The tracheotomy method of positive pressure anesthesia as described by Farris(3) has limited application. It was designed for use in terminal experiments where the survival of the animal is not necessary.

As an additional aid in the performance of the thoracotomy, a special operating table easily constructed from a standard cigar box is used. The extremities are tied in such a way that the animal is partly suspended and partly supported upon a soft-rubber sponge. This permits more freedom of thoracic movement and easier expansion of the unoperated lung. The use of the rat-board has proved to be more restricting and the results have not been as satisfactory.

Summary. A method of positive pressure anesthesia for the rat using a mask and an abdominal binder is described. It is applicable to the use of ether, intraperitoneal barbiturates or both as anesthetic agents. A closed system for the administration of the oxygen-ether mixture is also illustrated.

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Occurrence of a Sodium-Potassium Antagonism in Nerve Block.* (19478)

FREDERICK CRESCITELLI. (With the technical assistance of Robert J. Dellenback.)

From the Department of Zoology, University of California, Los Angeles.

It is recognized that sodium ions are required for activity of nerve fibers(1-6). It is also well known that potassium ions, in concentrations above normal, cause a depolarization of the nerve membrane, a decrease in excitability and a block of conduction(4, 7-9). A long series of experiments has culminated in the theory that entrance of sodium during the period of the rising phase of the action potential spike, and the exit of potassium during the falling phase, are central events in the mechanism of conduction(5). This theory recognizes the important point that the movements of these two ions are not independent and unrelated events but rather, integrated processes which occur in sequence. The sodium and potassium mechanisms thus interlock at some point in the chain of reactions of the nerve impulse. The problem, therefore, is to discover the means whereby such interlocking occurs. This communication will report an interaction between sodium and potassium in the maintenance of conduction in frog nerve fibers which seems to be of more than passing interest. At this stage of development it is only necessary to present a simple and factual report of the results without interpretation.

Procedure. Except for certain essential information, no details of procedure need be given, since these were reported in a previous communication(6). Isolated and desheathed sciatic-peroneal nerves of bullfrogs were laid across stimulating and recording electrodes in a moist lucite box. A 20 mm segment of nerve between the stimulating and the recording electrodes was set within a glass cup. Various test solutions were added to the cup, thus exposing the segment to the solutions. Phosphate Ringer's solution at a pH of 7.0-7.3 was used. This contained 0.11 M NaCl and 0.0018 M KCl in addition to the other usual components of frog Ringer's solution. Potassium enriched solutions, when required, were made by adding crystalline KCl to unaltered Ringer's fluid. As Shanes(10) has pointed out, this procedure is not only permissible but is requisite in a system which is

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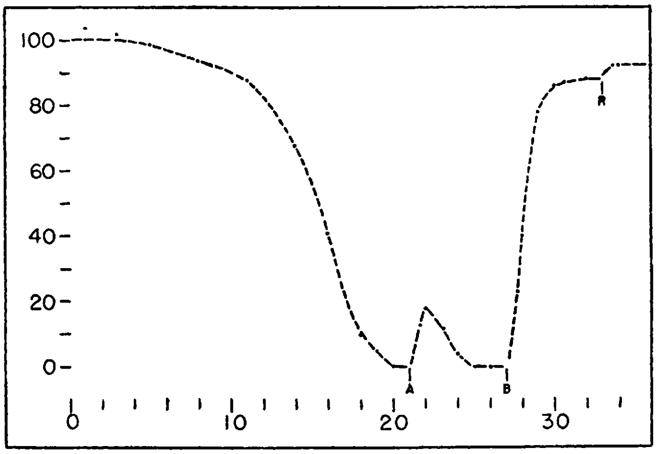

FIG. 1. The action of potassium ions on recovery of conduction at low, and at normal sodium ion concentrations. In all 3 figures the height of the compound A spike, in % of normal, is plotted against time, in min. At zero time the nerve segment was treated with .11 M tetramethyl ammonium bromide with no added NaCl. Block occurred, and at 21 min (A) a solution containing .0085 M NaCl and .0108 M KCl was added in place of the low sodium solution. At 27 min (B) a solution containing .11 M NaCl and .0108 M KCl replaced the previous solution. Ringer's solution with the normal concentration (.0018 M) of KCl was added at 33 min (R).

relatively impermeable to sodium and permeable to potassium and chloride. Sodium deficient solutions were prepared by osmotic replacement of the NaCl with compounds such as tetramethyl ammonium bromide or choline chloride. As already indicated(6) these compounds may be considered to be inert replacements for the NaCl. The significant experimental procedure involved the recording of the effect on conduction in the A fibers, of solutions with different potassium and sodium concentrations. Conduction in the A group of fibers was studied by oscillographic recording of the compound A spike in the nerve region beyond the treated segment.

The *results* are understandable by reference to the accompanying three figures and their legends. The nerve fibers were first blocked by a 0.11 M tetramethyl ammonium bromide solution containing no added NaCl. Except for Fig. 1, the time course of this low sodium block is not shown because this was described elsewhere(6). In addition to the block, Fig. 1 illustrates successively the effects of adding 2 solutions each with 6 times the normal concentration of KCl, the first solution (added at A) containing 0.0085 M NaCl, and the second solution (added at B) containing 0.11 M NaCl. This experiment brings out the point that a solution with potassium ions at 6 times the normal level was able to restore and to maintain conduction with the

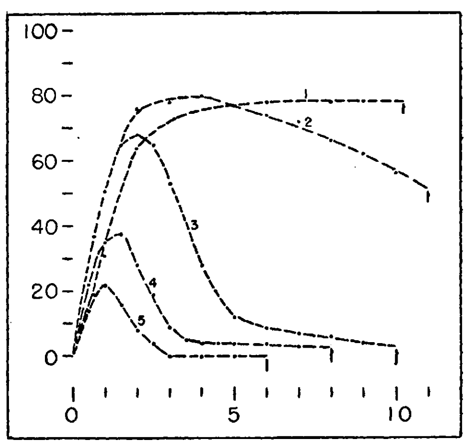

FIG. 2. Influence of potassium ions on recovery from low sodium block. Recovery was followed with solutions containing .0085 M NaCl and with varying concentrations of KCl. Each solution was added one min after completion of low sodium block. KCl concentrations were: .0036 M (1), .0054 M (2), .0072 M (3), .009 M (4) and .0108 M (5). To avoid complicating the figure the return of the A potential after the addition of Ringer's solution at the times indicated by vertical lines has not been drawn in.

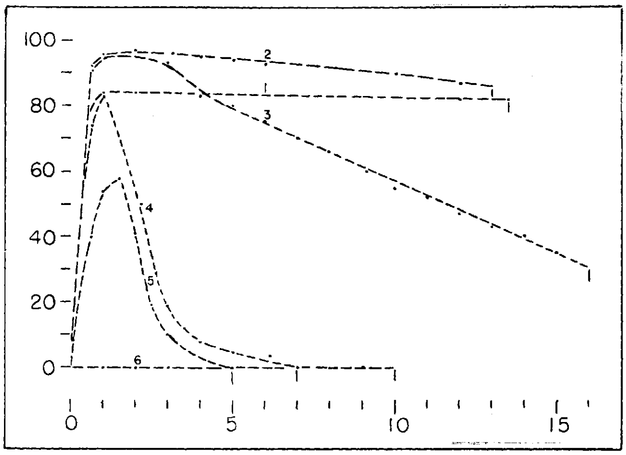

FIG. 3. Action of potassium ions in presence of varying concentrations of sodium. Recovery from low sodium block was followed with solutions containing .009 M KCl in the presence of NaCl at the following concentrations: .11 M (1), .066 M (2), .044 M (3), .022 M (4), .011 M (5) and .0055 M (6). Not drawn is the recovery which followed the addition of Ringer's solution at the times indicated by the vertical lines.

higher sodium concentration but not with the lower sodium concentration. It is known already(6) that a solution with 0.0085 M NaCl in the presence of normal potassium is able to maintain activity in many of the A fibers. This finding is supported by the graphical data of Fig. 2.

The second figure shows the course of recovery from low sodium block by means of different solutions all containing 0.0085 M NaCl but with varying potassium chloride concentrations. It is clear that though the 0.0085 M concentration of NaCl was able to effect recovery and to maintain conduction in the presence of 0.0036 M KCl, it was less able to do so, the higher the potassium concentration. Fig. 3 illustrates the converse effect, that is the action of different solutions with varying sodium concentrations and with one level of potassium chloride, *i.e.*, 0.009 M. This experiment shows that a concentration of 0.11 M NaCl was sufficient to antagonize the action of 0.009 M KCl so that recovery occurred and no secondary block developed, at least within the period of the experiment. However, lower concentrations of NaCl were unable to maintain activity in the presence of 0.009 M KCl.

The results of these experiments reveal the existence of a marked antagonism between these physiologically significant ions. Relatively small changes in potassium concentration were able to modulate the action of sodium. In addition to the primary problem of the mechanism of this antagonism, these results raise the interesting possibility that the sodium-potassium antagonism may be the basis for a number of physiological properties of nerve activity which are as yet unexplained. The refractory period, for example, is one such property. If sodium action is required for spike production, and if potassium leaves the nerve fiber at an accelerated rate during the falling phase of the spike, then the momentary rise in external potassium in the neighborhood of the fiber after an impulse may lead to a momentary antagonism to sodium and be the cause of refractoriness.

Summary. The conduction block in bullfrog A fibers which is produced by the presence of elevated potassium ions is dependent on the sodium concentration. At a fixed level of potassium the rate of block increases as the sodium concentration is decreased. This occurs at concentrations of sodium which, at normal potassium, have no blocking action.

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Influence of Hemagglutinating Viruses on Tumor Cell Suspensions: I. Growth Inhibition and Reversal of the Effect.* (19479)

ALICE E. MOORE AND LEILA C. DIAMOND.

From the Sloan-Kettering Institute for Cancer Research of the Memorial Cancer Center, New York.

In the course of experiments with mixtures of Newcastle Disease (NDV) virus and suspensions of sarcoma 180 cells it was found that after being kept at 4°C, the tumor cells failed to grow when inoculated subcutaneously into mice. This result was accompanied by a fall in hemagglutination titer of the virus. When similar mixtures were kept at 4°C for 10 minutes and subsequently at room temperature for 1, 2, or 3 hours, subcutaneous inoculation resulted in tumor growth. This was accompanied in most instances by the reappearance of hemagglutinating activity of the virus. The similarity of these findings to those observed when hemagglutinating viruses are brought in contact with red cells (1) has led to further experiments to determine to what extent the parallelism of the phenomenon holds. Most have been conducted with the NDV and the sarcoma 180 suspensions, but other tumor-virus systems have been used with success. The preliminary results are herewith reported.

Materials and methods. Viruses. The Massachusetts strain of NDV† was used in most of the experiments. In a few, however, the PR8† strain of influenza and the WS† strain of neuro-influenza were employed. All were used as pools of allantoic fluids. *Tumor suspensions.* Animals bearing 7-day-old sarcoma 180 were sacrificed, their tumors removed and made into a suspension by pushing them through a fine 40 mesh monel metal screen with a pestle. The suspensions were made to 10% in Locke-Ringer buffered with phosphate to make the pH 7.2. Glucose was added to make 600 mg %. The suspensions consisted of single cells, clumps of 8-10 cells and larger clumps up to the size of the apertures of the sieve. After the larger clumps had been permitted to settle a fairly uniform suspension was obtained. Both settled and unsettled suspensions were used in the experiment. Suspensions of Erhlich carcinoma were made in the same way.

Experimental procedure. In most experiments 10% sarcoma 180 and virus were mixed at the desired temperature. At different time intervals 0.5 cc of tumor-virus mixture was inoculated subcutaneously in each flank of Carworth Farm white mice. A readily palpable and visible bleb formed which gradually disappeared in about 1 hour. When tumor grew it assumed the form of the bleb. The mice were examined at weekly intervals and tracings of their tumors made. They were kept for 1 month after which they were autopsied to ascertain the presence of internal tumors. The results are expressed as per-

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