TITLE: EDROPHONIUM ALTERS THE PROPERTIES OF ION CHANNELS ACTIVATED BY ACETYLCHOLINE

AUTHOR: Ruth E. Wachtel, Ph.D.

AFFILIATION: Department of Anesthesia, College of Medicine, University of Iowa, Iowa City, IA 52242

Introduction. Edrophonium is frequently used to antagonize neuromuscular block produced by non-depolarizing agents such as curare. The actions of edrophonium are thought to result from inhibition of acetycholinesterase, the enzyme responsible for inactivation of acetylcholine (ACh) at cholinergic synapses. However, not all effects of edrophonium at the endplate can be explained solely on the basis of inhibition of cholinesterase.[1]

These experiments were undertaken to determine whether edrophonium might also act at other sites associated with the neuromuscular junction, such as postsynaptic ion channels activated by ACh.

Methods Patch clamp techniques were used to record single channel currents activated by ACh from BC_3H1 cells grown in culture. The recording electrode contained a modified Ringer solution and ACh (200-250nM), with or without edrophonium (0.5-10 μM).

In patch clamping, an electrode with a tip opening of about 2μ is pressed against a cell, resulting in formation of an extremely high-resistance seal between electrode and membrane. ACh that has been placed in the electrode will activate ion channels in the membrane patch underneath the electrode, resulting in current flow across the cell membrane. Patch clamping techniques allow measurement of ionic currents produced by the opening of single ion channels.

Several hundred channel openings were recorded from each membrane patch studied, and openings were analyzed to determine amplitude, open duration, and closed time (interval between events). Events were recorded with the membrane patch hyperpolarized +75 mV relative to cell resting potential.

Results Fig 1 shows sample recordings of single channel currents activated by ACh in the absence and presence of edrophonium. Channel openings appear as downward deflections of the trace.

With ACh alone in the recording electrode, channel openings resembled square pulses of inward current. In the presence of edrophonium, openings were much shorter in duration (see Table 1). In addition, successive openings were sometimes separated by very brief closures, and channel openings often appeared to occur in bursts. The burst shown in Fig 1 is an exceptionally long one containing 11 opening events, although some of the closures were so brief that the signal did not fully return to baseline levels during the closure. Average channel amplitude was unchanged by edrophonium.

Discussion These results demonstrate that edrophonium alters the kinetics of ion channels activated by ACh, and does so at therapeutic concentrations. Following a 2 minute intravenous infusion of 0.5 mg/kg, peak serum levels average about 5,000 ng/ml, or 25 μM.[2] In these experiments, profound alterations in channel properties were observed at 10 μM edrophonium. Such alterations in channel properties may help to account for some of the clinical effects of edrophonium at the neuromuscular junction.

References
1. Standaert, p. 861 in Miller, Anesthesia, 1986.
2. Morris et al, Anesthesiology, 54:399, 1981.

Figure 1

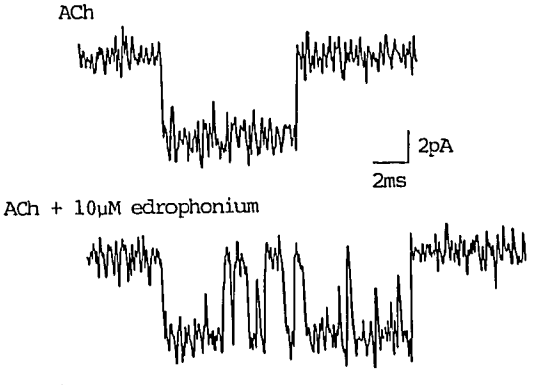

Table 1

	ACh	ACh + edrophonium
open time (ms)	6.32 ± .83	1.89 ± .49
openings per burst	1.10 ± .05	2.31 ± .36
amplitude (pA)	2.70 ± .16	2.61 ± .24

Each value is the mean±S.D. from 10-24 patches.