Experimental Prevention of Cerebral Vasculature Obstruction Produced by Ischemia*

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Marked increases in vascular resistance following ischemia have been demonstrated in the kidney, heart, adrenal gland, and hindlimb. In the brain, this impairment of circulation develops much earlier, becoming first apparent after only 5 to 7½ min of ischemia and involving more than 50% of the brain within 15 min. It is of particular importance in the brain as it may constitute the first irreversible change in the animal and lead directly to death. Previously reported investigations of this phenomenon, including light and electron microscopic studies of the vasculature, have indicated that the circulatory impairment results from shifts of water and electrolytes from plasma to perivascular cells with a resultant increase in blood viscosity and narrowing of vascular lumen.

Our experiments were undertaken to study factors that might modify this reaction and particularly to investigate measures that might prevent it. The effects of increasing serum osmolarity were investigated in detail. Other variables studied included production of cerebral vasodilation or vasoconstriction prior to ischemia by hypercapnia or hypocapnia; prevention of the presumed fall in pH during ischemia by increasing the pH and buffer capacity of the blood; prophylactic administration of glucocorticoids; and variation of arterial pressure in the post-ischemic period.

Significant protection against the vascular lesion was obtained in experiments in which serum osmolarity was increased with Mannitol or glucose and in experiments in which the cerebral vasculature was diluted prior to the ischemia by CO₂ inhalation.

Methods

The experiments were performed on New Zealand white rabbits weighing 2 to 3 kg. They were anesthetized with intravenous sodium pentobarbital, 25 mg/kg. A tracheostomy tube was inserted; following intravenous anectine, they were respired with a Harvard respirator using room air. Respiratory rate was adjusted on the basis of measurements of pH, pCO₂, and PO₂ made on samples of femoral arterial blood. The thorax was opened by splitting the sternum, and the ascending and descending portions of the aorta were exposed. Ligatures were placed loosely around the ascending aorta just above the aortic ring and around the descending aorta (Fig. 1).

Cerebral ischemia was produced by tightening the ligature around the ascending

FIG. 1. Experimental cerebral ischemia model.
aorta. During the ischemic period, the ascending aorta was incised just distal to the ligature but well proximal to the innominate artery, and a cannula of about the same size as the aorta was inserted and secured with a second ligature. This provided the means for perfusing the cerebral vasculature with a suspension of carbon in order to determine its degree of patency. The descending aorta was ligated and ligatures were placed at the base of each ear to reduce the portion of the vascular tree to be perfused with carbon. At the end of the period of ischemia, the rostral portion of the rabbit (excluding the ears) was perfused with a carbon suspension for 30 sec at 110 mm Hg pressure. The carbon suspension, the same as that used in previous studies, consisted of 10% soot, 9.5% gelatin, and 1.3% phenol (prepared by Pelikan Werke, Hannover, W. Germany).

At the end of the carbon perfusion, the animals were sacrificed. The brain was removed, fixed in 10% formalin, and cut into six coronal sections as follows: 1) mid-frontal lobe; 2) optic chiasm; 3) anterior edge of mammillary body; 4) posterior edge of mammillary body; 5) inferior colliculus; and 6) mid-cerebellum. The sections were examined under a dissecting microscope and scored for the percentage of the surface not perfused with carbon. The percentage figures thus obtained for each of the six sections were then averaged to obtain a total figure for the extent of vascular obstruction in the entire brain. These total figures were then used to compare results obtained under different experimental conditions.

Measurements were made on a total of 66 experimental animals divided into 12 groups as follows:

**Group 1 (4 rabbits).** Controls for the ischemia. The brains of these animals were examined after periods of ischemia of less than 5 min.

**Group 2 (6 rabbits).** Controls for the effects of experimental changes introduced before or after a 15-min ischemic period. These animals were subjected to no additional experimental variables.

All of the animals in the following groups were also subjected to 15 min of ischemia:

**Group 3 (6 rabbits).** Serum osmolarity increased with Mannitol. A solution of 20% Mannitol was infused into the left ventricle immediately before the aorta was occluded. Serum osmolarity of jugular venous blood, sampled at the end of the infusion, was between 404 and 497 mOsm/liter.

**Group 4 (6 rabbits).** Serum osmolarity increased with glucose. In one animal, 20% glucose was infused into the ventricle (as above). In five animals, 75 ml of 20% glucose (three with 40 units of insulin and three without) was administered intravenously over a 15-min period preceding the ischemia. Blood glucose at the time of ischemia was between 785 and 1700 mg%, and serum osmolarity was between 347 and 380 mOsm/liter.

**Group 5 (2 rabbits).** As a control for the hemodilution (about 12%) produced in Groups 3 and 4 above, isotonic saline was infused into the left ventricle at the same rate as the Mannitol or glucose infusions.

**Group 6 (4 rabbits).** Hypoglycemia. Regular insulin (20 units/kg) was administered intravenously 30 min before ischemia. Blood glucose levels were between 20 and 40 mg% when ischemia was produced.

**Group 7 (6 rabbits).** Hypercapnia. The rabbits were respired with a mixture of 20% \( \text{CO}_2 \), 20% \( \text{O}_2 \), 60% \( \text{N}_2 \) for 10 min before ischemia. Arterial blood analyses demonstrated the \( p\text{CO}_2 \) between 100 and 125 mm Hg and \( p\text{O}_2 \) between 98 and 120 mm Hg.

**Group 8 (4 rabbits).** Hypocapnia. The animals were hyperventilated for 10 min before ischemia. Arterial blood analyses demonstrated the \( p\text{CO}_2 \) between 15 and 20 mm Hg and \( p\text{O}_2 \) between 105 and 307 mm Hg.

**Group 9 (3 rabbits).** Hypoxia. The animals were respired at normal rates for 6 min before ischemia with 100% \( \text{N}_2 \). Arterial blood analyses demonstrated the \( p\text{O}_2 \) between 5 and 15 mm Hg.

**Group 10 (7 rabbits).** Glucocorticoids. Methylprednisolone, 40 mg, was administered intravenously from 45 min to 2 hours before ischemia.

**Group 11 (3 rabbits).** Prevention of acidosis. Isotonic tris acetate, 50 ml at pH