ANALYSIS OF OUTWARD POTASSIUM CURRENTS IN CULTURED MOUSE (MUS MUSCULUS) ASTROCYTES.

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Astrocytes, the most numerous cell type in the central nervous system, are thought to play an important role in the regulation of the ionic microenvironment, which is essential for normal neuronal function. In this respect, a high membrane permeability to potassium ions has been well established (Hertz, 1986, Ann. N.Y. Acad. Sci., 481:318), a function which could be mediated via voltage dependent ion channels. To study this further, we characterized the K^+ currents in single mouse astrocytes using the whole cell patch clamp technique. Our results indicated that at least two types of K^+ channel co-exist in the membrane of astrocytes.

Astrocytes were prepared as described by Hertz et al., 1989, J. Neurosci. Res., 22: 209;). For K^+ current recording, the astrocytes were dialyzed via the patch electrode with a solution composed of (in mM):120 KCl, 5 NaCl, 5 Mg-ATP, and 15 EGTA, and superfused with a solution composed of (in mM): 137 NaCl, 5.4 KCl, 1 $MgCl_2$, 2 $CaCl_2$, 10 HEPES and 10 glucose.

Two outward current profiles were consistently seen. In the first type, only a slowly activating outward current, that showed little or no inactivation during the time course of a 160 ms clamp pulse was observed. As shown in Figure 1 this current had an activation threshold of -10 mV and was on the whole insensitive to holding potential in the range -90 to -40 mV. In this respect, the current greatly resembles the cardiac delayed rectifier current i_K, and is similar to a non-inactivating current described in cortical astrocytes (Nowak et al., 1987, J. Neuroscience, 7: 101).

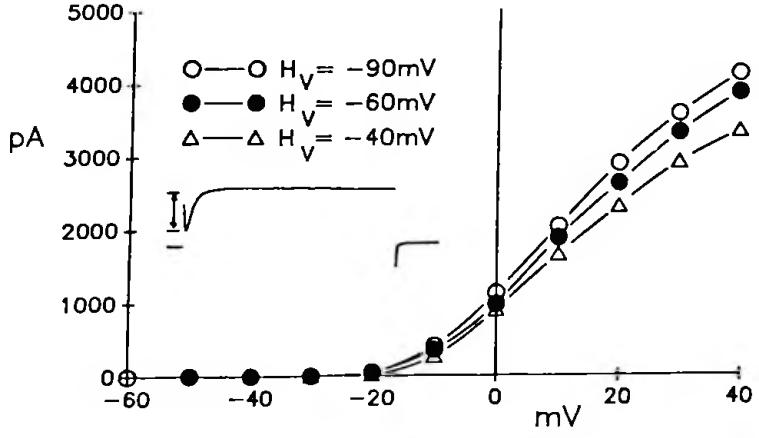
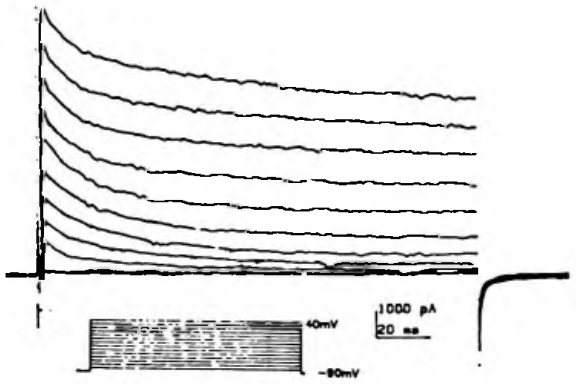


Figure 1. Characteristics of the slowly inactivating outward K^+ current, showing relative lack of sensitivity to changes in the holding potential.

In the second type, the outward current consisted of a more rapidly inactivating outward current superimposed on the slowly activating component. This transient component differed from the slowly inactivating component in two respects. First, the threshold of activation was 30 mV more negative, at -40 mV. Secondly, the transiently activating component was much more sensitive to holding potential. Thus, shifting the holding potential from -90 mV to -60 mV almost completely abolished the transiently activating component, leaving the slow component largely intact (Figure 2). In these respects the current resembles i_{to} recorded in rat ventricular myocytes (See Dukes & Morad, this Bulletin) and the A current that can be recorded from neurones and glial cells (Bevan & Raff, 1985, Nature, 315:229).



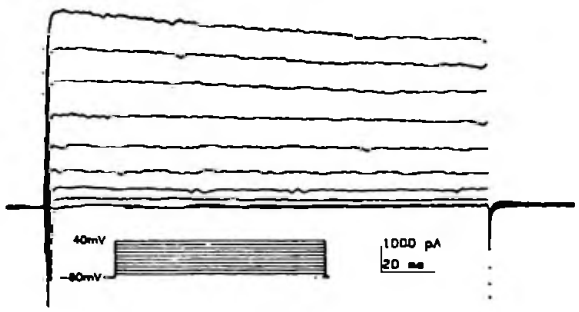


Figure 2. Effect of shifting the holding potential from -90 mV to -60 mV on the amount of transiently activating outward current produced by depolarizing pulses up to +40 mV. Note that the current remaining at the end of the pulse is largely the same at both holding potentials.

Other evidence supporting the existence of two separate K^+ channels in these cells comes from the effect of the addition of 10 mM TEA (tetraethyl-ammonium-chloride), in a cell expressing both types of channels. Application of TEA decreased the magnitude of the outward current without affecting the inactivation rate, and the difference current revealed that TEA only blocked the slowly inactivating component of outward current (Figure 3). This observation is in agreement with the findings in other tissues where TEA at this concentration blocks only the delayed rectifier type currents.

We conclude, therefore, that on the basis of activation threshold, voltage dependence and sensitivity to selective blockers, there appears to be two distinct types of voltage-dependent outward K^+ current in mouse cortical astrocytes. The relative contribution of these currents to the regulation of the K^+ concentration of the extracellular milieu remains to be determined.

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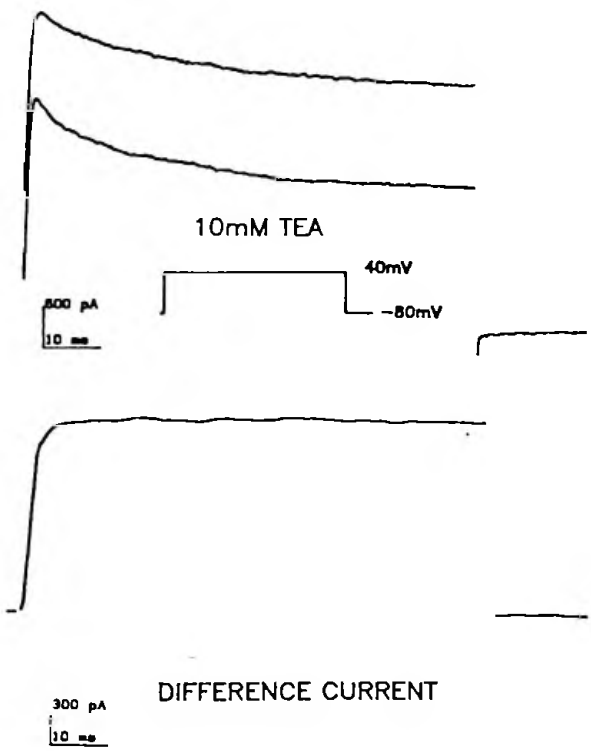

Figure 3. Effect of addition of 10 mM TEA. Upper panel; before (upper trace) and after (lower trace) addition of TEA. Lower panel; difference current after addition of TEA.