Increased Density and Periosteal Expansion of the Tibia in Young Adult Men following Short-Term Arduous Training

Authors:

Izard Rachel M[a], Fraser William D[b], Negus Charles[c], Sale Craig[d] and Greeves Julie P[a]

Affiliations:

[a]HQ Army Recruiting and Training Division, Upavon, UK

[b]Norwich Medical School, University of East Anglia, UK.

[c]L-3 ATI, Simulation, Engineering, and Testing, San Diego, CA, USA.

[d]Sport, Health and Performance Enhancement Research Centre, Nottingham Trent University, UK.

Corresponding author:

Dr Julie P Greeves

Army HQ, Marlborough Lines, Andover, Hampshire, SP11 8HJ

England

Email: Julie.Greeves143@mod.uk Tel: +44 1264 886827

Abstract

Purpose: Few human studies have reported early structural adaptations of bone to weight-bearing exercise, which provide a greater contribution to improved bone strength than increased density. This prospective study examined site- and regional-specific adaptations of the tibia during arduous training in a cohort of male military (infantry) recruits to better understand how bone responds *in vivo* to mechanical loading.

Methods: Tibial bone density and geometry were measured in 90 British Army male recruits (ages 21 ± 3 y, height 1.78 ± 0.06 m, body mass 73.9 ± 9.8 kg) in weeks 1 (Baseline) and 10 of initial military training. Scans were performed at the 4%, 14%, 38% and 66% sites, measured from the distal end plate, using pQCT (XCT2000L, Stratec Pforzheim, Germany). Customised software (BAMPack, L-3 ATI) was used to examine whole bone cross-section and regional sectors. *T*-tests determined significant differences between time points ($P<0.05$).

Results: Bone density of trabecular and cortical compartments increased significantly at all measured sites. Bone geometry (cortical area and thickness) and bone strength (i, MM_i and BSI) at the diaphyseal sites (38 and 66%) were also significantly higher in week 10. Regional changes in density and geometry were largely observed in the anterior, medial-anterior and anterior-posterior sectors. Calf muscle density and area (66% site) increased significantly at week 10 ($P<0.01$).

Conclusions: *In vivo* mechanical loading improves bone strength of the human tibia by increased density and periosteal expansion, which varies by site and region of the bone. These changes may occur in response to the nature and distribution of forces originating from bending, torsional and shear stresses of military training. These improvements are observed early in training when the osteogenic stimulus is sufficient, which may be close to the fracture threshold in some individuals.

Key words: Key words: pQCT, Exercise, Loading, Density, Geometry, Tibia

Introduction

Bone is sensitive to its mechanical environment and alters its shape and architecture to improve biomechanical strength and prevent structural fatigue [1]. These changes are achieved through the complex orchestration of bone modelling/remodelling [2-4], partly mediated by osteocyte signalling [5,6] Studies using the rat ulna have unravelled the osteogenic stimuli of mechanical loading, confirming that bone is most responsive to dynamic loading [1, 7], high strain rates [8-10] unfamiliar strain distributions [11], and to discrete rather than continuous bouts of loading [12,13].

The findings from animal models are corroborated, to a certain extent, in human studies that report increased areal bone mineral density following high impact exercise [14-17] Areal BMD is not a suitable surrogate measure of bone strength with exercise interventions [18] since animal studies report large changes in bone strength despite only modest changes in density [1, 19]. This is supported in cross-sectional studies of athletes using peripheral Quantitative Computed Tomography (pQCT), which describe a thicker cortex in the playing arm of tennis players [20], the tibia of triple jumpers [21] and in athletes from impact sports [22] compared with matched controls, with little or no differences in density [23].

Short-term exercise intervention studies examining the human tibia in young adults using pQCT have not detected changes in bone geometry, reporting only modest increases in trabecular density of the distal site following 8 weeks of weight-bearing aerobic exercise training [17], whereas significant improvements in bone structure are observed in the rodent ulnar following only 5 weeks of axial loading [1, 9, 24]. Differences in the magnitude and pattern of adaptations might be due, in part, to the unnatural loading model of the ulnar or to extrapolation across species. New bone formation is directly proportional to strain rate [9, 10] and variation in loading stimuli may account for differences between studies. Twelve weeks of initial military training, characterised by variable, dynamic and high impact activities, increased bone volume and whole bone cross sectional area of the femur in male recruits [25]. Other military training interventions have shown decreased areal BMD at the lumbar spine [14] and increased trabecular separation at the calcaneus [26], suggesting that early adaptations to exercise are site-specific, localised to sites of highest loading [27, 28].

The tibia experiences high loading during human locomotion [29]. The high trabecular content and larger cross sectional area at the distal site (4% of endplate) [30] is designed to *resist* high axial compressive forces resulting from impact, internal muscle forces, and to some extent posterior shear force generated throughout most of stance [31]. The primary forces experienced along the diaphysis result from the bending and torsional loads of eccentric muscle action and impact with the ground, resisted by its tubular structure and thicker outer cortical shell [30]. The loading is also likely to be highest at regions of peak compressive forces and, to a lesser extent, tensile forces [24] but no investigations have performed a prospective detailed examination of the tibia in response to an arduous training programme. This is clinically important because the tibia is susceptible to stress injuries early in military training [32], which typically present at the posterior border of the distal third [32-34]. The primary purpose of this study was to examine site- and regional-specific adaptations of the tibia to arduous training in a cohort of male infantry recruits to better understand how bone responds *in vivo* to mechanical loading.

Materials and Methods

Participants

One hundred and thirty six British male recruits volunteered to participate in week one of initial military training at the Infantry Training Centre, Catterick (ITC(C)). Forty-six participants were removed from training before their follow-up measurements in week 10 for reasons including voluntary discharge (32%), unsuitability for Army service (32%), medical discharge (13%) and voluntary exit from the study (23%). There were no significant differences in baseline body mass and bone density and geometry between participants dropping out and those completing the study. Complete datasets were available for 90 participants (age 21 ± 3 y, height 1.78 ± 0.06 m, body mass 74.0 ± 9.8 kg, tibial length 382 ± 24 mm). All recruits were free from medical illnesses or existing musculoskeletal injury, confirmed at the Army initial medical examination, and provided written informed consent prior to participation. The study was approved by the UK Ministry of Defence Research Ethics Committee (MODREC 0824/179).

Experimental Design and Procedure

Measurements were obtained at the same time point in week one, before physical training commenced, and week 10 of initial military training. The purpose of this training is to improve physical fitness and teach recruits basic military skills. In a 10-week period, infantry recruits typically undertake 27 periods of military drill, 45 physical training periods comprising of 8 continuous runs, 9 loaded marches (starting at 2 miles carrying a 10kg backpack building up to 5 miles carrying a 20 kg backpack), 6 swimming sessions and 22 40-min periods of military specific fitness (obstacle course, circuit training, and steeplechase run). Loading rates of British Army drill have been described by [35]. Additional skeletal loading is generated by frequent transits walking or marching around the camp between lessons and to meals. Steady state runs and circuit training were performed in Hi-Tec® 'Silver Shadow' standard issue Army trainers and all other physical training and transits were conducted in leather combat assault boots issued on arrival at ITC(C).

Anthropometric measurements

Height (m) was measured to the nearest 5mm using a portable stadiometer (Seca 225, Seca Ltd, Birmingham UK) and body mass (kg) was determined using a digital scale accurate to ± 0.1kg (Seca 770, Seca Ltd, Birmingham UK). For all measurements recruits wore light clothing and no footwear. Body composition was estimated from skinfold thickness measured at four sites (biceps, triceps, supra-iliac, and sub-scapular) with Harpenden skinfold callipers (Harpenden, UK) by the same investigator (RI). Percentage body fat was calculated from the skinfold measurements using previously established methods [36]. Tibial length (mm) was measured as the distance from the distal aspect of the medial malleolus to the medial joint line. Aerobic fitness was estimated from a 1.5-mile maximal run effort performed as part of the British Army physical fitness test assessment.

Injury classification

Tibial injury was classified as an injury caused during training that resulted in a visit to the medical centre and one or more days of light duties.

pQCT measurements

Peripheral QCT (XCT2000L, Stratec Pforzheim, Germany) was used to assess volumetric BMD, bone geometry and bone strength of the dominant leg at the distal (predominantly trabecular bone) and diaphyseal (predominantly cortical bone) sites of the tibia. Participants were seated comfortably with their lower leg extended through the scanning cylinder and were asked to remain still for the duration of the scanning procedure (10-15 min). All scans were undertaken within the framework for the safe use of radiation outlined in the Ionising Radiation Regulations and the Ionising Radiation (Medical Exposure) Regulations (IRMER). The effective dose of radiation to participants undertaking this study was 6 μSv, equivalent to one European flight, as determined by the manufacturer.

Initial scout scans were conducted at a speed of 40 mm·s^{-1} to identify the distal end plate of the tibia, following which scans of single axial slices (2.2 mm thickness, voxel size 0.5 mm, measure diameter 140 mm) were taken at a translation speed of 20 mm·s^{-1} at 4, 14, 38, and 66% distances of the approximate segment length, proximal to the distal endplate of the tibia. A quality assurance calibration check was undertaken on the morning of each data collection day by scanning standard phantoms with known densities of 168.5 (±0.20), 317.3 (±0.32), and 462.4 (±0.44) mg·cm^3.

The Bone Alignment and Measurement Package (BAMPack) software (L-3 ATI, San Diego, CA) was used to calculate whole and regional (60^o sectors) bone parameters using previously described methods and calculations [37, 38]. Briefly, BAMPack uses thresholds to determine boundaries, whereby contiguous voxels above 800 mg·mm^3 are considered cortical, and below 600 mg·mm^3 trabecular, bone. Voxels with density values between 600 and 800 mg·mm^3 are ignored to carefully delineate between cortical and trabecular regions. From the anterior aspect of the bone, moving in a clockwise direction, the regions are labelled in the following order: anterior (A); lateral-anterior (L-A); lateral-posterior (L-P); posterior (P); medial-posterior (M-P); and medial-anterior (M-A). Calculated parameters included: trabecular density and area (Tb.Dn and Tb.Ar); cortical density, area and thickness (Ct.Dn, Ct.Ar and Ct.Th); periosteal and endosteal perimeter (Ps.Pm and Es.Pm); anterior–posterior (AP) and medial–lateral (ML) widths; bone mineral content (BMC); and total area (Tt.Ar). The followingbone strength indices were also calculated: bone strength index (BSI); mass moment of inertia (MM_i); polar moment of inertia (i); and polar moment of inertia in the anterior–posterior ($_iAP$) and medial–lateral ($_iML$) planes. Only images with minimal motion artefacts (image quality >2) and alignment error (Root Mean Square of difference in the outer boundaries (<0.4mm)) were used for statistical analysis at each site. This alignment error criterion of <0.4mm, validated in 11 cadaveric tibia [38], captures average radius changes of approximately ±3%, based on the following equation:

Change in Area = $\pi(a_P^2 - a_0^2)$, where baseline radius = a_0, and post-exercise radius = a_P. An alignment error of ±0.4mm corresponds approximately to a change in area of ±40mm^2.

Repeatability studies using pQCT have typically reported a coefficient of variation (CV) at the tibia of <2% for total and Tb.Dn, and <1% for Ct.Dn [38, 39]. Using BAMPack we have achieved a CV of ≤1% for the test-retest measurements of Tb.Dn and Ct.Dn and geometry in a sample of $n = 31$ on two separate occasions, seven days apart (*unpublished data*).

Statistical analysis

All data are reported as mean ± 1SD. Descriptive statistics were performed on all variables and data were checked for assumptions of normality. Differences in normally distributed variables were calculated using Paired-samples *T*-tests, and data not normally distributed (Tb.Ar at 4% and CrtDn at 14, 38 and 66%) were analysed using the Wilcoxon-signed rank test. All statistical analyses were conducted on the Statistical Package for Social Sciences (SPSS) v 19.0; statistical significance was accepted at $P \leq 0.05$.

Results

Body fat (%) decreased during training (Week 1: 15.5 ± 3.8%; Week 10: 14.5 ± 2.8%; $P<0.001$), but no significant change in body mass was observed (Week 1: 74.9 ± 8.5 kg; Week 10: 75.6 ± 7.1 kg), indicating an increase in fat-free mass. 1.5-mile run time decreased during training (Week 1: 609 ± 46 s; Week 10: 576 ± 38 s; $P<0.001$). Calf muscle density (Mus.Dn) and area (Mus.Ar) increased from Week 1 (Mus.Dn: 75.6 ± 1.6 mg.cm^3; Mus.Ar: 8179.5 ± 834.9 mm^2) to Week 10 (M.Dn: 76.1 ± 1.5 mg.cm^3; M.Ar: 8521.7 ± 923.4 mm^2, *P*<0.01). Six participants (7%) suffered a tibial injury and had significantly lower Tb.Dn, Tt.Ar (14% site), Tt.Ar (38% site), Ct.Ar, Tt.Ar and BMC (66% site), and higher Ct.Ar (66% site) than non-injured counterparts. Differences in i, MMi, and BSI were also detected. Mean (+/-1SD) data for all measured sites of the tibia at Baseline and at Week 10 are shown in Table 1 (volumetric density and geometry) and Table 2 (bone strength). Regional results are provided in Table 3.

Bone Density

Trabecular bone

Trabecular density of whole bone cross-section increased at the distal tibia (4% site) following training (*P*<0.001) and regional increases in Tb.Dn were observed in all regions (*P*<0.05), with the exception of P (Figure 1A). The most significant changes in Tb.Dn were shown in the A, M-A and L-A regions (*P*<0.001). There was no significant effect of training on Tb.Dn at the 14% site.

Cortical bone

Cortical density increased at all sites along the tibial diaphysis (14%, 38% and 66% sites, *P*<0.01; Figure 1B-D). Increases at the 14% site were localised at the L-A, M-A (*P*<0.01) and P (*P*<0.05) regions. At the 38% site, regional analysis showed significant increases in the M-A (*P*<0.01) and P (*P*<0.05) regions and approached significance in the A region (*P*=0.057). At the 66% site there were increases in Ct.Dn in the A (*P*<0.01) and M-P (*P*<0.05) regions.

Bone Geometry

Area

Tt.Ar increased significantly at Week 10 of training at the 14% ($P<0.05$) and 38% ($P<0.01$) sites, resulting in a concomitant increase in area to length ratio (14%: $P<0.05$; 38%: $P<0.001$). At the 14% site, there was an increase in Tb.Ar ($P<0.01$), with Ct.Ar approaching significance ($P=0.059$). Significant increases in Ct.Ar were observed at the tibial diaphysis (38%: $P<0.01$; 66%: $P<0.001$).

Cortical thickness

Ct.Th at the 14% site did not change from baseline, although regional analysis revealed a significant increase in M-A ($P<0.01$). Training significantly increased Ct.Th at the 38% site ($P<0.001$) in all regions, with the exception of L-P, and lower baseline Ct.Th was associated with a greater change at Week 10 (-0.202, $P<0.05$). There was no significant change in Ct.Th at the 66% site.

Bone diameter

Tibial diameter increased significantly in both A-P and M-L axes at the 38% site, and in the A-P axis at the 66% site ($P<0.05$), resulting in an increase in periosteal perimeter (38%: $P<0.001$; 66%: $P<0.05$). There was no significant change in endosteal perimeter at any measured site.

Bone Strength

Moment of Inertia and Mass Moment of Inertia

All measures of bone strength, including i ($_i$AP and $_i$ML), MM$_i$ (MM$_i$AP and MM$_i$ML) and the resultant BSI, increased during training at the 14% and 38% sites ($P<0.001$). At the 66% site, i, $_i$ML, MM$_i$AP and MM$_i$ML, and BSI increased significantly with training ($P<0.001$) but $_i$AP did not change ($P>0.05$).

There were significant increases in bone density (Tb.Dn and Ct.Dn) at all measured sites, and bone geometry (Ct.Ar and Ct.Th) and bone strength (i, MM_i and BSI) at the 38 and 66% sites. Changes were predominantly observed in the A, M-A and A-P sectors.

Discussion

We examined site- and regional-specific adaptations of the tibia following 10 weeks of military training in young adult men using pQCT. Our findings demonstrate increased Tb.Dn at the distal tibia, and increased Ct.Dn and periosteal bone formation of the diaphysis, contributing to overall improved bone strength. No other study has characterised such anabolic effects of a short-term exercise intervention in humans, and the regional adaptations shown has enhanced our understanding of the pattern of load distribution from gravitational and muscle forces in response to *in vivo* mechanical loading. We observed similar gains in trabecular (metaphyseal) and cortical (diaphyseal) bone with loading, but disuse studies demonstrate greater losses in both compartments over the same timeframe, with earlier decrements of cortical bone [41].

No structural changes were observed at the distal tibia. This site is subject to predominantly compressive forces as indicated by its maximum circularity and lowest cortical thicknesses along the tibial diaphysis [30]. The difference in trabecular number and thickness of the ultra distal tibia between athletes of different sports [42] also suggests a possible contribution of trabecular microarchitecture to compressive bone strength with mechanical loading, which we could not directly assess using pQCT (due to its in-planar resolution of 0.4 mm). The increase in trabecular density is consistent with other studies, and is recognised to be an early adaptive response to mechanical loading [38]. Whilst important for shock absorption, increased trabecular density is not considered to be an important adaptation for biomechanical bone strength since higher strain rates are experienced by the tibial diaphysis under loading [43]. Analysis of regional sectors reveals fairly uniform increases in density, but we observed no change at the posterior region, which questions the short-term osteogenic contribution of shear forces during weight-bearing activities.

The significant increase in Ct.Dn at the diaphyseal sites (14, 38 and 66%) is in contrast to cross-sectional studies reporting no differences in volumetric Ct.Dn between athletic populations and controls [21, 23] or between the playing and contralateral limb in tennis players [44]. Changes in Ct.Dn have not been demonstrated with short exercise interventions [38] or long-term participation in high impact sports [23]. Increased cortical density is not perceived to be an exercise-related adaptation, possibly because Ct.Dn of young adults is optimised following growth, limiting further mineralisation [45,46]. Increased Ct.Dn was localised to the anterior and posterior sectors, with subtle differences between each diaphyseal site, reflecting regions of highest peak strains applied under axial loading, notably the anterior cortex under tensile, and the posterior surface under compressive stress [47].

Mineralisation of cortical bone increases stiffness and whole bone strength in bending [48], but ‘stiffer’ bones are more brittle, more prone to accumulation of microdamage [49], and are less able to withstand *repeated* loads. Bone tissue is brittle in narrower tibia, supporting the notion that increased Ct.Dn is a compensatory mechanism for a structurally weaker bone, as shown in male cadaveric specimens [50], *in vivo* in women [37], and in stress fracture cases [51,52]. In support of these previous findings, volumetric Ct.Dn was inversely related to periosteal (and endosteal) perimeter at Baseline, indicating that those with lower cross-sectional area had higher Ct.Dn.

An important finding of this study was the significant increase in Ct.Ar resulting from periosteal bone formation and cortical thickening of the tibial diaphysis, which confers a structural advantage to the tibia as periosteal expansion places the cortex further from the neutral axis to improve strength in bending [56] Similar architectural features are reported in the playing arm of tennis players [15, 20], in the tibia of triple jumpers [21, 55), and in runners compared with non-exercising controls [58, 59]. We detected no change in endocortical perimeter consistent with military training studies of the femur using MRI [60].

The changes in bone density and structure enhanced compressive (BSI) torsional (i) and bending ($_{i}$AP, $_{i}$ML) stiffness of the tibial diaphysis, and demonstrates that military training is not only anabolic to bone but possibly yields clinical benefits by *protecting* the tibia from the risk of fracture [61, 62]. However, this is not the case in individuals who begin training with a narrower tibial diaphysis, as indicated by lower CSA at the 38% site in our participants suffering tibial injury and in stress fracture cases of Royal Marine trainees [63]. The 38% site corresponds to the narrowest point of the tibial shaft (Table 1) [51] with lowest lateral bending strength [61] and high compressive and tensile stresses experienced during human locomotion [47].

We have shown that bone alters its shape and mineral composition to withstand the loads engendered by military training, and based on animal and some human studies we can propose, but cannot confirm, the osteogenic characteristics initiating new bone formation. Dynamic loading signals new bone formation [7], but the prevailing view is that the load must be high enough to initiate this response [64]. Military activities that are not performed often, but likely exceed the strain threshold, include downhill running and/or zigzag motions, which elicit up to 2000 microstrain at the tibial shaft [65], or foot drill which generates peak vertical forces up to 6.6 (±1.7) times body weight or 983 (±333) BW·s^{-1} [35]. The loading profile of other activities performed periodically such as gym sessions have not been measured, but may also contribute to high load, low frequency osteogenic events. Irregular orientation of loads can also augment the skeleton, even at relatively low magnitudes [66, 67], and cross sectional studies have shown that athletes engaged in sports such as soccer have significantly higher cortical area and periosteal circumference, but not volumetric BMD, of the tibia than those undertaking resistance training or in controls [42]. Further work to quantify the kinetic and kinematic characteristics of military training is required to help discern the osteogenic stimuli *in vivo* to enhance bone strength.

This study benefits from a large, homogenous population of young healthy men undertaking the same training under matched conditions. The main limitation is the lack of a control group, but we do not believe that changes in bone density and geometry are due to methodological errors for a number of reasons. Firstly, BAMPack is designed to minimise partial volume effects that may occur at the periosteal and subperiosteal boundaries by peeling away boundary voxels. Secondly, the algorithm for edge detection and boundary voxel stripping was the same for all participants at all time points, and other studies using BAMPack with similarly large sample sizes have shown no changes in cortical area [38]. Finally, the BAMPack alignment algorithm improves repositioning accuracy by excluding results with a root mean square of radial differences that exceed 0.4mm. Even though changes in bone density and area were small (~1%), we believe that these changes reflect mechanical adaptations rather than those associated with methodological errors.

In summary, *in vivo* mechanical loading improves bone strength of the human tibia by increased density and periosteal expansion, which varies by site and region of the bone. These changes may occur in response to the nature and distribution of forces originating from bending, torsional and shear stresses of military training. These improvements are observed early in training when the osteogenic stimulus is sufficient, which may be close to fracture threshold in some individuals.

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Authorship:

All authors contributed to the preparation of this manuscript. JG, RI, WDF and CS were responsible for the concept and design of this study. RI performed the data acquisition and data analysis. CN provided the BAMPack software and analysed the regional sectors. RI, CN and JG interpreted the results and accept responsibility for the integrity of the data analysis. JG oversaw the study.

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