Noninvasive measurements of body composition and body water via quantitative magnetic resonance, deuterium water, and dual-energy x-ray absorptiometry in awake and sedated dogs

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Objective—To compare quantitative magnetic resonance (QMR), dual-energy x-ray absorptiometry (DXA), and deuterium oxide (D₂O) methods for measurement of total body water (TBW), lean body mass (LBM), and fat mass (FM) in healthy dogs and to assess QMR accuracy.

Animals—58 Beagles (9 months to 11.5 years old).

Procedures—QMR scans were performed on awake dogs. A D₂O tracer was administered (100 mg/kg, PO) immediately before dogs were sedated, which was followed by a second QMR or DXA scan. Jugular blood samples were collected before and 120 minutes after D₂O administration.

Results—TBW, LBM, and FM determined via QMR were not significantly different between awake or sedated dogs, and means differed by only 2.0%, 2.2%, and 4.3%, respectively. Compared with results for D₂O dilution, QMR significantly underestimated TBW (10.2%), LBM (13.4%), and FM (15.4%). Similarly, DXA underestimated LBM (7.3%) and FM (8.4%). A significant relationship was detected between FM measured via D₂O dilution and QMR ($r^2 > 0.89$) or DXA ($r^2 > 0.88$). Even though means of TBW and LBM differed significantly between D₂O dilution and QMR or DXA, values were highly related ($r^2 > 0.92$).

Conclusions and Clinical Relevance—QMR was useful for determining body composition in dogs and can be used to safely and rapidly acquire accurate data without the need for sedation or anesthesia. These benefits can facilitate frequent scans, particularly in geriatric, extremely young, or ill pets. Compared with the D₂O dilution method, QMR correction equations provided accurate assessment over a range of body compositions. (Am J Vet Res 2013;74:733–743)

Obesity and healthy weight management in dogs is a growing concern in veterinary medicine and the companion animal nutrition industry, and the ability to accurately, safely, and rapidly obtain in vivo body composition measurements in animals is critical in health and nutrition studies. Most often, objective measures are obtained via DXA or D₂O dilution. However, both of these methods have distinct advantages and disadvantages. The accuracy of these 2 methods have been investigated in studies1,2 with dogs and validated against chemical analysis of carcasses as the criterion-referenced standard. Researchers in one of those studies1 found that the mean estimates of lean tissue mass ($r^2 = 0.998$) and FM ($r^2 = 0.964$) for DXA were strongly associated with chemical analysis of carcasses. For D₂O dilution, TBW mass ($r^2 = 0.99$), ash mass ($r^2 = 0.94$), and

**ABBREVIATIONS**

BCS Body condition score  
CV Coefficient of variation  
D₂O Deuterium oxide  
DXA Dual-energy x-ray absorptiometry  
FM Fat mass  
LBM Lean body mass  
QMR Quantitative magnetic resonance  
TBW Total body water
percentage of body fat ($r^2 = 0.96$) measurements were all highly related, compared with results of chemical analysis of carcasses.3 Noninvasive methods have also been reported for the validation of DXA in dogs against D2O dilution as the reference method.3,4 Consequently, other researchers have used DXA to validate the subjective assessment of body fat via body condition scoring in dogs4,5 and have effectively used DXA to monitor canine body composition during weight loss studies.6,7

Deuterium oxide dilution is routinely used as the reference method to determine body composition in humans and relies on a 2-compartment model approach, in which the body is partitioned into lean mass and FM fractions.9 The D2O dilution method can be used to determine TBW mass, which enables calculation of LBM on the basis of an LBM hydration constant and, thus, calculation of FM by subtraction. In comparison, the criterion-referenced standard for assessing body composition in humans is use of a 4-compartment model,9 which combines body mass, bone mineral mass, TBW mass determined by D2O dilution, and body volume.10 Consequently, the 4-compartment model has been used to validate accuracy of the QMR method in humans,8,11–13 However, a disadvantage is that the 4-compartment model requires the use of several measurement methods, including DXA, D2O dilution, and determination of body volume.

In 2003, the first study14 of the use of QMR to measure TBW and body composition of lean mass and FM was reported. Since then, QMR has been validated for use in many species, including cats,15 mice,16,17 rats,18–20 pigs,20,21 humans,22–24 and chickens.21 These studies for validation of QMR against carcass analysis in mice, rats, chickens, and pigs against D2O dilution in cats have all revealed that values for TBW, LBM, and FM are highly related ($r^2 = 0.80$ to 0.98) with results for the reference method. Overall, results of these studies indicate that the QMR method can be used to obtain accurate data that can be acquired quickly and easily without the need for sedation or anesthesia of subjects. These validation studies have also revealed that the QMR method has high precision (CV < 2%) for measurement of FM, LBM, and TBW. Combined, these positive attributes contribute to a considerable advantage in the ability to frequently assess body composition changes during nutrition studies and in veterinary medical patients. It is also possible to eliminate health concerns and decreased food intake associated with anesthesia, particularly in extremely young or old animals in which the health risks are greater.

Similar to results for other methods, QMR can underestimate or overestimate FM, TBW, and LBM, compared with results for reference methods. The underestimated or overestimated values can differ substantially by species, and in most cases, FM determined via QMR has the most variation. Although a small component of these species differences may be related to the use of different instruments and the small inherent variation for the QMR method, it is probable that the bulk of the species differences are attributable to inherent factors that have not yet been elaborated. Consequently, validation within a target species is required, and prediction equations for each measurement to correct for method bias appear to be necessary. Therefore, the objectives of the study reported here were to compare measurements of LBM, FM, and TBW for QMR and DXA with data generated via D2O dilution as the reference method in dogs with a range of ages and BCSs and to compare accuracy of the QMR method in awake and sedated dogs.

Materials and Methods

Animals—Fifty-eight Beagles were evaluated. Dogs were initially selected on the basis of age (9- to 12-month-old puppies [n = 16] and 1.5- to 11.5-year-old adult dogs [42]) and BCS for use in the study. The group of puppies was balanced on the basis of sex, whereas the group of adult dogs was not (26 males and 16 females). The study was conducted in accordance with approved animal care and use committee protocols.

Housing and assessment of BCS—The BCS was assigned on the basis of a 9-point scale, with a score of 4 or 5 representing an ideal body condition (characterized by a well-proportioned, observable waist caudal to the ribs, ribs palpable with a slight fat cover, and minimal abdominal fat pad).4,5 A BCS < 4 represented an assessment of diminished body fat tissue (lean category), and a BCS > 6 represented overweight dogs.

Before the study, dogs were housed in groups in pens (1.5 × 4.5 m; 2 to 4 dogs/pen). Dogs were grouped on the basis of age, compatibility, and sex. Dogs were housed indoors with natural lighting and exposure to natural light cycles. All dogs were housed at the same kennel location and could see other dogs in adjacent pens. All dogs had direct interaction and socialization with caretakers on a daily basis and had continuous access to multiple toys.

Dogs were fed once daily and had ad libitum access to water. Food was removed the night before the study in preparation for impending sedation of the dogs. This resulted in a food-withholding period of approximately 14 hours.

QMR—On the day of the study, a physical examination was performed on each dog to confirm good health status, and body weight was recorded to the nearest 0.01 kg. An initial QMR scan was performed on each dog, and a jugular blood sample was then collected and used for determination of background concentrations of deuterium. A dose of D2O (100 mg/kg; D2O as 99.8% deuterium) was administered orally as a one-tenth dilution in sterile water via a sterile syringe. Care was used (slow and intermittent administration of the diluted D2O) to ensure that all of the solution was swallowed by each dog. The volume administered was designed to provide a dose of 1 mL/kg; syringes were weighed before and after administration of D2O to determine the exact dose administered. Jugular blood samples were collected 120 minutes after D2O administration and evaluated to ensure equilibration. Serum was separated from blood samples and stored at –80°C until analyzed for deuterium content.

Immediately following collection of the blood sample at 120 minutes after D2O administration, dogs were sedated by IV administration of a combination of dexmedetomidine (0.03 mg/kg) and morphine sulfate (0.3 mg/kg) and were monitored to achieve anesthesia. Once conscious, dogs were transported to the QMR scanner located in the veterinary hospital. Care was taken to ensure that all dogs were positioned appropriately to avoid misregistration of the QMR image.

QMR scanning was performed 30 minutes after D2O administration. Dogs were sedated by IV administration of a combination of dexmedetomidine (0.03 mg/kg) and morphine sulfate (0.3 mg/kg) and were monitored to achieve anesthesia. Once conscious, dogs were transported to the QMR scanner located in the veterinary hospital. Care was taken to ensure that all dogs were positioned appropriately to avoid misregistration of the QMR image.

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mg/kg). A second QMR scan was performed within 15 minutes after dogs were sedated. Immediately after the second QMR scan, the dexmedetomidine was reversed by administration of atipamezole (0.115 mg/kg, IM).

For both scans (awake and sedated), dogs were placed in a polymethylmethacrylate crate that allowed only limited movement. The crate was placed within the magnet bore of the QMR unit, which was designed for scanning of animals weighing < 50 kg. Dogs were positioned at the magnet isocenter such that their long axis was perpendicular to the long axis of the magnet bore. Data were collected as a standard 3-minute fat and water acquisition in accordance with the manufacturer's protocol.

D2O dilution method—The D2O dilution method involves enrichment of body water with the stable (natural and nonradioactive) hydrogen isotope deuterium, followed by determination of the deuterium concentration in a physiologic fluid such as serum. The stable hydrogen isotope contents of the serum samples were measured via gas-isotope ratio mass spectrometry in accordance with previously validated procedures.\textsuperscript{23,24}\r
The precision (ie, SD) for the deuterium assay was 0.10% for samples with natural concentrations of deuterium and 0.18% for samples with enriched concentrations of deuterium.\textsuperscript{24}

The isotopic results were normalized against 2 international water standards: Vienna standard mean ocean water and standard light Antarctic precipitation.\textsuperscript{25} The isotopic dilution space for deuterium was calculated by use of the following equation:

\[
N_H (\text{mol}) = \frac{(d \times \text{A} \times \text{E}_D)}{\alpha \times \text{E}_D \times 18.02}
\]

where \(N_H\) is the isotopic dilution space for deuterium, \(d\) is the dose of D2O in grams, \(A\) is the amount of laboratory water (in grams) used in the dose dilution, \(E_\alpha\) is the increase in deuterium concentration in the laboratory water after the addition of the isotopic water, \(\alpha\) is the amount of D2O (in grams) added to the laboratory water in the dose dilution, and \(E_D\) is the increase in deuterium concentration in the serum samples at isotopic equilibrium at 120 minutes after D2O administration.

The use of a dose dilution in the calculation of isotopic dilution space was recommended by the International Dietary Energy Consultancy Groups to assure accuracy of the isotopic dilution calculations.\textsuperscript{26} The isotopic dilution space for deuterium was calculated by use of the following equation:

\[
\text{TBW} (\text{mol}) = \frac{N_H}{1.03}
\]

where 1.03 is the correction factor to account for the incorporation of deuterium into organic molecules during biosynthesis. Lean body mass is assumed to contain 73.2% moisture; therefore, by use of the hydration constant (0.732) for LBM, TBW was converted to LBM via the following equation: LBM = TBW/0.732. Consequently, FM was calculated as body weight minus LBM.

DXA—Because of unanticipated technical issues with the DXA unit, scans could not be performed on the same day as for the QMR and D\textsubscript{2}O dilution methods. Consequently, DXA was performed on the dogs within 7 days after QMR and D\textsubscript{2}O data collection. Dogs were sedated as described previously. Sedated dogs were placed in sternal recumbency in a DXA unit.\textsuperscript{4} Data acquisition was conducted by use of the manufacturer’s pediatric small algorithm. The associated software was subsequently used to determine the proportions of LBM, FM, and bone mineral. Sedation was reversed immediately after data collection.

Statistical analysis—All statistical analyses were conducted with statistical software.\textsuperscript{27,28} To characterize body composition measurements obtained via the reference method (D\textsubscript{2}O dilution) versus animal factors, an ANOVA was performed to assess the main effects of age, body weight, and BCS on the dependent variables of TBW, LBM, and FM determined via D\textsubscript{2}O dilution. Regression coefficients and prediction equations were generated via linear regression analysis. Paired \(t\) tests were used to assess differences in mean values for body composition components (LBM, FM, and TBW), as measured via the various techniques (QMR or DXA) versus the reference method (D\textsubscript{2}O dilution). Significance was determined at a value of \(\alpha = 0.05\).

To evaluate the predictive value of the correction equations generated to estimate body composition, a cross-validation (jackknife) analysis was performed as described elsewhere.\textsuperscript{15,28} Briefly, data for each dog were removed from the data set and a correction equation was generated for the remaining 57 dogs. The QMR or DXA values for each dog were entered into the equations to generate predicted measures of that particular dog’s LBM, FM, and TBW. This analysis was performed for each dog, which allowed for a predicted measurement to be generated for each body composition measure for each dog. Paired \(t\) tests were used to compare predicted values to observed values obtained with the D\textsubscript{2}O dilution method. Significance was determined at a value of \(\alpha = 0.05\).

Results

Animals—The study population included both puppies and adult dogs. Body weight of each dog differed by < 2% between the days of QMR and DXA scans (Figure 1).

Dogs were grouped on the basis of mean age and BCS in thin, ideal weight, or overweight categories (Table 1). Puppies had a mean ± SE body weight of 9.4 ± 0.3 kg, and all puppies had a BCS of 4 or 5. Mean body weight of adult dogs was 11.2 ± 1.2 kg, and BCS ranged from 3 to 8. Puppies and adult dogs were analyzed separately to assess the relationship between body weight, age, and BCS. A weak but significant relationship was detected between body weight and BCS for dogs > 1.5 years old (\(r^2 = 0.09; P = 0.04\)), but no relationship was detected between body weight and age (\(r^2 < 0.001; P = 0.99\)).

TBW determined via D2O dilution—For the puppies, the TBW determined via D2O dilution was 5.9 kg, which represented approximately 63.0% of total body weight (range, 58% to 68%; Table 1). For the adult dogs, the TBW differed significantly (\(P = 0.01\)) among

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body condition categories, specifically between dogs with an ideal weight versus dogs that were overweight or thin. As a percentage of body weight, TBW decreased significantly \((P < 0.001)\) with increasing BCS. Pairwise analysis of TBW as a percentage of body weight revealed a difference (although not significant \([P = 0.06]\) \(r^2 = 0.02\)) between the thin and ideal categories. Both thin dogs and dogs with an ideal BCS had significantly higher TBW as a percentage of body weight, compared with the value for overweight dogs. For all dogs, TBW and LBM were significantly related to body weight \((r^2 = 0.776 [P < 0.001] \text{ for both})\), but there was no relationship between age \((r^2 = 0.014; P = 0.33)\) or BCS \((r^2 = 0.005; P = 0.88)\) and TBW. However, TBW as a percentage of body weight was significantly related to BCS \((r^2 = 0.66; P < 0.001)\) and age \((r^2 = 0.29; P < 0.001)\). When data for the puppies were removed from the regression analysis for assessment of age versus TBW as a percentage of body weight, there was a still a significant but diminished relationship \((r^2 = 0.12; P = 0.02)\). This significant age relationship was largely influenced by the 1.5-year-old dogs. Lean body mass was not related to age \((r^2 = 0.016; P = 0.33)\) or BCS \((r^2 = 0.005; P = 0.96)\), but FM was significantly related to age \((r^2 = 0.273; P < 0.001)\), BCS \((r^2 = 0.625; P < 0.001)\), and body weight \((r^2 = 0.413; P < 0.001)\).

**QMR assessment of awake and sedated dogs—**
Quantitative magnetic resonance was used to determine TBW, LBM, and FM in awake dogs and subsequently in sedated dogs. The QMR measurements of TBW, LBM, and FM did not differ between awake and sedated dogs and differed by 2.0%, 2.2%, and 4.3%, respectively \(\text{Figure 1}\). Regression analysis of TBW determined via QMR in the awake dogs, compared with results for the sedated dogs, revealed a significant relationship \((r^2 = 0.944; P < 0.001)\), with a slope of 0.9873 that differed significantly \((P < 0.001)\) from 1 and an intercept of 0.049 that was not significantly \((P = 0.19)\) different from 0. Similarly, regression analysis of LBM determined via QMR, regardless of whether dogs were awake or sedated, revealed that the values were significantly related \((r^2 = 0.956; P < 0.001)\), with a slope of 0.961 that differed significantly \((P < 0.001)\) from 1 and an intercept of 0.128 that did not differ significantly \((P = 0.84)\) from 0. Regression analysis of FM determined via QMR in the awake dogs, compared with results for the sedated dogs, revealed a significant relationship \((r^2 = 0.996; P < 0.001)\) with a slope of 0.985 that differed significantly \((P < 0.001)\) from 1 and an intercept of 0.104 that did not differ significantly \((P = 0.48)\) from 0. All subsequent analyses of TBW, LBM, and FM were conducted with QMR data recorded in awake dogs.

The lean tissue hydration constant was calculated with and without the value for free water \(\text{Table 2}\). The mean estimate of LBM water content was not different between awake and sedated dogs, and there was significant agreement, regardless of whether the hydration constant was calculated with or without the value for free water \((r^2 = 0.209; P < 0.001)\) or excluded \((r^2 = 0.14; P = 0.004)\).

**Comparison of methods for assessment of TBW—**
The QMR significantly \((P < 0.001)\) underestimated TBW by 10.2%, compared with results for D\(_2\)O dilution \(\text{Table 2}\). However, regression analysis of TBW revealed that QMR was significantly related to D\(_2\)O dilution \((r^2 = 0.923; \text{Figure 2})\). The slope was significantly \((P < 0.001)\) different from 1 and an intercept was not significantly \((P = 0.42)\) different from 0. Evaluation of a Bland-Altman plot revealed that significant \((P < 0.001)\) differences were not observed between awake and sedated dogs, and there was significant agreement, regardless of whether the hydration constant was calculated with or without the value for free water \((r^2 = 0.209; P < 0.001)\) or excluded \((r^2 = 0.14; P = 0.004)\).

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**Figure 1—**Box-and-whisker plots of body weight recorded for 58 dogs on the day of QMR and D\(_2\)O analysis and within 7 days after QMR and D\(_2\)O data collection on the day of DXA analysis. Each box represents the interquartile range (25th to 75th percentiles), the square within each box represents the median, and the whiskers represent ± 1.5 times the upper or lower interquartile range. Body weights differed by <2% for all dogs between the days on which analyses were performed.

**Table 1—**Demographics of 58 dogs and TBW of those dogs determined via the D\(_2\)O dilution method.

<table>
<thead>
<tr>
<th>Group</th>
<th>Body weight (kg)</th>
<th>Age (y)</th>
<th>BCS†</th>
<th>TBW mass (kg)</th>
<th>Mean ± SD</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ideal weight puppies (n = 16)</td>
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<td></td>
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<td></td>
</tr>
<tr>
<td>Thin (n = 4)</td>
<td>9.6 ± 2.1±</td>
<td>5.5 ± 4.5±</td>
<td>3</td>
<td>6.0 ± 1.7±</td>
<td>61.3 ± 5.3±</td>
<td>55–67</td>
</tr>
<tr>
<td>Adult dogs</td>
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<td></td>
</tr>
<tr>
<td>Overweight (n = 19)</td>
<td>11.4 ± 2.2±</td>
<td>8.8 ± 1.6±</td>
<td>4–5</td>
<td>7.0 ± 1.3±</td>
<td>60.2 ± 2.7±</td>
<td>56–86</td>
</tr>
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<td>9.6 ± 2.1±</td>
<td>5.5 ± 4.5±</td>
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<td>60.2 ± 2.7±</td>
<td>56–86</td>
</tr>
</tbody>
</table>

Values reported are mean ± SD or range.

*Ideal weight puppies comprised 8 females and 8 males, thin adult dogs comprised 4 males, ideal weight adult dogs comprised 6 females and 13 males, and overweight adult dogs comprised 10 females and 9 males. The BCS was scored on a scale of 1 to 9; ideal body weight was a BCS of 4 or 5, thin was a BCS < 4, and overweight was a BCS > 6.

**Within a column, values with different superscript letters differ significantly \((P < 0.05)\).**
bias existed for assessment via QMR to underestimate TBW and that the underestimation became significantly greater with increasing TBW.

Comparison of methods for assessment of LBM and FM—Both QMR and DXA significantly \((P < 0.001)\) underestimated LBM by 13.4% and 7.3%, respectively, compared with values determined by \(D_2O\) dilution (Table 2). Regression analysis of LBM determined via QMR and \(D_2O\) dilution revealed that there was close agreement \((r^2 = 0.931 \ [P < 0.001];\) Figure 3). The slope and intercept both differed significantly from 1 \((P < 0.001)\) and 0 \((P = 0.008)\), respectively. Regression analysis also revealed that LBM determined via DXA and \(D_2O\) dilution was significantly related \((r^2 = 0.956 \ [P < 0.001];\)

### Table 2—Mean ± SD values for body composition of 58 dogs determined via the \(D_2O\) dilution method, QMR (scan was obtained with each dog awake but placed in a polymethylmethacrylate crate to minimize movement or sedated to eliminate movement), and DXA.

<table>
<thead>
<tr>
<th>Body composition component</th>
<th>(D_2O)</th>
<th>Awake</th>
<th>Sedated</th>
<th>DXA</th>
</tr>
</thead>
<tbody>
<tr>
<td>TBW (kg)</td>
<td>6.37 ± 1.21*</td>
<td>5.72 ± 1.01*</td>
<td>5.60 ± 1.02*</td>
<td>NA</td>
</tr>
<tr>
<td>LBM (kg)</td>
<td>8.70 ± 1.80*</td>
<td>7.54 ± 1.30*</td>
<td>7.30 ± 1.34*</td>
<td>8.07 ± 1.52*</td>
</tr>
<tr>
<td>FM (kg)</td>
<td>2.11 ± 0.99*</td>
<td>1.79 ± 0.95*</td>
<td>1.86 ± 0.93*</td>
<td>1.93 ± 1.05*</td>
</tr>
<tr>
<td>Free water (kg)</td>
<td>NA</td>
<td>0.08 ± 0.08</td>
<td>0.07 ± 0.05</td>
<td>NA</td>
</tr>
<tr>
<td>LBM hydration estimate (%)*</td>
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<tr>
<td>Excluding free water</td>
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</tr>
<tr>
<td>Including free water</td>
<td>79.0 ± 3.01*</td>
<td>75.9 ± 1.42*</td>
<td>76.0 ± 1.40*</td>
<td>NA</td>
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<tr>
<td>LBM hydration estimate (%)*</td>
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<td>Excluding free water</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Including free water</td>
<td>79.0 ± 3.01*</td>
<td>75.9 ± 1.42*</td>
<td>76.0 ± 1.40*</td>
<td>NA</td>
</tr>
</tbody>
</table>

*Lean body mass hydration estimate determined by excluding the value for free water was calculated as \(100 \times (TBW - \text{free water}) / LBM\). The LBM hydration estimate determined by including the value for free water was calculated as \(100 \times TBW / LBM\). Calculated with TBW determined via \(D_2O\) dilution and LBM determined via QMR in awake dogs.

NA = Not applicable.

Within a row, values with different superscript letters differ significantly \((P < 0.05)\).

![Figure 2](image2.png)

![Figure 3](image3.png)
The slope differed significantly \( (P < 0.001) \) from 1, but the intercept was not significantly \( (P = 0.43) \) different from 0. Evaluation of a Bland-Altman plot revealed that a significant bias existed for the QMR \( (P < 0.001) \) and DXA \( (P = 0.006) \) method to underestimate LBM, compared with results for D\(_2\)O dilution. For both QMR and DXA, the underestimation became greater with increasing LBM.

Similar to results for TBW and LBM, QMR and DXA both significantly \( (P < 0.001) \) underestimated FM by 13.4\% and 8.4\%, respectively, compared with results for the D\(_2\)O method (Table 2). Regression analysis of FM measured via QMR and D\(_2\)O dilution was significantly related \( (r^2 = 0.89 \; [P < 0.001]; \text{Figure 5}) \). Linear regression analysis indicated that the slope and intercept were significantly \( (P < 0.001) \) different from 1 and 0, respectively. Evaluation of a Bland-Altman plot indicated that no significant \( (P = 0.36) \) bias existed for the relationship between FM determined via QMR and D\(_2\)O dilution. Similar to results for QMR, regression analysis of FM determined via DXA and D\(_2\)O dilution was significantly related \( (r^2 = 0.88 \; [P < 0.001]; \text{Figure 6}) \). The slope and intercept both were significantly \( (P < 0.03) \) different from 1 and 0, respectively. Evaluation of a Bland-Altman plot indicated no significant \( (P = 0.21) \) bias for DXA to overestimate or underestimate FM.

The significant relationships between values obtained via D\(_2\)O dilution and QMR allowed for the generation of correction equations for TBW, LBM, and FM by linear regression analysis (Table 3). In addition, correction equations were also derived for values obtained via DXA. Prediction equations were evaluated via a cross-validation procedure and revealed that LBM, FM, and TBW could be predicted from QMR and DXA results in these dogs. The mean errors for LBM, FM, and TBW determined via QMR were 4.0\%, 14.8\%, and 4.4\%, respectively, whereas the mean errors of LBM and FM determined via DXA were 3.5\% and 16.2\%, respectively. For each body composition component, the mean of predicted values did not differ significantly \( (P > 0.98) \) from observed values, and the CV between predicted and observed values of LBM \( (r^2 > 0.93) \) and FM \( (r^2 > 0.87) \) determined via QMR or DXA were significantly \( (P < 0.001) \) related, as was TBW determined via QMR \( (r^2 = 0.92) \).

**Comparison of methods for generating the LBM hydration constant**—The proportion of water associated with the LBM for each dog was calculated by use of TBW and LBM determined via QMR in awake dogs or TBW determined via D\(_2\)O dilution and LBM determined via DXA. The LBM hydration constant determined via QMR data was significantly \( (P < 0.001) \) related to the...
percentage of body fat measured via QMR ($r^2 = 0.32$; Figure 7) or DXA ($r^2 = 0.32$; data not shown) and to BCS ($r^2 = 0.26$; data not shown). There was an inverse relationship, with decreasing values for the LBM hydration constant relative to increasing values for BCS or percentage of body fat. No relationship was detected with age ($r^2 = 0.05$; $P = 0.08$). For comparison purposes, the LBM hydration constant was calculated by use of TBW determined via $D_2O$ dilution and LBM determined via DXA. The estimate for the mean of the LBM hydration constant differed significantly ($P < 0.001$) when data obtained via the QMR or $D_2O$ dilution and DXA methods were used for calculation (Table 2). A different relationship was evident when comparing the LBM hydration constant determined via $D_2O$ dilution or DXA to the percentage of body fat determined via DXA ($r^2 = 0.137$; $P = 0.004$) or QMR ($r^2 = 0.10$; $P = 0.01$).

**Discussion**

The present study was designed to provide the initial evaluation of the QMR method in dogs. Results indicated the accuracy of QMR for measurement of TBW, LBM, and FM in awake and sedated dogs. Consequently, this study validated that QMR can be used as an effective method for rapidly and accurately measuring body composition of awake dogs.

In the study reported here, QMR involved use of a QMR system designed for acquisition of measurements from animals ranging in body weight from 4 to 50 kg. Highly precise body composition data were initially reported for dogs, in which a mean CV $< 2.0\%$ was observed for all measurements, except for significantly reduced precision in fat measurements for dogs with 5% body fat. That study was based on 5 scans for each of 8 dogs and indicated better results than a previous study involving the use of DXA in dogs and cats, which had a CV of approximately 6% for repeated scans.

Accuracy of the QMR results reported here was based on a single scan so that there would not be a bias for DXA, for which only a single scan also was acquired; thus, we believe this provided a fair evaluation of accuracy, compared with results for the reference standard. The QMR method was used to obtain highly accurate (< 4.3% difference) and highly related ($r^2 > 0.94$) values for FM, LBM, and TBW in awake dogs, compared with results for sedated dogs, which provided a high degree of confidence in routine acquisition of body composition components in awake animals. Although the QMR method underestimated body composition components in dogs by 10% to 15%, compared with results for $D_2O$ dilution as the reference method, the resulting data were highly related ($r^2 > 0.89$) between methods. When DXA was used, the data reaffirmed that both LBM and FM can be accurately measured and highly related ($r^2 > 0.88$) to values for $D_2O$ dilution, as determined in the same dogs used for QMR.

Data in the present study are consistent with other published results related to QMR. Analysis of results for the study reported here and in other studies indicates

Table 3—Correction equations for TBW, LBM, and FM prediction for QMR and DXA on the basis of results for the $D_2O$ dilution method.

<table>
<thead>
<tr>
<th>Dependent variable</th>
<th>Regression equation</th>
<th>Model $r^2$</th>
<th>Cross-validation*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>Difference (kg)</td>
</tr>
<tr>
<td>TBW QMR</td>
<td>(1.1506•QMR$_{TBW}$) + 0.2104</td>
<td>0.923T</td>
<td>$-0.06 \pm 0.05$</td>
</tr>
<tr>
<td>LBM QMR</td>
<td>(1.1659•QMR$_{LBM}$) + 0.088</td>
<td>0.931T</td>
<td>$-0.08 \pm 0.06$</td>
</tr>
<tr>
<td>LBM DXA</td>
<td>(1.0591•DXA LBM) + 0.1549</td>
<td>0.956T</td>
<td>$-0.06 \pm 0.05$</td>
</tr>
<tr>
<td>FM QMR</td>
<td>(0.9844•QMR$_{FM}$) + 0.3526</td>
<td>0.890T</td>
<td>$0.10 \pm 0.05$</td>
</tr>
<tr>
<td>FM DXA</td>
<td>(0.9885•DXA FM) + 0.3991</td>
<td>0.881T</td>
<td>$0.08 \pm 0.05$</td>
</tr>
</tbody>
</table>

*Mean $\pm$ SE values for the absolute difference and the percentage difference between predicted and actual measurements for the $D_2O$ dilution method. TValue of the model $r^2$ was significant ($P < 0.001$).
that FM, TBW, and LBM are underestimated or overestimated via QMR, compared with values for the reference method, and this underestimation or overestimation differs substantially by species. In this study, QMR underestimated all 3 body composition components in awake dogs, compared with data obtained via D₂O dilution. The TBW determined in this study was significantly underestimated by 10.2%. This is in contrast to TBW determined via QMR in most other species, which reportedly was underestimated by 5% in rats and 3.1% or 6.4% in baby pigs and overestimated by 4% in chickens; however, all of those studies involved the use of chemical analysis of carcasses as the reference method. On the basis of results for D₂O dilution as the reference method. In addition, no bias was evident with QMR or DXA when measuring FM in mice by 29% and LBM in infants, FM was overestimated by 10%, compared with results for the 4-compartment model, whereas it was underestimated by 4%, compared with results for D₂O dilution. Overall, the correction equations for LBM and TBW generated in the present study will be useful in improving the accuracy of data obtained in canine body composition studies that use QMR. However, use of correction equations to predict FM still requires caution during interpretation of QMR data because a 15% mean error was evident when the correction equations were evaluated in a cross-validation analysis. The high percentage difference observed with the cross-validation assessment of FM was influenced by QMR data for 5 dogs that had >35% error between predicted and observed FM determined via D₂O dilution. Post hoc analysis of the percentage mean difference without these 5 dogs reduced the mean value for QMR to 10%, which indicated that the QMR correction equation for FM can be used to predict FM for 90% of dogs in the present study within 90% of actual FM as determined with results for existing methods such as DXA. Analysis of the QMR method will allow for possible refinement of the use with larger breeds of dog, particularly given that the largest dog used in the present study was only 60% of the maximum recommended body weight range of the QMR unit. However, we detected a similar bias for determination of LBM via DXA. Therefore, the limitation may have been associated more with the use of D₂O dilution as the reference method. In addition, no bias was evident with QMR or DXA when measuring higher amounts of FM. Ultimately, these results indicated that QMR was capable of providing accurate data in dogs, likely across multiple breed sizes, which are highly related to reference measures and comparable to results for carcass analysis. However, QMR has also been associated with larger variation, compared with results for the reference method, in which measurements overestimated FM in mice by 29% and underestimated FM in chickens by 34% and underestimated FM in cats by 30.3%, compared with results for D₂O dilution. In humans, FM was underestimated more in males (13% to 18%) than females (2% to 8%). Depending on the reference method used for assessment in infants, FM was overestimated by 10%, compared with results for the 4-compartment model, whereas it was underestimated by 4%, compared with results for D₂O dilution.
ated with LBM is generally considered to be 73.2%32,33; calculated from data acquired via QMR with TBW and conclusions was minimal.

As a result, we believe that the effect on the no change in the distribution of the data between the 2 time points was < 2% and that there was no change in the distribution of the data between the 2 time points (Figure 1), we believe that the effect on the conclusions was minimal.

The LBM hydration constant can be effectively calculated from data acquired via QMR with TBW and LBM measurements. The hydration constant associated with LBM is generally considered to be 73.2%32,33; however, the QMR method can be used to provide a determination for each animal. Although investigators in another study1 confirmed the estimate of 73.2% in a group of cats and dogs, they used only 6 dogs, with no indication of body weight, age, or BCS that was specific to the dogs alone. In another study,2 investigators reported a similar, but slightly lower, mean LBM hydration constant in dogs of 71.3%, which was also based on carcass analysis and was representative of 75 lean and overweight dogs. Because the LBM hydration constant can be used to calculate the percentage of body fat (eg, use of D2O dilution to determine body composition measures), investigators in that latter study2 used both 71.3% and 73.2% and determined that the difference accounted for extremely little variation in the estimate of the percentage of body fat.

The ability to simultaneously and accurately measure TBW and the LBM hydration constant may provide insight on the hydration status, or at least influence the assumptions used to determine components of body composition of an animal, particularly in overweight animals. The LBM hydration constant has biological importance that is also applicable to growth, sex, body size, and acute or chronic illnesses.32 It is not surprising that the TBW proportion decreases with increasing obesity that results in increased body weight. Evaluation when the percentage of body fat is increased and the LBM hydration constant is reduced may offer insight into an animal's hydration status and reveals that the assumption of 73.2% may need to be used with caution in studies of overweight or obese populations of dogs.

The assumption of a standard LBM hydration constant and reference range of TBW values has also been a topic of discussion related to human nutrition and obesity, particularly because overweight or obese humans are currently more prevalent.32,33 A study35 in humans also found that the LBM hydration constant increases with age, but this was driven by a decrease in LBM, with age and TBW remaining unchanged. Additional studies will be necessary to examine how the LBM hydration component is influenced in an overweight or obese geriatric population for both companion animals and humans.

Calculation of the mean LBM hydration constant in the present study with the value for body water derived via QMR for each dog, which was divided by QMR-based LBM, yielded an estimate of 75.9% and 76.0% in awake and sedated dogs, respectively. Exclusion of the value for free water resulted in a reduction of 1% in these mean values. On the basis of QMR determination of the hydration constant for each animal, the observed means were extremely similar to values reported for cats.15 The mean LBM hydration constants reported here are slightly higher than those determined with values obtained via carcass analysis of dogs.1,2

Similar to values for cats reported in another study15 conducted by our research group, increased reliability of the hydration constant data is achieved when multiple QMR scans are performed, versus data obtained during a single scan. Minimal agreement (r² = 0.20) was observed in the relationship of values obtained for awake versus sedated dogs when only a single scan was performed. Analysis of data for assessment of the precision for QMR in dogs in another study26 revealed that the agreement (r² = 0.98) for values obtained in awake and sedated dogs improved considerably when the mean of 5 consecutive scans/dog was used. This indicated that both high precision and greater accuracy can be achieved for data obtained during multiple scans, which improves the reliability of measuring the LBM hydration constant in awake dogs.

Excessive body weight and an increase in obesity among pet cats and dogs is a growing concern. Methods such as DXA, bioelectrical impedance spectroscopy,36–38 and CT31 have been evaluated for their use in measuring body composition in dogs or cats. Every method has limitations and advantages. The use of DXA and CT routinely require that an animal be anesthetized to prevent movement, which has greater health risks for extremely young, old, or sick animals, which consequently limits routine use to healthy and adult animals.

Similar to results of another study15 conducted by our research group that involved the use of QMR to measure body composition in cats, the present study indicated that QMR is a useful technique for the determination of body composition in dogs and is suitable for longitudinal studies. Use of technology such as QMR in a research setting or at a teaching institution can allow veterinarians and animal nutritionists to safely and frequently assess loss of adiposity as well as evaluate maintenance of LBM following various interventions. Of particular advantage, QMR provides a safe, quick, and accurate method for assessment of healthy awake adult dogs and, more importantly, dogs that are extremely young or old or that have some degree

AJVR, Vol 74, No. 5, May 2013
of compromised health that would make them unsuitable for sedation or anesthesia procedures. Therefore, TBW and other body composition components can be measured in these groups of dogs. Finally, comparison with the D₂O dilution method allowed for the development of correction equations to provide accurate data over a range of body composition values. Ultimately, this provides a promising advancement for noninvasive assessment of weight loss and weight management in companion animals.

The QMR technique is readily applicable to clinical practice with distinct advantages, compared with the use of DXA and D₂O dilution, particularly because it eliminates the need for sedation or anesthesia of subjects and is thus completely noninvasive. None of the dogs in the present study had been acclimated to being placed in the polymethylmethacrylate crate, and it is highly likely that this would be accepted by most client-owned dogs, especially if they are used to being transported in a pet carrier. The greater precision of QMR (CV, 2%), compared with DXA (CV, 6%–28), implies that QMR will be a valuable technique for assessment of weight management in individual animals on a longitudinal basis. Although BCS determined by experienced personnel is a convenient and practical measure of body composition, it is qualitative in nature, whereas QMR provides a reliable quantitative measure that eliminates the subjectivity of BCS assessment among observers.

References


