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Bupivacaine is an effective potassium channel blocker in heart

Kenneth R. Courtney [a] and Joan J. Kendig [b]

[a] *Research Institute, Palo Alto Medical Foundation, Palo Alto, CA and* [b] *Department of Anesthesia, Stanford University, School of Medicine, Stanford, CA (U.S.A.)*

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The local anesthetic agent bupivacaine increases action potential duration in isolated frog atrial myocytes, and blocks two potassium conductances, I_K and I_{K1}. The effective concentrations, particularly for I_K, are similar to those which depress the sodium conductance. Potassium channel block may thus contribute to bupivacaine's reported cardiotoxicity.

Most studies of local anesthetic actions in excitable membrane have focussed on the sodium channel to explain conduction block in nerve and the antiarrhythmic and cardiotoxic properties of this class of agents in heart. However, early studies suggested that some agents are equieffective in blocking potassium channels in nerve [1,2]. Block of potassium channels might play a role in the overall effects of local anesthetics in heart either directly by altering conduction properties or indirectly by enhancing voltage- and time-dependent sodium channel block. We examined the effects of bupivacaine, an agent which has been suggested to be more cardiotoxic than other local anesthetic agents [3], on two potassium currents in isolated frog atrial myocytes.

Isolated cardiac myocytes, prepared using the technique of Hume and Giles [4], were used for whole cell recordings at 25°C. The standard Ringer's solution contained 106 mM NaCl, 2.5 mM KCl, 5 mM $MgCl_2$, 1.25 mM $CaCl_2$, 10 mM glucose, and 10 mM Hepes-NaOH buffer brought to a pH of 7.2. Electrodes were filled with an electrolyte solution containing 60 mM potassium gluconate, 12 mM KCl, 20 mM K_2EDTA, and 16 mM Hepes-NaOH buffer at pH 7.2. After 5–20 Gigaohm seals were established, the patch was ruptured to allow intracellular recording via a series resistance of 5–20 Megaohms. The single electrode voltage clamp (Dagan 8100) employed a 20 kHz switching rate and a 50% duty cycle.

Initial action potential (AP) durations in the myocytes were 400–700 ms, and resting potentials ranged from −75 to −90 mV. Bupivacaine was applied by dilution into the bathing solution from a concentrated stock solution. Action potential duration increased markedly within a few minutes (Fig. 1). The bupivacaine-related increase in AP duration was dose-dependent and averaged 14% for 23 μM ($n = 2$) and 53% for 47 μM ($n = 3$).

In order to test the role of potassium channel modulation in the increase in AP duration, I_K and I_{K1} were isolated in voltage clamp experiments. Tetrodotoxin (TTX, 10 μM) and $CdCl_2$ (100 μM) were used to block the two inward currents I_{Na} and I_{si} [5]. Step depolarizations from a holding potential of −60 mV were infrequently applied in order to evoke delayed outward currents (I_K); hyperpolarizing steps from the same holding potential were used to measure the time-

Correspondence: K.R. Courtney, Research Institute, Palo Alto Medical Foundation, 860 Bryant Street, Palo Alto, CA 94301, U.S.A.

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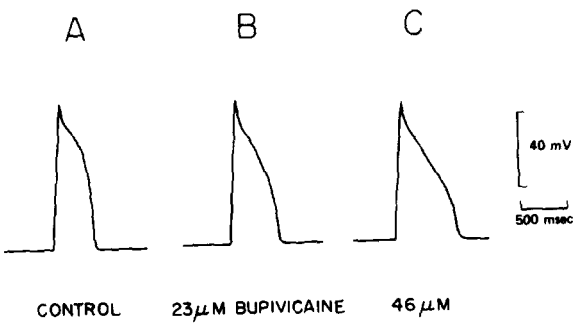

Fig. 1. Effects of bupivacaine on the action potential of a bullfrog atrial myocyte. Whole cell recording in current clamp mode. Action potentials were evoked at 3-s intervals by a 30 ms current pulse of 300 pA through the recording electrode. A, control; B and C, bupivacaine decreases the amplitude and increases the duration of the action potential.

independent background potassium current (I_{K1}). A test concentration of 20 μM was chosen based on the effect of bupivacaine on AP duration. Higher bupivacaine concentrations frequently produced irreversible large increases in cell leakiness, particularly when prolonged depolarizations were applied to evoke I_K; however, two experiments were successfully carried out on I_K at a bupivacaine concentration of 30 μM. Both I_K and I_{K1} were diminished by bupivacaine as illustrated in Fig. 2, I_K being consistently more depressed than I_{K1}. The results for the experiments at 20 μM are presented in Table I; bupivacaine at 20 μM depressed I_K by an average of 16% and I_{K1} by an average of 9%. In the two experiments at 30

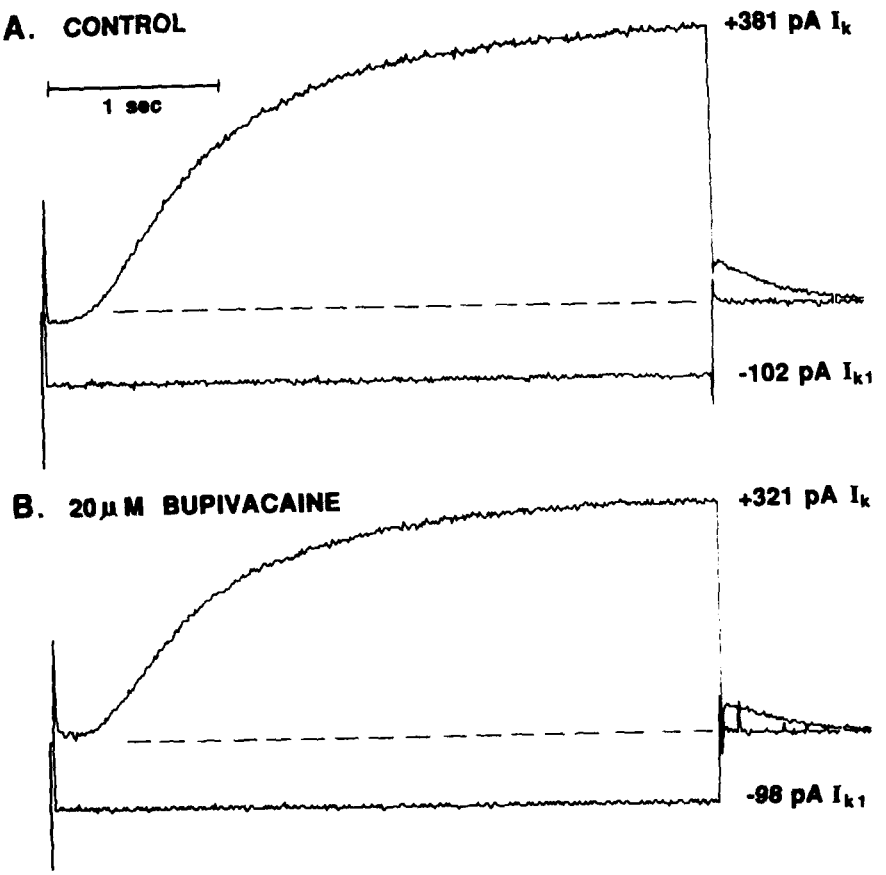

Fig. 2. Effects of 20 μM bupivacaine on two potassium currents. Whole cell voltage clamp of a bullfrog atrial myocyte, Na^+ and Ca^{2+}-dependent currents blocked by TTX and Cd^{2+}, clamp switching rate 20 kHz, duty cycle 50%. (A) Control; (B) bupivacaine. Upper traces in each, currents evoked by a 4-s long step depolarization from a holding potential of −60 to +40 mV (I_K); lower traces, currents evoked by a step hyperpolarization from −60 to −100 mV (I_{K1}). Both currents are depressed by bupivacaine; the depression of the outward current can also be seen in the tail currents following repolarization.

μM, bupivacaine depressed I_K by 21 and 23%.

In order to compare the sensitivity of the potassium channels to that of the sodium channel in this preparation, the actions of bupivacaine on sodium channels were assessed using either AP upstroke velocity ($\dot{V}_{max}$) as a measure of sodium channel availability ($n = 2$) or direct measurements of I_{Na} under voltage clamp ($n = 2$). Similar results were obtained in both kinds of experiments. Bupivacaine (20 μM) blocked 21% of sodium channel availability (Table I). Decreases in I_{Na} were voltage- and frequency-dependent; the rate of recovery following an AP was substantially slowed, an effect observed also in mammalian cardiac preparations [6]. Recovery followed an exponential time course with a 5 s time constant at a holding potential of -90 mV; hyperpolarization increased the rate of recovery, as has been observed in the case of other local anesthetics [7,8]. Bupivacaine effects were difficult to reverse on washing with drug-free solution, perhaps because of this drug's very high lipid solubility.

The results show that bupivacaine blocks both I_K and I_{K1} at concentrations associated with depression of the sodium conductance. The blocking site for neither potassium channel effect is characterized in heart. We have evidence that bupivacaine blocks I_K in nerve in a voltage- and time-dependent fashion [9] similar to the blocking by lipophilic quaternary ammonium ions [10,11]. Both I_K and I_{K1} regulate the duration of the action potential in heart. The kinetics and activation of I_K are consistent with the proposition that this current is responsible for initiating repolarization [12,13], whereas the strongly inwardly rectifying I_{K1} controls the final one-third of repolarization [5]. Since I_{K1} is negligible during the plateau phase of the action potential, only a small amount of I_K is required to initiate repolarization. Therefore small changes in this current will significantly affect AP duration. It is thus probable that prolongation of the action potential we have observed with bupivacaine is due to the blocking of potassium currents, although an additional blocking action on calcium channels cannot be ruled out. Tetracaine, another very lipophilic local anesthetic agent, has been reported to effectively block both potassium currents and calcium currents in guinea-pig ventricular myocytes [14].

These results are relevant to the question of enhanced cardiotoxicity by the more lipophilic local anesthetic agents. Enhanced cardiotoxicity by bupivacaine compared to another less lipid-soluble local anesthetic agent, lidocaine, has been attributed to rapid binding to sodium channels on depolarization, and slow unbinding following repolarization [15]. By blocking potassium channels, and thus prolonging the action potential, bupivacaine may enhance its own voltage-dependent sodium channel-blocking characteristics. In addition to enhancing sodium channel blocking, potassium channel blocking may directly contribute to arrhythmogenesis both by increasing AP duration and by increasing membrane resistance. Lidocaine, which shortens rather than lengthens AP duration in heart [16], may be less effective in blocking potassium channels than bupivacaine [9]. An important factor in bupivacaine cardiotoxicity may thus be its action on cardiac potassium channels.

TABLE I

BLOCKING ACTIONS OF 20 μM BUPIVACAINE ON THREE IONIC CURRENTS IN BULLFROG ATRIAL CELLS

I_{K1} was evoked by steps to -90 or -100 mV and I_K by steps to $+30$ or $+40$ mV from a holding potential of -60 mV in the presence of TTX and $CdCl_2$; I_{Na} was measured as either $\dot{V}_{max}$ or as peak current evoked by a step to -10 mV from a holding potential of -80 mV (TTX omitted).

Current	% decrease (mean ± S.E.)	N
I_{K1} (background)	8.7 ± 1.3	7
I_K (time dependent)	16.4 ± 1.3	5
I_{Na}	20.7 ± 2.0	4

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