EFFECT OF VARYING CONCENTRATIONS OF SODIUM ON ISOLATED MAMMALIAN HEART

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Experiments have been carried out on isolated rabbit hearts to study the role of extra-cellular sodium in the normal functioning of the heart. Modified Ringer-Locke solutions of varying NaCl content were used for perfusion and changes in the mechanical response were recorded. The isotonicity of the fluids was maintained by the addition of suitable amounts of sucrose.

It has been found that reducing the NaCl concentration down to 0.4% produces a marked and sustained increase in the force of contraction, without any change in rate or rhythm. Further reduction, below 0.4%, gives a biphasic response—initial stimulation followed by marked inhibition and cardiac arrest. Irregularities appear and there is a conspicous rise in tone. A heart which has ceased to beat in a low NaCl fluid shows evidence of some irreversible damage. It appears that, to the extent of about 0.4% NaCl, sodium ions are essential for the normal activity of the heart and may be directly concerned with impulse generation, conduction or contraction of the muscle.

Several reports in the past have suggested, directly or indirectly, that lowering the sodium concentration of the fluid medium can have a beneficial effect on the activity of the cardiac muscle (Daly and Clark, 1921 ; McDowall *et al.*, 1955 ; Kahali and Kothari, 1960). The present work has been undertaken to investigate this phenomenon by studying the mechanical response of a spontaneously beating isolated rabbit heart at different levels of sodium concentration in the perfusion fluid. The problem is conceived in the wider context of elucidating the role of sodium ions in the normal functioning of the mammalian heart.

METHODS

Experiments have been carried out on isolated hearts of adult healthy rabbits. The usual Martin-Langendorff technique of coronary perfusion was used with some minor modifications to suit the present problem. Six perfusion bottles, all raised to the same height, were employed instead of one. They were all connected to a glass spiral tube kept submerged in a large thermo-regulated water-bath. The spiral tube led down into the perfusion cannula which had 3 side-tubes for, (i) insertion of a thermometer, (ii) connection to a mercury manometer and (iii) for draining out the fluid when desired.

By means of suitably placed clamps, any one perfusion fluid could be allowed into the cannula without any intermixing of fluids or change of temperature and pressure.

Ringer-Locke solution was used as the basic perfusion fluid. A series of 9 modified fluids was prepared, such that, the NaCl content diminished progressively from 0.8 per cent to 0.0 per cent, in steps of 0.1. Each of these was made isotonic by the addition of osmo-equivalent amount of sucrose, calculated on the basis of the molecular weights of sucrose and NaCl (1 gm NaCl= 11.69 gm sucrose). The other constituents remained unaltered. The effect of any one of the five fluids could be studied, one after the other, at any one time; the remaining perfusion bottle was kept reserved for normal Ringer. Oxygen was kept bubbling through all the fluids.

Contractions were recorded on a slow moving kymograph by means of a Starling's heart-lever. Where rate had to be determined, a faster kymograph speed was used.

RESULTS

On changing from normal Ringer to a fluid of low NaCl content, there was a substantial increase in the amplitude of contraction in all cases. The lower the NaCl concentration of the fluid, more marked was this effect. The maximum augmentation recorded at various NaCl levels is shown in Table I.

TABLE 1

NaCl content of perfusion fluid, g/100 ml	Conc. of Na ions, mM/litre	Max. increase in amplitude, % of normal
0.9	153.8	——
0.8	136.7	60
0.7	119.6	68
0 6	102.5	75
0.5	85.4	100
0.4	68.3	140
0.3	51.2	190
0.2	34.1	350
0.1	17.0	450
0.0	0.0	800

The small amount of Na ions contributed by $NaHCO_3$ (0.015 %) was considered too small to be taken into account

Conversely, a change from low NaCl concentration to a higher one always produced an inhibitory effect and the contractions diminished.

Within the limits of this general observation it was, however, found that the pattern of response seen in the range of 0.9 to 0.4 per cent NaCl was significantly different from that observed at lower concentrations.

NaCl concentration between 0.9 – 0.4 per cent.—The increase in amplitude was well sustained over long periods of observation. There was no appreciable change in the frequency of the beats. The rhythm remained regular throughout.

NaCl concentration below 0.4 per cent.—The increase in amplitude was even greater but was only transitory. The stimulatory effect soon gave way to inhibition and the heart always came to a premature stand-still. Changes in the rate and rhythm were evident. Three phases could usually be discerned in the events that followed the starting of such a fluid. Firstly, there was marked increase in amplitude with some acceleration but no irregularities. The lower the concentration of NaCl, more pronounced and prolonged was this phase. Gradual fall in amplitude with decline in heart rate and some irregularity in the force and rhythm of the beats was next observed. This phase was quite variable but, in general, lower the concentration of NaCl less pronounced it was. Finally, there was abrupt cessation of beats.

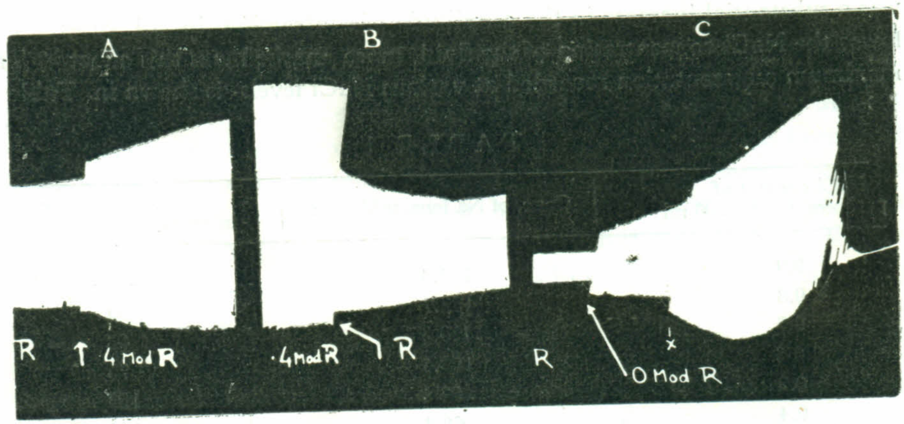

Fig. 1. Kymographic record of perfused rabbit heart showing effects of reduced NaCl concentration—(A) augmentation on changing from normal Ringer to modified Ringer with 0.4% NaCl; (B) sharp decline in amplitude on changing back to normal Ringer; (C) effect of NaCl-free modified Ringer, showing marked initial augmentation followed by inhibition and a rise in base-line. (kymograph stopped at **x**).

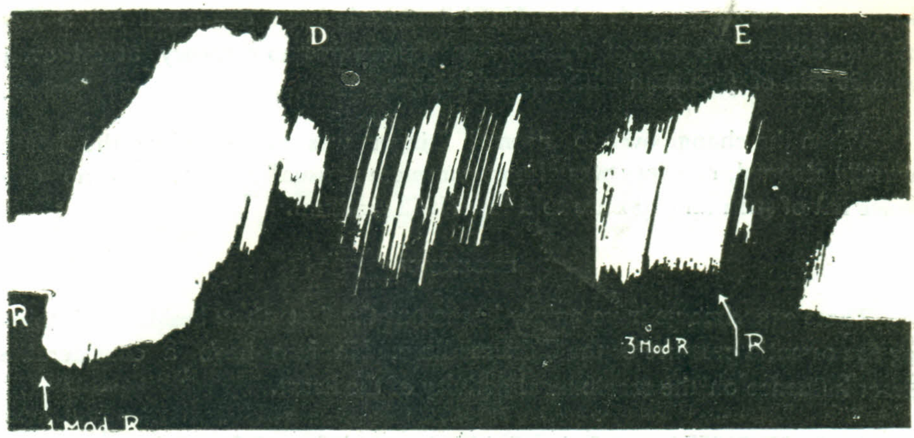

Fig. 2. Kymographic record of perfused rabbit heart. (D) marked increase in amplitude with 0.1% NaCl; the rising base line and sporadic contractions after cardiac arrest are evident; (E) sharp fall in base line and the quiescent pause on changing from 0.3% to normal NaCl concentration.

After the heart stopped beating there was a conspicous rise in the base line on the record. That this denoted an actual increase in tone rather than a mere mechanical effect of the quiescent heart becoming more globular under passive distension, was inferred from the following observations: The rise in the base-line was, usually, quite evident even before the beats ceased or became very feeble. The stopping of the perfusion fluid completely, as soon as the heart-beats ceased, did not prevent the rise in the base-line. On switching again to normal Ringer the base-line began to descend promptly, much before the contractions were resumed.

It was very often found that, when the fluid was continued even after the heart had stopped, short sporadic outbursts of vigorous contractions, with intervals of quiescence, occured several times before all activity finally ceased. Each outburst represented, in miniature, roughly the same 3 phases as described previously. The intervals between such sporadic bouts of activity gradually increased and they wore down till they totally disappeared.

While the heart was still beating in such a low NaCl fluid, a sudden change to normal Ringer stopped the heart completely for some time before the contractions were resumed again. During this pause the base line descended sharply. No definite corelation between the duration of the pause and the difference in NaCl content of the two fluids could be made out.

When perfusion was continued to the point of cardiac arrest, some permanent damage seemed to result. On changing to normal Ringer, recovery was only partial and the contractions never regained their original amplitude. In experiments where such a low NaCl fluid had been continued for a few minutes beyond the time of cardiac arrest, there was no recovery at all even with 30 min of perfusion with normal Ringer.

When the change over to normal Ringer was made before the heart actually stopped, recovery was almost complete ; usually there was no evidence left of such an exposure to a low NaCl medium.

DISCUSSION

These experiments have amply borne out the fact that, besides maintaining the osmotic pressure of the fluid medium, Na ions have a considerable direct influence on the mechanical activity of the heart.

As the NaCl in the perfusion fluid is lowered from 0.9 to 0.4 per cent the amplitude of contraction of the isolated heart increases progressively without any change in rate and rhythm ; most often it more than doubles. The mechanism which links the extracellular Na and the force of contraction of the muscle is as yet obscure and, therefore, it is difficult to say what is the precise cause of this positive inotropic effect. However, two broad possibilities may briefly be mentioned here.

It may be presumed that a reduction in the extracellular Na cuts down the metabolic energy which has normally to be spent for the active extrusion of Na by the cell, as suggested by McDowall, *et al.* (1955). The energy thus saved is diverted to the performance of mechanical work and leads to more powerful contractions.

Luttgau and Niedergerke (1958) have postulated that Ca and Na ions compete, at the cell surface, for some negatively charged receptor substance R, forming either CaR which activates contraction or NaR which is inert. Therefore, a reduction in extracellular Na would produce a relative preponderance of CaR and hence more vigorous contractions. Both these attractive hypotheses, however, involve many theoretical conjectures which are yet to be verified.

When the NaCl concentration is lowered below 0.4 per cent, the response becomes characteristically biphasic. There is, initially, a marked increase in the force of contraction associated with some acceleration. This is soon followed by inhibition, irregularities of rhythm, incomplete relaxation and, finally, cardiac arrest. The cardiac arrest seems to be associated with some irreversible damage to the muscle since there is hardly any recovery even after prolonged perfusion with normal Ringer.

It may be concluded that the presence of Na, to the extent of about 0.4 per cent NaCl (68.3 mM/litre Na) is essential for maintaining the normal performance of the heart. Above this level it somehow checks the force of contraction and its only use may be to make up the isotonicity of the fluid medium.

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