Magnetic resonance imaging studies in animals are usually performed with the animal anesthetized to avoid motion artifacts. The anesthetic agents and the type of ventilation used (spontaneous vs mechanical) can directly affect cerebrovascular and CSF dynamics. The cerebral vasculature is highly responsive to changes in PaCO₂ even in anesthetized animals.¹,² Hypocapnia induces cerebral vasoconstriction, which yields a decrease in CBF and CBV, whereas hypercapnia induces cerebral vasodilation, which yields an increase in CBF and CBV.³⁻⁵ There are conflicting results regarding the effects of hypercapnia, hypocapnia, and hyperoxemia on brain morphometrics determined by use of T1-weighted magnetic resonance imaging in isoflurane-anesthetized dogs

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Objective—To evaluate the effects of various combinations of Paco₂ and Pao₂ values on brain morphometrics.

Animals—6 healthy adult dogs.

Procedures—A modified Latin square design for randomization was used. Dogs were anesthetized with propofol (6 to 8 mg/kg, IV), and anesthesia was maintained with isoflurane (1.7%) and atracurium (0.2 mg/kg, IV, q 30 min). Three targeted values of Paco₂ (20, 40, and 80 mm Hg) and 2 values of Pao₂ (100 and 500 mm Hg) were achieved in each dog, yielding 6 combinations during a single magnetic resonance (MR) imaging session. When the endpoints were reached, dogs were given at least 5 minutes for physiologic variables to stabilize before T1-weighted MR images were obtained. Total brain volume (TBV) and lateral ventricular volume (LVV) were calculated from manually drawn contours of areas of interest by use of a software program, with each dog serving as its own control animal. Three blinded investigators subjectively evaluated the lateral ventricular size (LVS) and the cerebral sulci width (CSW). Brain morphometric values were compared among the target blood gas states.

Results—No significant differences in TBV were found among target states. The LVV was significantly greater during hypocapnia, compared with hypercapnia at the same Pao₂ value. With regard to the subjective evaluations, there were no significant differences among evaluators or among combinations of Pao₂ and Paco₂ values.

Conclusions and Clinical Relevance—The changes observed in LVV during hypocapnia and hypercapnia may serve as a potential confounding factor when neuromorphometric evaluations are performed in anesthetized dogs. (Am J Vet Res 2010;71:1011–1018)
influence of PaCO$_2$ values on CSF formation rate. Some studies have shown no significant changes in CSF production with varying values of PaCO$_2$,$^{6,7}$ whereas others have shown increased CSF production with hypercapnia and a decreased production with hypocapnia.$^{8,9}$ The cerebral vasculature is also responsive to changes in PaCO$_2$, with vasodilatation and therefore an increase in CBF and CBV during hypoxemia (< 50 mm Hg)$^{10}$ and vasoconstriction and a decrease in CBF and CBV during hyperoxemia (> 500 mm Hg).$^{11}$ These changes in CBF and CSF production with different values of PaO$_2$ and PaCO$_2$ could potentially induce parallel changes in global or regional cerebral volumes that may be detected with MR brain morphometric evaluations. However, to the authors’ knowledge, whether this occurs has not been established.

Brain morphometric MR evaluations are being performed in dogs as a research technique for evaluating human brain aging$^{12-14}$ and are increasingly being used clinically to diagnose and investigate the evolution of neurodegenerative diseases in humans.$^{15-17}$ Normal human brain aging is associated with cerebral shrinkage, ventricular enlargement, cortical atrophy, and sulcal widening as observed with MR.$^{16-21}$ In neurodegenerative diseases such as Alzheimer’s disease and frontotemporal dementia, these changes are more pronounced, occur at faster rates, and can vary regionally.$^{15,20}$ In dogs, cerebral morphometric MR evaluations have also shown a decrease in TBV, with the greatest change occurring in the frontal lobes, and an increase in LVV with increasing age.$^{12-14,27,28}$ In the veterinary literature, some evaluations have recently been published that used brain MR morphometrics as a tool to diagnose cerebellar cortical degeneration$^{29}$ and cerebellar atrophy.$^{30}$

Previous brain morphometric MR evaluations performed with dogs anesthetized with isoflurane in O$_2$ used either spontaneous ventilation$^{28}$ or mechanical ventilation$^{12-14,27,28,31}$, however, none of the associated reports included data on arterial CO$_2$ or O$_2$ values. Because variable PaCO$_2$ and PaO$_2$ values could potentially induce changes in TBV and LVV, such changes could represent a source of variability or a confounding factor in studies that use brain morphometrics.

The purpose of the study reported here was to evaluate brain volume changes as a consequence of hypercapnia, hypocapnia, and hyperoxemia in isoflurane-anesthetized dogs by use of brain morphometrics and subjective evaluations of T1-weighted MR images. The hypothesis was that brain volume changes would occur with varying values of PaCO$_2$ and PaO$_2$.

**Materials and Methods**

**Animals**—Six healthy mixed-breed adult castrated male dogs were included in the study. Mean ± SD age of dogs was 2.3 ± 0.5 years, and mean body weight was 28.5 ± 4.7 kg. Health status was determined on the basis of results of a physical examination, CBC, and serum biochemical analysis. Dogs were housed in individual runs in the Central Animal Facility building of the University of Guelph. Food but not water was withheld from all dogs for 12 hours prior to induction of anesthesia. The Animal Care Committee of the University of Guelph approved the procedures and the experimental design.

**Study design**—A split-plot design with modified Latin square design for randomization was used. Six different target combinations of PaCO$_2$ and PaO$_2$ values were sought in each dog during the same MR session: hypocapnia-normoxemia (PaCO$_2$ = 20 mm Hg; PaO$_2$ = 100 mm Hg), hypocapnia-hyperoxemia (PaCO$_2$ = 20 mm Hg; PaO$_2$ = 500 mm Hg), eucapnia-normoxemia (control treatment; PaCO$_2$ = 40 mm Hg; PaO$_2$ = 100 mm Hg), eucapnia-hyperoxemia (PaCO$_2$ = 40 mm Hg; PaO$_2$ = 500 mm Hg), hypercapnia-normoxemia (PaCO$_2$ = 80 mm Hg; PaO$_2$ = 100 mm Hg), and hypercapnia-hyperoxemia (PaCO$_2$ = 80 mm Hg; PaO$_2$ = 500 mm Hg). Each dog was randomly assigned to a different sequence of treatment combinations. T1-weighted images and susceptibility-weighted images were obtained for each of the 6 combinations.

**Achievement of blood gas states**—To achieve the 2 PaO$_2$ endpoints, the isoflurane’s carrier gas administered was switched from 23% O$_2$ for normoxemia (PaO$_2$ = 100 mm Hg) to 100% O$_2$ for hyperoxemia (PaO$_2$ = 500 mm Hg). To achieve the 3 PaCO$_2$ endpoints, the dogs were mechanically ventilated. A Bain circuit was used for hypercapnia (PaCO$_2$ = 80 mm Hg) by use of fresh gas flows between 50 and 130 mL/kg/min to permit rebreathing of CO$_2$, and a circle circuit was used for eucapnia (PaCO$_2$ = 40 mm Hg) and hypocapnia (PaCO$_2$ = 20 mm Hg) with fresh gas flows ranging from 100 to 400 mL/kg/min. The tidal volume and respiratory rate were adjusted to yield hyperventilation for hypercapnia, normoventilation for eucapnia, and hyperventilation for hypocapnia. Once the endpoints were reached as confirmed with an arterial blood gas analysis, at least 5 minutes was allowed for stabilization before the MR scan started.

**Anesthesia and monitoring**—The morning of the study, dogs were taken to the MR unit of the Ontario Veterinary College, where they were anesthetized only once. Anesthesia was induced with propofol$^b$ (6 to 8 mg/kg, IV) titrated to effect via a previously placed 20-gauge cephalic catheter. Once the dogs had lost palpebral reflexes, swallowing reflex, and jaw tone, they were endotracheally intubated and connected to a Bain circuit attached to an anesthetic machine. Isoflurane$^b$ was administered at a constant ETISO of 1.7% (approx 1.3 times the minimum alveolar concentration of isoflurane in dogs)$^{32}$ throughout the study period, which was initially delivered in 100% O$_2$, at a fresh gas flow of 200 mL/kg/min. Mechanical ventilation was initiated immediately after intubation with a volume ventilator$^c$ set up to deliver initially a tidal volume of 15 mL/kg and a respiratory rate of 10 breaths/min. A 20-gauge catheter was placed in a dorsal pedal artery for invasive blood pressure monitoring once the dogs were anesthetized, before moving into the MR scanner.

Dogs were positioned in sternal recumbency for the brain MR scanning. An MR-compatible pulse oximeter$^d$ was placed on the tongue for continuous monitoring of HR and SpO$_2$. A multiparameter monitor$^e$ was used to continuously monitor invasively assessed arterial blood pressures, HR, and inspired and expired concentrations of gases. Invasively assessed SAP, DAP, and MAP were monitored with a pressure transducer.
positioned at the level of the right atrium and zeroed at this level before every pressure recording. A side-stream capnograph with a sampling rate of 200 mL/min was used to measure inspired $O_2$, $ETCO_2$, and $ETIso$. The sampling line was positioned between the proximal end of the endotracheal tube and the anesthetic circuit and was advanced 10 cm distally through the endotracheal tube. The capnograph was calibrated with commercial calibration gases every morning before each MR scan. Rectal temperature was intermittently monitored with a digital thermometer. If the temperature became higher than 38.5°C, then a fan within the MR scanner was turned on, and if the temperature became lower than 37°C, then active warming with warm oat bags and blankets was initiated.

For replacement of fluid losses during anesthesia, a crystalloid solution was administered IV at a rate of 5 mL/kg/h. Dopamine was administered IV as needed at 7 µg/kg/min to maintain a constant MAP between 90 and 100 mm Hg. Atracurium was administered IV at a dose of 0.2 mg/kg before the MR scanning started, and the same dose was repeated every 30 minutes to avoid respiratory efforts or any other movement of the dogs during the study. At the end of the study, a dose of 5 µg of glycopyrrolate/kg followed 5 minutes later by a dose of 0.2 mg of edrophonium/kg was administered IV to reverse any residual effects of atracurium. Dogs were extubated when breathing spontaneously and a swallowing reflex had returned. On the evening of the same day, after complete recovery from anesthesia, they were returned to their runs in the Central Animal Facility.

Data collection—Values of physiologic variables including HR, $SpO_2$, SAP, DAP, MAP, $ETCO_2$, and $ETIso$ were recorded immediately before each MR sequence at each treatment combination and every 10 minutes during the MR scan. Arterial blood samples (collected from the dorsal pedal artery catheter in a 3-mL heparinized syringe) and rectal temperature were obtained immediately before and after each MR scan for each blood gas combination. The arterial blood samples were immediately analyzed for pH, $Paco_2$, $PaO_2$, $HCO_3^-$ concentration, and BE by use of a blood gas analyzer with a correction for rectal temperature. An oximeter was used to measure $CaO_2$. The Hct and TP were also measured in each arterial blood sample.

Image acquisition and processing—A 1.5-Tesla MR scanner with a quadrature transmit-receive knee coil was used. A 3-D, transverse, T1-weighted sequence was obtained (reception time, echo time, and inversion time = 10.6, 4.7, and 175 milliseconds, respectively; flip angle = 30°; slice thickness = 1 mm; 94 contiguous slices; field-of-view = 160 X 140 mm; matrix size = 256 X 160; acquisition time = 10:37 minutes), followed by a high-resolution susceptibility-weighted sequence (a T2*-weighted sequence) that was used in a previously published study. Manual planimetry procedures were performed in T1-weighted images, and a software tool designed to make precise quantitative measurements in volumetric MR brain images was used to assess changes in TBV. All measurements were performed in the transverse plane of the dogs’ brains on contiguous images once by 1 investigator (ER) blinded to the treatments. The brain contour was manually traced from the first slice at the rostral pole of the frontal lobe (not including the olfactory bulb) to the caudal margin of the cerebellum (Figure 1). The volume was calculated by the software multiplying the number of voxels enclosed within all the outlines by their volume and adding half of the volume of the voxels located on the outline itself.

Figure 1—Manually traced brain contours in a series of transverse, 3-D, T1-weighted images used to calculate the TBV in a healthy mixed-breed dog. The brain was measured from the rostral pole of the frontal lobe (upper left image) to the caudal margin of the cerebellum (bottom right image).
For the LVV, the outlined brain images or brain-only images were exported to a software library. An automated segmentation tool was used to visually segment the brain into its 3 components: gray matter, white matter, and CSF. After segmentation, the lateral ventricles were manually outlined on the images with the CSF component only by use of the corresponding transverse T1-weighted images as a reference (Figure 2), and their volume was calculated by summing the number of white voxels contained within the outlines and multiplying this number by the volume of each voxel.

Additionally, 3 blinded evaluators (HD, HJC, and RP) subjectively and independently assessed the LVS and the width of the cerebral sulci during the 6 combinations of $\text{Paco}_2$ and $\text{Pao}_2$ in the T1-weighted MR images of each dog as follows. The image series corresponding to eucapnia-normoxemia (control images) were known by the evaluators for each dog. The other 5 image series corresponding to the various combinations were unknown and compared with the control images in each dog by use of the following numeric scale: 0 = no difference with respect to the control image in LVS or CSW; –1 = mild decrease in LVS or CSW, compared with that in the control image; –2 = moderate decrease in LVS or CSW, compared with that in the control image; –3 = marked decrease in LVS or CSW, compared with that in the control image; +1 = mild increase in LVS or CSW, compared with that in the control image; +2 = moderate increase in LVS or CSW, compared with that in the control image; and +3 = marked increase in LVS or CSW, compared with that in the control image. The LVS and CSW were scored separately.

Statistical analysis—Statistical analysis was performed with statistical software. Data residuals were visually inspected, and the Shapiro-Wilk test was used to determine whether the data were normally distributed. The values of the physiologic variables HR, Sp$\text{o}_2$, ET$\text{CO}_2$, MAP, pH, $\text{Pao}_2$, $\text{Paco}_2$, $\text{CaO}_2$, $\text{HCO}_3^-$ concentration, BE, Hct, TP, and rectal temperature were analyzed with an ANOVA for repeated measures, in which the variable dog was included as a random effect and period and carryover (possible effect from the previous blood gas combination) were included as fixed effects. For each blood gas combination, the mean dose of dopamine used for the duration of the MR scanning was calculated. The mean dopamine dose and TBV were compared among blood gas combinations by use of an ANOVA, with dog as a random effect and period and carryover as fixed effects. The LVV was compared among blood gas combinations by means of an ANCOVA, including in the model the TBV as a covariable, dog as a random effect, and treatment, period, and carryover as fixed effects. The subjective scores for LVS and CSW obtained by each evaluator were compared among blood gas combinations with a Friedman test accounting for block effect of dog. The Wilcoxon Mann-Whitney test was used to determine possible differences between evaluators’ scores for each blood gas combination. Statistical significance was set at a value of $P \leq 0.05$. All results except those for subjective scores are reported as mean ± SD.

Results
The mean ± SD total duration of anesthesia (from induction until extubation) was 385 ± 21 minutes. The MR scan duration during each treatment was 25 ± 1 minutes. Values of physiologic variables during the 6 blood gas combinations of $\text{Paco}_2$ (20 [hypocapnia], 40...
[eucapnia], and 80 [hypercapnia] mm Hg) and PaO₂ (100 [normoxemia] and 500 [hyperoxemia] mm Hg) status were summarized (Table 1). Obtained values for PaCO₂ and PaO₂ were quite close to the targeted values. The ETCO₂ pH, HCO₃⁻ concentrations, and BE changed as expected in response to the changes in PaCO₂. The Hct and rectal temperature were significantly higher during both hypercapnic combinations. The HR, MAP, and TP were not significantly different among blood gas combinations. The SpO₂ values were significantly lower during eucapnia and hypercapnia combined with normoxemia, but CaO₂ was not different, compared with the other blood gas combinations. The dopamine dose used was not significantly different among blood gas combinations.

The TBV was not significantly different among the 6 combinations of PaCO₂ and PaO₂ values (Table 2). There was an overall significant effect of blood gas status on the LVV. After Tukey adjustments were made, significant differences were detected in LVV values between hypocapnia-normoxemia and hypercapnia-normoxemia and between hypocapnia-hyperoxemia and hypercapnia-hyperoxemia, with greater LVV detected during hypercapnia versus hypercapnia.

No significant differences were found in the subjective scores for LVS and CSW among the 6 combinations for any of the evaluators (Table 3). The LVS and CSW subjective scores for each combination were not significantly different among evaluators, except for the combination eucapnia-hyperoxemia for CSW (P = 0.002).

**Discussion**

In the present study, we objectively and subjectively evaluated the effects of hypoxemia, hypocapnia, and hyperoxemia on TBV, LVV, and CSW in isolurane-

**Table 1**—Mean ± SD values of physiologic variables during MR scanning of the brain in 6 isoflurane-anesthetized dogs in which 6 value combinations of PaCO₂ (20 [hypocapnia], 40 [eucapnia], and 80 [hypercapnia] mm Hg) and PaO₂ (100 [normoxemia] and 500 [hyperoxemia] mm Hg) were achieved.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Hypocapnia-normoxemia</th>
<th>Hypocapnia-hyperoxemia</th>
<th>Eucapnia-normoxemia</th>
<th>Eucapnia-hyperoxemia</th>
<th>Hypercapnia-normoxemia</th>
<th>Hypercapnia-hyperoxemia</th>
</tr>
</thead>
<tbody>
<tr>
<td>PaCO₂ (mm Hg)</td>
<td>21.6 ± 1.5</td>
<td>21.5 ± 1.3</td>
<td>41.7 ± 1.8</td>
<td>42.4 ± 0.9</td>
<td>81.5 ± 4.7</td>
<td>80.8 ± 3.7</td>
</tr>
<tr>
<td>ETCO₂ (mm Hg)</td>
<td>20.5 ± 0.7</td>
<td>21.1 ± 2.0</td>
<td>43.4 ± 2.3</td>
<td>43.9 ± 2.7</td>
<td>86.2 ± 3.1</td>
<td>86 ± 3.1</td>
</tr>
<tr>
<td>PaO₂ (mm Hg)</td>
<td>143.7 ± 14.3</td>
<td>152.3 ± 16.4</td>
<td>133.7 ± 15.9</td>
<td>513.8 ± 24.8</td>
<td>104.2 ± 16.2</td>
<td>488.6 ± 15.4</td>
</tr>
<tr>
<td>CaO₂ (ml of O₂/l of blood)</td>
<td>18.7 ± 1.4&lt;sup&gt;a&lt;/sup&gt;</td>
<td>19.1 ± 1.3&lt;sup&gt;a&lt;/sup&gt;</td>
<td>18 ± 2</td>
<td>18.5 ± 1.5&lt;sup&gt;b&lt;/sup&gt;</td>
<td>19.4 ± 1.8&lt;sup&gt;a&lt;/sup&gt;</td>
<td>19.7 ± 1.1&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>SpO₂ (%)</td>
<td>98.4 ± 0.9</td>
<td>98.8 ± 0.9</td>
<td>96.6 ± 0.9</td>
<td>99 ± 0.6&lt;sup&gt;a&lt;/sup&gt;</td>
<td>94.2 ± 0.9</td>
<td>98.9 ± 0.8&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>HR (beats/min)</td>
<td>133.8 ± 15.1&lt;sup&gt;c&lt;/sup&gt;</td>
<td>130.4 ± 18.1&lt;sup&gt;c&lt;/sup&gt;</td>
<td>137.4 ± 12.4&lt;sup&gt;c&lt;/sup&gt;</td>
<td>133.8 ± 12.6&lt;sup&gt;c&lt;/sup&gt;</td>
<td>126.5 ± 9.7&lt;sup&gt;c&lt;/sup&gt;</td>
<td>125.3 ± 10.3&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>MAP (mm Hg)</td>
<td>98.7 ± 12.9&lt;sup&gt;c&lt;/sup&gt;</td>
<td>105.9 ± 17.5&lt;sup&gt;c&lt;/sup&gt;</td>
<td>103.9 ± 15.4&lt;sup&gt;c&lt;/sup&gt;</td>
<td>100.3 ± 15.1&lt;sup&gt;c&lt;/sup&gt;</td>
<td>91 ± 10.7&lt;sup&gt;c&lt;/sup&gt;</td>
<td>92 ± 8.7&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>pH</td>
<td>7.359 ± 0.04&lt;sup&gt;c&lt;/sup&gt;</td>
<td>7.536 ± 0.05&lt;sup&gt;c&lt;/sup&gt;</td>
<td>7.241 ± 0.028&lt;sup&gt;c&lt;/sup&gt;</td>
<td>7.351 ± 0.037&lt;sup&gt;c&lt;/sup&gt;</td>
<td>7.128 ± 0.029&lt;sup&gt;c&lt;/sup&gt;</td>
<td>7.128 ± 0.028&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>HCO₃⁻ (mmol/L)</td>
<td>18.2 ± 1.9</td>
<td>18.0 ± 1.6</td>
<td>21.6 ± 1.5&lt;sup&gt;c&lt;/sup&gt;</td>
<td>21.9 ± 1.7&lt;sup&gt;c&lt;/sup&gt;</td>
<td>24.9 ± 1.6&lt;sup&gt;c&lt;/sup&gt;</td>
<td>24.8 ± 1.6&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>BE (mmol/L)</td>
<td>−1.9 ± 2.3</td>
<td>−1.7 ± 2.8&lt;sup&gt;c&lt;/sup&gt;</td>
<td>−3.1 ± 1.6&lt;sup&gt;c&lt;/sup&gt;</td>
<td>−2.8 ± 1.9&lt;sup&gt;c&lt;/sup&gt;</td>
<td>−6.1 ± 1.7&lt;sup&gt;c&lt;/sup&gt;</td>
<td>−6.0 ± 1.6&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Hct (%)</td>
<td>38.5 ± 3.4&lt;sup&gt;c&lt;/sup&gt;</td>
<td>39.3 ± 2.3&lt;sup&gt;c&lt;/sup&gt;</td>
<td>37.6 ± 3.8&lt;sup&gt;c&lt;/sup&gt;</td>
<td>38.7 ± 3.5&lt;sup&gt;c&lt;/sup&gt;</td>
<td>42.7 ± 3.9&lt;sup&gt;c&lt;/sup&gt;</td>
<td>41.4 ± 2.8&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>TP (g/dL)</td>
<td>5.6 ± 0.3&lt;sup&gt;c,d&lt;/sup&gt;</td>
<td>5.6 ± 0.3&lt;sup&gt;c&lt;/sup&gt;</td>
<td>5.6 ± 0.3&lt;sup&gt;c&lt;/sup&gt;</td>
<td>5.6 ± 0.2&lt;sup&gt;c&lt;/sup&gt;</td>
<td>5.8 ± 0.3&lt;sup&gt;c&lt;/sup&gt;</td>
<td>5.6 ± 0.3&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Temperature (°C)</td>
<td>37.9 ± 0.4&lt;sup&gt;c&lt;/sup&gt;</td>
<td>38.1 ± 0.6&lt;sup&gt;c&lt;/sup&gt;</td>
<td>38.1 ± 0.6&lt;sup&gt;c&lt;/sup&gt;</td>
<td>38.1 ± 0.6&lt;sup&gt;c&lt;/sup&gt;</td>
<td>38.4 ± 0.9&lt;sup&gt;c&lt;/sup&gt;</td>
<td>38.3 ± 0.8&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
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</table>

Dogs were maintained in each blood gas state for a mean ± SD of 25 ± 1 min. Mean values were calculated from the measurements recorded immediately before and after MR scanning (PaCO₂, PaO₂, CaO₂, pH, HCO₃⁻ concentration, BE, Hct, TP, and rectal temperature) or at 10-minute intervals during MR scanning (ETCO₂, SpO₂, HR, and MAP).

<sup>a</sup>Within a row, different superscript letters represent significant (P < 0.05) differences between blood gas combinations.

**Table 2**—Mean ± pooled SD TVB and LVV measured by use of transverse T1-weighted MR images of the brain in 6 isoflurane-anesthetized dogs in which 6 value combinations of PaCO₂ (20 [hypocapnia], 40 [eucapnia], and 80 [hypercapnia] mm Hg) and PaO₂ (100 [normoxemia] and 500 [hyperoxemia] mm Hg) were achieved.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Hypocapnia-normoxemia</th>
<th>Hypocapnia-hyperoxemia</th>
<th>Eucapnia-normoxemia</th>
<th>Eucapnia-hyperoxemia</th>
<th>Hypercapnia-normoxemia</th>
<th>Hypercapnia-hyperoxemia</th>
</tr>
</thead>
<tbody>
<tr>
<td>LVV (cm³)</td>
<td>1.1 ± 0.7&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.13 ± 0.7&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.99 ± 0.7</td>
<td>1.1 ± 0.7</td>
<td>0.97 ± 0.7&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1 ± 0.7&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup>Values for blood gas combinations with the same symbol are significantly (P ≤ 0.05) different.

**Table 3**—Median (range) subjective scores for LVS and CSW evaluated by 3 blinded independent investigators in coronal T1-weighted MR brain images of 6 isoflurane-anesthetized dogs in which 6 value combinations of PaCO₂ (20 [hypocapnia], 40 [eucapnia], and 80 [hypercapnia] mm Hg) and PaO₂ (100 [normoxemia] and 500 [hyperoxemia] mm Hg) were achieved.

<table>
<thead>
<tr>
<th>Evaluator</th>
<th>Variable</th>
<th>Hypocapnia-normoxemia</th>
<th>Hypocapnia-hyperoxemia</th>
<th>Eucapnia-normoxemia</th>
<th>Eucapnia-hyperoxemia</th>
<th>Hypercapnia-normoxemia</th>
<th>Hypercapnia-hyperoxemia</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>LVS</td>
<td>0.1 (−1)</td>
<td>0 (−1)</td>
<td>0</td>
<td>0</td>
<td>0 (−1)</td>
<td>0 (−1)</td>
</tr>
<tr>
<td>2</td>
<td>LVS</td>
<td>−0.1 (−1)</td>
<td>0 (−1)</td>
<td>0</td>
<td>0</td>
<td>0 (−1)</td>
<td>0 (−1)</td>
</tr>
<tr>
<td>3</td>
<td>LVS</td>
<td>−0.1 (−1)</td>
<td>−0.5 (−1)</td>
<td>0</td>
<td>0</td>
<td>−0.5 (−1)</td>
<td>−0.5 (−1)</td>
</tr>
</tbody>
</table>

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anesthetized dogs. Findings indicated that the TBV did not differ among the 6 combinations of PaCO₂ and PaO₂ values; however, the LVV was significantly greater during hypocapnia than during hypercapnia. Subjective evaluation of the MR images did not differentiate among the PaCO₂ and PaO₂ statuses.

Power calculations, based on previously published data, were performed before the study was conducted, indicating that a sample size of 6 dogs would provide a power of approximately 80% to detect statistically significant changes in TBV. Once the study was completed, the calculated power to detect significant differences among blood gas combinations was only 30% for TBV and 90% for LVV. Possible reasons for the low power encountered for TBV are high variability and small differences between combinations. A total of 18 dogs would have been needed to have a power of 84% to detect differences in TBV. However, the power to detect differences in LVV was quite high.

Intrinsic regulation of cerebral circulation involves several factors including chemical factors such as PaCO₂, PaO₂, and pH, which affect cerebral vascular tone and alter CBF; autoregulation, which maintains CBF in response to changes in cerebral perfusion pressure; changes in neuronal activity that may alter cerebral energy metabolism and CBF; rheological factors (primarily the viscosity of the blood); and neurogenic control of CBF by means of sympathetic innervation.

The present study was designed to control for all the other factors that affect cerebrovascular dynamics and therefore could potentially alter brain volumes to isolate the effects of PaCO₂ and PaO₂ status. For that purpose, the MAP was maintained within a physiologic range of 90 to 100 mm Hg with the aid of dopamine. Some MAP values were slightly out of this range, but this phenomenon occurred similarly in the 6 evaluated combinations, and therefore, it is unlikely that this factor significantly contributed to the results. Dopamine could also potentially have affected the CBF. The available data suggest that when dopamine is administered at a low dose (< 20 µg/kg/min) in healthy dogs and sheep, the predominant effect is slight vasodilation with minimal changes in CBF. Nonetheless, dopamine use was not different among the 6 blood gas combinations, making it unlikely that this factor significantly contributed to the results reported here.

Isoflurane can significantly affect cerebral vascular tone, directly causing cerebral vasodilation, whereas cerebral metabolism is depressed in a dose-related manner, producing a classic biphasic effect on CBF. Therefore, in the present study, the ETiso was maintained constant at 1.7% to avoid any possible effect of the concentration of isoflurane on brain volumes and also to guarantee an adequate plane of anesthesia. Nondepolarizing muscle relaxants do not have any direct effect on the cerebral vasculature, but some of them, such as atracurium, can cause histamine release after IV administration of very high doses (4 mg/kg), which may induce systemic and cerebral vasodilation, thus increasing the CBF. No clinical signs of histamine release (eg, hypotension, tachycardia, or urticaria) were detected in any dog in the present study after the administration of atracurium at low doses.

Cerebral blood flow is inversely related to in vitro whole blood viscosity, the major determinant of which is Hct. However, within the reference limits for Hct, the changes in CBF are minimal. It has been suggested that the increase or decrease in CBF when there is a decrease or increase in Hct, respectively, is not attributable to changes in blood viscosity but rather to changes in oxygen delivery to the brain. In our study, the Hct was slightly but significantly higher in both hypercapnic conditions, which could have been attributable to CO₂-induced catecholamine release with subsequent splenic contraction and release of RBCs into the systemic circulation. However, this increase in Hct was not accompanied by a clinically significant increase in the CaO₂; thus, its effect on CBF and therefore on brain volumes was probably unimportant.

Body temperature can also affect the CBF indirectly. Hypothermia decreases whereas hyperthermia increases cerebral metabolism, which will cause a parallel change in CBF. Additionally, a high blood viscosity would be induced because of the reduction of temperature, which may lead to an increase in cerebral vascular resistance and thus reduce the CBF. The rectal temperature of the dogs in the present study was maintained within reference limits, and effects on CBF were highly unlikely.

Hypercapnia is known to result in an increase in CBF and CBV, and the opposite effects are induced by hypocapnia. Hyperoxemia is also known to result in cerebral vasoconstriction, resulting in a reduction of CBF. It is expected that the brain volume will not expand to a great degree with the onset of hypercapnia because it is contained within the skull, which limits its expansion. However, a decrease in brain volume could be expected with the onset of hypocapnia, hyperoxemia, or both. In the present study, we could not demonstrate a decrease in TBV with hypocapnia and hyperoxemia, either separately or in combination. It is possible that no change in volume occurred or that the change in volume was too small to be detected with the methods used in the present study.

The cerebrovascular response to CO₂ appears to be preserved during isoflurane anesthesia, at least at low isoflurane concentrations. In dogs anesthetized with 1.4% isoflurane, the cerebral vasculature constricts with hypocapnia and dilates with hypercapnia, whereas with 2.8% isoflurane, vasoconstriction in response to hypocapnia is retained but vasodilation in response to hypercapnia is absent. At the concentration of isoflurane used in the present study (1.7%), there may have been partial blunting of the cerebrovascular responses to CO₂, but at least the vasoconstrictive response to hypocapnia would have been present. One possible reason for not having detected a change in TBV in our study is that 5 minutes of stabilization at each studied combination before the MR scan started was not enough time for the cerebrovascular responses to take place. However, this is unlikely given that in rats anesthetized with isoflurane, a cerebrovascular response could be detected after only 60 seconds of inhalation of CO₂.

In a study involving halothane-anesthetized dogs, the CSF production rate was of the order of 30 to 40 µL/min (0.03 to 0.04 cm³/min) and did not change dur-
ing 3 hours of hypcapnia. In rats anesthetized with enflurane, hypcapnia decreased the CSF turnover rate by 40%, and hypercapnia increased it by 23%.9 In contrast with this latter study in rats, the lateral ventricles were significantly smaller during hypercapnia than during hypcapnia in the present study, in which 5 minutes of stabilization was allowed after the target arterial blood gases were achieved. Therefore, even if the PaCO₂ truly affects the CSF production rate, there would be a difference in volume of only 30 to 80 µL (0.03 to 0.08 cm³), compared with the volume during eucapnia. This volume difference is too small to be detected with the methods used in our study. The reason for the smaller ventricular size during hypercapnia than during hypcapnia is not completely clear. We hypothesize that the CBF and CBV increased as a consequence of hypcapnia, and that any small increase in the TBV went undetected in our study. This increase in the blood component of the brain might have caused a compression of the lateral ventricles, probably causing an increase in the intracranial pressure caused by the limited compliance of the skull, which was not measured in our study. However, it was likely that the brain was still able to compensate for an increase in CBF and CBV in these healthy dogs because no evidence of brain herniation was evident in any dog and all recovered uneventfully from anesthesia.

Dogs are used as a means to study brain aging because their life span is shorter than the human life span. Brain morphometric studies in which MR images were used have revealed a decrease in TBV, particularly in the frontal lobes, and an increase in LVV with increasing age in dogs.12–14,27,28 The magnitude of these volume changes is small. For example, in 1 study12 the percentage of LVV relative to the TBV increased progressively from 1.1 ± 0.5% at the age of 8 years, 1.2 ± 0.7% at the age of 9 years, 1.6 ± 0.7% at the age of 10 years, and 2.8 ± 0.9% at the age of 11 years.12 All those studies were performed with the dogs anesthetized, usually with an inhalant anesthetic, and none of them reported arterial blood gas values, even though CO₂ and O₂ are important factors influencing cerebral hemodynamics and potentially brain volumes. In the clinical setting, it is unlikely that varying values of PaCO₂ and PaO₂ will affect or confound a diagnosis because the subjective evaluation of the brain MR images in our study did not result in detection of any differences among blood gas concentrations.

To our knowledge, the study reported here represents the first in which the effects of different PaCO₂ and PaO₂ values on brain morphometrics were examined in dogs. Although the TBV did not differ among treatments, the changes in LVV were significant and could serve as a confounding factor when neuromorphometric studies are conducted in anesthetized dogs. Therefore, PaCO₂ and PaO₂ should be controlled and values taken into account when these types of studies are performed. Furthermore, those values should be reported so findings of other studies can be compared.

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