potassium. Both cæsium and choline have been shown to enter the cell under our experimental conditions[1,3].

Are the postulated receptors for choline identical with the widely accepted acetylcholine receptors implicated in motor end-plate function[4,5]? Much more work is needed to clarify this situation, but the fact that excess external potassium results in liberation of acetylcholine from nerve endings is suggestive[6,7]. However, the apparent independence of the cæsium-choline depolarization from external sodium concentration, at least over the range 56–112 mM sodium chloride[1], is puzzling, since it prevents us from explaining the measured potential decrease as a result simply of increased sodium permeability. Sensitivity to acetylcholine is normally restricted to the motor end-plate region, but spreads over the whole muscle surface after denervation[8,9]. This spreading out process does not begin for at least 8 h after denervation, however (15 h under our laboratory conditions), so it is not likely to be involved in these choline effects. The scanning results indicate that the ability to react with choline is distributed over the entire muscle cell surface, including the motor end-plate region, but not limited to it.

This work was supported in part by grants from Consejo Nacional de Investigaciones Cientificas y Tecnicas, Argentina, the U.S. Public Health service, *AM*-07509, and U.S. Department of Defense, *DA-ARO*-49-092-64-*G*31.

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Lack of Artificial Acclimatization to Heat in Physically Fit Subjects

DURING investigations of artificial acclimatization to heat on three physically fit subjects, the usual changes in sweat rates, rectal temperatures and pulse rates were not observed. The acclimatization procedure consisted of five 2-h walks on an every-other-day sequence. The daily protocol was 50 min walking at 6·4 km/h on a level treadmill and 10 min rest each hour. The environmental conditions were dry bulb weight = 49° C, wet bulb weight = 28° C, and globe temperature = 49° C. Comparing the results of walk 1 with walk 5, the average pulse rate decreased 1 beat/min; the sweat rate decreased 66 g/h, and the rectal temperature decreased 0·3° C.

Other workers have shown that the major physiological changes occur after four or five exposures[1–3] and the optimal exposure time was 100 min[4]. It was hypothesized that the physiological changes induced in the subjects as a result of their high level of physical fitness precluded any heat effects. Thus the subjects were able to perform as though they were acclimatized to heat.

The question then arises when conducting heat acclimatization investigations that utilize physical exercise: which changes are due to environmental stimuli and which are due to the exercise? A further complication is that, since exercise also produces heat, what is the relationship between exercise heat and environmental heat on the response of the subject. Fox *et al.*[5] have recognized the possible separate effects of environment and exercise and commented that "a part of the large decrease in pulse rates normally observed when using the conventional technique (of heat acclimatization) may be due to the improved cardiovascular efficiency consequent on the repeated performance of a particular physical exercise. . . ."

In understanding the mechanisms of heat acclimatization it seems imperative that cognizance be taken of the level of physical fitness of the subjects as well as the relative contributions of the separate effects of the particular exercise and environment.

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Direct Evidence for an Action of Acetylcholine on Motor-nerve Terminals

SOME twenty-five years ago Masland and Wigton[1] showed that antidromic action potentials could be recorded from the ventral roots of cats during the muscular contraction provoked by the intra-arterial injection of acetylcholine. As a result of these experiments they suggested that acetylcholine might have an excitant effect on motor-nerve terminals. Later work[2] has shown that many other quaternary ammonium compounds have the same ability to provoke antidromic action potentials in motor nerves. In particular Werner[2] has shown, by ingenious but indirect means, that these drug effects are not due to excitation of the terminals by synchronous muscle action potentials. Despite these experiments which seem most satisfactorily explained by the presence of some cholinoceptive sites on the nerve terminals[3], it has recently been claimed that at the present time there is no direct evidence for an action of acetylcholine on motor-nerve terminals[4]. This problem is of special interest, for an excitant action of the acetylcholine released by nerve impulses on nerve terminals is the central feature of a new hypothesis of transmitter release at ganglionic and neuromuscular junctions put forward by Koelle[5]. This excitant action is thought to release further acetylcholine in a sort of positive feed-back action.

In the investigation recorded here the effect of muscle activity was eliminated by using rat diaphragm-phrenic nerve preparations *in vitro*, paralysed with either *d*-tubocurarine (Burroughs Wellcome) or an excess of magnesium chloride in the bathing Krebs-Ringer solution. The problem of acetylcholine action was attacked directly, using the methods developed by Hubbard and Schmidt[6] for stimulating nerve terminals. As in that investigation the terminals were located by the preliminary recording of extracellular miniature end-plate potentials in magnesium-poisoned preparations or by the recording of focal end-plate potentials in curarized preparations. The recording microelectrodes were filled with 4 M sodium chloride and had 1·5–3μ tips and 1–3 MΩ resistance. Stimulating pulses of 0·1 msec duration from a Grass isolation unit were then applied through the same electrodes and the single all-or-nothing antidromic impulses set up by the stimulation were recorded in the phrenic