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Effects of fluctuations on electrical properties of gap-junction connected cells in the turtle retina

E. Louis[a,*], C. Degli Esposti Boschi[a], G. Ortega[b], E. Andreu[c], E. Fernández[c], J.V. Sánchez-Andrés[d]

[a]Departamento de Física Aplicada and Unidad Asociada of the Consejo Superior de Investigaciones Científicas, Universidad de Alicante, Apartado 99, E-03080 Alicante, Spain
[b]Centro de Estudios e Investigaciones, Universidad Nacional de Quilmes, R. S. Peña 180, 1876, Bernal, Argentina
[c]Instituto de Bioingeniería, Universidad Miguel Hernández, Campus de San Juan, Apartado 18, E-03550 Alicante, Spain
[d]Departamento de Fisiología, Universidad de La Laguna (Campus de Ofra), E-38320 Tenerife, Spain

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Abstract

Electrical properties of gap-junction connected cells (input voltage and length constant) are shown to depend strongly on fluctuations in membrane and contact conductances. This opens new possibilities and incorporates a further difficulty to the analysis of electrophysiological data, since four, instead of two, parameters (the average values and the magnitude of fluctuations of the two conductances) have to be used in fitting the experimental data. The discussion is illustrated by investigating the effects of dopamine on signal spreading in horizontal cells of turtle retina, assuming a linear cell arrangement. It is shown that while a standard fitting with the average values of the two conductances leads to the conclusion that both are equally affected by dopamine, including fluctuations allows fitting the data by varying just the average contact conductance plus the magnitude of fluctuations.

Keywords: Electrophysiological methods; Gap-junctions; Fluctuations in membrane and contact conductance; Computational models

Electrophysiological methods are a useful tool to investigate the intrinsic electrical properties of excitable cells [8]. As cells are integrated in tissues, their properties can be modified due to inter-cell connections or gap-junctions. The study of the effects of cell coupling has deserved some attention using basically the following experimental approach. Current is injected into a given cell of the tissue and the subsequent voltage deflection at that site, and sites at increasing distances from it, is measured. These data give the input conductance g_0, and, combined with a measurement of the inter-cell distance, the length constant λ. From these two magnitudes, one can in principle derive the membrane and the contact conductances. These methods are commonly used, for instance, to investigate the effects of neurotransmitters on voltage spreading, or alternatively, signal propagation [11–13,16,20].

The procedure discussed above can be greatly complicated by the complex effects of tissue morphology, as discussed in Refs. [2,10,15,18], and/or the presence of fluctuations in the contact and membrane conductances. Conductance fluctuations are originated by the random opening and closing of ion channels and/or randomness in the reliability of synaptic buttons. In fact, it seems well established that the number of open channels at a given time may vary in a very wide range [8]. More specifically, fluctuations down to the millisecond scale in single gap-junction channels have been recently observed [4,19]. The Brownian motion of ions within the cells and in the intercellular spaces, among other factors, underlies this noisy behavior that may have a crucial role in determining cell properties. Note, for instance, the recent studies of the effects of noise in several neuron models [5–7,14,17].

If the time scale of fluctuations is much shorter than the time scale of the measurement (data are usually taken periodically with a not excessively short period to reduce noise, see for instance Refs. [1]), what is actually measured in the experiments described above is some time average of the relevant magnitudes (input resistances and voltage deflections). How this influences the whole fitting process is the

* Corresponding author. Tel.: +34-96-5903542; fax: +34-96-5903464.
E-mail address: enrique@fisic1.ua.es (E. Louis).

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Table 1
Experimental data [13] for the input conductance g_0 and the length constant λ [a] in horizontal cells of turtle retina [b,c]

Dopamine	g_0 (nS)	λ/a	$\langle g_c \rangle$ (nS)	$\langle g_m \rangle$ (nS)	σ_c	σ_m
No	1000	8.44	4210	59	0	0
Yes	900	4.67	2080	96	0	0
Yes	900	4.67	4210	59	1.8	2.16
Yes	900	4.67	2080	59	1.8	0.2

[a] In units of the inter-cell distance *a*.
[b] With and without dopamine.
[c] The data were fitted assuming that cells were arranged in a linear chain. The fitted average values of the membrane and contact conductances $\langle g_m \rangle$ and $\langle g_c \rangle$ and the magnitude of fluctuations (see text) in each of them σ_m and σ_c (see Eq. (2)) are also shown. In the text, we use the symbols g_m^0 and g_c^0 to denote the average values fitted, with no fluctuations, to the data without dopamine [2].

main question addressed in this article. An outstanding consequence of our analysis is that changes induced by dopamine on voltage spreading in horizontal cells of turtle retina [13] can be accounted for by decreasing the average contact conductance and changing the magnitude of fluctuations, while keeping the membrane conductance constant, which is consistent with experimental data and opposite to the results of fittings without fluctuations [2].

We assume that cells, with membrane conductance g_m, and coupled through junctions of contact conductance g_c, are arranged in a linear chain with periodic boundary conditions. Note that, although the details of the results may depend on tissue morphology [2], the main finding of our analysis does not, and therefore, a linear chain will suffice to illustrate our proposal. Besides, in a one-dimensional arrangement, the voltage decreases always exponentially with distance, making the length constant better defined [2]; moreover, as shown below, this exponential behavior is not modified by disorder.

If a current I is injected at site (cell) i, Ohm's law [9] applied to the model outlined above, leads to the following set of equations (Eq. (1)):

$$I - g_m^i V_i = -(g_c^{i,i-1} V_i - g_c^{i-1,i} V_{i-1}) - (g_c^{i,i+1} V_i - g_c^{i+1,i} V_i),$$

$$\text{.....} = \text{.....}$$

$$-g_m^j V_j = -(g_c^{j,j-1} V_j - g_c^{j-1,j} V_{j-1}) - (g_c^{j,j+1} V_j - g_c^{j+1,j} V_j), \qquad (1)$$

where j denotes an arbitrary cell. We assume that $g_c^{i,j} = g_c^{j,i}$ and that both the contact and the membrane conductances are randomly distributed in the range (Eq. (2)):

$$\log_{10} g_l = \log_{10} < g_l > \pm \sigma_l \qquad \text{for } l = m, c \qquad (2)$$

where $\langle g_l \rangle$ denotes the average values, and σ_l accounts for the corresponding magnitude of fluctuations. Then the calculation runs as follows: L cells of membrane conductance chosen randomly as in Eq. (2) are arranged in a linear chain with periodic boundary conditions through junctions

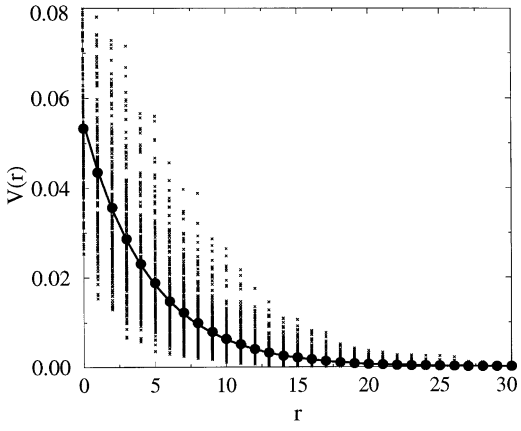

Fig. 1. Numerical results for the input voltage deflection (in units of I/g_m^0, where I is the injected current and g_m^0 is the membrane conductance for the turtle retina without dopamine and with no fluctuations included in the fitting, see Table 1) versus the distance (in units of the inter-cell distance) to the cell at which current was injected. The results correspond to average values of the membrane and contact conductances of $\langle g_m \rangle = 59$ and $\langle g_c \rangle = 4210$ nS, and fluctuations characterized by $\sigma_m = 2.16$ and $\sigma_c = 1.8$ (see Eq. (1)). Crosses give all results obtained for 200 realizations of disorder while filled circles represent their average. The continuous line gives the exponential fitted to the latter.

of contact conductance also randomly chosen as in Eq. (2). The voltage at each cell is then calculated by solving the system of linear equations in Eq. (1) and averaged over a number of realizations of disorder N_R (at least 200). Note that each realization accounts for the spatial distribution of conductance values at a given time, or alternatively, a frozen fluctuation around the average values. Calculations were carried out on linear chains of length $L = 500$, allowing the average conductances to vary around the values corresponding to horizontal cells of turtle retina, namely, $\langle g_c \rangle = g_c{}^0 = 4210$ nS and $\langle g_m \rangle = g_m{}^0 = 59$ nS (see Table 1). The chain length L used in the simulations is large enough for this value of the ratio $\alpha = g_m{}^0/g_c{}^0 \cong 0.014$ [2,3].

The results for the voltage $V(r)$ versus distance r to the cell at which current was injected are illustrated in Fig. 1. It can be readily noted that although fluctuations are noticeable, the average value can be nicely fitted by means of an exponential. From results such as those of Fig. 1, the length constant λ, defined in $V(r) = V(0)\exp(-r/\lambda)$, and the input voltage $V(0)$ (voltage at the cell at which current is injected) can be derived.

The effects of varying the average conductances, while keeping fixed the magnitude of disorder (or fluctuations), on the length constant and on the input conductance g_0, given by $g_0 = I/V(0)$, are illustrated in Fig. 2. It is interesting to note that the general trend of the results shown in Fig. 2, qualitatively agree with those found in a chain in which contact and membrane conductances do not vary from site to site (ordered uniform chain). The exact formulae that give the input conductance and the length constant in an infi-

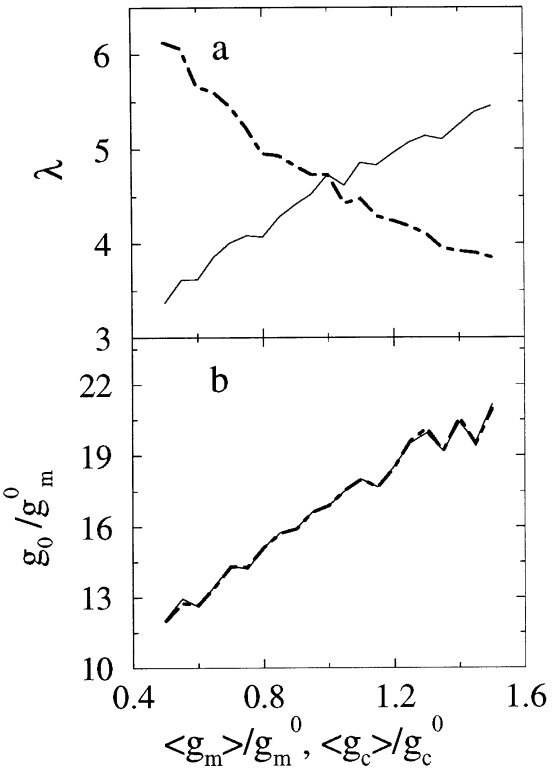

Fig. 2. Numerical results for the length constant (a) in units of the inter-cell distance and the input conductance (b) in units of g_m^0 (see Table 1), versus the membrane (chain line) and the contact (continuous line) conductances (both referred to its value for turtle retina without dopamine, see Table 1). The magnitude of fluctuations was kept, for both conductances, equal to 2. As one of the average conductances was varied, the other was kept equal to the value fitted to the experimental data for the turtle retina without dopamine given in Table 1.

nitely large ordered linear chain are:

$$g_0 = \frac{\sqrt{g_m g_c}}{2} \tag{3a}$$

$$\lambda = \sqrt{\frac{g_c}{g_m}} \tag{3b}$$

Thus, while the input conductance is equally affected by the two conductances, as in Fig. 2b, the effects are opposite in the length constant, exactly as in Fig. 2a.

The dependence of g_0 and λ on the magnitude of disorder are illustrated in Fig. 3. Now, while fluctuations in the membrane or contact conductance have the same effect on the length constant, their effects are opposite on the input conductance. These results can be understood as follows. The spread of the electrical signal is controlled by the maximum value of the membrane conductance and the minimum value of the contact conductance. Then, Eq. (3) tells that while the length constant should be equally affected by fluctuations in either of the two conductances, their effects will be opposite on the input conductance, as in Fig. 3. It is worth mentioning that although this behavior may depend on tissue architecture [2], the main result discussed here is unchanged, namely, the significant dependence of the electrical properties of gap-junction connected cells on fluctuations. A qualitative result that should not be affected by tissue architecture is the decrease in signal spreading promoted by fluctuations in any of the two conductances, a result of clear physiological relevance.

The above results indicate that fluctuations may increase the difficulty of the analysis of electrophysiological data. At the same time, they allow to cope with changes in voltage spreading without varying the average membrane and/or contact conductances. Hereafter, we discuss in more detail the effects that dopamine has on voltage spreading in horizontal cells of turtle retina. Table 1 shows the experimental data [13] for the input conductance and the length constant in horizontal cells of turtle retina with and without dopamine. As discussed in Ref. [2], in order to cope with the changes induced by dopamine, it is necessary to vary both the contact and the membrane conductances, regardless of the cell morphology. In particular, if a linear geometry is assumed, as done here, while dopamine decreases in a factor of two the

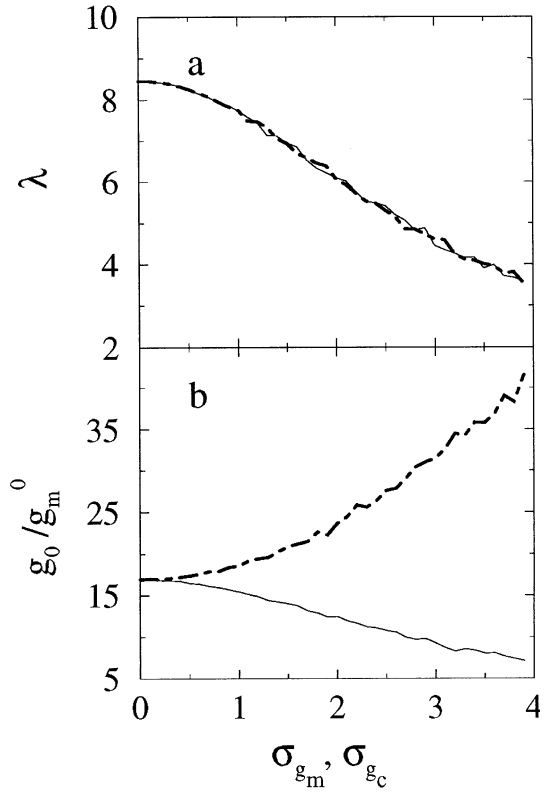

Fig. 3. Numerical results for the length constant (a) in units of the inter-cell distance, and the input conductance (b) in units of g_m^0 (see Table 1), versus the magnitude of fluctuations in the membrane (chain line) and the contact (continuous line) conductances as defined in Eq. (1). As the magnitude of fluctuations in one of the conductances varied, the other was kept equal to zero, while the average conductances were taken equal to g_m^0 and g_c^0 (see Table 1).

contact conductance, it also doubles the membrane conductance. Moreover, as reported in Ref. [2], if the fittings are carried out with two-dimensional networks, dopamine mainly affects the membrane conductance. This openly contradicts the general belief that neurotransmitters mainly modify the contact conductance [13]. Fluctuations allow a rather new interpretation. In Table 1, we report the results of fittings carried out including fluctuations, and either keeping constant the two average conductances, or varying only the average contact conductance, the latter being more consistent with experimental data. It is seen that changes induced by dopamine can be accounted for in both cases. This provides an alternative picture, namely, what neurotransmitters may do is to modify the average membrane and contact conductances *and/or the magnitude of fluctuations in each of them*.

An important consequence of the present analysis is that a full characterization of the electrical behavior of gap-junction connected cells will require measuring average magnitudes and their respective fluctuations. One may, for instance, measure the variance of the voltage $V(r)$ and how it changes with the distance to the input site r (see Fig. 1). Note that a large variability of the voltage has already been reported (see Fig. 4 of Ref. [1]); it is not unreasonable to suspect that the error bars are largely due to intrinsic fluctuations in the system parameters. Then, a complete fitting of the results would involve both the average of the conductances and the magnitude of their fluctuations. Available information on tissue morphology should also be incorporated into the analysis. Until detailed experimental studies along these lines are carried out, many uncertainties on the available data will remain.

As a final remark, we note that despite the good knowledge of the ionic mechanisms underlying cell excitability attained in recent years [8], the basis for intercellular signal propagation remains poorly understood. Understanding this basis seems essential to explain fundamental issues, such as cell coordination and synchronization. We have shown that fluctuations in parameters like membrane and coupling conductance can have per se already unexpected effects on input conductance and signal spreading. The current approach in physiological and pharmacological studies is to check the effects of drugs on the average values of these parameters. Our findings concerning the changes that fluctuations in the two conductances may induce on the input conductance and the length constant can open the way to use fluctuations as targets for active agents.

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