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The relationship between whole bone stiffness and strength is age and sex dependent

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ABSTRACT

Accurately estimating whole bone strength is critical for identifying individuals that may benefit from prophylactic treatments aimed at reducing fracture risk. Strength is often estimated from stiffness, but it is not known whether the relationship between stiffness and strength varies with age and sex. Cadaveric proximal femurs (44 Male: 18–78 years; 40 Female: 24–95 years) and radial (36 Male: 18–89 years; 19 Female: 24–95 years) and femoral diaphyses (34 Male: 18–89 years; 19 Female: 24–95 years) were loaded to failure to evaluate how the stiffness-strength relationship varies with age and sex. Strength correlated significantly with stiffness at all sites and for both sexes, as expected. However, females exhibited significantly less strength for the proximal femur (58% difference, $p < 0.001$). Multivariate regressions revealed that stiffness, age and PYD were significant negative independent predictors of strength for the proximal femur (Age: M: $p = 0.005$, F: $p < 0.001$, PYD: M: $p = 0.022$, F: $p = 0.025$), radial diaphysis (Age: M = 0.055, PYD: F = 0.024), and femoral diaphysis (Age: M: $p = 0.014$, F: $p = 0.097$, PYD: M: $p = 0.003$, F: $p = 0.091$). These results indicated that older bones tended to be significantly weaker for a given stiffness than younger bones. These results suggested that human bones exhibit diminishing strength relative to stiffness with aging and with decreasing PYD. Incorporating these age- and sex-specific factors may help to improve the accuracy of strength estimates.

1. Introduction

A fragility fracture is a mechanical event that occurs when a low-energy force applied to the bone, such as during a fall from a standing height, exceeds bone strength and results in structural failure (Bouxsein, 2008). Fractures occur through a process involving nonlinear material and structural behavior which leads to the accumulation of submicroscopic damage merging into a macroscopic "fatal" crack (Thurner et al., 2007). Since strength cannot be measured in situ, assessments of fracture risk rely on correlations between bone strength and surrogate indices, such as morphological traits (Ammann and Rizzoli, 2003) or results from engineering based finite element analyses (FEA) (Macneil and Boyd, 2008). Noninvasive linear-elastic estimates of strength depend on a strong association between the in situ stiffness (linear, elastic deformation) and strength (non-linear, plastic deformation and failure) (Zysset et al., 2015; Engelke et al., 2016). While some FEA models use linear and non-linear estimates to predict bone strength (Keaveny et al., 2012), those that rely on linear computational techniques may not accurately predict strength due to assumptions that ignore nonlinearities in structural behavior (Zysset et al., 2015; Engelke et al., 2016).

For tubular structures, like long bone diaphyses, stiffness is expected to correlate strongly with strength, because both measures depend on similar morphological and material properties (Kontulainen et al., 2008). Whether a similar correlation between stiffness and strength holds for fracture-prone cortical-cancellous structures (e.g., proximal femur) is not well understood. Correlations between stiffness and strength have been limited to studies conducted at the tissue-level, often for a single sex, and at the whole bone level but only for diaphyseal structures (Fyhrie and Vashishth, 2000, van Rietbergen and Ito, 2015; Jurist and Folts, 1977). Thus, the stiffness-strength relationship has been established at the tissue level but is not well understood at the whole bone level (McCalden et al., 1993; Zioupos and Currey, 1998). To

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our knowledge, no published studies tested how the relationship between whole bone stiffness and strength varies between sexes and with age at different anatomical sites in fresh-frozen cadavers.

The goal of this study was to test whether the relationship between stiffness and strength varies with sex and age. We tested the weight-bearing femoral diaphysis and proximal femur and the non-weight bearing radial diaphysis. Although men have stronger bones relative to body size compared to women (Schlecht et al., 2015), it is not known whether the relationship between stiffness and strength differs between sexes. With aging, bones become more brittle, thereby affecting crack toughening mechanisms (Zimmermann et al., 2011). Nawathe et al., (2015) reported that changing tissue-level material properties from fully ductile to brittle (i.e., no post-yield displacement) resulted in a ~40% decrease in the estimated whole bone strength (2015). We postulated that this age-related increase in brittleness, which we define as a decrease in post-yield displacement, would lead to premature propagation of the fatal crack and thus reduce whole bone strength in older bones beyond that, which is predictable from stiffness. Thus, we hypothesize that the relationship between stiffness and strength will depend on age and post-yield displacement. Knowing whether the stiffness-strength relationship varies with age and sex at multiple whole bone sites is important for refining strength estimates, which will benefit efforts aimed at reducing fragility fractures (Zysset et al., 2015).

2. Material and methods

2.1. Sample population

Fresh frozen cadaveric radii and femurs were collected from ScienceCare (Phoenix, AZ, USA), Anatomy Gifts Registry (Hanover, MD, USA), University of Michigan Anatomical Donations Program (Ann Arbor, MI, USA), and Ohio Valley Tissue and Skin Center (Cincinnati, OH, USA). The collection were primarily white male and female adults with no known or observable musculoskeletal pathologies. Paired femurs and radii were collected when possible. When paired femurs were collected, the right proximal femur and the left femoral diaphysis were prepared for mechanical testing. Table 1 shows a summary of the age distribution and the number of bones in all test groups. Body weight and height, measured at the time of death, were provided when medical history was present. Following procurement, bones were wrapped in PBS-soaked gauze and stored frozen at −40 °C.

2.2. Mechanical testing of long bone diaphyses

The proximal and distal metaphyses were embedded in square molds filled with acrylic resin (Ortho-Jet BCA, Lang Dental, Wheeling, IL, USA) using a custom alignment fixture (Fig. 1). Specimens were aligned so the anterior-posterior-medial-lateral quadrants coincided with the flat sides of the acrylic blocks. The acrylic blocks interfaced with parallel aluminum guide walls to prevent specimen rotation during testing. The diaphyses were loaded to failure in four-point bending using an Instron 8511 materials testing system (Instron, Inc., Norwood, MA, USA) (Jepsen et al., 2011). Lower loading points were positioned at 25% and 75% of bone length and upper loading points were positioned at one-third and two-thirds of the lower span length. Each sample was subjected to three pre-yield load-unload conditioning cycles before being loaded to failure at a displacement rate of 0.1 mm/s. The loading protocol was validated by testing aluminum cylinders and confirming that the derived material modulus was within 1% of textbook values. Femurs were loaded in the posteroanterior (PA) direction (anterior

Table 1
Distribution of bone samples relative to age, sex, and site.

Sex	Female			Male		
Bones	Number of samples	Mean Age ± SD (years)	Age range (years)	Sample number	Mean Age ± SD (years)	Age range (years)
Radius	19	59 ± 22	23–95	36	54 ± 23	18–89
Femur	19	57 ± 21	24–95	34	59 ± 20	18–89
Proximal Femur	40	63 ± 21	24–95	44	58 ± 19	18–89

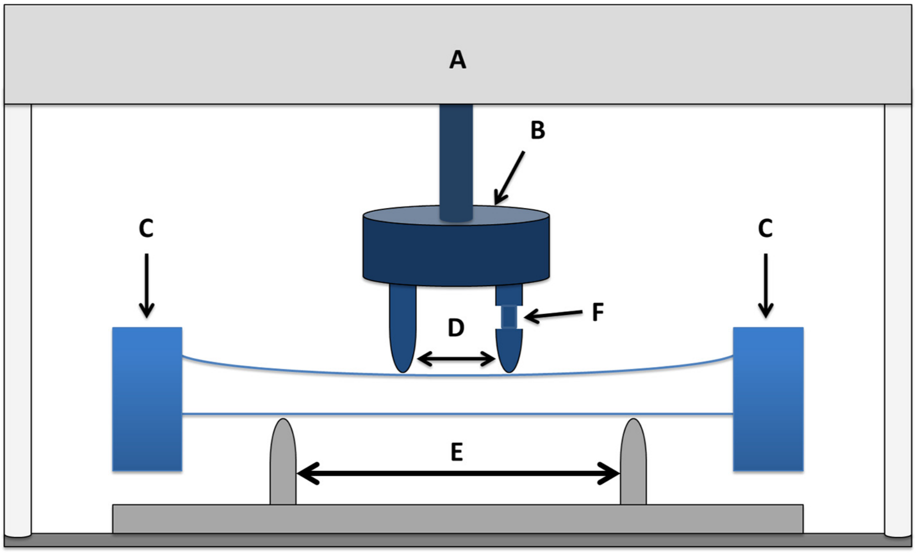


Fig. 1. Schematic of the 4-point bending testing fixture used to assess whole bone mechanical properties of the femoral and radial diaphysis. Elements of the system include the Instron 8511 material test frame (A), 10 kN load cell (B), square molds of acrylic resin used to prevent sample rotation during loading (C), upper loading points at 33% lower span length (D) and centered around lower span length, the lower span length (E) at 25% and 75% total bone length, and an adjustable upper loading point used to ensure contact at all 4 loading points along the non-uniform bone geometry (F).

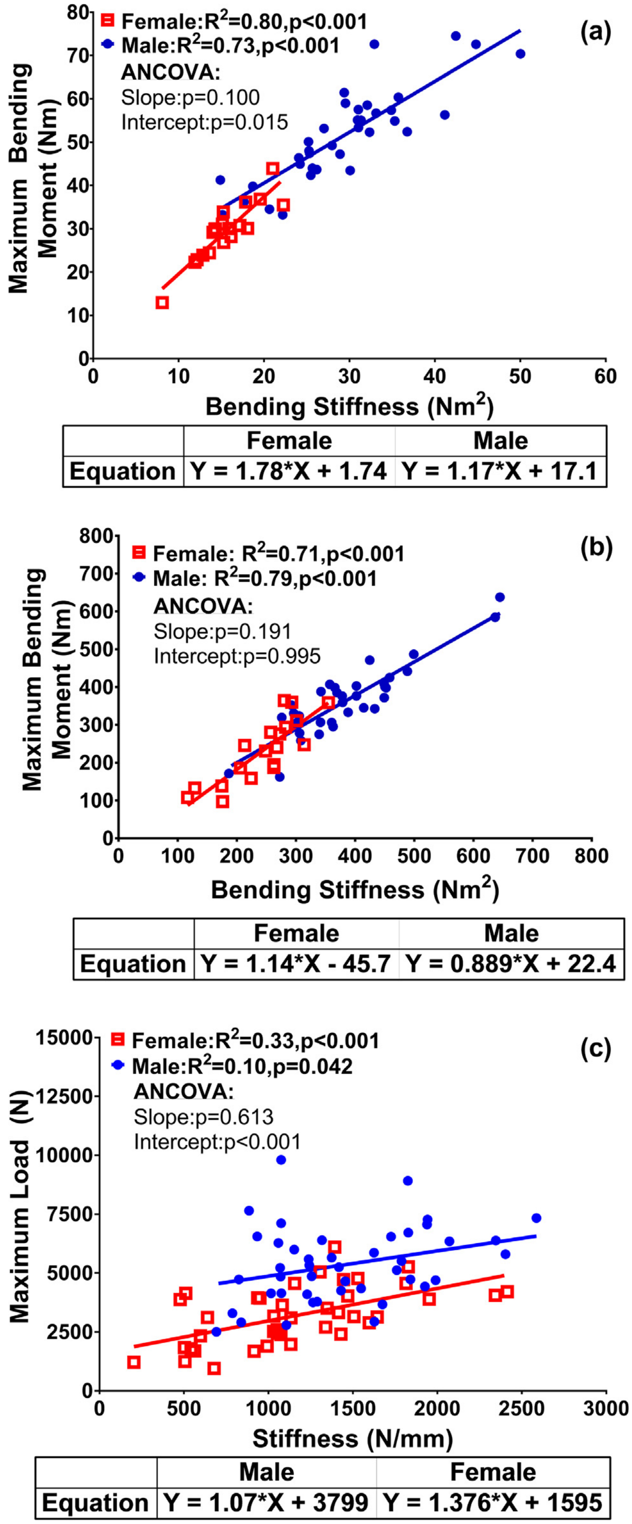
surface in tension) and radii were loaded in the medial-lateral direction (lateral surface in tension), as otherwise stated in the radial-ulnar direction (ulnar surface in tension). Loading directions were chosen to coincide with the natural curvature of the bones. Both the radial and femoral diaphyses lack symmetry so results may differ for other loading orientations. Load-displacement curves were adjusted for test fixture geometry, and used to determine bending stiffness (Nm^2), yield load (bending moment, Nm), post-yield deflection (1/m), post-yield load (Nm), and maximum bending moment (Nm) (Jepsen et al., 2011). Displacement was measured as the deflection of the upper loading points. The yield point was defined as the intersection between lines describing a 10% stiffness loss from the initial tangent stiffness with the load-displacement curve. Post-yield load (PYL) was calculated by subtracting the bending moment at yield from the maximum bending moment. Post-yield displacement (PYD) was calculated as the amount of deflection between the yield point and failure.

2.3. Mechanical testing of proximal femurs

Proximal femurs were cut 16.5 cm from the superior aspect of the femoral head. The femoral shaft was embedded in a 5 cm square aluminum channel filled with acrylic resin (Ortho-Jet BCA, Lang Dental, Wheeling, IL, USA) using a custom alignment fixture (Cody et al., 1999). Prior to mechanical testing, the proximal femurs were imaged using a nano-computed tomography system (nanotom-s, phoenix|x-ray, GE Measurement & Control; Wunstorf, Germany) (27 μm voxel size, 110 kV, 200 μA, 546 min). Morphological analyses of the proximal femurs will be conducted in future studies. To achieve a simulated fall-to-the-side loading configuration, proximal femurs were oriented with the shaft at 10° of inclination with respect to the horizontal surface and the femoral neck in 15° of internal rotation (Epelboym et al., 2012), as illustrated previously (Courtney, 1994). Custom Bondo (3M, Maplewood, MN, USA) molds were used to distribute the load applied to the greater trochanter during testing. Proximal femurs were subjected to a 100 N pre-load then loaded to failure at 100 mm/s through a metal acetabular cup that was best fit to the femoral head size. Stiffness (N/mm), yield load (N), PYD (mm), PYL (N), and maximum load (N) were calculated from load-displacement curves. A validation study, which involved indenting a rounded steel platen at 100 mm/sec into the Bondo pads, determined that the deflection attributable to the load cell and Bondo pads was 0.04 mm (0.02–0.1 mm), which accounted for 0.96% (0.56–2.2%) of the total displacement of the fractured femurs.

2.4. Statistical analysis

Whole bone strength for the diaphyses refers to the maximum bending moment, and whole bone strength for the proximal femur refers to maximum load. The relationship between stiffness and strength was assessed using a least-squares linear regression. Sex-specific differences in the slopes and y-intercepts of the linear regressions were determined by ANCOVA for each bone site. The degree to which strength varied for a given stiffness was determined by calculating the 90% prediction bands and measuring strength at the average stiffness value (Minitab 16 e-academy, Inc., State College, Pennsylvania USA). Multiple linear regression analysis was conducted to test if stiffness, age, PYD, and PYL were independent predictors of strength. Variance inflation factors (VIF) assessed if independent predictors of strength exhibited severe multicollinearity within the model. While there is no well-defined critical value to indicate severe multicollinearity, it is generally accepted that VIFs ranging from 5 to 10 signify a problem (Stine, 1995). Whole bone strength values were compared across sites using a least-squares linear regression. A regression analysis was also conducted between the residuals of the stiffness-strength regressions at different bone sites to test whether a bone that tended to be weak (or strong) for a given stiffness at one site was also weak (or strong) for a given stiffness at other sites. Sex-specific differences in the slopes and y-intercepts of these regressions were determined by ANCOVA.



	Female	Male
Equation	Y = 1.78*X + 1.74	Y = 1.17*X + 17.1

	Female	Male
Equation	Y = 1.14*X - 45.7	Y = 0.889*X + 22.4

	Male	Female
Equation	Y = 1.07*X + 3799	Y = 1.376*X + 1595

Fig. 2. Linear regressions between whole bone stiffness and strength for the (A) radial diaphysis (B) femoral diaphysis, and (C) proximal femur.

Table 2
Comparison of the maximum load for the proximal femur, femoral diaphysis, and radial diaphysis.

Bone site	Sex	Mean stiffness	Strength at mean stiffness	Minimum 90% PB	Maximum 90% PB	90% Prediction band strength range	% Range compare to average strength
Proximal Femur	F	1163 ± 509	3195	1451	4922	3471	**108%**
	M	1446 ± 460	5344	2721	7930	5209	**97%**
Femur	F	244 ± 62	232	149	313	164	**70%**
	M	386 ± 95	366	289	443	154	**42%**
Radius	F	16 ± 3	29	24	35	11	**38%**
	M	29 ± 3	52	42	61	19	**37%**

Stiffness values are given in Nm^2 for the femoral and radial diaphyses and N/mm for the proximal femur. Strength values are given in Nm for the diaphyses and in N for the proximal femur.

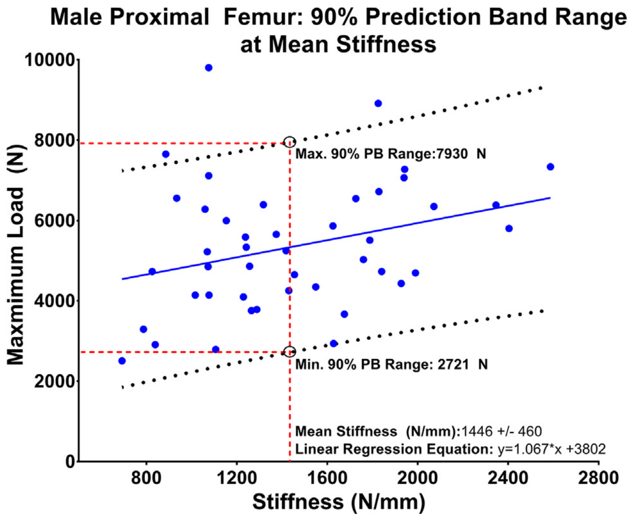


Fig. 3. Example (male proximal femurs) showing how the range in whole bone strength for a given stiffness was calculated from the 90% prediction bands (PB) and expressed relative to the mean stiffness.

3. Results

3.1. Relationship between stiffness and strength

Bone strength correlated significantly with stiffness for males and females at all three bone sites (Fig. 2). A comparison of the stiffness-strength regressions between males and females showed a significant difference in y-intercepts but not slopes for the radial diaphysis (Slope: $p = 0.100$, Intercept: $p = 0.015$) and the proximal femur (Slope: $p = 0.613$, Intercept $p < 0.001$), indicating that male radii and proximal femurs were significantly stronger for a given stiffness compared to females. Using regression equations (Fig. 2), it was determined that males were 158% ($\sim$1850 N) and 119% ($\sim$6 Nm) stronger than females for the proximal femur and radial diaphysis, respectively, when compared at the mean stiffness value for females. Although significant sex-specific differences were found for the radial diaphysis, there was limited overlap in stiffness values between sexes (15.12–21.02 Nm^2), suggesting that this particular sex-specific comparison was not appropriate. In contrast, the y-intercept and slope did not differ between males and females for the femoral diaphysis, even when the analysis was limited to the range of overlapping stiffness values (186.5–354.6 Nm^2). Thus, males and females showed a similar stiffness-strength relationship for the femoral diaphysis.

The degree to which strength varied for a given stiffness was determined for each bone site by calculating strength at the 90% prediction bands at the average stiffness (Table 2, Example Calculation: Fig. 3). The percent difference between the lowest and highest values of strength was 37–38% for the radial diaphysis, 42–70% for the femoral diaphysis, and 97–108% for the proximal femur. Qualitatively, high-resolution nanoCT images of proximal femurs obtained from young and elderly male and female donors showed large differences in bone morphology and internal microstructure for proximal femurs with similar stiffness but with a 50–100% difference in strength (Fig. 4).

3.2. The stiffness-strength relationship: age, sex, and brittleness effects

The relative contributions of stiffness, age, PYD, and PYL to bone strength were determined by conducting a multivariate regression analysis (Table 3). Stiffness remained a significant predictor of strength in all regressions, even when age, PYL, and PYD were included in the model. Age was a significant ($p < 0.05$) or borderline significant ($p < 0.1$) independent predictor of strength at all bone sites for both sexes except for the female radial diaphysis. PYD was a significant ($p < 0.05$) or borderline significant ($p < 0.1$) independent predictor of strength for both sexes at both femur sites and for female radial diaphyses. Post-yield load was a significant independent predictor for male femoral diaphyses and proximal femurs and female radial diaphyses. Adjusted R^2 values ranged from 46.9% to 91.5% among the test groups. There was only one case in which VIF values suggested a potential multicollinearity problem (Female radial diaphysis: PYL VIF = 5.647). However, this variable was not a significant independent predictor of bone strength and the impact on adjusted R-squared values was not further explored. Generalized linear models were conducted and revealed there were no significant 2-way, 3-way, and 4-way interactions among independent variables at any bone site or for either sex (data not shown). Thus, no strong interactions among the independent variables exist.

3.3. Comparing bone strength and residuals of the stiffness-strength relationship across anatomical sites

Strength correlated significantly across bone sites for the male donors (Fig. 5). These regressions were borderline significant for female donors. Regressions across bone sites were also conducted for using the stiffness-strength residuals (Fig. 6). Significant positive correlations were observed only when comparing the male radial diaphysis and femoral diaphysis ($R^2 = 0.18$, $p = 0.04$). Thus, in general, donors that tended to have low (or high) strength for a given stiffness at one site did not tend to show low (or high) strength values at other sites.

4. Discussion

Cadaveric femoral diaphyses, radial diaphyses, and proximal femurs were loaded to failure to test how sex, age, and brittleness affected the relationship between stiffness and strength. For the diaphyses, strength correlated well with stiffness, as expected for a tubular structure, with strength values varying by as much as

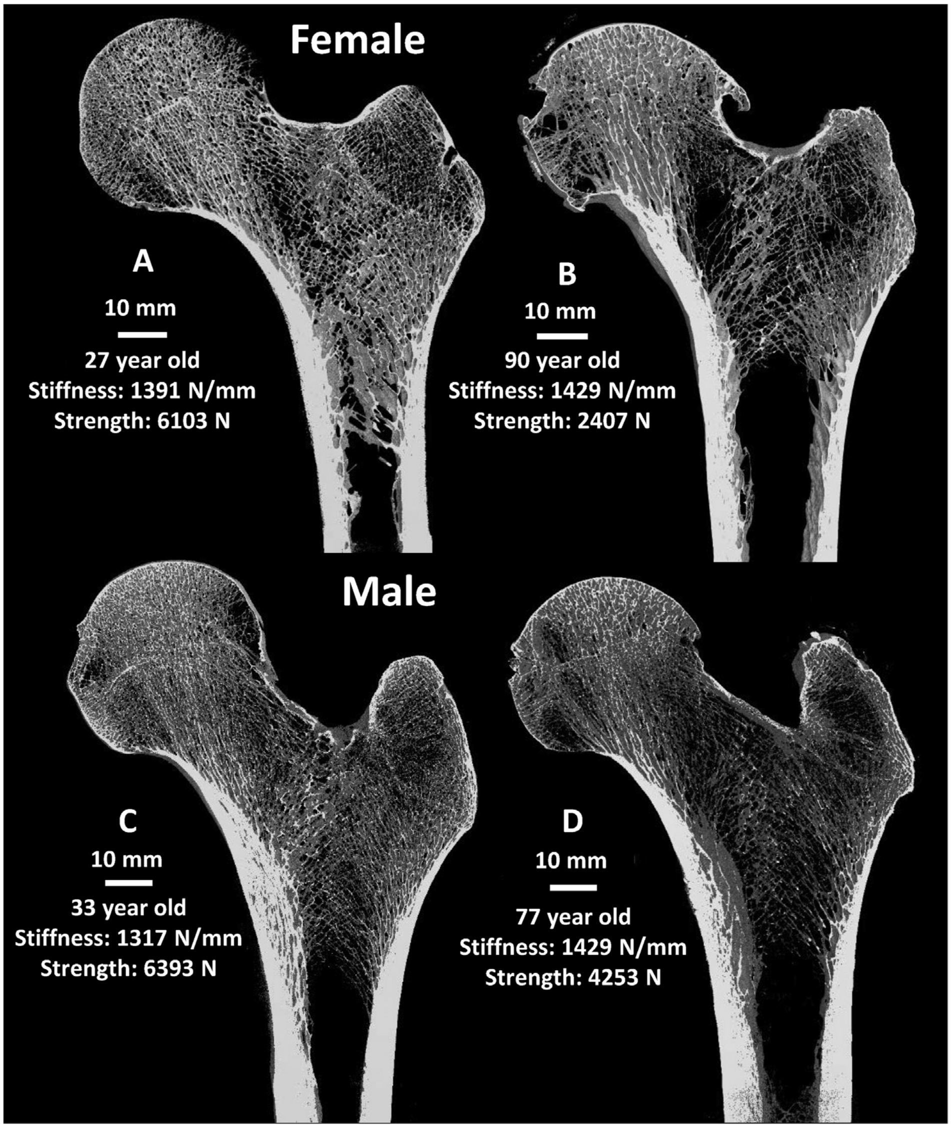


Fig. 4. Nano-CT images of proximal femurs showing similar whole bone stiffness by sex but different strength for a (A) 27 year old female [Stiffness: 1391 N/mm, Strength: 6103 N], (B) 90 year old female [Stiffness: 1429 N/mm, Strength: 2407 N], (C) 33 year old male [Stiffness: 1317 N/mm, Strength: 6393 N], and (D) 77 year old male [Stiffness: 1429 N/mm, Strength: 4253 N].

37–70% for a given stiffness. In comparison to the diaphyses, the relationship between stiffness and strength was weaker for the proximal femur, as evidenced by lower R-squared values, with strength varying as much as 97–108% (i.e., 2-fold) for a given stiffness. PYD (brittleness) and age had independent effects on the stiffness-strength relationship for all three bone sites, indicating that older and more brittle bones had a lower strength than would be predicted from stiffness alone. Finally, the relationship between stiffness and strength varied with sex for the proximal femur with males showing 158% ($\sim$1850 N) greater strength than females at matched stiffness values. To our knowledge, this is the first study to report significant sex, age, and brittleness effects on the stiffness-strength relationship of whole bones. Our findings suggest that surrogate indices of strength that rely on stiffness may be improved by adjusting for sex, age, and brittleness effects.

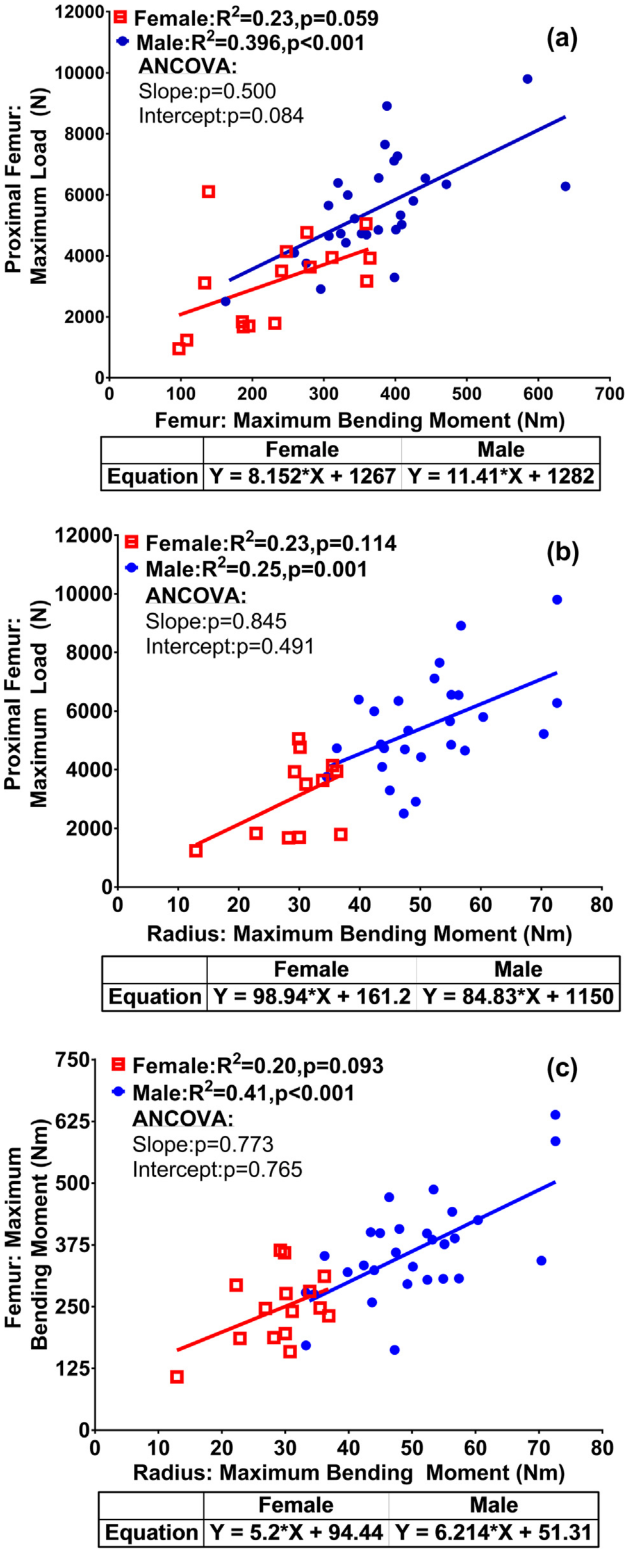
The sex-specific nature of the stiffness-strength relationship was observed for the proximal femur where males were approximately 158% stronger than females at matched stiffness values, respectively. Prior work reported sex-specific differences in bone stiffness and strength individually (Carpenter et al., 2011; Sherk and Bemben 2013; Schlecht et al., 2015), and it has been estimated that male bones are stronger relative to body size compared to female bones (Looker et al., 2001; Nieves et al., 2005; Schlecht et al., 2015). However, no studies have reported the sex-specific nature of the stiffness-strength relationship observed herein. Although it was not our goal to identify a mechanism that would explain the sex-specific differences in the stiffness-strength relationship, our results suggest that the physical bone traits that define bone stiffness may differ from those that define strength, and that these structure-function associations differ between men and women. Changes in porosity and external morphology may partially explain the decoupling of stiffness and strength. Likewise, sex specific changes in certain collagen cross-links may contribute to the decoupling of stiffness and strength (McNerny et al., 2015, Nyman et al., 2006). Variation in proximal femur strength between and within sexes may also be attributed in part to trabecular microstructural redundancy. Less microstructurally redundant bones require a smaller proportion of bone to fail, are less able to effectively transmit load, and thus are weaker (Nawathe et al., 2014; Fields et al., 2012). If females are less

Table 3
Results of a multiple linear regression analysis between whole bone strength and stiffness [Diaphysis: Nm^2, Proximal Femur: N/mm], age [years], PYD [Diaphysis: 1/m, Proximal Femur: mm], and post-yield load (PYL) [Diaphysis: Nm, Proximal Femur: N] (bold font, $p < 0.05$; italic font, $p < 0.10$).

Site	Sex	Predictive variable	Slope of coefficient (B)	Standardized slope of coefficient (Normalized B)	SE	t	P	VIF
Femur Diaphysis	Male	Constant	−20.94		49.23	−0.43	0.674	
		Stiffness	1.03	1.03	0.12	8.37	**<0.001**	3.788
		Age	−0.78	−0.17	0.30	−2.60	**0.014**	1.087
		PYD	57.58	0.25	17.74	3.25	**0.003**	1.501
		Post-Yield Load	−0.25	−0.23	0.15	−1.73	*0.094*	*4.307*
	Adjusted R^2: 86.7%							
	Female	Constant	−10.30		43.61	−0.24	0.817	
		Stiffness	0.66	0.48	0.20	3.29	**0.005**	2.991
		Age	−0.63	−0.17	0.36	−1.78	*0.097*	1.220
		PYD	37.01	0.26	20.36	1.82	*0.091*	*2.947*
		Post−Yield Load	0.54	0.32	0.40	1.58	0.137	5.647
	Adjusted R^2: 87.3%							
Radial Diaphysis	Male	**Constant**	15.46		6.11	2.53	**0.017**	
		Stiffness	1.26	0.92	0.18	7.02	**<0.001**	2.767
		Age	−0.09	−0.19	0.04	−1.99	*0.055*	*1.428*
		PYD	0.93	0.15	0.65	1.42	0.167	1.729
		Post-Yield Load	−0.015	−0.31	0.13	−0.12	0.909	2.669
	Adjusted R^2: 78.40%							
	Female	Constant	3.30		3.45	0.96	0.356	
		Stiffness	1.13	0.57	0.20	5.66	**<0.001**	2.125
		Age	−0.024	−0.80	0.02	−1.04	0.318	1.251
		PYD	0.79	0.19	0.31	2.52	**0.024**	1.175
		Post-Yield Load	0.42	0.35	0.12	3.49	**0.004**	2.154
	Adjusted R^2: 91.50%							
Proximal Femur	Male	**Constant**	5371		1077	4.99	**<0.001**	
		Stiffness	1.44	0.41	0.4098	3.5	**0.001**	1.088
		Age	−32.30	−0.37	10.87	−2.97	**0.005**	1.244
		PYD	−229.14	−0.32	95.66	−2.4	**0.022**	1.411
		Post-Yield Load	0.79	0.56	0.1869	4.25	**<0.001**	1.378
	Adjusted R^2: 46.9%							
	Female	**Constant**	3775.2		568.9	6.64	**<0.001**	
		Stiffness	1.29	0.54	0.26	4.97	**<0.001**	1.070
		Age	−28.67	−0.49	6.16	−4.65	**<0.001**	1.024
		PYD	−82.36	−0.26	35.23	−2.34	**0.025**	1.116
		Post-Yield Load	0.32	0.15	0.23	1.4	0.171	1.034
	Adjusted R^2: 57.5%							

microstructurally redundant than males, then this may explain why strength but not stiffness declines across the age-range examined. Women have an increased propensity to fracture throughout life compared to men (Wentz et al., 2011) and prior work has identified bone traits (external size, geometry, BMD, etc.) that may contribute to the increased strength indices of men compared to women (Cawthon, 2011). Our study suggested that the cumulative effect of these bone traits resulted in a stiffness-strength relationship that varied with sex.

Age, PYD, and PYL, in addition to stiffness, were significant independent predictors of strength for the proximal femur, and these variables were mostly significant at the femoral and radial diaphysis for both sexes. The adjusted R-squared values were relatively high (R^2 = 46.9–91.5%) for the multiple linear regressions, which only included measures of elastic and plastic mechanical behavior and no measures of bone morphology or tissue-level mechanical properties. This outcome indicated that older, more brittle bones tend to sustain a lower post-yield load, and thus a lower strength relative to stiffness. Previous studies examining age-related changes in bone mechanics were typically conducted at the tissue level (Nalla et al., 2006; Ding et al, 1997; Burr and Martin, 1983). Few cadaveric studies have reported age-changes in whole bone mechanical properties. Our results are consistent with prior work reporting that post-yield properties (e.g., strength, fracture toughness, post-yield strain) degrade with age (McCalden et al., 1993; Zioupos and Currey, 1998; Epelboym et al., 2012). This decrease in post-yield displacement, (i.e. increase in brittleness) reflects changes in crack tolerance of cortical and trabecular bone which may lead to premature failure and thus a proportionally lower strength of older bones (Zioupos and Currey, 1998; Nalla et al., 2006). Although most studies agree that bones tend to exhibit less PYD with age, stiffness has been shown to decrease (Courtney et al., 1995; Ding et al., 1997), not change (Burstein et al., 1976; McCalden et al., 1993; Epelboym et al., 2012), or even increase with age (Burr and Martin, 1983). Discrepancies in how stiffness changes with age among studies likely arise from differences in scale (whole bone level versus tissue-level), testing mode (compression, tension, torsion, 4-point bending), anatomic site, and/or tissue handling. Our results are in line with findings by Nawathe et al., (2015) who showed a ~40% decrease in simulated whole bone strength when tissue level post-yield behavior of bone was changed from a fully ductile to brittle behavior (2015). Our study utilized the natural variation in whole bone post-yield displacement across the adult age range to study how brittleness affected bone strength. The current study is unique because we tested a large number of samples to assess bone mechanical behavior at three different sites using consistent tissue handling methods. The clinical implication of finding that the age-related increase in brittleness may contribute to the age-related decrease in strength is that post-yield properties depend on material behavior, which are difficult to measure non-invasively. This outcome would suggest that the degree to which morphological traits can be used to predict strength becomes progressively limited with aging. Thus, determining how the relative contributions of material and



	Female	Male
Equation	Y = 8.152*X + 1267	Y = 11.41*X + 1282

	Female	Male
Equation	Y = 98.94*X + 161.2	Y = 84.83*X + 1150

	Female	Male
Equation	Y = 5.2*X + 94.44	Y = 6.214*X + 51.31

Fig. 5. Comparison of whole bone strength between the (A) femoral diaphysis and the proximal femur, (B) radial diaphysis and the proximal femur, (C) and the radial diaphysis and the femoral diaphysis. Comparison of residuals of whole bone stiffness-strength regressions between the (D) femoral diaphysis and the proximal femur, (E) radial diaphysis and the proximal femur, and (F) the radial diaphysis and the femoral diaphysis.

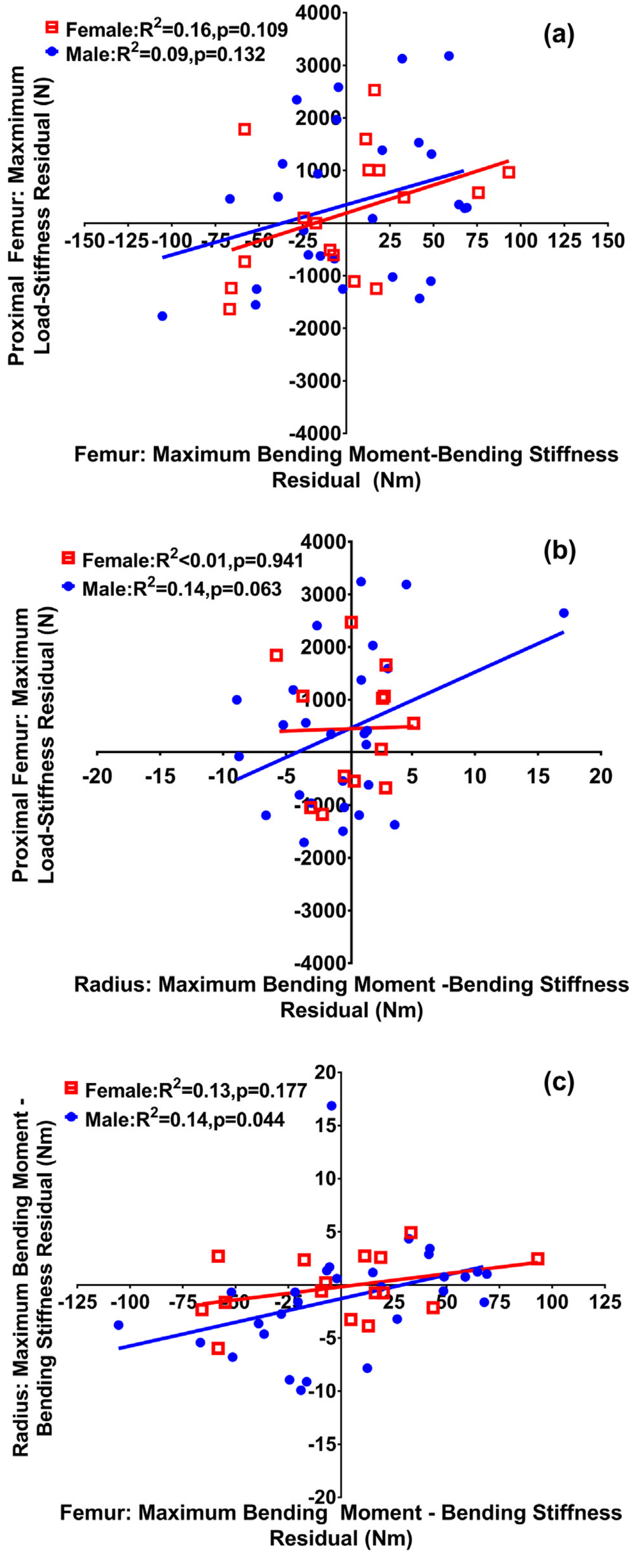


Fig. 6. Comparison of residuals calculated from the stiffness-strength regressions between the (A) femoral diaphysis and the proximal femur, (B) radial diaphysis and the proximal femur, and (C) the radial diaphysis and the femoral diaphysis.

morphological traits to whole bone strength change with age, site, and sex may benefit efforts to improve strength estimates and fracture risk.

Proximal femur strength varied as much as 97% and 108% at the mean stiffness for males and females, respectively, suggesting that bone strength may not be accurately predicted based solely on information arising within the linear-elastic range of loading (i.e., stiffness). Whole bone strength correlated across bone sites on an absolute basis but not relative to stiffness (Figs. 5 and 6), consistent with prior work (Vico et al., 2008; Liu et al., 2010; Schlecht et al., 2014). The lower R-squared values for the female bones may be partially attributed to the lower number of paired samples in this cohort. Intra-skeletal elements (cortical TMD and cortical area) are less highly correlated in females compared to males at the radial and femoral diaphysis (Schlecht et al, 2014). If bone material properties are less uniform across the female skeleton, differences in mechanical properties may be further accentuated when comparing across diaphyseal and the cortical-cancellous proximal femur. Intra-skeletal comparisons were studied to begin understanding whether peripheral sites can predict strength changes in the proximal femur. Herein, we tested whether strength correlated across sites (Fig. 5), leaving mechanistic details to follow up research. We believe that strength correlated across sites for three primary reasons: (1) body size effects (i.e., bigger people tend to have bigger, stronger bones), (2) bone morphology (i.e., bone robustness correlates across skeletal sites (Schlecht et al., 2014)), and (3) age-related changes in bone structure and material properties. For the latter factor, if bone structure and material properties change similarly similar across sites, then we would expect that bone strength would also show similar age-related declines across sites, and thereby contributing to the correlations in strength across sites. These factors remain to be teased out in future work to better understand whether peripheral bones provide a meaningful site to monitor the age-declines in strength of the fracture-prone proximal femur.

Similar correlations of whole bone strength across anatomical sites have been observed in formalin-fixed cadavers (Eckstein et al, 2002); these outcomes are limited because formalin affects bone mechanical properties (Currey et al., 1995; Ohman et al., 2008). Although we did not investigate the biomechanical mechanisms that would explain the correlation of strength across sites, similarities in stiffness measures at central and peripheral cortico-cancellous sites may be due to similarities in areal BMD, volumetric BMD, geometry, and microstructure (Liu et al., 2010). The lack of correlation of the residuals from the stiffness-strength regressions across bone sites could be attributed to the fairly narrow range of residual values for the radial and femoral diaphyses. The lack of strong correlations across bone sites between the stiffness-strength relationship suggest that age-related changes in strength and stiffness may arise through different rates of structural and material changes. Future work needs to tease out the material and geometrical contributions to whole bone stiffness and strength for both sexes to better explain the outcomes observed in this study. Clinically, this outcome would mean that site-specific strength estimates may be needed to predict fracture risk for women and that the sum of factors that affect the stiffness-strength relationship at one site may not be observed at another, despite the similarity in strength on an absolute value.

Directly measuring whole bone mechanical properties for a large cohort of cadaveric specimens is a strength of this study. However, some limitations need to be addressed. Because the cadaveric bones had no known musculoskeletal disease or injury, our donors may represent a stronger subgroup within the elderly population and thus may underestimate the declines in bone stiffness and strength with aging. Body weight and height were not available for all donors, which limited our ability to adjust for body size effects and investigate temporal trends. The proximal femur testing protocol (Courtney et al., 1995; Rezaei and Dragomir-Daescu, 2015; Dall'Ara et al. 2016) was limited to a constant loading rate for sideways falls. Although limitations such as loading condition, orientation, and rate exist for all ex vivo mechanical tests, the outcomes should provide a reasonable approximation of the in situ whole bone strength. Proximal femurs were loaded to failure at a rate that was three orders of magnitude greater than the diaphyseal sites. It is unclear how the stiffness-strength relationships would change with different loading modes. However, bones become more brittle at higher loading rates, which may partially explain the greater variation among the proximal femur mechanical properties compared to the diaphyses (Yu et al., 2011). McElhaney observed only a 12% change in compressive strength with a load increase of 300-fold suggesting that the mechanical test results observed here may not significantly differ from what would be observed in a clinical fracture (1966). Finally, relationships among material and geometrical properties were not explored, but which are needed to provide insight into the decoupling between whole bone stiffness and strength.

In conclusion, whole bone strength was impacted by stiffness and age-related declines in ductility and other age-related factors. Thus, bones appear to become weaker relative to stiffness with aging. Finally, the relationship between stiffness and strength varied between sexes for the proximal femur where males were twice as strong as stiffness-matched females. Both PYD and age affected the stiffness-strength relationship, to varying degrees, indicating that including these variables in addition to stiffness may improve estimates of whole bone strength. Future work will determine why the relationship between stiffness and strength changes with aging.

Conflict of interest

The authors have no conflicts of interest to declare.

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