ELECTROPHYSIOLOGICAL STUDY OF THE EFFECT OF ACETYLCHOLINE ON RENAL RECEPTORS

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Injection of small doses of acetylcholine (0.25-0.75 μg) into a branch of the renal artery of an anesthetized cat increased the renal blood flow and potentiated the fast flow of impulses in the afferent nerves of the kidney. Average doses (1-5 μg) had no significant effect on the blood flow, but potentiated the flow of fast impulses still further; slow activity also appeared under these conditions. Large doses (10-1000 μg) of acetylcholine reduced the blood flow and the fast activity but potentiated the slow activity still further. In the writer's opinion, acetylcholine has a direct depolarizing action on the kidney receptors and gives rise to effects indirectly through vascular changes and changes in the tone of the smooth muscle.

The mechanisms of action of acetylcholine on the receptors includes its action on the specialized cholinergic receptor [2-4], a nonspecific depolarizing action on various receptors and on nerve fibers, mainly the nonmedullated group C fibers [6, 7, 9, 14, 18, 19], and an action on receptors produced indirectly through contraction of neighboring smooth muscle [14, 21].

This paper deals with the effect of acetylcholine on the receptors of the kidney in connection with its action on the renal blood flow.

EXPERIMENTAL METHOD

Experiments were carried out on 29 cats anesthetized with hexobarbital (1% solution, intravenously). Activity was recorded from the peripheral ends of the divided renal nerves, amplified, and recorded on a CRO. Acetylcholine (0.1-1000 μg) in Ringer's solution (dilutions 10^{-3}-10^{-6}) was injected from a syringe into the renal vessels through a divided branch of the renal artery, the needle attached to the syringe being inserted as far as the hilus of the kidney. Activity was recorded 5 sec after each injection. The blood flowing from a divided branch of the renal vein, into which a thin glass cannula was introduced, was measured in drops (Fig. 1D).

EXPERIMENTAL RESULTS AND DISCUSSION

Injection of 0.25-0.75 μg acetylcholine into the renal artery increased the renal blood flow (Fig. 1A) and led to the appearance or potentiation of ungrouped fast activity (Fig. 2A). The amplitude of the individual spikes was 20-30 μV. After injection of 0.1-0.5 μg acetylcholine, the threshold dose causing the appearance or potentiation of this activity was as a rule 0.25 μg, but in 3 cases it was 0.5 μg.

Injection of 1-5 μg acetylcholine caused no significant change in the renal blood flow in the first 10 sec (Fig. 1B), but led to a further increase in the fast activity (Fig. 2B). Slow potentials appeared under these conditions, and the amplitude of the composite wave reached 10-20 μV.* The threshold dose causing

*These complex potentials will be described subsequently in the text as waves, by contrast with the simple potentials which are called spikes.

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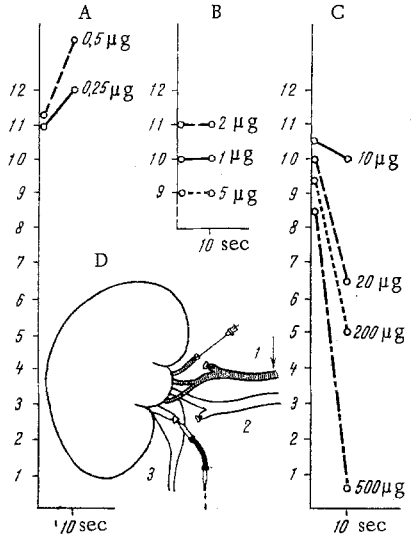

Fig. 1

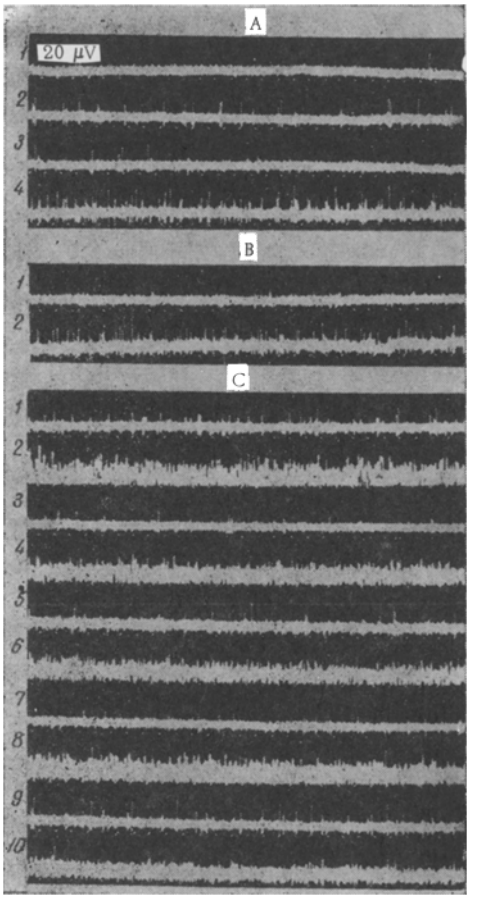

Fig. 2

Fig. 1. Renal blood flow (in drops per 10 sec) after injection of small (A), average (B), and large (C) doses of acetylcholine into the hilus of the kidney (mean results of 8 experiments); D) scheme showing method using to measure blood flow: 1) renal artery; 2) renal vein; 3) ureter.

Fig. 2. Afferent activity in renal nerves after injection of small (A), average (B), and large (C) doses of acetylcholine into the hilus of the kidney. In A: 1, 2) before, and 3, 4) 5 sec after injection of 0.25 and 0.5 µg acetylcholine respectively; in B: 1) before, 2) 5 sec after injection of 1 µg acetylcholine; in C: 1, 3, 5, 7, 9) before, and 2, 4, 6, 8, 10) 5 sec after injection of 10, 20, 50, 200, and 600 µg acetylcholine respectively. Time marker 0.02 sec.

the appearance of slow potentials was 1 µg acetylcholine (in Fig. 2B these potentials are shown as widening of the noise line).

Injection of 10-1000 µg acetylcholine reduced the renal blood flow, and the intensity of this decrease was directly proportional to the dose of acetylcholine (Fig. 1C). The frequency of the fast activity was considerably reduced (Fig. 2C). Injection of large doses of acetylcholine after the previous injection of smaller doses caused the complete disappearance of the fast activity. This fact has been observed previously in experiments on other organs [16, 20]. Against the background of the diminished renal blood flow and slowing of the fast activity, the slow activity continued to increase in amplitude and frequency (Fig. 2C).

All the changes in fast and slow activity during the action of these doses of acetylcholine are shown in Fig. 3. The differences between thresholds and maxima of the responses of the fast and slow activity are also illustrated. For instance, the threshold of appearance of fast activity was 0.25 µg acetylcholine, and of slow activity 1 µg; the maximum of the fast activity was recorded after injection of 5 µg acetylcholine and its frequency was 55 spikes/sec, while the slow activity reached a maximum after the injection of

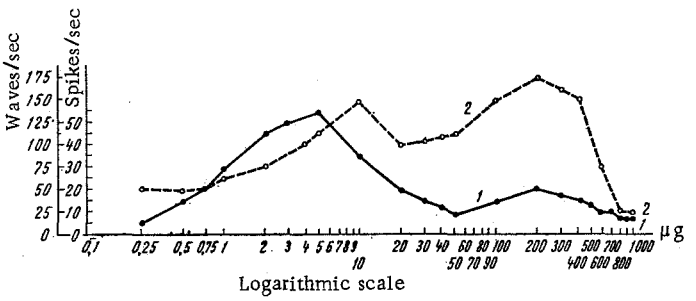

Fig. 3. Fig. 3. Afferent activity in renal nerves after injection of small, average, and large doses of acetylcholine into the hilus of the kidney (mean results of 15 experiments): 1) fast activity; 2) slow activity. Abscissa, logarithm of dose of acetylcholine; ordinate, frequency of activity.

10 μg acetylcholine, when its frequency was 150 waves/sec. With a further increase in the dose of acetylcholine in response to injection of 200 μg the activity reached a second maximum, corresponding to a frequency of 20 spikes/sec for the fast activity and 175 waves/sec for the slow activity.

In 3 experiments in which atropine hydrochloride (0.1 ml 10^{-3}) was injected 30 sec before the injection of acetylcholine, the responses of the receptors to acetylcholine were reduced. The inhibitory effect of atropine was greater on the slow activity.

In 2 of the 14 experiments in which a single injection of large doses (100-500 μg) of acetylcholine was given, the fast ungrouped activity was replaced by spikes grouped in the rhythm of the pulse. Grouping of the waves was observed only for 30-60 sec after the injection of acetylcholine, when the flow of both fast and slow activity was slightly weakened. Grouping of the spikes continued for 1.5-2 min, when it was replaced by the ordinary ungrouped activity observed under the original conditions.

In one case the acetylcholine was injected 2-3 cm away from the hilus of the kidney (in Fig. 1D the arrow shows the approximate site of injection). In this experiment the threshold doses of acetylcholine were increased (to 5 μg for the fast activity and 20 μg for the slow), and the responses of the receptors to acetylcholine were less marked.

The increase in blood flow and excitation of the renal receptors in response to injection of small doses of acetylcholine (0.25-0.75 μg) was due to vasodilatation and to the depolarizing action of acetylcholine directly on the receptors [17, 22]. When 0.1-0.25 μg acetylcholine was injected, the threshold dose causing the appearance or potentiation of activity was 0.25 μg.

Average doses of acetylcholine (1-5 μg) produced no significant change in the blood flow during the first 10 sec, but produced a further increase in the fast activity. Slow activity also appeared. The threshold of onset of this activity was 1 μg acetylcholine, i.e., four times higher than for the fast activity. This can be explained most probably by the lower sensitivity of the receptor systems of the group C fibers to the action of acetylcholine [16, 20]. For example, when acetylcholine is injected subcutaneously, the threshold dose for the appearance of activity in the group C afferent systems of the skin is 2 μg [13, 14], while for the group A fibers of the isolated receptors of the crab it is only 0.001 μg [22]. The results obtained after injection of average doses of acetylcholine prove that it has a direct depolarizing action on the renal receptors. The fast activity reached a maximum after injection of 5 μg acetylcholine, when its frequency was 55 spikes/sec.

Large doses of acetylcholine (10-1000 μg) reduced the renal blood flow and weakened the fast activity. The reasons for this weakening could be: a) constriction of the renal vessels due to the direct action of acetylcholine on them [8] and indirectly through a change in the general circulation, and b) the inhibitory action of large or repeated doses of acetylcholine on the receptors [16, 20]. After the initial weakening of the fast activity, however, further potentiation of the slow activity was observed, to reach a maximum occurred in response to injection of 10 μg acetylcholine, and corresponded to 150 waves/sec. The second maximum of the slow activity was recorded in response to injection of 200 μg acetylcholine and amounted to 175 waves/sec. It was probably due to the depolarizing action of acetylcholine directly on the nerve fibers (principally on nonmedullated group C fibers as being more accessible to the action of acetylcholine than the medullated group A fibers), and also to the activity of the receptors of the smooth muscles of the

vessel walls on account of their spasm. For instance, preliminary injection of atropine hydrochloride, which has no effect on the action of acetylcholine on afferent nerve systems but which blocks its action on smooth muscle, into the renal vessels weakened the effect of acetylcholine on both activities, but mainly on the slow activity.

The suggestion has recently been made that the potentiation of afferent activity by acetylcholine can be attributed partly to its stimulant action on efferent endings and, consequently, to antidromic activity in sympathetic nerve fibers [1, 5, 11, 15].

Grouping of the spike activity in rhythm with the pulse observed in 2 of the 14 experiments in response to a single injection of large doses of acetylcholine can evidently be explained by: a) increased excitability of the renal mechanoreceptors due to the depolarizing action of acetylcholine on them [6, 10, 12, 14, 21, 22] and b) weakening of the background electrical activity masking the pulse grouping of the spikes as a result of the vasoconstrictor action of acetylcholine [8].

When acetylcholine was injected into the artery 2-3 cm away from the renal hilus the threshold of the responses was increased by 20 times, while the maximal value of the responses of the receptors to acetylcholine was reduced by 1.5-2 times, because of its rapid destruction of the blood cholinesterase [3].

LITERATURE CITED

1. N. A. Anikina, in: Electrophysiology of the Nervous System [in Russian], Rostov-on-Don (1963), p. 19.
2. S. N. Nistratova, The Biochemical Nature of the Cholinergic Receptor and the Mechanism of Action of Acetylcholine, Author's Abstract of Candidate's Dissertation, Moscow (1959).
3. T. M. Turpaev, The Mediator Function of Acetylcholine and the Nature of the Cholinergic Receptor, Author's Abstract of Doctoral Dissertation, Moscow–Leningrad (1961).
4. T. M. Turpaev, The Mediator Function of Acetylcholine and the Nature of the Cholinergic Receptor [in Russian], Moscow (1962).
5. V. M. Khayutin, V. L. Shur, and Yu. E. Malyarenko, Fiziol. Zh. SSSR, 56, 84 (1970).
6. C. Armett and J. Ritchie, J. Physiol. (London), 152, 141 (1966).
7. C. Armett and J. Ritchie, J. Physiol. (London), 165, 141 (1963).
8. A. Aström, Y. Crafoord, and U. Samelius-Broberg, Acta Physiol. Scand., 61, 159 (1964).
9. T. Barness et al., Science, 104, 569 (1946).
10. G. Brown and I. Gray, J. Physiol. (London), 107, 306 (1948).
11. R. Carbera, R. Torrance, and H. Viveros, Brit. J. Pharmacol., 27, 51 (1966).
12. W. Douglas and J. Gray, J. Physiol. (London), 119, 118 (1953).
13. W. Douglas and J. Ritchie, J. Physiol. (London), 146, 46 (1959).
14. W. Douglas and J. Ritchie, J. Physiol. (London), 150, 501 (1960).
15. C. Ferry, J. Physiol. (London), 167, 487 (1963).
16. A. Iggo, Quart. J. Exp. Physiol., 42, 130 (1957).
17. A. Yarret, J. Physiol. (London), 133, 243 (1956).
18. P. Nordqvist, Acta Pharmacol. (Copenhagen), 8, 226 (1952).
19. P. Nordqvist, Acta Pharmacol. (Copenhagen), 8, 223 (1952).
20. A. Paintal, J. Physiol. (London), 126, 255 (1954).
21. A. Paintal, in: Twentieth International Physiological Congress, Abstracts, Vol. 1, Brussels (1956), p. 78.
22. C. Wiersma, E. Furshpan, and E. Florey, J. Exp. Biol., 30, 136 (1953).