INTRINSIC AND EXTRINSIC RISK FACTORS FOR STRESS FRACTURE AMONG COLLEGIATE CROSS COUNTRY RUNNERS

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ABSTRACT

Taylor Lyn Saimeri: Intrinsic and Extrinsic Risk Factors for Stress Fracture
Among Collegiate Cross Country Runners
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The research to date explored a combination of factors that may put high level athletes at risk for bone stress injuries (BSIs) such as dietary intake, menstrual disturbances, training volume, body composition, and biomechanical variables. The primary purpose of this study was to determine which risk factors play a role in BSIs in collegiate cross-country athletes. Two independent samples t-test assessed baseline bone quality and muscle quality measures between athletes with stress fracture history and those without. A multivariate logistic regression model assessed risk factors for incident BSI over the athlete's competitive season. Athletes with stress fracture history had higher baseline bone mineral content and lower echo intensity values when compared to athletes without stress fracture history. Athletes with a lower bone mineral density (z-score -1.5 or below) were at a higher risk for incident BSI adjusting for stress fracture history, sex and leg lean mass.

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LIST OF ABBREVIATIONS

ACSM	American College of Sports Medicine
A.U.	Arbitrary units
BIA	Bioelectrical Impedance Assessment
BMD	Bone mineral density
BMC	Bone mineral content
BMI	Body Mass Index
BMU	Basal multicellular unit
BSI	Bone stress injury
CM	centimeters
DEXA	Dual-energy X-ray Absorptiometry
EI	Echo Intensity
FM	Fat mass
G/CM^2	Grams per centimeter squared
GRF	Ground reaction force
IN	Inches
KG	kilograms
LBS	Pounds
LM	Lean mass
LLM	Lower leg lean mass
mCSA	Muscle cross sectional area
MRI	Magnetic resonance imaging

MPW	Miles per week
NCAA	National Collegiate Athletic Association
PPE	Pre-participation Examination
RPE	Rate of perceived exertion
TRIMP	Heart rate based training impulse
VL	Vastus Lateralis

CHAPTER 1

When bone undergoes repetitive sub-threshold loading on a daily basis micro-fracture can occur.[1] These micro-fractures over time can develop into a fully defined fracture in the bone, when loading continues to occur and new bone is delayed in being added to the site of tension.[1] Stress fractures are a common sports injury that affect high intensity athletes, such as military recruits and distance runners.[2-5] Stress fractures in the athletic population result in pain, loss of athletic participation and medical expenses.[6]

Stress fractures are injuries that plague both the military, during training, and collegiate level athletes. Stress fractures are estimated as 10% of all athletic injuries while in the military an incidence rate as high as 31 cases in 100 persons has been reported.[1] Injury surveillance conducted by Arendt et al. (2003) found the incidence of stress fractures in collegiate athletes over a ten year period was 1%.[7] Of those stress fractures 3.2% occurred in runners.[7] A study completed specifically with track athletes concluded that over the course of one season 20% of all the injuries were stress fractures.[3] A higher incidence rate of stress fractures have been found in cross-country athletes compared to other contact or non-contact team sports.[7] The study found that 3.2% of the stress fractures occurred in distance athletes, males and females examined as one cohort.[7] However studies have shown that in this specific cohort of distance athletes that women are typically at a higher risk for stress fracture incidents. A study completed by Nattiv et al., (2007) found the relative risk of stress fracture for women is 1.5-3.5 times higher, when compared to their male counterparts.[8] These findings were similar to the study completed Ardent et al. (2003), where females had a reported incidence rate of 1.9% and males of 0.8%.[7] These findings show that stress fractures affect other high performing individuals in the population, not only athletes. [7] The athletes most affected by stress fractures are distance runners, and more specifically female distance runners. All of this suggests that the collegiate distance runner population should be investigated when looking for risk factors that effect stress fracture outcomes.

The research to date has explored a combination of factors that may put athletes at a higher risk for stress fracture incidences such as dietary intake, menstrual disturbances, training volume, body composition, and bone content and geometry, as well as biomechanical variables.[4,6] The findings from these studies are still inconclusive and cannot be generalized to the athletic population as a whole.

The purpose of the study was to determine which risk factors play a key role in bone stress injuries (BSIs) in collegiate cross-country athletes. BSIs considered in the current study were both stress fractures and reactions.[9] The first aim of the study was to determine if a difference in bone composition, defined as bone mineral density (BMD) and bone mineral content (BMC), exists between collegiate cross-country runners with a history of stress fracture and athletes without. The second aim was to determine if a difference in muscle quality, defined as muscle cross sectional area (mCSA) and echo intensity (EI) of the vastus lateralis (VL), exists between athletes with a history of stress fracture and those without. The last aim of this study was to determine if an association exists between bone density measures (defined as lower z-scores) and BSI risk in collegiate cross-country athletes when controlling for confounding variables. Exploratory factors within the last aim were to determine the effects of other body composition variables including fat mass, lean mass and leg lean mass, training volume, injury history and menstrual history on the association between body composition and BSI risk.

There were three main research questions for the study.

1. Is there a difference in whole body bone mineral density and whole body bone mineral content at baseline between collegiate cross-country athletes with a history of stress fracture and those without?
    a. Hypothesis 1a: There is a difference in mean total body baseline BMD measured at the athlete's pre-participation physical, between athletes with a stress fracture history compared to those without.
    b. Hypothesis 1b: There is a difference in mean total body BMC at baseline between cross-country athletes with a history of stress fracture and those without.
2. Is there a difference in muscle quality specifically vastus lateralus (VL) muscle cross sectional area (mCSA) and echo intensity (EI) at baseline between cross-country athletes with a history of stress fracture and those without?
    a. Hypothesis 2a: There is a difference in VL muscle echo intensity, such that athletes with a history of stress fracture will have poorer mean baseline EI than those without a history of stress fracture.

    b. Hypothesis 2b: There is a difference in VL muscle cross sectional area, such that athletes with a history of stress fracture will have smaller cross sectional area than those without a history of stress fracture.

3. Is there an association between low bone mineral density, defined by lower z-scores (-1.5 or below) and BSI outcomes in cross-country athletes?
    a. Hypothesis 3: There is an association between low bone mineral density defined by lower z-scores and BSI risk.

Operational definitions

Bone stress injury: defined as a stress fracture or stress reaction

Competitive year: defined as an athlete's competition year starting August 1st and ending July 31st

Athlete-season: defined as an athlete participating in one competitive-year. Some athletes competed more than one competitive-year or athlete-season

CHAPTER 2

Stress fractures are common sports injuries that result from repeated cumulative stress to the bone at a submaximal level. [6] Athletes that are involved in high-intensity repetitive training, such as military recruits and distance runners, have increased incidence of stress fractures.[2-5] Development of a stress fracture is a function of the number of loading cycles, the amount of applied force, and the time allowed for bone remodeling.[10] The bone is at a much higher risk for injury when there is an imbalance in the bone-remodeling phase such that increased loading occurs during the time between absorption and remodeling. The bone is more susceptible to a stress fracture when the bone can no longer withstand the forces being applied to it in a repeated manner.[10]

Stress fractures in the athletic population result in pain, loss of athletic participation and medical expenses.[6] In military recruits the mean rehabilitation time per year for stress fracture in a study conducted by Wood et al. (2014) was found to be 814 weeks.[11] In distance athletes a study conducted by Kaeding et al (2005) conservatively managed low risk stress fractures can remove an athlete from activity anywhere from 4 to 6 weeks, while high risk stress fractures requiring surgical fixation can take up to 3 months.[12] Knowledge of the risk factors that may lead to stress fractures in the athletic population would improve the clinician's ability to develop injury prevention strategies. The research questions posed in this study seek to determine if factors such as, bone mineral density, bone mineral content, and muscle quality are risk factors for bone stress injuries (BSIs) among cross-country runners. An exploratory aim examined the associations between training volume, body composition, injury history and menstrual history during the competitive season that may increase the risk of BSI. The following review of the literature will explore both intrinsic and extrinsic risk factors that may play a role in BSI occurrence.

Pathophysiology and Epidemiology

Deformation of the lower extremity bone occurs during running when mechanical loads stress the skeleton. The amount of deformation depends on the amount of load applied and the ability of the bone to resist that load.[13] Mechanical loading or stress can be broken down into a smaller unit known as strain, or the change in length of a bone.[13] Bone remodeling takes place due to the activation of cells known as osteoclasts and osteoblasts when mechanical loading to the skeleton occurs. Osteoclasts break down bone while osteoblasts come in to lay down new bone during this process. The damaged tissues are removed and replaced with layers of new bone, maintaining homeostasis in the skeletal system. [13] Bone remodeling is a normal physiological process that helps to reduce tissue age, and allows bone to adapt over time to meet the changing demands being placed on the body.[13] The remodeling of bone is a time dependent process and typically occurs in a 3-4 month period for cortical bone during one remodeling period.[13]

During the cyclical chain of remodeling small bone regions are broken down and absorbed by cells called osteoclasts, while osteoblasts lay down new mineralized bone cells.[14] Osteoblastic and osteoclastic activity do not happen separately but in conjunction with one another, requiring communication between cells. Communication is completed by the basic multicellular unit (BMU).[14] The BMU is made up of a group of osteoblasts, osteoclasts and osteocytes embedded in the bone matrix.[14] The osteocytes are the matured osteoblasts that became trapped in the matrix to form the newly mineralized bone.[14]

Stress fracture development is thought to occur on a continuum, developing due to repetitive mechanical loading where degradation exceeds remodeling. [4] A repeated application of loading below the fracture threshold over short periods of time can contribute to stress fracture occurrence when the rate of stress being applied to the extracellular matrix of bone is not given sufficient time to repair.[15] The bone stress injury (BSI) continuum consists of stress reactions and stress fractures.[13] Stress reactions are defined as increased bone turnover and edema formation around the bone or in the periosteum. A stress fracture has a clear line of demarcation where the fracture is located.[13] Major functions of the bone remodeling cycle are to maintain bone mass and mechanical integrity of the bones, as well as homeostasis of the mineral composition in the skeleton.[14] Scintigrapy and MRI have improved the diagnostics for bony stress injury allowing these injuries to be diagnosed and addressed more quickly.[7] This has allowed for better identification of and diagnostic capability for stress reactions before an occult stress fracture develops.

Adaptive bone remodeling is the response of bone to mechanical load stimuli.[14] Overloading of bone occurs when bone is stressed beyond its upper limit, resulting in accumulated damage to the bone.[14] A BSI occurs when the magnitude, duration, and/or frequency of micro-trauma of the repetitive loads exceeds the pace of the bone's resorption and remodeling phase. [16] Bone's ability to adapt to mechanical load being placed upon it allows it to either gain or lose mechanical properties.[17] Wolff's law states that tissue changes in form or function due to definite changes of the composition in accordance to the forces being applied to them.[17] This explains the response bone has over time to the stress it is placed under.[16] Bone remodels as a response to stress so new bone is laid down where the greatest amount of stress occurs along the bone.[1] Continuous repetitive stress on a daily basis at sub-threshold loading will cause micro-trauma.[16] Bone that is stressed in this manner during any lag in the remodeling phase of bone growth results in a bone vulnerable to a stress injury.

The time lag between osteoclastic reabsorption and osteoblastic formation makes determining the exact time frame of when a BSI will occur after the onset of a training bout unpredictable.[1] Philipson et al. (2009) states that, "Bone is a dynamic tissue constantly remodeling under the influence of multiple hormonal and mechanical factors".[1] Many factors can play a role in BSI development across the training continuum. Symptoms typically appear within six to eight weeks after a change in training has occurred.[2] Due to this injury etiology, early identification of risk factors and diagnosis of a bony stress injury improves treatment effectiveness and patient outcomes.[2] Bone stress injuries will typically present with an insidious onset of pain. Common symptoms include pain associated with exercise that is relieved by rest, localized bony tenderness, and swelling overlying soft tissue and pain during loading. [4,10] Pain progresses from intermittent to continuous whether or not there has been activity completed that day, pain worsens at night.[2]

A prospective study by Arendt et al (2003) completed over a ten-year period found a one-percent incidence rate of stress fracture in athletes competing in organized sports.[7] Females were found to have a higher incidence rate than their male counterparts, at 1.9% compared to 0.8% in males. Cross-country athletes when compared to other contact or team sports have a higher rate of stress fracture incidence at 3.2%.[7]

Approximately one- to two-thirds of cross country athletes and long distance runners have some history of BSI. In about 10.3% to 12.6% of cases, athletes with a history of a BSI sustain subsequent BSIs when prospectively followed for 1-2 years.[13] A study that tracked injury incidence in track athletes demonstrated that 75 athletes sustained one or more stress fractures, 69% of which occurred in females and 81% of which occurred in males.[3] The study looked at overall injuries occurring over the course of the track season. Twenty percent of those injuries were stress fractures. [3] Another injury survey conducted by the NCAA estimates the incidence of stress fractures in athletes range from 1-2.6% with 15% of those occurring in runners.[16] The same study found the relative risk for women is 1.5-3.5 times higher when compared to their male counterparts.[16]

Distance athletes have also been studied and compared to their track athlete counterparts competing in shorter distance competitions. A study conducted by Warden et al. looked at the differences between distance athletes and sprinters.[13] Distance runners are typically rear foot strikers where the sprinters follow a forefootstriking pattern.[13] These differences in gait mechanics may correspond with the different areas of BSI found by the researchers in their study. Distance runners have been found to more commonly develop injury to long bone structures, such as the tibia and femur where sprinters are more prone to BSIs in the tarsal and metatarsal bones of the foot.[13]

Bone stress injuries can occur in any region in the lower extremity in runners, and should be considered in the differential diagnosis in these athletes if they complain of acute or chronic pain.[13] Bone stress injuries typically occur slowly over time with insidious onset of symptoms and may eventually prevent athletic participation. As with most overuse injuries, there are multiple risk factors that play a role in the development of the injury. Identifying which factors influence stress fracture incidence may help clinicians prevent stress fracture development in athletes.

Risk Factors

Numerous risk factors exist for BSI in the athletic population. Risk factors for any injury are typically classified as intrinsic or extrinsic. Extrinsic factors are those that are external to the athlete. Intrinsic factors are characteristics or attributes of the athlete.[10] Research to date has explored soft tissue composition, bone mineral density and geometry, dietary intake, menstrual disturbances, training volume and biomechanical factors and their effects on bone health in distance runners.[4,6] In this particular review of the literature both extrinsic and intrinsic factors will be considered. Intrinsic risk factors include; body composition, muscle composition and bone composition, and menstrual cycle changes in women, while extrinsic risk factors include disordered eating and training load.

Body Composition

Body composition can be measured a variety of ways. Brodie et al (1998) states that when choosing the correct method to measure body composition, one must consider the cost, availability, access, validity and intervention when researching body composition.[18] There are multiple types of body composition measurement systems: hydrodesitometry, anthropometry, Bioeletrical impedance assessment (BIA), and Dual-energy X-ray absorptiometry (DEXA).[18]

Hydrodensitometry or hydrostatic weighing is a method utilized to measure body volume. Due to the necessity of prolonged time submerged under water a high degree of water confidence is required which is a limitation of the technique.[18] Another limitation is the affect of food and hydration status on measurements affecting the percent body fat estimations. A similar limitation is its use of a two-compartment method for assessments. The calculations work under the assumption that the body is made up of only fat mass and fat free mass. It does not take into account muscle mass and bone mass estimates, which also increases prediction errors for fat and fat free mass estimates.[18,19]

The most portable method of assessing body composition is the use of skin calipers. All the researcher needs is the calipers themselves and the knowledge of the algorithms for each measurement site to calculate body fat percentage. This type of method is one measure of anthropometry; others include bone dimension and girth measurements. [18] This method is most effective in field testing and is a very non-invasive and mostly pain free way to estimate a subjects body fat percentage.[18] The limitations to this method include the assumption that adipose tissue layers represent total body fat percentage, as well as the variability between testing procedure and inter-rater reliability measures.[18] BIA is another semi-portable means of assessing body fat percentage and BMI of a subject. This technique is based off the relationship between the volume and length of a conductor as well as its components and impedance to current flow.[18] The limitation to this technique is that it assumes the conductor is a perfect cylinder, which is not the case for the human body. Variations in body proportions may in turn enhance error associated with body fat percentage and BMI calculations.[18]

The final technique assessed in the previous study is the DEXA scan. This technique is an improvement upon the two comportment method as it is a three compartment assessment of lean mass, fat mass and bone mineral content.[18,20] The DEXA scan uses tissue density to assess both whole body and regional fat mass, lean mass and bone mineral content. This technique allows for segmental measurements to be quantified.[21,22] The DEXA was a product of advancement in absorptiometry which started with single photon, moved to dual-photon which in turn became the DEXA.[19] The basic principle of the DEXA is the measurement of energy transmission in the body of x-ray at high and low levels.[20] The two energy levels of transmission allows the derivation between fat mass and lean mass without bone mass being accounted for.[22] The DEXA itself gives an output of the subjects fat mass, lean mass, bone mineral content, body fat percentages and bone mineral density for both the whole body and body regions for both the axial and appendicular skeleton.[22]

DEXA scans take only 10-15 minutes.[21] Each subject is exposed to very limited radiation. A strength of this method is its ability to give information on specific body segments, for both the axial and appendicular skeleton.[19,20,23] The technique can also be used with individuals who cannot undergo the procedures for hydrostatic weighing. All the individual must do is lay still for the entirety of the scan itself. The DEXA has no specific contraindications other than pregnancy making it widely available to a large population of individuals.[20,23] However, there are several limitations to DEXA scans. In some cases, the size of the table may be too small to accommodate a taller or wider individual and cause a deviation in measurements. Results may differ between machines due to differences in the type of X-ray utilized by the unit. [22] Precision of the scan has also been found to be lower in the obese population.[20] Other limitations of the DEXA scan can be caused by two types of errors these are either technical or biological. Technical errors may be generated by the machine, incorrect placement of the subject or inaccurate image post processing. Biological variations can be due to hydration status, exercise, food intake as well as long term changes due to diet an exercise.[20] These types of errors can be mostly avoided by following a standard protocol when scanning each subject.

Accurate analysis of body composition could help advance prevention, treatment and comprehension of injury.[19] Studies have shown that body composition will change through the course of the season.[24] One study in female track athletes showed that when compared to other female athletes actually start their pre-season at lower body fact percentages.[24] Determining if these fluctuations or values play a role in injury incidence may help determine prevention protocols in the future.

Bone mineral density, an output assessed by the DEXA is an important measure in the diagnosis of osteopenia, osteoporosis and other metabolic bone diseases as well as for prediction of fractures.[25] For the aforementioned reasons, DEXA is the most commonly used technique to assess bone mineral density and bone mineral content. The limitation however is that it cannot provide separate bone masses for the cortical versus the trabecular bone. For that typically Quantitative CT is utilized.[25] The DEXA as a test for fracture risk is the most valid in all areas of its scan except for the prediction of vertebral fracture.[25]

Bone mineral density, content and geometry

Current research has focused on bone mineral density (BMD) as a risk factor for BSI. DEXA is the current criterion for diagnosing osteopenia in patients by measuring BMD.[26] Advances in DEXA scan capabilities allow for both total body and regional bone density values to be assessed. This type of measurement may be instrumental in beginning to understand regional bone responses to sport specific loading demands.[26] Athletic bone mineral density is defined differently than that of the regular population. BMD is typically calculated by assessing z-scores. A z-score is a statistical measurement of a score's displacement about the average score, which is set to a standard of zero. The Z-score is related to the number of standard deviation above or below the population mean. Z-scores are used to compare bone density with age and sex matched controls.[16] In the general population, a z-score of less than or equal to -2 is considered risky bone mineral density. One study noted that athlete groups who sustained a stress fracture had a higher incidence of being osteopenic, having a t-score (different demographic matched measure from z-score) of less than -1.0.[6] Low bone density is theorized to increase the risk of stress fracture by reducing bone strength, making it easier during repetitive loading for micro trauma to occur. The bone is unable to withstand the forces acting upon it for extended periods of time.[10]

Current research demonstrates conflicting findings regarding bone density as a risk factor for stress fracture. One study conducted by Bennell et al. (1999) found bone density predicts stress fracture risk in females but is not as predictive in males[10,27] Another study by Bennell et al. (1996) found women who developed stress fractures had lower values for total bone mineral content (BMC), lumbar spine BMC and calcaneal BMD. The same pattern was not seen in males.[4] However, males with a history of stress fractures did follow a trend toward lower BMD scores.[3] The findings suggest sex differences should be considered when identifying stress fracture risk factors.[3]

Studies also have found that the BMD in specific anatomic locations may be important to consider in athletes with a history of stress fracture. Bone mineral density of the femoral neck, total hip (which includes the femoral neck), trochanteric region, inter-trochanteric region, and lumbar spine were significantly lower in a group with a history of stress fractures when compared to their uninjured counterparts.[6] The same study found that BMD at the site of a prior stress fracture was not significantly different between groups.[6] The findings in this study suggest that patients with a history of stress fracture are more likely to have an overall BMD deficit compared to healthy controls. The BMD at the sites of injury however are not significantly different between groups. The lack of difference may exist due to the bone remodeling and healing over time.

Examining the BMD at the injured anatomic site may not be enough to determine an athlete's risk of developing a recurrent stress fracture.[6] Roelofs et al. (2015) found significant correlations between BMC and lean mass in the lower legs of male and female cross-country athletes.[28] Additionally, there was a strong correlation between muscle cross sectional area (mCSA) and BMC, as well as BMD.[28] Theses studies show that BMD and BMC may not be enough information about the bones to determine if someone is at-risk for a BSI. Other factors may play a role in how the bone reacts to forces being applied to it on a regular basis.

Bone strength and shape are other factors that have been considered as risk factors for BSI incidence. A study conducted on female distance runners showed significantly larger bone strength estimates at mid-shaft cortical bone sites of the tibia. When adjusted for muscle cross sectional area the differences were no longer significant for females with a history of stress fracture and those without.[29] Beck et al. (2000) found that in both sexes, those with significantly larger bone geometries did not suffer from a history of stress fractures.[5] The previous findings suggest bone strength, defined based on bone content and also bone size, can be indicative of those individuals at-risk for a BSI. The studies also show however that the muscle surrounding the bone may also play a role in BSI risk and protection.

Other studies have also considered the correlation between BMD and measurements of body composition such as BMI, body fat percentage and total mass measurements.[9,26,30] Tenford et al. (2015) found that adolescent runners had both BMI and z-score values that were below average when compared to normative values for their age groups based on CDC reference values.[9] Egan et al. (2006) also found that distance runners had significantly lower fat mass and percent body fat than any other athlete group.[26] To support the hypothesis that these values are indicative of potential injury, the same study also found that total lean mass and total mass values were positively correlated with BMD at all sites in a DEXA scan.[26] The findings suggest that distance runners, due to their body types, are already at-risk to having lower BMD values that in turn puts them at a higher risk for BSI incidence. A similar study found that mass as well as BMI were significantly correlated with BMD measurements at each site.[30] The same study found that percent body fat was not correlated with BMD values at any site in the scans however and that total body mass was the best predictor of BMD specifically in female athletes.[30] Runners as a whole, both distance and sprinters were found to have lower BMI and mass measurements in conjunction with lower BMD values at multiple sites. Runners also had the lowest total and lumbar spine BMD out of any athletes.[30] The studies discussed above show that it may not only be the lean mass values in body composition that play a role in BMD values but total mass may also need to be taken into account when implementing prevention strategies.

Muscle size, cross sectional area and strength

Previous research has demonstrated that skeletal loading forces, such as bending, torsion and so on, can be mediated through the contraction of surrounding musculature. Certain muscle groups have also work to oppose bending and torsional forces acting on the bone.[5] The findings of a study conducted by Roelofs et al. (2015) suggest muscle size may have a protective role in bone health.[28] Other researchers have theorized muscle plays a protective role on bone during running by reducing shear forces acting on the bone being loaded during the running cycle.[6] Less lean mass and a smaller muscle girth may be indicative of lower muscular strength, increasing an athlete's risk of stress fracture.[6] Another study completed in military recruits found that for both males and females with stress fractures incidences had smaller muscle cross sectional area of the thigh musculature when compared to healthy control.[5] Shnackenburg et al. (2011) found similar results in female distance athletes. Knee extension torque was significantly lower in athletes who sustained a stress fracture versus those who did not.[6] Strength of the musculature in contact with the bone (either insertion or origin) should also be considered when identifying at-risk individuals for stress fractures[6]

Previous research demonstrates conflicting findings on the importance of muscle cross sectional area (mCSA) of the upper and lower leg as a factor that correlates to stress fracture incidence. A study conducted by Popp et al. (2009) found that mCSA was significantly lower in runners with a history of stress fracture.[29] When the data was corrected for the mCSA, bone strength estimates were no longer significantly different between athletes with a history of stress fractures or without. The data suggests that muscle strength may have a higher role in stress fracture risk than BMC and BMD. Another study by Roelofs et al. (2015) looking at both males and females with a history of stress fractures and comparing the subjects to those with no history of stress fracture found no significant difference between lean mass values.[28] In the same study, a high association between lean mass in the affected leg and boney integrity measured as BMD was found. The authors noted a potential difference in mCSA in the vastus lateralis between groups; those having a history of stress fractures typically did have smaller mCSA.[28]

Two studies conducted on military personnel and gymnasts had similar findings.[5,31] The study found that overall, military personnel with stress fracture had weaker lower limbs, which was derived from their mCSA data, and smaller mCSA of the thigh.[5] The gymnasts also had smaller mCSA of the upper and lower portions of the leg as well as weaker lower extremities when compared to healthy subjects.[31] If military personnel and gymnasts with weaker lower extremities, smaller mCSA and higher reported stress fracture incidences, it could potentially be inferred that mCSA and muscle strength plays a protective role against stress fractures.[5,31] The existing data suggests that mCSA and lean muscle mass may be important for prevention of stress fracture, suggesting that stress fracture risk is multifactorial and requires further investigation.

Contradictions in the literature also exist in the thoughts on the differences of affects of risk factors on BSI between the sexes. One theory suggests that one risk factor may be of more significance to one sex versus the other.[28] A recent study examined the relationships between mCSA, weight, lean mass in the lower extremity and performance and stratified their findings based on sex.[28] In males and females with a history of stress fracture, a significant correlation was found between mCSA and lower leg lean mass. Athletes with a lower lean mass and smaller mCSA were at higher risk for stress fracture injury.[28] Males and females differed, however, when total body composition was taken into account. Males with a history of stress fracture had lower body weight, fat mass, echo intensity and mCSA of the vastus lateralis when compared to uninjured male.[28] In contrast, females had higher fat mass and percent body fat, while mCSA was lower than their healthy counterparts.[28] Males and females examined as one cohort showed no significant lean mass value differences in healthy versus previously injured athletes.[28] Therefore, mCSA may play a greater role in BSI risk for one sex than the other. Currently the research is unclear as to which sex may have a higher risk of developing BSI when differences in muscle composition exist. Females may be at higher risk when mCSA in the vastus lateralis is lower where males' risk of BSI may be less affected by muscle composition. There are still disparities between the sexes and risk factors affected on BSI outcomes. This may suggest separate prevention models and future research may be needed to look at each separately.

Disordered eating and the female athlete triad

An extrinsic risk factor that can affect an athlete's bone health is disordered eating. Disordered eating is classified as a persistent disturbance of eating or eating related behaviors, which influences the consumption and/or absorption of food and significantly impacts physical health or psychosocial function.[32] Barrack et al. (2008) noted that primary components of disordered eating include weight concerns, shape concerns, eating concerns and dietary restraint.[33] In the same study, female cross country runners completed questionnaires regarding menstrual history and eating behaviors.[33] The authors found that runners with elevated weight or shape concerns actually had significantly higher BMI, body weight and percent body fat. These runners also reported running fewer miles per week during the season.[33] Runners who reported some form of dietary restriction had significantly lower bone mineral density than those who did not report any disordered eating changes.[33] The study did not report the incidence of stress fractures of their participants. However, the findings of decreased bone mineral density found by Barrack et al. are important as decreases in BMD has been previously linked to stress fracture risk.[3,5,10,27]

In 2007, the American College of Sports Medicine (ACSM) redefined the triad as a multifactorial disorder including three primary arms: energy availability, menstrual health and bone health.[16,34] Specifically, the Triad is a syndrome of three conditions, which exist along a continuum. These conditions are: 1) energy deficiency, with or without disordered eating, 2) menstrual disturbances or amenorrhea, and either 3) bone loss or osteoporosis.[35]

Energy deficiency is classified as any imbalance between energy consumption and energy expenditure. Inadequate caloric intake during intensive training leaves an athlete with depleted dietary energy and has be associated with reduced bone mass.[1] In one study of female adolescent endurance runners, dietary restriction was associated with lower bone mineral density scores.[33] This suggests poor and pathological eating habits, which may in turn increase the risk of future stress fracture. [33] Additionally, studies have examined the relationships between self-efficacy and disordered eating. Female marathon runners with a history of bone stress injuries had higher self-loathing screening scores. The self-loathing subscale highly correlates to self-reported eating disorder incidence, which may have a subsequent effect on bone health.[36]

Studies have found that disordered eating, low calcium and fat intake, and low dietary dairy product intake may be related to stress fracture risks in female athletes.[37] Female sprinters with a history of stress fractures had significantly lower fat intake and a higher calcium intake relative to their energy output.[4] The difference in nutrient levels is suggestive of a restricted diet. Four eating patterns were researched in the study completed by Nieves et al. (2010).[37] The only statistically significant pattern to reduce stress fracture incidence by increasing BMD was the diet that included an increased consumption of dairy product with decreased fat intake. [37] The study also found that calcium intake was positively related to annual gains in BMD of the hip and total body BMD. The previous study exemplifies the effect nutrition has on bone health. Better understanding of the influence of nutritional behaviors on bone health may allow clinicians help mitigate stress fracture incidence through early identification of modifiable risk factors.

Disordered or abnormal menstrual cycle is the second component of the triad. Menstrual disturbances can range from an irregular cycle to the cessation of the menstrual cycle, amenorrhea. Women with the risk factors of bone loss and osteoporosis are at much higher risk for lower bone mass and less bone strength. Bone loss and osteoporosis can lead to increased risk of BSI.[35] In female athletes, stress fracture incidence and dietary restriction are typically researched as outcomes due to deficiencies rendered by side effects of the female athlete triad. Female distance runners are identified as a group that have a higher risk of developing one or more triad related diagnoses.[16]

Normal menses at a cycle of approximately 28 days is defined as eummenorhea.[34] Deviations from this are know as amenorrhea or oligomenorrhea. Amenorrhea can be categorized as primary or secondary.[34] Primary amenorrhea is the absence of menarche after the age of fifteen, where secondary is the absence of a cycle for three or more consecutive months.[34] Oligomenorrhea is when menstruation occurs on a 35 day cycle or when a female has fewer than nine cycles in a year.[34] Female athletes with a history of stress fracture typically have a history of delayed menarche and report fewer menstrual cycles per year leading up to injury diagnosis.[4]

It is important that an athlete's past history and current cycle pattern are well-documented, as estrogen plays a large role in the physiology of bone formation.[34] Estrogen inhibits bone remodeling and resorption which then conversely helps to increase bone formation.[34] If an athlete is in an estrogen-deficient state, bone mineral density decreases, thus increasing the risk for bony stress injury.[34] Scofield et al. (2012) found that BMD was ten percent lower in amenorrheic athletes compared with eumenorrheic athletes.[16] Those ammenorrheic athletes lost two to three percent of their bone mass per year if the condition went untreated.[16] Female athletes with ogliomenorrhea or amenorrhea have a two to four-time higher risk of stress fracture than healthy menstruating counterparts.[16]

Training load, volume, intensity, baseline fitness

Training load is defined as the amount of training completed by an athlete during one training session.[38] There are two common methods used to quantify training load: internal load is the athlete's physiological and/or psychological response to an externally applied load. External load is training completed by the athlete without consideration of the physiological response.[38,39] There are three main methods of measuring internal load that are well reported in the literature: heart rate based training impulse (TRIMP), rate of perceived exertion (RPE) and session RPE. These internal load measures have all been found to be highly correlated with one another for quantifying the physiological response to training.[38]

The RPE method is based on the hypothesis that athletes can inherently monitor the stress their bodies experience during training.[40] The method has been utilized during steady state exercise and high intensity interval training and has been proven to be effective at quantifying internal training load responses during these activities.[40] Session RPE and HR-based methods have demonstrated a strong relationships between one another for being able to assess internal training load in Australian football athletes, endurance runners, cyclists and collegiate basketball athletes. [39,41,42] Session RPE is a modification of the RPE method that uses RPE and a marker of intensity. Session RPE is calculated by multiplying the athletes self-reported RPE by the duration of training session.[43] Monitoring training load is important to identify where injury may occur by detecting overreaching in training bouts.

Session RPE training loads have more recently been linked with injury and time loss in the athletic population.[43-46] A study completed in elite rugby athletes found that on field training loads were significantly associated with on field injures.[45] The findings suggest that the harder the athletes train the more likely they are to sustain and injury.[45] Another study completed by Gallo et al (2015) found that session RPE was positively correlated with distance, average speed and high speed running distance.[43] As training distance, average speed and high speed running distance increase so do session RPE reported values.[43] The study demonstrates that prescribing training based on external training loads will lead to differences in internal responses that could lead to overtraining in some athletes and thus injury.[43] A similar study was completed in a sample of distance runners. Garcin et al. (2002) measured session RPE over an 8-week training session and demonstrated that session RPE was able to detect periods of overreaching in training.[47,48] These studies suggest that training load and recovery status should be monitored to help reduce training related injuries in athletics.[45]

An important extrinsic factor is external training load. Training load is defined as a rapid increase in training activity or intensity and can increase an athlete's risk for injury.[16] For distance runners this could manifest as an increase in mileage, pace, change in training frequency or a change in running surface.[16] Abrupt changes, such as increases in training, during the time between bone reabsorption and reformation has been demonstrated to increase the risk of stress fracture in distance runners.[1] One study conducted in a cohort of college athletes found that approximately 48% of stress injuries to bone were correlated with a change in the training regimen, defined as entry to a new program or training with a new coach.[7] When a change in training load occurs, the effects it has on bone health may not be seen immediately. The time between bone absorption and reformation is not instantaneous, which can make BSI diagnosis even more difficult. It is important for clinicians to monitor athletes' training habits closely, and to be aware of changes in training habits and any associated symptomology.

In conjunction with training loads, baseline physical fitness prior to starting intense exercise has been found to play a role in stress fracture incidence. A study conducted on distance runners evaluated the effects of physical fitness on stress fracture incidence. The runners' fitness levels were tested with a Cooper's Run (a timed mile and a half), and a timed sit up test.[49] The study found in both genders those athletes who on average were less physically fit had a history of more stress fractures.[49] A study conducted with male military recruits had similar findings. The test group with worse Cooper's run times had a higher number of individuals in the cohort with history of stress fractures.[50] The same study also examined aforementioned risk factors, including muscle quality and bone mineral density in addition to physical fitness.

These findings demonstrated that changes in both internal and external training load over the course of a training regimen could lead to injury events. These factors should be taken into account when creating training plans an athlete's competitive season. When considering athletes who may be at-risk for BSI it is not only important to consider the amount of training they are doing but their physical fitness prior to increasing training intensity. In the case of cross-country athletes, who run on a continuous basis, it may not be a combination of their baseline fitness and their fitness levels at their current training mileage and intensity. Changes in training such as increased mileage, intensity and pace adjustments should be made gradually and mindfully in order to help reduce the risk of incurring injury potentials.

Biomechanics and biomechanical risk factors

Biomechanics are also an important factor to consider when studying BSI risk and prevention. Aberrant biomechanics can alter loading forces on the lower extremity thus altering internal training load spoken about above. Studies have been conducted to investigate the effect of changes in gait biomechanics during running in a fatigued state, at different grades of incline, and for specific fracture sites.[51-53] These studies found changes in peak hip adduction angle, abduction force moment at the hip and rear foot inversion angle are all correlated with stress fracture history.[51-53]

To determine the effects of changes in biomechanics on BSI risk, it would be clinically significant to know when these breakdowns occur, if not originally present. A study by Clansey et al. (2012) examined the biomechanical changes in the lower extremity when running in a fatigued state. After 40 minutes of intense running instantaneous and average vertical loading rates significantly increased.[52] Overall the majority of alterations in running mechanics were found to occur at the 40 minute mark of high intensity training, suggesting an increase in repeated runs at this level of high intensity increases the risk for injury.[52] These findings suggest that it may be more prudent to complete two shorter runs during the day than one complete bout of training, particularly if a runner is unable to maintain proper biomechanics during a given bout of running.

Changes in ground reaction forces during running have also been shown to be associated with BSI incidence. A study conducted on female subjects found an increase in ground reaction forces during 30 meters of running in patients with stress fracture history.[54] The study did not find any combination of variables that were statistically relevant in predicting possible BSI incidence. The authors hypothesize there may be other factors that play a more important role in stress fracture development in females.[54] Conversely a study explored differences in peak medial and lateral GRF, peak braking GRF, vertical impact GRF, average and peak vertical loading rate and between the injured and uninjured limbs.[55] The study did find significant differences, suggesting that ground reaction forces play a role in stress fracture incidence.[55] The study found peak braking, peak shock, and vertical ground reaction forces are significantly higher in the affected limb when compared to the unaffected side.[55] A study conducted in 2009 found that a 10% reduction in stride length could decrease peak tibial contact forces.[49] The reduction in forces being absorbed and transmitted through the tibia should also lead to a reduction in bone breakdown and in turn stress fracture occurrences.[49] With longer stride lengths or an abrupt increase in typical stride length, the athlete may be absorbing more force through the tibia and increasing stress placed on the bone. The study also found that the number of stress fracture occurrences rise with an increase in running mileage, even if the stride length is shortened.[49]

Many of these studies show that it is not adequate to adapt one piece of training but the training bout as a whole needs to be considered in order to prevent injury, instead summarizing that a multi-factorial approach to injury prevention is ideal.[49] These studies depict it is not enough to know how much the athlete is training but what forces their bodies are asked to absorb when they are training.

Gaps and Limitations in Current Research

The majority of studies to date have evaluated risk factors for BSIs using a retrospective approach. In order to better understand injury risk and develop clinical prevention strategies, prospective investigation of key risk factors that influence the development of BSI is critical.[10] In order to improve clinical screening best practice, clinicians must have a comprehensive knowledge of which factors are most strongly associated with the risk of injury. The current study seeks to determine the combination of risk factors that place athletes at highest risk for BSI. Such an investigation would help to provide information for future research and future development of screening tools during sport pre-participation examinations, to identify at-risk individuals.

Additionally, current research typically generalizes bone composition data to the whole lower extremity based on only two areal sections, such as the femur and the tibia alone.[5] For example, in the study conducted by Bennell et al., (1995) the researchers had technical issues with the DEXA scan that they used and only measured bone composition values at the sites of past injury. In this case their findings can't be generalized to other bones in the lower extremity because they only examined the femur and tibia, where this cohort's past injuries had occurred.[56]

Lastly, few studies have examined the influence of multiple factors on stress fracture incidence. Bone stress injury has a multifactorial etiology; hence, a single factor is not able to definitively determine if an athlete will sustain a stress fracture.[10] The majority of previous studies have looked at multiple risk factors, but analyzed them separately, looking at their individual effects on BSI incidence. The few studies that have combined the variables for analysis have had some success with determining a group of risk factors. Trends in different risk factor groupings have been noted in these studies.[4,29,56] However due to small numbers of outcomes have had to collapse event groups together as in the study by Bennell et al. (1996) that was completed over the course of one season. This study looked at multiple training years and seasons specifically in distance athletes who are typically found to incur the most BSI incidences in the track and field cohort.[3]

Therefore, identifying factors with high relative risk ratios, thus linking them to BSI outcomes in a multivariate model controlling for confounding variables, is essential to improve injury prevention strategies for bony stress injuries. If relative risk ratios can be estimated for each risk factor and subsequently analyzed as a part of a multi-factorial model, determining the combination of risk factors that most strongly associates with BSI risk is more likely. The findings from the current study may improve clinicians' knowledge of the most important risk factors for which to screen, and what cluster of risk factors place an athlete at a higher risk of BSI development.

CHAPTER 3

The purpose of the study was to determine which risk factors play a key role in bone stress injuries (BSIs) in collegiate cross-country athletes. BSIs considered in the current study were both stress fractures and reactions. The first aim of the study was to determine if a difference in bone composition, defined as bone mineral density (BMD) and bone mineral content (BMC), exists between collegiate cross-country runners with a history of stress fracture and athletes without. The second aim was to determine if a difference in muscle quality, defined as muscle cross sectional area (mCSA) and echo intensity (EI) of the vastus lateralis (VL), exists between athletes with a history of stress fracture and those without. The last aim of this study was to determine if an association exists between lower bone density scores defined as z-scores and BSI risk in collegiate cross-country athletes. Exploratory factors within the last aim will be to determine the effects of body composition (fat mass, lean mass and leg lean mass), training volume, injury history and menstrual history on the association between body composition and BSI risk.

There were three main research questions for the study.

1. Is there a difference in total body bone mineral density and total body bone mineral content at baseline between collegiate cross-country athletes with a history of stress fracture and those without?

a. Hypothesis 1a: There is a difference in mean baseline BMD measured at the athlete’s pre-participation physical, between athletes with a stress fracture history compared to those without.

b. Hypothesis 1b: There is a difference in mean BMC at baseline between cross-country athletes with a history of stress fracture and those without.

2. Is there a difference in muscle quality specifically muscle cross sectional area (mCSA) and echo intensity (EI) of the vastus lateralis (VL) at baseline between cross-country athletes with a history of stress fracture and those without?
   a. Hypothesis 2a: There is a difference in VL echo intensity, such that athletes with a history of stress fracture will have poorer mean baseline EI than those without a history of stress fracture.
   b. Hypothesis 2b: There is a difference in VL muscle cross sectional area, such that athletes with a history of stress fracture will have smaller cross sectional area than those without a history of stress fracture.
3. Is there an association between lower bone mineral density, defined by lower z-scores (-1.5 or below) and BSI incidence in cross-country athletes?
   a. Hypothesis 3: There is an association between low bone mineral density defined by lower z-scores and BSI risk controlling for confounding variables.

Study Design and Participants

All study procedures were reviewed and approved by the Institutional Review Board at the University of North Carolina – Chapel Hill. A cohort of 64 division-one collegiate distance runners (27 females and 37 males) was followed over the course of their competitive seasons, beginning the second week of August when the athletes report for pre-season until the follow august preseason date, at the University of North Carolina from 2013 to 2016 (a total of 4 competitive seasons). Each year, the specific variables listed in Table 1 below were collected during the pre season laboratory screenings and pre-participation examinations in the fall. In addition, during the 2016-2017 competitive seasons, athletes were asked to record their weekly mileage per coaching staff. Any athletes who chose not to take part in the two parent studies discussed below were excluded from this study. A participant flow diagram is provided for each of the study aims (Figures 1 and 2).

Figure 1: Diagram of participant flow for the study in aims 1 and 2

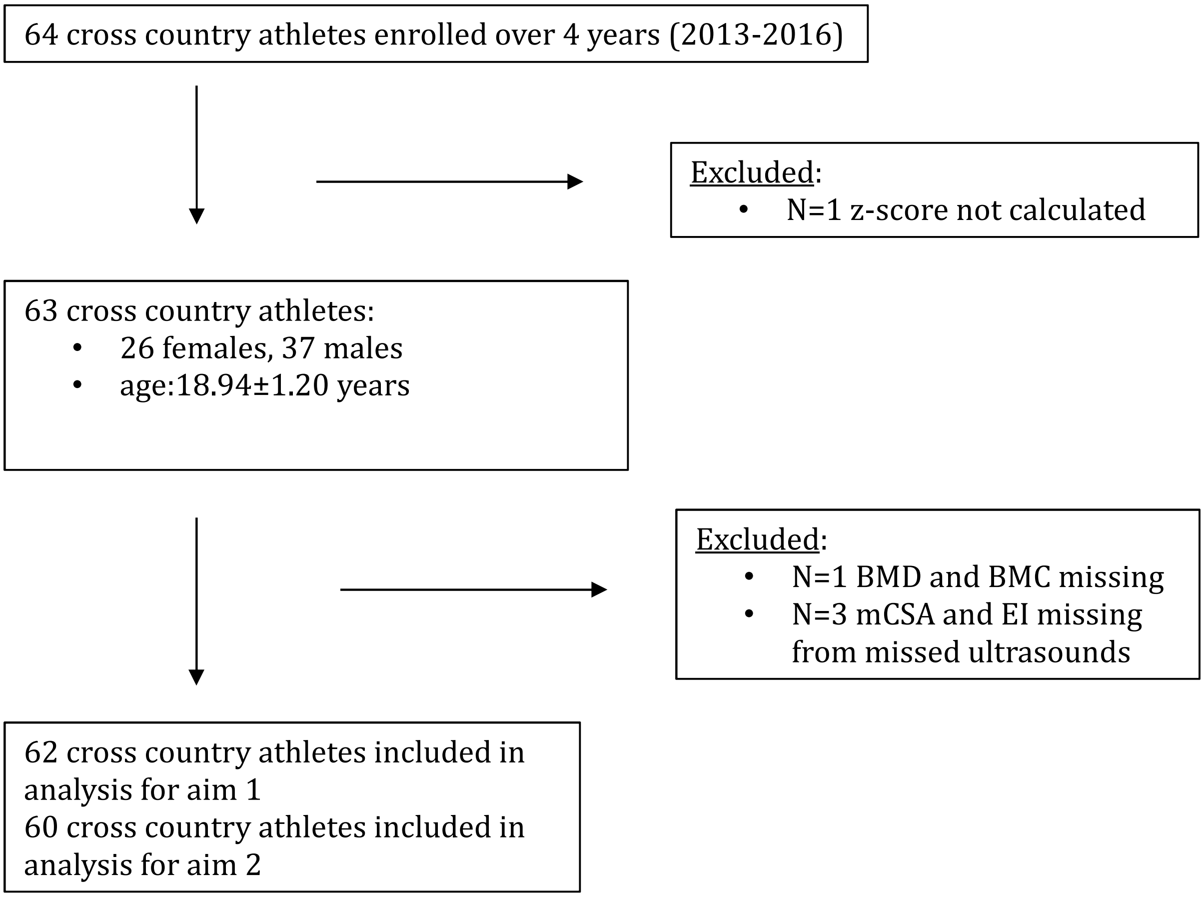
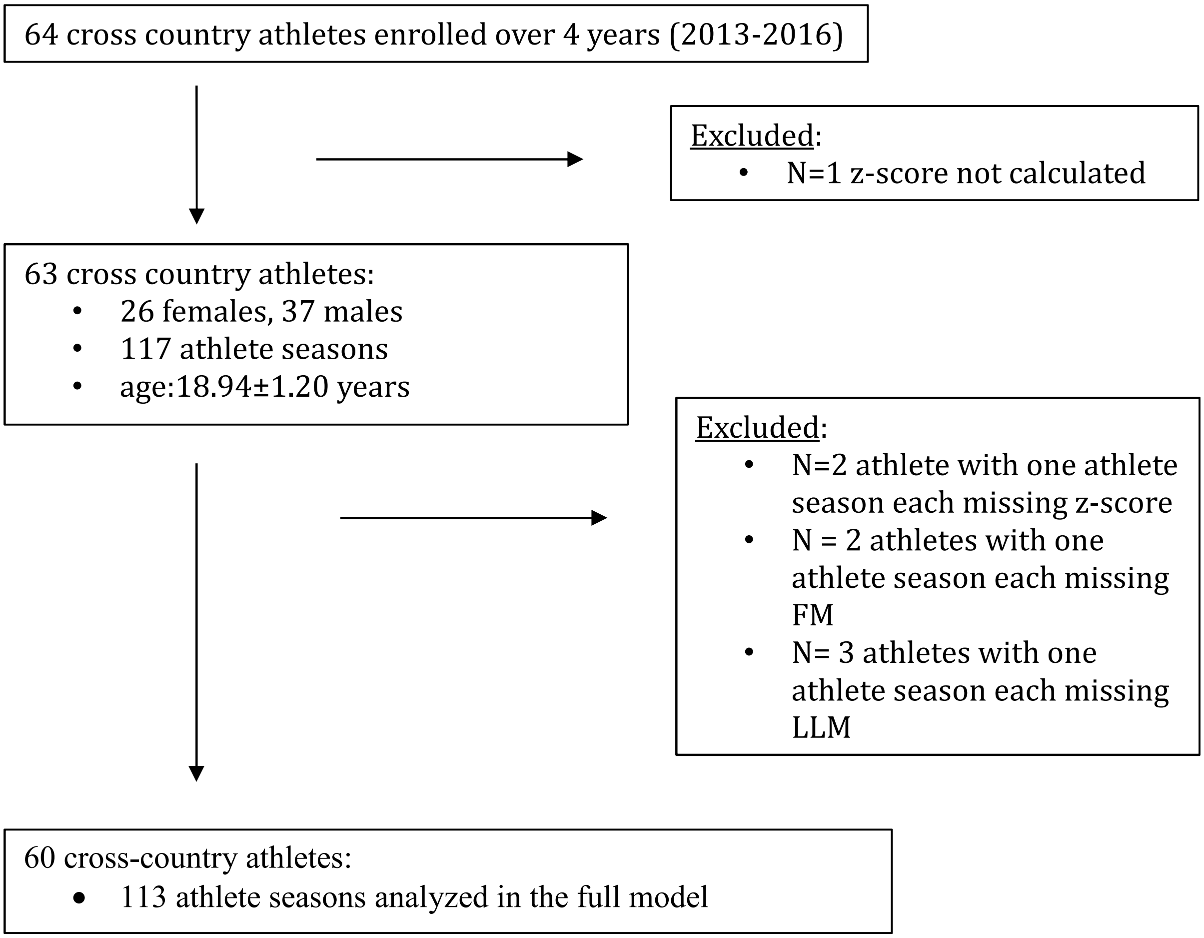


Figure 2: Diagram of participant flow through study for aim 3



Recruitment and Data Collection

Data for this thesis came from two UNC IRB approved parent research studies (PI: Smith-Ryan IRB 13-1664) [28] and (PI: Darin Padua IRB 13-2226). The participants who trained with the team during the 2016-2017 seasons were briefed on their role in each study. The returning athletes who were previously participants in the parent studies knew what the process entailed.[28] It was explained that each participant would complete laboratory screening prior to the season as well as a pre-participation screening. The laboratory screening included a Dual-energy x-ray (DEXA) scan for bone density, bone content and soft tissue composition (lean mass, leg lean mass, and fat mass) as well as ultrasound imaging over the quadriceps musculature (vastus lateralis).

The participants were told that in addition to the laboratory measures they would also complete paper surveys about past training and health history as a part of their pre-participation examination as directed by the sports medicine staff. BSI history, average training volume over the summer, and a question on menstrual status for females were on these forms. Each participant completed exit screenings post season including the same three questions of interest.

The Parent study (Smith-Ryan) had details on the laboratory screening process as it utilized data from these scans in previous research. [28] The subjects attended a single 30 minute testing session prior to training camp in early August.[28] The first measurements taken upon arrival were height, using a stadiometer (Perspective Enterprises, Portage, Michigan, USA) and weight with a digital scale (Health-o-meter, McCook, Illinois, USA).[28] Muscle quality and characteristics were measured by ultrasound and body composition measured by a whole body DEXA scan for the following variables BMC, BMD, FM, LM and LLM.[28] Detailed methods for collection these measures are on page 44 and 45.

Outcomes and Risk Factors of Interest

For aims one and two the independent variable examined was stress fracture history. For the purpose of this study, stress fracture history was defined as “any history of stress fracture prior to college.” The dependent variables or outcomes examined were bone mineral density, bone mineral content and muscle quality (mCSA and EI) respectively.

The main outcome of interest for aim three was incident BSI. The main exposure of interest was low bone mineral density defined by lower z-score. A z-score is a statistical measurement of a score's displacement about the average score, which is set to a standard of zero. The Z-score is related to the number of standard deviation above of below the population mean. Z-scores are used to compare bone density with ethnic, age and sex matched controls.[16] In the general population, a z-score of less than or equal to -2 is considered risky bone mineral density. Low bone density is theorized to increase the risk of stress fracture by reducing bone strength, making it easier during repetitive loading for micro trauma to occur.

Additional risk factors of interest included the following body composition measures: LM, LLM, FM, average mCSA, EI, BMD, and BMC; as well as sex, training volume, stress fracture history in the previous competitive season, and for females menstrual history. Table 1 separates the variables collected for this study into their respective measurement groups. These variables were all collected and analyzed in this study.

Table 1: Descriptive, Demographic and Research Variables of Interest

	Study	Years Collected			
		2013	2014	2015	2016
Demographic Variables					
Age (y)	Smith-Ryan	X	X	X	X
Sex	Smith-Ryan	X	X	X	X
Height (cm)	Smith-Ryan	X	X	X	X
Weight (kg)	Smith-Ryan	X	X	X	X
Variables from US					
Muscle Cross Sectional Area (mCSA) (cm^2)	Smith-Ryan	X	X	X	X
Echo Intensity (EI)	Smith-Ryan	X	X	X	X
Variables from DEXA					
Bone mineral content (BMC) (kg)	Smith-Ryan	X	X	X	X
Bone mineral density (BMD) (g/cm^2)	Smith-Ryan	X	X	X	X
Fat mass (FM) (kg)	Smith-Ryan	X	X	X	X
Lean mass(LM)(kg)	Smith-Ryan	X	X	X	X
Lower body lean mass (LLM) (kg)	Smith-Ryan	X	X	X	X
Health History Variables	Smith-Ryan	X	X	X	X
Menstrual cycle (normal/irregular)	Padua	X	X	X	X
Bone History (Stress fracture yes/no)	Padua	X	X	X	X
Training Volume/Intensity					
Weekly mileage (miles)	Padua				X
Average mileage pre-post season (miles)	Padua	X	X	X	X
Injury Surveillance Variables					
Incident stress reaction	Padua	X	X	X	X
Incident stress fracture	Padua	X	X	X	X

Instrumentation or variable measurement

The laboratory screening followed the methods of parent study (Smith-Ryan).[28] The subjects attended a single 30 minute testing session prior to training camp in early August.[28] The athletes had not begun training with the team and were training on their own for at least two months prior to the screening. The subjects reported to the testing session having fasted for two hours and had not completed any exercise for a minimum of two hours prior to testing. The first measurements taken upon arrival were height, using a stadiometer (Perspective Enterprises, Portage, Michigan, USA) and weight with a digital scale (Health-o-meter, McCook, Illinois, USA). Muscle quality and characteristics were measured by ultrasound and body composition measured by a whole body DEXA scan for BMC, BMD, FM, LM and LLM.

DEXA

Each subject underwent a full body DEXA scan (Apex Software version 3.3; Hologic Discovery W, Bedford, Ma, USA) or DEXA (GE Lunar iDXA, GE Medical Systems and Primary Care Diagnostics) performed by a trained technician. Before testing began the subjects were instructed to remove all metal, thick clothing and heavy plastic to reduce and noise or interference during the scan. Anthropometric variables such as age, height (cm), weight (kg) and ethnicity were entered into the system. Subjects were then asked to lie in a supine position on the center of the scanning table. Each independent variable of bone mineral content (kg) (BMC), bone mineral density (g/cm^2) (BMD), fat mass (kg) (FM), lean mass (kg) (LM), and leg lean mass, (LLM) (kg) were measured utilizing the scan. The test-retest reliability assessed by intraclass correlation coefficient (ICC) and standard error mean (SEM) in the laboratory for the parent study was: ICC = 0.98(kg) and SEM = 0.85(kg) for FM; ICC = 0.99(kg) and SEM = 1.07(kg) for LM.[28]

The bone mineral density data collected was utilized to create a profile of athletes who were “at-risk” versus those who were not “at-risk”. A z-score is a statistical measurement of a score’s displacement about the average score, which is set to a standard of zero. The Z-score is related to the number of standard deviation above of below the population mean. Z-scores are used to compare bone density with age, sex and ethnically matched controls.[16] In the general population, a z-score of less than or equal to -2 is considered risky bone mineral density. Low bone density is theorized to increase the risk of stress fracture by reducing bone strength, making it easier during repetitive loading for micro trauma to occur.

Due to these findings and for the purpose of this study analysis was completed by exploring two z-score cut points. The first was any athlete with a z score of -2.0 or lower was considered at-risk for a BSI. The data was then examined in quartiles and then designating the lower 25th percentile and below created the second cut point for at-risk athletes (z-score -1.5 or lower). Due to the small number of athletes with z-score cut point of -2 or less a multivariate analysis was not completed for this exposure variable.

Muscle Quality

Muscle cross sectional area (cm^2) (mCSA) of the vastus lateralis (VL) was determined using a GE Logic-e B-mode ultrasound (GE Healthcare Wisconsin, USA) from a panoramic scan. The settings of the scan were consistent for each.

(Frequency: 26 Hz, gain: 68, depth: 4.5 cm) Prior to completion of the scan the athlete was asked to assume a supine position for 5 minutes. A foam pad was strapped to the midpoint of the thigh in order to standardize the measurements. The right leg was then scanned for each patient three different times and the average mCSA was calculated by interpreting each scan. While the scan took place the patient was instructed to lie in a position allowing the right leg to be extended and relaxed on the table. The US probe (GE: 12L-RS) was aligned perpendicular to the direction of the muscle tissue and brought across the surface of the skin with an equal pressure from the lateral border to the medial fascia separation of the VL. Muscle cross sectional area was measured using imaging software (version 1.37; Image J, National Institutes of Health, Bethesda, MD, USA). The VL was traced and then analyzed by the Image J software. Test-retest reliability (October 2016) for the investigator for mCSA were ICC= 0.72cm^2 and SEM = 3.74 cm^2.

Echo intensity (EI) was determined using a gray scale imaging software, (Image J) in the standard histogram function of pixels ranging from 0-255. Prior to each measurement of EI, each image was calibrated by measuring the number of pixels within a known distance of 1cm. In order to measure EI, the primary investigator outlined each patient's VL along the fascia border to only capture the muscle itself. Test-retest reliability for the investigator (October 2016) for EI were ICC = 0.97 and SEM = 1.85. In the 2015 season there were 10 athletes who did not received their ultrasound scans so these athlete variables were not able to be included in the final analysis.

Menstrual History

The female subjects were asked to report their menstrual status at the beginning and end of the fall season. The subject's pre-participation examination questionnaire and exit surveys included a question on the status of the subject's menstrual health. The questions asked how frequently they have their period and if they are taking an oral contraceptive pill. Female athletes' menstrual cycle was designated as normal or "irregular" based upon their responses. Any athlete who answered they had their period regularly each month would be normal. Irregular cycles would be classified as an abnormal or absent cycle. Abnormal menstrual cycle included athletes that had a period but not on a normal cycle. Absent menstrual cycle was a response of not having a period in the past 4 months.

Injury History

In a similar format to the collection of menstrual history, all participating athletes were asked about their past injury history as it related to BSIs. During the pre-participation screening the subjects were asked to report if they ever had a history of BSI. If the athletes answered yes, they were asked to state how many, classify the injury and identify which bony structure in the lower extremity (sacrum, femur, tibia, fibula, ankle, foot, toes) the BSI was diagnosed in. The subject's BSI incidents while competing at the collegiate level were recorded and obtained from the Electronic Medical records system Blue Ocean as stated in the injury surveillance section.

Training Volume

Training volume data was collected in two ways for this study. In the first method training volume data was collected as self-reported baseline averages. The athletes were asked to report their average mileage per week prior to the beginning of the competitive year and at the end of the competitive year. The average mileage data from preseason allowed the athletes to be classified into one of two training groups, either above or below the median training MPW value for the cohort.

The second method of obtaining training volume was completed for only the 2016-2017 cohort of cross-country athletes. The athletes utilized an online software system in order to record their daily mileage. The software then computed their mileage for that week of training. The athlete's were given access to each of their own separate logins set up by the coaching staff prior to the season. At the end of the season, the documented mileage was collected and added to the athlete's profile for the current study. The mileage documented was then reduced to an average weekly mileage for the course of the season for each athlete.

Injury Surveillance

Injuries were monitored over the course of the competitive year by team athletic trainers and recorded in the electronic medical records (Blue Ocean, Dallas, TX).[28] When an injured athlete was initially evaluated they were entered into the system as a new injury incident. To be diagnosed with a BSI the athlete was seen by the team physician. The athlete first received an x-ray, which in most cases is not indicative of the injury at hand but must be used to rule out a full fracture of the bone in question. The athlete was then sent for a Magnetic Resonance Image (MRI) to determine the extent of the suspected BSI. The radiologist and physician interpreted the MRI and provided a report classifying the injury as a BSI. The specific site of BSI (includes sacrum, femur, tibia, fibula, ankle, foot, toes) and time lost was entered into the injury software. The data from new incoming athletes with past BSIs prior to their collegiate career was collected in the intake questionnaires.

In this study there were two BSI variables. In aims one and two the athlete’s history of stress fracture at baseline was utilized for analysis. An athlete, who had a stress fracture prior to their first year in the study, at any time, was included in the stress fracture history group. In aim three the stress fracture history variable is updated to include stress fracture and reactions, so both forms of BSI, incidence during the four competitive years of the study.

Statistical Analysis

All analyses were performed in Statistical Package for the Social Sciences (SPSS) (Version 23; IBM, Armonk, NY, USA) while the analysis for aim 3 was performed in the Statitsical Analysis System (SAS) (Version 94; SAS Institute, Cary, NC). To determine if a difference existed in BMD and BMC between collegiate cross-country athletes with a history of stress fractures compared to those without a history of stress fracture, a two-tailed independent sample t-test was performed. The grouping factor, or independent variable, utilized was stress fracture history and the dependent variable was mean bone mineral density in the first t-test and mean bone mineral content in the second. A t-test compares the means between groups. Statistical significance was set a priori at α=0.05.

For aim two, one-tailed independent samples t-tests was used to determine if there was a difference in muscle quality (mCSA and EI) between collegiate cross country-runners with or without a history of stress fractures. In this instance a one-tailed independent samples t-test was performed. The grouping variable for the test was again stress fracture injury history and the dependent variable utilized in the first t-test was mean mCSA. The second t-test evaluated mean EI as the dependent variable. A one-tailed independent samples t-test was chosen because the hypotheses for the second aim were given a direction of difference between the two groups. It was hypothesized that the mean of average mCSA in the stress fracture history group would be lower that the non-stress fracture history group. In opposition it was hypothesized that the mean of average echo intensity would be higher in the stress fracture history group compared to the non-stress fracture group. Statistical significance was set a priori at $\alpha=0.05$.

To address aim 3 a multivariate logistic regression was performed to determine the relative risk (expressed as an odds ratio) of BSI occurrence for athletes exposed to the risk factor of interest (z-score of -1.5 or below) compared to those who are unexposed (z-score greater than -1.5). The study assessed odd ratios to estimate the likelihood for a BSI occurrence over the competitive year. Odds ratios were defined as the odds of an incident BSI in the exposed group, divided by the odds of an incident BSI in the unexposed group.

First univariate analyses were completed to determine if each individual independent variable (BMD, BMC, mCSA, EI, LM, LLM, FM, training volume, stress fracture history and menstrual history) was associated with the odds of a BSI occurring. After this had been completed, presence of an interaction between sex and the effects of the main independent variable (z-score of -1.5 or below) on BSI incidence was evaluated. This was then used to evaluate whether the association between athletes with a z-score of -1.5 or below and BSI risk was different between males and females respectively. For example, if the odds ratio for the lower bone mineral density and BSI association was greater in females but had a lower value in males it was then determined prudent to create separate models for each sex.

For each of these models it had to be determined which confounders should be included in the initial multivariate model based on the findings of the univariate analysis. An odds ratio was assessed for each variable. A variable with an odds ratio 1.2 or greater, or 0.8 and below, with both the main exposure and outcome was considered an important factor to be included in the larger model. The initial multivariate model was then built by adding all associated variables at once and then completing backwards elimination technique of one variable at a time. Each variable was left in the model or removed based on its effect on the odds ratio of the main exposure of the model. The log of the confounding odds ratio equation was utilized to determine which variables to include in the final model: $\ln(coOR)=abs|\ln(unadjustedOR/adjustedOR)|*100$. When the variable was removed from the model, a 10% change or greater in the log of the confounding odds ratio for the main exposure-outcome and the variable was included as a confounder in the multivariate model. There were multiple competitive years per athlete in this study and Generalized Estimating Equations (GEE) were utilized to account for the lack of independence between the athletes who participated in multiple seasons.

Statistical Power

Statistical power estimations were conducted for each aim. To establish power for aims one and two, effect sizes (Cohen's *d*) for each variable of interest

BMD, BMC, and mCSA and EI of the vastus lateralis were calculated. A post-hoc power analysis was completed using G*Power statistical power analysis program (Düsseldorf, Germany).[57] This was completed by first calculating the pooled standard deviation between the groups for each variable. The pooled standard deviation and the mean of each group were then used to calculate Cohen's *d*, which was then entered into the G*Power software. A t-test for the difference in means between two independent groups was used. In previous research, comparisons were made between sexes, so the more conservative of the two power analyses was used for this study.

To calculate the Cohen's d for each variable a reference study had to be utilized in order to determine the effect size of the variable. The reference values for the variables BMD, mCSA and EI were all taken from the study completed by Roelofs et al. 2015.[28] The reference values for the last variable bone mineral density was taken from the study completed by Bennell et al. 1996.[4] Each study stratified their data by sex while the current study will look at the cohort as a whole. This being the case the more conservative of the two outcomes was utilized for the power analysis.

A sample size of 125 person years was set as the standard for the *n* of the group size. As investigators we have chosen to look at each individual at each year time point rather than grouping each data point as one. After computing the power for each variable it was determined that the study is 99.4% powered to detect changes in bone mineral density, 73.3% powered to detect changes in bone mineral content, 50.5 % powered to detect changes in muscle cross sectional area, and only 5.1% powered to detect changes in echo intensity.

For aim 3 of the study a cohort power analysis was completed. At an alpha level of 0.05 set *a priori*, with the unexposed to exposed ratio of 2.3 (more than twice the number of not at-risk body types compared to at-risk body types), and a 10 percent risk of stress fracture occurrence in the unexposed group (not at-risk body type), we estimated having at 80 percent power to detect risk ratios of 5.0 if 13 individuals are exposed to the at-risk body type and 10.0 if 5 individuals are exposed to the at-risk body type.

CHAPTER 4

Intrinsic and extrinsic risk factors for stress fracture among division one collegiate cross-country athletes

Introduction

When bone undergoes frequent and repetitive sub-threshold loading, micro-trauma can occur.[1] Over time, micro-trauma can develop into a fully defined fracture in the bone when excessive stress is applied to bone without adequate recovery for resoprtion and adaptation.[1] Lower extremity stress fractures are a common sports injury that affect high level athletes participating in endurance sports involving repetitive, high-intensity loading, such as military recruits and distance runners.[2-5] Stress fractures in the athletic population result in pain, loss of athletic participation, and high medical expenses.[6]

Stress fractures are injuries that plague a range of individuals, high volume athletes. Stress fractures are estimated as 10% of all athletic injuries while in the military an incidence rate as high as 31 cases in 100 persons has been reported.[1] Injury surveillance conducted by Arendt et al. (2003) found the incidence of stress fractures in collegiate athletes, in a ten year period, to be about 1%.[7] Of those stress fractures 15% occurred in runners.[7] A study completed specifically with track athletes concluded that over the course of one season 20% of all the injuries incurred were stress fractures.[3] A higher incidence rate of stress fractures have been found in cross-country athletes compared to other contact or non-contact team sports, with data demonstrating that 3.2% of the stress fractures occurred in distance athletes.[7] However, studies have shown that in this specific cohort of distance athletes, women are typically at a higher risk for developing stress fractures. A study completed by Nattiv et al. (2007), found the relative risk of stress fracture for women is 1.5-3.5 times higher when compared to their male counterparts.[8] These findings were similar to the study completed Ardent et al. (2003), where females had a reported incidence rate of 1.9% and males of 0.8%.[7] This prior research suggests that the collegiate distance runner population should be investigated when looking for risk factors that influence the development of bony stress injury.

Available research has explored a combination of factors that may put athletes at a higher risk for stress fracture incidences, including body composition, bone content, dietary intake, menstrual disturbances, training volume, and biomechanical variables.[4,6,7,34,35] Body composition measures of lower lean mass, smaller muscle girth and cross sectional area can be indicative of lower muscular strength and increased risk for stress fracture.[5,6,26] Athletes who have a history of stress fracture have been found to have significantly lower bone mineral density (BMD) and bone mineral content (BMC) scores when compared to healthy athletes.[3,4,9,24] A study conducted by Nieves et al. (2010) found that disordered eating patterns increased risk of stress fracture due to changes in BMD.[37] Specifically, increased consumption of dairy with low fat intake was significantly related to a reduced risk for stress fracture by an increased BMD.[37] Dietary disturbances can also affect menstrual disturbances.[6] Female athletes with a history of stress fracture typically have a history of delayed menarche and report fewer menstrual cycles per year leading up to injury diagnosis.[4] Athletes must also take into account training regimen as a risk factor. One study conducted in a cohort of college athletes found that approximately 48% of stress injuries to bone were correlated with a change in the training regimen.[7] Training changes and adaptations may also play a role in biomechanical changes in athletes.[51-53] Gait biomechanics have been related to higher stress fracture risk when evaluated during running in a fatigued state and at different grades of incline.[49-51]

Available incidence rates demonstrate the impact stress fractures may have on an athlete, affecting time loss and participation status.[6] The majority of studies to date have evaluated risk factors for stress fractures using retrospective approach. In order to better understand injury risk and develop clinical prevention strategies, prospective investigations of key risk factors that may influence the development of bony stress injury are critical[10] Bone stress injuries (BSI) have a multifactorial etiology; hence, a single factor is not able to definitively determine if an athlete will sustain a BSI.[10] The majority of previous studies have looked at multiple risk factors, but analyzed them separately, looking at their individual effects on stress fracture incidence. The few studies that have combined the variables for analysis have had little success in finding a significant grouping. Trends in different risk factor groupings have been noted in these studies, reporting relevant factors of sex, stress fracture history and bone mineral density (t-scores).[4,29,56] The current study sought to determine the combination of risk factors that place athletes at the highest risk for BSI. Such an investigation would help to provide information for future research and the future development of comprehensive screening tools to identify high-risk individuals during pre-participation examinations in sports medicine settings.

The purpose of this study was to determine which risk factors play a key role in the occurrence of BSIs in National Collegiate Athletic Association (NCAA) Division I (DI) cross-country athletes. The first aim of the study was to determine if a difference in bone composition, defined as bone mineral density (BMD) and bone mineral content (BMC), existed between collegiate cross-country runners with a history of stress fracture and those without. The second aim was to determine if a difference in muscle quality, defined as muscle cross sectional area (mCSA) and echo intensity (EI) of the vastus lateralis, existed between athletes with a history of stress fracture and those without. The last aim of this study was to determine if an association exists between body composition risk factors, and BSI risk in collegiate cross-country athletes. An exploratory aim evaluated the effects of training volume and menstrual history irregularities among females on the association between body composition risk factors and BSI risk. It was hypothesized that there would be a difference in mean BMD and BMD between groups. It was also hypothesized that athletes with a history of stress fracture would have smaller baseline VL mCSA and higher EI values indicating poor muscle quality. Lastly we hypothesized that there would be an association between body composition and BSI risk in collegiate cross country athletes when adjusting for other risk factors.

Methods

Participants

A cohort of 64 NCAA DI collegiate distance runners (27 females, 37 males; age:18.9±1.2 years) was followed over the course of their respective cross-country seasons at their institution from 2013 to 2016 (a total of 117 competitive athlete-years). Figures 1 and 2 depict the participant flow through the study for each aim. For this cohort, 63 of the 64 athletes data was utilized in the final analysis. Athlete demographics and descriptive statistics at baseline are shown in Tables 2-5.

Procedures

All study procedures were reviewed and approved by the Institutional Review Board at the University of North Carolina at Chapel Hill. Secondary data for this study came from two IRB-approved parent research studies. Data collected from the two previous studies was combined for analyses in the current study. The participants currently training with the team were briefed on their role in each study, and completed and signed informed consent. [28] Participants completed laboratory screening prior to the season as well as a routine pre-participation screening as a part of their annual pre-season sports medicine evaluation. The laboratory screening included measurements of body composition assessed from dual-energy X-ray (DEXA) and ultrasound imaging over the quadriceps musculature (vastus lateralis). The participants were also asked to complete paper based health history questions about past training and BSI history. The athletes filled these out as a part of their pre-participation examination.

The laboratory screening followed the methods of parent study which are described in a previously published study.[28] The subjects attended a single 30 minute testing session prior to training camp in early August.[28] The athletes had not begun training with the team and were training on their own for at least two months prior to the screening. The subjects reported to the testing session having fasted for two hours and had not completed any exercise for a minimum of two hours prior to testing. The first measurements taken upon arrival were height, using a stadiometer (Perspective Enterprises, Portage, Michigan, USA) and weight with a digital scale (Health-o-meter, McCook, Illinois, USA). Muscle quality and characteristics were measured by ultrasound and body composition measured by a whole body DEXA.

Instrumentation or variable measurement

DEXA

Each subject underwent a full body DEXA scan (Apex Software version 3.3; Hologic Discovery W, Bedford, Ma, USA or GE Lunar iDXA, GE Medical Systems and Primary Care Diagnostics) performed by a trained technician. Before testing began the subjects were instructed to remove all metal, thick clothing and heavy plastic to reduce and noise or interference during the scan. Anthropometric variables such as age, height (cm), weight (kg) and ethnicity were entered into the system. Subjects were then asked to lie in a supine position on the center of the scanning table. Bone mineral content (kg), BMD (g/cm^2) FM (kg) (LM (kg), %fat, and LLM were measured utilizing the scan. Test-retest reliability was assessed by interclass correlation coefficients (ICC) and standard error of the means (SEM). The test-retest reliability of these measures in the laboratory for the parent study was: ICC = 0.98 and SEM = 0.85 kg for FM; ICC = 0.99 and SEM = 1.07 kg for LM; and ICC = 0.98 and SEM = 1.06% for %fat.[28]

Muscle Quality

Muscle cross sectional area (cm^2) of the VL was determined using a GE Logic-e B-mode ultrasound (GE Healthcare Wisconsin, USA) from a panoramic scan. The settings of the scan were consistent for each. (Frequency: 26 Hz, gain: 68, depth: 4.5 cm) Prior to completing the scan the athlete was asked to assume a supine position for 5 minutes. A foam pad was strapped to the midpoint of the thigh in order to standardize the measurements. The right leg was then scanned for each patient. While the scan took place the patient was instructed to lie in a position allowing the right leg to be extended and relaxed on the table. The US probe (GE: 12L-RS) was aligned perpendicular to the direction of the muscle tissue and brought across the surface of the skin with an equal pressure from the lateral border to the medial fascia separation of the VL. Muscle cross sectional area was measured using imaging software (version 1.37; Image J, National Institutes of Health, Bethesda, MD, USA). The VL was traced (test-retest reliability for the investigator performed October 2016 for mCSA were ICC= 0.72 cm^2 and SEM = 3.74 cm^2) and then analyzed by the Image J software.

Utilizing the same panoramic scan taken of the VL EI was determined using a gray scale imaging software, (Image J) in the standard histogram function of pixels ranging from 0-255. Prior to each measurement of EI, each image was calibrated by measuring the number of pixels within a known distance of 1cm. In order to measure EI, the primary investigator outlined each patient’s VL along the fascia border to only capture the muscle itself. Test-retest reliability for the investigator performed October 2016 for EI were ICC = 0.97 a.u. and SEM = 1.85 a.u.

Covariates of Interest

Pre-participation examination questionnaire and exit surveys included questions on BSI history prior to collegiate career, average weekly mileage, and, for females, menstrual health. During the pre-participation screening the subjects were asked to report if they ever had a history of BSI. If the athletes answered yes, they reported how many, classified the injury as a stress fracture or reaction, and identified the location of BSI in the lower extremity (sacrum, femur, tibia, fibula, ankle, foot, toes). The athletes were asked to report their average mileage-per-week (MPW) prior to the beginning of the season. The average MPW data from preseason allowed the athletes to be classified into one of two training groups: above or below the median. Cut points were set for each of these groups by investigating the distribution of the variable at the end of the season. Females reported how frequently they had their menstrual cycle. Female athletes who reported they had their menstrual cycle regularly each month were classified as “regular”. Female athletes that had an irregular menstrual cycle or no menstrual cycle in the past 4 months were classified as “irregular”.

The second method of obtaining training volume was completed for only the 2016-2017 cohort of cross-country athletes. The athletes utilized an online software system (FinalSurge) in order to record their daily mileage. The software then computed their mileage for that week of training. The athlete’s were given access to each of their personal logins that the coaching staff set up prior to the season. At the end of the season, the documented mileage was collected and added to the athlete’s profile.

Incident BSI injuries were monitored over the course of the season by team athletic trainers and recorded in the electronic medical records (Blue Ocean, Dallas, TX).[28] Lower extremity BSI diagnoses were confirmed by the team physician, via x-ray and Magnetic Resonance Imaging (MRI). The radiologist and team physician interpreted the x-ray and MRI and diagnosed the BSI. The specific site of the BSI (sacrum, femur, tibia, fibula, ankle, foot, toes) was also recorded.

Statistical Analysis

Outcomes and Risk Factors of Interest

For aims one and two, the independent variable examined was stress fracture history. For the purpose of this study, stress fracture history was defined as "any history of stress fracture prior to college." The subject's stress fracture history prior to college from pre-participation questionnaire was used to classify participants into two groups: those with a stress fracture history and those without. The dependent variables examined were BMD, BMC and muscle quality (mCSA and EI) respectively. The main outcome of interest for aim three was incident BSI during the competitive year (August 1-July 31st). The risk factors of interest included: body composition variables (LM, LLM and FM), bone strength variables (BMD and BMC), sex (male and female), BSI history prior to the season (yes or no), training volume (above or below the median), and for females menstrual history (regular versus irregular).

All analysis for aims 1 and 2 were performed in Statistical Package for the Social Sciences (SPSS) (Version 23; IBM, Armonk, NY, USA). Analyses for aim 3 was performed in the Statistical Analysis System (SAS) (Version 94; SAS Institute, Cary, NC). For aim 1, a two-tailed independent sample t-test was used to determine differences in mean BMD and mean BMC between collegiate cross-country athletes with a history of stress fractures compared to those without a history of stress fracture. For aim 2, one-tailed independent samples t-tests were used to determine differences in mean muscle quality measures (mCSA and EI) between collegiate cross country-runners with or without a history of stress fractures. Statistical significance was set a priori at α=0.05.

To address aim 3 multivariate logistic regression was performed to determine the relative risk (expressed as an odds ratio) of BSI occurrence for athletes exposed to the risk factor of interest (lower bone mineral density scores) compared to those who are unexposed (higher bone mineral density scores) controlling for potential confounders. The BMD z-scores were taken from the DEXA output. In the general population, a BMD z-score of less than or equal to -2 is considered low or at risk.[16] Due to the published z-scores based on a general, but not highly active population, the present study explored two z-score cut points. Traditional cut-points (z-score of -2.0 or lower) were considered at-risk for an incident BSI. The second approach for cut-points examined the data in quartiles and then designated the lower 25th percentile and below as the cut point for at-risk athletes (z-score -1.5 or lower). Due to the small number of athletes with z-score cut point of -2 or less a multivariate analysis was not completed for this exposure variable.

First univariate analyses were completed to determine the association of each individual independent continuous body composition variable (BMD, BMC, mCSA, EI, LM, LLM, FM) followed by categorical variables sex, injury history, training volume, and menstrual irregularities with the odds of a stress fracture or reaction occurring. The presence of an interaction between sex and the effects of the main independent variable (lower bone mineral density) on BSI incidence was evaluated to determine if BSI risk was different between males and females respectively.

Confounders included in the initial multivariate model were based on the findings of the univariate analysis. A variable with an odds ratio of 1.2 or above and 0.8 or below was considered associated with the exposure or outcome. If the variable had an odds ratio, when assessed, as associated with both the main exposure and the outcome it was chosen as a covariate for the initial multivariate model. The multivariate model included all associated confounding variables and then backwards elimination technique, removing one variable at a time, was used to determine the final model. Each variable was left in the model or removed based on its effect on the odds ratio of the main exposure (Z-score ≤ -1.5) of the model. The log of the confounding odds ratio equation was utilized to determine which variables to include in the final model: ln(coOR)=abs|ln(unadjustedOR/adjustedOR)|*100. When the variable was removed from the model, a 10% change or greater in the log of the confounding odds ratio for the main exposure-outcome, and the variable was included as a confounder in the multivariate model. There were multiple competitive years per athlete in this study and Generalized Estimating Equations (GEE) were utilized to account for the lack of independence between the athletes who participated in multiple seasons. The initial multivariable model included the main exposure (z-score ≤ -1.5), along with stress fracture history; sex, FM and LLM as each were associated with the main outcome of interest incident BSI and main exposure of a z-score of -1.5 or below. After backwards elimination utilizing the log of the confounding odds ratio was completed and each covariate was assessed for confounding, the final model was determined.

Results

The athletes were followed for a total of 117 competitive years: 26 were followed for 1 competitive year, 23 followed for 2 competitive years, 11 followed for 3, and 3 competitive years followed for 4. (Table 2) Average age for the cohort at baseline was (19.3±1.4 years). Average height for the cohort was (68.5±3.5 in), average weight (134.4±18.2 lbs), and average training mileage pre-season (59.2±19.4 miles).

Aims 1 and 2

There was a significant difference between BMC at baseline (t =2.94, p = 0.02) between athletes with a history of stress fracture (2.1±0.3 kg) and athletes without (2.5±0.5 kg) (Table 3). There was no significant difference in BMD at baseline (t = 1.71, p = 0.09) between athletes with a history of stress fracture (1.1±0.1 g/cm^2) and athletes without (1.2±0.1 g/cm^2). Average mCSA at baseline was not significantly lower (t = 0.89, $p > 0.05$) among athletes with a history of stress fracture (19.0±3.9 cm^2) compared to athletes without (20.2±4.5 cm$^{2)}$). There was a significant difference between average EI at baseline (t = -1.83, $p < 0.05$), such that athletes with stress fracture history had higher average EI values (73.8±7.0 a.u), thus poorer muscle quality while athletes without a history of stress fracture had lower mean EI values (69.83±7.26 a.u) (Table 3).

Aim 3

There were 21 incident BSIs over the course of 117 competitive years distributed among 63 athletes: 17 stress fractures and 5 stress reactions (Table 4). Females had twice as many incident BSIs as males (n = 14 and n = 7 respectively). Athletes with a history of stress fracture had more incident BSIs (n = 13) when compared to athletes with no stress fracture history (n = 8). Athletes who were running 60 miles or below prior to the season (median cut point) had more incident BSIs (n=13) when compared to athletes above 60 miles a week (n=8) (Table 4).

All body composition variables were normally distributed except for lean mass. Lean mass had two notable outliers of 5.25 kg and 5.76 kg while the majority of lean mass measures were between 32 kg-62 kg. The reported variance for lean mass was also 91.63 kg. Table 5 shows the univariate odds ratios for association between incident BSIs and continuous body composition variables. BMC (OR=0.32; 95% CI: 0.09, 1.15) and BMD (OR=0.10; 95% CI: 0.00, 17.22) were associated with the outcome but were highly correlated with the z-score and were not included in the model (see Result Table 5 and Appendix Table 17). Fat mass (OR=1.21; 95% CI: 0.99, 1.48) and LLM (OR= 0.84; 95% CI: 0.70, 1.01) were also associated with incident BSI and were assessed as confounders in the model. None of the associations except for Average Echo Intensity were statistically significant as each of the 95% CIs reported included one. Average echo intensity was statistically significant but not associated with the outcome (OR= 1.09; 95% CI: 1.03; 1.15). Table 6 shows the univariate odds ratios between incident BSI and categorical variables. For the purposes of the current study, a z-score of -1.5 and below was utilized as the main exposure for the multivariate model. The unadjusted association between z-scores of -1.5 and below and BSI was 2.22 (95% CI: 0.61, 8.06). The unadjusted odds ratios for sex, and injury history and BSI were as follows: stress fracture history, with yes being the exposed group (OR=3.75; 95% CI: 1.41, 10.03), sex with females being the exposed group (OR=3.33; 95% CI: 1.23, 9.03).

Lastly the univariate odds ratios of association between the main exposure (z-score of -1.5 or below) and categorical variables of interest for the model were evaluated. Stress fracture history (OR=3.75; 95% CI: 1.41, 10.03) and sex (OR=0.82, 95% CI: 0.25, 2.69) were both associated with the main exposure. Both covariates were statistically significant (95% CI did not include one). The final covariates associated with incident BSI included in initial multivariate models were stress fracture history, with yes being the exposed group, sex with females being the exposed group, FM (FM as continuous variable) and LLM (LLM as a continuous variable).

Due to the fact that FM and LLM were correlated with one another (Appendix Table 17), these variables were not included in the total model together and LLM was evaluated after FM. After completing backwards elimination, FM was removed from the model as it did not result in a 10% change or greater in the odds ratio for z-score. Elimination of stress fracture history, sex and LLM resulted in changes in the odds ratio for z-score by 19.3%, 11.8% and 11.5% respectively. The three variables, stress fracture history, sex and LLM were all kept in the final model as covariates and the adjusted odds ratios for the final model (Table 6). Athletes with a z-score of less than or equal to -1.5 were over two times more likely to incur an incident BSI compared to athletes with z-score greater than -1.5 adjusting for sex, previous stress fracture injury, and LLM (OR=2.31; 95% CI: 0.58, 9.22). Athletes with a stress fracture history were over four times more likely to incur an incident BSI than athletes without, when adjusting for z-score, sex, and LLM (OR=4.29; 95% CI: 1.50, 12.25). Females were over two times more likely to incur an incident BSI when compared to males adjusting for z-score, stress fracture history and LLM (OR=2.46; 95% CI: 0.56, 10.78). Lastly, every one unit increase in leg lean mass increases the risk of stress fracture by 1.04 times when adjusting for the other variables in the model (OR=1.04; 95% CI: 0.79, 1.36).

The other variables explored for association between z-score of -1.5 and below and BSI were LM, training volume (above and below the median) and menstrual history. Menstrual history was unable to be evaluated as there were no females with an irregular menstrual cycle that had a z-score of -1.5 or below or had an incidence BSI (Tables 7and 8). Lean mass and training volume were not included in the final model. Lean mass was not associated with the exposure (z-score of -1.5 or below) (OR=1.01; 95% CI: 0.93, 1.11) or the outcome (BSI) (OR=0.95; 95% CI: 0.89, 1.00). Training volume was associated with BSI incident (OR=1.52; 95% CI: 0.58, 4.02) but was not associated with z-score -1.5 or below (OR=1.03; 95% CI: 0.32, 3.29).

Discussion

The study conducted in DI cross-country athletes found that there was a significant difference in BMC and EI between the stress fracture history group and non-stress fracture history group at baseline. However, there was no significant difference between BMD and mCSA between the two groups at baseline. The third aim of the study was focused on assessing BSI risk in the cohort when followed over four competitive years. The study conducted found that athletes who had a z-score of -1.5 or below when adjusting for stress fracture history, sex and LLM were two times more likely to incur a BSI incident. The same athletes with a history of stress fracture when adjusting for the variables mentioned prior were four times more likely to incur a subsequent BSI and female athletes were two times more likely to incur a BSI compared to males. Lastly each one-unit increase in LLM resulted in a 4% increased odds of a BSI.

Aims 1 and 2

The current study of DI cross-country athletes found there was no significant difference between athletes with a history of stress fracture and athletes without a history of stress fracture in BMD or mCSA at baseline. The study did find however that there were significant differences between BMC and EI between athletes with a history of stress fracture and those without.

These findings were similar to previous research[5, 26] that found BMD at the site of previous fracture was not significantly different between athletes with or without a history of stress fractures. This study did differ from the current study as the current study assessed BMD of the total body between athletes with a history of stress fracture and those without. The findings for mCSA in the current study were different than findings conducted in military recruits.[5] Beck et al., (2000) found that military recruits with stress fracture incidences had smaller mCSA of the thigh musculature when compared to healthy controls.[5] Similar findings in mCSA values were also noted in a study conducted by Popp et al. (2009) in runners with a history of stress fracture.[29] The findings of the current study and the literature referenced above suggested BMD and mCSA may take time to return to their pre-injury state. After injury, bone healing may take up to 4-6 weeks.[11] Athletes typically returned to activity after this time frame; pre-mature return to sport may lead to further breakdown of the bone mineral due to inadequate time for tissue healing.[11] This physiological process may explain why athletes with a history of stress fracture have BMD values less than athletes without stress fracture history. Muscle is thought to act on bones to oppose bending and torsional forces that occur during weight bearing activity.[26] After injury, athletes decrease activity and physical activity; an athlete with a smaller mCSA at baseline may be affected more dramatically due to lack of use and atrophy. As a result, previously injured athletes may lag behind in muscle development when compared to athletes with no previous stress fracture injuries.

In the present study, athletes with a history of stress fracture had a significant difference in BMC (lower) and EI (greater) when compared to athletes with no history of stress fracture. These results differed from one study similarly conducted in DI cross-country athletes, that reported no significant differences between athletes with stress fracture history and those without in any body composition variables.[28] A study by Bennell et al. (1996) found women who developed stress fractures had lower values for total BMC, however the same pattern was not seen in males.[4] The findings from the current study in conjunction with Bennell et al. (1996) may suggest athletes with stress fracture history may be at a greater risk of re-injury due to already lower BMC at entry into a new distance program. These findings indicate the need for further research into body composition values at baseline in distance athletes in order to determine their potential risk for recurrent injuries in the lower extremity.

Aim 3

Approximately one- to two-thirds of cross-country athletes and long distance runners have a history of BSIs.[10] In about 10.3% to 12.6% of cases athletes with stress fracture history sustain recurring BSIs when prospectively followed for 1-2 years.[13] The current study found in 11.1% of athletes with stress fracture sustain subsequent stress fractures or reactions.

In this study female athletes had two-times the number of BSI incidents when compared to their male counterparts. This data coincided with previous literature suggesting women are at a higher risk for incident stress fractures when compared to males.[7,16] A study conducted in Navy midshipman and midshipwomen also found that females had a higher incidence rate (8.4%) compared to males (2.3%).[58]

The current study examined the association between a z-score of -1.5 and below and incident BSI adjusted for stress fracture history, sex and fat mass (FM). The final logistic regression model indicated that cross-country athletes with a z-score of -1.5 or below were 2.06 times more likely to have an incident BSI during the year adjusted for sex and previous stress fracture history. Previous studies had conflicting results about the association of bone quality measures and stress fracture outcome. Bennell et al. (1999) found that BMD values are predictive of stress fracture risk in females, but was not always predictive in males.[10] Another study conducted by Bennell et al. (1996) found that males with a history of stress fracture did tend to follow a trend toward lower BMD scores.[4] However, these studies did not control for other factors such as mCSA, EI and total body fat mass when looking at the associations between stress fracture incidence or history respectively with BMD values.[6] Incidence in the present study was not stratified by sex due to low available sample size.

In a study by Armstrong et al., (2004) conducted in first-year military cadets who were matched for, age, weight and BMI with healthy subjects, cadets who suffered a bony stress injury lost more than four times as much weight as controls by the date of their stress fracture.[58] These findings were consistent in both male and female cadets.[58] In the current study, a one-unit increase in fat mass increased the chance of incurring a BSI incident by 1.2 times. In contrast to the current study, Armstrong et al., (2004) found that female cadets and controls had no significant difference in BMD values, but male cadets showed trends towards lower values in BMD at the non-dominant hip.[58] The current study, which assessed total body BMD values and z-score distribution in males and female as one cohort found that subjects with a z-score of -1.5 or below were 2.31 times more likely to incur a stress fracture or reaction.

The current study utilized whole body values due to the use of z-scores as the main exposure. Another study conducted in Israeli male military recruits evaluated the association between regional BMD and BMC and stress fracture incidence.[50] The study found that unadjusted BMC and BMD values were similar between stress fracture and non-fracture groups but when adjusted for age, height, weight, exercise, smoking, alcohol and calcium intake; values for femoral neck BMC and BMD were lower in stress fracture group.[50] This differed from the current study, which utilized total BMC and BMD values. Lower BMC and BMD were both associated with BSI incidents as well as lower z-score values. They were not included in the final analysis however as they were highly correlated with each other (Appendix Table 17). Z-scores are BMD measure transformed and reported as standard deviation above and below the population means, which explains why they would be so highly correlated with one another. The study completed in the military recruits also accounted for a number of covariates in their analysis that were not accounted for in the current study.

Strengths and Limitations

Strengths of the current study were its prospective nature, incident injury assessment, and baseline body composition measures. The current study was able to assess association between baseline risk factors and stress fracture and reaction incidences in the following season. Each of the body composition variables can be assessed by the completion of one DEXA scan. The scans took only 10-15 minutes. Each subject was exposed to very limited radiation. A strength of this method was its ability to give information on specific body segments.[19,20,23] The technique can also be used with individuals who cannot undergo the procedures for hydrostatic weighing. All the individual must do is lay still for the entirety of the scan itself. The DEXA had no specific contraindications other than pregnancy making it a technique that can be used with a wide population of individuals when available.[20,23] The variables included in the final model, stress fracture history, and sex were variables that are normally accounted for in pre-participation examinations of cross-country athletes preceding their training season. This was clinically relevant because athletic trainers at the pre-participation examination can gather each of the variables found in the study that were associated with BSI incidents.

However, there are several limitations to DEXA scans. In some cases, the size of the table may be too small to accommodate a taller or wider individual and cause a deviation in measurements. Results may have differed between machines due to differences in the type of X-ray utilized by the unit. [22] Precision of the scan has also been found to be lower in the obese population.[20] Other limitations of the DEXA scan can be caused by two types of errors these are either technical or biological. Technical errors may be generated by the machine, incorrect placement of the subject or inaccurate image post processing. Biological variations can be due to hydration status, exercise, food intake as well as long term changes due to diet an exercise.[20] These types of errors can be mostly avoided by following a standard protocol when scanning each subject.

One large limitation in the current study is attributed to the small sample size, resulting in a lack of statistical power for several analyses. As a result analyses stratified by sex could not be completed. A second limitation was the 10 missing ultrasound scans (2015) to evaluate vastus lateralis mCSA and EI. Two of these missing scans were from year-one participation scans. These athletes could not be used in the t-test analyses in aims 1 and 2 of the study. Another limitation related to the small sample size was the fact that only one team was followed for this study. This makes the findings of this study only relevant to this sample and not generalizable to the cross-country population as a whole. Another limitation in the study was the inability to look at menstrual status at baseline as a confounder in the model. There were no females with either a z-score of -1.5 or below or an incident BSI who were reported having an irregular menstrual cycle at baseline. As a result, odds ratio for the association between exposure and menstrual cycle irregularity and BIS and menstrual cycle irregularity were unable to be assessed. (Tables 15 and 16)

This study was also unable to address an original sub aim of assessing the effect weekly training volume during the season, had on the association between the covariates and incident BSIs. The training data reported over the course of the 2016-2017 season was incomplete as athlete compliance with recording their mileage was poor (only 1 completed logs for all weeks, 24 athletes completed partial logs). Therefore, the current study was only able to look at baseline average weekly training load and its association with the BSI outcome and covariates. A final limitation of the current study was the lack of accounting for dietary intake, hormonal changes, bone geometry and length and baseline training volume in the body during training, which was accounted for in the studies completed by Valimaki et al (2005) and Armstrong et al. (2004).[50,58]

Conclusions and Clinical Implications

It can be concluded from this study that athletes with a history of stress fracture had lower BMC and higher quadriceps EI values at baseline examinations. There was no significant difference between quadriceps mCSA and BMD at baseline in cross-country athletes who had a history of stress fracture and those who did not. However, when assessed prospectively athletes with z-scores -1.5 or lower, an adapted measurement of BMD to compare the athletes to the population means, was associated with an incident BSI.

More specifically athletes with a z-score of -1.5 or below were at an increased risk (2.31 times higher) for BSI when compared to athletes with a z-score above -1.5. Sex and stress fracture history were also associated with incident BSIs. Females were 2.46 times more likely to suffer a BSI incident while athletes with a history of stress fracture were 4.29 times more likely to suffer an incident BSI. Leg lean mass was also associated with BSI incident such that every one unit increase in LLM increased risk by 4 percent times.

Our results revealed that there are multiple risk factors that should be assessed when determining which athletes may be at an increased risk for BSIs. All variables in aims 1 and 2 were modifiable. BMD and BMC can be addressed by nutritional counseling and support while BMC, BMD, EI and mCSA can be addressed by normal bouts of resistance training. Wolfe's Law states that tissue changes in form or function due to definite changes of the composition of the tissue in accordance to forces being applied to them.[16] Resistance training can healthily stress the bone tissue and create positive bone growth as well as muscle development. Some of the variables included in aim 3 of the current study were however, non-modifiable at baseline such as sex and stress fracture history (patient history and biological makeup). However, these variables could be assessed at baseline and athletes with these risk factors can be more closely monitored during the course of the training and competition cycles to improve prevention of BSI. The subsequent risk from a positive stress fracture history can be affected and changed as the athlete is competing at the university level. These athletes should be educated on common signs and symptoms, as well as other factors that have proven to increase stress fracture risk such as diet, training, menstrual history in females and body composition measures that have been explored in previous research.[1, 3,18,34,35] Coaching staffs should also be made aware of which athletes they may have to adapt training styles to in order to accommodate healing time and bone remodeling in order to prevent overuse and thus incident BSIs.

The current pre-participation examinations (PPE) should also attempt to be as specific as possible. This is especially important in regards to means in assessing training volume, menstrual history and injury history. Athletes should be asked to specifically state their average weekly mileage for each month in the summer as they may be apt to misrepresent training if asked for only one number on average. Female athletes should be asked for the date of last menstrual cycle as well as if they take any oral contraceptives to determine the adequacy of their menstrual history responses. All athletes should be required to report any past BSI injuries, indicate whether they were a stress fracture or reaction, and indicate the date of injury, as athletes who report past BSIs could be indicating an injury that had occurred multiple years ago. It may be prudent to determine if an athlete with multiple BSIs or less BSIs closer to the current season are at a higher risk. In addition to the PPE if training load is of interest it would be pertinent to mandate that athletes record their weekly training mileage through out the season in order to determine trends in the injury occurrences. Each of these factors could play a key role in unlocking more pertinent variables in a BSI prevention model.

Other studies have explored the association of these variables with each other, as well as their association with bone content variables.[28] It would be beneficial for clinicians to focus on risk factors they can measure and implement change in order to create prevention strategies. Future studies should follow athletes over multiple seasons and with a larger participant pool. A more wide spread group of athletes such as different collegiate divisions or high school can broaden the generalizability of the study. Serial data collection can also allow researchers and athletic trainers to see patterns and changes in risk factors and BSI over time. The best scenario for NCAA D1 athletes would be serial collection four times over the competition year: at baseline for cross-country season, baseline going into indoor track, again at the beginning of outdoor and then at the end of the competitive year. This however may not be feasible due to differences in roster sizes and resources available to the team. This can also prove difficult, as the athletes will not all finish at the same time during the seasons. Athletes may qualify individually for post-season competition and train longer than others in their cohort. At the least baseline screenings should be completed each year to determine changes in athletes body composition and medical history that may put athletes at risk for a BSI the following competitive year.

Creating a more inclusive model would allow clinicians to be able to identify and work with modifiable risk factors to thus reduce stress fracture and reaction incidences during cross-country training. It would also be beneficial to assess differences in the model between the sexes, as menstrual history has also been found to be associated with females who have a history of stress fracture and stress fracture incidences. Males and females are biologically and hormonally different so it would make sense their bodies would react to potential risk factors differently. The current study provides a stepping-stone for future research in multi-factorial assessments of BSI risk in cross-country athletes. Future research should look to evaluate models in each sex for body composition variables, healthy history and nutrition, as this study was unable to include all risk factors currently being studied.

RESULTS TABLES

Table 2: Demographic statistics of the cohort of collegiate cross-country athletes as a whole

	n	%
Sex		
Males	37	58.7
Females	26	41.3
Seasons		
1	26	41.2
2	23	36.5
3	11	17.5
4	3	4.8
Bony Injury incidences		
Stress fracture		
Stress reaction	17	26.9
Number of fractures per athlete	5	7.9
0		
1	47	74.6
2	15	23.8
	1	1.60
Total athletes	63	100.
Total Athlete-seasons	117	

Table 3: Descriptive statistics for cross-country athletes at baseline stratified by stress fracture history

	Stress fracture history			No stress fracture history		
	N	Mean ± SD	95% CI	N	Mean ± SD	95% CI
Average muscle cross sectional area(cm$^{2)}$	15	19.0±3.9	(16.9, 21.2)	45	20.2±4.5	(18.8, 21.5)
Average echo intensity (a.u)	15	73.8±7.0	(69.9, 77.7)	45	69.8±7.3	(67.7, 72.0)
Fat mass (kg)	15	10.8±1.9	(9.9, 11.9)	48	10.1±2.4	(9.4, 10.8)
Lean mass (kg)	15	44.9±7.6	(40.6, 49.1)	48	47.7±10.2	(44.8, 50.6)
Lower leg lean mass (kg)	15	16.1±2.8	(14.6, 17.7)	47	17.9±3.2	(16.9, 18.8)
Bone mineral content (kg)	15	2.1±0.3	(1.9, 2.3)	48	2.5±0.5	(2.3, 2.6)
Bone mineral density (g/cm$^{2)}$	15	1.1±0.1	(1.0, 1.2)	48	1.2±0.1	(1.1, 1.2)
z-score	15	-0.4±1.2	(-1.1, 0.3)	48	-0.1±0.9	(-0.4, 0.1)

Table 4: Demographic distribution of categorical covariates stratified by incident bone stress injury (BSI) during the follow-up (Aug 1-Jul 31 each year)

Covariate	Incident BSI			
	Yes		No	
	Athlete-years	(%)	Athlete-years	(%)
Male athlete-seasons	7	(33.3)	60	(61.8)
Female athlete-seasons	14	(28.0)	36	(72.0)
Stress fracture history (Y)	13	(62.0)	29	(30.2)
Stress fracture history (N)	8	(38.0)	67	(69.7)
Menstrual History (Regular)	0	(0.0)	12	(75.0)
Menstrual History (Irregular)	30	(100.0)	4	(25.0)
Average miles pre (above median of 60)	8	(38.1)	44	(48.4)
Average miles pre (below median of 60)	13	(61.9)	47	(51.6)

Table 5: Univariate odds ratios for association between incident bone stress injuries (BSIs) and continuous body composition variables

	Beta (SE)	OR (95% CI)
Average muscle cross sectional area	-0.09 (0.08)	0.91 (0.77, 1.07)
Average echo intensity	0.08 (0.03)	1.09 (1.03, 1.15)
Bone Mineral Content (BMC)*	-1.13 (0.65)	0.32 (0.09, 1.15)
Bone Mineral Density (BMD)*	-2.29 (2.62)	0.10 (0.00, 17.22)
Lean mass*	-0.06 (0.03)	0.95 (0.89, 1.00)
Leg lean mass	-0.17 (0.09)	0.84 (0.70, 1.01)
Fat mass	0.19 (0.10)	1.21 (0.99, 1.48)

Table 6: Unadjusted and adjusted odds ratios for association between incident bone stress injury (BSI) and covariates

	Unadjusted OR (95% CI)	Adjusted OR (95% CI)
z-score		
-1.5 or lower	2.22 (0.61, 8.06)	2.31 (0.58,9.22)
> -1.5 (ref)	1.00	1.00
Stress Fracture History		
Yes	3.75 (1.41, 10.03)	4.29 (1.50, 12.25)
No (ref)	1.00	1.00
Sex		
Female	3.33 (1.23, 9.03)	2.46 (0.56, 10.78)
Male (ref)	1.00	1.00
Fat Mass	1.21 (0.99, 1.48)	-
Leg Lean Mass	0.84 (0.70, 1.01)	1.04 (0.79, 1.36)

Table 7: Stratified analysis of baseline menstrual history and z-score -1.5 or below versus above -1.5

		Menstrual History at Baseline		
		Normal	Absent/Irregular	Total
z-score (-1.5 or below)	Yes	5	0	5
	No	36	5	41
Total		41	5	46

Table 8: Stratified analysis of baseline menstrual history and incident bone stress injury (BSI) versus none

		Menstrual History at Baseline		
		Normal	Absent/Irregular	Total
Incident stress fracture/reaction	Yes	12	0	12
	No	30	4	34
Total		42	4	46

APPENDIX A: ADDITIONAL TABLES AND FIGURES

Table 9: Cross-country athlete categorical demographics at baseline stratified by sex

	All Participants (N=63)		Men (N=38)		Women (N=25)	
	n	%	n	%	n	%
Stress fracture history						
Yes	15	23.8	7	18.4	8	32.0
No	48	76.2	31	81.6	17	68.0
Stress fracture 12 mo. prior						
Yes	13	20.6	6	15.8	7	28.0
No	50	79.4	32	84.2	18	72.0
Stress Fracture during season						
Yes	9	14.3	3	7.9	6	24.0
No	54	85.7	35	92.1	19	76.0
Stress reaction history						
Yes	2	3.2	2	5.3	0	0.0
No	61	96.8	36	94.7	25	100.0
Stress reaction during season						
Yes	3	4.8	1	2.6	2	8.0
No	60	95.2	37	97.4	23	92.0
Menstrual history (pre)						
Yes	22	33.4			22	84.0
Irregular	3	4.8			3	12.0

Table 10: Cross-country athlete demographics at baseline stratified by stress fracture history

	Stress Fracture History			No Stress Fracture History		
	N	Mean ± SD	95% CI	N	Mean ± SD	95% CI
Age (years)	15	18.9±1.4	(18.5,19.2)	48	19.3±1.1	(18.5, 20.0)
Height (in)	15	68.9±2.4	(67.9, 70.1)	48	66.9±3.7	(65.6, 68.3)
Weight (lbs)	15	136.4±16.9	(130.9, 141.8)	47	128.1±18.4	(118.8, 137.5)
Average miles (pre)	15	59.0±22.6	(46.5, 71.5)	47	59.2±18.6	(53.8, 64.7)

Table 11: Descriptive Statistics for sample at baseline (year 1)

	(N)	Minimum	Maximum	Mean±SD
Height (in)	63	59.0	75.0	68.5±3.5
Weight (lbs)	62	90.2	167.0	134.4±18.2
Avg_mCSA(cm$^{2)}$	60	11.9	32.2	19.9±4.4
Avg_EI (a.u)	60	53.1	84.2	70.8±7.4
FM (kg)	63	5.7	15.9	10.3±2.3
LM (kg)	63	5.7	62.3	47.0±9.6
LLM (kg)	62	11.7	23.9	17.5±3.1
BMD (g/cm^2)	63	0.9	1.4	1.1±0.1
BMC (kg)	63	1.3	3.7	2.4±0.5
z-score	63	-2.4	1.9	-0.2±1.0
avg_miles (pre)	62	15.0	100.0	59.2±19.4

Table 12: T-test results comparing mean bone composition differences at baseline between cross-country athletes with stress fracture history at baseline and those without

	Mean difference ± SD	*t*	df	p-value
BMC	-0.3 ± 0.1	2.9	61	0.02
BMD	-0.1 ± 0.03	1.7	61	0.09

Table 13: T-test results comparing mean muscle composition differences at baseline between cross-country athletes with stress fracture history at baseline and those without

	Mean difference ± SD	*t*	df	p-value
Average muscle cross sectional area	1.2±1.3	0.9	58	$p > 0.05$
Average echo intensity	3.9±2.1	-1.8	58	$p < 0.05$

Table 14: Univariate odds ratios for association between z-score of -2 or below and continuous body composition variables odds ratios

	Beta (SE)	OR (95% CI)
Average muscle cross sectional area	-0.05 (0.11)	0.95 (0.77, 1.12)
Average echo intensity	-0.06 (0.06)	0.94 (0.83, 1.06)
Lean mass	0.01 (0.04)	1.01 (0.93, 1.11)
Leg lean mass	-0.17 (1.51)	0.84 (0.70, 1.01)
Fat mass	0.21 (0.23)	1.21 (0.78, 1.93)

Table 15: Univariate odd ratios for association between z-score of -1.5 or below and all continuous body composition variables

	Beta (SE)	OR (95% CI)
Average muscle cross sectional area	0.05 (0.08)	1.05 (0.90, 1.22)
Average echo intensity	-0.06 (0.04)	0.94 (0.87, 1.02)
Lean mass	0.04 (0.03)	1.04 (0.98, 1.09)
Leg lean mass	0.17 (0.11)	1.20 (0.96,1.47)
Fat mass	0.06 (0.13)	1.20 (0.98, 1.46)

Table 16: Univariate odds ratios of association between exposure (z-score of -1.5 or below) and categorical variables

	OR (95% CI)
Stress Fracture History	
Yes	3.75 (1.41, 10.03)
No (ref)	1.00
Sex	
Female	0.82 (0.25, 2.69)
Male (ref)	1.00
Average Miles per Week (MPW) (Pre-season)	
Below 60 MPW	1.03 (0.32, 3.29)
Above 60 MPW	1.00

Table 17: Correlations between continuous body composition variables

		avg_mCSA	avg_EI	BMC	BMD	z-score	FM	LM	LLM
avg_mCSA	Pearson Correlation	1	-.444**	.538**	.399**	.114	-.186	.465**	.602**
	Sig. (2-tailed)		.000	.000	.000	.257	.061	.000	.000
	N	104	104	101	102	100	102	101	102
avg_EI	Pearson Correlation	-.444**	1	-.299**	-.367**	-.147	.444**	-.310**	-.294**
	Sig. (2-tailed)	.000		.002	.000	.144	.000	.002	.003
	N	104	104	101	102	100	102	101	102
BMC	Pearson Correlation	.538**	-.299**	1	.830**	.484**	-.142	.615**	.804**
	Sig. (2-tailed)	.000	.002		.000	.000	.131	.000	.000
	N	101	101	114	114	111	114	113	112
BMD	Pearson Correlation	.399**	-.367**	.830**	1	.840**	-.127	.374**	.446**
	Sig. (2-tailed)	.000	.000	.000		.000	.175	.000	.000
	N	102	102	114	115	112	115	114	113
z-score	Pearson Correlation	.114	-.147	.484**	.840**	1	.164	.017	-.005
	Sig. (2-tailed)	.257	.144	.000	.000		.085	.857	.955
	N	100	100	111	112	112	112	111	110
FM	Pearson Correlation	-.186	.444**	-.142	-.127	.164	1	-.161	-.264**
	Sig. (2-tailed)	.061	.000	.131	.175	.085		.086	.005
	N	102	102	114	115	112	115	114	113
LM	Pearson Correlation	.465**	-.310**	.615**	.374**	.017	-.161	1	.709**
	Sig. (2-tailed)	.000	.002	.000	.000	.857	.086		.000
	N	101	101	113	114	111	114	114	112

LLM	Pearson Correlation	.602**	-.294**	.804**	.446**	-.005	-.264**	.709**	1
	Sig. (2-tailed)	.000	.003	.000	.000	.955	.005	.000	
	N	102	102	112	113	110	113	112	114

**. Correlation is significant at the 0.01 level

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