Histopathologic and Morphometric Studies of Leptomeningeal Vessels in Moyamoya Disease

Shinji Kono, MD, Kazunari Oka, MD, and Katsuo Sueishi, MD

To clarify the morphogenesis of vascular anastomoses in the leptomeninges, we examined the leptomeningeal vessels of six autopsied patients with moyamoya disease who died of cerebral hemorrhage. Sites examined in the brain (the frontal pole, frontoparietal convexity, and lateral occipital region) were selected by the presence of angiographic abnormalities on the convexity of the hemisphere. At all examined sites, neither the vascular density (number of blood vessels per unit length of underlying cortical surface) nor the arterial/venous ratio differed significantly between the patients and 14 controls. All patients showed dilatative changes of both arteries and veins; however, attenuation or disruption of the internal elastic lamina and fibrous intimal thickening were more prominent in those patients who had had moyamoya disease longer. Our results indicate that the leptomeningeal vessels of moyamoya disease are not newly formed but are merely dilated preexisting vessels and that the structural alteration of the vascular walls suggests their adaptability for participation in the collateral circulation at the cerebral surface. (Stroke 1990;21:1044–1050)

In moyamoya disease, leptomeningeal vessels have been thought to be important collateral channels for the ischemic brain, as have perforating vessels in the basal ganglia. These vessels participate in leptomeningeal anastomoses among the three main cerebral arteries and in transdural anastomoses from the external carotid artery via a rete mirabile. These anastomoses are frequently observed as abnormal vascular networks on cerebral angiographs. The pathophysiological role of leptomeningeal vessels has become more important in improving the prognosis of patients with moyamoya disease as the result of several kinds of extracranial-intracranial bypass operations that have recently been used to increase collateral circulation to the ischemic brain from the external carotid system. Several authors have described the pathologic changes of leptomeningeal vessels, but none have shown sufficient evidence as to whether these abnormal vessels were preexistent or newly formed. We took a morphometric approach to elucidate this problem.

Subjects and Methods

Six patients with moyamoya disease autopsied between 1979 and 1983 were collected from four hospitals in western Japan. Cerebral angiographs performed 1 week to 2 years before death were available in four patients. Angiography revealed nine abnormal vascular networks on the convexity of the hemisphere: three at the frontal pole, five at the frontoparietal convexity, and one at the lateral occipital region (Table 1, Figure 1). These predilectional sites were chosen for the histopathologic examinations. Fourteen controls who had no cerebrovascular alterations related to cause of death were selected from the autopsy file of Kyushu University Hospital (Table 1). The patient and control groups were divided by age into juvenile (those <20 years old) and adult (those >20 years old) subgroups.

We obtained sections of the cerebral cortex with an intact arachnoid membrane from the sites mentioned above, embedded them in paraffin, and routinely stained them with hematoxylin and eosin, Van Gieson's elastic stain, and Victoria blue. Areas of subarachnoid hemorrhage or cerebral infarction were avoided because it was difficult to identify leptomeningeal vessels within hemorrhage and the cortical surface in reactive gliomesodermal tissue. We counted all blood vessels in the subarachnoid space and measured vascular diameter at the maximal width of the lumen perpendicular to the long axis. The few extremely irregular vessels were eliminated from analysis.

Vascular density of the leptomeningeal vessels in each section was calculated as the number of blood...
vessels in the subarachnoid space per unit length of the underlying cortical surface and is expressed as number per centimeter. The length of cortical surface was measured with a digital analyzer (Cosmozone 98, Nikon, Tokyo, Japan) connected to a microcomputer (PC-9801F, NEC Corporation, Tokyo).

We counted the number of arteries and veins separately and calculated the ratio of arteries to veins (arterial/venous ratio) at each site in every case. Arteries were differentiated from veins by the presence in the former of an internal elastic lamina stained with Van Gieson's elastica stain or Victoria blue; however, some vessels <20 μm in diameter could not be classified. These small vessels were excluded from analysis of the arterial/venous ratio.

We compared both vascular density and arterial/venous ratio of the moyamoya patients and the controls using Student's unpaired t test. Since the vascular diameters had a positively skewed distribution, we made a logarithmic transformation. After logarithms of the diameters were proven to have an approximately normal distribution, we compared the log-transformed diameters of the moyamoya patients and the controls using Student's unpaired t test.

**Results**

There were 7,202 leptomeningeal vessels in the moyamoya patients and 25,201 in the controls. The diameter of these vessels ranged from a few to approximately 2,000 μm.

Mean±SD vascular density of the controls at each site is given in Table 2. To examine the influence of age on vascular density, juvenile controls and adult controls were also analyzed separately; the subgroups showed quite similar values (Table 2). Mean±SD vascular density of the moyamoya patients is also given in Table 2. There was no significant difference in vascular density between the moyamoya patients and the controls at any site.

Neither vascular density nor the arterial/venous ratio had any relation with age or the clinical period of the patients, and no particular findings were noted in sections from the angiographically abnormal regions.

The distribution of leptomeningeal vessels according to diameter is shown in Figure 2. Nearly half of the vessels in the controls were ≤20 μm in diameter, and vascular density decreased sharply with the
FIGURE 1. Left external carotid angiograph of case 2 with moyamoya disease. Three abnormal vascular networks are shown, one at frontal pole (a) and two at frontal convexity (b and c).

increase in diameter. In the moyamoya patients, vascular density of vessels ≤20 μm in diameter was lower than that of the controls, and density of the larger vessels was correspondingly greater at all sites, except for vessels 20–40 μm in diameter at the frontoparietal convexity. Mean±SD vascular diameter in the moyamoya patients was significantly greater than that in the controls at all sites (Table 4).

After classifying the vessels >20 μm in diameter into arteries and veins, both groups showed a similar distribution of vascular density (Figure 3), indicating that vascular dilatation with moyamoya disease occurred in both arteries and veins. Furthermore, when the moyamoya patients and controls were separately divided into juvenile and adult subgroups, the distributions of arterial and venous densities were still similar in patients and controls (Figure 4).

The walls of vessels in the moyamoya patients were thinner than those of the same sized vessels in the controls. We observed fibrous intimal thickening and disruption of the internal elastic lamina in arteries of the moyamoya patients but not the controls. Moreover, these changes were more prominent and widespread in those moyamoya patients with longer clinical periods (Figure 5). Fibrin deposits, microaneurysms, and thrombus formation were seen in neither the moyamoya patients nor the controls.

Discussion
While some investigators have referred to it in their pathologic findings, the histogenesis of abnormal leptomeningeal vessels in patients with moyamoya disease is still unsolved. It seems reasonable to consider that if these vessels were newly formed, either the vascular density or the arterial/venous ratio would be altered.

To quantify changes of leptomeningeal vascularity on the cortical surface, we defined the term "vascular

<table>
<thead>
<tr>
<th>TABLE 2. Vascular Density at Three Sites in Patients with Moyamoya Disease and in Controls</th>
</tr>
</thead>
<tbody>
<tr>
<td>Site</td>
</tr>
<tr>
<td>-----------------------------------------------</td>
</tr>
<tr>
<td>Frontal pole</td>
</tr>
<tr>
<td>Frontoparietal convexity</td>
</tr>
<tr>
<td>Lateral occipital region</td>
</tr>
</tbody>
</table>

Data are mean±SD blood vessels per centimeter of underlying cortical surface. Juvenile, <20 years of age; adult >20 years of age. No significant differences by Student's t test for unpaired data.
TABLE 3. Arterial/venous Ratio of Leptomeningeal Vessels >20 μm in Diameter at Three Sites in Patients With Moyamoya Disease and in Controls

<table>
<thead>
<tr>
<th>Site</th>
<th>Patients (n=6)</th>
<th>All (n=14)</th>
<th>Juvenile (n=4)</th>
<th>Adult (n=10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Frontal pole</td>
<td>1.65±0.36</td>
<td>1.92±0.58</td>
<td>1.80±0.65</td>
<td>1.98±0.61</td>
</tr>
<tr>
<td>Frontoparietal convexity</td>
<td>1.59±0.20</td>
<td>1.87±0.46</td>
<td>1.61±0.51</td>
<td>1.98±0.45</td>
</tr>
<tr>
<td>Lateral occipital region</td>
<td>1.97±0.63</td>
<td>1.96±0.46</td>
<td>2.17±0.80</td>
<td>1.88±0.28</td>
</tr>
</tbody>
</table>

Data are mean±SD. No significant differences by Student’s t test for unpaired data.

density" as the number of leptomeningeal vessels per unit length of underlying cortical surface. This method was applied to all specimens except those with infarction or hemorrhage, in which structures of the cortical surface and individual vessels were too obscure to be identified. Standard deviations of the vascular densities of the 14 controls at the three examined sites were fairly small, ensuring that vascular density calculated in this way was a useful parameter.

We differentiated arteries from veins by the existence in the former of an internal elastic lamina stained with Victoria blue or Van Gieson’s elastica stain. The elastic fibers in vessels ≤20 μm in diameter were so delicate that differentiation of these small vessels was impossible, although they comprised nearly half of the vessels in the controls and one third to one fifth of those in the moyamoya patients.

In the moyamoya patients, neither vascular density nor the arterial/venous ratio were significantly different from those of the controls at any site. Furthermore, there were fewer smaller vessels and correspondingly more larger ones in the patients than in the controls. These findings suggest that the leptomeningeal vessels in moyamoya disease are not newly-formed but are merely dilated preexisting vessels.

We also found that the fibrous intimal thickening and alterations of the internal elastic lamina were more prominent and frequent in moyamoya patients with longer clinical periods. An 8-year-old girl with a 7-month clinical history of the disease showed only focal disruption of the internal elastic lamina of a few arteries and almost no intimal thickening, while dilatation of the vessels in this patient occurred as frequently as in other moyamoya patients. The histologic changes of the intima and internal elastic lamina appeared to be secondary to dilatation. This progressive thickening of the intima might partly

TABLE 4. Logarithmically Transformed Values of Vascular Diameter at Three Sites in Patients with Moyamoya Disease and in Controls

<table>
<thead>
<tr>
<th>Site</th>
<th>Patients (n=6)</th>
<th>Controls (n=14)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Frontal pole</td>
<td>1.58±0.38*</td>
<td>1.38±0.39</td>
</tr>
<tr>
<td>Frontoparietal convexity</td>
<td>1.75±0.40*</td>
<td>1.43±0.41</td>
</tr>
<tr>
<td>Lateral occipital region</td>
<td>1.68±0.38*</td>
<td>1.36±0.40</td>
</tr>
</tbody>
</table>

Data are mean±SD μm.

*p<0.001 different from control by Student’s t test for unpaired data.

FIGURE 2. Histograms showing distribution of vascular density by diameter of leptomeningeal vessels at (top) frontal pole, (middle) frontoparietal convexity, and (bottom) lateral occipital region of six patients with moyamoya disease (shaded bars) and 14 controls (open bars). There are fewer vessels ≤20 μm in diameter and correspondingly more larger vessels at all sites in patients with moyamoya disease than in controls.
FIGURE 3. Histograms showing distribution of vascular density by diameter of leptomeningeal arteries and veins >20 μm in diameter at (top) frontal pole, (middle) frontoparietal convexity, and (bottom) lateral occipital region of six patients with moyamoya disease (shaded bars) and 14 controls (open bars). Both arteries and veins show similarly greater densities in moyamoya patients than in controls at each site except arteries 20–40 μm in diameter at frontal pole and frontoparietal convexity.

Histologic alterations of the leptomeningeal vessels differ greatly from those of the perforating arteries in moyamoya disease. Neither focal fibrin deposits nor the formation of microaneurysms, thought to be advanced lesions predisposed to rupture, are observed in leptomeningeal vessels, as shown in our study. This finding is thought to be fully consistent with the fact that most intracranial hemorrhages in patients with moyamoya disease are intracerebral and that rupture of the abnormal vessels in the subarachnoid space is rare.

Glionesodermal tissue containing many capillaries was observed near infarcted regions of the cerebrum. We excluded such regions from our analysis because the cortical surface was too obscure to measure its length. However, there is a possibility that the abnormal vascular networks observed on the angiographs arose from these capillaries. Further investigation is necessary to clarify how these capillaries might work as collateral channels to the adjacent ischemic brain.

In conclusion, the leptomeningeal vessels in patients with moyamoya disease were histologically characterized by the dilatation of preexisting arteries and veins and accompanied by intimal thickening and alterations of the internal elastic lamina as the clinical
period lengthened. Therefore, these dilated leptomeningeal vessels may participate in collateral circulation at the cerebral surface.

Acknowledgments

We wish to thank the following pathologists who contributed their autopsied cases to this investigation: Professor E. Tahara (Hiroshima), Professor T. Yumoto (Tottori), Dr. F. Uchino (Kokura), Dr. C. Yutani (Osaka), and Dr. T. Fukuhara (Hiroshima). We thank Professor Emeritus Kenzo Tanaka, MD, for scientific comments. We also thank Mr. B.T. Quinn for comments on the manuscript.

References


KEY WORDS • collateral circulation • moyamoya disease
Histopathologic and morphometric studies of leptomeningeal vessels in moyamoya disease.
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*Stroke*. 1990;21:1044-1050
doi: 10.1161/01.STR.21.7.1044

*Stroke* is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
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Print ISSN: 0039-2499. Online ISSN: 1524-4628

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