

A patch-clamp investigation of membrane currents in a novel mammalian retinal ganglion cell line

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We used patch-clamp recordings to characterise membrane currents in RGC-5 cells, a novel postnatal rat-derived retinal ganglion cell line recently used in in vitro models of apoptosis and glaucoma^{1,2}. Our initial studies have concentrated on voltage and stretch-activated channels in undifferentiated cells³. In current-clamp recordings in standard physiological solutions, no action potentials could be evoked and these undifferentiated cells had low resting membrane potentials (-9.7 ± 1.6 mV, $n=29$). In voltage-clamp recordings we could observe no voltage-dependent inward Na^+ or Ca^{2+} currents and the basal membrane currents were small and typically linear (27.5 ± 6.6 pA at a V_p of +60 mV; -25.9 ± 9.2 pA at a V_p of -60 mV, $n= 29$). Occasionally a hyperpolarization-activated inward current was seen. In three cells, dilution potential measurements revealed the basal membrane currents were predominantly anionic. Replacement of external NaCl with KCl revealed inwardly-rectifying K^+ currents in 8 out of 20 cells. Hypo-osmotic external solutions resulted in cell swelling, as seen under the light microscope, although clear hypo-osmotic activated currents (>100 pA) were only observed in two out of 15 cells and this current was insensitive to Gd^{3+} ($100 \mu\text{M}$). Direct application of negative pressure to membrane patches evoked some clear stretch-activated channel activity in two out of 14 patches with a conductance of about 20 pS and 45 pS at membrane potentials of approximately +60 and -60 mV, respectively. Our results indicate that undifferentiated RGC-5 cells are quite unexcitable and contain membrane currents not previously reported in mature mammalian retinal ganglion cells. These results therefore highlight the differences between the electrophysiological properties of undifferentiated RGC-5 cells and acutely dissociated retinal ganglion cells..

- (1) Krishnamoorthy, RR. et al. (2001) *Mol. Brain Res.* 86; 1-12.
- (2) Aoun, P. et al. (2003) *Invest. Ophthalmol. Vis. Sci.* 44; 2999-3004.
- (3) Moorhouse, AJ. et al., (2004) *Brain Res.*, 1003; 205-208.