DEPTH-CORRECTED VERSUS NON DEPTH-CORRECTED GFR

DETERMINATION BY QUANTITATIVE RENAL

SCINTIGRAPHY IN THE DOG

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DEPTH-CORRECTED VERSUS NON DEPTH-CORRECTED GFR DETERMINATION BY QUANTITATIVE RENAL SCINTIGRAPHY IN THE DOG

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May 9, 2009

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Gregory Thomas Almond

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Date of Graduation

VITA

Gregory Thomas Almond, son of Walter Rudolph Almond, Jr. and Patricia Irvin Almond, was born March 29, 1960 in Memphis, Tennessee. He graduated with honors from Briarcrest Baptist High School in 1978. He attended Baylor University, the University of Memphis, and the University of Tennessee at Knoxville as an undergraduate before entering the University of Tennessee College of Veterinary Medicine, where he received the degree of Doctor of Veterinary Medicine in 1984. He practiced veterinary medicine as an associate veterinarian in Memphis, Tennessee until April, 1996 when he founded Cordova Station Animal Hospital in Memphis, Tennessee. After 6 successful years of small animal private practice ownership, he decided to pursue a dream of furthering his education by entering a residency in radiology at Auburn University in August 2002. He completed his radiology residency in 2005 and began a residency in radiation oncology in the same year. He was awarded diplomat status in the American College of Veterinary Radiology in the fall of 2007. He completed his residency in radiation oncology in 2008. He married Tina, daughter of John Joseph Watkins and Linda Blakely Watkins in February 1993. They have 2 children, Seth Elliott and Sarah Elizabeth.

THESIS ABSTRACT

DEPTH-CORRECTED VERSUS NON DEPTH-CORRECTED GFR DETERMINATION BY QUANTITATIVE RENAL SCINTIGRAPHY IN THE DOG

Gregory Thomas Almond

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Using established methodology, 99mTc-diethylenetriaminepentaacetic acid (99mTc-DTPA) quantitative renal scintigraphy was performed on 22 healthy sedated dogs to measure glomerular filtration rate (GFR). Using each dog as its own control, GFR values were calculated with and without correction for photon attenuation due to kidney depth. The renal depth measurement was made from a right lateral scintigraphic image. Simultaneously, plasma clearance (PC) of 99mTc-DTPA was determined using a two-sample, one-compartment pharmacologic model with samples at 20 and 180 minutes. One to three days later, the dogs were sedated and computed tomographic (CT) images

were obtained of both kidneys with the dogs positioned as during scintigraphy. Measurements of renal depth were made from both the center of the kidney and the renal crest to the skin directly dorsal to the center of the kidney and to a line perpendicular to the skin at the dorsal midline. The dogs were manually lifted and positioned again and a second CT scan was performed to evaluate repeatability of these measurements. For all dogs, depth-corrected (DC) and non depth-corrected (nDC) GFR measurements were not different ($p=0.3926$). For small dogs, DC and nDC GFR measurements were not different ($p=0.2332$). Correlations between scintigraphy and CT depth measurements were poor for the left kidney and modest for the right kidney, making depth correction of doubtful value in dogs of this weight. For large dogs, DC and nDC GFR measurements were different ($p=0.0263$). Correlations between scintigraphy and CT depth measurements for the left kidney were modest, suggesting that using scintigraphy for depth correction may introduce error into depth corrected GFR measurement in large dogs. Before recommendations are made to abandon depth correction in large dogs it would be necessary to repeat the study with the gold standard of inulin clearance. Separate linear regression formulas for small and large dogs may improve the accuracy of GFR measurements by scintigraphy. DC and nDC GFR measurements were significantly different from PC when considering all dogs together ($p<0.0001$ and $p=0.0003$ respectively) and when considering small dogs alone ($p<0.0001$ for both). In large dogs, PC was also found to significantly differ from DC ($p=0.0016$). Only when comparing PC to nDC GFR in large dogs were the values significantly similar ($p=0.5179$). Because numerous other studies have indicated good agreement between scintigraphy and plasma clearance, the plasma clearance measurements obtained in this study are questionable.

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Style manual or journal used: Veterinary Radiology and Ultrasound

Computer software used: Microsoft Word 2003, Microsoft Excel 2003, Statistical Analysis System (SAS) 8.2, Mirage Processing Application 5.407 sp18

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I. INTRODUCTION

Because renal failure is a common debilitating clinical condition in dogs, early detection of decline in renal function is important for diagnosis and management of this condition. One of the most reliable tests of renal function is the measurement of glomerular filtration rate (GFR).[1] Methods that have been used to determine GFR in the dog include inulin clearance, endogenous and exogenous creatinine clearance, nuclear scintigraphy and plasma clearance of radioisotopes.[2] Although inulin clearance is generally considered the gold standard for GFR determination, technical considerations preclude its routine use in a clinical setting.[1,3] Quantitative renal scintigraphy using the radiopharmaceutical ^{99m}Tc-diethylenetriaminepentaacetic acid (commonly abbreviated as ^{99m}Tc-DTPA or DTPA) provides a reliable and relatively easy means of GFR determination. This procedure has been validated by comparison of scintigraphic ^{99m}Tc-DTPA uptake to inulin clearance. Using linear regression analysis, an equation was derived with scintigraphic ^{99m}Tc-DTPA uptake to calculate GFR,[3] which has gained wide acceptance.

For scintigraphic GFR quantification, a dorsal view dynamic acquisition is obtained after injection of DTPA into a peripheral vein. To improve correlation with inulin clearance in dogs, current protocols recommend correction for the attenuation of gamma radiation in the intervening soft tissues between each kidney and the gamma camera. Attenuation correction, commonly called depth correction, requires that a static

lateral image be obtained after the dynamic acquisition to measure the depth of the kidney from the dorsal skin surface. Based on the depth measurement, the ^{99m}Tc-DTPA uptake is mathematically increased prior to use in the linear regression formula in order to account for attenuation. Obtaining this additional image increases the length of time the dog must be restrained and requires additional computations in the post image processing.

Despite wide acceptance of the equation relating scintigraphic ^{99m}Tc-DTPA uptake to GFR, the accuracy of the depth measurement obtained from the right lateral scintigraphic image has not been validated. Also, lateral scintigraphic images of the abdomen frequently result in overlap of the kidneys so that the location of the center of each kidney must be estimated. Nuclear scintigraphy provides valuable functional information, but has inherently poor spatial resolution. In contrast, computed tomography has excellent spatial resolution. If it can be shown that the spatial measurements obtained by scintigraphy are inaccurate when compared to computed tomography, the validity of using the scintigraphic measurements to correct for gamma ray attenuation between the kidneys and the gamma camera becomes questionable.

It is also important to note that Krawiec *et al.* found only a slight improvement in predicting GFR by using depth correction in dogs.[3] The authors of that study noted that in humans, correlations improve "dramatically" with depth correction. They suggested that the difference in importance of depth correction between human patients and dogs is related to the fact that there is more tissue between the kidney and the camera in people than in dogs. Dog size was not addressed in Krawiec's study.[3] That study used 12 mixed

breed dogs, with no reference given to size. It is also possible that the size of the dog influences the importance of depth correction.

Therefore, failure of depth correction to more dramatically improve data correlation may be the result of inaccuracy of the renal depth measurement used in depth correction or failure to account for variability in dog size. Alternatively, the linear regression formula used previously[3] could be inaccurate in some cases. Consequently, it is important to assess the accuracy of the depth measurement obtained by renal scintigraphy in dogs and explore possible differences between dogs of different size. It is equally important to determine if depth correction is necessary.

The purposes of this study were 1) to determine if the additional step of depth correction for tissue attenuation is necessary in ^{99m}Tc-DTPA scintigraphy studies for determination of GFR in both large and small dogs and 2) to determine if the current depth correction procedure accurately assesses depth.

This study was designed to test the following hypotheses:

1. Scintigraphic measurements of global GFR in dogs using the radiopharmaceutical ^{99m}Tc-DTPA are not statistically different whether they are depth-corrected or non depth-corrected.
2. Both depth-corrected and non depth-corrected measurements correlate well with DTPA plasma clearance.
3. Renal depth measurement using DTPA scintigraphy is unreliable when compared to computed tomography measurements.

II. LITERATURE REVIEW

Detecting Renal Failure Without the Use of Nuclear Medicine

Veterinarians frequently are presented with dogs whose history suggests renal dysfunction. Whereas many of these patients may already have significant alterations in urine specific gravity (USG), blood urea nitrogen (BUN) and/or serum creatinine levels at time of clinical presentation, some animals are examined prior to alterations in these routine screening tests. The kidneys have tremendous reserve capacity. The loss of the ability to concentrate urine occurs only after two-thirds of the nephrons are affected, and BUN concentration increases only after 75% of the nephrons have failed. Clearly, diagnostic methods that detect early loss of renal function are desirable. Early detection may allow therapeutic intervention that can retard or halt the course of the disease, with treatment usually being more effective when initiated early in the course of the disease.[4]

Measurement of GFR is one of the most reliable indicators of renal function.[1] It is defined as the quantity of glomerular filtrate formed each minute by all nephrons of both kidneys.[5] Classical methods for determining GFR include inulin clearance measurement and endogenous or exogenous creatinine clearance measurement tests. These tests are cumbersome, difficult to perform accurately, and require multiple urinary bladder catheterizations.[1,3] These studies only provide measurement of global renal function and cannot quantify individual kidney function.[1]

Although BUN and creatinine provide valuable information in the investigation of renal disease, neither of these parameters truly assesses renal function. Passive reabsorption of urea in the renal tubules can occur, making BUN an unreliable indicator of GFR. Vascular volume depletion can result in decreased urea clearance without a decrease in GFR.[2] High-protein meals, gastrointestinal bleeding some drugs, and catabolic states can increase BUN concentration; low-protein diets, portosystemic shunts, severe liver disease, and some drugs can decrease BUN concentration.[2] Any of these factors can affect BUN without a change in GFR.[2] Although serum creatinine is predictably and inversely related to GFR, its usefulness in early detection of renal failure is hampered by the fact that the reference range for normal plasma creatinine concentration in dogs is wide and absolute changes in creatinine concentration are small in early chronic renal failure.[4]

Methods of GFR Determination Using Nuclear Medicine

The radiopharmaceutical ^{99m}Tc-diethylenetriaminepentaacetic acid (^{99m}Tc-DTPA) has been used successfully for determination of GFR using scintigraphic imaging[1,3,,6] and plasma clearance methods.[6,7,8] ^{99m}Tc-DTPA is fully filtered by the glomeruli, and there is no tubular secretion or absorption. There is minimal protein binding[3,9,10] and 20% of ^{99m}Tc-DTPA is extracted from the bloodstream during the first pass through the kidneys.[9,10] These properties make DTPA an ideal agent for GFR determination. The advantage of nuclear medicine studies over classical methods of GFR determination rests in their relative ease of performance, lack of invasiveness, availability of reagents, and

their ability to be performed a timely manner.[1,3] Additionally, patients encounter minimal stress during renal scintigraphy.[1]

In a study comparing the scintigraphic uptake and plasma clearance of ^{99m}Tc-DTPA to inulin clearance, Barthez *et al*. concluded that although both methods provide reasonable estimates of GFR, plasma clearance is more precise than scintigraphic uptake.[6] The correlation coefficient between DTPA plasma clearance and inulin clearance was 0.98, and the correlation coefficient between percentage kidney uptake and inulin clearance was 0.86. In two subsequent studies, Barthez *et. al.* investigated the effects of sample number and time on determination of GFR by plasma clearance methods.[7,8] Using a twelve-point ^{99m}Tc-DTPA plasma clearance curve as a reference, they determined that 1 or 2 blood samples produced results with a reasonable margin of error.[8]

As with inulin or creatinine clearance methods, ^{99m}Tc-DTPA plasma clearance methods provide measurement of global renal function.[6] Scintigraphy provides percentage uptake in each kidney.[6] For that reason it is superior to plasma clearance determination in situations where information on individual kidney function is necessary, such as prior to nephrectomy or nephrotomy.[6] Another advantage of scintigraphic uptake methods over plasma clearance methods is that information is obtained more quickly and with less patient discomfort.[1] Although scintigraphic methods are completed in less time than plasma clearance methods, there is still opportunity for improvement in the speed of this procedure.

Presently, scintigraphic determination of GFR in the dog involves intravenous injection of ^{99m}Tc-DTPA and immediately obtaining a dynamic study with 10 images per

minute for four minutes. With the dog kept in the same position the camera is rotated 90° to obtain a 45 to 60 second static right lateral image of the abdomen for depth attenuation correction.[9,10] ^{99m}Tc gamma rays are absorbed in tissue with a linear attenuation coefficient in soft tissue of 0.153/cm,[9,10] resulting in decreased counts from the deeper left kidney. The linear regression formula which serves as the standard for scintigraphy-based GFR determination in the dog is found in a 1986 study by Krawiec *et al*.[3] In that study it was noted that "tissue depth correction of percentage dose in each kidney only slightly improved the inulin clearance vs. percentage dose *r* value". Despite the fact that this study showed only a slight improvement with depth correction (correlation coefficient of 0.916 without depth correction versus 0.941 with depth correction), current protocols in the dog call for depth correction for attenuation. In a 1992 study by Uribe *et al*., GFR determination in cats via renal scintigraphy was compared with inulin and creatinine clearance methods.[11] It was found that correction for kidney depth slightly worsened correlation between scintigraphic methods and inulin clearance. The authors concluded that this may be related to species differences because cats have less tissue between the skin and kidneys than dogs and man. Based on that study, correction for attenuation due to renal depth is not recommended in cats.

Reliability of Spatial Measurements in Nuclear Medicine

Whereas nuclear medicine studies enjoy the advantage of providing valuable functional information, their spatial resolution is poorer than other imaging modalities. The reported intrinsic spatial resolution of modern scintillation cameras is between 3 and

7 mm.[10] Computed tomography has a reported spatial resolution of 0.5 mm[12]. To this author's knowledge, there are no published studies directly comparing the reliability of renal depth measurements obtained by nuclear scintigraphy and computed tomography. Because the currently accepted scintigraphic methodology for depth correction relies on the accuracy of renal depth measurement, it would be beneficial to assess renal depth measurements obtained in scintigraphy by comparison with those obtained in computed tomography.

Effect of Sedation Protocols on Scintigraphic GFR Determination

Several studies have examined the effects of different sedation protocols on scintigraphic determination of GFR. [13,14] During the examination, the patient is required to remain motionless for approximately 6 minutes, and any movement by the patient can result in inaccuracies in GFR determination.[13,14] Newel *et al*. measured the effects of three common sedative protocols on GFR in dogs: 1) butorphanol and diazepam, 2) acepromazine and butorphanol, and 3) diazepam and ketamine.[13] In that study it was found that the acepromazine and butorphanol protocol resulted in a significant decrease in mean arterial pressure and heart rate, but surprisingly yielded GFR values identical to those obtained in awake dogs. The authors proposed that maintenance of GFR despite a drop in mean arterial pressure with this protocol may be the result of autoregulation of renal blood flow or due to "preferential vasodilatation of renal versus systemic vasculature".[13] They recommended the acepromazine and butorphanol protocol for renal

scintigraphy in normal dogs because it provided optimal sedation without significant effect on GFR.

III. MATERIALS AND METHODS

Animal Subjects

Twenty-two purpose-bred, random source dogs were used in this study. Two dogs weighing less than 13.6 kg (30 lbs) were used as pilots before the study to refine technique. Of the remaining 20 dogs, 10 dogs weighed less than 13.6 kg (30 pounds) and 10 dogs weighed more than 22.7 kg (50 lbs). A complete physical exam, CBC, serum biochemical analysis, and urinalysis were performed on each dog prior to the study and no significant abnormalities were noted in any dog. During ultrasound-guided cystocentesis one dog was found to have multiple cystic calculi. Prior to the study, another dog was discovered to have canine dirofilariasis. No physical examination or screening test abnormalities were noted in these two dogs that would preclude their inclusion in the study.

Four dogs were housed in the Nuclear Medicine Isolation Ward, Auburn University College of Veterinary Medicine for a minimum of 48 hours after ^{99m}Tc-DTPA injection. The remaining 18 dogs were isolated in facilities maintained by the Division of Laboratory Animal Health (DLAH), Auburn University College of Veterinary Medicine, separate from other DLAH animals for a minimum of 48 hours after ^{99m}Tc-DTPA injection. Feeding and daily care for all dogs in the study were performed by the author. The change in housing was necessitated by the reluctance of the initial four dogs to urinate and defecate in the metabolism cages in the Nuclear Medicine Isolation Ward.

Nuclear Scintigraphy Acquisition

Quantitative renal scintigraphy study was performed on each dog after an overnight fast. An intravenous catheter was placed in a cephalic vein. Acepromazine was administered at a dosage of 0.05 mg/kg, and after 15 minutes butorphanol was administered at a dosage of 0.2 mg/kg. All quantitative renal scintigraphy studies were performed using one to three millicuries (mCi) of ^{99m}Tc-DTPA purchased from a local radiopharmacy[15]. During the course of the study, the labeling efficiency of the DTPA ranged from 95 to 99 percent, with an average of 98 percent.[16] Scintigraphy was performed using a gamma camera equipped with a low energy all purpose (LEAP) collimator.[17] Images were acquired using Mirage® Acquisition Application software.[18] The standard accepted protocol for ^{99m}Tc-DTPA scintigraphic determination of GFR as listed in the Textbook of Veterinary Nuclear Medicine[10] was followed with minor modifications.

1. Pre counts of the unshielded dose syringe were obtained a distance of 30 cm from the center of the gamma camera as a one minute dynamic acquisition.
2. The dog was placed in left lateral recumbency with the gamma camera positioned to obtain a dorsal view of the abdomen. Using manual restraint and/or medical tape, the dog was positioned with as much of the dog's dorsum in contact with the gamma camera as possible. The dog's spine and legs were not flexed or extended.
3. One to three millicuries (mCi) of ^{99m}Tc-DTPA was injected via the cephalic catheter and dynamic acquisition was begun simultaneously. Using six-second

frames the images were stored in a 64 x 64 x 16 matrix for a total of four minutes (Fig 1).

4. To obtain depth measurements, the dog was kept in the same position and the camera was rotated 90° to obtain a 60-second static right lateral image of the abdomen. This image was stored in a 128 x 128 x 16 matrix. Measurements of renal depth and image diameter were made by direct measurement of the images on the computer screen using a translucent ruler. Renal depth was considered to be from the dorsal surface of each dog to the midpoint of each kidney.
5. Separation of the left and right kidneys on the lateral image of the abdomen was assessed subjectively. The appearance was classified into one of the following four categories: left and right kidneys distinct (D), kidneys overlapped producing a single elongated image (OE), kidneys overlapped but discernable (OD), or kidneys completely superimposed (CS) (Fig 2).
6. To obtain the number of counts that were not injected into the patient (post injection counts), the gamma camera was rotated to its original position, and the dose syringe was taped to the dog's leg adjacent to the cephalic catheter. The rest of the dog's body was shielded from the camera. A one minute dynamic acquisition was obtained of the syringe and cephalic catheter at a distance of 30 cm from the center of the camera.

Figure 1. Still images of dynamic renal scintigraphy of dog #10.

The upper left-hand corner image was acquired immediately after injection began. Each of the 40 frames represents 6 seconds of time elapsed for a total of 240 seconds (4 minutes). Images proceed from left to right and from top to bottom. In each image cranial is at the top, the dog's right is to the right, and the dog's left is to the left. The dog was positioned in left lateral recumbency, and the gamma camera was positioned for a dorsal image.

The initial frame shows activity in the heart and lung with activity in the aorta appearing in the next two frames. Beginning in the third frame there is increase activity in the kidneys (first pass), then a brief decline in activity. This is followed by a prolonged increase in renal activity ending in a decline in activity indicating renal clearance.

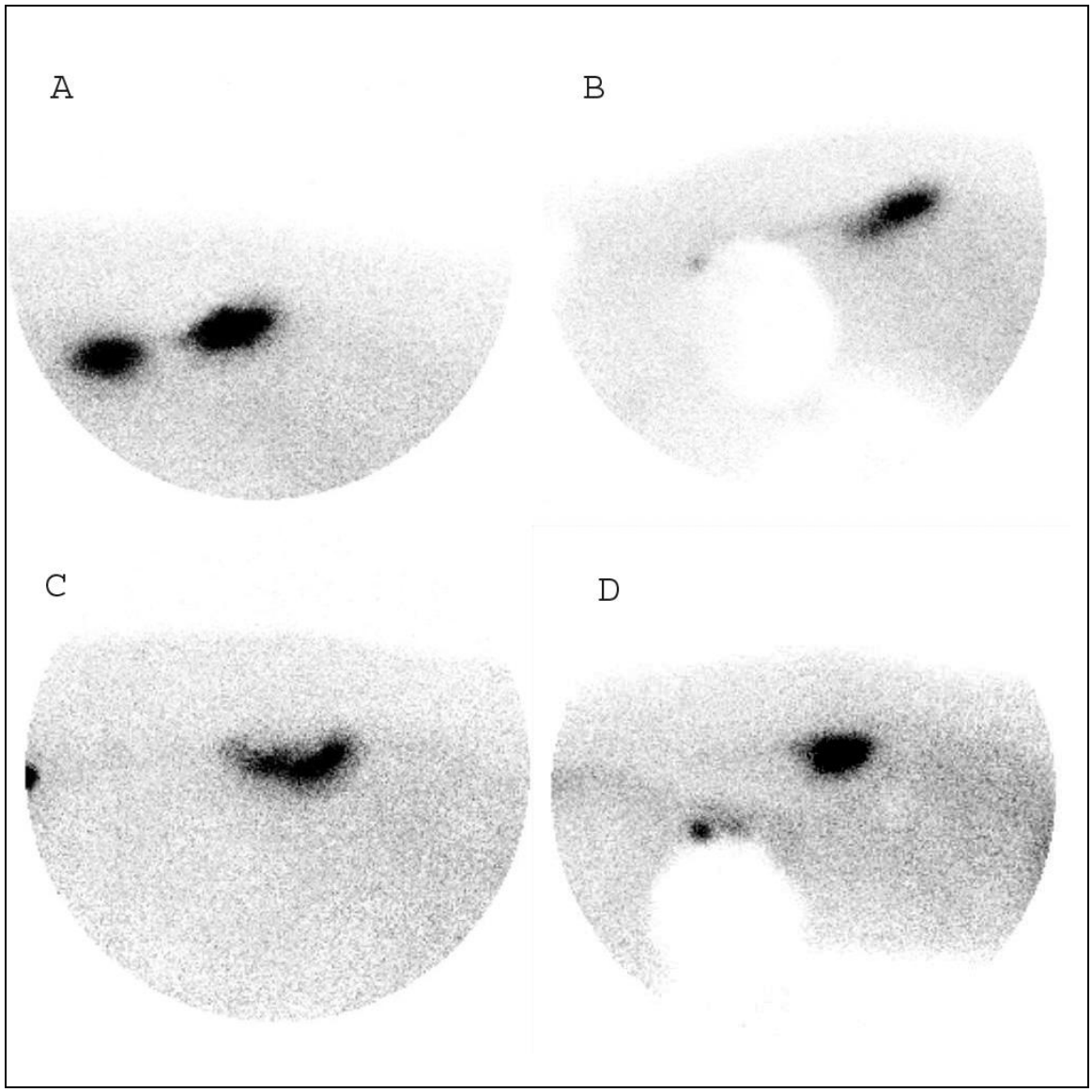

Figure 2. Variations in the appearance of the left and right kidneys in the right lateral image.

In each image, dorsal is at the top and cranial is to the right. Dog is positioned in left lateral recumbency with the gamma camera positioned for a right lateral image. In images A and C the left kidney is to the left and the right kidney is to the right.

(A) Kidneys appear distinct. (B) Kidneys overlapped, producing a single elongated image. (C) Kidneys overlapped but discernable. (D) Kidneys completely superimposed. The circular white areas in images B and D are the result of lead shielding used to block the activity in the urinary bladder.

Post Acquisition Processing and Calculations

Mirage® Processing Application software was used for post processing of the three dynamic studies.[19] To obtain pre counts, a ROI was drawn around the syringe activity in the summed image of the pre-injection dynamic acquisition. Using a summed image of the dynamic GFR study, individual regions of interest (ROIs) were drawn for each kidney, as well as background regions cranial and caudal to each kidney (Fig 3). Post counts were obtained from the summed image of the post injection dynamic study with a ROI drawn around the catheter and residual syringe, taking care not to include background activity from adjacent unshielded areas of the patient. The process of drawing ROIs for pre counts, GFR, and post counts was repeated 2 more times, so that 3 measures were made of each. Counts per ROI and number of pixels per ROI were exported from the Mirage® software to a spreadsheet.[20]

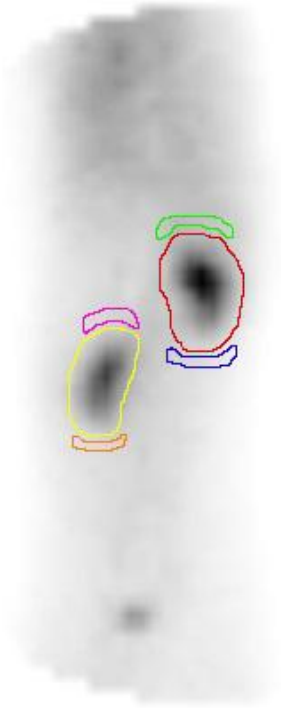

Figure 3. Summed image of the dynamic GFR study with regions of interest.

In each image cranial is at the top, the dog's right is to the right, and the dog's left is to the left. The dog was positioned in left lateral recumbency, and the gamma camera was positioned for a dorsal image. Red line = right kidney ROI; blue and green lines = background ROIs for right kidney; yellow line = left kidney ROI; magenta and orange lines = background ROIs for left kidney. The area of activity at the bottom of the image represents radiopharmaceutical in the urinary bladder.

The ROI results were used for GFR calculation as follows: [10]

1. The net kidney counts for each kidney were determined using the summed counts from 60 to 180 seconds according to the formula:

$$\text{Net Kidney cts} = (\text{Kidney cts}) - \left[\left\{ \frac{\text{Bkd cts}}{\text{\# Bkd pixels}} \right\} \times (\text{\# Kidney pixels}) \right]$$

where cts = counts and Bkd = background.

2. The percentage injected dose in each kidney was determined according to the formula:

$$\text{Injected dose (Kidney)} = \frac{\text{(net kidney cts)}}{\text{(pre-dose cts in cpm)} - \text{(post-dose cts in cpm)}} \times 100$$

where cts = counts and cpm = counts per minute.

3. In each dog, calculations of total (global) GFR were performed with and without correcting kidney counts for attenuation.
    a. Non depth-corrected GFR: Kraweic's 1986 article[3] showed a slight improvement with depth correction; therefore, the article only contains the depth-corrected linear regression formula. The following unpublished linear regression formula was used by Kraweic to predict non depth-corrected GFR.[21]

    GFR = 0.6355 x (% dose uptake right kidney + % dose uptake left kidney) – 0.351.

    b. Depth-corrected GFR:
        i. To correct for attenuation, the renal depths were calculated using measurements obtained from the lateral image by means of the following formula:

$$\text{renal depth} = \frac{\text{image depth x gamma camera field of view diameter}}{\text{image diameter}}$$

ii. Verification of the diameter of the gamma camera was made using visualization on the persistence scope of a radiation point source with the LEAP collimator in place.

iii. The renal depth in centimeters was used to depth correct the percent injected dose (%ID) for each kidney in the linear attenuation equation:

$$\%ID = \%ID \text{ non depth-corrected} \times e^{(O.153 \times \text{kidney depth in cm})}$$

iv. Depth-corrected total (global) GFR was calculated using an accepted linear regression formula[3]:

GFR = 0.194 x (% dose uptake right kidney + % dose uptake left kidney) – 0.37.

4. Individual kidney GFR for both depth-corrected and non depth-corrected methods was calculated according to the formula:

$$\text{Individual kidney GFR} = \frac{\%ID \text{ for the individual kidney}}{\%ID \text{ right kidney} + \%ID \text{ left kidney}} \times \text{global GFR } \%ID$$

Plasma Clearance Protocol

Plasma clearance of ^{99m}Tc-DTPA was performed simultaneously with quantitative renal scintigraphy using the following protocol:

1. Pre injection activity: Prior to injection of the ^{99m}Tc-DTPA, the activity in the dose syringe was measured with a dose calibrator[22].
2. Post injection activity: After the scintigraphy protocol was completed and the post injection static image was obtained, the IV catheter was carefully removed. The activity remaining in the syringe and catheter was measured with the dose calibrator.
3. The actual activity injected was calculated by subtracting the post injection activity from the pre injection activity. Pre-injection and post injection activity was obtained in a dose calibrator three times each, and an average reading was obtained.
4. Heparinized blood samples were obtained at 20 and 180 minutes post injection from a jugular vein.
5. The samples were centrifuged. A single-channel pipette with a reported accuracy of 0.8% and precision of 0.2%[23] was used to transfer 0.5 ml of plasma from each sample into a counting tube.
6. The radioactivity in each tube was determined using a multichannel sodium iodide well counter[24] with the photopeak region of interest centered over the 140 keV photopeak of ^{99m}Tc. The number of counts per minute (cpm) was measured sequentially three times for all samples, and an average value was obtained for each sample.
7. Well counter efficiency was determined by measuring the activity of a sample of ^{99m}Tc in the dose calibrator. The sample was allowed to decay until the expected counts per minute was less than 1.8 million, the highest cpm the well detector

could count without dead-time losses.[25] Efficiency was calculated by dividing actual cpm by theoretical cpm.

8. Plasma clearance was calculated from the 20 and 180 minute plasma sample values using the following formulas based on a monoexponential model[8]:

$$\alpha = \ln(C_1/C_2)/(T_2-T_1)$$

$$\ln(a) = [T_2\ln(C_1) - T_1\ln(C_2)]/(T_2 - T_1)$$

$$Cl_p = D(\alpha)/a$$

where

"a" is derived from math formula in line 2 above

C_1 = plasma concentration of tracer at time 1 (T_1)

C_2 = plasma concentration of tracer at time 2 (T_2)

Cl_p = plasma clearance

D = dose injected

Computed Tomography Protocol

Twenty-four to 72 hours after the scintigraphic procedure, computed tomography was performed. Each dog was sedated using the following protocol: premedication with glycopyrrolate 0.011 mg/kg subcutaneously followed 15 minutes later by a single intramuscular injection containing 1000mcg/M^2 medetomidine mixed with 0.2mg/kg butorphanol. If necessary after the procedure the medetomidine was reversed with atipamezole intramuscularly using a volume equal to that of the medetomidine. To obtain CT Scan 1, each dog was placed in left lateral recumbency on a wooden board on the CT table attempting to mimic the dog's position on the gamma camera table. The

wooden board was used to create a flat surface like the scintigraphic imaging table. The spine and legs were not flexed or extended. A scout image was obtained to aid in localization of the hilus of each kidney. Transverse CT slices through the hilus of each kidney were obtained. To obtain CT Scan 2 the dog was then manually lifted, placed back on the table in the same position, and scanned a second time. The repeat scan was performed to determine the effect of a subtle change in position on measurement of renal depth.

For both CT Scan 1 and CT Scan 2, the center of each kidney at the hilus was determined using two different methods. In the first method, the center of the kidney was considered to be the tip of the renal crest (Figure 4). In the second method, the center of the kidney was considered to be the intersection of two lines defining the longest and shortest dimensions of the kidney (Figure 5). Using each method, two separate measurements were obtained for each kidney: 1) the distance from the center of the kidney at the level of the hilus to the skin surface, and 2) the distance from the center of the kidney at the level of the hilus to a line perpendicular to the dorsal plane touching the skin at dorsal midline. The line perpendicular to the dorsal plane touching the skin at dorsal midline is referred to as the "dorsum".

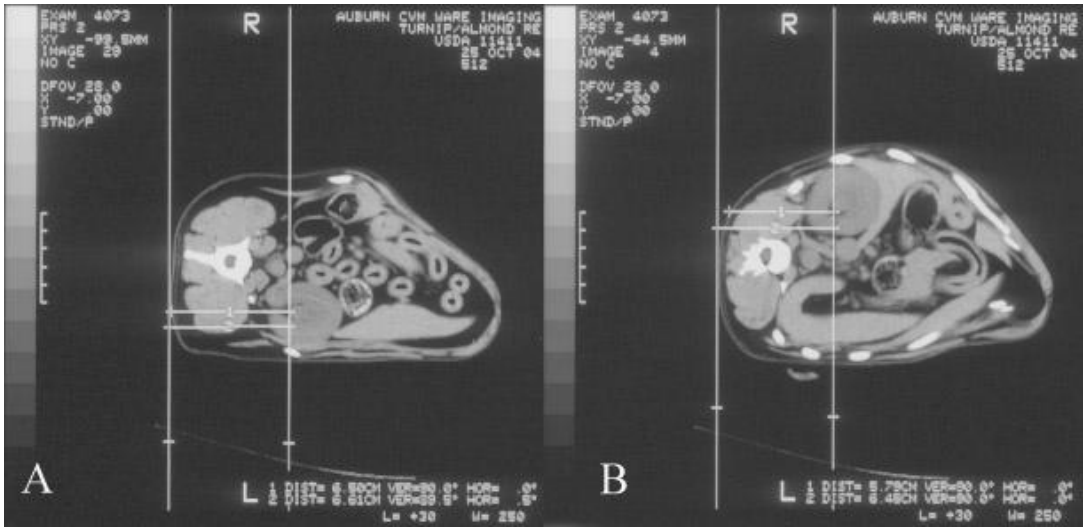

Figure 4. Computed tomography measurement method one.

Axial computed tomography images with patient in left lateral recumbency showing measurement of distances from renal crest to skin directly dorsal to center of kidney and to a line perpendicular to the skin at dorsal midline for (A) left kidney and (B) right kidney.

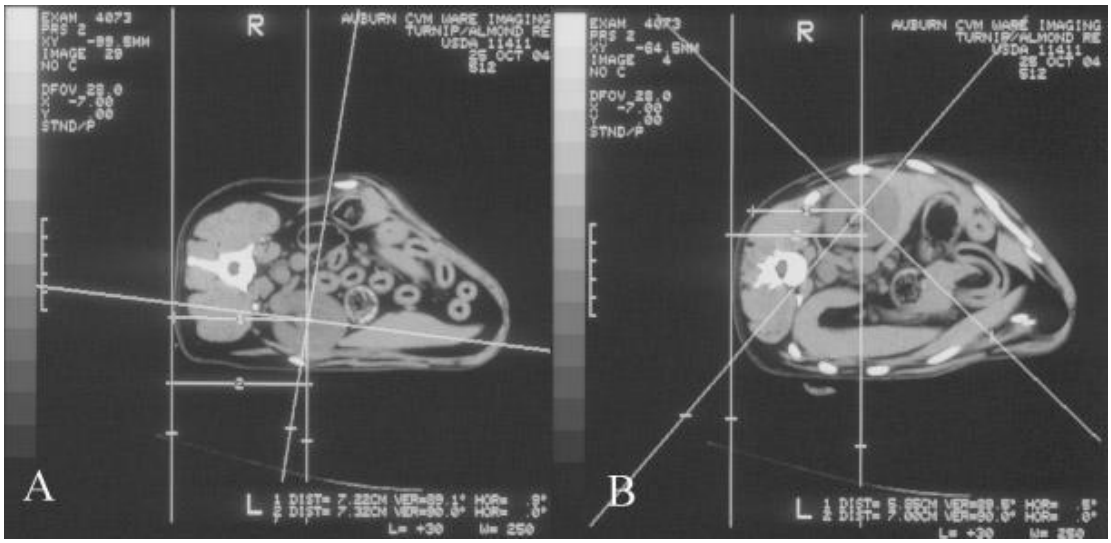

Figure 5. Computed tomography measurement method two.

Axial computed tomography images with patient in left lateral recumbency showing measurements of distances from center of kidney to skin directly dorsal to center of kidney and to a line perpendicular to the skin at dorsal midline for (A) left kidney and (B) right kidney.

IV. RESULTS

Glomerular filtration rate measurements

Depth-corrected (DC), non depth-corrected (nDC), and plasma clearance (PC) values are presented graphically in Figure 6 and in tabular form in the appendix. Depth-corrected (DC), non depth-corrected (nDC), and plasma clearance (PC) methods of GFR determination were compared using the mixed model for repeated measures of analysis of variance (ANOVA). This analysis was performed for all dogs together, for dogs less than 13.6 kg (small dogs), and for dogs greater than 22.7 kg (large dogs). A p value less than 0.05 was considered to be significant.

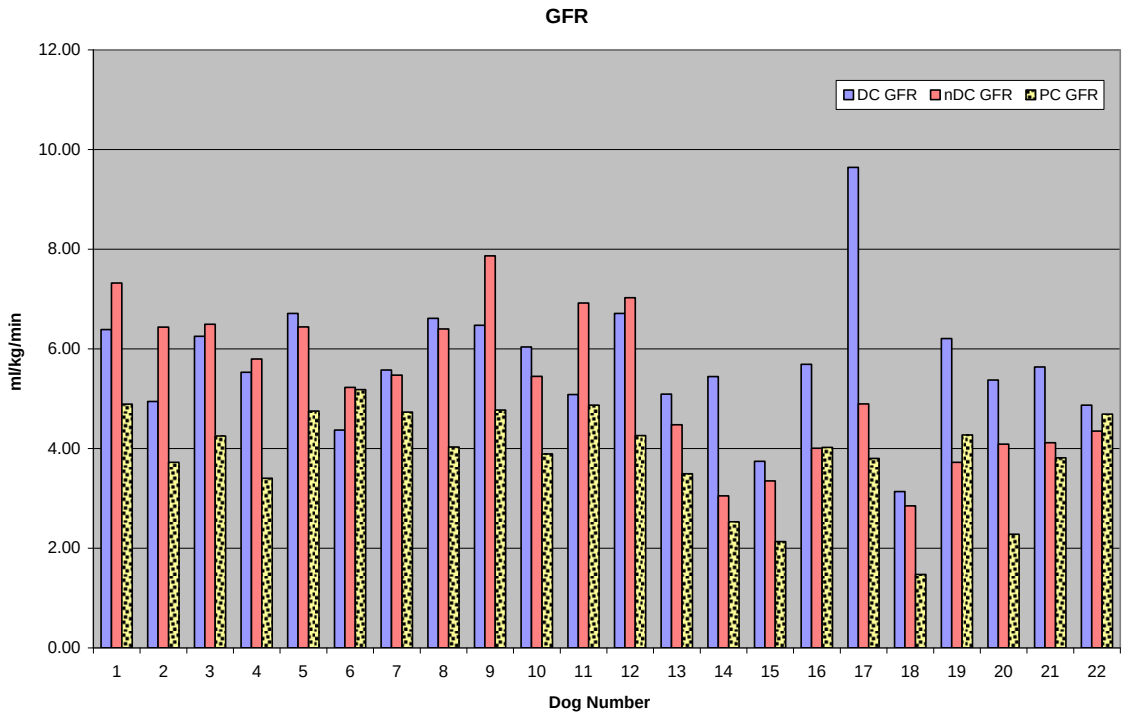

Figure 6. Depth-Corrected (DC), non Depth-Corrected (nDC), and Plasma Clearance (PC) values for GFR in all dogs.

Dogs 1-12 are small dogs, and dogs 13-22 are large dogs.

For all dogs the least square means estimate for DC, nDC, and PC were 5.6, 5.2, and 3.8 ml/kg/min respectively. DC and nDC were not different ($p = 0.3926$). PC was different from DC ($p < 0.0001$). PC was different from nDC ($p = 0.0003$).

For dogs less than 13.6 kg (30 lbs) the least square means estimate for DC, nDC, and PC were 5.9, 6.4, and 4.4 ml/kg/min respectively. DC and nDC were not different ($p = 0.2332$). PC was different from DC ($p < 0.0001$). PC was different from nDC ($p < 0.0001$).

For dogs greater than 22.7 kg (50 lbs) the least square means estimate for DC, nDC, and PC were 5.5, 3.9, and 3.2 ml/kg/min respectively. DC and nDC were different ($p = 0.0263$). PC was different from DC ($p = 0.0016$). PC and nDC were not different ($p = 0.5179$).

Pearson correlation coefficients were also calculated to determine the degree of correlation between depth-corrected GFR determination and plasma clearance and between non depth-corrected GFR determination and plasma clearance. These coefficients were determined for all dogs, for dogs smaller than 13.6 kg (30 lbs), and for dogs greater than 22.7 kg (50 lbs). When evaluating all dogs PC correlated better with nDC (0.682) than with DC (0.398). When evaluating dogs less than 13.6 kg (30 lbs) PC correlated poorly with both DC (-0.091) and nDC (0.204). In dogs greater than 22.7 kg (50 lbs) there was modest correlation between PC and DC (0.514) and between PC and nDC (0.685). Correlation between DC and nDC was modest for all dogs, small dogs, and large dogs (0.408, 0.517, and 0.693 respectively).

Renal Depth Measurements

Renal depth measurement using DTPA scintigraphy was compared to computed tomography measurements using Pearson correlation coefficients. These coefficients were calculated comparing computed tomography derived measurements of renal crest to dorsum, renal crest to skin, renal center to dorsum, renal center to skin, to renal scintigraphy depth measurement for both left and right kidneys. Pearson correlation coefficients were also calculated for all of these measurements for CT Scan 2. The correlation coefficients for CT versus scintigraphic renal depth measurements arranged

by right and left kidneys for all dogs, small dogs, and large dogs are presented in Table 1. The complete tables of Pearson correlation coefficients comparing all CT renal depth measurements and CT versus scintigraphic renal depth measurements are presented in Appendices D through I.

		Scintigraphic Measurements					
		All dogs		Small dogs		Large dogs	
		R	L	R	L	R	L
CT Scan 1	crest-dorsum	0.947	0.828	0.672	0.354	0.968	0.566
	crest-skin	0.921	0.843	0.611	0.469	0.905	0.610
	center-dorsum	0.940	0.738	0.636	0.022	0.959	0.473
	center-skin	0.909	0.753	0.641	0.145	0.892	0.530
CT Scan 2	crest-dorsum	0.932	0.838	0.597	0.323	0.961	0.602
	crest-skin	0.917	0.840	0.528	0.379	0.926	0.614
	center-dorsum	0.936	0.772	0.587	0.186	0.960	0.484
	center-skin	0.902	0.772	0.500	0.254	0.909	0.524

Table 1. Pearson correlation coefficients for CT versus scintigraphic renal depth measurements

Correlations between CT and nuclear scintigraphy depth measurements for the right kidney in all dogs were strong regardless of method of CT measurement. For the left kidney in all dogs, correlations were strong, with measurements from the renal crest correlating slightly better than those from the center of the kidney. In small dogs correlation between CT and nuclear scintigraphy depth measurements were modest for all methods of CT measurement of the right kidney and were poor for all methods of measurement of the left kidney. In large dogs correlation between CT and nuclear scintigraphy depth measurement were strong for all methods of CT measurement of the right kidney and modest to weak for all methods of measurement of the left kidney.

Using the static right lateral scintigraphic images, small dogs had a mean right kidney depth of 6.84 cm (range 5.24 to 8.03 cm) and a mean left kidney depth of 7.69 cm

(range 6.16 to 9.03 cm), giving a mean renal depth of 7.26 cm. In large dogs, the mean right kidney depth was 9.33 cm (range 7.80 to 12.32 cm) and the mean left kidney depth was 10.18 cm (range 8.60 to 11.70), resulting in a mean renal depth of 9.75 cm.

Subjective Assessment of Lateral Scintigraphic Images

In the subjective assessment of the separation of left and right kidneys on the lateral scintigraphic image of the abdomen distinct kidneys (D) and kidneys overlapped but discernible (OD) were considered readable for the sake of analysis. Kidneys that were overlapped producing a single elongated image (OE) and kidneys completely superimposed (CS) were considered unreadable for the sake of analysis. Of the 12 dogs weighing less than 13.6 kg (30 lbs), five were considered readable (dogs 3, 6, 7, 8, and 10) and six were considered unreadable (dogs 1, 2, 4, 5, 9, 11, and 12). Of the 10 dogs weighing greater than 22.7 kg (50 lbs), seven were considered readable (dogs 14, 15, 16, 17, 19, 20, and 21) and three were considered unreadable (dogs 13, 18, and 22). Fisher's exact test was performed to evaluate readability. There was no significant difference in readability between small dogs and large dogs ($p = 0.2305$).

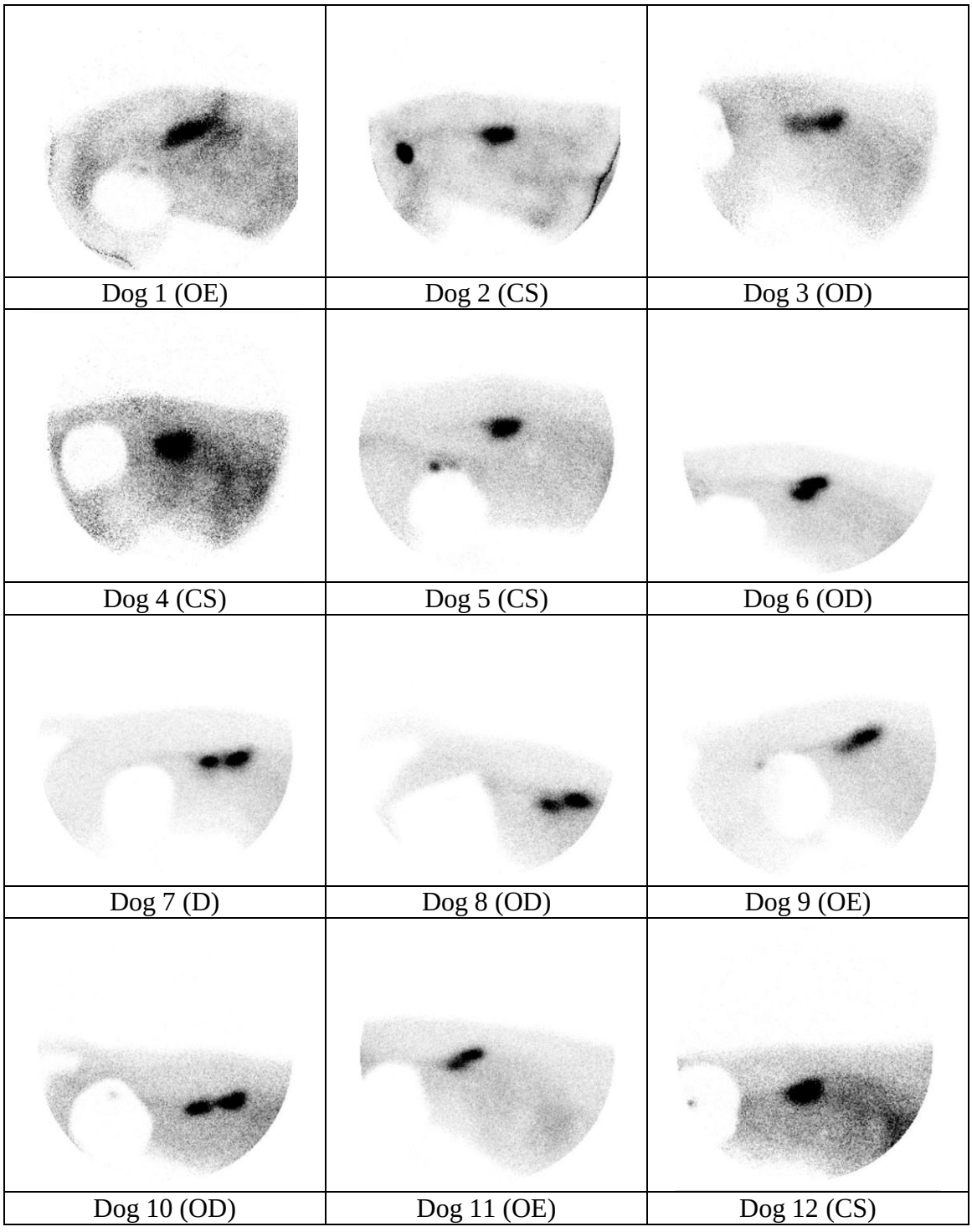

Figure 7. Lateral static scintigraphic image of all small dogs with readability assessment for each dog in parenthesis. Left and right kidneys distinct (D), kidneys overlapped producing a single elongated image (OE), kidneys overlapped but discernable (OD), or kidneys completely superimposed (CS).

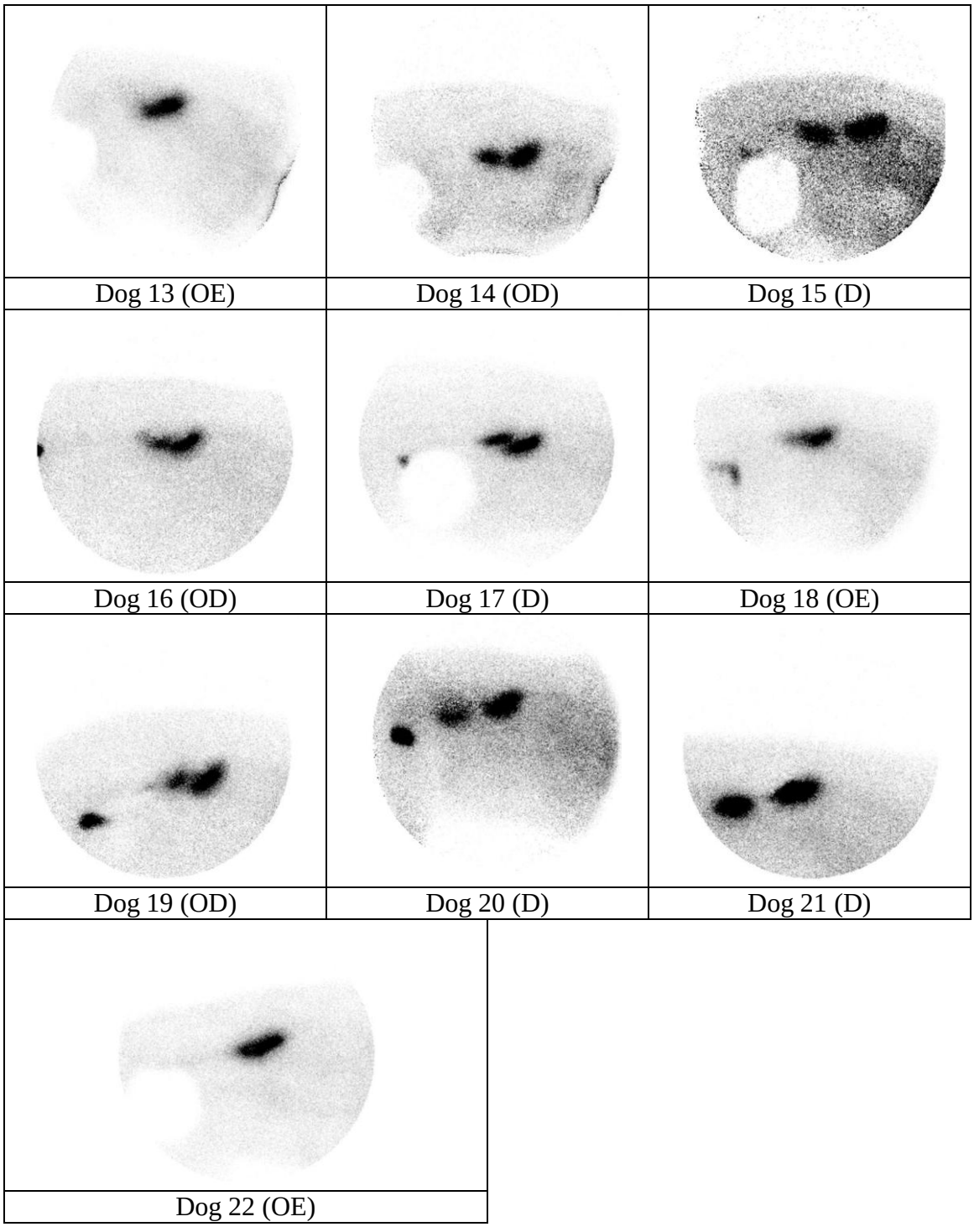

Figure 8. Lateral static scintigraphic image of all large dogs with readability assessment for each dog in parenthesis. Left and right kidneys distinct (D), kidneys overlapped producing a single elongated image (OE), kidneys overlapped but discernable (OD), or kidneys completely superimposed (CS).

V. DISCUSSION

Determination of glomerular filtration rate is beneficial in the early detection of renal failure in the dog.[4] Because GFR declines prior to alterations in urine specific gravity, BUN, and serum creatinine, detection of a decrease in GFR may allow therapeutic intervention early in the course of the disease.[4] Nuclear scintigraphy provides a relatively quick and easy method of determining GFR, unlike the classical methods of inulin or creatinine clearance which are cumbersome and difficult to perform.[1,3] An additional advantage of determination of GFR by nuclear scintigraphy is the ability to quantify individual kidney function.[6] Although scintigraphic determination of GFR is a relatively quick procedure, decreasing the time required to perform the study or decreasing post-imaging analysis time is desirable in a clinical setting. Current recommendations require obtaining a static lateral image of the abdomen to determine kidney depth, a value required for correcting for attenuation of radiation in the soft tissues between the kidney and the gamma camera.[3] The accuracy of kidney depth measurement is questionable because nuclear scintigraphy has poor spatial resolution. The current study was designed to determine if kidney depth measurement by scintigraphy is accurate and if this additional step is necessary.

Depth-corrected and Non Depth-Corrected GFR

The hypothesis, scintigraphic measurements of global GFR in dogs using the radiopharmaceutical ^{99m}Tc-DTPA are not statistically different whether they are depth-corrected or non depth-corrected, was confirmed. Depth-corrected and non depth-corrected values for GFR were not significantly different when evaluating all dogs. Likewise depth-corrected and non depth-corrected values for GFR were not significantly different in dogs weighing less than 13.6 kg (30 lbs). For dogs weighing greater than 22.7 kg (50 lbs), however, depth-corrected and non depth-corrected values for GFR were significantly different. This difference between large and small dogs is not surprising considering that mean kidney depth is greater in large dogs than in small dogs. Since the linear attenuation equation describes the exponential decrease in radiation as it traverses tissue, the formula for depth correction exponentially augments the counts coming from the kidneys. The deeper the kidneys are within the patient the greater the mathematical boost to GFR. This alone does not explain the difference between nDC and DC values; the linear regression formulas for nDC and DC are also different. Table 2 was prepared to demonstrate the combined effect of linear attenuation correction and the separate linear regression formulas for nDC and DC. The percentage injected doses and kidney depths used in this table reflect the ranges of those encountered in the dogs of this study.

%ID	6 cm		7 cm		8 cm		9 cm		10 cm		12 cm	
	nDC	DC	nDC	DC	nDC	DC	nDC	DC	nDC	DC	nDC	DC
1.5	0.6	0.4	0.6	0.5	0.6	0.6	0.6	0.8	0.6	1.0	0.6	1.5
2.0	0.9	0.6	0.9	0.8	0.9	0.9	0.9	1.2	0.9	1.4	0.9	2.1
2.5	1.2	0.8	1.2	1.0	1.2	1.3	1.2	1.6	1.2	1.9	1.2	2.7
3.0	1.6	1.1	1.6	1.3	1.6	1.6	1.6	1.9	1.6	2.3	1.6	3.3
3.5	1.9	1.3	1.9	1.6	1.9	1.9	1.9	2.3	1.9	2.8	1.9	3.9
4.0	2.2	1.6	2.2	1.9	2.2	2.3	2.2	2.7	2.2	3.2	2.2	4.5
4.5	2.5	1.8	2.5	2.2	2.5	2.6	2.5	3.1	2.5	3.7	2.5	5.1
5.0	2.8	2.1	2.8	2.5	2.8	2.9	2.8	3.5	2.8	4.1	2.8	5.7
5.5	3.1	2.3	3.1	2.7	3.1	3.3	3.1	3.9	3.1	4.6	3.1	6.3
6.0	3.5	2.5	3.5	3.0	3.5	3.6	3.5	4.2	3.5	5.0	3.5	6.9
6.5	3.8	2.8	3.8	3.3	3.8	3.9	3.8	4.6	3.8	5.5	3.8	7.5
7.0	4.1	3.0	4.1	3.6	4.1	4.2	4.1	5.0	4.1	5.9	4.1	8.1
7.5	4.4	3.3	4.4	3.9	4.4	4.6	4.4	5.4	4.4	6.3	4.4	8.8

Table 2: DC and nDC results for theoretical percent injected dose and various renal depths in the ranges encountered in the current study.

From this table it can be seen that regardless of percentage injected dose the obtained values for nDC and DC are in greater agreement if renal depths are between 7 and 9 cm. This agrees with the results of the current study. The small dogs in this study had a mean renal depth of 7.26 cm and the large dogs had a mean renal depth of 9.75 cm. The reason for the observation that the small dogs in this study with mean renal depth less than 7 cm (dogs 1, 2, 6, 9, and 11) showed the greatest difference in DC and nDC can also be seen in Table 2.

An obvious shortcoming of the current study is the lack of concurrent inulin clearance on these patients as the gold standard of GFR. In this study, nDC and DC were more disparate for dogs greater than 22.7 kg (50 lbs) than for dogs less than 13.6 kg (30 lbs). This finding suggests that depth correction is not necessary for dogs weighing less than 13.6 kg (30 lbs). Additional studies comparing nDC and DC to inulin clearance in

large dogs are necessary before the current recommendation of depth correcting in these patients is abandoned.

It should also be noted that this study only included healthy dogs. If nDC and DC are as disparate in large dogs with compromised renal function as they were in the normal large dogs in this study, a clinical conundrum could exist when attempting to determine if a diseased kidney could be removed. Clearly more research is needed to determine 1) whether nDC or DC is a more accurate predictor of GFR when compared with inulin clearance in large dogs and 2) if a new non depth-corrected linear regression formula should be derived solely for use in large dogs.

Another question raised by this study is the relationship between body weight and renal depth. Although dogs were divided into those weighing more than 30 pounds and those weighing less than 13.6 kg (30 lbs), there were no dogs weighing between 13.6 kg (30 lbs) and 22.7 kg (50 lbs). It is not certain what range of renal depths dogs in the 13.5-22.7 kg weight group would have. The majority of the smaller dogs in this study were Beagle mix and the majority of the larger dogs were hounds. It is not known if breed or body composition has a more profound effect on renal depth than absolute body weight.

It was hypothesized that depth-corrected and non depth-corrected GFR measurements would correlate well with DTPA plasma clearance. However, there was only modest correlation between PC and nDC in all dogs and between PC and both scintigraphy methods in large dogs. The remaining correlations were poor. The reason for these findings is unclear, but may be related to problems with the plasma clearance methodology as discussed in the next section. In all but 4 dogs, plasma clearance values were lower than nDC and DC values. If the plasma clearance values are accurate, then

depth correction may be introducing error into scintigraphic GFR measurement. If scintigraphic measurement is more accurate than plasma clearance, then depth correction in large dogs would provide more accurate results.

Plasma Clearance

Plasma clearance determination of GFR was shown by Barthez *et al*. to have a high degree of correlation with inulin clearance.[6] Therefore, plasma clearance was performed concurrent with nuclear scintigraphy in this study with the hopes of providing a close approximation to the gold standard of inulin clearance. According to Barthez *et al*. the correlation coefficient between inulin clearance and plasma clearance was 0.98, and the correlation coefficient between inulin clearance and nuclear scintigraphy was 0.86.[6] Based on that information, it was anticipated the plasma clearance values would not vary significantly from depth-corrected and non depth-corrected nuclear scintigraphy derived values. However, ANOVA revealed that DC and nDC were significantly different from plasma clearance when considering all dogs together and when considering small dogs alone. In large dogs plasma clearance was also found to significantly differ from DC. Only when comparing plasma clearance to non depth-corrected GFR in large dogs were the values not significantly different. The cause of general lack of agreement between plasma clearance and scintigraphy is unclear. Inaccuracies in the plasma clearance values could have occurred due to error in design, calculation, or execution of the procedure.

The design of the two-sample plasma clearance methodology was based upon that reported by Barthez *et al*.[8] in whose study subjects were administered 50-300 μCi of

^{99m}Tc DTPA. In the current study, dogs were injected with 1.5 to 2.8 mCi of ^{99m}Tc DTPA. In a previous study Barthez *et al*.[6] administered 5-10 mCi of ^{99m}Tc DTPA to subjects that were imaged by nuclear scintigraphy and concurrently evaluated with an eight-sample plasma clearance method. Despite using the larger dose, it was found that the eight-sample plasma clearance method correlated better with inulin than did percent DTPA uptake via nuclear scintigraphy.

Although these studies indicate that the range of activities of ^{99m}Tc DTPA used in the current study should produce acceptable results, one other factor must be considered. There is a finite period of time that must elapse between ionizations for a well detector to register the event; this period of time is called the dead time of the detector.[10] If the resulting activity in the plasma is too high, dead-time losses in the well detector could result in erroneous results. The well detector used in this study has dead-time losses if the count rate exceeds 1.8 million cpm. The dog that received the greatest dose in the current study had a 20 minute count rate of approximately 223,000 cpm, indicating that the activity used in the current study produced plasma counts well below the count rate that would produce dead-time losses in the well detector.

The formulas and spreadsheets used to calculate GFR for both methods were carefully evaluated, and no errors were found. One potential source of error in the plasma clearance methodology is in the calculation of well detector efficiency. Any change in well detector efficiency would have a profound effect on values obtained for plasma clearance. This procedure was performed several times during the course of this study. To reduce the likelihood of errors that can occur with dilution techniques for determining efficiency, a sample of ^{99m}Tc was placed in a dose calibrator to determine initial activity.

It was then allowed to decay until the expected counts per minute were less than 1.8 million. Efficiency was calculated by dividing actual cpm by theoretical cpm. Values obtained for well detector counting efficiency ranged from 0.68 to 0.71. These values were lower than anticipated. The technical support department for the manufacturer reported that counter efficiency varies with the detector manufacturer, and data on expected efficiency is not available.[25] Inherent in this method is the assumption that the reading obtained by the dose calibrator is correct. The dose calibrator had been recently calibrated at the local nuclear pharmacy.[26] Linearity was performed on the dose calibrator and was found to be within acceptable limits. Although great care was taken to collect samples on time and to carefully measure activity of the plasma samples, it is possible that human error occurred. A study on two-sample plasma clearance comparing low doses versus high doses may be useful to determine the accuracy of the procedure with the higher doses used for scintigraphy.

Renal Depth Measurements

Because nuclear scintigraphy has poor spatial resolution it was hypothesized that there would be poor correlation between renal depths measured on the static lateral scintigraphic image and those measured by computed tomography. There are several factors that interfere with the ability to obtain accurate measurement of renal depths from scintigraphic images. The exact margins of structures are often difficult to discern in scintigraphic images due to scattering and divergence of gamma rays. The farther an object is from the gamma camera the greater the likelihood of distortion due to gamma ray divergence. The lateral scintigraphic images of the kidneys in the study were

obtained with the patient in left lateral recumbency. Therefore the left kidney was farther from the gamma camera than the right kidney. As a result the left kidney frequently had "fuzzier" margins than the right kidney. This could have resulted in decreased accuracy when assessing left kidney depth.

Another factor affecting the ability to discern renal depth was the degree of superimposition of the left and right kidney in the lateral scintigraphic image. Although both kidneys are held in position in the retroperitoneal space, they are not rigidly fixed to the dorsal body wall and can move during respiration; the right kidney is more firmly attached than the left.[27] This would explain why the left kidney margin frequently appeared less distinct because the left kidney would be more likely than the right kidney to move with respiration during image acquisition. The degree of superimposition also may be related to individual body conformation. In images where the kidneys were completely superimposed it was assumed that both kidneys were of equal depth. If the kidneys were not in the exact same dorsal plane or if one kidney was smaller than the other this may have been an erroneous assumption. Partial overlapping of kidneys in some dogs created a degree of uncertainty as to exact renal depth. Although scintigraphic image readability was not statistically different between small and large dogs, 7 out of the 12 small dogs had kidneys that were difficult to delineate. The failure to achieve statistically significant difference between the groups may be due to small sample size.

With computed tomography measurements there was no difficulty determining the location of each kidney and the dorsal margin of the patient. Multiple measurements were made from the CT images to explore possible differences in depth measurements. Two different methods for estimating the center of each kidney were employed. The first

method considered the center of the kidney to be at the tip of the renal crest. In nuclear scintigraphy ^{99m}Tc DTPA is rapidly filtered by the glomerulus and quickly concentrates in the renal pelvis. This results in high numbers of counts in the region near the renal crest which could skew the observer's perception of kidney depth. The second method was designed to approximate the geometric center of the kidney at the level of the hilus. The high counts coming from the renal crest region should not skew the observer's perception of kidney depth using this method. Measurements were made from the renal crest and geometric center to both the skin and to the dorsal plane touching the dorsal midline. The dorsal plane touching dorsal midline was chosen because it most closely mirrors the dorsal edge of the patient used in the static lateral nuclear scintigraphic image. Measurements from the kidneys to the skin surface were made to account for the difference in the variable shape of the dorsum of patients and because it represents the true depth of attenuating tissue. However, there was strong correlation among all the different methods of assessing renal depths by computed tomography.

The sedation protocol used for the nuclear scintigraphy study was deemed to provide inadequate duration of sedation for inclusion of the CT study on the same day, necessitating the two studies being performed on sequential days. There was concern that laying an animal in left lateral recumbency at two different points in time would result in poor repeatability of renal depth measurements, especially in light of the greater mobility of the left kidney. For this reason the initial CT scan was followed by repeat positioning of the patient on the table and repeating the CT scan. A high degree of correlation between the two CT scans for all dogs, small dogs, and large dogs, indicate that there is minimal impact of repeat positioning of the dogs on the table.

Of interest is the result of the correlations between CT and scintigraphy renal depths. It is not surprising that correlations were good between right kidney depth measurements for all dogs in both imaging modalities. Right kidney correlations were strong for the large dogs and moderate for the small dogs. The margins of the right kidney are much easier to discern in nuclear scintigraphy, presumably making measurement of depth more reliable. The reason for the better correlations seen in the right kidneys of the large dogs versus the right kidneys of the small dogs is unclear. Correlations for left kidney depth measurements in both imaging modalities were modest in large dogs and poor in small dogs. Since the left kidney has less distinct margins in scintigraphy than the right kidney decreased accuracy in measuring the left kidney depth was anticipated. Because the left kidney scintigraphy depth measurement in small dogs correlated so poorly with CT measurements, using this value to depth-correct in small dog is at best of dubious value.

Because there is poor-to-modest correlation between CT and scintigraphic depth measurements for the left kidney of large dogs, depth correction could introduce error into determination of left kidney GFR in dogs weighting greater than 22.7 kg. Repeating Kraweic's study[3] with small and large dogs would be necessary to confirm these findings. It is possible that with further study it may be found that depth correction is not necessary, but that separate linear regression formulas for small and large dogs will improve accuracy of GFR measurements by scintigraphy.

VI. CONCLUSIONS

This study was designed to determine if depth correction is necessary in ^{99m}Tc-DTPA scintigraphy studies for determination of GFR for small and large dogs, and to determine if the current method used for depth correction accurately assesses renal depth. ^{99m}Tc-DTPA plasma clearance also was performed in hopes of providing a reference with which to compare the depth-corrected and non depth-corrected GFR values. The hypothesis that global GFR measurements using ^{99m}Tc-DTPA are not statistically different whether they are depth-corrected or non depth-corrected was found to be true when considering all dogs in the study; however, an important difference was discovered between dogs weighing less than 13.6 kg (30 lbs) and dogs weighing greater than 22.7 kg (50 lbs). Depth-corrected and non depth-corrected values were found to be similar in small dogs but significantly different in large dogs. There were no dogs between 13.6 and 22.7 kg in the study. Without the gold standard of inulin clearance, it is uncertain if depth correction can be abandoned in large dogs. Further research is needed to see if this difference between small and large dogs holds true in reference to the gold standard of inulin clearance.

Contrary to hypothesis neither depth-corrected nor non depth-corrected GFR measurements correlated well with plasma clearance determination of GFR. The reason

for this lack of correlation was not determined and points to the need to use a gold standard such as inulin clearance.

Renal depth measurement using DTPA scintigraphy was found to strongly correlate with computed tomography measurements only when comparing the depth of the right kidney of all dogs and the right kidney of large dogs. Renal depth measurement using DTPA scintigraphy was found to be unreliable when compared with CT for assessing left kidney depth in small dogs. This coupled with the finding that depth-corrected and non depth-corrected values are similar in small dogs makes obtaining a lateral scintigraphic image of questionable value in small dogs. In dogs greater than 22.7 kg (50 lbs) correlations between CT and scintigraphic depth measurements are modest and possibly introduce error into depth-corrected GFR values. It was also found that repeat positioning of dogs in lateral recumbency does not alter kidney depth measurements.

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APPENDICES

APPENDIX A

GFR and Nuclear Scintigraphy Depth Data

Dog #	Weight (lbs)	GFR (ml/kg/min)			Individual Kidney GFR (ml/kg/min)				Nuclear Scintigraphy			
									Appearance	Depth (cm)		
		DC	nDC	PC	R DC	R nDC	L DC	L nDC		R	L	Mean
1	27	6.4	7.3	4.9	2.8	3.2	3.6	4.1	OE	6.2	7.4	5.5
2	22	4.9	6.4	3.7	2.4	3.2	2.5	3.3	CS	6.2	6.2	4.3
3	29	6.3	6.5	4.3	3.6	3.7	2.7	2.8	OD	7.2	8.0	5.3
4	30	5.5	5.8	3.4	3.0	3.1	2.6	2.7	CS	7.5	7.5	5.0
5	27	6.7	6.4	4.8	3.4	3.3	3.3	3.1	CS	8.0	8.0	5.6
6	19	4.4	5.2	5.2	2.2	2.7	2.1	2.6	OD	5.8	7.5	4.8
7	22	5.6	5.5	4.7	3.1	3.0	2.5	2.5	D	7.7	8.1	5.3
8	21	6.6	6.4	4.0	3.5	3.4	3.1	3.0	OD	7.0	8.9	6.0
9	27	6.5	7.9	4.8	3.7	4.5	2.8	3.4	OE	6.0	7.3	5.0
10	22	6.0	5.4	3.9	3.2	2.9	2.8	2.6	OD	7.8	9.0	5.9
11	23	5.1	6.9	4.9	3.1	4.3	2.0	2.7	OE	5.2	6.8	4.4
12	23	6.7	7.0	4.3	3.8	4.0	2.9	3.0	CS	7.5	7.5	5.2
13	61	5.1	4.5	3.5	2.7	2.4	2.4	2.1	OE	8.2	8.9	5.7
14	64	5.4	3.1	2.5	3.1	1.7	2.3	1.3	OD	11.0	11.6	7.0
15	64	3.7	3.4	2.1	2.2	2.0	1.6	1.4	D	7.8	9.2	5.4
16	59	5.7	4.0	4.0	2.9	2.0	2.8	2.0	OD	9.9	10.0	6.4
17	56	9.6	4.9	3.8	4.2	2.1	5.5	2.8	D	12.3	11.7	8.6
18	50	3.1	2.9	1.5	1.2	1.1	2.0	1.8	OE	7.9	8.6	5.3
19	68	6.2	3.7	4.3	3.0	1.8	3.2	1.9	OD	10.9	10.9	7.0
20	65	5.4	4.1	2.3	3.6	2.7	1.8	1.4	D	8.7	10.7	6.2
21	58	5.6	4.1	3.8	3.5	2.6	2.1	1.5	D	8.5	11.3	6.7
22	62	4.9	4.3	4.7	2.5	2.2	2.4	2.2	OE	8.0	8.9	5.7

DC depth corrected
nDC non-depth corrected
PC plasma clearance

APPENDIX B

Computed Tomography Scan 1 Measurements

	Right Kidney 1st measurement				Left Kidney 1st measurement			
Dog #	crest to dorsum crd R1	crest to skin crs R1	center to dorsum ctd R1	center to skin cts R1	crest to dorsum crd L1	crest to skin crs L1	center to dorsum ctd L1	center to skin cts L1
1	6.2	5.7	6.6	5.7	7.0	6.9	7.2	6.9
2	5.8	5.2	6.6	5.3	6.5	6.0	7.3	6.7
3	6.3	5.6	6.6	5.6	8.6	8.4	9.6	9.3
4	6.8	6.1	7.0	6.0	8.7	8.3	9.1	8.7
5	6.3	6.0	6.4	5.9	7.1	6.9	7.8	7.5
6	5.2	4.6	5.6	4.8	6.3	6.1	5.9	5.7
7	6.5	5.9	6.9	5.7	6.7	6.6	7.5	7.4
8	5.6	5.0	6.2	5.2	7.6	7.6	6.9	6.9
9	5.9	5.4	6.1	5.2	7.2	7.1	7.9	7.8
10	7.5	6.2	8.1	6.8	7.6	7.6	7.5	7.5
11	6.1	5.7	6.2	5.6	7.0	7.0	7.4	7.4
12	6.9	6.2	7.3	6.3	7.3	7.3	7.1	7.1
13	8.3	7.7	8.5	7.8	10.5	10.2	11.2	10.9
14	9.6	7.9	10.0	8.0	12.1	11.7	12.3	12.2
15	7.0	6.2	7.3	6.1	10.4	9.8	10.7	9.9
16	9.0	8.3	9.6	9.0	9.0	8.9	8.4	8.2
17	11.0	10.3	11.5	10.9	12.0	12.0	12.2	12.2
18	7.7	7.1	7.7	6.6	9.2	8.8	9.4	8.9
19	10.2	9.3	11.1	10.2	11.9	11.7	12.7	12.5
20	7.9	6.9	7.9	6.4	10.5	10.4	10.5	10.4
21	7.6	7.1	7.6	6.8	8.8	8.5	9.1	8.5
22	7.8	6.9	7.8	6.6	9.0	8.5	9.2	8.5

APPENDIX C

Computed Tomography Scan 2 Measurements

	Right Kidney 2nd measurement				Left Kidney 2nd measurement			
Dog #	crest to dorsum crd R2	crest to skin crs R2	center to dorsum ctd R2	center to skin cts R2	crest to dorsum crd L2	crest to skin crs L2	center to dorsum ctd L2	center to skin cts L2
1	6.8	6.2	7.2	6.4	6.9	6.7	7.1	6.7
2	6.2	5.5	6.7	5.6	6.3	5.7	7.0	6.5
3	6.2	5.5	6.6	5.6	8.6	8.4	9.5	9.4
4	7.0	6.5	7.7	6.7	8.5	8.1	9.0	8.5
5	6.6	6.2	6.8	6.0	6.6	6.4	7.2	7.0
6	5.4	4.9	5.9	5.0	6.5	6.3	6.0	5.8
7	6.5	5.8	7.0	5.9	6.6	6.5	7.3	7.2
8	5.4	4.8	5.9	5.0	7.1	6.9	7.7	7.6
9	5.9	5.5	6.3	5.5	6.8	6.7	7.4	7.4
10	7.3	6.2	7.8	6.5	7.7	7.5	7.6	7.4
11	6.0	5.6	6.4	5.6	7.0	7.0	7.6	7.6
12	7.2	6.3	7.6	6.5	7.1	7.1	6.9	6.9
13	8.4	7.6	8.7	7.6	10.5	10.2	11.5	11.4
14	9.9	8.5	10.4	8.5	12.2	11.9	12.5	12.5
15	7.3	6.4	7.8	6.6	10.1	9.6	10.6	10.0
16	9.3	8.5	10.1	9.4	9.1	8.9	8.3	8.2
17	10.9	10.1	11.7	10.7	11.7	11.7	12.2	12.0
18	8.0	7.6	8.1	7.0	9.2	9.0	9.4	8.9
19	9.7	9.0	10.6	9.7	11.3	11.1	11.9	11.7
20	8.4	7.3	8.6	6.9	10.3	10.3	10.6	10.6
21	7.5	6.7	7.5	6.2	9.0	8.5	9.3	8.7
22	7.9	7.0	7.9	6.4	8.9	8.2	9.1	8.2

APPENDIX D

Pearson Correlation Coefficients for the Right Kidney of All Dogs

	crd R1	crs R1	ctd R1	cts R1	crd R2	crs R2	ctd R2	cts R2	R
crd R1	1.000	0.982	0.988	0.965	0.986	0.976	0.982	0.956	0.947
crs R1	0.982	1.000	0.965	0.979	0.966	0.980	0.962	0.963	0.921
ctd R1	0.988	0.965	1.000	0.977	0.966	0.956	0.982	0.966	0.940
cts R1	0.965	0.979	0.977	1.000	0.933	0.948	0.955	0.972	0.909
crd R2	0.986	0.966	0.966	0.933	1.000	0.989	0.988	0.957	0.932
crs R2	0.976	0.980	0.956	0.948	0.989	1.000	0.981	0.973	0.917
ctd R2	0.982	0.962	0.982	0.955	0.988	0.981	1.000	0.983	0.936
cts R2	0.956	0.963	0.966	0.972	0.957	0.973	0.983	1.000	0.902
R	0.947	0.921	0.940	0.909	0.932	0.917	0.936	0.902	1.000

Legend:

crd R1 = crest to dorsum CT Scan 1
crs R1 = crest to skin CT Scan 1
ctd R1 = center to dorsum CT Scan 1
cts R1 = center to skin CT Scan 1
crd R2 = crest to dorsum CT Scan 2
crs R2 = crest to skin CT Scan 2
ctd R2 = center to dorsum CT Scan 2
cts R2 = center to skin CT Scan 2
R = right kidney scintigraphy

APPENDIX E

Pearson Correlation Coefficients for the Left Kidney of All Dogs

	crd L1	crs L1	ctd L1	cts L1	crd L2	crs L2	ctd L2	cts L2	L
crd L1	1.000	0.995	0.971	0.970	0.991	0.989	0.979	0.970	0.828
crs L1	0.995	1.000	0.959	0.969	0.985	0.991	0.970	0.972	0.843
ctd L1	0.971	0.959	1.000	0.992	0.957	0.949	0.985	0.974	0.738
cts L1	0.970	0.969	0.992	1.000	0.956	0.958	0.981	0.986	0.753
crd L2	0.991	0.985	0.957	0.956	1.000	0.995	0.975	0.965	0.838
crs L2	0.989	0.991	0.949	0.958	0.995	1.000	0.967	0.969	0.840
ctd L2	0.979	0.970	0.985	0.981	0.975	0.967	1.000	0.991	0.772
cts L2	0.970	0.972	0.974	0.986	0.965	0.969	0.991	1.000	0.772
L	0.828	0.843	0.738	0.753	0.838	0.840	0.772	0.772	1.000

Legend:

crd L1 = crest to dorsum CT Scan 1
crs L1 = crest to skin CT Scan 1
ctd L1 = center to dorsum CT Scan 1
cts L1 = center to skin CT Scan 1
crd L2 = crest to dorsum CT Scan 2
crs L2 = crest to skin CT Scan 2
ctd L2 = center to dorsum CT Scan 2
cts L2 = center to skin CT Scan 2
L = left kidney scintigraphy

APPENDIX F

Pearson Correlation Coefficients for the Right Kidney of Small Dogs

	crd R1	crs R1	ctd R1	cts R1	crd R2	crs R2	ctd R2	cts R2	R
crd R1	1.000	0.924	0.951	0.979	0.923	0.810	0.918	0.868	0.672
crs R1	0.924	1.000	0.788	0.917	0.928	0.921	0.881	0.907	0.611
ctd R1	0.951	0.788	1.000	0.938	0.867	0.686	0.896	0.790	0.636
cts R1	0.979	0.917	0.938	1.000	0.912	0.798	0.891	0.854	0.641
crd R2	0.923	0.928	0.867	0.912	1.000	0.949	0.983	0.978	0.597
crs R2	0.810	0.921	0.686	0.798	0.949	1.000	0.914	0.971	0.528
ctd R2	0.918	0.881	0.896	0.891	0.983	0.914	1.000	0.968	0.587
cts R2	0.868	0.907	0.790	0.854	0.978	0.971	0.968	1.000	0.500
R	0.672	0.611	0. 636	0.641	0.597	0.528	0.587	0.500	1.000

Legend:

crd R1 = crest to dorsum CT Scan 1
crs R1 = crest to skin CT Scan 1
ctd R1 = center to dorsum CT Scan 1
cts R1 = center to skin CT Scan 1
crd R2 = crest to dorsum CT Scan 2
crs R2 = crest to skin CT Scan 2
ctd R2 = center to dorsum CT Scan 2
cts R2 = center to skin CT Scan 2
R = right kidney scintigraphy

APPENDIX G

Pearson correlation coefficients for Left Kidney of Small Dogs

	crd L1	crs L1	ctd L1	cts L1	crd L2	crs L2	ctd L2	cts L2	L
crd L1	1.000	0.976	0.832	0.860	0.952	0.921	0.912	0.894	0.354
crs L1	0.976	1.000	0.757	0.826	0.940	0.950	0.843	0.894	0.469
ctd L1	0.832	0.757	1.000	0.978	0.787	0.730	0.926	0.892	0.022
cts L1	0.860	0.826	0.978	1.000	0.821	0.800	0.940	0.942	0.145
crd L2	0.952	0.940	0.787	0.821	1.000	0.978	0.882	0.873	0.323
crs L2	0.921	0.950	0.730	0.800	0.978	1.000	0.843	0.871	0.379
ctd L2	0.912	0.873	0.926	0.940	0.882	0.843	1.000	0.981	0.186
cts L2	0.894	0.894	0.892	0.942	0.873	0.871	0.981	1.000	0.254
L	0.354	0.469	0.022	0.145	0.323	0.379	0.186	0.254	1.000

Legend:

crd L1 = crest to dorsum CT Scan 1
crs L1 = crest to skin CT Scan 1
ctd L1 = center to dorsum CT Scan 1
cts L1 = center to skin CT Scan 1
crd L2 = crest to dorsum CT Scan 2
crs L2 = crest to skin CT Scan 2
ctd L2 = center to dorsum CT Scan 2
cts L2 = center to skin CT Scan 2
L = left kidney scintigraphy

APPENDIX H

Pearson Correlation Coefficients for the Right Kidney of Large Dogs

	crd R1	crs R1	ctd R1	cts R1	crd R2	crs R2	ctd R2	cts R2	R
crd R1	1.000	0.964	0.991	0.950	0.978	0.974	0.972	0.945	0.968
crs R1	0.964	1.000	0.957	0.985	0.916	0.960	0.921	0.946	0.905
ctd R1	0.991	0.957	1.000	0.965	0.963	0.963	0.977	0.964	0.959
cts R1	0.950	0.985	0.965	1.000	0.897	0.941	0.925	0.963	0.892
crd R2	0.978	0.916	0.963	0.897	1.000	0.977	0.986	0.942	0.961
crs R2	0.974	0.960	0.963	0.941	0.977	1.000	0.972	0.969	0.926
ctd R2	0.972	0.921	0.977	0.925	0.986	0.972	1.000	0.976	0.960
cts R2	0.945	0.946	0.964	0.963	0.942	0.969	0.976	1.000	0.909
R	0.968	0.905	0.959	0.892	0.961	0.926	0.960	0.909	1.000

Legend:

crd R1 = crest to dorsum CT Scan 1
crs R1 = crest to skin CT Scan 1
ctd R1 = center to dorsum CT Scan 1
cts R1 = center to skin CT Scan 1
crd R2 = crest to dorsum CT Scan 2
crs R2 = crest to skin CT Scan 2
ctd R2 = center to dorsum CT Scan 2
cts R2 = center to skin CT Scan 2
R = right kidney scintigraphy

APPENDIX I

Pearson Correlation Coefficients for the Left Kidney of Large Dogs

	crd L1	crs L1	ctd L1	cts L1	crd L2	crs L2	ctd L2	cts L2	L
crd L1	1.000	0.991	0.972	0.985	0.983	0.979	0.956	0.961	0.566
crs L1	0.991	1.000	0.951	0.979	0.975	0.986	0.935	0.955	0.610
ctd L1	0.972	0.951	1.000	0.991	0.946	0.927	0.981	0.963	0.473
cts L1	0.985	0.979	0.991	1.000	0.968	0.962	0.977	0.978	0.530
crd L2	0.983	0.975	0.946	0.968	1.000	0.991	0.961	0.974	0.602
crs L2	0.979	0.986	0.927	0.962	0.991	1.000	0.936	0.966	0.614
ctd L2	0.956	0.935	0.981	0.977	0.961	0.936	1.000	0.986	0.484
cts L2	0.961	0.955	0.963	0.978	0.974	0.966	0.986	1.000	0.524
L	0.566	0.610	0.473	0.530	0.602	0.614	0.484	0.524	1.000

Legend:

crd L1 = crest to dorsum CT Scan 1
crs L1 = crest to skin CT Scan 1
ctd L1 = center to dorsum CT Scan 1
cts L1 = center to skin CT Scan 1
crd L2 = crest to dorsum CT Scan 2
crs L2 = crest to skin CT Scan 2
ctd L2 = center to dorsum CT Scan 2
cts L2 = center to skin CT Scan 2
L = left kidney scintigraphy