Possible Role for Vascular Cell Proliferation in Cerebral Vasospasm After Subarachnoid Hemorrhage

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Background and Purpose—During vasospasm after subarachnoid hemorrhage (SAH), cerebral blood vessels show structural changes consistent with the actions of vascular mitogens. We measured platelet-derived vascular growth factors (PDGFs) in the cerebrospinal fluid (CSF) of patients after SAH and tested the effect of these factors on cerebral arteries in vivo and in vitro.

Methods—CSF was sampled from 14 patients after SAH, 6 patients not suffering SAH, and 8 normal controls. ELISA was performed for PDGF-AB, transforming growth factor-β1, and vascular endothelial growth factor. A mouse model was used to compare cerebral vascular cell proliferation and PDGF staining in SAH compared with sham-operated controls. Normal human pial arteries were incubated for 7 days in vitro, 2 groups with human blood clot and 1 with and 1 without PDGF antibodies.

Results—PDGF-AB concentrations in CSF from SAH patients were significantly higher than those from non-SAH patients and normal controls, both during the first week after SAH and for all time points measured. Smooth muscle and fibroblast proliferation was observed after SAH in the mouse model, and this cellular replication was observed in conjunction with PDGF protein at the sites of thrombus. In human pial arteries, localized thrombus stimulated vessel wall proliferation, and proliferation was blocked by neutralizing antibodies directed against PDGFs.

Conclusions—Vascular mitogens are increased in the CSF of patients after SAH. Proliferation of cells in the vascular wall is associated with perivascular thrombus. Cellular proliferation and subsequent vessel wall thickening may contribute to the syndrome of delayed cerebral vasospasm. (Stroke. 2003;34:427-433.)

Key Words: cerebral arteries ■ cerebral vasospasm ■ growth factors ■ subarachnoid hemorrhage

Among patients with subarachnoid hemorrhage (SAH), delayed cerebral ischemia and infarction are the leading causes of death and major disability in patients surviving to hospitalization.1 The most common cause of delayed ischemia is cerebral arterial vasospasm.2-4 The prevention and treatment of vasospasm have emerged as major goals in the management of patients surviving SAH, but the exact etiology of vasospasm remains unknown.

Rupture of an intracranial aneurysm releases arterial blood into the subarachnoid space. Coagulation of subarachnoid blood activates platelets, which release potent growth factors for cells in the vascular wall.5 Among these growth factors are platelet-derived growth factor-AB (PDGF-AB), transforming growth factor-β1 (TGF-β1), and vascular endothelial growth factor (VEGF). The PDGFs and TGF-β are potent mitogens for smooth muscle cells in the vascular media and fibroblasts in the adventitia,6 whereas VEGF stimulates proliferation of vascular endothelium.7 Previous studies have shown that growth factors from platelets are increased in the cerebrospinal fluid (CSF) of patients with SAH.8,9

Cerebral blood vessels that are affected by vasospasm exhibit structural changes that are consistent with the actions of vascular mitogens. Smooth muscle and myofibroblast proliferation, as well as cellular necrosis and remodeling, are common features of vasospastic segments.8,9 Intimal hyperplasia, as well as collagen deposition and fibrosis, has also been extensively described.2,11 These changes may contribute to arterial wall thickening, and decreased vessel compliance after SAH. We hypothesized that growth factors, released from platelets, mediate vascular cell proliferation in cerebral arteries after SAH. Cellular proliferation and increased vessel wall thickness may in turn cause vascular stiffening that contributes to cerebral vasospasm. In this study, we asked whether increases in CSF growth factors and vascular cell proliferation occurred after SAH. The results show that CSF vascular mitogens are increased after SAH and that these mitogens are associated with significant vascular cell proliferation in vivo and in vitro.
Methods

CSF Analysis and Biopsy of Cerebral Arteries

Institutional review board approval was granted for human studies, and patients or families gave informed, written consent to participate. CSF was collected from lumbar or ventricular drainage devices after SAH (SAH patients) or after head injury, obstructive hydrocephalus, or intraparenchymal cerebral hemorrhage (non-SAH patients). Control samples were obtained from normal, healthy volunteers undergoing atraumatic lumbar puncture in a separate institutional review board–approved study (normal controls).

CSF samples were collected from the drainage apparatus by a sterile sampling technique. The dead-space volume from the drainage tubing was discarded before collecting 1 to 6 mL of fresh CSF in a polystyrene centrifuge tube. CSF samples were centrifuged at 270g for 15 to 30 minutes at room temperature to remove red blood cells, nonactivated platelets, and other cellular debris. The supernatant was then frozen at −80°C for later analysis by ELISA.

To collect segments of pial cerebral arteries, patients who were undergoing temporal lobectomy for intractable seizures were enrolled. Normal segments of pial artery overlying the resected brain parenchyma (3- to 4-mm lengths of artery, otherwise discarded) were dissected free of underlying brain tissue. The pial artery segments were stored immediately in culture medium at 37°C. To obtain fresh human blood for culture with human pial arteries in vitro, we collected 5-mL samples of venous blood from a single volunteer.

ELISA of CSF Samples

CSF samples were thawed from −80°C to room temperature and then assayed in duplicate for the presence of basic fibroblast growth factor (bFGF), PDGF-AB, and VEGF. ELISA procedures were conducted according to manufacturer’s instructions (R&D Systems).

Mouse Model of Endovascular Injury and SAH

The Duke University Animal Care and Use Committee approved all of the following animal experiments. Male C57BL/6J mice (8 to 10 weeks old; Jackson Laboratory, Bar Harbor, Me) were fasted for 12 hours to control blood glucose concentration. Anesthesia was induced with 5% halothane in 50% O2/balance N2. The tracheas were intubated and the lungs were mechanically ventilated to maintain normocapnia and moderate hyperoxemia. A femoral artery was cannulated for measurement of blood pressure and arterial blood gases. Percutaneous temperature was continuously monitored and the right common carotid artery was exposed by midline incision of the neck, and the external carotid artery was isolated and ligated. A blunted 5-0 monofilament nylon suture was introduced into the external carotid artery and advanced by way of the internal carotid artery distal to the right anterior cerebral artery (ACA)–middle cerebral artery bifurcation, where resistance was encountered, and then advanced 3 mm farther to perforate the right ACA. The suture was immediately withdrawn, allowing reperfusion and SAH. Sham-operated mice underwent identical procedures, except that the ACA was not perforated. After removal of the filament, the skin was closed with sutures and halothane anesthesia was discontinued. On recovery of spontaneous ventilation, the trachea was extubated. Mice were continuously observed until recovery of the righting reflex and were then returned to their cages. Eighteen mice were included in this study (SAH n=9; sham n=9).

Mouse Brain Harvesting

At 3, 24, or 72 hours after SAH, 3 mice from each group were anesthetized with halothane. The tracheas were intubated and the lungs mechanically ventilated. The chest was opened and the aorta was cannulated with a blunted, 23-gauge needle through the left ventricle. An incision was made in the right atrium to allow for outflow of perfusion solutions. Twenty milliliters of normal saline was infused, followed by 4% paraformaldehyde for 10 minutes. All perfusates were delivered at 60 to 80 mm Hg. Cadavers were refrigerated at 4°C for 3 hours. The brains were then harvested and placed in 70% alcohol. Brains were dehydrated, paraffin embedded, and sectioned.

Histological Analyses

To assess cellular proliferation, 5-mm coronal sections were deparaffinized and then immunostained for the presence of proliferating cell nuclear antigen (PCNA, DAKO). Primary antibody was applied at 1:300 dilution, followed by secondary staining with a Vectastain mouse kit (Vector Laboratories). A methyl green counterstain was used to visualize nonstaining nuclei. PCNA-positive nuclei were quantified by 2 blinded observers on 4 histological sections from each brain, counting only those positive nuclei within the wall of the ACA. For each brain, the number of positive nuclei per slide was averaged over all slides for both observers. To stain for PDGF, a primary anti–PDGF-B chain antibody (Chemical International) was used in combination with the Vectastain ABC peroxidase kit, according to the manufacturer’s instructions.

Human Pial Arteries In Vitro

Pial artery samples from 3 patients were divided into 4 segments, each 1 mm in length. The first segment was a “time zero” specimen that served as an initial control. This specimen was formalin fixed within 1 hour of tissue harvest. Remaining pial artery segments were cultured in growth medium, which consisted of Dulbecco’s modified Eagle’s medium with human serum (0.25%), ascorbic acid (50 μg/mL), copper sulfate (3 mg/mL), proline (50 μg/mL), glycine (50 μg/mL), alanine (20 μg/mL), and HEPES (10 mmol/L). All reagents were obtained from Sigma unless noted otherwise. The serum percentage (0.25%) was selected to result in PDGF-AB levels in the growth medium of 580 pg/mL, which is in the range of values measured in CSF of SAH patients (see below).

Pial artery segments were cultured in 3 groups: (1) 7-day-culture control, maintained for 7 days, with no blood contact (group 1); (2) 7-day blood exposure, maintained in contact with freshly clotted human blood (group 2); and (3) 7-day antibody treated, maintained with freshly-clotted human blood that had been mixed with inhibitory antibodies to PDGF-AB and PDGF-BB (group 3).

Pial artery segments were placed on the top of inserts of 6-well tissue-culture plates (Transwell, Corning, Costar Corp). For group 1, the well was filled with 13 mL of growth medium. For groups 2 and 3, 200 μL of human blood, with or without inhibitory antibodies, was allowed to coaggregate on top of the segments for 1 hour before the wells were filled with growth medium. For group 3, anti–PDGF-AB was used at 3.8 μg/mL (10-fold increase over the ND50; AB1486P, Chemicon), and anti–PDGF-BB was used at 6.3 μg/mL (300-fold increase over the ND50; AB1487P, Chemicon). The concentrations of inhibitory antibodies required to block action of targeted growth factors were determined from direct assay of PDGF levels in human serum.

Vessel segments for groups 1 through 3 were maintained in culture for 7 days and then were harvested, formalin fixed, and embedded in paraffin. To assess for cellular proliferation in the vessel walls, sections (5 μm) were stained for PCNA.

Statistical Analysis

Groups were compared with a 1-way ANOVA with a post hoc Scheffe test when indicated by a significant F ratio (JMP statistical software version 3.1, SAS Institute).

Results

Growth Factors in Human CSF

CSF was collected from 20 patients through ventricular or lumbar subarachnoid catheters and from 8 normal volunteers by lumbar puncture. The average patient age was 56 years and ranged from 16 to 86 years. The patients were 13 women
and 7 men; 14 were white and 6 were black. Fourteen patients suffered from SAH, 2 from cerebral contusions, 2 from an unruptured cerebral aneurysm, 1 from obstructive hydrocephalus, and 1 from intracerebral hemorrhage. Vasospasm was diagnosed angiographically in 11 patients, 9 of whom had also clinical symptoms of vasospasm.

PDGF-AB concentrations in CSF from SAH patients were significantly greater than in non-SA patients and normal controls, both during the first week after SAH and for all time points measured (Table 1). For patients with neurological injury who did not have SAH, PDGF-AB and VEGF concentrations were intermediate between SAH patients and controls. bFGF is a growth factor that is not secreted by activated platelets and hence, served as a negative control. When plotted as a function of time after SAH, PDGF-AB and VEGF concentrations began to approach control values 3 weeks after SAH. In contrast, bFGF was not changed after SAH (Figure 1C).

To examine whether PDGF-AB and VEGF concentrations were correlated with subarachnoid blood volume, we compared 7-day peak concentrations in Fisher grade 3 and 4 patients. Neither PDGF-AB nor VEGF were significantly greater in Fisher grade 4 patients compared with grade 3 (Table 2). Peak levels of PDGF-AB and VEGF in the first 7 days were not significantly different between patients with arteriographically documented cerebral vasospasm and those without evidence of vasospasm.

**Cellular Proliferation After SAH**

Immunohistochemical staining of mouse brains revealed clear differences between SAH and control, sham-operated animals (Figure 2A, 2B). Staining for PCNA, a nuclear cyclin involved in DNA replication, revealed marked increases in perivascular positive nuclei after SAH at all time points examined (3 hours, 24 hours, and 72 hours). Although many PCNA-positive smooth muscle cell nuclei were noted within the vascular media of the ACA and other cerebral arteries, the majority of positive nuclei were fibroblasts located in the adventitia and connective tissue surrounding cerebral vessels (arrows, Figure 2B). Counting of PCNA-positive nuclei revealed more smooth muscle cell replication in SAH animals, and this difference was significant when all time points were considered (P < 0.01, Figure 3).

To investigate whether PDGF protein was present at sites of vascular injury, we performed immunoperoxidase staining with a primary antibody directed against the PDGF B-chain. This stain (which would reveal both PDGF-AB, the major constituent of platelet α-granules, as well as PDGF-BB) indicated that there is extensive protein deposition of PDGF in the areas of thrombus formation in this SAH model (Figure 2D). PDGF staining was essentially absent in control specimens but abundantly present in the SAH group (Figure 2C). Thus, smooth muscle and fibroblast proliferation was observed after SAH, and this cellular replication was observed in conjunction with PDGF protein at the sites of thrombus.

**Cellular Proliferation in Human Pial Arteries**

When human pial arteries were examined, PCNA staining revealed little cellular replication in control segments that had been fixed within 1 hour of harvest (Figure 4A). Vessel segments that had been cultured in growth medium alone for 7 days as an additional control also showed low levels of vessel wall cellular proliferation (Figure 4B). In contrast, segments exposed to coagulated human blood displayed highly proliferative areas in the vessel wall, occurring most frequently in the adventitia (circled area, Figure 4C). This proliferation appeared to be blocked by admixing the human blood before coagulation with inhibiting concentrations of anti–PDGF-AB and anti–PDGF-BB antibodies (Figure 4D). These results provide further confirmation that localized thrombus can stimulate vessel wall proliferation in cerebral arteries and that proliferation is blocked by neutralizing antibodies directed against PDGFs.

**Discussion**

**Cellular Proliferation Occurs After SAH**

We have corroborated earlier studies showing that platelet-associated growth factors are elevated in patients with

<table>
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<th>Table 1. Growth Factor Concentrations in CSF</th>
<th>Table 2. Peak Vascular Growth Factor Levels in CSF Samples Within 7 Days After SAH</th>
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<tr>
<td><strong>Group</strong></td>
<td><strong>Peak PDGF-AB, pg/mL</strong></td>
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<tr>
<td>SAH patients</td>
<td></td>
</tr>
<tr>
<td>All time points</td>
<td>14</td>
</tr>
<tr>
<td>First 7 days</td>
<td>11</td>
</tr>
<tr>
<td>Non-SA patients</td>
<td>6</td>
</tr>
<tr>
<td>Normal controls</td>
<td>8</td>
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<tr>
<td><strong>Values are means ± SD.</strong></td>
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<tr>
<td>*P &lt; 0.003 for comparisons of SAH patients, non-SA patients, and controls.</td>
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<th>Fisher Grade</th>
<th>Angiographic Vasospasm</th>
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<tr>
<td>3 (n=3)</td>
<td>4 (n=9)</td>
</tr>
<tr>
<td>PDGF-AB, pg/mL</td>
<td>135 ± 49</td>
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<tr>
<td>VEGF, pg/mL</td>
<td>14 ± 21</td>
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<td><strong>Values are means ± standard deviations.</strong></td>
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Figure 1. ELISA determination of CSF growth factor concentrations in SAH patients. A, PDGF-AB; B, VEGF; C, bFGF. PDGF-AB and VEGF, both derived from platelets, are significantly increased over controls during the first week after SAH and then gradually decay. bFGF concentrations are not similarly elevated.
In a mouse model of SAH in which arterial intraluminal narrowing and resultant neurological deficits have been repeatedly observed at 72 hours after hemorrhage, we found that significant vascular and perivascular cellular proliferation occurs after SAH. This vascular proliferation is associated with PDGF protein in the areas of thrombus. Lastly, we showed that thrombus in proximity to human cerebral vessels in vitro can stimulate vascular cell proliferation. This proliferation is blocked by inhibiting concentrations of antibodies to PDGF. Vascular cell proliferation was observed in both mouse and human arteries, showing that this phenomenon may be generalizable across species.

Taken together, these results support the concept that cellular proliferation occurs in the media and adventitia of cerebral arteries that are near the site of subarachnoid thrombus. These data also support a putative role for platelet-associated growth factors as the stimulus for vascular cell proliferation.

Therapies That Are Effective in Treating Vasospasm

Multiple therapies have proven to be partially effective in alleviating delayed vasospasm, particularly in animal models. Endothelin, a potent vasoconstrictor, is increased in the CSF and plasma of patients with SAH. Animal studies have shown that endothelin inhibition attenuates vasospasm at 1 to 7 days after SAH. However, a recent clinical trial showed only modest efficacy of endothelin inhibition in the prevention of clinical vasospasm. Continuous intrathecal sodium nitroprusside infusion has been shown to be effective in human studies, with complete amelioration of vasospasm in some cases. In related studies, overexpression of nitric oxide synthase and superoxide dismutase was effective in reducing vasospasm. Antisense inhibition of the mitogen-activated protein (MAP) kinase pathway has been shown to attenuate vasospasm in rat models.

Vasospasm Therapies Decrease Cellular Proliferation

Interestingly, these diverse therapies that ameliorate vasospasm all share the feature of inhibiting smooth muscle and fibroblast proliferation. Endothelin is a potent mitogen for vascular smooth muscle, and its inhibition should decrease vascular wall cell proliferation and extracellular matrix synthesis. Nitric oxide, which is liberated by sodium nitroprusside, also blocks smooth muscle replication. Overexpression of superoxide dismutase is thought to increase nitric oxide bioavailability. Calcium channel blockers inhibit cell growth and matrix synthesis of vascular smooth muscle, as does blockade of the MAP kinase pathway. In a novel approach, vasospasm in a rodent model has even been prevented by systemic administration of the chemotherapeutic agent doxorubicin. The platelet-activating factor receptor antagonist, E5880, has shown efficacy in the treatment of patients with vasospasm. Hence, therapies that successfully treat vasospasm all have the physiological effect of decreasing vascular wall cellular proliferation and extracellular matrix synthesis.

Role for Cellular Proliferation in Vascular Stiffening

In vascular diseases like atherosclerosis and pulmonary hypertension, smooth muscle proliferation, wall thickening, and...
extracellular matrix synthesis cause vascular stiffening and decreased arterial compliance.\textsuperscript{31} However, the potential role of vascular cell proliferation in the syndrome of delayed vasospasm has not been extensively considered. Our experimental results show that cellular proliferation occurs in cerebral vessels that are adjacent to thrombus, both in vivo and in vitro. Although these studies focused on platelet-derived factors, there are multiple pathways that stimulate vascular cell proliferation in the vicinity of thrombus. Increased mitotic rate in the vascular wall may increase vessel wall thickness and cause stiffening over the days to weeks after SAH. From a theoretical perspective, it seems possible that thickening of the arterial wall by 3\% of its resting diameter could decrease arterial compliance by approximately one third. Hence, small changes in vessel wall dimensions can have substantial effects on arterial mechanics.

There are additional lines of evidence that point to a structural component of vascular stiffening in vasospasm. The time course of delayed cerebral vasospasm, peaking at 7 to 10 days after SAH, is consistent with the time course of cellular proliferation and tissue-remodeling processes. In contrast, changes in smooth muscle contractile state can occur over minutes to hours. In addition, Kim and coworkers\textsuperscript{32} have shown that the passive compliance of vasospastic canine arteries is decreased compared with control arteries. This decreased compliance is independent of smooth muscle contraction and hence, must be related to structural components of the vessel wall.

Interpretation of Data and Future Work
Growth factor concentrations that were measured from CSF were below concentrations required to stimulate cell replication in vitro.\textsuperscript{33} However, measured CSF concentrations must be assumed to be substantially lower than local concentrations near the site of the thrombus. In addition, CSF samples were often obtained from ventriculostomy drains. Residing in the lateral ventricles, ventriculostomy drains are “upstream” from the subarachnoid clot, and diffusion of growth factors to the ventriculostomy may be unpredictable. Therefore, growth factor concentrations measured from CSF are poorly sensitive indicators of actual concentrations near the site of subarachnoid clot. This fact may explain the inconsistent relation between CSF growth factor concentrations, Fisher grade, and clinical vasospasm.

The present study may provide an added dimension to the etiology of cerebral vasospasm. From these experiments, it is not clear whether cellular proliferation causes wall thickening and decreased arterial compliance in cerebral arteries affected by vasospasm. To make this determination will require future studies that use larger-animal models of SAH. Detailed studies of passive vascular mechanics after SAH and the effects of antimitotic interventions will be necessary to pinpoint the role of cellular proliferation and matrix deposition in the vasospasm syndrome. However, the antiproliferative effects of vasospasm therapies, in addition to their direct vasodilatory effects, should be considered.

Conclusions
Vascular mitogens are increased in the CSF of patients after SAH. Proliferation of cells in the vascular wall occurs after SAH in association with perivascular thrombus. Cellular proliferation and subsequent vessel wall thickening may contribute to the syndrome of delayed cerebral vasospasm after SAH.

Acknowledgments
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References
Cerebral vasospasm following subarachnoid hemorrhage (SAH) occurs in up to 45% of patients with intracranial aneurysmal rupture and is a major cause of mortality and morbidity. Treatment of patients with vasospasm remains a challenge in clinical practice. Targeted therapies have been lacking largely due to limited understanding of the pathogenetic mechanisms of this very complex disease. Over the past decade a variety of techniques. A particular strength of this study is its comprehensive approach that seeks to evaluate human disease and has resulted in important research contributions in this field.

The accompanying article by Borel et al. in this issue of *Stroke* reports the role of platelet-derived growth factor-AB (PDGF-AB), transforming growth factor-β1 (TGF-β1), and vascular endothelial growth factor (VEGF) in vasospasm following SAH. This is a technically sound and thorough study investigating the role of these vascular mitogens with a variety of techniques. A particular strength of this study is its comprehensive approach that seeks to evaluate human disease and animal models including (1) analysis of cerebrospinal fluid from SAH patients, (2) in vitro incubation of human cerebral arteries with blood and PDGF antibodies, and (3) immunohistochemical staining of PDGF over time in brains from a murine model of SAH. The data provide corroborative evidence that cellular proliferation in the media and adventitia of cerebral vessels is an important part of vasospastic pathology and that the proliferation can be effectively blocked by antibodies directed against PDGF. However, this study does not address the critical question: is PDGF simply

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**SAH-Induced Cerebral Vasospasm: Unraveling Molecular Mechanisms of a Complex Disease**

Cerebral vasospasm following subarachnoid hemorrhage (SAH) occurs in up to 45% of patients with intracranial aneurysmal rupture and is a major cause of mortality and morbidity. Treatment of patients with vasospasm remains a challenge in clinical practice. Targeted therapies have been lacking largely due to limited understanding of the pathogenetic mechanisms of this very complex disease. Over the past decade a variety of molecular mechanisms have been explored and a number of mediators implicated in its pathogenesis including (1) endothelium-derived mediators (nitric oxide, oxygen free radicals, endothelin, lipooxygenases, and cyclooxygenases and their metabolites); (2) vascular smooth-muscle–derived mediators (potassium channel inhibition, calcium channel activation, reduction in second messengers [cAMP and cGMP], PKC activation); (3) pro-inflammatory mediators involved in blood-brain barrier disruption (serotonin, histamine, bradykinin), cytokines (IL-1, TNF-α and IL-6), and adhesion molecules; and (4) stress-induced gene activation (heat shock proteins, hemeoxygenase-1).

One of the major difficulties in studying the pathogenesis of this complex disease is the use of appropriate animal models that simulate “delayed” vasospasm in human disease. Over the past 4 decades a search for an appropriate animal model of SAH has been pursued, and utility of each has been tailored toward testing specific hypothesis and desired end points. Each of these models has merits as well as limitations, and cost is an important consideration.

Large-animal models have mostly been used to study angiographic vasospasm with or without neurointerventional techniques (intracisternal injection of autologous blood in rabbit, pig, dog, and monkey) while rodents have been utilized to study pathophysiology of vasospasm using molecular biological techniques (femoral artery model in the rat or intravascular perforation in rat and mouse). The primate model seems to more closely simulate human disease and has resulted in important research contributions in this field.

The accompanying article by Borel et al. in this issue of *Stroke* reports the role of platelet-derived growth factor-AB (PDGF-AB), transforming growth factor-β1 (TGF-β1), and vascular endothelial growth factor (VEGF) in vasospasm following SAH. This is a technically sound and thorough study investigating the role of these vascular mitogens with a variety of techniques. A particular strength of this study is its comprehensive approach that seeks to evaluate human disease and animal models including (1) analysis of cerebrospinal fluid from SAH patients, (2) in vitro incubation of human cerebral arteries with blood and PDGF antibodies, and (3) immunohistochemical staining of PDGF over time in brains from a murine model of SAH. The data provide corroborative evidence that cellular proliferation in the media and adventitia of cerebral vessels is an important part of vasospastic pathology and that the proliferation can be effectively blocked by antibodies directed against PDGF. However, this study does not address the critical question: is PDGF simply
a “marker” of SAH-induced vasospasm? A logical follow-up study in the present SAH model would be treatment with a PDGF antagonist to demonstrate improved functional outcome in an animal model of SAH. For example, Hirashima et al demonstrated reduced incidence of symptomatic, as well as angiographic, vasospasm and improved functional outcome with a PDGF antagonist (E5880) in SAH patients as compared with historic controls.

The article by Borel et al is complementary to an extensive body of literature investigating a host of mediators in a variety of animal models of SAH-induced vasospasm. Targeted pharmacological inhibition of various molecular candidates ameliorates vasospasm in animals yet have not translated into benefit in human clinical trials. Thus the larger questions remain very much akin to neuroprotection for ischemic stroke. Firstly, do animal models in use presently simulate “delayed” SAH-induced vasospasm seen in human disease? Secondly, are the numerous mediators identified thus far merely “markers” rather than actual “causative factors” in this complex disease? Future experimental studies have to critically weigh these 2 important factors in carrying out hypothesis-driven experimental studies of this complex disease.

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References
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