ATP-DEPENDENT K^+-CHANNEL AND ITS POSSIBLE BLOCKERS IN ISOLATED GUINEA PIG VENTRICULAR MYOCYTES

Raman Mitra, James Weiss and Martin Morad, Department of Physiology, University of Pennsylvania School of Medicine, Philadelphia, PA 19104

Voltage clamp studies in multicellular and single cell cardiac preparations have identified a large increase in K^+ conductance following inhibition of oxidative metabolism by hypoxia, 2-4-dinitrophenol, and cyanide (Isenberg et al, Pflügers Archives 397:251,1983) Recently, evidence for a K^+ channel which appears when $[ATP]_i$ falls below 0.2mM was demonstrated in cell attached and inside out patch recordings from isolated guinea pig ventricular myocytes exposed to 5.4mM cyanide in a glucose free Tyrode solution (Noma , Nature 305:147,1983). The purpose of our study was to investigate the characteristics of this K^+ channel in the same preparation in more detail using a single electrode whole cell voltage clamp technique. This technique also allowed internal dialysis of the cell. Our findings indicate that within minutes of inhibiting carbohydrate metabolism with 5mM cyanide and 10mM 2-deoxyglucose in a glucose free Tyrode solution, the cells became inexcitable, Ca^{2+} and Na^+ currents disappeared, and a large increase in outward K^+ conductance occurred (Figure 1 A). Ba^{2+} and Cs^+ ,which are known to block the inward rectifier channel, blocked this current only in the inward direction but had little effect on it in the outward direction. Tetracaine, which is known to block both the inwardly and outwardly rectifying K^+ currents reversibly reduced the outward current by 20-30% (Figure 1B). Addition of 5mM ATP to the inside of the patch pipet prevented the development of this large outward current. However, even in the presence of 5mM $[ATP]_i$ the Ca^{2+} current was greatly reduced although the Na^+ current persisted. This finding suggests that intermediate concentrations of ATP inside the pipet were sufficient to inhibit the development of the large K^+ conductance ; however, these levels were insufficient to support the energy requirements of the Ca^{2+} channel. The effect of the metabolic inhibitors on the Ca^{2+} current was reversible only when ATP was inside the pipet. Our results confirm the single channel data and suggest that the large outward current that appeared when total cell metabolism was inhibited was not carried primarily through the inwardly rectifying K^+ channel.

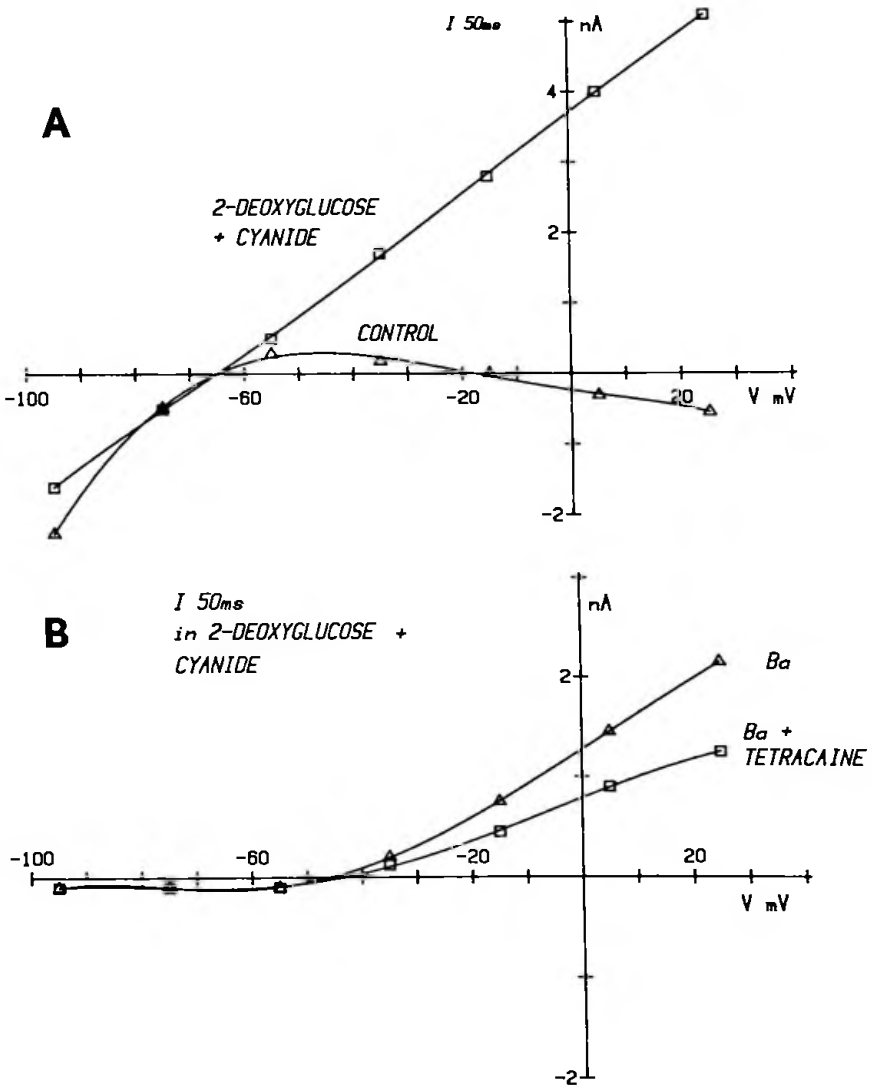
A
I 50ms
nA
4
2
-2
2-DEOXYGLUCOSE
+ CYANIDE
CONTROL
-100
-60
-20
20
V mV
B
I 50ms
in 2-DEOXYGLUCOSE +
CYANIDE
nA
2
-2
Ba
Ba +
TETRACAINE
-100
-60
-20
20
V mV

FIG. 1