

# Cerebrovascular and Metabolic Effects of SNP-induced Hypotension in Young and Aged Hypertensive Rats

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Cerebral blood flow (CBF) and cerebral oxygen consumption ( $CMR_{O_2}$ ) were measured in young (4 months) and aged (24-26 month) spontaneously hypertensive (SHR) rats and Wistar Kyoto (WKY) controls under control anesthetized conditions (70 per cent  $N_2O$ , 30 per cent  $O_2$ ) and during hypotension induced with intravenous sodium nitroprusside (SNP) infusions. CBF was measured with radioactive microspheres and cerebral arterial-venous blood  $O_2$  measurements were determined from arterial and sagittal sinus blood samples. Arterial blood  $P_{CO_2}$  was maintained at approximately 35 mmHg and body temperature at 37°C. Under control conditions blood pressure was increased in SHR but there was no significant difference in CBF or  $CMR_{O_2}$  between SHR and WKY or young and aged rats. CBF and  $CMR_{O_2}$  were maintained in WKY when mean blood pressure was decreased to 65 mmHg with SNP infusion. CBF was significantly decreased in young and aged hypertensive rats during SNP-induced hypotension.  $CMR_{O_2}$  was also decreased in both young and aged hypertensive animals. These results support previous reports that SNP-induced hypotension will maintain CBF and  $CMR_{O_2}$  in normotensive subjects, but suggest that the direct cerebrovasodilating effects of SNP are moderate and will not reverse the cerebrovascular changes induced by chronic hypertension. (Key words: Age factors. Anesthetic techniques: hypotension, induced. Blood pressure: hypertension; hypotension. Brain: blood flow; metabolism. Pharmacology: nitroprusside.)

IT HAS BEEN REPORTED that a chronic hypertensive state produces cerebrovascular changes which inhibit the ability of hypertensive subjects to maintain cerebral blood flow (CBF) during hypotensive stimuli.<sup>1-3</sup> Several studies have found decreased CBF and cerebral  $O_2$  consumption ( $CMR_{O_2}$ ) and signs of ischemia when moderate hypotension is induced with ganglionic blockade in hypertensive patients. Similar findings are reported in spontaneously hypertensive rats (SHR).<sup>4</sup> In addition, it was noted that the decrement in the ability of hypertensive rats to autoregulate CBF is increased as a function of age.

It is necessary for the anesthesiologist to control the blood pressure of the hypertensive patient during surgery and it is often desirable to induce hypotension to decrease blood loss and produce a dry surgical field. This presents a problem since signs of cerebral ischemia may be apparent at mean pressure levels above 100 mmHg in hypertensives.<sup>1</sup> A possible solution is to use a hypotensive drug which will also maintain CBF and  $CMR_{O_2}$  by inducing direct cerebrovasodilatation. Sodium nitroprusside (SNP) apparently is such a drug, reported to maintain CBF and  $CMR_{O_2}$  at blood pressure levels far below those which produce cerebral ischemia during ganglionic blockade.<sup>5,6</sup> It was the purpose of these experiments to test the ability of SNP infusion to maintain CBF and cerebral metabolism during moderate hypotension in young and aged SHR and normotensive Wistar Kyoto rats (WKY).

## Methods

Male SHR and WKY (Charles River Co.), 4 months and 24-26 months of age were used in these experiments. Rats were anesthetized initially with 1.2 per cent halothane in order to perform a tracheostomy and for the surgical preparation. PE50 tubing catheters containing heparinized saline were inserted into the right femoral artery and vein and a third catheter was inserted into the left ventricle via the right carotid artery. Pressure pulses were monitored during catheter placement, using a Hewlett-Packard pressure transducer and chart recorder, in order to verify the location of the left ventricular catheter. The rat was then placed in a stereotaxic head holder, the bone over the sagittal sinus drilled away, and a 23-gauge needle inserted into the sagittal sinus using the stereotaxic apparatus. After surgery was completed, anesthesia was maintained with 70 per cent  $N_2O$ , 30 per cent  $O_2$  and 1 mg · kg<sup>-1</sup> · h<sup>-1</sup> *d*-tubocurarine. One hour was allowed for the rat to stabilize while arterial  $P_{CO_2}$  was adjusted to approximately 35 mmHg. Rectal temperature was monitored using a temperature controlled Yellow Springs Inc. thermoprobe and body temperature was maintained at 37°C with the aid of overhead heat lamps. Each rat received 2 microsphere injections, one under control anesthetized conditions and the second after a 10-min intravenous (iv) infusion of SNP to maintain mean blood pressure at 65 mmHg.

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TABLE 1. Blood Gases, pH and O<sub>2</sub> Contents in Young and Aged SHR and WKY during Hypotensive Anesthesia

	Arterial P <sub>CO<sub>2</sub></sub> (mmHg)	Arterial P <sub>O<sub>2</sub></sub> (mmHg)	Arterial pH	CaO <sub>2</sub> (ml/dl)	CvO <sub>2</sub> (ml/dl)
4-month WKY (n = 11)					
Control	37 ± 1	122 ± 6	7.41 ± 0.01	17.5 ± 0.7	11.1 ± 1.0
Hypotension	36 ± 2	128 ± 11	7.39 ± 0.01	14.7 ± 0.5	8.8 ± 0.7
4-month SHR (n = 14)					
Control	36 ± 1	136 ± 4	7.40 ± 0.01	17.0 ± 0.4	9.2 ± 0.7
Hypotension	34 ± 1	141 ± 6	7.38 ± 0.01	15.7 ± 0.3	10.0 ± 0.9
24-26-month WKY (n = 11)					
Control	37 ± 2	107 ± 8	7.42 ± 0.02	17.6 ± 0.8	10.9 ± 0.5
Hypotension	35 ± 1	122 ± 7	7.37 ± 0.01*	16.0 ± 0.4	11.3 ± 0.7
24-26-month SHR (n = 10)					
Control	33 ± 1	109 ± 5	7.44 ± 0.01	18.2 ± 0.6	13.2 ± 0.5
Hypotension	31 ± 2	130 ± 9	7.36 ± 0.03*	15.2 ± 1.0	10.0 ± 1.0

Values are means ± SE.

\* *P* < 0.05 compared to control test in each group.

CaO<sub>2</sub> = arterial oxygen content; CvO<sub>2</sub> = cerebral venous O<sub>2</sub> content.

### MICROSPHERES

Microsphere injections were performed according to previously described techniques.<sup>7</sup> Briefly, a stock solution containing 500,000 15- $\mu$ m microspheres/ml was suspended in isotonic saline with 0.01 per cent Tween-80. Two labelled microsphere species were used, cobalt-57 and ruthenium-103 (New England Nuclear). Microsphere samples were vortexed for one minute, thereafter a 0.2-ml sample (100,000 microspheres) was withdrawn, injected into the left ventricle and flushed in with 0.2 ml saline over a 20-s period. Starting immediately before and continuing 45 s after the end of the microsphere injection, blood was withdrawn from the femoral artery at a rate of 0.4 ml/min using a Harvard infusion/withdrawal pump. Approximately 400 microspheres were trapped in the withdrawal blood sample and 1500 microspheres were trapped in brain tissue during each microsphere injection. This number of microspheres trapped in blood and tissue samples will provide accurate flow measures.<sup>8,9</sup> Arterial blood-gas measurements were made at the end of each microsphere injection procedure. Arterial and sagittal sinus blood samples were also taken after each microsphere test for measurement of cerebral arterial-venous O<sub>2</sub> content. Blood withdrawn during each microsphere test was replaced with rat donor blood. At the end of the second microsphere test the rat was killed and the whole brain removed and stored in 10 per cent formalin overnight. The following day the brain was weighed. The activity of each microsphere label in brain and blood samples was analyzed using a Nuclear Chicago 1035 gamma counter and a Nuclear Data 600 multi-channel analyzer. CBF was analyzed according to the methods of Heymann *et al.*<sup>10</sup> using an Interdata computer.

Oxygen content of blood was determined from mea-

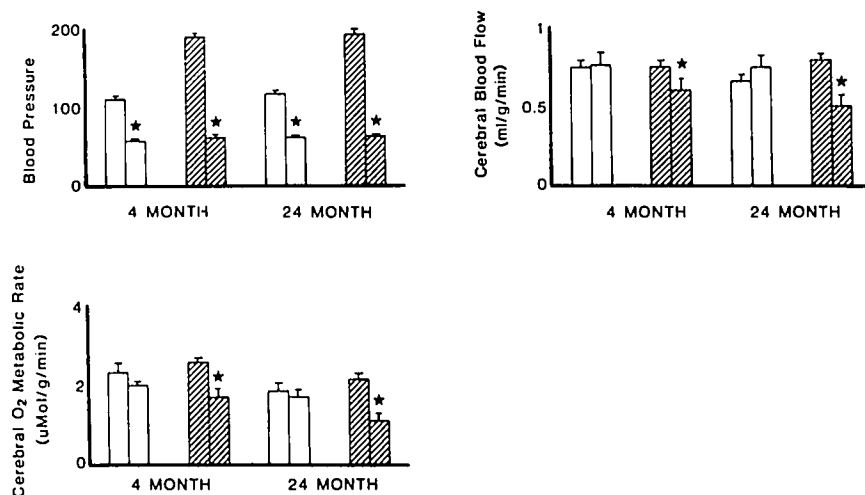
surements of hemoglobin and O<sub>2</sub> saturation using an Instrumentation Laboratory CO-oximeter. CMR<sub>O<sub>2</sub></sub> was calculated as the product of CBF, corrected for brain weight, and cerebral arterial-venous blood O<sub>2</sub> content. All data are reported as means ± SE. Statistical comparisons were performed using paired and unpaired *t* tests and a treatment × groups analysis of variance.

In previous experiments we have shown that three repeated injections of microspheres resulted in reproducible measures of CBF over the three tests.<sup>7</sup> Preliminary tests were also carried out here in order to determine whether reproducible measures could be obtained in nitrous oxide anesthetized rats. In 13 normotensive rats we observed that three repeated tests with radioactive microspheres produced no significant change in blood pressure, CBF or CMR<sub>O<sub>2</sub></sub> measures, indicating the reliability of these test procedures.

### Results

A total of 16 4-month WKY, 28 4-month SHR, 13 24-26 month WKY and 17 24-26 month SHR were surgically prepared for hypotensive drug treatment. All of these rats received the first microsphere test under control anesthetized conditions. Of these rats, five young WKY and two aged normotensive rats, 14 young and seven aged hypertensive rats developed tachyphylaxis during SNP infusion, requiring an SNP infusion rate greater than 1 mg · kg<sup>-1</sup> · min<sup>-1</sup> to maintain a mean blood pressure of 65 mmHg during the 10-min infusion period. Arterial blood-gas measurements in these rats, taken at the end of the 10 min hypotensive period, indicated arterial P<sub>O<sub>2</sub></sub> levels well over 100 mmHg, P<sub>CO<sub>2</sub></sub> values below 30 mmHg, an arterial pH in the range of 7.20 and elevated cerebral venous O<sub>2</sub> content, suggesting cyanide toxicity.<sup>11</sup> Development of tachyphylaxis was signifi-

FIG. 1. Mean blood pressure (mmHg), CBF, and  $CMR_{O_2}$  during control and SNP-induced hypotension in young and aged SHR and WKY. Values are reported as means  $\pm$  SE. The first bar graph in each group indicates control values with 70 per cent  $N_2O$ , 30 per cent  $O_2$  anesthesia. The second test was given during SNP-induced hypotension. Significance values indicate difference between experimental and control conditions within each group but not between groups. Young and aged SHR were significantly hypertensive compared to WKY ( $P < 0.01$ ). Under control conditions there was no significant difference in CBF or  $CMR_{O_2}$  between groups. SNP-induced hypotension produced a decrease in CBF and  $CMR_{O_2}$  in SHR but not WKY. Number of rats in each group are presented in table 1. \* $P < 0.05$ .  $\square$  = WKY.  $\square$  = SHR.



cantly more frequent in SHR than in WKY and in young rats compared to aged ( $P < .05$ ). Rats which developed tachyphylaxis were excluded from the study. The SNP infusion rate at the end of the 10-min hypotensive period in the rats which completed the experiment were as follows: 4-month WKY =  $187 \pm 94 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ , 4-month SHR =  $372 \pm 156 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ , 24–26-month WKY =  $241 \pm 82 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ , 24–26-month SHR =  $393 \pm 155 \mu\text{g} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$ . An analysis of variance indicated that SHR required significantly greater infusion rates of SNP to maintain the hypotensive level of 65 mmHg, compared to WKY ( $P < .05$ ). Arterial blood gases and pH of young and aged SHR and WKY are shown in table 1. Arterial blood gases were not significantly changed during SNP-induced hypotension. Arterial pH was slightly, but significantly decreased in aged SHR and WKY during SNP infusion. CBF and  $CMR_{O_2}$  changes during SNP-induced hypotension are shown in figure 1. Decreasing mean blood pressure to 65 mmHg did not produce a significant change in CBF or  $CMR_{O_2}$  in young or aged WKY. CBF and  $CMR_{O_2}$  decreased significantly in both young and aged hypertensive animals during SNP-induced hypotension.

### Discussion

Results presented here indicate that SNP-induced hypotension will maintain CBF and  $CMR_{O_2}$  in the young and aged normotensive rat. This is consistent with other studies.<sup>5,6</sup> In hypertensive rats, SNP-induced hypotension produced decreases in CBF.  $CMR_{O_2}$  was also significantly reduced in both young and aged SHR during SNP-induced hypotension. These results indicate that under the conditions of these experiments, SNP will maintain CBF and cerebral metabolism in normotensive rats during hypotensive anesthesia, but the cerebrovascular changes produced by chronic hypertension are not

reversed by SNP. Several differences were noted between SHR and WKY and between young and aged rats in these studies. First, it was noted that hypertensive rats had a higher incidence of tachyphylaxis and required a higher infusion rate of SNP to maintain hypotension than in normotensive animals. Young rats also had a higher incidence of tachyphylaxis compared to aged. The reason for these differences is not known. Development of tachyphylaxis during SNP infusion may be the result of baroreflex mechanisms and activation of the renin angiotensin system.<sup>12,13</sup> The differences observed between SHR and WKY and young and aged rats in the frequency of tachyphylaxis probably lies in differences in the ability of each group to induce reflex stimulation of the cardiovascular system. In SHR this may be due to an initially higher blood pressure and a reportedly elevated sympathetic tone.<sup>14</sup> In aged rats it may be due to a decrease in baroreflex activation.<sup>15</sup>

Several studies have investigated the effects of SNP on CBF and  $CMR_{O_2}$  in normotensive animals and patients. Stoyka and Schutz<sup>5</sup> found that SNP but not trimethaphan maintained CBF and  $CMR_{O_2}$  in dogs when cerebral perfusion pressure was lowered to as low as 30 mmHg. Griffiths *et al.*<sup>6</sup> reported that a 42 per cent decrease in blood pressure produced by SNP preserved CBF and  $CMR_{O_2}$  in patients undergoing cerebral aneurysm surgery. Fan *et al.*<sup>16</sup> showed that CBF was increased during SNP infusion in resistant dogs and maintained constant in SNP sensitive dogs during induced hypotension. Maekawa *et al.*<sup>17</sup> found in cats that SNP maintains better brain tissue oxygenation than trimethaphan-induced hypotension as measured with indwelling tissue  $O_2$  electrodes. Michenfelder and Theye<sup>18</sup> reported that hypotension induced with SNP to 40 or 50 mmHg maintained  $CMR_{O_2}$  but not CBF in dogs. Mil-etch *et al.*<sup>19</sup> reported in goats that SNP- and trimethaphan-induced hypotension to mean blood pressure

levels of 42 mmHg both decreased CBF. Although the results reported above appear consistent in indicating maintenance of brain tissue oxygenation and  $CMR_{O_2}$  with SNP during hypotension, the results reporting CBF changes appear more variable. The data suggest that while SNP is more effective than trimethaphan in maintaining CBF and  $CMR_{O_2}$  during moderate hypotension, CBF is not maintained by SNP infusion at very low pressure levels. This indicates that the direct cerebrovasodilator effects of SNP are limited. This may explain the inability of SNP in these studies to reverse the cerebrovascular changes induced by chronic hypertension.

In conclusion, results reported here indicate that hypotension induced with SNP maintains CBF and cerebral metabolism in normotensive WKY but not in SHR. Hypertensive rats are unable to maintain CBF during hypotension induced by ganglionic blockade.<sup>4</sup> This effect is increased as a function of age. Here we have shown that hypotensive anesthesia with SNP infusion does not reverse these changes in hypertensive animals and they remain unable to maintain control levels of CBF and  $CMR_{O_2}$  during hypotensive challenges. These results, together with previous studies, suggest that while SNP may have direct cerebrovasodilating effects, these effects are moderate and are unable to reverse the long-term cerebrovascular changes induced by chronic hypertension.

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