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Light intensity and the membrane parameters of Nitella translucens

Recent observations¹ of the temperature dependence of the membrane potential and resistance of *Nitella translucens* have been satisfactorily explained in terms of differential changes in the K⁺ and Na⁺ permeabilities of the plasmalemma. As an extension of this work we have conducted a preliminary investigation into the effect of light intensity on these membrane parameters.

The methods for measuring the membrane parameters have already been described¹ and the measurements were made for each cell under 'light' and 'dark' conditions. 'Light' conditions corresponded to those under which the cell was illuminated by the laboratory 'Daylight' fluorescent lighting plus the light admitted by the laboratory windows; this latter source could be controlled by suitable dark screens attached to the window frames. In these conditions of light the intensity of illumination was approx. 50 ft-candles; in the dark it was practically zero. The bathing medium for all the experiments was a standard artificial pond water made up of 1.0 mM NaCl, 0.1 mM KCl, 0.1 mM CaCl₂ at a temperature of around 18°. The changes in potential and resistance in going from light to dark conditions were practically instantaneous and in any case the new steady state was attained within 1 min. The speed of the changes indicates that the effects are not thermal and this point was confirmed by checking that no significant temperature changes occurred under the two extremes of light intensity.

TABLE I the values of the plasmalemma potential $(E_{\rm eo})$ and total membrane resistance $(R_{\rm m})$ obtained from 12 cells of N translucens

Cell	$E_{\mathbf{co}}(mV)$	1	$R_{\mathrm{m}} (k\Omega \cdot cm^{2})$		
No	Light	Dark	Light	Dark	
ı	-138	— 145	36.0	45.0	
2	- 125	-130	24 2	346	
3	-123	-126	24 9	33 4	
4	-138	— I 44	30.0	39 4	
5	- 124	-129	25 2	278	
6	146	<u> — 146 </u>	26 5	26 5	
7	-135	- I 37	23.1	25.7	
8	-120	-123	23.3	259	
9	-125	— 129	23 3	28.1	
10	-137	— 142	22.7	29.6	
II	-129	-134	28.4	38 3	
I 2	-136	-138	326	41.0	
Means	-131	-135	26.7	32.9	

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The results of experiments on 12 cells are summarised in Table I. The quoted membrane potentials are those of the plasmalemma and were obtained by adding the tonoplast potential (approx. 15 mV) to the measured potential difference between the vacuole and the external medium (see ref. 1). The membrane resistances are the combined values for the plasmalemma and the tonoplast but can be taken to be essentially the plasmalemma values since they are much the larger of the two². It can be seen that, with the exception of cell No. 6, the effect of reducing the light intensity is to give rise to a small depolarisation of the membrane potential of up to 5 % while the membrane resistance increases by as much as 30%. It is interesting to note that the membrane capacitance could also be obtained during the course of the resistance measurements: the values obtained were close to the accepted 1 μ F/cm² and were found not to be light sensitive.

The membrane resistance is a parameter which is normally associated with the passive permeability properties of the membrane and in view of the large changes that take place when the light intensity is reduced, it is clearly of some importance to determine whether these light effects can be interpreted in terms of permeability changes in the membrane. In order to do this we have used the equations for the membrane potential and resistance which were used in the interpretation of the temperature effects, viz.:

$$E_{co} = \frac{RT}{F} \ln \frac{P_{K}^{+}[K^{+}]_{o} + P_{Na}^{+}[Na^{+}]_{o}}{P_{K}^{+}[K^{+}]_{c} + P_{Na}^{+}[Na^{+}]_{c}}$$
(1)

$$R_{\rm m} = \frac{RT(^{1}/C_{\rm o} - ^{1}/C_{\rm c})}{F^{2} \ln (C_{\rm c}/C_{\rm o})}$$
(2)

 P_{K^+} and P_{Na^+} are, respectively, the K^+ and Na^+ permeability coefficients; $[K^+]_0$, $[Na^+]_o$, $[K^+]_c$ and $[Na^+]_c$ are the respective K^+ and Na^+ concentrations in the bathing medium and the cytoplasm; $C_0 = P_K[K^+]_0 + P_{Na}^+[Na^+]_0$ and $C_c = P_K^+[K^+]_c + P_{Na}^+[Na^+]_0$ $P_{Na}^{+}[Na^{+}]_c$; R, T and F have their usual significance. Eqn. 1 provides the value of the permeability ratio $\alpha = P_{Na}^+/P_K^+$ while Eqn. 2 gives the separate values of the permeability coefficients provided a is already known. The values of these three parameters calculated from the present experiments for both light and dark conditions are given in Table II; in making these calculations it was assumed that the cytoplasmic K⁺ and Na⁺ concentrations were 93 mM and 37 mM, respectively³. The changes in P_{K}^{+} in going from light to dark are generally quite small and there appears to be no consistent trend in these changes. On the other hand, these light intensity changes lower the values of P_{Na}^+ by as much as 30%, and in fact the P_{Na}^+ changes appear to follow closely the changes in membrane resistance. It might be mentioned that the temperature change required to give similar changes in $P_{
m Na^+}$ is about 4 $^\circ$ (see ref. I). Thus the results presented herein suggest that changes in light intensity lead to changes in P_{Na} but leave P_{K} largely unaffected. It would, however, be wrong to suggest that the effect of light intensity is entirely an effect on the passive ion transport mechanisms, since it has been clearly demonstrated4 that the active K+ and Cl-influxes at the plasmalemma are both highly light sensitive.

There is one point which is noteworthy and this concerns the values of α shown in Table II. There is a considerable spread in the values for the 12 cells, and it would appear that in the case of some of the cells there is very little difference between

TABLE II					
THE CALCULATED VALUES OF THE	PERMEABILITY	RATIO α AND	THE	SEPARATE	PERMEABILITY
COEFFICIENTS P_{κ}^+ AND $P_{N_0}^+$					

Cell No.	α	α		$P_{ m K}{}^+ imes$ 10 6 (cm·sec $^{-1}$)		$P_{ m Na}{}^+ imes$ 106 (cm·sec $^{-1}$)	
	Light	Dark	Light	Dark	Light	Dark	
ı	0 34	0.22	2 9	3.0	1.0	0.7	
2	o 74	0.55	2.5	2.1	1.9	1.7	
3	0.84	0.70	2.2	1.9	1.9	1.3	
4	0.34	0.22	3 4	3.6	1.2	0.8	
5	0.79	0 58	2.2	2.6	2.0	1.5	
5 6	0.21	0.21	5.3	5.3	I.I	II	
7	0.41	0.36	4.0	3.9	1.6	1.4	
7 8	1.00	0.84	2.1	2.I	2.I	1.8	
9	0.74	0.58	2.6	2.6	19	1.5	
10	o 36	0 27	4.4	4.I	1.6	1.1	
II	o 58	0.43	26	2.4	1.5	1.0	
12	0.38	0.34	3.0	26	1.2	0.9	
Means	0.48	0.44	3 І	3.1	r 6	1.2	

the K⁺ and Na⁺ permeabilities. This is rather misleading, since the α values were determined from single measurements of the membrane potential and not from a series of measurements obtained for different external K+ and Na+ concentrations (see, for example, refs. 3 and 5) which is a more satisfactory procedure. This point serves to illustrate the unsatisfactory nature of Eqn. 1 which, because of the logarithmic relationship between the potential and the permeability coefficients, makes a an extremely sensitive function of the potential. However, in the continued absence of any other usable expression for the membrane potential, Eqn. 1 provides the only means whereby the numerical value of the permeability ratio may be derived from electrical measurements.

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J. Hogg, E. J Williams and R J. Johnston, Brochim. Brophys Acta, 150 (1968) 640.
 R. M Spanswick and J. W. F. Costerton, J. Cell Sci., 2 (1967) 451.
 R. M. Spanswick, J Stolarek and E. J. Williams, J. Exptl. Botany, 18 (1967) 1.

⁴ E. A. C. MACROBBIE, J. Gen. Physiol, 45 (1962) 861.

⁵ A. B. HOPE AND N. A. WALKER, Australian J. Biol. Sci., 14 (1961) 26.