Effects of Hemorrhagic Hypotension on the Cerebral Circulation

III. Neuropathology

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SUMMARY A neuropathological examination was made of 15 cats anesthetized with alpha-chloralose, subjected to a stepwise reduction in mean arterial pressure by graded hemorrhagic hypotension. Five animals were sacrificed by perfusion-fixation at a mean arterial pressure of 75 mm Hg, 5 at a mean pressure of 35 mm Hg, and 5 at 25 mm Hg. Ischemic brain damage was seen in only 5 animals, one sacrificed at a mean arterial pressure of 35 mm Hg and 4 at a pressure of 25 mm Hg. It was maximal in the selectively vulnerable areas and included the boundary zones between the major arteries of the cerebral hemispheres, the Ammon's horn and the thalamus.

IN ACCOMPANYING PAPERS of this series local cerebral blood flow and the caliber of pial vessels, as well as the EEG and evoked cortical potential are related to changes in mean arterial pressure induced by graded hemorrhagic hypotension in the cat. Here, using the same model, the changes in cerebrovascular response and electrocortical function are correlated with the occurrence of ischemic brain damage.

Methods

General Description

The experiments were carried out on 15 cats of either sex, weighing between 1.5 and 4.3 kg. The animals were prepared as previously described and maintained at normocapnia (arterial PCO₂ ≈ 32 mm Hg) and normoxia, and with a stable body temperature of 37-38°C. Neither cerebral blood flow nor electrocortical function were measured in these animals.

Mean arterial pressure was measured continuously via one aortic catheter. A progressive stepwise reduction in mean arterial pressure was achieved over a 2 hour period by bleeding the cats into a heparinized reservoir. Mean arterial pressure was held at the desired level for at least 20 min. The animals were divided into 3 groups: group 1 in which the mean arterial pressure was lowered to 75 mm Hg; group 2 in which the mean arterial pressure was lowered to 35 mm Hg; and group 3 in which it was lowered to 25 mm Hg. The total period of hemorrhagic-induced hypotension ranged between 2 and 3 h.

Neuropathology

At the end of the procedure, i.e. some 20 min after the desired level of mean arterial pressure had been reached, the animals were heparinized (1000 I.U./kg) and perfusion-fixed with 1,000 ml of FAM fixative (40% formaldehyde — glacial acetic acid — absolute methanol = 1:1:8). After perfusion the cats were decapitated and the head was stored in the same fixative at 4°C for at least 4 h. After removing the brain, the hindbrain was detached by a cut through the midbrain and the cerebral hemispheres were cut into 6 mm coronal slices. The brain stem was cut perpendicular to its long axis into 6 mm slices and the cerebellum into 2 slices perpendicular to the folia of the dorsal surface of each hemisphere. Large representative bilateral blocks of brain were embedded in paraffin wax and sections (7-8 μm thick) were stained by a method combining cresyl violet and Luxol fast blue and by hemalum and eosin. The sections were examined by conventional light microscopy.

Results

As judged by the uniform hardening of the specimens and by the absence of blood in the vessels, perfusion fixation appeared to be good in all cats. There was no evidence of post-hypoxic brain swelling and internal herniation was not seen. Cytological artifacts, the "dark cell" and "hydropic cell or water change" were not seen.

Histological examination showed small foci of ischemic damage in the brains of 5 cats — none in group 1; one in group 2 and 4 in group 3 (table 1). The lesions appeared as small areas of selective neuronal necrosis comprising microvacuolation and ischemic cell change (fig. 1) as previously described in FAM-fixed material.

The ischemic damage was not distributed randomly but was accentuated in the "selectively vulnerable areas" (table 2), namely in the boundary zones of the cerebral cortex between the distributions of the common pericallosal and middle cerebral arteries (5 animals), in the upper portion of the striatum (3 animals) and in the Purkinje cells of the cerebellum (one animal). The brain stem was normal in all animals. The pattern of ischemic damage in one of the
NEUROPATHOLOGY OF HYPOTENSION IN CEREBRAL CIRCULATION/Graham et al.

TABLE 1

<table>
<thead>
<tr>
<th>Group (Number of animals)</th>
<th>Mean arterial pressure (mm Hg)</th>
<th>Ischemic brain damage</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 (5)</td>
<td>75</td>
<td>5</td>
</tr>
<tr>
<td>2 (5)</td>
<td>35</td>
<td>4</td>
</tr>
<tr>
<td>3 (5)</td>
<td>25</td>
<td>1</td>
</tr>
</tbody>
</table>

The finding of microvacuolation and ischemic cell change at the end of the 2–3 hour period of graded hypotension in some of the animals suggests that their response to low blood pressure was similar to that described in the FAM perfused and fixed brain of the rat and monkey.4, 8

Clinical evidence7, 8 and experimental studies on subhuman primates8 have shown that a major and abrupt reduction in the perfusion pressure of the brain supplied with normally oxygenated blood can result in ischemic damage within the boundary zones between territories of the major arteries of the cerebrum and cerebellum with variable involvement of deeper structures. Because of the profound fall in arterial pressure there is a failure of autoregulation and a severe reduction in cerebral blood flow in the regions most removed from the parent arterial stems, i.e. in the boundary zones. If the rate of fall of blood pressure is slower, then ischemic brain damage is not confined to the arterial boundary zones but becomes more generalized.7, 10

group 3 animals is shown in fig. 2. In none of the animals was there any evidence of vessel stenosis or occlusion.

Discussion

As in other models of hypoxia, ischemic cell change5 characterized the type of neuronal damage found in the present study. Even though the time course of neuronal necrosis was not investigated, the

TABLE 2

<table>
<thead>
<tr>
<th>Group (Animals with IBD)</th>
<th>CPA/MCA BZ</th>
<th>Ammon's Horn</th>
<th>Striatum</th>
<th>Thalamus</th>
<th>Cerebellum</th>
</tr>
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<tbody>
<tr>
<td>1 (0)</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2 (1)</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>3 (4)</td>
<td>4</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>0</td>
</tr>
</tbody>
</table>

IBD = Ischemic brain damage; CPA = Common pericallosal artery; MCA = Middle cerebral artery; BZ = Boundary zone.

FIGURE 1. Ischemic cell change in cortex of arterial boundary zone. The ischemic nerve cells are small and triangular and contain hyperchromatic nuclei (arrows). The cytoplasm was Luxolophilia. Combined Luxol fast blue and cresyl violet × 670.
Coronal Sections Of Cat Brain

From the first paper in this series it can be seen that as mean arterial pressure was reduced, cerebral blood flow remained constant until arterial pressure was reduced to 60-69 mm Hg. At a mean arterial pressure below 65 mm Hg, cerebral blood flow decreased with decreasing perfusion pressure. It was observed that at pressures of about 35 and 25 mm Hg, blood flow (as calculated from the initial slope index) had fallen to means of 28 and 10 ml/100g min respectively. A zero flow was also recorded in some electrodes over the pressure range of 18-49 mm Hg, and we have argued that this might be due to placement of the electrodes in the boundary zones between the territories of the common pericallosal and middle cerebral arteries, these regions being selectively vulnerable in severe hypotension. Intravascular sludging and stasis might account also for a zero flow, particularly at a mean arterial pressure of 35 mm Hg when the pial vessels were maximally dilated.

Cerebral blood flow was not measured in the animals used for neuropathological study, but by interpolation from the previous study it can be shown that ischemic brain damage occurred when flow had fallen to about 15-20% (8-11 ml/100g min) of control values (≈ 55ml/100g min). Presumably flow was reduced to critical levels in the single animal in group 2 with ischemic damage, and yet was not in the animal sacrificed at a mean arterial pressure of 25 mm Hg in which evidence of ischemic cell change was not found. It has not been possible from the available information in this study to give an adequate account for these exceptions. As the experimental profiles in these 2 animals were similar to the other animals in their respective groups the differences have been ascribed to biological variation.

In the parallel series of animals the evoked cortical potential remained essentially unchanged over the arterial pressure range of 120-40 mm Hg. At mean arterial pressures below 35-40 mm Hg the amplitude of the evoked potential decreased with a fall in arterial pressure and reached zero at 16-25 mm Hg. At a level of 30-35 mm Hg there was a marked slowing in the EEG in most animals. At lower levels of arterial pressure there was a progressive slowing of the EEG with further reduction in blood flow until the EEG became isoelectric at a flow close to zero. An isoelectric EEG was recorded over the pressure range of 10-30 mm Hg. Therefore, changes in the evoked cortical potential and in the EEG closely paralleled one another, and it seems likely that ischemic brain damage occurred only when there was marked loss of electrocortical function. It was decided not to study a further group of animals sacrificed between a mean pressure of 35-50 mm Hg as the cerebral blood flow and electrocortical data had suggested that the pathophysiology would not have become critical until the mean arterial pressure became 25-35 mm Hg.

As electrocortical function (either evoked or spontaneous) was not assessed in this group of animals, it is not possible to correlate directly either the evoked cortical potential or the EEG with the presence or absence of ischemic brain damage. However, our findings seem to be in general agreement with those of Brierley et al. who found in the normal rhesus monkey subjected to profound arterial hypotension that brain damage occurred only when the EEG became isoelectric and when the cerebral perfusion pressure had fallen rapidly to below 25 mm Hg and was sustained at this level for at least 15 min. It therefore seems reasonable to speculate that the EEG would not have been isoelectric in the animals in the present study in which ischemic brain damage was not identified.

Acknowledgment

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References
Correlated Electrical and Mechanical Responses of Isolated Rabbit Pial Arteries to Some Vasoactive Drugs


SUMMARY Simultaneous measurements were made of spike activity and perfusion pressure ($P_A$) in intact segments of rabbit middle cerebral artery in vitro. The segments were mounted on a Teflon tube designed so that the perfusing solution flowed in the annular space between the tube and the artery wall, thus magnifying the $P_A$ changes occurring when the artery constricted or dilated. A widened portion of the Teflon tube immobilized 1-2 mm of the artery segment for electrical recording with fine glass microelectrodes. Spontaneous spike activity (extra- and intracellular) was regularly observed. When a steady $P_A$ and spike discharge was obtained, tests were performed by substituting for the normal perfusion liquid, solutions containing 5 µg/ml norepinephrine, 5 µg/ml angiotensin II or 7.5 µg/ml isoproterenol. Norepinephrine and angiotensin each increased spike frequency (+293 and +126%) and $P_A$ (+6.6 and +7.9 mm Hg) whereas isoproterenol decreased spike frequency (−89%) and $P_A$ (−22.9 mm Hg). These results a) confirm the presence of receptors to these agents in pial arteries, and b) demonstrate a high degree of correlation between membrane electrical events and mechanical activity of these spontaneously-active myovascular cells.

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