ROLE OF Ca^{++} IONS IN THE FORMATION OF VASCULAR TONE AND IN THE CONSTRICTOR EFFECT OF SEROTONIN ON THE INTERNAL CAROTID ARTERIES

G. I. Mchedlishvili, P. Kometiani, and L. G. Ormotsadze

UDC 612.143 + 612.183].014.46: 612.018 + 546.41

The perfusion pressure in the internal carotid artery, isolated in situ, was recorded in experiments on dogs during perfusion of the artery with Ringer's solution by means of a constant output pump. The resting tone of the arterial wall rose with an increase in the Ca^{++} concentration in the perfusion fluid, and vice versa. The decrease in resting tone was particularly marked after elimination of Ca^{++} from the vessel wall by addition of EDTA to the perfusion fluid. The constrictor effect of serotonin on the internal carotid artery was considerably weakened if Ca^{++} was removed from the perfusion fluid and EDTA added to it.

The important role of the major arteries of the brain (internal carotid and vertebral) in the regulation of the blood supply to the brain and in the genesis of disturbances of blood supply due to spasm of these arteries [2], discovered in the last decade, compels detailed investigations to be made of the origin of the tone of these arteries in general and of their spasm in particular. Two modifications of the method using the internal carotid artery of a dog, isolated in situ [4, 5], provide excellent opportunities for research of this type.

Serotonin has been shown to be a specific endogenous constrictor agent in relation to the internal carotid artery [3, 5, 6], but the mechanism of its action has not yet been studied. This applies, in particular, to its role in the development of spasm (a sharp and persistent constriction) of these vessels. The need for a more detailed study of the effect of serotonin on the wall of the internal carotid artery will thus be understood.

Because of the important role of Ca^{++} ions in the manifestation of the serotonin effect on smooth-muscle elements [1, 7-10], the investigation described below was carried out to study the role of Ca^{++} in the formation of tone of the wall of the internal carotid artery and in the constrictor effect of serotonin on it.

EXPERIMENTAL METHOD

Experiments were carried out on 15 mongrel dogs weighing 15-25 kg, anesthetized with nembutal (about 0.04 g/kg, intraperitoneally). A preparation of the isolated internal carotid artery in situ [4] was used. All branches of the common carotid artery except the internal were ligated in the neck, after which the anastomoses between the internal carotid artery and the external orbital and inferior meningeal arteries were ligated. After wide trephining of the skull in the temporal region, the anterior cerebral and posterior communicating arteries were divided on the same side close to their origin from the internal carotid artery. One catheter was introduced into the common carotid artery in the direction towards the skull, and another (thin, polyethylene) was inserted in the retrograde direction inside the skull into the initial portion of the middle cerebral artery and passed along it to the internal carotid artery as far as the point

Department of Pathophysiology and Morphology and Department of Biochemistry, Institute of Physiology, Academy of Sciences of the Georgian SSR, Tbilisi. (Presented by Academician of the Academy of Medical Sciences of the USSR A. D. Ado.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 71, No. 6, pp. 3-5, June, 1971. Original article submitted October 27, 1970.

where it enters the skull. The internal carotid artery, with all its nervous communications intact, was thus hemodynamically isolated: the perfusion fluid injected by a pump from the common carotid artery passed only along the internal carotid artery and escaped without flowing through the other cerebral vessels.

The perfusion fluid consisted of Ringer's solution of the following composition, warmed to 37°C: NaCl 0.69%, KCl 0.035%, $CaCl_2$ 0.028%, KH_2PO_4 0.016%, $MgSO_4 \cdot 7H_2O$ 0.029%, $NaHCO_3$ 0.21%, glucose 0.05%; pH 7.4.

A constant output pump was used and the perfusion pressure, reflecting the hydraulic resistance of the internal carotid artery, was recorded. The investigated artery retained its sensitivity to physiologically active substances during several hours of the acute experiment.

EXPERIMENTAL RESULTS

During continuous perfusion of the dog's internal carotid artery, isolated in situ, the volume velocity (output) of flow of the Ringer's solution (V_{perf}) along the vessel was 13.5±1.2 ml/min (here and below, M±m), while the perfusion pressure (P_{perf}) averaged 100 mm Hg, i.e., the resistance in the vessel (R) was 8.5±1.05 mm Hg/ml/min. This resistance was due to the "resting tone" of the smooth-muscle fibers of the internal carotid arterial wall.

In the experiments of series I, after a stable background of perfusion pressure had been established during work with normal Ringer's solution, a Ringer's solution in which the Ca^{++} concentration was 2-4 times greater was passed through the artery (in all the experiments the perfusion fluid was isotonic; the salt composition was changed by reducing the concentration of Na^+ ions). The value of P_{perf} rose regularly (to 29.5±4.04 mm Hg), indicating an increase in tone of the vessel wall; the resistance increased under these conditions by 2.6±0.5 mm Hg/ml/min.

In the experiments of series II, after a period of perfusion of the vessel with normal Ringer's solution, the artery was perfused with Ca^{++}-free Ringer's solution. In this case as a rule the value of P_{perf} fell (on the average by 13.2±2.6 mm Hg) and R fell by 2.2±0.7 mm Hg/ml/min, indicating a decrease in tone of the smooth-muscle fibers of the arterial wall.

In the experiments of series III the artery was perfused (after establishment of the background) with Ca^{++}-free Ringer's solution with the addition of EDTA (ethylenediamine tetraacetate, a compound binding Ca^{++}) in a proportion of 40 mg to 100 ml perfusion fluid. Under these conditions the level of P_{perf} fell by a much greater degree than the experiments of series II (by 50.8±11.9 mm Hg), while R fell by 4.8±0.9 mm Hg/ml/min.

In all the series of experiments described above, after resumption of perfusion of the artery with normal Ringer's solution, the value of P_{perf} always had a tendency to return to its original level.

In the experiments of series IV the constrictor effects of serotonin during perfusion of the vessel with normal Ringer's solution (intra-arterial injection of serotonin under standard conditions in a dose of 0.4 μg in a volume of 2 ml) were compared with the effects of serotonin during the perfusion of the internal carotid artery with Ca^{++}-free Ringer's solution containing EDTA (40 mg/100 ml perfusion fluid). Whereas under the control conditions serotonin increased P_{perf} by 22.1±14.7 mm Hg and R by 3.4±1.1 mm Hg/ml/min, with Ca^{++} eliminated or its concentration significantly lowered in the vessel wall, the effect of serotinin was reduced on the average by 89%.

This weakening of the constrictor effect was specific for serotonin, for when vasopressin (0.1-0.01 unit) was injected into the artery or when Ringer's solution in which the K^+ concentration was increased 4-5 times was used to perfuse the vessel, constriction of the wall of the internal carotid artery always developed in about the same intensity as in the control.

The presence of Ca^{++} thus plays an important role in the formation of tone of the wall of the internal carotid artery. It is essential for the appearance of the vasoconstrictor effect of serotonin, the most active endogenous vasoconstrictor agent for this artery.

LITERATURE CITED

1. M. D. Kurskii and A. N. Fedorov, Uspekhi Sovr. Biol., 67, 190 (1969).
2. G. I. Mchedlishvili, Function of the Vascular Mechanisms of the Brain [in Russian], Leningrad (1968).

3. G. I. Mchedlishvili, P. A. Kometiani, and L. G. Ormotsadze, Pat. Fiziol., No. 3, 72 (1970).
4. G. I. Mchedlishvili, in: Cerebral Blood Flow, Berlin (1969), p. 98.
5. G. I. Mchedlishvili, L. G. Ormotsadze, and G. V. Amashukeli, Byull. Éksperim. Biol. i Med., 11, 3 (1967).
6. L. G. Ormotsadze, V. M. Samvelyan, L. S. Nikolaishvili, et al., Krovoobrashchenie, No. 4, 16 (1968).
7. M. E. Carsten, J. Gen. Physiol., 53, 414 (1969).
8. H. Gainer, J. Gen. Physiol., 52, 88 (1968).
9. J. Kozak and N. Lang, Nature, 197, 293 (1963).
10. A. P. Somlyo and A. V. Somlyo, Pharmacol. Rev., 20, 197 (1968).