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MAINTENANCE OF RESTING MEMBRANE POTENTIALS IN SLICES OF MAMMALIAN CEREBRAL CORTEX AND OTHER TISSUES *IN VITRO*

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Investigation of the mammalian brain would be greatly furthered if portions from it could be studied by electrophysiological methods while they were completely isolated from the body, as the peripheral nerves and spinal ganglia can be studied. Spontaneous potentials were observed in an isolated portion of the frog brain by Libet & Gerard (1939). Burns (1950, 1951) made observations on responses to electrical stimulation of cortical slabs of the cat which were connected to the brain only through pial vessels.

With completely isolated mammalian cerebral tissue adequate supply of metabolites for maintenance requires sections less than 0·35 mm in thickness, and electrical activity in such sections has not previously been recorded. It could be pictured as lost through damage to cells in preparation of the sections. However, such tissue under good metabolic conditions has proved to be remarkably autonomous, and has also shown metabolic responses to applied electrical impulses (McIlwain, 1951, 1956). With a variety of recording electrodes (but not intracellular electrodes), transmission of impulses could not be detected in such tissue. A reasonable explanation of the metabolic response is that polarized cell elements were maintained in the tissue and depolarized by the applied impulses. In this series of experiments evidence was sought for such polarized intracortical elements with the use of micropipette electrodes. Most observations were made on the cerebral cortex of guinea-pigs and cats. Slices from the liver, kidney and skeletal muscles were also examined.

METHODS

Tissue slices. The guinea-pigs were stunned by a blow on the neck and bled by cutting the neck. The cats were anaesthetized with intraperitoneal thiopental sodium. In each case, the scalp and calvarium were removed and the dura reflected. The brain was lifted with a spatula and placed on a filter paper which had been soaked in the experimental saline. Slices about 10 × 8 mm and

0·35 mm thick were cut from the outer convexity of the cerebral hemispheres with a strip of razor blade and a template. In some instances two or three successive slices were cut at increasing depths in the cortex.

After samples of brain tissues had been taken, a kidney and a lobe of liver were removed and sliced in the same way. The first slice from these organs was discarded in order to avoid fibrous membrane, and the second slice, consisting of tissue 0·35–0·7 mm from the surface, was taken for examination. Muscle samples were obtained from the gracilis anticus. The muscles were dissected and freed from fibrous tissue while the blood circulation of the animal was intact. They were then cut at the pelvic attachments and distal insertions.

The tissue slices were floated from the cutting blade or guide to a dish containing about 10 ml. of an oxygenated saline which contained glucose. The salines were based on those of Krebs as described by McIlwain (1951) and Rodnight & McIlwain (1954). A bicarbonate saline containing (mM) NaCl 124, KCl 5, KH_2PO_4 1·24, $MgSO_4$ 1·3, $CaCl_2$ 2·8, $NaHCO_3$ 26 and glucose 10 was used most frequently. The tissue was picked up from the dish by the glass-fibre grid of the slice chamber, described below. The grid with its tissue slices was placed in the chamber 2–3 min after the death of the animal.

Slice chamber. The chamber consisted of a glass vessel in the form of an inverted cone, with a removable grid (Fig. 1). Its design was based on metabolic data which indicated that slices of mammalian cerebral cortex could retain their metabolic response to applied electrical impulses when they had been held on the surface of a nutrient saline (McIlwain, Ochs & Gerard, 1952; Rodnight & McIlwain, 1954). In distinction from a previous model (McIlwain *et al.* 1952), the present chamber was designed to maintain the tissue-weight/fluid-volume relationships of typical metabolic experiments, by employing 30–100 mg of tissue with 3–10 ml. of saline. At the apex of the vessel were two openings, one connected to a 10 ml. syringe through flexible tubing, the other for the introduction of a Ag–AgCl wire which served as an indifferent electrode. In the wall of the glass vessel was an opening for passage of O_2, CO_2, or N_2 as desired. The glass vessel was covered by a transparent plastic lid so that the tissue could be studied in an atmosphere of pure or mixed gases. During experiments with N_2 or CO_2 5% (v/v) in N_2 no attempt was made to maintain strictly anaerobic conditions. The chosen gases were bubbled through the salines at the commencement of the experiments, and during incubation a good flow of the gas was maintained through the slice-chamber, so that the gas leaving through the holes in the lid of the chamber minimized inward diffusion of air. The recording micro-electrode and stimulating electrodes were passed through the holes in the lid into the chamber.

The removable grid in the slice chamber was made of glass fibres (about 0·2 mm diam.) cemented to a plastic ring 2·5 cm diam., shaped to fit the conical sides of the glass vessel. The apex of the glass vessel was immersed in a beaker of water. The beaker, provided with an inlet and outlet for the circulation of the water, was held between the claw feet of a binocular microscope. The slice chamber rested on supports from the microscope stage. The water in the beaker flowed, through a centrifugal pump, from a large tank equipped with a thermostat which kept the water at 39° C, and back into the tank. The tank was placed below the level of the beaker, and contained a vessel of saline, through which the oxygen or other gas supplied to the slice chamber was bubbled.

With the thermostat and pump running, the experimental saline was prepared and about 8 ml. introduced into the chamber by a syringe. (It was found in preliminary experiments that, when the water of the thermostat tank was kept at 39° C, the temperature of the saline in the slice chamber was 37·5° C; this was checked occasionally during experiments by replacing one of the electrodes by a thermometer.) The saline was not circulated continuously but was moved occasionally by the syringe, which was also used to withdraw and replace saline and to mix it after additions had been made. A flow of the moist gas was maintained with the lid of the slice-chamber in position but without the glass fibre grid. Phosphate saline was equilibrated with oxygen and bicarbonate salines with CO_2 5% (v/v) in O_2. A lamp just above the transparent lid illuminated the slice chamber and also warmed the lid sufficiently to prevent condensation of moisture on the under surface of the lid.

Tissue was then prepared and transferred on the grid to the slice chamber, where it was totally immersed in saline during a preliminary incubation period of 10–15 min. Two or three slices totalling some 2 cm² in area were employed in most experiments. After the preliminary incubation, part of the saline was withdrawn by the syringe, leaving the tissue on the grid. While the saline was being withdrawn, the tissue was guided into place with a spatula or fine brush to prevent folding or overlapping. Stimulating and recording electrodes were then lowered through holes in the lid. Opportunity was taken at approximately 30 min intervals to flood the surface of the tissues with saline to a depth of 1–2 mm. When it was necessary to observe the entry of an electrode into the tissue, the saline was withdrawn.

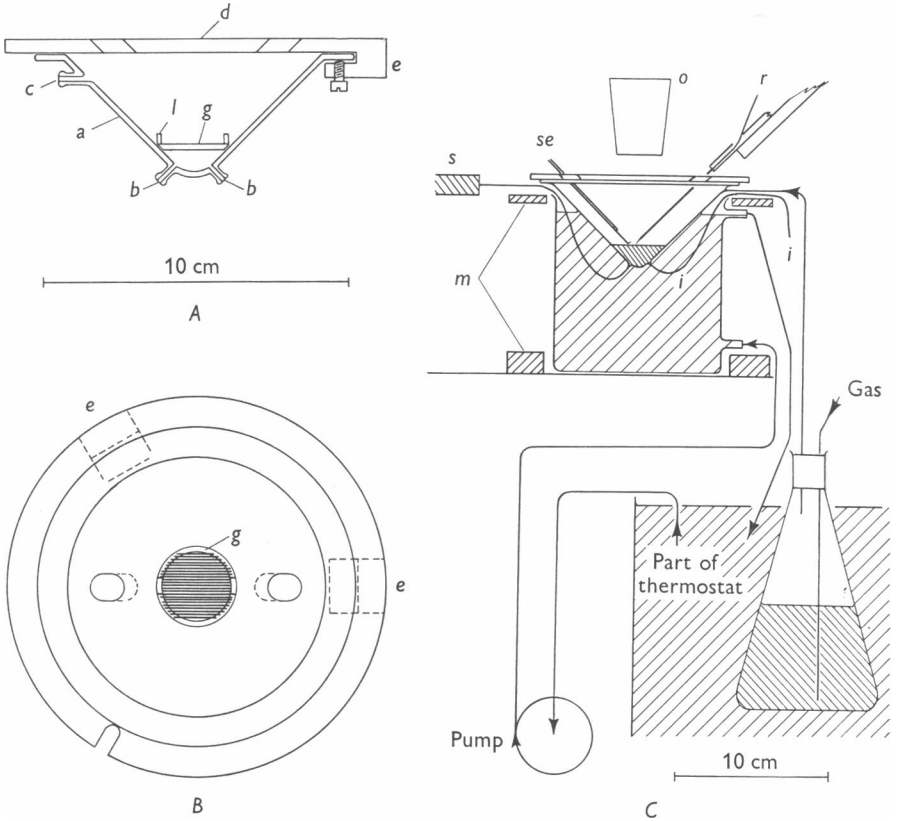

Fig. 1. Slice chamber: *A*, elevation; *B*, plan; *C*, assembled apparatus (semidiagrammatic).

The conical vessel *a* is of glass blown with two openings *b*, *b*, one for fluids the other for the indifferent electrode; and a third opening, *c*, for the passage of gas. Its upper surface is ground flat and on it rests a lid *d* of transparent plastic, held in position with two clips *e*; a third clip, removable, may be inserted in a slot in the lid. Through the lid two holes are drilled obliquely, directed towards the apex of the conical vessel. At the apex rests the grid *g* of glass fibres cemented to plastic ring. The grid is provided with lugs *l* by which it can be removed; it carries the slices.

In *C* the slice chamber is seen supported on a microscope stage (*m*, the feet and stage of the microscope) in a beaker of water supplied by a pump from a thermostat tank. The fluid in the chamber is adjusted by a syringe *s*, and contains the indifferent electrode *i*; the recording electrode *r* and stimulating electrodes *se* are seen on the slice; *o*, microscope objective.

Recording of steady potentials. For recording, glass micropipettes measuring less than 0·5 μ in outside diameter and filled with 3M-KCl solution were used. The d.c. resistance of these electrodes ranged between 10 and 30 MΩ. The micro-electrode was connected to the input grid of a cathode follower, which in turn was connected to a direct-coupled amplifier. A voltage-calibration signal was sent through the indifferent electrode, which was introduced into the flexible tubing connected to the apex of the slice chamber (or was attached to the temporalis muscles in the experiments *in vivo*, see below). The voltage calibration signal served also to indicate the resistance of the micro-electrode when a shunt resistor was interposed between the reference electrode and the input grid of the cathode-follower (Frank & Fuortes, 1955). The micro-electrode was attached to a micro-manipulator, and the initial contact of the micro-electrode tip with the tissue was observed under a binocular microscope. All the records were photographed with continuous moving film. Penetration of a polarized element was signalled by a sudden shift of the recorded potential to negativity. Junctional potentials, developed at the tip of the micro-electrode (del Castillo & Katz, 1955: Adrian, 1956), might have occurred after the electrode had penetrated the tissue. These potentials were not measured; but since they were usually accompanied by a change in the resistance of the micro-electrode (Li, Shy & Wells, 1957), records which showed change in micro-electrode resistance were discarded. Upon withdrawal of the micro-electrode, the steady potential returned to its original value, but if there was a difference which exceeded 5 mV the record was discarded.

Steady potential measurements from cortex of the guinea-pig and cat in vivo. To appraise results with separated tissue, the following experiments were performed. The guinea-pigs were initially anaesthetized with ether. The trachea and the right jugular vein were cannulated and the animal was maintained at a surgical anaesthetic level with intermittent injections of thiopental sodium. The tracheal cannula was attached to a plexiglass chamber equipped with a flutter valve (Li *et al.* 1957). After 20 min of positive-pressure artificial respiration with pure oxygen, continuous flow of oxygen into the plexiglass chamber was established and the animal became unconscious without further administration of thiopental sodium. No respiratory movement of the animal was observed, yet there was no change in cardiac rhythm. Elimination of respiratory movement was found to be essential in recording potentials from cortical units with an intracellular micro-electrode, since any movement in reference to the recording electrode would rapidly cause an irreparable damage to the unit, especially when the cortical units were small. After this preparation the cortex of one hemisphere was exposed and the animal fixed on the mechanical stage of a binocular microscope. Recording of electrical potentials from the guinea-pig cortex was carried out in a manner similar to that for isolated brain slices. Measurements from the cat were obtained in experiments designed for other purposes.

RESULTS

Resting membrane potentials of liver, kidney and muscle incubated in the slice chamber

With a slow insertion of the micro-electrode into a liver slice a steady negative potential was recorded. The potential remained stable during the travel of the micro-electrode for some tens of microns, then suddenly returned to the original value. As the micro-electrode was further advanced, a negative potential of similar magnitude was again recorded. The number of potential changes encountered on a single penetration of the micro-electrode was between 3 and 6. If the micro-electrode was slowly withdrawn a similar phenomenon was observed. In no instance was there a spike potential characteristic of injury discharges commonly seen in experiments with excitable tissues. The measurements thus recorded are shown in Figs. 2*A* and *B*; cat and guinea-pig liver gave similar values.

When the liver slices were incubated in N_2–CO_2 instead of O_2–CO_2, the steady potential rapidly dropped to half of its original size. If O_2–CO_2 was readministered after the tissue had been in N_2–CO_2 from 10 to 20 min, the steady potential rose again. The response of the tissue to N_2–CO_2 is shown in Fig. 2.

Slices of kidney cortex also showed steady potentials which were negative in relation to the saline environment. Frequently the penetrating micro-electrode registered a steady potential for the greater part of its travel through the slice. Further, the tissue could not be satisfactorily explored to a depth of

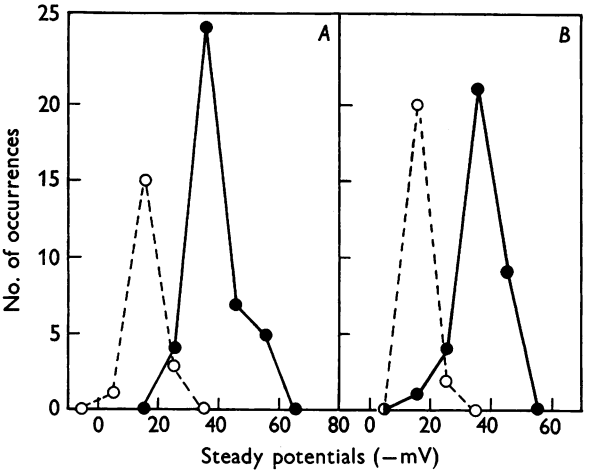

Fig. 2. Frequency polygons of steady potentials recorded from liver slices of guinea-pig (*A*) and cat (*B*). Values joined by dotted lines were obtained when the slices were incubated in N_2–CO_2 for 12–20 min and those by solid lines were obtained after O_2–CO_2 was readministered. From two guinea-pigs, four slices subjected to N_2–CO_2 showed steady potentials ranging from −7 to −21 mv with a mean value of -15 ± 4 (S.D.); in O_2–CO_2 they measured between −27 and −45 with a mean of -36 ± 6 mV. From two cats, four slices gave 15 ± 1 mV (from −10 to −25) and -34 ± 5 mV (from −15 to −45) respectively.

more than 50 or 100μ because at these depths changes in resistance of the micro-electrode tended to occur, and the magnitude of the negative potential increased progressively with the increase in extent and speed of the advance of the electrode. It was concluded that the micro-electrode had encountered an obstacle, such as fibrous tissue, since in these instances the micro-electrode frequently broke with a sudden decrease in its resistance to less than 1 MΩ. The possibility of junctional potentials (del Castillo & Katz, 1955; Adrian, 1956; Li *et al.* 1957) at the micro-electrode tips was also considered and these measurements were discarded. The values of steady negative potentials recorded with micro-electrodes of unvaried resistance are shown in Fig. 3.

From the gracilis anticus muscle of the guinea-pig steady potentials were recorded, occasionally with injury discharges which were not observed in the

liver and kidney. Stimulation of the motor nerve evoked action potentials which overshot the zero membrane potential by 15–40 mV. The steady potentials measured between −21 and −86 mV with a mean value of -63 ± 3 mV (S.D.), as shown in Fig. 3.

Resting membrane potentials of cerebral cortical tissue

Measurements in vivo. Experiments with cerebral cortex *in vivo* showed a wide distribution in negative steady potentials. Injury discharges were frequently observed. Spontaneous spike activity could be recorded with both extracellular and intracellular electrodes. A typical example of spontaneous spike discharges is illustrated in Fig. 4, which also shows the steady potential recorded from inside a cortical unit.

Of 148 negative steady potentials recorded from the guinea-pig cortex none could be maintained for longer than 1 sec, and on forty-two occasions the onset of the steady potential was associated with rapid, repetitive spike discharges which apparently resulted from mechanical injury to the cell. In the cat, despite visible movement of the brain due to respiration, the steady potentials recorded with an intracellular micro-electrode could be maintained for much longer periods than those recorded from guinea-pigs, and injury discharges were less frequently observed. The short-lasting steady potentials recorded from the guinea-pig cortex are probably due to the small size of the cell bodies. It is known that the size of cortical neurones in small mammals is smaller than that of the larger mammals, although the number of neurones in a unit volume of cortical tissue is greater (Tower, 1954). The steady potential measurements from the cortex of these two species are shown by the frequency polygons of Figs. 5 and 6.

Measurements in vitro. In the experiments with isolated cortical tissue in the slice chamber, similar steady potentials were recorded with the penetrating electrode. When the micro-electrode first entered the tissue surface, as observed under the microscope, little or no change of potential was recorded, but as the electrode reached a depth of from 10 to 200 μ, an abrupt swing of the steady potential toward negativity was seen. If a uniform speed of some 2–10 μ/sec of the penetrating micro-electrode was maintained, the potential remained relatively stable for distances which varied between 2 and 50 μ, then abruptly reverted to its original value. If the micro-electrode was halted while it was registering a negative potential, this in some cases slowly receded. In many instances, little change occurred for as long as 5 min. If the micro-electrode registering a negative potential was gradually withdrawn, the potential at first remained steady and then abruptly receded to its original level. The number of potential changes encountered on a single insertion of the micro-electrode into a slice of cortex was between 1 and 11. In some instances, the electrode appeared to be within regions of negative potential for fully half the

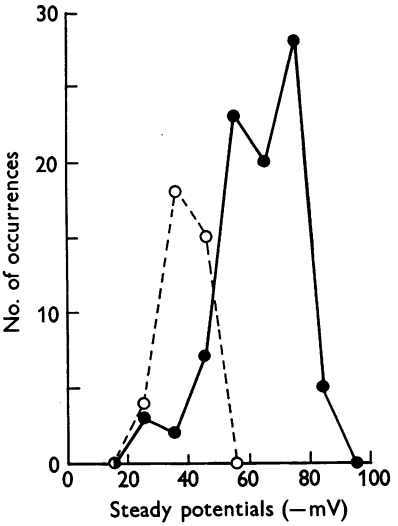

Fig. 3. Frequency polygons of steady potentials recorded from slices of guinea-pig kidney (broken line) and skeletal muscle (solid line), incubated in O_2–CO_2. Three kidney slices from two animals were used; potentials ranged from −21 to −48 with a mean of −35±1 mV. Five muscles from five animals showed potentials ranging from −21 to −86 with a mean of −63±3 mV.

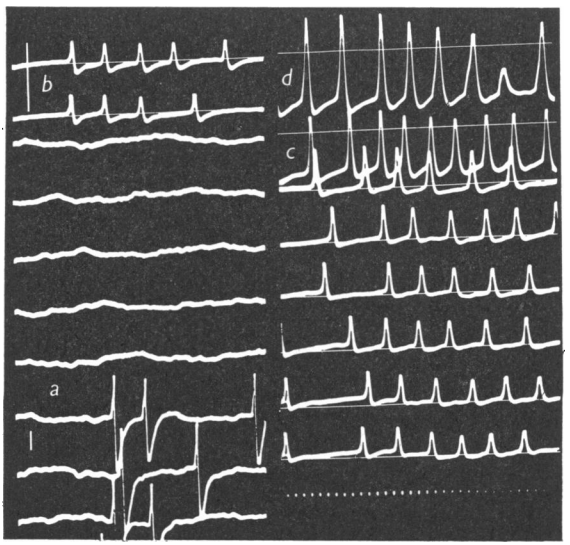

Fig. 4. Spontaneous spike activity and resting membrane potential recorded from a single cortical neurone in the cat approximately 10 hr after intraperitoneal pentothal sodium anaesthesia. *a*, Diphasic spike discharges recorded with micro-electrode outside the cell; *b*, *c* and *d*, as the micro-electrode was further advanced the positive phase of the spikes became larger and as the electrode had entered the cell the negative phase of the spike disappeared, while there was a shift of the steady potential from 0 to −44 mV. Voltage calibration in *a*, 2 mV; in *b*, *c* and *d*, 50 mV; time marker 1 msec. The thin lines in *b*, *c* and *d* represent zero membrane potential. In this and subsequent figures positivity is upwards.

distance travelled. A specific study has not been made of potentials recorded in tissue from different depths of the slice. However, in several instances, the tissue 0·35–0·7 mm beneath the cortical surface was sampled by cutting a second slice after removing the first. The second slice, with two cut surfaces, was also found to maintain potentials of the same order of magnitude as those recorded from the first slice. The steady potentials thus recorded are shown in Figs. 5 and 6.

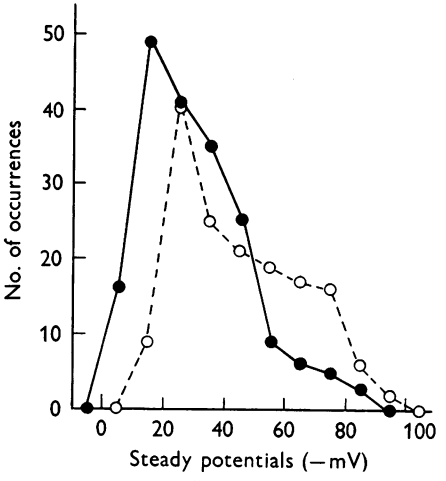

Fig. 5

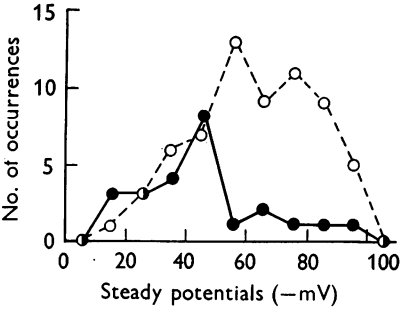

Fig. 6

Fig. 5. Frequency polygons of steady potentials recorded from guinea-pig cortex *in vivo* (two guinea-pigs) represented by dotted line; and *in vitro* (25 slices from fifteen guinea-pigs), by the solid line. The potential measurements *in vivo* ranged from −12 to −95 with a mean of −42±19 mV, while *in vitro* they were between −5 and −93 with a mean of −29±21 mV.

Fig. 6. Frequency polygons of steady potentials recorded from cat cortex *in vivo* (four cats) represented by the dotted line; and *in vitro* (three slices from two cats), by the solid line. The potential measurements *in vivo* ranged from −28 to −90 with a mean of −62±18 mV, while *in vitro* they were between −5 and −96 with a mean of −41±20 mV.

In the control experiments with cortex *in vivo*, spontaneous spike discharges were often recorded with an extracellular or intracellular micro-electrode. With the cortical slices in the chamber no spontaneous spike activity was recorded. However, in 13 out of 305 instances, injury discharges were observed concomitant with the onset of the steady potential. In one experiment they were observed $2\frac{1}{2}$ hr after the slice was removed from the brain of the animal, as shown in Fig. 7.

The injury discharges in response to mechanical irritation of the intracortical elements in the slice by the penetrating electrode prompted attempts at stimulation by application of electrical pulses. This was carried out through two silver electrodes resting on the slice surface adjacent to the entry of the recording micro-electrode. However, no response was elicited by stimuli of

various intensities and frequencies. Application of a slowly rising current also failed to evoke a response.

Metabolic factors. The cortical slices showed no response to electrical stimulation, but they were sensitive to a change in their metabolic conditions. Fifteen or 20 min after the omission of glucose from the saline in which the tissue was incubated, a slight reduction of the steady potential occurred. Addition of glucose to the saline resulted in an increase of the steady potential to its previous magnitude, as shown in Fig. 8. Partial replacement of the

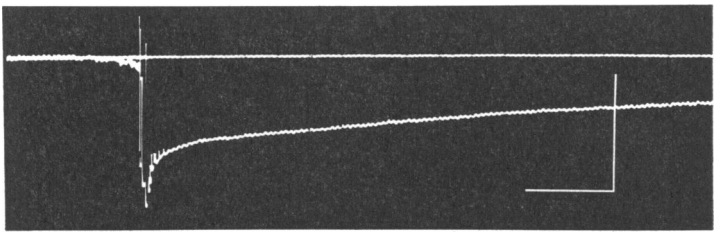

Fig. 7. Resting membrane potential and injury discharges recorded from guinea-pig cortical slice 2½ hr after removal from the brain. Voltage calibration, 50 mV, time 200 msec.

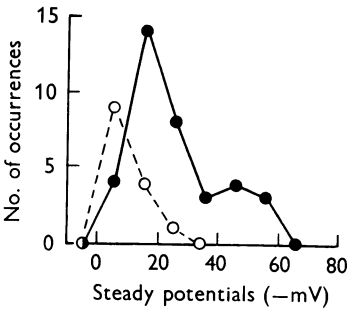

Fig. 8. Frequency polygons of steady potentials recorded from guinea-pig cortical slices incubated in saline lacking glucose (dotted line) and saline containing glucose (solid line); three slices from two animals in each case.

sodium salts of the saline by potassium salts produced an effect on the steady potential which was even more apparent. Thus, small steady potentials recorded from slices in saline with high concentration of potassium salts (130 mM) increased after the tissue was rinsed and incubated in low-potassium salines. If the slices were placed in high-potassium saline for 6–10 min the steady potentials recorded from guinea-pig cortical slices were not greater than −18 mV, mostly below −15 mV.

The effect of a change in the metabolic environment on the steady potentials was most readily observed when the experimental saline was equilibrated with N_2–CO_2 instead of O_2–CO_2. The steady potentials became smaller and in 10–15 min were no longer recordable from the slice. Supplying O_2–CO_2

mixture again in a few minutes resulted in a recovery of small steady potentials. This was found with seven slices (3 from 2 cats and 4 from 2 guinea-pigs). The largest steady potential recorded in N_2–CO_2 was −13 mV (slice from a cat) and its mean value, −7 mV. Thus the cortical slices were more sensitive to anoxia than were liver slices (Fig. 2).

The necessity for satisfactory metabolic conditions for the maintenance of steady potentials was also shown in experiments with thick slices. Slices of guinea-pig cortex were cut 0·9–1·0 mm thick and incubated in oxygenated glucose-salines together with others of the usual 0·3–0·4 mm thickness. Potentials recorded in the thick slices were small in magnitude, and in a given distance of travel of the micro-electrode were few. The thick slices presumably contained a greater proportion of structures which were undamaged by cutting, but this factor appeared to be of less importance than the limited access of oxygen or other substrates.

DISCUSSION

Although in the present study entry of the micro-electrode into tissue slices could readily be seen microscopically, its entry into individual cells (except in the muscle experiments) was not observed; however, there was little doubt that the discontinuity of the negative steady potential which was recorded by the penetrating micro-electrode indicated entry of the electrode into the cells.

Since the magnitude of resting membrane potentials recorded from the cortex varies with the degree of damage to the cells by the penetrating micro-electrode (Li, 1955), the wide distribution of potential measurements obtained from cortical slices is to be expected. Assuming that a large number of nervous elements in the slices were cut at the dendritic shafts, and that only a few could escape amputation of their axonal processes, it is not surprising to find a wide range in the distribution of resting membrane potentials. The variation of potential measurements from cortical slices is greater than the variation of those recorded from the liver and kidney slices. This is apparently due to the fact that in the liver and kidney the cells do not have processes and damage to the individual elements within the slices was minimum. The variation of measurements from the gracilis anticus muscle might have been due to similar damage to the fibres when the muscle was dissected and removed from its attachments.

The average thickness of the parietal cortex of the guinea-pig is about 1·2 mm and of the cat 1·8 mm. The first slice obtained from this area of the cortex consisted of layers I and II and the upper portion of layer III. The nervous elements in the slice which would be expected to be encountered by the penetrating micro-electrode were largely the horizontal cells and the small and medium-sized pyramidal neurones. The axons and dendrites of these neurones are extremely small, the largest being less than 2 μ in diameter. It is

unlikely that, when a micro-electrode could be maintained in a position from which a negative steady potential was recorded for 1 min or longer, the micro-electrode was located inside a dendrite or an axon, although intracellular dendritic potentials were recordable from the Purkinje cells of the cerebellum (Granit & Phillips, 1956, 1957).

The second slice obtained from the cortex after the first slice was cut consisted of cells in the lower portion of layer III, layer IV and possibly a small portion of layer V. In layer IV are Golgi type II cells whose dendritic branches are known to be confined to the same layer. It is possible that in the second slice many of these cells were relatively undamaged and contributed to the steady potentials recorded with the intracellular electrodes.

The question of whether the steady potentials from cortical slices were recorded from nerve cells or glial elements remains. On no occasions was it possible to evoke a response by depolarizing current or stimulating pulses. A similar observation was reported in experiments with intact animals (Li, 1955; Phillips, 1956) in which some of the cortical elements showed large steady potentials but failed to respond to electrical stimuli. It is to be noted that from these cortical elements (Li, 1955) injury discharges were not recorded. In the present study injury discharges were recorded from slices of the cortex and muscle but not from the liver and kidney. A reasonable assumption is that the cells of the liver and kidney are inexcitable, whereas the muscle fibres and some of the cells in the cortical slices are excitable.

Experiments with intact animals showed that the electrical activity of the cortex recorded with micro-electrodes (Li & Jasper, 1953) was extremely sensitive to a change in the general condition of the animal. A brief period of anoxia or a deepening of the anaesthesia would completely abolish the spike discharge, although the slow wave activity might not be affected by them. In the experiments with cortical tissue in the slice chamber, rhythmic slow wave activity has never been successfully demonstrated.

It is concluded that although the neuronal elements in the slice could maintain an intracellular steady potential, their electrical responsiveness depended upon factors which were not present in the isolated specimen. The potentials observed appear likely to represent the differential distribution of sodium and potassium salts between the tissue cells and the external fluids, a matter well documented in relation to excitable tissues in which the normal distribution and resting potentials are known to require oxygen (Keynes & Lewis, 1955). The data summarized by Leaf (1956) show maintenance of differential concentrations of sodium, potassium and chloride ions in slices of liver, kidney and brain cortex prepared and maintained under conditions similar to those of the present experiments. Again, lack of oxygen diminished the concentration gradients. In cerebral tissues, lack of glucose also has this effect (Terner, Eggleston & Krebs, 1950). In the present experiments lack of glucose and

oxygen diminished the resting membrane potentials observed in the tissue. So also did high concentrations of potassium salts in the external medium, this representing an attempt to decrease directly the gradient in potassium ions. Correlation between the metabolic and electrical events in cerebral tissues is illustrated by the way in which these changes in glucose, oxygen and potassium salts now found to lower resting membrane potentials were previously known to deplete the phosphocreatine of the tissue and to prevent metabolic response by the tissue to applied electrical pulses (McIlwain, 1952; Gore & McIlwain, 1952).

Many factors can be suggested as contributing to the failure to detect excitation in response to electrical stimulation, in cerebral slices during the present experiments. These include mechanical damage and the limited techniques at present employed. It is also to be emphasized that many additions to glucose salines are required to maintain the composition of such isolated tissues, at values observed *in vivo* (see McIlwain, Thomas & Bell, 1956; Thomas, 1956).

SUMMARY

1. A chamber is described for maintaining slices of isolated mammalian tissues under good metabolic conditions while electrical observations are made.

2. The resting membrane potential of the liver cells of the guinea-pig was found to be 36 ± 6 mV (S.D.) and of the cat, 34 ± 5 mV. They were both reduced to approximately half by anoxia, and recovery was accomplished by restoring oxygen. The resting membrane potential of kidney cells was 35 ± 2 mV. Injury potentials were not encountered by the penetrating micro-electrode in slices of kidney and liver.

3. The resting membrane potential recorded from cerebral cortical slices varied considerably, ranging from 1 to 91 mV in the guinea-pig and from 15 to 93 mV in the cat. They were reduced by lack of glucose and also by replacing part of the sodium salts of the experimental saline by potassium salts. Anoxia also greatly diminished the resting membrane potential and this change was found to be reversible. Injury discharges were occasionally recorded from cortical slices. Spontaneous spike activity was not observed. Electrical stimulation failed to evoke a spike discharge from cortical slices.

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