Experimental Cerebral Ischemia in Mongolian Gerbils

III. Behaviour of the Blood-Brain Barrier


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Summary. The behaviour of the BBB in cerebral ischemia was studied in symptom-positive Mongolian gerbils subjected to left common carotid artery occlusion using Evans Blue dye as indicator of BBB injury. The BBB damage was demonstrable grossly by the presence of areas of blue discoloration, and microscopically by the presence of a bright red fluorescent tracer, localized mostly in the neurons.

The survey of various groups of animals revealed a direct relationship between the incidence and time of appearance of the BBB lesions and the duration of the ischemic occlusion. This relationship can be interpreted as another example of the previously described "maturation" phenomenon.

A relatively late occurrence of the BBB injury in cerebral ischemia, at the time when the affected brain tissue shows severe, edematous histopathologic changes indicates that the brain edema, as the main complication of ischemia, could be regarded as being primarily of the cytotoxic type.

Key words: Cerebral ischemia — Blood-brain barrier — Mongolian gerbils.

Introduction

The behaviour of the blood-brain barrier (BBB) in cerebral ischemia has been a controversial subject. Broman (1949) was the first to stress a remarkable resistance of the BBB to anoxic-ischemic injury noticing that the function of the BBB with regard to Trypan Blue dye remains preserved for many hours post-mortem, or for more than 1 hr following an arterial occlusion. In later studies Olsson et al. (1971) and Hossmann and Olsson (1971) similarly emphasized the resistance of the BBB in cerebral ischemia. On the other hand, Denny-Brown and Meyer (1957) and Meyer (1959) observed changes in the BBB early after occlusion of the middle cerebral artery in the monkey. Also Plum et al. (1963) reported early BBB lesions in Levine preparations.

One of the main complications of cerebral ischemia is the edematous change affecting the brain tissue subjected to it. In the classification of brain edema into vasogenic vs. cytotoxic as the two main types (Klatzo, 1967), the behaviour of the BBB represents the most significant criterion by which these types can be differentiated. Thus evaluation of the BBB in cerebral ischemia may provide an important

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insight into overall pathomechanisms of the process, as well as providing clues for a proper clinical management of patients with cerebral ischemia.

Mongolian gerbils (Meriones unguiculatus), in which occlusion of the common carotid artery in the neck leads frequently to ischemic infarctions in ipsilateral hemispheres, were used as the experimental model in this study. The possibility of using large numbers of animals divided into various groups according to the duration of ischemia and the time intervals following release of occlusion offered special advantages for broad evaluation of the dynamics of the BBB changes in cerebral ischemia.

Materials and Methods

The gerbils were operated under light anesthesia introduced by intraperitoneal injection of pentobarbital (3 mgm/100 g). The left common carotid artery was occluded at the neck for various periods of time with the Heiffetz clip. Only symptom-positive animals (Kahn, 1969) were used in this study. The BBB was assessed by intravenous injection of 0.1 ml/100 g of 2% Evans Blue dye either immediately before release of the clip, or 5 hrs before sacrifice in the group with a one-week release period. The animals were sacrificed by perfusion with 10% buffered paraformaldehyde and the abnormal permeability of the BBB to Evans Blue tracer was assessed by visual inspection and photography of the perfused brains sectioned into coronal blocks. In some instances fluorescence microscopic observations were carried out on 10 μ frozen sections prepared from the paraformaldehyde-fixed tissue. The number of animals in each group defined by different occlusion and release periods, as well as the number of gerbils in these groups which showed abnormal permeability of the BBB, expressed by blue discoloration of the brain tissue, is indicated in Table 1.

Table 1. The groups of gerbils defined by different durations of occlusion and release time. The first number refers to the number of animals showing BBB damage, the second number denotes the total number of animals in a particular group

<table>
<thead>
<tr>
<th>Duration of Occlusion</th>
<th>5 sec</th>
<th>15 min</th>
<th>30 min</th>
<th>1 hr</th>
<th>2 hrs</th>
<th>3 hrs</th>
<th>5 hrs</th>
<th>20 hrs</th>
<th>1 week</th>
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<tbody>
<tr>
<td>15 min</td>
<td>0/4</td>
<td>0/5</td>
<td>0/5</td>
<td>0/6</td>
<td>0/6</td>
<td>0/7</td>
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<tr>
<td>30 min</td>
<td>0/4</td>
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<td>0/5</td>
<td>3/6</td>
<td>0/5</td>
<td></td>
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<td></td>
<td></td>
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<tr>
<td>1 hr</td>
<td>0/5</td>
<td>0/4</td>
<td>0/6</td>
<td>2/7</td>
<td>6/6</td>
<td>1/5</td>
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<td>3 hrs</td>
<td>0/3</td>
<td>0/5</td>
<td>1/5</td>
<td>3/5</td>
<td>4/5</td>
<td>5/5</td>
<td>5/6</td>
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<tr>
<td>6 hrs</td>
<td>1/6</td>
<td>4/6</td>
<td>5/6</td>
<td>7/7</td>
<td>3/3</td>
<td>2/2</td>
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Results

The BBB injury was demonstrable only on the side of carotid occlusion. Under visual inspection, the abnormal permeability of the BBB was conspicuous by regions of blue discoloration which ranged from small perivascular exudations to rather extensive areas involving large portions of cerebral cortex, hippocampus and basal ganglia (Fig. 1). The frequency and intensity of the BBB damage were greatest in the basal ganglia, less severe in the hippocampus and least in the cerebral cortex. The incidence of BBB damage in various groups of gerbils is evident in Table 1 and it is also represented graphically in percentages in Fig. 2. As these data indicate, gerbils in which the unilateral carotid occlusion lasted