NEURONAL RESPONSES OF INTACT AND ISOLATED STRIPS OF THE SENSORIMOTOR CORTEX TO THYROXINE INJECTION

M. G. Amiragova and N. É. Kuksgaus

UDC 612.825.1.014.2.014.46:
615.357.441:577.175.444

KEY WORDS: thyroxine; cerebral cortical neurons.

Under normal conditions of physiological activity cortical neurons are subject to the constant influence of impulses arriving from the various afferent systems and also from subcortical structures. Data obtained in the writers' laboratory, showing that subcortical neurons are more sensitive than cortical neurons to hormones [1, 2], suggest initial involvement of subcortical neurons in activation which, spreading along ascending pathways, ultimately extends to formations in the cortex, especially in its anterior zones. Evidence in support of this view is given by the later response of higher levels of the CNS to changes in hormonal homeostasis and the impression is gained that this response is subcortical in origin [3]. However, the question arises: What is the immediate response of cortical neurons to changes in the blood hormone composition?

Experiments on the intact cortex, retaining its connections with other parts of the brain, could not give a complete answer to this question because it is impossible to differentiate between the intrinsic electrical activity of the cortex and that due indirectly to subcortical influences. It was accordingly decided to use as the test object a neuronally isolated strip of cortex (ISC), which would seem to be a suitable method with which to study this problem.

EXPERIMENTAL METHOD

Experiments were carried out on 110 waking adult male rabbits, fixed in a stereotaxic frame. Altogether recordings were obtained from 230 neurons, 146 in the intact cortex and 84 in ISC. Only spontaneously active neurons were tested.

To record action potentials burr-holes were drilled in the skull under procaine anesthesia for microelectrodes. After removal of the dura the burr-holes were filled with a 3% solution of agar-agar. Activity of sensorimotor cortical neurons was always recorded at a depth of 800-1400 μ, which corresponds to the maximum of their activity. Microelectrodes made from Pyrex glass were filled with 2.5 M NaCl solution. The resistance of the electrodes was 30-50 MΩ and the diameter of their tip 1-2 μ. Unit activity was amplified by means of an MZ-4 dc cathode follower (Nihon Kohden, Japan) and led to one channel of a dual-beam VC-7 oscilloscope from the same firm. Data from the VC-7 was recorded on magnetic tape by a two-channel "Juniper 202" tape recorder and, at the same time, through a trigger device on paper tape by a seven-channel electroencephalograph (from VEB Zwönitz, East Germany).

The strip of cortex was isolated by the method described previously [7]. Electrical activity was recorded 2.5-3 h after the end of all the preliminary manipulations, in the same way as in experiments on the intact cortex. To inject the hormone intravenously into the marginal vein of the rabbit's ear, the injection needle with a stylet, rinsed with heparin, was first inserted. This was intended for subsequent injection of thyroxine solution in the course of the experiment. This procedure was adopted in order to avoid painful stimulation during the injection.

Thyroxine was injected in a dose of 0.02 mg/kg body weight. The hormone was diluted in 2 ml physiological saline at pH 8.0-9.0. Control animals received the same volume of physiological saline. The functional state of the neurons was judged from the change in

Laboratory of Neuroendocrine Regulation, P. K. Anokhin Institute of Normal Physiology, Academy of Medical Sciences of the USSR, Moscow. (Presented by Academician of the Academy of Medical Sciences of the USSR V. N. Orekhovich.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 94, No. 9, pp. 10-12, September, 1982. Original article submitted March 19, 1982.

0007-4888/82/9409-1171$07.50

their discharge frequency. Unit activity was analyzed by computer (from Nihon Kohden, Japan). The number of discharges of the test neurons was counted in 10-sec time cuts in the course of every 10 min of recording, with intervals of 20 sec. The response of a neuron was considered to be significant if the rate of discharge changed reproducibly by 30% compared with the previous period after injection of thyroxine. Graphs of mean discharge frequency were plotted from the processed numerical data. Student's test was used for statistical analysis.

EXPERIMENTAL RESULTS

The number of neurons tested in the intact sensorimotor cortex was 146 (136 after injection of thyroxine, 10 after injection of physiological saline).

Of the 146 neurons recorded in the intact cortex 52 (38%) had activity consisting of single discharges, 10 neurons (20%) had regular activity. A volley type of discharge was observed in 20 neurons (15%) and a mixed type in 64 neurons (47%). The mean spontaneous discharge frequency was 4.24 ± 0.95 spikes/sec, in agreement with data in the literature [4, 5].

After injection of thyroxine, spontaneous activity was recorded from only 136 neurons. Of this number, 62 (46% of all neurons) were activated, an inhibitory response was observed in 34 neurons (25%), and 40 neurons (29%) did not respond to injection of the hormone. The reactive population thus comprised 96 neurons (71%).

The mean percentage of changes on the activated neurons compared with spontaneous activity was 96.7. In all cases the frequency remained high until the end of observation, which lasted 2 h. Inhibition of unit activity under the influence of injected hormone took the form of a decrease in discharge frequency on average by 60%, and this also continued until the end of the investigation.

In control experiments with injection of physiological saline no significant changes were observed in unit activity. The latent periods of action of thyroxine could not easily be determined precisely in all cases. In most neurons tested the action of the hormone was manifested 1-2 min after injection. The action of thyroxine reached a maximum after 7-9 min.

The type of discharge was changed under the influence of thyroxine in 45 cases (33%). In 25 cases, neurons giving single discharges switched to a mixed type of activity 5 min after injection. Of the 20 neurons with a mixed type of discharge, 18 (90%) changed to the volley type, and two neurons (10%) began to give a single type of discharge.

Activity of 84 neurons was recorded from the neuronally isolated posterior zone of the sensorimotor cortex. During each insertion of the microelectrode with a step of 10 μ the number of spontaneously active neurons encountered in ISC in these experiments was only one-third to half of that in the intact cortex. This figure agrees with data of other workers [6, 8].

Of 74 neurons recorded in ISC 14 (19%) gave a single type of discharge, 32 neurons (43%) a mixed type of discharge, and 28 neurons (38%) a volley type of discharge.

The discharge frequency of the neurons varied from 0.1 to 28.3 spikes/sec. The mean discharge frequency of the neurons was 4.93 ± 0.73 spikes/sec. A similarity could thus be noted between the mean discharge frequency of the intact cortex and ISC. The same similarity also was observed by other workers in experiments on unimmobilized and unanesthetized rabbits [8].

Comparison of the type of spontaneous activity of neurons in the intact cortex and ISC showed a considerable decrease in the percentage of neurons with a single type of discharge and a more than twofold increase in the percentage of neurons with volley activity in ISC. Other workers [8] also have described an increase in the number of neurons with volley activity in ISC. This phenomenon can evidently be linked both with the absence of an organizing effect of the subcortex on the cortex and also with changes taking place in ISC as a result of the operation.

Activity of 74 neurons was recorded in ISC after injection of thyroxine into the animals. Of this number 30 neurons (41%) were activated, 14 (19%) were inhibited, and 30 neurons (42%) did not respond to the hormone. The reactive population thus comprised 44 neurons (58%).

The activation effect was expressed as an increase in frequency of spikes in the dis-

charge on average by 78.3% compared with initially. The firing rate of the neurons increased only once in all cases.

In the case of inhibition of unit activity the initial firing rate decreased on average by 69% below the spontaneous level. Just as in cases with activation of neurons, the decrease in firing rate occurred only once. After injection of physiological saline activity was recorded from 10 neurons in which no appreciable changes in the discharge were observed.

In the overwhelming majority of cases the response to thyroxine, just as in the experiments on the intact cortex, began in the first few minutes ofter injection of the hormone. The response of activation and inhibition of ISC neurons to thyroxine was considerably shorter than that of neurons in the intact cortex and it lasted about 1 h, after which the firing rate returned to its initial level.

The type of discharge of 16 neurons (22%) was changed after injection of the hormone. Nine of these neurons (56%) switched from a volley type of discharge to a mixed type, whereas the remaining seven neurons (44%), with a mixed type of discharge in their spontaneous activity, changed after injection of the hormone to the volley type, with a simultaneous increase in spike frequency.

When the character of response of neurons of the ISC and intact cortex to a change in hormonal homeostasis is compared, the first feature to note is the differences in the reactive neuron population in these structures. In ISC, for instance, it was 58%, in the intact cortex 71% ($P < 0.05$). Another important difference is in the duration of the response, which was only half as long in ISC as in the intact cortex. Finally, the change in discharge frequency in ISC averaged 78.3%, compared with 96.7% in the intact cortex ($P < 0.001$).

The basic differences in the pattern of response of the ISC neurons, reflecting the intrinsic response of cortical neurons to hormonal stimulation, are thus evidence that their functional capacity is lower than that of the intact cortex, and they demonstrate the importance of afferent volleys from subcortical structures, principally the hypothalamus, for the realization of adequate responses of cortical neurons to changes in hormonal homeostasis. Consequently, cortical neurons, receiving preformed nervous information on hormonal homeostasis from hypothalmic structures specialized in this respect thus acquire additional power to control hormone production at a level adequate to meet the needs of the body.

LITERATURE CITED

1. M. G. Amiragova, B. V. Stul'nikov, and N. É. Kuksgaus, Fiziol. Zh. SSSR, 66, No. 9, 1292 (1980).
2. M. G. Amiragova, B. V. Stul'nikov, S. I. Svirskaya, et al., in: Problems in Physiology of the Hypothalamus [in Russian], No. 11, Kiev (1977), pp. 40-47.
3. M. G. Amiragova and G. I. Svirskaya, Dokl. Akad. Nauk SSSR, 179, No. 5, 1246 (1968).
4. N. N. Vasilevskii, in: Mechanisms of Activity of the Central Neuron [in Russian], Moscow–Leningrad (1966), p. 149.
5. A. M. Melekhova, G. I. Shul'gina, and A. N. Lebedev, Zh. Vyssh. Nerv. Deyat., No. 5, 889 (1966).
6. M. M. Khananishvili and É. G. Zarkeshev, Neirofiziologiya, 3, 236 (1971).
7. Yu. A. Kholodov, Effect of Electromagnetic and Magnetic Fields on the Central Nervous System [in Russian], Moscow (1966).
8. G. A. Él'kina and Yu. A. Kholodov, in: Investigation of the Organization of Unit Activity in the Central Cortex [in Russian], Moscow (1971), pp. 14-37.