Can a death signal half-life be used to sense the distance to a lesion site in axons?

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Abstract Neuron response to injury depends on the distance to the lesion site, which means that neurons are capable of sensing this distance. Several mechanisms explaining how neurons can do this have been proposed and it is possible that neurons use a combination of several mechanisms to make such measurements. In this paper we investigate the feasibility of the simplest mechanism, which is based on the hypothesis that death signals, produced at the lesion site, propagate toward the neuron soma. The signals are propelled by dynein motors. If signals have a finite half-life, they decay as they propagate. By measuring the concentration of death signals arriving to the soma, neurons should thus be able to determine the distance to the injury site. We develop and solve a transport equation based on the above model. We investigate how a death signal distribution depends on the dynein velocity distribution. We evaluate the efficiency of such a mechanism by investigating the sensitivity of death signal concentration at the soma to the distance to the injury site. By using the hypothesis that system performance is optimized by evolution, we evaluate death signal half-lives that would maximize this sensitivity.

Keywords Neuronal injury · Death signals · Motor-dependent signals · Dynein motors · Retrograde axonal transport

1 Introduction

There is growing interest in simulating non-lethal axonal injuries as well as differences between normal and pathologically altered axons (Coggan et al. [1], Volman and Ng [2], Boucher et al. [3], Kuznetsov and Kuznetsov [4]). Accurate models may aid in finding cures to various debilitating neurological diseases (Medana and Esiri [5]).

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It is well known that the neuron response to axonal injury depends on the distance to the lesion site (Cancalon [6], You et al. [7]). Axonal injury can lead to neuronal apoptosis, axonal degeneration, or axonal regeneration (Shin et al. [8]). According to Cancalon [6], if the lesion site is less than 40 mm away from the cell body, all mature long garfish olfactory neurons die. However, if the distance is between 60 and 100 mm, an increasing number of neurons survive and regenerate (from 60 to 90%, respectively).

The question is then how neurons can measure distances to lesion sites. Normally cells can determine their location in tissues by measuring steady-state morphogen gradients. A morphogen is released from a spatially localized source and its distribution in a tissue is determined by its diffusivity as well as by the rates of its production, uptake, and destruction (Bollenbach et al. [9], Lander [10]).

However, axons are too long to effectively use diffusion to measure distances between the lesion site and the soma. Neurons can utilize different strategies to measure the distance to the lesion site (Rishal and Fainzilber [11]). Signal spreading over long distances due to dynein velocity distribution is one possible mechanism; sensing the time delay between the arrival of fast (electrophysiological) and slow (motor-driven chemical) signals is another possible mechanism (Kam et al. [12], Kuznetsov [13]). The third mechanism, recently proposed in Rishal et al. [14], is based on measuring the frequency of an oscillating signal produced by a feedback mechanism where the retrograde signal, generated at the axon synapse, periodically resets the original anterograde signal (see also Albus et al. [15]).

In this paper we discuss and quantify a more simple possibility. Prilloff et al. [16] discovered that the recovery of axonal transport in retinal ganglion cells leads to the second wave of cell death. Based on these results, Prilloff et al. [16] suggested the existence of a retrogradely transported death signal. The importance of a retrogradely transported JNK (c-Jun N-terminal kinase) signal activated at the lesion site immediately after an injury was also discussed in Fernandes et al. [17].

We evaluated, by means of mathematical modeling, the possibility that the decay of death signals produced at the lesion site, due to their finite half-life, is used to measure the distance to the lesion site. This possibility is mentioned in Rishal et al. [14] and our goal was to develop the continuum transport equation that would allow us to evaluate the feasibility and efficiency of such a mechanism. Based on the hypothesis that the system has been optimized by evolution to maximize sensitivity, we also evaluated possible death signal half-lives.

The advantage of the proposed mechanism of sensing the distance to the lesion site is its simplicity and robustness. There is no need for sophisticated sensors that would allow the neuron to measure a delay between the arrival of the fast and slow signals. There is also no need to measure the signal frequency. Since biological systems often utilize redundant mechanisms, it is also possible that neurons use this principle in combination with other mechanisms of length sensing, which are discussed above. A combination of a simple and a more sophisticated mechanism may improve the overall robustness of the system.

It should be noted that the signaling network responsible for retrograde injury signaling in axons is extremely complex and there is a great variety of phosphoproteins that are involved in this process (Michailevski et al. [18], Rishal and Fainzilber [19]). In addition, dynein motors, which transport retrograde injury signals, are ATP-powered. ATP production, in turn, is often compromised following axonal injury (Giza and Hovda [20]). This probable ATP deficit can make the situation even more complex. The presented modeling work should thus be viewed as only a feasibility study. More details can be incorporated into the mathematical model as more experimental data become available.

2 Governing equations

We modeled the retrograde transport of death signals from the lesion site by using a continuum approach. The approach is similar to that developed in Smith and Simmons [21] and Kuznetsov [13] but because of the small anticipated diffusivity of the death signals and the large transport distance, we neglected the diffusivity of the death signals. This assumption was justified by the following estimate.

Retrograde signals in axons are commonly transported in signaling endosomes (Ibanez [22], Murphy et al. [23], Miaczynska and Bar-Sagi [24]). The diameter range of signaling endosomes is estimated to be 50–220 nm (Schmieg et al. [25]). According to Swaminathan et al. [26], cytoplasmic viscosity is 1.5–3.2 times larger than the viscosity of water and the dynamic viscosity of water at room temperature is approximately 10^{-3} Pa s. Assuming a signaling endosome diameter of 100 nm and cytoplasmic viscosity of 3×10^{-3} Pa s, the Stokes–Einstein equation predicts the signaling endosome diffusivity to be $1.5 \mu\text{m}^2/\text{s}$. The diffusion time can be estimated as $(L^*)^2/2D^*$. It will thus take a death signal 3.33×10^9 s to travel a distance of 100 mm by diffusion. (100 mm is a typical distance between the soma and the lesion site, see Cancalon [6].)

It should be noted that not all of the intra-axonal space is available for diffusion. In addition to signaling endosomes, there are microtubules, intra-cellular structures, and mitochondria that occupy a significant portion of the axonal cross-section. All of these factors may further reduce the effective diffusivity of signaling endosomes. This will make the diffusion time even larger.

The average speed of signaling endosomes is between 0.2 and $3 \mu\text{m s}^{-1}$ (Cui et al. [27], Zhang et al. [28], Chowdary et al. [29]). Using the value of $1 \mu\text{m s}^{-1}$ for the average death signal velocity, we conclude that it will take a death signal 10^5 s to travel the same distance of 100 mm by motor-driven transport.

Since the motor-driven transport at these distances (~ 100 mm) is by several orders of magnitude faster than the diffusion-driven transport, we account only for transport of death signals by motor-driven transport.

The concentration of death signals is characterized by the linear number density of the death signals, which is the number of death signal particles per unit axon length (μm^{-1}). A schematic diagram of an axon with a lesion site is shown in Fig. 1. Protein degradation is a tightly regulated process (Berg et al. [30]). In the absence of further experimental information we assumed a constant rate of death signal loss as the signals travel toward the soma (Rishal et al. [14]). For the steady-state situation, the governing equation is:

$$v^* \frac{\partial n^*}{\partial x^*} - k^* n^* = 0, \quad (1)$$

where k^* is the rate of death signal degradation (s^{-1}), n^* is the concentration of death signals (μm^{-1}), v^* is the velocity of dynein motors ($\mu\text{m/s}$) (assuming that all dynein motors move with

Fig. 1 Schematic diagram of an axon with a lesion site

one and the same velocity; this assumption is relaxed later on) and x^* is the linear coordinate along the axon that originates at the neuron soma (μm).

Here

$$k^* = \frac{\ln 2}{T_{1/2}^*}, \quad (2)$$

where $T_{1/2}^*$ is the half-life of death signals (s).

Equation 1 was solved subject to the following boundary condition:

$$x^* = L^*, n^* = n_L^*, \quad (3)$$

where L^* is the distance between the neuron soma and the lesion site (μm) and n_L^* is the assumed constant concentration of the death signals at the axon lesion site (μm^{-1}).

Dimensionless quantities are defined as

$$n = \frac{n^*}{n_L^*}, \quad x = \frac{x^*}{L^*}, \quad k = \frac{k^* L^*}{v^*}. \quad (4)$$

The dimensionless form of Eq. 1 is

$$\frac{\partial n}{\partial x} - k n = 0. \quad (5)$$

The dimensionless boundary condition is

$$x = 1, n = 1. \quad (6)$$

The solution of Eq. 5 subject to boundary condition (6) is

$$n(x) = \exp[-k(1-x)]. \quad (7)$$

The dimensional version of Eq. 7 is

$$n^*(x^*; v^*) = n_L^* \exp \left[-\frac{k^* L^*}{v^*} \left(1 - \frac{x^*}{L^*} \right) \right]. \quad (8)$$

For small k^* (large $T_{1/2}^*$) the right-hand side of Eq. 8 can be expanded in power series in k^* . The result is

$$n^*(x^*; v^*) = n_L^* \left(1 - \frac{k^* (L^* - x^*)}{v^*} \right). \quad (9)$$

This means that for small values of k^* the concentration of death signals is linearly distributed between the lesion site and the neuron soma; for large values of k^* the distribution is exponential.

According to Eq. 8, the concentration of death signals arriving to the soma is

$$n_0^*(v^*) = n_L^* \exp \left[-\frac{k^* L^*}{v^*} \right]. \quad (10)$$

Equation 10 shows that the concentration of death signals at the soma exponentially decays as the distance from the lesion site increases. This suggests that the

concentration of death signals at the neuron soma can be used to evaluate the distance from the lesion site.

Equation 8 gives the concentration of death signals under the assumption that all death signals move with the same velocity v^* . This means that all dynein motors that propel the particles move with the same velocity v^* . Our next goal is to address the situation when the dynein velocity is characterized by a probability density function (PDF). We use the approach developed in Kuznetsov [13]. We first consider the situation when dynein velocity distribution is bimodal: some dynein motors move with velocity v_1^* and other dynein motors move with velocity v_2^* . We characterize signal particles by color, depending on the type of dynein motor they associate with. We assume that blue signal particles associate with dynein motors moving with velocity v_1^* while red signal particles associate with dynein motors moving with velocity v_2^* . If at $x^* = L^*$ the concentration of blue signal particles is λn_L^* then the concentration of red signal particles at $x^* = L^*$ is $(1-\lambda)n_L^*$, where $\lambda < 1$. In order to calculate the total concentration of signal particles, we calculate the sum with respect to different velocities, that is $\lambda n^*(x^*, t^*; v_1^*) + (1-\lambda)n^*(x^*, t^*; v_2^*)$. We next consider the case of a large number of colors. We assume that the concentrations of the colored particles at $x^* = L^*$ are distributed according to a stepwise version of $f^*(v^*)$. The total concentration of signal particles at $x^* = L^*$, given by the integral over the whole velocity range, is still equal to n_L^* . Physically we imagine that once signal particles are released from the lesion site, they are picked up by dynein motors whose velocity distribution is characterized by a PDF $f^*(v^*)$. In the limit of an infinite number of colors, we represent the total concentration by the following integral that expresses a superposition of concentrations corresponding to different dynein velocities:

$$n_{f^*(v^*)}^*(x^*) = \int_0^\infty n^*(x^*; v^*) f^*(v^*) dv^*. \quad (11)$$

If all dynein motors move with one and the same velocity v_0^* , then $f^*(v^*) = \delta(v^* - v_0^*)$, where δ is a Dirac delta function.

The concentration of the death signals at the neuron soma is then found from Eq. 11 as:

$$n_{f^*(v^*)}^*(0) = \int_0^\infty n_0^*(v^*) f^*(v^*) dv^*. \quad (12)$$

3 Parameter estimation

In Table 1 we summarized the estimated values of parameters involved in the problem.

If dynein motors have a velocity distribution, the following approach can be utilized. Kam et al. [12] presented two correlations for the dynein velocity distribution, based on experimental data obtained by Ross et al. [32] and Deinhardt et al. [33], respectively. Kuznetsov [13]

then normalized the correlations obtained by Kam et al. [12] so that $\int_0^\infty f^*(v^*) dv^* = 1$. This made it possible to obtain the following probability density functions describing the dynein velocity distributions. The first one (based on data reported in Ross et al. [32]) is

$$f_1^*(v^*) = 5.8145 v^* \exp \left[- \left(\frac{v^* - 0.2738}{0.3363} \right)^2 \right] \quad (13)$$

Table 1 Estimated values of parameters involved in the model

Parameter	Notation	Dimension	Value or range	Reference	Primary value(s) used in computations
Distance between the soma and the lesion site	L^*	μm	$40 \times 10^3 - 100 \times 10^3$ ^a	Cancalon [6]	100×10^3
Half-life of death signals	$T_{1/2}^*$	s	$10^3 - 10^7$ ^b	Coleman [31]	$10^4, 10^5, 10^6$
Average velocity of retrograde transport of signaling endosomes	v^*	$\mu\text{m s}^{-1}$	$0.2-3$ ^c	Cui et al. [27], Zhang et al. [28], Chowdary et al. [29]	1

^a According to Cancalon [6], if the lesion site is less than 40 mm away from the cell body, all mature olfactory neurons die. However, if the distance is between 60 and 100 mm, an increasing number of neurons survive and regenerate (from 60 to 90%, respectively)

^b According to Coleman [31], the half-life of various axonal proteins ranges from hours (or even minutes) to months

^c Signaling endosomes move with instantaneous velocities of $0.5-5 \mu\text{m s}^{-1}$. Since they pause approximately 30% of the time, this gives an average speed (including pauses) of $0.2-3 \mu\text{m s}^{-1}$ (Cui et al. [27], Zhang et al. [28], Chowdary et al. [29])

and the second one (based on data reported in Deinhardt et al. [33]) is

$$f_2^*(v^*) = 1.4616 v^* \exp \left[- \left(\frac{v^* + 0.6657}{1.7231} \right)^2 \right]. \quad (14)$$

In Eqs. 13 and 14 the velocity of dynein motors must be given in $\mu\text{m/s}$. The expected values of dynein velocity are $E_1(v^*)=0.445 \mu\text{m/s}$ and $E_2(v^*)=1.272 \mu\text{m/s}$, respectively. The PDFs given by Eqs. 13 and 14 are displayed in Fig. 2. In Fig. 2 we also display the PDF for the case when all dynein motors move with one and the same velocity of $1 \mu\text{m/s}$. In this case the PDF is given by the Dirac delta function, so that the condition $\int_0^\infty f^*(v^*) dv^* = 1$ is still satisfied.

4 Results and discussion

In Figs. 3 and 4 we present results for three death signal half-lives: 10^4 s (2.78 h), 10^5 s (27.8 h) and 10^6 s (278 h). For a large death signal half-life, the signal distribution in the axon is almost linear, in accordance with Eq. 9 (dash-dot line in Fig. 3a). The distribution is strongly dependent on the death signal half-life; for a small signal half-life the signal concentration exponentially decays from the lesion site to the neuron soma (Fig. 3a, b, c). It is interesting that the distribution of death signals is affected by the signal half-life much more than by the dynein velocity distribution (compare Fig. 3a, b, and c). This suggests that signal decay with the distance from the lesion site can be used to measure how far the lesion site is from the soma.

This idea is demonstrated in Fig. 4. Clearly there is a correlation between the death signal concentration at the soma and the distance to the lesion site. The system is more sensitive to the distance between the soma and the lesion site for small death signal half-lives but the price is

Fig. 2 A PDF of the dynein velocity distribution for the case when all dynein motors move with one and the same velocity (in this case the PDF is given by the Dirac delta function). Also, PDFs based on data reported in Ross et al. [32] and Deinhardt et al. [33] (the correlations for these data were originally given in Kam et al. [12]).

All PDFs were re-normalized by the condition $\int_0^{\infty} f^*(v^*) dv^* = 1$ in Kuznetsov [13]

that signals with a small half-life can only be used to measure small distances. For example, $T_{1/2}^* = 10^4$ s (2.78 h) can allow for the measurement of distances only up to 100 mm (Fig. 4a). A large signal half-life can easily allow for the measurement of distances up to 500 mm and above (see the curves for $T_{1/2}^* = 10^5$ s and $T_{1/2}^* = 10^6$ s in Fig. 4a). Again, a distributed dynein velocity has only a minor effect and does not negatively impact the ability of a neuron to measure distances to the lesion site. Since death signals are transported inside signaling endosomes, this may make them less susceptible to degradation and extend their half-life.

In Fig. 5 we plot the sensitivity, $-\frac{dn_0^*}{dL^*}$, versus the signal half-life. The sensitivity shows the change of the concentration of death signals in the soma with the distance between the soma and the lesion site. It is clear that the sensitivity is larger for smaller distances (smaller L^*). It is also interesting that all curves exhibit peaks that correspond to a maximum sensitivity.

Since signaling transport has been optimized by evolution, it can be expected that there is a certain optimality in the system. We propose that in addition to the major conservation laws (conservation of mass, momentum and energy), we can also use the optimality condition to obtain some additional information about the system. We used this condition to make the following estimates of the expected death signal half-life.

The sensitivity of soma concentration to the distance to the lesion site is found as

$$\frac{dn_0^*}{dL^*} = -\frac{n_L^* \ln 2}{T_{1/2}^* v^*} \exp \left[-\frac{L^* \ln 2}{T_{1/2}^* v^*} \right]. \quad (15)$$

We used Eq. 2 to explicitly introduce the death signal half-life into Eq. 15. The quantity $\frac{dn_0^*}{dL^*}$ is indicative of a neuron's ability to measure the distance to the lesion site by measuring the

Fig. 3 Dimensionless concentration of death signals (normalized by the condition that at the lesion site it is equal to 1) versus the distance from the neuron soma for various death signal half-lives. **a** Assuming that all dynein motors move at the same constant rate of $1 \mu\text{m/s}$, **b** assuming a dynein velocity distribution based on data reported in Ross et al. [32] and **c** assuming a dynein velocity distribution based on data reported in Deinhardt et al. [33]

Fig. 4 Dimensionless concentration of death signals at the neuron soma versus the distance between the soma and the lesion site for various death signal half-lives. **a** Assuming that all dynein motors move at the same constant rate of $1 \mu\text{m/s}$, **b** assuming a dynein velocity distribution based on data reported in Ross et al. [32] and **c** assuming a dynein velocity distribution based on data reported in Deinhardt et al. [33]

Fig. 5 Sensitivity of the system versus the death signal half-life.

The sensitivity is defined as the negative of the variation of the death signal concentration at the soma with the distance between the soma and the lesion site, $-\frac{dn_0^*}{dL^*}$. **a** Assuming that all dynein motors move at the same constant rate of $1 \mu\text{m/s}$, **b** assuming a dynein velocity distribution based on data reported in Ross et al. [32], **c** assuming a dynein velocity distribution based on data reported in Deinhardt et al. [33]

death signal concentration at the neuron soma. In order to find the death signal half-life that gives the largest sensitivity, we solved the following equation:

$$\frac{d}{dT_{1/2}^*} \left(\frac{dn_0^*}{dL^*} \right) = 0. \quad (16)$$

This gave

$$T_{1/2}^* = \frac{L^* \ln 2}{v^*}. \quad (17)$$

Equation 17 gives, for given values of L^* and v^* , the death signal half-life for which the concentration of death signals at the neuron soma will be most sensitive to the distance to the lesion site. The death signal half-life that results in optimal sensitivity thus depends on the distance that needs to be measured. According to Cancalon [6], for long garfish olfactory neurons, the cut-off distance (for smaller distances to the lesion site most axons degenerate, for larger most axons regenerate) is between approximately 60 and 100 mm. The system may be optimized to measure distances in that range. For example, for $L^* = 10^5 \mu\text{m}$ and $v^* = 1 \mu\text{m/s}$, Eq. 17 gives that the optimal value of $T_{1/2}^*$ is $6.93 \times 10^4 \text{ s}$ (19.3 h).

If there is a dynein velocity distribution, the above analysis can be extended as follows. Using Eq. 12, the following equation describing the sensitivity of the death signal concentration at the neuron soma to the distance between the soma and the lesion site was obtained:

$$\frac{dn_0^*}{dL^*} = -n_L^* \ln 2 \int_0^\infty \frac{1}{v^* T_{1/2}^*} \exp \left[-\frac{L^* \ln 2}{T_{1/2}^* v^*} \right] f^*(v^*) dv^*, \quad (18)$$

where n_0^* now means $n_{f^*(v^*)}^*(0)$, the total concentration of death signals (moving with different velocities) into the neuron soma ($x^* = 0$).

The value of the death signal half-life, $T_{1/2}^*$, which maximizes the sensitivity, can be obtained by solving the following equation:

$$\frac{d}{dT_{1/2}^*} \left(\frac{dn_0^*}{dL^*} \right) = -\frac{n_L^* \ln 2}{(T_{1/2}^*)^3} \int_0^\infty \frac{L^* \ln 2 - T_{1/2}^* v^*}{(v^*)^2} \exp \left[-\frac{L^* \ln 2}{T_{1/2}^* v^*} \right] f^*(v^*) dv^* = 0. \quad (19)$$

For example, for $L^* = 10^5 \mu\text{m}$ and the dynein velocity distribution based on data reported in Ross et al. [32] the optimal death signal half-life is $1.80 \times 10^5 \text{ s}$ (50.1 h) while for the dynein velocity distribution based on data reported in Deinhardt et al. [33] the optimal death signal half-life is $6.84 \times 10^4 \text{ s}$ (19.0 h).

5 Conclusions

We developed a model describing the decay of death signals as they propagate from the lesion site to the neuron soma. The model is based on a continuum approach, which was used to write

an equation expressing the conservation of death signals as they propagate toward the neuron soma. The signals are assumed to be propelled by dynein motors. We applied this model to investigate the feasibility of the hypothesis that neurons can determine the distance to the lesion site by measuring the concentration of death signals at the soma. A small distance to the injury site translates into a higher concentration of death signals at the soma, thus enhancing the possibility that apoptotic or axonal degeneration cascades will be triggered. By using the hypothesis that system sensitivity has been optimized by evolution, we estimated expected signal half-lives that optimize system performance.

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