^* = \vec{r}/L, \quad t^* = kt, \quad D^* = D/(kL^2)$$

$$D_c^* = D_c/(kL^2), \quad \lambda^* = \lambda/k, \quad c_m^* = c_m/c_0, \quad \alpha^* = \alpha/n_0.$$

With these definitions, the dimensionless model equations are (dropping the

asterisks for notational simplicity):

$$\frac{\partial n}{\partial t} = D\nabla^2 n + s(c)\cdot n\cdot(2-n) - n$$
$$\frac{\partial c}{\partial t} = D_c\nabla^2 c + \lambda g(n) - \lambda c, \qquad (1)$$

with initial conditions

$$n = c = 0 \quad \text{at } t = 0 \quad \text{inside the wound domain,}$$

and boundary conditions

$$n = 1 \quad \text{and} \quad c = 1 \quad \text{on the wound boundary, for all } t.$$

Here, for the activator

$$g(n) = \frac{n(1+\alpha^2)}{n^2+\alpha^2}, \qquad s(c) = \frac{2c_m(h-\beta)c}{c_m^2+c^2} + \beta \quad \text{where } \beta = \frac{1+c_m^2-2hc_m}{(1-c_m)^2},$$

and for the inhibitor

$$g(n) = n, \qquad s(c) = \frac{(h-1)c+h}{2(h-1)c+1},$$

where we assume $h > 1$ and $c_m > 1$.

We require the unwounded state to be stable to small perturbations, while the wounded state is unstable. Straightforward linear analysis shows that these conditions are satisfied provided $s(0) > 1/2$. For the activator, this condition is $c_m > (2h-1) + \sqrt{(2h-1)^2 - 1}$; for the inhibitor, it is simply $h > 1/2$.

4. Parameter values

It is possible to estimate the parameters λ and k from experimental data. We estimate λ in the case of a chemical inhibitor using data on chalones. Brugal and Pelmont (1975) found a decrease in the proliferation rate in intestinal epithelium during the 12 h after injection with epithelium extract. Also Hennings et al. (1969) were able to maintain suppression of epidermal DNA synthesis by repeated injection of epidermal extract at 12 h intervals. Based on these studies, we take the half-life of chemical decay as 12 h. If we consider only the decay term in the second equation this gives exponential decay with a half-life of $\lambda^{-1}\log 2$. We thus take $\lambda = 0.05$ ($\approx \frac{1}{12}\log 2$) h^{-1}.

In the case of a chemical activator, there is little quantitative experimental data. However, comparison of the work of Eisinger et al. (1988a,b) on chemical activators in wound healing and the clinical studies of chalone effects by Rytömaa and Kiviniemi (1969, 1970) suggests a longer time scale for the chalone activity, by a factor of about 6, so we take $\lambda = 0.3\ h^{-1}$ for the activator.

The parameter k is simply the reciprocal of the epidermal cell cycle time. This varies from species to species, but is typically about 100 h (Wright 1983), so we take $k = 0.01\ h^{-1}$. The diffusion coefficients D and D_c were estimated by Sherratt and Murray (1990) based on a best fit analysis with data on wound healing, since there is at present no direct experimental data from which they can be determined. This gave values $D = 3.5\times10^{-10}\ cm^2\,s^{-1}$, $D_c = 3.1\times10^{-7}\ cm^2\,s^{-1}$ for the activator, and $D = 6.9\times10^{-11}\ cm^2\,s^{-1}$, $D_c = 5.9\times10^{-6}\ cm^2\,s^{-1}$ for the inhibitor. These are not biologically unreasonable for cells and biochemicals of relatively low molecular weight, respectively.

5. Numerical solution of the model

We solved (1) numerically in a radially symmetric geometry using the method of lines, with a uniform space mesh of 201 points. The numerical solutions were compared with data from Van den Brenk (1956), one of the more careful experimental studies of such epidermal wounds. In this study the full thickness of epidermis was removed from a circular region, 1 cm in diameter, in the ears of rabbits. In particular, care was taken not to leave behind any hair follicles so that our model with no 'internal' sources of epidermal cells is appropriate. The change in wound radius with time was recorded. To capture the concept of 'wound radius' from our model, we take the wound as 'healed' when the cell density reaches 80% of its unwounded value, that is when $n = 0.8$ for the nondimensional equations. The choice of this level as 80% is somewhat arbitrary, but does not significantly affect the results since the solutions for n and c have travelling wave form, as discussed below.

Figs. 3 and 4 show the numerically calculated decrease in wound radius with time compared with the data, and plots of n and c against r at a selection of equally spaced times. As well as good overall agreement with the data, the numerical solutions exhibit the two phases (a lag phase and then a linear phase) that characterise epidermal wound healing (e.g. Snowden 1984). The (constant) speed of the linear phase can be approximately calculated visually from the graph of n against r. For a wound radius of 0.5 cm, this gives dimensional wave speeds of 2.6×10^{-3} mm h^{-1} for the activator and 1.2×10^{-3} mm h^{-1} for the inhibitor. These compare with the speed 8.6×10^{-3} mm h^{-1} found in Van den Brenk's (1956) study.

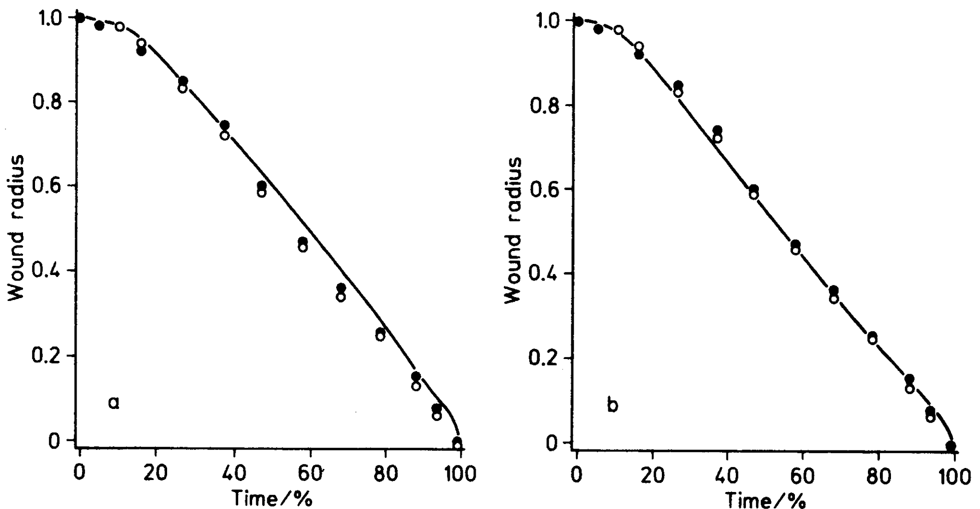

Fig. 3a,b. Numerically calculated decrease in wound radius with time from Eqs. (1) as compared to the data, denoted by ● and ○ (Van den Brenk 1956). Time is expressed as a percentage of total healing time, since this is how Van den Brenk's data is presented. **a** Biochemical activation of mitosis with parameter values $D = 5 \times 10^{-4}$, $D_c = 0.45$, $\lambda = 30$, $h = 10$, $\alpha = 0.1$, $c_m = 40$; **b** biochemical inhibition of mitosis with parameter values $D = 10^{-4}$, $D_c = 0.85$, $\lambda = 5$, $h = 10$

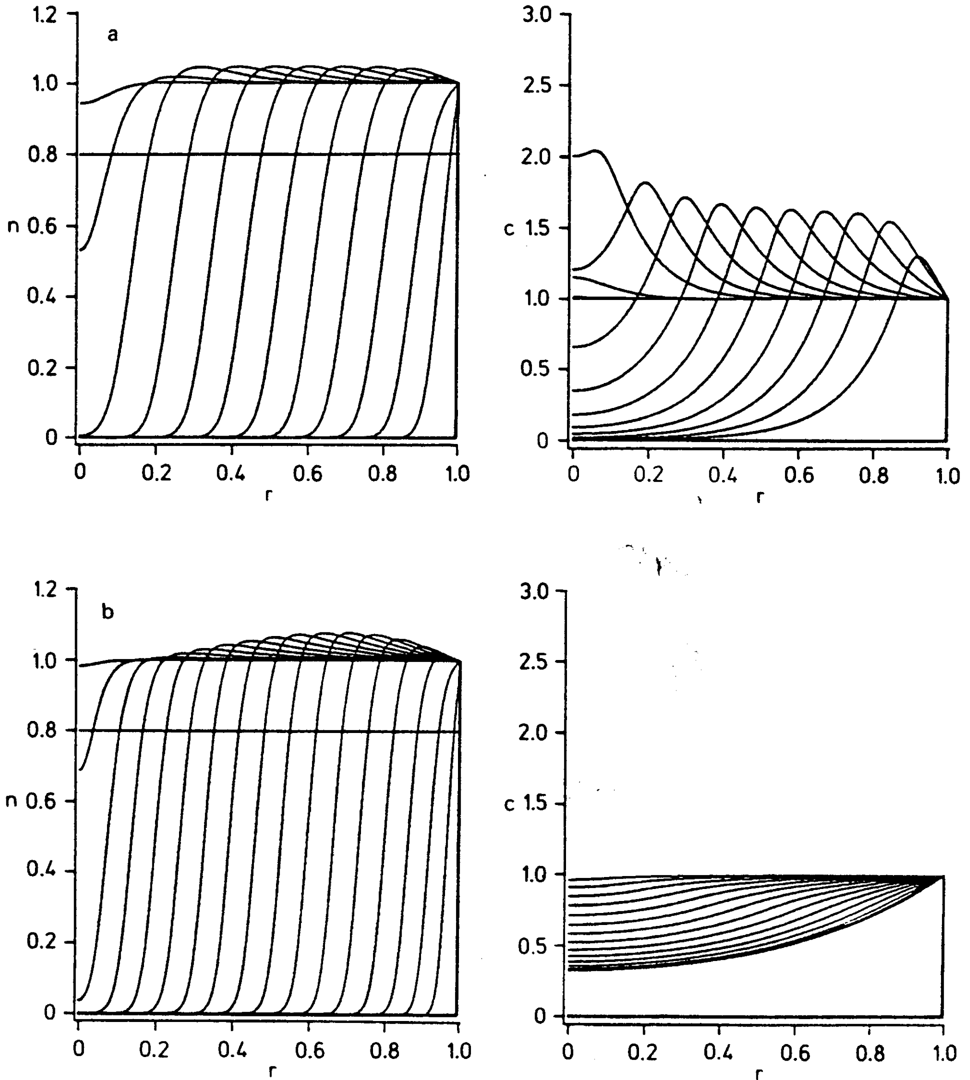

Fig. 4. Cell density (n) and chemical concentration (c) as a function of radius (r) at a selection of equally spaced times, from Eqs. (1). **a** Biochemical activation of mitosis with parameter values $D = 5 \times 10^{-4}$, $D_c = 0.45$, $\lambda = 30$, $h = 10$, $\alpha = 0.1$, $c_m = 40$; **b** biochemical inhibition of mitosis with parameter values $D = 10^{-4}$, $D_c = 0.85$, $\lambda = 5$, $h = 10$

6. Travelling wave solutions

For both types of chemical, the qualitative form of the solution in the linear phase is of a wave moving with constant shape and constant speed. Such a solution is amenable to analysis if we consider a one dimensional geometry, rather than the two dimensional radially symmetric geometry considered above. This is biologically relevant for large wounds of any shape, since to a good approximation these are one dimensional during much of the healing process. Numerical solutions of the model equations for this new geometry are not

significantly different from those in Figs. 3 and 4; the dimensionless wave speeds are approximately 0.048 for the activator and 0.03 for the inhibitor.

Mathematically, we look for travelling wave solutions of the form $n(x, t) = N(z)$, $c(x, t) = C(z)$, $z = x + at$, where a is the wave speed, positive since here we consider waves moving to the left. Substituting these forms into (1) gives the following system of ordinary differential equations:

$$\begin{aligned} aN' &= DN'' + s(C) \cdot N \cdot (2 - N) - N \\ aC' &= D_c C'' + \lambda g(N) - \lambda C \end{aligned} \tag{2}$$

where primes denote differentiation with respect to z. Biologically appropriate boundary conditions are $N(-\infty) = C(-\infty) = 0$, $N(+\infty) = C(+\infty) = 1$, $N'(\pm\infty) = C'(\pm\infty) = 0$.

Since the system of ordinary differential equations is fourth order, and thus difficult to analyse globally, we consider two reasonable approximations which reduce the order of the system: firstly treating λ as infinite and secondly treating D as zero. For the parameter values we are using, the values of these dimensionless parameters are $D = 5 \times 10^{-4}$, $\lambda = 30$ for the activator, and $D = 10^{-4}$, $\lambda = 5$ for the inhibitor. Biologically, the first approximation corresponds to the chemical kinetics always being in equilibrium, while the second corresponds to an absence of cellular diffusion, so that increase in cell density is due only to mitosis.

In the numerical solution $c \ll c_m$ for all r and t, and thus it is a good approximation in the activator case to take $s(c)$ as a simple linear function. Specifically, we approximate

$$s(c) \approx \gamma c + 1 - \gamma \quad \text{where } \gamma = \frac{2(h-1)}{c_m - 2}.$$

The numerical solution of (1) using this form differs negligibly from that with the original. We use this linear approximation in the subsequent analysis, since it makes this analysis much easier algebraically. The approximation is valid provided $c_m \gg 1$.

Travelling wave solutions with $\lambda = \infty$. Here we consider that the derivative terms in the second equation are negligible compared with the reaction terms. Intuitively this seems a reasonable approximation in the case of an inhibitor ($\lambda = 5$), and a good approximation in the case of an activator ($\lambda = 30$). The system then reduces to a second order ordinary differential equation for N, namely

$$N'' = \frac{a}{D} N' - \frac{1}{D} \psi(N) \quad \text{where } \psi(N) = s[g(N)] \cdot (2N - N^2) - N. \tag{3}$$

We look for a solution with boundary conditions $N(+\infty) = 1$, $N(-\infty) = 0$ and $N'(\pm\infty) = 0$, and of course with $N \geqslant 0$ throughout.

A straightforward plot shows that, on the interval $(0, 1)$, $\psi(N)$ has an essentially parabolic shape in both cases. Thus this equation can be analysed in an analogous way to the standard analysis for travelling wave solutions of the Fisher equation, $u_t = u_{xx} + u(1 - u)$. This shows that there is a unique solution of the required form for each wave speed $a \geqslant a_{\min} = 2\{D(2s(0) - 1)\}^{1/2}$ (see, for example, Murray (1989) for a summary of this standard analysis). In the usual way we expect the solution to evolve to a travelling wave with $a = a_{\min}$ for initial conditions such that $N = 1$ for sufficiently large z and $N = 0$ for sufficiently small z; biologically relevant initial conditions for our problem certainly satisfy this.

The parameter values we are using give dimensionless values $a_{min} = 0.01$ for the activator and $a_{min} = 0.09$ for the inhibitor. These compare to the wave speeds 0.05 and 0.03 respectively found in the numerical solution of (1). The discrepancy indicates inadequacies in the approximation of chemical decaying as it is formed; intuitively we expect this approximation to give a lower wave speed for the activator and a higher wave speed for the inhibitor.

In the inhibitor case an approximate analytic solution for the travelling wave can also be obtained. Rescaling the independent variable ($z_{new} = z_{old}/a$) gives

$$\epsilon N'' - N' + \psi(N) = 0 \tag{4}$$

where $\epsilon = D/a^2$ and primes denote $d/d\zeta$, $\zeta = z/a$. This looks intuitively like a singular perturbation problem in the small parameter $\epsilon \approx 0.01$, but it can in fact be solved by regular perturbation techniques. There is an analogous standard method for solving the travelling wave problem for the Fisher equation: see, for example, Murray (1989). In the activator case the method fails because $\epsilon \approx 5$. We look for a solution of (4) in the form

$$N(\zeta;\epsilon) = N_0(\zeta) + \epsilon N_1(\zeta) + \epsilon^2 N_2(\zeta) + \cdots.$$

Substituting this into (4) and equating coefficients of powers of ϵ gives

$$\begin{aligned} N_0' &= \psi(N_0) \\ N_1' &= N_1 \cdot \frac{d\psi(N_0)}{dN_0} + N_0'' \\ &\;\vdots \end{aligned}$$

The boundary conditions are

$$N_0(-\infty) = 0, \qquad N_0(0) = 1/2, \qquad N_0(+\infty) = 1$$
$$N_i(-\infty) = N_i(0) = N_i(+\infty) = 0 \quad \text{for } i \geq 1.$$

The value of $N(0)$ is arbitrary; it must be specified to give a unique solution (this simply fixes the origin of z). We choose $N(0) = 1/2$, so that

$$\begin{aligned} \zeta &= \int_{1/2}^{N_0} \frac{d\xi}{\psi(\xi)} \\ &= \left(\frac{1}{2h-1}\right)\ln[2N_0] - \left(\frac{2h-1}{3h-2}\right)\ln[2(1-N_0)] \\ &\quad + \frac{(4h-3)(h-1)}{(2h-1)(3h-2)}\ln\left[\frac{2(h-1)N_0 + 2(2h-1)}{5h-3}\right] \end{aligned}$$

for the inhibitor, assuming $h > 1$ as above. This cannot be inverted explicitly. However, observing that ζ is a monotonically increasing function of N_0, we consider N_0, rather than ζ, as the independent variable. Now

$$\frac{dN_1}{d\zeta} = N_1 \frac{d\psi}{dN_0} + \frac{d^2 N_0}{d\zeta^2},$$

which on dividing by $dN_0/d\zeta$ gives

$$\begin{aligned} \frac{dN_1}{dN_0} &= \frac{N_1}{dN_0/d\zeta} \cdot \frac{d\psi(N_0)}{dN_0} + \frac{d}{dN_0}\left(\frac{dN_0}{d\zeta}\right) \\ &= \frac{N_1}{\psi(N_0)} \frac{d\psi(N_0)}{dN_0} + \frac{d\psi(N_0)}{dN_0}, \quad \text{on using } \frac{dN_0}{d\zeta} = \psi(N_0). \end{aligned}$$

Dividing through by $\psi(N_0)$ gives

$$\frac{d}{dN_0}\left[\frac{N_1}{\psi(N_0)} - \ln[\psi(N_0)]\right] = 0.$$

Thus, since $N_1 = 0$ when $N_0 = 1/2$ (at $\zeta = 0$),

$$N_1 = \psi(N_0)\ \ln\left[\frac{\psi(N_0)}{\psi(1/2)}\right].$$

Plots of N_0 and $N_0 + \epsilon N_1$ against z are shown in Fig. 5 and compared to the numerical solution of (1) at a time in the middle of the linear phase of wound repair. These show that the first order correction is already small. The analytic solution agrees reasonably with the partial differential equation solution in regard to the slope of the linear portion of the wave-front. However, it fails to capture the important feature that $n > 1$ in part of the wave-front. This is an inadequacy in the approximation $\lambda = \infty$.

Travelling wave solutions with $D = 0$. Given the shortcomings of the previous approximation we consider now the approximation $D = 0$. Recall that $D = 5 \times 10^{-4}$ for the activator model and $D = 10^{-4}$ for the inhibitor model. The fourth order system (2) now reduces to a third order system:

$$N' = -\frac{N}{a} + \frac{1}{a}s(C)(2N - N^2)$$
$$C'' = \frac{a}{D_c}C' + \frac{\lambda}{D_c}C - \frac{\lambda}{D_c}g(N). \tag{5}$$

We look for a solution subject to boundary conditions $N(-\infty) = C(-\infty) = 0$, $N(+\infty) = C(+\infty) = 1$, $C'(\pm\infty) = 0$, with $N, C \geqslant 0$ for all z.

We solved this system numerically by treating $N(z_l) + C(z_l)$ and $N(z_r) + C(z_r) - 2$ as functions of $C(z_m)$ and $C'(z_m)$, where we consider the equations in the interval $[z_l, z_r]$ with z_m an intermediate point. The value of $N(z_m)$ was fixed at its value in the numerical solutions of (1). Initially we took

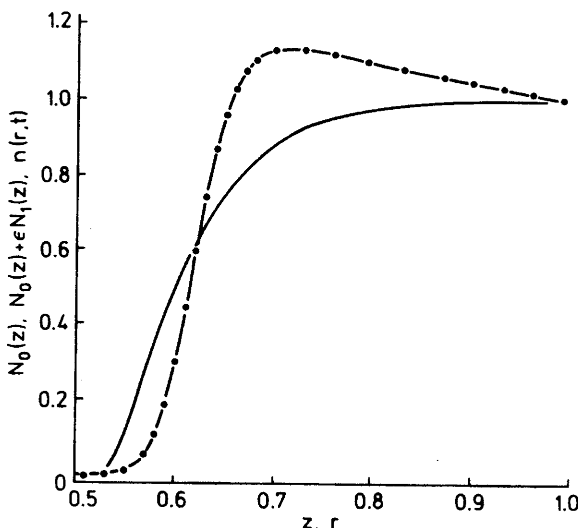

Fig. 5. Comparison of N_0 (*full curve*) and the numerical solution of (1) (*curve and dots*) for the inhibitor model. The $O(\epsilon)$ correction ϵN_1 to the leading order term N_0 in the asymptotic solution of (4) is so small that it is not visible in a graphical representation. The parameter values are $D = 10^{-4}$, $D_c = 0.85$, $\lambda = 5$, $h = 10$

$z_l = 0$, $z_r = 1$ and solved the equations using the Powell hybrid method. The initial estimates for $C(z_m)$ and $C'(z_m)$ were taken from the numerical solutions of (1). The ordinary differential equations were integrated both forwards and backwards from z_m using a Merson form of the Runge–Kutta method. With the solutions thus obtained for $C(z_m)$ and $C'(z_m)$ as initial estimates, we repeated the simulation with $z_l = -0.5$, $z_r = 1.5$ and continued this process of expanding the interval until the effect on the solution within [0, 1] was negligible. The solutions thus obtained are compared with the numerical solutions of (1) in Fig. 6: for both the activator and inhibitor models, the reasonably close agreement indicates that the approximation $D = 0$ is a good one.

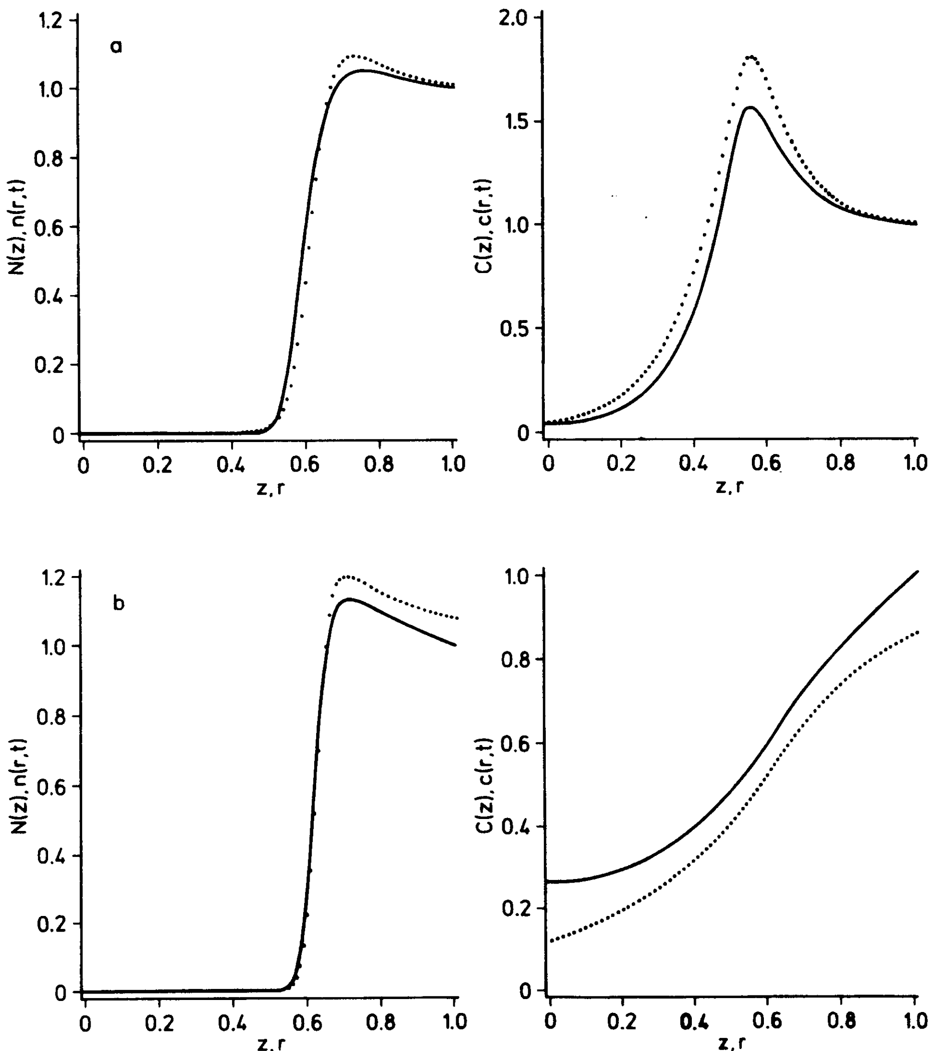

Fig. 6. Comparison of the numerical solutions of (1) (*full curves*) and (5) (*dotted curves*). **a** Activator model with parameter values $D = 5 \times 10^{-4}$, $D_c = 0.45$, $\lambda = 30$, $h = 10$, $\alpha = 0.1$, $c_m = 40$, $a = 0.048$; **b** inhibitor model with parameter values $D = 10^{-4}$, $D_c = 0.85$, $\lambda = 5$, $h = 10$, $a = 0.03$. These values for the wave speed a are those calculated from the numerical solutions of (1)

For the activator, phase space considerations give an upper bound on the wave speed. The solution of (5), obtained numerically, approaches the steady state monotonically. However, for parameter values close to those we are using, Eq. (5) has two eigenvalues with negative real parts at this steady state, with the third eigenvalue real and positive. Further, the two eigenvalues with negative real parts are complex unless $a \leqslant a_{max}$, where a_{max} is the value of a at which the eigenvalue equation has two equal negative roots. This condition gives a cubic in a_{max}:

$$\{[(1+\lambda)^2 - 4\Gamma]/4D_c\}a_{max}^6 + \{4\Gamma(3+D_c^2) - 18(1+\lambda)\Gamma + 2(1+\lambda)^2(1+2\lambda)\}a_{max}^4 + D_c\{(1+\lambda)^2 + 3\Gamma(6\lambda+2-9\Gamma)\}a_{max}^2 + 4D_c^2\Gamma = 0$$

where $\Gamma = \lambda[1 - s'(1)g'(1)]$.

For the parameter values we are using, this equation has a unique solution in $(0.0, 0.1)$; numerical solution gives the upper bound as $a_{max} \approx 0.0546$. This compares with the dimensionless wave speed 0.05 found in the numerical solution of (1).

Also for the activator, we can obtain an analytic solution of (5), again by regular perturbation theory. We write the equations as

$$aN' = \epsilon(C-1)(2N - N^2) + (N - N^2)$$
$$D_c C'' - aC' - \lambda C = -\lambda \cdot \frac{(1+\alpha^2)N}{N^2+\alpha^2}$$

and treat ϵ ($=2(h-1)/(c_m - 2) \approx 0.47$) as a small parameter. With this value for ϵ, we will require the $O(\epsilon)$ correction to the $O(1)$ solution. We look for a solution of the form

$$N(z;\epsilon) = N_0(z) + \epsilon N_1(z) + \epsilon^2 N_2(z) + \cdots$$
$$C(z;\epsilon) = C_0(z) + \epsilon C_1(z) + \epsilon^2 C_2(z) + \cdots. \tag{6}$$

Substituting this into the ordinary differential equations, changing the independent variable to $\xi = e^{z/a}$ and equating coefficients of ϵ^0 gives

$$\xi N_0' = N_0 - N_0^2$$
$$\kappa\xi(\xi C_0')' - \xi C_0' - \lambda C_0 = -\lambda \cdot \frac{(1+\alpha^2)N_0}{N_0^2+\alpha^2},$$

where $\kappa = a^{-2}D_c$ and primes denote $d/d\xi$. The relevant boundary conditions are $N_0(+\infty) = C_0(+\infty) = 1$, $N_0(0) = C_0(0) = 0$, with $N_0(1) = 1/2$ for uniqueness. Straightforward separation of variables gives $N_0 = \xi/(1+\xi)$. For C_0, the method of variation of parameters gives the general solution of

$$\kappa\xi[\xi y'(\xi)]' - \xi y'(\xi) - \lambda y(\xi) = F(\xi) \quad \text{as } y(\xi) = \gamma_+(\xi)\xi^{q^+} + \gamma_-(\xi)\xi^{q^-} \tag{7}$$

where

$$\gamma_\pm' = \frac{\pm 1}{\sqrt{1+4\lambda\kappa}} \cdot \frac{F(\xi)}{\xi^{(q^\pm+1)}} \quad \text{and} \quad q^\pm = \frac{1 \pm \sqrt{1+4\lambda\kappa}}{2\kappa}.$$

Using this,

$$C_0 = \frac{\lambda(1+\alpha^2)}{\sqrt{1+4\lambda\kappa}}\left[\xi^{q^+}\int_\xi^{+\infty} \frac{1}{x^{(q^++1)}} \cdot \frac{N_0(x)}{N_0(x)^2+\alpha^2}\,dx + \xi^{q^-}\int_0^\xi \frac{1}{x^{(q^-+1)}} \cdot \frac{N_0(x)}{N_0(x)^2+\alpha^2}\,dx\right]. \tag{8}$$

Here the values of the two constant limits of integration are necessary (but not sufficient) for convergence at $\xi = 0$ and $+\infty$.

We consider now the boundary conditions (which imply convergence) by investigating the behaviour of each of the two integrals in (8) as ξ approaches 0 and $+\infty$. We have

$$\begin{aligned}
&\lim_{\xi \to +\infty} \xi^{q^+} \int_\xi^{+\infty} \frac{1}{x^{(q^+ + 1)}} \cdot \frac{N_0(x)}{N_0(x)^2 + \alpha^2}\, dx \\
&= \lim_{\theta \to 0} \frac{1}{\theta^{q^+}} \int_{1/\theta}^{+\infty} \frac{1}{x^{(q^+ + 1)}} \cdot \frac{N_0(x)}{N_0(x)^2 + \alpha^2}\, dx \\
&= \lim_{\theta \to 0} \frac{1}{q^+ \theta^{(q^+ - 1)}} \cdot \frac{-\theta^{q^+}(1 + 1/\theta)}{\theta^{-2} + \alpha^2 (1 + 1/\theta)^2} \cdot (-\theta^{-2}) \\
&= \frac{1}{q^+(1 + \alpha^2)},
\end{aligned}$$

where we used L'Hôpital's rule and the expression for N_0 in the third step. Similarly

$$\lim_{\xi \to +\infty} \xi^{q^-} \int_0^\xi \frac{1}{x^{(q^- + 1)}} \cdot \frac{N_0(x)}{N_0(x)^2 + \alpha^2}\, dx = \frac{-1}{q^-(1 + \alpha^2)}.$$

Thus

$$\begin{aligned}
C_0(+\infty) &= \frac{\lambda(1 + \alpha^2)}{\sqrt{1 + 4\lambda\kappa}} \cdot \frac{1}{1 + \alpha^2} \cdot \left(\frac{1}{q^+} - \frac{1}{q^-} \right) \\
&= 1,
\end{aligned}$$

using the expressions for $q^\pm$. Similarly the condition at $\xi = 0$ is satisfied.

Now consider the first order perturbations. Equating coefficients of ϵ gives

$$\xi N_1' = N_1(1 - 2N_0) + (C_0 - 1)(2N_0 - N_0^2)$$

$$\kappa\xi(\xi C_1')' - \xi C_1' - \lambda C_1 = -\lambda(1 + \alpha^2) \cdot \left[\frac{\alpha^2 - N_0^2}{(\alpha^2 + N_0^2)^2} \right] \cdot N_1.$$

The relevant boundary conditions are now

$$N_1(0) = N_1(+\infty) = C_1(0) = C_1(+\infty) = N_1(1) = 0.$$

In the first equation, substituting for N_0, multiplying through by the integrating factor $(1 + 1/\xi)^2$ and integrating gives

$$N_1 = \frac{1}{\xi + 2 + 1/\xi} \int_1^\xi (C_0(x) - 1)(1 + 2/x)\, dx.$$

Use of L'Hôpital's rule as above shows that the boundary conditions are satisfied. Then (7) gives C_1, and again we use L'Hôpital's rule to confirm that the boundary conditions are satisfied. By repeating this process we can derive all the terms in the expansion, showing in particular that λ, D_c and a occur in each term of the series (6), and thus in the solution as a whole, only in the groupings $q^\pm$ and $\lambda/\sqrt{1 + 4\lambda\kappa}$.

7. Conclusion

We have constructed a simple model for epidermal wound healing in which the parameter values are based as far as possible on experimental fact. Numerical solutions of the model with either chemical activation or inhibition of mitosis compare well with experimental data on the normal healing of circular wounds, supporting the view that biochemical regulation of mitosis is fundamental to the process of epidermal migration in wound healing. Analytical investigation of the solutions has been possible because these solutions have approximately travelling wave form during most of the healing process. We have studied the travelling wave ordinary differential equations under two biologically relevant approximations. This provided information about the accuracy of these approximations and the roles of the various model parameters in the speed of healing of the wound.

Acknowledgements. We would like to thank Prof. Helene Sage (Dept. of Biological Structure, University of Washington) and Dr. Katherine Sprugel (Zymogenetics, Seattle) for several invaluable discussions on wound healing. JAS was supported in part by a graduate studentship of the Science and Engineering Research Council of Great Britain. JDM was supported in part by Grant DMS-900339 from the U.S. National Science Foundation.

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