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14. ABSTRACT The purpose of the project is to investigate whether a history of physical training with multi-axial loading (e.g. soccer) is associated with increased tibia mechanical strength and if a stronger tibia developed in soccer is resilient with load carriage. The first aim of the study is to examine the effect of past multi-axial physical training on tibia bone strength. The second aim is to examine whether a stronger tibia from past multi-axial physical training will exhibit less deformation during incremental load carriage. The third aim is to examine the relationship between tibia bone quality and the level of bone deformation during load carriage. 20 recreational female soccer players and 20 age, height, and weight matched sedentary women were recruited. pQCT were performed at various tibia sites. Motion capture, ground reaction force, and electromyography were collected during load carriage. CT scans of the tibias were collected. Subject-specific musculoskeletal models and tibia finite element models were created. Multi-body dynamic simulations and finite element analysis were conducted to examine tibia deformation during incremental load carriage. Within this final report, information concerning adherence to work objectives, research progress is presented.					
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INTRODUCTION

Due to the enormity of the tibia stress fracture (TSF) problem among female recruits in the military it is vitally important to develop a better understanding of how conditions experienced by female military personnel affect their risk for TSF. The overall purpose of this study is to determine whether history of physical training with multi-axial loading (e.g. soccer) is associated with increased tibia mechanical strength and if a stronger tibia developed in soccer is resilient with load carriage and TSF.

This research focuses on the effects of physical training history with multi-axial loading (e.g. soccer) on tibia mechanical strength and mechanical variables associated with increased risk of TSF in conditions encountered during basic training (e.g. load carriage). Two groups of participants were recruited. Group one consisted of female participants who have participated regularly in a minimum of two years of recreational soccer. Group two were age, body mass, body height matched sedentary women who did not regularly engaged in exercise with moderate intensity. Motion capture, ground reaction force, electromyographic (EMG), peripheral quantitative computed tomography (pQCT), and computed tomography (CT) data were collected. Participants' specific musculoskeletal and finite element models will be created. These models will be used to determine the tibia strains and strain rates during loaded walking conditions. It is the high mechanical loading and strains experienced by the tibia bone that ultimately leads to TSF. Thus, the immediate significance of this research is the determination of how a multi-axial loading history (e.g. soccer) affects tibia strength and tibia strains produced during militarily relevant conditions (load carriage). The results of this study can be used to evaluate the relative increases in bone strain and potential risk of TSF due to carrying additional loads. By understanding how physical training history (e.g. soccer) influences bone strength and strains, the appropriate measures can be taken in the future to minimize the risks to female recruits. This could be in form of altering load carriage, training programs, or a combination of the two.

The comprehensive scope of the proposed research will contribute significantly to the scarcity of published literature on in-vivo tibia strains and strain rates during activities consistent with what are required for female military personnel. These data are needed in order to develop a better understanding of physical training history with multi-axial loading on bone health and the dose-response relationship between loading conditions and bone's mechanical responses.

BODY

Tibia stress fracture is a common overuse skeletal injury in the military [1-3]. Female recruits face a higher risk of the injury than their male counterparts [1,4,5]. It is vitally important to develop a better understanding of how the physical conditions experienced by female recruits during basic training affect loading of the skeleton, and further, the influence of a history of multi-axial loading in attenuating loading of the skeleton during training-related activity. In order to assess the effect of multi-axial physical training history on tibia loading, two groups of female subjects, soccer players, and sedentary individuals will complete a loaded walking protocol. The loaded walking task will be analyzed using traditional gait analysis techniques in conjunction with subject-specific musculoskeletal modeling and FEA protocols in order to determine if differences in tibia strains and strain rates exist between the two groups. Such research will provide important information that may help identify novel countermeasures for stress fracture prevention in military recruits.

Therefore, The purpose of the project is to investigate whether history of physical training involved multi-axial loading is associated with increased tibia mechanical strength, and if a stronger tibia developed in physical training (soccer) signified by multi-axial loading is resilient to walking tasks with load carriage.

Summary of Methods

In order to determine the effects of multi-axial physical training history on tibia bone quality and tibia's mechanical responses to load carriage, traditional kinematic and kinetic analyses derived from motion capture and force plate data will be combined with a participant-specific finite element model of the tibia utilizing customized musculoskeletal modeling software and FEA (Figure 1).

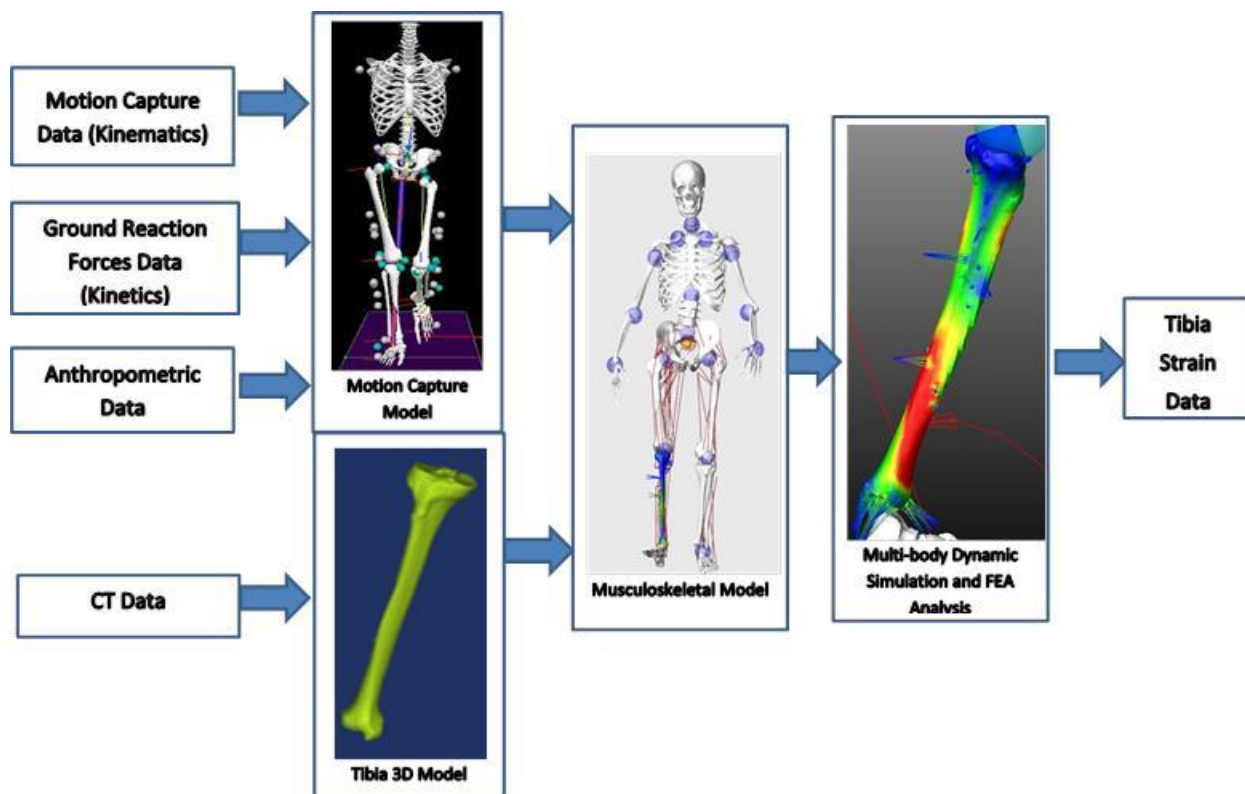


Figure 1. Diagram of project workflow

In order to test the research hypotheses that 1) Recreational female soccer players will have greater tibia bone density and more favorable tibia structural properties than those of sedentary women, 2) Recreational female soccer players will exhibit less tibia bone deformation than the controls for any given load during load carriage, and 3) Tibia bone density and structural properties quantified by pQCT will have strong correlations with tibia strains and strain rates, the following protocols and procedures have been implemented.

Participants

20 recreational female soccer players and 20 age-, height-, and weight-matched sedentary females between ages 18 and 27 years were recruited in the study. All participants met the current military height and weight enlisting standards and had no musculoskeletal conditions at the time of testing. All participants were classified as “low risk” for a cardiovascular event with exercise as defined by ACSM’s Guidelines for Exercise Testing and Prescription [6]. All participants had no tibia fractures and no history of smoking in the past two years. In addition, soccer players had been playing soccer regularly (≥ 2 hours/week) for a minimum of two years. Sedentary participants had not participated in any regular exercise training for the preceding 2 years. All participants signed an informed consent document approved by both the Institutional Review Board (IRB) at Ball State University (BSU) and the Human Research Protection Office (HRPO) of the U.S. ARMY.

The participants completed three data collection sessions; 1) peripheral quantitative computed tomography (pQCT) assessment of their tibias, 2) gait analysis during loaded walking conditions, and 3) computed tomography (CT) imaging of their tibias.

pQCT Assessments

The participants completed a pQCT assessment session which lasted approximately two hours.

A pQCT (XCT 2000; Stratec Medizintechnik, Pforzheim, Germany) was used to measure bone characteristics of the tibia. A standard phantom was used to calibrate the pQCT on each data collection day. Participants were positioned on a chair with the leg extended through the scanning cylinder for tibia scanning. Initial scout scans were conducted to determine the distal end plate of the tibia (reference plane). The following sites on the tibia were scanned: 4%, 14%, 38%, and 66% of the tibia length proximal to the reference plane [7]. In addition, the centers of the distal and middle thirds of the tibia bone shaft were scanned. Scanning speed was 20 mm/s. Slice thickness was 2.2 mm with a 0.5 mm voxel size. Custom Matlab programs (Mathworks, Natick, MA) were used to calculate the bone density, Young’s modulus, cross-sectional area, and area moment of inertia of the tibia bone at the seven scanned cross-sectional sites.

Load Carriage Protocol

The participants completed a set of walking trials under different loading conditions during which standard motion capture data were collected. This process took approximately two hours.

Fifteen VICON F-series cameras (200Hz) and VICON NEXUS 2.0 software (VICON Inc. Denver, CO) were used to collect three dimensional coordinates of reflective markers, ground reaction forces, and EMG of leg muscles. A modified standard plug-in gait marker set with cluster plates was used. A tandem force instrumented treadmill (AMTI Inc., Watertown, MA) was used to set the walking speed at

1.67 m/s and allowed ground reaction forces to be collected at 2000 Hz. A 16-channel Delsys wireless EMG system (Delsys Inc., Boston, MA) (2000 Hz) with bipolar single differential surface electrodes (41 mm x 20 mm electrode area, inter-electrode distance 1 cm) was used to monitor muscular activity of selected leg muscles.

Prior to data collection, basic participant anthropometric measurements were performed. These measurements include: height, weight, leg length, ankle width, knee width, and ASIS width.

Following the basic measurements and application of markers and electrodes, the participants warmed up by walking at a self-selected pace on the treadmill for 5 minutes.

The participants performed the walking tasks at 1.67 m/s on the force instrumented treadmill (AMTI, Watertown, MA) with: no backpack (0kg); a 10kg backpack; a 20kg backpack; and a 30kg backpack. Modular Lightweight Load-carrying Equipment (MOLLE) backpacks were used during the walking tasks. The participants wore standard military boots (Rocky S2V Tactical Military Boot, Rocky Brands, Inc.), shorts, and t-shirt during the experiment. Walking tasks were randomized and performed for 3 minutes each. Three seven-second trials per load condition were collected by a motion capture system (VICON Inc., Denver, CO) for biomechanical analysis. Participants rested for 5 minute between conditions.

CT Imaging Protocol

The participants visited Ball Memorial Hospital (Muncie, IN) accompanied by a member of the research team. During this visit, bilateral tibia CTs were recorded. This process took approximately one hour.

A Light Speed VCT machine (GE Medical System, Waukesha, WI) was used to obtain axial plane scans. CT slice thickness was 0.625 mm. The field of view (FOV) was 15cm x 15cm. Parameters of kVp and mAs were determined by machine algorithm (auto) based upon subject anthropometrics. Each leg was scanned separately from the calcaneus through the distal end of the femur. CT images were reconstructed in a 512x512 matrix with grey scale data stored and saved in a DICOM image format. A custom built phantom was scanned to establish a relationship between grey scale data and bone density.

Modeling and Finite Element Analysis

CT images of the tibia were imported into Mimics 16.0 (Materialise Inc., Leuven, Belgium). Segmentation of the tibias was then performed. A 3D tibia model was created and exported in drawing exchange format (DXF) for finite element modeling. The DXF file of the tibia model was imported into MARC 2013 (MSC Software Inc., Santa Ana, CA). A finite element (FE) model of the tibia was created to include hexahedral solid mesh elements (average size = 3mm x 3mm x 3mm) [8]. Material properties including density and Young's modulus were assigned to individual elements of the tibia FE model [8]. A modal neutral file (MNF) was generated to represent a flexible tibia. This MNF file was later used during multi-body dynamic simulations to elicit tibia bone strain information.

A subject specific musculoskeletal model was developed for each subject based on the height, mass, and gender of the participants in ADAMS 2012 (MSC Software Inc., Santa Ana, CA) through a LifeMOD 2010 plugin (LifeModeler Inc., San Clemente, CA). This model included seven segments; the pelvis, two thighs, two shanks, and two feet. All of these segments were initially modeled as rigid segments. The hip joint was modeled as a spherical joint, the knee joint was modeled as a revolute joint, and the

ankle joint was modeled as a universal joint. The passive joint stiffness properties were initially assigned based on equations described by Amankwah et al. [9]. These values were modified using an iterative design of experiments (DOE) approach with a cost function set to minimize the RMS error between the simulated and experimental ground reaction forces. 45 muscles were included in the model for each leg.

Lower-extremity joint position information obtained through the motion capture process were used to refine the musculoskeletal model. The motion capture data were then used to set the initial parameters of the model through an inverse kinematics simulation. After the initial parameters had been set and the model had been refined, a forward dynamics simulation was performed using inverse kinematics model states and ground reaction force data as inputs. During this simulation, the tibia segments of the model were replaced with correspondent flexible tibia bodies [10]. The tibia deformation data (stresses and strains) were calculated using the Durability plug-in in the ADAMS 2012. The tibia model and tibia bone strain data were divided into thirds along the longitudinal axis for the purpose of the statistical analyses.

Statistical Analyses

SPSS 21 (IBM Inc. Chicago, IL) was used to perform statistical analysis. To test the first hypothesis that soccer players' tibia bones would have greater mechanical properties than those of the controls, a one-way multivariate analysis of variance (MANOVA) was performed. The independent factor was the group effect between the soccer players and healthy controls. The dependent variables included bone density, cross-sectional area of cortical bone, area moments of inertia, Young's modulus of the tibia bone at the scanned sites. To test the second hypothesis of the study that soccer players would demonstrate smaller strains and strain rates during walking tasks with incremented load carriages, a mixed model repeated measures MANOVA was performed. The independent factors were the group effect (two levels: soccer players vs. healthy controls) and walking conditions (four load carriage levels: no load, 10 kg load, 20 kg load, and 30 kg load). The dependent variables included the maximal principal, minimal principal, and maximal shear strains and strain rates of the tibia bone. Significance level was at 0.05.

KEY RESEARCH ACCOMPLISHMENTS

- Ball State University IRB approval was obtained on June 3, 2015. (A Full Board Review)
- US ARMY HRPO approval was obtained on June 29, 2015.
- A keynote presentation titled “*Load carriage increases tibial strain rate*” was presented at the 2015 International Sport Science Congress in Gwangju, Korea on July 12, 2015. (Appendix A)
- A contract for a pQCT rental was signed between Ball State University and Bone Diagnostic Inc. on October 27, 2015. The leased pQCT scanner (Stratec XCT 3000, Germany) was delivered to Ball State University by Bone Diagnostic Inc. on November 5, 2015.
- A work flow for conducting experimental motion capture was created and tested in November 2015.
- A work flow for conducting pQCT scan on tibial bones was generated and tested in November 2015.
- Custom Matlab software programs were written and tested to perform the following computer modeling and simulation tasks: 1) Converting motion capture data into subject specific parameters for creating individualized musculoskeletal models in ADAMS. 2) Creating subject specific tibia models with bone density and mechanical properties assigned to individual hexahedral elements in MARC. 3) Exchanging data between MARC (an FEA program) and ADAMS (a multi-body dynamic simulation program). (August 2015 – November 2015)
- Forty participants were recruited and screened from January 2016 – May 2016.
- Experimental data collection for all forty participants was completed in May 2016.
- The presentation titled “*Tibia bone strain distribution during incremented load carriage*” was given at the 63rd Annual Meeting of American College of Sports Medicine on June 1, 2016. The abstract was published in *Medicine & Science in Sports & Exercise*. V. 48, No. 5 Supplement, S308 (Appendix B)
- Data processing of motion capture, pQCT, and CT data was completed in August 2016.
- The project titled “*Tibia strain patterns during walking and load carriage*” was presented at the 40th Annual Meeting of American Society of Biomechanics in August 2016. (Appendix C)
- The manuscript titled “*Influence of load carriage and physical activity history on tibial bone strain*” was published by the *Journal of Sport and Health Science* in September 2016. (Appendix D)
- A one-year no cost extension request for the project was approved by the ARMY in December 2016.

- The manuscript titled “*The effect of drop-landing height on tibia bone strain*” was published by the *Journal of Biomedical Science and Engineering* in January 2017. (Appendix E)
- The presentation titled “*The effect of soccer training on tibia bone properties in healthy young females*” was given at the 64th Annual Meeting of American College of Sports Medicine on June 1, 2017. The abstract was published in *Medicine & Science in Sports & Exercise*. V. 49, No. 5 Supplement, S398. (Appendix F)
- The project titled “*The effect of soccer training history and load carriage on joint mechanics*” was presented at the 2017 Congress of International Society of Biomechanics in July 2017. The abstract was published in *Proceedings of XXVI Congress of the International Society of Biomechanics*, 562 (Appendix G)
- The abstract titled “*The effect of soccer history on tibial strain during load carriage*” was submitted for presentation consideration at the 65th Annual Meeting of American College of Sports Medicine in October 2017. (Appendix H)
- The manuscript titled “*A novel approach to estimating bone strain during dynamic motion*” was submitted for publication consideration at *Journal of Visualized Experiments* in December 2017. (Appendix I)

REPORTABLE OUTCOMES

The following sections outline the results of this project. These data along with subsequent analyses will form the basis for forthcoming presentations and manuscripts documenting this research.

Participant Characteristics:

Twenty college-age female soccer players and twenty gender, body mass, and body height matched healthy sedentary controls were recruited in this study. Participants' demographic information and leg strength characteristics are presented in Table 1. The soccer players were slightly younger than the healthy controls (1 year younger). In addition, soccer players' knee and ankle muscles were stronger than those of healthy controls.

Table 1. Subject demographic information

	Soccer group	Control group
Age (yrs)	19.6(1.3) [#]	20.5(1.3)
Height (cm)	166.1(5.4)	165.5(4.1)
Mass (kg)	61.8(5.9)	60.8(10.4)
Knee Extensor Torque (Nm/kg)	4.19(0.64) ^{##}	2.99(0.71)
Knee Flexor Torque (Nm/kg)	1.99(0.29) ^{##}	1.29(0.46)
Ankle Plantar-flexor Torque (Nm/kg)	1.02(0.22) ^{##}	0.81(0.16)
Ankle Dorci-flexor Torque (Nm/kg)	0.56(0.08) ^{##}	0.47(0.08)

Mean (standard deviation) for basic participant demographics and results from leg strength assessments.

[#] $p < 0.05$, ^{##} $p < 0.001$, compared with healthy controls.

Gait Temporo-spatial Parameters and Ground Reaction Force:

Gait temporo-spatial parameters and ground reaction force of the four walking tasks performed are presented in table 2, which shows the means and standard deviations (SDs) of stride length, step rate, double support time, peak vertical ground reaction force, and peak vertical ground reaction loading rate during stance of walking. Two-way repeated-measures ANOVAs were used to determine the effects of incremented load carriage and soccer training history on the selected gait parameters and ground reaction force variables.

Table 2. Means (SDs) of temporo-spatial parameters and ground reaction forces during loaded walking

Variables\Conditions	WK00	WK10	WK20	WK30
<u>Soccer Players</u>				
Stride length (m)	1.54 (0.06)	1.54 (0.07)	1.51 (0.08)**	1.47 (0.08)**
Step frequency (steps/min)	130 (6)	130 (6)	133 (7)**	136 (8)**
Double support time (% gait cycle)	19(1)	21(1)**	23(1)** ^{##}	25(1)** ^{##}
Peak VGRF (BW)	1.29 (0.05)	1.53 (0.07)**	1.76 (0.12)**	2.04 (0.17)**
Peak VGRLR (BW/s)	40.0(4.8)	43.5(6.3)**	44.4(7.2)	46.2(7.4)*
<u>Healthy Controls</u>				
Stride length (m)	1.56 (0.07)	1.53 (0.09)**	1.51 (0.09)	1.44 (0.10)**
Step frequency (steps/min)	129 (6)	132 (9)*	134 (9)	140 (11)**
Double support time (% gait cycle)	19(2)	22(3)**	25(1)**	27(2)**
Peak VGRF (BW)	1.32 (0.09)	1.54 (0.10)**	1.81 (0.15)**	2.09 (0.19)**
Peak VGRLR (BW/s)	42.1(6.3)	45.7(7.9)**	47.1(9.8)	46.5(6.8)

Conditions: WK00 = unloaded walking, WK10 = 10 kg loaded walking, WK20 = 20 kg loaded walking, WK30 = 30 kg loaded walking. VGRF = vertical ground reaction force. VGRLR = vertical ground reaction instantaneous loading rate. [#] $p < 0.05$, ^{##} $p < 0.01$, compared with healthy controls; * $p < 0.05$, ** $p < 0.01$ compared with previous level of the loaded walking condition.

Significant interactions ($p < 0.05$) were found for stride length and step rate. Therefore, for each loaded walking condition, separate ANOVA tests were run to determine the effect of soccer training history on these gait parameters. Also, for each group of participants, separate ANOVA tests were run to determine the effect of load carriage on these gait parameters. It was revealed that load carriage in general leads to a decrease in stride length and an increase in step rate for each group. Furthermore, for both groups, load carriage leads to increases in double support time, peak vertical ground reaction force, and peak instantaneous ground reaction force loading rate.

In addition, the soccer players were found to use smaller double support time when carrying 20 kg and 30 kg loads than that of the healthy controls ($p < 0.01$).

Lower-extremity Joint Mechanics:

Table 3 shows the means and SDs of the lower-extremity peak moments and powers during stance of walking. Two-way repeated-measures ANOVAs were used to determine the effects of incremented load carriage and soccer training history on the selected lower-extremity joint mechanics variables.

Table 3. Means (SDs) of lower-extremity joint mechanics during loaded walking

Variables\Conditions	WK00	WK10	WK20	WK30
<u>Soccer Group</u>				
Peak ankle plantar-flexor moment (Nm/kg)	1.92(0.13)	2.14(0.15)**	2.32(0.16)**	2.35(0.22)
Peak knee extensor moment (Nm/kg)	1.02(0.14)	1.23(0.22)**	1.49(0.29)**	1.81(0.31)**#
Peak hip extensor moment (Nm/kg)	1.46(0.23)	1.59(0.22)**	1.73(0.23)**	1.87(0.23)**
Peak ankle power absorption(W/kg)	-1.82(0.35)	-1.20(0.34)**	-1.99(0.41)	-1.80(0.48)#
Peak knee power absorption (W/kg)	-3.01(0.80)	-3.63(1.16)**	-4.06(1.19)*	-4.05(0.95)##
Peak hip power absorption (W/kg)	-0.99(0.27)##	-1.19(0.40)**##	-1.39(0.35)**##	-1.56(0.38)**
Peak ankle power production(W/kg)	3.70(0.57)	4.10(0.51)**	4.46(0.51)**	4.48(0.55)
Peak knee power production (W/kg)	1.81(0.30)	1.84(0.32)#	2.09(0.45)**##	2.18(0.56)##
Peak hip power production (W/kg)	1.60(0.29)##	1.90(0.31)**##	2.07(0.31)**##	2.32(0.47)**
<u>Control Group</u>				
Peak ankle plantar-flexor moment (Nm/kg)	1.97(0.16)	2.16(0.20)**	2.34(0.23)**	2.24(0.23)**
Peak knee extensor moment (Nm/kg)	1.05(0.29)	1.29(0.30)**	1.67(0.35)**	2.08(0.48)**
Peak hip extensor moment (Nm/kg)	1.54(0.33)	1.70(0.37)**	1.88(0.40)**	2.06(0.36)**
Peak ankle power absorption(W/kg)	-1.96(0.39)	-1.99(0.47)	-2.02(0.53)	-1.52(0.29)**
Peak knee power absorption (W/kg)	-3.34(1.17)	-3.82(1.08)**	-4.54(0.82)**	-4.93(1.04)
Peak hip power absorption (W/kg)	-1.56(0.45)	-1.67(0.50)	-1.92(0.67)*	-2.11(0.85)
Peak ankle power production(W/kg)	3.99(0.73)	4.11(0.92)	4.55(0.81)**	4.33(0.71)*
Peak knee power production (W/kg)	1.99(0.35)	2.12(0.50)	2.51(0.61)**	2.94(1.11)*
Peak hip power production (W/kg)	1.94(0.33)	2.38(0.55)**	2.45(0.36)	2.57(0.52)

Conditions: WK00 = unloaded walking, WK10 = 10 kg loaded walking, WK20 = 20 kg loaded walking, WK30 = 30 kg loaded walking. # $p < 0.05$, ## $p < 0.01$, compared with healthy controls; * $p < 0.05$, ** $p < 0.01$ compared with previous level of the loaded walking condition.

Significant interactions ($p < 0.05$) were found for peak ankle planta-flexor moment, peak knee extensor moment, peak ankle power absorption and production. Therefore, for each loaded walking condition, separate ANOVA tests were run to determine the effect of soccer training history on these variables. Also, for each group of participants, separate ANOVA tests were run to determine the effect of load carriage on these variables.

It was determined that load carriage led to increases in peak ankle, knee, and hip moments and power absorption and production for both groups. When compared to the healthy controls, the soccer players were found to use less peak hip power absorption and production during loaded walking. In addition, the soccer players used less peak knee power production than those of healthy controls when carrying loads (≥ 10 kg).

Tibia Bone Density and Strength Indices:

Table 4 shows the means and SDs of the tibia bone density and strength indices at various levels (14%, 38%, and 66% tibia, centers of distal, middle, and proximal thirds of tibia) along the bone shaft. One way ANOVAs were used to determine the effects of soccer training history on the selected variables obtained from the pQCT measurement.

Table 4. Means(SDs) tibia bone density and strength indices

Variables\Groups	Soccer group	Control group
<u>Cortical bone density (mg/cm³)</u>		
14% tibia	1134.6(18.6) ^{##}	1151.4(20.1)
38% tibia	1170.9(16.4) ^{##}	1203.9(17.6)
66% tibia	1140.7(15.6) ^{##}	1166.8(20.9)
Center of distal third of tibia	1167.2(18.1) ^{##}	1183.7(18.8)
Center of middle third of tibia	1165.1(19.7) ^{##}	1199.6(18.6)
Center of proximal third of tibia	1125.6(16.0) ^{##}	1151.9(25.4)
<u>Bone strength indices (mm³)</u>		
14% tibia	1590.9(219.9) ^{##}	1348.9(233.4)
38% tibia	1733.3(242.5) ^{##}	1433.2(304.2)
66% tibia	2604.6(375.1) ^{##}	2187.7(440.8)
Center of distal third of tibia	1499.6(215.2) ^{##}	1293.6(239.6)
Center of middle third of tibia	1878.7(272.9) ^{##}	1558.6(335.8)
Center of proximal third of tibia	2774.6(383.2) ^{##}	2278.6(494.0)

[#] $p < 0.05$, ^{##} $p < 0.01$, compared with healthy controls.

The soccer players were found to have less cortical bone density along the tibia shaft than that of the healthy controls ($p < 0.01$). However, the tibia strength indices of the soccer players were stronger than that of the healthy controls ($p < 0.01$).

Tibia Bone Mechanical Properties:

Table 5 shows the means and SDs of the tibia bone mechanical properties represented by area moments of inertia in three principal directions at various levels of the tibia shaft (14%, 38%, and 66% tibia,

centers of distal, middle, and proximal thirds of tibia). One way ANOVAs were used to determine the effects of soccer training history on the selected variables obtained from the pQCT measurement.

Table 5. Means(SDs) tibia bone area moments of inertia

Variables\Groups	Soccer group	Control group
<u>Area moments of inertia (AP direction) (mm⁴)</u>		
14% tibia	12087.4(2967.0) ^{##}	9338.9(2311.9)
38% tibia	15814.5(3746.4) ^{##}	11684.5(3635.6)
66% tibia	33892.7(7552.4) ^{##}	25819.4(6845.9)
Center of distal third of tibia	10949.5(2785.4) ^{##}	8538.2(2204.4)
Center of middle third of tibia	19506.6(4474.5) ^{##}	14104.8(4425.6)
Center of proximal third of tibia	38853.8(9025.7) ^{##}	28738.9(8947.6)
<u>Area moments of inertia (ML direction) (mm⁴)</u>		
14% tibia	13714.8(2900.0) ^{##}	10594.1(2317.6)
38% tibia	11653.9(1933.9) ^{##}	9107.4(2475.3)
66% tibia	17510.5(3477.8) [#]	14552.5(4075.9)
Center of distal third of tibia	11028.3(1865.8) ^{##}	8955.7(2006.5)
Center of middle third of tibia	12711.0(2253.5) ^{##}	9703.8(2651.2)
Center of proximal third of tibia	20062.9(4352.7) [#]	16568.7(4954.7)
<u>Area moments of inertia (longitudinal direction) (mm⁴)</u>		
14% tibia	25802.2(5675.1) ^{##}	19932.9(4536.4)
38% tibia	27468.4(5129.9) ^{##}	20791.8(5722.8)
66% tibia	51403.3(10270.6) ^{##}	40371.9(10309.6)
Center of distal third of tibia	21977.87(4408.8) ^{##}	17493.9(4117.6)
Center of middle third of tibia	32217.6(6189.9) ^{##}	23808.6(6733.3)
Center of proximal third of tibia	58916.7(12390.0) ^{##}	45307.6(13392.3)

[#] $p < 0.05$, ^{##} $p < 0.01$, compared with healthy controls.

The soccer players were found to have greater area moments of inertia along the anteroposterior, mediolateral, and longitudinal directions in their tibia bones than those of healthy controls ($p < 0.01$).

Peak Strain and Strain Rate during Loaded Walking Tasks:

Peak strain during load carriages

For all the strain variables examined, significant interactions were found between the subject group and incremented load carriage ($p = 0.0001$). Therefore, separate ANOVA tests were run to determine the simple effects from the multi-axial loading history and incremented load carriage on tibia strain.

Table 6 shows the means and standard errors (SEs) of the peak compressive, tensile, and shear strain of the middle third of the tibia shaft during loaded walking. One-way repeated measures ANOVAs were run to examine the effects of the incremented load carriage on tibia bone strain for the soccer group and the control group, respectively. For both groups, significant differences in peak compressive strain, tensile strain, and shear strain (all $p = 0.0001$) were found among loaded walking tasks. Specifically, a dose-response relationship between the incremented load carriage and tibia strain existed for the soccer group from 0 kg to 30 kg loads and for the control group from 0 kg to 20 kg loads. As the load carriage increased, tibia compressive, tensile, and shear strain increased. However, strains of the 30 kg load were lower than those of the 20 kg load in the control group.

One-way repeated measures ANOVAs were run to examine the effects of multi-axial loading history on the tibia bone strain for each of the loaded walking tasks. Significant differences in peak compressive strain, tensile strain, and shear strain (all $p = 0.0001$) were found between the two groups. The soccer group consistently exhibited lesser peak compressive, tensile, and shear strain than the controls.

Table 6. Tibia bone strains during walking with load carriages of 0 kg, 10 kg, 20 kg, and 30 kg (mean $\pm$ SE).

Tibia bone strain (μs)	0 kg	10 kg	20 kg	30 kg
Soccer group				
Compressive strain	849 $\pm$ 3 ^{##}	960 $\pm$ 4 ^{**##}	1092 $\pm$ 10 ^{**##}	1180 $\pm$ 6 ^{**##}
Tensile strain	562 $\pm$ 2 ^{##}	634 $\pm$ 3 ^{**##}	736 $\pm$ 7 ^{**##}	825 $\pm$ 4 ^{**##}
Shear strain	651 $\pm$ 2 ^{##}	736 $\pm$ 2 ^{**##}	838 $\pm$ 7 ^{**##}	922 $\pm$ 4 ^{**##}
Control group				
Compressive strain	1039 $\pm$ 3	1179 $\pm$ 4 ^{**}	1749 $\pm$ 11 ^{**}	1152 $\pm$ 7 ^{**}
Tensile strain	684 $\pm$ 3	792 $\pm$ 3 ^{**}	1152 $\pm$ 7 ^{**}	1015 $\pm$ 4 ^{**}
Shear strain	799 $\pm$ 2	908 $\pm$ 2 ^{**}	1325 $\pm$ 7 ^{**}	1200 $\pm$ 4 ^{**}

* $p \leq 0.05$, ** $p \leq 0.0001$, compared with previous level of the loaded walking condition; # $p \leq 0.05$, ## $p \leq 0.0001$, compared with healthy controls.

Peak strain rate during load carriages

For all the strain rate variables examined, significant interactions were found between the subject group and incremented load carriage ($p = 0.0001$). Therefore, separate ANOVA tests were run to determine the simple effects from the multi-axial loading history and incremented load carriage on tibia strain rate.

Table 7 shows the means and SEs of the peak compressive, tensile, and shear strain rates of the middle third of the tibia shaft during loaded walking. One-way repeated measures ANOVAs were run to examine the effects of the incremented load carriage on tibia bone strain rate for the soccer group and the controls, respectively. For both groups, significant differences in compressive strain rate, tensile strain rate, and shear strain rate (all $p = 0.0001$) were found between loaded walking tasks. Specifically, a dose-response relationship between load carriage and strain rates existed for the soccer group from 0 kg to 30 kg load and for the control group from 0 kg to 20 kg load. Both the soccer group and control group demonstrated significant increases in peak compressive, tensile, and shear strain rate as load carriage increased. However, when load carriage was increased to 30 kg, the strain rates of the control group decreased from those of 20 kg load.

One-way ANOVA tests were run to examine the effect of multi-axial loading history on tibia strain rate for each of the loaded walking tasks. Significant differences in peak compressive strain rate, tensile strain rate, and shear strain rate (all $p = 0.0001$) were found between the soccer group and controls. Specifically, the soccer group demonstrated lesser strain rates than the controls during all loaded walking conditions (all $p = 0.0001$).

Table 7. Tibia bone strain rates during walking with load carriages of 0 kg, 10 kg, 20 kg, and 30 kg (mean $\pm$ SE).

Tibia bone strain rate ($\mu\text{s/s}$)	0 kg	10 kg	20 kg	30 kg
Soccer group				
Compressive strain rate	12757 $\pm$ 61 ^{##}	13549 $\pm$ 66 ^{**##}	16486 $\pm$ 253 ^{**##}	17744 $\pm$ 89 ^{**##}
Tensile strain rate	10509 $\pm$ 55 ^{##}	11076 $\pm$ 59 ^{**##}	13147 $\pm$ 119 ^{**##}	14525 $\pm$ 70 ^{**##}
Shear strain rate	10879 $\pm$ 37 ^{##}	11514 $\pm$ 41 ^{**##}	13803 $\pm$ 161 ^{**##}	15086 $\pm$ 53 ^{**##}
Control group				
Compressive strain rate	16279 $\pm$ 64	18840 $\pm$ 70 ^{**}	31433 $\pm$ 267 ^{**}	23530 $\pm$ 94 ^{**}
Tensile strain rate	12890 $\pm$ 58	14964 $\pm$ 62 ^{**}	20425 $\pm$ 125 ^{**}	17558 $\pm$ 74 ^{**}
Shear strain rate	13746 $\pm$ 39	15917 $\pm$ 43 ^{**}	24168 $\pm$ 170 ^{**}	19345 $\pm$ 56 ^{**}

* $p \leq 0.05$, ** $p \leq 0.0001$, compared with previous level of the loaded walking condition; # $p \leq 0.05$, ## $p \leq 0.0001$, compared with healthy controls.

SUMMARY AND DISCUSSION

The recreational soccer players and the healthy controls recruited in this study were gender, body mass, and body height matched. The healthy controls were slightly older than the soccer players. The age difference between the two groups was less than one year. The soccer players possessed stronger muscles around their knee and ankle joints than the healthy controls. When the quality of the tibia bone was assessed, it was found that the tibias of the healthy controls had higher cortical bone density than that of the soccer players. However, the soccer players had larger area moments of inertia along the tibia shaft than those of the healthy controls. Also, the strength indices of the tibias of the soccer players were greater than those of the healthy controls. The differences in leg muscle strength and tibia bone quality between the two groups appear to be related to the soccer training history. Regularly engaging in soccer could result in increases in leg muscle strength and alterations in tibia quality. Specifically, soccer training history leads to favorable changes in bone's mechanical properties without an increase in density. In fact, the cortical bone density of the soccer players was lower than that of the healthy controls. These findings suggest a co-adaptation of intra-cortical bone remodeling and bone formation modeling during soccer training such that bone geometry is optimized without increases in bone density, which would be metabolically expensive. The enhanced mechanical properties in soccer players could make the tibia more resilient to unaccustomed mechanical loading, such as load carriage.

When incremented load carriage was introduced to the two groups, it was found that load carriage resulted in a decreased stride length, increased step rate, increased double support time, and increased ground reaction force and loading rate. Greater lower-extremity joint moments and powers were elicited during load carriage. There were some differences in joint mechanics existing between the two groups during load carriage. The healthy controls were found to exert more hip and knee powers during incremented load carriage than those of soccer players. For a given load carriage task, using more lower-extremity joint power reflects higher energy expenditure, which is associated with higher metabolic cost. Thus, the healthy controls may experience fatigue sooner during load carriage than soccer players. On the contrary, the soccer players seemed to have more energy reserves for more physical demanding tasks such as carrying heavy loads for a longer duration.

The effects of soccer training history and incremented load carriage on tibia deformation were examined through computer modeling and finite element analysis. Significant differences in strains and strain rates were found among walking conditions and between the two experimental groups. The incremented load carriage resulted in large increases in strains and strain rates for both groups. A dose-response relationship between bone strain and load carriage was observed for both groups. Soccer players experienced this dose-response relationship from 0 kg to 30 kg load carriage. The healthy controls also experienced a similar dose-response relationship from 0 kg to 20 kg load carriage. This relationship highlights the direct influence of load carriage on tibial bone deformation. Interestingly, the healthy controls exhibited an alteration of bone strain patterns beyond the 20 kg load, specifically, tibial bone strain was reduced from the 20 kg level when a 30 kg load was carried. It appears that the 20 kg load could be a critical loading threshold where the healthy controls have to use movement strategies such as taking smaller steps and higher step rates to lower bone strains. It is possible that the human body could sense the danger of high strain to bone health and would use preventative strategies to lower bone strain during physically demanding tasks. On one hand, using movement strategies such as taking short steps and increasing step rates during heavy load carriage is effective to reduce bone strain. On the other hand, using these movement strategies might result in greater metabolic cost and energy consumption, which will accelerate the development of fatigue and will undoubtedly limit the participant's ability to achieve the physical training goals. Benefited from their enhanced tibia bone strength, soccer players did not

show a threshold prior to the 30 kg load. Thus, it is evident that a stronger tibia bone could help participants perform load carriage by reducing the need of employing significant gait changes. The current findings of a dose-response relationship between load carriage and strain as well as the threshold phenomenon in bone strain are in agreement with a previous study examining load carriage effect in male subjects [11].

The effect of soccer training history on tibial bone response is profound. The soccer group demonstrated significantly less bone strain (18-36% less in tensile strain, 18-38% less in compressive strain) and less bone strain rate (17-36% less in tensile strain rate, 22-48% less in compressive strain rate) than those of the healthy controls. For given load carriage, smaller bone strain and strain rate reflect greater bone resistance to external mechanical loads. Strain findings from this load carriage study well echo findings from the imaging study examining bone quality, which indicates that a history of soccer training results in a strong tibia bone. A strong tibia bone appears to be beneficial for soccer players to perform load carriage with less bone deformation than those of healthy controls. Moreover, less bone deformation during a given load carriage is advantageous because major gait modifications would not be required to protect the tibia and load carriage task could be performed in a state of low metabolic cost.

Soccer training is characterized with engaging in dynamic movements such as running, jumping, accelerations, decelerations, and cuttings. These dynamic movements introduce multi-axial ground impact forces to lower bodies. Hypothetically, under frequent multi-axial loadings, tibias would go through a remodeling process, which could improve bone strength uniformly and make the bones suitable for resisting unaccustomed loads in the future. However, there is a lack of experimental evidence to support this hypothesis. Thus, the first part of the current study was to examine the effect of a soccer training history on tibia bone quality in an effort to understand the influence of multi-axial loading on tibia bone strength. Outcomes of the study proves that a training history with multi-axial loadings such as soccer results in a significant enhancement of tibia bone quality. The second part of the current study was to quantify tibia deformations during incremented load carriage in an effort to understand whether a strong tibia bone is resistant to unaccustomed loading from load carriage. Outcomes of this study proves that a strong tibia bone is resilient to load carriage with small bone deformation exhibited.

Tibia stress fracture is very common in the military. Female recruits face a higher risk of developing tibia stress fracture than their male counterparts [1,4,5]. Thus, it is critical to develop effective preconditioning programs for female recruits to lower the incidence of stress fractures during basic training. In this study, it was determined that a training history with multi-axial loading (e.g. soccer) is beneficial for improving tibia bone quality. A strong tibia bone is resistant to deformation associated with heavy load carriage. These findings suggest that physical training involving multi-axial loading (e.g. soccer) may be an ideal exercise modality for preconditioning prior to basic combat training for female recruits.

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Appendix A

Abstract presented at the 2015 International Sport Science Congress in GwangJu, Korea on July 12, 2015

Title: Load Carriage Increases Tibial Strain Rate

Load Carriage Increases Tibial Strain Rate

Henry Wang * Clark Dickin * Mohammad Kia *Ball State University*

Purpose: To determine the effect of incremented load carriage on tibial bone strain rate.

Method: Forty recreational runners ($n=20$, 21 ± 2 yr.) and basketball players ($n=20$, 21 ± 2 yr.) participated in this study. Participants completed four walking tasks: walking with 0kg, 15kg, 25kg, and 35kg loads. Each task was performed for 5 min on a force instrumented treadmill (AMTI) at 1.67 m/s. Participants' tibial bone models obtained from CT scans were combined with subject-specific musculoskeletal models for forward-dynamic computer simulations and finite element analyses. Strain rate of the middle third of the tibial bone shaft were analyzed. One-way ANOVAs were performed, $\alpha = 0.05$.

Result: Significant differences in strain rate were found among walking conditions ($P<0.05$). The means $\pm$ SEs of runner's bone strain rates ($\mu\text{s/s}$) during load carriage (0kg, 15kg, 25kg, and 35kg) were 7956 ± 24 , 9478 ± 27 , 11562 ± 33 , and 11583 ± 37 in compression, respectively; and 6647 ± 30 , 7902 ± 23 , 9552 ± 29 , and 9617 ± 33 in tension, respectively. For the ball players, the 0kg, 15kg, 25kg, and 35kg load carriage result in compressive strain rates ($\mu\text{s/s}$) of 7883 ± 23 , 9034 ± 26 , 10293 ± 32 , and 11661 ± 36 , respectively; and tensile strain rates ($\mu\text{s/s}$) of 6557 ± 19 , 7432 ± 23 , 8632 ± 27 , and 10013 ± 32 , respectively. In addition, runners showed greater strain rates than the ball players when carrying 15kg and 25kg loads ($P<0.05$).

Conclusion: load carriage leads to increases in tibial bone strain rates, in particular, for people engaging in basketball, a dose-response relationship exists between the incremented load carriage from 0kg to 35kg and tibia strain rates. For people engaging in running, the dose-response relationship exists from 0kg to 35kg for the tensile strain rates and from 0kg to 25kg for the compressive strain rates. As the military recruits are often afflicted by lower-extremity overuse injuries such as tibial stress fracture, it is necessary to determine the mechanisms associate with the injuries. Load carriage is an essential task during military basic training. The increases in bone strain rates during load carriage appear to expose trainees to an increased risk of tibial stress fracture. Also, with small strain rates exhibited, it is evident that tibia may benefit from a history of playing basketball for resisting external load.

Key words: Stress fracture, load carriage, strain rate

Appendix B

Abstract was presented at the 63rd annual meeting of American College of Sports Medicine (ACSM) in Boston, MA. on June 1, 2016.

Title: Tibia Bone Strain Distribution during Incremented Load Carriage

CONCLUSIONS: No association was observed between serum 25(OH)D levels and markers of bone health in non-weight bearing athletes. Lower body weight was associated with osteopenia or osteoporosis regardless of serum 25(OH)D level.

1131 Board #5 June 1, 3:15 PM - 5:15 PM

Tibia Bone Strain Distribution during Incremented Load Carriage

Henry Wang¹, Mohammad Kia², Clark Dickin¹. ¹Ball State University, Muncie, IN. ²Hospital for Special Surgery, New York City, NY.

(No relationships reported)

Habitual runners and military recruits often develop tibia stress fracture (TSF). Continuous repetitive impact loading is a risk factor of TSF. Load carriage is essential to basic training and results in high bone strain, which may elicit TSF. Common TSF sites are at distal (DT) and middle (MT) thirds of the tibia. However, it is unknown whether incremented load carriage results in high strains in these regions. Also, it is important to determine if past physical activity results in different strain distributions during load carriage.

PURPOSE: To examine the effect of incremented load carriage on tibia strain distribution.

METHODS: Forty recreational runners (n=20, 21±2 yr.) and basketball players (n=20, 21±2 yr.) completed four tasks: walking with 0kg, 15kg, 25kg, and 35kg loads. Each task was performed on a force treadmill at 1.67 m/s. Participants' tibia models from CT scans were combined with subject-specific musculoskeletal models for forward-dynamic simulations and finite element analyses. Strains of the tibia shaft were analyzed. One-way ANOVAs were performed. $\alpha = 0.05$.

RESULTS: Significant differences in strain distribution were found ($P < 0.0001$). For the 15kg condition, peak compressive and shear strains were presented at the DT and the peak tensile strain was presented at the MT. Specifically, the means ± SEs of runner's peak bone strains were 840±2 µs, 562±2 µs, and 1255±3 µs in compression, tension, and shear, respectively. Ball players' peak bone strains were 803±2 µs, 519±2 µs, and 1185±3 µs in compression, tension, and shear, respectively. As load carriage was increased beyond 15kg, peak shear strain shifted toward the MT from the DT. In particular, this pattern of shear strain redistribution was observed at 25kg and 35kg levels for runners and ball players, respectively.

CONCLUSION: Load carriage results in high compressive and shear strains in the DT and high tensile strain in the MT. Incremented load carriage leads to a shift of peak shear strain from the DT to the MT. Ball players' tibias are resilient to load carriage with small strains exhibited and a delayed shift of the shear strain. This study supports the epidemiology that common TSF sites are at DT and MT. Preconditioning programs involved running may be less effective to prevent TSF than basketball. US ARMY #A81XVH-08-1-0587; #A81XVH-15-1-0006.

1132 Board #6 June 1, 3:15 PM - 5:15 PM

Eccentric Contractions Promote Osteoclast Activity in the Tibia

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(No relationships reported)

The notion that bone and muscle communicate at the biochemical and molecular levels in ways beyond simple mechanical interactions is leading to new insight into how bone and muscles work together to maintain health. We studied how injury in the skeletal muscle impacts bone remodeling.

PURPOSE: To determine osteoclast activity in the tibia following contraction-induced injury in the tibialis anterior muscle (TA).

METHODS: Using an established model, unilateral injury to the TA was induced by 15 lengthening contractions. Injury was confirmed by measuring changes in maximal dorsiflexor isometric torque before and after lengthening contractions. With the TA in situ, both tibias were harvested 2 or 7 days after injury, fixed, decalcified, paraffin embedded, and sectioned. We evaluated osteoclast number (Tartrate-resistant acid phosphatase staining) and assessed local muscle damage (labeling of cytoskeletal proteins). The contralateral, uninjured tibia served as a control.

RESULTS: Osteoclast number was similar at days 2 and 7 in injured tibia. Osteoclast number was markedly higher along the lateral surface of the diaphysis in injured tibia compared to control (osteoclast surface/bone surface ratios: 0.215 ± 0.03 in injured vs. 0.09 ± 0.02 control). At day 2, there was a localized loss of desmin labeling in muscle fibers adjacent to increased osteoclast activity.

CONCLUSIONS: Our results indicate a time-dependent metabolic change of bone resorption in response to forceful lengthening contractions, a model of muscle strain injury. Future experiments include dynamic histomorphometry to assess bone formation and detailed analysis of muscle and bone progenitor cell proliferation (e.g., Ki-67 labeling) and myokine release and activity (e.g., FGF-2, IGF-1 expression) in bone, muscle and/or the interface following eccentric injury. An understanding of underlying mechanisms is essential for developing targets for therapeutic interventions of musculoskeletal injury and disease.

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1133 Board #7 June 1, 3:15 PM - 5:15 PM

Do 6 Months Of Whole Body Vibration Training Improve Bone Mass Acquisition Of Adolescent Swimmers?

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(No relationships reported)

Whole body vibration (WBV) training has been suggested to be an effective type of training for improving bone mass.

PURPOSE: To determine the effects of WBV training on bone mass acquisition in adolescent swimmers.

METHODS: Sixty-four swimmers were followed over 8 months. Participants were divided in to two groups: The first group consisted of forty-one swimmers (14.2±1.8 y) who completed a WBV protocol 15 minutes of training 3 days per week during 6 months (WBV training increased each month starting with a peak acceleration of 3.6 g and ending at 11.6 during the last month) while continuing with their habitual water training (VIB). The second group consisted of twenty-three swimmers (15.0±2.2 y) who continued with their regular swimming training (SWI) (both groups performed an average of 10 hours per week of water training). Bone mineral density (BMD) and content (BMC) were measured longitudinally (8 months) by Dual Energy X-ray Absorptiometry at the whole body, lumbar spine and non-dominant hip. Analysis of covariance (ANCOVA) for repeated measures x2 (time) were performed to check differences within groups between pre and post intervention and to determine the effects of the intervention on BMD and BMC values adjusting by change in height and subtotal lean, initial age and final Tanner stage and calcium intake.

RESULTS: Six months of WBV training had little effect on bone mass, as no differences were found between VIB and SWI for absolute change or percentage change for BMD values. For BMC, VIB presented higher absolute and percentage changes in both trochanter (7% increase in VIB vs. 3% in SWI) and total hip (6% increase in VIB vs. 3% in SWI) than SWI, although there was no group by time interaction.

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Appendix C

Abstract presented at the 40th annual meeting of American Society of Biomechanics in Raleigh, NC,
August 2nd -5th, 2016

Title: Tibia Strain Patterns during Walking and Load Carriage

TIBIA STRAIN PATTERNS DURING WALKING AND LOAD CARRIAGE

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INTRODUCTION

Tibia stress fractures are severe musculo-skeletal injuries, which are common in sports and in military training [1]. The injury mechanism is not well understood. It is hypothesized that increased mechanical usage along with high strains and strain rates, stimulates bone remodeling, which results in locally increased porosity and decreased bone mass. Bone is weakened at this stage. If training continues, bone microdamages will occur and stress fractures are formed. The high incidence of tibia stress fracture in the military is related to large volume and intensity of physical training. Load carriage is an essential component of the basic training. In fact, the combined mileage of load carriage and running in a 12-week basic training generally exceeds 200 miles [2]. Although there seems to be a connection between intense training represented by load carriage and risk of tibia stress fracture, research is needed to examine the effect of load carriage on tibia bone deformation and bone strain patterns.

The purpose of this study was to examine incremented load carriage on tibia bone strains and to determine tibia bone strain patterns during loaded walking.

METHODS

Forty healthy male subject (Age = 21 ± 2 yr., height = 1.80 ± 7 m, and weight = 78 ± 11 kg) performed four walking tasks with load carriage of 0kg, 15kg, 25kg, and 35kg. A VICON motion capture system was used to record kinematics (240 Hz) and AMTI force plates were used to record ground reaction forces (2400 Hz). Computed tomography (CT) images of subjects' tibia bones were obtained. 3D tibias were segmented in MIMICS 14.0 (Materialise, Leuven, Belgium).

MARC 2012 (MSC Software, Santa Anna, CA) was used to generate finite element (FE) models of tibia bones. Mechanical properties were assigned based on bone density. The FE model was converted into a flexible tibia in MARC to incorporate the tibia geometry in LifeMod (LifeModeler Inc., San Clemente, CA). The subjects' weight, height, gender, and age as well as the relative positions of the ankle, knee, and hip joints, determined from the motion capture, were used to scale the generic lower extremity models based on the GeBOD anthropometric database. Generic tibia segments were replaced with the developed flexible tibias. A total of ninety muscles were added to the right/left legs. The measured kinematics, collected during walking tasks, were used to drive the model with an inverse dynamics algorithm [3], while the muscle shortening/lengthening patterns were recorded. Next, kinematic constraints were removed, and muscles served as actuators to replicate the motions during forward dynamic simulations.

The maximum and minimum principle strain values (tension/compression) for all surface nodes of the tibia bone were computed in ADAMS 2012 (MSC Software, Santa Ana, CA) during simulations. The cross sectional area of the middle third of the tibia was divided into the following six 60-degree sectors: anterior (AA); latero-anterior (LA); latero-posterior (LP); posterior (PP); medio-posterior (MP); medio-anterior (MA). Average peak principal compressive strains and tensile strains were computed for each sector. Repeated measures ANOVAs were run to determine the differences in bone strains between walking conditions and between tibia bone regions.

RESULTS AND DISCUSSION

Significant differences in bone strains were found among different bone regions ($P < 0.01$). The AA region exhibited the highest compressive strain followed by the MA and LA regions. The PP region exhibited the highest tensile strain followed by the LP and MP regions. Significant differences in bone strains were also observed among different levels of load carriage ($P < 0.01$). Incremented load carriage led to significant increases in bone strains.

Load carriage is an essential task in military training. Army recruits often suffer tibia stress fractures during military training. The cost associated with medical expenses and loss of training days is high. It is critical to develop a better understanding of how tibia bones are loaded and deform during load carriage. This will help design preventative exercise programs for recruits to reduce the incidence of stress fracture during training. In this study, we identified that carrying loads increases tibia bone strains, and incremented load carriage continues to increase bone strain to higher levels. Most importantly, we had identified that certain bone regions experiencing larger mechanical tension or compression than others. Specifically, the anterior and medio-anterior regions of the tibia bone are exposed to significantly high compression and the posterior and latero-posterior regions are exposed to significantly high tension. Experiencing large bone strains in these regions could put the bone at risk of stress fracture. In fact,

these bone regions are commonly reported to be sites of stress fractures in army recruits and athletes [1,4].

Future studies should focus on developing preventative exercise programs to enhance tibia bone mechanical strength for army recruits to lower the incidence of tibia stress fracture.

CONCLUSIONS

Incremented load carriage increases tibia bone strains. Large compressive strains are seen in anterior and medio-anterior regions and large tensile strains are seen in posterior and latero-posterior regions of the tibia.

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Table 1. Tibia bone strains of different regions during load carriage

Regions	Tensile Strains (micro strain)				Compressive Strains (micro strain)			
	00 kg	15 kg	25 kg	35 kg	00 kg	15 kg	25 kg	35 kg
MA	293(65)	346(73)	398(85)	444(112)	654(149)†	817(192)†	933(203)†	1057(227)†
AA	352(90)	415(99)	481(114)	538(144)	845(199)*	1026(269)*	1181(298)*	1361(391)*
LA	344(107)	410(118)	482(142)	537(184)	609(141)	714(186)	839(208)	951(327)
LP	479(136)†	605(165)†	708(183)†	799(198)†	492(131)	582(131)	652(163)	706(171)
PP	682(201)*	834(260)*	965(287)*	1115(380)*	585(198)	702(230)	781(268)	837(268)
MP	426(126)	510(165)	605(186)	684(270)	616(181)	738(202)	827(225)	897(257)

Note. MA: medio-anterior; AA: anterior; LA: latero-anterior; LP: latero-posterior; PP: posterior; MP: medio-posterior. * and † indicate the highest and second highest strains among regions, respectively.

Appendix D

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Title: Influences of Load Carriage and Physical Activity History on Tibial Bone Strain

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Original article

Influences of load carriage and physical activity history on tibia bone strain

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Abstract

Background: Military recruits are often afflicted with stress fractures. The military's strenuous training programs involving load carriage may contribute to the high incidence of tibia stress fractures in the army. The purpose of this study was to assess the influences of incremented load carriage and history of physical activity on tibia bone strain and strain rate during walking.

Methods: Twenty recreational basketball players and 20 recreational runners performed 4 walking tasks while carrying 0 kg, 15 kg, 25 kg, and 35 kg loads, respectively. Tibia bone strain and strain rate were obtained through subject-specific multibody dynamic simulations and finite element analyses. Mixed model repeated-measures analyses of variance were conducted.

Results: The mean $\pm$ SE of the runners' bone strain (μ s) during load carriages (0 kg, 15 kg, 25 kg, and 35 kg) were 658.11 ± 1.61 , 804.41 ± 1.96 , 924.49 ± 2.23 , and 1011.15 ± 2.71 , respectively, in compression and 458.33 ± 1.45 , 562.11 ± 1.81 , 669.82 ± 2.05 , and 733.40 ± 2.52 , respectively, in tension. For the basketball players, the incremented load carriages resulted in compressive strain of 634.30 ± 1.56 , 746.87 ± 1.90 , 842.18 ± 2.16 , and 958.24 ± 2.63 , respectively, and tensile strain of 440.04 ± 1.41 , 518.86 ± 1.75 , 597.63 ± 1.99 , and 700.15 ± 2.47 , respectively. A dose-response relationship exists between incremented load carriage and bone strain and strain rate. A history of regular basketball activity could result in reduced bone strain and reduced strain rate.

Conclusion: In conclusion, load carriage is a risk factor for tibia stress fracture during basic training. Preventative exercise programs, such as basketball, that involved multidirectional mechanical loading to the tibia bones can be implemented for military recruits before basic training commences.

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Keywords: Modeling; Multi-body dynamics; Strain; Strain rate; Stress fracture; Walking

1. Introduction

Stress fracture is a severe musculoskeletal injury requiring extended periods of recovery time and resulting in significant medical costs.^{1–3} Stress fractures are common in sports; the annual injury rate for track athletes has been reported to be 20%.⁴ The rate of stress fractures in the military is also high, reaching 6% in the U.S. Army and as high as 31% in the Israeli Army.^{1,5} The most common site of stress fractures is in the tibia.^{6–8} Tibia stress fractures account for 41%–55% of all stress fractures experienced by athletes involved in running.⁹ In the military, tibia stress fractures account for 50% of all stress fractures in male recruits and 35% in female recruits.^{6–8} The injury mechanism of stress fractures is not well understood.

One hypothesis posits that increased mechanical usage, signified by high strain and high strain rate, stimulates bone remodeling, which results in locally increased porosity and decreased bone mass. Bone mechanical strength is weakened during this stage. When the mechanical loading continues, local stresses are elevated in the bone regions with porosity. If the mechanical loading accumulates at a faster rate than the bone's remodeling process, bone microdamage occurs and microscopic cracks form in the bone,¹ resulting in a stress fracture.^{10,11} Thus, stress fractures may be a result of excessively repetitive loads acting on the bone, leading to fatigue-induced bone microdamage.^{10,11}

A high volume and intensity of training can result in an increased rate of overuse injuries. During physical training in the military and in sports, a high volume and intensity of training is required for trainees, which can lead to an increase in stress fractures. Stress fracture sites may vary among military trainees and those training for sports. However, tibia stress

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fracture has been identified as the most common type among military trainees and track and field athletes.⁶⁻⁹ The high incidence of tibia stress fracture in runners is related to the continuous, repetitive muscular activity and ground impacts.⁴ Similarly, the high rate of tibia stress fracture in the military is associated with the high volume and high intensity of physical training.¹¹⁻¹³ Loaded long-distance walking and running are major parts of the strenuous training protocol during basic training. The combined mileage of load carriage and running during 12 weeks of basic training can exceed 200 miles.¹¹

Several studies have examined the effect of load carriage on walking mechanics.¹⁴⁻²¹ Load carriage results in alterations in gait kinematics and kinetics. Kinematically, loaded walking reduces stride length, increases cadence, and increases pelvic tilt and knee and hip flexion.^{15,17} Kinetically, load carriage increases ankle, knee, and hip joint moment and power.²¹ Studies have also found that walking with additional load results in increased vertical ground reaction forces^{14,16,18} in proportion to the amount of load carried.¹⁷⁻¹⁹ In addition, load carriage results in a pronounced increase in ground reaction force loading rate.²⁰ Increases in ground reaction forces and loading rate combined with increased leg muscle forces present high mechanical stress to the tibia bone. Thus, in theory, tibia bones would be expected to experience elevated strain and increased strain rate during load carriage. However, the effect of load carriage on tibia bone deformation has yet to be determined experimentally. Because incremented load carriage results in increases in ground reaction forces, it is important to examine the tibia bone's response during load carriage by measuring bone strain and strain rate.

Given the significant impact of stress fracture on military and athletic populations, it is essential to determine possible preventative programs. Understanding the effect of past physical activity (PA) on the risk of stress fracture is important. However, findings from the literature are inconclusive. Mustajoki et al.²² did not find a correlation between past PA and stress fracture risk in the military population. Similarly, Giladi et al.²³ and Swissa et al.²⁴ could not establish a relationship between army recruits' PA history and the incidence of stress fractures in basic training. However, Gilbert and Johnson²⁵ reported that recruits with a sedentary lifestyle prior to basic training had a higher risk for stress fracture than those who played sports. Greaney et al.²⁶ found that Marine recruits who participated in running had fewer stress fracture indications than did nonrunners.⁶ Furthermore, Milgrom et al.²⁷ reported that recruits who played ball sports prior to enlistment had fewer incidences of stress fractures than recruits who did not play ball sports. Because it is evident that engaging in sports involving multidirectional impact loading (e.g., volleyball and gymnastics) results in higher bone mineral density than swimming and running,^{28,29} it is speculated that participating in ball sports leads to a general increase of bone mass and strength due to multidirectional impact loadings, which would make tibia bones more resilient to the mechanical loading introduced during basic training. Thus, the risk of stress fracture could be lowered. However, there is no direct evidence that participating in ball sports strengthens tibia bones for load carriage in basic

training. It is essential to determine whether ball sports, and the concomitant multidirectional loadings, help reduce bone deformation (strain and strain rate) during load carriage and lessen the chance of developing stress fracture compared with other sports lacking the pattern of multidirectional impacts (e.g., running).

The traditional approach to determining bone strain *in vivo* is to surgically implant strain gauges on bone surfaces. This approach is invasive and results in pain and discomfort. Thus, the number of subjects willing to participate in research examining bone strain has been very small, ranging from 1 to 6 subjects per study.³⁰⁻³² In recent years, a noninvasive approach has been implemented to examine bone strain *in vivo*.³³⁻³⁵ This method incorporates modeling and computer simulation techniques to perform finite element analysis on bone deformation during PAs. Specifically, a human body model is created based on an individual's gender, height, and weight. Bone structures to be examined are modeled based on medical imaging (e.g., computed tomography (CT) and magnetic resonance imaging). These bone models are then transformed into flexible bodies and incorporated into the human body model for multibody dynamic simulations. This flexible multibody dynamic simulation approach has been validated and was found to be comparable to the traditional strain gauge method.³³⁻³⁵ Given its noninvasive nature, the flexible multibody simulation approach can be used to examine bone strain for large cohorts taking part in PAs.

The purpose of this study was to investigate the influences of incremented load carriage and PA history on tibia bone strain and strain rate during walking by using flexible multibody dynamic simulations. We hypothesized that load carriage would lead to increases in tibia bone strain and in strain rate and that there would be a dose-response relationship between incremented load carriage and bone strain and strain rate. We also expected that PA history would influence the tibia bone strain and strain rate during load carriage. Specifically, we hypothesized that individuals with a history of regular basketball training would have less tibia bone strain and a lower strain rate than those who participated in running.

2. Methods

2.1. Participants

Forty healthy college male participants were recruited to the study. Participants were divided into 2 experimental groups (runners and basketball players). The runner group consisted of 20 recreational runners with a minimum of 2 years of regular running experience (≥ 15 miles per week). The basketball group consisted of 20 recreational basketball players with a minimum of 2 years of regular basketball playing experience (≥ 3 times per week). Participants' demographics are presented in Table 1. Participants were recreationally active, classified as low risk for cardiovascular diseases according to American College of Sports Medicine guidelines,³⁶ and free from known musculoskeletal injury. All participants met the military enlistment standards in terms of physical condition.³⁷ In addition, the age, body mass, body height, and fitness level maximal oxygen

Tibia bone strain during load carriage

3

Table 1
Subject demographic information (mean $\pm$ SD).

	Runners	Basketball players
Age (year)	20.7 $\pm$ 2.4	20.6 $\pm$ 1.7
Body height (cm)	180.1 $\pm$ 6.3	180.8 $\pm$ 8.3
Body mass (kg)	75.7 $\pm$ 9.6	80.1 $\pm$ 11.5
Leg press 1RM (lb)	492 $\pm$ 62	550 $\pm$ 127
VO _{2max} (mL/kg/min)	58 $\pm$ 7*	50 $\pm$ 5

* $p < 0.05$, compared with basketball players.

Abbreviations: 1RM = one-repetition maximum leg press; VO_{2max} = maximum oxygen consumption.

consumption (VO_{2max}) of our participants were comparable to those of military recruits entering U.S. Army Basic Training.³⁸ Approvals from the local Institutional Research Board and Human Research Protection Office at the U.S. Army Medical Research and Materiel Command were obtained prior to commencing the study, and all participants signed the informed consent document prior to testing.

2.2. Experimental protocol

The participants completed 3 data collection sessions: (1) assessment of aerobic and muscular strength, (2) motion capture while performing incremented load carriage, and (3) CT imaging of their tibias.

During the aerobic assessment, participants' VO_{2max} was assessed by means of a ParvoMedics TrueOne 2400 metabolic cart (Parvo Medics Inc., Sandy, UT, USA). A modified ramped version of the Bruce treadmill protocol was used.³⁹ During the muscular strength assessment, the participants' leg muscle strength was measured using a one-repetition maximum (1RM)-effort leg press similar to that described by Hoffman et al.⁴⁰

During motion capture of load carriage, a tandem force-instrumented treadmill (AMTI Advanced Mechanical Technology, Inc., Watertown, MA, USA) with 2 force platforms installed under the tandem belts was used to control the walking speed at 1.67 m/s while allowing the ground reaction forces to be detected. Reflective markers were attached on both sides of the body in the following locations to track the walking movement: acromion, sternum, anterior superior iliac spine, posterior superior iliac spine, lateral knee, lateral ankle, heel, base of the fifth metatarsal, and base of the second toe. In addition, 2 cluster marker sets were attached on the thigh and shank, respectively. Fifteen Vicon MX and F-20s series cameras (Vicon Motion Systems Ltd., Denver, CO, USA) were used to track the reflective markers in the space. Vicon Nexus Version 1.4.116 (Vicon Motion Systems Ltd.) was used to collect kinematic data at 240 Hz and ground reaction forces at 2400 Hz.

Participants wore compression shorts, a compression shirt, and cross-training shoes (Proto Speed Lite Trainer; Under Armour, Baltimore, MD, USA) during the experiment. They walked at a self-selected pace on the force-instrumented treadmill for 5 min to warm-up. The general experimental protocol consisted of the following walking tasks at a speed of 1.67 m/s: (1) 5 min of normal walking without load carriage, (2) 5 min of walking with a 15 kg rucksack (MOLLE; Specialty

Defense System, Dunmore, PA, USA), (3) 5 min of walking with a 25 kg rucksack, and (4) 5 min of walking with a 35 kg rucksack. The loads carried in this study were similar in range to the loads reported in previous studies.^{16,41} A 5 min rest break was given between tasks. Ten trials were collected during the 5 min intervals between each walking condition.

During the CT imaging session, axial plane scans of the tibia bones were obtained using a GE Light Speed VCT scanner (GE Healthcare; General Electric Company, Chicago, IL, USA). CT tube potential was set at 120 kV, and the tube current-time product was 144.54 mAs. The slice thickness was set at 0.625 mm. The field of view was 15 cm $\times$ 15 cm.

2.3. Computer modeling and simulations

CT images of tibias were segmented in Mimics 14 (Materialise NV, Leuven, Belgium). A surface mesh of the tibia was generated. Solid hexahedral meshing was performed in Marc 2013 (MSC Software Corp., Santa Ana, CA, USA) with an element size of 3 mm³. Bone density of individual elements was determined based on the average Hounsfield unit value of the pixels contained in the element. Young's modulus was calculated based on the element's density. Poisson ratio was set at 0.3. Individual element's density, Young's modulus, and Poisson ratio were assigned to each element through a custom MATLAB program (Mathworks Inc., Natick, MA, USA). The tibia bone model prepared in Marc 2013 was then converted into a modal-neutral file representing a flexible tibia. This subject-specific flexible tibia model was later used in a forward dynamic simulation of the walking movement to assess tibia bone strain.^{33–35}

Motion capture data were processed in Visual 3D Version 4.0 (C-Motion Inc., Germantown, MD, USA). Kinematic data and ground reaction forces data were filtered using a zero-lag fourth-order Butterworth filter with a cutoff frequency of 8 Hz and 40 Hz, respectively. Subjects' lower extremity musculoskeletal models, including generic geometries of pelvis, femur, tibia, and foot, were created in LifeMOD 2012 (LifeModeler Inc., San Clemente, CA, USA), which is a plug-in program in ADAMS 2012 (MSC Software Corp.). The generic lower extremity model was then scaled based on the subject's mass, height, gender, and age, as well as the relative positions of the ankle, knee, and hip joints determined from the kinematic data. The flexible tibia represented by the modal-neutral file was introduced to replace the generic tibia in the musculoskeletal model in ADAMS 2012 (MSC Software Corp.) through the LifeMOD plug-in. Tri-axis hinges combined with passive torsional spring-dampers (1 Nm/° and 0.1 Nm/s/°) were employed to model the hip joints. A hinge joint with a single degree of freedom was used for knee and ankle joints in the sagittal plane. A total of 90 muscles were assigned to the legs. The kinematics collected during walking tasks were used to drive the musculoskeletal model with an inverse dynamics algorithm while the muscles' shortening/lengthening patterns were recorded.^{33–35} Subsequently, kinematic constraints were removed, and a forward dynamics simulation was performed with muscles serving as actuators and experimental ground reaction forces applied to replicate the walking motion.

Table 2
Effect sizes (partial η^2) associated with strain and strain rate.

Variables	Load carriage effect	Past physical activity effect
Compressive strain	0.553	0.008
Tensile strain	0.467	0.005
Shear strain	0.589	0.011
Compressive strain rate	0.331	0.003
Tensile strain rate	0.312	0.001
Shear strain rate	0.368	0.004

A proportional-integral-derivative feedback controller was implemented to calculate each muscle force magnitude using the error signal between the current muscle length in the forward dynamics and the recorded muscle length during the inverse dynamics simulation.^{42–44} The force generated by the individual muscle was limited by its maximum force-generating potential.

Tibia bone strain was then calculated using the Durability plug-in in ADAMS 2012. Bone strain and strain rate from the surface of the middle third of the tibia were extracted for analysis. Because other bone strain studies commonly reported the standard error (SE) of the mean to indicate how accurate the estimate of the mean was likely to be,^{45–47} SEs were reported along with the means of the bone strain data in this study.

2.4. Statistical analysis

SPSS Version 19 (IBM, Armonk, NY, USA) was used to perform statistical analyses. The following dependent variables were examined: peak tensile strain (peak maximal principle strain) and strain rate, peak compressive strain (peak minimal principle strain) and strain rate, and peak shear strain (peak maximal shear strain) and strain rate during the stance of walking. For each of the dependent variables, a mixed model repeated-measures analysis of variance test was run to determine the effects of subject group (runners vs. ball players) and

load carriage (4 levels: 0 kg, 15 kg, 25 kg, and 35 kg) on bone strain and strain rate. Statistical effect size (partial η^2) was computed using the following formula: sum of squares of the effect/(sum of squares of the effect + sum of squares of the error).⁴⁸ Significance level was set at 0.05.

3. Results

For all the dependent variables examined, significant interactions were found between the subject group and the incremented load carriage ($p = 0.0001$). Therefore, separate analysis of variance tests were run to determine the simple effects from the PA history and incremented load carriage on tibia strain and strain rate. Effect sizes associated with each dependent variable are presented in Table 2.

3.1. Tibia bone strain

One-way repeated-measures analyses of variance were run to examine the effects of the incremented load carriage on tibia bone strain for the runner group and the basketball player group, respectively. For both groups, significant differences in peak compressive strain, tensile strain, and shear strain (all $p \leq 0.0001$) were found among loaded walking tasks. Specifically, a dose-response relationship between the incremented load carriage and the tibia strain existed. As the load carriage increased, tibia compressive, tensile, and shear strain increased. Table 3 presents the mean $\pm$ SE values of the peak compressive, tensile, and shear strain of the middle third of the tibia during loaded walking.

One-way repeated-measures analyses of variance were run to examine the effects of PA history on the tibia bone strain for each of the loaded walking tasks. Significant differences in peak compressive strain, tensile strain, and shear strain (all $p \leq 0.0001$) were found between the 2 groups. The runners consistently exhibited greater peak compressive, tensile, and shear strain than the basketball players.

Table 3
Tibia bone strain and strain rate during different loaded walking tasks with load carriages of 0 kg, 15 kg, 25 kg, and 30 kg (mean $\pm$ SE).

Variables/Conditions	0 kg	15 kg	25 kg	35 kg
Tibia bone strain (μs)				
<i>Runners</i>				
Compressive strain	658.11 $\pm$ 1.61 ^{##}	804.41 $\pm$ 1.96 ^{***##}	924.49 $\pm$ 2.23 ^{***##}	1011.15 $\pm$ 2.71 ^{***##}
Tensile strain	458.33 $\pm$ 1.45 ^{##}	562.11 $\pm$ 1.81 ^{***##}	669.82 $\pm$ 2.05 ^{***##}	733.40 $\pm$ 2.52 ^{***##}
Shear strain	1003.29 $\pm$ 2.02 ^{##}	1229.74 $\pm$ 2.40 ^{***##}	1444.68 $\pm$ 2.72 ^{***##}	1586.67 $\pm$ 3.48 ^{***##}
<i>Basketball players</i>				
Compressive strain	634.30 $\pm$ 1.56	746.87 $\pm$ 1.90 ^{**}	842.18 $\pm$ 2.16 ^{**}	958.24 $\pm$ 2.63 ^{**}
Tensile strain	440.04 $\pm$ 1.41	518.86 $\pm$ 1.75 ^{**}	597.63 $\pm$ 1.99 ^{**}	700.15 $\pm$ 2.47 ^{**}
Shear strain	972.28 $\pm$ 1.96	1155.32 $\pm$ 2.32 ^{**}	1309.96 $\pm$ 2.64 ^{**}	1519.01 $\pm$ 3.37 ^{**}
Tibia bone strain rate (μs/s)				
<i>Runners</i>				
Compressive strain rate	7955.99 $\pm$ 24.08 [#]	9477.93 $\pm$ 27.04 ^{***##}	11,562.32 $\pm$ 33.26 ^{***##}	11,583.09 $\pm$ 37.50
Tensile strain rate	6647.40 $\pm$ 20.07 ^{##}	7901.71 $\pm$ 23.27 ^{***##}	9551.94 $\pm$ 28.28 ^{***##}	9617.21 $\pm$ 33.14 ^{***##}
Shear strain rate	6668.60 $\pm$ 13.30 ^{##}	7828.50 $\pm$ 15.27 ^{***##}	9594.68 $\pm$ 20.05 ^{***##}	9662.64 $\pm$ 24.26 ^{***##}
<i>Basketball players</i>				
Compressive strain rate	7882.92 $\pm$ 23.34	9034.16 $\pm$ 26.22 ^{**}	10,293.25 $\pm$ 32.23 ^{**}	11,661.34 $\pm$ 36.36 ^{**}
Tensile strain rate	6557.76 $\pm$ 19.46	7432.32 $\pm$ 22.57 ^{**}	8632.39 $\pm$ 27.42 ^{**}	10,012.88 $\pm$ 32.13 ^{**}
Shear strain rate	6540.35 $\pm$ 12.90	7488.80 $\pm$ 14.80 ^{**}	8609.44 $\pm$ 19.44 ^{**}	9976.34 $\pm$ 23.53 ^{**}

* $p < 0.05$, ** $p < 0.0001$, compared with previous level of the loaded walking condition; [#] $p < 0.05$, ^{##} $p < 0.0001$, compared with basketball players.

3.2. Tibia bone strain rate

One-way repeated measures analyses of variance were run to examine the effects of the incremented load carriage on tibia bone strain rate for the runner group and the basketball player group, respectively. For both groups, significant differences in compressive strain rate, tensile strain rate, and shear strain rate (all $p \leq 0.0001$) were found between loaded walking tasks. Both the ball players and the runners demonstrated significant increases in peak compressive, tensile, and shear strain rate as load carriage increased from 0 kg to 15 kg, from 15 kg to 25 kg, and from 25 kg to 35 kg. The only exception was that the runners did not show a further increase in compressive strain rate when load carriage was increased beyond 25 kg. Table 3 presents the mean $\pm$ SE values of the peak compressive, tensile, and shear strain rate of the middle third of the tibia during loaded walking tasks.

One-way analysis of variance tests were run to examine the effect of PA history on tibia strain rate for each of the loaded walking tasks. Significant differences in peak compressive strain rate, tensile strain rate, and shear strain rate (all $p \leq 0.0001$) were found between the runners and basketball players. Specifically, the runners demonstrated greater strain rate than the ball players when carrying 15 kg and 25 kg loads. However, when load carriage was increased to 35 kg, the runners either showed no difference in strain rate (peak compressive strain rate, $p = 0.134$) or showed less strain rate (tensile strain rate and shear strain rate: both $p \leq 0.0001$) compared with the ball players.

4. Discussion

The purpose of the study was to assess the effects of incremented load carriage and PA history on tibia bone strain and strain rate during walking. A group of recreational runners and a group of recreational basketball players participated in the study. Incremented load carriage was introduced at the levels of 15 kg, 25 kg, and 35 kg. Flexible multibody dynamic simulations were performed to obtain tibia bone strain. It was found that significant differences in bone strain and strain rate existed between the runners and the basketball players and also among loaded walking conditions.

We hypothesized that load carriage would increase tibia bone strain. This hypothesis was supported. Compared with the unloaded walking, the 15 kg load carriage resulted in significant increases in tibia bone strain. For runners, the increases were 22%, 23%, and 23% in compressive, tensile, and shear strain, respectively. For ball players, they were 18%, 18%, and 19% in compressive, tensile, and shear strain, respectively. We further hypothesized that a dose-response relationship would exist between incremented load carriage and tibia bone strain. This hypothesis was also supported. We observed that as load carriage was increased to 35 kg from a 15 kg level in 10 kg increments, tibia bone strain continued to rise. High bone strain may trigger a bone remodeling process for building a strong bone to better resist the imposed higher level of mechanical loading. During basic training, military recruits regularly engage in prolonged walking with load carriage. The

amount of load carriage has been reported to be as high as 61 kg.¹³ The combined loaded walking and running distance during 12 weeks of basic training can reach 200 miles.¹¹ Repetitive high mechanical loading imposed on the tibia may increase the risk of developing stress fractures. The dose-response relationship between incremented load carriage and bone strain indicates that the risk of stress fracture could be even higher when heavy loads are carried.

We hypothesized that load carriage could lead to an increase in bone strain rate. Our hypothesis was supported. When compared with unloaded walking, carrying a 15 kg load resulted in significant increases in strain rate. For the runners, the increases were 19%, 19%, and 17% in compressive, tensile, and shear strain rate, respectively. For ball players, the increases were 15%, 13%, and 15% in compressive, tensile, and shear strain rate, respectively. We further hypothesized that there would be a dose-response relationship between the incremented load carriage and tibia bone strain rate. This hypothesis was partially supported. We observed that ball players exhibited increases in bone strain rate when load was increased from 15 kg to 35 kg in 10 kg increments. The runners also showed increases in bone strain rate when load was increased from 15 kg to 25 kg. Runners' tensile and shear strain rate continue to increase when load carriage reached 35 kg. However, runners' compressive strain rate remained unchanged when load was increased from 25 kg to 35 kg. Strain rate is a primary mechanical stimulus for bone remodeling.^{4,49} Bone is a viscoelastic tissue and is sensitive to strain rate.^{50–52} High strain rate could elicit bone remodeling processes for improving bone strength.² On the one hand, bone can remodel itself and become stronger to sustain the amount of mechanical load imposed if there is enough time for the remodeling process to be completed. On the other hand, if the bone continues to experience high strain rate in the middle of the remodeling process, microdamage may occur and lead to stress fracture. In this study, the increased strain rate owing to increased load carriage reflected an increased risk of tibia stress fracture. During military basic training, the loaded walking exercise is intense and introduces high strain rate to the tibia bones; thus, it is likely that the risk of tibia stress fracture will be elevated. Interestingly, runners in this study exhibited similar compressive strain rate when carrying 25 kg and 35 kg loads. It is unclear why this might occur. A possible explanation is that when the strain rate reaches a critical level that may be harmful to the bone, the body may regulate forces acting on the tibia to prevent a further increase in the strain rate at specific regions to avoid injury. Based on the current findings, a load carriage of 25 kg or greater may be unsafe to the tibias of runners and may result in a regulation of compressive strain rate. Because the basketball players in this study continued to experience increases in strain rate through the incremented load carriage, it is possible that their tibias were capable of sustaining heavy mechanical loads during load carriage training. Additional studies are needed to examine how strain rate is regulated as load carriage increases.

The effect of PA history on bone quality has been studied on a limited basis. One study reported that athletes engaged in impact-loading sports such as volleyball and gymnastics had

higher bone mineral density than swimmers and control subjects.²⁸ In addition, another study found that runners, swimmers, and divers had deficits in bone mineral density compared with gymnasts and softball players.²⁹ Furthermore, research demonstrated that a 14-week recreational soccer training program led to greater increases in tibia bone density among participants compared with runners and controls.⁵³ However, the effect of PA history on tibia bone strain and strain rate has not been fully determined. In this study, we hypothesized that during incremented load carriage the recreational basketball players would show less tibia bone strain compared with the runners. This hypothesis was supported. Compared with the runners, when carrying a 15 kg load, the basketball players exhibited less compressive strain (7% less), tensile strain (8% less), and shear strain (6% less). When carrying a 25 kg load, the ball players showed smaller bone strain in compression (9% less), tension (11% less), and shear (9% less). Furthermore, when load carriage was increased to 35 kg, the ball players continued to show less bone strain in compression (5% less), tension (5% less), and shear (4% less). These results indicate that the history of PA influences tibia bone strain. Individuals with a history of basketball training had less tibia bone strain compared with those with a history of running.

We further hypothesized that the basketball players would show a lower bone strain rate than that of runners during incremented load carriage. This hypothesis was partially supported. Compared with the runners, when carrying a 15 kg load, the ball players showed lower strain rate in compression (5% less), tension (6% less), and shear (4% less). When the load carriage increased to 25 kg, the ball players continued to show less strain rate in compression (11% less), tension (10% less), and shear (10% less). However, when the load carriage reached 35 kg, the runners showed no difference in compressive strain rate and lower strain rate in tension (4% less) and in shear (3% less) than those of the basketball players. It is speculated that a threshold in load carriage exists for individuals. When the load carriage results in a high level of strain rate that might be harmful to the tibia bone, the human body may adjust the individual's walking movement and regulate forces applied to the tibia to lower the strain rate to avoid injury. In this study, the runners may have experienced a threshold of strain rate when carrying a 25 kg load. Because ball players continued to show increases in strain rate when load carriage increased, it appears that the ball players' tibias were resilient to increased strain rate and could sustain mechanical loads of 35 kg or more. Future studies should examine whether the threshold effect of bone strain rate is accompanied with alterations of tibia bone strain distribution.

In this study, we confirmed that past PA has an effect on bone strain and strain rate. Our findings are consistent with previous studies investigating the effect of exercise on bone quality. Running movement introduces cyclic and uniaxial mechanical loading to the tibia bones. Over time, tibia bones adapt to the mechanical loading imposed on them by making some regions of the bone more dense and resilient to loading. However, the overall tibia bone density may not increase much because it is less metabolically costly to maintain a lighter bone. It was

reported that a 14-week running exercise program did not effectively increase tibia bone density.⁵³ However, a 14-week soccer training program did result in a significant increase in tibia bone density.⁵³ It was surmised that the movements used in soccer, such as acceleration, deceleration, jumping, and cutting, produced mechanical stimuli to tibia bones from different directions and contributed to changes in bone mass.⁵³ Similar to soccer, basketball introduces multiaxial mechanical loading to the tibia bone and could lead to positive osteogenesis along the whole bone. Subsequently, overall bone strength is improved. In this study, we observed that basketball players' tibia bones were stronger and that the players experienced less strain and a lower strain rate in their tibias than did runners during load carriage.

In this study, we recruited recreational runners and basketball players. In general, these participants did not participate in routine weight-training exercises. Although there was no statistical difference in maximal leg press between the groups, the greater leg strength exhibited by the ball players could be a result of playing basketball regularly, which comprises running, jump-landing, cutting, and sudden accelerations and decelerations of the body. In addition, the greater $\dot{V}O_{2\max}$ associated with runners appeared to be a result of their habitual running activity.

5. Conclusion

In summary, load carriage resulted in increases in bone strain and strain rate. A dose-response relationship existed between incremented load carriage and bone strain and strain rate. History of PA influenced bone strain and strain rate during load carriage. A history of regular basketball activity resulted in reduced bone strain and strain rate. Therefore, military administrators and recruits need to be aware that load carriage is a risk factor for tibia stress fracture during basic training. Preventative exercise programs involving multidirectional mechanical loading to the tibia bones, such as basketball and soccer, should be implemented for recruits before basic training. Furthermore, track coaches and distance runners should recognize the importance of strengthening the tibia bones to reduce the incidence of stress fractures. Runners should consider regularly engaging in exercises that involve multiaxial mechanical loadings to the lower extremity bones.

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Authors' contributions

HW carried out the study; participated in data collection, data processing, and data analysis; and drafted the manuscript. MK participated in data processing and data analysis and helped draft the manuscript; DCD participated in data collection and data analysis and helped draft the manuscript. All authors have read and approved the final version of the manuscript, and agree with the order of presentation of the authors.

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Competing interests

None of the authors declare competing financial interests.

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Appendix E

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Title: The Effect of Drop-landing Height on Tibia Bone Strain

The Effect of Drop-Landing Height on Tibia Bone Strain

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Abstract

Athletes and military recruits are often afflicted by stress fractures. Rigorous training programs consisting of increased repetitive mechanical loading may contribute to the high incidence of tibia stress fracture in the athletic and army populations. The purpose of this study was to assess the effect of incremented height on tibia bone strains and strain rates during landing. Seven healthy college males performed drop-landing tasks from 26 cm, 39 cm, and 52 cm, respectively. Tibia bone strains and strain rates were obtained through subject-specific multi-body dynamic computer simulations and finite element analyses. One-way repeated measures ANOVAs were conducted. Both 39 cm and 52 cm conditions resulted in larger tibia bone strains and strain rates than the 26 cm condition. The 52 cm condition also resulted in greater bone strains and strain rates than the 39 cm condition. A dose-response relationship exists between incremented landing height and bone strains and strain rates. Activities consisting of high impact landings are associated with increased risk of developing tibia stress fracture. When designing training programs involved high impact activities, athletes and military recruits should consider the effect of impact loading on tibia bone health and giving enough time for bones to adapt to new trainings.

Keywords

Stress Fracture, Bone Strain, Drop-Landing, Tibia Bone

1. Introduction

Stress fracture is a severe musculoskeletal injury requiring extended periods of recovery and incurring significant medical cost [1] [2]. Stress fractures are common in sports. Nearly 10% of all sports related injuries are stress fractures [3]. Track athletes experience an even higher annual injury rate at 20% [4]. Si-

Similarly, the rate of stress fractures in the military is also high. It reaches 6% in the US Army and is 31% in the Israeli Army [1] [5]. The most common site of stress fracture is tibia [6] [7]. Tibia stress fracture accounts for 41% - 55% of all stress fractures experienced by athletes involved in running [8]. In the military, tibia stress fracture accounts for 50% and 35% of all stress fractures in male and female recruits, respectively [6] [7].

Although the injury mechanism of stress fracture is not well understood, one possible hypothesis explains that increased mechanical usage, signified by high strains and strain rates, stimulates bone remodeling, which results in locally increased porosity and decreased bone mass. Bone mechanical strength is weakened during this stage. When the mechanical loading continues, local stresses are elevated in the bone region with porosity. If the mechanical loading accumulates at a faster rate than the bone's remodeling process, bone microdamages would occur and microscopic cracks would be formed and propagated in the bone [1]. A stress fracture would appear [9] [10]. Thus, stress fracture could be a result of excessively repetitive loads acting on the bone leading to fatigue induced bone microdamages [9] [10].

Rapid changes in training with increased volume and intensity result in an increased rate of overuse injuries. The high incidence of tibia stress fracture in runners is related to the continuous, repetitive muscular activity and increased ground impacts [4]. Similarly, the high rate of tibia stress fracture in the military is associated with the increased volume and high intensity of physical training [10] [11] [12], during which, recruits normally cover 200 miles of walking and running with load carriage in less than 12 weeks [10].

Landing is a basic component of many sports and physical activities. When the foot lands on the ground, there is an impact force introduced to the body. The body's ability to dissipate this landing impact will affect the stress and strain experienced by the various tissues of the body. The skeletal system, working mechanically as a series of levers, is responsible for much of this dissipation [13]. In situations where the musculoskeletal system cannot adequately absorb the impact mechanically, bone must dissipate the residual force. Absorption of residual force can cause microdamages in the bone tissue. These microscopic fractures will accumulate and may result in stress fractures. The tibia is a primary concern when examining stress fracture incidence as it has been identified as one of the most common locations of stress fracture in both military and athletic populations [5] [14] [15] [16]. Currently, the effect of ground impact forces on tibia bone's mechanical reaction is not well understood, one research question has yet to be answered is how tibia bone responds to the mechanical stress introduced by high impact activities.

Therefore, the purpose of this study was to investigate the effect of landing height on tibia bone strains and strain rates during drop-landing movements. We hypothesized that increased landing height would lead to increases in tibia bone strains and strain rates. Also, there would be a dose-response relationship between incremented height and bone strains and strain rates.

2. Methods

Seven healthy college male participants were recruited to the study. The means and standard deviations (SDs) of age, body mass, body height, one-rep max (1 RM) effort leg press, and maximal oxygen consumption ($\text{VO}_2 \text{ max}$) of participants were 21(2) yrs, 75.3(5.3) kg, 180(5) cm, 2217(471) N, and 51.5(5.9) $\text{ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$, respectively. Participants were recreationally active, classified as low risk for cardio-vascular diseases according to ACSM guidelines [17], and free from known musculoskeletal injury. All participants met the military enlistment standards in terms of physical conditions. In addition, the age, body mass, body height, and fitness level ($\text{VO}_2 \text{ max}$) of our participants were comparable to those military recruits entering the US Army Basic Training [18]. Approval from local Institutional Review Board (IRB) was obtained prior to commencing the study. Participants signed the informed consent before testing.

The participants completed three data collection sessions: 1) assessment of aerobic and muscular strength, 2) motion capture while performing drop-landing tasks, and 3) computed tomography imaging of their tibias.

During the aerobic assessment, participants' $\text{VO}_2 \text{ max}$ was assessed through a ParvoMedics metabolic cart (ParvoMedics, Sandy, UT). A modified ramped version of the Bruce treadmill protocol was used. During the muscular strength assessment, the participants' leg muscle strength was measured using a 1 RM effort leg press.

During motion capture of drop-landing movement, A 14 camera (Vicon F-series, Vicon, Oxford, UK) motion capture system and two AMTI multi-axis force platforms (Advanced Mechanical Technology, INC. Watertown, MA) controlled by Vicon Workstation software were used. Kinematic data were collected at 120 Hz. Kinetic data were collected at 2400 Hz.

Reflective markers were attached on both sides of the body in the following locations to track the drop-landing movement: Acromion, sternum, anterior superior iliac spine, posterior superior iliac spine, lateral knee, lateral ankle, heel, base of the fifth metatarsal, and base of the second toe. In addition, two cluster marker sets were attached on the thigh and shank, respectively.

Participants wore compression shorts and a compression shirt during the experiment. Participants walked at a self-selected pace on a treadmill for five minutes to warm up. Participants performed the drop-landing movement with the non-dominant foot on the box and the dominant foot suspended directly in front of the box. To begin the movement, the participant shifted his weight forward and stepped off from the height. The participant landed with each foot on a separate force plate simultaneously. The participant was instructed to land and remain standing until the research team completed capturing the trial. The participant performed landings at each height: 26, 39, and 52 cm [19]. Landing conditions were randomized. Three acceptable trials were collected from each height.

During CT imaging session, axial plane scans of the tibia bones were obtained using a GE Light Speed CT scanner (General Electric, WI). The slice thickness was set at 0.625 mm. The field of view was 15 cm $\times$ 15 cm.

CT images of tibias were segmented in Mimics 13.1 (Materialise, Belgium). A surface mesh of the tibia was generated. Solid hexahedral meshing was performed in MD Marc (MSC Software, Santa Ana, CA) with an element size of 3 mm³. Material properties were assigned assuming that bone is transversely isotropic within the ranges of strain we evaluated [20] [21] [22]. A Young's Modulus of 17 GPa and Poisson's Ratio of 0.3 were used based on previously reported values [23]. The tibia bone model prepared in MD Marc was then converted into a modal neutral file (MNF) representing a flexible tibia. This subject specific flexible tibia model was later used in a forward dynamic simulation of the drop-landing movement in order to assess tibia bone strain.

Motion capture data were processed in Visual 3D™ v4 (C-Motion, Germantown, MD, USA). Kinematic data and ground reaction forces data were filtered using a zero-lag fourth order Butterworth filter with a cutoff frequency of 10 Hz and 50 Hz, respectively. Subjects' lower-extremity musculoskeletal models including generic geometrics of pelvis, femur, tibia, and foot were created in LifeMod 2010 (LifeModeler Inc. San Clemente, CA), which is a plug-in program in ADAMS 2010 (MSC Software, Santa Ana, CA). The generic lower-extremity model was then scaled based on subject's mass, height, gender, and age as well as the relative positions of the ankle, knee, and hip joints determined from the kinematic data. The flexible tibia represented by the MNF file was introduced to replace the generic tibia in the musculoskeletal model in ADAMS 2010 (MSC Software, Santa Ana, CA). Tri-axis hinges combined with passive torsional spring-dampers (1 Nm/° and 0.1 Nm·s/°) were employed to model the hip joints. A hinge joint with a single degree of freedom was used for knee and ankle joints in the sagittal plane. The right side of the model was actuated by 17 muscles, while the left side was controlled using motion-replicating servo controlled joints. The kinematics collected during drop-landing tasks were used to drive the musculoskeletal model with an inverse dynamics algorithm while muscles' shortening/lengthening patterns were recorded [23]. Subsequently, kinematic constraints were removed and a forward dynamics simulation was performed with muscles serving as actuators and experimental ground reaction forces applied to replicate the drop-landing motion.

A proportional-integral-derivative feedback controller was implemented to calculate each muscle force magnitude using the error signal between the current muscle length in the forward dynamics and the recorded muscle length during the inverse dynamics simulation [24]. The force generated by individual muscle was limited by its maximum force generating potential.

Durability plug-in in ADAMS 2010 was used to measure bone strain during dynamic simulations. **Figure 1** describes a drop-landing simulation performed by a subject-specific lower-extremity skeletal model and bone strains experienced by the flexible tibia. Previous research suggests that most bone strain occurs at about 3 Hz [25]. As a result, the strain data in the current study were filtered using a second order, low-pass, Butterworth filter with a cutoff frequency of 5 Hz. The strain values were obtained from the location previously described:

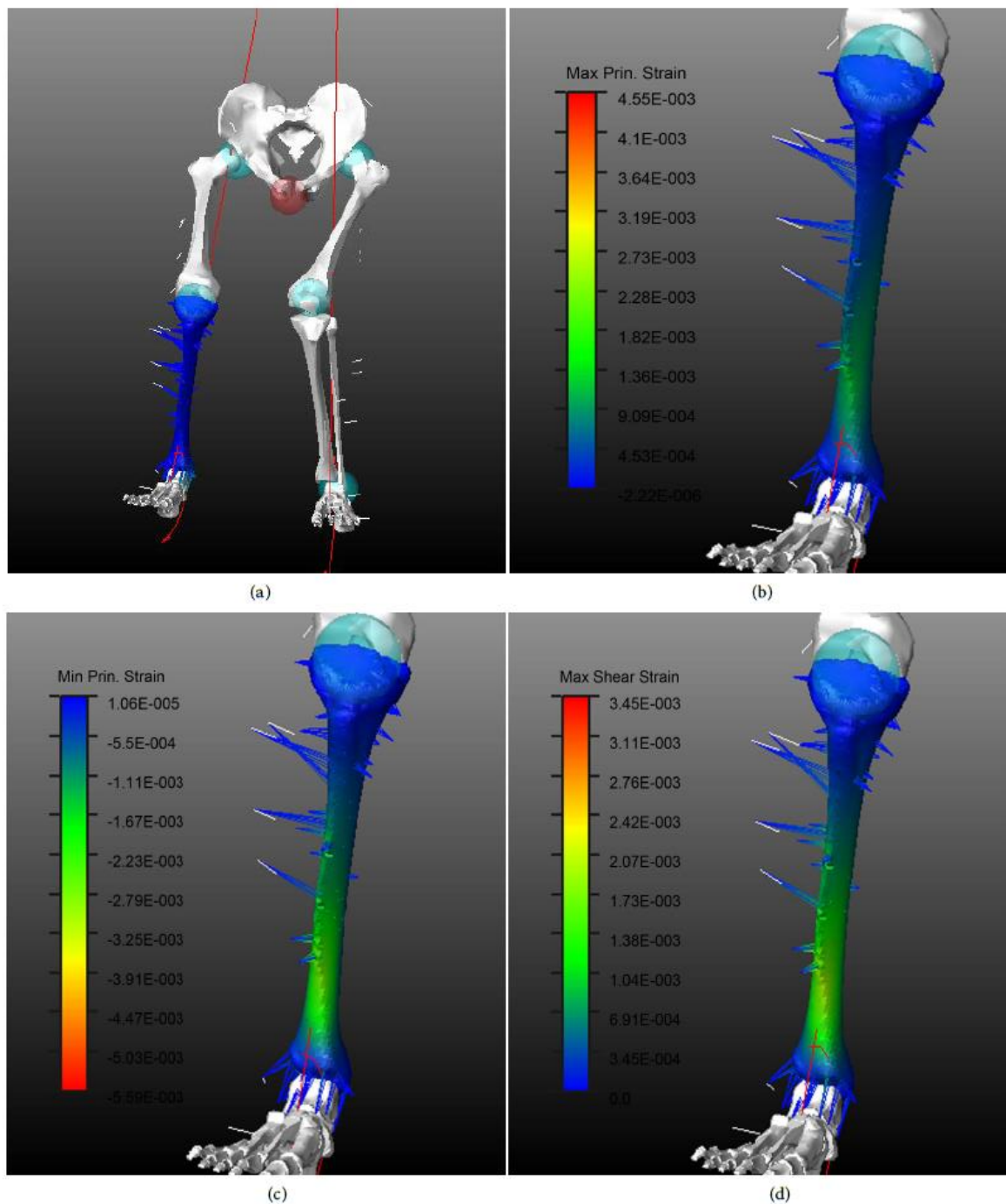


Figure 1. (a) Drop-landing simulation performed by a lower-extremity skeletal model with a right flexible tibia. (b) Maximal principal tensile strain of the flexible tibia at landing. (c) Minimal principal compressive strain of the flexible tibia at landing. (d) Maximal shear strain of the flexible tibia at landing.

the medial aspect of the mid-tibial diaphysis [19].

SPSS v19 (IBM Inc. Chicago, IL, USA) was used to perform statistical analysis. The following dependent variables were examined: peak tensile strain (peak maximal principle strain) and strain rate, peak compressive strain (peak minimal

principle strain) and strain rate, and peak shear strain (peak maximal shear strain) and strain rate during drop-landing tasks. For each of the dependent variables, a repeated measures analysis of variance (ANOVA) test was run to determine the effect of drop-landing height on bone strains and strain rates. Significance level was set at 0.05.

3. Results

One-way repeated measures ANOVAs were run to examine the effect of the drop-landing height on tibia bone strains. Significant differences in peak tensile strain ($p = 0.001$), compressive strain ($p = 0.001$), and shear strain ($p = 0.0001$) were observed among drop-landing tasks. As the height increased, tibia strains increased. Specifically, the 39 cm condition elicited larger increases in tensile strain ($p = 0.001$), compressive strain ($p = 0.002$), and shear strain ($p = 0.001$) from the 26 cm condition. Similarly, the 52 cm condition resulted in greater increases in tensile strain ($p = 0.001$), compressive strain ($p = 0.001$), and shear strain ($p = 0.001$) than the 26 cm condition. In addition, the 52 cm condition resulted in greater increases in tensile strain ($p = 0.073$), compressive strain ($p = 0.005$), and shear strain ($p = 0.033$) than the 39cm condition. **Table 1** presents the means and standard deviations (SDs) of the peak tensile, compressive, and shears strains of the tibia during drop-landings.

One-way repeated measures ANOVAs were run to examine the effect of the drop-landing height on tibia bone strain rates. Significant differences in tensile strain rate ($p = 0.006$), compressive strain rate ($p = 0.005$), and shear strain rate ($p = 0.001$) were found between drop-landing tasks. As the height increased, tibia tensile, compressive, and shear strain rates increased. Specifically, the 39 cm condition resulted in larger increases in tensile strain rate ($p = 0.034$), compressive strain rate ($p = 0.066$), and shear strain rate ($p = 0.013$) than those of the 26 cm condition. Similarly, the 52 cm condition led to larger increases in tensile strain rate ($p = 0.006$), compressive strain rate ($p = 0.005$), and shear strain rate ($p = 0.001$) than those of the 26 cm condition. In addition, the 52 cm condition led to larger increases in tensile strain rate ($p = 0.001$), compressive strain rate ($p = 0.001$), and shear strain rate ($p = 0.001$) than those of the 39 cm condition. **Table 2** presents the means and SDs of the peak tensile, compressive, and shears strain rates of the tibia during drop-landings.

4. Discussion

The purpose of the study was to assess the effect of incremented landing height

Table 1. Means (SDs) of tibia bone strain during drop-landing tasks.

Variables/Conditions	26 cm	39 cm	52 cm
Tensile strain (μs)	1348 (524)	1645 (593)*	1796 (597)
Compressive strain (μs)	654 (195)	797 (253)*	879 (256)*
Shear strain (μs)	988 (334)	1204 (397)*	1321 (401)*

*indicates a significant difference ($p \leq 0.05$) from the previous drop-landing condition.

Table 2. Means (SDs) of tibia bone strain rates during drop-landing tasks.

Variables/Conditions	26 cm	39 cm	52 cm
Tensile strain rate ($\mu\text{s/s}$)	13,808 (5329)	18,471 (7405)*	21,607 (7980)*
Compressive strain rate ($\mu\text{s/s}$)	7403 (2666)	9296 (3655)	10,913 (3733)*
Shear strain rate ($\mu\text{s/s}$)	11,115 (3858)	13,683 (5221)*	16,063 (5524)*

Significant differences between the current drop-landing condition and previous drop-landing condition were indicated (*indicates $p \leq 0.05$).

on tibia bone strain and strain rate during drop-landing. A group of healthy males took part in the study. Incremented height was introduced at the levels of 26 cm, 39 cm, and 52 cm. It was found that significant differences in bone strain and strain rate exist among landing conditions.

We had hypothesized that increasing landing height would increase tibia bone strain. This hypothesis was supported. Compared to the 26 cm condition, the 39 cm condition resulted in significant increases of tibia bone strain. There were 18.4%, 21.9%, and 21.9% increases in tensile, compressive, and shear strains, respectively. Compared to the 26 cm condition, the 52 cm condition showed significant increases in tibia bone strain. There were 33.2%, 34.4%, and 33.7% increases in tensile, compressive, and shear strains, respectively. In addition, the 52 cm condition also resulted in greater increases in tensile (9.2% more), compressive (10.3% more), and shear (9.7% more) strains than the 39 cm condition. As the increases in bone strains corresponded to the increases in landing height, a dose-response relationship between incremented heights and tibia bone strains was revealed. Increased bone strains could trigger bone remodeling process for building a strong bone to better adapt to the applied mechanical loading [26]. Bone remodeling process could be accelerated if bone continues to experience large strains. Thus, increased mechanical loading on the tibia during training may increase risk of developing stress fracture. The dose-response relationship between incremented heights and bone strain indicates that the risk of stress fracture could be even higher when training consists of high impact loadings.

We had hypothesized that incremented heights could lead to increases in bone strain rates. Our hypothesis was supported. When compared to the 26 cm condition, the 39 cm condition resulted in significant increases in strain rates. There were 33.8%, 25.6%, and 20.4% increases in tensile, compressive, and shear strain rates, respectively. Also, compared to the 26 cm condition, the 52 cm condition showed 58.0%, 47.4%, and 44.5% increases in tensile, compressive, and shear strain rates, respectively. Furthermore, the 52 cm condition exhibited higher tensile (11.6% more), compressive (17.4% more), and shear (17.4% more) strain rates than those of the 39 cm condition, respectively. In this study, a dose-response relationship between the incremented heights and tibia bone strain rates was evident. We observed that increases in bone strain rates corresponded to increases in landing heights. As bone is a viscoelastic tissue, which is sensitive to strain rate [27] [28] [29], its remodeling process is thought to be heavily influenced by strain rate [4] [30]. High strain rates could elicit bone re-

modeling process for improving bone strength [27]. On one hand, bone can remodel itself and become stronger to resist high impact loading if enough time is given for the remodeling process to complete. On the other hand, if the bone continues to experience high strain rate during the remodeling process, micro-damages may occur and evolve into stress fracture. In this study, the increased strain rates from increased impact loadings reflect an increased risk of tibia stress fracture. During athletic training or military basic training, if changes in training programs consist of unaccustomed high impact loading to the body, the tibias could experience increased strain rates and be exposed to increased risk of stress fracture.

Interestingly, Milgrom *et al.* also attempted to examine the effect of landing height on tibia bone strains [19]. However, the authors were not able to find statistical differences in bone strains among landing heights of 26 cm, 39 cm, and 52 cm. Several factors may have contributed to their non-significant findings. First, there were only four subjects examined in the study. A small sample size may have limited their ability to discover significant findings. Second, both male and female subjects were recruited in their study. There could be gender differences in landing mechanics, which could influence the tibia strains. Third, the mean age of the subject group was 44 years. Age may play a role of regulating the landing movement to dissipate the impacts. As landing height increases, potential energy increases, so does the amount of energy absorbed by the body upon landing. A loading threshold may be associated with individuals performing landing movement. When this loading threshold is exceeded due to high impact, human bodies must regulate the landing movement to lessen the impact force and avoid lower extremity injury. The aged subjects tested in their study may have experienced such a loading threshold when the landing height exceeds 26 cm.

There are some issues we like to address here. First, seven participants were recruited for this study. This seems to be a small sample size. However, the effect sizes (eta squared) of the strain variables calculated were found to range from 0.745 to 0.889, which were large. Thus, the sample size chosen in this study was justified. In fact, similar studies in the literature used sample sizes from 1 to 4 [19] [23]. Second, all our participants met the US Army enlistment standards. Outcomes of the study are applicable to military recruits entering basic training. Third, some bone strain studies have suggested the existence of loading thresholds, which could result in alterations of bone strain and strain rate patterns [19] [31]. The current study only examined landing heights from 26 cm - 52 cm for a group of healthy young males and did not identify such a loading threshold. It is possible that further increasing the drop-landing height could reveal a loading threshold that may change young adult's tibia bone response. Future studies should examine the effect of higher landing heights (>52 cm) on tibia bone strain to identify the loading threshold for the young adult population. Thus, accurate regression equations could be developed to improve understanding of the mechanism of impact related tibia stress fractures.

5. Conclusion

In summary, increased landing heights resulted in increases in bone strains and strain rates. A dose-response relationship existed between incremented heights and bone strains and strain rates. As bone remodeling is stimulated by increased strain and strain rate and bone is weakened during the remodeling stage, an increased risk of stress fracture presents if impact related training continues. Thus, when developing athletic training programs to improve performance, coaches and athletes should consider the effect of impact loading on bone health. In order to prevent tibia stress fractures, individualized training programs should be developed to give enough time to the trainees for building stronger bones to meet the new training challenges. Similarly, in the military, the administration and recruits need to be aware that high impact loading experienced in basic training is a risk factor of tibia stress fracture. Preconditioning programs for improving tibia bone strength would be helpful for military recruits before their basic training starts.

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Appendix F

Abstract was presented at the 64th annual meeting of American College of Sports Medicine (ACSM) in Denver, CO. May 30th – June 3rd, 2017

Title: The Effect of Soccer Training on Tibia Bone Properties in Healthy Young Females

1439 Board #114 June 1 8:00 AM - 9:30 AM

The Effect of Soccer Training on Tibia Bone Properties in Healthy Young FemalesHenry Wang¹, D. Clark Dickin¹, Daniel Chan¹, Jonathan Foster¹, Julie Hughes². ¹Ball State University, Muncie, IN. ²US Army Research Institute of Environmental Medicine, Natick, MA.

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(No relationships reported)

Tibia stress fracture (TSF) is very common in military recruits. Female Soldiers experience a higher rate than their male counterparts. TSF occurs frequently in basic training due to repetitive impact loading from tasks such as load carriage. Common TSF sites are at distal and middle thirds of the tibia. To date, it is unknown whether training involved multiaxial loading (e.g. soccer) could help improve tibia quality and potentially increase bone's resilience on TSF.

PURPOSE: To examine the effect of soccer history on tibia mechanical properties.

METHODS: 20 female soccer players (20±1 yr) and 20 mass and height matched sedentary women (21±1 yr) completed a pQCT scan on their tibias at seven locations along the bone shaft. Cortical bone density and mechanical strength of the bone at 14%, 38%, 66%, and centers of the proximal, middle, and distal thirds of the bone shaft were examined. One-way ANOVAs were performed.

RESULTS: Significant differences in area moments of inertia and bone strength index were found ($p < 0.05$). The area moments of inertia along the anteroposterior, mediolateral, and longitudinal axes of the soccer players' tibia were 33.5%, 24.7%, 18.4% greater than those of the sedentary women, respectively. In addition, the bone strength index of the soccer players was 19.6% higher than that of the sedentary women. Significant differences in cortical bone density were also found ($p < 0.01$). Sedentary women possessed an average of 2.2% higher cortical bone density in all seven locations of the bone shaft than those of the soccer players.

CONCLUSION: Participants with a soccer history had enhanced tibia mechanical properties, including greater bone strength, but reduced cortical bone density when compared to sedentary controls. These results suggest a co-adaptation of intracortical bone remodeling and bone formation modeling during training such that bone geometry is optimized without excessive increases in bone density, which would be metabolically expensive. Enhanced mechanical properties in soccer players could make the tibia more resilient to unaccustomed mechanical loading, such as load carriage during military training. Future studies should examine whether enhanced bone mechanical properties result in reduced bone strains and a lower risk of TSF. US ARMY #W81XWH-08-1-0587; #W81XWH-15-1-0006.

1440 Board #115 June 1 8:00 AM - 9:30 AM

Serum Sclerostin Levels Are Positively Correlated with Bone Mineral Density in Chinese Young AdultsZhaojing Chen¹, Meihua Su², Youg Zhou³, Breanne Baker¹, Samuel Buchanan¹, David Ross¹, Michael Bemben, FACSM¹, Debra Bemben, FACSM¹. ¹University of Oklahoma, NORMAN, OK. ²Minnan Normal University, Zhangzhou, China. ³University of Oklahoma Health Science Center, Oklahoma City, OK. (Sponsor: Debra Bemben, FACSM)

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(No relationships reported)

Sclerostin, exclusively secreted by osteocytes, is a potent inhibitor of the Wnt signaling pathway and bone formation. While it is well-recognized that differences in bone mineral density (BMD) and fracture rates exist between Asians and Caucasians, little is known about serum sclerostin concentrations and its relationship with BMD in Chinese young adults.

PURPOSE: 1) To compare sex differences in serum sclerostin in Chinese young adults; 2) To investigate associations between serum sclerostin and bone characteristics (areal BMD (aBMD), volumetric BMD (vBMD), bone strength).

METHODS: Fifty-three Chinese men (n=28) and women (n=25) aged 18 to 35 yrs, who had been living in the US ≤ 5 years, participated in this study. Body composition and aBMD of the total body, lumbar spine and hips were measured by DXA. vBMD and bone strength of non-dominant tibia at 4%, 38% and 66% sites were measured by peripheral Quantitative Computed Tomography (pQCT). Serum levels of sclerostin were measured by ELISA.

RESULTS: Serum sclerostin was significantly higher in males compared to females ($p = 0.003$). However, no significant differences were found after adjusting for total body bone mass. There were significant positive associations ($r = 0.39$ to 0.54 , $p < 0.01$) between serum sclerostin and total body bone mass, BMD at total body, lumbar and hips, bone strength at 4% of tibia. After adjusting for total body bone mass, serum sclerostin levels remained significantly correlated with BMD at all sites and bone strength at 4% of tibia.

CONCLUSION: Serum sclerostin levels were positively correlated with BMD and bone strength in Chinese young adults after controlling for total body bone mass, which is consistent with previous population based studies in US Caucasian and Chinese postmenopausal women.

Table 1. Bone Mineral Density and Sclerostin Levels in Chinese Young Adults (Mean ± SE)

	Men (n=28)	Women (n=25)	Total (n=53)
Total BMD (g/cm ²)	1.249 ± 0.017**	1.134 ± 0.023	1.195 ± 0.016
Lumbar Spine BMD (g/cm ²)	1.218 ± 0.084	1.183 ± 0.146	1.202 ± 0.016
Left Proximal Femur BMD (g/cm ²)	1.119 ± 0.133 ~	0.979 ± 0.183	1.053 ± 0.024
Bone Strength Index at 4% Tibia (mm ³)	143.7 ± 4.9**	88.0 ± 7.3	117.4 ± 5.8
Sclerostin (ng/mL)	0.442 ± 0.021~	0.353 ± 0.019	0.400 ± 0.015

~ p < 0.01 and ** p < 0.001 significant differences between men and women, respectively

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Appendix G

Abstract was presented at the 2017 Congress of International Society of Biomechanics in Brisbane, Australia. July 2017

Title: The Effect of Soccer Training History and Load Carriage on Joint Mechanics

THE EFFECT OF SOCCER TRAINING HISTORY AND LOAD CARRIAGE ON JOINT MECHANICS

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INTRODUCTION

Female recruits are afflicted by overuse injuries during basic training [1]. The injury rate of tibia stress fracture, one of the most common overuse injuries, is much higher in females than their male counterparts [2]. Load carriage is a major component in basic training. The repetitive elevated ground impact forces during walking with load carriage may contribute to the increased risk of tibia stress fracture. However, the injury mechanism of tibia stress fracture is still far from being understood. It is unclear how lower-extremity joints and bones respond to load carriage and whether a past history of multiaxial loading through soccer could enhance the function of lower-extremity bones and joints to sustain load carriage. The purpose of the study was to examine the effect of soccer history and incremented load carriage on lower-extremity joint mechanics.

METHODS

20 female recreational soccer players (20±1 yr) and 20 mass and height matched sedentary healthy women (21±1 yr) participated in the study. Walking tasks with load carriage of 0kg, 10kg, 20kg, and 30kg were performed on a force instrumented treadmill (AMTI) at 1.67m/s. A VICON motion capture system was used to record kinematics (200Hz) and AMTI force plates were used to record ground reaction forces (2000Hz).

Lower-extremity joint mechanics were computed for each walking task. Two way repeated measures ANOVAs were conducted to examine the effects of soccer history and incremented load carriage on joint mechanics. Significance level was at 0.05.

RESULTS AND DISCUSSION

It was found that incremented load carriage resulted in increases in most joint moments and power (Table 1). The healthy controls showed greater peak hip and knee power than those of the soccer players.

As the amount of load carriage increases, lower-extremity joint mechanics are altered accordingly, signified by increases in joint moment and power. In this study, compared to recreational soccer players, healthy controls revealed greater hip and knee power production during incremented load carriage. These results indicate that the healthy controls face bigger physical challenges during load carriage than soccer players. The greater increases in knee and hip power production suggest that the healthy controls experienced more energy expenditure, which would lead to higher metabolic cost. Thus, the healthy controls may encounter physical fatigue (which may contribute to altered gait mechanics) earlier than the soccer players during load carriage. Furthermore, attenuated increases in joint moments in soccer players may be protective from stress fracture and other musculoskeletal injuries, although this remains to be experimentally demonstrated. Future studies should examine the associations between joint mechanics, power and injury risk as well as determine if multiaxial loading is protective of tibia stress fracture.

CONCLUSIONS

Load carriage introduces significant physical demands on young females signified by increases in lower-extremity joint moment and power. Recreational soccer players demonstrated less alteration in joint mechanics such as smaller knee and hip power than those of the healthy controls. It appears that a history of multiaxial loading may better prepare military recruits for load carriage, which is an essential part of basic training.

ACKNOWLEDGEMENTS

US ARMY #W81XWH-08-1-0587; #W81XWH-15-1-0006.

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Table 1. Mean(SD) of lower-extremity joint mechanics

	Recreational Soccer Players				Healthy Controls			
	00 kg	10 kg	20 kg	30 kg	00 kg	10 kg	20 kg	30 kg
HM	1.46(0.23)	1.59(0.22) ^a	1.73(0.23) ^a	1.87(0.23) ^a	1.54(0.33)	1.70(0.37) ^a	1.88(0.40) ^a	2.06(0.36) ^a
KM	1.02(0.14)	1.23(0.22) ^a	1.49(0.29) ^a	1.81(0.31) ^{ab}	1.05(0.29)	1.29(0.30) ^a	1.67(0.35) ^a	2.08(0.48) ^a
AM	1.92(0.13)	2.14(0.15) ^a	2.32(0.16) ^a	2.35(0.22)	1.97(0.16)	2.16(0.20) ^a	2.34(0.23) ^a	2.24(0.23) ^a
HP	1.60(0.29) ^b	1.90(0.31) ^{ab}	2.07(0.31) ^{ab}	2.32(0.47) ^a	1.94(0.33)	2.38(0.55) ^a	2.45(0.36)	2.57(0.52)
KP	1.81(0.30)	1.84(0.32) ^b	2.09(0.45) ^{ab}	2.18(0.56) ^b	1.99(0.35)	2.12(0.50)	2.51(0.61) ^a	2.94(1.11) ^a
AP	3.70(0.57)	4.10(0.51) ^a	4.46(0.51) ^a	4.48(0.55)	3.99(0.73)	4.11(0.92)	4.55(0.81) ^a	4.33(0.71) ^a

HM=sagittal plane hip moment(Nm/kg); KM=sagittal plane knee moment(Nm/kg); AM=sagittal plane ankle moment(Nm/kg); HP=hip power(W/kg); KP=knee power(W/kg); AP=ankle power(W/kg); ^a p<0.05, compared with previous level of load carriage; ^b p<0.05 compared with healthy controls.

Appendix H

Abstract was submitted for presentation at the 65th Annual Meeting of American College of Sports Medicine (ACSM) in Minneapolis, Minnesota.

Title: The Effect of Soccer History on Tibial Strain during Load Carriage

The effect of soccer history on tibial strain during load carriage

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Tibia stress fracture (TSF) is common in military recruits. Female Soldiers experience a higher rate than their male counterparts. TSF occurs most frequently in Basic Combat Training (BCT) and has been partially attributed to repetitive impact loading from tasks such as load carriage. Tibia mechanical properties could be improved as a result of training with multi-directional loading (MDL) (e.g. soccer). However, it has yet to be determined whether the improved tibia strength observed with MDL could result in resistance of bone deformation (lower strains) due to load carriage.

Purpose: To examine the effects of a soccer history and incremented load carriage on tibial bone strain.

Methods: 20 female soccer players (20±1 yr) and 20 mass- and height-matched healthy women (21±1 yr) participated in this study. They completed four walking tasks with 0kg, 10kg, 20kg, and 30kg loads on a force instrumented treadmill at 1.67 m/s. Participants' tibia CT models were combined with subject-specific musculoskeletal models for forward-dynamic computer simulations and finite element analyses. Strains from the middle third of the bone shaft were analyzed. One-way ANOVAs were performed. $\alpha = 0.05$.

Results: Significant differences in strains were found among walking conditions and between the two groups (All $p < 0.0001$). The mean $\pm$ SE strains during load carriages (0kg, 10kg, 20kg, and 30kg) were 562±3 μ s, 634±3 μ s, 736±7 μ s, and 825±4 μ s in tension for soccer players, respectively, and 684±3 μ s, 791±3 μ s, 1152±7 μ s, and 1015±4 μ s, for controls, respectively, resulting in an 18-36% difference in strains during load carriage. Compressive strains were 849±4 μ s, 960±4 μ s, 1092±10 μ s, and 1180±7 μ s

in soccer players and $1039 \pm 4 \mu\text{s}$, $1179 \pm 7 \mu\text{s}$, $1749 \pm 11 \mu\text{s}$, and $1552 \pm 7 \mu\text{s}$ in controls showing similar percent differences as tensile strains (18-38%).

Conclusion: Participants with a soccer history benefited from significantly lower strains than healthy controls during incremental load carriage. Lower tibial strains during load carriage may be protective from TSF in those with a history of soccer, although this has yet to be demonstrated experimentally. These findings suggest that physical training involving MDL may be an ideal exercise modality for preconditioning prior to BCT for female recruits. US ARMY #W81XWH-15-1-0006.

Appendix I

Abstract of the Manuscript Submitted to Journal of Visualized Experiments

Title: A Novel Approach to Estimating Bone Strain During Dynamic Motion

ABSTRACT:

Bone stress injuries are common in sports and military trainings. Repetitive large ground impact forces during training could be the cause. It is essential to determine the effect of high ground impact forces on lower-body bone deformation to better understand mechanisms of bone stress injuries. Conventional strain gauge measurement has been used to study *in vivo* tibia deformation. This method is associated with limitations including invasiveness of the procedure, involvement of few human subjects, and limited strain data from small bone surface areas. The current study intends to introduce a novel approach to study tibia bone strain under high impact loading conditions. A subject-specific musculoskeletal model was created to represent a healthy male (19 years, 80 kg, and 1800 mm). A flexible finite element tibia model was created based on CT scan of the subject's right tibia. Laboratory motion capture was performed to obtain kinematics and ground reaction forces of drop-landings from different heights (26, 39, 52 cm). Multibody dynamic computer simulations combined with a modal analysis of the flexible tibia were performed to quantify tibia strain during drop-landings. Calculated tibia strain data were in good agreement with previous *in vivo* studies. It is evident that this non-invasive approach can be applied to study tibia bone strain during high impact activities for a large cohort, which will lead to a better understanding of injury mechanism of tibia stress fractures.