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The comparability of HR-pQCT bone measurements is improved by scanning anatomically standardized regions

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Abstract

Introduction—High-resolution peripheral quantitative computed tomography (HR-pQCT) measures of bone do not account for anatomic variability in bone length: a 1-cm volume is acquired at a fixed offset from an anatomic landmark. Our goal was to evaluate HR-pQCT measurement variability introduced by imaging fixed vs. proportional volumes and to propose a standard protocol for relative anatomic positioning.

Methods—Double-length (2-cm) scans were acquired in 30 adults. We compared measurements from 1-cm subvolumes located at the default fixed offset, and the average %-of-length offset. The average position corresponded to $4.0\% \pm 1.1$ mm for radius, and $7.2\% \pm 2.2$ mm for tibia. We calculated the RMS difference in bone parameters and T-Scores to determine the measurement variability related to differences in limb length. We used anthropometric ratios to estimate the mean limb length for published HR-pQCT reference data, and then calculated mean %-of-length offsets.

Results—Variability between fixed vs. relative scan positions was highest in the radius, and for cortical bone in general (RMS difference Ct.Th = 19.5%), while individuals had T-score differentials as high as +3.0 SD (radius Ct.BMD). We estimated that average scan position for published HR-pQCT reference data corresponded to 4.0% at the radius, and 7.3% at tibia.

Conclusion—Variability in limb length introduces significant bias to HR-pQCT measures, confounding cross-sectional analyses and limiting the clinical application for individual assessment of skeletal status. We propose to standardize scan positioning using 4.0% and 7.3% of total bone length for the distal radius and tibia, respectively.

Keywords

HR-pQCT; bone; osteoporosis; precision; standardization; multicenter study

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Conflict of interest

All authors report no conflict of interest.

Introduction

High-resolution peripheral quantitative computed tomography (HR-pQCT) is an *in vivo* imaging technique increasingly used to assess trabecular and cortical bone density, structure, and mechanics in the peripheral skeleton [1]. Because of challenges related to scanner geometry, patient immobilization, and dose considerations, *in vivo* imaging protocols for the distal radius and tibia typically span a limited length along the bone – approximately 1 cm. Scan localization is manually prescribed at scan time based on a planar scout radiograph (scout view image); the operator positions a reference line to intersect a visually identified anatomic landmark. By convention[2–4], the first slice of the volumetric scan is positioned at a fixed distance from this reference line (9.5 mm for radius and 22.5 mm for tibia).

This approach to scan localization does not account for anatomic variability due to limb length. As a consequence, the scan volume acquired for a long-limbed individual will be positioned at a relatively more distal region of the metaphysis compared to the volume for a short-limbed individual. In the forearm, scan positioning is of particular concern due to the large degree of densitometric and geometric variation along the distal-proximal axis of the ultradistal radius [5,6]. For example, cortical thickness varies by nearly an order of magnitude from the distal-most slice to the most proximal slice of a standard HR-pQCT scan (0.2 to 1.4 mm) [5]. Therefore, dependence on limb length likely introduces significant bias to group-wise comparisons between populations with different body sizes (e.g. gender and race comparison studies) and potentially diminishes the statistical power in discriminant analyses. Furthermore the dependence on bone length significantly limits the clinical utility of HR-pQCT for individual assessment of bone quality in patients with body sizes that differ from the mean of the reference population [7–11]. Adapting scan localization protocols to account for subject-specific limb length would therefore remove an important source of measurement variability, thereby improving the power of this technology in musculoskeletal research and relevance for clinical practice.

Clinical imaging systems that measure bone density in the peripheral skeleton have long utilized procedures to anatomically standardize scan positioning. International Society for Clinical Densitometry (ISCD)-sponsored efforts have demonstrated improved comparability for standardized (region of interest) ROI analysis for DXA of the ultradistal forearm [12]. Other standard analysis ROIs are based on total or fractional bone length (total radius; 1/3 distal radius). In conventional peripheral quantitative computed tomography (pQCT), single axial images are acquired at predefined percentages of total bone length; typically located distally (4%) or in the diaphysis (33%–66%) [13]. Relative scan positioning in HR-pQCT has been used by some research groups for studies of pediatric and adolescent subjects to account for different stages of growth [14,15]. To our knowledge, only Shanbhogue et al. [16] compared group-wise differences between study groups in an adult population based on scans acquired at both fixed and relative scan positions. They compared group-wise differences based on scans acquired separately at the standard fixed offset and at a relative offset. However, the relative offset positions selected for this study (4% for the radius and 7% for the tibia) were approximated from unpublished anthropometry data.

The first goal of our study was to directly determine the variability in HR-pQCT bone measures introduced by positioning the scan and analysis volumes at an absolute fixed distance offset (and thereby at variable relative anatomic locations). To isolate this specific effect, we collected double length *in vivo* scans and analyzed sub-volumes corresponding to scans positioned at: (1) the default fixed absolute offset; and (2) an anatomically standard position, relative to the directly measured total limb length. We also tested the measurement of a fixed scan length, compared to subvolume analysis of slices spanning a defined %-of-length. The second goal was to propose a standard procedure for prescribing HR-pQCT scans for the distal radius and tibia that adapts the measured volume of interest (VOI) to the subject's limb length. In defining an anatomically standardized scan location for these sites we aimed to provide approximate comparability to existing reference data in the literature that have previously been collected using the default fixed-offset protocols.

Material and Methods

Subjects

A total of 30 subjects were included in this study. Of these, 21 subjects were scanned at University of California, San Francisco (UCSF) and 9 at Mayo Clinic. Subjects included 13 women and 17 men, with age range 65–80 years. Exclusion criteria from the parent study included a history of metabolic bone disease, history of fracture at the region of interest, and the presence of metal implants that would interfere with imaging peripheral or axial skeletal sites. Descriptive details about the subjects' height and limb lengths are presented in Table 1(a). All subjects provided written informed consent to participate in this study. The Committees on Human Research of UCSF and Mayo Clinic approved all study procedures.

HR-pQCT imaging

At each site, a single trained operator imaged the non-dominant distal radius and tibia for each subject using the first generation XtremeCT system (Scanco Medical AG, Brüttisellen, Switzerland) following a modified protocol provided by the organizing center (UCSF). Each limb was immobilized and positioned to acquire a scout radiograph, used to prescribe the scan location. In contrast to the standard *in vivo* protocol [2,3], this modified procedure included the acquisition of two consecutive 9.02 mm stacks (18.04 mm, 220 slices in total), requiring two sequential orbits of the source-detector unit. The complete scan volume was positioned such that the central 110-slices coincided with the position of the default single-stack volume (Figure 1(a) and (b)). Accordingly, the first slice of the acquisition was offset 4.99 mm (radius) and 17.99 mm (tibia) proximal from the reference line. The total imaging time was 5.6 minutes per scan and resulted in 220 tomograms comprised of 82 μm isotropic voxels. Each orbit incurs an effective dose of approximately 4 μSv effective dose, therefore each scan incurred approximately 8 μSv . Following the acquisition, the operators graded image quality from 1 (no motion artifacts) to 5 (severe motion artifact) [17]. A single repeat scan was attempted if the image grade of the first scan exceeded 2. Scans graded 4 or higher were excluded from our analyses.

Image analysis and micro-finite element analysis

The complete 220 slice volume was analyzed using the manufacturer's semi-automated protocol for in vivo image analysis (IPL Version 5.08b, Scanco Medical) [4]. This procedure records mean values for standard parameters on a slice-by-slice basis to facilitate subsequent measurement of co-registered sub-volumes for longitudinal analyses. We utilized this feature to extract mean values for 110-slice sub-volumes where the position of the central slice corresponded to: (1) the position of the central slice for a scan located at the traditional fixed offset (e.g. the default "fixed offset"; and (2) a defined percentage of total limb length (e.g. the anatomically standardized position). In this case limb length was measured directly for each subject. In a second analysis we compared (1) fixed 110-slice sub-volumes centered at a defined percentage of total limb length and (2) subvolumes spanning a anatomically standard percentage of total limb length, and positioned at a defined percentage of total limb length. For each sub-volume, standard parameters were calculated. Integral BMD (Tt.BMD) and the individual density of the cortical and trabecular compartments (Ct.BMD and Tb.BMD) were measured using a phantom-derived density calibration [4]. We separated cortical and trabecular structure using a threshold-based binarization process [18]. From the binary image, we measured cortical thickness (Ct.Th) using an annular approximation [4,19], cortical porosity (Ct.Po) using direct volumetric methods [20–22], and trabecular number (Tb.N) and heterogeneity (Tb.Sp.SD) using the direct 3D distance transform method [23,24]. Linear-elastic micro-finite element analysis (μ FEA) (Scanco FE Software v1.12; Scanco Medical AG) was performed in corresponding 110-slice subvolumes to estimate failure load (L_{failure}) and calculate proximal and distal cortical load fraction (Ct.LF_{prox}, Ct.LF_{dist}) [25]. The μ FEA and Ct.Po measurements were calculated independently for every extracted subvolume because these measures are not incorporated into the manufacturer's patient analysis database.

Forearm and leg anthropometry

Measurements of forearm and lower leg length were performed using a standard protocol provided by the coordinating center (UCSF). Measurements were made superficially using a soft pencil to demarcate anatomic landmarks on the skin, and a meter tape to measure the distance between landmarks. Ulnar length was measured as a surrogate of radial length because of the greater detectability by palpation of the proximal and distal margins of the ulna. Ulnar length was measured on the medial side of the forearm (Figure 2(a)). The subject's elbow was positioned at the intersection of two perpendicular rigid surfaces: the elbow was rested on a horizontal surface (table) while the forearm was supported by a vertical surface. The palms were open and facing the subject, with the fingers extended vertically. The distal apex of the ulnar styloid process was demarcated on the lateral side of the arm with a soft pencil, and ulnar length was measured as the Euclidean distance between the horizontal surface and the styloid process demarcation. Tibial length was measured on the medial side of the lower leg (Figure 2(b)). Subjects were asked to sit on a chair with feet on the floor, and with legs slightly opened and perpendicular to the floor. Demarcations were made at the medial tibial plateau at the knee – identified by palpation – and the medial tibial malleolus at the ankle. Tibia length was measured as the Euclidean distance between these two anatomic landmarks. Operator reproducibility of the anthropometric measures was tested in 10 randomly selected volunteers by performing three repeat measurements at 30-

minute intervals. The superficial demarcations were erased after each measurement. Measurement precision (SD_{RMS}) was 3.2 mm for the forearm, and 7.0 mm for the lower leg.

Anatomically standard region

The average anatomical position of the scans collected for subjects in this study was calculated for the radius and tibia as the distance between a distal anatomic landmark and the central slice of the measured stack, expressed in fraction (%) of the total limb length directly measured in each subject. For the tibia, this percentage offset was determined with respect to the traditional reference line location for HR-pQCT: the distal margin of the tibial plafond. For the radius, we defined the relative offset as a percentage of ulnar length offset from the *proximal* margin of the articular surface of the radius (Figure 1c), consistent with other common procedures for measuring bone density at the distal radius (DXA, pQCT), but notably different from previous manufacturer recommendations for HR-pQCT. The position of the alternative reference landmark in the radius was determined retrospectively for each scan using software implemented for a complementary study of operator positioning precision [26]. Briefly, we developed a graphical user interface that reproduces the manufacturer's acquisition software. The scout view image corresponding to each scan is visualized in a simulated environment that mimics the positioning procedure performed during the actual scan. For each distal radius scout view image collected for this study, a single experienced operator at UCSF positioned a reference line on the proximal margin of the radial head according to our alternative landmark (Figure 1e–f). The simulation software recorded the position of the reference line in the native coordinate system of the scanner.

Estimation of average percent offset from published population studies

In an effort to establish standardized percent offsets that approximately correspond to existing reference data measured using fixed-offset scan regions, we estimated the mean relative scan location measured by the default protocol in major population studies from the literature. Our estimation was based on established anthropometric ratios between height and limb-length. We selected ten large HR-pQCT population studies comprised of different ethnicities, ages, and both sexes, where descriptive statistics for height are reported [3,21,27–34]. From each study, we estimated ulnar and tibial length as 16% and 22% of body height, respectively [35,36]. From limb length, we computed the average percentage position of the central slice of a standard 110-slice stack positioned in the default fixed-offset location, as described above. A weighted mean, by sample size, was calculated separately for male and female cohorts. Our standard %-of-length offset for each site was calculated as the unweighted mean of the male and female averages.

Statistics

For each subject scan, 110-slice subvolumes were extracted from the 220-slice scan at positions that correspond to the default fixed offset ROI, and the ROI at the mean %-of-length ROI for our study population. The slice-wise distance between the central slices of the fixed and relative subvolumes was determined. The root mean square (RMS) of these distances was calculated to summarize the average offset of the fixed position from the anatomically standard position. Furthermore, we calculated the percent difference between bone parameters measured in each subvolume. The range and RMS of these differences

were calculated to summarize the bias introduced by not accounting for limb length. The RMS difference between fixed length and %-of-length measurements performed at a standard %-of-length offset was additionally calculated to determine the additional bias of measuring a fixed number of slices. To determine the impact on the clinical application of HR-pQCT we calculated the corresponding RMS difference in gender-specific T-score, based on normative data for 20–29 year old men and women from the UCSF normative database [21,37]. To evaluate the variation in measurements in more detail we calculated the RMS percent difference for all parameters as a function of distance from the average relative scan position. This was accomplished by calculating mean measures in sequential 110-slice subvolumes, incrementally shifting the position of the central slice by two slices (164 μm) over 2.13 mm proximal and distal to the mean relative position. Linear or, where appropriate, non-linear fitting was performed to establish models to estimate measurement bias between studies or populations, according to group-wise differences in limb-length.

Results

Relative vs. fixed scan position

For the population enrolled in this study, scans acquired using the default fixed-offset positioning resulted in an average position of the central slice that corresponded to 4.0% of bone length in radius and at 7.2% in the tibia (Table 2a). On average (measured as RMSD), the 4.0% and 7.2% positions were 1.1 mm and 2.2 mm offset from the standard fixed scan positions, for the radius and tibia respectively. In general, percentage RMS differences for bone parameters measured at the fixed vs. the average relative scan position were greater for radius than for tibia. In particular, in the radius, parameter variations were greatest for integral BMD and cortical bone (Ct.BMD, Ct.Th and Ct.Po). The parameter most sensitive to limb-length variability was cortical thickness at the radius, where the RMS difference between the fixed offset scan location and a relative anatomic location at 4% was nearly 20% and ranged from –19% to +67%. At the tibia, we generally observed a lower magnitude of variability, but similar patterns to the radius. The variability for apparent bone strength and load distribution at the proximal margin of the scan volume was only moderately impacted by scan positioning at both radius and tibia, however the sensitivity of load fraction at the distal end of the scan was high.

Compared to the variability for a normal reference population of young men and women (20–29 years old, Table 2b), the RMS difference in T-Score between a fixed and relative offset was moderately high for all parameters, with the exception of Tb.BMD, Tb.N, and the estimated failure load by μFEA . Positioning variability was particularly high compared to normal biological variability for cortical bone measures: the RMS difference between Ct.BMD measured at fixed and relative scan positions was equivalent to 1.0 and 0.8 SDs, for the radius and tibia, respectively, with individual differences ranging from –1.8 to +3.0 SDs. Cortical thickness, porosity (tibia), and cortical load fraction exhibited similar magnitude variances with respect to the variability of our reference population. Of the trabecular bone parameters, only trabecular heterogeneity (Tb.Sp.SD) was found to have positioning variability that was high compared to the variability of our reference data (RMS T-score difference 0.7–1.2 SDs, ranging from –1.3 to +6.0 SDs).

Relative vs. fixed scan length

For the mixed-sex population of this study a relative length of 2.5% (radius) and 1.7% (tibia) could be measured in 99.7% of individuals from a routine 9.02 mm HR-pQCT scan. Compared to the fixed-length (9.02 mm) VOIs centered at the relative offset, the measurement of these relative length VOIs centered at the same relative offset resulted in relatively small differences (Table 3). In particular, RMS differences were generally less than 1% at the tibia, except for cortical porosity (1.5%) and trabecular heterogeneity (1.7%). At the radius the RMS differences were somewhat higher for Ct.Th (2.7%), Ct.Po (4.2%), and heterogeneity (2.8%).

Literature-derived anatomically standard scan position

Based on a review of published normal population studies, we calculated the arithmetic mean of male and female average heights to be 167.5 cm (Table 1b). Using anthropometric ratios for height to limb length we estimated that the average ulnar and tibial lengths of these populations to be 28.6 and 36.8 cm, respectively. Based on these limb lengths, the corresponding mean anatomic scan position for a fixed offset HR-pQCT scan would be centered at 4.0% of ulnar length from the distal end of the radius and 7.3% of tibial length from the tibial plafond.

Bias estimate for fixed offset scanning

The progression of bias for key parameters was plotted as a function of shift away from these standard anatomic positions (Figure 3). This models increasing differences in bone length between groups or individuals being compared. With respect to mean anatomic position of 4.0% at the radius (0mm offset in Figure 3a), the difference in Tt.BMD and cortical parameters (Ct.BMD, Ct.Th. and Ct.Po) increased as the measurement VOI was shifted more proximally, and decreased negatively as the measurement VOI shifted distally. Trabecular parameters (Tb.BMD and Tb.N) followed the opposite trend: they increased as the VOI shifted distally and decreased as the VOI shifted proximally. In the tibia (Figure 3b), all parameters followed the same trend, except Ct.Po, which had the opposite progression. When evaluated with respect to body height, the measurement bias in the radius for Tt.BMD and cortical bone parameters was positive for shorter subjects and negative for taller subjects. In contrast, the measurement bias of trabecular parameters was positive for taller subjects and negative for shorter subjects. Regression equations fit to the bias curves are presented in Table 4a and provided as a function of scan offsets and differentials by limb length and height.

Discussion

In this study, we analyzed the impact of anatomic variability in bone length on HR-pQCT measurements of bone density, structure, and strength in adults. In particular, we have evaluated the difference in measurements taken at a fixed absolute offset compared to a relative %-of-total-length offset based on direct measurement of forearm or leg length. Our data demonstrates that significant measurement bias is introduced by not taking into account individual bone length for measurement localization. A smaller bias is introduced by not measuring fixed-length VOIs rather than VOIs scaled to individual bone length. Using an

approximation based on anthropometric height:limb length ratios we estimated that the average anatomical position of the traditional fixed-offset scan site from existing reference data corresponds to 4.0% in the radius and at 7.3% in the tibia. Finally, we illustrated the significance of our findings by comparing our estimate of measurement bias to differences reported in the literature for relevant population comparisons.

The principle finding of our study is that the measurement of metaphyseal bone at the default fixed distance offset is significantly biased by variability in individual bone length. In particular, cortical bone parameters, and the radius more so than the tibia, are sensitive to bone length variability. The high degree of sensitivity of cortical bone measurements to bone length is explained by the large degree of metaphyseal variability of cortical bone along the bone axis [5,6]. Using paired fixed offset and relative offset scans, Shanbhogue and colleagues [16] found similar magnitude differences. These observations are also consistent with recent data that documents high precision errors for the measurement of cortical bone due to variability in operator scan positioning [26]. With RMS differences of 20% and 13% for the radius and tibia respectively, the variability confounding measurement of Ct.Th by not accounting for limb length is comparable to clinically relevant differences observed, for example, in fracture case-control studies [38,39]. These findings strongly suggest that accounting for variability in limb length, by using anatomically standardized %-of-length offsets, will reduce a significant source of variability in cross-sectional population studies. To further emphasize the clinical significance of these findings, we used differences in T-score between fixed and %-of-length measurements to relate the observed measurement bias to biological variability. While it has been suggested that centile curves, rather than T- or Z-scores, are more appropriate for the clinical application of HR-pQCT [7], our observations suggest that meaningful interpretation of an individual HR-pQCT score will require anatomically standardized measurements, regardless of the score formulation applied. For example, on average the fixed offset measurement of Ct.BMD deviated by 1.0 SD from what would be measured at a standard anatomic %-of-length position. For the shortest and tallest individuals in our study population we found T-score deviations as large as +3.0 and -1.8 SD for Ct.BMD, respectively. We observed similar patterns at both sites for cortical bone parameters and the cortical load fraction measured by μ FEA. These observations raise concern with the use of reference data to interpret HR-pQCT outcome measures in individual patients scanned at a fixed offset position. Indeed, several published case studies of adult individuals with various skeletal abnormalities have used normative reference data to characterize their bone quality status. While some of these cases involved patients of normal height relative to their respective reference population [8,9], it is not surprising that other cases involved subjects with abnormal stature [10,11]. The interpretation of new patient scans could be made more reliable if scan protocols are established to match the mean anatomic position (%-of-total-length) of the region of interest for the reference population. By calculating outcome measures in a sliding 110-slice volume of interest within our double stack scans, we have derived curves (Figure 3) that reflect the progression of measurement bias for increasing distal and proximal shifts of the volumes of interest. The corresponding regression equations provide a means to estimate measurement bias as a function of the differential for either height or limb-length (Table 4a). To illustrate that biases introduced by variability in limb length likely have significant consequences for the

comparability of data from populations with different average limb length, we plotted bias estimates for various studies in the literature over our empirical bias curves and compared bias estimates to the observed differences reported (Table 4b). For example, gender differences in Ct.Th at the distal radius are systematically underestimated, and should be more than double the observed difference based on fixed offset measurements (from Burghardt *et al* [21], observed difference: 80 μm vs. estimated bias of -110 μm). This is consistent with the fact that, compared to women, men are measured at a relatively more distal position on average, where the cortex is more thin. In contrast, our models indicate that the positioning bias leads to the observation of lower Ct.BMD in men compared to women, whereas it would be higher if measured at anatomically comparable positions (from Macdonald *et al* [32], observed: -25.8 mg/cm^3 , estimated bias: -43.6 mg/cm^3). As seen in Table 4b, estimated biases are also significant relative to the observed differences between ethnicities with different average height. Collectively, our observations indicate that taking into account bone length is essential for reliable comparisons between populations of different stature, and raises important questions about the conclusions drawn from such comparisons reported in the literature, including from our center.

Three critical methodological factors were considered in our development of a reliable procedure for standardized anatomic positioning: (1) the external measurement of limb length; (2) the definition of the anatomic landmark at the distal end of the bone used to position the region of interest; and (3) comparability to existing reference databases. Based on internal reproducibility data, precision error for the measurement of external forearm length was 3.2 mm. For a distal radius scan positioned at 4.0% of-total-length, the precision error for the limb-length measurement translates to variability in the position of the first slice of 0.128 mm (equivalent to 1–2 slices). This corresponds approximately to one tenth of the error that biological variability in individual limb-length introduces to fixed-offset scan positioning (i.e. 1.14 mm), and less than half the error introduced by the operator positioning the reference line to prescribe the scan (0.38 mm) [26]. Thus, despite moderate imprecision in limb length measurement, the effect on the positioning variability is relatively small. The second factor is the anatomic landmark used to define the distal reference point of the bone, from which the offset is defined. For the tibia we used the traditional landmark recommended by the manufacturer; it is both reliable from an operator precision perspective [26], and anatomically consistent with the distal extent of the tibia, lengthwise. For the radius, we selected to use the medial side and proximal margin of the radial head (Figure 1e–f) as our anatomic landmark. This feature is significantly more reliable in terms of operator precision [26], approximately reflects the distal margin of the metaphysis, and is consistent with common approaches for positioning in pediatric HR-pQCT studies [14,15] and other modalities that image the distal radius [13]. For our third consideration, we decided that it was important to attempt approximate comparability with existing reference databases collected using the default fixed offset position. The estimated mean %-of-length scan positions for population studies available in the literature at the time of our analysis was calculated to determine anatomic positions that provide approximate comparability. Taken together we believe these decisions will ultimately improve reliability of HR-pQCT measurements of bone.

Based on our analysis of positioning variability, and in light of the considerations above, we propose the adoption of anatomically standardized scan locations corresponding to 4.0% of ulnar length proximal to the proximal margin of the radial head for the forearm, and 7.3% of tibial length proximal to the tibial plafond for the lower leg. It is important to note that these scan positions represent the offset from their respective anatomic landmark to the central slice of the scan. HR-pQCT scans are constrained to a discrete height for each acquisition (i.e. 9.0 mm). As a result, the scan length corresponds to variable relative coverage in terms of %-of-length, depending on individual limb length. Therefore, centering on a relative %-of-length location will lead to equal variability in the distal and proximal extent of the scan, which is preferable to variable coverage in either the distal or proximal direction alone. This also provides compatibility across successive iterations of this technology that may include changes to the scan length (e.g. 9.0 and 10.2 mm for first and second generation XtremeCT, respectively). Definition of an anatomically standard %-of-length VOI size would additionally avoid bias introduced by measuring fixed-length VOIs. This necessitates a sub-volume analysis that constrains the number of slices measured to a standard percentage of limb length. This introduces some practical challenges to the data management and reporting tools for routine use, but would be possible with vendor software changes. We compared measures for a standard 110-slice volume centered at the mean anatomic offset of our population (4.0%, 7.2%) to measures calculated for sub-volumes centered at the same anatomic position, but with equal %-of-length coverage in the distal and proximal directions (radius: 2.5% of length, tibia 1.7% of length). Our findings indicate that the measurement biases due to the length of the VOI were generally equivalent or less than scan-rescan precision errors for the corresponding parameter. These observations suggest that scaling the length of the measured VOI by individual limb length will not significantly reduce measurement bias. That said, future software updates could provide the routine ability to retrospectively output measures for %-of-length VOIs. Strengths and limitations must be acknowledged in this study. The first strength is that this is the first study to explicitly propose an empirically driven standard anatomic offset. Previous pediatric studies [14,15] and one systematic adult study [16] report the use of an anatomic relative offset based on %-of-total-length. However, none of these studies aimed to propose optimal positioning based on a methodological analysis. The second strength of this study is that we used double-length scans from which we could extract virtual measurements at both fixed and relative offsets. By evaluating sub-volumes within single acquisitions, we fixed all sources of acquisition variability, including operator reference line positioning [26] and subject motion [17]. This allowed us to systematically investigate the impact of limb-length variability on bone measurements.

The first limitation of this study is that bone length was measured superficially with a meter tape, which is less precise than radiographic measurement of bone length. Due to the current limitations of the HR-pQCT scanner, it is not possible to obtain full-length scout scans of the radius or tibia. However, we demonstrated that consequent scan localization error due to intra-operator variability in measuring limb length is small compared to the bias due to inter-subject variability in limb length. In the future, it would be ideal for standard anthropometric tools and procedures to be made available, to minimize inter-operator variability in the limb length measurement. The second limitation is that we exclusively studied older adults. While

the patterns we observed are likely similar in children and adolescents, there are clearly other considerations that apply to imaging the growing skeleton. The mean ulnar and tibial length for the population studies identified in the literature were estimated as a percentage of the reported mean heights (ulna: 16%, tibia: 22%). We acknowledge that greater accuracy is accomplished by estimating bone lengths from regression lines that are sex and ethnicity specific and account for secular trends in anthropometry [40–43]. However, to our knowledge, published regression relations for all ethnicities represented by the studies considered here, were not available, nor was it feasible to account for study populations with a racial plurality. The use of these ratios was exclusively intended to determine an approximate position that will provide approximate comparability to existing data, however direct measures of limb length are essential component of our proposed standard protocol. Finally, this study has not independently tested the comparability of measurements performed at the traditional fixed offset and our proposed %-offset positions. This would be an important question to address prior to pooling data collected with both protocols, or using reference data interchangeably. Nevertheless, the data here provide compelling rationale to collect new study data using anatomically standard positions.

In conclusion, we have documented the significant variability introduced in HR-pQCT measurements of bone quality in adults by not accounting for bone length using the default scan localization procedure. Measurement bias related to variability in bone length can seriously confound cross-sectional comparisons of populations with different height, including studies of gender and racial differences. Furthermore, they seriously compromise the clinical utility of HR-pQCT to evaluate individual bone structure status by comparison to reference databases. Based on our results, we have proposed new recommendations for anatomically standardized positioning for HR-pQCT measurements of the distal radius and tibia, corresponding to offsets of 4.0% of ulnar length and 7.3% of tibial length. Additionally, anatomically standardized VOI lengths corresponding to 2.5% of ulnar length and 1.7% of tibial length can be measured, though this has a lesser impact on measurement variability. Our recommendations both improve the reliability of HR-pQCT measurements for research and clinical applications, and permit approximate comparability to existing reference data.

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Mini-Abstract

We investigated the sensitivity of distal bone density, structure, and strength measurements by HR-pQCT to variability in limb-length. Our results demonstrate that HR-pQCT should be performed at a standard %-of-total-limb-length to avoid substantial measurement bias in population study comparisons and the evaluation of individual skeletal status in a clinical context.

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Figure 1.

(a–d) Scout view images of radius and tibia illustrating the different actual and virtual scan positions evaluated in this study. In each sub-figure, the standard-length single-stack volume (blue) is virtually extracted for bone analysis from an actual double-length two-stack volume (green), acquired at a fixed offset (radius offset: 4.99 mm, tibia offset: 17.99 mm) from the reference line (green line). (a)–(b) Fixed scan positioning. In the standard acquisition protocol, the offset from the reference line to the virtual standard-length single stack volume was constant (radius: 9.5 mm; tibia: 22.5 mm). (c)–(d) Relative scan positioning. The offset was proportional to bone length (radius: 4.0%, tibia: 7.2%). (e)–(f) For training purposes, we use the skier metaphor to illustrate the correct position of the reference line at the proximal margin of the radiocarpal joint surface of the radial head to be used for our anatomically standardized positioning protocol in the forearm: (e) first, the operator visually identifies the edge (red) of the radial carpal head where it articulates with the carpal bones; (f) the operator then imagines the trajectory of a downhill skier following this curve, until they reach the edge and fly off; the point at which the skier leaves the curve should be intersected by the reference line (green) to define the scan position.

Figure 2.

Superficial measurement of ulnar and tibial length. (a) Ulnar length. The subject's upper limb is positioned with the elbow resting on a horizontal surface, with the forearm flexed at 90 degrees against a perpendicular rigid surface. The operator measures ulnar length as the Euclidean distance between horizontal surface and styloid process at the wrist (styloid process shown in subfigure). (b) Tibial length. The subject sits in a chair, with feet on the floor and legs perpendicular to the floor. The operator measures tibial length as the Euclidean distance between the tibial plateau at the knee (top subfigure) and tibial malleolus at the ankle (bottom subfigure).

Figure 3.

Difference/bias in densitometric and morphologic measurements of the radius (a) and tibia (b), plotted (lower x-axis) as a function of distance offset from the proposed anatomically standard scan position (4.0% for radius and 7.3% for tibia). Negative offsets represent measurements performed distal to the standard position, whereas positive offsets represent shifts in the proximal direction. The plotted line represents the fit of differences measured empirically by shifting a 110-slice VOI across 220-slice volumes collected for this study: the VOI was shifted up to ± 2.13 cm (26 slices) in 0.164 mm (2-slice) increments, in the distal

and proximal directions. The top x-axis represents difference in height for normative populations reported in the literature with respect to the cumulative average height (167.4cm). Positive values indicate greater height, whereas negative values indicate lesser height. The data points indicated by an asterisk represent the height-based estimation (from the equations of Table 4a) of the difference for the respective population compared to what would be measured at the standard 4.0% and 7.3% anatomic positions.

Table 1

(a) Data, including height and limb lengths, of the subjects of this study. (b) Ten population studies selected to calculate an anatomically standardized scan position based on %-of-length. Number of subjects and mean height are averaged over all the subjects involved in the studies. Ulnar and tibial lengths were estimated using reported anthropometric proportions as 16% and 22% of population height, respectively. The estimated relative positions of the default fixed offset position are estimated for each individual study, as well as a calculated mean over all studies.

Study	Number of Subjects	Mean Height [cm]	Radius		Tibia	
			Measured Length [cm]	Measured Relative Position [%]	Measured Length [cm]	Measured Relative Position [%]
Current study	30	170.4	26.7±1.9	4.0	37.9±3.1	7.2
(a)						
Study	Number of Subjects	Mean Height [cm]	Ulna		Tibia	
			Estimated Length (16% of Height) [cm]	Estimated Relative Position [%]	Estimated Length (22% of Height) [cm]	Estimated Relative Position [%]
Boutroy et al. [27]	192	161.5	25.8	4.2	35.5	7.6
Burghardt et al. [21]	68	168.0	26.9	4.0	37.0	7.3
Chevalley et al. [28]	196	164.6	26.3	4.1	36.2	7.5
Dalzell et al. [29]	132	170.5	27.3	3.9	37.5	7.2
Hansen et al. [30]	499	172.0	27.5	3.8	37.8	7.1
Hung et al. [31]	393	155.6	24.9	4.4	34.2	7.9
Khosla et al. [15]	602	169.9	27.2	3.9	37.4	7.2
Macdonald et al. [32]	642	168.3	26.9	4.0	37.0	7.3
Nilsson et al. [33]	597	175.0	28.0	3.8	38.5	7.0
Szulc et al. [34]	920	168.2	26.9	4.0	37.0	7.3
Mean value	424	167.4	28.6	4.0	36.8	7.3
(b)						

Table 2

Percentage RMS % difference (a) and RMS T-score difference (b) of bone parameters measured at fixed vs. relative scan position for radius and tibia.

Scan Positioning Average [%]		Bone Parameter Differences %RMS Difference [%range]					Biomechanics Differences %RMS Difference [%range]				
RMSD [mm]		Tt.BMD	Ct.BMD	Tb.BMD	Ct.Th	Tb.N	Ct.Po	Tb.Sp.SD	L _{failure}	Ct.LF _{prox}	Ct.LF _{dist}
Radius											
	4.0	7.0	5.9	2.7	19.5	5.8	14.3	11.0	2.4	3.2	9.9
	1.1	[-9.8; 20.8]	[-5.7; 20.2]	[-5.73; 5.15]	[-18.6; 66.7]	[-18.3; 4.5]	[-11.9; 41.1]	[-9.8; 32.6]	[-3.4; 6.6]	[-4.0; 9.5]	[-24.51; 21.34]
Tibia											
	7.2	3.9	2.9	4.7	13.2	5.5	12.4	12.5	1.6	5.03	15.1
	2.2	[-8.0; 6.6]	[-6.5; 5.4]	[-11.6; 10.9]	[-24.2; 27.5]	[-20.4; 9.5]	[-17.5; 35.4]	[-19.7; 53.7]	[-7.8; 2.0]	[-10.0; 13.3]	[-38.2; 34.8]
(a)											
		Bone Parameter Differences RMS T-Score Difference [range]					Biomechanics Differences RMS T-Score Difference [range]				
		Tt.BMD	Ct.BMD	Tb.BMD	Ct.Th	Tb.N	Ct.Po	Tb.Sp.SD	L _{failure}	Ct.LF _{prox}	Ct.LF _{dist}
Radius											
	0.4	1.0	0.1	0.6	11.0	0.3	11.0	0.7	0.1	0.4	0.6
	[-0.7; +0.7]	[-1.4; +3.0]	[-0.2; +0.3]	[-1.0; +0.9]	[-49.0; +17.7]	[-0.7; +0.4]	[-0.7; +2.3]	[-0.7; +1.2]	[-0.5; +1.2]	[-1.5; +1.0]	
Tibia											
	0.3	0.8	0.2	0.5	1.2	0.2	1.2	1.2	0.1	0.6	1.0
	[-0.7; +0.3]	[-1.8; +1.4]	[-0.4; +0.5]	[-1.2; +0.7]	[-2.1; +3.2]	[-0.7; +0.4]	[-1.3; +6.0]	[-0.5; +0.2]	[-1.1; +1.4]	[-2.7; +2.1]	
(b)											

Table 3

Comparison of RMS % differences for bone parameters due to scan position offset (fixed vs. relative) and related to measurement volume of interest length (fixed vs. relative) for the radius (top) and tibia (bottom).

Scan Positioning	Bone Parameter Differences %RMS Difference							
	Tl.BMD	Cl.BMD	Tb.BMD	Cl.Th	Cl.Po	Tb.N	Tb.Sp.SD	
Radius								
Variability due to offset Fixed vs. Relative offset equal fixed length VOI (9.02 mm)	7.0	5.9	2.7	19.5	14.3	5.8	11.0	
Variability due to VOI length Fixed vs. Relative VOI length equal relative scan offset (2.5% of Ulnar Length)	1.1	0.5	0.9	2.7	4.2	0.7	2.8	
Tibia								
Variability due to offset Fixed vs. Relative offset equal fixed length VOI (9.02 mm)	3.9	2.9	4.7	13.2	12.4	5.5	12.5	
Variability due to VOI length Fixed vs. Relative VOI Equal relative scan offset (1.7% of Tibial Length)	0.2	0.2	0.6	0.7	1.8	0.6	1.5	

Table 4

(a) Equations to calculate estimated bone parameter biases. The variable ϵ represents the estimated measurement bias for the corresponding bone parameter, whereas the variable z , l , and h correspond to the absolute difference in stack position, bone length, or height between groups or individuals to be compared, respectively. (b) Comparison of observed differences and estimated measurement bias bone measurements reported for five cross-sectional studies selected from literature. For each parameter, we report the reported group-wise difference and an estimate of the measurement bias based on the reported differences in height between groups. The bias estimations were performed using the equations in Table 3(a) (column: Bias Estimate (Height [cm])).

Parameter	Unit	Bias Estimate (Stack Offset [mm])	Bias Estimate (Bone Length [mm])	Bias Estimate (Height [cm])				
Radius								
Tt.BMD	[mgHA/cm ³]	e=17.19 z+0.24	e=-0.95 l+1.65	e=-1.73 h+1.64				
Ct.BMD	[mgHA/cm ³]	e=30.55 z-2.85	e=-1.69 l-0.34	e=-3.07 h-0.36				
Th.BMD	[mgHA/cm ³]	e=0.02 z ² -1.12 z-0.31	e=-0.07 l ² +0.06 l	e=-0.07 h ² +0.12 h				
Ct.Th	[μm]	e=81.06 z-3.02	e=-4.49 l+3.62	e=-8.15 h+3.58				
Ct.Po [*]	[%]	e=0.01 z ³ +0.22 z ² -0.10 z+0.03	e=0.02 l ³ -0.01 l ²	e=0.02 h ³ -0.20 h ²				
Th.N ^{**}	[mm ⁻¹]	e=-4.27 z-0.24	e=0.24 l-0.59	e=0.43 h-0.59				
Tibia								
Tt.BMD	[mmHA/cm ³]	e=5.42 z-0.03	e=-1.08 l+0.34	e=-1.08 h+0.34				
Ct.BMD	[mmHA/cm ³]	e=10.48 z-0.49	e=-2.10 l+0.22	e=-2.10 h+0.22				
Th.BMD	[mmHA/cm ³]	e=-2.98 z-0.01	e=0.60 l-0.22	e=0.60 h-0.22				
Ct.Th	[μm]	e=56.27 z-2.89	e=-11.25 l+0.94	e=-11.25 h+0.94				
Ct.Po [*]	[%]	e=-0.34 z-0.01	e=0.07 l-0.04	e=0.07 h-0.04				
Th.N ^{**}	[mm ⁻¹]	e=-3.21 z-0.08	e=0.64 l-0.30	e=0.64 h-0.30				
(a)								
Study	Compared Groups	Height Difference [cm]	Tt.BMD Obs. Diff./Est. Bias [mg/cm3]	Ct.BMD Obs. Diff./Est. Bias [mg/cm3]	Th.BMD Obs. Diff./Est. Bias [mg/cm3]	Ct.Th Obs. Diff./Est. Bias [μm]	Ct.Po Obs. Diff./Est. Bias [%]	Th.N Obs. Diff./Est. Bias [mm ⁻¹]
Radius								
Boutroy et al. [27]	Caucasian vs. Asian American	4.0	-54.0/-5.3	-34.0/-12.6	-6.5/-0.6	-164.5/-29.0	1.7/-1.9	NA/1.1
Burghardt et al. [21]	Male vs. Female	14.0	0.0/-22.6	-0.0/-43.3	NA/-12.0	90.0 ^a /-110.5	0.3/15.7	NA/5.4

Study	Compared Groups	Height Difference [cm]	Tt.BMD Obs. Diff./Est. Bias [mg/cm ³]	Ct.BMD Obs. Diff./Est. Bias [mg/cm ³]	Tb.BMD Obs. Diff./Est. Bias [mg/cm ³]	Ct.Th Obs. Diff./Est. Bias [μm]	Ct.Po Obs. Diff./Est. Bias [%]	Tb.N Obs. Diff./Est. Bias [mm ⁻¹]
<i>Dalzell et al.</i> [29]	Male vs. Female	13.0	17.0/-20.9	-19.0/-40.3	37.0/-10.3	80.0 [*] /-102.4	NA/10.1	0.2/5.0
<i>Hansen et al.</i> [30]	Male vs. Female	14.0	12.0/-22.6	-25.0/-43.3	39.0/-12.0	80.0 [*] /-110.5	0.5/15.7	0.1/5.4
<i>Macdonald et al.</i> [32]	Male vs. Female	14.1	25.8/-22.8	-25.8/-43.6	NA/-12.2	21.0 [*] /-111.3	1.5/16.3	0.2/5.5
<i>Tibia</i>								
<i>Boutroy et al.</i> [27]	Caucasian vs. Asian American	4.0	-22.5/-4.0	-33.5/-8.2	-3.0/2.2	-114.0/-44.1	1.8/0.2	NA/2.3
<i>Burghardt et al.</i> [21]	Male vs. Female	14.0	0.0/-14.8	0.0/-29.2	NA/8.2	210.0 [*] /-156.6	0.8/0.9	NA/8.7
<i>Dalzell et al.</i> [29]	Male vs. Female	13.0	-14.0/-13.7	23.0/-27.1	33.0/7.6	250.0 [*] /-145.3	NA/0.9	0.2/8.0
<i>Hansen et al.</i> [30]	Male vs. Female	14.0	16.0/-14.8	-26.0/-29.2	35.0/8.2	50.0 [*] /-156.6	1.2/0.9	0.2/8.7
<i>Macdonald et al.</i> [32]	Male vs. Female	14.1	35.1/-14.9	-16.0/-29.4	NA/8.2	350.0 [*] /-157.7	1.6/0.9	0.3/8.7

(b)

* x10⁻³

** x10⁻²

Obs. Diff./Est. Bias = Observed Differences/Estimated Bias

* original data reported in [mm]