Radiographic, ultrasonographic, and anatomic assessment of femoral trochlea morphology in red foxes (Vulpes vulpes)

James E. Miles, BVetMed, PhD; Ulrik Westrup, DVM; Eiliv L. Svalastoga, DVM, PhD; Thomas Eriksen, DVM, PhD

Objective—To compare repeatability and equivalency of measures of femoral trochlea depth and trochlear angle in red foxes (Vulpes vulpes) determined by use of radiography, ultrasonography, and digital photography of cadaver limbs.

Sample—24 pelvic limbs from 12 red fox cadavers.

Procedures—Cranio proximal-craniodistal oblique (skyline) and lateromedial radiographic views of the stifle joint and ultrasonographic images at 5 locations along the femoral trochlea were used in the study. Spacing of the 5 locations was determined on the basis of patellar position with the stifle joint at various caudal angles ranging from 96° to maximal extension (approx 170°). Ultrasonographic measurements were compared with those obtained at matched locations on photographs of anatomic preparations. Trochlear depth was assessed with all 3 image formats, and trochlear angle (measured between the trochlear ridges and sulcus) was assessed on radiographs and ultrasonographic images. Patellar thickness was measured on radiographs. Values obtained were compared by means of ANOVA, modified Bland-Altman plots, and repeatability testing.

Results—Depth measurement repeatability was considered good for all modalities. Small but significant differences between mean ultrasonographic trochlear depth and anatomic (photographic) measurements were found at 3 locations; 95% limits of agreement for paired anatomic and ultrasonographic measurements were wide. The ratio of trochlear depth to radiographic patellar thickness was approximately 30% for all modalities. Trochlear angle measurements were more variable than trochlear depth measurements, especially in the distal aspect of the trochlea.

Conclusions and Clinical Relevance—Paired anatomic and ultrasonographic measurements did not appear equivalent in this study, possibly attributable to imprecise probe location, which could limit quantitative use of ultrasonography in assessing proximal trochlear depth in a clinical setting. (Am J Vet Res 2014;75:1056–1063)

Medial patellar luxation in dogs requiring surgical treatment is frequently addressed by a combination of interventions, including trochleoplasty.1,2 Although the femoral trochlea can be assessed with cranioproximal-craniodistal oblique view (skyline) radiographs, the need for trochleoplasty is largely decided by clinical and visual inspection following arthroscopy.3–5 Typically, a minimum trochlear depth of 50% of radiographically determined patellar thickness is considered desirable following trochleoplasty in dogs.2,4 Reliably determining the need for trochleoplasty before surgery might avoid the morbidity associated with unwarranted trochleoplasty5 or arthroscopy.6

Trochlear depth measurement on lateral7 and skyline radiographic views2,6 and CT images9 has been reported. These methods require strict attention to positioning and may necessitate sedation or anesthesia, and CT may not always be available. Ultrasonographic examination offers a rapid, noninvasive means for investigation of canine stifle joints. Ultrasonographic measurements of femoral trochlea depth and the trochlear angle have been investigated in clinically normal dogs and dogs with medial patellar luxation;2 however, the repeatability and reproducibility of these measurements have not been determined.

The aim of the study reported here was to investigate the repeatability of measurements made on photographs of anatomic specimens, radiographs, and ultrason sound scan images and to assess agreement between anatomic and ultrasonographic measurements in pelvic limbs from red fox (Vulpes vulpes) cadavers, as a surrogate for the pelvic limbs of domestic dogs. Studies10–11 have shown substantial conservation of anatomy between domestic dogs and red foxes, making this
substitution acceptable for a method comparison study.

Materials and Methods

Sample—Twelve red fox cadavers (6 males and 6 females) were obtained from a commercial unit following euthanasia by electrocution according to Danish regulations. Both pelvic limbs from each animal were used. Physical and radiographic examination revealed no evidence of stifle joint abnormalities. Data (related to patellar positioning, quadriceps angle, and tibial tuberosity—trochlear groove offset measurements) obtained from the same cadavers have been reported previously.\textsuperscript{12–14} All measurements and image readings in the present study were performed by a single observer (JM).

Procedures—Lateromedial stifle joint radiographs (taken as part of another study\textsuperscript{12} with the same pelvic limbs positioned at caudal flexion angles of 96°, 113°, 130°, and 148° by use of a goniometer placed directly over selected bony landmarks) were assessed with the aid of freely available image analysis software.\textsuperscript{b} By use of the segmented line tool, the distance from the most cranial aspect of the intercondylar notch (distal landmark) to the convergence of the trochlear ridges with the line of the cranial diaphyseal cortex (proximal landmark) was measured along the base of the femoral trochlea. With the same tool, the distance from the distal landmark to the level of the distal pole of the patella along the femoral trochlea was measured and expressed as a ratio to the length of the sulcus itself (Figure 1). The level of the distal pole of the patella was defined as the point at which a line normal (perpendicular) to the base of the femoral trochlea touched the distal pole. Mean ratios for each of the 4 stifle joint flexion angles for all 24 limbs were calculated and used to create a custom acetate overlay for later measurements. The 4 ratios thus defined 4 locations (designated 1 to 4) between the distal and proximal aspects of the trochlea. An additional location (designated

\begin{figure}[h]
\centering
\includegraphics[width=\textwidth]{image1.png}
\caption{Lateromedial radiographic view of a stifle joint from a red fox (Vulpes vulpes) cadaver in a study to evaluate radiographic, ultrasonographic, and anatomic measures of femoral trochlea morphology. A computer software segmented line tool was used to measure distoproximal patellar position within the femoral trochlea. The distance from the distal landmark (cranial aspect of the intercondylar notch; gray square) to the point where a line normal to the trochlea touches the distal pole of the patella is shown. The distance to the cortical divergence at the start of the trochlea proximally was measured similarly, and the 2 measurements were used to calculate a ratio reflecting the position of the distal aspect of the patella along the length of the trochlea. A pin used as a positioning aid can be seen in the femoral condyles.}
\end{figure}

\begin{figure}[h]
\centering
\includegraphics[width=\textwidth]{image2.png}
\caption{Radiographic (A and B) and ultrasonographic (C and D) images depicting determination of trochlear depth (d) and trochlear angle (\(\theta\)). To measure trochlear depth, a tangent to the 2 trochlear ridges was drawn, and the furthest perpendicular distance to the base of the trochlea was determined to be the trochlear depth (A and C). The trochlear angle was measured between lines connecting the points where a tangent line touched the trochlear ridges and the deepest point of the trochlea (B and D).}
\end{figure}
ultrasonographic images were obtained from multiple levels in each stifle joint with a linear 10-MHz probe. Images were recorded at the level of the distal pole of the patella with the stifle joint flexed to caudal angles of 96°, 113°, 130°, and 148° and in forced maximal extension (approx 170°) on the basis of goniometric measurement between the greater trochanter, lateral epicondylar point, and lateral malleolus. The quadriceps muscles were manually tensioned by grasping the muscle bellies and moving them proximally to prevent patellar ligament laxity. Ultrasonographic image selection was based on the subjective impression of least trochlear depth to reduce the risk of an oblique scan angle relative to the trochlea. For comparison purposes, the positions for ultrasonographic examination were designated 1′ (96°) to 5′ (maximal extension [170°]).

Skyline radiographic views of the femoral trochlea were obtained with a cassette placed directly beneath the stifle joint according to a previously described technique; stifle joint flexion angle was not standardized for these images. A single image was obtained for each limb. Radiographic and ultrasonographic images were stored as DICOM (Digital Imaging and Communication in Medicine) files, anonymized with commercial software, and randomized for 2 readings with freely available software before randomization for 2 readings.

All measurements on DICOM files were made via freely available software with distances recorded to the nearest 0.1 mm and angles to the nearest 0.1°. Trochlear depth was measured by first drawing a line tangential to the lateral and medial trochlear ridges (Figure 2). A second line was extended perpendicular to the first to measure the maximal depth of the trochlea. The trochlear angle was measured with the vertex at the point of maximal depth and the 2 sides passing through the contact points of the ridges with the tangential line. Patella thickness, defined as the shortest distance across the patella at the distoproximal midpoint along the patella’s length, was measured for all stifle joints from the 96° caudal angle (100%) landmarks of the straightened trochlear baseline. The proximal landmark was the point of maximal depth of the trochlea. Trochlear depth was calculated as the mean of the medial and lateral depths for each location. The 5 locations for measurement derived from the lateromedial stifle radiographs were identified by use of a single customized acetate overlay that could be aligned with both the proximal and distal ends of the trochlea and the straightened baseline. The acetate overlay contained 2 lines diverging symmetrically from a central principal axis, for alignment with the distal (0%) and proximal (100%) landmarks of the straightened trochlear baseline. The principal axis was approximately 20 cm long and the 2 lines approximately 20 cm apart at their maximal divergence. Between these 2 lines, and diverging from the same point as them, were a series of lines corresponding to locations 1 to 5. A further series of parallel lines spaced 1 cm apart and perpendicular to the principal axis facilitated alignment of the overlay with the straightened trochlear baseline. By positioning the acetate sheet over the monitor displaying the straightened trochlear image with the outermost diverging lines crossing the distal and proximal landmarks of the trochlear baseline, the positions of the intervening locations could be readily and quickly identified.

Based on the reported focusing distance in the JPEG metadata, a magnification error of approximately 4% was calculated for the anatomic measurements from digital photographs; anatomic measurements were corrected by dividing by 1.04 prior to analysis.

Statistical analysis—Following breaking of randomization, data were assessed for homoscedasticity on the basis of plots of SD against the mean and by use of the White test. Agreement between readings for each modality was assessed on the basis of the within-subject SD and reliability coefficient (an indicator of the expected maximum difference between pairs of measurements in 99% of cases) with corrections for multiple readings. Agreement between anatomic and ultrasonographic measurements was assessed via Bland-Altman difference analysis with corrections for multiple readings. Agreement between modalities was assessed on the basis of paired measurements; if 1 measurement was missing, the pair was excluded.
Following confirmation of data normality by means of the Shapiro-Wilk test, comparisons between lateral and medial femoral trochlear ridge height (anatomic) and trochlear depths (anatomic and ultrasonographic) at various distoproximal locations were made with mixed-model ANOVA with fox as a random factor and limb (left or right), ridge (medial or lateral), and distoproximal location as fixed factors. Statistical testing was performed with the aid of a statistical package. When multiple comparisons were made, Bonferroni-corrected P values were reported. Values of P < 0.05 were considered significant. Morphometric data were reported as mean ± SD, and reliability data and location data as mean (95% CI).

![Diagram](Image 1)

**Figure 3**—Photographs depicting the straightening of the trochlea in an image for anatomic depth measurement. After photographing the femur (sectioned through the trochlea), the outline of the osteochondral junction was traced on the image through computer software with a segmented line tool (A), taking care to include sufficient segments that closely approximated the true curvature. The straighten function yielded an image aligned horizontally along the drawn baseline (B). Use of a custom acetate overlay sheet that could be aligned proximally, distally, and with the osteochondral baseline permitted accurate identification of measurement locations 1 to 4, representing trochlear locations of the distal pole of the patella in radiographs when the stifle joints were positioned at caudal flexion angles of 96°, 113°, 130°, and 148° by use of a goniometer placed directly over selected bony landmarks, and location 5, which was spaced at a distance 15% of the total trochlear length proximal to location 4 to approximate the position of the patellar landmark with the stifle joint in maximal extension (C).

**Results**

Although photographic images from all 24 stifle joints were obtained for the anatomic portion of the study, ultrasonographic images were only available from 22 stifle joints because of technical issues, and 12 of these were either missing or were deemed of non-diagnostic quality on review. Skyline and lateral radiographic images were analyzed for all stifle joints. One pair of values from the ultrasonographic measurement experiments was discarded because it was clear that trochlear depth had been misrecorded on 1 reading.

The distal pole of the patella was located at 19% (95% CI, 17% to 22%), 33% (95% CI, 30% to 35%), 47% (95% CI, 43% to 50%), and 62% (95% CI, 59% to 66%) of the length of the femoral trochlear sulcus (as measured from the distal landmark) at caudal stifle joint angles of 96°, 113°, 130°, and 148°, respectively. Location 5 was defined as 77% of the distance from the distal landmark. Mean radiographic patellar craniocaudal thickness was 5.5 ± 0.5 mm. Measurement repeatability between readings was considered good, with acceptably low within-subject SD and reliability coefficients for all modalities (Table 1).

**Anatomic trochlear depth**—Mean femoral trochlear depth over the 5 measurement locations determined by use of digital photographs (after correction for magnification) was 1.5 mm. Femoral trochlear depth for locations 1 (most distal), 2, and 3 differed significantly (corrected P < 0.004) from measurements for locations 4 and 5 (most proximal), with a maximal variation of 0.3 mm (Figure 4). These changes concealed an asymmetric variation in medial and lateral femoral trochlear ridge height, with the lateral ridge generally higher than the medial ridge distally and the medial ridge higher than the lateral ridge proximally. The difference in height between the 2 ridges was significantly (P < 0.08) different from 0 in locations 1, 3, and 4, with mean ± SD differences of up to 0.4 ± 0.4 mm. Mean ratios of anatomic trochlear depth to radiographic patellar thickness ranged from 25% to 31% across the 5 measurement locations.

**Ultrasonographic trochlear depth and angle**—Measurements of femoral trochlear depth were significantly (P < 0.05) greater at the 1′ location than at 2′ or 3′ by approximately 0.2 mm (Figure 5). Measurements of femoral trochlear angle also varied, with angles at 1′ significantly (corrected P < 0.014) smaller than those at 2′, 3′, or 4′, by approximately 5°. Data spread for the trochlear angle measurement decreased with more proximal measurement locations. Mean ratios of ultrasonographic trochlear depth to radiographic patellar thickness ranged from 28% to 34% across the 5 measurement locations.

**Skyline radiographic trochlear depth and angle**—Mean radiographic trochlear depth was 1.7 ± 0.3 mm, and mean radiographic trochlear angle was 131 ± 5°. The mean ratio of radiographic trochlear depth to radiographic patellar thickness was 30 ± 5%.

**Agreement between anatomic and ultrasonographic measurements**—Although mean values for trochlear depth at each measured location were similar
between the 2 modalities, and the Bland-Altman difference plot showed that ultrasonographic measurements were negligibly higher than anatomic measurements from digital photographs (mean difference between techniques [bias], 0.03 mm), the 95% limits of agreement were wide at –0.7 to 0.7 mm (Figure 6). Mean ultrasonographic measurements significantly exceeded mean anatomic measurements by approximately 0.2 mm at location pairs 1–1′ (P = 0.005) and 2–2′ (P = 0.02), whereas at location pair 4–4′, mean anatomic values exceeded mean ultrasonographic values by 0.2 mm (P = 0.01).

**Discussion**

Results of the present study showed that trochlear depth measurements could be made with an acceptable degree of repeatability by 1 observer using ultrasonographic and radiographic images. The limits of agreement between anatomic and ultrasonographic measurements were considered wide, suggesting that the clinical utility of ultrasonographic measurements may be limited, depending on the expected trochlear depth and clinically defined cutoffs for intervention. Repeatability was considered better for anatomic measurements (which were obtained from digital photographs) and for radiographic measurements than for ultrasonographic measurements. Because repeatability in this study was determined from single images, the influence of probe or patient repositioning was not a factor that affected results. We believe that the most likely cause was the axial resolution of the relevant images, which was markedly less for ultrasonography than for other methods. Ultrasonographic image quality is continually improving, and use of newer machines and high-frequency probes should help ameliorate this problem. Operator-induced errors in ultrasonographic measurements (attributable to incorrect caliper positioning) have been reported to be as high as 2% to 20%18; our results fit within this pattern.

Despite apparent similarities between ultrasonographic and anatomic depth measurements indicated by the low degree of bias on the Bland-Altman analysis, the limits of agreement for the 2 modalities appeared too wide for clinical use because they approximated the mean anatomic trochlear depth in this study population. The limits of agreement define a range within which 95% of differences between the 2 modalities can

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Table 1—Repeatability of trochlear depth and trochlear angle measurements obtained by use of photographic, ultrasonographic, and radiographic images of 24 stifte joints from 12 red fox (Vulpes vulpes) cadavers.

<table>
<thead>
<tr>
<th>Trochlear measurement</th>
<th>Within-subject SD</th>
<th>Repeatability coefficient</th>
</tr>
</thead>
<tbody>
<tr>
<td>Anatomic depth measured via digital photography (mm)</td>
<td>0.08 (0.08–0.09)</td>
<td>0.23 (0.21–0.26)</td>
</tr>
<tr>
<td>Ultrasonographic depth (mm)*</td>
<td>0.11 (0.10–0.13)</td>
<td>0.32 (0.28–0.37)</td>
</tr>
<tr>
<td>Ultrasonographic angle (°)*</td>
<td>2.60 (2.20–2.90)</td>
<td>7.30 (6.30–8.30)</td>
</tr>
<tr>
<td>Radiographic depth (mm)</td>
<td>0.08 (0.06–0.11)</td>
<td>0.25 (0.18–0.32)</td>
</tr>
<tr>
<td>Radiographic angle (°)</td>
<td>2.00 (1.4–2.60)</td>
<td>5.60 (3.90–7.30)</td>
</tr>
</tbody>
</table>

Values are given as mean (95% CI), rounded to 2 decimal places. The repeatability coefficient gives an indication of the expected maximum difference between pairs of measurements in 95% of cases. Anatomic depth measured in JPEG images was corrected for magnification by dividing by 1.04 prior to analyses.

*Ultrasonography was performed for 22 of 24 stifte joints, with 12 of these images unavailable or excluded from analysis for technical reasons.
be expected to fall. A number of potential causes for this poor agreement can be identified. A recent study of accuracy of ultrasonographic distance measurements found that, depending on the machine, probe, acoustic velocity of the tissues, spatial resolution, and aberration correction, computer-assessed accuracy (ie, with exclusion of operator-induced errors) was approximately 1% to 5%. Relative image resolution may have some role in this low degree of accuracy. Additionally, errors in probe positioning, where the probe is not positioned perpendicular to the trochlear base at the scan location, could result in artificial overestimation of ultrasonographic measurements. However, if this were consistently the case in our study, bias assessed with the Bland-Altman method would be expected to be clearly positive instead of nearly zero. It is also likely that the anatomic and ultrasonographic measurements were not obtained at equivalent levels in the trochlea and therefore the measurements were not truly paired. The anatomic locations selected for measurement on digital photographs were mean locations identified by use of lateromedial radiographs at accurately defined caudal flexion angles obtained with a goniometer placed directly on the bony landmarks. In comparison, the ultrasonographic locations were determined by palpation of landmarks through the soft tissues and use of a goniometer resting on the outer surface of the limb, before use of the distal pole of the patella as a landmark. Considerable inaccuracy may have been inadvertently introduced at this stage. This may have been compounded by use of mean locations instead of locations specific to each fox, given that patellar positioning was found to vary among these animals in another study. Thus, although the limits of agreement appear to preclude use of ultrasonographic measurements of trochlear depth, we suggest that these limits were artificially wide owing to the method application in the
study and do not necessarily indicate ultrasonographic inaccuracy, but rather spatial inaccuracy. Nonetheless, this raises an important question about the use of quantitative ultrasonographic measurements of trochlear depth, in that to ensure measurement repeatability and interobserver reproducibility, some reliable method of determining probe position relative to the underlying anatomy is essential. Assessment of ultrasonographic accuracy of trochlear measurements and of the necessity for spatial accuracy in the clinical setting appears warranted. Without this, trochlear evaluation by this method must be largely qualitative.

The ratio of trochlear depth to patellar thickness in this population was approximately 30%. Although we did not correct for magnification of the patella in radiographs when calculating our ratios (because data that would permit precise calculation were missing), subsequent estimates for the correction factor did not result in substantial changes in the ratios reported here (data not shown). Similar values have been reported before in a radiographic study of dogs with and without medial patellar luxation, and slightly lower ratios were reported following CT evaluation of dogs with medial patellar luxation in a clinical setting. In addition, we noted in the present study that the major contributor to trochlear depth proximally was the medial trochlear ridge. Although speculative, it might be that this represents a passive restraint to medial patellar luxation in the proximal portion of the trochlea in clinically normal (unaffected by medial patellar luxation) dogs and foxes in which a medially directed quadriceps angle can still be found. Evaluation of trochlear specimens from clinically normal dogs and dogs with patellar luxation might elucidate the relevance of these findings to the canine population.

A surprising degree of variability in ultrasonographic trochlear angle measurement was noted in relation to the values obtained for depth, particularly in the more distal region of the trochlea. Considering that medial patellar luxation is usually considered linked to deficiencies in the proximal aspect of the trochlea, this may be of little clinical relevance. However, variability was still considered quite high in the middle to mid-proximal trochlear regions, which may reflect both the described resolution problems and more subjectivity in placement of the arms of the angle at these locations. Although it is tempting to consider the trochlear angle as potentially being a size-independent marker of morphology, the femoral trochlea does not appear to scale uniformly with changes in body mass. This, in concert with the variability issues identified here, likely renders the trochlear angle less suited to assessment of the trochlea than measurement of depth and determination of the trochlear depth-to-patellar thickness ratio.

Although the skyline radiographic images were considered to have better measurement repeatability than did ultrasonographic images, it was similarly difficult to ascribe a position along the trochlea to these images. Clinically, the proximal part of the trochlea is of prime importance in development and treatment of medial patellar luxation. However, positioning the limb to obtain a view of this region is hampered by musculature at the cranial aspect of the thigh. On the basis of values obtained in the present study, we estimated the location of the skyline images to be close to location 4 (ie, at approx 62% of the total trochlear length as measured from the distal landmark, and therefore just at the edge of, or possibly outside, the area of primary interest).

In this study, we found clinically acceptable measurement repeatability for anatomic, ultrasonographic, and radiographic measurements of trochlear depth when taken individually. Overall, measurement of trochlear depth and comparison of trochlear depth with radiographically measured patellar thickness appeared to provide more clinically useful information than did trochlear angle measurement. Anatomic and ultrasonographic measurements could not be considered equivalent under the study conditions, and quantitative use of ultrasonography to measure trochlear depth requires development of a method to ensure repeatability of spatial location and avoid artifactual variation in measurements.


d. Sante DICOM Editor, Santesoft, Athens, Greece.


f. Synedra View Personal 3, Synedra IT, Immobrunck, Austria.


h. SAS, version 9.2, SAS Institute Inc, Cary, NC.

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