Stimulating Circle of Willis Nerve Fibers Preserves the Diffusion-Perfusion Mismatch in Experimental Stroke

Nils Henninger, MD; Marc Fisher, MD

Background and Purpose—Stimulation of the nerves traversing the ethmoidal foramen (including postsynaptic, parasympathetic projections from the sphenopalatine ganglion [SPG], henceforth referred to as “SPG-stimulation”) has been shown to elevate cerebral blood flow (CBF) and to be neuroprotective after permanent, middle cerebral artery occlusion (pMCAO).

Methods—Employing diffusion (DWI)- and perfusion (PWI) weighted MRI, the effect of SPG-stimulation (started at 60 minutes post-MCAO) on the spatiotemporal evolution of ischemia during and after pMCAO was investigated. In an additional experiment, regional CBF changes were investigated in the nonischemic brain.

Results—In the nonischemic brain, SPG stimulation significantly elevated CBF predominantly within areas supplied by the anterior cerebral artery (by 0.64 mL/g/min relative to baseline). In the ischemic brain, CBF only marginally increased within the penumbra and core (by up to 0.08 and 0.15 mL/g/min relative to prestimulation, respectively). However, the threshold-derived CBF lesion volume did not change significantly. Penumbral apparent diffusion coefficient (ADC)-values improved to almost baseline values and the threshold derived ADC/CBF-mismatch was preserved up to 180 minutes after MCAO. TTC-derived lesion volumes were significantly smaller in stimulated versus nonstimulated animals (120.4±74.1 mm³ versus 239.3±68.5 mm³, respectively).

Conclusion—This study demonstrates that unilateral SPG-stimulation increases CBF bilaterally within the normal brain, acutely preserves the CBF/ADC mismatch largely independent of altering cerebral blood flow, and reduces infarct size in the rat permanent suture model. (Stroke. 2007;38:2779-2786.)

Key Words: diffusion weighted imaging | focal cerebral ischemia | perfusion weighted imaging | pterygopalatine ganglion | suture model
stimulation. Serial determination of quantitative CBF and apparent diffusion coefficient (ADC) maps were performed up to 3 hours after pMCAO and correlated with 24-hour 2,3,5-triphenyltetrazolium chloride (TTC)-derived infarct volumes.

**Materials and Methods**

**Animal Preparation**

All procedures used in this study were performed in accordance with our institutional guidelines and all experiments were performed in a blinded, randomized manner. To alleviate pain, animals received 0.05 mg/kg subcutaneous buprenorphine immediately as well as 6 hours after the end of anesthesia. Male Sprague Dawley rats (n=21, Taconic Farms, New York) weighing 290±15 g were anesthetized with isoflurane (5% for induction, 2% for surgery, 1.5% for maintenance) in room air. PE-50 polyethylene tubing was inserted into the left femoral artery for continuous monitoring of mean arterial blood pressure (MABP) and for obtaining blood samples to measure pH, PaO2, PaCO2, and plasma glucose at baseline, 30, 60, and 180 minutes after pMCAO. Body temperature was maintained at 37.0±0.5°C with a thermostatically controlled water pad. Permanent focal cerebral ischemia was produced by intraluminal suture occlusion of the right middle cerebral artery using 4-0 silicon-coated monofilament sutures as previously described.11 Neurologic evaluation was performed at 24 hours as previously described.12

**Study Design**

The study consisted of 2 different experiments:

**Experiment 1** aimed to investigate the effect of SPG-stimulation on regional CBF of animals not subjected to pMCAO. Rats were randomized 2:1 to one of the following groups: After a baseline scan, SPG-stimulation was performed at 0 and 15 minutes, respectively (SPG, n=6). Control animals were operated and had the stimulating electrode implanted but were not stimulated (control, n=3). All animals were euthanized immediately after the end of the last imaging session.

**Experiment 2** aimed to investigate the effect of SPG-stimulation on regional CBF after induction of pMCAO. Rats were randomized to 1 of the following 2 groups: After a baseline scan, SPG-stimulation commenced at 60 minutes post-MCAO and was repeated every 15 minutes until after 3 hours after MCAO (SPG-MCAO, n=6). Control animals were subjected to pMCAO and had the stimulation electrode attached but were not stimulated (Control-MCAO, n=6). At 24 hours after MCAO animals were euthanized by an overdose of intraperitoneal injection of pentobarbital (200 mg/kg) followed by decapitation. Brains were removed and sectioned coronally into 7 1.5-mm-thick slices corresponding to the MR slices and stained with 2,3,5-triphenyltetrazolium chloride (TTC) for post mortem infarct volume calculation with edema correction.12 Absolute and relative swelling of the right (ischemic) versus left hemisphere was assessed. A corrected infarct volume was calculated by the following formula: corrected infarct volume = left hemisphere volume – right hemisphere volume – infarct volume.

**Electrical Stimulation**

The head of the animal was secured in an MR-compatible stereotactic frame and a fine, bipolar, hook-shaped stimulating electrode was put under the nerve bundle that extended from the sphenopalatine ganglion (SPG) to the ethmoidal foramen as previously described.3 The electrode was then immersed in mineral oil, the wound closed, and the electrode fastened to the stereotactic frame. Such secured animals were placed into the MR-scanner without risk of displacement of the stimulating electrode. The electrode was connected to an electronic stimulator (Neuropath, BrainGate LTD). Four sets of 60-second stimulations separated by 12-second intervals were administered at 10 Hz (pulse width=0.2 ms) with an intensity of 1.9 to 2.2 mA corresponding to a peak level of 60 mV. The stimulation paradigm was optimized during a previously published study, yielding robust neuroprotection without producing nerve injury, changes in animal physiology or disruption of the blood–brain barrier (BBB), that only occurred with higher (>10 mA) stimulation intensities and longer pulse widths (1 ms, data not shown).

**MRI Measurements**

MRI experiments were performed on a 4.7 T/40 cm horizontal magnet equipped with a Biospec Bruker console (Billerica), and a 20 G/cm gradient insert (ID=12 cm, 120-µs rise time). A surface coil (ID=2.3 cm) was used for brain imaging and an actively decoupled neck coil for perfusion labeling.12 Animals were imaged at baseline, 0, 5, 10, 15, 20, 25, 30, 60, 90, 120, 150, and 180 minutes (experiment 1) and at 30, 60, 90, 120, 150, and 180 minutes after MCAO (experiment 2), respectively. The ADC of water and CBF were recorded. Three ADC maps were separately acquired with diffusion-sensitive gradients applied along the x, y, and z direction.12 Single shot, echo-planar images (EPI) were acquired over 2.5 minutes with matrix=64×64, spectral width=200 kHz, TR=2 s (90° flip-angle), TE=37.3 ms, b=8 and 1400 s/mm², Δ=24 ms, δ=4.75 ms, FOV= 2.56×2.56 cm, 7 1.5-mm slices, and 16 averages. CBF measurements were made using the continuous arterial spin-labeling (CASL) technique with single-shot, gradient-echo, EPI acquisition.12 When scheduled at the same time, CBF measurements were made during SPG-stimulation. Note that in MCAO-animals, the 60-minute DWI scan started immediately after the first SPG-stimulation (ie, at 65 minutes post-MCAO). Paired images were acquired alternately—one with arterial spin labeling and the other without. MR parameters were as follows: matrix=64×64, TR=2 s (90° flip-angle), TE=13.5 ms, FOV= 2.56×2.56 cm, 7 1.5-mm slices, and 60 pairs of images.

**Region-of-Interest Analysis**

Only the 5 most anterior slices (ie, brain tissue from approximately Bregma +4 mm to −2 mm) were used for region-of-interest (ROI) analysis because the SPG is the source of parasympathetic innervation to most of the anterior part of the cerebral vasculature.5,7,13 Additionally, we did not observe overt CBF-changes within the parieto-occipital cortex (supplied through the posterior parts of the circle of Willis) during or after SPG-stimulation (data not shown). In experiment 1, hemispheric CBF was calculated to investigate the overall effect of SPG-stimulation on cerebral blood flow. In addition, relative CBF changes were calculated for the individual MR slices (with slice 1 being most anterior). Lastly, three ROIs (each 4×4 pixels) were manually defined in both experiments as follows: (ROI1) Caudate putamen (ischemic core in experiment 2), (ROI2) middle cerebral artery supplied cerebral cortex (ischemic penumbra in experiment 2), and (ROI3) anterior cerebral artery supplied cortex (non-ischemic tissue in experiment 2). Care was taken to ensure that ROIs were placed in the same location within the ipsi- and contralateral hemispheres, across groups, images, and time points. Further, ROIs were drawn conservatively avoiding the brain-skull interface as well as the ventricles.

**Calculation of In Vivo Lesion Size**

Images were acquired using STIMULATE (University of Minnesota) and QuickVol II (http://www.quickvol.com/).14 Quantitative average ADC and CBF maps and their corresponding threshold-derived lesion volumes were calculated as described elsewhere.12,15 Briefly, viability thresholds were used to identify all pixels with abnormal ADC and CBF characteristics on each of the 7 imaged slices at each time point. The corresponding ADC and CBF lesion volumes were then calculated by summing the abnormal area and multiplying by the slice thickness. The viability thresholds used were 0.53×10⁻³ mm²/s for ADC and 0.3 mL/g/min for CBF, as previously validated.15

**Statistical Analysis**

Data are presented as mean±SD unless otherwise stated. Statistical comparisons (Sigma-Stat 3.1, SPSS) were performed using repeated
measures analysis of variance (RM-ANOVA) with post hoc Holm-Sidak or Dunn test for multiple comparisons, 2-way RM-ANOVA, and 2-tailed paired or unpaired Student t test, where appropriate. P < 0.05 was considered significant.

## Results

### Physiological Parameters and Neurological Score

No animals died and basal physiological parameters did not differ significantly among groups or experiments at any time point (Table), and there was no significant difference in weight among animals of both groups (data not shown). No significant changes in MABP were observed during and after SPG-stimulation relative to values obtained at baseline as well as relative to controls (Table; Figure 1B), indicating sufficient depth of anesthesia to prevent nonspecific CBF-changes attributable to stimulation induced arousal. The 24-hour neurological scores (mean ± SD min, max, range)

### Table. Physiologic Parameters

<table>
<thead>
<tr>
<th></th>
<th>pH</th>
<th>P&lt;sub&gt;CO2&lt;/sub&gt; mm Hg</th>
<th>P&lt;sub&gt;O2&lt;/sub&gt; mm Hg</th>
<th>Temperature, °C</th>
<th>Glucose, mg/dL</th>
<th>MABP, mm Hg</th>
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<tbody>
<tr>
<td><strong>Nonstimulated</strong></td>
<td></td>
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<tr>
<td>Baseline</td>
<td>7.44±0.03</td>
<td>38.0±5.0</td>
<td>78±9</td>
<td>36.6±2.2</td>
<td>240±24</td>
<td>85±8</td>
</tr>
<tr>
<td>30 minutes</td>
<td>7.43±0.02</td>
<td>37.2±4.4</td>
<td>71±4</td>
<td>36.7±0.1</td>
<td>195±14</td>
<td>87±4</td>
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<tr>
<td>60 minutes</td>
<td>7.44±0.03</td>
<td>39.8±6.0</td>
<td>71±5</td>
<td>36.8±0.2</td>
<td>169±35</td>
<td>81±9</td>
</tr>
<tr>
<td>120 minutes</td>
<td>7.45±0.02</td>
<td>39.5±5.5</td>
<td>71±7</td>
<td>36.7±0.1</td>
<td>171±17</td>
<td>81±9</td>
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<tr>
<td>180 minutes</td>
<td>7.43±0.02</td>
<td>39.1±7.6</td>
<td>69±3</td>
<td>36.8±0.2</td>
<td>172±21</td>
<td>84±9</td>
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<tr>
<td><strong>SPG-stimulated</strong></td>
<td></td>
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<tr>
<td>Baseline</td>
<td>7.45±0.04</td>
<td>36.9±2.1</td>
<td>69±7</td>
<td>36.6±0.1</td>
<td>242±17</td>
<td>85±9</td>
</tr>
<tr>
<td>30 minutes</td>
<td>7.45±0.02</td>
<td>36.8±2.7</td>
<td>69±7</td>
<td>36.6±0.1</td>
<td>192±25</td>
<td>83±8</td>
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<td>60 minutes</td>
<td>7.42±0.05</td>
<td>40.0±3.5</td>
<td>71±8</td>
<td>36.8±0.1</td>
<td>173±33</td>
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<tr>
<td>120 minutes</td>
<td>7.43±0.04</td>
<td>40.8±4.5</td>
<td>75±2</td>
<td>36.9±0.1</td>
<td>160±14</td>
<td>86±7</td>
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<tr>
<td>180 minutes</td>
<td>7.42±0.03</td>
<td>43.7±3.2</td>
<td>72±4</td>
<td>36.8±0.2</td>
<td>161±9</td>
<td>88±4</td>
</tr>
</tbody>
</table>

Physiological parameters obtained from experiment 2 (pMCAO). There was no significant difference in glucose, pH, P<sub>CO2</sub>, and P<sub>O2</sub> levels within and between pMCAO-groups. Furthermore, continuous measurement of MABP as well as temperature did not reveal any changes within and between pMCAO-groups (for clarity, only data from selected time points are shown).
were improved ($P=0.041$) in the SPG group (2.5 ± 0.8, 2, 4, 2) relative to controls (3.7 ± 0.8, 3, 5, 2).

**Experiment 1: Quantitative Apparent Diffusion Coefficient and Cerebral Blood Flow Values Within ROIs**

One stimulated animal did not show any CBF changes throughout the experiment and was subsequently excluded from the data analysis as damage to the nerve bundle was observed. Stimulation induced a significant increase (by $\approx25\%$, range 14% to 76%) in hemispheric CBF relative to baseline. Though animals received only unilateral stimulation, CBF increased similarly in both hemispheres without significant side-by-side differences (data not shown). However, the observed CBF-increases were only significant during stimulation with almost immediate renormalization to baseline values after stimulation. Calculation of the absolute CBF-changes between baseline and subsequently measured CBF maps suggested greatest SPG-stimulation induced changes within anterior parts of the brain (Figure 1A).

Relative to baseline values, the first stimulation induced significant CBF-elevations only in slice 1 (1.4 ± 0.5 [range 0.9 to 2.2] mL/g/min). With the second stimulation, significant CBF-elevations were observed in slice 1 (1.4 ± 0.4 [range 0.9 to 2.0] mL/g/min), slice 2 (1.4 ± 0.2 [range 1.2 to 1.8] mL/g/min), and slice 3 (1.5 ± 0.3 [range 1.1 to 2.0] mL/g/min), respectively. These findings suggested that there was a cumulative effect on CBF with subsequent stimulations with a greater contribution from the anterior cerebral artery (ACA) than from the middle cerebral artery (MCA) territory. Indeed, calculation of relative CBF-changes (data not shown) clearly showed a more pronounced effect of SPG-stimulation in the anterior (mostly ACA-supplied) versus mid-posterior (mostly MCA-supplied) parts of the brain. Furthermore, in contrast to ROIs 1&2 (predominantly MCA-supplied, increases by $\approx16$ and 31%, respectively), only ROI3 (ACA-cortex, increase by $\approx56\%$ from baseline) showed significantly higher CBF values relative to control animals after the second SPG-stimulation (Figure 2).

**Quantitative Cerebral Blood Flow Values Within ROIs**

Figure 4A and 4C shows region specific CBF values within investigated ROIs of both pMCAO-groups. There was a statistically significant increase in core (ROI1) and mismatch (ROI2) CBF after SPG stimulation; however, these changes were only marginal ($\approx0.1$ mL/g/min) and elevated regional CBF only close to or slightly above the viability threshold. In contrast, SPG-stimulation significantly preserved the DWI/PWI-mismatch during treatment and significantly reduced the final TTC-derived lesion volume (120.4 ± 74.1 mm$^3$) relative to nonstimulated controls (239.3 ± 68.5 mm$^3$). After SPG stimulation, the CBF-derived lesion volumes in stimulated animals were approximately 10% smaller (nonsignificant, $P>0.05$) than in nonstimulated controls.
contrast to nonstroke animals, CBF did not change significantly within the ipsilesional (range 0.84 ± 0.24 to 0.95 ± 0.13 mL/g/min) and contralesional (range 0.89 ± 0.36 to 1.05 ± 0.15 mL/g/min) ACA-cortex (ROI3). Furthermore, CBF did not significantly differ within or between contralesional ROIs over the course of the experiment (data not shown).

**Quantitative Apparent Diffusion Coefficient Values Within ROIs**

Figure 4B and 4D shows region-specific ADC values within investigated ROIs of both pMCAO-groups. There was a significant improvement of mismatch (ROI2) but not core (ROI1) ADC values above the threshold for infarction after SPG-stimulation ($P<0.05$ vs nonstimulated controls). ADC values did not change significantly within the ipsilesional and contralesional ACA-cortex (ROI3). Furthermore, ADC values did not significantly differ within or between contralesional ROIs over the course of the experiment (data not shown).

**Discussion**

Several studies have shown that stimulation of the SPG in anesthetized animals induces vasodilation of cerebral arteries and elevations in CBF as assessed by laser Doppler flow studies,3,9 mass spectrometry,5 or angiography.6,7 Based on these findings it was hypothesized that electrical stimulation of the SPG may improve hemispheric CBF during focal cerebral ischemia and attenuate final infarct size.9 Indeed, Yarnitsky et al9 demonstrated a moderate improvement of cortical CBF during permanent MCAO as well as decreased 27-hour infarct volumes. However, this study did not systematically assess the acute evolution of the ischemic lesion. Using DWI and PWI, we investigated the effect of SPG-stimulation on the temporal evolution of CBF changes in the nonischemic as well as ischemic rat brain. In addition, we assessed the spatiotemporal evolution of ischemia during and after pMCAO as well as ascertained regional changes of quantitative ADC and CBF values within selected ROIs in stimulated versus nonstimulated MCAO rats.

The major results of this study were that: (1) SPG-stimulation significantly elevated CBF predominantly within the nonischemic brain supplied by the anterior cerebral artery; (2) the observed CBF-alterations were brief in nature (peristimulational) and less pronounced compared with previously reported results obtained by laser Doppler flow studies;3 (3) in the ischemic brain, CBF only marginally increased and did not contribute to a significant decrease in threshold derived CBF lesion volume; (4) intriguingly, stimulation improved penumbral ADC-values to almost baseline values preserving the DWI/PWI-mismatch; (5) lastly, final TTC-derived lesion volumes were significantly smaller in SPG-stimulated versus nonstimulated animals.

Electrical stimulation of the nasociliary nerve and SPG in the rat has been shown to transiently increase CBF together with MABP without affecting heart rate, and it was hypothesized that the rapid increase of MABP after stimulation could contribute to CBF elevation.3 Conversely, this as well as other studies did not observe significant changes in MABP during or after SPG stimulation,4,7 possibly because of interstudy differences in stimulation parameters and anesthesia. In this respect it is of note that isoflurane is a potent cerebrovasodilator16 leading to a global increase in CBF. This could have resulted in mitigation of MABP-associated CBF changes and such a “ceiling effect” may also explain the less pronounced stimulation-induced CBF changes in the ipsilateral parietal cortex of this study ($\approx 30\%$) relative to previously reported values ranging from $\approx 40\%$ to $100\%$.3-5 Indeed, whereas baseline CBF was higher in isoflurane versus alpha-chloralose anesthetized animals, CBF-changes after forepaw stimulation were much less than under alpha-chloralose, indicating that isoflurane reduces relative CBF-changes as a result of its vasodilating properties.17

Unexpectedly, we observed similar CBF-changes bilaterally as opposed to the previously reported predominantly

**Figure 3.** Spatiotemporal evolution of threshold-derived ADC and CBF lesion volumes of stimulated and nonstimulated pMCAO rats. *$P<0.05$ for significant CBF/ADC mismatch volume (paired t test); ‡$P<0.05$ for between-group differences of CBF or ADC lesion volumes (Student t test).
isphemispheric CBF-elevations.1–5 Fibers from each sphenopalatine ganglion innervate the contralateral hemisphere in the rat,18 which partly explains the albeit small, yet significant contralateral increases in CBF with SPG-stimulation.3,5 In addition, both anterior cerebral arteries in the rat fuse to form the azygos anterior cerebral artery, therefore, unilateral vasodilation of 1 anterior cerebral artery is expected to cause bilateral CBF-increases in the ACA-supplied brain regions as well as partly in the MCA-territory via anastomoses.

In contrast to marked CBF elevations in the nonischemic brain, we did not observe significant increases in the ACA territory the ischemic brain. Further, only a marginal improvement of ipsilesional CBF within the ischemic core and the penumbra were noted. These results corroborate the findings from a previous study showing greatly attenuated CBF responses during SPG stimulation,9 likely a result of ischemia-induced (sub)maximal vasodilation in the ischemic tissue with an attenuated capacity of the vessels to further dilate in response to SPG stimulation. Abrupt vascular occlusion such as in the model used can exhaust the capacity of the collaterals to sustain CBF to the hypoperfused brain areas,19 and hence these vessels may also “lose” their ability to respond to vasodilating stimuli. Because stimulation-induced CBF elevations were predominantly a result of ACA-vasodilation, it is likely that exhaustion of collaterals (azygos anterior cerebral artery) prevented bilateral CBF-increases in the ischemic brain.

Despite the fact that SPG stimulation induced some improvement of core as well as penumbral CBF, this was insufficient to significantly improve the threshold-defined CBF lesion volume und arguably may have contributed to—but was not the sole cause of—the impressive preservation of the ADC/CBF-mismatch as well as the neuroprotection seen on 24-hour TTC staining. Several potentially interacting phenomena may have contributed to those beneficial effects. First, because regional CBF was measured within an area comprising only a fraction of the threshold defined CBF lesion volume, a local steal phenomenon may have caused CBF declines in other, noninvestigated areas, rendering the overall CBF lesion volume unchanged. However, in light of the observed nonsignificant (∼10%) reduction in CBF lesion volume it seems more likely that the regional improvement in CBF was insufficient to affect the total lesion volume. Second, continued stimulation may

**Figure 4.** Region specific cerebral blood flow (CBF) and apparent diffusion coefficient (ADC) values within investigated ipsilesional ROIs of nonstimulated (A and B, respectively) and SPG-stimulated (C and D, respectively) pMCAO-groups. Dotted lines denote critical viability thresholds. Within each contralateral ROI, CBF and ADC values did not differ significantly between time points and were averaged for visual clarity in the figure (Contra). *P<0.05 vs nonstimulated controls (Student t test); †P<0.05 vs 30 minutes (RM-ANOVA).
induce subtle alterations of the BBB with subsequent vaso-
genic edema, which may have masked cytotoxic ADC de-
clines (“pseudo-normalization” of ADC). However, this pos-
sibility is remote because we did not observe any significant ADC changes within ipsilesional and contralesional ACA-
cortex over time or between hemispheres, and our prelimi-
ary results demonstrated preserved BBB integrity with the same stimulation paradigm. Nevertheless, to ascertain very subtle alterations of BBB-integrity inaccessible by the meth-
odology used, future studies should assess permeability or water content to further address this issue. Third, though unexpected, our results may not be implausible in light of several previous reports indicating that electrical stimulation of parts of the brain could provide central neurogenic neuro-
protection independent of CBF-impovermnt.20,21 Proposed mechanisms include decreased inflammation,22 spreading depression,23 and inhibition of apoptosis.24 For example, it was hypothesized that fastigial nucleus stimulation could render cerebral vessels less responsive to the proinflamma-
tory action of interleukin-1β by overproduction of inhibitor of nuclear factor κB-α (IkB-α), thus reducing the expression of inducible nitric oxide synthase (NOS-2) and intracellular adhesion molecule 1 (ICAM-1), and hence, attenuate inflation of immune cells.22 Additionally, penumbral apoptosis may be attenuated by modification of potassium channels and increased tolerance to depolarizing stimuli23 as well as reduction of caspase-3 activity by reduced mitochondrial cytochrome c release.24 However, it remains uncertain as to whether SPG stimulation provides stimulation through simi-
lar pathways. Nevertheless, given that SPG stimulation in the nonischemic brain causes vasodilation through release of NO,6-7 it is tempting to speculate that this may play a crucial role in the observed neuroprotection effect. NO can freely diffuse to adjacent cells and is a multimodal endogenous mediator that can prevent inflammation, oxidative damage, thrombosis, apoptosis, and platelet aggregation as well as improvement of neovascularization and mobilization of stem and progenitor cells.8 Importantly, administration of NO in models of permanent cerebral ischemia demonstrated neuro-
protective properties leading to a reduction in final infarct size.9 However, future studies should elucidate the precise neuroprotective mechanisms underlying SPG stimulation and examine its effects on complex functional outcome measures. For example, involvement of NO could be confirmed or ruled out by using a nitric oxide synthase inhibitor. Lastly, our findings warrant a lengthier observation period in a less severe ischemia model that may enhance the ability to show whether the tissue protection observed by MRI and brain pathology persists chronically. The importance of better understanding the ameliorative effects associated with SPG stimulation in ischemic stroke is highlighted by the recent development of an orally, minimally invasive implantable device (NeuroPath, BrainsGate LTD) that is currently under investigation for ischemic stroke treatment within an interna-
tional multicenter pilot study.

Summary
In conclusion our results demonstrate that SPG-stimulation (1) bilaterally elevates CBF within the normal brain, (2) acutely preserves the CBF/ADC mismatch largely indepen-
dent of altering CBF, and (3) reduces infarct size in the rat permanent suture model. These observations may be clini-
cally relevant as only a small minority of clinical stroke patients are currently eligible for reperfusion therapy and thrombolysis is often delayed or incomplete.

Acknowledgment
We gratefully acknowledge James Bouley for analyzing TTC-stains.

Source of Funding
This study was partly funded by BrainsGate LTD.

Disclosures
M.F. is a member of the advisory board and N.H. a consultant for BrainsGate LTD.

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*Stroke*. 2007;38:2779-2786; originally published online August 30, 2007;
doi: 10.1161/STROKEAHA.107.485581

*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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