METHODS

RECORDING THE ACTION POTENTIALS OF NERVES IN AN UNSCREENED ROOM

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(Received January 7, 1958. Presented by Active Member Acad.Med. Sci. USSR, V. V. Parin)

Until recently the only suitable method of protection against grid interference during work with weak electric signals such as nerve action potentials was careful screening of the room or even of the animal itself. However, working in a screened chamber, or placing the animal in a screened cage are both connected with great difficulty and inconvenience. This is an obstacle to the wide introduction of electrophysiological methods of investigation into practice. Recently a number of attempts have been made to construct special bioamplifiers and a method of working which would enable action potentials of an animal to be recorded in ordinary laboratory conditions without screening the object.

We set out to make a careful investigation of this problem and to determine what limits of sensitivity could be reached by recording nerve action potentials in an unscreened room. We divided the solution of the problem into 2 stages: 1) the construction of a bioamplifier possessing the highest possible coefficient of rejection, and 2) the development of a method of picking up the potentials, the selection of the optimal construction of the electrodes of the earthing system and so on.

Biological amplifiers, as a rule, are made according to a symmetrical circuit [1, 2, 4, 5, 8]. Each cascade contains 2 valves with separate anodic resistances of equal size and one general resistance R_k in the cathode circuit of both valves. The action potentials are picked up bipolarly and applied to the grids of two symmetrical valves in the first cascade. An amplified potential is taken from between the anodes of these valves and, as also in the opposite phase, is applied to the grid of the second symmetrical cascade and so on.

The main feature of the working of such an amplifier is that for an action potential which falls first on one valve of the input cascade and then on the other (as the wave of excitation moves along the nerve), the amplifying properties of the circuit are used to their full extent. The presence of the common, and usually high, cathode resistance R_k in this case does not lead to diminished amplification (there is no negative feedback). If, however, 2 electric signals, perfectly equal in amplitude, form and phase, reach the grids of both valves in the input cascade at the same time, then the resistance R_k will act as a negative feedback resistance and the amplification of this synphasic signal will be greatly diminished. Since grid interference is applied equally to both electrodes it will be weakened in comparison with the recorded potential of bioelectrical origin. This phenomenon is called rejection, and the ratio between the coefficient of amplification of the recorded antiphasic potential to the coefficient of amplification (in practice often weakening) of the synphasic potential, calculated for the amplifier as a whole, is called the coefficient of rejection.

As follows from the theory of the symmetrical amplifier [1, 2, 4, 5, 6, 8], the coefficient of rejection of such an amplifier is higher, the greater the magnitude of the common cathode resistance R_k and the higher the coefficient of amplification μ of the valves incorporated in it. The development of the amplifier circuit has therefore gone in the direction of maximum increase of both these parameters. Its final form is shown in Fig. 1.

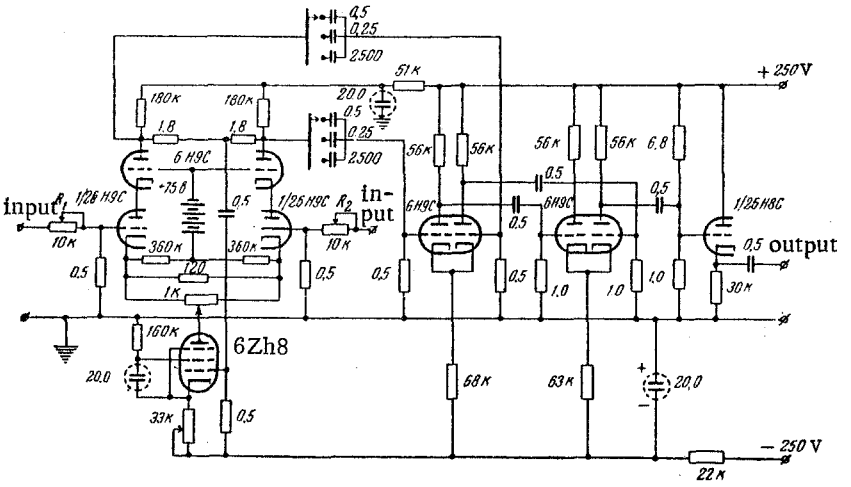

Fig. 1. Circuit showing the principle of the preamplifier.

As the common cathode resistance of the valves of the first cascade we used a pentode 6Zh8. This usage of the high dynamic resistance of a pentode has become a usual feature of the design of biological amplifiers in recent years [3, 5, 6, 8]. The dynamic resistance of the pentode is also increased on account of the negative feedback through resistances included in its cathode circuit.

Pentodes possess high intrinsic coefficients of amplification μ. For this reason some workers [5, 8] and foreign firms (for example, the American firm of Grass) use pentodes as the input valves of the bioamplifier. This solution does not seem justifiable to us, since pentodes differ from triodes in their higher noise level, which restricts the sensitivity of modern bioamplifiers.

In our amplifier, in place of each input valve of the symmetrical cascade we used a series combination of 2 triodes, the cathode of the second triode being directly connected to the anode of the first, the grid of which receives the potential to be amplified. The grids of the second triodes are connected together and their potential is relatively midway between the very close potentials of the cathodes of the first triodes, and is maintained constant by means of a dry battery. The amplified potential is obtained between the anodes of the second triodes, with which anode loads are included in series. A similar design including triodes was investigated by Klein [6]. He showed that such a combination of triodes possesses a very high coefficient of amplification, approximately equal to the product of the coefficients of amplification of each triode, and in this sense equivalent to a pentode; on the other hand it is distinguished by the characteristic low sound level of the triode [7]. Finally, following Richards [8], we led the negative feedback from the midpoint between the anodes of the second triodes to the controlling grid of the pentode, used as a cathode load. This lead profoundly lowers the change in the common level of anode potentials of both halves of the cascade during feeding of synphasic signals into its input; in this way the effect of the different dynamic characteristics of the valves is minimized and the rejection increased.

The second and third cascades of the amplifier are constructed according to the usual symmetrical design; the output signal is taken from the cathode repeater. Thermionic emission in the triodes of the first cascade is maintained by a low tension direct current (about 3 v). The second cascade is also supplied with direct current of normal tension. The voltages of +250 and −250 v are stabilized by electronic stabilized by electronic stabilizers.

In this design we obtained a value of more than one million for the coefficient of rejection, which was clearly adequate for carrying out the second part of the investigation.

In its construction the apparatus is produced in the form of easily transportable sections — a two-channel preamplifier section and a power section; neither is larger than the usual electric record player. The apparatus is wholly worked from the alternating current mains.

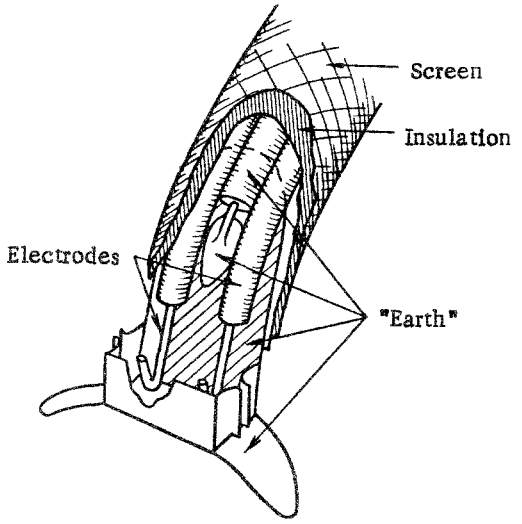

Fig. 2. Construction of the electrode for testing the action potentials of a nerve (magnified).

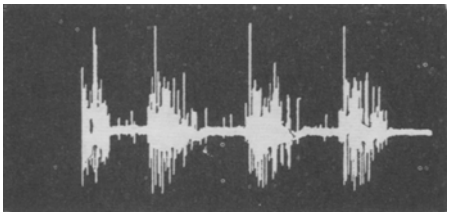

Fig. 3. Oscillogram of impulses from the depressor nerve of the cat taken without any form of screening of the object.

The preamplifier has a coefficient of amplification of 20,000 throughout a wide range of working frequencies of 20 cps to 8 kilocycles, and is designed for working with an industrial double-beam OK-17M oscillograph. The amplification is controlled in the oscillograph itself. The recordings on the oscillograph screen are transferred to a cinematograph film.

In developing a method of picking up the biopotentials we became convinced that earthing the animal itself is unnecessary and even harmful, since it creates the possibility of production by a magnetic field of an alternating current in the "loop" animal – electrode – "earth" of the amplifier – "earth" of the animal. We earth the nerve directly at its point of contact with the body of the animal, i. e., almost at the testing electrode itself. If during testing the nerve is not divided or if its divided end is in contact with the animal's body (even if only through a moist ligature), it is earthed on both sides. Earthing is done through a "wing" of a brass tin-covered box inside which and insulated from it are placed the platinum hooks of the electrodes (Fig. 2).

Emerging through lateral slits in the box, the nerve is placed on the "wing" and pressed against it by a cotton swab soaked in Ringer's solution. The box itself is connected by means of a third lead, which must be bound together with the two leads which come from the electrodes, to the "earth" inside the amplifier itself. All 3 leads are contained in a screened, flexible cover which leads almost to the electrodes, and is also earthed at one point only, close to the amplifier. This prevents the development of magnetic loops in the testing electrodes and the screen itself. After the nerve has been placed on the electrodes the box may be covered with vaseline. On account of the narrowness of the vertical slits the vaseline does not escape.

The method of picking up the potentials just described, together with the high rejection of the amplifier, enabled us to solve the problem of recording the action potentials of nerves in an animal without the use of a screen. As an illustration in Fig. 3 is shown the oscillogram of impulses taken from the depressor nerve of the cat. The power feed, transformer, illuminating lamp and other items were situated a few centimeters from the animal. The experimenter was able to manipulate the animal with his hands. In spite of this, the manipulations did not exceed the level of sound production. This level in Fig. 3 corresponds approximately to 20 μv "from peak to peak" (with a frequency range of 8 kilocycles).

In the course of further investigation of the apparatus we found that the figure of rejection obtained can be lowered more than ten times without interfering with its working. This was achieved by introducing into the input circuits of the apparatus balanced resistances R_1 and R_2 (controlled by the same handle). The importance of these resistances is that they equalize the amplitudes of the impulses reaching the grids of the input cascade of the amplifier from the electrodes, which are almost always unequal. Naturally this equalization must be carried out with the highest precision, and in consequence the preservation of such a high rejection as we had at first loses its purpose. Nevertheless we look upon the work of creating an amplifier with a very high coefficient of rejection as showing great promise, so that in the very near future it can evidently be expected that the level of sensitivity of bioamplifier will be further raised.

SUMMARY

By employing in each half of the symmetrical input cascade of the amplifier a series combination of two triodes and a pentode as a total cathode load with a deep inverse feedback – it becomes possible to bring the rejection factor of such an amplifier up to one million (without the matching of tubes).

A portable double-channel preamplifier using such a circuit has been developed for work with an industrial double-beam oscillograph. With the aid of this preamplifier and electrodes of special construction it was possible to register the potential of nerve action of animals in short-term experiments without screening the room and object in usual laboratory conditions.

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A METHOD OF STUDYING GASTRIC AND DUODENAL SECRETION AT THE SAME TIME**

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(Received April 22, 1957. Presented by Active Member Acad. Med. Sci. USSR, N. A. Rozhanskii***)

I. P. Pavlov (1897, 1902) repeatedly drew attention to the development of methods which would enable the simultaneous observation of the work of several digestive glands in the same normal animal.

The performance of this task is often beset by difficulties: on the one hand it is necessary to obtain, measure and test the juice from each gland separately, and on the other hand, the juices must enter the digestive tract in the same quantities as were secreted by the glands both outside and during the experiment, otherwise the normal course of the digestive process would be disturbed.

* In Russian.

** Delivered (with a demonstration on a dog) at the meeting of the Riazan section of the Society of Physiologists on December 27, 1955.

*** Deceased.