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In Vivo *Effects of Dexmedetomidine on Laser-Doppler Flow and Pial Arteriolar Diameter*

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Background: **The α_2-adrenergic agonist dexmedetomidine alters global cerebral blood flow (CBF). However, few studies have investigated the action of dexmedetomidine on the cerebral microcirculation. This investigation examined the effects of dexmedetomidine on (1) regional CBF in the rat cerebral cortex using laser-Doppler flowmetry and (2) on pial arteriolar diameter.**

Methods: **Halothane-anesthetized rats were fitted with instruments to measure CBF as determined by laser-Doppler flow (CBF_{ldf}) or to measure pial arteriolar diameter by preparing a cranial hollow deepened until a translucent plate of skull remained, thereby maintaining the integrity of the cranial vault. In both groups, 20 μg/kg dexmedetomidine was infused intravenously. Thirty minutes later, the mean arterial pressure was restored to control values with an infusion of phenylephrine (0.5 to 5 μg/kg/min).**

Results: **Administration of dexmedetomidine was associated with decreases in end-tidal and arterial carbon dioxide. The CBF_{ldf} and pial arteriolar diameter were measured during normocapnia (controlled carbon dioxide) and during dexmedetomidine-induced hypocapnia. Intravenous administration of dexmedetomidine significantly decreased systemic arterial pressure concurrent with a decrease in CBF_{ldf} (22% in normocapnic animals, 36% in hypocapnic animals). Restoration of mean arterial pressure increased CBF_{ldf} in normocapnic but not in hypocapnic animals. Similarly, dexmedetomidine significantly reduced pial vessel diameter in both normocapnic (9%) and hypocapnic animals (17%). However, vessel diameters remained decreased in the normocapnic and hypocapnic animals after the mean arterial pressure was restored.**

Conclusions: **These results suggest a modulation of cerebral vascular autoregulation by dexmedetomidine which may be mediated, in part, by alterations in carbon dioxide. Dexmedetomidine may have a direct action on the cerebral vessels to reduce the CBF during normo- or hypocapnia. The differences between CBF_{ldf} and pial arteriole responses to restoration of mean arterial pressure may reflect the difference in measurement techniques because laser-Doppler measurements reflect the net effect of several arterial segments on microvascular perfusion, whereas diameter measurements specifically examined individual pial arterioles, suggesting that dexmedetomidine vasoconstriction in the cerebral vasculature may be differentially and regionally mediated. (Key words: α_2-adrenergic agonists; brain; cerebral blood flow; microcirculation; microscopy.)**

DEXMEDETOMIDINE, a highly selective α_2-adrenergic agonist, possesses potentially useful perianesthetic properties, including sedation, analgesia, anxiolysis, muscle relaxation, hemodynamic stability, and a reduction in the mean alveolar concentration of inhalational anesthetics,[1–6] all mediated through specific presynaptic and postsynaptic α_2-adrenergic receptors within the central nervous system.[1–3,5–7] Other α_2-adrenergic agonists decrease the cerebral blood flow (CBF) in humans and animals.[8–10] Studies suggest that dexmedetomidine may produce significant reductions in CBF without specifically influencing the cerebral metabolic rate for oxygen.[11–13] Dexmedetomidine-induced reductions in CBF have also been observed in the presence of changes in

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the pressures of arterial carbon dioxide and oxygen.[14,15] Most previous studies have examined the action of dexmedetomidine on global CBF. Studies have suggested that certain volatile anesthetics may differentially effect global and regional CBF.[16] Thus it may be critically important to distinguish between specific regional erythrocyte flow and global CBF.

Laser-Doppler flowmetry (CBF_{ldf}) measures CBF to highly localized regions of the brain,[17–19] in a minimally invasive manner, and continuously monitors microvascular perfusion of tissues. Pial arteriolar diameter changes also may be valid indicators of local cerebral perfusion responses.[20,21]

Previous *in vitro* studies from this laboratory[22] showed that dexmedetomidine vasoconstricted canine middle cerebral artery vessels. In addition, recent *in vivo* canine studies showed a significant dexmedetomidine-induced constriction of pial vessels.[23] However, studies have not fully compared the effects of dexmedetomidine on regional CBF, autoregulation, and vessel diameter in the cerebral microcirculation. The purpose of the present investigation was to identify the specific actions of dexmedetomidine on (1) autoregulation of regional CBF in the rat cerebral cortex using laser-Doppler flowmetry and on (2) pial arteriolar diameter by direct microscopic observation through a thinned cranium. These studies were performed during alterations in systemic arterial pressure (SAP) and also during normocapnia and dexmedetomidine-induced hypocapnia. We hypothesized that dexmedetomidine would similarly alter pial vessel diameter and CBF_{ldf} in anesthetized rats.

Materials and Methods

Animal Preparation

All studies were approved by the Animal Use and Care Committee of the Medical College of Wisconsin and were in accordance with National Institutes of Health guidelines (*Guide for the Care and Use of Laboratory Animals*, DHEW [DHHS], NIH publication no. 85-23, revised 1985).

Anesthesia was induced in 33 adult, male Sprague-Dawley rats (weighing 250–450 g) by an intraperitoneal injection of pentobarbital (65 mg/kg; Sigma Chemical Co., St. Louis, MO). After a midline ventral neck dissection, the trachea was isolated and cannulated with polyethylene tubing (PE 240, Intramedic®, Becton Dickinson, Parsippany, NJ). The femoral artery and vein were cannulated with polyethylene catheters (PE 50, Intramedic®) filled with heparinized saline to transduce SAP, maintain fluids, and administer drugs, respectively. Intermittent positive-pressure ventilation with 70% nitrogen and 30% oxygen was instituted (model 707; Harvard Apparatus, South Natick, MA). The inspired gases were mixed using a calibrated 3-tube flowmeter (Linde; Union Carbide, Somerset, NJ). Anesthesia was maintained with 1 mean alveolar concentration halothane. Gallamine (80 mg/kg given intraperitoneally; Sigma Chemical) was used to maintain muscle paralysis. Inspired and expired oxygen, carbon dioxide, and volatile anesthetic concentrations were measured continuously (POET II; TM Criticare Systems, Milwaukee, WI). Arterial blood gas tensions were intermittently determined using a blood gas analyzer (Radiometer ABL3; Copenhagen, Denmark). Body temperature was continuously monitored using a rectal thermistor and maintained at 37 ± 0.2°C with a water-circulated heating pad (model 73 ATA; YSI, Yellow Springs, OH). Cranial/pericranial temperature was not monitored, but the cranial vault remained intact without breaching the transcranial bone or the dura in all experiments. End-tidal carbon dioxide (FET_{CO_2}), mean systemic arterial blood pressure (MAP), systolic SAP, diastolic SAP, cerebral blood flow (CBF_{ldf}), and the concentration of anesthetic vapor were displayed continuously and recorded on an eight-channel polygraph (Astro-Med, West Warwick, RI).

The rats were positioned prone and their heads secured in a stereotaxic apparatus (model 900; David Kopf, Tujunga, CA). After removal of a 2-cm^2 area of scalp and connective tissue, a cranial hollow, 2 mm in diameter, was drilled over the right parietal cerebral cortex, approximately 3 mm posterior to the bregma and 3 mm to the right of the midline. The drilling was done with the aid of a stereomicroscope (Stereo Zoom 5; Cambridge Instruments, Division of Leica, Inc., Deerfield, IL) using a low-speed (0–8,000 rpm) air drill (model Rhino XP; Midwest Dental, Des Plaines, IL), and the surface was simultaneously cooled by continuous air flow through a suction tip. The cranial hollow was deepened until a translucent plate of skull remained, as described previously by Hudetz *et al.*[18]

Measurement of Cerebral Blood Flow by Laser-Doppler Flow (Groups I_A and I_B)

In 18 rats, microvascular perfusion was monitored throughout the study using a laser-Doppler flowmeter

(PF3; Perimed, Stockholm, Sweden) and flowprobe (PF 316 dental probe; Perimed) with a tip diameter of 1 mm. Using a micromanipulator, the probe was lowered and positioned within the burr hole. Mineral oil was applied to improve optical coupling between the probe tip and brain tissue. Care was taken to avoid positioning the probe over large, visible vessels, which were also easily identified by extraordinarily high flow values. Once established, the probe position was not altered for the duration of the experimental protocol.

All animals were allowed to stabilize for 1 h during 1 mean alveolar concentration halothane administration and normocapnia (FET_{CO_2}, 34–37 mmHg). After baseline control measurements of CBF_{LDF}, MAP, SAP, diastolic SAP, and FET_{CO_2}, dexmedetomidine (20 μg/kg; Farmos Group, Turku, Finland), dissolved in 1 ml normal saline, was infused intravenously during a 15-min period. Subsequently, a 15-min period was allowed for equilibration after dexmedetomidine infusion and measurements were repeated. The MAP was restored to control values in the next 20–25 min using a titrated infusion of phenylephrine (0.5–5 $\mu g \cdot kg^{-1} \cdot min^{-1}$; Sigma Chemical Co., St. Louis, MO), and all parameters were again measured at 50% and 100% restoration of MAP. Experiments were successfully completed in 12 rats (groups I_A and I_B). Preliminary experiments suggested that during dexmedetomidine infusion there was a decrease in the $FET_{CO_2}2$ values. Consequently, in eight animals, the ventilation was continuously adjusted to maintain normocapnia (group I_A, n = 8), whereas in four animals, no adjustments in ventilation were made, and thus the FET_{CO_2} was allowed to vary (group I_B, n = 4).

Measurement of Pial Arteriolar Diameter (Groups II_A and II_B)

Fifteen rats were used to examine the effects of dexmedetomidine on the cerebral microvasculature by direct microscopic observation through a thinned cranium. A cranial burr hole was made to a maximum depth but without completely penetrating the skull. The cranial hollow was cleaned and dried by vacuum, filled with a drop of mineral oil, and sealed with a quartz cover slip (Fisher Scientific, Pittsburgh, PA). Pial arteries were visualized using a dark-field metallurgical microscope (model BHMJ; Olympus, Tokyo, Japan) equipped with a 50-W halogen lamp and a vertical epi-illuminator. The incident light was filtered using an infrared filter to minimize heating of the tissue. A 20× ultralong working distance, infinity-corrected lens (ULWD Neo SPlan 20, 0.40; Olympus) was used with a 2.5× TV projection lens. Pial arteries were differentiated from pial veins by lighter color, differences in the branching pattern, and the presence of pulsatile flow. The microscopic image was monitored using a CCD video camera (model VDC-2524; Sanyo, Osaka, Japan), which was continuously displayed and video recorded (model HR-D780V; JVC, Yokohama, Japan).

The experimental protocol was similar to that used in groups I_A and I_B. Briefly, control video recordings of the microvessels were made. Dexmedetomidine was infused and recording of the microvessels was repeated, phenylephrine administered, and recordings again made. Experiments were successfully completed in ten rats (groups II_A and II_B). In a manner similar to the CBF_{ldf} flow experiments of groups I_A and I_B, a decrease in FET_{CO_2} was observed with infusions of dexmedetomidine. Consequently, FET_{CO_2} was controlled within normocapnic limits in six rats (group II_A, n = 6), whereas in four rats (group II_B, n = 4) ventilation was not adjusted.

Using a JVC professional video recorder (model BR-S525U) the recorded video images were examined, selected at specified intervals, and grabbed using an image capturer board (Spectrum NTSC; Redlake Corp., Morgan Hill, CA). Hard copies of selected video images were made using the video graphics printer (UP-860; Sony Corp., Teaneck, NJ). Vessel diameter was measured for the digitized image contrast-enhanced images using Image I software (National Institutes of Health, Bethesda, MD). In each image, seven to eight lines were drawn perpendicular to the length of the vessel and across its opposite edges, and these distances were averaged and calibrated to give an average vessel diameter.

Dexmedetomidine, End-tidal Gas, and Arterial Gas Tensions (Group III)

To confirm that changes in expired end-tidal gas concentrations reflected systemic arterial gas tensions, seven additional adult, male Sprague-Dawley rats (group III) were prepared in a manner similar to that of groups I and II. The animals were anesthetized, ventilated, fitted with monitoring instruments, and positioned in an identical manner. The scalp was not incised and burr holes were not made. The experimental protocol was identical to that of groups I_B and II_B. A 0.25 to 0.5 ml sample of arterial blood was analyzed for *p*H and partial pressures of carbon dioxide and oxygen during control conditions, at the conclusion of dexmedetomidine infusion

Table 1. Effects of Dexmedetomidine during Normocapnia (Group I_A, N = 8)

		Dexmedetomidine Infusion			Post Dexmedetomidine			Phenylephrine Infusion	
	Control	5 min	10 min	15 min	20 min	25 min	30 min	50%	100%
SAP (mmHg)	143 ± 3	142 ± 8	127 ± 10*	119 ± 7*	105 ± 7*	101 ± 7*	102 ± 7*	122 ± 6*	144 ± 4
DAP (mmHg)	82 ± 3	87 ± 8	77 ± 8	75 ± 7	64 ± 7*	59 ± 8*	57 ± 5*	70 ± 4	79 ± 4
MAP (mmHg)	102 ± 3	105 ± 7	93 ± 8	88 ± 7*	75 ± 6*	70 ± 6*	69 ± 6*	87 ± 4*	102 ± 4
FET_{CO_2} (mmHg)	35 ± 0	35 ± 0†	35 ± 0†	35 ± 0†	35 ± 0†	35 ± 0†	35 ± 0†	35 ± 0†	35 ± 0†
RESIS (mmHg/ perfusion units)	0.66 ± 0.02	0.75 ± 0.04	0.72 ± 0.06	0.74 ± 0.08	0.65 ± 0.08	0.62 ± 0.09	0.60 ± 0.06	0.70 ± 0.06	0.68 ± 0.05

SAP = systolic arterial pressure; DAP = diastolic arterial pressure; MAP = mean arterial pressure; FET_{CO_2} = end-tidal carbon dioxide concentrations; RESIS = derived cerebrovascular resistance (MAP/CBF_{ldf}) (see fig. 1); 50%, 100% restoration of MAP by phenylephrine infusion by 50% or 100%.

* Significantly different ($P < 0.05$) from control.

† Significantly different ($P < 0.05$) from group I_B (hypocapnia).

(15 min), 15 min after completion of dexmedetomidine infusion (30 min), and after restoration (100%) of SAP with phenylephrine.

Statistical Analysis

All data are expressed as means ± SEM. All CBF_{ldf} values were normalized to percentages of control. Statistical analysis of data within and between groups during control conditions and during dexmedetomidine infusion, after infusion, and during phenylephrine administration was performed with a repeated measures analysis of variance and Duncan's modification of the *t* test, using the SAS procedure GLM software (SAS Institute, Cary, NC). Changes from control values were considered significant when the probability value was <0.05.

Results

Dexmedetomidine and Laser-Doppler Flow Determination of Cerebral Blood Flow

During control conditions with 1 mean alveolar concentration halothane equilibration and normocapnia, baseline MAP was 102 ± 3 mmHg and CBF_{ldf} was 156 ± 6 perfusion units in group I_A (normocapnia) (table 1, fig. 1). Values were similar in group I_B (hypocapnia) with MAP and CBF_{ldf} of 109 ± 3 mmHg and 163 ± 10 perfusion units, respectively (table 2, fig. 1). Subsequent to initiating the dexmedetomidine infusion, a transient, nonsignificant increase in MAP was observed in most animals. This was followed by a progressive and significant decrease in MAP in animals in groups I_A and I_B. Thirty minutes after initiating dexmedetomidine infusion, MAP decreased by 32% in group I_A rats (102 ± 3 to 69 ± 6 mmHg) and by 36% in group I_B rats (109 ± 3 to 70 ± 2 mmHg). During the dexmedetomidine infusion, a significant reduction in FET_{CO_2} was observed in group I_B rats (30 ± 2 mmHg 30 min after dexmedetomidine infusion compared with 35 ± 0 in control), whereas in group I_A the ventilation was adjusted to keep the FET_{CO_2} within normocapnic limits.

After starting the infusion of dexmedetomidine, sig-

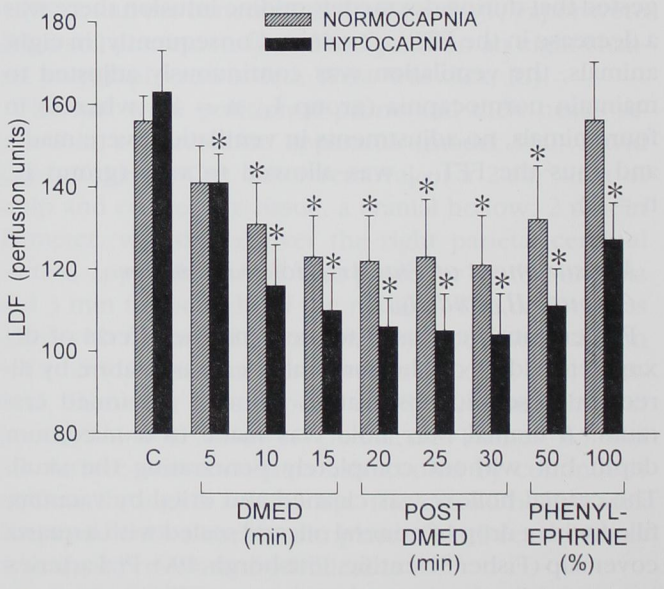

Fig. 1. The effect of dexmedetomidine (20 μg/kg) on laser-Doppler flow during normocapnia and hypocapnia and after restoration of systemic arterial pressure with phenylephrine. % refers to 50% or 100% restoration of control mean arterial pressure. *Significantly different ($P < 0.05$) compared with control values.

Table 2. Effects of Dexmedetomidine during Hypocapnia (Group I_B, N = 4)

		Dexmedetomidine Infusion			Post Dexmedetomidine			Phenylephrine Infusion	
	Control	5 min	10 min	15 min	20 min	25 min	30 min	50%	100%
SAP (mmHg)	144 ± 7	142 ± 7	134 ± 7	124 ± 8*	102 ± 11*	92 ± 2*	91 ± 1*	114 ± 10	142 ± 5
DAP (mmHg)	87 ± 3	98 ± 7	93 ± 7	82 ± 8	64 ± 8*	59 ± 3*	58 ± 3*	73 ± 3	92 ± 4
MAP (mmHg)	109 ± 3	114 ± 7	110 ± 6	99 ± 7	80 ± 10*	71 ± 2*	70 ± 2*	88 ± 6	111 ± 3
FET_{CO_2} (mmHg)	35 ± 0	33 ± 1*†	32 ± 1*†	30 ± 1*†	30 ± 1*†	29 ± 2*†	30 ± 2*†	31 ± 3*†	31 ± 3*†
RESIS (mmHg/ perfusion units)	0.68 ± 0.06	0.80 ± 0.03	0.95 ± 0.07*	0.90 ± 0.04*	0.75 ± 0.08	0.69 ± 0.06	0.68 ± 0.05	0.80 ± 0.07	0.88 ± 0.07*

SAP = systolic arterial pressure; DAP = diastolic arterial pressure; MAP = mean arterial pressure; FET_{CO_2} = end-tidal carbon dioxide concentrations; RESIS = derived cerebrovascular resistance (MAP/CBF_{ldf}) (see fig. 1); 50%, 100% restoration of MAP by phenylephrine infusion by 50% or 100%.

* Significantly different ($P < 0.05$) from control.

† Significantly different ($P < 0.05$) from group I_A (normocapnia).

nificant decreases in CBF_{ldf} values were observed in group I_A and I_B animals. The decreases were not significantly different among the normocapnic groups and hypocapnic groups. Thirty minutes after dexmedetomidine infusion, the decrease in CBF_{ldf} was 22% in group I_A rats (156 ± 6 to 121 ± 12 perfusion units) and 36% in group I_B rats (163 ± 10 to 104 ± 10 perfusion units). When MAP was restored with phenylephrine infusion, the CBF_{ldf} returned to control values in group I_A rats (normocapnic; 156 ± 14 perfusion units compared with 156 ± 6 at control) but remained significantly decreased in group I_B rats (hypocapnic; 127 ± 9 perfusion units compared with 163 ± 10 at control).

Dexmedetomidine and Pial Artery Diameter

The MAP at control was 105 mmHg in normocapnic animals (group II_A) and hypocapnic animals (group II_B; tables 3 and 4, respectively). Baseline vessel diameter was 59 ± 4 μm in group II_A animals, whereas in group II_B animals the vessel diameter was 72 ± 7 μm. Figure 2 shows the vessel diameters (as percentages of baseline control). There were no differences in vessel diameter (raw or normalized data) between groups during control or at any time during the experimental protocol. Similar to the observation in group I animals, the infusion of dexmedetomidine resulted in a significant decrease in MAP. Thirty minutes after starting dexmedetomidine infusion, the percentage decrease in MAP was 40% in group II_A (105 ± 1 to 63 ± 6 mmHg) and 40% (105 ± 3 to 63 ± 2 mmHg) in group II_B. In addition, a significant decrease in FET_{CO_2} was observed during dexmedetomidine infusion in the group II_B rats (30 ± 1 mmHg 30 min after dexmedetomidine infusion compared with 35 ± 0 mmHg at control), whereas the FET_{CO_2} was maintained at control levels in the group II_A animals.

Table 3. Effects of Dexmedetomidine during Normocapnia (Group II_A, N = 6)

		Dexmedetomidine Infusion			Post Dexmedetomidine			Phenylephrine Infusion	
	Control	5 min	10 min	15 min	20 min	25 min	30 min	50%	100%
SAP (mmHg)	152 ± 3	154 ± 3	122 ± 11*	119 ± 11*	101 ± 13*	92 ± 10*	88 ± 11*	107 ± 10	144 ± 4
DAP (mmHg)	88 ± 1	98 ± 6	76 ± 10	78 ± 10	65 ± 10*	55 ± 6*	52 ± 6*	64 ± 6*	89 ± 4
MAP (mmHg)	105 ± 1	115 ± 5	89 ± 8*	87 ± 8*	71 ± 8*	65 ± 6*	63 ± 6*	75 ± 5*	105 ± 2
FET_{CO_2} (mmHg)	36 ± 0	36 ± 0†	35 ± 0†	36 ± 0†	36 ± 0†	36 ± 0†	36 ± 0†	36 ± 0†	36 ± 0†
RESIS (mmHg/μm)	1.87 ± 0.12	2.16 ± 0.16*†	1.72 ± 0.22	1.71 ± 0.24	1.38 ± 0.24*	1.24 ± 0.20*	1.19 ± 0.20*	1.48 ± 0.19	2.04 ± 0.13

SAP = systolic arterial pressure; DAP = diastolic arterial pressure; MAP = mean arterial pressure; FET_{CO_2} = end-tidal carbon dioxide concentrations; RESIS = derived cerebrovascular resistance (MAP/DIA) (see fig. 2); 50%, 100% restoration of MAP by phenylephrine infusion by 50% or 100%.

* Significantly different ($P < 0.05$) from control.

† Significantly different ($P < 0.05$) from group II_B (hypocapnia).

Table 4. Effects of Dexmedetomidine during Hypocapnia (Group II_B, N = 4)

		Dexmedetomidine Infusion			Post Dexmedetomidine			Phenylephrine Infusion	
	Control	5 min	10 min	15 min	20 min	25 min	30 min	50%	100%
SAP (mmHg)	134 ± 5	139 ± 6	117 ± 9*	110 ± 5*	86 ± 3*	83 ± 3*	83 ± 3*	112 ± 8*	141 ± 3
DAP (mmHg)	87 ± 4	87 ± 3	78 ± 7	77 ± 6	56 ± 2*	55 ± 2*	55 ± 2*	74 ± 5	90 ± 5
MAP (mmHg)	105 ± 3	104 ± 2	91 ± 8*	88 ± 6*	66 ± 1*	63 ± 2*	63 ± 2*	88 ± 6*	109 ± 2
FET_{CO_2} (mmHg)	35 ± 0	33 ± 1*†	32 ± 1*†	31 ± 1*†	30 ± 1*†	29 ± 1*†	30 ± 1*†	30 ± 1*†	31 ± 2*†
RESIS (mmHg/μm)	1.53 ± 0.13	1.61 ± 0.20†	1.57 ± 0.29	1.65 ± 0.42	1.28 ± 0.31	1.18 ± 0.34	1.21 ± 0.37	1.68 ± 0.50	2.12 ± 0.55*

SAP = systolic arterial pressure; DAP = diastolic arterial pressure; MAP = mean arterial pressure; FET_{CO_2} = end-tidal carbon dioxide concentrations; RESIS = derived cerebrovascular resistance (MAP/DIA) (see fig. 2); 50%, 100% restoration of MAP by phenylephrine infusion by 50% or 100%.

* Significantly different ($P < 0.05$) from control.

† Significantly different ($P < 0.05$) from group II_A (normocapnia).

With dexmedetomidine infusion, a significant decrease in vessel diameter was observed in both group II_A and II_B rats. Within 15 min of dexmedetomidine infusion, there was a significant decrease in vessel diameter in both the normocapnic group (53 ± 4 compared with 59 ± 4; 10% of control) and the hypocapnic group (61 ± 10 compared with 72 ± 7; 17% of control; fig. 3). Thirty minutes after dexmedetomidine infusion, the vessel diameter remained significantly decreased by 16% of control in group II_B rats (62 ± 12 compared with 72 ± 7 μm at control; $P < 0.05$) but not in group II_A rats (56 ± 5 compared with 59 ± 4 μm at control; P = not significant). When MAP was restored to control values with phenylephrine infusion, the significant decreases in vessel diameter persisted in group II_B (hypocapnic) animals (60 ± 11 compared with 72 ± 7 μm at control) and reappeared in group II_A (normocapnic) animals (53 ± 4 compared with 59 ± 4 μm at control).

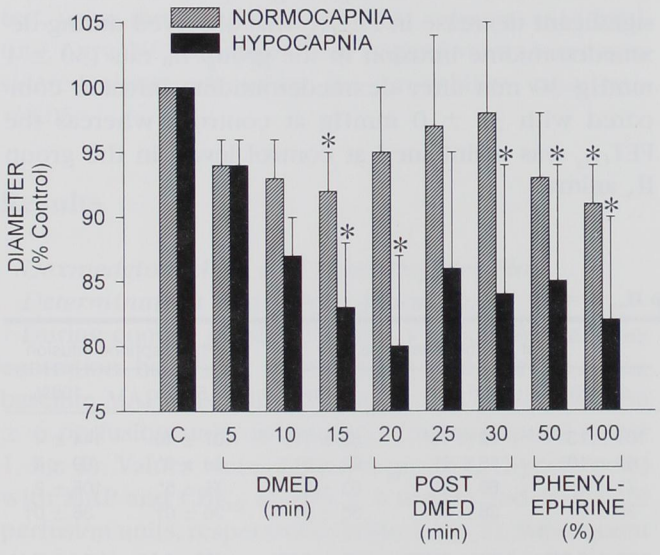

Fig. 2. The effect of dexmedetomidine (20 μg/kg) on pial arteriolar diameter during normocapnia and hypocapnia and after restoration of systemic arterial pressure with phenylephrine. % refers to 50% or 100% restoration of control mean arterial pressure. *Significantly different ($P < 0.05$) compared with control values.

Dexmedetomidine, End-tidal Gas, and Arterial Gas Tensions

Table 5 summarizes the effects of dexmedetomidine on end-tidal carbon dioxide concentrations and concurrent systemic arterial *p*H, carbon dioxide, and oxygen concentrations. As in groups I_B and II_B, dexmedetomidine reduced end-tidal carbon dioxide concentrations (36 ± 0 to 31 ± 1 mmHg; control compared with 30 min after dexmedetomidine; $P < 0.05$) with a concurrent decrease in the arterial partial pressure of carbon dioxide (36 ± 0 to 30 ± 2 mmHg; control compared with 30 min after dexmedetomidine; $P < 0.05$) without significantly altering *p*H or arterial oxygen partial pressures.

Discussion

This study examined the specific effects of dexmedetomidine on regional CBF in the rat cerebral cortex using laser-Doppler flowmetry and the effects on pial arterial diameter. In the present investigation, dexmedetomidine produced significant decreases in CBF_{ldf} and in pial vessel diameter. The slow intravenous infusion of dexmedetomidine that we used in these experiments

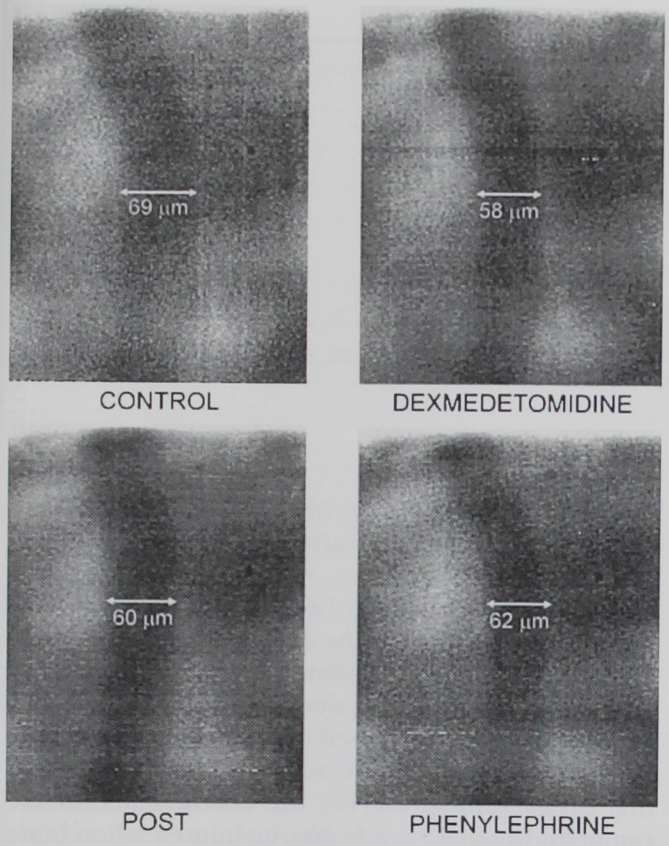

Fig. 3. The effect of dexmedetomidine on pial arteriolar diameter during control, immediately after dexmedetomidine (20 μg/kg) infusion, 30 min after infusion (post), and after restoration of systemic arterial pressure (phenylephrine). The measured diameter of the vessel is labeled in white. Note the decrease in diameter after dexmedetomidine infusion.

transiently increased then progressively and significantly reduced SAP, which is characteristic of α_2-adrenergic agonists. Restoration of SAP by phenylephrine returned CBF_{ldf} to near control values in normocapnic rats but not in hypocapnic rats. In contrast, after restoration of MAP to control values, pial vessel diameter remained significantly decreased in both normocapnic and hypocapnic animals.

The α_2-adrenergic agonists, including clonidine and dexmedetomidine, produce multiple actions within the central nervous system and reduce the dose requirements for anesthetics and narcotics.[1-6,24] Initially a transient increase in SAP occurs due to peripheral vascular α_2-adrenoceptor-mediated vasoconstriction.[25,26] These agents alter systemic hemodynamics through a central sympatholytic and parasympathomimetic action *via* specific α_2-adrenergic receptors at bulbar vasomotor centers, with contributions from supracollicular centers.[7,27,28] This central nervous system action decreases arterial pressure which, in the present investigation, was reversed with phenylephrine, which has no intrinsic effects on CBF.[29]

α_2-Adrenergic receptors are widely distributed within the cerebral vasculature,[30] and specific cerebral vasoconstrictive responses produced by activation of these receptors has been proposed.[31] Clonidine may significantly reduce CBF in both humans and animals[9-12] by stimulating postsynaptic vascular α_2-adrenergic receptors. Previous studies have suggested that dexmedetomidine may also reduce CBF, in both isoflurane- and halothane-anesthetized dogs, without specifically influencing the metabolic rate for oxygen.[11-13] This suggests that dexmedetomidine acts on specific brain regions with dense populations of α_2-receptors to directly vasoconstrict rather than indirectly vasoconstrict secondary to cerebral metabolic depressant effects. However, *in vitro* studies have shown an α_2-adrenergic receptor-mediated vasodilatation of rat middle cerebral arteries that involved[32] nitric oxide and a pertussis toxin-sensitive G protein as mediators. Thus vascular actions, altered sympathetic nerve or regional cerebral activity, and vessel specific actions all may influence the responses seen to α_2-adrenergic agonists.

The commonly used cerebral venous outflow technique to measure CBF requires extensive surgical preparation with possible cerebrovascular compromise.[11,12] Laser-Doppler flow determination of CBF is a sensitive and reliable method to measure regional CBF with a fast response time. To place the CBF_{ldf} probe, most studies use a cranial window technique.[17,20] However, the present studies used a minimally invasive model to monitor perfusion in the rat brain cortex without requiring craniotomy.[18] The CBF_{ldf} probe was applied over a small area of thinned cranium, without invading the tissues to be analyzed. The remaining cranium constitutes an effective barrier for gas diffusion, helps isolate the cerebral tissues from the atmosphere, minimizes or eliminates cerebral hyperemia, maintains local cerebral temperature, and preserves the intracranial pressure.

It has been suggested that CBF is maintained at nearly constant values despite wide variations in

Table 5. Effects of Dexmedetomidine on Arterial Blood Gas Tensions (Group III, N = 7)

	Control	Dexmedetomidine Infusion (15 min)	Post Dexmedetomidine (30 min)	Phenylephrine Infusion (100%)
SAP (mmHg)	126 ± 4	101 ± 8*	84 ± 5*	121 ± 5
DAP (mmHg)	82 ± 3	71 ± 6*	57 ± 3*	75 ± 4
MAP (mmHg)	97 ± 4	81 ± 7*	66 ± 4*	90 ± 4
FET_{CO_2} (mmHg)	36 ± 0	33 ± 1*	31 ± 1*	35 ± 2
*p*H	7.41 ± 0.02	7.45 ± 0.06	7.43 ± 0.02	7.38 ± 0.04
p_{CO_2} (mmHg)	36 ± 1	32 ± 1	30 ± 2*	33 ± 3
p_{O_2} (mmHg)	122 ± 6	120 ± 10	124 ± 11	116 ± 9

SAP = systolic arterial pressure; DAP = diastolic arterial pressure; MAP = mean arterial pressure; HR = heart rate; FET_{CO_2} = end-tidal carbon dioxide concentrations; p_{CO_2} = arterial partial pressure of carbon dioxide; p_{O_2} = arterial partial pressure of oxygen.

* Significantly different ($P < 0.05$) from control.

MAP by adjustments in cerebral vascular resistance.[33] Autoregulation of CBF_{ldf} has been observed between MAPs of 60–140 mmHg.[19] A linear relation between CBF_{ldf} and FET_{CO_2} levels has also been demonstrated.[17,18] Previous studies with volatile anesthetics, including halothane, have demonstrated dose-related increases in CBF_{ldf} in rats.[34] While autoregulation of CBF_{ldf} persisted, it was attenuated at low concentrations of halothane. Therefore, the use of halothane as the baseline anesthetic, and of pentobarbital as the initial anesthetic, may represent a potential limitation of the present investigation and may have affected the results obtained after dexmedetomidine administration, possibly by attenuating vascular responsiveness.

The finding that dexmedetomidine produced significant decreases in CBF_{ldf} despite an initial transient increase in MAP is consistent with previous studies in which a decrease in CBF was observed with dexmedetomidine with either no change or an increase in associated MAP.[11,12] This suggests that dexmedetomidine may have a direct action on the cerebral vessels to reduce the CBF. The alterations in the CBF_{ldf} seen with decreases and increases in MAP, in the autoregulatory range of 60–140 mmHg, suggest that dexmedetomidine might disrupt the autoregulatory mechanisms of the cerebral circulation. As expected, the decrease in CBF_{ldf} was more pronounced in the presence of hypocapnia. The cause of the hypocapnic response produced by dexmedetomidine, in animals in which ventilation was not adjusted, may represent an increase in the depth of anesthesia in these animals already anesthetized with only 1 mean alveolar concentration of halothane.[1–3] Previous studies have shown a species-specific decrease in FET_{CO_2} after administration of dexmedetomidine.[4,25,35]

Various methods have been used in the past to study diameter changes in cerebral microcirculation. Simultaneous measurements of pial diameter and CBF_{ldf} have also been performed.[17,20] Most previous studies describe the closed cranial window preparation for visualizing the cerebral cortex.[21] Although this preparation preserves the cerebral microenvironment, the pial vessels may be injured when bone and dura are lifted. To obviate this, we measured microvessel diameter without breaking the integrity of the cranium. Similar to the measurement of CBF_{ldf}, the pial diameters were also measured through the thinned cranial bone of rats with a fairly minimal surgical procedure. To our knowledge, this is the first time cerebrovascular behavior has been studied by direct microscopy through the thinned intact cranial bone of adult rats.

Potential limitations of the current technique include a possible erroneous simultaneous measurement of cortical, dural, and cranial bone flow rather than purely cortical microvascular flow and assessment of a single flow point rather than flow distribution under the cranial hollow. Extreme care was taken in each experiment to visualize cranial bone blood vessels during the microdissection of the cranial hollow. Vessels were encountered during drilling but not near the bottom when pial vessels were clearly visible. The bone vessels and pial vessels have well-distinguishable structures and, therefore, cannot be mistaken. In addition, the density of dural vessels and their blood flow is low. Furthermore, the

photon migration path distribution in brain tissue is banana shaped between the laser emitting and receiving fibers and, therefore, the measurement should be more sensitive to subsurface vessels. Only a single point measurement of cortical flow was obtained, and because the probe was always positioned at the site with the lowest reading, a higher flow would have been present at the other sites beneath the cranial hollow, although we did not do a systematic search in this investigation. An additional limitation of the present technique is the difficulty in performing topical pharmacologic studies, through a burr hole with an intact cranium in contrast to open preparations.

Studies evaluating the responsiveness of the cerebral vasculature to adrenergic-related agents have produced conflicting results. Norepinephrine has been shown to constrict pial vessels by stimulating α_2-adrenoceptors in pigs,[36] but it also may differentially affect the cerebral arteries, with constriction in rostral regions and dilatation in caudal regions of the bovine brain, resulting in an altered blood flow distribution.[37] Norepinephrine has also been shown to have no effect on cerebral arterioles under baseline conditions.[38]

Clonidine may exert cerebrovascular constrictive effects *in vivo*[39] and *in vitro*,[37] probably mediated by its actions on cerebral arterial α_2-receptors. However, clonidine also may inhibit neurosympathetic cerebroarterial constriction by activating prejunctional α_2-adrenoceptors.[40] A direct cerebral vasoconstrictor action has been proposed for dexmedetomidine due to its ability to decrease CBF.[11-13] Coughlan *et al.*[22] showed dexmedetomidine-induced direct vasoconstriction of canine middle cerebral arteries *in vitro*. Recently, Ishiyama *et al.*[23] showed that topical application of dexmedetomidine constricted pial arteries and veins. Although the vasoconstrictive effects of dexmedetomidine appear to be mediated by activating α_2-adrenoceptors, opposing vasodilatory effects may also occur when adenosine triphosphate-sensitive K^+ channels areactivated.[23] Similar to its effect on the CBF, the cerebrovascularconstrictive effects of dexmedetomidine do not appear to be metabolically mediated. It is also probably not influenced by nitric oxide release because the nitric oxide synthase inhibitor L-NAME did not attenuate dexmedetomidine-induced constriction of cerebral arteries.[22,23]

In the present study, we found that the infusion of dexmedetomidine initially caused cerebral vasoconstriction despite reductions in MAP. This suggests that dexmedetomidine directly vasoconstricts the pial vessels, similar to the observed *in vitro* effects.[22] Presumably, dexmedetomidine might affect the autoregulation of pial arterioles. Although the initial vasoconstriction after dexmedetomidine infusion was followed by vasodilatation as the MAP decreased further, the vessel diameters did not return to control values during either normocapnia or hypocapnia. Restoration of MAP then restored CBF_{ldf} to control levels during controlled normocapnia, but not during hypocapnia, after dexmedetomidine.

These results are consistent with a direct vasoconstrictor effect of dexmedetomidine overriding the cerebrovascular pressure autoregulation. The differences in the response patterns between CBF_{ldf} and pial vessel responses to restoration of MAP may reflect the difference in measurement techniques or the specific vascular segments whose responses are assessed. Changes in CBF are believed to be accompanied by adjustments in the cerebral arteriolar diameter. Haberi *et al.*[17] found a linear relation between CBF_{ldf} responses and arteriolar diameter responses to vasoconstrictor and vasodilator stimuli but suggest that diameter measurements, which are highly localized, may not precisely reflect events in other parts of the arterial tree. On the other hand, Ngai *et al.*[20] found that pial arteriolar diameter changes parallel CBF_{ldf} responses during sciatic nerve stimulation and suggest that arteriolar diameter changes can be considered valid indicators of local cerebral perfusion responses. However, it is still unclear whether surface vessel diameter changes may quantitatively predict flow responses in the cerebral cortex. Although flow within an arteriole has been linked proportionally to its internal diameter,[41] the distribution of cerebrovascular resistance may not be in a steady state in the vascular network. The region studied and the magnitude and direction of response of parenchymal vessels monitored by CBF_{ldf} may be different than that of pial arterioles.

In conclusion, this investigation shows the ability of dexmedetomidine to significantly affect the cerebromicrovascular system. During spontaneous hypocapnia and controlled normocapnia, dexmedetomidine decreased SAP, CBF_{ldf}, and pial arteriolar diame-

ter. Restoration of SAP did not reverse the dexmedetomidine-induced decrease in pial arteriolar diameter, but CBF_{ldf} measurements of CBF did return to control levels during normocapnia but not during hypocapnia. Further investigations are needed to fully delineate the differential actions of dexmedetomidine on various vascular segments of the cerebral circulation.

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